Highly Potent, Synthetically Accessible Prostratin Analogs Induce Latent HIV Expression *in vitro* and *ex vivo*

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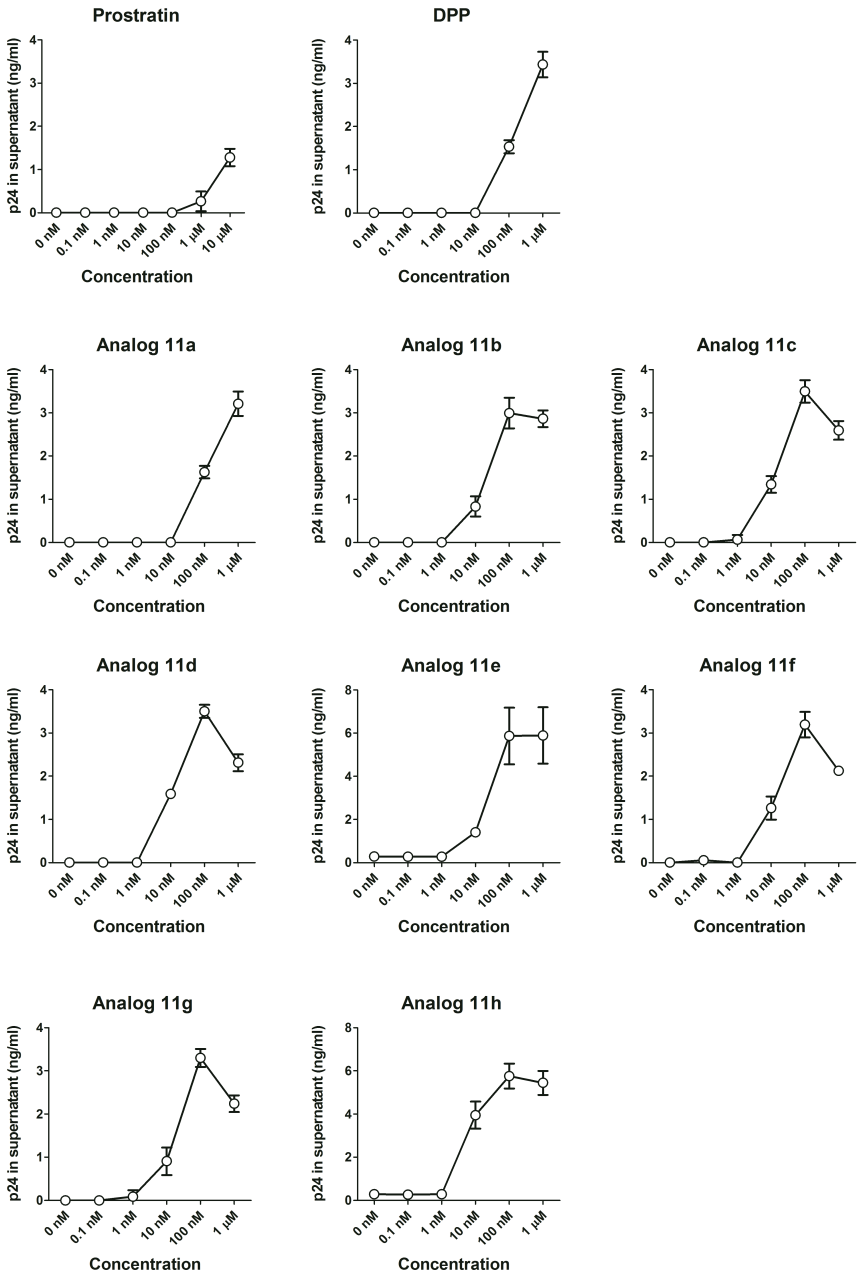


Figure S1: Curves showing HIV latency activation (p24 production) the U1 cell line after dosing with Prostratin, DPP or Analogs **11a-11f**.

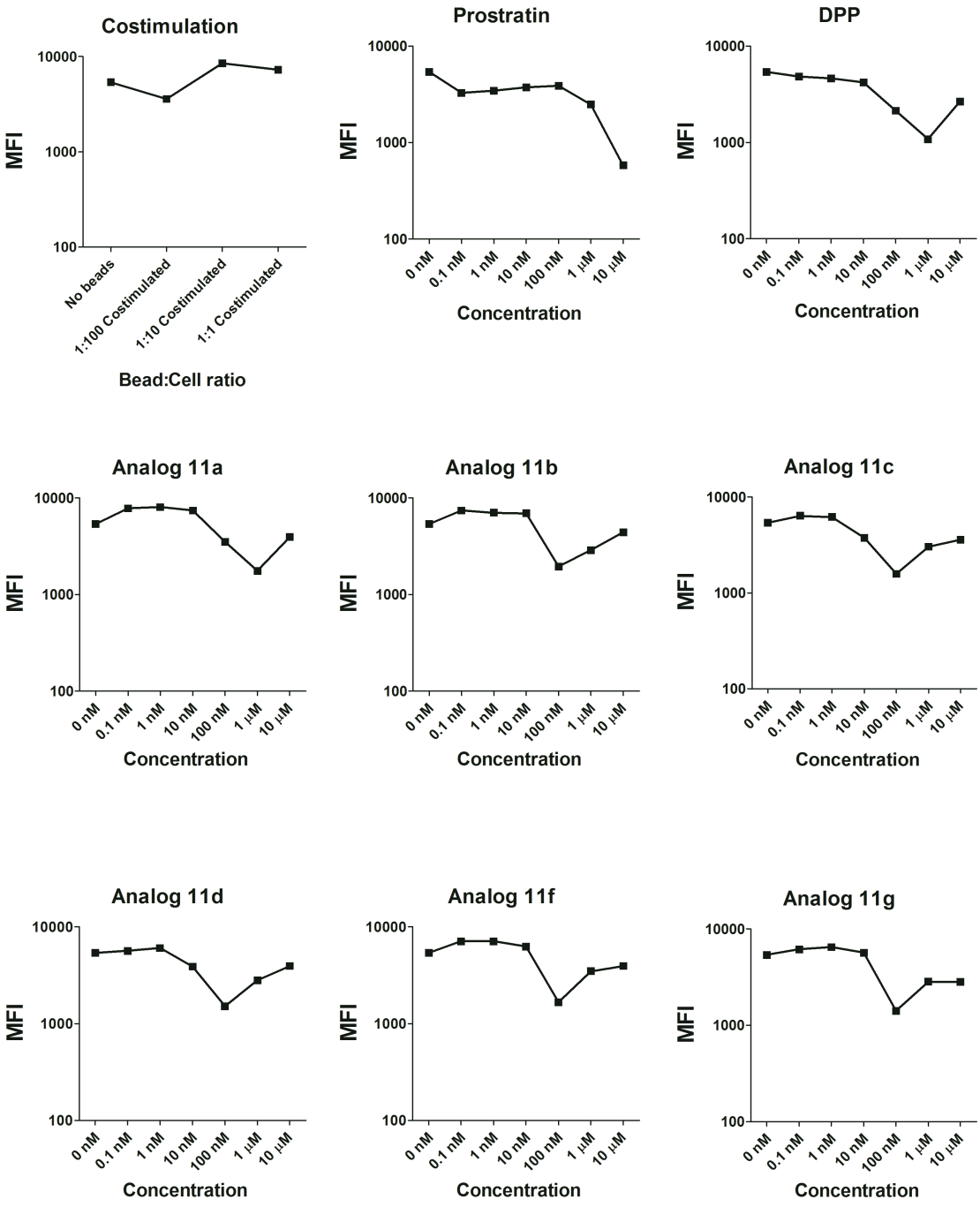


Figure S2: Treatment of resting CD4+ T cells isolated from HIV negative individuals with costimulation (via bead-based antibody ligation of CD3 and CD28), Prostratin, DPP, or analogs**.** CD4 mean fluorescence intensity (MFI) following 24h of treatment is shown.

General Synthetic Methods and Apparatus

Unless otherwise noted, all reactions were run with flame or oven dried glassware cooled under a dry nitrogen atmosphere prior to use. Reactions were stirred using Teflon-coated magnetic stirrer bars. Reactions were monitored using thin layer silica gel chromatography (TLC) using 0.25 mm silica gel 60F plates with fluorescent indicator from Merck. Plates were visualized by treatment with UV or acidic *p*-anisaldehyde stain with gentle heating. Products were purified via column chromatography using the solvent systems indicated. Silica gel 60, 230-400 mesh, was purchased from EM. Alternatively, a Teledyne-ISCO Combiflash Rf 75 system with pre-packed silica columns was used.

Unless otherwise noted, all commercial reagents were purchased from Sigma-Aldrich, Fischer Scientific, or VWR and used without additional purification. Tetrahydrofuran (THF) was distilled from sodium benzophenone ketyl under N_2. Dichloromethane (DCM) was passed through an alumina drying column (*Solv-Tek Inc.*) using nitrogen pressure. Ethyl acetate, petroleum ether, pentane, and methanol (MeOH) were obtained from Fisher Scientific. DMSO used in bioassays and used to prepare biological samples was obtained from Fisher BioReagents (Class III). Amine bases (Et_3N, pyridine, N,N-diisopropylethylamine) were distilled from CaH_2 under nitrogen. ^{3}H-phorbol dibutyrate (^{3}H-PDBu) was obtained from American Radiolabeled Chemicals, Inc. (St. Louis, MO) as a solution in acetone with a specific activity of 20 μCi/mmol. For noted compounds, samples were purified via preparative reverse-phase HPLC in a water/acetonitrile (MeCN) gradient using a Varian Pro-star(model 320) system equipped with an AllTech Alltima C18 column (10 μm, 10 x 250 mm).

NMR spectra were measured on a Varian INOVA 500 (^{1}H at 500 MHz, ^{13}C at 125 MHz), a Varian 400 (^{1}H at 400 MHz, ^{13}C at 100 MHz), or a Varian INOVA 600 MHz (^{1}H at 600 MHz, ^{13}C at 150 MHz) magnetic resonance spectrometer, as noted. ^{1}H chemical shifts are reported relative to the residual solvent peak (chloroform = 7.26 ppm) as follows: chemical shift (δ), (multiplicity (s = singlet, bs = broad singlet, d = doublet, bd = broad doublet, t = triplet, q = quartet, p = pentet, m = multiplet), integration, coupling constant(s) in Hz, proton ID [when available, designated by carbon number]). ^{13}C chemical shifts are reported relative to the residual deuterated solvent ^{13}C signals ($CDCl_3$ = 77 ppm). Infrared spectra were recorded on a Perkin-Elmer 1600 Series Fourier transform spectrometer (FTIR) and are reported in wavenumbers (cm^{-1}). Optical rotation data were obtained using a JASCO DIP are reported as $[\alpha]_D$ (c = grams/100 mL). Optical rotations are the average (± standard deviation) of 10 individual measurements. High resolution mass spectra were obtained at the Vincent Coates Mass Spectrometry Laboratory, Stanford, CA, 94305.

PKC Binding Assay Protocol

A. Preparation of Binding Assay Buffer

To a polypropylene vial was added Tris-HCl (pH 7.4, 1 M, 1 mL), KCl (1 M, 2 mL), $CaCl_2$ (0.1 M, 30 μL), and bovine serum albumin (40 mg, from Sigma). This mixture was diluted to 20 mL with de-ionized water. The buffer was stored on ice until use.

B. Preparation of Phosphatidylserine (PS) Vesicles

For each compound to be tested, 1.75 mg phosphatidylserine (Avanti Polar Lipids, porcine, obtained as a solution in $CHCl_3$) was isolated by removing chloroform. The solid PS was suspended as vesicles in binding assay buffer (1.75 mL) by sonicating (Branson Sonifier 250, power = 6, 40% duty cycle) four times for 30 s with a 30 s rest between sonications. The resulting cloudy mixture was stored on ice until use.

C. Preparation of PKC Mixture

Assay Protein Kinase C (PKC) was prepared by dissolving an aliquot of recombinant human PKCd (purchased from Invitrogen), in buffer solution (5.8 mL final volume for each compound to be tested).

D. Preparation of Compound and ^{3}H-PDBu Dilutions

^{3}H-PDBu (specific activity: 20 μCi/mmol) was diluted 10-fold in DMSO from a 1 mCi/mL commercial acetone solution (American Radiolabeled Chemicals, Inc.). This 500 nM stock solution was further diluted in DMSO to 30 nM for use in assays. The 500 nM and 30 nM stock solutions were stored in frozen DMSO at −20 ºC until use. Compound dilutions were also prepared in DMSO, serially diluting from a high concentration to the lowest concentration by factors of three. Thus, seven analog concentrations were used to define the inhibition curve.

E. Assay protocol

Triplicate data points were obtained for each analog concentration. For each data point, phosphatidylserine vesicles (60 μL of 1 mg/mL), diluted PKC (200 μL), and diluted test compound (20 μL) were added to a polypropylene vial. ^{3}H-PDBu (30 nM in DMSO, 20 μL) was then added to all vials. Non-specific ^{3}H-PDBu binding was assessed in triplicate by substitution of test compound with unlabeled PDBu (20 μL of a 75 μM stock, assay concentration: 5 μM). Maximal ^{3}H-PDBu binding was analyzed in triplicate by substitution of test compound with 20 μL DMSO. The solutions were mixed via vortexer, incubated at 37 °C for 10 min, and incubated on ice for at least 15 min prior to filtration.

Glass-fiber filters (Whatman GF/B, 21 mm) were prepared by soaking in a solution of aqueous poly(ethyleneimine) (10% by volume, 6 mL) diluted in water (200 mL) for ≥15 min. Rinsing buffer (500 mL, 20 mM Tris, pH 7.4) was cooled on ice for the duration of the incubation period and for the remainder of the assay.

Assay vial contents were vacuum-filtered through the poly(ethylenimine)-soaked filters, washing residual vial contents with 0.5 mL ice-cold rinse buffer. The filters were then washed dropwise with ice-cold buffer (4.5

mL), allowed to partially dry (~15 s) on the filter apparatus, and placed into scintillation vials. Scintillation vials were filled with Bio-Safe scintillation fluid (5 mL) and were measured for radioactivity using a Beckman LS 6000SC scintillation counter. Counts per minute (cpm) were averaged for each triplicate dilution. The data was then plotted (cpm vs. log(concentration)) using Prism® by GraphPad Software and an IC_{50} was determined using the built-in one-site competition least squares regression function. K_i values were calculated by the equation: $K_i = IC_{50}/(1+ [^3H\text{-PDBu}])/K_d$ of ^3H-PDBu). The K_d of ^3H-PDBu was measured via saturation binding under identical conditions to PKC-δ and was found to be 10.0 nM.

U1 Cell Stimulations Protocol

U1 cells[1] were cultured in RF10 media, which consisted of RPMI medium 1640 (Invitrogen) containing 10% fetal bovine serum (Omega Scientific), 100 units/ml of penicillin, and 100 μg/ml of streptomycin (Pen/Strep; Invitrogen). U1 stimulations were performed by seeding 20,000 cells per well in v-bottomed 96-well plates in a volume of 200 μl per well of RF10 media containing the relevant concentration of compound. The cultures were incubated for 2 days and then cell-free supernatant was removed and diluted with 2% Triton X-100 in phosphate buffered saline (PBS). Samples were stored at 4 ^oC before analysis. HIV p24 (capsid) protein concentrations in the supernatants were then quantified using the HIV p24 antigen enzyme-linked immunosorbent assay (ELISA) kit (Beckman Coulter) according to the manufacturer's instructions. The GraphPad Prism software package (version 5) was used to perform nonlinear regression analysis and calculate EC_{50} values.

Surface Receptor Expression Analysis in primary CD4+ T Cells

A. Assay Protocol

Peripheral blood mononuclear cells from HIV seronegative donors that had been isolated using Ficoll-Paque Plus separation (GE Healthcare) were obtained from the UCLA Virology Core. Resting CD4+ T cells were then separated in a two-step process. CD4+ T cells were first isolated by negative selection using the CD4+ T Cell Isolation Kit II (Miltenyi Biotec). The resting CD4+ T cell subset was then isolated by depletion of cells expressing CD25, CD69, and HLA-DR using the CD69 Microbead Kit (Miltenyi Biotec) according to the manufacturer's instructions, with an addition of CD25 Microbeads (Miltenyi Biotec) and Anti-HLA-DR Microbeads (Miltenyi Biotec) during the second incubation period. Cells were then placed in v-bottomed 96-well plates at a density of 100,000 cells per well in a 200 μl volume of RF10 media containing compound. For positive controls, 20 units/ml of interleukin 2 was added along with Dynabeads Human T-Activator CD3/CD28 (Invitrogen) at the indicated bead-to-cell ratios. Cells were incubated with compound or under relevant control conditions for 2 days before harvesting for analysis using flow cytometry.

B. Flow Cytometry

Cells were stained by first suspending them in 50 µl of a 1:1 mix of PBS and Human AB serum (Sigma). A combination of CD25-fluorescein isothiocyanate (Beckman Coulter), CD4-phycoerythrin (Beckman Coulter), and CD69-peridinin chlorophyll protein (Beckman Coulter) was then added, and the mixture was incubated at 4 ^{o}C for 15 minutes. The cells were then washed, resuspended in PBS containing 2% paraformaldehyde, and stored at 4 ^{o}C. Fixed cells were analyzed using a FC 500 flow cytometer (Beckman Coulter). The resultant list mode files were processed using FlowJo software (version 7.6). EC_{50} values for CD69 expression were calculated via nonlinear regression analysis using the GraphPad Prism software package.

HIV induction in CD4+ T cells from infected individuals receiving HAART

A. Clinical specimens

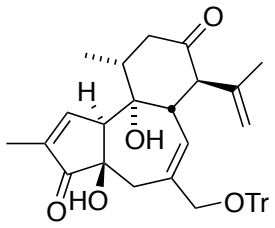
Six HIV-infected individuals receiving ART were included in this study. All individuals received various antiretroviral regimens and maintained undetectable levels of plasma viremia (<50 copies/mL) at the time of study. Leukapheresis products were obtained from the study participants in accordance with protocols approved by the Institutional Review Board of the National Institute of Allergy and Infectious Diseases. All participants signed an informed consent form.

B. Isolation and cultivation of resting $CD4^+$ T cells

Peripheral blood mononuclear cells (PBMCs) were obtained by leukapheresis and ficoll-hypaque centrifugation. $CD4^+$ T cells were isolated using a cell separation system (StemCell Technologies). Resting $CD4^+$ T cells were isolated by depleting $CD25^+$, HLA-DR^+ and $CD69^+$ $CD4^+$ T cells using PE-conjugated antibodies (BD Biosciences) and anti-PE microbeads (Miltenyi Biotec). Cells were cultured in RPMI 1640-based medium containing antiretroviral drugs consisting of T-20 (100µg/ml), tenofovir (1µM), and emtricitabine (1µM) in the absence or presence of the study compounds.

C. Measurements of virion-associated HIV RNA

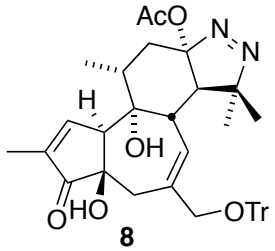
The copy number of viron-associated HIV RNA in the cell culture supernatants was determined using the Cobas Ampliprep/Cobas Taqman HIV-1 Test, Version 2.0 (Roche Diagnostics) following 48 hours of incubation of cells with the study compounds. The limit of detection for this system is 20 copies/ml.



7

To a 500 mL round-bottom flask containing a stirbar and **6** (2.00 g, 5.77 mmol, 1.0 eq.) at room temperature (23 °C) was added pyridine (58.0 mL, 0.10 M). After dissolution of **6**, trityl chloride (4.80 g, 17.3 mmol, 3.0 eq) was added. The mixture was heated to 50 °C. After 12 h, the mixture was cooled to room temperature and then diluted with EtOAc (200 mL). The resulting mixture was washed with water (50 mL), a saturated aqueous $NaHCO_3$ solution (50 mL) and brine (30 mL). The organic phase was dried over Na_2SO_4, filtered and concentrated under reduced pressure. The resulting residue was purified by flash chromatography (20%, 25%, 30%, 35%, 40% EtOAc/hexanes) to afford **7** (5.44 g, 94%) as a white solid. A reaction on a smaller scale (277 mg of product) proceeded in 96% yield.

Characterization data for **7**
^{1}H NMR ($CDCl_3$, 500 MHz): δ 7.52 (bs, 1H, C1-H), 7.41-7.38 (m, 6H, Ar-H), 7.30-7.22 (m, 9H, Ar-H), 5.69-5.67 (m, 1H, C7-H), 5.12 and 4.91 (2 x bs, C16-H), 3.67 (dd, J = 13.2 Hz, 5.7 Hz, 1H, C8-H), 3.60 (d, J = 13.3 Hz, 1H, C20-H), 3.46-3.43 (m, 2H, C14-H, C20-H), 2.90 (bs, 1H, C10-H), 2.69-2.63 (m, 1H, C11-H), 2.56-2.51 (m, 1H, C12-H), 2.44 (d, J = 19.0 Hz, 1H, C5-H), 2.35 (d, J = 0.9 Hz, 1H, OH) 2.28-2.24 (m, 1H, C12-H), 2.23 (d, J = 20.0 Hz, 1H, C5-H), 1.79 (dd, J = 2.9, 1.4 Hz, 3H, C19-H), 1.73 (s, 3H, C15-C$\underline{H_3}$), 1.43 (s, 1H, OH), 1.00 (d, J = 6.6 Hz, 3H, C18-H) ppm.
^{13}C NMR ($CDCl_3$, 125 MHz): δ 209.6, 208.2, 158.2, 144.0 (3C), 141.6, 139.4, 134.4, 128.5 (6C), 127.8 (6C), 127.1 (3C), 123.4, 116.6, 87.1, 76.5, 73.4 68.0, 58.3, 58.0, 46.6, 44.4, 38.6, 37.9, 19.7, 17.8, 10.2 ppm.
IR (thin film): ν 3445, 3061, 2968, 1694, 1627, 1491, 1449, 1374, 1336, 1222, 1154, 1056, 1014, 946, 909, 766, 733, 707, 646, 632 cm^{-1}
R_f = 0.7 (100 % EtOAc); visible under UV-lamp
$[\alpha]_D^{22.1}$ = + 121.4° (*c* 1.220, CH_2Cl_2)
HRMS (*m/z*): Calculated for $C_{39}H_{40}O_5Na^+$: 611.2768. Found: 611.2768.



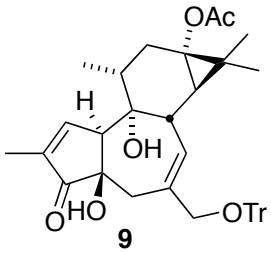
8

To a 2000 mL round-bottom flask containing a stirbar and **7** (4.15 g, 7.10 mmol, 1.0 eq) at room temperature was added MeOH (415 mL, 0.017 M). After dissolution of the solid, N_2H_4•(H_2O) (1.7 mL, 35 mmol, 5.0 eq) and AcOH (1.0 mL, 18 mmol, 2.5 eq) were added. After 2 hours, EtOAc (705 mL) and activated, basic Al_2O_3 (105 g)

were added, and the resulting mixture was stirred for 15 min. The mixture was filtered through a plug of sand (1.0 cm), Celite® (4 cm), sand (1.0 cm) layered in a fritted funnel (300 mL size) and then washed with EtOAc (300 mL). The filtrate was concentrated under reduced pressure.

The residue was transferred using EtOAc into a heavy-walled 350 mL pressure vessel and concentrated under reduced pressure; the residue was placed under high-vacuum. The flask was capped with a rubber septum and flushed with argon. Toluene (200 mL) and *i*Pr_2Net, (24 mL, 0.29 M) were added to the flask, and using a cannula, a gentle stream of argon was passed through the mixture for 15 min. The rubber septum was carefully exchanged for a threaded Teflon plug and the sealed reaction vessel placed into an oil bath preheated to 140 °C. After 16.5 h, the slightly turbid, yellowish mixture was cooled to room temperature and the threaded Teflon plug quickly exchanged for a rubber septum, and the flask was purged with argon.

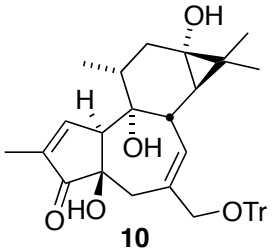
The mixture was immediately cooled to 0 °C, and a clear and colorless to slightly yellow solution of $Pb(OAc)_4$ (9.38 g, 21.1 mmol, 3.0 eq) in CH_2Cl_2 (75 mL) prepared ahead of time was added, and after 15 min, the mixture was warmed to ambient temperature (23 °C). After 1h, the mixture was diluted with EtOAc and then washed with a saturated aqueous solution of $NaHCO_3$, water and brine. The organic phase was dried over Na_2SO_4, filtered and concentrated under reduced pressure. The residue was purified by flash chromatography (20 %, 30%, 40% EtOAc/Hexanes) to afford **8** (2.34 g, 50%) whose spectral data matched that previously reported.[2] A reaction performed on a smaller scale (49 mg of product) with cooling to -78 °C for $Pb(OAc)_4$ addition proceeded in a 59% yield.



9

A gentle stream of argon was introduced into a vial with a solution of **8** (34 mg, 0.52 mmol) for 15 min. The reaction vial was placed into a Rayonett and exposed to 350 nM radiation for 40 min. The vial was removed from the Rayonett, the contents allowed to cool to room temperature and the solution concentrated under reduced pressure. The resulting residue was purified by flash chromatography (20%, 40% EtOAc/Petroleum Ether) to afford **9** (29 mg, 64%) as a white solid.

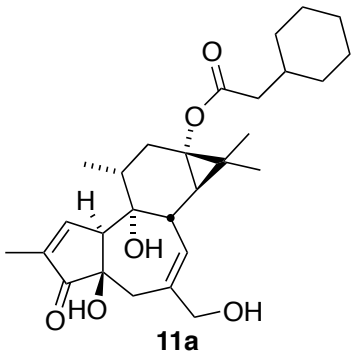
Characterization data for **9**
^{1}H NMR ($CDCl_3$, 500 MHz): δ 7.59 (s, 1H, C2-H), 7.43 - 7.26 (m, 6H, Ar-H), 7.30 – 7.26 (m, 6H, Ar-H), 7.24 – 7.20 (m, 3H, Ar-H), 5.62 (d, J = 4.1 Hz, 1H, C7-H), 5.24 (bs, 1H, C9-OH), 3.51 (s, 2H, C20-H), 3.27 (s, 1H, C10-H), 2.93 (dd, J = 5.3, 5.3 Hz, 1H, C8-H), 2.51 (d, *J* = 19.0 Hz, 1H, C5-H), 2.39 (d, *J* = 18.9 Hz, 1H, C5-H), 2.11 – 2.04 (m, 2H, C12-H, C-OH), 2.06 (s, 3H, COC$\underline{H_3}$), 1.94 (ddq, *J* = 11.3, 6.9, 6.6 Hz, 1H, C11-H), 1.77 (dd, *J* = 2.9, 1.2 Hz, 3H, C19-H), 1.57 (dd, *J* = 14.6, 11.3 Hz, C12-H), 1.19 (s, 3H, C15-C$\underline{H_3}$), 1.07 (s, 3H, C15-C$\underline{H_3}$), 0.88 (d, *J* = 6.5 Hz, 3H, C18-H), 0.82 (d, *J* = 5.3 Hz, 1H, C14-H) ppm
^{13}C NMR ($CDCl_3$, 100 MHz): δ 209.5, 173.4, 161.6, 144.2 (3C), 137.7, 132.8, 130.9, 128.9 (6C), 128.0 (6C), 127.2 (3C), 87.0, 76.0, 74.1, 69.6, 63.9, 55.9, 39.6, 39.4, 36.5, 32.6, 32.1, 23.5, 22.9, 21.5, 18.8, 15.6, 10.4 ppm.
IR (thin film): ν 3405, 3059, 2925, 2873, 1723, 1713, 1692, 1630, 1493, 1449, 1380, 1369, 1328, 1258, 1248, 1219, 1178, 1155, 1134, 1080, 1054, 1031, 1018, 982, 946, 899, 884, 754, 705, 667 cm^{-1}
R_f = 0.82 (50% EtOAc/Pentane), visible by UV-lamp
$[\alpha]_D^{20.0}$ = + 43.9 ° (*c* 0.915, $CHCl_3$)
HRMS (*m/z*): Calculated for $C_{41}H_{44}O_6Na^+$: 655.3030 Found: 655.3034.



10

To a 20 mL vial with a stirbar containing **9** (29.0 mg, 0.0458 mmol, 1.0 eq) and MeOH (7.6 mL, 0.0006 M) at room temperature (23 °C) was added $Ba(OH)_2$•$8H_2O$ (145 mg, 0.458 mmol, 10.0 eq). After completed reaction, the mixture was diluted with CH_2Cl_2, washed with pH 7 sodium potassium phosphate buffer, water and brine. The organic phase was then dried over Na_2SO_4, filtered and concentrated under reduced pressure. The residue was purified by flash chromatography (30% to 50% EtOAc/Petroleum Ether) to afford **10** (24.0 mg, 89%).

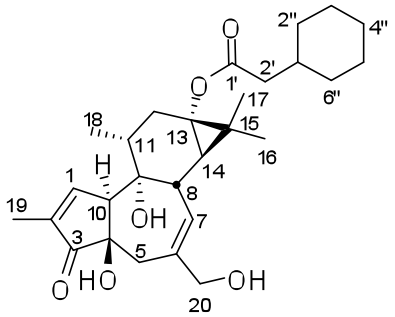
Characterization data for **10**
^{1}H NMR ($CDCl_3$, 500 MHz): δ 7.55 (s, 1H, C1-H), 7.45 – 7.42 (m, 6H, Ar-H), 7.31-7.28 (m, 6H, Ar-H), 7.31-7.28 (m, 6H, Ar-H), 7.26-7.22 (m, 3H, Ar-H), 5.89 (d, *J* = 3.9, 1H, C7-H), 3.57 (d, *J* = 11.8 Hz, 1H, C20-H), 3.54 (d, *J* = 11.8 Hz, 1H, C20-H), 3.09 (s, 1H, C10-H), 2.80 (dd, *J* = 5.5 Hz, 1H, C8-H), 2.50 (d, *J* = 19.0 Hz, 1H, C5-H), 2.35 (d, *J* = 19.0 Hz, 1H, C5-H), 2.24 (bs, 1H, C-OH), 2.03 (dd, *J* = 14.3, 7.3 Hz, 1H, C12-H), 1.98 (m, 1H, C11-H), 1.79 (s, 3H, C19-H), 1.60 (dd, *J* = 14.1, 9.1 Hz, 1H, C12-H), 1.26 (s, 3H, C15-C$\underline{H_3}$), 0.99 (s, 3H, C15-C$\underline{H_3}$), 0.95 (d, *J* = 6.6 Hz, 3H, C18-H), 0.64 (d, *J* = 6.5 Hz, 1H, C14-H) ppm
^{13}C NMR ($CDCl_3$, 125 MHz): δ 209.4, 160.5, 144.3 (3C), 137.8, 134.1, 130.9, 128.9 (6C), 128.0 (6C), 127.2 (3C), 92.3, 87.1, 74.0, 69.6, 59.4, 56.7, 39.6, 39.3, 36.7, 36.0, 33.5, 24.6, 22.5, 19.3, 16.3, 10.4 ppm.
IR (thin film): ν 3400, 2919, 2868, 1690, 1625, 1597, 1488, 1447, 1374, 1325, 1300, 1217, 1186, 1147, 1054, 1028, 1000, 943, 922, 897, 754, 703 cm^{-1}
R_f = 0.38 (50% EtOAc/Pentane), visible by UV-lamp
$[\alpha]_D^{20.0}$ = +40.1° (*c* = 1.120, $CHCl_3$)
HRMS (*m/z*): Calculated for $C_{39}H_{42}O_5Na^+$: 613.2924 Found: 613.2928.



11a

To a solution of the alcohol **10** (9.4 mg, 16 µmol) in CH_2Cl_2 (0.16 mL) was successively added 1-ethyl-3-(3'-dimethylaminopropyl)-carbodiimide hydrochloride (6.1 mg, 32 umol, 2.0 equiv.), cyclohexylacetic acid (2.7 mg, 19 µmol, 1.2 equiv.) and a catalytic amount of DMAP at room temperature. The reaction mixture was stirred at room temperature for 20 h until TLC-control indicated complete consumption of the alcohol **10**. The mixture was diluted with EtOAc (10 mL) and the org. Phase was washed with aq. sat. NH_4Cl-solution (5 mL). The aq. phase was extracted with EtOAc (3 × 5 mL) and the combined organic phases were dried over Na_2SO_4. The solvents were removed under reduced pressure and the residue was treated with $HClO_4$ (0.03 M in MeOH, 2 mL) at room temperature. The resulting mixture was stirred at room temperature for 3 h. The mixture was diluted with EtOAC (10 mL) and the org. Phase was washed with aq. sat. $NaHCO_3$-solution (3 mL). The aq. phase was extracted with EtOAc (4 × 5 mL) and the combined organic phases were dried over Na_2SO_4. After evaporation of the solvents in vacuo the residue was purified by flash chromatography (35% EtOAc/Hexanes) to give 12-deoxyphorbol-13-cyclohexylacetate **11a** (4 mg, 53%) as a white solid.

Characterization data for **11a**

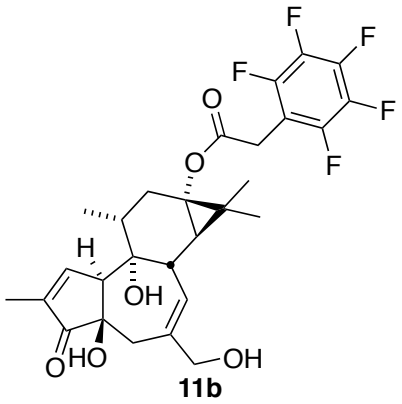


^{1}H NMR ($CDCl_3$, 500 MHz): δ 7.59 (br. q, $J \approx 1$ Hz, 1H, C1-H), 5.68 (br. d, $J = 4.7$ Hz, 1H, C7-H), 5.63 (br. s. 1H, C9-OH), 4.01 (m, 2H, C20-H_2), 3.27 (br. s. 1H, C10-H), 2.99 (br. dd, $J = 5.2$, 5.2 Hz, 1H, C8-H), 2.51 (d, $J = 18.9$ Hz, 1H, C5-H^B), 2.45 (d, $J = 18.9$ Hz, 1H, C5-H^A), 2.21 (s, 1H, OH), 2.17 (d, $J = 7.2$ Hz, 2H, C2'-H_2), 2.05 (dd, $J = 14.6$, 7.0 Hz, 1H, C12-H^2), 1.96 (m, 1H, C11-H), 1.77 (dd, $J = 2.9$, 1.3 Hz, C19-H_3), 1.55 (dd, $J = 14.6$, 11.3 Hz, 1H, C12-H^1), 0.90-1.02, 1.10-1.34, and 1.65-1.73 (3 x m, 11H, C1''-H, C2''-H_2, C3''-H_2, C4''-H_2, C5''-H_2, C6''-H_2), 1,06 and 1.18 (2 × s, 2 × 3H, C16-H_3, C17-H_3), 0.88 (d, $J = 6.5$ Hz, 3H, C18-H_3), 0.81 (d, $J = 5.3$ Hz, 1H, C14-H) ppm.

^{13}C NMR ($CDCl_3$, 125 MHz): δ 209.40, 175.57, 161.60, 139.98, 132.99, 130.47, 76.19, 73.96, 68.48, 63.49, 55.99, 42.49, 39.40, 38.90, 36.57, 35.08, 33.22, 33.16, 32.84, 32.06, 26.26, 26.18, 26.14, 23.51, 22.89, 18.80, 15.59, 10.36 ppm.

IR (thin film): ν 3395, 2919, 2852, 1708, 1628, 1447, 1377, 1354, 1328, 1287, 1261, 1238, 1217, 1183, 1165, 1121, 1107, 1013, 943, 886, 801, 754, 664 cm^{-i}

R_f = 0.2 (50% EtOAc/Hexanes; visible under UV-lamp)
$[\alpha]_D^{25}$ = +82.1 (*c* 0.18, $CHCl_3$).
HRMS (*m/z*): Calculated for $C_{28}H_{40}O_6Na^+$: 495.2717. Found: 495.2714.

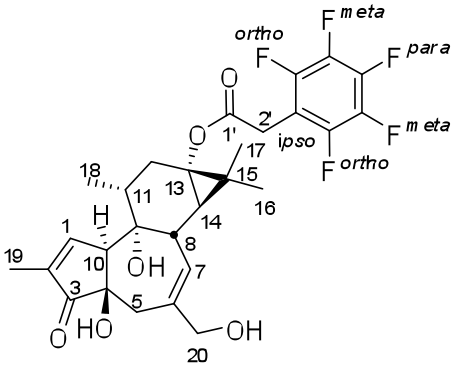


11b

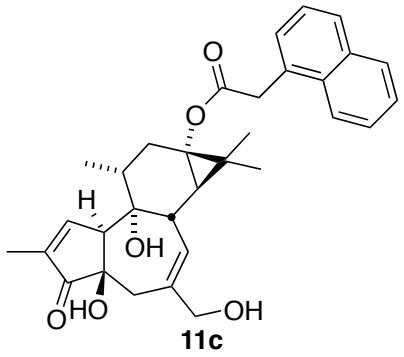
To a solution of the alcohol **10** (8 mg, 13 µmol) in CH_2Cl_2 (0.27 mL) were successively added 1-ethyl-3-(3'-dimethylaminopropyl)-carbodiimide hydrochloride (6.2 mg, 32 umol, 2.4 equiv.), pentaflourophenylacetic acid (3.7 mg, 16 µmol, 1.2 equiv.) and a catalytic amount of DMAP at room temperature. The reaction mixture was stirred at room temperature for 2 h until TLC-control indicated complete consumption of the alcohol **10**. The mixture was diluted with CH_2Cl_2 (10 mL) and the org. Phase was washed with aq. sat. NH_4Cl-solution (5 mL). The aq. phase was extracted with CH_2Cl_2 (3 × 5 mL) and the combined organic phases were dried over Na_2SO_4. After removal of the solvents in vacuo the residue was purified by flash chromatography (15% EtOAc/Hexanes) to give **C20-trityl-11b** (10.4 mg, 92%) as a white solid**.**

Trityl-protected 12-deoxyphorbol-13-pentafluorophenylacetate **C20-trityl-11b** (26 mg, 37 µmol) was treated with $HClO_4$ (0.03 M in MeOH, 3 mL) at room temperature. The resulting mixture was stirred at room temperature for 3 h until TLC-control indicated complete consumption of the starting material. The mixture was diluted with EtOAc (10 mL) and the org. Phase was washed with aq. half-sat. $NaHCO_3$-solution (7 mL). The aq. phase was extracted with EtOAc (3 × 10 mL) and the combined organic phases were dried over Na_2SO_4. After removal of the solvents in vacuo the residue was subjected to flash chromatography (35% EtOAc/Hexanes) to afford **11b** (5.9 mg, 88%; 81% over both steps) as a white solid.

Characterization data for **11b**



^{1}H NMR ($CDCl_3$, 500 MHz): δ 7.57 (s, 1H C1-H), 5.68 (d, *J* = 4.2 Hz, 1H, C7-H), 4.98 (br. s, 1H, C9-OH), 4.03 (m, C20-H_2), 3.76 (m, 2H, C2'-H_2), 3.27 (br. s. 1H, C10-H), 3.02 (m, 1H, C8-H), 2.54 (d, *J* = 19.0 Hz, 1H, C5-H^A), 2.46 (d, *J* = 19.0 Hz, 1H, C5-H^B), 2.23 (br. s. 1H, OH), 2.12 (dd, *J* = 14.9, 7.0 Hz, 1H, C12-H^1), 2.00 (m, 1H, C11-H), 1.79 (dd, *J* = 2.9, 1.4 Hz, C19-H_3), 1.58 (dd, J = 14.8, 11.3 Hz, 1H, C12-H^2), 1.09 and 1.16 (2 × s, 2 ×3H, C16-H_3, C17-H_3), 0.88-0.93 (m, 4H, C14-H, C18-H_3) ppm
^{13}C NMR ($CDCl_3$, 125 MHz): δ 209.31, 170.75, 161.21, 140.25, 133.20, 130.02, 76.22, 73.88, 65.58, 68.40, 55.92, 39.26, 38.85, 36.47, 32.62, 31.81, 28.25, 23.33, 23.27, 18.72, 15.48, 10.35 ppm. Resonance-peaks for the fluorinated aryl-ring (C^{ipso}, 2 x C^{ortho}, 2 x C^{meta}, C^{para}) could not be detected.
^{19}F-NMR (C_6D_6/$CFCl_3$ as external standard, 376 MHz): δ = –142.54 (dd, *J* = 23.0, 9.0 Hz, 2F), −154.95 (dd, *J* = 21.5, 21.5 Hz, 2F), −162.30 (m, 1F) ppm.
IR (thin film): ν 3421, 2956, 2919, 2873, 1718, 1700, 1656, 1625, 1522, 1506, 1457, 1416, 1374, 1349, 1325, 1305, 1261, 1235, 1214, 1176, 1129, 1093, 1077, 1036, 1013, 979, 946, 912, 886, 863, 798, 754, 664 cm^{-1}
R_f = 0.25 (50% EtOAc/Hexanes) one darkblue spot in p-anisaldehyde (visible under UV-lamp).
$[\alpha]_D^{23}$ = 124.19 (*c* 0.17, $CHCl_3$)
HRMS (*m/z*): Calculated for $C_{28}H_{29}F_5O_6Na^+$: 579.1776. Found: 579.1775.

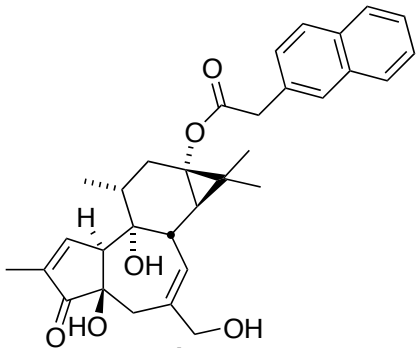


11c

To a solution of **10** (11.5 mgs 0.019 mmol) in CH_2Cl_2 (1.7 mL) in a disposable vial was added 1-naphthylacetic acid (6.6 mg, 0.037 mmol, 1.95 eq), followed by 1-ethyl-3-(3'-dimethylaminopropyl)-carbodiimide hydrochloride (7.4 mg, 0.037 mmol, 1.95 eq) and a catalytic amount of DMAP. This mixture stirred for 2 hours, at which point the reaction mixture was diluted with EtOAc, washed one time with a saturated aqueous solution of NH_4Cl, one time with a saturated aqueous solution of $NaHCO_3$, and one time with brine. The organic layer was dried over anhydrous sodium sulfate, filtered, and the solvent was removed *in vacuo*. The crude product was purified by flash chromatography (silica gel, 20% EtOAc/pentane) and carried forward.

The material was evenly divided into two disposable glass vials, and dissolved in methanol (0.8 mL each) and perchloric acid (8 mL each). After stirring at room temperature for 50 minutes, the reaction was stopped by stirring with solid sodium bicarbonate (85 mg each vial) for 15 minutes. The mixture was filtered through a plug of celite, washing with copious quantities of ethyl acetate, then dried *in vacuo*. The product was purified by preparative HPLC to afford **11c** (6 mg, 0.011 mmol, 61% two-step yield) as a white powder.

Characterization data for **11c**
^{1}H NMR ($CDCl_3$, 500 MHz): δ 7.97 (d, 1H, J = 8.2 Hz, Ar-H), 7.87 (d, 1H, J = 7.9 Hz, Ar-H), 7.80 (d, 1H, J = 7.9 Hz, Ar-H), 7.55-7.49 (m, 3H, C1-H, Ar-H), 7.44-7.39 (m, 2H, Ar-H), 5.58 (d, 1H, J = 4.4 Hz, C7-H), 5.33 (s, 1H, OH), 4.09 (d, 1H, J = 15.3 Hz, C20-H), 4.04 (d, 1H, J = 15.3 Hz, C20-H), 3.99 (dd, 1H, J = 4.6 Hz, 12.8 Hz, -C$\underline{H_2}$-Ar), 3.95 (dd, 1H, J = 5.3 Hz, 12.6 Hz, -C$\underline{H_2}$-Ar), 3.22 (s, 1H, C10-H), 2.92 (s, 1H, C8-H), 2.47 (d, 1H, J = 19.1 Hz, C5-H), 2.42 (d, 1H, J = 19.1 Hz, C5-H), 2.08 (s, 1H, OH), 2.02 (dd, 1H, J = 7.0 Hz, 14.7 Hz, C12-H), 1.95-1.89 (m, 1H, C11-H), 1.76 (bs, 3H, C19-H), 1.46 (t, 1H, J = 5.9 Hz, C12-H), 1.25 (s, 1H, OH), 0.97 (s, 3H, C15-C$\underline{H_3}$), 0.84 (d, 3H, J = 6.5 Hz, C18-H), 0.81 (s, 3H, C15-C$\underline{H_3}$), 0.67 (d, 1H, J = 5.3 Hz, C14-H) ppm
^{13}C NMR ($CDCl_3$, 125 MHz): δ 209.1, 173.7, 161.3, 161.2, 139.7, 133.8, 132.8, 132.0, 129.9, 128.8, 128.2, 125.5, 123.6, 75.9, 73.7, 68.2, 64.1, 55.8, 55.6, 39.6, 39.1, 39.0, 38.6, 36.3, 32.3, 31.6, 22.9, 22.7, 18.5, 15.3, 15.3, 10.1 ppm.
IR (thin film): ν 3390, 2920, 2868, 1704, 1333, 1273, 1243, 1133, 1017, 910, 785, 731 cm^{-1}
R_f = 0.43 (75% EtOAc/pentane) visible by UV-lamp
$[\alpha]_D$ = + 46.8° (*c* 0.24, MeOH)
HRMS (*m/z*): Calculated for $C_{32}H_{36}O_6Na^+$: 539.2410 Found: 539.2415.



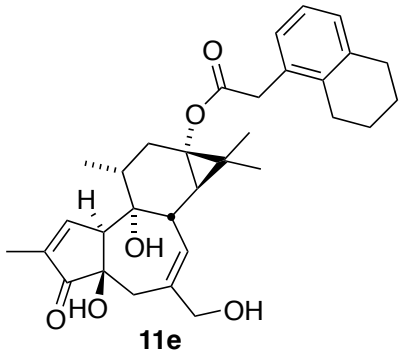
11d

To a solution of **10** (11.0 mg, 0.019 mmol) in CH_2Cl_2 (1.7 mL) in a disposable vial was added 2-naphthylacetic acid (6.9 mg, 0.037 mmol, 1.95 eq), followed by 1-ethyl-3-(3'-dimethylaminopropyl)-carbodiimide hydrochloride (7.6 mg, 0.037 mmol, 1.95 eq) and a catalytic amount of DMAP. This mixture stirred for 2 hours, at which point the reaction mixture was diluted with EtOAc, washed one time with a saturated aqueous solution of NH_4Cl, one time with a saturated aqueous solution of $NaHCO_3$, and one time with brine. The organic layer was dried over anhydrous sodium sulfate, filtered, and the solvent was removed *in vacuo*. The crude product was purified by flash chromatography (20% EtOAc/pentane) and carried forward.

In a disposable glass vial, the ester product was dissolved in a pre-made solution of 0.003 M $HClO_4$ in methanol (3 mL). After stirring at room temperature for 3 hours, the reaction was stopped by stirring with solid sodium bicarbonate (80 mg) for 15 minutes. The mixture was filtered through a plug of celite, washing with copious

quantities of EtOAc, then dried *in vacuo*. The product was purified by silica gel chromatography to afford **11d** (9.0 mg, 92% two-step yield) as a white powder.

Characterization data for **11d**
^{1}H NMR ($CDCl_3$, 600 MHz): δ 7.83-7.79 (m, 3H, Ar-H), 7.73 (s, 1H, Ar-H), 7.57 (s, 1H, C1-H), 7.50-7.45 (m, 2H, Ar-H), 7.40 (dd, 1H, J = 1.7 Hz, 8.3 Hz, Ar-H), 5.62 (d, 1H, J = 5.6 Hz, C7-H), 5.34 (bs, 1H, OH), 4.01 (d, 1H, J = 12.7, C20-H), 3.96 (d, 1H, J = 13.7, C20-H), 3.79 (d, 1H, J = 14.9 Hz, -C$\underline{H_2}$-Ar), 3.76 (d, 1H, J = 14.7 Hz, -C$\underline{H_2}$-Ar), 3.25 (s, 1H, C10-H), 2.96 (t, 1H, J = 5.4 Hz, C8-H), 2.49 (d, 1H, J = 19.0 Hz, C5-H), 2.43 (d, 1H, J = 19.1 Hz, C5-H), 2.07-2.04 (m, 2H, C12-H, OH), 1.98-1.92 (m, 1H, C11-H), 1.77 (dd, 3H, J = 1.5 Hz, J = 2.8 Hz, C19-H), 1.45 (bs, 1H, OH), 1.04 (s, 3H, C15-C$\underline{H_3}$), 1.03 (s, 3H, C15-C$\underline{H_3}$), 0.86 (d, 3H, J = 6.6 Hz, C18-H), 0.77 (d, 1H, J = 5.4 Hz, C14-H) ppm
^{13}C NMR ($CDCl_3$, 125 MHz): δ 209.4, 173.8, 161.5, 140.1, 133.6, 133.1, 132.8, 130.9, 130.4, 128.57, 128.40, 127.9, 127.6, 126.5, 126.2, 76.2, 74.0, 68.5, 64.4, 56.0, 42.1, 39.4, 38.9, 36.5, 32.7, 32.0, 23.40, 23.24, 18.8, 15.6, 10.4 ppm.
IR (thin film): ν 3391, 2921, 1694, 1633, 1336, 1189, 1172, 1017, 732 cm^{-1}
R_f = 0.49 (75% EtOAc/pentane) visible by UV-lamp
$[\alpha]_D$ = + 23.43° (*c* 0.24, MeOH)
HRMS (*m/z*): Calculated for $C_{32}H_{36}O_6Na^+$: 539.2410 Found: 539.2406.



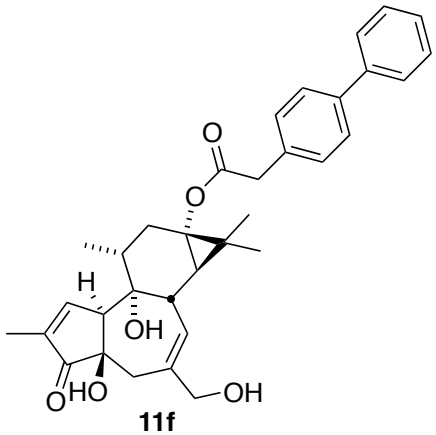
11e

To a 5 mL vial with **10** (5.5 mg, 0.0093 mmol) and THF (95 μL) at room temperature were added 1-ethyl-3-(3'-dimethylaminopropyl)-carbodiimide hydrochloride (4.5 mg, 0.023 mmol), 5,6,7,8-tetrahydro-1-naphthaleneacetic acid (2.2 mg, 0.011 mmol) and a catalytic amount of DMAP. The mixture was stirred 3 h 45 min and was then diluted with EtOAc (50 mL). The organic solution was washed with saturated aqueous NH_4Cl (10 mL), H_2O (10 mL), saturated aqueous $NaHCO_3$ (10 mL), H_2O (10 mL), and Brine (10 mL). The organic phase was dried over Na_2SO_4, filtered, and concentrated. The resulting residue was purified by flash chromatography (15% EtOAc/petroleum ether) to afford **C20-trityl-11e** (6.2 mg) as a residue.

To a solution of the aforementioned **C20-trityl-11e** (6.2 mg, 0.008 mmol) in MeOH (825 μL) was added $HClO_4$ (60% in H_2O, 8 μL). The resulting solution was stirred for 40 min and was then diluted with EtOAc (50 mL). The organic solution was washed with saturated aqueous $NaHCO_3$ (2 x 10 mL), H_2O (10 mL), and Brine (10 mL). The organic phase was dried over Na_2SO_4, filtered, and concentrated. The resulting residue was purified by flash

column chromatography (1% MeOH/CH_2Cl_2, 3% MeOH/CH_2Cl_2) and further purified by preparative reverse-phase HPLC (40% to 100% acetonitrile/H_2O) to afford **11e** (3.8 mg, 78% over two steps) as a white solid.

Characterization data for **11e**
^{1}H NMR ($CDCl_3$, 500 MHz): δ 7.56 (s, 1H, C1-H), 7.05 (m, 1H, Ar-H), 6.99-7.01 (m, 2H, Ar-H), 5.63 (d, 1H, J = 4.5 Hz, C7-H), 5.41 (s, 1H, C9-OH), 4.01 (d, 1H, J = 12.9 Hz, C20-H), 3.96 (d, 1H, J = 12.4 Hz, C20-H), 3.58 (s, 2H, COC$\underline{H_2}$-Ar), 3.23 (bs, 1H, C10-H), 2.96 (t, 1H, J = 5.1 Hz, C8-H), 2.77 (t, 2H, J = 6.2 Hz, Ar-C$\underline{H_2}$-CH_2-), 2.65-2.69 (m, 2H, Ar-C$\underline{H_2}$-CH_2-), 2.50 (d, 1H, J = 19.1 Hz, C5-H), 2.43 (d, 1H, J = 19 Hz, C5-H), 2.30 (s, 1H, OH), 2.05 (dd, 1H, J = 7 Hz, 14.7 Hz, C12-H), 1.91-1.99 (m, 1H, C11-H), 1.73-1.85(m, 4H, Ar-CH_2-C$\underline{H_2}$), 1.76 (dd, 3H, J = 1.0 Hz, 2.8 Hz, C19-H), 1.56 (dd, 1H, J = 11.3 Hz, 14.6 Hz, C12-H), 1.03 (s, 6H, C15-C$\underline{H_3}$ & C15-C$\underline{H_3}$), 0.86 (d, 3H, J = 6.5 Hz, C18-H), 0.75 (d, 1H, J = 5.3 Hz, C14-H) ppm
^{13}C NMR ($CDCl_3$, 125 MHz): δ 209.2, 173.8, 161.3, 139.8, 137.6, 135.6, 132.8, 132.0, 130.1, 128.7, 127.6, 125.4, 76.0, 73.7, 68.2, 63.8, 55.7, 39.1, 39.0, 38.6, 36.3, 32.4, 31.7, 30.0, 26.5, 23.3, 23.0, 22.9, 22.7, 18.5, 15.4, 10.1 ppm.
IR (thin film): ν 3401, 2924, 1704, 1628, 1462, 1377, 1333, 1263, 1133, 1079, 1016, 946, 910, 803, 763, 732 cm^{-1}
R_f = 0.59 (10% MeOH/CH_2Cl_2) visible by UV-lamp, one purple spot in p-anisaldehyde
$[\alpha]_D^{24.2}$ = + 38.48° (*c* 0.34, $CHCl_3$)
HRMS (*m/z*): Calculated for $C_{32}H_{40}O_6Na^+$: 543.2717. Found: 543.2713.



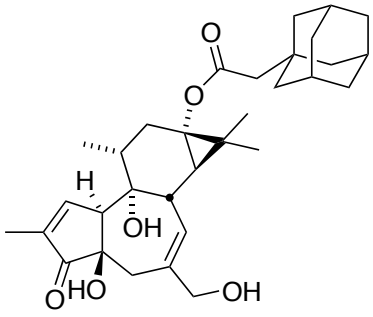
11f

To a 5 mL vial with **10** (4.9 mg, 0.0083 mmol, 1.0 eq) and THF (83 μL) at room temperature were added 1-ethyl-3-(3'-dimethylaminopropyl)-carbodiimide hydrochloride (4.0 mg, 0.0207 mmol, 2.5 eq), biphenylacetic acid (2 mg, 0.001 mmol, 1.2 eq) and a catalytic amount of DMAP. After stirring for 17 h, added additional 1-ethyl-3-(3'-dimethylaminopropyl)-carbodiimide hydrochloride (8.0 mg, 0.0414 mmol, 5 eq), biphenylacetic acid (4 mg, 0.002 mmol, 2.4 eq) and a catalytic amount of DMAP. After stirring for an additional 6 h, the mixture was diluted with EtOAc and a saturated aqueous NH_4Cl solution. The mixture was stirred for 10 min and diluted with EtOAc, washed with water and brine, and the organic phase was then dried over Na_2SO_4, filtered and concentrated under reduced pressure. The residue was purified by flash chromatography (25% EtOAc/petroleum ether) to afford **C20-trityl-11f** (5.8 mg, 89%).

To a 5 mL vial with the aforementioned **C20-trityl-11f** (4.8 mg, 0.0061 mmol, 1.0 eq) and MeOH (612 μL, 0.01 M) at room temperature was added $HClO_4$ (60% in H_2O; 6 μL). After 20 min, the solution was diluted with

EtOAc (20 mL). The organic phase was washed with a saturated aqueous $NaHCO_3$ solution (2 x 5 mL), water (5 mL) and brine (2 mL). The organic phase was dried over Na_2SO_4, filtered, concentrated under reduced pressure, and purified by flash chromatography (1%, 5% MeOH/CH_2Cl_2) to afford **11f** (3.2 mg, 96%; 85% over 2 steps) as a clear colorless film.

Characterization data for **11f**
^{1}H NMR ($CDCl_3$, 400 MHz): δ 7.60 – 7.55 (m, 5H, C1-H and Ar-H), 7.47 – 7.43 (m, 2H, Ar-H), 7.37 – 7.34 (m, 3H, Ar-H), 5.65 (d, *J* = 4.9 Hz, 1H, C7-H), 5.39 (s, 1H, C9-OH), 4.03 (d, *J* = 12.8 Hz, 1H, C20-H), 3.97 (d, *J* = 12.7 Hz, 1H, C20-H), 3.66 (s, 2H, -C$\underline{H_2}$Ar), 3.26 (s, 1H, C10-H), 3.00 (s, 1H, C8-H), 2.52 (d, *J* = 18.9 Hz, 1H, C5-H), 2.45 (d, *J* = 18.9 Hz, 1H, C5-H), 2.32 s, 1H, C-OH), 2.08 (dd, *J* = 14.6, 6.7 Hz, 1H, C12-H), 1.97 (dqd, *J* = 12.8, 6.7, 6.1 Hz, 1H, C11-H) 1.77 (m, 3H, C19-H), 1.58 (dd, *J* = 14.6, 11.6 Hz, 1H, C12-H), 1.08 (s, 3H, C15-C$\underline{H_3}$), 1.05 (s, 3H, C15-C$\underline{H_3}$), 0.88 (d, *J* = 6.1 Hz, 3H, C18-H), 0.80 (d, *J* = 4.8 Hz, 1H, C14-H) ppm
^{13}C NMR ($CDCl_3$, 100 MHz): δ 209.4, 173.8, 161.5, 140.8, 140.5, 140.1, 133.1, 132.4, 130.4, 130.0 (2C), 129.0 (2C), 127.6 (3C), 127.3 (2C), 76.2, 73.9, 68.5, 64.3, 56.0, 41.5, 39.3, 8.8, 36.5, 32.7, 32.0, 23.4, 23.2, 18.8, 15.6, 10.3 ppm.
IR (thin film): ν 3385, 2992, 2930, 2868, 1708, 1625, 1488, 1460, 1449, 1405, 1374, 1328, 1271, 1245, 1163, 1132, 1075, 1041, 1010, 940, 886, 757, 705 cm^{-1}
R_f = 0.89 (10% MeOH/CH_2Cl_2)
$[\alpha]_D^{20.0}$ = +26.5° (*c* 0.320, $CHCl_3$)
HRMS (*m/z*): Calculated for $C_{34}H_{38}O_6Na^+$: 565.2560. Found: 579.2560.

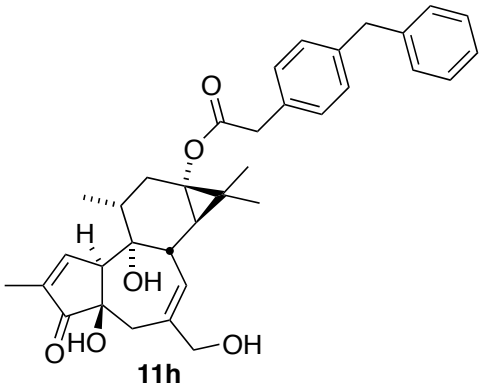


11g

To a solution of **10** (28.0 mg, 0.047 mmol) in CH_2Cl_2 (5 mL) in a disposable vial was added 1-adamantaneacetic acid (18.7 mg, 0.094 mmol, 2 eq, purchased from alfa aesar), followed by 1-ethyl-3-(3'-dimethylaminopropyl)-carbodiimide hydrochloride (19.4 mgs, 0.094 mmol, 2 eq) and DMAP (3.7 mg, 0.03 mmol, 0.6 eq). This mixture stirred for 3.75 hours at which point an additional small portion of DMAP was added. After 19.5 hours of additional stirring, additional 1-adamantaneacetic acid (18.4 mg, 0.094 mmol, 2 eq), 1-ethyl-3-(3'-dimethylaminopropyl)-carbodiimide hydrochloride (17.8 mg, 0.93 mmol, 2 eq), and DMAP (catalytic) were added. The reaction stirred for a further 3 hours at which point the reaction mixture was diluted with EtOAc, washed one time with a saturated aqueous solution of NH_4Cl, one time with a saturated aqueous solution of $NaHCO_3$, and one time with brine. The organic layer was dried over anhydrous Na_2SO_4, filtered, and the solvent was removed *in vacuo*. The crude product was purified by flash chromatography (silica gel, 20% EtOAc/pentane) and carried forward.

In a disposable glass vial, the ester product was dissolved in a pre-made solution of 0.003 M $HClO_4$ in methanol (3 mL). After stirring at room temperature for 5 hours, the reaction was stopped by stirring with solid sodium bicarbonate (80 mgs) for 10 minutes. The mixture was filtered through a plug of celite, washing with copious quantities of EtOAc, then dried *in vacuo*. The product was purified by flash chromatography (30%, 50% EtOAc/pentane) to afford **11g** (17.2 mg, 0.033 mmol, 70% two-step yield) as a white powder.

Characterization data for **11g**
^{1}H NMR ($CDCl_3$, 500 MHz): δ 7.60 (t, 1H, J = 1.8 Hz, Hz, C1-H), 5.74-5.68 (m, C7-H, OH), 4.04 (dd, 1H, J = 12.8 Hz, J = 5.7 Hz, C20-H), 3.99 (dd, 1H, J = 12.8 Hz, J = 6.4 Hz, C20-H), 3.27 (s, 1H, C10-H), 2.99 (t, 1H, J = 5.1 Hz, C8-H), 2.52 (d, 1H, J = 19.1 Hz, C5-H), 2.45 (d, 1H, J = 18.9 Hz, C5-H), 2.19 (s, 1H, C2'-H), 2.05 (d of t, 3H, J = 13.9 Hz, J = 6.5, C5'-H), 2.00-1.92 (m, 4H, C11-H, C12-H, OH, C2'-H), 1.77 (dd, 3H, J = 2.9 Hz, J = 1.3 Hz, C19-H), 1.73-1.54 (m, 13H, C12, C4', C6'), 1.20 (s, 3H, C15-CH_3), 1.06 (s, 3H, C15-CH_3), 0.89 (d, 3H, J = 6.5 Hz, C18-H), 0.82 (d, 1H, J = 5.3 Hz, C14-H) ppm
^{13}C NMR ($CDCl_3$, 125 MHz): δ 209.2, 174.2, 161.4, 139.8, 132.8, 130.3, 76.0, 73.7, 68.2, 63.3, 55.8, 48.7, 42.3, 39.2, 38.7, 36.6, 36.4, 33.2, 32.7, 31.9, 28.5, 23.5, 22.5, 18.6, 15.4, 10.1 ppm.
IR (thin film): ν 3401, 2904, 2848, 1704, 1436, 1374, 1335, 1263, 1199, 1137, 1095, 1017, 948 cm^{-1}
R_f = 0.51 (75% EtOAc/pentane), visible under UV-lamp
$[\alpha]_D$ = + 26.36° (*c* 0.09, $CHCl_3$)
HRMS (*m/z*): Calculated for $C_{32}H_{44}O_6Na^+$: 547.2998 Found: 547.3031



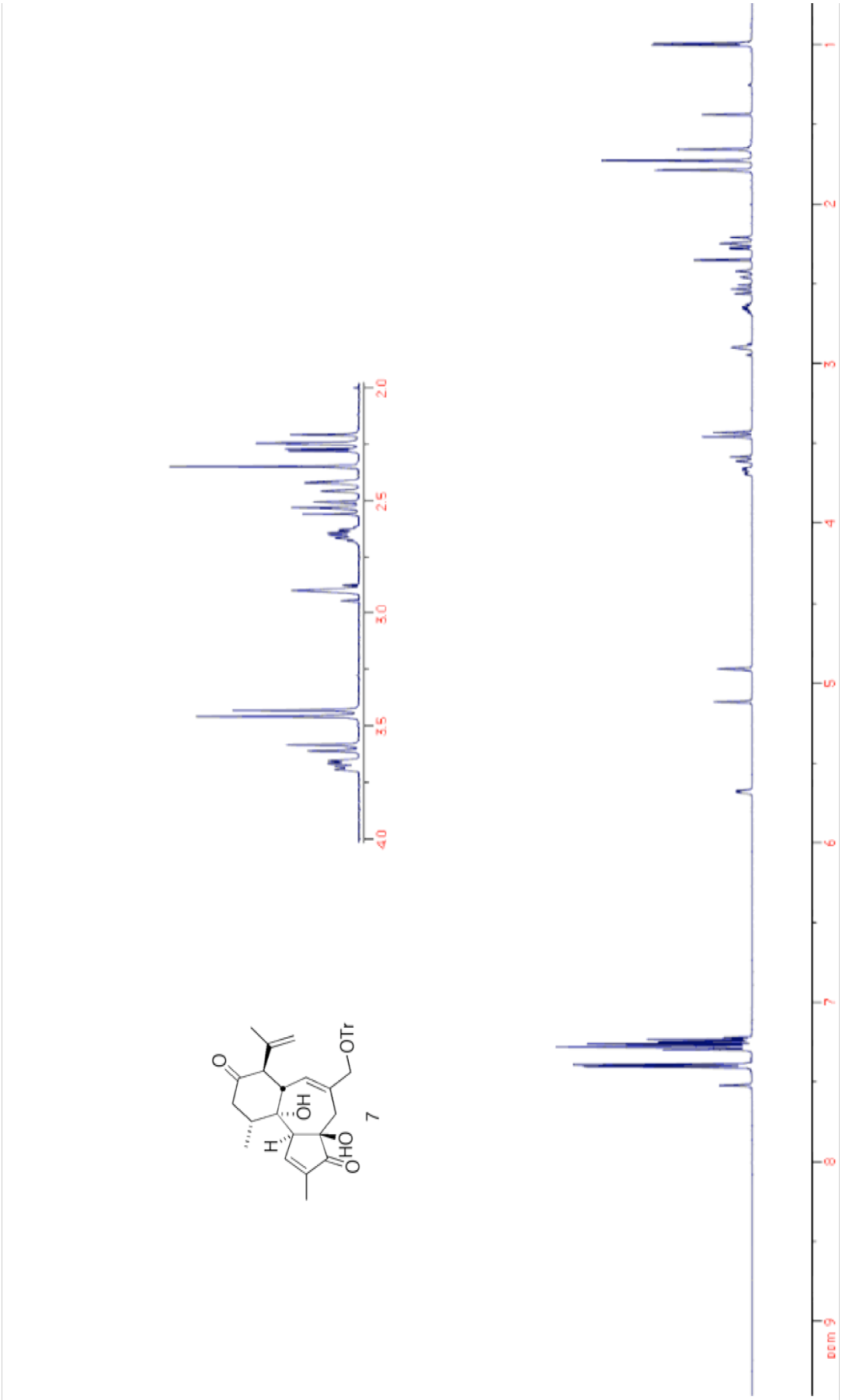
11h

To a 5 mL vial with **10** (4.0 mg, 0.0068 mmol, 1.0 eq) and THF (83 μL) at room temperature were added 1-ethyl-3-(3'-dimethylaminopropyl)-carbodiimide hydrochloride (13.0 mg, 0.068 mmol, 10 eq), p-benzyl-phenylacetic acid[3] (8 mg, 0.034 mmol, 5 eq) and a catalytic amount of DMAP. After completion of the reaction, the mixture was diluted with EtOAc and a saturated aqueous NH_4Cl solution. The mixture was stirred for 10 min and diluted with EtOAc, washed with water and brine, and the organic phase was then dried over Na_2SO_4, filtered and concentrated under reduced pressure. The residue was purified by flash chromatography (7.5%, 12.5%, 20%, 25% EtOAc/Hexanes) to afford **C20-trityl-11h** (5.0 mg, 92%).

To a 5 mL vial with the aforementioned **C20-trityl-11h** (4.3 mg, 0.0054 mmol, 1.0 eq) and MeOH (563 μL, 0.01 M) at room temperature was added $HClO_4$ (60% in H_2O; 5.6 μL). After 20 min, the solution was diluted with EtOAc (20 mL). The organic phase was washed with a saturated aqueous $NaHCO_3$ solution (2 x 5 mL), water (5 mL) and brine (2 mL). The organic phase was dried over Na_2SO_4, filtered, concentrated under reduced pressure, and

purified by flash chromatography (1%, 3% MeOH/CH_2Cl_2) to afford **11h** (2.4 mg, 80%; 74% over 2 steps) as a clear colorless film.

Characterization data for **11h**
^{1}H NMR ($CDCl_3$, 400 MHz): δ 7.57 (s, 1H, C1-H), 7.31-7.26 (m, 3H, Ar-H), 7.23-7.14 (m, 6H, Ar-H), 5.64 (d, J = 4.9 Hz, 1H, C7-H), 5.39 (bs, 1H, C9-OH), 4.02 (d, J = 15.8 Hz, 1H, C20-H), 3.97 (d, J = 15.8 Hz, 1H, C20-H), 3.97 (s, 2H, -Ar-C$\underline{H_2}$-Ar), 3.57 (s, 2H, -OCOC$\underline{H_2}$-Ar), 3.25 (s, 1H, C10-H), 2.98 (dd, J = 4.9 Hz, 4.9 Hz, 1H, C8-H), 2.52 (d, J = 18.9 Hz, 1H, C5-H), 2.44 (d, J = 18.9 Hz, 1H, C5-H), 2.34 (bs, 1H, -OH), 2.05 (dd, J = 14.0 Hz, 6.7 Hz, 1H, C12-H), 1.96 (dt, J = 6.7 Hz, 4.9 Hz, 1H, C11-H), 1.77 (s, 3H, C19-H), 1.55 (dd, J = 11.0 Hz, 14.6 Hz, 1H, C12-H), 1.04 (s, 6H, C15-C$\underline{H_3}$), 0.86 (d, J = 6.7 Hz, 3H, C18-H), 0.76 (d, J = 4.9 Hz, 1H, C14-H) ppm
^{13}C NMR ($CDCl_3$, 100 MHz): δ 209.4, 173.9, 161.5, 141.2, 140.4, 140.0, 133.1, 131.2, 130.4, 129.6 (2C), 129.4 (2C), 129.1 (2C), 128.7 (2C), 126.3, 76.2, 73.9, 68.5, 64.2, 55.9, 41.7, 41.5, 39.3, 38.8, 36.5, 32.7, 31.9, 23.3, 23.2, 18.9, 15.6, 10.3 ppm.
IR (thin film): ν 3379, 1702, 1625, 1514, 1493, 1447, 1416, 1374, 1325, 1269, 1238, 1134, 1075, 1036, 1013, 943, 878, 690 cm^{-1}
R_f = 0.84 (10% MeOH/CH_2Cl_2), visible under UV-lamp
$[\alpha]_D^{23.8}$ = + 20.7° (*c* 0.50, $CHCl_3$)
HRMS (*m/z*): Calculated for $C_{35}H_{40}O_6Na^+$: 579.2717. Found: 579.2714.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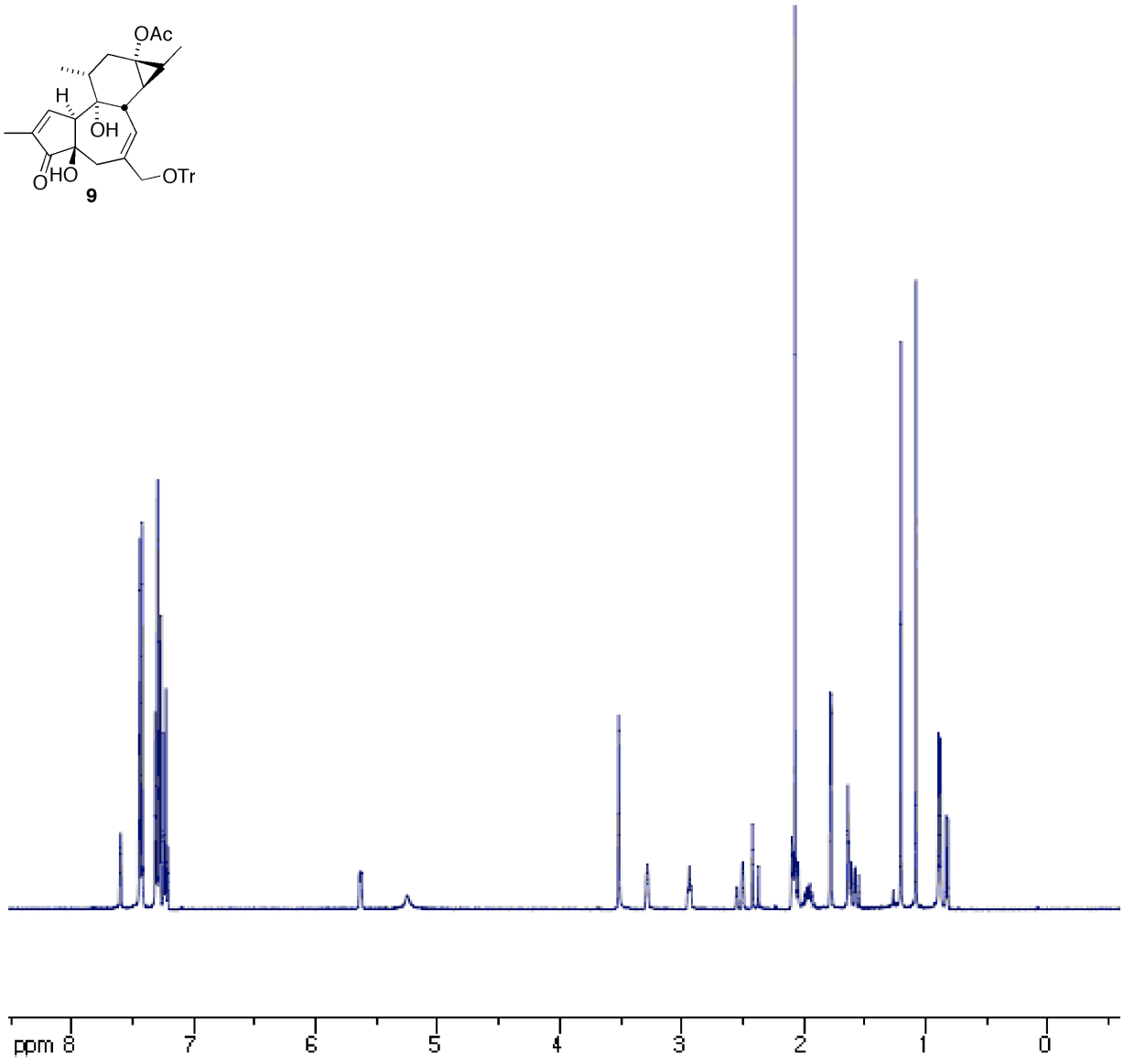
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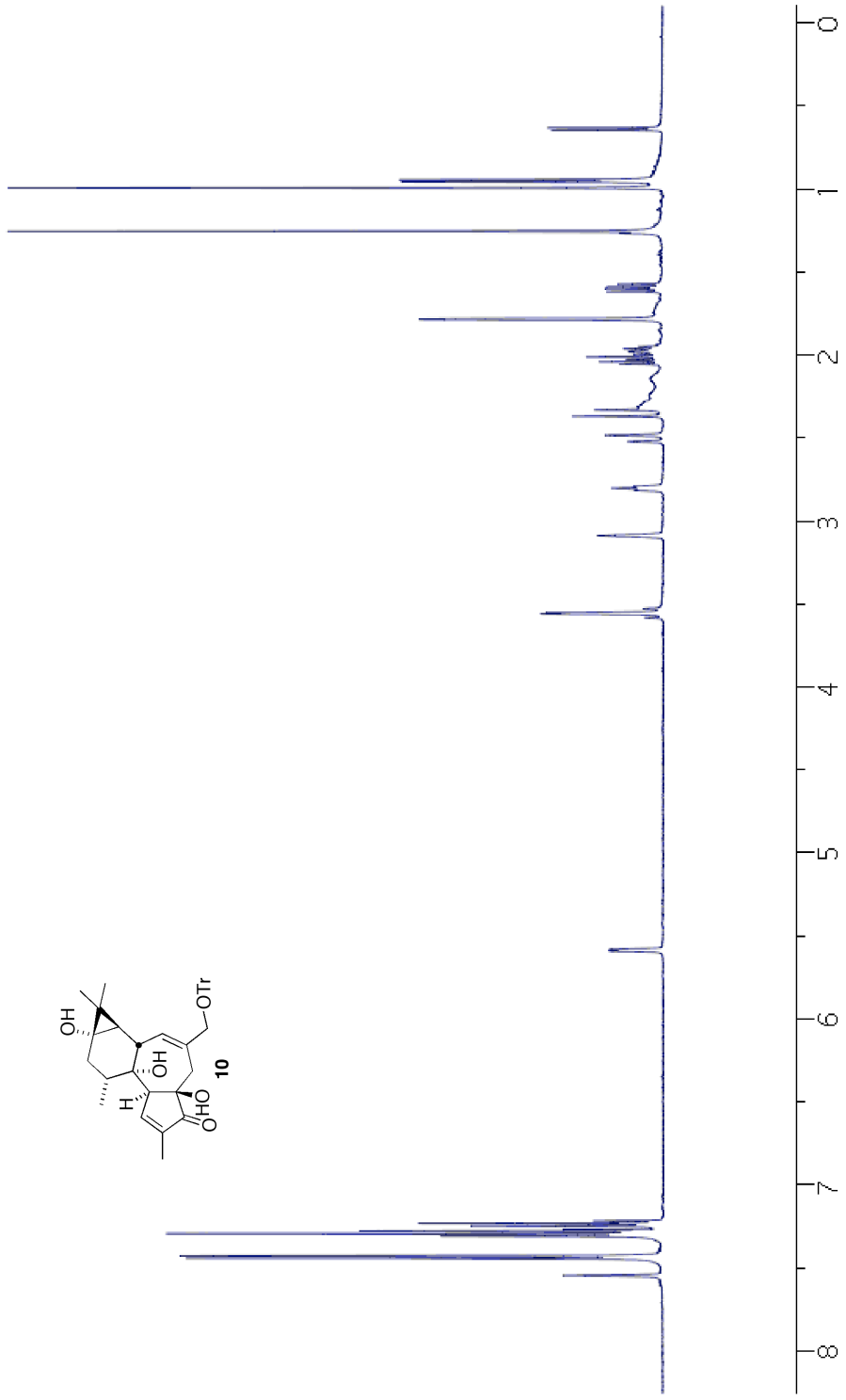
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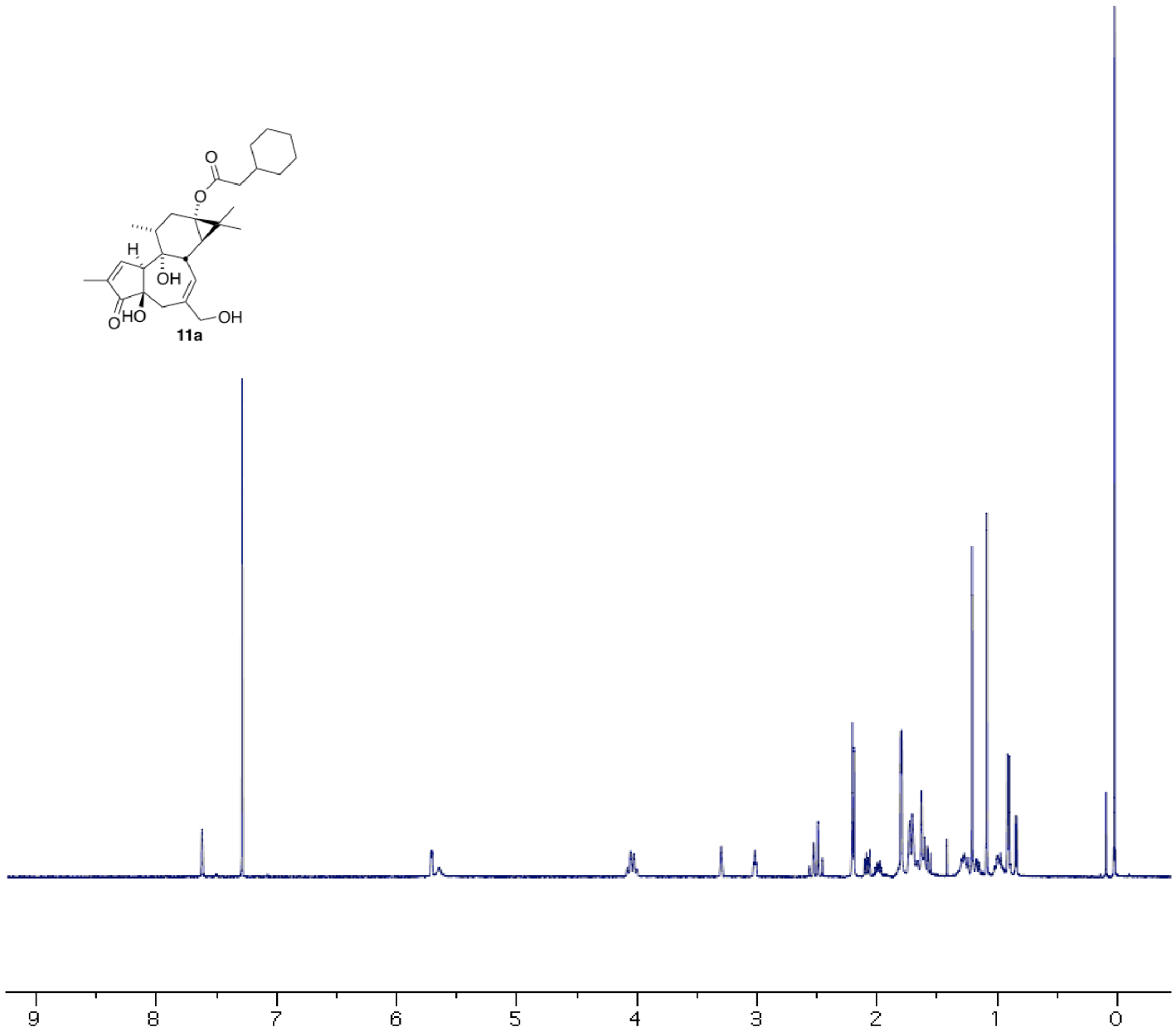
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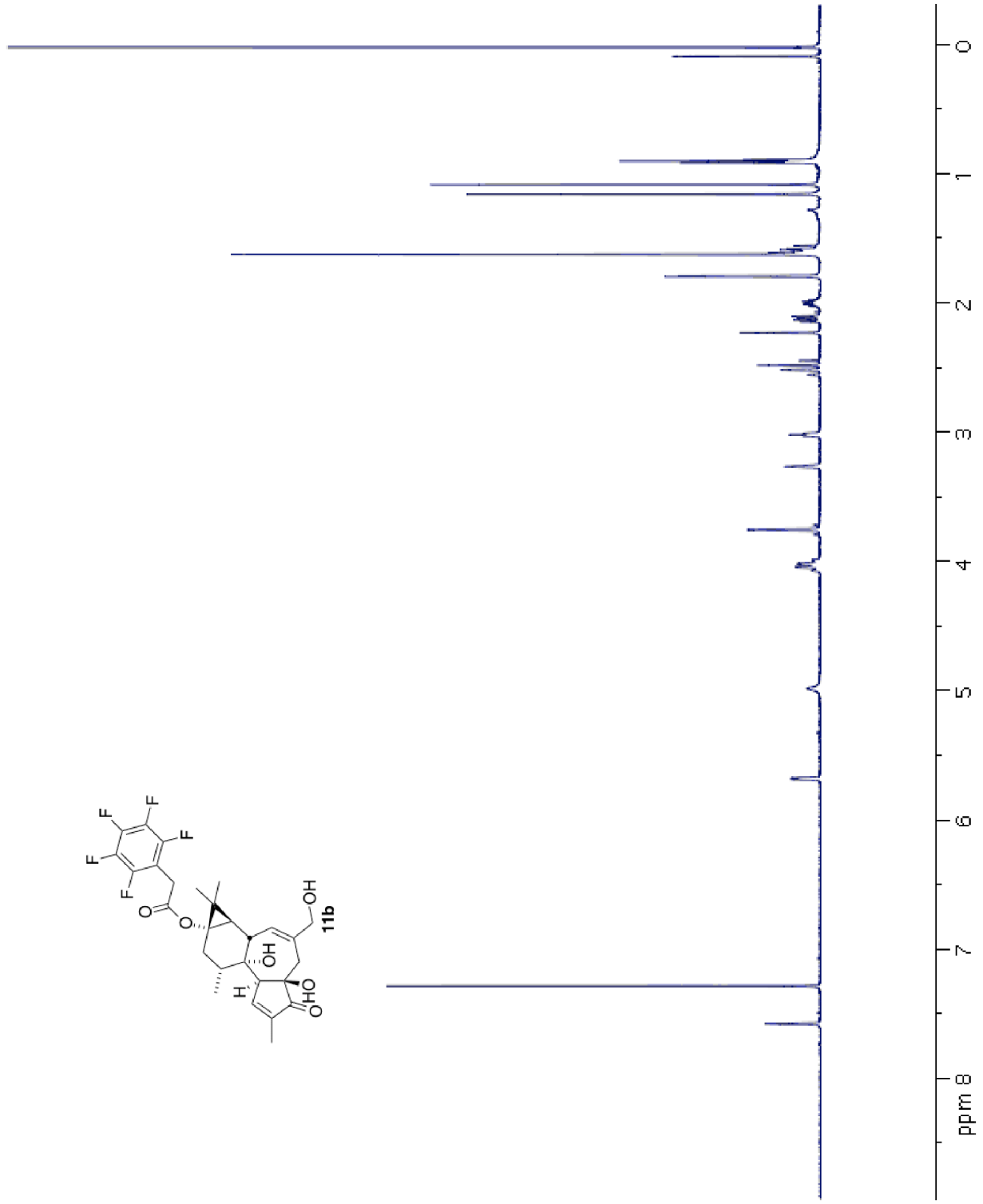
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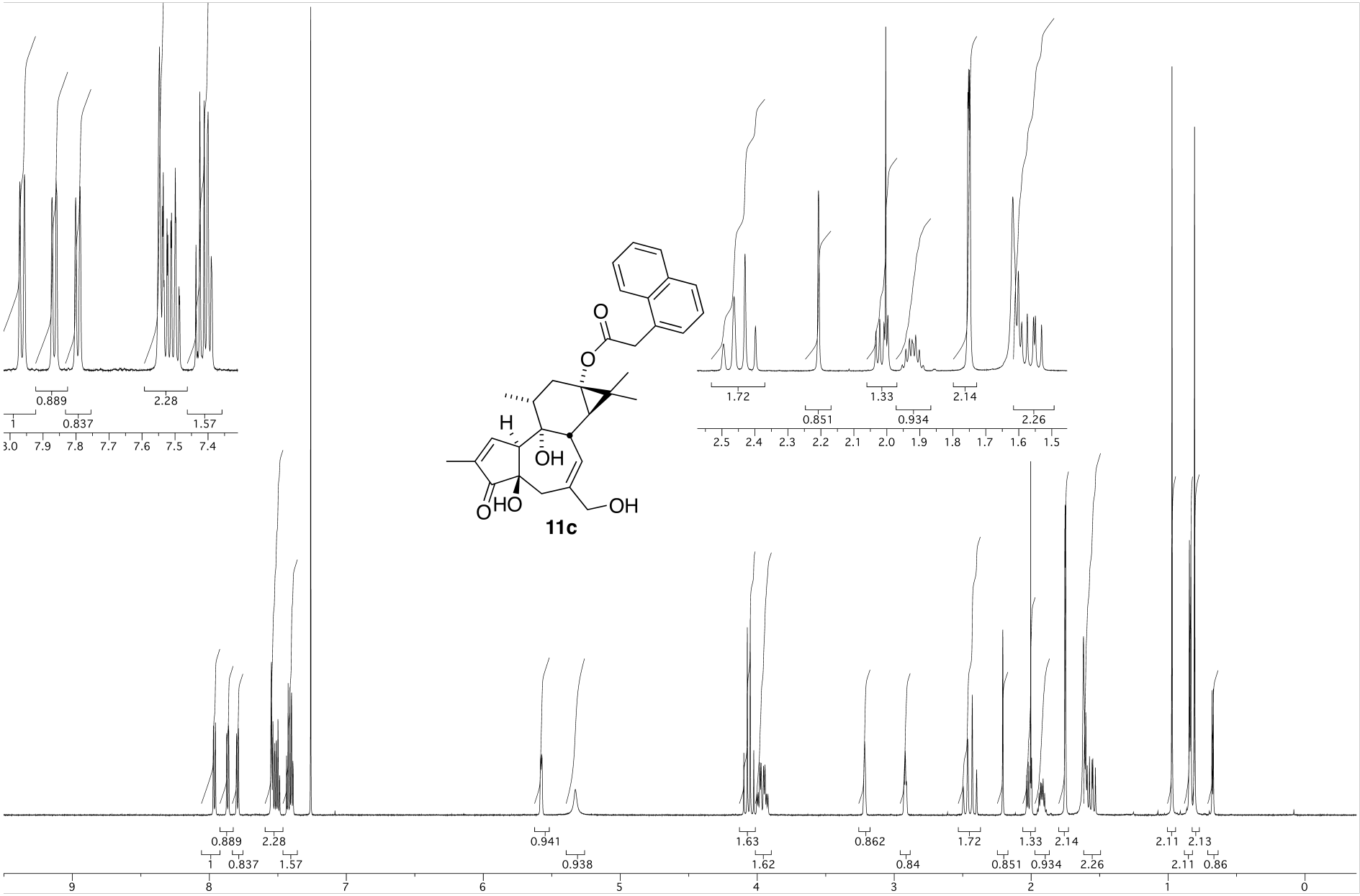
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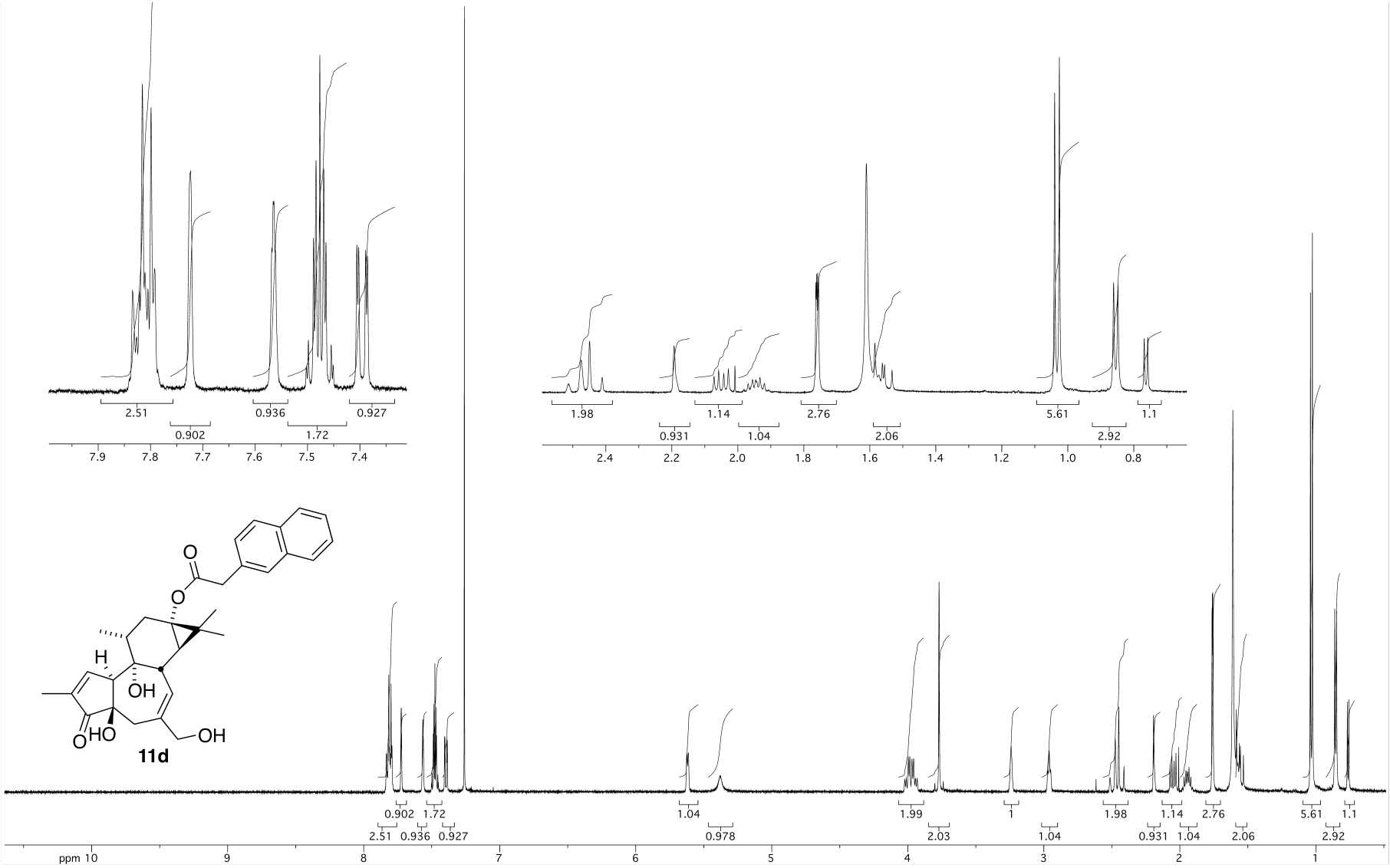
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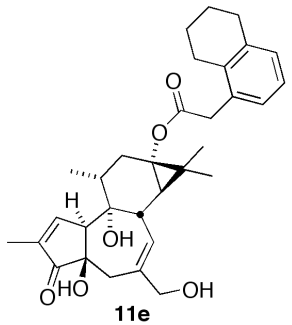
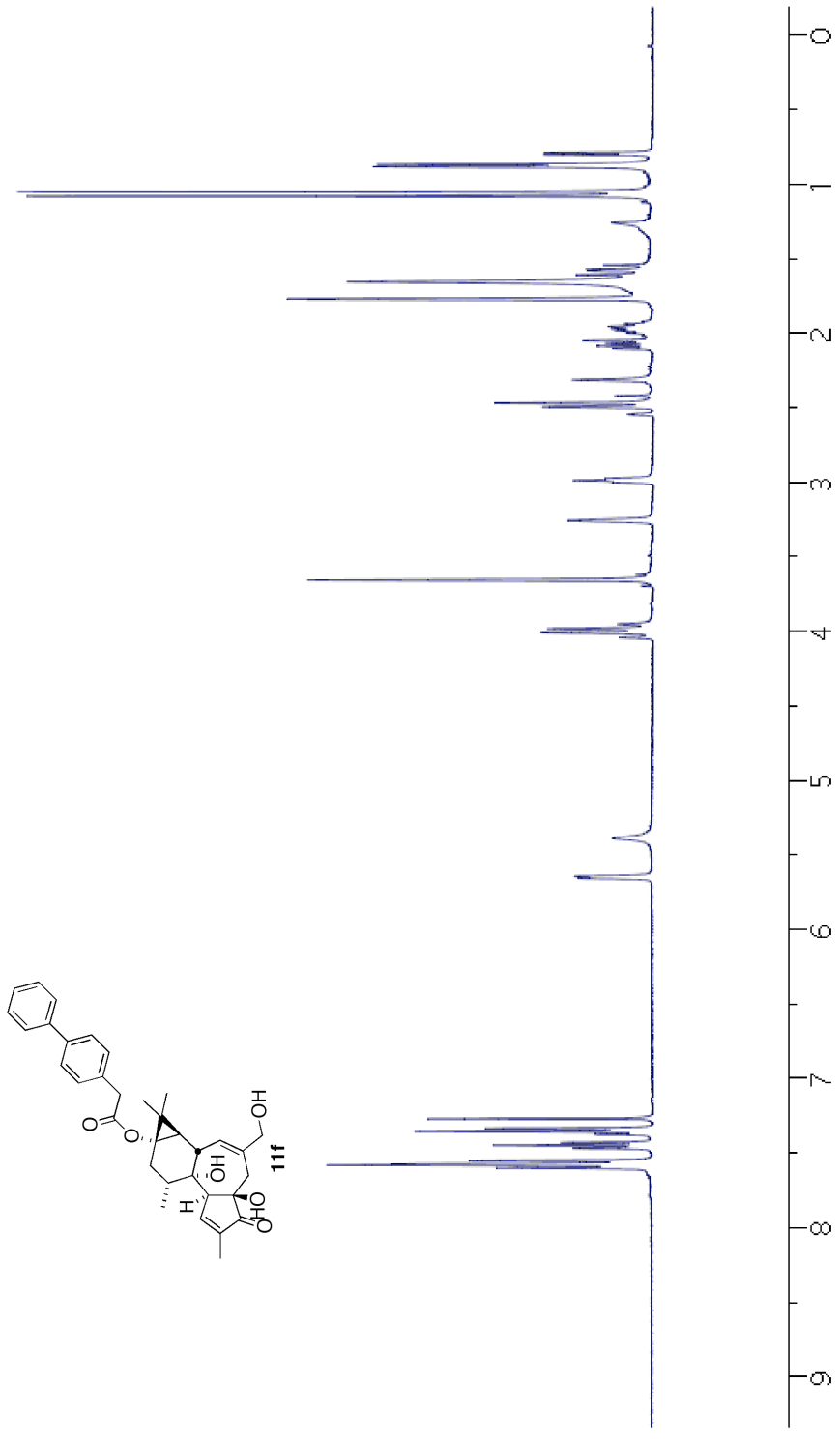


11e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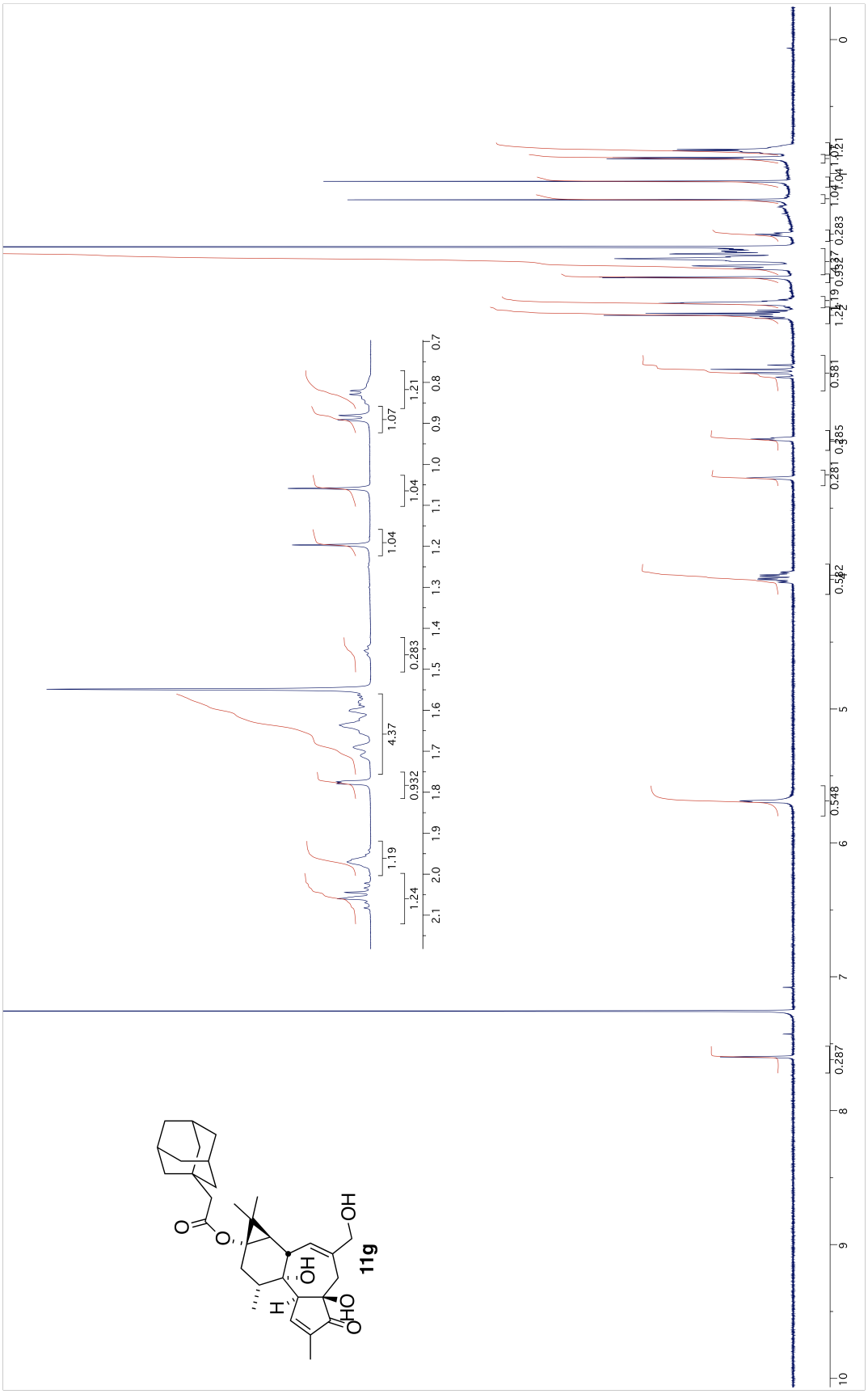
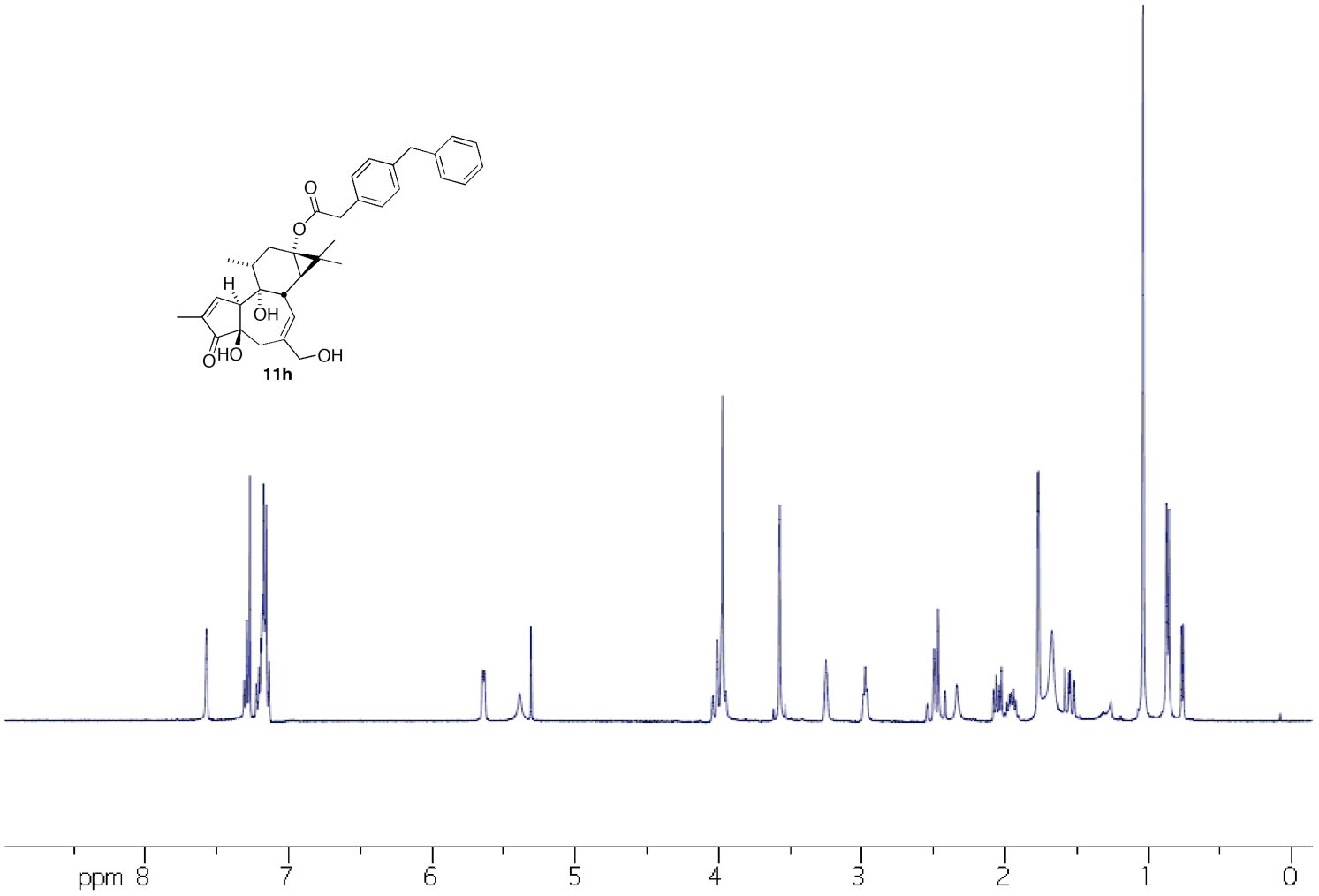

11g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References

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