tients who are receiving digoxin, steady-state studies in normal volunteers (7) correlate with the single-dose studies, suggesting that either protocol may be used.

The dose of digoxin appears to be important with regard to its absorption. Greenblatt *et al.* (12) used a single dose of 0.75 mg and found that the absolute bioavailability relative to an intravenous infusion of digoxin was 65% for the elixir and 55% for a digoxin tablet. Both of these values are considerably lower than the findings in this study. They administered the intravenous digoxin by a 1-hr infusion and found a slightly higher percent of the intravenous dose of digoxin in the urine, which may explain these differences.

Recent studies by Greenblatt *et al.* (13) indicate that the cumulative excretion of digoxin in the urine is 7% greater following a 1-hr intravenous infusion of digoxin when compared to an intravenous injection over 3 min. This finding accounts for the difference between the 66% cumulative excretion of digoxin following the intravenous dose in this study as compared with 76% in their study (12). When this difference is considered, however, there still remains an unexplained 13% difference in the bioavailability of digoxin in the two studies. Steady-state studies suggest that the bioavailability of oral digoxin may be dose dependent (7). The differences between this study and the earlier one (5) support this possibility. Therefore, a standard dose of digoxin should be chosen for future digoxin bioavailability studies.

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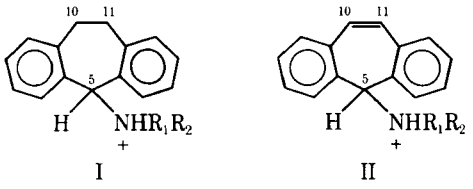
Alcoholysis of Medicinally Active 5-Aminodibenzo[*a,d*]cycloheptenes

J. NALLY, J. NAZARENO, J. POLESUK, and H. V. MAULDING ˣ

Abstract □ The rate constants for alcoholic solvolysis of the hydrochloride salts of diethylaminodibenzo[*a,d*]cycloheptene and related amino congeners were determined. The objective was a study of the comparative ease of cleavage of the C—N amino linkage by various aliphatic alcohols. The interaction of protonated amines of this series with alcoholic hydroxyls presumably leads to formation of the corresponding ethers in a manner somewhat analogous to alkoxide reaction with alkyl bromides. The methyl ether produced from solvolysis of diethylaminodibenzocycloheptene hydrochloride was isolated and identified. Methanol appears to react somewhat more rapidly with the amine hydrochlorides than other aliphatic alcohols. The latter produce almost invariant velocity constants with a given amine hydrochloride. The exception was *tert*-butanol, which resulted in k_{obs} values about one-third of those given by the other alcohols. Some velocity constants in formic and acetic acids were evaluated. Generation of carbonium ions of appreciable lifetime was indicated in formic acid by the formation of a highly colored (red-violet) solution. This color may be a manifestation of the dibenzotropylium ion.

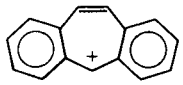
Keyphrases □ 5-Aminodibenzo[*a,d*]cycloheptenes—alcoholic solvolysis, rate constants □ Solvolysis, alcoholic—medicinally active 5-aminodibenzo[*a,d*]cycloheptenes, rate constants □ Alcohols—solvolysis of hydrochloride salts of diethylaminodibenzo[*a,d*]cycloheptene and related amino congeners, rate constants

Protonated members of the 5-aminodibenzo[*a,d*]-cycloheptane series (I) were found to be relatively unstable in aqueous solution, with rupture of the C—N linkage being the pertinent reaction (1). Insertion of a double bond between positions 10 and 11 of the cycloheptane moiety produced 5-aminodibenzo[*a,d*]cycloheptenes (II) and led to a nearly two-magnitude enhancement of the hydrolytic velocity constant. Velocity increases in each series were dependent upon the amino substituent at C-5. The rate constants for hydrolysis of these 5-amino compounds (I and II) were invariant with pH (where pH < pKa), being only a function of the nature of the various protonated amines and the specified temperature (1).



I II

R_1, R_2 = alkyl, alkyl; alkyl, hydrogen; cycloalkyl



III

Both series of compounds were previously found medicinally active as antihistaminic and anticonvulsant agents (2, 3).

The sensitivity of 5-aminodibenzocycloheptenes (II) to decomposition in aqueous solution is possibly mediated through a highly resonance-stabilized dibenzotropylium ion (III), with the positive charge being distributed over the carbon atoms of the entire molecule. However, on the basis of experimental evidence, the bimolecular attack of alcohol on the protonated amine cannot be discounted.

It became of interest to examine the reactivities of several aminodibenzocycloheptenes with ROH groups from a mechanistic viewpoint as well as from the standpoint of their tendency to function as alkylating agents. The aminodibenzocycloheptenes (II) react readily with nucleophiles such as alcoholic hydroxyls and carboxylic acid hydroxyl groups commonly found in proteins. These alkylation reactions may be related to the strong vesicatory properties of many members of this series. Sensitivity is noted with extremely dilute solutions or trace amounts of the pure material (2).

The work presented is mainly involved with the proclivity of several 5-aminodibenzocycloheptenes to interact with aliphatic alcohols, resulting in ether formation and simultaneous liberation of the primary or secondary amine during cleavage of the C—N bond (Scheme I).

EXPERIMENTAL

Preparation of Compounds—5-Aminodibenzo[*a,d*]cycloheptanes, either as the free base or hydrochloride salts, were synthesized as previously reported from 5-chlorodibenzocycloheptane[1] and the corresponding amine in benzene or toluene (1, 3).

The 5-aminodibenzo[*a,d*]cycloheptenes were isolated as the free bases or hydrochloride salts from the 5-chlorodibenzocycloheptene reaction with the corresponding liquid amine (4–6). The 5-chlorodibenzocycloheptene was prepared by treatment of the alcohol[1] with equimolar thionyl chloride in dry toluene. It was employed without further purification following removal of solvent.

Compounds utilized in this study were recrystallized and subjected to C, H, N, and Cl microanalysis. Actual values were found to be in good agreement with theoretical values (1, 6). Melting points corresponded to those previously reported for the compounds (1, 3–5) and TLC [chloroform–diethylamine (99:1)] showed only one spot.

Kinetic Procedures—Solutions of the various alcohols were equilibrated at the specified temperatures, followed by addition of 25 mg of the amine hydrochloride in solution to give a final volume of 100 ml. Four-milliliter samples were periodically withdrawn and evaporated to dryness under reduced pressure (rotary evaporator), followed by addition of 0.5 ml of 0.1 *N* HCl, 0.5 ml of 0.1 *N* NaOH, 4 ml of pH 3.44 citrate buffer[2], and 25 ml of methyl orange solution[3]. The aqueous solution was extracted three times with 30-ml portions of chloroform and made up to 100 ml in a volumetric flask. The chloroform solutions were scanned in the visible from 390 to 460 nm.

Neither the decomposition products nor their mixtures with the intact molecules in equimolar concentrations produced appreciable chloroform extractive color with methyl orange. This was noted since degraded solutions failed to interfere with the analytical procedure.

Solvent alcohols were reagent grade, anhydrous, except 95% ethanol. The other chemicals and materials used were reagent grade.

The analytical procedure was altered in the case of propylene glycol in that 2 ml of the propylene glycol containing 0.5 mg/ml amine was diluted to 50 ml, followed by adjustment of the pH to 3.44. The methyl orange was added and the method was followed as previously stated.

Degradation Products—A 250-mg sample of 5-diethylaminodibenzo[*a,d*]cycloheptene hydrochloride was heated at 50° for 18 hr in methanol solution. The resultant solution was filtered, and the filtrate was evaporated to dryness, acidified, and extracted into dry ether. The purity of the sample was checked by TLC[4] [chloroform–diethylamine (99:1)], with only one spot, R_f 0.75, being observed. The ether was evaporated, leaving an oily residue which was subjected to mass spectroscopy. The molecular ion, M^+, exhibited a value of 222, indicative of the methyl ether; NMR ($CDCl_3$): 3.3 (s, 3H, OCH_3) and 7.1 (m, 8H, aromatic protons) ppm.

Anal.—Calc. for $C_{16}H_{14}O$: C, 86.5; H, 6.3. Found: C, 86.2; H, 6.1.

RESULTS AND DISCUSSION

Rate constants were determined for the reactions depicted in Scheme I from either:

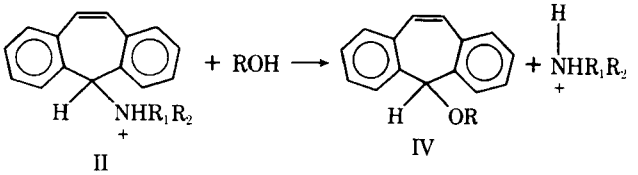
$$k_{obs} = 0.693/t_{1/2} \qquad \text{(Eq. 1)}$$

or:

$$\log (A_t - A_\infty) = \log (A_0 - A_\infty) - k_{obs}t/2.303 \qquad \text{(Eq. 2)}$$

where A_0, A_t, and A_∞ are absorbances at times = 0, t, and infinity, respectively; and k_{obs} is the observed first-order rate constant. The residual absorbance, A_∞, varied from negligible to about 2% A_0. Rate constants were evaluated in triplicate and were generally reproducible to within ±10%. Figure 1 illustrates the means of calculating k_{obs} by absorbance plots of the ion-pair complex (chloroform solution) *versus* time (Eq. 2) for several compounds in Table I.

Table I lists some alcohols examined for their effects on the reaction rate of the 5-aminodibenzocycloheptenes. Invariance of the velocity constants from alcohol to alcohol, with the exception of sterically hindered *tert*-butanol, is rather persuasive, but not conclusive, evidence of an S_N1 process; S_N processes, which for activation require only a dispersal of charge, are usually three or more times as fast in ethanol as in water (7). However, this phe-



II IV

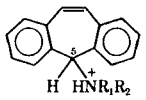
Scheme I

[1] Aldrich Chemical Co., Milwaukee, Wis.

[2] Prepared from 9.7 g of anhydrous citric acid in 900 ml of water. Thirty percent sodium hydroxide (10.5 ml) was added, and the pH was adjusted to 3.44 with concentrated hydrochloric acid, followed by dilution to 1 liter with distilled water.

[3] Two hundred and fifty milligrams of methyl orange (Eastman No. 432), 3.3 ml of 0.1 *N* NaOH, and 800 ml of water were placed in a 1-liter, volumetric flask and shaken to dissolve the contents. The pH was adjusted to 7.0 with 0.1 *N* HCl (dropwise), followed by dilution to 1 liter with distilled water.

[4] Silica gel FG, 250 μm, Analtech Inc., Newark, Del.



R_1 = alkyl, cycloalkyl, allyl
R_2 = hydrogen, alkyl

Table I—Rate Constants (k_{obs} in hours^{-1}) for Reaction of Various 5-Aminodibenzo[*a,d*]cycloheptenes in Alcohol Solution (Ether Formation) at Specified Temperatures

5-Substituent	Temperature	Water	k_{obs}[a], Alcohols: Methanol	Ethanol	Propanol	Isopropanol	Butanol	*sec*-Butanol	*tert*-Butanol	Propylene Glycol
1. Diethylamino hydrochloride	50°	0.49	1.50	1.27	1.32	2.04	1.61	1.54	0.68	1.31
2. Pyrrolidinoamino hydrochloride	60°	0.013	0.037	0.020	0.018	0.019	0.012	0.018	0.005	0.024
3. Allylamino hydrochloride	60°	0.24	0.108	0.116	0.103	0.082	0.059	0.053	0.015	0.159
4. Propylamino hydrochloride	60°	0.044	0.047	0.032	0.029	0.028	0.023	0.026	0.007	—
5. Piperidinoamino hydrochloride	60°	0.12	0.322	0.277	0.271	0.277	0.209	0.244	0.063	0.204

[a] Other conditions, listed in the order of compound, temperature, alcohol, and k_{obs}, are: diethylamine hydrochloride, 40°, ethanol, 0.26; propanol, 0.25 methanol, 0.33; and propylamino hydrochloride, 60°, neopentyl, 0.027.

nomenon also holds for reaction of neutral molecules with amine hydrochlorides. This phenomenon is manifested in the majority of the reactions of Table I.

Application of the mass law effect may be compelling evidence of an S_N1 reaction. For the sequence shown in Scheme II:

$$\overset{+}{R}NHR_1R_2 \underset{k_r}{\overset{k_f}{\rightleftharpoons}} NHR_1R_2 + R^+ \xrightarrow[k']{R'OH} ROR'$$

Scheme II

when assuming steady-state treatment, one obtains (7):

$$\text{rate} = [d\,ROR']/dt = k_f[\overset{+}{R}NHR_1R_2]\Big/\left(k_r\frac{[NHR_1R_2]}{k'} + 1\right) \text{(Eq. 3)}$$

Addition of excess amine should cause the rate of hydrolysis to slow down after some lag time. This is calculated by means of increasing the denominator in Eq. 3 through the $[NHR_1R_2]$ term. Unfortunately, the experiment cannot be carried out as with alkyl halides and halide ions because the proton exchange between added amine and protonated reactant produces precipitation of the 5-aminodibenzocycloheptene base, leaving the true mechanism to speculation. The allylic compound (Compound 3, Table I) argues against a pure S_N1 reaction due to its velocity decrease in alcoholic solution. Allylic compounds, however, are well known to undergo altered reaction mechanisms under many circumstances. This solvent effect would also be expected in the bimolecular attack of alcohol on an amine hydrochloride (7).

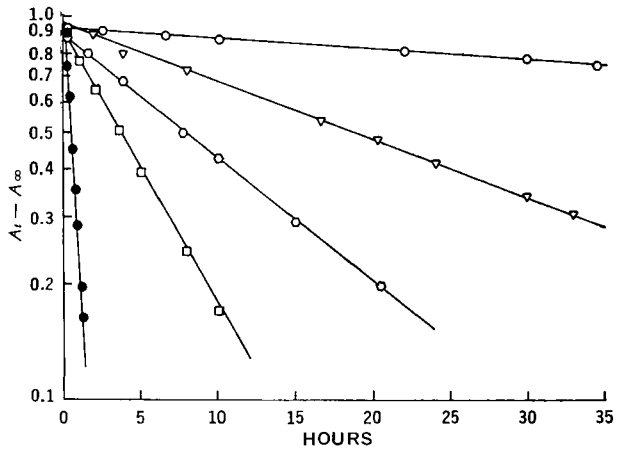


Figure 1—*Observed first-order plots for absorbance loss of various aminodibenzocycloheptenes as a function of time. The amine hydrochloride salts and alcohols utilized are listed in the key, along with the temperature for each. Key:* ●, *propylamine hydrochloride–methanol, 60°;* □, *allylamine hydrochloride–propylene glycol, 60°;* ○, *diethylamine hydrochloride–methanol, 50°;* ▽, *piperidino hydrochloride–*tert*-butanol, 60°; and* ○, *pyrrolidino hydrochloride–*tert*-butanol, 60°.*

The nonprotonated form of the compounds in Table I are unreactive, thus failing to undergo cleavage of the C—N bond which typifies this series. This could not be previously tested (1) due to the low aqueous solubility of the neutral species (free base) at pH's around and above the pKa. In alcoholic solution, this solubility problem was no longer existent and stability of the free base was observed. The addition of equimolar amounts of hydrochloric acid that protonated the amino nitrogen initiated the reaction in the usual manner.

Scheme I illustrates the expected products of the decomposition in alcoholic solution. The reaction of diethylaminodibenzocycloheptene hydrochloride in anhydrous methanol led to the anticipated methyl ether, which was isolated by preparative TLC; the structure was verified by mass spectroscopy microanalysis and NMR. In aqueous solution, three products were noted with the ether dominating (1). In the one case investigated, only the spot indicative of the methyl ether of dibenzocycloheptene was observed by TLC. No attempts were made to isolate the ethers presumably formed from the other alcohols. NMR spectroscopy shows δ 3.3 for the OCH_3 protons attached to the CH as compared to an expected value of δ 3.7 if attached to the phenyl ring. This finding corroborates a type IV structure (Scheme I).

As previously stated, the principal product in aqueous media is the bisether, which is formed both from the amine hydrochloride and from the intermediate alcohol. The ketone appears in traces and may be an artifact of oxidation of the alcohol by oxygen in the reaction solutions (1). The methyl ether appears as the only product in methanol, either through virtual absence of the alcohol in anhydrous methanol or through its low concentrations as an intermediate that prevent its observation by the methods used. The ethers, when formed, have an intrinsic stability in the reaction medium that prevents their further transformation under the reaction conditions.

Table II—Comparisons of the Observed First-Order Rate Constants (in hours^{-1}) for 5-Diethylaminodibenzocycloheptane (k_{ane}) with Those for the 5-Diethylaminodibenzocycloheptene Hydrochloride (k_{ene}) in Several Alcohols at 50°

Alcohol	k_{ane}	k_{ene}	k_{ene}/k_{ane}
Methanol	0.012	1.50	12.5
Ethanol	0.010	1.27	12.7
Isobutanol	0.0083	1.54	18.6
Isopropanol	0.0084	2.04	26.7
tert-Butanol	0.6079	0.68	8.5

Table III—Effects of Solvent Systems of Acetic Acid, Formic Acid, and Acetone on the Decomposition of 5-Diethylaminodibenzocycloheptene Hydrochloride with the k_{obs} Measured (in hours^{-1}) at 50°

Solvent[a]	k_{obs}
Acetic acid	1.46
Acetic acid + 0.5 ml of methanol	1.46
Acetic acid + 1.0 ml of methanol	1.72
Acetic acid + 5.0 ml of methanol	2.21
Formic acid, 99%	3.48
Formic acid, 99%, + 1 ml of methanol	3.52
Formic acid, 99%, + 5 ml of methanol	3.70
Reagent acetone[b]	0.41
Reagent acetone[b] + 1 ml of methanol	0.94
Reagent acetone[b] + 5 ml of methanol	1.40

[a] Run in 200-ml volumes with, for example, 5 ml of methanol added followed by formic acid addition (195 ml) to fill the volumetric flask. [b] Reagent acetone utilized from a freshly opened bottle; water content = 0.3%.

Table I shows alcohols and amines used in the investigation along with k_{obs} at specified temperatures. The following points suggest the reaction might proceed through a carbonium ion or its equivalent: *(a)* the general invariance of k_{obs} with most alcohols employed, *(b)* the slightly lower rate constant value for the hindered *tert*-butanol relative to other primary and secondary alcohols, and *(c)* the k_{ene}/k_{ane} values of greater than one magnitude. These facts, along with the comparative ease of tropylium-ion formation under the proper conditions (8), tend to support—but do not verify—a carbonium-ion hypothesis. The data for the allylic amine are disconcerting (Table I), because the observed first-order constant is smaller in alcoholic solution than in water.

Table II depicts the increase in the observed first-order rate constants with diethylaminodibenzocycloheptene hydrochloride compared with diethylaminodibenzocycloheptane hydrochloride in various alcohols at 50°. This enhancement of velocity constants in the unsaturated molecule is in agreement with the results in aqueous solution (1).

Table III shows the k_{obs} values for reaction of the diethylaminodibenzocycloheptene hydrochloride with the carboxylic hydroxyl groups of formic and acetic acids. In formic acid solution, a red-violet coloration was noted. The colored dibenzotropylium ion from the 5-chloro analog in anhydrous solvents was reported previously (9). This is circumstantial evidence for the presence of these stabilized carbonium ions under these conditions, which may be the consequence of a decreased rate of destruction of the ion rather than an increased rate of formation.

The reactivity of acetone (Table III) may be a consequence of reaction with the enolic hydroxyl as well as the water in solution.

CONCLUSIONS

The reaction of various 5-aminodibenzocycloheptenes with solvent alcohols was investigated. The k_{obs} values were found to be almost constant for a given amine in the several aliphatic alcohols studied excepting *tert*-butanol where a decrease in the observed first-order rate constant was detected.

The compounds were found to react in solutions of acetone, acetic acid, and formic acid with the latter producing a colored solution during the experiment. This may be indicative of the colored dibenzotropylium ion.

The increase in the velocity constants in alcoholic solution relative to water is characteristic of either a mechanism where only a separation of charge in the transition state is required or one representing bimolecular attack of neutral alcohol on the amine hydrochloride.

The unexpected results with the allylic compound where the k_{obs} is greater in water than in the alcohols leaves some doubts and is the principal anomaly encountered.

The magnitude of the k_{ene}/k_{ane} lends some credence to the S_N1-type mechanism, as does the general invariance of k_{obs} with alcohol employed for a given amine. Thus, the mechanism—S_N1 or S_N2—is still in doubt in this series, and a combination of the two cannot be discounted at the present time. Further experiments are in progress to elucidate the matter.

Many of these compounds are vesicant or extremely irritating. This may be a consequence of their reactivity with hydroxyls and other nucleophilic groups, such as SH, present in protein. Their relative ease of reaction may lead to alkylation of the protein materials, thus bringing about the strong response elicited upon contact with the skin.

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