Supporting information for

Synthesis, Anti-inflammatory and Pro-Resolving Activities of 22-OH-PD1, a Mono-Hydroxylated Metabolite of Protectin D1

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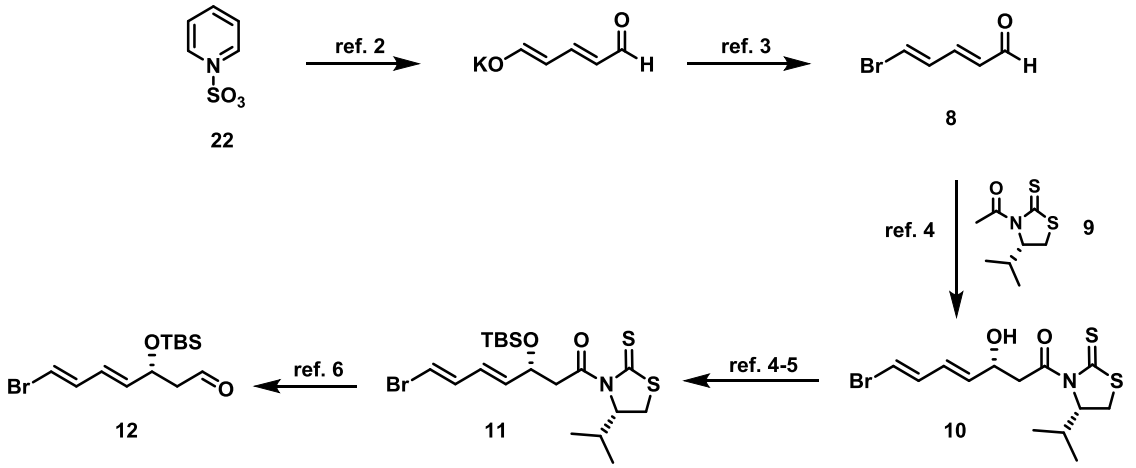
General Information

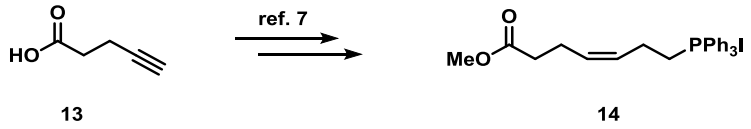
Unless stated otherwise, all commercially available reagents and solvents were used in the form they were supplied without any further purification. The stated yields are based on isolated material. Optical rotations were measured using a 1 mL cell with a 1.0 dm path length on a Perkin Elmer 341 polarimeter. The UV/Vis spectra from 190-900 nm were recorded using a Biochrom Libra S32PC spectrometer using quartz cuvettes. IR spectra (4000-600 cm-1) were obtained on a Perkin-Elmer Spectrum BX series FT-IR spectrophotometer. NMR spectra were recorded on a Bruker AVII400 spectrometer at 400 MHz for ^{1}H NMR, and at 101 MHz for ^{13}C NMR. Spectra are referenced relative to the central residual protium solvent resonance in ^{1}H NMR ($CDCl_3$ δ = 7.27 and DMSO-d_6 = δ 2.50) and the central carbon solvent resonance in ^{13}C NMR ($CDCl_3$ δ = 77.00 ppm and DMSO-d_6 = δ 39.43). Mass spectra were recorded at 70 eV on a Waters Prospec Q spectrometer using EI, ES or CI as the method of ionization. High-resolution mass spectra were recorded on a Waters Prospec Q spectrometer using EI or ES as the methods of ionization. Thin layer chromatography was performed on silica gel 60 F254 aluminum-backed plates fabricated by Merck. Flash column chromatography was performed on silica gel 60 (40-63 μm) produced by Merck. HPLC analyses for chemical purities were performed on an Agilent Technologies 1200 Series instrument with diode array detector set at 254 nm and equipped with a C18 stationary phase (Eclipse XDB-C18 5 μm 4.6 × 150 mm), applying the conditions stated. Diastereomeric ratios reported in this paper have not been validated by calibration; see Wernerova and Hudlicky for discussions and guidelines.[1]

(*R*,4*E*,6*E*)-7-Bromo-3-((*tert*-butyldimethylsilyl)oxy)hepta-4,6-dienal (12).



Aldehyde **12** was prepared in 5 steps from commercially available pyridinium-1-sulfonate **22** as previously reported,[2-6] in 29% overall yield. All spectroscopic and physical data were in full agreement with those reported in the literature.[6] $[\alpha]_D^{20}$ = 32 (c = 0.2, $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$) δ 9.75 (t, *J* = 2.2 Hz, 1H), 6.69 (dd, *J* = 13.4, 10.8 Hz, 1H), 6.33 (d, *J* = 13.6 Hz, 1H), 6.16 (ddd, *J* = 15.2, 10.6, 1.3 Hz, 1H), 5.75 (ddd, *J* = 15.3, 5.9, 0.8 Hz, 1H), 4.66 (dd, *J* = 6.8, 5.5 Hz, 1H), 2.75 – 2.41 (m, 2H), 0.87 (s, 9H), 0.06 (s, 3H), 0.04 (s, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 201.2, 136.6, 136.1, 127.6, 109.6, 68.5, 51.4, 25.9, 14.3, -4.2, -4.9.

(*Z*)-(7-Methoxy-7-oxohept-3-en-1-yl)triphenylphosphonium iodide (14).



Wittig salt **14** was prepared in five steps from commercially available pent-4-ynoic acid **13** as previously reported,[7] in 42% overall yield. All spectroscopic and physical data were in full agreement with those reported in the literature.[8] ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.93 – 7.74 (m, 15H), 5.49 – 5.36 (m, 2H), 3.69 – 3.58 (m, 2H), 3.56 (s, 3H), 2.37 – 2.26 (t, *J* = 7.2 Hz, 4H), 2.12 (q, *J* = 7.0 Hz, 2H); ^{13}C NMR (101 MHz, DMSO) δ 172.7, 134.9 (d, $^4J_{CP}$ = 3.1 Hz, 3C), 133.6 (d, $^3J_{CP}$ = 10.2 Hz, 6C), 130.2 (d, $^2J_{CP}$ = 12.4 Hz, 6C), 127.7, 127.45, 118.3 (d, $^1J_{CP}$= 85.7 Hz, 3C), 51.3, 32.8, 22.2, 20.3 (d, $^1J_{CP}$ = 48.2 Hz), 19.7 (d, $^2J_{CP}$ = 3.5 Hz).

Methyl (*R*,4*Z*,7*Z*,11*E*,13*E*)-14-bromo-10-((*tert*-butyldimethylsilyl)oxy)tetradeca-4,7,11,13-tetraenoate (15).

The Wittig salt **14** (581 mg, 1.04 mmol, 1.0 equiv.) in THF (9.5 mL) was added mol. sieves and HMPA (1.5 mL) before NaHMDS (0.6 M in toluene, 1.0 equiv.) was slowly added at -78 °C and then stirred for 5 min at 0 °C. Aldehyde **12** (prepared from DIBAL-H reduction of **11** as described above) was added at -78 °C. The solution was allowed to slowly warm up to room temperature in the dry ice/aceton bath for 24 h before it was quenched

with phosphate buffer (10 mL, pH = 7.2). Et_2O (15 mL) was added and the phases were separated. The aqueous phase was extracted with Et_2O (2 x 15 mL) and the combined organic layers were dried (Na_2SO_4), before concentrated *in vacuo*. The crude product was purified by column chromatography on silica (hexanes/EtOAc 95:5) to afford the title compound **15** as a yellow oil. Yield: 217 mg (47% for two steps); $[\alpha]_D^{20}$ = -9.4 (c = 0.1, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 6.68 (dd, *J* = 13.4, 10.9 Hz, 1H), 6.27 (d, *J* = 13.5 Hz, 1H), 6.09 (dd, *J* = 15.2, 10.8 Hz, 1H), 5.72 (dd, *J* = 15.2, 5.8 Hz, 1H), 5.48 – 5.32 (m, 4H), 4.16 (q, *J* = 6.0 Hz, 1H), 3.67 (s, 3H), 2.84 – 2.73 (m, 2H), 2.38 – 2.35 (m, 4H), 2.35 – 2.21 (m, 2H), 0.89 (s, 9H), 0.05 (s, 3H), 0.02 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.6, 138.0, 137.1, 129.9, 129.4, 128.0, 126.7, 125.6, 108.3, 72.6, 51.7, 36.3, 34.1, 26.0 (3C), 25.9, 23.0, 18.4, -4.4, -4.6. HRESTOFMS *m/z* 465.1436 $[M+Na]^+$ (calcd for $C_{21}H_{35}O_3Si^{79}BrNa$, 465.1436); TLC (hexanes/EtOAc 95:5, CAM stain): R_f = 0.29.

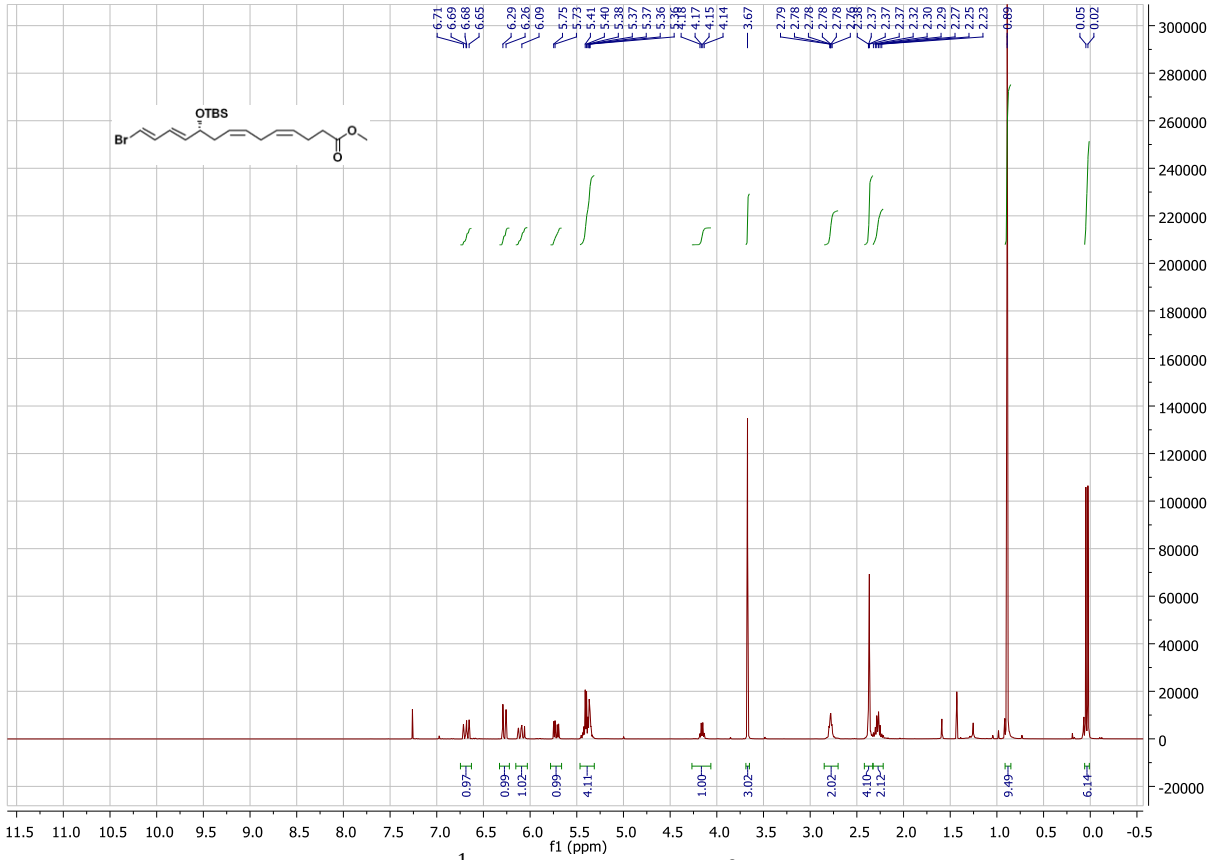


Figure S-1 ^{1}H-NMR spectrum of compound **15**.

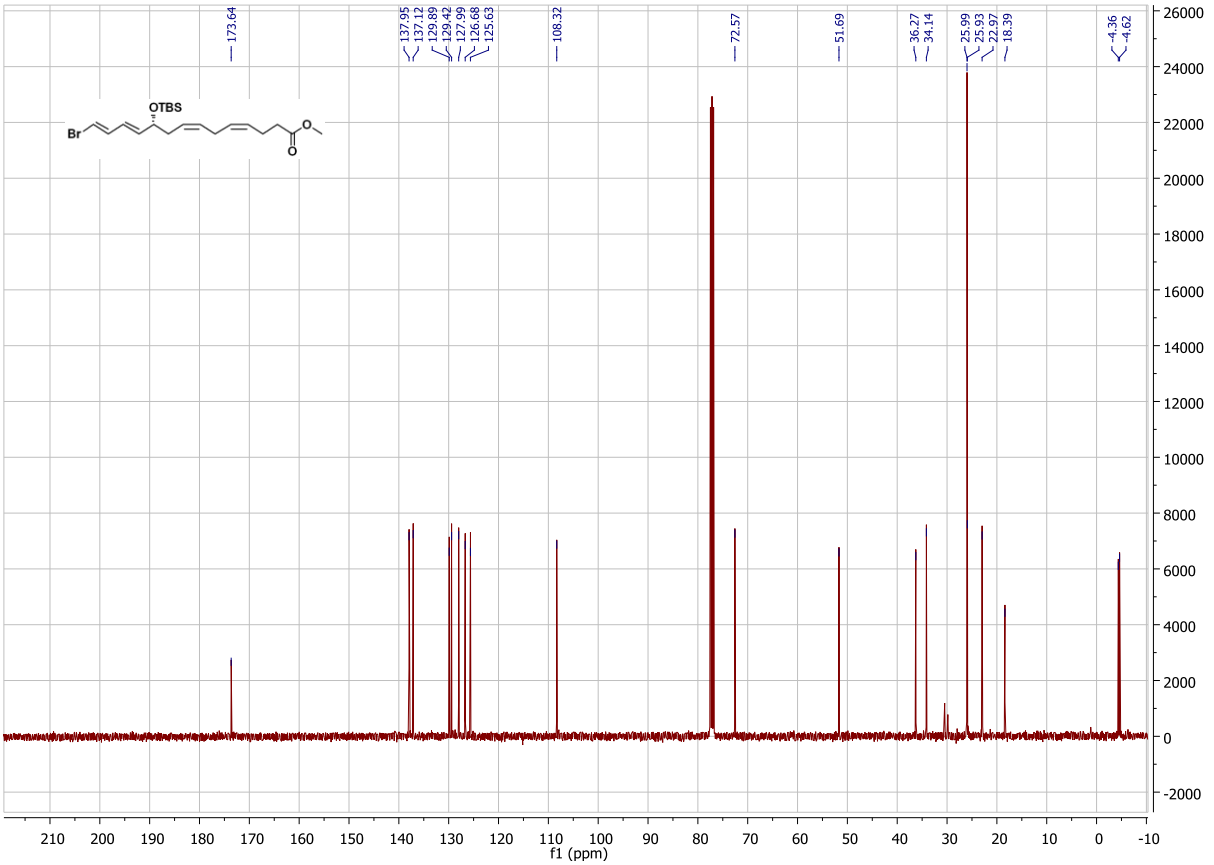


Figure S-2 ^{13}C-NMR spectrum of compound **15**.

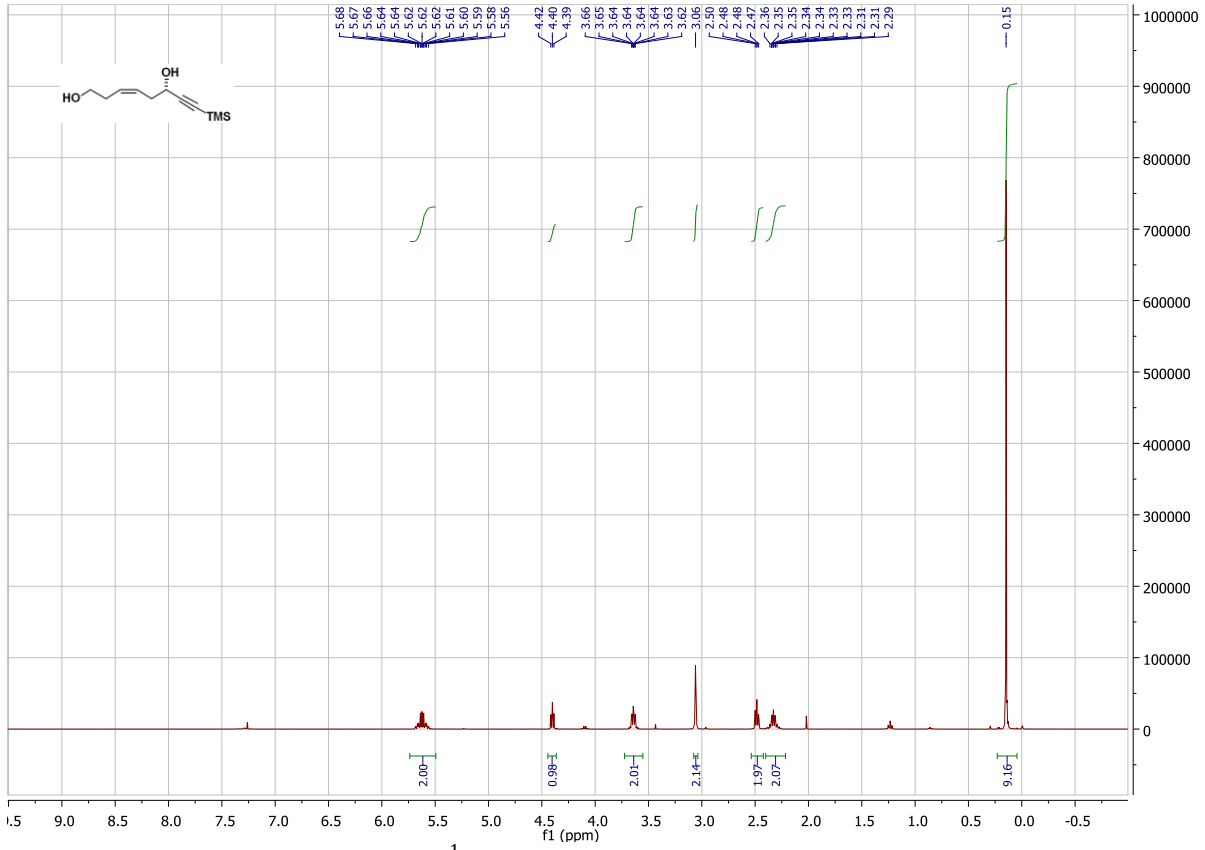


Figure S-3 ^{1}H-NMR spectrum of compound **18**.

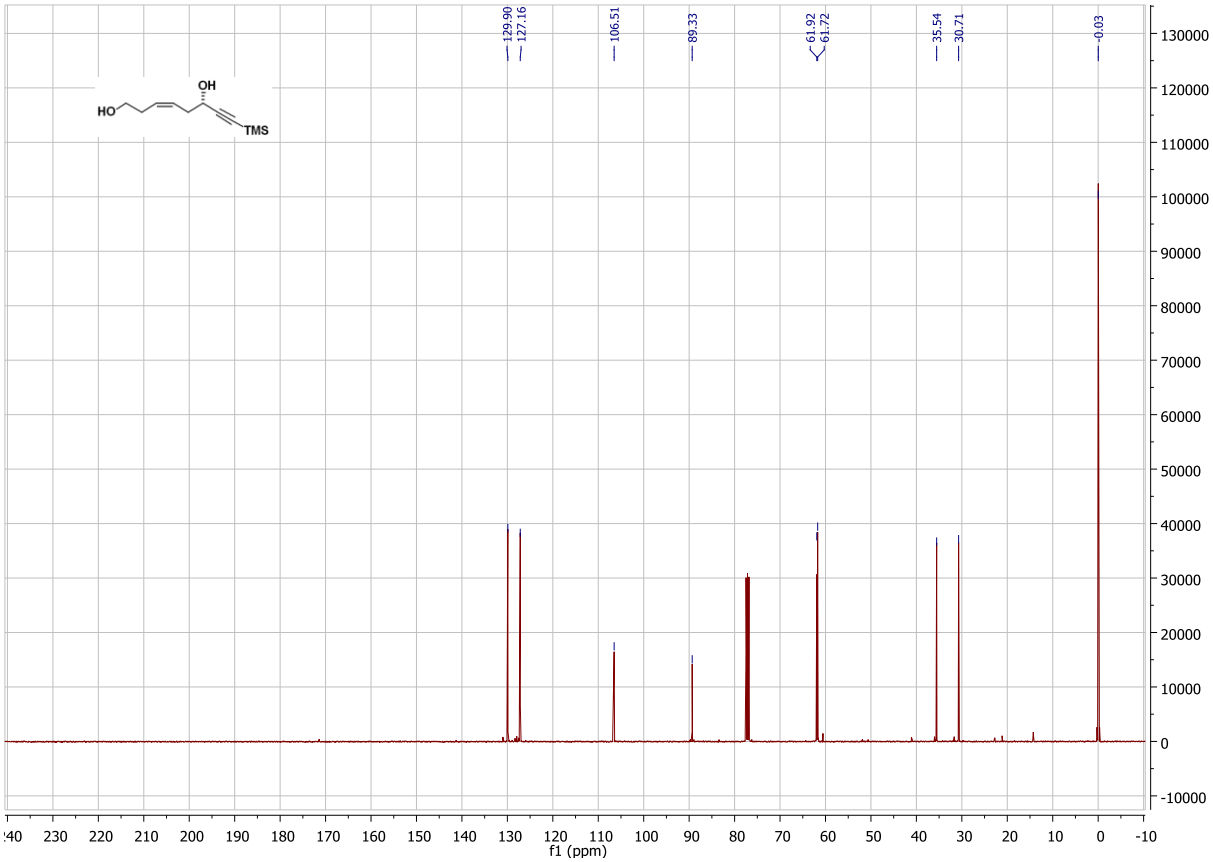


Figure S-4 ^{13}C-NMR spectrum of compound **18**.

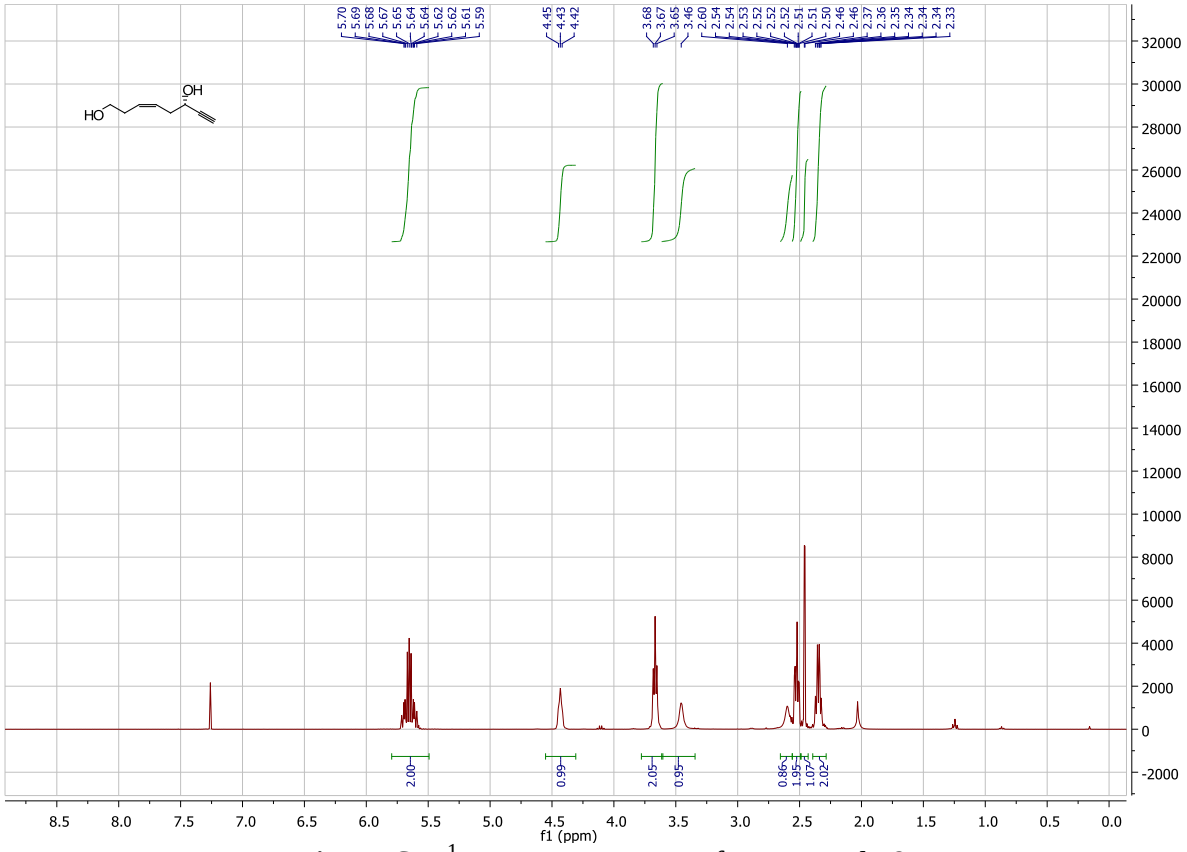


Figure S-5 ^{1}H-NMR spectrum of compound **19**.

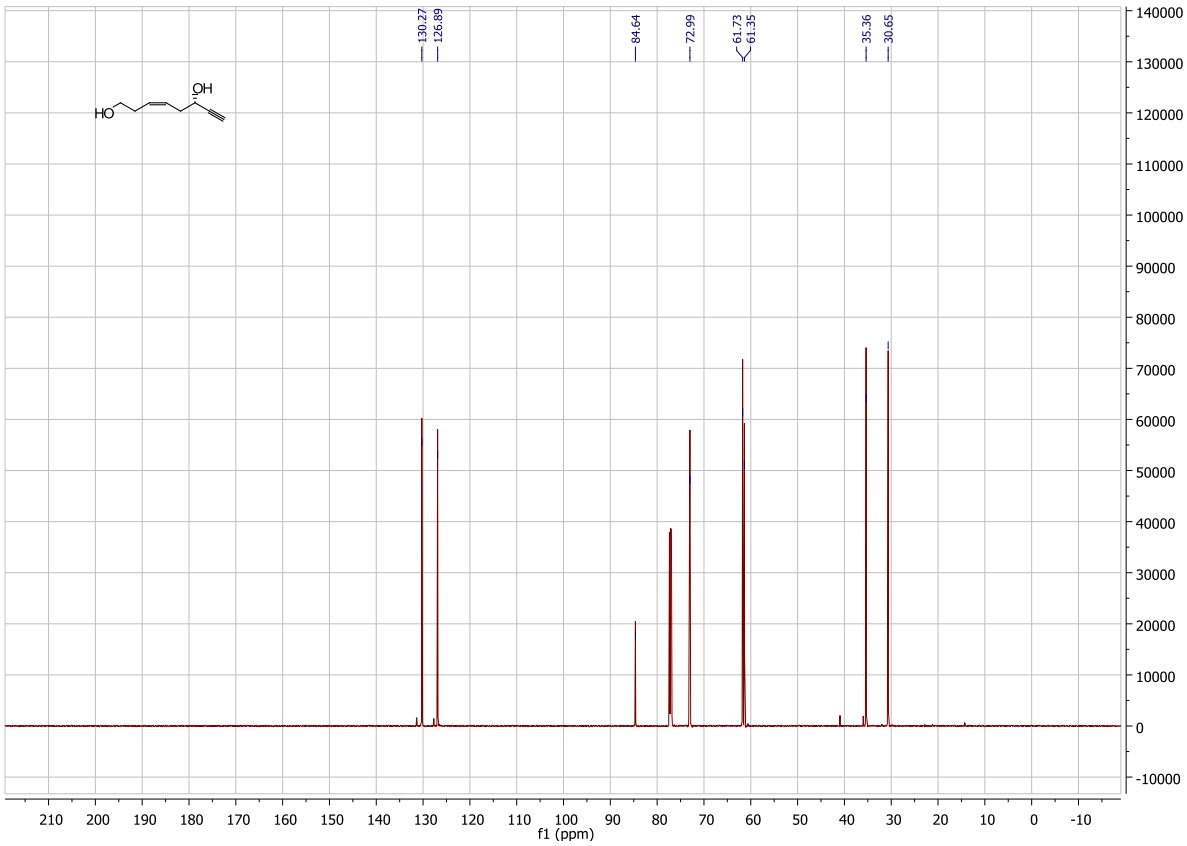


Figure S-6 ^{13}C-NMR spectrum of compound **19**.

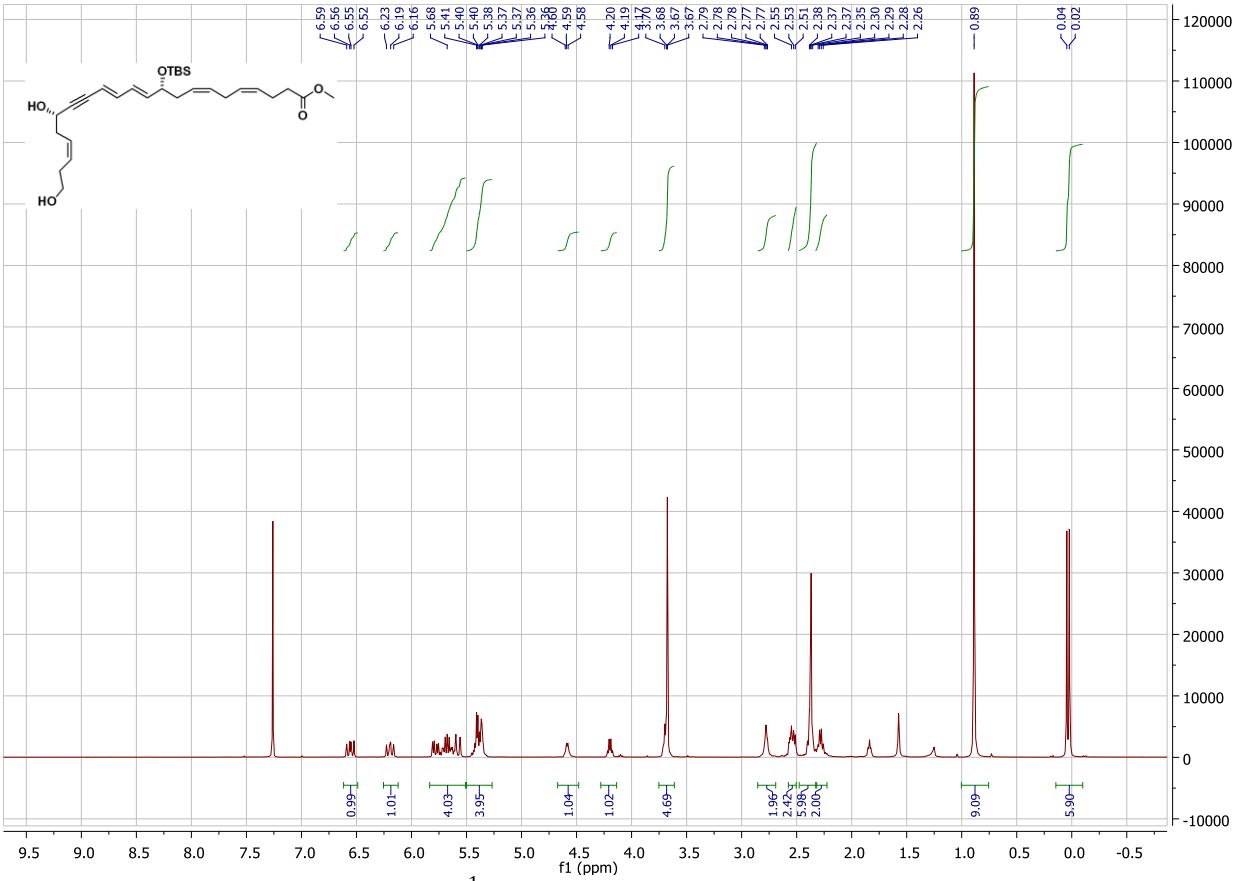


Figure S-7 ^{1}H-NMR spectrum of compound **20**.

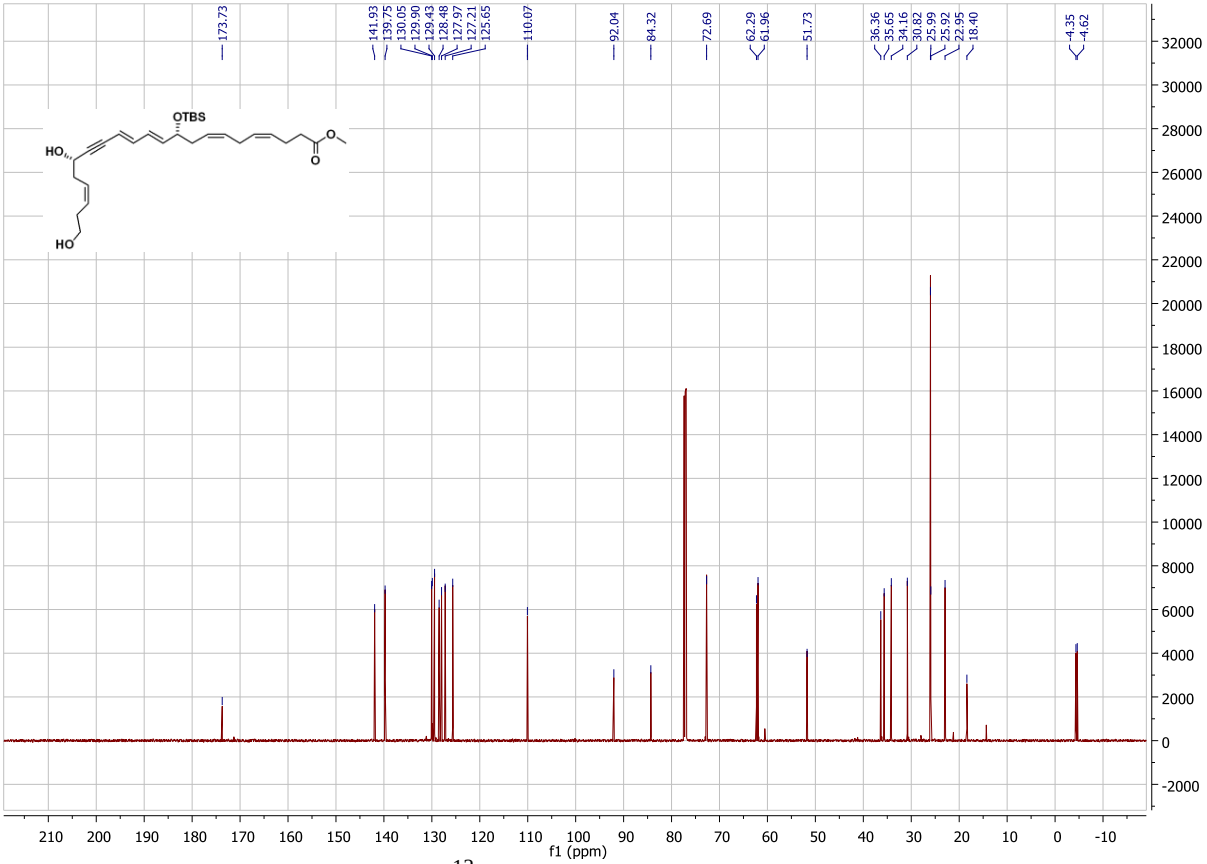


Figure S-8 ^{13}C-NMR spectrum of compound **20**.

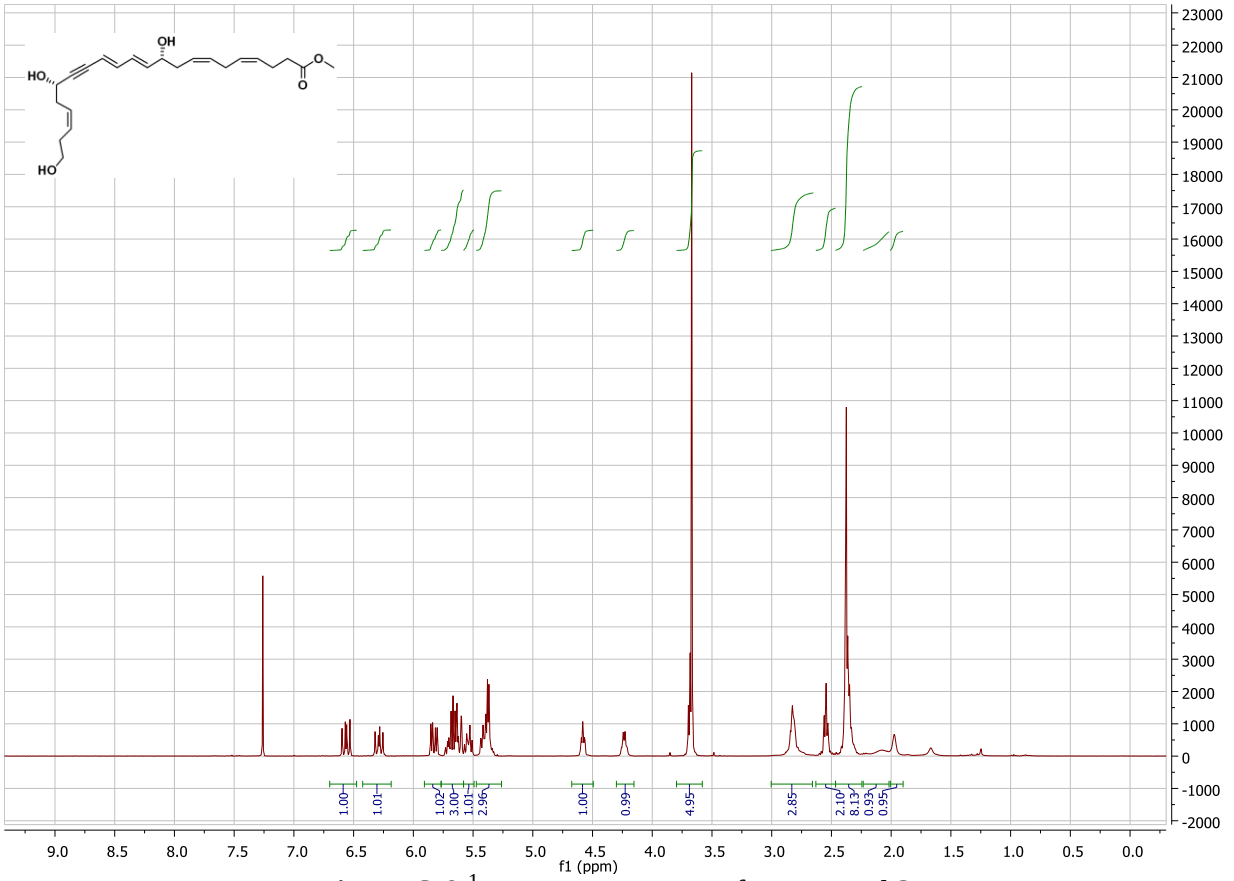


Figure S-9 ^{1}H-NMR spectrum of compound **21**.

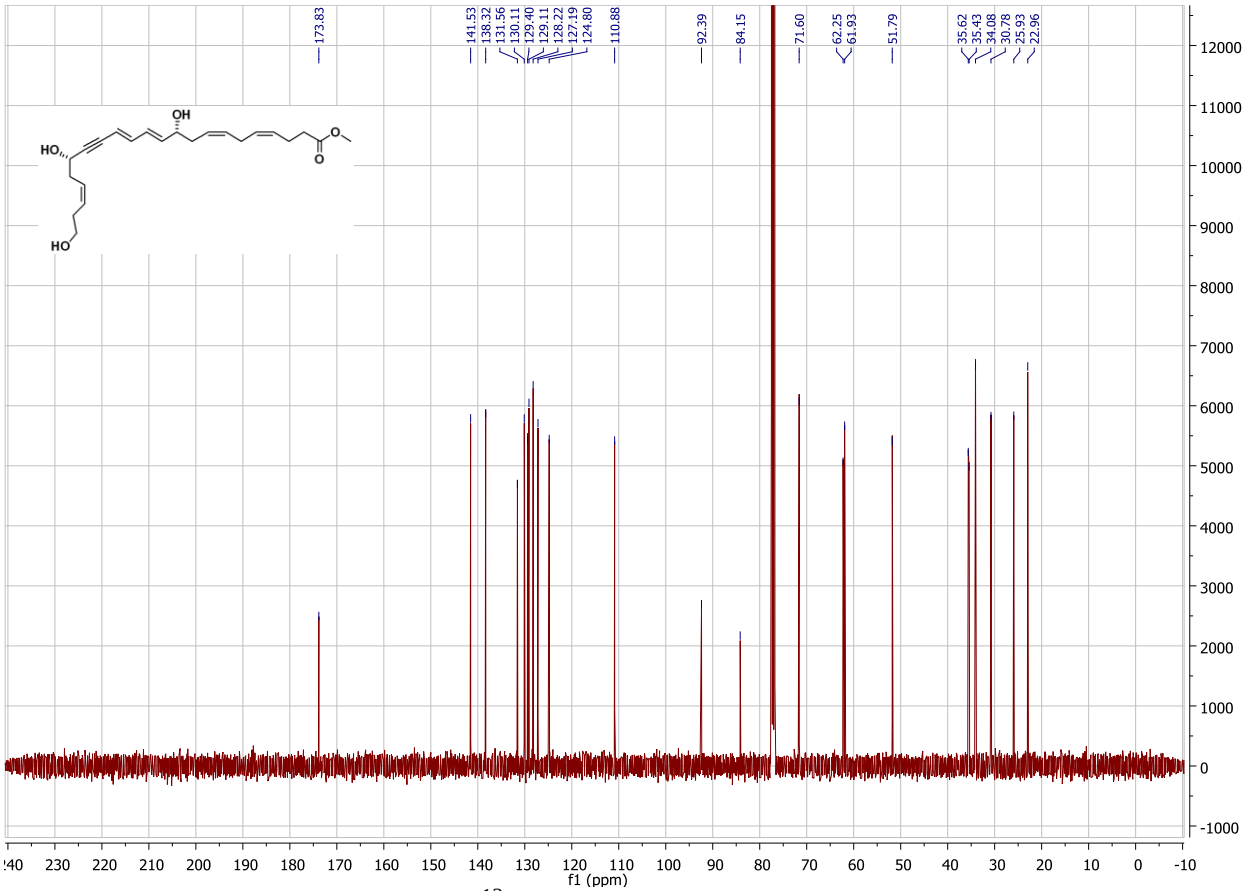


Figure S-10 ^{13}C-NMR spectrum of compound **21**.

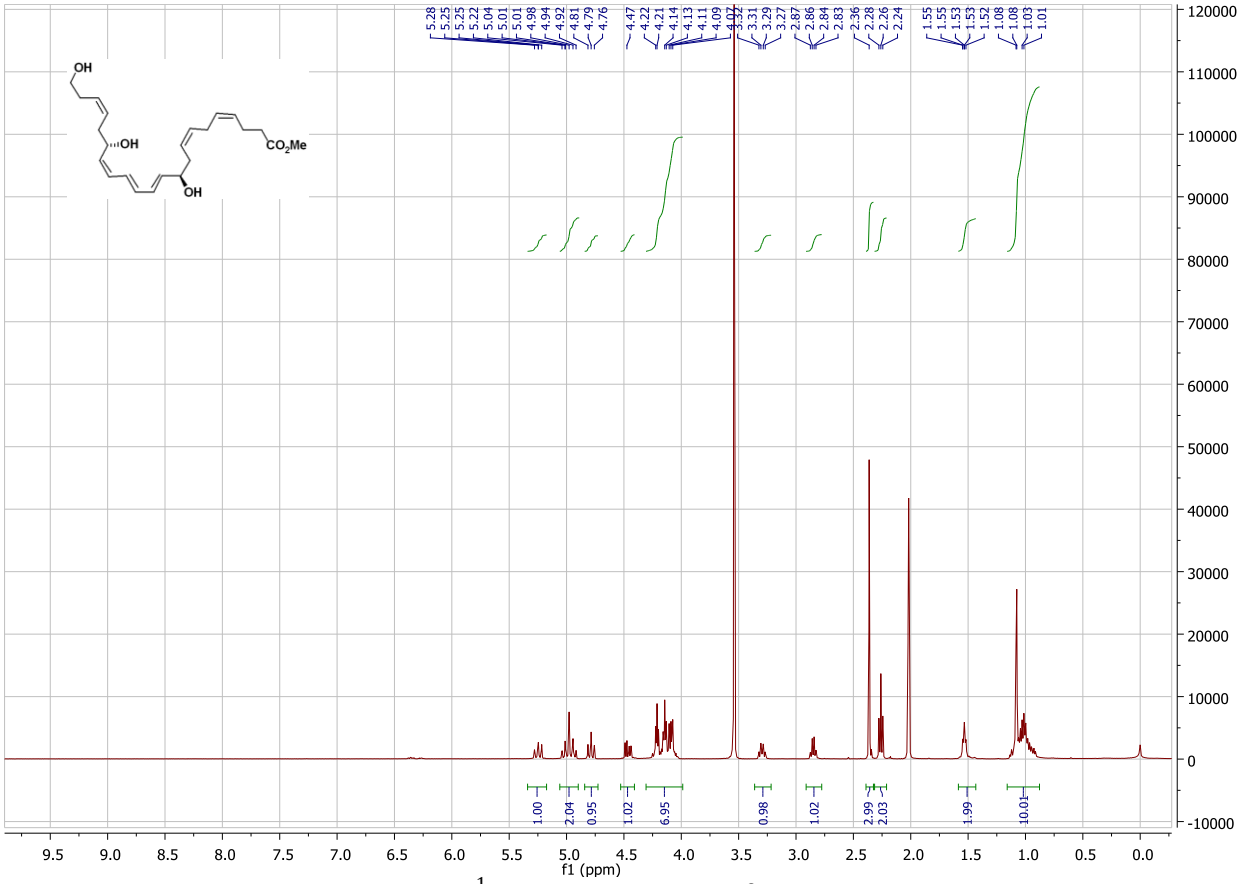


Figure S-11 ^{1}H-NMR spectrum of methyl ester **7**.

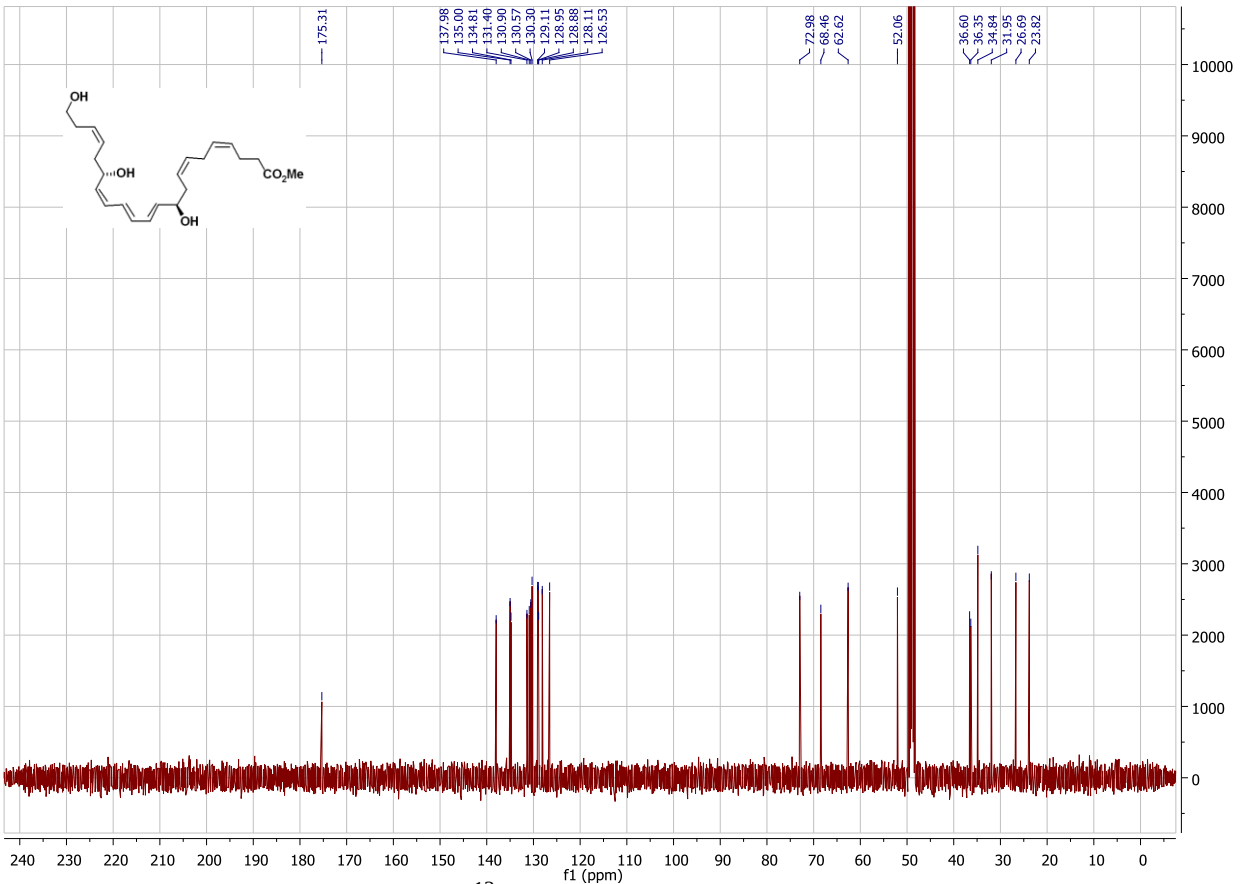


Figure S-12 ^{13}C-NMR spectrum of methyl ester **7**.

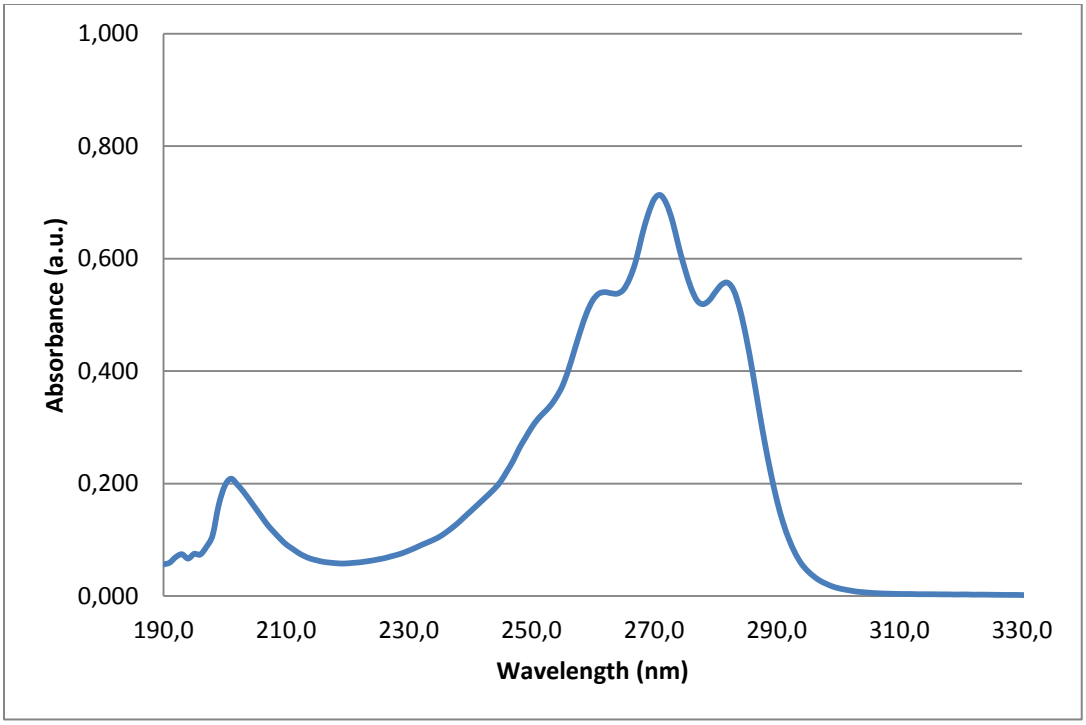
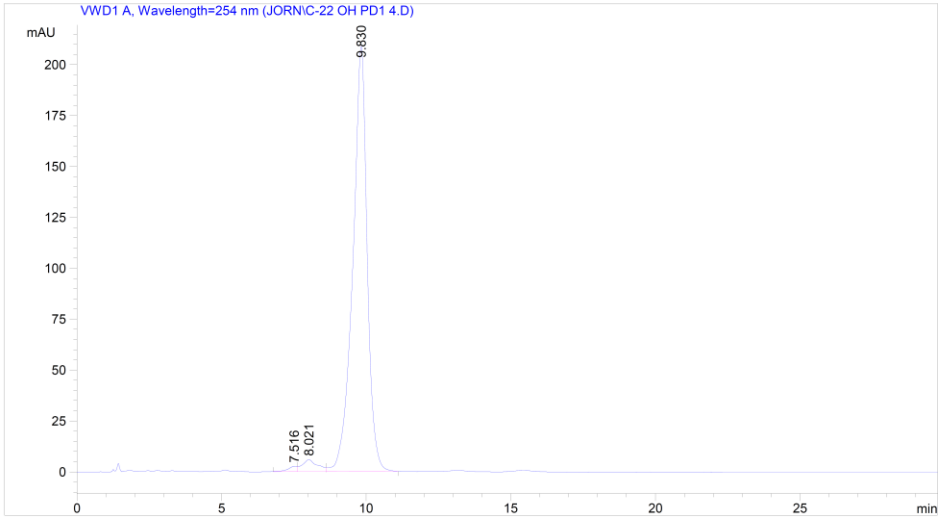


Figure S-13 UV-Vis chromatogram of methyl ester **7**.

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                    Area Percent Report
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Sorted By             :      Signal
Multiplier            :      1.0000
Dilution              :      1.0000
Sample Amount         :      1.00000  [ng/ul]   (not used in calc.)
Use Multiplier & Dilution Factor with ISTDs


Signal 1: VWD1 A, Wavelength=254 nm

Peak RetTime Type  Width     Area      Height     Area
  #   [min]       [min]  mAU   *s    [mAU   ]      %
----|-------|----|-------|----------|----------|--------|
   1   7.516 BV    0.3200   57.89385    2.60974   0.7711
   2   8.021 VV    0.5010  217.98421    5.87976   2.9033
   3   9.830 VB    0.4942 7232.14893  208.86168  96.3256

Totals :                  7508.02699  217.35118

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Figure S-14 HPLC chromatogram of methyl ester **7**.

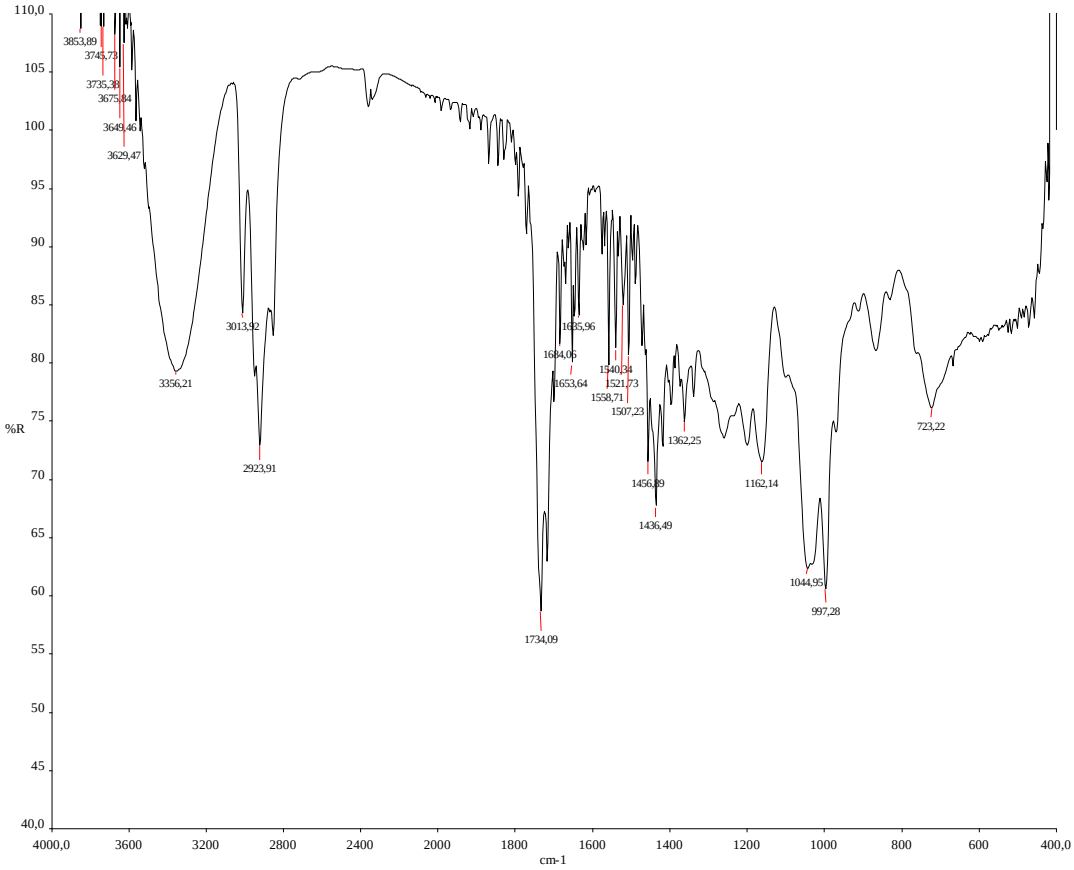

Figure S-15 IR spectrum of methyl ester **7**.

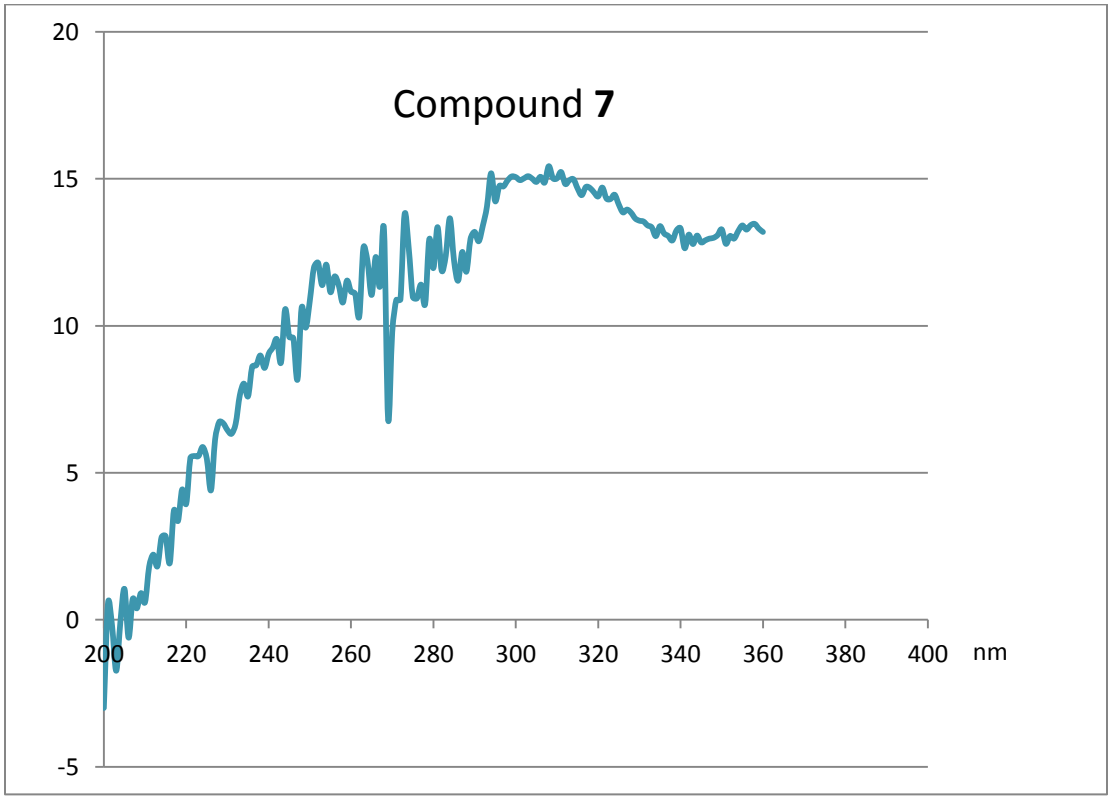

Figure S-16 CD spectrum of methyl ester **7**.

References

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