Supporting Information (SI)

Efficient regioselective enzymatic acylation of troxerutin: Difference characterization in vitro cellular uptake and cytotoxicity

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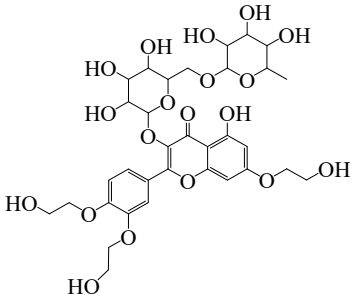
LC-MS diagram of ester formed after simulated digestion *in vitro*

Table S1 **PS23-S25**

The chemical shift comparation of TX and its esters in ^{13}C-NMR

HRMS, FT-IR and ^{13}C NMR spectra of troxerutin and its esters

Troxerutin



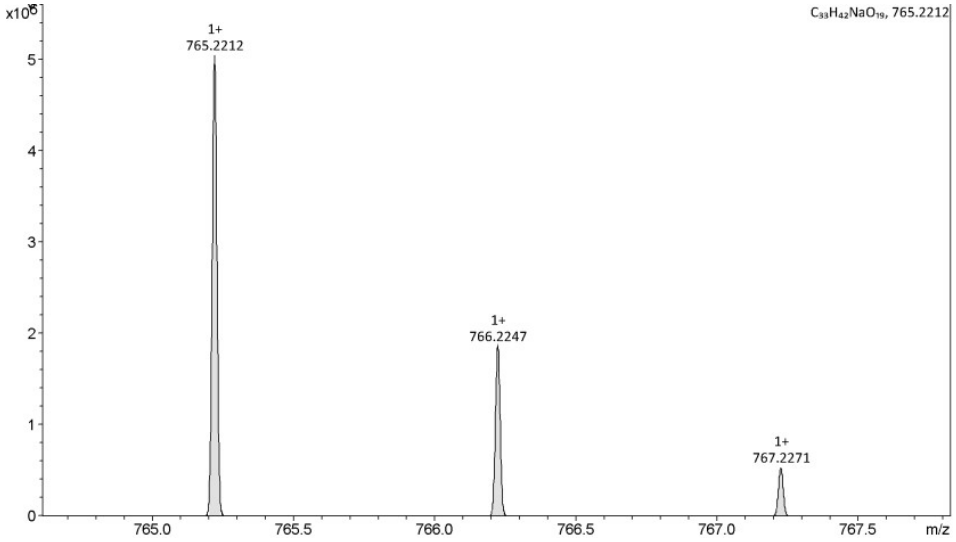


Figure S1. HRMS spectrum for troxerutin.

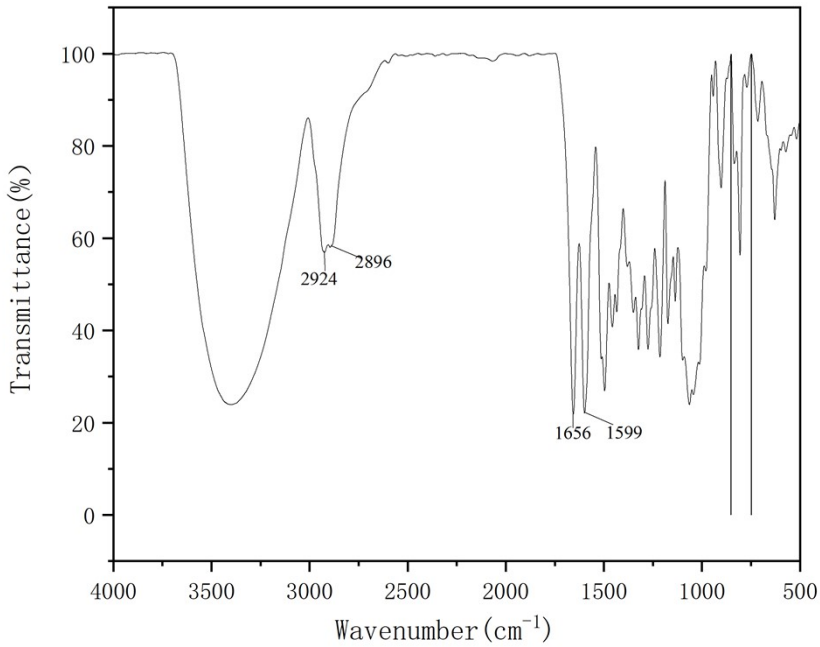


Figure S2. FT-IR spectrum for troxerutin.

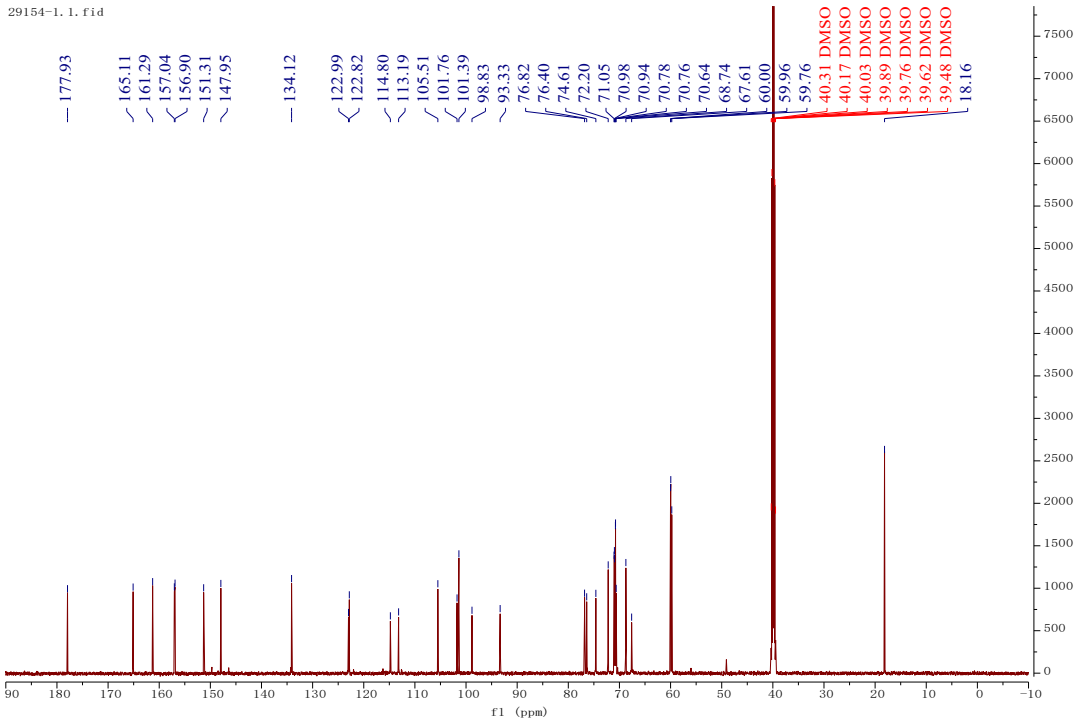
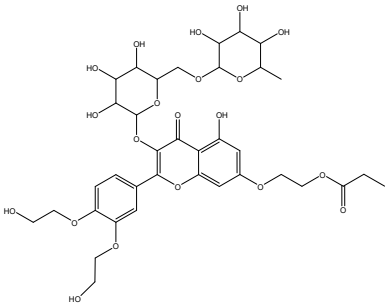


Figure S3. ^{13}C NMR spectrum for troxerutin in DMSO-d_6 (600 MHz).

Troxerutin monopropyl ester



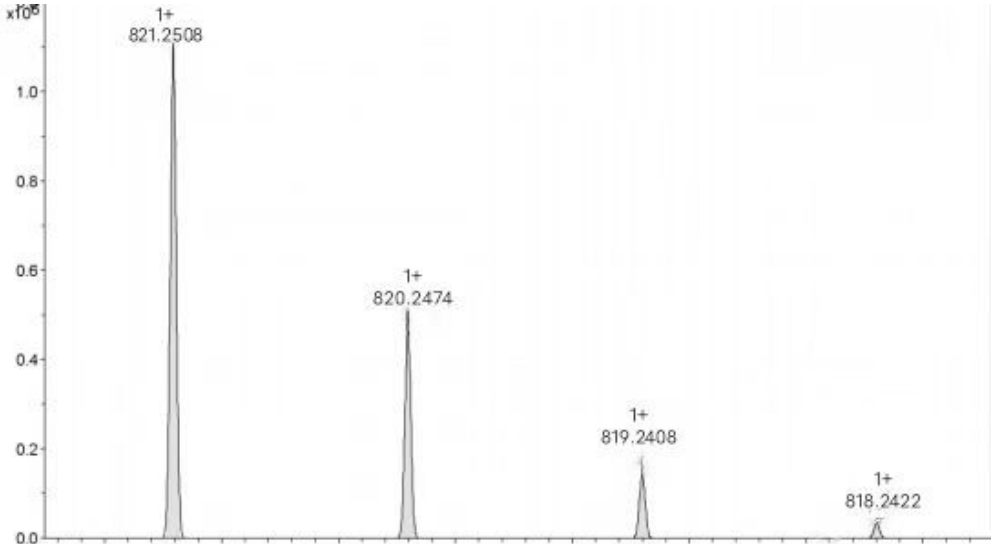


Figure S4. HRMS spectrum for troxerutin monopropyl ester.

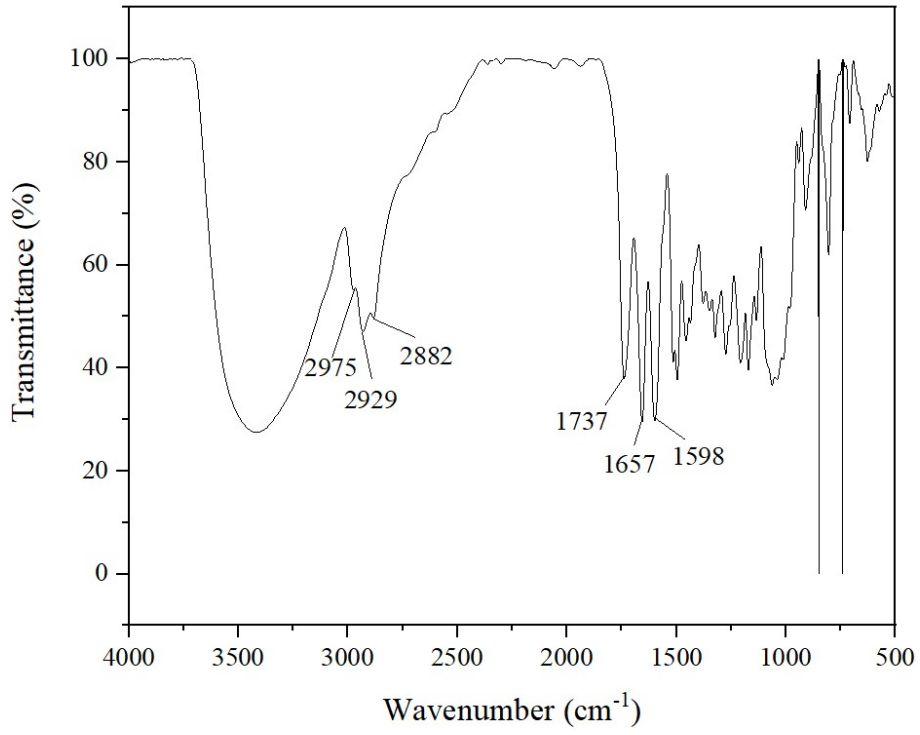


Figure S5. FT-IR spectrum for troxerutin monopropyl ester.

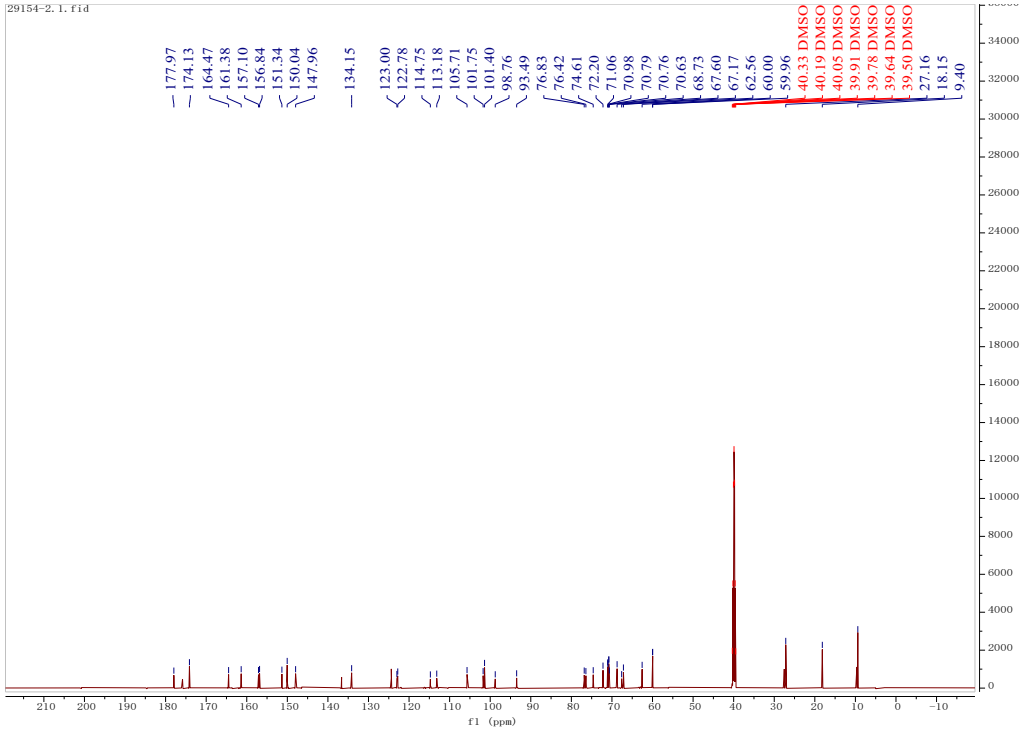
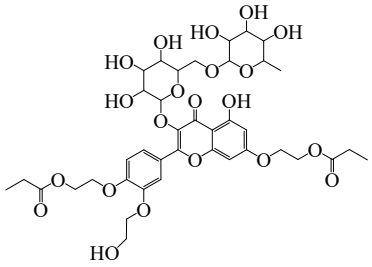


Figure S6. ^{13}C NMR spectrum for troxerutin monopropyl ester in DMSO-d_6 (600 MHz).

Troxerutin dipropyl ester



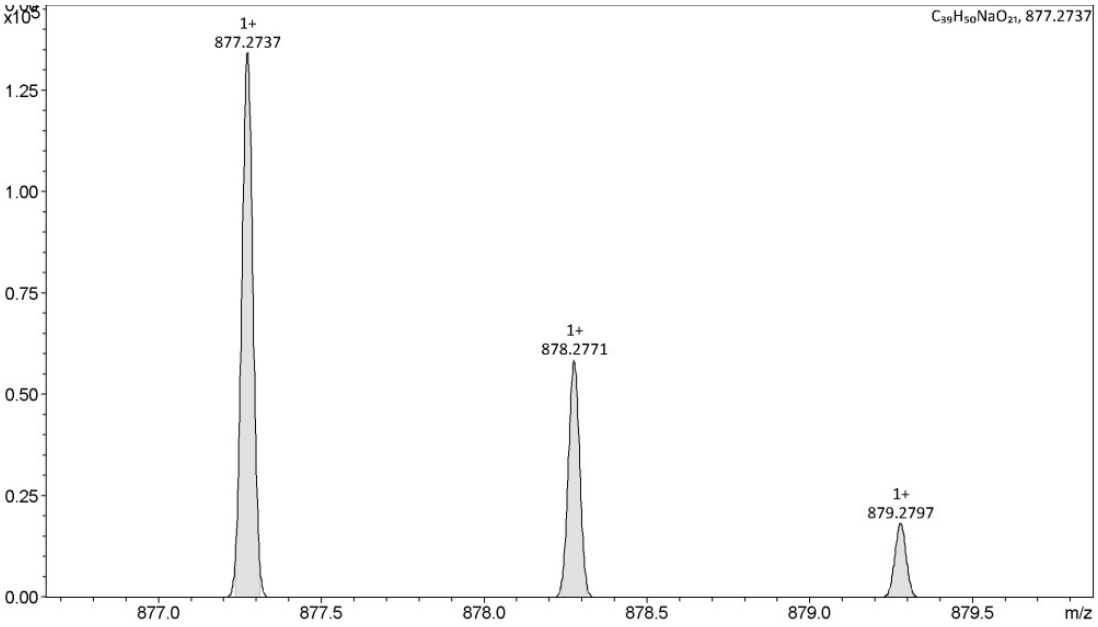


Figure S7. HRMS spectrum for troxerutin dipropyl ester.

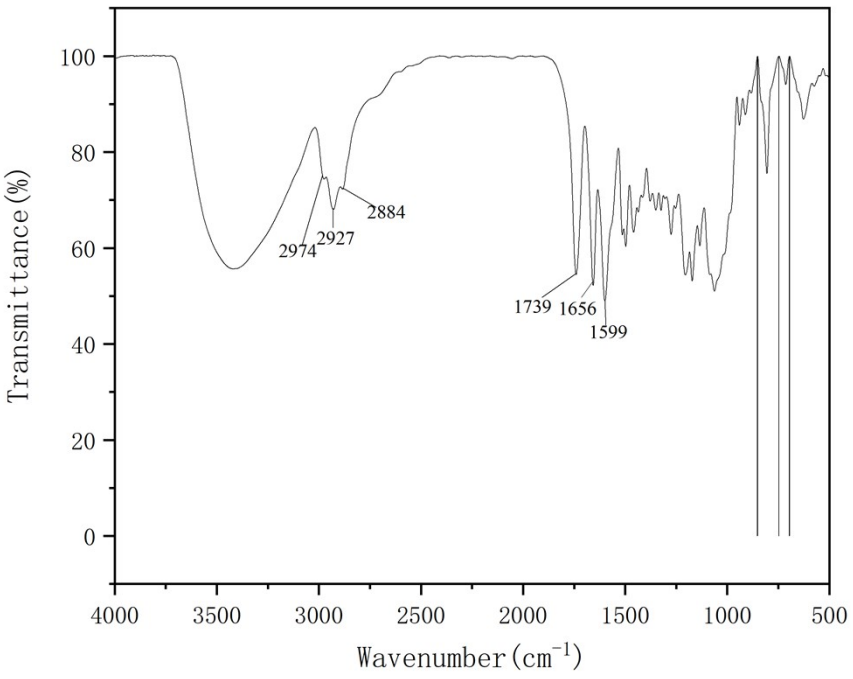


Figure S8. FT-IR spectrum for troxerutin dipropyl ester.

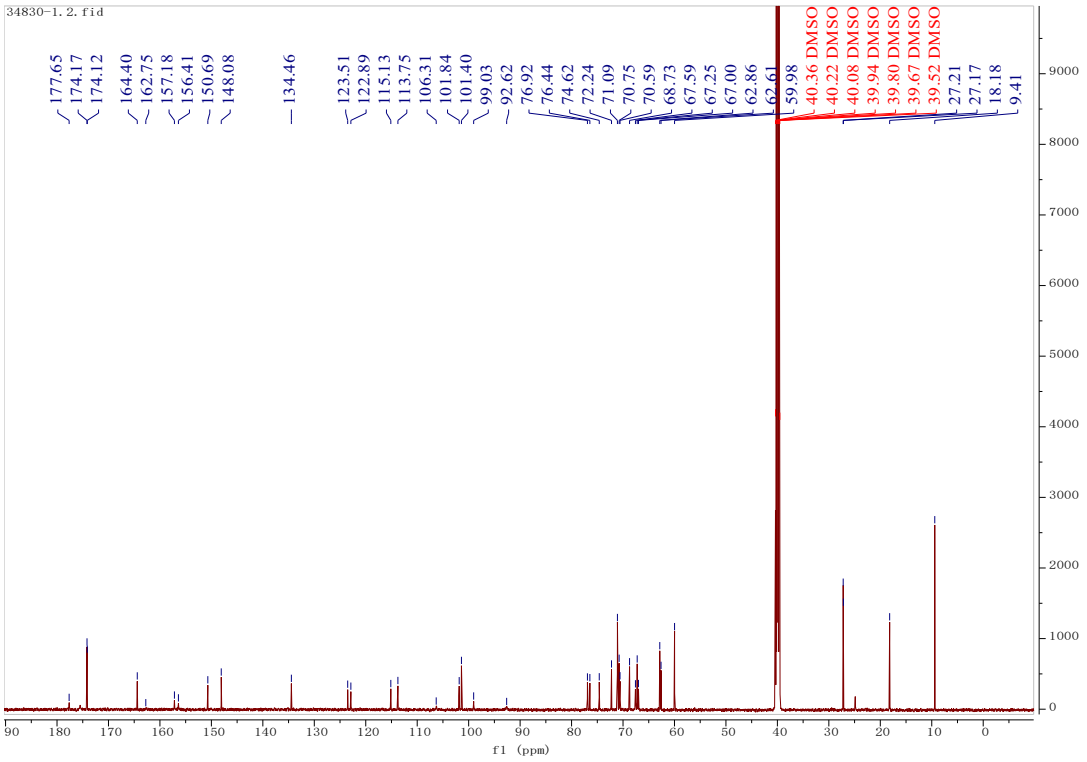
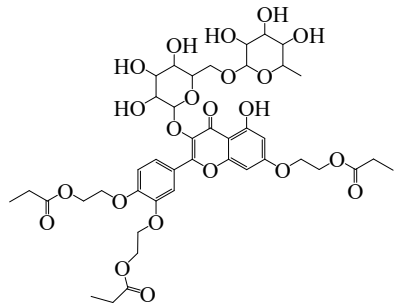


Figure S9. ^{13}C NMR spectrum for troxerutin dipropyl ester in DMSO-d_6 (600 MHz).

Troxerutin tripropyl ester



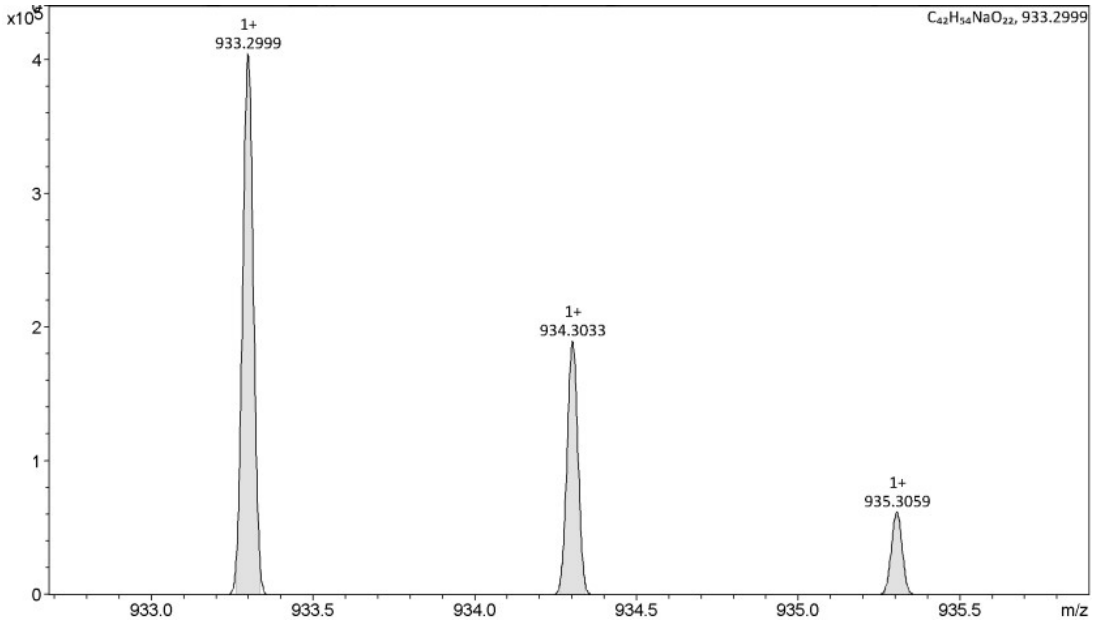


Figure S10. HRMS spectrum for troxerutin tripropyl ester.

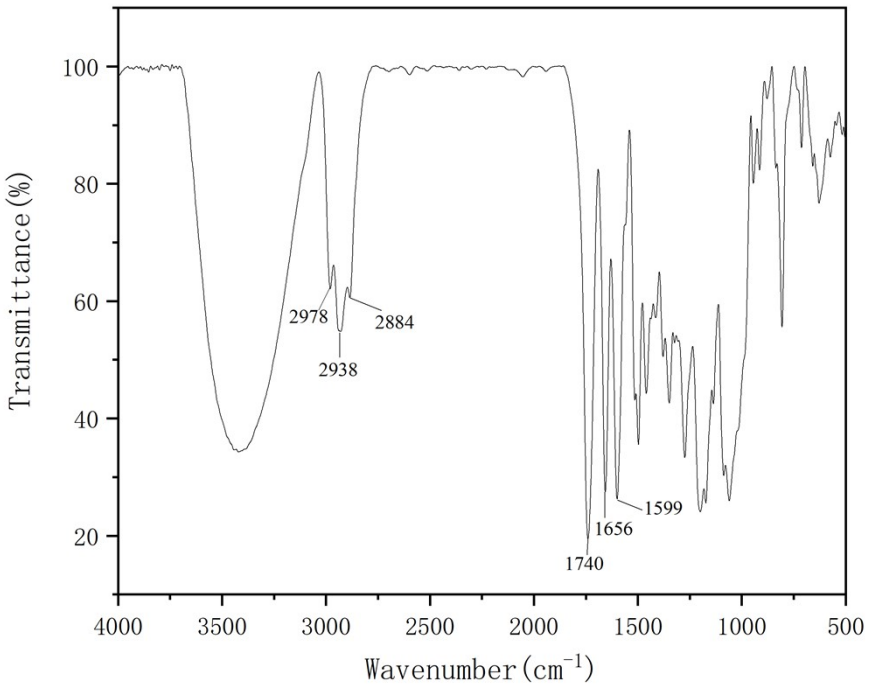


Figure S11. FT-IR spectrum for troxerutin tripropyl ester.

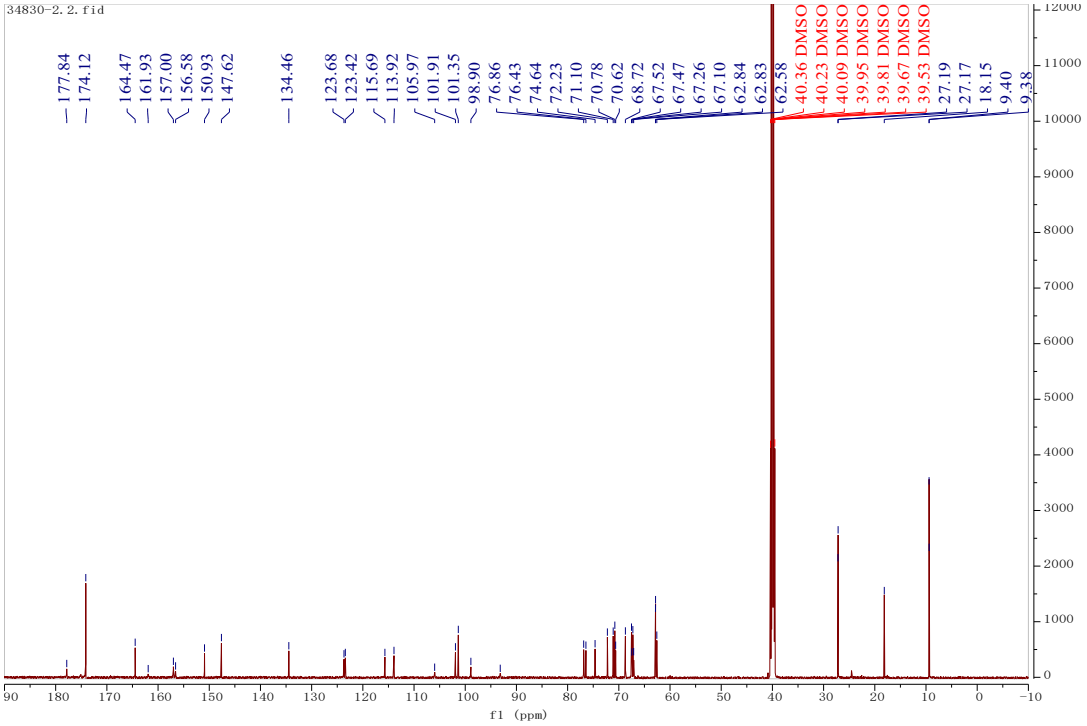
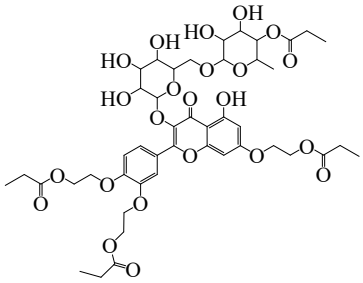


Figure S12. ^{13}C NMR spectrum for troxerutin tripropyl ester in DMSO-d_6 (600 MHz).

Troxerutin tetrapropyl ester



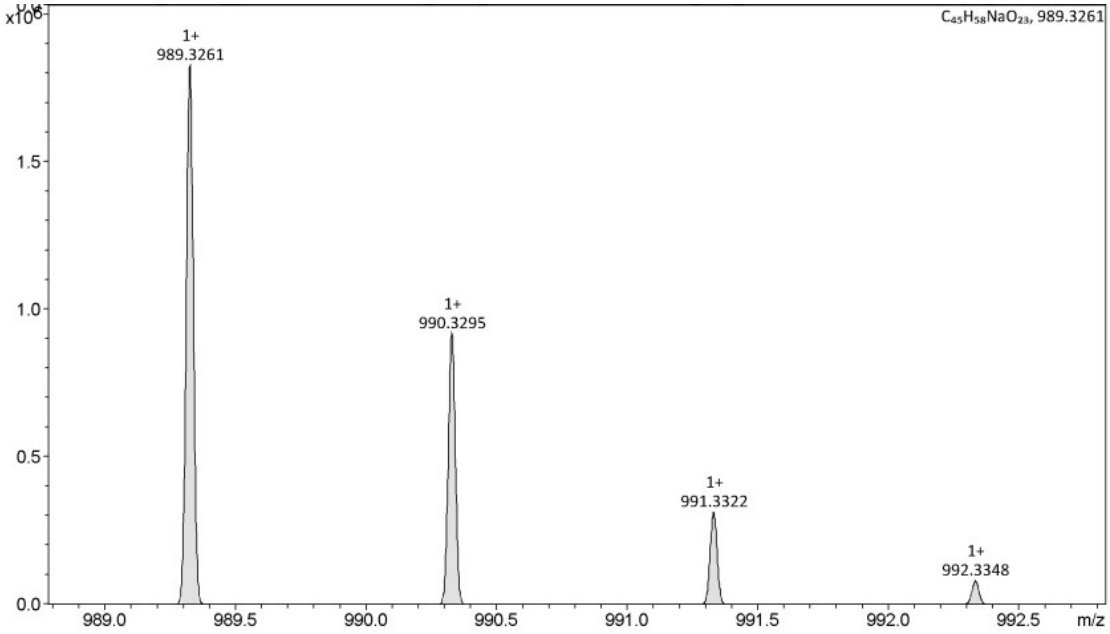


Figure S13. HRMS spectrum for troxerutin tetrapropyl ester.

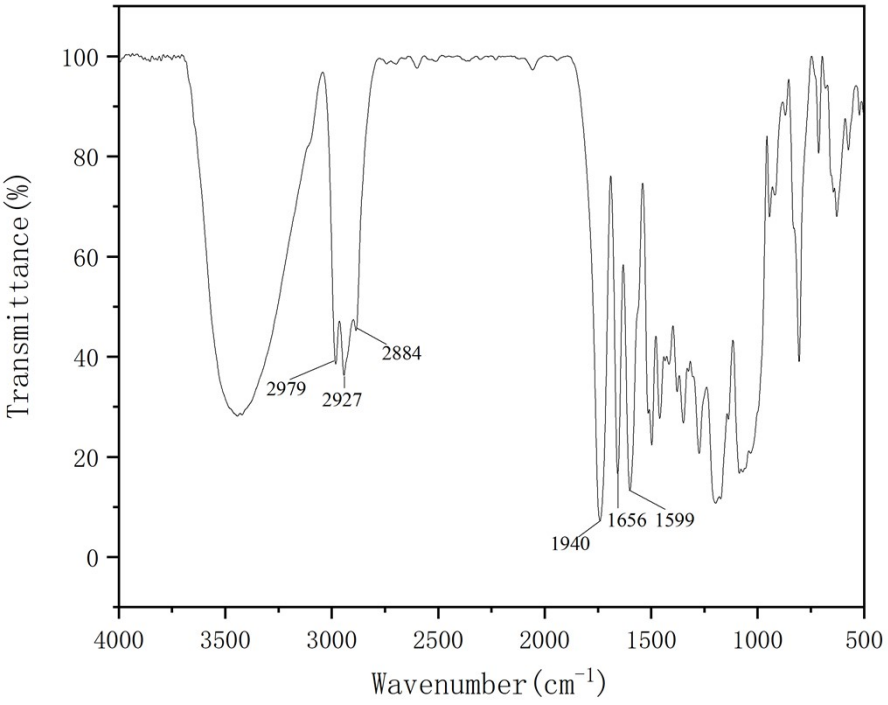


Figure S14. FT-IR spectrum for troxerutin tetrapropyl ester.

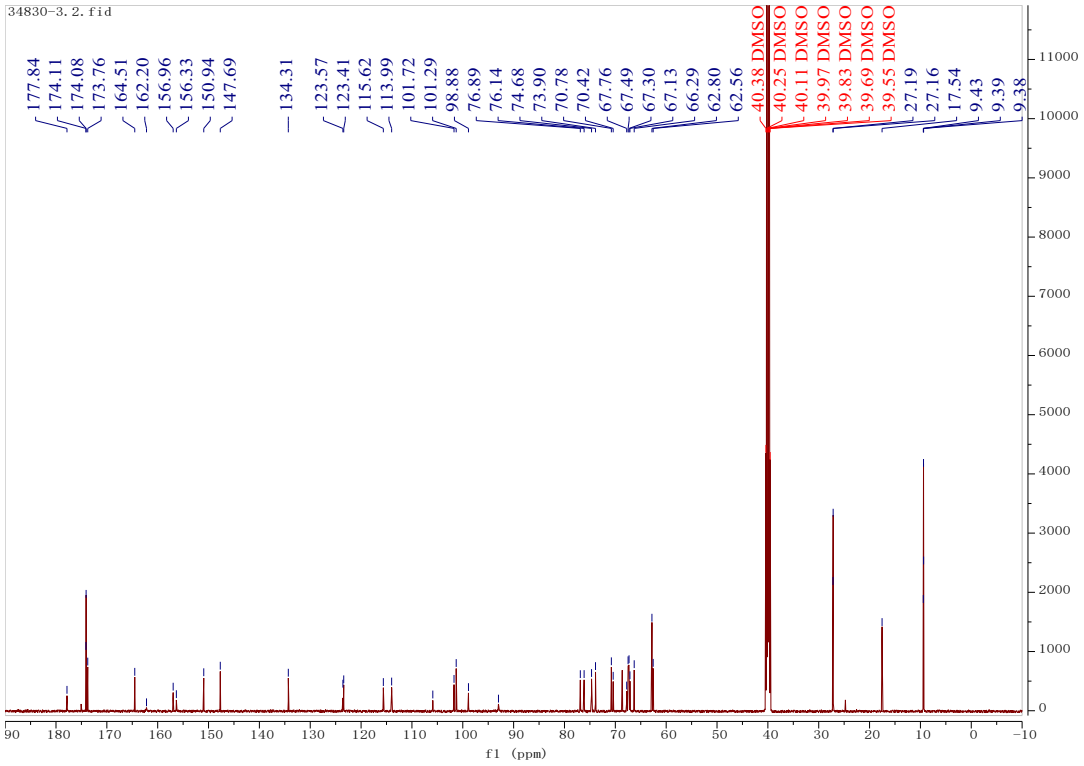
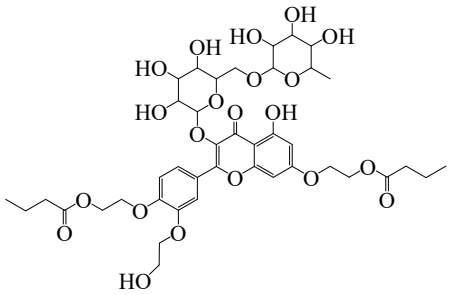


Figure S15. ^{13}C NMR spectrum for troxerutin tetrapropyl ester in DMSO-d_6 (600 MHz).

Troxerutin dibutyl ester



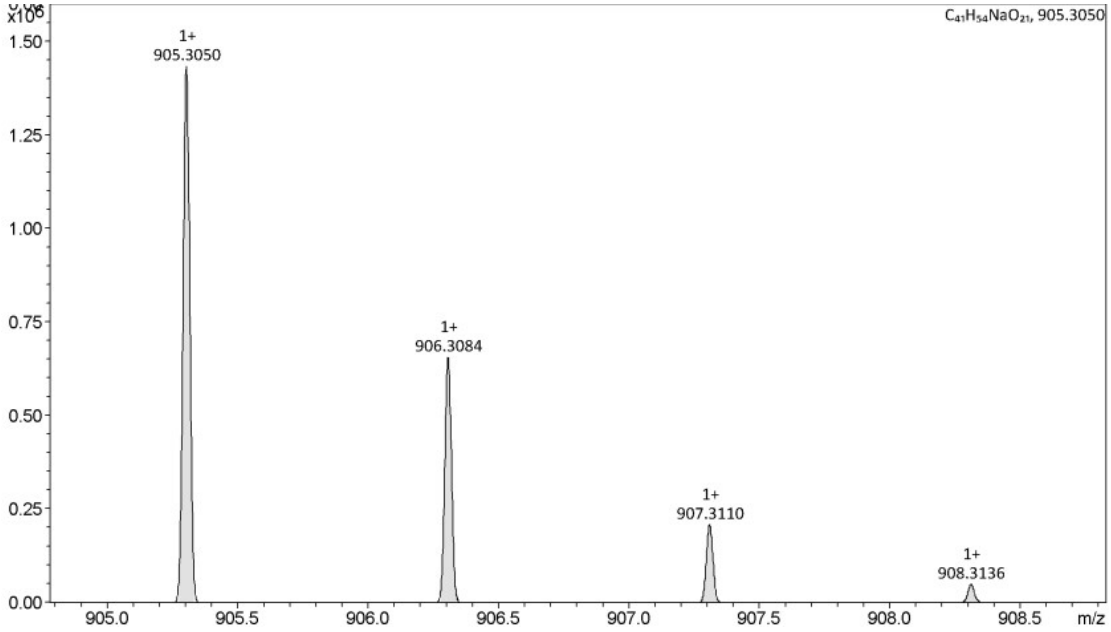


Figure S16. HRMS spectrum for troxerutin dibutyl ester.

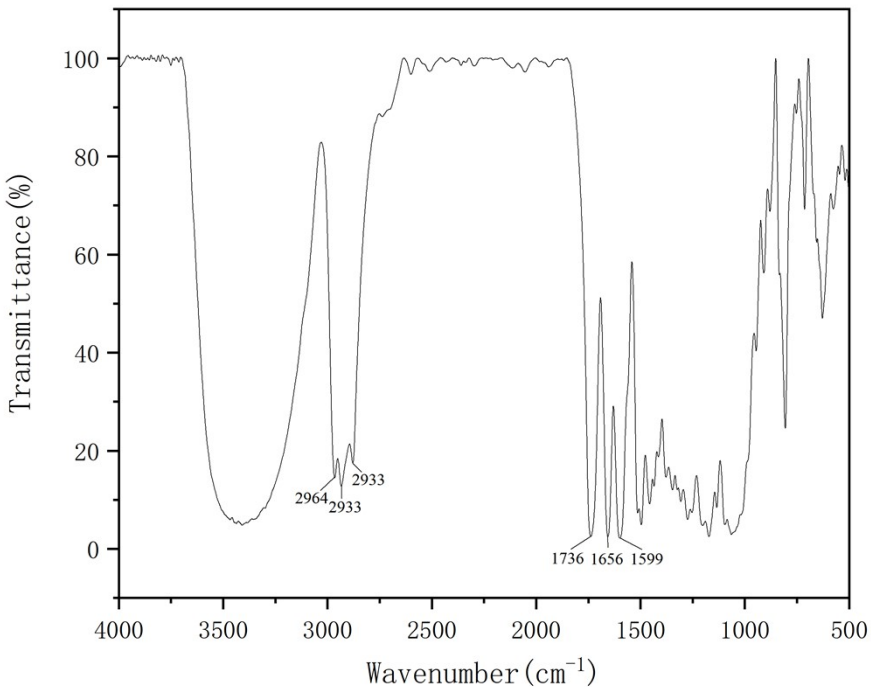


Figure S17. FT-IR spectrum for troxerutin dibutyl ester.

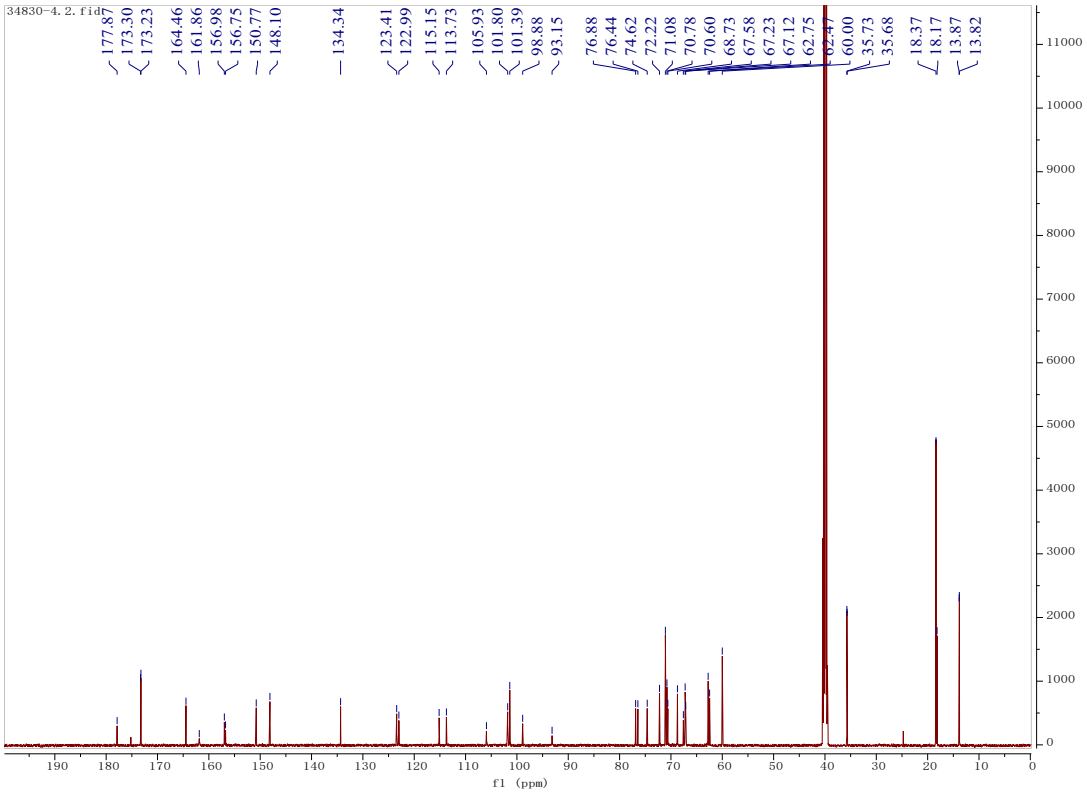
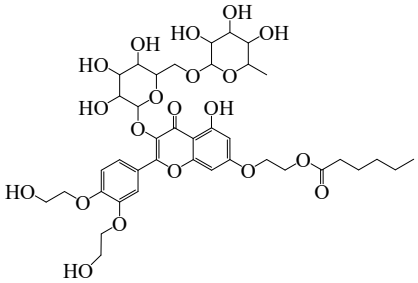


Figure S18. ^{13}C NMR spectrum for troxerutin dibutyl ester in DMSO-d_6 (600 MHz).

Troxerutin monohexyl ester



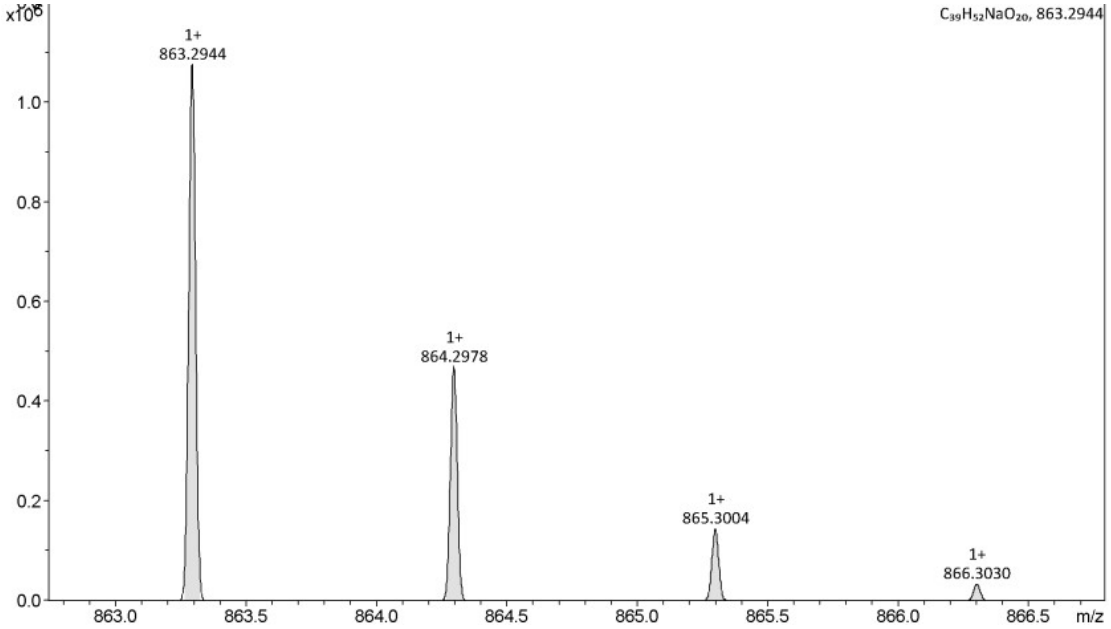


Figure S19. HRMS spectrum for troxerutin monohexyl ester.

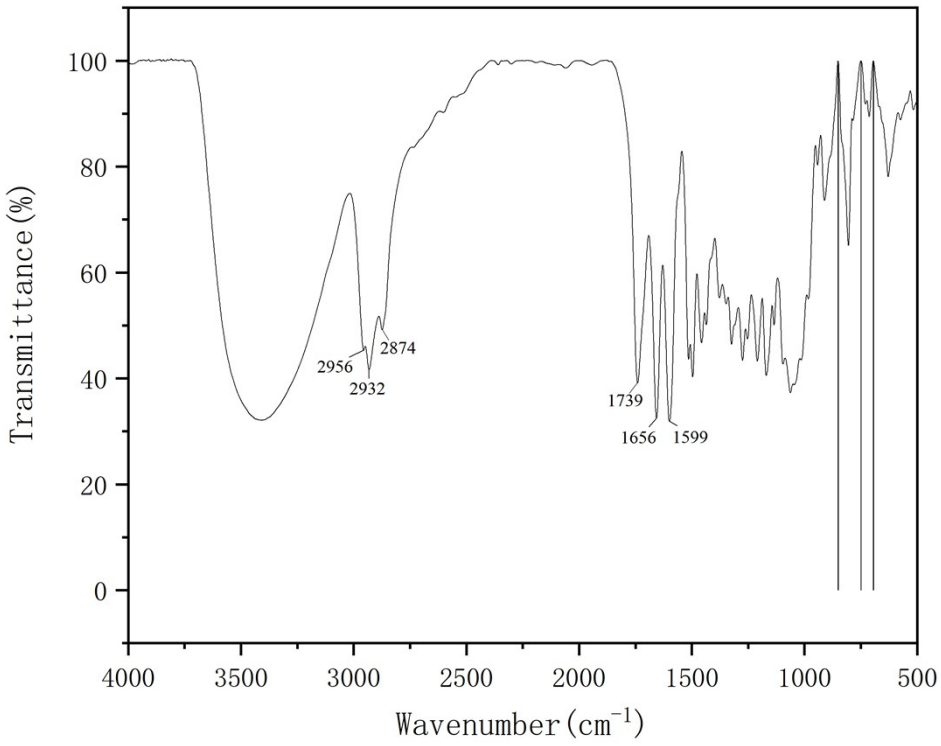


Figure S20. FT-IR spectrum for troxerutin monohexyl ester.

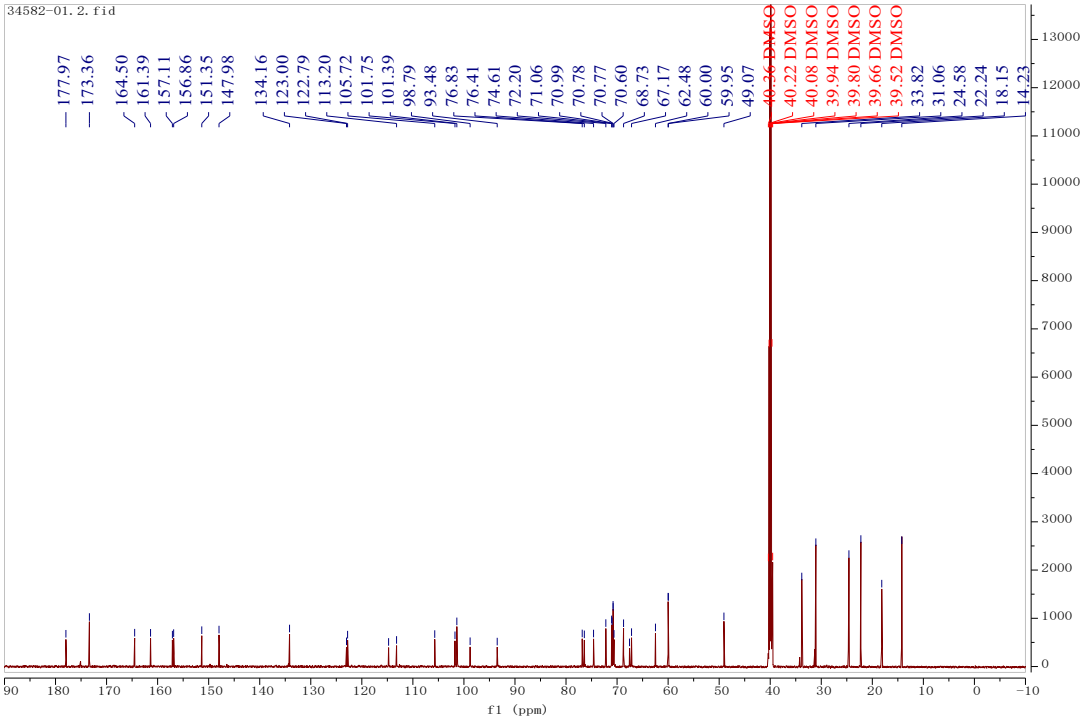
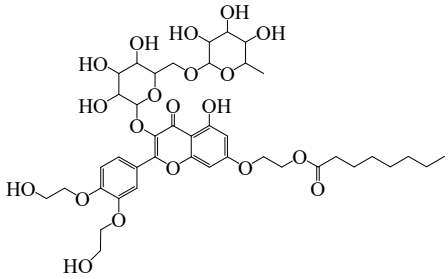


Figure S21. ^{13}C NMR spectrum for troxerutin monohexyl ester in DMSO-d_6 (600 MHz).

Troxerutin monooctyl ester



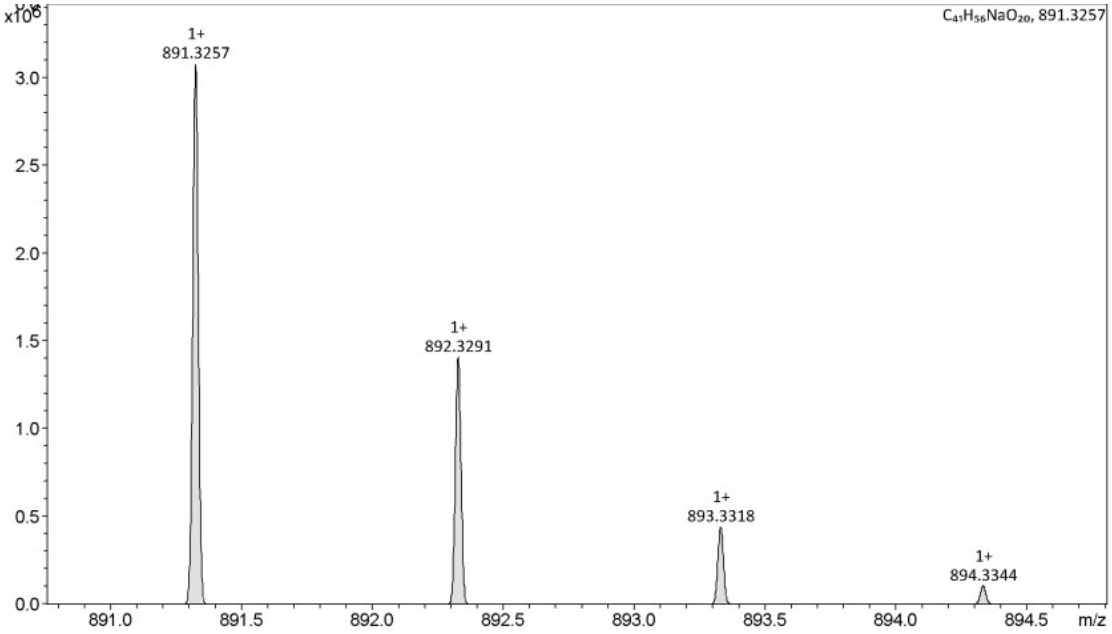


Figure S22. HRMS spectrum for troxerutin monooctyl ester.

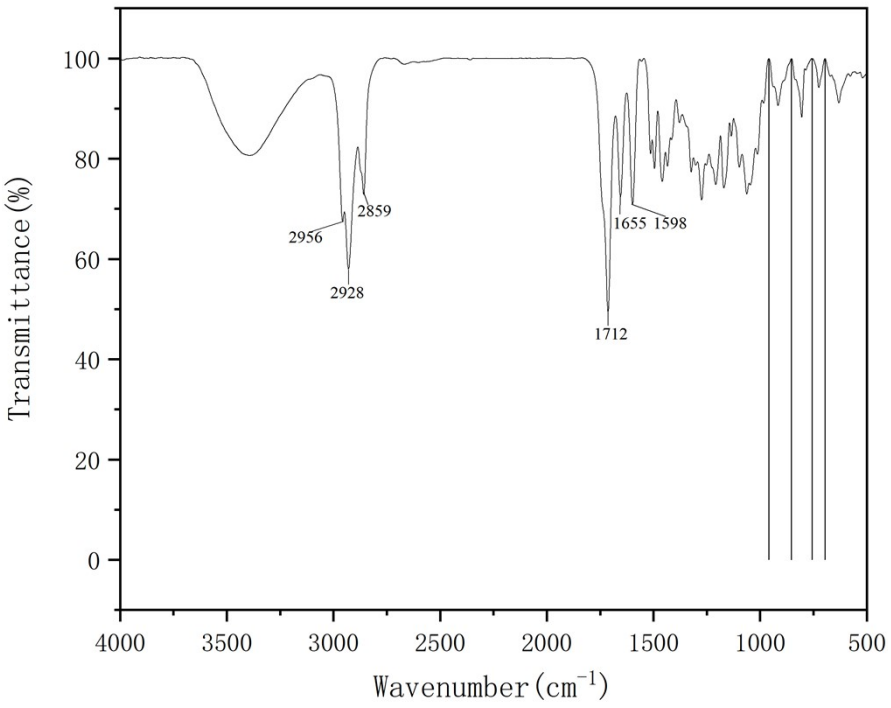


Figure S23. FT-IR spectrum for troxerutin monooctyl ester.

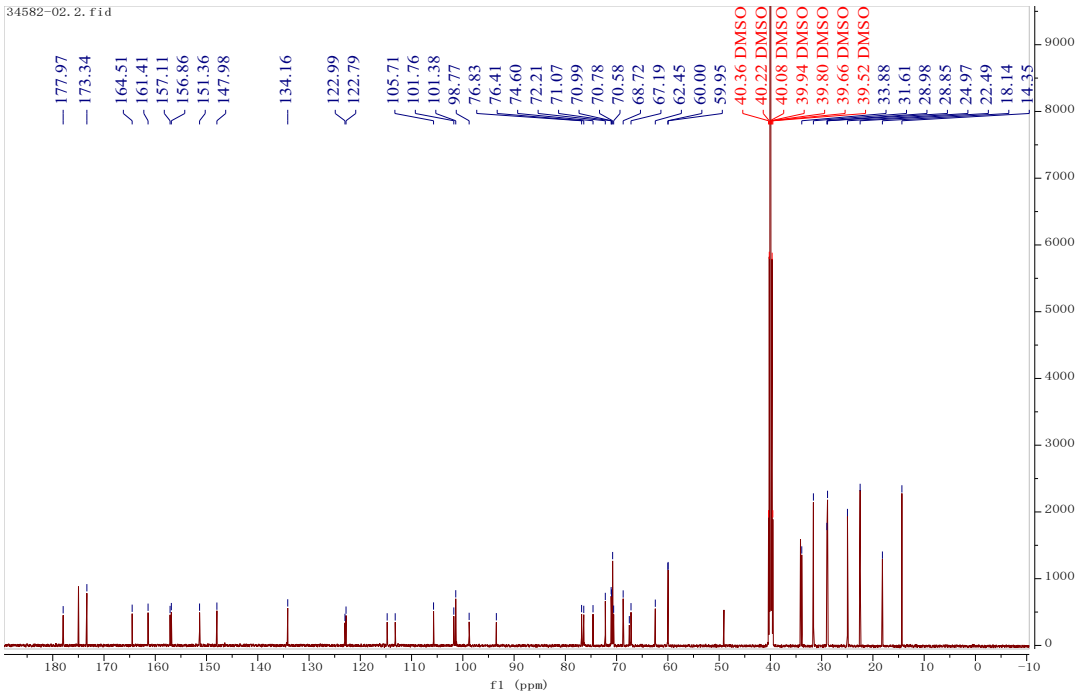
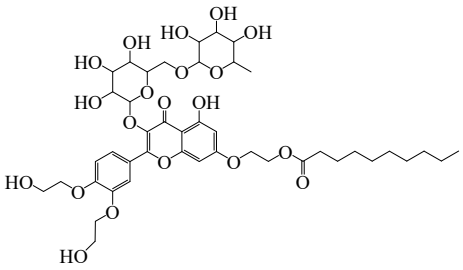


Figure S24. ^{13}C NMR spectrum for troxerutin monooctyl ester in DMSO-d_6 (600 MHz).

Troxerutin monodecyl ester



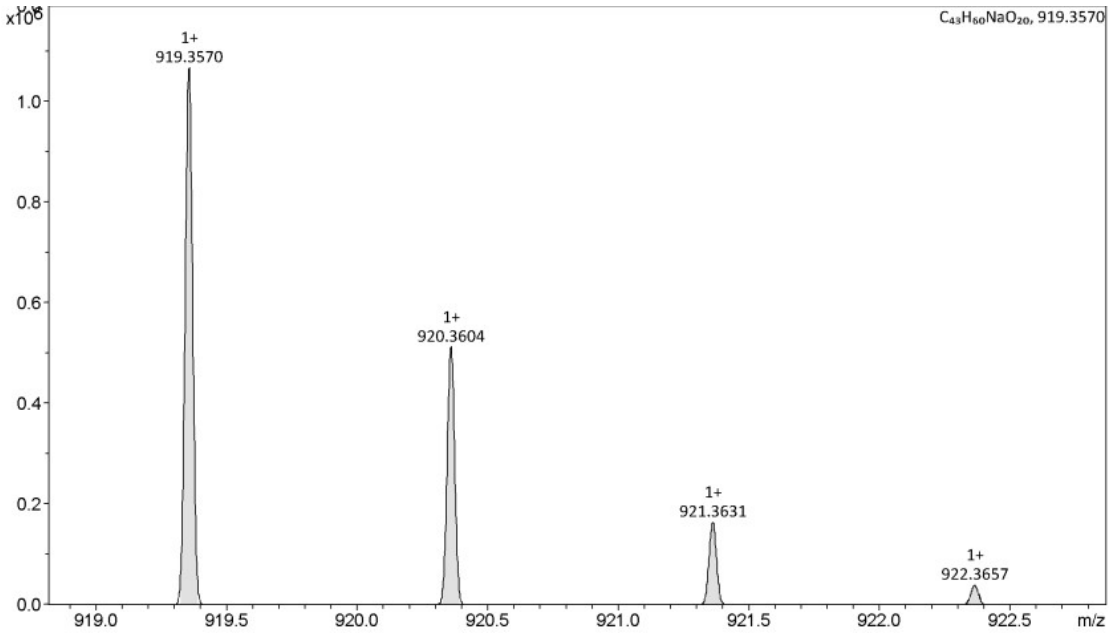


Figure S25. HRMS spectrum for troxerutin monodecyl ester.

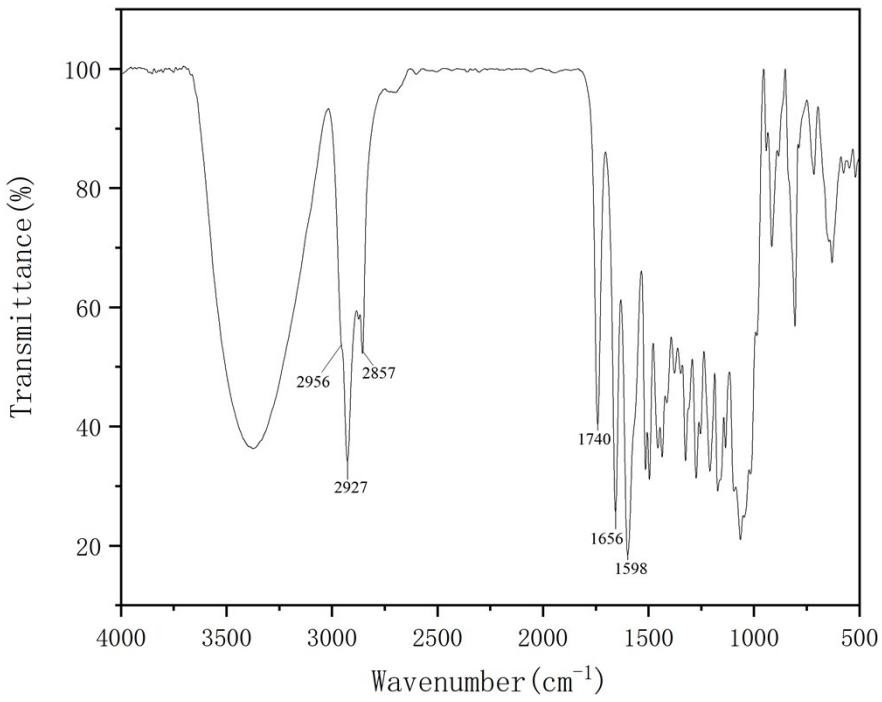


Figure S26. FT-IR spectrum for troxerutin monodecyl ester.

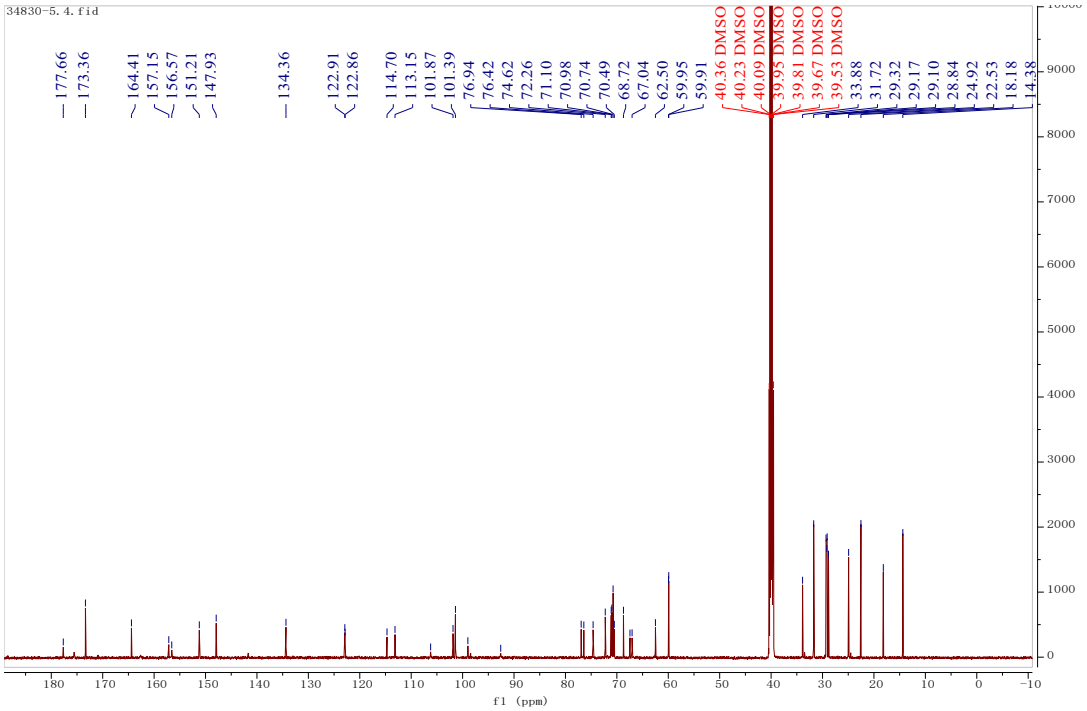
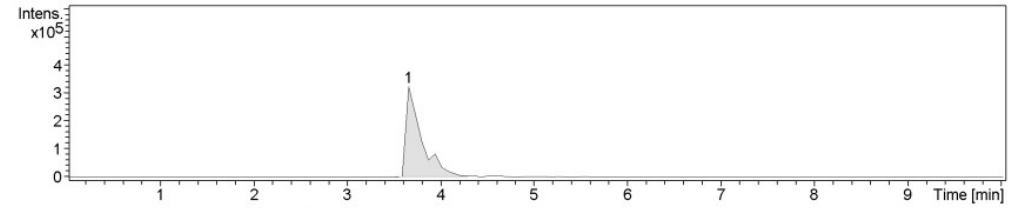
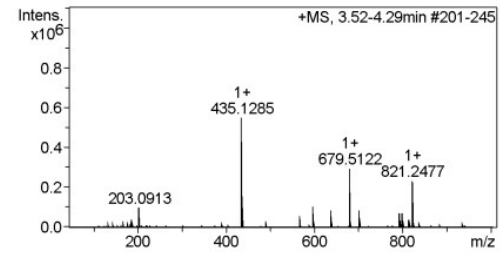


Figure S27. ^{13}C NMR spectrum for troxerutin monodecyl ester in DMSO-d_6 (600 MHz).



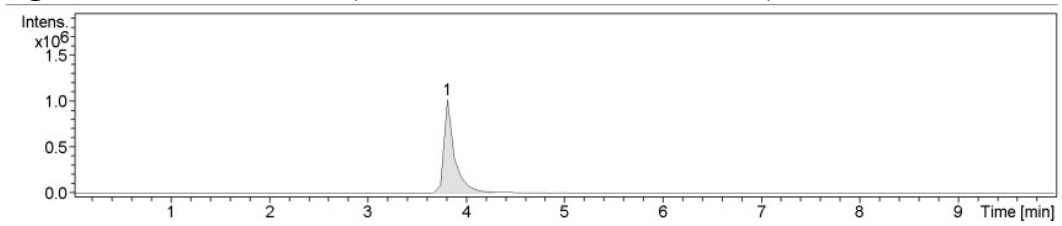
#	RT [min]	Area	S/N
1	3.66	3669290	169.2

Cmpd 1, 3.66 min



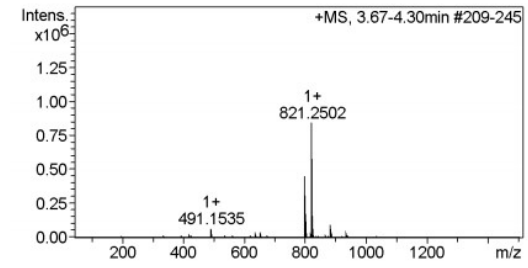
#	m/z	I	I %
1	203.0913	99202	18.1
2	435.1285	547151	100.0
3	436.1317	123091	22.5
4	597.1805	106846	19.5
5	637.2121	84623	15.5
6	679.5122	295253	54.0
7	680.5145	123651	22.6
8	701.4932	84987	15.5
9	821.2477	232792	42.5
10	822.2506	86107	15.7

Figure S28. LC-MS diagram of monopropyl ester formed after simulated digestion of TDP in vitro (HPLC retention time: 3.7 min)



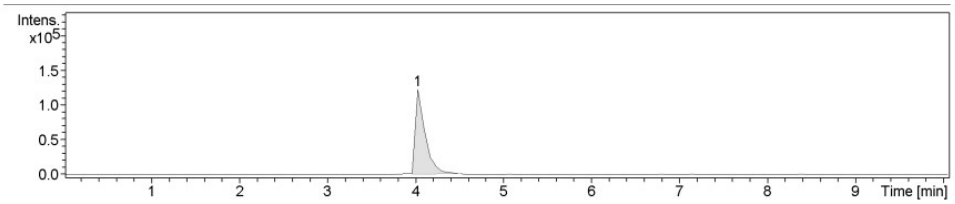
#	RT [min]	Area	S/N
1	3.81	7392777	442.0

Cmpd 1, 3.81 min



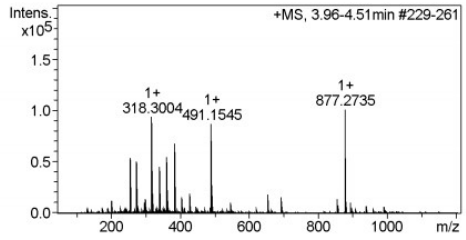
#	m/z	I	I %
1	491.1535	67682	8.0
2	653.2072	36540	4.3
3	799.2675	448565	53.3
4	800.2704	177607	21.1
5	801.2728	49951	5.9
6	821.2502	842273	100.0
7	822.2528	341236	40.5
8	823.2530	98469	11.7
9	883.2196	98099	11.6
10	884.2227	41282	4.9

Figure S29. LC-MS diagram of monopropyl ester formed after simulated digestion of TDP in vitro (HPLC retention time: 4.5 min)



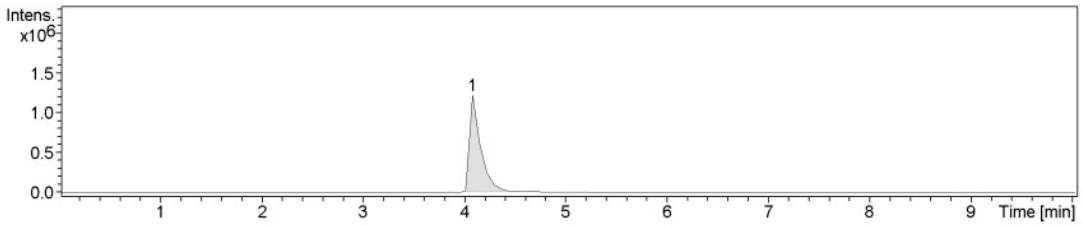
#	RT [min]	Area	S/N
1	4.03	931508	87.6

Cmpd 1, 4.03 min



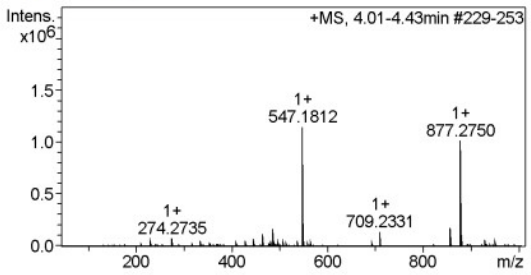
#	m/z	I	I %
1	256.2634	54043	53.6
2	274.2739	50867	50.4
3	318.3004	93746	92.9
4	340.2818	44955	44.5
5	362.3265	54403	53.9
6	384.3087	67729	67.1
7	491.1545	87044	86.3
8	492.1574	21155	21.0
9	877.2735	100915	100.0
10	878.2773	41765	41.4

Figure S30. LC-MS diagram of dipropyl ester formed after simulated digestion of TTP in vitro (HPLC retention time: 9.7 min)



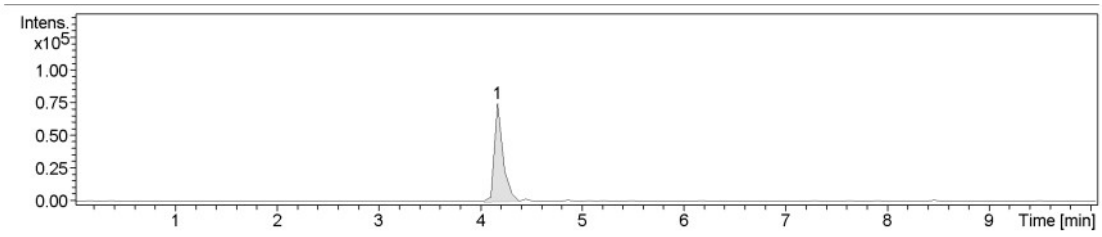
#	RT [min]	Area	S/N
1	4.08	8885964	415.2

Cmpd 1, 4.08 min



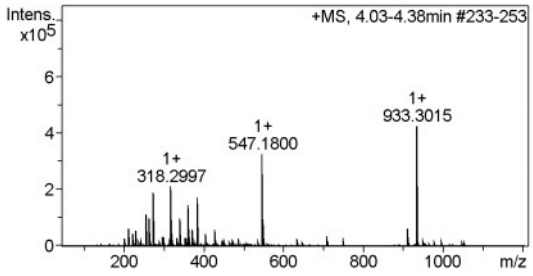
#	m/z	I	I %
1	274.2735	76242	6.7
2	464.2999	123900	10.8
3	486.2820	166626	14.6
4	547.1812	1143208	100.0
5	548.1839	319895	28.0
6	709.2331	144022	12.6
7	855.2906	177051	15.5
8	877.2750	1013450	88.6
9	878.2772	410216	35.9
10	879.2790	123626	10.8

Figure S31. LC-MS diagram of dipropyl ester formed after simulated digestion of TTP in vitro (HPLC retention time: 11.7 min)



#	RT [min]	Area	S/N
1	4.17	448163	26.8

Cmpd 1, 4.17 min



#	m/z	I	I %
1	256.2633	115114	27.2
2	263.0557	99154	23.4
3	274.2738	191696	45.2
4	318.2997	210385	49.7
5	340.2811	99803	23.6
6	362.3258	145652	34.4
7	384.3075	173353	40.9
8	547.1800	326097	77.0
9	933.3015	423718	100.0
10	934.3039	194189	45.8

Figure S32. LC-MS diagram of tripropyl ester formed after simulated digestion of TEP in vitro (HPLC retention time: 13.3 min)

Table S1. The chemical shift comparation of TX and its esters in ^{13}C-NMR

Carbon	TX	TMP	Chemical shift	TDP	Chemical shift	TTP	Chemical shift	TEP	Chemical shift	TDB	Chemical shift	TMH	Chemical shift	TMO	Chemical shift	TMD	Chemical shift
C-4	177.93	177.97	-0.04	177.65	0.28	177.84	0.09	177.84	0.09	177.87	0.06	177.97	-0.04	177.97	-0.04	177.66	0.27
C-7	165.11	164.47	0.64	164.4	0.71	164.47	0.64	164.51	0.6	164.46	0.65	164.5	0.61	164.51	0.6	164.41	0.7
C-9	161.29	161.38	-0.09	162.75	-1.46	161.93	0.64	162.2	-0.91	161.86	-0.57	161.39	-0.1	161.41	-0.12	162.72	-1.43
C-5	157.04	157.10	-0.06	157.18	-0.14	157	0.04	156.96	0.08	156.98	0.06	157.11	-0.07	157.11	-0.07	157.15	-0.11
C-2	156.9	156.84	0.06	156.41	0.49	156.58	0.32	156.33	0.57	156.75	0.15	156.86	0.04	156.86	0.04	156.57	0.33
C-4’	151.31	151.34	-0.03	150.69	0.62	150.93	0.38	150.94	0.37	150.77	0.54	151.35	-0.04	151.36	-0.05	151.21	0.1
C-3’	147.95	147.96	-0.01	148.08	-0.13	147.62	0.33	147.69	0.26	148.1	-0.15	147.98	-0.03	147.98	-0.03	147.93	0.02
C-3	134.12	134.15	-0.03	134.46	-0.34	134.46	-0.34	134.31	-0.19	134.34	-0.22	134.16	-0.04	134.16	-0.04	134.36	-0.24
C-1’	122.99	123.00	-0.01	123.51	-0.52	123.68	-0.69	123.57	-0.58	123.41	-0.42	123	-0.01	122.99	0	122.91	0.08
C-6’	122.82	122.78	0.04	122.89	-0.07	123.42	-0.6	123.41	-0.59	122.99	-0.17	122.79	0.03	122.79	0.03	122.86	-0.04
C-5’	114.8	114.75	0.05	115.13	-0.33	115.69	-0.89	115.62	-0.82	115.15	-0.35	114.76	0.04	114.76	0.04	114.7	0.1

C-2'	**113.19**	**113.18**	**0.01**	**113.75**	**-0.56**	**113.92**	**-0.73**	**113.99**	**-0.8**	**113.73**	**-0.54**	**113.2**	**-0.01**	**113.19**	**0**	**113.15**	**0.04**
C-10	**105.51**	**105.71**	**-0.2**	**106.31**	**-0.80**	**105.97**	**-0.46**	**105.89**	**-0.38**	**105.93**	**-0.42**	**105.72**	**-0.21**	**105.71**	**-0.2**	**106.22**	**-0.71**
C-1''	**101.76**	**101.75**	**0.01**	**101.84**	**-0.08**	**101.91**	**-0.15**	**101.72**	**0.04**	**101.8**	**-0.04**	**101.75**	**0.01**	**101.76**	**0**	**101.87**	**-0.11**
C-1'''	**101.39**	**101.40**	**-0.01**	**101.4**	**-0.01**	**101.35**	**0.04**	**101.29**	**0.1**	**101.39**	**0**	**101.39**	**0**	**101.38**	**0.01**	**101.39**	**0**
C-6	**98.83**	**98.76**	**0.07**	**99.03**	**-0.2**	**98.9**	**-0.07**	**98.88**	**-0.05**	**98.88**	**-0.05**	**98.79**	**0.04**	**98.77**	**0.06**	**98.98**	**-0.15**
C-8	**93.33**	**93.49**	**-0.16**	**92.62**	**0.71**	**93.16**	**0.17**	**92.98**	**0.35**	**93.15**	**0.18**	**93.48**	**-0.15**	**93.46**	**-0.13**	**92.59**	**0.74**
C-3''	**76.82**	**76.83**	**-0.01**	**76.92**	**-0.1**	**76.86**	**-0.04**	**76.89**	**-0.07**	**76.88**	**-0.06**	**76.83**	**-0.01**	**76.83**	**-0.01**	**76.94**	**-0.12**
C-5''	**76.4**	**76.42**	**-0.02**	**76.44**	**-0.04**	**76.43**	**-0.03**	**76.14**	**0.26**	**76.44**	**-0.04**	**76.41**	**-0.01**	**76.41**	**-0.01**	**76.42**	**-0.02**
C-2''	**74.61**	**74.61**	**0**	**74.62**	**-0.01**	**74.64**	**-0.03**	**73.9**	**0.71**	**74.62**	**-0.01**	**74.61**	**0**	**74.6**	**0.01**	**74.62**	**-0.01**
C-4'''	**72.2**	**72.20**	**0**	**72.24**	**-0.04**	**72.23**	**-0.03**	**74.68**	**-2.48**	**72.22**	**-0.02**	**72.2**	**0**	**72.21**	**-0.01**	**72.26**	**-0.06**
C-3'''	**71.05**	**71.06**	**-0.01**	**71.09**	**-0.04**	**71.1**	**-0.05**	**70.78**	**0.27**	**71.08**	**-0.03**	**71.06**	**-0.01**	**71.07**	**-0.02**	**71.1**	**-0.05**
C-2'''	**70.98**	**70.98**	**0**	**71.09**	**-0.11**	**70.78**	**0.2**	**70.78**	**0.2**	**71.08**	**-0.1**	**70.99**	**-0.01**	**70.99**	**-0.01**	**70.98**	**0**
C-4''	**70.94**	**70.79**	**0.15**	**70.59**	**0.35**	**70.62**	**0.32**	**70.42**	**0.52**	**70.6**	**0.34**	**70.6**	**0.34**	**70.58**	**0.36**	**70.49**	**0.45**
C-A'	**70.78**	**70.76**	**0.02**	**70.75**	**0.03**	**67.52**	**3.26**	**67.49**	**3.29**	**70.78**	**0**	**70.78**	**0**	**70.78**	**0**	**70.74**	**0.04**

C-A'	**70.76**	**70.73**	**0.03**	**67.25**	**3.51**	**67.26**	**3.5**	**67.3**	**3.46**	**67.23**	**3.53**	**70.77**	**-0.01**	**70.78**	**-0.02**	**70.74**	**0.02**
C-A	**70.64**	**67.17**	**3.47**	**67**	**3.64**	**67.1**	**3.54**	**67.13**	**3.51**	**67.12**	**3.52**	**67.17**	**3.47**	**67.19**	**3.45**	**67.04**	**3.6**
C-5'''	**68.74**	**68.73**	**0.01**	**68.73**	**0.01**	**68.72**	**0.02**	**66.29**	**2.45**	**68.73**	**0.01**	**68.73**	**0.01**	**68.72**	**0.02**	**68.72**	**0.02**
C-6''	**67.61**	**67.60**	**0.01**	**67.59**	**0.02**	**67.47**	**0.14**	**67.76**	**-0.15**	**67.58**	**0.03**	**67.56**	**0.05**	**67.52**	**0.09**	**67.45**	**0.16**
C-B'	**60**	**60.00**	**0**	**59.98**	**0.02**	**62.84**	**-2.84**	**62.8**	**-2.8**	**60**	**0**	**60**	**0**	**60**	**0**	**59.95**	**0.05**
C-B'	**59.96**	**59.96**	**0**	**62.86**	**-2.9**	**62.83**	**-2.87**	**62.8**	**-2.84**	**62.75**	**-2.79**	**59.95**	**0.01**	**59.95**	**0.01**	**59.91**	**0.05**
C-B	**59.76**	**62.56**	**-2.8**	**62.61**	**-2.85**	**62.58**	**-2.82**	**62.56**	**-2.8**	**62.47**	**-2.71**	**62.48**	**-2.72**	**62.45**	**-2.69**	**62.5**	**-2.74**
C-6'''	**18.16**	**18.15**	**0.01**	**18.18**	**-0.02**	**18.15**	**0.01**	**17.54**	**0.62**	**18.17**	**-0.01**	**18.15**	**0.01**	**18.14**	**0.02**	**18.18**	**-0.02**