mp 69–70°; on other samples, mps between 69° and 87° were obsd). *Anal.* ($C_{13}H_{15}NO_3$) C, H, N.

Ethyl 4-Anilino-1-phenyl-3-pyrroline-3-carboxylates (7). A soln of 2.33 g (0.01 mole) of **6**, 0.012–0.015 mole of an aniline, and 50 ml of C_6H_6 was allowed to stand at room temp for 4 days, washed with 1 *N* HCl, dried ($MgSO_4$), and concd to a solid which was recrystd (see Table I).

Ethyl 4-Anilino-1-phenyl-3-pyrrolecarboxylates (8). A mixt of 1.0 g of **7** and 1.0 g of S was heated at 140° for 1 hr, cooled, dild with 8 ml of $CHCl_3$, filtered, and concd. The residue was chromatogd (three Analtech, Inc. Uniplate silica gel GF plates, 1000 μ thickness) with C_6H_6. The product bands were washed with $CHCl_3$, the soln was concd, and the residue was recrystd (see Table II).

4-Anilino-1-phenyl-3-pyrrolecarboxylic Acids (9). A mixt of 2.5 g of **8**, 60 ml of EtOH, and 60 ml of 1 *N* NaOH was heated under reflux for 1 hr, distd until 75 ml remained, dild with H_2O, filtered, and acidified with HOAc. The solid which sepd was collected and recrystd (see Table III).

References

(1) W. C. Cutting, "Handbook of Pharmacology," 4th ed, Appleton-Century-Crofts, New York, N. Y., 1969, p 624.
(2) J. R. Boissier, R. Gentaz, J. Fichelle, and M. C. Piarroux, *Therapie*, **22**, 1257 (1967).
(3) A. S. Watnick, R. I. Taber, and I. A. Tobachnick, *Fed. Proc., Fed. Amer. Soc. Exp. Biol.*, **27**, 533 (1968).
(4) R. Z. Gussin, J. R. Cummings, E. H. Stokey, and M. A. Ronsberg, *J. Pharmacol. Exp. Ther.*, **167**, 194 (1969).
(5) H. G. Alpermann, *Arzneim.-Forsch.*, **20**, 293 (1970).
(6) R. C. Elderfield, W. J. Gensler, T. H. Bembry, C. B. Kremer, F. Brody, H. A. Hageman, and J. D. Head, *J. Amer. Chem. Soc.*, **68**, 1259 (1946).
(7) A. T. de Moulipied, *J. Chem. Soc.*, **87**, 435 (1905).
(8) C. A. Winter, E. A. Risely, and G. W. Nuss, *Proc. Soc. Exp. Biol. Med.*, 111, 544 (1962).
(9) B. B. Newbould, *Brit. J. Pharmacol.*, **21**, 127 (1963).
(10) L. C. Hendershot and J. Forsaith, *J. Pharmacol. Exp. Ther.*, **125**, 237 (1958).

Some Derivatives of 9-Amino-9*H*-purine-6(1*H*)-thione†

Carroll Temple, Jr.,* Conrad L. Kussner, and John A. Montgomery

Kettering-Meyer Laboratory, Southern Research Institute, Birmingham, Alabama 35205.

Received November 16, 1971

The anticancer activity of purine-6(1*H*)-thione, its 9-alkyl derivatives and compounds prepared from these thiones prompted the preparation and testing of some derivatives of 9-aminopurine-6(1*H*)-thione (**4**).[1]

The preparation and acidic hydrolysis of **3** to give **4** and the alkylation of the latter to give **1** and **2** has been reported.[2] Condensation of **1**, **2**, and **4** with C_6H_5CHO gave, respectively, **10**, **11**, and **14**. The reaction of **1** and **4** with 2,5-dimethoxytetrahydrofuran[3] gave the 9-pyrrol-1-yl-9*H*-purines **12** and **15**. Similarly condensation of **1** with 2,5-hexanedione gave **13**. Alkylation of the thione group of **3** with the appropriate alkyl halide gave **5**–**7**. A second alkylation of the acetamido group of **5** and **7** with $Br(CH_2)_4Cl$ gave **8** and **9**, respectively. Cyclization of **8** and **9** was effected with base to give the 9-pyrrolidin-1-yl-9*H*-purines **17** and **18**. Treatment of the latter with CF_3CO_2H removed the diphenylmethyl blocking group to give **16**.[4]

Compounds were tested against L1210 leukemic cells implanted ip in mice on single dose and chronic schedules.[5,6] The test results summarized in Table I indicate that the 9-aminopurines **2** and **4** and the 9-acetamidopurine **3** have activity and are less toxic than 6-mercaptopurine (6-MP). On the chronic schedule the activity of **2** is similar to that of 6-MP, whereas, the activities of **3** and **4** are lower. In con-

†This work was supported by funds from the C. F. Kettering Foundation, and Chemotherapy, National Cancer Institute, National Institutes of Health, Contract No. NIH-71-2021.

Scheme I

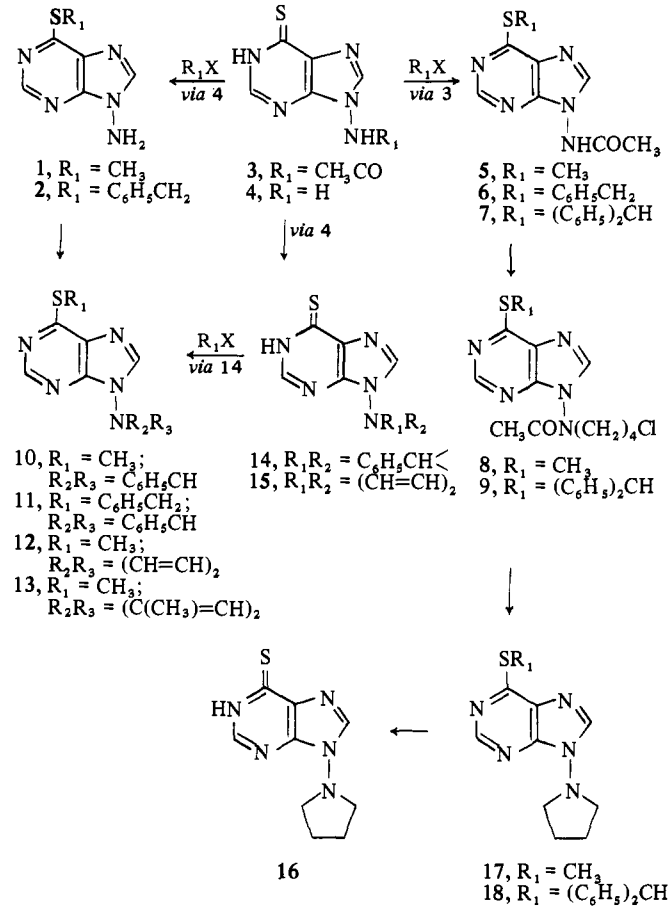

Table I. Activity of 9-Amino-9*H*-purine-6(1*H*)-thione and Derivatives against L1210 Leukemia Implanted Intraperitoneally

Compd	Dose, mg/kg per day	Schedule (ip)	ip L1210 (10^5 cells) Life span, days: Treated	Control	% ILS
6-MP[b]	380	day 2			39
	62	qd 2–16			60
2	62	qd 1–15	10.7	9.3	15
	93	qd 1–15	11.3	9.3	21
	140	qd 1–15	13.8	9.0	53[a]
	210	qd 1–15	12.7	9.3	36
3	177	day 1	9.3	8.7	6
	200	day 1	10.2	9.2	10
	266	day 1	13.4	8.7	54
	400	day 1	11.1	9.0	23[a]
	600	day 1		Toxic	
	100	qd 1–9	12.5	9.2	35
	200	qd 1–9	8.6	9.2	0
	400	qd 1–9		Toxic, chronic	
4	200	day 2	9.8	9.6	2
	266	day 2	9.8	8.5	15
	400	day 2	14.3	9.1	57[a]
	600	day 2		Toxic	
	72	qd 1–15	8.8	8.7	1
	120	qd 1–15	12.5	8.7	43
	200	qd 1–15	7.5	8.7	0
6	200	day 2	8.8	8.8	0
	266	day 2	10.6	9.2	15
	400	day 2	12.0	9.0	33
	600	day 2		Toxic, chronic	

[a] Average of 2 or 3 tests. [b] See ref 6.

trast to 2 and 6-MP both 3 and 4 showed higher activity on the single dose treatment. Compared to 3, the acetamido compounds 5-9 were less active on both dose schedules. In fact, no activity was observed for the N,N-disubstituted amides 8 and 9. Similarly, little or no activity was observed for the 9-benzylideneamino- (10, 11, and 14), 9-pyrrol-1-yl- (12, 13, and 15), and 9-pyrrolidin-1-yl- (16-18) 9*H*-purines. These results indicate that a proton on the 9-amino group is necessary for activity. Apparently these protons are involved either in enzymatic binding or removal of the 9-amino group to give the corresponding purine derivative.

Experimental Section‡

N-[6-(Methylthio)-9*H*-purin-9-yl]acetamide (5). A soln of 3 (6.0 g)[2] in DMF (120 ml) contg K_2CO_3 (4.1 g) and MeI (2.0 ml) was stirred at room temp for ~20 hr, dild with H_2O (600 ml), acidfd with dil HCl, and evapd to dryness *in vacuo*. The residue was washed with H_2O (90 ml) and recrystd from C_6H_6, and the resulting solid was dried (P_2O_5, 78°) *in vacuo*: yield, 3.4 g (53%); mp 185°. *Anal.* ($C_8H_9N_5OS$) C, H, N.

N-[6-(Benzylthio)-9*H*-purin-9-yl]acetamide (6) was prepd by a similar method from 3 (2.5 g),[2] K_2CO_3 (1.7 g), and $C_6H_5CH_2Cl$ (1.4 ml) in DMF (50 ml): yield, 2.3 g (C_6H_6, 64%); mp 200-201°. *Anal.* ($C_{14}H_{13}N_5OS$) C, H, N.

N-[6-[(Diphenylmethyl)thio]-9*H*-purin-9-yl]acetamide (7) was prepd from 3 (2.0 g),[2] K_2CO_3 (1.4 g), and $(C_6H_5)_2CHCl$ (2.0 ml) in DMF (40 ml): yield, 2.2 g (C_6H_6-hexane; 61%); mp 214-215°. *Anal.* ($C_{20}H_{17}N_5OS$) C, H, N.

N-(4-Chlorobutyl)-*N*-[6-(methylthio)-9*H*-purin-9-yl]acetamide (8). A soln of 5 (7.0 g) in DMF (140 ml) contg K_2CO_3 (4.4 g) and 1-bromo-4-chlorobutane (3.8 ml) was stirred at room temp for 18 hr, then dild with H_2O (700 ml). The resulting oily suspension was extd with Et_2O (3 × 1400-ml portions), and the combined ext was dried ($MgSO_4$) and evapd to dryness to give the product as an oil: yield, 9.8 g (99%). Elemental analyses were obtained on a sample dried at 56° *in vacuo* over P_2O_5. *Anal.* ($C_{12}H_{16}ClN_5OS$) C, H, N.

N-(4-Chlorobutyl)-*N*-[6-(diphenylmethyl)thio]-9*H*-purin-9-yl]-acetamide (9) was similarly prepd from 7 (12.0 g). The resulting oil was dried at 78° *in vacuo* over P_2O_5 to give a glass: yield, 14.5 g (97%). *Anal.* ($C_{24}H_{24}ClN_5OS$) C, H, N.

9-Benzylideneamino-6-(methylthio)-9*H*-purine (10) was prepd from 14 (5.0 g), K_2CO_3 (2.8 g) and MeI (1.3 ml) in DMF (100 ml): yield, 4.0 g (hexane, 76%): mp 200-201°. *Anal.* ($C_{13}H_{11}N_5S$) C, H, N.

9-Benzylideneamino-6-(benzylthio)-9*H*-purine (11) was prepd from 14 (3.0 g), K_2CO_3 (1.7 g), and $C_6H_5CH_2Cl$ (1.4 ml) in DMF (60 ml): yield, 3.1 g (C_6H_6, 76%); mp 208°. *Anal.* ($C_{19}H_{15}N_5S$) C, H, N.

6-(Methylthio)-9-pyrrol-1-yl-9*H*-purine (12). A mixt of 1 (1.0 g) and 2,5-dimethoxytetrahydrofuran (0.73 ml)[3] in glacial HOAc (10 ml) was refluxed for 2.5 hr and evapd to dryness *in vacuo*. The resulting residue was recrystd from hexane and dried at 78° *in vacuo* over P_2O_5: yield, 0.84 g (66%); mp 163°. *Anal.* ($C_{10}H_9N_5S$) C, H, N.

9-(2,5-Dimethylpyrrol-1-yl)-6-(methylthio)-9*H*-purine (13) was similarly prepd from 1 (1.0 g) and 2,5-hexanedione (0.66 ml) in glacial HOAc: yield, 1.1 g (77%). *Anal.* ($C_{12}H_{13}N_5S$) C, H, N.

9-Benzylideneamino-9*H*-purine-6(1*H*)-thione (14). A suspension of 4 (10.0 g)[2] and PhCHO (10 ml) in MeOH (500 ml) contg 5 drops of concd HCl was refluxed with stirring for 2 hr. The mixt was cooled, and the solid was collected by filtration and dried *in vacuo* over P_2O_5: yield, 14.5 g (95%); mp >264°. *Anal.* ($C_{12}H_9N_5S$) C, H, N.

9-(Pyrrol-1-yl)-9*H*-purine-6(1*H*)-thione (15) was prepd similarly to that of 12 from 4 (1.0 g) and 2,5-dimethoxytetrahydrofuran (0.78 ml)[3] in glacial HOAc (15 ml). The crude product was recrystd from H_2O and dried at 110° *in vacuo* over P_2O_5: yield, 0.71 g (55%); mp >264°. *Anal.* ($C_9H_7N_5S$) C, H, N.

9-Pyrrolidin-1-yl-9*H*-purine-6(1*H*)-thione Monohydrate (16). A soln of 18 (2.2 g) and PhOH (2.2 g) in CF_3CO_2H (22 ml) was refluxed with stirring for 30 min and evapd to dryness *in vacuo*. The residue was dissolved in dil NaOH and the soln was neutralized with dil HCl. A second repptn of the solid that deposited from a NaOH soln by addn of glacial HOAc gave pure 16: yield, 0.90 g; mp >264°. *Anal.* ($C_9H_{11}N_5S \cdot H_2O$) C, H, N.

6-(Methylthio)-9-pyrrolidin-1-yl-9*H*-purine (17). A soln of 8 (2.7 g) in dioxane (100 ml) contg pyridine (3 ml) was refluxed for 44 hr and evapd to dryness *in vacuo*. The residue was washed with H_2O and recrystd from EtOH: yield, 1.5 g (64%); mp 143°. *Anal.* ($C_{10}H_{13}N_5S$) C, H, N.

6-[(Diphenylmethyl)thio]-9-pyrrolidin-1-yl-9*H*-purine (18). A soln of 9 (10.6 g) in dioxane (220 ml) contg 1 *N* NaOH (55 ml) was heated at 55-60° for 20 hr and evapd to dryness *in vacuo*. The residue was extd with $CHCl_3$ (500 ml) and the solid obtd from evapn of the ext was recrystd from EtOH: yield, 3.80 g (39%); mp 151°. *Anal.* ($C_{22}H_{21}N_5S$) C, H, N.

Acknowledgment. The authors are indebted to Dr. W. C. Coburn, Jr., and members of the Molecular Spectroscopy Section of Southern Research Institute for the microanalytical results reported and to Dr. W. R. Laster and members of the Cancer Screening Division for the screening data reported.

References

(1) C. Temple, Jr., C. L. Kussner, and J. A. Montgomery, *J. Med. Chem.*, 11, 41 (1968).
(2) C. Temple, Jr., A. G. Laseter, and J. A. Montgomery, *J. Heterocycl. Chem.*, 5, 711 (1968).
(3) N. Elming and N. Clauson-Kaas, *Acta Chem. Scand.*, 6, 867 (1952).
(4) F. I. Carroll and A. Philip, *J. Org. Chem.*, 33, 3776 (1968).
(5) H. E. Skipper, J. A. Montgomery, J. R. Thomson, and F. M. Schabel, Jr., *Cancer Res.*, 19, 425 (1959).
(6) H. E. Skipper, F. M. Schabel, Jr., L. B. Mellett, J. A. Montgomery, L. J. Wilkoff, H. H. Lloyd, and R. W. Brockman, *Cancer Chemother. Rep.*, 54, 431 (1970).

‡Melting points were detd on a Kofler Heizbank apparatus. Where analyses are indicated only by symbols of the elements, analytical results obtained for those elements were within ±0.4% of the theoretical values.

Pyrido[2,3-*d*]pyrimidine-6-carboxamides as Potential Diuretic Agents

Arthur A. Santilli* and Dong Han Kim

Wyeth Laboratories, Inc., Research and Development Division, Radnor, Pennsylvania 19087. *Received October 20, 1971*

Previously we reported 4,7-diamino-*N*-(2-morpholinoethyl)-2-phenyl-6-pteridinecarboxamide (3)[1a] and related amides[1b] to have significant diuretic activity in rats. Further interest in the structural requirements for activity led us to prepare the 5-deaza isostere 4,7-diamino-*N*-(2-morpholinoethyl)-2-phenylpyrido[2,3-*d*]pyrimidine-6-carboxamide (6a) and related amides (6b-d). (See Scheme I and Table I.) The previously undescribed synthetic route to these compounds parallels the one used for preparing 3 except that the 4,6-diamino-5-pyrimidinecarboxaldehydes (5a-b) were used instead of 4,6-diamino-5-nitrosopyrimidines such as 1. Treatment of 4,6-dichloro-2-phenyl-5-pyrimidinecarboxaldehyde (4a)[2] with NH_4OH afforded 4,6-diamino-2-phenyl-5-pyrimidinecarboxaldehyde (5a). This intermediate, when allowed to react with 2-cyano-*N*-(2-morpholinoethyl)-acetamide (2) in refluxing EtOH containing an equivalent of NaOEt, afforded 6a. Similarly, the pyrido[2,3-*d*]pyrimidine-6-carboxamides 6b-d were prepared from 5a-b and the corresponding 2-cyano-*N*-(substituted)acetamides. When 5b was treated with *N,N'*-bis(2-methoxethyl)malonamide under the same conditions, 4-amino-7-hydroxy-*N*-(2-methoxyethyl)-2-phenylpyrido[2,3-*d*]pyrimidine-6-carboxamide (7) was formed.

Interestingly, neither 6a nor the other pyrido[2,3-*d*]-pyrimidines described in this report were active in the standard rat diuretic screen[3] used in our laboratories. Replacement of N at the 5 position with CH in compounds such as 3, therefore, must offer sufficient steric and/or