

PROPOSAL No.: SC4282

Title:

Thermally stabilized proteins: in situ encapsulation

User Names:

Sartbaeva, Chen & Daekhie

Local Contact:

Rejeer Detten

Start Date and Time: 6/7/16 8:00

Total number of shifts:

6

End Date and Time:

File path:

SC4282/1d02/

10m setup

- 4 Lupo CellWheel
- 5 PUMA CellWheel
- 6 Empty Beam

1.5m setup

- 7 Lupo CellWheel
- 8 AgRe CellWheel
- 9 Empty CellWheel

[DISTANCE 1.5M]

INITIAL SAMPLES

- 1) AIR (empty) 00010 - 00020
- 2) water ✓ 00020 - 00030
- 3) Tris pH7 ✓ 00030 - 00040
- 4) Release buffer 00040 - 00050
- 5) Tris pH8 ✓ 00050 - 00060
- 6) 0.1M HCl ✓ 00060 - 00070
- 7) Lysozyme pH7 00070 - 00080
- " - " - " 00080 - 00090
- 8) Tris pH7 00100

acquisition time

0.1 seconds

* 0.01 seconds

* 0.01

* exposure SAME AS prev.

Lysozyme pH7 90-99 0.1s exposure

100 0.03s exposure

101 0.04

102 0.05

103 0.06

104 0.07

Tris pH7 buffer 105 0.03s exposure

114

Lysozyme 115 - 124 0.03 sec
 pre-hydro TEOS 125 - 134 0.1 sec
 TTCT 135 - 144 0.03 sec
 TTCTF 145 - 154 0.03 sec
 Insuline 155 - 164 0.03 sec

PROPOSAL No.: SC4282

Title:

Thermally stabilized proteins: in situ ensilication

User Names:

Sartbaeva, Chen & Doekhie

Local Contact:

Rajeev Dettner

Start Date and Time: 6/7/16 8:00

Total number of shifts:

6

End Date and Time:

File path:

SC4282/1d02/

10m setup

- 4 Lys Cell Wheel
- 5 PMA Cell Wheel
- 6 Empty Beam

1.5m setup

- 7 Lys Cell Wheel
- 8 Azide Cell Wheel
- 9 Empty Cell Wheel

INITIAL SAMPLES

1) AIR (empty)	00010-00020	acquisition time 0.1 seconds
2) water	00020-00030	
3) Tris pH 7	00030-00040	
4) Release buffer	00040-00050	
5) Tris pH 8	00050-00060	
6) 0.1M HCl	00060-00070	0.01 seconds
7) Lysozyme pH 7	00070-00080	
8) Tris pH 7	00080-00090	
	00105	* exposure SAME AS prev.

Lysozyme pH 7 90-99 0.1s exposure

100	0.03s exposure
101	0.04
102	0.05
103	0.06
104	0.07

Tris pH 7 buffer 105 0.03s exposure

Lysozyme	115-124	0.03 sec
pre-hydro TEOS	125-134	0.1 sec
TTCF	135-144	0.03 sec
TTCF	145-154	0.03 sec
Insulin	155-164	0.03 sec

[DISTANCE 1.5M]

Insulin	165-174	0.015 sec
Insulin	175-184	0.015 sec
Insulin	185-194	0.012 sec
0.1M HCl	195-204	0.015 sec

* SHUTTER TIME correction applied.

Insulin	205-206	0.015 sec
20 μ m Plo	207-211	inserted

Insulin	212-221	0.01 sec (10 sec sleep) → GOOD
Insulin	222-231	0.03 → NOT GOOD

— RERUN BUFFERS —

0.1M HCl 232-241 0.03

tandem shutter inserted & test run 242-277

Tris pH 7 buffer 278-287

0.1M HCl 288-297

Water 298-307

Tris pH 8 308-317

Relax Buff 328-327

Insulin 328-337 (10s delay time) — Tris pH 8.

L2-ensilication 338-347 0.01s exposure

" 348-357 0.01s exposure

" 358-367 0.4s exposure

TTCF-ensilication 368-377 0.4s "

378-387 1.5s "

Insulin-ensilicated 388-397 1.5 sec "

Tris pH 7 398-407 1.5 sec "

Insulin + chitosan 408-x — single frame (10 sec delay between frames)

[NP] 409-417

Chitosan 418-427
Tip-Sonicated

[DISTANCE - 10M]

TRIS PH 7 428-437 1.5 sec

48

NAME	#	-	#	NOTES
release buffer	428	-	447	
lysozyme ensilication	448	-	457	
TTCF ensilication	458	-	467	
insulin ^{chi} ensilication	468	-	477	
in situ lysozyme ensili	478	-	535	
	536	-		

- Slit settings changed. - use new backgrounds from here.
next run is 544 - ~~553~~

Tris pH 7	544 - 553	frames 9 0.05
lysozyme 1mg/ml	554 - 613	
Tris pH 8	614 - 623	
chi-in nano.	624 - 683	

chi-insulin ensilication 684 - 743

Tris pH 7	744 - 753	0.05
TTCF	754 - 798 redo	- 0.02 (BUBBLES in CAP)
	799 - 848	
TTCF ensilication	849 - 888	
1.5% TEOS	889 - 898	
0.1 M HCL	899 - 917	→ 899-917 - ERROR -
chi + insulin (formation NP)	918 - 962	
water	963 - 972	
water	973 - 982	
prehydrolysed TEOS	983 - 992	
Tris pH 7	993 - 1002	

a bit noisy, clean and run again

[move to 1.5 meters distance]

48	NAME	# - #	NOTES
	release buffer	438 - 447	
	lysozyme ensilication	448 - 457	
	TTCF ensilication	458 - 467	
	insulin ^{chi} ensilication	468 - 477	
	in situ lysozyme ensili	478 - 535	
		536 -	

- Slit settings changed. - use new backgrounds from here
next run is 544 - 553

Tris pH 7 544 - 553 frames 9 0.05

lysozyme 1mg/ml 554 - 613

Tris pH 8 614 - 623

chi-in nano. 624 - 683

chi-insulin ensilication 684 - 743

Tris pH 7 744 - 753 0.05

TTCF 754 - 798 - 0.02 (BUBBLES in cap)
redo

TTCF ensilication 844 - 888

1.50 TEOS
0.1 M HCL 889 - 898

Chi + insulin
(formation NP) 918 - 962

water 963 - 972

water 973 - 982

prehydrolysed TEOS 983 - 992

Tris pH 7 993 - 1002

[move to 1.5 meters distance]

use this	Tris pH 7	1003 - 1012	(perhaps too much beam - 1 sec exp)
	Tris pH 7	1013 - 1022	(with 3 sec delay) <u>aborted</u>
	Tris pH 7	1015 - 1024	(with 3 sec. delay) - Empty capillary
	release buffer	1025 - 1034	0.1 sec exp
	release - lys	1035 - 1044	0.5 exp
	" "	1045 - 1054	0.75 exp
	released TTCF	1055 - 1064	0.75
		1065 - 1074	1 sec (BUBBLES)
		1075 - 1084	1 sec
		1085 - 1094	0.05
		1115 - 1124	0.09
		1125 - 1134	0.12
		1135 - 1144	0.17
		1145 - 1154	0.25
		1155 - 1164	0.3 ←
		1165 - 1174	0.35
	release buffer	1175 - 1184	0.3 sec exp.
	released insulin	1185 - 1194	0.01 sec exp
	lysozyme 1mg/ml	1195 - 1204	
		1205 - 1264	0.01 sec 60 fr / 60 delay.
	lysozyme ensilication		
	Tris buffer	1265 - 1274	0.01 (9 at 1 sec.)
	lysozyme ensilication 1mg/ml	1275 - 1334	
	Tris pH 7	1335 - 1344	1 sec (a bit dirty, clean again)
		1345 - 1354	1 sec (still dirty)
		1355 - 1364	1 sec (still)
		1365 - 1374	Cancel washed NaOH + ethanol + water
		1375 - 1384	1 sec washed NaOH + water
		1385 - 1394	1 sec washed only Ethanol
		1395 - 1404	1 sec washed NaOH + Tris pH 7
		1405 - 1414	1 sec
		1415 - 1424	1
		1425 - 1434	-1
		1435 - 1436	-1

a bit noisy, clean and run again

0	TJCF	1445 - 1504	0.01 sec	60 fr / 60 delay
	TTCF ensucation	1505 - 1564		
	Tris pH 8	1565 - 1575		not good, contaminate
	Tris pH 8	1576 - 1586		not good contaminate
	NaOH	1587 - 1597		leave NaOH for 10 min,
	Tris pH 8	1598 - 1608		good
	chitosan - insulin nano-ensucation	1609 - 1668		
	NaOH	1669 - 1678		
	release lysozyme	1679 - 1688	0.01	
	released lysozyme	1690 - 1699	0.03	
	released TJCF	1700 - 1709	0.01	
	released TTG	1710 - 1719	0.03	
	released insulin	1720 - 1729	0.01 (10 sec sleep)	
		1730 - 1739		
		1740 - 1749		
		1750 - 1759		
		1760 - 1769	0.05	
		1770 - 1779	0.1	
		1780 - 1789	0.3	
		1790 - 1799	0.1 sec	
	release buffer	1773 - 1782	0.3 sec.	

STATIONARY CAPILLARY TACK 1783 - 2722

release buffer	2723 - 2732	1 sec	[Data 04]
Tris pH 7			[Elder]
lysozyme 2mg/ml ensucation	2733 - 2792	0.01 sec	60 0.01 60
- Tris pH 7	2793 - 2804	1 sec	
lysozyme 2mg/ml	2805 - 2813	0.01 sec	16 0.01 -

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fr exp del.

Chi-Insulin-ensil-A 3059-3078

20 0.01 5

Tris pH 7 3079

1 1 ~

~~Insulin~~ Lysozyme pH 8 3080-3119

20

Tris pH 7 3120

1 1 -

TTCF ensil. 1:20
(immob) 10M

3121-3170

50 0.02 20

Tris pH 7 3171-3180

10 1 -

WATER

3181-3190

10 1

TTCF 1:100 ~~immob~~ 3191-3240

50 0.02 20

MOVE DETECTOR TO 1.5 M

Tris pH 7 3241-3250

10 1 -

TTCF 1:20 1mm 3251-3310

60 0.02 20

Tris pH 7 3311-3320

10 1 -

TTCF 1:100 1mm 3321-3380

60 0.02 20

Tris pH 7 3381-3400

10 1 -

TTCF 1:100 1mm
pH 6 3391-3450

60 0.02 20

~~Tris pH 7~~
TTCF pH 8 3451-3460
3570

60 0.02 20

Tris pH 7 3571-3580

10 1 -

Tris pH 8 3581-3590

10 1 -