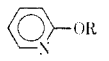
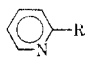
TABLE II

RELATIVE NICOTINE-LIKE ACTIVITY SUPERDELOCALIZABILITY AT RING POSITIONS 2 AND 6 AND THE ENERGY OF THE HIGHEST OCCUPIED MOLECULAR ORBITAL OF PHENYL AND *meta*-SUBSTITUTED-PHENYL CHOLINE ETHERS

	Rel nicotine-like act., *M* basis[b]			Superdelocalizability ring position		
Structure[a]	1	2	3	2	6	HOMO
C_6H_5OR	1.0	1.0	1.0	0.9751	0.9751	0.7692
m-IC_6H_4OR	5.4 ± 0.5	5.4 ± 0.4	6.3 ± 0.3	0.9947	0.9928	0.4812
m-BrC_6H_4OR	2.9 ± 0.2	3.5 ± 0.4	3.2 ± 0.3	0.9869	0.9837	0.7393
m-ClC_6H_4OR	1.6 ± 0.2	1.8 ± 0.2	2.4 ± 0.3	0.9779	0.9742	0.7671
m-FC_6H_4OR	1.1 ± 0.1	1.5 ± 0.2	1.5 ± 0.2	0.9746	0.9706	0.7685
m-$H_2NC_6H_4OR$	3.3 ± 0.3	3.7 ± 0.4	3.7 ± 0.4	1.2800	1.2781	0.5322
m-$O_2NC_6H_4OR$	0.4 ± 0.08	0.7 ± 0.1	0.4 ± 0.07	0.9483	0.9228	0.7738
			0.5 ± 0.05			0.8040
			0.2 ± 0.01			0.9467

[a] R = $(CH_2)_2N(CH_3)_3{}^+Br^-$. [b] Coleman, *et al.*[2b] 1: blood pressure of dog, 2: blood pressure of cat, 3: cat superior cervical ganglion (nictitating membrane).

increase or decrease the intensity of activity. If such be the case, what is the nature of the intermolecular forces involved? The ether could sterically approximate the receptor, be bound by electrostatic attraction, form H bonds, enter into complex formation, or act as an electron donor or acceptor. While steric factors are frequently important, they cannot explain the observed difference in activity of these compounds since they have a constant spatial disposition. The lack of correlation with π-charge distribution speaks against electrostatic interaction. Furthermore, if an electron-transfer mechanism is involved, the negative correlation with the energy of the lowest empty molecular orbital indicates that the ethers do not act *via* electron acceptance. Thus, despite the crudeness of the theoretical data discussed here, the close relationship between HOMO and superdelocalizability of atoms at the 2 and 6 ring positions and nicotine-like activity suggests that aromatic ring interacts with a secondary group(s) in the receptor by formation of a charge-transfer complex.

Comparison of Parameters Currently Used in the Study of Structure–Activity Relationships[1]

A. LEO, CORWIN HANSCH, AND C. CHURCH

Department of Chemistry, Pomona College, Claremont, California 91711

Received May 1, 1969

The reactivities of a large group of miscellaneous molecules, as measured in four different biological systems, were correlated using the following parameters: octanol–water partition coefficient, polarizability, molar attraction constant, parachor, adjusted parachor, and molecular weight. Three of the systems show a linear dependence upon these parameters and the fourth requires the addition of a squared term. Regression analysis shows that log *P* (octanol–water) correlates a greater percentage of the biological activity of the 70 compounds than the other parameters studied. Other reasons for the perferred use of log *P* are also given.

Ever since the work of Meyer and Overton at the turn of the century, efforts have been made to find suitable physicochemical parameters with which one could correlate the difference in biological activity of the members of a set of congeners.[2,3] These studies have usually found the best correlations in biochemical or pharmacological examples where "nonspecific toxicity" was being considered. In fact, the best definition of "nonspecific toxicity" might well be high correlation with a single physical constant such as an oil–water partition coefficient. While partition coefficients[4,5] have been the favorite parameter, others have also been studied. However, almost no comparisons have been made of the various parameters on the same biological data. At this stage of development it is quite important to have some idea of the relative merits of the different kinds of constants. In this report we are most interested in comparing octanol–water partition coefficients with other physical constants. A large number of systems have now been analyzed using log *P* or π from this system.[2,3,5]

In selecting sets of biological data, a number of criteria have guided our choice. We have looked for sets of data in the simplest systems where past experience has indicated that nonspecific toxicity appeared to follow lipophilic character of the drugs. We also chose data where a good variety of structural change was present in the set of congeners. As Meyer and Hemmi pointed out,[4] there is little to be gained by comparing homologous series. We also chose sets with relatively large numbers of drugs having a good spread in activity. The parameters we have selected for comparison with log *P* (octanol–water) are polarizability,

(1) This work was supported by Grant CA 11110 from the National Institutes of Health.

(2) C. Hansch, *Ann. Rept. Med. Chem., 1966*, 347 (1967).

(3) C. Hansch, *ibid., 1967*, 348 (1968).

(4) K. H. Meyer and H. Hemmi, *Biochem. Z.*, **277**, 39 (1935).

(5) C. Hansch in "Medicinal Chemistry," Vol. I, E. J. Ariëns, Ed., Academic Press, Inc., New York, N. Y., in press.

α,[6-8] molar attraction constant, F,[9] parachor, P_r,[10] adjusted parachor, P_r^*,[10,11] and molecular weight. As indicated in the associated references, all of these parameters have been under study in structure–activity investigations. Molecular weight has been included since it has been shown[12] to be a large component of many physical constants.

Thus a parameter such as log P or parachor only becomes interesting when correlations obtained with it are better than those obtained with molecular weight alone.

Method

The biological data and physical constants used in this study are given in Table I. Besides the direct comparison of a given parameter with log P, we determined whether each could add significantly to the correlation obtained with log P alone. Finally, we compared the correlations obtained with the various parameters in a set where the activities had been shown to be correlated by the exponential equation: log $(1/C) = a \log P + b(\log P)^2 + c$.

Polarizability has long been recognized as being important in the interaction of small molecules with proteins.[13] Agin, *et al.*,[6] used electronic polarizability to correlate the narcotic activity of a group of miscellaneous compounds on frog muscle. Of the 39 compounds they tested, we were able to obtain reliable parachor, molar attraction constant, and log P values for 23, which are reported in Table I. They considered the forces of absorption to be the sum of Keesom energy, Debye energy, Longon energy, and repulsion energy. For relatively nonpolar molecules they postulated that the Keesom, Debye, and repulsive energies are either small enough to be ignored, or that they cancel each other. The remaining London interaction energy is proportional to α, the electronic polarizability (expressed in cubic centimeters), and the ionization potential, I (expressed in electron volts). The correlation relationship used by Agin, *et al.*, is[14]

$$\log (1/C) = k\alpha I + \text{constant} \tag{1}$$

In the visible light range, molar refractivity results almost entirely from electronic polarizability. In 1880, Lorenz–Lorenz derived the following equation from the electromagnetic theory of light

$$R = \alpha = \frac{(n^2 - 1)M}{(n^2 + 2)d} \tag{2}$$

where n = refractive index of visible light, M = molecular weight, d = density (at the temperature quoted for n, usually 20°). For this study we used α values calculated from n, m, and d whenever these data were available.[15]

Molar Attraction Constant.—Recently, Ostrenga[9] has shown that there is a correlation between the biological activity of certain sets of congeners and the molar attraction constants (F). He noted that if only the derivatives of a given parent compound are being studied, one can use the calculated F value for the variable portion of the molecule. Mullins[16] calculated the energy necessary to desolvate a narcotic molecule (as it entered a lipophilic membrane) in the following manner

$$P_{nar}/P_0 = X_{nar}e^{w/kT} \tag{3}$$

assuming dilute solution where Raoult's law holds and where P_{nar} = partial pressure of narcotic in solution, P_0 = vapor pressure of narcotic in standard state, X_{nar} = mole fraction of narcotic, w = interchange energy, *i.e.*, the energy to exchange a molecule of solvent for one of solute. For solutions deviating from Raoult's law (*i.e.*, $A_{nar} \neq X_{nar}$), the deviation is usually expressed in terms of an activity coefficient, $\gamma = A/X$, or in terms of partial pressure, $\gamma_{nar} = (P_{nar}/P_0)/X_{nar}$. Making this substitution in eq 3 and rearranging gives

$$\ln \gamma_{nar} = w/kT \tag{4}$$

Hildebrand[17] has related w to the heat of vaporization and molar volume, specifically to the difference in the term $(H^{vap}/V_m)^{1/2}$ for solvent and solute. He designates the solubility parameter δ as $\delta = (H^{vap}/V_m)^{1/2}$. The term δ^2 is referred to as the cohesive energy density of the liquid. For dilute solutions, where solvent and solute have similar molar volumes (and entropy of mixing is negligible)

$$w = V_m(\delta_1 - \delta_2)^2 = \text{heat of mixing} \tag{5}$$

Mullins used the experimentally determined values of H^{vap} for the lower alcohols, and for V_m he used the molar volumes of the compounds as dilute solutions in heptane. Although preferred on theoretical grounds, these procedures gave molar attraction constants which were difficult to correlate with biological data, and so in this paper we use a simpler approximation suggested by Burrell.[18]

Except for compounds boiling below 55°, Burrell assumed that boiling point and ΔE were proportional and also that V_m could be calculated from the density of the pure compound. Thus

$$\delta = (\Delta E/V_m)^{1/2} \tag{6}$$

and

(6) D. Agin, L. Hersh, and D. Holtzman, *Proc. Natl. Acad. Sci. U. S.*, **53**, 952 (1965).

(7) A. Cammarata, *J. Med. Chem.*, **10**, 525 (1967).

(8) J. W. McFarland, L. Conover, A. Howes, J. L. Lynch, O. Courtney, and D. Morgan, 155th National Meeting of the American Chemical Society, San Francisco, Calif., 1968.

(9) J. A. Ostrenga, *J. Med. Chem.*, **12**, 349 (1969).

(10) J. C. McGowan, *J. Appl. Chem.*, **4**, 41 (1954).

(11) (a) J. C. McGowan, *Rec. Trav. Chim. Pays-Bas*, **75**, 193 (1956); (b) J. C. McGowan, *Nature*, **200**, 1317 (1963).

(12) O. Exner, *Collect. Czech. Chem. Commun.*, **32**, 1 (1967).

(13) L. Pauling and D. Pressman, *J. Am. Chem. Soc.*, **67**, 1003 (1945).

(14) Since the oscillation frequency should be used instead of I, I may be a poor approximation for polyatomic molecules. Even though an F test with the data of Agin, *et al.*,[6] shows that I adds significantly to the correlation obtained with α alone, the improvement was small, r^2 being increased from 0.92 to 0.96. The great difficulty in obtaining I values almost excludes it from consideration as a suitable parameter, and therefore we have used α values alone in this study.

(15) *E.g.*, for $PhNO_2$, d = 1.205, M = 123.1, and n = 1.553. Thus α = (1.41/4.41)(123.1/1.205) = 32.7. The sum of atomic polarizabilities is 6 × C(2.42) + 5 × H(1.1) + 1 × NO_2(7.30) + 3 double bonds(1.73) = 32.5. Atomic polarizabilities were taken from the compilation of R. Shriner, R. Fuson, and D. Curtin, "The Systematic Identification of Organic Compounds," 4th ed, John Wiley and Sons, Inc., New York, N. Y., 1956, p 50. Also see N. Bauer and S. Z. Lewin, "Techniques of Organic Chemistry," Vol. I, Physical Methods, Pt. II, A. Weissberger, Ed., 3rd ed, Interscience Publishers, Inc., New York, N. Y., 1960, pp 1162–1194.

(16) L. J. Mullins, *Chem. Rev.*, **54**, 289 (1954).

(17) J. H. Hildebrand and R. L. Scott, "The Solubility of Nonelectrolytes," 3rd ed, Reinhold Publishing Co., Inc., New York, N. Y., 1950.

(18) H. Burrell in "Polymer Handbook," J. Brandrup and E. H. Immergut, Ed., Interscience Publishers, Inc., New York, N. Y., 1966, Vol. IV, p 341.

TABLE I

Compd	Log P	α	F^*	P_f	P_f^*	Log (1/C) obsd
			Tadpole Narcosis[b]			
Methanol	−0.66	8.20	0.10	86.0	−0.14	0.24
Ethanol	−0.16	12.90	1.50	126.0	0.31	0.54
Propanol	0.34	17.50	2.80	166.0	0.79	0.96
Butanol	0.84	22.10	4.00	206.0	1.27	1.42
Octanol	2.84	40.60	8.70	366.1	3.20	3.40
2-Propanol	0.14	17.60	2.40	166.0	0.78	0.89
Isobutyl alcohol	0.64	22.10	3.75	206.0	1.27	1.35
t-Butyl alcohol	0.37	21.90	3.35	206.0	1.27	0.89
Isoamyl alcohol	1.14	26.80	5.00	246.0	1.75	1.64
t-Amyl alcohol	0.89	26.80	4.50	246.0	1.75	1.24
1,3-Chloro-2-propanol	0.92	27.20	5.20	227.2	1.52	1.92
Thymol	3.30	46.90	14.40	380.7	3.97	4.26
1,3-$(MeO)_2C_6H_4$	2.09	38.90	12.90	326.0	2.71	3.35
1,4-$(MeO)_2C_6H_4$	2.09	38.90	12.90	326.0	2.71	3.05
Acetone	−0.21	16.20	1.80	158.3	0.70	0.54
2-Butanone	0.29	20.80	3.10	199.0	1.19	1.04
3-Pentanone	0.79	25.00	4.20	236.0	1.63	1.54
2-Pentanone	0.79	25.00	4.20	236.0	1.63	1.72
Acetophenone	1.58	36.10	11.84	293.0	2.32	3.03
Acetal	0.86	33.20	5.60	311.6	1.34	1.98
Ethyl ether	0.77	22.50	2.65	217.0	1.34	1.57
Anisole	2.11	32.70	10.60	266.2	2.59	2.82
Methyl acetate	0.23	17.45	2.16	175.1	0.90	1.10
Ethyl formate	0.33	17.80	2.10	175.1	0.90	1.15
Ethyl acetate	0.73	22.20	3.34	215.0	1.38	1.52
Ethyl propionate	1.23	26.70	4.55	255.1	1.86	1.96
Propyl acetate	1.23	26.90	4.71	255.1	1.86	1.96
Ethyl butyrate	1.73	31.30	5.80	295.1	2.34	2.37
Ethyl isobutyrate	1.53	31.30	5.60	295.1	2.34	2.24
Butyl acetate	1.73	31.50	6.00	295.1	2.34	2.30
Isobutyl acetate	1.53	31.50	5.20	295.1	2.34	2.24
Ethyl valerate	2.23	36.20	7.10	335.1	2.82	2.72
Amyl acetate	2.23	36.10	7.20	335.1	2.82	2.72
Butyl valerate	3.23	45.30	9.40	415.1	3.78	3.60
Diethyl tartrate	−0.29	46.00	11.20	426.2	0.30	1.21
Methyl carbamate	−0.65	16.90	3.65	162.5	−0.45	0.57
Ethyl carbamate	−0.15	21.50	5.10	202.5	0.03	1.39
Phenyl carbamate	1.08	31.14	14.40	296.3	1.15	3.19
Pentane	2.50	25.20	8.10	231.4	2.77	2.55
Pentene	2.30	24.70	7.70	219.9	2.63	2.64
Benzene	2.13	26.20	8.19	206.1	2.47	2.68
Xylene	3.25	35.50	10.80	286.1	3.43	3.42
Naphthalene	3.37	41.65	11.85	307.6	3.68	4.19
Phenanthrene	4.46	57.01	16.40	425.8	5.10	5.43
Ethyl chloride	1.39	16.20	6.40	151.0	1.81	2.35
Ethyl bromide	1.60	18.90	6.54	164.5	1.97	2.57
Ethyl iodide	2.00	24.20	7.44	186.0	2.23	2.96
Ethylene chloride	1.78	20.90	7.70	190.5	2.28	2.64
Chloroform	1.97	21.40	7.50	190.0	2.28	2.85
Nitromethane	−0.33	12.40	6.85	129.8	1.55	1.09
Acetonitrile	−0.34	11.00	6.35	120.0	0.24	0.44
Azobenzene	3.84	48.40	15.40	434.1	5.20	4.74
Acetaldoxime	−0.13	16.10	2.26	139.0	0.47	0.92
			Frog Muscle Narcosis[6]			
Methanol	−0.66	8.2	0.10	86.0	−0.14	−0.09
Ethanol	−0.16	12.9	1.50	126.0	0.31	0.25
Acetone	−0.21	16.2	1.80	158.3	0.70	0.40
Propanol	0.34	17.5	2.80	166.0	0.79	0.60
Ethyl ether	0.77	22.5	2.65	212.0	1.34	1.07
Butanol	0.84	22.1	4.00	206.0	1.27	1.22
Benzyl alcohol	1.10	32.5	7.60	262.6	1.96	1.70
Pentanol	1.34	26.8	5.60	246.0	1.75	1.80
Hexanol	1.84	31.4	6.40	286.0	2.20	2.44
Heptanol	2.34	36.0	7.50	326.0	2.70	2.80
Octanol	2.84	40.6	8.70	366.0	3.20	3.16
2-Propanol	0.14	17.6	2.40	166.0	0.79	0.45
Urethan	−0.15	21.5	5.10	202.5	0.03	1.00
Pyridine	0.65	24.1	8.64	191.0	1.10	1.23

TABLE I (*Continued*)

Compd	Log P	α	F^*	P_r	P_r^*	Log (1/C) obsd
			Frog Muscle Narcosis[b]			
Aniline	0.90	30.5	5.40	234.5	1.60	1.70
Phenol	1.46	27.8	9.95	221.0	2.05	2.00
Nitrobenzene	1.85	32.5	11.40	264.5	3.17	2.53
Thymol	3.30	46.9	14.40	380.7	3.97	3.52
Acetanilide	1.16	38.1	13.40	319.0	2.60	1.83
2-Naphthol	2.84	45.4	13.50	339.0	3.46	3.00
Quinoline	2.03	42.1	12.80	309.0	2.52	2.70
Hydroquinone	0.59	29.4	11.30	235.2	1.62	1.60
Antipyrine	0.23	54.6	13.00[a]	422.0	2.66	1.22
			Complex with BSA[c]			
Phenol	1.46	27.8		220.6	2.05	3.32
3-Fluorophenol	1.93	26.7		231.0	2.17	3.86
4-Fluorophenol	1.77	24.9		231.0	2.17	3.52
3-Chlorophenol	2.50	32.7		260.2	2.53	4.30
4-Chlorophenol	2.39	32.7		260.2	2.53	4.00
4-Bromophenol	2.59	35.6		273.7	2.69	4.22
4-Iodophenol	2.91	40.1		295.3	2.94	4.40
4-Methylphenol	1.94	32.5		260.6	2.53	3.70
3-Ethylphenol	2.40	37.1		300.6	3.01	4.22
3-Trifluoromethylphenol	2.95	32.3		291.7	2.90	4.52
3-Cyanophenol	1.22	32.9		269.3	1.43	3.26
3-Hydroxyphenol	0.80	29.3		235.2	1.62	3.15
3-Methoxyphenol	1.58	33.9		280.7	2.17	3.54
4-Methoxyphenol	1.34	33.9		280.7	2.17	3.40
3-Nitrobenzonitrile	1.17	38.0		313.2	2.56	2.94
4-Methoxybenzyl alcohol	1.10	39.3		320.7	2.05	2.94
Benzonitrile	1.56	31.8		254.7	1.86	3.23
Acetophenone	1.58	36.1		293.0	2.32	3.31
Nitrobenzene	1.85	32.5		264.5	3.17	3.58
4-Bromoacetanilide	2.18	45.9		372.2	3.26	4.00
4-Nitroanisole	2.03	38.9		324.6	3.30	4.00
4-Chloronitrobenzene	2.39	37.4		304.1	3.65	4.07
2,4-$Cl_2C_6H_3NO_2$	2.93	42.2		343.7	4.12	4.59
Naphthalene	3.37	41.6		307.6	3.68	4.91
Azobenzene	3.82	48.4		434.1	4.00	5.29
Anisole	2.11	32.7		266.2	2.59	4.00
3-Fluoroaniline	1.30	30.5		244.4	1.73	3.09
4-Chloroaniline	1.83	35.6		273.6	2.08	3.68
4-Methoxyaniline	0.78	37.0		294.1	1.73	2.92
4-Bromoaniline	2.03	38.3		287.1	2.24	4.06
4-Methylaniline	1.39	35.1		274.0	2.09	3.30
1-Naphthylamine	2.23	46.0		336.0	2.83	3.94
Indole	2.14	37.6		272.4	3.66	4.07
Neopentyl alcohol	1.36	26.8		275.6	2.11	3.47
Camphorquinone	1.52	44.0		377.9	2.16	3.17
1-Hydroxyadamantane	2.14	43.3		352.0	3.02	3.94
Thymol	3.30	46.9		380.6	3.96	4.66
unsym-Methylphenyl-thiourea	0.85	51.5		378.3	2.15	3.30
n-Hexyl alcohol	1.84	31.2		286.2	2.24	3.94
Phenylethyl carbamate	2.30	45.5		377.2	2.13	3.83
Ethylamylbarbituric acid	2.24	57.6		510.5	2.54	3.66
2-Nonanone	2.79	43.5		398.0	3.58	4.33
			Chick Embryo Hatching[d]			
Acetone	−0.21	16.2	1.80		0.70	1.87
Ethanol	−0.16	12.9	1.50		0.31	1.77
Ethyl acetate	0.73	22.2	3.34		1.38	2.70
Isopropyl alcohol	0.14	17.6	2.40		0.78	2.18
Butyl acetate	1.73	31.5	6.00		2.34	2.81
Toluene	2.69	30.9	9.53		2.97	3.03
Methanol	−0.66	8.2	0.10		−0.14	1.61
Isoamyl alcohol	1.14	26.8	5.00		1.75	2.74
Butanol	0.84	22.1	4.00		1.27	2.67
Benzene	2.13	26.2	8.19		2.47	2.95

[a] Calculated from F^* for acetanilide, and group values as compiled by P. A. Small, *J. Appl. Chem.*, **3**, 75 (1953). [b] E. Overton, "Studien uber die Narkose," Fischer, Jena, Germany, 1901. [c] F. Helmer, K. Kiehs, and C. Hansch, *Biochemistry*, **7**, 2858 (1968). [d] J. McLaughlin, J. P. Marliac, M. J. Verrett, M. K. Mutchler, and O. G. Fitzhugh, *Ind. Hyg. J.*, **25**, 282 (1964).

$$F = V_m\delta = \text{molar attraction constant} \quad (7)$$

To correlate solvent–polymer miscibility data, Burrell found it necessary to *add* to the claculated values the following corrections for hydrogen bonding: alcohols = 1.4, esters = 0.6, and low-boiling ketones = 0.6. In correlating biological data, we found that a correction of opposite sign was required. We preferred to apply an equal *negative* correction of 5×10^2 to all calculated F values for aliphatic alcohols, aliphatic carbonyl groups, primary and secondary amines, nitriles, and aliphatic ethers. We applied no correction to aromatic ethers, carbonyls, or phenols.[19]

Parachor.—A third parameter which relates principally to molecular volume and which has been extensively studied by McGowan[10,11] is the parachor. It is defined as

$$P_r = (M/d)\gamma^{1/4} \quad (8)$$

where M = molecular weight, d = density, γ = surface tension in dynes/cm^2. The majority of parachor values used in this study were calculated from surface tension data.[20] Where this constant was not available, "bond" parameters of Vogel, *et al.*,[21] were employed.

McGowan has pointed out the relationship of physical toxicity and narcosis to parachor.[10] He also suggested a correction to the parachor of associated compounds which made it possible to use the same correlation as with nonpolar compounds. In this study we shall employ his correction with the following terminology: P_r = parachor calculated from eq 8, P_r^* = $0.012P_r$ for nonpolar compounds, $P_r^* = 0.012P_r - 0.6$ for compounds containing a phenolic OH or phenolic ether function, $P_r^* = 0.012P_r - 1.2$ for compounds containing carbonyl, ester, amine, nitrile, OH, or aliphatic ether functions; in contrast to the case of F^*, we obtained better correlations when we applied a multiple correction to compounds which contained multiple functional groups.

Molecular Weight.—Each of the three parameters discussed above depends a great deal on the molecular weight of the compound. Since a degree of cocorrelation between each of the parameters and molecular weight was expected,[12] it was decided to include molecular weight as a reference parameter. From the data in Table I we have derived, *via* the method of least squares, a linear relation between log $(1/C)$ (biological activity) and each of the parameters. These results are summarized in the first three sets of data in Table II. Both the square of the correlation coefficient (r^2) and the standard deviation from regression (s) are listed for comparing the quality of the correlation.

The best test for the value of a physical constant is Overton's data on tadpole narcosis[22] which contain 53 different drugs of rather widely varying structure and biological activity. Viewing the results with the six parameters, it is seen that while F^* gives a moderately good correlation, only log P and P_r^* give results which are clearly more significant than simple molecular weight. In this most rigorous test, log P is definitely superior to P_r^*. In the second most rigorous test, that of narcosis of frog muscle, we find essentially the same result. In this set of data[6] we have used only those molecules which were not significantly ionized under the test conditions.

The third test, that of binding to bovine serum albumin, encompasses a greater number of data points but not so great a spread in log $(1/C)$ as the frog muscle narcosis. However, since the system lends itself to much more accurate measurement, and since the standard deviation is the lowest of the three, it might be considered the most rigorous test. The superiority of log P in this case is clearly evident, and the performance of either parachor or polarizability is seen to be almost completely dependent upon the choice of "correction" to be applied for the polar nature of any functional group contained in the test substance.

Four other systems which gave linear correlations with the six parameters were also examined: frog heart narcosis,[23] C-mitosis,[24] enzyme inhibition,[25] and enzyme precipitation.[25] An examination of the original work in each case indicated a lower degree of accuracy in collecting the biological data than in the three cases we are presenting in detail, and the standard deviations from regression were greater, as expected. Nevertheless, none of the other parameters were found superior to log P in any case, although P_r^* gave essentially the same results for the enzyme systems.

In considering the two best types of constants, log P and P_r^*, there are a number of reasons for choosing log P over P_r^* besides the fact that log P appears to give the best correlations in the more rigorous tests. To obtain good correlations with parachor, one must make a number of arbitrary adjustments. The correlations with the unadjusted constant are no better than using molecular weight. It is likely that good correlations could be obtained using molecular weight with adjustment for hydrogen bonding. To obtain experimental values of parachor, one must make surface tension measurements which in general are more difficult than partitioning measurements. With compounds of complex structure it will be hard to decide what kind of correction on P_r must be made to obtain a suitable P_r^*. For instance, in the McGowan system there seems to be little to justify a full correction for aliphatic ethers, but only a half correction for phenols. Likewise, it is hard to see beforehand why a correction is assigned to aromatic secondary amines while phenols receive none.

Many instances have been found where biological activity [log $(1/C)$] is not linearly related to log P or other such constants.[26] One such example where there there was considerable variation in the structure of the drugs is that of inhibition of chick embryo hatching.[27] From the data in Table I the results in Table II are obtained. Again we find log P to yield the best correlation. In a variety of other examples which space limita-

(19) *E.g.*, for butanol, d = 0.81, M = 74.1, bp 117°, V_m = 91.5, ΔE = 8800 cal, δ 9.9, $F = 9.1 \times 10^2$, $F^* = 4.1 \times 10^2$.

(20) *E.g.*, for nitromethane, γ = 36.82, M = 61, d = 1.14, $P_r = (2.462/1.14)61 = 132$. The calculated value from Vogel, *et al.*,[21] bond parachors is 130.

(21) A. I. Vogel, W. T. Cresswell, G. J. Jeffery, and J. Leicester, *Chem. Ind.* (London), 358 (1950).

(22) See footnote *b* in Table I.

(23) H. Fühner, *Biochem. Z.*, **120**, 143 (1921).

(24) G. Ostergren, "Mechanisme de la Narcose," Centre National de la Recherche Scientific, Paris, 1950, p 77.

(25) F. Battelli and L. Stern, *Biochem. Z.*, **52**, 226 (1913).

(26) (a) C. Hansch, A. R. Steward, S. M. Anderson, and D. Bentley, *J. Med. Chem.*, **11**, 1 (1968); (b) E. J. Lien, C. Hansch, and S. M. Anderson, *ibid.*, **11**, 430 (1968).

(27) See footnote *a* in Table I.

Table II
Results from the Correlation Equation[a]

Biological system	n	X: Log P r^2	Log P s	α r^2	α s	Mol wt r^2	Mol wt s	F^* r^2	F^* s	P_r r^2	P_r s	P_r^* r^2	P_r^* s
						Log $(1/C) = aX + b$							
Tadpole narcosis[b]	53	0.913	0.343	0.683	0.654	0.567	0.765	0.758	0.571	0.556	0.775	0.861	0.434
Narcosis frog muscle[c]	23	0.944	0.242	0.630	0.623	0.569	0.672	0.617	0.634	0.659	0.598	0.841	0.408
1:1 M complex with bovine serum albumin[c]	42	0.920	0.159	0.094	0.536	0.163	0.515	Insufficient data		0.095	0.536	0.648	0.334
						Log $(1/C) = aX + bX^2 + c$							
I_{50} chick embryo hatching[d]	10	0.965	0.112	0.909	0.179	0.856	0.226	0.933	0.155			0.923	0.165

[a] See text for definition of parameters represented by X. n = number of data points used; r^2 = square of the correlation coefficient and can be taken as the per cent of the variance in the data "explained" by the regression; s = the standard deviation from regression. [b-d] See corresponding footnotes in Table I.

tions do not allow us to include, we have found results like those of Table II.

In making comparisons of the above type, care must be taken to select meaningful data. As Meyer and Hemmi[4] pointed out in comparing different solvent reference systems for log P, nothing is to be gained by using homologous series for comparisons. One can also see from a comparative study of different sets of biological data that if, say, only relatively apolar changes are made in a parent molecule, quite similar results can be obtained using a variety of parameters. This would appear to account for the rather close agreement Ostrenga[9] obtained in comparing F and π. The use of a variety of drugs as narcotics convinced Meyer and Hemmi that alcohols make better reference systems than esters such as olive oil. Our own studies[28] suggest that a variety of simple polar solvents would give reasonable results, but that hydrocarbons would not make good reference systems.[28] It still remains to be seen how close octanol–water fits the ideal for a reference system. In a study[5] of 54 different linear correlations based on log P (octanol–water), 47 had r values of 0.95 or better. This means that only 10% of the variance in the biological data is not accounted for. The 10% must be split between errors in determination of log P, errors in measuring log $(1/C)$, and the quality of the octanol–water model. It is not unreasonable to expect errors of 3–5% in even the best biological data and errors of 1–2% in log P. Thus it would seem that relatively little improvement could be obtained by selection of a better solvent reference system.

In extrathermodynamic correlations of the above type, the importance of choosing a reference system as close as possible to that of the one under study has been emphasized.[29] Thus it appears to us *a priori* that a model reference system such as octanol–water would be more able to account for drug distribution than a more abstract and artificial parameter such as parachor. It is our hope that log P can be used with some confidence to account for what these days is termed the hydrophobic[30] character of drugs. Not only does log P have the advantage of being relatively easily measured experimentally, it is also an additive and constitutive constant and thus may be estimated from known constants for the various constituents of a given drug.[5,31]

(28) C. Hansch, J. E. Quinlan, and G. L. Lawrence, *J. Org. Chem.*, **33**, 347 (1968).

(29) J. E. Leffler and E. Grunwald, "Rates and Equilibria of Organic Reactions," John Wiley and Sons, New York, N. Y., 1963, p 128.

(30) G. Némethy, *Angew. Chem.*, **6**, 195 (1967).

(31) C. Hansch and S. M. Anderson, *J. Org. Chem.*, **32**, 2583 (1967).

Potential Antitumor Agents. II. Effects of Modifications in the Side Chain of 1-Formylisoquinoline Thiosemicarbazone[1,2]

Krishna C. Agrawal and Alan C. Sartorelli

Department of Pharmacology, Yale University School of Medicine, New Haven, Connecticut 06510

Received February 20, 1969

A number of modifications have been made in the formyl thiosemicarbazone side chain of 1-formylisoquinoline thiosemicarbazone to ascertain the importance of this part of the molecule for antineoplastic activity; tumor-inhibitory potency and host toxicity of these compounds were assessed in mice bearing Sarcoma 180 ascites cells. Substitutions made on the different positions of the side chain resulted in either a diminution or a total loss of tumor-inhibitory activity, indicating that the intactness of this portion of the molecule was essential for 1-formylisoquinoline thiosemicarbazone to function as an inhibitor of the growth of malignant cells.

A number of α-N-heterocyclic aldehyde thiosemicarbazones, possessing the potential to form coordination compounds with certain transition metals, have been shown to be potent inhibitors of the growth of a variety of transplanted rodent neoplasms.[3] The meta-

(1) Presented in part before the Division of Medicinal Chemistry at the 155th National Meeting of the American Chemical Society, San Francisco, Calif., April 1968.

(2) This work was supported by Grant T-23 from the American Cancer Society and Grant CA-02817 from the National Cancer Institute, U. S. Public Health Service.

(3) (a) R. W. Brockman, J. R. Thomson, M. J. Bell, and H. E. Skipper, *Cancer Res.*, **16**, 167 (1956); (b) F. A. French and E. J. Blanz, Jr., *J. Med. Chem.*, **9**, 585 (1966); (c) F. A. French and E. J. Blanz, Jr., *Cancer Res.*, **25**, 1454 (1965).