Supporting Information

Plumisclerin A, a Diterpene with a New Skeleton from the Soft Coral *Plumigorgia terminosclera*

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Experimental Procedures

General Experimental Procedures. Optical rotations were determined in MeOH using a Jasco P-1020 polarimeter. UV spectra were obtained with a Perkin Elmer Lambda 15 UV/Vis spectrophotometer. IR spectra were measured on a Perkin Elmer Spectrum 100 FT-IR spectrometer. NMR spectra were recorded on a Varian "Unity 500" spectrometer at 500/125 MHz (^{1}H/^{13}C). Chemical shifts were reported in ppm using residual non-deuterated chloroform signals (δ 7.26 for ^{1}H and 77.0 for ^{13}C) as internal reference. Accurate mass analyses were performed by HREIMS on a VG Auto chromatograph spectrometer.

Sample Collection. Samples of *Plumigorgia terminosclera* Alderslade, 1986, were collected by SCUBA diving at Mayotte Island (12º44'45" S, 44º59' 70" E) at a depth of 25 m. A voucher specimen (ORMA026794) is deposited at PharmaMar.

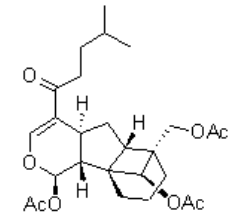
Extraction of organism: A sample (176 g) of *Plumigorgia terminosclera* was triturated and extracted with 2-propanol (4 × 500 mL). The combined extracts were concentrated to yield a crude extract of 5.42 g. This material was subjected to VLC on Lichroprep RP-18 with a stepped gradient from H_2O:MeOH 1:1 to MeOH and then to MeOH:$CHCl_3$ 1:1. The fraction eluted with MeOH was submitted to a new VLC on Lichroprep RP-18 with a stepped gradient from H_2O:MeOH 1:2 to MeOh and then to MeOH:CH_2Cl_2 1:1. Plumisclerin A (**1**) (5.0 mg) was isolated from the fraction eluting with MeOH by semipreparative HPLC (SymmetryPrep C-18, 7.8 × 150 mm, isocratic H_2O/MeCN 40:60, 3 mL/min, UV detection, t_R=16.3 min).

Plumisclerin A (1). white solid; $[\alpha]^{25}_D$ +125.0º (c 0.5, $CHCl_3$); UV (MeOH) λ_{max} 252 nm; IR (neat) ν_{max} 3016, 2970, 2949, 2869, 1738, 1435, 1366, 1228, 1217 cm^{-1}; (+)-HREIMS (m/z 476.2418 $[M]^+$ calcd for $C_{26}H_{36}O_8$ m/z 476.2405, Δ + 2.7 ppm); ^{1}H (500 MHz) and ^{13}C NMR (125 MHz) see Table 2.

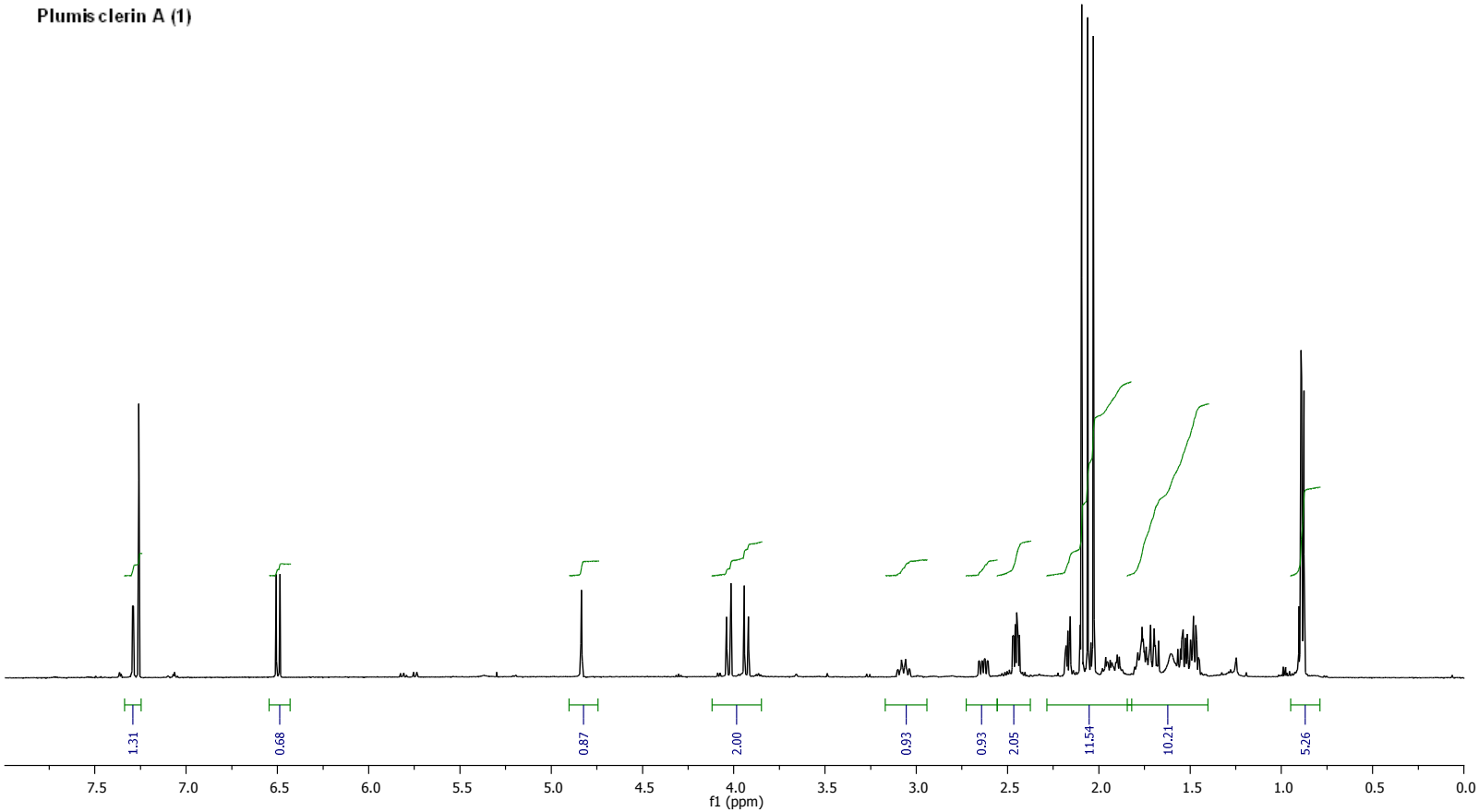
Biological Activity. A-549 (ATCC CCL-185), lung carcinoma; HT-29 (ATCC HTB-38), colorectal carcinoma and MDA-MB 231 (ATCC HTB-26), breast adenocarcinoma cell lines were obtained from the ATCC. Cell lines were maintained in RPMI medium supplemented with 10% fetal calf serum (FCS), 2 mM L-glutamine and 100 U/mL penicillin and streptomycin, at 37 ºC and 5% CO_2. Triplicate cultures were incubated for 72 h in the presence or absence of test compounds (at ten concentrations ranging from 40 to 0.01 μg/mL). For quantitative estimation of cytotoxicity, the colorimetric sulforhodamine B (SRB) method was used, essentially performed as described previously. Briefly, cells were washed twice with PBS, fixed for 15 min in 1% glutaraldehyde solution, rinsed twice in PBS, and stained in 0.4% SRB solution for 30 min at room temperature. Cells were then rinsed several times with 1% acetic acid solution and air-dried. Sulforhodamine B was then extracted in 10 mM trizma base solution and the absorbance measured at 490 nm. Results are expressed as GI_{50}, the concentration that causes 50% inhibition in cell growth after correction for cell count at the start of the experiment (NCI algorithm).

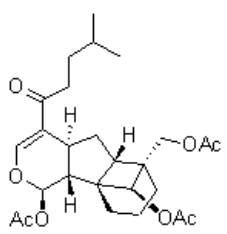
Table 2. ^{1}H and ^{13}C NMR data for Plumisclerin A (1) ($CDCl_3$).

	^{1}H, mult, *J* = Hz	^{13}C	COSY	HMBC
1	6.50, d, 9.8	95.9, d	H-11a	C-4a, C-11, C-24
3	7.29, d, 2.3	153.3, d	H-4a	C-1, C-4, C-4a, C-13
4		121.9, s		
4a	3.07, m	38.2, d	H-3, H-11a, H-5, H-5’	C-1, C-4, C-5, C-11a
5α	2.63, ddd,15.0, 8.4, 2.2	27.0, t	H-4a,H-5β, H-6	C-4a, C-6, C-7, C-11, C-11a
5β	1.54, m		H-4a H-5α, H-6	C-4, C-4a, C-6, C-7
6	2.17, m	42.5, d	H-5α, H-5β	C-4a, C-7, C-8, C-11
7		46.6, s		
8	1.95, m	28.3, t	H-8’, H-9	C-6, C-7, C-9
8’	1.70, m		H-8	C-9, C-10
9	1.76, m	15.7, t	H-8’, H-10β	C-10, C-11
9’	1.73, m			
10α	1.90, m	27.4, t	H-9, H-10β	C-6, C-9, C-11, C-12
10β	1.76, m		H-10α	C-9
11	-	53.2, s		
11a	1.69, dd, 12.4, 9.8	50.3, d	H-1, H-4a	C-1, C-4, C-4a, C-11, C-12
12	4.83, s	68.2, d		C-7, C-8, C-11, C-11a, C-19, C-22
13		198.5, s		
14	2.45, ddd, 7.7, 7.7, 3.8	35.4, t	H-15	C-13, C-15, C-16
15	1.49, m	34.1, t	H-14	C-14, C-16, C-17, C-18
15’	1.46, m			
16	1.53, m	27.8, d	H-17, H-18	C-15, C-17, C-18
17	0.89, d, 6.3	22.4, q	H-16	C-15, C-16, C-18
18	0.88, d, 6.5	22.4, q	H-16	C-15, C-16, C-17
19	3.93, d, 11.6	66.1, t		C-6, C-7, C-8, C-12, C-20
19’	4.02, d, 11.6			C-6, C-7, C-8, C-12, C-20
20		171.0, s		
21	2.06, s	20.9, q		C-19, C-20
22		170.2, s		
23	2.03, s	20.7, q		C-12, C-22
24		168.7, s		
25	2.09, s	20.8, q		C-1, C-24



Plumisclerin A (1)





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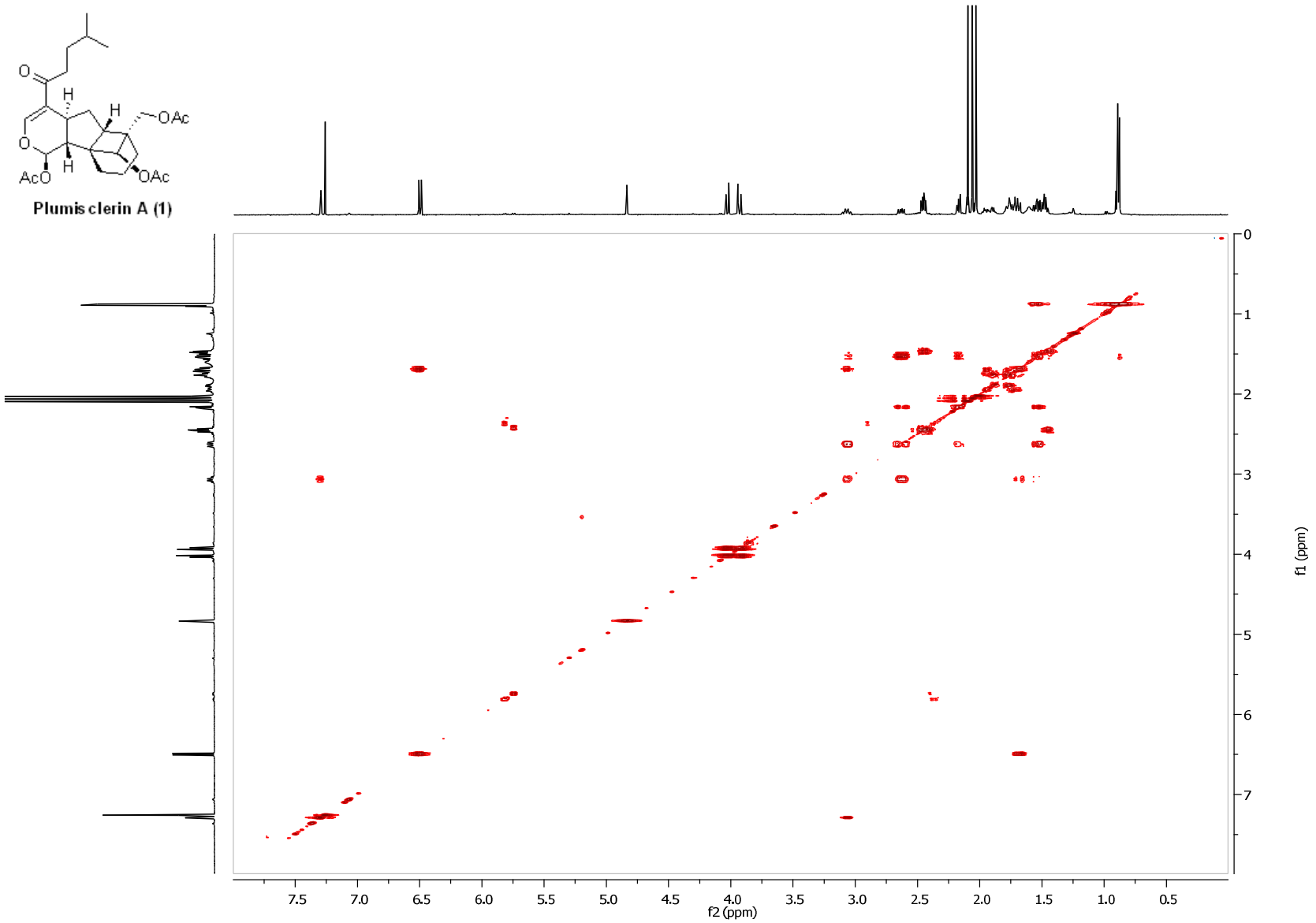
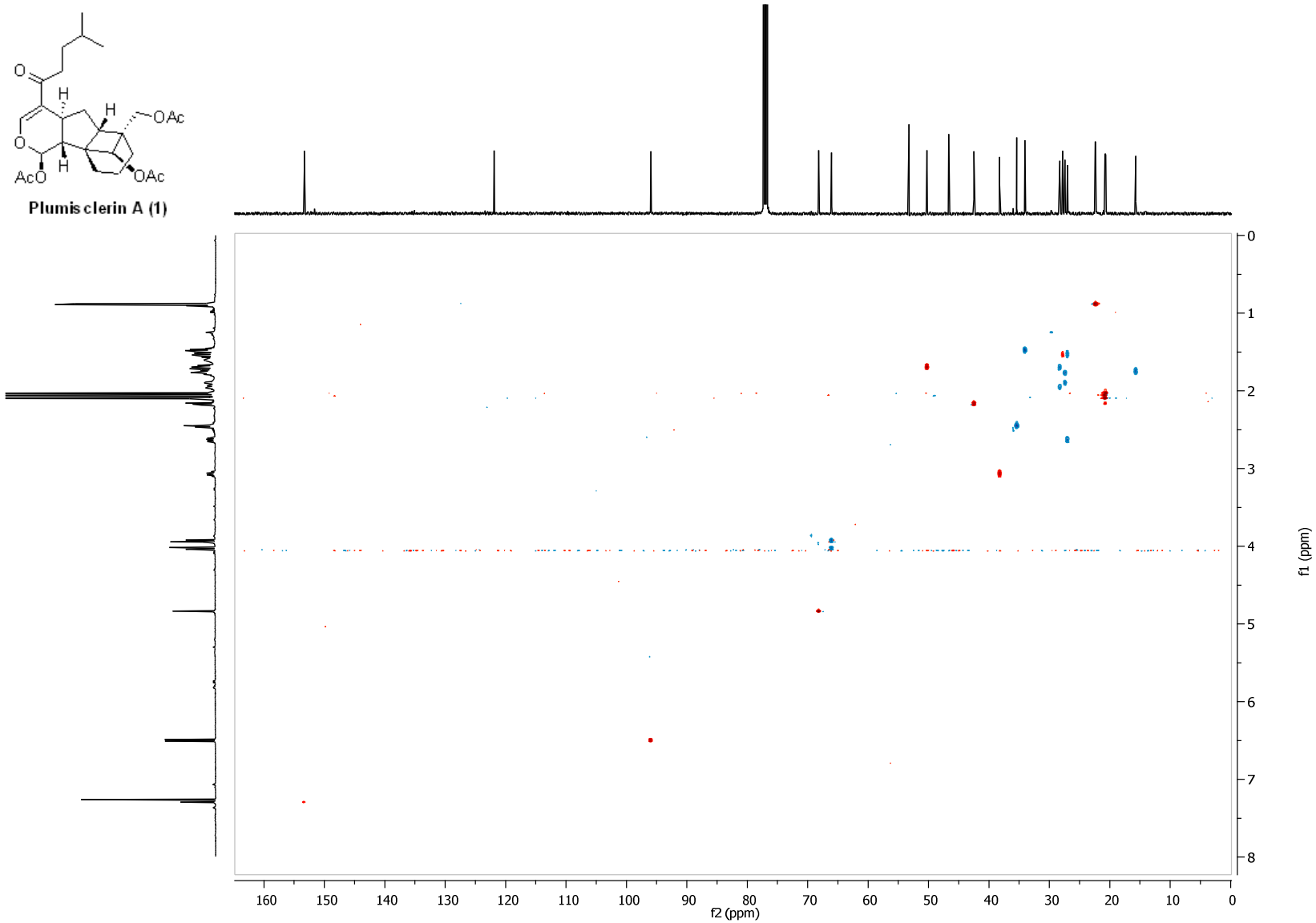

Plumisclerin A (1)
f2 (ppm)
f1 (ppm)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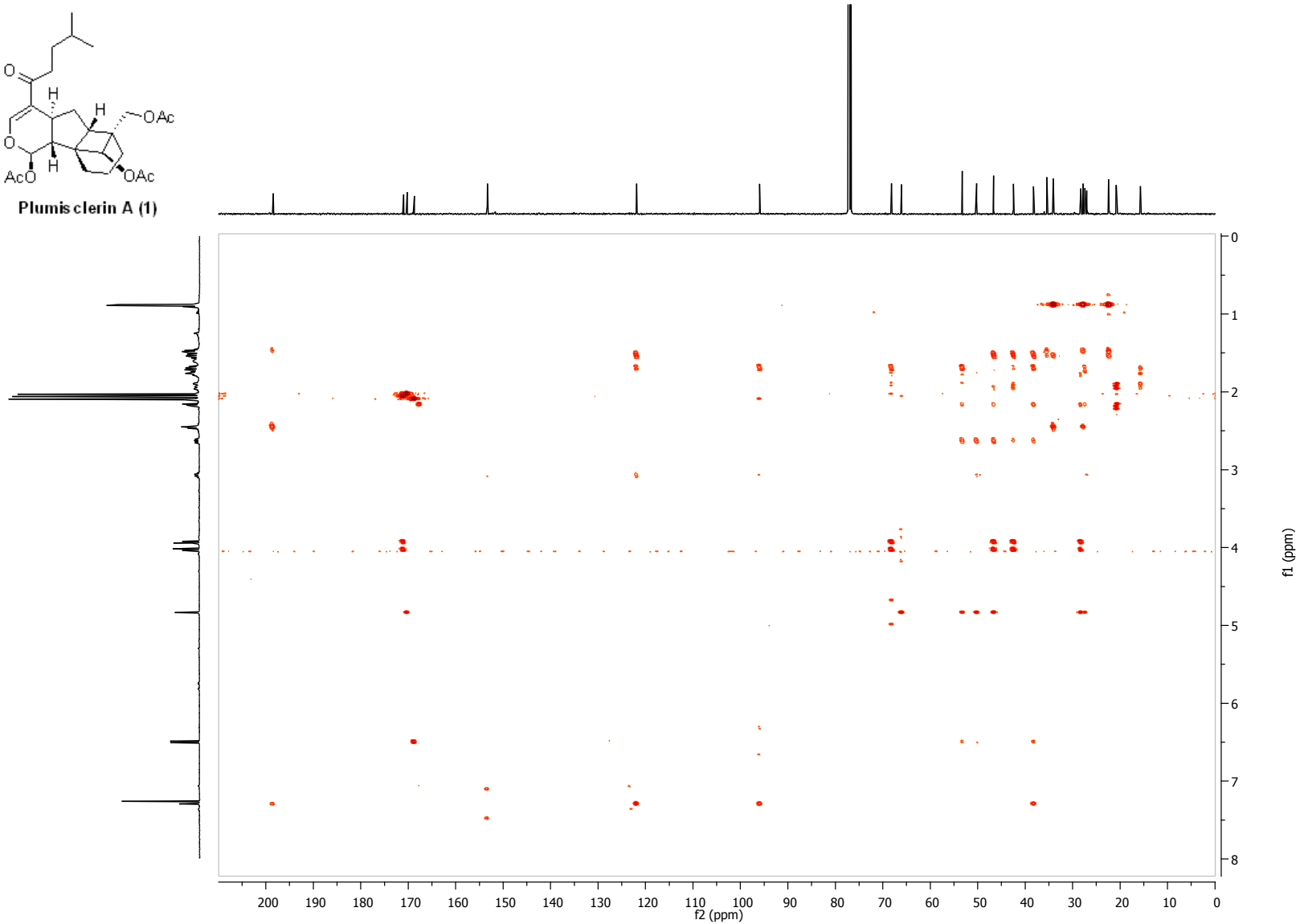
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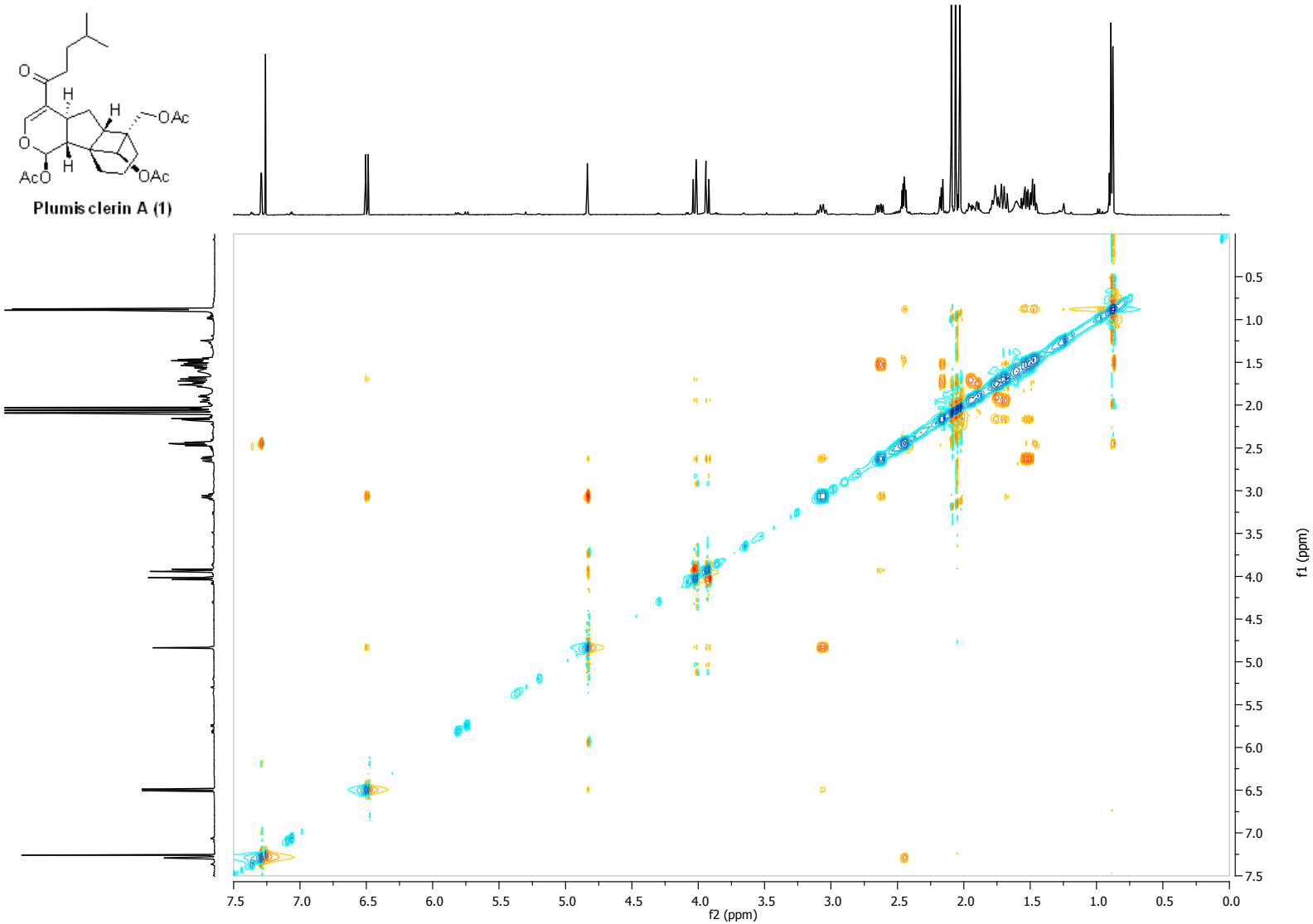
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