

Customer presentation 2D-LC SEC to RP

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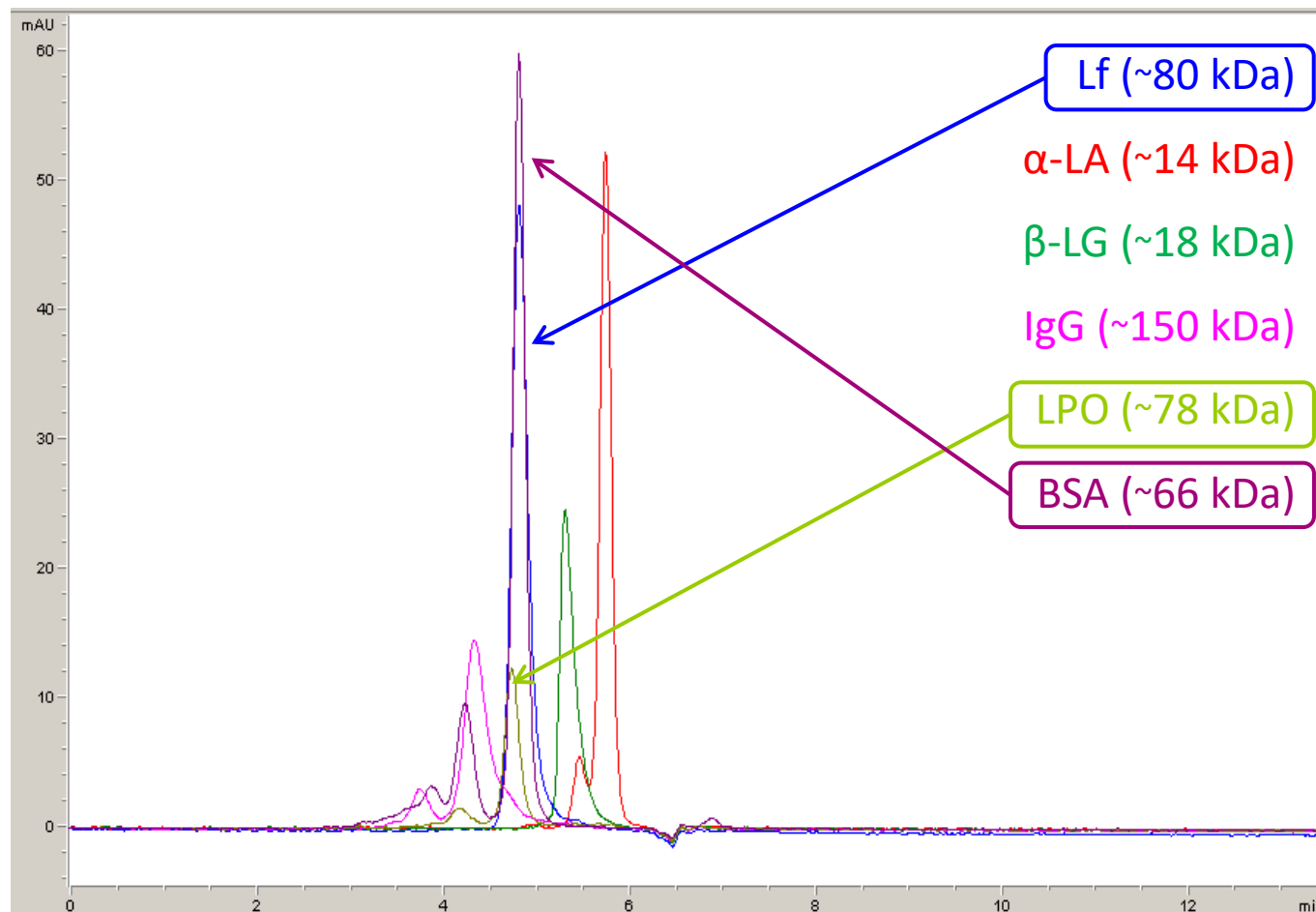
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About

- Zero waste project (circular economy)
- Innovative approaches for further processing of whey
- Fractionation and processing of whey proteins and exploitation of the residue for the formulation of new functional foods and food supplements
- The development of analytical methods for determination of main whey proteins → RP, SEC and IEX

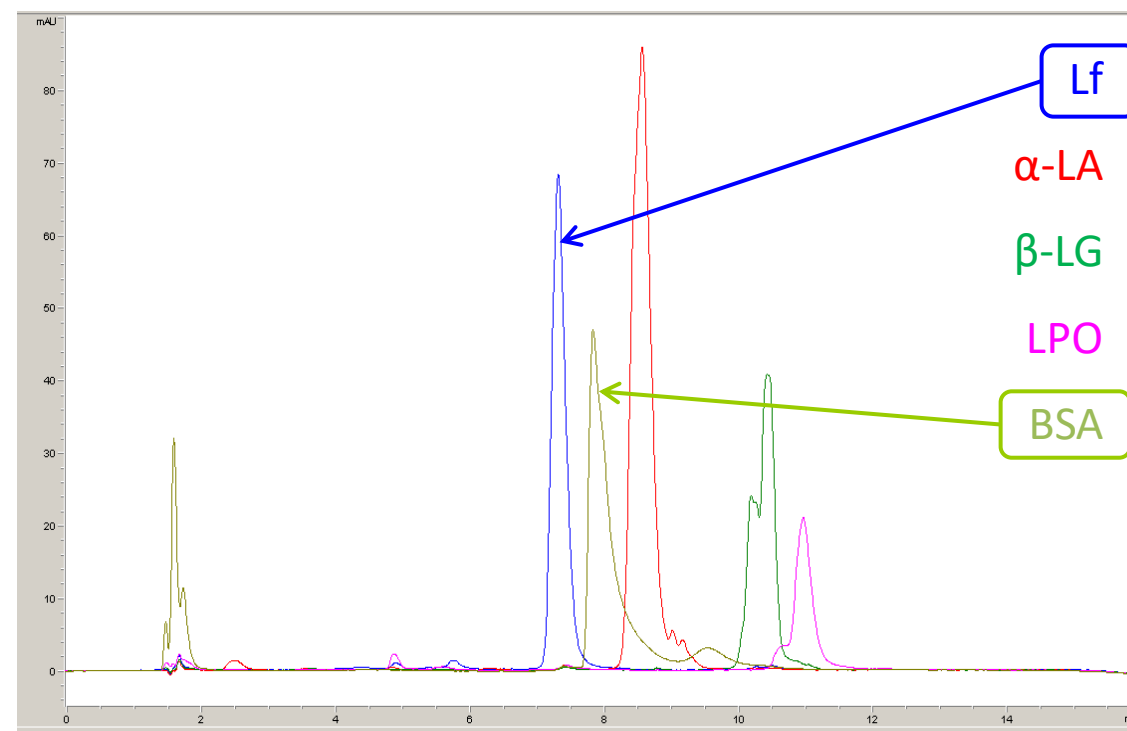
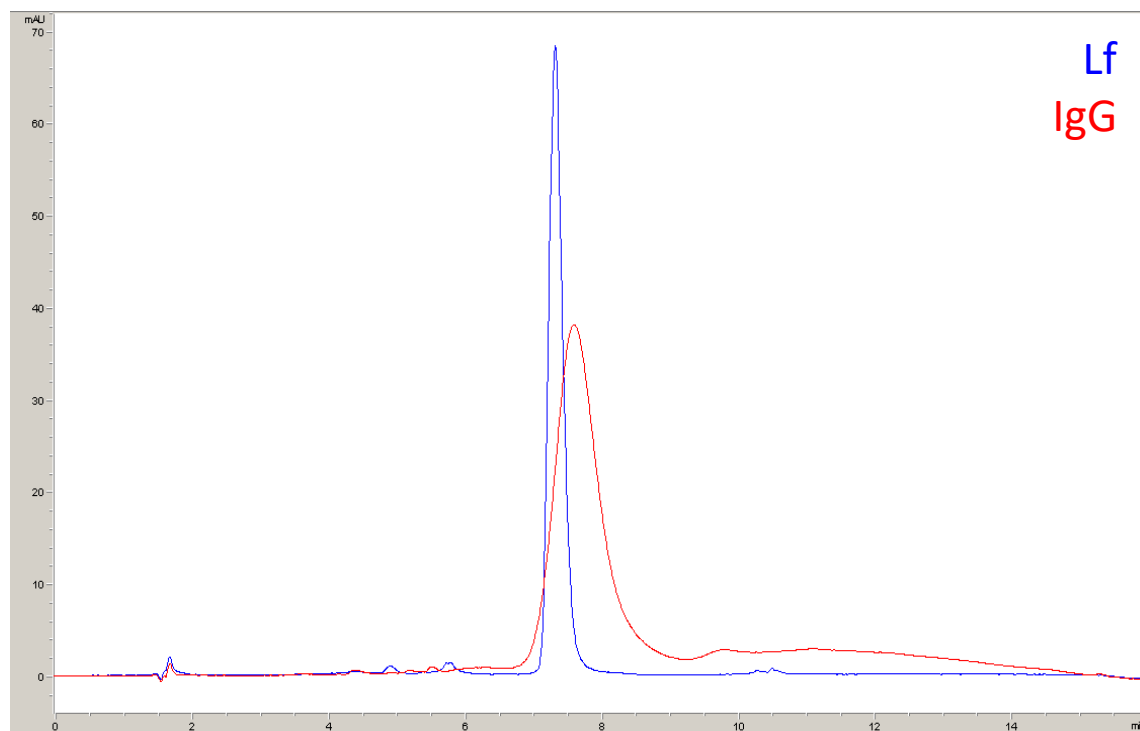
1D SEC method

- The basis for the 1st dimension of 2D-LC method



1D RP method

- Different RP stationary phases, mobile phases, column temperatures, gradient programs, flow rates
- Issues: detection of IgG, separation of BSA and LF



Method generation

- Inject on 1st D
- Start SEC elution
- 1st D separation
- Turn valve to capture peak
- Turn valve to continue
- Wash-off the solvents from 1st D
- Equilibrate 2nd D
- Inject on 2st D
- Start gradient elution
- 2nd D separation
- End gradient
- Equilibrate 2nd D

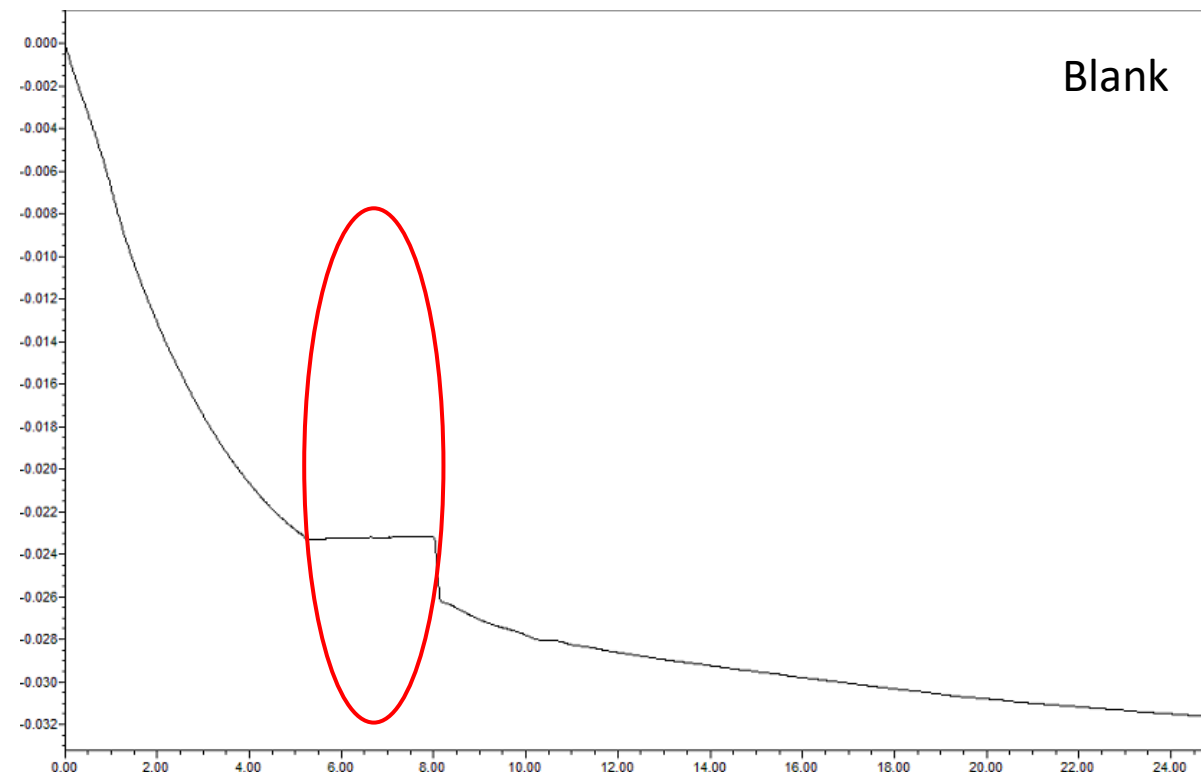
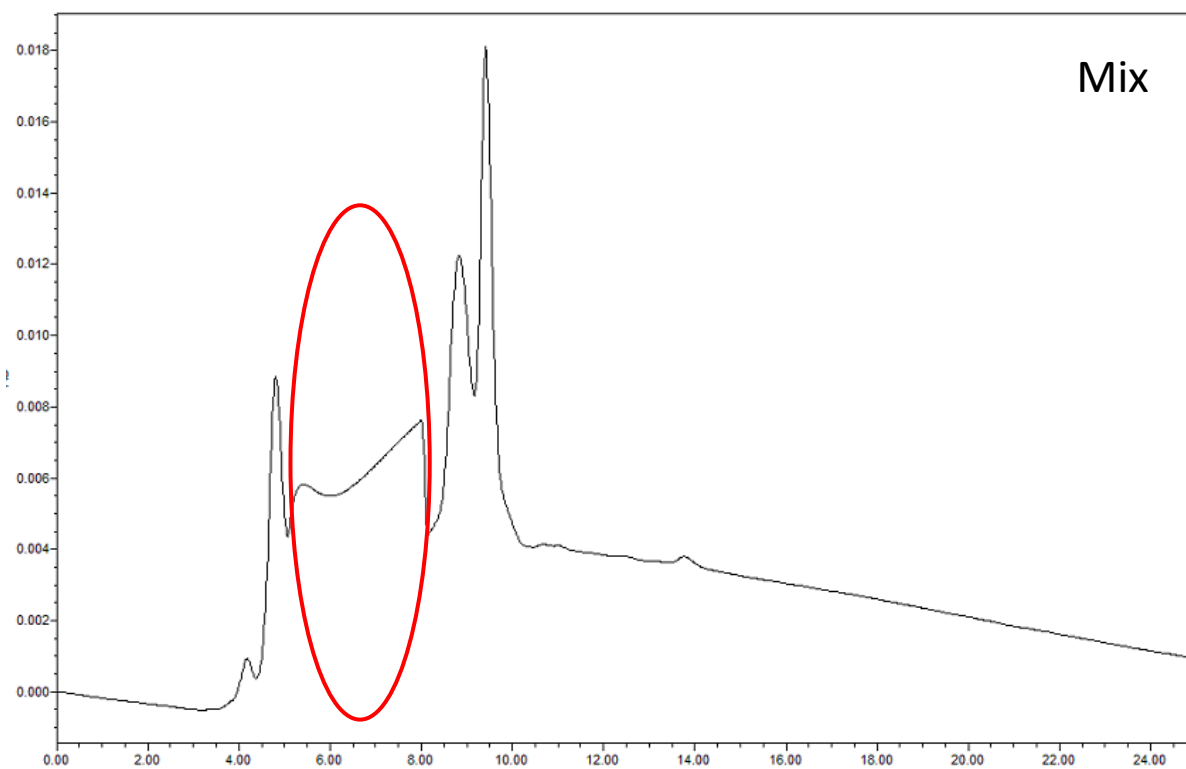
Method details

Dimension 1 - QSM				Dimension 2 - BSM			ISM	Valves	
Time	Flow	%A	%B	Flow	%A	%B	Flow	VL	VR
0	0.8	100	0	0.2	85	15	0	1	1
5	0.150	100	0	-	-	-	-	-	-
5.1	-	-	-	-	-	-	-	1	2
8	0.150	100	0	-	-	-	-	1	1
8.1	0.8	100	0	-	-	-	0.15	-	-
11	-	-	-	0.35	85	15	0.15	-	-
11.1	-	-	-	-	-	-	0	-	-
11.5	-	-	-	-	-	-	-	2	1
12	-	-	-	0.35	85	15	-	-	-
22	-	-	-	0.35	50	50	-	-	-
24	-	-	-	0.35	85	15	-	-	-

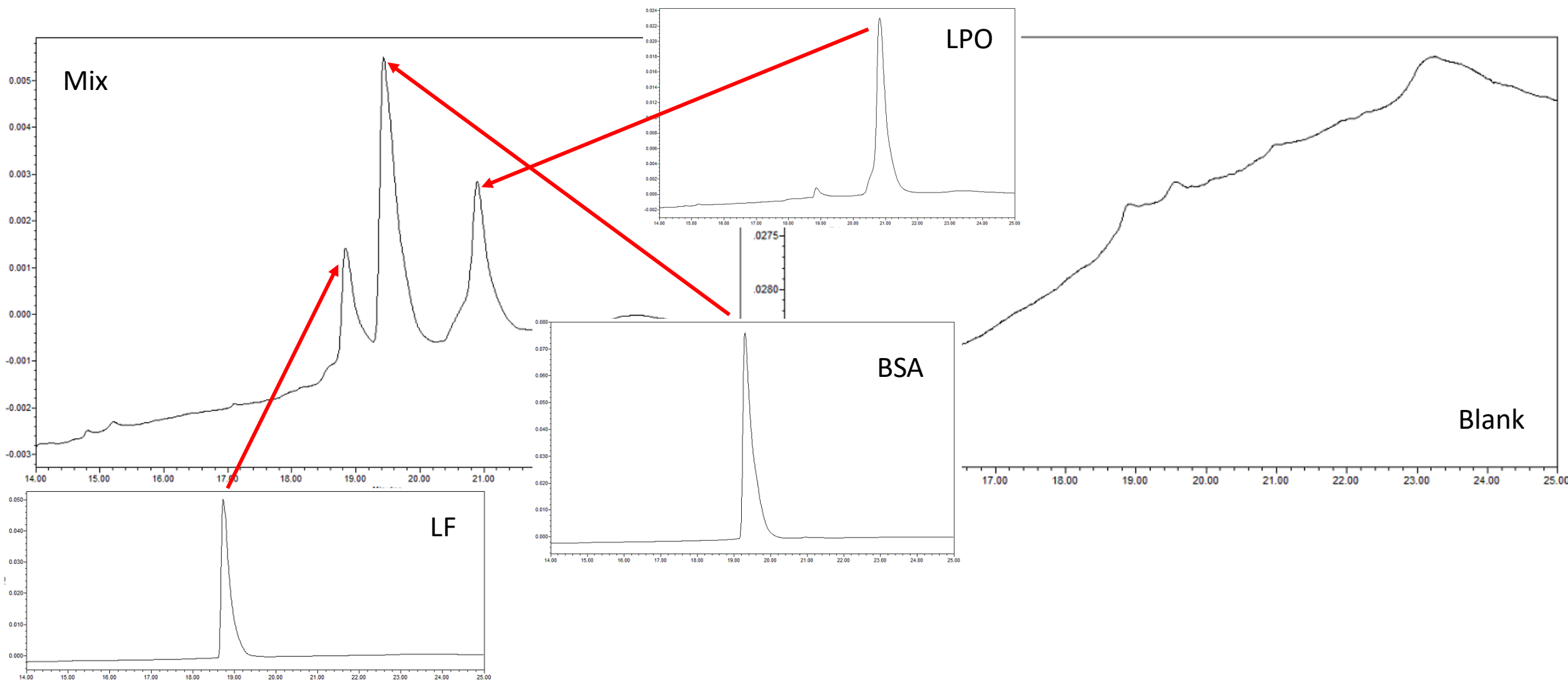
HC setup in practice

	Dimension 1
Column	XBridge Protein BEH SEC
Dimensions	7.8 x 150 mm, 3.5 μm , 200 Å
Mobile phase	(A) 0.2 NaP, 0.1 M NaCl, pH = 6,8
Temperature	Room temperature
Detection	PDA 210-400 nm
Flow	0.8 mL/min / 0.15 mL/min

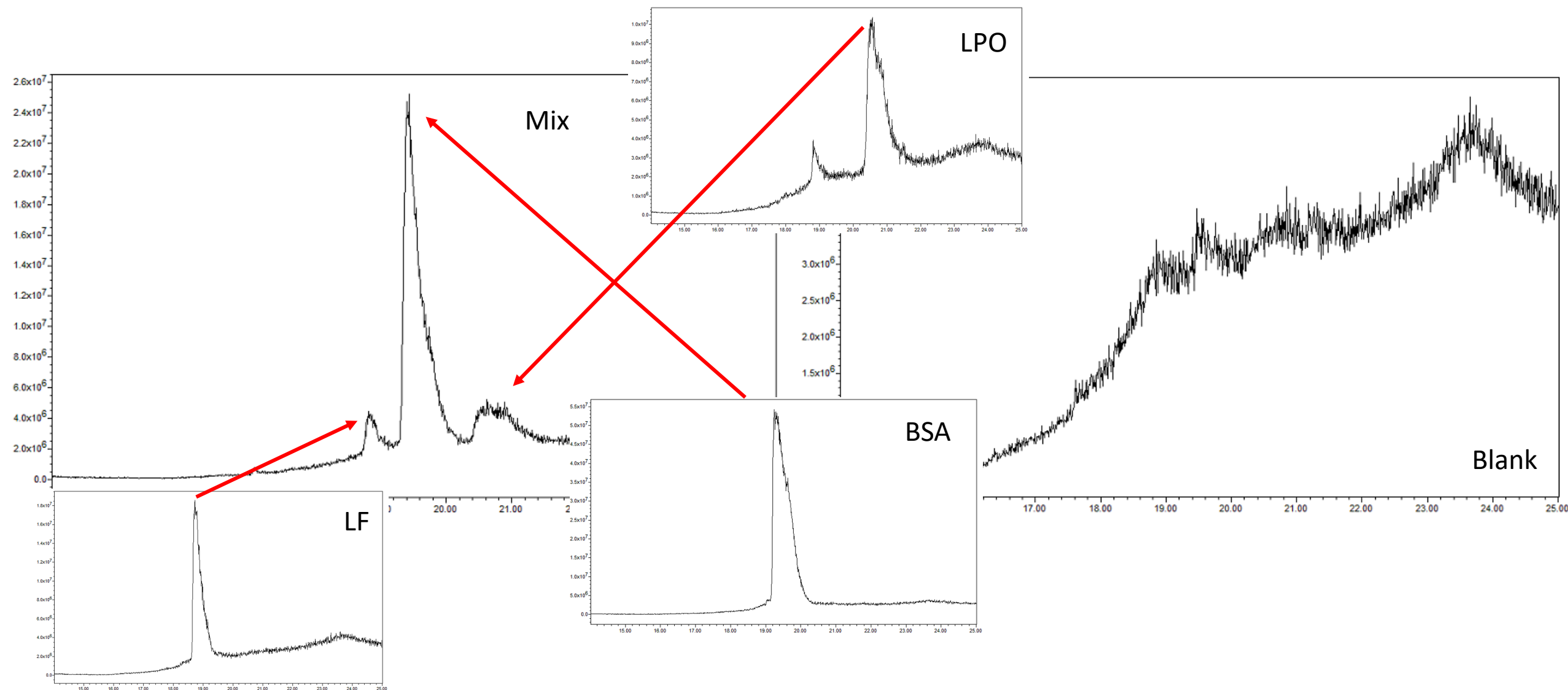
Starting chromatogram SEC



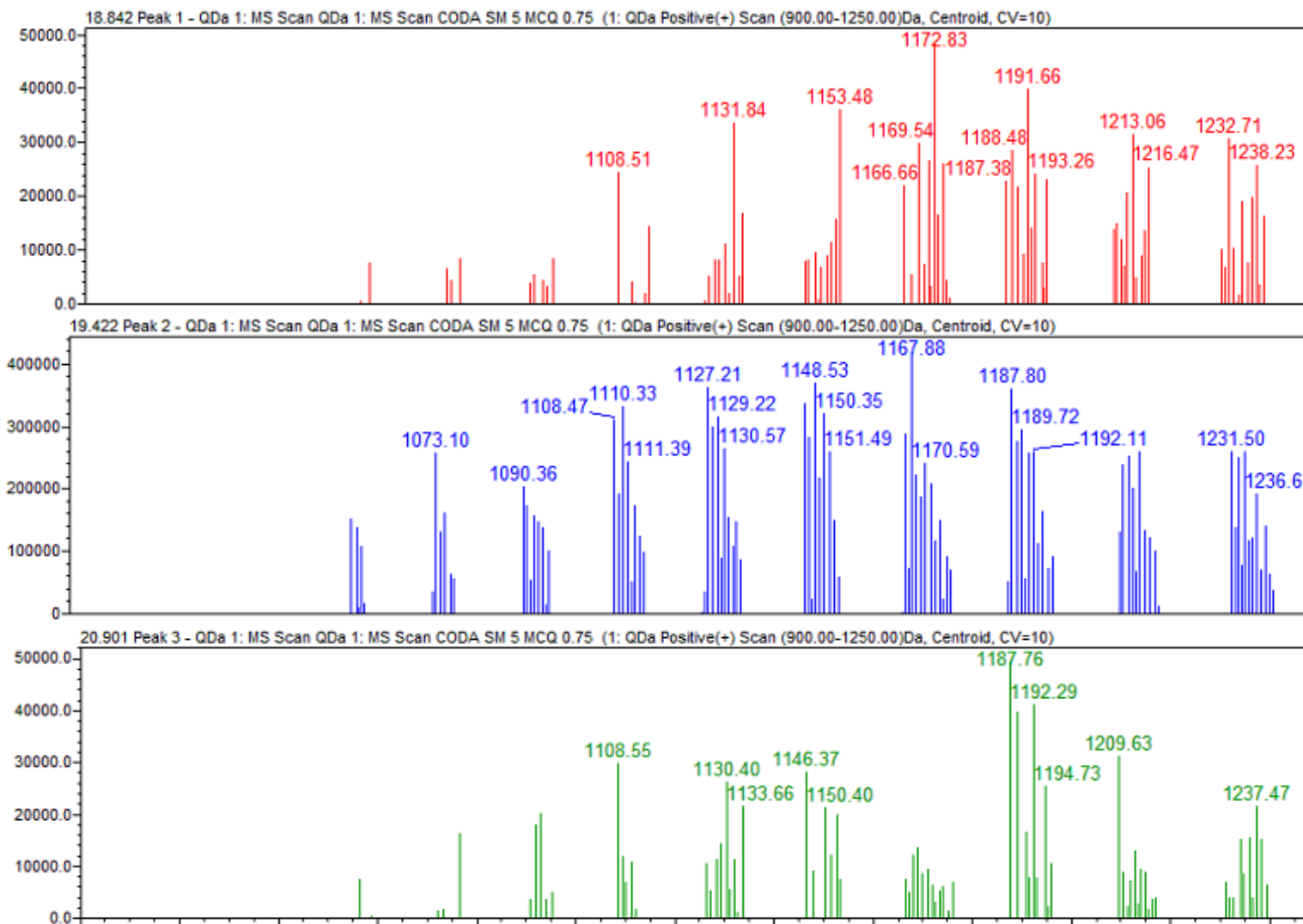
RP Dimension – UV @280 nm



RP Dimension – MS data



RP Dimension – MS data



Acknowledgment

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