Electronic Supplementary Material (ESI) for MedChemComm.

Identification of Gli-mediated transcription inhibitors through synthesis and evaluation of taepeenin D analogues

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Supplementary Data Part I

Synthetic Procedures, Characterization Data and Biological Evaluation

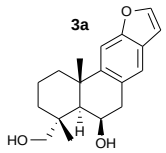
1. Synthetic Procedures, Characterization Data

1.1. General

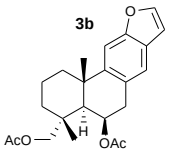
All reactions were carried out under a dry argon atmosphere with anhydrous, freshly distilled solvents unless otherwise noted. All reactions were magnetically stirred with Teflon stir bars, and temperatures were measured externally. Reactions requiring anhydrous conditions were carried out in oven dried (120 °C, 24 h) or flame dried (vacuum < 0.5 Torr) glassware. Yields refer to chromatographically and spectroscopically (^{1}H NMR) homogeneous materials unless otherwise noted. All reagents were obtained from Aldrich Chemical Co. Inc. and used without further purification. All reactions were monitored by thin layer chromatography (TLC) carried out on 0.25-mm E.Merck silica gel plates (60F-254). E.Merck silica gel (60, particle size 0.040-0.063 mm) was used for flash column chromatography.[1] Nuclear magnetic resonance (NMR) spectra were recorded on a Bruker Advance DRX-500 instrument. Chemical shifts are measured in parts per million (δ) and residual non-deuterated solvent peak(s) were used to reference NMR spectra.[2] Multiplicities are designated as singlet (s), doublet (d), triplet (t), or multiplet (m). Broad or obscured peaks are indicated as “br” or “obs” respectively. Copies of all spectra are provided in Part II. Electron spray ionization (ES) spectra were recorded on a Waters Q-TOF Ultima API, Micromass UK Limited high resolution mass spectrometer. Optical rotations were recorded using a Perkin-Elmer 241 polarimeter at the sodium-D line (589 nm) using a 10 cm path-length cell in the solvent and concentration indicated.

[1] W. C. Still, M. Kahn and A. Mitra, *J. Org. Chem.*, 1978, **43**, 2923–2925.
[2] H. E. Gottlieb, V. Kotlyar and A. Nudelman, *J. Org. Chem.*, 1997, **62**, 2923–2925.

1.2. Preparation of taepeenin D analogues.



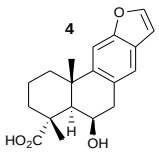
(4*R*,4a*R*,5*R*,11b*S*)-1,2,3,4,4a,5,6,11b-octahydro-4-(hydroxymethyl)-4,11b-dimethylphenanthro[3,2-*b*]furan-5-ol (**3a**). To a solution of ester **2a**[3] (16.6 mg, 50.6 µmol) in THF (2 mL) was added at 0 °C $LiAlH_4$ (6.0 mg, 156.7 µmol). After 2 h, 5% aqueous HCl was added dropwise and the mixture was allowed to gradually warm to ambient temperature. After 1 h the reaction mixture was filtered through Celite® and the filter cake was washed several times with EtOAc (4 × 5 mL), dried over Na_2SO_4 and concentrated under reduced pressure. The residue thus obtained was purified by flash column chromatography (SiO_2, hexane/EtOAc 9:1→8:2) to provide upon solvent removal diol **3a** as white foam (12.9 mg, 85% yield). **Optical Rotation**: $[\alpha]_D^{25}$ +35 (*c* 0.47, acetone). **^{1}H NMR** (500 MHz, $CDCl_3$) *δ* 7.54 (d, *J* = 1.5 Hz, 1H), 7.46 (s, 1H), 7.26 (s, 1H), 6.65 (s, 1H), 4.68−4.63 (m, 1H), 3.60 (d, *J* = 10.8 Hz, 1H), 3.37−3.29 (m, 2H), 3.02 (dd, *J* = 16.8, 2.9 Hz, 1H), 2.25 (d, *J* = 12.6 Hz, 1H), 2.00−1.85 (m, 3H), 1.76−1.68 (m, 2H), 1.58 (s, 3H), 1.53−1.47 (m, 1H), 1.42−1.36 (m, 1H), 1.29 (s, 3H). **^{13}C NMR** (125 MHz, $CDCl_3$) *δ* 154.5, 146.6, 145.0, 127.0, 125.2, 120.8, 107.2, 105.8, 72.9, 66.6, 48.6, 41.8, 40.6, 38.8, 38.2, 37.1, 26.7, 19.7, 18.9. **HR-ESI**: m/z: 323.16219; [*M*+*Na*$^+$] for the compound $C_{19}H_{24}O_3$ requires 323.16176.



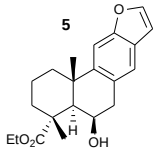
((4*R*,5*R*,11b*S*)-5-acetoxy-4,11b-dimethyl-1,2,3,4,4a,5,6,11b-octahydrophenanthro[3,2-*b*]furan-4-yl)methyl acetate (**3b**). To a solution of diol **3a** (12.9 mg, 42.9 µmol) in dichloromethane (0.7 mL) stirring at 0 °C, a solution of acetic anhydride (40.0 mg, 387 µmol) in dichloromethane (0.45 mL) and DMAP (47.2 mg, 387 µmol) were added. After 1h, water (5 mL) was added, followed by extractions with Et_2O (3 × 5 mL). The combined organic phases were washed with brine (10 mL), dried over Na_2SO_4 and concentrated under reduced pressure. The residue thus obtained was purified by flash column chromatography (SiO_2, hexane/ethyl acetate 95:5) to provide upon solvent removal diacetoxy analog **3b** as white foam (10.4 mg, 65% yield). **Optical Rotation**: $[\alpha]_D^{25}$ –13 (*c* 0.67, acetone). **^{1}H NMR** (500 MHz, $CDCl_3$) *δ* 7.56 (d, *J* = 2.1 Hz, 1H), 7.50 (s, 1H), 7.25 (s, 1H), 6.66 (d, *J* = 2.0 Hz, 1H), 5.64 (d, *J* = 5.1 Hz, 1H), 4.00 (d, *J* = 11.4 Hz, 1H), 3.84 (d, *J* = 11.4 Hz, 1H), 3.28 (dd, *J* = 17.9, 5.3 Hz, 1H), 3.12 (d, *J* = 17.7 Hz, 1H), 2.32 (d, *J* = 12.9 Hz, 1H), 2.03 (s, 3H), 2.02 (s, 3H), 1.96−1.88 (m, 2H), 1.76−1.70 (m, 1H), 1.62 (s, 3H), 1.55−1.42 (m, 2H), 1.37 (d, *J* = 13.3 Hz, 1H), 1.12 (s, 3H). **^{13}C NMR** (125 MHz, $CDCl_3$) *δ* 171.0, 170.7, 154.5, 145.4, 145.3,

[3] M. Chatzopoulou, A. Antoniou, E. N. Pitsinos, M. Bantzi, S. D. Koulocheri, S. A. Haroutounian and A. Giannis, *Org. Lett.*, 2014, **16**, 3344−3347

125.8, 125.6, 120.9, 107.4, 105.8, 72.3, 68.0, 46.5, 42.0, 38.4, 37.4, 37.2, 37.0, 27.7, 21.7, 20.9, 19.1, 18.8. **HR-ESI**: m/z: 407.18323; [*M*+*Na*$^+$] for the compound $C_{23}H_{28}O_5$ requires 407.18289.



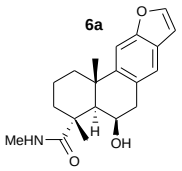
(4*R*,4a*R*,5*R*,11b*S*)-1,2,3,4,4a,5,6,11b-octahydro-5-hydroxy-4,11b-dimethylphenanthro[3,2-*b*]furan-4-carboxylic acid (**4**). To a solution of ester **2a**[3] (13.0 mg, 39.6 µmol) in DMSO (1 mL) stirring at ambient temperature ^{t}BuOK (18.0 mg, 158 µmol) was added. After 24 h, the reaction mixture was poured in 0.5 N HCl (2 mL) and stirred for 30 min, followed by extractions with Et_2O (5 × 2 mL). The organic phase was washed with water (5 mL), brine (5 mL), dried over Na_2SO_4 and concentrated under reduced pressure. The solid residue was purified by flash column chromatography (SiO_2, hexane/EtOAc 8:2→6:4) to provide upon solvent removal carboxylic acid **4** as white solid (10.2 mg, 82% yield). **^{1}H NMR** (500 MHz, $CDCl_3$) *δ* 7.54 (d, *J* = 2.1 Hz, 1H), 7.45 (s, 1H), 7.25 (s, 1H), 6.65 (d, *J* = 1.4 Hz, 1H), 4.39−4.34 (m, 1H), 3.36 (dd, *J* = 17.0, 4.9 Hz, 1H), 2.99 (d, *J* = 17.0 Hz, 1H), 2.34 (s, 1H), 2.27 (d, *J* = 12.8 Hz, 1H), 1.97−1.69 (m, 4H), 1.65 (s, 3H), 1.62 (s, 3H), 1.59−1.53 (m, 1H). **^{13}C NMR** (125 MHz, $CDCl_3$) *δ* 184.7, 154.5, 145.7, 145.1, 126.5, 125.6, 121.3, 107.2, 105.8, 69.0, 48.2, 47.7, 41.5, 41.2, 38.2, 37.8, 27.2, 19.0, 18.4.



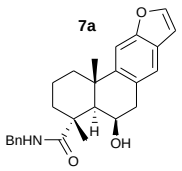
(4*R*,4a*R*,5*R*,11b*S*)-ethyl 1,2,3,4,4a,5,6,11b-octahydro-5-hydroxy-4,11b-dimethylphenanthro[3,2-*b*]furan-4-carboxylate (**5**). To a stirred solution of acid **4** (6.6 mg, 20.9 µmol) in DMF (1.0 mL) was added $NaHCO_3$ (5.2 mg, 62 µmol) and the mixture was cooled to 0 °C. Then, iodoethane (5 µL, 62 µmol) was added dropwise and the mixture was allowed to gradually warm up to ambient temperature over 5 h. The reaction was quenched by the addition of glacial acetic acid (5 µL, 88 µmol) and after 1 h it was diluted with diethyl ether (10 mL), washed with water (2 × 5 mL) and brine (5 mL), dried over Na_2SO_4, filtered, and concentrated under reduced pressure. The residue thus obtained was purified by flash column chromatography (SiO_2, hexane/EtOAc 95:5) to provide ethyl ester **5** (5.0 mg, 14.6 µmol; 70% yield) as white foam. **Optical Rotation**: $[\alpha]_D^{25}$ +12 (*c* 0.42, acetone). **1H NMR** (500 MHz, $CDCl_3$) δ 7.55 (d, *J* = 2.1 Hz, 1 H), 7.46 (s, 1 H), 7.27 (s, 1 H), 6.65 (dd, *J* = 2.0, 0.9 Hz, 1 H), 4.28–4.23 (br m, 1 H), 4.23−4.08 (m, 2 H), 3.33 (dd, *J* = 16.9, 5.1 Hz, 1 H), 3.01 (br d, *J* = 16.8 Hz, 1 H), 2.35 (s, 1 H), 2.28 (br d, *J* = 12.7 Hz, 1 H), 1.98–1.71 (m, 3 H), 1.69–1.56 (m, 3 H), 1.64 (s, 3 H), 1.62 (s, 3 H), 1.25 (t, *J* = 7.10 Hz, 3 H). **^{13}C NMR** (125 MHz, $CDCl_3$) δ 178.7, 154.5, 145.9, 145.1, 126.6, 125.5, 121.3, 107.3, 105.8, 69.0, 60.7, 48.2, 47.9, 41.6, 41.2, 38.0, 37.9, 27.2, 19.0, 18.6, 14.3. **HR-ESI**: *m/*z: 365.17228; [*M*+*Na*$^+$] for the compound $C_{21}H_{26}O_4$ requires 365.17288.

General procedure for the preparation of 6-hydroxy amido-analogs **6a**−**8a**.

To a solution of acid **4** (6.0 mg, 19 µmol) in dichloromethane (0.2 mL) stirring at ambient temperature, PyBOP (11.0 mg, 21.0 µmol) and DIPEA (4 µL, 23 µmol) were added. After 2 h, a solution of the appropriate amine (40 µmol) and DIPEA (7 µL, 40 µmol) in DCM (3.5 mL) was added and the reaction mixture was stirred at ambient temperature for 24−48 h. Then brine was added, and the mixture was extracted with Et_2O. The organic phases were dried over Na_2SO_4 and concentrated under reduced pressure. The solid residue was purified by flash column chromatography (SiO_2, hexane/EtOAc 8:2→5:5) to provide upon solvent removal the corresponding amides:

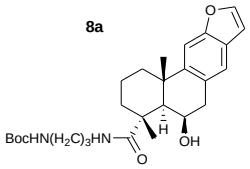


(4*R*,4a*R*,5*R*,11b*S*)-5-hydroxy-*N*,4,11b-trimethyl-1,2,3,4,4a,5,6,11b-octahydrophenanthro[3,2-*b*]furan-4-carboxamide (**6a**). White foam (4.2 mg, 68% yield). **Optical Rotation**: $[\alpha]_D^{25}$ +21 (*c* 0.18, acetone). **^{1}H NMR** (500 MHz, $CDCl_3$) *δ* 7.54 (d, *J* = 1.8 Hz, 1H), 7.44 (s, 1H), 7.25 (s, 1H), 6.64 (s, 1H), 6.06 (br s, 1H), 4.29−4.22 (m, 1H), 3.37 (dd, *J* = 17.0, 5.6 Hz, 1H), 2.96 (dd, *J* = 17.0, 2.3 Hz, 1H), 2.84 (d, *J* = 4.5 Hz, 3H), 2.28 (br d, *J* = 11.6 Hz, 1H), 2.23 (s, 1H), 2.00−1.84 (m, 3H), 1.81−1.74 (m, 1H), 1.66 (s, 3H), 1.60 (s, 3H), 1.57−1.51 (s, 1H). **^{13}C NMR** (125 MHz, $CDCl_3$) *δ* 179.3, 154.5, 146.3, 145.0, 126.8, 125.4, 121.0, 107.0, 105.8, 68.6, 48.7, 48.0, 41.3, 40.9, 38.1, 38.0, 26.9, 26.8, 19.1, 19.0. **HR-ESI**: m/z: 350.17287; [*M*+*Na*$^+$] for the compound $C_{20}H_{25}NO_3$ requires 350.17266.



(4*R*,4a*R*,5*R*,11b*S*)-*N*-benzyl-5-hydroxy-4,11b-dimethyl-1,2,3,4,4a,5,6,11b-octahydrophenanthro-[3,2-*b*]furan-4-carboxamide (**7a**). White foam (5.8 mg, 75% yield). **Optical Rotation**: $[\alpha]_D^{25}$ +27 (*c* 0.16, acetone). **^{1}H NMR** (500 MHz, $CDCl_3$) *δ* 7.55 (d, *J* = 0.7 Hz, 1H), 7.45 (s, 1H), 7.38-7.32 (m, 2H), 7.31-7.24 (m, 5H), 6.65 (s, 1H), 6.28 (s, 1H), 4.53 (dd, *J* = 14.7, 5.7 Hz, 1H), 4.43 (dd, *J* = 14.7, 5.1 Hz, 1H), 4.32−4.27 (m, 1H), 3.37 (dd, *J* = 17.0, 5.7 Hz, 1H), 2.97 (dd, *J* = 17.1, 2.0 Hz, 1H), 2.32−2.24 (m, 2H), 1.95−1.86 (m, 2H), 1.83−1.76 (m, 1H), 1.67 (s, 3H), 1.62-1.56 (s, 5H). ^{13}C NMR (125 MHz, $CDCl_3$) *δ* 178.56, 154.5, 146.2, 145.1, 138.4, 128.8, 127.8, 127.6, 126.8, 125.4, 121.0, 107.0, 105.8, 68.6, 48.7, 48.0, 44.0, 41.3, 40.9, 38.4, 38.0, 26.9, 19.1, 19.0. **HR-ESI**: m/z: 426.20441; [*M*+*Na*$^+$] for the compound $C_{26}H_{29}NO_3$ requires 426.20396.

8a

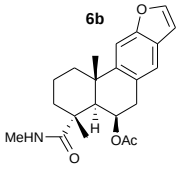


tert-butyl (3-((4*R*,4a*R*,5*R*,11b*S*)-5-hydroxy-4,11b-dimethyl-1,2,3,4,4a,5,6,11b-octahydrophenanthro[3,2-*b*]furan-4-carboxamido)propyl)carbamate (**8a**). White foam (5.8 mg, 66% yield). **Optical Rotation**: $[\alpha]_D^{25}$ +19 (*c* 0.15, acetone). **^{1}H NMR** (500 MHz, $CDCl_3$) *δ* 7.53 (d, *J* = 2.0 Hz, 1H), 7.44 (s, 1H), 7.24 (s, 1H), 6.74 (br s, 1H), 6.64 (d, *J* = 2.0 Hz, 1H), 4.86 (br s, 1H), 4.32−4.27 (m, 1H), 3.42−3.08 (m, 5H), 2.98 (dd, *J* = 16.9, 2.9 Hz, 1H), 2.29 (br d, *J* = 11.9 Hz, 1H), 2.20 (d, *J* = 2.2 Hz, 1H), 1.97−1.86 (m, 2H), 1.82−1.74 (m, 1H), 1.70 (s, 3H), 1.68−1.62 (m, 3H), 1.61 (s, 3H), 1.57−1.52 (m, 1H), 1.43 (s, 9H). **^{13}C NMR** (125 MHz, $CDCl_3$) *δ* 179.1, 156.7, 154.5, 146.5, 144.9, 127.1, 125.3, 120.8, 106.9, 105.8, 79.6, 68.3, 48.7, 48.0, 41.2, 40.8, 38.1, 38.1, 37.3, 36.1, 30.3, 28.4, 26.7, 19.1, 19.1. **HR-ESI**: m/z: 493.26750; [*M*+*Na*$^+$] for the compound $C_{27}H_{38}N_2O_5$ requires 493.26729.

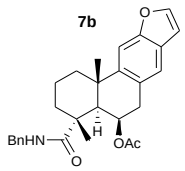
General procedure for the preparation of 6-acetoxy amido-analogs **6b**−**8b**.

To a solution of the appropriate alcohol **6a**−**8a** (8−10 µmol) in DCM (250 µL) stirring at 0 °C acetic anhydride (2 equiv) and DMAP (2 equiv) were added. After 24 h, water was added, followed by extractions with Et_2O. The combined organic phases were washed with brine, dried over Na_2SO_4 and concentrated under reduced pressure. The residue thus obtained was purified by flash column chromatography (SiO_2, hexane/EtOAc) to provide upon solvent removal the corresponding acetoxy analogs **6b**−**8b**.

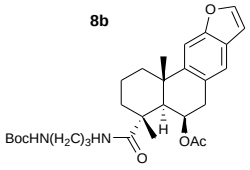
6b



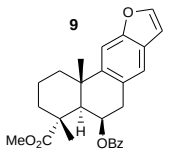
(4*R*,4a*R*,5*R*,11b*S*)-4,11b-dimethyl-4-(methylcarbamoyl)-1,2,3,4,4a,5,6,11b-octahydrophenanthro[3,2-*b*]furan-5-yl acetate (**6b**). White foam, 81% yield. **Optical Rotation**: $[\alpha]_D^{25}$ −26 (*c* 0.31, acetone). **^{1}H NMR** (500 MHz, $CDCl_3$) *δ* 7.54 (d, *J* = 2.1 Hz, 1H), 7.46 (s, 1H), 7.22 (s, 1H), 6.65 (d, *J* = 1.5 Hz, 1H), 5.96 (br d, *J* = 2.8 Hz, 1H), 5.30 (d, *J* = 5.0 Hz, 1H), 3.33 (dd, *J* = 17.8, 5.0 Hz, 1H), 3.01 (d, *J* = 17.7 Hz, 1H), 2.86 (d, *J* = 4.7 Hz, 3H), 2.62 (s, 1H), 2.31 (br d, *J* = 12.7 Hz, 1H), 2.05-1.97 (m, 1H), 2.01 (s, 3 H), 1.92−1.82 (m, 1H), 1.81−1.74 (m, 1H), 1.70−1.65 (m, 1H), 1.62 (s, 3H), 1.50 (br d, *J* = 13.0 Hz, 1H), 1.41 (s, 3H). **^{13}C NMR** (125 MHz, $CDCl_3$) *δ* 178.5, 170.9, 154.5, 145.3, 145.2, 126.0, 125.6, 121.0, 107.2, 105.9, 70.7, 47.9, 46.8, 41.6, 38.2, 38.0, 37.5, 27.7, 26.9, 21.8, 19.0, 18.1. **HR-ESI**: m/z: 392.18319; [*M*+*Na*$^+$] for the compound $C_{22}H_{27}NO_4$ requires 392.18322.



(4*R*,4a*R*,5*R*,11b*S*)-4-(benzylcarbamoyl)-4,11b-dimethyl-1,2,3,4,4a,5,6,11b-octahydrophenanthro[3,2-*b*]furan-5-yl acetate (**7b**). White foam, 78% yield. **Optical Rotation**: $[\alpha]_D^{25}$ −19 (*c* 0.47, acetone). **^{1}H NMR** (500 MHz, $CDCl_3$) *δ* 7.55 (d, *J* = 2.0 Hz, 1H), 7.47 (s, 1H), 7.39−7.27 (m, 5H), 7.23 (s, 1H), 6.65 (br s, 1H), 6.17 (br s, 1H), 5.39 (d, *J* = 4.5 Hz, 1H), 4.56 (dd, *J* = 14.7, 5.8 Hz, 1H), 4.44 (dd, *J* = 14.7, 4.9 Hz, 1H), 3.34 (dd, *J* = 17.8, 5.1 Hz, 1H), 3.03 (d, *J* = 17.8 Hz, 1H), 2.67 (s, 1H), 2.33 (d, *J* = 12.6 Hz, 1H), 2.08−2.02 (m, 1H), 1.99 (s, 3H), 1.94−1.83 (m, 1H), 1.82−1.74 (m, 1H), 1.67 (dd, *J* = 12.7, 2.7 Hz, 1H), 1.63 (s, 3H), 1.57 (br d, *J* = 12.6 Hz, 1H), 1.43 (s, 3H). **^{13}C NMR** (125 MHz, $CDCl_3$) *δ* 177.8, 170.7, 154.5, 145.3, 145.2, 138.3, 128.8, 127.9, 127.5, 126.1, 125.6, 121.0, 107.2, 105.9, 70.8, 47.8, 46.7, 44.2, 41.5, 38.6, 38.0, 37.4, 27.6, 21.8, 19.0, 18.2. **HR-ESI**: m/z: 468.21430; [*M*+*Na*$^+$] for the compound $C_{28}H_{31}NO_4$ requires 468.21453.



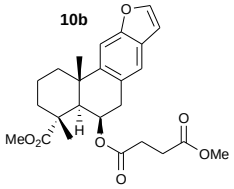
(4*R*,4a*R*,5*R*,11b*S*)-4-((3-((tert-butoxycarbonyl)amino)propyl)carbamoyl)-4,11b-dimethyl-1,2,3,4,4a,5,6,11b-octahydrophenanthro[3,2-*b*]furan-5-yl acetate (**8b**). White foam, 84% yield. **Optical Rotation**: $[\alpha]_D^{25}$ −15 (*c* 0.27, acetone). **^{1}H NMR** (500 MHz, $CDCl_3$) *δ* 7.54 (d, *J* = 1.9 Hz, 1H), 7.46 (s, 1H), 7.21 (s, 1H), 6.74 (br s, 1H), 6.64 (s, 1H), 6.17 (bs, 1H), 5.31 (d, *J* = 4.3 Hz, 1H), 4.98−4.87 (m, 1H), 3.48−3.42 (m, 1H), 3.34 (dd, *J* = 17.8, 5.2 Hz, 1H), 3.30−3.16 (m, 3H), 3.02 (d, *J* = 17.8 Hz, 1H), 2.63 (s, 1H), 2.32 (d, *J* = 12.5 Hz, 1H), 2.01 (s, 3H), 1.98−1.83 (m, 2H), 1.81−1.73 (m, 1H), 1.70-1.64 (m, 2H), 1.63 (s, 3H), 1.54−1.50 (m, 1H), 1.46 (s, 3H), 1.44 (s, 9H). **^{13}C NMR** (125 MHz, $CDCl_3$) *δ* 178.1, 170.9, 156.8, 154.5, 145.4, 145.2, 126.1, 125.5, 120.9, 107.2, 105.9, 79.3, 70.8, 47.8, 46.6, 41.5, 38.4, 38.0, 37.4, 36.4, 30.1, 28.4, 27.7, 21.8, 19.1, 18.0. **HR-ESI**: m/z: 535.27811; [*M*+*Na*$^+$] for the compound $C_{29}H_{40}N_2O_6$ requires 535.27786.



(4*R*,4a*R*,5*R*,11b*S*)-methyl 5-(benzoyloxy)-4,11b-dimethyl-1,2,3,4,4a,5,6,11b-octahydrophenanthro[3,2-*b*]furan-4-carboxylate (**9**). To a solution of alcohol **2a** (7.6 mg, 23 µmol) in pyridine (0.7 mL) stirring at 0 °C benzoyl chloride (5 mg, 35 µmol) and DMAP (28.3 mg, 231 µmol) were added. The reaction mixture was allowed to warm to ambient temperature and then heated to 70 °C. After 24 h, water (5 mL) was added, followed by extractions with Et_2O (3 × 2 mL). The combined organic phases were washed sequentially with 10% aqueous HCl solution (5 mL),

saturated aqueous $NaHCO_3$ solution (5 mL), and brine (10 mL), dried over Na_2SO_4 and concentrated under reduced pressure. The residue thus obtained was purified with by flash column chromatography (SiO_2, hexane/EtOAc 95:5) to produce upon solvent removal benzoyloxy analog **9** as a white foam (8.9 mg, 88% yield). **Optical Rotation**: $[\alpha]_D^{25}$ −52 (*c* 0.70, acetone). **^{1}H NMR** (500 MHz, $CDCl_3$) *δ* 7.97 (d, *J* = 7.3 Hz, 2H), 7.55 (d, *J* = 2.1 Hz, 1H), 7.54−7.48 (m, 2H), 7.42−7.36 (m, 2H), 7.24 (s, 1H), 6.64 (d, *J* = 1.6 Hz, 1H), 5.51 (d, *J* = 5.6 Hz, 1H), 3.72 (s, 3H), 3.48 (dd, *J* = 18.1, 5.6 Hz, 1H), 3.26 (d, *J* = 18.0 Hz, 1H), 2.67 (s, 1H), 2.41 (d, *J* = 12.6 Hz, 1H), 2.00−1.89 (m, 1H), 1.88−1.75 (m, 2H), 1.78 (s, 3H), 1.74-1.64 (m, 2H), 1.49 (s, 3H). **^{13}C NMR** (125 MHz, $CDCl_3$) *δ* 178.6, 166.7, 154.4, 145.3, 145.2, 132.9, 130.6, 129.6, 128.4, 126.0, 125.6, 121.0, 107.0, 105.8, 71.9, 52.3, 48.1, 46.9, 41.5, 38.6, 37.9, 37.1, 28.2, 18.9, 18.5. **HR-ESI**: m/z: 455.18324; [*M*+*Na*$^+$] for the compound $C_{27}H_{28}O_5$ requires 455.18289.

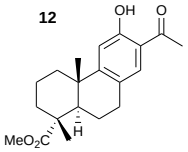
10b



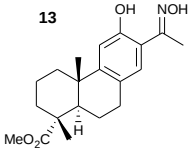
(4*R*,4a*R*,5*R*,11b*S*)-4-(methoxycarbonyl)-4,11b-dimethyl-1,2,3,4,4a,5,6,11b-octahydrophenanthro[3,2-*b*]furan-5-yl methyl succinate (**10b**). To a solution of alcohol **2a** (11.5 mg, 35.0 µmol) in DCM (1 mL) stirring at 0 °C succinic anhydride (56.5 mg, 574 µmol), DMAP (35.1 mg, 287 µmol) and pyridine (1 mL) were added. The reaction mixture was allowed to warm to ambient temperature and then heated to 60 °C. After 48 h, water (5 mL) was added, followed by extractions with AcOEt (3 × 2 mL). The combined organic phases were washed sequentially with 10% aqueous HCl solution (5 mL), saturated aqueous $NaHCO_3$ solution (5 mL), and brine (10 mL), dried over Na_2SO_4 and concentrated under reduced pressure. The residue thus obtained was partially purified by flash column chromatography (SiO_2, hexane/EtOAc/AcOH 90:9:1) to produce upon solvent removal a white foam (9.0 mg) that was used without further purification in the next step.

To a solution of the above derivative (**10a**) in a mixture of toluene (0.58 mL) and MeOH (0.42 mL) at ambient temperature, a 2.0 M $TMSCH_2N_2$ solution in hexanes was added dropwise until a stable yellow color remained in the reaction mixture. After 30 min the reaction was quenched with acetic acid and the solvents were removed under reduced pressure. The residue thus obtained was purified by preparative TLC (SiO_2, hexane/EtOAc 8:2) to provide the desired methyl aster **10b** as white foam (6.5 mg, 32% yield from alcohol **2a**). **Optical Rotation**: $[\alpha]_D^{25}$ −24 (*c* 0.30, acetone). **^{1}H NMR** (500 MHz, $CDCl_3$) *δ* 7.55 (d, *J* = 2.1 Hz, 1H), 7.46 (s, 1H), 7.23 (s, 1H), 6.65 (d, *J* = 1.9 Hz, 1H), 5.30−5.26 (m, 1H), 3.70 (s, 3H), 3.62 (s, 3H), 3.35 (dd, *J* = 17.8, 5.5 Hz, 1H), 3.08 (d, *J* = 17.6 Hz, 1H), 2.64−2.54 (m, 4H), 2.53 (s, 1 H), 2.34 (d, *J* = 13.3 Hz,1H), 1.94−1.86 (m, 1H), 1.82−1.72 (m, 2H), 1.72−1.65 (m, 1 H), 1.65-1.57 (m, 1H), 1.61 (s, 3H), 1.46 (s, 3H). **^{13}C NMR**

(125 MHz, $CDCl_3$) δ 178.5, 172.5, 171.9, 154.5, 145.3, 126.0, 125.6, 121.0, 107.1, 105.9, 71.4, 52.3, 51.8, 48.0, 46.7, 41.6, 38.6, 38.0, 37.0, 29.7, 28.9, 27.5, 18.9, 18.2. **HR-ESI**: m/z: 465.18910; [*M+Na*$^+$] for the compound $C_{25}H_{30}O_7$ requires 465.18837.



(1*R*,4a*S*)-methyl 7-acetyl-6-hydroxy-1,4a-dimethyl-1,2,3,4,4a,9,10,10a-octahydrophenanthrene-1-carboxylate (**12**). To a solution of intermediate **11**[4] (400 mg, 1.16 mmol) in DCM (5 mL) at 0 °C, acetyl chloride (0.74 mL, 10.4 mmol) was added. To this vigorously stirred mixture, $AlCl_3$ (934 mg, 7 mmol) was added in small portions over 30 min. The reaction mixture is gradually allowed to warm to ambient temperature. After 6 h, the mixture was slowly poured into ice-cold 10% aqueous HCl solution. The mixture was extracted with Et_2O (3 × 10 mL). The combined organic phases were washed sequentially with saturated aqueous $NaHCO_3$ solution (2 × 10 mL) and brine (10 mL), dried over Na_2SO_4 and concentrated under reduced pressure. The residue thus obtained was dissolved in DCM (19 mL), $AlCl_3$ (1.25 g, 9.4 mmol) was added and the stirred mixture was heated to reflux for 2 h. Work up of the reaction as described above provided an oily residue that was purified by flash column chromatography (SiO_2, hexane/EtOAc 9:1) to produce upon solvent removal **12** as a white solid (370.0 mg, 96%). **Optical Rotation**: $[\alpha]_D^{25}$ +79 (*c* 0.40, acetone). **^{1}H-NMR** (500 MHz, $CDCl_3$) δ 11.89 (s, 1 H,), 7.38 (s, 1 H), 6.85 (s, 1 H), 3.67 (s, 3 H), 2.91−2.78 (m, 2 H), 2.56 (s, 3 H), 2.24 (d, *J* = 11.8 Hz, 1 H), 2.18 (dd, *J* = 12.5, 2.0 Hz, 1 H), 1.88-1.60 (m, 5 H), 1.55−1.37 (m, 2 H), 1.27 (s, 3 H), 1.20 (s, 3 H). **^{13}C-NMR** (125 MHz, $CDCl_3$) δ 203.9, 178.8, 160.2, 159.2, 131.0, 125.8, 117.9, 113.4, 52.0, 47.6, 44.2, 38.0, 37.6, 36.6, 28.9, 26.4, 24.5, 21.5, 18.4, 16.6. **HR-ESI**: m/z: 353.17225; [*M+Na*$^+$] for the compound $C_{20}H_{26}O_4$ require 353.17233.

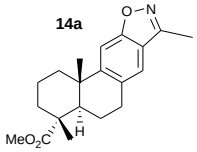


(1*R*,4a*S*,10a*R*)-methyl 6-hydroxy-7-(1-(hydroxyimino)ethyl)-1,4a-dimethyl-1,2,3,4,4a,9,10,10a-octahydrophenanthrene-1-carboxylate (**13**). A mixture of **12** (350 mg, 1.06 mmol) in MeOH/Pyr 1:5 (10.5 mL) and $NH_2OH{\cdot}HCl$ (1.5 g, 22 mmol) was heated to reflux for 1 h. It was then allowed to cool down to ambient temperature, diluted with EtOAc (20 mL) and washed with 5% aq. HCl solution, water and brine. The combined organic phases were dried over Na_2SO_4, and concentrated under reduced pressure. The residue thus obtained was purified by flash column chromatography (SiO_2, hexane/EtOAc 8:2) to produce upon solvent removal **13** as a white solid (330 mg, 93%). **Optical Rotation**: $[\alpha]_D^{25}$ +100 (*c* 0.44, acetone). **^{1}H-NMR** (500 MHz, $CDCl_3$) δ 11.10 (s, 1 H), 8.11 (s, 1 H), 7.05 (s, 1 H), 6.83 (s, 1 H), 3.69 (s, 3 H), 2.84−2.82 (m, 2 H), 2.30 (s,

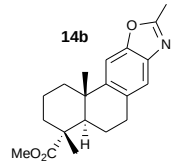
[4] R. H. Burnell and C. Côté, *Synth. Commun.*, 1988, **18**, 1753–1756.

3 H), 2.25 (d, *J* = 12.8 Hz, 1 H), 2.21 (dd, *J* = 12.5, 1.9 Hz, 1 H), 1.88−1.61 (m, 5 H), 1.57−1.46 (m, 1 H), 1.43−1.40 (m, 1 H), 1.28 (s, 3 H), 1.21 (s, 3 H). **^{13}C-NMR** (125 MHz, $CDCl_3$) *δ* 179.4, 159.1, 155.4, 152.6, 127.9, 125.5, 116.5, 112.5, 52.1, 47.7, 44.6, 37.7, 37.3, 36.7, 29.1, 24.7, 21.8, 18.5, 16.5, 10.6. **HR-ESI**: m/z: 368.18323; [*M*+*Na*$^+$] for the compound $C_{20}H_{27}NO_4$ requires 368.18271.

(4*R*,4a*R*,11b*S*)-methyl 4,8,11b-trimethyl-1,2,3,4,4a,5,6,11b-octahydrophenanthro[2,3-*d*]isoxazole-4-carboxylate (**14a**) and (4*R*,4a*R*,11b*S*)-methyl 4,9,11b-trimethyl-1,2,3,4,4a,5,6,11b-octahydrophenanthro[2,3-*d*]oxazole-4-carboxylate (**14b**). To a suspension of **13** (266 mg, 0.77 mmol) in pyridine (7.1 mL) at ambient temperature, TsCl (1.33 g, 6.98 mmol) was added in portions over 1 h. The reaction mixture was then diluted with water, followed by extractions with EtOAc (3 × 10 mL). The combined organic phases were washed with brine, dried over Na_2SO_4, and concentrated under reduced pressure. The residue thus obtained was subjected to flash column chromatography (SiO_2, hexane/EtOAc 9:1→8:2) to provide, in order of elution, **14a** (108 mg, 43%) and **14b** (101 mg, 40%) as white solids.



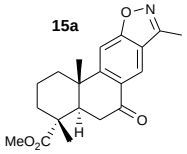
14a: Optical Rotation: $[\alpha]_D^{25}$ +63 (*c* 0.28, acetone). **^{1}H-NMR** (500 MHz, $CDCl_3$) *δ* 7.42 (s, 1 H), 7.27 (s, 1 H), 3.69 (s, 3 H), 3.11−2.97 (m, 2 H), 2.52 (s, 3 H), 2.36 (d, *J* = 11.1 Hz, 1 H), 2.25 (dd, *J* = 12.5, 2.5 Hz, 1 H), 1.94−1.85 (m, 1 H), 1.85−1.72 (m, 3 H), 1.72−1.63 (m, 1 H), 1.62−1.55 (m, 1 H), 1.53−1.45 (m, 1 H), 1.30 (s, 3 H), 1.24 (s, 3 H). **^{13}C-NMR** (125 MHz, $CDCl_3$) *δ* 178.9, 162.0, 154.4, 153.0, 130.8, 120.4, 120.3, 104.7, 52.0, 47.6, 44.5, 38.3, 38.2, 36.6, 29.6, 25.1, 21.5, 18.5, 16.6, 10.1. **HR-ESI**: m/z: 350.17255; [*M*+*Na*$^+$] for the compound $C_{20}H_{25}NO_3$ requires 350.17266.



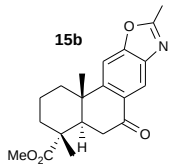
14b: Optical Rotation: $[\alpha]_D^{25}$ +87 (*c* 0.25, acetone). **^{1}H-NMR** (500 MHz, $CDCl_3$) *δ* 7.34 (s, 1 H), 7.27 (s, 1 H), 3.67 (s, 3 H), 3.02 (dd, *J* = 8,7, 4.6 Hz, 2 H), 2.59 (s, 3 H), 2.33 (d, *J* = 11.5 Hz, 1 H), 2.25 (dd, J= 12.5, 2.5 Hz, 1 H), 1.91−1.83 (m, 1 H), 1.82−1.72 (m, 3 H), 1.70−1.64 (m, 1 H), 1.61−1.52 (m, 1 H), 1.50−1.42 (m, 1 H), 1.29 (s, 3 H), 1.23 (s, 3 H). **^{13}C-NMR** (125 MHz, $CDCl_3$) *δ* 178.9, 163.9, 149.9, 147.0, 139.4, 131.2, 118.7, 105.3, 52.0, 47.6, 44.8, 38.6, 37.8, 36.7, 30.0, 25.4, 21.6, 18.6, 16.6, 14.5. **HR-ESI**: m/z: 350.17268; [*M*+*Na*$^+$] for the compound $C_{20}H_{25}NO_3$ requires 350.17266.

(4*R*,4a*R*,11b*S*)-methyl 4,8,11b-trimethyl-6-oxo-1,2,3,4,4a,5,6,11b-octahydrophenanthro[2,3-*d*]isoxazole-4-carboxylate (**15a**) and (4*R*,4a*R*,11b*S*)-methyl 4,9,11b-trimethyl-6-oxo-1,2,3,4,4a,5,6,11b-octahydrophenanthro[2,3-*d*]oxazole-4-carboxylate (**15b**)

A stirred solution of isoxazole **14a** or oxazole **14b** (121 mg, 0.370 mmol) in glacial acetic acid (2.1 mL) was cooled close to its freezing point (ca. 10 °C) and chromium trioxide (137 mg, 1.37 mmol) dissolved in 80% aqueous acetic acid (1.0 mL) was added dropwise over 60 min. Upon completion of this addition, the solution was allowed to gradually warm up to 15 ° and was kept at this temperature for 10 h. The solution was poured into ice-cold water (20 mL) and extracted with diethyl ether (3 × 10 mL). The combined organic phases were washed with saturated aqueous $NaHCO_3$ solution (10 mL) and brine (10 mL), dried over Na_2SO_4 and concentrated under reduced pressure. The residues thus obtained were subjected flash column chromatography (SiO_2, hexane/EtOAc 8:2, for **15a**; 9:1 for **15b**) to provide upon solvent removal the corresponding oxo-derivatives.



15a: yellow solid, 116 mg (92% yield). **Optical Rotation**: $[\alpha]_D^{25}$ +20 (*c* 0.29, acetone). **^{1}H-NMR** (500 MHz, $CDCl_3$) *δ* 8.40 (s, 1 H), 7.50 (s, 1 H), 3.67 (s, 3 H), 2.85−2.69 (m, 2 H), 2.59 (s, 3 H), 2.51−2.37 (m, 2 H), 1.92−1.67 (m, 5 H), 1.37 (s, 3 H), 1.30 (s, 3 H). **^{13}C-NMR** (125 MHz, $CDCl_3$) *δ* 197.2, 177.7, 165.5, 157.7, 156.0, 127.8, 122.8, 121.3, 104.4, 52.3, 46.7, 43.3, 38.4, 37.7, 37.5, 36.5, 24.1, 18.1, 16.5, 10.1. **HR-ESI**: m/z: 364.15158; [*M*+*Na*$^+$] for the compound $C_{20}H_{23}NO_4$ requires 364.15192.

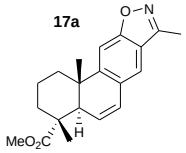


15b: white solid, 67 mg (78% yield; 68% conversion). **Optical Rotation**: $[\alpha]_D^{25}$ +38 (*c* 0.45, acetone). **^{1}H-NMR** (500 MHz, $CDCl_3$, *δ*): 8.31 (s, 1 H), 7.44 (s, 1 H), 3.66 (s, 3 H), 2.81−2.69 (m, 2 H), 2.64 (s, 3 H), 2.46−2.33 (m, 2 H), 1.88−1.58 (m, 5 H), 1.36 (s, 3 H), 1.28 (s, 3 H); **^{13}C-NMR** (125 MHz, $CDCl_3$) *δ* 197.4, 177.7, 165.3, 153.4, 140.1, 128.4, 119.3, 105.0, 52.3, 46.7, 43.7, 38.1, 37.7, 37.6, 36.5, 24.2, 18.2, 16.5, 14.6. **HR-ESI**: m/z: 364.15206; [*M*+*Na*$^+$] for the compound $C_{20}H_{23}NO_4$ requires 364.15192.

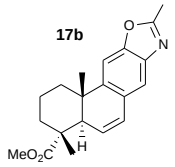
(4*R*,4a*R*,11b*S*)-methyl 4,8,11b-trimethyl-1,2,3,4,4a,11b-hexahydrophenanthro[2,3-*d*]isoxazole-4-carboxylate (**17a**) and (4*R*,4a*R*,11b*S*)-methyl 4,9,11b-trimethyl-1,2,3,4,4a,11b-hexahydrophenanthro[2,3-*d*]oxazole-4-carboxylate (**17b**). A stirred suspension of oxo derivative **15a** or **15b** (270 mg, 0.79 mmol) in methanol (4 mL) was cooled at 0 °C and sodium borohydride (77.4 mg, 2.05 mmol) was added in small portions over 30 min. The mixture was allowed to gradually warm up to ambient temperature over 30 min. After additional 15 min, ice was added and solvents were removed under reduced pressure. The residue thus obtained was diluted with EtOAc (10 mL) and washed with water (10 mL) and brine (10 mL). The combined organic phases were dried over

Na_2SO_4 and concentrated under reduced pressure. The white foam thus obtained (**16a** or **16b** respectively) was used without further purification in the next step.

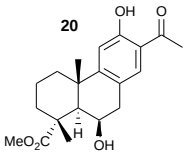
A mixture of the above crude alcohol and catalytic amount of *p*-toluene-sulfonic acid monohydrate in benzene (10 mL) was heated to reflux for 1 h. It was then allowed to cool down to ambient temperature, diluted with diethyl ether (15 mL) and was washed sequentially with saturated aqueous $NaHCO_3$ solution (15 mL) and brine (15 mL). The combined organic phases were dried over Na_2SO_4 and concentrated under reduced pressure. The oily residue thus obtained was purified by flash column chromatography (SiO_2, hexane/EtOAc 8:2) to produce upon solvent removal isoxazole derivative **17a** or oxazole derivative **17b**.



17a: white solid, 206 mg (80% yield from **15a**). **Optical Rotation**: $[\alpha]_D^{25}$ −46 (*c* 0.43, acetone). **^{1}H-NMR** (500 MHz, $CDCl_3$) *δ* 7.35 (s, 1 H), 7.26 (s, 1 H), 6.63 (dd, *J* = 9.6, 2.8 Hz, 1 H), 5.76 (dd, *J* = 9.5, 2.1 Hz, 1 H), 3.67 (s, 3 H), 2.94−2.92 (m, 1 H), 2.54 (s, 3 H), 2.31−2.22 (m, 1 H), 1.90−1.68 (m, 5 H), 1.42 (s, 3 H), 1.12 (s, 3 H). **^{13}C-NMR** (125 MHz, $CDCl_3$) *δ* 178.3, 163.2, 154.9, 151.5, 129.6, 129.4, 127.7, 120.1, 118.2, 103.5, 52.2, 46.3, 45.9, 38.5, 35.6, 29.7, 21.1, 18.3, 18.0, 10.1. **HR-ESI**: m/z: 348.15752; [*M*+*Na*$^+$] for the compound $C_{20}H_{23}NO_3$ requires 348.15701.



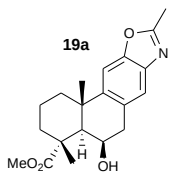
17b: white foam, 219 mg (85% yield from **15b**). **Optical Rotation**: $[\alpha]_D^{25}$ +12 (*c* 1.1, acetone). **^{1}H-NMR** (500 MHz, $CDCl_3$) *δ* 7.29 (s, 1 H), 7.27 (s, 1 H), 6.61 (dd, *J* = 9.6, 3.1 Hz, 1 H), 5.72 (dd, *J* = 9.6, 2.5 Hz, 1 H), 3.66 (s, 3 H), 2.92 (t, *J* = 2.8 Hz, 1 H), 2.60 (s, 3 H), 2.22 (d, *J* = 10.2 Hz, 1 H), 1.89−1.65 (m, 5 H), 1.41 (s, 3 H), 1.10 (s, 3 H). **^{13}C-NMR** (125 MHz, $CDCl_3$) *δ* 178.4, 163.7, 150.9, 145.7, 139.4, 129.8, 129.0, 128.2, 117.1, 104.0, 52.1, 46.4, 38.0, 35.7, 35.6, 21.2, 18.4, 17.9, 14.5. **HR-ESI**: m/z: 348.15714; [*M*+*Na*$^+$] for the compound $C_{20}H_{23}NO_3$ requires 348.15701.



(1*R*,4a*S*,10*R*,10a*R*)-methyl 7-acetyl-6,10-dihydroxy-1,4a-dimethyl-1,2,3,4,4a,9,10,10a-octahydro-phenanthrene-1-carboxylate (**20**). Derivative **17a** (56.9 mg, 175 μmol) was dissolved in a 10:1 mixture of THF and water (3.5 mL). Oxygen was removed by passing through the solution argon for 30 min, then the solution was cooled to 0 °C and NBS (49.8 mg, 280 μmol) was introduced. The mixture was allowed to stand in the dark and at 0 °C for 2 days. The reaction was quenched

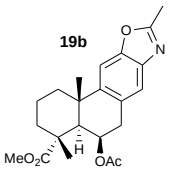
by addition of saturated aqueous solution of $Na_2S_2O_3$ (10 mL) and the mixture was extracted with EtOAc (3 × 10 mL). The combined organic phases were washed with water (2 × 10 mL) brine (10 mL), dried over Na_2SO_4 and concentrated under reduced pressure. The residue thus obtained was subjected to flash column chromatography (SiO_2, hexane/EtOAc 7:3) to provide a mixture of bromohydrins as a light yellow foam. A solution of the above bromohydrins mixture in THF (28 mL) was cooled to –78 °C and KO*t*Bu (30 mg, 260 μmol) was added in small portions. The resulting mixture was stirred at this temperature for 15 min and then it was allowed to gradually warm up to 0 °C over 1 h. The reaction mixture was diluted with ice cold water (30 mL) and extracted with EtOAc (3 × 10 mL). The combined organic phases were washed with brine (10 mL), dried over Na_2SO_4 and concentrated under reduced pressure. The residue thus obtained was used without further purification in the next step. A solution of the above crude epoxide **18a** in a mixture of methanol (33 mL) and triethanolamine (10 mL) was subjected to hydrogenolysis over 5% Pd/$BaSO_4$ at ambient temperature and pressure. After 1.5 h, the reaction mixture was filtered through Celite and the filter cake was washed several times with methanol (5 × 10 mL). The combined methanol filtrates were concentrated to approximately one sixth their original volume under reduced pressure. The concentrate was diluted with water (50 mL) and extracted with diethyl ether (3 × 10 mL). The combined organic phases were washed with brine (10 mL), dried over Na_2SO_4 and concentrated under reduced pressure. The crude product was diluted in a mixture of 9:1 MeOH/H_2O (10 mL), followed by the addition of Montmorillonite K-10 (100 mg). The reaction mixture was stirred overnight at ambient temperature and was then filtered through Celite. The filtrates were concentrated under reduced pressure and the residue thus obtained was purified by preparative TLC (SiO_2, hexane/EtOAc 8:2) to provide upon solvent removal intermediate **20** as light yellow solid (18.8 mg, 31% yield from **17a**). **Optical rotation**: $[\alpha]_D^{25}$ +18 (*c* 0.66, DCM). **^{1}H-NMR** (500 MHz, $CDCl_3$) *δ* 11.91 (s, 1 H), 7.41 (s, 1 H), 6.92 (s, 1 H), 4.21 (s, 1 H), 3.70 (s, 3 H), 3.17 (dd, *J* = 16.9, 4.9 Hz, 1 H), 2.84 (d, *J* = 16.8 Hz, 1 H), 2.59 (s, 3 H), 2.27 (s, 1 H), 2.17 (br d, *J* = 13.0 Hz, 1 H), 1.95−1.82 (m, 1 H), 1.82−1.69 (m, 2 H), 1.69−1.63 (m, 1 H), 1.62 (s, 3 H), 1.58 (s, 3 H), 1.55−1.46 (m, 2 H). **^{13}C-NMR** (125 MHz, $CDCl_3$) *δ* 203.7, 179.0, 160.5, 158.3, 131.7, 122.7, 118.3, 114.3, 68.6, 52.1, 48.4, 47.4, 40.7, 40.3, 38.1, 38.0, 26.5, 26.4, 18.8, 18.6. **HR-ESI**: m/z: 369.16682; [*M*+*Na*$^+$] for the compound $C_{20}H_{26}O_5$ requires 369.16724.

19a



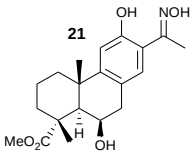
(4*R*,4a*R*,5*R*,11b*S*)-methyl 5-hydroxy-4,9,11b-trimethyl-1,2,3,4,4a,5,6,11b-octahydrophenanthro-[2,3-*d*]oxazole-4-carboxylate (**19a**). Derivative **17b** (29.3 mg, 90.0 μmol) was dissolved in a 10:1 mixture of THF and water (3 mL). Oxygen was removed by passing through the solution argon for

30 min, then the solution was cooled to 0 °C and NBS (24.9 mg, 140 μmol) was introduced. The mixture was allowed to stand in the dark and at 0 °C for 2 days. The reaction was quenched by addition of saturated aqueous solution of $Na_2S_2O_3$ (10 mL) and the mixture was extracted with EtOAc (3 × 10 mL). The combined organic phases were washed with water (2 × 10 mL) brine (10 mL), dried over Na_2SO_4 and concentrated under reduced pressure. The residue thus obtained was subjected to flash column chromatography (SiO_2, hexane/EtOAc 6:4) to provide a mixture of bromohydrins as a light yellow foam. A solution of the above bromohydrins mixture in THF (30 mL) was cooled to –78 °C and KO*t*Bu (15 mg, 135 μmol) was added in small portions. The resulting mixture was stirred at this temperature for 15 min and then it was allowed to gradually warm up to 0 °C over 1 h. The reaction mixture was diluted with ice cold water (30 mL) and extracted with EtOAc (3 × 10 mL). The combined organic phases were washed with brine (10 mL), dried over Na_2SO_4 and concentrated under reduced pressure. The residue thus obtained was used without further purification in the next step. A solution of the above crude epoxide **18b** in a mixture of methanol (7.5 mL) and triethanolamine (3.3 mL) was subjected to hydrogenolysis over 5% Pd/$BaSO_4$ at ambient temperature and pressure. After 2 h, the reaction mixture was filtered through Celite and the filter cake was washed several times with methanol (3 × 10 mL). The combined methanolic filtrates were concentrated to approximately one sixth their original volume under reduced pressure. The concentrate was diluted with water (25 mL) and extracted with diethyl ether (3 × 10 mL). The combined organic phases were washed with brine (10 mL), dried over Na_2SO_4 and concentrated under reduced pressure. The residue thus obtained was purified by flash column chromatography (SiO_2, hexane/EtOAc 6:4) to provide upon solvent removal alcohol **19a** as white solid (23.2 mg, 75 % from **17b**). **Optical rotation**: $[\alpha]_D^{25}$ +34 (*c* 0.54, DCM). **^{1}H-NMR** (500 MHz, $CDCl_3$) *δ* 7.40 (s, 1 H), 7.29 (s, 1 H), 4.22 (br s, 1 H), 3.70 (s, 3 H), 3.34 (dd, *J* = 17.1, 5.1 Hz, 1 H), 2.99 (d, *J* = 17.0 Hz, 1 H), 2.59 (s, 3 H), 2.32 (s, 1 H), 2.24 (br d, *J* = 12.6 Hz, 1 H), 1.98−1.85 (m, 1 H), 1.82 (dd, *J* = 12.9, 3.6 Hz, 1 H), 1.80−1.71 (m, 2 H), 1.64 (s, 3 H), 1.61 (s, 3 H), 1.60−1.53 (m, 2 H). **^{13}C-NMR** (125 MHz, $CDCl_3$) *δ* 179.2, 164.0, 150.3, 146.0, 139.7, 128.0, 119.3, 106.1, 68.7, 52.1, 48.4, 47.9, 41.7, 41.4, 38.1, 38.0, 27.0, 19.0, 18.5, 14.6. **HR-ESI**: m/z: 366.16762; [*M*+*Na*$^+$] for the compound $C_{20}H_{25}NO_4$ requires 366.16758.

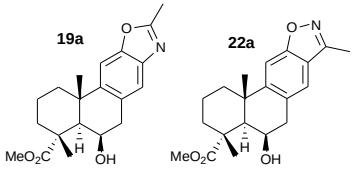


(4*R*,4a*R*,5*R*,11b*S*)-methyl 5-acetoxy-4,9,11b-trimethyl-1,2,3,4,4a,5,6,11b-octahydrophenanthro-[2,3-*d*]oxazole-4-carboxylate (**19b**). A solution of alcohol **19a** (20 mg, 58 μmol) in dichloromethane (1.5 mL) was cooled at 0 °C. 4-(Dimethylamino)pyridine (58 mg, 0.47 mmol) and acetic anhydride (45 μL, 0.47 mmol) were added sequentially and the mixture was stirred at 0 °C for 5 h. Water

(5 mL) was added and the mixture was extracted with diethyl ether (3 × 5 mL). The combined organic phases were washed with brine (5 mL), dried over Na_2SO_4 and concentrated under reduced pressure. The residue thus obtained was purified by flash column chromatography (SiO_2, hexane/EtOAc 6:4) to provide upon solvent removal acetate **19b** as colorless oil (18.3 mg, 82%). **Optical rotation**: $[\alpha]_D^{25}$ −34 (*c* 0.46, acetone). **^{1}H-NMR** (500 MHz, $CDCl_3$) *δ* 7.42 (s, 1 H), 7.27 (s, 1 H), 5.21 (dt, *J* = 3.6, 1.7 Hz, 1 H), 3.71 (s, 3 H), 3.34 (dd, *J* = 17.9, 5.5 Hz, 1 H), 3.09 (d, *J* = 17.8 Hz, 1 H), 2.60 (s, 3 H), 2.50 (s, 1 H), 2.29 (br d, *J* = 12.6 Hz, 1 H), 2.00 (s, 3 H), 1.94−1.86 (m, 1 H), 1.81−1.73 (m, 2 H), 1.71−1.54 (m, 2 H), 1.60 (s, 3 H), 1.45 (s, 3 H). **^{13}C-NMR** (125 MHz, $CDCl_3$) *δ* 178.5, 170.5, 164.2, 150.3, 145.2, 139.8, 127.5, 119.1, 106.1, 70.6, 52.3, 48.0, 46.6, 41.6, 38.4, 38.2, 37.2, 27.4, 21.6, 18.8, 18.1, 14.6. **HR-ESI**: m/z: 408.17836; [*M*+*Na*$^+$] for the compound $C_{22}H_{27}NO_5$ requires 408.17814.



(1*R*,4a*S*,10*R*)-methyl 6,10-dihydroxy-7-(1-(hydroxyimino)ethyl)-1,4a-dimethyl-1,2,3,4,4a,9,10,10a-octahydrophenanthrene-1-carboxylate (**21**). In analogy to the preparation of oxime **13** from ketone **12**, oxime **21** was prepared from ketone **20**. The desired product (**21**; 25 mg, 80%) was obtained as white solid after purification by flash column chromatography (SiO_2, hexane/EtOAc 7:3). **Optical Rotation**: $[\alpha]_D^{25}$ +24 (*c* 0.13, acetone). **^{1}H-NMR** (500 MHz, $CDCl_3$) *δ* 11.03 (br s, 1 H), 7.96 (br s, 1 H), 7.06 (s, 1 H), 6.89 (s, 1 H), 4.18 (d, *J* = 4.5 Hz, 1 H), 3.70 (s, 3 H), 3.16 (dd, *J* = 17.0, 5.0 Hz, 1 H), 2.82 (d, *J* = 16.8 Hz, 1 H), 2.29 (s, 3 H), 2.28 (s, 1 H), 2.17 (br d, *J* = 12.6 Hz, 1 H), 2.05 (s, 1 H), 1.93−1.83 (m, 1 H), 1.82−1.75 (m, 1 H), 1.74−1.68 (m, 1 H), 1.67−1.62 (m, 1 H), 1.62 (s, 3 H), 1.58 (s, 3 H), 1.51 (td, *J* = 12.8, 3.1 Hz, 1 H). **^{13}C-NMR** (125 MHz, $CDCl_3$) *δ* 179.4, 159.1, 155.8, 151.5, 128.6, 122.2, 117.0, 113.2, 68.9, 52.2, 48.5, 47.7, 40.8, 40.3, 38.0, 37.4, 26.5, 18.9, 18.5, 10.7. **HR-ESI**: m/z: 384.17790; [*M*+*Na*$^+$] for the compound $C_{20}H_{27}NO_5$ requires 384.17814.

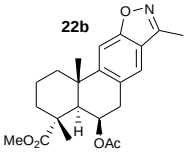


(4*R*,4a*R*,5*R*,11b*S*)-methyl 5-hydroxy-4,9,11b-trimethyl-1,2,3,4,4a,5,6,11b-octahydrophenanthro-[2,3-*d*]oxazole-4-carboxylate (**19a**) and (4*R*,4a*R*,5*R*,11b*S*)-methyl 5-hydroxy-4,8,11b-trimethyl-1,2,3,4,4a,5,6,11b-octahydrophenanthro[2,3-*d*]isoxazole-4-carboxylate (**22a**). In analogy to **14a** and **14b**, **22a** (6.8 mg, 30%; light yellow foam) and **19a** (5.5 mg, 24%; white solid) were obtained from **21** (24 mg, 66 µmol).

22a: Optical rotation: $[\alpha]_D^{25}$ +30 (*c* 0.33, DCM). **^{1}H-NMR** (500 MHz, $CDCl_3$) *δ* 7.49 (s, 1 H), 7.31 (s, 1 H), 4.28−4.23 (m, 1 H) 3.71 (s, 3 H), 3.35 (dd, *J* = 17.0, 5.2 Hz, 1 H), 3.04 (dd, *J* = 16.9, 1.8

Hz, 1 H), 2.53 (s, 3 H), 2.33−2.26 (m, 1 H), 2.32 (s, 1 H), 1.98−1.87 (m, 1 H), 1.86−1.73 (m, 2 H), 1.72−1.53 (m, 3 H), 1.65 (s, 3 H), 1.62 (s, 3 H). **^{13}C-NMR** (125 MHz, $CDCl_3$) *δ*; 179.1, 162.3, 154.4, 152.0, 127.8, 121.0, 120.6, 105.6, 68.6, 52.2, 48.4, 47.8, 41.4, 41.1, 38.5, 38.1, 27.0, 18.9, 18.7, 10.1. **HR-ESI**: m/z: 366.16740; [*M*+*Na*$^+$] for the compound $C_{20}H_{25}NO_4$ requires 366.16758.

22b



(4*R*,4a*R*,5*R*,11b*S*)-methyl 5-acetoxy-4,8,11b-trimethyl-1,2,3,4,4a,5,6,11b-octahydrophenanthro-[2,3-*d*]isoxazole-4-carboxylate (**22b**). In analogy to **19b**, **22b** (4.5 mg, 81%; light yellow foam) was obtained from **22a**. **Optical rotation**: $[\alpha]_D^{25}$ −24 (*c* 0.29, acetone). **^{1}H-NMR** (500 MHz, $CDCl_3$) *δ* 7.51 (s, 1 H), 7.28 (s, 1 H), 5.26−5.22 (m, 1 H) 3.71 (s, 3 H), 3.35 (dd, *J* = 17.7, 5.4 Hz, 1 H), 3.14 (d, *J* = 17.5 Hz, 1 H), 2.53 (s, 3 H), 2.52 (s, 1 H), 2.38−2.31 (m, 1 H), 2.01 (s, 3 H), 1.98−1.87 (m, 1 H), 1.82−1.73 (m, 2 H), 1.73−1.64 (m, 2 H), 1.61 (s, 3 H), 1.47 (s, 3 H). **^{13}C-NMR** (125 MHz, $CDCl_3$) *δ* 178.4, 170.5, 162.3, 154.4, 151.3, 127.2, 121.0, 120.8, 105.6, 70.5, 52.4, 48.0, 46.5, 41.4, 38.6, 38.4, 37.0, 27.4, 21.6, 18.8, 18.2, 10.1. **HR-ESI**: m/z: 408.17810; [*M*+*Na*$^+$] for the compound $C_{22}H_{27}NO_5$ requires 408.17814.

2. Biological assays

2.1. Gli-dependent luciferase assay

The biological investigation of the taepeenin D derivatives was accomplished using a Luciferase reporter assay. For that reason, Shh-LIGHTII cells that stably incorporate a Gli-dependent firefly luciferase reporter as well as a constitutive *Renilla* luciferase reporter were used. As positive and negative controls of the assay SAG (Smo agonist) and cyclopamine were correspondingly used.
Shh-LIGHTII cells (ATCC®, CRL-2795TM, LGC, Wesel, Germany), a clonal mouse fibroblast cell line derived from NIH 3T3 which are transfected with a Gli-dependent firefly luciferase reporter and a constitutive *Renilla* luciferase expression vector, were grown in 75 cm^2 cell culture flasks (TPP, Trasadingen, Switzerland) at 37 °C in a humid atmosphere with 5% CO_2.

Cell growth medium DMEM (Dulbecco's Modified Eagle's Medium, high glucose, sodium pyruvate, w/o glutamine) was supplemented with Zeocin™ Selection Reagent (0.15 mg/mL) and Geneticin® Selection Antibiotic (G418 sulfate, 0.4 mg/mL) which were obtained from Invitrogen, L-glutamine (4 mM) from PAA and FBS (fetal bovine serum, 10 %) from Sigma.

Assay medium consisted of DMEM with an additional amount of L-glutamine (4 mM), FBS (0.5 %) and HEPES (50 mM, pH 7.4). The latter was from Fluka.

Cell freeze medium contained 5 % DMSO from Fisher Scientific in complete growth medium.

Cell Culture: Before performing the assay, Shh-LIGHTII cells were firstly grown to reach 80% confluency. The cell culture was being checked under an optical microscope. Afterwards, the cells were washed with Versene (4 mL), detached with trypsin (1 mL) during 5 min and resuspended in growth medium (9 mL) by gently pipetting. Then, they were centrifuged at 3,000 rpm for 5 min, the supernatant was removed and the cell pellet dissolved in 4 mL growth medium. The amount of cells was counted and 50,000 cells were seeded per well in a 24-well plate and incubated for 48 hours.

To expose the cells to the candidate inhibitors, the growth medium was removed and assay medium containing several concentrations of the compounds was added to each well (500 µL/well). Therefore, stock solutions of the taepeenin D derivatives were prepared in DMSO and added to the assay medium to achieve the desired final concentrations. Furthermore, Shh-LIGHTII cells were co-exposed to 100 nM (and in case of **19a** and **5** also to 10 nM) of the Hh agonist SAG in order to activate the corresponding pathway. A SAG stock solution was prepared in DMSO and introduced into the medium. The final DMSO concentration (introduced by SAG and the test compound) was less than 0.1 %, thus not endangering the cell viability. Finally, a 100 nM solution of SAG was used as positive control for the experiment and an 8 µM solution of cyclopamine was utilized as negative control. Thereafter, the cells were incubated for 48 h.

After 48 h, the medium was removed and treated cells were washed with PBS (500 µL) and subsequently incubated with 1x passive lysis buffer (100 µL) for 15 min on a Belly Dancer Shaker. The suspensions of the lysed cells of each well were transferred into 1.5 mL reaction tubes, centrifuged at 14,000 rpm for 1 min at 4 °C and kept on ice until further use. The blank of luminescence of a 96-well plate was measured using the Orion Microplate Luminometer. Thence, luciferase assay reagent (LAR, 100 µL) and cell supernatant (20 µL) were carefully mixed per well by gently pipetting and the luminescence of firefly luciferase was recorded. Afterwards, Stop&Glo® reagent (100 µL) was added to each well and the *Renilla* luminescence was measured. The measurement was performed per row of the 96-well plate and the total duration between both the addition of LAR and Stop&Glo® reagent and the final measurement was no longer than 10 min.

Data Analysis: To determine the IC_{50} values, the quotient of firefly and *Renilla* luminescence (RLU/s) were plotted against the concentrations of the active compounds on a logarithmic (log-10) scale. Concentration-response curves of Gli-reporter gene luminescence were fitted with the Hill slope equation and IC_{50} values were calculated from modelled concentration-response curves. At first, all data were normalized so that individual dose-response curves converged to 0 and 100 and then they were used to calculate the IC_{50} value, corresponding SE (standard errors) and confidence intervals (2−3 replicates). Calculations were performed using the software OriginPro 8 (OriginLab Corporation). IC_{50} values with non-overlapping confidence intervals ($p<0.05$) were considered as statistically significantly different.

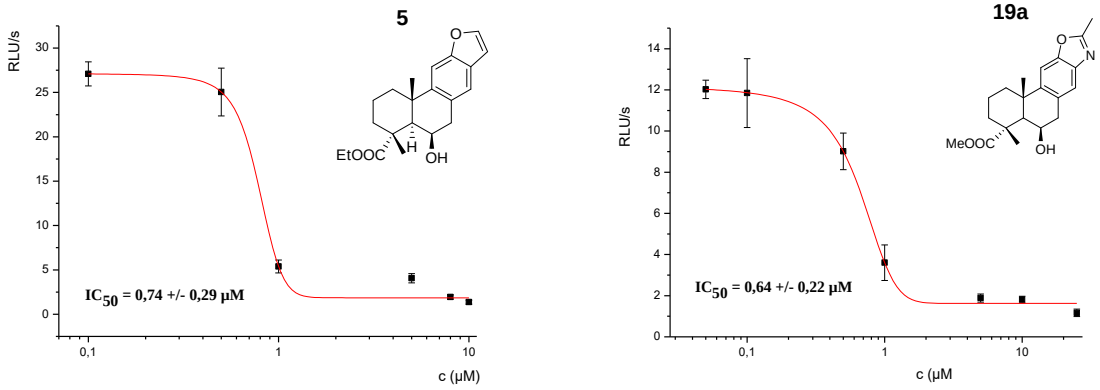


Figure S1. Determination of IC_{50} values for Gli-luc inhibition by compounds **5** and **19a**.

2.2. Colorimetric cytotoxicity assay

Cytotoxicity was measured in MCF-7 and HUVE cells employing the Cell Proliferation Reagent WST-1.

HUVE cells (PAN Biotech GmbH, HUVEC-cryo, Cat. No. P09-1000-1), a normal human umbilical vein endothelial cell line, were cultured in 25 cm^2 flasks at 37 °C in a humid atmosphere with 5% CO_2. Endothelial cell growth medium (C-22010), DetachKit (C-41200) and Cryo-SF medium (C-29910) were obtained from PromoCell.

The epithelial breast cancer cell line MCF-7 (HTB-22), were obtained from LGC Standards and were grown under standard conditions in 75 cm² flasks at 37 °C in a humid atmosphere with 5 % CO_2. Cell growth medium DMEM (Dulbecco's Modified Eagle's Medium, high glucose, sodium pyruvate, w/o glutamine) was obtained from Invitrogen and supplemented with FBS (fetal bovine serum, 10 %) and Insulin solution human from Sigma. Trypsin was obtained from PAA and Versene™ chelating agent from Gibco.

96-well plates (Item No. 655180) were purchased from Greiner Bio-One. The cell number was counted using a Hemocytometer (Neubauer-Zählkammer). For the centrifugation of the 15 mL falcon tubes (Corning Inc., 352095) and 1.5 mL reaction tubes (Eppendorf) a Heraeus™ Sepatech Labofuge™ 200 Centrifuge and a Beckman Avanti™ 30 Centrifuge were used respectively. Both compounds were dissolved in DMSO (Fluka, CAS No. 67-68-5).

Cell toxicity was measured using the cell proliferation reagent WST-1 (Roche, Product No. 05015944001). The assay was performed according to the assay kit instructions.

For performing the assay, 30000 MCF-7 and 8000 HUVE cells were seeded per well of a 96 well microplate with a total volume of 100 µL. The medium was removed after 24 h and the cells were incubated for additional 24 h with 100 µL of different concentrations of the compounds (1-100 µM; each concentration was checked three times). The solvent concentration was always lower than 0,5 % (v/v).

Subsequently, the WST-1 Reagent was added to each well and 2 h and 4 h later (concerning MCF-7 and HUVE respectively), the absorbance was measured using Optimax UV-VIS spectrometer (measured wavelengths: 440 nm and 650 nm).

The differences of the absorption at 440 nm and 650 nm were plotted against the inhibitor concentration.

No cytotoxicity was observed for either **5** or **19a** at concentrations up to 100 µM in either MCF-7 (data shown in Figure S2) or HUVE cells (data shown in Figure S3).

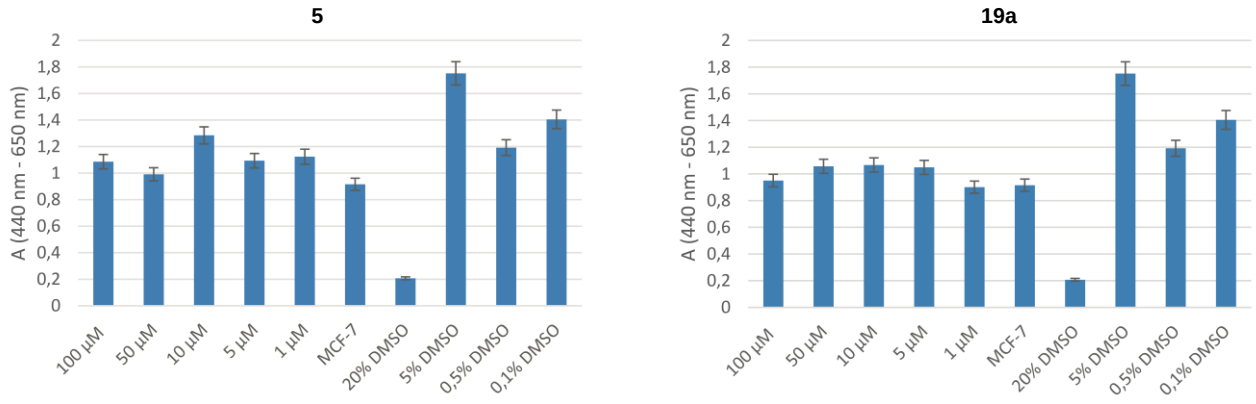


Figure S2. Effect of compounds **5** and **19a** on MCF-7 cells viability.

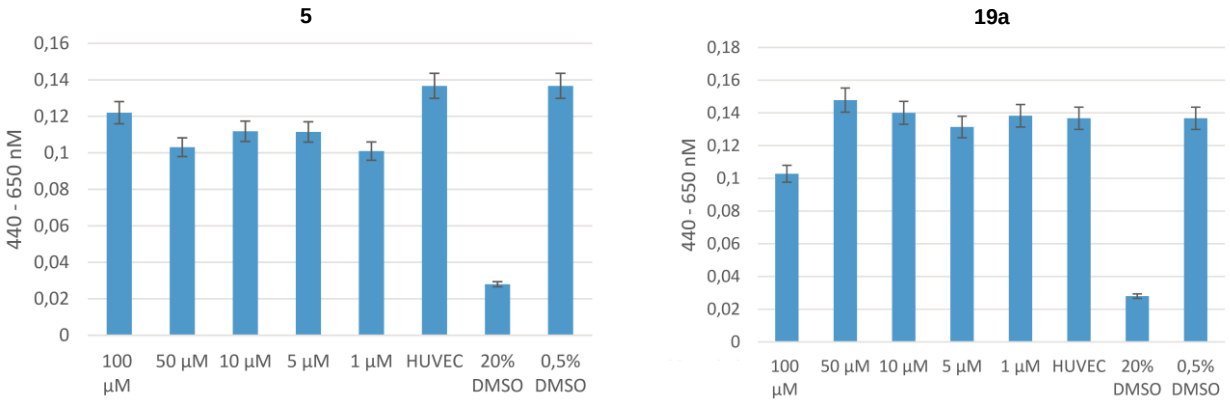


Figure S3. Effect of compounds **5** and **19a** on HUVE cells viability.