This dihydrochloride (1 g.) was dissolved with 20 ml. of anhydrous methanol, and the theoretical amount of silver carbonate was added slowly with stirring and cooling. Stirring was continued for 15 min., and the silver chloride was filtered. The methanol solution was evaporated under reduced pressure. The residue (VIII) was a white crystalline powder (0.75 g., 95%).

Anal. Calcd. for $C_{10}H_{20}Cl_2N_2O_2$: C, 44.29; H, 7.43; Cl, 26.15; N, 10.32. Found: C, 44.2; H, 7.6; Cl, 26.2; N, 10.21.

VIII dihydrochloride was thin layer chromatographed with cellulose powder, using 1-butanol–acetic acid–water (60:20:20) as a solvent. The chromatogram, developed with a 2% solution of ninhydrin, showed a single orange-red spot (R_f 0.67). The base showed a single violet spot (R_f 0.64).

Precipitation of the Reineckates of Valine, Phenylalanine, Ornithine, Hydroxyproline, and Aspartic Acid. Isolation of the Amino Acids from their Respective Salts Using a Cation-Exchange Resin.—These amino acids were precipitated as their reineckates from their respective aqueous solutions, acidified with HCl (pH 1–2), by the addition of the theoretical amount of 5% aqueous ammonium reineckate solution. These salts were all crystalline but without definite melting points.

The amino acids were isolated from their salts almost quantitatively. First, the salts were dissolved in acetone and diluted with a double volume of water. Then these solutions were percolated through 1.5 equiv. of Amberlite IR 120 (100–200 mesh). The resin was washed by percolation with water until the red color due to reinecke acid disappeared. The amino acid adsorbed by the resin was eluted with 5–10% hydrochloric acid recovered as the pure hydrochloride salt by concentration of the eluate.

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Further Investigations of Heterocyclic Alkylating Agents[1]

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The exceptional antitumor and mutagenic activities displayed by a quinacrine derivative of a monofunctional nitrogen mustard, 2-methoxy-6-chloro-9-[3-(ethyl-2-chloroethyl)aminopropylamino]acridine, led to the synthesis of 50 additional mono- and difunctional analogs of acridine, quinoline, and quinazoline. The acridine nucleus was found to exert a pronounced activating influence on the nitrogen mustard moiety. On a molar basis, the "half-mustard" 2-methoxy-9-[3-(ethyl-2-chloroethyl)aminopropylamino]acridine dihydrochloride was considerably more effective against the Ehrlich ascites tumor than methylbis(2-chloroethyl)amine hydrochloride; the corresponding bis analog was even more potent. Substitution of a 6-chloro group into 2-methoxyacridine decreased the molar activities of the mono and bis mustards. Several monofunctional nitrogen mustards of quinazoline and quinoline displayed moderate antitumor activity, but only at high molar dosages; other closely related analogs were inactive. The relationships between the chemical structures and antitumor activities of the compounds are presented.

From our earlier work[2–4] it was evident that the unusual antitumor activity of certain monofunctional nitrogen mustards was determined by the chemical structure of the heterocyclic nucleus that was attached through a side chain to the mono-2-chloroethylamino group. The first nitrogen "half-mustard" that displayed pronounced activity in prolonging the survival time of mice bearing several varieties of ascites tumors[2] and exhibited an extraordinary mutagenic capability in *Drosophila*[5] was 2-methoxy-6-chloro-9-[3-(ethyl-2-chloroethyl)aminopropylamino]acridine dihydrochloride.[4] On the other hand, the partial acridine structures, 7-chloro- and 6-methoxy-4-[3-(ethyl-2-chloroethyl)aminopropylamino]quinoline dihydrochloride,[2] and the secondary amine, 2-methoxy-6-chloro-9-[2-(2-chloroethyl)aminoethylamino]acridine dihydrochloride, showed no antitumor activity. Since their corresponding bis mustards were highly effective, it is apparent that both the heterocyclic nucleus and the presence of an alkyl group on the nitrogen containing the 2-chloroethyl group are of critical importance in activating the monofunctional mustard grouping.

It appeared worthwhile to determine whether the 2-methoxy or the 6-chloro group on the acridine nucleus played a significant role in this activation and whether any modifications of simpler heterocyclic nuclei, such as quinoline and quinazoline, would impart enhanced physiological activity to the "half-mustards." The effects of attachment of the nitrogen-mustard moiety at the 4-position of variously substituted quinolines, at the 2-position of quinoline and lepidine, at the 4-position of quinazoline and 6-chloroquinazoline, and at the 8-position of 6-methoxyquinoline, as well as the presence of an N-alkyl substituent on the 4-quinolyl nitrogen, were investigated both in the mono and bis forms, as shown in Table I. The letters A to X in the first column of Tables I and II represent the heterocyclic group Ar in the formula Ar-( )-N(R)CH_2CH_2X at the top of Table I. The heterocyclic structures corresponding to these letters are as follows.

Most of the tertiary amino side chains were added stepwise to the nucleus by condensing 4-chloroquinoline with an alkylaminoethanol, chlorinating, and condensing with diethanolamine, or an analog, to give the mustard precursor. However, when the readily crystallized nitrate salts[6] of the first hydroxy intermediate,

(1) Supported by research Grants CA 02975 and CA 06927 from the National Cancer Institute, National Institutes of Health, U. S. Public Health Service.

(2) H. J. Creech, E. Breuninger, R. F. Hankwitz, Jr., G. Polsky, and M. L. Wilson, *Cancer Res.*, **20**, 471 (1960).

(3) R. M. Peck, R. K. Preston, and H. J. Creech, *J. Am. Chem. Soc.*, **81**, 3984 (1959).

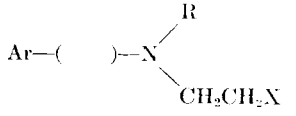
(4) R. M. Peck, R. K. Preston, and H. J. Creech, *J. Org. Chem.*, **26**, 3409 (1961).

(5) E. A. Carlson and I. I. Oster, *Genetics*, **47**, 561 (1962).

(6) R. M. Peck, *J. Org. Chem.*, **28**, 1998 (1963).

TABLE I

ANALYTICAL INFORMATION AND ANTITUMOR ACTIVITY



Compd. No.	Side chain —( )—	R	X	Salt	Yield, %	M.p., °C.	Calcd. % C	H	N	Halogen	Found,[a] % C	H	N	Halogen	Antitumor activity[c] Range, μmoles/kg.	Degree
A-1	$NHCH_2CH_2$	CH_3	Cl	$2HCl \cdot 0.5H_2O$	60	236.5–237.5	49.62	5.26	9.13	30.82	49.67	5.53	9.04	31.02	12–32	2.3
A-2	$NHCH_2CH_2$	CH_3	OH	...	27	135–136.5	63.43	6.17	11.67		63.95	6.36	11.75			
A-3	$NHCH(CH_3)(CH_2)_3CH_3$	C_2H_5	Cl	$2HCl \cdot H_2O$	57	148–152	52.60	6.33	8.00	27.00	52.80	6.41	8.55	26.79	2–6	2.3
A-4	$NHCH(CH_2)_3$	C_2H_5	OH	$2HCl \cdot 0.5H_2O$[b]	21	243–245 dec.	55.50	6.69	8.44	21.35	55.60	6.73	8.42	21.67		
B-1	$NH(CH_2)_3$	C_2H_5	Cl	2HCl	68	229.5–230	56.70	6.35	9.45	23.91	56.02	6.36	9.44	23.67	0.6–2.5	2.6
B-2	$NH(CH_2)_3$	C_2H_5	OH	2HCl	40	247.5–248.5	59.15	6.86	9.86	16.63	58.73	6.89	9.24	16.75		
B-3	$NH(CH_2)_3$	C_2H_4Cl	Cl	$2HCl \cdot 0.5H_2O$	66	219.5–221	51.65	5.78	8.66	29.05	52.01	5.88	8.45	29.03	0.15–1	2.7
B-4	$NH(CH_2)_3$	C_2H_4OH	OH	2HCl	20	237–238	57.01	6.61	9.50	16.03	56.09	6.98	9.51	16.30		
B-5	$NHCH_2CH_2$	C_2H_4Cl	Cl	2HCl	59	235–237	51.63	5.42	9.03	30.48	51.22	5.55	9.00	30.47	2–6	2.5
B-6	$NHCH_2CH_2$	C_2H_4OH	OH	2HCl	58	236–236.5	56.08	6.35	9.81	16.56	56.08	6.57	9.84	16.65		
C-1	$NH(CH_2)_3$	C_2H_5	Cl	2HCl	23	238–240 dec.	58.00	6.33	10.13	25.65	58.14	6.70	10.05	25.73	1.5–4	2.4
C-2	$NH(CH_2)_3$	C_2H_5	OH	$2HCl \cdot 0.5H_2O$	58	216–217	59.35	6.97	10.37	17.50	59.00	7.00	10.20	17.71		
C-3	$NH(CH_2)_3$	C_2H_4Cl	Cl	2HCl	70	225–228	53.44	5.61	9.36	31.53	53.31	5.74	9.40	31.34	0.2–1	2.6
C-4	$NH(CH_2)_3$	C_2H_4OH	OH	2HCl	81	218–219 dec.	58.20	6.60	10.18	17.20	57.40	6.98	10.25	17.44		
D-1	$N(CH_3)(CH_2CH_3)CH_3$	CH_3	Cl	2HCl	52	232–233.5 dec.	56.40	6.98	10.37	26.28	55.74	7.25	10.79	25.87	10–40	1.0
D-2	$N(CH_3)(CH_2CH_2CH_3)CH_3$	CH_3	OH	$2HCl \cdot 0.5H_2O$	40	241–243 dec.	57.76	7.65	10.62	17.92	57.32	7.88	10.63	18.29		
D-3	$N(CH_2CH_2CH_3)(CH_2CH_3)CH_3$	C_2H_4Cl	Cl	$2HCl \cdot 0.5H_2O$	48	200–201 dec.	51.90	6.54	9.08	30.68	51.41	6.88	8.48	30.78	12–40	2.4
D-4	$N(CH_2CH_2CH_3)(CH_2CH_3)CH_3$	C_2H_4OH	OH	2HCl	44	226.5–228.5	57.61	7.51	10.09	17.02	57.58	8.03	9.40	16.97		
D-5	$NH(CH_2)_3$	C_2H_4Cl	Cl	$2HCl \cdot 0.5H_2O$	75	206–207	51.98	6.54	8.85	30.70	51.27	6.72	8.98	31.06	1.2–6	2.6
D-6	$NH(CH_2)_3$	C_2H_4OH	OH	$2HCl \cdot 0.5H_2O$	32	214–215	56.24	7.59	9.88	16.68	56.70	7.67	10.08	17.07		

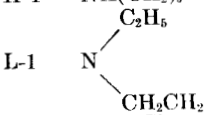
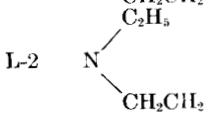
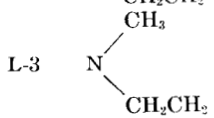
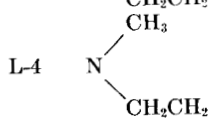
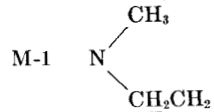
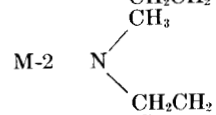
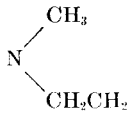
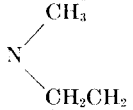
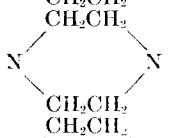
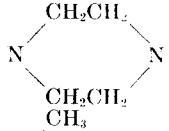
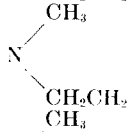
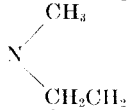
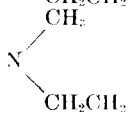
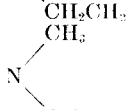
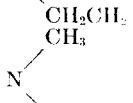
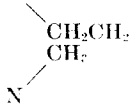
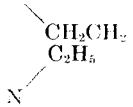
E-1	CH_2CH_2	C_2H_4Cl	Cl	$HCl \cdot Cl^-$	49	189–193 dec.	50.90	5.94	9.89	33.37	51.39	6.10	9.75	33.34	10–40	1.4
E-2	CH_2CH_2	C_2H_4OH	OH	$HCl \cdot Cl^-$	20	245.5–247	55.70	7.02	10.81	18.25	54.88	7.36	10.51	19.87		
F-1	$NH(CH_2)_3$	C_2H_5	Cl	2HCl	55	203–204	45.02	5.54	14.00	35.45	45.17	5.55	13.73	35.34	40–70	2.3
F-2	$NH(CH_2)_3$	C_2H_5	OH	2HCl	43	189–190	47.19	6.07	14.68	27.86	47.25	6.58	14.68	27.74		
F-3	$NHCH_2CH_2$	C_2H_4Cl	Cl	2HCl	89	212–213	39.98	4.55	13.32	42.15	40.12	4.54	13.54	42.05	10–60	2.5
F-4	$NHCH_2CH_2$	C_2H_4OH	OH	...	52	163.8–164.2	54.10	6.61	18.03	11.41	54.36	6.25	17.85	12.18		
G-1	$NHCH_2CH_2$	C_2H_4Cl	Cl	2HCl	90	206–209	43.54	5.22	14.51	36.73	43.29	5.56	14.64	36.44	8–32	2.6
G-2	$NHCH_2CH_2$	C_2H_4OH	OH	2HCl	50	175–177	48.14	6.35	16.04	20.30	47.74	6.76	15.77	21.18		
H-1	$NHCH_2CH_2$	C_2H_5	Cl	$2HCl \cdot H_2O$	68	142–145 dec.	48.15	6.58	10.52	26.69	47.95	7.07	9.57	25.09	40–100	2.0
H-2	$NHCH_2CH_2$	C_2H_5	OH	$2HCl \cdot 0.5H_2O$	42	182–184 dec.	51.75	7.06	11.32	19.12	51.55	7.00	11.40	19.05		
H-3	$NHCH_2CH_2$	C_2H_4Cl	Cl	$2HCl \cdot 0.5H_2O$	18	178–181 dec.	45.35	5.70	9.91	33.46	45.86	5.56	10.13	31.84	15–40	2.6
H-4	$NHCH_2CH_2$	C_2H_4OH	OH	2HCl	7.5	199–201 dec.	50.80	6.66	11.10	18.75	50.01	6.65	11.33	19.40		
I-1	$NH(CH_2)_3$	C_2H_5	Cl	2HCl	37	222–223.5	53.98	7.16	9.93	25.18	53.95	7.49	9.91	22.15	4–20	1.1
I-2	$NH(CH_2)_3$	C_2H_5	OH	2HCl	44	237–238.5	56.45	7.74	10.39	17.52	55.59	7.93	10.32	18.42		
J-1	$NHCH_2CH_2$	CH_3	Cl	$2HCl \cdot H_2O$	74	238–239	55.76	6.08	9.76	24.69	55.83	6.21	9.65	24.79	40–60	2.4
J-2	$NHCH_2CH_2$	CH_3	OH	$\cdot 0.5H_2O$	76	215–217	59.58	6.50	10.43	17.59	59.38	6.72	10.11	17.80		
J-3	$NH(CH_2)_3$	C_2H_5	Cl	$HCl \cdot 0.5H_2O$	97	195–197	58.76	6.49	9.35	23.66	58.25	6.77	9.24	23.69	10–15	2.0
J-4	$NH(CH_2)_3$	C_2H_5	OH	...	53	132–133	75.61	7.79	12.02		75.74	7.88	11.81			
K-1	$NHCH_2CH_2$	CH_3	Cl	$2HCl \cdot H_2O$	97	208–210	48.07	4.84	8.41	35.48	48.25	4.98	8.47	35.39	100–150	2.3
K-2	$NHCH_2CH_2$	CH_3	OH	$\cdot 0.75H_2O$	62	106–108	59.51	5.62	10.41	17.57	59.46	5.68	10.41	17.70		
K-3	$NH(CH_2)_3$	C_2H_5	Cl	$2HCl \cdot H_2O$	72	207–208	50.07	5.35	7.96	33.59	50.43	5.84	7.94	33.14	18–40	2.1
K-4	$NH(CH_2)_3$	C_2H_5	OH	$HCl \cdot 0.5H_2O$	25	246–248	52.84	5.64	8.40	28.36	52.72	5.93	8.33	28.37		
L-1		C_2H_4Cl	Cl	2HCl	81	185.5–190 dec.	42.32	4.82	8.71	44.15	42.60	5.26	8.67	44.19	24–80	2.4
L-2		C_2H_4OH	OH	2HCl	91	170–175 dec.	45.85	5.66	9.44	31.87	46.59	5.84	9.51	31.35		
L-3		C_2H_4Cl	Cl	2HCl	50	177–179	41.08	4.52	8.98	45.44	41.89	4.95	8.77	44.75	10–80	2.7
L-4		C_2H_4OH	OH	2HCl	63	171–172.5	44.58	5.38	9.75	32.90	45.05	5.87	10.11	32.98		
L-5	$NHCH_2CH_2$	C_2H_4Cl	Cl	$2HCl \cdot H_2O$	76	202–204	38.16	4.48	8.89	45.10	38.45	4.64	9.20	44.80	20–125	2.5
L-6	$NHCH_2CH_2$	C_2H_4OH	OH	...	64	120.5–121.5	52.32	5.56	12.21	20.61	52.74	5.65	12.25	21.72		
M-1		C_2H_4Cl	Cl	2HCl	85	206.5–209	42.34	4.81	8.71	44.14	42.61	4.99	8.74	43.48	20–60	2.6
M-2		C_2H_4OH	OH	2HCl	80	175–177	45.84	5.67	9.44	31.85	45.30	5.90	9.63	31.34		
M-3	$NH(CH_2)_3$	C_2H_5	Cl	2HCl	77	203.5–206	45.64	5.40	9.38	39.59	45.39	5.12	9.41	40.06	50–60	1.9
M-4	$NH(CH_2)_2$	C_2H_5	OH	2HCl	82	202–204.5	47.54	5.87	9.77	33.13	44.71	6.27	9.73	33.23		
M-5	$NH(CH_2)_3$	C_2H_4Cl	Cl	2HCl	72	204–206	42.34	4.81	8.76	44.14	42.53	4.98	8.85	43.71	6–30	2.6
M-6	$NH(CH_2)_3$	C_2H_4OH	OH	2HCl	79	194–196	45.85	5.67	9.44	31.84	46.60	6.23	9.70	31.59		
N-1	$NH(CH_2)_3$	C_2H_5	Cl	2HCl	69	257–258.5	49.41	6.10	10.17	34.32	49.44	6.13	10.38	34.63	1–6	1.1
N-2	$NH(CH_2)_3$	C_2H_5	OH	...	79	103–104	63.44	7.52	13.06	11.02	63.68	7.73	13.02	11.13		
N-3	$NHCH_2CH_2$	C_2H_5	Cl	2HCl	41	274–274.5	48.14	5.81	10.53	35.53	48.36	6.01	10.41	35.80	18–36	2.2
N-4	$NHCH_2CH_2$	C_2H_5	OH	2HCl	35	280–281	50.47	6.35	11.04	27.94	50.53	6.44	11.30	27.70		

TABLE I (*Continued*)

Compd. No.	Side chain —( )—	R	X	Salt	Yield, %	M.p. °C.	Calcd., % C	H	N	Halogen	Found,[a] % C	H	N	Halogen	Antitumor activity[c] Range, μmoles/kg.	De-gree
O-1		C_2H_4Cl	Cl	2HCl	29	209–201 dec.	45.65	5.39	9.39	39.59	45.82	5.78	9.19	38.65	15–72	2.4
O-2		C_2H_4OH	OH	2HCl	74	168–173	49.76	6.38	10.22	25.87	49.62	6.62	10.46	26.21		
P-1		...	Cl	2HCl	94	263–265 dec.	47.02	5.00	10.97	37.02	46.96	5.19	10.95	36.71	5–60	1.0
P-2		...	OH	2HCl	81	258–259	49.40	5.53	11.52	29.17	49.19	5.69	11.37	29.26		
P-3		CH_3	Cl	2HCl·H_2O	67	136–139 dec.	44.71	5.75	10.42	35.19	45.34	5.92	10.32	33.96	20–120	1.0
P-4		CH_3	OH	2HCl·0.5H_2O	95	183–186	48.00	6.17	11.19	28.32	48.20	6.31	11.33	28.26		
P-5		C_2H_5	Cl	2HCl	37	105–109	48.15	5.81	10.52	35.53	47.70	5.93	10.29	34.92	20–80	1.1
P-6		C_2H_5	OH	2HCl	78	155–159 dec.	50.45	6.35	11.03	27.95	50.10	6.73	10.72	27.41		
P-7		C_2H_4Cl	Cl	2HCl·0.5-C_2H_5OH	67	108–110°	44.71	5.50	9.23	38.92	44.62	5.51	9.34	38.50	18–70	2.7
P-8		C_2H_4OH	OH	2HCl	79	192–194	48.45	6.10	10.59	26.80	48.29	6.28	10.21	27.30		
P-9		C_2H_4Cl	Cl	2HCl	26	197–200	45.65	5.39	9.39	39.59	45.50	5.34	9.52	39.53	20–50	2.3

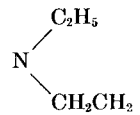
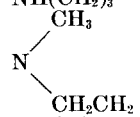
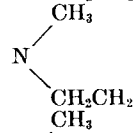
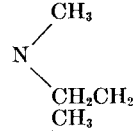
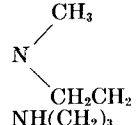
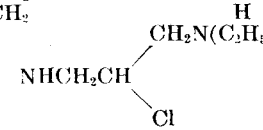
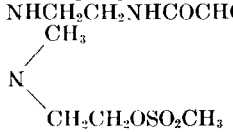
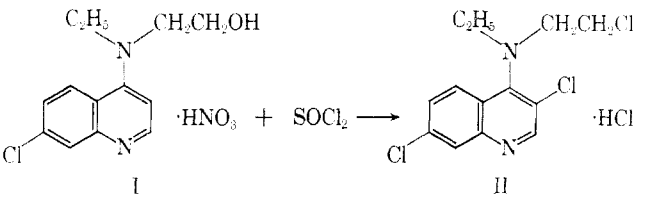
P-10		C_2H_4OH	OH	$2HCl \cdot 0.5H_2O$	70	175–177	48.65	6.49	9.99	25.38	48.53	6.73	10.26	25.22		
P-11	...	CH_3	Cl	HCl	78	186–189 dec.	49.50	4.50	9.62	36.40	49.68	4.39	9.47	36.43	10–100	1.0
Q-1	$NH(CH_2)_6$	C_2H_4Cl	Cl	2HCl	64	92–93	50.97	6.63	8.92	30.09	50.46	6.85	8.96	30.38	0.5–3.5	2.2
Q-2	$NH(CH_2)_6$	C_2H_4OH	OH	...	46	139–140	66.46	8.65	11.63		66.00	8.63	11.23			
Q-3	$NH(CH_2)_3$	CH_3	Cl	$2HCl \cdot 1.5H_2O$	90	*ca.* 100° clear at 180°	47.15	6.67	10.31	26.09	47.05	7.17	10.25	26.08	1–5	1.0
Q-4	$NH(CH_2)_3$	CH_3	OH	...	28	181.5–182.0	53.04	6.96	11.60	19.57	52.66	7.08	11.26	20.26		
R-1	$NH(CH_2)_3$	C_2H_5	Cl	$2HCl \cdot 0.5H_2O$	65	207–209	52.68	7.02	10.84	27.45	52.50	7.13	10.54	28.04	1–4.5	1.0
R-2	$NH(CH_2)_3$	C_2H_5	OH	$2HCl \cdot 0.5H_2O$	72	207–208.5	55.32	7.64	11.38	19.20	55.59	7.72	11.77	19.45		
S-1		CH_3	Cl	$2HCl \cdot 2H_2O$	82	221–222 dec.	47.92	7.04	10.49	26.52	47.56	6.97	10.35	26.20	20–240	1.1
S-2		CH_3	OH	$2HCl \cdot 1.25H_2O$	79	240–242.5	52.09	7.52	11.40	19.25	52.11	7.64	10.95	19.41		
S-3		C_2H_4Cl	Cl	$2HCl \cdot 0.5H_2O$	83	179–181 dec.	48.35	6.21	9.94	33.60	48.84	6.52	9.98	33.11	20–75	2.3
S-4		*C_2H_4OH*	*OH*	*2HBr*	*55*	*226–231*	43.87	5.86	9.03	34.36	43.88	5.90	8.75	34.44		
S-5	$NH(CH_2)_3$	*n*-C_3H_7	Cl	$2HCl \cdot 0.5H_2O$	62	131–133	53.85	7.28	10.47	26.58	53.70	7.30	10.73	26.23	25–50	2 0
S-6	$NH(CH_2)_3$	*n*-C_3H_7	OH	...	30	178–179	57.75	7.81	11.23	18.94	57.35	8.06	11.29	19.07		
S-7	$NH(CH_2)_3$	*i*-C_3H_7	Cl	2HCl	93	228–229	55.04	7.19	10.70	27.08	55.04	7.40	10.73	26.94	15–25	2.2
S-8	$NH(CH_2)_3$	*i*-C_3H_7	OH	...	69	206–207.5	57.75	7.81	11.23	18.94	57.70	7.94	11.13	18.41		
S-9	$NH(CH_2)_3$	C_2H_5	Cl	2HCl	44	202–203	53.90	6.92	11.10	28.08	53.53	6.61	10.79	28.53	36–60	2.1
S-10	$NH(CH_2)_3$	C_2H_5	OH	2HCl	36	203.5–204.5	56.66	7.55	11.66	19.68	56.42	7.79	11.67	20.92		
S-11	$NH(CH_2)_3$	CH_3	Cl	$2HCl \cdot H_2O$	70	115–116	50.20	6.85	10.98	27.79	50.60	7.04	10.88	27.33	100–125	1.9
S-12	$NH(CH_2)_3$	CH_3	OH	$2HCl \cdot 0.5H_2O$	75	238–239	54.09	7.38	11.83	19.96	53.96	7.63	11.73	20.13		
S-13	$NH(CH_2)_3$	C_2H_4Cl	Cl	$2HCl \cdot 0.5H_2O$	59	126–127	48.40	6.29	9.96	33.60	48.94	6.44	10.16	33.65	3–10	2.7
S-14	$NH(CH_2)_3$	C_2H_4OH	OH	2HCl	76	229–230.5	54.26	7.23	11.17	18.84	54.25	7.37	10.42	19.14		
S-15	$NHCH_2CH_2$	C_2H_4Cl	Cl	2HCl	93	212–213	48.14	5.81	10.53	35.53	48.07	5.88	10.47	35.31	8–36	2.6
S-16	$NHCH_2CH_2$	C_2H_4OH	OH	$2HCl \cdot 0.5H_2O$	87	183.5–184	51.69	7.06	11.32	19.10	51.93	7.12	11.20	19.05		
S-17	...	CH_3	Cl	HCl	67	150 dec.	57.60	5.95	10.32	26.12	57.40	6.14	10.67	26.69	10–60	1.0
S-18	...	CH_3	OH	...	73	58–59	72.20	7.46	12.95		72.14	7.61	12.99			
S-19	...	C_2H_4Cl	Cl	HCl	13	127–128.5	52.65	5.37	8.77	33.32	53.02	5.51	8.92	32.82	20–120	1.0
S-20	...	C_2H_4OH	OH	...	57	83–86	68.28	7.37	11.37		68.22	7.44	11.77			
T-1	$NHCH_2CH_2$	H	Cl	2HCl	85	235–236	48.39	5.62	13.02	32.97	48.43	5.65	13.34	32.85	100–300	1.0
T-2	$NHCH_2CH_2$	H	OH	2HCl	60	213–215	51.32	6.29	13.82	23.31	51.00	6.29	13.46	23.55		
U-1				$2HCl \cdot 1.75H_2O$	40	222–224	49.82	5.97	8.30	28.01	50.68	6.18	8.08	28.06	1–30	1.1
U-2	$NHCH_2CH_2NHCOCHCl_2$			...	49	187–191	52.43	3.91	10.18	25.76	52.87	4.14	9.91	25.53	8–60	1.0
V-1	$NHCH_2CH_2NHCOCHCl_2$			...	12	190–191	47.00	3.64	12.63	32.00	47.33	4.01	12.52	31.32	10–160	1.0
V-2				HCl	35	117.5–118.5	44.45 (S, 9.12)	4.59	7.98	20.20	44.96 (S, 8.72)	4.76	8.27	20.36	10–60	1.1

TABLE I (*Continued*)

Compd. No.	Side chain	Salt	Yield, %	M.p., °C.	Calcd., % C	H	N	Halogen	Found,[a] % C	H	N	Halogen	Antitumor activity[c] Range, μmoles/kg.	Degree
W-1		2HCl·H_2O	55[d]	198–200	51.32	6.30	13.81	23.31	52.72	6.54	13.84	22.84	15–90	1.0
X-1		HCl	53	155.5–157.5	59.00	6.37	9.81	24.84	57.45	6.77	9.60	25.30	30–200	1.0
X-2		HCl	94	235–236.5	63.04	7.18	10.50	13.29	63.18	7.22	10.32	13.70		
		2HCl	40	122–123	38.18	8.41	11.13	42.27	38.27	8.36	11.11	42.31	30–50	2.2

[a] Values are either single analyses or averages of checks. [b] The side-chain precursor of this compound was supplied by Dr. B. F. Tullar. [c] See text; negative, 1.0–4.4; best response, 2.5 and greater. [d] Prepared by the cyclization of the 2-chloroethyl compound (T-1) by a method analogous to those previously reported.

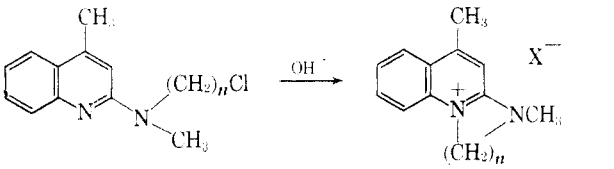
e.g., I, were used in the chlorination procedure, the products were found to contain an additional nuclear



I II

chlorine substituent resulting from the presence of nitrate as oxidizing agent in the chlorinating medium. By degradation to the corresponding 4-hydroxy compound and chlorination to the known 3,4,7-trichloroquinoline,[7] the position was established, as given in II. Further confirmation was furnished by elimination of the other isomeric 4,7,*x*-trichloroquinolines, all four of which are known, and by the analogous 3-halo substitution reaction of Surrey and Cutler.[7]

A different side reaction encountered in the 2-lepidyl series led to cyclization of the ω-chloro compound, which precluded its condensation with diethanolamine and necessitated presynthesis of the entire side-chain skeleton before condensation with the heterocyclic nucleus, as described in the Experimental part. Cycli-



IIIa, $n = 2$ IVa, $n = 2$; $X^- = I^-$
IIIb, $n = 3$ IVb, $n = 3$; $X^- = I^-$

zation of IIIa occurred spontaneously upon neutralizing its hydrochloride in aqueous solution; IIIb cyclized only on heating with excess amine. Analogs of 2,3-dihydro-3,5-dimethyl-1-H-imidazo[1,2-*a*]-quinolin-10-ium iodide (IVa) have been reported by Osbond[8]; although compounds having the skeleton of 1,2,3,4-tetrahydro-4,6-dimethylpyrimido-[1,2-*a*]quinolin-10-ium iodide (IVb) have been recorded, no compounds with analogous bond structure have been reported. By an adaptation of this reaction an analog of IVa bearing a mustard side chain was prepared for testing.

A number of compounds necessary to the synthesis of the listings in Table I, other than their immediate precursors, are given in Table II; representative procedures are described in the Experimental part.

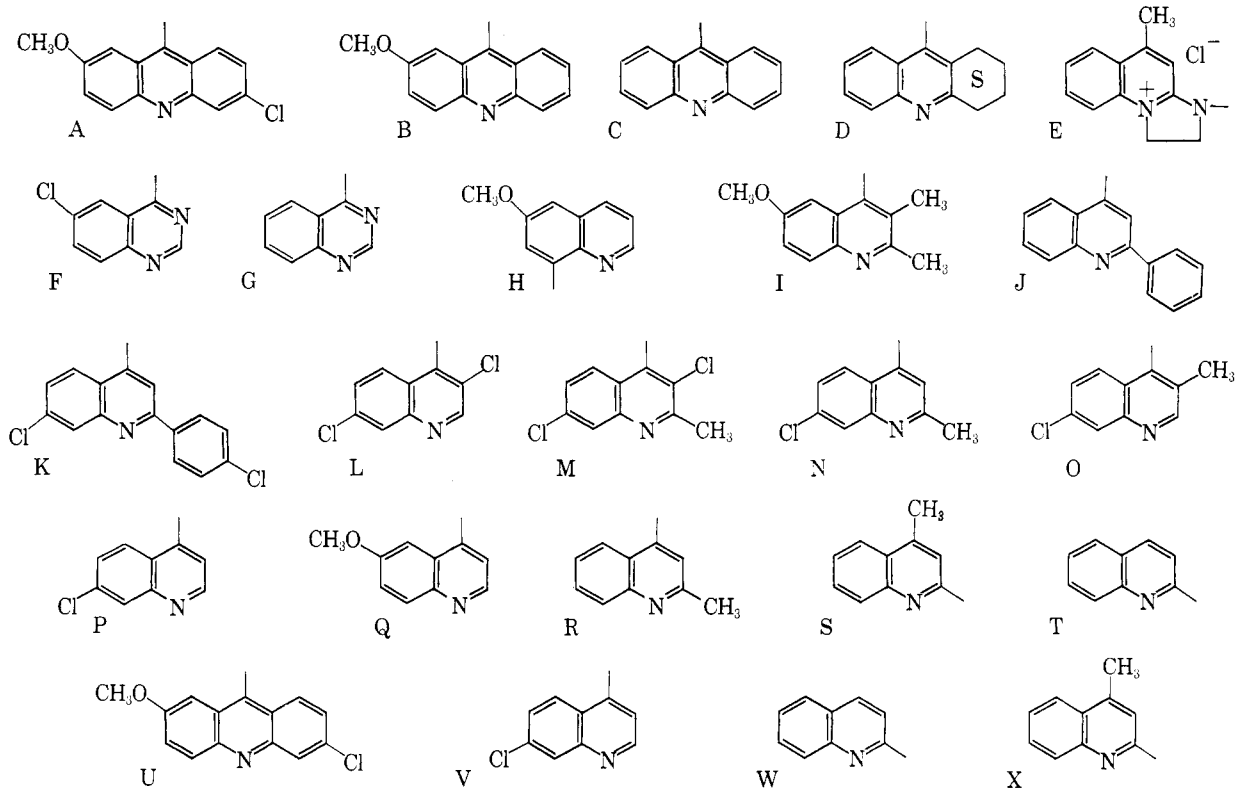
Experimental

Melting points were taken in open capillary tubes in a Hershberg apparatus using total immersion thermometers and are reported as uncorrected values.

All the 2-chloroethyl compounds in this paper were prepared by the action of excess thionyl chloride on their hydroxy precursors.[3] The precursors listed in Table I were, for the most part, prepared by interaction of the corresponding side chain and chloroheterocycle by known methods[3] and from known reactants except those few whose preparations are given below. Other precursors in Table I include those whose side chains were built up in two steps from compounds listed in Table II. A procedure

(7) A. R. Surrey and R. A. Cutler, *J. Am. Chem. Soc.*, **68**, 2570 (1946); A. R. Surrey and H. F. Hammer, *ibid.*, **68**, 1244 (1946); R. E. Lutz, G. Ashburn, J. A. Freek, R. H. Jordan, N. H. Leake, T. A. Martin, R. J. Rowlett, and J. W. Wilson, *ibid.*, **68**, 1285 (1946).

(8) J. M. Osbond, *J. Chem. Soc.*, 1853 (1950).



of general application to this sequence is included in the synthesis of compounds **4** and **5** in Table II and **M-2** in Table I. The hydrochlorides listed in the tables were recrystallized from ethanol or aqueous ethanol, with the addition of acetone and/or ether where necessary with the more soluble compounds. The preparation of two derivatives of 8-amino-6-methoxyquinoline is given below.

N-Methyl-N-2-hydroxyethylethylenediamine.[9]—The preparation of this compound from methylethanolamine and 2-bromoethylamine hydrobromide was carried out by the procedure used for the corresponding N-ethyl compound.[4] A second fractionation gave a 32% yield of product, b.p. 103–104° (8 mm.).

Anal. Calcd. for $C_5H_{14}N_2O$: C, 50.76; H, 11.95; N, 23.71. Found: C, 51.18; H, 12.12; N, 24.69.

N,N′-Dimethyl-N′-2-hydroxyethylethylenediamine and 2-[2-(2-Hydroxyethylmethylaminoethyl)methylamino]lepidine Dihydrochloride.—The first compound was prepared by an identical procedure from methylethanolamine and 2-chloroethylmethylamine hydrochloride. The redistilled fraction, b.p. 114–117° (9 mm.), was obtained in 44% yield. A mixture of 14 g. each of this product and of 2-chlorolepidine was stirred and heated at an internal temperature of 130–135° (exothermic), taken up in dilute acetic acid, and filtered from a small amount of unreacted 2-chlorolepidine. The filtrate was made alkaline, extracted with ether, and concentrated. A slight excess of concentrated hydrochloric acid was added to the residue, water was removed *in vacuo*, and acetone was added to precipitate crystalline **S-2**.

N-Methyl-N′,N′-bis(2-hydroxyethyl)ethylenediamine and 7-Chloro-4-[2-bis(2-hydroxyethyl)aminoethylmethylamino]-quinoline Dihydrochioride.—Substitution of diethanolamine in the above procedure gave the first compound. The redistilled fraction, b.p. 90–100° (10 μ), obtained in 25% yield, was condensed with 4,7-dichloroquinoline at 120° for 2 hr. to give **P-8** (Table I). The same compound was obtained by the reaction of 7-chloro-4-(2-chloroethyl)methylaminoquinoline hydrochloride (**P-11**, Table I) and diethanolamine in slightly higher yield.

8-[2-(2-Hydroxyethylethylamino)ethylamino]-6-methoxyquinoline Dihydrochloride.—This compound was prepared by the methanolic hydroxyethylation of 30 g. of 8-(2-ethylaminoethylamino)-6-methoxyquinoline[10] by methods previously employed.[4] The product was distilled twice *in vacuo*, and a 21-g. fraction boiling at 140–160° (50 μ) was accepted as product. A sample was converted to the hydrochloride (**H-2**, Table I).

2-[2-(6-Methoxy-8-quinolylamino)ethylimino]diethanol Dihydrochloride.—A crude mixture containing the necessary chloro side chain was prepared by dropwise addition of a chloroform solution of 0.5 mole of thionyl chloride into a stirred chloroform solution of 0.5 mole of triethanolamine, refluxing, decanting, slurrying the residue with ethanol, removing the crystalline triethanolamine hydrochloride by filtration, and precipitating the crude, oily, chlorinated product by dilution with ether. It weighed 14 g. and was condensed with 30 g. of 8-amino-6-methoxyquinoline by method I of Drake, *et al.*[11] The product was taken up in ethyl acetate (after 20 g. of excess 8-amino-6-methoxyquinoline was recovered), concentrated, and molecularly distilled twice at 160° (0.2 μ). It weighed 9.6 g. and formed a dihydrochloride (**H-4**, Table I).

7-Chloro-4-(2-hydroxyethyl)methylamino-2-methylquinoline.—A mixture of 21 g. (0.10 mole) of 4,7-dichloro-2-methylquinoline and 37 g. (0.5 mole) of methylethanolamine was stirred and heated for 3 hr. at 110–115° (internal), and taken up in dilute acetic acid. A solution of 60 ml. of saturated sodium nitrate precipitated 34 g. of crude product. This was dissolved in water and made alkaline to give 17.9 g. (71.5%) of the free base, m.p. 103–105°. An analytical sample melted at 102.5–104°.

Anal. Calcd. for $C_{13}H_{15}ClN_2O$: C, 62.26; H, 6.03; N, 11.17. Found: C, 62.00, 62.21; H, 6.05, 6.16; N, 11.19. The **nitrate** (**5**, Table II) was precipitated from a solution in dilute acetic acid with sodium nitrate and recrystallized from water.

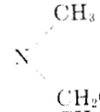
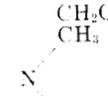
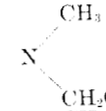
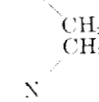
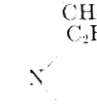
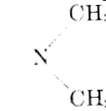
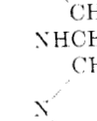
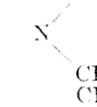
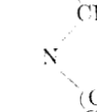
4-(2-Chloroethyl)methylamino-3,7-dichloro-2-methylquinoline Hydrochloride (4, Table II).—To 30 ml. of stirred thionyl chloride was added 5.0 g. of the nitrate salt of 4-(2-hydroxyethyl)-methylamino-7-chloro-2-methylquinoline, with cooling. The solution was kept for 40 hr. at room temperature, excess thionyl chloride was removed *in vacuo*, and the residue decomposed with a small amount of ethanol. Solvent was again removed *in vacuo* and the residue was slurried with 1:1 ethanol–acetone and filtered. The yield was 4.75 g., m.p. 196–200°; it was recrystallized in 80% recovery to give an analytical sample.

(9) In an equivocal reference, O. Eisleb and G. Ehrhart, German Patent 550,762 (Aug. 22, 1930), lists this compound with a melting point (no details).

(10) R. C. Elderfield, W. J. Gensler, J. D. Head, H. A. Hageman, C. H. Kremer, J. B. Wright, A. D. Holley, B. Williamson, J. Galbreath, L. Wiederhold, R. Froghardt, S. M. Kupchan, T. A. Williamson, and O. Birstein, *J. Am. Chem. Soc.*, **68**, 1524 (1946).

(11) N. L. Drake, R. A. Hayes, J. A. Garman, R. B. Johnson, G. W. Kelley, S. Melamed, and R. M. Peck, *ibid.*, **71**, 455 (1949).

TABLE II
ANALYTICAL INFORMATION ON INTERMEDIATES

Compd. no.	Side chain	Salt	Yield, %	M.p., °C.	Calcd., % C	Calcd., % H	Calcd., % N	Calcd., % Cl	Found,[a] % C	Found,[a] % H	Found,[a] % N	Found,[a] % Cl
D-1		HCl	70	191–195	61.76	6.47	9.00	22.79	61.70	6.62	8.99	22.52
D-2		...	22	155–157.5	75.00	7.86	10.92		75.20	8.16	11.39	
E-3		$Cl^- \cdot H_2O$	50	145–148	57.62	5.87	9.58	24.25	57.79	6.49	9.21	24.17
M-4		HCl	70	205–207	45.93	4.16	8.23	41.68	46.12	4.26	8.11	41.36
N-5		HNO_3	71	163.5–165	49.80	5.15	13.39		50.19	5.32	13.52	
L-6		HCl	73	151–154	45.85	4.16	8.24	41.65	46.13	4.32	8.36	41.70
L-7		...	40	68–70	49.75	3.83	9.68	36.74	49.81	3.86	9.51	37.22
L-8		...	98	85–87	47.93	3.29	10.15	38.62	48.31	3.40	10.24	38.03
O-9		HCl	68	198–201	51.15	4.95	9.17	34.80	51.37	5.10	8.46	34.84
P-10		HCl	95	198–201	51.15	4.95	9.17	34.80	51.34	5.15	9.00	34.41
P-11		HCl	50	191–193	54.39	5.62	9.75	24.67	54.63	5.72	9.37	24.90
P-12		HCl	99	185–187	51.14	4.95	9.17	34.80	51.48	5.38	9.15	34.33

[a] Values are either single analyses or averages of checks.

3,7-Dichloro-4-[2-bis(2-hydroxyethyl)aminoethylmethylamino]-2-methylquinoline Dihydrochloride (M-2, Table I).—A mixture of 2.1 g. of **4** (Table II) and 3 g. of diethanolamine was stirred in a heating bath (120–130°) for 3 hr., cooled, and partitioned between water and chloroform-ether. The organic layer was washed with water and then extracted with 10 ml. of 1 *N* hydrochloric acid. This extract plus a small water washing was concentrated *in vacuo*, and the residue was taken up in a small amount of ethanol plus 3 drops of concentrated hydrochloric acid. Addition of acetone precipitated the product which crystallized to give 2.2 g. (80%) of yellow crystals, m.p. 174.5–177°. Recrystallization gave the analytical sample reported in Table I.

Proof of Structure of Compound 6 (Table II).—All the 3-chloroquinolines in this paper were made by the oxidative chlorination reaction described above. A 3.0-g. sample of **6** was refluxed for 4.5 hr. in 15 ml. of 6 *N* hydrochloric acid and cooled overnight. The crude 3,7-dichloro-4-quinolinol was filtered, dissolved in 1 *N* sodium hydroxide, and reprecipitated with acetic acid, yielding 0.90 g. This was refluxed for 5 min. in 4 ml. of phosphorus oxychloride, cooled, decomposed with ice and water, and filtered. The trichloroquinoline obtained in almost quantitative yield was recrystallized from petroleum ether to yield pure 3,4,7-trichloroquinoline, m.p. 115.5–116°.[7]

2,3-Dihydro-3,5-dimethyl-1H-imidazo[1,2-*a*]quinolin-10-ium Iodide.—A solution of 2.1 g. of 2-(2-chloroethylmethylamino)-lepidine hydrochloride (**S-17**, Table I) in 10 ml. of water was neutralized slowly with an exact equivalent of 1 *N* sodium hydroxide. The base precipitated, redissolved as cyclization occurred, then precipitated again as the quaternary salt. After cooling and filtration, it was redissolved in warm water, and an excess of saturated potassium iodide was added. The sparingly soluble iodide precipitated, was filtered, and recrystallized from water. The yield was 0.50 g. (20%), m.p. 241–244°.

Anal. Calcd. for $C_{13}H_{15}IN_2$: C, 47.87; H, 4.64; N, 8.59; I, 38.91. Found: C, 47.41, 47.20; H, 4.83, 5.03; N, 8.04, 8.24; I, 38.69, 39.01.

1,2,3,4-Tetrahydro-4,6-dimethylpyrimido[1,2-*a*]quinolin-10-ium Iodide.—Compound **X-1** (Table II), 2-(3-chloropropylmethylamino)lepidine hydrochloride, did not cyclize on neutralization as did its analog. In an attempt, therefore, to alkylate ethylaminoethanol, 10.5 g. each of **X-1** and the amine were heated for 1 hr. on a steam cone. Under these conditions the self-alkylation reaction occurred and the reaction mixture was completely water soluble. Addition of excess, saturated potassium iodide precipitated 9.9 g. of product, m.p. 211–215°. Recrystallization from water gave 8.5 g. (68%), m.p. 215–217°.

Anal. Calcd. for $C_{14}H_{17}IN_2$: C, 49.44; H, 5.04; N, 8.23; I, 37.29. Found: C, 49.74, 49.54; H, 5.14, 5.20; N, 8.03, 8.18; I, 36.86, 37.00.

6-Chloro-2-methoxy-9-[2-(dichloroacetylamino)ethylamino]-acridine.—A mixture of 2.0 g. each of 6,9-dichloro-2-methoxy-

acridine and ethyl dichloracetate and 3 ml. of ethanol was warmed on a steam cone for 3 hr., cooled, and filtered. The product weighed 2.0 g.; recrystallization from ethanol gave 0.9 g., m.p. 187–191°. Analytical results are given in Table I, **U-1**.

N-(7-Chloro-4-quinolyl)-N-methylethanolamine Methanesulfonate.—To 25 ml. of methanesulfonyl chloride was added 2.0 g. of N-(7-chloro-4-quinolyl)-N-methylaminoethanol. The compound dissolved in about 1 hr. The solution was allowed to stand an additional 3 hr., concentrated *in vacuo*, taken up in ethanol and water, cooled, made alkaline, and extracted with ether to remove unchanged starting material. The supernatant liquid was decanted from the heavy oil which was then washed by decantation. Excess hydrochloric acid was added (cold), water was removed *in vacuo*, and the residue taken up in ethanol. Addition of ether precipitated the product, which was recrystallized from ethanol to give 1.0 g. (34%), m.p. 117.5–118.5°. Analytical results are reported in Table I, **V-2**.

Biological Results.—The results of our studies of the antitumor activity of the compounds are presented in the same summarized form used previously.[2] In the present report, however, only the observations with a hypotetraploid clone of Ehrlich ascites tumor (EF) in albino mice (ICR Swiss) are given. Mice, weighing 24–27 g., were inoculated intraperitoneally with 7 million cells of the EF ascites tumor; on the following day and for the next 2 days, the test compound, dissolved in physiological saline, was injected intraperitoneally into the mice.

The control series of mice for each experiment was injected with saline on days 1, 2, and 3 after tumor inoculation; the mean survival time of the control mice was 16 ± 1 days over the 2-year period. Survival data for each group of mice were recorded daily and the experiments were terminated between days 45 and 51, namely, at the end of the period that was 3 times the mean survival time of the controls for that particular series of tests. Approximately 300 mice were used in each weekly test; the results of the antitumor tests on each compound were based on 100–200 mice.

Dosages are expressed as the number of μmoles of compound (injected on each of the 3 days) per kg. of body weight of mouse. The activity range of a compound covers the lowest to the highest dosages that produced at least an 80% increase in survival time over that of its control group of mice. Dosages that were about 20% greater than the high level in the range usually killed 25–40% of the mice within 3 days of the last injection of compound.

The degree of activity of a compound was calculated statistically from the survival graphs. A value of 3.0 would indicate that all the mice in the experimental group had lived until the time of sacrifice at 45–51 days; with potent compounds, this level of activity was often noted at one or more intermediate levels within the dosage range. An average value of 3.0, however, cannot be attained by any compound because of our definition of the activity range, which utilizes a degree of 1.8 at the low and high ends of the dosage range. The average degree of activity is determined, of course, by the sharpness of the rise to, and fall from, the maximum effect and the existence of any plateau at the 5–8 intermediate testing levels between the lowest and highest effective dosages. A value between 1.0 and 1.4 means that the compound did not cause a significant increase in the survival time of the mice over that of the controls under our conditions of test; in these instances, the dosage range listed is simply that employed up to the toxic level for the compound. The ratio of the high to the low dosages in the activation range is an expression of possible therapeutic usefulness, compounds with a relatively wide range obviously having definite advantages.

Considerable variations were noted in the activities of the compounds, both on a molar basis and on an antitumor basis, dependent on the type of heterocyclic nucleus and its substituents, on the length of the alkyl side chain between the nitrogens, and whether the nitrogen mustard portion of the molecule was mono- or bifunctional.

For ready comparison of the current observations with earlier results,[2] it may be mentioned that the potent monofunctional nitrogen mustard, 6-chloro-2-methoxy-9-[3-(ethyl-2-chloroethyl)aminopropylamino]-acridine dihydrochloride, showed an activity range of 1.5–4 μmoles/kg. and an activity degree of 2.5 against the EF tumor. The analog in which ethylene replaced propylene between the nitrogens of the side chain had a range of 4–16 μmoles/kg. and a degree of 2.4. The corresponding bis nitrogen mustards showed values of 0.5–1.5 μmoles/kg. and a degree of 2.3 for the propyl derivative, and 4–24 μmoles/kg. and a degree of 2.4 for the ethyl derivative. Nitrogen mustard, itself, had values of 1.5–8 μmoles/kg. and a degree of 2.4, and the simple aminoalkyl half-mustard N-(2-chloroethyl)-N-ethyl-1,3-propylenediamine dihydrochloride had an activity degree of 2.3 at 25–60 μmoles/kg. The importance of the alkyl group on the nitrogen containing the 2-chloroethyl group was indicated by the fact that 6-chloro-2-methoxy-9-[3-(2-chloroethyl)aminopropylamino]acridine displayed only slight activity at high dosage levels (45–75 μmoles/kg.) and the ethylamino homolog was inactive. Although the bis nitrogen mustards of 7-chloro- and of 6-methoxyquinoline were highly active, their corresponding ethyl-2-chloroethyl forms were devoid of activity against ascites tumors.

From Table I (**A-1**) it is evident that activity in the 6-chloro-2-methoxyacridines was retained when a methyl group replaced the ethyl group on the nitrogen containing the 2-chloroethyl group, although the dosage requirement was about threefold greater than that of the N-ethyl analog. The homolog with a methylbutylamino side chain and an N-ethyl group (**A-3**) was highly active at a range which approximated that of the reference monofunctional mustard containing the propylamino side chain.

It was of interest to determine the importance of the 2-methoxy and the 6-chloro groups on the acridine nucleus. From a comparison of the mustards **B-1** and **B-3** with their reference compounds, it is clear that the nuclear chloro substituent depressed the molar activity. The 2-methoxyacridine derivatives were found to be the most potent antitumor compounds so far studied in our tests with ascites tumors. The ethylamino analog was also highly active at a reasonably low molar dosage (**B-5**). Deletion of the methoxy group to give **C-1** and **C-3** showed that the unsubstituted acridine nucleus was just as potent an activator of the nitrogen mustard grouping as 2-methoxy-6-chloroacridine. Hydrogenation of an end ring of acridine resulted in a considerably increased dosage require-

ment for a display of activity in the bis mustard (**D-5**). Further alteration in which a tertiary nitrogen was attached to the nucleus caused profound changes in that the monofunctional nitrogen mustard was inactive (**D-1**) and the bis form was active only at high dosage levels (**D-3**).

Although the monofunctional mustards of 6-methoxy- and 7-chloroquinoline obtained previously[2,4] had displayed no activity, we decided to explore a variety of nuclear substituted and unsubstituted two-ring structures to determine whether the acridine ring system was specifically required for an increase in the molar and biological activities of the nitrogen mustard moiety. Although not strictly comparable in structure, **E-1** was inactive. The quinazoline monofunctional nitrogen mustard (**F-1**) and the bis mustards (**F-3** and **G-1**) were active against the ascites tumor but only at high molar dosages. In the 6-methoxy-8-aminoquinoline series, moderate activity was shown by the monofunctional mustard (**H-1**) and high activity by the bis form (**H-3**). Replacement of the terminal benzene ring in 2-methoxyacridine by two methyl groups to give a 2,3-dimethyl-6-methoxyquinoline (**I-1**) resulted in loss of activity in the monofunctional mustard. On the other hand, the presence of a phenyl group at the 2-position of quinoline led to moderate antitumor activity in the N-ethyl monofunctional mustard (**J-3**) and the N-methyl analog (**J-1**). A similar degree of effectiveness against the Ehrlich tumor was noted with the *p*-chlorophenyl analogs (**K-1** and **K-3**) at higher dosage ranges.

The bis mustards (**L-1**, **L-3**, **L-5**, **M-1**, and **M-5**) of a series of 3,7-dichloroquinolines displayed exceptionally broad, effective ranges but the monofunctional mustard (**M-3**) had only slight activity. The effects of structural variations in the side chain of 7-chloroquinoline were also studied and it was found that the bis forms (**P-7** and **P-9**) were active but the monofunctional mustards (**P-1**, **P-3**, and **P-5**) were ineffective. A similar situation prevailed in the 6-methoxyquinoline series (**Q-1** and **Q-3**) with toxicity, however, being evident at low dosage levels.

In contrast, a methyl group in the 7-chloroquinoline nucleus occasionally conferred moderate activity on the monofunctional mustards (**N-3**, and **S-5** to **S-11**). In the case of the last four compounds, it is interesting that the greatest molar activity was imparted by N-isopropyl, followed in turn by N-propyl, N-ethyl, and N-methyl. The aromatic-type bis nitrogen mustard (**S-19**) and the monofunctional mustard with the secondary amine structure (**T-1**) were found to be inactive, in keeping with our earlier observations[2] on these types of structures.

The miscellaneous series of compounds (**U–X**), the components of some of which are effective against certain types of tumors, were found to be inactive in our tests with ascites tumors. The presence of quinacrine in the compounds **U-1** and **U-2** did not confer activity on the two chloro side chains; the methanesulfonate and ethyleneimino derivative (**V-2** and **W-1**) also displayed no activity. The simple alkylamino nitrogen "half-mustard" **Y**, which was prepared to complete the series described previously,[1,2] was moderately effective against the ascites tumor at a relatively high molar dosage similar to that of its N-ethyl reference compound.

Thus, although a moderate degree of antitumor activity at high molar dosage is retained in certain monofunctional nitrogen mustard derivatives of quinazoline and methylquinolines, the only powerful activator at the moment seems to be the intact acridine nucleus. This suggests the possibility that acridines may be unique in their ability to impart bifunctional character to nitrogen "half-mustard." The observations of Lerman[12] indicate that the spatial configuration of this heterocyclic nucleus plays an important role in the special reactivity of various acridines with deoxyribonucleic acid.

(12) L. S. Lerman, *J. Mol. Biol.*, **3**, 18 (1961).

Acridine and Quinoline Analogs of Nitrogen Mustard with Amide Side Chains[1]

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Several mono- and bifunctional nitrogen mustards attached to aminoalkyl derivatives of some acridines and quinolines through an amide linkage were synthesized and studied with the use of ascites tumors. Since the acridine nucleus was again found to exert a powerful activating influence on both the bis and mono nitrogen mustard moieties, the amide linkage was apparently not hydrolyzed to yield glycine mustard during these *in vivo* tests. The presence of a hydrazine linkage in the side chain led to considerably decreased antitumor effectiveness.

One of the initial reasons for our study of quinoline and acridine nitrogen mustards was based on the observation[2] that related carrier molecules (antimalarial drugs) exhibited preferential localization in different tissues dependent on the chemical structure of the heterocyclic base. Thus, the use of a variety of substituted quinoline and acridine carriers might permit the accumulation of the mustard moiety in specific tissues and presumably also in tumors of these tissues. The exceptionally great chemical and biological activities shown by the acridine mono- and bifunctional

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(2) L. H. Schmidt, "A Survey of Antimalarial Drugs," F. Y. Wiselogle, Ed., Edwards Bros., Ann Arbor, Mich., 1946.