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Article

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A Biomimetic Synthesis of Chaxine and its Related Compounds

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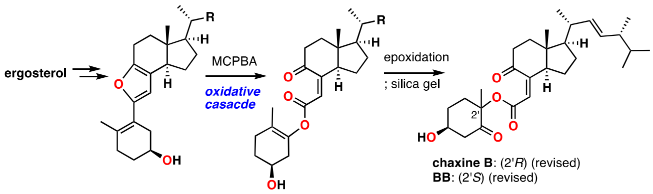
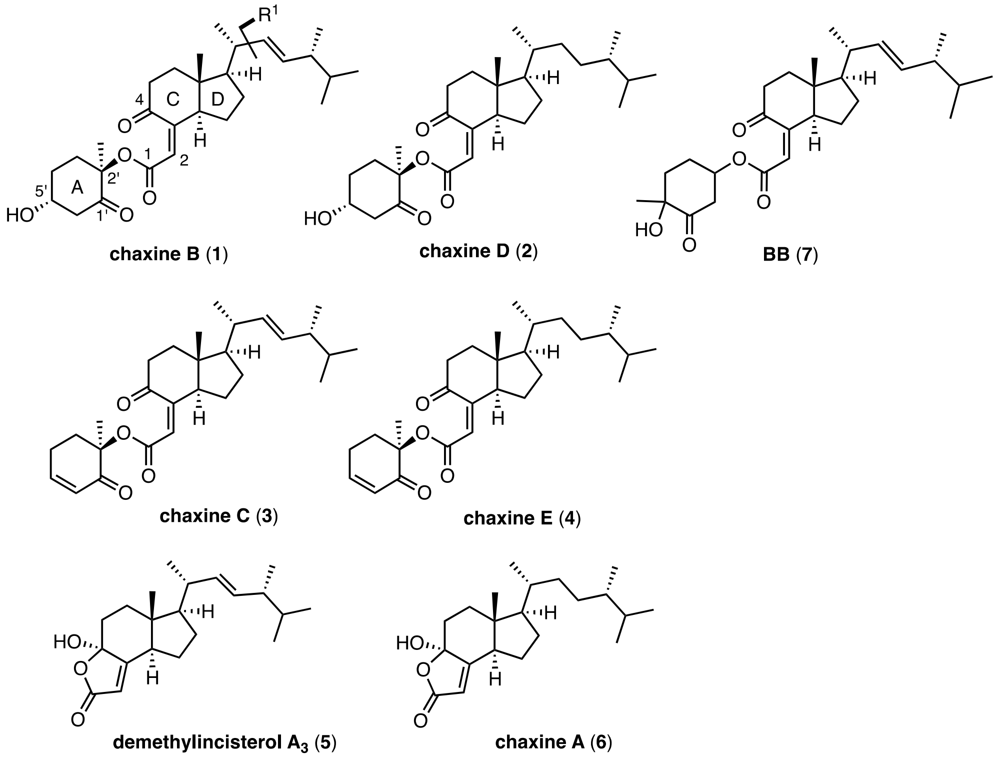


Abstract: The highly oxidized natural products chaxine B and BB have been synthesized from ergosterol in eight steps according to a route inspired by their proposed biosynthesis; key steps were an oxidative cascade from a furan intermediate to an enol ester using MCPBA, followed by diastereoselective epoxidation and acyloxy migration. This concise synthesis resulted in the revision of the structures of chaxine B and its naturally occurring analogs, and syntheses of the unnatural analogues of these natural products for biological investigations.

Keywords: chaxine, natural product, biosynthesis, electrocyclic reaction, Baeyer-Villiger oxidation, oxidative cascade

Introduction

Chaxine B (**1**) and its analogs were isolated from the Chinese edible mushroom *Agrocybe chaxingu* by Kawagishi[1,2] and are unprecedented oxidized steroidal compounds, wherein the A ring is connected to the CD ring moiety through an ester linkage, and the B ring is absent (**Figure 1**). The relative stereochemistry of the A ring was deduced by NOE experiments, but its absolute stereochemistry was not determined. Preliminary biological studies established these natural products to inhibit osteoclast formation[1,2] and induce fruiting body formation in several mushrooms,[3] but, the further details could not be clarified due to their very limited availability (only 2.1, 1.6, 0.5 and 0.4 mg of chaxines B, C, D and E respectively were isolated from 1.5 kg of the dried fruiting bodies of the mushroom).[4] Interestingly, the related compound BB (**7**) was isolated together with chaxine B (**1**) from *Penicilinim* sp. These two compounds were found to accelerate neural stem cell propagation, but the stereochemistry of the A ring moiety of **7** was not reported.[5] Detailed studies of the biological activity of BB (**7**) are also impossible, again due to its very limited availability. We were inspired to pursue total syntheses of these natural products by their interesting biological activities and unique chemical structures, to establish the absolute stereochemistry of the A ring, and support future biological investigations. We disclose herein full details of our synthetic efforts towards the chaxine family of natural products.[6]



chaxine B (**1**) **chaxine D** (**2**) **BB** (**7**)

chaxine C (**3**) **chaxine E** (**4**)

demethylincisterol A$_3$ (**5**) **chaxine A** (**6**)

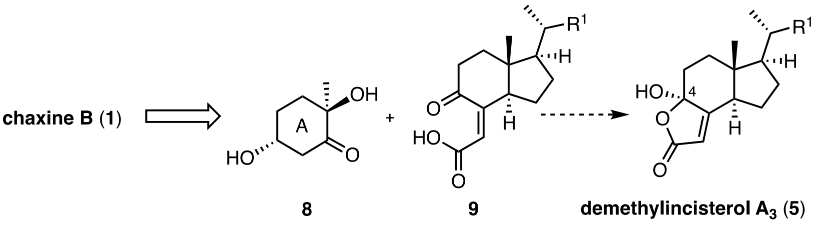
Figure 1. Initially proposed structures of chaxine B and its related natural products

Results and Discussion

Speculation of a biosynthetic route of chaxines and a synthetic plan based on this biosynthetic proposal

One possible retrosynthesis for chaxines B from demethylincisterol A$_3$ (**5**)[7] is depicted in **Scheme 1**, but was quickly discarded due to the extreme difficulties associated with esterification between a tertiary alcohol of the A ring moiety **8** and the carboxylic acid of the

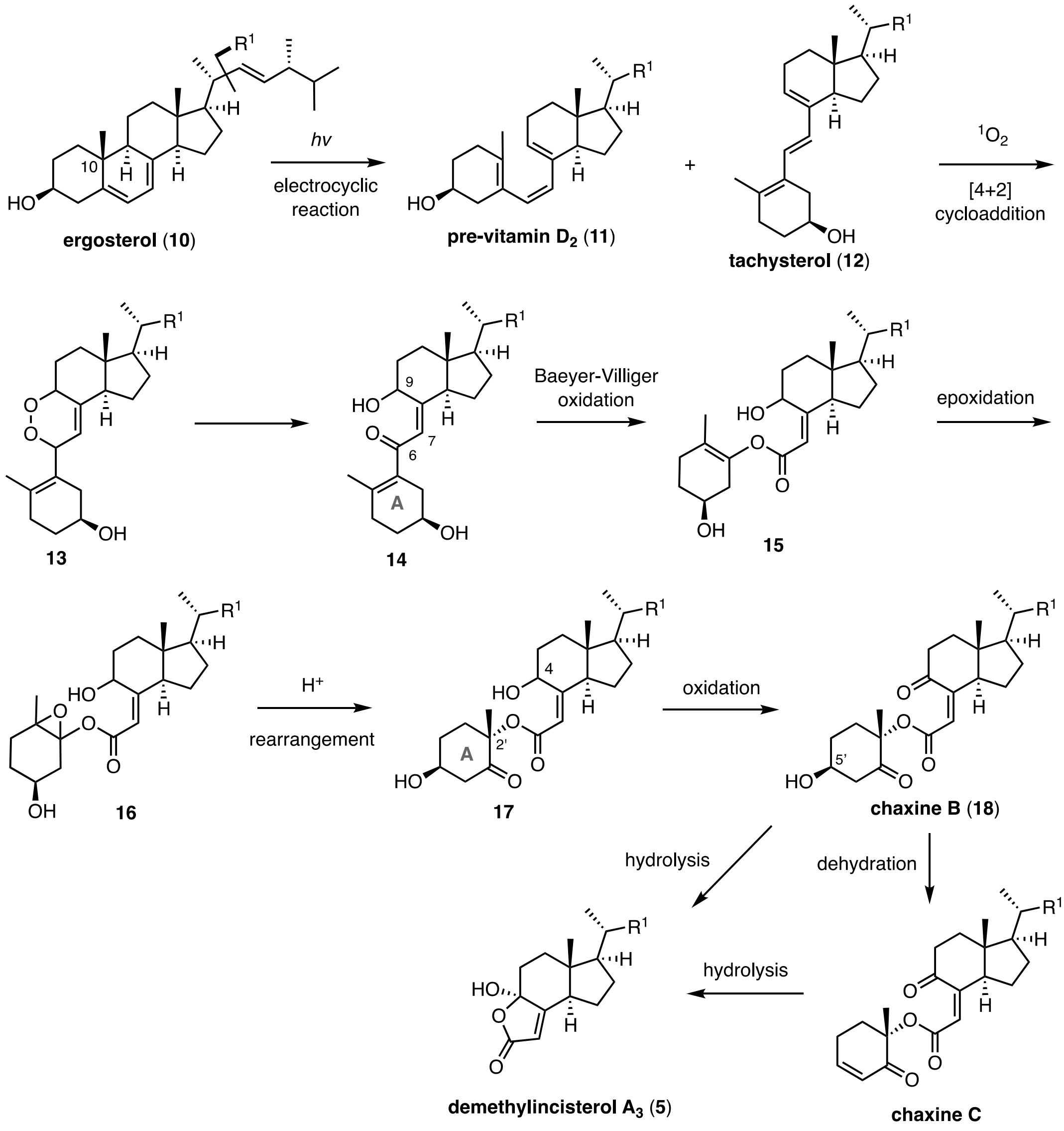
CD ring moiety **9**, considered likely to exist as hemiacetal **5** (demethylincisterol A_3).



Scheme 1. A possible retrosynthesis of chaxines B

Instead, we considered the most plausible biosynthetic route of chaxine B (**1**) as shown in **Scheme 2**.[8] Ergosterol (**10**) is a possible starting material having an identical side chain to that of the chaxine B, and is known to be abundant in many mushrooms. To construct the A ring structure of chaxine B from ergosterol (**10**), we proposed cleavage of the two C-C bonds of the B ring by an 6π-electrocyclic reaction and Baeyer-Villiger oxidation of the ketone group at the C-6 position after several oxidations. In the biosynthesis of vitamin D_2, the photoinduced electrocyclic reaction of ergosterol is known to give pre-vitamin D_2 (**11**) and/or tachysterol (**12**),[9] which might be further oxidized through endoperoxide **13** to γ-hydroxy enone **14**. The Baeyer-Villiger oxidation of **14** occurs regioselectively to give enol ester **15**,[10] which is epoxidized, and anticipated to undergo acyloxy rearrangement under acidic conditions to give α-acyloxy ketone **17**. Finally, oxidation of the hydroxy group of **17** at the C-4 position would give either chaxine B, or its 2'-epimer. Dehydration of chaxine B would give chaxine C. Hydrolysis of the esters of **1** or **3** gives demethylincisterol A_3 (**5**).[11] Since the relative stereochemistry of the A ring of chaxine B is as shown in **Figure 1**, our biosynthetic proposal suggests the absolute stereochemistry of chaxine B to be **18** as shown in **Scheme 2**. We would synthesize chaxines according to this proposed biosynthetic route.

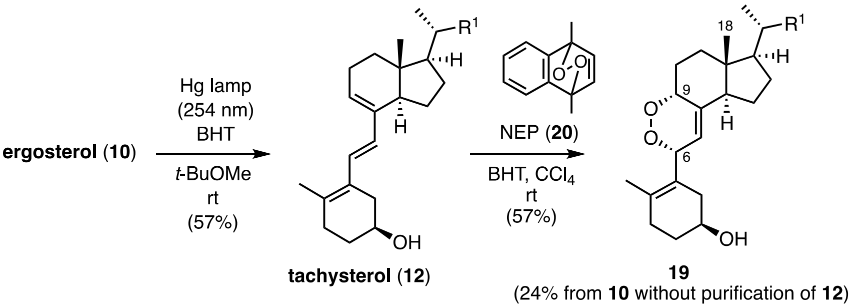
Prior to the initiation of experiments, we considered three potential issues with the biomimetic synthesis: (i) regioselectivity of the Baeyer-Villiger oxidation of **14**; (ii) diastereofacial selectivity of epoxidation of enol ester **15**; and (iii) the stereochemistry of the acyloxy migration of the enol ester epoxide **16**. In the Baeyer-Villiger oxidation of **14**, we anticipated the A ring alkene would preferentially migrate due to the higher alkyl substitution.[12] The stereochemistry of the epoxidation of enol ester **15** could be controlled by directed epoxidation of homoallylic alcohol with a metal catalyst for *syn* selective epoxidation, or by steric hindrance of protected homoallylic hydroxy group for *anti* selective epoxidation.[13] Since the acyloxy migration of a similar enol ester epoxide was reported to have occurred in a highly stereoselective manner,[14] we expected that two diastereomeric epoxides of **16** would give two different keto products, one of which would be identical to chaxine B after oxidation of hydroxy group at the C-4 position. Accordingly, determination of the absolute stereochemistry of the A ring of chaxine B and its analogs should be possible.



Scheme 2. A proposed biosynthetic pathway of chaxine B, chaxine C and demethylincisterol (**5**) from ergosterol (**10**)

Synthesis of chaxine B and BB

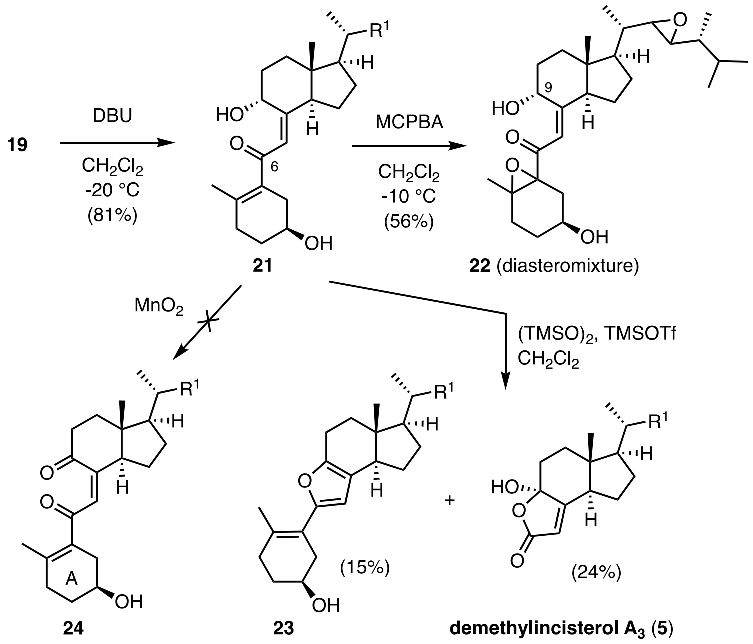
Our initial attempts to synthesize **14** from pre-vitamin D$_2$ (**11**) (easily obtainable from ergosterol (**10**)) failed due to its thermal instability. Accordingly, we sought to use tackysterol (**12**), a regioisomer of **11**, instead. However, the gram scale preparation of tachysterol **12** proved difficult. The photoinduced 6π-electrocyclic reaction of ergosterol (**10**) gave a mixture of three isomers; pre-vitamin D_2 (**11**), takysterol (**12**), and lumisterol, a epimer at C-10 of ergosterol. Irradiation of **10** using a shorter wave-length of UV (254 nm) was reported to yield tachysterol (**12**) as a major product,[15] however, tachysterol was not obtained using a photoreactor equipped with quartz glass and a low-pressure mercury lamp. After extensive experimentation, we found that the quartz glass of the reactor interfered the electrocyclic reaction. Direct irradiation of a solution of ergosterol in *tert*-BuOMe and a small amount of BHT (3,5-di-tert-butyl-4-hydroxytoluene) using a low pressure-mercury light under argon[16] gave a separable mixture (by flash column chromatography) of tachysterol (**12**) and pre-vitamin D_2 (**11**) in ca. 3:1 ratio; although the lability of tachysterol to oxygen and acids necessitated the use of neutralized silica gel, solvents containing Et_3N (3%), and nitrogen gas (**Scheme 3**). Nevertheless, we were finally able to prepare tachysterol (**12**) from ergosterol (**10**) on a gram-scale.



Scheme 3. Preparation of tachysterol (**12**) from ergosterol (**10**) and synthesis of endoperoxide **19**

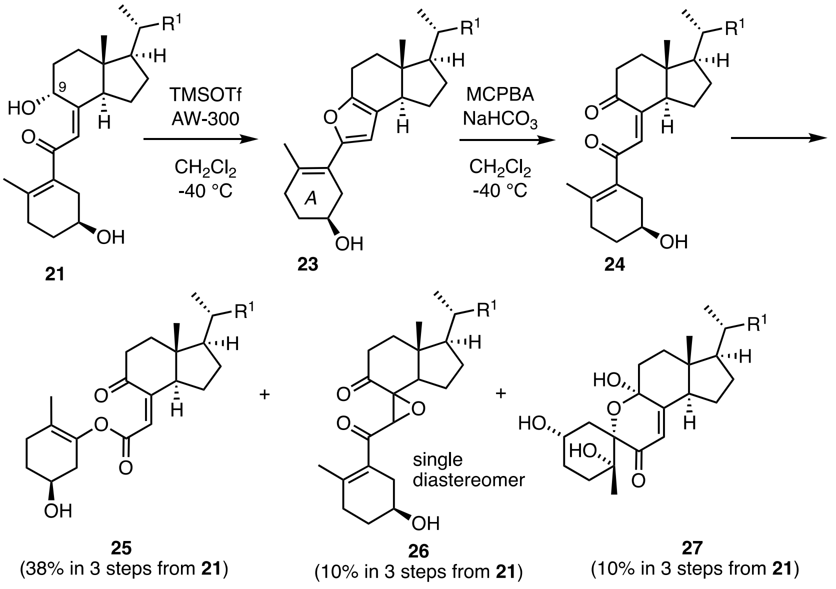
With sufficient **12** in hand, the synthesis of endoperoxide **19** from **12** using singlet oxygen could be investigated. Conventional conditions (light, oxygen gas, and the photosensitizer TPP (tetraphenylporphyrin)) resulted in a poor yield (< 30%) of **19**, presumably due to the lability of both starting material and the product to light and oxygen. Accordingly, we turned to NEP (1,4-dimethylnapthtalene endoperoxide, **20**),[17] which generates singlet oxygen quantitatively ($t_{1/2}$ = 5 h at rt), as the source of singlet oxygen. Carbon tetrachloride was selected as the solvent; the half-life of $^1O_2$ is much longer in CCl_4 than CH_2Cl_2;[18] and being non-polar, it allowed residual NEP to be easily removed by filtration through a plug of silica gel without evaporation of the solvent. A small amount of BHT was added to the reaction to prevent autoxidation, although its effect was not quantified. Under these conditions, and using 3 equivalents of NEP, **19** was obtained in 57% yield as a single diastereomer judged by ^1H NMR spectra (**Scheme 3**). The configurations of the newly generated stereocenters were determined by NOESY, a correlation being observed between the proton at C-9 and the 18-methyl group. The high diastereoselectivity is presumably due to the steric hindrance of the angular methyl group (18-CH_3). On a gram scale, **19** was obtained in a yield of 24% of two steps from **10**, where **12** was used without purification to avoid its decomposition during column chromatography.

Cleavage of the peroxy structure of **19** with DBU gave γ-hydroxy enone **21** in 81% yield (**Scheme 4**). The selective deprotonation of the proton at the C-6 position is probably due to its higher acidity than the proton at the C-9 position, being double allylic in nature. Initial attempts at the Baeyer-Villiger oxidation of **21** were less successful: treatment with MCPBA in CH_2Cl_2 resulted in epoxidation of the alkene of the A ring and the side chain unsaturation without Baeyer-Villiger oxidation. This indicates that the A ring alkene and the ketone of **21** are not fully conjugated, because steric hindrance between the methyl group and the enone precludes their coplanarity. In order to avoid the epoxidation, **21** was treated with $(TMSO)_2$ and TMSOTf, conditions reported to effect Baeyer-Villiger oxidation without alkene epoxidation,[19] to give demethylincisterol A_3 (**5**) and furan **23**. From the formation of **5**, we infer that the Baeyer-Villiger oxidation of **21** occurred with the desired selectivity to give **15** (in **Scheme 2**). However, hydrolysis of the ester **15** followed by oxidation of the C-9 position gave **5** under the reaction conditions – perhaps by lactonization of product **15** of the Baeyer-Villiger reaction, hydrolysis of the lactone and then oxidation of the hydroxy group. In order to prevent this, we could have attempted Baeyer-Villiger oxidation of 2-ene-1,4-dione **24** (obtainable from **21**), although the prediction of the site and regioselectivity of the Baeyer-Villiger oxidation of **24** would be difficult[12] as it incorporates two ketones conjugated through the same alkene, and therefore there are four possible Baeyer-Villiger products. To the best of our knowledge, the Baeyer-Villiger oxidation of such a complex substrate has not been reported.[12] Although the regioselectivity of the Baeyer-Villiger oxidation depends on many factors such as electron density, steric of the substituent and stereoelectronic effect, we optimistically anticipated the Baeyer-Villiger oxidation of **24** would take place preferentially at the ketone at C-6 position, and that the A ring alkene would preferentially migrate due to its increased substitution; as the rate-determining step of the Baeyer-Villiger oxidation is migration after addition of peroxide to a ketone,[12] the migratory aptitude of the substituents of the ketone determines the selectivity – and of the four substituents of the two ketones of **24**, the A ring alkene is tetrasubstituted with three alkyl groups, while the alkene between the two ketones is electron deficient.

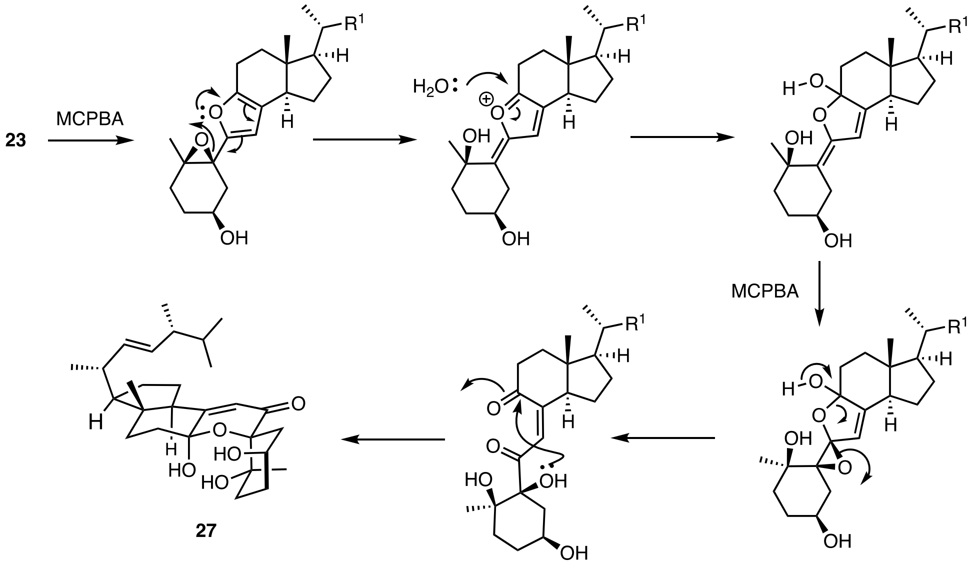


Scheme 4. Synthesis of **21** and attempted Baeyer-Villiger oxidation of **21**

However, preparation of **24** from **21** also proved problematic, even though production of **24** was observed when **21** was kept in a freezer for several months. Oxidation of **21** with MnO_2 gave a complex mixture of products, and the attempted protection of the hydroxy group of the A ring with TBSOTf and 2,6-lutidine in anticipation of the selective oxidation of the C-9 hydroxy group resulted in furan **23** (Paal-Knorr furan synthesis). At this point, we realized that 2-ene-1,4-dione **24**, the desired substrate of the Baeyer-Villiger oxidation, could be prepared by oxidation of furan **23**. Synthesis of furan **23** from **21** was best carried out by with TMSOTf in the presence of molecular sieves AW-300 at – 40 °C to give **23**, an unstable product that was used without purification (**Scheme 5**). Fortunately, upon treatment of **23** with MCPBA, we observed production of desired **24** as well as enol ester **25**, the desired product of the Baeyer-Villiger oxidation of **24**. Thus, the consecutive oxidation of furan **23** to give **24** and then **25** could be carried out in one-pot using MCPBA. Optimized conditions for this sequence were found to be 2.1 equivalent of MCPBA in CH_2Cl_2 at -40 °C, giving **25** in 38% yield in three steps from **21**. Two byproducts, epoxide **26** and spiro-product **27**, were also obtained, the both yields are 10%. and their structures were elucidated by NMR and X-ray analyses[20] (For details see the Supporting Information). Epoxide **26** is hypothesized to arise from **24** by MCPBA. A possible but unvalidated mechanism for the formation of spiro compound **27** is depicted in **Scheme 6**, based on the observation that **27** was not formed by treating purified samples of **24** with MCPBA.



Scheme 5. Synthesis of enol ester **25** by oxidative cascade of furan **23**

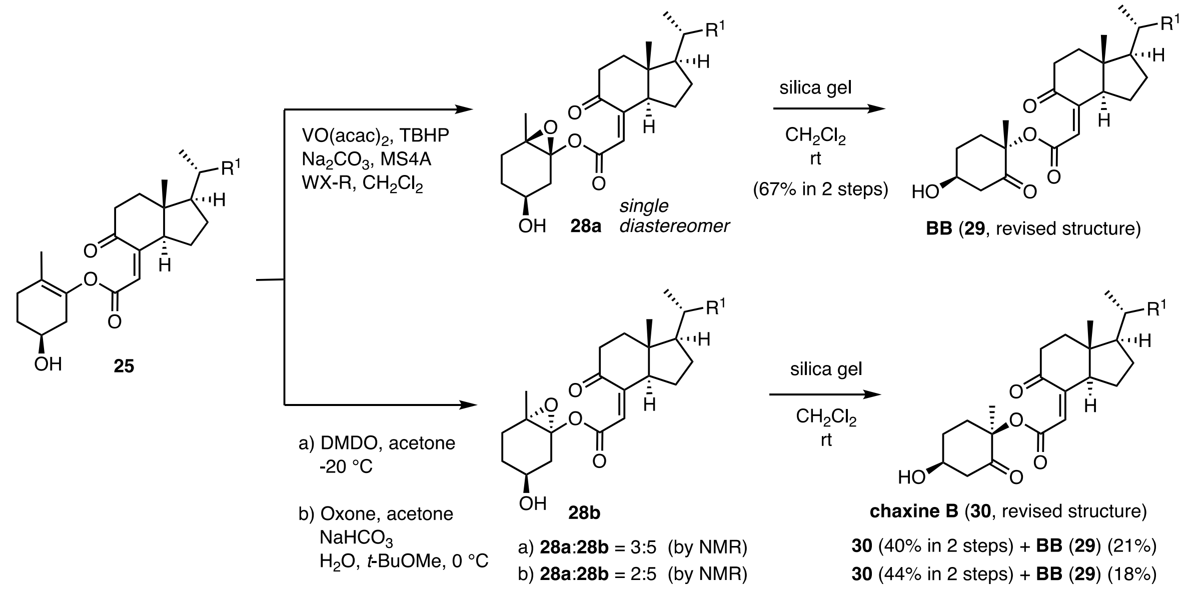


Scheme 6. A proposed mechanism for **27** from **23**

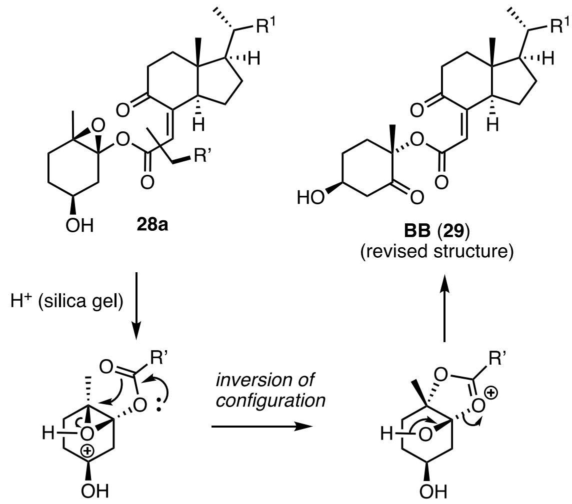
We then turned our attention to stereoselective epoxidation of enol ester **25** (**Scheme 7**). Epoxidation of **15** with MCPBA was first attempted with view to a development of an oxidative cascade from furan **23**, but an inseparable mixture of the products was obtained due to competitive epoxidation of the side chain. However, the selective, directed epoxidation of the A ring alkene was accomplished using TBHP and catalytic VO(acac)$_2$[21] in the presence of Na_2CO_3, MS4A and WX-R (4,4'-thiobis-(6-*tert*-butyl-3-methylphenol)).[22] to give epoxide **28a** as a single diastereomeric product, which was exposed to silica gel without any purification to give ester **29** as a single product in 67% yield over two steps from **25**. The NMR spectra of **29** was identical to that of BB, the stereochemistry of the A ring of which has not been reported. X-ray crystallographic analysis of **29** clarified its structure and definitively established it to incorporate a different ester linkage to that of **7** (in **Figure 1**). Since the relative stereochemistry of the A ring of BB (**29**) is the same as that of the proposed structure of chaxine B, the relative stereochemistry of the A ring of chaxine B (**1**) must be revised.

The highly stereoselective conversion of β-epoxide **28a** to BB (**29**) is rationalized by the mechanism shown in **Scheme 8**, and is consistent with a literature report of the stereochemical outcome of another, similar reaction.[23] We therefore hypothesized that chaxine B should be accessible by the action of silica gel on α-epoxide **28b**, a synthesis of which (from **25**) was now sought. Extensive exploration of the conditions for this reaction led us to conclude that DMDO (dimethyldioxirane) was the best oxidant; epoxidation with DMDO in acetone gave a mixture of **28a** and **28b** in ca. 3:5 ratio (by ^{1}H-NMR spectra). Exposure of this mixture to silica gel provided BB (**29**) and **30** in yields of 21 and 40% over two steps from **25**, respectively. As the NMR data of **30** was identical to those of natural chaxine B, the structure of chaxine B was revised as **30**, an epimer at the C-2' position of BB (**29**). Since chaxine B is noncrystalline, the structure of **30** was confirmed by spectroscopic analysis of the products of its reduction with $NaBH_4$[24] and hydrolysis products (vide infra). Thus, an eight-step synthesis of chaxine B and BB from ergosterol was accomplished.

To scale up the synthesis of chaxine B (**30**) and BB (**29**) (from **25**) for their biological evaluation, the epoxidation of **25** with DMDO generated *in situ*-from Oxone® and acetone was investigated in biphasic solvent of aqueous $NaHCO_3$ and several organic solvents.[25] Interestingly, the choice of organic solvent was found to influence the stereoselectivity of the epoxidation; the stereoselectivity (α/β) was 2.5:1 in *t*-BuOMe, 2.0:1 in AcOEt, 0.8:1 in benzene, and 0.6:1 in CH_2Cl_2, by analysis of the ^{1}H-NMR spectra. This observation is thought to be the result of hydrogen bonding between polar solvents such as *t*-BuOMe, acetone, and AcOEt and the allylic hydroxy group of the substrate which shields the β-face, resulting in preferential α-epoxidation.[26] On the other hand, nonpolar solvents such as CH_2Cl_2 and benzene do not significantly interact with the hydroxy group, and therefore epoxidation proceeds by hydrogen bonding between DMDO and the homoallylic alcohol to give β-epoxide preferentially. Thus, 576 mg of chaxine B (**30**) was obtained by the DMDO epoxidation of **25** in aqueous *t*-BuOMe followed by treatment of silica gel.



Scheme 7. Synthesis of chaxine B and BB

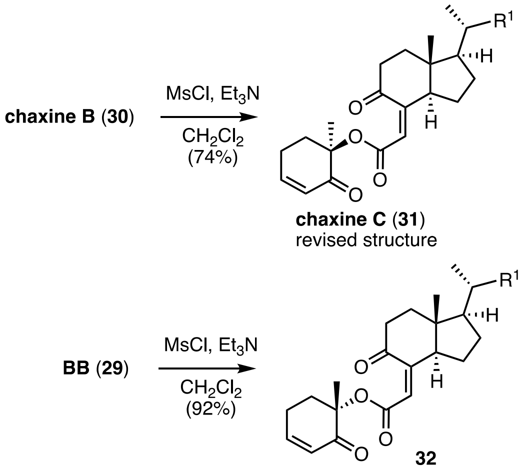


Scheme 8. A proposed mechanism for rearrangement of **28a** to **BB** (**29**)

Synthesis of analogs of chaxine B and BB

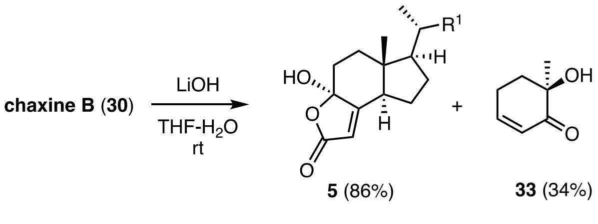
Several analogues of chaxine B and BB were also prepared (**Scheme 9**). Dehydration of synthetic chaxine B (**30**) and BB (**29**) with MsCl/Et$_3$N gave **31** and **32**, respectively.

Comparison of the NMR spectra of **31** and **32** with those of chaxine C led to the conclusion that **31** was chaxine C.[24]



Scheme 9. Synthesis of chaxine C (**31**) and its epimer **32**

Demethylincisterol A_3 (**5**) was obtained by hydrolysis of synthetic chaxine B (**30**) and BB (**29**). Alkaline hydrolysis of chaxine B (**30**) with aqueous lithium hydroxide gave demethylincisterol A_3 (**5**) and cyclohexenone **33** as a volatile compound[27] (**Scheme 10**).[28] Acid hydrolysis with *p*-TsOH in aqueous THF of chaxine B (**30**) gave a mixture of **5** and **34**; and of BB (**29**) gave **5** and **35** (**Scheme 11**). NMR analyses of **34** and **35** also confirmed the stereostructures of the A rings of chaxine B (**30**) and BB (**29**), both of which bear a methyl group at the equatorial position. Interestingly, the conformations of **34** and **35** are the opposite of those of the corresponding ester BB (**29**) and chaxine B (**30**).



Scheme 10. Alkaline hydrolysis of chaxine B (**30**)

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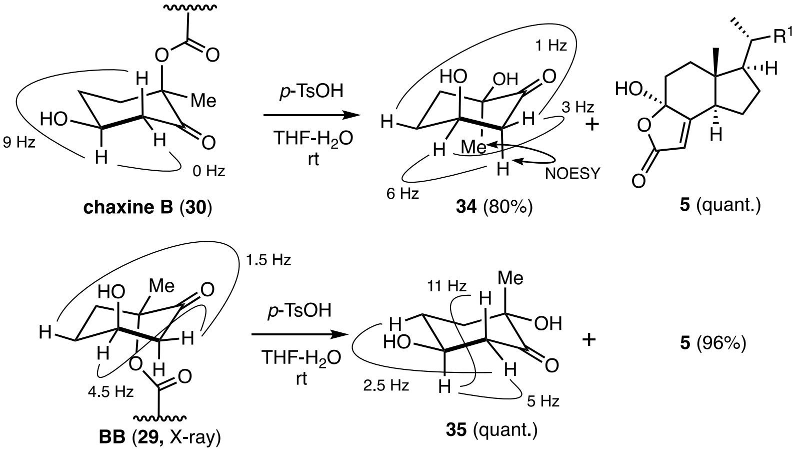

Scheme 11. Acid hydrolysis of chaxine B (**30**) and BB (**29**) and conformations of the A ring moieties

To undertake a SAR study of the side-chains of chaxine B and BB, 17-nor-chaxine D (**46**) and its 2’-epimer **45** were synthesized from 7-dehydrocholesterol (**36**) in a similar manner to that of chaxine B (**Scheme 12**; unoptimized conditions). Since the side chain of enol ester **42** does not contain an alkene in the side chain, we anticipated that the oxidative cascade from furan **41** could be expanded to effect epoxidation of enol ester **42**. Unfortunately, treatment of **41** with an excess amount of MCPBA resulted in an inseparable mixture of two diastereomers **45** and **46** along with two byproducts **43** and **44** after treatment of silica gel. Instead, we synthesized 17-nor-chaxines D (**46**) and its epimer **45** by DMDO oxidation of **42**, which was prepared from furan **40** in a similar manner to chaxine B and BB. Dehydration of **46** with MsCl and triethylamine gave 17-nor-chaxine E (**47**) in 86% yield. Alkaline hydrolysis of **46** with aqueous LiOH gave 17-nor-chaxine A (**48**).

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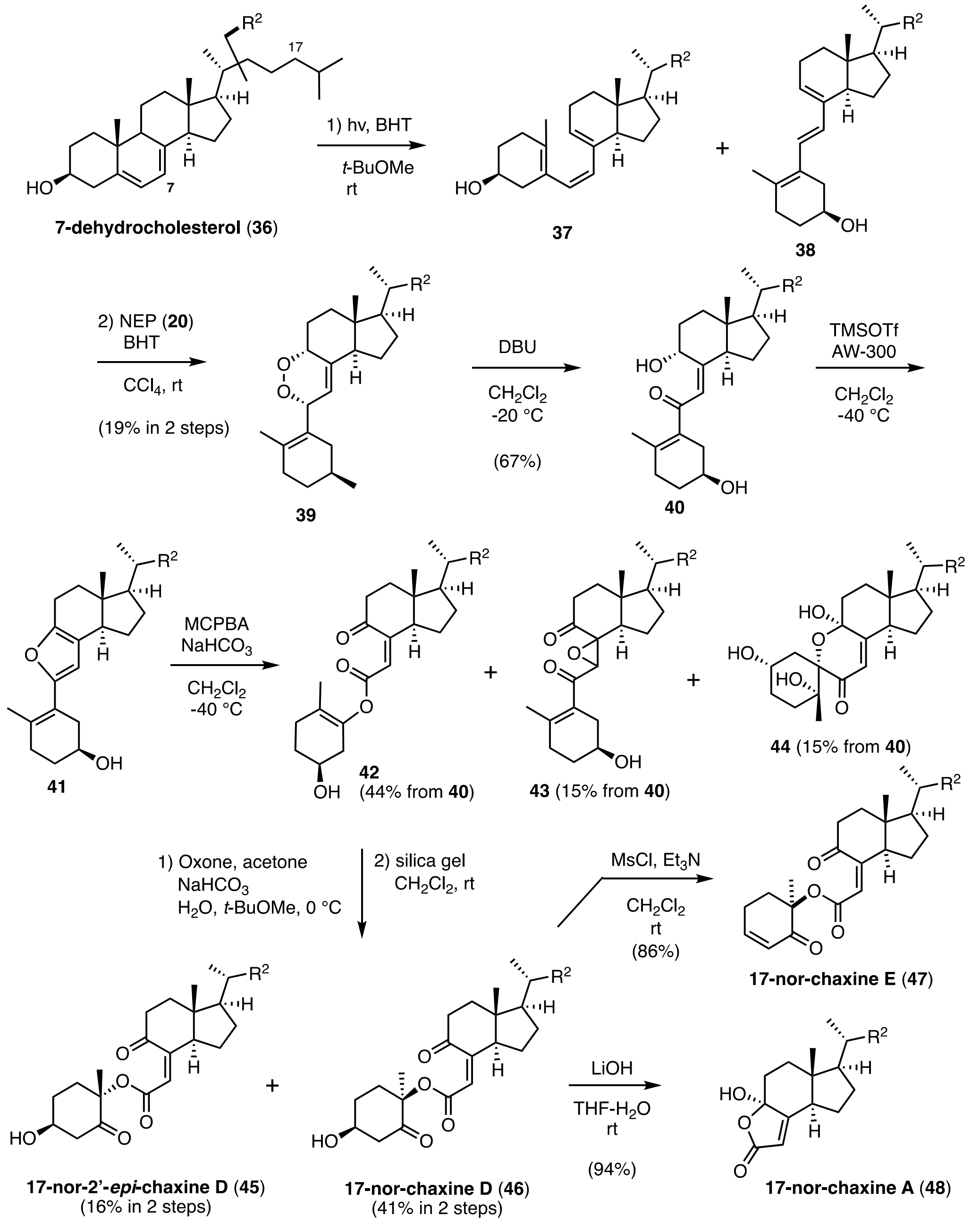

Scheme 12. Synthesis of 17-nor-chaxine D (**46**) and its analogs from 7-dehydrocholesterol (**36**) (The conditions were not optimized.)

Conclusion

The total syntheses of chaxine B and BB have been accomplished in eight steps starting from ergosterol and proceeding through a common intermediate in a manner inspired by their proposed biosynthesis. The key step is an oxidative cascade using MCPBA, which includes oxidation of the furan **23** followed by Bayer-Villiger oxidation of the resulting enedione **24**. The enol ester of the cascade product **25** was the common intermediate; its α-epoxidation gave chaxine B, and β-epoxidation gave BB after stereoselective rearrangement of the epoxide with silica gel. We believe that ergosterol is the precursor of the chaxines and the Baeyer-Villiger oxidation of enedione **24** and the epoxidation of **25** closely resemble the biosynthesis of these natural products. These two key reactions of the proposed biosynthesis are partially revised from those reactions shown in Scheme 2 during this investigation. This study also resulted in the revision of the stereostructures of the chaxines and BB; chaxine B and BB are 2'-epimers. Thus, stereostructures of the A ring of chaxines D and E should be identical to those of chaxines B and C, respectively. The scalable and concise nature of our synthesis enabled the synthesis of natural and unnatural analogs of chaxine B; biological studies of these products are ongoing, and results will be reported in due course.[29]

Experimental

General Experimental: Melting points (MP) were recorded on a Yanaco MP-S3 melting point apparatus and are not corrected. Optical rotations were measured on a JASCO DIP-370 digital polarimeter. Infrared spectra (IR) were recorded on a JASCO FT/IR-4100 spectrophotometer and are reported in wave number (cm^{-1}). Proton nuclear magnetic resonance (^{1}H NMR) spectra were recorded on a Bruker AVANCE-400 (400 MHz) spectrometer. NMR samples were dissolved in $CDCl_3$ or C_6D_6, and chemical shifts are reported in ppm relative to the residual undeuterated solvent ($CDCl_3$ as $\delta = 7.26$ ppm, C_6D_6 as $\delta = 7.16$ ppm). ^{1}H NMR data are reported as follows; chemical shift, integration, multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, br = broadened, m = multiplet), coupling constant, and assignment. Carbon nuclear magnetic resonance ($^{13}C\{^1H\}$ NMR) spectra were recorded on a Bruker AVANCE-400 (100 MHz) spectrometer. The samples were dissolved in $CDCl_3$ or C_6D_6, and chemical shifts are reported in ppm relative to the deuterated solvent ($CDCl_3$ as $\delta = 77.1$ ppm, C_6D_6 as $\delta = 128.0$ ppm). ^{1}H NMR and $^{13}C\{^1H\}$ NMR spectra were measured at 300 K unless otherwise noted. Elemental analyses were performed at the Analytical Laboratory of Graduate School of Bioagricultural Sciences, Nagoya University. High resolution mass spectra (HRMS) were recorded on an Applied Agilent 6220 Time of Flight (TOF) or an LTQ Orbitrap mass spectrometer (Thermo Fisher Scientific) equipped with an electro spray ionization probe and are reported in *m/z*. Reactions were monitored by thin-layer chromatography (TLC) on 0.25 mm silica gel coated glass plate 60 F_{254} (Merck, #1.05715). Silica gel 60 (spherical, particle size 40-50 μm, Kanto Chemical Co., Inc.) was used for flash-column chromatography. Unless otherwise noted, non-aqueous reactions were carried out in flame-dried glassware under nitrogen or argon. Dry CH_2Cl_2, THF, and Et_2O were purchased from Kanto Chemical Co., Inc. Et_3N and DBU were distilled from CaH_2. All other commercially available reagents were used as received. Activated AW-300, MS4A and dry silica gel (Merck, #1.07734.9025, 0.063-0.200 nm) were prepared by heating at 250 °C *in vacuo* with a Kugelrohr apparatus for 3 h.

Tachysterol (12): A flat dish (ф 18 cm, depth 3.6 cm, polypropylene) charged with ergosterol (**10**) (3.08 g, 7.77 mmol), BHT (168 mg, 0.766 mmol) and a magnetic stirrer bar, was placed on a magnetic stirrer. t-BuOMe (400 mL) was added, and three mercury lamps (GL 15, 15 W, 43.6 cm x ф 2.6 cm, Toshiba Corp. Ltd.) were placed on the flat dish. The whole apparatus was wrapped with a vinyl bag, into which N_2 gas was flowed. The flat dish was irradiated at room temperature with stirring. *t*-BuOMe (ca. 50 mL) was added at 30 min intervals to maintain the solvent volume. After 5 h, the solution was concentrated *in vacuo* at ice-bath temperature. The residue was purified by flash column chromatography (neutral silica gel 90 g, 3% Et_3N in Et_2O/hexane 5:9) to give tachysterol (**12**) (1.76 g, 57%) as a white amorphous solid.

Endoperoxide 19: A three-necked flask connected to a three-way stopcock was charged with tachysterol (**12**) (2.28 g, 5.75 mmol) and BHT (670 mg, 3.05 mmol), and then filled with argon gas. CCl_4 (180 mL) was degassed by freeze-thaw cycles (x3) in a separate flask and transferred to the reaction vessel with a syringe. 1,4-Dimethylnapthalene endoperoxide (NEP) (**20**) (2.14 g, 11.4 mmol) was added, the reaction vessel was wrapped with aluminum foil. After stirring at room temperature under argon atmosphere for 21 h, NEP (**20**) (1.05 g, 5.59 mmol) was added. Stirring was continued for 17 h, and then the solution was directly subjected to flash column chromatography (neutral silica gel 80 g, CH_2Cl_2 to AcOEt/hexane 1:3) to give **19** (1.41 g, 57%) as a white amorphous solid.

Synthesis of endoperoxide 19 from 10 without purification of 19: Two flat dishes (ф 18 cm, depth 3.6 cm, polypropylene) were each charged with ergosterol (**10**) (3.88 g, 9.80 mmol and 3.66 g, 9.24 mmol), BHT (210 mg, 0.953 mmol and 171 mg, 0.776 mmol) and a magnetic stirrer bar. *t*-BuOMe (450 mL) was added, and three mercury lamps were placed on the flat dish. The whole apparatus was wrapped with a vinyl bag, and N_2 gas was flowed into the bag. The flat dishes were irradiated with three lamps at room temperature for 5.5 h, with stirring. During this reaction, *t*-BuOMe (ca. 50 mL) was added at 30 min intervals to maintain the solvent volume. The solutions of the dishes were combined and evaporated at an ice-bath temperature. The residue was used for next reaction without purification. A three-necked flask connected to a three-way stopcock was charged with the crude product (7.55 g) and BHT (1.38 g, 6.26 mmol) and then filled with argon gas. CCl_4 (400 mL) was degassed by freeze-thaw cycles (x3) in a separate flask and transferred to the vessel with a syringe. NEP (**20**) (4.74 g, 25.2 mmol) was added, the reaction vessel was wrapped with aluminum foil. After stirring at room temperature under argon atmosphere for 14 h, NEP (**20**) (1.16 g, 6.17 mmol) was added. Stirring was continued for further 23 h, the solution was directly subjected to flash column chromatography (neutral silica gel 210 g, CH_2Cl_2 to AcOEt/hexane 1:5) to give **19** (1.92 g, 24% in 2 steps) as a white amorphous solid.

1,4-Dimethylnaphthalene endoperoxide (20): A flask (1 L) was charged with 1,4-dimethyl naphthalene (9.64 g, 61.7 mmol), methylene blue (44.7 mg, 0.139 mmol) and CH_2Cl_2 (1000 mL). LED light (10 W × 100, a irradiation device consisted of 100 LED lights (70 pcs: red, 20 pcs: blue, 8 pcs: white, 1 pcs: UV, 1 pcs: IR)) was placed next to the flask as close as possible. Irradiation was continued at 0 °C for 5 h with O_2 bubbling. During the reaction, CH_2Cl_2 was added at 30 min interval to maintain the solvent volume. The solution was evaporated at 0 °C to ca. 10 mL of the solvent, which was filtered through a glass filter packed with silica gel (20 g) under vacuum to remove methylene blue. The filtrate (250-300 mL) was concentrated to dryness *in vacuo* to provide orange crystals, which were washed with pentane to give NEP (**20**) (6.02 g, 32.0 mmol, 52%) as white crystals. The filtrate was concentrated at 0 °C, the residue was crystallized in the same way as already described to give NEP (**20**) (1.03 g, 5.48 mmol, 9%). The mother liqueur was recovered after decolorization with activated charcoal and evaporated to give starting material, 1,4-dimethyl naphthalene (3.38 g, 90% recovery yield).

Unsaturated ketone 21: DBU (4.70 mL, 31.9 mmol) was added to a stirred solution of **19** (3.03 g, 7.08 mmol) in dry CH_2Cl_2 (80 mL) at -20 °C. After stirring at the same temperature for 24 h, the reaction was quenched with saturated aqueous NH_4Cl (50 mL) and H_2O, and the mixture was allowed to warm to room temperature. After partitioning, the aqueous layer was extracted with CH_2Cl_2 (15 mL x3), and the combined organic layer was washed with H_2O (x1) and brine (x1). The solution was passed through a short column packed with anhydrous Na_2SO_4 and concentrated to dryness *in vacuo*. The residue was purified by flash column chromatography (silica gel 120 g, Et_2O/hexane 5:1 to Et_2O) to give **21** (2.44 g, 81%) as a white amorphous solid.

Diepoxide 22: To a stirred solution of **21** (16.2 mg, 0.0379 mmol) in CH_2Cl_2 (1.5 mL) was added MCPBA (27.4 mg, 0.159 mmol) at -10 °C. After stirring at the same temperature for 17 h, the reaction was quenched with saturated aqueous Na_2SO_3 (2 mL). The mixture was extracted with CH_2Cl_2 (x3). The combined organic layer was washed with saturated aqueous $NaHCO_3$ (x2) and brine (x1), dried over anhydrous Na_2SO_4, and evaporated under reduced pressure. The residue was purified by column chromatography (AcOEt/hexane 3:1) to give **22** (9.7 mg, 56%) as a white amorphous solid. **22**: $[\alpha]_D^{25}$ -47 (c 0.49, C_6H_6). IR (KBr) ν_{max} 3420, 2961, 2874, 1673, 1615, 1461, 1375, 1203, 1015, 970, 922, 903 cm^{-1}. HRMS (ESI) for $C_{28}H_{44}O_5Na$

[M+Na]$^+$, calcd 483.3081, found: 483.3082. ^{1}H and ^{13}C{^{1}H} NMR spectra (see the pdf of SI.) show disappearance of the side chain unsaturation, remaining of the unsaturated ketone, and four diastereomeric mixture.

Synthesis of 23 and demethylincisterol A$_3$ (5) from 21: TMSOTf (23 μL, 0.13 mmol) and (TMSO)$_2$ (0.21 mL, 0.48 M in CH_2Cl_2, dried over AW-300) were added to a stirred solution of **21** (21.5 mg, 0.050 mmol) in CH_2Cl_2 (1.0 mL) at -40 °C. After stirring at the same temperature for 2 h, additional (TMSO)$_2$ (0.21 mL, 0.48 M in CH_2Cl_2, dried over AW-300) was added. After stirring for 80 min, the reaction was quenched with Na_2SO_3 and the mixture extracted with EtOAc (x4). The combined organic layer was washed with brine (x1), dried over anhydrous Na_2SO_4, and evaporated under reduced pressure. The residue was purified by column chromatography (Et_2O/hexane 1:2 to 1:1) to give **23** (3.2 mg, 15%) and **5** (4.0 mg, 24%).

Synthesis of enol ester 25, 26 and 27 from 21: A suspension of **21** (2.18 g, 5.05 mmol) and activated AW-300 (1.08 g) in dry CH_2Cl_2 (50 mL) was stirred under argon atmosphere at -40 °C for 30 min. TMSOTf (2.20 mL, 12.6 mmol) was added at the same temperature, and after 20 min the reaction was quenched with saturated aqueous $NaHCO_3$ (20 mL). The mixture was diluted with H_2O and allowed to warm to room temperature. To remove AW-300, the mixture was filtered through a cotton plug. After partitioning, the aqueous layer was extracted with CH_2Cl_2 (15 mL x3). The combined organic layer was washed with H_2O (x1) and brine (x 1), and filtered through a short column packed with anhydrous Na_2SO_4. The solvent was evaporated to dryness *in vacuo,* and the residue was used for the next reaction without purification. MCPBA (1.67 g, 10.1 mmol) was added to a solution of this crude product (1.96 g) in dry CH_2Cl_2 (50 mL) at -40 °C under an argon atmosphere. After stirring for 40 min, the reaction was quenched with saturated aqueous Na_2SO_3 (15 mL) at -40 °C. H_2O was added and the mixture was allowed to warm to room temperature with stirring. After partitioning, the aqueous layer was extracted with CH_2Cl_2 (15 mL x3). The combined organic layer was washed with saturated aqueous $NaHCO_3$ (x1) and brine (x1), and passed through a short column packed with anhydrous Na_2SO_4. The solvent was evaporated to dryness *in vacuo*. The residue was purified by flash column chromatography (neutral silica gel 90 g, AcOEt/hexane 2:3) to give **25** (857 mg, 38%) as a yellow oil, **26** (230 mg, 10%) as a yellow solid, and **27** (289 mg, 10%) as a white solid. **26**: Mp 135-138 °C. $[\alpha]_D^{27}$ -14.8 (c 0.41, $CHCl_3$). IR (KBr) ν_{max} 3550, 2952, 2872, 1712, 1651, 1459, 1419, 1368, 1290, 1265, 1237, 966, 741, 707 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.26 (1H, dd, J = 15, 7.5 Hz, H-23), 5.15 (1H, dd, J = 15, 8.5 Hz, H-22), 4.03 (1H, m, H-4), 3.90 (1H, s, H-7), 2.86 (1H, brd, J = 16 Hz), 2.56-2.47 (2H, m), 2.41 (1H, dt, J =

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19, 6 Hz), 2.32 (1H, dd, $J = 12$, 7.5 Hz), 2.25-2.07 (4H, m), 2.04-1.96 (1H, m), 1.91 (3H, s, H-19), 1.89-1.77 (3H, m), 1.75-1.61 (2H, m), 1.57-1.36 (4H, m), 1.31- 1.20 (1H, m), 1.15 (3H, s, CH_3-18), 1.04 (3H, d, $J = 7$ Hz, CH_3-21), 0.91 (3H, d, $J = 7$ Hz, CH_3-28), 0.83 (3H, d, $J = 7$ Hz, CH_3-26 or 27), 0.81 (3H, d, $J = 7$ Hz, CH_3-26 or 27). $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$) δ 205.4, 199.6, 147.2, 134.8, 133.0, 128.5, 69.1, 66.0, 62.4, 55.7, 52.8, 45.5, 43.0, 39.9, 39.0, 37.2, 33.7, 33.2, 31.4, 29.8, 28.8, 21.7, 21.2, 20.3, 20.1, 19.8, 17.7, 12.2. HRMS (ESI) for $C_{28}H_{42}O_4Na$ $[M+Na]^+$, calcd 465.2975, found: 465.2954. **27**: Mp 182-190 °C. $[\alpha]_D^{26}$ -3.8 (c 0.52, $CHCl_3$). IR (KBr) ν_{max} 3359, 2956, 2870, 2076, 1666, 1456, 1141, 1104, 1028, 967, 827, 720, 709 cm^{-1}. ^1H NMR (400 MHz, $CDCl_3$) δ 6.51 (1H, br, O*H*), 5.78 (1H, d, $J = 2$ Hz, H-7), 5.23 (1H, dd, $J = 15$, 7.5 Hz, H-23), 5.14 (1H, dd, $J = 15$, 8 Hz, H-22), 4.35 (1H, brd, $J = 9$ Hz, O*H*), 3.98 (1H, br, H-4), 3.76 (1H, br, O*H*), 2.70 (1H, brt, $J = 9$ Hz, H-14), 2.47 (1H, td, $J = 13$, 4 Hz, H-11), 2.26 (1H, dd, $J = 15$, 3.5 Hz), 2.14-1.77 (8H, m), 1.74-1.63 (2H, m), 1.51-1.33 (6H, m), 1.26 (3H, s, CH_3-19), 1.01 (3H, d, $J = 7$ Hz, CH_3-21), 0.91 (3H, d, $J = 7$ Hz, CH_3-28), 0.83 (3H, d, $J = 7$ Hz, H-26 or 27), 0.81 (3H, d, $J = 7$ Hz, H-26 or 27), 0.65 (3H, s, CH_3-18). $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$) δ 195.0, 162.9, 135.0, 132.8, 120.5, 93.6, 88.2, 74.6, 65.9, 55.9, 52.8, 48.1, 43.0, 40.4, 39.2, 38.4, 36.1, 33.2, 31.6, 30.8, 28.7, 24.1, 21.1, 20.9, 20.1, 19.8, 17.7, 12.1. HRMS (ESI) for $C_{28}H_{44}O_5Na$ $[M+Na]^+$, calcd 483.3061, found: 483.3081.

BB (29): A flask was charged with enol ester **25** (92.6 mg, 0.209 mmol), dried MS4A (101 mg), WX-R (3.8 mg, 10 μmol), powdered Na_2CO_3 (133.7 mg, 1.26 mmol) and $VO(acac)_2$ (10.8 mg, 42 μmol), then filled with argon gas. Dry CH_2Cl_2 (4.5 mL) was added, and the reaction mixture was stirred at 0 °C for 30 min under argon atmosphere. TBHP (3.0 M in toluene, 205 μL, 0.628 mmol) was added at the same temperature, and the solution color turned to red from green. After stirring for 4 h, TBHP (210 mL, 0.628 mmol) was added. Stirring was continued for further 4 h, additional $VO(acac)_2$ (5.0 mg, 19 μmol) and TBHP (210 mL, 0.628 mmol) were added. After 16 h, the reaction was quenched with saturated aqueous Na_2SO_3 (10 mL) at 0 °C and then allowed to warm to room temperature. After partition, the aqueous layer was extracted with EtOAc (6 mL x4), the combined organic layer was washed with H_2O and brine (x1), and passed through a short column packed with anhydrous Na_2SO_4. The solvents were removed by evaporation. The residue was used for the next reaction without purification. To a stirred solution of the crude product in dry CH_2Cl_2 (5.0 mL) was added dry silica gel (1.0 g) at room temperature. Being stirred for 1.5 h at the same temperature, the reaction mixture was filtered through a short column packed with cotton to remove the silica gel, which was eluted with Et_2O. The eluate was evaporated to dryness *in vacuo*. The residue was purified by flash column chromatography (neutral silica gel 15 g, Et_2O/hexane 3:1 to 5:1) to give **29** (63.8 mg, 67% in 2 steps) as a colorless crystal.

***Chaxine B (30) and BB (29)**:* $NaHCO_3$ (8.69 g), H_2O (13 ml) and acetone (6.5 mL) were added to a solution of **25** (885 mg, 2.00 mmol) in *t*-BuOMe (13 mL) at 0 °C. A solution of Oxone® (6.18 g, 20.0 mmol) in H_2O (25.0 mL) was added dropwise over 1 h. The mixture was vigorously stirred for 6 h at 0 °C. The reaction was quenched with saturated aqueous Na_2CO_3 (30 mL) and CH_2Cl_2 (10 mL) was added. After separation of the organic layer, the aqueous layer was extracted with CH_2Cl_2 (10 mL x3). The combined organic layer was passed through a short column packed with anhydrous Na_2SO_4, and concentrated to dryness *in vacuo* to give a mixture of α & β epoxide **28a**, **28b** and enol ester **25** (2:5:1.2 ratio by ^{1}H NMR). The mixture (933 mg) was dissolved in dry CH_2Cl_2 (100 mL) and then silica gel (9.30 g) was added at room temperature. After being stirred vigorously for 1 h, the reaction mixture was filtered through a cotton plug, and then rinsed with AcOEt. The filtrate was concentrated and purified by flash column chromatography (neutral silica gel 25 g, AcOEt/hexane 2:5) to give a mixture (576 mg) of chaxine B (**30**) and BB (**29**) and recovered starting material **25** (76 mg, 9%). Further purification by flash column chromatography (neutral silica gel 40 g, $CHCl_3$/THF/pentane 9:1:3) gave chaxine B (**30**) (403 mg, 44% in 2 steps) and BB (**29**) (166 mg, 18% in 2 steps) as a white amorphous solid respectively.

***Alkaline hydrolysis of Chaxine B (30) with LiOH**:* An aqueous solution of LiOH (1.2 mL; 0.05 M) was added to a solution of chaxine B (**30**) (19.3 mg, 42.0 μmol) in THF (0.6 mL) at room temperature. After stirring at the same temperature for 3 h, the reaction was quenched with 1N HCl (5 mL) and H_2O. After separation of the organic layer, the aqueous layer was extracted with CH_2Cl_2 (3 mL x3). The combined organic layer was passed through a short column packed with anhydrous Na_2SO_4, and the solvent was removed by flowing argon gas. The residue was purified by flash column chromatography (neutral silica gel 3 g, Et_2O/pentane 1:1) to give demethylincisterol A_3 (**5**) (12.1 mg, 86%) as a white solid, and **33** (1.8 mg, 34%) as volatile compound. **Demethylincisterol A_3 (5)**: Mp 141-142 °C (lit. 144-145 °C). $[\alpha]_D^{27}$ +189.6 (c 0.57, $CHCl_3$). (lit. $[\alpha]_D$ +169.72 (c 0.58, $CHCl_3$)).[6] IR (KBr) ν_{max} 3304, 2957, 2925, 2870, 1760, 1727, 1661, 1456, 1446, 1433, 1114, 980, 970, 947, 901, 853 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.63 (1H, d, *J* = 2 Hz, H-2), 5.25 (2H, dd, *J* = 15, 8 Hz, H-16), 5.16 (2H, dd, *J* = 15, 8 Hz, H-15), 3.10 (1H, s, O*H*), 2.65 (1H, m, H-8), 2.28 (1H, ddd, *J* = 14, 4, 2.5 Hz, H-5), 2.11-2.01 (1H, m), 2.01-1.94 (1H, m), 1.94-1.81 (3H, m), 1.77-1.68 (1H, m), 1.65 (1H, dd, *J* = 13.5, 4 Hz), 1.61-1.56 (1H, m), 1.55-1.46 (4H, m), 1.04 (3H, d, *J* = 7 Hz, CH_3-14), 0.92 (3H, d, *J* = 7 Hz, CH_3-21), 0.84 (3H, d, *J* = 7 Hz, CH_3-19 or 20), 0.82 (3H, d, *J* = 7 Hz, CH_3-19 or

20), 0.61 (3H, s, CH_3-12). $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$) δ 171.2, 170.8, 134.8, 133.0, 112.4, 105.0, 55.5, 50.5, 49.0, 43.0, 40.3, 35.4, 35.2, 33.2, 29.0, 21.5, 21.2, 20.1, 19.8, 17.8, 11.9. HRMS (ESI) for $C_{21}H_{31}O_3$ [M-H]- calcd, 331.2268, found: 331.2284. **Cyclohexenone 33**: ^1H NMR (400 MHz, $CDCl_3$) δ 6.96 (1H, m, H-5'), 6.05 (1H, ddd, J = 10, 2.5, 1.5 Hz, H-6'), 3.65 (1H, s, O*H*), 2.52-2.45 (2H, m, H-4'), 2.13 (1H, m, H-3'), 2.05 (1H, ddd, J = 13, 10, 7 Hz, H-3'), 1.33 (3H, s, H-7'). $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$) δ 202.9, 151.1, 126.6, 73.3, 35.8, 25.6, 24.2.

Acid hydrolysis of chaxine B (30): *p*-TsOH·H_2O (40.8 mg) was added to a solution of chaxine B (**30**) (6.9 mg, 15 μmol) in THF-H_2O (3:1) (0.8 mL) at room temperature. After stirring at the same temperature for 44 h, the reaction was quenched with 1.46 M Pyridine-THF solution (0.4 mL), and the solvent was evaporated to dryness *in vacuo*. The residue was purified by flash column chromatography (neutral silica gel 1 g, CH_2Cl_2/MeOH 20:1) to give demethylincisterol A_3 (**5**) (6.4 mg, quant.) and **34** (1.8 mg, 80%) as a white solid respectively. **34**: Mp 118-120 °C. $[\alpha]_D^{27}$ +63.2 (c 0.095, $CHCl_3$). ^1H NMR (400 MHz, $CDCl_3$) δ 4.50 (1H, m, H-5'), 2.79 (1H, dd, J = 14.5, 3.5 Hz, H-6'), 2.61 (1H, brd, J = 14.5 Hz, H-6'), 2.22-2.13 (1H, m), 2.01-1.91 (3H, m), 1.41 (3H, s, H-7'). $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$) δ 212.5, 76.3, 70.0, 45.4, 35.9, 30.1, 24.8. HRMS (ESI) for $C_7H_{12}O_3Na$ $[M+Na]^+$, calcd 167.0678, found: 167.0680.

Acid hydrolysis of BB (29): TsOH-H_2O (40.0 mg) was added to a solution of BB (**29**) (7.5 mg, 16 μmol) in THF-H_2O (3:1) (0.8 mL) at room temperature. After stirring at the same temperature for 19 h, the reaction was quenched with 1.46 M pyridine-THF solution (0.4 mL), and the solvent was evaporated to dryness *in vacuo*. The residue was purified by flash column chromatography (neutral silica gel 1 g, CH_2Cl_2/MeOH 20:1) to give demethylincisterol A_3 (**5**) (5.2 mg, 96%) as a white solid and **35** (2.5 mg, quant) as an oil. **35**: $[\alpha]_D^{26}$ -23 (c 0.11, $CHCl_3$). ^1H NMR (400 MHz, $CDCl_3$) δ 3.96 (1H, tt, J = 11, 5 Hz, H-5'), 2.86 (1H, ddd, J = 13.5, 5, 2.5 Hz, H-6'), 2.64 (1H, dd, J = 13.5, 11 Hz, H-6'), 2.14 (1H, m), 2.08 (1H, dt, J = 13.5, 3.5 Hz), 1.77 (1H, m), 1.58 (1H, td, J = 13, 3.5 Hz), 1.42 (3H, s, H-7'). $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$) δ 211.1, 75.7, 70.7, 46.9, 35.6, 32.0, 25.1. HRMS (ESI) for $C_7H_{12}O_3Na$ $[M+Na]^+$, calcd 167.0678, found: 167.0677.

Purification of 7-dehydrocholesterol (36): 7-Dehydrocholesterol (**36**) (5.69 g, supplier: nacalai tesque) was suspended in MeOH (10 mL). The mixture was stirred for 1 h at room temperature and filtered. The filtride washed with MeOH to give purified 7-dehydrocholesterol (**35**) (2.93 g, recovered 52%) as a white solid.

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***Endoperoxide 39**:* Two flat-dishes (ф 18 cm, depth 3.6 cm, polypropylene) charged with 7-dehydrocholesterol (**36**) (2.84 g, 7.38 mmol and 2.92 g, 7.59 mmol respectively), BHT (166 mg, 0.754 mmol, 182 mg, 0.827 mmol) and a magnetic stirrer bar, were placed on magnetic stirrers. *t*-BuOMe (450 mL) was added, and three mercury lamps were placed on the flat dishes. The whole apparatus was wrapped with a vinyl bag, and nitrogen gas was flowed into the bag. The flat-dishes were irradiated with the lamps at room temperature for 5.5 h. During this reaction, *t*-BuOMe (ca. 50 mL) was added at 30 min intervals to maintain the solvent volumes. The solutions of the dishes were combined and evaporated at an ice-bath temperature to give a mixture of **38**, **36** and **37** (2:1:1 ratio by ^{1}H NMR). The residue was used for the next reaction without purification. A part of the crude product was purified by flash column chromatography (Et_2O/hexane 1:4) for spectral analysis of **38**. A three-necked flask connected to a three-way stopcock was charged with the crude product (5.75 g) and BHT (1.66 g, 7.54 mmol) and then filled with argon gas. CCl_4 (300 mL) was degassed by freeze-thaw cycles (x3) in a separate flask and transferred to the reaction vessel through a syringe. NEP (**20**) (4.41 g, 23.5 mmol) was added, the reaction vessel was wrapped with aluminum foil. After stirring at room temperature under argon atmosphere for 15 h, NEP (**20**) (1.11 g, 5.90 mmol) was added. Stirring was continued for further 32 h, the solution was directly subjected to flash column chromatography (neutral silica gel 210 g, AcOEt / hexane 1:5) to give **39** (1.19 g, 19% in 2 steps) as a white amorphous solid. **38**: $[\alpha]_D^{27}$ -24.7 (c 0.94, $CHCl_3$). IR (KBr) ν_{max} 3346, 2951, 2867, 2830, 1635, 1466, 1429, 1376, 1366, 1348, 1098, 1062, 1037, 954 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 6.72 (1 H, d, J = 16 Hz, H-6), 6.01 (1H, d, J = 16 Hz, H-7), 5.70 (1H, brd, J = 3 Hz, H-9), 3.97 (1H, br, H-3), 2.55 (1H, brd, J = 16.5 Hz), 2.30-2.15 (4H, m), 2.09 (1H, dd, J = 16.5, 7 Hz), 2.04-1.92 (3H, m), 1.86 (1H, m), 1.79 (3H, s, CH_3-19), 1.66-1.47 (4H, m), 1.45-1.30 (6H, m), 1.26-1.08 (4H, m), 1.07-0.98 (1H, m), 0.96 (3H, d, J = 6.5 Hz, CH_3-21), 0.87 (3H, d, J = 6.5 Hz, CH_3-26 or 27), 0.86 (3H, d, J = 6.5 Hz, CH_3-26 or 27), 0.69 (3H, s, CH_3-18). $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$) δ 138.4, 131.6, 128.0, 125.6, 125.1, 124.8, 67.7, 54.2, 50.3, 43.0, 39.7, 36.3, 36.2, 36.1, 34.7, 31.3, 31.2, 28.8, 28.2, 25.2, 24.5, 24.0, 23.0, 22.7, 19.1, 19.1, 11.4. Anal. Calcd for $C_{27}H_{44}O$: C, 84.31; H, 11.53. Found: C, 84.31; H, 11.51. **39**: $[\alpha]_D^{27}$ +32.2 (c 1.22, $CHCl_3$). IR (KBr) ν_{max} 3444, 2950, 2932, 2870, 1640, 1467, 1441, 1056, 1036 cm^{-1}. ^{1}H NMR (400 MHz, C_6D_6) δ 5.40 (1H, m, H-7), 5.24 (1H, q, J = 2.5 Hz, H-6), 5.09 (1H, m, H-9), 3.77 (1H, m, H-3), 3.03 (1H, m), 2.30-2.15 (2H, m), 1.95-1.77 (3H, m), 1.69-1.50 (5H, m), 1.47(3H, s, CH_3-19), 1.45-1.31 (6H, m), 1.30-1.15 (6H, m), 1.02 (2H, m), 0.94 (3H, d, J = 6.5 Hz, CH_3-26 or 27), 0.93 (3H, d, J = 6.5 Hz, CH_3-26 or 27), 0.83 (3H, d, J = 6.5 Hz, CH_3-21), 0.65 (3H, s, CH_3-18). $^{13}C\{^1H\}$ NMR (100 MHz, C_6D_6) δ 142.0, 129.4, 117.7, 78.3, 76.8,

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67.4, 57.3, 48.2, 41.3, 39.9, 36.3, 36.2, 35.6, 35.6, 31.5, 31.0, 29.3, 28.4, 24.4, 23.8, 23.3, 23.0, 22.8, 18.6, 18.5, 18.5. HRMS (ESI) for $C_{27}H_{44}O_3Na$ $[M+Na]^+$, calcd 439.3182, found: 439.3186.

Unsaturated ketone 40*:* DBU (2.00 mL, 12.8 mmol) was added to a stirred solution of **39** (1.10 g, 2.64 mmol) in dry CH_2Cl_2 (30 mL) at -20 °C. After stirring at the same temperature for 24 h, the reaction was quenched with saturated aqueous NH_4Cl (30 mL) and H_2O, and the mixture was allowed to warm to room temperature. After partitioning, the aqueous layer was extracted with CH_2Cl_2 (5 mL x3), and the combined organic layer was washed with H_2O (x1) and brine (x1). The solution was passed through a short column packed with anhydrous Na_2SO_4 and concentrated to dryness *in vacuo*. The residue was purified by flash column chromatography (silica gel 30 g, Et_2O/hexane 5:1 to Et_2O) to give **40** (737 mg, 67%) as a white amorphous solid. **40**: $[\alpha]_D^{28}$ -73.6 (c 1.13, $CHCl_3$). IR (KBr) ν_{max} 3397, 2869, 1609, 1467, 1444, 1380, 1366, 1265, 1237, 1215, 1158, 1068, 1055, 1016, 965, 922, 907 cm^{-1}. ^1H NMR (400 MHz, $CDCl_3$) δ 5.99 (1H, d, *J* = 2 Hz, H-7), 5.04 (1H, brs, H-9), 4.00 (1H, br, H-3), 2.75-2.67 (2H, m), 2.61 (1H, brs), 2.32 (1H, dt, *J* = 18, 5.5 Hz), 2.25-2.07 (2H, m), 2.00-1.72 (6H, m), 1.89 (3H, s, CH_3-19), 1.70-1.61 (2H, m), 1.57-1.44 (3H, m), 1.44-1.28 (5H, m), 1.21-1.09 (3H, m), 1.07-0.97 (1H, m), 0.93 (3H, d, *J* = 6 Hz, CH_3-21), 0.87 (3H, d, *J* = 6.5 Hz, CH_3-26 or 27), 0.86 (3H, d, *J* = 6.5 Hz, CH_3-26 or 27), 0.59 (3H, s, CH_3-18). $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$) δ 197.3, 160.5, 141.1, 131.6, 124.6, 66.6, 64.8, 56.6, 51.2, 46.1, 39.6, 36.2, 36.1, 35.4, 34.8, 31.3, 30.5, 29.8, 28.2, 27.7, 24.0, 23.0, 22.7, 22.4, 21.6, 18.9, 12.4. HRMS (ESI) for $C_{27}H_{44}O_3Na$ $[M+Na]^+$, calcd 439.3182, found: 439.3180.

Synthesis of enol ester 42, 43 and 44 from 40*:* A suspension of **40** (195 mg, 0.468 mmol) and activated AW-300 (94 mg) in dry CH_2Cl_2 (4.5 mL) was stirred under argon atmosphere at -40 °C for 30 min. TMSOTf (0.20 mL, 1.1 mmol) was added at the same temperature and the reaction was quenched with saturated aqueous $NaHCO_3$ (10 mL) after stirring for 20 min. The mixture was diluted with H_2O, allowed to warm to room temperature. To remove AW-300, the mixture was filtered through a cotton plug. The filtrate was partitioned, and the aqueous layer was extracted with CH_2Cl_2 (5 mL x3). The combined organic layer was washed with H_2O (x1) and brine (x1), and filtered through a short column packed with anhydrous Na_2SO_4. The solvent was evaporated to dryness *in vacuo* to give furan **41** (184 mg) as a white amorphous solid, a sample of which was purified by flash column chromatography (CH_2Cl_2/hexane 5:1) for spectral analysis. The residue was used for the next reaction without purification. The crude product (185 mg) was dissolved in dry CH_2Cl_2 (4.5 mL) and then MCPBA (157 mg, 0.910

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mmol) was added at -40 °C under argon atmosphere. After stirring for 40 min, the reaction was quenched with saturated aqueous Na_2SO_3 (10 mL) at -40 °C. H_2O was added and the mixture was allowed to warm to room temperature. After partitioning, the aqueous layer was extracted with CH_2Cl_2 (5 mL x3). The combined organic layer was washed with saturated aqueous $NaHCO_3$ (x2) and brine (x1), and passed through a short column packed with anhydrous Na_2SO_4. The solvent was evaporated to dryness *in vacuo*. The residue was purified by flash column chromatography (neutral silica gel 10 g, AcOEt/hexane 2:3 to 1:2) to give **42** (88.5 mg, 44%) as a yellow oil and a mixture of **43** and **44** (1:1 ratio by ^{1}H NMR, 61.7 mg, calculated 15% yield, respectively) as a solid. A part of the mixture was purified by flash column chromatography (AcOEt:CH_2Cl_2 1:5) for spectral analysis of **43** and **44**. **41**: $[\alpha]_D^{27}$ +15 (c 0.73, C_6H_6). IR (KBr) ν_{max} 3385, 2952, 2931, 2868, 1698, 1623, 1466, 1445, 1377, 1366, 1331, 1098, 1052 cm^{-1}. ^{1}H NMR (400 MHz, C_6D_6) δ 6.18 (1H, s, H-7), 3.78 (1H, m, H-3), 2.79 (1H, brd, *J* = 16.5 Hz), 2.72-2.56 (2H, m), 2.56-2.46 (1H, m), 2.38 (1H, ddd, *J* = 16, 7.5, 2 Hz), 2.15-1.90 (3H, m), 1.99 (3H, s, CH_3-19), 1.89-1.81 (1H, m), 1.67-1.33 (10H, m), 1.28-1.15 (5H, m), 1.07 (1H, m), 0.95 (9H, d, *J* = 6.5 Hz, CH_3-21 and 26 and 27), 0.62 (3H, s, CH_3-18). $^{13}C\{^1H\}$ NMR (100 MHz, C_6D_6) δ 154.0, 148.3, 129.1, 121.2, 120.0, 107.3, 66.9, 54.0, 47.8, 44.1, 39.9, 37.1, 36.7, 36.5, 35.8, 31.7, 31.4, 30.0, 28.4, 24.5, 23.9, 23.1, 22.8, 21.8, 21.6, 19.1, 11.0. Anal. Calcd for $C_{27}H_{42}O_2$: C, 81.35; H, 10.62. Found: C, 81.49; H, 10.79. **42**: $[\alpha]_D^{28}$ +86.9 (c 1.45, $CHCl_3$). IR (KBr) ν_{max} 3437, 2953, 2932, 2869, 1731, 1704, 1645, 1467, 1446, 1382, 1326, 1236, 1159, 1146, 1132, 1114, 1039 cm^{-1}. ^{1}H NMR (400 MHz, C_6D_6) δ 5.61 (1H, d, *J* = 2.5 Hz, H-7), 3.83 (1H, m, H-3), 2.67 (1H, brd, *J* = 16 Hz), 2.54-2.39 (2H, m), 2.34 (1H, ddd, *J* = 15, 5, 2 Hz), 2.04-1.89 (2H, m), 1.83-1.61 (3H, m), 1.65 (3H, brs, CH_3-19), 1.60-1.47 (4H, m), 1.39-1.05 (12H, m), 0.93 (3H, d, *J* = 6.5 Hz, CH_3-26 or 27), 0.92 (3H, d, *J* = 6.5 Hz, CH_3-26 or 27), 0.73 (3H, d, *J* = 6.5 Hz, CH_3-21), 0.57 (3H, s, CH_3-18). $^{13}C\{^1H\}$ NMR (100 MHz, C_6D_6) δ 202.1, 164.2, 155.9, 139.4, 120.5, 117.7, 66.9, 57.2, 55.5, 46.1, 39.9, 38.9, 37.9, 36.7, 36.2, 36.0, 30.8, 28.9, 28.4, 27.4, 24.3, 23.0, 22.8, 22.0, 18.7, 16.0, 11,6. HRMS (ESI) for $C_{27}H_{42}O_4Na$ $[M+Na]^+$, calcd 453.2975, found: 453.2964. **43**: Mp 91-96 °C. $[\alpha]_D^{27}$ -16.7 (c 1.13, $CHCl_3$). IR (KBr) ν_{max} 3520, 3441, 2953, 2932, 2869, 1723, 1645, 1467, 1419, 1378, 1366, 1237, 1055 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 4.03 (1H, m, H-3), 3.91 (1H, s, H-7), 2.85, (1H, brd, *J* = 15.5 Hz), 2.60-2.48 (2H, m), 2.41 (1H, dt, J = 19, 6 Hz), 2.32 (1H, dd, J = 12, 7 Hz), 2.28-2.13 (2H, m), 2.05-1.95 (2H, m), 1.91 (3H, brs, CH_3-19), 1.86-1.76 (2H, m), 1.75-1.62 (3H, m), 1.57-1.43 (4H, m), 1.41-1.23 (4H, m), 1.18-1.07 (2H, m), 1.14 (3H, s, CH_3-18), 1.03 (1H, m), 0.95 (3H, d, *J* = 6 Hz, CH_3-21), 0.86 (3H, d, *J* = 6.5 Hz, CH_3-26 or 27), 0.85 (3H, d, *J* = 6.5 Hz, CH_3-26 or 27). $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$) δ 205.5, 199.7, 147.2, 128.5, 69.1, 66.0, 62.4, 55.9, 52.8, 45.6, 39.5, 39.0, 37.3, 36.0, 35.6, 33.7, 31.4, 29.8, 28.6, 28.1, 23.9, 22.9, 22.7, 21.7, 20.3, 18.8, 11.9. HRMS (ESI) for $C_{27}H_{42}O_4Na$ [M+Na]$^+$, calcd 453.2975, found: 453.2972. **44**: Mp 180-183 °C. $[\alpha]_D^{28}$ +8.68 (c 0.95, $CHCl_3$). IR (KBr) ν_{max} 3462, 2956, 2868, 1670, 1642, 1464, 1444, 1381, 1297, 1234, 1137, 1106, 1084, 1025, 1006, 970, 954, 914 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 6.25 (1H, br, O*H*), 5.79 (1H, d, *J* = 2 Hz, H-7), 4.27 (1H, brd, 10 Hz, O*H*), 3.98 (1H, m, H-3), 3.07 (1H, br, O*H*), 2.70 (1H, t, *J* = 10 Hz, H-14), 2.51 (1H, td, *J* = 14, 4 Hz), 2.26 (1H, dd, *J* = 15, 4 Hz), 2.10 (1H, m), 2.05-1.89 (5H, m), 1.74-1.66 (2H, m), 1.56-1.46 (3H, m), 1.43-1.32 (6H, m), 1.29 (3H, s, CH_3-19), 1.18-1.09 (3H, m), 1.06-0.97 (1H, m), 0.92 (3H, d, *J* = 6 Hz, CH_3-21), 0.87 (3H, d, *J* = 6 Hz, CH_3-26 or 27), 0.86 (3H, d, *J* = 6 Hz, CH_3-26 or 27), 0.65 (3H, s, CH_3-18). $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$) δ 195.1, 162.6, 120.6, 93.6, 88.4, 74.3, 66.0, 56.1, 52.7, 48.2, 39.6, 39.2, 38.3, 36.2, 36.0, 31.5, 30.9, 28.5, 28.1, 24.0, 23.8, 22.9, 22.7, 20.9, 18.8, 11.8. HRMS (ESI) for $C_{27}H_{44}O_5Na$ [M+Na]$^+$, calcd 471.3081, found: 471.3071.

Synthesis of 17-nor-chaxine D (46) and 17-nor-2'-epi-chaxine D (45): Compound **42** (104 mg, 0.242 mmol) was dissolved in *t*-BuOMe (3 mL), $NaHCO_3$ (1.04 g), H_2O (3 mL) and acetone (1.5 mL) at 0 °C. After stirring at the same temperature, a solution of Oxone® (718 mg, 2.33 mmol) in H_2O (3 mL) was added dropwise over 20 min. The mixture was vigorously stirred at 0 °C for 8 h. The reaction was quenched with saturated aqueous Na_2SO_3 (15 mL) and AcOEt (5 mL). After separation of the organic layer, the aqueous layer was extracted with AcOEt (5 mL x3). The combined organic layer was washed with H_2O (x1) and brine (x1), and filtered through a short column packed with anhydrous $Na_2SO_{4,}$ and concentrated to dryness *in vacuo* to give a mixture of α & β epoxide. The ratio of α/β epoxide was determined to be ca. 5:2 by ^{1}H NMR spectra. To a stirred solution of the crude product (103 mg) in dry CH_2Cl_2 (10 mL) was added silica gel (1.02 g) at room temperature. Being stirred for 1 h, the reaction mixture was filtered through a cotton plug, and then eluted with AcOEt. The filtrate was concentrated and purified by flash column chromatography (neutral silica gel 10 g, AcOEt/hexane 2:5) to give a mixture (65.7 mg) of 17-nor-chaxine D (**46**) and 17-nor-2'-epi-chaxine D (**45**) and recovered **42** (4.2 mg). Further purification by flash column chromatography (neutral silica gel 10g, $CHCl_3$/THF/pentane 9:1:3) to give 17-nor-chaxine D (**46**) (44.3 mg, 41% in 2 steps) and 17-nor-*epi* chaxine D (**45**) (17.2 mg, 16% in 2 steps) as a white amorphous solid respectively. **17-nor-chaxine D (46)**: $[\alpha]_D^{25}$ +68 (c 0.67, $CHCl_3$). IR (KBr) ν_{max} 3427, 2869, 1711, 1644, 1467, 1456, 1446, 1377, 1331, 1285, 1258, 1166, 1143, 1127, 1075 cm $^{-1}$. ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.57 (1H, d, *J* = 2 Hz, H-2), 3.95 (1H, tt, *J* = 9, 5 Hz, H-5'), 2.87 (1H, dd, *J* = 13, 9 Hz), 2.74-2.63 (2H, m), 2.55-2.45 (2H, m), 2.33 (1H, td, *J* = 15, 5 Hz), 2.25 (1H, ddd, *J* = 13, 7, 2 Hz), 2.10-2.01 (1H, m), 1.99-1.86 (2H, m), 1.74-1.60 (3H, m), 1.59-1.47 (4H, m), 1.44 (3H, s, CH_3-7'), 1.43-1.30 (4H, m), 1.21-1.08 (3H, m), 1.02

(1H, m), 0.93 (3H, d, *J* = 6 Hz, CH_3-14), 0.87 (3H, d, *J* = 6.5 Hz, CH_3-19 or 20), 0.86 (3H, d, *J* = 6.5 Hz, CH_3-19 or 20), 0.86 (3H, s, CH_3-12). $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$) δ 204.7, 203.8, 165.4, 156.2, 117.4, 82.7, 70.3, 57.6, 55.7, 47.9, 46.5, 39.5, 39.0, 38.0, 35.9, 35.9, 33.4, 29.7, 28.9, 28.1, 23.9, 22.9, 22.7, 22.1, 20.4, 18.8, 12.0. HRMS (ESI) for $C_{27}H_{42}O_5Na$ $[M+Na]^+$, calcd 469.2924, found: 469.2921. **17-nor-*epi* chaxine D (45)**: $[\alpha]_D^{25}$ +69 (c 0.43, $CHCl_3$). IR (KBr) ν_{max} 3435, 2083, 1729, 1698, 1640, 1468, 1451, 1377, 1329, 1163, 1124, 1090, 957, 862 cm $^{-1}$. ^1H NMR (400 MHz, $CDCl_3$) δ 5.52 (1H, d, *J* = 2 Hz, H-2), 4.46 (1H, br, H-5’), 3.10 (1H, dd, *J* = 13.5, 3.5 Hz), 2.68 (1H, ddd, *J* = 15, 13, 7 Hz), 2.55-2.46 (2H, m), 2.39 (1H, dd, *J* = 13, 4 Hz), 2.26 (1H, brdd, *J* = 13, 7 Hz), 2.15 (1H, m), 2.10-1.97 (3H, m), 1.75-1.60 (4H, m), 1.57-1.47 (3H, m), 1.45 (3H, s, CH_3-7’), 1.42-1.30 (4H, m), 1.18-0.98 (4H, m), 0.94 (3H, d, *J* = 6.5 Hz, CH_3-14), 0.87 (3H, s, CH_3-12). 0.87 (3H, d, *J* = 6.5 Hz, CH_3-19 or 20), 0.86 (3H, d, *J* = 6.5 Hz, CH_3-19 or 20). $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$) δ 205.6, 204.0, 165.5, 154.9, 117.5, 84.5, 70.5, 57.5, 55.7, 46.6, 46.4, 39.5, 38.9, 38.0, 35.9, 34.7, 29.8, 28.9, 28.2, 28.1, 23.9, 22.9, 22.7, 22.1, 20.5, 18.8, 12.0. HRMS (ESI) for $C_{27}H_{42}O_5Na$ $[M+Na]^+$, calcd 469.2924, found: 469.2935.

***Oxidative cascade of 41 with MCPBA**:* MCPBA (34.0 mg, 0.198 mmol) was added to a solution of **41** (25.4 mg, 63.8 μmol) in dry CH_2Cl_2 (1.0 mL) at -40 °C. The reaction mixture was allowed to warm to room temperature gradually and stirred under argon atmosphere for 15 h. The reaction was quenched with saturated aqueous Na_2SO_3 (3 mL) and CH_2Cl_2 was added. After separation of the organic layer, the aqueous layer was extracted with CH_2Cl_2 (3 mL x3). The combined organic layer was filtered through a short column packed with anhydrous Na_2SO_4 and concentrated to dryness *in vacuo.* The residue was purified by flash column chromatography (neutral silica gel 5 g, $CHCl_3$/THF/pentane 9:1:3) to give a 3:4:1:2 (by ^1H NMR) mixture of 17-nor-chaxine D (**46**), 17-nor-2’*epi*-chaxine D (**45**), **43** and **44**. Separation of the products was difficult because of the similar polarity.

17-nor-chaxine E (47): MsCl (25 μL, 32 μmol) and NEt_3 (135 μL, 0.969 mmol) were added to solution of 17-nor-chaxine D (**46**) (14.2 mg, 31.8 μmol) in dry CH_2Cl_2 (1 mL) at 0 °C. The ice bath was removed and the reaction mixture was stirred for 45 min. Then the reaction was quenched with saturated aqueous $NaHCO_3$ (3 mL). The organic layer was separated and the aqueous layer was extracted with CH_2Cl_2 (3 mL x3). The combined organic layer was washed with H_2O (x1) and brine (x1), and filtered through a short column packed with anhydrous Na_2SO_4, and concentrated to dryness *in vacuo.* The residue was purified by preparative TLC (AcOEt/Hexane 1: 4) to give 17-nor-chaxine E (**47**) (11.8 mg, 86%) as a white solid. **17-nor-chaxine E** (**47**): Mp 84-86 °C. $[\alpha]_D^{28}$ +134 (c 0.59, $CHCl_3$). IR (KBr) ν_{max} 2954, 2869, 1729,

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1699, 1649, 1466, 1427, 1384, 1328, 1193, 1164, 1120, 1090, 1001, 866, 813 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 6.89 (1H, m, H-5'), 6.03 (1H, dd, *J* = 10, 2 Hz, H-6'), 5.64 (1H, d, *J* = 2 Hz, H-2), 2.93 (1H, td, *J* = 12, 6 Hz), 2.72 (1H, ddd, *J* = 16, 13, 7 Hz), 2.57-2.37 (4H, m), 2.23 (1H, ddd, *J* = 13, 7, 2 Hz), 2.10-1.98 (2H, m), 1.70-1.27 (9H, m) 1.47 (3H, s, CH_3-7'), 1.23-1.06 (3H, m), 1.01 (1H, m), 0.92 (3H, d, *J* = 6.5 Hz, CH_3-14), 0.87 (3H, d, *J* = 6.5 Hz, CH_3-19 or 20), 0.85 (3H, d, *J* = 6.5 Hz, CH_3-19 or 20), 0.85 (3H, s, CH_3-12). $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$) δ 204.9, 195.9, 164.4, 156.5, 149.0, 128.3, 117.5, 81.3, 57.9, 55.7, 46.8, 39.5, 39.2, 38.2, 35.9, 35.9, 32.2, 28.9, 28.1, 24.9, 23.9, 22.9, 22.7, 22.0, 21.8, 18.8, 11.9. HRMS (ESI) for $C_{27}H_{40}O_4Na$ $[M+Na]^+$, calcd 451.2819, found: 451.2808.

17-nor-Chaxine A (48)*:* To a solution of 17-nor-chaxine D (**46**) (9.0 mg, 20 μmol) in THF (0.50 mL) was added 0.05 N LiOH (0.60 mL) at room temperature. After stirring at the same temperature for 3 h, the reaction was quenched with 1 N HCl (3 mL) and H_2O. After separation of the organic layer, the aqueous layer was extracted with CH_2Cl_2 (3 mL x3). The combined organic layer was passed through a short column packed with anhydrous Na_2SO_4, and concentrated to dryness by spraying argon gas. The residue was purified by flash column chromatography (neutral silica gel 3 g, Et_2O/pentane 1:1) to give 17-nor-chaxine A (**48**) (5.7 mg, 94%) as a white solid and **33** (1.0 mg, 35%) as a volatile compound. **17-nor-chaxine A (48)**: Mp 101-103 °C. $[\alpha]_D^{28}$ +165 (c 0.29, $CHCl_3$). IR (KBr) ν_{max} 3413, 2955, 2930, 2869, 1742, 1662, 1467, 1381, 1177, 1128, 1048, 957, 934, 904, 853 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.62 (1H, d, *J* = 2 Hz, H-2), 3.21 (1H, br, O*H*), 2.64 (1H, ddd, *J* = 12, 7, 2 Hz, H-8), 2.27 (1H, ddd, *J* = 14, 4, 2.5 Hz), 2.07 (1H, m), 2.01 (1H, ddd, J = 13.5, 4.5, 2.5 Hz), 1.86 (1H, td, *J* = 14, 4.5 Hz), 1.75 (1H, m), 1.66- 1.57 (1H, m), 1.57-1.30 (6H, m), 1.20-1.01 (4H, m), 0.94 (3H, d, *J* = 6 Hz, CH_3-14), 0.87 (3H, d, *J* = 6.5 Hz, CH_3-19 or 20), 0.86 (3H, d, *J* = 6.5 Hz, CH_3-19 or 20), 0.59 (3H, s, CH_3-12). $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$) δ 171.2, 170.9, 112.4, 105.0, 55.7, 50.4, 49.2, 39.6, 36.1, 35.9, 35.4, 35.3, 28.8, 28.1, 23.9, 22.9, 22.7, 21.5, 18.8, 11.6. HRMS (ESI) for $C_{20}H_{31}O_3$ $[M-H]^-$, calcd 319.2268, found: 319.2276.

Supporting information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.joc.XXXXXXX.

An alternative biosynthetic route, X-ray crystallographic data for compound **27**, setup apparatus for electrocyclic reaction of ergosterol, and spectral data for all new compounds (PDF)

Crystal data for compound **27** (CIF)

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