

Whole-Cell Transformation of Cannabidiol by Selected Filamentous Fungi into Novel Polar Derivatives

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1. Cannabidiol (1)

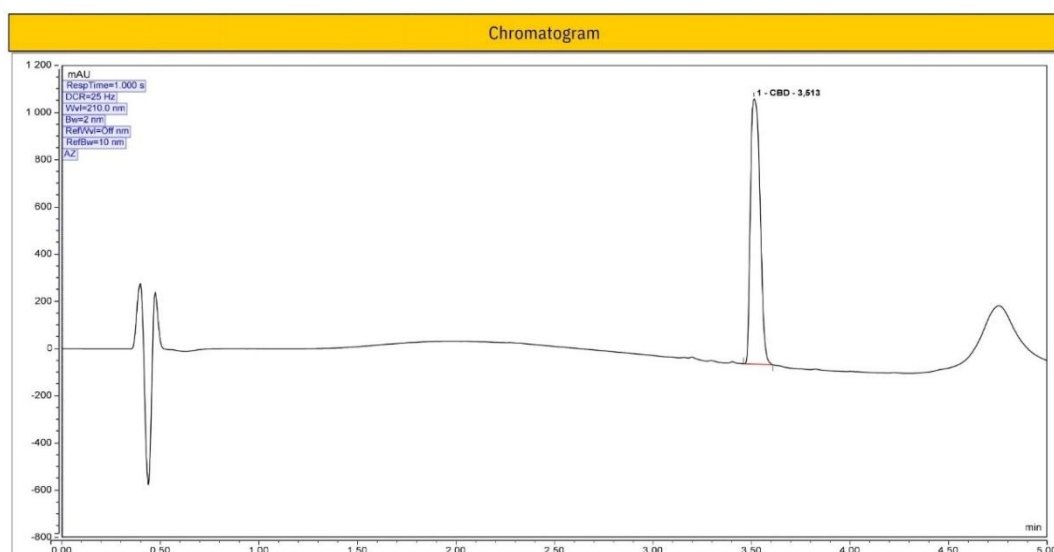


Figure S1. UHPLC-DAD chromatogram of substrate (CBD) (1)

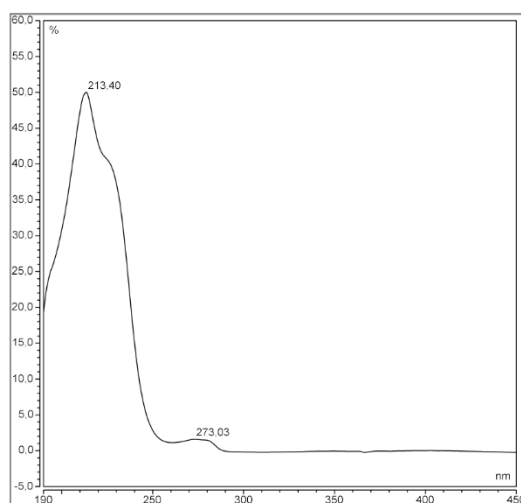


Figure S2. UV spectrum of substrate (CBD) (1)

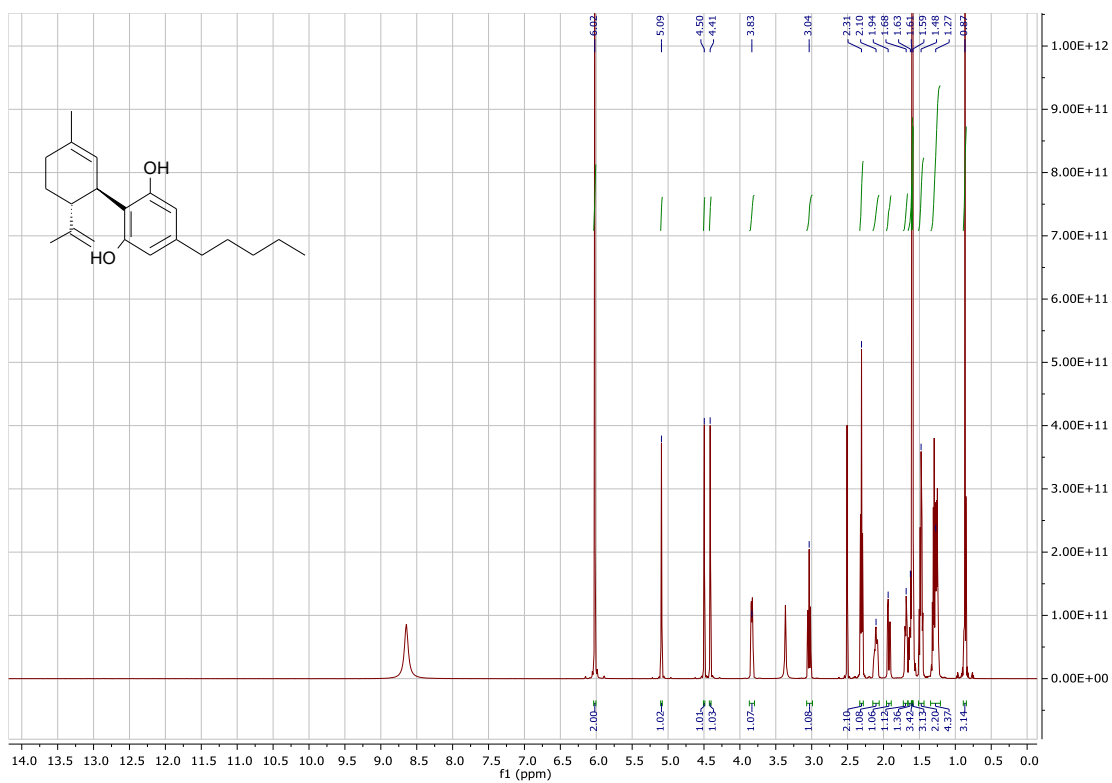


Figure S3. ^1H NMR spectrum of cannabidiol **1**

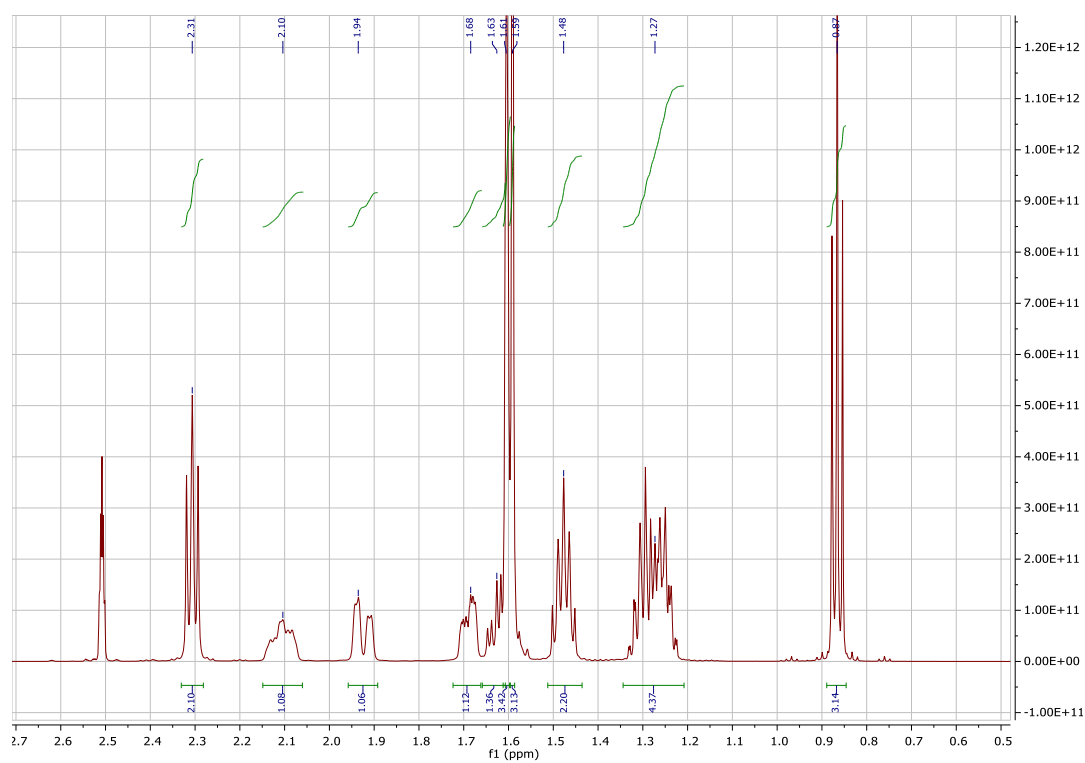


Figure S4. ^1H NMR (0.5-2.5 ppm) spectrum of cannabidiol **1**

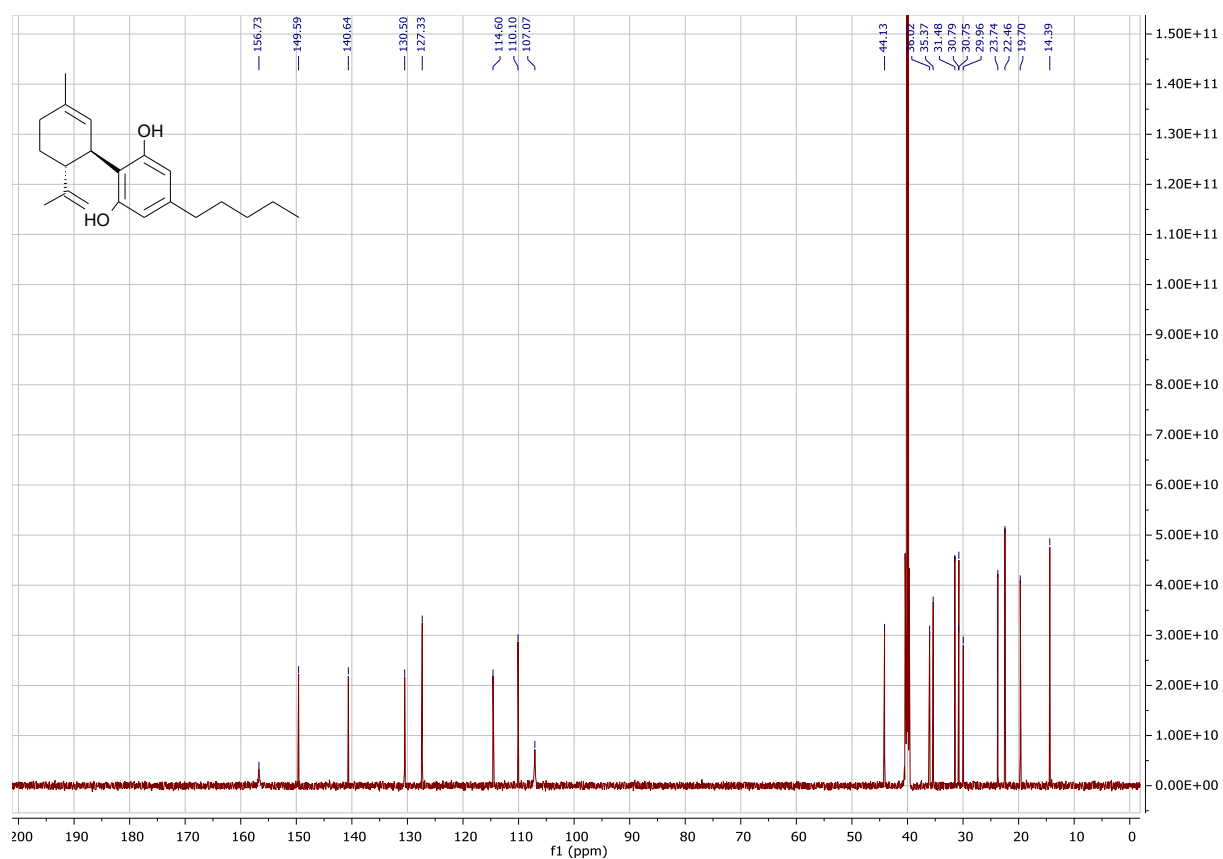


Figure S5. ^{13}C NMR spectrum of cannabidiol **1**

2. Metabolite 2: 2'-O-(β -D-glucopyranosyl)-cannabidiol

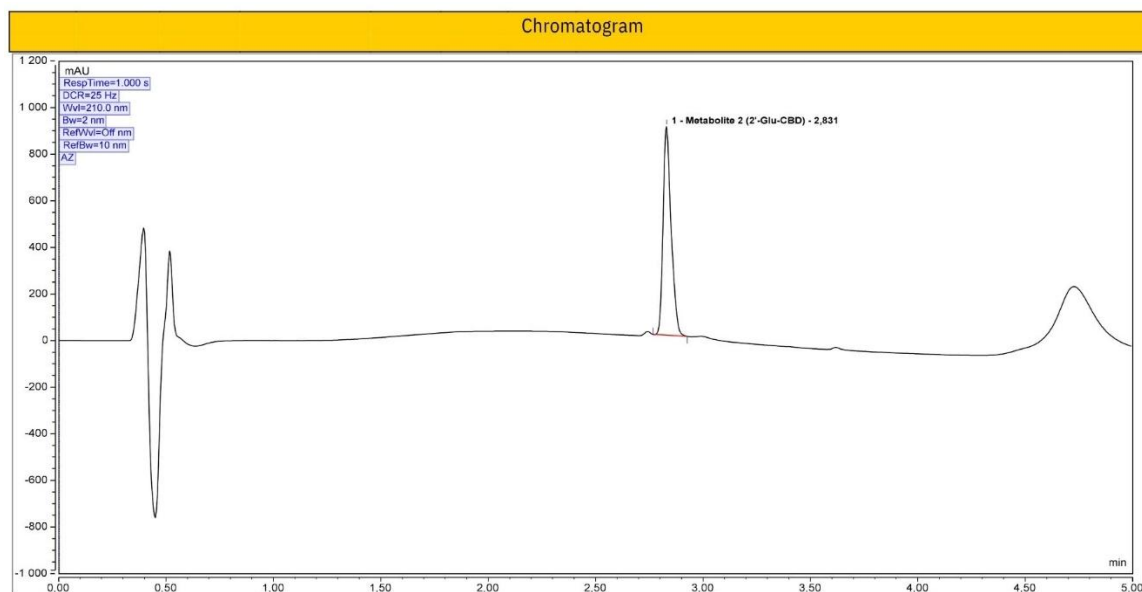


Figure S6. UHPLC-DAD chromatogram of metabolite 2

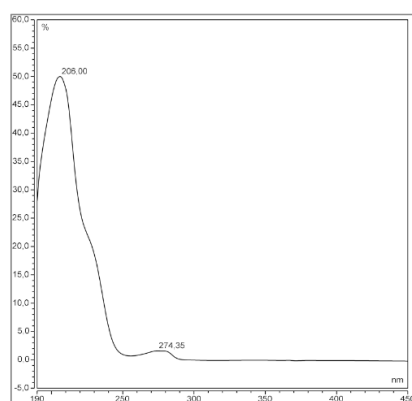


Figure S7. UV spectrum of metabolite 2

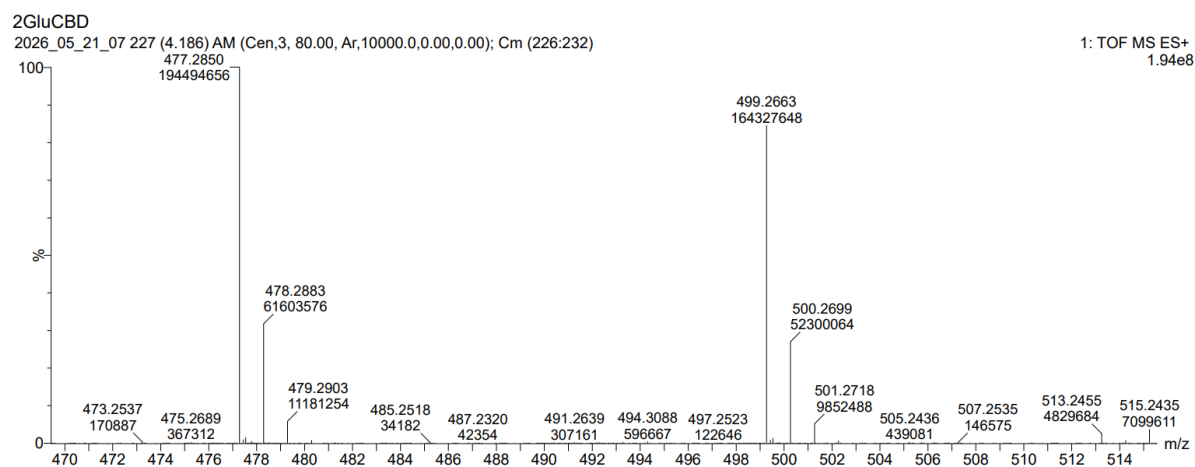


Figure S8. Mass spectrum of metabolite 2

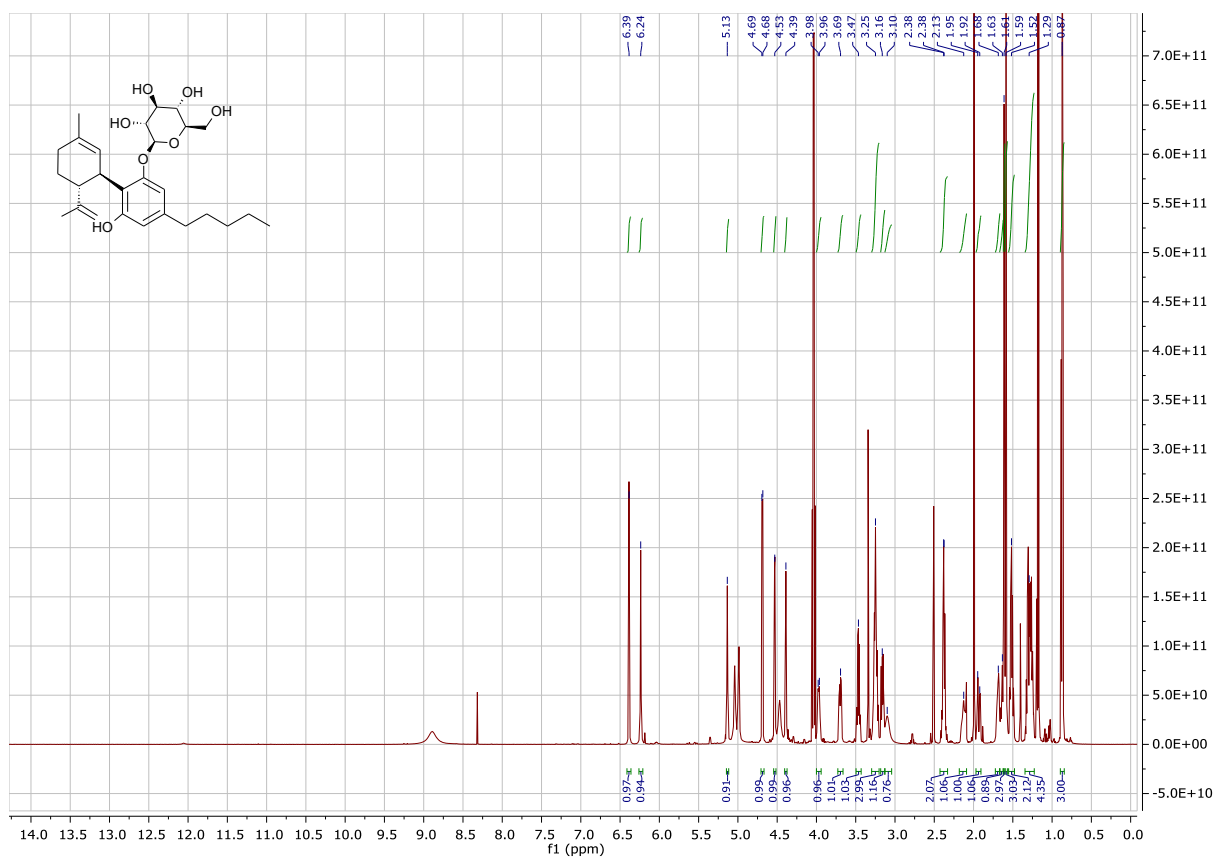


Figure S9. ^1H NMR spectrum of metabolite 2

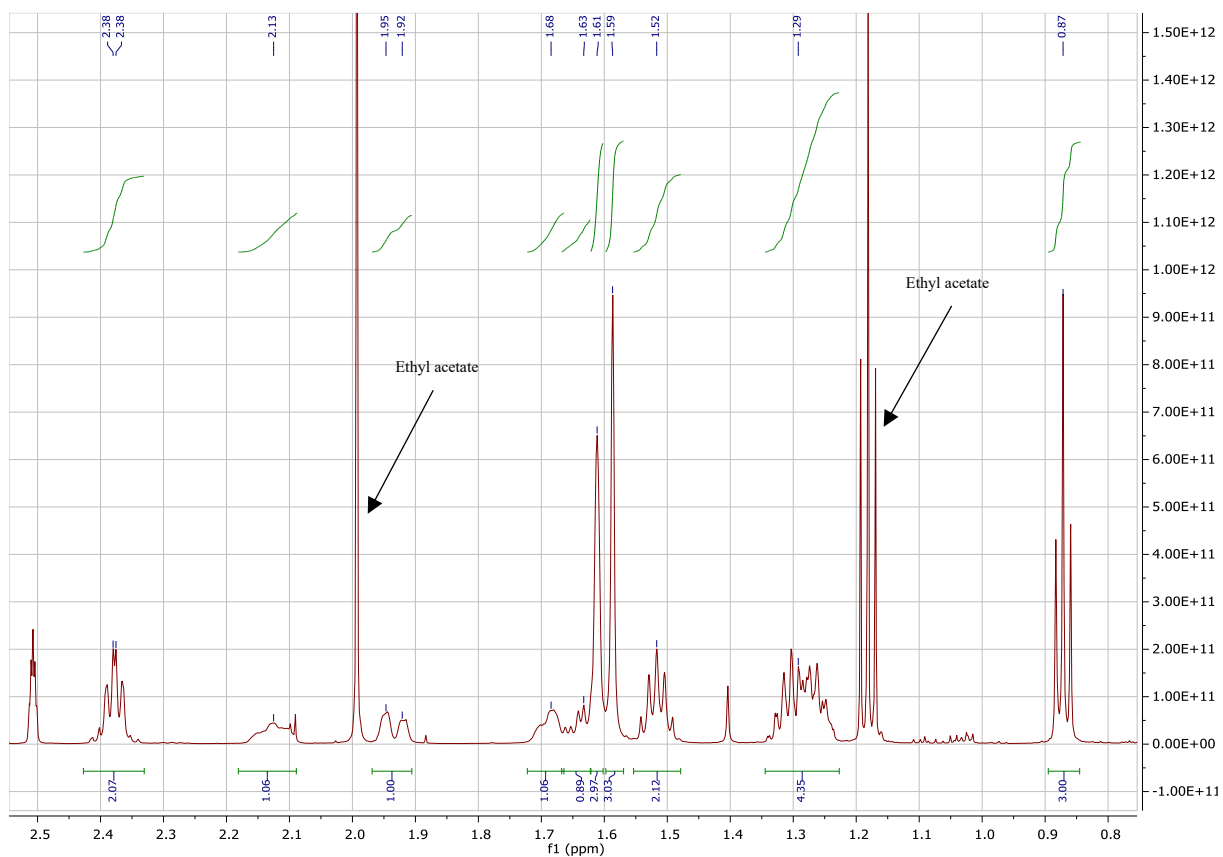


Figure S10. ^1H NMR spectrum of metabolite 2 (0.8-2.5 ppm)

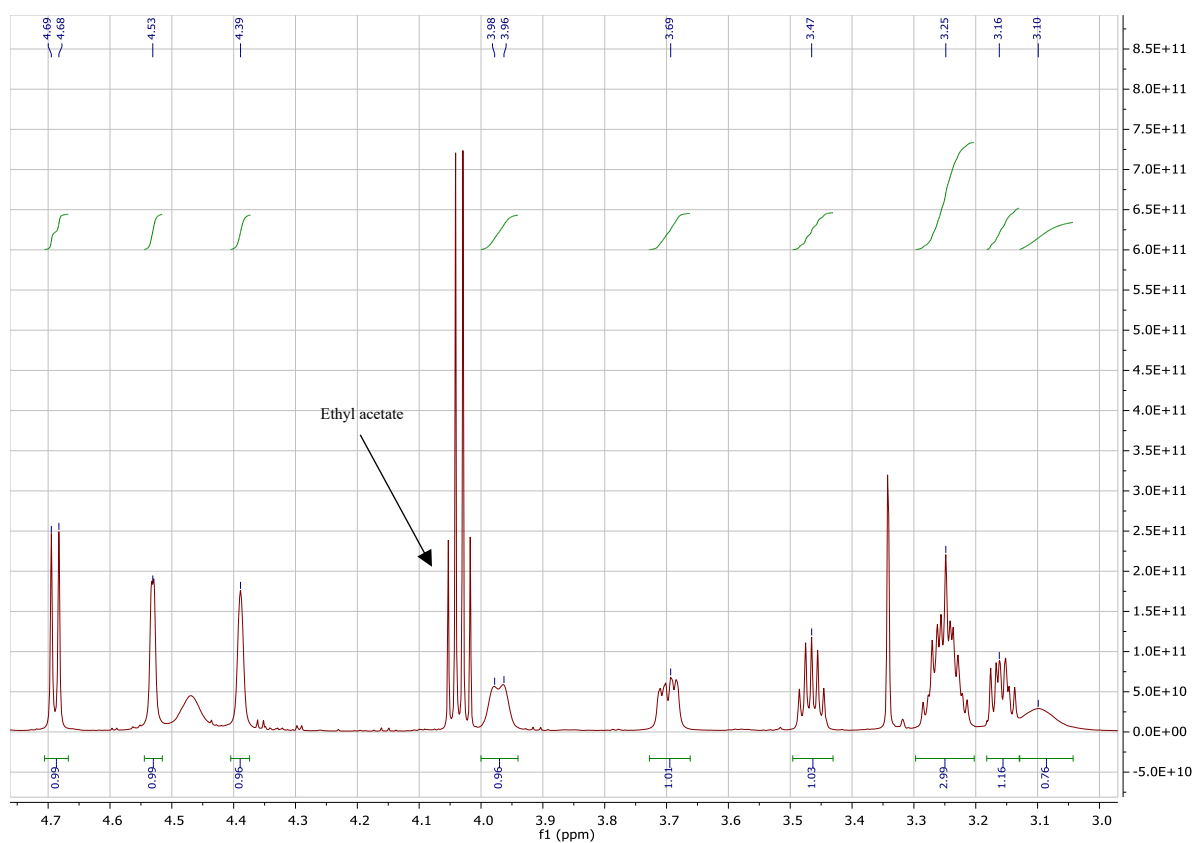


Figure S11. ¹H NMR spectrum of metabolite 2 (2.6-6.4 ppm)

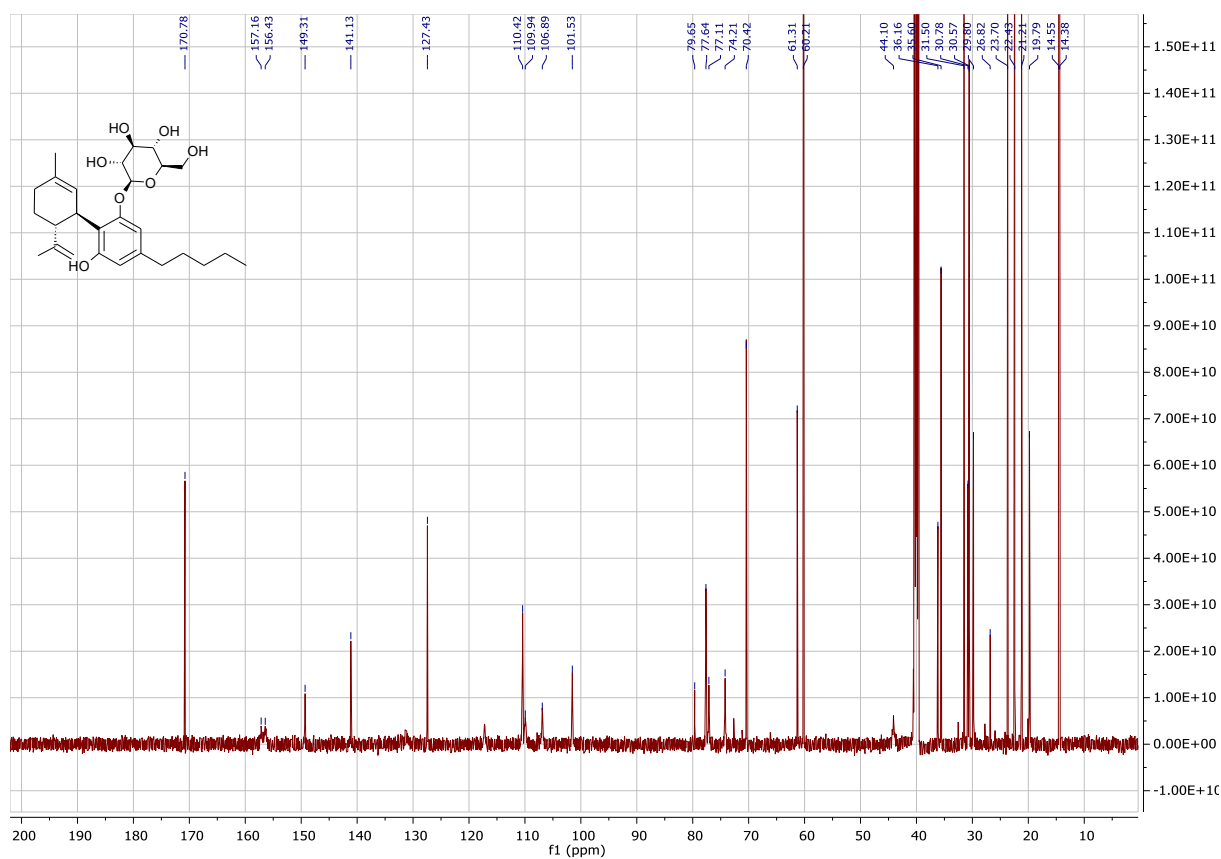


Figure S12. ¹³C NMR spectrum of metabolite 2

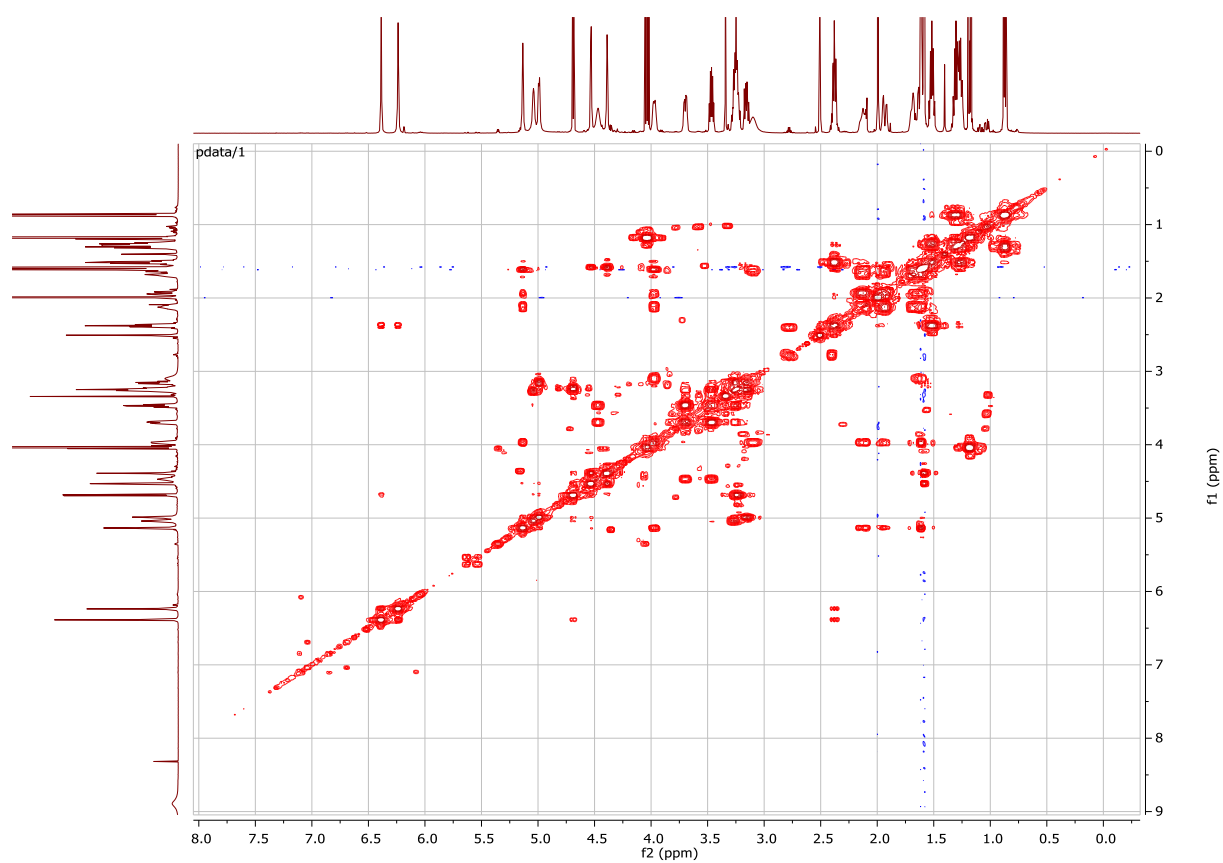


Figure S13. COSY (^1H - ^1H) NMR spectrum of metabolite 2

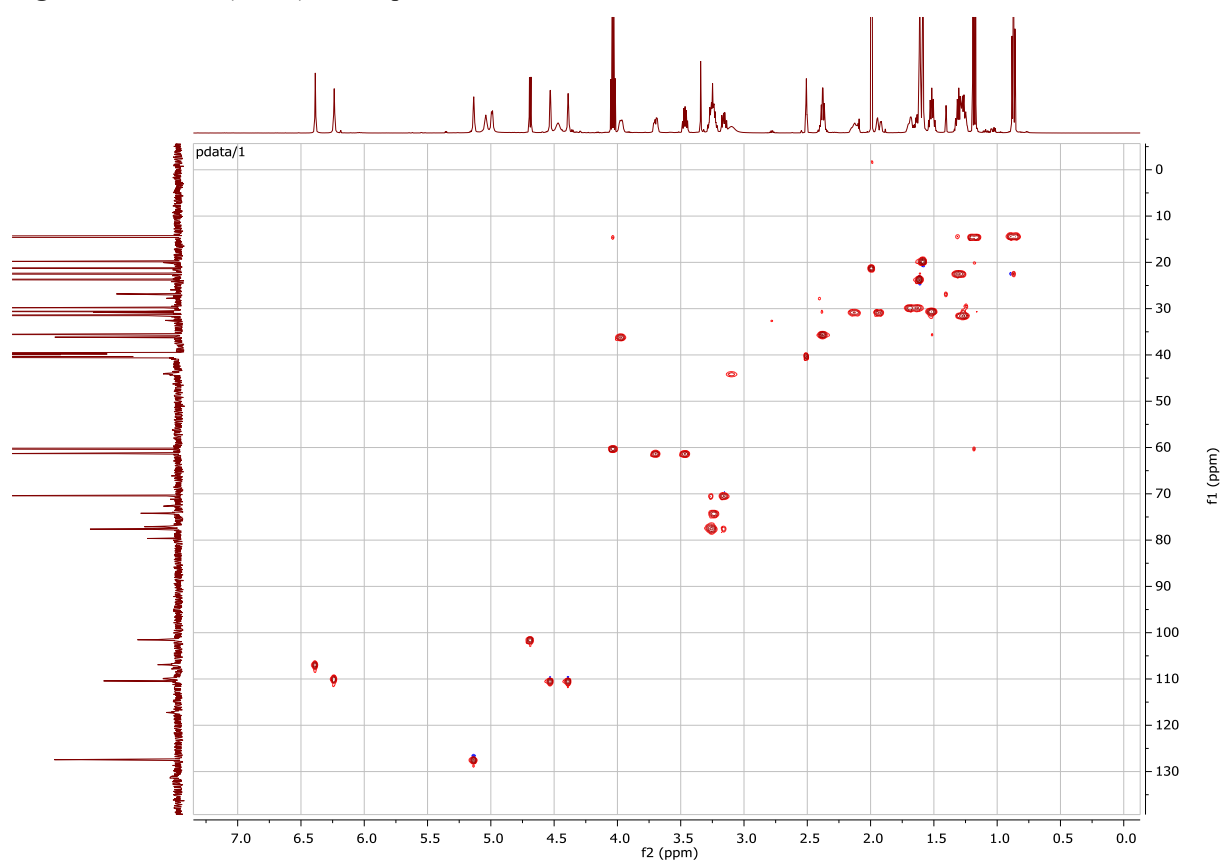


Figure S14. HSQC (^1H - ^{13}C) NMR spectrum of metabolite 2

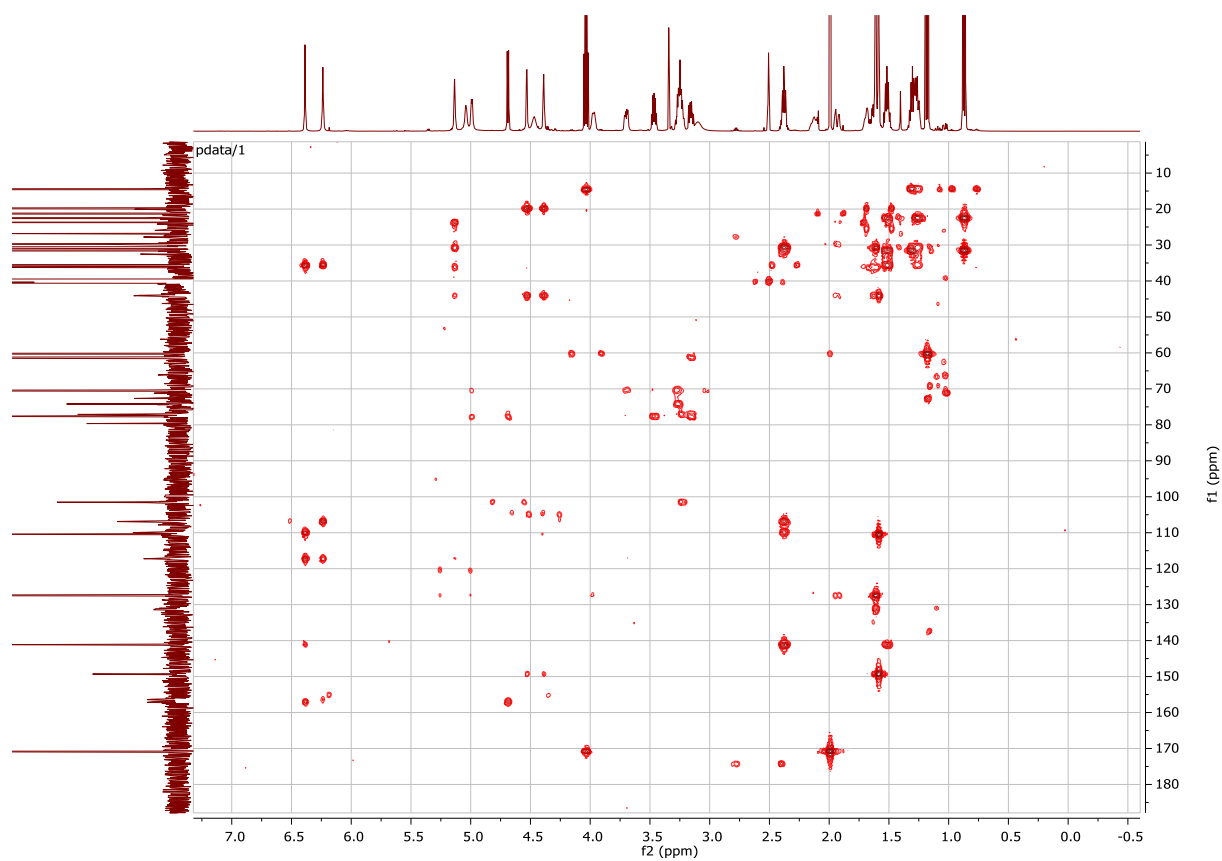


Figure S15. HMBC (long distance correlation ^1H - ^{13}C) spectrum of metabolite 2

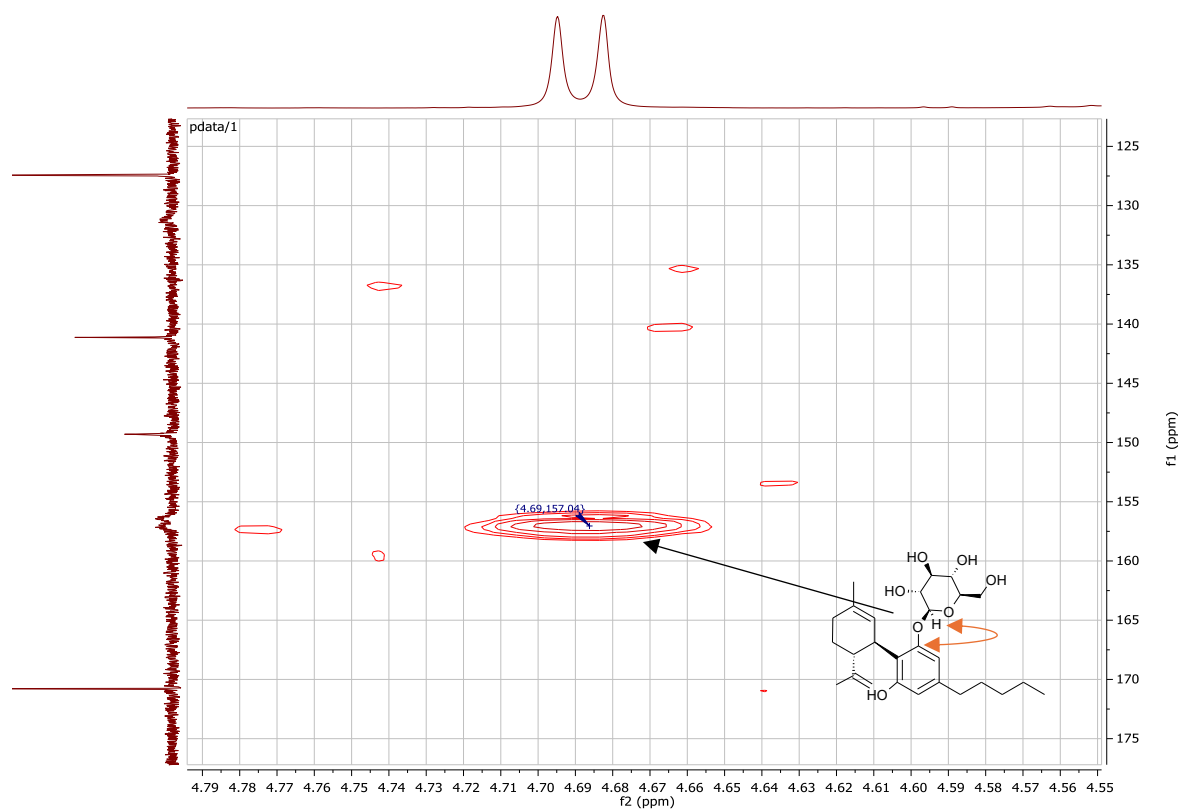


Figure S16. HMBC correlation between C1 carbon of glucose and C2 carbon of aromatic ring.

3. Metabolite 3: 2'-O-(β -D-glucopyranosyl)-4''-hydroxycannabidiol

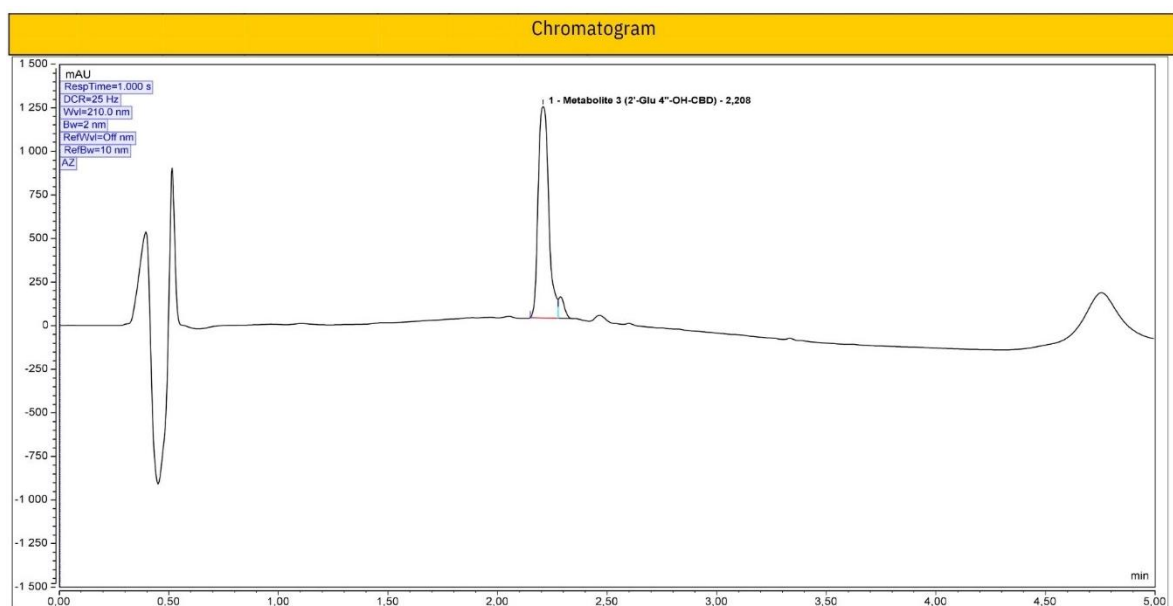


Figure S17. UHPLC-DAD chromatogram of metabolite 3

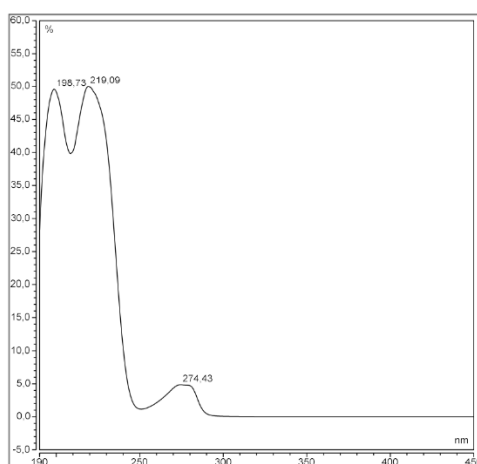


Figure S18. UV spectrum of metabolite 3

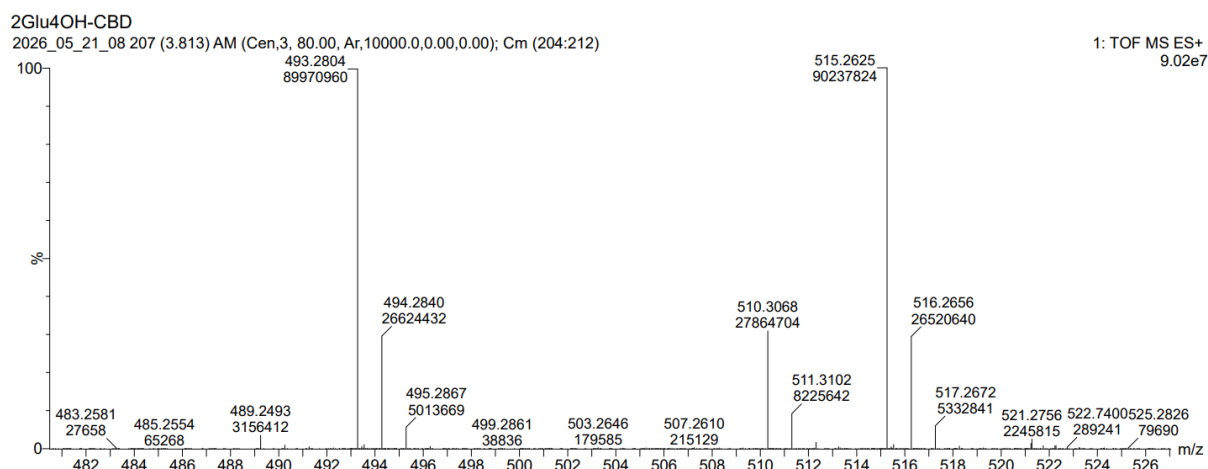


Figure S19. Mass spectrum of metabolite 3

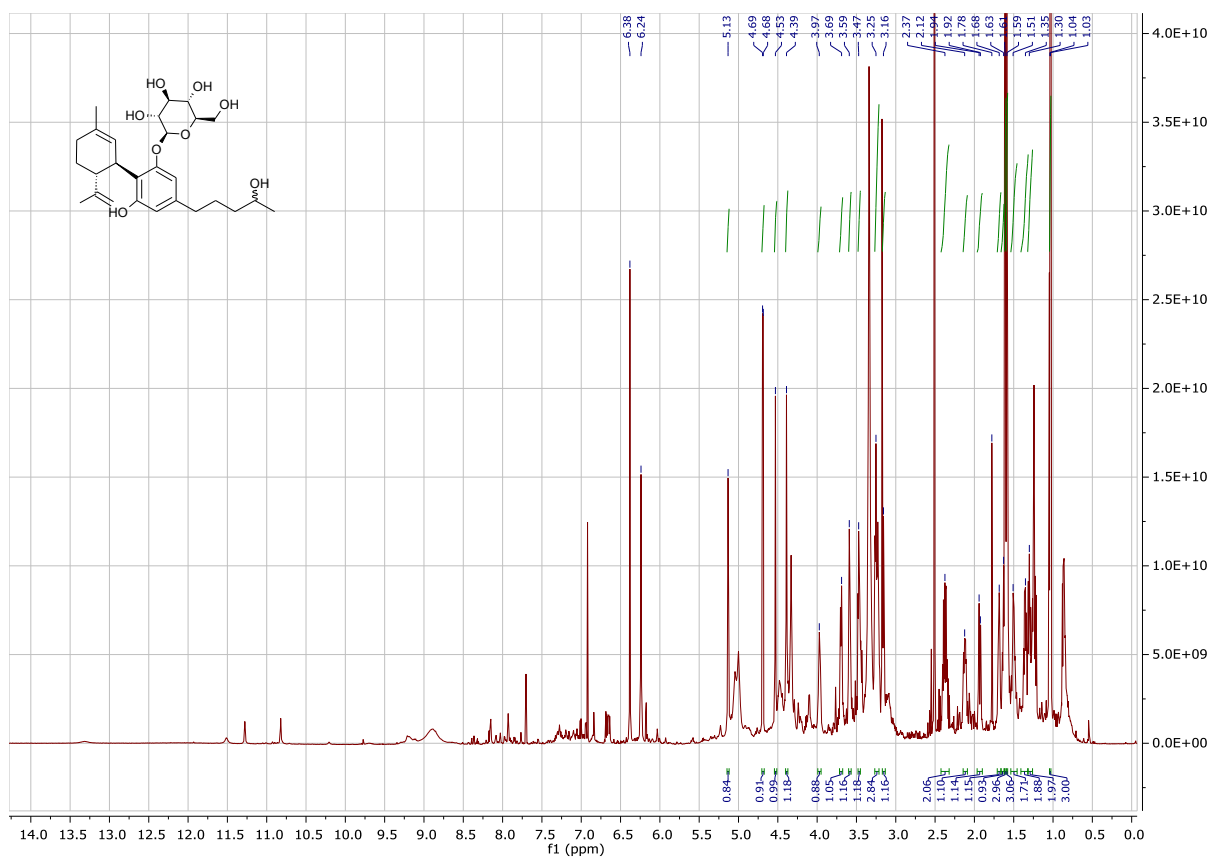


Figure S20. ¹H NMR spectrum of metabolite 3

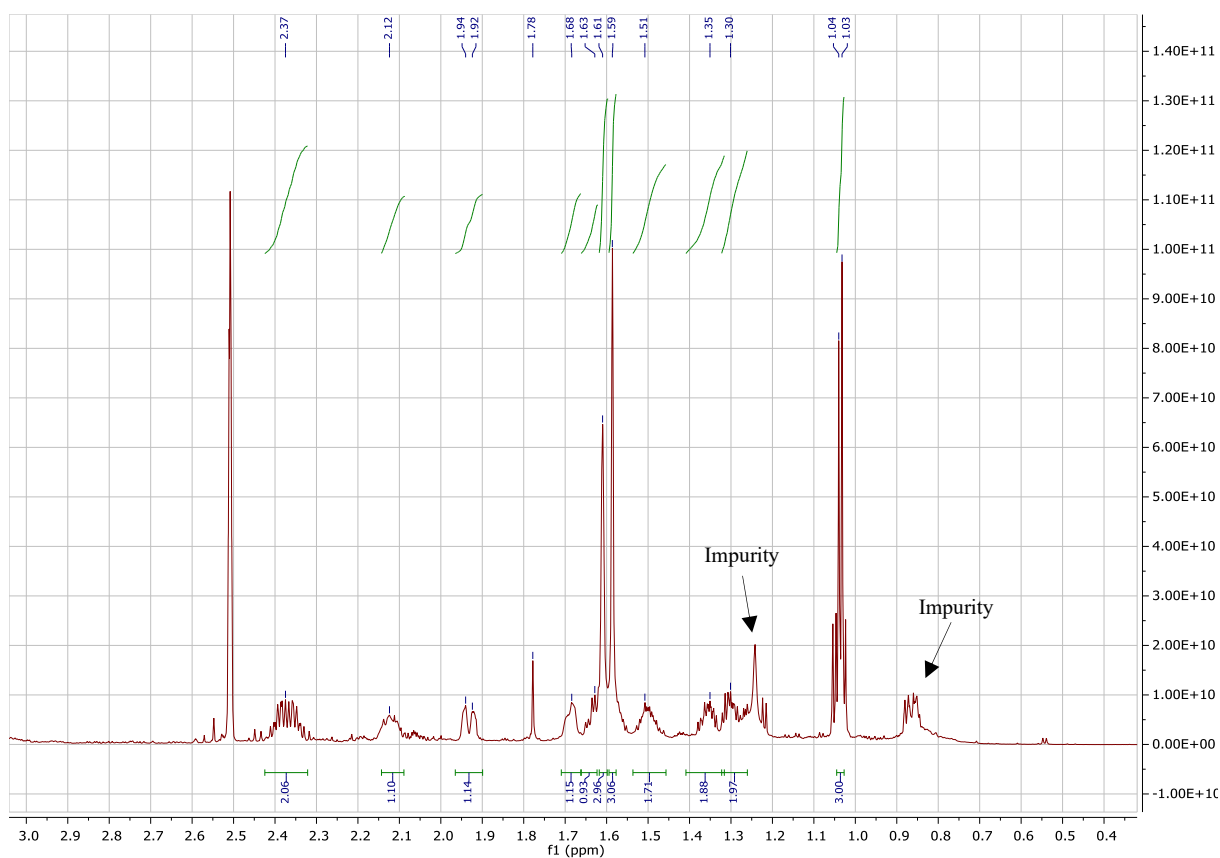


Figure S21. ^1H NMR (0.4-3.0ppm) spectrum of metabolite **3**

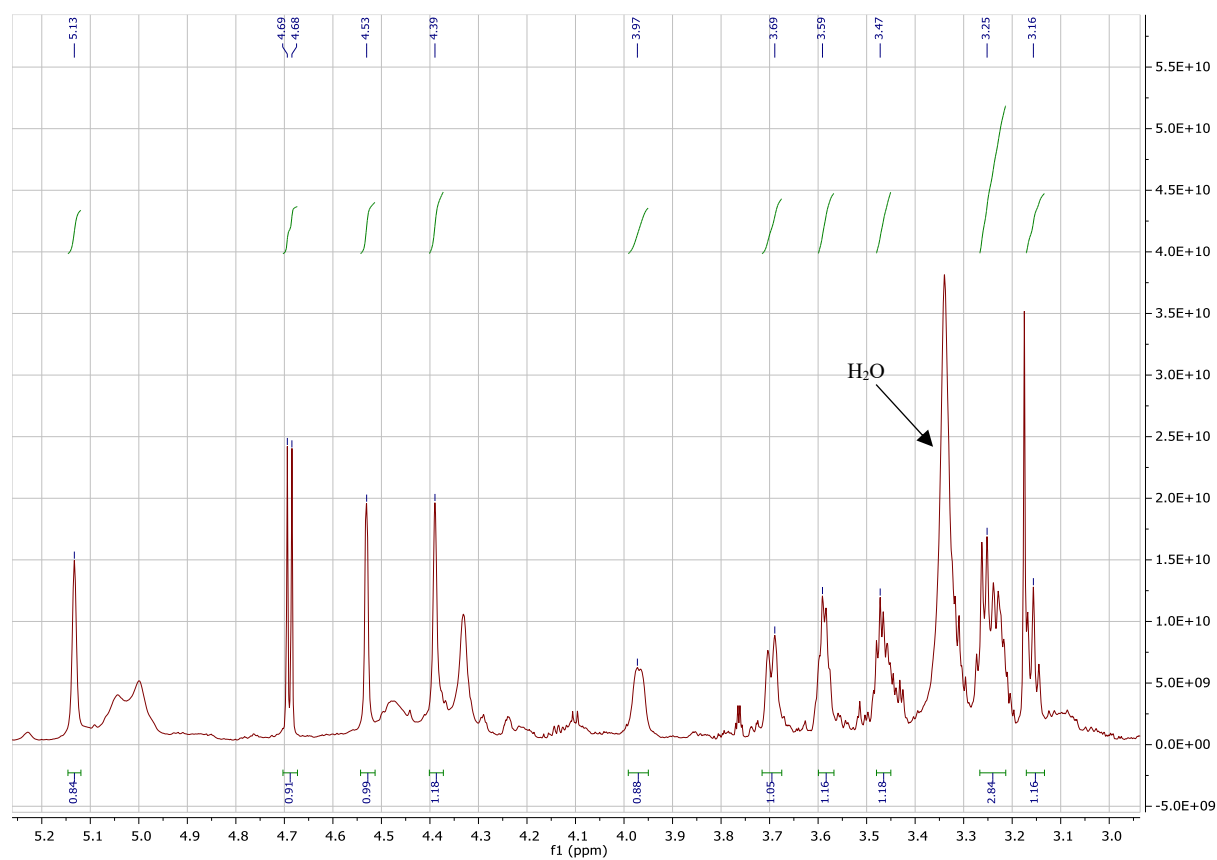


Figure S22. ^1H NMR spectrum of metabolite **3** (3.0-5.2 ppm)

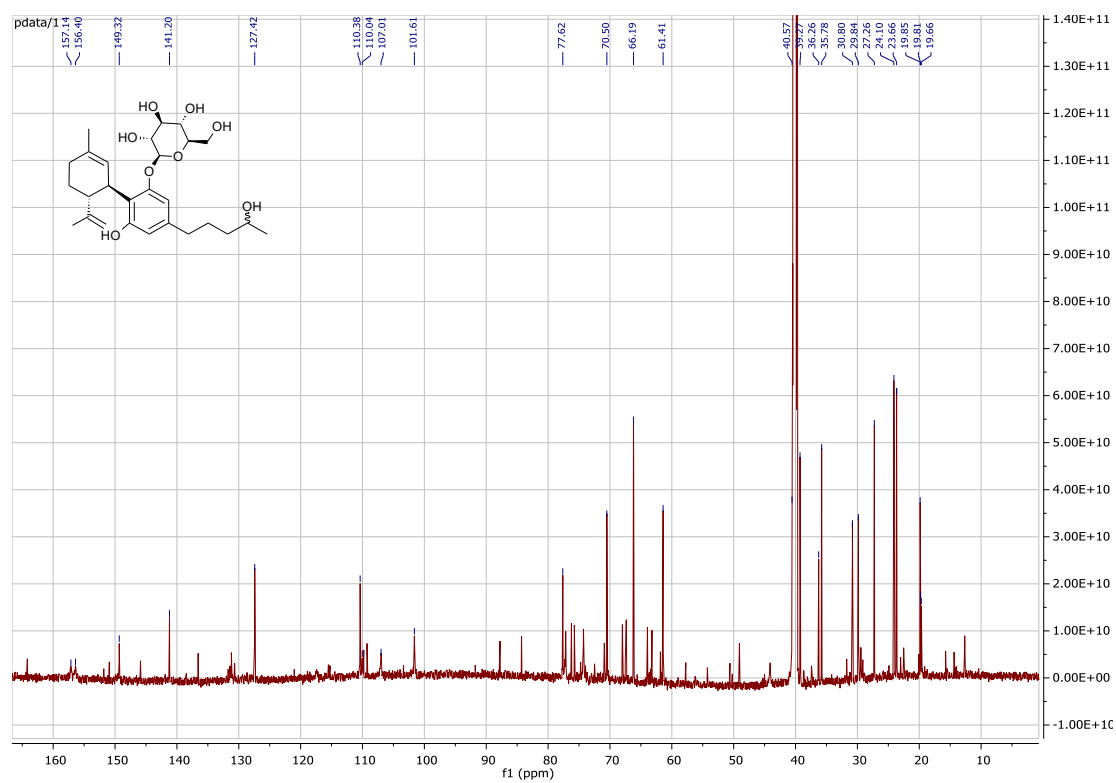


Figure S23. ^{13}C NMR spectrum of metabolite **3**

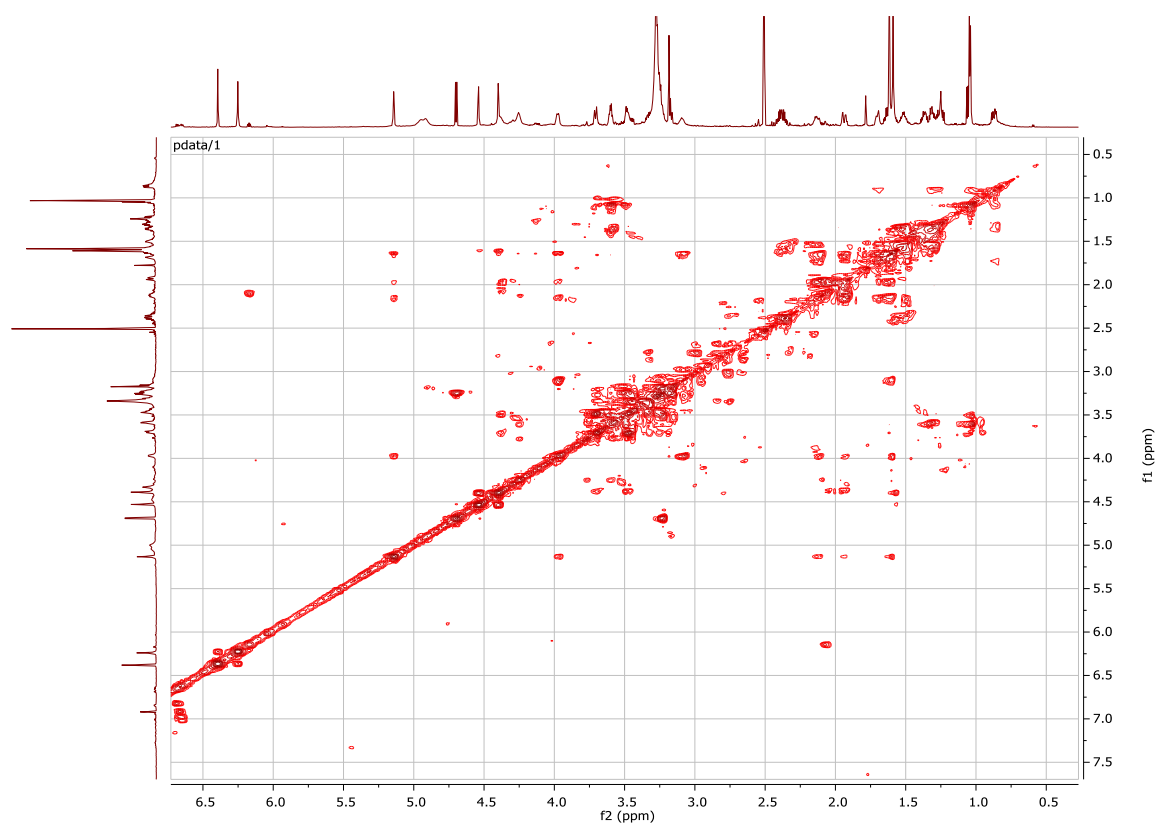


Figure S24. COSY (^1H - ^1H) NMR spectrum of metabolite **3**

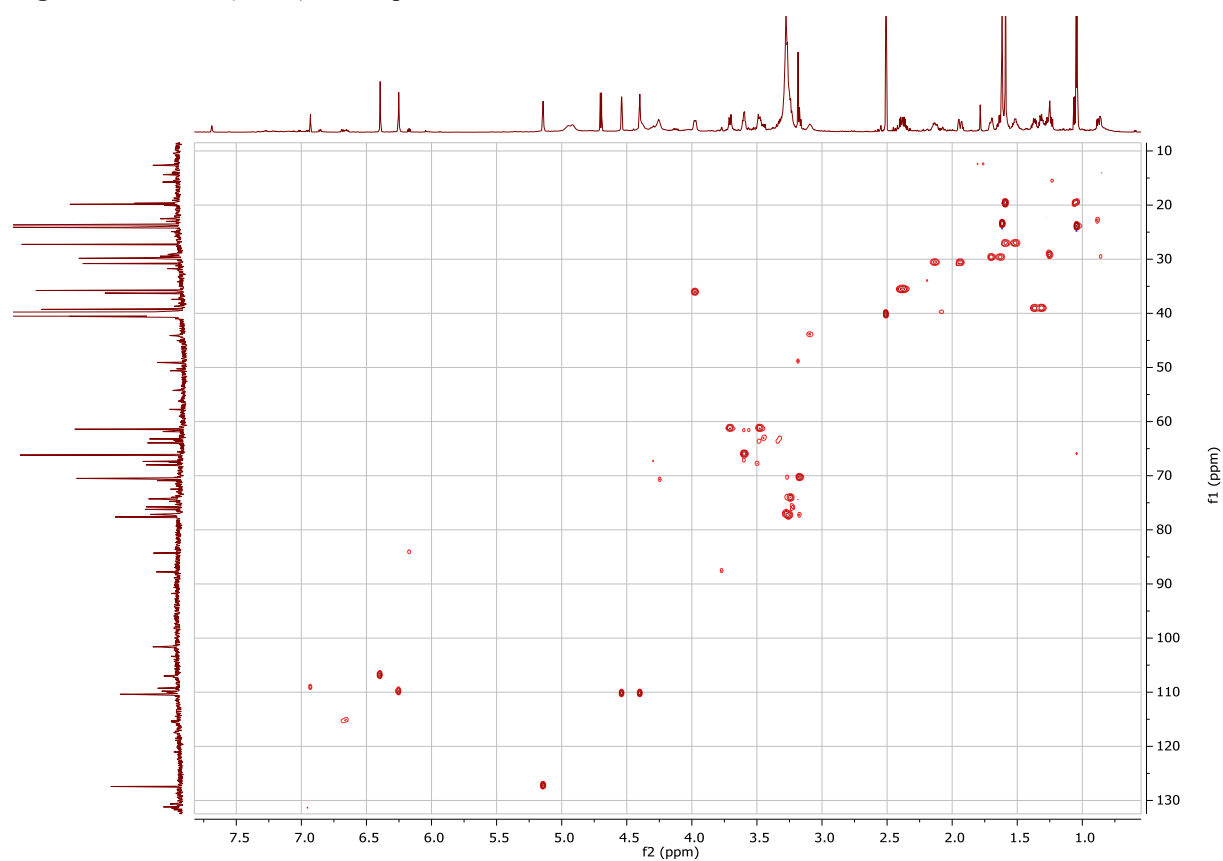


Figure S25. HSQC (^1H - ^{13}C) NMR spectrum of metabolite **3**

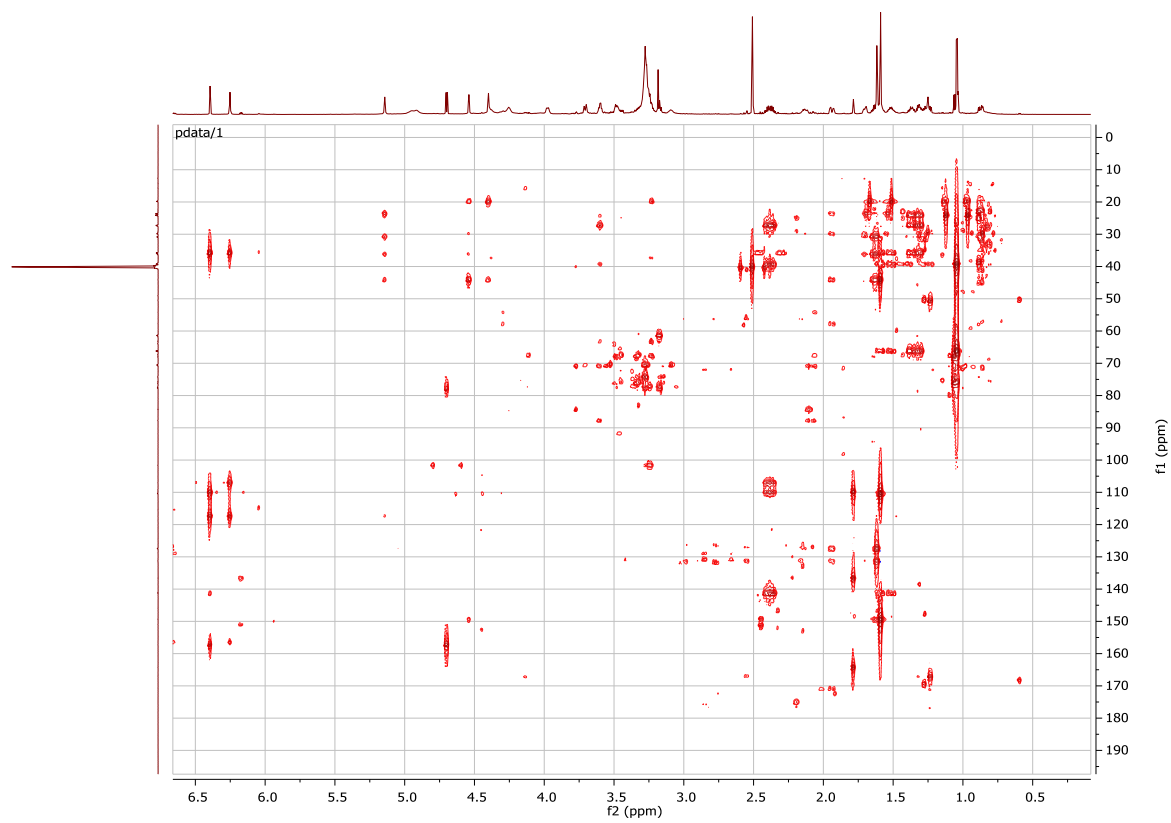


Figure S26. HMBC (long distance correlation ^1H - ^{13}C) spectrum of metabolite **3**

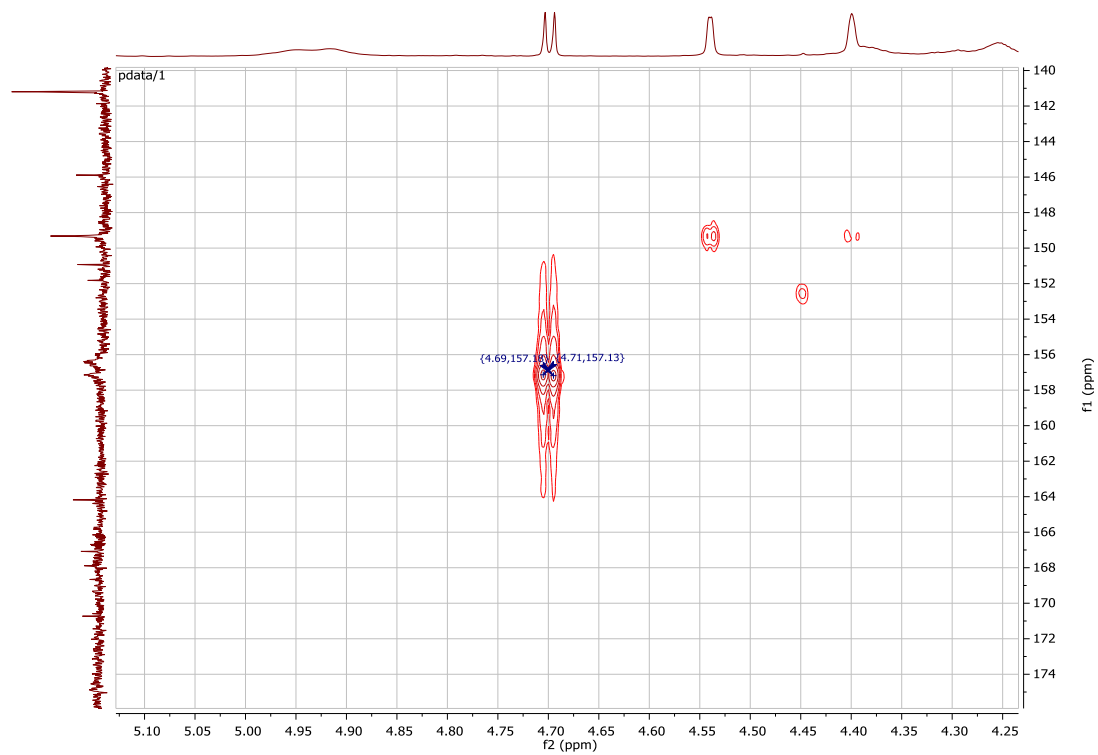


Figure S27. HMBC correlation between C1 carbon of glucose and C2 carbon of aromatic ring in metabolite **3**

4. Metabolite 4: 4''-hydroxycannabidiol

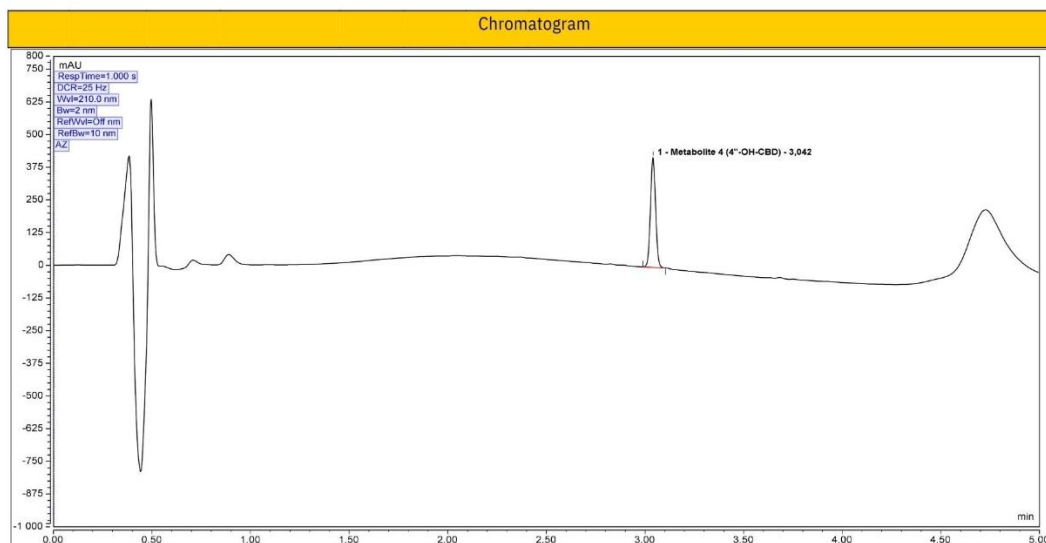


Figure S28. UHPLC-DAD chromatogram of metabolite **4**

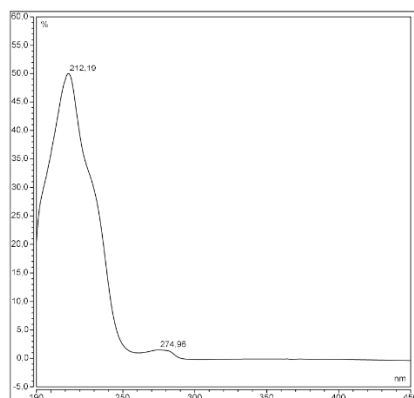


Figure S29. UV spectrum of metabolite **4**

4OH-CBD

2026_05_21_09 235 (4.332) Cm (232:241)

1: TOF MS ES+
1.41e8

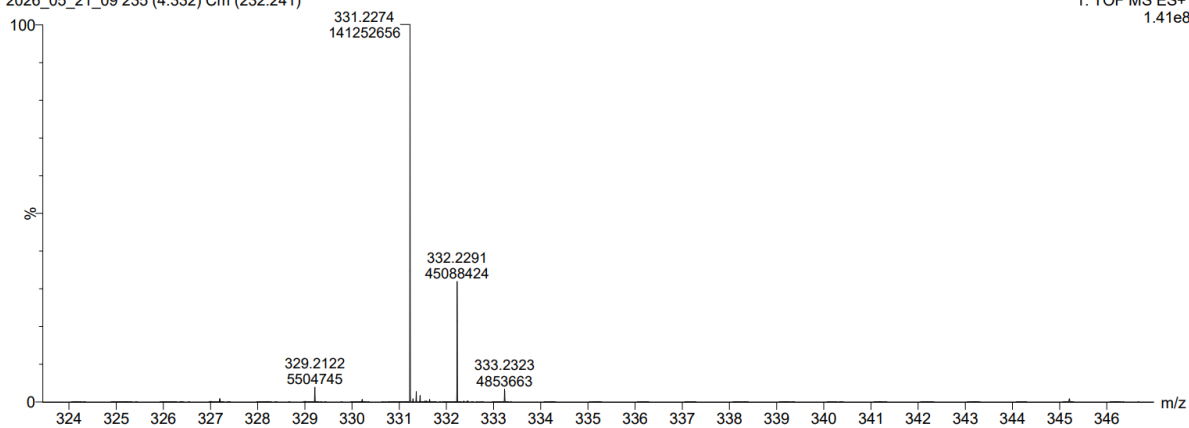


Figure S30. Mass spectrum of metabolite **4**

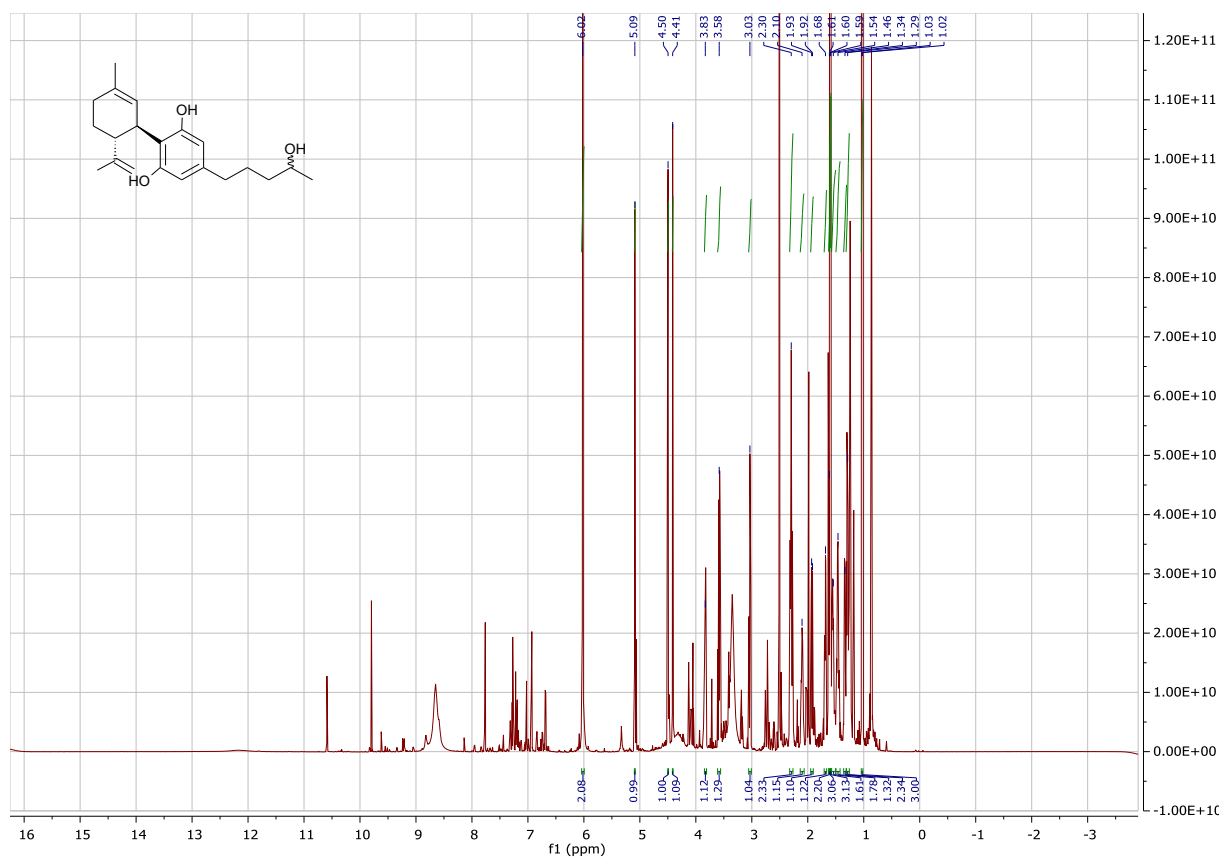


Figure S31. ^1H NMR spectrum of metabolite 4

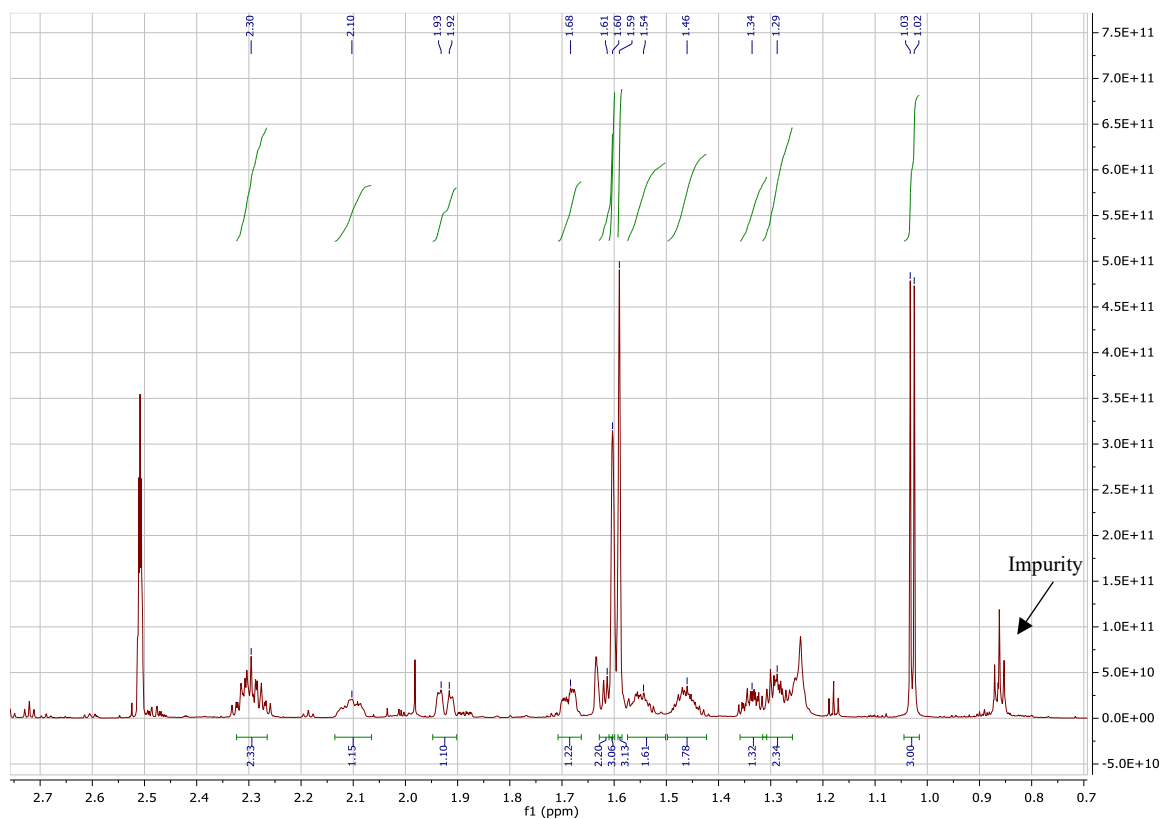


Figure S32. ^1H NMR (0.7-2.5) spectrum of metabolite 4

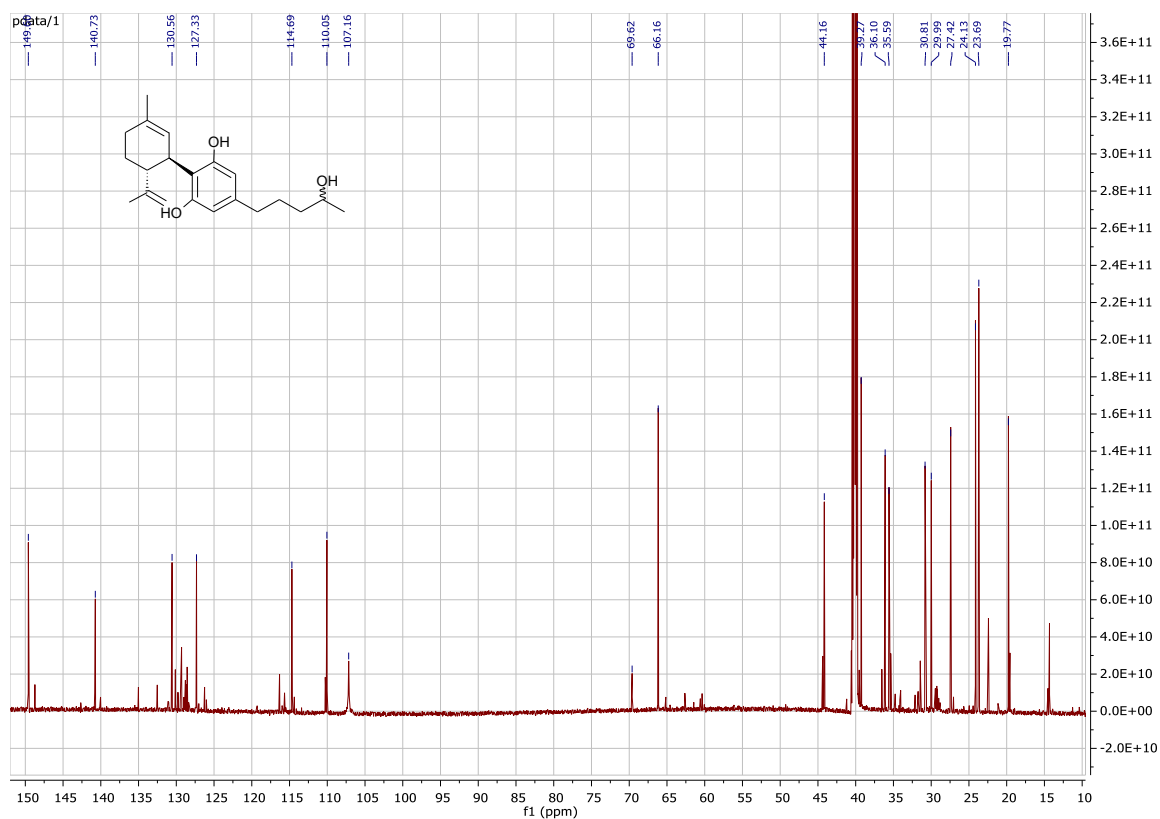


Figure S33. ^{13}C NMR spectrum of metabolite 4

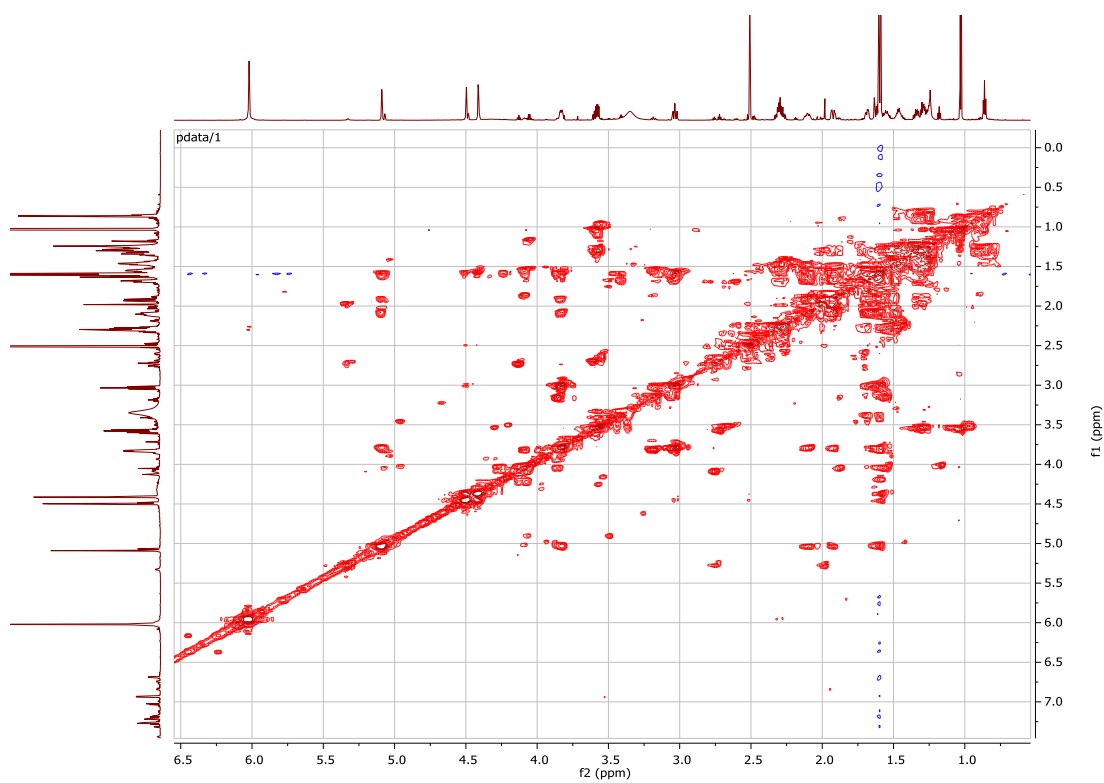


Figure S34. COSY (^1H - ^1H) NMR spectrum of metabolite 4

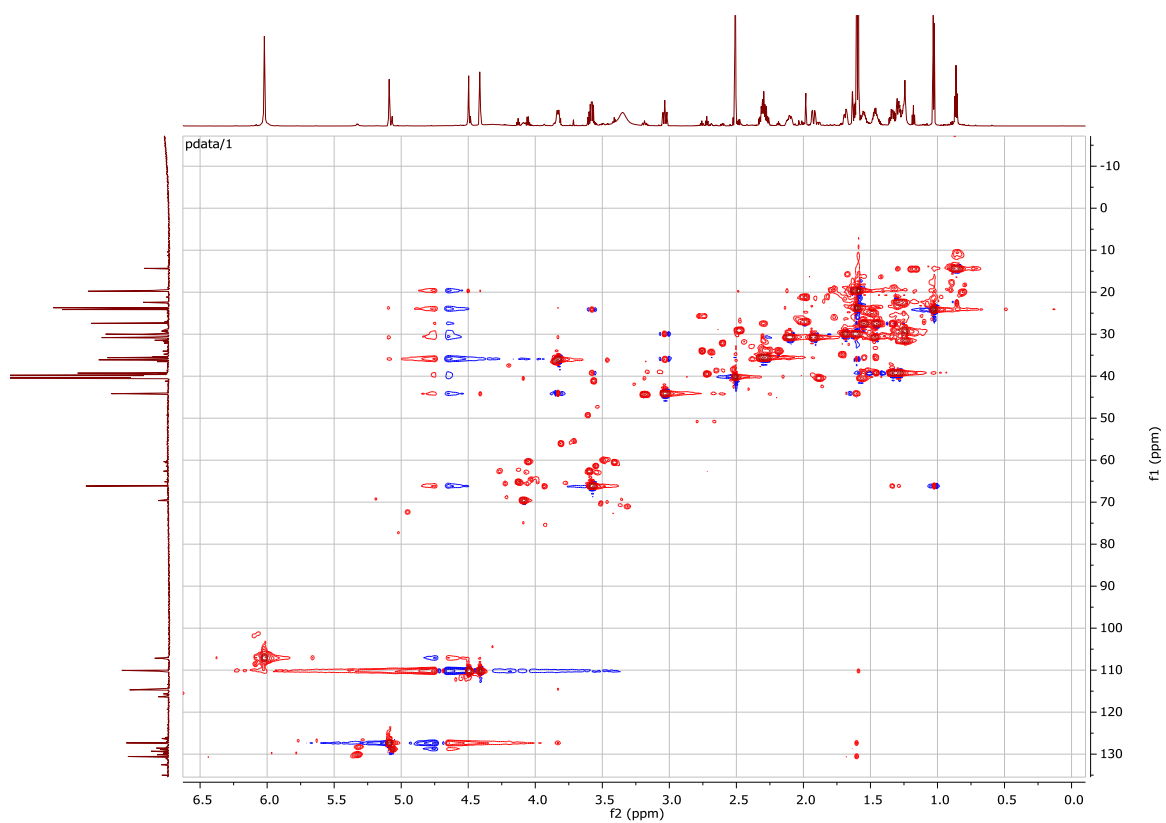


Figure S35. HSQC (^1H - ^{13}C) NMR spectrum of metabolite 4

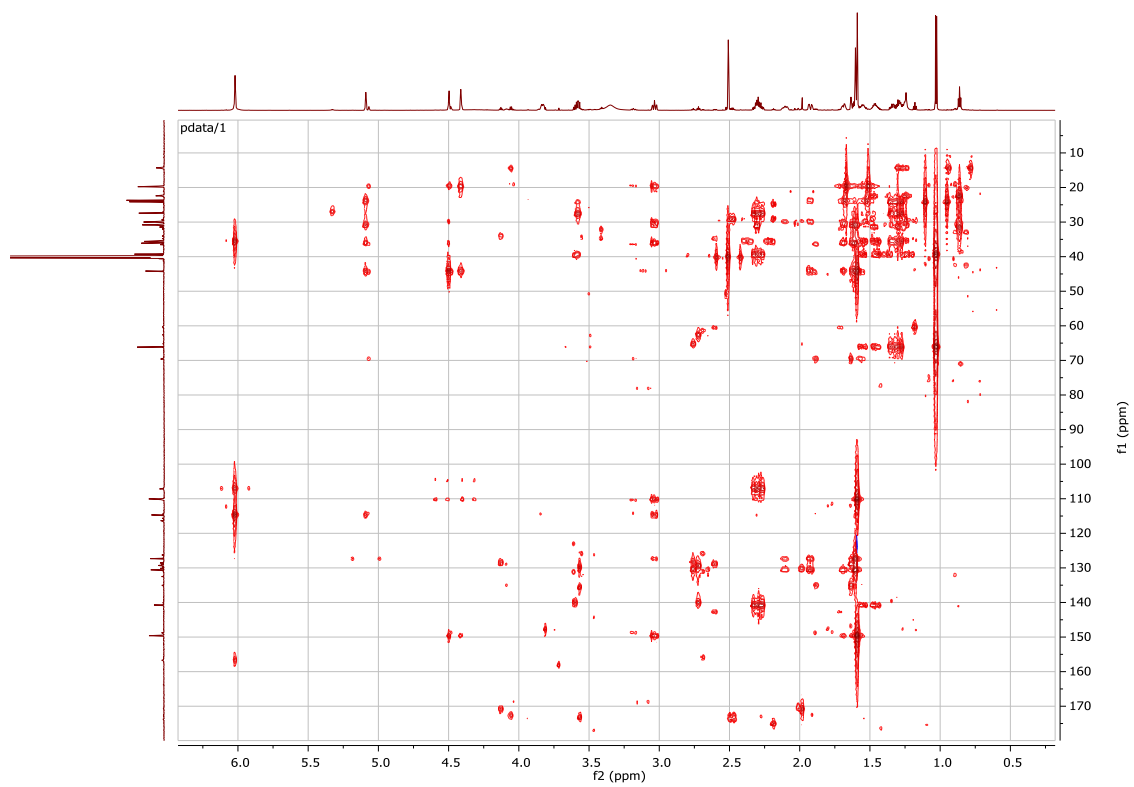


Figure S36. HMBC (long distance correlation ^1H - ^{13}C) spectrum of metabolite 4

5. Metabolite 5: 4,4''-dihydroxycannabidiol

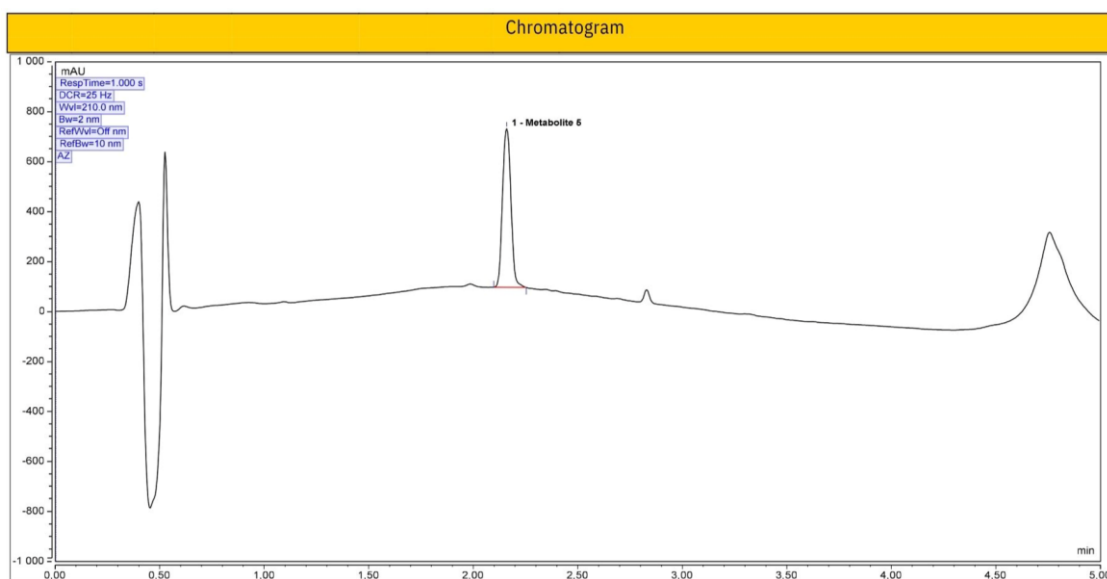


Figure S37. UHPLC-DAD chromatogram of metabolite **5**

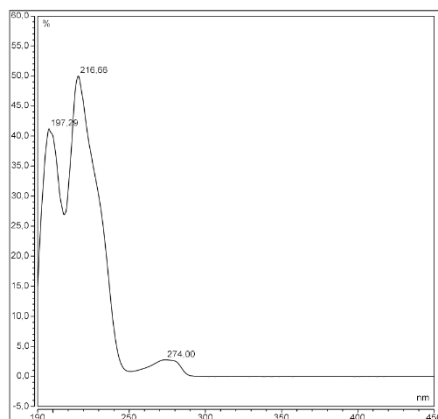


Figure S38. UV spectrum of metabolite **5**

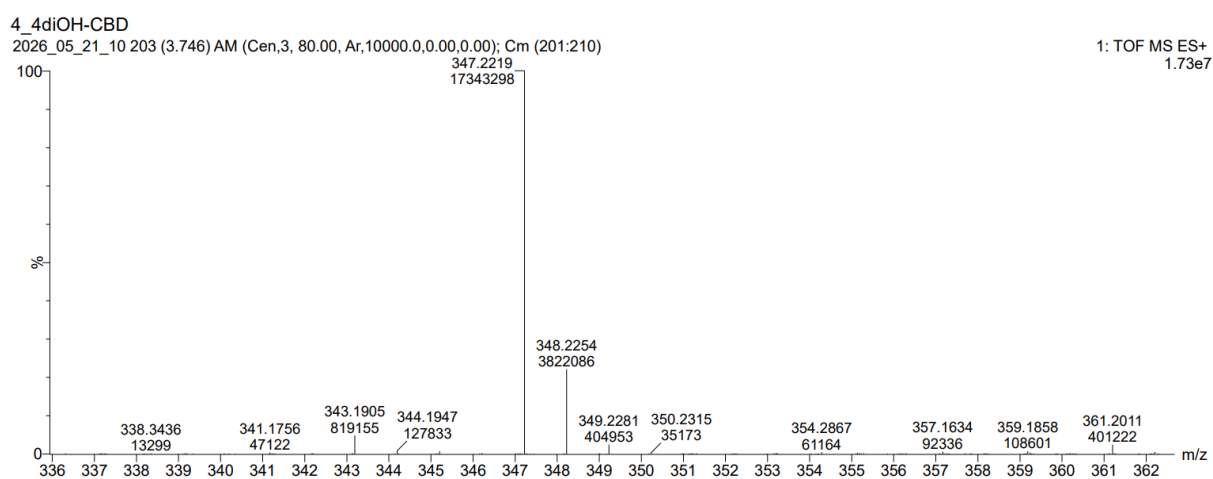


Figure S39. Mass spectrum of metabolite **5**

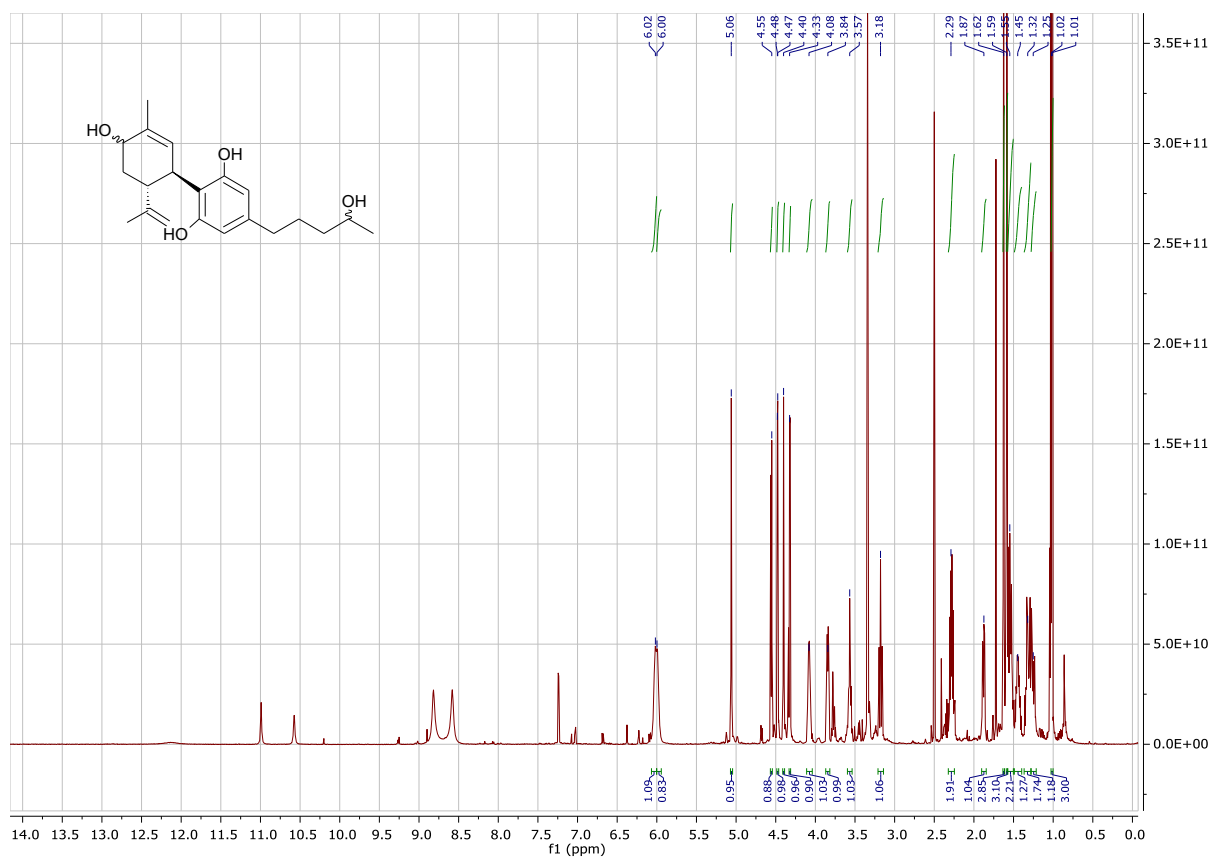


Figure S40. ^1H NMR spectrum of metabolite 5

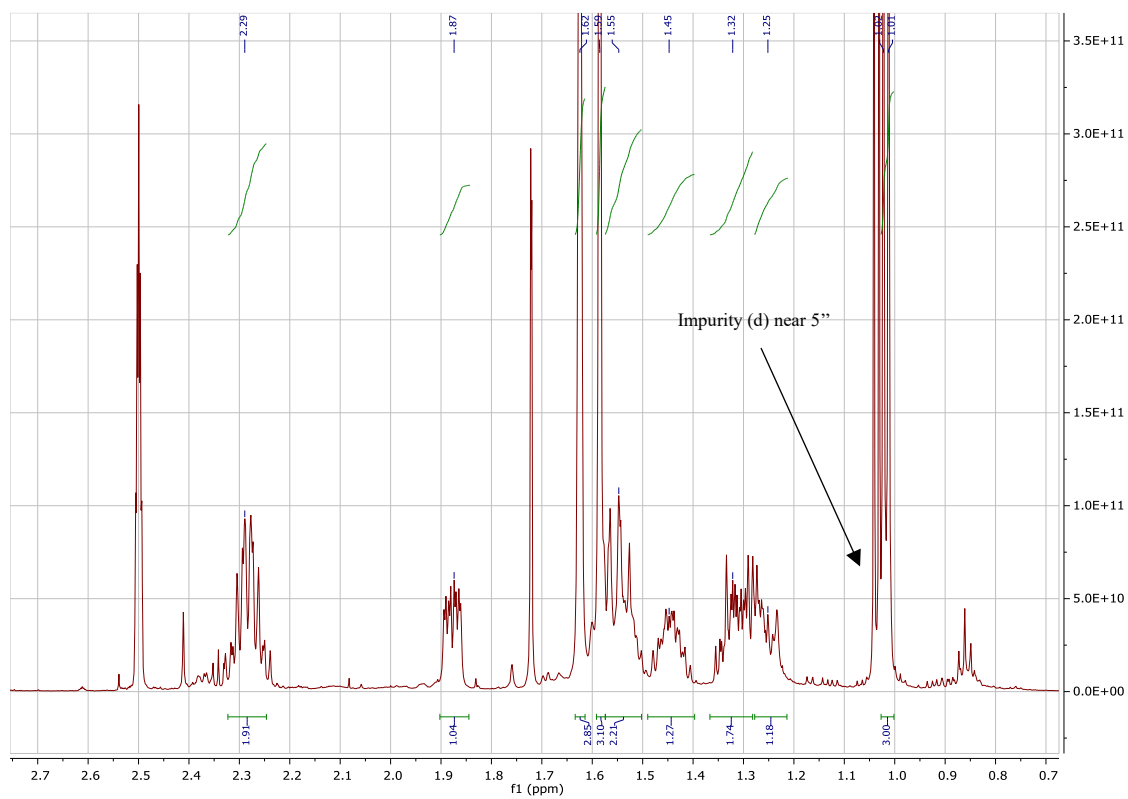


Figure S41. ^1H NMR (0.7-2.5) spectrum of metabolite 5

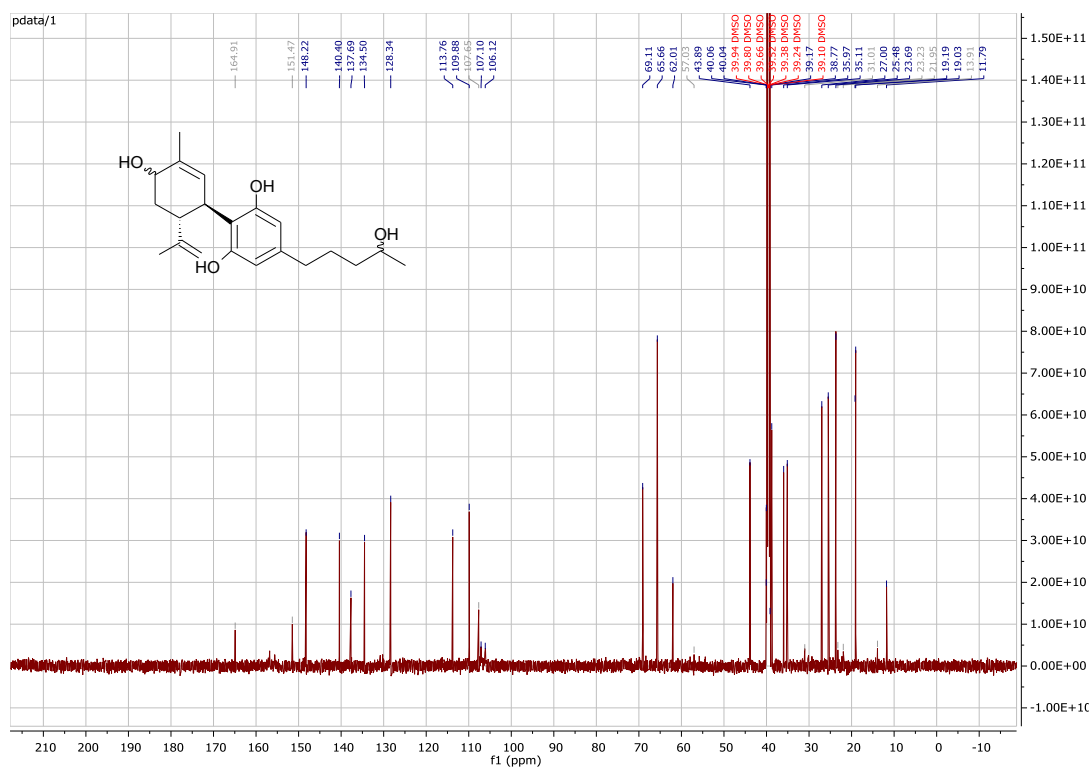


Figure S42. ^{13}C NMR spectrum of metabolite 5

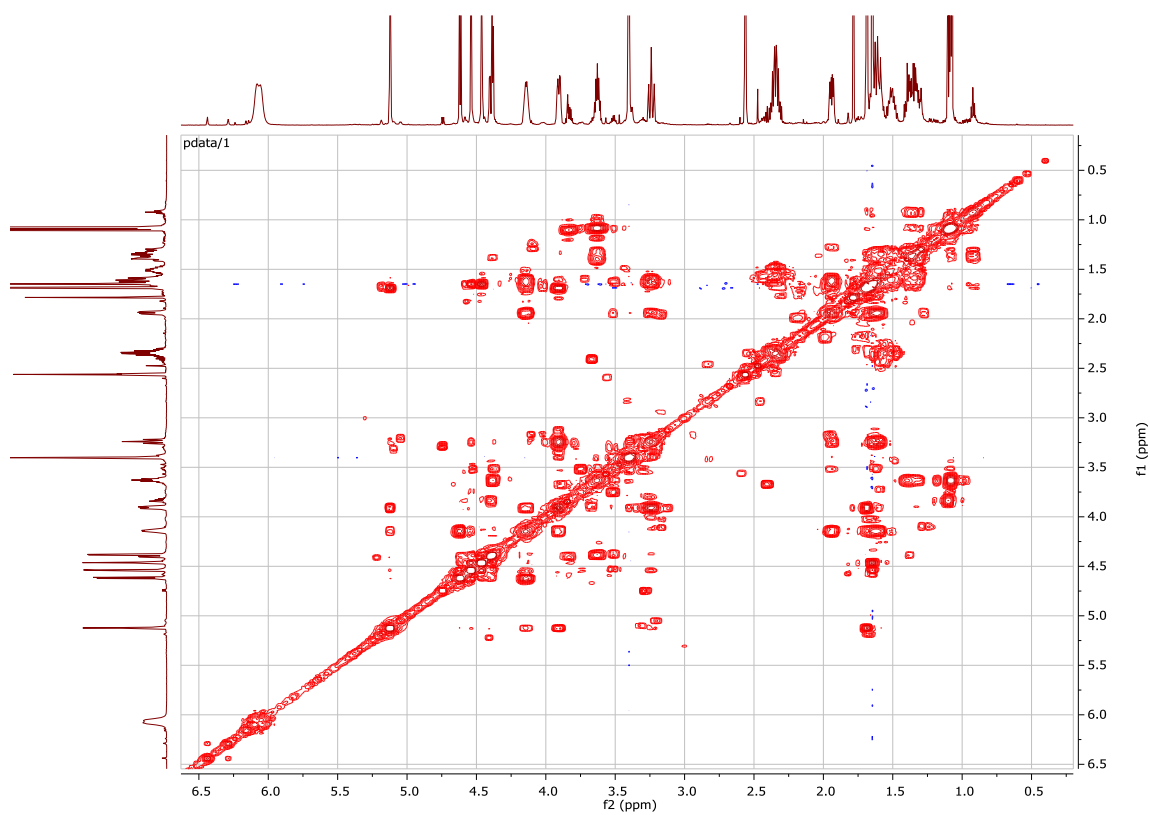


Figure S43. COSY (^1H - ^1H) NMR spectrum of metabolite 5

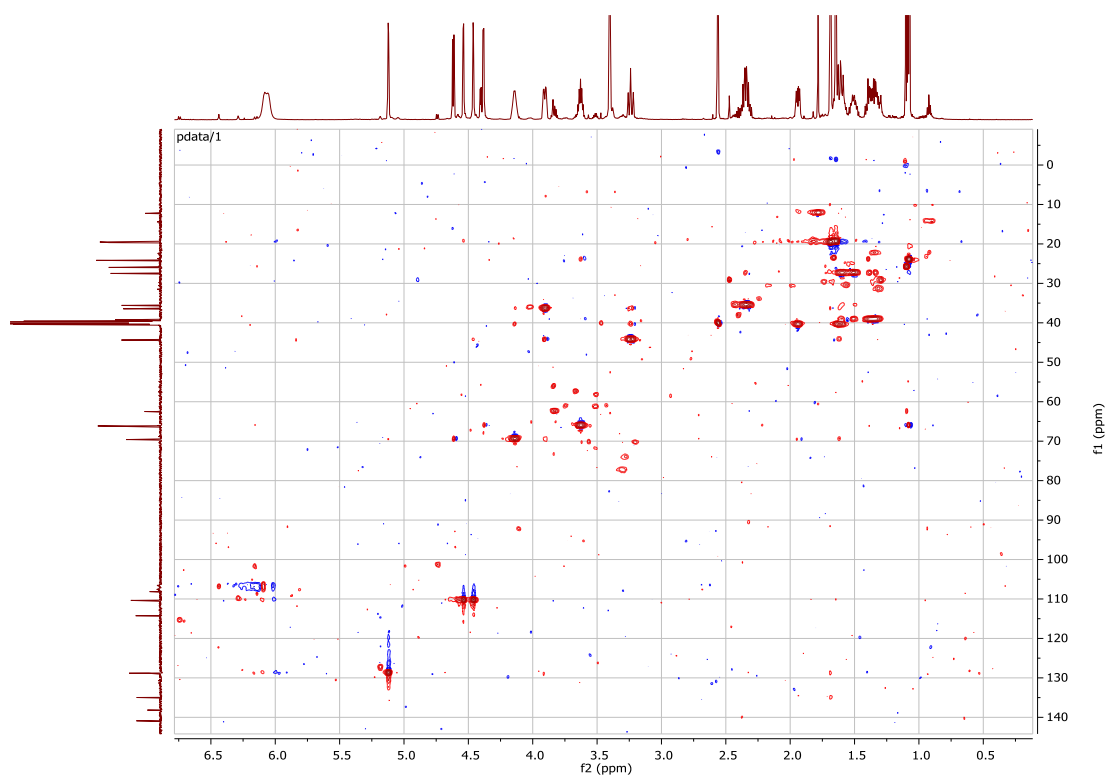


Figure S44. HSQC (^1H - ^{13}C) NMR spectrum of metabolite **5**

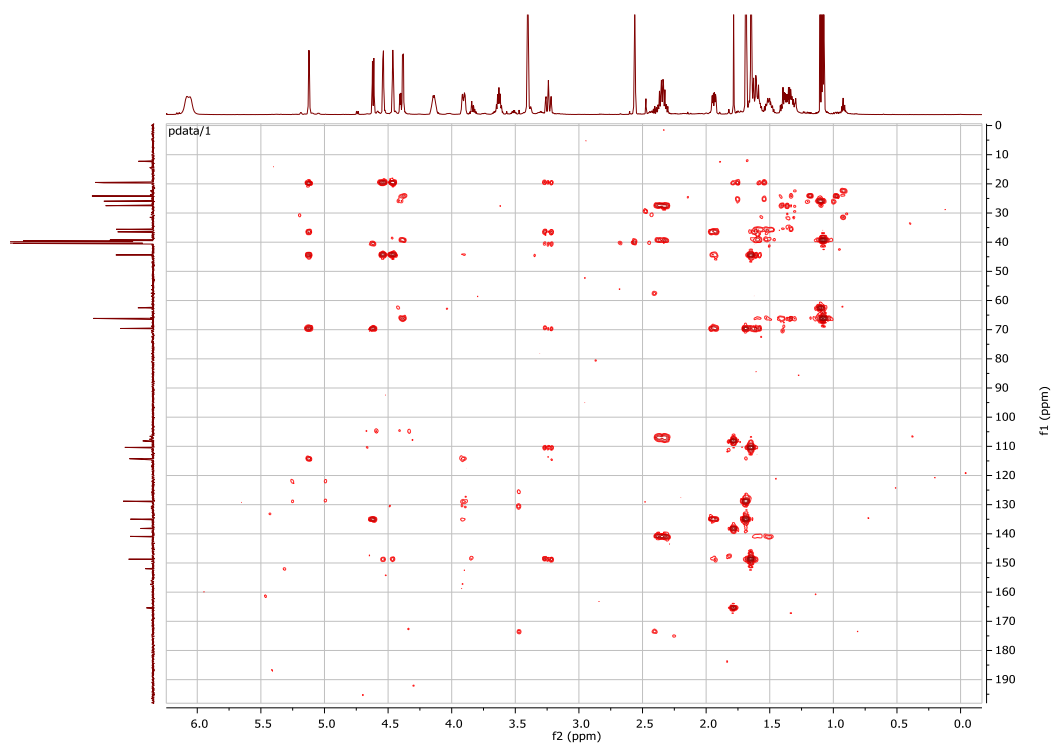


Figure S45. HMBC (long distance correlation ^1H - ^{13}C) spectrum of metabolite **5**

6. Metabolite 6: 7,4''-dihydroxycannabidiol

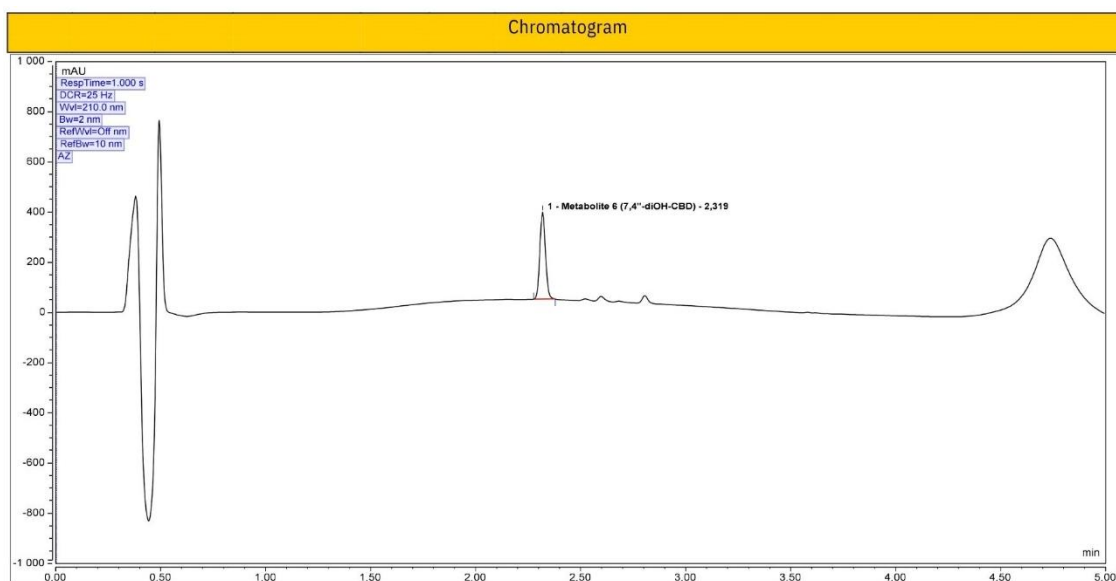


Figure S46. UHPLC-DAD chromatogram of metabolite 6

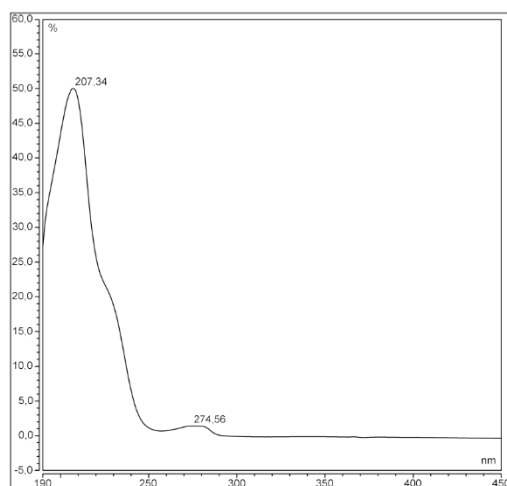


Figure S47. UV spectrum of metabolite 6

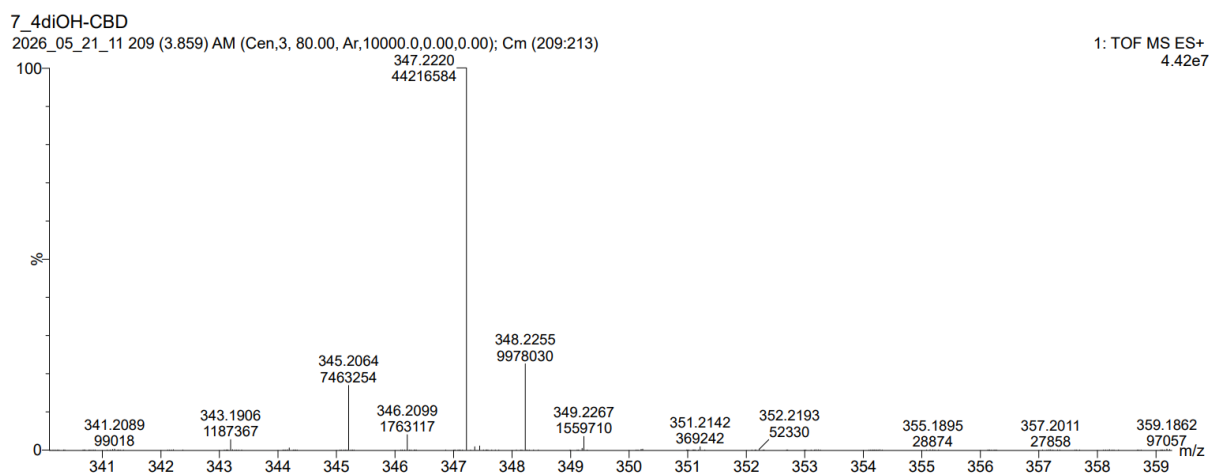


Figure S48. Mass spectrum of metabolite 6

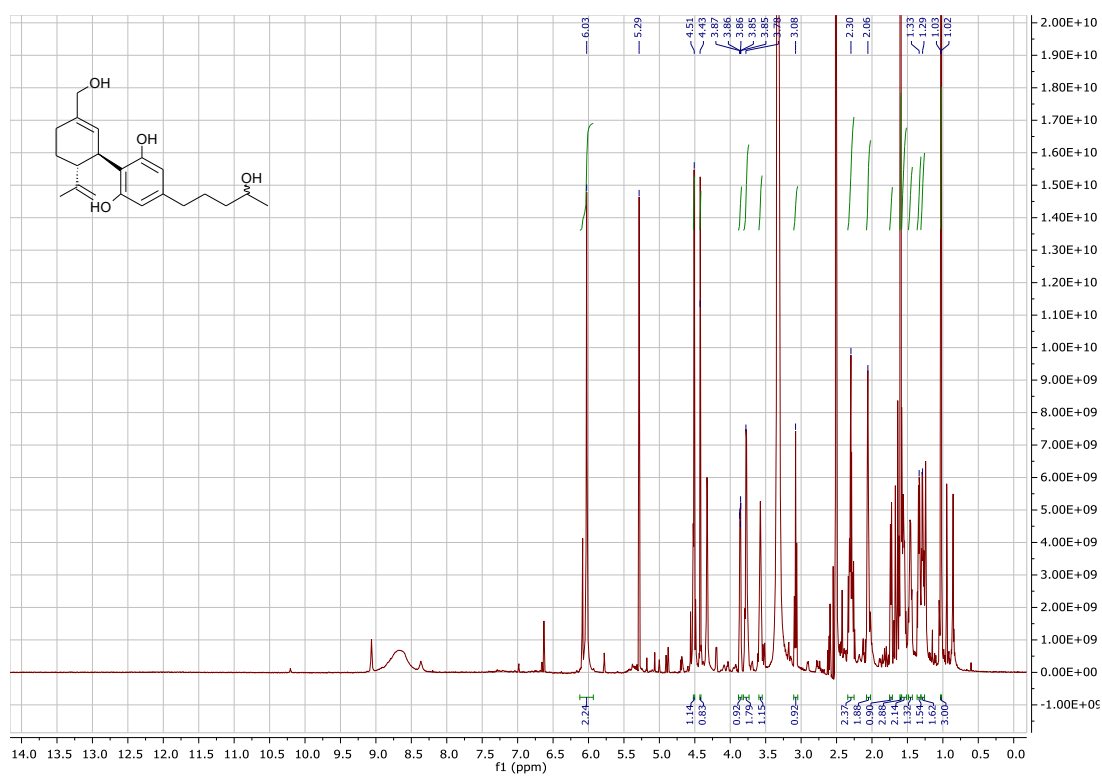


Figure S49. ¹H NMR spectrum of metabolite 6

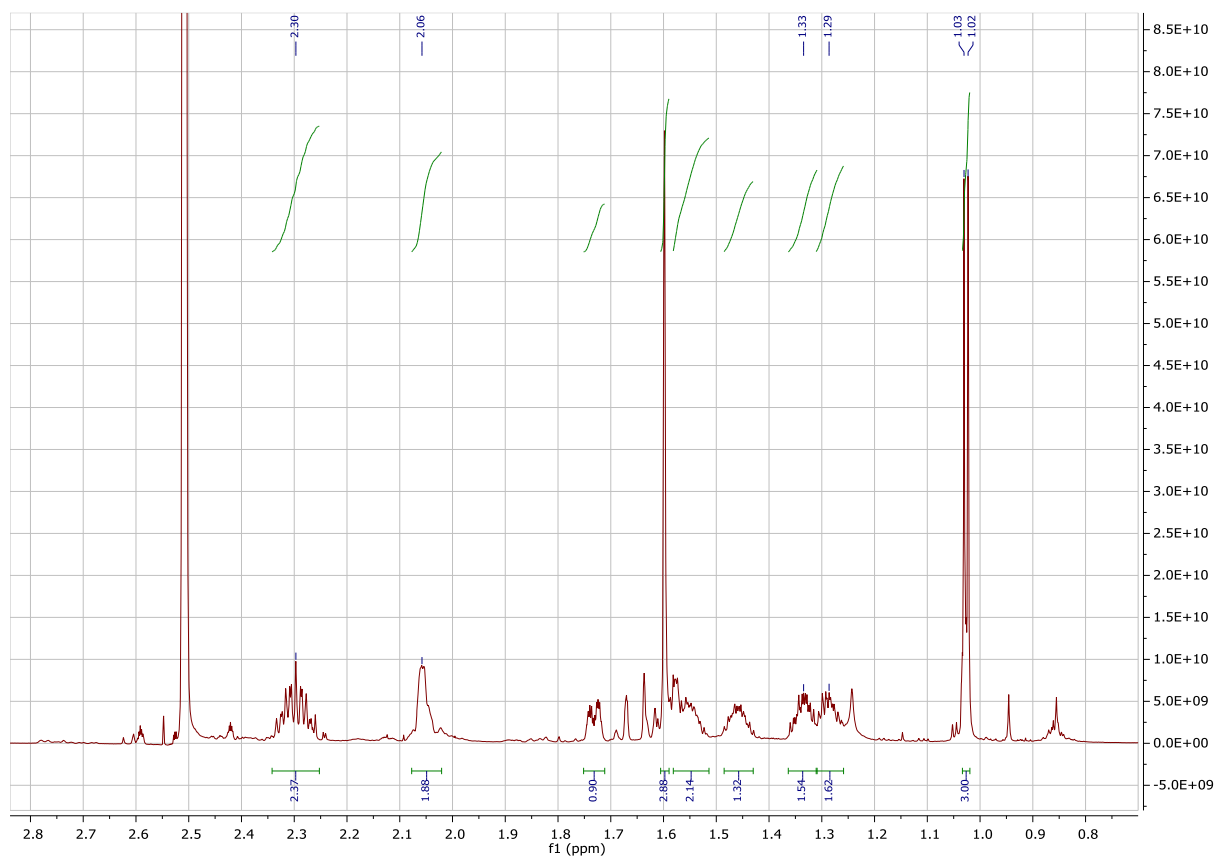


Figure S50. ¹H NMR (0.8-2.8) spectrum of metabolite 6

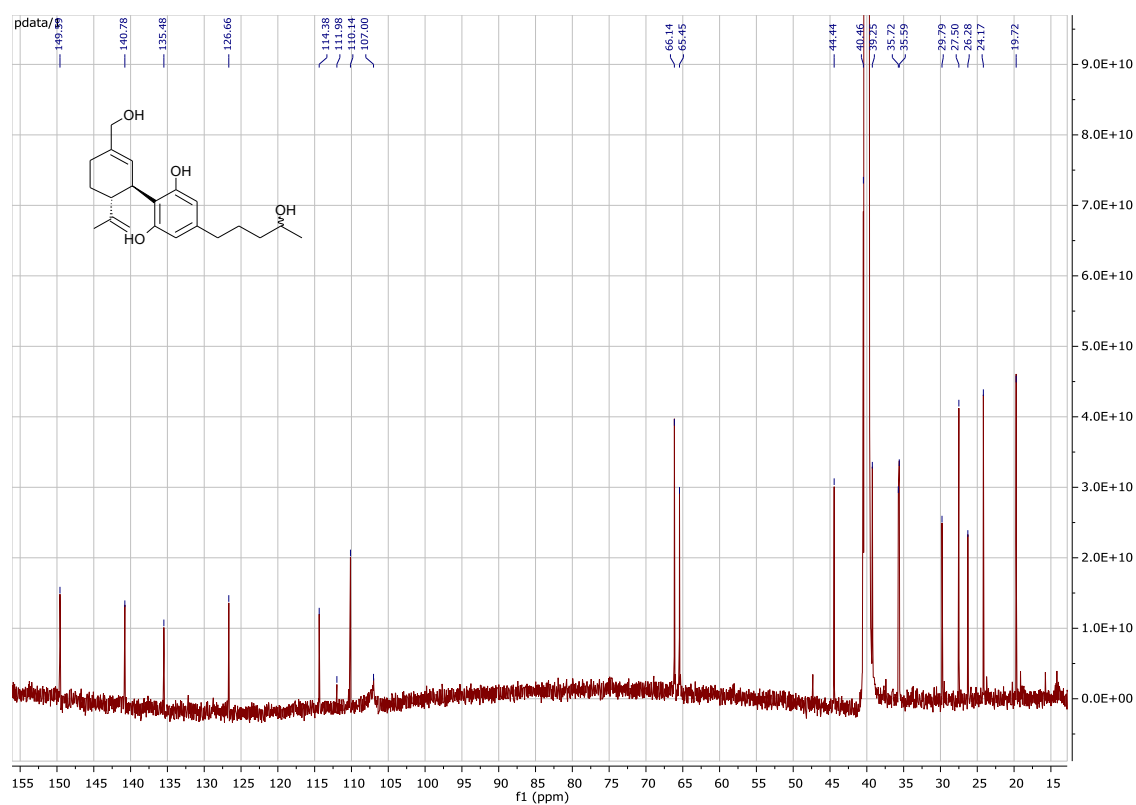


Figure S51. ¹³C NMR spectrum of metabolite 6

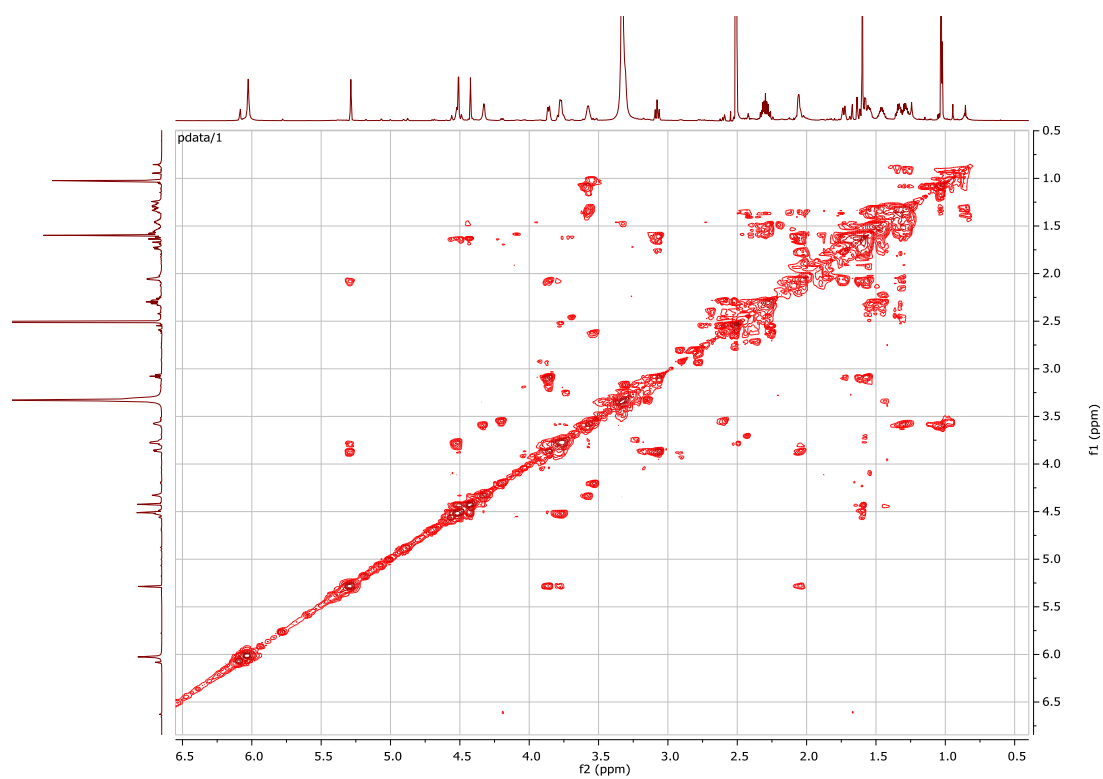


Figure S52. COSY (¹H-¹H) NMR spectrum of metabolite 6

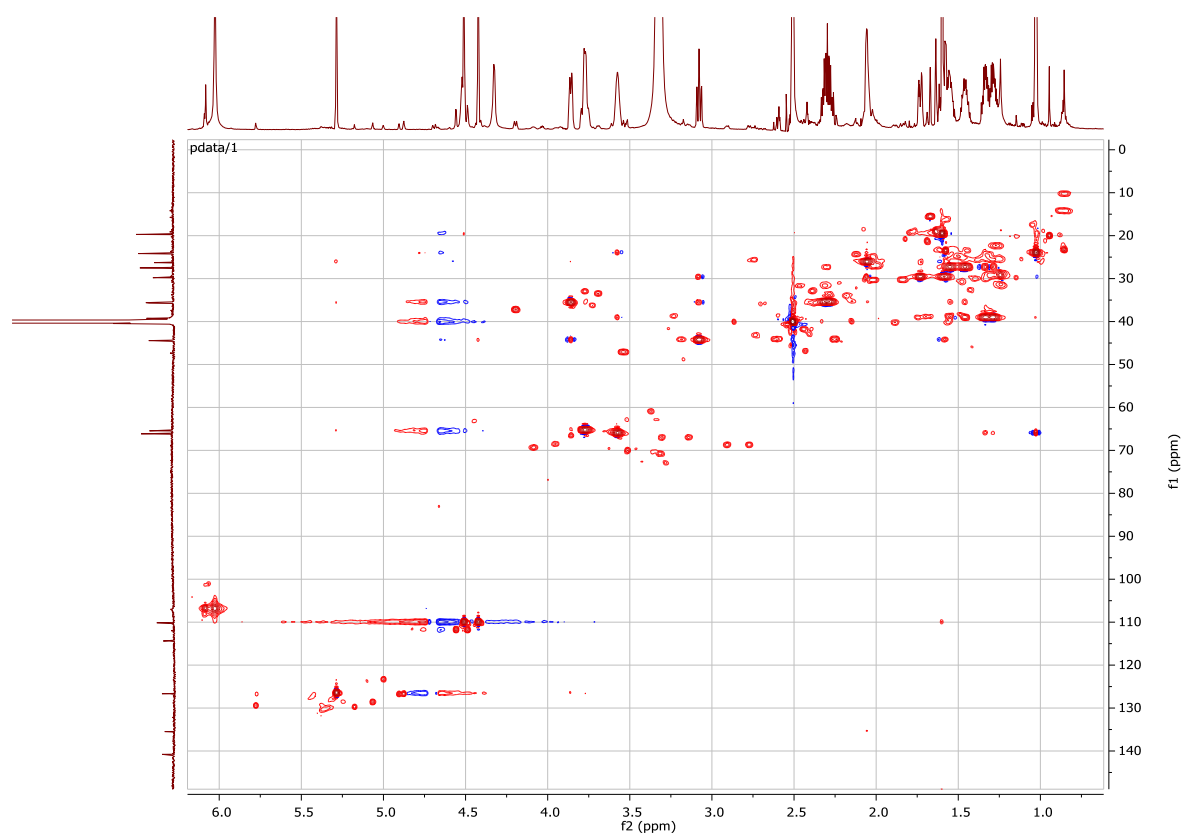


Figure S53. HSQC (^1H - ^{13}C) NMR spectrum of metabolite **6**

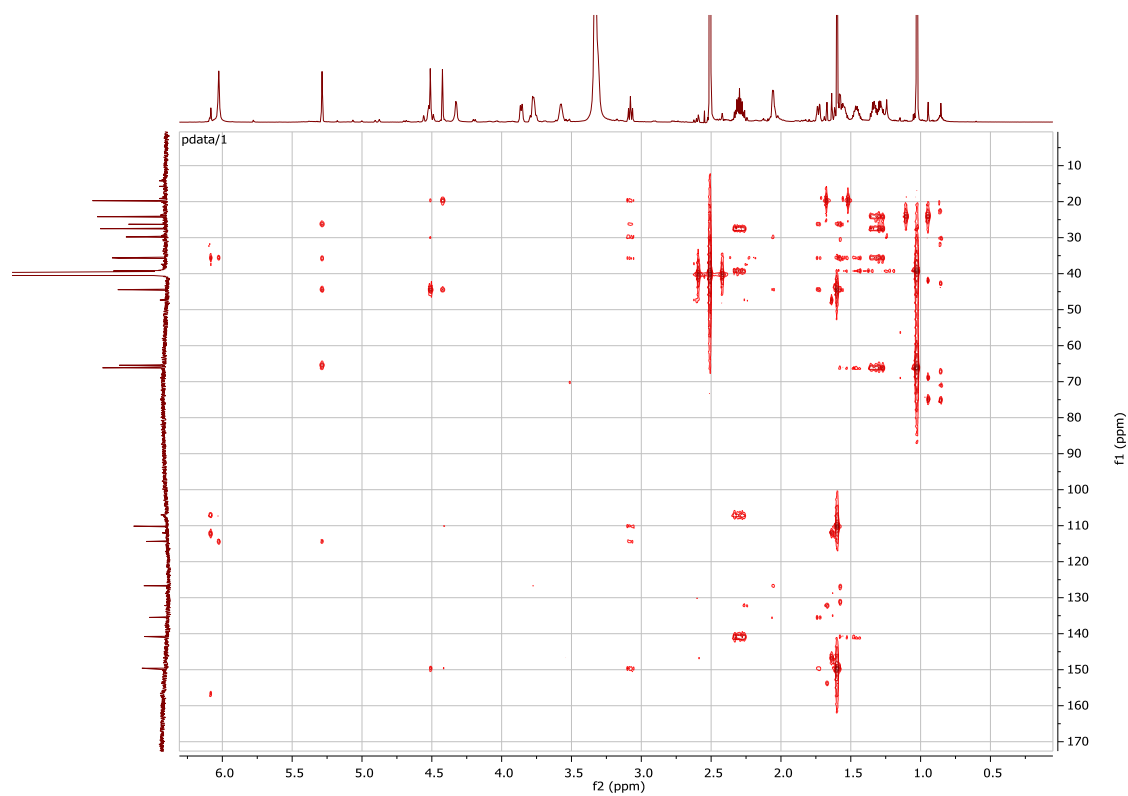


Figure S54. HMBC (long distance correlation ^1H - ^{13}C) spectrum of metabolite **6**

7. Metabolite 7: 2'-O-(4'''-O-methyl- β -D-glucopyranosyl)cannabidiol

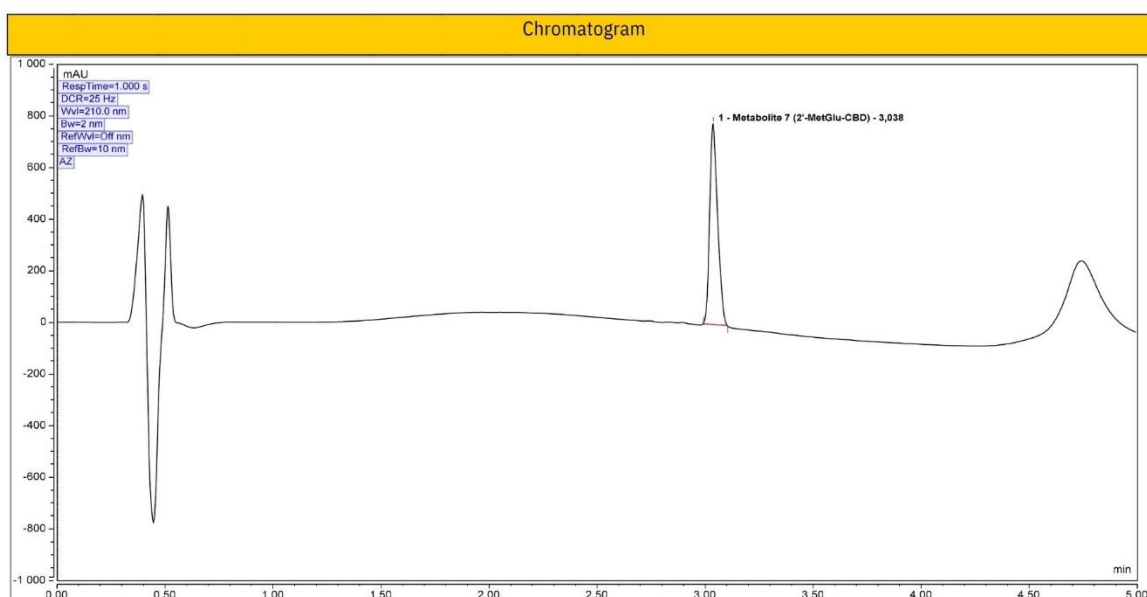


Figure S55. UHPLC-DAD chromatogram of metabolite 7

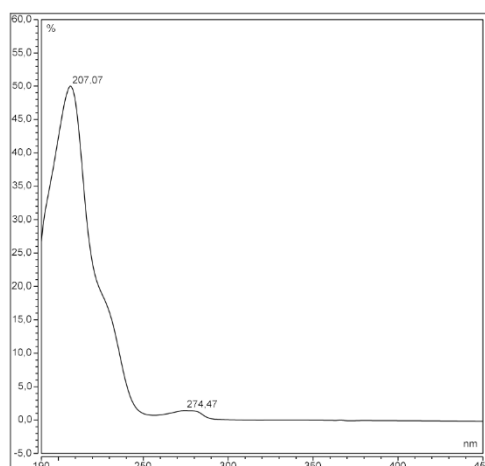


Figure S56. UV spectrum of metabolite 7

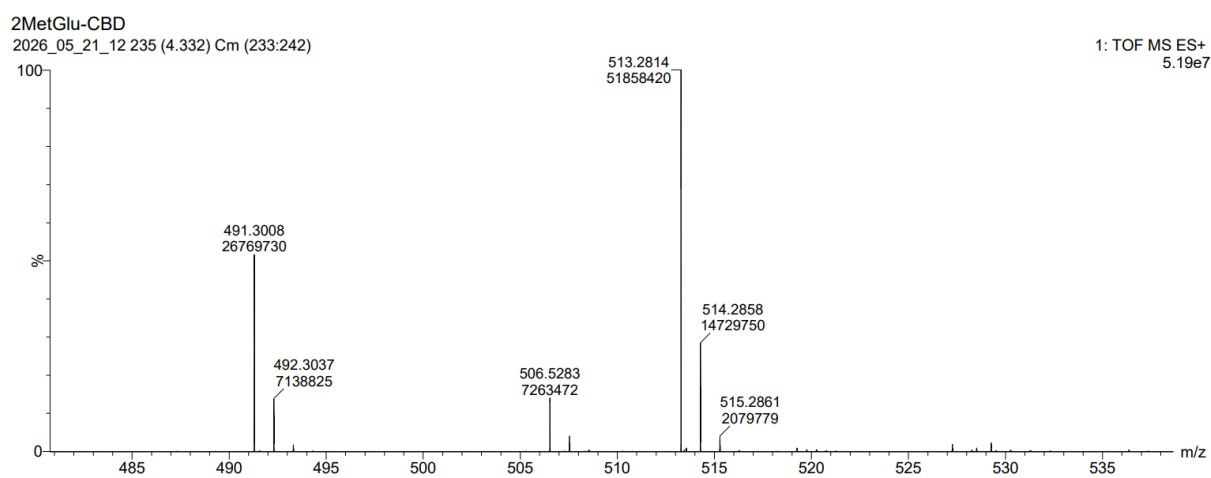


Figure S57. Mass spectrum of metabolite 7

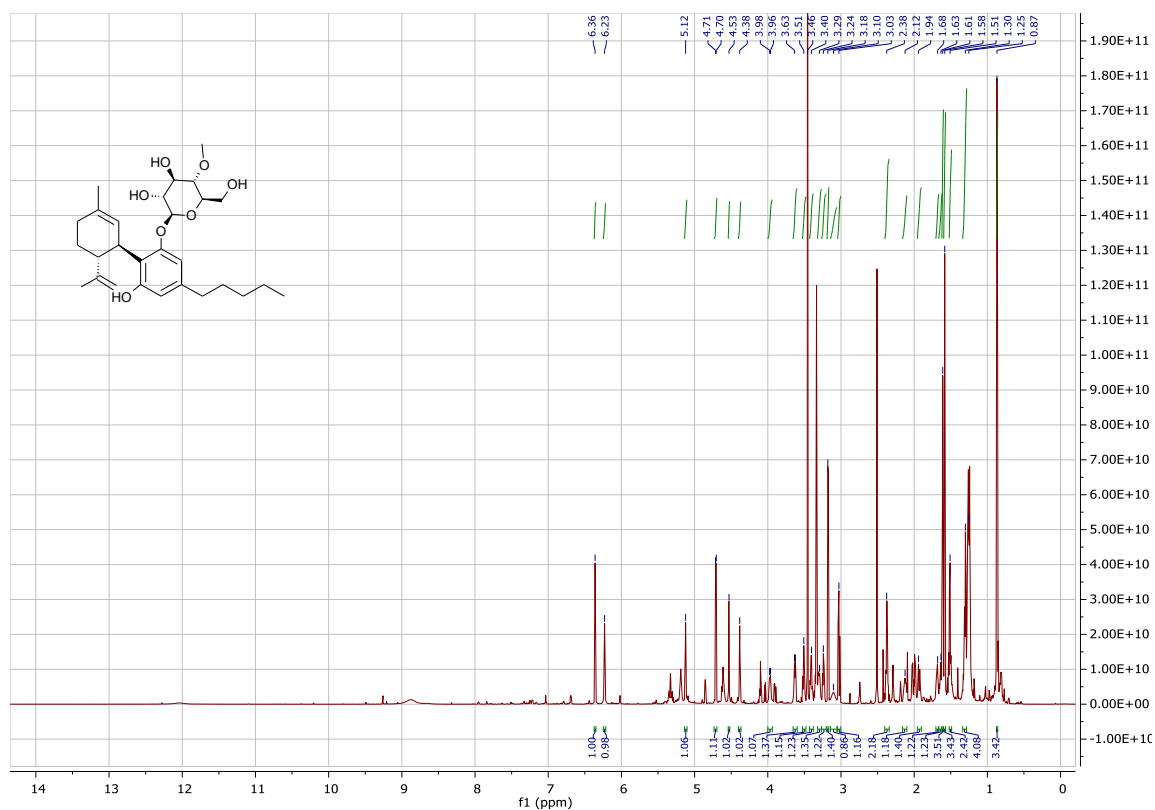


Figure S58. ^1H NMR spectrum of metabolite 7

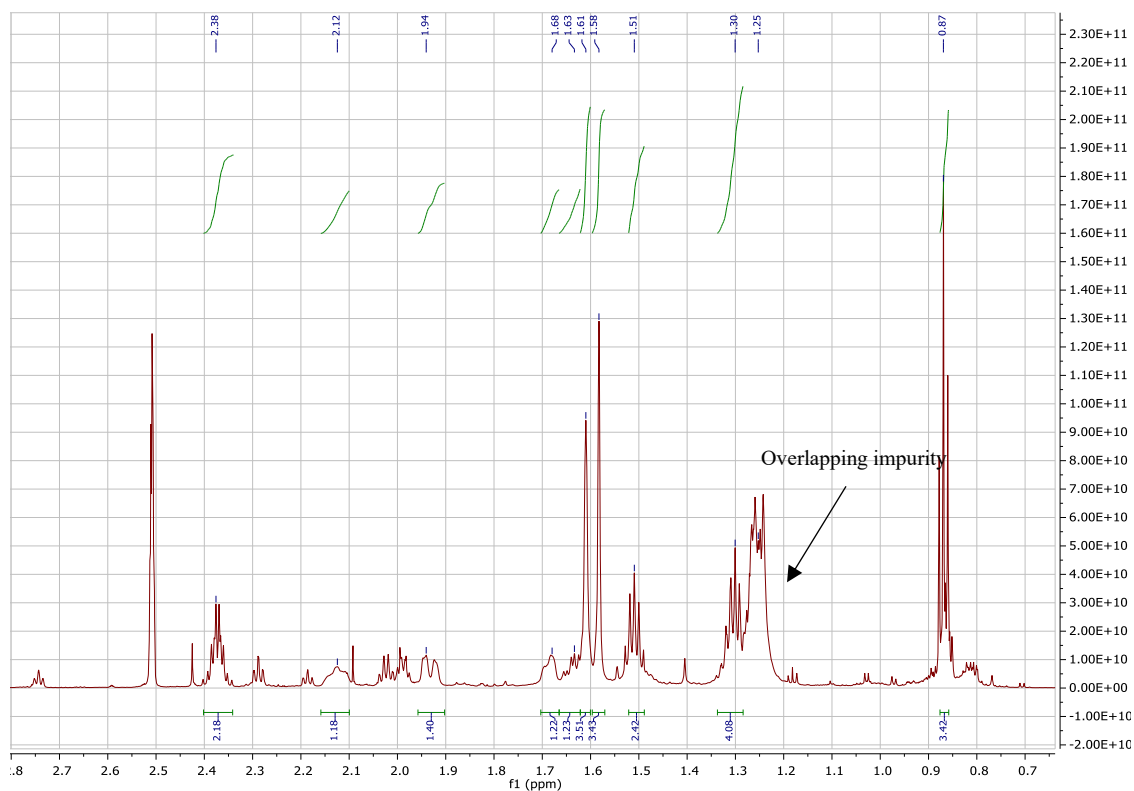


Figure S59. ^1H NMR (0.7-2.6) spectrum of metabolite 7

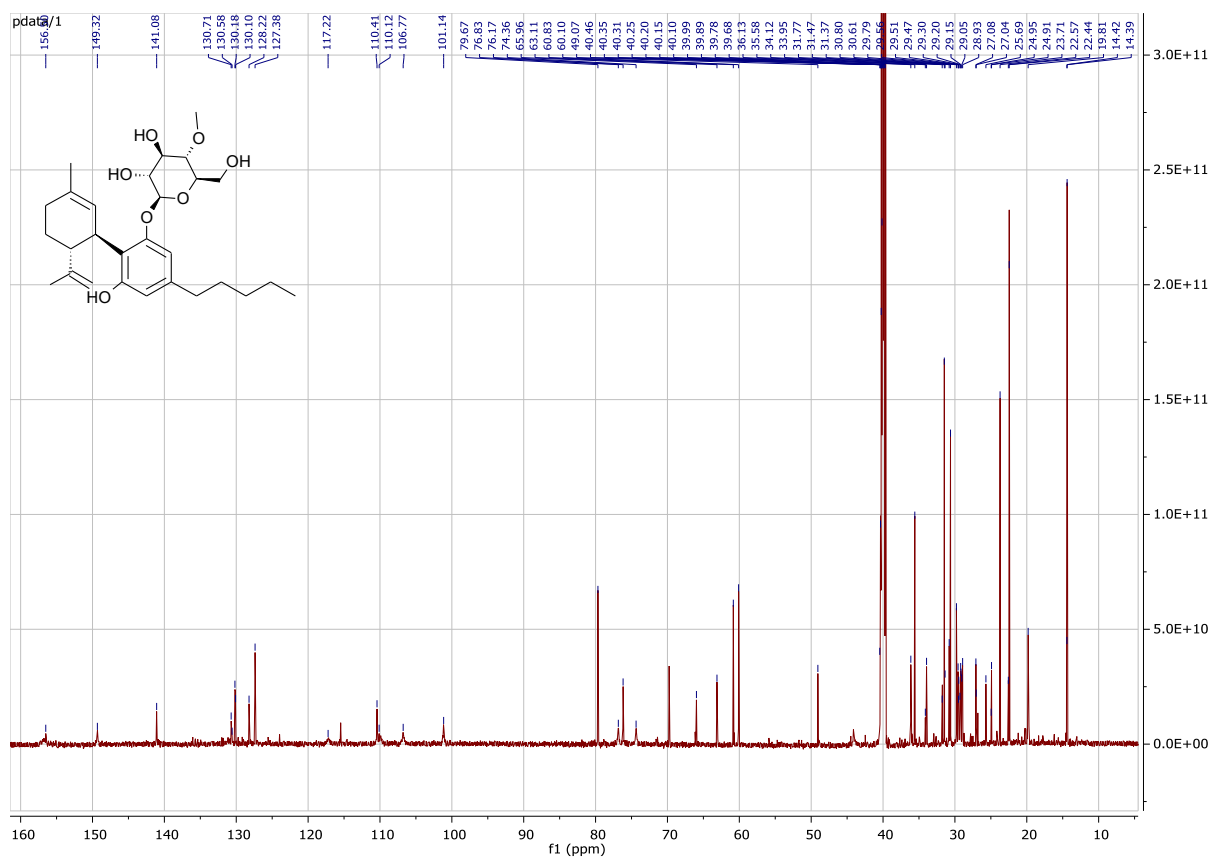


Figure S60. ^{13}C NMR spectrum of metabolite 7

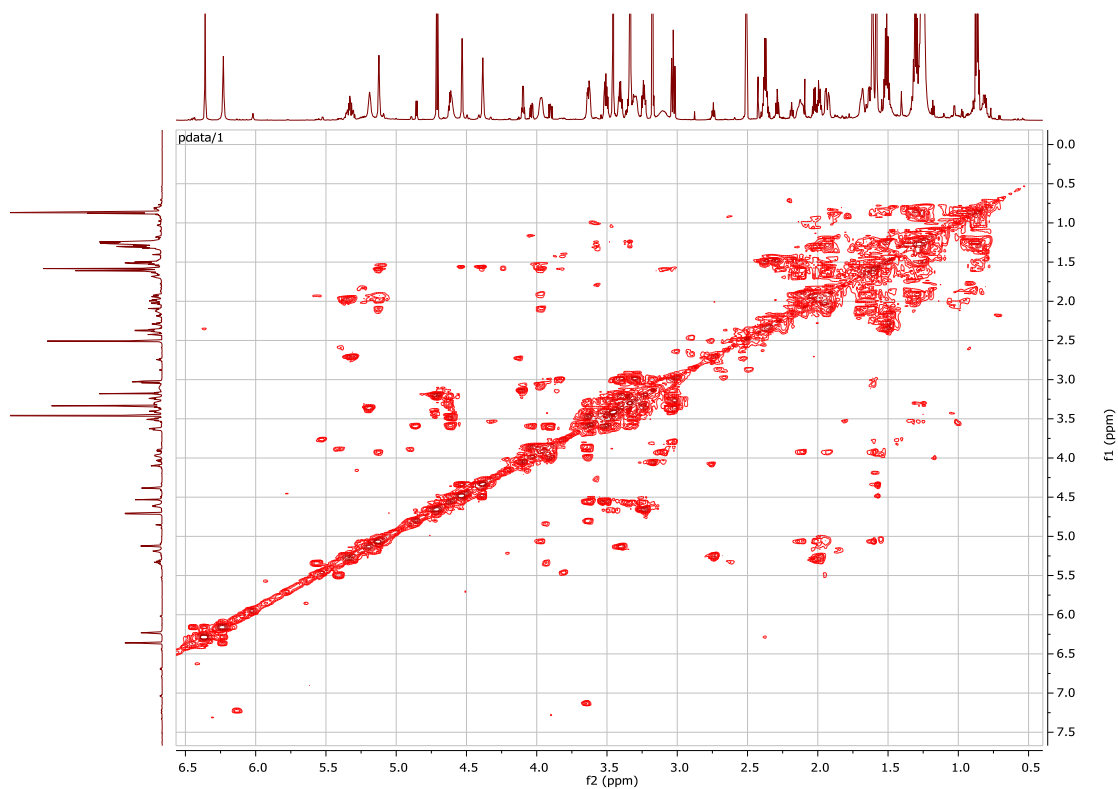


Figure S61. COSY (^1H - ^1H) NMR spectrum of metabolite 7

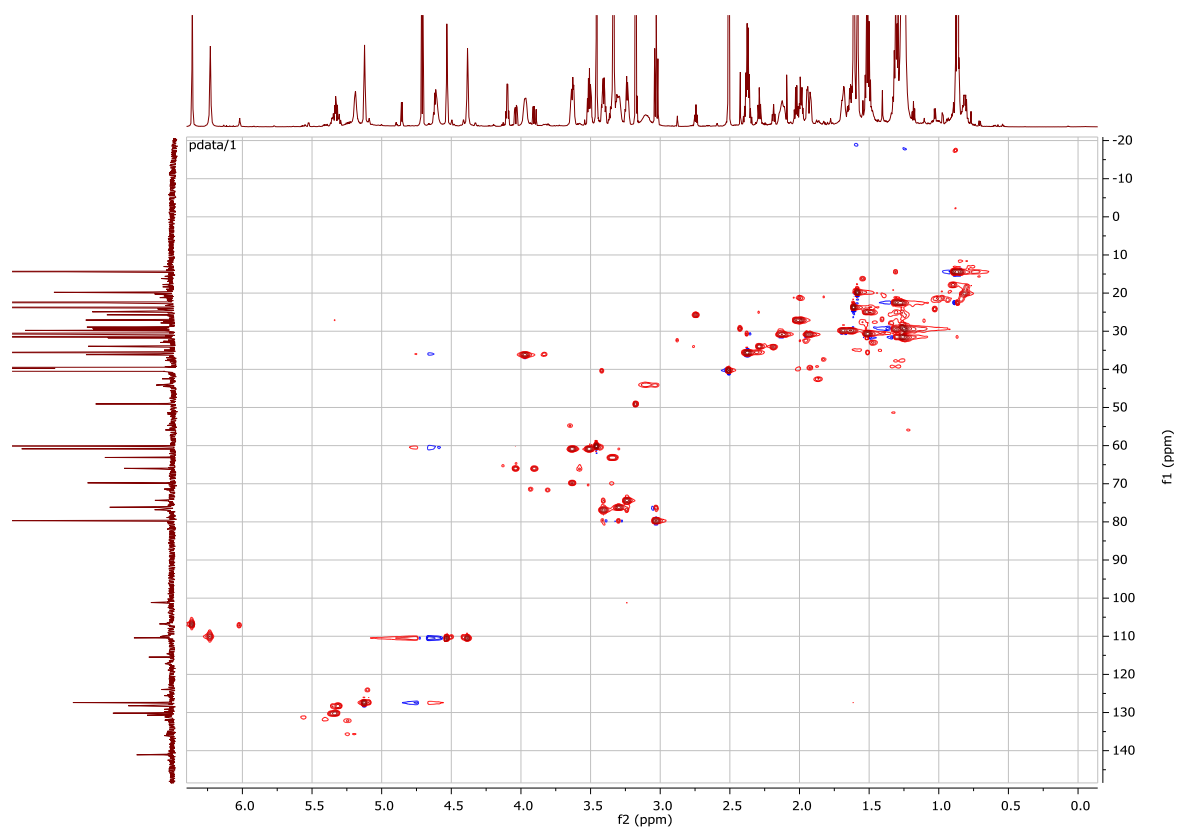


Figure S62. HSQC (^1H - ^{13}C) NMR spectrum of metabolite 7

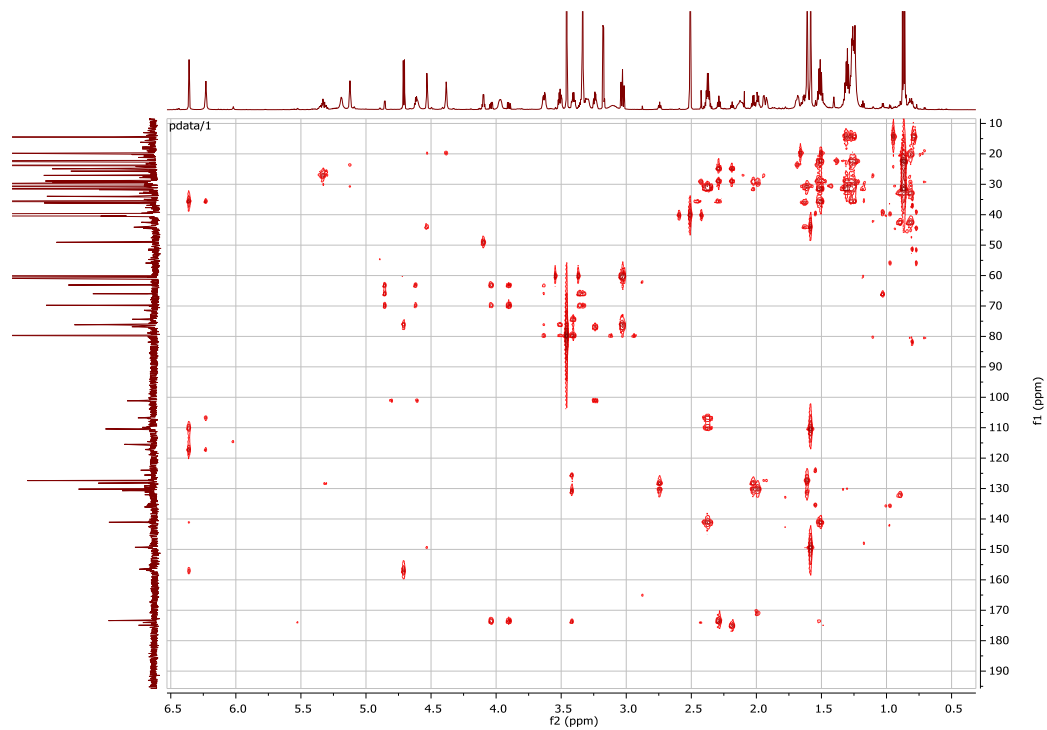


Figure S63. HMBC (long distance correlation ^1H - ^{13}C) NMR spectrum of metabolite 7

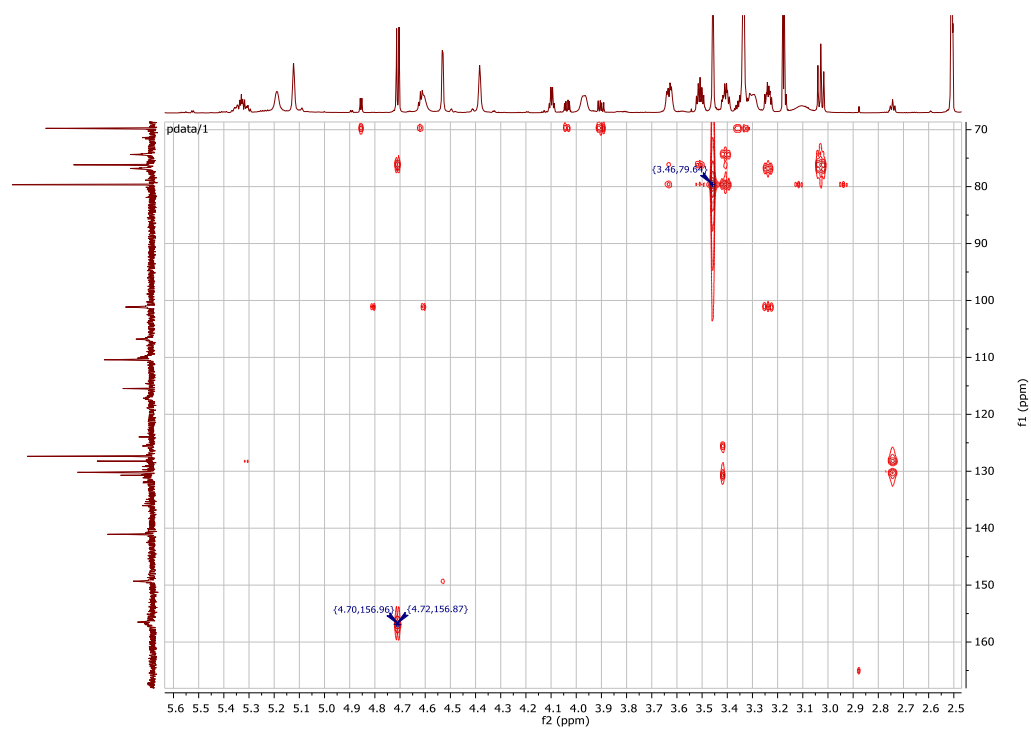


Figure S64. HMBC correlation between C1 carbon of glucose and C2 carbon of aromatic ring. Correlation between -OCH₃ methoxyl group and C4 carbon of glucose.

8. Metabolite 8: 2'-O-(4'''-O-methyl- β -D-glucopyranosyl) 5''-hydroxycannabidiol

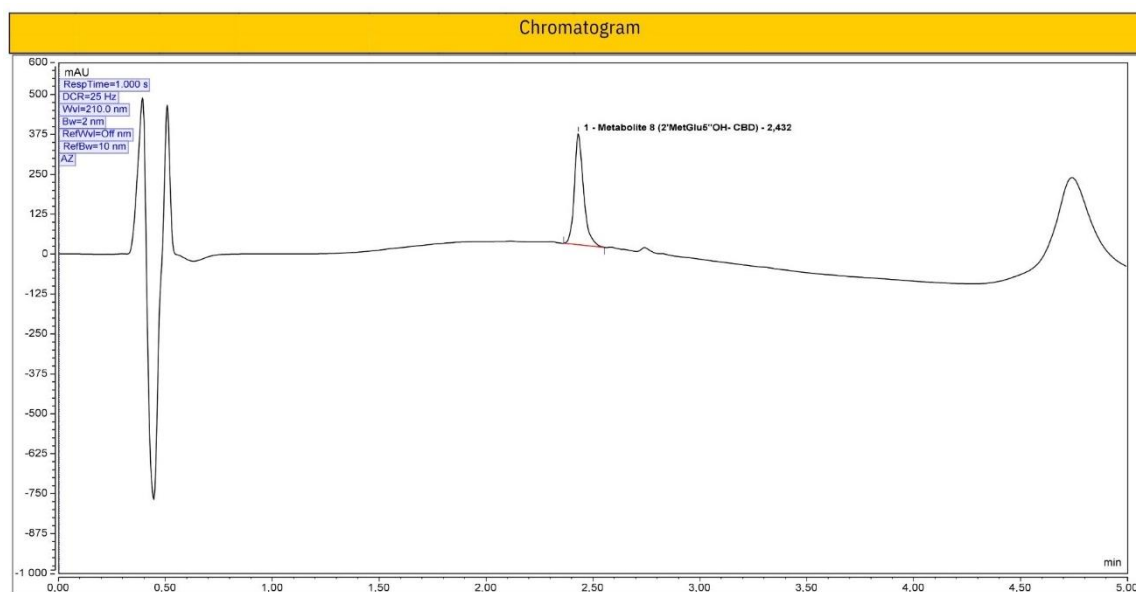


Figure S65. UHPLC-DAD chromatogram of metabolite 8

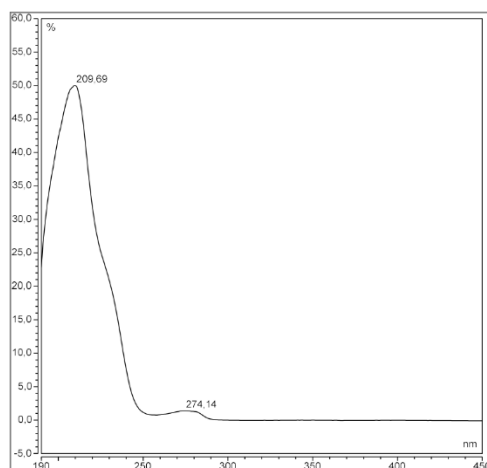


Figure S66. UV spectrum of metabolite 8

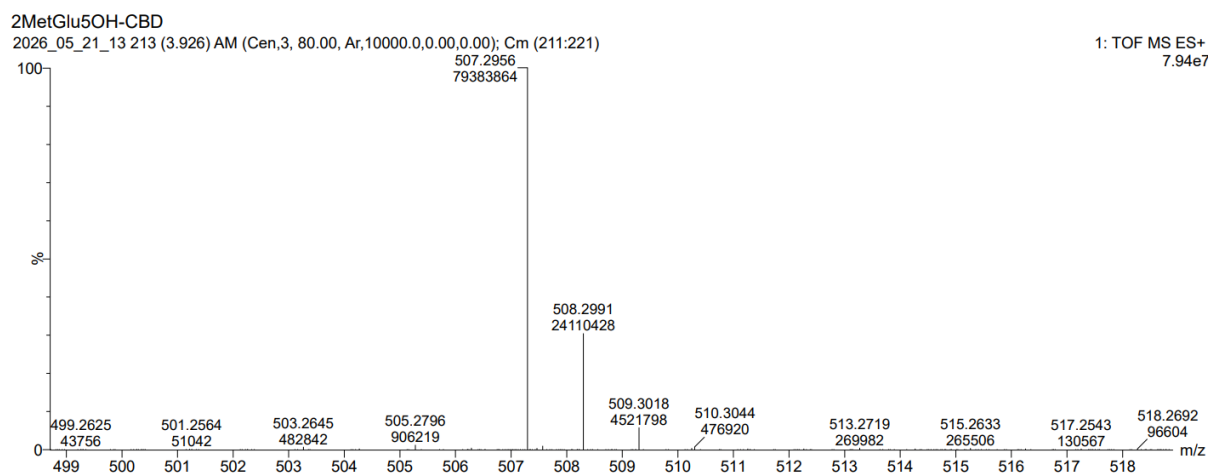


Figure S67. Mass spectrum of metabolite 8

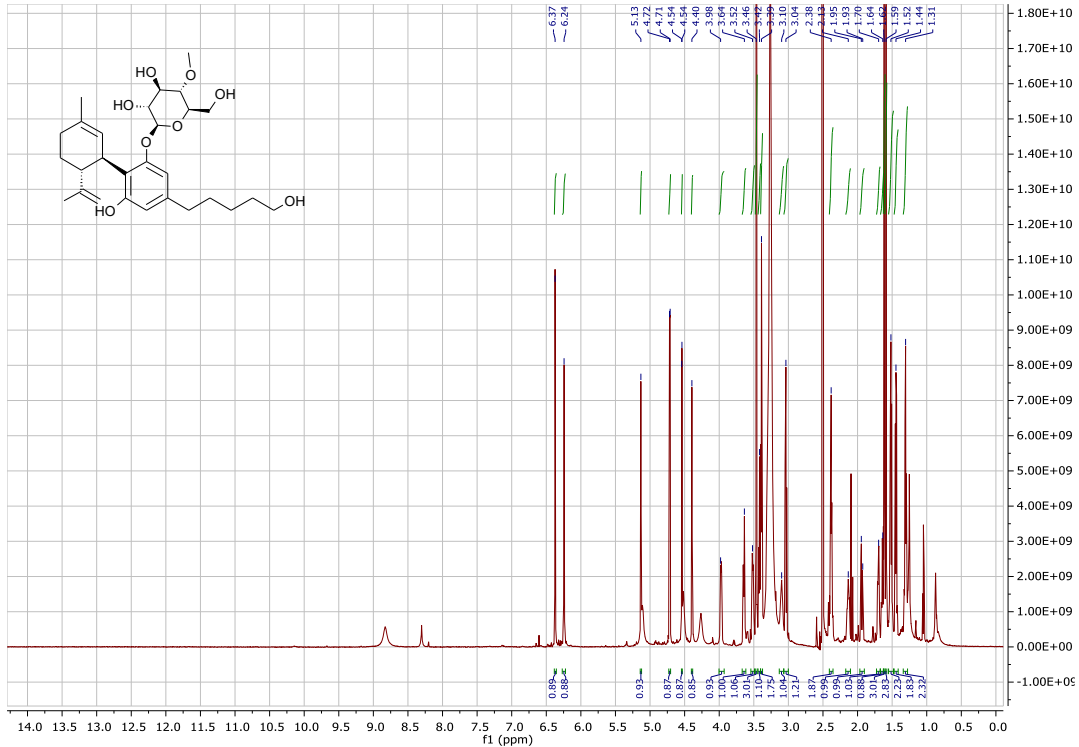


Figure S68. ^1H NMR spectrum of metabolite **8**

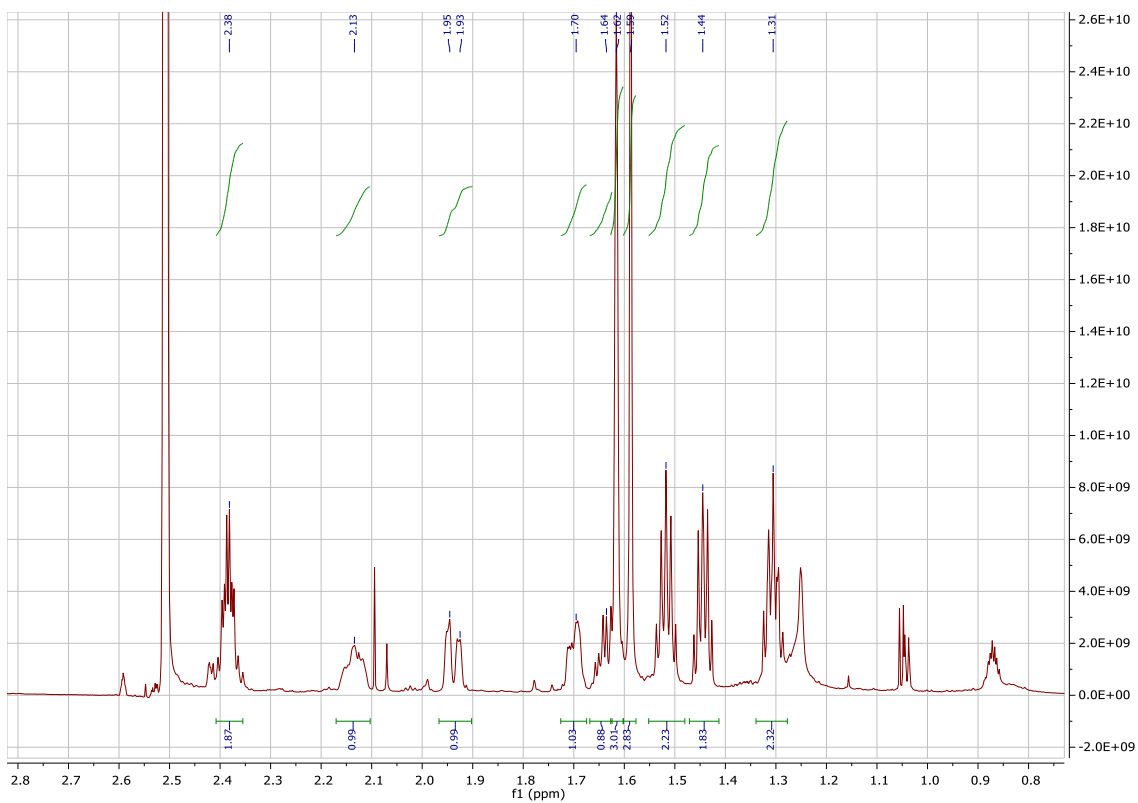


Figure S69. ^1H NMR (0.8-2.8) spectrum of metabolite **8**

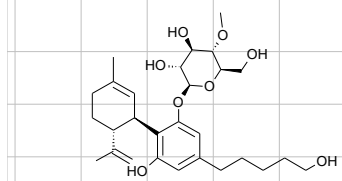


Figure S71. COSY (^1H - ^1H) NMR spectrum of metabolite **8**

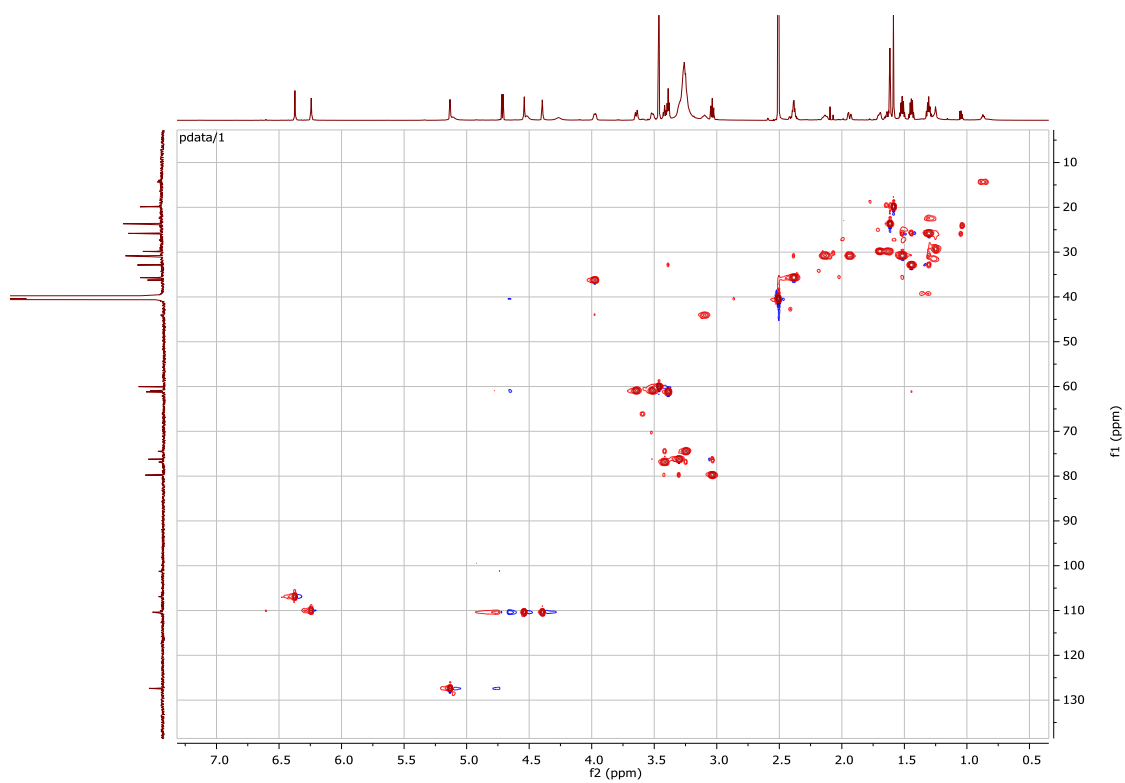


Figure S72. HSQC (^1H - ^{13}C) NMR spectrum of metabolite **8**

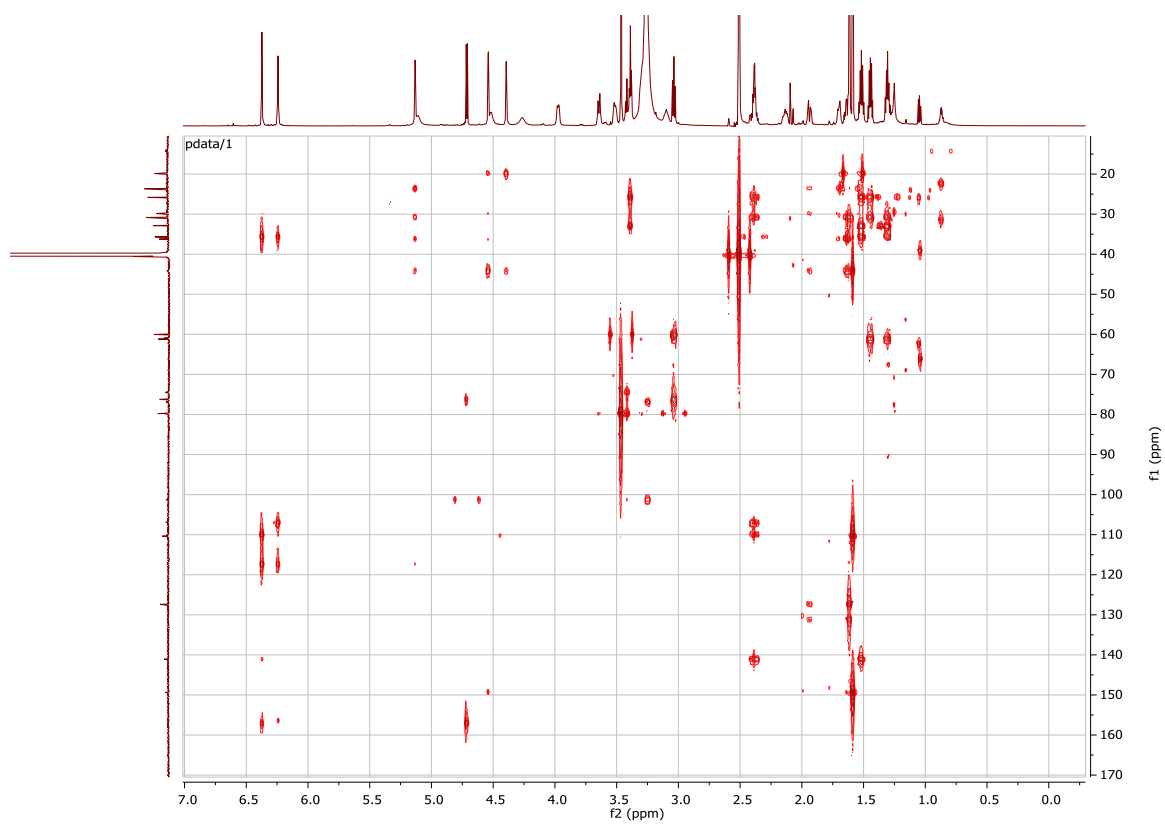


Figure S73. HMBC (long distance correlation ^1H - ^{13}C) NMR spectrum of metabolite **8**

9. Metabolite 9: 4,2''-dihydroxycannabidiol

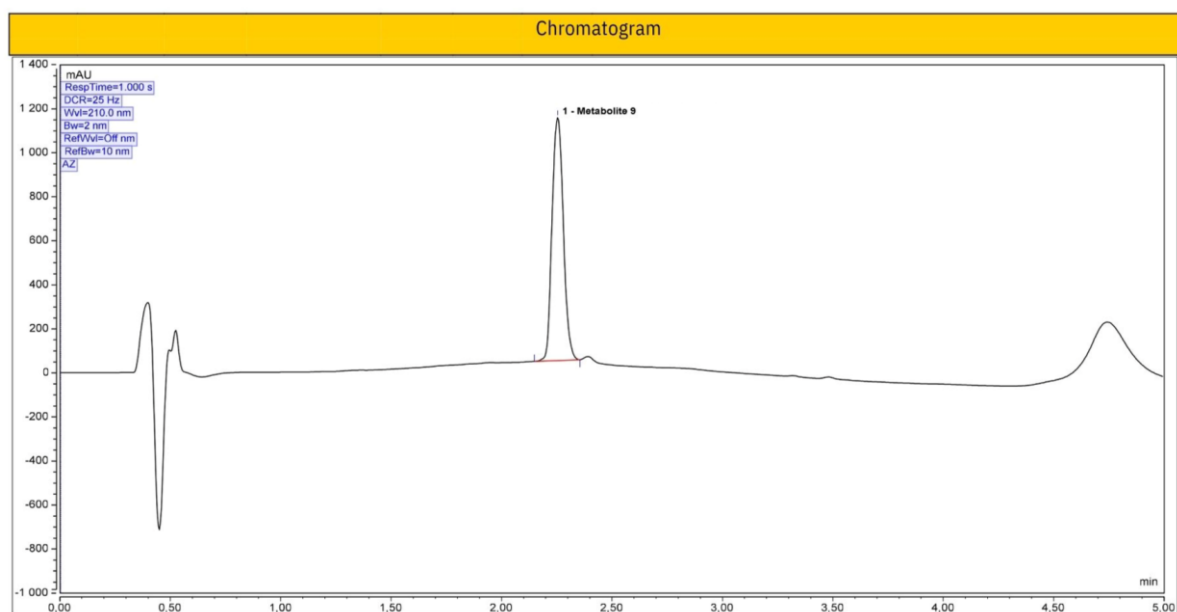


Figure S74. UHPLC-DAD chromatogram of metabolite 9

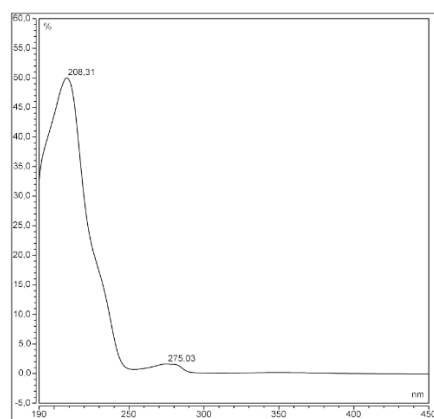


Figure S75. UV spectrum of metabolite 9

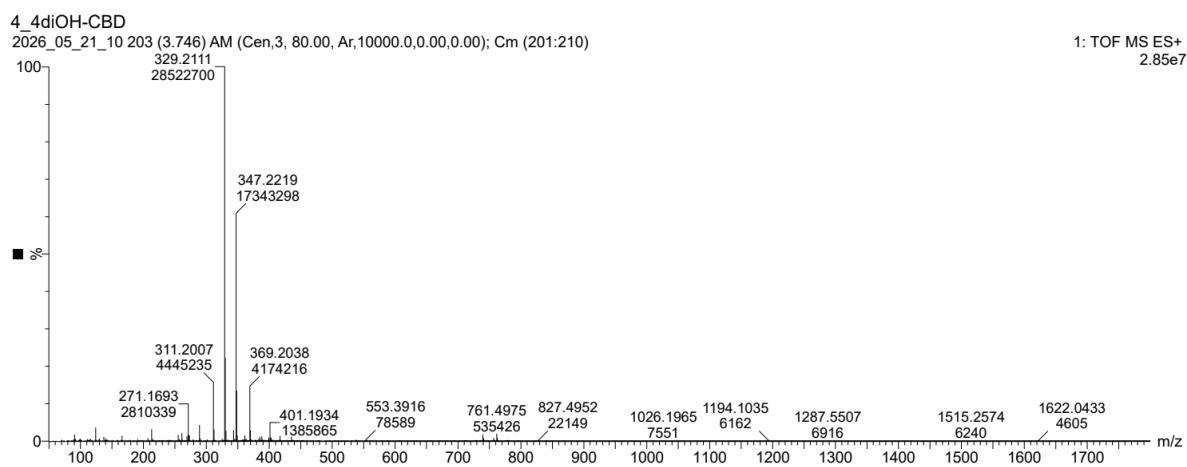


Figure S76. Mass spectrum of metabolite 9

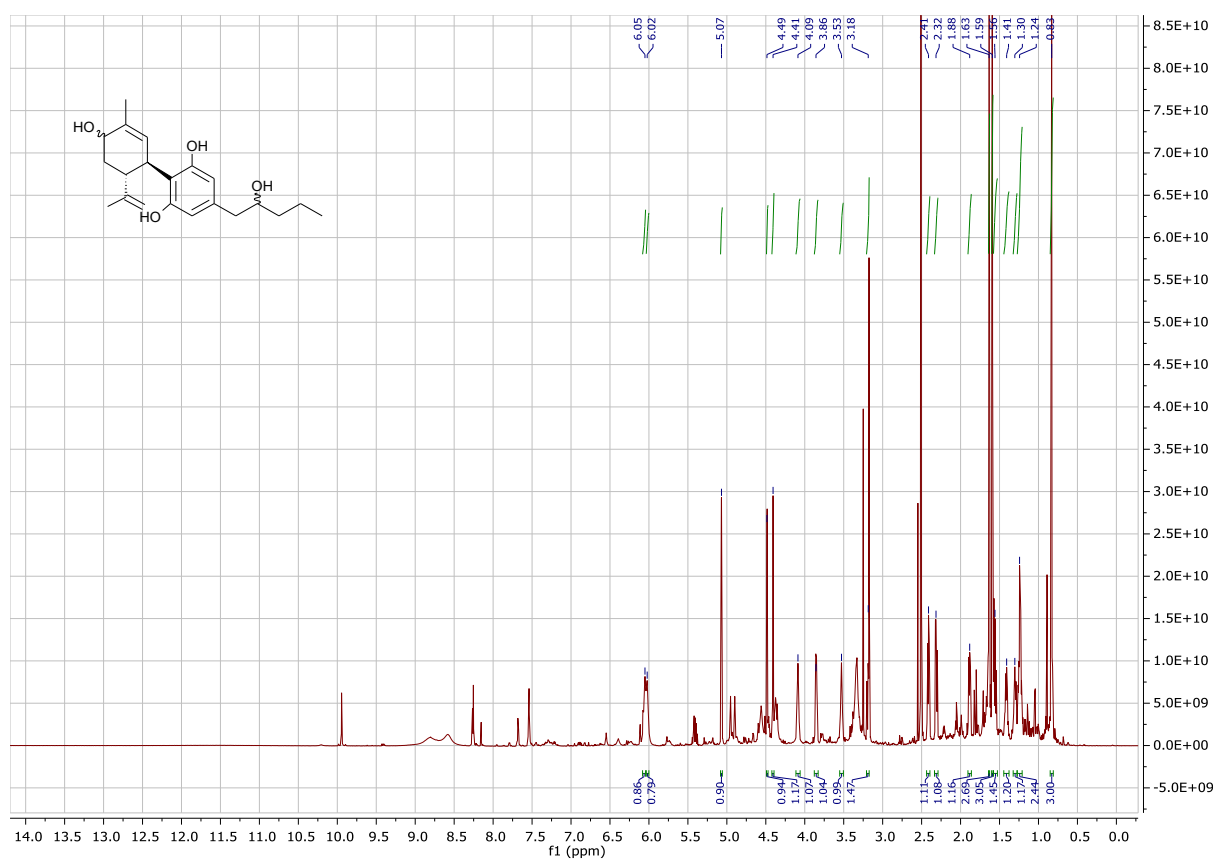


Figure S77. ^1H NMR spectrum of metabolite 9

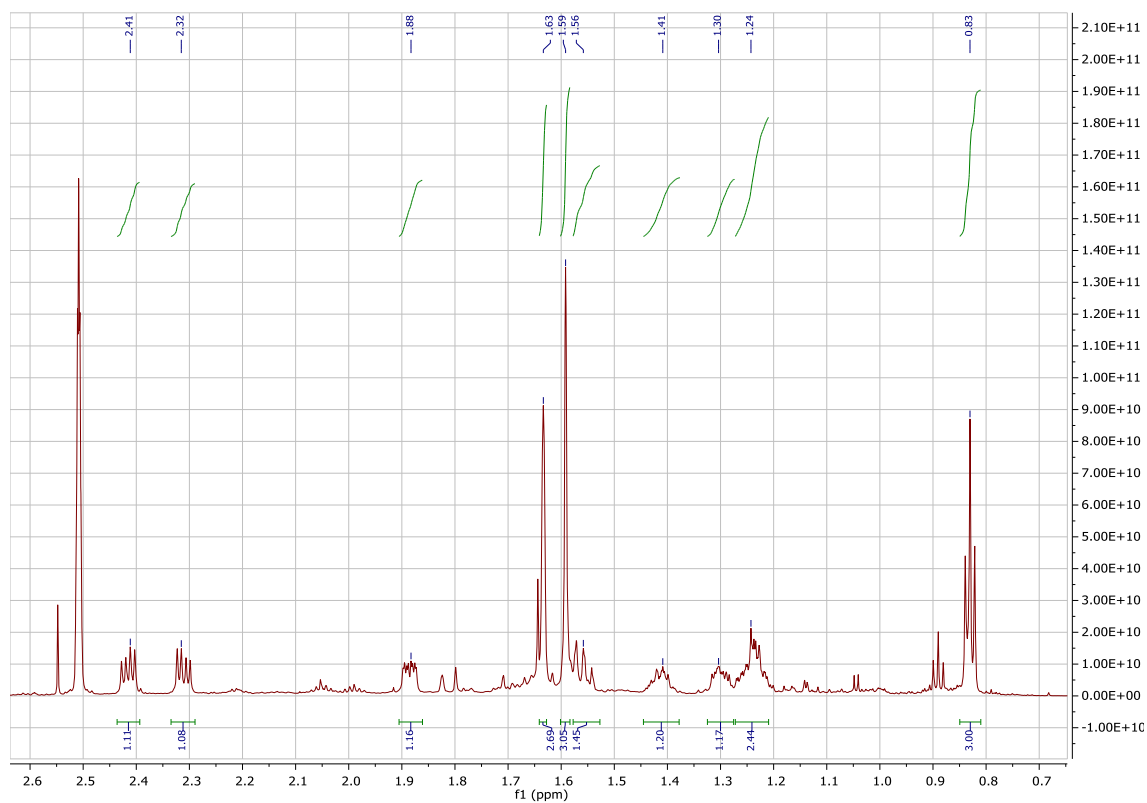


Figure S78. ^1H NMR (0.7-2.6 ppm) spectrum of metabolite 9

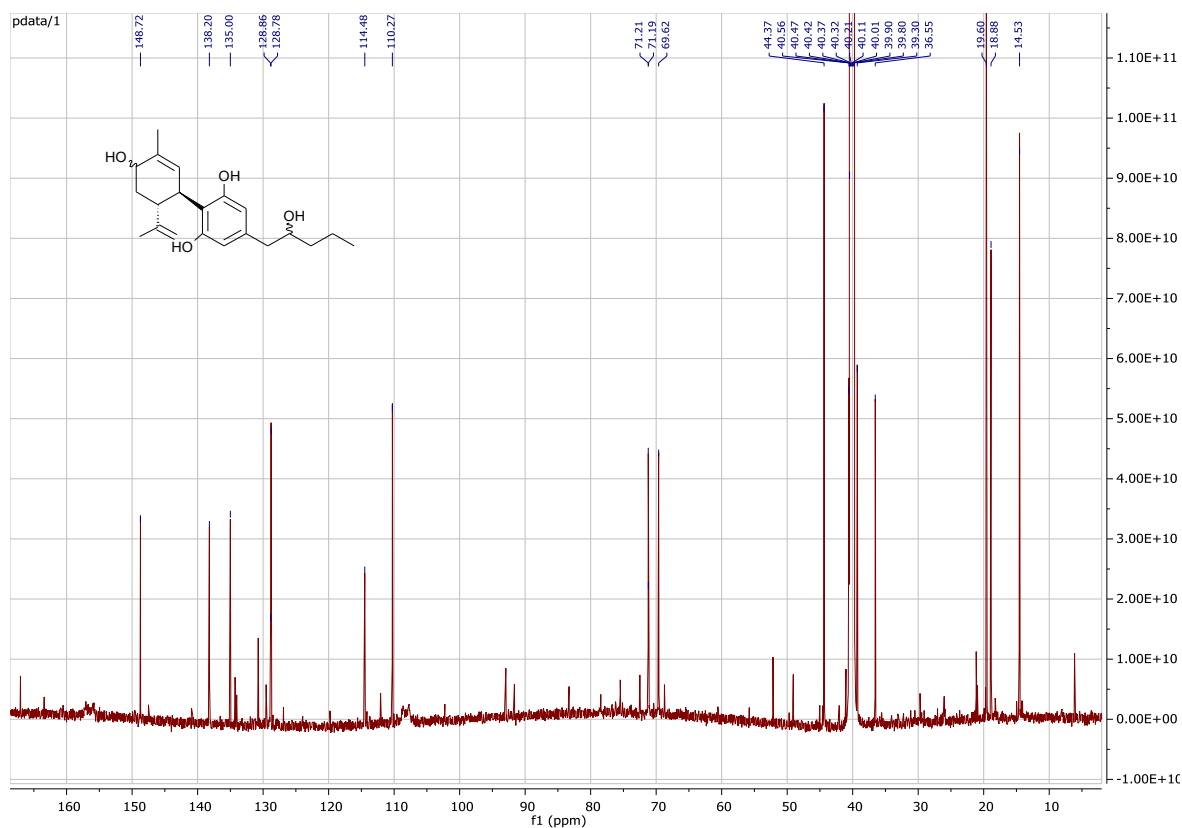


Figure S79. ^{13}C NMR spectrum of metabolite 9

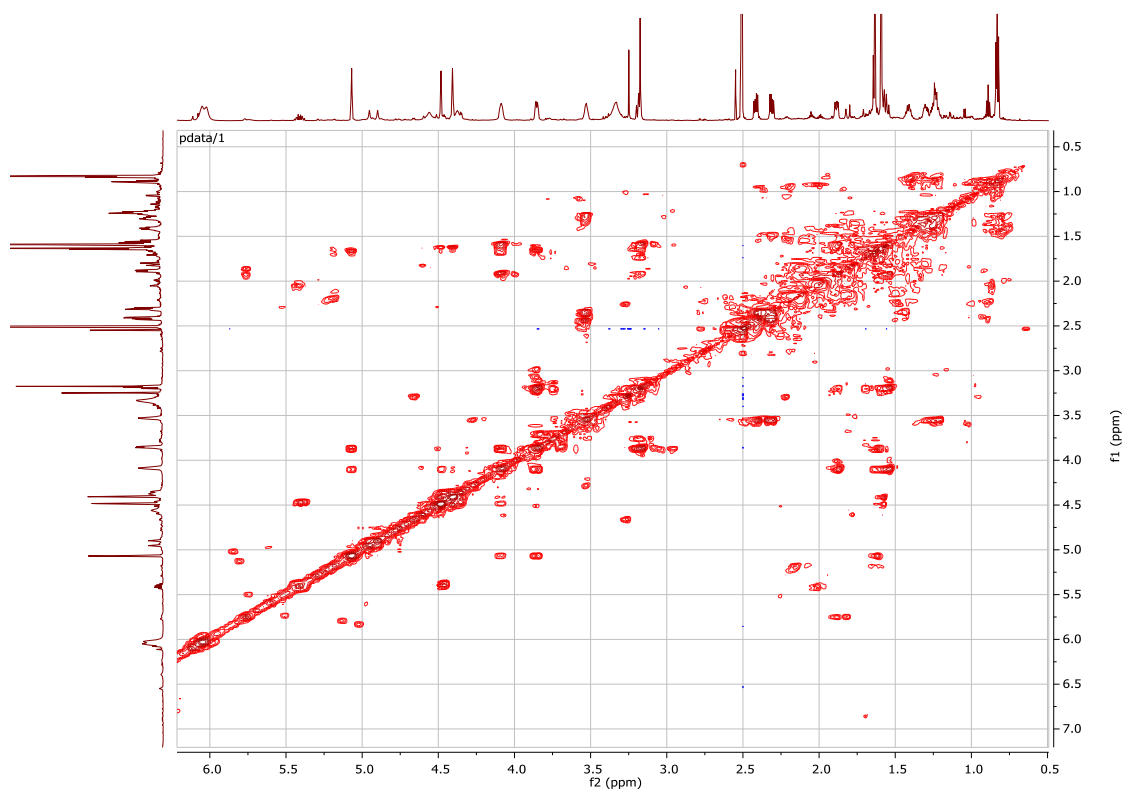


Figure S80. COSY (^1H - ^1H) NMR spectrum of metabolite 9

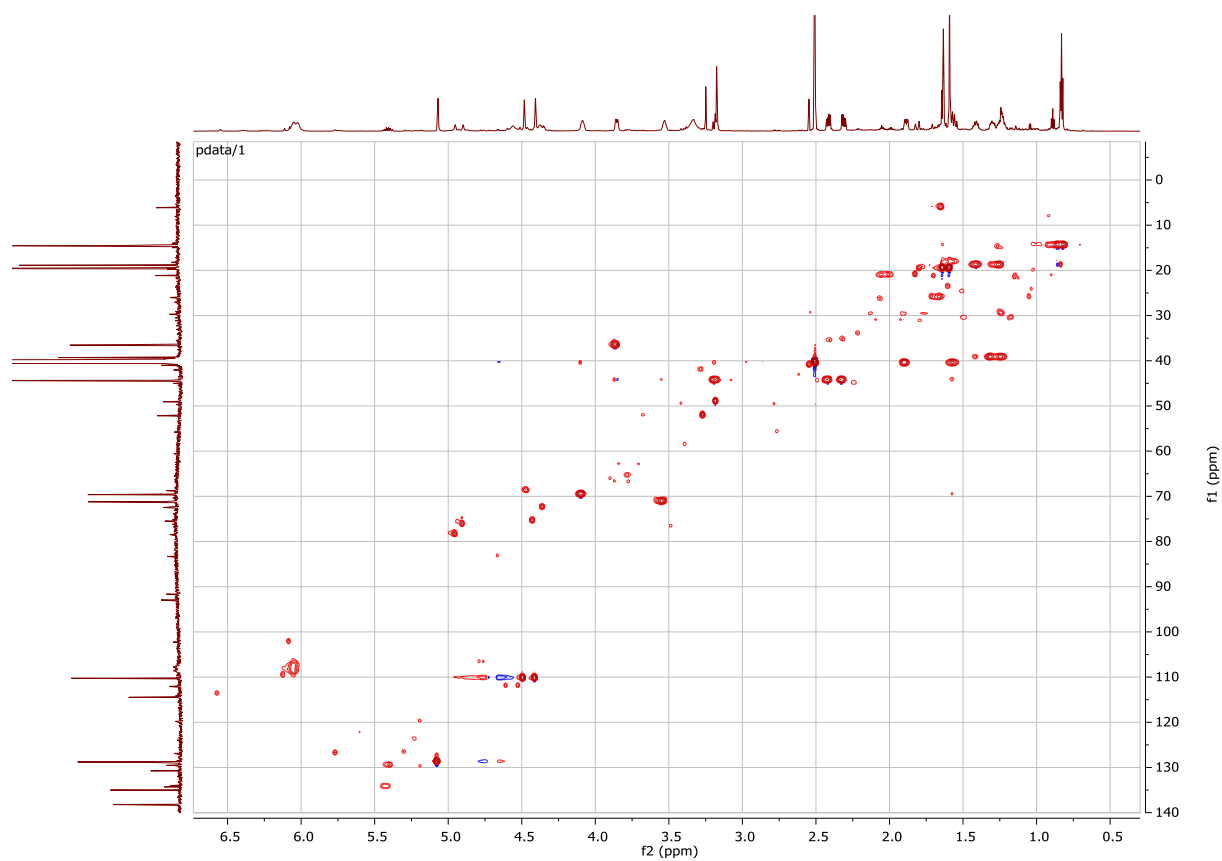


Figure S81. HSQC (^1H - ^{13}C) NMR spectrum of metabolite **9**

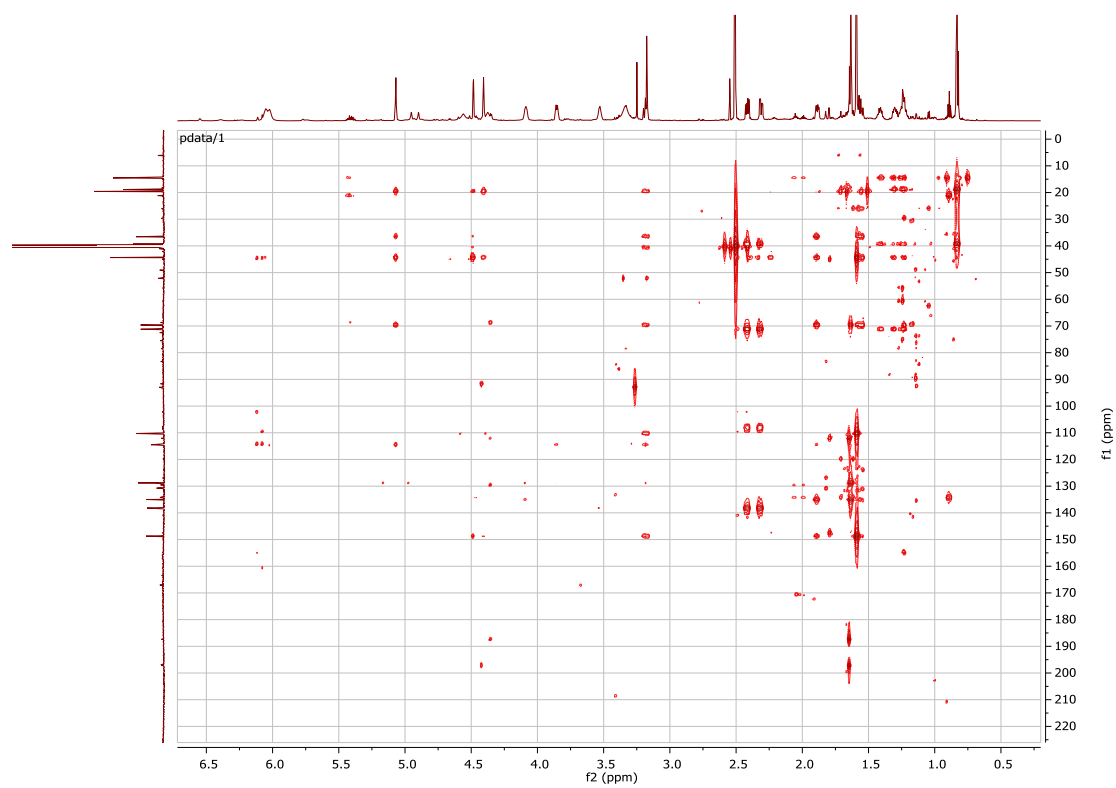


Figure S82. HMBC (long distance correlation ^1H - ^{13}C) NMR spectrum of metabolite **9**

10. Statistical analysis

Table S1. One-way ANOVA – Time significance

One-way ANOVA

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Mean</i>	<i>Variance</i>
Day 1	3	12.05135	4.017115	0.106401
Day 3	3	9.621467	3.207156	0.296782
Day 5	3	8.871741	2.957247	0.029211
Day 7	3	8.02416	2.67472	0.584577

One-way ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F critical</i>
Between Groups	3.005357	3	1.001786	3.940271	0.05368	4.066181
Within Groups	2.033943	8	0.254243			
Total	5.039299	11				

Table S2. One-way ANOVA and Student's t-test– Metabolite **2** over time – *I. fumosorosea* KCh J2

One-way ANOVA

SUMMARY				
Groups	Count	Sum	Mean	Variance
Day 1	3	8.916773	2.972258	0.154992
Day 3	3	8.743541	2.914514	0.027841
Day 5	3	6.885831	2.295277	0.003847
Day 7	3	5.308234	1.769411	0.042277

One-way ANOVA						
Source of Variation	SS	df	MS	F	P-value	F critical
Between Groups	2.909794	3	0.969931	16.94518	0.000794	4.066181
Within Groups	0.457915	8	0.057239			
Total	3.367708	11				

Metabolite 2		
Day 1	Day 7	T-student
3.411478	1.682688	
2.651127	2.004179	0.018109
2.854169	1.621366	

Table S3. One-way ANOVA and Student's t-test– Metabolite **8** over time - *I. fumosorosea* KCh J2

One-way ANOVA

SUMMARY				
Groups	Count	Sum	Mean	Variance
Day 1	3	3.398848	1.132949	0.672694
Day 3	3	6.046763	2.015588	0.137246
Day 5	3	6.820451	2.273484	0.062326
Day 7	3	7.398666	2.466222	0.066547

One-way ANOVA						
Source of Variation	SS	df	MS	F	P-value	F critical
Between Groups	3.123161	3	1.041054	4.435615	0.040871	4.066181
Within Groups	1.877627	8	0.234703			
Total	5.000788	11				

Metabolite 8		
Day 1	Day 7	T-Student
2.079442	2.470639	
0.631272	2.206074	0.095257
0.688135	2.721953	

Table S4. One-way ANOVA and Student's t-test– Metabolite 7 over time - *I. fumosorosea* KCh J2

One-way ANOVA

SUMMARY				
Groups	Count	Sum	Mean	Variance
Day 1	3	8.599751	2.866584	0.07391
Day 3	3	11.50696	3.835655	0.014395
Day 5	3	12.31786	4.105952	0.000467
Day 7	3	12.50768	4.169226	0.013407

One-way ANOVA						
Source of Variation	SS	df	MS	F	P-value	F critical
Between Groups	3.270257	3	1.090086	42.67314	2.87E-05	4.066181
Within Groups	0.20436	8	0.025545			
Total	3.474617	11				

Metabolite 7		
Day 1	Day 7	T-student
3.160399	4.080753	
2.815409	4.12665	0.006657
2.623944	4.300274	

Table S5. Student's t-test– *M. hiemalis* strains

			Ln	Ln	
	KCh W2	AM 450	KCh W2	AM450	T-Student (p)
CBD	1.82	35.6	0.598836501	3.572345638	
	1.36	14.02	0.3074847	2.640484882	0.003124556
	0.6	15.8	-0.510825624	2.76000994	
Metabolite 2	69.76	25.99	4.24506078	3.257711849	
	64.14	29.08	4.161068194	3.370050653	0.002289354
	61.5	22.11	4.119037175	3.096029995	
Metabolite 3	21.66	1.39	3.075467242	0.329303747	
	23.59	0.74	3.160822893	-0.301105093	0.023931245
	27.27	0.18	3.305787197	-1.714798428	
Metabolite 4	2.07	31.24	0.727548607	3.441699325	
	0.31	49.01	-1.171182982	3.892024359	0.014433334
	0.38	53.07	-0.967584026	3.971611797	
Metabolite 5	3.06	2.97	1.118414916	1.088561953	
	4.53	4.16	1.510721939	1.425515074	0.665629202
	3.6	5.42	1.280933845	1.690095815	
Metabolite 6	1.63	2.82	0.488580015	1.036736885	
	6.08	2.99	1.805004696	1.095273387	0.606964905
	6.65	3.42	1.894616855	1.229640551	