residue, in H_2O, was made basic with 12 M NH_4OH and extracted with Et_2O. Drying and evaporation of the extracts gave 17 mg of oil which was converted to 11 mg (58%) of 13-oxalate, mp 196–197°, identical with authentic material.[2]

Interconversion of 9 and 10. Compound **10** (10 mg, 0.034 mmol) and the MeOH–NaOMe prepared from 2 mg (0.087 mmol) of Na and 0.2 ml of MeOH were stirred overnight at room temperature. Removal of MeOH in vacuo, solution of the residue in $CHCl_3$, and evaporation of the H_2O-washed, dried $CHCl_3$ solution gave 8 mg of **9**, identical (TLC, ir) with that described above.

Ac_2O (10 drops), 8 mg (0.032 mmol) of **9**, and 1 drop of C_5H_5N were stirred at room temperature for 1 day, poured into ice–H_2O, basified with K_2CO_3, and extracted with $CHCl_3$. The dried, evaporated (in vacuo below 20°) extract gave 10 mg of solid which was recrystallized from ligroine (bp 30–60°) yielding 6 mg of **9**, mp 94–96°, identical with that described above.

Acknowledgment. We are indebted to Drs. Arthur Jacobson and Kenner Rice for valuable suggestions and to the Section on Instrumentation and Analytical Services for analytical and spectral data.

References and Notes

(1) Visiting Associate from Tanabe Laboratories, Tokyo.
(2) H. Inoue, T. Oh-ishi, and E. L. May, *J. Med. Chem.*, 18, 787 (1975).
(3) The α and β designations used in this paper are with respect to the hydroaromatic ring; see ref 9 for preferred Chemical Abstracts nomenclature.
(4) H. Hodjat, A. Lattes, J. P. Laval, J. Moulines, and J. J. Perie, *J. Heterocycl. Chem.*, **9**, 1081 (1972), and pertinent references cited therein.
(5) Fieser and Fieser, "Reagents for Organic Synthesis", Vol. I, Wiley, New York, N.Y., 1967, p 645; K. B. Wiberg and S. D. Nielsen, *J. Org. Chem.*, **29**, 3353 (1964).
(6) D. P. Vaughan and A. H. Beckett, *J. Pharm. Pharmacol.*, **25**, 895 (1973).
(7) The dihedral angle between C-1 H and C-8 H is about 90° in **9**, **10**, and **11**.
(8) S. E. Fullerton, E. L. May, and E. D. Becker, *J. Org. Chem.*, **27**, 2144 (1962).
(9) T. Oh-ishi, A. E. Jacobson, R. S. Wilson, H. J. C. Yeh, and E. L. May, *J. Org. Chem.*, **39**, 1347 (1974).
(10) T. D. Perrine, L. Atwell, I. B. Tice, A. E. Jacobson, and E. L. May, *J. Pharm. Sci.*, **61**, 86 (1972).
(11) J. H. Ager, S. E. Fullerton, and E. L. May, *J. Med. Chem.*, **6**, 322 (1963).
(12) E. L. May and M. Takeda, *J. Med. Chem.*, **13**, 805 (1970).
(13) N. Yokoyama, F. B. Block, and F. H. Clarke, *J. Med. Chem.*, **13**, 488 (1970).
(14) H. H. Swain and M. H. Seevers, private communication from the Department of Pharmacology, University of Michigan; Committee on Problems of Drug Dependence, National Academy of Sciences, Bulletin on Narcotics, Vol. XXV (no. 2), 1972, p 25.

Interferon Inducing Activities of Derivatives of 1,3-Dimethyl-4-(3-dimethylaminopropylamino)-1*H*-pyrazolo[3,4-*b*]quinoline and Related Compounds

R. R. Crenshaw,* George M. Luke, and Paul Siminoff

Research Division, Bristol Laboratories, Division of Bristol-Myers Company, Syracuse, New York 13201.
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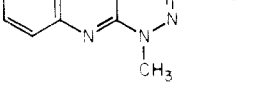
Syntheses and interferon inducing activities are reported for 137 relatives of 1,3-dimethyl-4-(3-dimethylaminopropylamino)-1*H*-pyrazolo[3,4-*b*]quinoline (1). Three different generalized synthetic schemes for the preparation of pyrazolo[3,4-*b*]quinolines are presented and limitations contrasted. Other heterocyclic nuclei containing the 3-dimethylaminopropylamino side chain include pyridine, quinoline, acridine, pyrazolo[3,4-*b*]pyridine, pyrazolo[3,4-*b*][1,8]naphthyridine, pyrazolo[4',3':5,6]pyrido[2,3-*d*]pyrimidine, dipyrazolo[3,4-*b*:4',3'-*e*]pyridine, pyrrolo[2,3-*b*]quinoline, isothiazolo[5,4-*b*]quinoline, and pyrido[2,3-*h*]pyrazolo[3,4-*b*]quinoline. Structural requirements for interferon induction in this series are discussed and two of the more active compounds (**172** and **196**) are compared directly with tilorone.

The capacity of an organism to produce interferon in response to infection by viruses or certain protozoan parasites is thought to be an important nonspecific defense mechanism. The interferon system is the earliest component of the host defense to become operative following virus infection and may also play a role in the later stages of recovery.[1] It is now well established that, once evoked, interferon will inhibit the replication of a wide variety of both RNA- and DNA-containing cytopathic and oncogenic viruses.[2] An agent which stimulates release and/or in vivo synthesis of interferon thus has implications for the development of a clinically useful broad-spectrum antiviral drug. Since the discovery of the interferon system in 1957,[3] a number of polymeric substances have been reported which stimulate interferon production, but all of these have limitations in clinical utility.[4] Impetus for the search for a monomeric low-molecular-weight inducer of interferon was provided by reports that 2,7-bis[2-(diethylamino)-ethoxy]fluoren-9-one (tilorone) and congeners induce interferon activity in mice.[5,6] A preliminary report from our laboratories disclosed the interferon-eliciting activity of an entirely different structural type, viz. the pyrazolo[3,4-*b*]quinoline 1 (BL-20803).[7] We now report results

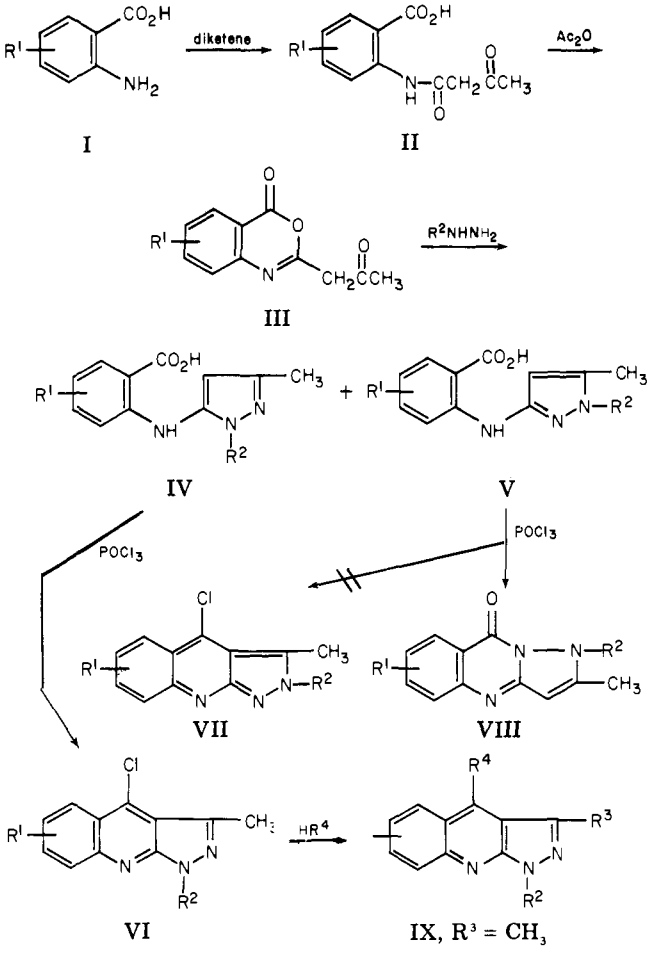


1

of a research program designed to (i) define structural features of 1 responsible for the interferon-eliciting activity and (ii) increase potency relative to 1 with a concomitant improvement in therapeutic ratio.

Chemistry. Relatives of 1 which were prepared for biological evaluation are listed in Tables III and IV. Some of the simpler fragment moieties of 1 and related acridines were prepared by literature procedures as indicated in Table III. The amide **65**, which may be viewed as a hydrolytically cleaved form of 1, was prepared from the corresponding acid chloride and 3-dimethylaminopropylamine. Treatment of **65** with diborane in THF gave the reduced derivative **66**.

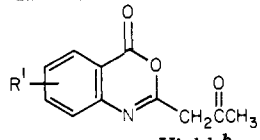
Scheme I



Most of the pyrazolo[3,4-*b*]quinolines were prepared as indicated in Scheme I. This route was first used by Wolfrum et al.[8,9] for preparation of 4-halo-, alkoxy-, or aryloxy-substituted pyrazolo[3,4-*b*]quinolines useful as optical brightening agents. The procedure was subsequently used by Stein and coworkers for the preparation of 1 which was initially prepared as a potential antimalarial agent[10] and later found to possess hypocholesterolemic activity.[11] Although neither group reported encountering the isomeric pyrazolylanthranilic acid V, we usually obtained V as a contaminant of the desired isomer IV in varying amounts depending on the substituents in the benzenoid ring and the reaction conditions used. We found that it generally was not necessary to isolate the intermediates II–VI in pure form prior to conversion to IX (cf. Experimental Section). Intermediate benzoxazinones (III) and acids (IV and V) which were isolated and characterized are listed in Tables I and II, respectively. Three pure isomers V (**52**, **53**, and **54**) were isolated for attempted conversion to a 2-methyl-substituted pyrazolo[3,4-*b*]-quinoline derivative VII. However, treatment with $POCl_3$ produced the corresponding cyclic amide VIII rather than VII. As an incidental point, this experimental result confirms the isomeric assignments of the acids **52**, **53**, and **54** and also supports the assignment of pyrazolo[3,4-*b*]-quinolines made from this route by us and by others[8-11] as the 1-alkyl rather than 2-alkyl isomers.

Scheme I permitted variation of R^1 and R^2 through choice of appropriately substituted anthranilic acids and hydrazines, respectively. This route could not be used for synthesis of 1-unsubstituted pyrazolo[3,4-*b*]quinolines (R^2 = H). When hydrazine was condensed with the benzoxazinone III (R^1 = H), the only product isolated was VIII

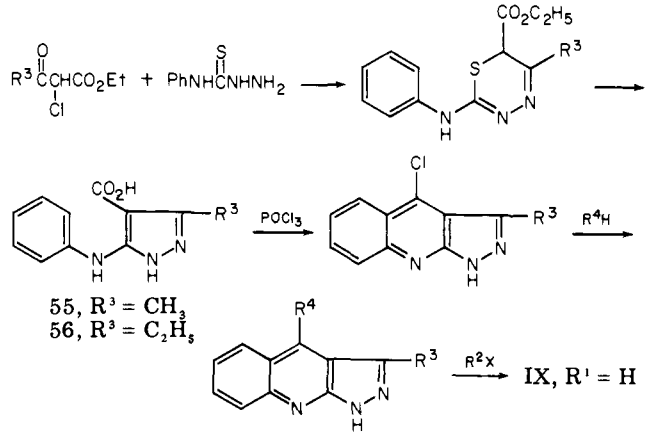
Table I. Substituted 2-Acetonyl-4*H*-3,1-benzoxazin-4-ones



No.	R^1	Mp, °C[a]	Yield,[b] %	Formula[c]
2	6-Cl	158–163	84	$C_{11}H_8ClNO_3$
3	6-I	168–170	36	$C_{11}H_8INO_3$
4	5-CH_3	143–145	60	$C_{12}H_{11}NO_3$
5	6-CH_3	138.5–140	74	$C_{12}H_{11}NO_3$
6	7-CH_3	142–144	48	$C_{12}H_{11}NO_3$
7	8-CH_3	152.5–154.5	5	$C_{12}H_{11}NO_3$
8	6,7-$(CH_3)_2$	126–129	53	$C_{13}H_{13}NO_3$
9	6,8-$(CH_3)_2$	156–159	63	$C_{13}H_{13}NO_3$
10	7,8-$(CH_3)_2$	188.5–192	58	$C_{13}H_{13}NO_3$
11	7-C_2H_5	93–97	54	$C_{13}H_{13}NO_3$[d]
12	7-CF_3	107–109	18	$C_{12}H_8F_3NO_3$[e]
13	5-CH_3O	158.5–161	49	$C_{12}H_{11}NO_4$[f]
14	6-CH_3O	154.5–157	8	$C_{12}H_{11}NO_4$
15	5,7-$(CH_3O)_2$	178–180	85	$C_{13}H_{13}NO_5$
16	5-NO_2	150–153	65	$C_{11}H_8N_2O_5$
17	7-NO_2	172–174	77	$C_{11}H_8N_2O_5$
18	7-CO_2H	>300	79	$C_{12}H_9NO_5$[g]

[a] Broad melting ranges may reflect a mixture of tautomeric forms (cf. Experimental Section). [b] Yields of analytically pure products. All compounds were recrystallized from MeCN except 18 which was purified by trituration under warm AcOH. [c] All new compounds were analyzed for C, H, and N. Analytical results were within ±0.4% of the theoretical values unless otherwise indicated. [d] C: calcd, 67.52; found, 68.12. [e] C: calcd, 53.15; found, 53.58. [f] C: calcd, 61.80; found, 61.30. [g] C: calcd, 58.30; found, 55.98. Mass spectrum (70 eV) *m/e* 247.

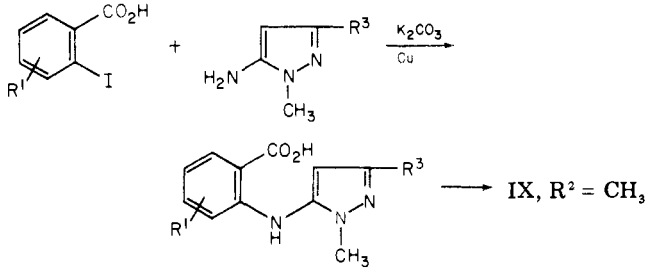
Scheme II



(R[1], R[2] = H).[12] Additionally, the C-3 position could not be varied by Scheme I because the 3-methyl group in IX is derived from diketene.

Both of the aforementioned variations not permitted by Scheme I were possible by the alternative synthesis outlined in Scheme II. Synthesis of the acid **55** as indicated in Scheme II was reported by Bulka et al.[13] We used the same procedure for the preparation of **56** starting with ethyl α-chloropropionylacetate and 4-phenylthiosemicarbazide. Acids **55** and **56** were converted to the

Scheme III



1-unsubstituted pyrazolo[3,4-*b*]quinolines **136** and **152**, respectively. When the sodium salt of **136** in DMF was treated with 1 equiv of CH_3I, alkylation occurred exclusively at the 1 position to give a quantitative yield of **1**. Scheme II thus provided an alternative to Scheme I for variation of R^2, as well as permitting variation of R^3.

Neither Scheme I nor Scheme II was applicable for preparation of a 3-unsubstituted pyrazolo[3,4-*b*]quinoline (IX, R^3 = H). This variation was made possible via Scheme III. In an Ullmann type reaction, 5-amino-1-methylpyrazole[14] was condensed with *o*-iodobenzoic acid to yield the acid **29** which then was converted through the usual sequence to **154**. A similar reaction using 5-amino-1,3-dimethylpyrazole[15] provided an alternative synthesis of the acid **19** and, thus, an alternative to Scheme I for the synthesis of **1**. A variation of Scheme III starting with 5-amino-3-methylisothiazole was used for the preparation of **75**. The general approach of Scheme III was also used for the preparation of **70**, **71**, and **162** (cf. Experimental Section).

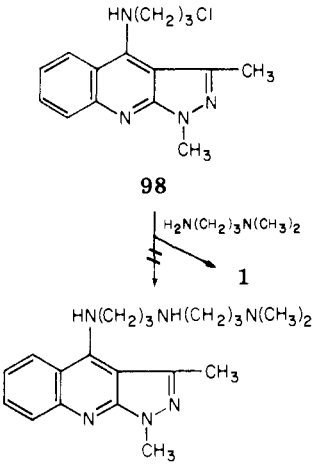
The 6-nitropyrazolo[3,4-*b*]quinoline **162** thus obtained via Scheme III was identical with the product obtained from direct nitration of **1**. By inference, the product obtained from sulfonation of **1** was assigned as the 6-SO_3H derivative **181**. The related sulfonamide derivatives **182** and **183** were prepared from **181**.

Problems were encountered in initial attempts to obtain the 5- and 7-nitro derivatives **161** and **163**. Treatment of the acid **38** with $POCl_3$ under even very mild conditions gave black tars from which none of the intermediate VI (R^1 = 5-NO_2; R^2 = CH_3) could be isolated. Treatment of **38** with H_2SO_4 gave the corresponding 4-hydroxy intermediate which condensed satisfactorily with $POCl_3$ to give VI (R^1 = 5-NO_2; R^2 = CH_3) which subsequently was converted to **161**. Heating VI (R^1 = 7-NO_2; R^2 = CH_3) in excess 3-dimethylaminopropylamine gave none of the expected 7-nitro derivative **163**, but rather a product which we have characterized as the tetracyclic derivative **77**.[16] Reaction of VI (R^1 = 7-NO_2; R^2 = CH_3) with an equimolar quantity of the amine gave **163**.

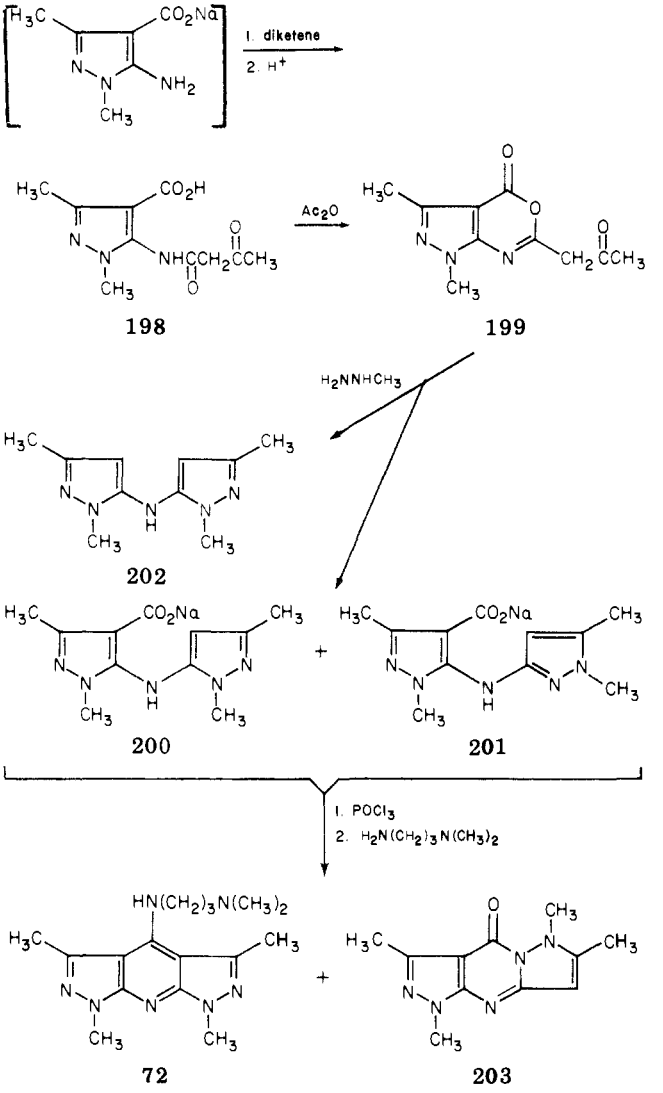
A particularly noteworthy result of our chemical investigation is illustrated in Scheme IV. Heating **98** in excess 3-dimethylaminopropylamine resulted in none of the expected triamine side-chain derivative shown but rather yielded **1** in essentially quantitative yield. Another example in which displacement of the entire 3-chloropropylamine side chain resulted involved reaction of **98** with cyclopropylamine to yield **86**. On the other hand, reaction of **98** with excess pyrrolidine, methylamine, isopropylamine, *tert*-butylamine, *N,N,N'*-trimethyl-1,3-propanediamine, piperidine, and piperazine resulted in displacement of only the chloro group of **98** to yield **117**, **103**, **104**, **105**, **115**, **119**, and **122**, respectively.

Synthesis of the symmetrical dipyrazolo[3,4-*b*:4',3'-*e*]-pyridine derivative **72** (Scheme V) presented special problems. 5-Amino-1,3-dimethylpyrazole-4-carboxylic acid required for preparation of **72** analogously to Scheme I is

Scheme IV

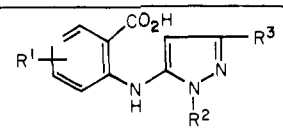


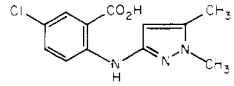
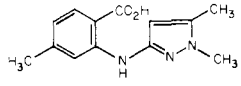
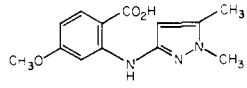
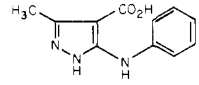
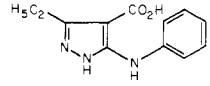
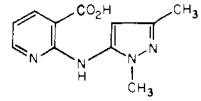
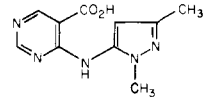
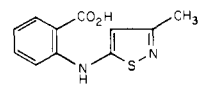
Scheme V



unknown. Reported attempts by Taylor and Hartke to isolate this acid were unsuccessful owing to its instability to decarboxylation.[15] We thus carefully treated without isolation the presumed salt of the acid (obtained from basic hydrolysis of 5-amino-1,3-dimethylpyrazole-4-carbonitrile[15]) with diketene and obtained the *N*-acetoacetyl derivative **198**. This product was converted to the oxazinone **199**. Attempts to obtain the free acids of **200** and **201** by reaction of **199** with methylhydrazine in acetic acid

Table II. Substituted *N*-(Pyrazolyl)anthranilic Acids and Related Heterocyclic Acids



No.	R^1	R^2	R^3	Mp, °C	Recrystn solvent[a]	Yield,[b] %	Formula[c]
19	H	CH_3	CH_3	208-210.5[d]	A	46	$C_{12}H_{13}N_3O_2$
20	H	C_2H_5	CH_3	185-187	A	45	$C_{13}H_{15}N_3O_2$
21	H	CH_2CF_3	CH_3	189-191.5	B	63	$C_{13}H_{12}F_3N_3O_2$
22	H	$CH(CH_3)_2$	CH_3	212-215[e]	A	62	$C_{14}H_{17}N_3O_2$
23	H	CH_2CH_2OH	CH_3	216.5-218[f]	A	71	$C_{13}H_{15}N_3O_3$
24	H	$CH_2C_6H_5$	CH_3	193-194.5[g]	A	69	$C_{18}H_{17}N_3O_2$
25	H	C_6H_5	CH_3	214-216.5	A	85	$C_{17}H_{15}N_3O_2$
26	H	*p*-$C_6H_4OCH_3$	CH_3	204-207	A	66	$C_{18}H_{17}N_3O_3$[h]
27	H	*p*-$C_6H_4CO_2H$	CH_3	>300	C	90	$C_{18}H_{15}N_3O_4$
28	H	$(CH_2)_3N(CH_3)_2$	CH_3	194-196	D	62	$C_{16}H_{22}N_4O_2$
29	H	CH_3	H	174-176.5	A	42	$C_{11}H_{11}N_3O_2$
30	4-F	CH_3	CH_3	250.5-251.5	A	33	$C_{12}H_{12}FN_3O_2$
31	5-F	CH_3	CH_3	228.5-230.5	A	44	$C_{12}H_{12}FN_3O_2$
32	3-Cl	CH_3	CH_3	185-187.5	E	37	$C_{12}H_{12}ClN_3O_2$
33	4-Cl	CH_3	CH_3	237-238[i]	A	92	$C_{12}H_{12}ClN_3O_2$
34	5-Cl	CH_3	CH_3	241-242	A	79	$C_{12}H_{12}ClN_3O_2$
35	5-I	CH_3	CH_3	239-240.5 dec	C	67	$C_{12}H_{12}IN_3O_2$
36	4-NO_2	CH_3	CH_3	255-257	A	67	$C_{12}H_{12}N_4O_4$
37	5-NO_2	CH_3	CH_3	278-280 dec	A	90	$C_{12}H_{12}N_4O_4$
38	6-NO_2	CH_3	CH_3	236-248 dec	A	62	$C_{12}H_{12}N_4O_4$
39	4-CH_3	CH_3	CH_3	188-192	A	83	$C_{13}H_{15}N_3O_2$
40	5-CH_3	CH_3	CH_3	194-196	A	69	$C_{13}H_{15}N_3O_2$
41	6-CH_3	CH_3	CH_3	171.5-173.5	A	47	$C_{13}H_{15}N_3O_2$
42	4-CF_3	CH_3	CH_3	226-229	A	62	$C_{13}H_{12}F_3N_3O_2$
43	4-C_2H_5	CH_3	CH_3	166-168	B	40	$C_{14}H_{17}N_3O_2$
44	5-C_2H_5	CH_3	CH_3	195-197	B	37	$C_{14}H_{17}N_3O_2$
45	3,5-$(CH_3)_2$	CH_3	CH_3	223-226 dec	A	60	$C_{14}H_{17}N_3O_2$[j]
46	4,5-$(CH_3)_2$	CH_3	CH_3	220-221.5	A	32	$C_{14}H_{17}N_3O_2$[k]
47	3-CH_3O	CH_3	CH_3	173-176.5 dec	F	26	$C_{13}H_{15}N_3O_3$
48	4-CH_3O	CH_3	CH_3	225-226.5 dec	A	46	$C_{13}H_{15}N_3O_3$
49	3-CH_3O, 6-Cl	CH_3	CH_3	216-218.5 dec	A	42	$C_{13}H_{14}ClN_3O_3$
50	4,5-$(CH_3O)_2$	CH_3	CH_3	211.5-213 dec	A	93	$C_{14}H_{17}N_3O_4$
51	4,5-benzo	CH_3	CH_3	245-247.5 dec	A	55	$C_{16}H_{15}N_3O_2$
52				195-196 dec	A	14[l]	$C_{12}H_{12}ClN_3O_2$
53				227-229	A	33[l]	$C_{13}H_{15}N_3O_2$
54				208.5-210	A	25[l]	$C_{13}H_{15}N_3O_3$
55				198 dec[m]	A	74	$C_{11}H_{11}N_3O_2$
56				199-200 dec	A	24	$C_{12}H_{13}N_3O_2$
57				263-265 dec	A	64	$C_{11}H_{12}N_4O_2$
58				284-286 dec	G	58	$C_{10}H_{11}N_5O_2$
59				212-213 dec	D	17	$C_{11}H_{10}N_2O_2S$

[a] A, EtOH; B, MeCN; C, AcOH; D, absolute EtOH; E, *i*-PrOH; F, $MeNO_2$; G, DMF. [b] For acids which were prepared via Scheme I, in which isomers were always encountered, yields are for crude products which were used directly in the next step except as noted in the Experimental Section. The melting points given for these acids are for chromatographically pure samples (GLC analysis after treatment with CH_2N_2) obtained by recrystallization of an aliquot of the crude acid from the indicated solvent. Yields for acids made by routes other than Scheme I (cf. Experimental Section) represent pure products. [c] See footnote *c* in Table I. [d] Lit.[11] mp 212-213°. [e] Lit.[11] mp 206-208°. [f] Lit.[11] mp 210-211°. [g] Lit.[11] mp 192-193°. [h] C: calcd, 66.86; found, 66.16. [i] Lit.[10] mp 232-233°. [j] N: calcd, 16.21; found, 15.78. [k] N: calcd, 16.21; found, 15.61. [l] Percent of product relative to the corresponding 1-methyl isomer present in the crude product as determined by GLC assay (cf. Experimental Section), isolated analogous to the procedure described for 53. [m] Lit.[13] mp 195°.

led to the recovery of the symmetrical amine **202** as the only identifiable product. Experimental conditions then were developed which permitted preparation of the salts **200** and **201** and their subsequent elaboration without isolation into the tricyclic derivatives **72** and **203**.

Biology. Details of oral or parenteral dosing of mice and the assay of serum interferon have been published.[7] Briefly, compounds were dissolved or suspended in sterile distilled water and administered to 15-g female CD-1 mice. At least three mice were treated at each dose level of compound. Later (18–20 hr), mice were bled and their sera collected, pooled, and quantitatively assayed for interferon activity by a plaque reduction method employing mouse L cells and challenge by vesicular stomatitis virus. The reliability of the assay was verified by including a reference mouse interferon standard provided by the National Institute of Allergy and Infectious Diseases. In our hands, the standard assayed $10^{4.7}$ units/ml of interferon as compared to the assigned potency of $10^{4.5}$ units/ml.

For simplicity, the interferon data listed in Tables III and IV are expressed as the lowest dose of compound required to elicit a significant level of circulating interferon, i.e., ≥32 units/ml.[7] The highest doses given to the animals were 400 mg/kg po or 300 mg/kg ip. In general, no major differences in activity were obtained between routes of administration with the exception that the parenteral route evinced toxicity more frequently than did the oral route.

For convenience in discussing structure–activity correlations, compounds assayed may be divided into the following groups: (i) fragment moieties of **1** (**60**, **61**, **64a–c**, **65–68**, the acid **19**, 1,3-dimethyl-5-aminopyrazole, and 3-dimethylaminopropylamine); (ii) side-chain variants (**79–135**); (iii) ring A (benzenoid) nuclear modifications (**69–73**); (iv) ring C (pyrazole) modifications (**62**, **63**, **74**, **75**, **136–154**); (v) ring A substituent variants (**76**, **77**, **155–197**); and (vi) bis compounds related to **1** (**78**).

Results obtained indicated that activity in this series is confined within rather narrow limits. Excepting the marginal activity shown by **70**, fragment moieties of group i and ring A nuclear modifications of group iii were devoid of activity. Especially noteworthy is the inactivity seen with the progression **60**, **61**, **64a–c**, and with the tetrahydro derivative **69**. These results suggested that the nucleus requirement for activity is at least three appropriately fused aromatic or heteroaromatic rings. The one bis tricyclic compound prepared (**78**) in group vi was inactive or nearly so.

A number of compounds of group ii were active within certain structural limitations. Results seen among compounds **80–135** indicated that for activity the substitution at the 4 position of the pyrazolo[3,4-*b*]quinoline nucleus has to be NH attached to a side chain of at least three carbons in length terminating in a second amino function. Particularly noteworthy in this group are the inactive compounds **89** and **90** which differ from the parent **1** only by substitution for NH by O and S, respectively. For retention of activity, the side chain may be varied in length (cf. **92–94**) and some variation of the terminal amine function is permissible although none of these changes resulted in an analog significantly more active than **1**. Interestingly, even changing the terminal amine function to diethylamino (**110**) resulted in lower activity relative to **1**. All derivatives of **1** having a tertiary rather than secondary amine function at the 4 position of the tricyclic nucleus were inactive (**131–135**).

Very little change of the pyrazole moiety in **1** was permissible for retention of activity (group iv) and no member of this group proved more active than **1**. Even the minimal change of the 1-normethyl (**136**) or 1-ethyl (**137**) or 3-ethyl (**153**) derivatives of **1** resulted in inactive or nearly inactive compounds. All other changes at the 1 position resulted in compounds inactive at nontoxic doses. Removal of the 3-methyl in **1** (**154**) resulted in a derivative of comparable activity to **1**. Changing the pyrazole for benzenoid (**62**, **63**) or isothiazole (**75**) resulted in inactivity. Interestingly, substitution of pyrazole by pyrrole (**74**) resulted in an active derivative, but one shown by quantitation of units of serum interferon to be less active than the pyrazole relative **154**.

Success in enhancing potency of **1** was achieved with the preparation of certain ring A substituted derivatives of **1** in group v. Again, however, considerable structural specificity was seen within this group as was encountered in the other groups. With the exception of the 7-Cl derivative **158**, substitution of halogen (**155–160**) resulted in considerable loss of activity. Nitro (**161–163**) and amine (**164–169**) derivatives were inactive with the exception of the 6-C_2H_5NH derivative **167** which was about four times as active as **1**. The 6-CH_3 (**171**) and 7-CH_3 (**172**) derivatives proved to be among the most active compounds seen, yet the 5- (**170**) and 8- (**173**) CH_3 derivatives were devoid of activity. While the 8-CH_3 derivative was inactive, the 7,8- (**179**) and 6,8- (**180**) di-CH_3 derivatives were active. Combination of the two active monomethyl derivatives (**171** and **172**) into the 6,7-disubstituted derivative **178** resulted in one of the more active compounds in the series. Additional examples of the structural specificity in this series are seen in the 7-CF_3 (**174**), 7-C_2H_5 (**176**), and 7-CH_2OH (**177**) relatives of **172**, all of which are either inactive or considerably less active than **172**. The only two tetracyclic compounds prepared (**76** and **77**) have been considered within this class of compounds comprising ring A substituted derivatives. Both compounds were active and again reflect, analogously to compounds **178** and **179**, that the 6,7 and 7,8 positions may be disubstituted with retention of activity. Acid derivatives at positions 6 and 7 (**181–187**) were uniformly inactive. In contrast to the four monomethyl derivatives discussed above, each of the monomethoxy derivatives (**188–191**) was active. The 5,7-dimethoxy derivative **196** was among the most active compounds prepared in the series. Surprisingly, the trimethoxy derivative **197** was inactive.

Of the compounds described herein, the most active appear to be **167**, **171**, **172**, **178**, **188**, **190**, **195**, and **196**. Two of the more active compounds, **172** and **196**, were compared with tilorone in five separate dose–response studies (Figure 1). Each compound received at least four separate evaluations at each of the dose levels employed. There was considerable spread in the interferon response at dose levels required to elicit significant interferon activity in the circulation. It should also be noted that at 50 mg/kg, compound **172** failed to stimulate measurable amounts of interferon in three out of five trials. On the other hand, compound **196** and tilorone were active in one instance at 25 mg/kg and in all instances at 50 mg/kg. At the latter dose level, the response to tilorone tended to distribute at the higher end of the spread than did the response to **196**. We conclude that an MED range of 25–50 mg/kg should be assigned to **196** and to tilorone while the MED of **172** is 50–100 mg/kg.

We have previously reported a good correlation between interferon-inducing activity and suppression of vaccinia virus infection by compound **1** in mice.[7] Other compounds in this series have also shown this same close relationship. For example, the interferon-inducing and vaccinia virus-inhibiting MED's for compounds **171** and **172** were

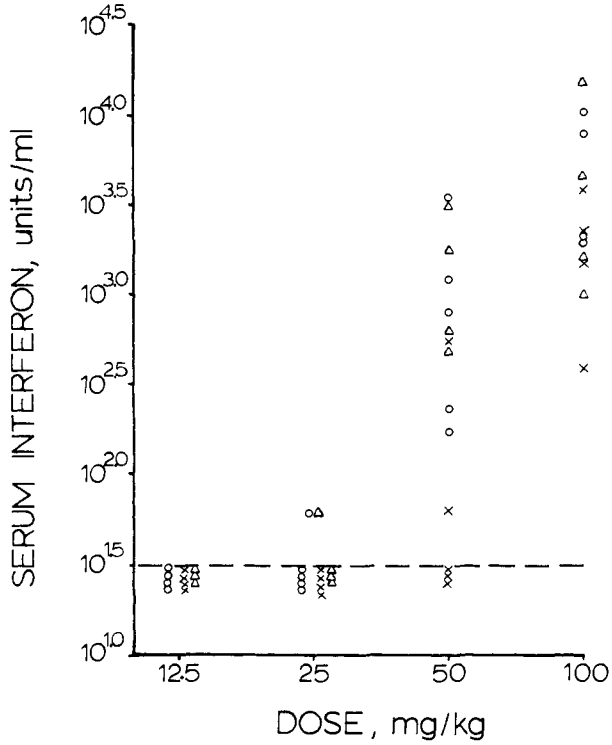


Figure 1. The distribution of mouse serum interferon activities following oral dosing with various concentrations of 172 (×), 196 (○), and tilorone (△) in five separate experiments. The interrupted line represents the lower limit of assay sensitivity.

identical, i.e., 50 mg/kg given orally or intraperitoneally.

Acute toxicity of compound 172 in mice is approximately equal to that reported for compound 1 (250 mg/kg ip and 1600 mg/kg po).[7] Thus, based on MED's of 200 and 50 mg/kg for 1 and 172, respectively, a fourfold improvement in the therapeutic ratio has been achieved.

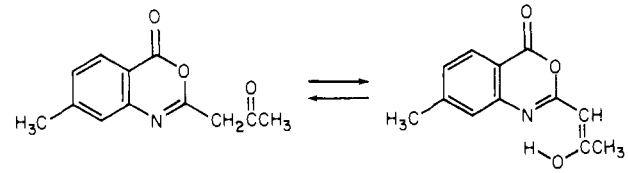
Discussion

At the time we started our research on the "lead" compound 1, tilorone was the only significant low-molecular-weight inducer of interferon. Since the inception of our program, a few low-molecular-weight inducers other than the fluorenone type have been reported. Among these are a series of diamines,[17] e.g., *N*,*N*-bis(2-hydroxyethyl)-*N'*,*N'*-dioctadecyl-1,3-propanediamine,[18-20] certain basic dyes, e.g., toluidine blue,[21] and the antimalarial drugs quinacrine and acridine.[22] Where the activity levels were indicated, the amounts of interferon produced were low relative to levels (Figure 1) produced in this series of compounds and with tilorone. The compounds of the present study thus add a new class of compounds to those known to elicit circulating interferon and some members of the series are among the most potent low-molecular-weight inducers reported to date. Furthermore, compounds of the present series appear to afford an advantage over other reported interferon inducers in exhibiting a delay in onset of a hyporesponsive state after multiple dosing.[7] Work is continuing in our laboratories and elsewhere to investigate the full therapeutic potential of this class of compounds.

Experimental Section

Melting points are capillary and are uncorrected. All compounds had consistent NMR and ir spectra. Where elemental analyses are indicated by symbols of the elements, analytical results were within ±0.4% of the theoretical values. All required anthranilic acids and hydrazines are previously described compounds. For brevity, detailed procedures are given for one representative example of a general type, and additional compounds synthesized from the general procedures are so indicated. Where reaction procedures were not straightforward and/or where compounds do not fit into generalized class reactions, the experimental details are given in full. "Alumina" refers to Merck & Co. reagent aluminum oxide (product no. 71707); "neutral alumina" and "basic alumina" refer to J. T. Baker Co. aluminum oxide, neutral (catalog no. 0540), and aluminum oxide, basic (catalog no. 0539), respectively.

2-Acetonyl-7-methyl-4*H*-3,1-benzoxazin-4-one (6). A mixture of 4-methylanthranilic acid (25.0 g, 0.17 mol) and diketene (13.95 g, 0.17 mol) in CCl_4 (250 ml) was heated at reflux for 2.5 hr. Acetic anhydride (18.3 g, 0.18 mol) was added and the mixture was stirred under reflux for 16 hr. The mixture then was cooled and the product isolated by filtration; recrystallization (MeCN) gave 6 (17.4 g, 48%), mp 142–144°. NMR analysis indicated that in solution ($CDCl_3$) the product is a mixture of the tautomeric forms A and B in a ratio of approximately 1:2, respectively: δ (A) 3.80 (s, CH_2), 2.50 (s, CH_3-aro), 2.32 ppm (s, CH_3CO); δ (B)



A B

5.23 (s, C*H*=COH), 2.45 (s, CH_3-aro), 2.11 ppm (s, C*H*$_3$COH). All 3,1-benzoxazin-4-ones prepared were a mixture in solution of these two forms. Similarly prepared by this procedure starting with the appropriate anthranilic acids were 2–8, 11–14, and 17; for 3 and 13, THF was added to the CCl_4 to increase solubility. The benzoxazinone 15 was prepared by this procedure except the intermediate *N*-acetylacetamide was prepared and isolated (by the procedure which follows for 17) prior to cyclization in CCl_4.

2-Acetonyl-7-nitro-4*H*-3,1-benzoxazin-4-one (17). Diketene (17.30 g, 0.21 mol) was added during 30 min to a solution of 4-nitroanthranilic acid (37.60 g, 0.20 mol) in 1.33 *N* aqueous NaOH (150 ml, 0.20 mol) while maintaining the temperature at 28–30° with external cooling. The solution was stirred at 25° for 90 min and then was cooled and acidified with 1.5 equiv of 6 *N* aqueous HCl to yield *N*-(acetoacetyl)-4-nitroanthranilic acid (47.8 g, 90%), mp 172–175° dec. This product (47.5 g, 0.18 mol) was stirred with Ac_2O (25 ml, 0.27 mol) in AcOH (190 ml) at 80° for 2.5 hr. The solution was cooled and filtered to yield 17 (34.0 g, 77%): mp 172–174° (MeCN); NMR (Me_2SO) indicated an approximately 1:1 mixture of the tautomeric forms.

Similarly prepared by this procedure from the appropriate anthranilic acids were 9, 10, 16, and 18.

***N*-(1,3-Dimethylpyrazol-5-yl)-4-methylanthranilic Acid (39) and *N*-(1,5-Dimethylpyrazol-3-yl)-4-methylanthranilic Acid (53).** The benzoxazinone 6 (15.31 g, 0.071 mol) was added to a solution of methylhydrazine (3.90 g, 0.085 mol) in acetic acid (150 ml) and the resultant solution was stirred at 80° for 2.5 hr. The solvent was removed and the product was triturated under MeCN at 0° to yield 14.50 g (83%) of crude 39 containing about 33% of the isomeric acid 53 (determined by GLC analysis after CH_2N_2 treatment). A 9.50-g aliquot was triturated under 300 ml of hot MeCN and the insoluble solid (2.23 g, about 85% of 53) isolated by filtration; additional recrystallization (EtOH) gave pure 53, mp 227–229°. The product obtained from the 300 ml of MeCN filtrate was recrystallized successively from MeCN and EtOH to yield pure 39, mp 188–192°.

Similarly prepared from the appropriate benzoxazinones and known hydrazines were acids 19–28, 30–36, and 38–54.

4-(3-Dimethylaminopropylamino)-1,3,7-trimethyl-1*H*-pyrazolo[3,4-*b*]quinoline Dihydrochloride (172). A mixture of the above crude acid 39 (5.00 g, 0.02 mol) and $POCl_3$ (50 ml) was stirred at reflux for 3 hr. The cooled mixture was concentrated at 5 mm to a syrup which was poured onto ice. The mixture was basified with 4 *N* NaOH and then was extracted with chloroform. The chloroform extracts were washed, dried, and evaporated to yield crude 4-chloro-1,3,7-trimethyl-1*H*-pyrazolo[3,4-*b*]quinoline. The product was treated with 3-dimethylaminopropylamine (50 ml) and the mixture was stirred at 130° for 12 hr. The mixture

Table III. Heterocycle Derivatives Related to 1

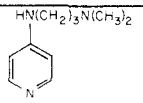
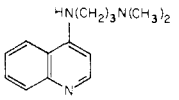
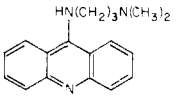
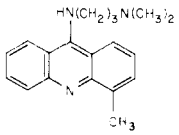
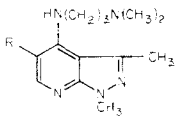
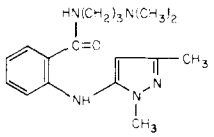
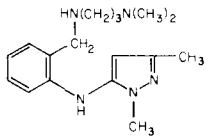
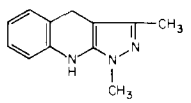
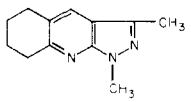
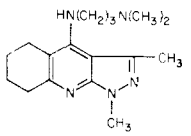
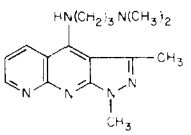
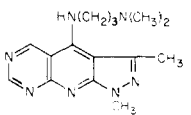
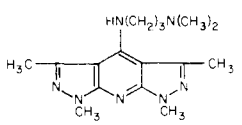
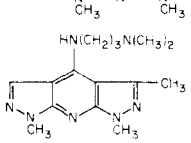
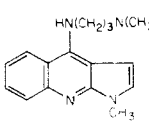
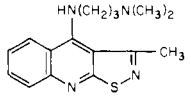
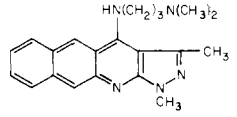
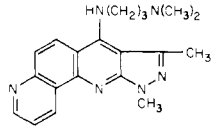
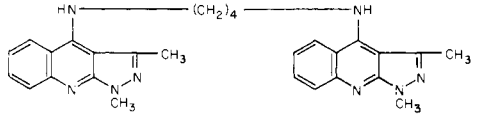
No.	Structure	Mp, °C	Recrystn solvent[a]	Formula[b]	Interferon assay,[c] mg/kg ip	po
60		188–191[d]	A	$C_{10}H_{17}N_3 \cdot 2HCl$[e]	NA	
61		58–60[f]		$C_{14}H_{19}N_3 \cdot 2HCl$	NA	
62		250[g]	E	$C_{18}H_{21}N_3 \cdot 2HCl$	⩾300	NA
63		241–243.5[h]	C	$C_{19}H_{23}N_3 \cdot 2HCl$		NA
64	 a, R = $CO_2C_2H_5$ b, R = CO_2H c, R = H	196–200 dec 268–272 dec 272.5–276 dec	D E D	$C_{16}H_{25}N_5O_2 \cdot 2HCl \cdot 0.5H_2O$[i] $C_{14}H_{21}N_5O_2 \cdot 2HCl$ $C_{13}H_{21}N_5 \cdot 2HCl$		NA (100) NA NA
65		Foam		$C_{17}H_{25}N_5O \cdot HCl$[j]	NA	
66		222–224	C	$C_{17}H_{27}N_5 \cdot 2HCl$	NA	
67		219–235	D	$C_{12}H_{13}N_3 \cdot HCl$[k]		NA
68		215–218.5 dec	B	$C_{12}H_{15}N_3 \cdot HCl$		NA
69		269–271.5 dec	A	$C_{17}H_{27}N_5 \cdot 2HCl$	NA (100)	NA
70		298 dec	E	$C_{16}H_{22}N_6 \cdot 2HCl$	⩾300	
71		279–281 dec	E	$C_{15}H_{21}N_7 \cdot 2HCl$	NA	
72		272–275 dec	D	$C_{16}H_{25}N_7 \cdot 2HCl$		NA
73		265–270 dec	D	$C_{15}H_{23}N_7 \cdot 2HCl$		NA
74		Gum		$C_{17}H_{22}N_4$[l]		200

Table III (*Continued*)

No.	Structure	Mp, °C	Recrystn solvent[a]	Formula[b]	Interferon assay,[c] mg/kg ip	po
75		>300	E	$C_{16}H_{20}N_4S \cdot 2HCl$		NA
76		>300	E	$C_{21}H_{25}N_5 \cdot 2HCl$	100	
77		293–296 dec	D	$C_{20}H_{24}N_6 \cdot 2HCl$	100	100
78		194–195.5	D	$C_{28}H_{30}N_8$	>300	

[a] A, *i*-PrOH; B, MeCN; C, absolute EtOH; D, EtOH; E, aqueous EtOH. [b] See footnote *c* in Table I. [c] Minimal effective dose (mg/kg) required to elicit detectable serum interferon in mice by the indicated route of administration; cf. text for more complete description of assay. NA means not active at 300 mg/kg ip or 400 mg/kg po. NA qualified by a dose level shown in parentheses signifies that toxicity was encountered at higher doses and that the compound was not active at the indicated dose level. [d] Prepared from 4-chloropyridine using the procedure described for compound 172. [e] N: calcd, 16.66, found, 17.13. [f] Lit. mp hydrate (base) 73°; the HCl salt was purified by trituration under Me_2CO; J. K. Landquist, *J. Chem. Soc.*, 1038 (1951). [g] Lit. mp 249–250°: J. Peryt, E. Zylkiewicz, and A. Ledochowski, *Rocz. Chem.*, 43, 623 (1969), *Chem. Abstr.*, 71, 12990b (1969). [h] Lit. mp 243–244°: A. Ledochowski, B. Stefanska, and B. Kozinska, *Rocz. Chem.*, 38, 421 (1964); *Chem. Abstr.*, 61, 1830d (1964). [i] N: calcd, 17.45; found, 18.12. [j] C: calcd, 58.03; found, 59.10. [k] Analyzed as the free base. [l] Not analyzed, cf. Experimental Section.

was cooled and excess amine was evaporated at 5 mm. The residue was dissolved in chloroform and the resultant solution was washed in succession with aqueous K_2CO_3, water, and brine. Drying (Na_2SO_4) and evaporation gave a syrup which was dissolved in 1-propanol (330 ml) containing aqueous 1 *N* HCl (55 ml). The salt **172** was separated by filtration, washed with acetone, and recrystallized from aqueous EtOH: yield 3.78 g (48% based on weight of starting acid or 72% based on estimated content of pure **39** in the starting material); mp 290–293° dec.

Similarly prepared from the appropriate starting acids were **76**, **136–139**, **143–147**, **149** (from **23**), **150** (4-Cl intermediate), **152**, **154**, **155–160**, **162**, **171–176**, **178–180**, **186**, **188–191**, and **194–197**. Similar treatment of IV (R^1 = H; R^2 = $CH_2CO_2C_2H_5$) gave **148**.

1,2,6-Trimethylpyrazolo[5,1-*b*]quinazolin-9(1*H*)-one (VIII, R^1 = 6-CH_3; R^2 = CH_3) hydrochloride was prepared by refluxing the acid **53** for 3 hr in $POCl_3$: mp 245–248° dec after recrystallization (EtOH). Anal. ($C_{13}H_{13}N_3O \cdot HCl$) H, Cl, N; C: calcd, 59.20; found, 58.73.

Similarly prepared from the acid **52** was the 7-chloro relative (VIII, R^1 = 7-Cl; R^2 = CH_3): mp >300°. Anal. ($C_{12}H_{10}ClN_3O \cdot HCl$) C, H, N.

1,3-Dimethyl-4-(4-dimethylaminobutylamino)-1*H*-pyrazolo[3,4-*b*]quinoline Dihydrochloride (92). A mixture of 4-chloro-1,3-dimethyl-1*H*-pyrazolo[3,4-*b*]quinoline[10] (**81**, 4.63 g, 0.02 mol), K_2CO_3 (2.76 g, 0.02 mol), and 4-dimethylaminobutylamine (2.32 g, 0.02 mol) in DMF (60 ml) was stirred at 130° for 16 hr. The solvent was removed at 5 mm and the residue was partitioned between water and chloroform. The organic phase was separated, washed with water, dried, and evaporated to leave an oil (6.02 g, 97%). The dihydrochloride salt was prepared as described above for **172**; recrystallization (EtOH) gave mp 294° dec.

Similarly prepared from **81** and the appropriate amines were **87** (except heating was at 144° for 64 hr), **88**, **91**, **93**, **94**, **97**, **100**, **102**, **106**, **109–113**, **118**, **120**, **121**, **124**, **126**, **127**, **129**, **131**, and **133–135**; also **78** and **79** except 0.5 equiv of amine was used. Similarly prepared from **81** and the indicated nucleophiles were **82** (NaSH), **83** ($PhCH_2SH$), **89** [$Me_2N(CH_2)_3ONa$], and **90** [$Me_2N(CH_2)_3SH$]. Also prepared from **81** using excess of the appropriate amine as solvent and heating at 125–135° for 16 hr were **101**, **114**, **116**, **123**, **125**, and **128**. For a number of the preceding products, standard chromatographic procedures were used for final purification before formation of salts. In some of the preceding examples, the 4-dimethylamino derivative **130** was encountered as a by-product, presumably arising from the DMF.

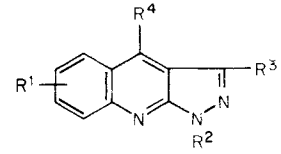
4-(3-Chloropropylamino)-1,3-dimethyl-1*H*-pyrazolo[3,4-*b*]quinoline Hydrochloride (98). A solution of the hydroxy compound **97** (11.50 g, 0.038 mol) and thionyl chloride (110 ml) was heated at 60° for 2 hr. Evaporation gave **98**, mp 210–214° after recrystallization (absolute EtOH).

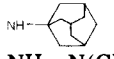
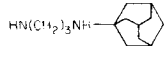
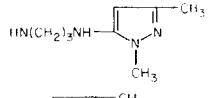
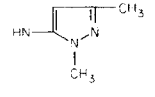
1,3-Dimethyl-4-(3-pyrrolidinopropylamino)-1*H*-pyrazolo[3,4-*b*]quinoline Dihydrochloride (117). A solution of **98** (2.00 g, 5.5 mmol) in pyrrolidine (20 ml) was heated at 80° for 2 hr. Usual work-up gave 117 free base (1.64 g, 92%) from which the salt was made as described above: mp >300° (aqueous EtOH). Similarly prepared from **98** and the appropriate amine except heating was at 80–100° for up to 16 hr in a steel bomb were **103** (absolute EtOH used as solvent), **104**, **105**, **115**, **119**, and **122**. Also prepared from **98** and 1 equiv of appropriate amine and K_2CO_3 in *N*,*N*-dimethylacetamide (DMAC) at 135° were **107** and **108**.

4-Cyclopropylamino-1,3-dimethyl-1*H*-pyrazolo[3,4-*b*]-quinoline Hydrochloride (86). A mixture of **98** (3.25 g, 0.01 mol) and cyclopropylamine (9 ml) was heated in a bomb at 100° for 64 hr. The crude product was purified by chromatography on alumina (elution with 1:1 Et_2O–CH_3Ph), then converted to the HCl salt, and recrystallized (absolute EtOH) to yield **86** (1.57 g, 54%), mp 237–239° dec. Similarly, **98** (2.50 g) in 3-dimethylaminopropylamine (25 ml) was heated at 125° for 16 hr. Work-up gave pure **1** (2.68 g, 98%).

4,9-Dihydro-1,3-dimethyl-1*H*-pyrazolo[3,4-*b*]quinoline Hydrochloride (67) and 1,3-Dimethyl-1*H*-pyrazolo[3,4-*b*]-quinoline Hydrochloride (80). A mixture of **81**[10] (6.00 g, 0.026 mol), $NaOAc \cdot 3H_2O$ (3.54 g, 0.026 mol), and 5% Pd/C (0.5 g) in AcOH (200 ml) was shaken under H_2 at 50 psi of initial pressure for 2.5 hr. Usual work-up gave a product indicated by GLC to be a mixture of **80** (base), **81**, and **67** (base) in an approximate ratio of 47:17:36, respectively. Chromatography on alumina gave initial fractions (elution with toluene) of mixtures of **81** and **80** (base) and later fractions (elution with 98:2 Et_2O–EtOH) of mainly

Table IV. Pyrazolo[3,4-*b*]quinolines



No.	R¹	R²	R³	R⁴	Mp, °C	Recrystn solvent[a]	Formula[b]	Interferon assay,[c] MED (mg/kg) ip	po
1	H	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	306-309 dec	D	$C_{17}H_{23}N_5 \cdot 2HCl$	100	200
79	5-$HN(CH_2)_3N(CH_3)_2$	CH_3	CH_3	H	264-272 dec	A	$C_{17}H_{23}N_5 \cdot 2HCl$		NA
80	H	CH_3	CH_3	H	252-255 dec	B	$C_{12}H_{11}N_3 \cdot HCl$		NA
81	H	CH_3	CH_3	Cl	118-120[d]	C	$C_{12}H_{10}ClN_3$		NA
82	H	CH_3	CH_3	SH	297-299	D	$C_{12}H_{11}N_3S$	NA	
83	H	CH_3	CH_3	SCH_2Ph	156-162	C	$C_{19}H_{18}ClN_3S$		NA
84	H	CH_3	CH_3	NH_2	>320[e]	C	$C_{12}H_{12}N_4 \cdot HCl$		NA
85	H	CH_3	CH_3	$NHCH_3$	>300	E	$C_{13}H_{14}N_4 \cdot HCl \cdot 0.5H_2O$		NA
86	H	CH_3	CH_3	NH-c-C_3H_5	237-239 dec	B	$C_{15}H_{16}N_4 \cdot HCl$	NA (100)	
87	H	CH_3	CH_3		>300	D	$C_{22}H_{26}N_4 \cdot HCl$[f]	NA	
88	H	CH_3	CH_3	NH-c-$N(CH_2CH_2)_2NCH_3$	>290 dec	A	$C_{17}H_{22}N_6 \cdot 2HCl \cdot 1.5H_2O$	300	
89	H	CH_3	CH_3	$O(CH_2)_3N(CH_3)_2$	235-245	F	$C_{17}H_{22}N_4O \cdot HCl \cdot H_2O$	NA (50)	
90	H	CH_3	CH_3	$S(CH_2)_3N(CH_3)_2$	202-204	C	$C_{17}H_{22}N_4S \cdot HCl$	NA (50)	NA (200)
91	H	CH_3	CH_3	$HN(CH_2)_2N(CH_3)_2$	>300	A	$C_{16}H_{21}N_5 \cdot 2HCl$		400
92	H	CH_3	CH_3	$HN(CH_2)_4N(CH_3)_2$	294 dec	D	$C_{18}H_{25}N_5 \cdot 2HCl$		200
93	H	CH_3	CH_3	$HN(CH_2)_5N(CH_3)_2$	269-271 dec	B	$C_{19}H_{27}N_5 \cdot 2HCl$	150	200
94	H	CH_3	CH_3	$HN(CH_2)_6N(CH_3)_2$	215-219	B	$C_{20}H_{29}N_5 \cdot 2HCl$		200
95	H	CH_3	CH_3	$HN(CH_2)_3SCH_3$	145-148	G	$C_{16}H_{20}N_4S \cdot HCl$		NA
96	H	CH_3	CH_3	$HN(CH_2)_3S(O)CH_3$	190-192 dec	F	$C_{16}H_{20}N_4OS \cdot HCl \cdot H_2O$		NA
97	H	CH_3	CH_3	$HN(CH_2)_3OH$	252.5-254.5	D	$C_{15}H_{18}N_4O \cdot HCl$		NA
98	H	CH_3	CH_3	$HN(CH_2)_3Cl$	210-214	B	$C_{15}H_{17}ClN_4 \cdot HCl$		NA
99	H	CH_3	CH_3	$HN(CH_2)_3CO_2^-Na^+$	230-240		$C_{15}H_{15}N_4NaO_2$	NA	
100	H	CH_3	CH_3	$HN(CH_2)_3CO_2CH_3$	133-142 dec	B	$C_{16}H_{18}N_4O_2 \cdot HCl$	NA	
101	H	CH_3	CH_3	$HN(CH_2)_3NH_2$	311-314 dec	A	$C_{15}H_{19}N_5 \cdot 2HCl$	⩾300	
102	H	CH_3	CH_3	$HN(CH_2)_3NHC(=O)H$	222-225 dec	A	$C_{16}H_{19}N_5O \cdot HCl$[g]	NA	
103	H	CH_3	CH_3	$HN(CH_2)_3NHCH_3$	307-309 dec	D	$C_{16}H_{21}N_5 \cdot 2HCl$	150	200
104	H	CH_3	CH_3	$HN(CH_2)_3NHCH(CH_3)_2$	>300	A	$C_{18}H_{25}N_5 \cdot 2HCl$	⩾300	
105	H	CH_3	CH_3	$HN(CH_2)_3NHC(CH_3)_3$	>305	A	$C_{19}H_{27}N_5 \cdot 2HCl$	⩾300	
106	H	CH_3	CH_3	$HN(CH_2)_3NH$-c-C_6H_{11}	307-309 dec	A	$C_{19}H_{29}N_5 \cdot 2HCl$	NA (100)	
107	H	CH_3	CH_3		>300	A	$C_{25}H_{33}N_5 \cdot 2HCl$	NA (100)	
108	H	CH_3	CH_3		288-290 dec	D	$C_{20}H_{25}N_7 \cdot 2HCl$	NA	
109	H	CH_3	CH_3		264 dec	C	$C_{37}H_{18}N_6 \cdot HCl \cdot H_2O$[h]	NA	
110	H	CH_3	CH_3	$HN(CH_2)_3N(C_2H_5)_2$	297-299 dec[i]	A	$C_{19}H_{27}N_5 \cdot 2HCl$	300	400

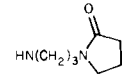
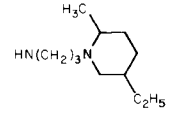
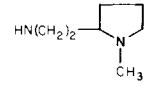
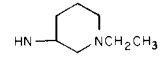
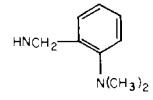
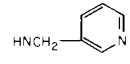
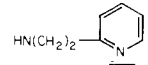
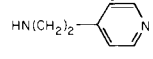
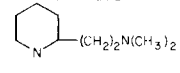
111	H	CH_3	CH_3	$HN(CH_2)_3N(CH_2CH_2 \cdot OH)_2$	244–246.5 dec	A	$C_{19}H_{27}N_5O_2 \cdot 2HCl$	NA		
112	H	CH_3	CH_3	$HN(CH_2)_3N(n\text{-}C_4H_9)_2$	100–105 dec	C	$C_{23}H_{35}N_5 \cdot 2HCl$[j]	NA (100)		
113	H	CH_3	CH_3	$HNCH_2CH(OH)CH_2 \cdot N(C_2H_5)_2$	290 dec	A	$C_{19}H_{27}N_5O \cdot 2HCl$		400	
114	H	CH_3	CH_3	$HNCH(CH_3)(CH_2)_3 \cdot N(C_2H_5)_2$	236–239.5 dec	H	$C_{21}H_{31}N_5 \cdot 2HCl$		NA	
115	H	CH_3	CH_3	$HN(CH_2)_3N(CH_3)(CH_2)_3 \cdot N(CH_3)_2$	263–266	D	$C_{21}H_{32}N_6 \cdot 3HCl$	>300		
116	H	CH_3	CH_3	$HN(CH_2)_2 \cdot c\text{-}NC_4H_8$	295–298 dec	D	$C_{18}H_{23}N_5 \cdot 2HCl$	NA (100)		
117	H	CH_3	CH_3	$HN(CH_2)_3 \cdot c\text{-}NC_4H_8$	>300	A	$C_{19}H_{25}N_5 \cdot 2HCl$[k]		200	
118	H	CH_3	CH_3		234.5–237	B	$C_{19}H_{23}N_5O \cdot HCl$	NA (100)		
119	H	CH_3	CH_3	$HN(CH_2)_3 \cdot c\text{-}NC_5H_{10}$	>300	A	$C_{20}H_{27}N_5 \cdot 2HCl$	NA (100)	200	
120	H	CH_3	CH_3	$HN(CH_2)_3 \cdot c\text{-}N(CH_2CH_2)_2O$	>300	A	$C_{19}H_{25}N_5O \cdot 2HCl$	300	NA	
121	H	CH_3	CH_3	$HN(CH_2)_3 \cdot c\text{-}N(CH_2CH_2)_2NCH_3$	266–270 dec	D	$C_{20}H_{28}N_6 \cdot 2HCl$	300		
122	H	CH_3	CH_3	$HN(CH_2)_3 \cdot c\text{-}N(CH_2CH_2)_2NH$	282–286 dec	A	$C_{19}H_{26}N_6 \cdot 3HCl$	100		
123	H	CH_3	CH_3		180–210 dec	C	$C_{23}H_{33}N_5 \cdot 2HCl$[l]	NA (100)		
124	H	CH_3	CH_3		289–292 dec	A	$C_{19}H_{25}N_5 \cdot 2HCl$	NA (100)		
125	H	CH_3	CH_3		298–305 dec	D	$C_{19}H_{25}N_5 \cdot 2HCl$	⩾300		
126	H	CH_3	CH_3		>300	D	$C_{21}H_{23}N_5 \cdot 2HCl$	NA		
127	H	CH_3	CH_3		303–306 dec	A	$C_{18}H_{17}N_5 \cdot 2HCl$	NA (100)		
128	H	CH_3	CH_3		>300	D	$C_{19}H_{19}N_5 \cdot 2HCl$	NA (50)		
129	H	CH_3	CH_3		>300	A	$C_{19}H_{19}N_5 \cdot 2HCl$	NA (17)		
130	H	CH_3	CH_3	CH_3NCH_3	235	D	$C_{14}H_{16}N_4 \cdot HCl$[m]	NA		
131	H	CH_3	CH_3	$CH_3N(CH_2)_3N(CH_3)_2$	220–224 dec	B	$C_{18}H_{25}N_5 \cdot 2HCl$	⩾300		
132	H	CH_3	CH_3	$C_2H_5N(CH_2)_3N(CH_3)_2$	186–188.5 dec	I	$C_{19}H_{27}N_5 \cdot 2HCl$		NA	
133	H	CH_3	CH_3	$(CH_3)_2N(CH_2)_3N(CH_2)_3N(CH_3)_2$	227–230	I	$C_{22}H_{34}N_6 \cdot 3HCl$	NA		
134	H	CH_3	CH_3		140–150	C	$C_{21}H_{29}N_5 \cdot 2HCl$[n]	NA		

Table IV (*Continued*)

No.	R¹	R²	R³	R⁴	Mp, °C	Recrystn solvent[a]	Formula[b]	Interferon assay,[c] MED (mg/kg) ip	po
135	H	CH_3	CH_3		219-223	D	$C_{21}H_{29}N_5 \cdot 2HCl$	NA (50)	
136	H	H	CH_3	$HN(CH_2)_3N(CH_3)_2$	>270	A	$C_{16}H_{21}N_5 \cdot 2HCl \cdot 0.5H_2O$[o]	NA	
137	H	C_2H_5	CH_3	$HN(CH_2)_3N(CH_3)_2$	278-280 dec	D	$C_{18}H_{25}N_5 \cdot 2HCl$	NA (100)	NA
138	H	CH_2CF_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	285-288 dec	D	$C_{18}H_{22}F_3N_5 \cdot 2HCl$	NA (100)	
139	H	$CH(CH_3)_2$	CH_3	$HN(CH_2)_3N(CH_3)_2$	275 dec	D	$C_{19}H_{27}N_5 \cdot 2HCl$	NA (50)	
140	H	CH_2CH_2OH	CH_3	$HN(CH_2)_3N(CH_3)_2$	251-254	A	$C_{18}H_{25}N_5O \cdot 2HCl$[p]	NA	
141	H	$CH_2CO_2CH_3$	CH_3	$HN(CH_2)_3N(CH_3)_2$	211-212	E	$C_{19}H_{25}N_5O_2 \cdot 2HCl$[q]	NA (100)	
142	H	CH_2CO_2H	CH_3	$HN(CH_2)_3N(CH_3)_2$	295 dec	A	$C_{18}H_{23}N_5O_2 \cdot 2HCl \cdot 2H_2O$[r]	NA	
143	H	CH_2Ph	CH_3	$HN(CH_2)_3N(CH_3)_2$	248-250	E	$C_{23}H_{27}N_5 \cdot 2HCl$	NA (100)	
144	H	Ph	CH_3	$HN(CH_2)_3N(CH_3)_2$	268-271	D	$C_{22}H_{25}N_5 \cdot 2HCl$	NA	
145	H	$-C_6H_4 \cdot p \cdot OCH_3$	CH_3	$HN(CH_2)_3N(CH_3)_2$	220-223 dec	D	$C_{23}H_{27}N_5O \cdot 2HCl$	NA	
146	H	$-C_6H_4 \cdot p \cdot CO_2H$	CH_3	$HN(CH_2)_3N(CH_3)_2$	147-158	J	$C_{23}H_{25}N_5O_2$	NA	
147	H	$-(CH_2)_3N(CH_3)_2$	CH_3	$HN(CH_2)_3N(CH_3)_2$	225-227.5	D	$C_{21}H_{32}N_6 \cdot 3HCl$	NA (100)	NA
148	H	$-CH_2C(=O)NH(CH_2)_3N(CH_3)_2$	CH_3	$HN(CH_2)_3N(CH_3)_2$	283-286 dec	D	$C_{23}H_{35}N_7O \cdot 3HCl$	NA	
149	H	$-CH_2CH_2NH(CH_2)_3N(CH_3)_2$	CH_3	$HN(CH_2)_3N(CH_3)_2$	250-255 dec	D	$C_{23}H_{37}N_7 \cdot 4HCl \cdot 0.5H_2O$	NA (9)	
150	H	$-(CH_2)_3N(CH_3)_2$	CH_3	Cl	240-242	I	$C_{19}H_{19}ClN_4 \cdot HCl$	NA (17)	
151	H	$-(CH_2)_3N(CH_3)_2$	CH_3	H	165-175	I	$C_{16}H_{20}N_4 \cdot 1.7HCl$	NA (100)	
152	H	H	C_2H_5	$HN(CH_2)_3N(CH_3)_2$	233-234	D	$C_{17}H_{23}N_5 \cdot 2HCl$	NA (100)	
153	H	CH_3	C_2H_5	$HN(CH_2)_3N(CH_3)_2$	285-287 dec	D	$C_{18}H_{25}N_5 \cdot 2HCl \cdot 0.5H_2O$	NA (100)	200
154	H	CH_3	H	$HN(CH_2)_3N(CH_3)_2$	280-282 dec	A	$C_{16}H_{21}N_5 \cdot 2HCl \cdot H_2O$		200
155	6-F	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	266-269 dec	D	$C_{17}H_{22}FN_5 \cdot 2HCl$		⩾400
156	7-F	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	268-276 dec	E	$C_{17}H_{22}FN_5 \cdot 2HCl \cdot H_2O$		⩾400
157	6-Cl	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	288-291 dec	D	$C_{17}H_{22}ClN_5 \cdot 2HCl$	NA (100)	
158	7-Cl	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	303-305 dec	A	$C_{17}H_{22}ClN_5 \cdot 2HCl$	⩾100	⩾100
159	8-Cl	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	246-252	D	$C_{17}H_{22}ClN_5 \cdot 2HCl$	NA	
160	6-I	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	298 dec	A	$C_{17}H_{22}IN_5 \cdot 2HCl$	NA (100)	⩾400
161	$5\text{-}NO_2$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	261-265 dec	D	$C_{17}H_{22}N_6O_2 \cdot 2HCl$		NA
162	$6\text{-}NO_2$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	262-264 dec	A	$C_{17}H_{22}N_6O_2 \cdot 2HCl$[s]	NA (100)	
163	$7\text{-}NO_2$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	261-265 dec	A	$C_{17}H_{22}N_6O_2 \cdot 2HCl$	NA	
164	$5\text{-}NH_2$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	>300	D	$C_{17}H_{24}N_6 \cdot 3HCl$[t]	NA	
165	$6\text{-}NH_2$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	>300	D	$C_{17}H_{24}N_6 \cdot 2HCl$[u]	NA	
166	$6\text{-}CH_3C(=O)NH-$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	65-76	K	$C_{19}H_{26}N_6O$	NA	
167	$6\text{-}CH_3CH_2NH-$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	210-214	D	$C_{19}H_{28}N_6 \cdot 3HCl$	25	50
168	$7\text{-}NH_2$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	301-304 dec	A	$C_{17}H_{24}N_6 \cdot 2.5HCl$		NA
169	$7\text{-}CH_3C(=O)NH-$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	171-173.5 dec	C	$C_{19}H_{26}N_6O$		NA
170	$5\text{-}CH_3$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	>300	A	$C_{18}H_{25}N_5 \cdot 2HCl$	NA (100)	NA
171	$6\text{-}CH_3$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	274 dec	D	$C_{18}H_{25}N_5 \cdot 2HCl \cdot 0.5H_2O$	50	50
172	$7\text{-}CH_3$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	290-293 dec	D	$C_{18}H_{25}N_5 \cdot 2HCl$	50	50
173	$8\text{-}CH_3$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	272 dec	D	$C_{18}H_{25}N_5 \cdot 2HCl$	NA (100)	NA
174	$7\text{-}CF_3$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	258-260	D	$C_{18}H_{22}F_3N_5 \cdot 2HCl$	NA (100)	
175	$6\text{-}C_2H_5$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	248-252	D	$C_{19}H_{27}N_5 \cdot 2HCl \cdot H_2O$	NA (100)	200
176	$7\text{-}C_2H_5$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	248-251 dec	I	$C_{19}H_{27}N_5 \cdot 2HCl$	NA (100)	200
177	$7\text{-}CH_2OH$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	296-298 dec	A	$C_{18}H_{25}N_5O \cdot 2HCl$	100	200
178	$6,7\text{-}(CH_3)_2$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	>300	E	$C_{19}H_{27}N_5 \cdot 2HCl$	25	50
179	$7,8\text{-}(CH_3)_2$	CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	240-245 dec	I	$C_{19}H_{27}N_5 \cdot 2HCl \cdot 2H_2O$	200	200

180	6,8-$(CH_3)_2$	CH_3	$HN(CH_2)_3N(CH_3)_2$	251–253 dec	D	$C_{19}H_{27}N_5 \cdot 2HCl$		400
181	6-SO_3H	CH_3	$HN(CH_2)_3N(CH_3)_2$	310–313 dec	D	$C_{17}H_{23}N_5O_3S$	NA	
182	6-SO_2NH_2	CH_3	$HN(CH_2)_3N(CH_3)_2$	194.5–196.5	D	$C_{17}H_{24}N_6O_2S^v$	NA	
183	6-$(CH_3)_2N(CH_2)_3NHSO_2$	CH_3	$HN(CH_2)_3N(CH_3)_2$	233–237 dec	D	$C_{22}H_{35}N_7O_2S \cdot 3HCl$	NA	
184	7-CO_2H	CH_3	$HN(CH_2)_3N(CH_3)_2$	>300	A	$C_{18}H_{23}N_5O_2 \cdot 2HCl$	NA	
185	7-CO_2CH_3	CH_3	$HN(CH_2)_3N(CH_3)_2$	206–210 dec	A	$C_{19}H_{25}N_5O_2 \cdot 2HCl$	NA	
186	7-$(CH_3)_2N(CH_2)_3NH(C{=}O)$	CH_3	$HN(CH_2)_3N(CH_3)_2$	259–267 dec	A	$C_{23}H_{35}N_7O \cdot 3HCl \cdot 2H_2O$	NA	
187	7-$C(=O)NH_2$	CH_3	$HN(CH_2)_3N(CH_3)_2$	282–285 dec	A	$C_{18}H_{24}N_6O \cdot 2HCl \cdot 2H_2O$		NA
188	5-CH_3O	CH_3	$HN(CH_2)_3N(CH_3)_2$	278 dec	A	$C_{18}H_{25}N_5O \cdot 2HCl$	100	100
189	6-CH_3O	CH_3	$HN(CH_2)_3N(CH_3)_2$	281–282 dec	D	$C_{18}H_{25}N_5O \cdot 2HCl$	NA (100)	200
190	7-CH_3O	CH_3	$HN(CH_2)_3N(CH_3)_2$	261–263 dec	D	$C_{18}H_{25}N_5O \cdot 2HCl$	50	50
191	8-CH_3O	CH_3	$HN(CH_2)_3N(CH_3)_2$	261.5–263 dec	D	$C_{18}H_{25}N_5O \cdot 2HCl$	50	100
192	6-HO^w	CH_3	$HN(CH_2)_3N(CH_3)_2$	>300	x	$C_{17}H_{23}N_5O \cdot 2HCl^y$	NA	
193	7-HO^w	CH_3	$HN(CH_2)_3N(CH_3)_2$	301–304 dec	A	$C_{17}H_{23}N_5O \cdot 2HCl^z$	NA	
194	5-Cl, 8-CH_3O	CH_3	$HN(CH_2)_3N(CH_3)_2$	178–180 dec	I	$C_{18}H_{24}ClN_5O \cdot 2HCl$		NA (100)
195	6,7-$(CH_3O)_2$	CH_3	$HN(CH_2)_3N(CH_3)_2$	259–261 dec	D	$C_{19}H_{27}N_5O_2 \cdot 2HCl \cdot 0.5H_2O$	50	50
196	5,7-$(CH_3O)_2$	CH_3	$HN(CH_2)_3N(CH_3)_2$	285 dec	A	$C_{19}H_{27}N_5O_2 \cdot 2HCl$	25	25
197	5,6,7-$(CH_3O)_3$	CH_3	$HN(CH_2)_3N(CH_3)_2$	235–239 dec	E	$C_{20}H_{29}N_5O_3 \cdot 2HCl$		NA (200)

[a] A, aqueous EtOH; B, absolute EtOH; C, MeCN; D, EtOH; E, n-PrOH; F, absolute EtOH–Et_2O; G, MeOH–Et_2O; H, MeOH–Me_2CO; I, i-PrOH; J, DMF; K, H_2O. [b] Cf. footnote c in Table I. [c] Cf. footnote c in Table III. [d] Lit.[10] mp 115–118°. [e] Lit.[10] mp 350°. [f] C: calcd, 69.01; found, 69.62. [g] C: calcd, 57.57; found, 56.87. [h] N: calcd, 23.29; found, 23.76. [i] Lit.[10] mp for free base only. [j] C: calcd, 60.79; found, 60.33. [k] C: calcd, 57.58; found, 57.16. [l] C: calcd, 61.06; found, 59.24. [m] C: calcd, 69.97; found, 69.01. [n] N: calcd, 16.50; found, 17.36. [o] H: calcd, 6.62; found, 7.13. [p] H: calcd, 6.80; found, 6.27. [q] N: calcd, 16.35; found, 15.89. [r] H: calcd, 6.49; found, 6.20. N: calcd, 15.55; found, 14.83. Cl: calcd, 15.74; found, 15.39. [s] C: calcd, 49.17; found, 48.68. [t] C: calcd, 48.41; found, 52.29. N: calcd, 19.93; found, 19.30. [u] C: calcd, 52.99; found, 52.52. [v] N: calcd, 22.32; found, 21.83. [w] Prepared from demethylation of the corresponding methyl ethers using BBr_3 in CH_2Cl_2 by the procedure of J. F. W. McOmie, M. L. Watts, and D. E. West, *Tetrahedron*, 24, 2289 (1968). [x] Insufficient sample for recrystallization. [y] H: calcd, 6.52; found, 5.88. Mass spectrum, M^+ 313. [z] C: calcd, 52.85; found, 50.40. N: calcd, 18.13; found, 17.48. Mass spectrum, M^+ 313.

67 (base). The early fractions (4.30 g) were recrystallized from MeCN to remove the less soluble 81; the filtrate was evaporated and the residue was converted to the HCl salt which was recrystallized (absolute EtOH) to yield pure **80** (1.71 g, 28%), mp 252–255° dec. The latter fractions were recrystallized in succession from MeCN and PhH; the HCl salt then was prepared and recrystallized (EtOH) to yield **67** (1.65 g, 27%), mp 219–235°, indicated by GLC to be 97% pure.

1-(3-Dimethylaminopropyl)-3-methyl-1H-pyrazolo[3,4-b]quinoline Dihydrochloride (151). A solution of **150** (3.03 g, 0.01 mol) in EtOAc (200 ml) and Et_3N (1.39 ml, 0.01 mol) containing 10% Pd/C (0.6 g) was shaken under H_2 at 50 psi until 1 equiv was absorbed (25 min). Usual work-up gave a residue which was triturated under Me_2CO to remove $Et_3N \cdot HCl$. Evaporation of the Me_2CO gave the free amine of the title compound which was treated with 2 equiv of HCl. Recrystallization (i-PrOH) of the resultant di-HCl salt resulted in partial loss of HCl: yield 1.49 g (45%); mp 165–175°. Anal. Calcd for $C_{16}H_{20}N_4 \cdot 1.7HCl$: C, 58.25; H, 6.59; Cl, 18.19; N, 16.98. Found: C, 58.54; H, 6.99; Cl, 18.23; N, 17.03; M^+ 268.

1,3-Dimethyl-5,6,7,8-tetrahydro-1H-pyrazolo[3,4-b]-quinoline Hydrochloride (68). The free amine of the 4-benzylthio compound **83** (3.72 g, 0.0117 mol) in absolute EtOH (150 ml) containing Raney nickel (4 tsp) was stirred at reflux for 64 hr. Usual work-up gave a mixture of products from which the major component was separated by chromatography on alumina (elution with MeCN): yield 0.88 g (38%) from which the HCl salt was prepared; recrystallized (MeCN) to give **68**, mp 215–218.5°.

1,3-Dimethyl-4-(3-dimethylaminopropylamino)-5,6,7,8-tetrahydro-1H-pyrazolo[3,4-b]quinoline Dihydrochloride (69). A mixture of the di-HCl salt of 1[10] (3.70 g, 0.01 mol), NaOAc·$3H_2O$ (2.72 g, 0.02 mol), and PtO_2 in AcOH was shaken under H_2 (50 psi initially) for 16 hr. Usual work-up gave quantitatively the title free base (pure by GLC assay) which was converted to the di-HCl salt and recrystallized (i-PrOH) to yield **69**, mp 269–271.5°.

4-(3-Dimethylaminopropylamino)-3-ethyl-1-methyl-1H-pyrazolo[3,4-b]quinoline Dihydrochloride (153). A mixture of 4-(3-dimethylaminopropylamino)-3-ethyl-1H-pyrazolo[3,4-b]quinoline (**136**, 2.24 g, 0.0076 mol) and NaH (0.0076 mol) dispersion in oil was stirred under N_2 in DMF (32 ml) at 55° for 30 min. Methyl iodide (1.07 g, 0.0076 mol) was added and the mixture was heated at 42° for 18 hr. Usual work-up gave 1.99 g (85%) of **153** free amine containing a small amount of starting material; the latter was removed by chromatography on alumina; the product was converted to the salt and recrystallized to give **153**, mp 285–287° dec.

Similarly prepared from **136** and the indicated alkylating agent were **140** ($BrCH_2CH_2OTHP$)[23] and **141** ($BrCH_2CO_2CH_3$). Basic hydrolysis of **141** gave **142**.

1,3-Dimethyl-4-(3-dimethylaminopropylamino)-5-nitro-1H-pyrazolo[3,4-b]quinoline Dihydrochloride (161). A mixture of the 6-nitro acid **38** (5.00 g, 0.018 mol) and concentrated H_2SO_4 (30 ml) was stirred at 95° for 1.25 hr. The mixture was cooled, poured onto ice, and, after stirring 30 min, basified with concentrated NH_4OH. The precipitate (3.48 g, 75%) was separated by filtration, washed, dried, and then stirred in refluxing $POCl_3$ (40 ml) for 1 hr. Work-up as described above (cf. **172**) gave the 4-chloropyrazolo[3,4-b]quinoline intermediate (2.32 g, 62%). Treatment of this product with equimolar quantities of 3-dimethylaminopropylamine and K_2CO_3 in DMAC at 90° for 4.5 hr gave, after usual work-up, **161**: mp 261–265° dec after recrystallization (EtOH); yield 2.08 g (60%).

1,3-Dimethyl-5-(3-dimethylaminopropylamino)-1H-pyrazolo[3,4-b]quinoline Dihydrochloride (79). A solution of 4-chloro-1,3-dimethyl-5-nitro-1H-pyrazolo[3,4-b]quinoline (7.95 g, 0.029 mol; cf. preceding example), Et_3N (4 ml, 0.029 mol), and 10% Pd/C (0.8 g) in EtOAc (200 ml) was shaken under H_2 at 50 psi for 16 hr. Work-up gave a product recrystallized from CH_3NO_2 to yield 5.08 g (75%) of 5-amino-1,3-dimethyl-1H-pyrazolo-[3,4-b]quinoline, mp 227.5–231.5°. A 3.92-g (0.019 mol) sample of this product in DMAC (80 ml) containing NaH (0.019 mol) was heated at 50° for 1.5 hr. A solution of 3-dimethylaminopropyl chloride (2.31 g, 0.019 mol) in DMAC (60 ml) was added dropwise and the resultant mixture heated at 85° for 16 hr. After usual

work-up, the product **79** was obtained by chromatography on basic alumina (elution with Et_2O): yield 1.40 g (20%).

1,3-Dimethyl-4-(3-dimethylaminopropylamino)-6-nitro-1*H*-pyrazolo[3,4-*b*]quinoline Dihydrochloride (162). A mixture of 70% nitric acid (1.2 ml) and 96% sulfuric acid (13.6 ml) was added dropwise over 30 min to a solution at 5° of **1** (4.91 g, 0.017 mol) in nitromethane (30 ml). After the addition, the mixture was stirred at 10–20° for 2 hr and then was diluted with cold water and neutralized with ammonium hydroxide. The product was isolated by extraction into chloroform and worked up in the usual manner; yield 4.61 g (82%); mp 189–192°. The dihydrochloride salt had mp 262–264° dec (aqueous EtOH), identical with **162** prepared from the acid **37**.

6-Amino-1,3-dimethyl-4-(3-dimethylaminopropylamino)-1*H*-pyrazolo[3,4-*b*]quinoline Dihydrochloride (165) and Derivatives. The above nitro compound **162** (4.00 g, 0.012 mol) suspended in EtOH (100 ml) containing 5% Pd/C (0.4 g) was shaken under hydrogen at an initial pressure of 50 psi for 2.5 hr. The catalyst was removed by filtration and the solvent was evaporated to leave the title compound. It was necessary to store the compound under nitrogen because it rapidly darkens upon exposure to air. The compound was converted to the salt in the usual manner and recrystallized (EtOH): mp 305° dec.

Similarly prepared from **161** and **163** were **164** and **168**, respectively. Treatment of **165** or **168** with 1 equiv of Ac_2O in pyridine at 28° for 16 hr gave the 6- and 7-acetamido derivatives **166** and **169**, respectively. The 6-ethylamino derivative **167** was prepared by reduction of **166** using BH_3 in THF.

1,3-Dimethyl-4-(3-dimethylaminopropylamino)-7-nitro-1*H*-pyrazolo[3,4-*b*]quinoline Dihydrochloride (163). The 4-nitro acid **36** (5.00 g, 0.018 mol) was converted to the 4-chloropyrazolo[3,4-*b*]quinoline derivative VI ($R^1 = 7\text{-}NO_2$; $R^2 = CH_3$) by the procedure described above for **172**. This intermediate (2.76 g, 0.01 mol) was heated at 90° for 4.5 hr in DMAC (30 ml) containing 3-dimethylaminopropylamine (1.02 g, 0.01 mol) and K_2CO_3 (1.38 g, 0.01 mol). Usual work-up gave 3.33 g (97%) which was converted to the salt and recrystallized (aqueous *n*-PrOH) to **163**, mp 261–265° dec.

10,8-Dimethyl-7-(3-dimethylaminopropylamino)-10*H*-pyrido[2,3-*h*]pyrazolo[3,4-*b*]quinoline Dihydrochloride (77). A mixture of the 4-nitro acid **36** (15.00 g, 0.054 mol) and $POCl_3$ (150 ml) was heated at reflux for 2 hr. Excess $POCl_3$ was evaporated (5 mm) and the residue was treated (cautious addition with cooling) with 3-dimethylaminopropylamine (150 ml). The mixture was stirred at 130° for 16 hr and then was worked up as described for **172**. The crude product was chromatographed on alumina (elution with 98:2 CH_2Cl_2–EtOH) to yield 5.3 g which was recrystallized (CH_3NO_2) to mp 137–138.5°; the structural assignment **77** was made on the basis of the spectral and analytical data and was proved by an independent synthesis.[16]

1,3-Dimethyl-4-(3-dimethylaminopropylamino)-6-sulfo-1*H*-pyrazolo[3,4-*b*]quinoline (181) and Derivatives. The di-HCl salt of **1** (3.00 g, 0.008 mol) was added slowly to cold $ClSO_3H$ (15 ml), and then the mixture was heated at 70° for 1 hr. Excess $ClSO_3H$ was removed (5 mm) and the residue was cautiously treated with cold dilute NH_4OH. The resultant precipitate was collected by filtration, washed with water, and recrystallized (EtOH) to yield **181** (2.38 g, 78%), mp 310–313° dec. Similarly prepared were **182** (except the intermediate $6\text{-}SO_2Cl$ was treated with concentrated NH_4OH; yield 47%) and **183** (except treatment was with 3-dimethylaminopropylamine followed by refluxing for 8 hr; yield 64%).

7-Carbomethoxy-1,3-dimethyl-4-(3-dimethylaminopropylamino)-1*H*-pyrazolo[3,4-*b*]quinoline Dihydrochloride (184) and Derivatives. A mixture of *N*-(1,3-dimethylpyrazolo-5-yl)-4-carboxyanthranilic acid (20.0 g, 0.073 mol) and $POCl_3$ (200 ml) was stirred at reflux for 2 hr. Excess $POCl_3$ was evaporated and the residue was divided into two parts: (i) part treated with excess 3-dimethylaminopropylamine as described above for **172** to give **186** (79%); (ii) part treated with excess cold MeOH and then worked up to give VI ($R^1 = 7\text{-}CO_2CH_3$; $R^2 = CH_3$) which was treated with 1 molar equiv of 3-dimethylaminopropylamine in DMF as described above (cf. **92**) to give **184** (58%). Basic hydrolysis of **184** gave the acid **185**. Treatment of **185** with $SOCl_2$ followed by NH_4OH gave **187**.

1,3-Dimethyl-4-(3-dimethylaminopropylamino)-5-ethoxycarbonyl-1*H*-pyrazolo[3,4-*b*]pyridine Dihydrochloride (64a). A mixture of 4-chloro-1,3-dimethyl-5-ethoxycarbonyl-1*H*-pyrazolo[3,4-*b*]pyridine[24] (10.0 g, 0.04 mol), 3-dimethylaminopropylamine (4.24 g, 0.04 mol), and K_2CO_3 (5.45 g, 0.04 mol) in DMAC (100 ml) was stirred at 100° for 10 hr. Usual work-up followed by treatment with aqueous HCl gave **64a** (10.9 g, 70%) after recrystallization (EtOH).

The acid **64b** was prepared by refluxing crude **64a** base in aqueous ethanolic NaOH for 15 hr, followed by usual work-up; the yield of **64b** after recrystallization (aqueous EtOH) was 30%.

1,3-Dimethyl-4-(3-dimethylaminopropylamino)-1*H*-pyrazolo[3,4-*b*]pyridine Dihydrochloride (64c). 1,3-Dimethyl-4-hydroxy-1*H*-pyrazolo[3,4-*b*]pyridine-5-carboxylic acid (3.70 g, 0.018 mol; prepared from the corresponding ethyl ester[24]) was kept at 230° for 20 min. The crude decarboxylated product was heated at reflux in $POCl_3$ (30 ml) for 4 hr. Usual work-up gave the 4-chloro derivative which was dissolved in 3-dimethylaminopropylamine (40 ml) and the resultant solution heated at 115° for 15 hr. The product was chromatographed on basic alumina (elution with 99:1 Et_2O–absolute EtOH) to yield **64c** base (1.02 g, 23%) which was converted to **64c** by treatment with 2 equiv of HCl.

2-(1,3-Dimethylpyrazol-5-ylamino)nicotinic Acid (57). A mixture under nitrogen of 5-amino-1,3-dimethylpyrazole (4.85 g, 0.05 mol) and NaH (0.05 mol) in hexamethylphosphoramide (HMPA, 50 ml) was stirred at 45° for 2 hr. Similarly, 2-chloronicotinic acid (7.85 g, 0.05 mol) and NaH (0.05 mol) were stirred in HMPA (60 ml) at 25° for 2 hr. The two salt solutions were mixed and stirred at 80° for 16 hr. The mixture was poured onto ice water, and the resultant solution was extracted twice with $CHCl_3$ and then was acidified with 1 *N* HCl to precipitate **57** (7.38 g 64%): mp 263–265° dec after recrystallization (EtOH).

***N*-(1,3-Dimethylpyrazol-5-yl)-5-nitroanthranilic acid (37)** was prepared from the sodium salts of 5-amino-1,3-dimethylpyrazole and 2-chloro-5-nitrobenzoic acid in HMPA by the preceding procedure used for **57**. The yield of **37** of mp 275–278° dec was 90%. Recrystallization of an aliquot from a large volume of EtOH gave the analytical sample having mp 278–280° dec.

1,3-Dimethyl-4-(3-dimethylaminopropylamino)-1*H*-pyrazolo[3,4-*b*][1,8]naphthyridine Dihydrochloride (70). A mixture of the acid **57** (5.30 g, 0.023 mol) and $POCl_3$ (60 ml) was heated at reflux for 3 hr. Excess $POCl_3$ was evaporated (5 mm) and the residue was cooled and treated with 3-dimethylaminopropylamine (75 ml). The mixture was heated at 130° for 16 hr and then worked up as described above for **172**: yield 5.10 g (60%); mp 298° dec (aqueous EtOH).

4-(1,3-Dimethyl-5-pyrazolylamino)pyrimidine-5-carboxylic Acid (58). A mixture of 5-carbethoxy-4-chloropyrimidine[25] (18.6 g, 0.1 mol), 5-amino-1,3-dimethylpyrazole (9.70 g, 0.1 mol), and Et_3N (10.1 g, 0.1 mol) in benzene (100 ml) was heated at reflux for 16 hr. Usual work-up and recrystallization (MeCN) gave **58** ethyl ester (7.48 g, 29%) which was hydrolyzed in refluxing aqueous ethanolic NaOH to yield **58** (3.88 g, 58%), mp 284–286° dec (DMF).

1,3-Dimethyl-4-(3-dimethylaminopropylamino)-1*H*-pyrazolo[4',3':5,6]pyrido[2,3-*d*]pyrimidine dihydrochloride (71) was prepared from the acid **58** by the procedure described above for **163** with the exception that in the last step heating was at 80° for 4 hr: yield 74%; mp 276–280° dec.

5-Acetoacetylamino-1,3-dimethyl-4-pyrazolecarboxylic Acid (198). A mixture of 5-amino-1,3-dimethyl-4-pyrazolecarbonitrile[15] (16.32 g, 0.12 mol) and NaOH (30.12 g, 0.754 mol) in H_2O (120 ml) was stirred at reflux for 15 hr. The solution was cooled to −5° and then treated with aqueous HCl (200 ml of 1 *N*; 100 ml of 4 *N*; 25 ml of 1 *N*; added in succession by dropwise addition). Diketene (11.10 g, 0.13 mol) was added dropwise at 15–30°. The mixture was stirred 2 hr at 25° (evolution of CO_2 was detected during addition of the last portions of the diketene at 30°) and then was cooled and acidified. Filtration yielded **198** (10.9 g, 38%), mp 194° dec. An aliquot dissolved in DMF (60°) was filtered and the filtrate diluted with Et_2O to give an analytical sample, mp 198° dec. Anal. ($C_{10}H_{13}N_3O_4$) H, N; C: calcd, 50.21; found, 49.75.

2-Acetonyl-5,7-dimethyl-4,7-dihydropyrazolo[4,3-*d*]-3,1-oxazin-4-one (199). A mixture of the acid **198** (4.94 g, 0.021 mol) and Ac_2O (2.28 g, 0.022 mol) in DMF (50 ml) was stirred at 85°

for 16 hr. The solvent was evaporated (5 mm) and the residue was extracted with CH_2Cl_2 to yield crude **199** (4.24 g) which was recrystallized (MeCN) to yield **199** as white needles (0.76 g, 17%): mp ~130–165° (NMR indicates that in $CDCl_3$ about 75% is in the enol form). Anal. ($C_{10}H_{11}N_3O_3$) C, H, N; M^+ 221.

Bis(1,3-dimethylpyrazol-5-yl)amine (202). In an attempt to obtain the acid **200**, the crude oxazinone **199** was treated with methylhydrazine in AcOH as described above for **39**. The product was chromatographed on neutral alumina (elution with CH_2Cl_2) to give **202** (yield, 39%): mp 156.5–158.5° (MeCN); NMR ($CDCl_3$) δ 2.18 (s, 6, CH_3C), 3.60 (s, 6, CH_3N), 5.50 (s, 2, pyrazole ring CH), 6.14 ppm (s, 1, NH; exchangeable). The sharp single absorbances for the two methyl groups and the ring proton are considered as evidence to support assignment as the symmetrical structure **199** as opposed to the unsymmetrical isomer which would result from decarboxylation of **201**. Anal. ($C_{10}H_{15}N_5$) C, H, N; M^+ 205.

1,7-Dihydro-4-(3-dimethylaminopropylamino)-1,3,5,7-tetramethyldipyrazolo[3,4-b:4',3'-e]pyridine Dihydrochloride (72) and 1,2,5,7-Tetramethyl-1*H*,5*H*-dipyrazolo[1,5-a:5',4'-d]pyrimidin-8-one (203). A mixture of the crude oxazinone **199** (2.24 g, 0.01 mol) and methylhydrazine (0.93 g, 0.02 mol) was stirred in DMF (25 ml) at 80° for 2 hr. The DMF was removed and the residue was partitioned between water containing NaOH (0.01 mol) and $CHCl_3$. Evaporation (4 mm) of the water left a foam (2.7 g). $POCl_3$ (60 ml) was cautiously added with cooling and the mixture was heated at reflux for 2.5 hr. Excess $POCl_3$ was removed and the residue was treated with 3-dimethylaminopropylamine (30 ml). The mixture was heated at reflux for 15 hr and then was worked up as described above (cf. **172**) to yield 1.04 g; chromatography on neutral alumina (elution with CH_2Cl_2) gave in initial fractions **203** (326 mg, 14%), mp 178.5–180.5 (MeCN). Anal. ($C_{11}H_{13}N_5O$) C, H, N; M^+ 231. Later fractions gave **72** free base (265 mg, 8%), M^+ 315, which was converted to the di-HCl salt and recrystallized (EtOH) to **72**, mp 272–275° dec.

The 3-normethyl relative **73** was similarly prepared.

***N*-(3-Methylisothiazol-5-yl)anthranilic Acid (59).** A mixture of *o*-iodobenzoic acid (45.0 g, 0.18 mol), 5-amino-3-methylisothiazole hydrochloride (30.0 g, 0.20 mol), K_2CO_3 (54.8 g, 0.4 mol), and Cu (4.5 g, 1 μ) in water (250 ml) was stirred at reflux for 5 hr. Standard work-up and recrystallization (absolute EtOH) gave **59** (7.38 g, 17%), mp 212–213° dec.

4-(3-Dimethylaminopropylamino)-3-methylisothiazolo[5,4-b]quinoline dihydrochloride (75) was prepared from the acid **59** by the general procedure described above for **172**; purified by chromatography on alumina prior to conversion to the salt, mp >300°.

4-(3-Dimethylaminopropylamino)-1-methylpyrrolo[2,3-b]quinoline (74). A mixture of 4-chloro-1-methylpyrrolo[2,3-*b*]quinoline[26,27] (134 mg, 0.62 mmol) and KI (100 mg, 0.6 mmol) in 3-dimethylaminopropylamine (25 ml) was heated at reflux for 15 hr. The mixture was worked up as described for **172**. Chromatography (neutral alumina; elution with Et_2O plus 0.5% EtOH) gave **74** (86 mg, 49%) as a gum; elemental analyses were not obtained owing to scarcity of material; NMR ($CDCl_3$) δ 1.8 (m, 2, $CH_2CH_2CH_2$), 2.24 [s, 6, $(CH_3)_2N$], 2.45 (t, 2, CH_2N), 3.74 (s, 3, NCH_3), 3.86 (t, 3, $HNCH_2$), 6.59 (d, 1, NCH=CH), 6.87 (d, 1, NCH=CH), 7.0–8.0 (m, 4, aro); M^+ 282.

***N*-(1-Methylpyrazol-5-yl)anthranilic Acid (29).** A mixture of *o*-iodobenzoic acid (20.6 g, 0.08 mol), 5-amino-1-methylpyrazole hydrochloride[14] (12.0 g, 0.09 mol), K_2CO_3 (24.8 g, 0.18 mol), and Cu (2.25 g, 0.035 g-atom; 1-μ mesh) in water (47 ml) was stirred at reflux for 16 hr. Additional water (135 ml) was added; the mixture was refluxed 30 min, then filtered, and worked up in the usual manner; recrystallization (EtOH) gave **29** (7.62 g, 42%), mp 174–176.5°.

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References and Notes

(1) S. Baron, "Interferons and Interferon Inducers", N. B. Finter, Ed., North-Holland Publishing Co., Amsterdam-London, 1973, pp 267–293.
(2) M. N. Oxman in ref 1, pp 391–480.
(3) A. Issacs and J. Lindenmann, *Proc. Roy. Soc., Ser. B*, **147**, 258 (1957).
(4) T. C. Merigan, E. DeClercq, and G. H. Bausek, "Nonviral Interferon Inducers", Proceedings of the New York Heart Association Symposium on Interferon, Little, Brown and Co., Boston, Mass., 1970, pp 57–75.
(5) G. D. Mayer and R. F. Krueger, *Science*, **169**, 1214 (1970).
(6) W. L. Albrecht, R. W. Fleming, S. W. Horgan, B. A. Deck, J. W. Hoffman, and G. D. Mayer, *J. Med. Chem.*, **17**, 1150 (1974), and preceding papers.
(7) P. Siminoff, A. M. Bernard, V. S. Hursky, and K. E. Price, *Antimicrob. Agents Chemother.*, **3**, 742 (1973).
(8) G. Wolfrum, R. Pütter, H. Hanke, and K. Menzel, U.S. Patent 3234142 (1966).
(9) R. Pütter, G. Wolfrum, and H. Hanke, U.S. Patent 3257410 (1966).
(10) R. G. Stein, J. H. Biel, and T. Singh, *J. Med. Chem.*, **13**, 153 (1970).
(11) R. E. Graeve, J. R. Pociask, and R. G. Stein, U.S. Patent 3600393 (1971).
(12) R. Puetter, G. Wolfrum, and K. H. Menzel, German Patent 1139123 (1962); *Chem. Abstr.*, **58**, 9095 (1963).
(13) E. Bulka, H. G. Rohde, and H. Beyer, *Chem. Ber.*, **98**, 259 (1965).
(14) N. J. Cusack, G. Shaw, and G. J. Litchfield, *J. Chem. Soc. C*, 1501 (1971).
(15) E. C. Taylor and K. S. Hartke, *J. Am. Chem. Soc.*, **81**, 2456 (1959).
(16) R. R. Crenshaw, G. M. Luke, and D. F. Whitehead, *J. Heterocycl. Chem.*, in press.
(17) British Patents 1369247, 1369248, 1369249, 1369250 (1974).
(18) W. W. Hoffman, J. J. Korst, J. F. Niblack, and T. H. Cronin, *Antimicrob. Agents Chemother.*, **3**, 498 (1973).
(19) R. G. Douglas and R. F. Betts, *Infect. Immun.*, **9**, 506 (1974).
(20) C. Panusarn, E. D. Stanley, V. Dirda, M. Rubenis, and G. G. Jackson, *N. Engl. J. Med.*, **291**, 57 (1974).
(21) J. Diederich, E. Lodemann, and A. Wacker, *Arch. Gesamte Virusforsch.*, **40**, 80 (1973).
(22) E. T. Glaz, E. Szolgay, I. Stoger, and M. Talas, *Antimicrob. Agents Chemother.*, **3**, 537 (1973).
(23) W. E. Parham and E. L. Anderson, *J. Am. Chem. Soc.*, **70**, 4187 (1948).
(24) H. Hohn, T. Denzel, and W. Janssen, *J. Heterocycl. Chem.*, **9**, 235 (1972).
(25) H. Bredereck, F. Effenberger, and E. H. Schweizer, *Chem. Ber.*, **95**, 803 (1962).
(26) T. Tanaka, T. Iwakuma, M. Miyazaki, M. Wagatsuma, and I. Iijima, *Chem. Pharm. Bull.*, **20**, 109 (1972).
(27) T. Tanaka, T. Iwakuma, M. Wagatsuma, and I. Iijima, *J. Heterocycl. Chem.*, **9**, 1355 (1972).