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STRUCTURE AND STEREOCHEMISTRY OF PARISTERONE, A NOVEL PHYTOECDYSONE FROM THE TUBERS OF *PARIS POLYPHYLLA*

S. B. SINGH and R. S. THAKUR*
Central Institute of Medicinal and Aromatic Plants, Lucknow-226010, India

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Abstract—Paristerone, a novel phytoecdysone has been isolated from the tubers of *P. polyphylla* and its structure and stereochemistry has been established as 2α, 3β, 14α, 20(R), 22(R), 25-hexahydroxy-5β-cholest-7-en-6-one (2-epiecdysterone) on the basis of chemical and physical evidence.

Paris polyphylla Sm. (Liliaceae) is a temperate plant distributed throughout the central southern slopes of Himalayas. In early reports[1,2] depressant action on carotid pressure has been described in the tuber extract. Re-examination of the total alcoholic extract of the tubers confirmed the activity. Consequently our interest was revived in the isolation and study of its chemical constituents. Recently[3-5] we have reported a series of new steroidal saponins. During the course of these studies, a phytoecdysone designated as paristerone was isolated.

Paristerone 1, m.p. 216–20° (decomp), $[\alpha]_D$ +41.9°, possesses the composition $C_{27}H_{44}O_7$ (FDMS, M^+ at *m/z* 480). It gave positive colour reactions typical of steroids.[6] The UV spectrum of 1 showed absorption at 240 nm (ϵ = 10314) and an intense IR band at 1650 cm^{-1} corresponding to an enone system. ^{1}H-NMR spectrum displayed a broad singlet at δ 5.65 which could be assigned to a proton α- to the enone carbonyl. These data together with ^{13}C-NMR chemical shift at δ202.7 strongly suggested the presence of a β, β-substituted-α,β-unsaturated carbonyl moiety in the molecule.[7] Furthermore a strong IR band at 3300 cm^{-1} indicated this compound to be an ecdysone.

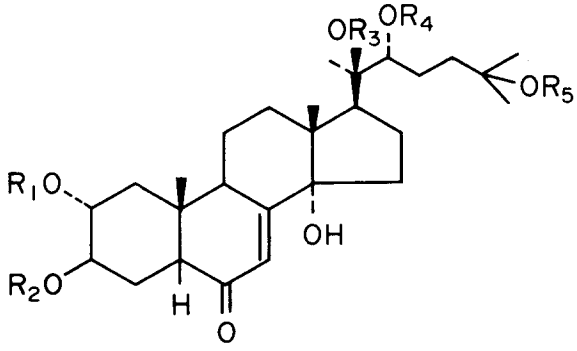
When paristerone was heated in methanolic HCl the resultant product showed UV maxima at 294 and 245 nm. Such behaviour is compatible with the 7-en-6-one system having a 14α-hydroxyl group.[8]

The EIMS of 1 did not show M^+ due to facile loss of water and the ions formed were *m/z* 462 ($M-H_2O$), 444 ($M-2H_2O$) and 426.2788 ($C_{27}H_{38}O_4$, $M-3H_2O$) (Scheme 1). Successive elimination of H_2O followed by the increase in the intensity of the signals are indicative of the presence of at least three tertiary hydroxyl groups. Loss of a methyl group was seen at *m/z* 411 (426–15).

Presence of a vicinal dihydroxy group at C-20, C-22 was indicated by abundant ions at *m/z* 363.2166 ($C_{21}H_{31}O_5$) and 117 due to cleavage between C-20 and C-22 position (Scheme) which readily eliminates water and produces ions at *m/z* 345, 327, 99 and 81. These ions are characteristic for ecdysones bearing 20,22-dihydroxy groups with the remaining three hydroxy function in steroid nucleus and one in the side chain.[8,9] This stipulated fact was substantiated by the characteristic fragments obtained after cleavage of the molecule at C-17, C-20, which in EIMS afforded ions at *m/z* 319, 301, 300, 161, 143, 125, 59 and 43. The peaks at *m/z* 59 (C_3H_7O) and 43 (C_2H_3O) could only be possible when the molecule bears a hydroxy group at C-25 and cleavage[8] takes place between C-24, C-25. The presence of the OH group at C-25 was also supported by downfield appearance of a singlet at δ1.10 of the methyl protons at C-26 and C-27 and ^{13}C-NMR at δ75.8 for C-25.

The mass spectrum discussed above and chemical shift of methyl protons in the ^{1}H-NMR spectrum (Table 1) and ^{13}C-NMR chemical shift of 1 are similar (with few exceptions) to ecdysterone having hydroxyl groups at C-20, C-22 and C-25 along with 5β-configuration and C-2, C-3-diol moiety.

Acetylation of paristerone with Ac_2O/C_5H_5N furnished a mixture of five acetates which have been designated as **1a**, **1b**, **1c**, **1d** and **1e** in their decreasing R_f values.



1 $R_1 = R_2 = R_3 = R_4 = R_5 = H$

1a $R_1 = R_2 = R_4 = R_5 = Ac$, $R_3 = H$

1b $R_1 = R_2 = R_4 = Ac$, $R_3 = R_5 = H$

1c $R_1 = R_2 = Ac$, $R_3 = R_4 = R_5 = H$

1d $R_1 = R_4 = Ac$, $R_2 = R_3 = R_5 = H$

1e $R_2 = R_4 = Ac$, $R_1 = R_3 = R_5 = H$

1f $R_1 R_2 = CMe_2$, $R_3 R_4 = CMe_2$, $R_5 = H$

1g $R_3 = R_4 = R_5 = H$, $R_1 R_2 = CMe_2$

1h $R_1 = R_2 = R_5 = H$, $R_3 R_4 = CMe_2$

The MS of **1a** did not show the expected molecular ion. Characteristic fragments at *m/z* 447, 429 and 385 of the diacetylated steroid nucleus of paristerone and diacetylated side chain fragments at *m/z* 201 and 185

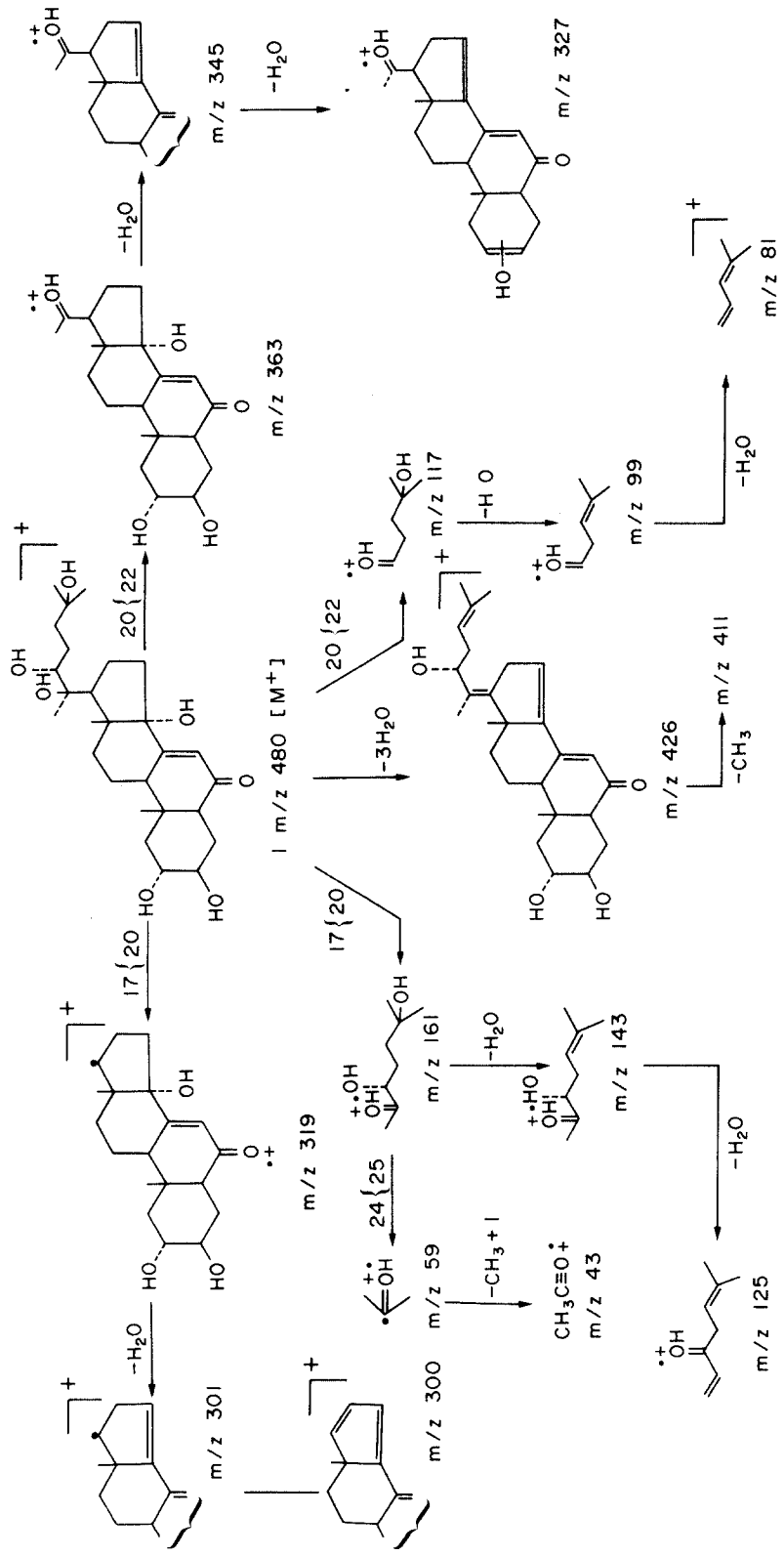

m/z 480 [M+]
m/z 363
m/z 345
m/z 327
m/z 319
m/z 301
m/z 300
m/z 426
m/z 411
m/z 161
m/z 143
m/z 125
m/z 117
m/z 99
m/z 81
m/z 59
CH3C≡O+
m/z 43
-H2O
-3H2O
-CH3
-H O
17{20
20{22
24{25
-CH3+1

Table 1. ^{1}H-NMR chemical shifts (δ,ppm from TMS) of paristerone and derivatives

Sl No.	Compound	18-CH_3 s	19-CH_3 s	21-CH_3 s	26,27 CH_3,s	C-2H,brs, ($W_{\frac{1}{2}}$)	C-3H,brs, ($W_{\frac{1}{2}}$)	C-22H,brs, ($W_{\frac{1}{2}}$)	C-7H,brs, ($W_{\frac{1}{2}}$)	C-9H m	Others
1	Ecdysterone	0.77	0.85	1.08	1.08	-	-	-	-	-	-
2.	Paristerone	0.77	0.84	1.10	1.10	3.75 (8)	3.75 (8)	3.20 (10)	5.65	-	4.60 (14-OH) 3.32 all other OH,D_2O exchange.
3	Ecdysterone-2, 3,22,25-tetra-acetate	0.86	1.02	1.26	1.42	4.91 (20)	5.29 (7)	-	5.85	-	-
4.	Ecdysterone,2, 3,22-triacetate	0.86	1.02	1.24	1.20	5.04 (20)	5.31 (7)	4.79	5.85	3.10	-
5	Paristerone-2, 3,22,25-tetra-acetate.	0.78	0.98	1.20	1.35	4.90 (10)	5.27 (9)	4.70 (11)	5.90 (6)	3.05	1.92,1.95,2.02, 2.08 (4x$OCOCH_3$)
6	Paristerone-2, 3,22-triacetate.	0.78	0.95	1.28	1.16	4.77 (7)	5.25 (12)	4.68 (7)	5.75 (7)	3.05	1.94 (3H), 2.05 (6H) (3x$OCOCH_3$)
7	Paristerone-2, 3-diacetate.	0.78	0.96	1.10	1.10	4.95 (7)	5.40 (11)	-	5.65	-	1.75, 1.95 (2x$OCOCH_3$)
8	Paristerone-2, 22-diacetate	0.71	0.90	1.08	1.12	4.85 (11)	4.05 (m)	4.68 (12)	5.75	3.05	1.82, 1.95, 2.0 ($OCOCH_3$)
9	Paristerone-3, 22-diacetate.	0.71	0.90	1.08	1.12	4.05 (m)	5.15 (9)	4.65 (12)	5.75	3.05	1.82,1.95, 2.0 ($OCOCH_3$).
10	Paristerone-2, 3-monoacetonide	0.82	0.90	1.20 -.40	1.20 -.40	3.50 -3.90	3.50 -3.90	3.50 -3.90	5.75	3.00	1.20-.40 $(CH_3)_2$:C:$(O)_2$
11	Paristerone-20 22-monoacetonide	0.78	0.88	1.15 - .35	1.15 -.35	3.55 -3.80	3.55 - 3.80	3.55 -3.80	5.75	-	1.15-.35 $(CH_3)_2$:C:$(O)_2$
12	Paristerone-2,3, 20,22-diacetonide	0.70	0.90	1.15 -.38	1.15 -.38	3.50	3.50	3.50	5.65	3.00	1.15-.38 $(CH_3)_2$:C:$(O)_2$

The Chemical shift data of S.No.1 (DMSO-d_6) 3 and 4 ($CDCl_3$) have been taken from Ref.No. 16,6 and 15 respectively. The spectrum of paristerone have been recorded on 60 MHz in DMSO-d_6 while others on 90 MHz NMR spectrophotometer in $CDCl_3$. The figures in parentheses are in Hz.

after cleavage at C-20, C-22 with other ions at *m/z* 169, 140, 99 and 81 indicated it to be a tetraacetate. A deshielded singlet at δ1.35 was obtained in the ^{1}H-NMR spectrum assignable to the 26 and 27 methyl protons. This deshielding is only possible after acetylation of the C-25 hydroxyl group, which is favoured due to lesser hindrance to the tertiary hydroxy group located at the far end of the side chain. Thus **1a** could be identified as paristerone-2, 3, 22, 25-tetracetate.

Similarly **1b** displayed 26 and 27 methyl proton signal at δ1.16 in ^{1}H-NMR spectrum and fragments at *m/z* 447, 385, 185, 141, 99 and 81 in MS are consistent for paristerone-2, 3, 22-triacetate.

1c has been established as paristerone -2, 3-diacetate on the basis of its ^{1}H-NMR spectrum (Table 1).

1d and **1e**, which are position isomers were obtained as a mixture. The MS showed fragments at *m/z* 406, 387, 185 and 141 for monoacetylated steroid nucleus and monoacetylated side chain as expected from both the acetates. The ^{1}H-NMR spectrum exhibited signals at δ4.65 (1H), 4.85 (1/3 H) and 5.15 (2/3 H). The appearance of these three signals lead to suspicions about the homogeneity of the compound which were strengthened by observation of a multiplet in ^{1}H-NMR at δ4.05 (HO–C_2–H and HO–C_3–H). The peak area measurement of the AcO–C_2–H and AcO–C_3–H established the ratio of 2,22-diacetate (**1d**) and 3,22-diacetate (**1e**) as 1 : 2. Every possible effort at separation has failed to separate this mixture.

The presence of vicinal dihydroxy groups at C-2, C-3 and C-20, C-22 was further confirmed by the formation of diacetonide, paristerone-2,3,20,22-diacetonide (**1f**) together with paristerone-2,3-monoacetonide (**1g**) and paristerone-20,22-monoacetonide (**1h**).

The above evidence coupled with ^{13}C-NMR (Table 2) led us to postulate that paristerone may be an isomer of ecdysterone. A significant difference in the ^{13}C-NMR chemical shift of C-25 was noticed which was shifted downfield by ≈6 ppm from that of ecdysterone. It is worth pointing out that this type of chemical shift (≈73 ppm) for C-25 has been reported[10] in C_5D_5N for the compounds containing a methyl group at C-24 and of shidasterone[7] (75.4 ppm). The possibility of containing a methyl group at C-24 and structure like shidasterone at C-25 in **1** is ruled out on the basis of other spectral data. Therefore, it is assumed that this downfield shift may possibly due to solvent (DMSO-d_6) effect which deshielded the C-25 through less hindered hydroxyl group. The main stereochemical difference was observed at C-2 position which is discussed below.

Stereochemistry of paristerone. The stereochemistry of paristerone was deduced as follows:

The CD spectrum of paristerone in dioxan (Figure 1) showed a characteristic positive Cotton effect at 343 nm $[\theta] = +5070$ due to the $n \rightarrow \pi^*$ transition of enone group of A/B cis (5β) ring junction[11,12]. A negative Cotton effect at 246 nm $[\theta] = -16490$ due to $\pi \rightarrow \pi^*$ transition was also observed which supports A/B cis junction. The slight bathochromic shifts (see reported values 243, 340 nm[11,12]) and high negative ellipticity of the latter suggests the stereochemical variation of the substituents in ring A.

The CD spectrum of **1** in MeOH similarly exhibited two Cotton effects at 332 nm $[\theta] = +6820$ and at 252 nm $[\theta] = -17060$. The hypsochromic shift (11 nm) of former and bathochromic shift (6 nm) of latter wavelength and consequent increase in the ellipticity of both Cotton effects in MeOH may be due to high solvation power of the solvent (solvent effect). Cases of solvent effect are reported[13] in saturated steroids but no such account is available in the case of ecdysones.

Thus the ellipticity and Cotton effects in the CD curve of the paristerone indicated A/B cis ring junction (5β

Table 2. ^{13}C-NMR chemical shifts (δ, ppm from TMS) of paristerone

Carbon Number	Ecdysterone	Paristerone**
1.	37.8	37.6
2.	68.0	66.8*
3.	68.0	68.8*
4.	32.3	33.2
5	51.1	50.1
6	203.2	202.7
7	121.6	120.5
8	165.7	165.2
9	34.5	36.7
10	38.5	38.5
11	21.0	20.3
12	31.5	30.9
13	48.1	46.9
14	84.2	83.1
15	32.0	31.5
16	21.5	23.8
17	50.1	48.8
18	17.7	17.1
19	24.3	26.1
20	76.9	75.8
21	21.5	20.9
22	77.5	76.2
23	27.2	29.0
24	42.4	41.3
25	69.7	75.8
26	29.9	29.8
27	29.9	29.8

* Assignments may be reversed.

**In DMSO-d_6

Chemical shifts of Ecdysterone(in C_5D_5N) has been taken from ref.17. The slight difference in the chemical shifts may be due to solvent effect[18].

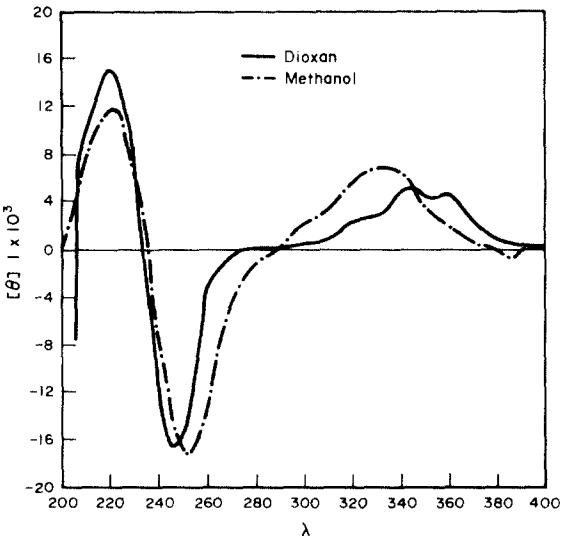


Fig. 1. CD spectrum of paristerone.

hydrogen). Almost all the known ecdysones have A/B cis ring junction.[8,11]

The configuration of C-2 and C-3 hydroxyl groups in paristerone was found to be different from any of the reported ecdysones. This was evident from the comparative study of the ^{1}H-NMR spectra of the acetate derivatives of ecdysterone and paristerone, particularly the signals due to C-2 and C-3 protons (Table 1). The ^{1}H-NMR spectrum of paristerone-2,3,22-triacetate (others also) did not display any proton signal with $W_{\frac{1}{2}}^{1}$ 20 Hz while it was observed in the similar acetate of ecdysterone for 2α-H and ponasterone B and C for 3β-H.[6,12,14] This fact eliminates the possibility of identical configuration at C-2 and C-3 *viz* 2β, 3β or 2α, 3α of hydroxyl-groups. Alternatively, the configuration may be 2β, 3α or 2α, 3β for hydroxyl functions. The former possibility is ruled out because the hydrogens at C-2 and C-3 would be α and β respectively and by virtue of 5β configuration these hydrogens would undergo axial–axial coupling giving rise to $W_{\frac{1}{2}}^{1} \sim 20$ Hz. Hence the only possibility which would not give $W_{\frac{1}{2}}^{1} \sim 20$ Hz is 2α, 3β which is assigned to the paristerone. In this configuration both the hydroxyls are axial leaving hydrogen to be equatorial which would be anticipated to give $W_{\frac{1}{2}}^{1} \sim 7$–12 Hz (chair conformation of ring A). This type of low $W_{\frac{1}{2}}^{1}$ values was observed in the acetates of paristerone, e.g. paristerone-2,3,22-triacetate displayed signals at δ4.77 and 5.25 ($W_{\frac{1}{2}}^{1} = 7$ and 12 Hz) for 2β and 3α-hydrogen respectively. This observation was also supported by ^{13}C-NMR data (Table 2) which exhibited different chemical shifts for both the carbons while they were found indistinguishable in the ecdysones containing 2β, 3β or 2α, 3α-configuration with 5β hydrogen. The data obtained suggested the configuration of C-2 and C-3 as 2α and 3β, both axial (5β) in paristerone. This type of configuration present in any ecdysone is being reported for the first time.

^{1}H-NMR chemical shifts of 18 and 21 methyl as well as C-22 H of paristerone and its acetates are comparable with the ecdysterone and its acetates having R, R configuration at C-20 and C-22 (Table 1). Thus R, R-configuration has been assigned to paristerone which is also supported by comparable ^{13}C-NMR chemical shifts (Table 2) with ecdysterone and the formation of monacetonide in fairly good yield.

Thus on the basis of above data, the structure of paristerone is established as 2α, 3β, 14α, 20(R), 22(R), 25-hexahydroxy-5β-chloest-7-en-6-one (**1**) (2-epiecdysterone). The novel configuration at C-2 and C-3 may lead to isolation of a new series of phytoecdysones which may suggest the role of these compounds in moulting activity.

EXPERIMENTAL

M.ps were determined in open capillaries and are uncorrected. IR spectra were determined on Perkin–Elmer-177 in KBr and UV spectra in MeOH on Pye Unicam SP-8–100 spectrophotometers. The ^{1}H-NMR spectra were recorded on Perkin–Elmer R-32 (90 MHz) or Varian A-60D (60 MHz) spectrophotometer in the $CDCl_3$ unless otherwise stated using TMS as internal reference. All chemical shift values are reported in δ(ppm). The MS were done on JEOL JMS-D300 mass spectrophotometer with JMA-2000 data processing unit. High resolution mass spectra were recorded on Varian MAT-311 instrument. The circular dichroism spectra were taken on Jobin–Yvon dichrograph III at 22°. Tlc was performed on silica gel G (BDH) and spots were visualized with iodine and/or 10% H_2SO_4.

Extraction and isolation. Dried and powdered tubers (1 kg) of *P. Polyphylla* (collected from Nepal) were extracted successively with boiling n-hexane (16 h) and methanol (17 h) (3l each). A voucher specimen has been deposited in Herbareum of National Botanical Research Institute, Lucknow (India). The methanolic extract was concentrated on water bath under *vacuo*. The concentrate (91 g) was diluted with water and extracted with $CHCl_3$ (5 × 500 ml). The aqueous layer was again extracted with n-butanol (5 × 250 ml). The butanol layer on complete removal of the solvent under *vacuo* furnished crude solid (40.6 g, 4% dwb). This crude mixture was chromatographed on silica gel (1 kg) column and eluted successively with $CHCl_3$ and $CHCl_3$ with varying percentage of methanol. Early fractions of MeOH–$CHCl_3$ (1:9) yielded trillin while latter fractions gave paristerone.

Paristerone, crystallized from methanol as colourless needles (550 mg, 0.099%) m.p. 216–20° (decomp) $[\alpha]_D^{25}$ + 41.9° (Pyridine, C, 0.5), R_f 0.63 ($CHCl_3$: MeOH: H_2O, 70:35:7). λ_{max}^{MeOH} 240(ε = 10314) λ_{max}^{MeOH} + dil HCl 245, 294 nm. IR of acid treated product ν_{max} 3400, 2900, 1710, 1650(w), 1460, 1380 and 1060 cm^{-1}. IR of **1** ν_{max} 3300 (broad), 1650 (s), 1435, 1380 (CH_3), 1260, 1150, 1060, 1030, 940, 920, 900 and 888 cm^{-1}. FDMS *m/z* 480.4 (M^+, 34%, $C_{27}H_{44}O_7$), MS: *m/z* (rel. int.%) 462, 444, 426, 411(5), 363(33), 345(62), 327(23), 320(4), 309(6), 301(8), 300(5), 285(5), 269(12), 250(9), 225(3), 213(4), 191(8), 173(12), 161(6), 143(10), 117(6), 99(40), 81(27), 69(28), 58(100). The ions at *m/z* 462, 444 and 426 were of very low intensity and could only be detected faintly on UV paper. In HR-MS the lower peaks were observed at *m/z* 59 (24%) and 43(100%). The other peaks are almost similar.

HR-MS (Accurate masses and composition)

Observed mass	Calculated mass	Composition
426.2788	426.2770	$C_{27}H_{38}O_4$
363.2166	363.2171	$C_{21}H_{31}O_5$
345.2052	345.2066	$C_{21}H_{29}O_4$
327.1962	327.1960	$C_{21}H_{27}O_3$

Acetylation of paristerone. A mixture of paristerone (400 mg), pyridine (10 ml) and acetic anyhydride (5 ml) were kept at room temperature for 24 h, finally heated on water bath for 2 h, (The reaction mixture exhibited four spots on the tlc) was diluted with ice cold water and extracted with chloroform. The organic layer was repeatedly washed with water and dired over anhydrous Na_2SO_4. The solvent was removed under reduced pressure. The mixture (403 mg) was resolved on plc ($CHCl_3$: $(CH_3)_2$ CO, 4:1) and the acetates were isolated and characterised.

Paristerone-2,3,22,25-tetraacetate (**1a**; 60 mg) was crystallized from acetone-hexane to give an amorphous powder m.p. 130–1° (decomp), R_f 0.60 ($CHCl_3$–$(CH_3)_2CO$, 4:1), λ_{max}^{EtOH} 238 nm. IR ν_{max} 3400, 1725, 1650, 1425, 1370 and 1230 cm^{-1}. MS: *m/z* (rel. int.%) 510(13.2, M-2 AcOH–H_2O), 495(4.0, M–H_2O–2AcOH–CH_3), 492(7.6, M–2AcOH–2H_2O), 447(3.7), 429(6.2), 385(4.2), 334(8.6), 327(8.2), 309(2.2), 232(5.7), 201(2.3), 185(2.7), 169(6.5), 161(4.2), 149(5.8), 140(2.1), 119(5.9), 99(9.7), 81(9.5), 59(2.5), 43(100). Found: C, 63.52; H, 8.00, $C_{35}H_{52}O_{11}$. H_2O requires C, 63.61; H, 8.10%.

Paristerone-2,3,22-triacetate (**1b**; 125 mg) was obtained as amorphous powder from acetone–hexane, m.p. 172–3° (decomp), R_f 0.43 ($CHCl_3$: $(CH_3)_2$ CO, 4:1) λ_{max}^{EtOH} 242 nm. IR ν_{max} 3450 (broad), 1735, 1660, 1450, 1370 and 1250 cm^{-1}. MS: *m/z* (rel. int.%) 510(6.4, M–AcOH–2H_2O), 495(2.0, M–AcOH–2H_2O–CH_3), 492(3.2, M–AcOH–3H_2O), 447(3.1), 429(4.2), 385(2.5), 334(8.7), 327(6.4), 257(2.4), 232(4.1), 185(4.0), 183(3.1), 169(4.6), 161(3.2), 141(6.0), 99(18.8), 59(7.2), 57(100). Found: C, 63.51; H, 8.25, $C_{33}H_{50}O_{10}$. H_2O requires C, 63.46; H, 8.33%. Paristerone-2,3-diacetate (**1c**; 35 mg) was obtained as amorphous powder on crystallization from acetone-hexane. m.p. 123–5° (decomp), R_f 0.19 ($CHCl_3$: $(CH_3)_2$ CO, 4:1). λ_{max}^{EtOH} 243 nm. Found C,63.61; H, 8.69; $C_{31}H_{48}O_9$. H_2O requires C, 63.75; H, 8.89%.

Paristerone-2,22 and 3,22-diacetate (**1d** and **1e**; 73 mg), all efforts to separate this mixture failed and they were obtained as amorphous powder, m.p. 160–2° (decomp) R_f 0.11 ($CHCl_3$: $(CH_3)_2$ CO, 4:1) λ_{max}^{EtOH} 242 nm. IR ν_{max} 3400, 1735, 1650, 1450, 1370 and 1240 cm^{-1} MS: *m/z* (rel. int.%) 529(0.1, M–2H_2O + H), 511 (0.2, M–3H_2O + H), 510(0.9, M–3H_2O), 495(0.6, M–3H_2O–CH_3),

492(0.1, M–4H_2O), 407(0.3, 405 + 2H), 406(0.1, 405 + H), 393(M–2AcOH–2H_2O–CH_3), 387(0.2, M–AcOH–4H_2O–3CH_3), 362(0.2), 335(0.4), 334(1.6), 327(0.4), 323(0.4), 232(1.4), 185(1.7), 169(3.0), 161(2.1), 149(3.2), 141(5.4), 99(18.3), 81(23.5), 57(100). Found: C, 63.52; H, 8.73; $C_{31}H_{48}O_9$. H_2O requires C, 63.75; H, 8.89%.

Preparation of acetonide of paristerone. A suspension of paristerone (111 mg) in anhydrous acetone (150 ml) was refluxed with anhydrous $CuSO_4$ (10 g) for 6 h and the reaction was monitored by tlc. After completion of the reaction, the mixture was filtered. Removal of the solvent from the filtrate afforded the mixture of acetonide along with a small amount of unreacted product. This mixture was resolved by plc on silica gel G (CH_2Cl_2–MeOH, 9:1). The acetonides numbered **1f** (22 mg), **1g** (15 mg) and **1h** (110 mg) were separated with decreasing R_f values.

Paristerone-2,3,20,22-diacetonide. **1f** was crystallized from acetone-hexane as colourless powder, m.p. 166–7°, R_f 0.72 (CH_2Cl_2: MeOH, 9:1) λ_{max}^{EtOH} 240 nm IR ν_{max} 3400, 1645(s), 1440, 1380(s), 1200, 1100 and 1040 cm^{-1}. MS: *m/z* (rel int.%) 361 (23.2), 345(21.9), 341(5.1), 329(13.0), 328(28.3), 301(77.6), 300(58.9), 267(10.1), 199(4.5), 159(4.7), 143(30.3), 117(6.4), 99(64.9) and 59(100), MW 560. Found C, 70.5; H, 9.2; $C_{33}H_{52}O_7$ requires C, 70.7, H, 9.4%.

Paristerone-2,3-monoacetonide. (**1g**) was crystallized from acetone-hexane as amorphous powder, m.p. 228–30°, R_f 0.54(CH_2Cl_2: MeOH, 9:1), λ_{max}^{EtOH} 240 nm, IR ν_{max} 3400, 1650(s), 1440, 1360(s) and 1200 cm^{-1}. Found C, 68.3; H, 9.35; $C_{30}H_{48}O_7$. $\frac{1}{2}H_2O$ requires C, 68.05; H, 9.24%.

Paristerone-20,22-monoacetonide. (**1h**) on crystallization from acetone-hexane afforded colourless crystals, mp 180–2°, R_f 0.27(CH_2Cl_2: MeOH, 9:1), λ_{max}^{EtOH} 241 nm. IR ν_{max} 3400, 1650(s). 1440, 1370(s), 1200 and 1045 cm^{-1}. MS: *m/z* (rel. int.%) 426 (3.3), 363(55.9), 345(11.2), 327(3.2), 301(13.2), 300(21.2), 199(2.5), 197(2.5), 185(2.9), 159(3.7), 143(21.0), 117(13.0), 99(59.3), 81(16.8), 59(100) and 43(55.6), MW(520). Found C, 66.78; H, 9.35; $C_{30}H_{48}O_7$. H_2O requires C, 66.9; H, 9.29%.

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REFERENCES

[1]R. N. Chopra, J. C. Gupta, B. C. Bose and I. C. Chopra, *Ind. J. Med. Res.* **30**, 103 (1942).
[2]A. T. Dutt, N. R. Chatterjee, S. Ghosh and R. N. Chopra, *Arch. Pharm.* **276**, 343 (1938).
[3]S. B. Singh and R. S. Thakur, *Planta Med.* **40**, 301 (1980).
[4]S. B. Singh and R. S. Thakur, *Phytochemistry* (communicated).
[5]S. B. Singh and R. S. Thakur, *Ibid.* (communicated).
[6]M. N. Galbraith and D. H. S. Horn, *Aust. J. Chem.* **22**, 1045 (1969).
[7]H. Hikino, T. Okuyama, S. Arihara, Y. Hikino, T. Takemoto, H. Mori and K. Shibata, *Chem. Pharm. Bull.* **23**, 1458 (1975).
[8]A. Prakash and S. Ghosal, *J. Sci. Ind. Res.* **38**, 632 (1979).
[9]N. Ikekawa, T. Ikeda, T. Mizuno, E. Ohnishi and S. Sakurai, *J. Chem. Soc. Chem. Commun.* 448 (1980).
[10]J. W. Blunt, G. A. Lane, M. H. G. Murno and G. B. Russell, *Aust. J. Chem.* **32**, 779 (1979).
[11]H. Hikino and Y. Hikino, *Progress in the Chemistry of Organic Natural Products* (Edited by W. Herz, H. Grisebach and A. I. Scott), Vol. 28, p. 256. Springer Verlag, Wein (1970).
[12]M. Koreeda and K. Nakanishi, *J. Chem. Soc., Chem. Commun.* 351 (1970).
[13]P. Carabbé, *Optical Rotatory Dispersion and Circular Dichroism in Organic Chemistry.* Holden-Day, San Francisco (1965).
[14]K. Nakanishi, M. Koreeda, M. L. Chang and H. Y. Hsu, *Tetrahedron Letters* 1105 (1968).
[15]T. Takemoto, Y. Hikino, K. Nomoto and H. Hikino, *Ibid.* 3191 (1967).
[16]J. Jizba, V. Herout and F. Sorm, *Ibid.* 5139 (1967).
[17]J. Krepinsky, J. A. Findlay, B. Danieli, G. Pamisano, P. Beynon and S. Murakami, *Org. Magn. Reson.* **10**, 255 (1977).
[18]G. C. Levy and G. L. Nelson, *Carbon-13. Nuclear Magnetic Resonance for Organic Chemists*, p. 200. Wiley-Interscience, New York (1972).