The synthesis of some 9-furfuryl-6-substituted purines as potential antimetabolites[1,2]

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Several 9-furfuryl-6-substituted purines were synthesized by reacting 9-furfuryl-6-chloropurine (**5**) with the corresponding nucleophilic reagent. The mother compound (**5**) was prepared by direct cyclization of 4-furfurylamino-5-amino-6-chloropyrimidine (**4**) with a mixture of ethyl orthoformate and acetic anhydride. The diaminopyrimidine (**5**) was prepared by treatment of 4,6-dichloro-5-aminopyrimidine with furfurylamine in boiling water.

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In the synthesis of various potential antagonists of the natural purines 9-methyl-6-chloropurine (1) has shown the same order of activity against Adenocarcinoma 755 in C-57 black mice as 6-chloropurine[3] (2), while two other 9-methyl-6-substituted purines have shown less activity against this tumor. 9-Ethyl-6-chloropurine (3) and 9-propyl-6-chloropurine (4), synthesized by Montgomery and Temple, have also shown the same kind of activity.

Due to the structural similarity between naturally occurring metabolic purine ribonucleosides, deoxyribonucleosides, and 9-furfuryl-6-substituted purines, the investigation of the biological activity of 9-furfuryl-6-substituted purines could be of some interest. A comparison of the biological activity of 9-furfuryl-6-chloropurine with that of 9-methyl-, 9-ethyl-, 9-propyl-, and 9-phenyl-6-chloropurine (5) would be especially desirable.

Thirteen such derivatives of purine were synthesized through the method devised by Robins and Lin (1) and sent to the Cancer Chemotherapy National Service Center, National Institutes of Health, Bethesda, Maryland, for screening their biological activities. The mother compound 9-furfuryl-6-chloropurine (**5**) was prepared in good yield by direct cyclization of 4-furfurylamino-5-amino-6-chloropyrimidine (**4**) with a mixture of ethyl orthoformate and acetic anhydride.

Treatment of several aliphatic and aromatic amines with **5** in boiling amyl alcohol yielded corresponding amino derivatives in fairly good yield. Cyclization of 4-furfurylamino-5-amino-6-chloropyrimidine by refluxing with 97% formic acid failed to yield 9-furfuryl-6-hydroxypurine (**6**). A black sticky polymeric residue was the only product that could be isolated. The ring cleavage of furfuryl ring in the boiling formic acid could cause the formation of this polymeric residue. 9-Furfuryl-6-hydroxypurine (**6**) was prepared easily by treatment of the mother compound (**5**) with 1 *N* sodium hydroxide in good yield. 9-Furfuryl-6-methoxypurine (**9**) was prepared by treatment of **5** with sodium methoxide. 9-Furfuryl-6-aminopurine (9-furfuryladenine) (**8**) was prepared by heating **5** with excess concentrated ammonium hydroxide at 130° in a steel Parr bomb. 9-Furfuryl-6-mercaptopurine (**7**) was prepared in high yield by treatment of **5** with thiourea. The ultraviolet absorption spectra of the 9-furfuryl-6-substituted purines are in general very similar to that of 9-methyl-6-substituted purines.

Biological Activities

9-Furfuryl-6-mercaptopurine (**7**) was found to be active at 112 mg/Kg (T/C, 31%) against Adenocarcinoma 755. 4,6-Difurfurylamino-5-nitropyrimidine (**1**) was found to be non-toxic and inactive at 125 mg/Kg in the cell culture and cell line SA. 9-Furfuryl-6-chloropurine (**5**) and 9-furfuryl-6-hydroxypurine (**6**) were found to be inactive at 100 mg/Kg against Adenocarcinoma

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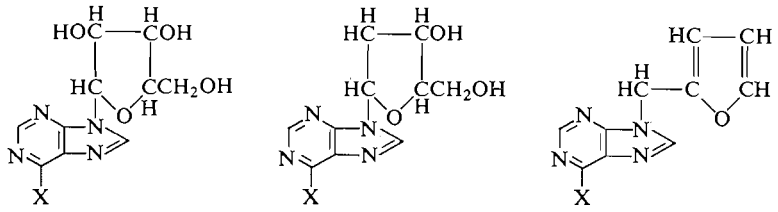
[2]The data published here is taken from the theses submitted by H. C. Hamann and V. T. Spaziano in partial fulfillment for the degree of Master of Science at Saint Joseph's College, Philadelphia, Pennsylvania.

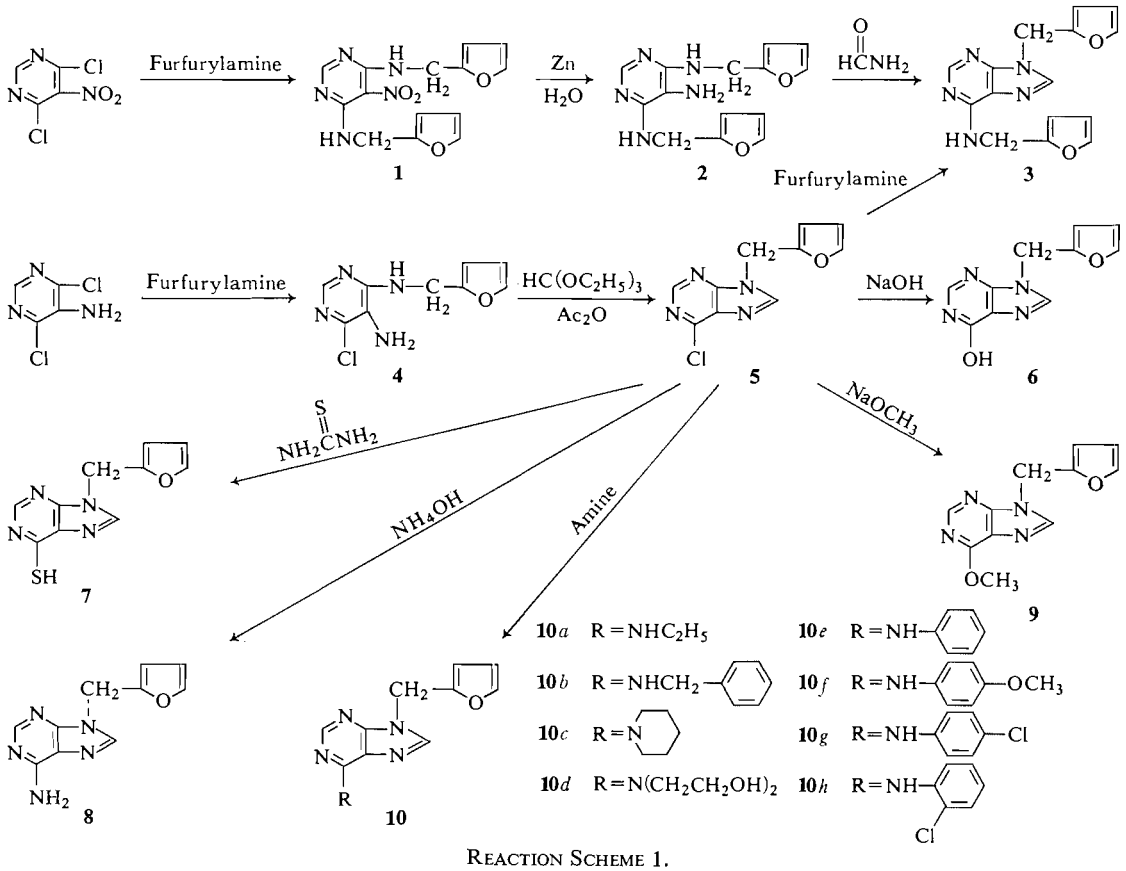
[3]H. E. Skipper, J. R. Thompson, and R. K. Robins. The Southern Research Institute, Birmingham, Alabama. Unpublished data.



Ribonucleoside
X = —OH, —NH_2

Deoxyribonucleoside
X = —OH, —NH_2

9-Furfuryl-6-substituted purine
X = —OH, —NH_2



REACTION SCHEME 1.

755. 9-Furfuryl-6-ethylaminopurine (**10***a*) and 9-furfuryl-6-methoxyanilinopurine (**10***f*) were found to be inactive at 10^{-3} mg/ml against the Leukemia L1210 and Walker 256 tumors.

Experimental

4,6-Difurfurylamino-5-nitropyrimidine (1)

A mixture of 5.8 g of 4,6-dichloro-5-nitropyrimidine and 11.6 g of furfurylamine in 150 ml of absolute ethanol was refluxed for 1 h. Upon cooling the reaction mixture in the refrigerator, yellow crystalline needles separated out. The crystals were filtered with suction and dried, yield 5.4 g. Upon evaporation of filtrate to half its original volume, further crystals (1.8 g) were collected, m.p. 128–131°. A small portion of crude product was recrystallized from a mixture of ethanol and water to yield yellow needles, m.p. 131–132°; total yield was 77%.

Anal. Calcd. for $C_{14}H_{13}N_5O_4$: C, 53.33; H, 4.17; N, 22.21. Found: C, 53.61; H, 4.31; N, 22.13.

4,6-Difurfurylamino-5-aminopyrimidine (2)

To constantly stirred boiling water (500 ml) containing 200 g of zinc dust was added in small portions 4 g of 4,6-difurfurylamino-5-nitropyrimidine. The reaction mixture was digested for an additional 10 min.

After the addition, the mixture was filtered with suction while still hot. Slightly tan needles separated out upon cooling. The crystals were collected and dried, gave a crude product of 1.1 g, m.p. 114–117°. A small amount of crude product was recrystallized from water to give white crystals, m.p. 116–117°, yield 31%.

Anal. Calcd. for $C_{14}H_{15}N_5O_2$: C, 58.94; H, 5.29; N, 24.55. Found: C, 59.02; H, 5.16; N, 24.40.

9-Furfuryl-6-furfurylaminopurine (3)

Method A

4,6-Difurfurylamino-5-aminopyrimidine (2 g) was added to 6 ml of formamide. The mixture was then heated to boiling on a hot plate for 15 min. As the triaminopyrimidine dissolved in the hot formamide, the resultant solution became dark and viscous. The liquid was then extracted with 150 ml of hot boiling benzene. The benzene solution was then treated with activated charcoal and filtered. The filtrate was then evaporated to half its original volume and *n*-heptane was added until it became cloudy. Upon cooling, tan solid separated out, m.p. 123–126°, yield 1.2 g. A portion of the crude product was recrystallized from a mixture of ethanol and water to give m.p. 127–128°, yield 57%.

Anal. Calcd. for $C_{15}H_{13}N_5O_2$: C, 61.02; H, 4.43; N, 23.72. Found: C, 61.04; H, 4.42; N, 23.60.

Method B

A mixture of 9-furfuryl-6-chloropurine (0.59 g) and furfurylamine (0.49 g) in 20 ml of 2-ethoxyethanol was refluxed for 3 h. The residue was then distilled to dryness *in vacuo* and recrystallized from a mixture of water and ethanol to give tan crystals, m.p. 120–124°, yield, 0.42 g. Further recrystallization gave m.p. 127–128°. A mixture melting point showed that it is identical with the product prepared from Method A, yield 74%.

4-Furfurylamino-5-amino-6-chloropyrimidine (4)

A mixture of 3.28 g of 4,6-dichloro-5-aminopyrimidine and 4.0 g of furfurylamine in 60 ml water was heated on a steam bath for 4 h. The dark-brown oil separated and crystallized upon cooling the reaction mixture in the refrigerator. The crude product was collected and dried, m.p. 126–129°, yield 4.1 g. A small portion of the crude product was recrystallized from a mixture of water and ethanol to give tan prisms of m.p. 130–131°, yield 91%.

Anal. Calcd. for $C_9H_9N_4ClO$: C, 48.11; H, 4.04; N, 24.94; Cl, 15.78. Found: C, 48.43; H, 4.20; N, 25.15; Cl, 15.59.

9-Furfuryl-6-chloropurine (5)

4-Furfurylamino-5-amino-6-chloropyrimidine (6 g) was added to a mixture of 60 ml of triethylorthoformate and 60 ml of acetic anhydride. The mixture was refluxed for 3 h and distilled to near dryness *in vacuo*. The viscous black residue was then dissolved in 200 ml of hot benzene, and charcoal was added. The mixture was filtered and the filtrate was evaporated to half of its original volume. To the hot solution, *n*-hexane was added until the mixture became slightly cloudy. Upon cooling of the solution, a sticky brownish-yellow solid crystallized out. Further scratching of the sticky semicrystalline mass yielded more crystalline solid. The crude product was collected, m.p. 105–109°, yield 4.5 g. A portion of the sticky solid was recrystallized again from a mixture of benzene and hexane, m.p. 109–112°, yield 72%.

Anal. Calcd. for $C_{10}H_7N_4ClO$: C, 51.18; H, 3.00; N, 23.87; Cl, 15.11. Found: C, 51.02; H, 2.80; N, 23.81; Cl, 15.68.

9-Furfuryl-6-hydroxypurine(9-furfurylhypoxanthine) (6)

9-Furfuryl-6-chloropurine was added to 40 ml of 1 *N* NaOH. The mixture was heated on a steam bath for ½ h. The chloropurine dissolved completely into the alkaline solution at the end of heating. The dark-brown solution was treated with charcoal and filtered. The filtrate was acidified with glacial acetic acid. Light-tan solids precipitated, were collected, and dried, m.p. 260–264°, yield 0.6 g. A small portion of the crude product was recrystallized from water, m.p. 265° (decomp.), yield 65%.

Anal. Calcd. for $C_{10}H_8N_4O_2$: C, 55.55; H, 3.73; N, 25.92. Found: C, 55.81; H, 3.92; N, 25.59.

9-Furfuryl-6-mercaptorpurine (7)

A mixture of 9-furfuryl-6-chloropurine (2.35 g) and thiourea (0.8 g) in 60 ml of 2-ethoxyethanol was refluxed for 3 h. A light-tan solid separated and was filtered. The filtrate was distilled to dryness *in vacuo*. The residue was combined with the light-tan solid and dissolved in 50 ml of 5% hot NaOH solution. The solution was treated with activated charcoal and filtered. The filtrate was acidified with glacial acetic acid to yield 2.1 g of light-tan solid, which became pale-yellow upon drying, m.p. 315° (decomp.). A small portion of the product was suspended in hot water with a minimum amount of NaOH to ensure complete dissolution. The solution was filtered and acidified with glacial acetic acid. White crystalline solids separated, were collected, and dried, m.p. 315° (decomp.), yield 90%.

Anal. Calcd. for $C_{10}H_8N_4SO$: C, 51.72; H, 3.44; N, 24.13; S, 13.79. Found: C, 51.52; H, 3.64; N, 24.18; S, 13.59.

9-Furfuryl-6-aminopurine(9-furfuryladenine) (8)

A mixture of 9-furfuryl-6-chloropurine (1.0 g) and 20 ml of concentrated NH_4OH was sealed in a steel Parr bomb (40 ml). The bomb was heated to 130° in a steel high pressure bomb (500 ml) for 3 h. The mixture was taken out from the bomb upon cooling to room temperature. White crystalline prisms separated and were filtered, m.p 195–201°, yield 0.6 g. A small portion of the crude product was recrystallized from hot water to give m.p. 202–205°, yield 65%.

Anal. Calcd. for $C_{10}H_9N_5O$: C, 55.81; H, 4.22; N, 32.54. Found: C, 55.90; H, 4.41; N, 32.25.

9-Furfuryl-6-methoxypurine (9)

9-Furfuryl-6-chloropurine (1.4 g) was added to 100 ml of absolute methanol in which 0.13 g of sodium metal was dissolved. The mixture was refluxed on the steam bath for ½ h. A small amount of sodium chloride which precipitated was filtered off. The filtrate was then evaporated to about 25 ml, then allowed to cool. Colorless crystalline needles separated, were collected, and were dried, m.p. 100–103°, yield 0.8 g. A small portion of the crude product was recrystallized from water (0.7 g), m.p. 103–104°, yield 58%.

Anal. Calcd. for $C_{11}H_{10}N_4O_2$: C, 57.38; H, 4.37; N, 24.33. Found: C, 57.64; H, 4.15; N, 24.38.

TABLE I

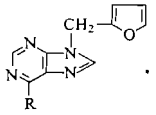
9-Furfuryl-6-anilinopurines

Compound	Yield %	Melting point °C	Formula	Recrystallization solvent	Analysis							
					Calculated, %				Found, %			
					C	H	N	Cl	C	H	N	Cl
10*a*	55	125–127	$C_{12}H_{13}N_5O$	water	59.25	5.39	28.79		59.02	5.27	28.75	
10*b*	76	124–125	$C_{17}H_{15}N_5O$	water and ethanol mixture	66.88	4.95	22.94		67.26	5.36	22.75	
10*c*	52	151–153	$C_{15}H_{18}N_5OCl$	ethanol	56.33	5.67	21.89	11.09	56.37	5.94	22.18	11.06
10*d*	59	109–110	$C_{14}H_{17}N_5O_3$	ethyl acetate	55.43	5.65	23.08		55.36	5.43	23.18	
10*e*	53	151–152	$C_{16}H_{13}N_5O$	water and ethanol mixture	65.95	4.49	24.08		66.11	4.47	24.08	
10*f*	31	137–139	$C_{17}H_{15}N_5O_2$	water and ethanol mixture	63.54	4.70	21.79		63.53	4.79	21.99	
10*g*	62	155–158	$C_{16}H_{12}N_5OCl$	water and ethanol mixture	58.99	3.71	21.49	10.88	59.08	3.79	21.61	10.60
10*h*	73	143–144	$C_{16}H_{12}N_5OCl$	water and ethanol mixture	58.99	3.71	21.49	10.88	59.06	3.73	21.28	10.50

TABLE II

The ultraviolet absorption maxima of 9-furfuryl-6-substituted purines*

R	0.1 *N* HCl, λ_{max} mμ	ε	0.1 *N* NaOH, λ_{max} mμ	ε
OH	248	1.5×10^4	254	1.7×10^4
SH	insoluble		311	2.0×10^4
Cl	264	1.0×10^4	264	1.1×10^4
OCH_3	264	1.5×10^4	255	1.2×10^4
$-NHCH_2CH_3$	266	1.7×10^4	270	1.9×10^4
$-NHCH_2C_6H_5$	268	1.9×10^4	270	2.0×10^4
$-NC_5H_{10}$	279	0.69×10^4	282	2.4×10^4
$-N(CH_2CH_2OH)_2$	276	2.5×10^4	280	2.5×10^4
p-$C_6H_4-OCH_3$	277	2.5×10^4	286	2.3×10^4
$-NHCH_2$-furyl	267	1.5×10^4	268	2.0×10^4

*Compounds of structure  .

9-Furfuryl-6-ethylaminopurine (10a)

A mixture of 9-furfuryl-6-chloropurine (1.4 g) and 20 ml of aqueous ethylamine (70%) was heated in a steel Parr bomb at 120° for 3 h. The mixture was then evaporated to dryness on a steam bath. The brown residue was recrystallized from water to give analytically pure product.

9-Furfuryl-6-benzylaminopurine (10b), 9-Furfuryl-6-piperidinopurine Monohydrochloride (10c), 9-Furfuryl-6-N,N-bis(2-hydroxyethyl)amino Purine (10d), 9-Furfuryl-6-anilinopurine(10e), 9-Furfuryl-6-p-methoxyanilinopurine (10f), 9-Furfuryl-6-p-chloroanilinopurine (10g), and 9-Furfuryl-6-o-chloroanilinopurine (10h)

A mixture of 9-furfuryl-6-chloropurine (0.005 mole) and corresponding aniline (0.01 mole) in 50 ml of *n*-amyl alcohol was refluxed for 3 h. The solution was distilled to dryness under reduced pressure. The residue was then recrystallized from a mixture of water and ethanol to give light-tan crystals. The crude products were further purified to analytically pure compounds. The data for each compound are tabulated in Table I.

Acknowledgments

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