presumably the diiodate IIc which was removed and dried in air. The material was then suspended in ethanol (60 ml), the solid was removed, and the filtrate was retained. The extraction was repeated three times, the filtrates were combined, and the residue was discarded. To the combined filtrates excess NaI was added, precipitating IIa (3.39 g, 4.17 mmoles, 13.8%), mp 145–148° dec. Washing IIa with ethanol raised the melting point to 155–157° dec inserted at 145°, 6°/min; lit.[20] mp 158° dec.

To the solution remaining after IIc had been removed was added excess NaBr, precipitating a white solid which was washed with water until the washings were no longer basic and dried to give Ie (3.5 g, 9.7 mmoles, 32.4%), the infrared spectrum of which was superimposable with that of Ie prepared by another method.

Diphenyl-4,4'-biphenylyldiiodonium Dibromide (IIb).—A suspension of 4,4'-diiodoxybiphenyl (7.89 g, 16.5 mmoles), iodosobenzene (7.3 g, 33 mmoles), and 1 *N* NaOH (62.1 ml) was stirred for 3 hr, and the yellow residue IIc was removed from the reaction mixture and washed with 1.4 l. of water at 60°. To the filtrate was added excess NaBr, precipitating IIb (4.17 g, 36%), mp 183–186° dec. It was washed with water (50 ml) and ethanol (25 ml), raising the melting point to 185–186° dec inserted at 175°, 5°/min; lit.[20] mp 185° dec.

Acknowledgment.—We are grateful to Professor E. C. Jorgensen of the University of California for encouraging us to undertake this work.

Synthesis of Potential Antineoplastic Agents. XXXV. Phosphorus-Containing Structural Analogs of Myleran[1]

ROBERT F. STRUCK

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

Received September 16, 1965

Three classes of phosphorus-containing structural analogs of myleran have been synthesized in which the methanesulfonate group of myleran has been replaced by phosphonate groups (II) and by disubstituted phosphinate (III) and phosphinite groups (IV). Five of the analogs displayed cytotoxic activity at a concentration of 100 μg/ml against Eagle's KB cells. No significant *in vivo* activity was observed.

Among antineoplastic agents of the alkylating agent class, several compounds with chemical alkylating activity weaker than that of the nitrogen mustard type have been synthesized. Myleran (I) is a well-known representative of this type of alkylating agent, and in order to determine whether replacement of sulfur by phosphorus in myleran-type structures would produce compounds with antineoplastic activity, three groups of phosphorus-containing analogs have been synthesized in which the methanesulfonate group of myleran (I) has been replaced by phosphonate groups (II) and by disubstituted phosphinate (III) and phosphinite groups (IV).

$CH_3SO_2O(CH_2)_4OSO_2CH_3$
I

$R(C_2H_5O)P(O)O(CH_2)_4O(O)P(OC_2H_5)R$
IIa, R = CH_3
b, R = C_2H_5
c, R = n-C_4H_9
d, R = C_6H_5

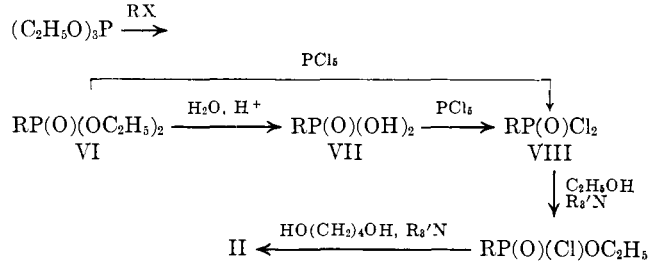
$R_2P(O)O(CH_2)_4O(O)PR_2$
IIIa, R = CH_3
b, R = C_2H_5
c, R = n-C_4H_9
d, R = C_6H_5

$R_2PO(CH_2)_4OPR_2$
IVa, R = n-C_4H_9
b, R = C_6H_5

$RP(O)(Cl)OC_2H_5$
V

The phosphonate (II), phosphinate (III), and phosphinite (IV) analogs were synthesized by reaction of 1,4-butanediol with the appropriate phosphorus acid chloride in the presence of a tertiary amine. The phosphonates (II) were obtained as shown in Scheme I. Alkylphosphonate esters (VI) were converted to phosphonic dichlorides (VIII) with or without prior conversion to phosphonic acids (VII); somewhat

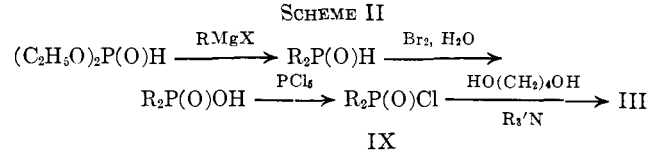
SCHEME I



higher yields of the chlorides (VIII) were generally obtained when the esters were hydrolyzed to the acids before treatment with phosphorus pentachloride.

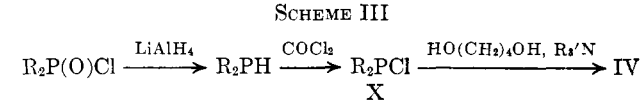
Scheme II outlines the reactions used to prepare the phosphinates (III). Dimethylphosphinic chloride

SCHEME II



(IX, R = CH_3) was prepared by the convenient method of Pollart and Harwood[2] from tetramethyl bi(phosphine sulfide). The phosphinites (IV) were prepared as shown in Scheme III. Diphenylphosphinous chloride (X, R = C_6H_5) was secured from commercial sources.

SCHEME III



(1) This investigation was supported by the Cancer Chemotherapy National Service Center, National Cancer Institute, National Institutes of Health, Contract No. PH-43-64-51, and by the C. F. Kettering Foundation.

(2) K. A. Pollart and H. J. Harwood, *J. Org. Chem.*, **27**, 4444 (1962).

Quin and Anderson[3] published their synthesis of tetramethylene bis(diphenylphosphinite) (IVb) and 1,4-dimethyltetramethylene bis(diphenylphosphinite) after the former compound had been prepared here, and Pudovik, *et al.*,[4] prepared diethyl tetramethylene bis(ethylphosphonate) (IIb) and a closely related derivative, dibutyl tetramethylene bis(butylphosphonate), by an Arbuzov reaction of the corresponding bis(phosphites) with the appropriate alkyl halide. Other related tetramethylene bis(phosphites),[4] bis(phosphates),[5] bis(phosphorothioates),[4] and bis(phosphonates)[6] have been described in the literature.

Myleran (I), functioning as an alkylating agent, reacts by an SN2 mechanism.

$$MeSO_3(CH_2)_4O_3SMe + 2HY \longrightarrow Y(CH_2)_4Y + 2MeSO_3^- + 2H^+$$

Chemically, the phosphorus analogs can react by two routes, undergoing either C–O or P–O bond cleavage. In biological systems, the esters might presumably serve as alkylating agents (1) upon rupture of the C–O bond. Alternatively, P–O bond rupture *in vivo* would produce groups which might be capable of serving as phosphorylating agents (2).

$$>P(O)O(CH_2)_4O(O)P< + 2HY \longrightarrow Y(CH_2)_4Y + 2>P(O)O^- + 2H^+ \quad (1)$$

$$>P(O)O(CH_2)_4O(O)P< + 2HY \longrightarrow 2>P(O)Y + HO(CH_2)_4OH \quad (2)$$

Strong alkylating properties have been reported for certain phosphorus esters such as diethyl phosphorofluoridate,[7] and there is evidence that triethyl phosphate will alkylate the thiol group of cysteine *in vivo*.[8] In addition, Lapidot, *et al.*,[9] have demonstrated that *t*-butyl phosphate undergoes both P–O and C–O bond fission at pH 7, the latter fission amounting to 23.5% of the combined P–O and C–O bond fission reaction.

Many phosphorus esters display a tendency to serve as phosphorylating agents. Dialkylphosphinites, alkyl- and arylphosphinates, dialkyl phosphonates, and phosphites have been used in the phosphorylation of mono- and polyhydric alcohols,[10] and more recently Wasserman and Cohen[11] have described phosphorylating activity displayed by 1-alkoxyvinyl phosphates. Because of this tendency, which appears to be general for phosphorus esters, it appears that the series of phosphonates, phosphinates, and phosphinites (II–IV) will similarly exhibit phosphorylating activity.

Compounds IId, IIIc, IIId, IVa, and IVb displayed cytotoxic activity at a concentration of 100 μg/ml against Eagle's KB cells; cell growth in these tests was 50% or less than the growth of the controls. All of the analogs have been screened in the Sarcoma 180 system, and the majority have been tested in the Walker 256, Adenocarcinoma 755, and leukemia L1210 systems. Compound IIc was active in the initial test in the Walker 256 system at 50 mg/kg per day, but the activity was not confirmed upon further testing. Compound IVb showed marginal activity in one test in the Adenocarcinoma 755 system. Screening results are listed in Table I.[12]

TABLE I

TUMOR AND CYTOTOXICITY TEST DATA[a]

Compd	Tumor	Dose, mg/kg per day	Mortality	T/C, %[b]	Cytotoxicity data Eagle's KB cell line (T − C_0)/(C − C_0)[c]
IIa	S180	20	1/6	72	0.95
	Ca755	16	0/10	64	
	Wa256	100	0/6	111	
	L1210	64	0/6	93	
IIb	S180	8	0/6	117	0.69
	Ca755	32	1/10	81	
	Wa256	2.5	0/6	98	
IIc	S180	10	1/6	87	0.58
	Ca755	8	0/10	114	
	Wa256	50	0/10, 6/6, 0/6	43, toxic, 90[d]	
	L1210	32	0/6	98	
IId	S180	125	0/6	80	0.24
	Ca755	100	1/10	95	
	Wa256	250	0/6	150	
	L1210	100	0/6	108	
IIIa	S180	250	0/6	95	0.85
	Ca755	100	0/10	80	
	Wa256	250	0/6	134	
	L1210	200	0/6	94	
IIIb	S180	62	1/6	111	1.00
	Ca755	50	0/10	99	
	Wa256	125	0/6	79	
	L1210	100	0/6	98	
IIIc	S180	500	0/6	123	0.13
	Ca755	400	0/10	78	
	Wa256	250	0/6	97	
	L1210	400	0/6	98	
IIId	S180	500	1/6	109	−0.16
	L1210	400	0/6	97	
IVa	S180	400	0/6	70	0.50
IVb	S180	500	0/6	99	−0.01
	Ca755	450	1/10	48	

[a] Test results for the mouse tumors were obtained in accordance with standard CCNSC protocol. For the Walker 256 system, only the highest nontoxic doses are listed. [b] T/C = treated/controls. T and C represent tumor weights for S180, Ca755, and Wa256 and survival time for L1210. [c] (T − C_0) and (C − C_0) represent micrograms of protein in treated and control systems in tests at a dose level of 100 μg/ml. [d] Compound IIc was toxic at 75 mg/kg per day. The variable results obtained at 50 mg/kg per day suggest that this dose is essentially the maximum tolerable dose and that the activity is only borderline.

Experimental Section

Melting points were determined with a Kofler Heizbank melting point apparatus and are corrected. Boiling points are uncorrected. Infrared spectra were determined with a Perkin-Elmer Model 221G or 521 spectrophotometer.

(3) L. D. Quin and H. G. Anderson, *J. Org. Chem.*, **29**, 1859 (1964).

(4) A. N. Pudovik, I. M. Aladzheva, I. A. Sokolova, and G. A. Kozlova, *Zh. Obshch. Khim.*, **33**, 102 (1963).

(5) T. M. Moshkina and A. N. Pudovik, *ibid.*, **32**, 1671 (1962).

(6) E. Cherbuliez, R. Prince, and J. Rabinowitz, *Helv. Chim. Acta*, **47**, 338 (1964).

(7) C. Stolzer and A. Simon, *Chem. Ber.*, **96**, 288 (1963).

(8) J. J. Roberts and G. P. Warrick, *Ann. Rep. Brit. Empire Cancer Campaigne*, **36**, 53 (1958); W. C. J. Ross, "Biological Alkylating Agents," Butterworth and Co. Ltd., London, 1962, p 8.

(9) A. Lapidot, D. Samuel, and M. Weiss-Broday, *J. Chem. Soc.*, 637 (1964).

(10) K. A. Petrov, E. E. Nifant'ev, and R. G. Gol'tsova, *Zh. Obshch. Khim.*, **32**, 3716 (1962); G. Kamai and E. T. Mukmenev, *ibid.*, **33**, 3197 (1963).

(11) H. Wasserman and D. Cohen, *J. Org. Chem.*, **29**, 1817 (1964).

(12) Tumor and tissue culture test data were obtained by the Chemotherapy Division of Southern Research Institute under the direction of Drs. F. M. Schabel, Jr., W. R. Laster, and G. J. Dixon.

TABLE II

BIS(PHOSPHONATES) (II)

$R(C_2H_5O)(O)PO(CH_2)_4OP(O)(OC_2H_5)R$

Compd	Bp, °C (mm)	Yield, %	Formula	C, % Calcd	C, % Found	H, % Calcd	H, % Found	P, % Calcd	P, % Found
IIa	141–145 (0.03)	47	$C_{10}H_{24}O_6P_2$	39.73	39.49	8.00	7.97	20.5	20.5
IIb	150–151 (0.02)[a]	56	$C_{12}H_{28}O_6P_2$	43.63	43.60	8.48	8.39	18.7	18.5
IIc	167 (0.5 μ)	70	$C_{16}H_{36}O_6P_2$	49.73	49.75	9.39	9.16	16.0	16.1
IId	200–205 (0.05)	82	$C_{20}H_{28}O_6P_2$	56.33	56.15	6.62	6.35	14.5	14.6

[a] Pudovik, *et al.*,[9] reported bp 173–174° at 3.5 mm.

TABLE III

BIS(PHOSPHINATES) (III)

$R_2(O)PO(CH_2)_4OP(O)R_2$

Compd	Mp, °C	Bp, °C (mm)	Yield, %	Formula	C, % Calcd	C, % Found	H, % Calcd	H, % Found	P, % Calcd	P, % Found
IIIa	77–78	...	48	$C_8H_{20}O_4P_2$	39.67	39.60	8.32	8.50	25.58	25.31
IIIb	...	172 (0.1)	53	$C_{12}H_{28}O_4P_2$	48.31	48.10	9.46	9.37	20.77	20.62
IIIc	...	219 (0.2)	74	$C_{20}H_{44}O_4P_2$	58.51	58.46	10.80	10.83	15.08	15.18
IIId	117–118	...	72	$C_{28}H_{28}O_4P_2$	68.56	68.70	5.75	5.87	12.6	12.9

TABLE IV

BIS(PHOSPHINITES) (IV)

$R_2PO(CH_2)_4OPR_2$

Compd	Mp, °C	Bp, °C (μ)	Yield, %	Formula	C, % Calcd	C, % Found	H, % Calcd	H, % Found	P, % Calcd	P, % Found
IVa	...	126–130 (1)	52	$C_{20}H_{44}O_2P_2$	63.46	63.19	11.72	11.47	16.37	16.23
IVb	73–75[a]	...	52	$C_{28}H_{28}O_2P_2$	73.35	73.10	6.16	6.15	13.51	13.70

[a] Lit.[8] mp 74–75°.

General Procedure.—Phosphonochloridates, phosphinic chlorides, and phosphinous chlorides (*ca.* 0.22 mole) in 400 ml of ether were cooled in an ice bath. A solution of 1,4-butanediol (0.1 mole) and triethylamine (0.2 mole) in 50 ml of ether was added dropwise with stirring in 3 hr. The mixture was stirred overnight at 0° to room temperature (cooling bath allowed to warm to room temperature) and filtered. After removal of the ether *in vacuo* (water pump), the residue was purified by distillation *in vacuo* or crystallization from benzene (for IIIa and IIId) or ether (for IVb). In the preparation of the bis(phosphinites), all operations were performed in an atmosphere of dry nitrogen. Analytical results, yields, and boiling points or melting points for II–IV are listed in Tables II–IV.

Alkylphosphonates were prepared from triethyl phosphite and an alkyl iodide by means of the Arbuzov reaction[13] and were converted to phosphonic dichlorides by reaction of the esters or the corresponding phosphonic acids with 2 molar equiv of PCl_5 in benzene.[13] By a procedure analogous to that described in the General Procedure, phosphonochloridates (V)[14] (see Table V) were prepared by addition of 1 molar equiv of ethanol and of a tertiary amine in ether solution to an ether solution of the phosphonic dichlorides at 0°. Diethyl-, dibutyl-, and diphenylphosphinic acids were prepared by the procedure of Kosolapoff and Struck[15] and were converted to phosphinic chlorides by reaction with 1 molar equiv of PCl_5 in benzene.[13] Tetramethyl bi(phosphine sulfide) was synthesized according to the method of Reinhardt, *et al.*,[16] and was converted to dimethylphosphinic chloride by the procedure of Pollart and Harwood.[2] Reduction of dibutylphosphinic chloride with $LiAlH_4$[17] produced dibutylphosphine which was chlorinated with phosgene as described by Henderson, *et al.*[18] Phenylphosphonic dichloride and diphenylphosphinous chloride were obtained from Aldrich Chemical Co.

TABLE V

PHOSPHONOCHLORIDATES (V)

RP(O)(Cl)OEt

R	Bp, °C (mm)	Yield, %	Cl, % Calcd	Cl, % Found
CH_3	37–40 (1)[a]	70	24.9	28.3
C_2H_5	38 (0.7)	70	22.7	24.0
n-C_4H_9	67–70 (1)	70	19.2	20.4
C_6H_5	...[b]	90	17.3	16.2

[a] Lit.[14b] bp 40–41° (1 mm). [b] Since distillation caused decomposition and polymerization, the crude product was used in the synthesis of IId.

Infrared Spectra.—P→O infrared absorption occurred in the 1240–1250-cm^{-1} range for the phosphonates (II) and in the 1255–1260-cm^{-1} range for the phosphonochloridates (V). P-O-C_2H_5 absorption appeared at 1155–1160 cm^{-1} in the phosphonates (II) and phosphonochloridates (V). P-O-C (aliphatic) absorption was generally broad and was observed as two peaks for most of the compounds (II–V) in the 950–1050-cm^{-1} range. P-CH_3 absorption occurred at 1315, 1310 and 1300, and 1305 cm^{-1}, respectively, in the spectra of IIa, IIIa, and Va; and P-C_6H_5 absorption was observed at 1440, 1440, and 1435 cm^{-1}, respectively, in the spectra of IId, IIId, and IVb. Absorption frequency assignments listed above are consistent with assignments given by Bellamy.[19] The alkyl phosphinates (III) showed two prominent bands in the P→O absorption region at 1195–1200 and 1240 cm^{-1}; for the methyl (IIIa) and ethyl (IIIb) derivatives, the 1200-cm^{-1} band was more intense than the 1240-cm^{-1} band, whereas the order of intensity was reversed for the butyl compound (IIIc). P→O absorption appeared at 1230 cm^{-1} for the phenyl phosphinate (IIId). Thomas and Chittenden[20] reported P→O absorption at 1181–1220 cm^{-1} for alkyl

(13) G. M. Kosolapoff, "Organophosphorus Compounds," John Wiley and Sons, Inc., New York, N. Y., 1950, pp 61, 121.

(14) (a) A. E. Arbuzov and A. I. Razumov, *Izv. Akad. Nauk SSSR, Otd. Khim. Nauk*, 167 (1945); (b) R. F. Hudson and L. Keay, *J. Chem. Soc.*, 2463 (1956).

(15) G. M. Kosolapoff and R. F. Struck, *ibid.*, 3950 (1959).

(16) H. Reinhardt, D. Bianchi, and D. Mölle, *Chem. Ber.*, **90**, 1656 (1957).

(17) L. Horner, H. Hoffmann, and P. Beck, *ibid.*, **91**, 1583 (1958).

(18) W. A. Henderson, Jr., S. A. Buckler, N. E. Day, and M. Grayson, *J. Org. Chem.*, **26**, 4770 (1961).

(19) L. J. Bellamy, "The Infrared Spectra of Complex Molecules," 2nd ed, John Wiley and Sons, Inc., New York, N. Y., 1958, Chapter 18.

(20) L. C. Thomas and R. A. Chittenden, *Spectrochim. Acta*, **20**, 467 (1964).

dialkylphosphinates and 1230–1238 cm^{-1} for phosphinates of the type ROP(O)(R′)(R″) in which R, R′, R″ = mixed alkyl and aryl.

Acknowledgment.—The author wishes to express his appreciation to Drs. Y. F. Shealy and J. A. Montgomery for encouragement in this work, and to the members of the Analytical Section of Southern Research Institute who performed the spectral and most of the microanalytical determinations reported under the direction of Dr. W. J. Barrett. Some of the analyses reported were performed by Galbraith Laboratories, Knoxville, Tenn.

Notes

Nucleosides from Homoribose[1]

JOHN A. MONTGOMERY AND KATHLEEN HEWSON

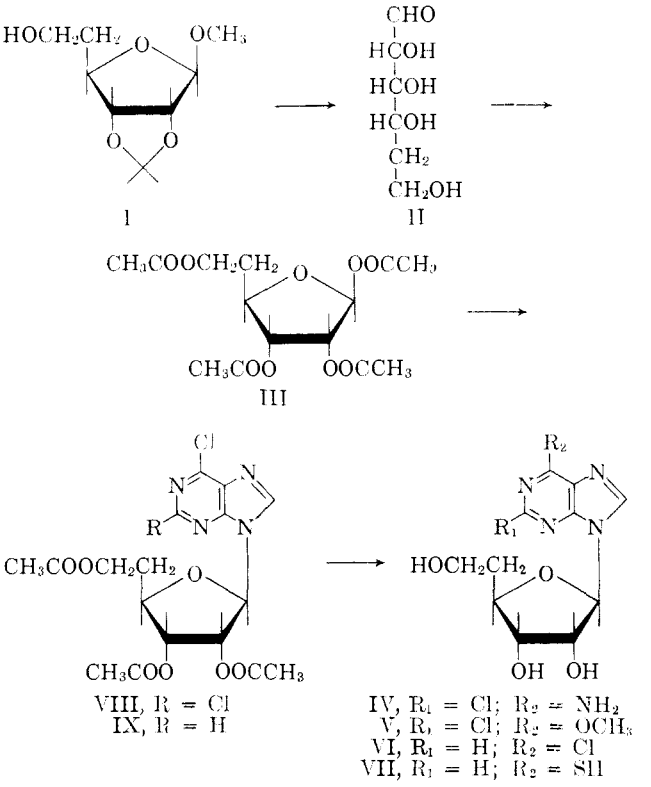
Kettering-Meyer Laboratory, Southern Research Institute, Birmingham, Alabama

Received August 2, 1965

In a previous paper[2] we described the synthesis of homoribose (5-deoxy-D-*ribo*-hexose) from methyl 2,3-*O*-isopropylidene-β-D-ribofuranoside and its proof of structure by proton magnetic resonance spectroscopy. We have now prepared some purine nucleosides from homoribose.[3]

Methyl 5-deoxy-2,3-*O*-isopropylidene-β-D-*ribo*-hexofuranoside (I)[2] was hydrolyzed in a mixture of dilute hydrochloric acid and ethanol, but concentration of the reaction mixture caused the resulting 5-deoxy-D-*ribo*-hexose (II) to condense with itself. Treatment of this material with acetic anhydride resulted in a low yield of impure tetra-*O*-acetyl-5-deoxy-D-*ribo*-hexose (III). Neutralization of the acid hydrolysis media with ion-exchange resin before concentration did not prevent self-condensation, but the use of dilute sulfuric acid followed by neutralization with barium hydroxide and then freeze-drying gave a high yield of II, which was readily converted to the tetraacetate III, a light yellow oil (see Scheme I). The β-configuration was assigned to III on the basis of the comparison of its proton magnetic resonance spectrum with that of tetra-*O*-acetyl-β-D-ribofuranose.[5] Furthermore the fact that the absorption due to the proton at C-1 appears as a singlet ($J \leq 1$ cps)[6] also indicates the β-configuration.[6] Compound III was allowed to react with 2,6-dichloropurine by the fusion technique using *p*-toluenesulfonic acid as catalyst.[7] From this reaction a 34% yield of 9-(2,3,6-tri-*O*-acetyl-5-deoxy-β-D-*ribo*-hexofuranosyl)-2,6-dichloropurine (VIII) was isolated as a crystalline solid. The β-configuration was assigned to this nucleoside on the basis of the comparison of its proton magnetic resonance spectrum with that of 9-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)-2,6-dichloropurine (X).[8] The striking similarity of the

SCHEME I



spectra of these two nucleosides (See Figure 1) provides the best available evidence for this anomeric assignment. Although attempts have been made to relate the coupling constant for the proton at C′-1 ($J_{1'2'}$) of purine nucleosides to the anomeric configuration at C′-1 by use of the Karplus equation,[9] these attempts have unfortunately been entirely unsuccessful.[10–12] Lemieux and Lineback[13] have pointed out that in a furanose ring the coupling constant for vicinal *cis* protons can vary from 3.5 to 8.0 cps and for vicinal *trans* protons from 0 to 8.0 cps. Consequently, the coupling constant $J_{1'2'}$ for VIII, 5.0 cps, provides no definitive information concerning its anomeric configuration; however, from our investigation of the proton magnetic resonance spectra of a number of 9-β-ribofuranosylpurines we have found the chemical shift of the C′-1 hydrogen to be character-

(1) This work was supported by the C. F. Kettering Foundation and by the Cancer Chemotherapy National Service Center, National Cancer Institute, National Institutes of Health, Contract No. PH-43-64-51.

(2) J. A. Montgomery and K. Hewson, *J. Org. Chem.*, **29**, 3436 (1964).

(3) Ryan, *et al.*,[4] have described the synthesis of homoribose and homoadenosine by a different route.

(4) K. J. Ryan, H. Arzoumanian, E. M. Acton, and L. Goodman, *J. Am. Chem. Soc.*, **86**, 2503 (1964).

(5) H. Zinner, *Ber.*, **83** 153 (1950); **86**, 817 (1953).

(6) K. L. Rinehart, Jr., W. S. Chilton, M. Hicheus, and W. von Phillipsborn, *J. Am. Chem. Soc.*, **84**, 3216 (1962).

(7) T. Sato, T. Shimadate, and Y. Ishido, *Nippon Kagaku Zasshi*, **81**, 1440 (1960).

(8) J. A. Montgomery and K. Hewson, *J. Heterocyclic Chem.*, **1**, 213 (1964).

(9) M. Karplus, *J. Chem. Phys.*, **30**, 11 (1959).

(10) L. Goldman and J. W. Marsico, *J. Med. Chem.*, **6**, 413 (1963).

(11) N. J. Leonard and R. A. Laursen, *J. Am. Chem. Soc.*, **85**, 2026 (1963).

(12) J. A. Montgomery and H. J. Thomas, *ibid.*, **87**, 5442 (1965).

(13) R. U. Lemieux and D. R. Lineback, *Ann. Rev. Biochem.*, **32**, 155 (1963).