moval of solvent the solid was recryst (C_6H_6–hexane); yield 4.2 g (41%), mp 151–152°. Hydrogenation of 1.0 g with 20% Pd–C gave the amino compound isolated as the HCl salt from EtOH–Et_2O, mp 193–199°; yield 0.83 g (69%).

3,4-Dihydroisoquinolines.—The amino-substituted dihydroisoquinolines were prepared by a slight modification of the Bischler–Napieralski reaction[32] using the corresponding *N*-phenethylnitrobenzamides followed by hydrogenation. Noncrystalline bases were converted into crystalline HCl salts.

7-Methoxy-1-(*o*-aminophenyl)-3,4-dihydroisoquinoline (46).—To a solution of 18 g (0.060 mol) of *N*-(*p*-methoxyphenethyl)-*o*-nitrobenzamide in 60 ml of $(CHCl_2)_2$ and 40 ml $POCl_3$ was added 14 g of P_2O_5 with rapid stirring. The solution was stirred under reflux 1 hr and most of the solvent removed *in vacuo*. The residue was poured into ice–H_2O and extracted 3 times with Et_2O. The aq layer, after filtration, was made alkaline with NaOH and extracted with CH_2Cl_2. After drying and evaporation *in vacuo* the nitro compound was obtained as a yellow gum (13.5 g). The product was dissolved in MeOH and hydrogenated with 0.5 g of Raney Ni at 3.5 kg/cm². Filtration and concentration afforded 8.7 g (58%) of product, mp 118–119° (MeOH–H_2O).

1-(*o*-Aminophenyl)isoquinoline (58).—A suspension of 5.3 g (0.024 mol) of 1-(*o*-aminophenyl)-3,4-dihydroisoquinoline and 0.3 g of 20% Pd–C in 50 ml of decalin was stirred at reflux for 20 hr. The cooled solution was diluted with C_6H_6, filtered, and coned *in vacuo*. The solid was filtered and recrystd twice (C_6H_6–hexane): yield 2.5 g (47%); mp 153–154°; λ_{max}^{MeOH} 322 mμ (ε 5100), 310 (4530), 283 (6500), 273 (6820), and 22 (66700).

(32) *Org. React.*, **6**, 74 (1951).

Acknowledgment.—The authors wish to express their appreciation to Dr. Frank Brandon for aid in the viral assays, Dr. J. M. Vandenbelt for spectral analyses, Mr. C. E. Childs for microanalyses, and Mr. W. Pearlman for hydrogenations.

Synthesis and Pharmacological Evaluation of Some Pyridylmethyl Substituted Ethylenediamines

SEIJI MIYANO, AKIKO ABE,

Department of Synthetic Chemistry, Faculty of Pharmaceutical Sciences, Fukuoka University, Fukuoka, Japan

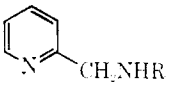
YOSHITOSHI KASÉ, TOMOKAZU YUIZONO, KUNIOMI TACHIBANA, TAKESHI MIYATA, AND GO KITO

Department of Chemico-Pharmacology, Faculty of Pharmaceutical Sciences, Kumamoto University, Kumamoto, Japan

Received August 22, 1969

A series of new 2-pyridylmethyl (picolyl) substituted ethylenediamines has been prepared. Comparative pharmacological screening of the series included tests for antihistaminic and antispasmodic activity. *N,N*-Dimethyl-*N'*-phenyl-*N'*-(2-picolyl)ethylenediamine was the most active antihistaminic and had about one-third the activity of tripelennamine. *N*-(2-Picolyl)-*N*-phenyl-*N*-(2-piperidinoethyl)amine was the most active antitussive compound, being somewhat more potent in this respect than codeine. Comparison of structure–activity relationships in the series supports the hypothesis of Kasé and Yuizono that a piperidino group in the molecule enhances antitussive activity.

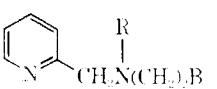
The ready preparation[1,2] of a number of *N*-(2-picolyl)anilines (I) and *N*-(2-picolyl)-2-aminopyridines (II) in our laboratory prompted us to attempt the preparation of a series of new ethylenediamine-type antihistaminic with a 2-picolyl group attached to N and to ascertain whether the introduction of a 2-picolyl group may provide desirable pharmacological activities.



I, R = phenyl, substituted phenyl
II, R = 2-pyridyl

The favorable effect of the replacement of benzyl by a thenyl group in the ethylenediamine series has been reported by previous workers.[3–5] However, no synthesis of the isosteres in which a 2-picolyl group is introduced in place of benzyl has been reported.

In this paper the synthesis of a series of 2-picolyl substituted ethylenediamines which are represented as a general formula (III) in which R is phenyl, substituted phenyl, or 2-pyridyl, and B is Me_2N, Et_2N, pyrrolidino, or piperidino, and their pharmacological screening results are recorded. Compound **1** (Table I) is an isomer of the known antihistamine, tripelennamine, since the position of two groups, phenyl and 2-pyridyl, are reversed.



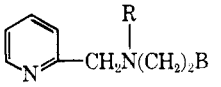
III

The syntheses of these antihistaminic tertiary amines were carried out by alkylating the *N*-picolyl secondary amines with dialkylaminoethyl chloride. The condensation was conducted by refluxing the reactants in toluene in the presence of $NaNH_2$. The intermediate secondary amines were prepared according to the general procedure we recently reported.[1,2] One *N*-(4-picolyl) analog **10** was also prepared.

Pharmacological screening included studies of acute toxicity, antihistaminic, antispasmodic, and antitussive activities.

(1) S. Miyano, *Chem. Pharm. Bull.*, **13**, 1135 (1965).
(2) S. Miyano, A. Uno, and N. Abe, *ibid.*, **15**, 515 (1967).
(3) A. W. Weston, *J. Amer. Chem. Soc.*, **69**, 980 (1947).
(4) R. C. Clapp, J. H. Clark, J. R. Vaughen, J. P. English, and G. W. Anderson, *ibid.*, **69**, 1549 (1947).
(5) L. P. Kyrides, F. C. Meyer, F. B. Zienty, J. Harvey, and L. W. Bannister, *ibid.*, **72**, 745 (1950).

TABLE I
2-PICOLYL SUBSTITUTED ETHYLENEDIAMINES



			Free base			HCl salt	
Compd	R	B	Yield, %	Bp, °C (mm)	Formula	Mp,[e] °C	Formula
1	Ph	$N(CH_3)_2$	50	186–189 (4)	$C_{16}H_{21}N_3$[g]	263–265[f]	$C_{16}H_{22}ClN_3$[h]
2	Ph	*a*	76.3	195–196 (4)	$C_{19}H_{25}N_3$	183–185	$C_{19}H_{26}ClN_3$
3	Ph	*b*	49.5	193–196 (5)	$C_{18}H_{23}N_3$	196–197	$C_{18}H_{24}ClN_3$
4	*p*-MeOPh	$N(C_2H_5)_2$	47.0	176–178 (4.5)	$C_{19}H_{17}N_3O$	186–188	$C_{19}H_{28}ClN_3O$
5	*p*-MeOPh	*a*	34.5	187–195 (4.5)	$C_{20}H_{27}N_3O$	178–179	$C_{20}H_{28}ClN_3O$
6	*p*-MeOPh	*b*	22.9	200–203 (4)	$C_{19}H_{25}N_3O$	260–262	$C_{19}H_{26}ClN_3O$
7	2-Py	$N(CH_3)_2$	60.1	161–162 (4)	$C_{15}H_{20}N_4$	228–229[f]	$C_{15}H_{21}ClN_4$
8	2-Py	$N(C_2H_5)_2$	66.5	185–186 (4.5)	$C_{17}H_{24}N_4$	222–224[f]	$C_{17}H_{25}ClN_4$
9	2-Py	*b*	57.5	198–204 (4)	$C_{17}H_{22}N_4$	204–205	$C_{17}H_{23}ClN_4$
10[c]	Ph	*a*	42.5	201–203 (2)[d]	$C_{19}H_{25}N_3$	204–205	$C_{19}H_{26}ClN_3$

[a] Piperidino. [b] Pyrrolidino. [c] 4-Picolyl substituted ethylenediamine. [d] Mp 83.5–84.5° (from Et_2O). [e] All salts were recrystallized from EtOH–Et_2O. [f] With decomposition. [g] All bases were analyzed for C, H, N. [h] All HCl salts were analyzed for C, H, Cl, N.

Experimental Section

Chemistry. Intermediate Secondary Amines.—All the amines were prepared by condensation between aromatic amines and 2- and 4-pyridinemethanol in the presence of KOH at 225–240°.[1,2]

2- and 4-Picolyl Substituted Ethylenediamines.—2- and 4-Picolyl-substituted ethylenediamines listed in Table I were prepared by a procedure illustrated by the following example.

***N*-(2-Picolyl)-*N*-phenyl-*N*-(2-piperidinoethyl)amine (III, R = C_6H_5, B = piperidino).**—To a stirred and refluxing suspension of 5.6 g (0.144 mol) of $NaNH_2$ in 60 ml of dry toluene was added dropwise a solution of 18.4 g (0.1 mol) of *N*-(2-picolyl)aniline in 20 ml of dry toluene. After the addition was complete the reaction mixture was refluxed for 2 hr under constant stirring. β-Piperidinoethyl chloride (14.9 g, 0.101 mol) in 20 ml of dry toluene was then added dropwise with stirring. After refluxing for 2 hr, the mixture was cooled and H_2O was added carefully to decompose excess $NaNH_2$. The oily layer which separated was dried (Na_2SO_4), and the toluene removed under reduced pressure. The residual oil was distilled *in vacuo* and a fraction boiling at 185–198° (4 mm) was collected. Redistillation afforded of 22.5 g (76.3%) of pale yellow liquid, bp 195–196° (4 mm).

Pharmacology.—Hydrochlorides of the test compounds (Table I) were used for pharmacological tests.

Toxicity.—dd-Strain male mice weighing 15–20 g were used. Toxic symptoms were observed after subcutaneous administration of the test compounds and the LD_{50} with fiducial limits (p = 0.05) were calculated by the method of Litchfield and Wilcoxon.[6]

Antihistaminic, Anticholinergic, and Anti-$BaCl_2$ Activities.—The inhibitory effects on histamine-, ACh-, and $BaCl_2$-induced contractions of the isolated guinea pig ileum were examined by the method of Magnus.[7] The ED_{50}, which inhibits maximal contraction to 50% by the pretreatment of the compounds (3 min before), was calculated by plotting per cent inhibition against log dose.

Antitussive Activity.—The "coughing dog" method[8] was used. Coughing was induced by mechanical stimulation on the mucosa tracheal bifurcation through a chronically built fistula. The evaluation was made according to the criterion previously reported[8] and the effect was expressed in terms of 50% antitussive dose (AtD_{50}).

Studies on the following pharmacological activities were further carried out with **1**, **2**, and **3**.

Antihistaminic Activity.—(a) Protective action against death induced by a fatal dose of histamine in the guinea pig. The test compounds were administered ip to animals weighing 180–250 g. Histamine·2HCl (1.21 mg/kg) was given iv 60 min later. The ED_{50}, which was effective to prevent 50% of the guinea pigs used from death within 24 hr after histamine injection, was calculated. (b) Antianaphylactic action in the guinea pig. Male guinea pigs (300–350 g) were treated ip with 5 ml of 25 % v/v solution of egg white in physiological saline; 20 days later, 0.25 ml of the same antigen solution was given iv to all the animals, 60 min after ip administration of the test compounds. The guinea pigs which survived for 24 hr after the reinjection were considered to be protected.

Effect on the Bronchial Muscles *in Vivo*.—In the urethanized male rabbit, the effect on the normal tone of bronchial muscles and spasmolytic actions of the test compounds were studied by the method of Jackson.[9] Bronchospasms were induced by iv administration of histamine·2HCl (5–15 μg/kg) or methacholine chloride (2–8 μg/kg).

Local Anesthetic Activity.—Surface anesthesia was examined by the corneal reflex method in the guinea pig,[10] infiltration anesthesia by the intracutaneous wheal method in the guinea pig.[10]

Analgetic, anticonvulsant, and antiemetic activities were also studied. Each of the testing methods used will be described where pertinent.

Results and Discussion

The results obtained are given in Tables II and III.

A. Acute Toxic Symptoms.—The acute toxic symptoms of all the test compounds, tripelennamine, and diphenhydramine in mice were chiefly those of central excitation; death occurred with respiratory paralysis during and after clonic convulsions. Occasional elevation of the tail, tremor of the whole body, and clonic convulsions were observed with all of the test compounds, tripelennamine, and diphenhydramine. The remarkable salvation that occurred with tripelennamine was not observed in any of the test compounds and diphenhydramine.

As shown in Table III, it appears that the replacement of the benzyl in tripelennamine by 2- and 4-picolyl results in a decrease in toxicity in mice.

B. Antihistaminic, Anticholinergic, and Anti-Ba Activities of the Isolated Guinea Pig.—Compounds **1** and **2** possess potent antihistaminic effects of an order similar to that of diphenhydramine. The other compounds were much less active. These results indicate that the reversal of Ph and 2-pyridyl in tripelennamine leads to a considerable decrease in antihistaminic ac-

(6) J. T. Litchfield and F. Wilcoxon, *J. Pharmacol. Exp. Ther.*, **96**, 99 (1949).

(7) R. Magnus, *Pflügers Arch.*, **102**, 123 (1904).

(8) Y. Kasé, *Jap. J. Pharmacol.*, **4**, 130 (1955); "Selected Pharmacological Testing Methods," A. Burger, Ed., Marcel Dekker, Inc., New York, N. Y., 1968, p 363.

(9) C. Jackson, "Experimental Pharmacology," C. V. Mosby Co., St. Louis, Mo., 1917, p 287.

(10) E. Bülbring and I. Wajda, *J. Pharmacol. Exp. Ther.*, **85**, 78 (1945).

TABLE II

PHARMACOLOGICAL ACTIVITIES OF PICOLYL SUBSTITUTED ETHYLENEDIAMINES

	Toxicity (mice)		Antihistaminic activity[a]			Antispasmodic activity[a]				Antitussive activity[b]	
						Acetylcholine		$BaCl_2$			
Compd	LD_{50}, mg/kg sc	Ratio of toxicity to tripelennamine	Ratio of toxicity to codeine	ED_{50}, g/ml	Ratio of effect to tripelennamine	Ratio of effect to diphenhydramine	ED_{50}, g/ml	Ratio of effect to atropine	ED_{50}, g/ml	Ratio of effect to papaverine	AD_{50}, mg/kg i.v.	Ratio of effect to codeine
1	290 (254–331)	27	66	8×10^{-8}	31.3	107.5	2.5×10^{-5}	0.2	1.5×10^{-5}	33.3	3.98 (3.55–4.46)	63
2	210 (185–239)	37	91	9×10^{-8}	27.8	95.6	3×10^{-5}	0.2	9×10^{-6}	55.5	2.36 (2.08–2.68)	106
3	194 (164–229)	40	98	2.5×10^{-7}	10.0	34.4	4×10^{-5}	0.1	1.2×10^{-5}	41.7	2.86 (2.46–3.32)	87
4	550[c]	14	35	8×10^{-5}	<0.1	0.1					Ineffective up to 10.0	
5	450[c]	17	42	1.8×10^{-4}	<0.1	<0.1	$>1 \times 10^{-4}$	<0.1	6×10^{-5}	8.3	5.90 (5.16–6.74)	42
6	500[c]	16	38	8×10^{-5}	<0.1	0.1	$>1 \times 10^{-4}$	<0.1	3×10^{-4}	1.7	6.00 (5.12–7.03)	42
7	420[c]	19	45	9×10^{-6}	0.3	1.0	$>1 \times 10^{-4}$	<0.1	1×10^{-4}	5.0	Ineffective up to 20.0	
8	550[c]	14	35	2×10^{-5}	0.1	0.4	$>1 \times 10^{-4}$	<0.1	1.5×10^{-4}	3.3	Ineffective up to 20.0	
9	260[c]	30	73	8×10^{-7}	3.1	10.8	$>1 \times 10^{-4}$	<0.1	1.5×10^{-4}	3.3	5.60 (4.73–6.63)	45
10	140 (116–169)	56	136	5×10^{-6}	0.5	1.7	$>1 \times 10^{-4}$	<0.1	6×10^{-5}	8.3	3.64 (2.96–4.48)	69
Tripelennamine·HCl	78 (67–91)	100	245	2.5×10^{-5}	100.0	344.0	9×10^{-6}	0.6	2.5×10^{-5}	20.0	2.74 (2.32–3.24)	91
Diphenhydramine·HCl	175 (145–199)	45	109	8.6×10^{-8}	29.1	100.0	7×10^{-6}	0.7	8×10^{-9}	62.5	4.36 (3.43–5.32)	57
Codeine phosphate	191 (178–205)	41	100								2.50 (2.31–2.70)	100
						Atropine·H_2SO_4		Papaverine·HCl				
						5×10^{-8}	100.0	5×10^{-6}	100.0			

[a] Antihistaminic and antispasmodic activities were examined using the isolated guinea pig ileum by the method of Magnus. Spasms were induced by histamine·2HCl (10^{-6}), acetylcholine chloride (10^{-6}), and $BaCl_2$ (4×10^{-4} g/ml), respectively. [b] Antitussive activity was evaluated by the "coughing dog" method. Figures in parenthesis show the fiducial limits ($p = 0.05$). [c] Minimal lethal dose.

TABLE III
PHARMACOLOGICAL ACTIVITIES OF N-(2-PICOLYL)-N-PHENYLETHYLENEDIAMINES

Drugs	Protection against lethal dose of histamine (guinea pig) ED_{50}, mg/kg ip	Antianaphylactic activity (guinea pigs) ED_{100}, mg/kg ip	Effects on the bronchial muscles[a] (rabbits, *in vivo*) Normal tone	Anti-histamine[b] ED_c, mg/kg iv	Antimethacholine[b] ED_c, mg/kg iv	Local anesthetic activity (guinea pigs) Surface (relative potency)	Infiltration (relative potency)
1	2.9[d]	10.0	Nc	0.2	5.0	0.6	2.1
2	31.5 (20.2–49.1)	Ineffective up to 40.0	Nc	0.2	2.0	3.0	2.9
3	2.8[d]	15.0	Nc	0.1	2.0	1.7	2.1
Tripelennamine·HCl	0.16 (0.09–0.27)	1.0	Nc	0.01	2.0		
Diphenhydramine	3.8 (2.3–6.2)	10.0	Nc	0.05	0.5	2.1	1.1
						Procaine·HCl 1.0	Procaine·HCl 1.0

[a] Effect on the bronchial muscles was examined by the method of Jackson. [b] Spasms were induced by histamine·2HCl (5–15 μg/kg iv) and methacholine chloride (2–8 μg/kg iv), respectively. [c] Dose causing complete relaxation; Nc = no change. [d] Minimal effective dose. Figures in parenthesis mean the fiducial limits ($p = 0.05$).

tivity. However, **1** and **2** retained significant antihistaminic activities.

The test compounds, in general, had no significant antagonistic effects against ACh-induced spasms, that is, the *N*-(2-picolyl)-*N*-phenyl derivatives (**1**, **2**, and **3**) were only 0.002–0.0001 as active as atropine, and the potency of the other compounds was much lower. Spasmolytic effects of the test compounds on Ba^{2+}-induced spasms were relatively weak as compared with that of papaverine. The efficacies of the most active compounds (**1**, **2**, **3**) were about $^1/_3$, $^1/_2$, and $^2/_5$ that of papaverine, respectively.

C. Antitussive Activity.—Compounds **1, 2, 3,** and **10** exhibited potent antitussive effects in the dog, **5, 6,** and **9** being moderately potent antitussives. With the exception of **1** they all have a piperidino or pyrrolidino group; the potency of **2**, in particular, which possesses a piperidino group, was 1.1 times that of codeine, hence the most potent of this series.

These results have given conclusive evidence to a working hypothesis on antitussive activity presented by Kase and Yuizono[11,12] that introduction of a piperidino or pyrrolidino group as a basic moiety to a compound showing any actions on the CNS can produce antitussive activity which has been latent otherwise, or strengthen it if such activity has already been manifest.

The above observations show that the reversal of the positions of the phenyl and 2-pyridyl group in the tripelennamine structure results in some decrease in antitussive activity, and that the introduction of a piperidino or pyrrolidino group can restore the potency to a certain degree and cancel out the interchange.

Other pharmacological activities of **1**, **2**, and **3** were studied further seeking for promising antihistaminic or antitussive agents. The results are shown in Table III.

D. The protective effect against an intravenous lethal dose of histamine in the guinea pig was especially noticeable with **1** and **3**; the effects were more pronounced than for diphenhydramine, but far less potent than for tripelennamine. On the other hand, **2** which showed the most potent antitussive activity was found to be the weakest antihistaminic of the three compounds.

E. Antianaphylactic Action in the Guinea Pig.—Antianaphylactic activities of **1** and **3** were equal to $^2/_3$ that of diphenhydramine, respectively, while **2** was not active at the dose levels used. Of the compounds employed, tripelennamine was the most potent in this respect.

F. Effect on the Bronchial Muscles.—The relaxation of contracted bronchial muscles results in a decrease in the intensity of coughs, therefore, the compounds relax the bronchial muscles. On the other hand, they showed potent spasmolytic effects on histamine-induced contraction, but were considerably less active than diphenhydramine and tripelennamine.

Spasmolytic effects of the three compounds on methacholine-induced contraction were relatively weak.

G. Local Anesthetic Activity.—The compounds showed surface and infiltration anesthesia. Of the three compounds tested, **2** showed the strongest activity.

H. Analgetic Activity.—No significant analgesic effect in mice was observed with the three test compounds in doses up to $0.5LD_{50}$ (sc), when tested by the method of Haffner.[13] Determined by the same method, codeine showed a definite effect, and the ED_{50} was 16(12–20) mg/kg.

I. Anticonvulsant Activity.—Maximal electroshock seizure (MES) was induced in mice using the apparatus of Woodbury and Davenport.[14] The test compounds were given subcutaneously before an application of electroshock and 50% effective doses for inhibiting MES were calculated.

Compounds **1** and **3** showed no effect with doses up to 150 mg/kg, and **2** showed a definite inhibitory effect and ED_{50} was found to be 71(56–90) mg/kg. Both diphenhydramine and phenobarbital were more potent than **2**, and ED_{50} were 30(22–41) and 29(23–37) mg/kg, respectively. Tripelennamine, however, showed no significant effect in doses up to $0.5LD_{50}$.

J. Antiemetic Activity.—The depressing activity of

(11) Y. Kasé and T. Yuizono, *Chem. Pharm. Bull.*, **7**, 378 (1959).
(12) Y. Kasé, T. Yuizono, and M. Muto, *J. Med. Chem.*, **6**, 118 (1963).
(13) F. Haffner, *Deut. Med. Wochschr.*, **55**, 731 (1929).
(14) L. A. Woodbury and V. D. Davenport, *Arch. Int. Pharmacodyn. Ther.*, **92**, 97 (1952).

the three compounds on apomorphine-induced emesis in dogs was not observed with doses up to 10 mg/kg (sc), while chlorpromazine was able to block the emesis completely with 1.5 mg/kg.

In summary, among the ten compounds tested, **2** with favorable pharmacological properties seems to be a promising antitussive agent in that it shows approximately equal antitussive effect to codeine with slightly less toxicity. Here also the role of piperidino group in manifestation of antitussive activity was illustrated. This substance is now undergoing clinical trials as an antitussive agent.

Synthesis of 5-Mercaptouridine[1,2]

GABOR L. SZEKERES AND THOMAS J. BARDOS[3]

Department of Medicinal Chemistry, School of Pharmacy, State University of New York at Buffalo, Buffalo, New York 14214

Received November 14, 1969

Reaction of 5-acetylmercapto-2,4-bis(trimethylsiloxy)pyrimidine (I) with (anomeric) 2,3,5-tri-*O*-benzoyl-D-ribofuranosyl chloride led to an anomeric mixture of the blocked nucleoside. Reaction of I with 2,3,5-tri-*O*-*p*-anisoyl-D-ribofuranosyl chloride resulted, instead of coupling with the blocked ribosyl group, in *p*-anisoylation of the N_1 position of the pyrimidine. Stereoselective synthesis of 5-mercaptouridine (MUR) was achieved by fusion *in vacuo* of I with 2,3,5-tri-*O*-*p*-chlorobenzoyl-α-D-ribofuranosyl bromide (prepared from the anomerically pure β-1-*p*-nitrobenzoate), followed by removal of the blocking groups. MUR showed inhibitory activity on preliminary testing in bacterial and tissue culture assays.

Among a number of 5-substituted uridine derivatives which showed growth inhibition in bacterial and viral (tissue culture) assay systems, 5-hydroxyuridine was found to be the most effective antimetabolite[4] and to possess significant *in vivo* activity as an antitumor agent.[5] This compound undergoes most of the biochemical reactions of uridine including conversion into the mono-, di-, and triphosphates and minor incorporation into RNA.[6] Its 5′-monophosphate, 5-hydroxyuridylic acid, is a strong inhibitor of orotidylic acid decarboxylase,[6] and its triphosphate, 5-hydroxy-UTP, acts as a competitive inhibitor of UTP in the RNA polymerase reaction.[7] Roy-Burman, *et al.*, demonstrated that the inhibitory effect of 5-hydroxy-UTP is lowest at pH 7.0 and increases with increasing pH values to a maximum at pH 9.0, while the ability of this analog to replace UTP as a substrate for RNA polymerase (*i.e.*, to incorporate into RNA) showed the opposite pH dependence.[7] Thus, the inhibitory effect of the analog is predominant when its 5-hydroxyl group (pK_a 7.2) is in the ionized form, in contrast to its utilization as a substrate which appears to be suppressed by ionization.[7]

In view of the much lower pK_a value (pK_a 5.0)[8] of the previously synthesized 5-mercapto-2′-deoxyuridine (MUdR)[9] which, in its essentially ionized form (pH 7.4), was shown to undergo phosphorylation by thymidine kinase[10] and, subsequently, to act as a potent competitive inhibitor of dUMP in thymidylate synthetase,[11] it appeared of interest to prepare the corresponding uridine analog, 5-mercaptouridine (MUR; XIV). For the synthesis of this compound, the silyl modification of the Hilbert–Johnson reaction[12] appeared to be the method of choice.

Reaction of 5-acetylmercapto-2,4-bis(trimethylsiloxy)pyrimidine[9] (I) with 2,3,5-tri-*O*-benzoyl-D-ribofuranosyl chloride (*II*)[13] yielded, after hydrolysis of the 4-*O*-silyl group, an anomeric mixture of the blocked nucleoside III (Scheme I). The nmr spectrum of this product showed two close but separate peaks for the CH_3 of the *S*-acetyl group, at δ 2.30 and 2.36 ppm. Examination of the nmr spectra of the previously synthesized pure α and β anomers of *S*-acetyl-N_1-[3′,5′-di-*O*-*p*-chlorobenzoyl-2′-deoxy-D-ribofuranosyl]-5-mercaptouracil[9] similarly revealed a difference (of 2 Hz) between the chemical shifts for the *S*-acetyl protons of the two anomers, the corresponding resonance peaks being located at δ 2.38 ppm for the α, and 2.35 ppm for the β anomer. By analogy, in the spectrum of III the resonance peak at the higher field (2.30 ppm) would correspond to the β anomer; integration indicated that the ratio of α and β anomers was approximately 1:2. Separation of the two anomers of III proved to be quite difficult, and therefore, a more stereoselective method for the preparation of the β anomer was desired.

The above result is in agreement with previous reports[14] indicating that the *trans* rule is not applicable

(1) This work was supported by U.S. Public Health Service Research Grant R01-CA 06695-8 from the National Cancer Institute, National Institutes of Health, Bethesda, Md.

(2) A preliminary report on part of this work was presented to the Division of Medicinal Chemistry, 158th National Meeting of the American Chemical Society, New York, N. Y., Sept 1969.

(3) To whom inquiries should be addressed.

(4) I. J. Slotnik, D. W. Visser, and S. C. Rittenbert, *J. Biol. Chem.* **203**, 647 (1953).

(5) K. Sugiura and H. J. Creech, *Ann. N. Y. Acad. Sci.*, **63**, 962 (1955).

(6) D. A. Smith and D. W. Visser, *J. Biol. Chem.*, **240**, 446 (1965).

(7) S. Roy-Burman, P. Roy-Burman, and D. W. Visser, *ibid.*, **241**, 781 (1966).

(8) T. J. Bardos and T. I. Kalman, *J. Pharm. Sci.*, **55**, 606 (1966).

(9) (a) T. J. Bardos, M. P. Kotick, and C. Szantay, *Tetrahedron Lett.*, 1759 (1966); (b) M. P. Kotick, C. Szantay, and T. J. Bardos, *J. Org. Chem.*, **34**, 3806 (1969).

(10) T. J. Bardos, J. L. Ambrus, K. Baranski, T. I. Kalman, and C. Ambrus, *Proc. Amer. Ass. Cancer Res.* **8**, 4 (1967).

(11) (a) T. I. Kalman and T. J. Bardos, *Fed. Proc.*, **27**, 650 (1968); (b) K. Baranski, T. J. Bardos, A. Bloch, and T. I. Kalman, *Biochem. Pharmacol.*, **18**, 347 (1969).

(12) (a) T. Nishimura, B. Shimuzu, and I. Iwai, *Chem. Pharm. Bull.*, **11**, 1470 (1963); (b) T. Nishimura and I. Iwai, *ibid.*, **12**, 357 (1964); T. Nishimura, B. Shimuzu, and I. Iwai, *ibid.*, **12**, 1471 (1964); (d) E. Wittenburg, *Z. Chem.*, **4**, 303 (1964); and more recent publications.

(13) H. M. Kissman, C. Pidacks, and B. R. Baker, *J. Amer. Chem. Soc.*, **77**, 18 (1955).

(14) (a) J. D. Stevens, R. K. Ness, and H. B. Fletcher, Jr., *J. Org. Chem.*, **33**, 1806 (1968); (b) J. Farkas, L. Kaplan, and J. J. Fox, *ibid.*, **29**, 1469 (1964); (c) M. Pyrstas and F. Sorm, *Collect. Czech. Chem. Commun.*, **31**, 1035 (1966).