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Diels–Alder reaction of androsta-14,16-dien-17-yl acetates with nitroethylene: Product distribution and selected adduct transformations

Alexander V. Baranovsky [a,*], Dmitry A. Bolibrukh [a], Vladimir A. Khripach [a], Bernd Schneider [b]

[a] Institute of Bioorganic Chemistry, National Academy of Sciences of Belarus, Kuprevich str. 5/2, 220143 Minsk, Belarus
[b] Max Planck Institute for Chemical Ecology, Beutenberg Campus, Hans Knöll Str. 8, D-07745 Jena, Germany

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ABSTRACT

The Diels–Alder cycloaddition of nitroethylene to some androsta-14,16-dien-17-yl acetates has been studied. The addition occurs stereoselectively, giving predominantly head-to-head-adduct. 14β-Cyanomethyl steroids were obtained via the reductive cleavage reaction of bridged nitro compounds. The structures of the new compounds have been fully characterized by 2D NMR and tandem mass-spectrometry methods.

1. Introduction

We have recently described how the Diels–Alder adduct of 3-methoxyestra-1,3,5(10),14,16-pentaen-17-yl acetate with nitroethylene could undergo various unusual chemical transformations under relatively mild conditions [1,2] probably with the involvement of the nitrile oxide intermediate. Here, we tried expanding the procedures on androstane-type steroids in order to analyze the product distribution of the Diels–Alder reaction of steroidal dienyl acetates with nitroethylene and to verify that the method can be applied to more functionalized steroids for exploring the approach to C-14-modified natural steroids such as brassinosteroids.

2. Experimental

2.1. General

Melting points were measured using a Boetius apparatus and are uncorrected. IR spectra were recorded using a Michelson Bomem 100 FTIR spectrometer. ^{1}H NMR (500.13 MHz) and ^{13}C NMR (125.77 MHz) spectra were recorded on a Bruker AVANCE-500 NMR spectrometer. $CDCl_3$ was used as a solvent and the residual solvent signals (δ 7.26 ppm for ^{1}H NMR and 77.16 ppm for ^{13}C NMR) served as an internal reference standards. COSY, HSQC, HMBC, TOCSY and NOESY experiments were carried out with the use of the standard Bruker program package. Mass spectra (APCI MS and MS^2, positive mode, CID 35%) were recorded on an Accela HPLC system coupled with LCQ Fleet mass-detector, mass-to-charge ratio (m/z) and relative intensities (%) are indicated for the significant peaks. Microanalyses were determined using a Eurovector EA3000 CHNS-O instrument. Accurate masses (EI) were obtained with a VG-70E mass spectrometer.

TLC was performed on precoated aluminum backed TLC sheets (silica gel 60 F_{254}) and visualized by UV and/or exposure to $Ce(NH_4)_4(SO_4)_4$ in 8 M H_2SO_4. Column chromatography was conducted with Merck silica gel 60: 70–230 mesh. Solvents were dried and freshly distilled according to common practice [3]. All reactions were conducted under positive nitrogen pressure.

2.1.1. Cycloaddition of dienyl acetate *1* with nitroethylene

A solution of dienyl acetate **1** (0.809 g, 2.19 mmol) and nitroethylene (0.318 g, 4.35 mmol) in dry benzene (7 ml) was refluxed for 3 h and after which more nitroethylene (0.511 g, 7 mmol) was added. Refluxing was continued for another 2 h and the solution was cooled, diluted with dichloromethane and filtered through a Celite plug. The filtrate was evaporated and the residue was crystallized from benzene to give 3β-acetoxy-14,17-etheno-16α-nitro-androst-5-en-17β-yl acetate (**5**) (0.734 g, 76%): m.p. 219–221 °C (hexane). IR (KBr, cm^{-1}) 2940, 1735 (OAc), 1730 (OAc), 1550 and 1370 (NO_2), 1240, 1040. ^{1}H NMR δ: 0.98 (3H, s, 19-Me), 1.00 (3H, s, 18-Me), 1.08 (1H, m, 12β-H), 1.14 (1H, m, 1α-H), 1.19 (1H, m, 9α-H), 1.28 (1H, m, 11β-H), 1.54 (1H, m, 11α-H), 1.57 (1H, m, 8β-H), 1.58 (1H, m, 2β-H), 1.87 (1H, m, 1β-H), 1.88 (1H, m, 2α-H),

* Corresponding author. Tel.: +375 17 267 9103; fax: +375 17 267 8761.
E-mail address: baranovsky@iboch.bas-net.by (A.V. Baranovsky).

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1.97 (2H, m, 7-H), 1.99 (1H, m, 15α-H), 2.03 (3H, s, 3-AcO), 2.13 (1H, m, 15β-H), 2.14 (3H, s, 17-AcO), 2.19 (1H, m, 12α-H), 2.35 (2H, m, 4-H), 4.61 (1H, m, 3α-H), 5.38 (1H, dd, J = 3 and 9 Hz, 16β-H), 5.42 (1H, m, 6-H), 6.19 (1H, J = 6 Hz, 17^1-H), 6.29 (1H, d, J = 6 Hz, 17^2-H); ^{13}C NMR δ: 14.92 (C-18), 19.37 (C-19), 20.90 (C-11), 21.53 (3-OC(O)*C*H$_3$), 21.55 (17-OC(O)*C*H$_3$), 26.41 (C-7), 27.77 (C-2), 28.65 (C-12), 31.46 (C-8), 34.96 (C-15), 36.48 (C-10), 36.84 (C-1), 38.22 (C-4), 46.08 (C-9), 55.44 (C-14), 62.12 (C-13), 73.68 (C-3), 87.34 (C-16), 96.24 (C-17), 121.53 (C-6), 129.72 (C-17^1), 135.13 (C-17^2), 139.70 (C-5), 169.74 (17-O*C*(O)CH$_3$), 170.59 (3-O*C*(O)CH$_3$). LRMS m/z (I,%): 444 ([MH]$^+$, 39), 402 ([MH−CH$_2$CO]$^+$, 21), 384 ([MH−AcOH]$^+$, 26), 342 ([MH−CH$_2$CO−AcOH]$^+$, 15), 324 ([MH−2AcOH]$^+$, 100), 307 (25), 283 (19). MS^2 (444): 402 ([MH−CH$_2$CO]$^+$, 11), 358 (13), 330 (100), 324 ([MH−2AcOH]$^+$, 20). MS^2 (324): 307 (50), 306 ([MH−2AcOH−H$_2$O]$^+$, 100), 296 ([MH−2AcOH−C$_2$H$_4$]$^+$ 47), 279 (24), 278 ([MH−2AcOH−C$_2$H$_4$−H$_2$O]$^+$,19), 265 (52), 247 (33). Anal. calcd. for $C_{25}H_{33}NO_6$: C, 67.70; H, 7.50; N, 3.16. Found. C, 67.38; H, 7.43; N, 3.40. Mother liquor residues were chromatographed on a silica gel column (petroleum ether: EtOAc = 90: 10) to give three fractions: A – 0.018 g of compound **5**; B – 0.163 g of mixture of adducts **5** and **6** (1:1 based on integration curve in ^{1}H NMR spectrum); C – 0.048 g of mixture of adducts **6** and **7** (1:1 based on integration curve in ^{1}H NMR spectrum). Fraction C was repeatedly separated to afford pure 3β-acetoxy-14,17-etheno-15α-nitroandrost-5-en-17β-yl acetate (**6**): m.p.194–196 °C (hexane). IR (KBr, cm^{-1}) 2920, 1740 (OAc) 1730 (OAc), 1550 and 1365 (NO$_2$), 1245, 1030. ^{1}H NMR δ: 0.99 (3H, s, 19-Me), 1.02 (3H, s, 18-Me), 1.13 (1H, m, 1α-H), 1.17 (1H, m, 9α-H), 1.18 (1H, m, 12β-H), 1.27 (1H, m, 11β-H), 1.52 (1H, m, 11α-H), 1.57 (1H, m, 2β-H), 1.85 (1H, m, 1β-H), 1.87 (1H, m, 2α-H), 1.87 (1H, m, 7α-H), 1.95 (1H, m, 8β-H), 2.01 (1H, m, 7β-H), 2.02 (3H, s, 3-AcO), 2.09 (1H, m, 12α-H), 2.09 (3H, s, 17-AcO), 2.33 (2H, m, 4-H), 2.49 (1H, dd, J = 9 and 13 Hz, 16β-H), 2.61 (1H, dd, J = 4 and 13 Hz, 16α-H), 4.58 (1H, m, 3α-H), 5.04 (1H, dd, J = 4 and 9 Hz, 15β-H), 5.31 (1H, m, 6-H), 6.05 (1H, J = 6 Hz, 17^2-H), 6.58 (1H, d, J = 6 Hz, 17^1-H); ^{13}C NMR δ: 15.18 (C-18), 19.26 (C-19), 20.57 (C-11), 21.33 (17-OC(O)*C*H$_3$), 21.51 (3-OC(O)*C*H$_3$), 25.71 (C-7), 27.79 (C-2), 28.21 (C-12), 32.7 (C-8), 36.44 (C-10), 37.02 (C-1), 37.99 (C-4), 39.93 (C-16), 46.23 (C-9), 60.9 (C-14), 62.15 (C-13), 73.61 (C-3), 89.52 (C-15), 92.54 (C-17), 121.52 (C-6), 129.7 (C-17^2), 135.49 (C-17^1), 138.95 (C-5), 170.6 (O*C*(O)CH$_3$). LRMS m/z (I,%): 444 ([MH]$^+$, 43), 384 ([MH−AcOH]$^+$, 38), 324 ([MH−2AcOH]$^+$, 100. Anal. calcd. for $C_{25}H_{33}NO_6$: C, 67.70; H, 7.50; N, 3.16. Found. C, 67.14; H, 7.35; N, 3.26. and 3β-acetoxy-14,17-etheno-15β-nitroandrost-5-en-17α-yl acetate (**7**): m.p.205–207 °C. (hexane, dec.). IR (KBr, cm^{-1}) 2945, 1735 (OAc) 1730 (OAc), 1550 and 1365 (NO$_2$), 1245, 1040. ^{1}H NMR δ: 1.01 (3H, s 18-Me), 1.06 (3H, m, 19-Me), 1.25 (1H, m, 1α-H), 1.47 (2H, m, 12-H), 1.54 (1H, m, 11β-H), 1.58 (1H, m, 2β-H), 1.61 (1H, m, 9α-H), 1.69 (1H, m, 11α-H), 1.78 (1H, m, 7α-H), 1.89 (1H, m, 1β-H), 1.90 (1H, m, 2α-H), 1.98 (1H, m, 8β-H), 2.04 (3H, s, 3-AcO), 2.05 (1H, m, 7β-H), 2.10 (3H, s, 17-AcO), 2.35 (2H, m, 4-H), 2.58 (2H, m, 16-H), 4.62 (1H, m, 3α-H), 5.37 (1H, m, 6-H), 5.49 (1H, dd, J = 5 and 8 Hz, 15α-H), 6.10 (1H, d, J = 6 Hz, 17^2-H), 6.50 (1H, d, J = 6 Hz, 17^1-H). ^{13}C NMR δ: 15.51 (C-18), 19.07 (C-11), 19.53 (C-19), 21.38 (17-OC(O)*C*H$_3$), 21.52 (3-OC(O)*C*H$_3$), 26.94 (C-12), 27.21 (C-7), 27.77 (C-2), 30.49 (C-8), 37.14 (C-1), 37.48 (C-16), 37.58 (C-10), 38.11 (C-4), 43.78 (C-9), 58.88 (C-14), 61.87 (C-13), 73.62 (C-3), 86.33 (C-15), 92.49 (C-17), 122.07 (C-6), 132.05 (C-17^2), 135.97 (C-17^1), 139.18 (C-5), 170.56 (3-O*C*(O)CH$_3$), 170.66 (17-O*C*(O)CH$_3$). LRMS m/z (I,%): 444 ([MH]$^+$, 100), 384 ([MH−AcOH]$^+$, 35), 324 ([MH−2AcOH]$^+$, 68), 306 ([MH−2AcOH−H$_2$O]$^+$, 13). MS^2 (444): 402 ([MH−CH$_2$CO]$^+$, 92), 384 ([MH−AcOH]$^+$, 100), 324 ([MH−2AcOH]$^+$, 86). Anal. calcd. for $C_{25}H_{33}NO_6$: C, 67.70; H, 7.50; N, 3.16. Found. C, 66.84; H, 7.40; N, 3.40. Therefore, the overall yield of the adducts was 87% of compound **5**, 11% of compound **6** and 2% of compound **7**.

2.1.2. 3β-Benzoyloxy-14,17-etheno-16α-nitroandrost-5-en-17β-yl acetate (**8**)

Following the same procedure as for compound **5**, compound **2** (0.868 g, 2 mmol) was converted into steroid **8** and its two isomers. Crystallization of the reaction mixture gave 0.531 g (53%) of nitro compound **8**: m.p. 223–225 °C (benzene). IR (KBr, cm^{-1}): 2940, 1740 (OAc), 1710 (OBz), 1560 and 1370 (NO$_2$), 1275, 1240, 1110. ^{1}H NMR δ: 1.02 (3H, s, 18-Me), 1.03 (3H, s, 19-Me), 1.10 (1H, m, 12β-H), 1.23 (1H, m, 1α-H), 1.25 (1H, m, 9α-H), 1.29 (1H, m, 11β-H), 1.58 (1H, m, 11α-H), 1.59 (1H, m, 8β-H), 1.73 (1H, m, 2β-H), 1.94 (1H, dt, J = 3 and 13 Hz, 1β-H), 2.01 (2H, m, 7-H), 2.01 (1H, m, 15α-H), 2.03 (1H, m, 2α-H), 2.15 (1H, m, 15β-H), 2.15 (3H, s, 17-AcO), 2.20 (1H, m, 12α-H), 2.50 (2H, m, 4-H), 4.87 (1H, m, 3α-H), 5.39 (1H, dd, J = 3 and 9 Hz, 16β-H), 5.48 (1H, m, 6-H), 6.20 (1H, J = 6 Hz, 17^1-H), 6.33 (1H, d, J = 6 Hz, 17^2-H), 7.44 (2H, m, m-Ph), 7.55 (1H, m, p-Ph), 8.04 (2H, d, J = 7 Hz, o-Ph); ^{13}C NMR δ: 14.95 (C-18), 19.46 (C-19), 20.94 (C-11), 21.58 (17-OC(O)*C*H$_3$), 26.47 (C-7), 27.90 (C-2), 28.68 (C-12), 31.49 (C-8), 34.99 (C-15), 36.57 (C-10), 36.90 (C-1), 38.33 (C-4), 46.11 (C-9), 55.47 (C-14), 62.15 (C-13), 74.29 (C-3), 87.36 (C-16), 96.27 (C-17), 121.68 (C-6), 128.44 (Ph C-3), 129.69 (Ph C-2), 129.75 (C-17^1), 130.90 (Ph C-1), 132.93 (Ph C-4), 135.18 (C-17^2), 139.71 (C-5), 166.11 (3-O*C*(O)Ph), 169.78(17-OC(O)CH$_3$). LRMS m/z (I,%): 506 ([MH]$^+$, 45), 464 ([MH−CH$_2$CO]$^+$, 14), 384 ([MH−BzOH]$^+$, 24), 342 ([MH−BzOH−CH$_2$CO]$^+$, 25), 324 ([MH−BzOH−AcOH]$^+$, 100). MS^2 (506): 392 (100), 324 ([MH−BzOH−AcOH]$^+$, 10). MS^2 (324): 307 (56), 306 ([MH−BzOH−AcOH−H$_2$O]$^+$, 100), 296 ([MH−BzOH−AcOH−C$_2$H$_4$]$^+$ 40), 279 (29), 278 ([MH−BzOH−AcOH−C$_2$H$_4$−H2O]$^+$, 17), 265 (52), 247 (15). HRMS calcd for $C_{23}H_{29}NO_4$ (M^+−BzOH): 383.2097. Found. 383.2094. Mother liquor residues were chromatographed on a silica gel column (petroleum ether: EtOAc = 90: 10) to give two fractions: A – 0.147 g of compound **8**, thus elevating the overall yield of **8** to 67%; B – 0.191 g of two minor adducts.

2.1.3. 14,17-Etheno-6,6-ethylenedioxy-2α,3α-isopropylidendioxy-16α-nitro-5α-androstan-17β-yl acetate (**9**)

A solution of steroid **3** (0.066 g, 0.15 mmol) and nitroethylene (0.54 g, 7.4 mmol) in dry benzene (7 ml) was refluxed for 10 h and some more nitroethylene (0.27 g, 3.7 mmol) was added. Refluxing was continued for a further 6 h and the solution was cooled, evaporated and residue was chromatographed on a silica gel column (petroleum ether: EtOAc = 90:10) to give steroid **9** (0.038 g, 49%): m.p.132–134 °C (hexane). IR (KBr, cm^{-1}): 2940, 1745 (OAc), 1550 and 1370 (NO$_2$). ^{1}H NMR δ: 0.80 (3H, s, 19 Me), 0.98 (3H, s, 18 Me), 1.03 (1H, m, 9α-H), 1.08 (1H, m, 1α-H), 1.08 (1H, m, 12β-H), 1.13 (1H, m, 11β-H), 1.34 (3H, s, β-CH$_3$ acetonide), 1.44 (1H, m, 7α-H), 1.48 (3H, s, α-CH$_3$ acetonide), 1.58 (1H, m, 11α-H), 1.59 (1H, m, 8β-H), 1.70 (1H, m, 7β-H), 1.81 (1H, m, 4β-H), 1.87 (1H, m, 5α-H), 1.93 (1H, m, 1β-H), 1.97 (1H, m, 15α-H), 2.12 (1H, m, 12α-H), 2.13 (1H, m, 15β-H), 2.15 (3H, s, OAc), 2.23 (1H, d, J = 15 Hz, 4α-H), 3.80 and 3.94 (2H, m, α-CH$_2$ dioxolane), 3.92 and 3.99 (2H, m, β-CH$_2$ dioxolane), 4.08 (1H, m, 2β-H), 4.27 (1H, m, 3β-H), 5.37 (1H, dd, J = 2 and 9 Hz, 16β-H), 6.18 (1H, d, J = 6.4, 17^1-H), 6.28 (1H, d, J = 6.4, 17^2-H). ^{13}C NMR δ: 13.43 (C-19), 15.04 (C-18), 20.41 (C-11), 21.57 (OC(O)*C*H$_3$), 22.04 (C-4), 26.70 (β-CH$_3$ acetonide), 28.64 (C-12), 28.77 (α-CH$_3$ acetonide), 32.78 (C-8), 34.82 (C-15), 36.61 (C-7), 38.21 (C-10), 42.69 (C-1), 45.28 (C-5), 48.68 (C-9), 55.18 (C-14), 62.13 (C-13), 64.42 (α-CH$_2$ dioxolane), 65.79 (β-CH$_2$ dioxolane), 72.78 (C-3), 72.84 (C-2), 87.30 (C-16), 96.02 (C-17), 107.83 (Me$_2$*C* acetonide), 109.36 (C-6), 129.74 (C-17^1), 134.61 (C-17^2), 169.78 (O*C*(O)CH$_3$). LRMS m/z (I,%): 518 ([MH]$^+$, 23), 460 ([MH−Me$_2$CO]$^+$, 40), 400 ([MH−Me$_2$CO−AcOH]$^+$, 40), 382 ([MH−Me$_2$CO−AcOH−H$_2$O]$^+$, 51), 338 ([MH−Me$_2$CO−AcOH−H$_2$O−CH$_2$CH$_2$O]$^+$, 20). HRMS calcd for $C_{28}H_{39}NO_8$: 517.2676. Found. 517.2679.

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2.1.4. 2α,3α-Diacetoxy-14,17-etheno-16α-nitro-5α-androstan-6-on-17β-yl acetate (10)

Following the same procedure as for compound **9**, compound **4** (0.016 g, 0.04 mmol) was converted into steroid **10** (0.013 g 70%): m.p. 245–247 °C (hexane). IR (KBr, cm^{-1}): 2945, 1741 (CO), 1715 (CO), 1550 and 1370 (NO_2). ^1H NMR δ: 0.80 (3H, s, 19 Me), 0.98 (3H, s, 18 Me), 1.15 (1H, m, 12β-H), 1.16 (1H, m, 11β-H), 1.65 (1H, m, 1α-H), 1.65 (1H, m, 9α-H), 1.66 (1H, m, 11α-H), 1.86 (1H, m, 8β-H), 1.88 (2H, m, 4-H), 1.79 (1H, dd, J = 5 and 12 Hz, 1β-H), 1.94 (1H, dd, J = 3 and 13 Hz, 15α-H), 1.99 (3H, s, 2-OAc), 2.11 (3H, s, 3-OAc), 2.12 (1H, m, 15β-H), 2.16 (3H, s, 17-OAc), 2.21 (1H, td, J = 5 and 14 Hz, 12α-H), 2.32 (2H, m, 7-H), 2.64 (1H, dd, J = 5 and 11 Hz, 5α-H), 4.92 (1H, ddd, J = 3, 5 and 12 Hz, 2β-H), 5.33 (1H, dd, J = 3 and 9 Hz, 16β-H), 5.39 (1H, m, 3β-H), 6.29 (1H, d, J = 6 Hz, 17^1-H), 6.38 (1H, d, J = 6 Hz, 17^2-H). ^{13}C NMR δ: 13.40 (C-19), 15.03 (C-18), 20.90 (C-11), 21.18 (2-OC(O)CH_3), 21.32 (3-OC(O)CH_3), 21.54 (17-OC(O)CH_3), 24.94 (C-4), 28.58 (C-12), 34.42 (C-15), 37.28 (C-8), 37.49 (C-1), 42.18 (C-7), 42.47 (C-10), 49.51 (C-9), 51.62 (C-5), 55.38 (C-14), 62.38 (C-13), 67.96 (C-3), 68.96 (C-2), 87.14 (C-16), 95.68 (C-17), 130.56 ($C-17^1$), 133.18 ($C-17^2$), 169.79 (17-OC(O)CH_3), 170.15 (3-OC(O)CH_3), 170.46 (2-OC(O)CH_3), 209.34 (C-6). LRMS *m/z* (*I*,%): 518 ([*M*H]$^+$, 43), 458 ([*M*H−AcOH]$^+$, 63), 416 ([*M*H−AcOH−CH_2CO]$^+$, 71), 398 ([*M*H−2AcOH]$^+$, 59), 356 ([*M*H−2AcOH−CH_2CO]$^+$, 100). MS^2 (518): 476 ([*M*H−CH_2CO]$^+$, 13), 432 (10), 404 (100), 356 ([*M*H−2AcOH−CH_2CO]$^+$, 17). HRMS calcd for $C_{27}H_{35}NO_9$: 517.2312. Found. 517.2301.

*2.1.5. Solvolysis of nitro compound **5***

A mixture of nitro compound **5** (0.263 g, 0.59 mmol) and $NaHCO_3$ (0.346 g, 4.13 mmol) in aqueous ethanol (66 ml, 10:1) was refluxed for 1 h. The yellow solution was cooled to 0 °C and poured into a cooled 1 M HCl solution. The resulting mixture was stirred for 10 min and extracted with chloroform. The combined organic phase was washed with brine, dried (Na_2SO_4), and concentrated. The oily residue (0.266 g) was dissolved in pyridine (3 ml) and benzoyl chloride (0.21 g, 1.5 mmol) was added. The resulting mixture was stirred for 24 h, then poured out in cold water and extracted with chloroform. The combined organic phase was washed with 1 M HCl solution, saturated $NaHCO_3$, brine, dried (Na_2SO_4), and concentrated. The residue was chromatographed on a silica gel column with ethyl acetate as eluent to afford 3β-acetoxy-1′-benzoyloxy-2′-oxopyrrolidino-[4′,5′:14β,15β]-androst-5-en-17-one (**12b**) (0.062 g, 20%). m.p. 157–158 °C (EtOAc-hexane). IR ν_{max} (KBr), cm^{-1}: 2945, 1770 (OBz), 1740 (CO), 1725 (OAc and NC = O), 1245. ^1H NMR δ: 1.05 (3H, s, 19 Me), 1.13 (3H, s, 18 Me), 1.20 (1H, m, 1α-H), 1.38 (1H, m, 12α-H), 1.42 (1H, m, 9α-H), 1.42 (1H, m, 11β-H), 1.49 (1H, m, 12β-H), 1.63 (1H, m, 2β-H), 1.71 (1H, m, 11α-H), 1.72 (1H, m, 8β-H), 1.91 (1H, m, 2α-H), 1.92 (1H, m, 1β-H), 2.04 (3H, s, 3-OAc), 2.06 (1H, m, 7β-H), 2.21 (1H, d, J = 17 Hz, 14^1α-H), 2.30 (1H, d, J = 17 Hz, 14^1β-H), 2.34 (1H, m, 4β-H), 2.40 (1H, ddd, J = 1.5, 5 and 13 Hz, 4α-H), 2.52 (1H, dd, J = 3 and 19 Hz, 16β-H), 2.61 (1H, m, 7α-H), 3.02 (1H, dd, J = 9 and 19 Hz, 16α-H), 4.49 (1H, dd, J = 4 and 9 Hz, 15α-H), 4.64 (1H, m, 3α-H), 5.54 (1H, m, 6-H), 7.49 (2H, m, *m*-Ph), 7.65 (1H, m, *p*-Ph), 8.09 (2H, d, J = 8 Hz, *o*-Ph); ^{13}C NMR δ: 14.19- (C-18), 19.60 (C-11), 19.74 (C-19), 21.54 (3-OC(O)CH_3), 26.23 (C-7), 27.66 (C-2), 32.84 (C-12), 36.85 (C-8) 36.95 (C-1), 37.32 (C-10), 37.86 (C-4), 38.37 ($C-14^1$), 38.72 (C-16), 45.21 (C-9), 48.44 and 54.07 (C-13 and C-14), 57.16 (C-15), 73.61 (C-3), 122.40 (C-6), 126.49 (Ph C-1), 128.94 (Ph C-3), 130.38 (Ph C-2), 134.59 (Ph C-4), 138.01 (C-5), 163.81 (PhCO), 168.94 ($C-14^2$), 170.63 (3-OC(O)CH_3), 215.82 (C-17). LRMS *m/z* (*I*,%): 506 ([*M*H]$^+$, 100), 446 ([*M*H−AcOH]$^+$, 15). MS^2 (506): 446 ([*M*H−AcOH]$^+$, 100). MS^3 (446): 324 ([*M*H−AcOH−BzOH]$^+$, 73), 296 ([*M*H−AcOH−BzOH−CO]$^+$, 10), 282 ([*M*H−AcOH−BzOH−CH_2CO]$^+$, 100). Further elution with EtOAc: EtOH (97:3) gave 3β-acetoxy-2′-oxopyrrolidino-[4′,5′:14β,15β-androst-5-en-17-one (**11**) (0.060 g, 26%): m.p. 256–258 °C (EtOAc). IR ν_{max} (KBr), cm^{-1}: 3241 (NH), 2930, 1735 (CO and OAc), 1700 (NC = O), 1670, 1245. ^1H NMR δ: 1.03 (3H, s, 19 Me), 1.11 (3H, s, 18 Me), 1.17 (1H, m, 1α-H), 1.34 (1H, m, 9α-H), 1.37 (1H, m, 12α-H), 1.41 (1H, m, 11β-H), 1.43 (1H, m, 12β-H), 1.61 (1H, m, 2β-H), 1.68 (1H, m, 11α-H), 1.69 (1H, m, 8β-H), 1.90 (1H, m, 2α-H), 1.92 (1H, dt, J = 3 and 13 Hz, 1β-H), 2.03 (2H, m, 7-H), 2.04 (3H, s, 3-OAc) 2.01 (1H, d, J = 17 Hz, 14^1α-H), 2.13 (1H, dd, J = 3 and 19 Hz, 16β-H), 2.21 (1H, d, J = 17 Hz, 14^1β-H), 2.34 (2H, m, 4-H), 3.15 (1H, dd, J = 9 and 19 Hz, 16α-H), 4.10 (1H, ddd, J = 1, 4, 7 Hz, 15α-H), 4.62 (1H, m, 3α-H), 5.42 (1H, m, 6-H), 5.85 (1H, br s, NH); ^{13}C NMR δ: 15.17 (C-18), 19.65 (C-11), 19.67 (C-19), 21.54 (3-OC(O)CH_3), 26.49 (C-7), 27.64 (C-2), 32.64 (C-12), 36.98 (C-8), 37.01 (C-1), 37.30 (C-10), 37.83 (C-4), 41.15 ($C-14^1$), 43.20 (C-16), 45.25 (C-9), 51.53 (C-15), 52.86 and 53.95 (C-14 and C-13), 73.60 (C-3), 121.94 (C-6), 138.56 (C-5), 170.70 (3-OC(O)CH_3), 176.32 ($C-14^2$), 217.12 (C-17). LRMS *m/z* (*I*,%): 386 ([*M*H]$^+$, 100). MS^2 (386): 326 ([*M*H−AcOH]$^+$, 100). MS^3 (326): 280 ([*M*H−AcOH−H_2O−CO]$^+$, 100), 249 ([*M*H−AcOH−H_2O−CH_3-$CONH_2$]$^+$, 16). Anal. calcd. for $C_{23}H_{31}NO_4$: C, 71.66; H, 8.11; N, 3.63. Found. C, 70.61; H, 7.86; N, 3.93.

*2.1.6. 3β-Benzoyloxy-14β-cyanomethylandrosta-5,15-dien-17-one (**13**)*

A mixture of nitro compound **8** (0.22 g, 0.44 mmol), $NaHCO_3$ (0.259, 3.18 mmol), and triphenylphosphine (150 mg, 0.57 mmol) in aq. ethanol (30 ml, 10:1) was deoxygenated by bubbling nitrogen for 10 min and then refluxed for 1 h. The solution was cooled, diluted with dichloromethane and filtered. The precipitate was washed with dichloromethane and the combined filtrate was dried (Na_2SO_4) and evaporated. The concentrated solid residue was chromatographed on a silica gel column. Elution with ethyl acetate–toluene (5:95) gave nitrile **13** (0.188 g, 99%): m.p. 201–202 °C (MeOH). IR (KBr, cm^{-1}): 2940, 2250 (CN), 1720 (CO) 1710 (OBz), 1275, 1115. ^1H NMR δ: 1.07 (1H, m, 1α-H), 1.10 (3H, s, 19-Me), 1.25 (1H, m, 9α-H), 1.29 (3H, s, 18-Me), 1.29 (1H, m, 11β-H), 1.40 (1H, m, 11α-H), 1.59 (1H, m, 12β-H), 1.74 (1H, m, 2β-H), 1.76 (1H, m, 12α-H), 1.84 (1H, dt, J = 3 and 13 Hz, 1β-H), 1.99 (1H, m, 2α-H), 2.15 and 2.25 (2H, m, 7-H), 2.25 (1H, m, 8β-H), 2.34 and 2.59 (2H, two d, J = 17 Hz, 14^1-H), 2.42 (1H, m, 4β-H), 2.52 (1H, ddd, J = 2, 5 and 13 Hz, 4α-H), 4.83 (1H, m, 3α-H), 5.48 (1H, m, 6-H), 6.40 (1H, d, J = 6 Hz, 16-H), 7.44 (2H, m, *m*-Ph), 7.56 (1H, m, *p*-Ph), 7.64 (1H, d, J = 6 Hz, 15-H), 8.04 (2H, d, J = 7 Hz, *o*-Ph); ^{13}C NMR δ:17.85 (C-19), 18.87 (C-11), 18.95 (C-18), 24.81 ($C-14^1$), 27.11 (C-7), 27.46 (C-2), 33.75 (C-8), 35.59 (C-12), 36.20 (C-1), 37.76 (C-4), 38.88 (C-10), 42.81 (C-9), 51.45 (C-13), 52.57 (C-14), 74.17 (C-3), 117.29 (CN), 120.83 (C-6), 128.45 (Ph C-3), 129.68 (Ph C-2), 130.76 (Ph C-1), 133.00 (Ph C-4), 135.25 (C-16), 140.42 (C-5), 164.14 (C-15), 166.13 (3-OC(O)Ph), 213.49 (C-17). LRMS *m/z* (*I*,%): 430 ([*M*H]$^+$, 1), 308 ([*M*H−BzOH]$^+$, 100), 291 ([*M*H−BzOH−OH]$^+$, 19). MS^2 (430): 412 ([*M*H−H_2O]$^+$, 31), 308 ([*M*H−BzOH]$^+$, 100), 292 ([*M*H−BzOH−CH_4]$^+$, 11), 228 (17). MS^2 (308): 290 ([*M*H−BzOH−H_2O]$^+$, 100), 280 ([*M*H−BzOH−CO]$^+$, 50), 267 (21). HRMS calcd for $C_{21}H_{25}NO$ (M^+−BzOH): 307.1936. Found. 307.1924.

*2.1.7. 3β-Benzoyloxy-14β-cyanomethylandrost-5-en-17-one (**14**)*

To a solution of steroid **13** (0.167 g, 0.39 mmol) and ammonium formate (0.123 g, 1.95 mmol) in methanol (5 ml), palladium on charcoal (10%, 75 mg) was added. The resulting mixture was stirred for 12 h and then the solvent was evaporated. The obtained residue was dissolved in EtOAc, the organic phase was washed with brine, dried (Na_2SO_4) and concentrated. The solid residue was chromatographed on a silica gel column (toluene: EtOAc = 95:5) to afford nitrile **14** (0.134 g, 80%): m.p. 227–229 °C (MeOH). IR (KBr, cm^{-1}) 2945, 2240 (CN), 1740 (CO), 1715 (OBz), 1277, 1120.

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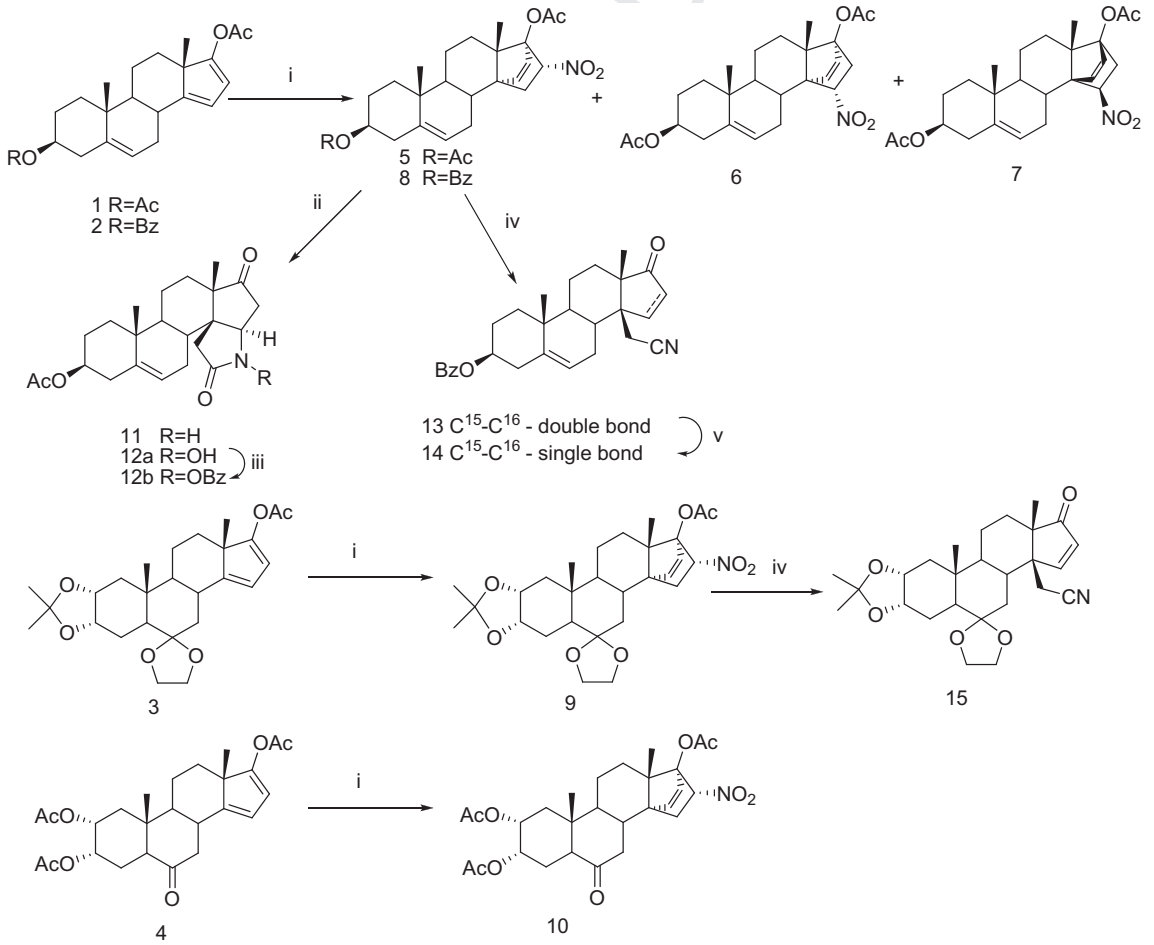
^{1}H NMR δ: 1.11 (3H, s, 19 Me), 1.18 (3H, s, 18 Me), 1.26 (1H, m, 1α-H), 1.31 (1H, m, 12β-H), 1.40 (1H, m, 11β-H), 1.41 (1H, m, 9α-H), 1.51 (1H, m, 12α-H), 1.71 (1H, m, 11α-H), 1.76 (1H, m, 2β-H), 1.79 (1H, m, 15β-H), 1.96 (1H, m, 8β-H), 1.98 (1H, m, 1β-H), 2.00 and 2.17 (2H, m, 7-H), 2.05 (1H, m, 2α-H), 2.27 (1H, m, 15α-H), 2.27 and 2.32 (2H, two d, J = 17 Hz, 14^1-H), 2.28 (1H, m, 16-H), 2.52 (2H, m, 4-H), 2.57 (1H, m, 16-H), 4.88 (1H, m, 3α-H), 5.45 (1H, m, 6-H), 7.45 (2H, m, *m*-Ph), 7.57 (1H, m, *p*-Ph), 8.05 (2H, d, J = 7 Hz, *o*-Ph); ^{13}C NMR δ: 14.85 (C-18), 19.51 (C-19), 19.95 (C-11), 25.85 and 25.88 (C-7 and C-15), 26.28 (C-14^1), 27.81 (C-2), 31.68 (C-12), 32.71 (C-16), 34.95 (C-8), 37.07 (C-1), 37.12 (C-10), 38.00 (C-4), 44.41 (C-9), 47.07 and 53.26 (C-13 and C-14), 74.19 (C-3), 118.19 (CN), 121.57 (C-6), 128.46 (Ph C-3), 129.70 (Ph C-2), 130.80 (Ph C-1), 133.00 (Ph C-4), 139.12 (C-5), 166.16 (3-OC(O)Ph), 219.57 (C-17). LRMS *m/z* (*I*,%): 432 ([*M*H]$^+$, 4), 310 ([*M*H−BzOH]$^+$, 100). MS^2 (432): 414 ([*M*H−H$_2$O]$^+$, 10), 310 ([*M*H−BzOH]$^+$, 100), 228 (15). MS^2 (310): 293 ([*M*H−BzOH−OH]$^+$, 100), 292 ([*M*H−BzOH−H$_2$O]$^+$, 87), 282 ([*M*H−BzOH−CO]$^+$, 40), 275 (41), 267 (86), 254 ([*M*H−BzOH−CH$_2$CHCHO]$^+$, 22). Anal. calcd. for $C_{28}H_{33}NO_3$ C, 77.93; H, 7.71; N, 3.25. Found. C, 77.48; H, 7.63; N, 3.14.

2.1.8. 14β-Cyanomethyl-6,6-ethylenedioxy-2α,3α-isopropylidendioxy-5α-androst-15-en-17-one (**15**)

Following the same procedure as for compound **13**, compound **9** (0.018 g, 0.04 mmol) was converted over 4 h into steroid **15** (0.009 g, 58%): m.p.107–109 °C (EtOAc-hexane). IR (KBr, cm^{-1}): 2940, 2250 (CN), 1720 (CO). ^{1}H NMR δ: 0.84 (3H, s, 19 Me), 0.93 (1H, m, 1α-H), 0.94 (1H, m, 9α-H), 1.25 (3H, s, 18-Me), 1.31 (3H, s, β-CH$_3$ acetonide), 1.32 (2H, m, 11-H), 1.44 (3H, s, α-CH$_3$ acetonide), 1.45 (1H, m, 7α-H), 1.54 (1H, ddd, J = 7, 9, 14 Hz, 12β-H), 1.66 (1H, m, 12α-H), 1.78 (1H, m, 5α-H), 1.80 (1H, dd, J = 4 and 8 Hz, 4β-H), 1.82 (1H, dd, J = 7 and 13 Hz, 1β-H), 1.97 (1H, dd, J = 3 and 13, 7β-H), 2.17 (1H, m, 4α-H), 2.21 (1H, m, 8β-H), 2.30 (1H, d, J = 17 Hz, 14^{1}β-H), 2.57 (1H, d, J = 17 Hz, 14^{1}α-H), 3.79 and 3.98 (2H, m, α-CH$_2$ dioxolane), 3.99 and 4.04 (2H, m, β-CH$_2$ dioxolane), 4.09 (1H, m, 2β-H), 4.27 (1H, m, 3β-H), 6.33 (1H, d, J = 6 Hz, 16-H), 7.48 (1H, d, J = 6 Hz, 15-H). ^{13}C NMR δ: 12.50 (C-19), 18.47 (C-18), 19.26 (C-11), 22.10 (C-4), 25.42 (C-14^1), 26.65 (β-CH$_3$ acetonide), 28.72 (α-CH$_3$ acetonide), 35.00 (C-12), 35.37 (C-8), 36.66 (C-7), 39.16 (C-10), 42.18 (C-1), 45.28 (C-5), 45.65 (C-9), 51.49 (C-13), 52.30 (C-14), 64.60 (α-CH$_2$ dioxolane), 65.73 (β-CH$_2$ dioxolane), 72.71 (C-3), 72.76 (C-2), 107.84 (Me$_2$*C* acetonide), 109.17 (C-6), 116.90 (CN), 134.26 (C-16), 162.99 (C-15), 212.74 (C-17). LRMS *m/z* (*I*,%): 441 ([*M*]$^+$, 0.1), 384 ([*M*H−Me$_2$CO]$^+$, 100), 322 ([*M*H−Me$_2$CO−CH$_2$O−H$_2$O]$^+$, 11). MS^2 (384): 340 ([*M*H−Me$_2$CO−CH$_2$O]$^+$, 100), 322 ([*M*H−Me$_2$CO−CH$_2$O−H$_2$O]$^+$, 45). HRMS calcd for $C_{26}H_{35}NO_5$: 441.2515. Found 441.2523.

3. Results and discussion

Dienyl acetates **1–4** in this work were synthesized from appropriate enones [4]. The cycloaddition of nitroethylene to steroids **1** and **2** in refluxing benzene gave each of the three adducts in excel-



Scheme 1. Reagents and conditions: (i) nitroethylene, PhH, reflux; (ii) $NaHCO_3$, H_2O, EtOH, reflux; (iii) BzCl, Py, rt.; (iv) $NaHCO_3$, Ph_3P, H_2O, EtOH, reflux; (v) HCO_2NH_4, Pd/C, MeOH, rt.

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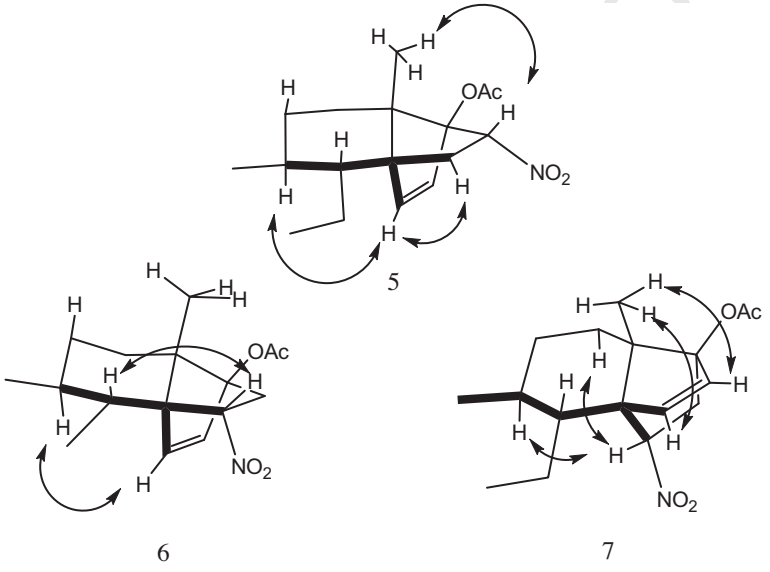
lent overall yield (86–99%), and the β-side head-to-head adducts **5** and **8** were predominant (about 85% of the mixture). In the case of functionalized steroids **3** and **4**, the result was less unambiguous: we were able to isolate only compounds **9** and **10** in 49% and 70% yield due to a heavy contamination of the reaction mixtures by degradation products. Here, a higher excess of the dienophile was used and the reaction took 16 h to complete (See Scheme 1).

The low efficiency of the reaction with **3** can be attributed to a steric hindrance [5] caused by substituents in the B ring as well as to lability. Product distribution was established using the Diels–Alder reaction with steroid **1** as an example. Products **5–7** were separated and their structures were proven by 2D NMR spectroscopy. Key interactions in NOESY spectra of the compounds for structure assignment are presented in Scheme 2.

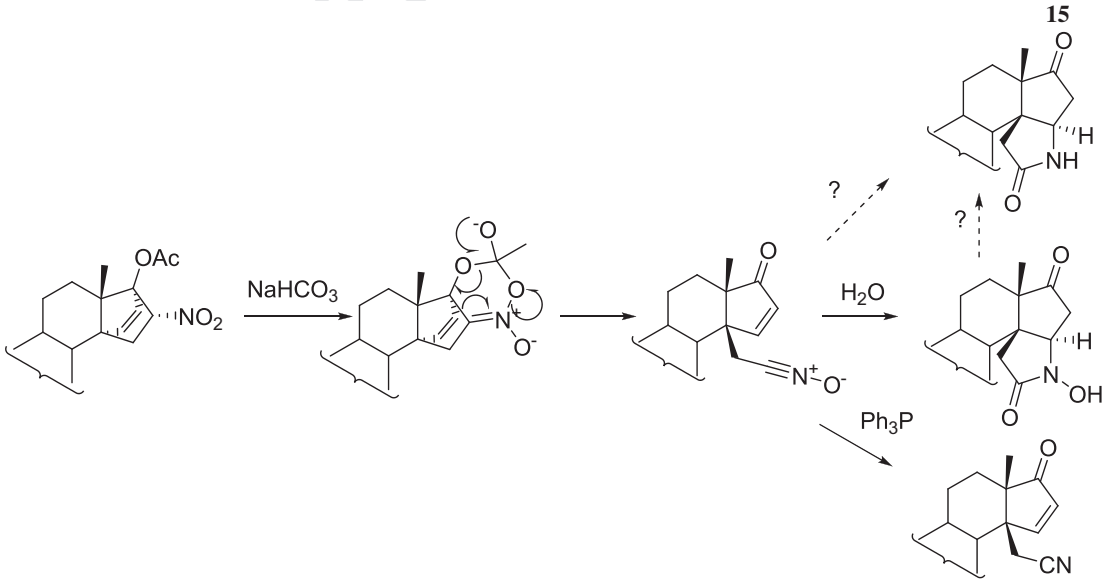
Based on obtained data, the ratio of the isomers in the cycloaddition reaction was found to be **5:6:7** = 87:11:2. Similar data were received in the cycloaddition reaction of steroid **2** with nitroethylene. Therefore, the reaction proceeded in accordance with previous observations of steroidal 14,16-dienes [6,7]; the addition of nitroethylene happens from β-face (compounds **5** and **6**) and highly selectively gives head-to-head adduct. Structures of major and two minor products correspond to those obtained in the reaction of 14,16-dienolacetate with phenyl vinyl sulphone [8] where three isomers had been isolated but with different ratio.

Tandem mass-spectrometry analysis revealed an interesting common feature of the fragmented protonated molecules of the nitroadducts. No ions of retro Diels–Alder reaction were found among daughter ions. Instead, after the elimination of acetic acid, the bridged part of the molecules lost ethylene but not nitroethylene.

Nitroadducts **5, 9** and **10** underwent several representative reactions; these had been characteristic for the nitroadduct cleavage in the estrane series [1,2] in order to allow the transformations, distribution and yields of products to be compared. The reproducibility of these procedures in the androstane series is important for



Scheme 2. Ring C/D region of cycloadducts **5–7** and interactions in NOESY spectra.



Scheme 3. Pathways of the nitroadducts' fragmentation.

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the development of an approach to D-ring modified analogs of steroidal hormones.

The solvolysis of steroid **5** in aqueous ethanol in the presence of $NaHCO_3$ followed by column chromatography gave an inseparable mixture of two major products; these products were formulated as lactam **11** and hydroxylactam **12a** based on preliminary NMR examination. The separation of components for a complete characterization was accomplished by exploiting different chemical properties. In contrast to compound **11**, that hydroxylactam **12a** can be easily esterified at ambient temperature. Thus, when the crude residue was treated with benzoyl chloride in pyridine, only compound **12a** reacted, and, upon work-up and separation, steroids **12b** and **11** were isolated in 20% and 26% yield respectively. The result of the reaction differed from that obtained in the estrane series where the ratio of products was 1:2 and overall yield was 60%. Possibly, a lower yield of compound **11** was connected with certain lability of 3-acetoxy group under basic condition of the reaction and, for the synthesis of nitriles, benzoyloxy derivative **8** was used.

We consider nitrile group [9–11] to be an attractive functionality for further lengthening the chain at C-14 (in the case of hapten synthesis) as well as a stable function during the formation of the steroidal side chain (in the case of brassinosteroid analog synthesis). Accordingly, we subjected nitro compounds **8** and **9** to reductive cleavage with triphenylphospine in the presence of $NaHCO_3$. Both compounds reacted to give 14-cyanomethyl derivatives as expected. However, again, as with cycloaddition, Δ^5-derivative **13** was isolated almost quantitatively and steroid **15** only in 58% yield. The conversion of steroid **9** lasted much longer, and though minor components were not analyzed, judging from their polarity, one can expect formation of lactam to accompany the fragmentation of nitroadducts in the estrane series.

The saturation of the 15-double bond in steroid **13** was fulfilled regioselectively by the transfer hydrogenation [12,13] to afford steroid **14** in 80% yield and some minor polar compounds. The minor compounds were neither isolated nor analyzed but their formation can be attributed to the simultaneous reduction of carbonyl or nitrile groups [14].

A unique role played by a combination of functional groups and bridged moiety in compounds **5** and **8–10** for the course of the above transformations were confirmed by using the mixture of minor 15-nitro derivatives. Thus, when the mixture of minor isomers of adduct **8** was treated either with Ph_3P or underwent solvolysis, neither nitrile nor lactam derivatives were found. A slow course of reaction led to the partial hydrolysis of protective groups at C-3 and C-17. This observation favors our suggestion that the acetoxy group migrates to nitro group with the formation of 6-membered cyclic mixed anhydride as the first step of the process [1] (Scheme 3).

The obtained results prove the high selectivity of the cycloaddition of nitroethylene to the steroidal 14,16-dienyl acetates and reproducibility of the reductive cleavage reaction of bridged nitro compounds. Consequently, nitroadducts **5, 9** and **10** can serve as an effective starting point for the synthesis 14β-substituted analogs of natural steroids.

Acknowledgments

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