# Syntheses of 22,23-Dihydro-1α,25-dihydroxyvitamin $D_2$ and Its 24*R*-Epimer, New Vitamin $D_2$ Derivatives

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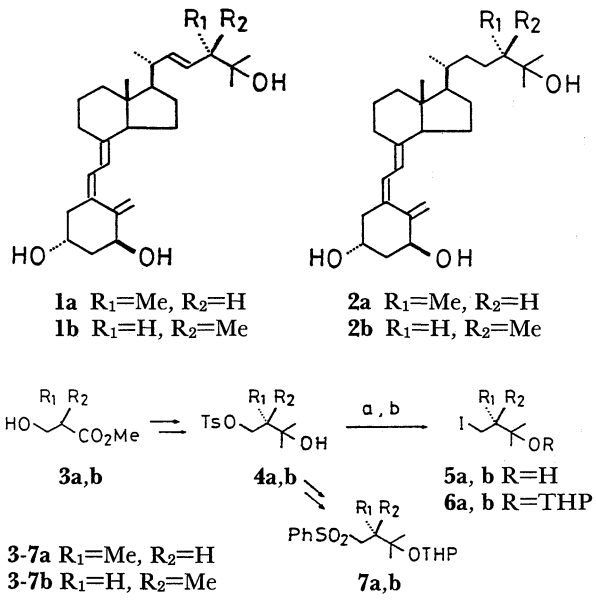
New 1α,25-dihydroxyvitamin $D_2$ derivatives (22,23-dihydro-1α,25-dihydroxyvitamin $D_2$ (**2a**) and its 24*R*-epimer (**2b**)), were synthesized by two procedures. 22-Oxo-5α,8α-(4-phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-23,24-dinor-6-cholene-1α,3β-diyl diacetate (**8**), obtainable readily from ergosterol, was converted to 22-phenylsulfonyl-5α,8α-(4-phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-1α,3β-bis(tetrahydropyranyloxy)-23,24-dinor-6-cholene (**14**) or 22-iodo-5α,8α-(4-phenyl-3, 5-dioxo-1,2,4-triazolidine-1,2-diyl)-1α,3β-bis(tetrahydropyranyloxy)-23,24-dinor-6-cholene (**16**). Condensation of the C-22 sulfone (**14**) with (3*R*)-4-iodo-2,3-dimethyl-2-butanol THP ether, or the C-22 iodide (**16**) with (3*R*)-2,3-dimethyl-4-phenylsulfonyl-2-butanol THP ether followed by desulfonylation and successive deprotection gave 5,7-ergostadiene-1α,3β,25-triol (**21a**), which led to **2a** upon irradiation and subsequent thermal isomerization. Similarly, a 24*R*-epimer of **2a** was synthesized.

It is well established that vitamin $D_3$ and vitamin $D_2$ must be hydroxylated at the C-25 position in the liver, and subsequently at the C-1α position in the kidney, before eliciting their physiological activity.[1] Biological testing indicated that the activity of 1α,25-dihydroxyvitamin $D_2$ (**1a**) is similar to that of the corresponding vitamin $D_3$ derivative in manmals, though the former is 1/5—1/10 less active than the latter in birds.[2] Recently, DeLuca et al. reported that though 1α-hydroxyvitamin $D_2$ is equally potent to 1α-hydroxyvitamin $D_3$ regarding biological activity, the former is 5—10 times less toxic than the latter in rats.[3] These findings seem to be responsible for the difference of the side chain moiety between these "active" vitamin $D_2$ and vitamin $D_3$.

In order to study the functional importance of the 24-methyl group, the (24*R*)-1α,25-dihydroxyvitamin $D_2$[4-6] (**1b**) and (22*E*)- and (22*Z*)-dehydro-1α-hydroxyvitamin $D_3$[7] were synthesized and their binding affinities investigated.[8] However, the influence of the C-22 double bond of these active vitamin $D_2$ derivatives on the physiological activity has not yet been clarified. These observations prompted us to study the effect of unsaturation at C-22,23 of 1α,25-dihydroxyvitamin $D_2$. In the present work, the syntheses of 22,23-dihydro-1α,25-dihydroxyvitamin $D_2$ (**2a**) and its 24*R*-epimer (**2b**), the new active vitamin $D_2$ derivatives, are described.

Our syntheses of **2a** and **2b** were achieved by two procedures. One was based on a condensation of the steroidal C-22 sulfone with optically active iodide derivatives to constitute the side chain part; the other was based on a coupling of the steroidal C-22 iodide with the corresponding side chain sulfone derivatives.

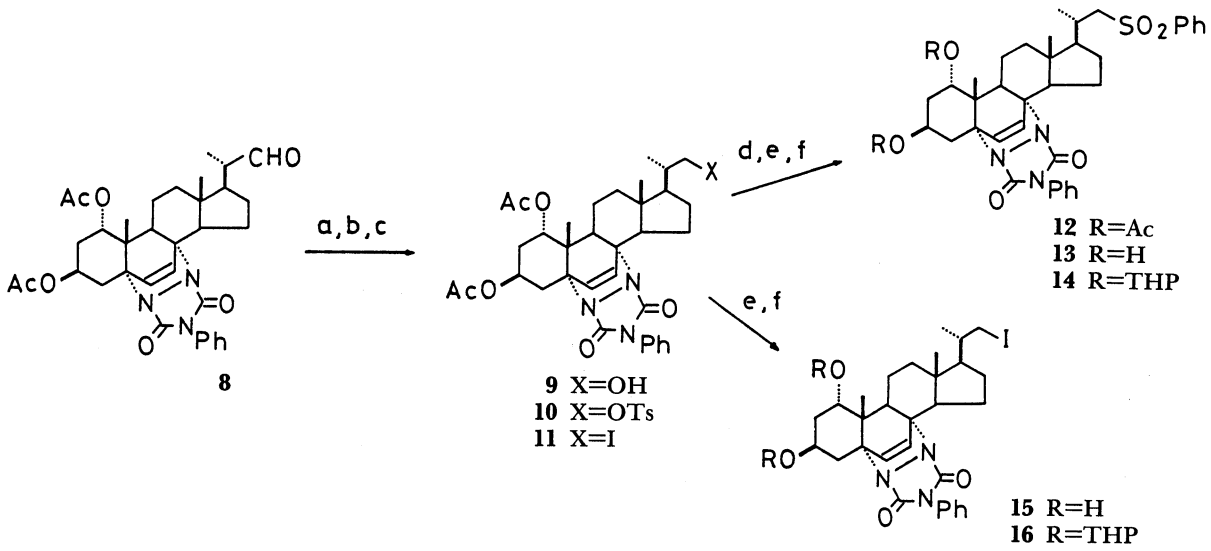
The side chain fragments (optically active iodides (**6a** and **6b**) and sulfones (**7a** and **7b**)) were synthesized, respectively, starting from methyl (*S*)- and (*R*)-3-hydroxy-2-methylpropionate (**3a** and **3b**) as follows (Scheme 1). The syntheses of **7a** and **7b** were reported previously.[6] The tosylate (**4a**), an intermediate of the




Scheme 1. a) NaI/acetone; b) DHP, PPTS.

sulfone (**7a**), was reacted with sodium iodide[9] in refluxing acetone to give the hydroxy iodide (**5a**) in 70% yield. The hydroxyl group of **5a** was protected as a tetrahydropyranyl (THP) ether in the usual manner to afford the iodide (**6a**) in 93% yield. Similarly, **6b** was prepared via the tosylate (**4b**).

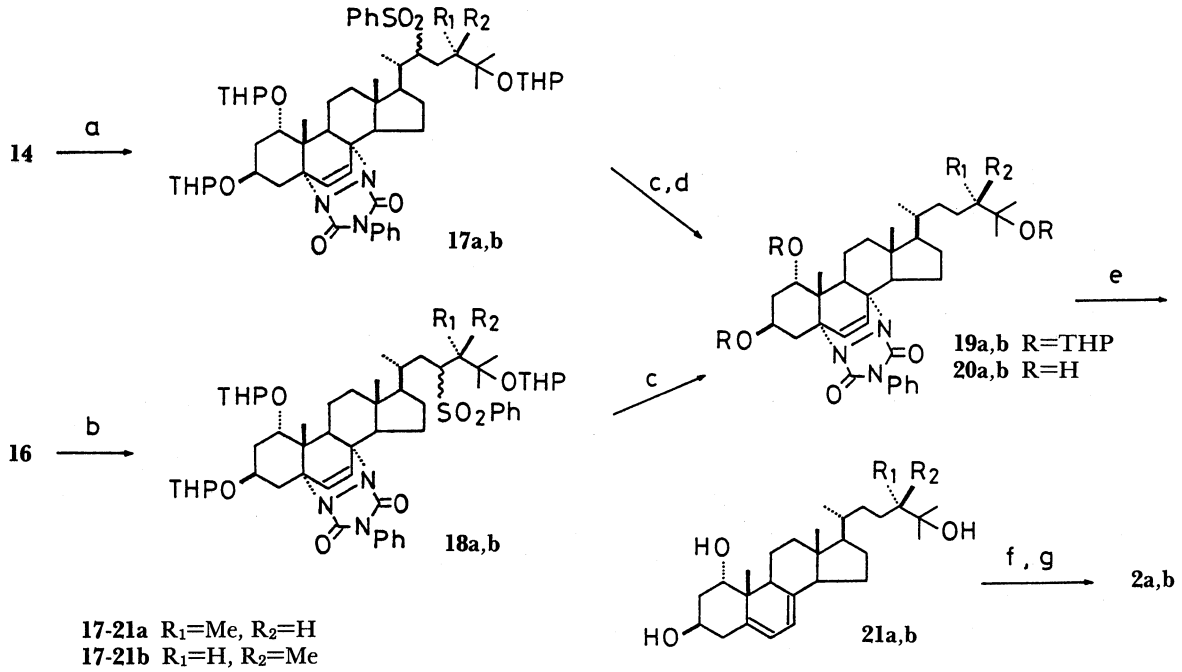
The C-22 steroidal blocks, the C-22 sulfone (**14**) and the C-22 iodide (**16**) were synthesized as follows (Scheme 2). 22-Oxo-5α,8α-(4-phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-23,24-dinor-6-cholene-1α,3β-diyl diacetate (**8**), prepared by a method described in our previous paper,[6] seemed to be a good key intermediate for the preparation of **2a** and **2b** as the steroidal block. Thus, the aldehyde (**8**) was reduced by $NaBH_4$ in MeOH to afford the alcohol (**9**),[10] which was tosylated in the conventional manner to give the tosylate (**10**) in 78% yield from **8**. Treatment of **10** with sodium iodide in DMF was followed by a reaction with




Scheme 2. a) $NaBH_4$/MeOH; b) *p*-TsCl, Py; c) NaI/DMF; d) $PhSO_2Na$/DMF; e) NaOH or KOH/MeOH; f) DHP, PPTS or *p*-TsOH.

sodium benzenesulfinate to yield the sulfone (**12**) in 82% yield.[11] The acetyl groups of **12** were converted to stable protective groups under the basic conditions employed in the following steps.[10] Thus, the hydrolysis of **12** yielded the diol (**13**), which was protected as THP groups in the usual manner to give the steroidal C-22 sulfone (**14**) in 71% yield. Similarly, the conversion of the acetyl groups of **11** to the THP groups via the diol (**15**) gave the steroidal C-22 iodide (**16**) in 73% yield.

The next task was to introduce the side chain moiety (Scheme 3). Lithiation of the C-22 sulfone (**14**) by butyllithium in the presence of hexamethylphosphoric triamide (HMPA) in THF at −20 °C followed by the addition of the iodide (**6a**) gave the 22-phenylsulfonyl derivative (**17a**) in 44% yield (72% based on the consumed **14**) together with the recovered starting sulfone (**14**) in 38% yield. Reductive desulfonylation with sodium amalgam in buffered MeOH ($Na_2HPO_4$) and subsequent deprotection of the THP groups of the resulting **19a** provided the triol (**20a**) in 43% yield. The triol (**20a**) was treated with $LiAlH_4$ in refluxing THF to remove the triazoledione protective group,[12] giving 5,7-ergostadiene-1α,3β,25-triol (**21a**)




**17-21a** $R_1$=Me, $R_2$=H
**17-21b** $R_1$=H, $R_2$=Me

Scheme 3. a) **6a** or **6b**, *n*-BuLi, HMPA/THF; b) **7a** or **7b**, *n*-BuLi, HMPA/THF; c) Na-Hg/MeOH ($Na_2HPO_4$); d) *p*-TsOH/EtOH e) $LiAlH_4$/THF; f) *hν*; g) reflux/EtOH.

in 71% yield.

The introduction of a side chain was carried out by an alternative procedure. Condensation of the C-22 iodide (**16**) with the sulfone (**7a**) was achieved under the same conditions described above to yield the 23-phenylsulfonyl derivative (**18a**) in 60% yield. Desulfonylation of **18a** gave **19a** in 35% yield.

Irradiation of the 5,7-diene-1$\alpha$,3$\beta$,25-triol (**21a**) with a high-pressure mercury lamp using an aq 1.5% $KNO_3$ solution as a filter followed by thermal isomerization of the resulting previtamin D in refluxing EtOH furnished crystalline 22,23-dihydro-1$\alpha$,25-dihydroxyvitamin $D_2$ (**2a**) in 25% yield after purification by preparative HPLC.

Similarly, (24R)-5,7-ergostadiene-1$\alpha$,3$\beta$,25-triol (**21b**) was prepared from C-22 sulfone (**14**) and iodide (**6b**) in place of **6a** in four steps with a 32% yield from **14**. Irradiation of **21b** and subsequent thermal isomerization of the resulting previtamin D gave crystalline (24*R*)-22,23-dihydro-1$\alpha$,25-dihydroxyvitamin $D_2$ (**2b**) in 17% yield.

Since 22,23-dihydro-1$\alpha$,25-dihydroxyvitamin $D_2$ (**2a**) and its 24*R*-epimer (**2b**) were prepared by two synthetic methods, the present study provides efficient routes to 1$\alpha$,25-dihydroxyvitamin D derivatives having the C-22,23 single bond.

The biological activities of **2a** and **2b** will be reported elsewhere.

## Experimental

All melting and boiling points are uncorrected. IR spectra were measured on a Jasco IR-810 spectrometer. $^1$H NMR spectra were recorded with TMS as an internal standard and $CDCl_3$ as a solvent at 200 MHz on a JEOL JNM-FX 200 spectrometer, unless otherwise stated. Optical rotations were measured with $CHCl_3$ as a solvent on a Jasco DIP-370 polarimeter, unless otherwise stated. Mass spectra were recorded on a Hitachi M-80 spectrometer at 70 eV. Merck Kieselgel 60 (Art 7734, 70—230 mesh) or Merck Kieselgel 60 (Art 9385, 230—400 mesh) were used for $SiO_2$ column chromatography.

**(3*R*)-4-Iodo-2,3-dimethyl-2-butanol THP Ether (6a).** The tosylate (**4a**) was prepared from (*S*)-2,3-dimethyl-1,3-butanediol[6] (6.40 g, 54.2 mmol) in the same manner as described previously.[6] A solution of **4a** and sodium iodide (24.4 g, 0.16 mol) in acetone (180 ml) was stirred for 5 h at reflux temperature. After removing the acetone in vacuo, water was added to the residue and the mixture was extracted with ether. The ether solution was washed with a 10% $Na_2S_2O_3$ solution and brine, dried ($MgSO_4$), and concentrated in vacuo. The residue was distilled to give 8.26 g (70%) of **5a**: bp 69—71 °C/4 mmHg (1 mmHg≈133.322 Pa); $n_D^{25}$ 1.5192; $[\alpha]_D^{23}$ −37.7° (*c* 1.98); IR (film) 3400, 1470, 1380, 1190, 1135, 1110, 950 cm$^{-1}$; $^1$H NMR $\delta$=1.11 (3H, d, *J*=6.8 Hz), 1.16 (3H, s), 1.26 (3H, s), 1.69 (1H, s), 1.87 (1H, m), 2.91 (1H, dd, *J*=9.5 and 10.5 Hz), 3.67 (1H, dd, *J*=9.5 and 7.2 Hz).

A solution of **5a** (7.73 g, 33.9 mmol), dihydropyran (5.70 g, 67.8 mmol) and pyridinium *p*-toluenesulfonate (PPTS) (0.85 g, 3.4 mmol) in dry $CH_2Cl_2$ (70 ml) was stirred for 3 h at room temperature. The mixture was washed with a sat. $NaHCO_3$ solution and brine, dried ($MgSO_4$), and concentrated in vacuo. The residue was chromatographed over $SiO_2$ (130 g) eluting with hexane–ether (19 : 1) to give 9.88 g (93%) of **6a**: IR (film) 1470, 1390, 1375, 1200, 1130, 1075, 1035, 1025, 985 cm$^{-1}$. This was employed in the next step without further purification.

**(3*S*)-4-Iodo-2,3-dimethyl-2-butanol THP Ether (6b).** In the same manner as described for **5a**, (*R*)-2,3-dimethyl-1,3-butanediol[6] (6.85 g, 58.1 mmol) was converted to 10.20 g (77%) of **5b**: bp 69—71 °C/4 mmHg; $n_D^{23}$ 1.5190; $[\alpha]_D^{23}$ +38.7° (*c* 1.93). Its IR and $^1$H NMR spectra were identical with those of **5a**.

In the same manner as described for **6a**, **5b** (8.60 g, 37.7 mmol) was converted to 10.88 g (92%) of **6b**. Its IR spectrum was identical with that of **6a**.

**5$\alpha$,8$\alpha$-(4-Phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-23,24-dinor-6-cholene-1$\alpha$,3$\beta$,22-triyl 1$\alpha$,3$\beta$-Diacetate 22-*p*-Toluenesulfonate (10).** To a stirred solution of **8** (9.50 g, 15.8 mmol) in MeOH (100 ml) was added $NaBH_4$ (0.30 g, 7.9 mmol) portionwise over 10 min at room temperature; the mixture was stirred for an additional 10 min. To the mixture was added AcOH (0.3 ml); the mixture was stirred for 10 min. After removal of MeOH in vacuo, water was added to the residue and the mixture was extracted with $CHCl_3$. The $CHCl_3$ solution was washed with water and brine, dried ($MgSO_4$), and concentrated in vacuo to give 9.50 g of crude alcohol (**9**).[10] This was employed in the next step without further purification.

To a stirred, ice-cooled solution of **9** (9.50 g) in dry pyridine (45 ml) was added *p*-toluenesulfonyl chloride (4.50 g, 23.6 mmol); the mixture was stirred for 4 h at room temperature. To the reaction mixture was added a small amount of water and the mixture was stirred for 1 h at that temperature. The mixture was next poured into ice water and extracted with $CHCl_3$. The $CHCl_3$ solution was washed with water, 5% HCl, sat. $NaHCO_3$ solution and brine, dried ($MgSO_4$), and concentrated in vacuo. The residue was chromatographed over $SiO_2$ (250 g) eluting with hexane–EtOAc (1 : 1—1 : 2) to give 9.33 g (78% from **8**) of **10**: IR (KBr) 1750, 1700, 1600, 1505, 1400, 1245, 1180, 1030 cm$^{-1}$; $^1$H NMR $\delta$=0.80 (3H, s, 18-H$_3$), 1.01 (3H, d, *J*=6.6 Hz, 21-H$_3$), 1.05 (3H, s, 19-H$_3$), 2.01 (3H, s, Ac), 2.02 (3H, s, Ac), 2.44 (3H, s, -CH$_3$(tosyl)), 3.25 (1H, dd, *J*=13.4 and 5.6 Hz, 9-H), 3.73 (1H, dd, *J*=8.8 and 6.6 Hz, 22-H), 4.01 (1H, dd, *J*=8.8 and 2.4 Hz, 22-H), 5.09 (1H, m, 1-H), 5.87 (1H, m, 3-H), 6.33 and 6.41 (2H, ABq, *J*=8.3 Hz, 6-H and 7-H), 7.32—7.50 (8H, m, Ph), 7.77 (2H, d, *J*=8.1 Hz, Ph); MS *m/z* (rel intensity) 524 (M$^+$-PTAD-AcOH; 3), 464 (58), 292 (43), 277 (16), 177 (62), 155 (100), 119 (78).

**22-Iodo-5$\alpha$,8$\alpha$-(4-phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-23,24-dinor-6-cholene-1$\alpha$,3$\beta$-diyl Diacetate (11).** A solution of **10** (2.61 g, 3.44 mmol) and sodium iodide (2.57 g, 17.1 mmol) in dry DMF (20 ml) was stirred for 30 min at 80 °C. After cooling, the mixture was poured into water and extracted with $CHCl_3$. The $CHCl_3$ solution was washed with water, a 5% $Na_2S_2O_3$ solution and brine, dried ($MgSO_4$), and concentrated in vacuo. The residue was chromatographed over $SiO_2$ (50 g) eluting with hexane–EtOAc (3 : 2—1 : 1) to give 2.33 g (95%) of **11**: mp 173—174 °C (from hexane–EtOAc, rods); $[\alpha]_D^{23}$ −64.4° (*c* 1.12); IR (KBr) 1740, 1685, 1600, 1505, 1410, 1250, 1230, 1030 cm$^{-1}$; $^1$H NMR $\delta$=0.87 (3H, s, 18-H$_3$), 1.04 (3H, d, *J*=6.6 Hz, 21-H$_3$), 1.06

(3H, s, 19-$H_3$), 2.01 (3H, s, Ac), 2.04 (3H, s, Ac), 3.12—3.36 (3H, m, 9-H and 22-$H_2$), 5.11 (1H, m, 1-H), 5.88 (1H, m, 3-H), 6.34 and 6.44 (2H, ABq, $J$=8.3 Hz, 6-H and 7-H), 7.28—7.51 (5H, m, Ph); MS $m/z$ (rel intensity) 540 ($M^+$-PTAD; 0.3), 480 (8), 420 (95), 251 (20), 141 (100), 119 (65). Found: C, 57.25; H, 5.93; N, 5.82%. Calcd for $C_{34}H_{42}N_3O_6I$: C, 57.07; H, 5.92; N, 5.87%.

**22-Phenylsulfonyl-5α,8α-(4-phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-23,24-dinor-6-cholene-1α,3β-diyl Diacetate (12).** A solution of **10** (9.30 g, 12.3 mmol) and sodium iodide (9.19 g, 61.3 mmol) in dry DMF (80 ml) was stirred for 30 min at 80 °C. To the mixture was next added sodium benzenesulfinate (4.20 g, 24.5 mmol); this mixture was stirred for 30 min at 80 °C. The mixture was then poured into ice water and extracted with $CHCl_3$. The $CHCl_3$ solution was washed with water, a 5% $Na_2S_2O_3$ solution and brine, dried ($MgSO_4$), and concentrated in vacuo. The residue was chromatographed over $SiO_2$ (200 g) eluting with hexane-EtOAc (1:1—1:2) to give 7.31 g (82%) of **12**: mp 176—178 °C (from hexane-EtOAc, needles); $[\alpha]_D^{23}$ −99.3° ($c$ 1.20); IR (KBr) 1750, 1695, 1600, 1505, 1400, 1305, 1250, 1235, 1145 $cm^{-1}$; $^1H$ NMR δ=0.83 (3H, s, 18-$H_3$), 1.05 (3H, s, 19-$H_3$), 1.21 (3H, d, $J$=6.4 Hz, 21-$H_3$), 2.01 (3H, s, Ac), 2.02 (3H, s, Ac), 2.84 (1H, dd, $J$=13.8 and 9.4 Hz, 22-H), 3.13 (1H, d, $J$=13.8 Hz, 22-H), 3.25 (1H, dd, $J$=13.9 and 5.6 Hz, 9-H), 5.09 (1H, m, 1-H), 5.89 (1H, m, 3-H), 6.33 and 6.41 (2H, ABq, $J$=8.3 Hz, 6-H and 7-H), 7.30—7.70 (8H, m, Ph), 7.89 (2H, m, Ph); MS $m/z$ (rel intensity) 494 ($M^+$-PTAD-AcOH; 10), 435 (100), 251 (18), 177 (48), 141 (82). Found: C, 65.75; H, 6.53; N, 5.78%. Calcd for $C_{40}H_{47}N_3O_8S$: C, 65.82; H, 6.49; N, 5.76%.

**22-Phenylsulfonyl-5α,8α-(4-phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-23,24-dinor-6-cholene-1α,3β-diol (13).** A mixture of **12** (7.31 g, 10.0 mmol) and potassium hydroxide (1.12 g, 20.0 mmol) in MeOH (100 ml) was stirred at reflux temperature for 30 min. After cooling, a crystallized material was filtered off to give 5.12 g (79%) of **13** as needles: mp 240—242 °C; $[\alpha]_D^{25}$ −87.3° ($c$ 0.49); IR (KBr) 3540, 3470, 1740, 1675, 1505, 1410, 1310, 1155, 1090, 1040 $cm^{-1}$; $^1H$ NMR δ=0.82 (3H, s, 18-$H_3$), 0.90 (3H, s, 19-$H_3$), 1.23 (3H, d, $J$=6.4 Hz, 21-$H_3$), 2.82 (1H, dd, $J$=13.7 and 8.1 Hz, 22-H), 3.10 (2H, m, 9-H and 22-H), 3.81 (1H, m, 1-H), 4.84 (1H, m, 3-H), 6.25 and 6.36 (2H, ABq, $J$=8.1 Hz, 6-H and 7-H), 7.30—7.70 (8H, m, Ph), 7.91 (2H, m, Ph); MS $m/z$ (rel intensity) 470 ($M^+$-PTAD; 5), 452 (2), 434 (4), 239 (21), 177 (53), 119 (100). Found: C, 66.74; H, 6.75; N, 6.47%. Calcd for $C_{36}H_{43}N_3O_6S$: C, 66.95; H, 6.71; N, 6.51%.

**22-Phenylsulfonyl-5α,8α-(4-phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-1α,3β-bis(tetrahydropyranyloxy)-23,24-dinor-6-cholene (14).** A solution of **13** (5.12 g, 7.9 mmol), dihydropyran (2.67 g, 31.8 mmol) and PPTS (0.40 g, 1.6 mmol) in dry $CH_2Cl_2$ (50 ml) was stirred for 24 h at room temperature. The mixture was washed with a sat. $NaHCO_3$ solution and brine, dried ($MgSO_4$), and concentrated in vacuo. The residue was chromatographed over $SiO_2$ (100 g) eluting with hexane-EtOAc (2:1), and then crystallized from hexane-ether to give 5.81 g (90%) of **14** as needles: mp 181—186 °C; $[\alpha]_D^{24}$ −84.9° ($c$ 1.03); IR (KBr) 1750, 1695, 1605, 1505, 1400, 1305, 1150, 1030 $cm^{-1}$; $^1H$ NMR δ=0.83 (3H, s, 18-$H_3$), 0.95 and 0.98 (3H, pair of s, 19-$H_3$), 1.23 (3H, d, $J$=6.4 Hz, 21-$H_3$), 2.85 (1H, m, 22-H), 3.15 (2H, m, 9-H and 22-H), 3.50 (2H, m, $CH_2$(THP)), 3.70 (1H, m, 1-H), 3.90 (2H, m, $CH_2$(THP)), 4.75 (2H, m, CH(THP)), 4.85 (1H, m, 3-H), 6.32 (2H, m, 6-H and 7-H), 7.3—7.7 (8H, m, Ph), 7.91 (2H, m, Ph); MS $m/z$ (rel intensity) 638 ($M^+$-PTAD; 0.5), 554 (2), 536 (2), 239 (13), 177 (62), 119 (100). Found: C, 67.39; H, 7.31; N, 5.05%. Calcd for $C_{46}H_{59}N_3O_8S$: C, 67.87; H, 7.30; N, 5.16%.

**22-Iodo-5α,8α-(4-phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-1α,3β-bis(tetrahydropyranyloxy)-23,24-dinor-6-cholene (16).** A mixture of **11** (1.23 g, 1.72 mmol) and sodium hydroxide (0.14 g, 3.5 mmol) in MeOH (20 ml) was stirred for 30 min at reflux temperature. After removal of MeOH in vacuo, water was added to the residue and the mixture was extracted with $CHCl_3$. The $CHCl_3$ solution was washed with brine, dried ($MgSO_4$), and concentrated in vacuo to give 1.05 g of crude **15**. This was employed in the next step without further purification. An analytical sample was prepared by recrystallization from $CHCl_3$-EtOAc: mp 172—174 °C (rods); $[\alpha]_D^{23}$ −65.4° ($c$ 1.14); IR (KBr) 3420, 1745, 1680, 1600, 1505, 1400, 1150, 1090, 1030 $cm^{-1}$; $^1H$ NMR δ=0.84 (6H, br s, 18-$H_3$ and 19-$H_3$), 1.05 (3H, d, $J$=5.6 Hz, 21-$H_3$), 3.02—3.35 (3H, m, 9-H and 22-$H_2$), 3.70 (1H, m, 1-H), 4.80 (1H, m, 3-H), 6.20 and 6.34 (2H, ABq, $J$=8.3 Hz, 6-H and 7-H), 7.29—7.40 (5H, m, Ph); MS $m/z$ (rel intensity) 456 ($M^+$-PTAD; 13), 438 (5), 436 (11), 420 (10), 410 (20), 328 (5), 251 (15), 177 (68), 119 (100).

A solution of crude **15** (1.05 g), dihydropyran (0.43 g, 5.12 mmol) and a catalytic amount of $p$-TsOH·$H_2O$ in dry $CH_2Cl_2$ (20 ml) was stirred for 24 h at room temperature. The mixture was washed with sat. $NaHCO_3$ solution and brine, dried ($MgSO_4$), and concentrated in vacuo. The residue was chromatographed over $SiO_2$ (30 g) eluting with hexane-EtOAc (2:1) to give 1.00 g (73% from **11**) of **16** as a foam: IR (KBr) 1750, 1690, 1600, 1505, 1400, 1130, 1115, 1030 $cm^{-1}$; $^1H$ NMR δ=0.87 (3H, s, 18-$H_3$), 0.96 and 0.99 (3H, pair of s, 19-$H_3$), 1.05 (3H, d, $J$=5.9 Hz, 21-$H_3$), 3.10—3.65 (5H, m, 9-H, 22-$H_2$ and $CH_2$ (THP)), 3.70 (1H, m, 1-H), 3.90 (2H, m, $CH_2$(THP)), 4.75 (2H, m, CH(THP)), 4.95 (1H, m, 3-H), 6.30—6.45 (2H, m, 6-H and 7-H), 7.30—7.50 (5H, m, Ph); MS $m/z$ (rel intensity) 624 ($M^+$-PTAD; 0.8), 540 (2), 454 (10), 437 (48), 420 (23), 382 (15), 309 (10), 251 (12), 177 (48), 119 (100).

**22-Phenylsulfonyl-5α,8α-(4-phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-1α,3β,25-tris(tetrahydropyranyloxy)-6-ergostene (17a).** To a stirred solution of **14** (3.50 g, 4.3 mmol) in dry THF (35 ml) was added successively butyllithium (1.5 mol $dm^{-3}$ in hexane, 3.4 ml, 4.3 mmol) and dry HMPA (2.26 ml, 12.9 mmol) at −78 °C under Ar, and the mixture was stirred for 20 min at −20 °C. To the mixture was added a solution of **6a** (4.30 g, 12.9 mmol) in dry THF (12 ml); the mixture was stirred for 1.5 h at −20 °C. The mixture was then poured into a sat. $NH_4Cl$ solution and extracted with $CHCl_3$. The $CHCl_3$ solution was washed with brine, dried ($MgSO_4$), and concentrated in vacuo. The residue was chromatographed over $SiO_2$ (200 g) eluting with hexane-EtOAc (2:1—3:2—1:1). The earlier fraction gave 1.90 g (44%; 72% based on the consumed **14**) of **17a**, and the latter fraction gave 1.34 g (38%) of the recovered **14**: **17a**, IR (KBr) 1750, 1695, 1605, 1505, 1400, 1150, 1130, 1075, 1030, 985 $cm^{-1}$; $^1H$ NMR δ=3.05 (1H, m, 22-H), 3.22 (1H, m, 9-H), 3.48 (3H, m, $CH_2$(THP)), 3.69 (1H, m, 1-H), 3.93 (3H, m, $CH_2$ (THP)), 4.78 (3H, m, CH(THP)), 4.93 (1H, m, 3-H), 6.33 (2H, m, 6-H and 7-H), 7.3—7.9 (10H, m, Ph); MS $m/z$ (rel intensity) 552 ($M^+$-PTAD-DHP×3-$H_2O$; 16), 534 (21), 177 (60), 119 (100).

**(24*R*)-22-Phenylsulfonyl-5α,8α-(4-phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-1α,3β,25-tris(tetrahydropyranyloxy)-6-ergostene (17b).** In the same manner as described for **17a**, **14** (2.55 g, 3.1 mmol) and **6b** (2.93 g, 9.4 mmol) was converted to 1.43 g (46%; 84% based on the consumed **14**) of **17b** together with 1.17 g (46%) of the recovered **14**: **17b**, IR (KBr) 1750, 1695, 1605, 1505, 1400, 1150, 1130, 1080, 1030, 985 $cm^{-1}$; $^1H$ NMR δ=3.18 (2H, m, 9-H and 22-H), 3.48 (3H, m, $CH_2$(THP)), 3.70 (1H, m, 1-H), 3.93 (3H, m, $CH_2$(THP)), 4.80 (3H, m, CH(THP)), 4.92 (1H, m, 3-H), 6.32 (2H, m, 6-H and 7-H), 7.3—7.9 (10H, m, Ph).

**23-Phenylsulfonyl-5α,8α-(4-phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-1α,3β,25-tris(tetrahydropyranyloxy)-6-ergostene (18a).** To a stirred solution of **7a** (326 mg, 1.0 mmol) in dry THF (3 ml) was added successively butyllithium (1.5 mol $dm^{-3}$ in hexane, 0.67 ml, 1.0 mmol) and dry HMPA (0.17 ml, 1.0 mmol) at −78 °C under Ar; the mixture was stirred for 20 min at −20 °C. To the mixture was next added a solution of **16** (400 mg, 0.50 mmol) in dry THF (4 ml). After stirring for 2 h at −20 °C, the mixture was stirred for 2 h at room temperarure, poured into a sat. $NH_4Cl$ solution, and extracted with $CHCl_3$. The $CHCl_3$ solution was washed with brine, dried ($MgSO_4$), and concentrated in vacuo. The residue was chromatographed over $SiO_2$ (20 g) eluting with hexane-EtOAc (4:1) to give 317 mg (64%) of **18a**: IR (KBr) 1750, 1695, 1600, 1500, 1400, 1140, 1125, 1030 $cm^{-1}$; $^1H$ NMR δ=3.1—3.6 (5H, m, 9-H, 23-H and $CH_2$(THP)), 3.67 (1H, m, 1-H), 3.87 (3H, m, $CH_2$(THP)), 4.75 (3H, m, CH(THP)), 4.95 (1H, m, 3-H), 6.33 (2H, m, 6-H and 7-H), 7.3—7.9 (10H, m, Ph); MS *m/z* (rel intensity) 570 ($M^+$-PTAD-DHP×3; 3), 552 (4), 534 (4), 177 (57), 119 (100).

**5α,8α-(4-Phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-1α,3β,25-tris(tetrahydropyranyloxy)-6-ergostene (19a).** (a) (from **17a**) To a solution of **17a** (1.20 g, 1.2 mmol) in MeOH saturated with $Na_2HPO_4$ (120 ml) was added sodium amalgam (5%, 16.6 g, 36.0 mmol), and the mixture was stirred for 16 h at room temperature. After removal of MeOH in vacuo from the supernatant, water was added to the residue and the mixture extracted with $CHCl_3$. The $CHCl_3$ solution was washed with water and brine, dried ($MgSO_4$), and concentrated in vacuo. The residue was chromatographed over $SiO_2$ (20 g) eluting with hexane-EtOAc (3:1—2:1) to give 0.51 g (50%) of **19a**: IR (KBr) 1750, 1700, 1605, 1505, 1400, 1135, 1080, 1030, 985 $cm^{-1}$; $^1H$ NMR δ=3.22 (1H, m, 9-H), 3.47 (3H, m, $CH_2$(THP)), 3.70 (1H, m, 1-H), 3.92 (3H, m, $CH_2$(THP)), 4.78 (3H, m, CH(THP)), 4.93 (1H, m, 3-H), 6.37 (2H, m, 6-H and 7-H), 7.3—7.5 (5H, m, Ph); MS *m/z* (rel intensity) 598 ($M^+$-PTAD-DHP; 4), 580 (1), 412 (80), 239 (18), 177 (85), 119 (100).

(b) (from **18a**) In the same manner as described above, **18a** (300 mg, 0.30 mmol) was converted to 90 mg (35%) of **19a**. Its IR and $^1H$ NMR spectra were identical with those described above.

**(24*R*)-5α,8α-(4-Phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-1α,3β,25-tris(tetrahydropyranyloxy)-6-ergostene (19b).** In the same manner as described for **19a**, **17b** (1.17 g, 1.7 mmol) was converted to 0.78 g (53%) of **19b**: IR (KBr) 1750, 1695, 1600, 1505, 1395, 1130, 1075, 1025, 985 $cm^{-1}$; $^1H$ NMR δ=3.20 (1H, m, 9-H), 3.47 (3H, m, $CH_2$(THP)), 3.70 (1H, m, 1-H), 3.92 (3H, m, $CH_2$(THP)), 4.78 (3H, m, CH(THP)), 4.93 (1H, m, 3-H), 6.37 (2H, m, 6-H and 7-H), 7.3—7.5 (5H, m, Ph).

**5α,8α-(4-Phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-6-ergostene-1α,3β,25-triol (20a).** A solution of **19a** (0.51 g, 0.60 mmol) and *p*-TsOH·$H_2O$ (23 mg, 0.12 mmol) in 95% EtOH (5 ml) was stirred for 4 h at 80 °C. After removal of EtOH in vacuo, brine was added to the residue and the mixture was extracted with $CHCl_3$. The $CHCl_3$ solution was dried ($MgSO_4$) and concentrated in vacuo. The residue was chromatographed over $SiO_2$ (25 g) eluting with $CHCl_3$-EtOAc (1:3) and then EtOAc to give 0.31 g (85%) of **20a**: mp 211—214 °C (from EtOAc, rods); $[\alpha]_D^{22}$ −97.7° (*c* 0.31); IR (KBr) 3530, 3460, 1745, 1680, 1505, 1410, 1320, 1150, 1035 $cm^{-1}$; $^1H$ NMR δ=0.81 (3H, s, 18-$H_3$), 0.88 (3H, d, *J*=7.1 Hz, 28-$H_3$), 0.92 (3H, s, 19-$H_3$), 0.94 (3H, d, *J*=6.4 Hz, 21-$H_3$), 1.14 and 1.15 (6H, each s, 26-$H_3$ and 27-$H_3$), 3.12 (1H, dd, *J*=15.6 and 6.1 Hz, 9-H), 3.85 (1H, m, 1-H), 4.88 (1H, m, 3-H), 6.25 and 6.41 (2H, ABq, *J*=8.5 Hz, 6-H and 7-H), 7.3—7.4 (5H, m, Ph); MS *m/z* (rel intensity) 430 ($M^+$-PTAD; 13), 412 (12), 394 (11), 251 (17), 199 (41), 119 (100). Found: *m/z* 430.3444. Calcd for $C_{28}H_{46}O_3$ ($M^+$-PTAD): M, 430.3449.

**(24*R*)-5α,8α-(4-Phenyl-3,5-dioxo-1,2,4-triazolidine-1,2-diyl)-6-ergostene-1α,3β,25-triol (20b).** In the same manner as described for **20a**, **19b** (0.77 g, 0.90 mmol) was converted to 0.45 g (83%) of **20b**: mp 216—218 °C (from EtOAc-MeOH, needles); $[\alpha]_D^{22}$ −79.0° (*c* 0.36); IR (KBr) 3520, 1745, 1680, 1505, 1410, 1325, 1155, 1035 $cm^{-1}$; $^1H$ NMR δ=0.82 (3H, s, 18-$H_3$), 0.87 (3H, d, *J*=6.8 Hz, 28-$H_3$), 0.93 (3H, s, 19-$H_3$), 0.95 (3H, d, *J*=6-7 Hz, 21-$H_3$), 1.15 and 1.17 (6H, each s, 26-$H_3$ and 27-$H_3$), 3.15 (1H, dd, *J*=16.4 and 7.1 Hz, 9-H), 3.88 (1H, m, 1-H), 4.90 (1H, m, 3-H), 6.27 and 6.42 (2H, ABq, *J*=6.8 Hz, 6-H and 7-H), 7.30—7.42 (5H, m, Ph); MS *m/z* (rel intensity) 430 ($M^+$-PTAD; 8), 251 (15), 177 (44), 119 (100). Found: C, 71.39; H, 8.52; N, 7.01%. Calcd for $C_{36}H_{51}N_3O_5$: C, 71.37; H, 8.49; N, 6.94%.

**5,7-Ergostadiene-1α,3β,25-triol (21a).** To a suspension of $LiAlH_4$ (0.40 g) in dry THF (30 ml) was added a solution of **20a** (0.44 g, 0.73 mmol) in dry THF (10 ml); the mixture was stirred for 1.5 h at reflux temperature. Then to the ice-cooled mixture was added successively water (0.4 ml), a 10% NaOH solution (0.4 ml) and water (1.2 ml); the mixture was stirred for 30 min at room temperature. After addition of $MgSO_4$, the mixture was stirred for an additional 30 min and filtered through Celite. The filtrate was concentrated in vacuo; thus, the residue was recrystallized from EtOH-THF to give 0.22 g (71%) of **21a** as needles: mp 228—231 °C; $[\alpha]_D^{22}$ −89° (*c* 0.11, THF); IR (KBr) 3520, 3360, 1655, 1605, 1465, 1380, 1135, 1070, 1045 $cm^{-1}$; $^1H$ NMR (DMSO-$d_6$+$CDCl_3$) δ=0.60 (3H, s, 18-$H_3$), 0.84 (3H, d, *J*=6.6 HZ, 28-$H_3$), 0.85 (3H, s, 19-$H_3$), 0.95 (3H, d, *J*=6.1 Hz, 21-$H_3$), 1.07 and 1.08 (6H, each s, 26-$H_3$ and 27-$H_3$), 3.62 (1H, m, 1-H), 3.91 (1H, m, 3-H), 5.30 (1H, m, 7-H), 5.56 (1H, m, 6-H); MS *m/z* (rel intensity) 430 ($M^+$; 55), 412 (85), 394 (31), 251 (40), 197 (64), 157 (100), 145 (68); UV (EtOH) 282 nm (ε 9200). Found: C, 77.59; H, 10.70%. Calcd for $C_{28}H_{46}O_3$: C, 78.09; H, 10.77%.

**(24*R*)-5,7-Ergostadiene-1α,3β,25-triol (21b).** In the same manner as described for **21a**, **20b** (0.45 g, 0.74 mmol) was converted to 0.28 g (86%) of **21b**: mp 154—157 °C (from EtOH, rods); $[\alpha]_D^{22}$ −17° (*c* 0.12, MeOH); IR (KBr) 3400, 1655, 1605, 1465, 1385, 1155, 1055 $cm^{-1}$; $^1H$ NMR δ=0.63 (3H, s, 18-$H_3$), 0.88 (3H, d, *J*=6.6 Hz, 28-$H_3$), 0.95 (3H, d, *J*=6.1 Hz, 21-$H_3$), 0.95 (3H, s, 19-$H_3$), 1.16 and 1.17 (6H, each s, 26-$H_3$ and 27-$H_3$), 3.78 (1H, m, 1-H), 4.08 (1H, m, 3-H), 5.40 (1H, m, 7-H), 5.73 (1H, m, 6-H); MS *m/z* (rel intensity) 430 ($M^+$; 32), 412 (20), 394 (18), 251 (35), 197 (64), 157 (100), 145 (65). Found: *m/z* 430.3443. Calcd for $C_{28}H_{46}O_3$: M, 430.3449.

**22,23-Dihydro-1α,25-dihydroxyvitamin $D_2$ (2a).** A solution of **21a** (100 mg, 0.23 mmol) in ether-THF (19:1, 1000 ml) was irradiated for 3 min under Ar at water-cooled temperature with a high-pressure mercury lamp (Ushio, UM-452) using 1.5% $KNO_3$ solution as a filter. The mixture was concentrated in vacuo. A solution of the residue containing the previtamin D in EtOH (30 ml) was stirred for 1 h at reflux temperature under Ar, and then concentrated in vacuo. The residue was chromatographed on HPLC (Merck, LiChrosorb® Si60 (7 μm), 25×250 mm) eluting with 6% MeOH-$CH_2Cl_2$ (6.0 ml $min^{-1}$) to give 25 mg (25% from **21a**) of crystalline **2a**. This was recrystallized from hexane-$CH_2Cl_2$ to give **2a** as needles: mp 93—95 °C; $[\alpha]_D^{22}$ +33° (*c* 0.15, EtOH); $^1H$ NMR δ=0.54 (3H, s, 18-$H_3$), 0.90 (3H, d, *J*=6.8 Hz, 28-$H_3$), 0.94 (3H, d, *J*=5.9 Hz, 21-$H_3$), 1.15 and 1.17 (6H, each s, 26-$H_3$ and 27-$H_3$), 4.23 (1H, m, 3-H), 4.43 (1H, m, 1-H), 5.00 (1H, narrow m, 19-H), 5.33 (1H, narrow m, 19-H), 6.02 (1H, d, *J*=11.2 Hz, 7-H), 6.38 (1H, d, *J*=11.2 Hz, 6-H); MS *m/z* (rel intensity) 430 ($M^+$; 8), 412 (10), 394 (11), 285 (6), 251 (5), 134 (100), 105 (34); UV (EtOH) 265 nm (ε 16900).

**(24*R*)-22,23-Dihydro-1α,25-dihydroxyvitamin $D_2$ (2b).** A solution of **21b** (100 mg, 0.23 mmol) in ether (1000 ml) was irradiated for 3 min under the same conditions as described above, and then concentrated in vacuo. The residue was chromatographed on HPLC (Merck, LiChrosorb® Si60 (7 μm), 25×250 mm) eluting with 6% MeOH-$CH_2Cl_2$ (6.0 ml $min^{-1}$) to give 25.0 mg (25%) of the previtamin D: $^1H$ NMR δ=0.70 (3H, s, 18-$H_3$), 0.88 (3H, d, *J*=6.8 Hz, 28-$H_3$), 0.95 (3H, d, *J*=6.1 Hz, 21-$H_3$), 1.17 (6H, br s, 26-$H_3$ and 27-$H_3$), 1.77 (3H, s, 19-$H_3$), 4.06 (1H, m, 3-H), 4.20 (1H, m, 1-H), 5.50 (1-H, m, 9-H), 5.78 and 5.92 (2H, ABq, *J*=12.2 Hz, 6-H and 7-H).

A solution of the previtamin D (25.0 mg) in EtOH (15 ml) was stirred for 1 h at reflux temperature, and concentrated in vacuo. The residue was chromatographed on HPLC under the same conditions as mentioned above to give 16.7 mg (67%; 17% from **21b**) of crystalline **2b**. This was recrystallized from hexane-$CH_2Cl_2$ to give **2b** as rods: mp 172—174 °C; $[\alpha]_D^{22}$ +63° (*c* 0.11, EtOH); $^1H$ NMR δ=0.54 (3H, s, 18-$H_3$), 0.88 (3H, d, *J*=6.8 Hz, 28-$H_3$), 0.93 (3H, d, *J*=6.1 Hz, 21-$H_3$), 1.16 and 1.17 (6H, each s, 26-$H_3$ and 27-$H_3$), 4.23 (1H, m, 3-H), 4.44 (1H, m, 1-H), 5.01 (1H, narrow m, 19-H), 5.33 (1H, narrow m, 19-H), 6.02 (1H, d, *J*=11.2 Hz, 7-H), 6.38 (1H, d, *J*=11.2 Hz, 6-H); MS *m/z* (rel intensity) 430 ($M^+$; 5), 412 (11), 394 (18), 285 (5), 251 (5), 134 (100), 105 (32); UV (EtOH) 265 nm (ε 17600).

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