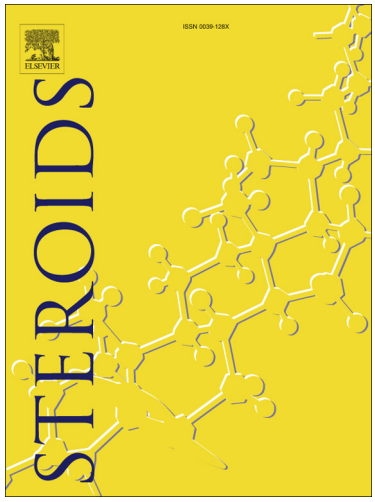

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PII: S0039-128X(14)00132-9
DOI: http://dx.doi.org/10.1016/j.steroids.2014.05.019
Reference: STE 7581

To appear in: *Steroids*

Received Date: 11 March 2014
Revised Date: 19 May 2014
Accepted Date: 25 May 2014

Please cite this article as: Mótyán, G., Kádár, Z., Kovács, D., Wölfling, J., Frank, É., Regio- and stereoselective access to novel ring-condensed steroidal isoxazolines, *Steroids* (2014), doi: http://dx.doi.org/10.1016/j.steroids.2014.05.019

Regio- and stereoselective access to novel ring-condensed steroidal isoxazolines

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Abstract

Novel 5α-androstanes containing an isoxazoline moiety condensed to ring A or D were efficiently synthetized by 1,3-dipolar cycloadditions of aryl nitrile oxides to steroidal α,β-unsaturated ketones. During the ring closures, regioisomers in which the *O* terminus of the nitrile oxide dipoles is attached to the β-carbon of the dipolarophile were formed in a stereoselective manner to furnish exclusively 1α,2α- or 15β,16β-condensed heterocycles. The cyclic enone moiety of the six-membered ring A proved to be less reactive than that of the five-membered ring D, but all the reactions were affected significantly by the substitution pattern of the nitrile oxide. 17-Deacetylation of the primary products resulted in aromatization or simultaneous hydroxylation, depending on the base applied for the ring A-fused heterocycles, while retro-Dieckman-like fragmentation was observed partially or completely for the ring D-fused analogues during 3-deacetylation.

Keywords: 5α-Androstanes, Cycloaddition,Isoxazolines, Stereocontrol, Regioselectivity

1. Introduction

In view of the diversity of their biological activities, heterocyclic steroids continue to be attractive target molecules for stereoselective syntheses [1-6]. The main driving force towards the preparation of such compounds is the modification of the receptor-binding ability by chemical transformation of the extant functional groups and therefore the reduction or elimination of the undesirable hormonal effects [7]. Significant changes in the pharmacodynamic and pharmacokinetic properties can result from the introduction of a hetero ring into the sterane core.

Among the five-membered heterocyclic compounds, 2-isoxazolines have attracted interest as structural building blocks of biologically active molecules and they also serve as versatile intermediates in organic chemistry [8-10]. Thanks to their stability, the hetero ring can be functionalized or can be reductively cleaved to furnish a number of important bifunctional units, such as α,β-unsaturated ketones [11], β-hydroxycarbonyl compounds [12] or 1,3-aminoalcohols [13]. The outstanding role of 2-isoxazolines, however, is due to their straightforward synthetic availability from chemically divergent alkenes and nitrile oxides by 1,3-dipolar cycloadditions [14].

In consequence of their instability and high reactivity, most nitrile oxides have to be prepared *in situ*. In the absence of a dipolarophile, sterically unhindered nitrile oxides tend to dimerize to furoxanes at ambient or lower temperatures, or (especially the sterically stabilized ones) undergo rearrangement to isocyanates at elevated temperature [15]. Although most aryl nitrile oxides possess relatively long half-lives at room temperature, the presence of an electron-withdrawing substituent on the aromatic ring facilitates dimerization, while the resistance to furoxane formation is enhanced by electron-donating groups and sterically by *ortho* substituents [5]. These side-reactions may also occur to a certain extent even in the presence of an alkene component, which can reduce the yield of the desired isoxazoline.

Hydroximidoyl chlorides, the most frequently applied precursors of nitrile oxides, are usually prepared from aldoximes by oxidative halogenation with *N*-chlorosuccinimide and subsequent base-induced dehydrohalogenation [16].

A number of steroidal isoxazolines with diverse pharmacological activities (*i.e.* anti-inflammatory, hypocholesterolemic, antiviral, antibacterial, antifungal and antiproliferative effects) have been reported to date [17-19], but there are no examples of isoxazolines condensed to positions 1,2 or 15,16 of sterane ring A or D. The addition of steric bulk adjacent to the extant functional groups on C-3 or C-17, essential for hormone-receptor binding, may contribute to a change in biological activity and these derivatives may therefore deserve attention from a pharmacological aspect. Moreover, the vicinity of the angular methyl groups on C-10 and C-13 to the reaction centre and also the rigidity of the sterane skeleton overall may have a significant influence on the stereo- and regiocontrol of the processes.

As an extension of our work on the synthesis of steroid-fused heterocycles [20-23], we set out to prepare novel ring-fused isoxazolines from α,β-unsaturated steroidal 17-ketones with aryl nitrile oxides via intermolecular 1,3-dipolar cycloadditions. A further goal was to investigate the regio- and stereoselectivity of the processes and to compare the reactivities of rings A and D against nitrile oxides. The influence of steric and electronic factors on the ring closures and the behaviour of the cycloadducts under conventional deacetylation conditions were also studied. Determination of the stereostructures of the synthetized compounds was also planned.

2. Experimental

2.1. General

Melting points (Mps) were determined on an SMS Optimelt digital apparatus. Elemental analysis data were obtained with a Perkin Elmer CHN analyser model 2400. NMR spectra were recorded at room temperature with a Bruker DRX 500 instrument. Chemical shifts are reported in ppm (δ scale), and coupling constants (J) in Hz. For the determination of multiplicities, the *J*-MOD pulse sequence was used. Automated flow injection analyses were performed by using an HPLC/MSD system. The system comprised an Agilent 1100 micro vacuum degasser, a quaternary pump, a micro-well plate autoinjector and a 1946A MSD equipped with an electrospray ion source (ESI) operated in positive ion mode. The ESI parameters: nebulizing gas N_2, at 35 psi; drying gas N_2, at 350 °C and 12 L/min; capillary voltage 3000 V; and fragmentor voltage 70 V. The MSD was operated in scan mode with the mass range *m/z* 60–620. Samples (0.2 μL) were injected with an automated needle wash directly into the solvent flow (0.3 mL/min) of MeCN/H_2O 70:30 (v/v) supplemented with 0.1% formic acid. The system was controlled by Agilent LC/MSD Chemstation software. All solvents were distilled immediately prior to use. Reagents and materials were obtained from reliable commercial suppliers and were used without purification. The reactions were monitored by TLC on Kieselgel-G (Merck Si 254 F) layers (0.25 mm thick); solvent systems (ss): (A) CH_2Cl_2, (B) EtOAc/CH_2Cl_2 (2:98 v/v), (C) EtOAc/CH_2Cl_2 (5:95 v/v) or (D) EtOAc/CH_2Cl_2 (10:90 v/v). The spots were detected by spraying with 5% phosphomolybdic acid in 50% aqueous phosphoric acid. The R_f values were determined for the spots observed by illumination at 254 and 365 nm. Flash chromatography: Merck silica gel 60, 40–63 μm.

*2.2. General procedure for the synthesis of ring-condensed isoxazolines (**5**, **6**, **9** and **11**) in the Δ^5 androstene series*

17β-Acetoxy-5α-androst-1-en-3-one (**1**, 330 mg, 1.00 mmol), 3β-acetoxy-5α-androst-15-en-17-one (**2**, 330 mg, 1.00 mmol), 17β-hydroxy-5α-androst-1-en-3-one (**13**, 288 mg, 1.00 mmol) or 3β-hydroxy-5α-androst-15-en-17-one (**14**, 288 mg, 1.00 mmol) and the appropriate aromatic imidoyl chloride (**3a–g**, 1.50 mmol) were dissolved in toluene (15 mL), and DIPEA (0.52 mL, 3.00 mmol) was added dropwise to the reaction mixture at room temperature, with subsequent refluxing for 5 h (for **1** and **13**) or for 2 h (for **2** and **14**). The solvent was then evaporated off *in vacuo* and the resulting crude product was purified by column chromatography with CH_2Cl_2 for **5** and **6** and with EtOAc/CH_2Cl_2 = 5:95 for **9** and **11**.

*2.2.1. 17β-Acetoxy-3'-phenyl-2'-isoxazolino[4',5'-d:2α,1α]-5α-androstan-3-one (**5a**)*

Compound **1** and *N*-hydroxybenzenecarboximidoyl chloride (**3a**, 233 mg) were used for the synthesis as described in the General Procedure. The crude product **5a** (247 mg, 55%) was obtained as a white precipitate. Mp 222–224 °C; R_f = 0.54 (ss B). Anal. Calcd. for $C_{28}H_{35}NO_4$ (449.58): C, 74.80; H, 7.85. Found: C, 74.92; H, 7.97. ^{1}H NMR (500 MHz, $CDCl_3$): δ0.81 (s, 3H, 18-H_3), 0.95 (s, 3H, 19-H_3), 0.98 (m, 1H), 1.12 (m, 1H), 1.26–1.32 (m, 3H), 1.40–1.79 (m, 9H), 2.04 (s, 3H, Ac-CH_3), 2.12 (m, 1H), 2.19–2.36 (m, 3H), 4.31 (d, 1H, J = 10.0 Hz, 2-H), 4.59 (t, 1H, J = 8.5 Hz, 17-H), 4.86 (d, 1H, J = 10.0 Hz, 1-H), 7.38 (overlapping m, 3H, 3"-H, 4"-H and 5"-H), 7.75 (d, 2H, J = 7.6 Hz, 2"-H and 6"-H) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ 12.0 (C-18), 12.2 (C-19), 20.4 (CH_2), 21.2 (Ac-CH_3), 23.6 (CH_2), 27.5 (CH_2), 28.7 (CH_2), 30.5 (CH_2), 34.8 (CH), 35.7 (CH), 36.4 (CH_2), 38.7 (C-10), 42.4 (C-13), 42.8 (CH_2), 46.2 (CH), 50.2 (CH), 58.7 (C-2), 82.7 (C-17), 88.6 (C-1), 127.5 (2C), 128.5 (2C), 128.7 (C-1"), 130.2 (C-4"), 155.2 (C-5'), 171.1 (Ac-CO), 206.1 (C-3) ppm; ESI-MS 450 $[M+H]^+$.

*2.2.2. 17β-Acetoxy-3'-4"-methoxyphenyl-2'-isoxazolino[4',5'-d:2α,1α]-5α-androstan-3-one (**5b**)*

Compound **1** and *N*-hydroxy-4-methoxybenzenecarboximidoyl chloride (**3b**, 279 mg) were used for the synthesis as described in the General Procedure. The crude product **5b** (360 mg, 75%) was obtained as a white precipitate. Mp 166–168 °C; R_f = 0.50 (ss C). Anal. Calcd. for $C_{29}H_{37}NO_5$ (479.61): C, 72.62; H, 7.78. Found: C, 72.85; H, 7.90. ^{1}H NMR (500 MHz, $CDCl_3$): δ0.80 (s, 3H, 18-H_3), 0.93 (s, 3H, 19-H_3), 0.97 (m, 1H), 1.12 (m, 1H), 1.24–1.32 (m, 3H), 1.40–1.79 (m, 9H), 2.03 (s, 3H, Ac-CH_3), 2.10 (m, 1H), 2.16–2.34 (m, 3H), 3.81 (s, 3H, 4"-OCH_3), 4.26 (d, 1H, *J* = 10.3 Hz, 2-H), 4.59 (t, 1H, *J* = 8.3 Hz, 17-H), 4.82 (d, 1H, *J* = 10.3 Hz, 1-H), 6.88 (d, 2H, *J* = 8.7 Hz, 3"-H and 5"-H), 7.70 (d, 2H, *J* = 8.7 Hz, 2"-H and 6"-H) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ 12.0 (C-18), 12.1 (C-19), 20.4 (CH_2), 21.1 (Ac-CH_3), 23.6 (CH_2), 27.5 (CH_2), 28.7 (CH_2), 30.5 (CH_2), 34.8 (CH), 35.7 (CH), 36.4 (CH_2), 38.7 (C-10), 42.4 (C-13), 42.8 (CH_2), 46.3 (CH), 50.3 (CH), 55.3 (OCH_3), 59.1 (C-2), 82.7 (C-17), 88.3 (C-1), 114.0 (2C), 121.4 (C-1"), 129.1 (2C), 154.7 (C-5'), 161.2 (C-4"), 171.0 (Ac-CO), 206.2 (C-3) ppm; ESI-MS 480 $[M+H]^+$.

*2.2.3. 17β-Acetoxy-3'-2"-tolyl-2'-isoxazolino[4',5'-d:2α,1α]-5α-androstan-3-one (**5c**)*

Compound **1** and *N*-hydroxy-2-methylbenzenecarboximidoyl chloride (**3c**, 255 mg) were used for the synthesis as described in the General Procedure. The crude product **5c** (338 mg, 73%) was obtained as a white precipitate. Mp 182–184 °C; R_f = 0.53 (ss B). Anal. Calcd. for $C_{29}H_{37}NO_4$ (463.61): C, 75.13; H, 8.04. Found: C, 75.20; H, 7.95. ^{1}H NMR (500 MHz, $CDCl_3$): δ0.81 (s, 3H, 18-H_3), 0.93 (s, 3H, 19-H_3), 0.99 (m, 1H), 1.12 (m, 1H), 1.24–1.34 (m, 3H), 1.40–1.79 (m, 9H), 2.03 (s, 3H, Ac-CH_3), 2.11 (m, 1H), 2.18–2.30 (m, 3H), 2.49 (s, 3H, 2"-CH_3), 4.37 (d, 1H, *J* = 10.1 Hz, 2-H), 4.59 (t, 1H, *J* = 8.2 Hz, 17-H), 4.84 (d, 1H, *J* = 10.1 Hz, 1-H), 7.18–7.29 (overlapping m, 3H), 7.38 (d, 1H, *J* = 6.9 Hz) ppm. ^{13}C NMR (125 MHz,

$CDCl_3$): δ 12.0 (C-18), 12.2 (C-19), 20.4 (CH_2), 21.1 (Ac-CH_3), 22.3 (2"-CH_3), 23.6 (CH_2), 27.5 (CH_2), 28.7 (CH_2), 30.5 (CH_2), 34.8 (CH), 35.8 (CH), 36.4 (CH_2), 38.8 (C-10), 42.4 (C-13), 42.7 (CH_2), 46.2 (CH), 50.2 (CH), 60.8 (C-2), 82.7 (C-17), 87.5 (C-1), 125.7 (CH), 127.7 (C-1"), 129.4 (CH), 129.7 (CH), 131.3 (CH), 137.7 (C-2"), 155.9 (C-5'), 171.1 (Ac-CO), 205.4 (C-3) ppm; ESI-MS 464 $[M+H]^+$.

2.2.4. 17β-Acetoxy-3'-3"-tolyl-2'-isoxazolino[4',5'-d:2α,1α]-5α-androstan-3-one ***(5d)***

Compound **1** and *N*-hydroxy-3-methylbenzenecarboximidoyl chloride (**3d**, 255 mg) were used for the synthesis as described in the General Procedure. The crude product **5d** (260 mg, 56%) was obtained as a white precipitate. Mp 213–216 °C; R_f = 0.48 (ss B). Anal. Calcd. for $C_{29}H_{37}NO_4$ (463.61): C, 75.13; H, 8.04. Found: C, 75.25; H, 8.10. ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.81 (s, 3H, 18-H_3), 0.94 (s, 3H, 19-H_3), 0.98 (m, 1H), 1.13 (m, 1H), 1.24–1.33 (m, 3H), 1.39–1.80 (m, 9H), 2.04 (s, 3H, Ac-CH_3), 2.12 (m, 1H), 2.17–2.32 (m, 3H), 2.36 (s, 3H, 3"-CH_3), 4.30 (d, 1H, *J* = 10.0 Hz, 2-H), 4.60 (t, 1H, *J* = 8.4 Hz, 17-H), 4.85 (d, 1H, *J* = 10.0 Hz, 1-H), 7.20 (d, 1H, *J* = 7.3 Hz), 7.26 (t, 1H, *J* = 7.3 Hz, 5"-H), 7.53 (d, 1H, *J* = 7.3 Hz), 7.58 (s, 1H, 2"-H) ppm; ^{13}C NMR (125 MHz, $CDCl_3$): δ 12.0 (C-18), 12.1 (C-19), 20.4 (CH_2), 21.1 (Ac-CH_3), 21.4 (3"-CH_3), 23.6 (CH_2), 27.5 (CH_2), 28.7 (CH_2), 30.5 (CH_2), 34.8 (CH), 35.8 (CH), 36.4 (CH_2), 38.7 (C-10), 42.4 (C-13), 42.8 (CH_2), 46.2 (CH), 50.2 (CH), 58.8 (C-2), 82.7 (C-17), 88.6 (C-1), 124.7 (CH), 127.9 (CH), 128.4 (CH), 128.7 (C-1"), 131.0 (CH), 138.2 (C-3"), 155.4 (C-5'), 171.1 (Ac-CO), 206.1 (C-3) ppm; ESI-MS 464 $[M+H]^+$.

2.2.5. 17β-Acetoxy-3'-4"-tolyl-2'-isoxazolino[4',5'-d:2α,1α]-5α-androstan-3-one ***(5e)***

Compound **1** and *N*-hydroxy-4-methylbenzenecarboximidoyl chloride (**3e**, 255 mg) were used for the synthesis as described in the General Procedure. The crude product **5e** (283

mg, 61%) was obtained as a white precipitate. Mp 215–217 °C; R_f = 0.60 (ss B). Anal. Calcd. for $C_{29}H_{37}NO_4$ (463.61): C, 75.13; H, 8.04. Found: C, 74.96; H, 8.13. ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.80 (s, 3H, 18-H_3), 0.93 (s, 3H, 19-H_3), 0.97 (m, 1H), 1.12 (m, 1H), 1.25–1.31 (m, 3H), 1.38–1.78 (m, 9H), 2.03 (s, 3H, Ac-CH_3), 2.11 (m, 1H), 2.15–2.32 (m, 3H), 2.35 (s, 3H, 4"-CH_3), 4.28 (d, 1H, J = 10.2 Hz, 2-H), 4.58 (t, 1H, J = 8.5 Hz, 17-H), 4.83 (d, 1H, J = 10.2 Hz, 1-H), 7.17 (d, 2H, J = 7.9 Hz, 3"-H and 5"-H), 7.63 (d, 2H, J = 7.9 Hz, 2"-H and 6"-H) ppm; ^{13}C NMR (125 MHz, $CDCl_3$): δ 12.0 (C-18), 12.1 (C-19), 20.4 (CH_2), 21.2 (Ac-CH_3), 21.4 (4"-CH_3), 23.6 (CH_2), 27.5 (CH_2), 28.7 (CH_2), 30.5 (CH_2), 34.8 (CH), 35.7 (CH), 36.4 (CH_2), 38.7 (C-10), 42.4 (C-13), 42.8 (CH_2), 46.2 (CH), 50.2 (CH), 58.8 (C-2), 82.7 (C-17), 88.4 (C-1), 125.9 (C-1"), 127.4 (2C), 129.2 (2C), 140.4 (C-4"), 155.2 (C-5'), 171.1 (Ac-CO), 206.1 (C-3) ppm; ESI-MS 464 $[M+H]^+$.

*2.2.6. 17β-Acetoxy-3'-4"-chlorophenyl-2'-isoxazolino[4',5'-d:2α,1α]-5α-androstan-3-one (**5f**)*

Compound **1** and *N*-hydroxy-4-chlorobenzenecarboximidoyl chloride (**3f**, 292 mg) were used for the synthesis as described in the General Procedure. The crude product **5f** (194 mg, 40%) was obtained as a white precipitate. Mp 200–202 °C; R_f = 0.62 (ss B). Anal. Calcd. for $C_{28}H_{34}ClNO_4$ (484.03): C, 69.48; H, 7.08. Found: C, 69.59; H, 6.97. ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.80 (s, 3H, 18-H_3), 0.95 (s, 3H, 19-H_3), 0.98 (m, 1H), 1.12 (m, 1H), 1.24–1.33 (m, 3H), 1.39–1.80 (m, 9H), 2.04 (s, 3H, Ac-CH_3), 2.13 (m, 1H), 2.18–2.34 (m, 3H), 4.23 (d, 1H, J = 10.1 Hz, 2-H), 4.59 (t, 1H, J = 8.3 Hz, 17-H), 4.85 (d, 1H, J = 10.1 Hz, 1-H), 7.34 (d, 2H, J = 8.3 Hz, 3"-H and 5"-H), 7.70 (d, 2H, J = 8.3 Hz, 2"-H and 6"-H) ppm; ^{13}C NMR (125 MHz, $CDCl_3$): δ 12.0 (C-18), 12.2 (C-19), 20.4 (CH_2), 21.2 (Ac-CH_3), 23.6 (CH_2), 27.5 (CH_2), 28.6 (CH_2), 30.5 (CH_2), 34.8 (CH), 35.7

(CH), 36.3 (CH_2), 38.5 (C-10), 42.4 (C-13), 42.7 (CH_2), 46.3 (CH), 50.2 (CH), 58.3 (C-2), 82.7 (C-17), 88.7 (C-1), 127.3 (C-1"), 128.7 (2C), 128.8 (2C), 136.3 (C-4"), 154.6 (C-5'), 171.1 (Ac-CO), 205.8 (C-3) ppm; ESI-MS 485 $[M+H]^+$.

*2.2.7. 17β-Acetoxy-3'-4"-nitrophenyl-2'-isoxazolino[4',5'-d:2α,1α]-5α-androstan-3-one (**5g**)*

Compound **1** and *N*-hydroxy-4-nitrobenzenecarboximidoyl chloride (**3g**, 300 mg) were used for the synthesis as described in the General Procedure. The crude product **5g** (94 mg, 19%) was obtained as a pale-yellow precipitate. Mp 206–209 °C; R_f = 0.64 (ss B). Anal. Calcd. for $C_{28}H_{34}N_2O_6$ (494.58): C, 68.00; H, 6.93. Found: C, 68.16; H, 6.82. ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.81 (s, 3H, 18-H_3), 0.96 (m, 1H), 1.00 (s, 3H, 19-H_3), 1.13 (m, 1H), 1.26–1.36 (m, 3H), 1.42–1.80 (m, 9H), 2.04 (s, 3H, Ac-CH_3), 2.16–2.27 (m, 3H), 2.32 (m, 1H), 4.25 (d, 1H, *J* = 9.6 Hz, 2-H), 4.60 (t, 1H, *J* = 8.6 Hz, 17-H), 4.89 (d, 1H, *J* = 9.6 Hz, 1-H), 7.94 (d, 2H, *J* = 8.9 Hz, 3"-H and 5"-H), 8.22 (d, 2H, *J* = 8.9 Hz, 2"-H and 6"-H) ppm; ^{13}C NMR (125 MHz, $CDCl_3$): δ 12.0 (C-18), 12.2 (C-19), 20.4 (CH_2), 21.1 (Ac-CH_3), 23.6 (CH_2), 27.5 (CH_2), 28.5 (CH_2), 30.5 (CH_2), 34.8 (CH), 35.9 (CH), 36.3 (CH_2), 38.4 (C-10), 42.4 (C-13), 42.7 (CH_2), 46.4 (CH), 50.2 (CH), 57.5 (C-2), 82.6 (C-17), 89.3 (C-1), 123.7 (2C), 128.3 (2C), 135.0 (C-1"), 148.4 (C-4"), 154.6 (C-5'), 171.1 (Ac-CO), 205.4 (C-3) ppm; ESI-MS 495 $[M+H]^+$.

*2.2.8. 3β-Acetoxy-3'-phenyl-2'-isoxazolino[4',5'-d:16β,15β]-5α-androstan-17-one (**6a**)*

Compound **2** and *N*-hydroxybenzenecarboximidoyl chloride (**3a**, 233 mg) were used for the synthesis as described in the General Procedure. The crude product **6a** (423 mg, 94%) was obtained as a white precipitate. Mp 281–283 °C; R_f = 0.43 (ss A). Anal. Calcd. for $C_{28}H_{35}NO_4$ (449.58): C, 74.80; H, 7.85. Found: C, 74.95; H, 7.74. ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.82

(m, 1H), 0.90 (s, 3H, 19-H_3), 1.06 (m, 1H and s, 3H, 18-H_3), 1.13–1.33 (overlapping m, 3H), 1.35–1.44 (overlapping m, 4H), 1.52 (m, 1H), 1.61–1.85 (overlapping m, 6H), 2.03 (s, 3H Ac-CH_3), 2.08 (m, 1H), 2.15 (m, 1H), 4.14 (d, 1H, J = 9.2 Hz, 16-H), 4.71 (m, 1H, 3-H), 5.42 (dd, 1H, J = 9.2 Hz, J = 4.3 Hz, 15-H), 7.41 (m, 3H, 3″-H, 4″-H and 5″-H), 7.93 (m, 2H, 2″-H and 6″-H) ppm; ^{13}C NMR ($CDCl_3$, 125 MHz): δ 12.2 (C-19), 17.8 (C-18), 20.2 (CH_2), 21.4 (Ac-CH_3), 27.4 (CH_2), 28.0 (CH_2), 30.4 (CH_2), 32.1 (CH), 33.3 (CH_2), 33.9 (CH_2), 35.8 (C-10), 36.6 (CH_2), 44.8 (CH), 48.9 (C-13), 54.3 (CH), 55.0 (CH), 62.1 (CH), 73.4 (C-3), 84.6 (C-15), 127.6 (2C, C-3″ and C-5″), 128.3 (C-1″), 128.7 (2C, C-2″ and C-6″), 130.3 (C-4″), 155.3 (C-3′), 170.6 (Ac-CO), 212.3 (C-17) ppm; ESI-MS 450 $[M+H]^+$.

*2.2.9. 3β-Acetoxy-3'-4"-methoxyphenyl-2'-isoxazolino[4',5'-d:16β,15β]-5α-androstan-17-one (**6b**)*

Compound **2** and *N*-hydroxy-4-methoxybenzenecarboximidoyl chloride (**3b**, 279 mg) were used for the synthesis as described in the General Procedure. The crude product **6b** (470 mg, 98%) was obtained as a white precipitate. Mp 232–234 °C; R_f = 0.33 (ss A). Anal. Calcd. for $C_{29}H_{37}NO_5$ (479.61): C, 72.62; H, 7.78. Found: C, 72.45; H, 7.70. ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.81 (m, 1H), 0.90 (s, 3H, 19-H_3), 1.05 (m, 1H and s, 3H, 18-H_3), 1.11–1.32 (overlapping m, 3H), 1.34–1.43 (overlapping m, 4H), 1.52 (m, 1H), 1.59–1.69 (overlapping m, 3H), 1.75 (m, 2H), 1.83 (m, 1H), 2.02 (s, 3H Ac-CH_3), 2.07 (m, 1H), 2.14 (m, 1H), 3.82 (s, 3H, 4″-OMe), 4.11 (d, 1H, J = 9.1 Hz, 16-H), 4.70 (m, 1H, 3-H), 5.38 (dd, 1H, J = 9.1 Hz, J = 4.2 Hz, 15-H), 6.92 (d, 2H, J = 8.8Hz, 3″-H and 5″-H), 7.86 (d, 2H, J = 8.8 Hz, 2″-H and 6″-H) ppm; ^{13}C NMR ($CDCl_3$, 125 MHz): δ 12.1 (C-19), 17.9 (C-18), 20.2 (CH_2), 21.4 (Ac-CH_3), 27.4 (CH_2), 28.0 (CH_2), 30.4 (CH_2), 32.1 (CH), 33.2 (CH_2), 33.9 (CH_2), 35.8 (C-10), 36.6 (CH_2), 44.8 (CH), 48.9 (C-13), 54.3 (CH), 55.0 (CH), 55.3 (4″-OMe), 62.4 (CH), 73.4

(C-3), 84.1 (C-15), 114.1 (2C, C-3″ and C-5″), 120.9 (C-1″), 129.2 (2C, C-2″ and C-6″), 154.8 (C-4″), 161.2 (C-3′), 170.6 (Ac-CO), 212.6 (C-17) ppm; ESI-MS 480 $[M+H]^+$.

*2.2.10. 3β-Acetoxy-3′-4″-tolyl-2′-isoxazolino[4′,5′-d:16β,15β]-5α-androstan-17-one (**6e**)*

Compound **2** and *N*-hydroxy-4-methylbenzenecarboximidoyl chloride (**3e**, 255 mg) were used for the synthesis as described in the General Procedure. The crude product **6e** (450 mg, 97%) was obtained as a white precipitate. Mp 273–276 °C; R_f = 0.27 (ss A). Anal. Calcd. for $C_{29}H_{37}NO_4$ (463.61): C, 75.13; H, 8.04. Found: C, 75.25; H, 8.10. ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.81 (m, 1H), 0.90 (s, 3H, 19-H_3), 1.05 (m, 1H and s, 3H, 18-H_3), 1.12–1.32 (overlapping m, 3H), 1.35–1.42 (overlapping m, 4H), 1.52 (m, 1H), 1.60–1.67 (overlapping m, 3H), 1.78 (m, 2H), 1.83 (m, 1H), 2.03 (s, 3H Ac-CH_3), 2.07 (m, 1H), 2.15 (m, 1H), 2.37 (s, 3H, 4″-CH_3), 4.12 (d, 1H, *J* = 9.1 Hz, 16-H), 4.71 (m, 1H, 3-H), 5.39 (dd, 1H, *J* = 9.1 Hz, *J* = 4.1 Hz, 15-H), 7.21 (d, 2H, *J* = 7.8 Hz, 3″-H and 5″-H), 7.81 (d, 2H, *J* = 7.8 Hz, 2″-H and 6″-H) ppm; ^{13}C NMR ($CDCl_3$, 125 MHz): δ 12.1 (C-19), 17.8 (C-18), 20.1 (CH_2), 21.4 (2C, Ac-CH_3 and 4″-CH_3), 27.4 (CH_2), 28.0 (CH_2), 30.4 (CH_2), 32.1 (CH), 33.2 (CH_2), 33.9 (CH_2), 35.8 (C-10), 36.6 (CH_2), 44.8 (CH), 48.9 (C-13), 54.3 (CH), 55.0 (CH), 62.2 (CH), 73.4 (C-3), 84.3 (C-15), 125.5 (C-1″), 127.6 (2C, C-2″ and C-6″), 129.4 (2C, C-3″ and C-5″), 140.6 (C-4″), 155.3 (C-3′), 170.6 (Ac-CO), 212.4 (C-17) ppm; ESI-MS 464 $[M+H]^+$.

*2.2.11. 3β-Acetoxy-3′-4″-chlorophenyl-2′-isoxazolino[4′,5′-d:16β,15β]-5α-androstan-17-one (**6f**)*

Compound **2** and *N*-hydroxy-4-chlorobenzenecarboximidoyl chloride (**3f**, 292 mg) were used for the synthesis as described in the General Procedure. The crude product **6f** (378 mg, 78%) was obtained as a white precipitate. Mp 242–245 °C; R_f = 0.50 (ss A). Anal.

Calcd. for $C_{28}H_{34}ClNO_4$ (484.03): C, 69.48; H, 7.08. Found: C, 69.37; H, 7.18. ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.82 (m, 1H), 0.90 (s, 3H, 19-H_3), 1.03 (s, 3H, 18-H_3), 1.04 (m, 1H), 1.13–1.43 (overlapping m, 7H), 1.52 (m, 1H), 1.61–1.84 (overlapping m, 6H), 2.02 (s, 3H Ac-CH_3), 2.07 (m, 1H), 2.14 (m, 1H), 4.09 (d, 1H, J = 8.9 Hz, 16-H), 4.70 (m, 1H, 3-H), 5.42 (dd, 1H, J = 8.9 Hz, J = 4.2 Hz, 15-H), 8.11 (d, 2H, J = 7.3 Hz, 3″-H and 5″-H), 8.26 (d, 2H, J = 7.3 Hz, 2″-H and 6″-H) ppm; ^{13}C NMR ($CDCl_3$, 125 MHz): δ 12.1 (C-19), 17.9 (C-18), 20.1 (CH_2), 21.4 (Ac-CH_3), 27.3 (CH_2), 28.0 (CH_2), 30.4 (CH_2), 32.1 (CH), 33.2 (CH_2), 33.8 (CH_2), 35.8 (C-10), 36.6 (CH_2), 44.8 (CH), 48.9 (C-13), 54.3 (CH), 54.9 (CH), 61.9 (CH), 73.3 (C-3), 84.8 (C-15), 126.8 (C-1″), 128.8 (2C, C-3″ and C-5″), 128.9 (2C, C-2″ and C-6″), 136.4 (C-4″), 154.4 (C-3′), 170.6 (Ac-CO), 212.3 (C-17) ppm; ESI-MS 485 $[M+H]^+$.

*2.2.12. 3β-Acetoxy-3'-4"-nitrophenyl-2'-isoxazolino[4',5'-d:16β,15β]-5α-androstan-17-one (**6g**)*

Compound **2** and *N*-hydroxy-4-nitrobenzenecarboximidoyl chloride (**3g**, 300 mg) were used for the synthesis as described in the General Procedure. The crude product **6g** (223 mg, 45%) was obtained as a yellow precipitate. Mp > 250 °C (decomp.); R_f = 0.38 (ss A). Anal. Calcd. for $C_{28}H_{34}N_2O_6$ (494.58): C, 68.00; H, 6.93. Found: C, 67.87; H, 7.05. ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.82 (m, 1H), 0.90 (s, 3H, 19-H_3), 1.03 (s, 3H, 18-H_3), 1.06 (m, 1H), 1.19–1.43 (overlapping m, 7H), 1.51 (m, 1H), 1.65–1.81 (overlapping m, 6H), 2.03 (s, 3H Ac-CH_3), 2.08 (m, 1H), 2.14 (m, 1H), 4.14 (d, 1H, J = 9.1 Hz, 16-H), 4.70 (m, 1H, 3-H), 5.50 (dd, 1H, J = 9.1 Hz, J = 4.0 Hz, 15-H), 8.11 (d, 2H, J = 8.6 Hz, 2″-H and 6″-H), 8.26 (d, 2H, J = 8.6 Hz, 3″-H and 5″-H) ppm; ^{13}C NMR ($CDCl_3$, 125 MHz): δ 12.2 (C-19), 17.9 (C-18), 20.1 (CH_2), 21.4 (Ac-CH_3), 27.3 (CH_2), 28.0 (CH_2), 30.4 (CH_2), 32.1 (CH), 33.2 (CH_2), 33.8 (CH_2), 35.8 (C-10), 36.6 (CH_2), 44.8 (CH), 49.0 (C-13), 54.4

(CH), 54.9 (CH), 61.4 (CH), 73.3 (C-3), 85.8 (C-15), 123.9 (2C, C-3″ and C-5″), 128.3 (2C, C-2″ and C-6″), 134.4 (C-1″), 148.6 (C-4″), 154.0 (C-3′), 170.7 (Ac-CO), 212.0 (C-17) pppm; ESI-MS 495 $[M+H]^+$.

*2.2.13. 17β-Hydroxy-3'-phenyl-2'-isoxazolino[4',5'-d:2α,1α]-5α-androstan-3-one (**9a**)*

Compound **13** and *N*-hydroxybenzenecarboximidoyl chloride (**3a**, 233 mg) were used for the synthesis as described in the General Procedure. The crude product **9a** (179 mg, 44%) was obtained as a white precipitate. Mp 206–208 °C; R_f = 0.41 (ss B). Anal. Calcd. for $C_{26}H_{33}NO_3$ (407.55): C, 76.62; H, 8.16. Found: C, 76.77; H, 8.26. ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.76 (s, 3H, 18-H_3), 0.95 (s, 3H, 19-H_3), 1.04 (m, 1H), 1.18–1.31 (m, 4H), 1.40–1.84 (m, 9H), 2.04–2.16 (m, 2H), 2.23–2.35 (m, 2H), 3.68 (t, 1H, J = 8.5 Hz, 17-H), 4.30 (d, 1H, J = 9.8 Hz, 2-H), 4.86 (d, 1H, J = 9.8 Hz, 1-H), 7.38 (overlapping m, 3H, 3"-H, 4"-H and 5"-H), 7.75 (d, 2H, J = 7.4 Hz, 2"-H and 6"-H) ppm; ^{13}C NMR (125 MHz, $CDCl_3$): δ 11.1 (C-18), 12.2 (C-19), 20.6 (CH_2), 23.4 (CH_2), 28.7 (CH_2), 30.3 (CH_2), 30.6 (CH_2), 35.1 (CH), 35.9 (CH), 36.2 (CH_2), 38.7 (C-10), 42.7 (CH_2), 42.8 (C-13), 46.4 (CH), 50.5 (CH), 58.7 (C-2), 81.7 (C-17), 88.7 (C-1), 127.5 (2C), 128.5 (2C), 128.8 (C-1"), 130.2 (C-4"), 155.4 (C-5'), 206.0 (C-3) ppm; ESI-MS 408 $[M+H]^+$.

*2.2.14. 17β-Hydroxy-3'-4"-methoxyphenyl-2'-isoxazolino[4',5'-d:2α,1α]-5α-androstan-3-one (**9b**)*

Compound **13** and *N*-hydroxy-4-methoxybenzenecarboximidoyl chloride (**3b**, 279 mg) were used for the synthesis as described in the General Procedure. The crude product **9b** (289 mg, 66%) was obtained as a white precipitate. Mp 225–228 °C; R_f = 0.31 (ss D). Anal. Calcd. for $C_{27}H_{35}NO_4$ (437.57): C, 74.11; H, 8.06. Found: C, 73.96; H, 8.12. ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.75 (s, 3H, 18-H_3), 0.94 (s, 3H, 19-H_3), 1.03 (m, 1H), 1.17–1.30 (m, 4H),

1.37–1.84 (m, 9H), 2.02–2.14 (m, 2H), 2.22–2.34 (m, 2H), 3.67 (t, 1H, J = 8.3 Hz, 17-H), 3.81 (s, 3H, 4"-OCH_3), 4.27 (d, 1H, J = 10.3 Hz, 2-H), 4.83 (d, 1H, J = 10.3 Hz, 1-H), 6.88 (d, 2H, J = 8.7 Hz, 3"-H and 5"-H), 7.70 (d, 2H, J = 8.7 Hz, 2"-H and 6"-H) ppm; ^{13}C NMR (125 MHz, $CDCl_3$): δ 11.0 (C-18), 12.2 (C-19), 20.5 (CH_2), 23.4 (CH_2), 28.7 (CH_2), 30.3 (CH_2), 30.6 (CH_2), 35.1 (CH), 35.8 (CH), 36.2 (CH_2), 38.7 (C-10), 42.7 (CH_2), 42.8 (C-13), 46.4 (CH), 50.6 (CH), 55.3 (OCH_3), 58.9 (C-2), 81.7 (C-17), 88.3 (C-1), 114.0 (2C), 121.3 (C-1"), 129.1 (2C), 154.8 (C-5'), 161.1 (C-4"), 206.3 (C-3) ppm; ESI-MS 438 $[M+H]^+$.

*2.2.15. 17β-Hydroxy-3'-2"-tolyl-2'-isoxazolino[4',5'-d:2α,1α]-5α-androstan-3-one (**9c**)*

Compound **13** and *N*-hydroxy-2-methylbenzenecarboximidoyl chloride (**3c**, 255 mg) were used for the synthesis as described in the General Procedure. The crude product **9c** (261 mg, 62%) was obtained as a white precipitate. Mp 163–166 °C; R_f = 0.40 (ss B). Anal. Calcd. for $C_{27}H_{35}NO_3$ (421.57): C, 76.92; H, 8.37. Found: C, 77.05; H, 8.25. ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.76 (s, 3H, 18-H_3), 0.94 (s, 3H, 19-H_3), 1.04 (m, 1H), 1.18–1.32 (m, 4H), 1.40–1.85 (m, 9H), 2.04–2.18 (m, 2H), 2.22–2.32 (m, 2H), 2.50 (s, 3H, 2"-CH_3), 3.67 (t, 1H, J = 8.2 Hz, 17-H), 4.38 (d, 1H, J = 10.1 Hz, 2-H), 4.84 (d, 1H, J = 10.1 Hz, 1-H), 7.20–7.30 (overlapping m, 3H), 7.38 (d, 1H, J = 6.9 Hz) ppm; ^{13}C NMR (125 MHz, $CDCl_3$): δ 11.0 (C-18), 12.2 (C-19), 20.6 (CH_2), 22.3 (2"-CH_3), 23.4 (CH_2), 28.8 (CH_2), 30.3 (CH_2), 30.6 (CH_2), 35.1 (CH), 35.9 (CH), 36.2 (CH_2), 38.8 (C-10), 42.7 (CH_2), 42.8 (C-13), 46.4 (CH), 50.6 (CH), 60.7 (C-2), 81.7 (C-17), 87.6 (C-1), 125.7 (CH), 127.7 (C-1"), 129.5 (CH), 129.7 (CH), 131.3 (CH), 137.7 (C-2"), 156.0 (C-5'), 205.5 (C-3) ppm; ESI-MS 422 $[M+H]^+$.

*2.2.16. 17β-Hydroxy-3'-3"-tolyl-2'-isoxazolino[4',5'-d:2α,1α]-5α-androstan-3-one (**9d**)*

Compound **13** and *N*-hydroxy-3-methylbenzenecarboximidoyl chloride (**3d**, 255 mg) were used for the synthesis as described in the General Procedure. The crude product **9d** (215

mg, 51%) was obtained as a white precipitate. Mp 175–177 °C; R_f = 0.42 (ss B). Anal. Calcd. for $C_{27}H_{35}NO_3$ (421.57): C, 76.92; H, 8.37. Found: C, 76.80; H, 8.22. ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.76 (s, 3H, 18-H_3), 0.95 (s, 3H, 19-H_3), 1.04 (m, 1H), 1.18–1.30 (m, 4H), 1.40–1.85 (m, 9H), 2.04–2.16 (m, 2H), 2.22–2.31 (m, 2H), 2.36 (s, 3H, 3"-CH_3), 3.68 (t, 1H, *J* = 8.4 Hz, 17-H), 4.29 (d, 1H, *J* = 10.0 Hz, 2-H), 4.85 (d, 1H, *J* = 10.0 Hz, 1-H), 7.20 (d, 1H, *J* = 7.3 Hz), 7.26 (t, 1H, *J* = 7.3 Hz, 5"-H), 7.53 (d, 1H, *J* = 7.3 Hz), 7.58 (s, 1H, 2"-H) ppm; ^{13}C NMR (125 MHz, $CDCl_3$): δ 11.0 (C-18), 12.2 (C-19), 20.6 (CH_2), 21.4 (3"-CH_3), 23.4 (CH_2), 28.7 (CH_2), 30.3 (CH_2), 30.6 (CH_2), 35.1 (CH), 35.9 (CH), 36.2 (CH_2), 38.7 (C-10), 42.8 (C-13), 42.9 (CH_2), 46.4 (CH), 50.5 (CH), 58.8 (C-2), 81.7 (C-17), 88.6 (C-1), 124.7 (CH), 127.9 (CH), 128.4 (CH), 128.7 (C-1"), 131.0 (CH), 138.2 (C-3"), 155.5 (C-5'), 206.1 (C-3) ppm; ESI-MS 422 $[M+H]^+$.

*2.2.17. 17β-Hydroxy-3'-4"-tolyl-2'-isoxazolino[4',5'-d:2α,1α]-5α-androstan-3-one (**9e**):*

Compound **13** and *N*-hydroxy-4-methyl-benzenecarboximidoyl chloride (**3e**, 255 mg) were used for the synthesis as described in the General Procedure. The crude product **9e** (223 mg, 53%) was obtained as a white precipitate. Mp 232–234 °C; R_f = 0.45 (ss B). Anal. Calcd. for $C_{27}H_{35}NO_3$ (421.57): C, 76.92; H, 8.37. Found: C, 77.08; H, 8.45. ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.76 (s, 3H, 18-H_3), 0.94 (s, 3H, 19-H_3), 1.05 (m, 1H), 1.18–1.31 (m, 4H), 1.40–1.84 (m, 9H), 2.02–2.14 (m, 2H), 2.22–2.33 (m, 2H), 2.36 (s, 3H, 4"-CH_3), 3.68 (t, 1H, *J* = 8.5 Hz, 17-H), 4.28 (d, 1H, *J* = 10.2 Hz, 2-H), 4.84 (d, 1H, *J* = 10.2 Hz, 1-H), 7.17 (d, 2H, *J* = 7.9 Hz, 3"-H and 5"-H), 7.63 (d, 2H, *J* = 7.9 Hz, 2"-H and 6"-H) ppm; ^{13}C NMR (125 MHz, $CDCl_3$): δ 11.0 (C-18), 12.2 (C-19), 20.5 (CH_2), 21.4 (4"-CH_3), 23.4 (CH_2), 28.7 (CH_2), 30.3 (CH_2), 30.6 (CH_2), 35.1 (CH), 35.9 (CH), 36.2 (CH_2), 38.7 (C-10), 42.7 (CH_2), 42.8 (C-13), 46.4 (CH), 50.6 (CH), 58.8 (C-2), 81.7 (C-17), 88.5 (C-1), 125.9 (C-1"), 127.4 (2C), 129.2 (2C), 140.5 (C-4"), 155.3 (C-5'), 206.1

(C-3) ppm; ESI-MS 422 $[M+H]^+$.

*2.2.18. 17β-Hydroxy-3'-4"-chlorophenyl-2'-isoxazolino[4',5'-d:2α,1α]-5α-androstan-3-one (**9f**):*

Compound **13** and *N*-hydroxy-4-chlorobenzenecarboximidoyl chloride (**3f**, 292 mg) were used for the synthesis as described in the General Procedure. The crude product **9f** (164 mg, 37%) was obtained as a white precipitate. Mp 210–212 °C; R_f = 0.41 (ss B). Anal. Calcd. for $C_{26}H_{32}ClNO_3$ (441.99): C, 70.65; H, 7.30. Found: C, 70.82; H, 7.52. ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.76 (s, 3H, 18-H_3), 0.96 (s, 3H, 19-H_3), 1.04 (m, 1H), 1.17–1.32 (m, 4H), 1.40–1.84 (m, 9H), 2.04–2.34 (m, 4H), 3.67 (t, 1H, *J* = 8.3 Hz, 17-H), 4.23 (d, 1H, *J* = 10.1 Hz, 2-H), 4.85 (d, 1H, *J* = 10.1 Hz, 1-H), 7.34 (d, 2H, *J* = 8.3 Hz, 3"-H and 5"-H), 7.70 (d, 2H, *J* = 8.3 Hz, 2"-H and 6"-H) ppm; ^{13}C NMR (125 MHz, $CDCl_3$): δ 11.1 (C-18), 12.2 (C-19), 20.5 (CH_2), 23.4 (CH_2), 27.5 (CH_2), 28.6 (CH_2), 30.5 (CH_2), 34.9 (CH), 35.8 (CH), 36.3 (CH_2), 38.5 (C-10), 42.4 (C-13), 42.7 (CH_2), 46.4 (CH), 50.4 (CH), 58.3 (C-2), 81.7 (C-17), 88.7 (C-1), 127.4 (C-1"), 128.7 (2C), 128.8 (2C), 136.3 (C-4"), 154.7 (C-5'), 205.8 (C-3) ppm; ESI-MS 443 $[M+H]^+$.

*2.2.19. 3β-Hydroxy-3'-phenyl-2'-isoxazolino[4',5'-d:16β,15β]-5α-androstan-17-one (**11a**):*

Compound **14** and *N*-hydroxybenzenecarboximidoyl chloride (**3a**, 233 mg) were used for the synthesis as described in the General Procedure. The crude product **11a** (375 mg, 92%) was obtained as a white precipitate. Mp > 280 °C (decomp.); R_f = 0.43 (ss D). Anal. Calcd. for $C_{26}H_{33}NO_3$ (407.55): C, 76.62; H, 8.16. Found: C, 76.85; H, 8.22. ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.81 (m, 1H), 0.90 (s, 3H, 19-H_3), 1.02 (m, 1H), 1.07 (s, 3H, 18-H_3), 1.19 (m, 2H), 1.26–1.47 (overlapping m, 6H), 1.62 (m, 2H), 1.68–1.85 (overlapping m, 4H), 2.08 (m, 1H), 2.16 (m, 1H), 3.61 (m, 1H, 3-H), 4.14 (d, 1H, *J* = 9.2 Hz, 16-H), 5.43 (dd, 1H, *J* = 9.2 Hz, *J* =

4.3 Hz, 15-H), 7.42 (m, 3H, 3″-H, 4″-H and 5″-H), 7.94 (m, 2H, 2″-H and 6″-H) ppm; ESI-MS 408 $[M+H]^+$.

*2.2.20. 3β-Hydroxy-3'-4"-methoxyphenyl-2'-isoxazolino[4',5'-d:16β,15β]-5α-androstan-17-one (**11b**)*

Compound **14** and *N*-hydroxy-4-methoxybenzenecarboximidoyl chloride (**3b**, 279 mg) were used for the synthesis as described in the General Procedure. The crude product **11b** (424 mg, 97%) was obtained as a white precipitate. Mp 205–208 °C; R_f = 0.24 (ss D). Anal. Calcd. for $C_{27}H_{35}NO_4$ (437.57): C, 74.11; H, 8.06. Found: C, 73.95; H, 8.22. ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.79 (m, 1H), 0.89 (s, 3H, 19-H_3), 1.06 (m, 1H and s, 3H, 18-H_3), 1.12–1.33 (overlapping m, 3H), 1.35–1.43 (overlapping m, 4H), 1.53 (m, 1H), 1.62 (m, 2H), 1.67–1.84 (overlapping m, 4H), 2.08 (m, 1H), 2.14 (m, 1H), 3.61 (m, 1H, 3-H), 3.84 (s, 3H, 4″-OMe), 4.11 (d, 1H, J = 9.1 Hz, 16-H), 5.39 (dd, 1H, J = 9.1 Hz, J = 4.2 Hz, 15-H), 6.93 (d, 2H, J = 8.8 Hz, 3″-H and 5″-H), 7.88 (d, 2H, J = 8.8 Hz, 2″-H and 6″-H) ppm; ESI-MS 438 $[M+H]^+$.

*2.2.21. 3β-Hydroxy-3'-4"-tolyl-2'-isoxazolino[4',5'-d:16β,15β]-5α-androstan-17-one (**11e**)*

Compound **14** and *N*-hydroxy-4-methylbenzenecarboximidoyl chloride (**3e**, 255 mg) were used for the synthesis as described in the General Procedure. The crude product **11e** (396 mg, 94%) was obtained as a white precipitate. Mp 221–224 °C; R_f = 0.29 (ss D). Anal. Calcd. for $C_{27}H_{35}NO_3$ (421.57): C, 76.92; H, 8.37. Found: C, 76.80; H, 8.44. ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.79 (m, 1H), 0.89 (s, 3H, 19-H_3), 1.06 (m, 1H and s, 3H, 18-H_3), 1.14–1.33 (overlapping m, 3H), 1.35–1.43 (overlapping m, 4H), 1.56–1.84 (overlapping

m, 7H), 2.07 (m, 1H), 2.15 (m, 1H), 2.37 (s, 3H, 4″-CH_3), 3.61 (m, 1H, 3-H), 4.13 (d, 1H, *J* = 9.1 Hz, 16-H), 5.40 (dd, 1H, *J* = 9.0 Hz, *J* = 4.1 Hz, 15-H), 7.22 (d, 2H, *J* = 7.7 Hz, 3″-H and 5″-H), 7.82 (d, 2H, *J* = 7.7 Hz, 2″-H and 6″-H) ppm; ESI-MS 422 $[M+H]^+$.

*2.2.22. 3β-Hydroxy-3'-4"-chlorophenyl-2'-isoxazolino[4',5'-d:16β,15β]-5α-androstan-17-one (**11f**):*

Compound **14** and *N*-hydroxy-4-chlorobenzenecarboximidoyl chloride (**3f**, 292 mg) were used for the synthesis as described in the General Procedure. The crude product **11f** (340 mg, 77%) was obtained as a white precipitate. Mp > 280 °C (decomp.); R_f = 0.31 (ss D). Anal. Calcd. for $C_{26}H_{32}ClNO_3$ (441.99): C, 70.65; H, 7.30. Found: C, 70.47; H, 7.21. ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.80 (m, 1H), 0.89 (s, 3H, 19-H_3), 1.04 (m, 1H), 1.05 (s, 3H, 18-H_3), 1.18 (m, 2H), 1.26–1.47 (overlapping m, 6H), 1.62 (m, 2H), 1.68–1.85 (overlapping m, 4H), 2.08 (m, 1H), 2.15 (m, 1H), 3.61 (m, 1H, 3-H), 4.09 (d, 1H, *J* = 9.2 Hz, 16-H), 5.43 (dd, 1H, *J* = 9.2 Hz, *J* = 4.3 Hz, 15-H), 7.39 (d, 2H, *J* = 8.5 Hz, 3″-H and 5″-H), 7.87 (d, 2H, *J* = 8.5 Hz, 3″-H and 5″-H) ppm; ESI-MS 442 $[M+H]^+$.

*2.2.23. 3β-Hydroxy-3'-4"-nitrophenyl-2'-isoxazolino[4',5'-d:16β,15β]-5α-androstan-17-one (**11g**):*

Compound **14** and *N*-hydroxy-4-nitrobenzenecarboximidoyl chloride (**3g**, 300 mg) were used for the synthesis as described in the General Procedure. The crude product **11g** (199 mg, 44%) was obtained as a yellow precipitate. Mp 210–212 °C; R_f = 0.30 (ss D). Anal. Calcd. for $C_{26}H_{32}N_2O_5$ (452.54): C, 69.01; H, 7.13. Found: C, 69.16; H, 7.01. ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.81 (m, 1H), 0.90 (s, 3H, 19-H_3), 1.02 (m, 1H), 1.04 (s, 3H, 18-H_3), 1.19 (m, 2H), 1.26–1.47 (overlapping m, 6H), 1.62 (m, 2H), 1.68–1.85 (overlapping m, 4H), 2.08–2.17 (m, 2H), 3.61 (m, 1H, 3-H), 4.14 (d, 1H, *J* = 9.0 Hz, 16-H), 5.51 (dd,

1H, *J* = 9.0 Hz, *J* = 4.5 Hz, 15-H), 8.12 (d, 2H, *J* = 9.0 Hz, 2″-H and 6″-H), 8.27 (d, 2H, *J* = 9.0 Hz, 3″-H and 5″-H) ppm; ESI-MS 453 $[M+H]^+$.

*2.3. Deacetylation of 17β-acetoxy-3'-4"-methoxyphenyl-2'-isoxazolino[4',5'-d:2α,1α]-5α-androstan-3-one (**5b**)*

Method A: Compound **5b** (150 mg, 0.31 mmol) was dissolved in MeOH (10 mL), and KOH (50 mg, 0.89 mmol) was added. The solution was stirred at room temperature for 3 h and then diluted with water, and the precipitate that formed was filtered off, washed with water and purified by flash chromatography with EtOAc/CH_2Cl_2 = 20:80 as eluent to obtain **7b** (100 mg, 74%) and **8b** (25 mg, 18%) as white crystals (sequence of elution: **7b** > **8b**).

Method B: Compound **5b** (150 mg, 0.31 mmol) was dissolved in DMSO (10 mL), and ^{t}BuOK (80 mg, 0.71 mmol) was added. The solution was stirred at 80 °C for 1 h and then diluted with water and extracted with EtOAc (2 × 10 mL). The crude product was purified by flash chromatography with EtOAc/CH_2Cl_2 = 20:80 as eluent to obtain **7b** (120 mg, 89%).

7b: Mp 128−131 °C; R_f = 0.38 (ss D). Anal. Calcd. for $C_{27}H_{33}NO_4$ (435.56): C, 74.45; H, 7.64. Found: C, 74.60; H, 7.48. ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.83 (s, 3H, 18-H_3), 0.96−1.11 (m, 3H), 1.32 (s, 3H, 19-H_3), 1.40−1.63 (m, 7H), 1.70−1.79 (m, 2H), 1.92 (m, 1H), 2.06 (m, 1H), 2.20 (m, 1H), 2.38 (m, 1H), 2.56−2.68 (m, 2H), 3.68 (t, 1H, *J* = 8.3 Hz, 17-H), 3.84 (s, 3H, 4"-OCH_3), 6.97 (d, 2H, *J* = 7.7 Hz, 3"-H and 5"-H), 7.94 (d, 2H, *J* = 7.7 Hz, 2"-H and 6"-H) ppm; ^{13}C NMR (125 MHz, $CDCl_3$): δ 11.4 (C-18), 13.6 (C-19), 23.4 (CH_2), 23.8 (CH_2), 26.5 (CH_2), 29.7 (C-10), 30.2 (CH_2), 30.4 (CH_2), 36.2 (CH), 36.6 (CH_2), 41.4 (C-13), 42.9 (CH_2), 45.1 (CH), 49.5 (CH), 50.8 (CH), 55.3 (4″-OCH_3), 81.6 (C-17), 113.2 (C-2), 113.7 (2C, C-3″ and C-5″), 119.7 (C-1″), 130.8 (2C, C-2″ and C-6″), 159.5 (C-3′), 161.3 (C-4″), 189.8 (C-1), 191.9 ppm; ESI-MS 436 $[M+H]^+$.

8b: Mp 239–242 °C; R_f = 0.34 (ss E). Anal. Calcd. for $C_{27}H_{35}NO_5$ (453,57): C, 71.50; H, 7.78. Found: C 71.37; H, 7.92. ^{1}H NMR (500 MHz, DMSO-d_6): δ 0.64 (s, 3H, 18-H_3), 0.66 (s, 3H, 19-H_3), 0.88 (m, 2H), 1.01 (m, 1H), 1.12–1.50 (overlapping m, 7H), 1.61 (m, 3H), 1.78 (m, 2H), 1.92 (m, 1H), 2.21 (m, 1H), 2.74 (dd, *J* = 17.1 Hz, *J* = 6.4 Hz, 1H), 3.44 (m, 17-H), 3.78 (s, 3H, 4″-OMe), 4.19 (s, 1H, 1-H), 4.46 (d, J = 4.3 Hz, 17-OH), 6.95 (d, 2H, *J* =8.4 Hz, 3″-H and 5″-H), 7.02 (s, 1H, 2-OH), 7.64 (d, 2H, *J* = 8.4 Hz, 2″-H and 6″-H) ppm; ^{13}C NMR (125 MHz, $CDCl_3$): δ 7.3 (C-19), 11.2 (C-18), 22.6 (CH_2), 23.0 (CH_2), 27.3 (C-10), 29.7 (CH_2), 30.4 (2×CH_2), 34.6 (CH), 36.4 (CH_2), 37.6 (CH), 40.8 (CH_2), 42.4 (C-13), 50.1 (CH), 54.7 (CH), 55.1 (4″-OMe), 79.9 (C-17), 89.0 (C-2), 97.4 (C-1), 113.7 (2C, C-3″ and C-5″), 120.8 (C-1″), 128.8 (2C, C-2″ and C-6″), 156.3 (C-3′), 160.3 (C-4″), 207.0 (C-3) ppm; ESI-MS: 454 $[M+H]^+$.

Method C: Compound **5b** (150 mg, 0.31 mmol) was dissolved in DMSO (10 mL), and ^{t}BuOK (80 mg, 0.71 mmol) was added under a N_2 atmosphere. The solution was stirred at 80 °C for 1 h and then diluted with water and extracted with EtOAc (2 × 10 mL). The crude product was purified by flash chromatography with EtOAc/CH_2Cl_2 = 5:95 as eluent to obtain **9b** (115 mg, 85%). See. *2.2.14.*

*2.4. Deacetylation of 3β-acetoxy-3'-phenyl-2'-isoxazolino[4',5'-d:16β,15β]-5α-androstan-17-one (**6a**)*

Method A: Compound **6a** (135 mg, 0.30 mmol) was dissolved in MeOH (10 mL), and KOH (50 mg, 0.89 mmol) was added. The solution was stirred at room temperature for 8 h and then diluted with water, and the precipitate that formed was filtered off, washed with water and purified by flash chromatography with EtOAc/CH_2Cl_2 = 10:90 as eluent to obtain **10a** (73 mg, 55%) and **11a** (45 mg, 37%) as white crystals (sequence of elution: **11a** > **10a**).

10a: Mp 192–195 °C; R_f = 0.25 (ss D). Anal. Calcd. for $C_{27}H_{37}NO_4$ (439.59): C, 73.77; H, 8.48. Found: C, 73.85; H, 8.60. ^{1}H NMR ($CDCl_3$, 500 MHz): δ0.82 (m, 3H, 19-H_3), 0.85 (m, 1H), 0.98 (m, 1H), 1.11 (m, 2H), 1.29 (m, 4H and s, 3H, 18-H_3), 1.39 (m, 1H), 1.55–1.75 (overlapping m, 8H), 1.81 (m, 1H), 2.26 (m, 1H), 3.09 (dd, 1H, J = 16.5 Hz, J = 9.0 Hz, one of 16-H_2), 3.44 (dd, 1H, J = 16.5 Hz, J = 11.2 Hz, the other 16-H_2), 3.60 (m, 1H, 3-H), 3.62 (s, 3H, 17-OMe), 4.42 (m, 1H, 15-H), 7.38 (m, 3H, 3″-H, 4″-H and 5″-H), 7.63 (m, 2H, 2″-H and 6″-H) ppm; ^{13}C NMR ($CDCl_3$, 125 MHz): δ12.3 (C-19), 16.0 (C-18), 20.1 (CH_2), 28.5 (CH_2), 31.3 (CH_2), 32.6 (CH_2), 35.6 (C-10), 35.7 (CH), 36.9 (CH_2), 37.7 (CH_2), 37.8 (CH_2), 42.4 (C-16), 44.1 (CH), 45.3 (C-13), 51.7 (17-OMe), 52.7 (CH), 53.7 (CH), 71.0 (C-3), 83.0 (C-15), 126.5 (2C, C-2″ and C-6″), 128.6 (2C, C-3″ and C-5″), 129.7 (C-1″), 129.8 (C-4″), 156.1 (C-3′), 178.8 (C-17) ppm; ESI-MS: 440 $[M+H]^+$.

Method B: Compound **6a** (135 mg, 0.30 mmol) was dissolved in ^{t}BuOH (10 mL), and KOH (50 mg, 0.89 mmol) was added. The solution was stirred at room temperature for 3 h and then diluted with water, and the precipitate that formed was filtered off, washed with water and dried. The crude polar product **12a** (111 mg, 87%) was obtained as a white precipitate. Anal. Calcd. for $C_{26}H_{35}NO_4$ (425.56): C, 73.38; H, 8.29. Found: C, 73.27; H, 8.37. ^{1}H NMR (DMSO-d_6, 500 MHz): δ0.76 (m, 3H, 19-H_3), 0.80 (m, 2H), 0.91 (m, 2H), 1.07 (m, 4H and s, 3H, 18-H_3), 1.39 (m, 1H), 1.54–1.74 (overlapping m, 8H), 2.28 (m, 1H), 3.25 (dd, 1H, J = 17.4 Hz, J = 12.3 Hz, one of 16-H_2), 3.46 (dd, 1H, J = 17.4 Hz, J = 12.3 Hz, the other 16-H_2), 3.60 (m, 1H, 3-H), 4.42 (d, 1H, J = 4.5 Hz, 3-OH), 4.52 (t-like m, 1H, 15-H), 7.43 (m, 3H, 3″-H, 4″-H and 5″-H), 7.64 (m, 2H, 2″-H and 6″-H), 12.13 (bs, 1H, COOH) ppm; ESI-MS: 426 $[M+H]^+$.

Method C: Compound **6a** (135 mg, 0.30 mmol) was dissolved in DMSO (10 mL), and ^{t}BuOK (80 mg, 0.71 mmol) was added. The solution was stirred at 80 °C for 1 h and then diluted with water and extracted with EtOAc (2 × 10 mL). The crude product was purified by flash

chromatography with EtOAc/CH_2Cl_2 = 5:95 as eluent to obtain **11a** (106 mg, 87%). See. *2.2.19.*

Results and Discussion

Since conjugation of the alkenyl moiety with a C=O bond has been demonstrated to have a strong promoting effect on the reactivity of the dipolarophile [14], the steroidal unsaturated ketones 17β-acetoxy-5α-androst-1-en-3-one (**1**) [24] and 3β-acetoxy-5α-androst-15-en-17-one (**2**) [25], readily available from 5α-dihydrotestosterone and dehydroepiandosterone, respectively, in a multistep pathway, were applied for the transformations (Table 1). Aromatic hydroximidoyl chlorides (**3a–g**), as relatively stable precursors of nitrile oxides (**4a–g**), were synthetized in two steps by the general protocol from benzaldehyde or its substituted derivatives [26]. The 1,3-dipoles (**4a–g**) can be generated *in situ* from **3a–g** by dehydrochlorination with a base.

Preliminary ring-closure experiments on **1** and **2** with benzonitrile oxide **4a** were first carried out to determine the optimum conditions. The steroidal enone (**1** or **2**) and an excess of *N*-hydroxybenzenecarboximidoyl chloride (**3a**) were dissolved in toluene and 3 equivalents of *N*,*N*-diisopropylethylamine (DIPEA) was slowly added in order to ensure a low stationary concentration of the dipole and to minimize undesired furoxane formation. After the completion of DIPEA addition at room temperature, the solution was refluxed to achieve complete conversion for compound **2** within 2 h, leading to a single product **6a** in a yield of 94% after chromatographic purification (Table 1, entry 8). However, the similar reaction of **1** with **4a** was not complete after a 5-h heating period, as indicated by TLC, and the yield of the purified isoxazoline (**5a**) was only 55% (entry 1), together with a considerable amount (42%) of the residual starting material **1**. The rate of cycloaddition was much slower at room temperature or below, and refluxing of the mixtures was therefore necessary for sufficient

conversions. Similar cycloadditions of **1** and **2** with various substituted benzonitrile oxides (**4b–g**) were subsequently performed under the same conditions to furnish novel ring-condensed isoxazolines (**5b–g**, **6b** and **6e–g**) in moderate to excellent yields (entries 2–7 and 9–12).

Depending on the dipole orientation relative to the double bond, two regioisomers, each involving four diastereoisomers (two *cis* and two *trans*) as concerns the newly-formed stereogenic centres on C-1 and C-2 in **5a–g** or on C-15 and C-16 in **6a–e** can be conceived as possible products of the cycloadditions. However, the *trans*-like connection of the 1,3-dipole in both orientations is precluded because of the ring strain. The attack of the dipole from above the general plane of the sterane framework (the β side) is unlikely in **1** in consequence of the same spatial orientation of the 18-CH_3 [27]. Similarly, the introduction of the nitrile oxide in **2** is unlikely to occur from beneath (the α side) in consequence of the steric interaction between the dipole and 14-H_α [28, 29]. Accordingly, for both reactions only two regioisomers are probable (depicted in Table 1), containing a 1α,2α-*cis* (**5** and **5'**) or a 15β,16β-*cis* ring junction (**6** and **6'**). The formation of regioisomers (**5'** and **6'**) in which the *O* terminus of the dipole is attached to the α-carbon of the dipolarophile, and the aromatic ring is therefore bent towards the sterane portion, is considered to be hampered by steric repulsions. The most facilitated isomers are undoubtedly **5** and **6**, in which the anionic pole of the nitrile oxide is connected to the β carbon of **1** or **2**. Both the regio- and the stereoselectivity of the ring closures are therefore influenced by steric factors, in good agreement with earlier observations that the electronic character of the dipolarophile has only a minor effect on such reactions [30].

The overall yields of the products (**5a–g**, **6a**, **6b** and **6e–g**) were found to depend on the electronic features of the substituents on the aromatic ring of the dipoles **4a–g** (Table 1), and

this effect was more pronounced in the case of the ring A-fused derivatives **5a–g**. The electron-donating substituents in **4b** and **4e** favoured cycloaddition to **1** and **2** (Table 1, entries 2, 5, 9 and 10), in consequence of the lower tendency of these dipoles to undergo dimerization to furoxanes, while the yields of the desired products (**5f** and **6f**, or **5g** and **6g**) were decreased by the presence of the electron-withdrawing groups on the aromatic moiety in **4f** and **4g** (entries 6, 7, 11 and 12). The lowest conversions were achieved for the reactions of **1** and **2** with *p*-nitrobenzonitrile oxide **4g**, which resulted in the desired products (**5g** and **6g**) in yields of only 19% and 45%, respectively. In order to investigate both the electronic and the steric effects of the substituents on the reactivity of the dipoles, the cycloadditions of *o*- and *m*-methyl-substituted benzonitrile oxides (**4c** and **4d**) to **1** were also carried out (entries 3 and 4). The highest yield of the tolyl-substituted cycloadducts (**5c–e**) was that for **5c**, which was comparable to that of the methoxy-substituted analogue **5b** (entry 3). This is indicative of the steric hindrance of the *o*-methyl group and hence the increased resistance of this dipole (**4c**) against dimerization to furoxane [14]. The five-membered ring D of **2** proved to be more reactive against nitrile oxides than the six-membered ring A of **1**, and the related products were obtained in higher yields within shorter reaction times. A similar difference in reactivity was earlier observed for the reactions of cyclopentene and cyclohexene, the former being more reactive due to ring strain and conformational effects [16].

The structures of all the synthetized compounds were confirmed by ^{1}H and ^{13}C NMR measurements. The presence of the phenyl or substituted phenyl ring derived from the nitrile oxides (**4a–g**) was demonstrated by the signals in the aromatic range of the ppm scale in **5a–g**, **6a**, **6b** and **6e–g**. In the ^{1}H NMR spectra of **5a–g**, the signals of 2-H and 1-H appeared as two doublets at around 4.27 and 4.85 ppm. The coupling constant $^{2}J_{H,C,H}$ of about 10.0 Hz was consistent with the 1α,2α-(*cis*)-anellation of the hetero ring. At the same time, the doublet of 16-H and the double doublet of 15-H were detected at around 4.12 and 5.42 ppm in

the spectra of **6a**, **6b** and **6e–g**. The exact configurations of the newly formed stereocentres were established with the aid of homonuclear 2D NMR (COSY and NOESY) and heteronuclear 2D NMR (HSQC and HMBC) measurements.

The deacetylations of one cycloadduct from both sets of compounds (**5b** and **6a**) were carried out by using general conditions so as to obtain the corresponding 17-OH analogs of **5** and 3-OH derivatives of **6**. However, the transformations led to unexpected products in particular cases. 17-Deacetylation of **5b** in alkaline MeOH at room temperature resulted in the simultaneous formation of a heteroaromatic isoxazole (**7b**) and a 4′-hydroxylated isoxazoline (**8b**) in an approximate ratio of 4:1, the structures of which were confirmed by NMR spectroscopy after separation (Scheme 1). The formation of the 4′-OH derivative **8b** can be explained by the oxidation of the corresponding enolate produced in alkaline medium. Such hydroxylation has already been observed, especially for ketones containing a tertiary carbon at the α position [31]. The spontaneous aromatization of the isoxazoline ring of **5b** to isoxazole **7b** is quite unusual, however, since the oxidation of such rings is more difficult than that of pyrazolines to pyrazoles [32], and the application of different oxidizing reagents is usually needed [33] even for the dehydrogenation of six-membered ring-condensed analogues [34]. Interestingly, repeated deacetylation of **5b** with ^{t}BuOK in DMSO at 80 ^{o}C led to the formation of **7b** alone, which can be attributed to the fact that the enolate needed for α-hydroxylation is less favoured in more polar solvents. Although **8b** proved to be quite stable in alkaline medium, it could be converted to **7b** by elimination in the presence of *p*-toluenesulfonic acid or sulfuric acid at elevated temperature. However, similar reaction by applying an inert atmosphere led to the desired product **9b** exclusively.

Scheme 1

When the 3-deacetylation of the ring D-fused analogue **6a** was carried out at room temperature, the major formation of a D-*seco* ester (**10a**) was observed, together with the desired product (**11a**) in a ratio of about 6:4 (Scheme 2), while **10a** was the sole product when either **6a** or **11a** was refluxed in alkaline MeOH. However, exclusively the *seco*-carboxylic acid **12a** was obtained when MeOH was replaced by ^{t}BuOH presumably due to the poor nucleophilicity of $^{t}BuO^{-}$ which precludes its attack on C-17. The desired 3β-hydroxy compound **11a** was obtained as sole product by applying ^{t}BuOK in DMSO for repeated deacetylation.

Scheme 2

The observed (*a1a*) fragmentation is similar to the retro-Dieckmann reactions of 1,3-diketones and ketoesters [35], where the attack of the MeO^{-} or HO^{-} nucleophile, derived from either the solvent or the reagent, on the carbonyl-C induces the cleavage of ring D between C-16 and C-17, as depicted in Figure 1. Simultaneous deacetylation on C-3 also occurs to furnish **10a** or **12a**. The formation of the D-*seco* derivatives (**10a** and **12a**) serves as indirect evidence of the regioselectivity of the 1,3-cycloaddition of **2** with **4a–e**, because the fragmentation would not be possible in the cases of **6′a–e** (see Table 1).

Figure 1

The different behavior observed for **5b** and **6a** under deacetylation conditions may be attributed to the higher rigidity and sterically more hindered character of ring D in **6a** compared to the flexible six-membered ring A in **5b**. Aromatization of the hetero ring should

further enhance the ring strain of ring D in **6a**, therefore, fragmentation induced by a nucleophile attack on C-17 instead of oxidation is more favorable in this case.

The 1,3-cycloadditions of nitrile oxides generated *in situ* from **3a–g** were also carried out with the corresponding 17- and 3-OH analogues (**13** and **14**) of **1** and **2** under the same conditions as applied earlier (Table 2). Similar tendencies concerning the reaction rates and the electronic and steric effects of the substituents on the aromatic ring of the nitrile oxides **4a–g** and therefore the yields of the desired products (**9a–g**, **11a**, **11b** and **11e–g**) were found to be repeated here too. The lower reactivity of ring A than that of ring D was again manifested. Since the *p*-nitrophenyl-substituted compound **5g** was isolated in only a moderate yield (19%) from the reaction of **1** with **4g**, the similar cycloaddition of **13** and **4g** was not performed in this case.

Table 2

Conclusions

In summary, novel types of steroidal isoxazolines condensed to either ring A or ring D of the sterane framework were synthetized through the 1,3-dipolar cycloaddition of 5α-androstenones and aromatic nitrile oxides. The ring closures proved to occur in a regio- and stereoselective manner to furnish a single isomer in all cases, in moderate to excellent yields. The higher reactivity of the five-membered ring D led to higher yields of the corresponding products, though all the reactions were affected by the substitution pattern of the nitrile oxides. The 17- and 3-deacetylation of the isoxazolines in alkaline medium opened the way for interesting oxidation and fragmentation pathways, resulting in aromatization of the hetero ring for the ring A-fused derivatives, and skeletal cleavage of the ring D-condensed analogues without affecting the isoxazoline moiety.

Acknowledgements

The financial support by the Hungarian Scientific Research Fund (OTKA K-109107) is gratefully acknowledged. The research by D. Kovács was supported by the European Union and the State of Hungary, co-financed by the European Social Fund in the framework of TÁMOP 4.2.4. A/2-11/1-2012-0001 ″National Excellence Program″.

References

[1] Singh R, Panada G. An overview of synthetic approaches for heterocyclic steroids. Tetrahedron 2013;69:2853–84.

[2] Dua R, Shrivastava S, Sonwane SK, Srivastava SK. Pharmacological significance of synthetic heterocycles scaffold: A review. Adv Biol Res 2011;5:120–44.

[3] Stulov SV, Misharin AYu. Synthesis of steroids with nitrogen-containing substituents in ring D. Chem Heterocycl Compd 2013;48:1431–72.

[4] Mohareb RM, Elmegeed GA, Abdel-Salam OME, Doss SH, William MG. Synthesis of modified steroids as a novel class of non-ulcerogenic, anti-inflammatory and anti-nociceptive agents. Steroids 2011;76:1190–203.

[5] Frank É, Mucsi Z, Zupkó I, Réthy B, Falkay G, Schneider G, Wölfling J. Efficient approach to androstene-fused arylpyrazolines as potent antiproliferative agents. Experimental and theoretical studies of substituent effects on BF_3-catalyzed intramolecular [3+2]cycloadditions of olefinic phenylhydrazones. J Am Chem Soc 2009;131:3894–904.

[6] Kovács D; Wölfling J, Szabó N, Szécsi M, Kovács I, Zupkó I, Frank É. An efficient approach to novel 17-5′-(1′,2′,4′)-oxadiazolyl androstenes via the cyclodehydration of

cytotoxic *O*-steroidacylamidoximes, and an evaluation of their inhibitory action on 17α-hydroxylase/$C_{17,20}$-lyase. Eur J Med Chem 2013;70:649–60.

[7] Frank É, Schneider G. Synthesis of sex hormone-derived modified steroids possessing antiproliferative activity. J Steroid Biochem Mol Biol 2013;137:301–15.

[8] Tangallapally RP, Sun D, Rakesh, Budha N, Lee REB, Lenaerts AJM, Meibohm B, Lee RE. Discovery of novel isoxazolines as anti-tuberculosis agents. Bioorg Med Chem Lett 2007;17:6638–42.

[9] Habeeb AG, Rao PNP, Knaus EE. Design and synthesis of 4,5-diphenyl-4-isoxazolines: novel inhibitors of cyclooxygenase-2 with analgesic and antiinflammatory activity. J Med Chem 2001;44:2921–7.

[10] Safaei-Ghomi J, Ghasemzadeh MA. Synthesis of some 3,5-diaryl-2-isoxazoline derivatives in ionic liquids media. J Serb Chem Soc 2012;77:733–9.

[11] Jäger V, Grund H. Eliminative ring opening of 2-isoxazolines: a new route to α,β-unsaturated ketones. Angew Chem Int Ed 1976;15:50–1.

[12] Curran DP. Reduction of Δ^2-isoxazolines: a conceptually different approach to the formation of aldol adducts. J Am Chem Soc 1982;104:4024–6.

[13] Jäger V, Buss V, Schwab W. Syntheses via isoxazolines III. Diastereoselective synthesis of γ-amino-alcohols with 2 and 3 chiral centres. Tetrahedron Lett 1978;19:3133–6.

[14] Caramella P, Grünanger P. Nitrile oxides and imines. In: Padwa A, editor. 1,3-Dipolar cycloaddition chemistry, vol. 1. New York: John Wiley & Sons; 1984. p. 291–392 [chapter 3].

[15] Belen'kii LI, Zelinksy ND. Nitrile oxides. In: Feuer H, editor. Nitrile oxides, nitrones, and nitronates in organic synthesis: novel strategies in synthesis. Hoboken: John Wiley & Sons; 2008. p. 1–8 [chapter 1].

[16] Jäger V, Colinas PA. Nitrile oxides. In: Padwa A, Pearson WH, editors. Synthetic application of 1,3-dipolar cycloaddition chemistry toward heterocycles and natural products. Hoboken: John Wiley & Sons; 2003. p. 361–472 [chapter 6].

[17] Camoutsis Ch, Nikolaropoulos S. Steroidal isoxazoles, isoxazolines and isoxazolidines. J Heterocycl Chem 1998;35:731–59.

[18] Drach SV, Litvinovskaya RP, Khripach VA. Steroidal 1,2-oxazoles. Synthesis and biological activity (Review). Chem Heterocycl Compds 2000;36:233–55.

[19] Banday AH, Singh S, Alam MS, Reddy DM, Gupta BD, Kumar HMS. Synthesis of novel steroidal D-ring substituted isoxazoline derivatives of 17-oxoandrostanes. Steroids 2008;73:370–4.

[20] Frank É, Kazi B, Mucsi Z, Ludányi K, Keglevich G. New steroid-fused P-heterocycles Part II. Synthesis and conformational study of oxazaphosphorino[16,17-e]estrone derivatives. *Steroids* 2007;72:446–58.

[21] Frank É, Kardos Zs, Wölfling J, Schneider G. Stereoselective synthesis of novel Δ^5-androstenoarylpyrazolyne derivatives by $BF_3.OEt_2$-induced intramolecular 1,3-dipolar cycloaddition. Synlett 2007;1311–3.

[22] Frank É, Wölfling J, Aukszi B, König V, Schneider TR, Schneider G. Stereoselective synthesis of some novel heterocyclic estrone derivatives by intramolecular 1,3-dipolar cycloaddition. Tetrahedron 2002;58:6843–9.

[23] Frank É, Mucsi Z, Szécsi M, Zupkó I, Wölfling J, Schneider G. Intramolecular approach to some new D-ring-fused steroidal isoxazolidines by 1,3-dipolar cycloaddition: synthesis, theoretical and in vitro pharmacological studies. New J Chem 2010;34:2671–81.

[24] Zhang H, Qiu Z. An efficient synthesis of 5α-androst-1-ene-3,17-dione. Steroids 2006;71:1088–90.

[25] Matsui M, Kinuyama Y. Synthesis of isomeric 5α-androstane-3,15,17β-triols. J Chem Soc, Perkin Trans 1 1976;1429–32.

[26] Himo F, Lowell T, Hilgraf R, Rostovtsev VV, Noodleman L, Sharpless KB, Fokin VV. Copper(I)-catalyzed synthesis of azoles. DFT study predicts unprecedented reactivity and intermediates. J Am Chem Soc 2005;127:210–6.

[27] Kádár Z, Baji Á, Zupkó I, Bartók T, Wölfling J, Frank É. Efficient approach to novel 1α-triazolyl-5α-androstane derivatives as potent antiproliferative agents. Org Biomol Chem 2011;9: 8051–7.

[28] Li C, Qiu W, Yang Z, Luo J, Yang F, Liu M, Xie J, Tang J. Stereoselective synthesis of some methyl-substituted steroid hormones and their *in vitro* cytotoxic activity against human gastric cancer cell line MGC-803. Steroids 2010;75:859–69.

[29] Kádár Z, Molnár J, Schneider G, Zupkó I, Frank É. A facile ″click″ approach to novel 15β-triazolyl-5α-androstane derivatives, and an evaluation of their antiproliferative activities *in vitro*. Bioorg Med Chem 2012;20:1396–402.

[30] Grundmann C. Synthesis of heterocyclic compounds with the aid of nitrile oxides. Synthesis 1970;344–59.

[31] Jones AB. Oxidation adjacent to C=X bonds by hydroxylation methods. In: Trost BM, editor. Comprehensive organic synthesis, vol. 7. Oxford: Pergamon Press; 1991. p. 159–60.

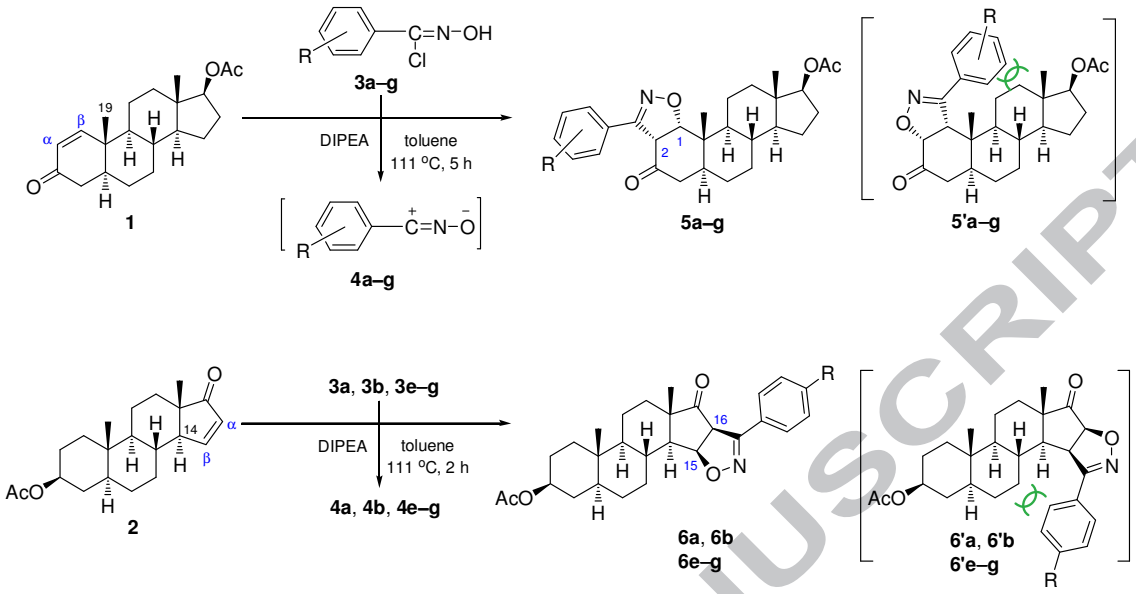
[32] Bianchi G, Grünanger P. Conversion of 2-isoxazolines to isoxazoles. Tetrahedron 1965;21:817–22.

[33] Azarifar D, Khosravi K, Veisi R-A. An efficient oxidation of 2-pyrazolines and isoxazolines by bis-bromine-1,4-diazabicyclo[2.2.2]octane complex (DABCO-Br_2). ARKIVOC 2010;(ix):178–84.

[34] Hashimoto Y, Takada A, Takikawa H, Suzuki K. Synthesis of isoxazoles *en route* to semi-aromatized polyketides: dehydrogenation of benzonitrile oxide–*para*-quinone acetal cycloadducts. Org Biomol Chem 2012;10:6003–9.

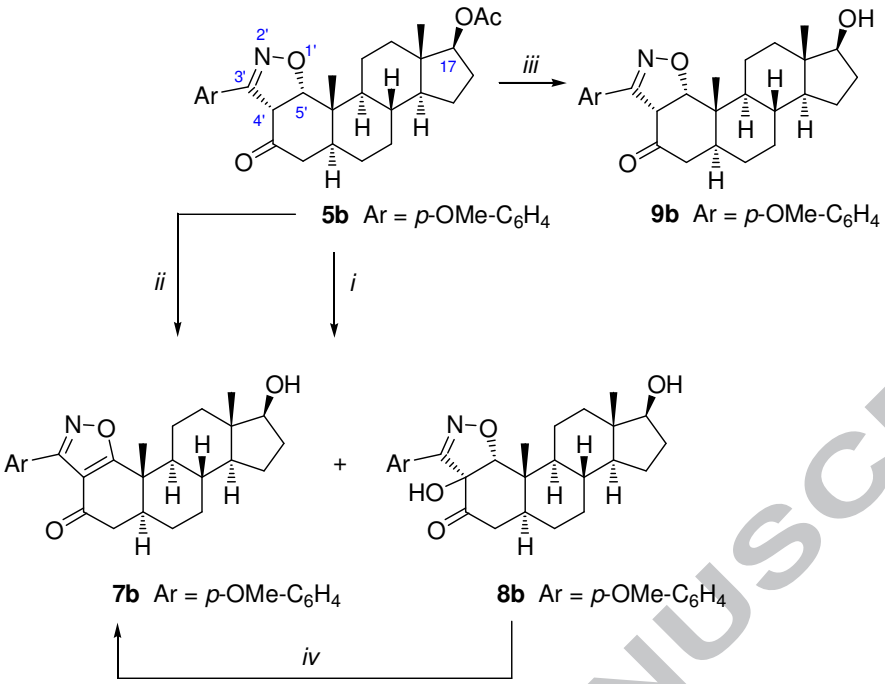
[35] Sano S, Shimizu H, Nagao Y. Facile generation method for conjugated allenyl esters based on retro-Dieckmann-type ring-opening reactions. Tetrahedron Lett 2005;46:2883–6.

Table 1. Regio- and stereoselective synthesis of ring A- and ring D-fused steroidal isoxazolines

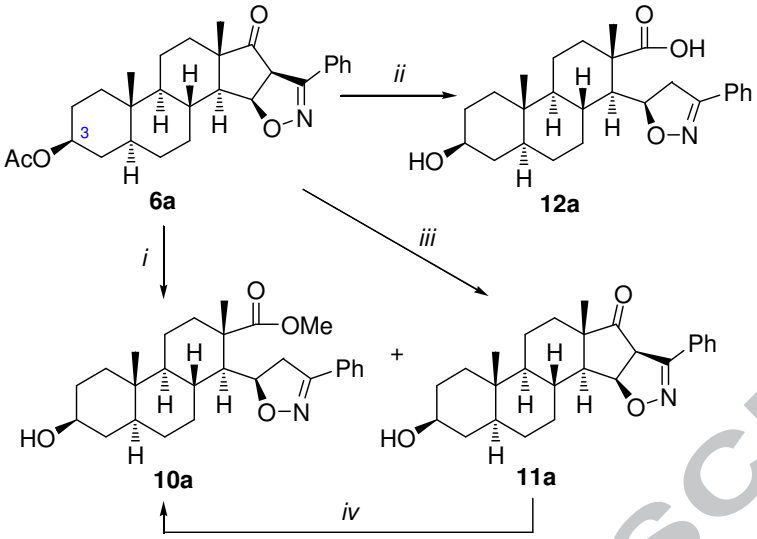


Entry	Substrate	Hydroximidoyl chloride/Nitrile oxide	R	Product	Yield[a] (%)
1	**1**	**3a/4a**	H	**5a**	55
2	**1**	**3b/4b**	*p*-OMe	**5b**	75
3	**1**	**3c/4c**	*o*-CH_3	**5c**	73
4	**1**	**3d/4d**	*m*-CH_3	**5d**	56
5	**1**	**3e/4e**	*p*-CH_3	**5e**	61
6	**1**	**3f/4f**	*p*-Cl	**5f**	40
7	**1**	**3g/4g**	*p*-NO_2	**5g**	19
8	**2**	**3a/4a**	H	**6a**	94
9	**2**	**3b/4b**	*p*-OMe	**6b**	98
10	**2**	**3e/4e**	*p*-CH_3	**6e**	97
11	**2**	**3f/4f**	*p*-Cl	**6f**	78
12	**2**	**3g/4g**	*p*-NO_2	**6g**	45

[a] Determined after purification by column chromatography.



Scheme 1



Scheme 2

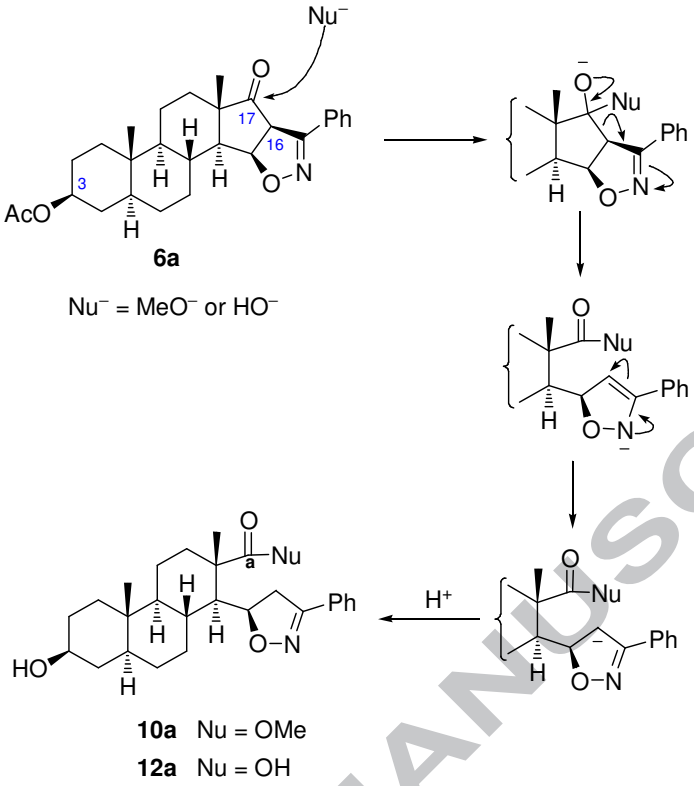
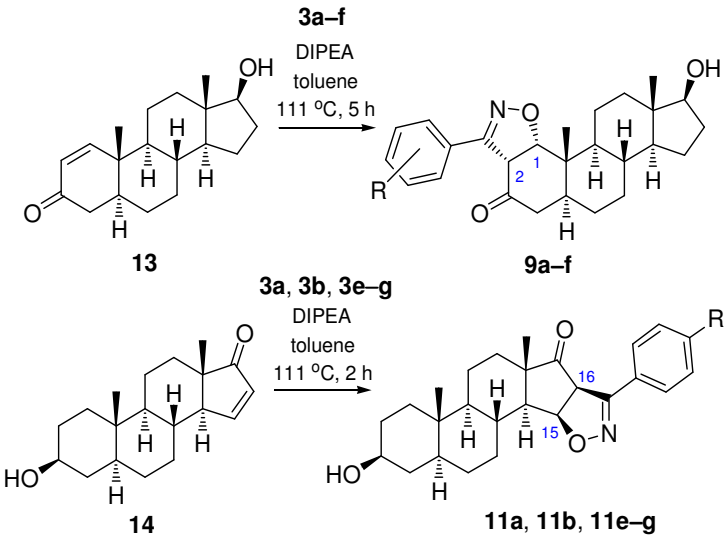


Figure 1

Table 2. Synthesis of 17β- and 3β-OH analogues of ring-fused isoxazolines



Entry	Substrate	R	Product	Yield[a] (%)
1	**13**	H	**9a**	44
2	**13**	*p*-OMe	**9b**	66
3	**13**	*o*-CH_3	**9c**	62
4	**13**	*m*-CH_3	**9d**	51
5	**13**	*p*-CH_3	**9e**	53
6	**13**	*p*-Cl	**9f**	37
8	**14**	H	**11a**	92
9	**14**	*p*-OMe	**11b**	97
10	**14**	*p*-CH_3	**11e**	94
11	**14**	*p*-Cl	**11f**	77
12	**14**	*p*-NO_2	**11g**	44

[a] Determined after purification by column chromatography.

Legends for Figures and Schemes

Scheme 1. Reagents and conditions: (*i*) KOH, MeOH, rt, 3 h; (*ii*) ^{t}BuOK, DMSO, 80 °C, 1 h; (*iii*) ^{t}BuOK, DMSO, 80 °C, 1 h, N_2 atm; (*iv*) PTSA or H_2SO_4, MeOH, 65 °C.

Scheme 2. Reagents and conditions: (*i*) KOH, MeOH, rt, 8 h; (*ii*) KOH, ^{t}BuOH, rt, 3 h; (*iii*) ^{t}BuOK, DMSO, 80 °C, 1 h; (*iv*) KOH, MeOH, 65 °C, 2 h.

Figure 1. Proposed mechanism of the (*a1a*)-type fragmentation

Highlights

> Isoxazoline hetero rings were introduced into the sterane nucleus.

> Intermolecular alkene–nitrile oxide cycloadditions were carried out.

> The reactivities of the enone moiety of rings A and D were compared.

> Behavior of the synthetized compounds under deacetylation was studied.

> The structures of all novel compounds were confirmed by NMR measurements.