

**Cholinesterase Inhibitory Arisugacins L-Q from a *Penicillium* sp. Isolate Obtained through a Citizen
Science Initiative and their Activities in a Phenotype-Based Zebrafish Assay**

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Table S1. Identification and Associated Information for the Fungal Isolate OK0263 PDA-20.

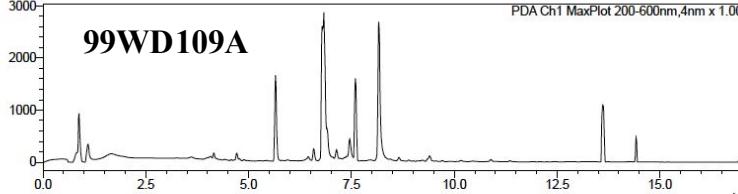
Fungus name used in this report:	<i>Penicillium</i> sp.
Full ID code for the sample from which the fungal isolated was obtained:	10263, soil
SHAREOK link for citizen-science-derived sample (if available):	https://shareok.org/handle/11244/300790
Internal lab sample ID code:	OK0263
Full fungal isolate ID code:	OK0263 PDA-20
Plate number and well for fungal initial extract:	87-B1
First chemistry notebook code assigned to fungal extract and PDA chromatogram of the crude extract:	 LCMS condition: MeCN-H ₂ O gradient containing 0.1% HCOOH (10:90-100:0 in 15 min)
Pictures of fungal isolate: picture shows the fungus growing on Cheerios.	
ITS sequence used to assign taxonomy:	GCGGGTGACAAAGCCCCATACGCTCGAGGACCGGACGCG GTGCCGCCGCTGCCTTCGGGCCGTCGGGGAAATCGG AGGACGGGGCCAACACACAAGCCGGCTTGAGGGCAG CAATGACGCTCGGACAGGCATGCCCGGAATACCAGG GGCGCAATGTGCGTTCAAAGACTCGATGATTCACTGAA TTTGCATTACATACGTATCGCATTGCTGCGTTCTTC ATCGATGCCGAACCAAGAGATCCGTTGAAAGTTT AAATAATTATATTTCACTCAGACTCAATCTTCAGACA GAGTCGAGGGTGTCTCGCGGGCGCGGGCCGGGGC GTGAGCCCCCGGCCAGTTAAGGCGGGCCGCCGAA GCAACAAGGTAAAATAAACACGGGTGCGGAGGTTGGACC CAGAGGGCCCTCACTCGTAATGATCCTCCGAGGTCA CCTACGGAAACCTTGTACGACTTTACTTCC
GenBank accession number:	MK722362
Notes on the taxonomic affiliation of the fungal isolate:	Multiple <i>Penicillium</i> species all at 98.90% identity, 100% query cover, cannot assign species level identification, BLAST date April 1 2019

Table S2. Dose curve analysis of toxic crude fungal extracts.

Sample ID	Phenotype Observed ^a	Dose, µg/mL ^b
P86-B12	Edema	0.1
P86-C4	Missed tail	1
P86-C6	Missed tail, necrosis	2
P86-D7	Missed tail, necrosis	1
P86-D10	Edema, necrosis, enlarged hindbrain	0.2
P86-E9	Hemorrhage	0.2
P86-F1	Edema, misspelled tail, necrosis, lack of blood circulation	0.1
P87-A3	Edema	1
P87-A5	Missed tail	2
P87-A7	Growth arrest, necrosis	1
P87-A8	N	-
P87-A9	Necrosis	0.1
P87-A11	N	-
P87-A12	N	-
P87-B2	N	-
P87-B5	N	-
P87-B8	Missed tail and body	1
P87-B10	N	-
P87-B12	N	-
P87-C2	N	-
P87-C8	N	-
P87-C9	Hemorrhage	0.1
P87-C10	N	-
P87-D8	N	-
P87-E8	N	-
P87-F12	N	-
P87-H6	Hemorrhage	2
P87-H8	N	-
GL6-A8	N	-
GL6-B8	N	-
GL6-B12	N	-
GL6-F9	Small embryo	10
GL6-F10	Loss of pigmentation, impaired motility	2
GL7-H8	N	-

^aN – no phenotype observed at all concentrations tested.^bDose when phenotype was initially observed.

Table S3. ROESY correlations of compound 4.

Proton	ROESY correlations
H-12 (δ_H 2.71)	H-25 (δ_H 1.44), H-26 (δ_H 1.19)
H-12 (δ_H 3.14)	OH-13 (δ_H 6.72), H-19 (δ_H 4.91)
OH-13 (δ_H 6.72)	H-12 (δ_H 3.14), H-15 (δ_H 2.38), H-19 (δ_H 4.91), H-23 (δ_H 1.10)
H-15 (δ_H 2.38)	OH-13 (δ_H 6.72), OH-17 (δ_H 6.81)
H-16 (δ_H 1.73)	H-23 (δ_H 1.10)
H-16 (δ_H 1.94)	H-24 (δ_H 1.04), H-25 (δ_H 1.44), H-26 (δ_H 1.19)
OH-17 (δ_H 6.81)	H-15 (δ_H 2.38), H-19 (δ_H 4.91), H-23 (δ_H 1.10)
H-19 (δ_H 4.91)	H-12 (δ_H 3.14), OH-13 (δ_H 6.72), OH-17 (δ_H 6.81), OH-21(δ_H 5.44)
OH-19 (δ_H 3.63)	H-12 (δ_H 3.14), H-26 (δ_H 1.19)
H-20 (δ_H 2.16)	H-24 (δ_H 1.04), H-26 (δ_H 1.19), OH-19 (δ_H 3.63)
H-20 (δ_H 2.05)	OH-19 (δ_H 3.63)
H-21 (δ_H 3.66)	H-23 (δ_H 1.10), H-24 (δ_H 1.04),H-20 (δ_H 2.05, δ_H 2.16)
OH-21 (δ_H 5.44)	H-23 (δ_H 1.10), H-19 (δ_H 4.91)

Table S4. ROESY correlations of compound **5**.

Proton	ROESY correlations
H-12 (δ_{H} 2.19)	H-19 (δ_{H} 1.14), OH-13 (δ_{H} 6.66)
H-12 (δ_{H} 2.60)	H-25 (δ_{H} 1.34), H-26 (δ_{H} 1.07)
OH-13 (δ_{H} 6.66)	H-12 (δ_{H} 2.19), H-15 (δ_{H} 2.28), H-19 (δ_{H} 2.11), H-23 (δ_{H} 0.91)
H-15 (δ_{H} 1.57)	H-25 (δ_{H} 1.37)
H-15 (δ_{H} 2.28)	OH-13 (δ_{H} 6.66), OH-17 (δ_{H} 6.08)
H-16 (δ_{H} 1.61)	H-23 (δ_{H} 0.91)
H-16 (δ_{H} 1.89)	H-24 (δ_{H} 0.84), H-25 (δ_{H} 1.34), H-26 (δ_{H} 1.07)
OH-17 (δ_{H} 6.08)	H-15 (δ_{H} 2.28), H-16 (δ_{H} 1.61), H-19 (δ_{H} 2.11), H-23 (δ_{H} 0.91)
H-19 (δ_{H} 1.14)	H-26 (δ_{H} 1.07)
H-19 (δ_{H} 2.11)	H-21 (δ_{H} 3.72), OH-17 (δ_{H} 6.08)
H-21 (δ_{H} 3.72)	H-19 (δ_{H} 2.11), H-23 (δ_{H} 0.91)
OH-21 (δ_{H} 4.19)	H-23 (δ_{H} 0.91)

Table S5. ROESY correlations of compound **8**.

Proton	ROESY correlations
H-12 (δ_{H} 2.65)	H-25 (δ_{H} 1.38), H-26 (δ_{H} 1.10)
H-12 (δ_{H} 2.95)	H-19 (δ_{H} 4.74)
OH-13 (δ_{H} 6.63)	H-19 (δ_{H} 4.74)
H-15 (δ_{H} 1.63)	H-24 (0.96), H-25 (δ_{H} 1.38)
H-15 (δ_{H} 2.23)	OH-17 (δ_{H} 6.46)
H-16 (δ_{H} 1.63)	H-23 (δ_{H} 1.10)
H-16 (δ_{H} 1.96)	H-25 (δ_{H} 1.38), H-26 (δ_{H} 1.10)
OH-17 (δ_{H} 6.46)	H-15 (δ_{H} 2.23), H-19 (δ_{H} 4.74)
H-19 (δ_{H} 4.74)	H-23 (δ_{H} 1.10), H-12 (δ_{H} 2.95), OH-13 (δ_{H} 6.63), OH-17 (δ_{H} 6.46)
OH-19 (δ_{H} 5.05)	H-26 (δ_{H} 1.10)
H-20 (δ_{H} 2.54)	H-23 (δ_{H} 1.10)
H-20 (δ_{H} 2.74)	H-24 (δ_{H} 0.91)

Table S6. ROESY correlations of compound **12**.

Proton	ROESY correlations
H-12 (δ_{H} 2.72)	H-25 (δ_{H} 1.37), H-26 (δ_{H} 1.24)
H-12 (δ_{H} 3.39)	OH-13 (δ_{H} 6.44)
OH-13 (δ_{H} 6.44)	H-12 (δ_{H} 3.39), H-15 (δ_{H} 2.22)
H-15 (δ_{H} 1.60)	H-25 (δ_{H} 1.37)
H-15 (δ_{H} 2.22)	OH-13 (δ_{H} 6.44), OH-17 (δ_{H} 6.73)
H-16 (δ_{H} 1.74)	H-23 (δ_{H} 0.94), H-24 (δ_{H} 1.11), OH-17 (δ_{H} 6.73)
H-16 (δ_{H} 1.92)	H-24 (δ_{H} 1.11), H-25 (δ_{H} 1.37), H-26 (δ_{H} 1.24)
OH-17 (δ_{H} 6.73)	H-15 (δ_{H} 2.22), H-16 (δ_{H} 1.74), H-23 (δ_{H} 0.94)
H-20 (δ_{H} 2.65)	H-21 (δ_{H} 3.94), H-24 (δ_{H} 1.11), H-26 (δ_{H} 1.24)
H-20 (δ_{H} 2.79)	H-23 (δ_{H} 0.94), H-23 (δ_{H} 0.94),
H-21 (δ_{H} 3.94)	H-23 (δ_{H} 0.94), H-24 (δ_{H} 1.11), H-26 (δ_{H} 1.24)
OH-21 (δ_{H} 5.10)	H-23 (δ_{H} 0.94)

Table S7. ROESY correlations of compound **13**.

Proton	ROESY correlations
H-12 (δ_{H} 2.31)	H-25 (δ_{H} 1.30), H-26 (δ_{H} 1.10)
H-12 (δ_{H} 2.67)	H-13 (δ_{H} 1.78)
H-13 (δ_{H} 1.78)	H-12 (δ_{H} 2.67)
H-15 (δ_{H} 2.11)	H-25 (δ_{H} 1.30)
H-16 (δ_{H} 1.62)	H-24 (δ_{H} 1.02), H-25 (δ_{H} 1.30), H-26 (δ_{H} 1.10)
H-16 (δ_{H} 1.74)	H-23 (δ_{H} 1.09)
H-17 (δ_{H} 1.86)	H-23 (δ_{H} 1.09)

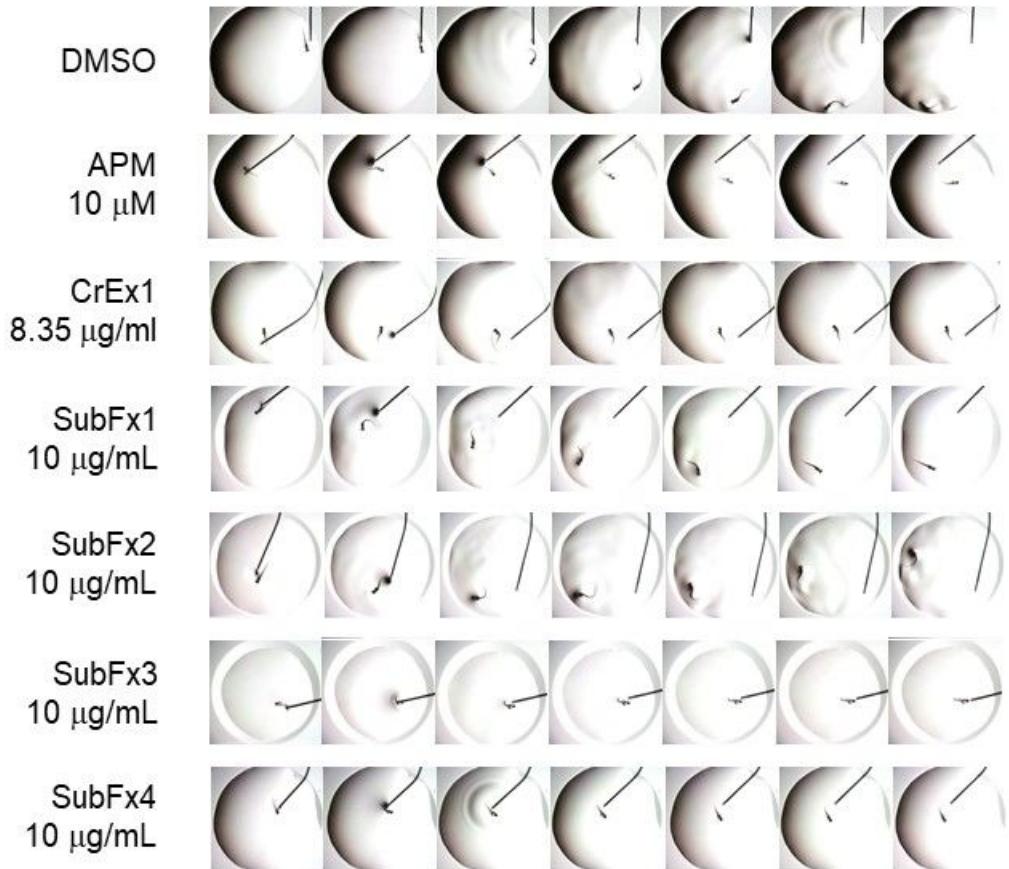


Figure S1. Zebrafish phenotype screen reveals fungal extracts that impaired motility. Frame-by-frame images representing response of zebrafish embryos to external stimulus (represented by black line).

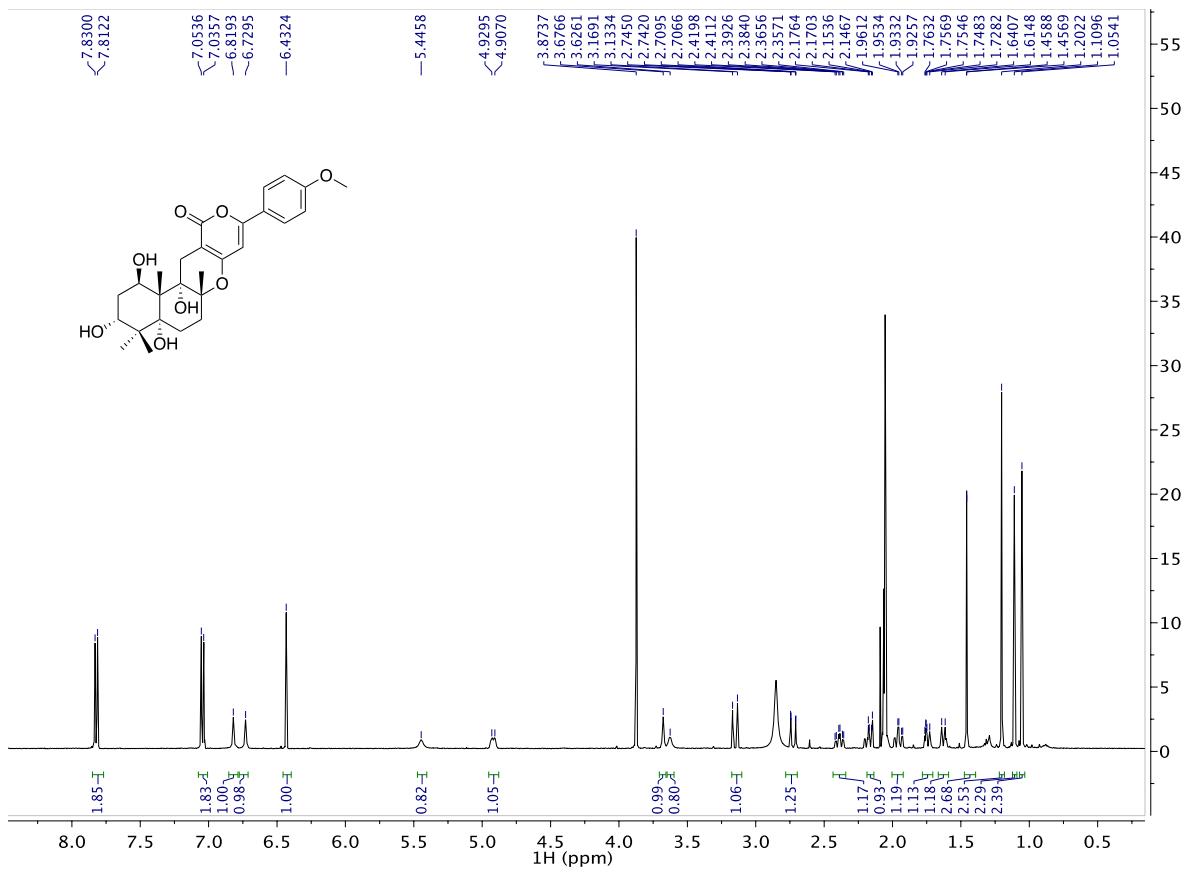


Figure S2. ¹H-NMR spectrum of compound 4 in acetone-*d*₆ (500 MHz).

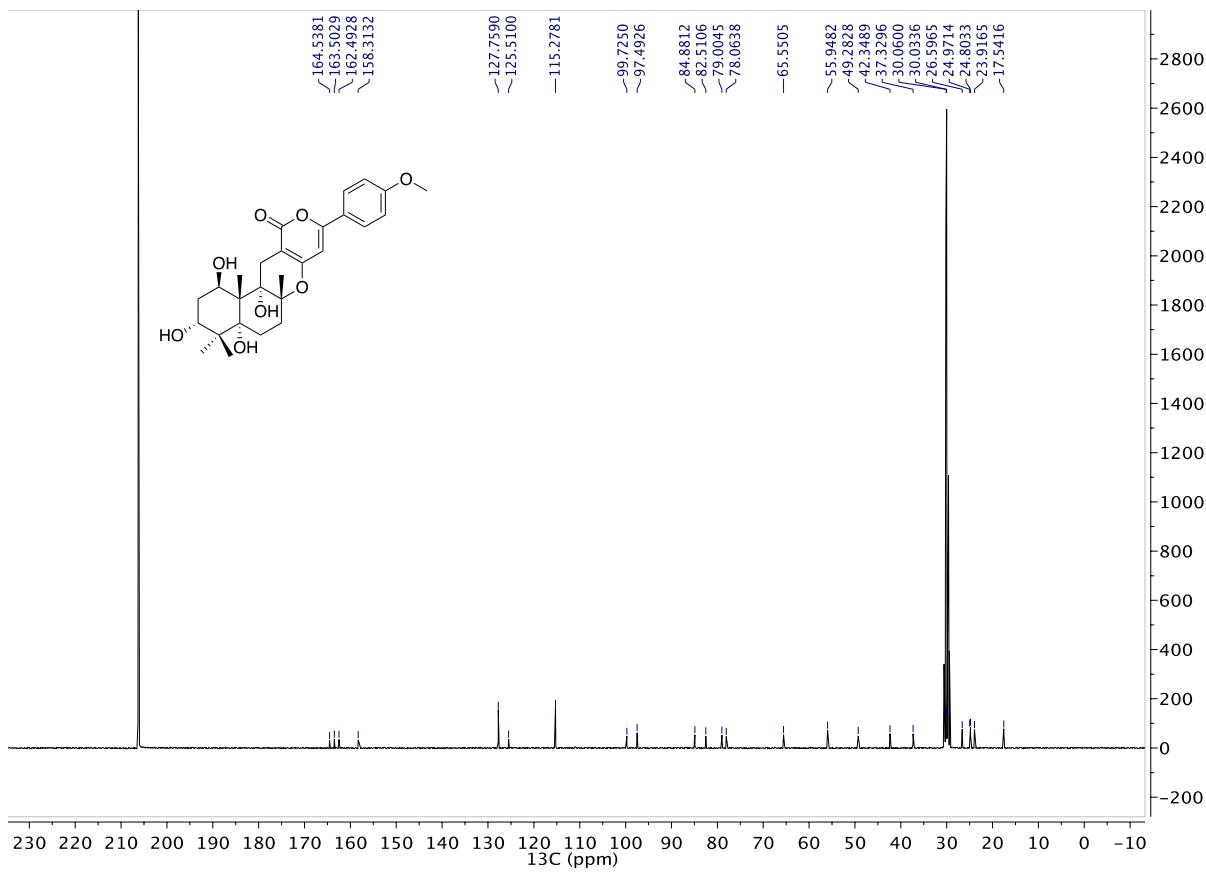


Figure S3. ^{13}C -NMR spectrum of compound 4 in acetone- d_6 (100 MHz).

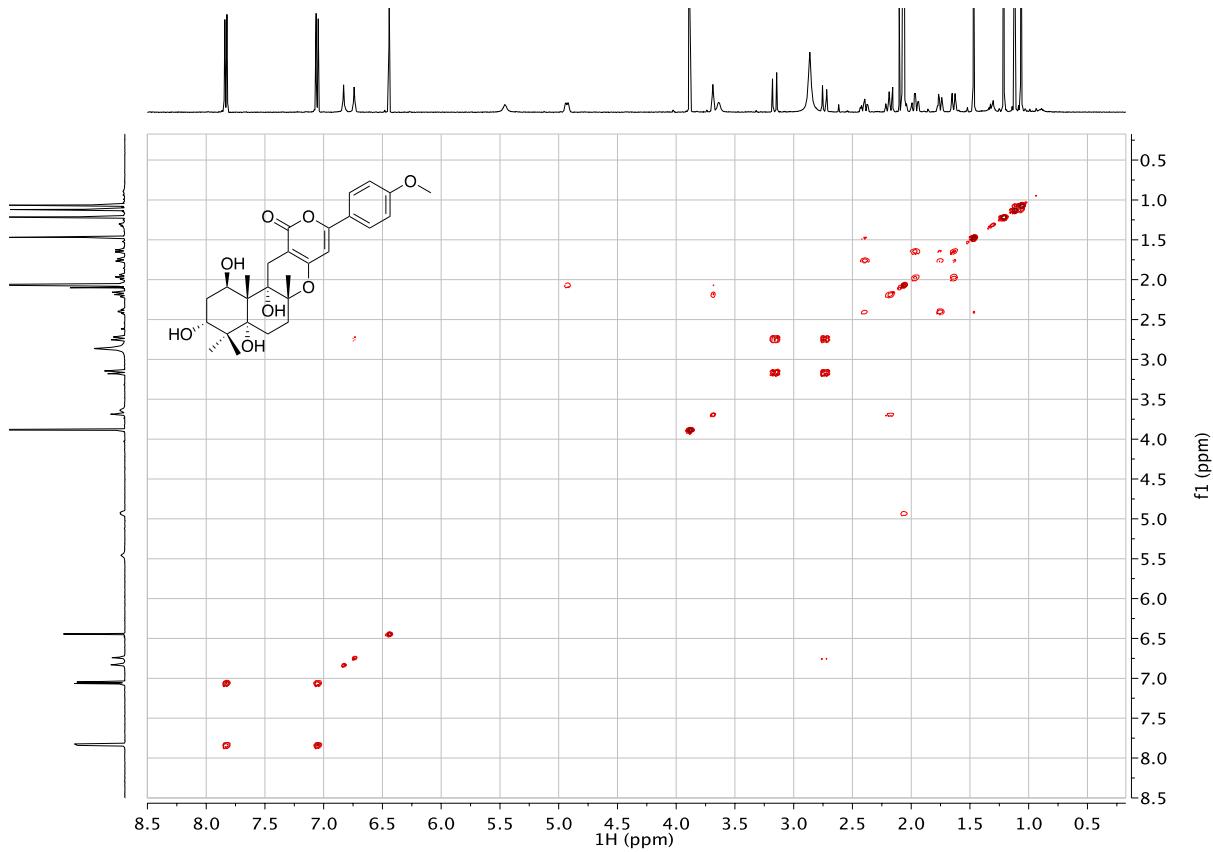


Figure S4. ^1H - ^1H COSY spectrum of compound 4 in acetone- d_6 (500 MHz).

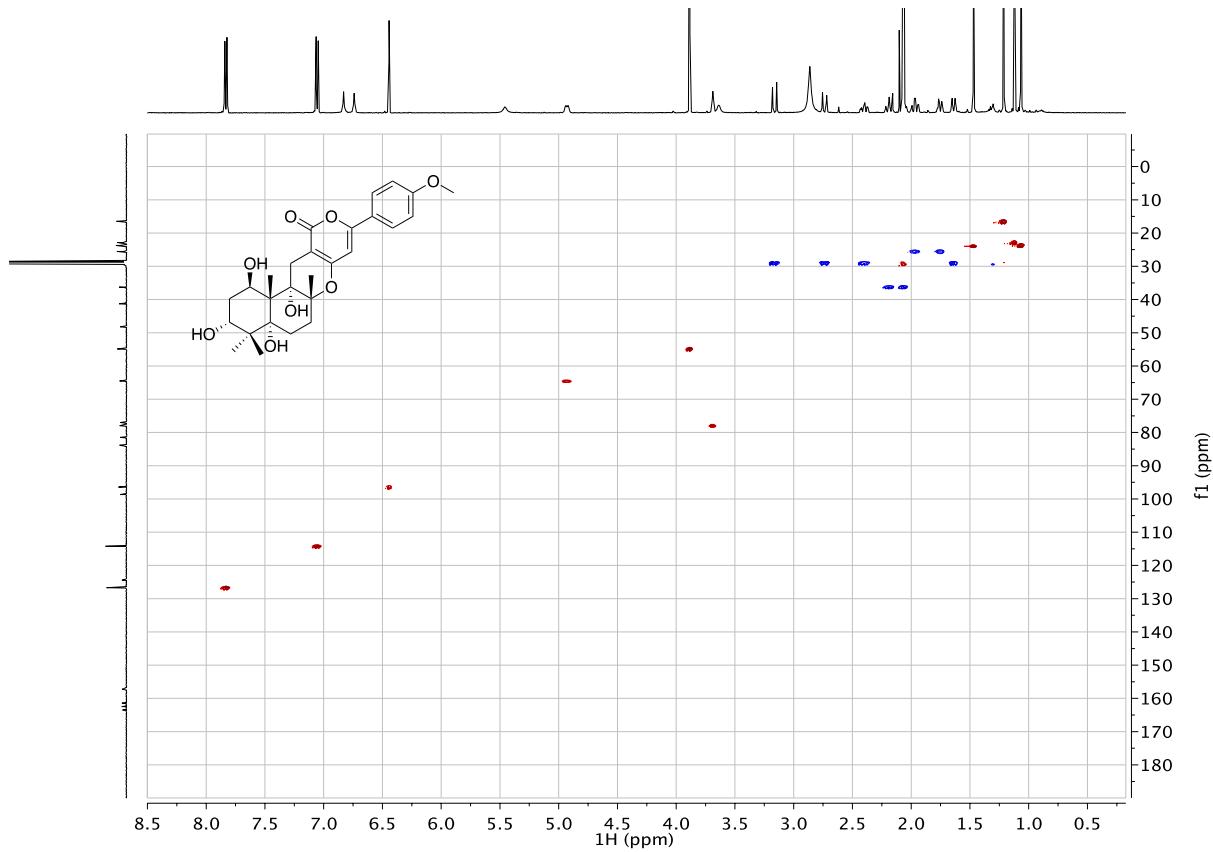


Figure S5. HSQC spectrum of compound 4 in acetone-*d*₆ (500 MHz).

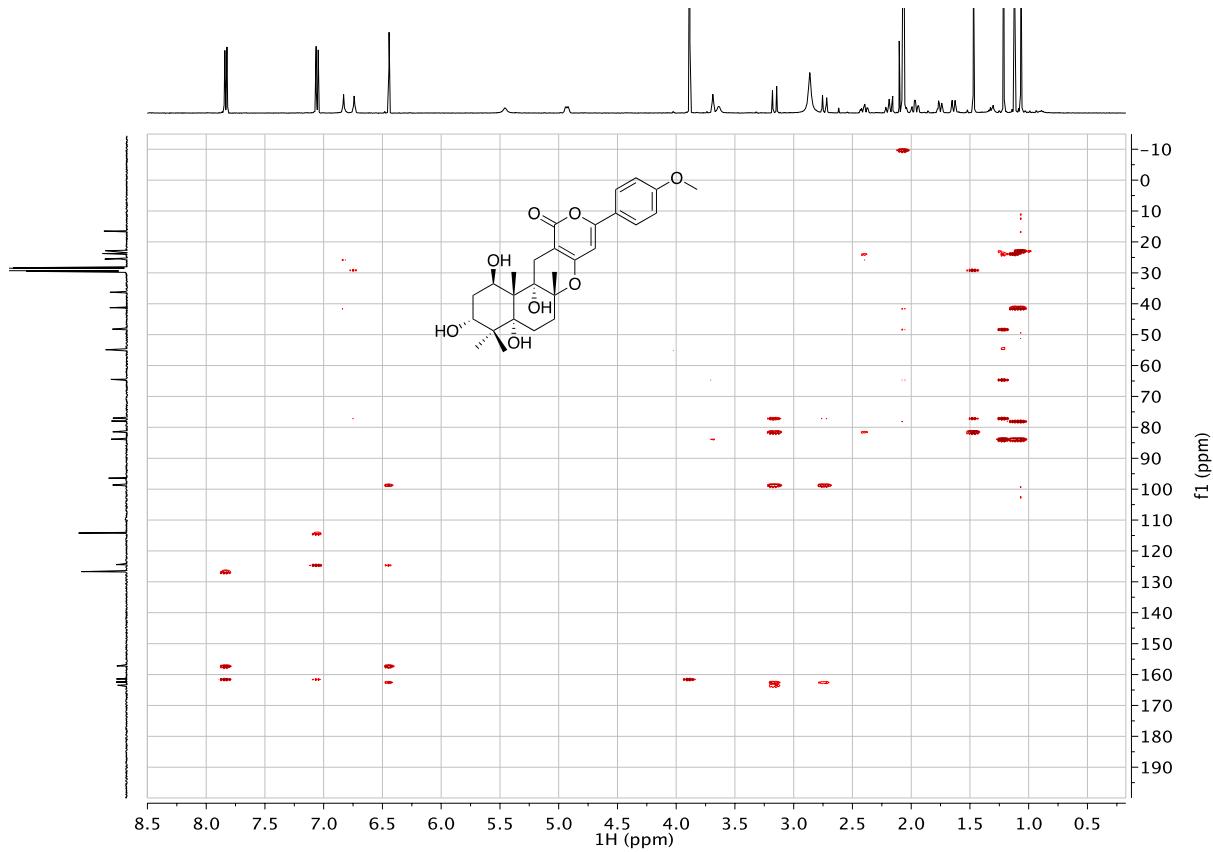


Figure 6. HMBC spectrum of compound **4** in acetone- d_6 (500 MHz).

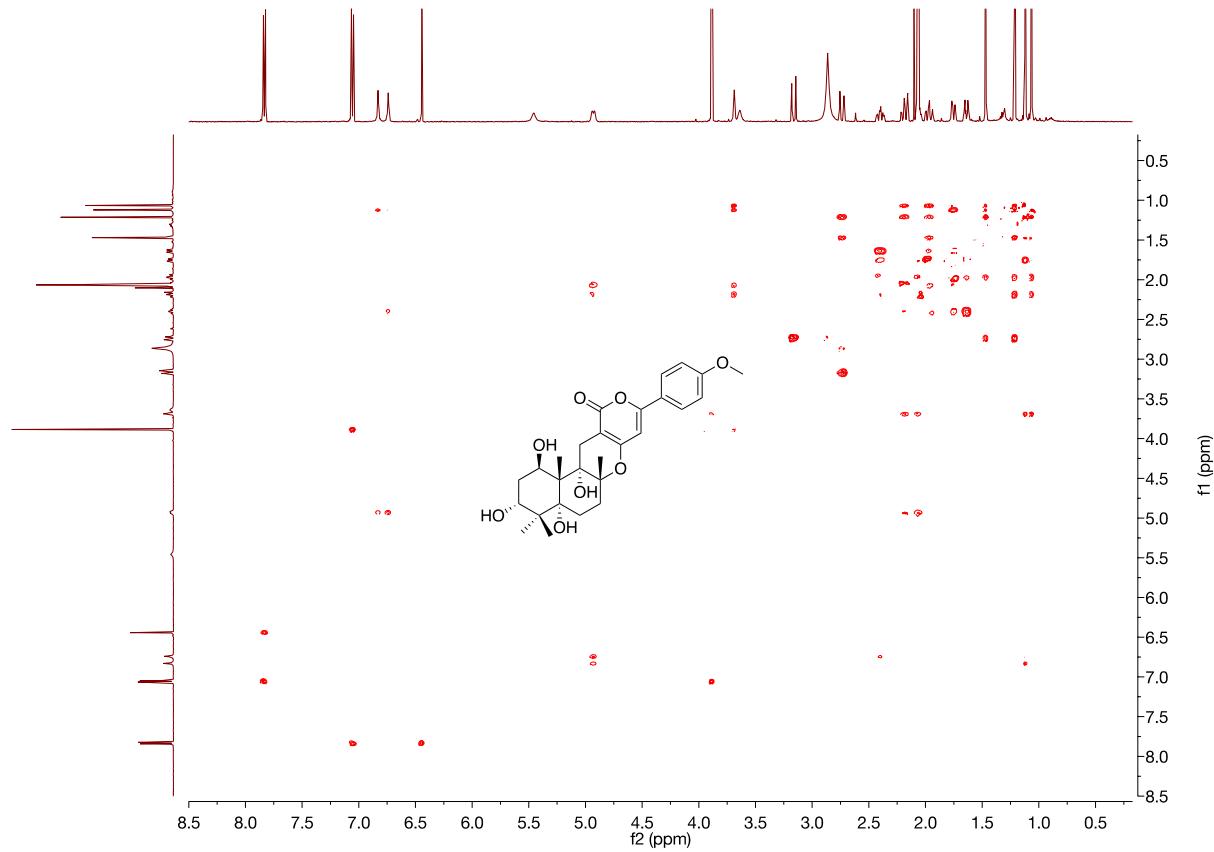


Figure S7. ROESY spectrum of compound **4** in acetone- d_6 (500 MHz).

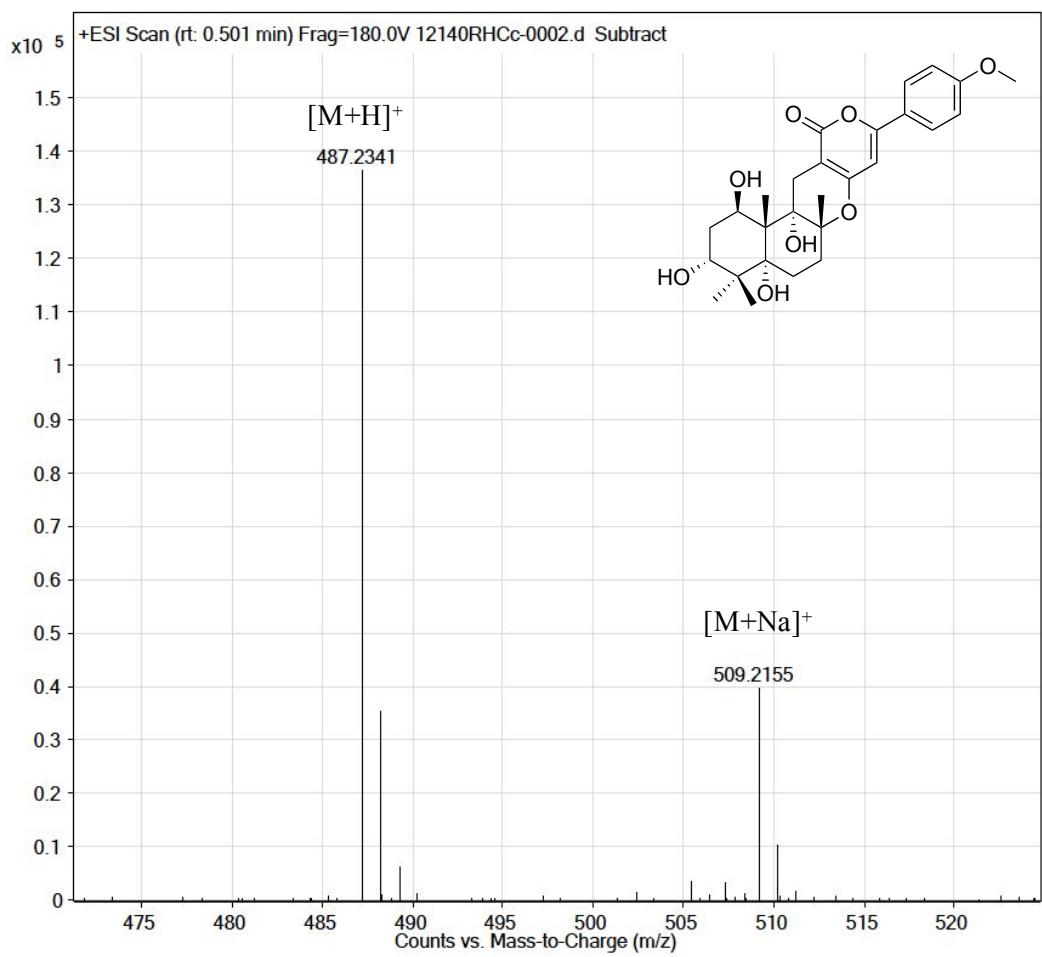


Figure S8. HRESIMS data for compound 4.

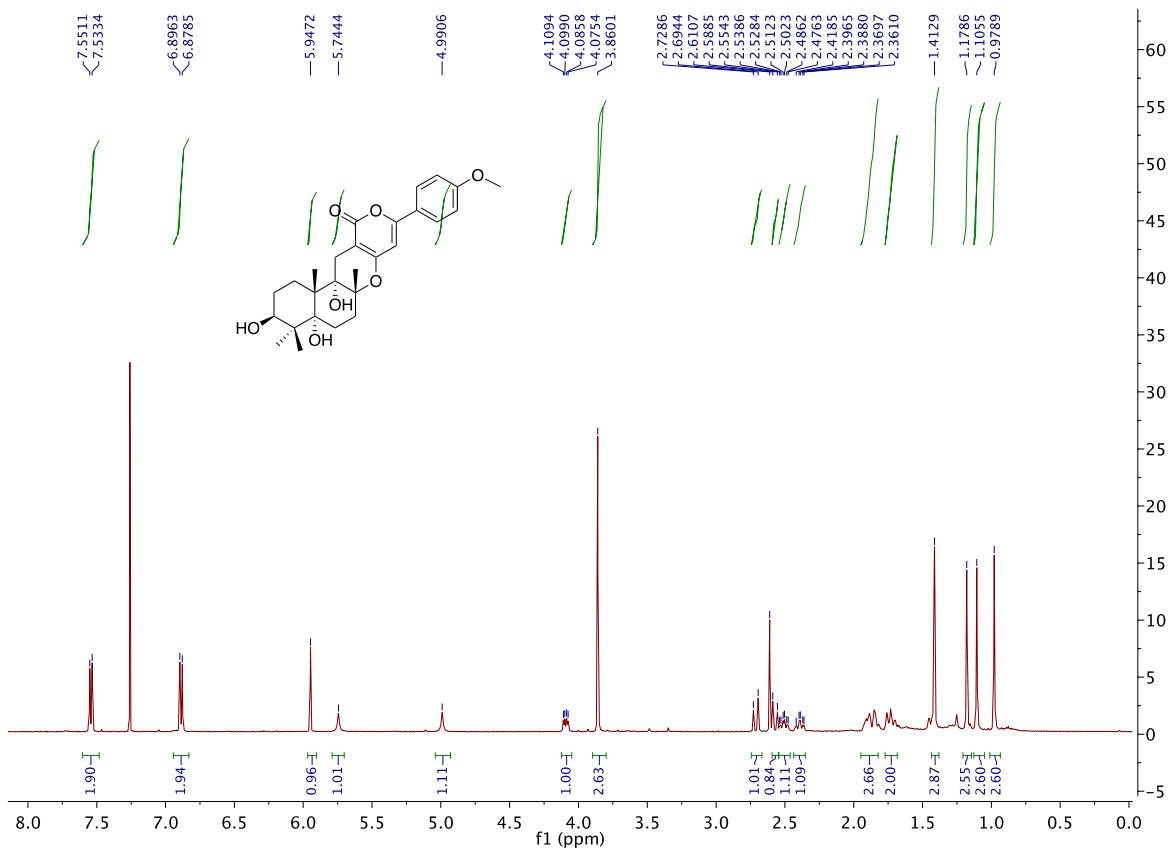


Figure S9. ^1H -NMR spectrum of compound **5** in CDCl_3 (500 MHz).

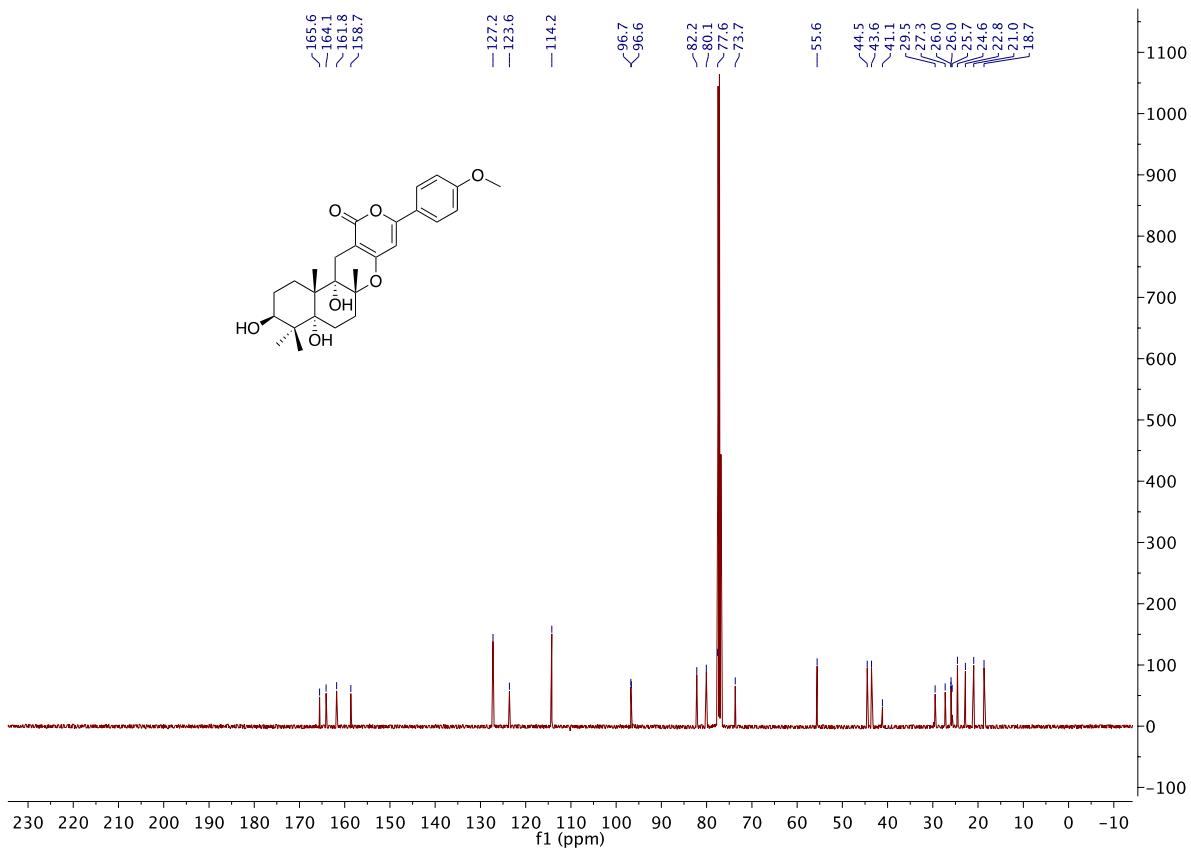


Figure S10. ^{13}C -NMR spectrum of compound 5 in CDCl_3 (500 MHz).

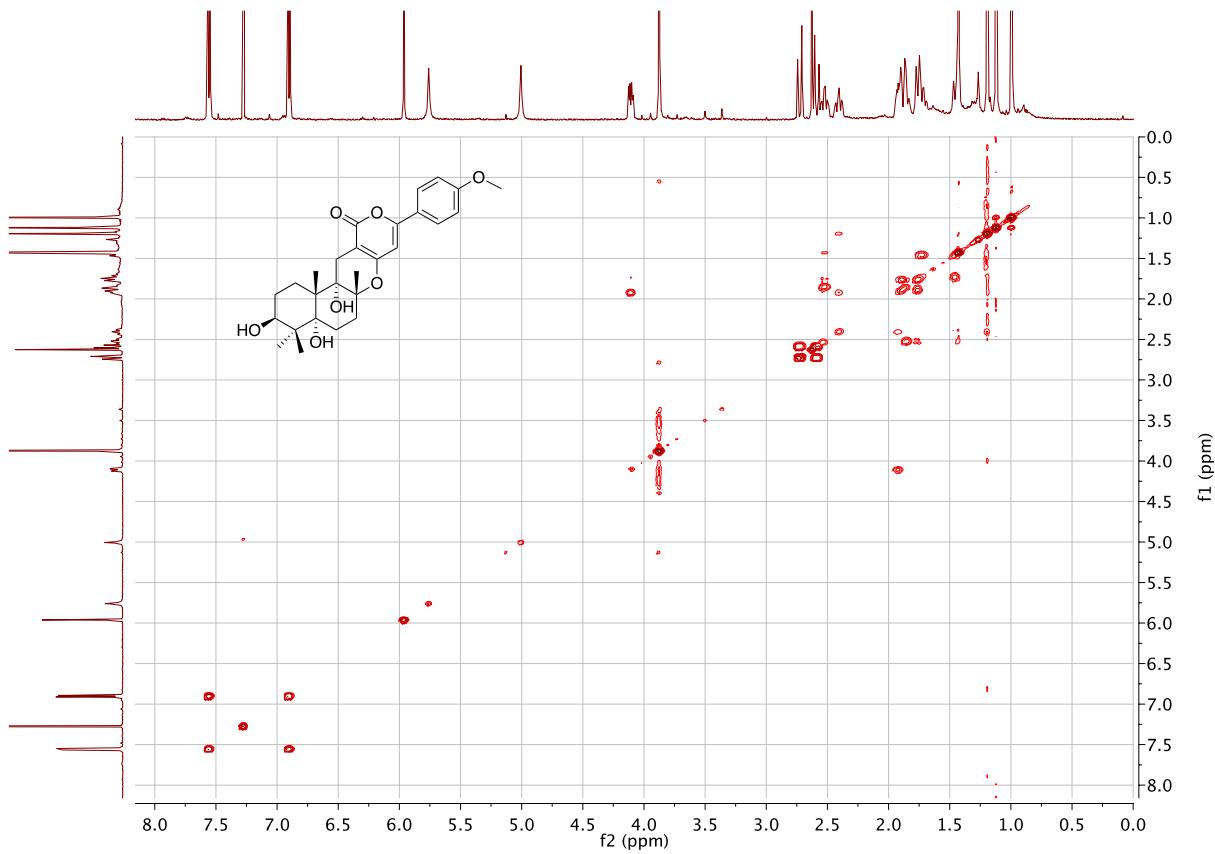


Figure S11. ^1H - ^1H COSY spectrum of compound **5** in CDCl_3 (500 MHz).

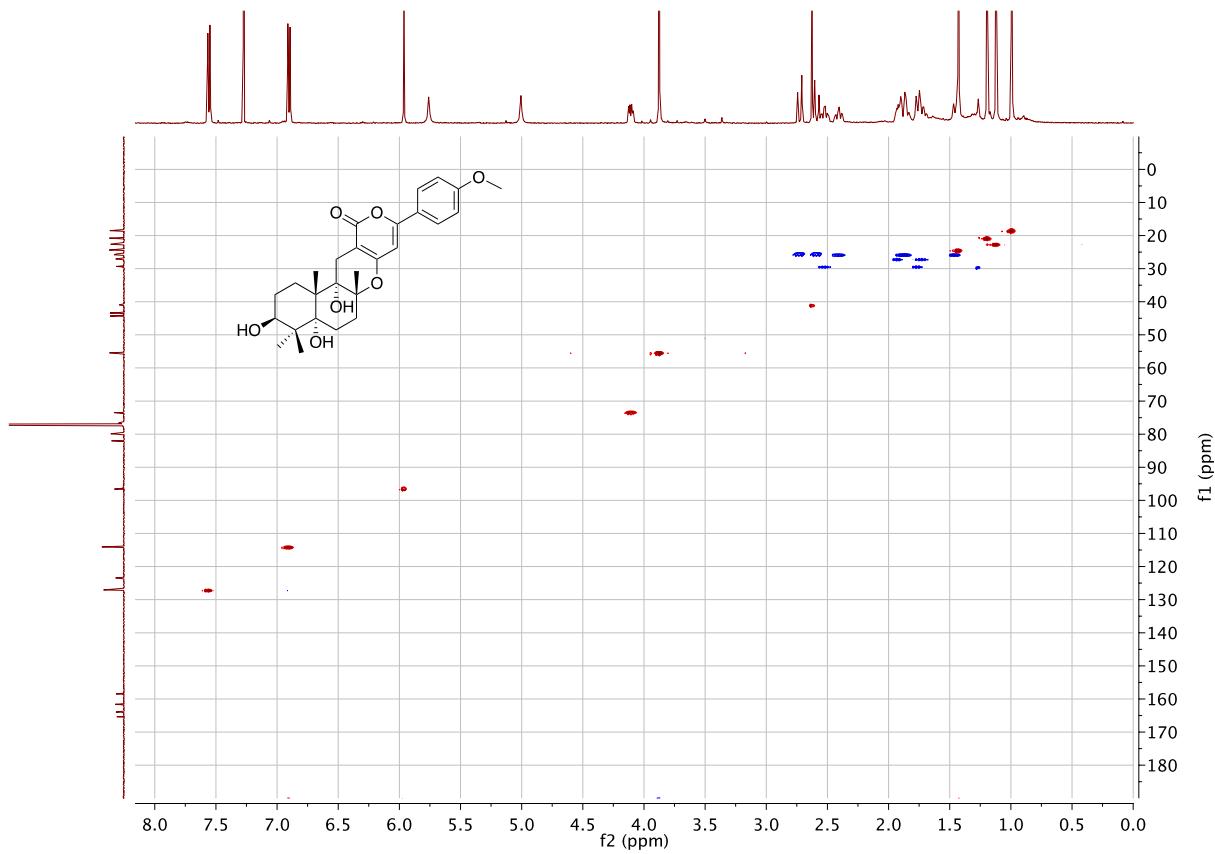


Figure S12. HSQC spectrum of compound **5** in CDCl_3 (500 MHz).

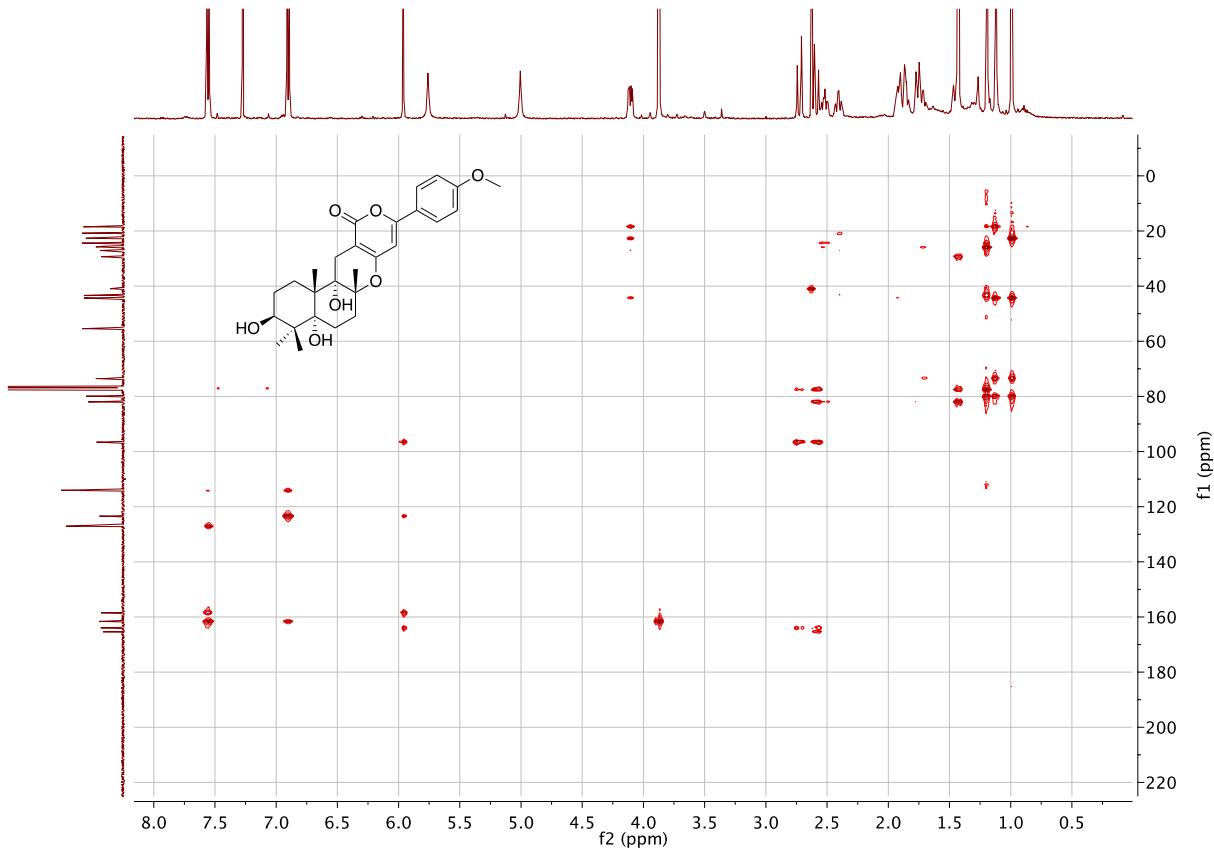


Figure S13. HMBC spectrum of compound **5** in CDCl_3 (500 MHz).

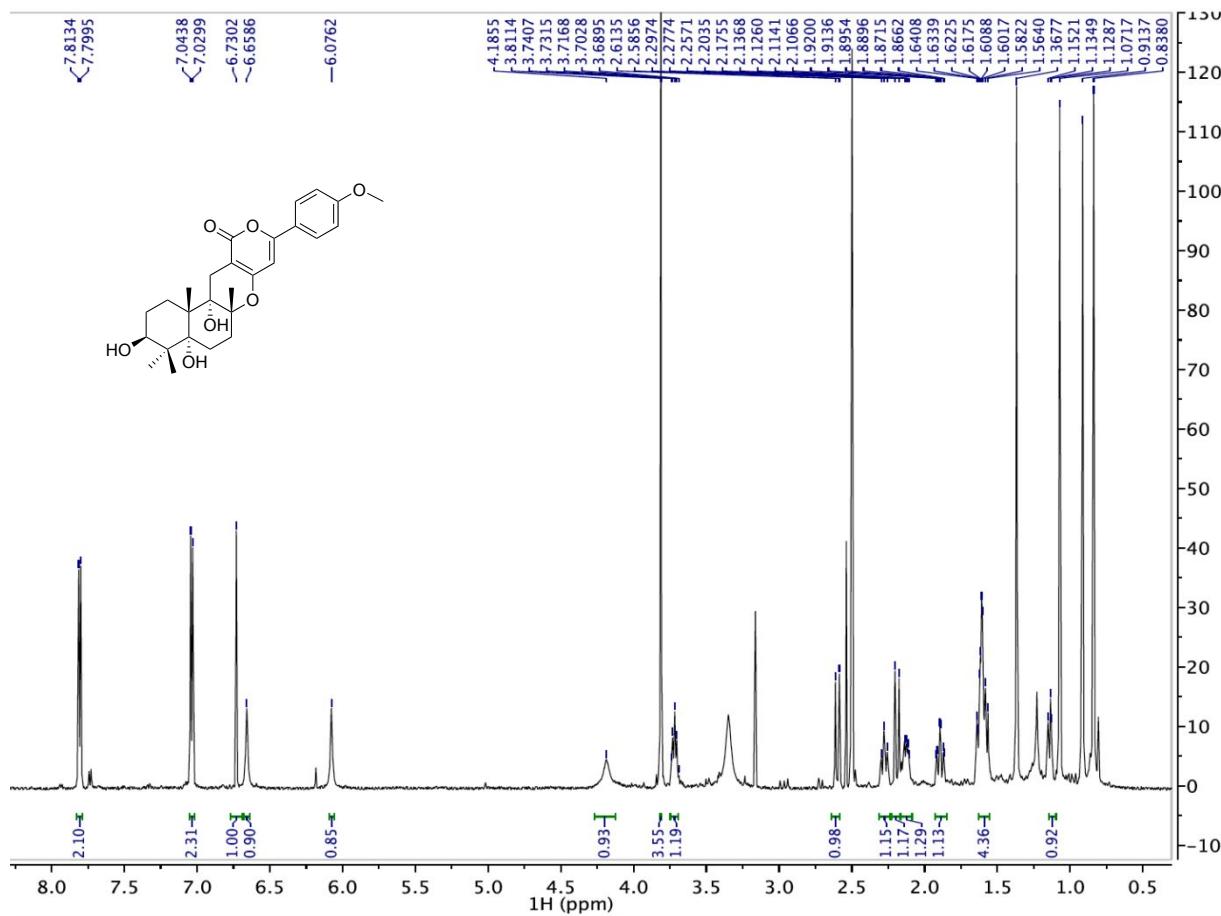


Figure S14. ^1H -NMR spectrum of compound 5 in $\text{DMSO}-d_6$ (600 MHz).

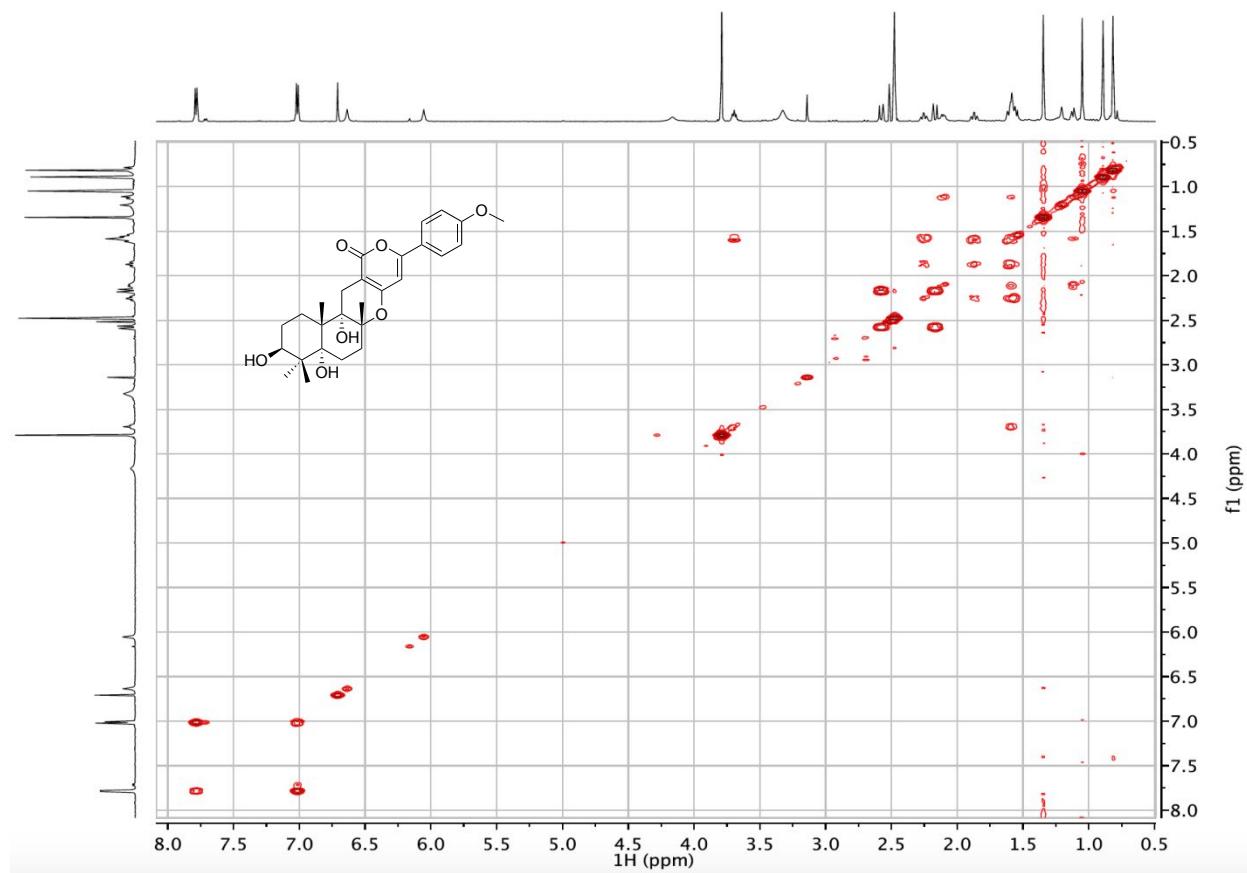


Figure S15. ^1H - ^1H COSY spectrum of compound 5 in $\text{DMSO}-d_6$ (600 MHz).

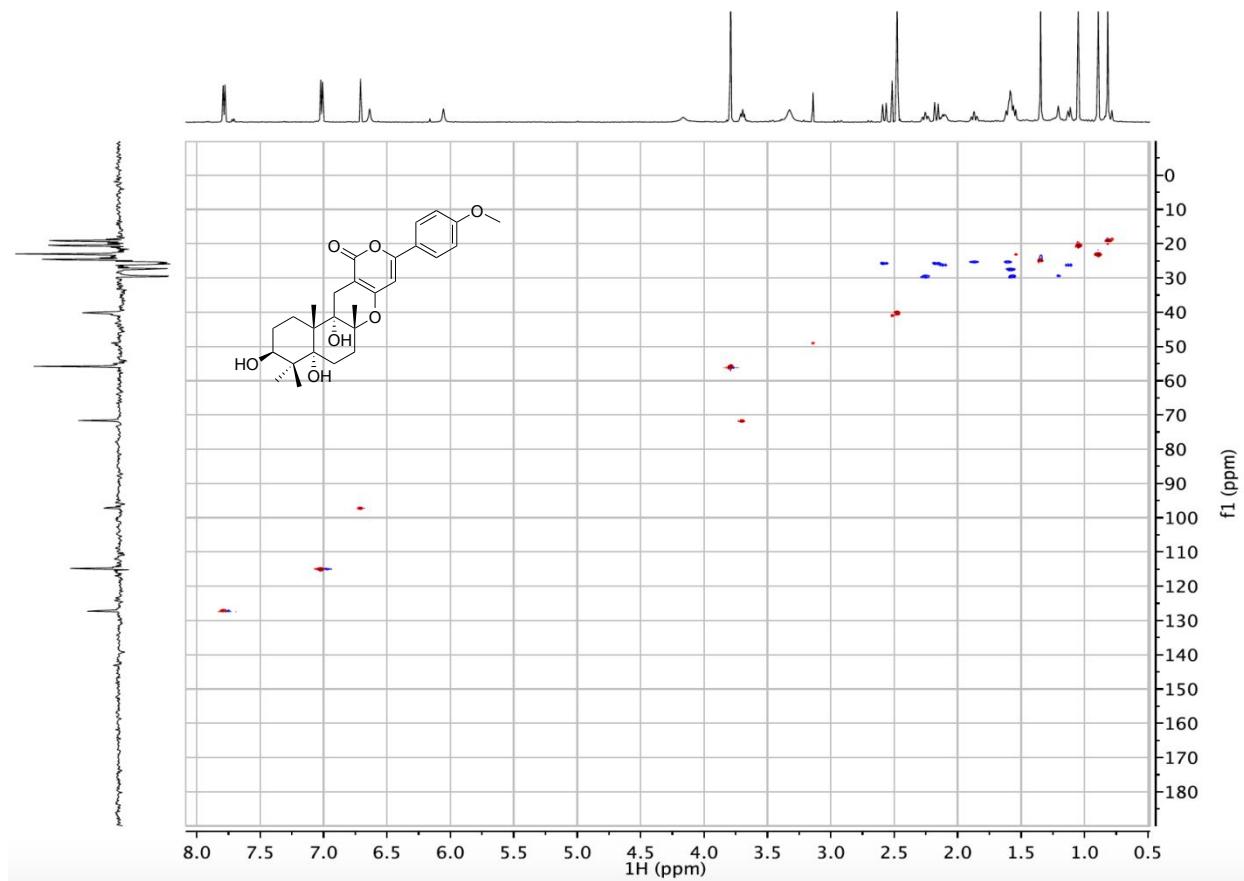


Figure S16. HSQC spectrum of compound **5** in $\text{DMSO}-d_6$ (600 MHz).

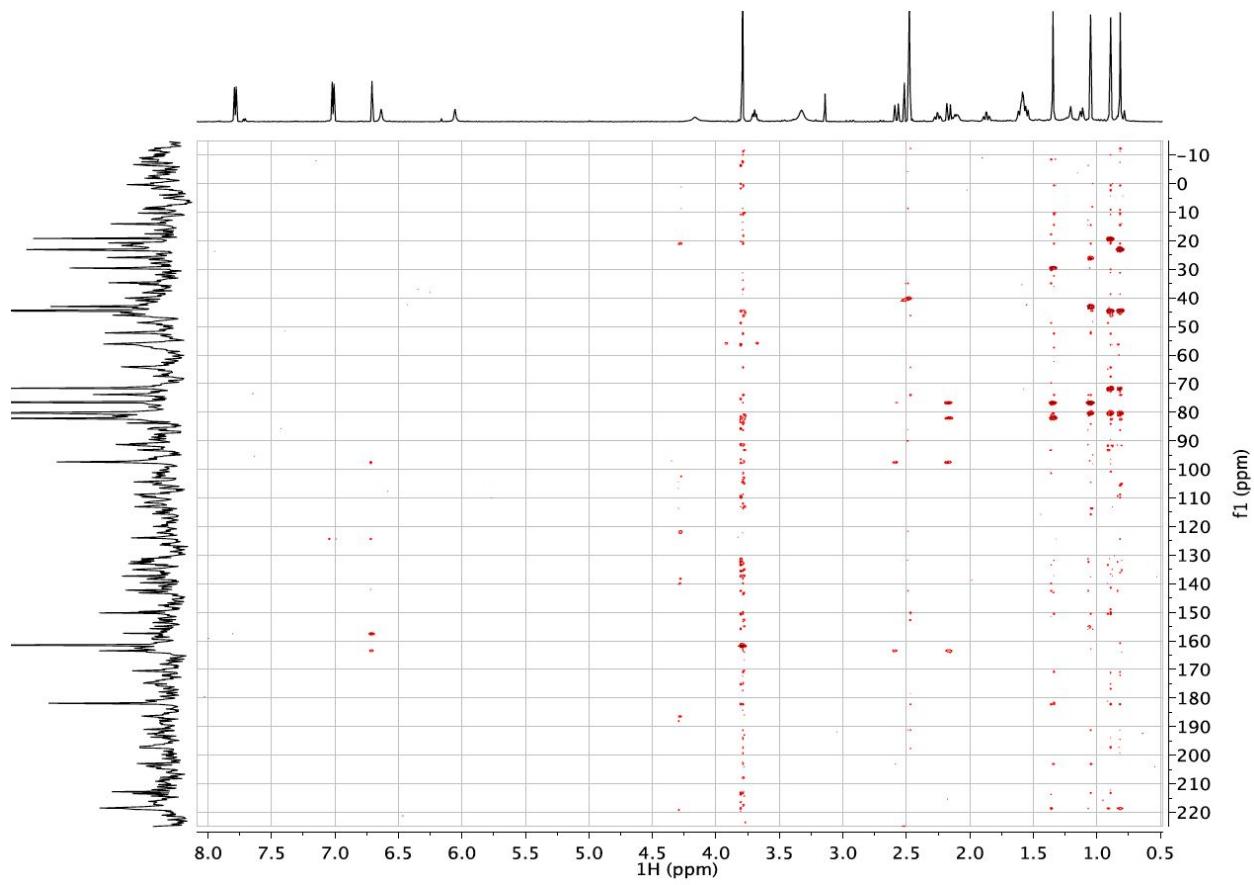


Figure S17. HMBC spectrum of compound **5** in $\text{DMSO}-d_6$ (600 MHz).

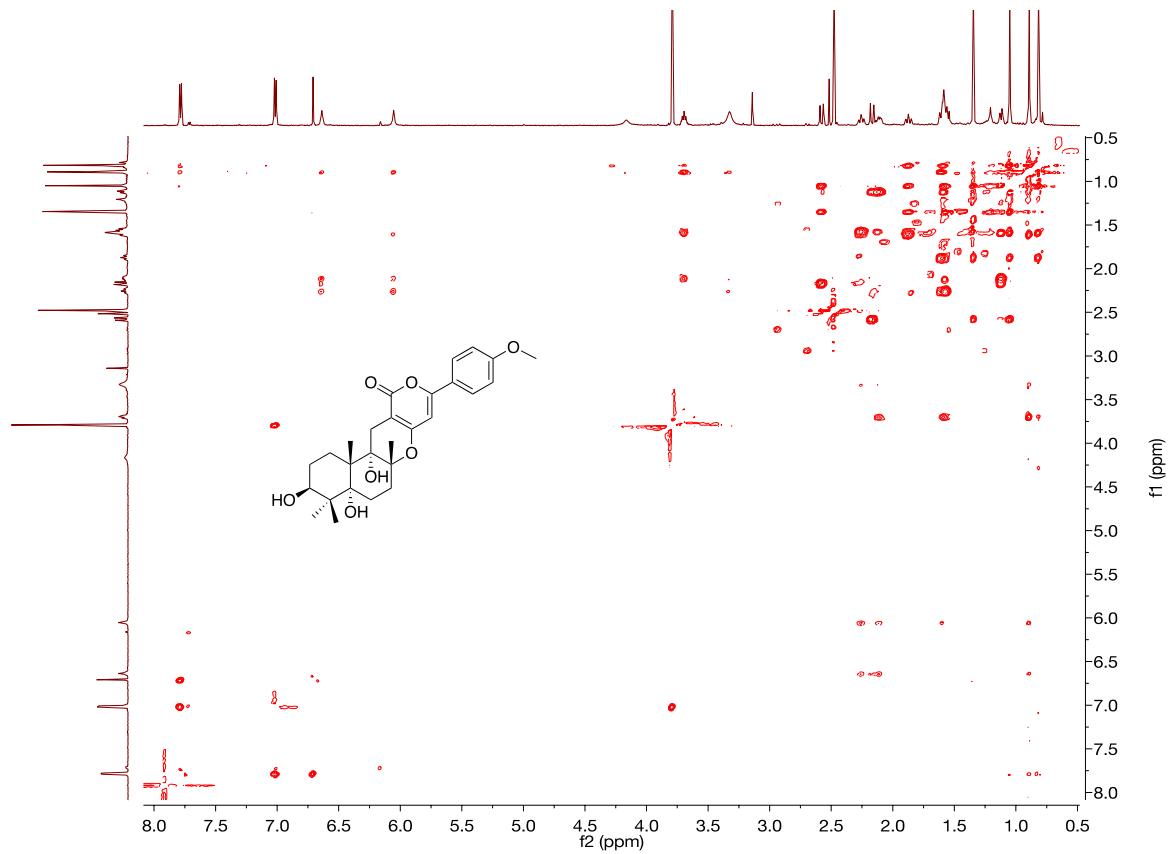


Figure S18. ROESY spectrum of compound **5** in $\text{DMSO}-d_6$ (500 MHz).

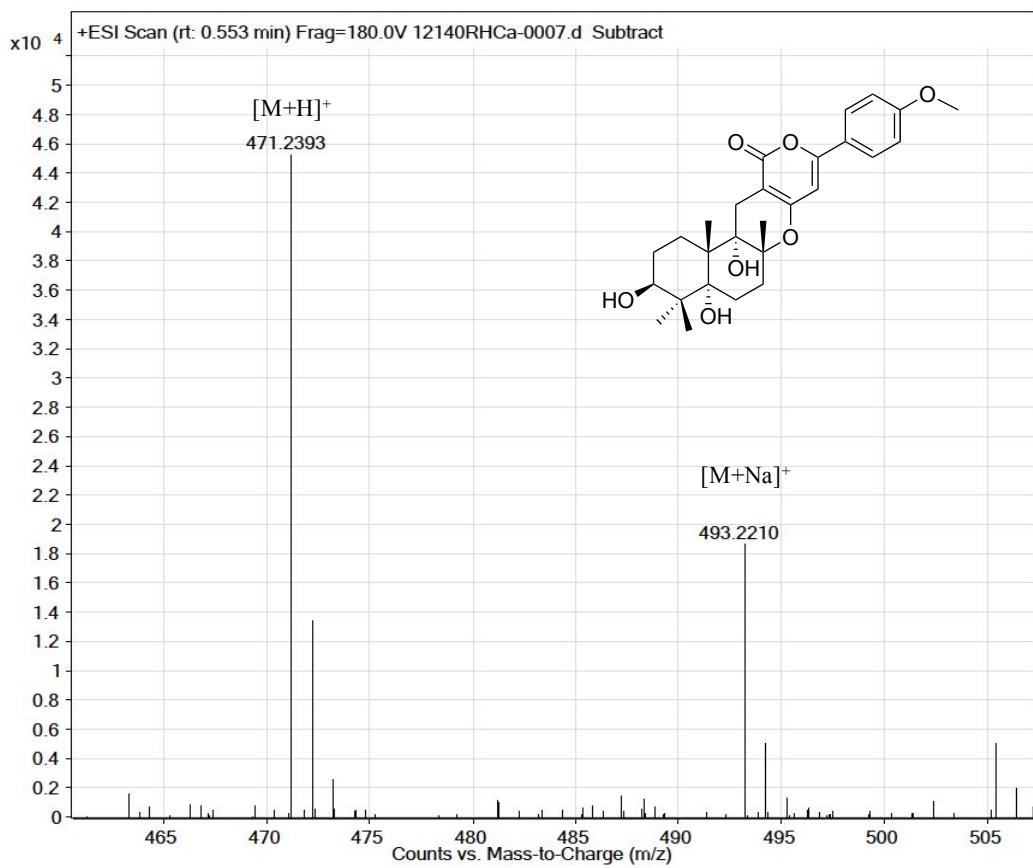


Figure S19. HRESIMS data for compound 5.

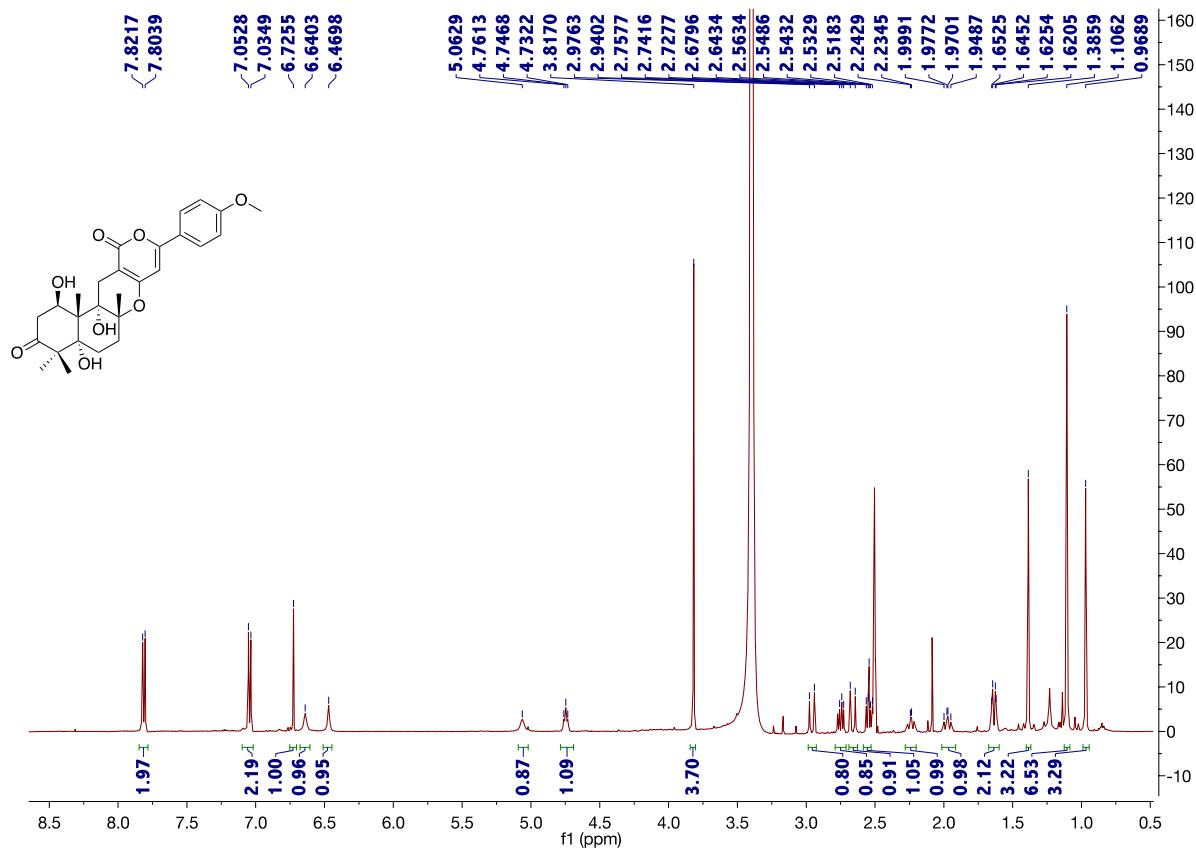


Figure S20. ^1H -NMR spectrum of compound 8 in $\text{DMSO}-d_6$ (500 MHz).

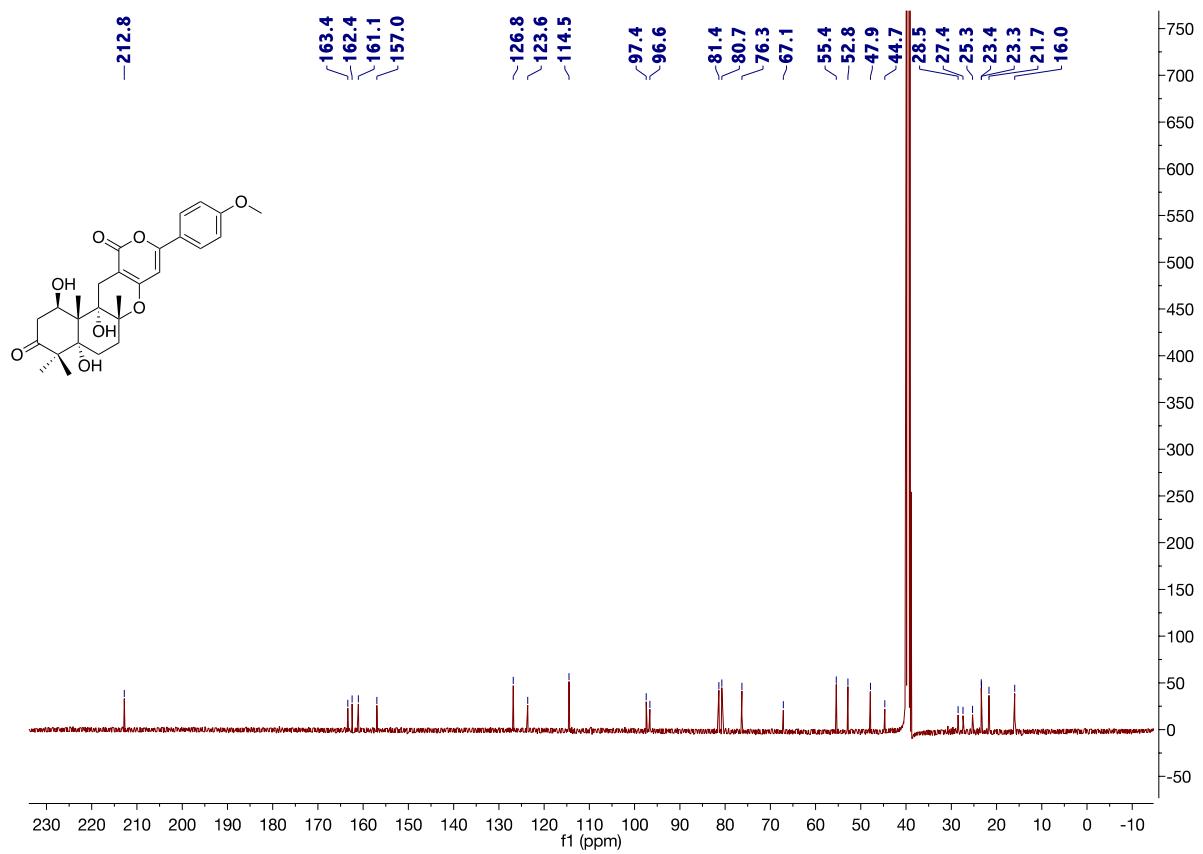


Figure S21. ^{13}C -NMR spectrum of compound 8 in $\text{DMSO}-d_6$ (100 MHz).

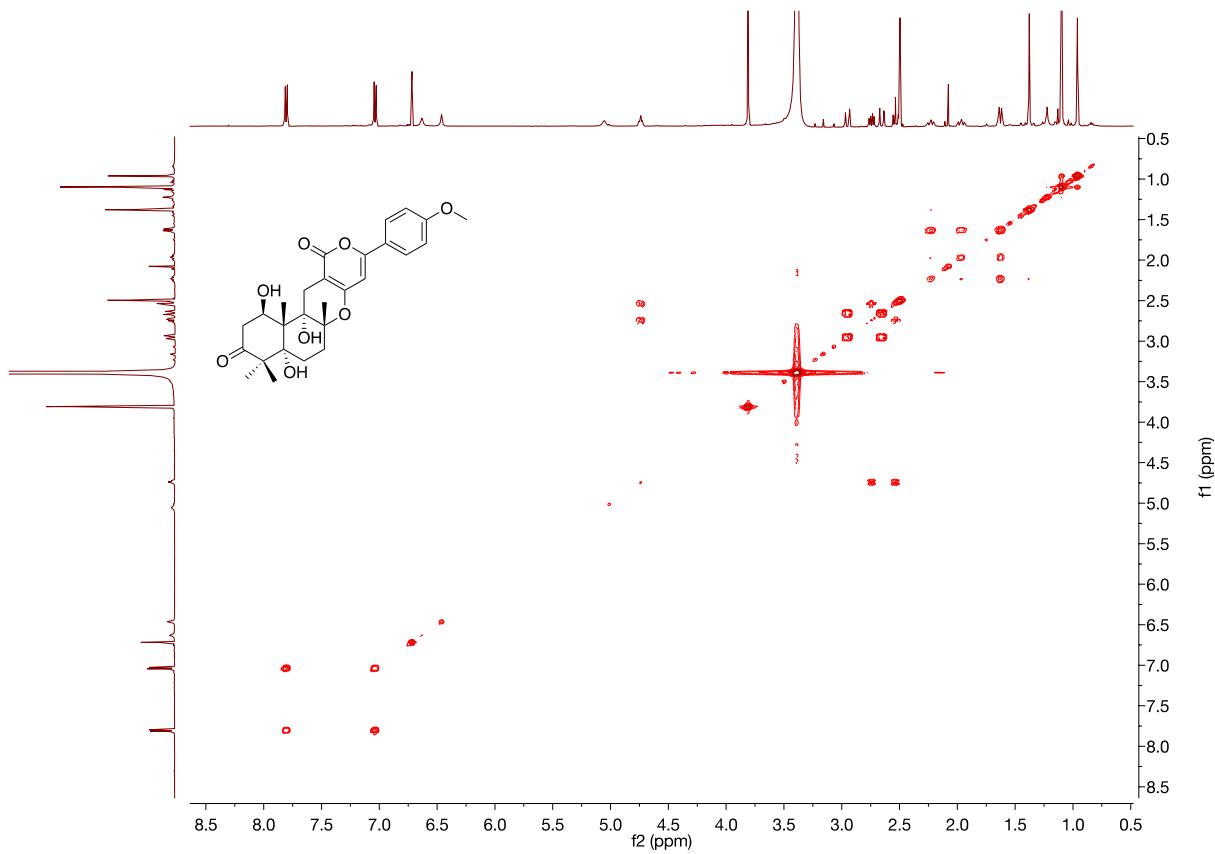


Figure S22. ^1H - ^1H spectrum of compound **8** in $\text{DMSO}-d_6$ (500 MHz).

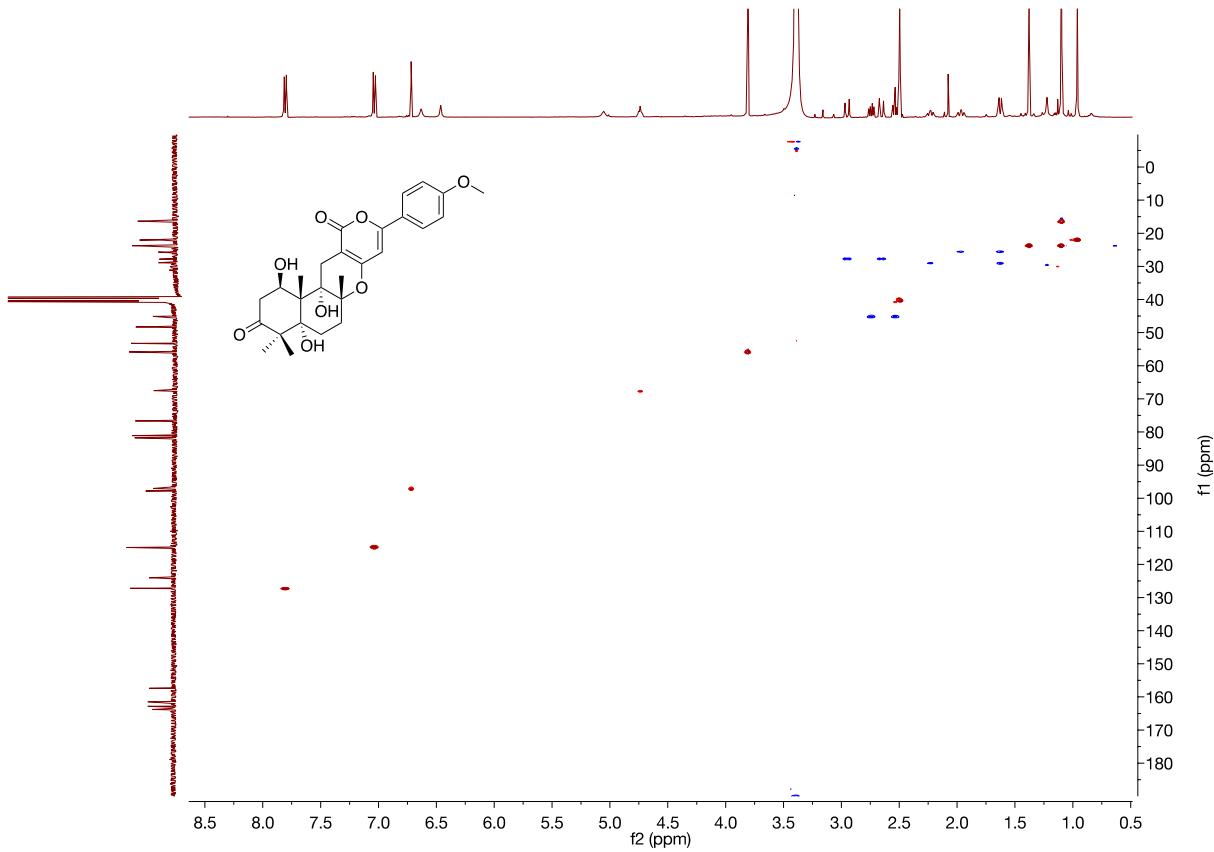


Figure S23. HSQC spectrum of compound **8** in $\text{DMSO}-d_6$ (500 MHz).

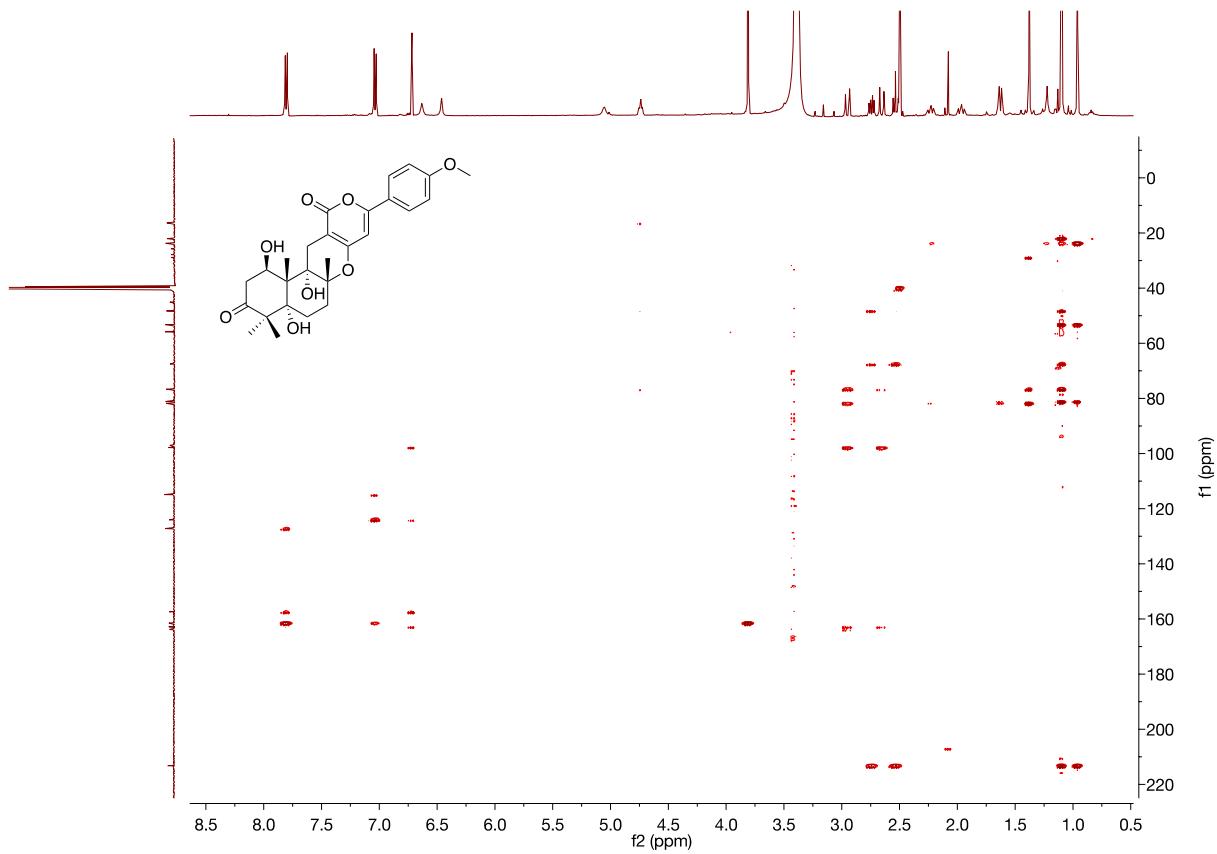


Figure S24. HMBC spectrum of compound **8** in $\text{DMSO}-d_6$ (500 MHz).

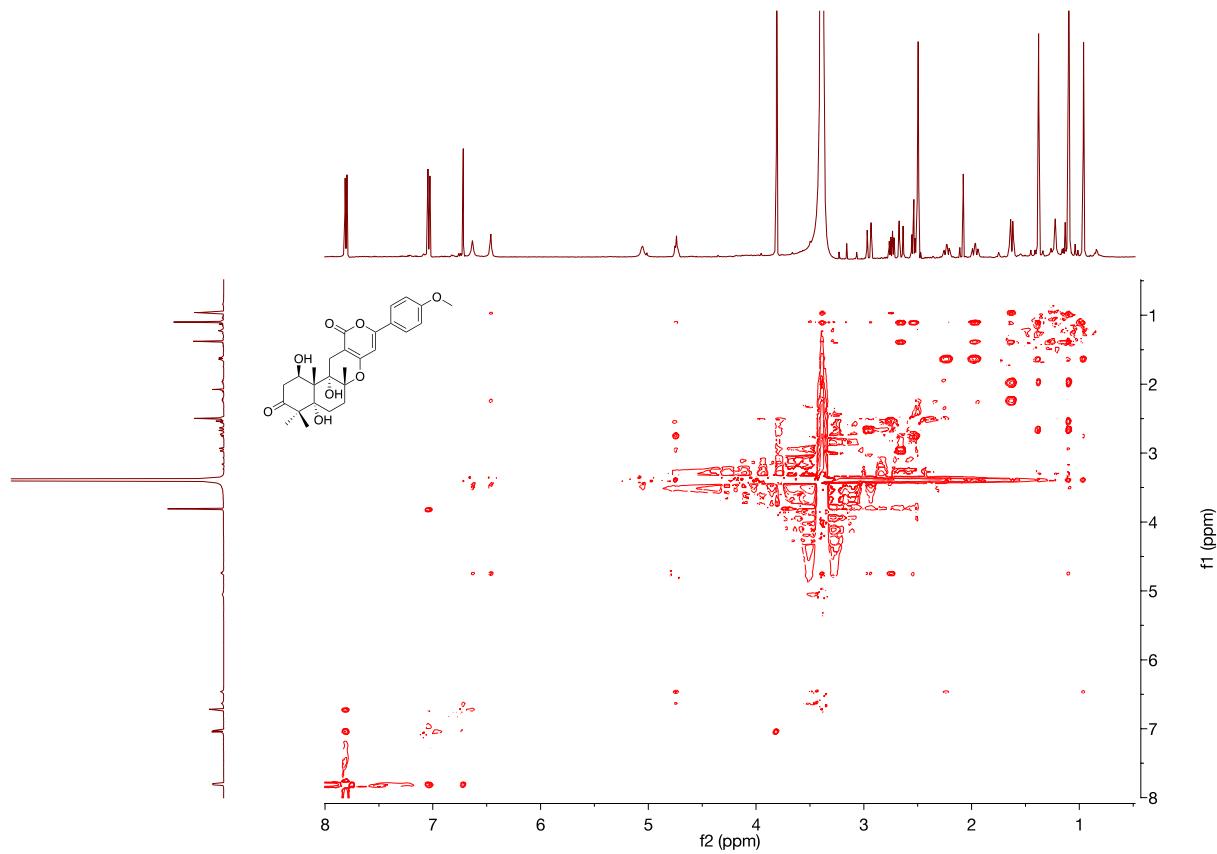


Figure S25. ROESY spectrum of compound **8** in $\text{DMSO}-d_6$ (500 MHz).

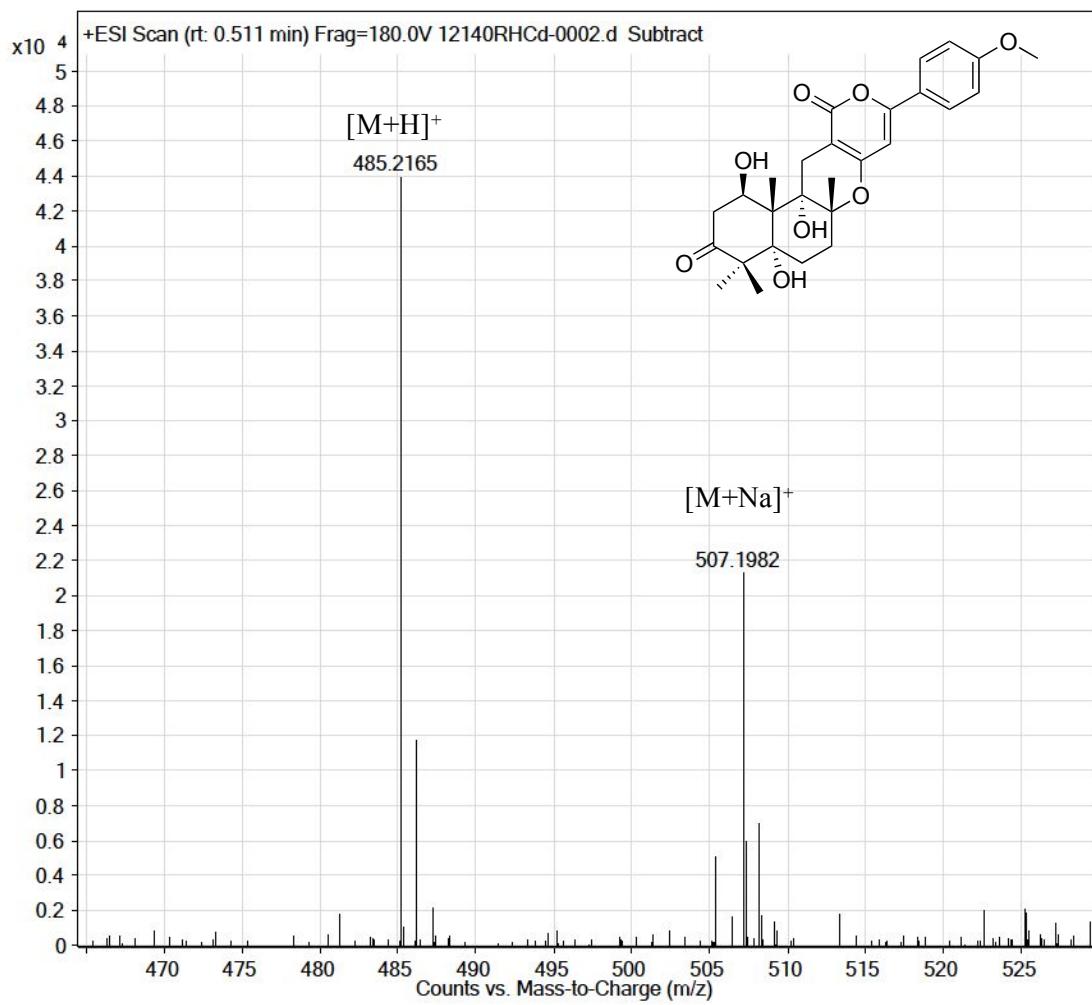


Figure S26. HRESIMS data for compound 8.

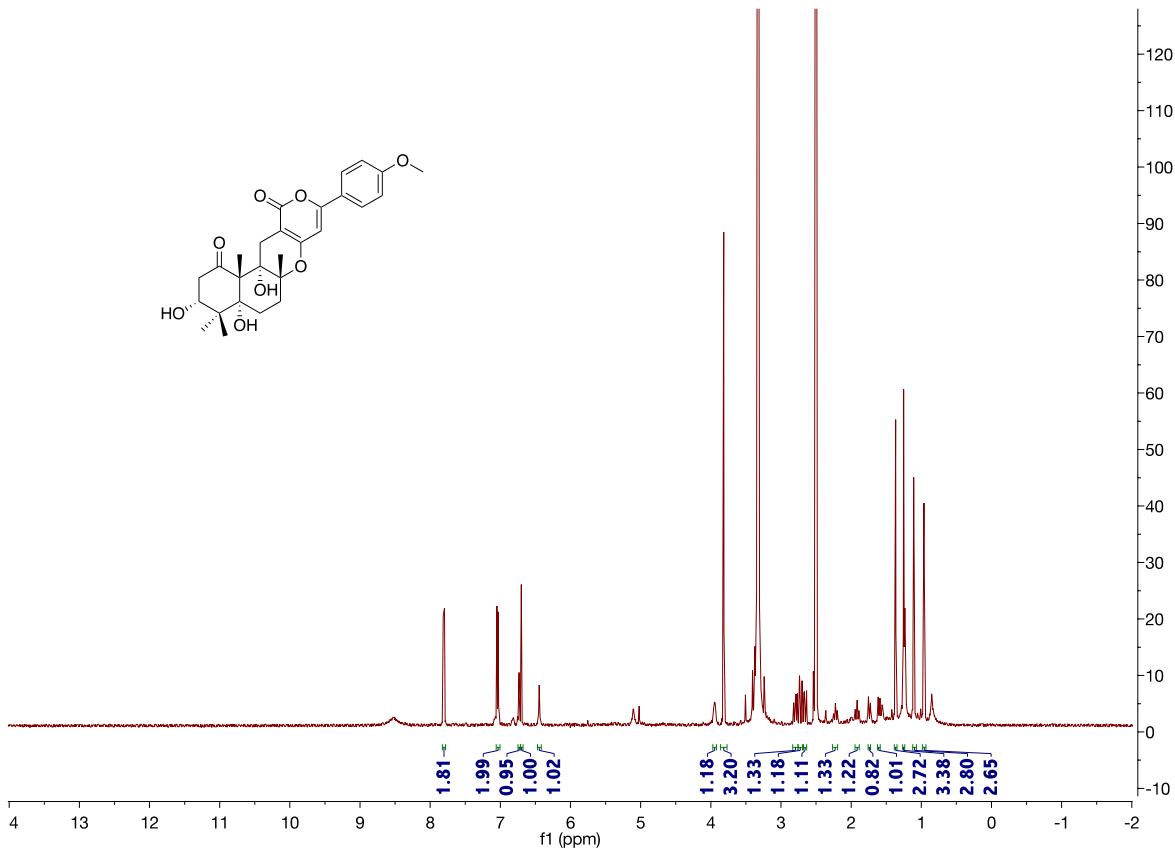


Figure S27. ¹H-NMR spectrum of compound **12** in DMSO-*d*₆ (500 MHz).

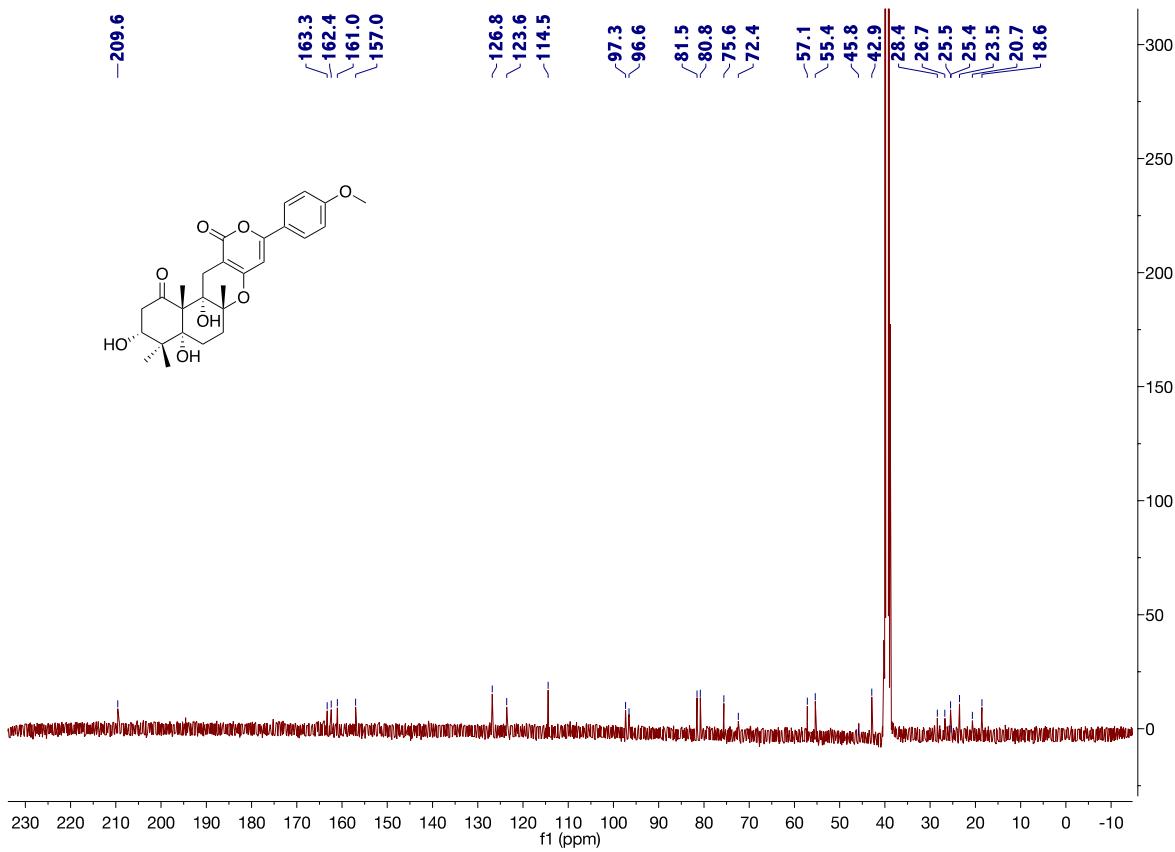


Figure S28. ^{13}C -NMR spectrum of compound **12** in $\text{DMSO}-d_6$ (100 MHz).

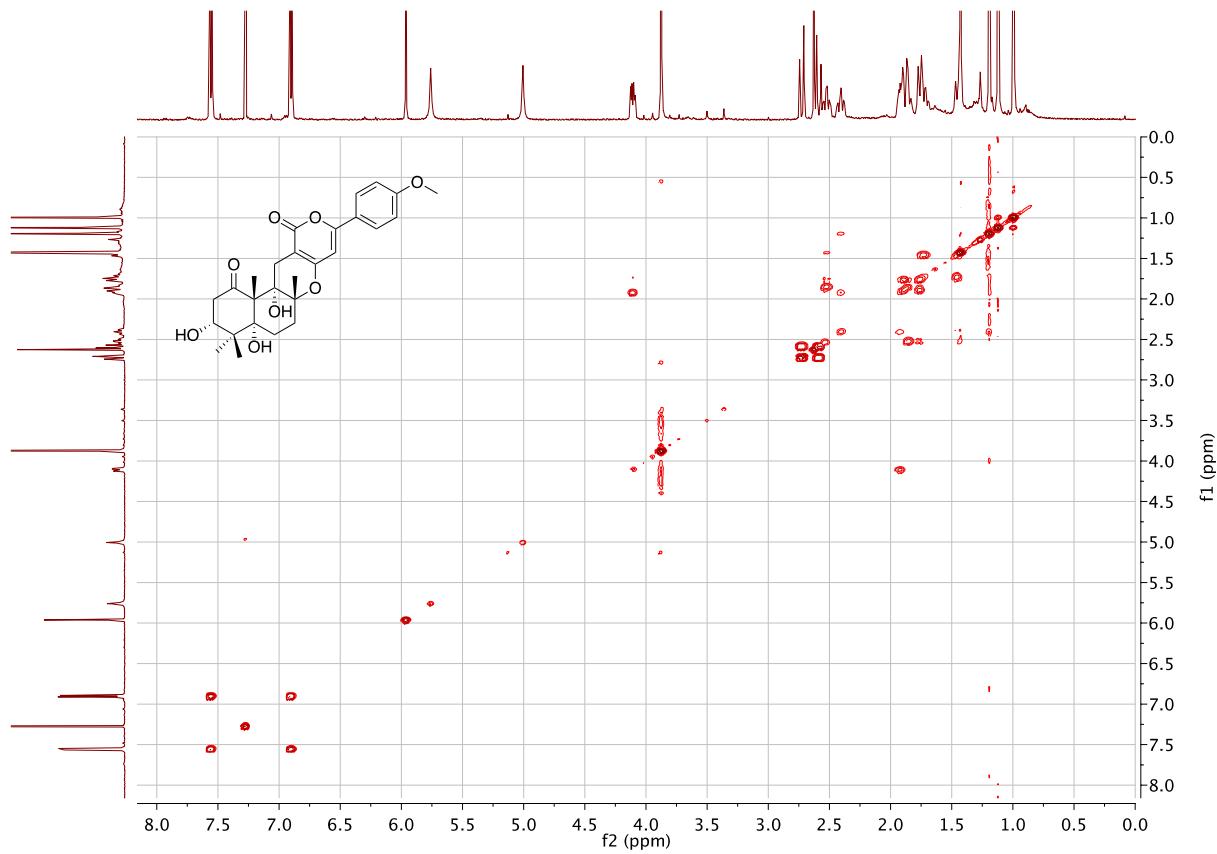


Figure S29. ^1H - ^1H spectrum of compound **12** in $\text{DMSO}-d_6$ (500 MHz).

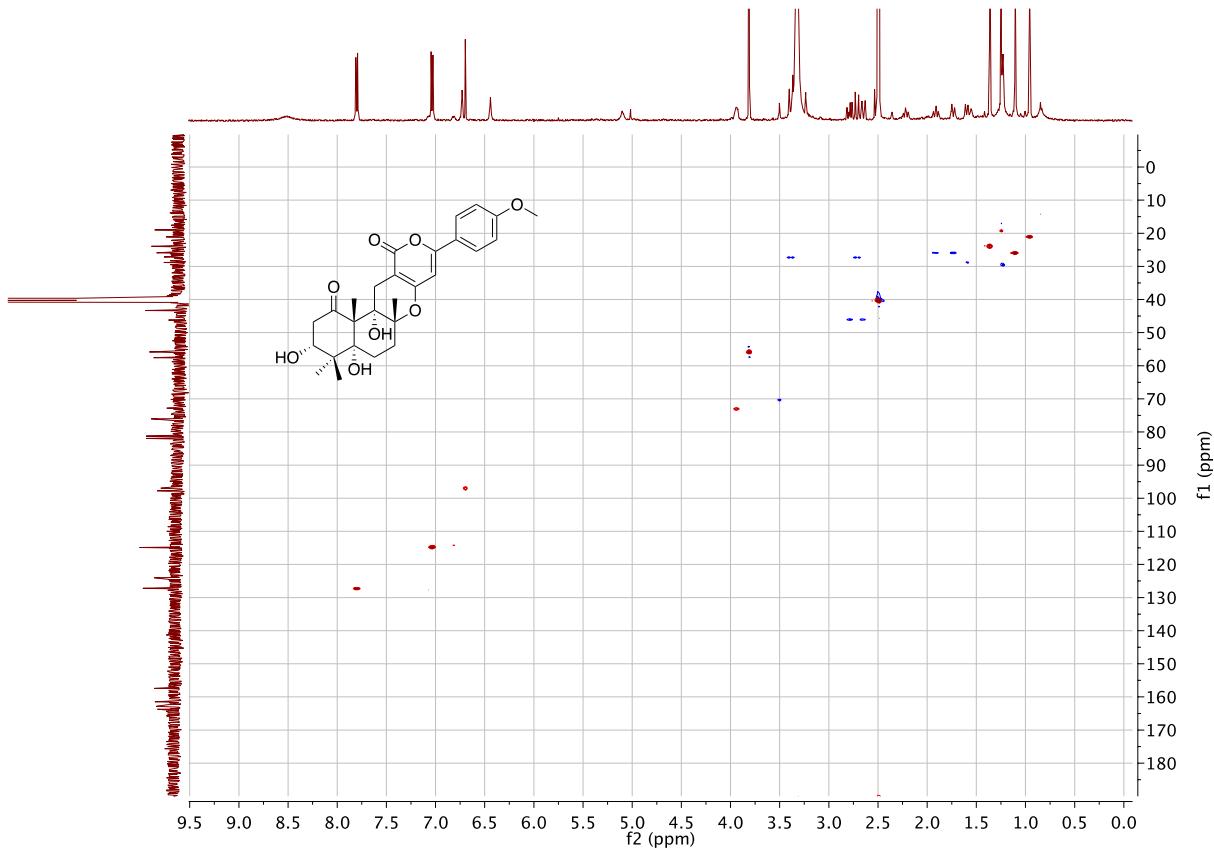


Figure S30. HSQC spectrum of compound **12** in DMSO-*d*₆ (500 MHz).

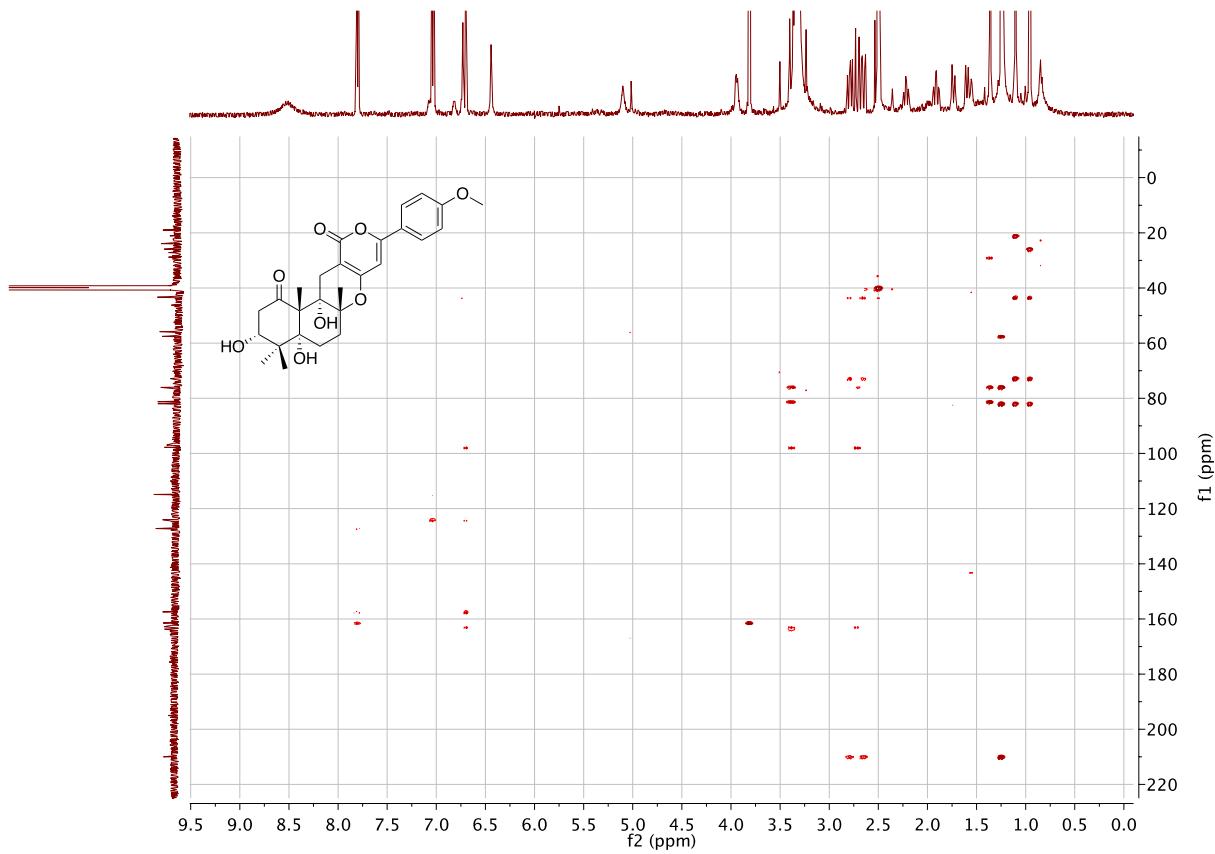


Figure S31. HMBC spectrum of compound **12** in $\text{DMSO}-d_6$ (500 MHz).

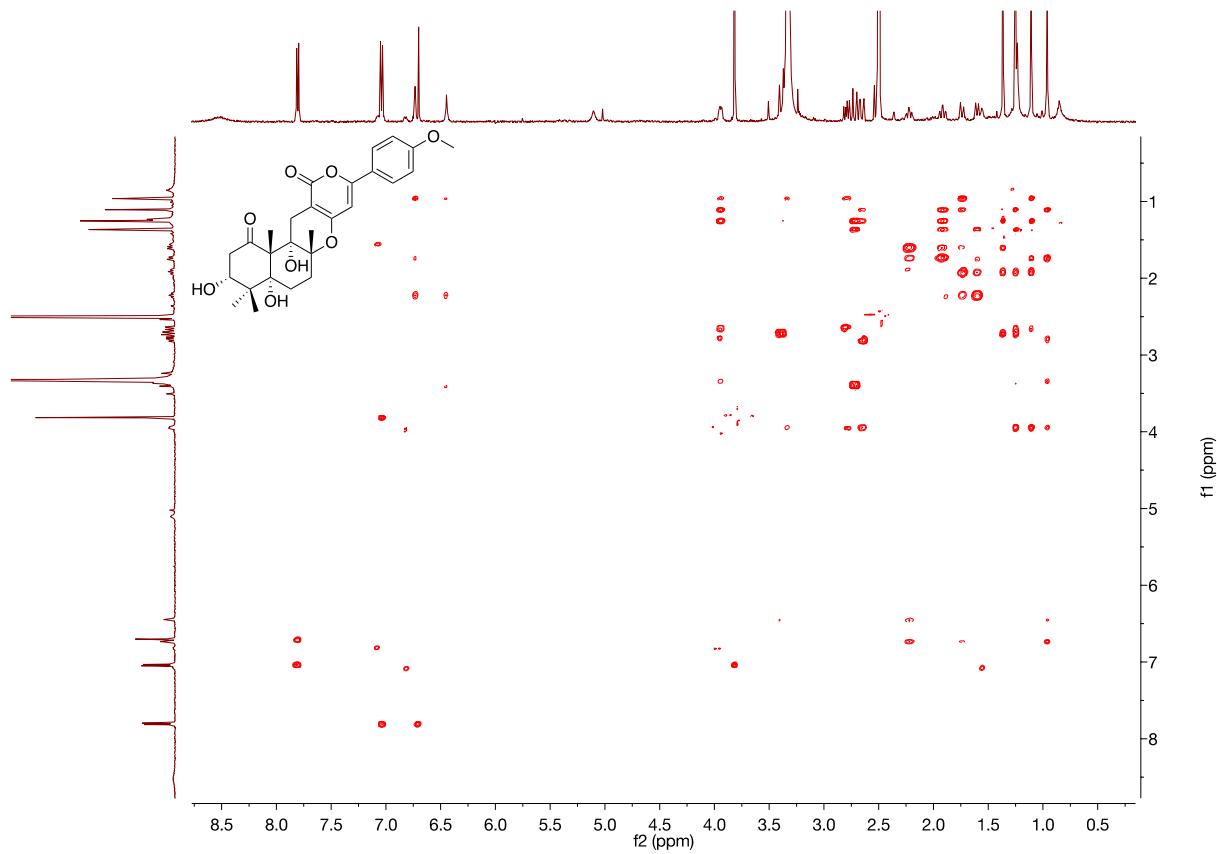


Figure S32. ROESY spectrum of compound **12** in DMSO-*d*₆ (500 MHz).

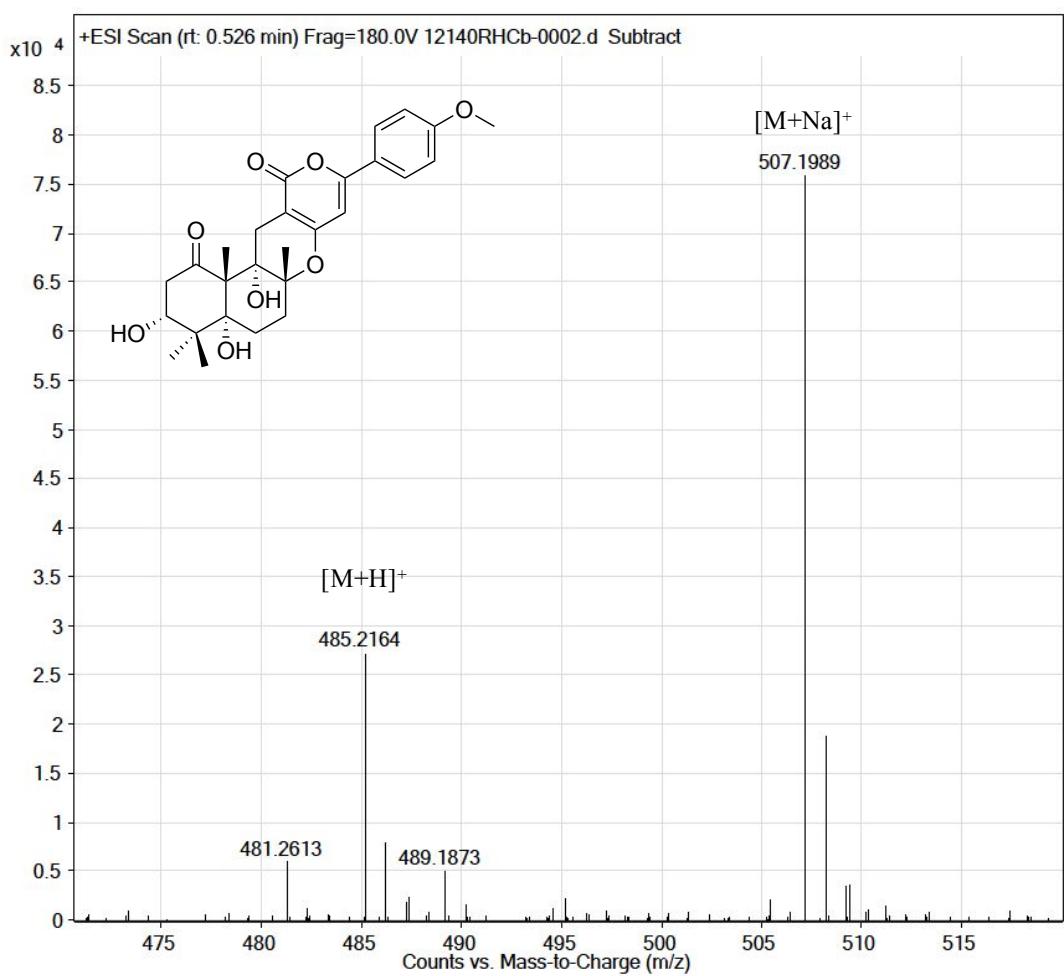


Figure S33. HRESIMS data for compound **12**.

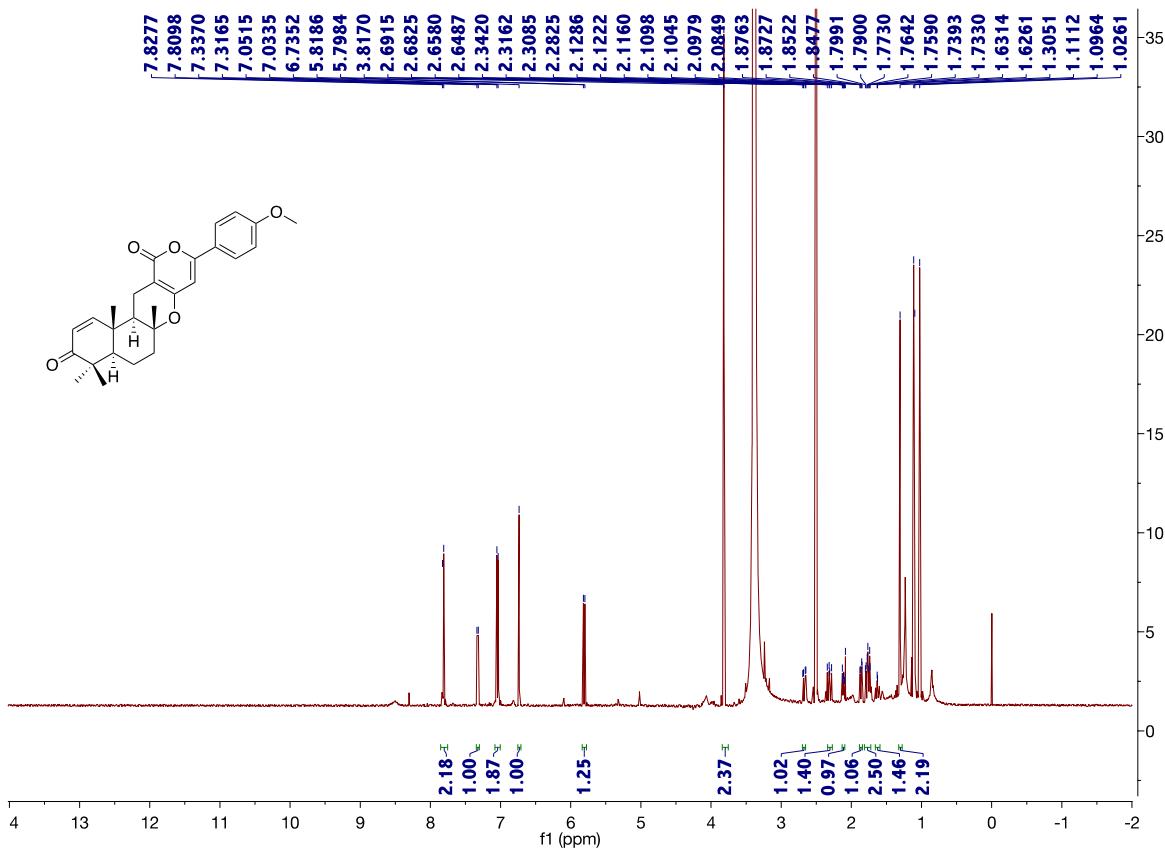


Figure S34. ^1H -NMR spectrum of compound **13** in $\text{DMSO}-d_6$ (500 MHz).

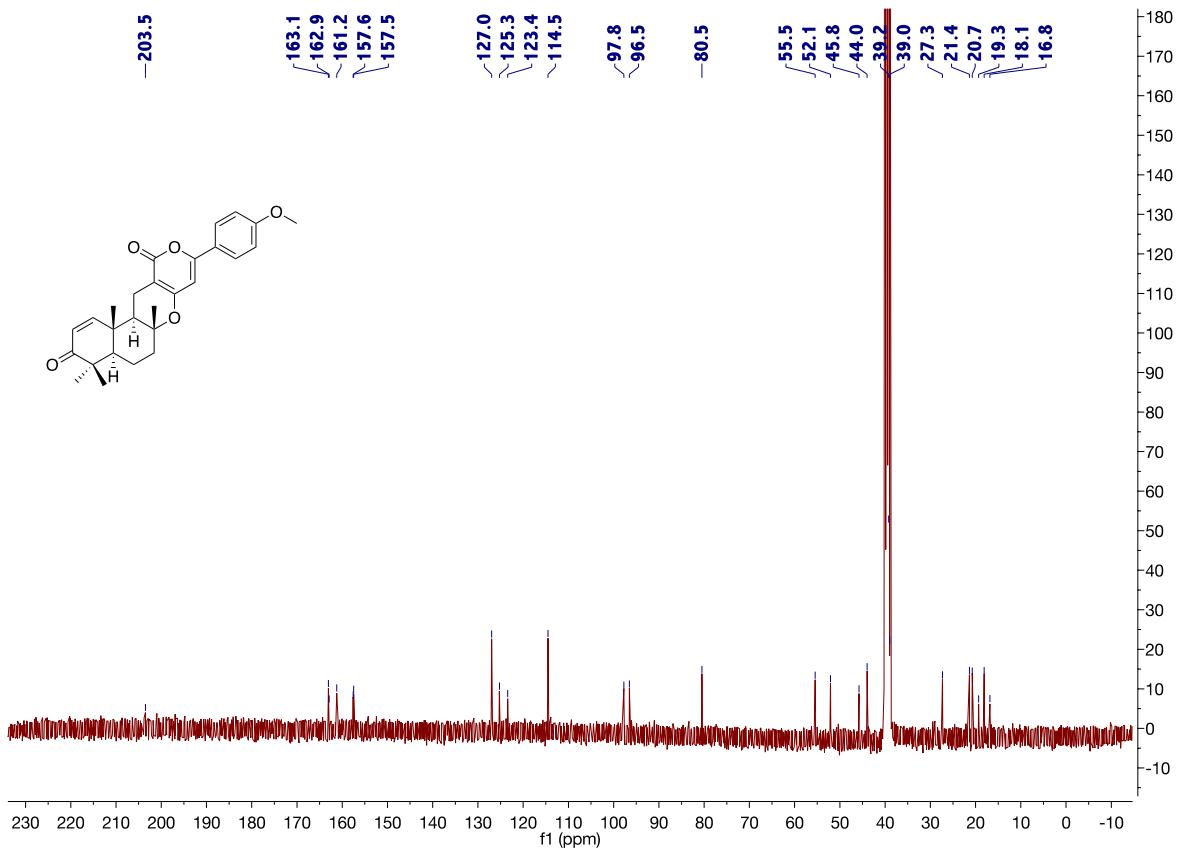


Figure S35. ^{13}C -NMR spectrum of compound 13 in $\text{DMSO}-d_6$ (100 MHz).

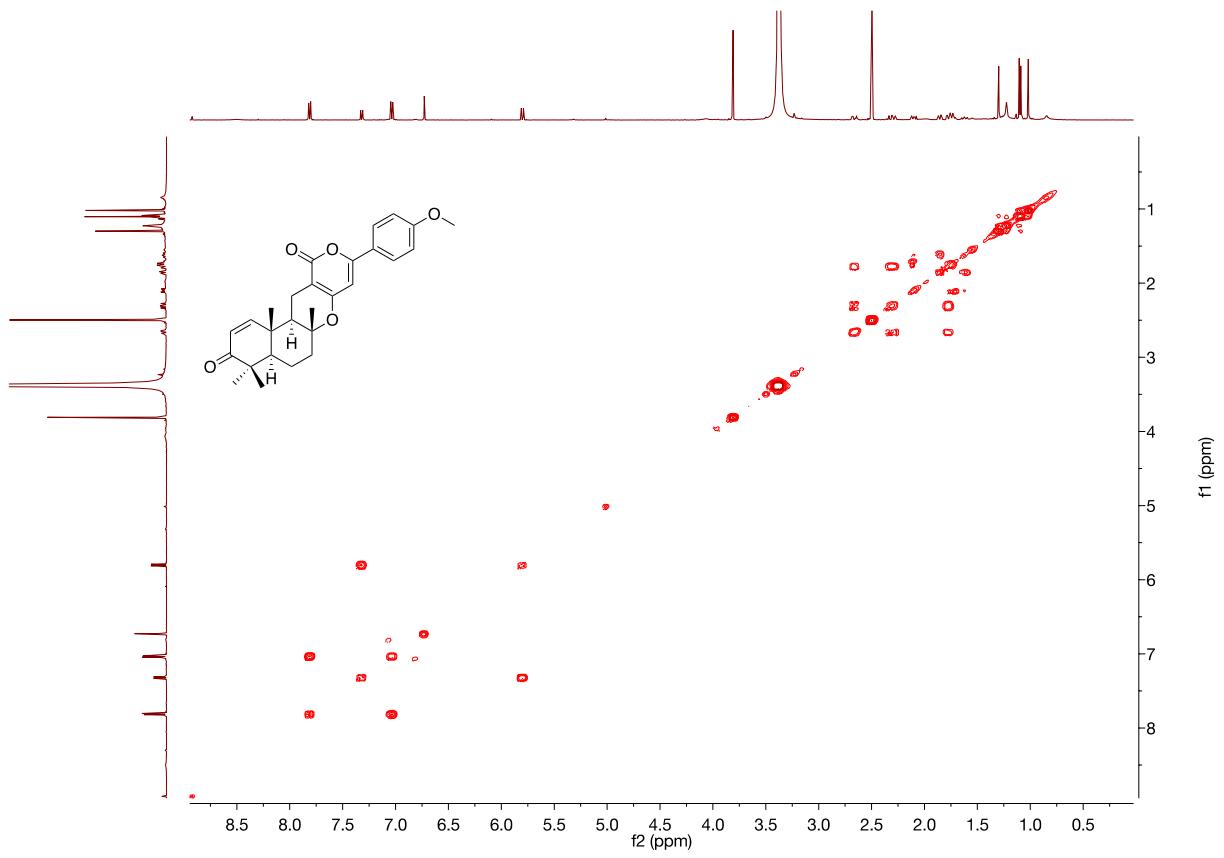


Figure S36. ^1H - ^1H spectrum of compound **13** in $\text{DMSO}-d_6$ (500 MHz).

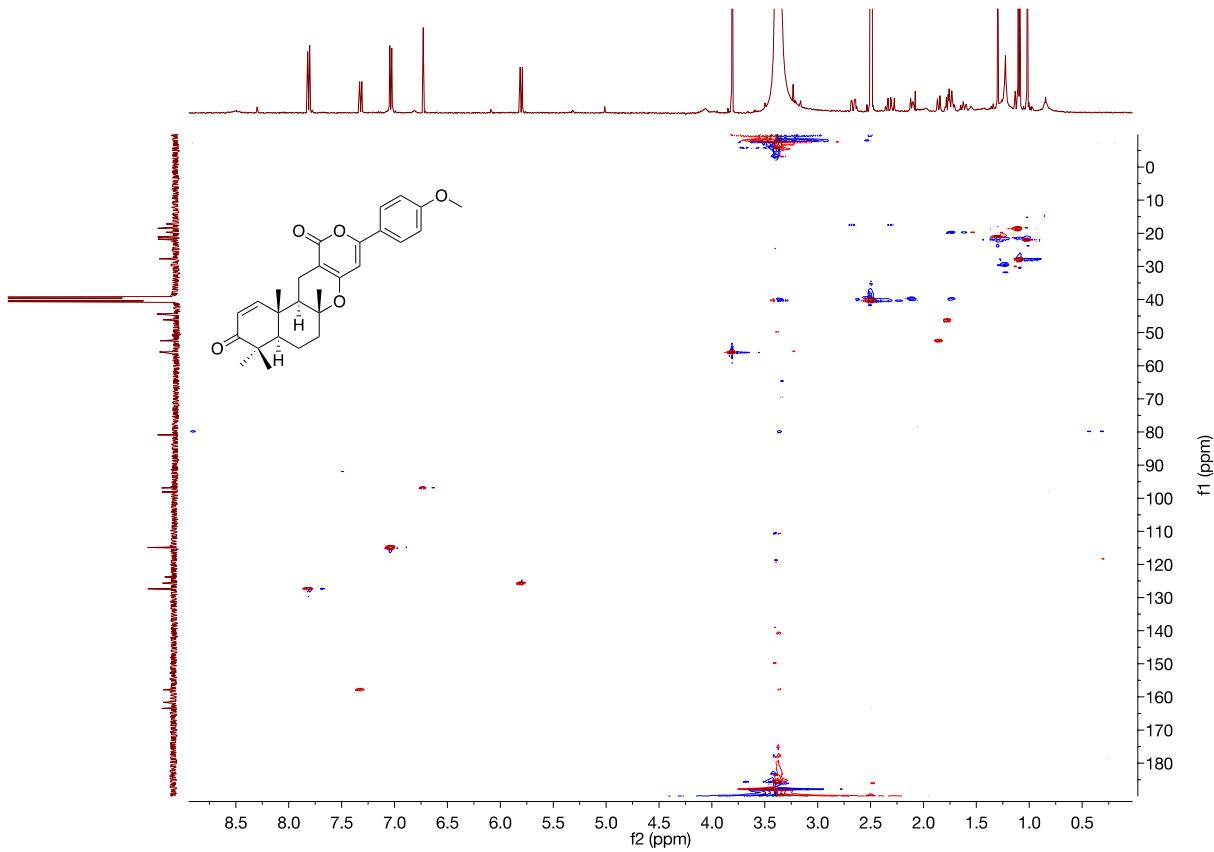


Figure S37. HSQC spectrum of compound **13** in $\text{DMSO}-d_6$ (500 MHz).

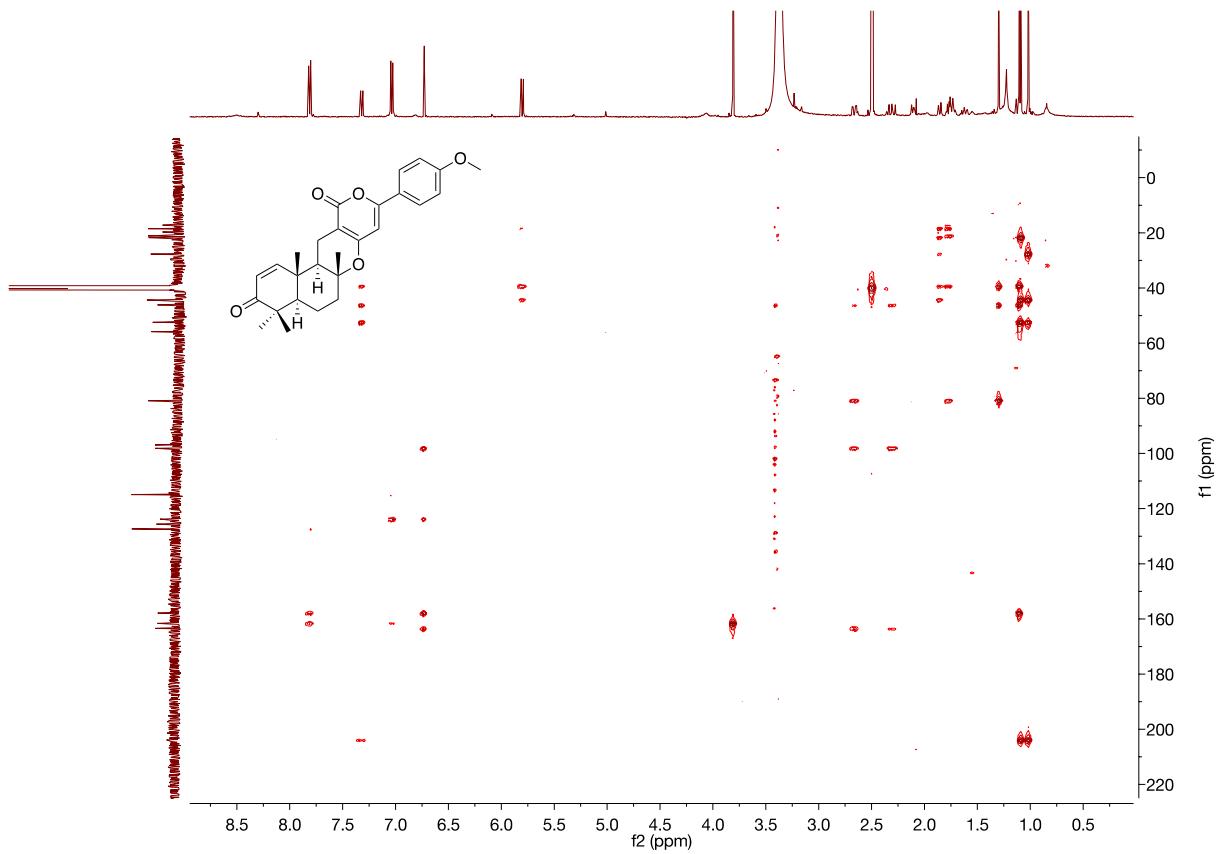


Figure S38. HMBC spectrum of compound **13** in $\text{DMSO}-d_6$ (500 MHz).

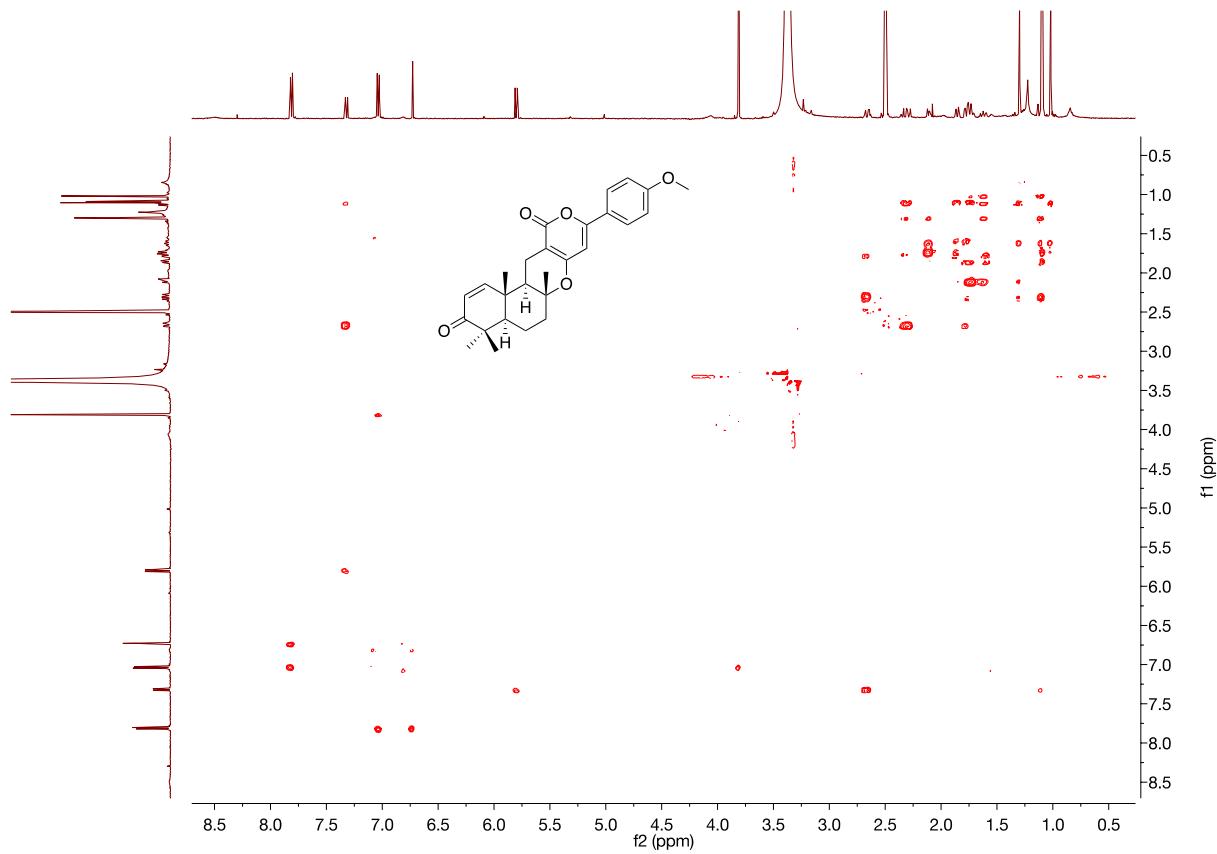


Figure S39. ROESY spectrum of compound **13** in $\text{DMSO}-d_6$ (500 MHz).

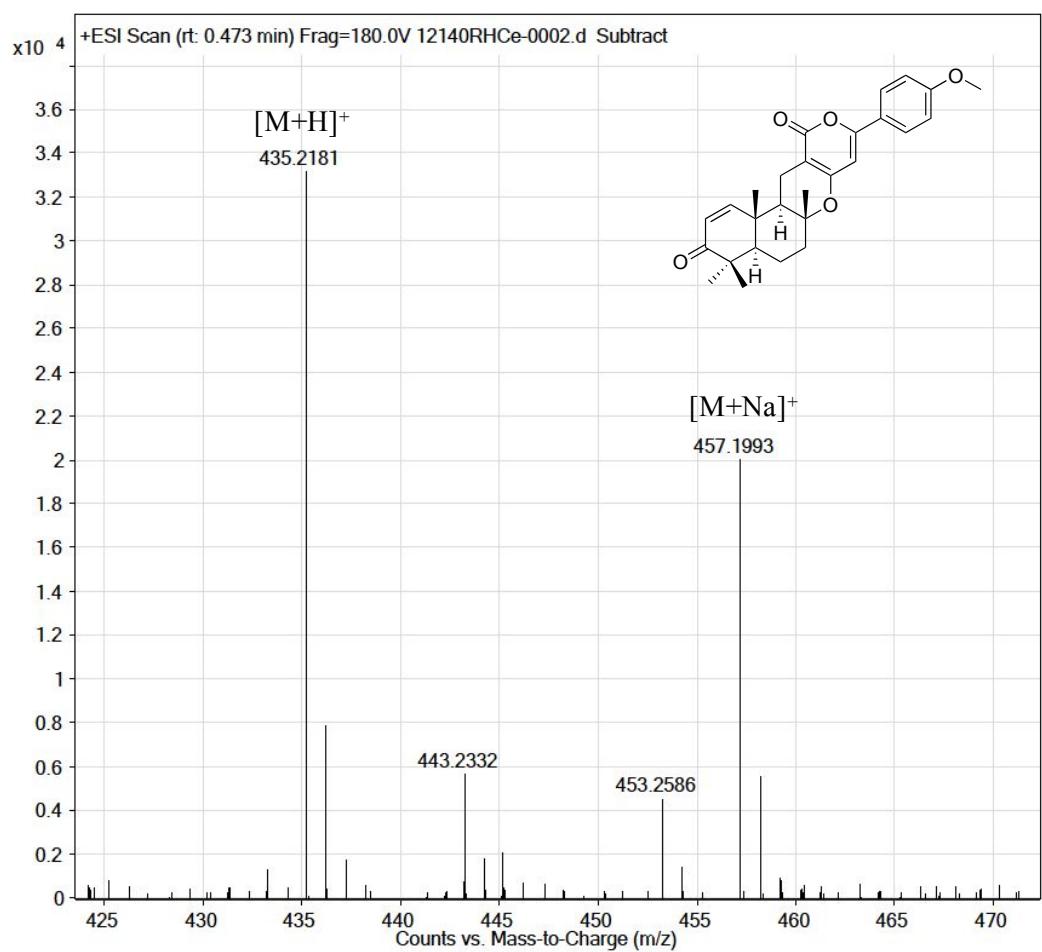


Figure S40. HRESIMS data for compound **13**.

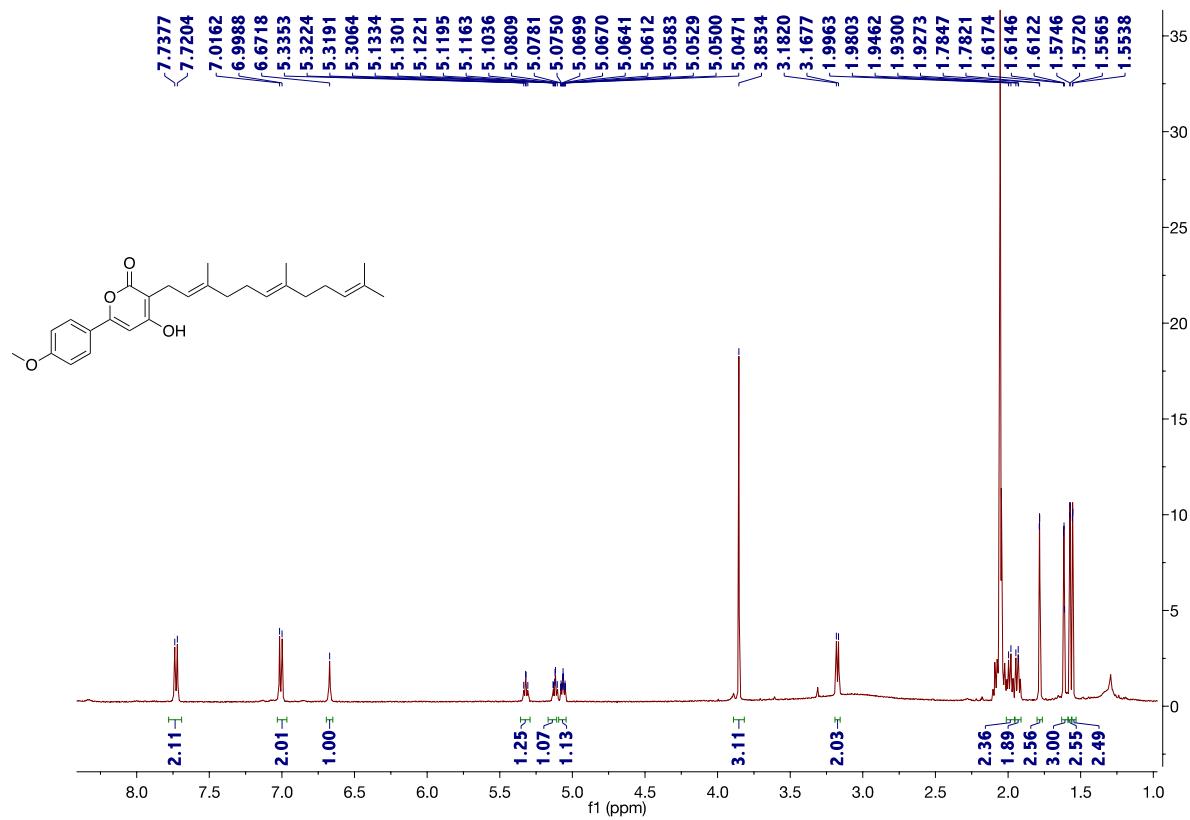


Figure S41. ^1H -NMR spectrum of compound **14** in acetone- d_6 (500 MHz).

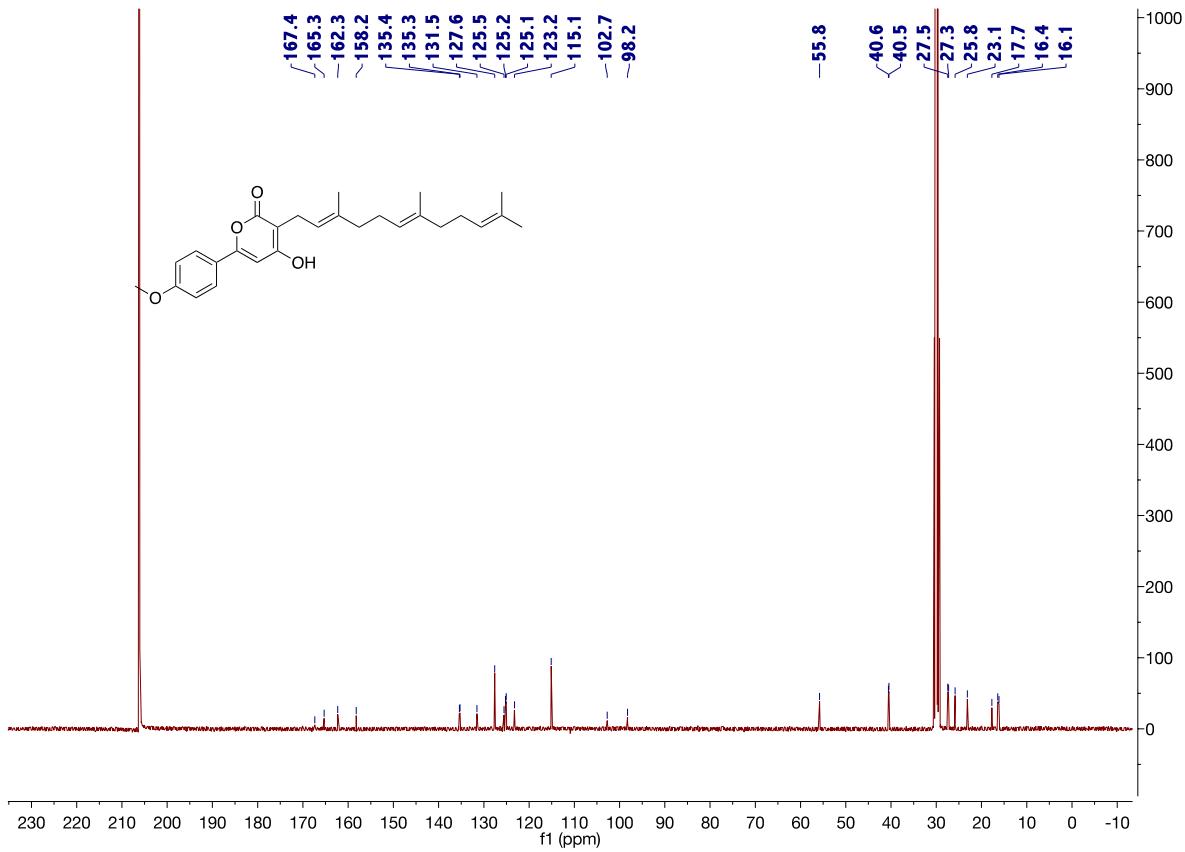


Figure S42. ^{13}C -NMR spectrum of compound 14 in acetone- d_6 (100 MHz).

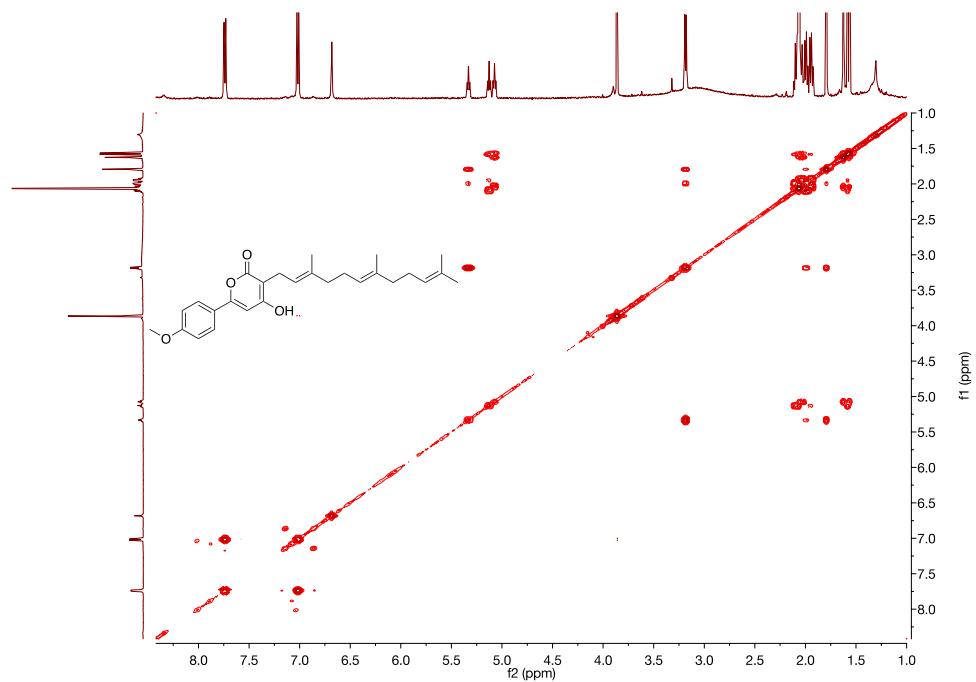


Figure S43. ^1H - ^1H spectrum of compound **14** in acetone- d_6 (500 MHz).

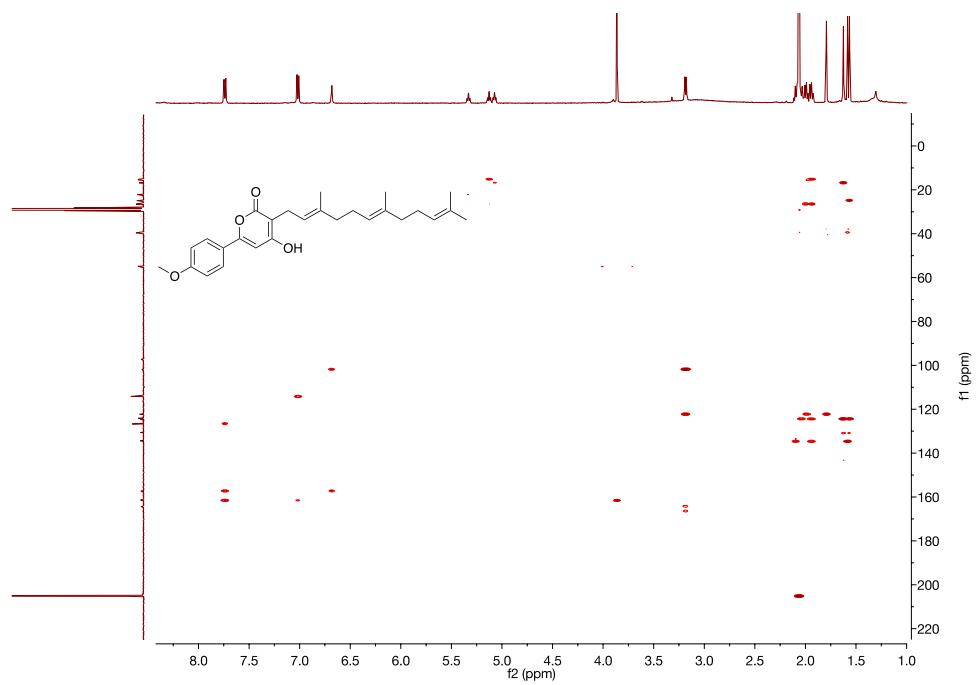


Figure S44. HSQC spectrum of compound **14** in acetone-*d*₆ (500 MHz).

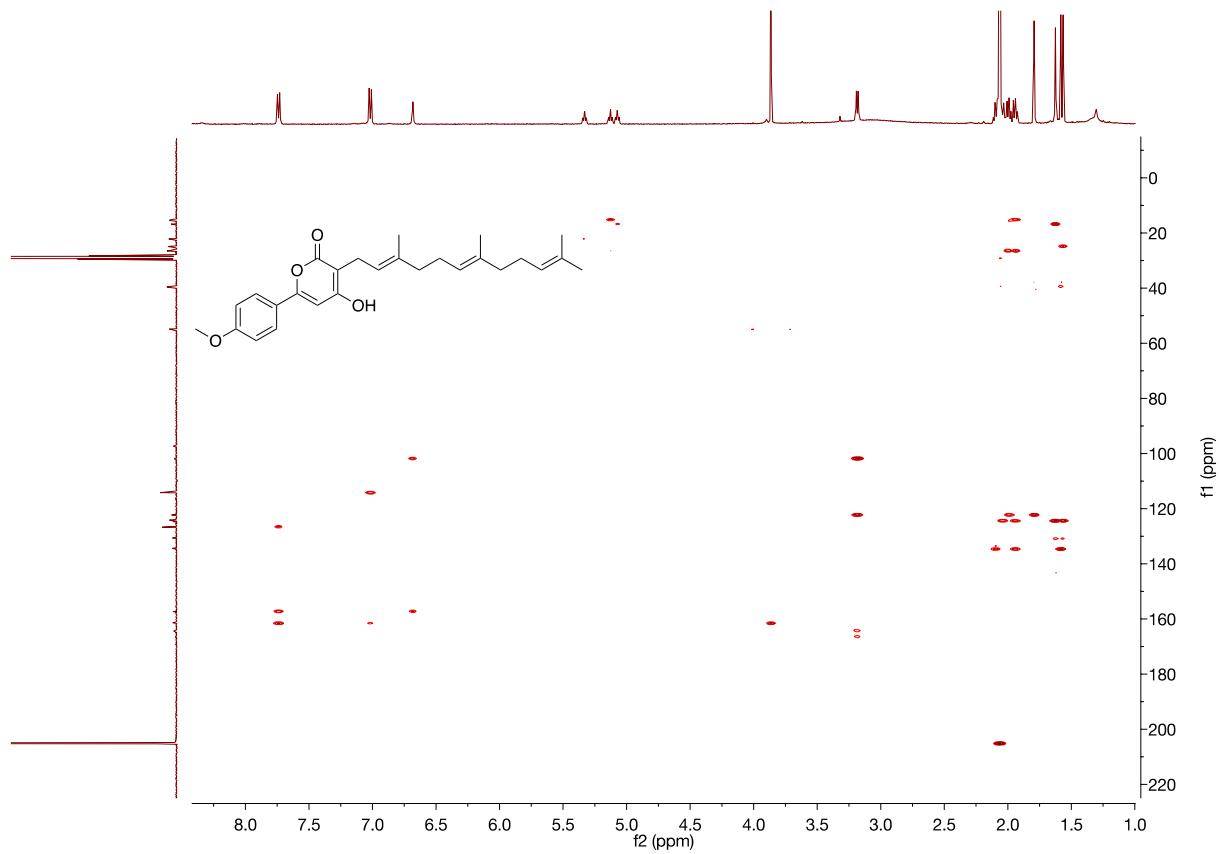


Figure S45. HMBC spectrum of compound **14** in acetone-*d*₆ (500 MHz).

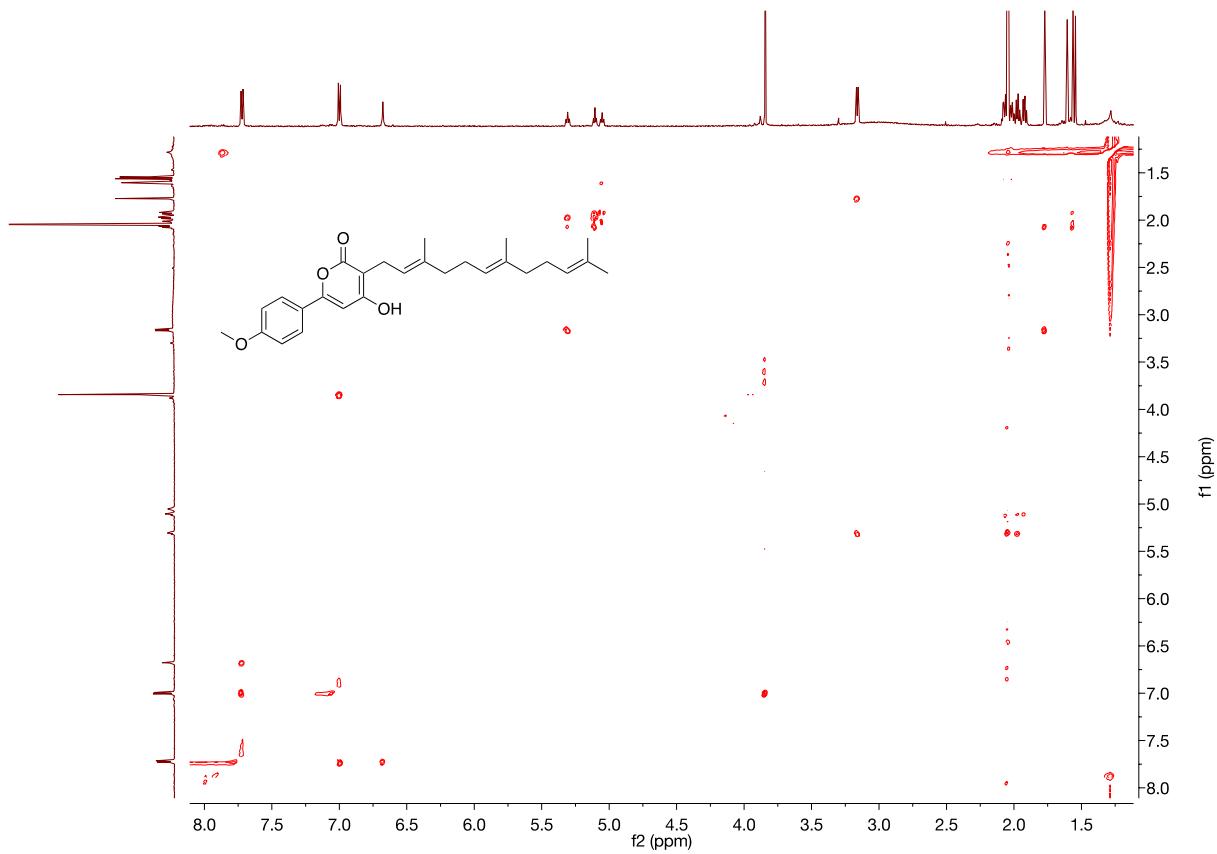


Figure S46. ROESY spectrum of compound **14** in acetone-*d*₆ (500 MHz).

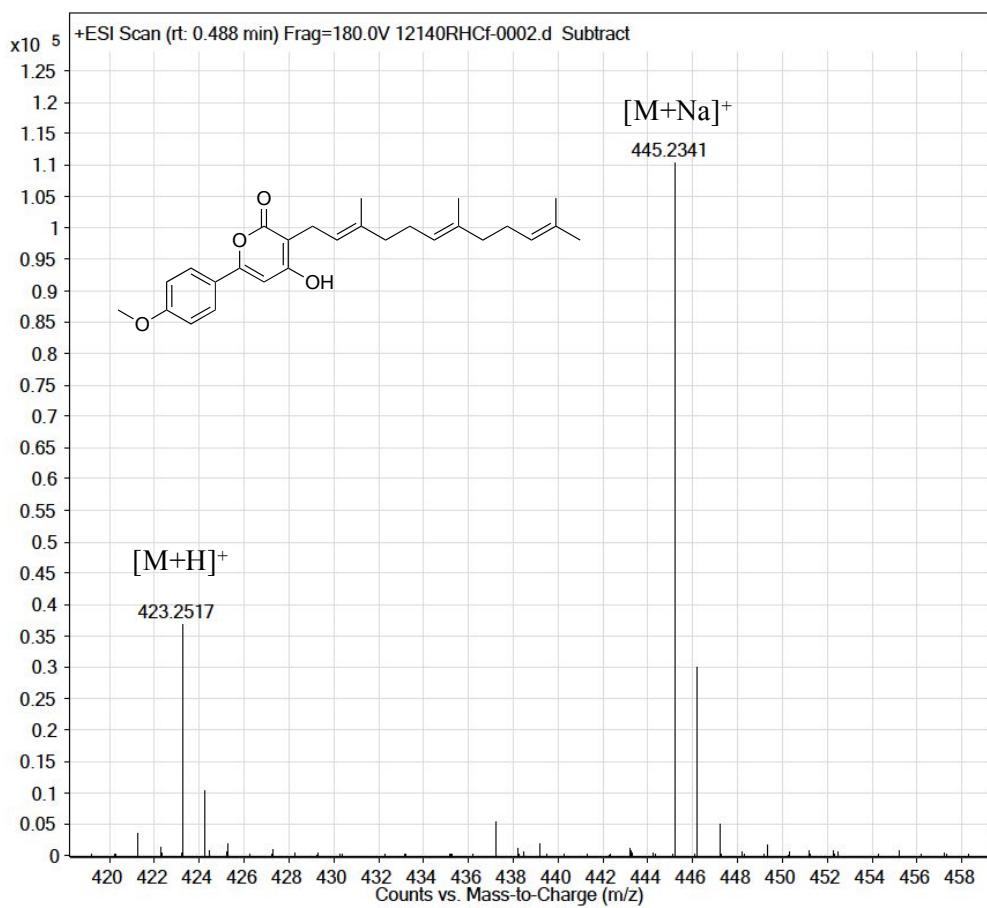


Figure S47. HRESIMS data for compound 14.