α-bromo-β-methoxyisovaleric acid, which is reported to melt at 77°.[15]

Methyl β-Methoxyisovalerate.—This ester was prepared in 45% yield by esterification of the acid with methanolic hydrogen chloride, but it was found advantageous to esterify the mixture from the haloform reaction directly without isolation of the acid.

The ether residue, obtained from the haloform reaction as described above, was refluxed for eight hours with 100 g. of dry methanol containing 10% of dry hydrogen chloride; about two-thirds of the methanol was removed under reduced pressure, the residue poured into water and the solution neutralized with solid sodium carbonate. The ester layer was separated, the water layer washed with three portions of ether, and the combined ester-ether solutions dried and distilled. The ester was obtained in 29% yield (based on the methyl ketone); b. p. 57–64° (15 mm.), $n^{22}D$ 1.4158; the values agree with those of Wagner.[14]

(15) Schrauth and Geller, *Ber.*, **55**, 2788 (1922).

The second fraction, b. p. 70–85 (15 mm.), when redistilled, yielded 17% of what appeared from the analysis to be methyl α-bromo-β-methoxyisovalerate. The analytical sample boiled at 86° (15 mm.), $n^{23}D$ 1.4618.

Anal. Calcd. for $C_7H_{13}BrO_3$: C, 37.35; H, 5.82. Found: C, 37.62; H, 6.01.

Summary

The acid-catalyzed methanolysis and hydrolysis of 2,2-dimethyl-1-(3-aminopropyl)-ethyleneimine led in each case to cleavage of the bond between the tertiary carbon and the nitrogen of the imine ring. The structure of the products has been proved by synthesis, and the possible causes for the course of the cleavage reaction have been discussed.

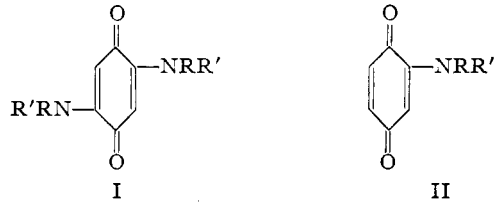
ROCHESTER, N. Y. RECEIVED DECEMBER 5, 1949

[CONTRIBUTION FROM THE STERLING-WINTHROP RESEARCH INSTITUTE]

Amino- and Ammonium-alkylaminobenzoquinones as Curarimimetic Agents

BY CHESTER J. CAVALLITO, ALBERT E. SORIA AND JAMES O. HOPPE

Recent investigations have led to the view that quaternary salts of known structure with a high curare-like physiological activity require the presence of at least two quaternary groups situated approximately 12 to 14 Å. apart in the molecule.[1,2,3,4] This communication describes a group of mono and bis quaternary derivatives of intense activity as well as the corresponding amines, some of which show unusually high curare-like activity. The active compounds described belong to class I or II in which R is H



and R′ is an alkyl chain containing a tertiary amine or quaternary group.

Quinones (or hydroquinones and oxygen) are known to react with primary or secondary amines to yield mono- and 2,5-bis-substituted quinones, I.[5,6,7,8] These compounds also may be prepared by substitution of halogen or alkoxy groups in quinones by amines.[9,10] In such compounds the nitrogen is amide-like in character. By using amino-substituted acids there have been described the preparations of acidic amino-substituted quinones by these methods.[8,11] No published information was found describing quinone bases such as might be formed from dialkylaminoalkylamines. A few such bases appear to have been prepared in Germany during the last war for study as antibiotic types.[12]

It was found that the reaction of dialkylaminoalkylamines with hydroquinone and oxygen in aqueous or ethanol solutions was not as satisfactory as the reaction of simple amines[7] under these conditions. Yields of 2,5-bis-(dialkylaminoalkylamino)-benzoquinone could be markedly improved by conducting the reaction with amine, quinone and oxygen in dioxane, acetonitrile, benzene or similar solvents. With hydroquinone, the reaction was slower and depended upon prior oxidation to quinone. With a number of amines, the monosubstituted dialkylaminoalkylaminoquinone could be isolated by carrying out the reaction in a concentrated solution from which the product crystallized. The mono- and bis-substituted aminoalkylaminoquinones described were orange or red crystalline substances which formed orange or red quaternary salts that usually crystallized.

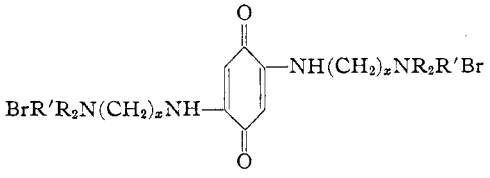
An interesting property of the 2-alkylaminoalkylaminoquinones is the ability to undergo disproportionation to the 2,5-bis derivative. In an inert solvent or neutral aqueous solutions, 2-piperidylpropylaminobenzoquinone gave a 90% yield of the 2,5-bis derivative after four days at 25°.

(1) Bovet and Bovet-Nitti, *Experientia*, **4**, 325 (1948).
(2) Barlow and Ing, *Brit. J. Pharmacol.*, **3**, 298 (1948).
(3) Kimura, Unna and Pfeiffer, *J. Pharmacol.*, **95**, 149 (1949).
(4) Paton, *J. Pharmacy and Pharmacol.*, **1**, 273 (1949).
(5) Hofmann, *Proc. Roy. Soc., London*, **13**, 4 (1863).
(6) Suida and Suida, *Ann.*, **416**, 113 (1918).
(7) Harger, THIS JOURNAL, **46**, 2540 (1924).
(8) Martynoff and Tsatsas, *Bull. soc. chim.*, **29**, 52 (1947).
(9) Kehrmann, *J. prakt. Chem.*, [2] **43**, 260 (1891).
(10) Jackson and Torrey, *Am. Chem. J.*, **20**, 395 (1898).
(11) Suchanek, *J. prakt. Chem.*, [2] **90**, 467 (1914).
(12) P. B. Report 981, 33-35 (1945), mentions that the following were prepared for study as antibiotic types: 2,5-bis-(piperidylethylamino)-benzoquinone, 2-hydroxy-5-piperidylethylaminobenzoquinone, 2,5-dichloro-3,6-bis-(diethylaminoethylamino)-benzoquinone and 2,5-bis-[*p*-(diethylaminoethoxy)-phenylamino]-benzoquinone.

The basic quinones and their quaternary salts were of mediocre antibacterial activity.[13]

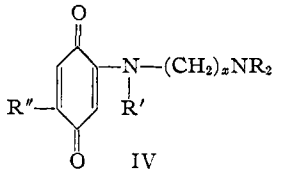
Tests for curare-like activity were carried out first with mice, then with rabbits, the data being summarized in the table. The activity of the bis-quaternary quinones, which were prepared first, was found to be greater than that of a number of bis-onium compounds of similar distance between quaternary groups. This suggested involvement of the quinone structure in eliciting the physiological response, a point borne out by the equally high activity of the mono-onium derivatives and more surprisingly so with the activity of some of the quinoneamines. A sufficiently extensive series of compounds has been prepared to allow drawing some conclusions and raising speculations relative to the relationship of chemical structure, physical–chemical properties and curare-like activity of this series.

Evidence has accumulated that the higher curare-like activity of bis-onium compounds as compared with the mono-onium type results from an ionic bonding at two points at the site of action with the former thus producing a more firm attachment than would be obtained if only one coulombic bond were involved. With the bis-onium quinones of type *III*, the distance



III

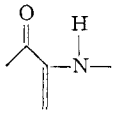
between onium nitrogen centers[14] is approximately 11 Å. where $x = 2$, 14 Å. where $x = 3$, 17 Å. where $x = 4$ and 20 Å. where $x = 5$. The two test methods show some variations in order of activity, but where R is methyl or ethyl or NR_2 is piperidyl, all of the compounds are of high activity without very much variation with change in x. Similarly, in the mono-quaternary series, activity is high and does not vary much with chain length. With the mono- and bis-aminoquinones, changes in chain length produce more dramatic changes in activity, although there is little difference between the activity of corresponding mono- and bis-substituted derivatives. With derivatives of type IV, where R′ is H and R″ is H or the same as the side chain at position 2 of the quinone, compounds where x is 2 are inert but where x



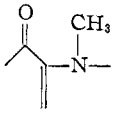
IV

is 3 to 5 may be quite active (for amines). Some correlation is possible between basicity of the nitrogen atom and activity; for example, the morpholinopropyl- and 2-(2-pyridyl)-ethyl- derivatives are much less active than the more basic piperidylpropyl- and diethylaminopropyl- derivatives. This relationship carries over somewhat to the quaternary series, in which the morpholino derivatives are less active than the others. It is also apparent that the nature of the 5-substituent in a 2-substituted member is not critical as evidenced by the similarity of activity of the mono- and bis - diethylaminobutylaminobenzoquinones and 2-diethylaminobutylamino-5-methylaminobenzoquinone. A profound loss in activity results when R′ in IV is changed from H to CH_3 whether this be with the mono or bis derivative. The activity, nevertheless, appears to be as great whether R′ is H or CH_3 when the bis derivative is quaternized. Another point of interest was the observation that with amines or quaternaries in which x is 4 or 5, the duration of head-drop in the rabbit was much more prolonged and onset was slower than with the shorter group where x is 2 or 3.

The data with the bis-onium quinones is compatible with the suggestion that bis-onium curarimimetic compounds are bound to the site of action by two acidic groups located approximately 14 Å. apart at this site. With the bis-onium quinones the difference between structure V and VI is not important in contributing to high



V



VI

activity. With the mono- or bis-alkylaminoalkylaminoquinones, structure V is essential for high activity but is insufficient in itself and requires a basic nitrogen atom at least three carbons removed from the nitrogen attached to the ring. This indicates that both structure V and the basic group are involved in bonding at the site of action. Structure V could bond by two apparent methods; by chelation with a metal atom or by hydrogen bonding through the quinone oxygen with a hydrogen atom at the site of action. Structure VI could not form metal chelates. In the series of diethylaminoalkylaminobenzoquinones it is seen that curarimimetic activity increases with increase in length of the alkyl chain up to four carbon atoms. With these, activity increases as the distance between basic nitrogen atom

(13) Tests by John W. Klimek of these laboratories showed the bacteriostatic concentrations (in mg. per cc.) with *Streptococcus pyogenes*, *Salmonella typhi* and *Mycobacterium tuberculosis* (H37Rv), respectively, of the following compounds to be: 2-(dimethylaminopropylamino)-benzoquinone, 0.05, 1.0 and 0.25; 2-(diethylaminobutylamino)-benzoquinone. 0.08, >1 and 0.35; 2,5-bis-(diethylaminoethylamino)-benzoquinone, 0.5, 0.5 and 0.8; 2,5-bis-(diethylaminopropylamino)-benzoquinone, 0.8, >1 and >1; and the bis benzochloride of the last, 0.25, >1 and 0.25.

(14) The molecule is assumed to be approximately maximally distended due to mutual repulsion of the two similarly charged groups.

and hydrogen bonding or chelating structure increases to at least approximately 9 Å. As the distance between the structure V and the basic amino group in IV increases, one might expect that physiological activity would also increase until the distance reached coincided with the distance between the two corresponding points of attachment at the site of action. Compounds of greater length would show little change in activity since the molecule could flex to the required distance between the points involved. Since the amines show greater changes in activity with changes in molecular structure than do the quaternaries, the former may be more useful in yielding information as to the nature of the site of action. This would mean that the shortest distance (9 ± Å.) between amine and structure V which yields high activity gives a clue as to the distance between the points of attachment at the site of action.

The differences in curare-like activity of the amines and quaternaries observed with changes in distance between bonding structures might be accounted for as follows. The quaternary ions of type R_4N^+ are much more basic in character than ions of type R_3HN^+.[15] At physiological *p*H values, the amines form ions of the latter type; these may be considered to have a much smaller electrostatic field than do the quaternary type. One would expect, as a result, that the onium type could vary from optimum configuration much more than could the amines, which have a smaller electrostatic field and correspondingly shorter bonding radius, without producing as marked a change in bonding ability. An anionic group at the site of action would not be able to bind an ion of type R_3NH^+ unless this group came within its immediate vicinity.[15a] This hypothesis would explain (a) the greater curare-like activity of the more basic tertiary amine quinones which could bind at greater distances from the optimum than could weaker basic groups and (b) the increase in activity upon quaternarization. In addition to basicity, steric hindrance may be involved with the amines in some of the effects produced. The lower activity of the dipropylaminopropylamino- as compared with diethylaminopropylamino- or more compact piperidylpropylamino- derivatives may be attributed to the steric hindrance offered by the alkyl groups on the basic nitrogen to approach to the site of action by the short-range bonding amine.

A remarkable correlation exists in the complementary structure of the metaloporphyrins and these curarimimetic compounds.[16]

(15) Moore and Winmill, *J. Chem. Soc.*, **101**, 1635 (1912).

(15a) The attractive force between an anion at the site of action and a highly dissociated cationic group of the curarimimetic agent is proportional to $1/r^2$ where r is the distance between these ionic groups. The attractive force between an anion and a weakly dissociated cation is proportional to a value lying between $1/r^2$ and $1/r^6$ where the latter is proportional to the attractive force between two non-ionic dipoles. This would explain the more rapid drop in activity in the amine series as compared with the quaternaries as these varied from optimum configuration.

Experimental

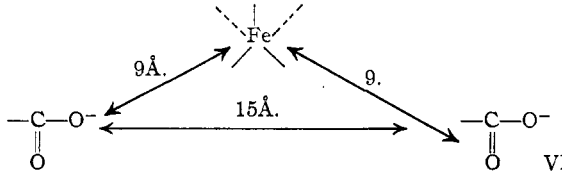
Diamines.—Diethylaminoethylamine,[17] diethylaminopropylamine[18] and aminoethylmorpholine[19] are commercially available. Diethylaminobutylamine[20] and 2-pyridylethylamine[21] were prepared by published procedures. The dimethylamino-, dipropylamino-, morpholino- and piperidyl-propylamines were prepared by addition of the appropriate secondary amine to acrylonitrile and reduction to the diamine.[22] Diethylamino- and piperidylamylamines were synthesized by reaction of diethylamino- and piperidylpropyl chlorides, respectively, with sodium ethyl cyanoacetate and proceeding as described for the preparation of the butylamino derivative.[20] The diethylaminoamylamine distilled at 80–81° at 8 mm., $n^{25}D$ 1.4450.[23] Piperidylamylamine distilled at 113–115° at 11 mm., $n^{25}D$ 1.4730.

Anal. Calcd. for $C_{10}H_{22}N_2$: N, 16.48. Found: N, 16.47.

3-Piperidylpropylmethylamine was prepared by treating piperidylpropyl chloride with methylamine; b. p. 95° at 15 mm., $n^{27}D$ 1.4642.

2-(Aminoalkylamino)-benzoquinones.—To 10.8 g. (0.1 mole) of *p*-benzoquinone in 125 cc. of dioxane was added with cooling, 0.1 mole of the diamine. Oxygen was bubbled into the dark red solution through a fritted-glass gas-absorption tube for at least three hours and up to twenty hours (overnight). During the first hour, red or orange crystalline reaction product may begin to separate out of solution. The reaction mixture was cooled, the product was filtered off and recrystallized from hot ethanol–water solution. In the preparation of 2-(piperidylamylamino)-benzoquinone it was necessary to concentrate the

(16) Using hemin as an example, one has a relationship shown by VII which includes a metal group capable of forming chelate structures and two carboxyl groups which could be involved in salt formation with the curarimimetic agent resulting from an ionic exchange for the cation normally bound to the carboxyl groups. The impor-



VII

tance of the metaloporphyrins in cellular respiration is well recognized and marked differences in specificity exist in the properties of natural physiological constituents containing such groups depending upon the characteristics of the protein fragment to which they are attached (Theorell, *Advances in Enzymology*, **7**, 268 (1947), Interscience Publishers, Inc., New York, N. Y.). It may be rational to consider the possibility that curarimimetic agents act by blocking a metaloporphyrin group attached to a specific protein or at a particularly accessible location at the neuro-muscular junction. The distance between carboxyl groups is compatible with the activity of the quinone derivatives and other bis-quaternaries and the 9 Å. distance between carboxyl and chelating groups fits excellently the relationship pointed out with the mono-onium quinones and quinone amines. Experiments with 2,5-bis-(methylamino)-benzoquinone and 2,5-bis-(dimethylamino)-benzoquinone showed that the former yielded a chelate complex with cupric acetate in aqueous solution but the latter did not.

(17) Royal Organic Chemical Co., Roselle, N. J.

(18) Sharples Chemicals, Inc.

(19) Carbide and Carbon Chemicals Corp.

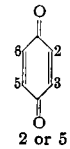
(20) Huber, Clinton, Boehme and Jackman, THIS JOURNAL, **67**, 1618 (1945).

(21) Kirchner, McCormick, Cavallito and Miller, *J. Org. Chem.*, **14**, 388 (1949).

(22) Whitmore, Mosher, Adams, Taylor, Chapin, Weisel and Yanko, THIS JOURNAL, **66**, 725 (1944).

(23) Magidson and Grigorowsky, *Ber.*, **69**, 402 (1936), give b. p., 205–208°, $n^{20}D$ 1.4540.

TABLE I



2 or 5	3,6	Quaternizing agent	Cor. m. p., °C.	Analyses, % Nitrogen Calcd.	Nitrogen Found	Cl or Br Calcd.	Cl or Br Found	Carbon Calcd.	Carbon Found	Hydrogen Calcd.	Hydrogen Found	Curarimimetic dose, mg. per kg. Inclined screen, mouse, S. C. LD_{50}	Inclined screen, mouse, S. C. ED_{50}	Head drop, rabbit, i. v ALD_{50}	Head drop, rabbit, i. v AHD_{50}
2-NH(CH$_2$)$_2$N(C$_2$H$_5$)$_2$	H		119	12.60	12.51							400	335 ± 30		
2-NH(CH$_2$)$_3$N(CH$_3$)$_2$	H		159–160	13.44	13.29			63.40	63.18	7.74	8.05	160	83 ± 6		
2-NH(CH$_2$)$_4$N(C$_2$H$_5$)$_2$	H		123–125	11.19	11.23			67.16	67.24	8.86	9.08	21	14 ± 0.7		
2-NH(CH$_2$)$_5$N(C$_2$H$_5$)$_2$	H		84–86	10.60	11.19							16	13 ± 0.6		
2-NH(CH$_2$)$_3$–NC$_5$H$_{10}$[c]	H		200–201 dec.	11.29	10.97			67.71	67.38	8.12	8.38	20	13 ± 1		
2-N–(CH$_2$)$_3$–NC$_5$H$_{10}$[c] (CH$_3$)	H		160–161	10.69	10.72			68.69	68.65	8.44	8.67	490	375 ± 40		
2-NH(CH$_2$)$_5$–NC$_5$H$_{10}$[c]	H		176–177	10.14	9.98			69.53	69.63	8.75	8.97	22	16 ± 0.8		
2-NH(CH$_2$)$_3$–NC$_5$H$_5$[d]	H		195–196	12.22	11.94			68.40	68.22	5.29	5.36	300	130 ± 12		
2-NH(CH$_2$)$_4$N(C$_2$H$_5$)$_2$ 5-NHCH$_3$	H		103–106					64.48	64.28	9.02	9.04	33	16 ± 1		
2,5-NH(CH$_2$)$_2$N(C$_2$H$_5$)$_2$	H		134–135	16.65	16.65			64.20	64.21	9.58	9.40	535	400		
2,5-NH(CH$_2$)$_3$N(CH$_3$)$_2$	H		125–126	18.17	17.65			62.35	61.85	9.15	9.16				
2,5-NH(CH$_2$)$_3$N(C$_2$H$_5$)$_2$	H		122–124	15.37	15.18			65.89	65.91	9.96	9.68	50	34 ± 2		
2,5-NH(CH$_2$)$_3$N(n-C$_3$H$_7$)$_2$	H		65–66	13.34	13.52							425	350		
2,5-NH(CH$_2$)$_4$N(C$_2$H$_5$)$_2$	H		105–106	14.28	14.12			67.45	67.69	10.28	10.41	22	11 ± 1		
2,5-NH(CH$_2$)$_5$N(C$_2$H$_5$)$_2$	H		92–95	13.32	12.96							11	9.6		
2-NH(CH$_2$)$_2$N(C$_2$H$_5$)$_2$ 5-NH(CH$_2$)$_2$–NC$_5$H$_5$[d]	H		169–172	15.71	15.20										
2,5-NH(CH$_2$)$_2$–NC$_4$H$_8$O[e]	H		189–190	15.38	15.38			59.32	59.31	7.74	7.82				
2,5-NH(CH$_2$)$_3$–NC$_4$H$_8$O[e]	H		200–201	14.28	14.16			61.19	60.98	8.22	8.50	1500	750		
2,5-NH(CH$_2$)$_3$–NC$_5$H$_{10}$[c]	H		179–181	14.42	14.26			68.00	68.24	9.34	9.59	19	9 ± 0.5		
2,5-N–(CH$_2$)$_3$–NC$_5$H$_{10}$[c] (CH$_3$)	H		117–118	13.47	13.75			69.19	69.48	9.68	9.74	1200	800		
2,5-NH(CH$_2$)$_2$N(C$_2$H$_5$)$_2$	Cl		136–137	12.93	12.92	16.36	15.92								
2-NH(CH$_2$)$_2$N(C$_2$H$_5$)$_2$	H	A	dec.[a] >225	8.84	9.28							3.6	0.9 ± 0.08		
2-NH(CH$_2$)$_6$N(C$_2$H$_5$)$_2$	H	A	113[a]	7.79	7.57	22.22	22.92					0.9	.4 ± 0.03	0.152	0.07
2-NH(CH$_2$)$_3$–NC$_5$H$_{10}$[c]	H	A	255	8.17	8.13	23.28	23.70					0.9	.33 ± 0.02	.10	.074
2,5-NH(CH$_2$)$_2$N(C$_2$H$_5$)$_2$	H	B	[a]	9.50	9.49	12.02	12.36					3.7 ± 0.3	.32 ± 0.02	.71	.34
2,5-NH(CH$_2$)$_3$N(CH$_3$)$_2$	H	B	205–225[a]	9.38[b]	9.38	11.87	11.87					1.5	.5 ± 0.05	.12	.084
2,5-NH(CH$_2$)$_3$N(C$_2$H$_5$)$_2$	H	B	191–195[a]	9.08	9.28	11.48	11.68					2.5 ± 0.1	.8 ± 0.06	.042	.029
2,5-NH(CH$_2$)$_3$N(C$_2$H$_5$)$_2$	H	A	225	10.10	10.38	28.82	28.95					1.8	.4 ± 0.02	.126	.09
2,5-NH(CH$_2$)$_4$N(C$_2$H$_5$)$_2$	H	A	230 dec.	9.66	9.31	27.53	27.65					0.7 ± 0.05	.3 ± 0.02	.146	.073
2,5-NH(CH$_2$)$_5$N(C$_2$H$_5$)$_2$	H	A	225–228 dec.	9.18	9.07	26.19	25.68					0.6	.4 ± 0.02		
2,5-NH(CH$_2$)$_3$N(C$_2$H$_5$)$_2$	Cl	B	[a]	8.17	8.22	20.65	20.60					3.5 ± 0.2	1.2 ± 0.2	.126	.080
2-NH(CH$_2$)$_2$N(C$_2$H$_5$)$_2$ 5-NH(CH$_2$)$_2$–NC$_5$H$_5$[d]	H	A	242	10.25	10.24	29.25	28.85					4.5	2.4 ± 0.2		
2,5-NH(CH$_2$)$_2$–NC$_4$H$_8$O[e]	H	A	231–234 dec.	10.10	9.84	28.02	28.12					95	33 ± 3		
2,5-NH(CH$_2$)$_3$–NC$_4$H$_8$O[e]	H	A	248 dec.	9.63	9.33	27.47	27.20					9	3.6 ± 0.1		
2,5-NH(CH$_2$)$_3$–NC$_5$H$_{10}$[c]	H	A	253 dec.	9.68	9.63	27.60	27.35					0.9	0.3 ± 0.02	.095	.066
2,5-N–(CH$_2$)$_3$–NC$_5$H$_{10}$[c] (CH$_3$)	H	A	245–250 dec.	9.25	8.71	26.39	25.95					2.0	.9 ± 0.05	.140	.080
2,5-NH(CH$_2$)$_5$–NC$_5$H$_{10}$[c]	H	A	264–270 dec.	8.83	8.90	25.19	25.25					.6	.2 ± 0.01	.132	.056
d-tubocurarine												.6 ± 0.02	.4 ± 0.02	.187	.146

[a] Melting point indefinite. [b] Dihydrate; H_2O calcd., 6.03%; found (Karl Fischer Method), 5.49%. [c] –NC_5H_{10} = piperidyl. [d] –NC_5H_5 = 2-pyridyl. [e] –NC_4H_8O = morpholino; A = methyl bromide; B = benzyl chloride.

reaction mixture to obtain separation of the product. The products progressed from orange-yellow to dark red in color with increase in molecular weight. Yields ranged from 45 to 65%. The 2-monosubstituted aminoalkylaminoquinones obtained are shown in the table; their preparation is facilitated by the low solubility in dioxane which allows separation of the desired product before reaction with a second molecule of diamine can take place.

2,5-Bis-(aminoalkylamino)-benzoquinones.—To 0.1 mole of *p*-benzoquinone in 125 to 200 cc. of dioxane was added with cooling, 0.2 mole of diamine and oxygen was bubbled into the solution for twenty hours. The reaction mixture was kept warm, where necessary, to prevent separation of the monosubstituted derivative. Upon cooling the reaction mixture the 2,5-disubstituted derivatives crystallized from solution with some of the preparations and these were recrystallized from dioxane or from hot ethanol–water solution. The solubility in dioxane of the bis derivatives obtained from dimethylaminopropyl-, dipropylaminopropyl-, diethylaminobutyl- and diethylaminoamyl-amines made it necessary to first concentrate the reaction solution and dilute with water to obtain satisfactory yields. This group could also be recrystallized from hot Skellysolve C.

These derivatives were orange to red crystalline products obtained in from 30 to 50% yield.

Mixed as well as symmetric disubstituted derivatives were also prepared by treating the isolated 2-mono substituted derivatives with a second molar equivalent of diamine and oxygen in warm dioxane (45 to 65% yields).

The substituted quinones also could be prepared from hydroquinone, diamine and oxygen, but longer reaction

time was required. The reaction of 2-chlorohydroquinone with diethylaminopropylamine yielded primarily 2,5-bis-(diethylaminopropylamino)-benzoquinone by substitution and addition of diamine.

With the use of diethylaminopropylamine, hydroquinone and oxygen in aqueous solution only a 5% yield of the bis-derivative was obtained; when a mole of acetic acid was added with each mole of diamine, an 8% yield was obtained.

2-Mono- and 2,5-Bis-(3-piperidylpropylmethylamino)-benzoquinone.—These two compounds were prepared in dioxane in the usual manner but generally were obtained as a mixture. The products were separated by concentration of the reaction mixture and recrystallization of the precipitate from hot benzene from which the monosubstituted derivative separated upon cooling. Concentration of the benzene solution and addition of Skellysolve B resulted in crystallization of the bis derivative. These two compounds were more soluble in non-polar solvents than were the analogs with a hydrogen on the nitrogen attached to the quinone ring.

2,5-Dichloro-3,6-bis-(diethylaminopropylamino)-benzoquinone.—A solution of 7.3 g. (0.03 mole) of chloranil and 15.6 g. (0.12 mole) of diethylaminopropylamine in 100 cc. of dioxane was heated on a steam-bath for six hours. The solution was concentrated to one-half volume, cooled and the crystalline precipitate filtered off. The desired product was recrystallized from hot ethanol as bronze-colored needles, yield 7.5 g. (60%).

Quaternarization

A. Benzyl Chloride Quaternaries.—A solution of 0.02 mole of dibasic quinone and 0.08 mole of benzyl chloride in 75 cc. of 95% ethanol was refluxed for four hours, cooled and diluted with ether to precipitate the bis-quaternary. The amorphous precipitate was dissolved in alcohol, treated with charcoal, filtered and reprecipitated with ether. The reprecipitation was repeated once more. The products were dried at 60° over phosphorus pentoxide and paraffin *in vacuo*. Yields were approximately 80%. The bis benzochloride from the dimethylaminopropylamine derivative formed a crystalline dihydrate; the diethylaminopropylamino derivative also crystallized. The bis-benzochlorides were red in color excepting the 3,6-dichloro- derivative which was brown and gave purple aqueous solutions.

B. Methyl Bromide Quaternaries.—Methyl bromide gas was passed into a warm solution of 0.02 mole of the aminoalkylaminoquinone in 50 cc. of ethanol and 100 cc. of dioxane for about one hour. Brick-red crystalline products separated during the reaction in some instances, in others it was necessary to add ether which precipitated amorphous quaternaries that soon crystallized upon standing. Yields of pure products were approximately 90%.

In the reaction of methyl bromide with the mono-substituted quinones to produce mono-quaternaries, the time of reaction and the heating should be kept to a minimum, otherwise considerable disproportionation to the bis compound takes place.

Basicity of the Alkylaminoalkylaminoquinones.—The relative basicity of some of the amines was determined by dissolving one millimole of the aminoquinone in 75 to 85 cc. of water to which was added enough 0.1 *N* hydrochloric acid solution to supply a 50% excess of acid above the quantity required for each basic amino group. The solution was titrated potentiometrically with 0.1 *N* sodium hydroxide solution and the *p*H was observed at which the excess acid was neutralized. This represented the pK (salt) and was a much sharper end-point than could be obtained for the pK_B. For the 2-monosubstituted quinones, the pK(salt) values were: piperidylpropylamino, 5.2; pyridylethylamino, 3.5. The 2,5-bis derivatives gave the following values for the bis hydrochlorides: morpholinopropylamino-, 3.7; diethylaminoethylamino-, 4.5; diethylaminopropylamino-, 5.5; dipropylaminopropylamino-, 5.5; piperidylpropylamino-, 5.2; piperidylpropylmethylamino, 6.0.

Biological Tests.—Preliminary tests for curare-like activity were made in mice and rabbits. Thompson's[24] inclined screen procedure for the biological assay of insulin was modified and adapted to the quantitative evaluation of curarimimetic activity in mice. Groups of ten mice were injected subcutaneously with graduated doses given in a volume of 0.01 cc. per gram of body weight and placed in 6 by 18 inch stalls on a screen inclined at an angle of 50° from the horizontal. Those mice developing typical skeletal muscle paralysis and abruptly sliding off the screen were considered positive reactors. The effective dose producing paralysis in 50% of the mice (ED_{50}) and the dose causing death by subcutaneous injection in a similar manner in 50% of the mice (LD_{50}) were estimated by the method of Miller and Tainter.[25] The more active compounds were tested by the rabbit "head-drop, HD_{50}" procedure. Graduated doses were injected intravenously at a rate of 12 cc. per minute into groups of three to ten rabbits per dose level. A positive response occurred when the head dropped forward to the supporting surface and could not be raised in response to a light tap on the back of the animal. The dose producing head-drop in 50% of the rabbits (HD_{50}) and the intravenous (LD_{50}) were also estimated by the method of Miller and Tainter.

Acknowledgments.—We wish to acknowledge the technical assistance of Mrs. J. Prudente, Miss Kathleen Kraft and Mr. D. K. Seppelin and are indebted to Mr. M. E. Auerbach, Mr. K. Fleischer and Staff for the microanalyses.

Summary

2,5-Bis-onium-alkylaminobenzoquinones are potent curarimimetic compounds. The 2-mono-onium derivatives are likewise active and represent the first examples of monoquaternaries of known structure of such order of activity (ED_{50} < 1 mg. per kg.).

Mono- and bis-aminoalkylaminobenzoquinones demonstrate unusually high curare-like activity. The activity of these is more susceptible to structural variations than is that of the quaternaries. Factors involved include basicity of the tertiary amino group, distance between the tertiary amine and quinone group and size of the alkyl groups on the basic nitrogen. The activity of the quinone amines is dependent upon the presence of a hydrogen on the nitrogen attached to the quinone ring.

Some speculations are made relative to the site of action of these drugs.

RENSSELAER, N. Y. RECEIVED OCTOBER 11, 1949

(24) Thompson, *Endocrinology*, **39**, 62 (1946).

(25) Miller and Tainter, *Proc. Soc. Exp. Biol. Med.*, **57**, 261 (1944).