

Supplementary data

Title: NF- κ B inhibitors from *Eurycoma longifolia*

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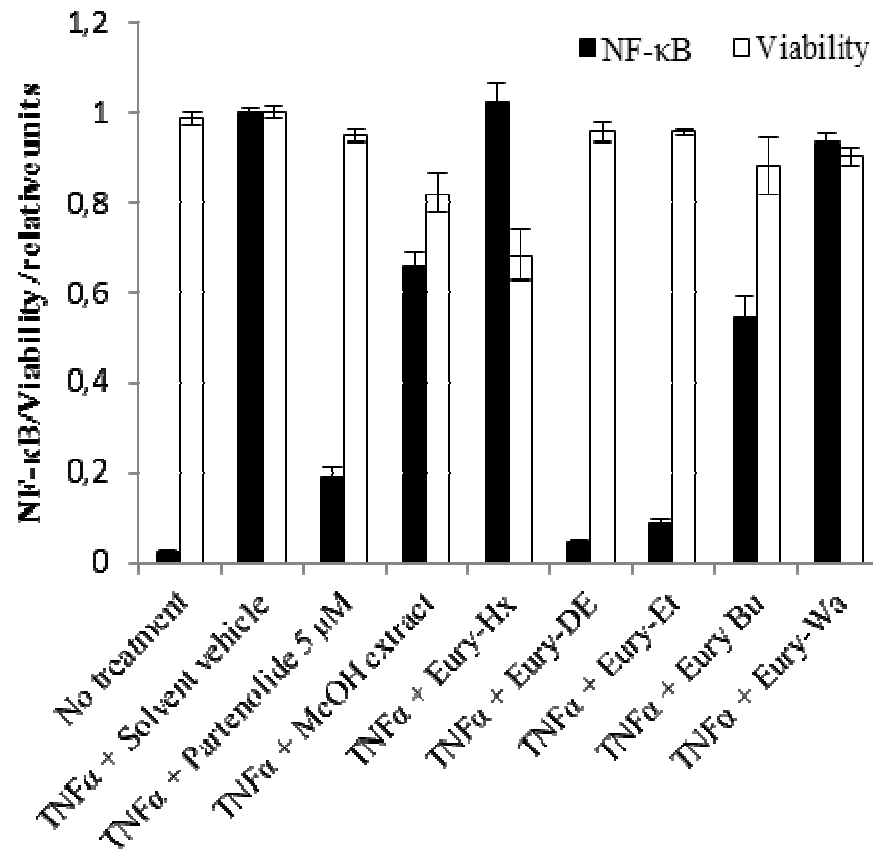
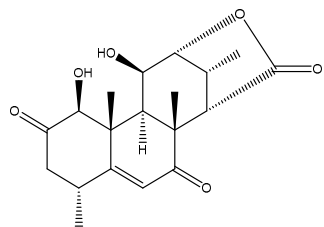


Figure S1: Effect of fractions (Eury-Hx: *n*-hexane fraction, Euy-DE: diethyl ether fraction, Eury-Et: ethyl acetate fraction, Eury-Bu: *n*-butanol fraction, Eury-Wa: water fraction) on NF-κB activity in HEK-293/NF-κB-luc cells; negative control: cells were treated with solvent vehicle (0.1% DMSO; the final concentration of DMSO in all treatment groups was equal) and stimulated with TNF-α (2 ng/mL); positive control: cells were treated with parthenolide (5 μM) and stimulated with TNF-α. The fractions were tested at a concentration of 10 μg/mL, data points represent mean ± S.E.M.; *n* = 3, * *p* < 0.01 (ANOVA/Tukey).

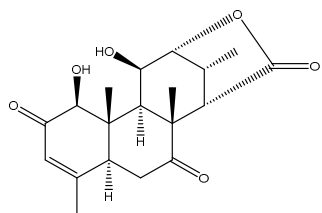
Table 1: NF- κ B Inhibition and Cell Viability of Isolated Compounds (at 30 μ M) on TNF- α -Stimulated HEK-293/NF- κ B-luc Cells and IC₅₀ Values and the corresponding CI₉₅ Values of Compounds which exhibited more than 50% inhibition at 30 μ M. Data shown as mean $\pm$ S.E.M./S.D.; n = 3.

compound	NF- κ B inhibition (%) at 30 μ M	S.E.M.	S.D.	cell viability (%) at 30 μ M	S.E.M.	S.D.	IC ₅₀ (μ M)	CI ₉₅ (μ M)
eurycomalide C (1)	79.2	1.0	4.6	80.1	1.8	7.7	18.4	16.9-20.1
eurycomalactone (2)	99.9	0.3	1.4	66.7	5.1	21.8	0.5	0.3-0.7
7 α -hydroxyeurycomalactone (3)	99.6	0.4	1.6	76.0	2.4	10.1	1.5	1.3-1.6
5,6-dehydroeurycomalactone (4)	92.2	0.7	2.9	74.8	2.7	10.6	6.2	5.3-7.4
eurycolactone E (5)	96.8	0.3	1.1	97.4	9.4	32.6	3.8	3.2-4.5
longilactone (6)	97.8	0.3	1.1	96.6	7.4	25.5	4.7	3.7-5.9
14,15 β -dihydroxyklaianone (7)	96.8	0.4	1.7	96.6	7.8	31.0	1.0	0.8-1.2
11-dehydroklaianone (8)	99.2	0.2	0.6	96.9	6.7	23.3	1.9	1.8-2.1
eurycomanone (9)	97.8	0.2	0.6	88.4	4.8	16.7	2.4	2.0-2.9
13,21-dehydroeurycomanone (10)	98.0	0.2	0.6	96.9	6.7	23.3	0.7	0.6-0.9
laurycolactone A (11)	-29.4	10.5	36.5	90.0	9.6	33.4	-	-
laurycolactone B (12)	17.8	9.2	32.0	71.4	13.1	45.4	-	-
1-methoxycarbonyl- β -carboline (13)	50.6	4.3	19.3	105.7	9.1	40.6	29.3	20.3-42.4
9-hydroxycanthin-6-one (14)	90.3	0.5	2.2	112.0	9.4	37.5	3.8	3.3-4.4
9-methoxycanthin-6-one (15)	95.6	0.5	2.0	110.5	10.1	41.5	7.4	6.6-8.2
9,10-dimethoxycanthin-6-one (16) ^a	62.1	1.1	3.7	83.4	1.4	4.8	19.5	13.0-29.4
4-hydroxy-5-methoxycanthin-6-one (17)	-19.7	5.8	22.6	111.4	11.7	45.3	-	-
9-O- β -D-glucosylcanthine-6-one (18)	0.2	1.7	6.0	101.5	8.3	28.8	-	-
scopoletin (19)	-2.3	5.0	20.1	129.5	13.2	52.9	-	-
fraxidin (20)	8.2	2.4	0.1	130.3	11.8	44.1	-	-
eurylene (21)	-42.9	10.2	35.4	89.4	7.4	25.7	-	-
pedunculoside (22)	7.3	4.4	15.2	76.7	4.8	16.5	-	-
vanilic acid (23)	0.1	2.7	10.5	132.6	10.1	39.1	-	-
vanilic aldehyde (24)	-10.7	3.1	10.7	144.8	10.7	36.9	-	-
syringic acid (25)	1.9	4.6	17.2	129.6	14.8	55.4	-	-
1,1'-biphenyl-3,3'-dicarboxylic acid (26)	-15.1	3.2	10.5	138.8	11.2	37.3	-	-
isoaloeresine D (27)	-0.4	5.2	18.2	77.6	6.4	22.1	-	-
3,5,6,7,8,3',4'-heptamethoxyflavone (28)	63.9	2.0	6.8	88.4	4.9	17.0	23.3	18.4-29.4
parthenolide (5 μ M)	75.8	2.8	10.3	98.0	5.4	23.3	1.5	1.3-1.8

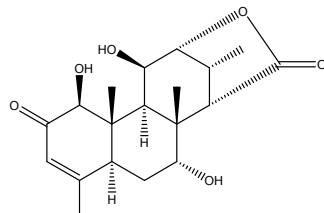
^atested as the corresponding HCl-salt, dissolved in water.



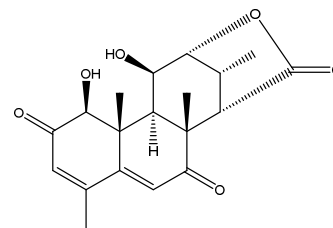
1. eurycomalide C



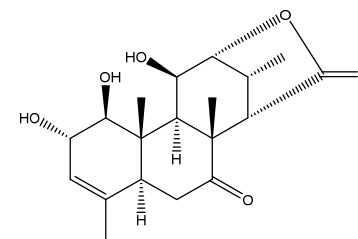
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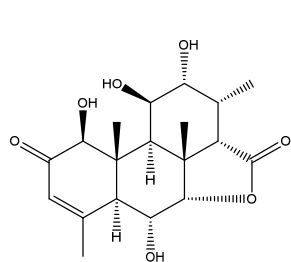
3. 7 α -hydroeurycomalactone



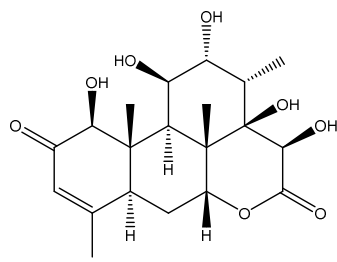
4. 5,6-dehydroeurycomalactone



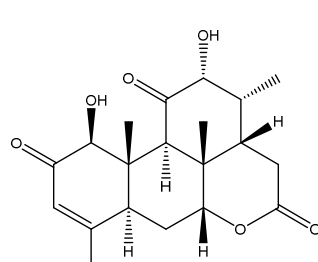
5. eurycolactone E



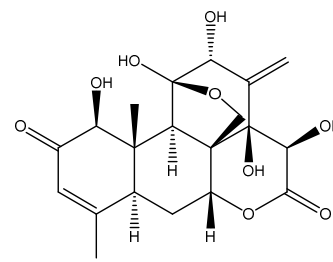
6. longilactone



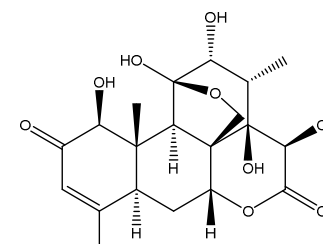
7. 14, 15 β -dihydroklaieanone



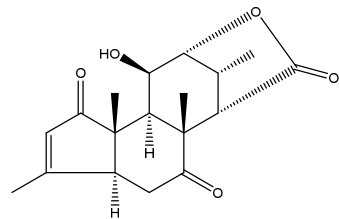
8. 11-dehydroklaieanone



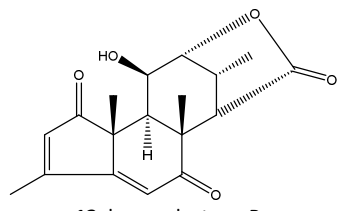
9. eurycomanone



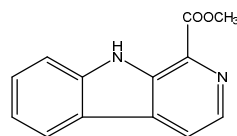
10. 13, 21-dehydroeurycomanone



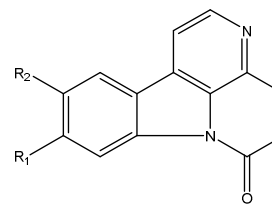
11. laurycolactone A



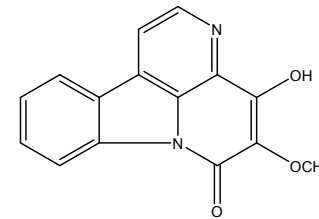
12. laurycolactone B



13. 1-methoxy-carbonyl- β -carboline



14: $R_1 = \text{OH}$, $R_2 = \text{H}$
15: $R_1 = \text{OCH}_3$, $R_2 = \text{H}$
16: $R_1 = \text{OCH}_3$, $R_2 = \text{OCH}_3$
18: $R_1 = \text{glucose}$, $R_2 = \text{H}$



17. 5-methoxy-4-hydroxycanthin-6-one

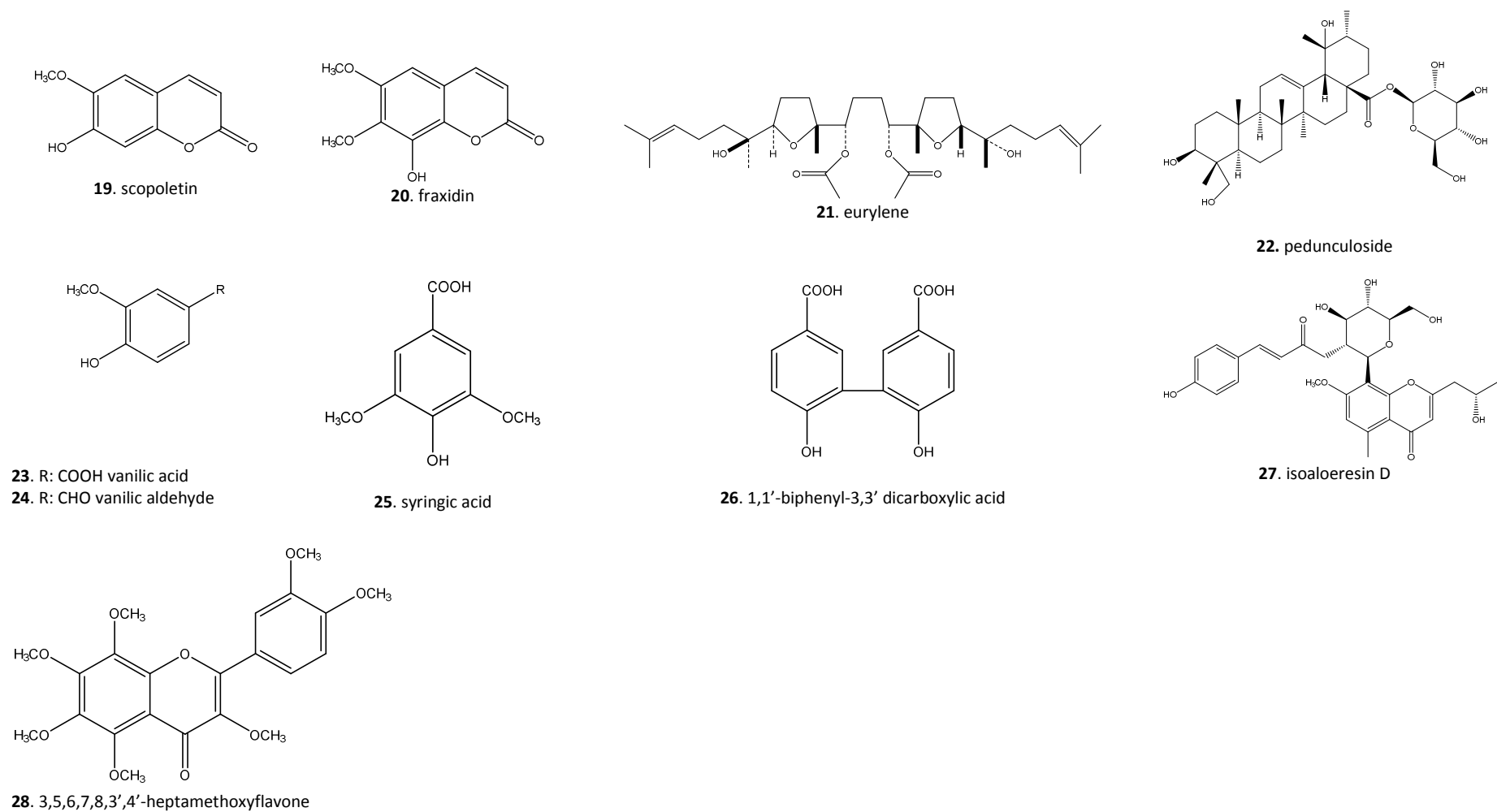


Figure S2: Isolated compounds from diethyl ether and ethyl acetate fractions of the extract of *Eurycoma longifolia*.

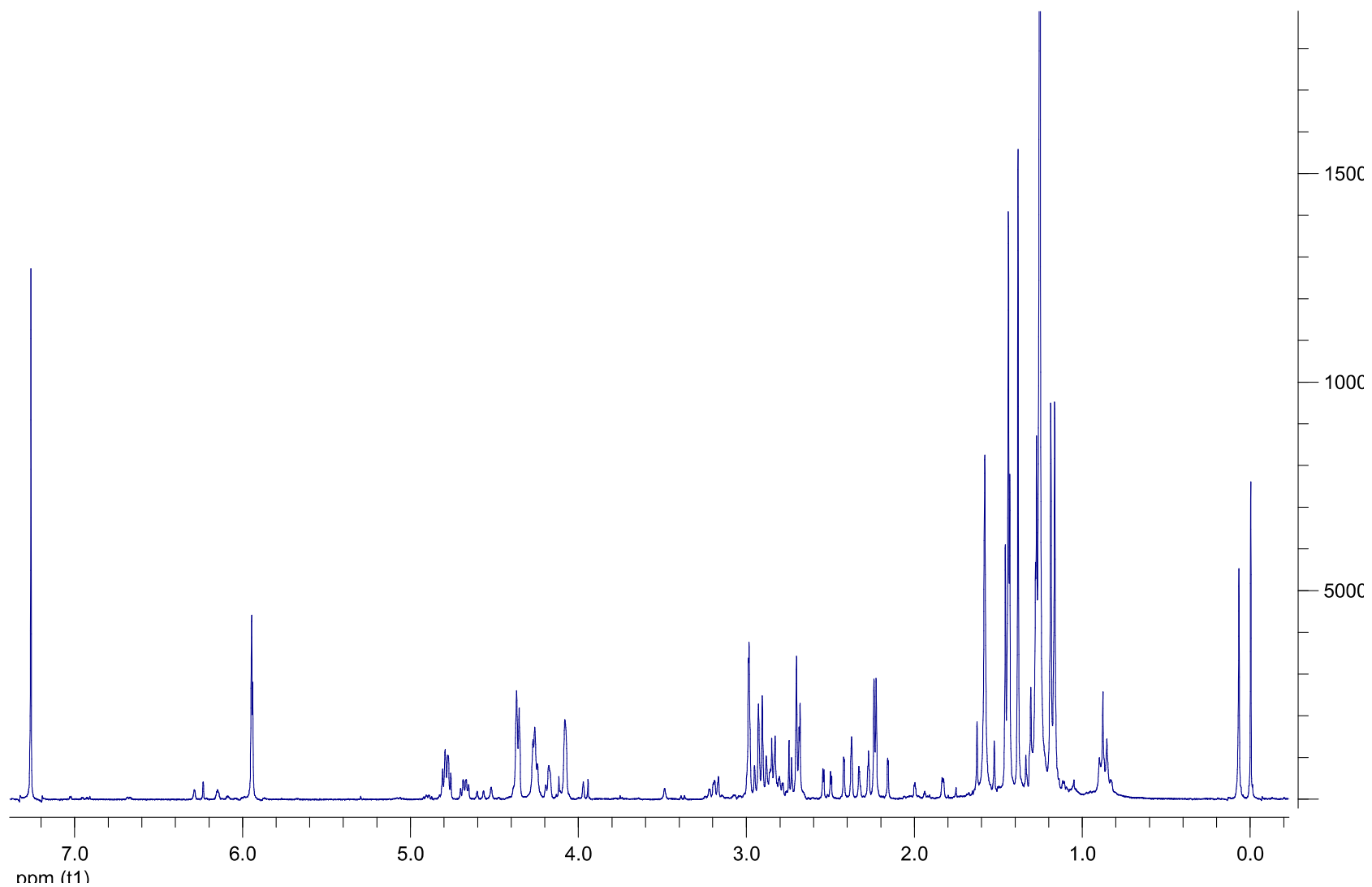


Figure S3: ^1H NMR spectrum of **1** in CDCl_3 (300 MHz).

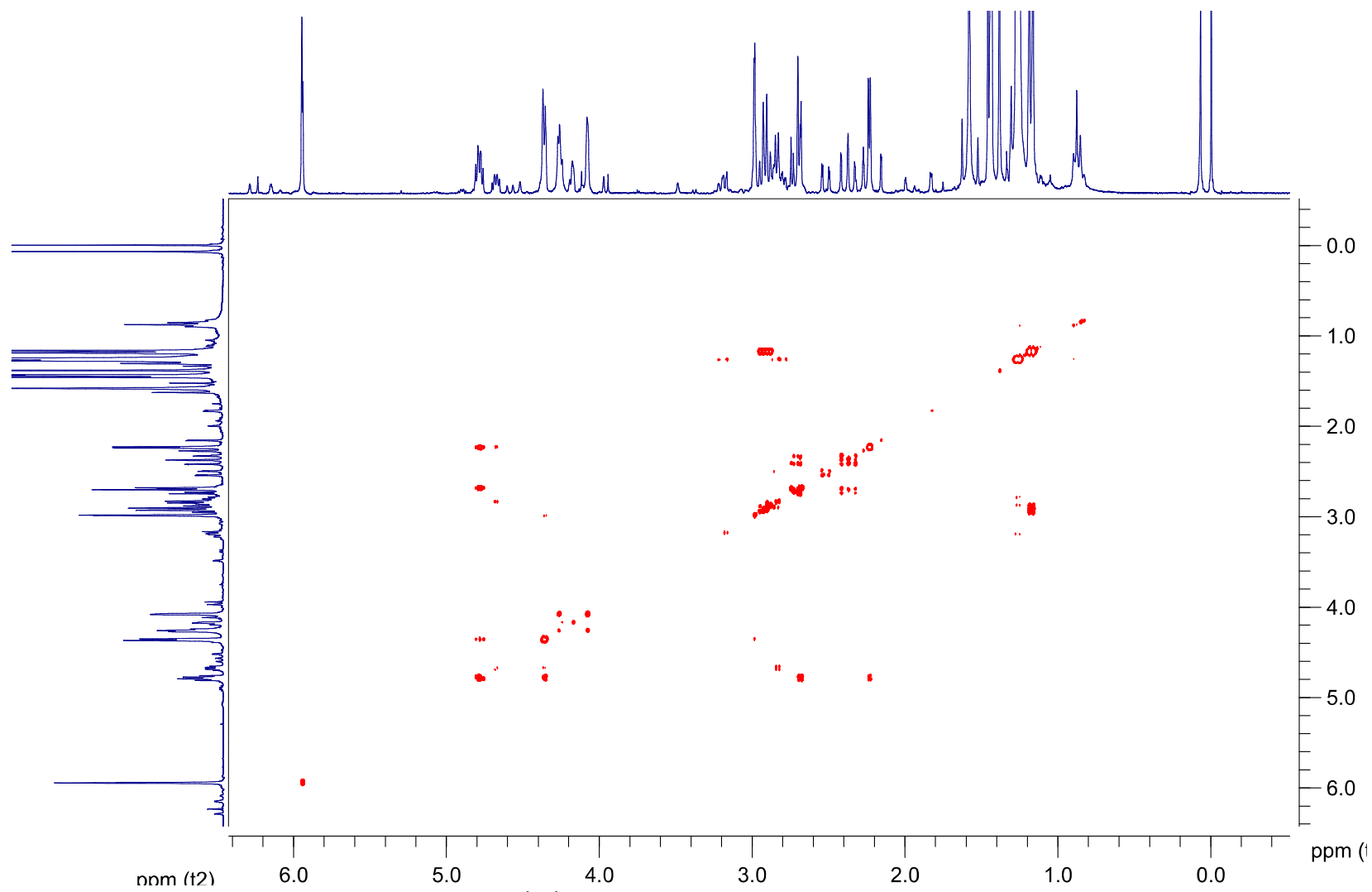


Figure S4: ^1H - ^1H COSY spectrum of **1** in CDCl_3 (300 MHz).

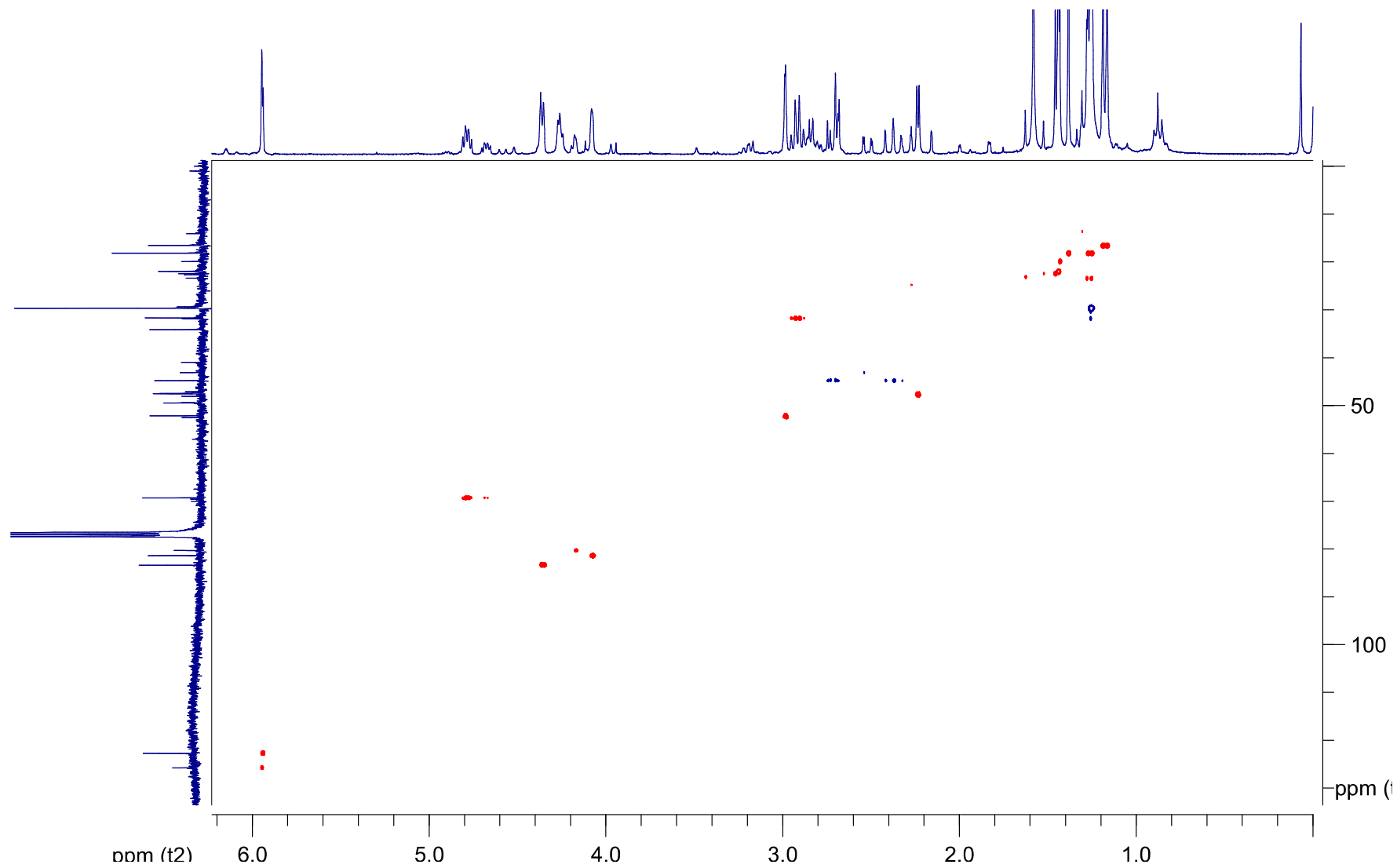


Figure S5: HSQC spectrum of **1** in CDCl_3 (300 MHz/ 75 MHz).

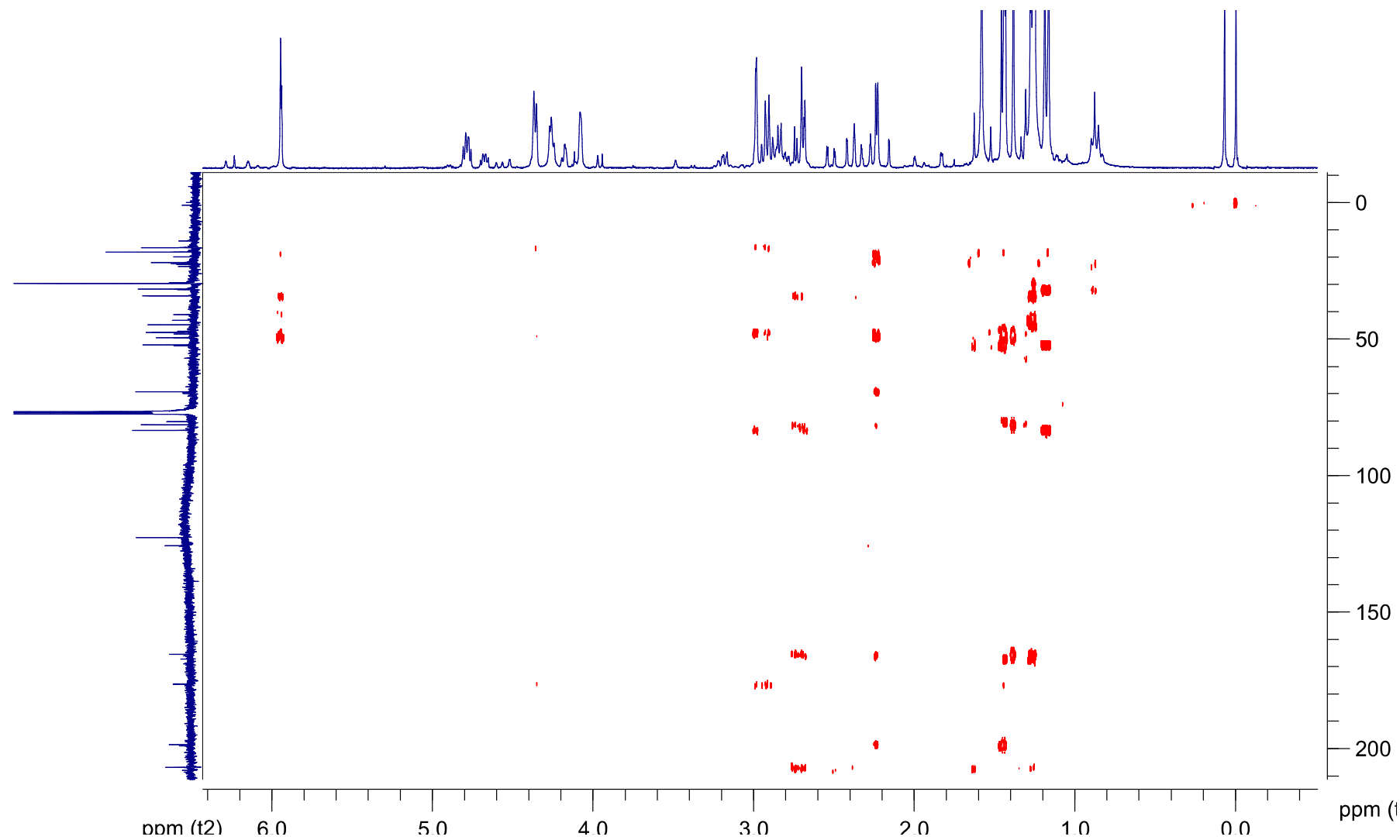


Figure S6: HMBC spectrum of **1** in CDCl₃ (300 MHz/75 MHz).

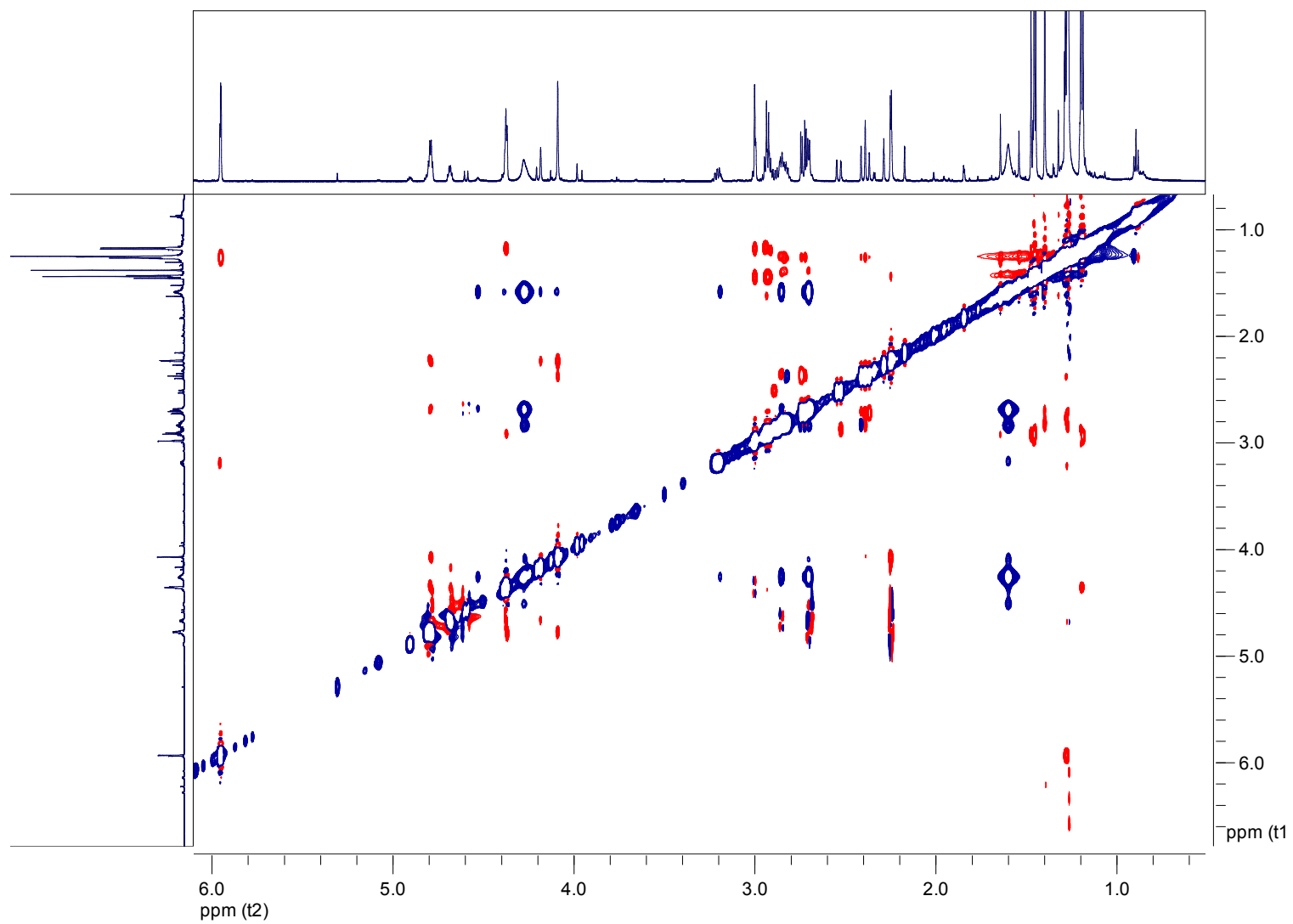


Figure S7: ROESY spectrum of **1** in CDCl₃ (600 MHz/150 MHz).

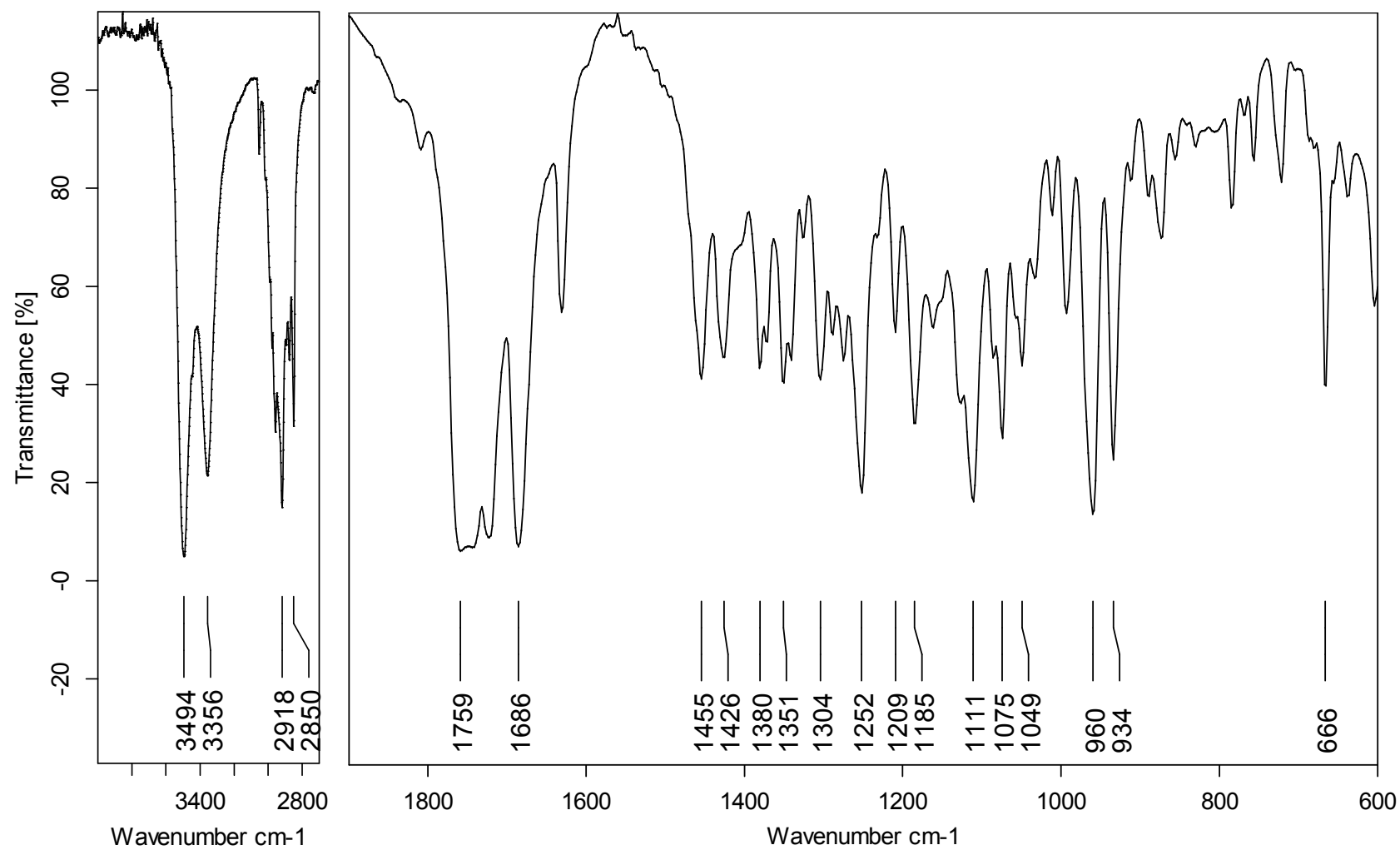


Figure S8: IR spectrum of **1**.