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AN UNUSUAL FLUORIDE-MEDIATED OXIDATION OF GITOXIN[1)]

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An unusual oxidative transformation of α,β-unsaturated γ-lactone of the cardenolide gitoxin (**1a**) by tetra-*n*-butylammonium fluoride (TBAF) is reported. The structure of the product (**2a**) was finally confirmed by an X-ray crystal structure analysis of its genin (**2b**). The oxidation proceeded also in the presence of CsF, and KF + 18-crown-6.

KEY WORDS gitoxin; α,β-unsaturated γ-lactone; oxidation; fluoride ion

Gitoxin (**1a**) is a cardiac glycoside isolated from *Digitaris purupurea* as one of its major components along with digitoxin, but is not used clinically because of its low solubility to water and alcohol and its low uptake from the digestive system. Thus, we attempted to create more potent compounds from **1a**. In the course of this study, we found that treatment of gitoxin (**1a**) only with tetra-*n*-butylammonium fluoride (TBAF) caused oxidative transformation of the α,β-unsaturated γ-lactone moiety. In this paper, we report identification of this product obtained by the unusual oxidation induced by fluoride anion.

Gitoxin (**1a**, 50 mg) was treated with TBAF (5 eq)[2)] in freshly distilled THF, DMF, or CH_2Cl_2 (5 ml) at r.t. for an hour to give **2a**[3)] in 86% yield. FAB-MS spectrum of **2a** indicated quasimolecular peaks at 795 ($M+H^+$) and 817 ($M+Na^+$). In conjunction with elemental analysis data, the molecular formula of **2a** was determined as $C_{41}H_{62}O_{15}$. In the ^{1}H- and ^{13}C-NMR spectra of **2a**, the methylene signals of C-21 disappeared, while an additional quaternary carbon signal appeared and two proton signals of H-16 (δ 5.34) and H-22 (δ 6.29) were shifted to downfield. The IR spectrum indicated two absorption bands of carbonyl groups at 1660 and 1730 cm^{-1} in addition to the broad absorption band at 3450 cm^{-1}. These facts suggested that **2a** possessed an additional carbonyl group instead of C-21 methylene of **1a**. With the UV spectrum taken into consideration, one carbonyl group was supposed to exist as α,β-unsaturated carboxyl group. The other was esterified with 16-OH group. The lactone structure was supported by ^{1}H-detected heteronuclea multiple bond correlation (HMBC) spectrum ($J_{C\text{-}H}$ = 8.3 Hz), which indicated the correlation from 16-H to C-21 (δ_C 172.9), from 17-H (δ 3.15) to C-20 (δ_C 140.8), C-21, and C-22 (δ_C 131.9), and from olefinic H-22 to C-20 and C-21 (Fig. 1A). As the NOE enhancement was observed between 17-H and 22-H and between 18-H_3 (δ 0.81) and 22-H, the olefin structure of 20 and 22 position was cleared to be *Z*-form (Fig. 1B). Thus the structure of **2a** was estimated as indicated in Chart 1.

In order to confirm the structure, X-ray crystal structure analysis of the genin of **2a** (**2b**) was carried out. Compound **2b**[4)] was prepared from gitoxigenin (**1b**)[5)] in a similar manner. The ^{1}H- and ^{13}C-NMR spectral data of **2b** were almost the same as those of **2a**, except for the signals due to the sugar moieties. As shown in Fig. 2, the structure of **2b** was unambiguously clarified[6)], and at the same time, the structure of **2a** was established.

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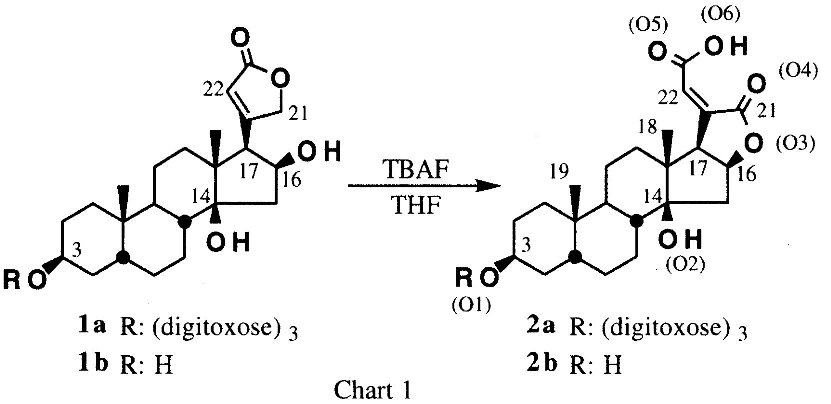

1a R: (digitoxose) $_3$
1b R: H

2a R: (digitoxose) $_3$
2b R: H

Chart 1

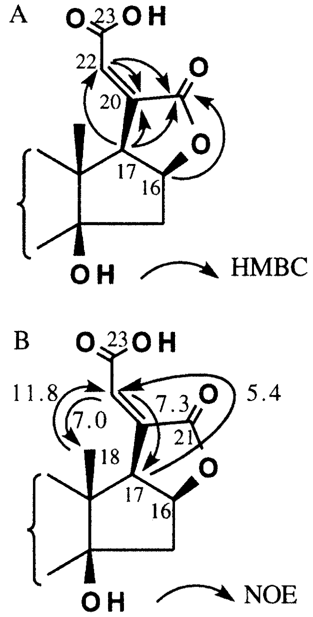

Fig. 1. ^{1}H-^{13}C Long-Range Correlations in the HMBC Spectrum (A) and NOE Interactions in Differential NOE Experiments (B) of **2a**

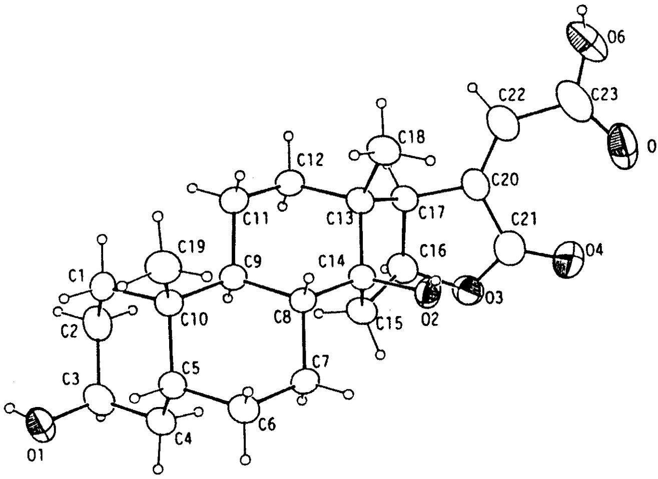

Fig. 2. ORTEP Structure of **2b**

Table 1. Positional Parameters and Their Estimated Standard Deviations

Atom	x	y	z	Beq(A2)[a]	Atom	x	y	z	Beq(A2)[a]
O1	0.0872(5)	0.000	0.6221(3)	3.34(7)	C11	0.3836(6)	0.3199(3)	0.7558(5)	2.70(9)
O2	-0.1160(4)	0.4687(3)	0.6519(3)	2.63(6)	C12	0.3558(6)	0.3812(3)	0.8600(4)	2.62(9)
O3	-0.1873(5)	0.5037(3)	0.9234(3)	3.32(7)	C13	0.2085(6)	0.4467(3)	0.7988(4)	2.25(8)
O4	-0.2451(5)	0.6293(3)	0.8644(4)	4.51(9)	C14	0.0122(6)	0.4080(3)	0.7225(4)	2.14(8)
O5	-0.0265(7)	0.7497(3)	0.8790(4)	5.0(1)	C15	-0.0800(6)	0.3810(3)	0.8377(4)	2.56(9)
O6	0.2899(7)	0.7706(3)	0.9297(4)	5.4(1)	C16	-0.0292(7)	0.4450(3)	0.9451(4)	2.8(1)
C1	0.3453(6)	0.1433(3)	0.6704(5)	2.83(9)	C17	0.1528(6)	0.4896(3)	0.9230(4)	2.43(9)
C2	0.2661(7)	0.1004(3)	0.7787(5)	3.1(1)	C18	0.2969(7)	0.5019(3)	0.7094(5)	2.9(1)
C3	0.0681(7)	0.0627(3)	0.7146(4)	2.9(1)	C19	0.2991(7)	0.2376(4)	0.4763(5)	3.1(1)
C4	-0.0731(6)	0.1240(3)	0.6360(5)	2.8(1)	C20	0.0849(7)	0.5743(3)	0.9055(4)	2.72(9)
C5	0.0074(6)	0.1717(3)	0.5314(4)	2.27(8)	C21	-0.1315(7)	0.5739(4)	0.8928(5)	3.3(1)
C6	-0.1414(6)	0.2347(3)	0.4622(4)	2.68(9)	C22	0.2021(8)	0.6375(3)	0.9138(5)	3.4(1)
C7	-0.1614(6)	0.3019(3)	0.5600(4)	2.52(9)	C23	0.155(1)	0.7244(4)	0.9071(5)	4.0(1)
C8	0.0370(6)	0.3413(3)	0.6228(4)	2.00(8)	O25[b]	0.6313(5)	0.4014(3)	0.2148(4)	4.04(9)
C9	0.1898(6)	0.2786(3)	0.6918(4)	2.16(8)	C26[b]	0.4571(7)	0.3762(4)	0.2523(5)	3.5(1)
C10	0.2106(6)	0.2093(3)	0.5940(4)	2.21(8)	C27[b]	0.3924(9)	0.4400(4)	0.3336(7)	5.0(2)

a Anisotropically refined atoms are given in the form of the isotropic equivalent displacement parameter defined as: (4/3) x [a^2 x B(1,1) + b^2 x B(2,2) + c^2 x B(3,3) + ab(cos γ) x B(1,2) + ac(cos β) x B(1,3) + bc(cos α) x B(2,3)]. b Atoms of ethanol.

Since compounds **2a**-**b** must be produced by the oxidation of **1a**-**b** in the presence of either tetrabutylammonium ion or fluoride ion, **1b** was treated with tetra-*n*-butylammonium bromide (TBAB), tetra-*n*-butylammonium chloride (TBAC), CsF, and KF. The reaction never proceeded in the presence of TBAB and TBAC. The reaction with CsF gave **2b** in high yield. Although the reaction with KF proceeded very slowly, the addition of 18-crown-6 to the mixture enhanced the reaction speed. These facts indicated that fluoride ion must be deeply involved in the oxidation. In order to confirm what is the oxidant, **1b** was treated with TBAF under argon or oxygen atmosphere.[7] The reaction under oxygen was completed within 30 min to give **2b**, while that under argon for 24 h afforded a trace amount of **2b**, which was probably yielded in the reaction of **1b** with trace oxygen remaining in the reaction system, and another major product as yet unidentified. Although oxidations using fluoride ion and oxygen were reported, the reactions were essentially the photooxidation which was performed by use of strong visible-light and dye-sensitizer in the oxygen-purged well.[8] Compounds **1a-b** were converted to **2a-b** in air and in the presence of only TBAF, CsF, or KF+18-crown-6, and the reaction proceeded also in the dark. An investigation on the mechanism of this unusual oxidative transformation is now in progress.

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2) TBAF used for the reaction was purchased from Wako Pure Chemical Industries Ltd. (Osaka, Japan) as trihydrate, Tokyo Kasei Kogyo Co., Ltd. (Tokyo, Japan) as 1 M THF solution, and Aldrich Co. Inc. (Milwaukee, U.S.A.) as 1 M THF solution.
3) *Data for* **2a**: colourless crystalline powder. m.p. 169-171°C (EtOH-diisopropyl ether). $[\alpha]_D^{25}$ -21.4°(*c*=0.5, MeOH). IR (KBr, cm^{-1}): 3450 (OH), 1730 (CO), 1660 (CO). UV (λ_{max}, MeOH) nm (ε): 232.7 (7.0×10^3). ^1H-NMR ($CDCl_3$, 500 MHz, δ): 3.15 (1H, dd, *J*=2.4, 6.7 Hz, 17-H), 4.02 (1H, br s, 3-H), 4.13 (1H, dd, *J*=3.1, 6.1 Hz, 3'''-H), 4.24 (2H, dd, *J*=3.1, 8.0 Hz, 3',3''-H), 4.89 (3H, m, 1', 1'', 1'''-H), 5.34 (1H, dd, *J*=6.7, 6.7 Hz, 16-H), 6.29 (1H, d, *J*=2.4 Hz, 22-H). ^{13}C-NMR ($CDCl_3$, 125 MHz, δ): 56.7 (C-17), 84.3 (C-16), 131.9 (C-22), 140.8 (C-20), 163.6 (C-23), 172.9 (C-21). FAB-MS (*m/z*) 795 $(M+H)^+$, 817 $(M+Na)^+$. Anal. Calcd for. $C_{41}H_{62}O_{15}$: C, 61.93; H, 7.87. Found: C, 62.08; H, 7.94.
4) *Data for* **2b**: colourless plates. m.p. 252-253°C (EtOH-diisopropyl ether). $[\alpha]_D^{25}$ -38.8° (*c*=0.5, MeOH). IR (KBr, cm^{-1}): 3450 (OH), 1720 (CO), 1660 (CO). ^1H-NMR ($CDCl_3$, 270 MHz, δ): 0.97 (3H, s, 19-CH_3), 1.07 (3H, s, 18-CH_3), 3.14 (1H, dd, *J*=2.2, 6.9 Hz, 17-H), 4.01 (1H, br s, 3-H), 5.32 (1H, dd, *J*=6.6, 6.9 Hz, 16-H), 6.27 (1H, d, *J*=2.2 Hz, 22-H). FAB-MS (m/z) 405 $(M+H)^+$, 427 $(M+Na)^+$. Anal. Calcd for. $C_{23}H_{32}O_6$: C, 68.28; H, 7.98. Found: C, 68.08; H, 7.94.
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6) *Crystal data for* **2b**: $C_{23}H_{32}O_6 \cdot C_2H_6O$, monoclinic, a = 7.049(1), b = 16.861(2), c = 10.088(1) Å, β = 103.81(1)°, V = 1164(3) $Å^3$, Z = 2, space group $P2_1$, Dc = 1.285 gcm^{-1}, μ (Mo-Kα) = 0.9 cm^{-1}. Data were collected on an Enraf-Nonius CAD4 diffractometer with monochromated Mo-Kα radiation, and corrected for Lorentz-polarization. No absorption correction was made. Full-matrix least-squares refinement of 288 parameters gave R = 0.045 for 2249 observed reflections. Atomic coordinates, bond lengths and angles, and thermal parameters have been deposited at the Cambridge Crystallographic Data Centre.
7) In each case, 100 mg of **1b** was treated with 5 eq of TBAF in 8 ml of freshly distilled THF at r.t..
8) a) Wasserman H. H., Pickett J. E., *J. Am. Chem. Soc.*, **104**, 4695-4596 (1982); b) *idem*, *Tetrahedron*, **41**, 2155-2162 (1985).

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