5-Arylaminopyrimidines[1]

FRED R. GERNS, AGOSTINO PERROTTA, AND GEORGE H. HITCHINGS

Burroughs Wellcome & Co., Inc., The Wellcome Research Laboratories, Tuckahoe, New York

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A series of 5-anilino-2,4-disubstituted pyrimidines was prepared by the reaction of aromatic amines with 5-bromo-4-oxopyrimidines. The 5-anilinouracils were chlorodehydroxylated and aminated to form 5-anilino-2,4-diaminopyrimidines. The latter exhibited moderate antimetabolic effects as folate antagonists and sporadic, unimpressive antibacterial activities.

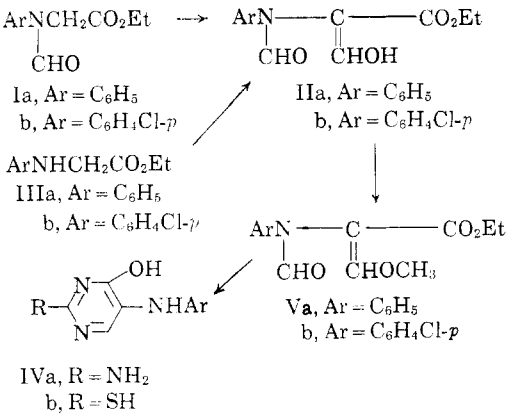
Potent antifolic acid activity (inhibition of folic and dihydrofolic acid reductases[2]) is displayed by 2,4-diaminopyrimidines with a heavy substituent in the 5-positions.[3] Several isologous series of pyrimidines bearing an aryl group either directly in the 5-position or connected to it through O, CH_2, or S have been synthesized previously,[4] and these have yielded compounds with notable antimicrobial activity, *e.g.*, the coccidiostat, diaveridine [2,4-diamino-5-(3,4-dimethoxybenzyl)-pyrimidine][2]; the antibacterial, trimethoprim [2,4-diamino-5-(3,4,5-trimethoxybenzyl)pyrimidine][4e,5]; and the antimalarial, pyrimethamine (2,4-diamino-5-*p*-chlorophenyl-6-ethylpyrimidine).[6] The present paper deals with the further extension of the group to the 5-arylaminopyrimidines.

The first reported syntheses of 5-anilinopyrimidines were those of 5-anilinobarbituric acid and its 2-thio analog by condensation of diethyl anilinomalonate with urea and thiourea, respectively.[7] An extension of this procedure was reported in a recent paper on a series of 2,4,6-trisubstituted and 4,6-disubstituted 5-arylaminopyrimidines.[8] This method, however, does not lend itself to the synthesis of 2,4-diamino derivatives because of the difficulty of subsequent elimination of a 6-hydroxyl group from the pyrimidine ring.

Three approaches to the synthesis of 5-anilino-2,4-diaminopyrimidines have been studied in this laboratory: (a) the condensation of guanidine with α-anilino-β-methoxyacrylonitriles, (b) the interaction of halobenzenes with 5-aminopyrimidines, and (c) the reaction of 5-halopyrimidines with anilines.

The condensation of guanidine with α-aryl-β-alkoxyacrylonitriles is a well-established route to 2,4-diamino-5-substituted pyrimidines.[4e,9] Similarly, α-arylthio-β-alkoxyacrylic esters[10] condense with guanidine to give 5-substituted isocytosines. Compounds of this type may be converted to the 2,4-diamino derivatives by chlorodehydroxylation and amination.[4a-e] This route was investigated with selected α-anilino-β-methoxyacrylonitriles[11] but in each case the condensation of the nitrile with guanidine failed to yield a pyrimidine. Attention was then turned to the preparation of 5-anilinoisocytosines. The intermediate enol esters (II) were prepared here by formylation of N-aryl-N-formylglycine esters (I) or by diformylation of N-arylglycine esters (III) (see Scheme I). The preparation of IIa from Ia has been reported.[12] This compound was not isolated, however. The formyl derivatives (II) did not condense with guanidine. Their enol esters (V), prepared from II by reaction with diazomethane or with dimethyl sulfate and sodium bicarbonate in aqueous dioxane, however, did condense with guanidine with loss of the N-formyl group, to give 5-anilinoisocytosines (IVa), identical with 5-anilinoisocytosine prepared by the method described below. This proved to be a poor intermediate for preparation of the desired 2,4-diamino analog, since on treatment with phosphoryl chloride it yielded intractable phosphorus complexes which did not aminate satisfactorily. The enol esters (V) reacted with thiourea, again with loss of the N-formyl group, to give the 2-mercapto-4-hydroxypyrimidines (IVb). The identity of the products was established by elemental analysis and comparison of ultraviolet absorption spectra with those prepared by the method described below. 5-*p*-Chloroanilino-2-thiouracil was prepared in 67% yield by this method.

SCHEME I



(1) Paper presented in part at the Metropolitan Regional Meeting, of the American Chemical Society, New York, N. Y., Jan. 1962.

(2) G. H. Hitchings in "Drugs, Parasites and Hosts," L. G. Goodwin and R. H. Nimmo-Smith, Ed., J. and A. Churchill Ltd., London, 1962, p. 196.

(3) G. H. Hitchings, G. B. Elion, H. VanderWerff, and E. A. Falco, *J. Biol. Chem.*, **174**, 765 (1948); G. H. Hitchings, E. A. Falco, H. VanderWerff, P. B. Russell, and G. B. Elion, *ibid.*, **199**, 43 (1952).

(4) (a) E. A. Falco, P. B. Russell, and G. H. Hitchings, *J. Am. Chem. Soc.*, **73**, 3753 (1951); (b) E. A. Falco, S. DuBreuil, and G. H. Hitchings, *ibid.*, **73**, 3758 (1951); (c) P. B. Russell and G. H. Hitchings, *ibid.*, **73**, 3763 (1951); (d) E. A. Falco, B. Roth, and G. H. Hitchings, *J. Org. Chem.*, **26** 1143 (1961); (e) B. Roth, E. A. Falco, G. H. Hitchings, and S. R. M. Bushby, *J. Med. Plarm. Chem.*, **5**, 1103 (1962).

(5) G. H. Hitchings and S. R. M. Bushby, Vth International Congress of Biochemistry, Moscow, August 1961, Paper No. 7.42.1167.

(6) E. A. Falco, L. G. Goodwin, G. H. Hitchings, I. M. Rollo, and P. B. Russell, *Brit. J. Pharm.*, **6**, 185 (1951); G. H. Hitchings, *Trans. Roy. Soc. Trop. Med. Hyg.*, **46**, 467 (1952).

(7) T. B. Johnson and N. A. Shepard, *J. Am. Chem. Soc.*, **35**, 994 (1913).

(8) D. E. O'Brien, F. Baiocchi, R. K. Robins, and C. C. Cheng, *J. Med. Chem.*, **5**, 1085 (1962).

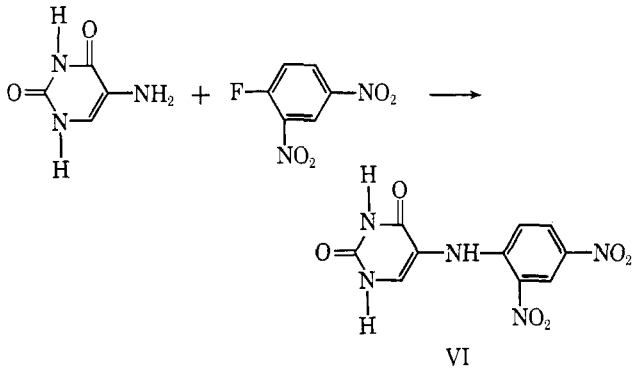
(9) P. B. Russell and N. Whittaker, *J. Am. Chem. Soc.*, **74**, 1310 (1952); B. H. Chase, J. P. Thurston, and J. Walker, *J. Chem. Soc.*, 3439 (1951); B. H. Chase and J. Walker, *ibid.*, 3518 (1953).

(10) B. Roth and G. H. Hitchings, *J. Org. Chem.*, **26**, 2770 (1961).

(11) Prepared by formylation of phenylglycinonitriles followed by O-alkylation in the usual way.

(12) R. G. Jones, *J. Am. Chem. Soc.*, **71**, 644 (1949).

The synthesis of 5-anilinopyrimidines by reaction of aryl halides with 5-aminopyrimidines[13] has only limited application because the aryl halide must be activated. Thus, 2,4-dinitrofluorobenzene was found here to react with 5-aminouracil to yield 5-(2,4-dinitroanilino)-uracil (VI) which was readily chlorodehydroxylated in 66% yield. Subsequent amination afforded the 2,4-diaminopyrimidine derivative.

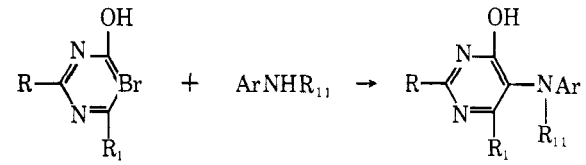


VI

While this work was in progress, a synthesis of VI from dinitrobromobenzene and 5-aminouracil was reported.[8] The ultraviolet absorption spectrum was not in agreement with that found in this laboratory, and since the reaction conditions for the two had been somewhat different, the literature method was repeated here. The compounds, obtained under the two sets of different conditions, were identical on the basis of elemental analysis, ultraviolet spectra, and mixture melting points. These results have been independently confirmed by E. A. Falco (personal communication).

Nucleophilic substitution of the halogen of 5-halopyrimidines by aromatic amines was examined as another approach to the synthesis of 5-arylaminopyrimidines. Apparently this reaction has been successful in only one instance, where aniline reacted with 5-bromo-4-carboxy-2-methylpyrimidine to give the 5-anilino derivative.[13] On the other hand, aniline displaces the ethylthio group preferentially from 5-bromo- and 5-iodo-2-ethylthio-4-hydroxypyrimidine.[14] 5-Arylamino-5,6-dihydrouracils were prepared by the reaction of arylamines with 5-bromo-5,6-dihydrouracil,[15] but their dehydrogenation to 5-anilinouracils could not be effected.[15b] Although various aliphatic amines react with 5-bromouracil (VIIa)[16] and 5-bromoisocytosine (VIIc),[17] aniline is reported not to react with VIIa.[16] It was found by the present authors, however, that aniline *does* react with 5-bromouracil, under somewhat more drastic conditions than those previously employed. Using refluxing ethylene glycol as solvent, VIIa was indeed found to react with a variety of anilines to form 5-anilinouracils. In many cases the yields were excellent.

5-Bromouracil (VIIa) was by far the most reactive of the pyrimidines. Here it was possible to find a rough correlation between the basicity of the arylamines[18] and the yield of anilinouracils (VIIIa). The nitroanilines as well as *o*- and *p*-bromoanilines failed to react. The introduction of *ortho* substituents in one or both reactants and N-alkylation of the arylamines, as well as replacement of the 2-hydroxy function of the pyrimidine by an amino (VIIc) or mercapto group (VIId), retarded the reaction. In such cases, extension of the reflux time or the use of a large excess of the aniline improved the yields. The high temperature necessary for the reaction causes undesirable side reactions and considerable decomposition in VIId and in many of the aromatic amines.

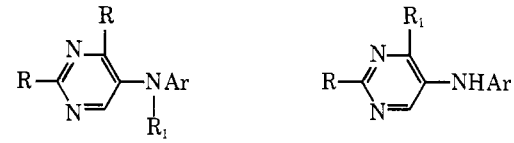


VIIa, R = OH; R_1 = H
b, R = OH; R_1 = CH_3
c, R = NH_2; R_1 = H
d, R = SH; R_1 = H

VIIIa, R = OH; R_1 = R_{11} = H
b, R = OH; R_1 = CH_3; R_{11} = H
c, R = OH; R_1 = H; R_{11} = alkyl
d, R = NH_2; R_1 = R_{11} = H
e, R = SH; R_1 = R_{11} = H

The order of reactivity of the 5-bromopyrimidines toward aromatic amines decreases in the order 5-bromouracil (VIIa) > 5-bromo-6-methyluracil (VIIb) > 5-bromoisocytosine (VIIc) > 5-bromo-2-thiouracil (VIId). The reaction of VIIc with anilines produces 5-anilinouracils (VIIIa) unless sufficient sodium acetate is present to prevent hydrolysis of the 2-amino group. The importance of a 4-hydroxy function to the activation of the 5-bromo group was shown by the failure of the following to react with aromatic amines at temperatures to 190°: 2-hydroxy, 4-amino-2-hydroxy, 2,4-diamino, and 2,4-diamino-6-hydroxy (IX). In compound IX deactivation by the two amino groups on the ring appears to outweigh the effect of the 6-hydroxyl group.

Since 5-bromo-2,4-diaminopyrimidine did not produce the desired 5-anilino derivatives directly, transformation of the corresponding uracils (VIIIa and c) was investigated. Chlorodehydroxylation of VIIIa was effected in yields of 20–36% in refluxing phosphoryl chloride to which had been added either 3–5 moles of water or 2 moles of N,N-diethylaniline/mole of uracil. The N-alkylanilinouracils (VIIIc), on the other hand, gave yields of 65–88% of 2,4-dichloro derivatives (IXa). Treatment of IXa with alcoholic ammonia



IXa, R = Cl; R_1 = H or alkyl
b, R = NH_2; R_1 = H or alkyl

Xa, R = NH_2; R_1 = SH
b, R = R_1 = SH

at 140° in an autoclave afforded the desired 5-anilino-2,4-diaminopyrimidines (IXb). Thiation of VIIId and VIIIe by phosphorus pentasulfide in pyridine afforded Xa and Xb, respectively.

The ultraviolet absorption spectra of the pure forms of three typical 5-anilinouracils are presented in Table I. Protonation of the bases results in a lowering of the

(13) D. M. Besley and A. A. Goldberg, *J. Chem. Soc.*, 4997 (1957).
(14) H. L. Wheeler and H. S. Bristol, *Am. Chem. J.*, **35**, 437 (1905); T. B. Johnson and C. O. Johns, *ibid.*, **34**, 175 (1905).
(15) (a) S. Gabriel, *Ber.*, **38**, 630 (1905); (b) K.-Y. Zee-Cheng, R. K. Robins, and C. C. Cheng, *J. Org. Chem.* **26**, 1877 (1961).
(16) A. P. Phillips, *J. Am. Chem. Soc.*, **73**, 1061 (1951).
(17) A. P. Phillips, *ibid.*, **75**, 4092 (1953).
(18) L. F. Fieser and M. Fieser, "Advanced Organic Chemistry," Reinhold Publishing Corp., New York, N. Y., 1961, p. 709; J. Hine, "Physical Organic Chemistry," 2nd Ed., McGraw-Hill Book Co., Inc., New York, N. Y., 1962, p. 87.

TABLE I

ULTRAVIOLET ABSORPTION AND pK_a VALUES OF 5-ANILINOPYRIMIDINES

Compd.	Species	pK_a	pH	λ_{max}, mμ	$\epsilon \times 10^{-3}$
2	Neutral	...	7.0	242.5, 312	12.2, 3.0
	Cation	−1.34[a]	[b]	262.5	7.7
	Anion	9.12[c]	13.0	237.5, 280	12.6, 8.0
4	Neutral	...	7.0	247, 310[d]	13.1, 3.5
	Cation	−0.66[a]	[b]	260	8.1
	Anion	9.12[c]	11.0	237.5, 282[d]	12.7, 8.1
30	Neutral	...	7.0	246, 288[d]	16.2, 3.9
	Cation	−0.99[a]	[e]	257.5	9.0
	Anion	9.57[c]	11.6	246, 284	15.2, 7.5
42	Neutral	...	5.0	235, 315	12.3, 16.6
	Anion	7.74[c]	10.7	237.5, 297.5	14.1, 14.7
48	Neutral	...	[f]	263, 325, 414.5	12.9, 33.0, 4.8
	Anion	[g]	8.6	248, 307.5	7.3, 11.7
	Dianion	10.9[a]	13.0	282.5, 357.5[d]	9.6, 2.9
60	Neutral	...	10.0	240g, 280[d]	7.5, 4.1
	Cation	7.04[h]	1.0	234	9.6
	Dication	−1.47[h]	[e]	225, 278	8.6, 6.6
71	Neutral	...	10.0	246, 295[d]	8.5, 3.9
	Cation	6.94[a]	5.0	240	10.5
	Dication	−1.49[a]	[h]	232.5, 282	9.4, 6.0
35	Neutral	...	7.0	273	10.2
	Cation	4.18[a]	1.8	238, 273, 332[d]	10.3, 11.6, 5.1

[a] ± approximately 0.1 pH units. [b] 8 *N* HCl. [c] ± approximately 0.05 pH units. [d] Shoulder. [e] 9 *N* HCl. [f] In EtOH. [g] Compound precipitates. [h] 12 *N* HCl.

intensity of the entire spectrum, so that no isosbestic points are present at wave lengths greater than 220 mμ. There is at the same time a bathochromic shift of the main peak. Formation of the monoanion results in a hypsochromic shift of the low wave length peak, and increased absorption in the 280-mμ region of the spectrum. Three isosbestic points are present.

The anilinouracils are weaker bases than diphenylamine (pK_a = 0.85) by factors greater than one H_0 unit (Table I). That protonation occurs first on the anilino nitrogen is suggested by the fact that the parent uracil is a much weaker base (pK_a = 3.38, according to the data of Katritzky and Waring[19]). Furthermore, the anilinouracils are Hammett bases, as indicated by the fact that plots of log ([S]/[SH$^+$]) against H_0 gave straight lines with unit gradient. This is not the case with uracil.[19] As might be expected from steric considerations, the *o*-toluidinouracil is a weaker base than the *p*-toluidino derivative. The introduction of a 6-methyl substituent into the pyrimidine ring also lowers the base strength of the *p*-toluidino derivative. The acidic pK_a values of the anilinouracils are very close to those of the corresponding 5-unsubstituted uracils (uracil, pK_a = 9.38[20]; 6-methyluracil, pK_a = 9.7[21]).

The ultraviolet absorption spectra and pK_a values for thio- and dithiouracil analogs also are presented in Table I. These compounds exhibit the typical absorption of thiones in the 300–330-mμ region of the spectrum as the neutral species, which is decreased in intensity as the mono- and dianions are formed. The acidic pK_a values for these compounds are again very close to those which have been reported for the corresponding 5-unsubstituted thiouracils (2-thiouracil, pK_a = 7.74, 12.7[22]; 2,4-dithiouracil, pK_a = 6.4, 11.2[23]).

The ultraviolet absorption spectra of two typical 5-anilino-2,4-diaminopyrimidines also are given. The neutral species show no well-defined absorption maxima but two nondescript shoulders in the 240- and 280-mμ regions. Monoprotonation results in a hypsochromic shift with lowered intensity in the 290-mμ region and increased intensity of the low wave length peak. Two isosbestic points result. The addition of a second proton increases the intensity once more in the 280-mμ region and creates a well-defined maximum. The low wave length peak, on the other hand, undergoes a hypso- and hypochromic shift.

The basic strength of the 5-anilino-2,4-diaminopyrimidines is lower than that of 2,4,5-triaminopyrimidine (pK_a = 7.63, 2.56[24]), as is to be expected from the introduction of the electron-withdrawing aromatic group, and is similar to that of 2,4-diaminopyrimidine (pK_a = 7.26[25a]; no second pK_a value reported). The pK_a value representing the second protonation for either of these two pyrimidines is approximately one H_0 value lower than that representing the first protonation of the corresponding uracil compound **2**. Methylation on the aniline nitrogen does not appreciably affect the basicity. These pyrimidines also behave like Hammett bases in this region of acidity, although the changes in absorption with acidity are not large, rendering quantitative interpretation more difficult.

Spectral data and pK_a values for one 5-anilinoisocytosine are reported (Table I). The basic pK_a value for this compound is very similar to that for the parent isocytosine (pK_a = 4.01[25b]). The acidic pK_a value could not be determined with any degree of accuracy because of instability of the compound in alkali. This is a common characteristic of 2,5-diaminopyrimidines which oxidize rapidly to dark red products in alkaline medium.

Antimetabolic Effects.—The 5-anilinopyrimidines show only moderate inhibitory effects in the *Lactobacillus casei* system (Table II). Among the uracil derivatives (**1**, **16**, VI, **33**), only the dinitro derivative VI is inhibitory at the 100-μg./ml. level and it is essentially inactive at a concentration of 5 μg./ml. A 2-mercapto-4-hydroxy derivative (**44**) and an isocytosine derivative (**38**) also were essentially inactive. A somewhat greater level of activity was shown by the dimercapto derivative (**49**) but it also failed to inhibit at a concentration of 5 μg./ml. The diamino derivatives (**58**–**66**) were more potent. All were inhibitory at a concentration of 5 μg./ml., and all showed one of the characteristics of folate antagonism, *i.e.*, reversal of the effects by folate as evidenced by lesser inhibitions in the high folate (FA_+) than in the low folate (OFA) medium. None of these, however, exhibited the bypass reversal (growth in PT medium) characteristic of uncomplicated folate antagonism. The dinitrodiamino derivative (**67**) was the least active of the group, in contrast to the corresponding dihydroxy derivative which was

(19) A. R. Katritzky and A. J. Waring, *J. Chem. Soc.*, 1540 (1962).
(20) A. Albert and J. W. Phillips, *ibid.*, 1294 (1956).
(21) J. R. Marshall and J. Walker, *ibid.*, 1004 (1951).
(22) H. N. Christensen, *J. Biol. Chem.*, **160**, 425 (1945); D. Shugar and J. J. Fox, *Bull. soc. chim. Belges*, **61**, 293 (1952).
(23) D. J. Brown, *J. Appl. Chem.* (London), **9**, 203 (1959); H. G. Mautner, *J. Am. Chem. Soc.*, **78**, 5292 (1956).
(24) S. F. Mason, *J. Chem. Soc.*, 2071 (1954).
(25) A. Albert, R. Goldacre, and J. Phillips, *ibid.*, 2240 (1948); (b) P. A. Levene, L. W. Bass, and H. S. Simms, *J. Biol. Chem.*, **70**, 229 (1926).

TABLE II

GROWTH INHIBITORY ACTIVITY OF 5-ANILINOPYRIMIDINES *vs. L. casei*

Compd.	Concn., μg./ml.	Inhibition (%) in medium indicated— OFA	FA_+	PT	PFA
6	100	−23	0	0	0
16	100	−30	0	−20	0
VI	100	−86	−100	0	0
	5	−29	−9	0	0
33	100	−16	0	−27	0
44	100	−23	0	0	0
38	100	0	0	0	0
49	100	−92	−44	−85	−70
	5	−7	0	15	−8
58	100	−96	−95	−50	−95
	5	−58	0	−27	−31
59	100	−96	−94	−87	−94
	5	−81	−65	−74	−68
60	100	−85	−90	−84	−93
	5	−45	−10	−37	−57
61	100	−95	−94	−91	−93
	5	−74	−34	−40	−67
62	100	−94	−96	−92	−96
	5	−96	−87	−92	−96
	1	−69	−30	−70	−70
	0.1	0	−24	−22	−25
63	100	−93	−96	0	−93
	5	−64	−14	−20	−73
64	100	−91	−93	−87	−93
	5	−69	−16	−21	−70
65	100	−90	−90	−82	−96
	5	−75	−13	−10	−72
66	100	−96	−97	−80	−94
	5	−90	−71	−61	−84
	1	−56	0	0	−37
67	100	−56	−18	0	−58
	5	−12	0	0	−10
69	100	−83	−21	−58	−61
	5	−42	0	0	−10
70	100	−49	0	33	−46
	5	0	0	0	0

the most active of the substituted uracils. Substitution of the anilino nitrogen diminished activity in both of the cases tested (**69** and **70**).

Antibacterial Activities.—Although activities of a minimal nature were exhibited sporadically by the 5-anilino-2,4-diaminopyrimidines, the results on the whole were unimpressive in comparison to those of other groups of 5-substituted pyrimidines (*e.g.*, 5-benzylpyrimidines[4e]) (see Table III).

TABLE III

ANTIBACTERIAL EFFECTS OF 5-ANILINOURACILS BY DISK–PLATE METHOD

Compd.	Zones of inhibition (mm.) of bacteria[a] Sa/s	Sa/r	S.f.	E.c.	A.a.	P.v.	Ps.a.
58	0	0	24	0	0	0	0
59	12	13	20	0	12	12	0
60	0	0	30	12	0	12p	0
61	0	0	33	0	12	12p	0
63	0	0	33	12	0	12	Tr
64	0	0	0	0	0	0	0
65	0	0	36	12	12	15p	0
66	15	13	0	0	0	0	11p
70	0	0	17	0	0	13	0

[a] Sa/s = *Staphylococcus aureus* CN 209, Sa/r = *Staphylococcus aureus* Bennett, S.f. = *Streptococcus faecalis* ATCC 6043, E.c. = *Escherichia coli* CN 601, A.a. = *Aerobacter aerogenes* ATCC 8308, P.v. = *Proteus vulgaris* ATCC 9920, Ps.a. = *Pseudomonas aeruginosa* P-20.

Antitumor Activities.—None of the 5-anilino-2,4-diaminopyrimidines showed reproducible activity against S180 or Ad755 in mice. A few of the 5-anilinouracils exhibited activity against S180, but failed to show activity against Ad755. The results of the tests of compounds active against S180 are shown in Table IV.

TABLE IV

ACTIVITIES OF 5-ANILINOURACILS AGAINST SARCOMA 180

Compd.	Dose, mg./kg.	TWI (T/C)[a]	T/N[b]	Toxicity[c]	BWI[d] (T/C)
17	50	1.01	6/6	0/6	0.92
	100	0.20	4/6	0/6	0.98
	200	Toxic			
4	100	0.54	6/6	0/6	0.87
	150	0.28	5/6	0/6	0.98
	250	0.21	6/6	0/6	1.23
	250	0.66	6/6	0/6	1.10
	400	0.44	5/6	0/6	0.96
8	50	0.51	6/6	0/6	1.14
	100	0.39	4/5	1/6	0.98
	100	0.30	5/5	1/6	0.91
VIa	25	0.41	6/6	0/6	1.07
	50	0.26	6/6	0/6	0.94
	100	Toxic			

[a] TWI = tumor weight index; ratio of tumor weights of treated/control. [b] T/N = tumors/number of animals at termination. [c] Toxicity = number dying of toxicity/number in group. [d] BWI = body weight index; ratio of animal weights at termination, treated/control.

Experimental Section[26]

Antibacterial Tests.—Filter paper disks (10 mm. in diameter) were saturated with solutions of the compounds to be tested (1 mg./ml.) and applied to agar plates seeded with the appropriate organism. After incubation of the plates, the diameters of the zones of inhibition were measured. The disks retain *ca.* 0.02 ml. of solution, and, therefore, approximately 20 μg. of compound.

pK_a Determinations.—pK_a values were determined spectrophotometrically using the procedure of Roth and Schloemer.[28] Values below pH 1 were determined in HCl of varying normalities, and the H_0 scale of Paul and Long[29] was used as the acidity function. In the cases of pK_a values in the H_0 range, curves were plotted at a series of acidities, and the optical densities at single wave lengths where the greatest differences occurred were plotted against the acidity function. Sigmoid curves were obtained from which the midpoints could be used to estimate the pK_a values, and it was found that the calculated curves for the pK_a values in question matched the experimental points.[30] Similarly, plots of the log of fractional protonation *vs.* H_0 produced straight lines of unit gradient, indicating that the compounds were Hammett bases, and that the H_0 scale was satisfactory for use here. The anilinouracils were somewhat unstable in alkali, which renders the extinction coefficients and hence acidic pK_a values open to some uncertainty. It appeared that a second proton was lost between pH 13 and 14, but it was not possible to give any closer estimate of this second pK_a value, since the spectra did not change sufficiently with pH in this region and the stability was also too serious.

(26) Melting points (corrected) were taken in an open capillary, using a silicone bath. Above 260° a Melt-Meter® was used. Ultraviolet absorption spectra were determined on a Beckman DU spectrophotometer with matched quartz cuvettes. Infrared absorption spectra were determined from pressed potassium bromide disks on a Beckman IR-4 spectrophotometer. The use of *Lactobacillus casei* for the evaluation of potential antagonists of folic acid, purines, and thymine has been described in previous publications.[27]

(27) G. H. Hitchings, G. B. Elion, E. A. Falco, P. B. Russell, and H. VanderWerff, *J. Biol. Chem.*, **183**, 1 (1950); G. H. Hitchings, *Ann. N. Y. Acad. Sci.*, **76**, 490 (1958).

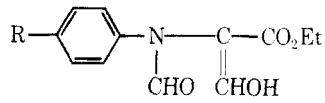
(28) B. Roth and L. A. Schloemer, *J. Org. Chem.*, **28**, 2659 (1963).

(29) M. A. Paul and F. A. Long, *Chem. Rev.*, **57**, 12 (1957).

(30) B. Roth and J. F. Bunnett, *J. Am. Chem. Soc.*, **87**, 334 (1965).

TABLE V

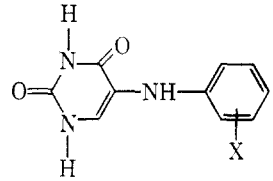
ETHYL α-(N-FORMANILIDO)-β-HYDROXYACRYLATES (II)



Compd.	R	Route	Yield, %	M.p., °C.	Formula	Calcd., % C	Calcd., % H	Calcd., % N	Found, % C	Found, % H	Found, % N
IIa	H	A	40	99–101	$C_{12}H_{13}NO_4$	61.27	5.57	5.95	61.64	5.47	5.95
IIb	Cl	A	85	102–104	$C_{12}H_{12}ClNO_4$	53.44	4.48	5.19	53.16	4.17	5.15
IIb	Cl	B	56	102–104	$C_{12}H_{12}ClNO_4$	53.44	4.48	5.19	53.38	4.29	5.29

TABLE VI

5-ANILINOURACILS



Compd.	X	Conditions[a]	Yield, %	M.p., °C.[b]	Formula	Calcd., % C	Calcd., % H	Calcd., % N	Found, % C	Found, % H	Found, % N
1	H	2	76	317–319	$C_{10}H_9N_3O_2$	59.11	4.46	20.68	59.11	4.53	20.56
2	2-CH_3	4	56	299–300	$C_{11}H_{11}N_3O_2$	60.82	5.10	19.35	60.58	5.09	19.26
3	3-CH_3	2	53	324–326	$C_{11}H_{11}N_3O_2$	60.82	5.10	19.35	60.62	4.89	19.16
4	4-CH_3	2	91	323–326	$C_{11}H_{11}N_3O_2$	60.82	5.10	19.35	60.62	5.05	19.25
5	2,4,6-$(CH_3)_3$	4	25	314–317	$C_{13}H_{15}N_3O_2$	63.40	6.55	17.06	63.76	6.21	17.36
6	2-Cl	4	12	309–311	$C_{10}H_8ClN_3O_2$	50.54	3.39	17.68	50.65	3.03	17.42
7	3-Cl	2	24	304–305	$C_{10}H_8ClN_3O_2$	50.54	3.39	17.68	50.73	3.10	17.75
8	4-Cl	2	45	338–340	$C_{10}H_8ClN_3O_2$	50.54	3.39	17.68	50.65	3.47	17.64
9	3-Br	2	27	310–311	$C_{10}H_8BrN_3O_2$	42.57	2.86	14.90	42.65	2.78	15.22
10	4-F	2	66	327–329	$C_{10}H_8FN_3O_2$	54.30	3.65	19.00	54.28	3.65	18.80
11	3,4-$(Cl)_2$	2	31	325–326	$C_{10}H_7Cl_2N_3O_2$	44.14	2.59	15.44	44.17	2.44	15.48
12	2-CH_3-3-Cl	4	16	310–311	$C_{11}H_{10}ClN_3O_2$	52.50	4.01	16.70	52.56	4.00	16.67
13	2-CH_3-4-Cl	4	19	317–318	$C_{11}H_{10}ClN_3O_2$	52.50	4.01	16.70	52.88	3.89	16.90
14	2-OCH_3	4	50	300–302	$C_{11}H_{11}N_3O_3$	56.65	4.75	18.02	56.74	4.76	17.92
15	3-OCH_3	2	54	295–297	$C_{11}H_{11}N_3O_3$	56.65	4.75	18.02	56.87	5.14	18.06
16	4-OCH_3	2	88	312–315	$C_{11}H_{11}N_3O_3$	56.65	4.75	18.02	56.55	4.74	17.80
17	3,4,5-$(OCH_3)_3$	2	28	271–273	$C_{13}H_{15}N_3O_5$	53.24	5.16	14.33	53.28	5.08	14.47
18	2,4-$(OCH_3)_2$	4	67	279–281	$C_{12}H_{13}N_3O_4$	54.75	4.98	15.96	54.90	5.02	15.92

[a] In all cases 3.5 moles of arylamine/mole of 5-bromouracil was used. Figures in this column refer to reflux time in hours. [b] With decomposition. Recrystallized from acetic acid.

N-Aryl-N-formylglycine Ethyl Esters (I).—The formyl esters were prepared by reaction of the sodium salt of the formanilides and ethyl chloracetate according to known procedures.[31] The use of NaOH in toluene was found to facilitate the reaction. Ia was obtained as an oil, b.p. 110–111 (0.1 mm.), lit.[31] b.p. 290–295°.

The ***p*-chloro compound (Ib)** melted at 56–57° (from benzene–petroleum ether); λ_{max} 2.88, 3.32, 5.68, 5.88, 6.22 μ.

Anal. Calcd. for $C_{11}H_{12}ClNO_3$: C, 54.67; H, 5.00; N, 5.80. Found: C, 54.71; H, 4.51; N, 5.66.

Ethyl α-(N-Formanilido)-β-hydroxyacrylates (II, Table V). A. By C-Formylation of N-Formyl Esters (I).—The method used has been described.[12] The diformyl esters were recrystallized from benzene–petroleum ether; IIb has λ_{max} 2.85, 3.15, 3.27, 5.9, 6.21, 6.62 μ.

B. By Diformylation of Arylglycine Esters (III).—The preparation of the *p*-chloro derivative (IIb) is presented as an example. To a solution of 21.3 g. (0.1 mole) of N-*p*-chlorophenylglycine ethyl ester (IIIb)[32] and 37.0 g. (0.5 mole) of ethyl formate (dried over anhydrous K_2CO_3) in 60 ml. of dry benzene was added 2.3 g. (0.1 g.-atom) of sodium. The mixture was stirred under nitrogen for 6 hr. then refrigerated for 3 days. Water was added, followed by $NaHCO_3$ solution. The organic phase was extracted with fresh $NaHCO_3$ solution. The combined aqueous phase was then extracted with ether and acidified. The oil which separated was taken up in ether, washed with water, and dried. After elimination of the solvent, a solid was obtained, m.p. 95–97°. The analytical sample was obtained by recrystallization from benzene–petroleum ether: λ_{max} 2.85, 3.15–3.3, 5.9, 6.21, 6.62 μ.

Ethyl α-(N-Formanilido)-β-methoxyacrylates (V).—This procedure, which is essentially that of Baltzly and Russell[33] is exemplified by the preparation of **ethyl α-(N-formyl-p-chloroanilido)-β-methoxyacrylate (Vb).** A mixture of 46 g. (0.17 mole) of IIb, and 43 g. (0.51 mole) of $NaHCO_3$ in 135 ml. of dioxane and 13 ml. of water was stirred at room temperature for 1 hr. Dimethyl sulfate (43 g., 0.34 mole) was added, and the reaction flask was immersed in a water bath at 75°. The mixture was vigorously stirred as the temperature of the bath was raised to 90°. After 3.5 hr., the reaction mixture was cooled, poured on ice, and extracted with ether. The ether extract was washed once with 10% $NaHCO_3$ and three times with water and dried ($MgSO_4$). Evaporation of the ether gave the crude product which was triturated with hexane and filtered to give 48.3 g. (100%) of cream-colored solid, melting at 88–90°. An analytical sample was obtained by recrystallization from benzene–hexane. It melted at 92–94°; λ_{max} 2.85, 3.30, 5.78–5.83, 5.97, 6.21, 6.62 μ.

Anal. Calcd. for $C_{13}H_{14}ClNO_4$: C, 55.03; H, 4.97. Found: C, 55.02; H, 4.60.

Ester Va was obtained only as a crude oil.

Synthesis of 5-Anilinopyrimidines *via* Acrylates (V). 5-*p*-Chloranilino-2-thiouracil (43, Table IX).—A solution of 30 g. (0.106 mole) of Vb, 8.4 g. (0.11 mole) of thiourea, and 12.0 g. (0.22 mole) of commercial sodium methoxide in 100 ml. of methanol was refluxed for 7 hr. The solution was concentrated to dryness *in vacuo*, and water was added to the residue, followed by

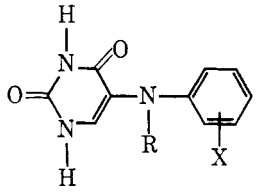
(31) C. Paal and G. Otten, *Ber.*, **23**, 2587 (1890).

(32) W. Baker, W. D. Ollis, and V. D. Poole, *J. Chem. Soc.*, 307 (1949).

(33) R. Baltzly and P. B. Russell, *J. Org. Chem.*, **21**, 912 (1956).

TABLE VII

5-N-SUBSTITUTED ANILINOURACILS

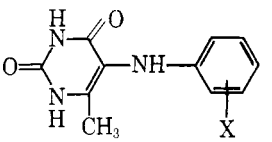


Compd.	R	X	Conditions[a]	Yield, %	M.p., °C. dec.	Formula	Calcd., % C	Calcd., % H	Calcd., % N	Found, % C	Found, % H	Found, % N
19	$COCH_3$		...	91	276–278[b]	$C_{12}H_{11}N_3O_3$	58.77	4.52	17.13	59.13	4.57	17.10
20	$COCH_3$	4-Cl	...	99	247.5–250.5[b]	$C_{12}H_{10}ClN_3O_3$	51.53	3.60	15.02	51.47	3.43	14.90
21	CH_3		5	47	261–263[c]	$C_{11}H_{11}N_3O_2$	60.82	5.10	19.35	60.65	4.86	19.20
22	C_2H_5		5	52	294–297[c]	$C_{12}H_{13}N_3O_2$	62.32	5.67	18.17	62.12	5.69	18.02
23	n-C_4H_9		6	34	273–274.5[c]	$C_{14}H_{17}N_3O_2$	64.85	6.61	16.20	64.70	6.16	16.42
24	CH_3	2-CH_3	6	22	293–294[c]	$C_{12}H_{13}N_3O_2$	62.32	5.67	18.17	62.57	5.73	18.01
25	CH_3	4-Cl	4.5	27	271–273.5[c]	$C_{11}H_{10}ClN_3O_2$	52.50	4.01	16.70	52.18	3.97	17.03
26	CH_3	2-OCH_3	6	63	298–300	$C_{12}H_{13}N_3O_3$	58.29	5.30	17.00	58.45	5.24	16.91

[a] Compounds **19** and **20** were prepared as described below. Compounds **21–26** were prepared from 3.5 moles of N-alkylaniline/mole of 5-bromouracil. Figures in this column refer to reflux time in hours. [b] Recrystallized from aqueous ethanol. [c] Recrystallized from aqueous 2-methoxyethanol.

TABLE VIII

5-ANILINO-6-METHYLURACILS



Compd.	X	Conditions[a]	Yield, %	M.p., °C.[b]	Formula	Calcd., % C	Calcd., % H	Calcd., % N	Found, % C	Found, % H	Found, % N
27		6	27	315–317	$C_{11}H_{11}N_3O_2$	60.82	5.10	19.35	61.09	4.76	19.22
28	2-CH_3	4	20	320–324	$C_{12}H_{13}N_3O_2$	62.32	5.67	18.17	62.59	5.71	18.02
29	3-CH_3	2	53	272–275.5	$C_{12}H_{13}N_3O_2$	62.32	5.67	18.17	62.35	5.89	18.13
30	4-CH_3	2	42	319–321	$C_{12}H_{13}N_3O_2$	62.32	5.67	18.17	62.50	5.34	18.34
31	3-Cl	2	15	305–308	$C_{11}H_{10}ClN_3O_2$	52.50	4.01	16.70	52.24	4.00	16.57
32	4-Cl	2	12	321–322	$C_{11}H_{10}ClN_3O_2$	52.50	4.01	16.70	52.54	3.67	16.95
33	4-OCH_3	2	60	278–280	$C_{12}H_{13}N_3O_3$	58.29	5.30	17.00	58.23	4.97	17.09

[a] In all cases 10 moles of the aniline/mole of 5-bromo-6-methylureacil was used. Figures in this column refer to reflux time in hours. [b] With decomposition. Recrystallized from acetic acid.

acetic acid. The crude product was collected by filtration and washed with water, methanol, and ether. A cream-colored solid (18 g., 67%) was obtained. It was purified by solution in dilute alkali, followed by treatment with charcoal and reprecipitation with acetic acid. Several recrystallizations from aqueous 2-methoxyethanol gave the analytically pure product.

5-Anilinoisocytosine.—A warm mixture of 107 g. (1.12 moles) of guanidine hydrochloride and 90.5 g. (1.68 moles) of commercial sodium methoxide in 300 ml. of methanol was stirred for 1 hr. and filtered through diatomaceous earth. The filter cake was washed with additional methanol. To the filtrate was added a solution of 140 g. (0.56 mole) of ethyl α-(N-formanilido)-β-methoxyacrylate in 100 ml. of methanol. The solution became hot. It was refluxed for 4 hr. After standing at room temperature overnight the mixture was concentrated to dryness at reduced pressure. The residue was taken up in hot, dilute alkali, cooled, filtered clear, and treated with charcoal. The product was reprecipitated by the addition of acetic acid to pH 5.5. Reprecipitation from alkali gave a yellow solid, 60 g. Several recrystallizations from aqueous 2-methoxyethanol gave an analytical sample, melting at 294–295°, undepressed by admixture with compound prepared by general method (see below); λ_{max} 2.1–2.8, 3.15, 5.8–6.8, 6.23, 6.5–7.5 μ; ultraviolet absorption characteristics were as follows [mμ ($\epsilon \times 10^{-3}$)]: (a) 0.1 N HCl, λ_{max} 235 (9.5), 272 (11.2), 332 sh (4.7); λ_{min} 222 (8.6), 246.5 (8.9); (b) 0.1 N NaOH, λ_{max} 240 (11.8), 280 sh (8.5); λ_{min} 222.5 (10.4).

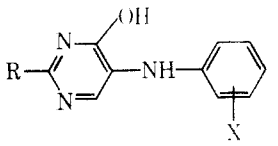
Anal. Calcd. for $C_{10}H_{10}N_4O$: C, 59.40; H, 4.98; N, 27.71. Found: C, 59.75; H, 4.99; N, 27.70..

General Method of Synthesis of 5-Anilinopyrimidines by Reaction of 5-Bromopyrimidines with Aromatic Amines.—An excess of the appropriate aniline (distilled from zinc) and 5-bromopyrimidine (VII) were heated at 185–200° in 5–10 vol. of refluxing ethylene glycol for the 2–6 hr. in the presence of hydroquinone. Nitrogen was introduced below the surface of the reaction mixture to provide agitation and to minimize oxidation. On cooling to room temperature, the reaction mixture often solidified. In any case, it was diluted with several volumes of water to precipitate all of the product. The crude product was washed with ether and acetone to remove excess anilines and dissolved in alkali. In some cases the aqueous alkaline solution required additional ether extraction. After treatment with charcoal, the product was reprecipitated with acid, washed with water, and dried. The yields indicated in Tables VI–IX were calculated at this point for the 5-anilinopyrimidines prepared by this method. Further purification was accomplished by a second reprecipitation from alkali and repeated recrystallization from the appropriate solvent (see tables). These compounds were obtained as white or off-white crystalline solids, with the exception of the 2-thio derivatives which were pale yellow to yellow. The general method is exemplified by the synthesis of 5-anilinouracil and 5-anilinoisocytosine.

5-Anilinouracil (1, Table VI).—A solution of 250 g. (1.3 moles) of 5-bromouracil and 363 g. (3.9 moles) of aniline in 2.5 l. of ethylene glycol was refluxed for 2 hr. at 190–195° in a nitrogen atmosphere. The product precipitated on cooling. The reaction mixture was diluted with a large volume of water and refrigerated. The product was collected by suction filtration and washed with water and acetone. It was further purified by solution in aqueous base, followed by treatment with charcoal, and reprecipitation with concentrated HCl to give 201.4 g. (76%) of off-white needles. Several recrystallizations from glacial acetic acid gave the product as the free base (colorless needles).

TABLE IX

2-AMINO- AND 2-MERCAPTO-4-HYDROXY-5-ANILINOPYRIMIDINES



Compd.	R	X	Conditions[a]	Yield, %	M.p., °C.[b]	Formula	Calcd., % C	Calcd., % H	Calcd., % N	Found, % C	Found, % H	Found, % N
34	NH_2		2[c,d]	43	294–295	$C_{10}H_{10}N_4O$	59.40	4.98	27.71	59.13	4.88	27.71
35	NH_2	3-CH_3	2[c]	40	291–293	$C_{11}H_{12}N_4O$	61.10	5.59	25.91	61.08	5.35	25.77
36	NH_2	4-CH_3	2[c]	40	277–280	$C_{11}H_{12}N_4O$	61.10	5.59	25.91	61.35	5.68	25.84
37	NH_2	3-Cl	4.5[c]	25	292–295	$C_{10}H_9ClN_4O$	50.75	3.83	23.68	50.44	4.00	23.48
38	NH_2	4-OCH_3	3[c]	61	240–243	$C_{11}H_{12}N_4O_2$	56.89	5.21	24.12	56.78	5.04	24.33
39	SH		e,f	11	280–282	$C_{10}H_9N_3OS$	54.78	4.14	19.16	54.98	4.37	19.03
40	SH	2-CH_3	2[f]	6	280–283	$C_{11}H_{11}N_3OS$	56.63	4.75	18.01	56.64	5.19	17.62
41	SH	3-CH_3	2[f]	12	246–248	$C_{11}H_{11}N_3OS$	56.63	4.75	18.01	56.82	4.89	17.85
42	SH	4-CH_3	2[f]	41	300–302	$C_{11}H_{11}N_3OS$	56.63	4.75	18.01	56.64	4.40	18.07
43	SH	4-Cl	g		300–301	$C_{10}H_8ClN_3OS$	47.34	3.18	16.56	47.54	3.14	16.22
44	SH	4-OCH_3	2[f]	30	274–276	$C_{11}H_{11}N_3O_2S$	53.00	4.45	16.86	52.85	4.08	16.72

[a] Figures in this column refer to reflux time in hours. [b] With decomposition. Recrystallized in all cases from aqueous 2-methoxyethanol. [c] In each case 6 moles of the aniline/mole of 5-bromoisocytosine [H. L. Wheeler and T. B. Johnson, *Am. Chem. J.*, **29**, 504 (1903)] and sodium acetate (2.5 moles) were used. [d] Synthetic procedure given in detail. [e] From crude ethyl α-(N-formanilido)-β-methoxyacrylate and thiourea. [f] Ten moles of the aniline/mole of 5-bromo-2-thiouracil [H. W. Barrett, I. Goodman, and K. Dittmer *J. Am. Chem. Soc.*, **70**, 1753 (1948)] was used. [g] From purified ethyl α-(N-formyl-*p*-chloroanilido)-β-methoxyacrylate and thiourea. Synthetic procedure given in detail.

5-Anilinoisocytosine (34, Table IX).—A mixture of 20 g. (0.105 mole) of 5-bromoisocytosine,[34] 20 g. (0.245 mole) of sodium acetate, 100 g. (1.07 moles) of aniline, and 100 ml. of ethylene glycol was refluxed at 180–190° for 2 hr. Upon cooling and dilution with water, acetic acid was added to pH 5.5. The mixture was refrigerated overnight, ether was added to dissolve a large fraction of the excess aniline, and the brown solid was collected. The product was washed with ether and acetone, dissolved in alkali, treated with charcoal, and reprecipitated with acetic acid to give 9 g. (43%) of beige solid. The product was again reprecipitated from alkali and repeatedly recrystallized from aqueous 2-methoxyethanol. It melted at 294–295°; λ_{max} 2.1–2.8, 3.15, 5.8–6.8, 6.23, 6.5–7.5 μ.

Anal. Calcd. for $C_{10}H_9N_3O_2$: C, 59.11; H, 4.46; N, 20.68. Found: C, 59.35; H, 5.10; N, 20.49, 20.73.

A solution of 5 g. (0.026 mole) of 5-bromoisocytosine and 20 g. (0.215 mole) of aniline in 85 ml. of ethylene glycol was refluxed for 2 hr. at 190–195°. The product was isolated and purified as before. It melted at 310–315°. The analysis and ultraviolet absorption spectrum indicated that the substance was 5-anilinouracil.

5-(2,4-Dinitroanilino)uracil (VI).—A solution of 20 g. (0.16 mole) of 5-aminouracil and 30 g. (0.16 mole) of dinitrofluorobenzene in 500 ml. of ethylene glycol was stirred at 175–180° (internal) for 1.5 hr. Upon cooling, the reaction mixture solidified. Water was added with stirring, and the product (38 g., 83%) was filtered and washed with water, ethanol, and ether. It was purified by recrystallization from aqueous dimethylformamide. An analytically pure sample was obtained from boiling glacial acetic acid. It melted at 297–298° (yellow needles); ultraviolet absorption characteristics were as follows [mμ (ε × 10^{-3})]: (a) 0.1 *N* HCl, λ_{max} 262 (14.9), 351 (16.3); λ_{min} 250 (14.0), 297.5 (5.8); (b) pH 11 glycine buffer, λ_{max} 227.5 (13.3), 269 (12.0), 362 (16.2); λ_{min} 247 (9.2), 310 (6.5).

Anal. Calcd. for $C_{10}H_7N_3O_8$: C, 40.96; H, 2.40; N, 23.88. Found: C, 40.90; H, 2.92; N, 24.14.

The literature[8] gives m.p. 312–313 dec.; ultraviolet [mμ (ε × 10^{-3})]: at pH 1, λ_{max} 246 (15.8), sh 285 (10.0), 340 (5.8); at pH 11, λ_{max} 286 (12.4) and 340 (10.0). Using the published procedure,[8] a 73% yield of yellow VI was obtained.

5-(4-Carboxy-2-nitro)anilinouracil was prepared by the reaction of 5-aminouracil with 4-fluoro-3-nitrobenzoic acid, as described above for VI. The orange-red product, recrystallized from HOAc, did not decompose at 315°; yield, 21%.

Anal. Calcd. for $C_{11}H_8N_4O_8$: C, 45.21; H, 2.76; N, 19.71. Found: C, 45.19; H, 2.91; N, 18.94.

(34) H. L. Wheeler and T. B. Johnson, *Am. Chem. J.*, **29**, 504 (1903).

2,4-Dichloro-5-anilinopyrimidines (Table X).—A mixture of 1 mole of the 5-anilino- or 5-N-alkylanilinouracil and 2 moles of N,N-diethylaniline in phosphoryl chloride (12 ml./g. of uracil) was refluxed for 2 hr. The dark solution was concentrated at reduced pressure to one-third of the original volume and poured on ice–water. While the temperature was maintained below 10°, the mixture was stirred and neutralized by addition of NH_4OH. Stirring was continued for 2 hr. and NH_4OH was added as necessary to maintain pH 6–7. The mixture was then extracted with ethyl acetate four times, and the combined extracts were extracted with 6 *N* HCl to remove N,N-diethylaniline. The ethyl acetate was then washed with water, until the washings were neutral, and dried. The residue, obtained after filtering and concentrating the filtrate to dryness, was taken up in boiling hexane, filtered, and concentrated to a small volume. On cooling the 2,4-dichloro derivative crystallized in sufficiently pure state for use in subsequent amination. Analytically pure samples were obtained by further recrystallization from hexane. In an equally effective method, but offering no advantages, 3–5 moles of water was first added to phosphoryl chloride. The work-up and yields were the same. Phosphoryl chloride alone was found to require very much longer reflux times and afforded very poor yields, except in the case of 5-(2,4-dinitroanilino)uracil.

5-Anilino-2,4-diaminopyrimidines. General Method.—A mixture of 2,4-dichloro-5-anilino- or 5-N-alkylanilinopyrimidine and ethanol saturated with ammonia at 5° was heated at 140° for 16 hr. in an autoclave. The solvent was removed under diminished pressure. The residue was taken up in dilute acetic acid, treated with charcoal, and reprecipitated with alkali. The product was collected, washed well with water, dried, and recrystallized from benzene containing a little methanol (except as noted in Table XI). The 2,4-diaminopyrimidines are listed in Table XI.

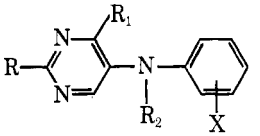
5-Acetanilidouracil.—A mixture of 5 g. (0.0246 mole) of 5-anilinouracil and 20 ml. of acetic anhydride was refluxed for 2 hr., concentrated to half the original volume, and poured onto ice. The lumps were broken up, and the solid product was collected and washed with water and two small portions of ethanol. After drying, there was obtained 5.5 g. (87.5%) melting at 270°. An analytically pure sample was obtained by repeated recrystallization from aqueous ethanol.

5-*p*-Chloroacetanilidouracil was prepared by the same procedure. These compounds are listed in Table III.

Thiation of 5-Anilinoisocytosines and 5-Anilino-2-thiouracils (Table VI). 2-Amino-5-anilino-4-mercaptopyrimidine.—A mixture of 5 g. (0.025 mole) of 5-anilinoisocytosine and 15 g. of crude phosphorus pentasulfide powder in 50 ml. of redistilled pyridine containing 4 drops of water was stirred under reflux for 7 hr. After standing overnight at room temperature, the mixture was

TABLE X

MISCELLANEOUS 2,4-DISUBSTITUTED 5-ANILINOPYRIMIDINES

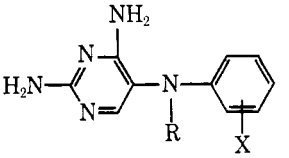


Compd.	R	R_1	R_2	X	Yield, %	M.p., °C. dec.	Formula	Calcd., % C	H	N	Found, % C	H	N
45	NH_2	SH	H		..	223–224	$C_{10}H_{10}N_4S$[a]	55.02	4.62	25.67	55.22	4.43	25.59
46	NH_2	SH	H	3-Cl	..	242–243	$C_{10}H_9ClN_4S$[b]	47.52	3.59	22.17	47.49	3.86	22.01
47	NH_2	SH	H	4-OCH_3	..	241–242	$C_{11}H_{12}N_4OS$[c]	53.21	4.87	22.56	53.55	4.93	22.73
48	SH	SH	H	4-CH_3	..	276–278	$C_{11}H_{11}N_3S_2$	52.98	4.45	16.85	53.02	4.35	16.73
49	SH	SH	H	4-Cl	..	287–289	$C_{10}H_5ClN_3S_2$	44.52	2.99	15.58	44.77	2.87	15.51
50	Cl	Cl	H		34	95.5–96.5	$C_{10}H_7Cl_2N_3$	50.02	2.94	17.50	50.19	2.94	17.38
51	Cl	Cl	H	3-CH_3	21	74–75.5	$C_{11}H_9Cl_2N_3$	51.99	3.57	16.54	51.70	3.41	16.33
52	Cl	Cl	H	3-Br	36	143–145	$C_{10}H_6BrCl_2N_3$	37.65	1.90	13.17	37.77	2.00	13.12
53	Cl	Cl	H	4-OCH_3	20	121–122.5	$C_{11}H_9Cl_2N_3O$	48.91	3.36	15.56	48.76	3.02	15.63
54	Cl	Cl	H	2,4-$(NO_2)_2$	66	168–170	$C_{10}H_5Cl_2N_5O_4$	36.38	1.53	21.22	36.31	1.53	20.95
55	Cl	Cl	C_2H_5		66	51–52	$C_{12}H_{11}Cl_2N_3$	53.75	4.13	15.67	53.44	4.39	15.66
56	Cl	Cl	CH_3	2-CH_3	82	115–117	$C_{12}H_{11}Cl_2N_3$	53.75	4.13	15.67	53.54	4.20	15.47
57	Cl	Cl	CH_3	4-CH_3	88	90–91	$C_{12}H_{11}Cl_2N_3$	53.75	4.13	15.67	54.08	3.87	15.67

[a] *Anal.* Calcd.: S, 14.69. Found: 15.05. [b] *Anal.* Calcd.: S, 12.69. Found: 12.75. [c] *Anal.* Calcd.: S, 12.91. Found: 13.19

TABLE XI

2,4-DIAMINO-5-ANILINOPYRIMIDINES



Compd.	R	X	M.p., °C.[a]	Formula	Calcd., % C	H	N	Found, % C	H	N
58	H	H	201–203[b]	$C_{10}H_{11}N_5$	59.69	5.51	34.80	59.48	5.32	35.10
59	H	3-CH_3	189–190[b]	$C_{11}H_{13}N_5$	61.38	6.09	32.54	61.11	5.94	32.27
60	H	4-CH_3	231–232[b]	$C_{11}H_{13}N_5$	61.38	6.09	32.54	61.53	6.36	32.28
61	H	4-Cl	263–265[b]	$C_{10}H_{10}ClN_5$	50.96	4.28	29.72	50.70	4.23	29.37
62	H	3,4-$(Cl)_2$	260–261[b]	$C_{10}H_9Cl_2N_5$	44.46	3.36	25.93	44.93	3.32	25.90
63	H	3-Br	226–227[b]	$C_{10}H_{10}BrN_5$	42.87	3.60	25.00	43.11	3.91	25.15
64	H	3-OCH_3	202–203[b]	$C_{11}H_{13}N_5O$	57.13	5.66	30.28	57.01	5.53	30.32
65	H	4-OCH_3	216–219[b]	$C_{11}H_{13}N_5O$	57.13	5.66	30.28	57.00	5.35	30.05
66	H	2,4-$(OCH_3)_2$	212–213[b]	$C_{12}H_{15}N_5O_2$	55.16	5.79	26.81	54.98	5.64	26.88
67	H	2,4-$(NO_2)_2$	257–258[b,c]	$C_{10}H_9N_7O_4$	41.24	3.11	33.67	41.41	3.44	33.40
68	CH_3		179–181	$C_{11}H_{13}N_5$	61.38	6.09	32.54	61.13	5.90	31.92
69	C_2H_5		155–156	$C_{12}H_{15}N_5$	62.86	6.59	30.55	63.09	6.39	30.64
70	CH_3	2-CH_3	212–214	$C_{12}H_{15}N_5$	62.86	6.59	30.55	63.06	6.52	30.59
71	CH_3	4-CH_3	143.5–144.5[d]	$C_{12}H_{15}N_5$	62.86	6.59	30.55	62.71	6.23	30.92
72	CH_3	4-Cl	196–198[c]	$C_{11}H_{12}ClN_5$	52.91	4.85	28.05	53.39	4.53	28.85

[a] Recrystallized from benzene (methanol), except as noted. [b] With decomposition. [c] Recrystallized from aqueous 2-methoxyethanol. [d] Recrystallized from benzene.

poured into water, and the bright yellow solid was collected and washed with water. The crude product was purified by solution in dilute alkali, treatment with charcoal, and reprecipitation by the addition of acetic acid. Repeated recrystallization from aqueous ethanol gave a pure product.

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