acted with bases at a CH_3 group rather than at the methine position, Barlett[16] assumed steric hindrance to base approach to be the overriding factor. Also steric hindrance to carbon protonation by protonated secondary amines has been reported for the reaction of benzylidene Meldrum's acid with morpholinium ion.[5,17]

Recently, morpholine and piperidine have been used as reference points to determine Brönsted $\beta^{RR'NH}$ values that were assumed to be typical for the behavior of secondary amines in various types of reaction.[9-12,18] Such $\beta^{RR'NH}$ values obtained for studies of the ionization of numerous carbon acids were generally identical with those (β^{RNH_2}) found for primary amine catalysts.[9-12] For example, β values of 0.65 and 0.69 have been reported for the deprotonation of phenylnitromethane by primary amines and the piperidine–morpholine pair, respectively, in 90% DMSO–10% H_2O.[10] Similar situations hold for the deprotonation of diphenylmethanes, e.g., 2,4,2',4'-tetranitrodiphenylmethane ($\beta^{RNH_2} = \beta^{RR'NH} = 0.45$), (α-cyanodiphenylmethane)bis(tricarbonylchromium(0)) ($\beta^{RNH_2} = 0.75$, $\beta^{RR'NH} = 0.72$), and of diketones, e.g., acetylacetone ($\beta^{RNH_2} = 0.45$, $\beta^{RR'NH} = 0.42$), 1,3-indanedione ($\beta^{RNH_2} = 0.42$, $\beta^{RR'NH} = 0.40$ in 50% DMSO–50% H_2O).[9,11b,12] These observations have implied that there is no major difference in the degree of proton transfer in the transition states for the corresponding reactions (with primary and secondary amines). However, the use of the piperidine–morpholine pair in the present work affords a $\beta^{RR'NH}$ value of 0.6 (line B of Figure 1). This value is markedly greater than that derived from line A for the primary amines (0.47), apparently indicating that proton transfer has made more progress in the transition states involving secondary amines that in those involving primary amines.

These results suggest that β values derived from data for secondary amines may not always reflect those for other classes of amines, especially in reactions involving highly sterically hindered substrates.

From the plots of Figure 1, values for the intrinsic rate constants for deprotonation of I (k_0) may be obtained.[3,19] These intrinsic rate constants are the values of k when

$$pK_a^{BH} + \log p/q = pK_a^{CH} = 8.25$$

and are found to be $\log k_0 = -2.95$ from plot A and $\log k_0 = -3.75$ from plot B, placing them among the lowest reported values.[3,11b,20] Low intrinsic rate constants are normally observed in those carbon acid ionizations that either occur with extensive solvent reorganization and/or give rise to highly delocalized carbanions.[3,21] The anion II is extremely stable in 50% aqueous DMSO and has $\lambda_{max} = 720$ nm ($\epsilon = 29\,300$), indicating extensive charge delocalization. Formation of this anion will involve considerable structural–electronic–solvational reorganization, and this will contribute significantly to the extremely low k_0 values found for I. In addition, the twist angle of each ring of II is presumably greater than that found for the triphenylmethyl anion (31–2° av) or, possibly, at any given instant one ring is orthogonal to the other two and these could then be coplanar, c.f. the 9-phenylfluorenyl anion.[22-24] In either situation there will be a considerable barrier to protonation of II and therefore a low intrinsic reactivity for I, as is observed.

Experimental Section

Materials. Tris(2,4-dinitrophenyl)methane was parepared according to the procedure of Margerum et al.[25] mp 260 °C (lit.[25] mp 256–8 °C). Solvents were purified and solutions made up as previously described.[12] Buffers were purified commercial products.

Measurements. Kinetic studies were made at 720 nm on a Durrum-Gibson 135 stopped-flow spectrophotometer with a thermostatted cell compartment (±0.5 °C). pH determinations were carried out as in other studies, on a Tacussel Isis 20000 pH meter. pK_a^{BH} values for the buffers in 50% aqueous DMSO were taken from previous studies,[12] except where otherwise indicated in Table I.

Registry No. I, 3626-18-4; aniline, 62-53-3; aminoacetonitrile, 540-61-4; glycine ethyl ester, 459-73-4; glycinamide, 598-41-4; allylamine, 107-11-9; 2-methoxyethylamine, 109-85-3; *n*-butylamine, 109-73-9; morpholine, 110-91-8; *N*-methylaminoethanol, 109-83-1; piperidine, 110-89-4; pyrrolidine, 123-75-1.

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Stereoselective Synthesis of (24*S*)- and (24*R*)-24-(Hydroxymethyl)cholesta-5,22(*E*)-dien-3β-ol: Model Compounds for Stereochemical Assignments of Polyhydroxylated Marine Steroids

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During our continuing work on polyhydroxysteroids and steroidal glycosides from echinoderms[1] we isolated a series of steroids with unusual oxygenation of the side chain. The latest additions are glycosides of polyhydroxysteroids isolated from the starfish *Coscinasterias tenuispina*[2] [coscinasteroside C; i.e. 28-*O*-β-D-glucopyranosyl-24-methyl-5α-cholest-22(*E*)-ene-3β,6α,8,15β,16β,28-hexol 4'-sulfate], *Pisaster brevispinus*,[3] and a steroid isolated from the ophiuroid *Ophiolepis superba*,[4] all possessing a Δ^{22}-24-hydroxymethyl side chain. The structures of these compounds were determined by means of spectral data and some chemical transformations, but the stereochemistry at C-24 remained to be defined. The growing number of naturally occurring steroids with such a structural feature, and the limited amount of natural compound usually available, made it desirable to develop a technique using

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Table I. 250-MHz ^{1}H NMR Data ($CDCl_3$) for Side Chains Signals in (24*S*)- and (24*R*)-24-Hydroxymethyl Steroids[a]

compd	21-Me	22-H	23-H	26,27-Me's	28-H_2	
5a	1.03 d (6.5)	5.38 dd (8.8, 15)	5.09 dd (9.3, 15)	0.84, 0.88 d's (6.5)	3.32 t (10)	3.62 m
5b	1.04 d (6.50)	5.39 dd (8.8, 15)	5.09 dd (9.3, 15)	0.85, 0.89 d's (6.5)	3.33 t (10)	3.62 m
5a$_1$	0.89 d (6.50)	5.22 dd (8.8, 15)	5.06 dd (8.8, 15)	0.80, 0.85 d's (6.5)	4.20 dd (7.3, 10.5)	4.29 dd (5.5, 10.5)
5b$_1$	0.97 d (6.50)	5.31 dd (7.5, 15)	5.12 dd (7.5, 15)	0.80, 0.84 d's (6.5)	4.23 dd (4, 10.5)	4.29 dd (4.5, 10.5)
5a$_2$	0.90 d (6.50)	5.25 dd (8.8, 15)	5.08 dd (8.8, 15)	0.80, 0.83 d's (6.5)	4.24 br d (6.2)	
5b$_2$	0.94 d (6.50)	5.25 dd (7.5, 15)	5.06 dd (8.5, 15)	0.78, 0.84 d's (6.5)	4.20 dd (6.7, 10.5)	4.30 dd (5.5, 10.5)
7a	0.92 d (6.5)	–	–	0.88, 0.91 d's (6.5)	3.58 br d (5)	
7b	0.93 d (6.5)	–	–	0.89, 0.90 d's (6.5)	3.53 dd (6.5, 10)	3.61 dd (6.5, 10)

[a] The chemical shift values are given in δ ppm and were referred to $CHCl_3$ (7.27). The coupling constants, in parentheses, are given in hertz.

Table II. 250-MHz ^{1}H NMR Data (CD_3OD) for Side Chains Signals in (24*S*)- and (24*R*)-24-Hydroxymethyl Steroids[a]

compd	21-Me	22-H	23-H	26,27-Me's	28-H_2	
5a	1.07 d (6.5)	5.33 dd (8.8, 15)	5.20 dd (9.3, 15)	0.88, 0.94 d's (6.5)	3.49 dd (6.5, 10)	3.57 dd (6.5, 10)
5b	1.08 d (6.50)	5.35 dd (8.8, 15)	5.20 dd (9.3, 15)	0.87, 0.93 d's (6.5)	3.51 dd	3.58 dd
5a$_1$	0.96 d (6.50)	5.28 dd (8.8, 15)	5.14 dd (8.8, 15)	0.87, 0.94 d's (6.5)	4.25 dd (7.5, 10.5)	4.39 dd (5, 10)
5b$_1$	1.03 d (6.50)	5.38 dd (7.5, 15)	5.20 dd (7.5, 15)	0.87, 0.91 d's (6.5)	4.34 d (6.5)	
5a$_2$	0.96 d (6.50)	5.33 dd (8.8, 15)	5.18 dd (8.8, 15)	0.88, 0.92 d's (6.5)	4.32 br d (6.2)	
5b$_2$	1.01 d (6.50)	5.26 dd (7.5, 15)	5.14 dd (8.5, 15)	0.87, 0.93 d's (6.5)	4.23 br dd (7, 10.5)	4.43 dd (5.5, 10.5)
7a	0.98 d (6.5)	–	–	0.91, 0.94 d's (6.5)	3.52 br d (5)	
7b	0.99 d (6.5)	–	–	0.93, 0.92 d's (6.5)	3.47 dd (6.5, 10)	3.56 dd (6.5, 10)

[a] The chemical shift values are given in δ ppm and were referred to CHD_2OD (3.34). The coupling constants, in parentheses, are given in hertz.

^{1}H and/or ^{13}C NMR spectroscopy which would permit unambiguous differentiation between 24*R* and 24*S* epimers or alternatively if this could be achieved after derivatization with a chiral reagent [e.g. (*R*)-(+)- or (*S*)-(−)-α-methoxy-α-(trifluoromethyl)phenylacetic acid].

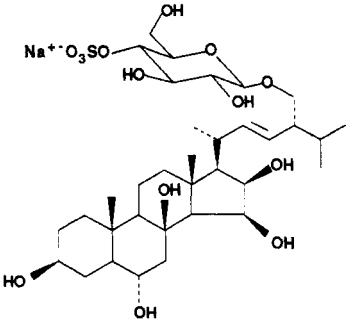
In designing the synthesis of **6a** and **6b** (Scheme I) we have used the methods developed by Sucrow and coworkers,[5] which permits construction of the chiral center at C-24 in a predictable way from a cis allylic C-22 alcohol via Claisen rearrangement. This approach has been used by different authors for stereospecific construction of functionalized steroidal side chains.[6]

The 250-MHz ^{1}H NMR spectra of the two epimeric alcohols **5a** and **5b** as well those of the two epimeric diols **6a** and **6b** were virtually identical, and the ^{13}C NMR spectra were equally useless for differentiating between the C-24 epimers. However, the ^{1}H NMR spectra of the (*R*)-(+)-α-methoxy-α-(trifluoromethyl)phenylacetates (MTPA) **5a$_1$** and **5b$_1$** showed differences in the side chain signals (Tables I and II) which permitted differentiation between the original epimers. The most noticeable feature was a large difference in the chemical shift of the C-21 methyl signal, which in the 24*S* isomer **5a$_1$** was shifted upfield by more than 0.1 ppm if compared with the underivatized compound, whereas in the 24*R* isomer **5b$_1$** it was affected only to a minor extent; moreover the signals due to the C-28 protons were separated more in the spectrum of the 24*S* isomer **5a$_1$** than in that of the 24*R* isomer **5b$_1$**. Likewise signals of the C-26 and C-27 protons were more separated in **5a$_1$** then in **5b$_1$**. The relatively large upfield shift exhibited by the C-21 methyl signal in the 24*S* isomer **5a$_1$** can be explained in terms of a preferred anti conformation with the 21-methyl group in the shielding cone of the phenyl ring of the MTPA residue. The Me-21 signals of the (*S*)-(−)-α-methoxy-α-(trifluoromethyl)phenylacetates (MTPA) **5a$_2$** and **5b$_2$** exhibited the same behavior. In the 24*S* isomer **5a$_2$** the C-21 proton signal is again shifted upfield, whereas the differences observed for the C-26, C-27, and C-28 proton signals are, as expected, completely reversed and are even larger than those observed in the (*R*)-(+)-MTPA derivatives **5a$_1$** and **5b$_1$**. Thus the signals due to the C-28 protons are more separated in the spectrum of the 24*R* isomer **5b$_2$** than in that of the 24*S* isomer **5a$_2$** and the same is true for the C-26 and C-27 proton signals.

Thus ^{1}H NMR measurements of the MTPA derivatives provide a highly reliable means for making stereochemical assignments in Δ^{22}, 24-hydroxymethyl steroids.

In the series with a saturated side chain, **7a** and **7b**, the two C-24 epimers can be differentiated directly by their ^{1}H NMR spectra. Thus the C-28 protons resonate as a broad doublet in the ^{1}H NMR spectrum of the 24*R* isomer **7a**, while they appear as two well-separated signals in the spectrum of the 24*S* isomer **7b**. Small differences are also observed in the chemical shifts of the side chain methyl protons (Tables I and II). Examination of the ^{13}C NMR data shows small differencies in the chemical shifts of side chain signals between the two isomers which also offer useful information about the C-24 stereochemistry. What appears to be of greatest diagnostic value is the difference in the chemical shifts of the isopropyl methyl carbons: this is larger in the spectrum of the 24*R* isomer **7a** than in that of the 24*S* isomer **7b**.

With these data in hand it was possible to assign the stereochemistry at C-24 of coscinasteroside C (**8**).



8 coscinasteroside C

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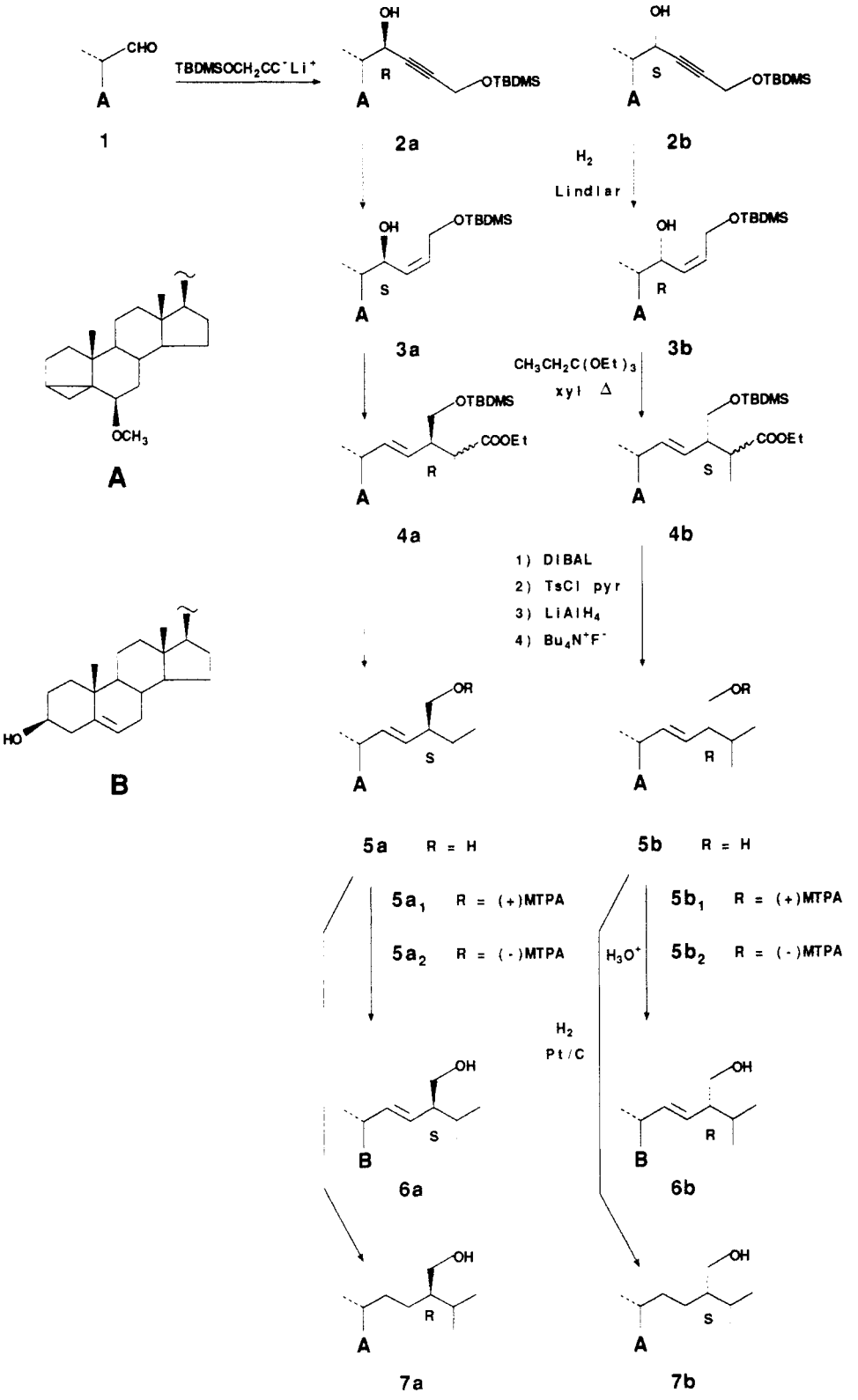
Scheme I



The 24-methyl-5α-cholest-22(E)-ene-3β,6α,8,15β,16β-,28-hexol derived from coscinasteroside C after removing the glucose unit by enzymatic hydrolysis was converted to a 3β,6α,28-(R)-(+)-MTPA derivative. In the ^{1}H NMR spectrum (CD_3OD) of the latter the resonances of the C-28 protons (d at δ 4.37, J = 6.5 Hz) and of the 21-Me (d at δ 1.06, J = 7 Hz), this last essentially at the same chemical shift as in the underivatized steroid, were in good agreement with the shifts of **5b**$_1$ in CD_3OD, but significantly different from those of **5a**$_1$, thus establishing the stereochemistry at C-24 in coscinasteroside C to be 24R.

A similar procedure was also used to establish the stereochemistry at C-24 in pisasteroside A from *P. brevispinus*[3] and in a 24-hydroxymethyl steroid isolated from the ophiuroid *O. superba*.[4]

Experimental Section

The following instruments were used. For NMR, Bruker

WM-250 Fourier transform spectrometer; for mass spectra, Kratos MS50; for optical rotations, Perkin Elmer polarimeter Model 141. Melting point were determined on a Kofler apparatus and are uncorrected.

"The usual workup" refers to dilution with water, extraction with diethyl ether, washign to neutrality, drying over $MgSO_4$, filtration, and evaporation under vacuum.

(22*R*)- and (22*S*)-25-((*tert*-Butyldimethylsilyl)oxy)-26,27-dinor-3α,5-cyclo-6β-methoxy-5α-cholest-23-yn-22-ol (2a,b). 3-((*tert*-Butyldimethylsilyl)oxy)propyne (6 g) was added under N_2 to a solution of *n*-butyllithium (35 mmol) in 60 mL of dry THF at −15 °C, and the mixture was stirred for 30 min at −15 °C and then at room temperature for 1 h. Benzene (25 mL) was added, and the solution was kept at 0 °C during the addition of the aldehyde 1[7] (4.5 g) in THF (25 mL). The mixture was stirred at room temperature for 1 h, and then a saturated ammonium chloride solution was added. The usual workup gave 8.2 g of crude product, which was chromatographed on silica gel. Gradual elution with petroleum ether–ethyl acetate from 98:2 to 95:5 gave in order of polarity the 22*R* isomer **2a** (2 g) and the 22*S* isomer **2b** (1.3 g).[8]

Less polar product **2a** (22*R* isomer): MS *m/z* 514 (M^+); ^1H NMR ($CDCl_3$) δ 0.11 (6 H, s, $SiMe_2$), 0.42 (1 H, m, 4-H), 0.63 (1 H, m, 4-H), 0.71 (3 H, s, 18-Me), 0.90 (9 H, s, $SiCMe_3$), 1.01 (3 H, s, 19-Me), 1.09 (3 H, d, *J* = 7 Hz, 21-Me), 2.76 (1 H, t, *J* = 3 Hz, 6-H), 3.31 (3 H, s, OMe), 4.34 (2 H, br s, 25-H_2), 4.49 (1 H, br, $W_{1/2}$ = 6 Hz, 22-H) ppm.

More polar product **2b** (22*S* isomer): MS *m/z* 514 (M^+); ^{1}NMR ($CDCl_3$) δ 0.13 (6 H, s, $SiMe_2$), 0.43 (1 H, m, 4-H), 0.64 (1 H, m, 4-H), 0.73 (3 H, s, 18-Me), 0.91 (9 H, s, $SiCMe_3$), 1.02 (3 H, s, 19-Me), 1.04 (3 H, d, *J* = 7 Hz, 21-Me), 2.74 (1 H, t, *J* = 3 Hz, 6-H), 3.32 (3 H, s, OMe), 4.35 (2 H, br s, 25-H_2), 4.47 (1 H, br, $W_{1/2}$ = 9 Hz, 22-H) ppm.

(22*S*,23*Z*)- and (22*R*,23*Z*)-25-((*tert*-Butyldimethylsilyl)oxy)-26,27-dinor-3α,5-cyclo-6β-methoxy-5α-cholest-23-en-22-ol (3a,b). A mixture of the (22*R*)-acetylenic alcohol **2a** (1.5 g) and Lindlar catalyst (Aldrich Chemical Co., 150 mg) in toluene (40 mL) was stirred at room temperature under a hydrogen atmosphere for 3 h. The suspension was filtered, and the solvent was evaporated under reduced pressure to give **3a**: MS *m/z* 516 (M^+, 5), 501 (15), 461 (40), 369 (40), 201 (100); ^1H NMR ($CDCl_3$) δ 0.08 (6 H, s, $SiMe_2$), 0.43 (1 H, m, 4-H), 0.65 (1 H, m, 4-H), 0.72 (3 H, s, 18-Me), 0.90 (9 H, s, $SiCMe_3$), 0.96 (3 H, d, *J* = 6.5 Hz, 21-Me), 1.02 (3 H, s, 19-Me), 2.77 (1 H, t, *J* = 3 Hz, 6-H), 3.32 (3 H, s, OMe), 4.22–4.29 (2 H, m, 25-H_2), 4.49 (1 H, br, $W_{1/2}$ = 8.8 Hz, 22-H), 5.59 (2 H, m, 23,24-H) ppm.

The (22*S*)-acetylenic alcohol **2b** (1 g) was hydrogenated under the same conditions as for **2a** to afford the 22*R*,23*Z* allylic alcohol **3b**: MS *m/z* 516 (M^+, 5), 501 (20), 461 (50), 369 (50), 201 (20); ^1H NMR ($CDCl_3$) δ 0.10 (6 H, s, $SiMe_2$), 0.43 (1 H, m, 4-H), 0.66 (1 H, m, 4-H), 0.76 (3 H, s, 18-Me), 0.92 (9 H, s, $SiCMe_3$), 1.01 (3 H, d, *J* = 6.5 Hz, 21-Me), 1.03 (3 H, s, 19-Me), 2.77 (1 H, t, *J* = 3 Hz, 6-H), 3.32 (3 H, s, OMe), 4.24 (1 H, dd, *J* = 5, 12.5 Hz), 4.34 (1 H, dd, *J* = 6, 12.5 Hz) (25-H_2), 4.46 (1 H, br dd, *J* = 3.7, 8 Hz, 22-H), 5.61 (1 H, br dd, *J* = 8.7, 11 Hz, 23-H), 5.74 (1 H, m, 24-H) ppm.

Ethyl (22*E*,24*R*,25*R*)- and (22*E*,24*R*,25*S*)-24-(((*tert*-Butyldimethylsilyl)oxy)methyl)-3α,5-cyclo-6β-methoxy-5α-cholest-22-en-26-oate (4a). The (22*S*,23*Z*)-allylic alcohol **3a** (700 mg) was heated under reflux in xylene (30 mL) with triethyl orthopropionate (4 mL) and propionic acid (0.25 mL), 5 mL of solvent was removed by distillation after 1.5 h, and reflux was continued for additional 1.5 h. The usual workup gave the epimeric mixture **4a** (650 mg): MS *m/z* 543 ([M − *tert*-butyl]$^+$); ^1H NMR ($CDCl_3$) δ 0.03 (6 H, s, $SiMe_2$), 0.43 (1 H, m, 4-H), 0.65 (1 H, m, 4-H), 0.72 (3 H, s, 18-Me), 0.90 (9 H, s, $SiCMe_3$), 1.02 (3 H, s, 19-Me), 2.77 (1 H, t, *J* = 3 Hz, 6-H), 3.32 (3 H, s, OMe), 3.50–3.60 (m's, 28-H_2), 4.11 (q, *J* = 6 Hz, OEt), 5.10 (dd, *J* = 7, 13 Hz, 23-H), 5.29 (dd, *J* = 7, 13 Hz, 22-H) ppm, overlapping with a multiplet for 22- and 23-H of the other C-25 epimer.

Ethyl (22*E*,24*S*,25*R*)- and (22*E*,24*S*,25*S*)-24-(((*tert*-Butyldimethylsilyl)oxy)methyl)-3α,5-cyclo-6β-methoxy-5α-cholest-22-en-26-oate (4b). The (22*R*,23*Z*)-allylic alcohol **3b** (300 mg) was heated under reflux in xylene (12 mL) with triethyl orthopropionate (2 mL) and propionic acid (0.25 mL), 2 mL of solvent was removed by distillation after 1.5 h, and reflux was continued for additional 1.5 h. The usual workup gave the epimeric mixture **4b** (250 mg): MS *m/z* 543 ([M − *tert*-butyl]$^+$); ^1H NMR ($CDCl_3$) δ 0.03 (6 H, s, $SiMe_2$), 0.43 (1 H, m, 4-H), 0.65 (1 H, m, 4-H), 0.72 (3 H, s, 18-Me), 0.90 (9 H, s, $SiCMe_3$), 1.02 (3 H, s, 19-Me), 2.77 (1 H, t, *J* = 3 Hz, 6-H), 3.32 (3 H, s, OMe), 3.50–3.60 (m's, 28-H_2), 4.11 (q's, *J* = 6 Hz, OEt), 5.10–5.29 (m's, 22- and 23-H) ppm.

(22*E*,24*S*)-3α,5-Cyclo-6β-methoxy-5α-ergost-22-en-28-ol (5a). A 1 M solution of diisobutylaluminum hydride in toluene (8 mL) was slowly added to a solution of ester mixture **4a** (800 mg) stirred at −10 °C under a N_2 atmosphere. After 2 h the solution was allowed to warm to room temperature and, after addition of a further 4 mL of DIBAL, was stirred at room temperature for 3 more hours before being quenched with methanol. The solution was then poured into a saturated ammonium chloride solution, and usual workup afforded the crude 26-ol mixture (710 mg). This material was dissolved in dry pyridine (2 mL) and treated overnight at 5 °C with 500 mg of *p*-toluenesulfonyl chloride. The usual workup afforded the crude *p*-toluenesulfonate (850 mg; reaction checked by TLC), which was dissolved in 5 mL of dry ethyl ether and added dropwise to a suspension of lithium aluminum hydride (600 mg) in dry ethyl ether (10 mL). The mixture was stirred at room temperature and the reaction was monitored by TLC. After 5 h, when any trace of starting material had disappeared, the mixture was quenched by adding a few drops of methanol. The crude residue obtained after usual workup (560 mg) was dissolved into 6 mL of dry THF, added with 1.5 mL of a 1 M solution of tetrabutylammonium fluoride, and kept at room temperature for 8 h.[9] The solution was concentrated under vacuum and then diluted with ethyl ether, washed with water, and evaporated to dryness. Chromatography of the residue on a silica gel (12 g) column using as eluant mixtures of petroleum ether–ethyl acetate of increasing polarity (from 95:5 to 85:15) afforded pure **5a** (325 mg): $[\alpha]_D$ +19° (*c* 1, $CHCl_3$); HRMS calcd for $C_{29}H_{48}O_2$ (M^+) *m/z* 428.3654, found 428.3642; ^1H NMR ($CDCl_3$) δ 0.43 (1 H, m, 4-H), 0.65 (1 H, m, 4-H), 0.73 (3 H, s, 18-Me), 1.01 (3 H, s, 19-Me), 2.76 (1 H, t, *J* = 3 Hz, 6-H), 3.31 (1 H, s, OMe), remaining side chain signals in Tables I and II; ^{13}C NMR ($CDCl_3$) C-1 33.4, C-2 25.0, C-3 21.5, C-4 13.1, C-5 35.3, C-6 82.4, C-7 35.1, C-8 30.5, C-9 48.1, C-10 43.4, C-11 22.7, C-12 40.2, C-13 42.9, C-14 56.6, C-15 24.2, OMe 56.5 ppm; (CD_3OD) C-1 34.5, C-2 25.9, C-3 22.8, C-4 13.9, C-5 36.5, C-6 84.0, C-7 36.2, C-8 31.8, C-9 49.5, C-10 44.6, C-11 23.8, C-12 41.5, C-13 43.9, C-14 57.5, C-15 25.3, OMe 57.9 ppm, remaining signals in Table III.

(22*E*,24*R*)-3α,5-Cyclo-6β-methoxy-5α-ergost-22-en-28-ol (5b). An analogous reaction sequence was used to convert ester mixture **4b** (500 mg) to **5b** (140 mg): $[\alpha]_D$ +22.5° (*c* 1, $CHCl_3$); HRMS calcd for $C_{29}H_{48}O_2$ (M^+) *m/z* 428.3654, found 428.3635; ^1H NMR ($CDCl_3$) δ 0.43 (1 H, m, 4-H), 0.65 (1 H, m, 4-H), 0.73 (3 H, s, 18-Me), 1.02 (3 H, s, 19-Me), 2.76 (1 H, t, *J* = 3 Hz, 6-H), 3.32 (1 H, s, OMe), remaining side chain signals in Tables I and II; ^{13}C NMR C-1 to C-15 as in **5a** ±0.1 ppm, remaining signals in Table III.

Preparation of (*R*)-(+)- and (*S*)-(−)-MTPA Derivatives $5a_1$–$5b_2$ for NMR Measurements. The required alcohol **5a** or **5b** (5 mg) was treated with (+)- or (−)-α-methoxy-α-(trifluoromethyl)phenylacetyl (MTPA) chloride (3–5 μL) in dry pyridine (0.1 mL) for 1–3 h at room temperature. After solvent removal, the product was eluted with CH_2Cl_2 through a Pasteur pipet filled (ca. 2 cm) with Si gel. Significative ^1H NMR spectral data are reported in Tables I and II.

(22*E*,24*S*)-Ergosta-5,22-diene-3β,28-diol (6a). The *i*-sterol **5a** (15 mg) was refluxed for 3 h in 4 mL of a 1:1 mixture water–dioxane acidified with a few milligrams of *p*-toluenesulfonic acid. After cooling and usual workup the product was purified

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Table III. ^{13}C NMR Spectral Data[a]

	5a		5b		7a		7b	
C	$CDCl_3$	CD_3OD	$CDCl_3$	CD_3OD	$CDCl_3$	CD_3OD	$CDCl_3$	CD_3OD
16	28.9	30.0	28.9	29.6	28.4	29.3	28.3	29.3
17	55.9	56.8	56.0	56.8	56.4	56.8	56.4	56.8
18	12.4	12.9	12.5	12.9	12.3	12.6	12.3	12.6
19	19.2	19.8	19.3	19.7	19.3	19.7	19.3	19.7
20	40.5	41.7	40.4	41.5	36.2	37.3	36.3	37.5
21	20.9	21.5	20.8	21.4	18.7	19.3	18.9	19.7
22	141.9	141.0	141.6	140.8	34.2	35.3	34.3	35.5
23	126.5	127.9	126.8	127.7	24.3	25.1	24.3	25.5
24	52.5	53.0	52.6	52.9	47.2	50.0	47.5	50.0
25	29.0	29.5	29.1	29.3	28.4	29.1	28.2	29.0
26	19.7	19.1	19.6	18.9	19.3	19.2	19.3	19.3
27	21.1	21.3	20.9	21.4	20.0	20.4	19.6	19.9
28	64.1	65.2	64.3	65.2	63.1	63.8	64.2	64.1
OCH_3	56.5	57.9	56.5	57.8	56.7	57.8	56.7	57.8

[a] At 62.9 MHz; values relative to $CDCl_3$ = 77.00 ppm and CD_3OD = 49.00 ppm (central peaks); assignment aided by DEPT technique and comparison with known reference compounds; carbon signals 1–15 in the Experimental Section.

by HPLC on a Whatman ODS-2 M9 10/50 column eluting with methanol, to afford **6a** (10 mg): $[\alpha]_D$ −51° (*c* 1, $CHCl_3$); mp 194–196 °C; HRMS calcd for $C_{28}H_{46}O_2$ (M^+) *m/z* 414.3498, found 414.3490; ^1H NMR ($CDCl_3$) δ 0.70 (3 H, s, 18-Me), 0.85–0.89 (each 3 H, d, *J* = 7 Hz, 26- and 27-Me), 1.01 (3 H, s, 19-Me), 1.04 (3 H, d, *J* = 7 Hz, 21-Me), 3.35 (1 H, dd, *J* = 9, 11 Hz, 28-H), 3.50 (1 H, m, 3-H), 3.62 (1 H, dd, *J* = 5, 11 Hz, 28-H), 5.10 (1 H, dd, *J* = 9, 15 Hz, 23-H), 5.35 (1 H, m, 6-H), 5.39 (1 H, dd, *J* = 8.5, 15 Hz, 22-H).

(22*E*,24*R*)-Ergosta-5,22-diene-3β,28-diol (6b). The *i*-sterol **5b** (10 mg) was treated as described above to afford **6b** (6 mg): $[\alpha]_D$ −36° (*c* 0.6, $CHCl_3$); mp 196–198 °C; HRMS calcd for $C_{28}H_{46}O_2$ (M^+) *m/z* 414.3498, found 414.3502; ^1H NMR ($CDCl_3$) δ 0.71 (3 H, s, 18-Me), 0.85–0.89 (each 3 H, d, *J* = 7 Hz, 26- and 27-Me), 1.01 (3 H, s, 19-Me), 1.04 (3 H, d, *J* = 7 Hz, 21-Me), 3.34 (1 H, dd, *J* = 9, 11 Hz, 28-H), 3.50 (1 H, m, 3-H), 3.62 (1 H, dd, *J* = 5, 11 Hz, 28-H), 5.10 (1 H, dd, *J* = 9, 15 Hz, 23-H), 5.35 (1 H, m, 6-H), 5.39 (1 H, dd, *J* = 8.5, 15 Hz, 22-H).

(24*R*)-3α,5-Cyclo-6β-methoxy-5α-ergostan-28-ol (7a). Alcohol **5a** (15 mg) was hydrogenated at atmospheric pressure over 10% Pt/C in 10 mL of ethanol for 5 h. Removal of the catalyst by filtration and evaporation of solvent gave the noncrystalline saturated alcohol **7a** (13 mg): $[\alpha]_D$ +45° (*c* 1, $CHCl_3$); HRMS calcd for $C_{29}H_{50}O_2$ (M^+) *m/z* 430.3798, found 430.3794; ^1H NMR ($CDCl_3$) δ 0.42 (1 H, m, 4-H), 0.64 (1 H, m, 4-H), 0.71 (3 H, s, 18-Me), 1.02 (3 H, s, 19-Me), 2.77 (1 H, t, *J* = 3 Hz, 6-H), 3.32 (3 H, s, OMe), remaining side chain signals in Tables I and II; ^{13}C NMR C-1 to C-15 as in **5a** ±0.1 ppm, remaining signals in Table III.

(24*S*)-3α,5-Cyclo-6β-methoxy-5α-ergostan-28-ol (7b). Alcohol **5a** (15 mg) was converted to **7b** as described above: $[\alpha]_D$ +39° (*c* 1, $CHCl_3$); HRMS calcd for $C_{29}H_{50}O_2$ (M^+) *m/z* 430.3798, found 430.3789; ^1H NMR ($CDCl_3$) δ 0.42 (1 H, m, 4-H), 0.64 (1 H, m, 4-H), 0.71 (3 H, s, 18-Me), 1.02 (3 H, s, 19-Me), 2.77 (1 H, t, *J* = 3 Hz, 6-H), 3.32 (3 H, s, OMe), remaining side chain signals in Tables I and II; ^{13}C NMR C-1 to C-15 as in **5a** ±0.1 ppm, remaining signals in Table III.

Enzymic Hydrolysis of Coscinasteroside C (8) and Preparation of MTPA Derivative for NMR Measurement. The glycoside sulfate **8** (5 mg), after solvolysis at 130 °C in pyridine–dioxane, 1:1, was incubated at 37 °C with a glycosidase mixture (5 mg) from *Charonia lampas* in citrate buffer (2.0 mL; pH 4.5). After reaction for 24 h, TLC analysis (SiO_2 with 1-butanol–acetic acid–water, 60:15:25) showed that the starting material had disappeared. The mixture was then extracted with 1-butanol and evaporated, and the residue was fractionated by hplc on a C-18 μ-Bondapack column (30 cm × 3.9 mm i.d.) using methanol–water (70:30) as eluent to give 24-methyl-5α-cholest-22(*E*)-ene-3β,6α,8,15β,16β,28-hexol (1 mg): negative ion FABMS *m/z* 479 ($[M - H]^-$); ^1H NMR (CD_3OD) δ 0.88 (3 H, d, *J* = 6.5 Hz, 26- or 27-H_3), 0.94 (3 H, d, *J* = 6.5 Hz, 27- or 26-H_3), 1.03 (3 H, s, 19-H_3), 1.09 (3 H, d, *J* = 6.5 Hz, 21-H_3), 1.32 (3 H, s, 18-H_3), 1.64 (1 H, m, 25-H), 2.18 (1 H, m, 24-H), 2.43 (1 H, dd, *J* = 5, 12 Hz, 7-H), 3.46 (1 H, dd, *J* = 9, 10 Hz, 28-H), 3.50 (1 H, m, 3α-H), 3.67 (1 H, dd, *J* = 5, 10 Hz, 28-H), 3.73 (1 H, td, *J* = 3, 12 Hz, 6β-H), 4.16 (1 H, t, *J* = 6.5 Hz, 16α-H), 4.39 (1 H, dd, *J* = 5.6, 6.7 Hz, 15α-H), 5.29 (1 H, dd, *J* = 9, 15 Hz, 23-H), 5.49 (1 H, dd, *J* = 9, 15 Hz, 22-H).

The polyhydroxylated sterol was then treated with (+)-α-methoxy-α-(trifluoromethyl)phenylacetyl (MTPA) chloride (3 μL) as described above for preparation of **5a**$_1$–**5b**$_2$ to give (+)-MPTA triester; negative ion FABMS *m/z* 1127 ($[M - H]^-$); ^1H NMR (CD_3OD) δ 0.88 (3 H, d, *J* = 6.5 Hz, 26- or 27-H_3), 0.92 (3 H, d, *J* = 6.5 Hz, 27- or 26-H_3), *1.06 (3 H, d, J = 6.5 Hz, 21-H3)*, 1.12 (3 H, s, 19-H_3), 1.31 (3 H, s, 18-H_3), 2.64 (1 H, m, 20-H), *4.37 (2 H, d, J = 6.5 Hz, 28-H$_2$)*, 5.37 (1 H, dd, *J* = 9, 15 Hz, 23-H), 5.79 (1 H, dd, *J* = 9, 15 Hz, 22-H).

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Registry No. **1**, 25819-77-6; **2a**, 91535-67-0; **2b**, 91509-33-0; **3a**, 91509-34-1; **3b**, 91509-36-3; **4a** (isomer 1), 125413-89-0; **4a** 26-ol derivative (isomer 1), 125413-90-3; **4a** *p*-toluenesulfonate derivative (isomer 1), 125413-91-4; **4a** (isomer 2), 125413-94-7; **4a** 26-ol derivative (isomer 2), 125413-92-5; **4a** *p*-toluenesulfonate derivative (isomer 2), 125413-93-6; **4b**, 125473-23-6; **4b** (isomer 2), 125473-24-7; **5a**, 125413-95-8; **5a1**, 125413-95-8; **5a2**, 125473-26-9; **5b**, 125473-25-8; **5b1**, 125514-81-0; **5b2**, 125473-27-0; **6a**, 125413-97-0; **6b**, 125473-28-1; **7a**, 68844-34-8; **7b**, 68889-65-6; **8**, 105377-96-6; (+)-MTPA chloride, 20445-33-4; (−)-MTPA chloride, 39637-99-5; 3β,6α,28-(*R*)-(+)-MTPA, 125413-98-1; 3-((*tert*-butyldimethylsilyl)oxy)propyne, 76782-82-6; triethyl orthopropionate, 115-80-0; (22*E*,24*S*)-24-(((*tert*-butyldimethylsilyl)oxy)methyl)-3α,5-cyclo-6β-methoxy-5α-cholest-22-ene, 125413-99-2; 24-methyl-2α-cholest-22(*E*)-ene-3β,6α,8,15β,16β,28-hexol, 125414-00-8; pisasteroside A, 123154-33-6.

An Improved Method for Reductive Alkylation of Amines Using Titanium(IV) Isopropoxide and Sodium Cyanoborohydride[1]

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The reductive alkylation of amines is one of the fundamental reactions of synthetic organic chemistry. The

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