Supporting Information

Addressing Structural Flexibility at the A-Ring on Salvinorin A: Discovery of a Potent Kappa Opioid Agonist with Enhanced Metabolic Stability

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[1] Department of Medicinal Chemistry, School of Pharmacy, The University of Kansas, Lawrence, KS, USA. [2] School of Biological Sciences, Centre for Biodiscovery, Victoria University of Wellington, New Zealand. [3] Department of Chemistry, The University of Kansas, Lawrence, KS, USA. [†]These authors contributed equally to this work.

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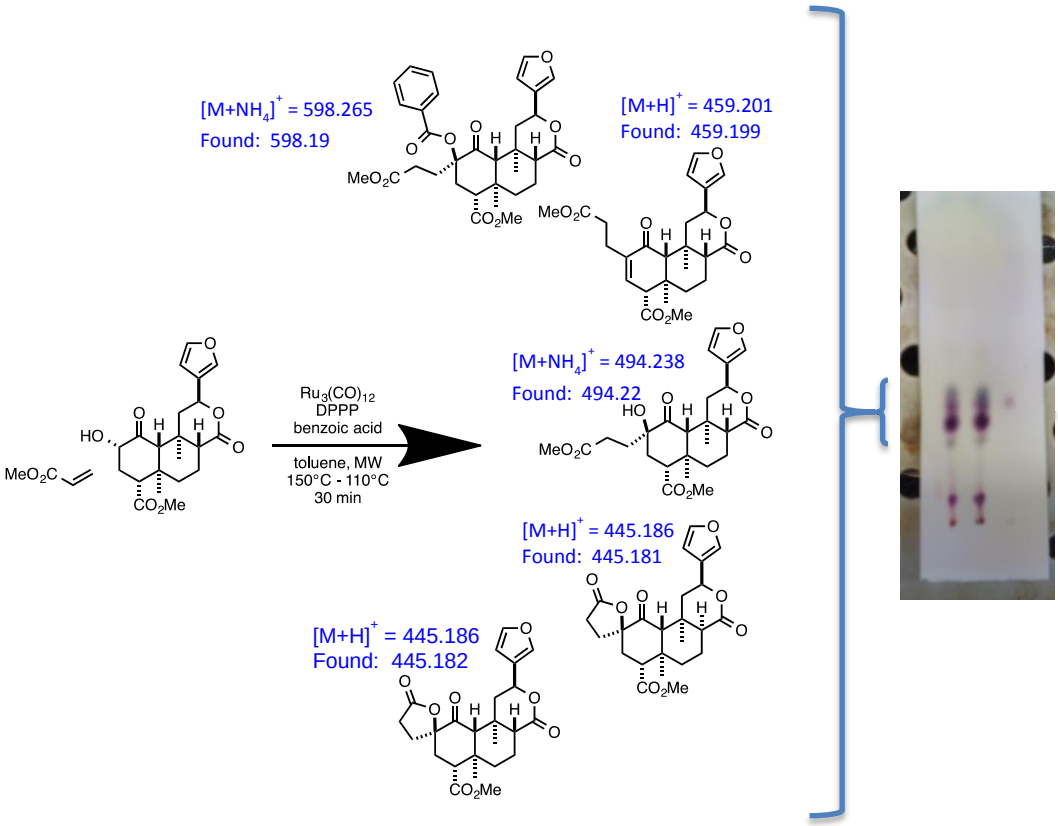


Figure S1. Thin layer chromatography analysis of crude reaction mixture (30 % EtOAc in hexanes; vanillin stain). Proposed structures identified by mass are shown.

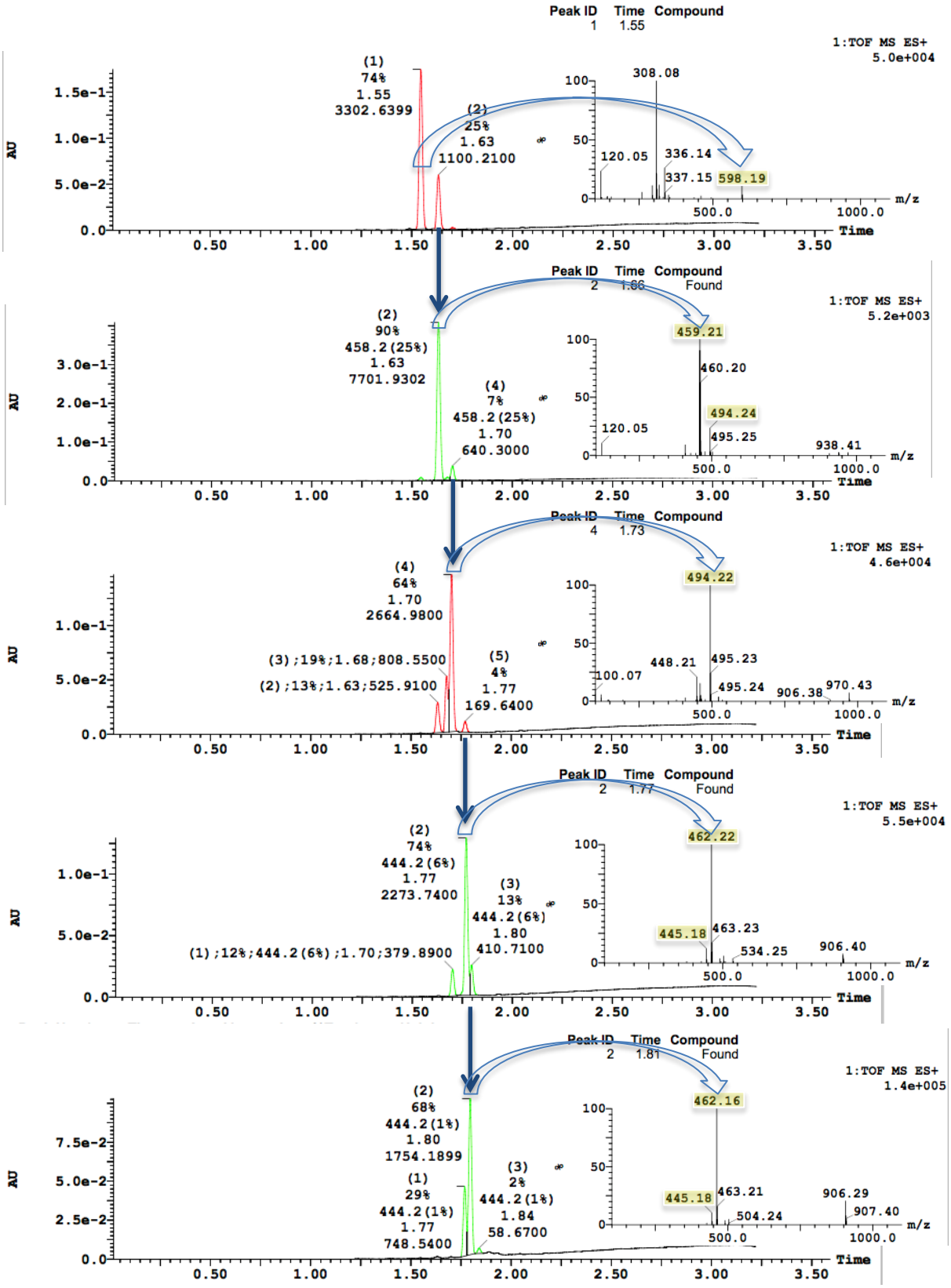
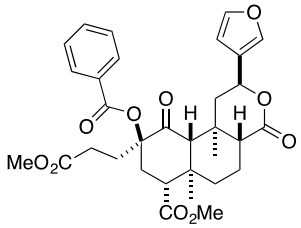
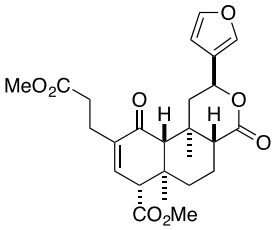
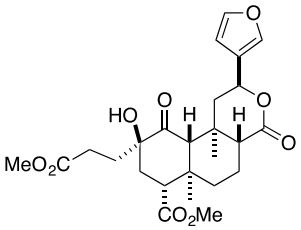
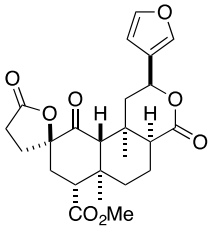
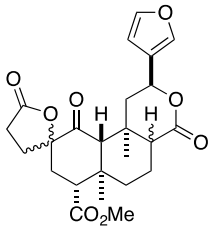


Figure S2. HPLC analysis for fractionated proposed structures (~70 % single compound) aligned by retention time. Mass spectrum for major component shown on right with mass highlighted in yellow.

Proposed structure	RT (min)	% mass recovery	% enriched	Adduct	Calcd.	Found	kOR activity Potency ± SEM[a,b] (nM)
	1.55	2.4	74	$[M+NH_4]^+$	598.2647	598.1988	160 ± 20
	1.63	24.5	90	$[M+H]^+$	459.2013 494.2385	459.1987	>1000 [c]
	1.70	4.5	64	$[M+NH_4]^+$	494.2385	494.2284	100 ± 40
	1.77	11.6	74	$[M+NH_4]^+$ $[M+H]^+$	462.2122 445.1857	462.2286 445.1813	0.6 ± 0.2
	1.80	2.3	68	$[M+NH_4]^+$ $[M+H]^+$	462.2122 445.1857	462.2287 445.1816	22 ± 7

[a]Mean ± standard error of the mean; n ≥ 2 individual experiments run in triplicate. [b]KOR E_{max} = 100%. [c]KOR E_{max} = 0% up to 10uM

Table S1. Proposed structures from Figures S1 and S2 organized by retention time with mass characterization shown and corresponding activity at KOR.

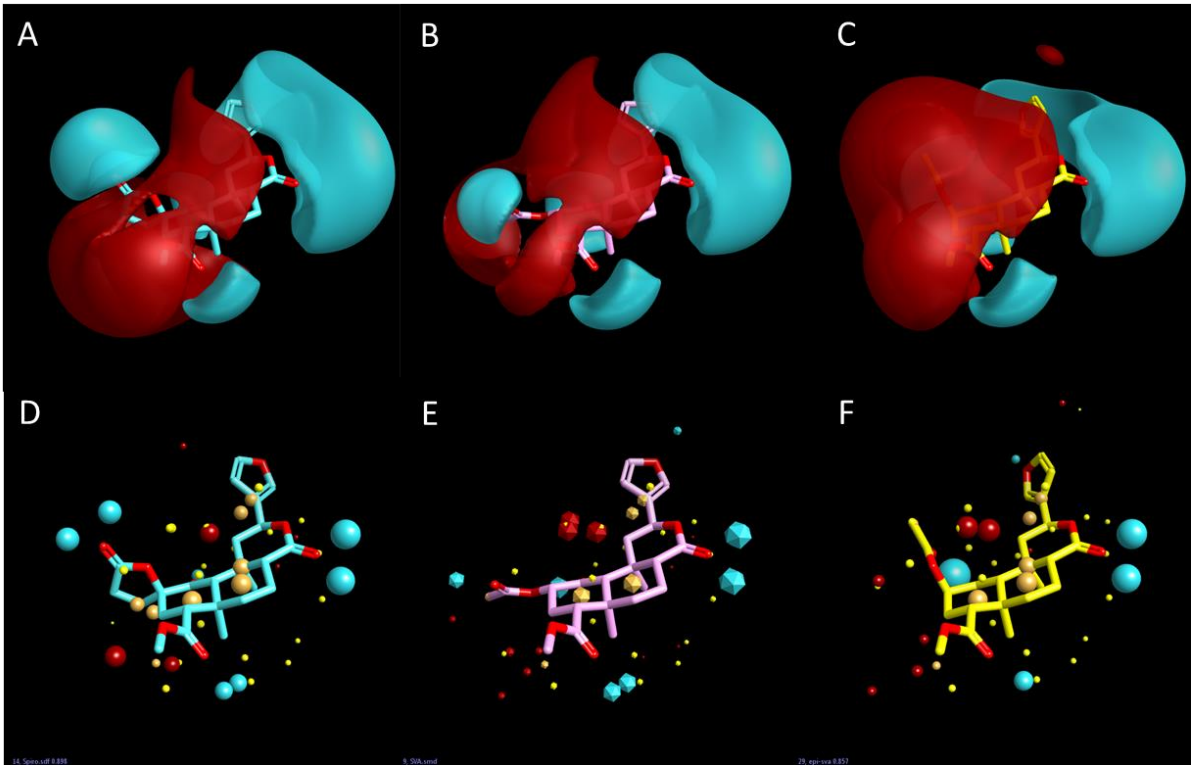
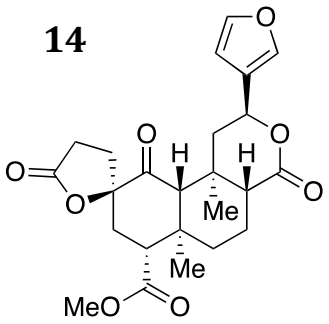
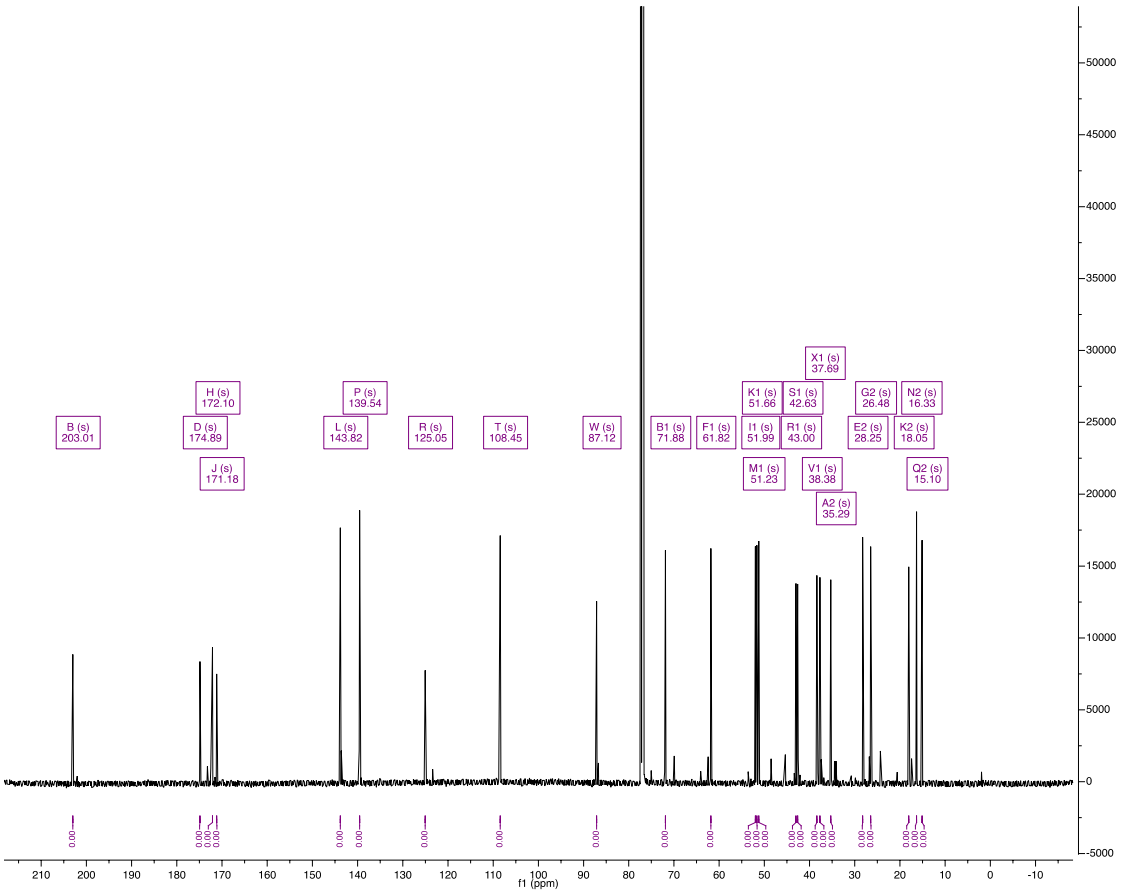


Figure S3. A-C) Surface view. Blue and red indicate negative and positive electrostatic surfaces, respectively. D-E) Field points of compounds. The size of the points indicates relative strength of interaction. Blue and red field points indicate negative and positive electrostatic potential, respectively. Gold and yellow points indicate regions favoring hydrophobic and van der Waals interactions, respectively. A & D) compound **14**, spirobutyrolactone; B & E) compound **9**, salvinorin A; C & F) compound **29**, 2-epi-salvinorin A.

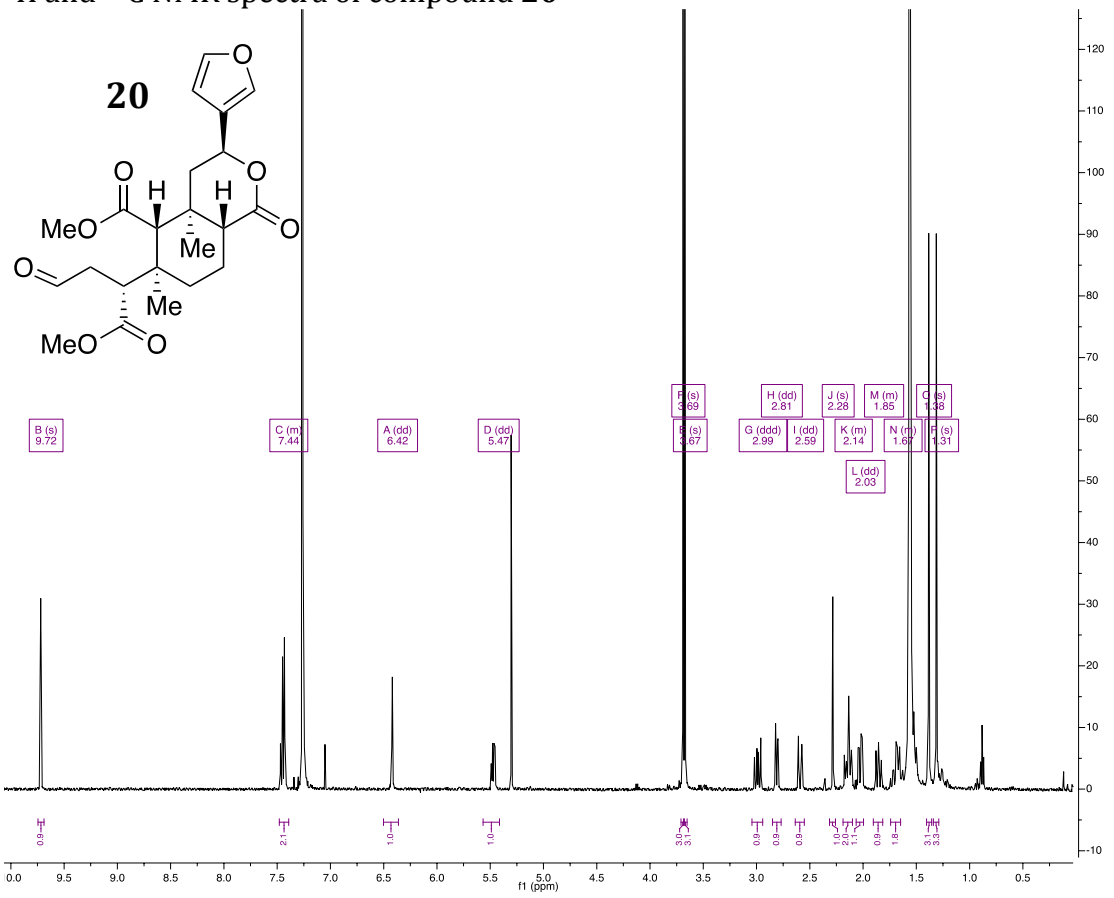
^{1}H and ^{13}C NMR spectra of compound **14**

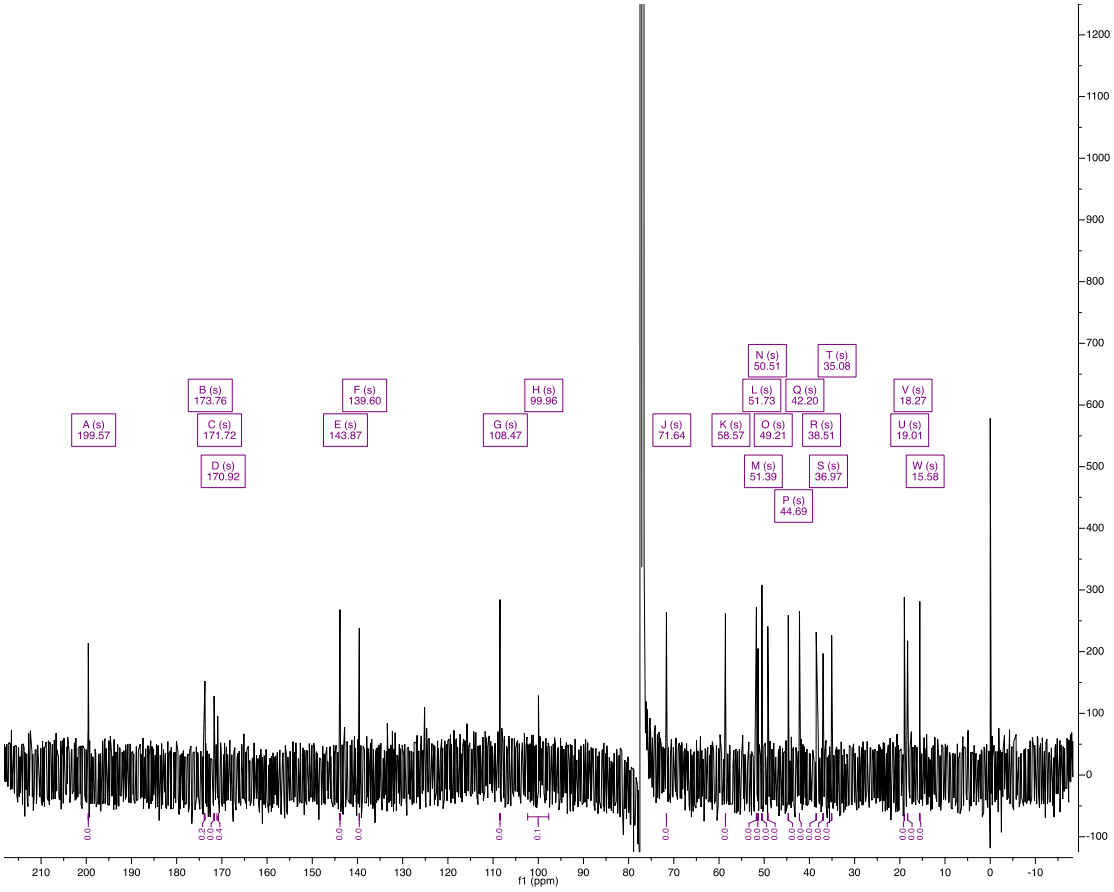
14





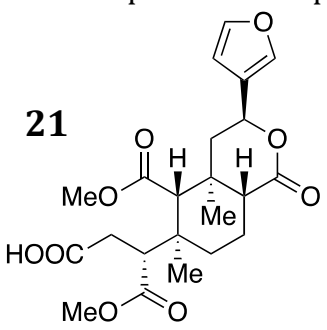
^{1}H and ^{13}C NMR spectra of compound **20**

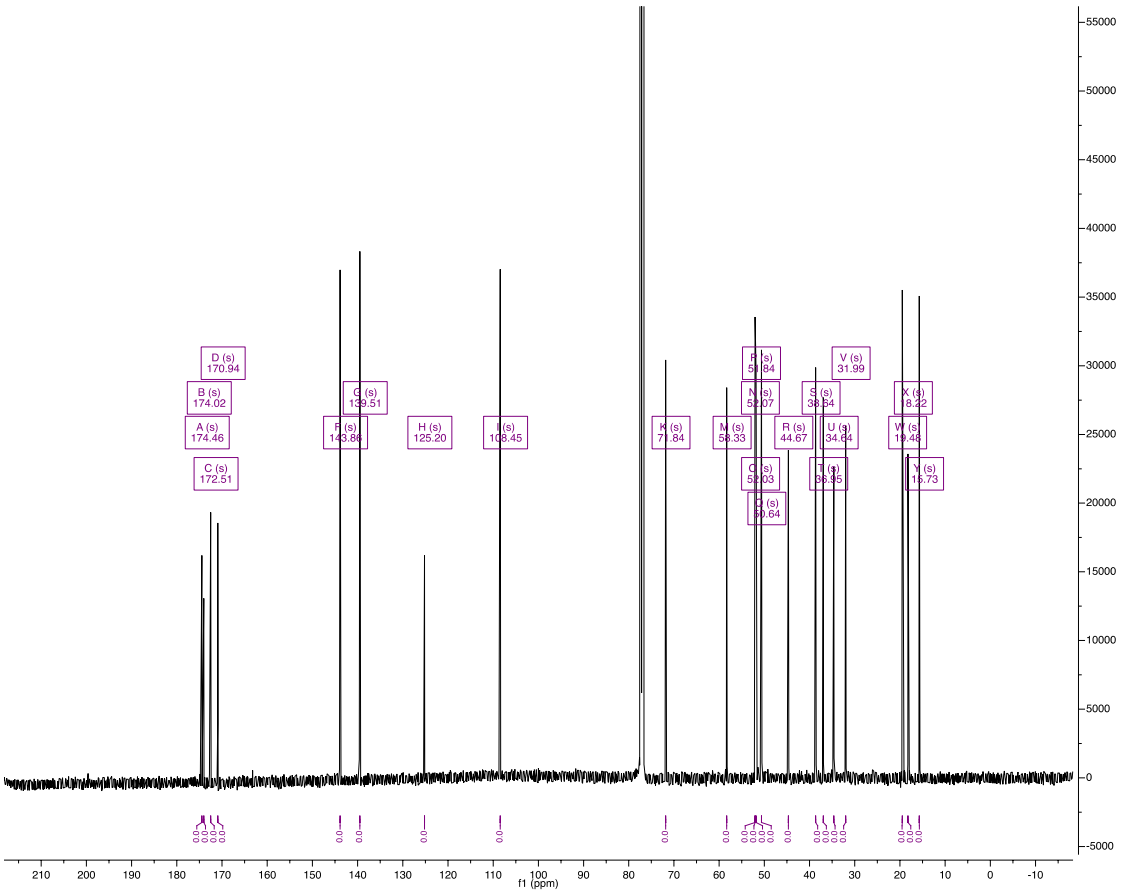




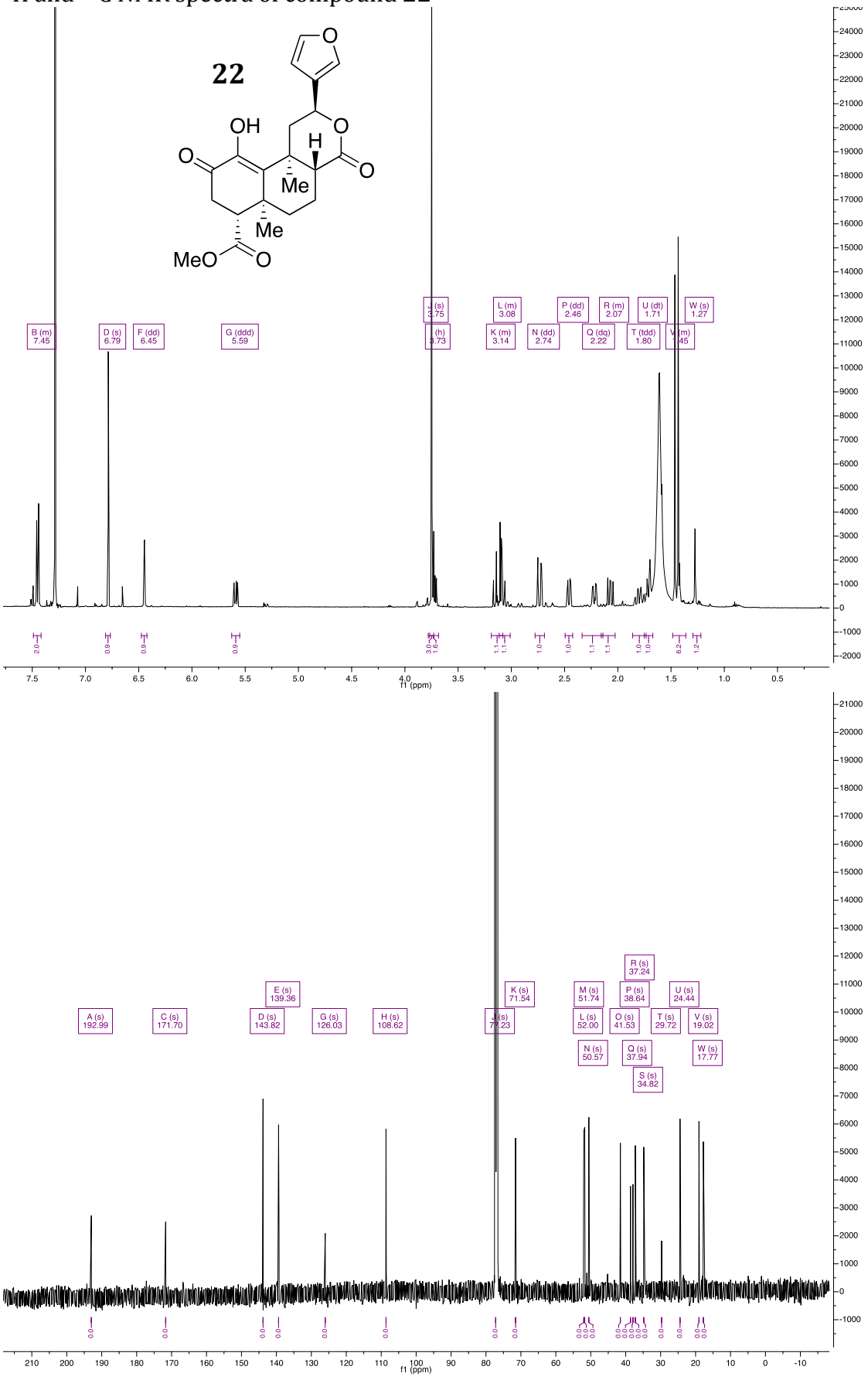
^{1}H and ^{13}C NMR spectra of compound **21**

21

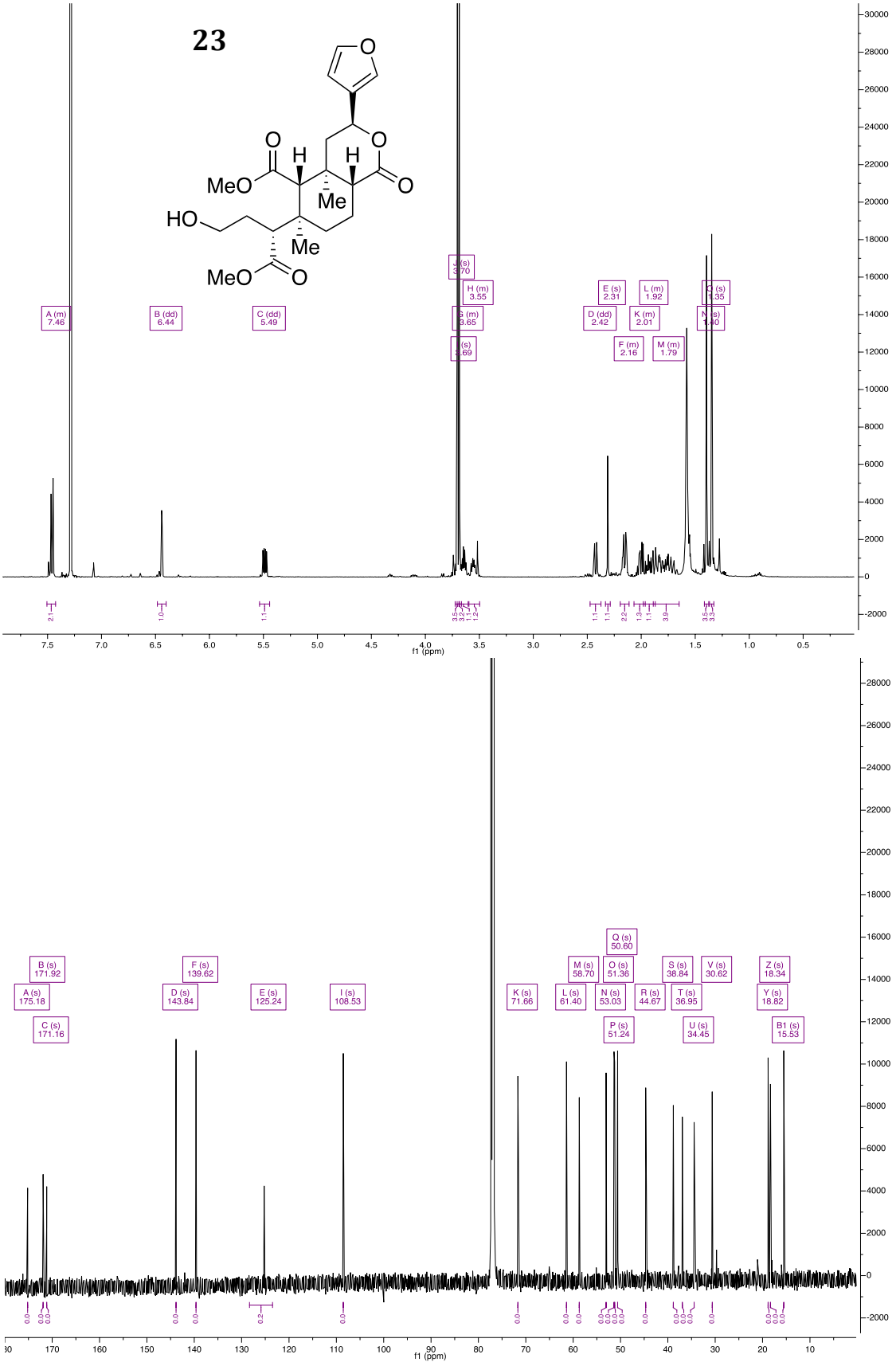




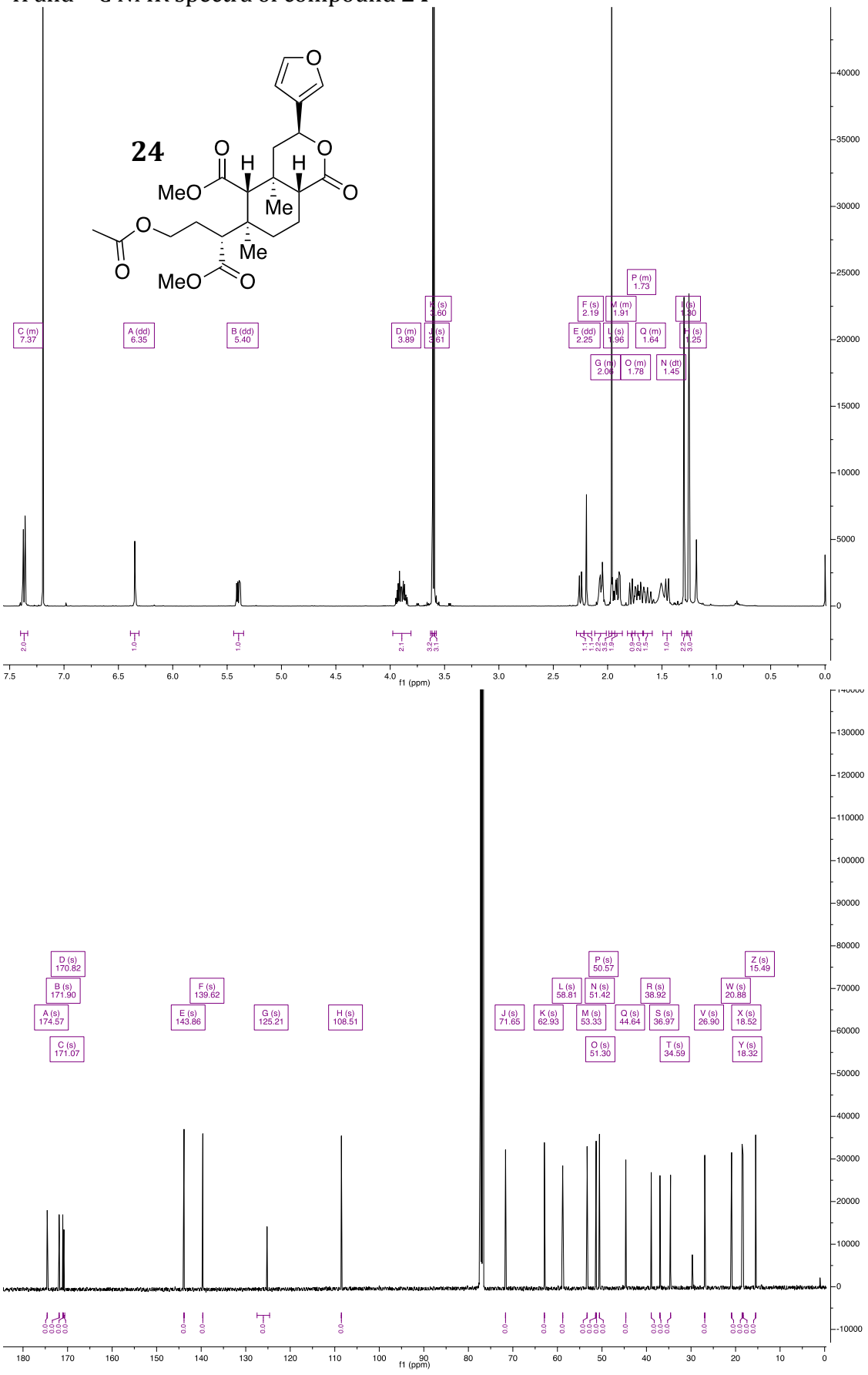
^{1}H and ^{13}C NMR spectra of compound **22**



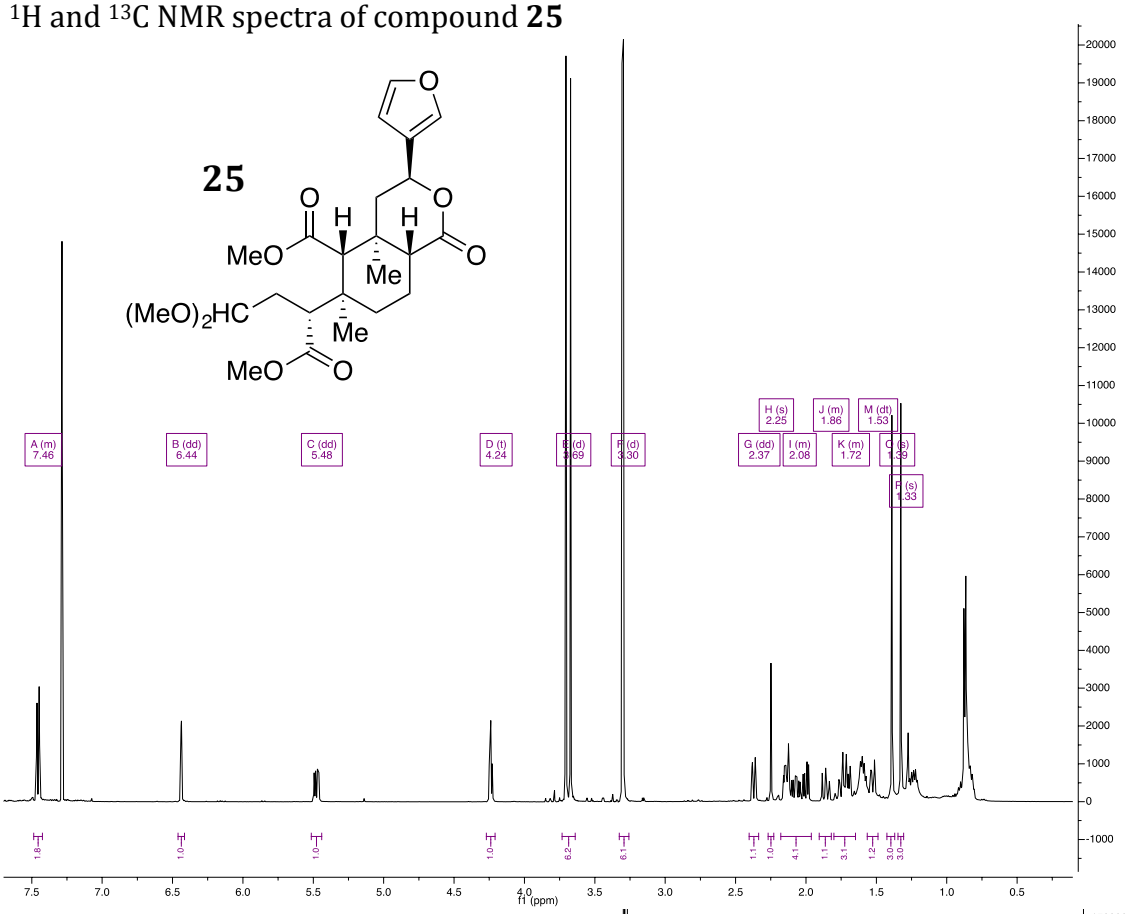
^{1}H and ^{13}C NMR spectra of compound **23**

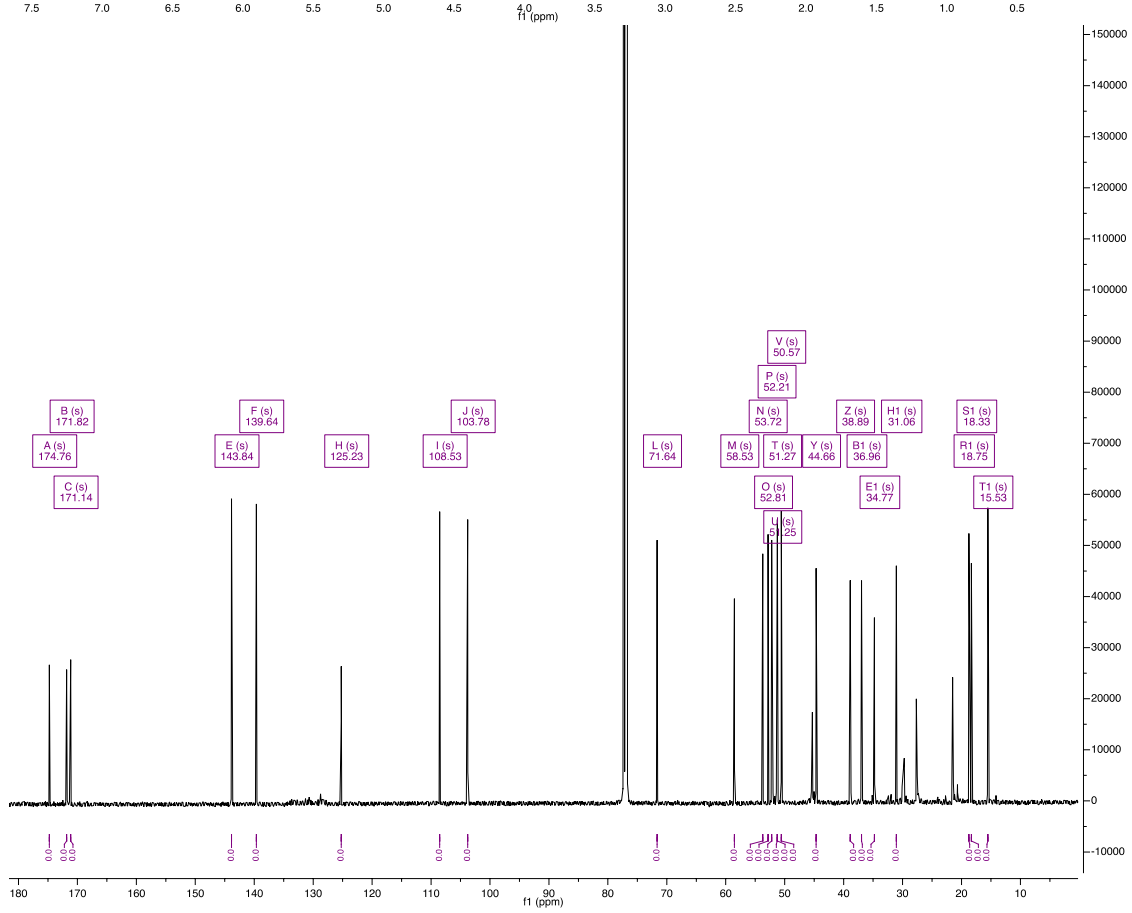


^{1}H and ^{13}C NMR spectra of compound **24**

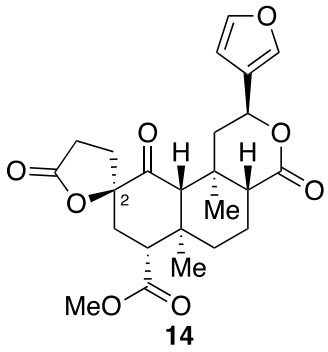


^{1}H and ^{13}C NMR spectra of compound **25**





Chromatogram and purity assessment for compound **14**



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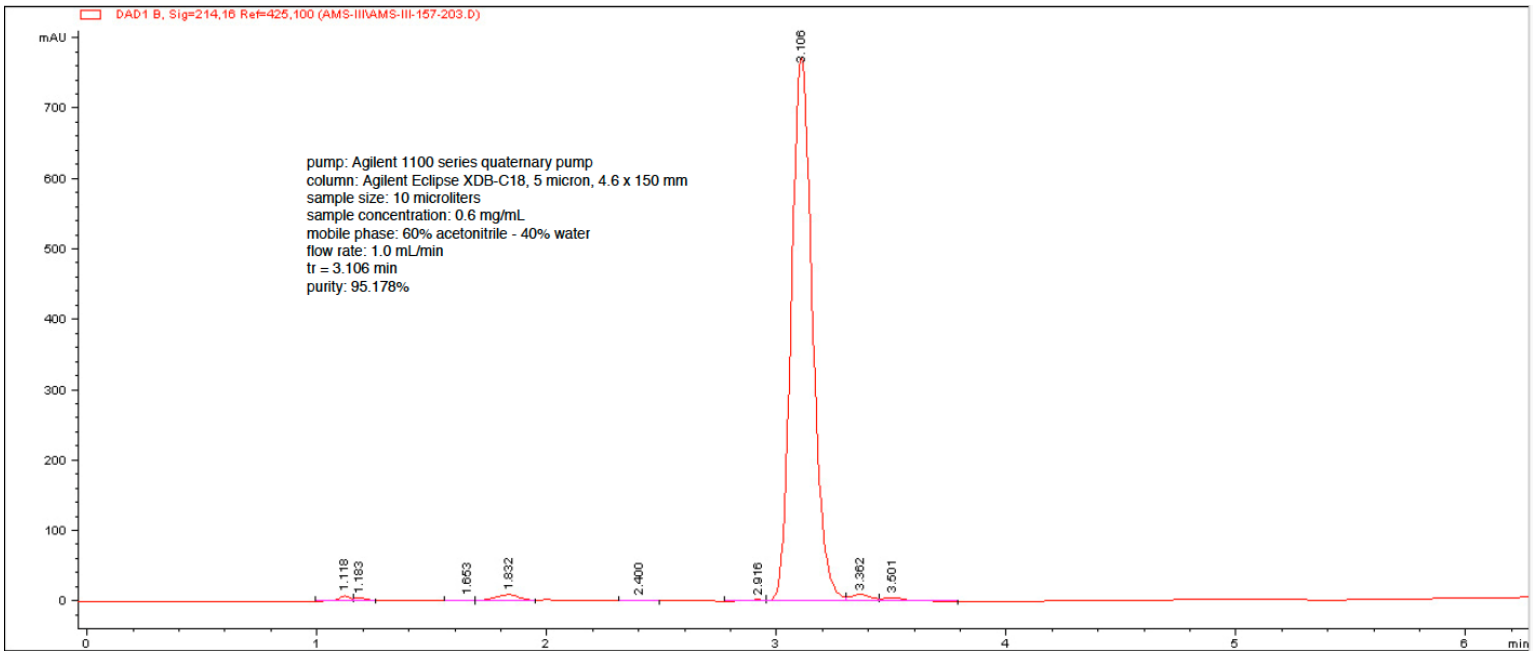
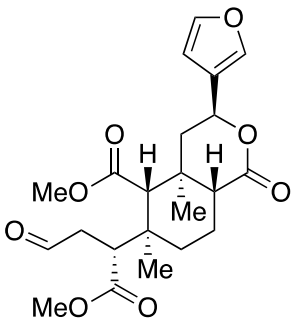


Chromatogram and purity assessment for compound **20**



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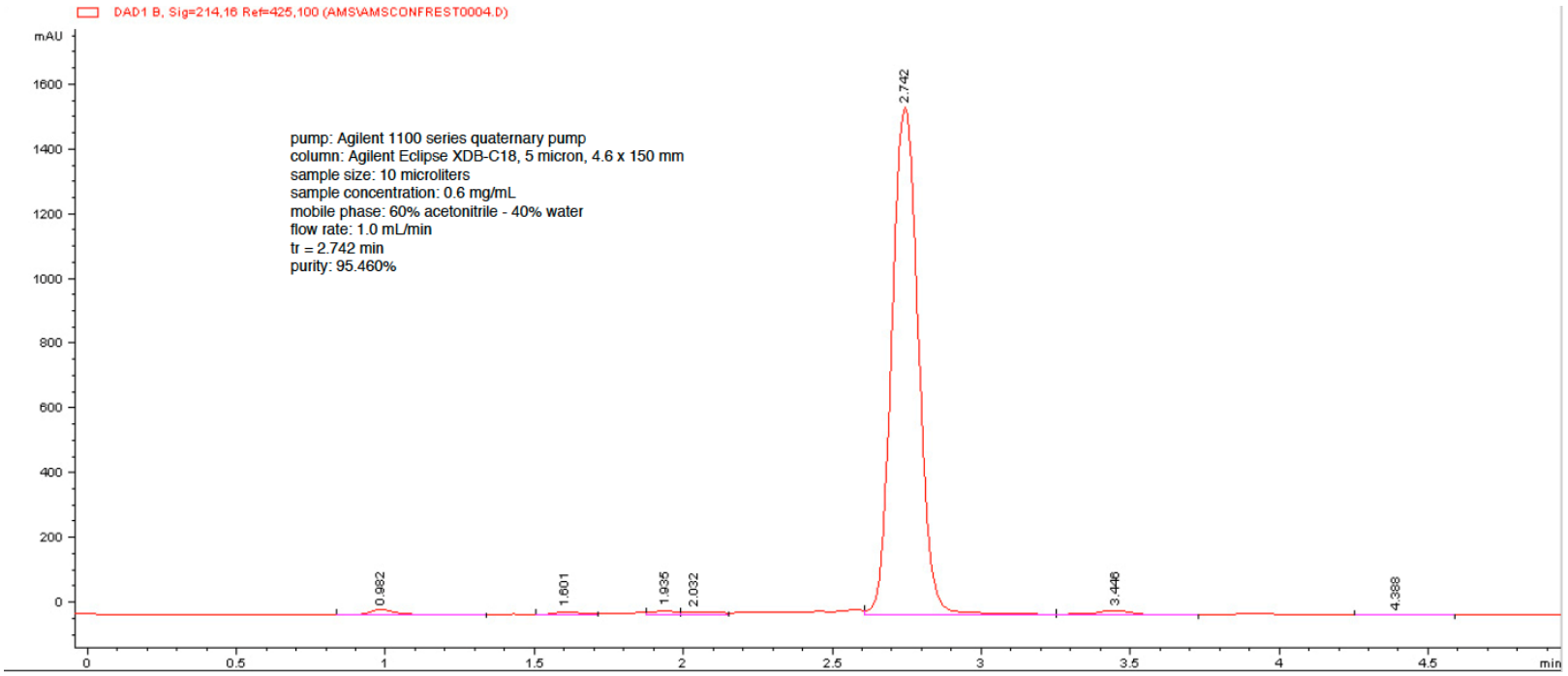
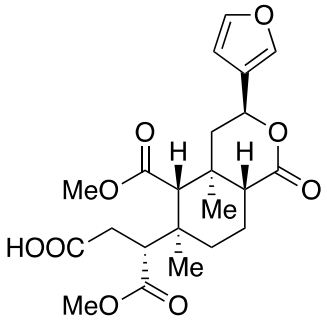


Chromatogram and purity assessment for compound **21**



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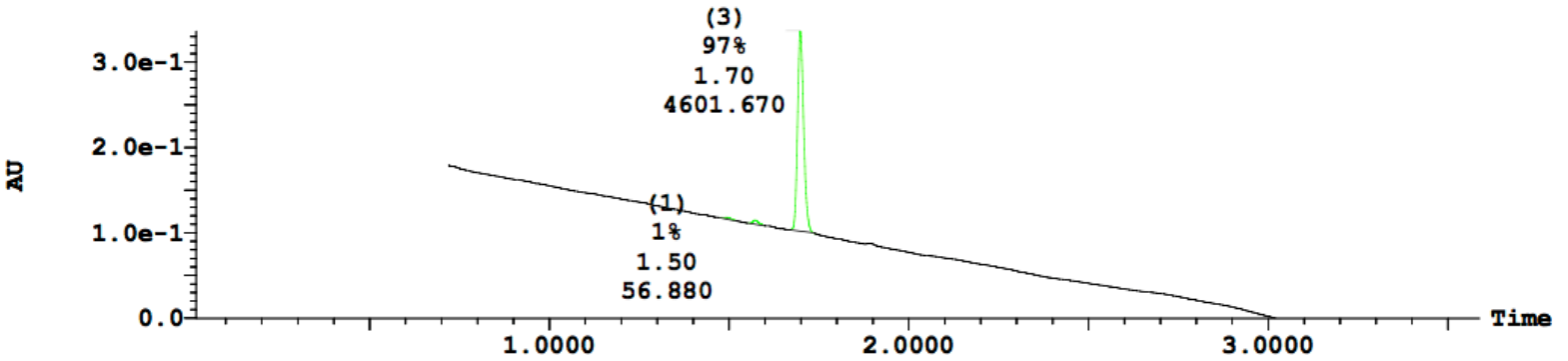

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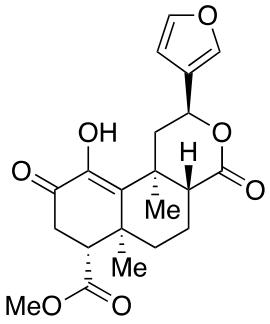
1	1.50	57	1.20	2241
2	1.57	93	1.96	4731
3	1.70	4602	96.84	234708

1: TOF MS ES+ :437.181 Smooth (SG, 2x3) **3.7e+004**

Chromatogram and purity assessment for compound **22**



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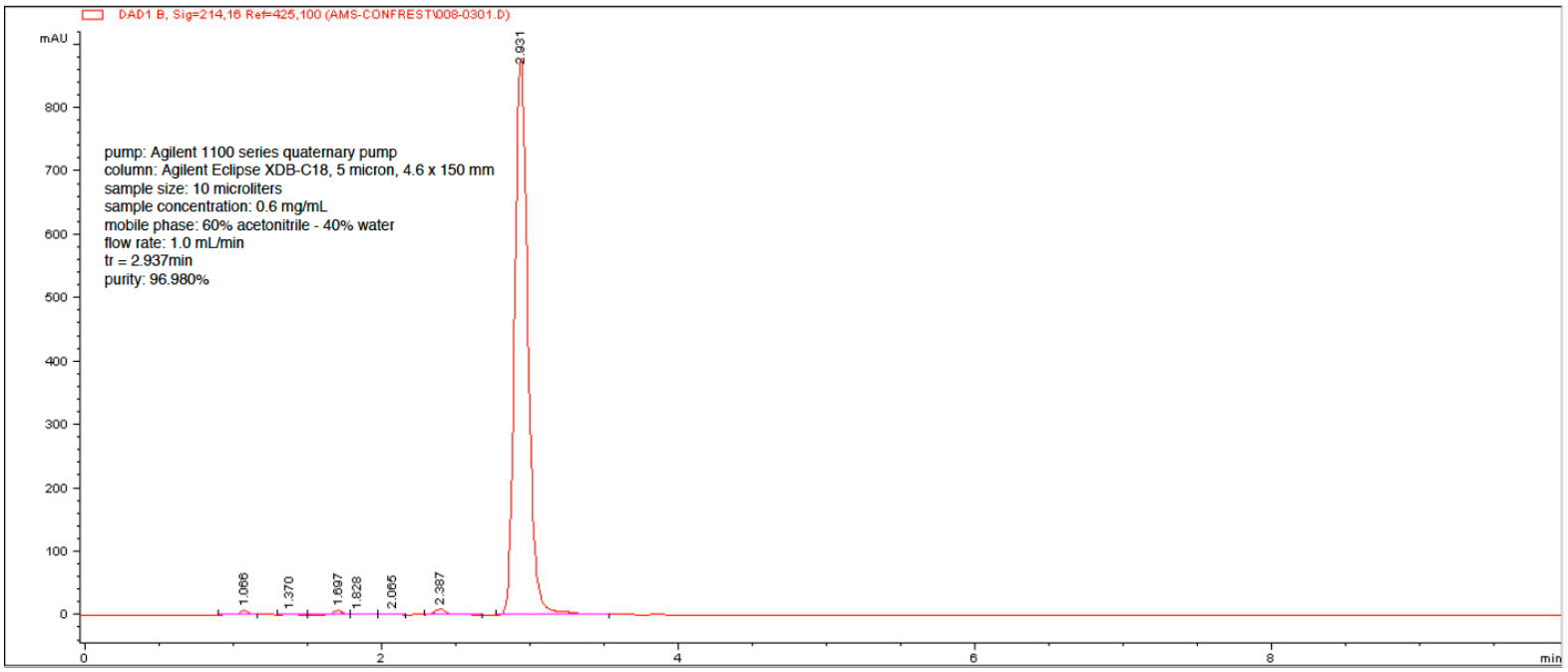
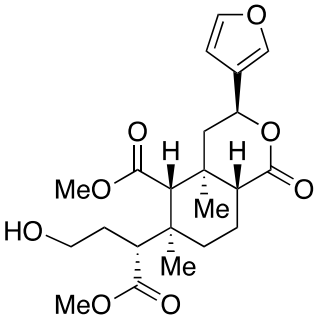
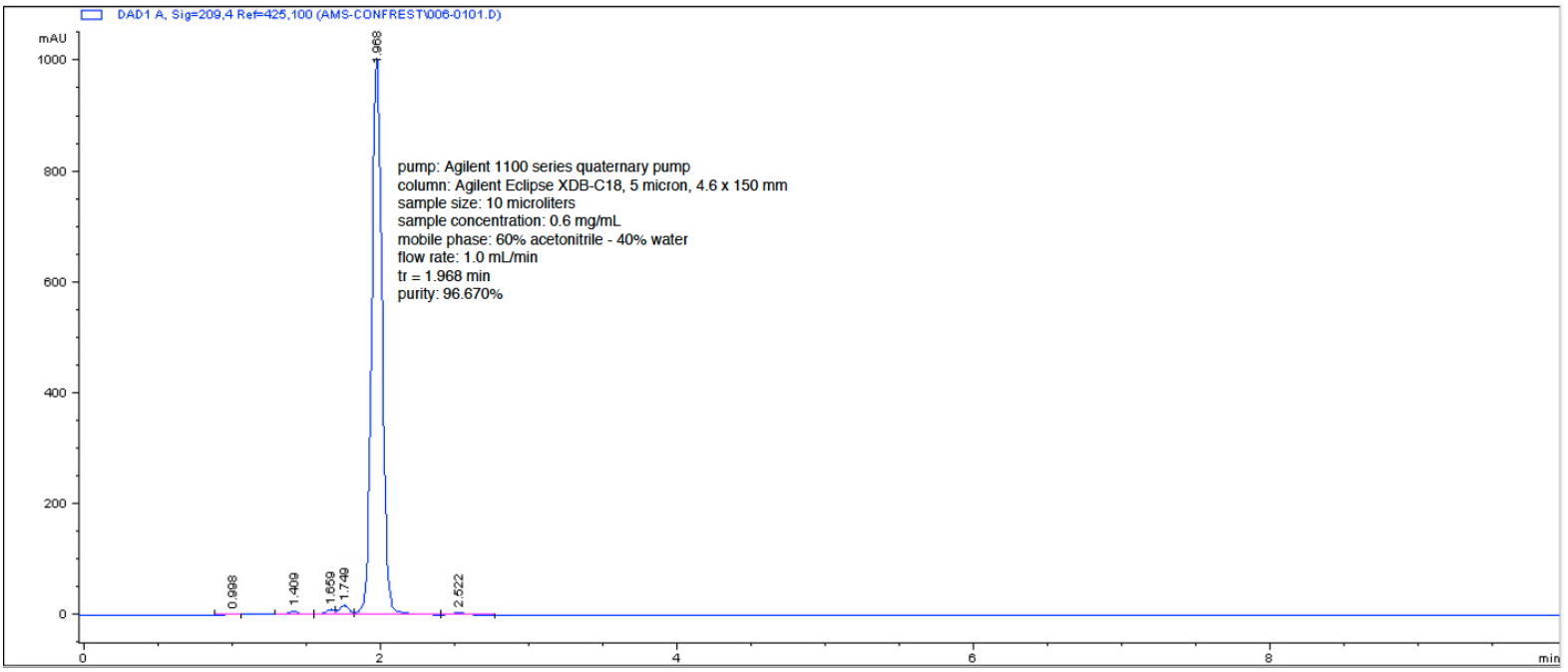


Chromatogram and purity assessment for compound **23**

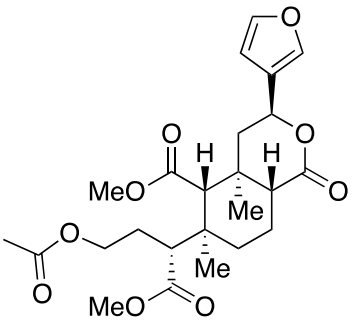


23

(flappy-SVB)



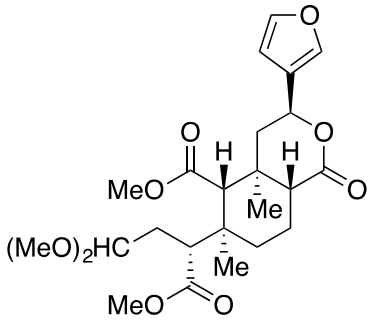
Chromatogram and purity assessment for compound **24**



24

(flappy-SVA)

Chromatogram and purity assessment for compound **25**



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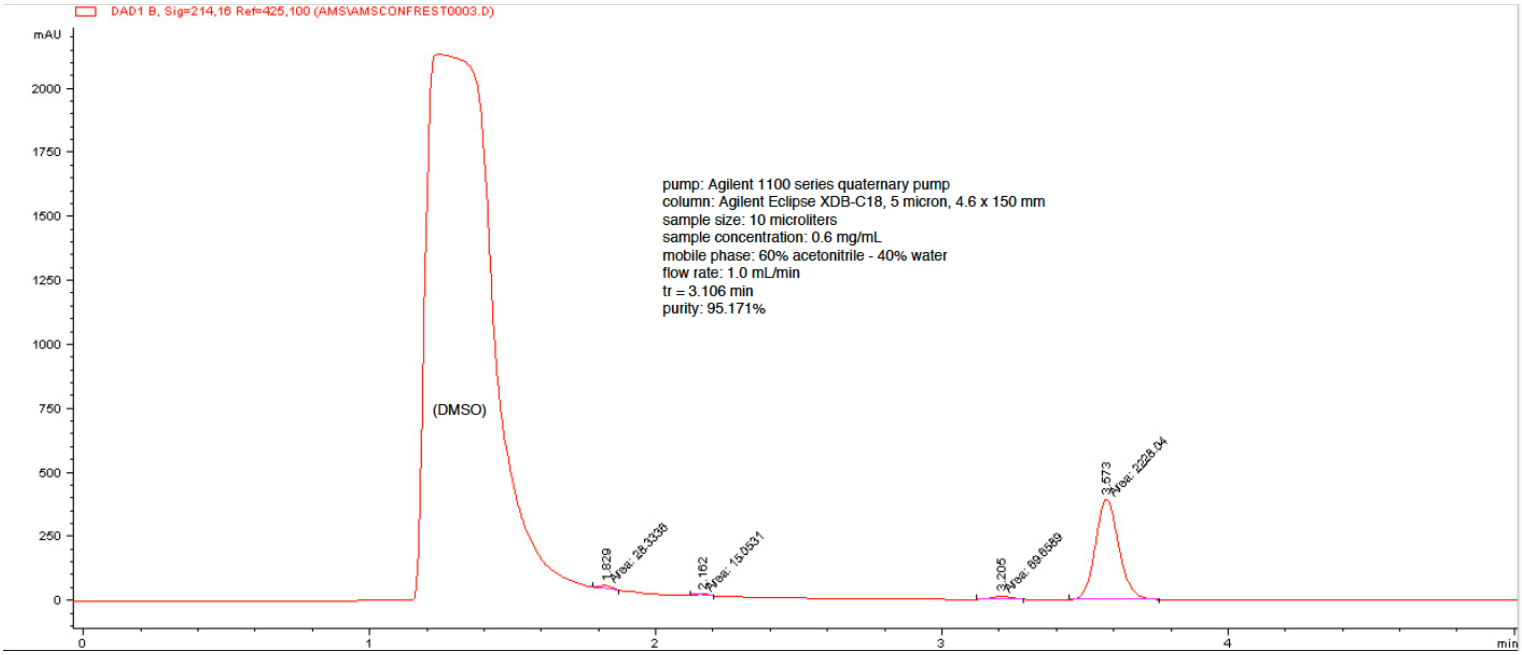
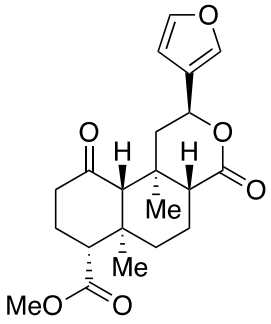


Chromatogram and purity assessment for compound **14**



11
(desacetoxy-SVA)