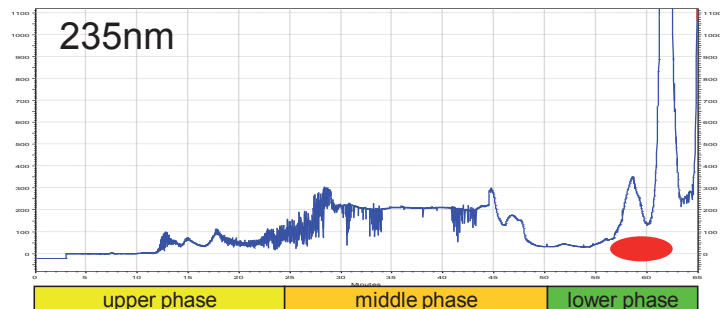
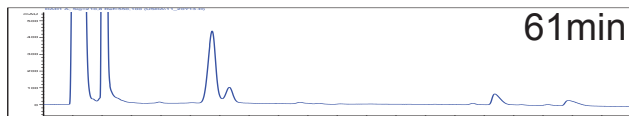
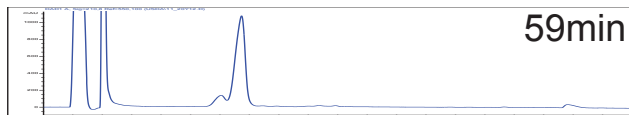
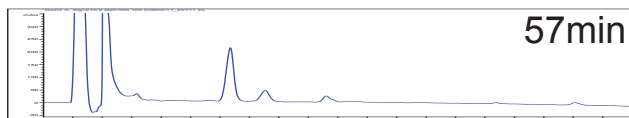
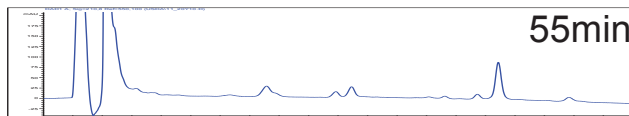
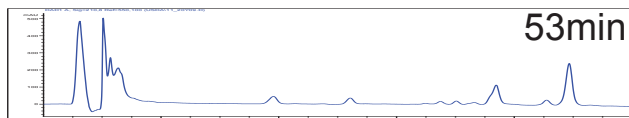
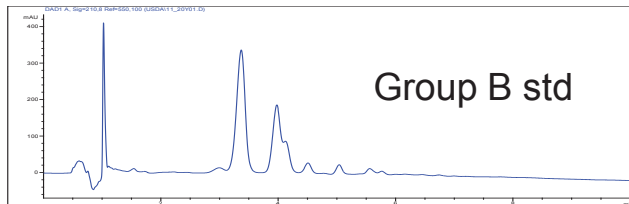


Saponin Separation – 3-Phase Solvent System

Hexane:MTBE:CH₃CN:0.1% aq. formic acid
(2:2:3:2); NP mode, elution as shown; 54mg analytical coil



HPCCC Conditions

Sample	Crude extract					
Concentration	54mg					
Solvent	UP/MP/LP 0.3/0.3/0.3					
Injection Volume	0.9					
Solvent System	hex:MTBE:ACCN:0.1%aq. FA (2:2:3:2)					
Solvent A	UP					
Solvent B	MP					
Solvent C	LP					
Solvent D						
Mixing Mode	premixed					
Pump	SSI					
Elution Flow Rate	1					
Extraction Flow Rate	2					
Flow Direction	P->C(NP)					
Method	3phase_FE					
	Time	%A	%B	%C	%D	Flow
	0	0	70	30	0	6
	7	0	70	30	0	6
	7.1	100	0	0	0	1
	20	100	0	0	0	1
Method	Time	%A	%B	%C	%D	Flow
	0	100	0	0	0	1
	25	100	0	0	0	1
	25.1	0	100	0	0	1
	50	0	100	0	0	1
	50.1	0	0	100	0	2
	65	0	0	100	0	2
Detection	235nm					
Coil Volume	25.5					
Instrument	SSI					
System Dead Volume	4					
Stationary Phase Displaced	12.5					
Sf	0.666667					
Bleed, Stripping?	F8to11	0.5				
	F11-14	1.7				
	F14to17	1				
	F17to20	1				
	F20to23	1				
	F23to26	0.8				
	F26to29	0.5				
	F29to end	0				
	total	6.5				
Sf at end	0.41					
Notes:	Not bad. Run should have been slightly longer, 66min, but material collected. Fractions conc to dryness, dissolved in 0.5mL for HPLC (seq USDA07An.s)					



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