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152. Michiya Kimura, Masahiko Tohma, and Itsuo Yoshizawa : Constituents of Convallaria. Ⅶ.*1 Structure of Convallagenin-A.

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Convallagenin-A (I) was isolated from the flowers of *Convallaria keisukei* Miq. and was shown to be a new 25L,1,3,5-trihydroxyspirostane due to the conversion of it to $\Delta^{1,4}$-tigogenone (XII). All of the hydroxyl groups in I were indicated to have β-configuration as a result of forming the 1,5-carbonate (XIII), the 3,5-carbonate (XIX), and the orthoester (XX). The chemical structure of convallagenin-A (I) was thus elucidated as 25L,5β-spirostan-1β,3β,5β-triol.

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In the previous paper of this series,[1] the authors reported on a steroidal sapogenin, convallagenin-A (I), $C_{27}H_{44}O_5$, m.p. 268~269°, $[\alpha]_D$ −28.0° ($CHCl_3$), obtained by the hydrolysis of convallasaponin-A which was isolated from the flowers of *Convallaria keisukei* Miq., Japanese lily of the valley (SUZURAN). The infrared absorption spectrum, ν_{max} : 982, 922, 897 and 852 cm^{-1}, indicated this sapogenin to be in the 25L-series.[2] Acetylation of I gave monoacetate (II), $C_{29}H_{46}O_6$, m.p. 190~191°, and diacetate (III), $C_{31}H_{48}O_7$, m.p. 208~210°, both still showing hydroxyl bands in the infrared spectra. Thus, I was assumed to be a new 25L-trihydroxysapogenin. The present paper deals with the structure elucidation of I as 25L,5β-spirostan-1β,3β,5β-triol from the following evidences.

Kutney[3] reported that nuclear magnetic resonance (NMR) spectroscopy can provide an immediate information regarding the stereochemistry of C-25 in steroidal sapogenins.

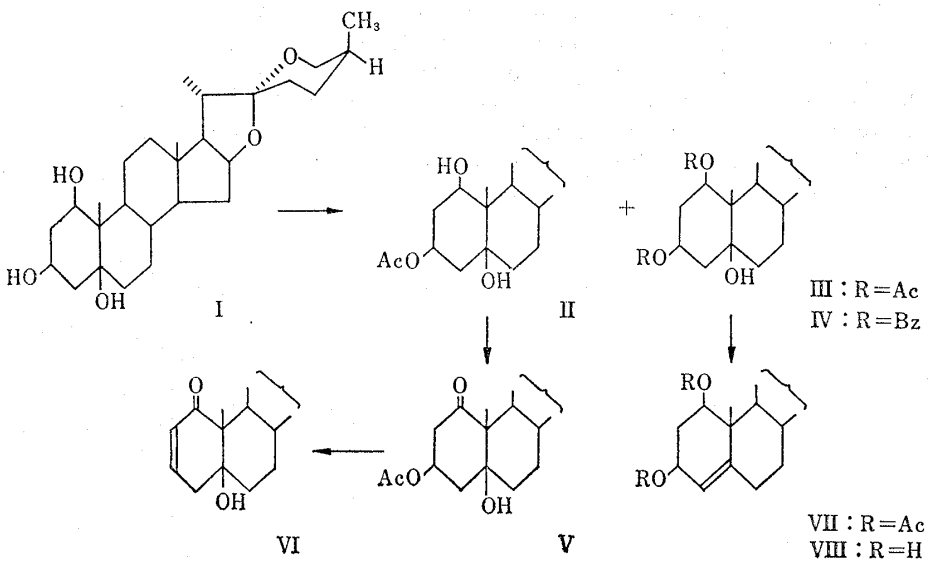

Chart 1.

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1) M. Kimura, M. Tohma, I. Yoshizawa : This Bulletin, **14**, 50 (1966).

2) M. E. Wall, C. R. Eddy, M. L. McClennan, M. E. Klumpp : Anal. Chem., **24**, 1337 (1952); C. R. Eddy, M. E. Wall, M. K. Scott : *Ibid.*, **25**, 266 (1953).

3) J. P. Kutney : Steroids, **2**, 225 (1963).

The same spectra as well as the infrared spectra of I and its acetates represented typical indications of the 25L-series; the 27-axial methyl signal (τ 8.97) in the former spectra was well separated from that of the 18-methyl at higher field (Table I).

As described previously,[1] acetylation of I by heating on a steam bath in acetic anhydride-pyridine gave a mixture of II and III in a ratio of 1.7:1. When the reaction mixture was allowed to stand at room temperature or was kept in a diluted chloroform solution under lower temperature, the ratios were shown as 4:1 or 16:1, respectively. Benzoylation of I by usual method, however, gave only a dibenzoate (IV). The monoacetate (II) was oxidized with chromium trioxide-sulfuric acid in acetone to give a ketone (V), still showing a hydroxyl band at 3585 cm^{-1}. The diacetate (III) in thionyl-chloride-pyridine mixture was readily converted to the anhydro compound (VII) which showed an isolated C=C band at 1657 cm^{-1} but no hydroxyl absorption in the infrared spectrum. The compound (VII) gave an unsaturated diol (VIII), $C_{27}H_{42}O_4$, by alkaline hydrolysis, showing the absorption bands at 3600~3200, 3060 and 1658 cm^{-1}. The structural assignment of VII was further confirmed by the NMR band at τ4.70 as a sharp singlet which is attributed to one olefinic proton at C-4. These results indicated that I contains one tertiary and two secondary hydroxyl groups, one of which is readily acetylable, the other less reactive.

Table I. Chemical Shifts of 18- and 19-Methyl Groups in Pyridine

Convallagenin-A monoacetate	19·CH_3 (τ)	Diff.[a] (p.p.m.)	18·CH_3 (τ)	Diff.[a] (p.p.m.)
Convallagenin-A (I)	8.41	−0.67	9.14	−0.01
Convallagenin-A monoacetate (II)	8.46	−0.62	9.13	−0.02
Convallagenin-A diacetate (III)	8.78	−0.30	9.17	+0.02
Convallagenin-A-1-oxo-3-acetate (V)	8.47	−0.61	9.15	0.00
Convallagenin-A-Δ^4-diacetate (VII)	8.82	−0.26	9.14	−0.01
Convallagenin-A-1-deoxy-3-acetate (XVIII)	8.85	−0.23	9.15	0.00
5β-Spirostane[7]	9.08		9.15	

a) Difference in chemical shifts of the angular methyl group from that of the 5β-spirostane.

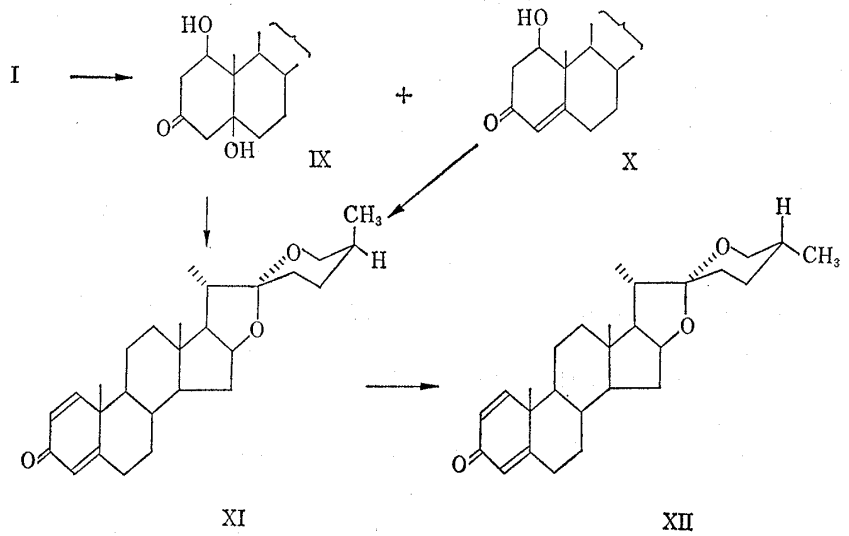


Chart 2.

Recently, in NMR studies of steroids, the substituent effects on the angular methyl shifts have been reported by several workers.[4~7] The additivity principle of the effects is thus held on these shifts, if no conformational change of the ring is caused by the substituents. Table I shows the chemical shifts of the angular 18- and 19-methyl groups of I as well as its derivatives in pyridine and the difference from these values of 5β-spirostane. These data showed that the 19-methyl signals are affected by their substituents and shifted to the lower field, contrary to those of the 18-methyl group. Therefore, all of the hydroxyl groups in I appeared to be located at neighboring positions of the 19-methyl group. With consideration on the acetylation described above, this may indicate I to be a 1,3,5-trihydroxy steroid, that was confirmed by the conversion of I to the 1,4-dien-3-one (XII) as described below. Selective oxidation of the 3-hydroxyl group in I with molecular oxygen and platinum catalyst,[8] followed by chromatography on alumina, afforded the 3-ketone (IX), ν_{max}: 1717 cm^{-1}, and a small amount of the 4-en-3-one (X), ν_{max}: 1667 and 1624 cm^{-1}, λ_{max}: 241 mμ (log ε 4.16). By refluxing with 10% methanolic potassium hydroxide or acetic acid, these ketones were readily converted to the expected dienone (XI) which showed maximum absorptions at 244.5 mμ (log ε 4.19) as well as at 1663, 1624 and 1605 cm^{-1} corresponding to the 1,4-dien-3-one.[9,10] The isomerization of the 25L-dienone (XI) to the 25D-series was carried out by refluxing with hydrochloric acid in 80% ethanol solution for 40 hours. The main product of the reaction was identified as the 25D-isomer (XII) by melting point on admixture with $\Delta^{1,4}$-tigogenone prepared from diosgenone,[10] by the ultraviolet as well as the infrared spectra and by Rf values on thin-layer chromatogram.

The optical rotatory dispersion curves of the 3-ketone (IX) and the 1-ketone (V) are shown in Fig. 1. Table II shows the signs and molecular amplitudes of the Cotton

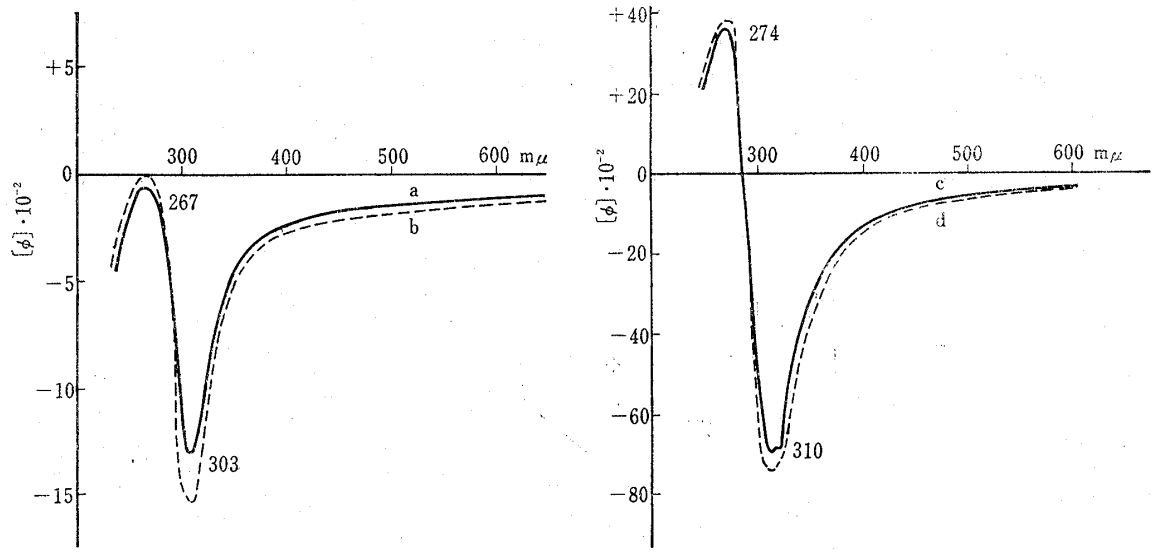


Fig. 1. Optical Rotatory Dispersion Curves of 3- and 1-Ketones (in Methanol)

a) Convallagenin-A-3-one (IX) *b*) Smilagenone
c) 3-Acetoxyconvallagenin-A-1-one (V) *d*) isorhodeasapogenone-1

4) R. F. Zürcher: Helv. Chim. Acta, **44**, 1380 (1961); **46**, 2054 (1963).
5) A. I. Cohen: Steroids, **3**, 243 (1964).
6) L. L. Smith: *Ibid.*, **4**, 395 (1964).
7) K. Tori, K. Aono: Ann. Repts. Shionogi Res. Lab., **14**, 136 (1964).
8) R. P. A. Sneeden, R. B. Turner: J. Am. Chem. Soc., **77**, 190 (1955).
9) D. Burn, B. Ellis, V. Petrow: J. Chem. Soc., **1958**, 795.
10) K. Takeda, T. Okanishi, K. Igarashi, A. Shimaoka: Tetrahedron, **15**, 183 (1961).

Table II. Optical Rotatory Dispersion (in Methanol)

	A/B-Junction	Mol. Amplitude	Ref.
3-Ketones			
Convallagenin-A-3-one (IX)	*cis*	− 12.8	
Smilagenone	*cis*	− 16.6	12)
1-Methoxyisorhodeasapogenone-3	*cis*	− 28	13)
Coprostanone-3	*cis*	− 27	11)
Cholestanone-3	*trans*	+ 65	11)
1-Ketones			
3-Acetoxyconvallagenin-A-1-one (V)	*cis*	−104.4	
Isorhodeasapogenone-1	*cis*	−113.4	
Methyl-1-oxo-5β-etianate	*cis*	−136	11)
Cholestanone-1	*trans*	plain	11)

effect curves for these compounds and the related ketones.[11~13] The A/B ring junction of V as well as IX may reasonably suggested as *cis*-configuration from the fact that the former has smaller and the latter has larger negative amplitudes. The skeleton of I is, therefore, the 25L,5β-spirostane, that represents the hydroxyl group at C-5 to be in the β-configuration. Treatment of the monoacetate (II) with phosgene in pyridine afforded a carbonate(XIII) in good yield. The infrared spectrum of the product showed absorption bands at 1737 and 1106 cm^{-1} as characteristic of the carbonate, but no hydroxyl band. This may reveal that the hardly acetylable 1-hydroxyl group formed a 1,3-diaxial glycol with the 5β-hydroxyl group, showing inevitably the β-orientation of the former.

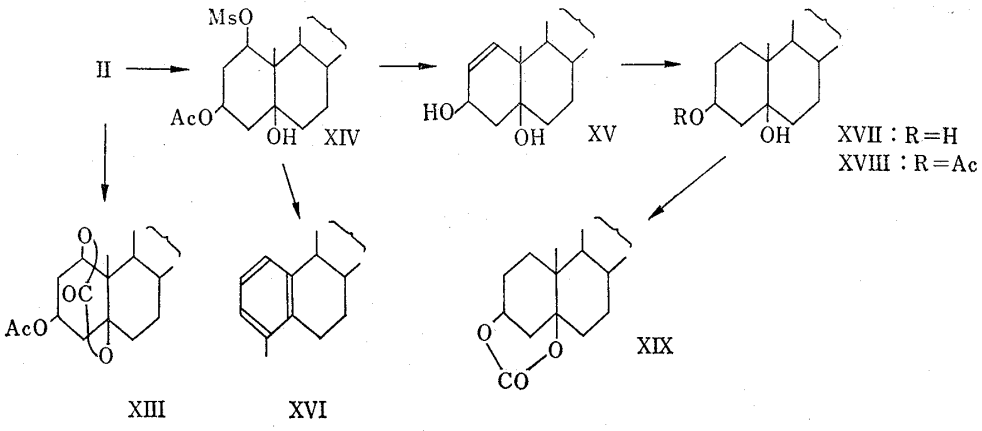
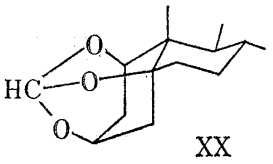


Chart 3.

Finally, the axial (β) configuration of the hydroxyl group at C-3 was suggested by the finding that the 3-acetoxy-1-one (V) readily gave an α,β-unsaturated ketone (VI) through the β-elimination of acetoxyl group with alkaline alumina in benzene; that was confirmed further by forming the 3,5-carbonate (XIX) from II as mentioned below. In order to eliminate the functional group at C-1, the pyridine solution of the methanesulfonate (XIV), ν_{max} : 1350 and 1167 cm^{-1}, was refluxed as usual, but the starting material was entirely recoverd. Nace's method[14] of the effective demesylation

11) M. Moffitt, R. B. Woodward, A. Moscowitz, W. Klyne, C. Djerassi : J. Am. Chem. Soc., **83**, 4013 (1961).
12) K. Takeda, H. Minato : Steroids, **1**, 345 (1963).
13) M. Kimura, M. Tohma, I. Yoshizawa : This Bulletin, **14**, 55 (1966).
14) H. R. Nace : J. Am. Chem. Soc., **81**, 5428 (1959).

by heating in dimethylsulfoxide solution was then attempted instead. An aromatic compound (XVI), however, was unexpectedly isolated from the solution as colorless crystalline leaflets. Takeda, *et al.*[10] reported that luvigenin, a steroidal sapogenin with an aromatic ring in the molecule, was isolated from *Methanarthecium luteo-viride* Maxim. and proved to be 25D, 19-nor-4-methylspirosta-1,3,5(10)-triene. The compound (XVI) thus obtained was assumed to be an 25L-isomer of luvigenin, since both compounds showed a close similarity in their physical properties as described in the experimental part. The demesylation was completed finally by refluxing XIV with lithium aluminum hydride in tetrahydrofuran or by allowing to stand with potassium *tert*-butoxide in dimethylsulfoxide to give the unsaturated 3,5-diol (XV) which showed yellow color with tetranitromethane and exhibited the absorption bands at 3050 as well as 1650 cm^{-1} due to the isolated double bond, and no sulfonyl absorption in the infrared spectrum. The saturated 3,5-diol (XVII) was obtained by the hydrogenation of XV on platinum catalyst in ethylacetate. The diol (XVII) gave two sharp hydroxyl bands at 3620 as well as 3540 cm^{-1}, and its acetate (XVIII) showed a hydroxyl (ν_{max} : 3600 cm^{-1}) and an acetoxyl band (ν_{max} : 1748 cm^{-1}) which was found at higher wave number region than those of ordinary acetates. This may indicate the intramolecular hydrogen bond between hydroxyl and acetoxyl groups, on which Dalton, *et al.*[15] studied by infrared spectrometry. The hydroxyl groups in XVII was, therefore, regarded to have the 1,3-diaxial configuration. The 3,5-carbonate (XIX), ν_{max} : 1740 and 1106 cm^{-1}, was obtained as expected, suggesting the β-orientation of the hydroxyl group on C-3.



XX

Chart 4.

From these results, it was concluded that the three hydroxyl groups of I are all considered to have β-configuration. Further evidence for this assignment was obtained from the formation of the orthoester (XX) with ethylorthoformate in anhydrous methanolic hydrogen chloride. The chemical structure of convallagenin-A (I) can, therefore, be elucidated as 25L,5β-spirostan-1β,3β,5β-triol. It seems to be noteworthy that I is one of the few 25L-trihydroxysapogenins such as neodigitogenin, 2α,3β,15β-triol[16] and reineckiagenin, 1β,3β,25-triol,[17] contrary to the 25D-series ordinarily isolated from natural plant sources.

Experimental

Melting points were determined on a micro hot-stage and are uncorrected. Ultraviolet spectra were recorded with a Hitachi EPS-2U and infrared spectra with a Koken-DS-301 Spectrophotometer. Optical rotatory dispersion curves were measured with a Nihon Bunko ORD-UV-5 Recording Spectrophotometer. Nuclear magnetic resonance spectra were recorded on a Varian A-60 spectrometer by using 5% solution containing $(CH_3)_4Si$ as an internal refference.

Acetylation of Convallagenin-A (I)——a) I (173 mg.) dissolved in a mixture of pyridine (2 ml.) and Ac_2O (2 ml.) was allowed to stand for 15 hr. at room temperature (14~20°). The reaction mixture was treated in the usual way to give a crude acetate (173 mg.) which showed two spots on the thin-layer chromatography (TLC) using $CHCl_3$-acetone (9:1) as solvent. The product was chromatographed on alumina (5.2 g.). Elution with *n*-hexane-benzene (3:2) afforded the diacetate (III; 32 mg.) which was recrystallized from MeOH as colorless plates, m.p. 208~210°. Further elution with *n*-hexane-benzene (1:1) gave the monoacetate (II; 125 mg.), m.p. 190~191°. These acetates were identical with those reported previously[1] in comparison of melting points, IR spectra and TLC.

b) A mixture of I (889 mg.), Ac_2O (9 ml.), pyridine (9 ml.), and $CHCl_3$ (20 ml.) was allowed to stand in an ice box (1~3°) for 20 hr. The product (927 mg.) was chromatographed on alumina (28 g.) and gave the diacetate (III; 53 mg.), m.p. 207~210°, and the monoacetate (II; 836 mg.), m.p. 190~191°.

Benzoylation of I——To a mixture of I (340 mg.), $CHCl_3$ (5 ml.) and pyridine (5 ml.), benzoylchloride (0.3 ml.) was added dropwise under cooling in ice-water. After the solution was allowed to stand at room

15) F. Dalton, J.I. McDougall, G.D. Meakins : J. Chem. Soc., **1963**, 4068; **1962**, 1566.
16) D.L. Klass, M. Fieser, L.F. Fieser : J. Am. Chem. Soc., **77**, 3829 (1955).
17) K. Takeda, T. Okanishi, H. Minato, A. Shimaoka : Tetrahedron, **19**, 759 (1963).

temperature for 14 hr., the excess reagent was then decomposed by the addition of 2*N* H_2SO_4 and the reaction mixture was extracted with $CHCl_3$. The organic layer was washed with H_2O, 5% $NaHCO_3$ and H_2O successively, dried and evaporated. The residue (633 mg.) was chromatographed on alumina (10 g.). Elution with benzene and crystallization from MeOH afforded the dibenzoate (IV; 360 mg.) as colorless needles, m.p. 207~208°, $[\alpha]_D^{23}$ −38.0° (c=0.665, $CHCl_3$). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3555 (OH, sharp), 1720 (C=O), 1604, 1589 (C=C). *Anal.* Calcd. for $C_{41}H_{52}O_7$: C, 74.97; H, 7.98. Found: C, 74.96; H, 8.02.

Oxidation of Monoacetate (II)——To a stirred solution of II (178 mg.) in Me_2CO (20 ml.) was added dropwise CrO_3–H_2SO_4 solution*3 (0.135 ml., containing 36.2 mg. of CrO_3, 1 equiv.) at 2°. After 2 min. the reaction mixture was diluted with H_2O and extracted with ether. The organic layer was washed with 5% $NaHCO_3$ and H_2O, dried over Na_2SO_4 and evaporated. Recrystallization of the residue (167 mg.) from Me_2CO gave the 1-oxoconvallagenin-A-3-acetate (V) as colorless needles, m.p. 210~213°, IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3585 (OH, sharp), 1724 (OAc), 1707 (C=O). NMR: Table I. ORD: Fig. 1, $[\alpha]_{589}^{21}$ −100.8° (c=0.273, MeOH), a=−104.3. *Anal.* Calcd. for $C_{29}H_{44}O_6$: C, 71.28; H, 9.07. Found: C, 71.24; H, 9.08.

Elimination of Acetic Acid from 3-Acetoxy-1-one (V)——A solution of V (122 mg.) in benzene (7 ml.) was stirred with Al_2O_3 (Brockman, 1.5 g.) at room temperature for 18 hr. The reaction mixture was filtered and evaporated *in vacuo*, and crystallization from MeOH–Me_2CO gave 25L,5β-hydroxyspirost-2-en-1-one (VI) as colorless needles (91 mg.), m.p. 234~236° (decomp.), UV λ_{max}^{EtOH} mμ: 225.5 (log ε 3.90), 332 (log ε 1.75), IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH, sharp), 1669, 1605 (α,β-unsaturated ketone). *Anal.* Calcd. for $C_{27}H_{40}O_4$: C, 75.66; H, 9.41. Found: C, 75.53; H, 9.32.

Dehydration of Diacetate (III)——To a solution of III (88 mg.) in pyridine (2 ml.), $SOCl_2$ (0.1 ml.) was added under ice cooling. After 1 hr. at room temperature, the reaction mixture was diluted with ice-cold water and ether. The organic layer was washed successively with 2*N* HCl, H_2O, 5% $NaHCO_3$ and H_2O, dried over Na_2SO_4, and evaporated. The product was recrystallized from MeOH to give anhydroconvallagenin-A diacetate (VII) as colorless needles, m.p. 207~208°, $[\alpha]_D^{21}$ −109.6° (c=0.526, $CHCl_3$), giving a positive tetranitromethane color test. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1730 (OAc), 3060, 1657 (C=C). NMR (in $CDCl_3$): 4.70 τ (C_4-H; singlet). *Anal.* Calcd. for $C_{31}H_{48}O_6$: C, 72.06; H, 9.36. Found: C, 71.83; H, 9.18.

Anhydroconvallagenin-A (VIII)——The unsaturated diacetate (VII, 66 mg.) was saponified by refluxing for 1.5 hr. with 3% KOH–MeOH (5 ml.). After the cooled reaction mixture was diluted with water, the precipitate formed was filtered and recrystallized from Me_2CO to give anhydroconvallagenin-A (VIII) as needles, 55 mg., m.p. 212~214°, $[\alpha]_D^{24}$ −87.8° (c=0.660, $CHCl_3$). IR ν_{max}^{Nujol} cm^{-1}: 3600~3200 (OH; broad), 3060, 1658 (C=C). *Anal.* Calcd. for $C_{27}H_{42}O_4$: C, 75.31; H, 9.83. Found: C, 75.25; H, 9.64.

Oxidation of Convallagenin-A (I)——A suspension of $PtO_2 \cdot H_2O$ (100 mg.) in a mixture of H_2O (10 ml.) and AcOH (1 ml.) was shaken under an atmosphere of H_2 until the reduction was completed giving metalic Pt, and the solvent was decanted off. A solution of I (150 mg.) in Me_2CO–AcOEt (1:2; 60 ml.) was then added and the mixture was shaken in O_2 atmosphere for 35 hr. The catalyst was filtered off and the filtrate was evaporated to dryness under reduced pressure. The residue (137 mg.) was chromatographed on alumina (4.1 g.). Elution with benzene–$CHCl_3$ (4:1) gave the α,β-unsaturated ketone (X; 16 mg.) which was recrystallized from MeOH as colorless needles, m.p. 229~231°. UV λ_{max}^{EtOH} mμ (log ε): 241.2 (4.16). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600, 3430 (OH), 1667 (C=O), 1624 (C=C). *Anal.* Calcd. for $C_{27}H_{40}O_4$: C, 75.66; H, 9.41. Found: C, 75.47; H, 9.34. Further elution with benzene–$CHCl_3$ (2:3) gave the 3-ketone (IX; 108 mg.) which was recrystallized from MeOH as colorless needles, m.p. 199~200°, $[\alpha]_D^{21}$ −44.7° (c=0.900, $CHCl_3$). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600, 3390 (OH; broad), 1717 (C=O). ORD: a=−12.8 (c=0.272; MeOH; Fig. 1). *Anal.* Calcd. for $C_{27}H_{42}O_5$: C, 72.61; H, 9.48. Found: C, 72.77; H, 9.46.

25L-Spirosta-1,4-dien-3-one (XI)——a) From the 3-ketone (IX): A solution of IX (126 mg.) in AcOH (6 ml.) was heated on a steam bath for 30 min., and the resulting solution was evaporated under reduced pressure. The residue (115 mg.) was chromatographed on alumina (3.5 g.). Elution with *n*-hexane–benzene (3:2) gave the 1,4-dien-3-one (XI; 80 mg.) which was recrystallized from MeOH as colorless needles, m.p. 176~179°, $[\alpha]_D^{23}$ −69.4° (c=0.490, $CHCl_3$). UV λ_{max}^{EtOH} mμ (log ε): 244.5 (4.19). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1663 (C=O), 1625, 1605 (C=C), 980, 920, >897, 850 (25L-spiroketal). *Anal.* Calcd. for $C_{27}H_{38}O_3$: C, 78.98; H, 9.33. Found: C, 79.06; H, 9.41.

b) From the 4-en-3-one (X): A solution of X (11 mg.) in 5% KOH–MeOH (4 ml.) was refluxed in water bath for 2 hr. The solution was diluted with H_2O, acidified with 2*N* HCl, and extracted with ether. The etheral solution was washed, dried and evaporated to dryness under reduced pressure. After being crystallized from MeOH, the 1,4-dien-3-one (XI), colorless needles (9 mg.), m.p. 175~178°, thus obtained was proved to be identical with that prepared as described above.

Isomerization of 25L-Spirosta-1,4-dien-3-one (XI)——The compound XI (150 mg.) was refluxed for 40 hr. in 80% EtOH (24 ml.) containing conc. HCl (5.1 ml.). After cooling, two volumes of H_2O were added to give the precipitate which was filtered, dried (124 mg.), and was then chromatographed on alumina (3.8 g.). Elution with *n*-hexane–benzene (3:2) gave 25D-spirosta-1,4-dien-3-one (XII; 85 mg.), which was recrystallized

*3 A solution of CrO_3 (6.7 g.) in conc. H_2SO_4 (5.3 ml.), diluted with H_2O to 25 ml., was used as a standard (CrO_3=268 mg./ml.). cf. C. Djerassi, R. R. Engle, A. Bowers: J. Org. Chem., **21**, 1547 (1956).

from MeOH as colorless needles, m.p. 197~200°, $[\alpha]_D^{23}$ −77.5°(c=0.521, $CHCl_3$). UV λ_{max}^{EtOH} mμ (log ε): 245 (4.15). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1659 (C=O), 1621, 1604 (C=C), 982, 918, <896, 861 (25D-spiroketal). These physical data were identical to those of the specimen prepared from dicyanodichlorobenzoquinone dehydrogenation of diosgenone.[10]

Treatment of Convallagenin-A Monoacetate (II) with Phosgene——II (139 mg.) was dissolved in alcohol-free $CHCl_3$ (20 ml.), and 3 ml. of the solvent was distilled off. After pyridine (10 ml.) was added to this solution, the mixture was cooled to −20° and 10% $COCl_2$-toluene solution (15 ml.) was added dropwise. The reaction mixture was allowed to stand at room temperature for 45 hr. After decomposing an excess of $COCl_2$ with ice, H_2O was added and the solution was extracted with ether. The organic layer was washed successively with 2*N* HCl, 5% $NaHCO_3$ and H_2O, dried over Na_2SO_4, and evaporated to dryness *in vacuo*. The residue (127 mg.) was chromatographed on alumina (3.7 g.) to give colorless prisms of 3β-acetylconvallagenin-A-1β,5β-carbonate (XIII; 98 mg.), m.p. 164~167°. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1736, 1106 (C=O). *Anal.* Calcd. for $C_{30}H_{44}O_7$: C, 69.74; H, 8.58. Found: C, 69.57; H, 8.42.

Mesylation of Monoacetate (II)——To a solution of II (239 mg.) in pyridine (8 ml.), CH_3SO_2Cl (2 ml.) was added dropwise under cooling with ice, and the solution was allowed to stand at 0° for 20 hr. The mixture was then diluted with ice-water and extracted with ether. After treatment in the usual way, the product (266 mg.) was recrystallized from *n*-hexane, giving a pure sample of the mesylate (XIV; 243 mg.) as colorless needles, m.p. 160° (decomp.). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3580 (OH; sharp), 1740 (C=O), 1350, 1167 (S=O). *Anal.* Calcd. for $C_{30}H_{48}O_8S$: C, 63.34; H, 8.50. Found: C, 63.28; H, 8.48.

Formation of 1-en-3,5-diol (XV) from Mesylate (XIV)——a) XIV (266 mg.) was dried by azeotropic distillation with benzene (10 ml.), dissolved in dimethylsulfoxide (6 ml.), and treated with potassium *tert*-butoxide reagent (20 ml.)*4 at room temperature for 5 hr. The reaction mixture was diluted with ice-water, acidified with 2*N* H_2SO_4 and extracted with ether. The etheral solution was washed with H_2O, dried and evaporated to dryness under reduced pressure. The residue (175 mg.) was chromatographed on alumina (5.3 g.) to yield the 1-en-3,5-diol (XV), which gave yellow color with tetranitromethane, and was recrystallized from MeOH as colorless needles (85 mg.), m.p. 229~231°. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3050, 1657 (C=C), 3590, 3550 (OH; sharp). *Anal.* Calcd. for $C_{27}H_{42}O_4$: C, 75.31; H, 9.83. Found: C, 75.15; H, 9.70.

b) To a stirred suspension of $LiAlH_4$ (280 mg.) in dry ether (18 ml.) was added dropwise a solution of XIV (110 mg.) in dry tetrahydrofuran (11 ml.). The mixture was refluxed for 3 hr. After destruction of the excess reagent with ice-water, the solution was acidified with 2*N* HCl and extracted with ether. The etheral layer was washed with 5% $NaHCO_3$ and H_2O, dried and evaporated. Chromatography of the residue (75 mg.) on alumina (2.3 g.) gave the 1-en-3,5-diol (XV; 53 mg.) which was recrystallized from MeOH, m.p. 229~231°, and identical in all respects with the compound (XV) mentioned above.

c) A solution of XIV (103 mg.) in dimethylsulfoxide (3 ml.) was heated at 95~100° for 2 hr. until the crystals separated, and the reaction was continued for additional 2 hr. The mixture was cooled, neutralized with 5% $NaHCO_3$, and poured into ice-water. The precipitate was collected, washed with H_2O, and dried *in vacuo*. Recrystallization from ether-EtOH gave 4-methyl-25L-spirosta-1,3,5(10)-triene (XVI) as colorless plates (79 mg.), m.p. 199~202°. $[\alpha]_D^{21}$ −53.1°(c=0.490, $CHCl_3$); tetranitromethane test: positive. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1591 (C=C), UV λ_{max}^{THF} mμ (ε): 259 (240; shoulder), 265 (260), 271 (240, shoulder). NMR ($CDCl_3$; τ): 7.78 (aromatic CH_3; singlet), 2.90 (aromatic H; complex). *Anal.* Calcd. for $C_{27}H_{38}O_2$: C, 82.18; H, 9.71. Found: C, 82.06; H, 9.55.

25L,5β-Spirostan-3β,5β-diol (XVII)——A solution of the unsaturated diol (XV; 125 mg.) in AcOEt (12.5 ml.) was shaken with platinum oxide (120 mg.) under atmospheric pressure and room temperature until 1 mole of H_2 was absorbed. The catalyst was removed by filtration and the filtrate was evaporated under reduced pressure. The residue (121 mg.) was recrystallized from MeOH to give the 3β,5β-diol (XVII) as colorless needles, m.p. 252~254°, $[\alpha]_D^{24}$ −17.2°(c=0.470, $CHCl_3$-MeOH, 1:1). IR $\nu_{max}^{CHCl_3}$ cm^{-3}: 3620, 3540 (OH, sharp). *Anal.* Calcd. for $C_{27}H_{44}O_4$: C, 74.95; H, 10.25. Found: C, 74.75; H, 10.09.

Acetylation of 3β,5β-diol (XVII)——A solution of XVII (74 mg.) in a mixture of pyridine (2 ml.) and Ac_2O (1 ml.) was heated at 95~100° for 2 hr. After treatment in the usual way, the product (78 mg.) was recrystallized from MeOH to give the acetate (XVIII) as colorless needles, m.p. 165~166°. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH), 1748 (OAc). *Anal.* Calcd. for $C_{29}H_{46}O_5$: C, 73.38; H, 9.77. Found: C, 73.33; H, 9.69.

3,5-Carbonate (XIX) of 25L,5β-Spirostan-3β,5β-diol (XVII)——The reaction of XVII (61 mg.) with $COCl_2$ was carried out in the similar manner to that of the monoacetate (II) described above. Recrystallization of the product (68 mg.) from MeOH-$CHCl_3$ afforded the 3,5-carbonate (XIX) as colorless needles, m.p. 222~224°. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1740, 1106 (C=O). *Anal.* Calcd. for $C_{28}H_{42}O_5$: C, 73.32; H, 9.23. Found: C, 73.12; H, 9.15.

Orthoester (XX) of Convallagenin-A (I)——I (88 mg.) was dissolved in warm MeOH (30 ml.). After cooling, 28.8% HCl-MeOH (1 ml.) and ethylorthoformate (3 ml.) was added, and the mixture was allowed to

*4 The potassium *tert*-butoxide reagent used was of 0.5*N*, freshly prepared by dissolving potassium *tert*-butoxide (1.400 g.) in dimethylsulfoxide (25 ml.). cf. F. C. Chang, N. F. Wood: Steroids, **4**, 55 (1964).

stand at room temperature for 24 hr. Evaporaton under reduced pressure and dilution with H_2O gave precipitates which were recrystallized from $CHCl_3$–MeOH to yield the orthoester (XX; 77 mg.) as colorless needles, m.p. 197~200°, $[\alpha]_D^{23}$ −56.6°(c=0.495, $CHCl_3$). IR $\nu_{max}^{CHCl_3}$ cm^{-1} : 1142, 991, 985 (orthoester), 979, 919, >896, 846 (25L-spiroketal). NMR ($CDCl_3$; τ) : 9.22 (C_{18}–CH_3), 9.04 (27–CH_3; doublet), 8.65 (19–CH_3), 4.35 (orthoester–H). *Anal.* Calcd. for $C_{28}H_{42}O_5$: C, 73.32; H, 9.23. Found : C, 73.29; H, 9.25.

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