LAAO Kinetiken für SAM, SAH, Methionin

1.1 Reaction solutions

1600

LAAO-Reaction-Mix (prepare fresh)	Final Volume: 4500 μl	
Add	Final concentration	
233 μl - 193 U/mL HRP	10 U/mL HRP	
90 μl - 5 mg/mL TMB-HCl	0.1 mg/mL TMB-HCl	
0.852 mL – 1.32 mg/mL LAAO	0.25 mg/mL LAAO	1.39
3325 μl Buffer (0.1 M Tris-HCl pH 7.5)		

1.2 Substrate concentrations

	1	2	3	4	5	6
	SAH		SAM		L-Met	
A	0	0	0	0	0	0
В	5	5	5	5	5	5
C	25	25	25	25	25	25
D	75	75	75 75		75	75
Ε	250	250	250	250	250	250
F	500	500	500	500	500	500
G	1250	1250	1250 1250		1250	1250
Н	2000	2000	2000	2000	2000	2000

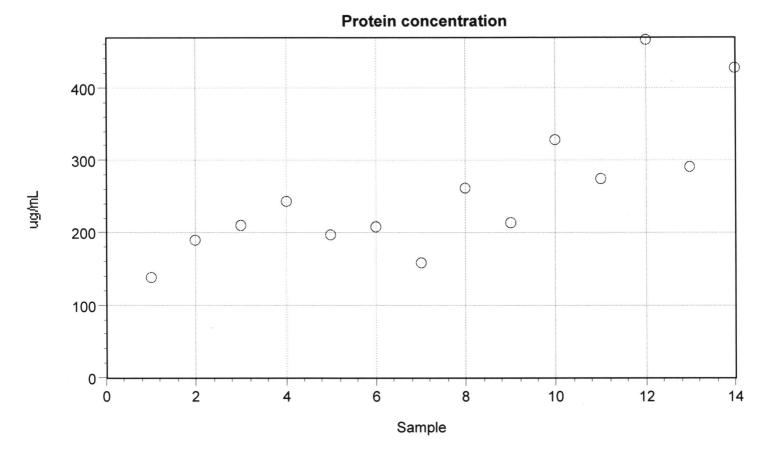
1.3 Pipetting

Stock solution (SAM, SAH, L-Met) are 2 mM in ddH₂O

Final concentration	Stock solution	ddH₂O	
[μM]	[μl]	[µl]	
0	-	100	
5	1 (500 μΜ)	99	
25	5 (500 μM)	95	
75	15 (500 μΜ)	85	
250	12.5	87.5	
500	100	300	
1250	62.5	37.5	
2000	100	-	

RHy 1:2.92 100pl fon 182pl H2U

Experiment#1



- O ÄKTA Fractions (Fractions: Sample# vs Concentration)
- ☐ Samples (Group01: Sample# vs Concentration)

Experiment#1

Fractions (I)

Sample	Wells	Sample#	Values	MeanValue	sd	Concentration
Fr01	A3	Sample#			0,007	Concentration
FIUT	A4	1	0,826	0,019	0,007	138,822
E-02	† · · · · · · · · · · · · · · · · · · ·	2	0,813	0.072	0.004	100 220
Fr02	B3	2	0,877	0,873	0,004	190,329
	B4	•	0,869		0 005	
Fr03 C3	1	3		0,893	0,005	209,374
4	C4		0,888			040054
Fr04	D3	4	0,930	0,927	0,003	242,654
	D4		0,924			
Fr05	E3	5	0,879	0,880	0,001	197,255
	E4		0,881		ļ	
Fr06	Fr06 F3	6	0,893	0,891	0,002	208,027
	F4		0,890			
Fr07	G3	7	0,845	0,840	0,004	159,069
	G4		0,836			
Fr08	Fr08 H3	8	0,893	0,948	0,055	262,228
	H4		1,003			
Fr09	Fr09 A5	9	0,892	0,898	0,006	214,520
	A6		0,904			
Fr10	Fr10 B5	10	1,013	1,016	0,004	328,403
	В6		1,020			
Fr11 C5 C6	11	0,967	0,960	0,007	274,203	
	C6		0,953			
Fr12 D5	12	1,173	1,159	0,014	465,563	
	D6		1,145			
Fr13	E5	13	0,988	0,979	0,010	292,093
	E6		0,969			
Fr14	F5	14		1,119	0,009	427,089
	F6		1,110			