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Structure Elucidation

A Geomimetic Approach to the Formation and Identification of Fossil Sterane Biomarkers in Crude Oil: 18-*nor*-D-*homo*-Androstane and 5α,14β-Androstane

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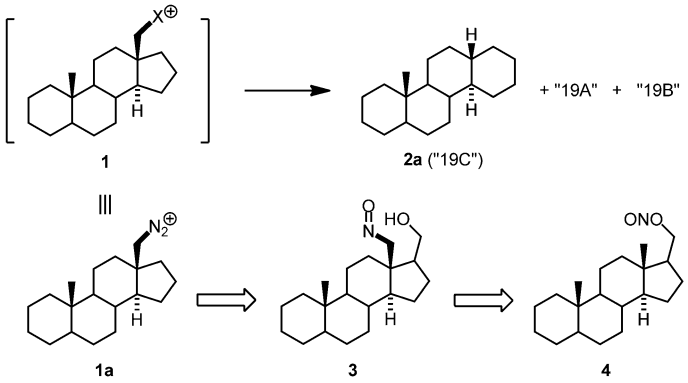
Abstract: A diazonium ion derived from 18-aminoandrostane rearranged upon decomposition by a carbonium and a carbenium ion to furnish a mixture of a cyclopropanated compound and two D-*homo*-androstenes. Hydrogenation of this mixture gave the saturated hydrocarbons, 18-*nor*-D-*homo*-androstane and 5α,14β-androstane, which are both fossil sterane biomarkers in Neoproterozoic crude oil. The so far unknown constitution and configuration as well as the geochemical genesis were established by this experiment. The starting material for this investigation, 18-aminoandrostane, was prepared in twelve steps from androstan-17-one (12.5% overall yield) with a Barton reaction as the key step.

Introduction

Molecular fossils (geochemical biomarkers) are widely used in modern organic geochemistry for studying geological, geobiological, and geothermal processes. Their structures, distribution patterns, and reaction pathways reveal insight into geological history.[1] With the structures of those biomarkers in hand, organic geochemists are able to link them to potential biogenic precursors and the organisms that biosynthesized them and, in many cases, to the depositional environment of sediment deposition.

Mass spectrometric analyses show that approximately 550-million-year-old crude oils and sedimentary rocks from the Oman Salt Basin contain (at least) three gas chromatographically separated isomers of putative low-molecular-weight steranes ($C_{19}H_{32}$, *m/z*: 260; "19A", "19B", and "19C" in order of elution) with modified androstane carbon skeletons and unknown constitutions.[2] Co-eval sediments from Eastern Siberia, India, and Australia contain the same hydrocarbons.[2c] Since intense organic geochemical investigations provided evidence that the occurrence and relative abundance of the unknown biomarkers correlate with geological age and the salinity of the waters during sediment accumulation, there is considerable interest in revealing the precise structures of the unknowns. The concentrations of the unknown biomarkers in the Oman samples are too low for them to be readily isolated as pure compounds for rigorous spectroscopic structural assignment.



Scheme 1. Geochemical precursor compound **1** leading to 18-*nor*-D-*homo*-androstane **2** and the two putative unknown $C_{19}H_{32}$ steranes "19A" and "19B"; synthetic approach to diazonium ion **1a** by Barton reaction.

In extensive synthetic studies,[3] we were recently able to identify 5α,13β,14α-18-*nor*-D-*homo*-androstane (**2a**, Scheme 1) as the hitherto unknown sterane "19C" by unequivocal GC co-elution experiments.[4] Conceivably, enlargement of the D-ring may have occurred by Wagner–Meerwein rearrangement of a species **1** with a leaving group X at C-18, which could be

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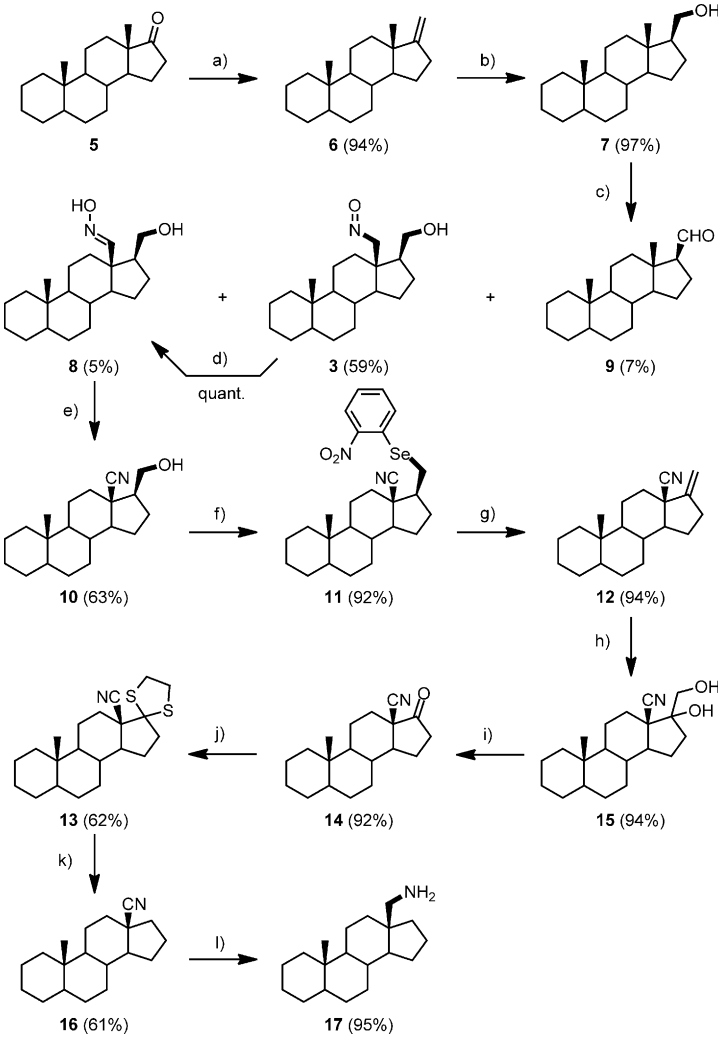
either a protonated alcohol or a halogen atom activated by a Lewis acid.[5] These precursors could have been formed by marine organisms in aerobic, saline environments by oxidative hydroxylation or halogenation of the C-18 methyl group and are well-known from laboratory synthesis.[6] It therefore appears possible that isomers "19A" and "19B" may have been formed from the same reactive intermediate **1**.

Accordingly, we set up the following synthetic strategy for the identification of compounds "19A" and "19B": A diazonium ion **1a** could be a suitable analogue of the postulated geochemical precursor **1**. An amino function at C-18 could be installed from nitroso compound **3** which can be synthesized by a Barton reaction from nitrous acid ester **4**. A prerequisite for such a key transformation would be the installation of a hydroxymethyl substitutent at C-17 and its removal after nitrosation of carbon atom C-18. Thus, androstan-17-one would be a suitable synthetic precursor for this investigation, since the 17-hydroxymethyl group can be introduced by a Wittig reaction followed by *anti*-Markownikoff hydroxylation.

Results and Discussion

Synthesis of 18-aminoandrostane **17** started with commercially available dihydrotestosterone. Using literature procedures, the carbonyl group at C-3 was removed by a Wolff–Kishner reduction and the secondary alcohol at C-17 oxidized with pyridinium chlorochromate (PCC) furnishing 5α-androstan-17-one **5** (Scheme 2).[7] We then started to build up the side chain required for Barton reaction by Wittig olefination, which gave 17-methylene derivative **6** (94%).[8] Subsequent hydroboration with 9-BBN yielded the primary alcohol **7** (97%) after oxidative workup. The Barton reaction[9] was accomplished in two consecutive reactions on a 500 mg scale: Under light exclusion nitrous acid ester **4** was formed by reaction of alcohol **7** with *t*BuONO in $CHCl_3$. All volatile materials were removed in the dark and the residue redissolved in anhydrous acetone, transferred to a bubble column reactor (stream of nitrogen) with cooling jacket (water, 20 °C) and an inner mercury UV-lamp (150 W). After 20 min of irradiation, the reaction mixture was separated by column chromatography yielding four fractions: Starting material **7** (29%), aldehyde **9**[10] (7%), oxime **8** (5%), and nitroso compound **3** (59%) (yields based on recovered starting material **7** were 10% aldehyde **9**, 7% oxime **8**, and 83% nitroso compound **3**). The latter could be quantitatively transformed into oxime **8** by heating it in isopropanol. We then converted the oxime to nitrile **10** (63%) with the Mukaiyama reagent[11] in order to preserve the nitrogen functionality in a protected form. Actually, we had initially investigated alternative routes with other protective groups at the primary amino function (Cbz or phthalimide), but they always led to pyrrolidine ring formation upon attempts of alcohol elimination (i.e. the formation of norconanine derivatives).[12] Since water elimination under acidic conditions was not fruitful, we prepared selenyl ether **11** (92%) from *ortho*-nitrophenylselenocyanate[13] and eliminated it oxidatively to furnish methylene compound **12** (94%). Degradation of the side chain was then accomplished in two steps by osmium-catalyzed dihydroxylation (product **15**, 94%) and periodate cleavage. Since Wolff–Kishner reduction of the ketone failed, we removed this functionality by dithioacetal formation (product **13**, 62%) and reduction with Raney nickel (product **16**, 61%).[14] Finally, the nitrile was reduced with $LiAlH_4$ to furnish the primary amine **17** (95%; 0.5 mmol scale). The overall yield of the twelve-step sequence was 12.5%.



Scheme 2. Synthesis of 18-aminoandrostane **17**: a) *n*BuLi (3 equiv), $MePPh_3Br$ (3 equiv), THF, 66 °C, 16 h; b) 1. 9-BBN (3 equiv), THF, 66 °C, 3 h; 2. NaOH, H_2O_2, H_2O, 23 °C, 16 h; c) 1. *t*BuONO (10 equiv), $CHCl_3$, 23 °C, 0.5 h; 2. removal of all volatiles, 3. acetone, UV light, 20 min, 20 °C, 29% of starting material **7** recovered; d) *i*PrOH, 82 °C, 2 h; e) 2-chloro-1-methylpyridiniumiodide (1.3 equiv), NEt_3 (1.3 equiv), CH_2Cl_2, 40 °C, 24 h; f) 2-$(O_2N)C_6H_4SeCN$ (1.5 equiv) and nBu_3P, THF, 3 h, 66 °C; g) H_2O_2, H_2O, THF, 66 °C, 4 h; h) $K_2OsO_4 \cdot 2H_2O$ (0.02 equiv), NMO (2 equiv), THF, acetone, *t*BuOH, H_2O, 23 °C, 40 h; i) $NaIO_4$ (5 equiv), THF, H_2O, reflux, 4 h; j) $(CH_2SH)_2$ (1.3 equiv), *p*TosOH·H_2O (0.03 equiv), C_6H_6, 80 °C, 16 h; k) Raney Ni, EtOH, 3.5 h, 78 °C; l) $LiAlH_4$ (5 equiv), THF, 20 h, 66 °C. 9-BBN = 9-borabicyclo[3.3.1]nonane; NMO = *N*-methylmorpholine-*N*-oxide.

Diazotation of primary amine **17** was accomplished with aqueous $NaNO_2$ in $AcOH/CH_2Cl_2$. A hydrocarbon fraction (57% yield) was extracted from the reaction mixture consisting of three components with *m/z*: 258 (GCMS). 92% (area of GC signal) of this mixture was cyclopropane compound **18**, which could actually be separated from the two other components by column chromatography. Formation of the cyclopropane ring proceeded by approach of C-18 (during the release of N_2 from the diazonium ion) to C-14 on the β-face. As reported in the literature, a carbocation ion is formed, which releases a proton to the solvent with inversion of configuration at C-14.[15]

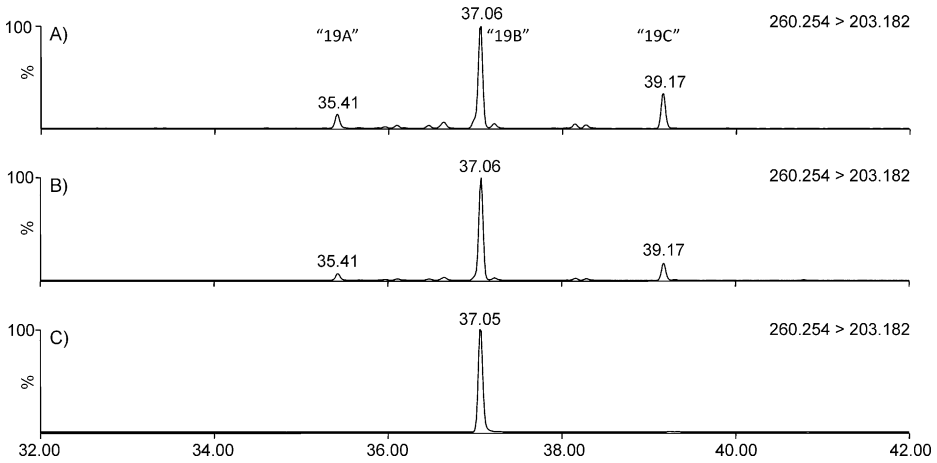
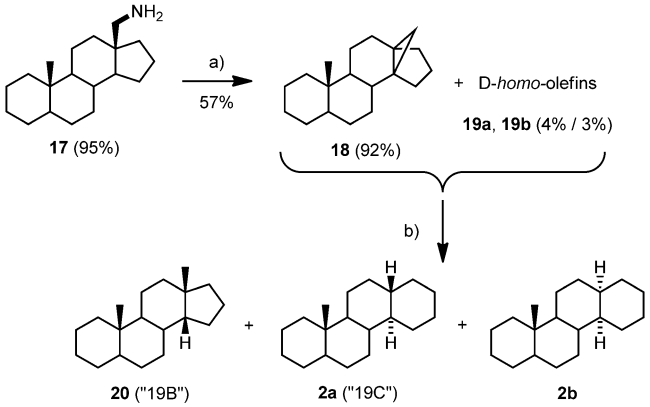


Figure 1. GCMS coinjection experiments of **20** and the sterane "19B". Chromatogram A is the *m/z*: 260→203 transition for the saturated hydrocarbon fraction of an Oman crude oil (OMO 037), chromatogram C is compound **20**. Chromatogram B is the admixture of the crude oil hydrocarbons containing "19B" plus **20** and showing an increase in the intensity of the peak at 37.06 min relative to the other peaks. The absence of peak broadening confirms a perfect match in this data for a DB-5MS column. Experiments conducted with DB-1MS and DB-XLB liquid phases yielded matching results.

The other two components of the mixture, named **19a** and **19b** (4 and 3% GC area in the mixture), were not separable from each other and residual amounts of compound **18**. They are presumably D-*homo*-androstenes with one C–C double bond located either at C-13(17) or C-12, as was indicated by ^{13}C NMR spectroscopy of the mixture of the three compounds **18**, **19a**, and **19b**. These D-*homo*-sterenes result from an 1,2-alkyl-shift of C-17 to C-18 under formation of a tertiary carbenium ion at C-13 with subsequent deprotonation of either C-17 or C-18. The D-*homo*-constitution of **19a**/**19b** was furthermore proven by the following derivatization: Hydrogenation of this mixture (after acidic pre-treatment) yielded three components with *m/z*: 260 consisting of the saturated hydrocarbons **20**, **2a**, and **2b** (ratio 81, 16, and 3% from GC). The structures of compounds **2a** and **2b** were unambiguously confirmed by comparison (GC coinjection) with authentic standards previously prepared in our laboratory.[4] As mentioned in the introduction, compound **2a** had already been identified to be component "19C" in natural crude oil. Furthermore, GC coinjection experiments confirmed by that hydrocarbon **20** was identical to crude oil component "19B" (Figure 1), which is actually the most important key finding of this manuscript.

To elucidate the constitution and configuration of compound **20** we started with cyclopropane derivative **18**, which as mentioned was separable from olefins **19a**/**19b**. The main ^1H NMR spectroscopic feature of this compound is the AB-system with $\delta_A = 0.05$, $\delta_B = 0.44$ ppm and $J_{AB} = (-)4.4$ Hz. Compound **18** was, however, inert to direct hydrogenation, but could be isomerized under acidic conditions to two unknown olefins, which then were both hydrogenated to give compound **20**, which is actually the reason for acidic pretreatment at step (b) in Scheme 3. Anyhow, cyclopropane derivative **18** could be reduced under ionic conditions with TFA/Et_3SiH[16] to a clean product **20** as the only saturated hydrocarbon (*m/z*: 260). It was separated from residual amounts of olefins by



Scheme 3. Diazotization and rearrangement of amine **17** with subsequent hydrogenation: a) $NaNO_2$ (20 equiv), H_2O, AcOH, CH_2Cl_2, 23 °C, 2 h; b) 1. TFA, $CHCl_3$, −20 °C, 2.5 h; 2. H_2 (4 bar), Pd/C, 50 °C, 2 days. TFA = trifluoroacetic acid.

chromatography on SiO_2 impregnated with $AgNO_3$ (81% yield).[17] This process presumably proceeded by protonation of the C-18 methylene group under opening of the three-membered ring with concurrent delivery of a hydride ion to C-14 with inversion of configuration. Structural elucidation by NMR spectroscopy turned out to be impossible. Fortunately, we were able to grow single crystals of this compound which allowed for an X-ray structure determination.[18] Figure 2 gives an

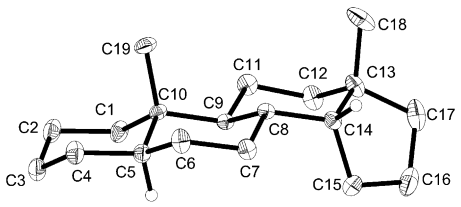
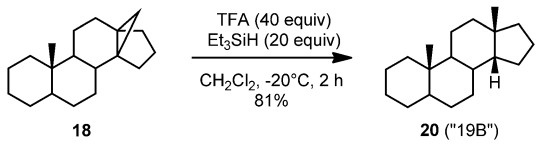


Figure 2. ORTEP-representation of the structure of 5α,14β-androstane (**20**).

ORTEP representation of compound **20** in the solid state. The key structural issue is the *cis*-annulation of rings C and D with both, 13-Me and 14-H in β-configuration, thus compound **20** ("19B") is 5α,14β-androstane (Scheme 4).[19]



18 **20** ("19B")

Scheme 4. Selective conversion of methano-derivative **18** furnishing fossil natural product 14β-androstane (**20**).

Conclusion

Based on the identification of 18-*nor*-D-*homo*-androstane (**2a**) as a fossil C_{19} sterane in our previous investigation, we made the assumption that diazonium ion **1a** derived from primary amine **17** could be a geomimetic precursor to compound **2a** as well as other fossil C_{19} steranes ($C_{19}H_{32}$, *m/z*: 260) with so far unknown constitutions. The decomposition of diazonium salt **1a** proceeded to a mixture of cyclopropane derivative **18** and two D-*homo*-androstenes **19a** and **19b**. Subsequent hydrogenation of the mixture indeed gave saturated hydrocarbons **20**, **2a**, and **2b**. The formation of 18-*nor*-D-*homo*-androstane (**2a**) confirmed our presumption of the geochemical genesis of this biomarker. Compound **2b** was its 13α-epimer, which is actually not present in recognizable amount in natural samples. We furthermore proved by GC-coinjection that 5α,14β-androstane (**20**) was identical to the fossil biomarker "19B", the molecular structure of which was so far unknown. Constitution and configuration of compound **20** was established by an X-ray single-crystal structure analysis.

Synthesis of the starting material **17** of this study began with androstanone **5**. To achieve the installation of a nitrogen function at C-18 by Barton reaction, a hydroxymethyl side chain had to be assembled at C-17 by Wittig olefination and hydroboration. After Barton nitrosation, the nitrogen functionality was preserved as a nitrile and the C-17 side chain uninstalled by elimination, dihydroxylation, and periodate cleavage. Finally, the carbonyl group was removed by reduction of the respective dithioacetal and the nitrile reduced to the primary amino group. The overall yield of this twelve-step sequence was 12.5%.

Experimental Section

General

Preparative column chromatography was carried out using Merck SiO_2 (35–70 μm, type 60 A) with hexane, *tert*-butyl methyl ether (MTBE), and ethyl acetate (EtOAc) as eluents. TLC was performed on Merck aluminum plates coated with SiO_2 F_{254}. ^{1}H- and ^{13}C NMR spectra were recorded on a Bruker Avance DRX 500 instrument. Multiplicities of carbon signals were determined with DEPT experiments. MS and HRMS spectra of synthesis products were obtained with a Finnigan MAT 95 (EI and CI) and a Waters Q-TOF Premier (ESI) spectrometer. IR spectra were recorded on a Bruker Tensor 27 spectrometer equipped with a "GoldenGate" diamond ATR unit. Elemental analyses were determined with a Euro EA-CHNS instrument from HEKAtech and optical rotations with a Perkin–Elmer polarimeter 343. GCMS retention time experiments on petroleum hydrocarbons, both alone and in admixture with synthetic standards, were conducted in multiple reaction monitoring (MRM) mode and targeting the compound-specific 260 Da molecular ion to 203 Da fragment transition. We used a Micromass AutoSpec Ultima mass spectrometer interfaced to an Agilent 6890 N gas chromatograph and the GC was fitted with a fused silica capillary column (60 m; 0.25 mm i.d.; 0.25 μm film thickness; J&W Scientific) with He as carrier gas. The phases tested were DB-5MS, DB-1MS and DB-XLB (30 m for this column) and identity was assured by co-elution on all three. The GC temperature program was: 60 (2 min) to 150 °C at 10 K min^{-1}, to 330 °C (held 19 min) at 3 K min^{-1}. The AutoSpec source was operated in electron ionization (EI, 70 eV) mode at 250 °C, with 8 kV accelerating voltage. Additional full scan analyses were conducted over a range of *m/z*: 50–600. Data were acquired and processed using MassLynx 4.0 (Micromass Ltd.).

17-Methylene-5α-androstane (6)

A solution of *n*BuLi (46.8 mmol, 18.7 mL, 2.5 mol L^{-1} in hexane) was slowly added to a suspension of $[Ph_3PMe]Br$ (16.9 g, 46.8 mmol) in abs. THF (200 mL) under N_2 at −10 °C. The reaction mixture was stirred for 10 min at 23 °C. A solution of ketone **5** (4.28 g, 15.6 mmol) in abs. THF (100 mL) was then added at 23 °C. After heating of the mixture to 66 °C for 16 h, water (100 mL) was added. The layers were separated and extracted with MTBE (3× 100 mL). The combined organic layers were dried ($MgSO_4$), filtered, and the solvent was evaporated. The crude product was purified by column chromatography (SiO_2, hexane, R_f = 0.75) to give olefin **6** (3.97 g, 14.6 mmol, 94%) as colorless crystals. M.p. 66 °C; $[\alpha]_D^{20}$ = +10 (CH_2Cl_2, 1 g L^{-1}) (lit. m.p. 69–70 °C, $[\alpha]_D^{22}$ = +12 (0.7 g L^{-1})].[20] ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.66–0.72 (m, 1H), 0.77 (s, 3H), 0.80 (s, 3H), 0.86–1.07 (m, 4H), 1.16–1.31 (m, 8H), 1.36–1.45 (m, 2H), 1.48–1.53 (m, 1H), 1.61–1.73 (m, 5H), 1.79 (dt, *J* = 12.7, 3.0 Hz, 1H), 2.22 (qt, *J* = 8.7, 1.8 Hz, 1H), 2.47–2.50 (m, 1H), 4.61–4.62 ppm (m, 2H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ = 12.25 (CH_3), 18.56 (CH_3), 20.71 (CH_2), 22.20 (CH_2), 24.12 (CH_2), 26.82 (CH_2), 29.03 (CH_2), 29.07 (CH_2), 29.41 (CH_2), 32.04 (CH_2), 35.50 (CH), 35.80 (CH_2), 36.40 (C), 38.72 (CH_2), 44.15 (C), 47.11 (CH), 54.49 (CH), 54.99 (CH), 100.50 (C), 162.19 ppm (CH_2); IR (ATR): $\tilde{\nu}$ = 3065 (w), 2967 (m), 2921 (s), 2849 (m), 1652 (m), 1467 (m), 1449 (m), 873 cm^{-1} (s); MS (EI, 70 eV): *m/z* (%): 272 (100) [M^+], 257 (100), 216 (40), 215 (17), 202 (16), 189 (13), 175 (12), 163 (63), 162 (96), 149 (34), 147 (38), 135 (25), 133 (18), 121 (26), 109 (41), 108 (67), 107 (49), 95 (34), 93 (39), 91 (35), 81 (51), 79 (34), 67 (32), 55 (24); HRMS (EI): calcd 272.2499 (for $C_{20}H_{32}$); found: 272.2503 [M^+]; 272.48 ($C_{20}H_{32}$).

17β-(Hydroxymethyl)-5α-androstane (7)

A solution of 9-BBN (43 mmol, 86 mL, 0.5 mol L^{-1} in THF) was added to a cooled (ice/water bath) solution of alkene **6** (3.90 g, 14.3 mmol) in abs. THF (100 mL). The reaction mixture was heated to 66 °C for 3 h. After cooling to 0 °C (ice/water bath) a solution of NaOH (143 mmol, 47.0 mL, 3 mol L^{-1}) and H_2O_2 (143 mmol, 16.0 mL, 30% in H_2O) was added and the reaction mixture was stirred vigorously for 16 h at ambient temperature. The layers were separated and extracted with MTBE (2×100 mL). The combined organic layers were dried ($MgSO_4$), filtered, and the solvent was evaporated. The crude product was purified by column chroma-

tography (SiO_2, hexane/MTBE = 3:1, R_f = 0.26) to yield alcohol **7** (4.02 g, 13.8 mmol, 97%) as colorless crystals. M.p. 143 °C. $[\alpha]_D^{20}$ = +6.7 (CH_2Cl_2, 1 g L^{-1}) (lit. mp.: 145–146 °C. $[\alpha]_D^{20}$ = +6.3 ($CHCl_3$, 0.5 g L^{-1})).[21] ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.59 (s, 3H), 0.65 (ddd, *J* = 14.7, 10.6, 4.0 Hz, 1H), 0.74 (s, 3H), 0.79–0.90 (m, 2H), 0.95–1.01 (m, 2H), 1.04–1.24 (m, 10H), 1.28–1.40 (m, 2H), 1.42–1.50 (m, 2H), 1.53–1.64 (m, 5H), 1.72–1.82 (m, 2H), 3.48 (dd, *J* = 10.5, 7.6 Hz, 1H), 3.66 ppm (dd, *J* = 10.5, 6.8 Hz, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ = 12.24 (CH_3), 12.63 (CH_3), 20.54 (CH_2), 22.19 (CH_2), 24.45 (CH_2), 25.28 (CH_2), 26.82 (CH_2), 29.06 (CH_2), 29.07 (CH_2), 32.25 (CH_2), 35.27 (CH), 36.35 (C), 38.74 (CH_2), 38.95 (CH_2), 41.95 (C), 47.12 (CH), 53.15 (CH), 55.07 (CH), 56.16 (CH), 64.70 ppm (CH_2); IR (ATR): $\tilde{\nu}$ = 3378 (m), 2969 (m), 2937 (s), 2913 (s), 2868 (s), 2853 (s), 1469 (m), 1447 (m), 1002 cm^{-1} (s); MS (EI, 70 eV): *m/z* (%): 290 (38) [M^+], 275 (42), 257 (28), 235 (9), 232 (23), 218 (79), 217 (100), 203 (14), 189 (7), 180 (10), 162 (20), 149 (55), 135 (19), 121 (22), 109 (32), 95 (35), 81 (36), 67 (30), 55 (30); HRMS (EI): calcd 290.2604 (for $C_{19}H_{34}O$); found: 290.2607 [M^+]; 290.49 ($C_{20}H_{34}O$).

Barton reaction of alcohol 7

A solution of alcohol **7** (500 mg, 1.72 mmol) in $CHCl_3$ (17 mL) was treated with *tert*-butyl nitrite (2.00 mL, 1.77 g, 17.2 mmol) and the mixture was stirred for 30 min in the dark. All volatile materials were evaporated and the crude solid was dried in high vacuum for 3 h. The residue was dissolved in anhydrous acetone (350 mL, 22 mol L^{-1}). Portions of 80 mL each were placed successively in a bubble column reactor with an inner 150 W Hg-lamp and an outer cooling jacket and were irradiated for 20 min, while a laminar stream of N_2 was passed slowly through the solution and the temperature was maintained at 20 °C. The colorless precipitate was filtered off to give a first portion of nitroso compound **3** (176 mg, 0.55 mmol, 32%). The combined filtrates were concentrated and submitted to column chromatography (SiO_2, hexane/MTBE = 3:1). In the first fraction, aldehyde **9** (35 mg, 0.12 mmol, 7%; R_f = 0.58) was obtained as a colorless oil. Secondly, starting material **7** (143 mg, 0.49 mmol, 29%; R_f = 0.26) was recovered. The third fraction contained oxime **8** (30 mg, 0.09 mmol, 5%; R_f = 0.14) as colorless crystals. Finally, a second portion of nitroso compound **3** (148 mg, 0.46 mmol, 27%; R_f = 0.08) was received in the fourth fraction.

17β-(Hydroxymethyl)-18-nitroso-5α-androstane (3)

M.p. 151 °C; $[\alpha]_D^{20}$ = +40.0 (CH_2Cl_2, 1 g L^{-1}); ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.70–0.75 (m, 1H), 0.79 (s, 3H), 0.83–0.99 (m, 4H), 1.01–1.05 (m, 1H), 1.17–1.25 (m, 7H), 1.30–1.48 (m, 4H), 1.53–1.59 (m, 2H), 1.65–1.79 (m, 4H), 1.88–1.95 (m, 1H), 2.28 (dt, *J* = 13.7, 2.6 Hz, 1H), 3.39 (d, *J* = 13.9 Hz, 1H), 3.66–3.71 (m, 2H), 3.76–3.79 (m, 1H), 5.46 ppm (d, *J* = 13.9 Hz, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ = 12.44 (CH_3), 20.74 (CH_2), 22.04 (CH_2), 23.50 (CH_2), 23.76 (CH_2), 26.71 (CH_2), 28.80 (CH_2), 28.86 (CH_2), 31.76 (CH_2), 34.29 (CH_2), 35.66 (CH), 36.33 (C), 38.82 (CH_2), 46.85 (CH), 47.21 (C), 52.86 (CH), 54.32 (CH_2), 54.62 (CH), 57.77 (CH), 64.02 ppm (CH_2); IR (ATR): $\tilde{\nu}$ = 3384 (m), 2913 (s), 2850 (s), 1465 (m), 1445 (m), 1381 (m), 1227 (m), 1198 (m), 1177 (s), 1050 (m), 1026 (m), 1009 cm^{-1} (m); MS (ESI+): *m/z*: 320 [*M*+H^+], 342 [*M*+Na^+]; HRMS (ESI+): calcd 342.2404 (for $C_{20}H_{33}NNaO_2$); found: 342.2410 [*M*+Na^+]; 319.49 ($C_{20}H_{33}NO_2$).

17β-(Hydroxymethyl)-18-oximino-5α-androstane (8)

Small amounts were obtained from the Barton reaction. It was also prepared by the following isomerization: A solution of nitroso compound **3** (324 mg, 1.01 mmol) in *i*PrOH (50 mL) was heated to reflux for 2 h. The solvent was removed in vacuum to give oxime **8** (324 mg, 1.01 mol, quant.) as colorless crystals. M.p. 140 °C; $[\alpha]_D^{20}$ = +17.0 (CH_2Cl_2, 1 g L^{-1}); ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.72 (s, 3H), 0.74–0.79 (m, 1H), 0.83–0.89 (m, 1H), 0.91–0.97 (m, 1H), 0.99–1.05 (m, 1H), 1.14–1.26 (m, 6H), 1.27–1.43 (m, 6H), 1.46–1.50 (m, 1H), 1.57 (dq, *J* = 13.4, 3.7 Hz, 1H), 1.62–1.71 (m, 3H), 1.78–1.85 (m, 2H), 1.91–1.97 (m, 1H), 2.42 (dt, *J* = 12.1, 3.1 Hz, 1H), 2.82 (br s, 1H), 3.55–3.62 (m, 2H), 7.36 (s, 1H), 8.46 ppm (br s, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ = 12.21 (CH_3), 21.21 (CH_2), 22.09 (CH_2), 24.47 (CH_2), 24.77 (CH_2), 26.74 (CH_2), 28.74 (CH_2), 28.97 (CH_2), 32.05 (CH_2), 35.90 (CH_2), 36.30 (C), 36.74 (CH), 38.65 (CH_2), 46.72 (CH), 49.71 (C), 52.30 (CH), 54.86 (CH), 56.33 (CH), 63.97 (CH_2), 153.96 ppm (CH); IR (ATR): $\tilde{\nu}$ = 3320 (m), 2919 (s), 2852 (s), 1446 (m), 1034 (m), 1012 cm^{-1} (m); MS (ESI+): *m/z*: 320 [*M*+H^+], 342 [*M*+Na^+]; HRMS (ESI): *m/z*: calcd 342.2404 (for $C_{20}H_{33}NNaO_2$); found: 342.2413 [*M*+Na^+]; 319.49 ($C_{20}H_{33}NO_2$).

5α-Androstan-17β-carboxaldehyde (9)

M.p. 106 °C; $[\alpha]_D^{20}$ = 12.4 (CH_2Cl_2, 2.7 g L^{-1}); ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.68–0.77 (m, 1H), 0.73 (s, 3H), 0.78 (s, 3H), 0.85–0.94 (m, 2H), 1.00–1.06 (m, 1H), 1.08–1.15 (m, 1H), 1.18–1.27 (m, 7H), 1.35–1.42 (m, 3H), 1.46–1.52 (m, 1H), 1.59 (ddd, *J* = 13.5, 7.0, 3.8 Hz, 1H), 1.64–1.75 (m, 5H), 1.96 (dt, *J* = 12.3, 3.3 Hz, 1H), 2.05–2.14 (m, 1H), 2.26–2.29 (m, 1H), 9.67 ppm (d, *J* = 2.1 Hz, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ = 12.21 (CH_3), 13.96 (CH_3), 20.29 (CH_2), 20.97 (CH_2), 22.12 (CH_2), 24.79 (CH_2), 26.74 (CH_2), 28.91 (CH_2), 28.98 (CH_2), 32.11 (CH_2), 35.05 (CH), 36.32 (C), 38.57 (CH_2), 38.67 (CH_2), 44.96 (C), 47.00 (CH), 54.72 (CH), 56.39 (CH), 63.00 (CH), 205.24 ppm (CH); IR (ATR): $\tilde{\nu}$ = 2923 (s), 2849 (m), 1718 (vs), 1446 cm^{-1} (m); MS (EI, 70 eV): *m/z* (%): 288 (10) [M^+], 273 (8), 255 (8), 243 (5), 231 (5), 217 (10), 203 (5), 189 (3), 175 (8), 162 (7), 149 (22), 135 (43), 133 (16), 119 (16), 109 (44), 106 (10), 95 (70), 91 (51), 81 (81), 77 (31), 67 (100), 53 ppm (28); HRMS (EI): calcd 288.2448 (for $C_{20}H_{32}O$); found: 288.2453 [M^+]; 288.48 ($C_{20}H_{32}O$).

17β-(Hydroxymethyl)-5α-androstano-18-nitrile (10)

2-Chloro-1-methylpyridinium iodide (2.15 g, 8.43 mmol) was added to a solution of oxime **8** (2.07 g, 6.48 mmol) in CH_2Cl_2 (40 mL). After stirring the mixture for 10 min, NEt_3 (1.17 mL, 853 mg, 8.43 mmol) was added and the resulting mixture was heated to 40 °C for 24 h. After cooling to ambient temperature, the solvent was evaporated. The residue was column chromatographed (SiO_2, hexane/EtOAc = 2:1, R_f = 0.37) to furnish nitrile **10** (1.24 g, 4.11 mmol, 63%) as colorless crystals. M.p. 177 °C; $[\alpha]_D^{20}$ = +10 (CH_2Cl_2, 1 g L^{-1}); ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.75 (ddd, *J* = 12.1, *J* = 10.7, *J* = 3.7 Hz, 1H), 0.81 (s, 3H), 0.87 (td, *J* = 13.3, *J* = 4.7 Hz, 1H), 0.92–0.98 (m, 1H), 0.99–1.06 (m, 1H), 1.18–1.35 (m, 7H), 1.39–1.51 (m, 5H), 1.59 (qd, *J* = 10.9, 4.0 Hz, 1H), 1.64–1.69 (m, 2H), 1.71–1.91 (m, 5H), 1.94–2.02 (m, 1H), 2.37 (dt, *J* = 12.9, 3.2 Hz, 1H), 3.78 (dd, *J* = 10.8, 6.2 Hz, 1H), 3.87 ppm (dd, *J* = 10.8, 8.3 Hz, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ = 12.18 (CH_3), 22.01 (CH_2), 22.18 (CH_2), 24.85 (CH_2), 25.43 (CH_2), 26.64 (CH_2), 28.48 (CH_2), 28.84 (CH_2), 31.44 (CH_2), 34.61 (CH_2), 36.21 (C), 38.21 (CH), 38.67 (CH_2), 46.75 (CH), 49.91 (CH), 50.04 (C), 53.78 (CH), 55.62 (CH), 64.43 (CH_2), 121.27 ppm (C); IR (ATR): $\tilde{\nu}$ = 3472 (m), 2917 (s), 2851 (s), 2224 (w), 1445 (m), 1381 (s), 1042 (m), 1030 (m), 1015 (m), 1003 cm^{-1} (m); MS (EI): *m/z* (%): 301 (40) [M^+], 286 (42), 259 (7), 245 (27), 244 (61), 217 (9), 216 (7), 199 (4), 165 (5), 147 (6), 133 (7), 110 (100), 95 (26), 81 (23), 67 (19), 55 (17); HRMS (ESI+): calcd 324.2298 (for $C_{20}H_{31}NNaO$); found: 324.2309 [*M*+Na^+]; 301.47 ($C_{20}H_{31}NO$).

17β-[(2-Nitrophenyl)selenomethyl]-5α-androstano-18-nitrile (11)

2-Nitrophenyl selenocyanate (1.50 g, 6.60 mmol) and tributylphosphane (1.63 mL, 1.34 g, 6.60 mmol) were added successively to a solution of alcohol **10** (1.24 g, 4.13 mmol) in THF (40 mL). The reaction mixture was heated to 66 °C for 3 h and then cooled to ambient temperature. The solvent was evaporated and the residue column chromatographed (SiO_2, hexane/MTBE = 5:1, R_f = 0.30) to give the selenoether **11** (1.84 g, 3.80 mmol, 92%) as yellow crystals. M.p. 170–174 °C; $[\alpha]_D^{20} = -25$ (CH_2Cl_2, 1 g L^{-1}); ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.73–0.78 (m, 1H), 0.82 (s, 3H), 0.88 (td, J = 13.1, 4.1 Hz, 1H), 0.93–0.99 (m, 1H), 1.00–1.06 (m, 1H), 1.18–1.33 (m, 7H), 1.38–1.69 (m, 8H), 1.75 (dq, J = 10.4, J = 3.1 Hz, 1H), 1.84–1.90 (m, 2H), 2.01 (qd, J = 9.4, 4.7 Hz, 1H), 2.20–2.11 (m, 1H), 2.35 (dt, J = 12.7, 3.2 Hz, 1H), 2.97–3.04 (m, 1H), 3.23 (dd, J = 10.8, 4.7 Hz, 1H), 7.33 (ddd, J = 8.2, 6.9, 1.5 Hz, 1H), 7.50–7.56 (m, 2H), 8.28 ppm (dd, J = 8.3, 1.1 Hz, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ = 12.18 (CH_3), 22.00 (CH_2), 22.16 (CH_2), 25.35 (CH_2), 26.61 (CH_2), 27.72 (CH_2), 28.42 (CH_2), 28.81 (CH_2), 29.31 (CH_2), 31.31 (CH_2), 33.45 (CH_2), 36.22 (C), 38.56 (CH), 38.66 (CH_2), 46.65 (CH), 46.74 (CH), 52.02 (C), 53.68 (CH), 55.26 (CH), 120.80 (C), 125.53 (CH), 126.43 (CH), 128.91 (CH), 133.22 (C), 133.73 (CH), 146.67 ppm (C); IR (ATR): $\tilde{\nu}$ = 3089 (w), 2921 (m), 2846 (m), 2230 (w), 1588 (m), 1562 (m), 1502 (s), 1463 (m), 1449 (m), 1328 (s), 1302 (s), 1025 (m), 1099 (m), 851 (m), 730 (s), 701 cm^{-1} (m); MS (ESI+): m/z: 509 [M+Na^+]; HRMS (EI): calcd 509.1678 (for $C_{26}H_{34}N_2NaO_2Se$); found: 509.1685 [M+Na^+]; 485.53 ($C_{26}H_{34}N_2O_2Se$).

17-Methylene-5α-androstano-18-nitrile (12)

H_2O_2 (4.31 mL, 38.0 mmol, 30% in H_2O) was slowly added to a cooled (ice/water bath) solution of selenoether **11** (1.84 g, 3.80 mmol) in THF (60 mL). The reaction mixture was heated to 66 °C for 4 h, cooled to ambient temperature, and then treated with sat. $NaHCO_3$ soln (30 mL). The layers were separated and extracted with MTBE (3×30 mL). The combined organic layers were dried ($MgSO_4$) and concentrated after filtration. The residue was purified by column chromatography (SiO_2, hexane/MTBE = 5:1, R_f = 0.63) to yield olefin **12** (1.01 g, 3.56 mmol, 94%) as colorless crystals. M.p. 104 °C; $[\alpha]_D^{20} = -58$ (CH_2Cl_2, 1 g L^{-1}). ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.75 (ddd, J = 12.3, 10.5, 3.7 Hz, 1H), 0.84 (s, 3H), 0.89 (td, J = 12.9, 4.2 Hz, 1H), 0.96–1.07 (m, 2H), 1.14 (ddd, J = 12.6, 10.7, 6.1 Hz, 1H), 1.21–1.27 (m, 4H), 1.28–1.34 (m, 2H), 1.38–1.47 (m, 1H), 1.50–1.59 (m, 3H), 1.64–1.72 (m, 3H), 1.80 (dq, J = 12.6, 3.7 Hz, 1H), 1.85–1.93 (m, 2H), 2.29–2.37 (m, 2H), 2.64 (ddq, J = 17.5, 10.0, 2.3 Hz, 1H), 4.98–5.01 ppm (m, 2H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ = 12.18 (CH_3), 22.05 (2 CH_2), 25.49 (CH_2), 26.66 (CH_2), 28.50 (CH_2), 28.88 (CH_2), 29.17 (CH_2), 31.22 (CH_2), 32.19 (CH_2), 36.31 (C), 38.19 (CH), 38.73 (CH_2), 46.86 (CH), 48.87 (C), 53.82 (CH), 55.21 (CH), 107.73 (CH_2), 121.18 (C), 150.72 ppm (C); IR (ATR): $\tilde{\nu}$ = 3071 (w), 2963 (w), 2948 (m), 2925 (s), 2850 (m), 2221 (w), 1445 (s), 897 cm^{-1} (s); MS (EI, 70 eV): m/z (%): 283 (4) [M^+], 268 (5), 226 (11), 198 (4), 171 (4), 159 (4), 146 (10, 131 (10), 117 (8), 110 (100), 95 (33), 91 (33), 81 (36), 79 (31), 67 (43), 55 (38); HRMS (EI): calcd 283.2295 (for $C_{20}H_{29}N$); found: 283.2289 [M^+]; 283.46 ($C_{20}H_{29}N$).

17-(Hydroxymethyl)-17-hydroxy-5α-androstano-18-nitrile (15)

Potassium osmate(VI) dihydrate (26 mg, 70 μmol) and NMO (408 mg, 3.49 mmol) were successively added to a solution of alkene **12** (990 mg, 3.49 mmol) in a mixture of THF (5 mL), acetone (5 mL), H_2O (5 mL), and *tert*-butyl alcohol (5 mL). The reaction mixture was stirred for 20 h at 23 °C and then a second portion of NMO (408 mg, 3.49 mmol) was added. After stirring for a further 20 h at 23 °C, the mixture was treated with sat. aqueous Na_2SO_3 soln. (40 mL). The layers were separated and extracted with MTBE (3×60 mL). The combined organic layers were dried ($MgSO_4$), filtered, and the solvent was evaporated. The crude product (1.04 g, 3.28 mg, 94%) was used at the next step (periodate cleavage) without further purification. M.p. 162 °C; $[\alpha]_D^{20} = -5.0$ (CH_2Cl_2, 1 g L^{-1}); ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.73–0.78 (m, 1H), 0.80 (s, 3H), 0.87 (td, J = 13.0, 4.2 Hz, 1H), 0.95–1.06 (m, 2H), 1.20–1.26 (m, 5H), 1.36–1.46 (m, 3H), 1.47–1.52 (m, 1H), 1.53–1.62 (m, 2H), 1.63–1.78 (m, 5H), 1.80–1.85 (m, 2H), 1.87–1.93 (m, 1H), 2.07 (dt, J = 12.7, 3.1 Hz, 1H), 2.99–3.19 (m, 2H), 3.69 (d, J = 11.1 Hz, 1H), 4.01 ppm (d, J = 11.1 Hz, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ = 12.13 (CH_3), 21.92 (CH_2), 21.96 (CH_2), 24.86 (CH_2), 26.57 (CH_2), 28.44 (CH_2), 28.79 (2 CH_2), 31.35 (CH_2), 33.94 (CH_2), 36.13 (C), 38.43 (CH), 38.66 (CH_2), 46.64 (CH), 50.95 (CH), 53.33 (CH), 54.05 (C), 67.02 (CH_2), 82.12 (C), 121.88 ppm (C); IR (ATR): $\tilde{\nu}$ = 3498 (m), 3379 (m), 3260 (m), 2966 (m), 2923 (s), 2856 (m), 2235 (w), 1465 (m), 1450 (m), 1379 (m), 1361 (m), 1303 (m), 1285 (m), 1275 (m), 1056 (m), 1041 (m), 1026 cm^{-1} (m); MS (ESI+): m/z: 318 [M+H^+], 340 [M+Na^+], 356 [M+K^+]; HRMS (ESI): calcd 340.2247 (for $C_{20}H_{31}NNaO_2$); found: 340.2247 [M+Na^+]; 317.47 $C_{20}H_{31}NO_2$.

17-Oxo-5α-androstano-18-nitrile (14)

Sodium periodate (3.51 g, 16.4 mmol) was added to a solution of dihydroxy compound **15** (1.04 g, 3.28 mmol) in a mixture of THF (25 mL) and H_2O (5 mL). The mixture was heated to reflux for 4 h, cooled to ambient temperature, and treated with brine (50 mL). The layers were separated and extracted with MTBE (3×50 mL). The combined organic layers were dried ($MgSO_4$), filtered, and the solvent was removed in vacuum to give ketone **14** (864 mg, 3.03 mmol, 92%) as colorless crystals. M.p. 129 °C; $[\alpha]_D^{20} = +56.7$ (CH_2Cl_2, 1 g L^{-1}) [lit. m.p. 139–141 °C, $[\alpha]_D^{20} = +42$ ($CHCl_3$, 0.3 g L^{-1})].[22] ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.76 (ddd, J = 12.3, 10.5, 3.7 Hz, 1H), 0.83 (s, 3H), 0.89 (td, J = 13.1, 4.2 Hz, 1H), 0.95–1.08 (m, 2H), 1.17–1.46 (m, 9H), 1.48–1.54 (m, 1H), 1.64–1.69 (m, 2H), 1.75–1.92 (m, 4H), 2.10–2.15 (m, 1H), 2.20–2.29 (m, 2H), 2.66 ppm (ddd, J = 19.9, 9.0, 0.5 Hz, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ = 12.10 (CH_3), 21.68 (CH_2), 21.93 (CH_2), 23.01 (CH_2), 26.63 (CH_2), 28.18 (CH_2), 28.76 (CH_2), 28.86 (CH_2), 30.15 (CH_2), 36.00 (CH_2), 36.31 (C), 38.30 (CH), 38.59 (CH_2), 46.71 (CH), 52.08 (CH), 52.33 (C), 53.60 (CH), 116.72 (C), 207.19 ppm (C); IR (ATR): $\tilde{\nu}$ = 2981 (w), 2958 (w), 2921 (s), 2880 (m), 2853 (m), 2230 (w), 1758 (s), 1446 cm^{-1} (m); MS (EI, 70 eV): m/z (%): 285 (18) [M^+], 270 (15), 242 (5), 228 (19), 214 (4), 200 (6), 175 (5), 158 (5), 121 (15), 110 (58), 109 (100), 95 (47), 81 (40), 67 (49); HRMS (ESI): calcd 308.1985 (for $C_{19}H_{27}NNaO$); found: 308.1986 [M^+]; 285.43 ($C_{19}H_{27}NO$).

17,17-(1,2-Ethylenedithio)-5α-androstano-18-nitrile (13)

A solution of ketone **14** (915 mg, 3.21 mmol), 1,2-ethanedithiol (0.35 mL, 390 mg, 4.2 mmol), and *p*-TosOH·H_2O (25 mg, 0.13 mmol) in benzene (60 mL) was heated under reflux for 16 h in a Dean–Stark trap. After cooling to ambient temperature, the reaction mixture was diluted with H_2O (100 mL) and the layers were separated. The organic layer was dried ($MgSO_4$), filtered, and the solvent was evaporated. The crude product was recrystallized from hexane (ca. 10 mL) to give thioacetal **13** (724 mg, 2.00 mmol, 62%) as colorless crystals. M.p. 172 °C; $[\alpha]_D^{20} = -67.7$ (CH_2Cl_2, 1 g L^{-1}); ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.76 (ddd, J = 12.7, 9.9, 3.5 Hz, 1H), 0.81 (s, 3H), 0.89 (td, J = 13.0, 4.2 Hz, 1H), 0.93–1.07 (m, 2H), 1.17–1.27 (m, 5H), 1.37–1.46 (m, 2H), 1.48–1.59 (m, 4H), 1.66–1.70 (m, 2H), 1.72–

1.80 (m, 2H), 1.82–1.89 (m, 2H), 2.05 (dt, $J=12.5$, 3.4 Hz, 1H), 2.36–2.42 (m, 1H), 2.73–2.79 (m, 1H), 3.12–3.17 (m, 1H), 3.25–3.36 ppm (m, 3H); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): $\delta=12.18$ (CH_3), 21.72 (CH_2), 22.01 (CH_2), 24.37 (CH_2), 26.64 (CH_2), 28.45 (CH_2), 28.82 (CH_2), 28.95 (CH_2), 31.08 (CH_2), 36.22 (C), 38.70 (CH_2), 39.41 (CH), 39.68 (CH_2), 40.82 (CH_2), 41.04 (CH_2), 46.76 (CH), 52.44 (CH), 53.24 (CH), 59.08 (C), 75.40 (C), 121.26 ppm (C); IR (ATR): $\tilde{\nu}=2967$ (m), 2919 (s), 2874 (m), 2857 (m), 2840 (m), 2226 (w), 1467 (m), 1458 (m), 1441 (m), 1421 (m), 850 (m), 822 cm^{-1} (m); MS (ESI+): *m/z*: 362 [*M*+H^+], 384 [*M*+Na^+], 400 [*M*+K^+]; HRMS (ESI+): calcd 384.1790 (for $C_{21}H_{31}NNaS_2$); found: 384.1790 [*M*+Na^+]; 361.61 ($C_{21}H_{31}NS_2$).

5α-Androstano-18-nitrile (16)

Dithioacetal **13** (300 mg, 0.83 mmol) was added to a suspension of freshly prepared W2-Raney nickel[14] (5 g, 50% Al, 50% Ni) in abs. EtOH (40 mL). The reaction mixture was heated to 100 °C for 3.5 h. After cooling to ambient temperature, the suspension was filtered through Al_2O_3 (2 cm, washed with 100 mL EtOH). The filtrate was concentrated in vacuum and the residue column chromatographed (SiO_2, hexane/MTBE = 20:1, $R_f=0.38$) to yield nitrile **16** (138 mg, 0.51 mmol, 61%) as colorless crystals. M.p. 99 °C; $[\alpha]_D^{20}=+8.3$ (CH_2Cl_2, 1 g L^{-1}); ^1H NMR (500 MHz, $CDCl_3$): $\delta=0.74$ (ddd, $J=12.3$, 10.7, 3.7 Hz, 1H), 0.82 (s, 3H), 0.88 (td, $J=12.9$, 4.3 Hz, 1H), 0.94–1.04 (m, 2H), 1.09 (ddd, $J=12.2$, 11.1, 6.7 Hz, 1H), 1.18–1.27 (m, 6H), 1.39–1.60 (m, 6H), 1.66–1.70 (m, 2H), 1.72–1.93 (m, 5H), 2.13 (ddd, $J=12.6$, 8.6, 1.9 Hz, 1H), 2.21 ppm (dt, $J=12.9$, 3.3 Hz, 1H); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): $\delta=12.20$ (CH_3), 20.62 (CH_2), 22.05 (CH_2), 22.36 (CH_2), 26.36 (CH_2), 26.67 (CH_2), 28.87 (CH_2), 31.71 (CH_2), 34.40 (CH_2), 36.26 (CH_2), 36.26 (C), 36.66 (CH_2), 38.44 (CH), 38.73 (CH_2), 46.76 (CH), 46.83 (C), 53.80 (CH), 55.07 (CH), 123.44 ppm (C); IR (ATR): $\tilde{\nu}=2951$ (m), 2923 (s), 2869 (m), 2854 (s), 2225 (w), 1444 (m) cm^{-1}; MS (EI, 70 eV): *m/z* (%): 271 (7) [M^+], 256 (7), 215 (6), 214 (17), 179 (2), 165 (3), 151 (5), 149 (6), 137 (7), 125 (11), 123 (11), 110 (42), 97 (35), 95 (33), 85 (54), 84 (82), 71 (33), 69 (39), 57 (53), 55 (44), 49 (100); HRMS (EI): calcd 271.2295 (for $C_{19}H_{29}N$); found: 271.2298 [M^+]; 271.45 ($C_{19}H_{29}N$).

18-Amino-5α-androstane (17)

$LiAlH_4$ (97 mg, 2.6 mmol) was added at 23 °C to a stirred solution of abs. THF (6 mL). A solution of nitrile **16** (138 mg, 0.51 mmol) in THF (6 mL) was added dropwise. The reaction mixture was heated to 66 °C for 20 h. After cooling to 0 °C (ice/water bath) the mixture was treated with $MgSO_4 \cdot 7H_2O$ and stirred for 30 min. The suspension was filtered through $MgSO_4$ (4 cm, washed with MTBE (150 mL) and the solvent was removed under vacuum. Amine **17** (134 mg, 0.49 mmol, 95%) was obtained as colorless crystals. M.p. 83 °C; $[\alpha]_D^{20}=+23$ (CH_2Cl_2, 1 g L^{-1}); ^1H NMR (500 MHz, $CDCl_3$): $\delta=$ 0.72 (ddd, $J=12.4$, 10.8, 4.2 Hz, 1H), 0.78 (s, 3H), 0.84–0.94 (m, 4H), 1.00–1.06 (m, 1H), 1.08–1.27 (m, 9H), 1.32–1.45 (m, 3H), 1.47–1.51 (m, 1H), 1.55 (dq, $J=13.6$, 3.9 Hz, 1H), 1.59–1.71 (m, 6H), 1.74–1.79 (m, 1H), 2.04 (dt, $J=13.2$, 3.3 Hz, 1H), 2.24 (d, $J=13.1$ Hz, 1H), 2.68 ppm (d, $J=13.1$ Hz, 1H); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): $\delta=$ 12.21 (CH_3), 20.22 (CH_2), 20.43 (CH_2), 22.14 (CH_2), 25.22 (CH_2), 26.76 (CH_2), 28.97 (CH_2), 28.99 (CH_2), 32.64 (CH_2), 32.97 (CH_2), 34.44 (CH_2), 34.95 (CH), 36.35 (C), 38.67 (CH_2), 39.23 (CH_2), 45.25 (C), 47.04 (CH), 54.97 (CH), 55.15 ppm (CH); IR (ATR): $\tilde{\nu}=3320$ (br w), 2918 (s), 2850 cm^{-1} (s); MS (EI, 70 eV): *m/z* (%): 275 (7) [M^+], 258 (3), 245 (52), 244 (100), 229 (31), 189 (5), 163 (14), 149 (57), 135 (49), 134 (25), 121 (15), 109 (34), 95 (28), 81 (34), 67 (28), 55 (20); HRMS (EI): calcd 275.2608 (for $C_{19}H_{33}N$); found: 276.2612 [M^+]; 275.48 ($C_{19}H_{33}N$).

13β,14β-Methano-18-*nor*-5α-androstane (18)

An aqueous solution of sodium nitrite (7.8 mmol, 3.0 mL, 2.5 mol L^{-1}) was added to a vigorously stirred solution of amine **17** (107 mg, 0.388 mmol) in a mixture of AcOH (8 mL) and CH_2Cl_2 (1.6 mL). The reaction mixture was stirred at 23 °C for 2 h and then diluted with water (20 mL). The solution was extracted with hexane (3×10 mL) and the combined organic layers were washed with sat. $NaHCO_3$ soln (3×50 mL, in water). After filtration through Al_2O_3 (washing with hexane), the solvent was removed under vacuum to yield cyclopropane derivative **18** along with D-*homo*-olefins **19a** and **19b** (58 mg, 22 μmol, 57%) in a ratio of 92:4:3 (**18**/**19a**/**19b**). The cyclopropane derivative **18** (15 mg, 0.06 mmol, 15%) was partly separated by column chromatography (SiO_2, hexane, $R_f=0.71$) as a colorless oil. $[\alpha]_D^{20}=19.8$ (CH_2Cl_2, 1 g L^{-1}); ^1H NMR (500 MHz, $CDCl_3$): $\delta=0.05$ (d, $J=4.4$ Hz, 1H; 18-H), 0.44 (d, $J=4.4$ Hz, 1H; 18-H), 0.54 (td, $J=11.4$, 2.0 Hz, 1H), 0.66 (s, 3H), 0.73 (qd, $J=12.6$, 4.3 Hz, 1H), 0.83 (td, $J=12.9$, 4.4 Hz, 1H), 0.98–1.04 (m, 1H), 1.05–1.12 (m, 1H), 1.16–1.29 (m, 6H), 1.35 (dd, $J=12.7$, 4.0 Hz, 1H), 1.38–1.53 (m, 6H), 1.61–1.76 (m, 5H), 1.88 (dq, $J=12.3$, 3.3 Hz, 1H), 2.01 ppm (dt, $J=12.8$, 3.3 Hz, 1H); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): $\delta=12.31$ (CH_3, 19-C), 18.61 (CH_2, 18-C), 20.82 (CH_2), 21.61 (CH_2), 22.44 (CH_2), 27.26 (CH_2), 28.37 (C), 29.29 (CH_2), 29.99 (CH_2), 30.23 (CH_2), 30.36 (CH_2), 31.00 (C), 31.66 (CH_2), 35.97 (CH_2), 37.07 (C), 39.04 (CH_2), 40.49 (CH), 47.31 (CH), 52.00 ppm (CH); IR (ATR): $\tilde{\nu}=3049$ (w), 2919 (s), 2851 (s), 1447 cm^{-1} (m); MS (EI, 70 eV): *m/z* (%): 258 (31) [M^+], 243 (42), 215 (2), 202 (3), 201 (2), 188 (2), 187 (2), 176 (7), 162 (18), 149 (74), 135 (41), 121 (35), 108 (55), 95 (73), 94 (94), 91 (64), 81 (60), 79 (100), 67 (65), 55 (50); HRMS (EI): calcd 258.2342 (for $C_{19}H_{30}$); found 258.2347 [M^+]; 258.45 ($C_{19}H_{30}$).

Acid treatment of a mixture of 18, 19a, and 19b followed by hydrogenation

Trifluoroacetic acid (0.09 mL, 1.2 mmol) was slowly added to a solution of cyclopropane derivative **18** and D-*homo*-olefins **19a** and **19b** (15 mg, 58 μmol (ratio **18**/**19a**/**19b** = 92:4:3)) in $CHCl_3$ (3 mL) at −20 °C. The reaction mixture was stirred for 2.5 h at −20 °C and then diluted with sat. aqueous $NaHCO_3$ soln. (5 mL). The layers were separated and the solvent removed under vacuum. The crude product (10 mg) was dissolved in EtOAc (4 mL) and palladium on charcoal (1 mg, 0.9 μmol) was added. The suspension was degassed and hydrogenated (4 bar H_2) at 50 °C for 2 days. After filtration through SiO_2 the solvent was removed in vacuum. A mixture (10 mg, 38 μmol, 66%) of 14β-androstane (**20**), 13β,14α-D-*homo*-androstane (**2a**), and 13α,14α-D-homo-androstane (**2b**) was obtained in a ratio of 81:16:3 (**20**/**2a**/**2b**) as a colorless oil.

5α,14β-Androstane (20)

Trifluoroacetic acid (0.50 mL, 6.5 mmol) and triethylsilane (0.51 mL, 3.2 mmol) were successively added at −20 °C to a solution of cyclopropane derivative **18** (42 mg, 16 μmol; prior to that purified by column chromatography) in CH_2Cl_2 (10 mL). The reaction mixture was stirred for 2 h at −20 °C and then diluted with sat. aqueous $NaHCO_3$ soln. (20 mL). The layers were separated and the organic layer was washed with sat. aqueous $NaHCO_3$ solution (2×20 mL), dried ($MgSO_4$), and evaporated after filtration. The crude product was purified by column chromatography with silver nitrate impregnated silica gel (SiO_2/$AgNO_3$ (12%), hexane) to give *epi*-androstane **20** (33 mg, 13 μmol, 81%) as colorless crystals. M.p. 48 °C; $[\alpha]_D^{20}=$ +35.0 (CH_2Cl_2, 1 g L^{-1}); ^1H NMR (500 MHz, $CDCl_3$): $\delta=0.76$ (s, 3H), 0.83–0.92 (m, 3H), 0.96–1.04 (m, 1H), 0.99 (s, 3H), 1.06–1.71 ppm

(m, 22H); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): $\delta = 12.18$ (CH_3), 20.39 (CH_2), 21.21 (CH_2), 22.13 (CH_2), 24.54 (CH_3), 25.00 (CH_2), 26.92 (CH_2), 29.11 (CH_2), 29.43 (CH_2), 32.71 (CH_2), 32.78 (CH_2), 33.93 (CH), 36.21 (C), 38.89 (CH_2), 40.50 (C), 41.86 (CH_2), 46.59 (CH), 46.93 (CH), 51.05 ppm (CH); IR (ATR): $\tilde{\nu} = 2920$ (s), 2852 (s), 1446 (m), 1377 cm^{-1} (m); MS (EI, 70 eV): *m/z* (%): 260 (67) [M^+], 245 (78), 231 (7), 217 (13), 204 (49), 203 (100), 189 (19), 178 (8), 179 (10), 176 (9), 175 (13), 163 (13), 161 (13), 150 (9), 149 (32), 148 (25), 147 (16), 135 (58), 121 (34), 107 (35), 95 (54), 93 (29), 91 (19), 81 (49), 79 (30), 67 (39), 55 (22); HRMS (EI): calcd 260.2499 (for $C_{19}H_{32}$); found: 260.2493 [M^+]; 260.46 $C_{19}H_{32}$.

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