ibid., **34**, 981 (1974). Two-step fragmentation of the 4-hydroxy derivative of **2** into phosphoramide mustard [H_2N-(HO)P(O)N($CH_2CH_2Cl)_2$] and *o*-hydroxybenzaldehyde is conceivable. The importance of phosphoramide mustard production in the metabolism of 1 has been recently discussed: P. J. Cox, P. B. Farmer, and M. Jarman, *Biochem. Pharmacol.*, **24**, 599 (1975).
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Synthesis and Antimicrobial Activity of Certain Imidazo[1,2-*a*]pyrimidines

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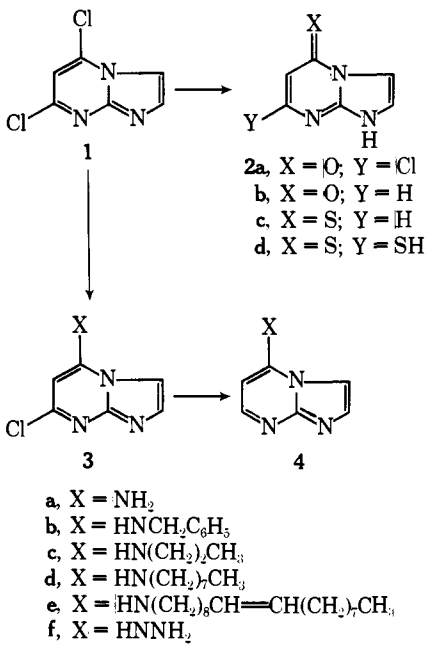
A series of 5-substituted and 5,7-disubstituted imidazo[1,2-*a*]pyrimidines has been prepared. The in vitro antimicrobial activity of these compounds against a variety of microorganisms is reported. 5-*n*-Octylaminoimidazo[1,2-*a*]pyrimidine exhibited significant activity against all the microorganisms studied.

In continuation of our investigations on the purine antagonists, we recently described [1-5]the synthesis and antiviral activity of nucleosides resembling inosine and guanosine of various heterocyclic systems containing a bridgehead nitrogen atom. We now wish to report the synthesis and antimicrobial activity of some amino-substituted imidazo[1,2-*a*]pyrimidines.

Scheme I



Chemistry. The compounds described herein (Scheme I) were prepared by treating the reactive 5,7-dichloroimidazo[1,2-*a*]pyrimidine (1) with the appropriate amine. The synthesis of 1 has been achieved as described earlier[4] by the successful chlorination of 5,7-dihydroxyimidazo[1,2-*a*]pyrimidine.[6] Treatment of 1 with 5% aqueous sodium hydroxide solution at reflux gave 7-chloroimidazo[1,2-*a*]pyrimidin-5-one[4] (**2a**) which on subsequent catalytic dehalogenation with 10% palladium on carbon in a hydrogen atmosphere at room temperature furnished imidazo[1,2-*a*]pyrimidin-5-one (**2b**). Chlorination of **2b** with phosphorus oxychloride in the presence of *N,N*-dimethylaniline gave the intermediate 5-chloroimidazo[1,2-*a*]pyrimidine, which without isolation was treated with thiourea in ethanol to obtain the 6-mercaptopurine analog, imidazo[1,2-*a*]pyrimidine-5-thione (**2c**). There was observed an absorption band in the ir spectrum (KBr) at 1625 cm^{-1} which was assigned to C=S stretching[7] indicating that **2c** exists in the thione rather than the thiol form. Reaction of 1 with 2 molar equiv of thiourea produced the dimercapto derivative **2d**.

The difference in reactivity between the two chlorine atoms of 1 was made use to prepare a number of 5-substituted imidazo[1,2-*a*]pyrimidines. Treatment of 1 with concentrated ammonium hydroxide at room temperature gave 5-amino-7-chloroimidazo[1,2-*a*]pyrimidine (**3a**) which on catalytic dehalogenation converted to the adenine analog, 5-aminoimidazo[1,2-*a*]pyrimidine (**4a**). Similarly, treatment of 1 with benzylamine, propylamine, *n*-octylamine, and oleylamine gave the corresponding 5-substituted 7-chloroimidazo[1,2-*a*]pyrimidines (**3b–e**) which on subsequent catalytic dehalogenation with 10% palladium on carbon in a hydrogen atmosphere furnished 5-benzylamino- (**4b**), 5-propylamino- (**4c**), 5-*n*-octylamino- (**4d**), and 5-oleylamino- (**4e**) imidazo[1,2-*a*]pyrimidines, respectively. Displacement of chlorine adjacent to the bridgehead nitrogen of 1 by hydrazine at room temperature gave almost quantitative yield of 7-chloro-5-hydrazinoimidazo[1,2-*a*]pyrimidine (**3f**). Physical constants of the 16 compounds screened for antimicrobial activity are listed in Table I.

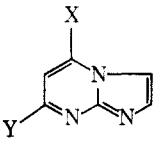
Of the 16 compounds screened in vitro, only 5-*n*-octylaminoimidazo[1,2-*a*]pyrimidine (**4d**) exhibited significant activity against all the microorganisms[8] tested [MIC (μmol/ml) *Ca*, 0.08; *Ef*, 0.08; *Mc*, 0.04; *Sa*, 0.16; *Tm*, 0.08] and is presently under further evaluation. All other compounds tested were not active.

In conclusion, a series of purine analogs in the imidazo[1,2-*a*]pyrimidine ring system has been synthesized by an unambiguous method. These compounds resemble 1-deazapurines with a bridgehead nitrogen atom in which C-5 and N-7 are interchanged. Of all the compounds examined, only **4d** exhibited significant in vitro antimicrobial activity.

Experimental Section

Melting points were taken on a Thomas-Hoover capillary melting point apparatus and are uncorrected. Nuclear magnetic resonance (^1H NMR) spectra were recorded at 60 MHz on a Hitachi Perkin-Elmer R-20A spectrometer in Me_2SO-d_6 using DSS as an internal standard and infrared (ir) spectra on a Perkin-Elmer 257 spectrophotometer (KBr pellets). ICN-Woelm silica gel (70–230

Table I. Physical Data of Some Imidazo[1,2-*a*]pyrimidines



Compd	X	Y	Mp, °C	% yield	Formula[a]	Uv, λ_{max} (pH 7), nm ($\epsilon \times 10^3$)[b]
1	Cl	Cl	255–256	72.3	$C_6H_3Cl_2N_3$	280 (4.0) 317 (4.3)
2a	OH	Cl	266–267 dec	61.0	$C_6H_4ClN_3O$	294 (10.7)
2b	OH	H	222	45.2	$C_6H_5N_3O$	247 (3.7) 296 (11.4)
2c	SH	H	285–287 dec	68.4	$C_6H_5N_3S$	249 (11.9) 345 (17.7)
2d	SH	SH	>220 dec	85.5	$C_6H_6N_3S_2$	257 (14.5) 343 (13.0)
3a	NH_2	Cl	>320	85.5	$C_6H_5ClN_4$	305 (8.4)
3b	$HNCH_2C_6H_5$	Cl	>320	85.0	$C_{13}H_{11}ClN_4$	260[c] (2.2) 310 (10.4)
3c	$HN(CH_2)_2CH_3$	Cl	210 softens >300	92.6	$C_9H_{11}ClN_4$	260[c] (2.2) 310 (11.6)
3d	$HN(CH_2)_7CH_3$	Cl	170	85.4	$C_{14}H_{21}ClN_4$	235 (14.9) 310 (5.8)
3e	$HN(CH_2)_8CH{=}CH(CH_2)_7CH_3$	Cl	135–136	87.5	$C_{24}H_{39}ClN_4$	243 (24.2) 323 (13.6)
3f	$HNNH_2$	Cl	>300	97	$C_6H_6ClN_5$	262[c] (2.8) 308 (10.7)
4a	NH_2	H	260 dec	53.3	$C_6H_6N_4 \cdot H_2O$	305 (10.0)
4b	$HNCH_2C_6H_5$	H	220–221	71.5	$C_{13}H_{12}N_4$	260[c] (2.0) 312 (13.8)
4c	$HN(CH_2)_2CH_3$	H	148 softens 180	83.6	$C_9H_{12}N_4$	306[c] (1.9) 311 (13.4)
4d	$HN(CH_2)_7CH_3$	H	101–102	68.4	$C_{14}H_{22}N_4$	227 (26.7) 312 (13.3)
4e	$HN(CH_2)_8CH{=}CH(CH_2)_7CH_3$	H	98	71.5	$C_{24}H_{40}N_4$	239 (25.5) 323 (14.1)

[a] All compounds were analyzed for C, H, and N and the results were within ±0.3% of theoretical values. [b] Ultraviolet spectra were obtained on a Cary Model 15 spectrophotometer. [c] Shoulder.

mesh) was used for column chromatography. Elemental analyses were performed by MHW Laboratories, Garden City, Mich.

7-Chloroimidazo[1,2-*a*]pyrimidin-5-one (2a). A solution of 5,7-dichloroimidazo[1,2-*a*]pyrimidine[4] (1, 2.0 g, 0.0106 mol) in 5% aqueous sodium hydroxide (20 ml) was heated on a steam bath for 45 min, before it was evaporated in vacuo to dryness. The residue was washed with cold ethanol, followed by ether, and dried. The sodium salt thus obtained was dissolved in minimum volume of water and decolorized and cooled filtrate acidified with 6 *N* hydrochloric acid to pH 4. The product was collected, washed with cold water, and crystallized from aqueous ethanol as needles: ir λ_{max} 1670 (C=O), 3100 cm^{-1}.

Imidazo[1,2-*a*]pyrimidin-5-one (2b). To a solution of **2a** (0.50 g, 0.0029 mol) in 50% aqueous ethanol (25 ml) containing 1 ml of concentrated ammonium hydroxide was added palladium on carbon (150 mg, 10%) and the mixture was hydrogenated at 45 psi at room temperature for 5 hr after which the catalyst was removed by filtration on a Celite pad and washed with hot ethanol (3 × 10 ml). The combined filtrate and washings were evaporated to dryness and the residue was crystallized from aqueous ethanol as needles: ir λ_{max} 1670 (C=O), 3100 cm^{-1}.

Imidazo[1,2-*a*]pyrimidine-5-thione (2c). A suspension of dry **2b** (0.50 g, 0.0037 mol) in phosphorus oxychloride (10 ml) containing dimethylaniline (0.5 ml) was heated under reflux for 2 hr under anhydrous conditions. The clear solution was concentrated in vacuo and the residual syrup poured over crushed ice (~25 g). The aqueous solution was neutralized before it was evaporated to dryness. Coevaporation with absolute ethanol several times gave dry residue which was suspended in ethanol (25 ml) and treated with thiourea (250 mg). The suspension was heated under reflux for 1.5 hr and filtered and the filtrate was evaporated to dryness. The residue was chromatographed over a silica gel column (2 × 30 cm) using ethyl acetate–water–1-propanol (4:2:1, upper phase) as the eluent. The appropriate fraction was evaporated and the residue crystallized from aqueous ethanol: ir λ_{max} 1625 cm^{-1} (C=S).

Imidazo[1,2-*a*]pyrimidine-5,7-dithione (2d). A solution of 1 (1.88 g, 0.01 mol) in ethanol (50 ml) was treated with thiourea (1.52 g, 0.02 mol) and the mixture was heated under reflux for 1 hr. After cooling the product precipitated out which was collected and crystallized from aqueous ethanol: ir λ_{max} 1640 cm^{-1} (C=S).

General Synthesis of 7-Chloro-5-substituted Imidazo[1,2-*a*]pyrimidines (3a–f). A solution of 1 (1.88 g, 0.01 mol) in ethanol (25 ml) was treated with the appropriate amine (0.02 mol) and the mixture was heated under reflux for 2–15 hr. The solvent was removed in vacuo and the residue triturated with water (20 ml). After filtration, the residue was crystallized from ethanol.

General Synthesis of 5-Substituted Imidazo[1,2-*a*]pyrimidines (4a–e). A solution of **3a–e** (1.0 g) in 50% aqueous ethanol (50 ml) containing 1 ml of concentrated ammonium hydroxide was treated with palladium on carbon (150 mg, 10%) and the mixture hydrogenated at 45 psi at room temperature for 5–15 hr after which the catalyst was removed by filtration on a Celite pad and washed with hot ethanol (3 × 20 ml). The combined filtrate and washings were evaporated to dryness and the residue was chromatographed over a silica gel column (100 g of silica gel/1 g of compound) using ethyl acetate–water–1-propanol (4:2:1, upper phase) as the eluent. The appropriate fraction was evaporated and the residue crystallized from ethanol.

Antimicrobial Activity. The compounds synthesized for this study were assayed for the antimicrobial activity. Clinical isolates of *Candida albicans* (*Ca*), *Escherichia coli* (*Ec*), *Epidermophyton floccosum* (*Ef*), *Microsporum canis* (*Mc*), *Pseudomonas aeruginosa* (*Pa*), *Staphylococcus aureus* (*Sa*), and *Trichophyton mentagrophytes* (*Tm*) were used for this study. In vitro sensitivity of these organisms to this series of imidazo[1,2-*a*]pyrimidines was quantitatively determined by broth dilution assay.[9]

Serial dilutions were prepared in chemically defined medium in a range from 0.4 to 0.005 μmol/ml. The minimal inhibitory concentration (MIC) was recorded as the highest dilution of compound which prevented visible growth of the pathogen. Bacterial and yeast MIC's were read following 24 hr of incubation at 35°. Dermatophyte inhibition was read after 48 hr of incubation at 30°.

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Synthesis of Some Glycoside Analogs and Related Compounds from 9-Amino-6-(methylthio)-9*H*-purine

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Additional information on the anticancer activity of 9-amino-9*H*-purine-6(1*H*)-thione and its derivatives was sought by the synthesis of some 9-(substituted amino)-6-(methylthio)-9*H*-purines in which the 9-substituent contained functional groups capable of either reversible or irreversible binding with an enzymatic site. Condensation of 9-amino-6-(methylthio)-9*H*-purine (**1**) with some carbonyl compounds followed by hydride reduction of the azomethine linkage in the intermediates leads to the 2-pyrrolylmethyl (**8**), 2,3,4-trihydroxybutyl (**10**), and the 1,5-dihydroxy-2- and -3-pentyl (**11** and **12**) compounds. A 4-hydroxybutyl derivative (**13**) was obtained by alkylation of **18**, the 9-acetyl derivative of **1**, with 4-chlorobutyl acetate followed by saponification. The cyclization of **13** and **11** with a sulfonyl chloride gave the 9-pyrrolidin-1-yl (**27**) and the 9-[2-(tosyloxymethyl)pyrrolidin-1-yl] (**28**), respectively. Acylation of **1** with ethyl L-2-pyrrolidine-5-carboxylate and ethyl 1-methyl-5-pyrrolidone-3-carboxylate, respectively, in Me_2SO containing NaH gave the corresponding amides **15** and **17**. Alkylation of **18** with 1-bromo-2-chloroethane and epichlorohydrin gave the *N*-(2-chloroethyl) and *N*-(1,2-epoxy-3-propyl) derivatives **19** and **20**. The chloro group of the chlorobutyl derivative of **18** was displaced with KSCN and NaN_3, respectively, to give the thiocyanate and azido derivatives **23** and **24**. Hydrogenation of the latter gave the amine (**25**), which was acylated with ethyl chloroformate to give the (ethoxycarbonyl)amino compound **26**. None of these compounds showed activity against L1210 leukemia cells implanted ip in mice on a single-dose schedule, suggesting that the activity observed in the simpler 9-aminopurines resulted from cleavage of the hydrazino linkage to give 9*H*-purine-6(1*H*)-thione.

Previously, we reported that 9-amino-9*H*-purine-6(1*H*)-thione and some of its derivatives showed anticancer activity against leukemia L1210 in mice.[1] Although these compounds or their anabolic products might possess intrinsic activity, it is possible that they are degraded to the active agent, 9*H*-purine-6(1*H*)-thione.[2] To provide additional information on this type of compound, one series of 9-(substituted amino)-6-(methylthio)-9*H*-purines was prepared in which the 9-substituent was either a pyrrolylmethyl or a (poly)hydroxyalkyl group, which might lead to tighter binding with an enzymatic site. In another series the 9-substituent contained a reactive center, which might bind irreversibly with an enzymatic site.

The condensation of **1** and pyrrole-2-carboxaldehyde gave **2**, which was treated with $NaBH_4$ to reduce the -N=CH- linkage to give **8**. Similarly, **9** was prepared by the condensation of **1** with 2,4-*O*-ethylidene-D-erythrose[3] to give **3** followed by reduction of this product with $NaBH_4$. Hydrolysis of the ethylidene blocking group of **9** with hot 60% aqueous HOAc gave the 2,3,4-trihydroxybutyl compound **10**. The dihydroxypentyl derivatives **11** and **12** were prepared by the condensation of **1** with diethyl 2-oxoglutarate (**4**)[4] and 3-oxoglutarate (**6**),[5] respectively, to give **5** and **7** followed by reduction of both the -N=C= linkage and the ester moieties of these intermediates with $NaBH_4$. Of interest is the observation that **5** is cleaved in Me_2SO-d_6, but not $CDCl_3$, to 6-(methylthio)-9*H*-purine (^1H NMR). Further, reduction of the azomethine linkage of **5** gave a compound (**11**) that was stable in Me_2SO-d_6 (^1H NMR). The 4-hydroxybutyl compound **13** was prepared by saponification of **21** (see below).

The cyclization of **13** to the known 9-pyrrolidin-1-yl derivative **27** was effected in dioxane with methanesulfonyl chloride in the presence of Et_3N.[1] The isolation of the mesyloxy intermediate in this reaction was unsuccessful. Similarly, treatment of **11** in pyridine with excess *p*-toluenesulfonyl chloride gave the 9-[2-(tosyloxymethyl)pyrrolidin-1-yl] compound, **28** (Scheme I).

For the preparation of some acyl derivatives, the mild conditions of Singh were used.[6] Reaction of **1** with ethyl L-2-pyrrolidone-5-carboxylate (**14**)[7] and ethyl 1-methyl-5-pyrrolidone-3-carboxylate (**16**), respectively, in Me_2SO containing NaH gave **15** and **17**.

The synthesis of the compounds containing a reactive center was effected by alkylation of the known 9-acetamido compound **18**.[1] Treatment of the latter with 1-bromo-2-chloroethane, epichlorohydrin, and 4-chlorobutyl acetate, respectively, in a dipolar aprotic solvent in the presence of