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Studies on Chemotherapeutic Agents. V.[1,2] A Synthesis of Benzimidazole Nucleosides of D-Glucopyranuronamide

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Direct condensation of D-glucuronolactone with *o*-nitroaniline (Ia) or 5-nitro-*o*-4-xylidine (Ib) afforded *o*-nitroanilino- or 5-nitro-*o*-4-xylidino-D-glucuronolactone, IIa or IIb in an excellent yield. On treatment with ammoniacal MeOH, IIa and IIb were amidated to give VIIa and VIIb, respectively. It was found that they held a furanose structure. VIIa and VIIb underwent conversion of the lactol ring from furanoside to pyranoside form by the presence of AcOH, affording *o*-nitroanilino- and 5-nitro-*o*-4-xylidino-D-glucopyranuronamide, XIa and XIb. Hydrogenation of the nitro group in the acetylated derivative XIIa or XIIb was effectively made over a palladium or platinum catalyst. Ring closure of the phenylenediamine derivative XIIIa or XIIIb with ethyl orthoformate or ethyl formimidate hydrochloride followed by acid or alkali hydrolysis gave 1-deoxy-1-(1-benzimidazolyl)- or 1-deoxy-1-(5,6-dimethyl-1-benzimidazolyl)-β-D-glucopyranuronamide, XVa or XVb. The corresponding benzimidazole nucleosides of D-glucuronolactone and D-glucofuranuronamide could not be obtained by the same ring closure procedure. XVa, XVb and 1-deoxy-1-(5,6-dichloro-1-benzimidazolyl)-β-D-glucopyranuronamide (XVc) were also formed by the chloromercury method of Davoll and Brown.

Many nucleoside-type antibiotics have been isolated and extensively investigated, some of them having been proved to be of chemotherapeutic value.

Recently, three antibiotic nucleosides, gougerotin,[4] blasticidin S,[5] and polyoxin complex[6] which contain hexouronic acid derivatives as the sugar component have been isolated. The finding of these nucleosides led the authors to a study of the pyrimidine and purine nucleosides of hexouronic acid derivatives. In the previous papers,[7] the authors described the syntheses of pyrimidine and purine nucleosides of D-glucuronic acid, which is an indispensable component of a higher animal. This paper deals with a synthesis of benzimidazole nucleosides of D-glucopyranuronamide.

As is well known, 5,6-dimethyl-1-(α-D-ribofuranosyl)benzimidazole is a nucleoside moiety in vitamin B_{12} and the importance of the vitamin as a cofactor in many biochemical reaction have been well established.[8]

1) A part of this paper was read at the 88th Annual Meeting of Pharmaceutical Society of Japan in Tokyo, April 5, 1968.
2) Part IV: T. Kishikawa, K. Furuta, and S. Takitani, *Tetrahedron Letters*, **1969**, 1961.
3) Location: *Ichigayafunagawara-machi, Shinjuku-ku, Tokyo.*
4) T. Kanzaki, *J. Antibiotics* (Tokyo), Ser. A, **15**, 93 (1962): H. Iwasaki, *Yakugaku Zasshi*, **82**, 1358 (1962); J.J. Fox, Y. Kuwada, K.A. Watanabe, T. Ueda, and E.B. Whipple, *Antimicrobial Agents and Chemotherapy*, **1964**, 518; J.J. Fox, Y. Kuwada, and K.A. Watanabe, *Tetrahedron Letters*, **1968**, 6029.
5) N. Otake, S. Takeuchi, T. Endo, H. Yonehara, *Tetrahedron Letters*, **1965**, 1405, 1411; J.J. Fox, K.A. Watanabe, *Tetrahedron Letters*, **1966**, 897.
6) K. Isono, S. Suzuki, *Tetrahedron Letters*, **1968**, 203, 1133.
7) T. Kishikawa, and H. Yuki, *Chem. Pharm. Bull.* (Tokyo), **12**, 1259 (1964); *ibid.*, **14**, 1354, 1360 (1966).
8) J.M. Buchanan, H.L. Elford, R.E. Loughlin, B.M. McDougall, and S. Rosenthal, *Ann. N. Y. Acad. Sci.*, **112**, 756 (1964); K. Bernhauer, O. Muller, and F. Wagner, *Angew. Chem. Intern. Ed Engl.*, **3**, 200 (1964); H. Weissbach and H. Dickerman, *Physiol. Rev.*, **45**, 80 (1965).

On the other hand, some benzimidazole nucleosides, including 5,6-dichloro-1-(β-D-ribofuranosyl)benzimidazole have been shown to exhibit antiviral activity.[9] Lately, this compound has been proved to exhibit specific inhibition of chromosomal RNA synthesis.[10] Many synthetic methods[11] for benzimidazole nucleosides have been presented, since the finding of the 5,6-dimethyl-benzimidazole nucleoside in 1950.

The authors undertook the synthesis of both the pyranose and furanose forms of the benzimidazole nucleosides containing D-glucuronamide as the sugar component (Chart 1).

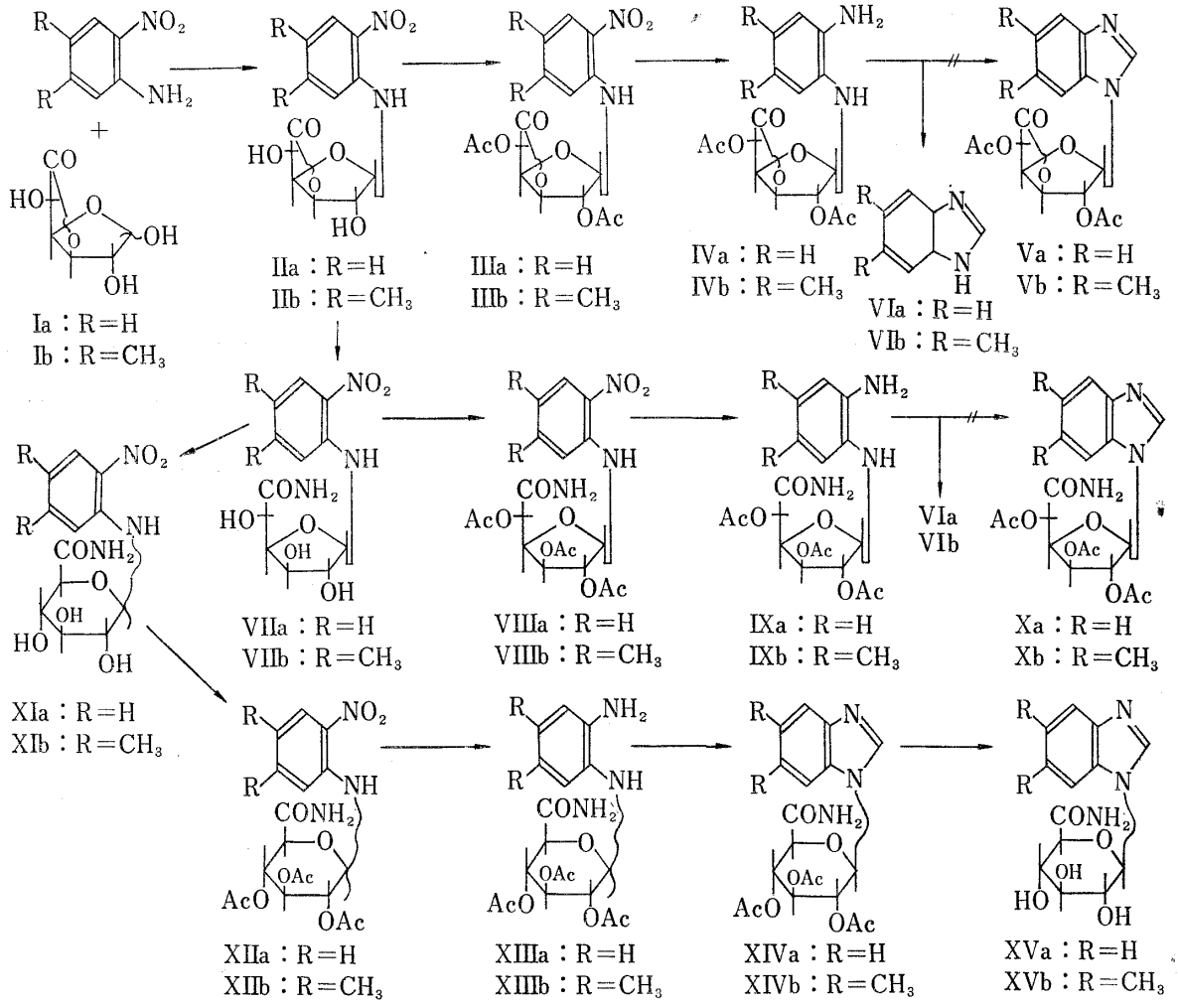

Chart 1

9) I. Tamm, K. Folkers, and C.H. Shunk, *J. Bacteriol.*, **72**, 54 (1956); I. Tamm, *Yale J. Biol. Med.*, **29**, 33 (1956).

10) M.L. Birnstein, J.J. Sirlin, and J. Jacob, *Biochem. J.*, **94**, 10 (1965).

11) *a*) N.G. Brink, F.W. Holly, C.H. Shunk, E.W. Peel, J.J. Cahill, and K. Folkers, *J. Am. Chem. Soc.*, **72**, 1866 (1950); F.W. Holly, C.H. Shunk, E.W. Peel, J.J. Cahill, J.B. Lavigne, and K. Folkers, *ibid.*, **74**, 4521 (1952); P. Mamalis, V. Petrow, and S. Sturgeon, *J. Pharm. Pharmacol.*, **2**, 491, 503, 512 (1950); *b*) G. Cooley, B. Ellis, P. Mamalis, V. Petrow and B. Sturgeon, *ibid.*, **2**, 579 (1950); J.G. Buchanan, A.W. Johnson, J.A. Mill, and A.R. Todd, *J. Chem. Soc.*, **1950**, 2845; D. Heyl, E.C. Chase, C.H. Shunk, M.U. Moore, G.A. Emerson, and K. Folkers, *J. Am. Chem. Soc.*, **76**, 1355 (1954); *c*) F. Weygand, A. Wacker, and F. Wirth, *Z. Naturforsch.*, **66**, 25 (1951); F. Weygand and F. Wirth, *Chem. Ber.*, **85**, 1000 (1952); A.J. Cleaver, A.B. Foster, and W.G. Overend, *J. Chem. Soc.*, **1959**, 409; *d*) A.W. Johnson, G.W. Miller, J.A. Mills, and A.R. Todd, *J. Chem. Soc.*, **1953**, 3061; *e*) J. Davoll and G.B. Brown, *J. Am. Chem. Soc.*, **73**, 5781 (1951); *f*) H. Bräuniger and A. Koine, *Arch. Pharm.*, **296**, 668 (1963); *ibid.*, **298**, 641 (1965); *g*) C.P. Whittle and R.K. Robins, *J. Am. Chem. Soc.*, **87**, 4940 (1965).

The glycosidation of *o*-nitroaniline (Ia) or 5-nitro-*o*-4-xylidine (Ib) by boiling with D-glucurono-6,3-lactone in methanol or in aqueous ethanol was effectively made, and 1-deoxy-1-(*o*-nitroanilino)-D-glucurono-6,3-lactone (IIa) and 1-deoxy-1-(5-nitro-*o*-4-xylidino)-D-glucurono-6,3-lactone (IIb) were obtained in the yields of 72 and 45%, respectively.

When the compound IIa or IIb was treated with a mixture of methanol and concentrated ammonium hydroxide at room temperature, bitter orange crystals melting at 169—170° (decomp.), $[\alpha]_D^{24}$ +102.1° or at 158—159° (decomp.), $[\alpha]_D^{24}$ +89.6° were obtained after recrystallization from methanol. Compound VIIa and VIIb thus obtained exhibited infrared absorption of type A, B, C and D[12] at 935 and 933, 878 and 875, 870 and 854 and, 798 and 806 cm^{-1}, respectively (Fig. 1 and 2), which are characteristic of a furanose ring.

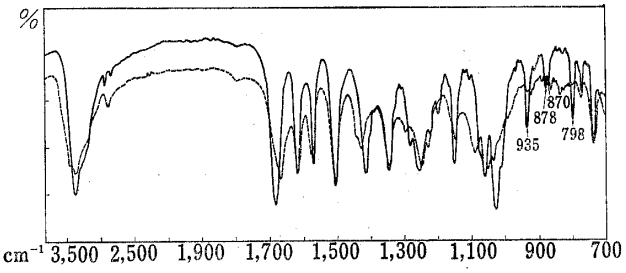

Fig. 1. Infrared Absorption Spectra of 1-Deoxy-1-(*o*-nitroanilino)-D-glucuronamide (KBr)

——: furanose VIIa ------: pyranose XIa

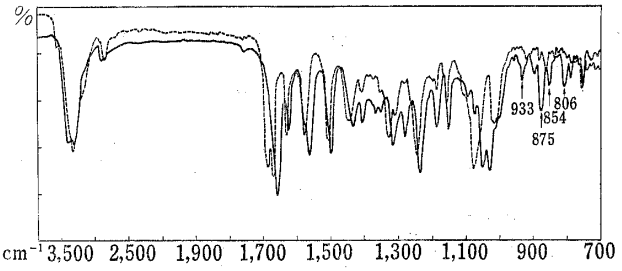

Fig. 2. Infrared Absorption Spectra of 1-Deoxy-1-(*o*-4-xylidino)-D-glucuronamide (KBr)

——: furanose VIIb ------: pyranose XIb

The nuclear magnetic resonance (NMR) spectral data of the acetylated compound VIIIa and VIIIb (Table I) are also consistent with the furanose structure.[13] Furthermore, this furanoside character of the lactol ring in compound VIIa and VIIb was unequivocally clarified

Table I. The 100 MHZ NMR Spectral data for the Ring Protons in the Sugar Moiety of *o*-nitroanilino- and 5-nitro-*o*-4-xylidino-derivatives of D-Glucuronolactone and D-Glucofuranuronamide (in $DMSOd_6$)

IIIa		IIIb	
H 1′δ 5.99 Q	$J_{NH,1'}$=8.5 HZ $J_{1',2'}$=4.6 HZ	H 1′δ 5.98 Q	$J_{NH,1'}$=8.6 HZ $J_{1',2'}$=4.6 HZ
H 2′δ 5.50 D		H 2′δ 5.46 D	
H 3′δ 5.27 D		H 3′δ 5.24 D	
H 4′δ 5.01 Q	($J_{3',4'}$=4.0 HZ)	H 4′δ 5.00 Q	($J_{3',4'}$=4.2 HZ)
H 5′δ 5.72 D	($J_{4',5'}$=5.1 HZ)	H 5′δ 5.67 D	($J_{4',5'}$=5.2 HZ)

VIIIa		VIIIb	
H 1′δ 5.90 Q	$J_{NH,1'}$=8.5 HZ $J_{1',2'}$=5.0 HZ	H 1′δ 5.90 Q	$J_{NH,1'}$=8.2 HZ $J_{1',2'}$=4.8 HZ
H 2′δ 5.54 Q	($J_{2',3'}$=2.5 HZ)	H 2′δ 5.53 Q	($J_{2',3'}$=2.1 HZ)
H 3′δ 5.42 Q	($J_{3',4'}$=4.4 HZ)	H 3′δ 5.48 Q	($J_{3',4'}$=4.3 HZ)
H 4′δ 4.56 Q	($J_{4',5'}$=8.8 HZ)	H 4′δ 4.53 Q	($J_{4',5'}$=8.7 HZ)
H 5′δ 5.07 D		H 5′δ 5.03 D	

Chemical shifts (δ) were measured in ppm from internal tetramethylsilane.
D: Doublet; Q: Quartet

12) S.A. Barker, R. Stephens, *J. Chem. Soc.*, **1954**, 4550.
13) *a*) R.J. Abraham, K.A. McLauchlan, *Chem. Ind.* (London), **1962**, 213; R.J. Abraham, L.D. Hall, L. Hough, K.A. McLauchlan, *J. Chem. Soc.*, **1962**, 3699. *b*) G. Huber, A. Rossi, *Helv. Chim. Acta*, **51**, 1185 (1968).

by obtaining 2,3,5-tri-O-acetyl-D-glucofuranuronamide (XVI)[14] by acid hydrolysis of VIIIa or VIIIb (Chart 2).

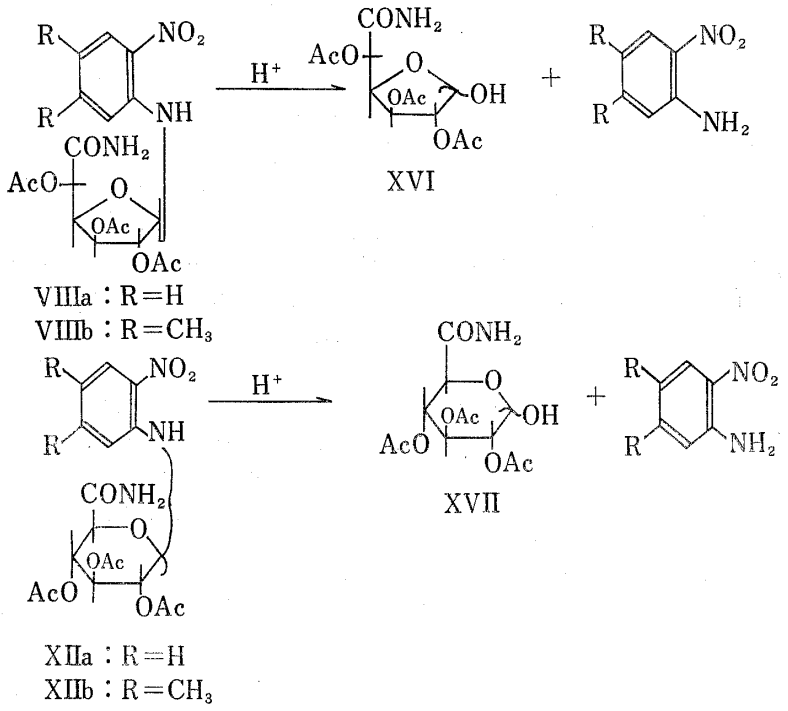

Chart 2

It should be noted that the furanose structure in compound IIa and IIb survived ammoniacal treatment without transformating into the pyranose structure. For it has been observed[14,15] that 1-deoxy-1-anilino-(or *p*-nitroanilino)-D-glucuronolactone is easily amidated by treatment with ammoniacal methanol, always, being accompanied with conversion of the lactol ring from furanoside to pyranoside form.

Compound VIIa and VIIb were comparatively stable and were not easily subject to the ring transformation in an aqueous solution. However, they underwent ring expansion from furanoside to pyranoside form by dissolving in warm methanol solution containing glacial acetic acid. After complete removal of the methanol and acetic acid, yellow needles melting at 176—177° (decomp.), $[\alpha]_D^{24}$ −126.3° and at 173—174° (decomp.), $[\alpha]_D^{24}$ 0° were obtained. Infrared absorption spectra of XIa and XIb (Fig. 1) differed from D-glucofuranuronamide derivative VIIa and VIIb, and the product XVII obtained by acid hydrolysis of XIIa or XIIb (Chart 2), was indistinguishable with 2,3,4-tri-O-acetyl-D-glucopyranuronamide,[16] which had been prepared by treatment of 1-bromo-1-deoxy-2,3,4-tri-O-acetyl-α-D-glucopyranuronamide with silver carbonate in aqueous acetone. Therefore it was established that the compound XIa and XIb had a pyranose structure.

Acetylation of compound IIa, IIb, VIIa, VIIb, XIa or XIb with a mixture of pyridine and acetic anhydride was carried out in a yield of 80—90%.

Hydrogenation of the nitro group in the acetylated derivative IIIa, IIIb, VIIIa, VIIIb, XIIa or XIIb was achieved over a palladized charcoal or platinum catalyst at atmospheric pressure, and the phenylenediamine derivatives IVa, IVb, IXa, IXb, XIIIa, and XIIIb were obtained as colorless needles or a pale yellow glass after evaporation of the solvent.

14) Y. Nakajima, *Yakugaku Zasshi*, **81**, 1094 (1961).
15) *a*) S. Takitani, *Chem. Pharm. Bull.* (Tokyo), **9**, 222 (1961). *b*) I. Ide, *Yakugaku Zasshi*, **85**, 213 (1965).

Attempts to cyclise IVa, IVb, IXa and IXb to their corresponding benzimidazole nucleoside Va, Vb, Xa and Xb with a various ring closing agents were unsuccessful, presumably due to the very ease of hydrolysis at the glycosidic linkage of the phenylenediamine derivatives. In these cases of ring closures, the free bases, benzimidazole (VIa) and 5,6-dimethylbenzimidazole (VIb) were produced in reasonable yields.

However, treatment of compound XIIIa or XIIIb with ethyl formimidate hydrochloride or with ethyl orthoformate followed by heating with 0.05N hydrochloric acid afforded the protected benzimidazole nucleoside XIVa or XIVb. XIVa or XIVb was then hydrolyzed with ethanolic ammonia or hydrochloric acid to give 1-deoxy-1-(1-benzimidazolyl)-β-D-glucopyranuronamide (XVa), mp 192—193° (decomp.), $[\alpha]_D^{24}$ +21.3° or 1-deoxy-1-(5,6-dimethyl-1-benzimidazolyl)-β-D-glucopyranuronamide (XVb), mp 263—264° (decomp.), $[\alpha]_D^{24}$ +15.5°. The overall yields of XVa and XVb were about 16 and 20%, calculated from XIIa and XIIb.

Though the glycosidation reaction of *o*-nitroaniline or 5-nitro-*o*-4-xylidine involves the anomeric center of D-glucuronolactone, one isomer is formed to the complete exclusion of the other. IIa and IIb, and their acetyl derivatives, IIIa and IIIb all have high specific rotation, varying from +120° to +260° and hence may be assumed to have α-configuration. Similarly, the amidated compound VIIa, VIIb, VIIIa and VIIIb which were obtained by treatment of IIa and IIb with ammonium hydroxide, and by the subsequent acetylation are tentatively assigned to α-configuration.

Recently, the NMR spectra of the D-glucuronolactone[17] and D-glucofuranose derivatives[13] have been reported. The anomeric proton signal has appeared as a doublet in the α-anomer compounds, and as a singlet in the β-anomer compounds. The NMR spectral data of D-glucuronolactone and D-glucofuranuronamide derivatives mentioned above are listed in Table 1 and $J_{1,2}$ of 4.6—5.0 Hz supports the α-configuration of the anomeric carbon atom. This assignment based on the NMR data coincides with that deduced from optical rotation.

On the other hand, the configuration at the anomeric center in compound XIa and XIb which were established to have a pyranoside structure could not be argued on the basis of optical rotation because they showed $[\alpha]_D^{24}$ −126° and 0.° The NMR spectrum of the acetylated derivative XIIb was not sufficiently resolved to allow the assignment of the anomeric carbon atom, however, compound XIIa in $CDCl_3$ gave two anomeric proton triplets at 5.76 and 5.54 (ppm) with J=4.3 and 8.7 Hz, which were identified utilizing the double-resonance experiment. The two triplet patterns for the anomeric proton in XIIa might arise from the coupling of H-1′$_{ax}$ and H-1′$_{eq}$, with H-2′ and secondary –NH proton at C-1. Irradiation of the –NH proton signals located at δ=8.55 (N–H$_{ax}$) and 8.46 (–NH$_{eq}$) caused a collapse of each of the triplets at 5.76 and 5.54 (ppm) to a corresponding doublet with $J_{H-1',H-2'}$=4.3 and 8.7 Hz. Since the C 1 conformation for (XIIa) should be most probable, the width of these two doublets shows an axial–equatorial and an axial–axial orientation[18] of the $C_{1'}$ and $C_{2'}$ protons, and thus axial (α-) and equatorial (β-) arrangement of the 1-substituent. The intesity ratio indicate that (XIIa) is approximately 1:2 mixture of α- and β-anomer.

Lately, Ide has reported[15] that *p*-nitroanilino-D-glucopyranuronamide obtained by treatment of *p*-nitroanilino-D-glucuronolactone with methanolic ammonia has α-configuration, while the direct condensation product of D-glucopyranuronamide and *p*-nitroaniline has β-configuration.

The author's attempts to codense D-glucopyranuronamide directly with *o*-nitroaniline (Ia) or 5-nitro-*o*-4-xylidine (Ib) were unsuccessful. Assignment of the anomeric configuration to the benzimidazole nucleosides of D-glucopyranuronamide, XVa and XVb was made on the basis of NMR spectra.[19] XVa and XVb gave an anomeric proton doublet at δ=5.55 and

16) Y. Nitta, J. Ide, A. Momose, Y. Nakajima, *Yakugaku Zasshi*, **82**, 578 (1962).
17) A. Momose, K. Kamei, Y. Nitta, *Chem. Pharm. Bull.* (Tokyo), **14**, 199 (1966).
18) R. Lemieux, R. Kullnig, H. Bernstein, G. Schneider, *J. Am. Chem. Soc.*, **80**, 6098 (1958).
19) The spectra of XVa and XVb were determined in $DMSOd_6$ with tetramethylsilane as internal reference.

5.48 (ppm) with J=8.8 and 8.7 Hz, which proved the di-axial arrangement[18] of the $C_{1'}$ and $C_{2'}$ protons. Thus, the β-configuration at the glycosidic linkage in these compounds was confirmed.

Compound XVa, XVb and 5,6-dichlorobenzimidazole derivatives (XVc) could also be formed from acetobromoglucuronic acid (Chart 3) by application of the chloromercury proce-

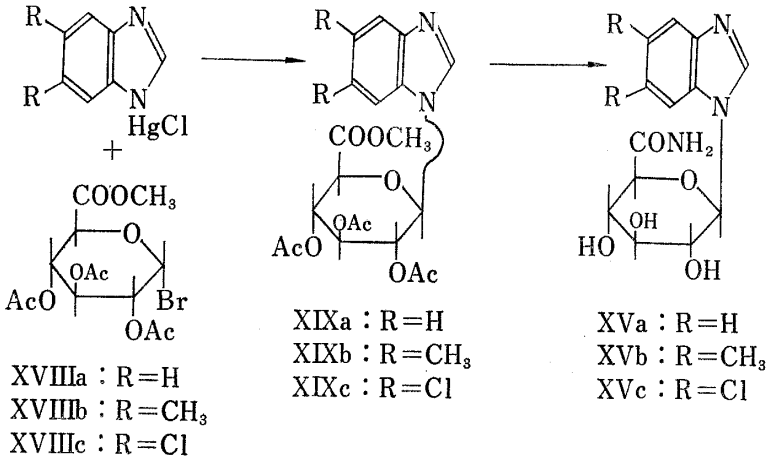

Chart 3

dure of Davoll and Brown.[11e] Condensation of methyl 1-deoxy-1-bromo-(2,3,4-tri-O-acetyl)-β-D-glucopyranuronate[20] with a chloromercuribenzimidazole, XVIIIa, XVIIIb and XVIIIc[11e] in boiling xylene followed by deacetylation and amidation with ammoniacal methanol afforded XVa, XVb and XVc in the yields of 31.8, 23.5 and 41.2%, calculated from XVIIIa, XVIIIb and XVIIIc, respectively.

Experimental[21]

1-Deoxy-1-(*o*-nitroanilino)-*α*-D-glucofuranurono-6,3-lactone (IIa)——A solution of D-glucuronolactone (10 g) and *o*-nitroaniline (Ia, 10 g) in a mixture of EtOH (30 ml) and H_2O (20 ml) was heated under reflux on a water bath for 6 hr. The solvent was completely removed under reduced pressure to leave a residual solid, which on recrystallization from MeOH afforded 11.2 g (71.9 g) of orange pillars, melting at 153—154°. $[\alpha]_D^{22}$ +142.7° (c=3.00, dioxane). *Anal.* Calcd. for $C_{12}H_{12}O_7N_2$: C, 48.61; H, 4.05; N, 9.45. Found: C, 48.90; H, 4.25; N, 9.29.

1-Deoxy-1-(5-nitro-*o*-4-xylidino)-*α*-D-glucofuranurono-6,3-lactone (IIb)——A solution of 5-nitro-*o*-4-xylidine (Ib, 15 g) and D-glucuronolactone (15 g) in MeOH (300 ml) was heated under reflux on a water bath for 5 hr, and then allowed to stand at room temperature. Orange needles, mp 143—144° separated. Recrystallization from MeOH raised the melting point to 145—146°. Yield, 14 g (45.2%). $[\alpha]_D^{22}$ +119.2° (c=3.00, dioxane). *Anal.* Calcd. for $C_{14}H_{16}O_7N_2$: C, 51.85; H, 4.93; N, 8.64. Found: C, 51.63; H, 4.96; N, 8.64.

1-Deoxy-1-(*o*-nitroanilino)2,5-di-O-acetyl-*α*-D-glucofuranurono-6,3-lactone (IIIa)——13 g of (IIa) was dissolved in a mixture of pyridine (20 ml) and Ac_2O (60 ml) previously cooled to 0° and the solution was allowed to stand at 0° for 2 hr, and then poured into crushed ice. A yellow precipitate formed was filtered off and recrystallized from AcOEt. Yellow plates, melting at 172—173° were obtained in a yield of 83.8% (14 g). $[\alpha]_D^{23}$ +253.1° (c=1.54, $CHCl_3$). *Anal.* Cacld. for $C_{16}H_{16}O_9N_2$: C, 50.50; H, 4.21; N, 7.36. Found: C, 49.90; H, 4.41; N, 7.55.

1-Deoxy-1-(5-nitro-*o*-4-xylidino)-2,5-di-O-acetyl-*α*-D-glucofuranurono-6,3-lactone (IIIb)——Essentially the same procedure as described for the preparation of IIIa was used. Yellow needles (from AcOEt), mp 192—193°. $[\alpha]_D^{23}$ +260.3° (c=1.52, $CHCl_3$). *Anal.* Calcd. for $C_{18}H_{20}O_9N_2$: C, 52.94; H, 4.90; N, 6.86. Found: C, 52.63; H, 4.80; N, 6.63.

1-Deoxy-1-(*o*-nitroanilino)-*α*-D-glucofuranuronamide (VIIa)——To a stirred suspension of (IIa, 10 g) in MeOH (100 ml) was dropwise added 25 ml of concentrated ammonium hydroxide under ice-water cooling. The suspension went into a clear solution and then immediately orange precipitate separated. The stirring

20) G.N. Bollenbach, J.W. Long, D.G. Benjamin, and J.A. Lindquist, *J. Am. Chem. Soc.*, **77**, 3310 (1955).
21) All melting points were not corrected.

was continued for additional 2 hr. After cooling, the precipitate was filtered off, washed with MeOH and then dried. Yield, 8.7 g (84.5%). mp 157—161° (decomp.). Recrystallization from MeOH raised the melting point to 169—170° (decomp.). $[\alpha]_D^{24}$ +102.1° (c=1.56, DMF). *Anal.* Calcd. for $C_{12}H_{15}O_7N_3$: C, 46.01; H, 4.83; N, 13.42. Found: C, 46.34; H, 4.93; N, 13.40.

1-Deoxy-1-(5-nitro-*o*-4-xylidino)-α-D-glucofuranuronamide (VIIb)——A stirred suspension of (IIb, 2 g) in MeOH (20 ml) was treated with concentrated ammonium hydroxide as mentioned above. Recrystallization from MeOH gave 1.5 g of orange needles, mp 158—159° (decomp.). $[\alpha]_D^{24}$ +89.6° (c=1.50, DMF). *Anal.* Calcd. for $C_{14}H_{19}O_7N_3$: C, 49.26; H, 5.57; N, 12.31. Found: C, 49.10; H, 5.35; N, 12.30.

1-Deoxy-1-(*o*-nitroanilino)-2,3,5-tri-O-acetyl-α-D-glucofuranuronamide (VIIIa)——12 g of VIIa was dissolved in a mixed solution of pyridine (65 ml) and Ac_2O (65 ml) pre-cooled to 0°. The solution was left to stand at 5° for 1 hr and then poured into ice water. The separated oil (crystallized in part) was solidified by addition of a small volume of EtOH. Recrystallization from EtOH gave 15 g of yellow needles melting at 172—173°. $[\alpha]_D^{20}$ +183.6° (c=2.91, $CHCl_3$). *Anal.* Calcd. for $C_{18}H_{21}O_{10}N_3$: C, 49.20; H, 4.78; N, 9.57. Found: C, 48.98; H, 4.77; N, 9.56.

1-Deoxy-1-(5-nitro-*o*-4-xylidino)-2,3,5-tri-O-acetyl-α-D-glucofuranuronamide (VIIIb)——Essentially the same procedure as described for the preparation of (VIIa) was used. The precipitated product was recrystallized from MeOH in yellow needles. mp 213—214°. $[\alpha]_D^{20}$ +169.7° (c=1.50, $CHCl_3$). *Anal.* Calcd. for $C_{20}H_{25}O_{10}N_3$: C, 51.39; H, 5.35; N, 8.99. Found: C, 51.20; H, 3.03; N, 8.88.

1-Deoxy-1-(*o*-nitroanilino)-D-glucopyranuronamide (XIa)——5 g of VIIa was dissolved in a warmed solution of MeOH (200 ml) containing 10 ml of glacial AcOH and the solution was left to stand at room temperature overnight. After evaporation of the solvent, the remaining acid was removed *in vacuo* by distillation with EtOH 3—5 times to give a syrup, which on recrystallization from MeOH yielded 3.1 g (62%) of yellow needles. mp 176—177° (decomp.). $[\alpha]_D^{20}$ −126.3° (c=1.52, DMF). *Anal.* Calcd. for $C_{12}H_{15}O_7N_3$: C, 46.01; H, 4.83; N, 13.42. Found: C, 45.93; H, 4.50; N, 13.31.

1-Deoxy-1-(5-nitro-*o*-4-xylidino)-D-glucopyranuronamide (XIb)——2.6 g of VIIb was dissolved in a warmed solution of MeOH (80 ml) containing glacial AcOH (8 ml) and the solution was left to stand at room temperature for 3 dyas. After complete removal of the solvent and AcOH, the residue was recrystallized from MeOH in fine yellow needles. Yield, 1.1 g (42.3%). mp 177—178° (decomp.). $[\alpha]_D^{24}$ 0° (c=1.55, DMF). *Anal.* Calcd. for $C_{14}H_{19}O_7N_3$: C, 49.26; H, 5.57; N, 12.31. Found: C, 49.11; H, 5.53; N, 12.27.

1-Deoxy-1-(*o*-nitroanilino)-2,3,4-tri-O-acetyl-D-glucopyranuronamide (XIIa) and 1-Deoxy-1-(5-nitro-*o*-xylidino)-2,3,4-tri-O-acetyl-D-glucopyranuronamide (XIIb)——Essentially the same procedure as described for the preparation of VIIIa or VIIIb was employed. XIIa: mp 165—166°. $[\alpha]_D^{23}$ +72.8° (c=1.50, $CHCl_3$). *Anal.* Calcd. for $C_{18}H_{21}O_{10}N_3$: C, 49.20; H, 4.78; N, 9.57. Found: C, 49.03; H, 4.65; N, 9.34. (XIIb): mp 213—214°. $[\alpha]_D^{23}$ − 42.7° (c=1.52, $CHCl_3$). *Anal.* Calcd. for $C_{20}H_{25}O_{10}N_3$: C, 51.39; H, 5.35; N, 8.99. Found: C, 51.26; H, 5.16; N, 8.77.

1-Deoxy-1-(*o*-phenylenediamino)-2,3,5-tri-O-acetyl-α-D-glycofuranuroamide (IXa) and 1-Deoxy-1-(4,5-dimethyl-*o*-phenylenediamino)-2,3,5-tri-O-acetyl-α-D-glucofuranuronamide (IXb)——0.05 mole of VIIIa or VIIIb dissolved in 250 ml of AcOEt was hydrogenated by shaking with 0.5 g of PtO_2 catalyst in an atmosphere of hydrogen under water cooling until there was no absorption of hydrogen. After collection of the catalyst, the solvent was removed by distillation under reduced pressure. A pale yellow syrup thus obtained was azeotropically dried by distillation with EtOH and toluene several times, and then used for the subsequent ring closure for benzimidazole derivatives without further purification.

1-Deoxy-1-(*o*-phenylenediamino)-2,5-di-O-acetyl-α-D-glucofuranurono-6,3-lactone (IVa) and 1-Deoxy-1-(4,5-dimethyl-*o*-phenylenediamino)-2,5-di-O-acetyl-α-D-glucofuranurono-6,3-lactone (IVb)——These compounds were obtained as a slightly brown syrup by essentially the same procedure described above and likewise used for the subsequent ring closure for benzimidazole derivatives without furhter purification.

An Attempted Cyclisation of IXa or IXb with Ethyl Formimidate Hydrochloride——The hydrogenated and dried syrup (IXa or IXb) (prepared from 0.01 mole of VIIIa or VIIIb) was dissolved in 50 ml of MeOH and this solution was heated under reflux with 2.1 g of ethyl formimidate hydrochloride[22] on a water bath for 48 hr. After removal of MeOH, the residue was dissolved in H_2O followed by neutralization with $NaHCO_3$, and the solution was left standing in a refrigerator. The separated crystals were recrystallized from H_2O. Benzimidazole[23] melting at 169—170° and 5,6-dimethylbenzimidazole[24] melting at 203—204° were obtained in the yields of 53 and 57%, calculated from IXa and IXb, respectively.

1-Deoxy-1-(1-benzimidazolyl)-β-D-glucopyranuronamide (XVa)——A) Ethyl Orthoformate Method (*via* the Ring Closure of Phenylenediamine Derivative): 2.2 g (0.005 mole) of XIIa was dissolved in 60 ml of AcOEt and this solution was shaken with hydrogen in the presence of a platinum catalyst (PtO_2, 0.1 g) until hydrogen uptake was complete. The catalyst was filtered off and the filtrate was evaporated *in vacuo* to give a crystalline solid melting at 133—134°, which was dissolved in 50 ml of EtOH containing 5 ml of

22) L.F. Cavalieri, J.F. Tinker, A. Bendich, *J. Am. Chem. Soc.*, 71, 533 (1949).
23) *Org. Synth.*, Coll. Vol. II, 65 (1943).
24) K. Folkers, N.G. Brink, *J. Am. Chem. Soc.*, 72, 442 (1950).

ethyl orthoformate. The mixture was heated on a water bath in an open flask for 2 hr. Evaporation of the excess ethyl orthoformate afforded a syrup, which was then treated with 20 ml of 0.05N HCl on a water bath for 15 minutes. After neutralisation with $NaHCO_3$, the mixture was extracted with $CHCl_3$ (20 ml) five times. The $CHCl_3$ extract was washed with H_2O and then dried over Na_2SO_4. After removal of the solvent, the residual syrup was dissolved in ammoniacal EtOH (30 ml) saturated at 0° and this solution was allowed to stand in a refrigerator overnight. The solvent was evaporated *in vacuo* to give a residue, which on recrystallization from 90% EtOH afforded 0.24 g (16.4% calculated from (XIIa)) of colorless needles, mp 191—192° (decomp.). $[\alpha]_D^{24}$ +19.6° (c=1.81, 0.1N HCl). *Anal.* Calcd. for $C_{13}H_{15}O_5N_3$: C, 53.24; H, 5.12; N, 14.33. Found: C, 53.54; H, 5.01; N, 14.39.

B) Chloromercuribenzimidazole Procedure: A suspension of 5.3 g (0.015 mole) of the finely powdered chloromercuribenzimidazole in 420 ml of xylene was dried by slow distillation of *ca.* 120 ml of the solvent and then cooled to 100°. To the warm suspension was added 6.3 g (0.016 mole) of methyl 1-deoxy-1-bromo-2,3,4-tri-O-acetyl-α-D-glucopyranuronate[16] and the mixture was heated under reflux with mechanical stirring for 1.25 hr, then chilled and diluted with 500 ml of petroleum ether. The precipitate formed was filtered, washed with petroleum ether, dried and then extracted with $CHCl_3$. The $CHCl_3$ extract was washed with 30% KI, subsequently with H_2O and dried over Na_2SO_4. Evaporation of the solvent afforded a syrup, which on recrystallization from EtOH yielded 2.6 g of fine needles melting at 185—186°. 1.3 g portions of the above product was dissolved in 70 ml of ammoniacal MeOH saturated at 0° and this solution was left to stand in a refrigerator overnight. The solvent was removed under reduced pressure to give a residue. Recrystallization from 90% EtOH afforded 0.7 g (31.8%) of fine needles. mp 191—192° (decomp.). $[\alpha]_D^{22}$ +22.6° (c=1.38, 0.1N HCl).

1-Deoxy-1-(5,6-dimethyl-1-benzimidazolyl)-β-D-glucopyranuronamide (XVb)——This compound was obtained in the yields of 20.1% (method A) and 23.5% (method B) by exactly the same procedure mentioned above. mp 264—265° (decomp.) (colorless needles from H_2O). $[\alpha]_D^{22}$ +16.2° (c=1.23, 0.1N HCl). *Anal.* Calcd. for $C_{15}H_{19}O_5N_3$: C, 56.07; H, 5.91; N, 13.08. Found: C, 55.87; H, 5.65; N, 12.91.

1-Deoxy-1-(5,6-dichloro-1-benzimidazolyl)-β-D-glucopyranuronamide (XVc)——This compound was obtained in a yield of 41.2% by method B. mp 247—248° (decomp.) (recrystallized from MeOH). $[\alpha]_D^{22}$ +15.7° (c=1.05, 0.1N HCl). *Anal.* Calcd. for $C_{13}H_{13}O_5N_3Cl_2$: C, 43.09; H, 3.59; N, 11.60 Found: C, 42.79; H, 3.44; N, 11.58.

Hydrolysis of VIIIa or VIIIb with Aqueous AcOH——A solution of 3 g of finely powdered VIIIa or VIIIb in 40 ml of 5% AcOH was heated at 80° with intermittent swirling for 20 minutes, during which times hydrolytic cleavage occurred to liberate *o*-nitroaniline or 5-nitro-*o*-4-xylidine. After cooling, the reaction mixture was filtered and the filtrate was treated with 20 ml (wet volume) of Amberlite IR-120 (H^+ Form). After removal of the resin, the slightly brown filtrate was decolored with activated carbon and then evaporated under diminished pressure to a syrup, which was taken up in a small volume of AcOEt and the solution was chilled in a refrigerator overnight. Separated crystals were recrystallized from 95% EtOH in colorless needles. Yield, 0.8 g. mp 155—158°. $[\alpha]_D^{23}$ +63.1° (c=1.10, H_2O). No depression of the melting point of this compound was observed on admixture with 2,3,5-tri-O-acetyl-D-glucofuranuronamide,[14] which had been synthesized from *p*-nitroanilino-2,3,5-tri-O-acetyl-D-glucofuranuronamide by mild acid hydrolysis. Infrared absorption spectra of both compounds were identical.

Hydrolysis of XIIa or XIIb with aqueous AcOH——Exactly the same procedure described above was employed. The product melted at 148—149° and showed $[\alpha]_D^{24}$ +148.4° (c=2.40, $CHCl_3$). The melting point of this compound was undepressed on admixture with 2,3,4-tri-O-acetyl-D-glucopyranuronamide,[16] which had been prepared by treatment of 1-bromo-1-deoxy-2,3,4-tri-O-acetyl-α-D-glucopyranuronamide with silver carbonate in aqueous acetone. Infrared absorption spectra of both compounds were identical.