It is seen that there is good correlation between the localization energies and the experimental values. All compounds having an O.L.E. of less than 2.00β for their K region are carcinogenic unless they have an L region with a P.L.E. of less than approximately 2.9β. The correlation is considerably better than that obtained using *ortho* and *para* localization energies obtained from the mathematically more complicated Hückel molecular orbital method. The quantitative correlation is, in fact, somewhat better than that obtained in many of the more sophisticated calculations.[3,4] This is quite possibly due to the fact that Dewar's approximation is more closely related to those reaction indices which involve charge-transfer mechanisms[19] than it is to true localization energies.

When working with substituted aromatic systems and heteroaromatic systems, the approximations which were used in the derivation of the Coulson, Longuet-Higgins perturbation theory are no longer valid; therefore, as might be expected, correlations based on Dewar's approximate localization energies are no longer as close as in the nonsubstituted aromatic cases. Certain generalizations can be made, however.

Substitution by methyl groups will result in compounds which are at least as active as the parent compound and usually more active, due to hyperconjugation, unless such substitution sterically blocks the K region. Activity is particularly enhanced if the L region is blocked. For example, 1,2-benzanthracene is inactive under the cited conditions due to a highly reactive L region. 9,10-Dimethyl-1,2-benzanthracene, on the other hand, has high activity.

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Cyano Analogs of Phenolic Nitrogen Mustards[1]

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Cyano derivatives of both antimetabolites and alkylating agents have recently shown promise as anticancer agents in preliminary studies. For example, 6-(cyanomethylthio)purine exhibited marked inhibition of Adenocarcinoma 755[3] in mice, and *p*-[bis(2-chloroethyl)]aminobenzylidenemalononitrile and related compounds were active against Dunning leukemia[4] in rats. In addition, a series of bicyclic nitriles and related compounds were recently prepared by the Diels–Alder reaction for evaluation as antitumor agents.[5]

Work in this laboratory[6] has utilized the Mannich reaction as a route to phenolic nitrogen mustards. One of these compounds, 2,5-bis{[bis(2-chloroethyl)amino]-methyl}hydroquinone, was effective against Carcinoma 755, Dunning leukemia, Lymphoma 8, Ehrlich EF, Yoshida hepatoma, and Walker 256 ascites in preliminary tests and is currently being evaluated clinically.[7] Just recently Kuehne and Konopka[8] have shown that related phenolic Mannich bases devoid of 2-chloroethylamino groups also possess antitumor activity.

In view of these results and the unusually low toxicity of the hydroquinone mustard in comparison with other agents of this type, the synthesis of cyano analogs of phenolic nitrogen mustards was undertaken. Condensation of hydroquinone with formaldehyde and bis(2-cyanoethyl)amine in the required proportions in refluxing methanol gave a 33% yield of the desired 2,5-bis-{[bis(2-cyanoethyl)amino]methyl}hydroquinone (XII, Table I). Somewhat higher yields were obtained in the synthesis of analogous disubstituted compounds from resorcinol (74%) and 2-methylresorcinol (83%) by a similar procedure at 5°. Efforts to prepare a monosubstitution derivative of resorcinol by condensation of equimolar proportions of the reactants led to the isolation of only the same disubstituted product. The infrared spectrum showed bands at 11.4 and 11.6 μ, which is characteristic[9] of isolated ring hydrogens. In view of this and the presence of a moderate band at 13.4 μ, which is not generally shown by aromatic compounds with two adjacent hydrogens, it was assumed that the [bis(2-cyanoethyl)amino]methyl groups entered the 4 and 6 positions of resorcinol. This assignment was consistent with the observation that two substituents were introduced into 2-methylresorcinol while only a monosubstituted derivative was obtained from 4-chlororesorcinol, even when the reactant ratio was that required for disubstitution.

As indicated in Table I, several *ortho*-monosubstituted [bis(2-cyanoethyl)amino]methyl derivatives of monohydric phenols were also prepared. Under the conditions used in the condensation of 4-substituted phenols with two free *ortho* positions, only a monosubstituted product was isolated even when sufficient amine and formaldehyde were present to give a disubstituted product. Further treatment of 2-{[bis(2-cyanoethyl)-amino]methyl}-4-chlorophenol (I) with formaldehyde and bis(2-cyanoethyl)amine resulted in the recovery of the original Mannich base in high yield.

A 2,6-disubstituted product XV was obtained readily and in good yield, however, by the reaction of 2,6-bis-(chloromethyl)-4-chlorophenol with excess bis(2-cyanoethyl)amine in benzene at 65°. In a similar manner 2-{[bis(2-cyanoethyl)amino]methyl}-4,6-dichlorophenol (IX) was prepared in 87% yield from 2-chloromethyl-4,6-dichlorophenol. The compound was also prepared directly from 2,4-dichlorophenol by the Mannich reaction but in lower yield.

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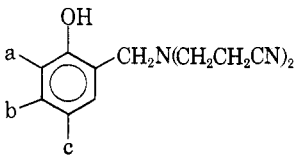
TABLE I

ortho-SUBSTITUTED {[BIS(2-CYANOETHYL)AMINO]METHYL}PHENOLS



	Substituent			M.p., °C.	Yield, %	Molecular formula	Carbon, %		Hydrogen, %		Neut. equiv.	
	a	b	c				Calcd.	Found	Calcd.	Found	Calcd.	Found
I	H	H	Cl	123–124	87[a]	$C_{13}H_{14}ClN_3O$	59.20	58.85	5.35	5.40	263.7	263.0
II	H	H	Br	138–139	50[a]	$C_{13}H_{14}BrN_3O$	50.66	50.86	4.58	4.63	308.2	311.3
III	H	H	CH_3	83–84	34[a]	$C_{14}H_{17}N_3O$	69.11	69.03	7.06	6.96	243.3	246.9
IV	H	H	OC_4H_9	75–76	41[b]	$C_{17}H_{23}N_3O_2$	67.75	67.94	7.69	7.71	301.4	304.3
V	H	CH_3	CH_3	82–83	77[a]	$C_{15}H_{19}N_3O$	70.01	70.00	7.44	7.62	257.3	257.2
VI	Cl	H	$C(CH_3)_3$	105–106	69[a]	$C_{17}H_{22}ClN_3O$	63.84	63.85	6.93	6.75	319.8	321.7
VII	CH_3	H	CH_3	108–109	55[a]	$C_{15}H_{19}N_3O$[c]	70.01	69.84	7.44	7.62	257.3	258.4
VIII	CH_3	H	Cl	115–116	40[a]	$C_{14}H_{13}ClN_3O$	60.53	60.41	5.81	5.73	277.8	281.1
IX	Cl	H	Cl	87–88	87[a]	$C_{13}H_{13}Cl_2N_3O$	52.36	52.27	4.39	4.38	298.2	295.1
X	H	CH_3	OH	116–117	16[d]	$C_{14}H_{17}N_3O_2$	64.84	64.78	6.61	6.67	...	...
XI	H	OH	Cl	146–147	95[a]	$C_{13}H_{14}ClN_3O_2$	55.82	55.99	5.04	5.45	279.7	286.0
XII	H	e	OH	198–199	33[f]	$C_{20}H_{24}N_6O_2$[g]	63.14	62.89	6.36	6.40	...	...
XIII	H	OH	e	171–172	74[f]	$C_{20}H_{24}N_6O_2$[h]	63.14	63.22	6.36	6.70	...	...
XIV	CH_3	OH	e	191–192	83[f]	$C_{21}H_{26}N_6O_2$	63.92	63.95	6.64	6.74	197.2	200.6

[a] Recrystallized from 95% ethanol. [b] Recrystallized from propanol-1. [c] *Anal.* Calcd.: N, 16.33. Found: N, 16.31. [d] Recrystallized from methanol. [e] b or c = $-CH_2N(CH_2CH_2CN)_2$. [f] Recrystallized by dissolving in hot dimethylformamide and then adding methanol. [g] *Anal.* Calcd.: N, 22.09. Found: N, 21.85. [h] *Anal.* Calcd.: N, 22.09. Found: N, 21.86.

Representative examples of N,N-bis(2-cyanoethyl)-aminomethylphenols described in this paper were screened under the direction of the Cancer Chemotherapy National Service Center, for anticancer activity in mice in doses up to 200 mg./kg. and in cell culture tests. All compounds tested were nontoxic at these levels. Against Walker 256, slight activity of the order of 25 to 50% inhibition was shown by compounds II and XIII in Table I. Otherwise no significant inhibition was shown by compounds I–IX, XII, XIII, or XV against Sarcoma 180, Solid Friend Virus Leukemia, Leukemia 1210, Walker 256, or in cell culture tests. Compound XVI showed slight activity against Adenocarcinoma 755, but was inactive against Sarcoma 180 and Leukemia 1210.

Experimental[10]

2,5-Bis{[bis(2-cyanoethyl)amino]methyl}hydroquinone (XII).—Bis(2-cyanoethyl)amine (24.6 g., 0.2 mole) in 50 ml. of methanol was added dropwise to an ice-cooled, stirred solution of 30 ml. of 37% aqueous formaldehyde (0.4 mole) in 30 ml. of methanol. Hydroquinone (11 g., 0.1 mole) was added and the solution stirred for 10 min. on an ice bath. After the solution was refluxed for 4 hr., the solvents were removed by evaporation under the hood. The crude solid was dissolved in 20 ml. of hot dimethylformamide and sufficient methanol was added to the cooled solution to induce crystallization. The product (12.5 g., 33%, m.p. 180–185°) melted at 198–199° after four recrystallizations from dimethylformamide–ethanol.

2,6-Bis{[bis(2-cyanoethyl)amino]methyl}-4-chlorophenol (XV).—To a solution of 2,6-bis(chloromethyl)-4-chlorophenol (22.6 g., 0.10 mole) in 600 ml. of benzene was added 50 ml. of bis(2-cyanoethyl)amine (0.40 mole) with shaking. The solution was warmed at 65° for 15 min. and then cooled. The resulting solid, bis(2-cyanoethyl)amine hydrochloride (32.8 g., m.p. 144–147°), was removed by filtration. Evaporation of benzene from the filtrate gave 25.2 g. (63.2%) of solid; m.p. 123–124° after 5 recrystallizations from 95% ethanol. A 20° depression of melting point was observed for a mixture of this product and the monosubstituted product I, which also melted at 123–124°.

(10) All melting points are uncorrected. This work was completed before the requirement for melting point corrections went into effect for this journal.

Anal. Calcd. for $C_{20}H_{23}ClN_6O$: C, 60.22; H, 5.81; N, 21.07; neut. equiv., 199.5. Found: C, 60.62; H, 5.65; N, 20.63; neut. equiv., 202.1.

1-{[Bis(2-cyanoethyl)amino]methyl}-2-naphthol (XVI).—This compound was prepared in 74% yield from 2-naphthol by the Mannich reaction; m.p. 101–102° after recrystallization from ethanol.

Anal. Calcd. for $C_{17}H_{17}N_3O$: neut. equiv., 279.3. Found: neut. equiv., 280.6.

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Alkyl and Aryl Thiolsulfonates

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The first thiolsulfonate was reported in 1840 and in 1949 Small, *et al.*, demonstrated antimicrobial activity of thiolsulfonates.[1,2] The structural relationship to allicin (allylthiolsulfinate), the antibiotic found in garlic, stimulated further interest in thiolsulfonates.[3–5] Grishko and Gur'yanova discussed whether the RSO_2- or the RS- is the active moiety, concluding that the former could be substituted without affecting activity while the RS- could not and was responsible for the inhibition towards microorganisms.[6] In order to investi-

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