Fig. 1

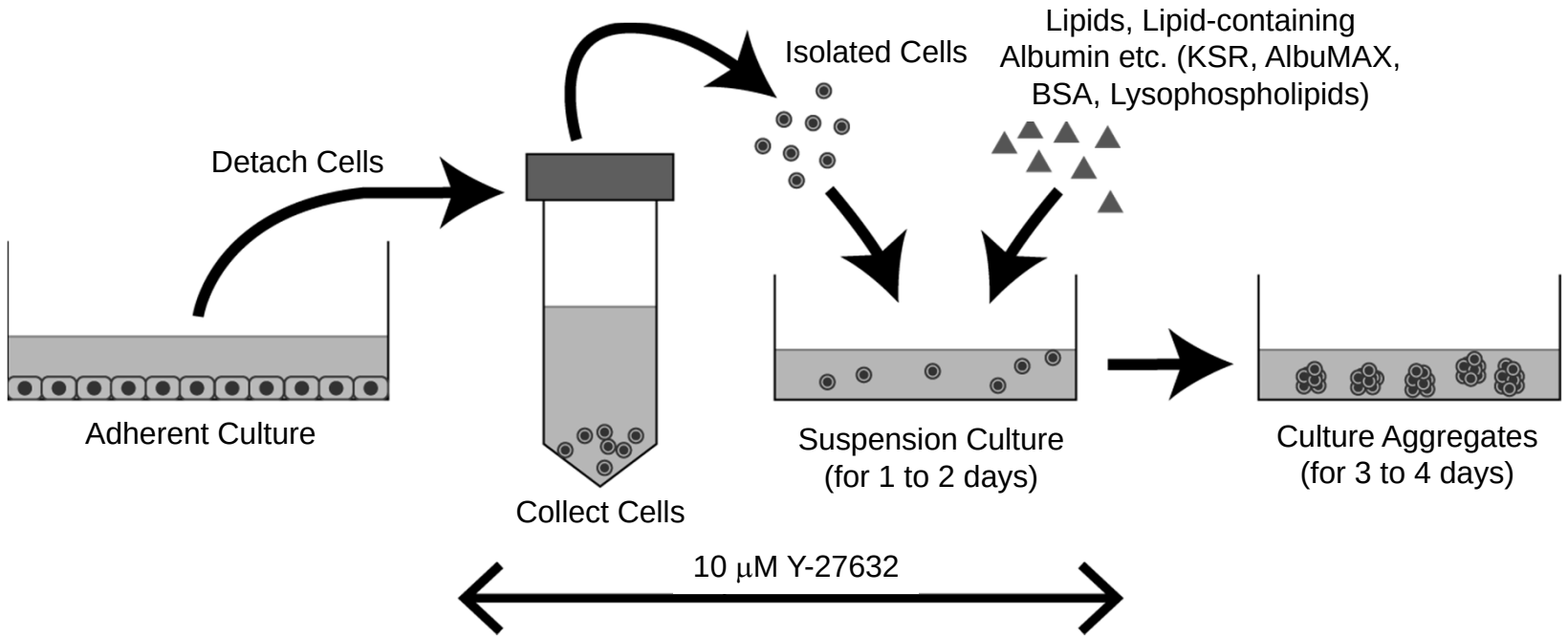

Isolated Cells
Lipids, Lipid-containing
Albumin etc. (KSR, AlbuMAX,
BSA, Lysophospholipids)
Detach Cells
Adherent Culture
Collect Cells
Suspension Culture
(for 1 to 2 days)
Culture Aggregates
(for 3 to 4 days)
10 μM Y-27632

Fig. 2

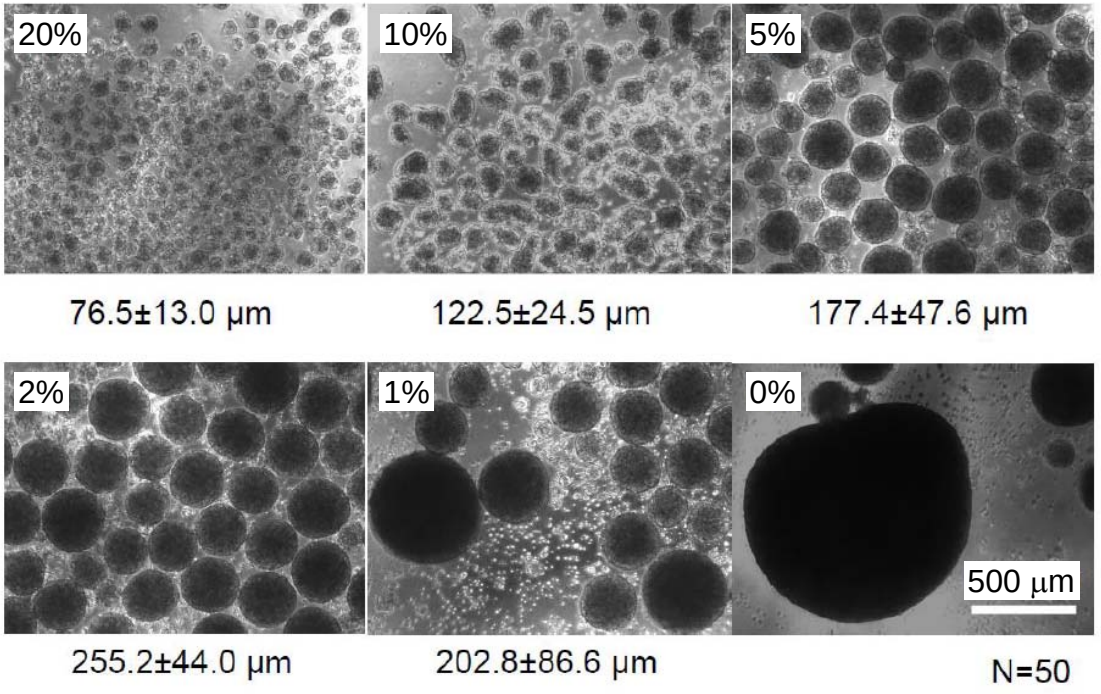

20%
10%
5%
76.5±13.0 µm
122.5±24.5 µm
177.4±47.6 µm
2%
1%
0%
500 µm
255.2±44.0 µm
202.8±86.6 µm
N=50

Fig. 3

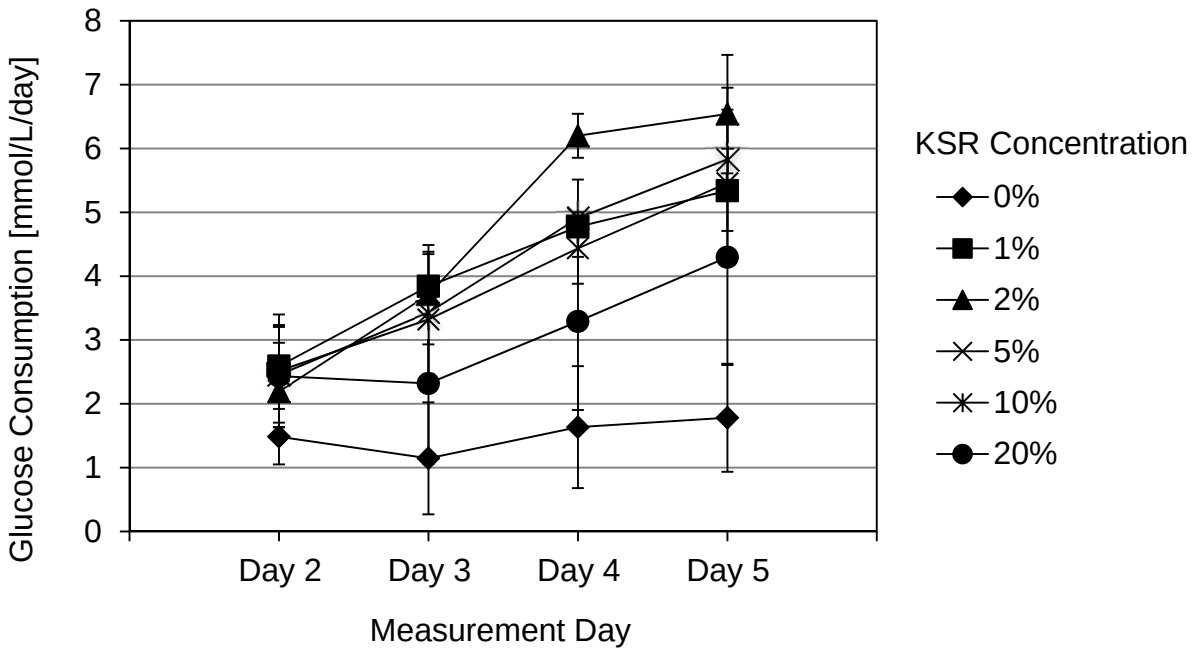

Glucose Consumption [mmol/L/day]
0
1
2
3
4
5
6
7
8
Day 2
Day 3
Day 4
Day 5
Measurement Day
KSR Concentration
0%
1%
2%
5%
10%
20%

Fig. 4

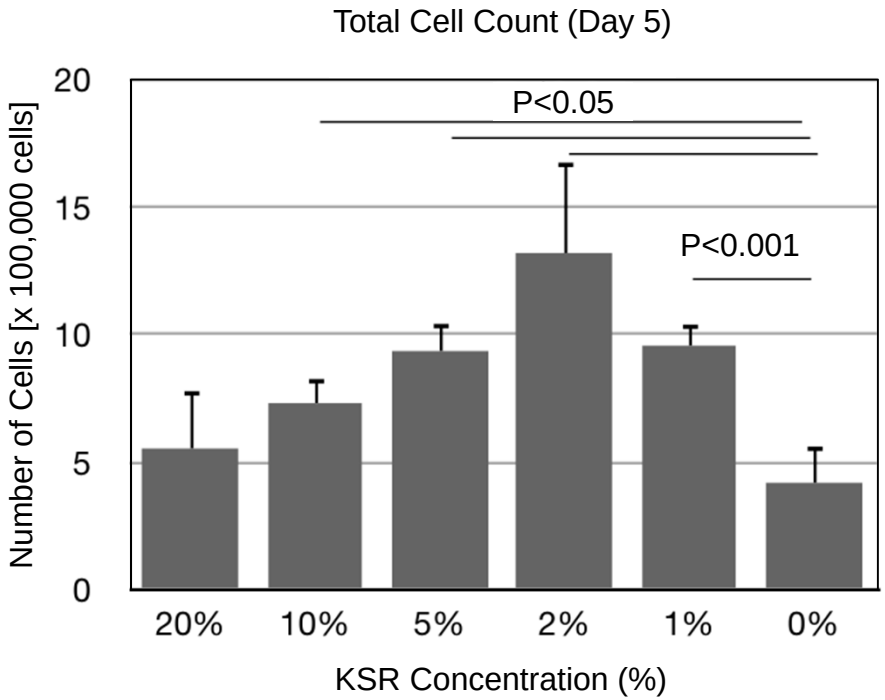

Total Cell Count (Day 5)
Number of Cells [x 100,000 cells]
20
15
10
5
0
P<0.05
P<0.001
20%
10%
5%
2%
1%
0%
KSR Concentration (%)

Fig. 5

Amino Acids	Glycine, L-histidine, L-isoleucine, L-methionine, L-phenylalanine, L-proline, L-hydroxyproline, L-serine, L-threonine, L-tryptophan, L-tyrosine, L-valine
Vitamins/Antioxidants	Thiamine, reduced glutathione, ascorbic acid 2-PO_4
Trace Elements	Ag^{+}, $Al3^{+}$, Ba^{2+}, Cd^{2+}, Co^{2+}, Cr^{3+}, Ge^{4+}, Se^{4+}, Br^{-}, I^{-}, F^{-}, Mn^{2+}, Si^{4+}, V^{5+}, Mo^{6+}, Ni^{2+}, Rb^{+}, Sn^{2+}, Zr^{4+}
Proteins	Transferrin (iron-saturated), insulin, lipid-rich albumin (AlbuMAX)

Fig. 6

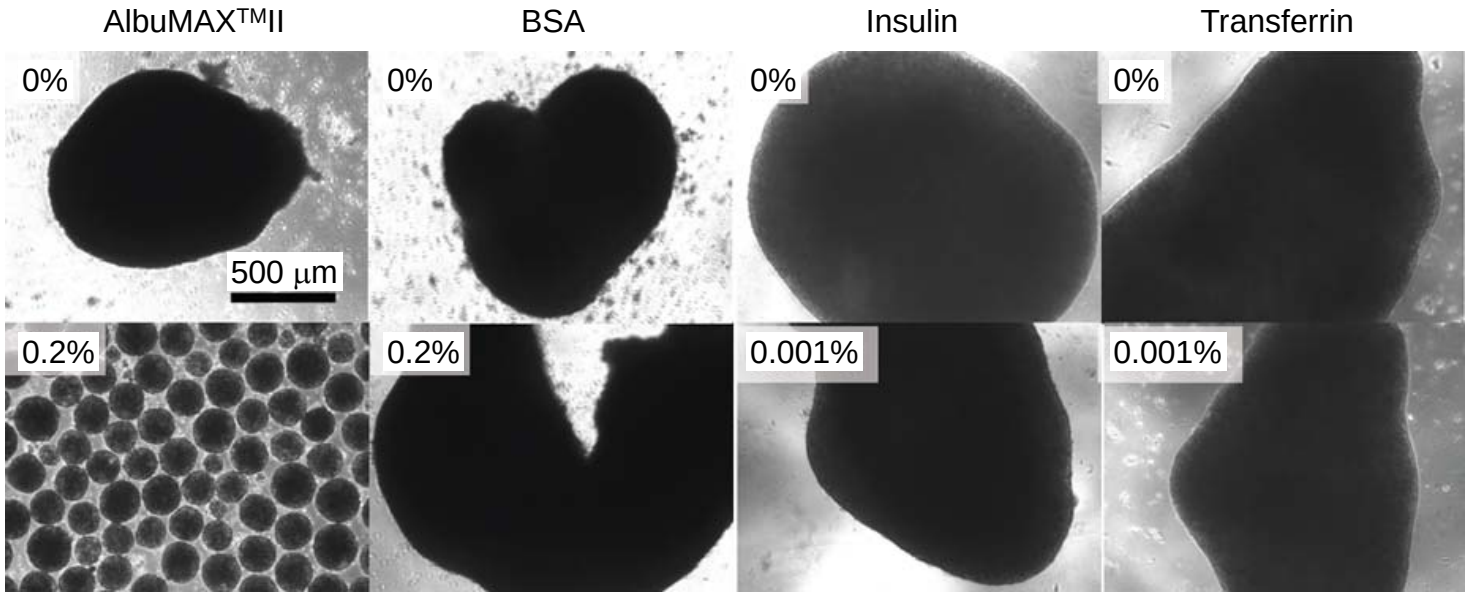
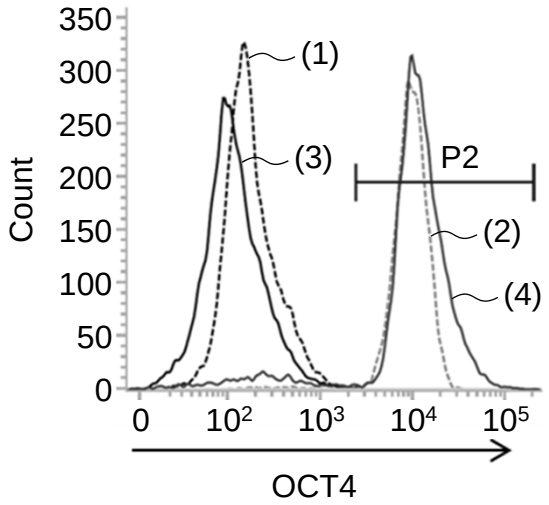
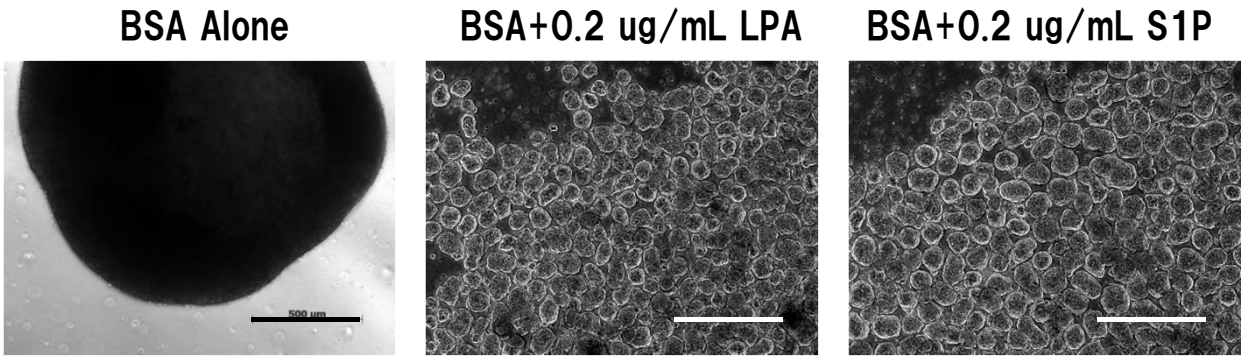

AlbuMAX™II
BSA
Insulin
Transferrin
0%
0%
0%
0%
500 μm
0.2%
0.2%
0.001%
0.001%

Fig. 7



		Percentage of Cells Positive for OCT4
(1)	Adherent (Isotype cont.)	0%
(2)	Adherent (Anti-OCT4)	98%
(3)	Suspension (Isotype cont.)	0%
(4)	Suspension (Anti-OCT4)	96%

Fig. 8



Scale bar: 500 um

Fig. 9

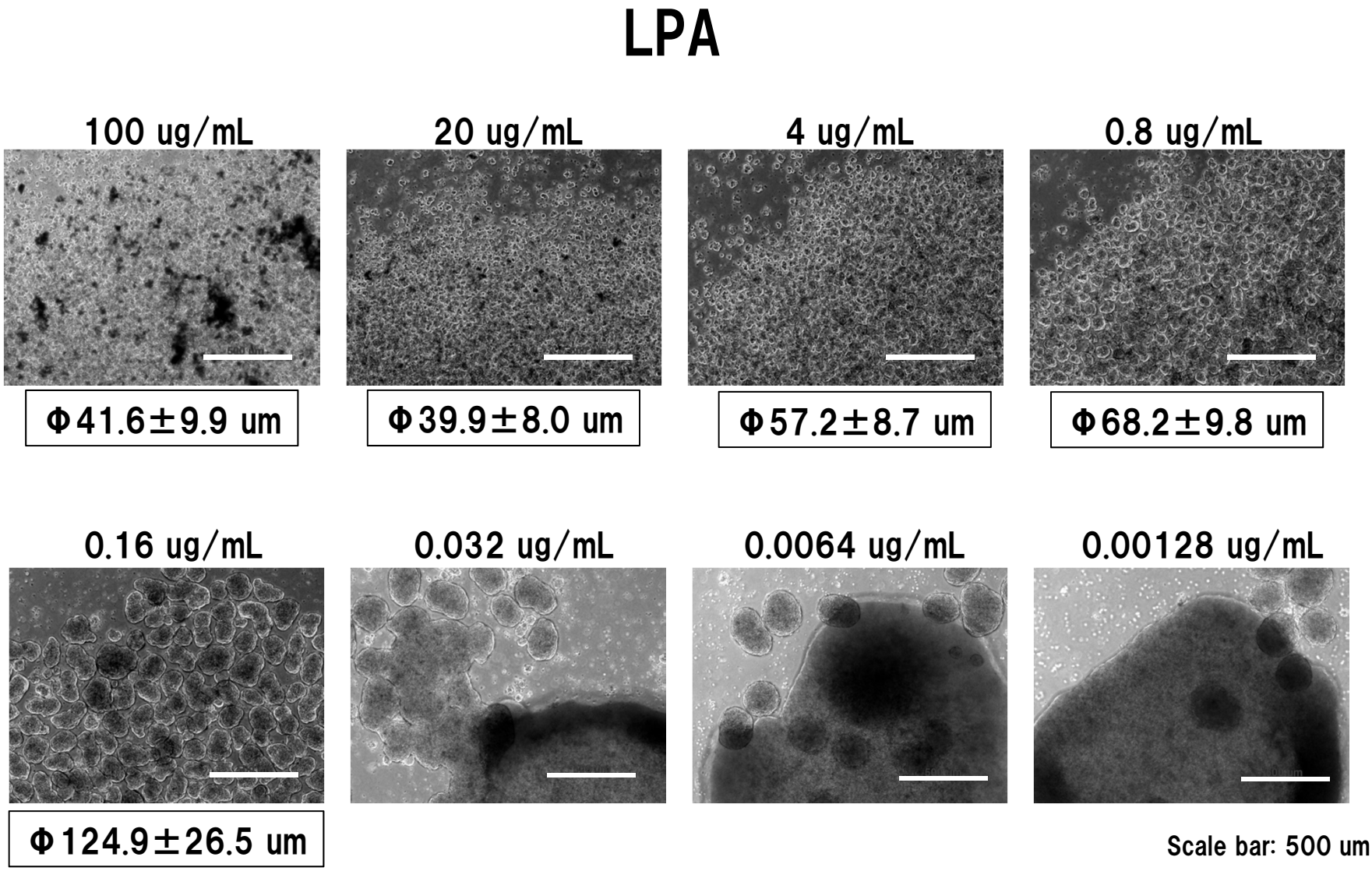

LPA
100 ug/mL
20 ug/mL
4 ug/mL
0.8 ug/mL
Φ41.6±9.9 um
Φ39.9±8.0 um
Φ57.2±8.7 um
Φ68.2±9.8 um
0.16 ug/mL
0.032 ug/mL
0.0064 ug/mL
0.00128 ug/mL
Φ124.9±26.5 um
Scale bar: 500 um

Fig. 10

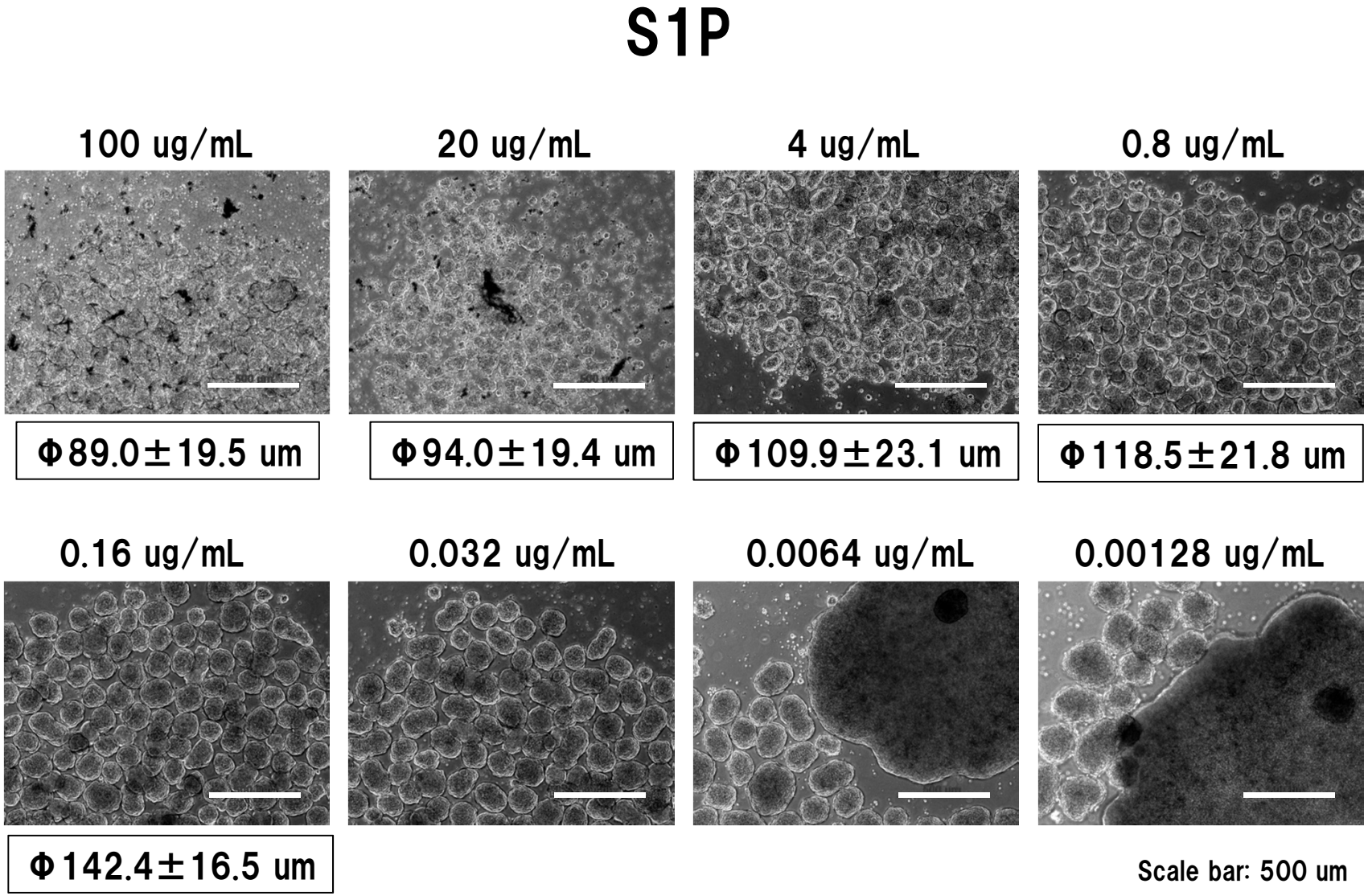

S1P
100 ug/mL
20 ug/mL
4 ug/mL
0.8 ug/mL
Φ89.0±19.5 um
Φ94.0±19.4 um
Φ109.9±23.1 um
Φ118.5±21.8 um
0.16 ug/mL
0.032 ug/mL
0.0064 ug/mL
0.00128 ug/mL
Φ142.4±16.5 um
Scale bar: 500 um

Fig. 11

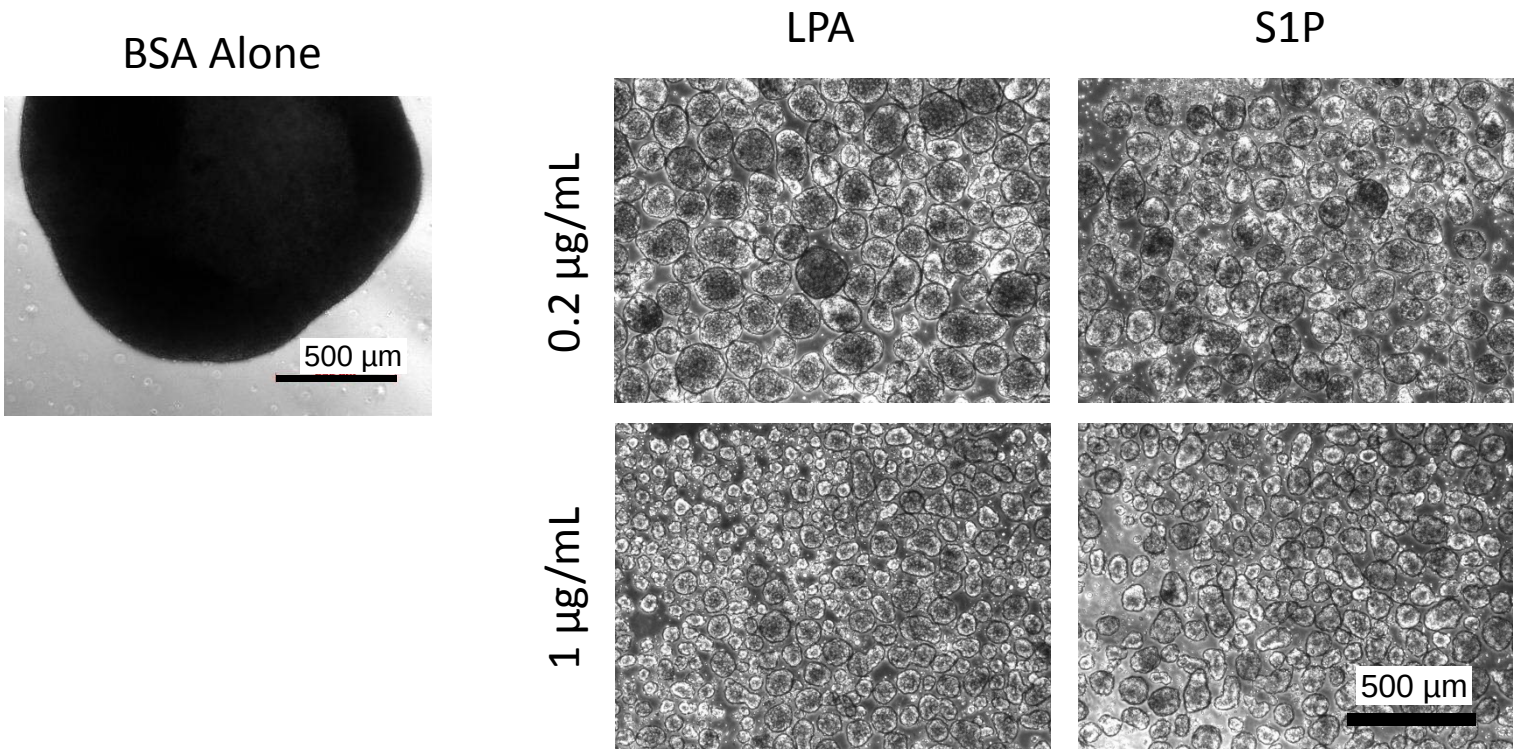
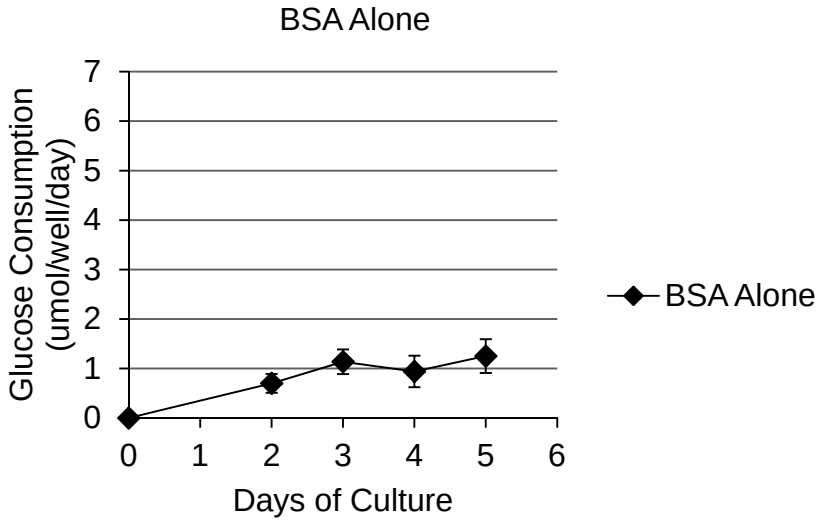
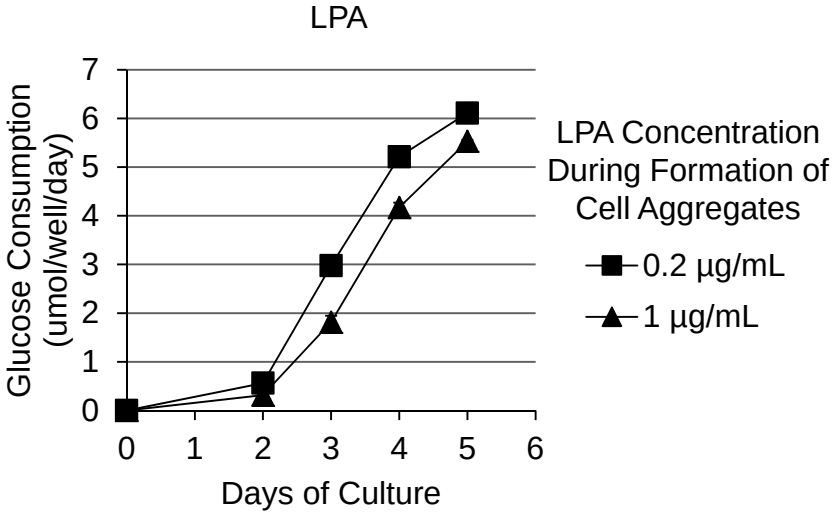


Fig. 12


BSA Alone
Glucose Consumption (umol/well/day)
0
1
2
3
4
5
6
7
Days of Culture
BSA Alone


LPA
Glucose Consumption (umol/well/day)
Days of Culture
LPA Concentration During Formation of Cell Aggregates
0.2 µg/mL
1 µg/mL

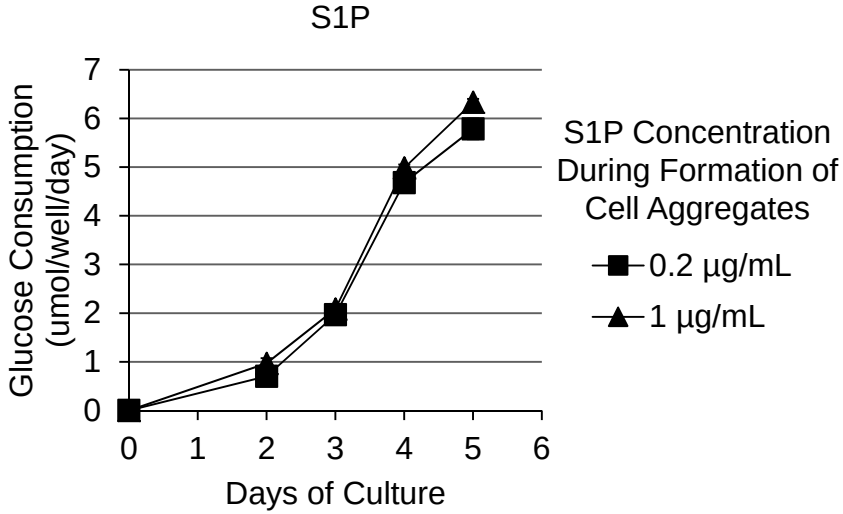

S1P
Glucose Consumption (umol/well/day)
Days of Culture
S1P Concentration During Formation of Cell Aggregates
0.2 µg/mL
1 µg/mL

Fig. 13

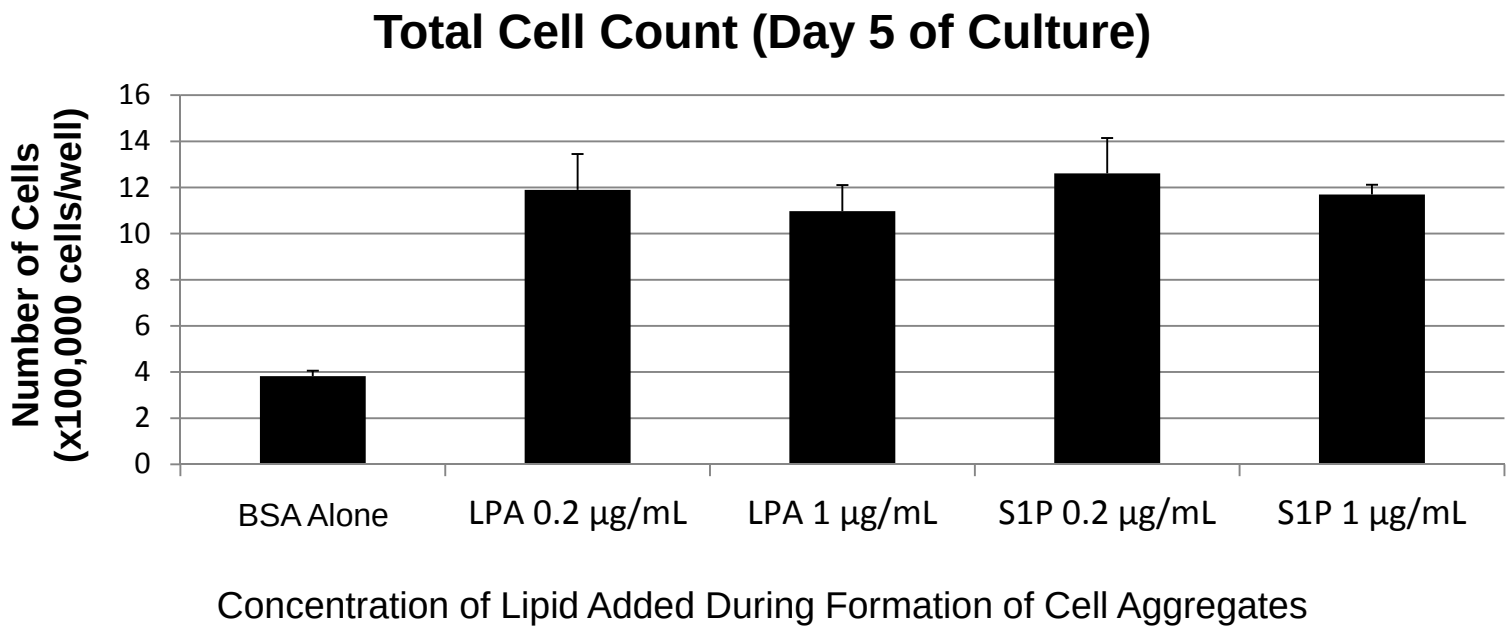

Total Cell Count (Day 5 of Culture)
Number of Cells (x100,000 cells/well)
16
14
12
10
8
6
4
2
0
BSA Alone
LPA 0.2 µg/mL
LPA 1 µg/mL
S1P 0.2 µg/mL
S1P 1 µg/mL
Concentration of Lipid Added During Formation of Cell Aggregates

Fig. 14

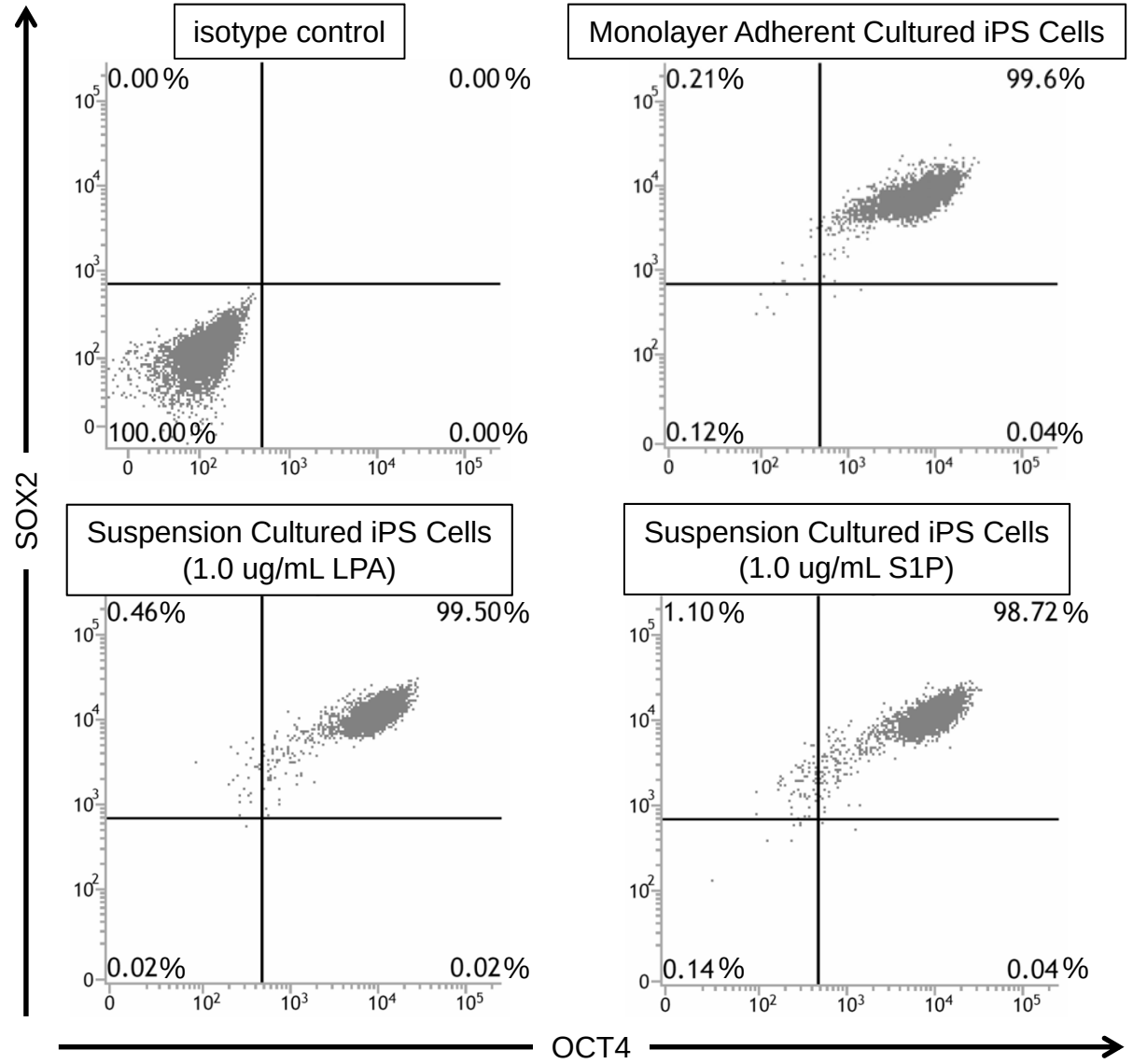

isotype control
0.00%
0.00%
100.00%
0.00%
Monolayer Adherent Cultured iPS Cells
0.21%
99.6%
0.12%
0.04%
Suspension Cultured iPS Cells
(1.0 ug/mL LPA)
0.46%
99.50%
0.02%
0.02%
Suspension Cultured iPS Cells
(1.0 ug/mL S1P)
1.10%
98.72%
0.14%
0.04%
SOX2
OCT4

Fig. 15

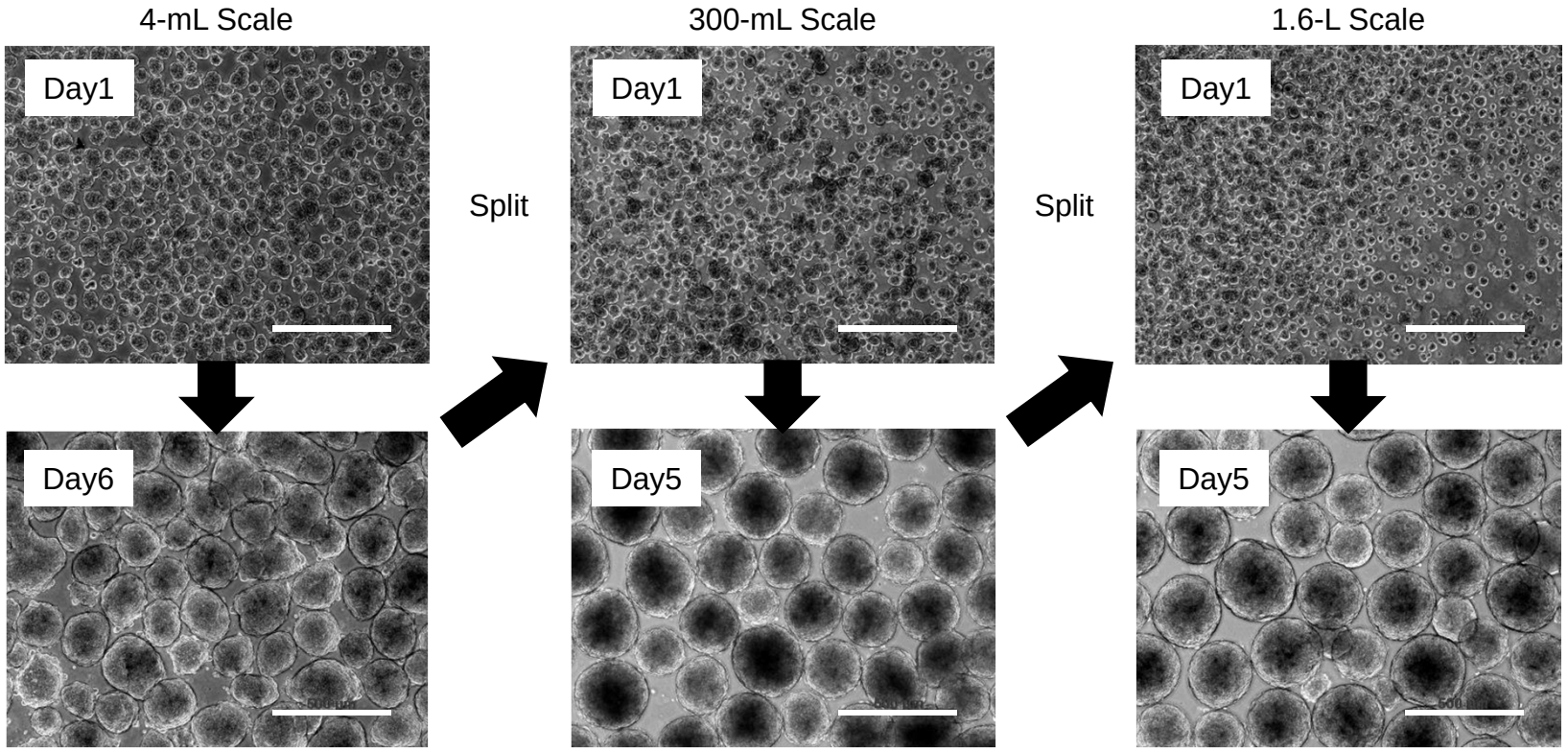

4-mL Scale
300-mL Scale
1.6-L Scale
Day1
Split
Day1
Split
Day1
Day6
Day5
Day5
Scale bar; 500um

Fig. 16

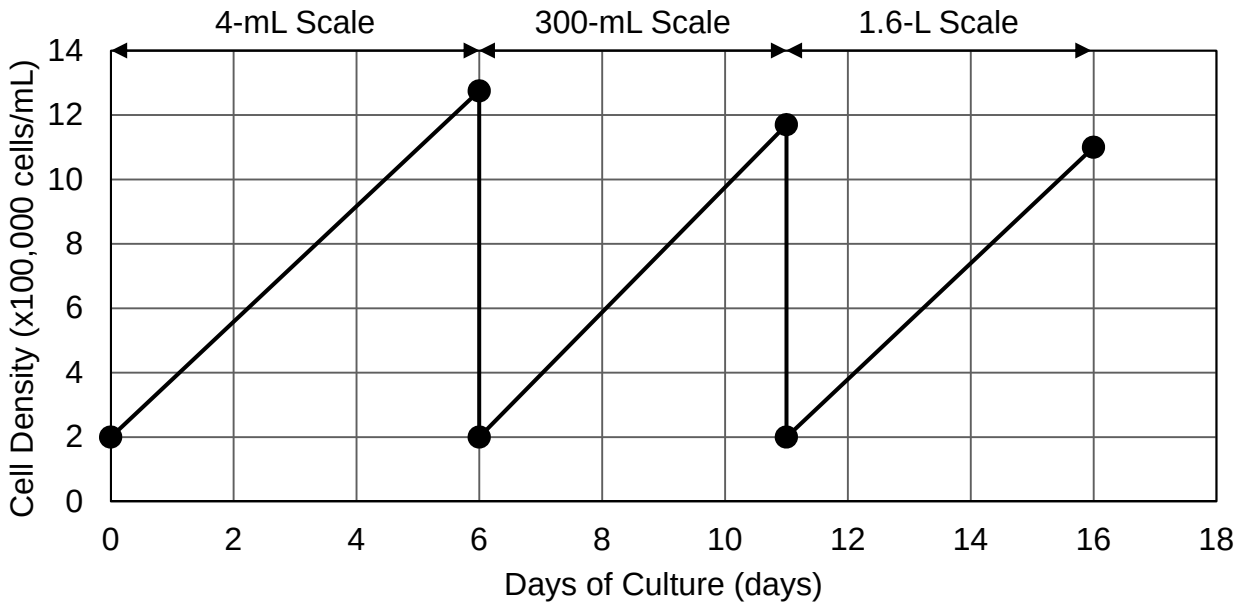
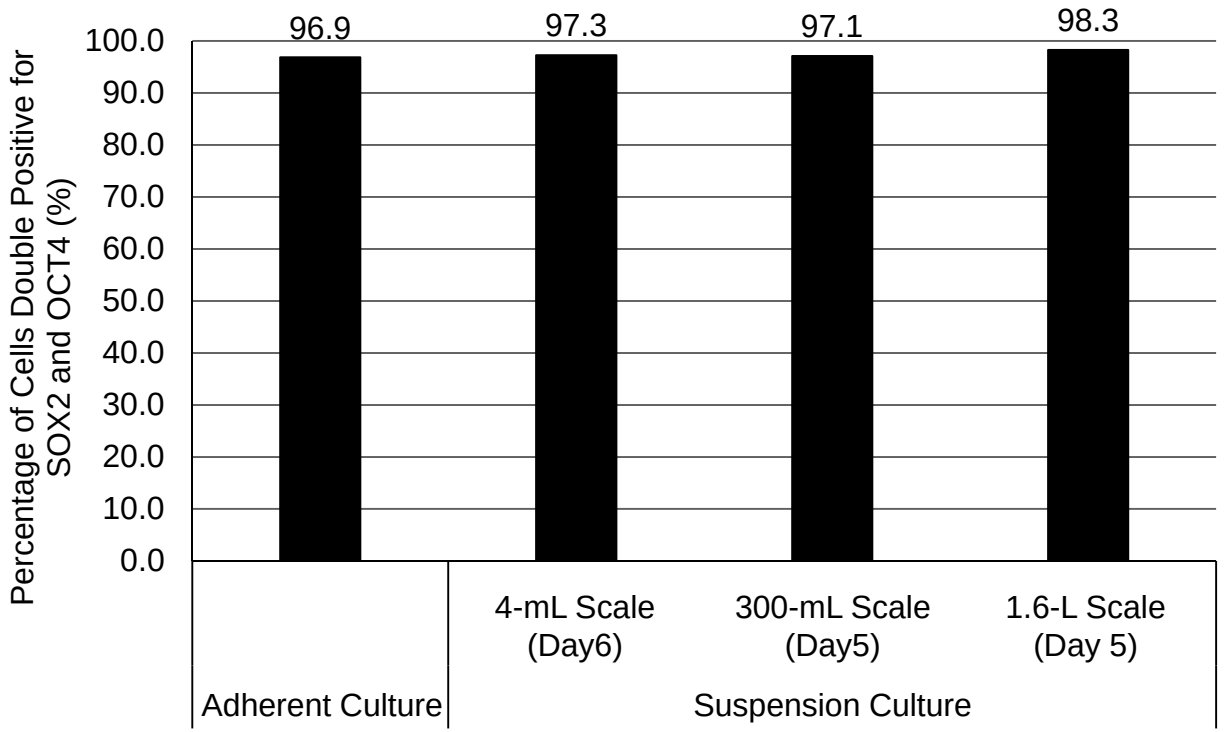

4-mL Scale
300-mL Scale
1.6-L Scale
Cell Density (x100,000 cells/mL)
0
2
4
6
8
10
12
14
0
2
4
6
8
10
12
14
16
18
Days of Culture (days)

Fig. 17


Percentage of Cells Double Positive for SOX2 and OCT4 (%)
100.0
90.0
80.0
70.0
60.0
50.0
40.0
30.0
20.0
10.0
0.0
96.9
97.3
97.1
98.3
4-mL Scale (Day6)
300-mL Scale (Day5)
1.6-L Scale (Day 5)
Adherent Culture
Suspension Culture