Supplementary Materials: Acute Toxicity of Gambierone and Quantitative Analysis of Gambierones Produced by Cohabitating Benthic Dinoflagellates

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Table S1. Comparison of the ^{13}C (125 MHz) and ^{1}H (500 MHz) NMR chemical shifts (ppm), multiplicity and coupling constants (Hz) for gambierone purified from *Gambierdiscus cheloniae* CAWD232, generated in d4-MeOH, and those published by Rodriguez *et al*., 2015 [38].

Atom	Purified gambierone		Published gambierone[b]	
	δ_C[a]	δ_H (multi, *J* in Hz)	δ_C	δ_H (multi, *J* in Hz)
1	67.6	3.47 (-)[c], 3.42 (m)	67.9	3.47 (dd, 11.0, 4.5), 3.42 (m)
2	69.8	4.10 (m)	70.3	4.11 (m)
3	39.6	2.01 (m), 1.70 (dd, 14.5, 10.1)	39.6	2.01 (m), 1.70 (dd, 14.5, 10.0)
4	-[d]	-	101.0	-
5	73.0	4.21 (d, 3.2)	73.1	4.21 (d, 3.2)
6	77.7	4.70 (dd, 10.0, 3.2)	77.9	4.70 (dd, 10.0, 3.2)
7	77.1	3.37 (-)[c]	77.4	3.37 (t, 10.0)
8	67.9	3.76 (m)	68.0	3.77 (m)
9	37.8	2.19 (m), 1.58 (-)[c]	37.8	2.17 (m), 1.58 (q, 12.0)
10	79.6	3.35 (-)[c]	79.8	3.35 (m)
11	82.6	3.80 (m)	82.6	3.79 (m)
12	132.7	5.64 (dd, 12.5, 2.4)	133.2	5.64 (dd, 12.5, 2.5)
13	132.9	5.74 (dd, 12.4, 2.5)	133.5	5.75 (dd, 12.5, 2.5)
14	82.9	3.81 (m)	83.2	3.81 (m)
15	80.0	3.45 (m)	80.0	3.44 (m)
16	47.2	1.98 (m), 1.49 (t, 10.9)	47.4	1.99 (m), 1.49 (t, 11.0)
17	-[d]	-	76.7	-
18	86.8	3.0 (dd, 10.9, 2.5)	87.0	3.00 (dd, 11.0, 2.5)
19	25.1	1.77 (-)[c], 1.62 (-)[c]	25.3	1.78 (m), 1.62 (m)
20	34.0	1.95 (-)[c], 1.81 (-)[c]	34.2	1.95 (m), 1.80 (m)
21	87.3	3.53 (m)	87.6	3.54 (m)
22	75.8	3.53 (m)	75.9	3.54 (m)
23	32.6	1.82 (-)[c], 1.64 (m)	32.7	1.82 (m), 1.64 (m)
24	29.5	1.94 (-)[c], 1.77 (-)[c]	29.5	1.92 (m), 1.77 (m)
25	35.5	2.19 (-)[c]	35.6	2.19 (m)
26	86.0	3.11 (-)[c]	86.1	3.11 (m)
27	77.5	3.51 (m)	77.9	3.51 (m)
28	39.7	2.20 (-)[c], 1.33 (q, 11.2)	39.8	2.20 (m), 1.33 (q,11.3)
29	70.0	3.12 (-)[c]	70.2	3.12 (td, 9.5, 4.3)
30	77.8	2.94 (ddd, 11.8, 9.6, 4.6)	78.3	2.94 (ddd, 11.5, 9.5, 4.5),
31	34.7	1.89 (-)[c], 1.54 (q, 11.7)	34.9	1.90 (m), 1.55 (q, 11.5)
32	72.4	3.78 (m)	72.6	3.77 (m)
33	-[d]	-	77.4	-
34	54.0	2.36 (d, 12.1), 2.14 (-)[c]	54.2	2.35 (d, 12.0), 2.14 (d, 12.0)
35	-[d]	-	144.0	-
36	43.3	2.55 (d, 14.0), 2.22 (d, 14.1)	43.3	2.54 (d, 14.0), 2.22 (d, 14.0)

37	- [d]	-	80.0	-
38	73.1	4.06 (dd, 8.7, 3.6)	73.5	4.06 (dd, 8.7, 3.5)
39	45.7	2.63 (dd, 12.0, 3.0), 2.60 (dd, 12.1, 9.0)	45.8	2.62 (dd, 12.0, 3.0), 2.60 (dd, 12.0, 9.0)
40	- [d]	-	212.2	-
41	43.7	2.61 (t, 7.0)	43.7	2.61 (t, 7.0)
42	27.2	2.33 (q, 7.0)	27.3	2.33 (q, 7.0)
43	134.3	5.70 (dt, 15.1, 7.2)	134.9	5.70 (dt, 15.0, 7.0)
44	132.9	6.08 (dd, 15.1, 10.4)	133.4	6.08 (dd, 15.0, 10.5)
45	138.6	6.28 (dt 16.9, 10.3)	138.9	6.29 (dt, 17.0, 10.4)
46	115.4	5.07 (dd, 17.1, 1.7), 4.94 (dd, 10.3, 1.7)	115.9	5.08 (dd, 17.0, 1.8), 4.94 (dd, 10.3, 1.8)
47	16.2	1.21 (3H, s)	16.2	1.20 (3H, s)
48	13.3	1.00 (3H, d, 7.3)	13.3	1.00 (3H, d, 7.3)
49	16.6	1.19 (3H, s)	16.7	1.19 (3H, s)
50	118.5	4.98 (br s), 4.85 (br s)	119.0	4.98 (br s), 4.86 (br s)
51	20.6	1.13 (3H, s)	20.6	1.13 (3H, s)

[a] Carbon chemical shifts determined from an HSQC spectrum. [b] Values from Rodriguez *et al*., 2015 [38]. [c] Coupling pattern unable to be determined using current dataset. [d] Quaternary carbon, therefore unable to determine chemical shift from HSQC.

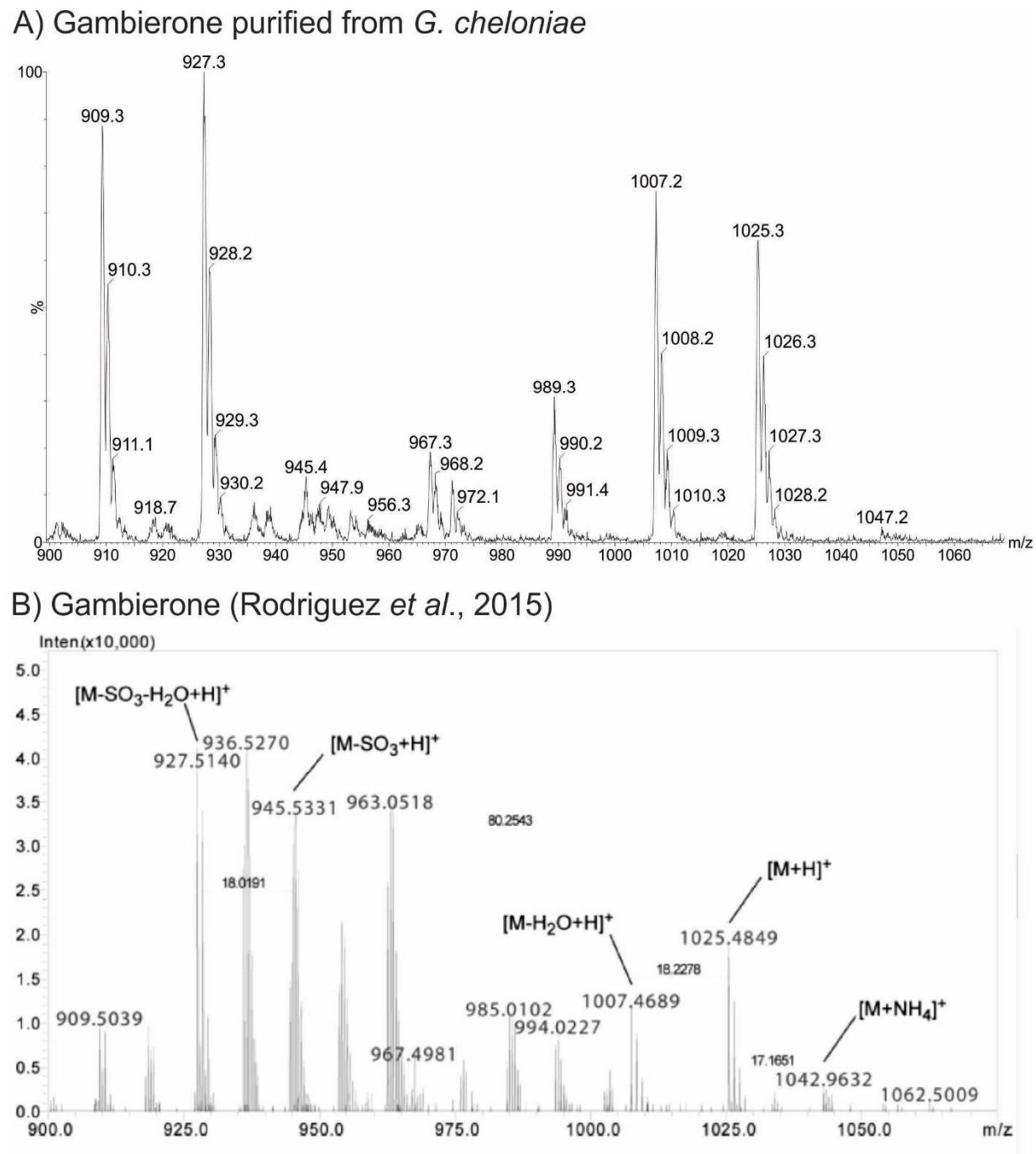

Figure S1. Comparison of the mass spectra of gambierone in positive electrospray ionization mode (**A**) purified from *Gambierdiscus cheloniae* CAWD232 and (**B**) published by Rodriguez *et al*., 2015 [38].

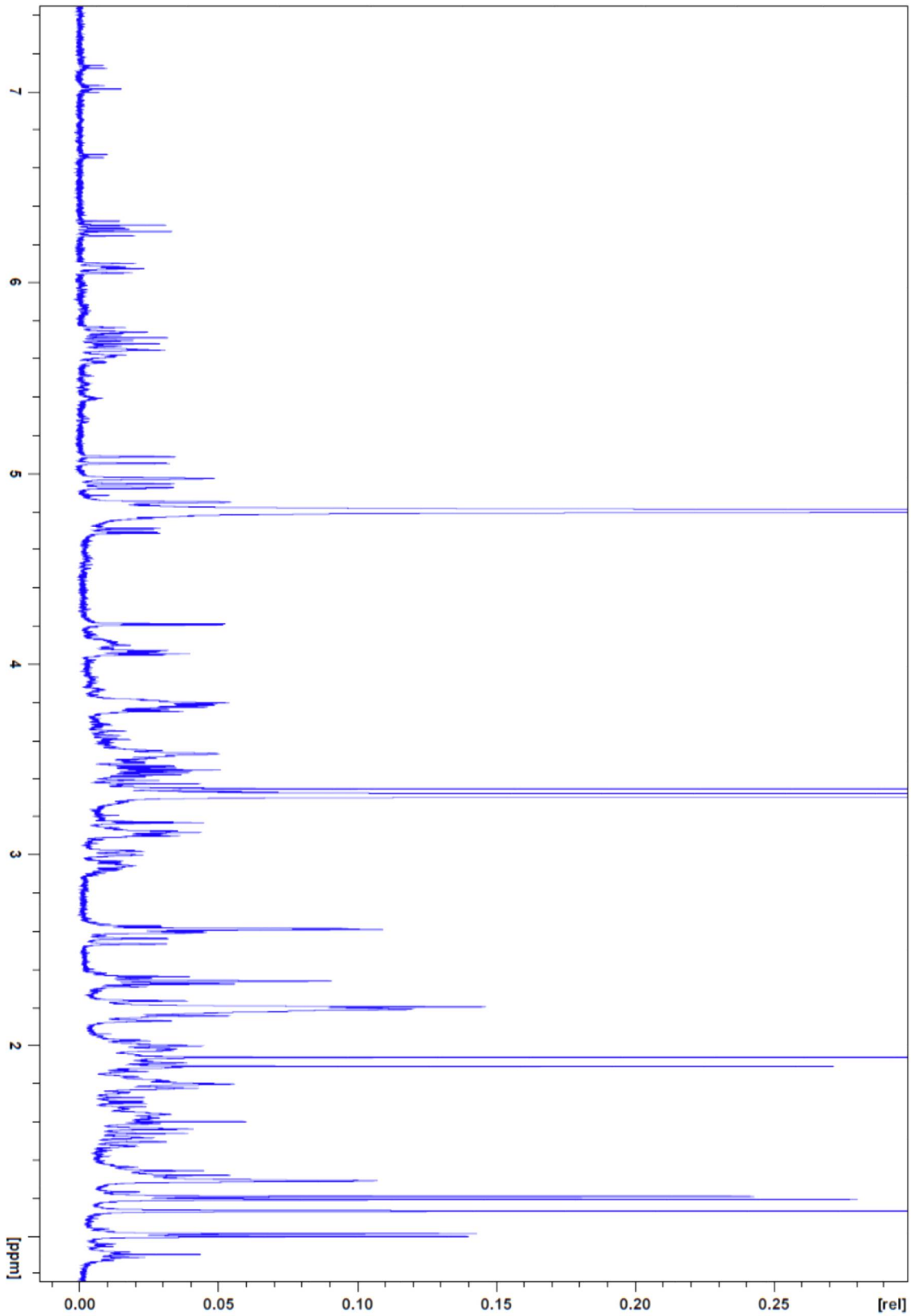

Figure S2. ^{1}H NMR spectrum of gambierone purified from *Gambierdiscus cheloniae* CAWD232 acquired on a Bruker Advance III 500 MHz instrument in CD_3OH (≥99.8% atom D).

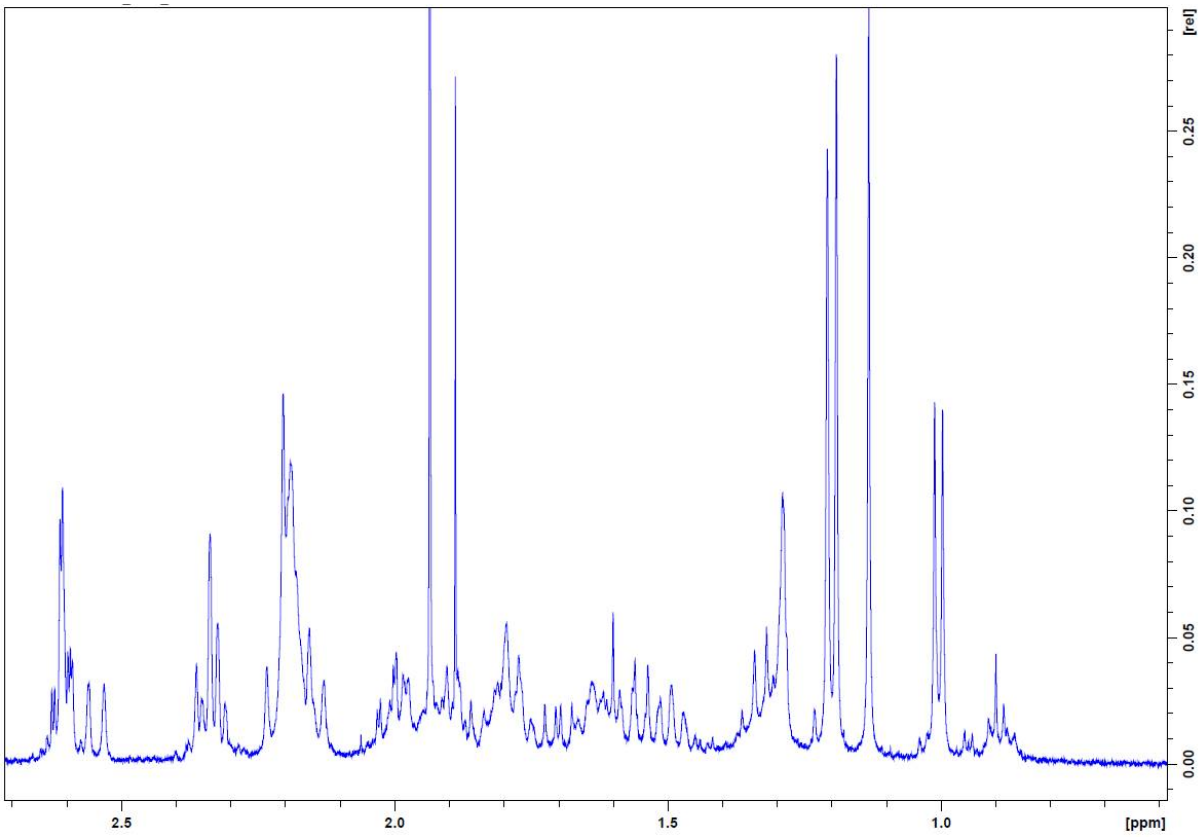

Figure S3. Expansion of the ^{1}H NMR spectrum (0.6–2.7 ppm) of gambierone in CD_3OD (≥99.8% atom D) at 500 MHz.

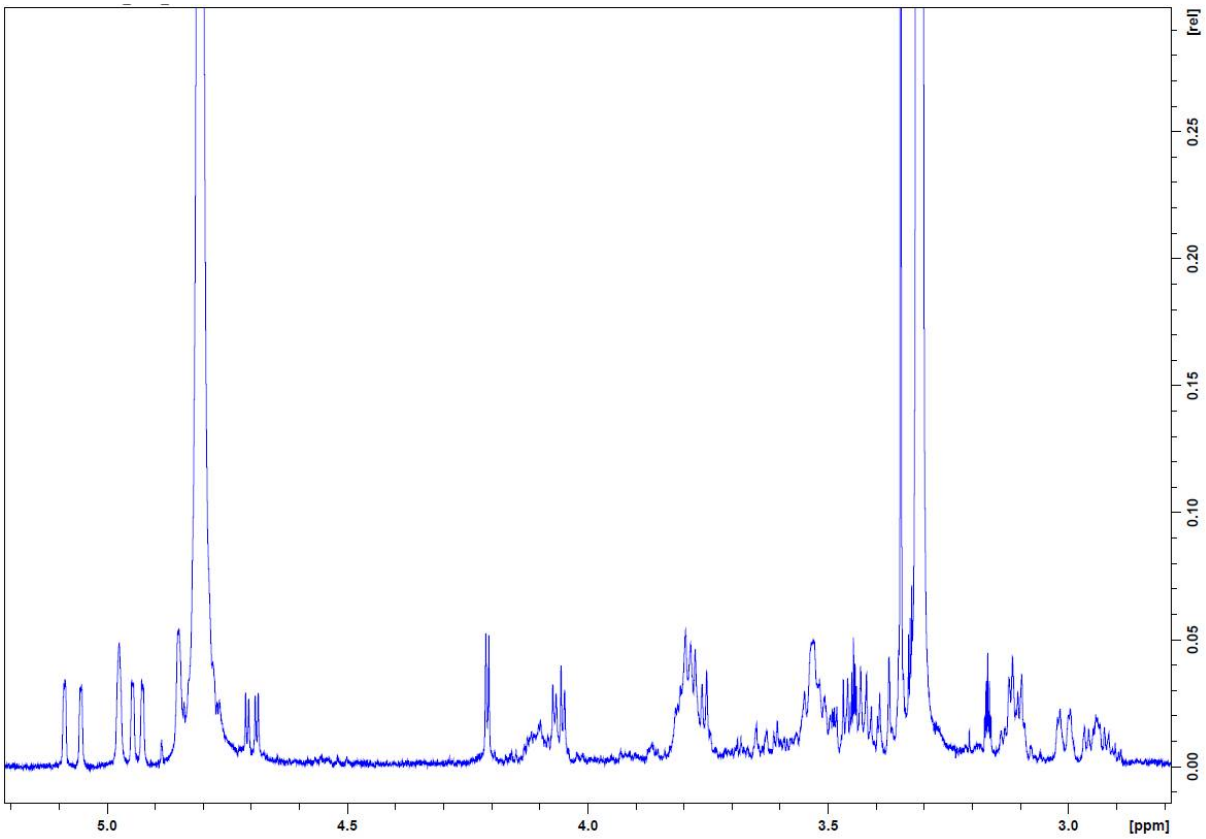

Figure S4. Expansion of the ^{1}H NMR spectrum (2.8–5.2 ppm) of gambierone in CD_3OD (≥99.8% atom D) at 500 MHz.

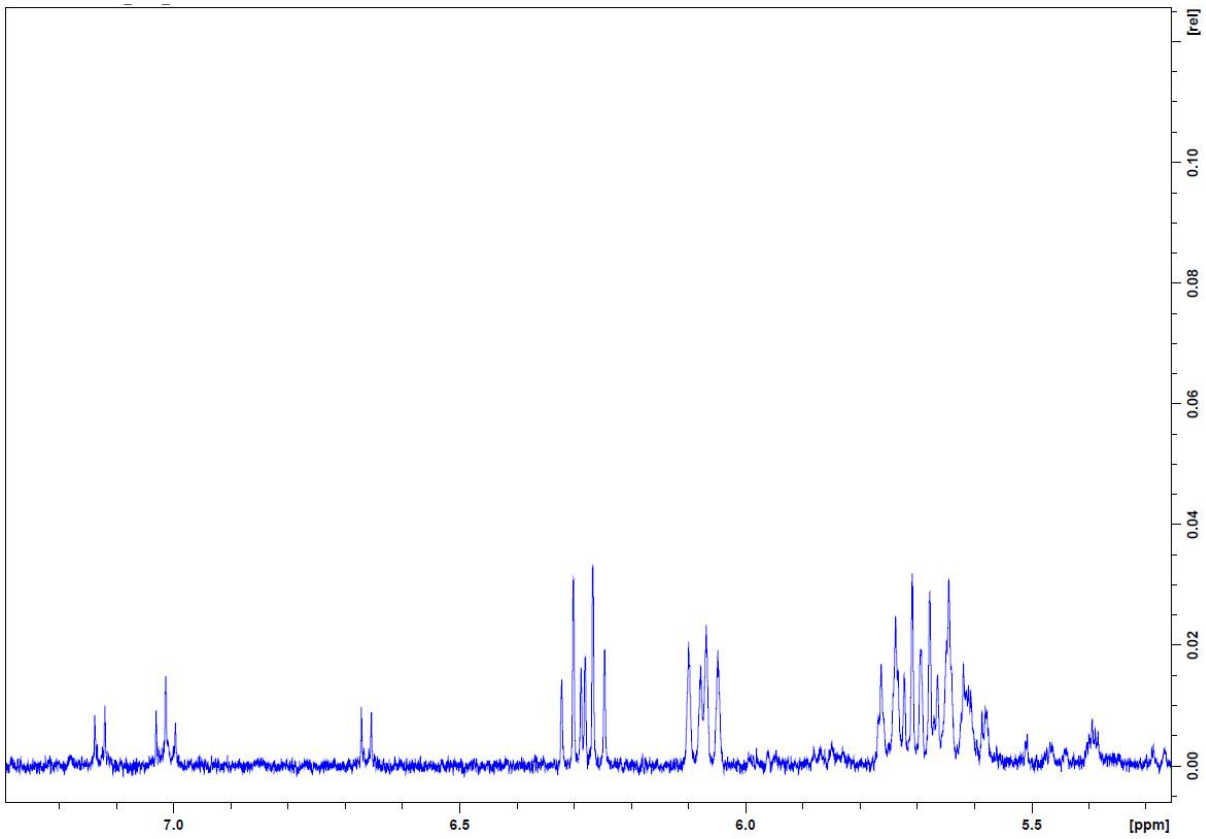

Figure S5. Expansion of the ^{1}H NMR spectrum (5.3–7.3 ppm) of gambierone in CD_3OD (≥99.8% atom D) at 500 MHz.

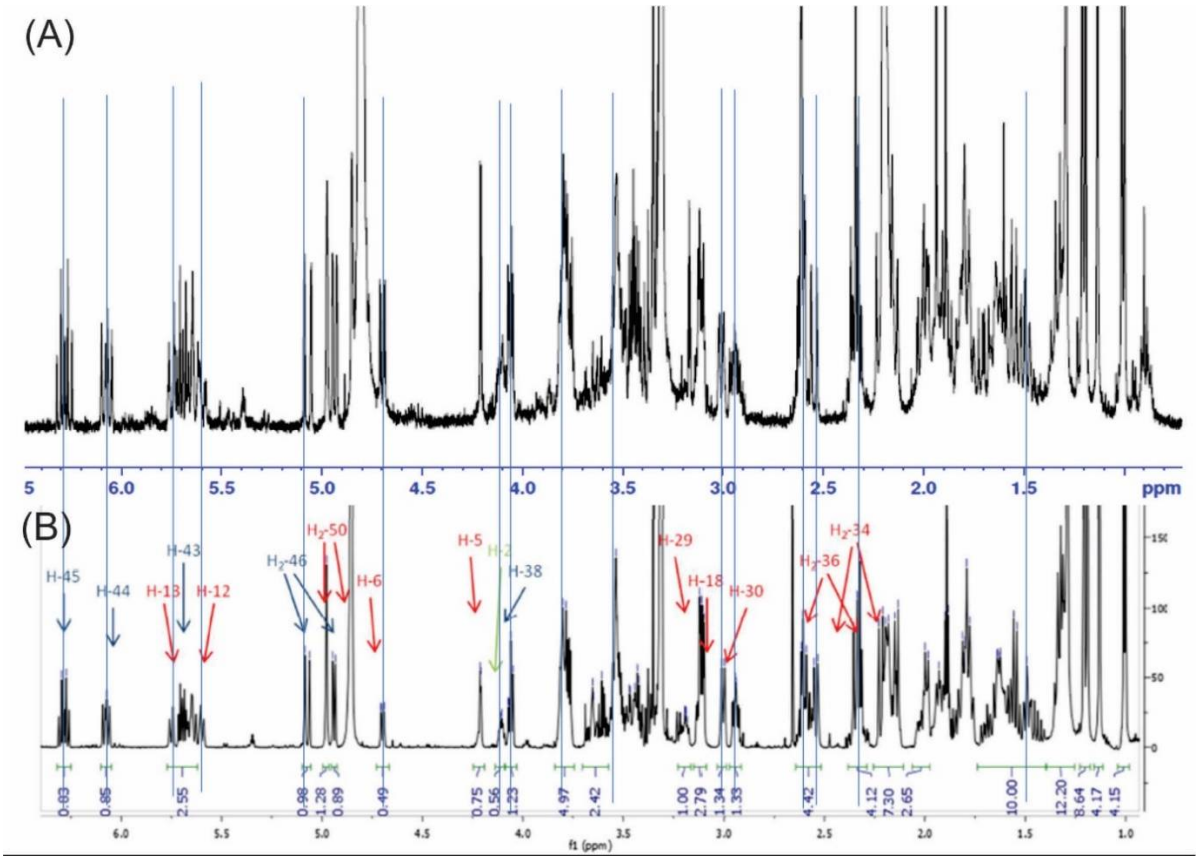

Figure S6. Comparison of the ^{1}H NMR spectrum of (**A**) gambierone purified from *Gambierdiscus cheloniae* CAWD232 acquired on a Bruker Advance III 500 MHz instrument and (**B**) the published spectrum from Rodriguez *et al*., acquired on a Varian Inova 750 MHz instrument [38].

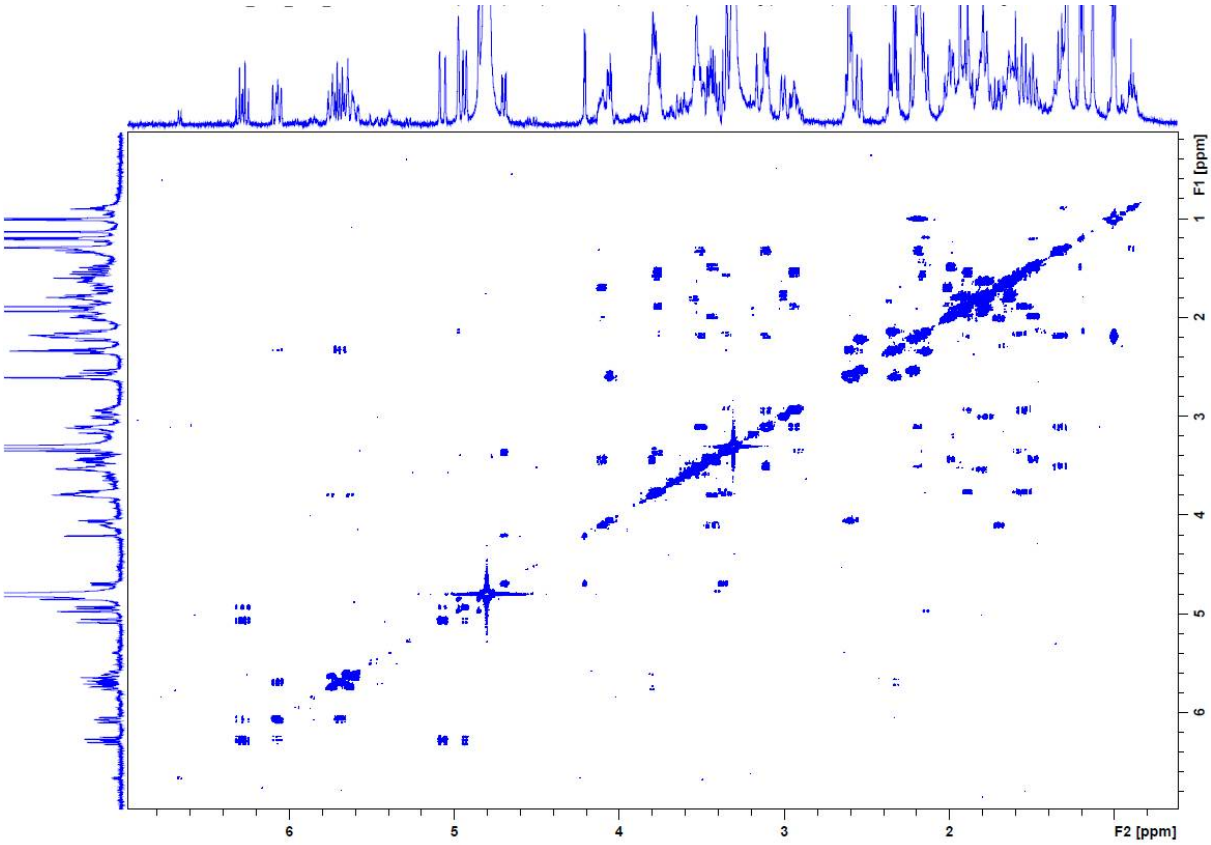

Figure S7. COSY NMR spectrum of gambierone purified from *Gambierdiscus cheloniae* CAWD232 in CD_3OD (≥99.8% atom D) at 500 MHz.

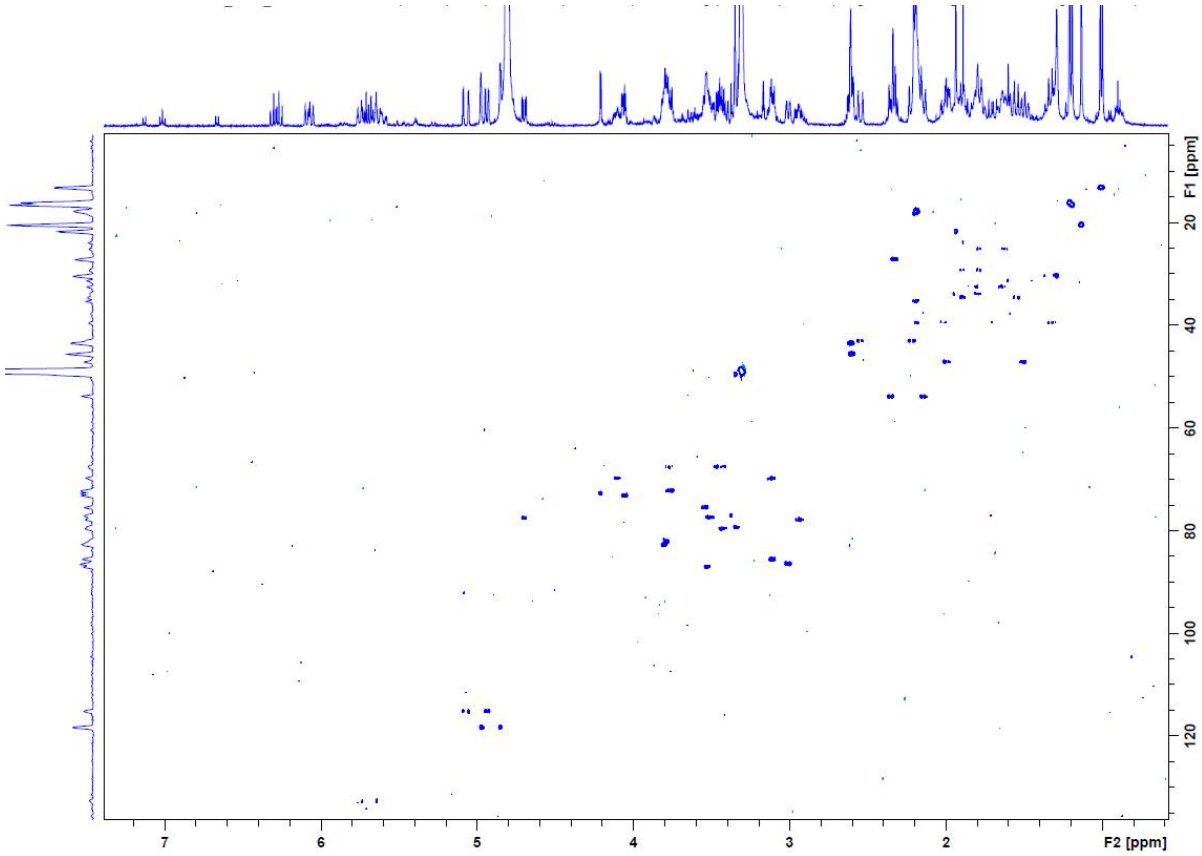

Figure S8. HSQC NMR spectrum of gambierone purified from *Gambierdiscus cheloniae* CAWD232 in CD_3OD (≥99.8% atom D) at 500 MHz.

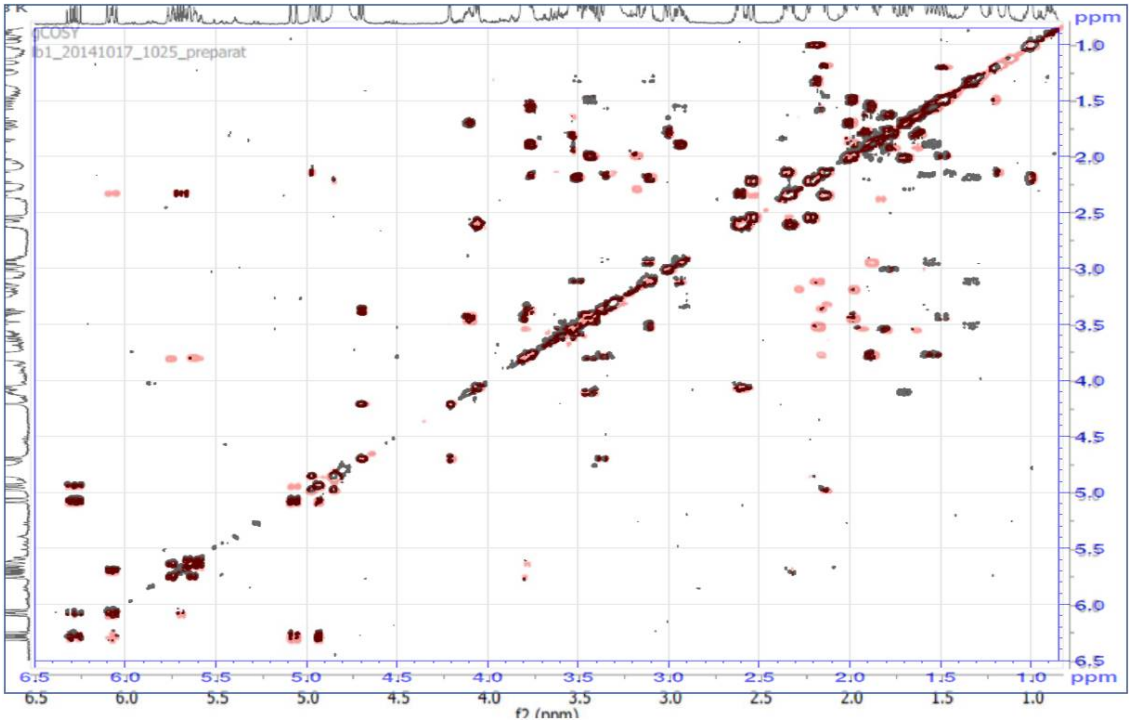

Figure S9. Comparison of the COSY NMR spectrum (1.0–6.5 ppm) of gambierone purified from *Gambierdiscus cheloniae* CAWD232 (black) and that published by Rodriguez *et al*., 2015 (red) [38]. Long range (~2 Hz) couplings not displayed.

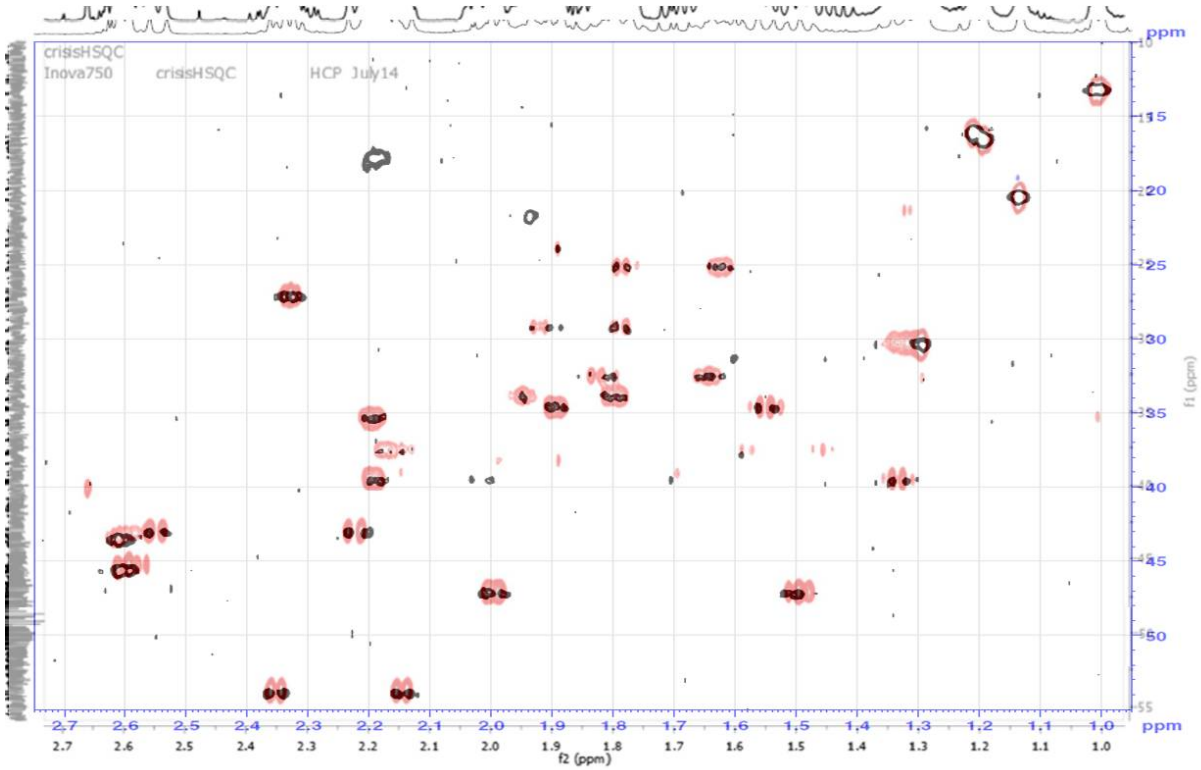

Figure S10. Expansion (^{1}H: 1.0–2.7 ppm; ^{13}C: 10–55 ppm) of the HSQC spectrum of gambierone purified from *Gambierdiscus cheloniae* CAWD232 (black), and the published spectrum from Rodriguez *et al*., 2015 (red) [38].

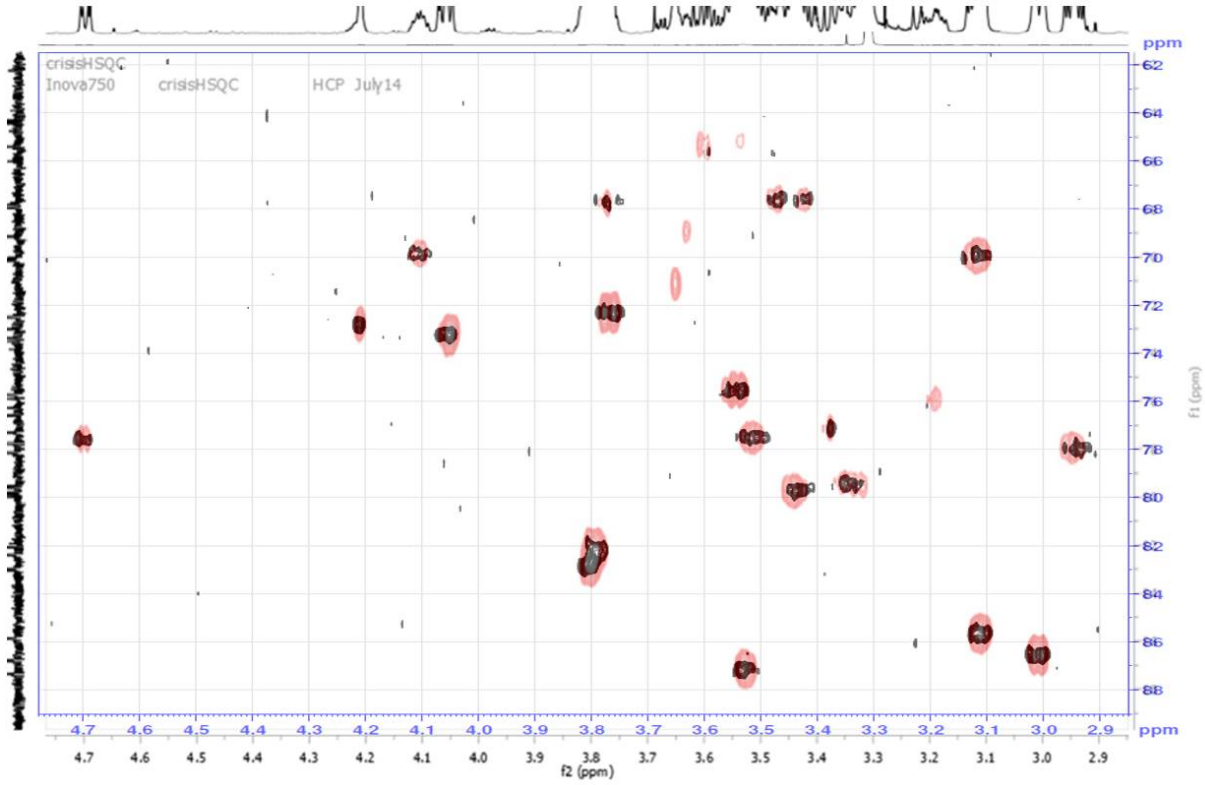

Figure S11. Expansion (^{1}H: 2.5–4.7 ppm; ^{13}C: 62–85 ppm) of the HSQC spectrum of gambierone purified from *Gambierdiscus cheloniae* CAWD232 (black), and the published spectrum from Rodriguez *et al*., 2015 (red) [38].

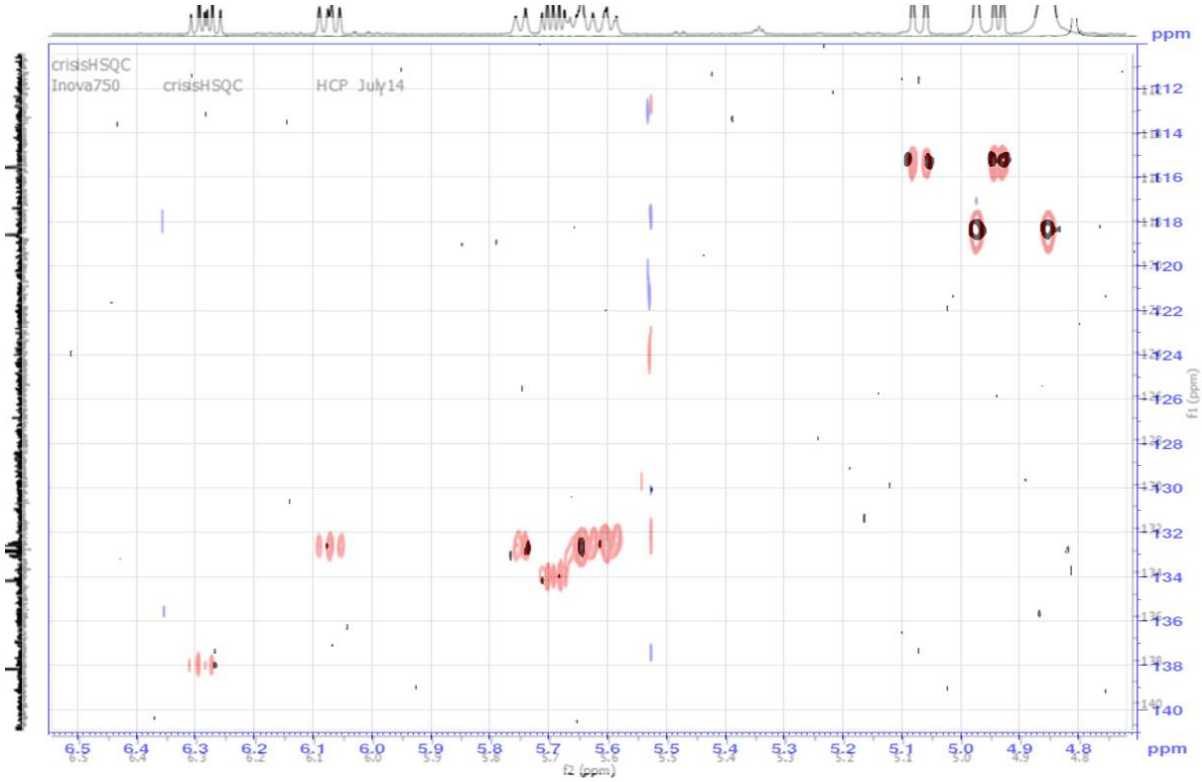

Figure S12. Expansion (^{1}H: 4.7–6.5 ppm; ^{13}C: 110–140 ppm) of the HSQC spectrum of gambierone purified from *Gambierdiscus cheloniae* CAWD232 (black), and the published spectrum from Rodriguez *et al*., 2015 (red) [38].

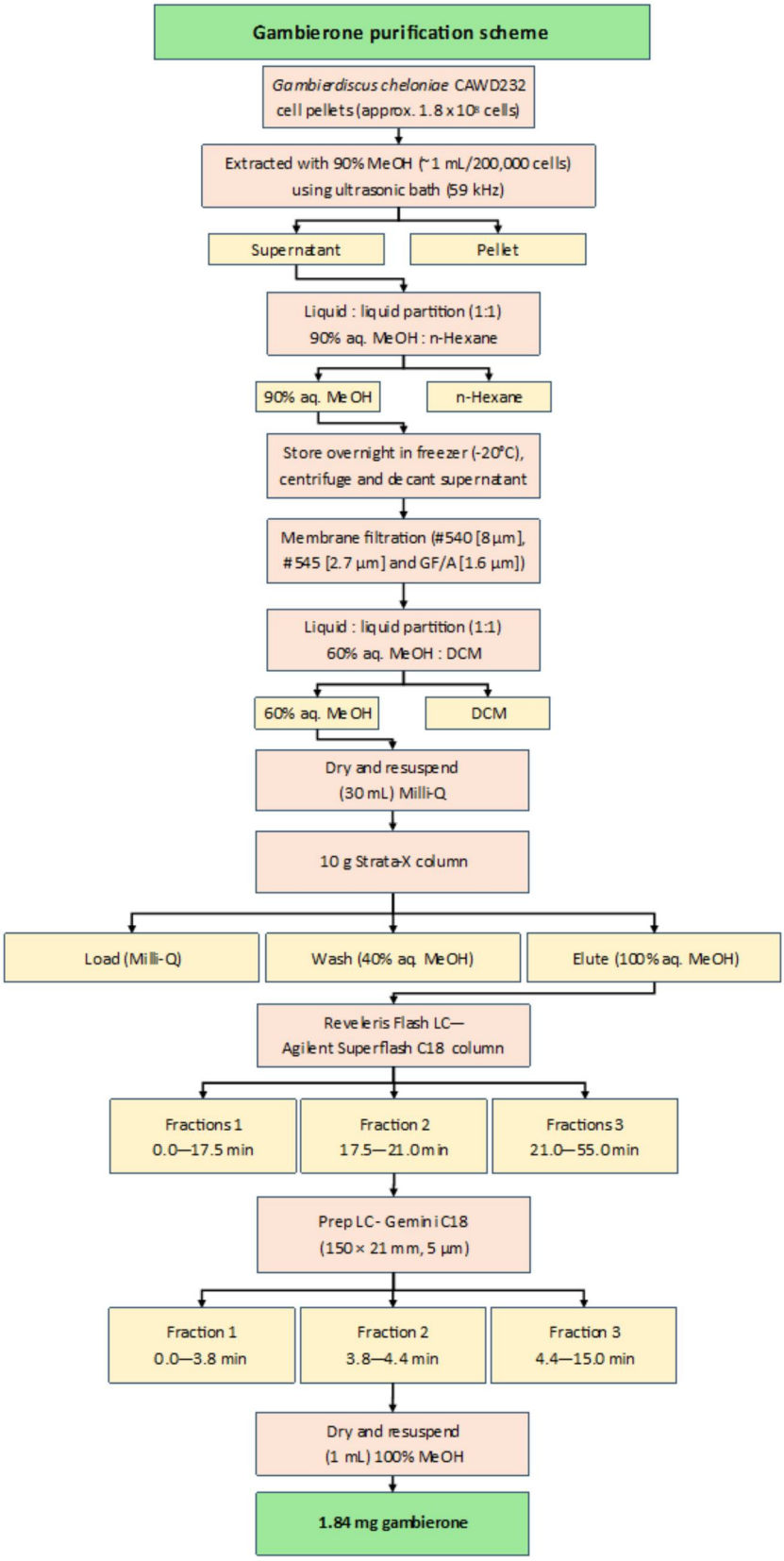

Figure S13. Purification scheme for the isolation of gambierone from *Gambierdiscus cheloniae* CAWD232.