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Structure-Activity Relationships in Membrane-Perturbing Agents

Hemolytic, Narcotic, and Antibacterial Compounds

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SUMMARY

Extrathermodynamic structure-activity correlations have been formulated for various sets of congeners causing hemolysis. The similarity of these equations to those correlating bacterial action and narcosis is discussed.

INTRODUCTION

In extending our studies (1–6) of the correlation of chemical structure with biological activity of organic compounds, it seemed that an analysis of the studies on hemolysis would yield interesting comparative results. The erythrocyte has often been used as a model system for the study of cellular processes. Our interest is, in particular, centered on the action of simple organic compounds on the rupture of the red cell and comparison of this process with the toxicity of such compounds toward bacteria. It is also of interest to compare action on erythrocytes with the binding of organic compounds to proteins in order to obtain a better understanding of the dynamics of the processes.

In correlating the effects of a set of congeners on a given biological system with chemical structure we have, as a first approximation, divided chemical properties into three classes: electronic, steric, and hydrophobic. In order to deal with large numbers of different systems, it is necessary to express the above qualitative properties in numerical form. This we have done by employing Hammett *sigma* constants or pK_a values to account for variation in electronic properties of a set of congeners, Taft's E_s constants to describe steric effects, and log P values to account for differences in hydrophobicity, where P is the octanol/water partition coefficient. In the present report our concern is with the relative lipophilic character of organic compounds as defined by log P.

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Extending the line of thinking started by Meyer and Overton, we have found that the biological action of many sets of compounds can be correlated with two types of equations:

$$\text{RBR} \equiv \log \frac{1}{C} = a \log P + b \quad (1)$$

$$\text{RBR} = \log \frac{1}{C} \equiv -d(\log P)^2 + e \log P + f \quad (2)$$

In the above equations, C is the molar concentration of drug causing a standard biological response, RBR = relative biological response, and the constants a, b, d, e, and f are characteristic of a given system and are evaluated by the method of least squares.

While Meyer and Overton and their successors looked for a linear relation between RBR and P, there is now abundant experi-

mental evidence as well as theoretical support (1) for a parabolic relationship between log $1/C$ and log P. One of the advantages of casting experimental results in the form of Eq. 1 or 2 is of course the fact that large amounts of data can be stored very compactly. Most important is the fact that rapid and more or less quantitative comparisons can be made by simple comparison of parameters.

For this report we have searched the literature for examples in which *sets* of organic compounds have been used to hemolyze red cells and in which the data have been expressed in quantitative terms. From the experimental data of Table 1, the equations in Tables 2 and 3 have been derived.

METHOD

The biological data (7–23) and physicochemical constants are given in Table 1. Partition coefficients were not measured for every molecule; those marked with an asterisk were calculated by means of additivity principles (28).

It has been found that each CH_2 in a homologous series increases log P by about 0.5. Thus log P for butanol is calculated from log P for methanol: $\log P_{\text{methanol}} + 3(CH_2) = -0.66 + 1.50 = 0.84$. A branched chain near a functional group usually decreases log P by 0.2. Hence $\log P_{\text{2-propanol}} = \log P_{\text{propanol}} - 0.2 = 0.14$. The calculated values for the alcohols in Table 1*A*, *C*, *D*, *G*, and *BB* were made in this fashion.

The carbamates of Table 1*B* were similarly based on log P for ethyl carbamate, while log P for the amides was based on butyramide. The values for the esters in Table 1*C* were based on propyl formate and ethyl acetate.

In Table 1*E* and *F*, values for the substituents are from the benzene system (28). Log P for naphthalene and benzene was used as the basis for these sets. For example,

$$\text{Log } P_{\text{1,2,4-trichlorobenzene}} = \log P_{\text{o-dichlorobenzene}} + \pi_{Cl} = 3.38 + 0.71 = 4.09$$

$$\pi_X = \log P_X - \log P_H$$

Thus π_{Cl} as used above is calculated as $\pi_{Cl} = \log P_{C_6H_5Cl} - \log P_{C_6H_6} = 2.84 - 2.13 = 0.71$. The data in Table 1*E* and *F* are not directly comparable to those in the other parts of Table 1. In Table 1*E*, saponin was used as the lysin and the compounds were tested for their ability to accelerate hemolysis. Table 1*F*, taurocholate was used as the lysin.

In Table 1*H*, the calculated partition coefficients were based on the experimental value of -0.17 found for monobutyrin. The heptyl derivative was calculated as follows: $-0.17 + 2.00 = 1.83$.

In Table 1*I* and *J*, log P for *p*-chlorocresol was calculated by adding 0.50 to *p*-chlorophenol. Log P for *p*-chlorophenylethyl alcohol was calculated by adding π_{Cl} from the benzyl alcohol system (28) to log P of phenylethyl alcohol: $0.86 + 1.36 = 2.22$.

All the data in Table 1*K* are from the work of Rogers (14). The log P values for ionic compounds such as the *N*-alkylpyridinium halides of Table 1*L* present a special problem. In the aqueous phase these molecules are completely ionized, while in the organic phase they must be in the form of the ion pairs. Since the partition coefficient, in the proper sense of the word, refers to the partitioning of a single species, we cannot call these "true" partition coefficients. However, the apparent partition coefficients of (presumably) the ion pair do constitute a useful reference scale of relative lipophilicity. The values of Table 1*L* are based on the experimental value of -0.95 for octylpyridinium bromide.

In Table 1*M*, the calculated partition coefficients were based on the experimental value of 1.77 found for *N*-decylpiperidine hydrochloride. The partition coefficient for this compound was measured in 0.01 M HCl. Log P values of amine hydrochlorides were found to vary linearly with the hydrochloric acid concentration; however, values extrapolated to infinite dilution were essentially those obtained with 0.01 M HCl. The analytical technique used was that of Mukerjee and Mukerjee (29). The assumption is that it is primarily the protonated form of these derivatives which is responsible for hemolysis.

In Table 1*N*, the molecule for the base is the decyl derivative. In Table 1*O*, *P*, and *Q*,

TABLE 1

Isohemolytic concentrations and partition coefficients used in derivation of equations in Table 2

Asterisks indicate calculated values.

A. Compound	Log *P*	Observed log $1/C^a$	Calculated log $1/C$ from Eq. 8	$\lvert \Delta \log 1/C \rvert$
Methyl alcohol	−0.66	−0.46	−0.55	0.09
Ethyl alcohol	−0.16*	−0.23	−0.13	0.10
Propyl alcohol	0.34	0.21	0.29	0.08
Isopropyl alcohol	0.14*	0.21	0.12	0.09
Butyl alcohol	0.84*	0.60	0.71	0.11
Isobutyl alcohol	0.64*	0.60	0.54	0.06
Pentyl alcohol	1.34*	1.27	1.13	0.14
tert-Pentyl alcohol	0.89	0.68	0.75	0.07

B. Compound	Log *P*	Observed log $1/C^b$	Calculated log $1/C$ from Eq. 9	$\lvert \Delta \log 1/C \rvert$
Methyl carbamate	−0.65*	−0.39	−0.36	0.03
Ethyl carbamate	−0.15	−0.02	−0.05	0.03
Propyl carbamate	0.35*	0.45	0.26	0.19
Formamide	−1.71*	−0.89	−1.02	0.13
Acetamide	−1.21*	−0.68	−0.71	0.03
Propionamide	−0.71*	−0.62	−0.40	0.22
Butyramide	−0.21	−0.21	−0.08	0.13

C. Compound	Log *P*	Observed log $1/C^b$	Calculated log $1/C$ from Eq. 7	$\lvert \Delta \log 1/C \rvert$
Methyl alcohol	−0.66	−0.87	−0.83	0.04
Ethyl alcohol	−0.16*	−0.51	−0.38	0.13
Propyl alcohol	0.34	−0.03	0.07	0.10
Butyl alcohol	0.84*	0.50	0.52	0.02
Pentyl alcohol	1.34*	1.04	0.97	0.07
Heptyl alcohol	2.34*	1.92	1.87	0.05
Octyl alcohol	2.84*	2.40	2.32	0.08
Methyl formate	−0.17*	−0.17	−0.39	0.22
Ethyl formate	0.33*	0.15	0.06	0.09
Propyl formate	0.83	0.64	0.51	0.13
Methyl acetate	0.18	−0.06	−0.08	0.02
Ethyl acetate	0.73	0.33	0.42	0.09
Propyl acetate	1.23*	0.80	0.87	0.07
Butyl acetate	1.73*	1.33	1.32	0.01
Methyl propionate	0.71*	0.47	0.40	0.07
Ethyl propionate	1.21	0.77	0.85	0.08
Propyl propionate	1.71*	1.22	1.30	0.08
Methyl butyrate	1.23*	0.85	0.87	0.02
Ethyl butyrate	1.73*	1.22	1.32	0.10

D. Compound	Log *P*	Observed log $1/C^c$	Calculated log $1/C$ from Eq. 6	$\lvert \Delta \log 1/C \rvert$
Methyl alcohol	−0.66	−0.87	−0.93	0.06
Ethyl alcohol	−0.16*	−0.51	−0.45	0.06
Propyl alcohol	0.34	−0.03	0.02	0.05
Butyl alcohol	0.84*	0.50	0.50	0.00
Pentyl alcohol	1.34*	1.04	0.98	0.06
Heptyl alcohol	2.34*	1.92	1.93	0.01

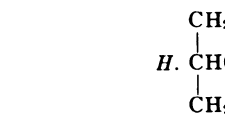
TABLE 1—*Continued*

E. Compound	Log *P*	Observed log 1 − *R*/*C*[d]	Calculated log 1 − *R*/*C* from Eq. 20	|Δ log 1 − *R*/*C*|
Benzene	2.13	−1.60	−1.61	0.01
Chlorobenzene	2.84	−0.74	−0.71	0.04
Bromobenzene	2.99	−0.70	−0.54	0.16
Iodobenzene	3.25	−0.21	−0.28	0.07
1,2-Dichlorobenzene	3.38	−0.11	−0.16	0.05
1,3-Dichlorobenzene	3.38	−0.16	−0.16	0.00
1,4-Dichlorobenzene	3.39	−0.19	−0.15	0.04
1,4-Dibromobenzene	3.85*	0.34	0.22	0.12
1,2,4-Trichlorobenzene	4.01*	0.18	0.38	0.20
Naphthalene	3.37	0.08	0.17	0.25
α-Chloronaphthalene	4.08*	0.23	0.37	0.14
β-Chloronaphthalene	4.08*	0.30	0.37	0.07
α-Bromonaphthalene	4.23*	0.40	0.46	0.06
β-Bromonaphthalene	4.23*	0.48	0.46	0.02
1,4-Diiodobenzene	4.37*	0.59	0.53	0.06
α-Iodonaphthalene	4.49*	0.63	0.59	0.05
β-Iodonaphthalene	4.49*	0.67	0.59	0.09

F. Compound	Log *P*	Observed log 1 − *R*/*C*[d]	Calculated log 1 − *R*/*C* from Eq. 4	|Δ log 1 − *R*/*C*|
Benzene	2.13	−1.58	−1.45	0.13
Chlorobenzene	2.84	−0.74	−0.86	0.12
Bromobenzene	2.99	−0.70	−0.73	0.03
Iodobenzene	3.25	−0.55	−0.51	0.04
1,2-Dichlorobenzene	3.38	−0.22	−0.41	0.19
1,3-Dichlorobenzene	3.38	−0.52	−0.41	0.11
1,4-Dichlorobenzene	3.39	−0.60	−0.40	0.20
1,4-Dibromobenzene	3.85*	−0.10	−0.01	0.09
1,2,4-Trichlorobenzene	4.01*	−0.16	0.19	0.35
Naphthalene	3.37	0.00	−0.41	0.41
α-Chloronaphthalene	4.08*	0.11	0.18	0.07
β-Chloronaphthalene	4.08*	0.20	0.18	0.02
α-Bromonaphthalene	4.23*	0.32	0.31	0.01
β-Bromonaphthalene	4.23*	0.40	0.31	0.09
1,4-Diiodobenzene	4.37*	0.50	0.42	0.08
α-Iodonaphthalene	4.49*	0.50	0.52	0.02
β-Iodonaphthalene	4.49*	0.58	0.52	0.06

G. Compound	Log *P*	Observed log 1 − *R*/*C*[e]	Calculated log 1 − *R*/*C* from Eq. 3	|Δ log 1 − *R*/*C*|
Butyl alcohol	0.84*	−2.85	−2.78	0.07
Pentyl alcohol	1.34*	−2.21	−2.25	0.04
Hexyl alcohol	1.84*	−1.60	−1.72	0.12
Heptyl alcohol	2.34*	−1.12	−1.19	0.07
Octyl alcohol	2.84*	−0.94	−0.66	0.28
Nonyl alcohol	3.34*	−0.01	−0.13	0.12

TABLE 1—*Continued*

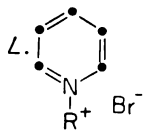
H. 	Log P^f	Observed log $1/C^g$	Calculated log $1/C$ from Eq. 26	$\lvert \Delta \log 1/C \rvert$
R = heptyl	1.83*	2.95	2.97	0.02
R = octyl	2.33*	3.40	3.44	0.04
R = nonyl	2.83*	3.70	3.72	0.02
R = decyl	3.33*	3.95	3.82	0.13
R = undecyl	3.83*	3.82	3.74	0.08
R = tridecyl	4.83*	2.80	3.03	0.23
R = pentadecyl	5.83*	1.70	1.60	0.10

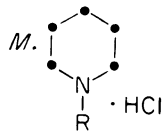
I. Compound	Log *P*	Observed log $1/C^h$	Calculated log $1/C$ from Eq. 10	$\lvert \Delta \log 1/C \rvert$
Phenol	1.46	1.24	1.22	0.02
Cresols (*o*, *m*, *p*)	1.95[i]	1.62	1.61	0.01
m-Cresol	1.96	1.54	1.61	0.07
p-Chlorophenol	2.39	1.93	1.95	0.02
p-Chloro-*m*-cresol	2.89*	2.37	2.36	0.01
Benzyl alcohol	1.10	0.93	0.94	0.01
Phenethyl alcohol	1.36	1.17	1.14	0.03
p-Chlorophenethyl alcohol	2.22*	1.85	1.82	0.03

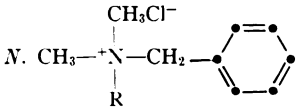
J. Compound	Log *P*	Observed log $1/C^h$	Calculated log $1/C$ from Eq. 11	$\lvert \Delta \log 1/C \rvert$
Phenol	1.46	1.29	1.32	0.03
Cresols (*o*, *m*, *p*)	1.95[i]	1.66	1.70	0.04
m-Cresol	1.96	1.66	1.70	0.04
p-Chlorophenol	2.39	2.01	2.03	0.02
p-Chloro-*m*-cresol	2.89*	2.41	2.44	0.03
Benzyl alcohol	1.10	1.04	1.05	0.01
Phenethyl alcohol	1.36	1.28	1.25	0.03
p-Chlorophenethyl alcohol	2.22*	2.04	1.90	0.14

K. Compound	Log P^j	Observed log $1/C^k$	Calculated log $1/C$ from Eq. 5	$\lvert \Delta \log 1/C \rvert$
Diphenylamine	3.22	3.06	3.06	0.00
Thianaphthene	3.09	3.06	2.92	0.14
Naphthalene	3.01	3.00	2.84	0.16
5-Bromoindole	3.00	2.84	2.82	0.02
1,2-Dimethylindole	2.82	2.69	2.63	0.06
Indene	2.80	2.69	2.61	0.08
5-Methylindole	2.68	2.17	2.48	0.31
3-Methylindole	2.60	2.17	2.39	0.22
2-Methylindole	2.56	2.17	2.35	0.18
5-Cyanoindole	2.37	2.19	2.14	0.05
Indole	2.25	1.97	2.01	0.04
Toluene	2.11	1.93	1.86	0.07
5-Methoxyindole	2.10	1.93	1.85	0.08
Quinoline	2.06	1.74	1.81	0.07
Anisole	2.04	1.69	1.78	0.09
Benzothiazole	2.03	1.91	1.77	0.14
Indizole	1.82	1.63	1.54	0.09
Benzoxazole	1.59	1.39	1.30	0.09
Benzene	1.56	1.19	1.26	0.07

TABLE 1—*Continued*

L. 	Log P^l	Observed log $1/C^m$	Calculated log $1/C$ from Eq. 21	$\lvert\Delta \log 1/C$
R = nonyl	−0.45*	2.18	2.00	0.18
R = decyl	0.05*	2.55	2.65	0.10
R = undecyl	0.55*	3.02	3.22	0.20
R = dodecyl	1.05*	3.63	3.69	0.06
R = tridecyl	1.55*	4.16	4.08	0.08
R = tetradecyl	2.05*	4.49	4.39	0.10
R = hexadecyl	3.05*	4.82	4.74	0.08
R = heptadecyl	3.55*	4.77	4.78	0.01
R = octadecyl	4.05*	4.67	4.74	0.07

M. 	Log P^n	Observed log $1/C^m$	Calculated log $1/C$ from Eq. 23	$\lvert\Delta \log 1/C\rvert$
R = heptyl	0.27*	1.61	1.41	0.20
R = octyl	0.77*	1.97	2.07	0.10
R = nonyl	1.27*	2.55	2.65	0.10
R = decyl	1.77	3.02	3.14	0.12
R = undecyl	2.27*	3.40	3.54	0.14
R = dodecyl	2.77*	3.92	3.85	0.07
R = tridecyl	3.27*	4.18	4.07	0.11
R = tetradecyl	3.77*	4.38	4.20	0.18
R = pentadecyl	4.27*	4.28	4.24	0.04
R = hexadecyl	4.77*	4.12	4.19	0.07
R = heptadecyl	5.27*	3.97	4.05	0.08

N. 	Log P	Observed log $1/C^o$	Calculated log $1/C$ from Eq. 24	$\lvert\Delta \log 1/C\rvert$
R = octyl	−1.08*	1.76	1.76	0.00
R = decyl	−0.08[p]	2.95	2.94	0.01
R = dodecyl	0.92*	3.82	3.82	0.00
R = tetradecyl	1.92*	4.40	4.43	0.03
R = hexadecyl	2.92*	4.79	4.67	0.12
R = octadecyl	3.92*	4.52	4.58	0.06

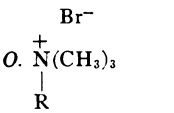
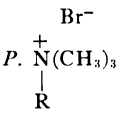
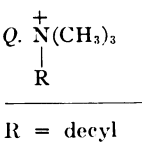
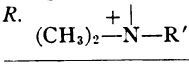
O. 	Log P	Observed log $1/C^q$	Calculated log $1/C$ from Eq. 22	$\lvert\Delta \log 1/C\rvert$
R = octyl	−1.07[r]	1.08	1.17	0.09
R = decyl	−0.16[r]	2.72	2.56	0.16
R = dodecyl	0.84*	3.78	3.75	0.03
R = tetradecyl	1.84*	4.40	4.58	0.18
R = hexadecyl	2.84*	5.11	5.04	0.07
R = octadecyl	3.84*	5.15	5.14	0.01

TABLE 1—*Continued*

P. 	Log *P*	Observed log $1/C^q$	Calculated log $1/C$ from Eq. 17	$\lvert \Delta \log 1/C \rvert$
R = decyl	−0.16[r]	2.68	2.76	0.08
R = dodecyl	0.84*	3.81	3.92	0.11
R = tetradecyl	1.84*	5.55	5.10	0.45
R = hexadecyl	2.84*	6.00	6.26	0.26

Q. 	Log *P*	Observed log $1/C^q$	Calculated log $1/C$ from Eq. 16	$\lvert \Delta \log 1/C \rvert$
R = decyl	−0.16[r]	2.73	2.82	0.09
R = dodecyl	0.84*	3.73	3.59	0.14
R = tetradecyl	1.84*	4.34	4.36	0.02
R = hexadecyl	2.84*	5.10	5.13	0.03

R. 	Log *P*	Observed log $1/C^s$	Calculated log $1/C$ from Eq. 25	$\lvert \Delta \log 1/C \rvert$
R = decyl, R′ = benzyl	−0.08[p]	3.11	3.15	0.04
R = decyl, R′ = 2-chlorobenzyl	0.63*	3.47	3.60	0.13
R = decyl, R′ = 4-chlorobenzyl	0.63*	3.70	3.60	0.10
R = decyl, R′ = 2,4-dichlorobenzyl	1.34*	3.85	3.97	0.12
R = dodecyl, R′ = benzyl	0.92*	4.04	3.90	0.14
R = dodecyl, R′ = 2,4-dichlorobenzyl	2.34*	4.43	4.43	0.00
R = tetradecyl, R′ = benzyl	1.92*	4.52	4.31	0.21
R = tetradecyl, R′ = 2,4-dichlorobenzyl	3.34*	4.39	4.62	0.23
R = hexadecyl, R′ = benzyl	2.92*	4.54	4.56	0.02
R = hexadecyl, R′ = 2-chlorobenzyl	3.63*	4.66	4.64	0.02
R = octadecyl, R′ = benzyl	3.92*	4.62	4.66	0.04
R = octadecyl, R′ = 2-chlorobenzyl	4.63*	4.74	4.63	0.11

S. R—NH_2·HCl	Log P^t	Observed log $1/C^u$	Calculated log $1/C$ from Eq. 13	$\lvert \Delta \log 1/C \rvert$
R = butyl	−2.15*	−0.12	−0.42	0.30
R = hexyl	−1.15*	0.32	0.53	0.21
R = octyl	−0.15*	1.12	1.48	0.36
R = decyl	0.85*	2.57	2.44	0.13
R = dodecyl	1.85	3.52	3.39	0.13

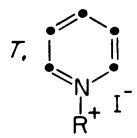
T. 	Log P^l	Observed log $1/C^u$	Calculated log $1/C$ from Eq. 15	$\lvert \Delta \log 1/C \rvert$
R = butyl	−2.95*	0.03	−0.24	0.27
R = hexyl	−1.95*	0.33	0.64	0.31
R = octyl	−0.95	1.42	1.51	0.09
R = decyl	0.05*	2.40	2.39	0.01
R = dodecyl	1.05*	3.39	3.27	0.12

TABLE 1—*Continued*

U. $ROSO_3Na$	Log P	Observed log $1/C^o$	Calculated log $1/C$ from Eq. 27	$\lvert\Delta \log 1/C\rvert$
R = ocytl	−0.40*	1.79	1.80	0.01
R = decyl	0.60*	2.95	3.00	0.05
R = dodecyl	1.60	4.00	3.87	0.13
R = tetradecyl	2.60*	4.44	4.41	0.03
R = hexadecyl	3.60*	4.44	4.62	0.18
R = octadecyl	4.60*	4.60	4.51	0.09

V. $ROSO_3Na$	Log P	Observed log $1/C^o$	Calculated log $1/C$ from Eq. 14	$\lvert\Delta \log 1/C\rvert$
R = octyl	−0.40*	1.44	1.48	0.04
R = decyl	0.60*	2.54	2.56	0.02
R = dodecyl	1.60	3.79	3.63	0.16
R = tetradecyl	2.60*	4.60	4.70	0.10

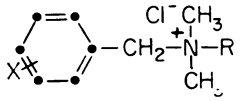
W. RCOOH	Log P	Observed log $1/C^w$	Calculated log $1/C$ from Eq. 12	$\lvert\Delta \log 1/C\rvert$
R = butanoic	0.88	1.41	1.26	0.15
R = 2-methylpropanoic	0.68*	1.40	1.00	0.40
R = 3-methylbutanoic	1.18*	1.69	1.64	0.05
R = pentanoic	1.38*	1.79	1.89	0.10
R = 4-methylpentanoic	1.68*	2.00	2.26	0.26
R = hexanoic	1.88	2.04	2.52	0.48
R = heptanoic	2.38*	2.66	3.14	0.48
R = octanoic	2.88*	4.31	3.77	0.54
R = nonanoic	3.38*	4.58	4.49	0.18

X. $R{-}\underset{\displaystyle Br}{\underset{\vert}{C}}H{-}COO^-K^+$	Log P^x	Observed log $1/C^y$	Calculated log $1/C$ from Eq. 19	$\lvert\Delta \log 1/C\rvert$
R = hexyl	−0.68*	2.20	1.92	0.28
R = octyl	0.32*	2.50	2.85	0.35
R = decyl	1.32*	3.41	3.51	0.10
R = dodecyl	2.32*	3.71	3.89	0.18
R = tetradecyl	3.32*	4.32	4.00	0.32
R = hexadecyl	4.32*	4.32	3.83	0.49
R = octadecyl	5.32*	2.80	3.39	0.59
R = eicosyl	6.32*	2.80	2.67	0.13

Y. RCOOH	Log P^z	Observed log $1/C^{aa}$	Calculated log $1/C$ from Eq. 18	$\lvert\Delta \log 1/C\rvert$
R = octanoic	−1.20*	1.46	1.26	0.20
R = nonanoic	−0.70*	1.72	1.88	0.16
R = decanoic	−0.20*	2.36	2.41	0.05
R = undecanoic	0.30*	2.67	2.85	0.18
R = dodecanoic	0.80*	3.22	3.20	0.02
R = tetradecanoic	1.80*	3.93	3.61	0.32
R = hexadecanoic	2.80*	3.51	3.64	0.13
R = octadecanoic	3.80*	3.30	3.31	0.01

TABLE 1—*Continued*

Z. Inhibition of *S. aureus* by quaternary compounds



	Log *P*	Observed log 1/C[bb]	Calculated log 1/C from Eq. 28	| Δ log 1/C |
X = H, R = $C_{10}H_{21}$	−0.08[p]	2.79	2.88	0.09
X = H, R = $C_{12}H_{25}$	0.92*	3.74	3.62	0.12
X = H, R = $C_{14}H_{29}$	1.92*	4.17	4.02	0.15
X = H, R = $C_{16}H_{33}$	2.92*	3.92	4.06	0.14
X = H, R = $C_{18}H_{37}$	3.92*	3.34	3.76	0.42
X = 2-Cl, R = C_8H_{17}	−0.49*	2.02	2.48	0.46
X = 2-Cl, R = $C_{10}H_{21}$	0.51*	3.41	3.36	0.05
X = 2-Cl, R = $C_{12}H_{25}$	1.51*	4.14	3.90	0.24
X = 2-Cl, R = $C_{14}H_{29}$	2.51*	4.14	4.08	0.06
X = 2-Cl, R = $C_{16}H_{33}$	3.51*	3.74	3.93	0.19
X = 2-Cl, R = $C_{18}H_{37}$	4.51*	3.43	3.42	0.01
X = 4-Cl, R = C_8H_{17}	−0.38*	2.60	2.60	0.00
X = 4-Cl, R = $C_{10}H_{21}$	0.62*	3.60	3.44	0.16
X = 4-Cl, R = $C_{12}H_{25}$	1.62*	4.04	3.93	0.11
X = 4-Cl, R = $C_{14}H_{29}$	2.62*	4.34	4.08	0.26
X = 4-Cl, R = $C_{16}H_{33}$	3.62*	3.85	3.89	0.04
X = 4-Cl, R = $C_{18}H_{37}$	4.62*	3.17	3.35	0.18
X = 4-NO_2, R = C_8H_{17}	−0.84*	2.11	2.09	0.02
X = 4-NO_2, R = $C_{10}H_{21}$	0.16*	3.11	3.09	0.02
X = 4-NO_2, R = $C_{12}H_{25}$	1.16*	4.20	3.75	0.45
X = 4-NO_2, R = $C_{14}H_{29}$	2.16*	4.23	4.06	0.17
X = 4-NO_2, R = $C_{16}H_{33}$	3.16*	4.00	4.02	0.02
X = 4-NO_2, R = $C_{18}H_{37}$	4.16*	3.46	3.64	0.18
X = 2,4-Cl_2, R = C_8H_{17}	0.21*	2.63	3.13	0.50
X = 2,4-Cl_2, R = $C_{10}H_{21}$	1.21*	3.65	3.77	0.12
X = 2,4-Cl_2, R = $C_{12}H_{25}$	2.21*	4.25	4.06	0.19
X = 2,4-Cl_2, R = $C_{14}H_{29}$	3.21*	4.28	4.01	0.27
X = 2,4-Cl_2, R = $C_{16}H_{33}$	4.21*	3.41	3.61	0.20
X = 2,4-Cl_2, R = $C_{18}H_{37}$	5.21*	3.30	2.86	0.44
X = 3,4-dioxymethylene, R = C_8H_{17}	−1.13*	2.04	1.74	0.30
X = 3,4-dioxymethylene, R = $C_{10}H_{21}$	−0.13*	3.08	2.84	0.24
X = 3,4-dioxymethylene, R = $C_{12}H_{25}$	0.87*	4.00	3.59	0.41
X = 3,4-dioxymethylene, R = $C_{14}H_{29}$	1.87*	4.23	4.00	0.23
X = 3,4-dioxymethylene, R = $C_{16}H_{33}$	2.87*	4.20	4.07	0.13
X = 3,4-dioxymethylene, R = $C_{18}H_{37}$	3.87*	3.23	3.78	0.55
X = 3,4-dimethoxy, R = $C_{12}H_{25}$	1.00*	3.71	3.67	0.04
X = 3,4-dimethoxy, R = $C_{14}H_{29}$	2.00*	4.11	4.03	0.08
X = 3,4-dimethoxy, R = $C_{16}H_{33}$	3.00*	3.41	4.05	0.64
X = 3,4-Cl_2, R = C_8H_{17}	0.38*	2.92	3.27	0.35
X = 3,4-Cl_2, R = $C_{10}H_{21}$	1.38*	3.79	3.85	0.06
X = 3,4-Cl_2, R = $C_{12}H_{25}$	2.38*	4.36	4.08	0.28
X = 3,4-Cl_2, R = $C_{14}H_{29}$	3.38*	4.04	3.97	0.07
X = 3,4-Cl_2, R = $C_{16}H_{33}$	4.38*	3.39	3.51	0.12
X = 3,4-Cl_2, R = $C_{18}H_{37}$	5.38*	3.11	2.70	0.41
X = 2-OH-5-NO_2, R = $C_{12}H_{25}$	0.49*	2.70	3.35	0.65
Omitted from correlation				
X = 3,4-dimethoxy, R = $C_{10}H_{21}$	0.00*	1.88	2.96	1.08
X = 2-OH-5-NO_2, R = $C_{14}H_{29}$	1.49*	2.63	3.89	1.26

TABLE 1—*Continued*

AA. Inhibition of *S. aureus* by sodium alkyl sulfates

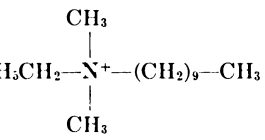
$ROSO_3^-Na^+$	Log *P*	Observed log $1/C^{cc}$	Calculated log $1/C$ from Eq. 29	$\lvert \Delta \log 1/C \rvert$
R = butyl	−2.40*	0.08	0.02	0.06
R = pentyl	−1.90*	0.36	0.37	0.01
R = hexyl	−1.40*	0.51	0.72	0.21
R = heptyl	−0.90*	0.94	1.06	0.12
R = octyl	−0.40*	1.27	1.41	0.14
R = nonyl	0.10*	1.79	1.76	0.03
R = decyl	0.60*	2.25	2.11	0.14
R = dodecyl	1.60	3.27	2.80	0.47
R = tetradecyl	2.60*	3.88	3.49	0.39
R = hexadecyl	3.60*	3.58	4.19	0.61

BB. Inhibition of resting potential of lobster axon

Compound	Log *P*	Observed log $1/C^{dd}$	Calculated log $1/C$ from Eq. 30	$\lvert \Delta \log 1/C \rvert$
Methanol	−0.66	−0.60	−0.67	0.07
Ethanol	−0.16*	−0.26	−0.24	0.02
Propanol	0.34	0.09	0.19	0.10
Butanol	0.84*	0.62	0.63	0.01
Pentanol	1.34*	1.12	1.06	0.06

[a] From Wirgin (7).
[b] From Führer and Neubauer (8).
[c] From Vernon (9).
[d] From Ponder (10).
[e] From Ponder and Hyman (11).
[f] Monobutyrin measured, log $P = -0.17$; W. R. Glave, unpublished observations.
[g] From Breusch and Hersek (12).
[h] From Ansel and Cadwallader (13).
[i] Average value for *meta* and *para* isomers.
[j] Log *P* values taken from Rogers (14).
[k] From Rogers (14).
[l] Octylpyridinium bromide measured, log $P = -0.95$; D. Soderberg, unpublished observations.
[m] From Breusch and Hersek (15).
[n] *N*-Decylpiperidine hydrochloride measured, log $P = 1.77$; W. P. Glave, unpublished observations.
[o] From Ross and Silverstein (16).
[p] D. Soderberg, unpublished observations.
[q] From Hooghwinkel, DeRooij, and Dankmeijer (17).
[r] From Eldefrawi and O'Brien (24).
[s] From Cadwallader and Ansel (18).
[t] Dodecylamine hydrochloride measured, log $P = 1.85$; W. P. Glave, unpublished observations.
[u] From Kondo and Tomizawa (19).
[v] From Rideal and Taylor (20).
[w] From Bodansky (21).
[x] α-Bromopropanoic acid log *P* for carboxylate anion = − 3.18; for each additional CH_2 unit, add 0.50.
[y] From Eggerth (22).
[z] Hexanoic acid log *P* for carboxylate anion = −2.20; for each additional CH_2 unit, add 0.50.
[aa] From Breusch and Bodur (23).
[bb] From Ross, Kwartler, and Bailey (25).
[cc] From Cowles (26).
[dd] From Houck (27).

the experimental values of the octyl and decyl derivatives were available from the work of Eldefrawi and O'Brien (24). In Table 1*R*, π_{Cl} from the benzene system (28) was added to the log *P* calculated from the basic structure:



In Table 1*S*, log *P* values are based on the experimental value of 1.85 found for dodecylamine hydrochloride in 0.01 M HCl. The technique used was the same as for the compounds of Table 1*M*.

In Table 1*T*, it is assumed that the iodide does not have a significantly different log *P* from the corresponding bromide. This is based on two experimental facts. The difference of log *P* for hexadecylpyridinium bromide is 0.10. It is assumed that the difference between bromide and iodide would not be much greater. The difference between π_{Br} and π_I in RX is 0.4. A difference of this size is not serious for our present purpose.

The values in Table 1*U*, *V*, and *AA* are based on the experimental value for $CH_3(CH_2)_{11}OSO_3^-Na^+$ (1.60); as with the pyridinium compounds, we are assuming that it is the ion pair which is partitioned. In determining log *P* for this compound we have employed the analytical technique of Uno *et al.* (30).

In Table 1*W* and *Y*, the calculated log *P* values refer to the un-ionized and ionized forms of the acid, respectively, and are based on butanoic and hexanoic acids. Log *P* values for Table 1*X* were calculated as follows: log $P_{CH_3(CH_2)_4CO_2^-}$ − log $P_{CH_3(CH_2)_4CO_2H}$ = −2.20 − (1.90) = −4.10, the difference between ionized and un-ionized hexanoic acid. It is assumed that −4.10 would be a constant for all simple aliphatic acids. Log $P_{\alpha\text{-bromopropanoic acid un-ionized}}$ = 0.92, while the ionized log *P* = −4.10 + 0.92 = −3.18. Where R = hexyl, log *P* ionized = 2.50 − 3.18 = −0.68.

Table 1*Z* contains a series of substituted benzyldimethylalkylammonium chlorides used for killing *Staphylococcus aureus*. The log *P* values of the ring-substituted molecules were calculated by adding the π values for the substituents derived from the phenoxyacetic acid system and the phenylacetic acid system (28) to the parent benzyldimethylalkylammonium chloride. The π value for 3,4-dioxymethylene was calculated as follows:

$$\pi_{CH_2O_2} = \log P_{\text{3,4-dioxymethylene benzyl alcohol}} - \log P_{\text{benzyl alcohol}}$$
$$= 1.05 - 1.10 = -0.05$$

RESULTS AND DISCUSSION

Turning first to the slopes of the linear equations of Table 2, we find a surprisingly constant value of mean and standard deviation of 0.93 ± 0.17 for 15 different examples. The constancy of slope is most interesting in view of the fact that such a variety of different red cells and organic compounds have been employed and that the studies were carried out in many different laboratories utilizing different definitions of hemolysis as well as different experimental conditions with respect to buffers, temperature control, and precision of analysis. Thus a given increment in hydrophobicity, as defined by log *P* of the octanol/water reference system, results in a corresponding constant increase in hemolytic ability of the congener as defined by log 1/*C*. Whether the molecule is neutral like the alcohols or ionic like the pyridinium or sulfate ions has little influence on the slope. In this sense, hemolysis is a very nonspecific process dependent simply on the amount of apolar material partitioning into the red cell membrane. Even Eqs. 3 and 4 conform to the pattern. In this work saponin and taurocholate, respectively, were used to cause hemolysis. The effect of the organic molecules is that of expediting the process. It is of interest to compare these slopes with other processes in which hydrophobic interactions appear to be the primary agents. Table 4 offers such comparisons. The process of hemolysis most closely resembles that of narcosis. Considering the standard deviation on the two sets of slopes, one can say, as a first approximation from the point of view of hydrophobic effects, that the two processes appear to be identical. There is a slightly greater spread in the average slopes for

TABLE 2

Linear relationships between hemolysis and log P

Type of compound	Erythrocyte source	Log $1/C = a \log P + b$					Location of data in Table 1	Temperature and pH	Equation
		a	b	n[a]	r[b]	s[c]			
Alcohols[d]	Rabbit	1.060(±0.23)	−3.670(±0.50)[e]	6	0.988	0.171	1*G*	25°, —	3
Benzene derivatives[f]	Rabbit	0.836(±0.14)	−3.230(±0.52)	17	0.958	0.172	1*F*	25°, 7.2	4
Aromatic	Rabbit	1.085(±0.13)	−0.430(±0.32)	19	0.974	0.133	1*K*	38°, —	5
Alcohols	Rabbit	0.955(±0.07)	−0.302(±0.08)	6	0.999	0.059	1*D*	18°, —	6
Alcohols and esters	Bovine	0.901(±0.05)	−0.238(±0.07)	19	0.993	0.096	1*C*	18–24°, —	7
Alcohols	Rabbit	0.841(±0.16)	0.006(±0.12)	8	0.983	0.110	1*A*	37°, —	8
Amides and carbamates	Bovine	0.625(±0.23)	0.046(±0.21)	7	0.951	0.155	1*B*	18–24°, —	9
Alcohols and phenols	Human	0.778(±0.06)	0.087(±0.11)	8	0.997	0.037	1*I*	37°, —	10
Alcohols and phenols	Rabbit	0.765(±0.10)	0.206(±0.20)	8	0.991	0.065	1*J*	37°, —	11
RCOOH	Dog	1.258(±0.35)	0.151(±0.71)	9	0.954	0.386	1*W*	25°, 3.1–3.8	12
$RNH_2 \cdot HCl$	Dog	0.953(±0.32)	1.625(±0.46)	5	0.984	0.318	1*S*	30°, 7.1	13
$ROSO_3^-Na^+$	Human	1.073(±0.27)	1.912(±0.42)	4	0.997	0.139	1*V*	23–25°, 7.2	14
N-Alkylpyridinium I^-	Dog	0.879(±0.25)	2.349(±0.43)	5	0.988	0.252	1*T*	30°, 7.1	15
Alkyltrimethylammonium Br^-	Bovine	0.772(±0.23)	2.941(±0.40)	4	0.995	0.120	1*Q*	37°, —	16
Alkyltrimethylammonium Br^-	Human	1.170(±0.74)	2.942(±1.3)	4	0.979	0.385	1*P*	37°, —	17

[a] Number of data points used in deriving the equation.

[b] Correlation coefficient.

[c] Standard deviation.

[d] Saponin was used to cause hemolysis. Activity is not expressed as log $1/C$ but as $\log(1 - R/C)$.

[e] Numbers in parentheses are 95% confidence intervals.

[f] Taurocholate was used to cause hemolysis. Activity is not expressed as log $1/C$ but as $\log(1 - R/C)$.

TABLE 3

Nonlinear relationships between hemolysis and log P

Type of compound	Erythrocyte source	Log $1/C = d(\log P)^2 + e \log P + f$							Location of data in Table 1	Temperature and pH	Equation
		d	e	f	$\log P_0$	n[a]	r[b]	s[c]			
RCOOH	Dove	−0.186(±0.08)	0.894(±0.25)	2.600(±0.24)	2.40(2.0–3.4)	8	0.978	0.214	1*Y*	38°, 7.5	18
RCHBrCOOK	Sheep	−0.137(±0.09)	0.881(±0.52)	2.583(±0.67)	3.21(2.56–4.38)	8	0.891	0.436	1*X*	37°, 7.5	19
Benzene derivatives	Rabbit	−0.210(±0.13)	2.324(±0.91)	−5.608(±1.54)	5.52(4.7–8.8)	17	0.983	0.118	1*E*	25°, 7.2	20
N-Alkylpyridinium bromides	Dove	−0.173(±0.06)	1.231(±0.24)	2.591(±0.18)	3.56(3.1–4.6)	9	0.993	0.139	1*L*	—, —	21
Alkyltrimethylammonium bromides	Sheep	−0.181(±0.08)	1.309(±0.26)	2.777(±0.26)	3.63(2.9–5.6)	6	0.997	0.152	1*O*	37°, —	22
N-Alkylpiperidine chlorides	Dove	−0.179(±0.04)	1.519(±0.25)	1.010(±0.30)	4.25(3.9–4.8)	11	0.992	0.141	1*M*	—, —	23
$RN^+(CH_3)(CH_2C_6H_5)Cl^-$	Sheep	−0.152(±0.04)	1.003(±0.14)	3.021(±0.15)	3.30(2.9–4.0)	6	0.998	0.086	1*N*	37°, 7.2	24
Quaternary benzylammonium Cl^-	Rabbit	−0.090(±0.06)	0.721(±0.28)	3.219(±0.26)	4.00(3.2–7.7)	12	0.959	0.167	1*R*	37°, 7.5	25
α-Monoglycerides	Dove	−0.362(±0.10)	2.435(±0.80)	−0.268(±1.4)	3.36(3.1–3.5)	7	0.988	0.152	1*H*	—, —	26
$ROSO_3^-Na^+$	Sheep	−0.164(±0.08)	1.232(±0.34)	2.320(±0.32)	3.75(3.2–5.2)	6	0.995	0.144	1*U*	37°, 7.2	27

[a] Number of data points used in deriving the equation.

[b] Correlation coefficient.

[c] Standard deviation.

TABLE 4

Biochemical processes correlated by log P

$$\text{Log RBR} \equiv \log \frac{1}{C} = a \log P + b$$

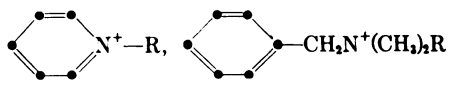
System	No. of examples	Slope: mean ± S.D.	Reference
Narcosis	15	1.04 ± 0.13	31
Hemolysis	15	0.93 ± 0.17	
Toxicity to Gram-positive bacteria	17	0.73 ± 0.17	2
Toxicity to Gram-negative bacteria	8	0.65 ± 0.25	2

toxicity with Gram-positive organisms and a still greater difference with Gram-negative organisms. The striking similarlity between the narcotic and hemolytic processes is also quite evident in the intercepts of the equations of Table 2. Omitting Eqs. 12–17, in which the hemolytic agents are ionic in character, and omitting Eqs. 3 and 4 because of the quite different definition of activity, we are left with a set of seven equations with intercept of mean and standard deviation −0.09 ± 0.23. The mean intercept for six different equations correlating (31) neutral molecules (alcohols, esters, ethers, ketones) in the narcosis of tadpoles is 0.76 ± 0.16. The difference in intercepts is 0.85; that is, for tadpoles narcosis requires a 7-fold lower concentration than does hemolysis. From the similarity of the equations it would seem that both processes involve perturbations of cell membranes by the neutral molecules. Unfortunately, most of the work reported for the toxic action of simple neutral compounds on bacteria has been in terms of phenol coefficients rather than log $1/C$. For two examples of the action of alcohols and phenols on *S. aureus*, intercepts of 0.062 and 0.480, respectively, are found (31). This gives an average value of 0.27, which falls between those for narcosis and hemolysis; that is, a 2–3-fold higher concentration is needed to inhibit the growth of *S. aureus* than to cause narcosis.

The value of the intercepts in Eqs. 1 and 2 will be determined by two factors: the sensitivity of the system and the intrinsic activity of the set of congeneric drugs under consideration. The sensitivity of the system will be determined in part by the level of standard response. One finds a lower intercept for the same set of drugs in the same system when LD_{100} is measured rather than LD_{50}. As considered above, sensitivity varies with the type of cell under consideration. However, if one studies the effects of a variety of sets of drugs on a given system, the intercept is a convenient quantitative measure of the intrinsic stereoelectronic character of the common functional group in the set of congeners. Comparing intercepts from different equations means comparison on an isolipophilic basis; that is, activities are being compared when $P = 1$ or $\log P = 0$. It is assumed under these conditions that, since hydrophobic contributions to activity are equal, differences in intercepts are due to the stereoelectronic character of the pharmacophoric function which is common to the set of congeners (setting aside, of course, differences in intercepts due to experimental error; confidence intervals on the intercepts must not be overlooked).

The intercepts of Eqs. 14–17 are particularly interesting. If we can assume that the octanol/water partition coefficient of ion pairs is comparable to that of neutral molecules, the ions are about 100–1000 times as active as neutral molecules in disrupting cell membranes. There are two examples in which anions were employed, one in Table 2 and one in Table 3 (Eqs. 14 and 27). The average for these two intercepts is 2.11. There are seven examples (Eqs. 15–17, 21, 22, 24, and 25) in which the intercepts are available for positive ions. Omitting the single case of the iodides (there is some uncertainty about their log P values; Eq. 15), for which the intercept is considerably lower, we find a mean value and standard deviation for the other six of 2.92 ± 0.21. For those interested in the quantitative description of structure-activity relationships, there are a number of satisfying aspects of the relatively small standard deviation associated with the mean value.

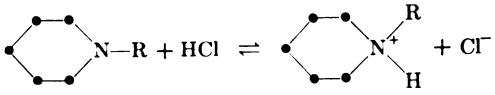
Most reassuring is the fact that structures as different as



and $RN^+(CH_3)_3$ all yield essentially the same intercept. Assuming that this result is not fortuitious, it strongly supports the use of log P as a suitable reference system for ion pairs as well as neutral molecules. The log P values for the pyridinium and benzylammonium compounds were measured in our laboratory, while the alkyltrimethylammonium values are from O'Brien's laboratory (see Table 1).

Using the above extrathermodynamic approach to structure-activity relationships, we can now classify the positively charged nitrogen atom as being roughly 10 times as effective as the negatively charged sulfate anion in the disorganization of cell membranes. It is no wonder that so many quaternary ammonium salts have been found to have a paralytic action on animal nervous systems.

After consideration of the completely unionized and the completely ionized sets of congeners, the partially ionized compounds can be discussed with more confidence. The *N*-alkylpiperidinium chlorides of Eq. 23 present an interesting example. While the following equilibrium is rather far to the right, at pH 7.4 (*N*- methylpiperidine has a pK_a of 10.1) which form of the amine is the biologically active one is open to question:



Although the neutral form would be expected from the above considerations to be intrinsically less active, and would be present in very small amounts, its partition coefficient would be serveral powers of 10 higher than the protonated form. If the protonated form is about 1000 times as active as the neutral form, and if it is present in about 1000 times the concentration, one might expect the correlation equation to resemble most closely that for completely charged ions, as indeed it does. This is true in terms of the intercept as well as log P_0 values.

The carboxylate anions of Eqs. 18 and 19 present another case in which two species are present, either or both of which might be an active agent: $COOH \rightleftharpoons COO^- H^+$. The ion would be expected to be 100 times as active, and present in very large amounts compared to the neutral molecule. Using log P values for the ion pair (RCOONa) gives an average intercept of 2.6 for Eqs. 18 and 19, near that of the positive ammonium ion. Using log P for the neutral molecules gives intercepts of -3.33 and -4.14. Moreover, log P_0 values are more in line with other sets of congeners when log P for ion pairs is used. With log P for RCOOH, log P_0 values of 7.31 and 6.48 are obtained. These are quite out of line with the other values of Tables 2 and 3.

In comparing the intercepts of Eqs. 18 and 19 with that of Eq. 12, a great difference is apparent. The work on which Eq. 12 is based was done at a low pH, so that the acids were essentially un-ionized. For these molecules we have employed log P for the neutral species of RCOOH. The intercept for Eq. 12 then fits in with the intercepts for the other equations based on neutral molecules.

An important parameter in drug design is log P_0, the ideal lipophilic character for a set of congeners acting in the same fashion on a given system. This constant is found by taking the derivative of Eq. 2 with respect to log P and setting this equal to zero to obtain the point of intersection of the tangent to the parabola relating log $1/C$ and log P.

From Table 3, omitting Eq. 20, in which hemolysis is defined in quite different terms, nine values of log P_0 can be calculated. The mean value and standard deviation for these are 3.50 ± 0.53. It is interesting to compare this value with results obtained by testing drugs *in vitro* on Gram-positive and Gram-negative cells. In that work (2) it was found that log P_0 for seven sets of Gram-negative bacteria had a mean and standard deviation of 4.37 ± 0.18. For parallel sets of Gram-positive bacteria the log P_0 was 5.87 ± 0.20. The value of log P_0 thus appears to be set by the system in which the drugs are tested. By system is meant not only the organism but also, in the case of work *in vitro*, the surrounding medium. For the three types of cells mentioned above, log P_0 appears to decrease with increasing lipid content. The lipid content of Gram-positive cell walls is quite low (about 2.5%) (32), while Gram-negative organisms have about 10 times this lipid content (25%) (33). The lipid content of the red blood cell is even higher (40–50%) (34–36).

However, as discussed below, further work on this problem is needed, since the value of log P_0 is also a function of molecular structure (37).

Although our method of comparing hemolysis, narcosis, and the inhibition of bacteria may be open to question, the parallel structure-activity relationships as defined by the extrathermodynamic approach are striking. It is our feeling that the growth inhibition of bacteria may be related to perturbations in the bacterial membranes.

Very lipophilic molecules tend to be randomly bound when they hit the extracellular lipid on the surface of the cell wall or membrane. Although this process of escaping from noncritical lipid pools into critical lipophilic sites appears to be important in determining the value of log P_0, apparently it is not the only decisive factor. Thus the average value mentioned above for log P_0 for Gram-positive cells was found primarily for neutral molecules, of which there is only one example (Table 3, Eq. 26). While this agrees well with the other log P_0 values in Table 3, it may be an exception related in some way to the glyceride structure. This is best appreciated by considering Eqs. 28 and 29.

Benzyldimethylalkylammonium chlorides acting on *S. aureus* (Table 1*Z*):

$$\text{Log } \frac{1}{C} = -0.173(\pm 0.03)(\log P)^2 + 0.884(\pm 0.14) \log P + 2.956(\pm 0.14) \quad (28)$$

$$\text{Log } P_0 = 2.55(2.4\text{–}2.7),$$

$$n = 45, \quad r = 0.898, s = 0.288$$

$ROSO_3Na$ acting on *S. aureus* (Table 1*AA*):

$$\text{Log } \frac{1}{C} = 0.694(\pm 0.13) \log P + 1.689(\pm 0.24), \quad (29)$$

$$n = 10, \quad r = 0.976, \quad s = 0.325$$

The intercepts correlating minimum inhibitory concentration in Eqs. 28 and 29 are remarkably close to the corresponding equations for hemolysis. For hemolysis by $ROSO_3Na$, the average intercept is 2.1 ± 0.2, compared with 1.7 ± 0.2. From the crude data at hand it is difficult to say how much difference there is in the intercepts. It would appear from the single Eq. 29 that bacteria are slightly more resistant than red cells. The intercept of Eq. 28 of 2.96 is quite close to the average value of 2.91 for hemolysis, showing great similarity in the two processes. The log P_0 value for Eq. 28 is lower than that found for hemolysis. Moreover, it is not out of line with many other unpublished values now in our data bank of structure-activity equations. The fact that these values of log P_0 in the range 2.5–3.0 apply to Gram-negative *and* Gram-positive cells and are much lower than log P_0 for neutral molecules acting on these systems indicates that log P_0 is a function of both the system and the type of compound. Charged molecules in particular appear to differ from neutral compounds. For the latter, a rather large variation in structure appears (6) to have little influence on log P_0. Further studies of the factors influencing log P_0 are in progress.

A more precise way of comparing hemolysis and narcosis can be made using Eq. 30.

ROH causing −5-mV change in resting potential of lobster axon (Table 1*BB*):

$$\text{Log } \frac{1}{C} = 0.864(\pm 0.16) \log P - 0.099(\pm 0.13), \quad (30)$$

$$n = 5, \quad r = 0.995, \quad s = 0.082$$

While an enormous amount of work has been done on the effect of various drugs on nerve potential (38), that of Houck (27), embodied in Eq. 30, shows nicely to what degree the resting potential must be changed to compare with hemolysis of alcohols as correlated by Eq. 8, and of other narcotic effects as correlated (31) by Eqs. 31–34.

ROH toxicity to red spider:

$$\text{Log } \frac{1}{C} = 0.69 \log P + 0.16, \quad (31)$$

$$n = 14, \quad r = 0.979, \quad s = 0.087$$

ROH I_{50} for lung oxygen consumption:

$$\text{Log } \frac{1}{C} = 0.90 \log P + 0.16, \quad (32)$$
$$n = 7, \quad r = 0.995, \quad s = 0.106$$

ROH I_{50} for paramecium mobility:

$$\text{Log } \frac{1}{C} = 0.96 \log P + 0.33, \quad (33)$$
$$n = 8, \quad r = 0.998, \quad s = 0.086$$

Narcosis of frog heart by miscellaneous neutral compounds:

$$\text{Log } \frac{1}{C} = 0.93 \log P + 0.11, \quad (34)$$
$$n = 28, \quad r = 0.975, \quad s = 0.182$$

The slopes and intercepts of Eqs. 31–34 are indeed close to those of Eqs. 30 and 6–10.

In summarizing the above work, we can compare the intrinsic membrane-perturbing character of different chemical functions on an isolipophilic basis. The average intercepts, from the equations on hemolysis summarized in Table 5, constitute a tentative reference scale. Consideration of this scale with respect to drug design is worthwhile. It is of course well known that functions such as R_4N^+, R_3NH^+, $R_2NH_2^+$, and $R—NH_3^+$ have profound effects on the nervous systems of the higher organisms. Our results support the view that this is to be attributed to nerve membrane perturbation. If this is so, why do not anions such as $RCOO^-$ and $ROSO_3^-$ find use as muscle relaxants, analgesics, etc.? The answer appears to be that anions are strongly bound by serum protein while cations are not. Thus serum protein serves as a buffer to protect membranes from lipophilic anions.

In an excellent review Seeman (39) discusses the effects of many drugs on membranes. In general, most organic compounds appear to stabilize membranes at low concentrations, but at higher concentrations they cause disruption of the membrane. Seeman summarizes evidence suggesting that it is the protonated form of analgesics and tranquilizers which is biologically active. If indeed these compounds bring about their action through membrane perturbation, as the evidence suggests, this is in line with the results summarized in Table 5. Both neutral and ionic molecules appear to perturb the red cell membrane in the same way (i.e., they have the same coefficient with log P); however, the much greater intercept with the positive ions indicates that these molecules are strongly attracted to electron-rich sites in the membrane. This in itself no doubt perturbs the membrane, but in addition each increment of lipophilic character added to the charge causes further perturbation up to the point of log P_0. Our work supports the idea that the protonated amine is the more active form. If the extensive studies of Seeman *et al.* (40, 41) with erythrocyte membranes can be used as a general model for the influence of drugs on membranes, as our results seem to indicate, then the application of extrathermodynamic correlations (1) to hemolysis and stabilization against hemolysis (39) should provide greater insight into the molecular features of drugs which stabilize and/or destabilize membranes.

TABLE 5

Relative pharmacophoric scale

Function	Relative membrane-perturbing ability on logarithmic scale
R_4N^+	2.9
$RCOO^-$	2.8
$ROSO_3^-$	2.1
R_3NH^+, RNH_3^+	1.3
Neutral functions such as ROH, RCOOH, RCOOR, RCOR, ROR	0.0

The above extrathermodynamic relationships correlating hemolysis, narcosis, and antibacterial action serve to illustrate the advantages of using partition coefficients from the octanol/water system as a common language for comparing quite different sets of congeners acting on quite different systems. Not only are the data stored in compact, machine-retrievable form, but the set of equations also serves to predict hemolytic, narcotic, and antibacterial action for an enormous number of drugs not yet tested. Since partition coefficients can themselves be

calculated (28, 31), the activities of molecules not yet synthesized can be predicted within the limits set by the above experimental systems. It is hoped that as more experimental work is cast in such terms, a serious start can be made in placing comparative pharmacodynamics on a numerical basis.

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