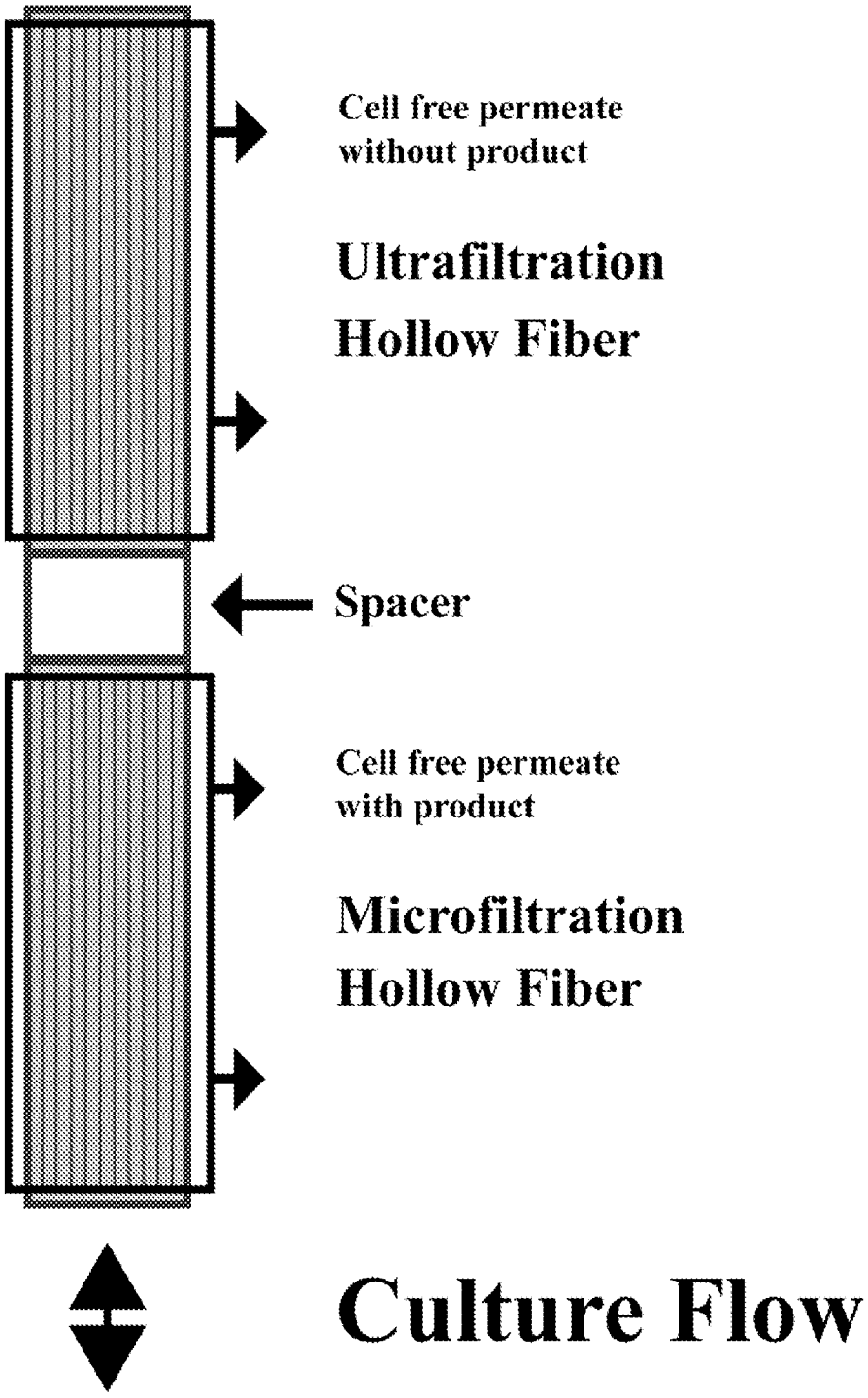

Cell free permeate
without product
Ultrafiltration
Hollow Fiber
Spacer
Cell free permeate
with product
Microfiltration
Hollow Fiber
Culture Flow

Figure 1

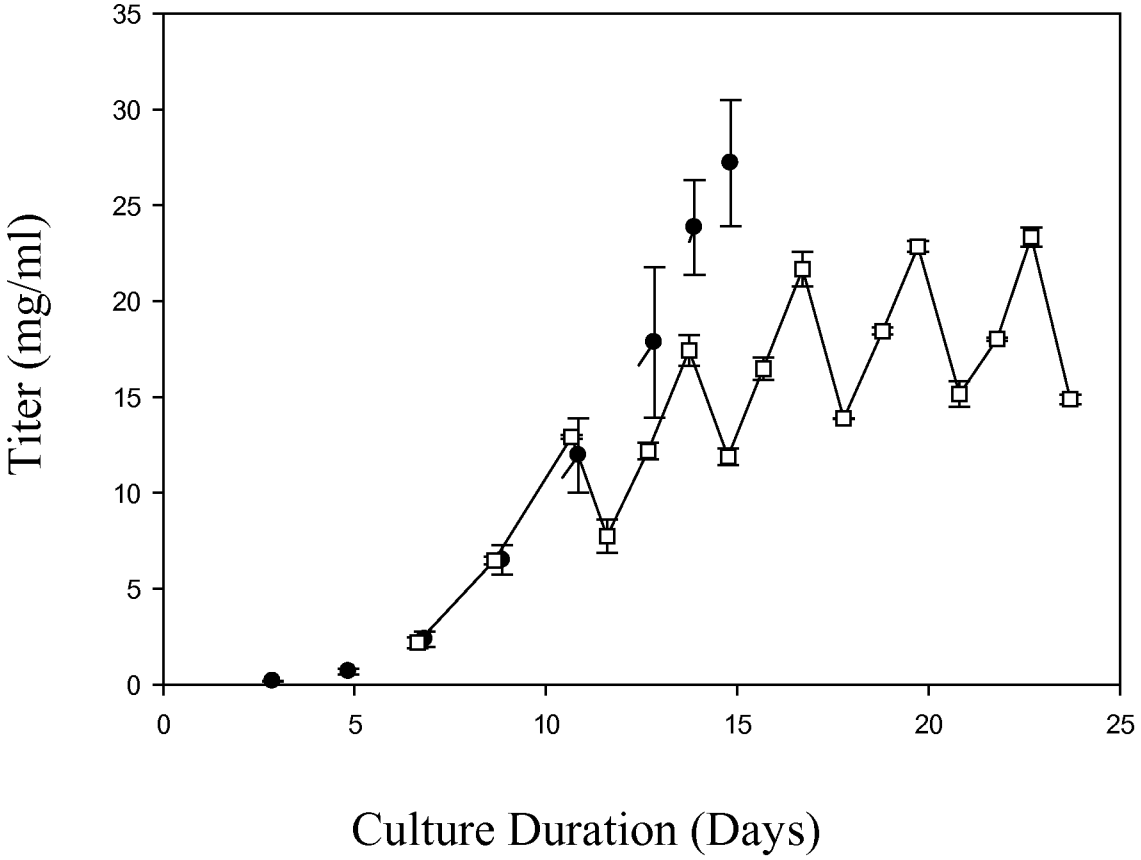

Titer (mg/ml)
35
30
25
20
15
10
5
0
0
5
10
15
20
25
Culture Duration (Days)

Figure 2

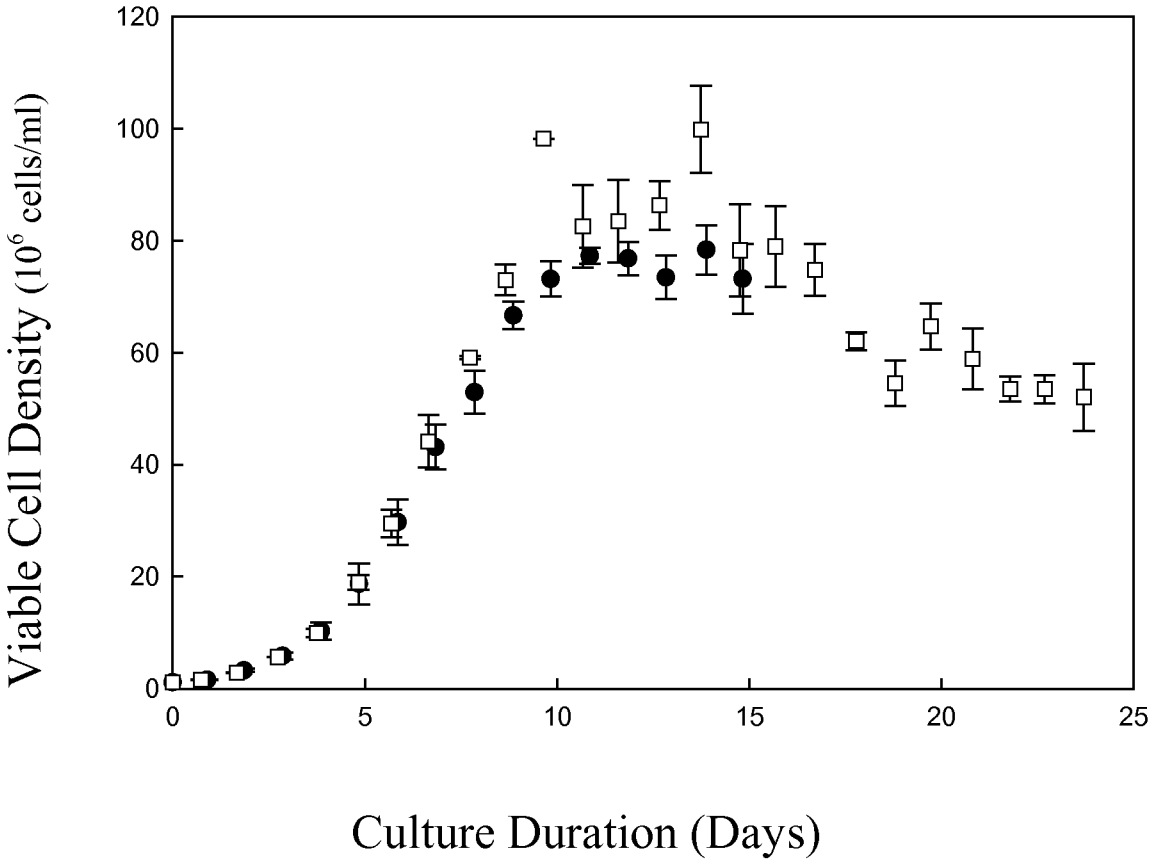

Viable Cell Density (10^6 cells/ml)
120
100
80
60
40
20
0
0
5
10
15
20
25
Culture Duration (Days)

Figure 3

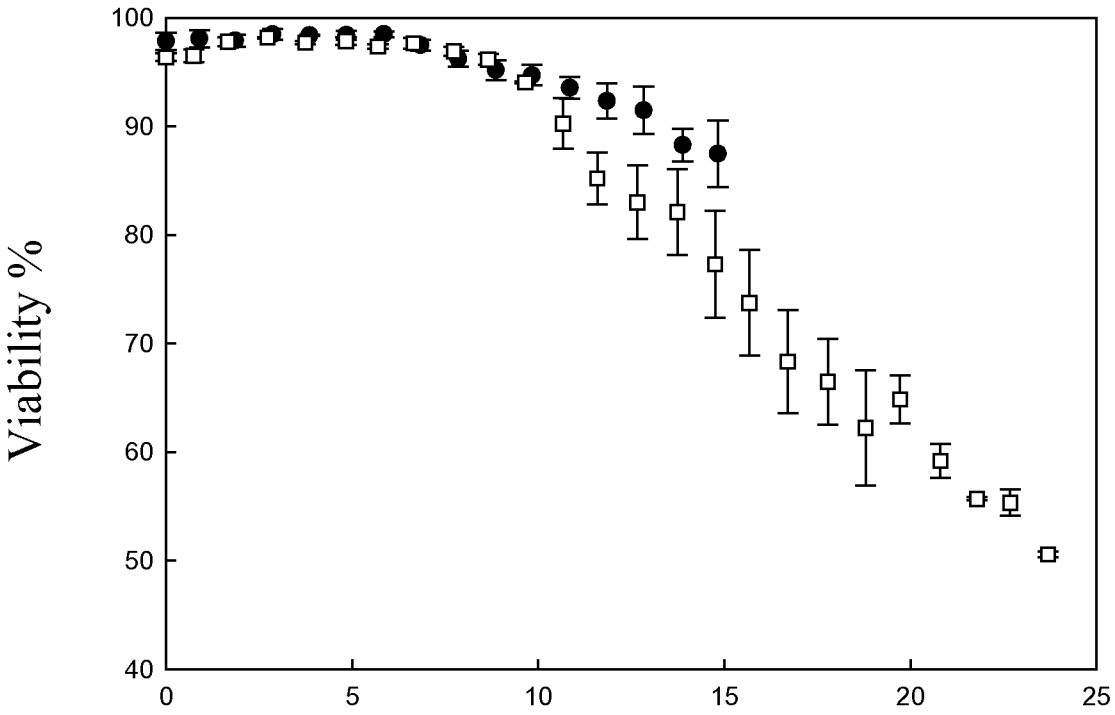

Viability %
100
90
80
70
60
50
40
0
5
10
15
20
25
Culture Duration (Days)

Figure 4

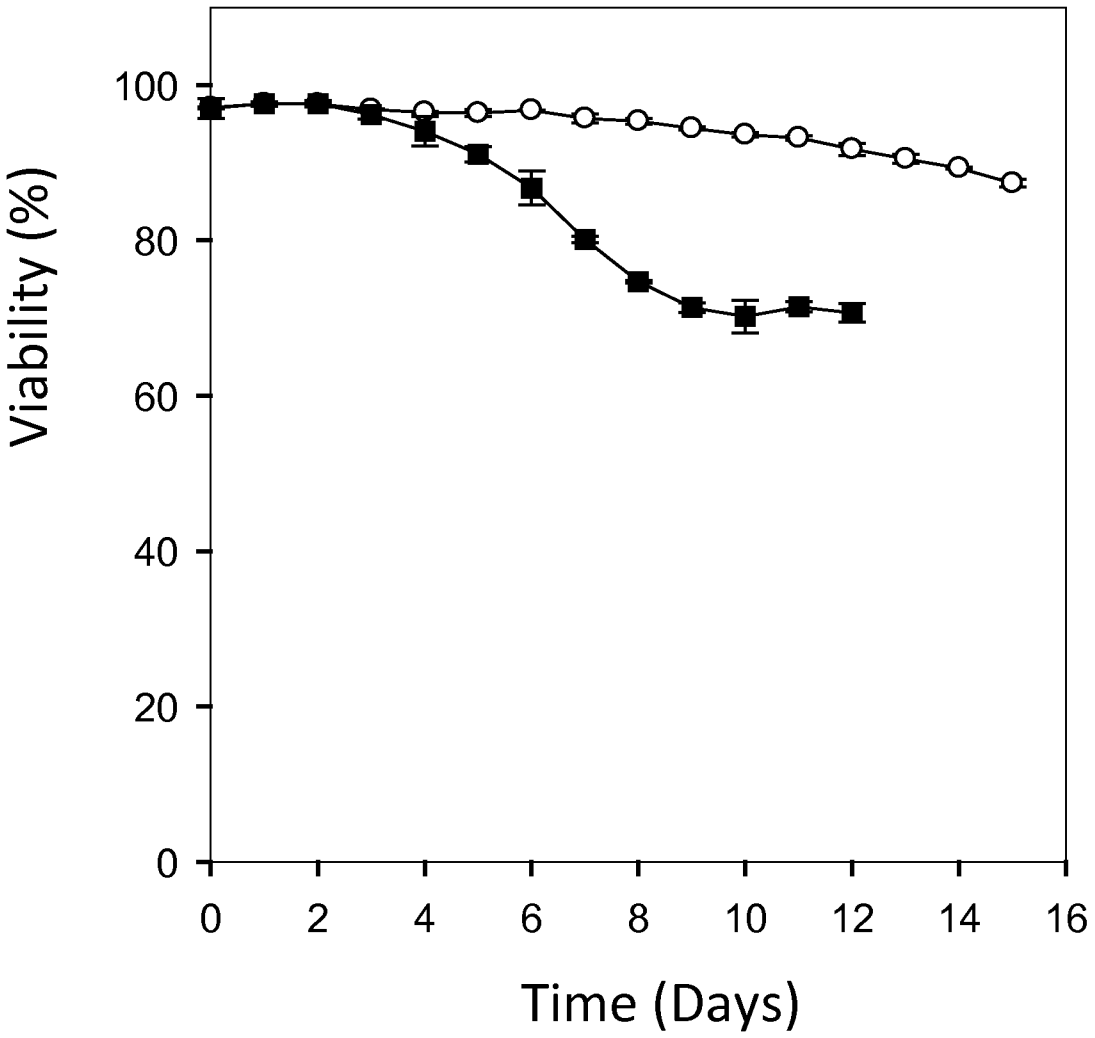
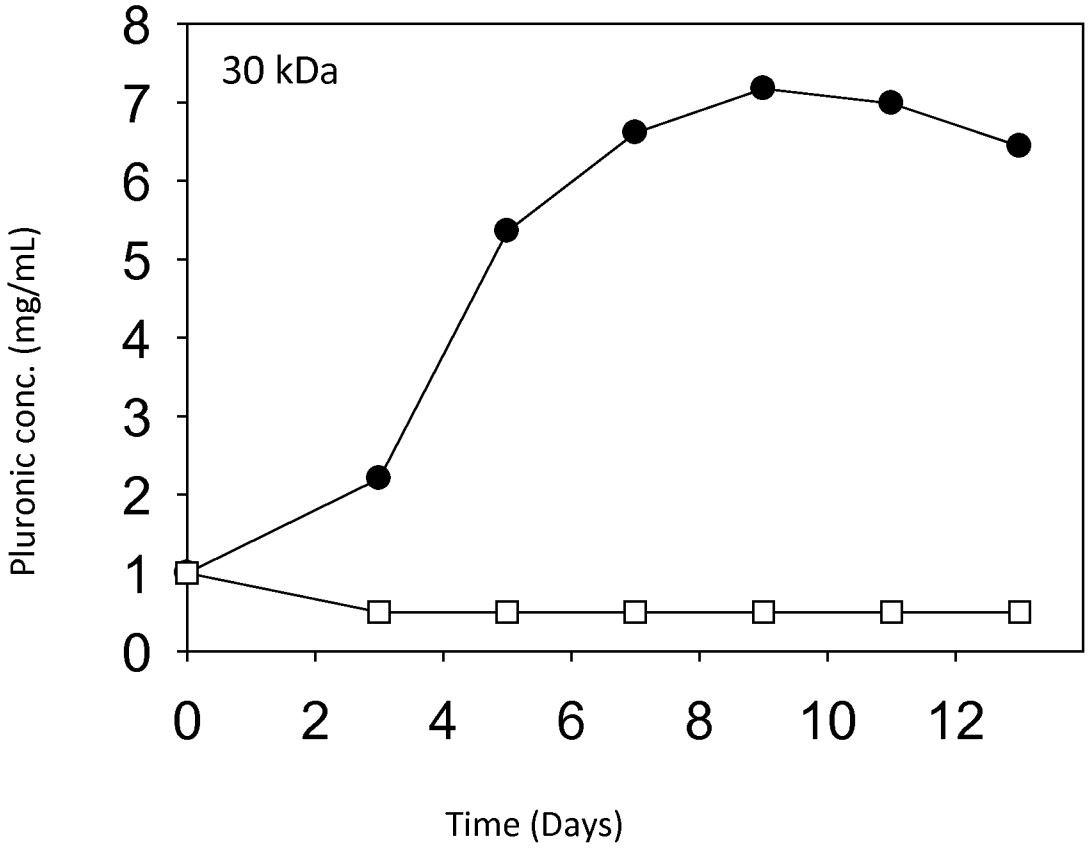


Figure 5


30 kDa
Pluronic conc. (mg/mL)
0
1
2
3
4
5
6
7
8
0
2
4
6
8
10
12
Time (Days)

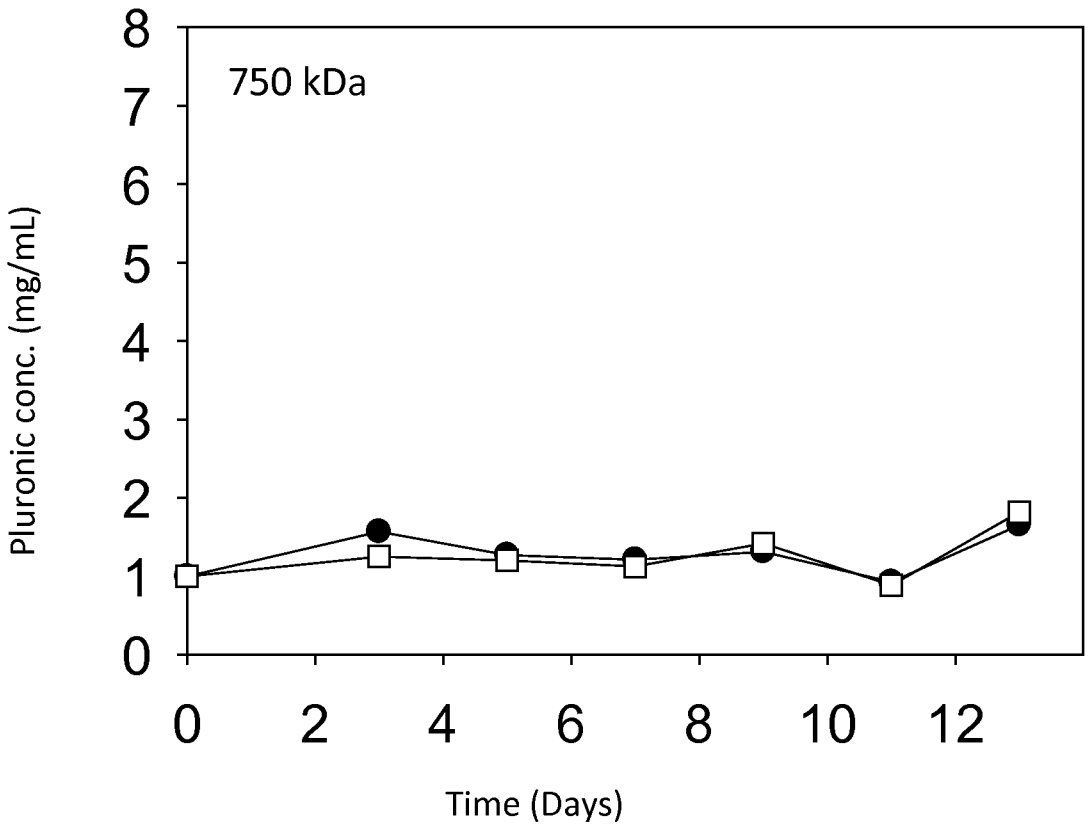
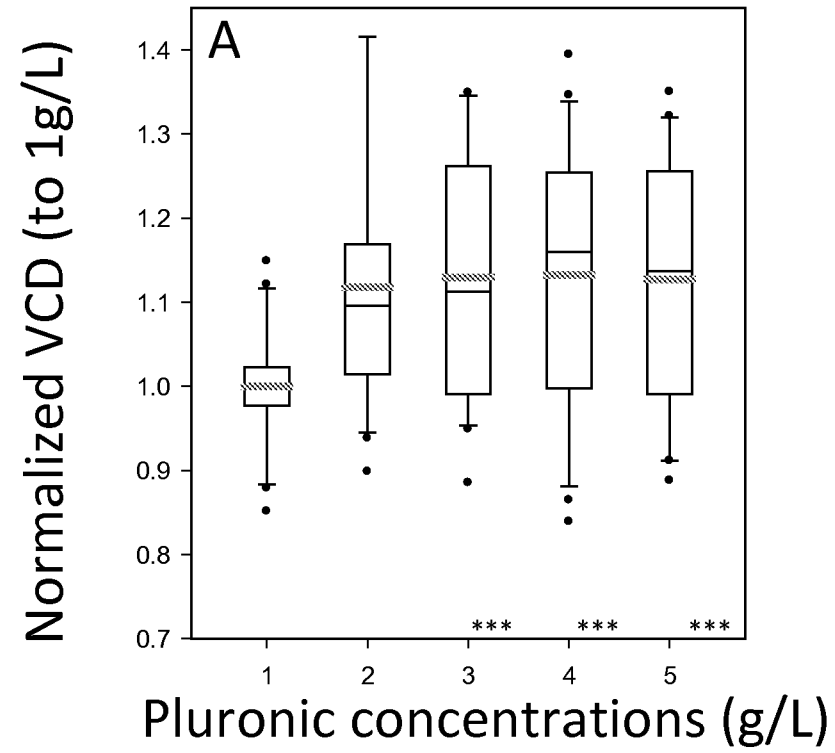

750 kDa
Pluronic conc. (mg/mL)
0
1
2
3
4
5
6
7
8
0
2
4
6
8
10
12
Time (Days)

Figure 6


A
Normalized VCD (to 1g/L)
1.4
1.3
1.2
1.1
1.0
0.9
0.8
0.7
***
***
***
1
2
3
4
5
Pluronic concentrations (g/L)

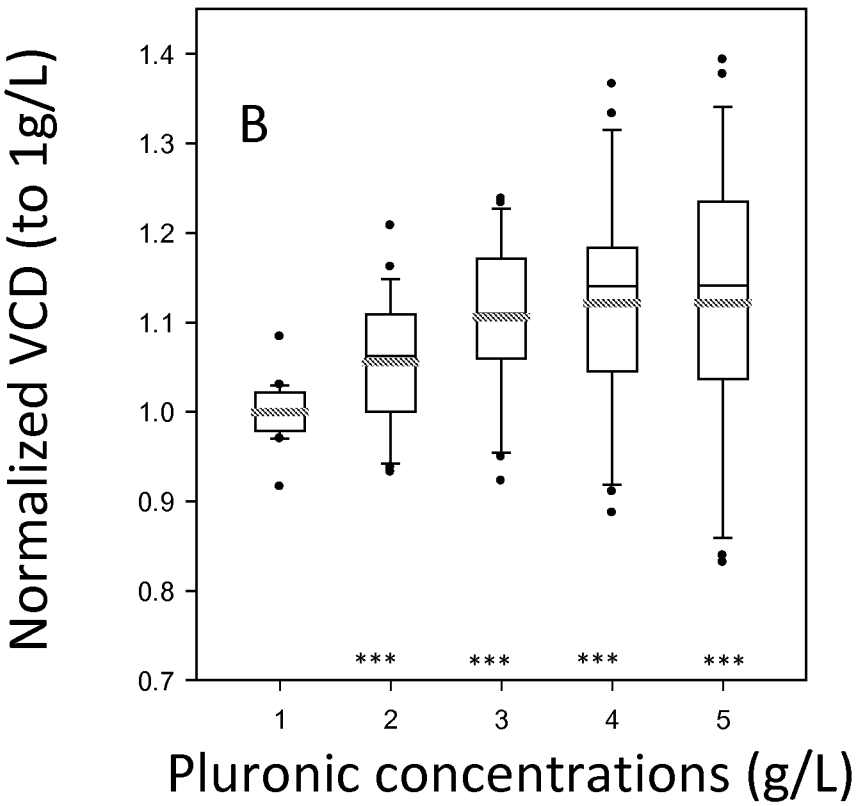
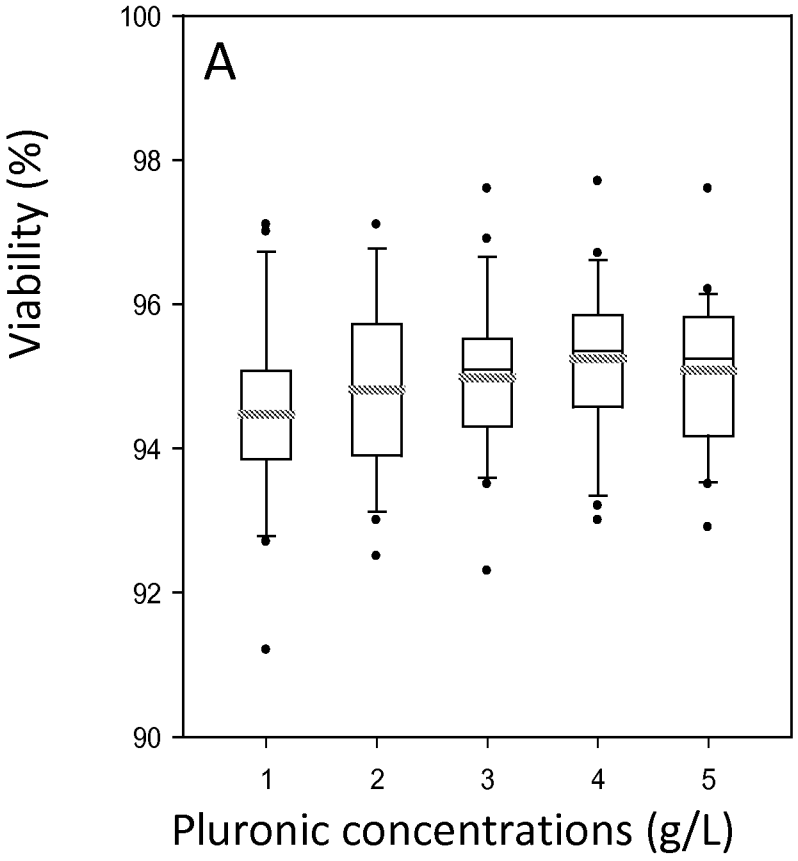

B
Normalized VCD (to 1g/L)
1.4
1.3
1.2
1.1
1.0
0.9
0.8
0.7
***
***
***
***
1
2
3
4
5
Pluronic concentrations (g/L)

Figure 7A


A
Viability (%)
100
98
96
94
92
90
1
2
3
4
5
Pluronic concentrations (g/L)

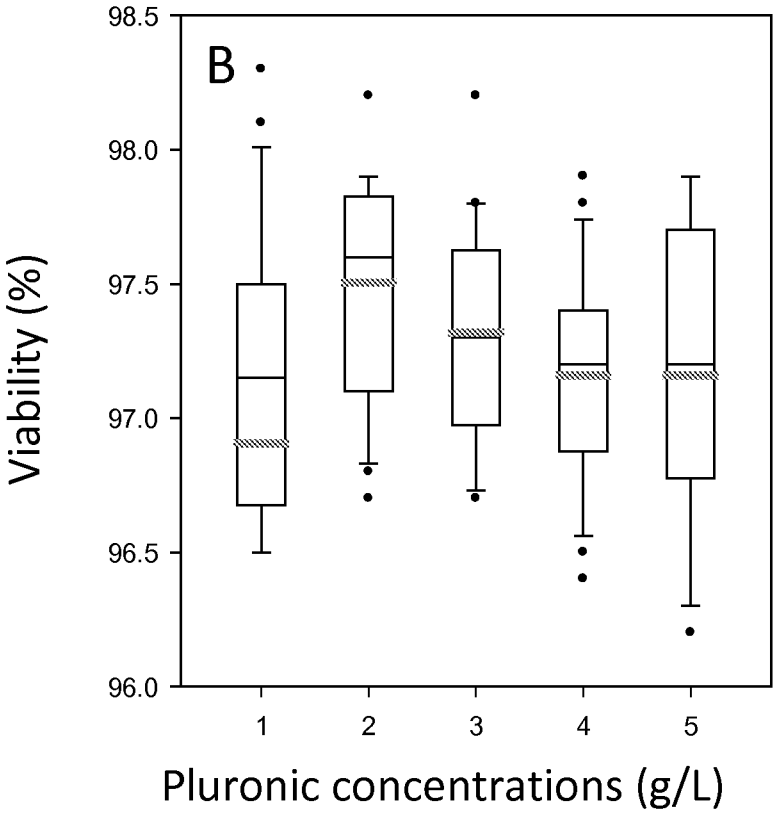
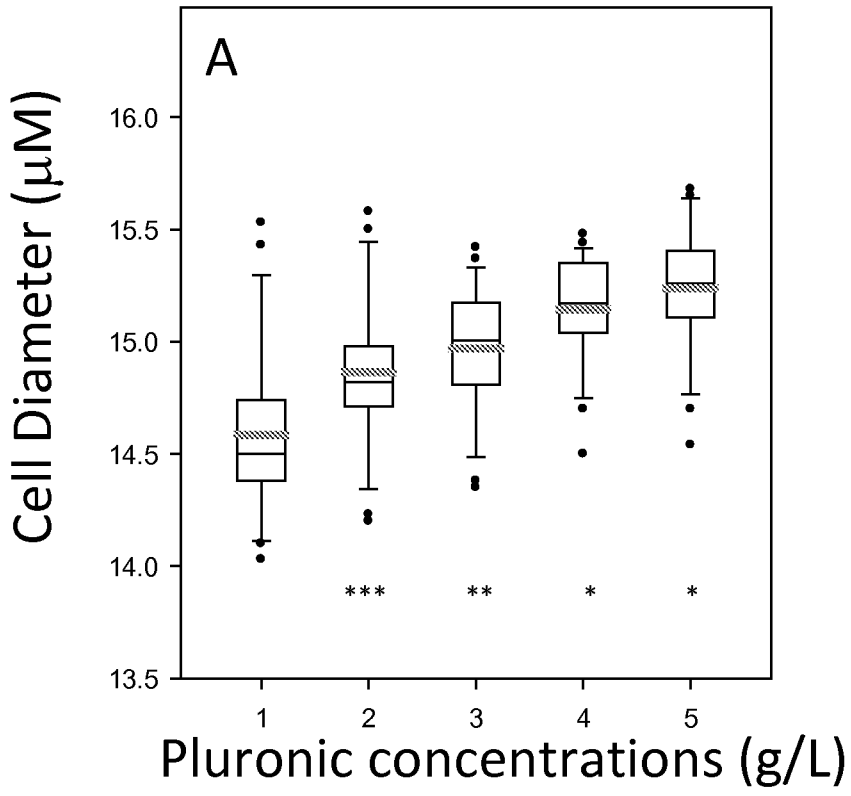

B
Viability (%)
98.5
98.0
97.5
97.0
96.5
96.0
1
2
3
4
5
Pluronic concentrations (g/L)

Figure 7B


A
Cell Diameter (µM)
16.0
15.5
15.0
14.5
14.0
13.5
***
**
*
*
1
2
3
4
5
Pluronic concentrations (g/L)

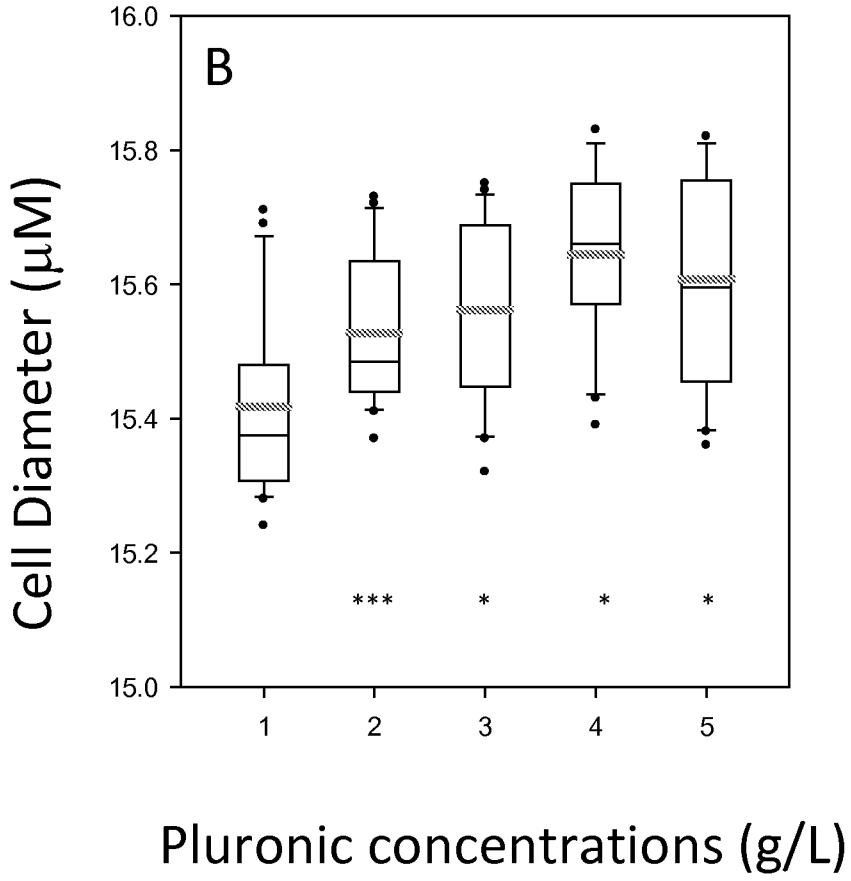

B
Cell Diameter (µM)
16.0
15.8
15.6
15.4
15.2
15.0
***
*
*
*
1
2
3
4
5
Pluronic concentrations (g/L)

Figure 7C

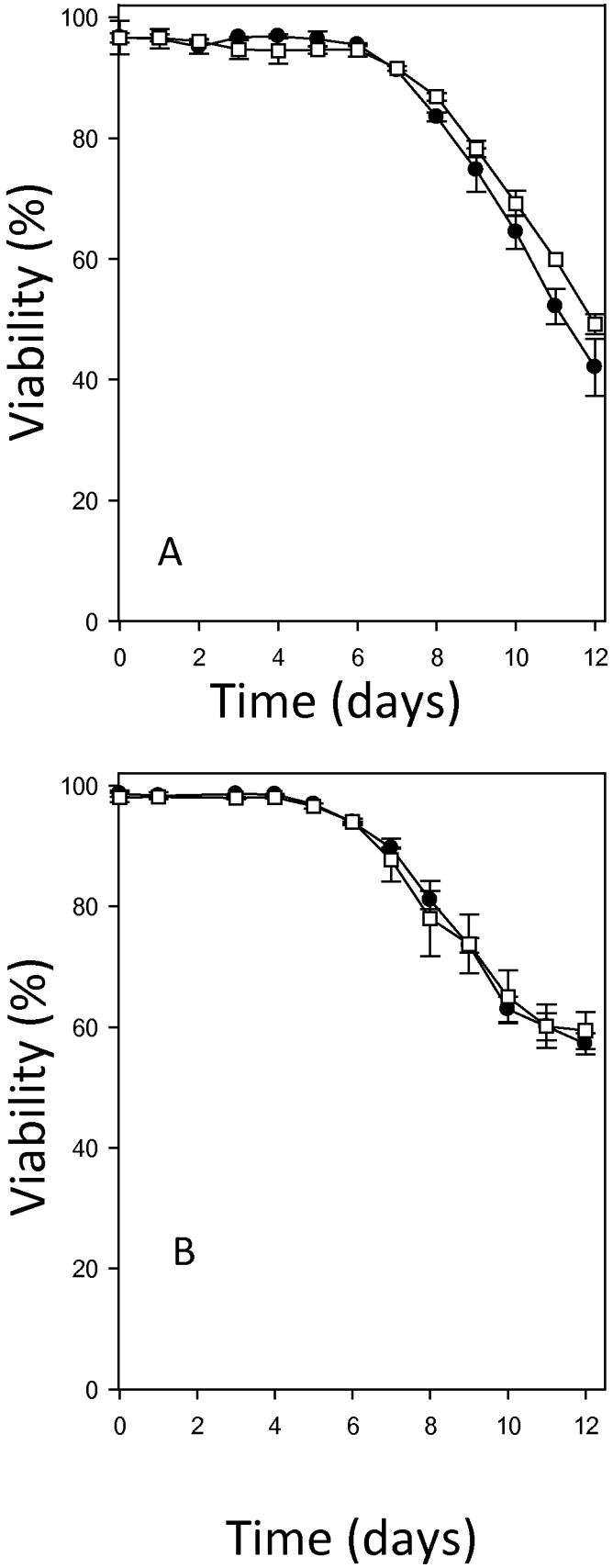

100
80
60
40
20
0
Viability (%)
A
0
2
4
6
8
10
12
Time (days)
100
80
60
40
20
0
Viability (%)
B
0
2
4
6
8
10
12
Time (days)

Figure 8

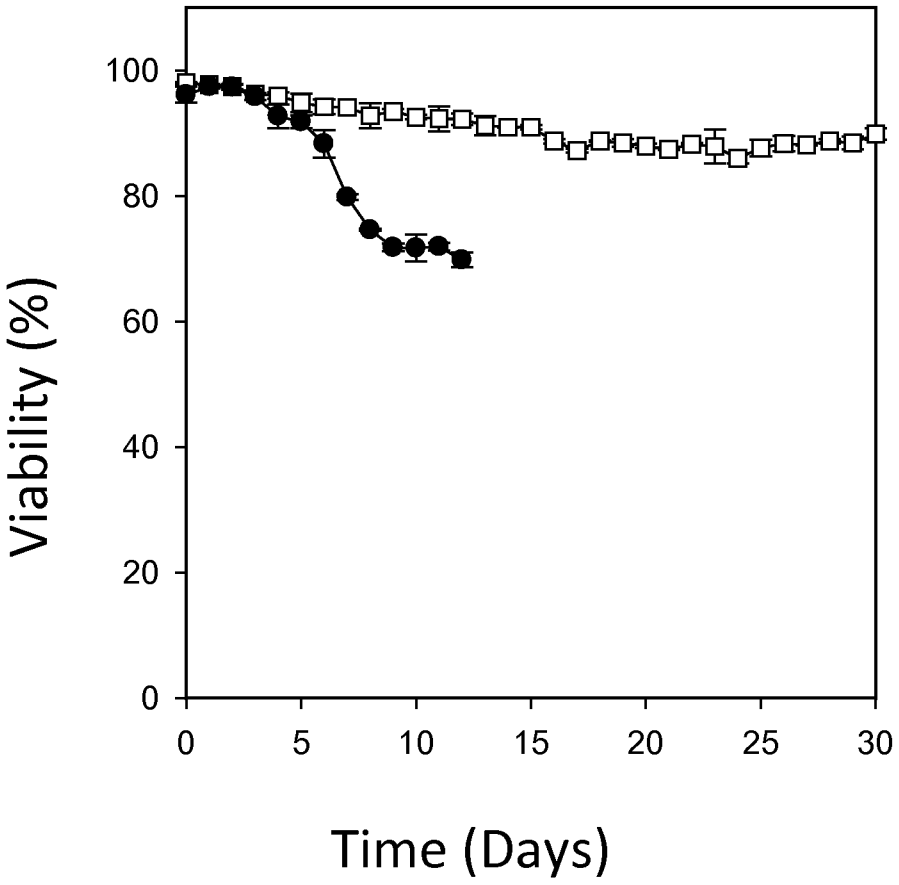


Figure 9

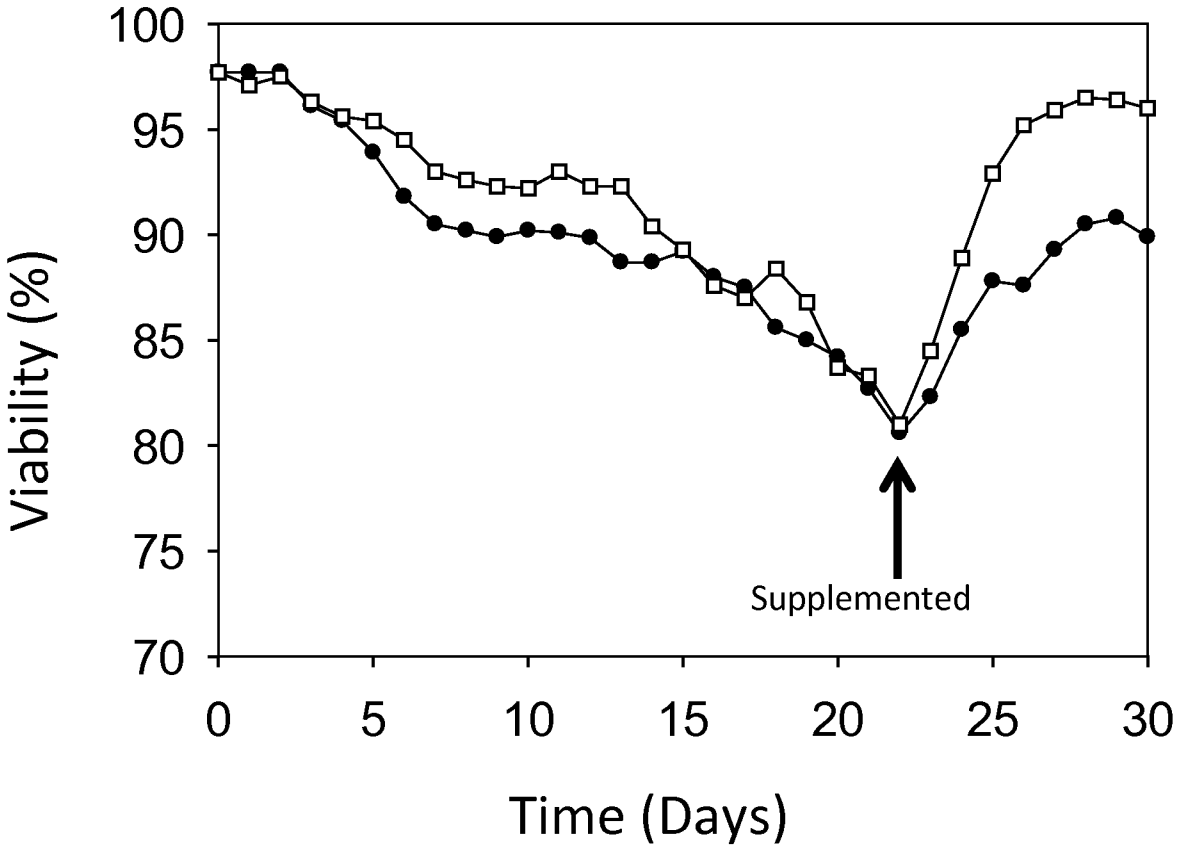

100
95
90
85
80
75
70
Viability (%)
0
5
10
15
20
25
30
Time (Days)
Supplemented

Figure 10

Figure 11

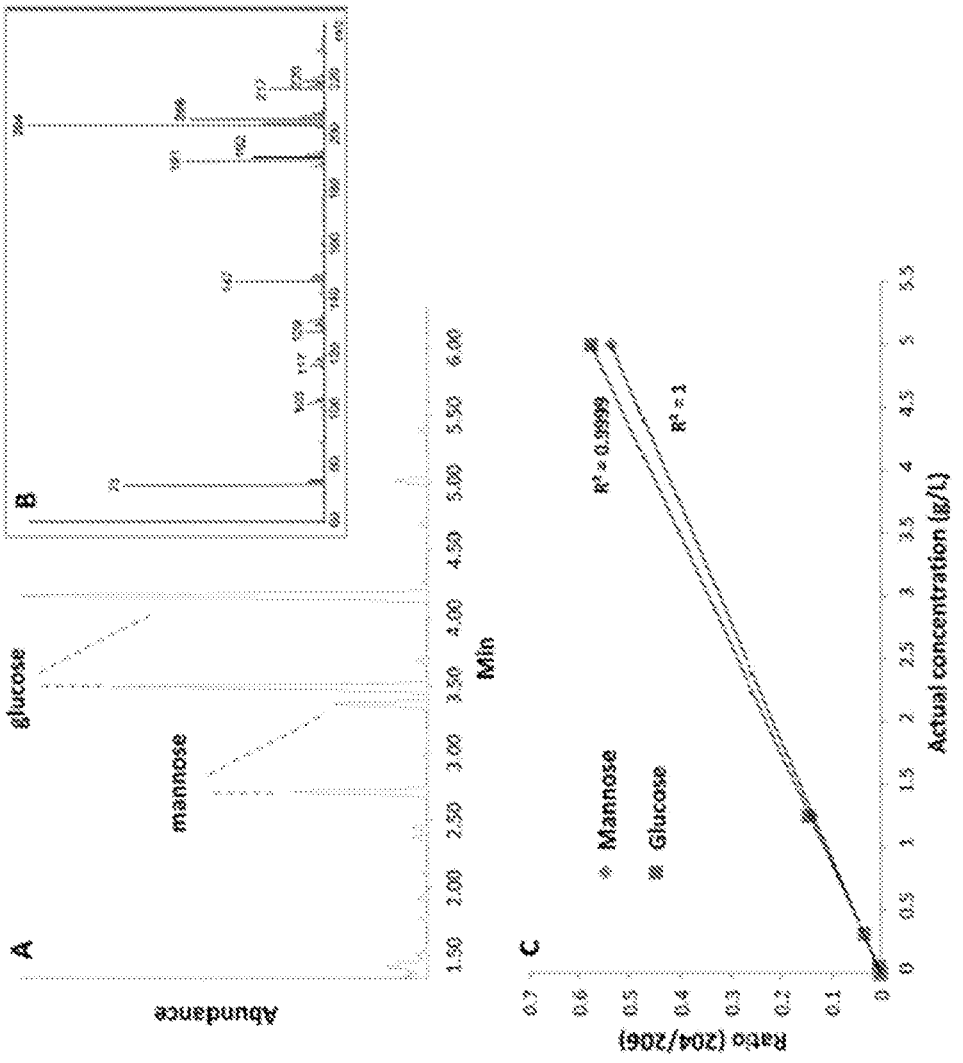

A
glucose
mannose
Abundance
Min
B
C
Mannose
Glucose
Ratio (204/206)
Actual concentration (g/L)

Figure 12

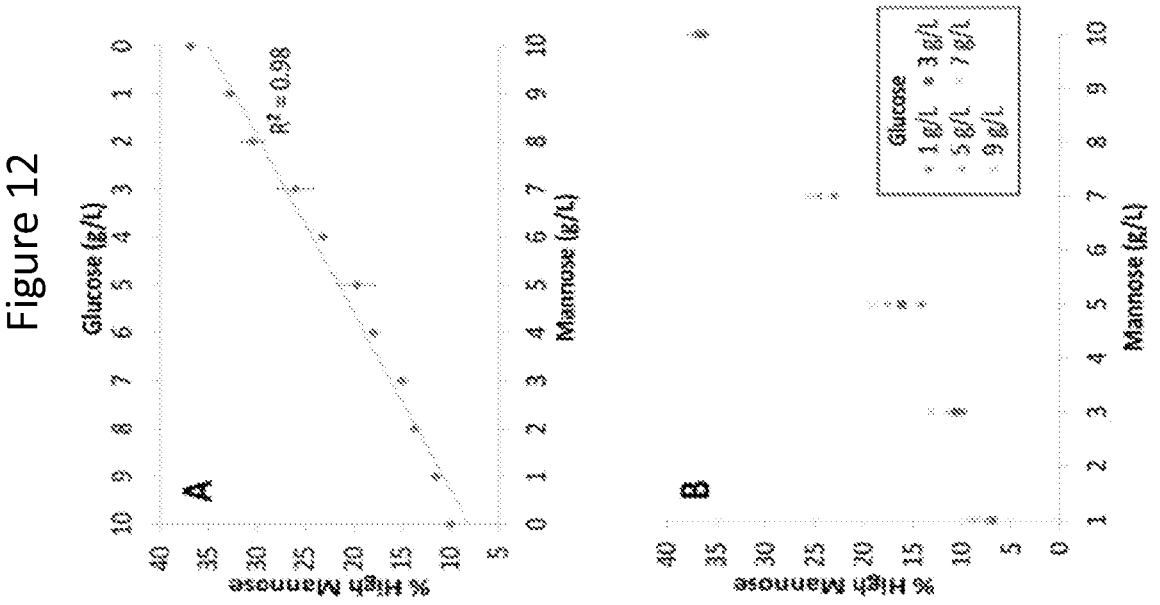

A
Glucose (g/L)
% High Mannose
Mannose (g/L)
$R^2 = 0.98$
B
% High Mannose
Mannose (g/L)
Glucose
1 g/L
3 g/L
5 g/L
7 g/L
9 g/L

Figure 13

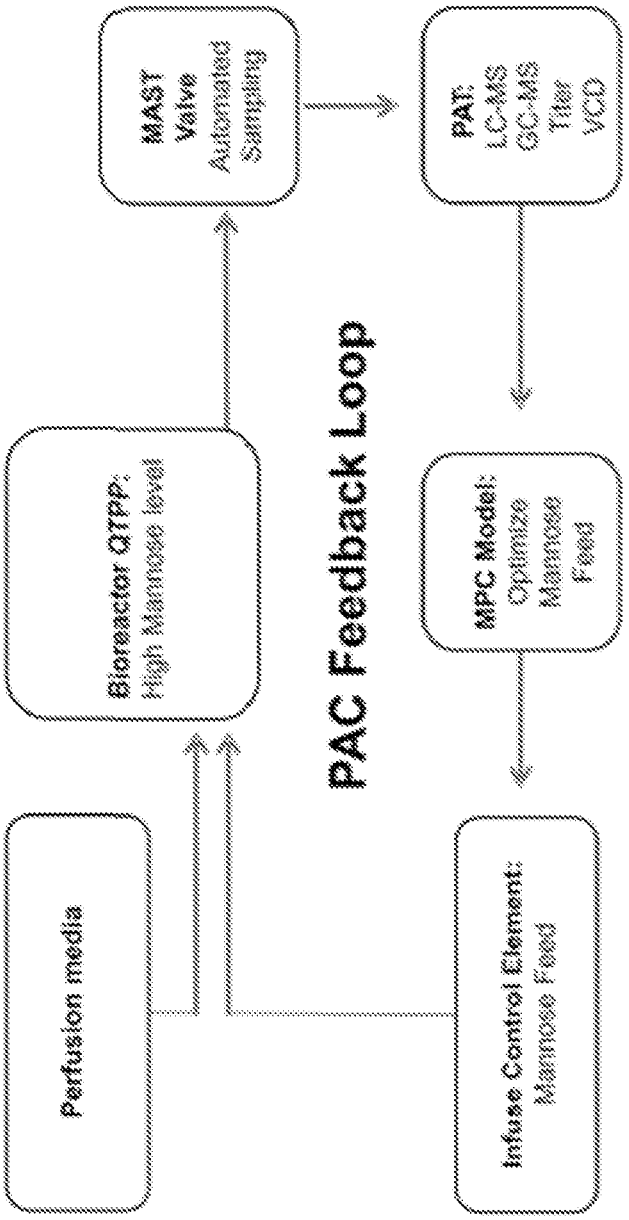

PAC Feedback Loop
MAST
Valve
Automated
Sampling
PAT:
LC-MS
GC-MS
Titer
VCD
Bioreactor QTPP:
High Mannose level
MPC Model:
Optimize
Mannose
Feed
Perfusion media
Infuse Control Element:
Mannose Feed

Figure 14

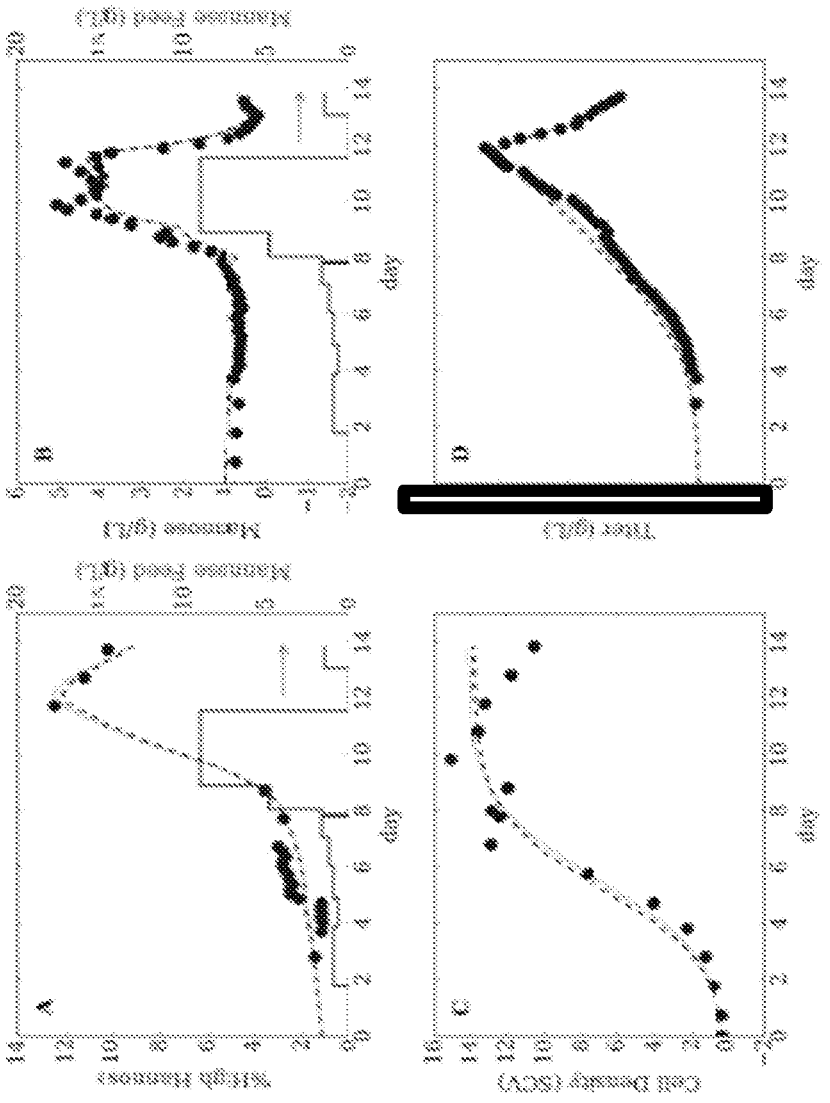

A
B
C
D
day
Mannose (g/L)
Mannose Feed (g/L)
Titer (g/L)
Cell Density (SCV)

Figure 15

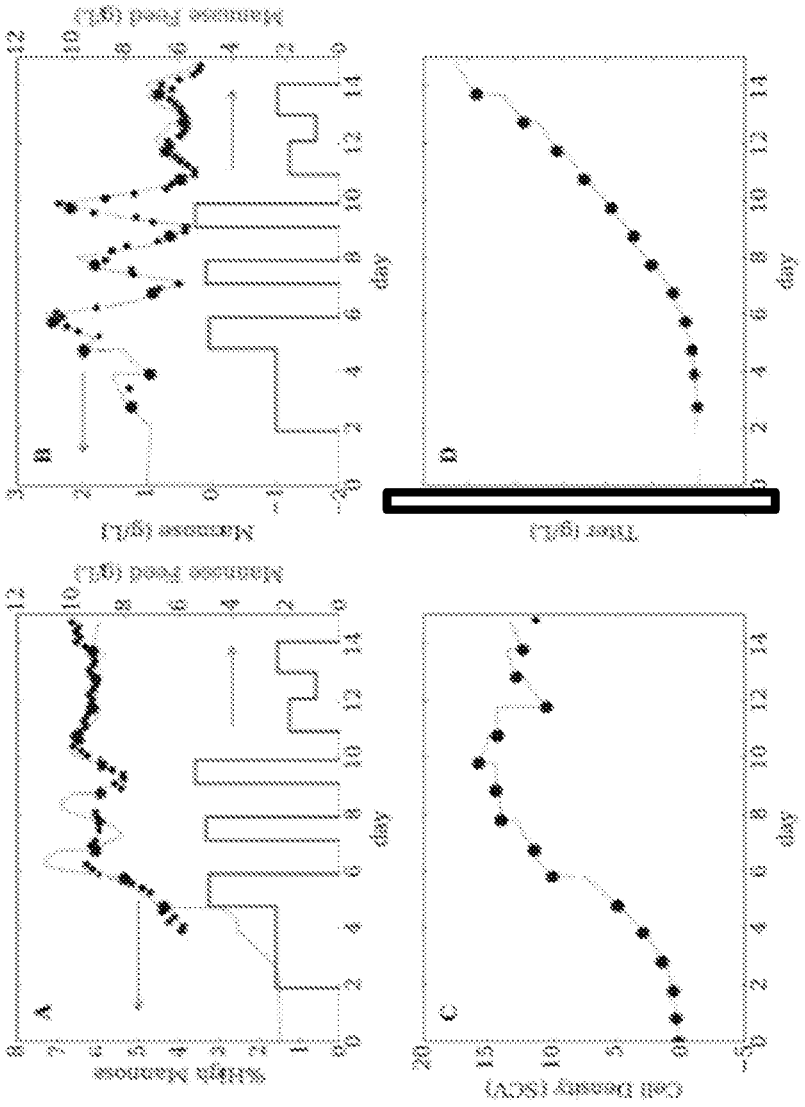
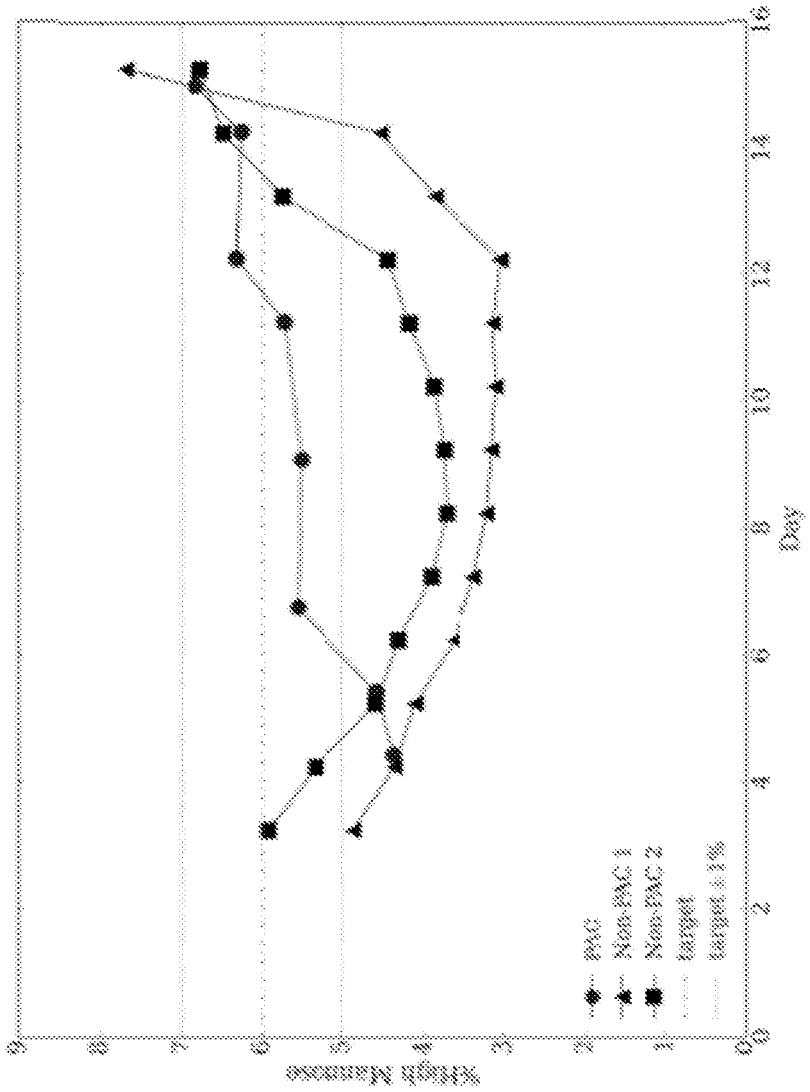

A
Viable Cell Density (MVCD)
Mannose Feed (g/L)
day
B
Titer (g/L)
day
C
Cell Density (MVCD)
day

Figure 16


%High Mannose
Day
0
2
4
6
8
10
12
14
16
18
20
22
24
26
28
30
0
7
14
21
28
ENG
Non-ENG 1
Non-ENG 2
target
target ± 5%