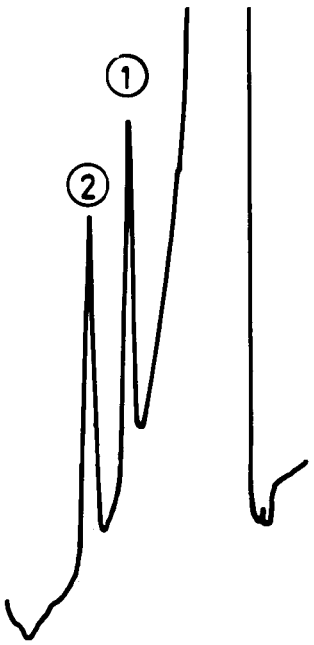


Figure 1—*Separation of the ditazole derivative (2) from the internal standard, diazepam (1).*

Ditazole also was found in high concentration in the liver 5 min after treatment; ditazole levels in the liver decreased with a slope similar to that seen in the brain. The heart levels, with a maximum at 5 min, decreased rapidly so that only small amounts were detected 1 hr after treatment.

Small amounts of ditazole were found in the adipose tissue, with a maximum of 5 μg at 30 min after administration. No measurable amounts were present after 1 hr.

The pharmacokinetic parameters of ditazole in rat blood following a 20-mg/kg iv administration were calculated using the NONLIN program (8) on a digital computer[5]. The apparent ditazole half-life in the rat blood was 41 min, its distribution volume was about 2 liters/kg, and its clearance was about 0.03 liter/kg/min. Areas under the blood ditazole concentration–time curves at time 0 and ∞ were 490 and 578 μg/ml × min, respectively.

[5] UNIVAC 1106.

Table I—Ditazole Levels[a] in the Blood, Brain, Liver, and Adipose Tissue of Rats after Administration of 20 mg/kg iv

Minutes after Ditazole Administration	Blood, μg/ml ± *SE*	Brain, μg/g ± *SE*	Heart, μg/g ± *SE*	Liver, μg/g ± *SE*	Adipose Tissue, μg/g ± *SE*
5	8.7 ± 1.0	79.7 ± 4.0	18 ± 0.2	29.6 ± 1.0	3.4 ± 0.1
30	4.9 ± 0.7	15.4 ± 1.0	10.2 ± 0.5	24.7 ± 0.7	5.0 ± 0.5
60	3.6 ± 0.7	7.6 ± 0.3	1.7 ± 0.2	7.0 ± 0.7	1.1 ± 0.1
120	1.5 ± 0.4	2.6 ± 0.1	<0.4	2.5 ± 0.3	<0.4
240	<0.4	<0.4	<0.4	<0.4	<0.4

[a] Each figure is the average of at least four determinations.

Studies on ditazole metabolism in rats after intravenous administration are lacking. Marchetti *et al.* (5) reported that ditazole elimination occurred slowly through urine and feces after oral administration; most of the drug was excreted unchanged. Among its metabolites, 4,5-diphenyl-4-oxazolin-2-one, 4,5-diphenyl-2-(2-oxyethyl)aminoxazole, and benzil were identified. Preliminary *in vitro* studies on ditazole metabolism by rat liver microsomal enzymes indicate that this drug is metabolized into two or three compounds not yet identified.

It is well known that drugs and other foreign compounds combine with hepatic cytochrome P-450 to produce difference spectra of two general types, I and II (9). With hepatic cytochrome P-450, ditazole gave a type II spectrum with λ_{max} and λ_{min} falling within the usual range.

REFERENCES

(1) E. Marchetti, G. Mattalia, and V. Rosnati, *J. Med. Chem.*, **11**, 1092 (1968).

(2) L. Caprino, F. Borrelli, and R. Falchetti, *Arzneim.-Forsch.*, **23**, 1972 (1973).

(3) *Ibid.*, **23**, 1277 (1973).

(4) G. de Gaetano, M. C. Tonolli, M. P. Bertoni, and M. C. Roncaglioni, *Haemostasis*, **6**, 127 (1977).

(5) E. Marchetti, G. Mattalia, and G. Bergesi, *Arzneim.-Forsch.*, **23**, 1291 (1973).

(6) T. Omura and R. Sato, *J. Biol. Chem.*, **239**, 2370 (1964).

(7) *Ibid.*, **239**, 2379 (1964).

(8) C. M. Metzler, "A User's Manual for NONLIN," Tech. Rep. 7292/69/7292/005, The Upjohn Co., Kalamazoo, Mich., Nov. 25, 1969.

(9) B. N. La Du, M. C. Mandel, and E. L. Way, "Fundamentals of Drug Metabolism and Drug Disposition," Williams & Wilkins, Baltimore, Md., 1971, p. 206.

Electronic Structure–Activity Relationships of Antibacterial Acridines

P. SINGH and S. P. GUPTA ˣ

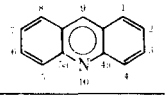
Received June 9, 1977, from the *Department of Chemistry, Birla Institute of Technology and Science, Pilani 333031, India.* Accepted for publication August 3, 1977.

Abstract □ The antibacterial activity of a series of amino- and fluorinated acridines was studied in the framework of their electronic structures. To calculate the electronic structure, a simple Hückel molecular orbital theory was used. A statistical regression analysis revealed linear correlations between the activity and the electronic indexes, particularly the electron density at the ring nitrogen.

Keyphrases □ Acridines, various—antibacterial activity related to electronic structure □ Antibacterial activity—various acridines, related to electronic structure □ Electronic structure—various acridines, related to antibacterial activity □ Structure–activity relationships—various acridines, antibacterial activity related to electronic structure

The antibacterial activity of acridines has been found to be proportional to the fraction ionized as the cation (1–3). The simplest interpretation of the mode of action of acridine cations is that they compete with hydrogen ions for a vitally important anionic group on the bacterium (4). The vital activity of the vulnerable anionic group (A^-) of the bacterium is supposed to be reduced by the formation of a feebly dissociated complex (ABH) with the cation

Table I—Electronic Parameters for Regression Analysis and Observed and Calculated Antibacterial Activity of Aminoacridines



Substituent	q_N	q_{4a}	q_{5a}	log 1/C Obs. (3)	log 1/C Calc. (Eq. 7)	log 1/C Calc. (Eq. 4)	$\Delta E_{\pi b}$	log 1/C Calc. (Eq. 10)
4-Amino	1.234	0.966	0.961	3.699	3.884	4.012	2.537	3.968
2-Amino	1.239	0.964	0.961	4.000	4.047	4.119	2.543	4.089
3-Amino	1.256	0.957	0.958	4.903	4.490	4.485	2.562	4.475
9-Amino	1.294	0.959	0.959	5.204	5.216	5.301	2.604	5.328
4,5-Diamino	1.224	0.970	0.970	<3.699	3.913	3.797	2.531	3.846
2,7-Diamino	1.236	0.968	0.968	4.301	4.135	4.055	2.542	4.069
3,7-Diamino	1.253	0.961	0.965	5.204	4.578	4.420	2.560	4.435
3,6-Diamino	1.270	0.958	0.958	5.204	4.734	4.785	2.576	4.759
3,9-Diamino	1.305	0.960	0.960	5.204	5.441	5.537	2.615	5.551
4-Amino-5-methyl	1.232	0.967	0.963	<3.699	3.891	3.969	2.536	3.947
2-Amino-9-methyl	1.248	0.965	0.962	4.301	4.232	4.313	2.555	4.333
9-Amino-2-methyl	1.293	0.960	0.960	5.204	5.204	5.279	2.603	5.308
9-Amino-3-methyl	1.296	0.959	0.959	5.204	5.256	5.344	2.606	5.368
9-Amino-4-methyl	1.291	0.948	0.958	5.505	5.471	5.236	2.600	5.247
9-Amino-3-chloro	1.289	0.960	0.959	5.204	5.085	5.193	2.599	5.226
9-Amino-2-chloro	1.295	0.954	0.958	5.204	5.357	5.322	2.602	5.287
9-Amino-4-chloro	1.297	0.948	0.958	5.204	5.589	5.365	2.602	5.287
2-Amino-6-chloro	1.234	0.957	0.958	<3.699	4.055	4.012	2.538	3.988
3-Amino-9-chloro	1.234	0.964	0.963	<3.699	4.027	4.012	2.541	4.049
3-Amino-6-chloro	1.251	0.959	0.958	4.602	4.327	4.377	2.557	4.374

(BH^+) of the drug. The more feebly the complex is dissociated into A^- and BH^+ ions, the greater is the reduction in the vital activity of the anion. The dissociation of the complex is suppressed only when the cation (BH^+) is present in excess. Therefore, the activity of the drug (B) depends on its degree of cationization as BH^+.

BACKGROUND

Since the cationization of the acridines largely depends on the electron density of their ring nitrogen, q_N (the greater the q_N, the more the attraction for protons), it is expected that a correlation must exist between their antibacterial activity and q_N. The electron density is calculated by using a simple Hückel molecular orbital (HMO) method.

In the HMO method, the coulomb integral for a heteroatom x, α_x, is defined as:

$$\alpha_x = \alpha_0 + h_x\beta_0 \qquad \text{(Eq. 1)}$$

and the resonance integral for a bonded pair of carbon and heteroatom, β_{cx}, is defined as:

$$\beta_{cx} = k_{cx}\beta_0 \qquad \text{(Eq. 2)}$$

where α_0 and β_0 are the standard coulomb and resonance integrals for the carbon atom and the carbon–carbon bond, respectively; and h_x and k_{cx} are the semiempirical parameters under discussion. The values of these parameters for the present purpose have been taken from the literature (5).

If some heteroatoms were adjacent to a carbon atom, the coulomb integral for that carbon atom was taken as:

$$\alpha_c = \alpha_0 + 0.1 \sum_x^{adj} h_x\beta_0 \qquad \text{(Eq. 3)}$$

Equation 3 takes into account the inductive effect produced on the carbon atom by heteroatoms (6). The methyl group was treated as a hyperconjugation model.

The molecular orbital techniques have been successfully utilized in drug research (7). Like several physicochemical parameters, such as hydrophobicity, partition coefficients, and polarizability (8–13), many molecular orbital indexes (*e.g.*, charge density, free valence, delocalization energy, and energies of the highest occupied and lowest unoccupied molecular orbitals) have been found (7, 14–17) to be well correlated with biological responses of organic compounds. Recently, a new molecular parameter, known as molecular connectivity, was shown (18–23) to possess significant correlations with many physical properties and biological activities of compounds. The molecular connectivity index, χ (24), signifies the degree of branching or connectivity in a molecule.

RESULTS AND DISCUSSION

Table I gives the calculated electron densities at the ring nitrogen and the two neighboring carbon atoms (4a and 5a) of the bridges as well as the antibacterial activity for a series of aminoacridines. Similarly, Table II gives the same values for a series of fluorinated acridines. In Table I, log 1/C [C is the minimal bacteriostatic concentration (3) for *Streptococcus pyogenes* after 48 hr of incubation in 10% serum broth at 37° and pH 7.3], as expected, appears to be linearly correlated with q_N. Likewise, the mean K.D. time[1], t, in Table II appears to be proportional to q_N. A regression analysis (25) reveals the following three equations relating biological responses with q_N:

$$\log 1/C = 21.48q_N - 22.478$$
$$n = 20 \quad r = 0.896 \quad s = 0.309 \quad F^1_{18} = 73.30 \qquad \text{(Eq. 4)}$$

$$t_1 = 142.8q_N - 168.23$$
$$n = 33 \quad r = 0.605 \quad s = 1.419 \quad F^1_{31} = 17.91 \qquad \text{(Eq. 5)}$$

$$t_2 = 158.0q_N - 185.64$$
$$n = 33 \quad r = 0.551 \quad s = 1.806 \quad F^1_{31} = 13.55 \qquad \text{(Eq. 6)}$$

The statistical parameters, r, s, and F (F ratio between the variances of the calculated and observed activities) show that correlation between q_N and log 1/C is highly significant. In Eq. 4, F is highly significant at the 99% level [F^1_{18} (0.01) = 8.28]. In Eqs. 5 and 6, F also is significant at the 99% level [F^1_{31} (0.01) = 7.53] but not as highly as in Eq. 4. In the latter two equations, the correlation coefficient is also comparatively low (the correlation between log t and q_N was still less significant). The calculated values of activity from these equations are listed in the respective tables. There is good agreement between the calculated and observed values.

With acridines, q_N appears to be most important. The inclusion of q_{4a} and q_{5a}, the electron densities at the nearest neighbors of nitrogen that might affect the degree of cationization, in the regression analysis makes no significant improvement in the correlation in any case (compare the statistical parameters of Eqs. 7–9 with those of Eqs. 4–6, respectively, and the results obtained with them):

$$\log 1/C = 19.77q_N - 32.10q_{4a} + 39.42q_{5a} - 27.34$$
$$n = 20 \quad r = 0.910 \quad s = 0.305 \quad F^3_{16} = 24.34 \qquad \text{(Eq. 7)}$$

$$t_1 = 147.5q_N - 29.96q_{4a} + 14.25q_{5a} - 158.72$$
$$n = 33 \quad r = 0.638 \quad s = 1.42 \quad F^3_{29} = 6.11 \qquad \text{(Eq. 8)}$$

$$t_2 = 159.9q_N - 32.20q_{4a} + 9.66q_{5a} - 166.10$$
$$n = 33 \quad r = 0.585 \quad s = 1.82 \quad F^3_{29} = 4.69 \qquad \text{(Eq. 9)}$$

With aminoacridines, one more independent electronic index, delocalization energy, can be correlated with the activity. Since the amino-

[1] K.D. time stands for knockdown time of cockroaches after injecting the compound.

Table II—Electronic Parameters for Regression Analysis and Observed and Calculated Antibacterial Activity of Fluorinated Acridines

Substituent	q_N	q_{4a}	q_{5a}	Mean K.D. Time, t_1 (hr), 0.5% Concentration			Mean K.D. Time, t_2 (hr), 0.1% Concentration		
				Obs. (26)[a]	Calc. (Eq. 5)	Calc. (Eq. 8)	Obs. (26)	Calc. (Eq. 6)	Calc. (Eq. 9)
5-Chloro-3-fluoro	1.240	0.957	0.946	11.00	8.75	8.84	13.00	10.13	10.28
5-Chloro-8-fluoro	1.242	0.956	0.946	10.50	9.03	9.17	12.00	10.45	10.63
5-Chloro-6-fluoro	1.241	0.955	0.947	10.00	8.89	9.06	12.00	10.29	10.51
5-Chloro-3,6-difluoro	1.234	0.956	0.947	9.00	7.89	8.00	10.00	9.18	9.36
5-Chloro-3,8-difluoro	1.236	0.957	0.946	10.00	8.17	8.25	12.50	9.50	9.64
5-Amino-3-fluoro	1.227	0.963	0.966	9.50	6.89	7.03	10.75	8.08	8.20
5-Amino-8-fluoro	1.228	0.962	0.966	9.00	7.03	7.20	10.50	8.24	8.39
5-Amino-6-fluoro	1.227	0.961	0.966	8.50	6.89	7.09	9.50	8.08	8.26
5-Amino-3,6-difluoro	1.220	0.963	0.966	7.00	5.89	6.00	9.00	6.97	7.08
5-Amino-3,8-difluoro	1.222	0.963	0.965	7.50	6.18	6.28	9.50	7.29	7.39
3-Fluoro-5-phenoxy	1.232	0.960	0.958	8.00	7.60	7.74	9.00	8.87	9.02
8-Fluoro-5-phenoxy	1.234	0.960	0.958	8.00	7.89	8.04	9.50	9.18	9.34
6-Fluoro-5-phenoxy	1.232	0.959	0.959	7.00	7.60	7.79	8.00	8.87	9.06
3,6-Difluoro-5-phenoxy	1.226	0.959	0.958	7.00	6.75	6.89	8.50	7.92	8.09
3,8-Difluoro-5-phenoxy	1.227	0.961	0.958	6.50	6.89	6.97	8.00	8.08	8.19
3-Fluoro-5-*p*-fluorophenoxy	1.232	0.960	0.958	7.00	7.60	7.74	8.50	8.87	9.02
8-Fluoro-5-*p*-fluorophenoxy	1.230	1.050	1.031	6.50	7.32	5.79	7.50	8.55	6.51
6-Fluoro-5-*p*-fluorophenoxy	1.232	0.959	0.959	6.00	7.60	7.79	6.50	8.87	9.06
3,6-Difluoro-5-*p*-fluorophenoxy	1.226	0.960	0.959	6.50	6.75	6.87	8.50	7.92	8.07
3-Fluoro-5-phenylamino	1.228	0.963	0.963	5.75	7.03	7.13	6.50	8.24	8.33
8-Fluoro-5-phenylamino	1.229	0.962	0.964	5.50	7.17	7.32	6.50	8.39	8.53
6-Fluoro-5-phenylamino	1.227	0.975	0.953	5.50	6.89	6.48	7.00	8.08	7.69
3,6-Difluoro-5-phenylamino	1.222	0.977	0.952	5.50	6.18	5.67	5.75	7.29	6.81
3,8-Difluoro-5-phenylamino	1.223	0.963	0.963	6.00	6.32	6.40	6.25	7.45	7.53
3-Fluoro-5-*p*-fluorophenylamino	1.228	0.963	0.963	5.00	7.03	7.13	5.00	8.24	8.33
8-Fluoro-5-*p*-fluorophenylamino	1.229	0.962	0.963	4.50	7.17	7.31	5.50	8.39	8.52
6-Fluoro-5-*p*-fluorophenylamino	1.228	0.961	0.964	5.00	7.03	7.21	6.00	8.24	8.41
3,6-Difluoro-5-*p*-fluorophenylamino	1.221	0.963	0.964	5.50	6.03	6.11	5.75	7.13	7.22
3,8-Difluoro-5-*p*-fluorophenylamino	1.204	0.963	0.961	5.00	3.60	3.56	6.00	4.41	4.47
3-Fluoro-5-α-naphthylamino	1.228	0.963	0.963	8.00	7.03	7.13	9.00	8.24	8.33
8-Fluoro-5-α-naphthylamino	1.229	0.962	0.963	7.00	7.17	7.31	8.50	8.39	8.52
6-Fluoro-5-α-naphthylamino	1.245	0.976	0.943	8.00	9.46	8.97	8.50	10.92	10.44
3,6-Difluoro-5-α-naphthylamino	1.228	0.975	0.943	6.00	7.03	6.49	8.00	8.24	7.75

[a] The activity is against male and female cockroaches.

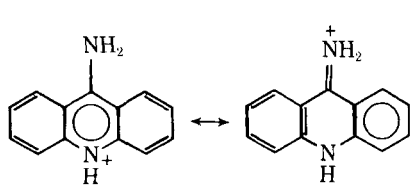
acridine cations exhibit the resonance phenomenon as shown in Scheme I, the degree of cationizations of aminoacridines depends (27) on the extra stabilization the cations gain by this resonance (delocalization). Hence, the difference in the delocalization energies of the ionized and unionized forms of aminoacridines, ΔDE, should be proportional to their activity. For the aminoacridines listed in Table I, ΔDE can be replaced by $\Delta E_{\pi b}$, the difference in the π-bond energies of their ionized and unionized forms, because all of these compounds have the same number of delocalized bonds of one kind in either form. A regression analysis reveals the following equation correlating log $1/C$ with $\Delta E_{\pi b}$:

$$\log 1/C = 20.52\ \Delta E_{\pi b} - 48.08$$
$$n = 20 \quad r = 0.897 \quad s = 0.307 \quad F_{18}^{1} = 74.42 \qquad \text{(Eq. 10)}$$

The statistical parameters of Eq. 10 are almost equal to those of Eq. 4, and the results obtained by the two equations nearly tally with each other. The reason is that a nearly perfect correlation exists between q_N and $\Delta E_{\pi b}$ (in Eq. 11, $r \cong 1$ and $s \cong 0.0$):

$$\Delta E_{\pi b} = 1.057 q_N + 1.235$$
$$n = 20 \quad r = 0.998 \quad s = 0.001 \quad F_{18}^{1} = 6299 \qquad \text{(Eq. 11)}$$

This result also shows that, from the antibacterial activity point of view, q_N is more important for acridines than the charge at any other atoms.



Scheme I

This electronic index can be successfully utilized to predict the antibacterial activity of any acridine before its synthesis.

REFERENCES

(1) A. Albert, S. Rubbo, and R. Goldacre, *Nature (London)*, **147**, 332 (1941).

(2) A. Albert, S. Rubbo, R. Goldacre, M. Davey, and J. Stone, *Br. J. Exp. Pathol.*, **26**, 160 (1945).

(3) A. Albert, in "Drug Design," vol. III, E. J. Ariëns, Ed., Academic, New York, N.Y., 1972.

(4) A. Albert "Selective Toxicity," Chapman and Hall, London, England, 1973.

(5) A. Streitwieser, Jr., "Molecular Orbital Theory for Organic Chemists," Wiley, New York, N.Y., 1961.

(6) K. Higasi, H. Baba, and A. Rembaum, "Quantum Organic Chemistry," Interscience, New York, N.Y., 1965, p. 148.

(7) L. B. Kier, "Molecular Orbital Theory in Drug Research," Academic, New York, N.Y., 1971.

(8) M. S. Tute, *Adv. Drug Res.*, **6**, 1 (1971).

(9) C. Hansch, in "Drug Design," vol. I, E. J. Ariëns, Ed., Academic, New York, N.Y., 1971, p. 271.

(10) L. B. Kier, *J. Pharm. Sci.*, **61**, 1394 (1972).

(11) H. D. Holtje and L. B. Kier, *J. Med. Chem.*, **17**, 814 (1974).

(12) H. D. Holtje and L. B. Kier, *J. Pharm. Sci.*, **63**, 1435 (1974).

(13) H. D. Holtje and L. B. Kier, *J. Theoret. Biol.*, **48**, 197 (1974).

(14) F. Peradejordi, A. N. Martin, and A. Cammarata, *J. Pharm. Sci.*, **60**, 577 (1971).

(15) T. K. Lin, Y. W. Chien, H. B. Desai, and P. K. Yonan, *Chem. Pharm. Bull.*, **24**, 2739 (1976).

(16) E. Mizuta, K. Nishikawa, K. Omura, and Y. Oka, *ibid.*, **24**, 2078 (1976).

(17) S. P. Gupta, S. S. Sharma, and P. Singh, *Indian J. Chem.*, **15B**, 731 (1977).
(18) L. B. Kier, L. H. Hall, W. J. Murray, and M. Randić, *J. Pharm. Sci.*, **64**, 1971 (1975).
(19) L. H. Hall, L. B. Kier, and W. J. Murray, *ibid.*, **64**, 1974 (1975).
(20) W. J. Murray, L. H. Hall, and L. B. Kier, *ibid.*, **64**, 1978 (1975).
(21) L. B. Kier, W. J. Murray, and L. H. Hall, *J. Med. Chem.*, **18**, 1272 (1975).
(22) L. B. Kier, W. J. Murray, M. Randić, and L. H. Hall, *J. Pharm. Sci.*, **65**, 1226 (1976).
(23) W. J. Murray, L. B. Kier, and L. H. Hall, *J. Med. Chem.*, **19**, 573 (1976).
(24) M. Randić, *J. Am. Chem. Soc.*, **97**, 6609 (1975).
(25) R. A. Fisher, "Statistical Methods for Research Workers," Oliver and Boyd, Edinburg, Scotland, 1970.
(26) K. C. Joshi and M. K. Tholia, *Agr. Biol. Chem.*, **38**, 1165 (1974).
(27) A. Pullman and B. Pullman, "Quantum Biochemistry," Interscience, New York, N.Y., 1963.

ACKNOWLEDGMENTS

The authors thank CSIR, New Delhi, India, for providing a Senior Research Fellowship to P. Singh.

Synthesis and Antitumor Evaluation of 4-Ethoxycarbonyl Cyclophosphamide Analogs

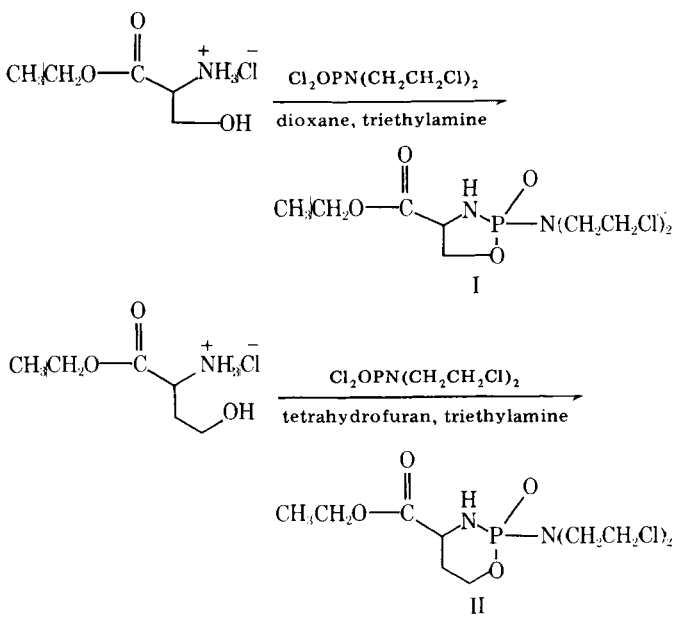
EMERSON L. FOSTER

Received January 7, 1977, from the *Veterans Administration Hospital, Indianapolis, IN 46202.* Accepted for publication March 31, 1977.

Abstract □ 4-Ethoxycarbonyl analogs of cyclophosphamide and its five-membered ring homolog were synthesized utilizing the cyclization method previously described. *N,N*-Bis(2-chloroethyl)-4-ethoxycarbonyl-1,3,2-oxazaphospholidin-2-amine 2-oxide demonstrated activity against L-1210 lymphoid leukemia whereas *N,N*-bis(2-chloroethyl)-4-(ethoxycarbonyl)tetrahydro-2*H*-1,3,2-oxazaphosphorin-2-amine 2-oxide did not. The oxazaphosphorin-2-amine was evaluated against human epidermoid carcinoma of the nasopharynx (cell culture). The results again were negative: $ED_{50} = 2.8 \times 10$.

Keyphrases □ Cyclophosphamide analogs—synthesized, antitumor activity evaluated □ Antitumor activity—evaluated in cyclophosphamide analogs □ Structure–activity relationships—4-ethoxycarbonyl cyclophosphamide analogs evaluated for antitumor activity

Previous studies demonstrated that cyclophosphamide is an active antitumor agent (1). The active principle responsible for significant inhibitory activity against Yashida sarcoma in rats and L-1210 leukemia in mice is 4-hydroxycyclophosphamide (1, 2). Compounds such as *N,N*-bis(2-chloroethyl) -4- ethoxycarbonyl -1,3,2- oxazaphospholidin-2-amine 2-oxide (I) and *N,N*-bis(2-chloroethyl)-4-(ethoxycarbonyl)tetrahydro -2*H*- 1,3,2-oxazaphosphorin-2-amine 2-oxide (II) were readily synthesized, as described previously (3), by reaction of *N,N*-bis(2-chloroethyl)phosphinamide dichloride (III) with DL-serine ethyl ester hydrochloride and DL-homoserine ethyl ester hydrochloride, respectively, in the presence of triethylamine (Scheme I). All products were isolated as oils. Elemental analysis data of I and II are shown in Table I. The antitumor activity of I and II was evaluated on the basis of survival[1] (Table II).



Scheme I

Table I—Physical Data for the 4-Ethoxycarbonyl Cyclophosphamide Analogs

Compound	Yield, %	Formula		Analysis, % Calc.	Found
I	35.4	$C_9H_{17}Cl_2N_2O_4P$	C	33.87	33.42
			H	5.37	5.43
			Cl	22.71	22.38
			N	8.77	8.41
			P	9.70	9.60
II	23.1	$C_{10}H_{19}Cl_2N_2O_4P$	C	36.05	36.04
			H	5.75	5.21
			Cl	21.28	21.13
			N	8.40	8.68
			P	9.29	9.45

EXPERIMENTAL[2]

Compound I—DL-Serine ethyl ester hydrochloride, 1.25 g (0.002 mole), was dissolved in warm dioxane and 4 ml (0.029 mole) of triethylamine and then added slowly to a solution of 2.43 g (0.009 mole) of III in dioxane while being stirred under a nitrogen atmosphere. The mixture was stirred overnight at room temperature.

The reaction mixture was then filtered, and the solvent was evaporated under reduced pressure. Separation on magnesium silicate[3] gave 1.5 g

[1] National Cancer Institute Drug Research and Development, National Institutes of Health, Bethesda, MD 20014.

[2] IR spectra were run on a Perkin-Elmer 247 spectrophotometer. Elemental analyses were performed by Galbraith Laboratories, Knoxville, Tenn.

[3] Florisil.