Conversion of Spirostane and Solanidane into Pregnane (1) Introduction of Oxygen Function into C-23

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A spirosolane derivative possessing a hydroxyl group at C-23, esculeogenin A, a sapogenol of tomato saponin, was found to be easily converted into the corresponding pregnane derivative by refluxing with aqueous pyridine. Therefore, introduction of a hydroxyl group into the C-23 of diosgenin (as representative of spirostane derivatives) and solasodine (as representative of spirosolane derivatives) was attempted by the reaction of $NaNO_2$–$BF_3 \cdot Et_2O$. In diosgenin, the objective compound was obtained by the reaction in AcOH. However, in solasodine, we obtained a 23-nitroso derivative by the reaction in AcOH and 23,24-bisnorcholanic acid 22-16 lactone, or vespertilin, in AcOH and $CHCl_3$.

Key words spirost-23-hydroxylation; diosgenin; solasodine; nitroso derivative; bisnorcholanic acid

Nohara *et al.* found that the spirosolane sapogenol of tomato saponin, (5*α*,22*S*,23*S*,25*S*)-3*β*,23,27-trihydroxyspirosolane, or esculeogenin A (**1**),[1] was converted to 3*β*,16*β*-dihydroxy-5*α*-pregna-20-one (**2**) by refluxing with diluted aqueous pyridine.[2,3] Its reaction was considered to be triggered by the existence of the 23-hydroxyl group on the spirosolane skeleton. Preparation of steroidal hormones depends mainly upon the Marker degradation.[4] When the 23-hydroxyspirostane, spirosolane, and tomatidane compounds are prepared, the pregnane compound might be derived from them by a similar simple reaction. Therefore, we first tried to introduce an oxygen group at C-23 in them by the reported method.[5,6] In this report, diosgenin (a representative spirostane) and solanidine (a representative spirosolane derivative) were used as crude original substances.

First, to a suspension of diosgenin (**3**, 1 g) in acetic acid (40 mL), $NaNO_2$ (3.0 g) and $BF_3 \cdot Et_2O$ in ether (4.5 mL) were added gradually with stirring for 1 h at rt. After 1.5 h, the reaction mixture was treated in the usual manner to give a product that was chromatographed on silica gel with *n*-hexane : acetone = 4 : 1 → 3 : 1 to give compounds **4** (349 mg, 33.8%) and **5** (137 mg, 11.9%).

Compound **4** consisted of colorless needles (mp 199—202°C; $[\alpha]_D$ −84.4° [$CHCl_3$]). High-resolution fast atom bombardment mass spectrometry (HR-FAB-MS) revealed a molecular ion at *m/z* 451.2826, indicating a molecular formula of $C_{27}H_{40}O_4$+Na (Calcd: 451.2824), and a carbonyl group appeared at 1732 cm^{-1} in the IR spectrum.

The ^{1}H-NMR spectrum exhibited two tertiary methyl groups at δ 0.79 (3H, s, H_3-18) and 1.02 (3H, s, H_3-19); two secondary methyl groups at δ 0.93 (6H, d, J=6.7 Hz, H_3-21, H_3-27); two methylene protons at δ 3.59 and 3.78 (1H, m and

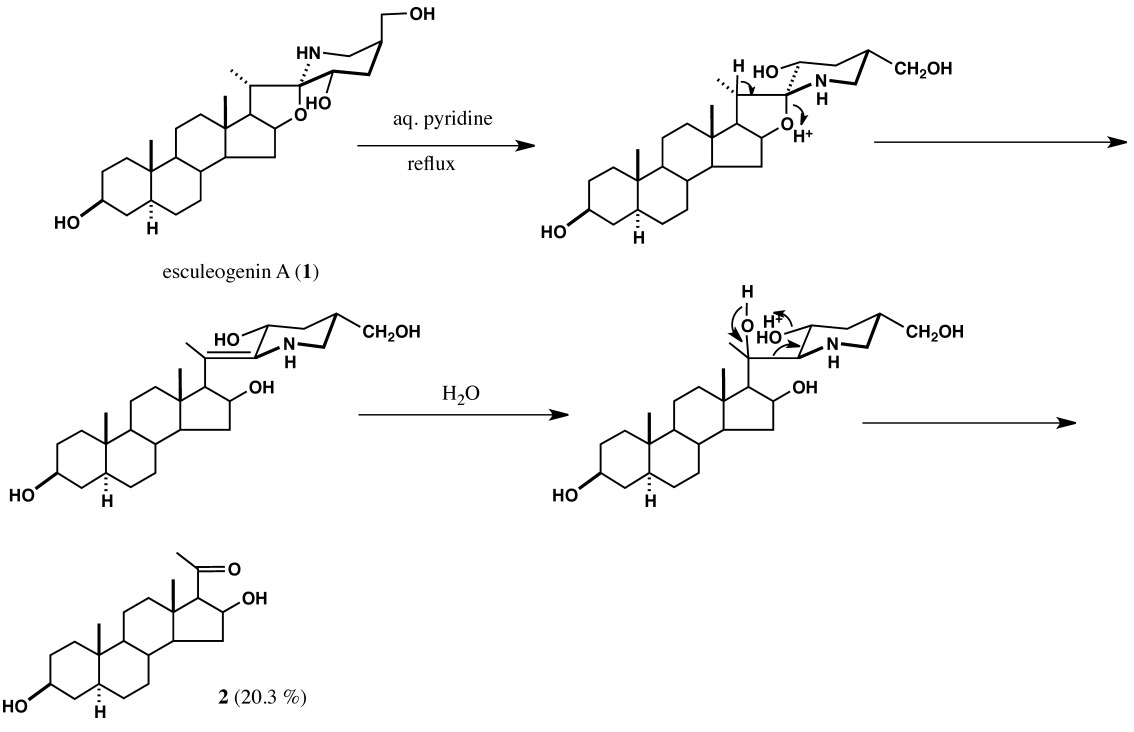

Chart 1. Mechanism for Conversion of Esculeogenin A (**1**) into Pregnane Derivative (**2**)

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1H, t-like, J=11.3 Hz, Heq-26, Hax-26, respectively); two oxygen-bearing methine protons at δ 3.51 (1H, m, H-3) and 4.61 (1H, dd, J=6.8, 15.7 Hz, H-16); and one olefinic proton at δ 5.34 (1H, m, H-6). In the ^{13}C-NMR spectrum (in $CDCl_3$), one carbonyl carbon newly occurred at δ 201.8, which correlated with H-20 at δ 2.89 (1H, m). Therefore, the structure of compound **4** was characterized as 23-oxodiosgenin.[5,6)]

For compound **5**, an amorphous powder having $[\alpha]_D$ −99.6° ($CHCl_3$), HR-FAB-MS revealed a molecular ion at m/z 540.2935, indicating a molecular formula of $C_{29}H_{43}NO_7$+Na (Calcd: 540.2937). A carbonyl group appeared at 1730 cm^{-1} in the IR spectrum. The ^{1}H-NMR spectrum showed signals due to H_3-18 (3H, s) at δ 0.76, H_3-19 (3H, s) at δ 1.02, H_3-27 (3H, d, J=4.3 Hz) at δ 0.93, and H_3-21 (3H, d, J=4.3 Hz) at δ 0.94; one acetyl signal (3H, s) at δ 2.17; H-3 (1H, m) at δ 3.77; Heq-26 (1H, t-like, J=11.2 Hz) at δ 3.77; and H-16 (1H, m) at δ 4.51. The ^{13}C-NMR spectrum revealed the presence of one hydroxyimino group at δ 178.0, together with one carbonyl carbon at δ 201.6 and one acetyl carbonyl carbon at δ 169.5. Furthermore, since heteronuclear multiple bond coherence (HMBC) analysis indicated the existence of a hydroxyl group at C-5, the presence of an imino group at C-6 is suggested. The carbonyl group is located at C-23, as in compound **4**. The acetyl group was estimated to be bound to the hydroxyimino group because the H-3 was not down shifted. Therefore, the structure of compound **5** was characterized as 3β,5-dihydroxy-6-acetoxyimino-23-oxo-(25R)-spirostane.

Next, compound **4** (250 mg) was reduced with $NaBH_4$ (200 mg) in MeOH (6 mL) for 30 min at rt. After a small amount of AcOH was added, its reaction mixture was evaporated to give a residue, which was then chromatographed on silica gel with n-hexane–acetone = 4 : 1 → 3 : 1 to provide compound **6** (160 mg, 64.0%). Compound **6** appeared as colorless needles (mp 212—214°C; $[\alpha]_D$ −112.4° [$CHCl_3$]). HR-FAB-MS revealed a molecular ion at m/z 494.2877, indicating a molecular formula of $C_{27}H_{42}O_4$+Na (Calcd: 494.2880), and a hydroxyl group appeared at 3550 cm^{-1} in the IR spectrum. The ^{1}H-NMR spectrum (in pyridine-d_5) displayed signals due to H_3-21 (d, J=6.7 Hz) at δ 0.73, H_3-18 (s) at δ 0.99, H_3-19 (s) at δ 1.02, H_3-27 (d, J=6.7 Hz) at δ 1.18, Hax-26 (t-like, J=11.0 Hz) at δ 3.46, Heq-26 (m) at δ 3.54, H-3 (m) at δ 3.82, H-16 (dd, J=7.6, 15.6 Hz) at δ 4.64, H-6 (m) at δ 5.36, and H-23 (dd, J=4.5, 11.3 Hz) at δ 3.82. Their coupling constants indicated that the hydroxyl group was oriented equatorially (S). In the ^{13}C-NMR spectrum (in pyridine-d_5), one hydroxyl carbon newly occurred at δ 67.4. The structure of compound **6** was characterized as (22S,23S,25R)-3β,23-dihydroxy-spirost-5-ene,23S-hydroxydiosgenin.

Next, to a suspension of solasodine (**7**, 307 mg) in acetic acid (18 mL), $NaNO_2$ (924 mg) and $BF_3 \cdot Et_2O$ (4.5 mL) were added gradually with stirring for 1 h at rt. After 1.5 h, the reaction mixture was treated in the usual manner to give a product that was chromatographed on silica gel with n-hexane–acetone = 4 : 1 → 3 : 1 to give compound **8** (184 mg, 56.2%) as a major product.

Compound **8** exhibited $[\alpha]_D$ −98.2° (MeOH) and a molecular formula of $C_{27}H_{42}N_2O_3$+Na at m/z 465.3090 (Calcd: 465.3093) in positive HR-FAB-MS. The ^{1}H-NMR spectrum showed two tertiary groups at δ 0.82 (3H, s, H_3-18) and 1.00 (3H, s, H_3-19); two secondary methyl groups at δ 0.90 (3H, d, J=6.8 Hz, H_3-27) and 1.17 (3H, d, J=7.5 Hz, H_3-21); two oxygen-bearing methine protons at δ 3.50 (1H, m, Hax-26) and 3.97 (1H, m, Heq-26); one olefinic proton at δ 5.31 (1H, m, H-6), along with a down-shifted proton at δ 4.66 (1H, dd, J=3.6, 13.8 Hz), which was assigned to H-23 because HMBC was observed between H-23 and C-22 at δ 102.0; and H-25 at δ 44.1. The ^{13}C-NMR spectrum of **7** displayed a total of 27 carbon signals, including four methyl carbons at δ 15.0, 16.4, 19.1, and 19.5; two oxygen-bearing carbons at δ 74.8 and 83.1; one hemiacetal carbon at δ 102.0; one double bond at δ 121.4 and 140.9; and one nitrogen-bearing methine carbon at δ 44.1. The chemical shifts of H-23 in the ^{1}H- and ^{13}C-NMR spectra indicated that an imino group was attached to C-23; its configuration was determined to be equatorial by its coupling constants.

Next, to increase the solubility of the reaction mixture, a small amount of $CHCl_3$ was added to the reaction. To a solution of solasodine (**7**, 304 mg) in AcOH (14 mL) and $CHCl_3$ (5 mL), $NaNO_2$ (890 mg) and BF_3 ethelate (4.5 mL) were added gradually and left standing at rt for 2 h. The usual treatment yielded compound **9** (120.5 mg, 48.2%) having $[\alpha]_D$ −82.6° (MeOH) and a molecular formula of $C_{22}H_{32}O_3$+Na at m/z 367.2252 (Calcd: 367.2249), as revealed by positive HR-FAB-MS. The ^{1}H-NMR spectrum of **9** showed two tertiary methyl groups at δ 0.90 (3H, s, H_3-18) and 0.95 (3H, s, H_3-19); one secondary methyl group at δ 1.23 (3H, d, J=7.5 Hz, H_3-21); two oxygen-bearing methine protons at δ 3.46 (1H, m, H-3) and 4.89 (1H, m, H-16); and one olefinic proton at δ 5.31 (1H, m, H-6). The ^{13}C-NMR spectrum displayed a total of 22 carbon signals including two oxygen-bearing carbons at δ 71.7

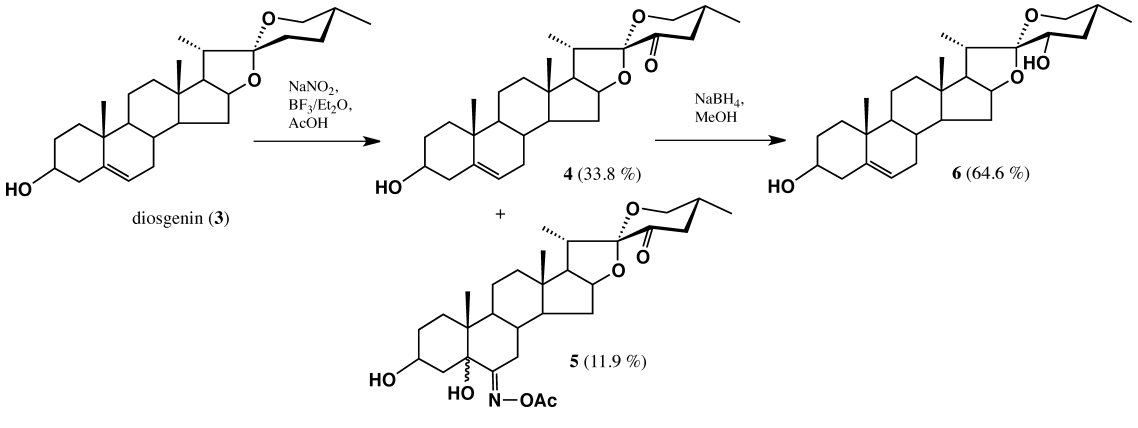

Chart 2. Introduction of Oxygen Function into C-23

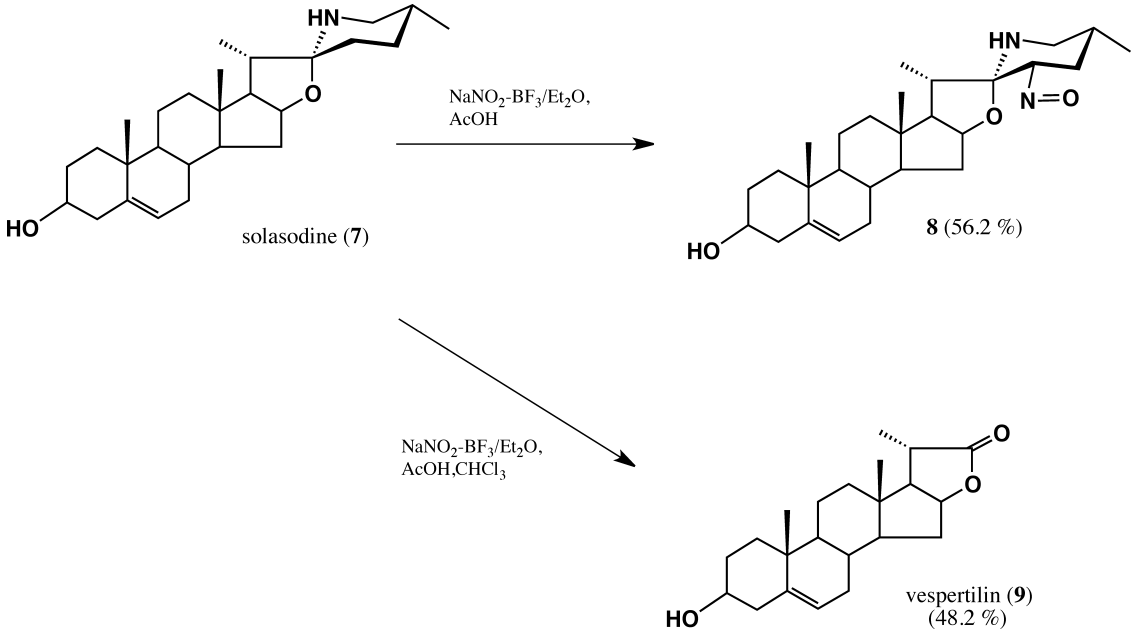

Chart 3. Reaction of Solasodine

and 82.9; one trisubstituted double bond at δ 121.3 and 141.0; and one lactone carbonyl carbon at δ 181.5. This compound was coincident with vespertilin.[7)]

Consequently, in the reaction of solasodine with $NaNO_2$ and BF_3, the results when only AcOH was used in suspension state as a solvent differed from those obtained when AcOH and $CHCl_3$ were used in solution state, thus they would depend on the solubility.

We are currently preparing tomatidine produced from the aerial part of *Solanum lycopersicum*.

Experimental

General Procedure Optical rotations were measured with a JASCO P-1020 (l=0.5) automatic digital polarimeter. FAB-MS were obtained with a glycerol matrix in the positive ion mode using a JEOL JMS-DX300 and a JMS-DX 303 HF spectrometer. The ^{1}H- and ^{13}C-NMR spectra were measured in pyridine-d_5 with JEOL α-500 spectrometer, and chemical shifts are given on a δ (ppm) scale with tetramethylsilane (TMS) as the internal standard. Column chromatographies were carried out on a Diaion HP-20 (Mitsubishi Chemical Ind.), and silica gel 60 (230—400 mesh, Merck). TLC was performed on silica gel plates (Kieselgel 60 F_{254}, Merck) and RP C_{18} silica gel plates (Merck). The spots on TLC were visualized by UV light (254/366 nm) and sprayed with 10% H_2SO_4, followed by heating.

Reaction of Diosgenin with $NaNO_2$ and BF_3-Ethelate in AcOH Firstly, to a solution of diosgenin (**3**, 1 g) in acetic acid (40 mL), $NaNO_2$ (3.0 g) and $BF_3 \cdot Et_2O$ (4.5 mL) was gradually added with stirring for 1 h at rt. After 1.5 h, the reaction mixture was treated in an usual manner to give the product which was chromatographed on silica gel with *n*-hexane–acetone = 4 : 1 → 3 : 1 to give compounds **4** (349 mg, 33.8%) and **5** (137 mg, 11.9%).

Compound **4**, colorless needles, mp 199—202°C, $[\alpha]_D$ −84.4° (c=0.5, $CHCl_3$), IR ν_{max} 1732 cm^{-1} (carbonyl carbon). HR-FAB-MS (*m/z*): 451.2826 [$C_{27}H_{40}O_4$+Na, Calcd: 451.2824]. ^{1}H-NMR spectrum (in $CDCl_3$) δ: 0.79 (3H, s, H_3-18), 1.02 (3H, s, H_3-19), 0.93 (6H, d, J=6.7 Hz, H_3-21, H_3-27), 3.59, 3.78 (1H, m, and 1H, t-like, J=11.3 Hz, Heq-26 and Hax-26, respectively), 3.51 (1H, m, H-3), 4.61 (1H, dd, J=6.8, 15.7 Hz, H-16), 5.34 (1H, m, H-6). ^{13}C-NMR spectrum (in $CDCl_3$): C-1—C-27, δ 37.3, 29.7, 71.8, 43.5, 141.0, 121.2, 31.7, 31.5, 50.1, 36.7, 20.8, 39.6, 40.8, 56.7, 31.7, 83.4, 61.8, 16.0, 19.4, 34.8, 14.4, 109.9, 201.8, 43.9, 35.8, 65.7, 17.1.

Compound **5**, an amorphous powder, $[\alpha]_D$ −99.6° (c=0.5, $CHCl_3$), IR ν_{max} 1730 cm^{-1} (carbonyl carbon). HR-FAB-MS (*m/z*): 540.2935 ($C_{29}H_{43}NO_7$+Na, Calcd: 540.2937). ^{1}H-NMR spectrum ($CDCl_3$) δ: 0.76 (3H, s, H_3-18), 1.02 (3H, s, H_3-19), 0.93 (3H, d, J=4.3 Hz, H_3-27), 0.94 (3H, d, J=4.3 Hz, H_3-21), 2.17 (3H, s, acetyl signal), 3.77 (1H, m, H-3), 3.77 (1H, t-like, J=11.2 Hz, Heq-26), 4.51 (1H, m, H-16). ^{13}C-NMR spectrum ($CDCl_3$): C-1—C-27, δ 38.9, 29.8, 68.0, 45.6, 87.1, 178.0, 33.7, 34.5, 55.1, 36.9, 21.2, 39.7, 42.2, 56.7, 32.7, 83.5, 61.9, 15.8, 17.3, 34.6, 14.6, 109.9, 201.6, 45.2, 35.9, 65.6, 17.1, acetyl group, 169.5, 18.2.

Reaction of Compound 4 with $NaBH_4$ Compound **4** (250 mg) was reduced with $NaBH_4$ (200 mg) in MeOH (6 mL) for 30 min at rt. After addition of a small amount of AcOH, its reaction mixture was evaporated to give a residue, which was then chromatographed on silica gel with *n*-hexane–acetone = 4 : 1—3 : 1 to provide compound **6** (160 mg, 64.0%).

Compound **6** exhibited colorless needles, mp 212—214°C, $[\alpha]_D$ −112.4° (c=0.5, $CHCl_3$), IR ν_{max} 3550 cm^{-1} (hydroxyl). HR-FAB-MS (*m/z*): 467.3017 ($C_{27}H_{42}NO_4$+Na, Calcd: 467.3012). ^{1}H-NMR spectrum (in pyridine-d_5) δ: 0.73 (3H, d, J=6.7 Hz, H_3-21), 0.99 (3H, s, H_3-18), 1.02 (3H, s, H_3-19), 1.18 (3H, d, J=6.7 Hz, H_3-27), 3.46 (1H, t-like, J=11.0 Hz, Hax-26), 3.54 (1H, m, Heq-26), 3.82 (1H, m, H-3), 3.82 (1H, dd, J=4.5, 11.3 Hz, H-23), 4.64 (1H, dd, J=7.6, 15.6 Hz, H-16), 5.36 (1H, m, H-6). ^{13}C-NMR spectrum (in pyridine-d_5): C-1—C-27, δ 37.8, 29.8, 71.2, 43.4, 142.0, 120.9, 33.7, 31.7, 50.5, 37.0, 21.2, 40.3, 41.0, 56.8, 32.7, 81.6, 62.5, 35.8, 14.7, 111.7, 67.4, 38.8, 31.7, 65.2, 16.9.

Reaction of Solasodine (7) with $NaNO_2$ and $BF_3 \cdot Et_2O$ in AcOH To a solution of solasodine (**7**, 307 mg) in acetic acid (18 mL), $NaNO_2$ (924 mg) and $BF_3 \cdot Et_2O$ (4.5 mL) was gradually added with stirring for 1 h at rt. After 1.5 h, the reaction mixture was treated in the usual manner to give the product which was chromatographed on silica gel with *n*-hexane–acetone = 4 : 1 → 3 : 1 to give compound **8** (184 mg, 56.2%) as a major product.

Compound **8**, an amorphous powder, $[\alpha]_D$ −98.2° (*c*=1.0, MeOH). HR-positive FAB-MS (*m/z*): 465.3090 (Calcd for $C_{27}H_{42}N_2O_3$+Na: 465.3093). ^{1}H-NMR spectrum ($CDCl_3$) δ: 0.82 (3H, s, H_3-18), 0.90 (3H, d, *J*=6.8 Hz, H_3-27), 1.00 (3H, s, H_3-19), 1.17 (3H, d, *J*=7.5 Hz, H_3-21), 3.50 (1H, m, Hax-26), 3.50 (1H, m, H-3), 3.97 (1H, m, Heq-26), 4.66 (1H, dd, *J*=3.6, 13.8 Hz, H-23), 5.31 (1H, m, H-6). ^{13}C-NMR spectrum ($CDCl_3$) C-1—C-27, δ 37.2, 29.7, 71.8, 42.3, 140.9, 121.4, 31.7, 31.5, 50.1, 36.7, 21.0, 39.8, 40.4, 56.5, 32.1, 83.1, 61.6, 16.4, 19.3, 44.1, 15.0, 102.1, 44.1, 42.3, 44.1, 42.3, 19.0.

Reaction of Solasodine (7) with $NaNO_2$ and $BF_3 \cdot Et_2O$ in AcOH and $CHCl_3$ To a solution of solasodine (**7**, 304 mg) in AcOH (14 mL) and $CHCl_3$ (5 mL), $NaNO_2$ (890 mg) and $BF_3 \cdot Et_2O$ (4.5 mL) was added gradually and reft stand at rt for 2 h. Usual work up gave compound **9** (120.5 mg, 48.2%), $[\alpha]_D$ −82.6° (*c*=1.0, MeOH). HR-positive FAB-MS (*m/z*): 361.2252 (Calcd for, $C_{22}H_{32}O_3$+Na: 361.2249). ^{1}H-NMR spectrum ($CDCl_3$) δ: 0.70 (3H, s, H_3-18), 0.95 (3H, s, H_3-19), 1.23 (3H, d, *J*=7.5 Hz, H_3-21), 3.46 (1H, m, H-3), 4.89 (1H, m, H-16), 5.27 (1H, m, H-6). ^{13}C-NMR spectrum ($CDCl_3$): C-1—C-22, δ 37.3, 32.0, 71.7, 42.3, 141.0, 121.0, 31.6, 31.3, 50.2, 36.7, 20.4, 38.3, 41.5, 59.0, 36.5, 82.9, 61.6, 18.1, 19.5, 54.9, 13.8, 181.5.

References and Notes

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