Steroids 78 (2013) 1339–1346

Contents lists available at ScienceDirect

Steroids

journal homepage: www.elsevier.com/locate/steroids

The regio- and stereo-selective reduction of steroidal 4-en-3-ones using $Na_2S_2O_4/NaHCO_3$ and $CuCl/NaBH_4$

Chunli Wang, Xiaoyu Chen, Yaoqing Huang, Jesse Yang [1], Ying Chen *

Department of Medicinal Chemistry, School of Pharmacy, Fudan University, Shanghai 201203, China

ARTICLE INFO

Article history:
Received 22 April 2013
Received in revised form 19 September 2013
Accepted 29 September 2013
Available online 12 October 2013

Keywords:
4-En-3-one
Conjugated reduction
Sodium dithionite
Cuprous chloride

ABSTRACT

This paper describes the regio- and stereoselective reduction of $\triangle^4$-3-keto moiety in certain steroids using $Na_2S_2O_4/NaHCO_3$ and $CuCl/NaBH_4$, respectively. Using either one of the two reduction agents in the reaction, the 17-substituents in the D ring were observed to have clearly influenced the stereoselective reduction of 4-ene in the A ring by the so-called conformational transmission effect. $Na_2S_2O_4/NaHCO_3$ regioselectively reduced C=C at 4-position of 17-substituted-androst-4-en-3-one derivatives to 5α-*H*-3-one as the main isomer. And as an extended application, Epiandrosterone (**11**) was further synthesized from androst-4-en-3,17-dione (**AD**) *via* four steps. The total yield from this was about 45%. In the presence of $CuCl/NaBH_4$, $\triangle^4$-3-keto conjugated reduction of 17-spirocyclic ethylene ketal protected androst-4-en-3-one derivatives mainly produced 3α-hydroxy-5β-*H* isomers, at a yield around 81%. Considering the scaffold configuration of 3α-hydroxy-5β-*H* moiety coincided with that of bile acid analogs, this selective reduction could also be used as an alternative method for the synthetic study of bile acids using **AD** and its derivatives, which are from the microorganism degradation of natural sterols, as the potential materials. Meanwhile, configurations of the reductive compounds **5b**, **6b**, **9**, **10** and **17e** were identified by X-ray diffraction.

1. Introduction

Finding a new natural resource for the semi-synthesis of steroidal drugs has been an important research topic. Since androst-4-en-3,17-dione (**AD**) and androsta-1,4-dien-3,17-dione (**ADD**, Fig. 1) have been obtained from the degradation of natural sterols by microorganisms with a relatively good yield, using **AD** and **ADD** as the starting materials to synthesize various steroidal drugs has attracted the attention of many organic synthetic chemists [1–5]. Because many sterols are available from industrial waste, the synthesis of valuable steroidal drugs from this resource will have great economic and scientific advantages. However, due to the lack of effective and highly stereoselective reduction method for 4-ene in the conjugated enone, synthesizing steroidal drugs using saturated A ring from **AD** and **ADD** is still a great challenge.

Although several hydrogenation catalysts were reported for the selective reduction of conjugated enone-steroids such as catalytic systems based on Pd, Cu/Al_2O_3, Pt/TiO_2, homogeneous rhodium catalysts and so on [6–10], most of them have no practical use in industrial manufacturing due to their high cost, harsh reaction condition or low stereo-selectivity, etc. In this paper, we explored two common reduction systems, $Na_2S_2O_4/NaHCO_3$ and $CuCl/NaBH_4$, to reduce steroidal 4-en-3-one and 1,4-dien-3-one.

Akamanchi [11] reported that **AD** could be reduced to 5α-androstane-3,17-dione with a yield of 88% by refluxing a solution of steroid in toluene with an aqueous solution of $Na_2S_2O_4/NaHCO_3$ in the presence of PTC (Aliquat 336). The high stereoselectivity, good yield, inexpensive reductant, mild condition and easy operation of this phase transformation reaction presented a strong attraction to us. However, when we repeated their work, the experiments resulted in a product that was a 2:1 mixture of 5α- and 5β-isomer. It was not a pure 5α-androstane-3,17-dione, as was expected. Since 5α- and 5β-androstane-3,17-dione isomers show very similar physical properties, including very close R_f values on thin-layer chromatography (TLC), similar chemical shifts on ^1H NMR spectra and same melting points, it was thus suspected that the author might not have separated those two isomers, and reported them as a single product in the literature.

Coupled with our attempt to synthesize Epiandrosterone from AD and to improve the stereoselectivity of 5α-isomer, six different 17-*O*-substituted cyanohydrins (**3a–c**, **4a–c**) and one 17-spirocyclic ethylene ketal **AD** (**14**) were prepared to conduct the stereoselective reduction of 4-ene using $Na_2S_2O_4/NaHCO_3$. The experiments showed that the yield and stereoselectivity of the reduction are closely related to 17-substituents. The highest ratio of 5α-:5β-isomer was 2.2:1, which was in agreement with that of other catalytic

* Corresponding author. Tel.: +86 021 51980116.
E-mail address: yingchen71@fudan.edu.cn (Y. Chen).
[1] Intern from Henry M. Gunn High School, Palo Alto, CA, USA.

0039-128X/$ - see front matter © 2013 Elsevier Inc. All rights reserved.
http://dx.doi.org/10.1016/j.steroids.2013.09.011

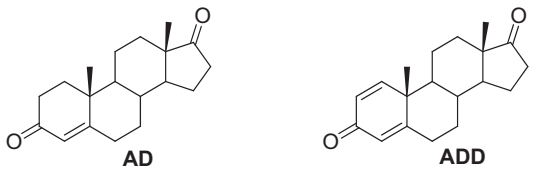


Fig. 1. The structures of **AD** and **ADD**.

reduction based on Pd [7]. Applying one of the 5α-isomers (**5b**) as key intermediates, Epiandrosterone was further synthesized with a yield of 92%.

Moreover, many literatures reported that a number of complex Cu–H reagents can reduce the C=C of α,β-unsaturated carbonyl compound selectively [12,13], but rarely for steroidal 4-en-3-one. $NaBH_4$–CuCl/MeOH is a Cu–H reagent formed *in situ*, which can selectively reduce the C=C of α,β-unsaturated esters but does not attack the ester group [14]. This inspired us to explore the C=C selective reduction of 4-en-3-one in **AD** and **ADD** derivatives under the $NaBH_4$–CuCl condition. Interestingly enough, with this reagent, both C=C and C=O of 4-en-3-one were directly saturated into 5β-*H*-3α-hydroxy as main isomer by conjugated reduction. It is noticed that the spatial structures of 17-substituents in **AD** and **ADD** derivatives, seemingly by a conformational transmission effect [15,16], markedly influenced the ratio of 5α-*H*-3β-hydroxy and 5β-*H*-3α-hydroxy isomers. With the 17-*O*-acyl cyanohydrin protected group, 4-en-3-one of **AD** was reduced to 5β-*H*-3α-hydroxy- and 5α-*H*-3β-hydroxy-androstane isomers with ratios around 2.8:1–1:1. While the **AD** and **ADD** derivatives with 17-spirocyclic ethyleneketal protected were reduced to 5β-*H*-3α-hydroxy- and 5α-*H*-3β-hydroxy-androstane isomers at a 9:1 predominant ratio, and nearly 80% diastereo-selectivity. And the configuration of one reduction product **17e** was identified by X-ray diffraction. The 3α-hydroxy-5β-androstane isomer with two chiral centers at 3- and 5-position is the common structural pattern of bile acid derivatives. Because of the low cost of reagents and easy acquisition of **AD** and **ADD** as starting materials, this reduction method has great potential in the synthetic investigation of bile acid analogs.

Herein, we would like to report the details of this research.

2. Experimental

Melting points were measured with a microscope (SGW X-4) melting apparatus without correction. The ^{1}H NMR was measured on a Bruker-DPX spectrometer operating at 400 MHz for ^{1}H and 101 MHz for ^{13}C, expressed in ppm. ^{1}H assignments were made using 2D *g*COSY experiments, while ^{13}C assignments were made using 2D *g*HSQC and *g*HMBC experiments. The solvents used were $CDCl_3$ and DMSO-d6. Mass spectrum was measured with HP5973N analytical mass spectrometers. Optical rotation was measured on a WZZ-2S polarimeter. Column chromatography (CC) was carried out on silica gel (300–400 mesh, Shanghai Sanpont, Co., Ltd, China) and TLC analyses were carried out on silica gel GF254 glass plates (20 × 20 cm with 250 μm layer, Yantai Jiangyou Company, China).

2.1. General protection procedure of 17-one

2.1.1. 17β-Cyano-17α-hydroxyandrost-4-en-3-one (*1*)

AD (4.0 g, 14 mmol) was dissolved in the mixture of methanol (20 mL) and water (2 mL). To the solution, sodium carbonate (150 mg, 1.4 mmol) and acetone cyanohydrin (4.1 mL, 44.9 mmol) were added. The reaction mixture was stirred at 40 °C for 4 h and then water (6 mL) was added. The mixture was stirred for another 5 h and kept overnight at room temperature. Solid was filtered, washed with diethyl ether and then poured into 0.2 N HCl (30 mL). After stirring for 3 h, the product was collected by filtration, washed with water and dried to give **1** (3.9 g, 89%). m.p. 171–174 °C; Recrystallization from acetone gave white crystal: m.p. 177–178 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.73 (1H, s, 4-CH), 1.20 (3H, s, 19-CH_3), 0.98 (3H, s, 18-CH_3). ESI-MS *m/z* (%): 313 $[M]^+$.

2.1.2. 17α-Cyano-17β-hydroxyandrost-4-en-3-one (*2*)

AD (2.4 g, 8.4 mmol) was dissolved in acetone cyanohydrin (3.6 mL, 39.4 mmol). The mixture was stirred at 40 °C for 8 h and then a few drops of triethylamine were added to adjust pH to 8.8–9.0. Stirring was continued for another 0.5 h. The solid was filtered, washed with diethyl ether, followed with acetone adjusted by *p*-toluenesulfonic acid to pH = 6, and dried to give **2** (1.7 g, 65%), m.p. 162–164 °C. ESI-MS *m/z* (%): 313 $[M]^+$.

2.1.3. 17β-Cyano-17α-acetoxyandrost-4-en-3-one (*3b*) and 17α-cyano-17β-acetoxyandrost-4-en-3-one (*4b*)

Acetic anhydride (6.0 mL, 64 mmol) was added under nitrogen to a solution of **1** (4.0 g, 12.8 mmol) and dimethylaminopyridine (DMAP) (0.5 g, 4.1 mmol) in dry pyridine (40 mL). The resulting mixture was refluxed for 3 h and poured into ice water (150 mL). The resulting brown solid was filtered, washed with water till neutral, dried to give **3b** (4.35 g, 96%), m.p. 219–222 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.77 (1H, s, 4-CH), 2.11 (3H, s, –$COCH_3$), 1.22 (3H, s, 19-CH_3), 1.12 (3H, s, 18-CH_3). ESI-MS *m/z* (%): 356.2 (M+1).

The synthesis procedure of **4b** was the same as that used for the preparation of **3b**. Yield: 93%, m.p. 157–160 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.75 (1H, s, 4-CH), 2.11 (3H, s, –$COCH_3$), 1.20 (3H, s, 19-CH_3), 0.98 (3H, s, 18-CH_3). ESI-MS *m/z* (%): 356.2 (M+1).

2.1.4. 17β-Cyano-17α-benzoyloxyandrost-4-en-3-one (*3c*) and 17α-cyano-17β-benzoyloxyandrost-4-en-3-one (*4c*)

Trifluoroacetic anhydride (4.5 mL, 32 mmol), *p*-toluenesulfonic acid (0.5 g, 2.9 mmol) and benzoic acid (4.0 g, 32 mmol) were added to $CHCl_3$ (15 mL). After stirring for 0.5 h at room temperature, compound **1** (2.0 g, 6.4 mmol) was added. The reaction was stirred at room temperature and monitored by TLC until the substrate disappeared. Then the mixture was alkalized to pH 7–8 with saturated sodium bicarbonate solution and extracted with $CHCl_3$ (3 × 15 mL). The organic layer was dried (Na_2SO_4), filtered, and concentrated in vacuum to give the crude product, which was purified by column chromatography using CH_2Cl_2 as eluent to give a white solid **3c** (1.5 g, 56%), m.p. 200–202 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.49–8.08 (5H, m, –COC_6H_5), 5.77 (1H, s, 4-CH), 1.22 (3H, s, 19-CH_3), 1.16 (3H, s, 18-CH_3). ESI-MS *m/z* (%): 418.2 (M+1).

The synthesis procedure of **4c** was the same as that used for the preparation of **3c**. Yield: 62%, m.p. 246–249 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.49–8.08 (5H, m, –COC_6H_5), 5.77 (1H, s, 4-CH), 1.22 (3H, s, 19-CH_3), 1.16 (3H, s, 18-CH_3). ESI-MS *m/z* (%): 418.2 (M+1).

2.1.5. Synthesis of *14*, *15e*, *15f* and *18*

The compound **14** was prepared by the ketalization of **AD** (1.0 g, 3.5 mmol) in refluxing toluene (10 ml) using ethylene glycol (0.7 mL, 12.3 mmol). After 2 h, *p*-toluenesulfonic acid (0.05 g, 0.3 mmol) was added to the mixture. The reaction mixture was worked up after another 1 h and then alkalized to pH 7–8 with saturated sodium bicarbonate solution. The organic layer was dried (Na_2SO_4), filtered, and concentrated in vacuum to give the crude product, which was purified by column chromatography using 17% ethyl acetate in petroleum ether as eluent to give a white solid **14** (0.53 g, 46%), m.p. 178–181 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.73 (s, 1H, 4-H), 3.89 (m, 4H, $(OCH_2)_2$), 1.22 (s, 3H, 19-CH_3), 0.79 (s, 3H, 18-CH_3). ESI-MS *m/z* (%): 331.2 (M+1). The data were compared with that of the literature [17].

The synthesis procedure of **15e** was the same as that used for the preparation of **14** with **ADD** as the starting material, and the product was purified by crystallizing from acetone with a yield of 73%, m.p. 170–172 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.12 (d, J = 9.5 Hz, 1H, 2-H), 6.20 (d, J = 10.5 Hz, 1H, 1-H), 6.02 (s, 1H, 4-H), 3.89 (m, 4H, $(OCH_2)_2$), 1.24 (s, 3H, 19-CH_3), 0.85 (s, 3H, 18-CH_3). ESI-MS m/z (%): 329.2 (M+1). The data were compared with that of the literature [18].

The synthesis procedure of **15f** was the same as that used for the preparation of **15e** with 11α-hydroxy-androsta-1,4-diene-3,17-dione as the starting material, and the product was crystallized from methanol with a yield of 88%, m.p. 250–252 °C. ESI-MS m/z (%): 345.2 (M+1).

The synthesis procedure of **18** was the same as that used for the preparation of **15e** with 6-methyleneandrost-4-en-3,17-dione as the starting material, and the product was crystallized from methanol with a yield of 90%, m.p. 170–172 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.98 (s, 1H, 4-H), 5.05 (s, 1H, 6-CH_2), 4.93 (s, 1H, 6-CH_2), 3.89 (m, 4H, $(OCH_2)_2$), 1.14 (s, 3H, 19-CH_3), 0.89 (s, 3H, 18-CH_3). ESI-MS m/z (%): 343.2 (M+1).

2.2. General reduction procedure with sodium dithionite

A solution of steroidal substances **AD**, **3a**–**c**, **4a**–**c**, **14** (1 equiv) in toluene, together with a solution of sodium dithionite (9 equiv) and sodium bicarbonate (9 equiv) in water in presence of PTC (5% equiv), was refluxed and stirred for 3 h. The aqueous layer was separated and organic phase was concentrated in vacuum. Products were purified by the column chromatography using 9–10% ethyl acetate in petroleum ether.

2.2.1. Reduction of androst-4-en-3,17-dione (*AD*)

AD was reduced following the general reduction procedure, and yielded **9** and **10** in 78% with the ratio of 2:1.

Compound **9**: m.p. 134–137 °C. Optical rotation: $[\alpha]_D^{25}$ = +159.7° (CH_2Cl_2, 0.01 g/mL). IR (KBr): ν 1452, 1713, 1746, 2832, 2939 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.03 (3H, s, 19-CH_3), 0.88 (3H, s, 18-CH_3), 0.79 (1H, m, 9-H). ESI-MS m/z (%): 289.3 (M+1).

Compound **10**: m.p. 134–137 °C. Optical rotation: $[\alpha]_D^{25}$ = +97.2° (CH_2Cl_2, 0.01 g/mL). IR (KBr): ν 1446, 1708, 1734, 2862, 2928 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.05 (3H, s, 19-CH_3), 0.89 (3H, s, 18-CH_3). ESI-MS m/z (%): 289.3 (M+1).

2.2.2. Reduction of 17β-cyano-17α-acetoxyandrost-4-en-3-one (*3b*) and 17α-cyano-17β-acetoxyandrost-4-en-3-one (*4b*)

Compound **3b** was reduced by the general reduction procedure, and yielded **5b** and **6b** in 85% with the ratio of 2:1.

Compound **5b**: m.p. 226–230 °C. Optical rotation: $[\alpha]_D^{25}$ = +32.9° (CH_2Cl_2, 0.01 g/mL). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.11 (3H, s, –$COCH_3$), 1.03 (6H, d, J = 1.8 Hz, 18-CH_3, 19-CH_3), 0.79 (1H, m, 9-H). ESI-MS m/z (%): 358.3 (M+1).

Compound **6b**: m.p. 225–229 °C. Optical rotation: $[\alpha]_D^{25}$ = +19.3° (CH_2Cl_2, 0.01 g/mL). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.14 (3H, s, –$COCH_3$), 1.04 (6H, d, J = 6.7 Hz, 18-CH_3, 19-CH_3). ESI-MS m/z (%): 375.2 (M+18).

Compound **4b** was reduced by the general reduction procedure, and yielded **7b** and **8b** in 94% (**7b**:**8b** = 1:1).

Compound **7b**: m.p. 205–207 °C. Optical rotation: $[\alpha]_D^{25}$ = +6.6° (CH_2Cl_2, 0.01 g/mL). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.11 (3H, s, –$COCH_3$), 1.02 (3H, s, 19-CH_3), 0.92 (3H, s, 18-CH_3), 0.84 (1H, m, 9-H). ESI-MS m/z (%): 358.2 (M+1).

Compound **8b**: m.p. 167–170 °C. Optical rotation: $[\alpha]_D^{25}$ = −1.2 (CH_2Cl_2, 0.01 g/mL). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.10 (3H, s, –$COCH_3$), 1.04 (3H, s, 19-CH_3), 0.92 (3H, s, 18-CH_3). ESI-MS m/z (%): 358.2 (M+1).

2.2.3. Reduction of 17β-cyano-17α-benzoyloxyandrost-4-en-3-one (*3c*) and 17α-cyano-17β-benzoyloxyandrost-4-en-3-one (*4c*)

Compound **3c** was reduced by the general procedure, and yielded **5c** and **6c** in 80% with the ratio of 2.2:1.

Compound **5c**: m.p. 207–209 °C. Optical rotation: $[\alpha]_D^{25}$ = +6.8° (CH_2Cl_2, 0.01 g/mL). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.51–8.02 (5H, m, –COC_6H_5), 1.12 (3H, s, 19-CH_3), 1.09 (3H, s, 18-CH_3), 0.92 (1H, m, 9-H). ESI-MS m/z (%): 420.2 (M+1).

Compound **6c**: m.p. 211–213 °C. Optical rotation: $[\alpha]_D^{25}$ = +5.8° (CH_2Cl_2, 0.01 g/mL). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.49–8.01 (5H, m, –COC_6H_5), 1.12 (3H, s, 19-CH_3), 1.09 (3H, s, 18-CH_3). ESI-MS m/z (%): 420.2 (M+1).

Compound **4c** was reduced by the general procedure, and yielded **7c** and **8c** in 89% (**7c**:**8c** = 1.8:1).

Compound **7c**: m.p. 264–267 °C. Optical rotation: $[\alpha]_D^{25}$ = +39.7° (CH_2Cl_2, 0.01 g/mL). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.47–8.01 (5H, m, –COC_6H_5), 1.06 (6H, d, J = 8.9 Hz, 18-CH_3, 19-CH_3), 0.94 (1H, m, 9-H). ESI-MS m/z (%): 420.1 (M+1).

Compound **8c**: m.p. 245–247 °C. Optical rotation: $[\alpha]_D^{25}$ = +37.9° (CH_2Cl_2, 0.01 g/mL). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.47–8.01 (5H, m, –COC_6H_5), 1.07 (6H, d, J = 8.9 Hz, 18-CH_3, 19-CH_3). ESI-MS m/z (%): 420.1 (M+1).

2.2.4. Reduction of 17-ethylendioxyandrost-4-en-3-one (*14*)

Compound **14** was reduced by the general procedure, and the original product was then added to 2 N HCl solution to form 17-ketone. Products **9** and **10** were obtained with the yield of 77% and with the ratio of 1:1.4.

2.2.5. Reduction of 17β-cyano-17α-(1-butoxy-ethoxy)-androst-4-en-3-one (*3a*) and 17α-cyano-17β-(1-butoxy-ethoxy)-androst-4-en-3-one (*4a*)

Compound **1** (1.0 g, 3 mmol) and *n*-Butyl vinyl ether (4.2 mL, 30 mmol) were added to anhydrous tetrahydrofuran containing PTS (0.07 g). The mixture was stirred for 0.5 h at room temperature under nitrogen atmosphere. The reaction solution was adjusted to pH 8–9 with triethylamine and concentrated in vacuum to get a brown oil. Then toluene (10 mL) together with a solution of sodium dithionite (5.0 g, 28.7 mmol) and sodium bicarbonate (2.4 g, 28.7 mmol) in water (15 mL) in presence of PTC (0.07 g, 0.17 mmol) were added into the brown oil and refluxed with stirring for 3 h. Monitored by TLC, but no product was appeared.

The reduction procedure of **4a** was same as that used for the preparation of **3a**. But the mixture of **7a** and **8a** was obtained with the yield of only 10%. Further study on their ratio was not conducted.

2.3. Synthesis of Epiandrosterone (*11*)

A solution of sodium borohydride (17 mg, 0.45 mmol) in methanol (4 mL) was added slowly into the methanol (10 mL) solution of **5b** (0.16 g, 0.45 mmol) at −10 °C. After stirring for 0.5 h at this temperature, the mixture was added with 20% KOH (12 mL) aqueous solution, stirred for additional 1 h at 40 °C and then poured into water (15 mL). The resulting solid was filtered, washed with water till neutral, and dried to give the target compound **11** (0.12 g, 92%), m.p. 170–173 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.61 (1H, s, 3-H), 0.85 (6H, d, J = 10.4 Hz, 18-CH_3, 19-CH_3), 0.75 (1H, m, 9-H); ESI-MS m/z (%): 291.3 (M+1).

2.4. General reduction procedure with CuCl/$NaBH_4$

Steroidal substances (**AD**, **3b**–**c**, **4b**, **14**, **15e**–**f**, **18**, 1 equiv) and CuCl (1–2 equiv) were added to dry ethanol. The mixture was stirred for 2 h at room temperature under nitrogen atmosphere. $NaBH_4$ (2–6 equiv) was added slowly to the solution. Monitored

by TLC and stop the reaction when the substrate disappeared. The black solution was then filtered and concentrated in vacuum to give original product. The original product was then added to saturated NaOH solution (**3b–c**, **4b**) or 2 N HCl (**14**, **15e–f**, **18**) solution to form 17-ketone. The resulting white solid was filtered, washed with water till neutral, and dried. The crude solid was chromatographed by using 9–10% ethyl acetate in petroleum ether as eluent to obtain eight target compounds.

*2.4.1. 3β,17β-dihydroxy-5α-androstane (**12**) and 3α,17β-dihydroxy-5β-androstane (**13**)*

AD was reduced by the general procedure, and yielded **12** and **13** in 73% with the ratio of 4:3.

Compound **12**: m.p. 158–160 °C ESI-MS *m/z* (%): 315.1 (M+23). ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.60 (dt, *J* = 11.1, 7.3 Hz, 2H, 3-H, 17-H), 0.83 (s, 3H, 19-CH_3), 0.77(s, 3H, 18-CH_3), 0.63 (m, 1H, 9-H). ^{13}C NMR (101 MHz, DMSO-d6) δ 79.99, 69.24, 54.01, 50.53, 44.39, 42.49, 38.80, 38.11, 36.63, 36.57, 35.12, 31.28, 29.78, 28.27, 23.03, 20.39, 12.09, 11.27.

Compound **13**: m.p. 241–243 °C ESI-MS *m/z* (%): 315.1 (M+23). ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.60 (dt, *J* = 11.1, 7.3 Hz, 2H, 3-H, 17-H), 0.98 (s, 3H, 19-CH_3), 0.83 (s, 3H, 18-CH_3). ^{13}C NMR (101 MHz, DMSO-d6) δ 80.00, 69.77, 50.60, 42.57, 41.53, 40.17, 36.74, 36.22, 35.44, 35.18, 34.26, 30.30, 29.87, 26.72, 25.76, 23.22, 23.05, 19.95, 11.23.

*2.4.2. 3β-hydroxy-5α-androstane-17-one (**16e**) and 3α-hydroxy-5β-androstane-17-one (**17e**)*

Compound **3b** was reduced by the general procedure, and yielded **16e** and **17e** in 95% with the ratio of 2.8:1.

Compound **16e**: m.p. 180–183 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.65 (s, 1H, 3-H), 0.90 (s, 3H, 19-CH_3), 0.77 (s, 3H, 18-CH_3), 0.83 (m, 1H, 9-H). ESI-MS *m/z* (%): 291.3 (M+1). ^{13}C NMR (101 MHz, $CDCl_3$) δ 221.43, 71.10, 54.38, 51.39, 47.75, 44.79, 38.02, 36.89, 35.81, 35.60, 35.00, 31.51, 31.39, 30.84, 28.35, 21.74, 20.46, 13.77, 12.26.

Compound **17e**: m.p. 143–145 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.65 (s, 1H, 3-H), 0.90 (s, 3H, 19-CH_3), 0.77 (s, 3H, 18-CH_3). ESI-MS *m/z* (%): 291.3 (M+1). ^{13}C NMR (101 MHz, $CDCl_3$) δ 221.36, 71.90, 51.51, 47.87, 42.04, 40.40, 36.35, 35.90, 35.69, 35.39, 34.77, 31.75, 30.52, 26.91, 25.38, 23.29, 21.83, 20.10, 13.80.

*2.4.3. Reduction of 17α-cyano-17β-acetoxyandrost-4-en-3-one (**4b**)*

The procedure was same as that of **3b**, and produced **16e** and **17e** with 1:1 ratio in the total yield of 50%.

*2.4.4. Reduction of 17β-cyano-17α-benzoyloxyandrost-4-en-3-one (**3c**)*

Compound **3c** was reduced by the general procedure and yielded **16e** and **17e** in 29% without being separated.

*2.4.5. Reduction of 17-ethylendioxyandrost-4-en-3-one (**14**)*

Compound **14** was reduced by the general procedure, and yielded **16e** and **17e** in 90% with the ratio of 1:9 (**16e**:**17e**).

*2.4.6. Reduction of 17-ethylendioxyandrosta-1,4-dien-3-one (**15e**)*

The procedure was same as that of **14**, and yielded **16e** and **17e** in 89% with the ratio of 1:9 (**16e**:**17e**).

*2.4.7. 3α,11α-dihydroxy-5β-androstane-17-one (**17f**)*

The reduction procedure of **15f** was same as that of **14**, and yielded **17f** in 81% with no 5α-isomer (**16f**) obtained.

Compound **17f**: m.p. 99–102 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.95 (s, 1H, 11-H), 3.62 (s, 1H, 3-H), 1.04 (s, 3H, 19-CH_3), 0.90 (s, 3H, 18-CH_3). ESI-MS *m/z* (%): 329.2 (M+23). ^{13}C NMR (101 MHz, $CDCl_3$) δ 219.50, 71.91, 68.02, 50.39, 48.08, 47.12, 43.61, 43.22, 38.18, 36.82, 35.95, 35.84, 34.49, 31.69, 27.34, 25.13, 23.54, 21.86, 14.02.

*2.4.8. 3β-hydroxy-6-methyl-androst-5-en-17-one (**19**) and 3α-hydroxy-6-methylen-5β-androstan-17-one (**20**)*

Compound **18** was reduced by the general procedure, and yielded **19** (57%) together with **20** (30%).

Compound **19**: m.p. 152–155 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.55 (s, 1H, 3-H), 1.82 (s, 3H, 6-CH_3), 1.04 (s, 3H, 19-CH_3), 0.90 (s, 3H, 18-CH_3). ESI-MS *m/z* (%): 303.3 (M+1). ^{13}C NMR (101 MHz, $CDCl_3$) δ 221.20, 132.59, 125.21, 70.86, 51.44, 50.35, 47.47, 37.75, 37.49, 36.93, 35.90, 35.24, 31.61, 31.50, 31.32, 21.88, 20.47, 19.64, 19.56, 13.58.

Compound **20**: m.p. 113–116 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 4.80 (s, 1H, 6-CH_2), 4.50 (s, 1H, 6-CH_2), 3.62 (s, 1H, 3-H), 1.04 (s, 3H, 19-CH_3), 0.90 (s, 3H, 18-CH_3). ESI-MS *m/z* (%): 303.3 (M+1). ^{13}C NMR (101 MHz, $CDCl_3$) δ 220.90, 148.52, 106.62, 71.31, 54.75, 51.37, 49.31, 47.88, 40.74, 37.82, 36.97, 36.49, 35.81, 33.67, 31.46, 31.22, 21.88, 20.47, 13.83, 12.47.

3. Results and discussion

3.1. Regioselective reduction C=C of 4-En-3-one moiety with $Na_2S_2O_4$/$NaHCO_3$

With the desire of regio- and stereo-selectively reducing C=C double band of 4-en-3-one steroids in mind, androst-4-en-3,17-dione (**AD**) was selected as model substrate to repeat its reduction with $Na_2S_2O_4$/$NaHCO_3$ according to Akamanchi's method mentioned in the literature [11]. Unexpectedly, two configuration isomers, 5α- and 5β-androstane-3,17-dione (**9** and **10**) were produced at a 2:1 ratio with a total yield of 78%. This is different from only 5α-androstane-3,17-dione isomer formed at 88% yield as reported in the literature. The configurations of 5α- and 5β-androstane-3,17-diones (**9**, **10**) were confirmed by the X-ray crystal diffraction analysis (Fig. 2). Duax et al [15,16] had reported that the 17-substituents of 17α-acetoxy progesterone could change the A-ring conformation by conformational transmission, which then affect the achieved reaction at the ring A. In order to investigate and improve the ratio of 5α-androstane-3,17-dione isomer in the synthesis of Epiandrosterone, six different *O*-substituted 17-cyanohydrin (**3a–c**, **4a–c**) and one 17-spirocyclic ketal of **AD** (**14**) were further prepared as substrates to monitor their stereoselective reduction with $Na_2S_2O_4$/$NaHCO_3$.

As Scheme 1 shown, in the Na_2CO_3/MeOH-H_2O solvent of acetone cyanohydrin and pure acetone cyanohydrin, the configuration of pure 17α-hydroxy-17β-cyano-androst-4-en-3-one (**1**) and 17α-cyano-17β-hydroxy-androst-4-en-3-one (**2**) were synthesized with yields of 89% and 65%, respectively. Then two 17-ether-protected cyanohydrins (**3a**, **4a)**, four 17-*O*-acyl-protected cyanohydrins (**3b–c**, **4b–c**) and one 17-spirocyclic ketal (**14**) were prepared further to undergo PTC reduction. The results were shown in Table 1. Surprisingly, **4a** yielded only 10% of reductive products, while **3a** had no reaction at all. This implied that the bulky 17-substituent might hinder the reduction of 4-ene in A ring. As for the others, the **3b** and **3c** produced **5b**, **5c** and **6b**, **6c** with ratios of 2:1 and 2.2:1 for 5α-:5β-isomer and at yields of 85% and 80%, respectively. The **4b** and **4c** resulted in **7b**, **7c** and **8b**, **8c** at ratios of 1:1 and 1.8:1 for 5α-:5β-isomer and with yields of 94% and 89%, respectively. And the substrate **14** also created 5α-:5β-isomer at a ratio of 1:1.4 and with the yield of 77%. The configurations of **5b** (5α-isomer) and **6b** (5β-isomer) were confirmed by crystal X-ray diffraction (Fig. 3). The configurations of other reduction products were confirmed *via* 17-deprotection to form 17-ketone. They were then

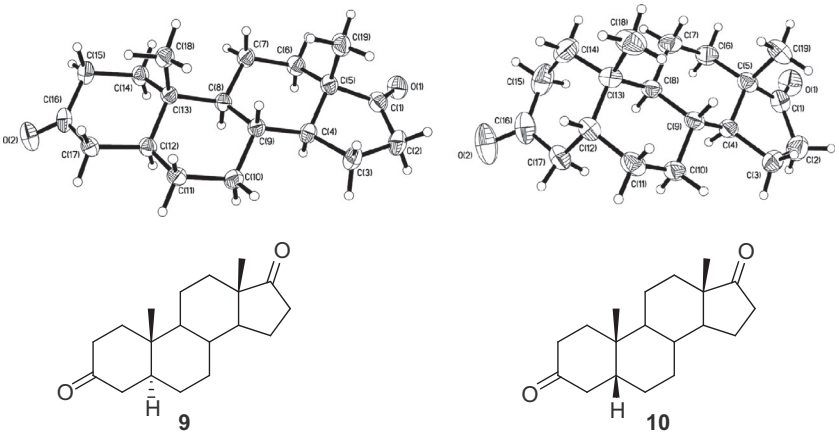
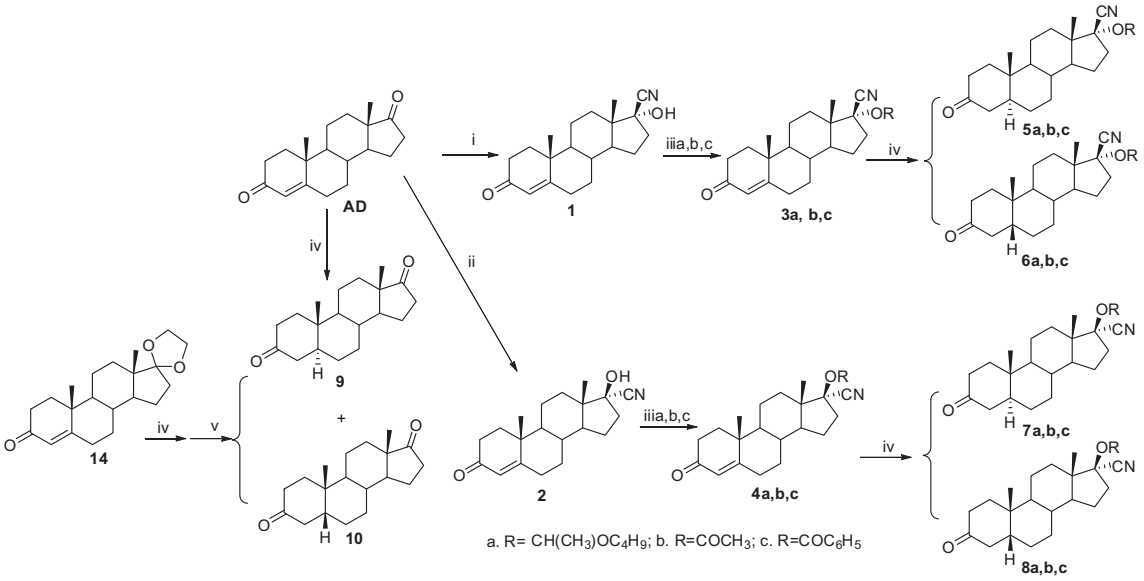


Fig. 2. X-ray diffraction crystal structures of **9** and **10**.



Scheme 1. Reagents and conditions: (i). $(CH_3)_2C(OH)CN$, Na_2CO_3, MeOH-H_2O, 40 °C for 9 h, 0.2 N HCl for 3 h, RT; (ii). $(CH_3)_2C(OH)CN$, 40 °C for 8 h, $N(Et)_3$, 0 °C for 0.5 h; (iiia). $C_2H_3OC_4H_9$, PTS, THF; (iiib). $(CH_3CO)_2O$, Py, DMAP, refluxed for 3 h; (iiic). $(CF_3CO)_2O$, PTS, C_6H_5COOH; (iv). $Na_2S_2O_4$, $NaHCO_3$, toluene-water, refluxed for 3 h; (v). 2 N HCl aq, RT.

Table 1
The selective PTC reduction of 4-ene-3-one with sodium dithionite.

Entry	Substrate	R	T	Ratio[a] (mol)	Time (h)	Yield (%)	5-α:5-β
1	**4-AD**	–	Reflux	1:9:9	3	78	2:1
2	**3a**	$CH_3CHOC_4H_9$	Reflux	1:9:9	3	0	–
3	**3b**	$COCH_3$	Reflux	1:9:9	3	85	2:1
4	**3c**	COC_6H_6	Reflux	1:9:9	3	80	2.2:1
5	**4a**	$CH_3CHOC_4H_9$	Reflux	1:9:9	3	10	–
6	**4b**	$COCH_3$	Reflux	1:9:9	3	94	1:1
7	**4c**	COC_6H_6	Reflux	1:9:9	3	89	1.8:1
8	**14**[b]	–	Reflux	1:9:9	3	77	1:1.4

[a] The molar ratio refers to substrate: $Na_2S_2O_4$: $NaHCO_3$ = 1:9:9.
[b] The 17-one was protected by ethylendioxy group.

compared with **9** and **10**, of which the configurations were measured by single crystal X-ray diffraction (Fig. 2).

The data in Table 1 showed that the structural pattern at 17-position has considerable effect on the yield and stereoselectivity of 4-ene reduction. The 17-position in steroidal skeleton is far from the reaction center of 4-ene reduction. This remote effect should have been resulted from the so-called conformation transmission effect. The conformation change of D ring caused by different 17-substitutes or structural patterns could have transmitted to A ring and therefore resulted in its conformation change, which might influence the stereoselectivity of reduction.

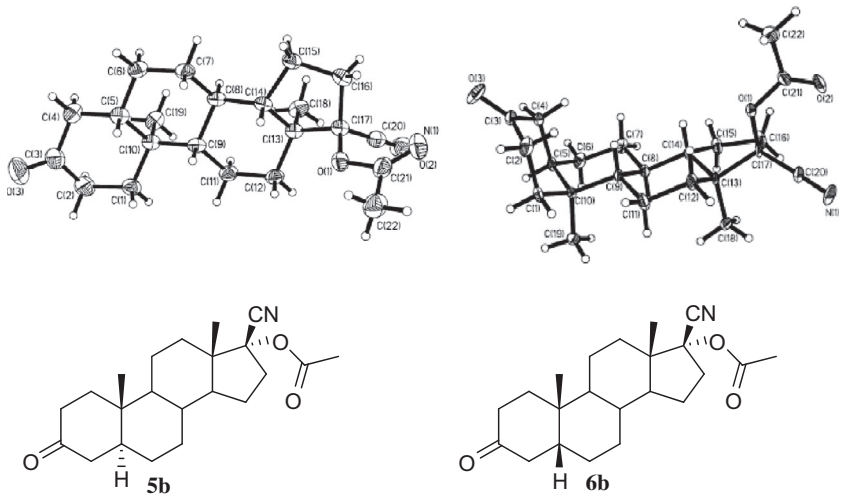
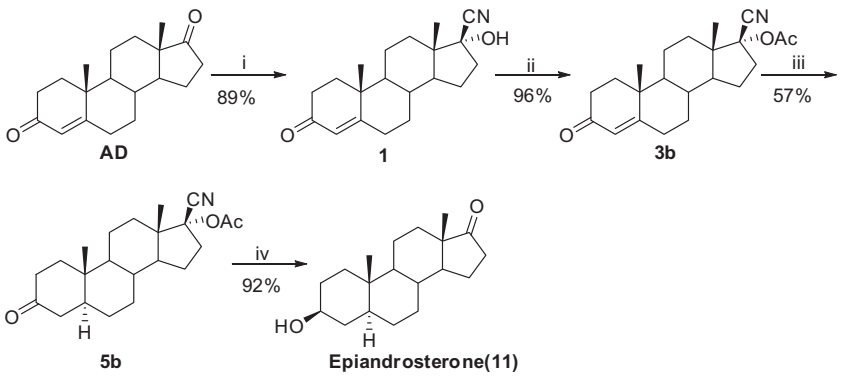


Fig. 3. X-ray diffraction crystal structures of **5b** and **6b**.



Scheme 2. Synthesis of Epiandrosterone. (i). $(CH_3)_2C(OH)CN$, Na_2CO_3, MeOH-H_2O, 40 °C for 9 h, 0.2 N HCl for 3 h, RT; (ii). $(CH_3CO)_2O$, Py, DMAP, refluxed for 3 h; (iii). $Na_2S_2O_4$, $NaHCO_3$, toluene-water, refluxed for 3 h; (iv). $NaBH_4$, MeOH, −10 °C for 0.5 h, 20% KOH in water, 40 °C for 1 h.

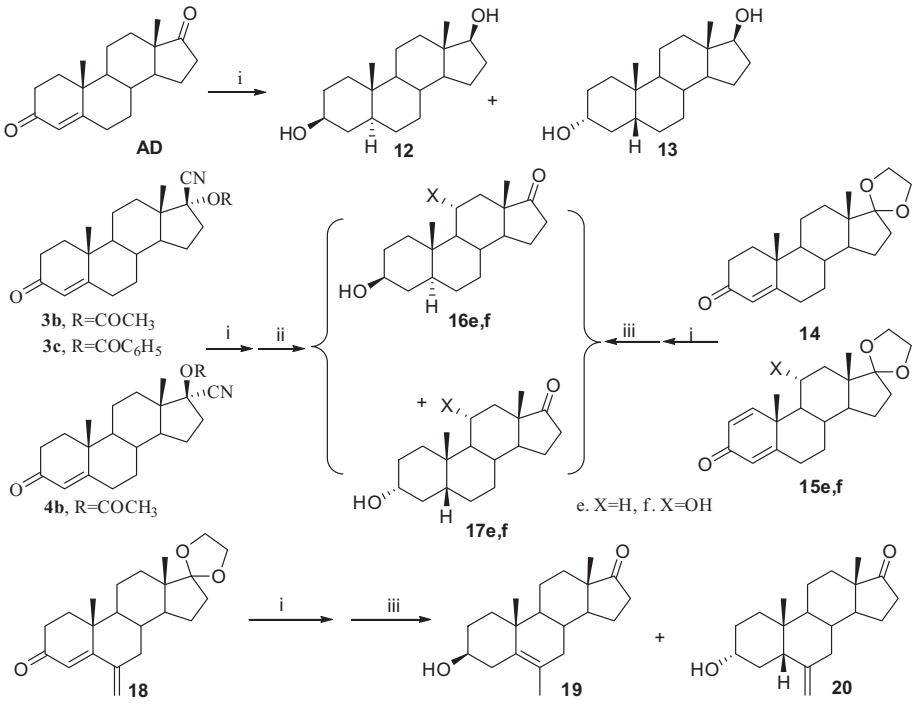
As an extended use of C=C stereoselective reduction product in 4-en-3-one steroids, **3b** was selected as an intermediate product for further synthesizing Epiandrosterone (**11**), which was a key starting material for synthesizing a series of steroidal drugs, such as Rocuronium Bromide, Vecuronium Bromide, etc. As Scheme 2 below showed, the overall yield of the 4-step synthesis from **AD** was 45%.

3.2. Conjugated reduction of 4-En-3-one moiety with CuCl/$NaBH_4$

AD, three 17-ester-protected cyanohydrins (**3b**, **3c** and **4b**) and four 17-spirocyclic ketal (**14**, **15e**, **15f** and **18**) compounds were used as the substrates to perform their reduction with CuCl/$NaBH_4$. The general procedure was described in Scheme 3 below. At room temperature, the mixture of substrate and CuCl in absolute ethanol was added with $NaBH_4$ slowly and stirred for a couple of hours with TLC monitoring. After hydrolysis of 17-protected group, 17-ketone was formed in the aqueous solution of NaOH or HCl. The products were isolated by common workup mentioned in the experimental section.

The experimental data and results listed in Table 2 above illustrate that the reduction of 4-en-3-one was a little bit complicated. In the reduction reaction, **AD** was reduced to 3β,17β-dihydroxy-5α-androstane (**12**) and 3α,17β-dihydroxy-5β-androstane (**13**) at a ratio of 4:3 and with a yield of 73%. Due to conformational transmission, the spatial orientation and bulk of 17-substituent in D ring remarkably influenced the ratio and yield of 5α and 5β-*H* isomers again for selective reduction of 4-en-3-one in A ring. 17α-Acetyloxy-17β-cyano-androst-4-en-3-one (**3b**) reacted with CuCl/$NaBH_4$ in ethanol to produce 3β-hydroxy-5α-androstane-17-one (**16e**) and 3α-hydroxy-5β-androstane-17-one (**17e**) at the ratio of 2.8:1 and a total yield of 95%. Similarly, 17β-acetyloxy-17α-cyano isomer (**4b**) generated those two isomers at the ratio of 1:1 and the yield of 50%. On the other hand, 17α-benzoyloxy-17β-cyano derivative (**3c**) was converted into corresponding **16e** and **17e** with a yield of 29% only. Interestingly, the conjugated reduction of **14** and **15e** bearing 17,17-ethylendioxy spiro-ketal as protecting group produced 5β-androstane-3α-hydroxy compound (**17e**) at high stereoselectivity, but formed only 9% of 5α-androstane-3β-hydroxy isomer (**16e**) at a ratio of 9:1 (**17e:16e**). In particular, the introduction of 11α-hydroxy and 6-double bond also dramatically increased the ratio of 5β-androstane-3α-hydroxy isomer. Based on these facts, it was believed that compound **15f** was almost reduced into the desired isomer **17f** at a yield of 81%, and **18** was reduced to 3β-hydroxy-6-methyl-androst-5-en-17-one (**19**) and 3α-hydroxy-6-methylen-5β-androstane-17-one (**20**) at a yield of 86%. In both of these two reactions, no 5α-isomer was separated. The above indicates that the 17-spirocyclic ketal protecting group was more favorable to stereoselectively form 5β-configuration products by using CuCl/$NaBH_4$ as reduction agent.

The two isomers **12** and **13** obtained were identified by ^{1}H NMR, ^{13}C NMR, HSQC and HMBC. Nunes [10] reported that the H-3 chemical shifts of the axial alcohols in 5β-*H*-3β-hydroxyand 5α-*H*-3α-hydroxy isomers tended to move to low field due to lack of diaxial ^{3}*Ja, a* couplings, presenting a peak at 4.0 ppm. On the other



Scheme 3. Reagents and conditions: (i).CuCl/$NaBH_4$, dry ethanol; (ii). NaOH aq, RT; (iii).2 N HCl aq, RT.

Table 2
The conjugate reduction of steroids by CuCl/$NaBH_4$.

Entry	Substrate	T (°C)	Ratio (S:CuCl:$NaBH_4$)	Time (h)	Yield (%)	Ratio
1	**AD**	rt	1:1:4	2.5	73 (**12** + **13**)	4:3 (**12:13**)
2	**3b**	−10	1:2:2	4	95 (**16e** + **17e**)	2.8:1 (**16e:17e**)
3	**3c**	rt	1:2:4	2.5	29 (**16e** + **17e**)	–
4	**4b**	0	1:2:2	2.5	50 (**16e** + **17e**)	1:1 (**16e**:**17e**)
5	**14**	rt	1:2:4	4	90 (**16e** + **17e**)	1:9 (**16e**:**17e**)
6	**15e**	rt	1:2:4	4	89 (**16e** + **17e**)	1:9 (**16e**:**17e**)
7	**15f**	rt	1:2:4	4	81 (**17f**)	–
8	**18**	rt	1:2:6	4	86 (**19** + **20**)	1.9:1 (**19:20**)

hand, the corresponding C-3 chemical shifts tended to move to high field, presenting a peak at 66.1 ppm. While the thermodynamically more stable alcohols, 5β-*H*-3α-hydroxy and 5α-*H*-3β-hydroxy isomers, presented ^{1}H NMR peak at δ = 3.6 (m, 3-CH) with ^{13}C NMR at δ = 71 (C3) around. Furthermore, Kanchithalaivan [19] reported that the ^{13}C NMR of 5α-*H*-3β-hydroxy isomer presented C-9 peak at 54.6 ppm. In this study, the ^{1}H NMR of **12** presented a peak at δ = 3.60 (dt, *J* = 11.1, 7.3 Hz, 2H, 3&17-H) with diaxial ^{3}*Ja, a* couplings and ^{13}C NMR at δ = 79.99 (C17), 69.24 (C3). ^{1}H NMR of **13** also showed peaks at δ = 3.60 (dt, *J* = 11.1, 7.3 Hz, 2H, 3&17-H) and ^{13}C NMR at δ = 80.00 (C17), 69.77 (C3). The HSQC of **12** showed that the C-9 carbon signal at 54.01 ppm assigned a multiplet at 0.63 ppm to H-9 proton; and in the HSQC of **13**, the chemical shift of C-9 moved to 40.17 ppm assigned a multiplet to H-9 at 1.38 ppm. Furthermore, the HMBC of **12** showed the H-19 correlation with a carbon signal at 54.01 ppm (C-9), and the H-9 at 0.63 ppm had a correlation with C-1 at 35.12 ppm. Analyzing all the data mentioned above, it was assumed that the structure of **12** was 3β,17β-dihydroxy-5α-androstane and **13** was 3α,17β-dihydroxy-5β-androstane. Summarizing all the ^{1}H NMR spectrum of the 5α steroids synthesized in our previous studies, the chemical shift of H-9 was observed to present a peak at 0.63–0.94 ppm with a clear multiplet signal in all of the 5α-isomers, including the 5α-androstane (**5b–c**, **9**, **7b–c**, **11**, **12**, **16e**) reported in this study, whereas there was no corresponding chemical shift signals in ^{1}H NMR of all 5β isomers. Meanwhile, the configuration of **17e** was confirmed by X-ray spectrum (Fig. 4), and the structures of the others were also confirmed by comparing their ^{1}H and ^{13}C NMR spectra with **12**, **13**, **17e** and the data reported in the literatures [10,19].

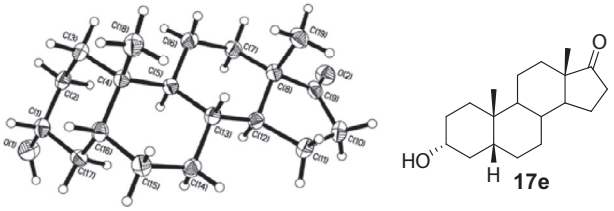


Fig. 4. X-ray diffraction crystal structure of **17e**.

The two chiral centers, 3α-hydroxyl and 5β-*H*, associated with that of bile acid analogs, were introduced simultaneously in one-step reaction with CuCl/$NaBH_4$. The reduction system might be an alternative method for the synthetic investigation of bile acid analogs from **AD**, **ADD** and their derivatives.

4. Conclusion

The two reduction systems, $Na_2S_2O_4$/$NaHCO_3$ and CuCl/$NaBH_4$, were studied to regio- and stereoselectively reduce 4-en-3-one ste-

roids. $Na_2S_2O_4$/$NaHCO_3$ reduced 4-ene with high regioselectivity to give 5α-saturated-3-one compounds as main products, while CuCl/$NaBH_4$ converted both 3-one and 4-ene into 5β-*H*-3α-hydroxy with high stereoselectivity. For the both reduction agents, the ratios and yields of isomers formed by stereoselective reaction at 5-position of A ring were highly related to the spatial conjoint styles of 17-substituentsin D ring. This remote phenomenon is believed to be resulted from the so-called conformational transmission effect. The 17-spirocyclicethylendioxy protected ketal was a preferred group for forming 5β-androstane-3α-hydroxy isomer at a preponderant ratio. Its configuration, including two chiral centers, was just required for bile acid analogs. Furthermore, Epiandrosterone, a key intermediate for synthesizing a number of steroidal drugs, was synthesized from androst-4-en-3,17-dione (**AD**) in four steps *via* 17-hydrocyanation, acylation, reduction of 4-ene with sodium dithionite and 17-deprotection with an overall yield of 45%. Because the starting materials **AD** and **ADD** are readily available from microorganism degradation of natural sterols, the results of this study could be significant for the use of natural sterols, and may lead to the potential application in the synthetic investigation of bile acid analogs.

Acknowledgement

This research was supported by Zhejiang Xianju Pharmaceutical Co., Ltd.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.steroids.2013.09.011.

References

[1] Marsheck WJ, Kraychy S, Muir RD. Microbial degradation of sterols. Appl Microbiol 1972;23:72–7.
[2] Sih CJ, Lee SS, Tsong YY, Wang KC, Chang FN. An efficient synthesis of estrone and 19-norsteroids from cholesterol. J Am Chem Soc 1965;87:2765–6.
[3] Sih CJ, Wang KC. A new route to estrone from sterols. J Am Chem Soc 1965;87:1387–8.
[4] Li PK, Brueggemeier RW. Synthesis and biochemical studies of 7-substituted 4,6-androstadiene-3,17-diones as aromatase inhibitors. J Med Chem 1990;33:101–5.
[5] Zhang BN, Chen Y, Zhang Q, Xia P. Some unreported by-products from the reaction of 1,4-dien-3-one steroids with 2-(methylammo)benzethiol/BF3. Steroids 2005;70:111–6.
[6] Oliveto BE, Erold O, Hershberg EB. Allopregnan-17α,21-diol-3,11,20-trione-21-acetate. J Am Chem Soc 1952;74:2248–50.
[7] Keinan E, Greenspoon N. Highly chemoselective palladium-cata1yzed conjugate reduction of α,β-unsaturated carbonyl compounds with silicon hydrides and zinc chloride cocatalyst. J Am Chem Soc 1986;108:7314–25.
[8] Nunes RMD, Machado BF, Pereira MM, Moreno MJSM, Faria JL. Platinum supported on TiO_2 as a new selective catalyst on heterogeneous hydrogenation of α,β-unsaturated oxosteroids. J Mol Catal A: Chem 2010;333:1–5.
[9] Ravasio N. Selective hydrogenations promoted by copper catalysts. Chemoselectivity, regioselectivity, and stereoselectivity in the hydrogenation of 3-substituted steroids. J Org Chem 1991;56:4329–33.
[10] Nunes RMD, Peixoto AF, et al. Selective hydrogenation of α,β-unsaturated oxosteroids with homogeneous rhodium catalysts. J Mol Catal A: Chem 2006;247:275–82.
[11] Akamanchi KG, Patel HC, Meenakshi R. Reduction of steroidal ketones and enones by sodium dithionite in presence of phase transfer catalyst. Synth Commun 1992;22(12):1655–60.
[12] Mahoney WS, Brestensky DM, Stryker JM. Selective hydride-mediated conjugate reduction of α,β-unsaturated carbonyl compounds using $[(Ph_3P)CuH]_6$. J Am.Chem Soc 1988;110:291–3.
[13] Comins DL, Abdullah AH. Synthesisof l-Acyl-1,4-dihydropyridinevs via copper hydride reduction of 1-acylpyridinium salts. J Org Chem 1984;49:3392–4.
[14] Narisada M, Horibe I, Watanabe F, Takeda K. Selective reduction of Aryl halides and α,β-unsaturated esters with sodium borohydride-cuprous chloride in methanol and its application to deuterium labeling. J Org Chem 1989;54:5308–13.
[15] Duax WL, Cody V, Griffin J, Hazel J, Weeks CM. Steroid structure and function-II. Conformational transmission and receptor binding of medroxyprogesteroneacetate. J Steroid Biochem 1978;9(10):901–7.
[16] Duax WL, Cody V, Hazel J. Steroid structure and function I. Conformational transmission in 17α-acetoxy progesterone. Steroids 1977;30:471–80.
[17] Ács P, Takács A, Kiss M, et al. Systematic investigation on the synthesis of androstane-based 3-, 11- and 17-carboxamides via palladium-catalyzed aminocarbonylation. Steroids 2011;76(3):280–90.
[18] Marwah P, Marwah A, Lardy HA, et al. Steroids as androgen receptor modulators: design, discovery, and structure-activity relationship of new steroidal androgen receptor antagonists. Bioorg Med Chem 2006;14(17):5933–47.
[19] Kanchithalaivan S, Kumar RR, Perumal S. Synthesis of novel 16-spiro steroids: Spiro-7′-(aryl)tetrahydro-1H-pyrrolo[1,2-c][1,3]thiazolo-trans-androsteronehybrid heterocycles. Steroids 2013;78:409–17.