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Nuclear magnetic resonance spectroscopy of the trimethylsilyl ethers of some hydroxyphenylalkylamines*

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A number of hydroxyphenylalkylamines have been converted into their trimethylsilyl ethers by the action of hexamethyldisilazane in pyridine. The derivatives are completely soluble in carbon tetrachloride and these solutions are suitable for nuclear magnetic resonance spectroscopy. The nmr spectra of 16 potentially biologically active phenylalkylamines have been obtained in this way. The spectral data are presented and the significance of the results discussed.

In recent years nuclear magnetic resonance (nmr) spectroscopy has been widely used in medicinal and pharmaceutical chemistry for the identification and stereochemical investigation of biologically important molecules (cf. Review by Casy, 1967). However relatively little work has been reported dealing with nmr studies on phenylalkylamines in general and the catecholamines in particular.

The nmr spectrum of adrenaline (I) in D_2O/DCl was first discussed by Weiner, Pappas & Jardetzky (1961) and Weiner & Jardetzky (1964) in the course of their studies on the interaction of adrenaline and ATP. More recently the nmr spectra of adrenaline (I) and a number of other phenylalkylamines have been reported (Clauder, Radics & others, 1968; Reisch, Alfes & Möllmann, 1968). In addition to D_2O, trifluoroacetic acid and hexadeuterodimethylsulphoxide (containing trifluoroacetic acid) have been used as solvents in these studies. However all these solvents have certain disadvantages for use in nmr spectroscopy. Solutions of catecholamines in strong acids, such as trifluoroacetic acid, often undergo slow decomposition, whilst the inevitable proton resonance peaks due to HOD or incompletely deuterated dimethyl sulphoxide often tend to mask important spectral regions.

The twin problems of involatility and low non-polar solvent solubility often encountered when investigating both the chromatographic properties of hydroxylated phenylalkylamines and their behaviour in the mass spectrometer have often been overcome in the past by the formation of suitable derivatives. Several have been prepared for this purpose including acetyl derivatives (cf. Waldi, 1962; Brooks & Horning, 1964; Stern, Franklin & Meyer, 1967); trifluoroacetates (cf. Greer, Sprinkle & Williams, 1968; Kawai & Tamura, 1968) and trimethylsilyl derivatives (Sen & McGeer, 1963; cf. Review by Pierce, 1968). The possible use of the trifluoroacetates was not considered in the course of the present investigation since they are difficult to prepare in the pure state and tend to be unstable. Preliminary experiments made by the authors with tri- and tetra-acetyladrenaline (Prepared by the methods of Welsh, 1952, and Bretschneider, 1947, respectively) suggested that these acetyl derivatives would be unsuitable for nmr studies, since, despite their high solubility

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in non-polar solvents, their nmr spectra were unexpectedly complex, probably due to the existence of more than one conformation of the molecule in both cases.

The trimethylsilyl ethers of the catecholamines and related compounds are relatively easy to prepare and since this type of derivative has been successfully used in the past for nmr studies on other phenolic compounds, e.g. flavonoids (Waiss, Lundin & Stern, 1964), it was decided to investigate the usefulness of this type of derivative for nmr studies on the catecholamines and some related phenylalkylamines. The trimethylsilyl derivatives of hydroxyphenylalkylamines in which all the hydroxy groups are silylated are readly prepared by the action of hexamethyldisilazane on the amine in pyridine at 95° (cf. Review by Pierce, 1968).

EXPERIMENTAL

Phenylalkylamines

Commercial samples of the phenylalkylamine derivatives were used whenever possible. In some cases it was necessary to recrystallize the samples before use. If the sample was not available commercially it was prepared by one of the methods described in the literature. The following phenylalkylamines were used (the source or literature reference is indicated in parenthesis): adrenaline (Th. Schuchardt); noradrenaline hydrochloride, *N*-ethylnoradrenaline hydrochloride, *N*-isopropylnoradrenaline hydrochloride; metanephrine hydrochloride, phenylephrine hydrochloride (Winthrop Laboratories); *N*-n-butylnoradrenaline hydrochloride, synephrine (Regis Chemical Co.); epinine hydrochloride (Wellcome Research Laboratories); adrenaline methyl ether hydrochloride, adrenaline ethyl ether hydrochloride (Hukki & Seppäläinen, 1958); adrenaline n-butyl ether hydrochloride, β-*p*-hydroxyphenylepinine, β-3,4-dihydroxy-2-methylphenylepinine, adrepine, adnamine (Forrest, Kăspárek & others, 1969); O^3,O^4-dimethyladrenaline (a gift from Dr. B. Jaques).

Preparation of trimethylsilyl derivatives

Hexamethyldisilazane* (2·4 ml) was added to a suspension of the phenylalkylamine (free base or hydrochloride: 100 mg) in anhydrous pyridine (2 ml) in an atmosphere of nitrogen. The reaction flask was loosely stoppered and heated, with stirring, on an oil-bath at 80–100° for 30–60 min. In cases where the free base was used, the reaction mixture was heated for 5 min after a clear solution was obtained. The reaction mixture was cooled to room temperature, and if solid was present, filtered rapidly through a fine sintered glass funnel. The flask and residue were washed with dry carbon tetrachloride (B.D.H. "Analar" grade). The combined filtrate and washings were concentrated to dryness *in vacuo* below 30°. Residual pyridine was removed from the product, usually a pale yellow oil, by repeated evaporation with carbon tetrachloride (2 ml portions). A solution of the product, so obtained, in carbon tetrachloride (1 ml) was used directly for nmr analysis.†

Nuclear magnetic resonance spectroscopy

The nmr spectra were obtained on a Varian A-60-A instrument. Chemical shifts are reported as τ values. Tetramethylsilane was used as an internal reference. The spectra were recorded both in the presence and absence of the reference compound.

* In practice it was observed that the best results were obtained when fresh samples of this reagent were used.

† These solutions tend to decompose on storage.

This was necessary in view of the proximity of some of the peaks in the spectrum to the reference peak.

RESULTS AND DISCUSSION

The nmr spectra of the trimethylsilyl derivatives of 16 hydroxyphenylalkylamine derivatives (freshly prepared in carbon tetrachloride solution) were recorded and the spectral data are given in Table 1.

This method has proved to be very useful for obtaining the nmr spectra of a number of hydroxyphenylalkylamines, containing a secondary amino-group. However difficulties were encountered when primary amines were used. The fact that complex and variable spectra were obtained with the trimethylsilylated primary amines was probably due to partial or inconsistent silylation of the primary amino-group. This was not altogether surprising since the silylation reaction was carried out with hexamethyldisilazane in pyridine and incomplete silylation of primary amino-groups has previously been reported with this reagent mixture. A detailed consideration of the procedures available for the silylation of phenylalkylamines in general and catecholamines in particular is outside the scope of this paper. The subject has however been reviewed in a recent book by Pierce (1968).

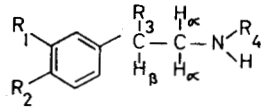
The following general conclusions can be drawn from consideration of the spectra of the trimethylsilyl derivatives of the 16 secondary hydroxyphenylalkylamines investigated. It was found that in all cases the hydroxyl groups were fully silylated but the secondary amino-groups were unaffected.

The aromatic ether trimethylsilyl protons were observed as sharp singlets in the region 9·75–9·83 τ, whilst the benzylic ether trimethylsilyl protons were seen at 9·94–9·99 τ, also as sharp singlets. The chemical shift of the trimethylsilyl protons appears to be a reliable indication of the type of hydroxyl group which has been silylated. The N-CH_3 group, when present, was observed in the range 7·59–7·62 τ, except for one of the N-CH_3 groups in adrepine (XV) and that in adnamine (XVI) which were found at 7·80 τ and 7·83 τ respectively. The N-CH_3 signal usually appeared as a sharp singlet, but extensive broadening of the N-CH_3 peaks was observed in two cases where the molecule contained a methoxyl group at the 3-position of the aromatic ring, i.e. metanephrine (VIII) and O^3,O^4-dimethyladrenaline (IX).

The N-H proton was observed as a broad signal in the 5·5–9·0 τ range; the position of the signal appeared to be markedly affected by concentration and temperature.

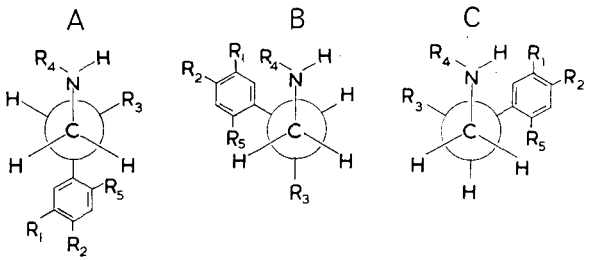
The positions and multiplicity of the signals due to the protons on the carbon atoms of the side-chain varied with the substitution pattern. In the compounds with a trimethylsilyl ether group on the carbon atom β to the nitrogen (i.e. I–IV; VIII–XI, XV) the signal due to the methine hydrogen [Hβ] on this carbon was observed between 5·23 and 5·42 τ, with a variety of different ring and N-alkyl substituents. In compounds with an O-alkyl group in this position (i.e. V, VI and VII), the corresponding methine proton signal was observed at a position about 0·5 ppm upfield in the range 5·75 to 5·85 τ. In compounds with either an alkyl or trimethylsilyl ether group in the β-position, the protons [Hα] on the carbon α to the nitrogen were seen between 7·30 and 7·43 τ, whereas in the β,β-diarylethylamine derivatives (i.e., XIII–XV), in which a second aryl group replaces the β-ether, these protons were seen in the 6·95–6·99 τ range. In the tricyclic compound adnamine the α proton is seen at 7·35 τ and the β proton at 6·22 τ.

Table 1. *Hydroxyphenylalkylamine trimethylsilyl ethers. Nuclear magnetic resonance spectra data*



Phenylalkylamine derivative	Compound No.	R_1	R_2	R_3	R_4	H_α	H_β	Notes
Adrenaline	I	9·77 (s)	9·77 (s)	9·99 (s)	7·61 (s)	7·43 (d), 7·39 (d) $J = 5·0$, $J = 7·5$	5·40 (dd) $J = 5·0$ & 7·5	R_1,R_2,R_3 = OTMS; $R_4 = CH_3$
N-Ethylnoradrenaline ..	II	9·78 (s)	9·78 (s)	9·99 (s)	CH_3CH_2 (see notes)	7·33 (d), 7·31 (d) $J = 5·0$, $J = 7·0$	5·38 (dd) $J = 5·0$ & 7·0	R_1,R_2,R_3 = OTMS; R_4 = $-CH_2$ $-CH_3$ 7·35 (q), 8·94 (t)
N-Isopropylnoradrenaline ..	III	9·79 (s)	9·79 (s)	9·99 (s)	$(CH_3)_2CH$ (see notes)	7·39 (d), $J = 6·5$,	5·42 (t) $J = 6·5$	R_1,R_2,R_3 = OTMS; R_4 = $(CH_3)_2$ $-CH$ 8·99 (d), 9·01 (d) 7·25 (sp)
N-n-Butylnoradrenaline ..	IV	9·78 (s)	9·78 (s)	9·99 (s)	$CH_3CH_2CH_2CH_2$ (see notes)	7·32 (m)	5·42 (dd) $J = 5·0$ & 7·0	R_1,R_2,R_3 = OTMS; R_4 = CH_3 – CH_2CH_2 – CH_2 9·17 (t), 8·70 (m), 7·47 (t)
Adrenaline methyl ether ..	V	9·77 (s)	9·77 (s)	6·71 (s)	7·60 (s)	7·39 (d), 7·33 (d) $J = 5·0$, $J = 8·0$	5·85 (dd) $J = 5·0$ & 8·0	R_1,R_2 = OTMS; $R_3 = OCH_3$; $R_4 = CH_3$
Adrenaline ethyl ether ..	VI	9·75 (s)	9·75 (s)	OCH_2CH_3 (see notes)	7·61 (s)	7·42 (d), 7·35 (d) $J = 5·0$, $J = 8·0$	5·75 (dd) $J = 5·0$ & 8·0	R_1,R_2 = OTMS; $R_4 = CH_3$; R_3 = – O–CH_2 – CH_3 6·64 (q), 6·67 (q), 8·84 (t)
Adrenaline n-butyl ether ..	VII	9·75 (s)	9·75 (s)	O-*n*-butyl (see notes)	7·61 (s)	7·40 (d), 7·33 (d) $J = 5·0$, $J = 8·0$	5·77 (dd) $J = 5·0$ & 8·0	R_1,R_2 = OTMS; $R_4 = CH_3$; R_3 = CH_3–CH_2CH_2–CH_2O 9·12 (t), 8·55 (m), 6·68 (m)
Metanephrine	VIII	6·22 (s)	9·78 (s)	9·98 (s)	7·61 (bs)	7·42 (bm)	5·33 (t) $J = 6·0$	$R_1 = OCH_3$; R_2,R_3 = OTMS; $R_4 = CH_3$
O^3,O^4-Dimethyladrenaline ..	IX	6·23 (s)	6·20 (s)	9·98 (s)	7·61 (bs)	7·35 (bm)	5·29 (t) $J = 6·5$	$R_1,R_2 = OCH_3$; R_3 = OTMS; $R_4 = CH_3$
Synephrine	X	H (aromatic)	9·76 (s)	9·99 (s)	7·59 (s)	7·30 (m)	5·23 (t) $J = 6·5$	R_1 = H; R_2,R_3 = OTMS, $R_4 = CH_3$ Aromatic AA′BB′ at 3·09, $J = 9·0$
Phenylephrine	XI	9·78 (s)	H aromatic	9·97 (s)	7·62 (s)	7·39 (d), 7·41 (d) $J = 7·0$, $J = 5·0$	5·31 (dd) $J = 5·0$ & 7·0	R_1 = OTMS; R_2 = H; R_3 = OTMS; $R_4 = CH_3$
Epinine	XII	9·79 (s)	9·79 (s)	7·33 (bt)	7·62 (s)	7·33 (bt)	7·33 (bt)	R_1,R_3 = OTMS; R_2 = H; $R_4 = CH_3$

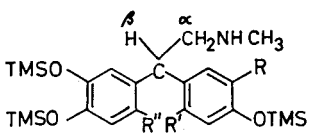
Notes: The aromatic proton signals occurred as multiplets in the region of 3·09 to 3·34.
The position of the N–H signal was concentration dependent and was found within the range of 5·57 to 8·95.
(s) = singlet; (d) = doublet; (dd) = doublet of doublets;
(t) = triplet; (q) = quartet; (b) = broad; (sp) = septet; (bs) = broad singlet.



The conformational populations of the various compounds could be estimated from the coupling constants of the side-chain protons which depend upon the percentage distribution of the individual conformers (cf. Emsley, Feeney & Sutcliffe, 1965). Values for the *trans* and *gauche* coupling constants, required for these calculations were obtained from consideration of the spectrum of adrepine [XV] (Forrest, Kăspárek & others, 1969). This amine which has an extra bulky substituent [R_5] in the *ortho*-position, causing it to exist mainly as one of conformer A or B, shows a *trans* coupling of 9·0 Hz (dihedral angle of 180°) and a *gauche* coupling of 2·5 Hz (dihedral angle of 60°). Using these values for the *gauche* and *trans* coupling constants of all conformers, estimates of the conformer populations could be made. The approximate distribution was found to vary from a 50 : 50 mixture of A and B, as in the case of *N*-isopropylnoradrenaline [III], to a 70 : 30 mixture as in the case of adrenaline [I]. It must be emphasized that in view of the first order analysis of the spectra and the use of *trans* and *gauche* coupling constants from a model compound, these values must be considered as approximations only.

This procedure for determining the nmr spectra of catecholamines should prove a useful adjunct to mass spectrometry and gas liquid chromatography for the identification of this type of compound.

Table 2. *Diphenylalkylamine trimethylsilyl ethers. Nuclear magnetic resonance spectral data*



XIII—XVI

Diphenylalkylamine derivative	Compound number	Aromatic OTMS	N–CH_3	Hα	Hβ	Aromatic H	Others
β-*p*-Hydroxyphenylepinine R=R′=R″=H	XIII	9·77(s) 9·79(s) 9·83(s)	7·59(s)	6·95(d) J=7·5	6·00(t) J=7·5	3·29(m) 3·08 AA′BB′ J=9·0	
β-3,4-Dihydroxy-2-methylphenylepinine R=OTMS; R′= CH_3, R″=H	XIV	9·78(s) 9·87(s)	7·60(s)	6·96(d) J=7·0	5·83(t) J=7·0	3·30(m)	C–CH_3 7·87(s)
Adrepine R=OTMS; R′=$C(OTMS)(H)\cdot CH_2\cdot NHCH_3$; R″=H	XV	9·82(s) 9·79(s) 9·77(s) 9·74(s)	7·59(s) 7·80(s)	6·99(d) J=7·0	5·74(t) J=7·0	3·29(m)	Aliphatic OTMS 9·94(s) Hα′ 8·12(dd), J=12·0 and 2·5 Hα′ 7·47(dd), J=12·0 and 9·0 Hβ′ 4·99(dd), J=2·5 and 9·0
Adnamine R=OTMS; R′–R″=–CH=CH–	XVI	9·83(s) 9·81(s)	7·83(s)	7·35(d) J=7·5	6·22(t) J=7·5	3·44(bs)	H olefinic 3·44(bs)

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