Potential Antihypertensive Agents. III.[1] 3,4-Dihydro-2H-1,4-benzothiazine Derivatives

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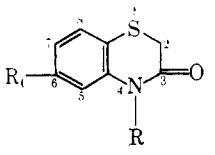
A series of N-acyl derivatives of 3,4-dihydro-2H-1,4-benzothiazine has been prepared. The carbonyl groups in some of these have been reduced by diborane. Oxidation of a few of the acyl derivatives with $KMnO_4$ gave the corresponding sulfones. Benzylation of 2H-1,4-benzothiazine-3(4H)-thione (**42**) resulted in dethiation to 1 (R = R^1 = H). The latter, on further benzylation, gave the N-benzyl derivative (**44**). Benzylation of 2H-1,4-benzothiazine-3(4H)-thione 1,1-dioxide (**32**) gave the corresponding S-benzyl derivative (**33**). In animal screening compounds **3**, **4**, **12**, **26**, and **27** had antihypertensive activity.

Pharmacological screening in our laboratories showed that 2H-1,4-benzothiazin-3(4H)-one (**1**, R = R^1 = H), its 4-allyl (**1**, R = $CH_2CH{=}CH_2$; R^1 = H), 4-allyl-6-trifluoromethyl (**1**, R = $CH_2CH{=}CH_2$; R^1 = CF_3), 4-allyl-6-chloro (**1**, R = $CH_2CH{=}CH_2$; R^1 = Cl), and 6-chloro-4-propargyl (**1**, R = $CH_2C{\equiv}CH$; R^1 = Cl) derivatives,[2] and 4-ethyl-3,4-dihydro-2H-1,4-benzothiazine[2] (**2**, R = C_2H_5) had blood pressure reducing properties of short duration in experimental animals.



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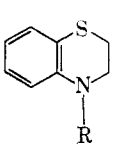

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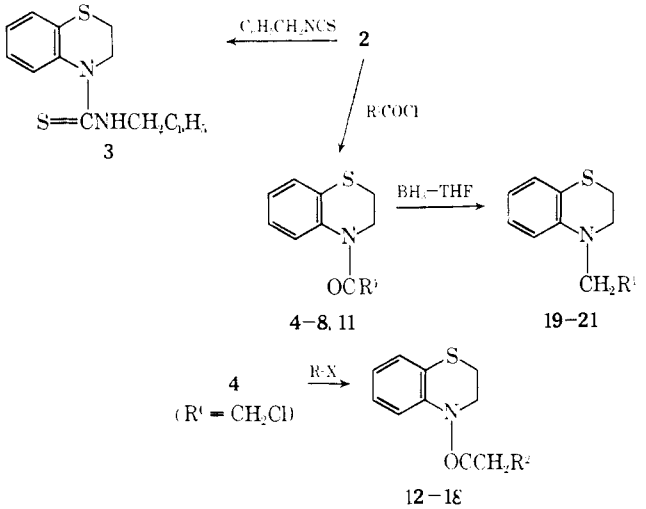
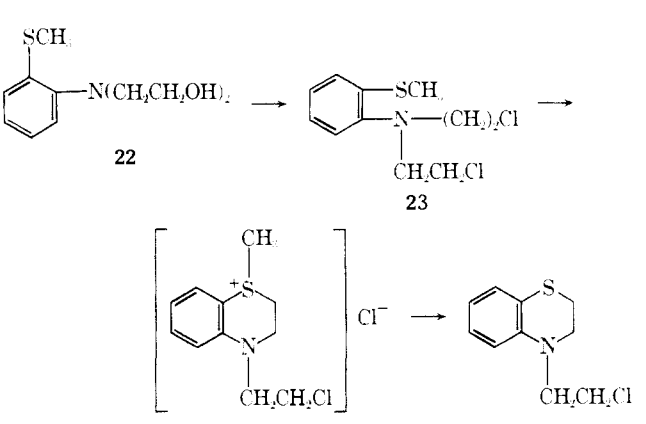
In view of this suggestive antihypertensive activity, it seemed advisable to explore the hypotensive properties of 3,4-dihydro-2H-1,4-benzothiazine derivatives somewhat further.

Chemistry.—This paper describes the synthesis and pharmacological properties of a number of new N-substituted 3,4-dihydro-2H-1,4-benzothiazines (**3–21, 26, 27**) as well as a few other new derivatives (**32, 33, 35, 37**) of 3(4H)-oxo-2H-1,4-benzothiazine 1,1-dioxide (**31**).

SCHEME I

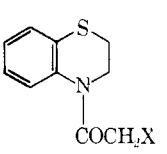


SCHEME II

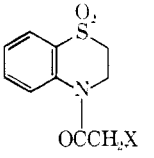


The synthetic steps leading to the formation of compounds **3–21** (Table I) are outlined in Scheme I, (where OCR^1, CH_2R^1, and $OCCH_2R^2$ = R of Table I), and are described in the Experimental Section. A different route for the preparation of 4-(β-chloroethyl)-dihydrobenzothiazine (**19**) is shown in Scheme II.

The attempted distillation of **23** led to its cyclization to **19** with the elimination of methyl chloride, probably through the sulfonium ion intermediate **24**. This reaction is similar to the one reported previously[2] for the formation of 2H-1,4-benzothiazin-3(4H)-one from 2-(α-chloroacetamido)phenylalkyl (and -aralkyl) sulfide. $KMnO_4$ oxidation of N-chloroacetyl (**4**) and N-azidoacetyl (**12**) derivatives of **2** (R = H) gave the corresponding sulfones (**26, 27**). Preparative details



4, X = Cl
12, X = N_3



26, X = Cl
27, X = N_3

of the sulfones **26–30** (Table II) are described in the Experimental Section.

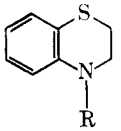
The preparation of S-benzyl (**33**) and C-benzyl (**35**) (Table III) derivatives of 3-thiosulfones are outlined in Scheme III. The proof that the benzylation of **32** gave the S-benzylated derivative (**33**) and not the C-benzylated thione (**35**) was found in the reaction of

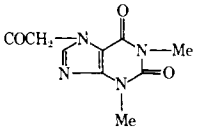
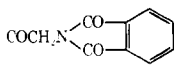
1) For paper II, see R. N. Prasad, L. R. Hawkins, and K. Tiedje, *J. Med. Chem.*, 11, 1144 (1968).

2) R. N. Prasad and K. Tiedje, *Can. J. Chem.*, **44**, 1247 (1966).

TABLE I

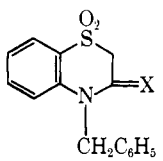
3,4-DIHYDRO-4-SUBSTITUTED 2H-1,4-BENZOTHIAZINES



No.	R	Method	Recrystn solvent[a]	Yield, %[b]	Mp or bp (mm), °C	Formula	Analyses
3	$CSNHCH_2C_6H_5$		P_2	66	92–92.5	$C_{16}H_{16}N_2S_2$	C, H, N, S
4	$COCH_2Cl$	A	E + P_1	50	57–58	$C_{10}H_{10}ClNOS$	C, H, Cl, N, S
5	$COCH_2OC_6H_5$	B	M	63	96.5–98	$C_{16}H_{15}NO_2S$	C, H, N, S
6	$COCH(CH_3)OC_6H_5$	B	M	90	127–129	$C_{17}H_{17}NO_2S$	C, H, N, S
7	$COCH{=}CH_2$[c]	B		60	125–130 (0.3)	$C_{11}H_{11}NOS$	C, H, N, S
8	$COCH{=}CHC_6H_5$	B	E + P_1	45	77.5–80	$C_{17}H_{15}NOS$	C, H, N, S
9	$SO_2C_6H_4NO_2$-*p*		Et	56	130–131	$C_{14}H_{12}N_2O_4S_2$	C, H, N, S
10	$SO_2C_6H_4NH_2$-*p*		M	53	158.5–160	$C_{14}H_{14}N_2O_2S_2$	C, H, N, S
11	$COOC_2H_5$			30	146 (0.1)	$C_{11}H_{13}NO_2S$	C, H, N, S
12	$COCH_2N_3$	D	Ea + P_2	99	105–106	$C_{10}H_{10}N_4OS$	C, H, N, S
13		E	C + B	90	201–203	$C_{17}H_{17}N_5O_3S$	C, H, N, S
14	$COCH_2NH_2$		Et	71	261–264	$C_{10}H_{12}N_2OS \cdot HCl$	C, H, Cl, N, S
15	$COCH_2SC_6H_4NH_2$-*o*	F	P_2	85	117–117.5	$C_{16}H_{16}N_2OS_2$	C, H, N, S
16	$COCH_2SC_6H_5$	F	E + P_1	64	62–63	$C_{16}H_{15}NOS_2$	C, H, N, S
17	$COCH_2I$	C	B + P_2	45	114–116	$C_{10}H_{10}INOS$	C, H, N, I
18		E	Et	53	242–243	$C_{18}H_{14}N_2O_3S$	C, H, N, S
19	CH_2CH_2Cl	G	E + P_1	50	55–57, 142–143 (0.4)	$C_{10}H_{12}NS$	C, H, Cl, N, S
20	$CH_2CH_2OC_6H_5$	G		80	175–179 (0.15–0.20)	$C_{16}H_{17}NOS$	C, H, N, S
	$CH_2CH_2OC_6H_5$[d]		A		112–113.5	$C_{24}H_{27}NO_4S_2$	C, H, N, S
21	$CH_2CH(CH_3)OC_6H_5$	G			165–169 (0.15–0.17)	$C_{17}H_{19}NOS$	C, H, N, S
25	CH_2CH_2I	C	Pn	61	81–82	$C_{10}H_{12}INS$	C, H, I, N

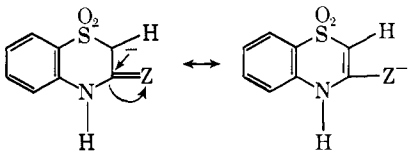
[a] A, Me_2CO; B, C_6H_6; C, $CHCl_3$; E, Et_2O; Ea, EtOAc; Et, EtOH; M, MeOH; P_1, petroleum ether (30–60); P_2, petroleum ether (60–80); Pn, pentane. [b] Yields given are those of crude solids or once-distilled liquid. [c] $n^{25}D$ 1.6427. [d] Methyl tosylate (from 3,4-dihydro-4-(phenoxyethyl)-2H-1,4-benzothiazine and methyl *p*-toluenesulfonate in boiling MeOH) of **20**.

34[2] with P_2S_5, which yielded **35**. The alternate possibility (**37**) was eliminated by its synthesis from the N-benzylated 3-oxo derivative **36**[2] and P_2S_5. This



36, X = O
37, X = S

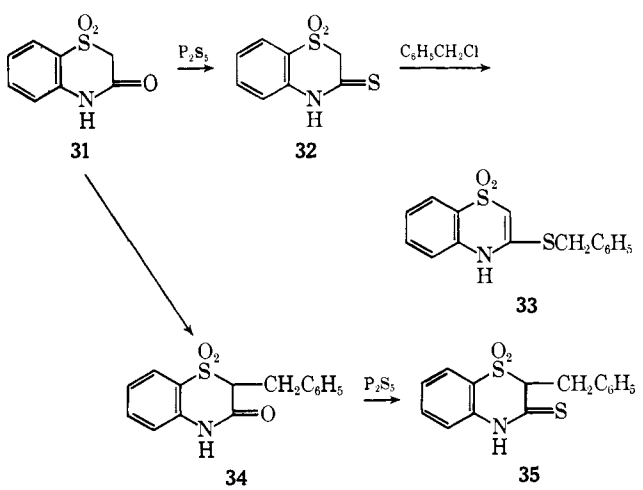
difference in benzylation of **32** (which gives the S-benzyl derivative **33**) compared to the benzylation of **31** (which gives the C-benzyl derivative **34**) is noteworthy. The presence of the sulfonyl function in **31** and **32** clearly renders the CH_2 group sufficiently acidic to give a stabilized anion in the presence of alcoholic NaOH in both cases. Alkylation of the anion (**38** ↔ **40**) then occurs at the more nucleophilic negative carbon



38, Z = O
39, Z = S

40, Z = O
41, Z = S

SCHEME III



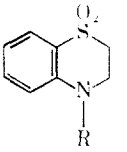
atom to give the C-benzyl derivative **34**, in analogy to the usual course of enolate ion alkylations.[3] In contrast, a negatively charged sulfur atom is known to be strongly nucleophilic and therefore alkylation of the anion (**39** ↔ **41**) leads to the S-benzyl derivative **33**.

The sulfur atom is more nucleophilic than the

(3) H. O. House, "Modern Synthetic Reactions," W. A. Benjamin, Inc., New York, N. Y., 1965, p 163.

TABLE II

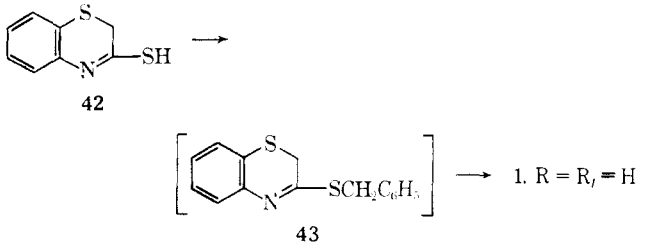
3,4-DIHYDRO-4-SUBSTITUTED 2H-1,4-BENZOTHIAZINE 1,1-DIOXIDE



No.	R	Method	Recrystn solvent[a]	Yield, %[b]	Mp, °C	Formula	Analyses
26	$COCH_2Cl$	H	B + P_1	73	98–99	$C_{10}H_{10}ClNO_3S$	C, H, Cl, N, S
27	$COCH_2N_3$	D, H	Ea + P_2	54	111–112	$C_{10}H_{10}N_4O_3S$	C, H, N, S
28	$COC_6H_4NO_2$-*p*	H	A + B	84	206–207	$C_{15}H_{12}N_2O_5S$	C, H, N, S
29	$COCH_2SO_2C_6H_5$	H	B	100	161–162	$C_{16}H_{15}NO_5S_2$	C, H, N, S
30	$COC_6H_4NH_2$-*p*		Ea	30	216–217	$C_{15}H_{14}N_2O_3S$	C, H, N, S

[a] A, Me_2CO; B, C_6H_6; Ea, EtOAc; P_1, petroleum ether (30–60°); P_2, petroleum ether (60–80°). [b] Yields given are those of crude solids or once-distilled liquid.

nitrogen atom (*viz.* in thiourea). This may account for the fact that attempted benzylation of 2H-1,4-benzothiazine-3-thiol[4] (**42**) resulted in isolation of only 3(4H)-oxo-2H-1,4-benzothiazine (**1**, R = R¹ = H) in over 80% yield, since the S-alkylated product **43**, which may have formed first, would very easily be cleaved.



42

43

There was no formation of the possible N-benzyl derivative (**45**, X = S), which was prepared from 4-benzyl-3-oxo-3,4-dihydro-2H-1,4-benzothiazine[2] (**44**, X = O) and P_2S_5. Similar alkylation of **1** (R = R¹ = H) gave only 10% of **44** after a 1-hr reaction period, but this yield was raised to 50% after 6 hr. Failure to

44, X = O
45, X = S

isolate any N-benzylation product (**36**) during the alkylation of **31**, may be due to faster alkylation of the carbon atom and the presence of the sulfonyl function (to give **34**).

Pharmacology.—All the compounds reported have been tested for their antihypertensive activity by the method described before.[5] The criterion for activity was a fall in blood pressure in cats of at least 20 mm for at least 15 min. Of all the compounds tested, only the N-chloroacetyl derivatives **4** and **26** and the thioureido derivative **3** were found to exhibit activity. Neither of the two chloroacetyl derivatives altered the pressor response to carotid occlusion, but **26** was found to reduce the pressor action of epinephrine. The azides **12** and **27** showed moderate hypotensive properties, **27** having a more sustained effect than **12**. The rest of the compounds showed either very weak hypotensive properties or were inactive.

Experimental Section[6]

4-(N-Benzylthiocarbamoyl)-3,4-dihydro-2H-1,4-benzothiazine (3).—A mixture of 3,4-dihydro-2H-1,4-benzothiazine (7.6 g, 0.05 mole) and benzyl isothiocyanate (7.45 g, 0.05 mole) in absolute EtOH (80 ml) was left overnight at room temperature. The mixture was then heated slowly on the steam bath until most of the EtOH had evaporated (4 hr). The oily residue, on trituration with Et_2O–petroleum ether (30–60°), solidified. Two recrystallizations gave the pure product as white shining crystals.

Method A. 4-(Chloroacetyl)-3,4-dihydro-2H,1,4-benzothiazine (Table I, 4).—A solution of chloroacetyl chloride (22.6 g, 0.2 mole) in Et_2O (100 ml) was added dropwise (1 hr) to a solution of 3,4-dihydro-2H-1,4-benzothiazine (**2**, R = H) (30.2 g, 0.2 mole) in dry Et_2O (500 ml) containing Et_3N (21.2 g, 0.21 mole), at 10–15°. After completion of the addition, the mixture was refluxed for 1 hr and filtered. The filtrate was washed (dilute HCl, cold H_2O), dried, and evaporated (35–40°) and the viscous residue was triturated with Et_2O–petroleum ether (30–60°) to give the desired product.

Method B was the same as method A, except that pyridine was used as the acid acceptor and the reaction mixture was allowed to stand overnight at room temperature.

4-Carbethoxy-3,4-dihydro-2H-1,4-benzothiazine (11).—A mixture of **2** (R = H) (15.1 g, 0.1 mole) and ethyl chloroformate (10.8 g, 0.1 mole) was heated over a steam bath for 4 hr. At the end of this period, the mixture was distilled under reduced pressure, and the fraction (19.2 g, 86%) boiling at 150–160° (1.3 mm) was collected and redistilled.

3,4-Dihydro-4-(*p*-nitrobenzenesulfonyl)-2H-1,4-benzothiazine (9).—A solution of *p*-nitrobenzenesulfonyl chloride (22.2 g, 0.1 mole) in pyridine (30 ml) was added dropwise to a solution of **2** (R = H) (15.1 g, 0.1 mole) in pyridine (30 ml) at 10–20°. The mixture was then heated and a temperature of 85° was maintained for 5 min. The mixture was poured onto ice-water and the

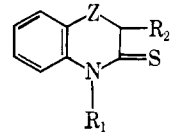
(4) A. I. Kiprianov and T. M. Verboskaya, *Zh. Obshch. Khim.*, **32**, 3946 (1962); *Chem. Abstr.*, **58**, 12707 (1963).

(5) F. Fried, R. N. Prasad, and A. P. Gaunce, *J. Med. Chem.*, **10**, 278 (1967).

(6) Melting points were determined with a Thomas–Hoover capillary melting point apparatus. Both melting points and boiling points are uncorrected. Ir spectra were obtained with a Beckman IR-8 spectrophotometer. Analyses were carried out by the Abbott Microanalytical Laboratory, North Chicago, Ill. The nmr spectra, kindly provided by Dr. M. I. Levenberg and R. Egan of the Chemical Physical Laboratory, Abbott Laboratories, North Chicago, Ill., were recorded on a Varian A-60 spectrometer at 60 MHz. The spectra were measured on approximately 10% (w/v) solutions in DMSO-d_6 with Me_4Si as an internal standard. General synthetic procedures given refer to the compounds listed in Tables I and II. Physical properties of analytical samples are also recorded in the tables. Where analyses are indicated either in the experiments described herein or in the tables only by symbols of the elements, analytical results obtained for these elements were within ±0.4% of the theoretical values.

TABLE III

3-THIO-3,4-DIHYDRO-2H-1,4-BENZOTHIAZINES



No.	R_1	R_2	Z	Recrystn solvent[a]	Yield, %[b]	Mp, °C	Formula	Analyses
32	H	H	$>SO_2$	A + P_2	66	184–186	$C_8H_7NO_2S_2$	C, H, N, O, S
35	H	$CH_2C_6H_5$	$>SO_2$	C + P_1	60	188–190	$C_{15}H_{13}NO_2S_2$	C, H, N, O, S
37	$CH_2C_6H_5$	H	$>SO_2$	C + P_1	100	134–135	$C_{15}H_{13}NO_2S_2$	C, H, N, O, S
42	H	H	S	Et	39	126–128[c]	$C_8H_7NS_2$	N, S
45	$CH_2C_6H_5$	H	S	E + H	63	92–94	$C_{15}H_{13}NS_2$	C, H, N, S

[a] A, Me_2CO; C, $CHCl_3$; E, Et_2O; Et, EtOH; H, hexane; P_1, petroleum ether (30–60°); P_2, petroleum ether (60–80°). [b] Yields given are those of crude solids. [c] Lit.[3] mp 128°.

precipitate was filtered and washed thoroughly (H_2O) to give the product.

4-(*p*-Aminobenzenesulfonyl)-3,4-dihydro-2H-1,4-benzothiazine (10).—A mixture of **9** (33.2 g, 0.1 mole) and Sn granules (23.7 g, 0.2 g-atom) in 15% aqueous HCl (300 ml) was stirred at 90–95° for 2 hr. At the end of this period, the reaction mixture was cooled and basified with 20% aqueous NaOH solution. The gummy residue, so obtained, was thoroughly washed (H_2O) to give the product.

Method C. 4-(Iodoacetyl)-3,4-dihydro-2H-1,4-benzothiazine (17).—A solution of NaI (6.0 g, 0.04 mole) in dry MeOH (200 ml) was boiled gently with **4** (6.82 g, 0.03 mole) on the steam bath, until the volume was nearly halved. The concentrated solution was refluxed for 24 hr and filtered hot. The filtrate, on cooling, deposited 4.4 g (45%) of **17**.

4-(β-Iodoethyl)-3,4-dihydro-2H-1,4-benzothiazine (25) was similarly prepared by refluxing **19** and NaI in Me_2CO for 40 hr.

Method D. 4-(Azidoacetyl)-3,4-dihydro-2H-1,4-benzothiazine (12).—A solution of **4** (6.82 g, 0.03 mole) in Me_2CO (50 ml) was added to a well-stirred suspension of NaN_3 (2.6 g, 0.04 mole) in 15% aqueous Me_2CO (180 ml). The mixture was refluxed for 5 hr and filtered hot. The filtrate on evaporation gave a quantitative yield of **12**. Three recrystallizations gave the pure product.

Method E. 3,4-Dihydro-4-(phthalimidoacetyl)-2H-1,4-benzothiazine (18).—A mixture of **4** (11.37 g, 0.05 mole), potassium phthalimide (9.3 g, 0.05 mole), and Et_3N (15 ml) in DMF (100 ml) was stirred on the steam bath for 24 hr. At the end of this period, the mixture was poured into cold H_2O (1 l.), with stirring, and the product was filtered. Two recrystallizations from EtOH (4 l.) gave the pure product.

4-(Aminoacetyl)-3,4-dihydro-2H-1,4-benzothiazine hydrochloride (14) was obtained by reducing **18** in absolute EtOH (100 ml) with 1 equiv of 95% $(NH_2)_2$.[7]

Method F. 4-(*o*-Aminobenzenethioacetyl)-3,4-dihydro-2H-1,4-benzothiazine (15).—A solution of **4** (11.37 g, 0.05 mole) in EtOH (200 ml) was added dropwise to a refluxing solution of *o*-aminobenzenethiol (6.25 g, 0.05 mole) in ethanolic KOH (3.08 g, 0.055 mole in 100 ml EtOH) in 15 min. After a 4-hr reflux period, the mixture was filtered hot and the filtrate was evaporated. The residue, on trituration with EtOH–Et_2O, gave the product.

Method G. (i) **4-(β-Chloroethyl)-3,4-dihydro-2H-1,4-benzothiazine (19)** was obtained in 50% yield by the reduction of the crude **4** (obtained from 0.2 mole of **2**, R = H) in THF (100 ml) by diborane (0.446 mole) in THF, as described before.[2] The product was isolated from the final basic solution by filtration.

Compounds **20** and **21** were obtained similarly by the reduction of **5** and **6**, respectively.

(ii) **By Rearrangement of 23.**—$SOCl_2$ (139 g, 1.16 moles) in $CHCl_3$ (300 ml) was added dropwise to a solution of N,N-bis(2-hydroxyethyl)-*o*-methylthioanisidine[8] (**22**) (132.5 g, 0.583 mole) in $CHCl_3$ (300 ml), at −5°, over a 30-min period. The mixture was stirred at room temperature for 4 hr and allowed to stand at room temperature overnight. The solvent was then evaporated under reduced pressure in a bath at 30–40°. The residue was diluted with C_6H_6 and the mixture was poured onto cold H_2O. The organic phase was separated and the aqueous layer was extracted (C_6H_6, three 50-ml portions). The combined organic layer was washed (H_2O), dried, and filtered. The filtrate was evaporated (below 40°) to give a viscous residue. The viscous residue was extracted with *n*-hexane and the solution was evaporated under reduced pressure below 40° to give the product (31 g, 21%). Two crystallizations from Et_2O containing some MeOH, gave N,N-bis(β-chloroethyl)-*o*-methylthioanisidine (**23**), mp 116–119°. *Anal.* ($C_{11}H_{15}Cl_2NS$) C, H, Cl, N, S.

When an attempt was made to distil the viscous residue (**23**), the only product isolated was **19**, which solidified on standing, melting at 56–59°. This did not depress the melting point of **19** obtained by the first method (i).

Method H. 4-(Chloroacetyl)-3,4-dihydro-2H-1,4-benzothiazine 1,1-dioxide (Table II, 26) was prepared in 73% yield by the $KMnO_4$ oxidation of **4**, as described before.[2]

Similar oxidations of **12**, 3,4-dihydro-**4**-(*p*-nitrobenzoyl)-2H-1,4-benzothiazine,[2] and **16** gave **27**, **28**, and **29**, respectively.

4-(*p*-Aminobenzoyl)-3,4-dihydro-2H-1,4-benzothiazine 1,1-Dioxide (30).—$SnCl_2$ (33.18 g, 0.17 mole) in concentrated HCl (100 ml) was added dropwise to a stirred solution of **28** (16.6 g, 0.05 mole) in concentrated HCl (200 ml) at 60°. After the initial tendency to frothing was over, the mixture was refluxed for 5 hr and then filtered. The residue (14.5 g, 43%) was washed with concentrated HCl and Et_2O. This hydrochloride salt (mp 280–285°) was dissolved in H_2O (400 ml) and basified with NaOH and the free base was extracted ($CHCl_3$). The extract was washed (cold H_2O), dried, and evaporated. The residue was crystallized once with ether–petroleum ether (30–60°) and then with EtOAc to give pure **30**.

2H-1,4-Benzothiazine-3(4H)-thione 1,1-Dioxide (32).—A mixture of 3(4H)-oxo-2H-1,4-benzothiazine 1,1-dioxide[2] (9.85 g, 0.05 mole) and P_2S_5 (11.10 g, 0.05 mole) in pyridine (160 ml) was stirred and refluxed for 1 hr, and then the reaction mixture was concentrated under reduced pressure. The residual liquid (*ca.* 30 ml) was stirred with a 10% NaOH solution (160 ml) at 10° for 30 min and filtered (Norit). The filtrate, on acidification, gave the product.

3-Benzylthio-4H-1,4-benzothiazine 1,1-Dioxide (33).—A mixture of benzyl chloride (3.2 g, 0.025 mole) and **32** (5.3 g, 0.025 mole) in 50% EtOH (50 ml) containing NaOH (1.0 g, 0.025 mole) was refluxed for 1 hr and then allowed to stand at room temperature for 4 days. The reaction mixture was then diluted (H_2O, 100 ml) and extracted successively with petroleum ether (30–60°, two 50-ml portions) and Et_2O (30 ml). The aqueous layer was filtered, and the cream-colored residue (6.2 g, 81%, mp 176–177°) was recrystallized twice from dilute EtOH to give the pure product: mp 177–178°; nmr, 658 Hz (s, NH). *Anal.* ($C_{15}H_{13}NO_2S_2$) C, H, N, S.

2-Benzyl-2H-1,4-benzothiazine-3(4H)-thione 1,1-dioxide (35) was prepared in 69% yield from 2-benzyl-3(4H)-oxo-2H-1,4-benzothiazine 1,1-dioxide[2] (**34**) by a method similar to that used for the preparation of **32**, except that the product was isolated by extraction with $CHCl_3$; nmr, 582 (NH), 302 (q, CH), 200 Hz (m, CH_2). Addition of D_2O exchanged the methine proton in **35**,

(7) L. I. Smith and O. M. Emerson, "Organic Syntheses," Coll. Vol. III, John Wiley and Sons, Inc., New York, N. Y., 1955, p 153.

(8) M. Freifelder and G. R. Stone, *J. Org. Chem.*, **26**, 1477 (1961).

and, as the CH coupling was removed, the ABX pattern simplified to a AB quartet centered at 198 Hz.

Similarly, thiation of 4-benzyl-3-oxo-3,4-dihydro-2H-1,4-benzothiazine 1,1-dioxide[2] (**36**) (2.87 g, 0.01 mole) by P_2S_5 (2.22 g, 0.01 mole) in refluxing dioxane (50 ml) gave 4-benzyl-2H-1,4-benzothiazine-3(4H)-thione 1,1-dioxide (**37**).

2H-1,4-Benzothiazine-3(4H)-thione[4] (**42**) was obtained as yellowish green crystals in 39% yield from 1 (R = R' = H) and P_2S_5 using dioxane as a solvent instead of toluene as reported by Kiprianov, *et al.*,[4] mp 126–128° (EtOH).

4-Benzyl-3,4-dihydro-2H-1,4-benzothiazine-3-thione (**45**) was prepared from 4-benzyl-3-oxo-3,4-dihydro-2H-1,4-benzothiazine[2,8] (**44**) (5.1 g, 0.02 mole) by the method used for the preparation of **32**, except that the product was isolated by extraction with Et_2O.

(8) Compound **44** was obtained in 55% yield by refluxing **1** (R = R' = H) for 6 hr with 10% excess benzyl chloride and NaOH in 60% EtOH. Reported[2] method required the use of NaH in DMF.

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The Activity of Phenothiazine Anthelmintics as Related to Semiquinone Formation

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Substitutions of various groups on the phenothiazine nucleus have been studied with respect to their effects on standard electrode potential, semiquinone free-radical stability, and anthelmintic activity. The electrode potentials of the two oxidation steps are correlated on Hammett plots. An expression is derived for the relative semiquinone concentration occurring in a biological system having a definite oxidation potential and pH. The anthelmintic activity is shown to be related to the semiquinone concentration.

Craig, and coworkers,[1] studied a series of substituted phenothiazines with regard to potentiometric titration, electrode potentials, and their correlation with anthelmintic activity as measured in the biological assay using mixed infestations of *Syphacia obvelata* and *Aspiculuris tetraptera* in mice. From these studies it appeared that two factors were necessary for activity, namely, the ability to form a high proportion of a stable semiquinone radical (as measured by the index potential in aqueous AcOH), and the presence of a free 3 or 7 position.

In addition to the two factors above, Craig, *et al.*,[1] also noted that only those compounds with electrode potentials in the range of 550–850 mV in aqueous AcOH had significant activity. If the toxic or paralyzing effect of the phenothiazines is due to an inhibition by the semiquinone of an oxidation–reduction system in the parasite, it would seem reasonable that the active phenothiazines would have reduction potentials corresponding to those of the oxidation–reduction enzyme system or systems which they inhibit. At the similar potentials the semiquinone concentration would be maximal and thus facilitate or compete with the electron transfers in the enzyme system involved. For example, it has been suggested that the semiquinone of chlorpromazine is responsible for the inhibition of certain oxidoreductases *in vitro*[2-4] and that some of the biological activities of phenothiazines correlate with the formation of their semiquinones *in vivo*.[5,6]

In this report the substituted phenothiazines studied by Craig, *et al.*,[1] are examined to discover a possible relationship between the calculated relative concentration of their semiquinones *in vivo* and their anthelmintic activities. It is first established that the electrode potentials for the two one-electron oxidation steps leading to the phenazothionium ion are linear functions of the Hammett substituent constants. The semiquinone concentration then becomes a function of the Hammett constant and the prevailing electromotive force and pH at the site of action. The results are shown to be not inconsistent with the supposition that the biological action is a result of the interference of the semiquinone with an essential oxidation–reduction system in the parasite.

Reduction Potentials.[7]—Let E represent the local

(1) (a) J. Cymerman-Craig, M. E. Tate, G. P. Warwick, and W. P. Rogers, *J. Med. Pharm. Chem.*, **2**, 659 (1960); (b) J. Cymerman-Craig, M. E. Tate, F. W. Donovan, and W. P. Rogers, *ibid.*, **2**, 669 (1960); (c) J. Cymerman-Craig and M. E. Tate *Progr. Drug Res.*, **3**, 76 (1961); (d) W. P. Rogers, J. Cymerman-Craig, and G. P. Warwick, *Brit. J. Pharmacol.*, **10**, 340 (1955).

(2) M. Wolleman and P. Elodi, *Biochem. Pharmacol.*, **6**, 228 (1961).

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(5) H. Laborit, U. S. Psychopharmacology Service Center Bulletin, Vol. 2, 1962–1963, p 34.

(6) I. S. Forrest and F. M. Forrest, *Exp. Med. Surg.*, **21**, 231 (1963).

(7) The following treatment of the electrode potentials makes use of the conventions and definitions adopted at the XVIIth Conference of the International Union of Pure and Applied Chemistry, Stockholm, 1953; see J. A. Christiansen, *J. Amer. Chem. Soc.*, **82**, 5517 (1960). In addition, all potentials are referred to the standard H electrode, and the electrode potentials for the two univalent oxidation steps are defined as follows: E_1 is the normal electrode potential under the condition that phenothiazine and its semiquinone are of equal activity; E_2 is the normal electrode potential at a specified pH under the condition that the semiquinone and the phenazothionium ions are of equal activity. As considered in greater detail in eq 5 and 6, the potentials E_1 and E_2 are related to the standard electrode potentials $E_1°$ and $E_2°$ and the bivalent midpoint electrode potential E_m (called the "mean normal potential" by L. Michaelis and M. P. Schubert, *Chem. Rev.*, **22**, 437 (1938)) as follows.

$$E_1 = E_1° \quad E_2 = E_2° + (RT/F) \ln [H^+] \quad E_m = (E_1 + E_2)$$

In the factor RT/F, R is the gas constant, T the absolute temperature, and F is the value of the faraday, all expressed in consistent units.