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5. Atsushi Yamada : Studies on the Derivatives of Cardiac Aglycones of Digitalis.*1

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Several derivatives of cardiac aglycones of digitalis, digitoxigenin, gitoxigenin, and digoxigenin, including those reported in the past literature, were prepared for the purpose of investigating the relationship between the specific heart action of these cardiac aglycones and their chemical configuration as well as their structure.

3-Epidigitoxigenin (IV) was prepared in a good yield by the sodium borohydride reduction of digitoxigenone[1] (III), which was obtained almost quantitatively by the oxidation of digitoxigenin (I) with the Jones reagent.[2] 3-Dehydrodigoxigenin[3] (XV), which was obtained by the microbiological hydroxylation of digitoxigenone*3 (III), was treated with sodium borohydride and gave 3-epidigoxigenin[3] (XX). When oleandrigenone (X) was reduced with sodium borohydride and the reduction product was chromatographed on a mixture of Florisil and Celite, 3-epioleandrigenin (XI) was obtained as the main product, accompanied with a small quantity of 3-epigitoxigenin (XII), possibly produced by hydrolysis of the acetyl group during reduction. Treatment of (XI) in hydrous methanol with potassium hydrogencarbonate afforded (XII). The diacetate of (XII) as well as of (XX) could not be crystallized. Table I shows the molecular rotation difference between natural cardiotonic aglycones with 3β-hydroxyl group and their corresponding synthetic C-3 epimers described above.

Table I. Molecular Rotation Differences between C-3 Epimers

Compound	$[\alpha]_D$	$[M]_D$	ΔC-3(α-OH) - (β-OH)
3-Epidigitoxigenin (IV)	+27 (MeOH)	+101	+30
Digitoxigenin (I)	+19 (〃)	+ 71	
3-Epigitoxigenin (XII)	+39.9 (MeOH)	+156	+27
Gitoxigenin (V)	+33 (〃)	+129	
3-Epioleandrigenin (XI)	0 (EtOH)	0	+43
Oleandrigenin (VII)	−10 (MeOH)	− 43	
3-Epidigoxigenin (XX)	+27 (MeOH)	+105	+42
Digoxigenin (XVII)	+16 ($CHCl_3$)	+ 63	

Partial hydrolysis of diacetylgitoxigenin (VIII) reported by Cardwell, *et al.*[4] gave 3-monoacetylgitoxigenin (VI), while oleandrigenin (VII) was prepared from gitoxin according to the procedure reported by Meyer, *et al.*[5] 12-Monoacetyldigoxigenin (XIII) was obtained in a fairly good yield in the present work by a mild acid hydrolysis of digoxin acetate, though Shindler[6] reported the preparation of this compound by the partial acetylation of digoxigenin (XVII). When diacetyldigoxigenin (XVIII) was treated in hydrous methanol with potassium hydrogencarbonate at room temperature, followed by chromatography

*1 A preliminary account of a part of the present work has already appeared in This Bulletin, **4**, 420(1956).

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*3 The results obtained concerning the microbiological hydroxylation of cardiotonic aglycones and their derivatives will be reported at a later date.

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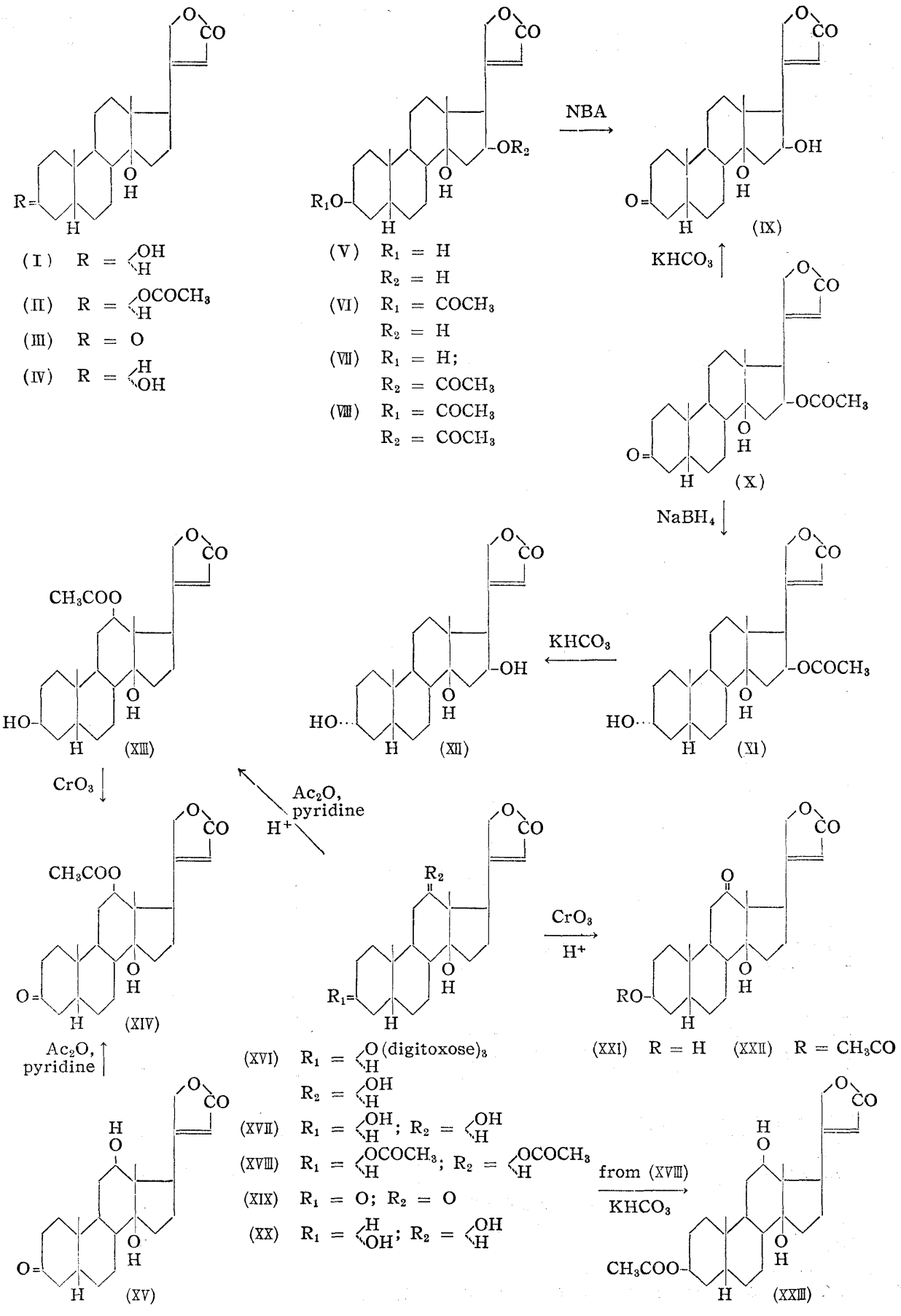

(I) R = 6-OH
(II) R = 6-OCOCH3
(III) R = O
(IV) R = 6-H, OH

(V) R1 = H; R2 = H
(VI) R1 = COCH3; R2 = H
(VII) R1 = H; R2 = COCH3
(VIII) R1 = COCH3; R2 = COCH3

(IX) (X) (XI) (XII) (XIII) (XIV) (XV)

(XVI) R1 = 3-O(digitoxose)3; R2 = 6-OH
(XVII) R1 = 3-OH; R2 = 6-OH
(XVIII) R1 = 3-OCOCH3; R2 = 6-OCOCH3
(XIX) R1 = O; R2 = O
(XX) R1 = 3-H, OH; R2 = 6-OH

(XXI) R = H (XXII) R = CH3CO

(XXIII)

on alumina, a monoacetyldigoxigenin, m.p. 260~264.5°, $[\alpha]_D^{21}$ +18°(chloroform), different from (XIII), m.p. 275~280°, $[\alpha]_D^{21}$ +52°(chloroform), was obtained. That this new monoacetyldigoxigenin is 3-monoacetyldigoxigenin (XXIII), as reasonably anticipated, is evident from the molecular rotation data shown in Table II.

Oxidation of gitoxigenin (V) with N-bromoacetamide gave 3-dehydrogitoxigenin (IX), which was proved to be identical with that obtained from oleandrigenone (X) by its hydrolysis with potassium hydrogencarbonate. Tamm and his co-workers[7] recently

Table II. Molecular Rotation Contribution of Acetoxyl Groups at C-3 and C-12

Compound	$[\alpha]_D$	$[M]_D$	ΔC-3(β-OCOCH$_3$) - (β-OH)
Acetyldigitoxigenin (II)	+ 21 ($CHCl_3$)	+ 88	+ 17
Digitoxigenin (I)	+ 19 (MeOH)	+ 71	
3-Monoacetylgitoxigenin (VI)	+ 33 ($CHCl_3$)	+143	+ 14
Gitoxigenin (V)	+ 33 (MeOH)	+129	
3-Monoacetyldigoxigenin (XXIII)	+ 18 ($CHCl_3$)	+ 78	+ 15
Digoxigenin (XVII)	+ 16.2 (〃)	+ 63	
3-Acetyl-12-dehydrodigoxigenin (XXII)	+112 ($CHCl_3$)	+482	+ 43
12-Dehydrodigoxigenin (XXI)	+113 (〃)	+439	

Compound	$[\alpha]_D$	$[M]_D$	ΔC-12(β-OCOCH$_3$) - (β-OH)
12-Monoacetyldigoxigenin (XIII)	+ 52 ($CHCl_3$)	+225	+162
Digoxigenin (XVII)	+ 16.2 (〃)	+ 63	
12-Acetyl-3-dehydrodigoxigenin (XXII)	+ 72 ($CHCl_3$)	+310	+186
3-Dehydrodigoxigenin (XV)	+ 32 ($CHCl_3$·MeOH)	+124	

Compound	$[M]_D$	Δ-Values	
3-Monoacetyldigoxigenin (XXIII)	+ 78	ΔC-3(β-OCOCH$_3$) - (β-OH)	+ 15
Digoxigenin (XVII)	+ 63		
12-Monoacetyldigoxigenin (XIII)	+225	ΔC-12(β-OCOCH$_3$) - (β-OH)	+162
Digoxigenin (XVII)	+ 63		
Δ[C-3(β-OCOCH$_3$)] + Δ[C-12(β-OCOCH$_3$)]			+177
Diacetyldigoxigenin (XVIII) $[\alpha]_D$ +50.4 ($CHCl_3$)	+239		+176
Digoxigenin (XVII)	+ 63		

Table III. Molecular Rotation Contribution of Keto Groups at C-3 and C-12

Compound	$[\alpha_D]$	$[M]_D$	ΔC-3(=O) - (β-OH)
Digitoxigenone (III)	+ 33 ($CHCl_3$)	+123	+ 52
Digitoxigenin (I)	+ 19 (MeOH)	+ 71	
3-Dehydrogitoxigenin (IX)	+ 48.5 ($CHCl_3$)	+189	+ 60
Gitoxigenin (V)	+ 33 (MeOH)	+129	
Oleandrigenone (X)	− 4.5 ($CHCl_3$)	− 19	+ 24
Oleandrigenin (VII)	− 10 (MeOH)	− 43	
3-Dehydrodigoxigenin (XV)	+ 32 ($CHCl_3$·MeOH)	+124	+ 61
Digoxigenin (XVII)	+ 16.2 ($CHCl_3$)	+ 63	
3-Dehydro-12-acetyldigoxigenin (XIV)	+ 72 ($CHCl_3$)	+310	+ 85
12-Monoacetyldigoxigenin (XIII)	+ 52 (〃)	+225	

Compound	$[\alpha]_D$	$[M]_D$	ΔC-12(=O) - (β-OH)
12-Dehydrodigoxigenin (XXI)	+113 ($CHCl_3$)	+440	+377
Digoxigenin (XVII)	+ 16.2 (〃)	+ 63	
12-Dehydro-3-acetyldigoxigenin (XXII)	+112 ($CHCl_3$)	+482	+404
3-Monoacetyldigoxigenin (XXIII)	+ 18 (〃)	+ 78	

Compound	$[M]_D$	Δ—Values	
3-Dehydrodigoxigenin (XV)	+124	ΔC-3(=O) - (β-OH)	+ 61
Digoxigenin (XVII)	+ 63		
12-Dehydrodigoxigenin (XXI)	+440	ΔC-12(=O) - (β-OH)	+377
Digoxigenin (XVII)	+ 63		
Δ[C-3(=O)] + Δ[C-12(=O)]			+438
Bisdehydrodigoxigenin (XIX) $[\alpha]_D$ +130 (Me_2CO)	+503		+440
Digoxigenin (XVII)	+ 63		

7) Ch. Tamm, A. Gubler : Helv. Chim. Acta, **41**, 1762(1958).

reported the preparation of this compound by selective dehydrogenation of the hydroxyl group at C-3 of gitoxigenin (V) with platinum oxide. Oxidation of digoxigenin (XVII) with the Jones reagent afforded bisdehydrodigoxigenin[8] (XIX) in almost quantitative yield. The preparation of 12-dehydrodigoxigenin (XXI) was achieved by a mild acid hydrolysis of the oxidation product of digoxin (XVI) in acetic acid with chromium trioxide. It showed ultraviolet maximum at 217 mμ (log ε 4.23) with a weak shoulder at ca. 280 mμ, and an absorption for a six-membered ketone at 5.87 μ (KBr) in its infrared spectrum.

On the other hand, 3-dehydrodigoxigenin (XV) was prepared by the microbiological hydroxylation of digitoxigenone.*3 Contribution of molecular rotation due to the keto group in digoxigenin is summarized in Table III.

Acetylation of (XXI) in the usual way with acetic anhydride in pyridine gave the acetate (XXII), m.p. 228~229.5°, $[\alpha]_D^{21}$ +112° (chloroform), the physical constants of which agreed quite well with those of the acetoxy-ketone, m.p. 228~229°, $[\alpha]_D$ +113°, of Cardwell *et al.*,[5] prepared from diacetyldigoxigenin (XVIII) by its partial hydrolysis with alkali and subsequent oxidation with chromium trioxide.

3-Dehydrodigoxigenin acetate (XIV) was obtained as leaflets either by the acetylation of 3-dehydrodigoxigenin (XV) with acetic anhydride in pyridine, or by oxidation of 12-monoacetyldigoxigenin (XIII) with chromium trioxide, while Shindler[6] failed in crystallization of (XIV).

Contributions of molecular rotation due to the keto and acetoxyl groups in (XIV) and (XXII) are indicated in Tables II and III, respectively.

In Table IV the toxicity of various derivatives of cardiac aglycones of digitalis described in this paper are compared with those of the parent compounds.*4

Acetylation of 3β-hydroxyl group of the parent aglycones resulted in the decrease

Table IV. Toxicity by Pigeon Method*

	Compound	No. of animals	LD (mg./kg.)
A-series	Digitoxigenin (I)	6	0.60
	Acetyldigitoxigenin (II)	5	1.87
	Digitoxigenone (III)	7	3.21
	3-Epidigitoxigenin (IV)	1	10.0
B-series	Gitoxigenin (V)	6	5.40
	3-Monoacetylgitoxigenin (VI)	5	6.53
	Oleandrigenin (VII)	6	0.95
	Diacetylgitoxigenin (VIII)	5	1.28
	3-Dehydrogitoxigenin (IX)	1	—**
	Oleandrigenone (X)	7	3.01
	3-Epioleandrigenin (XI)	7	5.785
	3-Epigitoxigenin (XII)	1	—**
C-series	Digoxigenin (XVII)	5	1.62
	3-Monoacetyldigoxigenin (XXIII)	5	2.66
	12-Monoacetyldigoxigenin (XIII)	1	—**
	Diacetyldigoxigenin (XVIII)	2	10.0
	3-Dehydrodigoxigenin (XV)	4	4.94
	12-Dehydrodigoxigenin (XXI)	5	4.00
	Bisdehydrodigoxigenin (XIX)	1	—**
	3-Acetyl-12-dehydrodigoxigenin (XXII)	1	9.3
	12-Acetyl-3-dehydrodigoxigenin (XIV)	1	—**
	3-Epidigoxigenin (XX)	1	—**

* This was performed according to the procedure of U.S.P. XIV, except the injection period which was in 2.5-min. intervals.

** This indicates that the animals did not die until 10 mg./kg. was injected.

*4 The toxicity determination was carried out by Mr. C. Isono of this Institute under the direction of Prof. K. Tokita, to whom the author's thanks are due.

8) S. Smith : J. Chem. Soc., **1935**, 1305.

of toxicity to some extent, while the dehydrogenation of this group reduced the toxicity remarkably. The β-configuration of the hydroxyl group at C-3 of these cardiotonic aglycones is considered essential to their characteristic effect, because the 3-epimers except 3-epioleandrigenin (XI) were found to be inactive.

A marked increase of the toxicity resulted by acetylating the 16β-hydroxyl group in the B-series, while inactivation of compounds in the C-series was observed by acetylation of 12β-hydroxyl group, the dehydrogenation of which, on the other hand, caused a striking decrease of toxicity.

Experimental

All melting points are uncorrected. Infrared spectra were obtained with Perkin-Elmer 21 Double-beam Spectrophotometer and ultraviolet spectra were measured with Beckman Model DU Spectrophotometer.

Digitoxigenone (III)—437 mg. of digitoxigenin (I) was dissolved in 59 cc. of Me_2CO (previously distilled with $KMnO_4$) and the solution after cooling to about 10° was treated rapidly and with vigorous stirring with 0.39 cc. of standard CrO_3 reagent (a solution of 8*N* CrO_3 in H_2SO_4) and diluted with water after 2 min. The product was extracted with AcOEt, the extract was washed with 5% Na_2CO_3 and water, dried (Na_2SO_4), and evaporated. Crystallization of the residue from Me_2CO-petr. ether yielded 416 mg. (96%) of (III), m.p. 197~199°/203~204° (reported[1] m.p. 203~205°).

Bisdehydrodigoxigenin (XIX)—This was prepared from digoxigenin (XVII) by its oxidation with standard CrO_3 reagent described above. 200 mg. of (XVII) gave 184 mg. (93%) of (XIX), m.p. 260.5~261° (from Me_2CO) (reported[8] m.p. 265°).

3-Epidigitoxigenin (IV)—To a solution of 230 mg. of digitoxigenone (III) in 14 cc. of 80% MeOH, 180 mg. of $NaBH_4$ was added and the solution was allowed to stand for 5 hr. at room temperature. The solution was acidified to ca. pH 3 with *N* H_2SO_4 and allowed to stand overnight. Water was added and the solution was concentrated under a reduced pressure, the separated crystals were collected, and recrystallized from MeOH. Yield, 192 mg., m.p. 282~284° (reported[1] m.p. 274~282°).

3-Dehydrogitoxigenin (IX)—To a solution of 200 mg. of gitoxigenin (V) in 10.7 cc. of *tert*-BuOH, 1.07 cc. of pyridine, and 1.07 cc. of water, 353 mg. of N-bromoacetamide was added. The oxidation in the resulting solution was allowed to proceed at room temperature for 11 days protected from light. At the end of this time, 1.34 g. of KI in 6.7 cc. of water was added, followed by 10 cc. of 0.1 *N* $Na_2S_2O_3$, and the mixture was extracted with $CHCl_3$. After successive washing with alkali, acid, and water, the dried solution was evaporated, leaving 214 mg. of a gummy residue which provided 146 mg. of crystals (from MeOH-Et_2O) melting at 170~200°. Chromatographic purification of the product over acid-washed alumina and recrystallization from Me_2CO-Et_2O afforded 133 mg. of (IX) as prisms, m.p. 210~217°. Admixture with a sample of (IX), m.p. 174~178°/207~213°, prepared by hydrolysis of (X) (see below) melted at 175~189°/203~217°, $[\alpha]_D^{23}$ +56° ($CHCl_3$) (reported[7] m.p. 177~178°/206~211°, $[\alpha]_D^{25}$ +48.5° ($CHCl_3$)). UV: λ_{max}^{EtOH} 217 mμ (log ε 4.19). IR λ_{max}^{Nujol} μ: 5.53, 5.63, 5.90, 6.15. *Anal.* Calcd. for $C_{23}H_{32}O_5$: C, 71.10; H, 8.30. Found: C, 71.07; H, 7.92.

Preparation of (IX) from Oleandrigenone (X)—A solution of 74 mg. of (X) in 18 cc. of MeOH was treated with 74 mg. of $KHCO_3$ in 18 cc. of water and the solution was stored at room temperature for 18 days. After the usual work-up, 36 mg. of (IX), m.p. 174~178°/207~213°, was obtained.

3-Epioleandrigenin (XI)—To a solution of 223 mg. of oleandrigenone (X) in 10.5 cc. of 80% dioxane, 70 mg. of $NaBH_4$ in 7 cc. of 80% dioxane was added dropwise over 2-hr. period and the mixture was allowed to stand for 5 hr. at about 20°. The solution was acidified to pH ca. 3 with 2*N* H_2SO_4 and again allowed to stand overnight. Water was then added, the solution was evaporated to a small volume under reduced pressure, and extracted with $CHCl_3$. The extract was washed with dil. Na_2CO_3 and water, and evaporated to dryness. The residue was refluxed for 30 min. in a mixture of 550 mg. of *d*-mannitol in 25 cc. of MeOH and 25 cc. of 0.1*N* H_2SO_4. After evaporating MeOH *in vacuo* the product was extracted with $CHCl_3$ and dried *in vacuo*. The residue was chromatographed on 4 g. of Florisil-Celite (535) mixture (2:1) by successive elution with benzene, $CHCl_3$, and MeOH. The fraction eluted with benzene-$CHCl_3$ mixture (1:1) and $CHCl_3$ gave, after recrystallization from MeOH-Et_2O, 187 mg. of (XI), as prisms, m.p. 207~212°; $[\alpha]_D^{15}$ ±0° (EtOH). UV: λ_{max}^{EtOH} 217 mμ (log ε 4.16). *Anal.* Calcd. for $C_{25}H_{36}O_6$: C, 69.42; H, 8.39. Found: C, 69.38; H, 8.20.

The fraction eluted with $CHCl_3$-MeOH (1:1) gave a small amount of crystals of m.p. 222~226°, which was identical with (XII).

65 mg. of (XI) was acetylated in the usual way with Ac_2O and pyridine to prepare the diacetate of (XII), but it could not be crystallized.

3-Epigitoxigenin (XII)—To a solution of 79 mg. of (XI) in 20 cc. of MeOH, 79 mg. of $KHCO_3$ in 4

cc. of water was added and the solution allowed to stand at room temperature for 15 days. After evaporating MeOH *in vacuo* the product was extracted with $CHCl_3$, the organic phase was washed with water, and dried. Recrystallization of the residue from $CHCl_3$-Et_2O afforded (XII), m.p. 223~226°, $[\alpha]_D^{20}$ +39.9° (MeOH). UV: λ_{max}^{EtOH} 217 mμ (log ε 4.19). *Anal.* Calcd. for $C_{23}H_{34}O_5$: C, 70.74; H, 8.78. Found: C, 70.70; H, 8.56. Admixture with gitoxigenin (V), m.p. 224~226°, showed m.p. 205~214°.

12-Monoacetyldigoxigenin (XIII)—500 mg. of digoxin (XVI) was acetylated with Ac_2O in pyridine. The acetylated product obtained in the usual way was refluxed for about 45 min. in a mixture of 50 cc. of MeOH and 50 cc. of 0.1*N* H_2SO_4. After evaporating MeOH at 40° under reduced pressure, the product was extracted with $CHCl_3$, the $CHCl_3$ layer was washed with water, dried over anhyd. Na_2SO_4, and the solvent evaporated. The residue was crystallized from Me_2CO-Et_2O-petr. ether to 210 mg. of (XIII), m.p. 268~279° The analytical sample melted at 275~280°; $[\alpha]_D^{21}$ +52° ($CHCl_3$) (reported[6] m.p. 283~286°, $[\alpha]_D^{22}$ +56.3° ($CHCl_3$)). UV: λ_{max}^{EtOH} 218 mμ (log ε 4.21). IR: $\lambda_{max}^{CH_2Cl_2}$ μ: 5.60, 5.73, 6.15, 8.10. *Anal.* Calcd. for $C_{25}H_{36}O_3$: C, 69.42; H, 8.39. Found: C, 69.11; H, 8.11.

3-Dehydro-12-acetyldigoxigenin (XIV)—A solution of 100 mg. of the 12-monoacetyldigoxigenin (XIII) in AcOH was oxidized with 2% CrO_3-AcOH reagent by the procedure previously described for the preparation of (XXI). There was obtained 92 mg. of (XIV), which could not be crystallized, while chromatographic purification on acid-washed alumina gave 46 mg. of crystalline (XIV), m.p. 183~192°. The analytical sample was recrystallized from Me_2CO-Et_2O-petr. ether, m.p. 193~197°. The melting point of the mixture with the acetate prepared from (XV) by acetylation showed no depression. $[\alpha]_D^{21}$ +72° ($CHCl_3$). UV: λ_{max}^{EtOH} 217 mμ (log ε 4.20). IR $\lambda_{max}^{CH_2Cl_2}$ μ: 5.63, 5.77, 5.87, 6.15, 8.12. *Anal.* Calcd. for $C_{25}H_{34}O_6$: C, 69.74; H, 7.96. Found: C, 69.35; H, 7.96.

Acetylation of 3-dehydrodigoxigenin (XV) in the usual way with Ac_2O and pyridine yielded (XIV), m.p. 193~197°.

3-Dehydrodigoxigenin (XV)—This compound was prepared by microbial hydroxylation of digitoxigenone (III) as crystals of m.p. 251~254°, $[\alpha]_D^{21}$ +32° ($CHCl_3$·MeOH) (reported[3] m.p. 247~252°). UV: λ_{max}^{EtOH} 219 mμ (log ε 4.24). IR λ_{max}^{KBr} μ: 5.63, 7.54, 5.94, 6.14.

3-Epidigoxigenin (XX)—A solution of 66 mg. of 3-dehydrodigoxigenin (XV) in 5 cc. of MeOH was treated at about 20° with ca. 55 mg. of $NaBH_4$ in 1 cc. of water and the solution was allowed to stand for 5 hr. at this temperature. m.p. 258~265°, $[\alpha]_D^{21}$ +37° (EtOH) (reported[3] m.p. 249~262°, $[\alpha]_D^{26}$ +27° (MeOH)). UV: λ_{max}^{EtOH} 218 mμ (log ε 4.20). IR λ_{max}^{KBr} μ: 5.53, 5.76, 6.17.

12-Dehydrodigoxigenin (XXI)—To a solution of 500 mg. of digoxin (XVI) in 60 cc. of AcOH 3.25 cc. of 2% CrO_3-AcOH solution was added during 2 hr. After standing at room temperature for 1 hr., an additional 6.0 cc. of the above reagent was added during 2 hr. and again allowed to stand for 1 hr. 7.5 cc. of MeOH was added to decompose the excess CrO_3. After standing the reaction mixture for 15 hr. at room temperature, the solvent was evaporated under a reduced pressure, the residue was extracted with AcOEt, the extract was washed with 5% Na_2CO_3 and water, dried over Na_2SO_4, and evaporated. The residue was refluxed for 45 min. in a mixture of 70 cc. of MeOH and 70 cc. of 0.1*N* H_2SO_4. After evaporating MeOH *in vacuo* the product was extracted with AcOEt and the solvent evaporated to dryness *in vacuo*. The product, which crystallized from Me_2CO-Et_2O, was purified by recrystallization from Me_2CO yielding 209 mg. (84%) of (XXI), m.p. 269~272°, $[\alpha]_D^{21}$ +113° ($CHCl_3$). IR λ_{max}^{KBr} μ: 5.55, 5.75, 5.87, 6.12. *Anal.* Calcd. for $C_{23}H_{32}O_5$: C, 71.10; H, 8.30. Found: C, 71.03; H, 8.26.

12-Dehydro-3-acetyldigoxigenin (XXII)—Acetylation of 45 mg. of (XXI) with Ac_2O and pyridine in the usual way yielded 50 mg. of the acetate (XXII). The analytical sample was crystallized from hydr. MeOH. UV: λ_{max}^{EtOH} 217 mμ (log ε 4.24). IR $\lambda_{max}^{CH_2Cl_2}$ μ: 5.60, 5.75, 5.79, 5.87, 6.13, 8.12. *Anal.* Calcd. for $C_{25}H_{34}O_6$: C, 69.74; H, 7.96. Found: C, 69.76; H, 7.91.

3-Monoacetyldigoxigenin (XXIII)—To a solution of 200 mg. of diacetyldigoxigenin (XVIII) (m.p. 219~223°) in 50 cc. of MeOH 200 mg. of $KHCO_3$ in 9.8 cc. of water was added and the solution was stored at room temperature for 3 weeks. After evaporating MeOH *in vacuo* the product was extracted with AcOEt. The organic phase was washed with water, dried, and the solvent was evaporated. The residue (195 mg.) was chromatographed on 6 g. of acid-washed alumina by successive elution with benzene-$CHCl_3$. The fraction (82 mg.) eluted with benzene-$CHCl_3$ mixture (9:1, 4:1, and 3:1) gave 55 mg. of diacetyldigoxigenin (m.p. 215~221°). The fraction (85 mg.) eluted with denzene-$CHCl_3$ mixture (1:1 and 1:2) and $CHCl_3$ gave 57 mg. of 3-monoacetyldigoxigenin (XXIII). The analytical sample was recrystallized from Me_2CO-Et_2O-petr. ether. UV: λ_{max}^{EtOH} 218 mμ (log ε 4.23). IR λ_{max}^{KBr} μ: 5.53, 5.71, 5.83, 6.14, 8.09. *Anal.* Calcd. for $C_{25}H_{36}O_6$: C, 69.42; H, 8.39. Found: C, 69.23; H, 8.22.

The presence of digoxigenin (XVII) in the fraction eluted with $CHCl_3$ containing 3% MeOH was indicated by paper chromatography.

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Summary

Acetyl, dehydro, and 3-epi derivatives of the cardiac aglycones of digitalis, digitoxigenin, gitoxigenin, and digoxigenin, including those reported in past literature, were prepared. The toxicity of these derivatives by the pigeon method was compared and discussed.

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6. Eiji Ochiai und Yutaka Kawazoe: Polarisation der heterozyklischen Ringe mit aromatischem Charakter. CXXXVI. Beitrag zur Nitrierung der Isochinolin-Derivate.[1]

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Die Nitrierung von Isochinolin selbst unter milder Bedingung ergibt hauptsächlich das 5-Nitroderivat und nebenbei das 8-Nitroderivat in sehr kleiner Menge.[3] Die Entstehung von 8-Nitroderivat bei dieser Reaktion sowie bei analoger Nitrierung von 5-Chlor[4]-, 7-Chlor-[5] und 5-Acetamido-isochinolin[4] zeigt die Reaktivität der 8-Stellung gegen die elektrophile Substitution, obwohl in untergeordnetem Grad. Ferner wurde gezeigt, dass die Isochinolin-Derivate, die in ihrem Pyridin-Teil einen Substituent mit einem kleinen +M Effekt wie Halogen- oder Alkyl-gruppe tragen, hauptsächlich auch an der 5-Stellung nitriert werden. Nämlich ergibt 1-Chlor-[3,6] sowie 4-Brom-isochinolin[7,8] dabei das 5-Nitroderivat und 3-Methylisochinolin ein Mononitroderivat, welches als 5-Nitroderivat vermutet wurde.[9] 1-Benzylisochinolin wird zuerst an der *para*-Stellung des Benzylrestes[10] und dann an der 5-Stellung des Isochinolin-Teils[11] nitriert. Noch zu bemerken ist die Tatsache, dass 3-Methyl-4-methoxyisochinolin auf dem Benzol-Teil nitriert wird.[12] Der polare Effekt des Kernstickstoffes scheint so gross zu sein, dass die *para*-Stellung der Methoxylgruppe unsubstituiert bleibt. Ferner wurde nun gezeigt, dass 1-Äthoxyisochinolin (I) an der 5-Stellung nitriert wird. (I) ergab nämlich bei der Nitrierung mit Salpeter in konz. Schwefelsäure bei Zimmertemperatur ein schwachgelbes nadelförmiges Mononitroderivat (II) vom Schmp. 141°, dessen Konstitution

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