Supplemental Data

A new ultrasensitive bioluminescence-based method for assaying Monoacylglycerol lipase

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^{1}H and ^{13}C-NMR data

Instruments

^{1}H-NMR spectra were recorded in $CDCl_3$ (isotopic enrichment 99.95%) solutions at 300 K using a Bruker AVANCE 500 instrument (500.13 MHz for ^{1}H, 125.76 MHz for ^{13}C) using 5 mm inverse detection broadband probes and deuterium lock. Chemical shifts (d) are given as parts per million relative to the residual solvent peak (7.26 ppm for ^{1}H and 77.0, central line, for ^{13}C) and coupling constants (*J*) are in Hertz. The experimental error in the measured ^{1}H-^{1}H coupling constants is ± 0.5 Hz. The splitting pattern abbreviations are as follows: s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet, and bs, broad peak. For two-dimensional experiments, Bruker microprograms using gradient selection (gs) were applied. Acquisition parameters for 1D were as follows: ^{1}H spectral width of 5000 Hz and 32 K data points providing a digital resolution of ca. 0.305 Hz per point, relaxation delay 2 s; ^{13}C spectral width of 29412 Hz and 64 K data points providing a digital resolution of ca. 0.898 Hz per point, relaxation delay 2.5 s.

For two-dimensional experiments, standard Bruker microprograms using gradient selection (gs) were applied. COSY-45 experiments were acquired with 512 t1 increments; 2048 t2 points; spectral/spectrum width 10.0 ppm. The acquisition data for HSQC and HMBC experiments were obtained with 512 t1 increments; 2048 t2 points; spectral/spectrum width 10.0 ppm for 1H and 220 ppm for 13C. Delay values were optimized for $^{1}J_{C,H}$ 140.0 Hz and $^{1}J_{C,H}$ 3.0 Hz. Zero filling in F1 to 1 K, π/2 shifted sine-bell squared (for gHSQC) or sinebell (for gHMBC) apodization functions were used for processing.

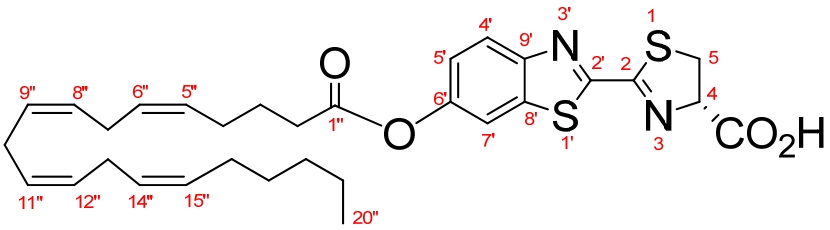
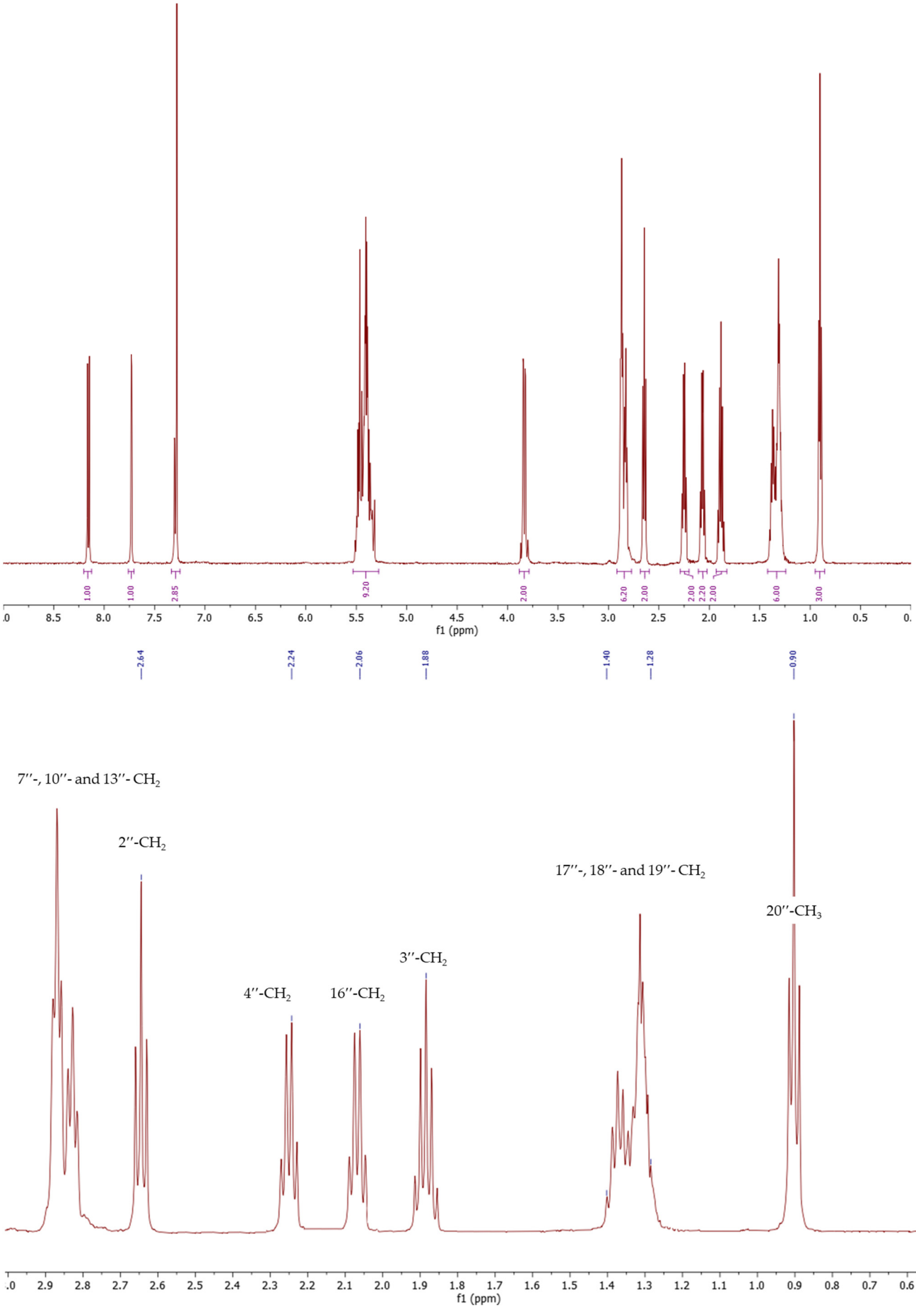
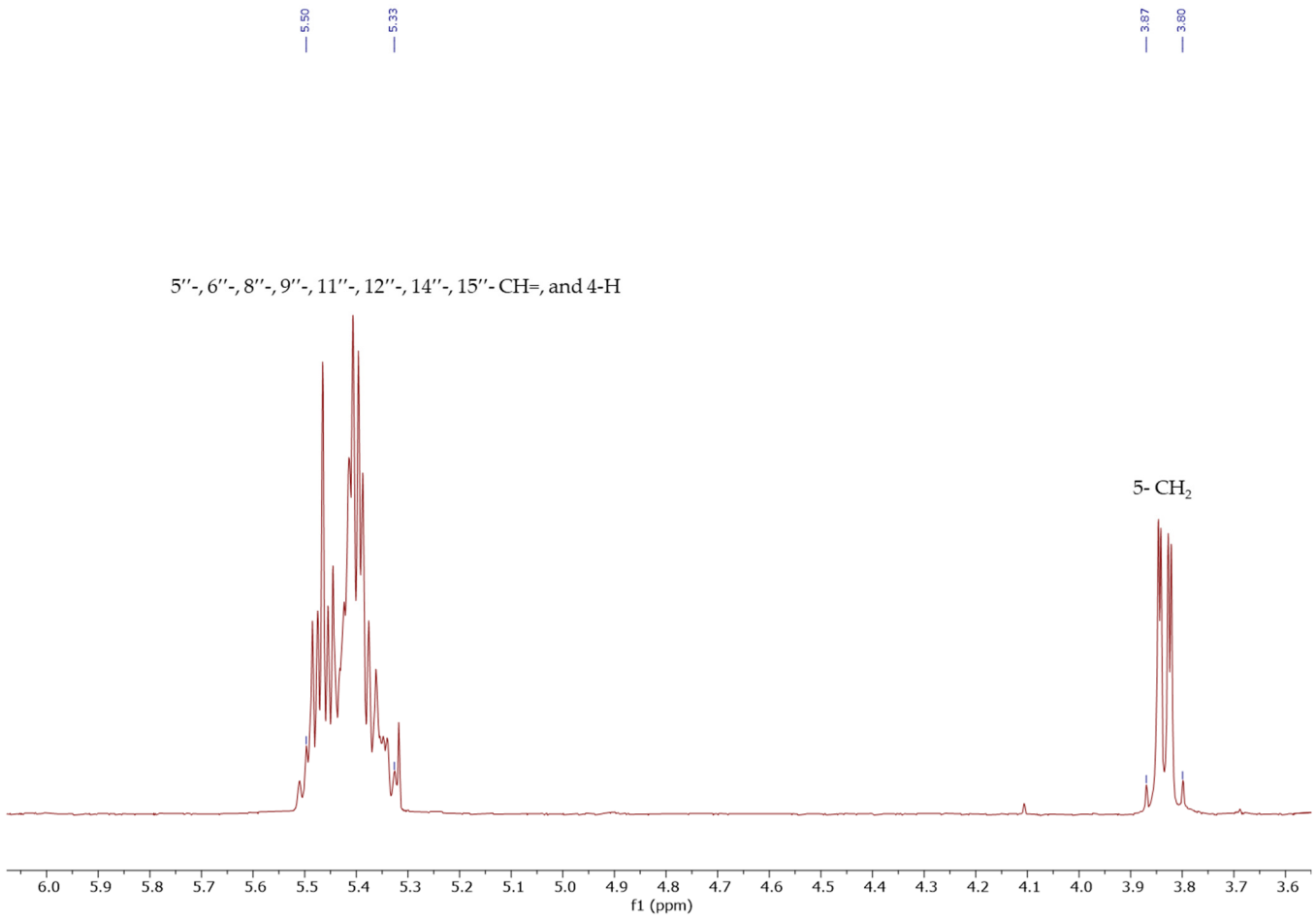
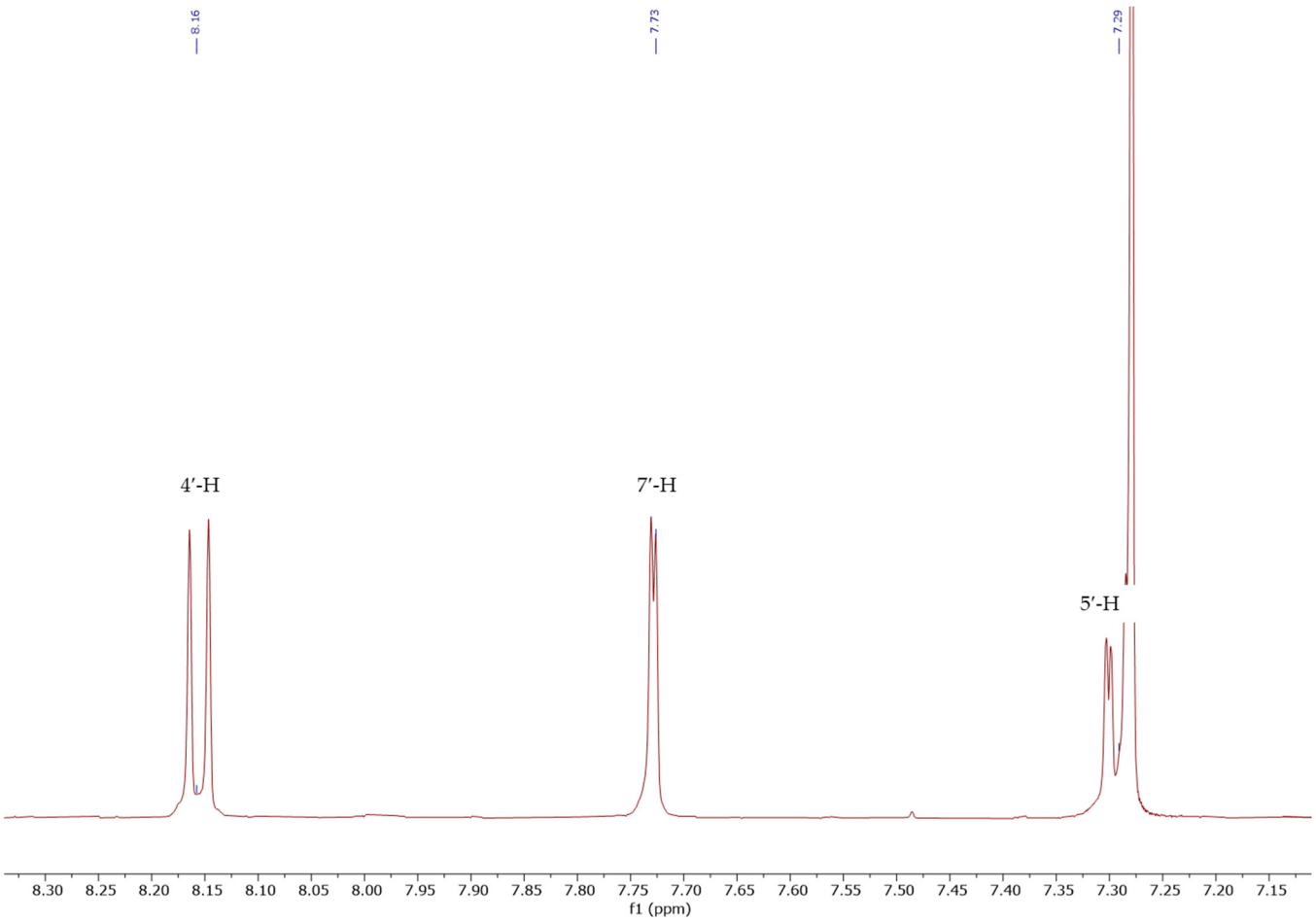


Figure S1. IUPAC numbering of 6-*O*-arachidonoylluciferin

^{1}H spectrum of 6-*O*-arachidonoylluciferin (**1**)

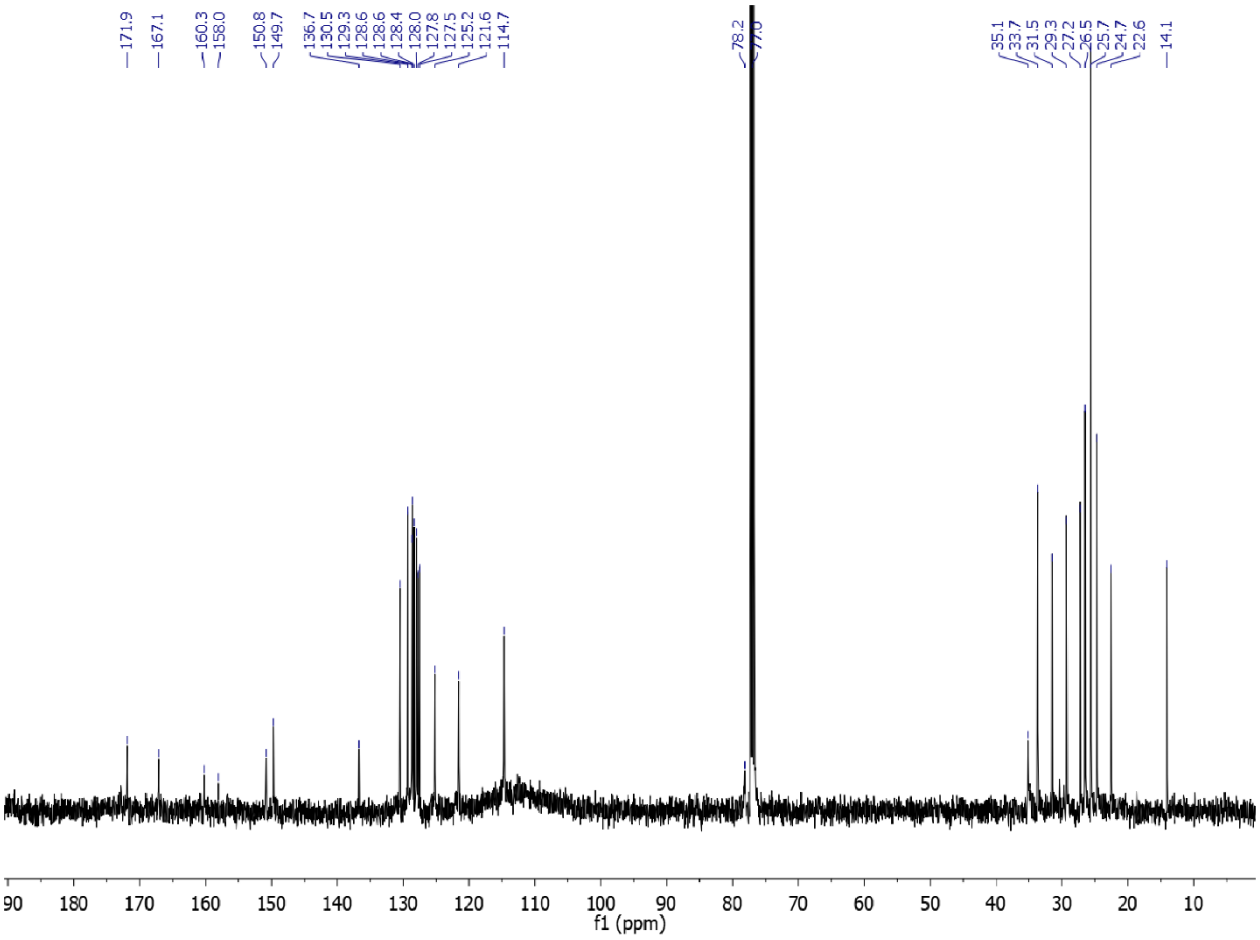

1.00
1.00
2.85
9.20
2.00
6.20
2.00
2.00
2.20
2.00
6.00
3.00
f1 (ppm)
2.64
2.24
2.06
1.88
1.40
1.28
0.90
7''-, 10''- and 13''- CH2
2''-CH2
4''-CH2
16''-CH2
3''-CH2
17''-, 18''- and 19''- CH2
20''-CH3
f1 (ppm)


5.50
5.33
3.87
3.80
5''-, 6''-, 8''-, 9''-, 11''-, 12''-, 14''-, 15''- CH=, and 4-H
5- CH2
6.0 5.9 5.8 5.7 5.6 5.5 5.4 5.3 5.2 5.1 5.0 4.9 4.8 4.7 4.6 4.5 4.4 4.3 4.2 4.1 4.0 3.9 3.8 3.7 3.6
f1 (ppm)


8.16
7.73
7.29
4′-H
7′-H
5′-H
8.30 8.25 8.20 8.15 8.10 8.05 8.00 7.95 7.90 7.85 7.80 7.75 7.70 7.65 7.60 7.55 7.50 7.45 7.40 7.35 7.30 7.25 7.20 7.15
f1 (ppm)

^{1}H NMR ($CDCl_3$, 500 MHz): δ (ppm) 0.90 (3H, t, J=7.6, 20′′-CH_3), 1.28-1.40 (6 H, m, 17′′-, 18′′-, 19′′- CH_2), 1.88 (2H, tt, J=7.0, 7.6 Hz, 3′′-CH_2), 2.06 (2H, dt, J=7.6, 7.6 Hz, 16′′-CH_2), 2.24 (2H, dt, J=7.0, 7.4 Hz, 4′′-CH_2), 2.64 (2H, t, J=7.0 Hz, 2′′-CH_2), 2.82-2.88 (6H, m, 7′′- 10′′- and 13′′-CH_2), 3.80-3.87 (2H, AB part of ABX system, 5a- and 5b-H), 5.32-5.50 (9H, m, 5′′- 6′′- 8′′- 9′′- 11′′- 12′′- 14′′- 15′′-CH and 4-H), 7.29 (1H, dd, J=2.3, 8.9 Hz, 5′-H), 7.73 (1H, d, J=2.3 Hz, 7′-H), 8.16 (1H, d, J=8.9 Hz, 4′-H).

^{13}C NMR spectrum of 6-*O*-arachidonoylluciferin (**1**)



^{13}C NMR ($CDCl_3$, 125.76 MHz): δ (ppm) 14.1 (20′′), 22.6 (19′′), 24.7 (3′′), 25.7 (7′′, 10′′, 13′′), 26.5 (4′′), 27.2 (16′′), 29.3 (17′′), 31.5 (18′′), 33.7 (2′′), 35.1 (5), 78.2 (4), 114.7 (7′), 121.6 (5), 125.2 (4), 127.5, 127.8, 128.0, 128.4, 128.6, 128.6, 129.3, 130.5 (5′, 6′, 8′, 9′, 11′, 12′, 14′, 15′), 136.7 (8′), 149.7 (6′), 150.8 (9′), 158.0 (2), 160.3 (2′), 167.1(1′′), 171.9 (*C*OOH).