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CORRELATION OF THE CONFIGURATION OF SOME SULPHOXIDES WITH (+)S-METHYL-L-CYSTEINE S-OXIDE

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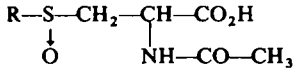
Abstract—The preparation of some (+)- and (−)-S-alkyl-L-cysteine S-oxides and (+)- and (−)-dicyclohexylammonium alkylsulphinylacetates from the dicyclohexylammonium salts of (+)- and (−)-N-acetyl-S-alkyl-L-cysteine S-oxides is described. On the basis of the ORD of the alkylsulphinylacetates, and their relationship to (+)-S-methyl-L-cysteine S-oxide, the configuration S is assigned to the (+)-dicyclohexylammonium N-acetyl-S-alkyl-L-cysteine S-oxides and to (+)-dicyclohexylammonium methylsulphinylacetate, (−)-dicyclohexylammonium ethylsulphinylacetate and (−)-dicyclohexylammonium propylsulphinylacetate.

A NUMBER of dextrorotatory sulphoxides derived from S-alkyl-L-cysteines have been



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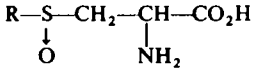

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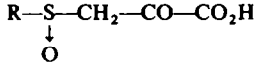

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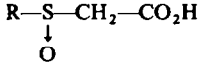


IV

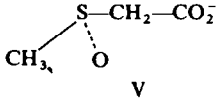
isolated from plant material,[1] and from animal sources (+)-N-acetyl-S-ethyl-L-cysteine S-oxide,[2] (+)-methionine sulphoxide[3] and (+)-2-hydroxy-3-methylsulphinylpropionic acid[4] have been isolated. Other sulphoxides from animals (chlorpromazine sulphoxide,[5] methylsulphinylacetic acid,[4] and N-acetyl-S-methyl-L-cysteine S-oxide[4]) have been isolated as mixtures of sulphoxide isomers, and N-acetyl-S-propyl-L-cysteine S-oxide[6,7] has been detected but has not been examined stereochemically. The S-alkyl-L-cysteine S-oxides from plant material are all dextrorotatory, and because of this factor it has been suggested by Klyne[8] that they have sterically related sulphoxide groups. Confirmation of this has been obtained recently by Gaffield *et al.*[9] who have made ORD measurements between 190 and 270 mμ, the region in which sulphoxide absorption occurs. It is probable that (+)-N-acetyl-S-alkyl-L-cysteine S-oxides have the same sulphoxide configuration as the corresponding (+)-S-alkyl-L-cysteine S-oxides, for (+)-N-acetyl-S-methyl-L-cysteine S-oxide has been prepared by the acetylation of the (+)-amino acid sulphoxide.[10]

In order to extend this observation experimentally and to confirm the correlation of sulphoxide conformations among the S-alkyl-L-cysteine S-oxides, the following experiments were carried out. Some N-acetyl-S-alkyl-L-cysteine S-oxides (I) were resolved and crystallized as their dicyclohexylammonium salts by methods which have already been reported,[2,4,6,10] and their properties are summarized in Table 1.

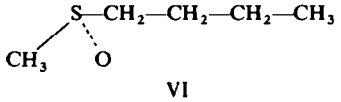
Deacetylation of these sulphoxides, which like others reported in the literature are decomposed by acids,[11–13] was effected under mild conditions with an enzyme (acylase I) to give the amino acid sulphoxide (II). (+)- and (−)-N-Acetyl derivatives gave (+)- and (−)-S-alkyl-L-cysteine S-oxides respectively, and their properties are recorded in Table 1. The amino acid sulphoxides were then converted to derivatives, which no longer contained an asymmetric C atom, by incubation with L-amino acid oxidase in the presence of oxygen. The first products of this reaction, hydrogen peroxide and a 2-oxo-acid (III), reacted together with the formation of an alkylsulphinylacetate[14] (IV) which was isolated as the dicyclohexylammonium salt (Table 2). These reactions, i.e. oxidative deamination followed by oxidative decarboxylation, are undergone by L-amino acids generally,[14] and are not likely to involve the sulphoxide group in the present compounds.

The corresponding (+)- and (−)-alkylsulphinylacetates gave optical rotatory dispersion curves that were, within experimental error, mirror images (Fig. 1). Furthermore, those alkylsulphinylacetates derived from (+)-S-alkyl-L-cysteine S-oxides showed the beginning of a positive Cotton effect which, in the absence of any other asymmetric centre in the compound, showed that they all had the same sulphoxide configuration and confirmed the correlation of the sulphoxide configurations of the amino acid sulphoxides. It should be noted, however, that although the optical rotation of a methylsulphinylacetate at the sodium D line had the same sign as that of the S-methyl-L-cysteine S-oxide from which it was prepared, the ethyl- and propyl-sulphinylacetates had rotations of opposite sign (Table 2). Another homologous series of sterically related sulphoxides examined previously ((−)-methylsulphinylalkylisothiocyanate derivatives[15]) had the same sign of rotation at the sodium D line, and so the present results with alkylsulphinylacetates serve to emphasize the fact that correlations should not be made on the basis of the sign of rotation at a single wavelength.

The absolute configuration of the sulphoxide group of (+)-S-methyl-L-cysteine S-oxide has been examined by X ray methods and shown to be S,[16] and so the configuration S can now be assigned to the following series of compounds: (+)-S-methyl-, (+)-S-ethyl- and (+)-S-propyl-L-cysteine S-oxide, the (+)-dicyclohexylammonium N-acetyl derivatives of these compounds, and to (+)-dicyclohexylammonium methylsulphinylacetate, (−)-dicyclohexylammonium ethylsulphinylacetate and (−)-dicyclohexylammonium propylsulphinylacetate. These latter compounds may serve as intermediates in the correlation of sulphoxide configurations with that of (+)-S-methyl-L-cysteine S-oxide, and the lowest homologue, (+)-methylsulphinylacetate (V) appears to be in agreement with another standard, (+)-methyl n-butylsulphoxide (VI), for which the configuration S has been determined.[17]



V



VI

EXPERIMENTAL

Elementary analyses were carried out by Weiler and Strauss, Oxford; m.ps are uncorrected. Optical rotations at the sodium D line were determined using a 1 dm tube in a manual polarimeter; data for the ORD curves were obtained with a Bellingham and Stanley Polarmatic 62 spectropolarimeter. IR absorption spectra were determined in KBr with a Perkin–Elmer 237 IR spectrophotometer. Enzyme preparations

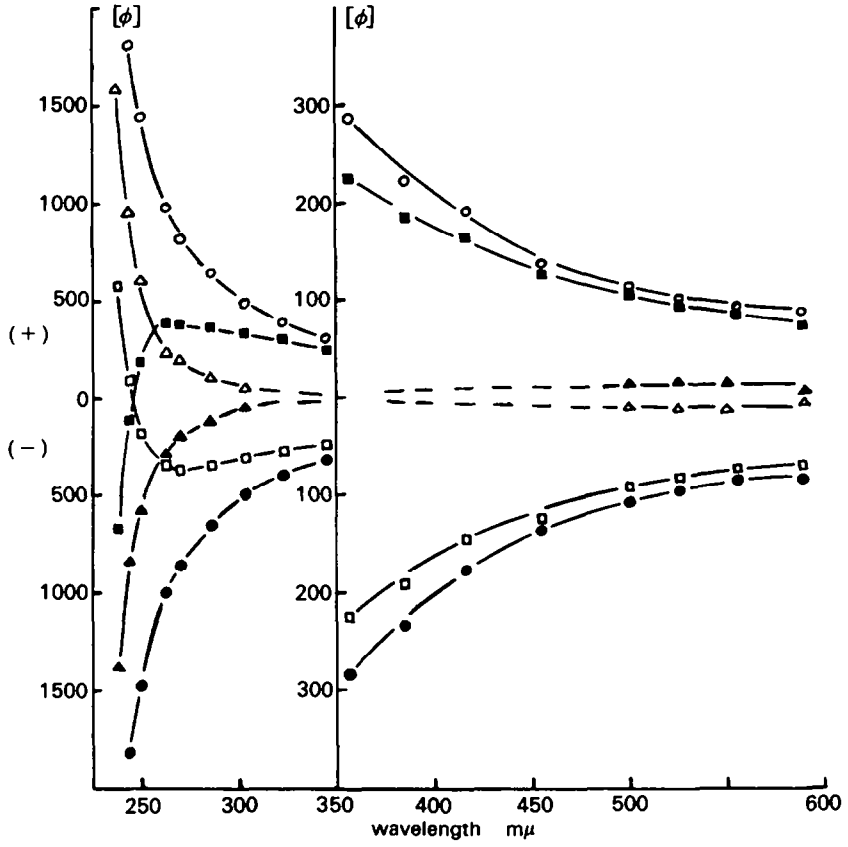


FIG. 1 ORD of dicyclohexylammonium alkylsulphinylacetates.

○, △, □; methyl-, ethyl- and propyl-derivatives from (+)-S-alkyl-L-cysteine S-oxides respectively.

●, ▲, ■; methyl-, ethyl- and propyl-derivatives from (−)-S-alkyl-L-cysteine S-oxides respectively.

(Concn 0·25 g/100 ml in water at 25°.) The magnitude of rotation of the ethyl-derivatives between 300 and 500 mμ is very small and of uncertain sign, and this is indicated by the dashed lines.

were obtained from commercial sources and used without further purification: hog-kidney acylase from British Drug Houses Ltd.; L-amino acid oxidase (*Crotalus adamanteus* venom) from L. Light & Co.

Deacetylation of N-*acetyl*-S-*alkyl*-L-*cysteine* S-*oxides.* The (+)- and (−)-dicyclohexylammonium salts of N-acetyl-S-alkyl-L-cysteine S-oxides were prepared as described previously[2,4,6,10] and some of their properties are given in Table 1. About 8 g of the salt was dissolved in water (100 ml) and dicyclohexylammonium ions were removed from the soln by passing it over a column of Zeo-Karb 225 resin (40 ml, H^+ form). The column was washed with water until the pH of the eluate rose to about 5, and the total eluate was neutralized with N NaOH. Phosphate buffer (50 ml, 0·44 M, pH 7·0) was added, and the soln was diluted to about 500 ml. Acylase (0·4 g) was added, and the soln was incubated at 37° for 24 hr. Samples were removed at intervals and diluted, and an amount equivalent to 5 μl of the incubation mixture was assayed for amino acids by the method of Synge and Wood.[11] Reaction was 90% complete after a period varying between 6–10 hr. The soln was passed over a column of Zeo-Karb 225 resin (100 ml, H^+ form), the column was washed with water until the pH rose to 4–5, and a further 500 ml of water was passed through the column. Aqueous ammonia (0·2 N) was passed through the column to displace the amino acid, and after the eluate became alkaline a further 200 ml of eluate was collected. The eluate was evaporated under reduced press at 50°. The residue was evaporated with acetone to effect crystallization, and then dissolved

TABLE 1. PROPERTIES OF S-ALKYL-L-CYSTEINE S-OXIDES AND THEIR DICYCLOHEXYLAMMONIUM N-ACETYL DERIVATIVES

Alkyl group	Dicyclohexylammonium N-acetyl deriv		Corresponding S-alkyl-L-cysteine S-oxide								
					Elementary analyses						
					Found %				Calculated %		
	$[\phi]_D$[a]	m.p.[c]	$[\phi]_D$[b]	m.p.[c]	C	H	N	S	C	H	N	S
Me	+138 ± 6°	166–167	+203 ± 15°[d]	163[d]	31·5	6·1	9·2	21·3	31·8	6·0	9·3	21·2
Me	−251 ± 6°	168–169	−202 ± 6°	165[e]	32·3	6·0	9·3	21·4				
Et	+ 63 ± 2°	163·5–164·5	+ 97 ± 7°	158	36·3	7·2	8·0	18·9	36·3	6·7	8·5	19·4
Et	−151 ± 2°	167–168	−127 ± 7°	155	36·6	6·9	8·4	18·9				
Pr	+ 4 ± 4°	167	+ 54 ± 7°[f]	163	40·0	7·7	7·8	17·8	40·2	7·3	7·8	17·9
Pr	− 80 ± 6°	166	− 59 ± 7°[g]	162	40·6	7·3	7·5	17·5				

[a] Concn 1·5 g/100 ml in water at room temp.
[b] Concn 0·4 g/100 ml for the Me compounds, and 1·0 g/100 ml for the others, in water at room temp.
[c] All m.ps occurred with dec, that of the amino acids was very dependent on the rate of heating.
[d] $[\phi]_D^{25} = +187°$, m.p. 174–175° have been reported previously.[20]
[e] $[\phi]_D^{25} = -192°$, m.p. 174–175° have been reported previously.[20]
[f] $[\phi]_D = +57°$, m.p. 164–168° have been reported previously.[21]
[g] $[\phi]_D = -51°$, m.p. 161–162° have been reported previously.[21]

TABLE 2. PROPERTIES OF DICYCLOHEXYLAMMONIUM ALKYLSULPHINYLACETATES

Alkyl group	$[\phi]_D$[a]	$[\phi]_D$ of corresponding S-alkyl-L-cysteine S-oxide[b]	m.p.[c]	Elementary analyses								Solvent for Recrystallization
				Found %				Calculated %				
				C	H	N	S	C	H	N	S	
Me	+88 ± 6°	+203°	151·5	58·9	9·5	4·6	10·4	59·4	9·6	4·6	10·6	Ethanol/acetone
Me	−85 ± 6°	−202°	150·5	59·5	9·7	4·7	11·1					Ethanol/acetone
Me	racemic	—	151	58·9	9·5	4·9	10·8					Ethanol/acetone
Et	+ 6 ± 4°	−127°	128–128·5	60·8	10·3	4·9	10·4	60·5	9·8	4·4	10·1	Ethyl acetate
Et	− 3 ± 3°	+ 97°	128	60·6	9·8	4·4	10·1					Ethyl acetate
Et	racemic	—	123	60·1	10·1	4·5	10·4					Ethanol/acetone
Pr	+69 ± 6°	− 59°	144·5	61·3	10·4	4·3	9·4	61·6	10·0	4·2	9·7	Ethyl acetate
Pr	−69 ± 6°	+ 54°	144·5	61·5	10·0	4·3	9·8					Ethyl acetate
Pr	racemic	—	134·5	61·3	9·9	4·4	10·0					Ethyl acetate

[a] Concn 1 g/100 ml in water; [b] see Table 1; [c] all compounds melt with dec.

in the minimum volume of 50% (v/v) aqueous EtOH or aqueous acetone. Protein was removed by centrifugation, and the amino acid was precipitated by the addition of EtOH. The crude product was usually recrystallized from aqueous EtOH, but (+)-S-ethyl- and (+)-S-propyl-L-cysteine S-oxide were recrystallized from aqueous MeOH by the addition of acetone. Aqueous EtOH gave gelatinous products. The properties of the products are recorded in Table 1. Although the yields of crude products were about 75%, recrystallization to give analytically and chromatographically pure material reduced the final yield to about 30%. The impurities appeared to be traces of other amino acids, probably derived from the acylase, and the major product lost in recrystallization was the required amino acid. Polarimetric examination of the mother liquors from recrystallization gave no evidence for the presence of a sulphoxide isomer other than the one being isolated.

Oxidative deamination of S-alkyl-L-cysteine S-oxides. About 1 g of S-alkyl-L-cysteine S-oxide was dissolved in 250 ml water in a 750 ml conical flask. The pH of the soln was adjusted to 7·0 with dil NaOH, and 50 mg *Crotalus adamanteus* venom was added. The flask was lightly plugged with cotton wool, and the contents were aerated by swirling. The temp was maintained at 35°. The course of the reaction was followed by removing samples (0·5 ml) and immediately estimating their ammonia content by the method of Conway.[18] Formation of 90% of the theoretical amount of ammonia took between 24 and 72 hr, depending upon the activity of the venom preparation. When the reaction was proceeding slowly (30–40% ammonia release in 24 hr) a further 50 mg of venom, and, in order to prevent bacterial growth, 10 mg of benzylpenicillin and 30 mg of streptomycin were added. The reaction mixture was heated in a boiling water bath for 5–10 min to coagulate protein, and was filtered. The filtrate was passed through a column of Zeo-Karb 225 resin (30 ml, H^+ form), and the column was washed with water until the pH of the eluate rose to 5. The total eluate was neutralized with a soln of dicyclohexylamine in acetone, and was evaporated at 50° under reduced press. The residue was extracted with EtOH, the extract was evaporated, and the dicyclohexylammonium alkylsulphinylacetate was recrystallized from the appropriate solvent (Table 2).

Preparation of racemic dicyclohexylammonium alkylsulphinylacetates. These compounds were prepared for comparison with the enantiomers derived from S-alkyl-L-cysteine S-oxides. Alkylthioacetic acids were prepared by a method similar to that described by Maw and du Vigneaud[19] for the Me derivative. The acid was dissolved in water, neutralized with dicyclohexylamine, and oxidized with one molar proportion of H_2O_2 for 24 hr at room temp. The soln was evaporated at 50° under reduced press, and the product was recrystallized from the appropriate solvent (Table 2). The IR spectra of the products were identical with those of the corresponding alkyl enantiomers.

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