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Synthesis of Lanosterol Analogs with Lengthened Side Chains and Their Effects on Cholesterol Biosynthesis from Lanosterol¹⁾

YOSHIHIRO SATO* and YOSHIKO SONODA

*Kyoritsu College of Pharmacy, Shibakoen 1-chome,
Minato-ku, Tokyo 105, Japan*

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Starting from 3 β -acetoxy-25,26,27-trinorlanost-8-en-24-al (1), eight lanosterol analogs (10—17) with longer side chains than that of lanosterol were synthesized by Wittig reaction followed by catalytic hydrogenation. Cholesterol biosynthesis was examined in rat hepatic subcellular preparation (S₁₀) incubated with [24-³H]-lanosterol in the presence of each of the eight lanosterol analogs. Some of the analogs (10 and 12) caused slight inhibition, but 16 and 17 showed no inhibitory effect. The structure-inhibitory activity relationship of lanosterol analogs on cholesterol biosynthesis from lanosterol is discussed.

Keywords—cholesterol biosynthesis; [24-³H]-lanosterol; lanosterol analogs; rat hepatic subcellular preparation; inhibitory activity

The biosynthesis of cholesterol from lanosterol involves the removal of three methyl groups,²⁾ reduction of the Δ^{24} -double bond, and the migration of double bonds.^{3,4)} However, very few studies⁵⁾ have been carried out on the inhibition of cholesterol biosynthesis from lanosterol.

Recently, we reported the effects of lanosterol analogs,⁶⁾ cholesterol analogs,¹⁾ and oxygenated lanosterol derivatives⁷⁾ on cholesterol biosynthesis from lanosterol. From these studies, it was clear that both the side chain and skeleton structures are important in relation to the inhibitory effect. This study was carried out in order to determine whether lanosterol analogs with longer side chains than that of lanosterol could have an inhibitory effect.

The effects of eight lanosterol analogs (10—17) with longer side chains than that of lanosterol on cholesterol biosynthesis from [24-³H]-lanosterol in rat hepatic subcellular 10000 $\times$ g supernatant (S₁₀) fraction were studied. Compounds 10 and 12, with a little change in the side chains, exhibited slight inhibitory effects, but 16 and 17, which have long side chains, showed no activity.

Materials and Methods

Proton nuclear magnetic resonance (¹H-NMR) spectra, mass spectra (MS), and infrared (IR) spectra were recorded as described previously.⁸⁾ High-performance liquid chromatography (HPLC) was performed on a μ Bondapak-C₁₈ reverse-phase column (3.9 mm $\times$ 30 cm), using a Waters pump (model 510) and a Waters detector (model 480 spectrophotometer, set at 229 nm). Acetonitrile or acetonitrile–water (85:15, v/v) was used as an eluent (flow rate 2.0 ml/min, pressure 100 kg/cm²).

General Procedure for the Wittig Reaction—A solution of *n*-butyl lithium (14%, 1.2 ml) was added to a suspension of the appropriate alkyl triphenylphosphonium bromide (in the preparations of 4 and 6, the corresponding iodides were used) (1.2 g) in anhydrous benzene (20 ml) and the mixture was stirred at room temperature for 10 min. A solution of the aldehyde (1) (0.5 g) in anhydrous benzene (10 ml) was then added, and the mixture was stirred at room temperature for 24 h. After extraction of the neutral product with benzene, the extract was concentrated and the residue was column-chromatographed on silica gel (50 g). Elution with benzene gave the Wittig products (2a, b—9a, b) and their acetates. The yields of the Wittig products (2a, b—9a, b) and their acetates

were 43—55% and 18—31%, respectively, with the exception of **4a, b** and the acetates (17 and 5%). The Wittig products (**2a, b**—**9a, b**), which consisted of 24-*E* and 24-*Z* isomers, were recrystallized from MeOH to give colorless needles.

26-Methyl-27-norlanosta-8,24-dien-3 β -ol (2a, b)—mp 130—131 °C. *Anal.* Calcd for $C_{30}H_{50}O$: C, 84.44; H, 11.81. Found: C, 84.73; H, 11.73. MS m/z : 426 (M^+), 411, 393. IR $\nu_{\max}^{KBr}$ cm^{-1} : 962. 1H -NMR δ (ppm): 0.69 (3H, s, 18-CH₃), 0.81 (3H, s, 4 β -CH₃), 0.88 (3H, s, 14-CH₃), 0.98 (3H, s, 4 α -CH₃), 0.99 (3H, s, 19-CH₃), 3.06—3.30 (1H, m, 3-H), 5.20—5.40 (2H, m, 24, 25-H).

26-Ethyl-27-norlanosta-8,24-dien-3 β -ol (3a, b)—mp 131—132 °C. *Anal.* Calcd for $C_{31}H_{52}O$: C, 84.48; H, 11.89. Found: C, 84.13; H, 11.85. MS m/z : 440 (M^+), 425, 407. IR $\nu_{\max}^{KBr}$ cm^{-1} : 962. 1H -NMR δ (ppm): 0.69 (3H, s, 18-CH₃), 0.81 (3H, s, 4 β -CH₃), 0.88 (3H, s, 14-CH₃), 0.98 (3H, s, 4 α -CH₃), 0.99 (3H, s, 19-CH₃), 3.08—3.32 (1H, m, 3-H), 5.24—5.42 (2H, m, 24, 25-H).

26,26-Dimethyl-27-norlanosta-8,24-dien-3 β -ol (4a, b)—mp 130—131 °C. *Anal.* Calcd for $C_{31}H_{52}O$: C, 84.48; H, 11.89. Found: C, 84.60; H, 11.68. MS m/z : 440 (M^+), 425, 407. IR $\nu_{\max}^{KBr}$ cm^{-1} : 964. 1H -NMR δ (ppm): 0.68 (3H, s, 18-CH₃), 0.81 (3H, s, 4 β -CH₃), 0.88 (3H, s, 14-CH₃), 0.96 (6H, d, isopropyl-CH₃, $J=6.5$ Hz), 0.98 (3H, s, 4 α -CH₃), 0.99 (3H, s, 19-CH₃), 3.08—3.32 (1H, m, 3-H), 5.28—5.44 (2H, m, 24, 25-H).

26-n-Propyl-27-norlanosta-8,24-dien-3 β -ol (5a, b)—mp 130—131 °C. *Anal.* Calcd for $C_{32}H_{54}O$: C, 84.51; H, 11.97. Found: C, 84.22; H, 12.11. MS m/z : 454 (M^+), 439, 421. IR $\nu_{\max}^{KBr}$ cm^{-1} : 955. 1H -NMR δ (ppm): 0.69 (3H, s, 18-CH₃), 0.81 (3H, s, 4 β -CH₃), 0.88 (3H, s, 14-CH₃), 0.98 (3H, s, 4 α -CH₃), 0.99 (3H, s, 19-CH₃), 3.08—3.32 (1H, m, 3-H), 5.20—5.42 (2H, m, 24, 25-H).

26-Isopropyl-27-norlanosta-8,24-dien-3 β -ol (6a, b)—mp 131—132 °C. *Anal.* Calcd for $C_{32}H_{54}O$: C, 84.51; H, 11.97. Found: C, 84.19; H, 12.00. MS m/z : 454 (M^+), 439, 421. IR $\nu_{\max}^{KBr}$ cm^{-1} : 965. 1H -NMR δ (ppm): 0.69 (3H, s, 18-CH₃), 0.81 (3H, s, 4 β -CH₃), 0.87 (3H, s, 14-CH₃), 0.88 (6H, d, isopropyl-CH₃, $J=6.5$ Hz), 0.98 (3H, s, 4 α -CH₃), 0.99 (3H, s, 19-CH₃), 3.08—3.32 (1H, m, 3-H), 5.20—5.42 (2H, m, 24, 25-H).

26-n-Butyl-27-norlanosta-8,24-dien-3 β -ol (7a, b)—mp 126—127 °C. *Anal.* Calcd for $C_{33}H_{56}O$: C, 84.54; H, 12.04. Found: C, 84.33; H, 11.90. MS m/z : 468 (M^+), 453, 435. IR $\nu_{\max}^{KBr}$ cm^{-1} : 963. 1H -NMR δ (ppm): 0.69 (3H, s, 18-CH₃), 0.81 (3H, s, 4 β -CH₃), 0.88 (3H, s, 14-CH₃), 0.98 (3H, s, 4 α -CH₃), 0.99 (3H, s, 19-CH₃), 3.08—3.32 (1H, m, 3-H), 5.20—5.44 (2H, m, 24, 25-H).

26-n-Pentyl-27-norlanosta-8,24-dien-3 β -ol (8a, b)—mp 120—121 °C. *Anal.* Calcd for $C_{34}H_{58}O$: C, 84.58; H, 12.11. Found: C, 84.28; H, 12.00. MS m/z : 482 (M^+), 467, 449. IR $\nu_{\max}^{KBr}$ cm^{-1} : 963. 1H -NMR δ (ppm): 0.69 (3H, s, 18-CH₃), 0.81 (3H, s, 4 β -CH₃), 0.88 (3H, s, 14-CH₃), 0.98 (3H, s, 4 α -CH₃), 0.99 (3H, s, 19-CH₃), 3.08—3.32 (1H, m, 3-H), 5.20—5.44 (2H, m, 24, 25-H).

26-n-Hexyl-27-norlanosta-8,24-dien-3 β -ol (9a, b)—mp 118—119 °C. *Anal.* Calcd for $C_{35}H_{60}O$: C, 84.61; H, 12.17. Found: C, 84.52; H, 11.96. MS m/z : 496 (M^+), 481, 463. IR $\nu_{\max}^{KBr}$ cm^{-1} : 962. 1H -NMR δ (ppm): 0.69 (3H, s, 18-CH₃), 0.81 (3H, s, 4 β -CH₃), 0.88 (3H, s, 14-CH₃), 0.98 (3H, s, 4 α -CH₃), 3.08—3.32 (1H, m, 3-H), 5.24—5.44 (2H, m, 24, 25-H).

General Procedures for the Reduction of the Wittig Products (2a, b—9a, b)—A solution of a Wittig product (**2a, b**—**9a, b**) (80 mg) in MeOH (100 ml) was hydrogenated in the presence of 5% Pd-C (150 mg) at room temperature. After removal of the catalyst by filtration, the filtrate was concentrated under reduced pressure. Recrystallization of the residue from MeOH gave colorless needles (**10**—**17**, respectively) (yield, 80—90%).

26-Methyl-27-norlanost-8-en-3 β -ol (10)—mp 144—145 °C. *Anal.* Calcd for $C_{30}H_{52}O$: C, 84.04; H, 12.23. Found: C, 83.68; H, 12.08. MS m/z : 428 (M^+), 413, 395. 1H -NMR δ (ppm): 0.69 (3H, s, 18-CH₃), 0.81 (3H, s, 4 β -CH₃), 0.88 (3H, s, 14-CH₃), 0.98 (3H, s, 4 α -CH₃), 0.99 (3H, s, 19-CH₃), 3.06—3.30 (1H, m, 3-H).

26-Ethyl-27-norlanost-8-en-3 β -ol (11)—mp 139—140 °C. *Anal.* Calcd for $C_{31}H_{54}O$: C, 84.09; H, 12.29. Found: C, 83.75; H, 11.83. MS m/z : 442 (M^+), 427, 409. 1H -NMR δ (ppm): 0.68 (3H, s, 18-CH₃), 0.80 (3H, s, 4 β -CH₃), 0.87 (3H, s, 14-CH₃), 0.98 (3H, s, 4 α -CH₃), 0.99 (3H, s, 19-CH₃), 3.08—3.32 (1H, m, 3-H).

26,26-Dimethyl-27-norlanost-8-en-3 β -ol (12)—mp 140—141 °C. *Anal.* Calcd for $C_{31}H_{54}O$: C, 84.09; H, 12.29. Found: C, 84.11; H, 12.14. MS m/z : 442 (M^+), 427, 409. 1H -NMR δ (ppm): 0.68 (3H, s, 18-CH₃), 0.81 (3H, s, 4 β -CH₃), 0.85 (6H, d, isopropyl-CH₃, $J=6.5$ Hz), 0.88 (3H, s, 14-CH₃), 0.98 (3H, s, 4 α -CH₃), 0.99 (3H, s, 19-CH₃), 3.08—3.32 (1H, m, 3-H).

26-n-Propyl-27-norlanost-8-en-3 β -ol (13)—mp 136—137 °C. *Anal.* Calcd for $C_{32}H_{56}O$: C, 84.14; H, 12.36. Found: C, 83.79; H, 12.05. MS m/z : 456 (M^+), 441, 423. 1H -NMR δ (ppm): 0.69 (3H, s, 18-CH₃), 0.81 (3H, s, 4 β -CH₃), 0.88 (3H, s, 14-CH₃), 0.98 (3H, s, 4 α -CH₃), 0.99 (3H, s, 19-CH₃), 3.08—3.32 (1H, m, 3-H).

26-Isopropyl-27-norlanost-8-en-3 β -ol (14)—mp 137—138 °C. *Anal.* Calcd for $C_{32}H_{56}O$: C, 84.14; H, 12.36. Found: C, 83.81; H, 11.91. MS m/z : 456 (M^+), 441, 423. 1H -NMR δ (ppm): 0.69 (3H, s, 18-CH₃), 0.81 (3H, s, 4 β -CH₃), 0.85 (6H, d, isopropyl-CH₃, $J=6.5$ Hz), 0.88 (3H, s, 14-CH₃), 0.98 (3H, s, 4 α -CH₃), 0.99 (3H, s, 19-CH₃), 3.08—3.32 (1H, m, 3-H).

26-n-Butyl-27-norlanost-8-en-3 β -ol (15)—mp 131—132 °C. *Anal.* Calcd for $C_{33}H_{58}O$: C, 84.18; H, 12.42. Found: C, 83.68; H, 12.01. MS m/z : 470 (M^+), 455, 437. 1H -NMR δ (ppm): 0.69 (3H, s, 18-CH₃), 0.81 (3H, s, 4 β -CH₃), 0.88 (3H, s, 14-CH₃), 0.98 (3H, s, 4 α -CH₃), 0.99 (3H, s, 19-CH₃), 3.08—3.32 (1H, m, 3-H).

26-n-Pentyl-27-norlanost-8-en-3 β -ol (16)—mp 122—123 °C. *Anal.* Calcd for $C_{34}H_{60}O$: C, 84.23; H, 12.47.

Found: C, 83.93; H, 12.37. MS m/z : 484 (M^+), 469, 451. $^1\text{H-NMR}$ δ (ppm): 0.69 (3H, s, 18- CH_3), 0.81 (3H, s, 4 β - CH_3), 0.88 (3H, s, 14- CH_3), 0.98 (3H, s, 4 α - CH_3), 0.99 (3H, s, 19- CH_3), 3.08—3.32 (1H, m, 3-H).

26-*n*-Hexyl-27-norlanost-8-en-3 β -ol (17)—mp 121—122 °C. Anal. Calcd for $\text{C}_{35}\text{H}_{62}\text{O}$: C, 84.26; H, 12.53. Found: C, 84.28; H, 12.12. MS m/z : 498 (M^+), 483, 465. $^1\text{H-NMR}$ δ (ppm): 0.69 (3H, s, 18- CH_3), 0.81 (3H, s, 4 β - CH_3), 0.88 (3H, s, 14- CH_3), 0.98 (3H, s, 4 α - CH_3), 0.99 (3H, s, 19- CH_3), 3.08—3.32 (1H, m, 3-H).

[24- ^3H]-Lanosterol—This material was synthesized from 3 β -acetoxylanost-8-en-24-one as described previously.⁶⁾

Preparation of Rat Hepatic Subcellular Fraction and Incubation Procedure—Hepatic subcellular 10000 $\times g$ supernatant fractions (S_{10}) were prepared from Wistar male rats weighing 125—150 g as described by Gibbons *et al.*⁹⁾ The incubation mixture consisted of S_{10} fraction (4 ml, 20.0—20.6 mg protein/ml), 1 mM EDTA, 30 mM nicotinamide, 10 mM GSH, 2 mM NADP^+ , 12 mM glucose-6-phosphate, 1 unit of glucose-6-phosphate dehydrogenase, 1.3 mM NADH, 0.8 mM NAD^+ , 1.3 mM ATP, 4 mM MgCl_2 and 0.1 M potassium phosphate buffer (pH 7.4) in a total volume of 5 ml. The control incubation was started by the addition of [24- ^3H]-lanosterol (18 μM) in phosphate buffer emulsion (0.1 ml) containing Tween 80 (3 mg). In the inhibition experiments, [24- ^3H]-lanosterol (18 μM) and lanosterol analogs (40 μM each) were added at the same time in each experiment. Incubations were carried out at 37 °C for 3 h, and MeOH and KOH were added to final concentrations of 50% and 10%, respectively. The mixture was heated at 70 °C for 1 h, then extracted with CH_2Cl_2 (20 ml $\times$ 2). The CH_2Cl_2 extracts containing tritium-labeled products were washed with water, dried over sodium sulfate, and concentrated to a few milliliters. After addition of carrier lanosterol (1.0 mg) to the solution, it was subjected to silica gel thin-layer chromatography (TLC) with CH_2Cl_2 as the mobile phase and the radioactive 4,4-dimethyl sterol fraction and 4,4-demethyl sterol fraction were separated. Appropriate amounts of lanosterol were added to the eluate of the 4,4-dimethyl sterol fraction and it was recrystallized several times to constant specific activity. The 4,4-demethyl sterol fraction separated by silica gel TLC was isolated as the digitonin-precipitable sterols as described by Popják¹⁰⁾ and counted with a liquid scintillation spectrometer (Aloka LSC-502). The amount of cholesterol biosynthesis was determined from the radioactivity of the 4,4-demethyl sterol fraction (*i.e.*, cholesterol fraction).

Results and Discussion

Synthesis of Lanosterol Analogs

We have synthesized eight lanosterol analogs (10—17) with lengthened side chains (total number of carbon atoms: C_{30} — C_{35}). Wittig reactions of the aldehyde (1)⁸⁾ with the appropriate phosphoranes in benzene gave the lanosterol analogs (2a, b—9a, b) as shown in Chart 1. The structures of these products were confirmed by their $^1\text{H-NMR}$ spectra and MS. These lanosterol analogs (2a, b—9a, b) each showed one peak on gas liquid chromatography (GLC) with 1.5% OV-17. Further, their IR spectra exhibited absorption bands in the region of 955—965 cm^{-1} .

In order to determine the geometry of the 24,25-double bonds in 2a, b—9a, b, reverse-

phase HPLC was performed. As model compounds, the 24*E*-isomer and 24*Z,E*-mixture of 27-norlanosta-8,24-dien-3 β -ol 3-benzoate were analyzed by reverse-phase HPLC. The 24*Z*-isomer could be separated from the 24*E*-isomer, indicating that the 24*Z*-isomer has a slightly

TABLE I. Cholesterol Biosynthesis during Incubation of S₁₀ Fraction of Rat Liver Homogenate with [24-³H]-Lanosterol in the Presence of Various Lanosterol Analogs

Compound		Lanosterol Fr. (%)	Cholesterol Fr. (%)	Inhibition (%)
None (control)		24.9	22.1	—
10 (C ₃₀)	29.8	17.9	19
11 (C ₃₁)	28.5	20.2	9
12 (C ₃₁)	32.4	16.0	28
13 (C ₃₂)	26.9	20.4	8
14 (C ₃₂)	33.6	20.3	8
15 (C ₃₃)	27.3	19.6	12
16 (C ₃₄)	27.8	22.3	0
17 (C ₃₅)	21.6	24.7	0
18 (C ₃₀) ^{a)}	32.6	18.3	17
19 (C ₂₉) ^{a)}	75.2	5.1	77

[24-³H]-Lanosterol (90600 dpm; 0.43 μ Ci/ μ mol, 18 μ M) was incubated with rat liver S₁₀ fraction (20.0–20.6 mg protein/ml) at 37 °C for 3 h. The incubation mixture contained, in a total volume of 5 ml, 4 ml of S₁₀ fraction and cofactors. Incubation was started by the addition of the substrate and test compounds as an emulsion (0.1 ml) with Tween 80 (3 mg). Analytic methods for incubation products are described in Materials and Methods. Results are expressed as the percentage inhibition, as follows: Percent inhibition of cholesterol synthesis = [(percent yield of cholesterol isolated by TLC in control – percent yield in run with test compound)/percent yield in control] $\times 10^2$. Each incubation was carried out in triplicate and the standard deviation of each value listed was less than 5%.

a) These compounds were tested as references; the results were somewhat different from those reported⁶⁾ previously.

shorter retention time than the 24*E*-isomer. HPLC of the 3-benzoates of the lanosterol analogs (**2a**, **b**—**9a**, **b**) showed two peaks and the ratios of 24*Z* to 24*E*-isomers were approximately 3:1. As noted previously,⁸⁾ the 24*Z*-isomer of 27-norlanosta-8,24-dien-3 β -ols was isomerized to the 24*E*-isomer during 10% silver nitrate-impregnated silica gel column chromatography. The same treatment of **6a**, **b** (*Z*:*E*=76:24) yielded a *Z*,*E*-mixture (*Z*:*E*=60:40) with a slight isomerization of the 24*Z*-isomer to the 24*E*-isomer, (HPLC analysis showed an increase of the relative peak area corresponding to the 24*E*-isomer). Catalytic reduction of the Wittig products (**2a**, **b**—**9a**, **b**) afforded the corresponding 24,25-dihydro compounds (**10**—**17**) (Chart 1).

Biological Activity of Lanosterol Analogs

The effects of the lanosterol analogs on cholesterol biosynthesis from lanosterol were examined and the results are shown in Table I. Further, 24,25-dihydrolanosterol (**18**) and 27-nor-24,25-dihydrolanosterol (**19**) were tested as references.

Analog **10** and **12** showed slight inhibitory effects (19 and 28%, respectively). Analog **11**, **13**, **14**, and **15** were less inhibitory than **10** and **12**. Further, **16** and **17** showed no inhibitory effects. 27-Nor-24,25-dihydrolanosterol, which lacks the methyl group at the 27-position of the side chain of 24,25-dihydrolanosterol, was the most potent inhibitor in the series of compounds. Increase in the length of the side chain greatly reduced the inhibitory effect, as compared with that of 27-nor-24,25-dihydrolanosterol (**19**).

When the side chains were lengthened to yield the C₃₄ and C₃₅ compounds, no inhibitory effect was seen. When the side chain of 24,25-dihydrolanosterol was shortened to the C₂₂ compound, again there was no inhibitory effects, as described previously.⁶⁾ Analog **12** showed 28% inhibition but **11** showed 9% inhibition, despite having the same carbon number (C₃₁). However, **13** and **14**, having the same carbon number (C₃₂), showed the same inhibitory effects (8%). These results suggest that in the analogs with long side chains, branching at the end of the side chain was not essential for inhibitory effect.

The present results coupled with our previous ones may be summarized as follows. Among the lanosterol analogs, the 24-ethylidene-, nor-, dinor-, trinor-, tetranor-, and

Fig. 1. Relationship between Side Chain Structure and Inhibition of Cholesterol Biosynthesis.

pentanor-compounds showed inhibitory effects. In particular, 27-nor-24,25-dihydrolanosterol (**19**) showed the most potent inhibitory effect in the series of analogs. However, the hexanor-, heptanor-, and octanor-compounds, the 20-iso-compound, and the analogs with longer side chains than that of lanosterol showed only slight inhibitory effects. These results are summarized in Fig. 1. Further, cholesterol analogs with various sizes of side chains showed no inhibitory activity.¹⁾

On the other hand, in a series of oxygenated lanosterol derivatives, 7-oxo-24,25-dihydrolanosterol was the most active inhibitor (98% inhibition of cholesterol synthesis from lanosterol).⁷⁾

In the experiments in the presence of active inhibitors, recovery yields of the substrate ([24-³H]-lanosterol) increased in parallel to the extents of inhibitions. The results suggest that a potent inhibitor such as the 7-oxo-compound or 27-nor-compound may inhibit 14 α -demethylation of lanosterol, which is the first step of transformation of lanosterol to cholesterol, although the S₁₀ fraction used in this study contains many enzymes.

Aoyama *et al.*¹¹⁾ reported that lanosterol interacts with yeast cytochrome P-450 as the initial step of 14 α -demethylation, and incubation of lanosterol with a reconstituted system containing cytochrome P-450 gives the 14-demethylated product. Gaylor *et al.*¹²⁾ also reported that incubation of 24,25-dihydrolanosterol with a reconstituted system containing rat cytochrome P-450 gives the 14-demethylated product.

From our studies together with other results,^{11,12)} the enzyme involved in the initial step of the 14-demethylation is thought to be a cytochrome P-450. The substrate binding site contains at least two pockets involved in the binding of the lanosterol skeleton and its side chain. The pocket for the side chain is thought to reach the region of C-22 from the terminal area of the side chain. In the case of the hexanor-, heptanor-, and octanor-compounds, thus, no inhibitory effect is observed since their side chains are too short to interact with the binding site at the pocket. Further, no inhibitory effect is observed with the analogs having longer side chains than lanosterol, since their side chains are too long to be satisfactorily accommodated in the binding site.

On the other hand, 20-iso-24,25-dihydrolanosterol, having a different orientation at the 20-position from 24,25-dihydrolanosterol, showed no inhibitory effect. This result indicates that the side chain structure with the unnatural configuration cannot fit in the side chain pocket. In the cholesterol analogs, the inhibitory effect was not observed, suggesting that the skeletal structure is more important than the side chain structure for this activity. Among the oxygenated lanosterol derivatives studied, 7-oxo-24,25-dihydrolanosterol showed the highest inhibitory activity, and it is suggested that this effect is due to its interaction with an active center of cytochrome P-450, based on previously reported results.¹³⁾

In summary, it is suggested that the important features for an inhibitory effect of lanosterol and cholesterol derivatives on cholesterol biosynthesis from lanosterol are the side chain and skeletal structures, and the configuration at C-20.

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