Stereoselective Synthesis of the C-24 and C-25 Stereoisomeric Pairs of 24-Ethyl-26-hydroxy- and 24-Ethyl-[26-^{2}H]sterols and their Δ^{22}-Derivatives: Reassignment of ^{13}C N.m.r. Signals of the *pro-R* and the *pro-S* Methyl Groups at C-25 of 24-Ethylsterols

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Two C-24 and C-25 epimeric pairs of 24-ethyl-26-acid sterol derivatives (**22**)—(**25**) were stereoselectively synthesized *via* an ester-enolate Claisen rearrangement of the (22*R*)-(**20**) and (**21**) and (22*S*)-23*E*-ene derivatives (**18**) and (**19**). The absolute configuration at C-24 and C-25 of methyl (22*E*,24*S*,25*S*)-6β-methoxy-3α,5-cyclo-5α-stigmast-22-en-26-oate (**22a**) and (22*E*,24*R*,25*R*)-6β-methoxy-3α,5-cyclo-5α-stigmast-22-en-26-oic acid (**24**) were confirmed by *X*-ray crystallography. Four C-24 and C-25 epimeric pairs of 24-ethyl-[26-^{2}H]-(**34**)—(**37**) and 24-ethyl-26-hydroxy-sterols (**42**)—(**45**) and their Δ^{22}-derivatives (**38**)—(**41**) and (**46**)—(**49**) have been synthesized in order to study the stereochemistry in the biosynthesis of (24*R*)- and (24*S*)-24-ethylsterol side-chains. The data showed that C-26 and C-27 arise biosynthetically from C-6 and C-2 of MVA, respectively.

Plant sterols have a methyl or an ethyl group at C-24 with *R* or *S* configuration. The alkyl groups are known to be introduced by transmethylation from *S*-adenosylmethionine onto the 24(25)-double bond of the side-chain (**1**). As shown in Scheme 1, the proposed biosynthetic mechanism[1] for 24α- (**7**) and (**8**) and 24β-ethylsterol side-chains (**9**)—(**11**) involves 1,2-hydride shifts and double bond reductions. In the course of our studies on the biosynthesis of sterols by ^{13}C n.m.r. spectroscopy, we found that the process operates stereoselectively and that one methyl group at C-25 originates from C-2 of mevalonic acid (MVA) and another from C-6 of MVA.[2] We reported that both 24α-and 24β-ethylsterols (**7**)—(**11**) had the *pro-R* methyl group (C-26) and the *pro-S* methyl group (C-27) originating from C-2 and C-6 of MVA, respectively.[3,4] This conclusion was based on isofucosterol (**5**) having the 24-ethylidene side-chain (**5**) reported by Nicotra *et al.*[5] Later, we found that the two methyl groups at C-25 of 24-methylenecycloartanol and 24-methylenecholesterol having the side-chain (**3**) had carbons of the opposite origin from MVA.[6] The discrepancy of the biosynthetical origin of the methyl groups between the two sterols might suggest a stereochemical inversion mechanism from (**3**) to (**5**). In order to study more precisely the biosynthetic stereoselectivity, we synthesized the sterols (**34**)—(**41**) deuteriated stereospecifically at one of the two methyl groups at C-25 to establish the ^{13}C n.m.r. signal assignments of the methyl groups and also synthesized the (25*R*)- and (25*S*)-24-ethyl-26-hydroxysterols (**42**)—(**49**).

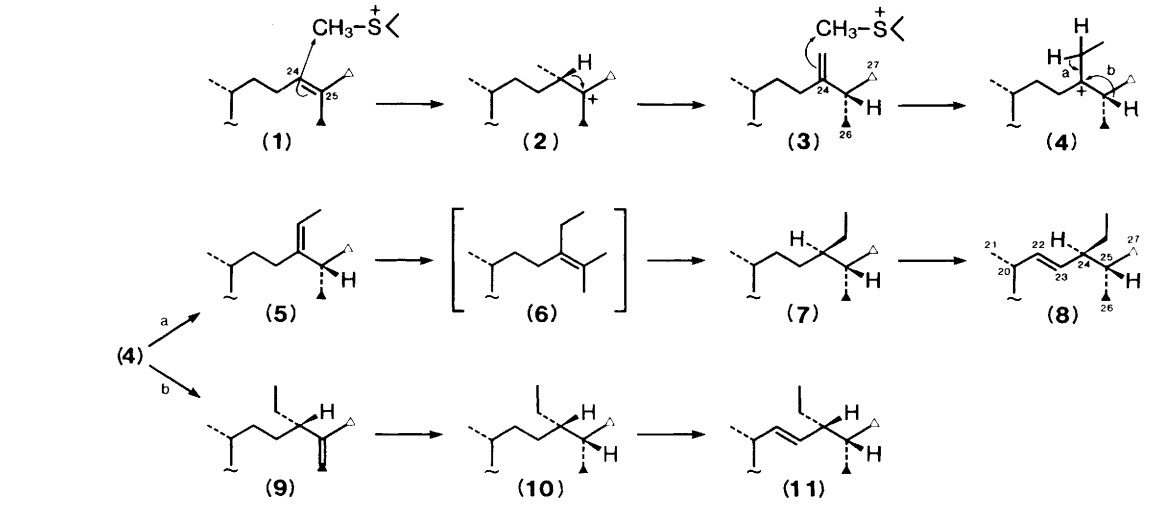
Sucrow *et al.*[7] have successfully applied the Claisen rearrangement to appropriate allylic alcohols for construction of the side-chain of 24-ethylsterols. However, when this rearrangement with the amide acetals[8] or orthoesters,[9,10] of Δ^{23}-22-hydroxy steroids was applied to the synthesis of other sterols, it failed to control the stereochemistry at the C-25 position owing to the nonselective formation of the intermediate ketene acetals. In order to achieve better stereochemical control, the Ireland's enolate-Claisen rearrangement[11] seemed more promising from the few examples of its application, *i.e.*, in the synthesis of ergosterol[12] and the vitamin E side-chain.[13] Our synthetic strategy is the induction of the desired stereochemistry at the C-24 and C-25 positions by this rearrangement.

Results

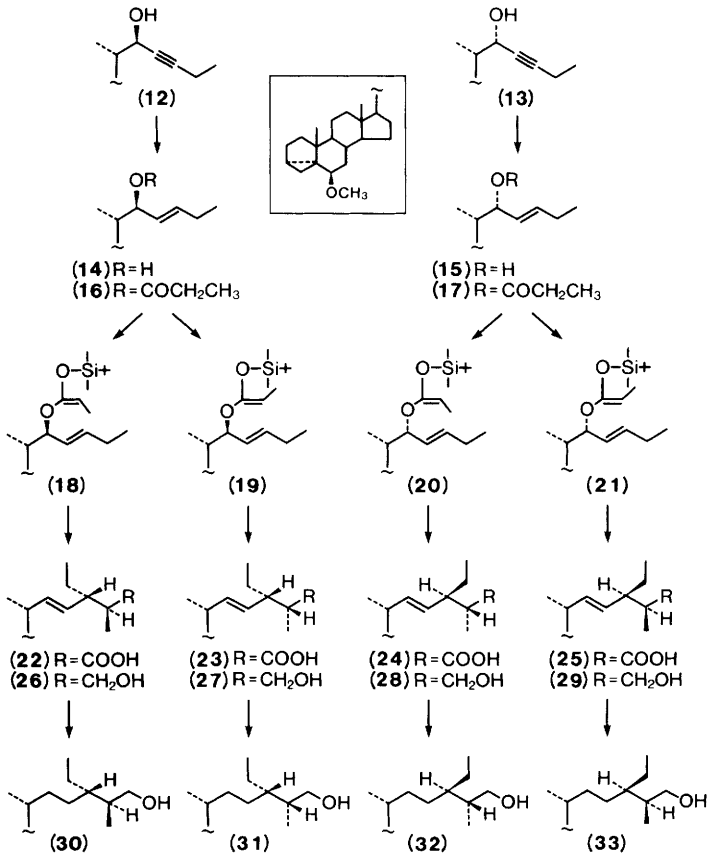
The starting material chosen for our synthesis was the (22*R*)-(**12**) and (22*S*)-acetylenic alcohols (**13**), which are derived from stigmasterol by a literature procedure.[9,10] Compounds (**12**) and (**13**) were reduced independently with an excess of lithium in liquid ammonia to give the *E*-allylic alcohols (**14**) and (**15**), respectively, together with *ca.* 10% of each *Z* isomer[10] as a by-product. These alcohols were purified by chromatography on silica gel. The 23*E* stereochemistry was confirmed by the olefinic region of the ^{1}H n.m.r. spectra. Acylation of (**14**) and (**15**) with propionic anhydride in pyridine gave the esters (**16**) and (**17**), respectively, in quantitative yield.

Claisen rearrangement proceeds through the chair-like transition state which has the smallest number of nonbonded interactions. As shown in Scheme 2, the esters (**16**) and (**17**) should give products (**22**) and (**24**), respectively, when the rearrangement is carried out *via* the *E* enolate,* whereas the product (**23**) and (**25**) should be produced if the rearrangement involves the intermediate *Z* enolates.[11] The selective enolization of the (22*S*)-ester (**16**) was actually carried out in tetrahydrofuran (THF) and in a mixture of hexamethylphosphoramide (HMPA) and THF at −78 °C to generate the *E*- and *Z*-enolates, respectively, which were trapped as t-butyldimethylsilylketene acetals (**18**) and (**19**). The Claisen rearrangement of these ketene acetals proceeded easily even at room temperature. The rearrangement products were isolated as acids (**22**) and (**23**) after hydrolysis of the silyl esters with tetraethylammonium fluoride in THF. The (22*R*)-ester (**17**) was rearranged similarly *via* the *E*-(**20**) and *Z*-ketene acetal

* *E* and *Z* Ester enolate follow Evans, regardless of the nature of the metal which associates with oxygen. See: D. A. Evans, 'Asymmetric Synthesis', ed. J. D. Morrison, Academic Press: New York, 1984, vol. 3.



Scheme 1. Proposed biosynthetic mechanism of (24α)- and (24β)- ethylsterol side-chains. △ and ▲: Carbons originate from C-2 and C-6 of MVA



Scheme 2.

intermediates (**21**) to give the acids (**24**) and (**25**). The stereoselectivity of the rearrangement was determined as the corresponding alcohols (**26**)—(**29**) by high performance liquid chromatography (h.p.l.c.). The results are summarized in Table 1. Selectivity of the *E* enolate in THF (*ca.* 90%) is higher than that of the *Z* enolate in HMPA–THF (*ca.* 80%) in both cases.

The acids (**22**) and (**24**) were obtained in pure form by recrystallization from each reaction mixture of the rearranged *E* enolate (**18**) and (**20**), respectively, in *ca.* 60% yield. Since the diastereoisomeric acids (**23**) and (**25**), which were predicted as major products from the *Z* enolate, could not be crystallized in both cases, each reaction mixture was transformed into the corresponding alcohol (**27**) and (**29**) after esterification followed by lithium aluminium hydride ($LiAlH_4$) reduction. The alcohols (**27**) and (**29**) were purified by preparative h.p.l.c. Other diastereoisomeric alcohols (**26**) and (**28**) were also obtained from (**22**) and (**24**) by esterification followed by $LiAlH_4$ reduction, respectively, in nearly quantitative yield.

In order to confirm the configurations at C-24 and C-25 estimated by the reaction mechanism, the acid (**24**) and the

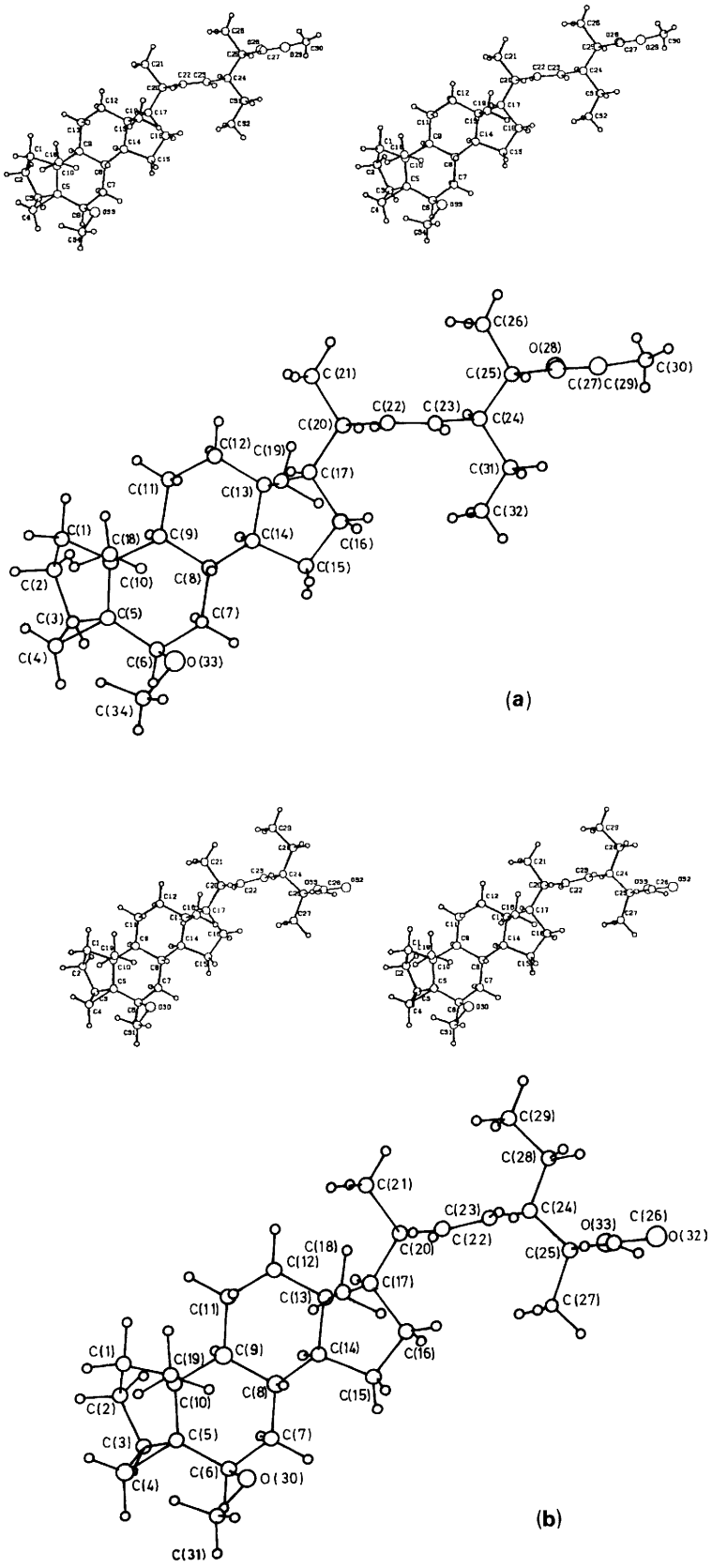


Figure 1. Stereoview of (a) (**22a**) and (b) (**24**)

methyl ester (**22a**), obtained from (**22**), were analysed by *X*-ray crystallography. As shown in Figure 1, compounds (**22a**) and (**24**) were determined as each to have a 22*E* double bond and the (24*S*,25*S*) and (24*R*,25*R*) configurations at the two stereocentres, respectively.

Hydrogenation of the alcohols (**26**)—(**29**) using 10% Pd/C as

Table 1. Enolate Claisen rearrangement of the esters (**16**) and (**17**)

Ester	Enolate	Solvent	Yield (%)	Product ratio[a] (22)	(23)	(24)	(25)
(**16**)	*E*	THF	92	92	8		
(**16**)	*Z*	HMPA–THF	90	22	78		
(**17**)	*E*	THF	88			90	10
(**17**)	*Z*	HMPA–THF	87			20	80

[a] The product ratios were determined by h.p.l.c. as the corresponding alcohols.

a catalyst afforded the 22-dihydro compounds (**30**)—(**33**) in 91—97% yield. After tosylation, reduction by means of $LiAl^2H_4$, and then deprotection with toluene-*p*-sulphonic acid (*p*-TsOH) in aqueous dioxane, the alcohols (**26**)—(**33**) were converted into the 24-ethylsterols stereospecifically deuteriated at one of the methyl groups at C-25, [25*R*,26-^{2}H]-(**38**) and [25*S*,26-^{2}H]-poriferasterol (24*R*) (**39**), [25*S*,26-^{2}H]-(**40**) and [25*R*,26-^{2}H]-stigmasterol (24*S*) (**41**), [25*R*,26-^{2}H]-(**34**) and [25*S*,26-^{2}H]-clionasterol (24*S*) (**35**), and [25*S*,26-^{2}H]-(**36**), and [25*R*,26-^{2}H]-sitosterol (24*R*) (**37**).

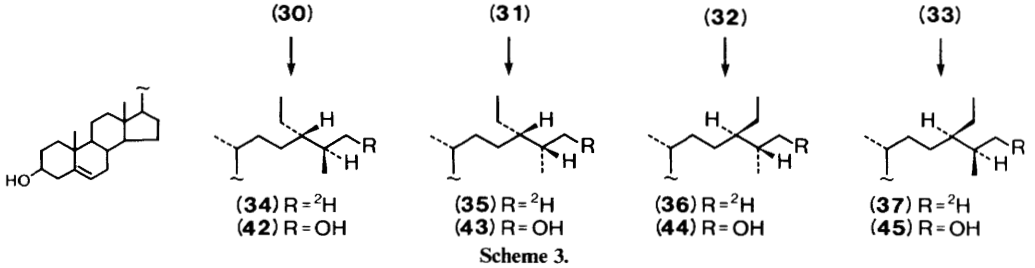
Although the configurations at C-24 and C-25 of the carboxylic acids (**22**) and (**24**) were determined by *X*-ray analysis, those of (**23**) and (**25**) were estimated on the basis of the reaction mechanism. These were confirmed by ^{13}C n.m.r. spectroscopy of the eventually deuteriated sterols (**35**) and (**39**), and (**37**) and (**41**). The ^{13}C n.m.r. data are shown in Table 2 and 3. Since the methyl group bearing a deuterium atom in compounds (**34**), (**36**), (**38**), and (**40**), [derived from the acids (**22**) and (**24**) which were subjected to examination by *X*-ray crystallography], appeared as a triplet ($J_{C,D}$ 19 Hz) with α-deuterium isotope shift (*ca.* −0.3 p.p.m.) in each compound, the signals at δ_C 18.98 of (**34**; R = H) and δ_C 20.91 of (**38**; R = H) were assigned to the *pro-R* methyl group (C-26) at C-25 of each compound, and the signals at δ_C 19.82 of (**36**; R = H) and δ_C 21.09 of (**40**; R = H) were assigned to the *pro-S* methyl group (C-27) at C-25 of each compound. Consequently, the *pro-S* methyl group could be assigned to the signal at δ_C 19.60 of (**34**; R = H) and δ_C 18.94 of (**38**; R = H), and the *pro-R* methyl group to δ_C 19.05 of (**36**; R = H), and δ_C 19.00 of (**40**; R = H). These signals of the corresponding (25*S*)-26-deuteriated isomers (**35**) and (**39**) and those of the (25*R*)-26-deuteriated isomers (**37**) and (**41**) were observed as triplets ($J_{C,D}$ 19 Hz) with an α deuterium isotope shift.

The 26-hydroxylated 24-ethylsterol derivatives obtained from the pollen of *Podocarpus macrophylla*[14] and from biological transformation of sitosterol[15] have been reported, but the configurations at the positions C-24 and C-25 have yet to be determined. We synthesized the set of C-24 and C-25 diastereoisomers of the 24-ethyl-26-hydroxysterols, (25*S*)-(**46**) and (25*R*)-26-hydroxyporiferasterol (**47**), (25*R*)-(**48**) and (25*S*)-26-hydroxystigmasterol (**49**), (25*S*)-(**42**) and (25*R*)-26-hydroxyclionasterol (**43**), and (25*R*)-(**44**) and (25*S*)-26-hydroxysitosterol (**45**), by deprotection of the corresponding 26-hydroxy derivatives (**26**)—(**33**), respectively. ^{13}C N.m.r. assignments of those 26-hydroxysterols are shown in Table 4 and 5 (see Experimental section).

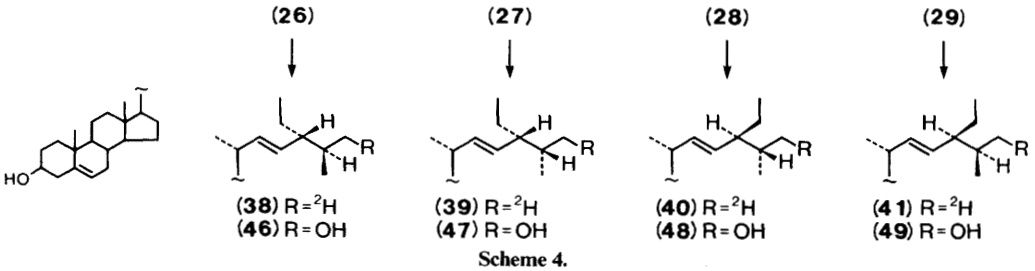
Discussion

The diastereotopic methyl groups, C-26 and C-27, of most sterols have different chemical shifts. The two signals of cholesterol were assigned for the first time by Popják and co-workers[16] with the C-26 resonating at higher field than the C-27 according to the biosynthetic evidence. The results were confirmed by preparing (25*R*)- and (25*S*)-[26-^{2}H]cholesterol.[17] In the case of 24-methylsterols, Arigoni assigned the methyl groups of ergosterol by synthesizing [*pro-S*-methyl ^{13}C]ergosterol.[12] Based on his assignments, we assigned these carbons of campesterol and dihydrobrassicasterol.[6] These assignments agreed with the results[18,19] predicted by the conformational analysis developed by Beierbeck and co-workers.[20]

In a similar way, the methyl carbons of 24-ethylsterols were predicted by Wright and co-workers,[19,21] but their results on sitosterol and poriferasterol need to be revised according to the assignments obtained here. It is noteworthy that the ^{13}C chemical shifts of the C-26 and C-27 are strongly affected by changes in the configuration at C-24, *S* to *R*, when the double bond at C-22 exists (see Figure 2). The C-26 resonating at δ_C 19.00 in (**40**; R = H) shifts by 1.91 p.p.m. downfield and appears at δ_C 20.91 in (**38**; R = H). In contrast, the C-27 shifts by 2.15 p.p.m. upfield. In the case of 22-saturated sterols, sitosterol (**36**; R = H, 24*R*) and clionasterol (**34**; R = H, 24*S*), the effects of the configuration at C-24 on the ^{13}C chemical shifts of the methyl groups are much smaller, −0.08 p.p.m. (C-26) and −0.21 p.p.m. (C-27). These chemical shift comparisons are in good agreement with the values reported on sitosterol,[3]



Scheme 3.



Scheme 4.

Table 2. ^{13}C N.m.r. data of the deuteriated side-chains of sterols (**34**)—(**41**)

Sterols	C-24	C-25	C-26	C-27	C-28	C-29
(**34**; R = H)	46.07	28.96	18.98	19.60	23.03	12.32
(**34**)	46.04	28.87	18.68[d]	19.59	23.01	12.32
	(−0.03)[a]	(−0.09)[b]	(−0.30)[c]	(−0.01)[a]		
(**35**)	46.04	28.87	18.95	19.29[d,e]	23.02	12.32
	(−0.03)[a]	(−0.09)[b]	(−0.03)[a]	(−0.31)[c]		
(**36**; R = H)	45.85	29.18	19.05	19.82	23.08	11.99
(**36**)	45.83	29.09	19.02	19.51[d,e]	23.07	11.98
	(−0.02)[a]	(−0.09)[b]	(−0.03)[a]	(−0.31)[c]		
(**37**)	45.83	29.09	18.78[d]	19.80	23.08	11.98
	(−0.02)[a]	(−0.09)[b]	(−0.27)[c]	(−0.02)[a]		
(**38**; R = H)	51.22	31.83	20.91	18.94	25.38	12.41
(**38**)	51.20	31.75	20.61[d]	18.92	25.38	12.42
	(−0.02)[a]	(−0.08)[b]	(−0.30)[c]	(−0.02)[a]		
(**39**)	51.21	31.76	20.90	18.66[d,e]	25.40	12.43
	(−0.01)[a]	(−0.07)[b]	(−0.01)[a]	(−0.28)[c]		
(**40**; R = H)	51.25	31.91	19.00	21.09	25.41	12.25
(**40**)	51.23	31.81	18.97	20.77[d,e]	25.20	12.24
	(−0.02)[a]	(−0.10)[b]	(−0.03)[a]	(−0.32)[c]		
(**41**)	52.22	31.80	18.68[d]	21.07	25.41	12.25
	(−0.03)[a]	(−0.11)[b]	(−0.32)[c]	(−0.02)[a]		

^{13}C N.m.r. spectra were determined at 50 MHz in [^2H]chloroform and data are shown in p.p.m. [a,b] and [c] γ, β, and α Deuterium isotope shifts, respectively, are shown in parenthesis. [d] These signals were observed as a triplet ($J_{C,D}$ 19 Hz). [e] These signals are C-26, but for easy comparison, we put them in the C-27 column in accordance with non-deuteriated compounds.

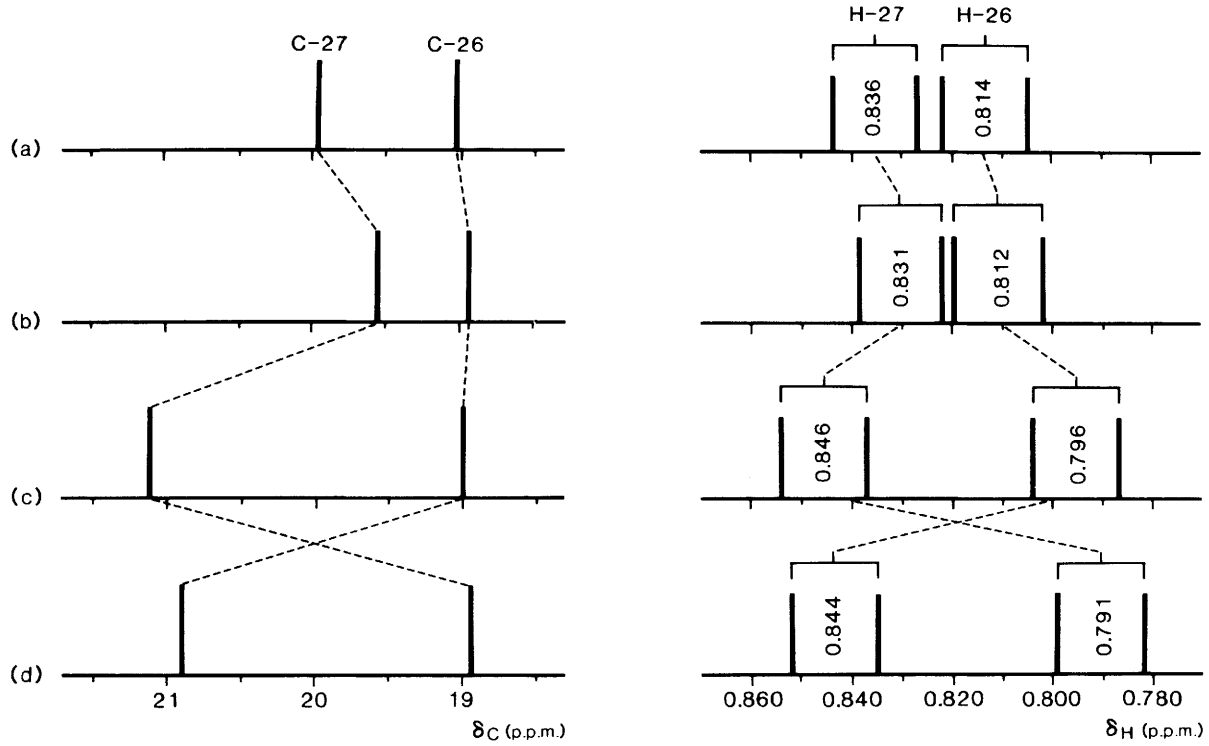
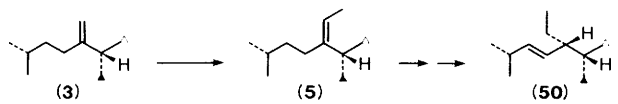


Figure 2. A diagram of the ^{13}C and ^1H n.m.r. signals for C-26 and C-27 in (a) sitosterol (**36**; R = H), (b) clionasterol (**34**; R = H), (c) stigmasterol (**40**; R = H), and (d) poriferasterol (**38**; R = H)



Scheme 5. Proposed biosynthetic mechanism of poriferasterol side-chain (**50**) in *O. malhamensis*

clionasterol,[3,21] stigmasterol,[3] and poriferasterol,[21] but the previous assignments of the two signals need to be reversed.

^1H N.m.r. signals of deuteriated methyl group appeared as a doublet of triplets (J_{HH} 7.0 Hz and J_{HD} 1.8 Hz) with an upfield shift ($\Delta\delta^2_H$ −0.017 to −0.019). As shown in Figure 2, the 26-H signal appeared upfield of the 27-H signals of sitosterol (**36**; R = H), clionasterol (**34**; R = H), and stigmasterol (**40**; R = H); only poriferasterol (**38**; R = H) has its 26-H signal downfield of the 27-H signal. The 29-H chemical shifts of the 24-epimeric pairs are the diagnostic for the 24-configurations [δ_H 0.846 for (**36**) and δ_H 0.855 for (**34**), and δ_H 0.805 for (**40**) and δ_H 0.811 for (**38**)], although the 26-H and 27-H signals are too proximate to distinguish between the corresponding pairs.

Table 3. ^{13}C N.m.r. signal assignments of clionasterol (**34**; R = H), sitosterol (**36**; R = H), poriferasterol (**38**; R = H), and stigmasterol (**40**; R = H)

C-No	(**34**; R = H)	(**36**; R = H)	(**38**; R = H)	(**40**; R = H)
C-1	37.26	37.27	37.27	37.28
C-2	31.68	31.64	31.67	31.66
C-3	71.81	71.77	71.82	71.78
C-4	42.32	42.29	42.31	42.31
C-5	140.76	140.76	140.75	140.76
C-6	121.71	121.69	121.70	121.69
C-7	31.92	31.92	31.91	31.91
C-8	31.92	31.92	31.91	31.91
C-9	50.14	50.15	50.18	50.18
C-10	36.51	36.51	36.52	36.52
C-11	21.09	21.10	21.08	21.09
C-12	39.77	39.80	39.70	39.70
C-13	42.32	42.33	42.23	42.22
C-14	56.77	56.78	56.86	56.88
C-15	24.31	24.31	24.35	24.38
C-16	28.24	28.25	28.79	28.92
C-17	56.04	56.08	55.96	55.97
C-18	11.88	11.86	12.06	12.06
C-19	19.40	19.39	19.39	19.40
C-20	36.27	36.16	40.41	40.50
C-21	18.84	18.80	21.17	21.23
C-22	33.93	33.96	138.24	138.31
C-23	26.39	26.11	129.33	129.28
C-24	46.07	45.85	51.22	51.25
C-25	28.96	29.18	31.83	31.91
C-26	18.98	19.05	20.91	19.00
C-27	19.60	19.82	18.94	21.09
C-28	23.03	23.08	25.38	25.41
C-29	12.32	11.99	12.41	12.25

^{13}C N.m.r. spectra were determined at 50 MHz in [^{2}H]chloroform and the data are shown in p.p.m.

Table 4. ^{13}C N.m.r. signal assignments of 26-hydroxysterols (**42**)—(**45**)

C-No	(**42**)	(**43**)	(**44**)	(**45**)
C-1	37.27	37.27	37.27	37.26
C-2	31.68	31.67	31.67	31.67
C-3	71.81	71.80	71.80	71.80
C-4	42.34	42.31	42.31	42.33
C-5	140.77	140.77	140.77	140.76
C-6	121.70	121.70	121.69	121.69
C-7	31.92	31.91	31.91	31.91
C-8	31.92	31.91	31.91	31.91
C-9	50.14	50.14	50.14	50.13
C-10	36.52	36.52	36.52	36.51
C-11	21.09	21.09	21.09	21.09
C-12	39.78	39.80	39.79	39.77
C-13	42.34	42.35	42.31	42.33
C-14	56.75	56.77	56.77	56.75
C-15	24.31	24.31	24.30	24.30
C-16	28.26	28.28	28.27	28.25
C-17	55.97	56.06	56.06	55.98
C-18	11.86	11.86	11.88	11.88
C-19	19.40	19.40	19.39	19.39
C-20	36.35	36.12	36.09	36.14
C-21	18.88	18.76	18.73	18.85
C-22	34.12	33.78	34.00	33.90
C-23	25.95	26.97	25.68	26.97
C-24	41.78	41.26	41.02	41.56
C-25	37.40	37.35	37.30	37.76
C-26	66.63	66.83	66.82	66.59
C-27	12.94	12.31	12.47	13.15
C-28	24.07	22.41	23.74	22.42
C-29	12.26	12.39	11.98	12.12

^{13}C N.m.r. spectra were determined at 50 MHz in [^{2}H]chloroform and the data are shown in p.p.m.

The firm assignments established here, indicate the need to revise the biosynthetic stereochemistry of the 24-ethylsterol side-chains. As shown in Scheme 5, the C-26 and C-27 of poriferasterol must originate from C-6 and C-2 of MVA, respectively, instead of *vice versa* as suggested by Nicotra on the basis of evidence from labelling patterns of [1,2-$^{13}C_2$]acetate in *Ochromanas malhamensis*.[21] Here we must also revise the C-26 and C-27 origin of sitosterol and stigmasterol in some higher plants,[2,22] and 24β-sterols in *Trichosanthes kirilowii*[4,23] as arising from C-6 and C-2 of MVA, respectively (see Scheme 1).

Experimental

All reactions were carried out in dry solvents under a nitrogen atmosphere. M.p.s were measured with a Yanagimoto micro melting point apparatus and are uncorrected. Unless otherwise stated, ^1H n.m.r. spectra were taken for solutions in [^{2}H]chloroform with Varian VXR-200 and XL-400 spectrometers, i.r. spectra for Nujol mulls, and optical rotations for solutions in chloroform. ^{13}C N.m.r. spectra were determined on a Varian XL-200 spectrometer operating at 50.057 MHz in [^{2}H]chloroform using a 10-mm spinning spherical tube. Chemical shifts are given in p.p.m. downfield from internal tetramethylsilane. Mass spectra were recorded on a Hitachi RMU-8GN spectrometer.

(22S,23E)-6β-*Methoxy*-3α,5-*cyclo*-26-*nor*-5α-*cholest*-23-*en*-22-*ol* (**14**).—A solution of compound (**12**) (1.37 g, 3.44 mmol) in ethanol (30 ml) and THF (35 ml) was added to liquid ammonia (70 ml) at −78 °C. To this solution was added portionwise lithium (460 mg, 66 mmol) over 1 h, and the mixture was stirred for 1 h. Solid ammonium chloride (4.0 g, 75 mmol) was added cautiously and the ammonia was allowed to evaporate. The residue was diluted with water and extracted with ether. The extract was washed with water, dried (Na_2SO_4), and evaporated under reduced pressure to give the crude crystals, which was recrystallized from acetone–hexane to give (**14**) (962 mg, 70%), m.p. 136—137 °C; $[\alpha]_D^{24}$ +29.8° (±1.4°) (*c* 0.506); $\nu_{max.}$ 3 555, 1 098, 1 020, and 974 cm^{-1}; δ_H 0.43 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.65 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.73 (3 H, s, 18-H), 0.90 (3 H, d, *J* 6.5 Hz, 21-H), 1.00 (3 H, t, *J* 7.5 Hz, 27-H), 1.02 (3 H, s, 19-H), 2.78 (1 H, t, *J* 2.5 Hz, 6-H), 3.33 (3 H, s, OMe), 4.22 (1 H, d, *J* 5 Hz, 22-H), and 5.50 and 5.66 (2 H, AB part of $ABMX_2$, *J* 15.5, 6, and 5 Hz, 23- and 24-H) (Found: C, 80.95; H, 11.2. $C_{27}H_{44}O_2$ requires C, 80.95; H, 11.05%).

The mother liquid (437 mg) was purified by chromatography on silica gel [Lobar B, toluene–ethyl acetate (49:1) as eluant] to give additional (**14**) (189 mg, 14%) and its *Z*-isomer[10] (145 mg, 11%).

(22R,23E)-6β-*Methoxy*-3α,5-*cyclo*-26-*nor*-5α-*cholest*-23-*en*-22-*ol* (**15**).—Compound (**13**) (1.75 g, 4.40 mmol) was reduced in the same manner as described above to give the crude product (1.461 g, 83%), which was shown by ^1H n.m.r. to be a 9:1 mixture of (**15**) and its *Z*-isomer.[10] The product was purified by chromatography on silica gel [Lobar C and B, toluene–ethyl acetate (19:1) as eluant] to give (**15**) (968 mg, 55%). Crystallization from light petroleum gave an analytical sample, m.p. 101—102 °C; $[\alpha]_D^{25}$ +48.5° (±2.9°) (*c* 0.303), $\nu_{max.}$ 3 390, 1 098, 1 086, 1 019, 997, and 975 cm^{-1}; δ_H 0.43 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.65 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.75 (3 H, s, 18-H), 0.95 (3 H, d, *J* 7 Hz, 21-H), 1.00 (3 H, t, *J* 7.5 Hz, 27-H), 1.02 (3 H, s, 19-H), 2.77 (1 H, t, *J* 2.5 Hz, 6-H), 3.32 (3 H, s, OMe), 4.13 (1 H, dd, *J* 7.5 and 3.5 Hz, 22-H), 5.46 (1 H, ddt, *J* 15.5, 7.5, and 1.4 Hz, 23-H), and 5.70 (1 H, dt, *J* 15.5 and 6 Hz, 24-H) (Found: C, 80.8; H, 10.85. $C_{27}H_{44}O_2$ requires C, 80.95; H, 11.05%).

Table 5. ^{13}C N.m.r. signal assignments of Δ^{22}-26-hydroxysterols (**46**)—(**49**)

C-No	(46)	(47)	(48)	(49)
C-1	37.28	37.26	37.27	37.27
C-2	31.67	31.67	31.66	31.66
C-3	71.79	71.80	71.80	71.79
C-4	42.27	42.28	42.31	42.29
C-5	140.78	140.76	140.76	140.76
C-6	121.67	121.66	121.67	121.66
C-7	31.91	31.90	31.90	31.90
C-8	31.91	31.90	31.90	31.90
C-9	50.16	50.15	50.15	50.15
C-10	36.53	36.52	36.52	36.52
C-11	21.08	21.07	21.08	21.08
C-12	39.54	39.68	39.69	39.68
C-13	42.32	42.31	42.27	42.29
C-14	56.84	56.82	56.84	56.81
C-15	24.35	24.35	24.38	24.38
C-16	28.81	28.82	28.94	28.83
C-17	55.81	55.84	55.79	55.88
C-18	12.07	12.06	12.04	12.04
C-19	19.40	19.39	19.40	19.40
C-20	40.40	40.30	40.54	40.42
C-21	21.08	20.97	21.10	21.08
C-22	139.16	138.57	139.22	138.66
C-23	128.09	130.08	127.89	129.83
C-24	46.04	48.02	45.84	47.84
C-25	39.69	39.96	39.55	39.94
C-26	67.26	66.85	67.31	66.77
C-27	12.42	15.08	12.23	15.15
C-28	25.96	24.92	25.92	24.90
C-29	11.95	12.15	12.00	12.00

^{13}C N.m.r. spectra were determined at 50 MHz in [^{2}H]chloroform and the data are shown in p.p.m.

(22S,23E)-6β-*Methoxy*-3α,5-*cyclo*-26-*nor*-5α-*cholest*-23-*en*-22-*yl Propionate* (**16**).—To a stirred solution of the alcohol (**14**) (1.151 g, 2.88 mmol) in pyridine (8 ml) at 0 °C were added propionic anhydride (1.0 ml, 7.8 mmol) and 4-dimethylaminopyridine (30 mg, 0.25 mmol), and the mixture was left at room temperature for 16 h. Water (0.5 ml) was added to destroy the excess of anhydride after which the product was extracted with ether. The extract was successively washed with 1M hydrochloric acid, water, 1M sodium carbonate, and water, dried (Na_2SO_4), and evaporated to give the oily product. This was purified by flash chromatography [5 g of silica gel, toluene–ethyl acetate (19:1) as eluant] to give (**16**) (1.31 g, 100%) as an oil; $\nu_{max.}$($CHCl_3$) 1 711, 1 177, 1 091, 1 075, and 965 cm^{-1}; δ_H 0.42 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.64 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.71 (3 H, s, 18-H), 0.97 (3 H, d, *J* 7 Hz, 21-H), 0.97 (3 H, t, *J* 7.5 Hz, 27-H), 1.01 (3 H, s, 19-H), 1.14 (3 H, t, *J* 7.5 Hz, COEt), 2.78 (1 H, br s, 6-H), 3.32 (3 H, s, OMe), and 5.23—5.85 (3 H, m, 22-H, 23-H, and 24-H).

(22R,23E)-6β-*Methoxy*-3α,5-*cyclo*-26-*nor*-5α-*cholest*-23-*en*-22-*yl Propionate* (**17**).—The alcohol (**15**) (905 mg, 2.26 mmol) was treated with propionic anhydride (0.60 ml, 4.52 mmol) in the same manner as described above to give the ester, which was purified by flash chromatography [4 g of silica gel, toluene–ethyl acetate (19:1) as eluant] to give (**18**) (1.03 g, 100%) as an oil; $\nu_{max.}$($CHCl_3$) 1 709, 1 177, 1 089, 1 072, and 965 cm^{-1}; δ_H 0.41 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.62 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.72 (3 H, s, 18-H), 0.96 (3 H, d, *J* 7 Hz, 21-H), 0.98 (3 H, t, *J* 7 Hz, 27-H), 1.00 (3 H, s, 19-H), 1.11 (3 H, t, *J* 7.5 Hz, OEt), 2.75 (1 H, br s, 6-H), 3.31 (3 H, s, OMe), and 5.20—5.94 (3 H, m, 22-H, 23-H, and 24-H).

(22E,24S,25S)-6β-*Methoxy*-3α,5-*cyclo*-5α-*stigmast*-22-*en*-26-*oic Acid* (**22**).—A stirred solution of di-isopropylamine (0.52 ml, 3.73 mmol) in THF (10 ml) at 0 °C was treated with a 1.4M solution of butyl-lithium in hexane (2.34 ml, 3.28 mmol). After 20 min the solution was cooled to −78 °C, and a solution of the ester (**16**) (1.00 g, 2.19 mmol) in THF (10 ml) was added dropwise. The solution was stirred for 10 min and then a solution of t-butyldimethylsilyl chloride (562 mg, 3.73 mmol) in THF (1.5 ml) and HMPA (1.5 ml) was added over 5 min. After 5 min the cooling bath was removed and the mixture was allowed to warm over 30 min to room temperature. The Claisen rearrangement reaction was allowed to proceed at this temperature for 30 min, while progress was followed by t.l.c.; the reaction mixture was then extracted with ether. The extract was washed with water, dried (Na_2SO_4), and evaporated to leave the crude silyl ester (1.30 g). This was dissolved in THF (20 ml) and tetraethylammonium fluoride hydrate (510 mg, 3.42 mmol) was added. The mixture was stirred at room temperature for 1 h, and then poured into cold 5% aqueous sodium hydroxide (15 ml). The aqueous solution was washed with ether and acidified with 4M hydrochloric acid, and the product was extracted with ether to give (**22**) [*ca.* 9:1 mixture with (**23**)] (920 mg, 92%) as a solid. Crystallization from ether–light petroleum gave pure (**22**) (580 mg, 58%), m.p. 144—145 °C; $\nu_{max.}$ 1 703, 1 102, and 972 cm^{-1}; δ_H 0.45 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.65 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.73 (3 H, s, 18-H), 0.86 (3 H, t, *J* 7.5 Hz, 29-H), 1.00 (3 H, d, *J* 7 Hz, 21-H), 1.03 (3 H, s, 19-H), 1.08 (3 H, d, *J* 7 Hz, 27-H), 2.78 (1 H, t, *J* 2.5 Hz, 6-H), 3.33 (3 H, s, OMe), and 4.99 and 5.30 (2 H, AB part of ABMN, *J* 15, 9, and 9 Hz, 22-H and 23-H) (Found: C, 79.05; H, 10.5. $C_{30}H_{43}O_3$ requires C, 78.9; H, 10.6%).

(22E,24R,25R)-6β-*Methoxy*-3α,5-*cyclo*-5α-*stigmast*-22-*en*-26-*oic Acid* (**24**).—Lithium di-isopropylamide solution was prepared by dropwise addition of an 1.4M solution of butyl-lithium in hexane (1.05 ml, 1.48 mmol) to a stirred solution of di-isopropylamine (0.25 ml, 1.65 mmol) in THF (4 ml) at 0 °C. To this solution at −78 °C was added a solution of the ester (**17**) (450 mg, 0.99 mmol) in THF (5 ml). After 10 min, a solution of t-butyldimethylsilyl chloride (248 mg, 1.65 mmol) in THF (0.8 ml) and HMPA (0.8 ml) was added and the mixture was allowed to warm over 30 min to room temperature. Stirring was continued for 30 min after which extractive (ether) work-up gave the silyl ester (560 mg).

A solution of the silyl ester (560 mg) in THF (8.0 ml) was treated with tetraethylammonium fluoride hydrate (220 mg, 1.48 mmol) at room temperature for 1 h. Work-up as described above for the preparation of (**22**) gave (**24**) [*ca.* 9:1 mixture with (**25**)] (400 mg, 88%), which was crystallized from ether–light petroleum to give pure (**24**) (257 mg, 57%), m.p. 156—157 °C; $\nu_{max.}$ 1 707, 1 004, and 976 cm^{-1}; δ_H 0.43 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.65 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.75 (3 H, s, 18-H), 0.87 (3 H, t, *J* 7.5 Hz, 29-H), 1.04 (3 H, d, *J* 6.5 Hz, 21-H), 1.04 (3 H, s, 19-H), 1.11 (3 H, d, *J* 7 Hz, 27-H), 2.78 (1 H, t, *J* 2.5 Hz, 6-H), 3.35 (3 H, s, OMe), and 5.00 and 5.31 (2 H, AB part of ABMN, *J* 15, 9, and 9 Hz, 22-H and 23-H) (Found: C, 78.95; H, 10.65. $C_{30}H_{48}O_3$ requires C, 78.9; H, 10.6%).

(22E,24S,25S)-6β-*Methoxy*-3α,5-*cyclo*-5α-*stigmast*-22-*en*-26-*ol* (**26**).—To a solution of the acid (**22**) (562 mg, 1.23 mmol) in ether (15 ml) at 0 °C was added an excess of ethereal diazomethane. After 30 min the solvent was evaporated and the residue was purified by flash chromatography [4 g of silica gel, toluene–ethyl acetate (9:1) as eluant] to give *methyl* (22E,24S,25S)-6β-*methoxy*-3α,5-*cyclo*-5α-*stigmast*-22-*en*-26-*oate* (**22a**) (560 mg, 97%). Crystallization from ethanol gave an analytical sample, m.p. 85—86 °C; δ_H 0.42 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.64 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.74 (3 H, s, 18-H), 0.83 (3 H, t, *J* 7.5 Hz, 29-H), 1.00 (3 H, d, *J* 7 Hz, 21-H), 1.03 (3 H, s, 19-H), 1.06 (3 H, d, *J* 7 Hz, 27-H), 2.74 (1 H, br s, 6-H), 3.31 (3 H,

s, OMe), 3.64 (3 H, s, OMe), and 4.94 and 5.24 (2 H, AB part of ABMN, *J* 15, 8, and 8 Hz, 22-H and 23-H).

To a solution of the above ester (460 mg, 0.99 mmol) in ether (7 ml) and THF (7 ml) at 0 °C was added $LiAlH_4$ (70 mg, 1.84 mmol). The mixture was stirred at 0 °C for 1 h and then water (1.0 ml) was added. The mixture was dried ($MgSO_4$), filtered, and evaporated to give (**26**) (435 mg, 100%), m.p. 86—87 °C (from light petroleum); $[\alpha]_D^{24}$ +27.9° (±1.6°) (*c* 0.426); $\nu_{max.}$ 3 285, 1 100, 1 029, and 975 cm^{-1}; δ_H 0.43 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.65 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.73 (3 H, s, 18-H), 0.83 (3 H, d, *J* 7 Hz, 27-H), 0.84 (3 H, t, *J* 7 Hz, 29-H), 1.02 (3 H, d, *J* 6.5 Hz, 21-H), 1.02 (3 H, s, 19-H), 2.77 (1 H, t, *J* 2.5 Hz, 6-H), 3.32 (3 H, s, OMe), 3.44 and 3.52 (2 H, AB part of ABX, *J* 11, 6.5, and 6 Hz, 26-H), and 5.09 and 5.25 (2 H, AB part of ABMN, *J* 15, 9, and 8 Hz, 22-H and 23-H) (Found: C, 81.3; H, 11.3. $C_{30}H_{50}O_2$ requires C, 81.4; H, 11.4%).

(22E,24S,25R)-6β-*Methoxy*-3α,5-*cyclo*-5α-*stigmast*-22-*en*-26-*ol* (**27**).—A solution of di-isopropylamine (0.64 ml, 4.53 mmol) in HMPA (3 ml) and THF (10 ml) at 0 °C was treated with a 1.4M solution of butyl-lithium in hexane (2.80 ml, 3.94 mmol). The solution was stirred at 0 °C for 20 min and then cooled to −78 °C. To this solution was added a solution of the ester (**16**) (1.38 g, 3.03 mmol) in HMPA (0.23 ml) and THF (0.77 ml), and the mixture was stirred for 10 min. A solution of t-butyldimethylsilyl chloride (684 mg, 4.53 mmol) in THF (4 ml) was added slowly and the mixture was allowed to warm over 30 min to room temperature. Stirring was continued for an additional 30 min after which the mixture was extracted with ether. The extract was washed with water, dried (Na_2SO_4), and evaporated, to leave the crude silyl ester (1.73 g).

A solution of the silyl ester (1.73 g) in THF (20 ml) was treated with tetraethylammonium fluoride hydrate (540 mg, 3.62 mmol) at room temperature for 1 h and then poured into cold 5% aqueous sodium hydroxide (20 ml). The aqueous solution was washed with ether and acidified with 4M hydrochloric acid, and the product was extracted with ether to give the acid (**23**) [*ca.* 4:1 mixture with (**22**)] (1.246 g, 90%).

The above mixture of acids was converted by the same manner as described for the preparation of (**26**) into a similar mixture of the alcohols (1.18 g, 98%), which was shown by h.p.l.c. analysis [column, μ-Bondasphere 5μ Si-100 Å, 150 × 3.9 mm i.d., hexane–ethyl acetate–chloroform (6:1:1), 1 ml/min, as eluant] to be a 78:22 mixture of (**27**) and (**26**), respectively. The mixture of the alcohols was chromatographed on preparative h.p.l.c. [column, μ-Bondasphere 5μ Si-100 Å, 150 × 19 mm i.d.; hexane–ethyl acetate–chloroform (20:1:1), 10 ml/min, as eluant] to give the alcohol (**27**) (600 mg, 50%), m.p. 93—94 °C (from light petroleum); $[\alpha]_D^{25}$ +20.8° (±1.2°) (*c* 0.500); $\nu_{max.}$ 3 492, 1 100, 1 092, 1 044, 1 023, and 976 cm^{-1}; δ_H 0.43 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.65 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.73 (3 H, s, 18-H), 0.83 (3 H, t, *J* 7.5 Hz, 29-H), 0.93 (3 H, d, *J* 6.5 Hz, 27-H), 1.02 (3 H, d, *J* 6.5 Hz, 21-H), 1.025 (3 H, s, 19-H), 2.77 (1 H, t, *J* 2.5 Hz, 6-H), 3.33 (3 H, s, OMe), 3.44 and 3.61 (2 H, AB part of ABX, *J* 10.5, 6, and 5 Hz, 26-H), and 5.10 and 5.25 (2 H, AB part of ABMN, *J* 15.5, 8.5, and 8 Hz, 22-H and 23-H) (Found: C, 81.3; H, 11.45. $C_{30}H_{50}O_2$ requires C, 81.4; H, 11.4%).

(22E,24R,25R)-6β-*Methoxy*-3α,5-*cyclo*-5α-*stigmast*-22-*en*-26-*ol* (**28**).—The acid (**24**) (225 mg, 0.49 mmol) was converted by the same manner as described for the preparation of (**26**) into (**28**) (210 mg, 96%), a colourless oil; $[\alpha]_D^{24}$ +33.2° (± 1.5°) (*c* 0.482); $\nu_{max.}$($CHCl_3$) 3 630, 3 435, 1 090, 1 078, 1 012, and 974 cm^{-1}; δ_H 0.44 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.65 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.73 (3 H, s, 18-H), 0.83 (3 H, d, *J* 7 Hz, 27-H), 0.84 (3 H, t, *J* 7 Hz, 29-H), 1.02 (3 H, s, 19-H), 1.02 (3 H, d, *J* 6.5 Hz, 21-H), 2.77 (1 H, t, *J* 2.5 Hz, 6-H), 3.33 (3 H, s, OMe), 3.44 and 3.51 (2 H, AB part of ABX, *J* 10.5, 7, and 6.5 Hz, 26-H), and 5.08 and 5.24 (2 H, AB part of ABMN, *J* 15, 9, and 8.5 Hz, 22-H and 23-H).

(22E,24R,25S)-6β-*Methoxy*-3α,5-*cyclo*-5α-*stigmast*-22-*en*-26-*ol* (**29**).—To a solution of di-isopropylamine (0.27 ml, 1.91 mmol) in HMPA (1.15 ml) and THF (3.85 ml) at 0 °C was added dropwise a 1.4M solution of butyl-lithium in hexane (1.18 ml, 1.65 mmol). After 20 min the solution was cooled to −78 °C, and treated with a solution of the ester (**17**) (580 mg, 1.27 mmol) in HMPA (1.15 ml) and THF (3.85 ml). After 10 min a solution of t-butyldimethylsilyl chloride (288 mg, 1.91 mmol) in THF (2 ml) was added, and the mixture was allowed to warm over 30 min to room temperature. After being stirred for 30 min, the mixture was diluted with water and extracted with ether to give the silyl ester (740 mg).

A solution of the silyl ester (740 mg) in THF (10 ml) was treated with tetraethylammonium fluoride hydrate (285 mg, 1.91 mmol) at room temperature for 1 h. Work-up as described above for the preparation of (**27**) gave (**25**) [*ca.* 4:1 mixture with (**24**)] (503 mg, 87%).

The above mixture of the acids (503 mg, 1.10 mmol) was converted as described for the preparation of (**28**) into a similar mixture of the alcohols (487 mg, 100%), which was shown by h.p.l.c. analysis (column, TSK gel ODS-120T, 250 × 4.5 mm i.d.; MeOH 1 ml/min as eluant) to be a 80:20 mixture of (**29**) and (**28**), respectively. The mixture of the alcohols was chromatographed on recycling preparative h.p.l.c. (column, TSK gel ODS-120T, 250 × 20 mm i.d.; MeOH, 8 ml/min, as eluant; pump, LC-908 Nihon Bunsekikogyo) to give the alcohol (**29**) (340 mg, 70%), a colourless oil; $[\alpha]_D^{24}$ +40.3° (±1.5°) (*c* 0.529); $\nu_{max.}$ 3 692, 3 440, 1 092, 1 013, and 969 cm^{-1}; δ_H 0.44 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.65 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.73 (3 H, s, 18-H), 0.82 (3 H, t, *J* 7.5 Hz, 29-H), 0.94 (3 H, d, *J* 7 Hz, 27-H), 1.02 (3 H, d, *J* 6.5 Hz, 21-H), 1.02 (3 H, s, 19-H), 2.77 (1 H, t, *J* 2.5 Hz, 6-H), 3.33 (3 H, s, OMe), 3.44 and 3.61 (2 H, AB part of ABX, *J* 10.5, 6.5, and 5 Hz, 26-H), and 5.09 and 5.24 (2 H, AB part of ABMN, *J* 15, 8.5, and 8 Hz, 22-H and 23-H).

General Procedure for Hydrogenation of Alcohols (**26**)—(**29**).—(24S,25S)-6β-*Methoxy*-3α,5-*cyclo*-5α-*stigmastan*-26-*ol* (**30**).—A solution of the alcohol (**26**) (340 mg, 0.77 mmol) in ethyl acetate (10 ml) was stirred with 10% Pd/C (45 mg) under a hydrogen atmosphere at room temperature for 16 h. The catalyst was filtered off and the filtrate was evaporated under reduced pressure. The residue was purified by chromatography on silica gel [Lobar B, toluene–ethyl acetate (9:1) as eluant] to give (**30**) (324 mg, 95%) as an oil, δ_H 0.43 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.65 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.71 (3 H, s, 18-H), 0.86 (3 H, d, *J* 7 Hz, 27-H), 0.88 (3 H, t, *J* 7 Hz, 29-H), 0.92 (3 H, d, *J* 6.5 Hz, 21-H), 1.02 (3 H, s, 19-H), 2.78 (1 H, t, *J* 2.5 Hz, 6-H), 3.32 (3 H, s, OMe), and 3.45 and 3.59 (2 H, AB part of ABX, *J* 10.5, 7.5, and 6 Hz, 26-H).

(24S,25R)-6β-*Methoxy*-3α,5-*cyclo*-5α-*stigmastan*-26-*ol* (**31**). This compound, prepared from the alcohol (**27**) (303 mg, 0.68 mmol) by the foregoing procedure in 91% yield, was a colourless oil; δ_H 0.42 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.64 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.70 (3 H, s, 18-H), 0.82 (3 H, d, *J* 7 Hz, 27-H), 0.87 (3 H, t, *J* 7 Hz, 29-H), 0.91 (3 H, d, *J* 6.5 Hz, 21-H), 1.01 (3 H, s, 19-H), 2.77 (1 H, t, *J* 2.5 Hz, 6-H), 3.31 (3 H, s, OMe), and 3.45 and 3.57 (2 H, AB part of ABX, *J* 10.5, 7, and 6 Hz, 26-H).

(24R,25R)-6β-*Methoxy*-3α,5-*cyclo*-5α-*stigmastan*-26-*ol* (**32**). This compound, prepared from the alcohol (**28**) (198 mg, 0.45 mmol) by the foregoing procedure in 93% yield, was a colourless oil; δ_H 0.43 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.65 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.72 (3 H, s, 18-H), 0.85 (3 H, d, *J* 7 Hz, 27-H), 0.87 (3 H, t, *J* 7 Hz, 29-H), 0.91 (3 H, d, *J* 6.5 Hz, 21-H), 1.02 (3 H, s, 19-H), 2.77 (1 H, t, *J* 2.5 Hz, 6-H), 3.32 (3 H, s, OMe), and 3.46 and 3.58 (2 H, AB part of ABX, *J* 10.5, 7, and 6 Hz, 26-H).

(24R,25S)-6β-*Methoxy*-3α,5-*cyclo*-5α-*stigmastan*-26-*ol* (**33**).

This compound, prepared from the alcohol (**29**) (242 mg, 0.55 mmol) by the foregoing procedure in 97% yield, was a colourless oil; δ_H (0.43 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.65 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.72 (3 H, s, 18-H), 0.87 (3 H, d, *J* 7 Hz, 27-H), 0.88 (3 H, t, *J* 7 Hz, 29-H), 0.92 (3 H, d, *J* 6 Hz, 21-H), 1.02 (3 H, s, 19-H), 2.77 (1 H, t, *J* 2.5 Hz, 6-H), 3.33 (3 H, s, OMe), and 3.45 and 3.61 (2 H, AB part of ABX, *J* 10.5, 7, and 6 Hz, 26-H).

General Procedure for Conversion of Alcohols (**26**)—(**33**) *into* [26-$^2H_1$]*Sterols* (**34**)—(**41**).—(25R)-[26-$^2H_1$]*Clionasterol* (**34**). To a solution of the alcohol (**30**) (49 mg, 0.11 mmol) in pyridine (1.0 ml) was added toluene-*p*-sulphonyl chloride (42 mg, 0.22 mmol), and the mixture was left at room temperature for 16 h. Work-up (extraction with ether) gave the tosylate (63 mg, 95%) as an oil; δ_H 0.43 (1 H, dd, *J* 8 and 5 Hz, 4-H), 0.65 (1 H, dd, *J* 5 and 3.5 Hz, 4-H), 0.70 (3 H, s, 18-H), 0.81 (3 H, t, *J* 7.5 Hz, 29-H), 0.82 (3 H, d, *J* 7 Hz, 27-H), 0.85 (3 H, d, *J* 6 Hz, 21-H), 1.02 (3 H, s, 19-H), 2.45 (3 H, s, ArMe), 2.78 (1 H, t, *J* 2.5 Hz, 6-H), 3.33 (3 H, s, OMe), 3.85 and 3.95 (2 H, AB part of ABX, *J* 9.5, 7.5, and 6 Hz, 26-H), 7.34 (2 H, d, *J* 8.5 Hz, ArH), and 7.80 (2 H, d, *J* 8.5 Hz, ArH).

To a suspension of $LiAl^2H_4$ (41 mg, 1.0 mmol) in ether (2.0 ml) at 0 °C was added a solution of the tosylate (63 mg, 0.105 mmol) in ether (2.0 ml). The mixture was stirred at room temperature for 2 h, cooled to 0 °C, and then diluted with water (0.5 ml). The mixture was dried ($MgSO_4$) and filtered, and the solvent was evaporated to give [26-$^2H_1$]*clionasterol* (45 mg, 100%) as an oil. This was dissolved in 2.0 ml of stock solution composed of *p*-TsOH (20 mg), water (2.0 ml), and dioxane (8.0 ml), and heated at 85 °C for 1.5 h. Pyridine (1 drop) was added and the mixture was extracted with ether. The product was purified by chromatography on silica gel [Lobar A, dichloromethane–ethyl acetate (1:1) as eluant] to give (**34**) [40 mg, 87% from (**30**)], m.p. 139.5—141 °C (from ethanol), *m/z* 415 (M^+); δ_H (±0.0006 p.p.m. at 400 MHz) 0.680 (3 H, s, 18-H), 0.793 (2 H, dt, *J* 7 and 1.8 Hz, 26-H), 0.830 (3 H, d, *J* 7 Hz, 27-H), 0.855 (3 H, t, *J* 7.5 Hz, 29-H), 0.926 (3 H, d, *J* 6.5 Hz, 21-H), 1.010 (3 H, s, 19-H), 3.526 (1 H, m, 3-H), and 5.354 (1 H, d, *J* 5.5 Hz, 6-H).

(25S)-[26-$^2H_1$]*Clionasterol* (**35**). The alcohol (**31**) (41 mg, 0.09 mmol) was similarly converted into (**35**) (30 mg, 78%), m.p. 139—141 °C (from ethanol), *m/z* 415 (M^+); δ_H(±0.0006 p.p.m. at 400 MHz) 0.680 (3 H, s, 18-H), 0.811 (3 H, d, *J* 7 Hz, 27-H), 0.814 (2 H, dt, *J* 7 and 1.8 Hz, 26-H), 0.855 (3 H, t, *J* 7.5 Hz, 29-H), 0.926 (3 H, d, *J* 6.5 Hz, 21-H), 1.010 (3 H, s, 19-H), 3.526 (1 H, m, 3-H), and 5.354 (1 H, d, *J* 5.5 Hz, 6-H).

(25S)-[26-$^2H_1$]*Sitosterol* (**36**). The alcohol (**32**) (40 mg, 0.09 mmol) was similarly converted into (**36**) (31 mg, 83%), m.p. 137.5—138 °C (from ethanol), *m/z* 415 (M^+); δ_H (±0.0006 p.p.m. at 400 MHz) 0.681 (3 H, s, 18-H), 0.813 (3 H, d, *J* 7 Hz, 27-H), 0.817 (2 H, dt, *J* 7 and 1.8 Hz, 26-H), 0.846 (3 H, t, *J* 7.5 Hz, 29-H), 0.922 (3 H, d, *J* 6.5 Hz, 21-H), 1.009 (3 H, s, 19-H), 3.526 (1 H, m, 3-H), and 5.353 (1 H, d, *J* 5.5 Hz, 6-H).

(25R)-[26-$^2H_1$]*Sitosterol* (**37**). The alcohol (**33**) (40 mg, 0.09 mmol) was similarly converted into (**37**) (34 mg, 90%), m.p. 137—138 °C (from ethanol); *m/z* 415 (M^+); δ_H(±0.0006 p.p.m. at 400 MHz) 0.681 (3 H, s, 18-H), 0.796 (2 H, dt, *J* 7 and 1.8 Hz, 26-H), 0.835 (3 H, d, *J* 7 Hz, 27-H), 0.846 (3 H, t, *J* 7.5 Hz, 29-H), 0.922 (3 H, d, *J* 6.5 Hz, 21-H), 1.009 (3 H, s, 19-H), 3.526 (1 H, m, 3-H), and 5.353 (1 H, d, *J* 5.5 Hz, 6-H).

(25R)-[26-$^2H_1$]*Poriferasterol* (**38**). The alcohol (**26**) (40 mg, 0.09 mmol) was similarly converted into (**38**) (32 mg, 85%), m.p. 158—159 °C (from ethanol), *m/z* 413 (M^+); δ_H(±0.0006 p.p.m. at 400 MHz) 0.697 (3 H, s, 18-H), 0.790 (3 H, d, *J* 7 Hz, 27-H), 0.811 (3 H, t, *J* 7.5 Hz, 29-H), 0.826 (2 H, dt, *J* 7 and 1.8 Hz, 26-H), 1.011 (3 H, s, 19-H), 1.024 (3 H, d, *J* 6.5 Hz, 21-H), 3.525 (1 H, m, 3-H), 5.022 and 5.158 (2 H, AB part of ABMN, *J* 15, 9, and 8.5 Hz, 22-H and 23-H), and 5.351 (1 H, d, *J* 5.5 Hz, 6-H).

(25S)-[26-$^2H_1$]*Poriferasterol* (**39**). The alcohol (**27**) (36 mg, 0.08 mmol) was similarly converted into (**39**) (29 mg, 86%), m.p. 157—158.5 °C (from ethanol); *m/z* 413 (M^+); δ_H(±0.0006 p.p.m. at 400 MHz) 0.698 (3 H, s, 18-H), 0.773 (2 H, dt, *J* 7 and 1.8 Hz, 26-H), 0.811 (3 H, t, *J* 7.5 Hz, 29-H), 0.843 (3 H, d, *J* 7 Hz, 27-H), 1.011 (3 H, s, 19-H), 1.025 (3 H, d, *J* 6.5 Hz, 21-H), 3.525 (1 H, m, 3-H), 5.022 and 5.158 (2 H, AB part of ABMN, *J* 15, 9, and 8.5 Hz, 22-H and 23-H), and 5.351 (1 H, d, *J* 5.5 Hz, 6-H).

(25S)-[26-$^2H_1$]*Stigmasterol* (**40**). The alcohol (**28**) (29.7 mg, 0.067 mmol) was similarly converted into (**40**) (24 mg, 86%), m.p. 169—170 °C (from ethanol); *m/z* 413 (M^+); δ_H(±0.0006 p.p.m. at 400 MHz) 0.699 (3 H, s, 18-H), 0.795 (3 H, d, *J* 7 Hz, 27-H), 0.805 (3 H, t, *J* 7.5 Hz, 29-H), 0.827 (2 H, dt, *J* 7 and 1.8 Hz, 26-H), 1.011 (3 H, s, 19-H), 1.022 (3 H, d, *J* 6.5 Hz, 21-H), 3.525 (1 H, m, 3-H), 5.018 and 5.151 (2 H, AB part of ABMN, *J* 15, 9, and 8.5 Hz, 22-H and 23-H), and 5.351 (1 H, d, *J* 5.5 Hz, 6-H).

(25R)-[26-$^2H_1$]*Stigmasterol* (**41**). The alcohol (**29**) (30 mg, 0.068 mmol) was similarly converted into (**41**) (13 mg, 46%), m.p. 169—170 °C (from ethanol), *m/z* 413 (M^+); δ_H(±0.0006 p.p.m. at 400 MHz) 0.699 (3 H, s, 18-H), 0.778 (2 H, dt, *J* 7 and 1.8 Hz, 26-H), 0.805 (3 H, t, *J* 7.5 Hz, 29-H), 0.845 (3 H, d, *J* 7 Hz, 27-H), 1.012 (3 H, s, 19-H), 1.022 (3 H, d, *J* 6.5 Hz, 21-H), 3.525 (1 H, m, 3-H), 5.018 and 5.151 (2 H, AB part of ABMN, *J* 15, 9, and 8.5 Hz, 22-H and 23-H), and 5.351 (1 H, d, *J* 5.5 Hz, 6-H).

General Procedure for Preparation of 26-*Hydroxysterols* (**42**)—(**49**).—(24S,25S)-*Stigmast-5-ene*-3β,26-*diol* (**42**). To a stirred solution of the alcohol (**30**) (153 mg, 0.34 mmol) in dioxane (4.8 ml) and water (1.2 ml) was added *p*-TsOH (12 mg, 0.07 mmol). The mixture was heated at 85 °C for 1.5 h, cooled to room temperature, and then diluted with ethyl acetate (30 ml) containing pyridine (0.1 ml). The organic layer was washed with water, dried (Na_2SO_4), and evaporated under reduced pressure. The residue was purified by flash chromatography [1 g of silica gel, dichloromethane–ethyl acetate (1:1) as eluant] to give (**42**) (137 mg, 93%) as a solid, which was recrystallized from acetone to give an analytical sample, m.p. 188—189 °C; $[\alpha]_D^{25}$ −43.9° (±1.7°) (*c* 0.506); $\nu_{max.}$ 3 280, 1 049, 1 015, and 950 cm^{-1}; δ_H 0.68 (3 H, s, 18-H), 0.86 (3 H, d, *J* 7 Hz, 27-H), 0.88 (3 H, t, *J* 7.5 Hz, 29-H), 0.92 (3 H, d, *J* 6 Hz, 21-H), 1.01 (3 H, s, 19-H), 3.45 and 3.60 (2 H, AB part of ABX, *J* 10.5, 7.5, and 6 Hz, 26-H), and 5.35 (1 H, d, *J* 5 Hz, 6-H) (Found: C, 80.9; H, 11.65. $C_{29}H_{50}O_2$ requires C, 80.85; H, 11.7%).

(24S,25R)-*Stigmast-5-ene*-3β,26-*diol* (**43**). The alcohol (**31**) (250 mg, 0.56 mmol) was similarly treated with *p*-TsOH in aqueous dioxane to give (**43**) (224 mg, 93%), m.p. 173—175 °C (from acetone); $[\alpha]_D^{24}$ −30.8° (±1.4°) (*c* 0.500); $\nu_{max.}$ 3 325, 1 057, and 1 022 cm^{-1}; δ_H 0.67 (3 H, s, 18-H), 0.82 (3 H, d, *J* 7 Hz, 27-H), 0.87 (3 H, t, *J* 7 Hz, 29-H), 0.91 (3 H, d, *J* 6.5 Hz, 21-H), 1.00 (3 H, s, 19-H), 3.45 and 3.57 (2 H, AB part of ABX, *J* 10.5, 7.5, and 6 Hz, 26-H), and 5.34 (1 H, d, *J* 5 Hz, 6-H) (Found: C, 80.6; H, 11.65. $C_{29}H_{50}O_2$ requires C, 80.85; H, 11.7%).

(24R,25R)-*Stigmast-5-ene*-3β,26-*diol* (**44**). The alcohol (**32**) (144 mg, 0.32 mmol) was similarly treated with *p*-TsOH in aqueous dioxane to give (**44**) (135 mg, 97%), m.p. 166—167 °C (from acetone–dichloromethane); $[\alpha]_D^{24.5}$ −29.4° (±1.4°) (*c* 0.504); $\nu_{max.}$ 3 274, 1 058, and 1 026 cm^{-1}; δ_H 0.68 (3 H, s, 18-H), 0.85 (3 H, d, *J* 7 Hz, 27-H), 0.87 (3 H, t, *J* 7 Hz, 29-H), 0.92 (3 H, d, *J* 6.5 Hz, 21-H), 1.01 (3 H, s, 19-H), 3.47 and 3.59 (2 H, AB part of ABX, *J* 10.5, 7, and 6 Hz, 26-H), and 5.36 (1 H, d, *J* 5 Hz, 6-H) (Found: C, 80.8; H, 11.7. $C_{29}H_{50}O_2$ requires C, 80.85; H, 11.7%).

(24R,25S)-*Stigmast-5-ene*-3β,26-*diol* (**45**). The alcohol (**33**) (194 mg, 0.44 mmol) was similarly treated with *p*-TsOH in aqueous dioxane to give (**45**) (173 mg, 92%), m.p. 193—195 °C (from acetone–dichloromethane); $[\alpha]_D^{24.5}$ −41.6° (±1.6°) (*c* 0.510); $\nu_{max.}$ 3 255, 1 047, and 1 017 cm^{-1}; δ_H 0.68 (3 H, s, 18-H), 0.87 (3 H, d, *J* 7 Hz, 27-H), 0.88 (3 H, t, *J* 7 Hz, 29-H), 0.93 (3 H, d, *J* 6.5 Hz, 21-H), 1.01 (3 H, s, 19-H), 3.53 (1 H, m, 3-H), 3.45 and 3.61 (2 H, AB part of ABX, *J* 10.5, 7.5, and 6 Hz, 26-H), and

Table 6. Crystallographic details for (**22a**) and (**24**)

	(**22a**)	(**24**)
Formula	$C_{31}H_{50}O_3$	$C_{30}H_{48}O_3$
M	470.73	456.71
Crystal system	Orthorhombic	Monoclinic
Space group	$P2_12_12_1$	$C2$
a/Å	11.193(1)	21.497(2)
b/Å	35.701(6)	9.098(1)
c/Å	7.234(1)	14.797(2)
β/°		96.34(1)
V/Å^3	2 890.6(7)	2 876.3(6)
Z	4	4
D_x/g cm^{-1}	1.082	1.055
μ(Cu-K_α)/cm^{-1}	5.3	5.2
No. of observed reflections	1 680 [$I > 1.5\sigma(I)$]	1 899 [$I > 2\sigma(I)$]
R	0.065	0.049
R_w	0.067	0.039
S	1.1778	0.985

Table 7. Atomic co-ordinates for (**22a**) with their e.s.d.s in parentheses

	x	y	z
C(1)	−0.283 3(6)	0.843 4(2)	1.258 0(10)
C(2)	−0.350 5(6)	0.837 1(2)	1.080 5(14)
C(3)	−0.260 7(7)	0.821 3(2)	0.944 7(10)
C(4)	−0.183 1(7)	0.790 0(2)	1.022 9(10)
C(5)	−0.136 3(6)	0.829 9(1)	1.024 1(9)
C(6)	−0.037 8(6)	0.840 5(2)	0.891 9(8)
C(7)	−0.033 2(7)	0.882 0(2)	0.854 5(8)
C(8)	−0.031 8(5)	0.904 5(1)	1.038 2(7)
C(9)	−0.141 0(5)	0.895 1(1)	1.153 2(8)
C(10)	−0.152 2(5)	0.852 2(1)	1.201 5(7)
C(11)	−0.148 2(6)	0.920 1(2)	1.328 0(9)
C(12)	−0.144 8(6)	0.962 1(1)	1.281 4(9)
C(13)	−0.032 2(5)	0.971 6(1)	1.170 4(7)
C(14)	−0.034 3(5)	0.946 8(1)	0.996 7(7)
C(15)	0.064 5(7)	0.963 1(2)	0.873 1(8)
C(16)	0.055 7(6)	1.006 2(2)	0.914 7(9)
C(17)	−0.029 0(5)	1.011 2(1)	1.078 1(8)
C(18)	−0.063 7(6)	0.840 1(2)	1.350 4(8)
C(19)	0.079 9(6)	0.966 4(1)	1.286 8(9)
C(20)	0.003 8(5)	1.045 2(1)	1.197 3(9)
C(21)	−0.088 8(8)	1.052 7(2)	1.347 6(12)
C(22)	0.016 1(5)	1.080 3(1)	1.083 0(9)
C(23)	0.111 2(5)	1.100 6(1)	1.064 4(10)
C(24)	0.128 4(5)	1.136 0(1)	0.956 3(9)
C(25)	0.172 2(5)	1.167 3(2)	1.086 1(9)
C(26)	0.081 7(7)	1.175 3(2)	1.238 8(11)
C(27)	0.194 7(6)	1.203 3(2)	0.982 0(11)
O(28)	0.124 3(5)	1.219 2(1)	0.886 0(10)
O(29)	0.303 8(4)	1.216 0(1)	1.014 6(6)
C(30)	0.335 9(8)	1.252 4(2)	0.933 5(10)
C(31)	0.213 6(6)	1.130 8(2)	0.794 9(10)
C(32)	0.166 7(7)	1.103 7(2)	0.652 4(11)
O(33)	0.080 1(4)	0.831 9(1)	0.966 8(6)
C(34)	0.107 3(7)	0.793 1(2)	0.975 5(12)

5.35 (1 H, d, J 5 Hz, 6-H) (Found: C, 80.85; H, 11.8. $C_{29}H_{50}O_2$ requires C, 80.85; H, 11.7%).

(22E,24S,25S)-*Stigmasta*-5,22-*diene*-3β,26-*diol* (**46**). The alcohol (**26**) (100 mg, 0.23 mmol) was similarly treated with *p*-TsOH in aqueous dioxane to give (**46**) (95 mg, 98%), m.p. 183—184 °C (from acetone); $[\alpha]_D^{25}$ −56.1° (±1.9°) (*c* 0.503); $\nu_{max.}$ 3 240, 1 068, 1 050, 1 040, and 972 cm^{-1}; δ_H 0.70 (3 H, s, 18-H), 0.83 (3 H, d, J 7 Hz, 27-H), 0.84 (3 H, t, J 7 Hz, 29-H), 1.01 (3 H, s, 19-H), 1.02 (3 H, d, J 6 Hz, 21-H), 3.45 and 3.52 (2 H, AB part of ABX, J 10.5, 6.5, and 6 Hz, 26-H), 5.09 and 5.26 (2 H, AB part of ABMN, J 15, 9, and 8 Hz, 22-H and 23-H), and 5.35 (1 H, d, J 5 Hz, 6-H) (Found: C, 81.25; H, 11.45. $C_{29}H_{48}O_2$ requires C, 81.25; H, 11.3%).

Table 8. Fractional atomic co-ordinates for (**24**) with their e.s.d.s in parentheses

	x	y	z
C(1)	0.355 9(4)	0.6758	0.974 9(4)
C(2)	0.414 8(4)	0.757 1(16)	0.993 3(5)
C(3)	0.401 9(4)	0.905 3(15)	0.953 1(6)
C(4)	0.340 6(4)	0.983 2(13)	0.972 1(6)
C(5)	0.344 4(3)	0.896 5(12)	0.884 6(4)
C(6)	0.341 0(4)	0.977 6(12)	0.798 9(5)
C(7)	0.371 6(4)	0.892 2(12)	0.726 4(5)
C(8)	0.342 4(3)	0.740 5(11)	0.713 6(4)
C(9)	0.351 0(2)	0.655 9(11)	0.802 5(3)
C(10)	0.323 3(2)	0.737 3(11)	0.883 1(3)
C(11)	0.329 2(3)	0.497 2(12)	0.790 1(4)
C(12)	0.358 4(3)	0.416 1(11)	0.714 0(4)
C(13)	0.346 8(2)	0.498 0(11)	0.624 2(3)
C(14)	0.373 0(3)	0.654 1(11)	0.641 1(4)
C(15)	0.372 3(4)	0.720 4(13)	0.545 7(5)
C(16)	0.386 5(4)	0.588 2(12)	0.486 8(4)
C(17)	0.386 3(3)	0.449 8(11)	0.546 5(3)
C(18)	0.277 4(3)	0.499 5(13)	0.588 1(5)
C(19)	0.251 9(3)	0.722 2(13)	0.875 7(5)
C(20)	0.367 6(3)	0.310 7(12)	0.492 0(3)
C(21)	0.369 3(4)	0.172 7(12)	0.549 7(5)
C(22)	0.410 2(2)	0.288 6(11)	0.418 0(3)
C(23)	0.392 5(3)	0.263 0(12)	0.334 0(3)
C(24)	0.433 1(3)	0.231 3(12)	0.259 1(3)
C(25)	0.421 1(3)	0.349 8(12)	0.185 9(4)
C(26)	0.455 1(2)	0.319 6(12)	0.103 6(3)
C(27)	0.440 6(4)	0.504 6(13)	0.221 5(5)
C(28)	0.420 5(4)	0.080 4(13)	0.219 6(5)
C(29)	0.436 7(6)	−0.043 8(14)	0.292 0(7)
O(30)	0.279 1(3)	1.010 3(11)	0.757 4(3)
C(31)	0.245 4(4)	1.111 5(15)	0.802 1(7)
O(32)	0.425 7(2)	0.314 8(11)	0.027 5(2)
O(33)	0.514 5(2)	0.306 1(12)	0.117 2(2)

(22E,24S,25R)-*Stigmasta*-5,22-*diene*-3β,26-*diol* (**47**). The alcohol (**27**) (90 mg, 0.20 mmol) was similarly treated with *p*-TsOH in aqueous dioxane to give (**47**) (81 mg, 93%), m.p. 187—189 °C (from ethyl acetate); $[\alpha]_D^{24}$ −63.7° (±2.0°) (*c* 0.512); $\nu_{max.}$ 3 350, 1 064, 1 030, and 979 cm^{-1}; δ_H 0.70 (3 H, s, 18-H), 0.82 (3 H, t, J 7 Hz, 29-H), 0.93 (3 H, d, J 6.5 Hz, 27-H), 1.01 (3 H, s, 19-H), 1.03 (3 H, d, J 6 Hz, 21-H), 3.44 and 3.61 (2 H, AB part of ABX, J 11, 6, and 5 Hz, 26-H), 5.11 and 5.26 (2 H, AB part of ABMN, J 15, 8.5, and 8 Hz, 22-H and 23-H), and 5.35 (1 H, d, J 5.5 Hz, 6-H) (Found: C, 81.0; H, 11.2. $C_{29}H_{48}O_2$ requires C, 81.25; H, 11.3%).

(22E,24R,25R)-*Stigmasta*-5,22-*diene*-3β,26-*diol* (**48**). The alcohol (**28**) (61 mg, 0.14 mmol) was similarly treated with *p*-TsOH in aqueous dioxane to give (**48**) (54 mg, 92%), m.p. 181—182 °C (from acetone); $[\alpha]_D^{24.5}$ −46.8° (±1.7°) (*c* 0.500); $\nu_{max.}$ 3 255, 1 058, 1 022, 980, and 957 cm^{-1}; δ_H 0.70 (3 H, s, 18-H), 0.83 (3 H, d, J 7 Hz, 27-H), 0.84 (3 H, t, J 7 Hz, 29-H), 1.01 (3 H, s, 19-H), 1.03 (3 H, d, J 6.5 Hz, 21-H), 3.44 and 3.51 (2 H, AB part of ABX, J 11, 7, and 6.5 Hz, 26-H), 5.08 and 5.24 (2 H, AB part of ABMN, J 15, 9, and 8.5 Hz, 22-H and 23-H), and 5.35 (1 H, d, J 5 Hz, 6-H) (Found: C, 81.3; H, 11.25. $C_{29}H_{48}O_2$ requires C, 81.25; H, 11.3%).

(22E,24R,25S)-*Stigmasta*-5,22-*diene*-3β,26-*diol* (**49**). The alcohol (**29**) (102 mg, 0.23 mmol) was similarly treated with *p*-TsOH in aqueous dioxane to give (**49**) (88 mg, 90%), m.p. 163—164 °C (from acetone); $[\alpha]_D^{24.5}$ −40.4° (±1.6°) (*c* 0.500); $\nu_{max.}$ 3 330, 1 052, 1 020, and 972 cm^{-1}; δ_H 0.70 (3 H, s, 18-H), 0.82 (3 H, t, J 7 Hz, 29-H), 0.94 (3 H, d, J 7 Hz, 27-H), 1.01 (3 H, s,

19-H), 1.03 (3 H, d, J 6 Hz, 21-H), 3.58 (1 H, m, 3-H), 3.43 and 3.62 (2 H, AB part of ABX, J 10.5, 6, and 5 Hz, 26-H), 5.09 and 5.24 (2 H, AB part of ABMN, J 15, 8.5, and 8 Hz, 22-H and 23-H), and 5.35 (1 H, d, J 5 Hz, 6-H) (Found: C, 81.05; H, 11.3. $C_{29}H_{48}O_2$ requires C, 81.25; H, 11.3%).

X-*Ray Structure Determination of Compound* (**22a**).—Suitable crystals were grown by recrystallization from ethanol. A crystal with dimensions of 0.40 × 0.15 × 0.03 mm was used. *X*-Ray measurements were performed on a Rigaku AFC-5 diffractometer with graphite-monochromated Cu-K_α radiation (λ = 1.541 78 Å). Cell dimensions were refined on diffractometer angles for 24 automatically centred reflections ($19 < \theta < 32°$). Integrated intensities were recorded in the range $\theta \leq 70°$ ($+h$, $+k$, $-l$) with $\omega/2\theta$ mode (ω scan width = $1.2° + 0.2° \tan\theta$). Three standard reflections monitored every 100 reflections showed no significant change during data collection. Of the 3 061 independent reflections measured, 1 680 with $I \geq 1.5\sigma(I)$ were considered as observed. The data were corrected for Lorentz and polarization factors but not for absorption effects. The crystal structure were solved using the program MULTAN87.[24] All hydrogen atoms could be located on difference density maps and their positions were refined with fixed isotropic thermal parameters. The structure was refined by block-diagonal least squares with anisotropic thermal parameters for the non-hydrogen atoms. The function $\Sigma w(\Delta F)^2$ was minimized with a weighting scheme $w = [\sigma^2(F_o) + 0.0011F_o^2]^{-1}$ for 1 645 reflections with $w^{\frac{1}{2}}|\Delta F| < 3$ and $w = 0$ otherwise. No correction was for dispersion effects. Final residual densities were in the range $-0.18 \leq \Delta\rho \leq 0.19$ e Å^{-3}. Atomic scattering factors were calculated using the analytical expression $f = \Sigma[a_i\exp(b_i\sin^2\theta/\lambda^2)] + c$ (i = 1, ···, 4).[25] Calculations were performed on a FACOM M-730 computer.

Crystallographic data are summarized in Table 6. Atomic co-ordinates are listed in Table 7.* A PLUTO[26] stereoview of the molecule is shown in Figure 1.

X-*Ray Structure Determination of Compound* (**24**).—Suitable crystals were grown by recrystallization from ethanol. A crystal with dimensions of 0.20 × 0.35 × 0.15 mm was used. Cell dimensions were refined using 24 reflections ($23 < \theta < 28°$). Integrated intensities were recorded in the range $\theta \leq 60°$ ($+h$, $-k$, $\pm l$). Of the 2 297 independent reflections measured, 1 899 with $I \geq 2\sigma(I)$ were considered as observed. The structure was refined by minimizing $\Sigma w(\Delta F)^2$ with $w = 1$. Isotropic type-I extinction correction[27] was applied [the final extinction parameter $g = 1.50(6) \times 10^{-4}$]. Final residual densities were in the range $-0.13 \leq \Delta\rho \leq 0.14$ e Å^{-3}. Calculations were made on a VAX11/780 computer using the program system XTAL2.4[28] with the scattering factors as included in the program. The other features of data collection and structure determination are the same as with (**22a**). Crystallographic data are summarized in Table 6. Atomic co-ordinates are listed in Table 8.* A stereoview of the molecule is shown in Figure 1.

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* Supplementary data (see section 5.6.3. on Instruction for Authors, in *J. Chem. Soc., Perkin Trans. 1*, 1989, Issue 1). List of bond lengths, bond angles, hydrogen-atom co-ordinates, and thermal parameters have been deposited at the Cambridge Crystallographic Data Centre.

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