Antitumor 1-(X-Aryl)-3,3-dialkyltriazenes. 1. Quantitative Structure–Activity Relationships vs. L1210 Leukemia in Mice[1]

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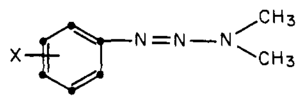
Quantitative structure–activity relationships (QSAR) have been formulated for phenyl-, pyrazolyl-, and imidazolyltriazenes acting against L1210 leukemia in mice. All three sets of congeners have the same ideal lipophilicity ($\log P_0 \approx 1$). Electron-releasing substituents increase potency; ortho substitution decreases activity. The synthesis of a number of new triazenes and some of their partition coefficients are reported.

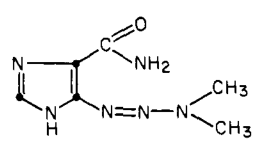
Since the discovery in 1955 that 1-(X-phenyl)-3,3-dimethyltriazenes I acted to inhibit[2] murine Sarcoma 180,



I

many attempts have been made to find more effective derivatives for cancer chemotherapy. Rondestvedt and Davis[3] were the first to make an extensive study of derivatives of I which were tested as antitumor agents at the Sloan-Kettering Institute. A conclusion from this study, which has also been drawn by others,[4] was that at least one *N*-methyl group appeared essential for antitumor activity.

The synthesis[5] and discovery of the anticancer activity[6] of 5-(3,3-dimethyl-1-triazeno)imidazole-4-carboxamide (DTIC, II) ushered in a new era of triazene study. DTIC



II

was soon shown to be clinically effective against malignant melanoma. A recent review[7] shows that in a number of separate studies involving 851 patients in all, there was an average response of 24% (complete and partial response). The only other antitumor agents showing significant antimelanoma activity are the nitrosoureas[7] and actinomycin D.[8] Combination chemotherapy using DTIC, BCNU, and vincristine has achieved a 63% response in a limited set of 16 patients[9] but this has not yet been confirmed by other studies.

A more promising treatment for malignant melanoma is the combined chemotherapy with DTIC, Me-CCNU, and immunotherapy.[10] One of the distinct advantages of DTIC in this combined modality is that it is minimally immunosuppressive in man. A negative aspect of the superficial success of DTIC is that once in the clinic, its use tends to discourage attempts to find a better triazene.

Although DTIC is the most widely used drug against melanoma, it is far from being a satisfactory agent. Connors et al.[11] have recently pointed out that one of the reasons for the lack of success with DTIC is its poor in vivo stability. Loo and his associates have made extensive studies of the fate of DTIC in humans as well as in animals.[12]

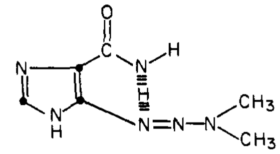
With the realization that DTIC is not going to solve the problem of malignant melanoma, both Connors' and Loo's groups have initiated new and more systematic studies of congeners of I.[11,13] It is clear from their work, as well as that of others, that derivatives of I are just as active as DTIC and, moreover, it is easier to do systematic SAR with phenyltriazenes than with heterocyclic triazenes.

One of the serious disadvantages of the phenyltriazenes,[14] DTIC,[15] and many other of the present antitumor drugs[14] is their known carcinogenicity as well as mutagenicity[16–18] in animals. This carcinogenicity is particularly disturbing in view of the evidence that chemical carcinogens appear to have a cumulative effect.[19]

The above rather bleak picture of triazene cancer chemotherapy suggests to us that careful quantitative structure–activity relationships must be formulated to discover those structural features which contribute to efficacy and those which contribute to toxicity. If these two features cannot be separated, the future of triazene chemotherapy seems severely limited.

Opportunities for the variation of I are manifold. For example, considering only substitution in the 2, 3, and 4 positions of the benzene ring and using 90 substituents for which complete sets of physicochemical constants are available,[20] we have the possibility of making 90^3 or 729 000 derivatives. One can easily think of 100 variations to make in place of one of the *N*-methyl groups or in the phenyl ring which would lead to 70 290 000 possible derivatives. Clearly, the job of making and testing a truly representative sample of this vast number of possibilities is a serious challenge for which medicinal chemistry, as of today, has no complete answer. Our present intention is to attempt to delineate the role of hydrophobic, electronic, and steric substituent effects on anticancer activity.

A number of monosubstituted ortho derivatives were made in the hope that either a beneficial steric or hydrogen-bonding effect could be uncovered. The X-ray crystallographic study of DTIC by Edwards et al.[21] showed strong hydrogen bonding as in III. It was thought that



III

this might have some role in the anticancer activity but results from compounds synthesized to test this hypothesis make this seem unlikely.

In an analysis[4] of the activity of derivatives of DTIC in

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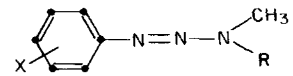
Table I. Physicochemical and Antitumor Data for Phenyltriazenes of Equation 2



No.	X[a]	R	Log 1/C: Obsd	Log 1/C: Calcd	|Δ log 1/C|	E_s-R[b]	$\Sigma\sigma^+$[c]	ΣMR-2,6[b]	Log P	NSC no.
1	4-NHCOCH$_3$	CH$_3$	4.04	3.84	0.20	−1.24	−0.60	0.20	1.54	157031
2	4-NHCONH$_2$	CH$_3$	3.97	3.88	0.09	−1.24	−0.69[d]	0.20	1.25[e]	268492
3	3-CONH$_2$, 6-OCH$_3$	CH$_3$	3.95	3.68	0.27	−1.24	−0.50	0.84	0.44[e]	276374
4	4-NHCONH$_2$	CH$_2$CH$_3$	3.87	3.84	0.03	−1.31	−0.69[d]	0.20	1.75	279501
5	4-NHCOH	CH$_3$	3.85	3.84	0.01	−1.24	−0.60[f]	0.20	1.53	276376
6	H	CH$_3$	3.85	3.58	0.27	−1.24	0.00	0.20	2.59[e]	3094
7	3-CONH$_2$	CH$_3$	3.80	3.57	0.23	−1.24	0.28	0.20	1.21	140017
8	4-CH$_3$	CH$_3$	3.76	3.63	0.13	−1.24	−0.31	0.20	2.93	48821
9	4-NHCONH$_2$	CH$_2$CH=CH$_2$	3.77	3.76	0.01	−1.45[g]	−0.69[d]	0.20	2.14	279502
10	4-SO$_2$NH, 2-pyrimidyl	CH$_3$	3.74	3.48	0.26	−1.24	0.57[h]	0.20	1.41	166759
11[i]	2-CO$_2$H	n-C$_3$H$_7$	3.74	3.10	0.64	−1.60	−0.02	0.71	−1.66	173201
12[j]	4-CONH$_2$	CH$_2$CH$_3$	3.66	3.51	0.15	−1.31	0.36	0.20	1.70	276375
13[i]	2-COOH	CH$_3$	3.64	2.95	0.69	−1.24	−0.02	0.71	−2.66	210718
14	2,6-F$_2$	CH$_3$	3.63	3.59	0.04	−1.24	−0.15	0.16	2.87	251241
15	4-SO$_2$NH$_2$	CH$_3$	3.60	3.48	0.12	−1.24	0.57	0.20	0.98[e]	157030
16	4-(CH$_2$)$_3$CONHNH$_2$	CH$_3$	3.60	3.76	0.16	−1.24	−0.31[k]	0.20	1.46	83693
17	H	H	3.60	3.64	0.04	−1.24	0.00	0.20	1.94	136056
18	m-COCH$_3$	CH$_3$	3.58	3.50	0.08	−1.24	0.38	0.20	2.19	226086
19	4-(CH$_2$)$_3$CO$_2$C$_2$H$_5$	CH$_3$	3.54	3.45	0.09	−1.24	−0.31[k]	0.20	3.87	80637
20	4-CONH$_2$	CH$_3$	3.51	3.55	0.04	−1.24	0.36	0.20	1.20[e]	86441
21[j]	4-CONH$_2$	n-C$_4$H$_9$	3.47	3.33	0.14	−1.63	0.36	0.20	2.46	87429
22*[p]	2-COOH	n-C$_8$H$_{17}$	3.47	3.33	0.14	−1.77	−0.02	0.71	0.24[e]	173202
23	2-CONH$_2$, 4-CN	CH$_3$	3.46	3.18	0.28	−1.24	1.02	1.08	1.53	258831
24	3-Pyridyl nucleus	CH$_3$	3.46	3.45	0.01	−1.24	0.67[l]	0.20	1.39	125093
25	2-CONH$_2$	CH$_2$CH=CH$_2$	3.43	3.22	0.21	−1.45[g]	0.36	1.08	2.62	145123
26	3-CO$_2$CH$_3$	CH$_3$	3.42	3.45	0.03	−1.24	0.37	0.20	2.72	140015
27*	3-CH$_3$	CH$_3$	3.40	3.56	0.16	−1.24	−0.07	0.20	2.85	136067
28	4-CONH$_2$	CH$_2$CH=CH$_2$	3.38	3.43	0.05	−1.45[g]	0.36	0.20	2.09[e]	276372
29	2-CONH$_2$, 4-SO$_2$NH$_2$	CH$_3$	3.32	3.17	0.15	−1.24	0.93	1.08	0.12	258833
30[j]	2-CONH$_2$	CH$_3$	3.31	3.38	0.07	−1.24	0.36	1.08	1.73[e]	136896
31	2-NO$_2$	CH$_3$	3.28	3.22	0.06	−1.24	0.79	0.77	2.71	136066
32	2-CONH$_2$, 4-CONH$_2$	CH$_3$	3.27	3.25	0.02	−1.24	0.72	1.08	0.34	261725
33	3-CONH$_2$, 5-CONH$_2$	CH$_3$	3.27	3.45	0.18	−1.24	0.56	0.20	0.22[e]	268495
34	2-CONH$_2$, 4-NO$_2$	CH$_3$	3.26	3.13	0.13	−1.24	1.15	1.08	1.85	143908
35	2-Cl	CH$_3$	3.26	3.41	0.15	−1.24	0.11	0.68	2.97	515127
36	2-CONH$_2$	n-C$_3$H$_7$	3.26	3.15	0.11	−1.60	0.36	1.08	2.73	145137
37	2-CONHCH$_3$	CH$_3$	3.25	3.29	0.04	−1.24	0.36[m]	1.56	1.83[e]	261059
38	2-CONHCH$_2$CONH$_2$	CH$_3$	3.25	3.15	0.10	−1.24	0.36[m]	2.44	1.17[e]	263462
39	2-CONHCH$_2$CN	CH$_3$	3.22	3.20	0.02	−1.24	0.36[m]	2.09	1.79[e]	263461
40	4-COOH	CH$_3$	3.22	3.30	0.08	−1.24	−0.02	0.20	−1.77	228635
41	4-CO$_2$CH$_3$	CH$_3$	3.20	3.40	0.20	−1.24	0.49	0.20	2.77	140016
42	4-CONH$_2$	i-C$_3$H$_7$	3.17	3.34	0.17	−1.71	0.36	0.20	2.00	240524
43	2-COOH	n-C$_4$H$_9$	3.15	3.19	0.04	−1.63	−0.02	0.71	−1.16	210719
44	2-CO$_2$H, 4-Cl	n-C$_8$H$_{17}$	3.14	3.33	0.19	−1.77	0.09	0.71	0.97	183740
45	2-CONH$_2$, 4,6-Cl$_2$	CH$_3$	3.14	3.12	0.02	−1.24	0.59	1.56	2.85	146372
46*	2-CO$_2$CH$_3$	i-C$_4$H$_9$	3.12	2.64	0.48	−2.17	0.49	1.39	3.83	103540

47	2-CONH$_2$	n-C$_4$H$_9$	3.10	3.10	0.00	−1.63	0.36	1.08	2.99	136892
48	2-CONH$_2$, 4-NO$_2$	n-C$_4$H$_9$	3.08	2.84	0.24	−1.63	1.15	1.08	3.11	143149
49	2-CONH$_2$	CH$_2$CH$_2$OH	3.07	3.25	0.18	−1.60[n]	0.36	1.08	0.85	142025
50	2-CONHNHCOCH$_3$	CH$_3$	3.05	3.15	0.10	−1.24	0.36[m]	2.43	1.49[e]	260617
51	4-OCH$_3$	COC$_6$H$_5$	3.05	3.34	0.29	−2.43[o]	−0.78	0.20	2.70	156202
52	3-CONH$_2$, 2,5-Cl$_2$	CH$_3$	3.04	3.26	0.22	−1.24	0.79	0.68	2.42	153183
53	H	CH$_2$C$_6$H$_5$	3.03	3.11	0.08	−1.69	0.00	0.20	4.15	136060
54	4-(CH$_2$)$_2$CO$_2$C$_2$H$_5$	n-C$_4$H$_9$	3.02	3.15	0.13	−1.63	−0.31[k]	0.20	4.45	77587
55	4-CO$_2$C$_2$H$_5$	CH$_2$CH$_3$	3.01	3.20	0.19	−1.31	0.48	0.20	3.78	93192
56*	2-CONHNH$_2$	Cyclohexyl	2.99	2.76	0.23	−2.03	0.36[m]	1.41	3.67	103537
57	2-CONHCH$_2$CH$_3$	CH$_3$	2.92	3.17	0.25	−1.24	0.36[m]	2.03	2.33	261060
58*	4-CONH$_2$	s-C$_4$H$_9$	2.90	3.06	0.16	−2.37	0.36	0.20	2.26	93194
59	4-CONH$_2$	n-C$_8$H$_{17}$	2.84	2.82	0.02	−1.77	0.36	0.20	4.70	276741
60	4-CO$_2$C$_2$H$_5$	i-C$_4$H$_9$	2.80	2.73	0.07	−2.17	0.48	0.20	4.34	93191
61[i]	4-CH$_3$	COCH$_3$	2.78	3.41	0.63	−1.71	−0.31	0.20	3.13[e]	208827
62	3,5-(CN)$_2$	CH$_3$	2.78	3.27	0.49	−1.24	1.12	0.20	2.18[e]	284698
63	4-CN	CH$_2$C$_6$H$_4$-4-OCH$_3$	2.72	2.99	0.27	−1.69	0.66	0.20	3.80	220338
64*	2-CONHNHCOCH$_2$CN	CH$_3$	2.63	3.05	0.42	−1.24	0.36[m]	2.96	1.44[e]	261061

[a] COOH is treated as COO$^-$ due to physiological ionization. [b] S. H. Unger and C. Hansch, *Prog. Phys. Org. Chem.*, 12, 92 (1976), and reference 10; E_s referenced to $H = 0$ and MR-2,6 scaled by 0.1. [c] J. E. Leffler and E. Grunwald, "Rates and Equilibria of Organic Reactions", Wiley, New York, N.Y., 1963, p 204; σ^+ values for ortho substituents are assumed, as a best approximation, to be equal to σ^+ for the para isomer; σ^+ values for electron-withdrawing substituents are assumed equal to normal σ when the actual value is not available. [d] Estimated from the σ^+ and σ values for 4-OCH$_3$ and σ for 4-NHCONH$_2$. [e] Experimentally determined log P. [f] Estimated from the σ^+ and σ values for 4-NHCOCH$_3$ and σ for 4-NHCOH. [g] Estimated from the values for -CH$_2$CH$_2$CH$_3$ and -CH$_2$CH$_3$. [h] σ^+ values assumed equal to that for 4-SO$_2$NH$_2$. [i] These data points are not used in deriving equations. [j] Prepared according to the method of D. E. V. Wilman, Ph.D. Thesis, University of London, London, England, 1974. [k] σ^+ value assumed similar to that for 4-CH$_3$. [l] 3-Pyridyl nucleus assumed equal to 3-nitrophenyl system. [m] σ^+ value assumed similar to 2-CONH$_2$. [n] Estimated from the E_s value for -CH$_2$CH$_2$CH$_3$. [o] Estimated from the E_s value for -CH(CH$_3$)C$_6$H$_5$. [p] An asterisk indicates that these molecules did not achieve a T/C of 125.

Table II. Development of Equation 2 for Phenyltriazenes

Intercept	E_s-R	$\Sigma\sigma^+$	MR-2,6	(Log P)^2	Log P	r	s	$F_{1,X}$[a]
4.05	0.50					0.459	0.299	15.74
4.20	0.55	0.39				0.710	0.239	34.21
4.31	0.56	0.32	0.15			0.762	0.221	10.56
4.22	0.41	0.30	0.18	0.02		0.816	0.199	14.34
4.12	0.39	0.31	0.18	0.04	0.10	0.836	0.191	5.95

[a] $F_{1,40;\alpha=0.001} = 12.6$; $F_{1,40;\alpha=0.025} = 5.42$.

Table III. Squared Correlation Matrix for Variables of Equation 2

	E_s-R	σ^+	MR-2,6	Log P
E_s-R	1.00	0.01	0.01	0.12
σ^+		1.00	0.09	0.00
MR-2,6			1.00	0.01
Log P				1.00

which one N–CH$_3$ was replaced by a variety of alkyl groups, we were able to formulate a QSAR from which an optimum log P of 1.1 (0.8–2.4) was found. Since it is important to establish this boundary for the phenyltriazenes, considerable variation in log P was built into the congeners in Table I. We were especially concerned with making more hydrophilic drugs since this class has been somewhat neglected in the past.

The problem of relating optimum lipophilicity to antitumor activity is a complex one which must receive much more systematic study. Cancer is different from most diseases with which medicinal chemists must contend. For example, it has been shown that the introduction of *one* cell of L1210 leukemia is sufficient to produce fatal cancer in a mouse; this means that the cancer chemotherapist must have drugs which will penetrate into every cavity of the body and selectively destroy all cancer cells without causing irreparable damage to sensitive normal cells. We have pointed out[22] that it is unlikely that a single drug can be designed to penetrate all hydrophobic as well as hydrophilic barriers to reach every body compartment in concentrations sufficient to destroy all tumor cells. In designing combination chemotherapy (using a number of drugs), one should take into account relative lipophilicities of drugs as well as their different modes of action on the cell cycle.

The importance of proper hydrophobicity can be illustrated with triazene data. As mentioned above, log P_0 of 1.1 was found for a set of derivatives which produced a response of test/control on L1210 leukemia in mice of 150%. In a study of similar derivatives against brain tumors, log P_0 could not be defined accurately but was obviously[23] much higher than 1.1.

Nonspecific toxicity is also a function of log P and does not necessarily parallel antitumor activity.[4] This too must be minimized.

The electronic effect of substituents on the ring and the electronic effect of the ring on the triazene side chain must also be studied in order to take advantage of any possible increase in antitumor activity by electron-releasing or -withdrawing substituents. Therefore, a range in σ of the substituents attached to the phenyl ring was an important objective in preparing the set of triazenes of this report.

In this report we discuss the formulation of QSAR (eq 2, 4, and 5) for several classes of triazenes and compare the results from our earlier study[4] (eq 1) and the study of Dunn[13b] (eq 3). Equation 1 was formulated[4] for congeners

$$\log 1/C = -0.28 (\log P)^2 + 0.59 \log P + 3.45 \quad (1)$$

$$n = 10; r = 0.929; s = 0.146; \log P_0 = 1.1$$

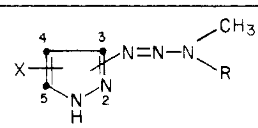
Table IV. Inactive and Toxic Phenyltriazenes[a] vs. L1210 Leukemia in Mice

$X{-}C_6H_4{-}N{=}N{-}N(CH_3)R$

No.	X	R	Results[b]	Log 1/*C* calcd	NSC no.
1	2-$CONHOCH_3$	CH_3	Toxic	3.26	260618
2	2-$CONH_2$, 4-Cl	CH_3	Toxic	3.29	143907
3	2-$CONHNH_2$	CH_3	Toxic	3.33	102247
4	2-OCH_3	CH_3	Toxic	3.74	173097
5	2-CH_2OH	CH_3	Toxic	3.59	183741
6	2-Cl	CH_2CH_2OH	Toxic	3.37	180040
7	2-CN	CH_2CH_2OH	Toxic	3.23	180041
8	H	CH_2CH_2OH	Toxic	3.51	180039
9	2-CO_2H	Cyclohexyl	Toxic	3.18	173203
10	2-CO_2H, 4-OCH_3	CH_3	Toxic	3.13	233877
11	3-NO_2	CH_3	Toxic	3.36	82309
12	3-Cl	CH_3	Toxic	3.32	515463
13	3-$NHCONH_2$	CH_3	Toxic	3.69	284697
14	4-CH_3	H	Toxic	3.71	183741
15	4-$CONH_2$	$NHCH_3$	Toxic	3.73	284696
16	4-$CONH_2$	Diallyl[c]	Toxic		284695
17	4-$CONH_2$	OCH_3	Toxic	3.82	279831
18	4-$CONH_2$	CH_2CN	Toxic	3.12	279830
19	4-NO_2	CH_3	Toxic	3.32	408428
20	4-Cl	CH_3	Toxic	3.43	115221
21	4-CF_3	CH_3	Toxic	3.23	157033
22	4-CN	CH_3	Toxic	3.40	157034
23	4-$COCH_3$	CH_3	Toxic	3.46	157032
24	2-I	CH_3	Inactive	3.16	173095
25	2-CN	CH_3	Inactive	3.32	180036
26	2-CF_3	CH_3	Inactive	3.11	180038
27	2-SCH_3	CH_3	Inactive	3.46	173098
28	2-$CONHCH_2CF_3$	CH_3	Inactive	3.09	260619
29	2-$CONH_2$, 4-SCN	CH_3	Inactive	3.17	258832
30	2-CO_2H, 4-Cl	CH_3	Inactive	3.14	233879
31	2-CO_2H, 4-NO_2	CH_3	Inactive	2.75	233878
32	4-$CONH_2$	$C(CH_3)_3$	Inactive	2.89	276373
33	4-COOH	*n*-C_8H_{17}	Inactive	3.41	183739
34	2-$CONHNHCSNH_2$	CH_3	Erratic	3.00	258834

[a] This work. [b] Toxic molecules include those which result in severe weight loss in the test animals and usually in death of one or more of the test subjects during the first 5 days of the experiment; inactive molecules include those which do not result in significant increases in life span (T/C > 110); the erratic molecule produced such a scattered antitumor result that a reliable value of log 1/*C* could not be obtained. [c] 4-$CONH_2$-C_6H_4-N=N-N($CH_2CH{=}CH_2$)$_2$.

Table V. Physicochemical and Antitumor Data for Pyrazolyltriazenes of Equation 4



No.	X	Position of triazene	R	Log *P*	*I*-2	Log 1/*C* Obsd	Log 1/*C* Calcd	|Δ log 1/*C*|	NSC no.
1	3-$CONH_2$, 5-CH_3	4	CH_3	0.44	0.0	3.84	3.73	0.11	123145
2	3-$CONH_2$, 5-CH_3	4	*n*-C_4H_9	1.96	0.0	3.83	3.63	0.20	136879
3	4-$CONH_2$	3	*n*-C_4H_9	1.40	0.0	3.81	3.76	0.05	121239
4	4-$CONH_2$	3	Allyl	0.77	0.0	3.77	3.78	0.01	153186
5	4-$CONH_2$	3	*n*-C_3H_7	0.90	0.0	3.70	3.79	0.09	145929
6	4-$CONH_2$	3	$CH_2CH_2C_6H_5$	2.11	0.0	3.52	3.58	0.06	145930
7	4-CO_2CH_3	3	CH_3	0.04	1.0	3.49	3.27	0.22	117122
8	4-$CONH_2$	3	CH_3	−0.12[a]	0.0	3.49	3.57	0.08	114924
9	3-$CONH_2$	4	CH_3	−0.12	0.0	3.42	3.57	0.15	131260
10	4-$CO_2CH_2CH_3$	3	CH_3	0.56	1.0	3.35	3.40	0.05	115757
11	3-CO_2CH_3, 5-*i*-C_3H_7	4	CH_3	1.57	1.0	3.30	3.38	0.08	131255
12	4-$CO_2CH_2CH_3$	3	*n*-C_4H_9	2.08	1.0	3.16	3.24	0.08	118319
13	4-$CONH_2$	3	CH_2CH_2OH	−1.00	0.0	3.10	3.09	0.01	133729

[a] Log *P* determined experimentally.

of II in which one *N*-methyl group was replaced by various alkyl groups. *C* in eq 1 is moles per kilogram producing a T/C (T = life span of test animal inoculated with leukemia and treated with drug; C = life span of control animal inoculated intraperitoneally but receiving no drug; see Geran et al.[24] for experimental details) of 150. In this report *n* represents the number of data points upon which the equation is based, *r* is the correlation coefficient, *s* is the standard deviation, and log P_0 is the optimum log *P* for a given set of congeners.

Table VI. Development of QSAR of Equation 4

Intercept	I-2	Log P	(Log P)^2	r	s	$F_{1,X}$[a]
3.61	0.28			0.538	0.224	4.47
3.54	0.32	0.09		0.641	0.213	2.07
3.61	0.35	0.35	0.17	0.895	0.131	17.56

[a] $F_{1,9;\alpha=0.005} = 13.61$; $F_{1,10;\alpha=0.25} = 1.49$.

In the formulation of eq 2, we have used data from molecules prepared in our laboratory, as well as from the National Cancer Institute[24] (NCI). Equations 4 and 5 are based on NCI data.

Results

Equation 2 has been derived from data in Table I for

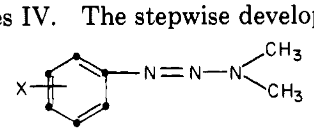
$$\log 1/C = 0.100\ (\pm 0.08) \log P - 0.042\ (\pm 0.02)\ (\log P)^2 - 0.312\ (\pm 0.11)\ \Sigma\sigma^+ - 0.178\ (\pm 0.08)\ \text{MR-2,6} + 0.391\ (\pm 0.18)\ E_s\text{-R} + 4.124\ (\pm 0.27) \quad (2a)$$

$n = 61$; $r = 0.836$; $s = 0.191$; $\log P_0 = 1.18\ (0.36\text{–}1.68)$

$$\log 1/C = -0.002 \log P - 0.022\ (\log P)^2 - 0.295\ \sigma^+ - 0.166\ \text{MR-2,6} + 0.422\ E_s\text{-R} + 4.244 \quad (2b)$$

$n = 64$; $r = 0.789$; $s = 0.217$; $\log P_0 = -0.001\ (-4.9 \text{ to } 0.93)$

phenyltriazenes IV. The stepwise development of eq 2



IV

is given in Table II and the collinearity among the variables is shown in Table III. C in eq 2 is moles per kilogram producing a T/C of 140 (the drug administration regimen in this paper consisted of daily injections on days 1–9, followed by evaluation on day 30), MR-2,6 refers to the sum of molar refractivity of the substituents flanking the triazene side chain (MR is scaled by 0.1), and E_s is the Taft steric parameter for the largest R (when R = H, E_s for CH_3 is used). The negative coefficient with $\Sigma\sigma^+$ indicates that electron release via through resonance increases activity and the negative weighting factor with MR-2,6 shows that large X groups in the ortho position depress activity; large R groups also reduce activity. There are almost 15 data points/variable supporting eq 2a.

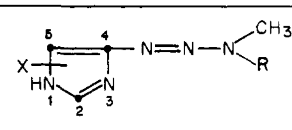
The most interesting aspect of this equation is the value of log P_0 which agrees surprisingly well with that of eq 1. Although the ring systems are quite different, hydrophobic requirements are the same.

Three data points in Table I have not been used in the derivation of eq 2a; two of these contain carboxyl groups and one contains an unusual side chain: $N{=}NN(CH_3)COCH_3$. The triazenes are extremely toxic compounds and toxicity parallels activity rather closely. In many instances, toxicity masks antitumor activity, preventing the determination of a T/C; such toxic compounds and inactive compounds are listed in Table IV.

Equation 2b contains all data points of Table I, while eq 2a has been formulated without the three most poorly fit points. Except for the log P coefficients, the parameters of eq 2a and 2b do not differ significantly. Equation 2b suggests a lower log P_0, but the poor confidence limits on this parameter discount its value.

Six of the compounds in Table I (**22**, **27**, **46**, **56**, **58**, and **64**) did not achieve T/C values of 125; however, two data points were available in the range of 112–120. Since a T/C of 140 for these congeners was estimated by extrapolation, eq 2a was derived without these data points. This yielded

Table VII. Physicochemical and Antitumor Data for Imidazolyltriazenes of Equation 5



No.	X	R	Log P	Log 1/C Obsd	Log 1/C Calcd	|Δ log 1/C|	NSC no.
1	5-$CO_2C_6H_5$	CH_3	0.44[a]	3.98	3.57	0.41	98 662
2	5-$CONH_2$	s-C_4H_9	1.08[b]	3.87	3.59	0.28	144 216
3	5-CO_2CH_3	CH_3	−0.06[c]	3.86	3.49	0.37	87 982
4	5-$CONH_2$[b]	$CH_2C{\equiv}CH$	0.33	3.61	3.56	0.05	173 351
5	5-$CONH_2$	n-C_5H_{11}	1.78	3.58	3.52	0.06	87 981
6	5-$CONH_2$	n-C_3H_7	0.78	3.58	3.59	0.01	76 418
7	5-$CONH_2$	n-C_4H_9	1.28	3.54	3.58	0.04	70 874
8	5-$CO_2C_6H_5$	CH_3	1.44	3.53	3.57	0.04	112 477
9	2-i-C_4H_9, 5-$CONH_2$	CH_3	1.56	3.53	3.55	0.02	127 836
10	5-$CONH_2$	i-C_4H_9	1.08	3.51	3.59	0.08	83 113
11	5-$CONH_2$	$CH_2CH_2C_6H_5$	1.99	3.49	3.48	0.01	146 371
12	2-C_6H_5, 5-$CO_2C_2H_5$	CH_3	2.40	3.48	3.38	0.10	166 721
13	5-$CONH_2$	$CH_2C_6H_5$	1.54[a]	3.43	3.56	0.13	83 695
14	2-n-C_3H_7, 5-$CONH_2$	CH_3	1.31	3.42	3.58	0.16	127 837
15	5-$CO_2CH_2CH_3$	CH_2CH_2OH	−0.44	3.38	3.41	0.03	105 766
16	2-CH_3, 5-$CO_2C_2H_5$	CH_3	1.00	3.35	3.59	0.24	166 722
17	5-CO_2CH_3	H	−0.89	3.35	3.27	0.08	105 530
18	5-$CONH_2$	CH_3	−0.24[a]	3.29	3.46	0.17	45 388
19	5-CO-c-NC_4H_8	H	−0.86	3.28	3.28	0.00	145 924
20	5-$CONH_2$	CH_2CH_2OH	−1.12	3.12	3.18	0.06	83 112
21	2-CH_3, 5-$CONH_2$	CH_3	0.32[c]	3.06	3.56	0.50	140 406
22	5-$CO_2C_8H_{17}$	CH_3	3.44	3.06	2.99	0.07	100 863
23	5-$CONH_2$	n-C_8H_{17}	3.22[a]	3.00	3.09	0.09	208 826
24	2-$CH_2C_6H_5$, 5-$CONH_2$	CH_3	1.77[c]	2.89	3.52	0.63	127 838

[a] Log P determined experimentally. [b] Log P determined, ref 29. [c] Not utilized in the correlation equation.

Table VIII. Development of QSAR for Equation 5

Intercept	$(\log P)^2$	$\log P$	r	s	$F_{1,X}$[a]
3.54	0.04		0.534	0.202	7.58
3.51	0.10	0.18	0.780	0.154	14.88

[a] $F_{1,17;\alpha=0.005} = 10.4$.

an equation whose parameters did not differ significantly from eq 2a with $r = 0.871$, $s = 0.168$, and $\log P_0 = 1.20$. Of these six data points, two (46 and 64) are rather badly fit which can be seen in Table I.

A T/C of 150 was used as the end point in eq 1; in this report we felt that a T/C of 140 was a slightly better standard. We have found that small differences in the T/C standard do not affect the parameters of the correlation equation except for the intercept.

Dunn and his colleagues have studied[13b] a set of phenyltriazenes of type II on Sarcoma 180 tumor in mice and derived eq 3. *In this study on Sarcoma 180 tumor, the* hydrophobic character of the drugs as modeled by π played no role in the antitumor activity. The coefficient with σ is similar to that of eq 2a. In the case of Dunn's work, there was little difference in σ and σ^+ for the substituents considered so that it is not possible to make a strict comparison between the two equations. The difference in π dependence may be a result of the different types of tumors and the difference in mode of inoculation.

$$\log 1/C = -0.69\,\sigma + 3.41 \qquad (3)$$

$$n = 13;\ r = 0.922;\ s = 0.09$$

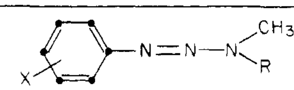
A second set of NCI data on pyrazolyltriazenes (Table V) yields eq 4, the development of which is given in Table VI.

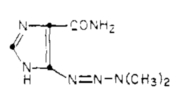
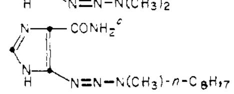
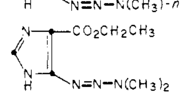
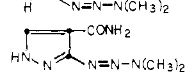
$$\log 1/C = 0.350\ (\pm 0.17) \log P - 0.173\ (\pm 0.09)\ (\log P)^2 - 0.349\ (\pm 0.18)\ I + 3.610\ (\pm 0.12) \qquad (4)$$

$$n = 13;\ r = 0.895;\ s = 0.131;\ \log P_0 = 1.01\ (0.75\text{-}1.49)$$

The variables I and $\log P$ in eq 4 are reasonably orthogonal ($r^2 = 0.03$) with the indicator variable given the value of 1 for congeners containing an ester group. The most interesting aspect of eq 4 is the value of $\log P_0$ of 1.01 which is in good agreement with that found for eq 1 and 2.

Table IX. Experimentally Determined Partition Coefficients of Selected Triazenes[a]



X	R	NSC no.	Log P	No. of determinations
H[b]	CH_3	3094	2.59 ± 0.02	4
H[c]	CH_2CH_2OH	180039	1.71 ± 0.01	4
4-$CONH_2$	CH_3	86441	1.20 ± 0.01	4
4-$CONH_2$	$CH_2CH{=}CH_2$	276372	2.09 ± 0.01	4
4-$CONH_2$	n-C_4H_9	87429	2.46 ± 0.01	4
4-$NHCONH_2$	CH_3	268492	1.25 ± 0.02	4
4-SO_2NH_2	CH_3	157030	0.98 ± 0.01	4
4-CN	CH_3	157034	2.39 ± 0.02	4
3-NO_2	CH_3	83209	2.75 ± 0.03	4
2-$CONH_2$[b]	CH_3	136896	1.73 ± 0.01	6
2-$CONHCH_3$	CH_3	261059	1.83 ± 0.01	4
2-CONHCN	CH_3	268490	0.80 ± 0.07	3
2-$CONHNH_2$	CH_3	102247	1.28 ± 0.03	4
2-$CONHCH_2CONH_2$	CH_3	263462	1.17 ± 0.02	4
2-$CONHCH_2CN$	CH_3	263461	1.79 ± 0.03	6
2-$CONHNHCOCH_2CN$	CH_3	261061	1.44 ± 0.14	4
2-$CONHNHCOCH_3$	CH_3	260617	1.49 ± 0.01	4
2-SCH_3[b]	CH_3	173098	3.13 ± 0.02	4
2-OCH_3[b]	CH_3	173097	2.34 ± 0.03	3
2-Cl[c]	CH_3	515127	2.97 ± 0.01	4
2-I[c]	CH_3	173095	3.50 ± 0.02	3
2-CH_2OH[c]	CH_3	180037	1.73 ± 0.01	4
2-COOH[c]	n-C_3H_7	173201	−1.66 ± 0.01	2
2-COOH[c]	n-C_8H_{17}	173202	0.24 ± 0.01	4
3-$CONH_2$, 6-OCH_3	CH_3	276374	0.44 ± 0.01	5
3,5-$(CONH_2)_2$	CH_3	268495	0.22 ± 0.01	4
3,5-$(CN)_2$	CH_3	284698	2.18 ± 0.01	3
4-CH_3	$COCH_3$	208827	3.23 ± 0.02	3
		45388	−0.24 ± 0.02	4
		208826	3.22 ± 0.04	4
		98662	0.44 ± 0.02	4
		117122	−0.12 ± 0.01	4

[a] Determined with octanol-saturated water and water-saturated octanol unless otherwise indicated. [b] Aqueous phase, 7.4 phosphate buffer. [c] Aqueous phase, 0.01 M NaOH.

Table X. Data for and Predicted π Values of Equation 7

$X{-}C_6H_4{-}N{=}NN(CH_3)_2$

				π		
No.	X	σ	π_{arom}[a]	Obsd[b]	Calcd	$\lvert\Delta\pi_X\rvert$
1	3-CF_3	0.43	0.88	1.19[c]	1.04	0.15
2	3-Cl	0.37	0.71	0.85[c]	0.83	0.02
3	3-SCH_3	0.15	0.61	0.39[c]	0.59	-0.20
4	3-CH_3	-0.07	0.56	0.26[c]	0.40	-0.14
5	3-NO_2	0.71	-0.28	0.16[d]	0.08	0.08
6	H	0.00	0.00	0.00[d]	-0.10	-0.10
7	4-CN	0.66	-0.57	-0.20[d]	-0.24	-0.04
8	3-$NHCOCH_3$	0.21	-0.97	-0.98[c]	-0.91[a]	-0.07
9	4-$NHCONH_2$	-0.24	-1.30	-1.34[d]	-1.52	-0.18
10	4-$CONH_2$	0.36	-1.49	-1.39[d]	-1.33	-0.06
11	4-SO_2NH_2	0.57	-1.82	-1.61[d]	-1.52	-0.09
12	4-$NHCOCH_3$	0.00	-0.97		-1.05[e]	
13	4-NHCOH	0.00	-0.98		-1.06[e]	
14	3-$CONH_2$	0.28	-1.49		-1.38[e]	
15	4-CH_3	-0.17	0.56		0.34[e]	
16	4-NO_2	0.78	-0.28		0.12[e]	
17	3-$COCH_3$	0.38	-0.55		-0.40[e]	
18	3-CO_2CH_3	0.37	-0.01		0.13[e]	
19	4-$CO_2CH_2CH_3$	0.45	0.51		0.69[e]	
20	4-OCH_3	-0.27	-0.02		-0.29[e]	
21	4-COOH	0.00	-4.36[e]		-4.36	
22	4-I	0.18	1.12[e]		1.11	
23	4-F	0.06	0.14[e]		0.08	
24	4-Br	0.23	0.86[e]		0.92	
25	4-Cl	0.23	0.71[e]		0.74	
26	3-Cl	0.37	0.71[e]		0.83	
27	4-CF_3	0.54	0.88[e]		1.11	

[a] Reference 10. [b] $\pi_{obsd} = \log P_{X-C_6H_4-N=NN(CH_3)_2} - \log P_{C_6H_5-N=N-N(CH_3)_2}$. [c] Calculated from appropriate log P values from ref 13. [d] Calculated from appropriate log P values in Table VIII. [e] Predicted π value for substituted triazenes using eq 7.

Table XI. Development of Equation 7

Intercept	π obsd	σ	r	s	$F_{1,X}$[a]
0.08	0.96		0.970	0.243	144.12
-0.10	0.98	0.64	0.991	0.139	32.33

[a] $F_{1,8;\alpha=0.001} = 25.4$.

Equation 5 has been derived from data on a set of

$$\log 1/C = 0.180\ (\pm 0.10) \log P - 0.096\ (\pm 0.04) (\log P)^2 + 3.507\ (\pm 0.09) \quad (5a)$$

$$n = 21;\ r = 0.780;\ s = 0.154;\ \log P_0 = 0.93\ (0.59\text{-}1.25)$$

$$\log 1/C = 0.137 \log P - 0.084 (\log P)^2 + 3.487 \quad (5b)$$

$$n = 24;\ r = 0.566;\ s = 0.235;\ \log P_0 = 0.82\ (-0.12 \text{ to } 1.38)$$

imidazolyltriazenes (Table VII). Equation 5b is based on all of the data points of Table VII. The parameters of eq 5a and 5b are quite close; however, eq 5a is a much poorer correlation. In both eq 4 and 5, C is the concentration of drug producing a T/C of 140 in the 1–9-day regimen, as in eq 2. While eq 5 is a rather poor correlation in terms of r, it is interesting that the same log P_0 is found as in the case of eq 1, 2, and 4. The development of eq 5 is given in Table VIII.

A result of these studies which seems quite firm is that triazenes acting against L1210 leukemia have a common ideal log P of essentially 1.

Equations 2 and 3 bring out the advantage of incorporating electron-releasing groups on the aromatic nucleus. The congeners upon which eq 1, 4, and 5 are based do not contain sufficient variation with respect to σ for ring substituents to confirm the findings of eq 2 and 3.

Correlation equations 2, 4, and 5 are not as sharp as one would like. Part of the reason for this is that the compounds have been supplied to the NCI over a period of years and tested in a number of different laboratories; also, some of the congeners are quite unstable in solution and, hence, difficult to test. In addition, the highly toxic nature of these substances makes it difficult to find the narrow region of activity between toxicity and inactivity. Although the quality of fit obtained with eq 2, 4, and 5 is not high in terms of r, the standard deviation is not bad for the type of testing involved.

The problem of defining the relative activity of antitumor drugs is the most difficult of any class of drugs. Efficacy and toxicity often parallel each other so closely that curative activity can easily be masked by toxicity unless extensive gradation of dose is carefully studied. At present, the great expense of such testing precludes its use.

One might question the use of selecting some arbitrary T/C to define activity in terms of log 1/C. The mathematical model we have elected to employ dictates defining activity in terms of molar concentration producing a standard response.[25] One might elect to use some other definition, such as the dose which produces a maximum response. Our experience suggests that this would be a mistake from the point of view of QSAR. We firmly believe in the principles of the extrathermodynamic approach to structure–activity relationships. So little QSAR has been done with other definitions of activity and such a large amount of self-consistent results have been obtained with the approach used in this paper that we see no reason to treat antitumor drug data in a different manner from other drug data.

Given that one is to use the molar concentration producing a standard response, one is still left with the vexing problem of defining the "standard response". This is the problem central to all pharmacology. From the SAR point

Chart I. Selected Log P Calculations for Tables I, V, and VII

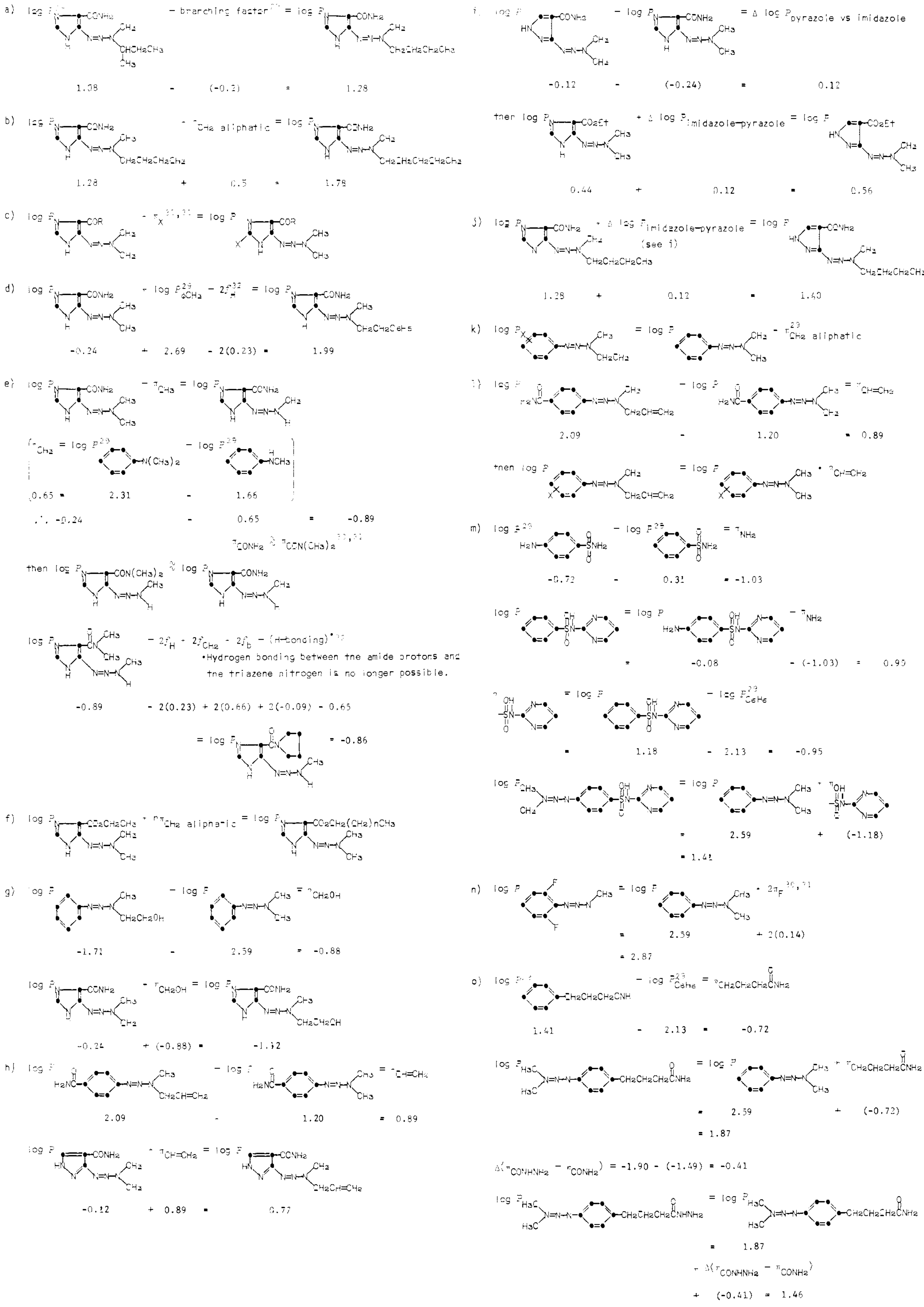
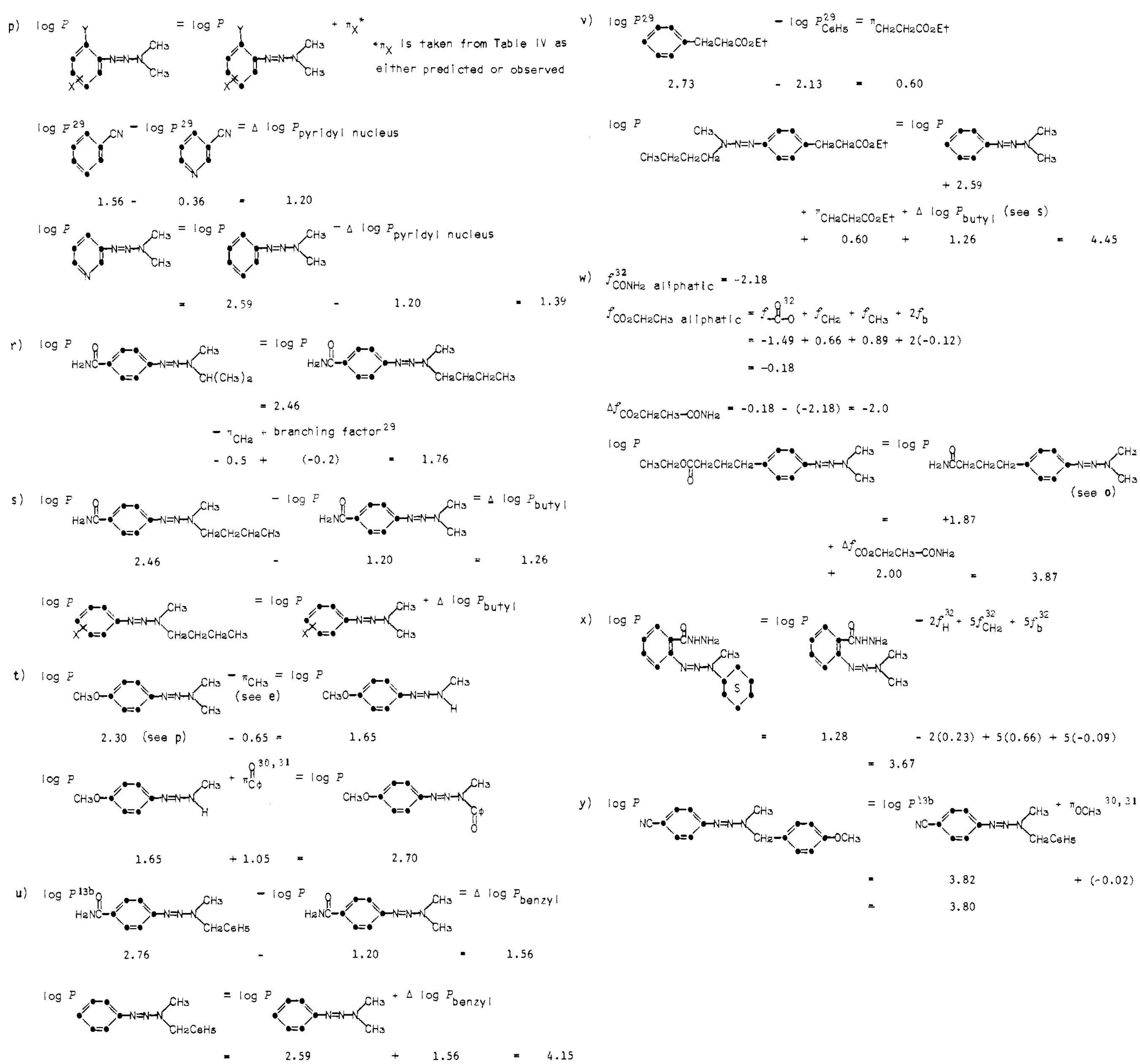


Chart I *(Continued)*



of view, selecting different types of responses will lead to different SAR or QSAR and, in the end, to different drugs. Success in drug research depends more on selecting the proper system for testing and the best end point than on any other factor.

In the present case we have little choice. We have selected a T/C so that we would minimize large extrapolations; that is, if one selects a T/C too high, then many compounds must be dropped because they do not achieve such activity or large extrapolations must be made to estimate the proper concentration.

There is a great amount of scatter in the test results obtained on the triazenes discussed in this paper and, in fact, this is the usual problem with antitumor drugs. We have had to exercise some subjectivity in drawing the "best" line for our relatively small extrapolations. Our largest extrapolation is from a T/C of 120–140.

Errors in estimating the proper T/C will show up in the standard deviations from the regression equation along with other errors. For example, the standard deviation of eq 2 (~0.2) suggests that our QSAR would estimate the molar concentration producing a T/C of 140 within a factor of ±1.6. Despite this rather large error, we believe that the self-consistency among the QSAR of eq 1, 2, 4, and 5 in the estimation of log P_0 shows their value in antitumor research.

The above QSAR lead to a dead end as far as obvious routes for the development of more potent triazenes are concerned. Log P_0 has been firmly established as about 1 so that there is no room for further improvement in potency by manipulation of lipophilic character. Equation 2 indicates that electron-releasing substituents could be used to increase potency; however, these increase the instability of the triazenes.

Equation 6 has been formulated from the work of Kolar

$$\log k_X/k_H = -4.42\ (\pm 0.29)\ \sigma - 0.016\ (\pm 0.13) \qquad (6)$$

$$n = 14;\ r = 0.995;\ s = 0.171$$

and Preussmann[26] who elegantly demonstrated that the rate of hydrolysis of a set of 14 X-phenyldimethyltriazenes correlates with the electronic effect of ring substituents. They found a value of −4.7 for the Hammett ρ in their study. In order to obtain eq 6, we have included three data

Table XII. Summary of Data on New $X{-}C_6H_4N{=}NNCH_3R$

No.	X	R	Mp or bp (mm), °C	Solvent for recrystn	% yield	NCI no.
1	4-SO_2NH_2	CH_3	181-182	Benzene-ethanol	75	157030
2	4-$NHCOCH_3$	CH_3	153-155	Acetone-hexane	98	157031
3	4-CN	CH_3	109.5-111.5	Ethanol	99	157034
4	2-I	CH_3	100-102 (0.17)		76	173095
5	2-CN	CH_3	37-38	Pentane	79	180036
6	2-Cl	CH_2CH_2OH	133-136 (0.15)		90	180040
7	2-CH_2OH	CH_3	127-130 (0.4)		79	180037
8	2-CN	CH_2CH_2OH	51-52.5	Pentane-ether	50	180041
9	4-$NHCONH_2$[a]	CH_3	178.5-180.5 dec	Ethanol-water	51	268492
10	2-COOH	Cyclohexyl	108.5-110	Hexane-benzene	76	173203
11	2-COOH	*n*-Propyl	58-60	Hexane-ether	94	173201
12	2-COOH	*n*-Butyl	58-59.5	Hexane-ether	85	210719
13	2-COOH	*n*-Octyl	46.5-48	Pentane	34	173202
14	3,5-$(COOH)_2$	CH_3	173 dec	Ethanol-water	45	
15	2,4-$(COOH)_2$[b]	CH_3	173 dec	Methanol	84	
16	4-COOH	*n*-Octyl	91-92	Pentane	72	183739
17	2-COOH, 4-Cl	CH_3	140-141.5 dec	Ethanol	67	233879
18	2-COOH, 4-Cl	*n*-Octyl	53-55	Pentane-ether	48	183740
19	2-COOH, 4-NO_2[c]	CH_3	186-187 dec	Me_2SO-methanol	73	233878
20	2-COOH, 4-CN[b]	CH_3	185-186 dec	Methanol-water	70	
21	2-COOH, 4-SO_2NH_2[d]	CH_3	195-197 dec	DMF-water	81	
22	2-COOH, 4-OCH_3	CH_3	134-135	CH_3OH	89.5	233877
23	4-$CONH_2$[e]	Allyl	114-116	Ethanol	65	276372
24	4-$CONH_2$[f]	*n*-Octyl	104.5-105.5	Benzene-hexane	19	276741
25	4-NHCHO[g]	CH_3	105-107	Ethyl acetate-hexane	30	276376
26	4-$NHCONH_2$[a,e]	CH_2CH_3	149-151 dec	Acetone-hexane	19	276375
27	4-$NHCONH_2$[a,e]	Allyl	111-113	Ethyl acetate-hexane	13	279502
28	4-$CONH_2$	OCH_3[h]	118-119.5	Ethyl acetate	73	279831
29	4-$CONH_2$	CH_2CN[h]	85 dec	Acetone	95	279830
30	3-$CONH_2$, 6-OCH_3	CH_3	184-186	Ethanol	74	276374
31	4-$CONH_2$	$NHCH_3$[h]	115-117	Methanol-pentane-ether	85	284696
32	4-$CONH_2$	Diallyl[i]	112.5-114	Ethyl acetate-hexane	61	284695
33	3-$NHCONH_2$[j]	CH_3	165-166 dec	Acetone	40	284697
34	H[f]	*n*-Octyl	153-155 (0.6)		35	284699
35	3,5-$(CN)_2$[k]	CH_3	161.5-163	Ethyl acetate	50	284698

[a] *p*-Nitrophenylurea was obtained by warming KOCN with *p*-nitroaniline in aqueous acetic acid; the aniline was then obtained by catalytic hydrogenation with Pd/C in ethanol. [b] 4-Aminoisophthalic acid was obtained via basic hydrolysis of *N*-acetyl-4-cyanoanthranilic acid which was prepared from *N*-acetyl-4-nitroanthranilic acid [A. Cohen, H. King, and W. Strangeways, *J. Chem. Soc.*, 3236 (1931)] via reduction, diazotization, and treatment with $NaCu(CN)_2$. [c] 5-Nitroanthranilic acid was prepared according to the procedure of E. Baly, W. Tuck, and E. Marsden, *J. Chem. Soc.*, 97, 1494 (1910). [d] 4-Amino-3-carboxybenzenesulfonamide was obtained according to the method of L. Szabo, *Bull. Soc. Chim. Fr.*, 771 (1953). [e] Purified by column chromatography, ethyl acetate/alumina. [f] The appropriate aniline was converted to the diazonium tetrafluoroborate salt and the salt reacted with *N*-methyloctylamine in methanol. [g] *N*-Formyl-4-nitroaniline was obtained by refluxing *p*-nitroaniline in excess formic acid and the desired amino compound was prepared by catalytic hydrogenation with Pd/C in ethanol. [h] The amine precursors were generated from an aqueous mixture of their hydrochlorides and Na_2CO_3. [i] $H_2NCO{-}C_6H_4{-}N{=}N{-}N(CH_2CH{=}CH_2)_2$. [j] Same as footnote *a*, except *m*-nitroaniline was utilized. [k] 5-Nitroisophthalonitrile was prepared from 5-nitroisophthalic acid according to the procedure of M. Kimura and M. Thoma, *Yakugaku Zasshi*, 78, 1401 (1958); the aniline was then obtained by reduction with $SnCl_2 \cdot HCl$, similar to a procedure reported by N. V. Philips-Gloeilamdenfabrieken, Netherlands Patent 6613164 (March 18, 1968); *Chem. Abstr.*, 69, 86660z (1968).

points which they omitted. Equation 6 correlates the half-life of the reaction

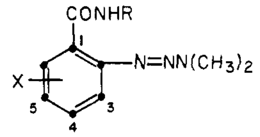
$$X{-}C_6H_4N{=}NN(CH_3)_2 + H_2O \rightarrow X{-}C_6H_4\overset{+}{N}_2 + HN(CH_3)_2$$

Kolar and Preussmann noted that the 4-OCH_3 derivative has a half-life of about 12 min, while the 3-NO_2 congener has a half-life of 3.6×10^5 min. Clearly, a group more electron releasing than OCH_3 would produce a drug too unstable to work with.

In Table IV, containing the inactive compounds, we have included a value of log $1/C$ calculated using eq 2. All of these predicted values fall within the limits covered by the analogues in Table I. The majority, 23 out of 34, were simply too toxic to detect any activity; activity could possibly be found by very careful variation of the dose. A large number of the molecules of Table IV contain ortho substituents or, in some instances, unusual side chains (**6–8** and **15–18**). A number of the unusual ortho substituents (**1**, **2**, **28**, and **34**) were prepared with the hope that some sort of favorable hydrogen-bonding interaction with the triazene moiety might result in increased activity; no such effect could be uncovered. In fact, the negative coefficient with the MR-2,6 term in eq 2 indicates that all ortho substitution is detrimental to activity. There are no interesting examples in Table I where activity turns out to be much greater than eq 2 would predict. Such cases would constitute points of departure for new synthetic efforts.

Many of the most poorly predicted molecules of Table I are those of lowest activity (**61–64**), again indicating the difficulties involved in testing.

Although the overall results of eq 2 for phenyltriazenes are rather disappointing to the theorist trying to account for a large fraction of the variation in log $1/C$ with a sharp mathematical model, there are some generally useful conclusions. Ideal lipophilicity for triazenes acting against leukemia seems to be well established. There is no special potency associated with heterocyclic rings. Electron release via through resonance increases potency. Large R groups are the most important determinate of activity (see Table II), depressing it greatly. When R = *tert*-butyl, all activity is lost (Table IV, **32**). No positive ortho effects were uncovered. There seems to be no obvious way of increasing potency; however, one way in which better drugs could be

Table XIII. Summary of Data on 

No.	X	R	Mp or bp (mm), °C	Solvent for recrystn	% yield	NCI no.
36	H	$NHCSNH_2$	188-189 dec	Chloroform-ethanol	55	258834
37	H	$NHCOCH_3$	185.5-187 dec	Chloroform-hexane	88	260617
38	H	$NHCOCH_2CN$	190 dec	Acetone	90	261061
39	H	OCH_3	60-62	Ether-hexane	90	260618
40	H	CH_2CF_3	87-88	Ethanol-ether	86	260619
41	H	CH_3	69-71	Ether-hexane	76	261059
42	H	CH_2CH_3	81-83	Ether-hexane	63	261060
43	H	CH_2CN[a]	106.5-109	Chloroform-hexane	76	263461
44	H	CH_2CONH_2[a]	189-190.5	Chloroform-hexane	39	263462
45	H	CN[b]	143-145 dec	Chloroform-hexane	35	268490
46	5-SO_2NH_2[c]	H	212-214.5	DMF-water	30	258833
47	5-$CONH_2$[c,d]	H	230-232 dec	Ethanol	41	261725
48	5-SCN[c]	H	172-173	DMF-water	69	258832
49	5-Cl[c]	H	189-191	Ethanol	50	143907
50	5-NO_2[c]	H	190 dec	DMF-water	66	143908
51	5-CN[e]	H	178.5-179.5	Methanol-water	86	258831
52	3,5-$(CONH_2)_2$[f]		234-236 dec	Ethanol	45	268495

[a] Amine precursor generated from its hydrochloride salt and NEt_3 in dioxane and added to the reaction mixture in the form of this mixture. [b] Aqueous Na_2NCN was added to the reaction mixture, followed by acidification with dilute HCl. [c] See Table XII for carboxylic acid precursor. [d] Purified with column chromatography, DMF-ethanol/alumina. [e] The anthranilic acid precursor was obtained from anthranilic acid according to the method of R. Pohloudek-Fabini and M. Schuessler, *Pharm. Zentralhalle Dtschl.*, 107, 116 (1968); *Chem. Abstr.*, 68, 1114480 (1968). [f] Also prepared according to method A from 5-aminoisophthalamide.

obtained would be by minimizing toxicity. This possibility is explored in the following paper.[33]

Method

The necessary biological data and substituent constants for eq 2, 4, and 5 are given in Tables I, V, and VII. (See Chart I.) In our initial studies, a T/C (T = survival time of test animal; C = survival time of control animal) of 140 was established by plotting concentration of drug (moles per kilogram) vs. survival time for compounds showing a T/C of at least 125. Compounds were not included unless activity was obtained at at least two different concentrations. A line was drawn between these points and the origin. After the formulation of an equation from these initial studies, it was observed that compounds showing an initial T/C of as low as 112 with a higher T/C, generally in the region of 120, gave extrapolated values of T/C of 140 which were moderately well fit by our initial equation. A few such examples (**22**, **27**, **46**, **56**, **58**, and **64**) are included in Table I.

Experimental log *P* values for a variety of triazenes are given in Table IX. It was necessary to calculate some log *P* values using data in Table X as starting points.

In the case of simple substituted phenyltriazenes it was possible to use eq 7 for estimating[27] the appropriate

$$\pi_{\text{estimate}} = 0.977\ (\pm 0.11)\ \pi_{\text{aromatic}} + 0.636\ (\pm 0.33)\ \sigma - 0.099\ (\pm 0.14) \quad (7)$$

$n = 11; r = 0.991; s = 0.139$

substituent contribution to log *P*. The development of eq 7 is given in Table XI.

Preparation of Phenyltriazenes. Method A. The compounds of Table XII were prepared according to the procedure of Rondestvedt and Davis[3] by treating the proper aryldiazonium salt with the appropriate amine. The appropriately substituted anilines were commercially available unless otherwise stated in Table XII.

Method B. All of the compounds in Table XIII were made from the corresponding carboxytriazenes using dioxane as a solvent (except compounds **47** and **52** in which DMF was used), according to the method of Lin et al.[13] In this method the carboxyl is converted to the mixed anhydride by treating its triethylammonium salt with ethyl chloroformate. The mixed anhydride is then treated with the appropriate amino compound to produce the substituted triazene in the indicated yield. Concentrated ammonia (28%) was found to be satisfactory for the preparation of compounds **46**–**52**.

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Antitumor 1-(X-Aryl)-3,3-dialkyltriazenes. 2. On the Role of Correlation Analysis in Decision Making in Drug Modification. Toxicity Quantitative Structure–Activity Relationships of 1-(X-Phenyl)-3,3-dialkyltriazenes in Mice[1]

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A series of 11 triazenes ($X—C_6H_4N{=}NNRCH_3$) was characterized for toxicity in mice (LD_{50}). The quantitative structure–activity relationship (QSAR) obtained for toxicity was compared with the QSAR for antitumor activity. The close correspondence of the two QSAR leaves essentially no means for the synthesis of more potent, less toxic triazenes.

In the previous paper in this series,[2] eq 1 was formulated

$$\log 1/C = 0.10 \log P - 0.04 (\log P)^2 - 0.31\ \Sigma\sigma^+ - 0.18\ \text{MR-2,6} + 0.39\ E_s\text{-R} + 4.12 \quad (1)$$

$$n = 61;\ r = 0.836;\ s = 0.191;\ \log P_0 = 1.18$$

for the antitumor activity of $X–C_6H_4N{=}N–NR_1R_2$ acting against L1210 leukemia in mice. C in eq 1 is the concentration (moles per kilogram) producing a T/C of 140, MR-2,6 is the sum of molar refractivity of substituents in the two ortho positions, and E_s-R is the Taft steric parameter for the larger of R_1 and R_2. The $\log P_0$ of 1.18 sets the upper limit of potency which can be obtained in this series by manipulation of the lipophilic/hydrophilic balance. Essentially the same $\log P_0$ was found for imidazolyl- and pyrazolyltriazenes.[2] The only advantage to be gained from the MR-2,6 term is obtained when both ortho positions are unsubstituted. For practical purposes, the E_s-R term limits one to the $N(CH_3)_2$ since $NHCH_3$ compounds are so unstable.

At first glance, one presumes that more activity could be obtained by introducing more electron-releasing groups (large negative $\Sigma\sigma^+$); however, the QSAR of eq 2 effectively

$$\log k_X/k_H = -4.42\ \sigma - 0.16 \quad (2)$$

$$n = 14;\ r = 0.995;\ s = 0.171$$

limits this avenue. Equation 2 correlates the rate of hydrolysis of phenyltriazenes.[2] This is so enormously promoted by electron-releasing groups that it is not possible in practice to go beyond the 4-OCH_3 (half-life = 12 min) in the use of electron-releasing functions. Attempts to increase potency through steric and/or hydrogen-bonding effects of ortho substituents have reached

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