Novel Polycyclic Heterocycles. IV.[1] Structure of the Dimer of 5,11-Dihydrodibenz[*b*,*e*][1,4]oxazepine. Infrared, Proton Magnetic Resonance, and Mass Spectral Studies

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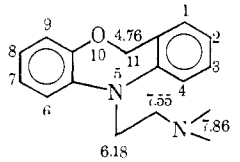
In the synthesis of 5-[2-(dimethylamino)ethyl]-5,11-dihydrodibenz[*b*,*e*][1,4]oxazepine (**1**), by the reaction of the anion of the heterocycle with 2-dimethylaminoethyl chloride, one of the by-products isolated from the residue from the distillation of **1** has been identified as 5-(*o*-[*o*-[2-(dimethylamino)ethoxy]-*N*-[2-(dimethylamino)ethyl]-anilino}benzyl)-5,11-dihydrodibenz[*b*,*e*][1,4]oxazepine (**2**). It has been shown in addition that, in the absence of 2-dimethylaminoethyl chloride, the anion of the heterocycle forms the parent dimer, 5-[*o*-(*o*-hydroxyanilino)-benzyl]-5,11-dihydrodibenz[*b*,*e*][1,4]oxazepine. The infrared, proton magnetic resonance, and mass spectra of these and related compounds are discussed.

In an earlier paper[2] we reported on a series of 5-(dialkylaminoalkyl)-5,11-dihydrodibenz[*b*,*e*][1,4]oxazepines. In the synthesis of one member of this series, 5-[2-(dimethylamino)ethyl]-5,11-dihydrodibenz-[*b*,*e*][1,4]oxazepine (**1**), by the reaction of the anion of the heterocycle, prepared by means of sodium hydride in tetrahydrofuran, with 2-dimethylaminoethyl chloride,[3] one of the by-products which could be isolated from the residue from the distillation of **1** was a crystalline substance (**2**), mp 123–125°. Analysis by gas–liquid partition chromatography and by paper chromatography showed that both **1** and **2** were homogeneous; elemental analyses indicated that **2** had the identical empirical formula as **1**, $C_{17}H_{20}N_2O$; a molecular weight determination established that **2** was a dimer of **1**.

The structure of **2** was of interest since it showed biological properties different from **1**; the latter, for example, was a potent antihistaminic[2] while **2** was devoid of this activity. Of particular significance, however, was the observation that **2** inhibited the multiplication of Earle's L cells of mouse fibroblasts growing in suspension,[3,4] having an ED_{50} of 1 μg/ml while **1** had an ED_{50} of 50 μg/ml.

The infrared and ultraviolet spectra of **1** and **2** showed only minor differences which *per se* offered no clues as to the structure of **2**. In the pmr spectrum[5] of **1**, the methylene group at position 11 appeared as a two-proton singlet at τ 4.76, while the 1-methylene group of the basic side chain appeared as a two-proton triplet centered at τ 6.18. In **2**, four two-proton signals were of special significance since they established **2** as an unsymmetrical dimer of **1**:[6] (1) the singlet at



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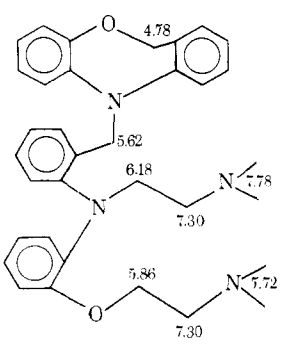

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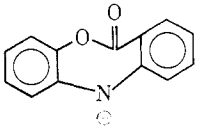

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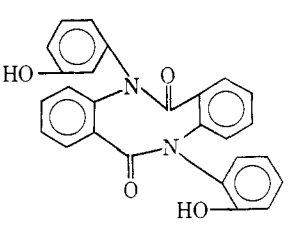

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τ 4.78 indicated one intact heterocycle; (2) the singlet at τ 5.62, shifted upfield from τ 4.76, indicated that the methylene group originally present in the second heterocyclic molecule (a) was now exocyclic, revealing a base-catalyzed benzyl ether type of cleavage[7] and (b) that this methylene group was not adjacent to any other methylene group; (3) the triplet centered at τ 6.18 indicated a methylene group attached to nitrogen; and (4) the triplet centered at τ 5.86, shifted downfield from τ 6.18, established that one methylene group was attached not to nitrogen but to the more electronegative oxygen atom. An examination of the pmr spectra of several authentic reference structures **5**–**7** offered convincing evidence for the proposed structure of **2**; what remained somewhat anomalous was the upfield shift of *ca.* 0.5 ppm of the "benzyl-type" protons, when the reference structures **8**–**11** were used for comparison, although it was obvious from these structures that these "benzyl-type" protons were particularly sensitive to the shielding and deshielding effects associated with

(1) For paper III, see H. L. Yale and F. Sowinski, *Arzneimittel-Forsch.*, **16**, 550 (1966).

(2) Paper II: H. L. Yale and F. Sowinski, *J. Med. Chem.*, **7**, 609 (1964).

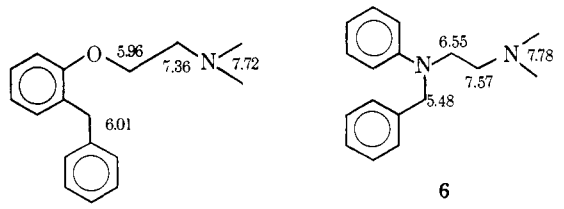
(3) D. Perlman, N. A. Guiffre, and P. W. Jackson, *Proc. Soc. Exptl. Biol. Med.*, **102**, 290 (1959).

(4) H. L. Yale and M. Kalkstein, *J. Med. Chem.*, **10**, 334 (1967).

(5) The pmr spectra of the compounds described in this paper were determined by Dr. Allen I. Cohen of these laboratories.

(6) It is of interest that H. Gorien, D. H. Malarek, and A. I. Raciillo, *J. Heterocyclic Chem.*, **3**, 527 (1966), have reported that the dibenz[*b*,*e*][1,4]-oxazepin-6(11H)-one anion (**3**), prepared by means of NaH in xylene, gave the symmetrical dimer **4**.

(7) M. S. Kharasch and R. L. Huang, *J. Org. Chem.*, **17**, 669 (1952), have reported that alkyl aryl ethers were not cleaved by the Grignard–CoCl$_2$ reagent but that benzyl phenyl ether was to give phenol and toluene. The earlier report by G. Wittig and L. Löhmann, *Ann.*, **550**, 260 (1942), that phenyllithium and benzyl ethyl ether gave benzyl alcohol and ethylene, but that phenyllithium and benzyl methyl ether gave methylphenylcarbinol, led to the recognition of the Wittig rearrangement. The difficulty in applying any single mechanistic scheme to the Wittig rearrangement has been discussed by P. T. Lansbury and V. A. Pattison, *J. Org. Chem.*, **27**, 1933 (1962); a similar difficulty exists in delineating a mechanism for the formation of **2**. In this connection, S. O. Winthrop and his co-workers [*J. Med. Chem.*, **5**, 1207 (1962); *Tetrahedron Letters*, 1113 (1965)] have discussed the Wittig-type rearrangement of 11-keto-6,11-dihydrodibenzo[*b*,*e*]oxepine.



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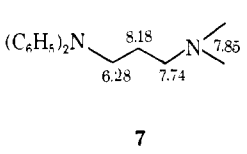

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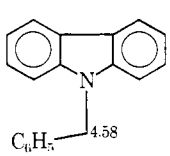

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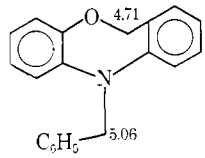

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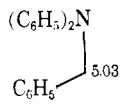

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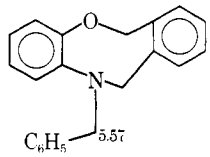

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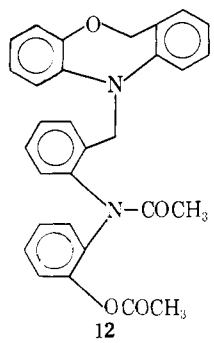
the conformation of the molecule. Thus, it is probable that in the preferred conformation of **2**, the shielding influence of the strongly basic side chains and of the substituted diphenylamine portion of the molecule were responsible for the upfield shift observed with these protons.[8]

Confirmation of the proposed structure of **2** was obtained by dimerization of the dibenzoxazepine anion in the absence of 2-dimethylaminoethyl chloride. The reaction product could be separated arbitrarily into two fractions based on solubility in 1% aqueous NaOH: the major product was an insoluble amorphous brown polymer; the alkali soluble product could be isolated by acidification, purified by chromatography, and then acetylated to give a crystalline diacetyl derivative whose structure as **12** was confirmed by its infrared and pmr spectra. The former showed two strong absorption bands at 1750 and at 1670 cm^{-1}, indicative, respectively, of the C=O stretching vibrations of the O-acetyl and N-acetyl groups of the dimer.[9a] The pmr spectrum showed two CH_2 singlets at τ 4.74 and 5.55



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(8) A comparison of the pmr spectra of toluene and *o*-toluidine add substance to this proposal: in the former, the three CH_3 appear as a singlet at τ 7.68, while in the latter these protons are shifted upfield *ca.* 0.5 ppm and appear as a singlet at τ 8.17.

(9) (a) For comparison, the C=O stretch absorption in N-acetyldiphenylamine (**13**) is found at 1670 cm^{-1}, and in O,N-diacetyl-4-hydroxydiphenylamine (**14**) the absorptions are at 1760 and 1670 cm^{-1}, respectively. (b) In the pmr spectrum of **13**, the three methyl protons are seen as a singlet at τ 7.97; in **14**, the two three proton signals are at τ 7.77 and 7.98, respectively.

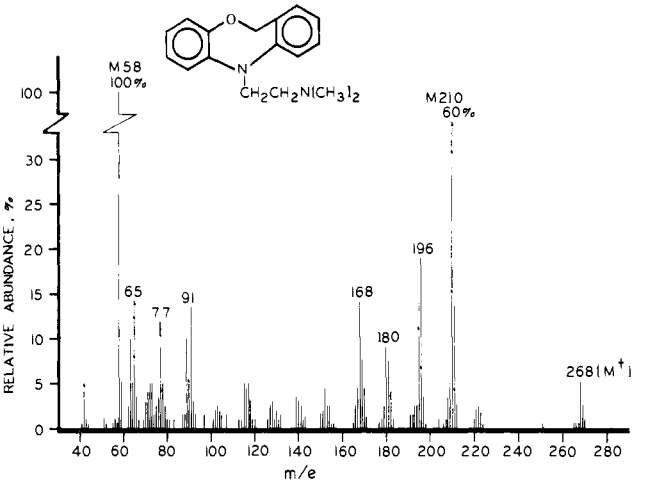


Figure 1.—Mass spectrum of **1**.

while the two singlets of the CH_3 of the dissimilar acetyl groups appeared at τ 7.83 and 8.00.[9b]

Mass Spectra of 1 and 2.—To our knowledge there are no reports in the literature concerning the electron-impact fragmentation patterns of N-dialkylaminoalkyl polycyclic heterocycles,[10] and it was of interest in connection with the studies on the elucidation of the structure of **2** to examine the mass spectra of **1** and **2**. The fragmentation patterns of a number of steroids[11] and terpenes[12] containing the amino and dimethylamino groups have been of considerable interest since these basic groups have demonstrated powerful fragmentation directive influence. Thus, N,N-dimethylbornylamine[12] showed the M^+ peak at *m/e* 181 and *two* base peaks (relative abundance difference only *ca.* 3%)[13] at *m/e* 58 and 95. The *m/e* 58 peak was shown to be due to the $CH_2{=}N^+(CH_3)_2$ fragment arising *via* β cleavage[14] while the *m/e* 95 peak was due to the hydrocarbon fragment arising *via* loss of $CH_2{=}CHN(CH_3)_2$ and a CH_3 group. The next most abundant peak was at *m/e* 110 (90%), attributable to the $(CH_2{=}CH)_2C{=}N^+(CH_3)_2$ cation. The remaining peaks of interest at *m/e* 71 (22%), 72 (42%), and 98 (26%) were represented by the fragments $-CH_2CH{=}N^+(CH_3)_2$,  , and $CH_2{=}CHCH_2CH{=}N^+(CH_3)_2$, respectively. Thus, in this bicyclic structure, the hydrocarbon framework has a fragmentation-directing ability which can compete with the very strong fragmentation-directing dimethylamine group.

The mass spectra of **1** and **2** are shown in Figures 1 and 2. The mass spectrum[15] of **1** showed the M^+ peak at *m/e* 268, the base peak at *m/e* 58, and other peaks at the following *m/e*: 210 (60%), 197 (3%), 196 (19%),

(10) K. G. Das, P. T. Funk, and A. K. Bose [*J. Am. Chem. Soc.*, **86**, 3729 (1964)] and N. P. Buu-Hoi, M. Mangane, and P. Jacquignon [*J. Heterocyclic Chem.*, **3**, 149 (1966)] have reported on the mass spectrum of carbazole but not on any of its N-substituted derivatives.

(11) For discussion and references, see H. Budzikiewicz, C. Djerassi, and D. H. Williams, "Structure Elucidation of Natural Products by Mass Spectrometry," Vol. 2, Holden-Day, Inc., San Francisco, Calif., 1964, Chapter 18.

(12) D. S. Weinberg and C. Djerassi, *J. Org. Chem.*, **31**, 3832 (1966).

(13) The use of per cent relative abundance with the most intense peak (base peak) being taken as 100% is used throughout the entire text of ref 11.

(14) R. M. Silverstein and G. C. Bassler, "Spectrometric Identification of Organic Compounds," John Wiley and Sons, Inc., New York, N. Y., 1966, p 9.

(15) The spectra of **1** and **2** were determined at the Department of Chemistry, Stanford University, through the cooperation of Dr. Carl Djerassi.

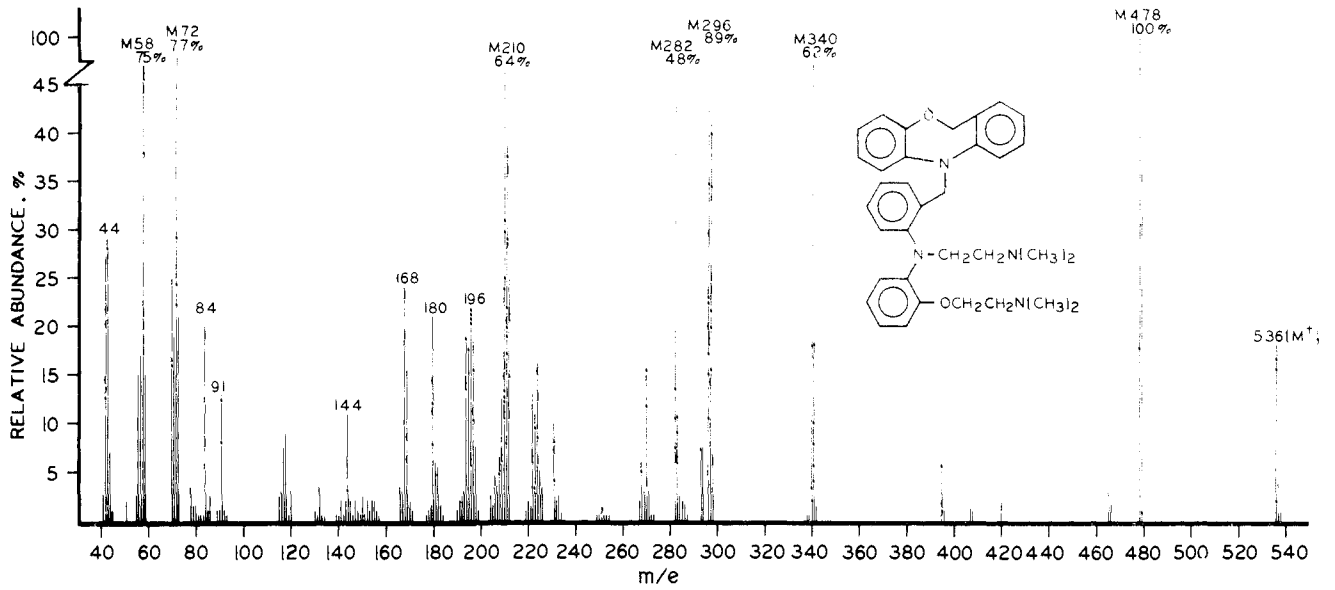
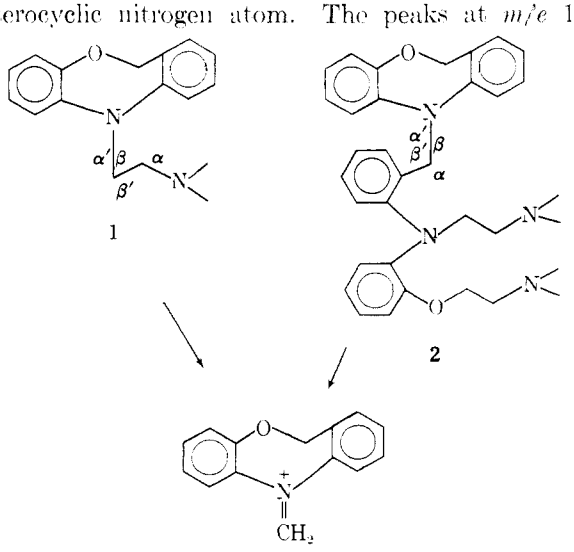


Figure 2.—Mass spectrum of **2**.

168 (14%), 91 (14%), 77 (11%), 72 (5%), 71 (4%), 70 (3%), and 65 (15%). The spectrum of **2** showed the M^+ peak at *m/e* 536, the base peak at *m/e* 478, and other peaks at the following *m/e*: 340 (62%), 296 (89%), 282 (48%), 210 (64%), 197 (17%), 196 (22%), 168 (22%), 91 (13%), 77 (4%), 72 (82%), 71 (19%), 70 (25%), 65 (6%), and 58 (75%).

Thus, electron impact on **1** and **2** resulted in the elimination of the $CH_2{=}N^+(CH_3)_2$, $-CH_2CH{=}N^+(CH_3)_2$, and  fragments due to the anticipated directing influence of the dimethylamino group. Of equal significance were the peaks in **1** and **2** at *m/e* 210 representing the stable cation **15**, arising *via* reinforced β cleavage directed from both the heterocyclic nitrogen atom and from the dimethylamino nitrogen atom in **1**, but by an unprecedented β fragmentation in **2**, *i.e.*, one not favoring formation of a "phenylalkyl" ion,[16] and, thus, representing a striking example of the powerful directive influence of the heterocyclic nitrogen atom. The peaks at *m/e* 196 in **1** and **2** represented the anticipated α-directed cleavage of the heterocyclic nitrogen atom, the peaks at *m/e* 340 in **2** represented the anticipated "phenylalkyl" ion fragmentation, and the peaks at *m/e* 168, 91, 77, and 65 in both **1** and **2** were associated with further fragmentation and/or rearrangement[17] of the heterocyclic portion of the molecule. The greater abundance of the *m/e* 72 peak in **2** as compared to **1** supports the assignment of one of the dimethylaminoethyl groups as being attached to an oxygen rather than a nitrogen atom. Finally, contrary to what was observed in the spectrum of *N,N*-dimethylbornylamine,[12] no $M-CH_3$ peak was seen in the spectrum of either **1** or **2**. Thus, the analysis of the mass spectrum of **2** confirmed the structure previously assigned on the basis of its pmr spectrum and served to illustrate an unanticipated fragmentation pattern associated with this derivative of 5,11-dihydrodibenz[*b,e*][1,4]oxazepine.



Experimental Section

5-{*o*-{*o*-[2-(Dimethylamino)ethoxy]-*N*-[2-(dimethylamino)ethyl]-anilino}benzyl)-5,11-dihydrodibenz[*b,e*][1,4]oxazepine.—To 98.8 g (0.5 mole) of 5,11-dihydrodibenz[*b,e*][1,4]oxazepine in 1 l. of tetrahydrofuran (THF, dried over $LiAlH_4$) under N_2 was added in small portions during 0.5 hr 30.0 g (0.625 mole) of a 50% NaH dispersion in mineral oil. Subsequently, the mixture was stirred for 0.5 hr, 80.6 g (0.75 mole) of 2-dimethylaminoethyl chloride was added in 0.5 hr, and the mixture was stirred and heated under reflux for 2 hr, then stirred overnight at room temperature. A second portion of 24.0 g of 50% NaH was added in small portions followed by 53.5 g of 2-dimethylaminoethyl chloride, and the mixture was stirred and heated under reflux for 5 hr and filtered. The filtrate was concentrated to dryness *in vacuo*, the residue was dissolved in 500 ml of ether, and the ether solution was extracted successively with 500 and 250 ml of 10% aqueous phosphoric acid. The aqueous acid extracts were treated with an excess of solid K_2CO_3 and the liberated base was extracted into ether. The ether extracts were dried ($MgSO_4$), treated with Darco, and filtered, and the filtrate was concentrated to give 134.3 g (quantitative yield) of a viscous oil. The products from this and a second identical experiment were combined. Attempts to obtain a pure maleate salt from this oil were unsuccessful. Therefore, the base was distilled to give 145.6 g (54%

(16) H. M. Grubb and S. Meyerson in "Mass Spectrometry of Organic Ions," F. W. McLafferty, Ed., Academic Press Inc., New York, N. Y., 1963, pp 453–527.

(17) The skeletal and hydrogen rearrangements in aryl alkyl ether ions have most recently been discussed by F. W. McLafferty, M. M. Bursey, and S. M. Kimball, *J. Am. Chem. Soc.*, **88**, 5022 (1966).

yield) of the desired I, bp 151–153° (0.5 mm), $n^{20}D$ 1.5998; maleate salt, mp 141–142°, alone or mixed with authentic material.[2] This material was found to be homogeneous by paper chromatography and gave satisfactory analyses.

The residue from the distillation was recrystallized from acetone repeatedly to give 24 g (12% yield) of **2**, mp 123–125°. This material was homogeneous by paper chromatography and glpc.

Anal. Calcd for $C_{38}H_{40}N_4O_2$: C, 76.08; H, 7.51; N, 10.44; mol wt, 537. Found: C, 75.80; H, 7.78; N, 10.79; mol wt (osmometrically), 541.

The dimer did not give a crystalline salt with either HCl or maleic acid.

5-[*o*-(*o*-Acetoxy-N-acetylanilino)benzyl]-5,11-dihydrodibenz[*b,e*][1,4]oxazepine.—A mixture of 4.90 g (0.025 mole) of 5,11-dihydrodibenz[*b,e*][1,4]oxazepine, 1.44 g (0.030 mole) of 50% NaH dispersion in mineral oil, and 50 ml of xylene was stirred and heated under reflux for 8 hr and filtered, and the filtrate was concentrated to dryness *in vacuo*. The residue was extracted with 500 ml of 1% aqueous NaOH. The insoluble brown amorphous polymeric material was separated by filtration. The filtrate was treated with 1 g of Darco and refiltered, and the filtrate was acidified with 10% aqueous H_3PO_4. The precipitated solid was collected, dried, dissolved in 15 ml of benzene, chromatographed on a 1 × 20 cm column of Florisil, and eluted with benzene to give 1.0 g of material, mp 75–84°. In the infrared this material showed a single strong broad OH plus NH absorption band centered at *ca.* 3330 cm^{-1}. The solid, 1.0 g, 10 ml of Ac_2O, and 0.5 ml of pyridine were kept 18 hr at room temperature and concentrated *in vacuo*, and the residue was dissolved in 20 ml of 1:10 benzene–acetone, chromatographed on Florisil, and eluted with the same solvent to give a crystalline product, mp 92–95° dec, ν 1670 and 1750 cm^{-1} with no absorption at 3330 cm^{-1}.

Anal. Calcd for $C_{30}H_{26}N_2O_4$: C, 75.28; H, 5.47; acetyl, 17.9. Found: C, 75.26; H, 5.58; acetyl, 15.4.

5-Benzyl-5,11-dihydrodibenz[*b,e*][1,4]oxazepine.—To 2.50 g (0.0125 mole) of 5,11-dihydrodibenz[*b,e*][1,4]oxazepine in 25 ml of anhydrous THF was added 4.28 g (0.025 mole) of benzyl bromide. To the stirred solution was added in 0.5 hr 0.96 g (0.02 mole) of 50% NaH dispersion. The mixture was stirred 18 hr at room temperature and filtered, the filtrate was concentrated *in vacuo*, and the residual solid was recrystallized from pentane to give 1.65 g (46% yield) of product, mp 84–86°.

Anal. Calcd for $C_{20}H_{17}NO$: C, 83.58; H, 5.97; N, 4.88. Found: C, 83.55; H, 6.14; N, 4.95.

O,N-Diacetyl-4-hydroxydiphenylamine.—A solution of 6.0 g (0.033 mole) of 4-hydroxydiphenylamine, 50 ml of Ac_2O, and 0.5 ml of pyridine was refluxed for 1 hr and concentrated to dryness *in vacuo*. The residue was recrystallized from hexane to give 7.2 g (83% yield) of product, mp 117–118°.

Anal. Calcd for $C_{16}H_{15}NO_3$: C, 71.35; H, 5.62; N, 5.21. Found: C, 71.13; H, 5.49; N, 5.26.

12-Benzyl-11,12-dihydro-6H-dibenz[*b,f*][1,4]oxazocine.—To a solution of 0.90 g (0.004 mole) of 11,12-dihydro-6H-dibenz[*b,f*]-[1.4]oxazocine in 40 ml of anhydrous THF was added 2.5 ml of 1.6 *N* butyllithium in hexane. The mixture was stirred 1 hr at room temperature, 0.70 g (0.004 mole) of benzyl bromide in 5 ml of anhydrous hexane was added dropwise, and the stirring at room temperature was continued for 48 hr. The solution was concentrated *in vacuo*, and the residue was recrystallized from petroleum ether (bp 30–60°) to give 0.4 g (33% yield) of product, mp 70–72°.

Anal. Calcd for $C_{21}H_{19}NO$: C, 83.64; H, 6.35; N, 4.64; neut equiv, 301. Found: C, 83.19; H, 6.81; N, 4.58; neut equiv ($HClO_4$ in glacial acetic acid), 298.

The Chemistry and Biological Activity of Derivatives of Strophanthidin[1]

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A number of new synthetic derivatives of the cardenolide, strophanthidin, have been prepared in an attempt to delineate and compare the structural requirements for activity in three biological systems; *i.e.*, cytotoxic activity against human carcinoma of the nasopharynx in tissue culture (KB), inhibition of a brain transport ATPase, and cardiotonic activity. Strophanthidin was converted to the monoanhydroacetate derivative Vb and the dianhydroacetate derivative VIb. Epoxidation of Vb yielded a mixture of epoxides (X and XI) in which the α-epoxide X predominated. Treatment of the mixture with hydrogen chloride in chloroform gave the 5α-hydroxy-6β-chloro derivative VII and the hemiacetal of the 5α-chloro-6β-hydroxy derivative IXa. Upon attempted acetylation, VII was largely recovered unchanged, whereas IXa gave the diacetate IXb. Potassium acetate treatment of VII gave pure X, and similar treatment of IXa gave XI. Epoxidation of VIb gave the 5,6α-14,15α-diepoxide XIII and the 5,6β-14,15α-diepoxide XIV. Diepoxide XIII was also obtained by dehydration of X to XII, followed by epoxidation of XII to XIII. Strophanthidin 3-iodoacetate (XVIIb) was converted to the azidoacetate XVIII; the latter was reduced to the 3-aminoacetate hydrochloride XIX, which was directly converted to the 3-diazoacetate XX. The results of the biological tests of the foregoing and other derivatives of strophanthidin indicate that similar structural features are important for activity in each of the three systems. These results indicate that the receptors for the cardenolides in the three systems may be structurally very similar.

The cardiotonic steroids exert a specific and powerful action on the heart muscle and have been successfully used in heart therapy for almost two centuries. Most therapeutically useful members of the group are glycosides, although many aglycones show potent activity. Extensive studies have shed considerable light upon relationships between cardiotonic activity and structure and configuration of the cardiotonic steroids.[3–5]

Cardiotonic steroids have also been shown to be specific and reversible inhibitors of adenosine triphosphatases (ATPases) involved in the active transport of Na^+ and K^+.[6–8] A preliminary study of structure–activity relationships among 21 digitalis derivatives revealed that, in most cases, the ability of a cardiotonic steroid to block a transport ATPase *in vitro* paralleled its activity and toxicity in the intact animal.[9]

(1) (a) Tumor Inhibitors. XXVI. Part XXV of the series: S. M. Kupchan and M. Mokotoff, *J. Med. Chem.*, **10**, 977 (1967). (b) This investigation was supported by grants from the National Institutes of Health (CA-04500 and NB-01730) and the American Cancer Society (T-275).

(2) (a) Fellow of the American Foundation for Pharmaceutical Education, 1963–1965. (b) Research Career Awardee of the National Institutes of Health (5-K6-GM-1347).

(3) K. K. Chen, Proceedings of the 1st International Pharmacological Meeting, Stockholm, 1961, Vol. 3, p 27.

(4) Ch. Tamm, ref 3, p 11.

(5) F. G. Henderson and K. K. Chen, *J. Med. Chem.*, **8**, 577 (1965).

(6) H. J. Schatzmann, *Helv. Phys. Acta*, **11**, 346 (1953).

(7) I. M. Glynn, *Pharmacol. Rev.*, **16**, 381 (1964).

(8) J. C. Skou, *Physiol. Rev.*, **45**, 596 (1965).

(9) K. Repke in "Drugs and Enzymes," B. B. Brodie and J. R. Gillette, Ed., Pergamon Press Ltd., Oxford, 1965, p 65.