TABLE III—ACTIVITY OF THE ACTIVE FRACTION FROM *C. persicum* MILL. AGAINST THE WALKER INTRAMUSCULAR CARCINOSARCOMA 256

Fraction	Dose, mg./Kg.	Survivors	Animal Wt. Change, Gm. Diff. (T-C)	Tumor Wt., mg. (Test/Control)	T/C × 100
E′	50	2/4	−14	1400/7400	Toxic
	40	4/4	−11	1900/7100	26%
	30	4/4	−7	4400/7100	61%

A larger batch of fraction A′ (9.67 Gm.), prepared from crude extract (84 Gm.), was dissolved in 5% methanol in chloroform (400 ml.) and introduced onto a column of silicic acid (1 Kg.) in chloroform. The column was eluted with 5% methanol in chloroform (4 L.), 7% methanol in chloroform (6 L.), and with 10% methanol in chloroform (10 L.), to yield fractions which were examined by TLC as before. The fractions richest in the one-spot active material were combined (E′, 4.84 Gm.) and tested in the Walker carcinosarcoma 256 assay (see Table III).

Fraction E′ was a white amorphous powder, (m.p. 229–233°); $[\alpha]_D^{28}$ = +0.8° (c 1.20 MeOH), I.R. (Nujol) 3.04 μ, 5.81 μ, 6.10 μ.

Determination of the Hemolytic Index of Fractions P, Q, and E′—The determination was performed by the method outlined in the Swiss Pharmacopoea (4) using a 1:20 dilution of blood with normal saline and Fisher standard saponin as reference. Using these conditions, fraction P had a hemolytic index of 0.8; fraction Q, 0.5; and fraction E′, 316; with reference to standard saponin at 100. Hence the *Acer negundo* active principles have low hemolytic activity.

Test for the Glycosidic Nature of Fractions P, Q, and E′—Fractions P, Q, and E′ (5 mg. of each, separately), when heated for 2 min. with a 0.5% aqueous solution of triphenyltetrazolium chloride (2 drops) and 0.5 *N* sodium hydroxide (1 drop), gave no pink color or precipitate.

Further 5 mg. portions of fractions P, Q, and E′ were hydrolyzed by refluxing for 30 min. with 0.5 *N* hydrochloric acid in 50% aqueous alcohol (0.2 ml). Each hydrolyzate was neutralized and the triphenyltetrazolium chloride test performed. The deep red solutions and red precipitates indicated the presence of reducing sugars in each of the hydrolyzates (5).

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Molecular Orbital Calculations on Sulfonamide Molecules

By E. C. FOERNZLER* and A. N. MARTIN†

The LCAO-MO method has been used to compute the electronic characteristics for a series of 50 sulfonamide derivatives. The results of the calculations have been compared with experimental parameters through correlation with pKa. For those compounds where the substituent R group was an alternate ring, the correlation of pKa and the electronic charge at the ionizing (N^1) nitrogen atom was considered significant. However, it was necessary to classify the compounds by type of R group, plotting each class separately. The simple Hückel MO treatment was inadequate for those molecules where the substituent R group attached to the N^1 nitrogen atom was a nonalternate ring or a straight chain.

IN RECENT YEARS it has become possible to study the electronic structure of large organic molecules by using the principles of quantum mechanics. This field is known as quantum chemistry, and when applied to molecules of biological interest, has been called quantum biology or quantum biochemistry (1–4). The molecular orbital (MO) method, involving a linear combination of atomic orbitals (LCAO), is ordinarily used for calculating the energies of the π or delocalized electrons in a molecule, and the abbreviation LCAO-MO is used to designate this type of calculation. In this study, a modification of the LCAO-MO method, the simple Hückel molecular orbital (HMO) approximation (3–6), has been employed.

Actually, the reactivity of drug molecules should be discussed in terms of their dynamic

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behavior in going into a transition state along with a specific receptor or reagent (7). However, where such an approach is either impossible or too complex, useful information may be obtained by considering various static properties of the molecules, such as the electronic charge densities relative to the total π or mobile electron systems of the molecules in their ground states. Calculation of the distribution of electronic charge indicates the average density of mobile electrons present on different atoms in the molecule. The higher the calculated charge, the greater the π electron density associated with a particular atom. It is also possible to compute bond orders, free valence, and the relative values of the highest occupied (homo) and lowest empty (lemo) molecular orbitals. This information, together with other chemical knowledge may be useful for predicting various physical-chemical properties and even biological activities of drug molecules.

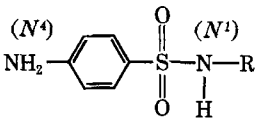
During the past two and one-half decades, a number of theories have been advanced on the mechanism of sulfonamide action. Bell and Roblin (8), Kumler and associates (9–11), Klotz and Bordwell (12–13), and Krüger-Thiemer *et al.* (14) have proposed possible modes of action. Although these theories present different explanations of the mechanism of sulfonamide action, they are all based on the original suggestion of Woods (15) and Fildes (16) that the sulfonamides interfere with the utilization of *p*-aminobenzoic acid in enzyme systems through competitive enzyme inhibition.

In this study the authors have computed the electronic charge densities for 50 sulfonamide compounds. The results of the computations have allowed an examination of the utility of HMO calculations applied to complex drug molecules, and have resulted in additional knowledge which may be useful for further elucidating the mechanism of sulfonamide action.

TREATMENT OF SULFONAMIDE MOLECULES

Compounds Studied—The 50 sulfonamide molecules selected for the calculations were those studied by Bell and Roblin (8). These molecules are all N^1-substituted sulfanilamide type compounds of the general structure I, and are listed in Table I according to nomenclature and numbering used by Bell and Roblin. The chemical structures of these compounds are given in a text by Northey (17). The experimental data, including the acid dissociation constants and the *in vitro* activities, C_R, were taken from the published results of Bell and Roblin.

The activity index, C_R, in Table I represents the minimum molar concentration of drug necessary to cause bacteriostasis of *E. coli* in a buffered (pH 7) synthetic medium under standardized conditions. The smaller the number, the greater the activity. As pointed out by Bell and Roblin, it is difficult to obtain a high degree of precision in such tests and small differences in bacteriostatic activity (a factor of 2) are within experimental error.



I

Sulfanilamide type compounds are weak acids because the amide (N^1) nitrogen atom of these molecules is sufficiently negative in solution to liberate a proton. Due to their high electronegativity, the oxygen atoms of the sulfonyl (SO_2) group greatly attract electrons. The resulting electron deficiency of the sulfur atom results in the electrons of the N:H bond being held more closely to the nitrogen atom. The hydrogen, therefore, is bound less firmly and readily ionizes to liberate a proton in solution. In addition to the acid dissociation constants, Bell and Roblin also measured the basic dissociation constants which are due to the ionization of the *p*-amino group or amino (N^4) nitrogen atom. These values were found to be approximately constant [$(0.5–2.3) \times 10^{-12}$]. A striking difference was thus observed between the acid and basic dissociation constants.

The R group is the only variable involved in the N^1-substituted sulfonamide derivatives, and it must be the factor controlling the acid dissociation constants of these compounds (8). In other words, the greater the electronegativity of the R group, the more R attracts electrons from the adjacent amide (N^1) nitrogen so that the hydrogen escapes more easily as a proton in solution leaving the anion form of the molecule. However, according to Bell and Roblin, as the electron-attracting power of different R groups increases, the SO_2 group should become less negative (thereby decreasing activity), because under these conditions a greater part of the ionic charge of the amide (N^1) nitrogen will be taken by the R group which will be competing more strongly with the SO_2 group for the ionic charge. As the SO_2 group becomes less negative, its ion should be less active than an ion whose R group is a weaker electron-attracting group. Similar conclusions are obtained by applying the same reasoning to the molecular form.

By calculating the electronic charge densities of the sulfanilamide type compounds, we would expect to find a relationship between the pKa and the electronic charge at the ionizing (N^1) nitrogen atom. The relationship of pKa to electronic charge has been discussed by Daudel *et al.* (3, 18–20), Longuet-Higgins (21), the Pullmans (22–24), and others (25).

Calculations—The linear combination of atomic orbitals-molecular orbital (LCAO-MO) method was employed for the calculations, using the Hückel molecular orbital (HMO) approximation with neglect of overlap (3–6). For the heteroatom (or substituent group) X, the Coulomb integral (α_X) and resonance or bond integral (β_{CX}) were given the usual forms shown in Eqs. 1 and 2:

$$\alpha_X = \alpha_C + \delta_X\beta_{CC} \qquad \text{(Eq. 1)}$$

TABLE I—SULFONAMIDE DERIVATIVES USED FOR LCAO-MO CALCULATIONS (8)

No.	Compd.[a]	pKa	K_a	$C_R \times 10^5$
1	*p*-Aminobenzoic acid	4.68	2.1×10^{-5}	...
2	Sulfanilamide	10.43	3.7×10^{-11}	20.0
3	N^1-Methylsulfanilamide	10.77	1.7×10^{-11}	30.0
4	N^1,N^1-Dimethylsulfanilamide	...	...	30.0
5	N^1-Hydroxyethylsulfanilamide	10.92	1.2×10^{-11}	50.0
6	Sulfanilylglycine	3.52	3.0×10^{-4}	[>90.0
7	N^1-Phenylsulfanilamide	9.60	2.5×10^{-10}	3.0
8	N^1-*o*-Tolylsulfanilamide	9.96	1.1×10^{-10}	10.0
9	N^1-*m*-Tolylsulfanilamide	9.74	1.8×10^{-10}	5.0
10	N^1-*p*-Tolylsulfanilamide	9.82	1.5×10^{-10}	5.0
11	N^3-Sulfanilylmetanilamide	8.23	5.9×10^{-9}	2.0
12	N^4-Sulfanilylmetanilamide	7.85	1.4×10^{-8}	0.5
13	N^1-*p*-Aminophenylsulfanilamide	10.22	0.6×10^{-10}	5.0
14	N^1-Furfurylsulfanilamide	10.88	1.3×10^{-11}	20.0
15	Sulfapyridine	8.43	3.7×10^{-9}	0.6
16	3-Sulfanilamidopyridine	7.89	1.3×10^{-8}	0.2
17	2-S-5-Bromopyridine	7.15	7.1×10^{-8}	0.5
18	5-S-2-Bromopyridine	7.12	7.6×10^{-8}	0.2
19	2-S-5-Aminopyridine	8.47	0.34×10^{-8}	0.6
20	5-S-2-Aminopyridine	8.82	0.15×10^{-8}	2.0
21	2-Sulfanilamidoimidazole	9.72	1.9×10^{-10}	40.0
22	3-Sulfanilamidopyridazine	7.06	0.87×10^{-7}	0.08
23	Sulfadiazine	6.48	3.3×10^{-7}	0.08
24	2-S-4-Methylpyrimidine	7.06	0.87×10^{-7}	0.2
25	2-S-4,6-Dimethylpyrimidine	7.37	0.43×10^{-7}	0.3
26	2-S-4-Aminopyrimidine	9.44	3.6×10^{-10}	20.0
27	4-S-Pyrimidine	6.17	6.7×10^{-7}	0.1
28	5-S-Pyrimidine	6.62	2.4×10^{-7}	0.2
29	5-S-2-Chloropyrimidine	5.80	1.6×10^{-6}	0.1
30	2-Sulfanilamidopyrazine	6.04	0.91×10^{-6}	0.08
31	4-S-1,2,4-Triazole	4.66	2.2×10^{-5}	>80.0
32	2-Sulfanilamidooxazole	6.50	3.2×10^{-7}	0.08
33	5-S-3-Methylisoxazole	4.20	6.3×10^{-5}	0.6
34	Sulfathiazole	7.12	7.6×10^{-8}	0.08
35	2-S-4-Methylthiazole	7.79	1.6×10^{-8}	0.2
36	3-S-4-Methylfurazan	4.10	7.9×10^{-5}	1.0
37	3-S-5-Methyloxadiazole	4.40	4.0×10^{-5}	2.0
38	2-S-1,3,4-Thiadiazole	4.77	1.7×10^{-5}	0.6
39	2-S-5-Methylthiadiazole	5.45	3.5×10^{-6}	0.2
40	Sulfanilylcyanamide	2.92	1.2×10^{-3}	100
41	Sulfanilylurea	5.42	3.8×10^{-6}	10.0
42	Sulfanilylguanidine	...	...	10.0
43	Sulfanilylaminoguanidine	...	...	0.9
44	N^1-Acetylsulfanilamide	5.38	4.2×10^{-6}	0.7
45	N^1-Chloroacetylsulfanilamide	3.79	1.6×10^{-4}	10.0
46	N^1-Benzoylsulfanilamide	4.57	2.7×10^{-5}	0.3
47	N^1-*p*-Aminobenzoylsulfanilamide	5.20	6.3×10^{-6}	0.5
48	N^1-Ethylsulfonylsulfanilamide	3.10	7.9×10^{-4}	1000
49	N^1-Sulfanilylsulfanilamide	2.89	1.3×10^{-3}	60.0
50	4,4′-Diaminodiphenylsulfone	...	...	2.0

[a] S = Sulfanilamido.

$$\beta_{CX} = \eta_{CX}\beta_{CC} \qquad \text{(Eq. 2)}$$

where δ_X (heteroatom Coulomb integral coefficient) and η_{CX} (heteroatom bond integral coefficient) are the adjustable, semiempirical parameters of the HMO treatment. The δ and η values used in this study were similar to those which have been suggested by the Pullmans (24). A complete listing of these parameters is given in Table II.

For each molecule, the appropriate δ_X and η_{CX} values must be selected. The HMO matrix (A) must then be set up and solved for the eigenvalues and eigenvectors. The electronic charges may then be computed from the coefficients contained in the eigenvectors. Although a number of different procedures may be employed to manipulate such matrices, the most reasonable approach for large molecules as those studied here is through the use of a digital computer. To solve the matrices on a computer, a program is required which will determine the eigenvalues (x) and eigenvectors (v) of a real symmetric matrix (A) for an eigenvalue problem which has the form $Av = xv$ (26). Initial calculations were made using a Jacobi routine (27) and an IBM 1620 computer. However, this method was too slow, and the procedure was changed to a modified Givens routine (28) written for an IBM 7090. In this case a subroutine for the Gram-Schmidt orthogonalization procedure (29) was added to the main program in order to insure an orthonormal basis for the eigenvectors computed by the Givens method. A second subroutine was also added to the main program which computed the electronic charges directly according to Eq. 3, thereby eliminating most of the hand computations. In Eq. 3, q_J is the electronic charge density of atom J,

$$q_J = \sum_{i=1}^{n} e_i c_{iJ}^2 \qquad \text{(Eq. 3)}$$

and c_{iJ} is the LCAO coefficient of atom J in the i^{th} molecular orbital which is occupied by e_i electrons. The sum is taken over all molecular orbitals, $i = 1, 2, \ldots, n$, however, only the occupied MO's contribute to the charge, since for the unoccupied orbitals, $e_i = 0$. For the HMO matrix, A, the roots (x) of the characteristic equation of A, *i.e.*, $|A - xI| = 0$, were obtained in the form shown in Eq. 4, where E_i is the energy level of the i^{th} molecular orbital, α is the Coulomb integral, β is the resonance integral, and x_i is the molecular orbital energy coefficient of the i^{th} MO.

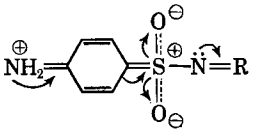
$$E_i = \alpha + x_i\beta \text{ or } x_i = \frac{\alpha - E_i}{\beta} \qquad \text{(Eq. 4)}$$

TABLE II—δ AND η VALUES

Atom[a]	δX	Bond	ηCX
$=\dot{N}-$	0.4	C=N	1.0
$-\ddot{N}-$	1.0	C—N	0.9
$\equiv \dot{N}$	1.5[b]	C≡N	2.0[b]
$=\dot{N}^+-$	2.0[c]	C=N$^+$	1.0[c]
NO_2 $-\ddot{N}^+$	0.6	C—N$^+$	1.0
$\cdots\dot{O}^-$	1.6	N$^+$ ··· O$^-$	1.0
$=\dot{O}$	1.2	C=O	2.0
$-\ddot{O}-$	2.0	C—O	0.9
$=\dot{S}$	0	C=S	1.2
$-\ddot{S}-$	0[d]	C—S	0.9[d]
$-\ddot{F}$	3.0[e]	C—F	0.7[e]
$-\ddot{Cl}$	1.4–2.0	C—Cl	0.4
$-\ddot{Br}$	1.5	C—Br	0.3
$\dot{C}_{arom.} - \dot{C}_{aliph.} = \dot{H}_2$			
$C_{arom.}$	−0.1		
$C_{aliph.}$	0	$C_{arom.}-C_{aliph.}$	0.7
$=\dot{H}_2$	−0.2[f]	C=H$_2$	2.0[f]
$\dot{C}_{arom.} - \dot{C}_{aliph.} = \dot{H}_2$			
$C_{arom.}$	−0.1		
$C_{aliph.}$	0	$C_{arom.}-C_{aliph.}$	0.7
$=\dot{H}_2$	−0.2[f]	C=H$_2$	2.0[f]
$\dot{C}_{arom.}=\dot{H}_2$			
$C_{arom.}$	0		
$=\dot{H}_2$	−0.2[f]	C=H$_2$	2.0
$\dot{C}_{arom.}=\dot{C}H-\dot{C}_{arom.}$			
$C_{arom.}$	0	$C_{arom.}$=CH	1.0
=CH	0	=CH—$C_{arom.}$	1.0
$-\dot{N}^+-\dot{C}-\dot{C}\equiv\dot{H}$			
$-\dot{N}^+-$	2.0	N$^+$—C	1.0
—C—	0.2	C—CH$_3$	1.0
CH_3 C	0	C≡H	2.0[f]
H	−0.2[f]		
$\dot{C}_{arom.}-\dot{C}H=\dot{C}H_2$			
$C_{arom.}$	0		
$-CH=$	0	$C_{arom.}-CH=$	1.0
$=CH_2$	0	$-CH=CH_2$	1.0
		$C_{aliph.}-C_{aliph.}$	0.7
		$C_{aliph.}-N^+-$	0.7
		$C_{aliph.}-N=$	0.7
		$=N-N-$	0.9
		$C_{arom.}$—COOH	0.7
		$C_{arom.}$—CONH$_2$	0.7

[a] The dots above the atoms refer to the number of π electrons initially assigned to the atom. [b] For $C_{arom.}-C_{aliph.}$ ≡N; $C_{arom.}$ has $\delta C = 0.15$, $C_{aliph.}$ has $\delta C = 0.30$, $C_{arom.}-C_{aliph.}$ has $\eta CC = 0.7$. [c] For C atoms adjacent to $=N^+-$, $\delta C = 0.3$. [d] For the most part, $\eta CS = 0.9$ has been used for a ring sulfur, *e.g.*, in thiazole or phenothiazine, but $\eta CS = 0.6$ has also been used in a few cases. [e] For $C_{arom.}-F$; $C_{arom.}$ has $\delta C = 0.2$. [f] Hyperconjugation.

The molecular form of the sulfonamide molecules was used as a model for the LCAO-MO calculations.[1] In order to consider the lone pair of electrons at the N^1 nitrogen atom as part of the mobile electron system, the resonance structure (II) was assumed.



II

The number of molecular orbitals and the number of π electrons considered for each molecule are given in Table III. The HMO matrix A for any particular compound listed in Table I may be set up by referring to Northey (17) for the structure of the R group, by allocating to the molecule the number of π electrons shown in Table III, and by using the δ and η values given in Table II. An example of the HMO matrix is given in Table IV for sulfapyridine (sulfa No. *15*). The numbering of the rows and columns in the matrix corresponds to the numbering of the atoms shown in structure III.[2] All other molecules were treated in the same manner as the sulfapyridine example, with exception of the δ and η values for the R groups. The δ and η values for the sulfanilamido part of the molecule in each case were the same as those given for the sulfapyridine example, that is, up to and including δ_{11} and η_{11-12}.

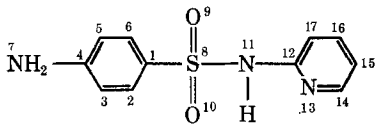
Results—The results of the LCAO-MO calculations on 30 of the sulfonamides studied are given in Table V. The tabulation includes the electronic charges at the amide nitrogen (q_{N1}), the *p*-amino nitrogen (q_{N4}), the SO_2 group oxygen atoms (q_{oxy}), and the sulfur atom (q_S). The formal π charges at these atoms may be computed by subtracting the tabulated electronic charges (the q's) from 2, 2, 1, and 2, respectively, according to Eq. 5, where f_J denotes the net or formal charge on atom J, atom J contributing e_J π electrons to the π electron pool.

[1] Calculations on the ionic forms did not differ appreciably from the molecular forms, and the authors were not able to improve the correlations by accounting for the ionic species in the calculations. Also, the omega technique (5, 30) was used for another series of compounds; however, it did not change the results significantly. The authors have found more success in accounting for ionic forms by moderately adjusting the δx parameter rather than by using the iterative process of the omega method.

[2] The *d*-orbitals of sulfur have been neglected. There is some controversy in the literature about the contribution of these orbitals (34). However, an article by Cammarata [*J. Pharm. Sci.*, **55**, 1469(1966)] suggests that if the effects of the *d*-orbital overlap are taken into account (using $\delta x = -2.60$ and $\beta CX = 0.99$ for the entire SO_2 group), small variations in the electronic charge at the *p*-amino position of the sulfonamides could be noted.

$$f_J = (e_J - q_J) \qquad \text{(Eq. 5)}$$

The results of the MO calculations on the sulfanilamide type compounds generally agree with chemical facts. In terms of the formal charge for sulfanilamide, for example (sulfa No. *2*), the electronic configuration shown in structure IV is found. The results indicate: (*a*) a deficiency (+) of π electrons at the *p*-amino group due to the fact that this group has donated its electrons to the adjacent benzene ring; (*b*) that due to their high electronegativity, the sulfonyl oxygen atoms have withdrawn electrons from the sulfur atom and also from the N^1 nitrogen atom, resulting in an excess (−) of electrons at these atoms; and (*c*) that the N^1 nitrogen atom is deficient (+) in π charge with



III

Table III—Data Used for LCAO-MO Calculations on Sulfonamide Derivatives

Sulfa	No. of MO's	No. of π Electrons
1 (PABA)	10	12
2	11	14
3	13	16
4	15	18
5	16	20
6	16	20
7	17	20
8	19	22
9	19	22
10	19	22
11	21	26
12	21	26
13	18	22
14	18	22
15	17	20
16	17	20
17	18	22
18	18	22
19	18	22
20	18	22
21	16	20
22	17	20
23	17	20
24	19	22
25	21	24
26	18	22
27	17	20
28	17	20
29	18	22
30	17	20

Table V—Electronic Charges for Sulfonamide Derivatives

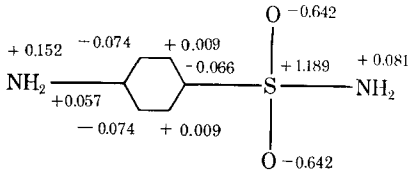
Sulfa	Electronic Charges			
	q_{N^1}	q_{N^4}	q_{oxy}	q_S
1 (PABA)	...	...	...	...
2	1.919	1.848	1.642	0.811
3	1.853	1.848	1.640	0.811
4	1.795	1.848	1.638	0.812
5	1.851	1.848	1.642	0.811
6	1.855	1.848	1.640	0.811
7	1.794	1.848	1.637	0.810
8	1.791	1.848	1.637	0.809
9	1.792	1.848	1.637	0.811
10	1.793	1.848	1.638	0.810
11	1.792	1.848	1.637	0.811
12	1.790	1.848	1.637	0.811
13	1.805	1.848	1.638	0.811
14	1.856	1.848	1.640	0.811
15	1.781	1.848	1.636	0.810
16	1.793	1.848	1.637	0.810
17	1.781	1.848	1.637	0.810
18	1.794	1.848	1.638	0.811
19	1.793	1.848	1.637	0.811
20	1.805	1.848	1.638	0.811
21	1.790	1.848	1.637	0.811
22	1.780	1.848	1.637	0.810
23	1.770	1.848	1.636	0.810
24	1.770	1.848	1.635	0.808
25	1.769	1.848	1.638	0.810
26	1.769	1.848	1.637	0.810
27	1.768	1.848	1.636	0.809
28	1.793	1.848	1.637	0.811
29	1.797	1.848	1.636	0.810
30	1.779	1.848	1.636	0.810

Table IV—HMO Matrix for Sulfapyridine[a]

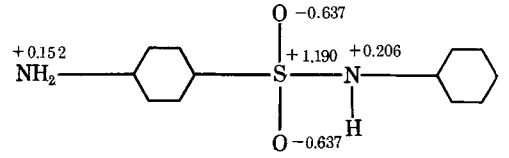
$A =$

Atom ←No.	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	↓
	0	1.0	0	0	0	1.0	0	0.6	0	0	0	0	0	0	0	0	0	1
	1.0	0	1.0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2
	0	1.0	0	1.0	0	0	0	0	0	0	0	0	0	0	0	0	0	3
	0	0	1.0	0	1.0	0	0.9	0	0	0	0	0	0	0	0	0	0	4
	0	0	0	1.0	0	1.0	0	0	0	0	0	0	0	0	0	0	0	5
	1.0	0	0	0	1.0	0	0	0	0	0	0	0	0	0	0	0	0	6
	0	0	0	0.9	0	0	1.0	0	0	0	0	0	0	0	0	0	0	7
	0.6	0	0	0	0	0	0	0	2.0	2.0	0.9	0	0	0	0	0	0	8
	0	0	0	0	0	0	0	2.0	1.2	0	0	0	0	0	0	0	0	9
	0	0	0	0	0	0	0	2.0	0	1.2	0	0	0	0	0	0	0	10
	0	0	0	0	0	0	0	0.9	0	0	1.0	0.9	0	0	0	0	0	11
	0	0	0	0	0	0	0	0	0	0	0.9	0	1.0	0	0	0	1.0	12
	0	0	0	0	0	0	0	0	0	0	0	1.0	0.4	1.0	0	0	0	13
	0	0	0	0	0	0	0	0	0	0	0	0	1.0	0	1.0	0	0	14
	0	0	0	0	0	0	0	0	0	0	0	0	0	1.0	0	1.0	0	15
	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1.0	0	1.0	16
	0	0	0	0	0	0	0	0	0	0	0	1.0	0	0	0	1.0	0	17

[a] Numbering of rows and columns corresponds to the numbering of structure III.



IV



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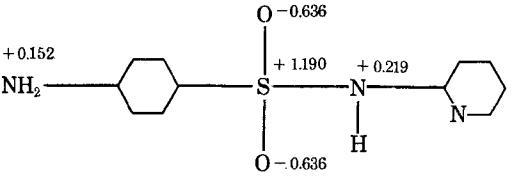

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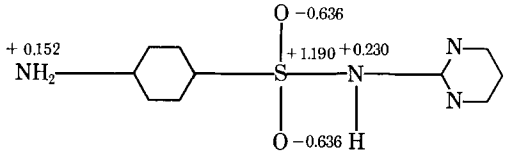
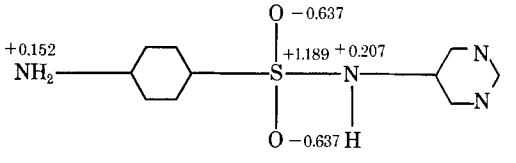


VII



VIII

respect to its original contribution of 2π electrons to the π electron pool.

Now looking at the derivative whose R group is a phenyl radical, N^1-phenylsulfanilamide (sulfa No. 7) (V), the results change accordingly. In this case, the electronegative phenyl group as well as the SO_2 group oxygen atoms withdraw electrons from the sulfanilamide system. A much greater deficiency of π charge results at the N^1 nitrogen atom. The phenyl group has also reduced the charge on the oxygen atoms, further indicating its effect as an electron acceptor.

An even greater deficiency of π charge results at the N^1 nitrogen atom where the R group is pyridine with the pyridine nitrogen at the *ortho* position in sulfapyridine (sulfa No. *15*) (VI). The greater electronegativity of this R group is thereby accounted for. A still greater change occurs for sulfadiazine (sulfa No. *23*) (VII). The excess π charge at the oxygen atoms reacts accordingly when these atoms as well as the electronegative R group compete for the available delocalized electrons. For the compound 5-sulfanilamidopyrimidine (sulfa No. *28*) (VIII), the electron attracting effect of the R group is less than that for sulfadiazine, apparently because the electronegative nitrogen atoms in this substituent group are further removed from the point of attachment at the N^1 nitrogen.

Table V also shows that for all of the sulfonamide derivatives studied, the electronic charge at the N^4 nitrogen atom is constant, $q_{N^4} = 1.848$ (formal π charge $= +0.152$). Although this group seems to be necessary for biological activity, it appears that no correlation exists between the degree of activity and the electronic charge density calculated by the HMO method for this position of the compounds studied. This seems to agree with Bell and Roblin's finding of constant basic dissociation; however, it is possible that differences in the charges at the N^4 nitrogen atom of various sulfonamide derivatives would become evident if a more refined method of calculation were established.[2]

CORRELATION AND DISCUSSION OF RESULTS

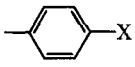
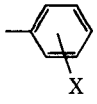
A plot of pKa *versus* the electronic charge at the ionizing N^1 nitrogen atom (q_{N^1}) for the 50 sulfanilamide type compounds indicated considerable scatter. The correlation expected in this case would be a linear relationship between pKa and q_{N^1}. The scatter was due, at least in part, to the effect of solvation energy which was not included in the calculations. More important, however, is the fact that the R groups of the 50 sulfonamide derivatives are of widely varying types, such as alternate rings, nonalternate rings, rings with and without substituent groups, and straight chains with and without heteroatoms. The correlation of pKa and electronic charge depends upon whether certain partition functions cancel (3, 19, 20). If this is not the case, the relationship may be much less direct, especially in the case of changing molecular skeletons. Also, the Hückel approximation does not provide equally good results in all of these cases. It is known, for instance, that the simple Hückel treatment gives a better account of alternate than nonalternate hydrocarbons because in the latter case the presence of a nonself-consistent field really requires that the Coulomb integral of each atom be corrected for the charges on neighboring atoms (3, 4). Also, the treatment of straight chains requiring the use of hyperconjugation is often less reliable. The R groups of those compounds which were poorly accounted for by the simple Hückel method were all of the nonalternate or straight chain types (types *D*, *E*, and *F* as defined below), and hence, less reliable results were to be expected in these cases.

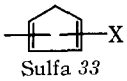
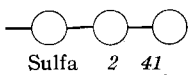
In order to examine the relationship between pKa and electronic charge in more detail, the 50 sulfonamide derivatives were classified into groups dependent on the type of substituent R group attached to the N^1 nitrogen atom. The classes are given in Table VI. Following this classification, individual plots were constructed for each type of R group. The results are shown in Figs. 1–3, for the *A*, *B*, and *C* type substituents. Accordingly, the categorization by the type of R group seems to be an important factor in explaining the correlation of

the pKa–electronic charge relationship. The results illustrate that if one anticipates using the electronic charge at the ionizing atom of sulfanilamide type compounds (or similarly complex drug molecules) for theoretically predicting pKa, it will probably be necessary to employ a more exact treatment in the cases of nonalternate R groups and hyperconjugation than the simple Hückel approximation. Procedures for the treatment of hyperconjugation and nonalternate molecules are available (5, 30, 31); however, the problems involved become increasingly difficult with the number and types of heteroatoms.

A knowledge of the electronic characteristics of a molecule may also allow prediction of drug activity. This would be possible even for compounds which have never been synthesized, once a basic relationship has been established by pharmacological testing of a representative sample of compounds. Table VII shows the relationship between sulfonamide activity and the charge $q_N{}^1$. The trend exhibited by the data indicates that, in general, a decrease in bacteriostatic activity may be associated with a decrease in the formal π charge at

TABLE VI—CLASSIFICATION OF SULFONAMIDE DERIVATIVES ACCORDING TO TYPE OF R GROUP

Type A[a]	Type B	Type C
		
Sulfa *7*	Sulfa *10*	Sulfa *8*
15	*12*	*9*
16	*13*	*11*
22	*17*	*24*
23	*18*	*25*
27	*19*	*26*
28	*20*	
30	*29*	

Type D	Type E	Type F
		
Sulfa *14*	Sulfa *33*	Sulfa *2* *41*
21	*35*	*3* *42*[b]
31	*36*	*4*[b] *43*[b]
32	*37*	*5* *44*
34	*39*	*6* *45*
38		*40* *48*

Compd. Not Classified
Sulfa *1* (PABA)
46[c]
47[c]
49
50[b]

[a] The ring may contain heteroatoms. This note applies to B, C, D, and E also. [b] No pKa available. [c] These compounds may not be permitted to form the theorized resonance structure, and therefore, may require a slightly different MO treatment.

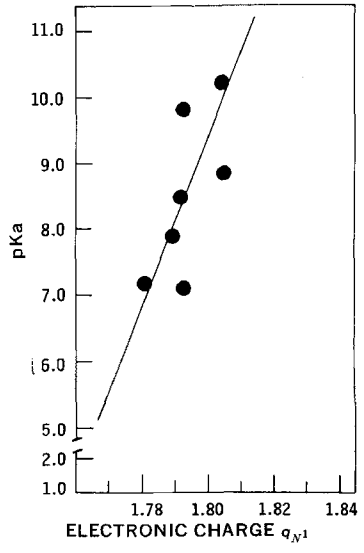


Fig. 2—pKa-electronic charge relationship for the type B sulfonamide derivatives.

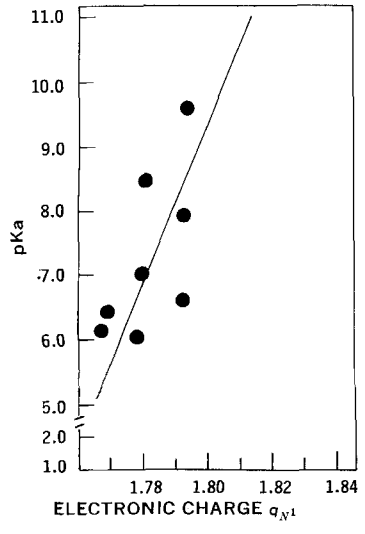


Fig. 1—pKa-electronic charge relationship for the type A sulfonamide derivatives.

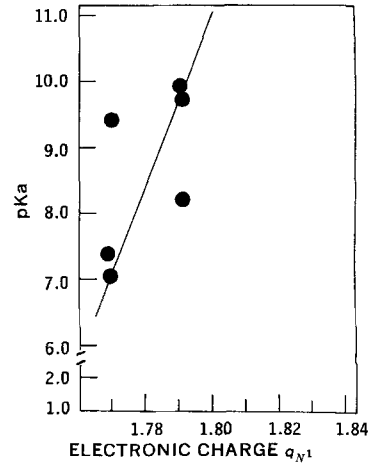


Fig. 3—pKa-electronic charge relationship for the type C sulfonamide derivatives.

TABLE VII—RELATIONSHIP BETWEEN BIOLOGICAL ACTIVITY AND ELECTRONIC CHARGE

Sulfa	Activity Index ($C_R \times 10^5)^{-1}$	Decreasing Activity	Formal π Charge $f_N{}^1$	Decreasing Charge
23	12.5		(+) 0.230	
30	12.5		0.221	
22	12.5		0.220	
27	20.0		0.232	
29	10.0		0.203	
16	5.0		0.207	
28	5.0		0.207	
18	5.0		0.206	
17	2.0		0.219	
12	2.0		0.210	
15	1.67		0.219	
19	1.67		0.207	
11	0.50		0.208	
20	0.50		0.195	
7	0.33		0.206	
9	0.20		0.208	
10	0.20		0.207	
13	0.20		0.195	

N^1 (or an increase in $q_N{}^1$). A better correlation of these parameters is probably dependent upon a more complete calculation procedure, including the use of partition coefficients as substituent constants (32, 33) in order to account for the polar characteristics of the molecules. Further work is being performed to correlate the electronic characteristics of the sulfonamide molecules with biological activity.

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