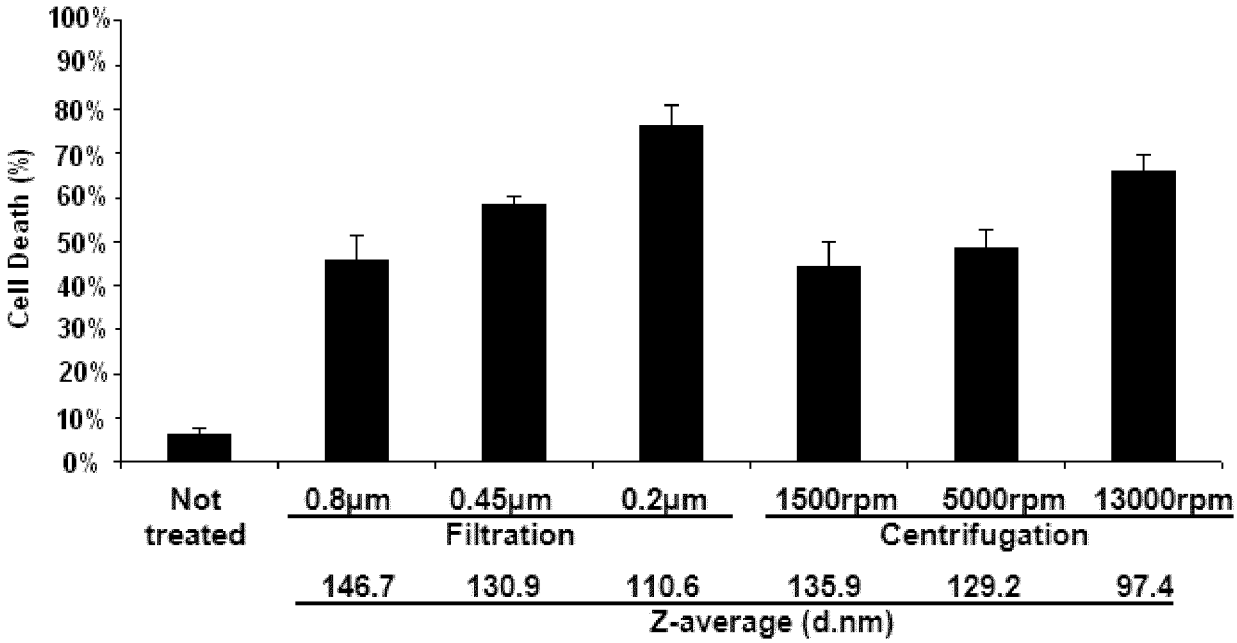
Figure 1

A)


100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
Cell Death (%)
Not treated
0.8µm
0.45µm
0.2µm
Filtration
1500rpm
5000rpm
13000rpm
Centrifugation
146.7
130.9
110.6
135.9
129.2
97.4
Z-average (d.nm)

B)

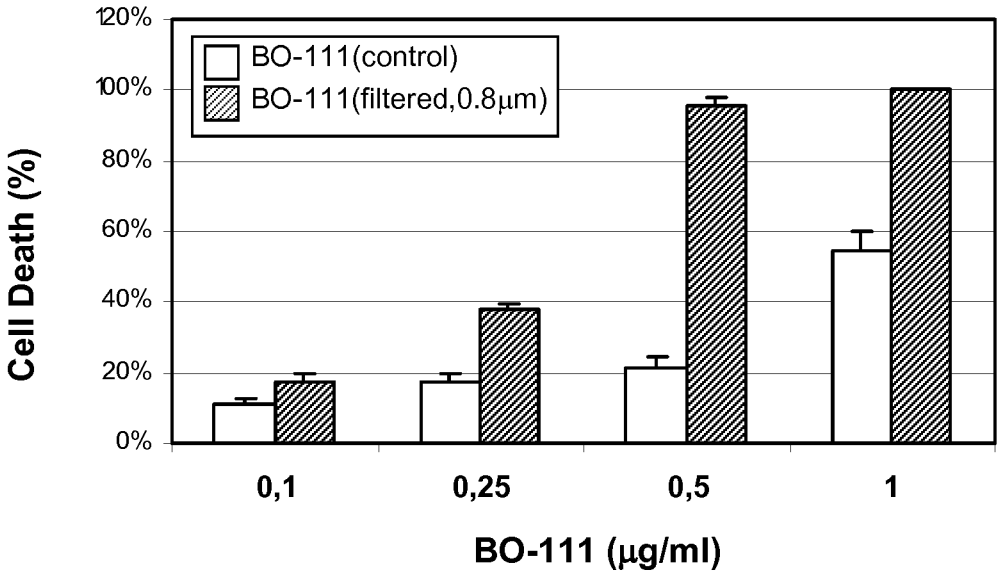
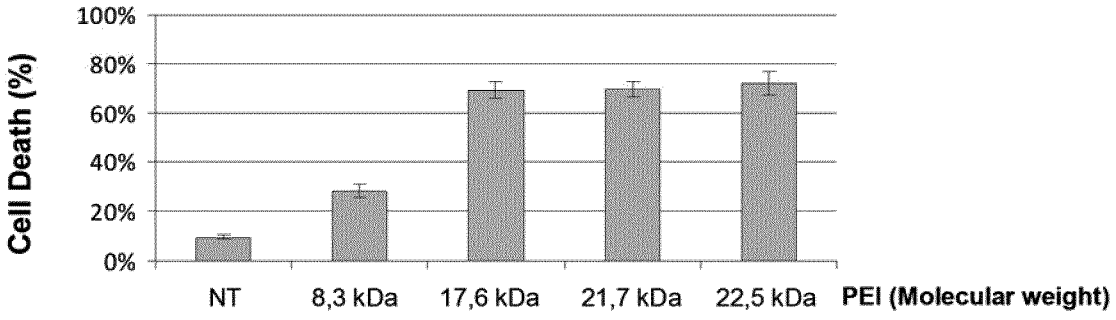

120%
100%
80%
60%
40%
20%
0%
Cell Death (%)
BO-111(control)
BO-111(filtered,0.8µm)
0,1
0,25
0,5
1
BO-111 (µg/ml)

Figure 2

A)


100%
80%
60%
40%
20%
0%
Cell Death (%)
NT
8,3 kDa
17,6 kDa
21,7 kDa
22,5 kDa
PEI (Molecular weight)

B)

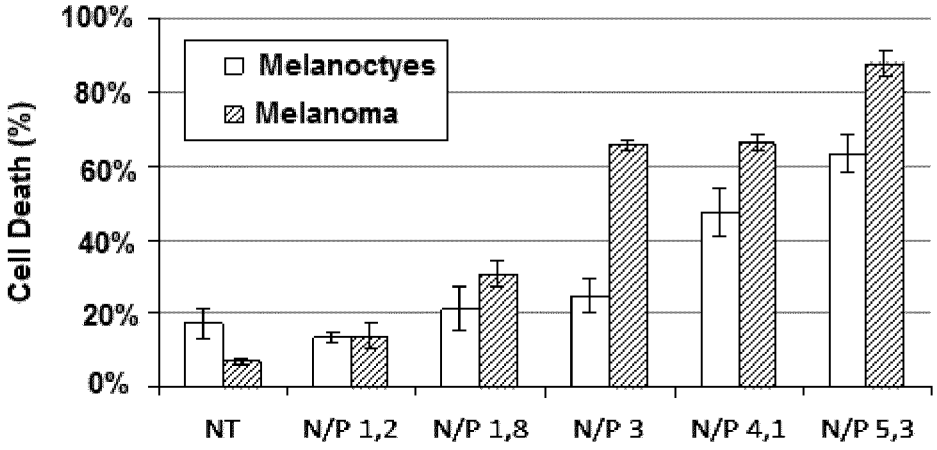

100%
80%
60%
40%
20%
0%
Cell Death (%)
Melanoctyes
Melanoma
NT
N/P 1,2
N/P 1,8
N/P 3
N/P 4,1
N/P 5,3

Figure 3

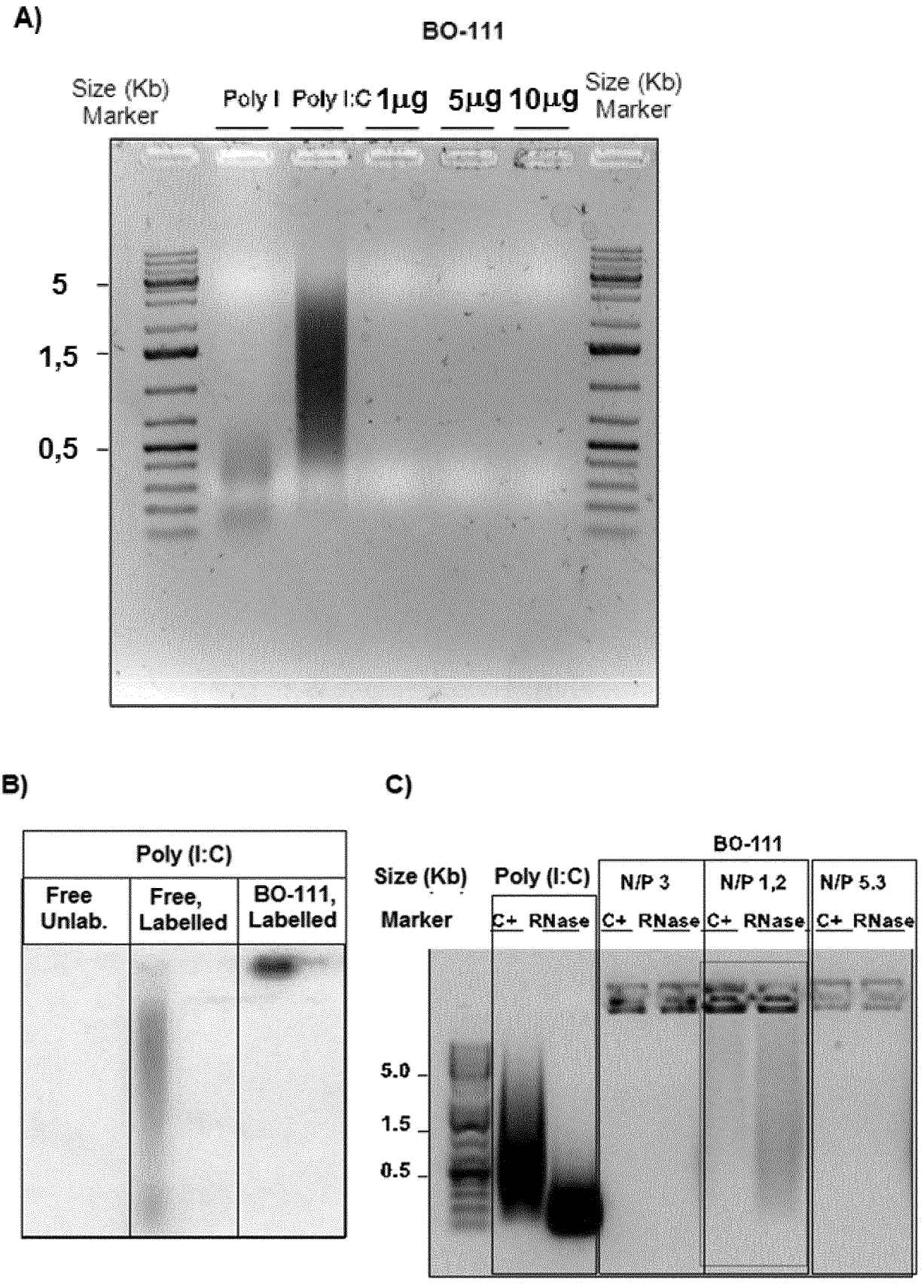

A)
BO-111
Size (Kb) Marker
Poly I
Poly I:C
1µg
5µg
10µg
Size (Kb) Marker
5
1,5
0,5
B)
Poly (I:C)
Free Unlab.
Free, Labelled
BO-111, Labelled
C)
BO-111
Size (Kb) Marker
Poly (I:C)
N/P 3
N/P 1,2
N/P 5.3
C+ RNase
C+ RNase
C+ RNase
C+ RNase
5.0
1.5
0.5

Figure 4

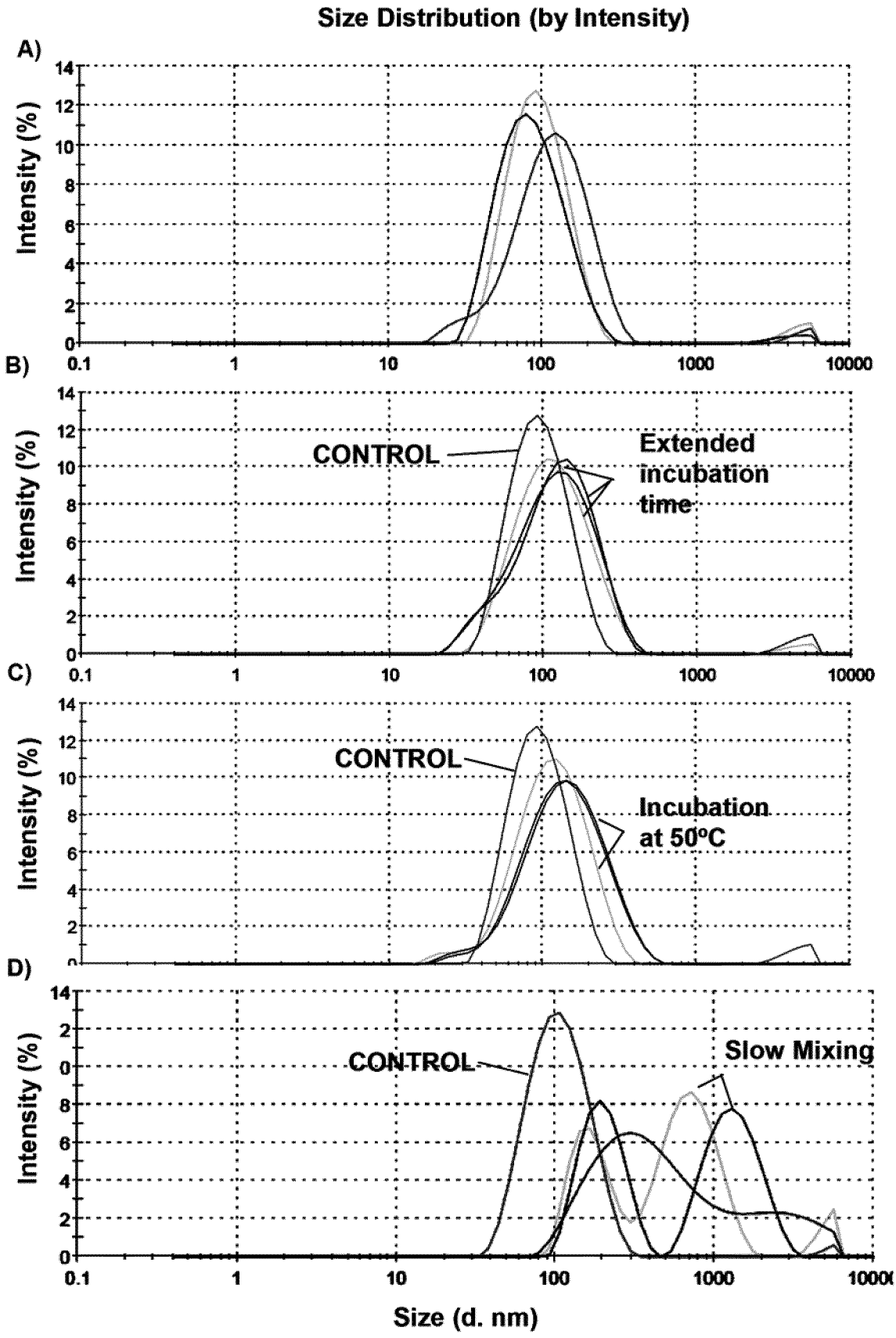
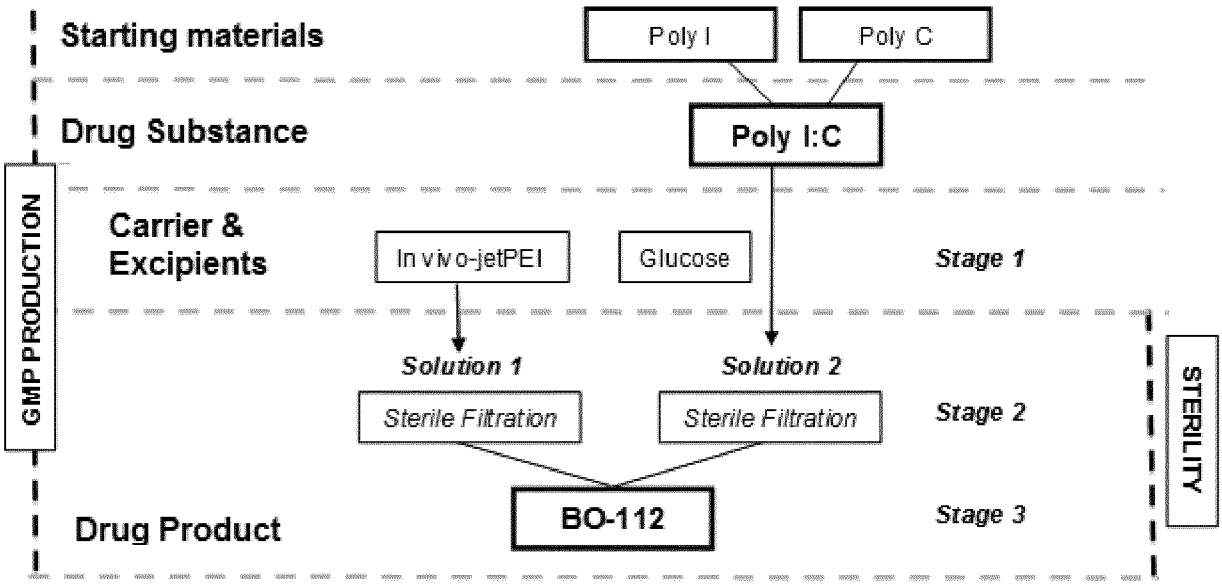
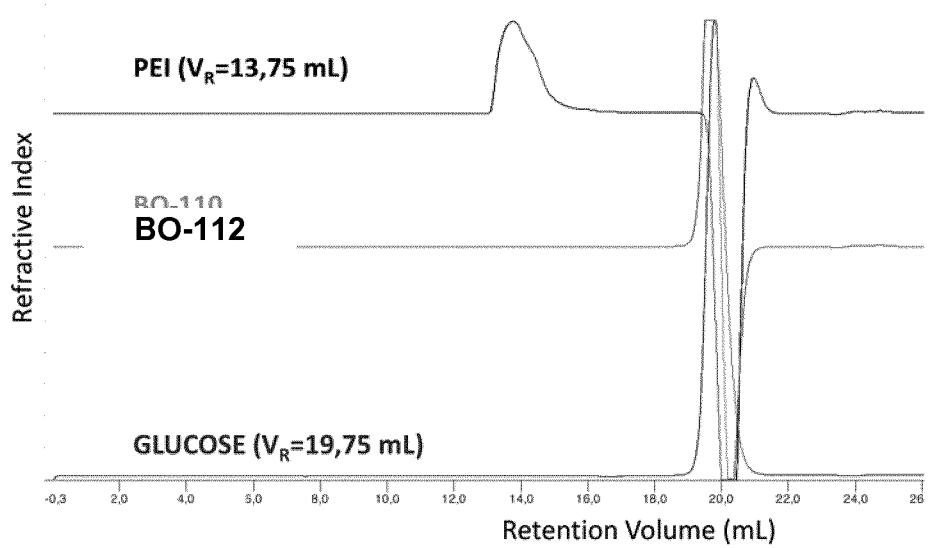

Size Distribution (by Intensity)
A)
B)
C)
D)
Intensity (%)
CONTROL
Extended incubation time
CONTROL
Incubation at 50°C
CONTROL
Slow Mixing
0.1
1
10
100
1000
10000
Size (d. nm)

Figure 5

A)


Starting materials
Poly I
Poly C
Drug Substance
Poly I:C
GMP PRODUCTION
Carrier & Excipients
In vivo-jetPEI
Glucose
Stage 1
Solution 1
Solution 2
Sterile Filtration
Sterile Filtration
Stage 2
STERILITY
BO-112
Stage 3
Drug Product

B)


PEI (V_R=13,75 mL)
BO-112
GLUCOSE (V_R=19,75 mL)
Refractive Index
-0,3
2,0
4,0
5,0
8,0
10,0
12,0
14,0
16,0
18,0
20,0
22,0
24,0
26
Retention Volume (mL)

C)

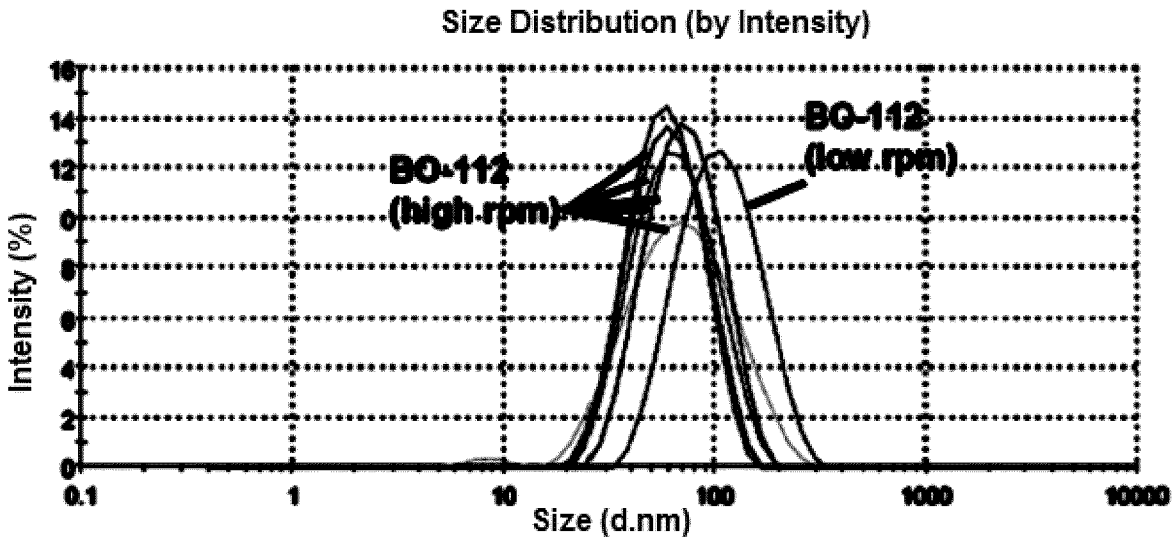
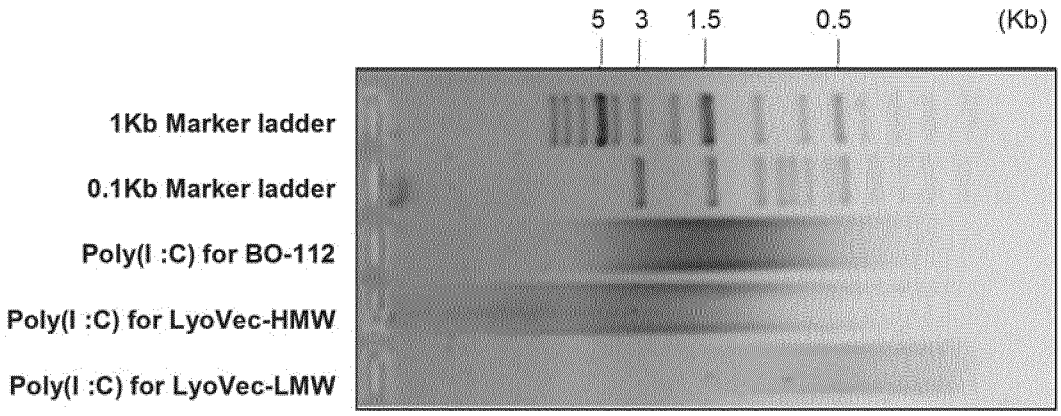

Size Distribution (by Intensity)
BO-112 (high rpm)
BO-112 (low rpm)
Intensity (%)
16
14
12
0
8
6
4
2
0
0.1
1
10
100
1000
10000
Size (d.nm)

Figure 6

A)


5
3
1.5
0.5
(Kb)
1Kb Marker ladder
0.1Kb Marker ladder
Poly(I :C) for BO-112
Poly(I :C) for LyoVec-HMW
Poly(I :C) for LyoVec-LMW

B)

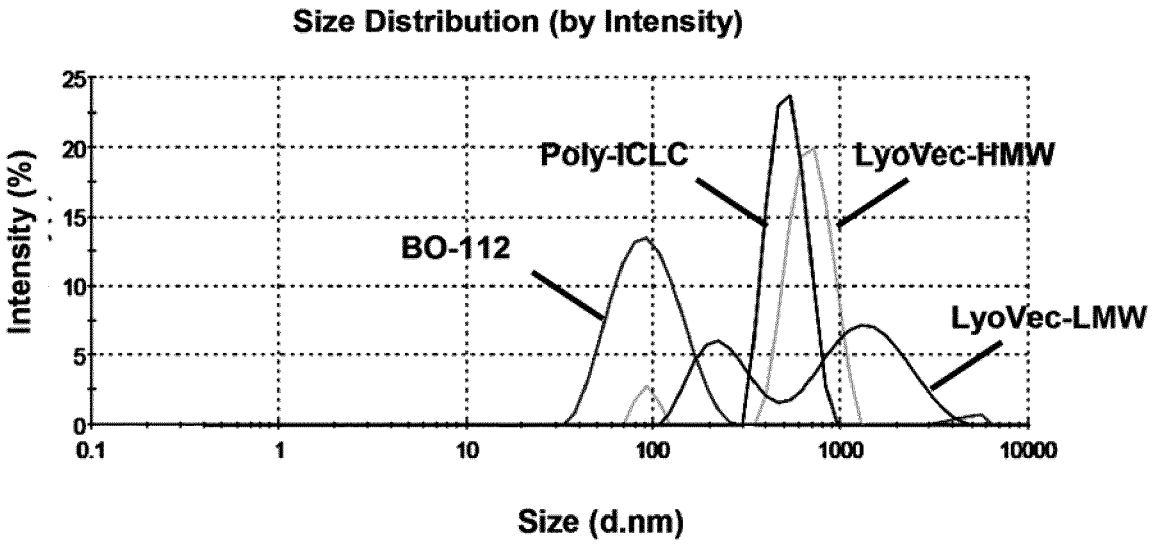

Size Distribution (by Intensity)
Intensity (%)
25
20
15
10
5
0
0.1
1
10
100
1000
10000
Size (d.nm)
Poly-ICLC
LyoVec-HMW
BO-112
LyoVec-LMW

Figure 7

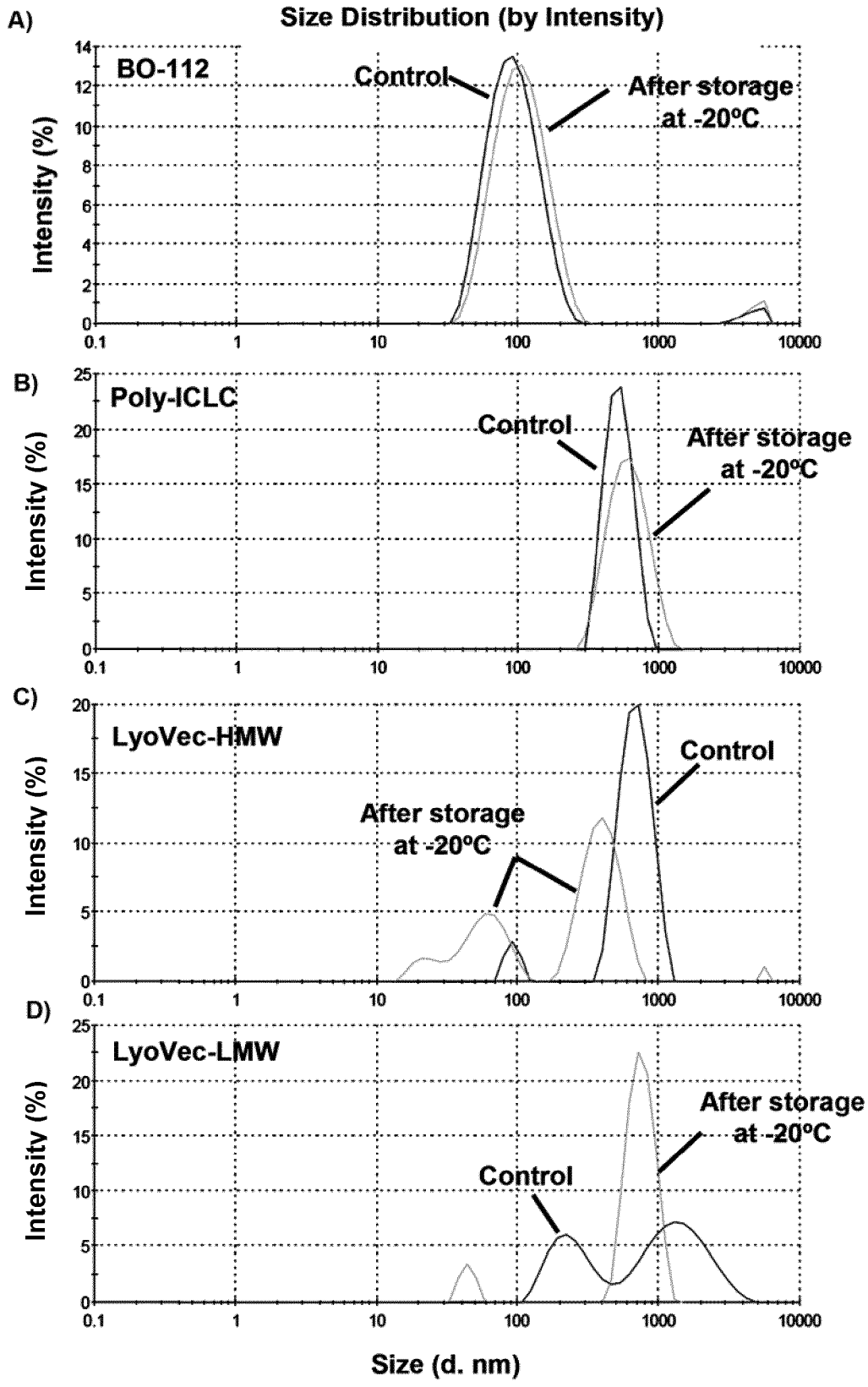
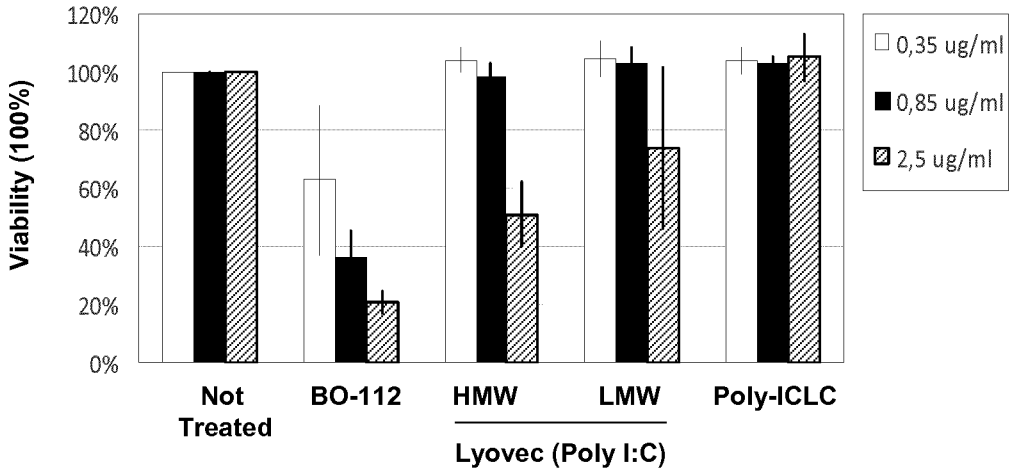
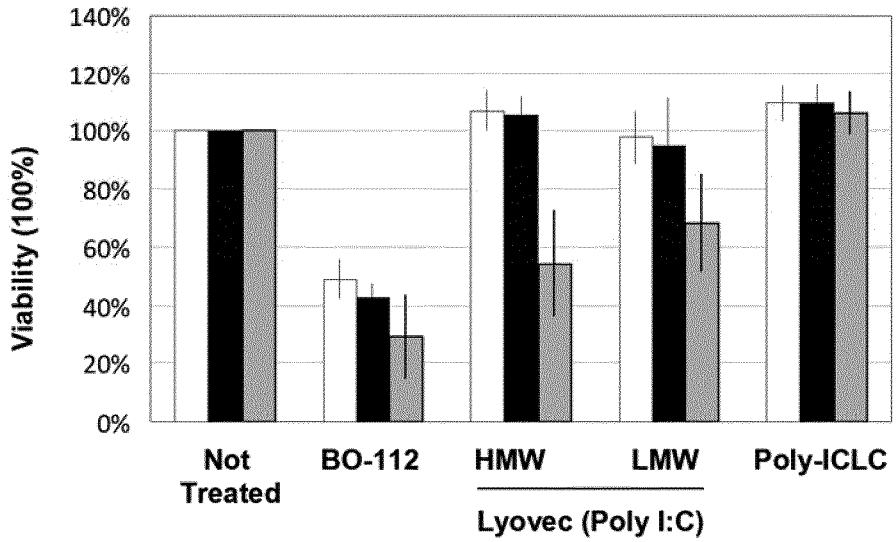

A)
Size Distribution (by Intensity)
BO-112
Control
After storage at -20ºC
Intensity (%)
B)
Poly-ICLC
Control
After storage at -20ºC
Intensity (%)
C)
LyoVec-HMW
Control
After storage at -20ºC
Intensity (%)
D)
LyoVec-LMW
After storage at -20ºC
Control
Intensity (%)
0.1
1
10
100
1000
10000
Size (d. nm)

Figure 8

A)


120%
100%
80%
60%
40%
20%
0%
Viability (100%)
0,35 ug/ml
0,85 ug/ml
2,5 ug/ml
Not Treated
BO-112
HMW
LMW
Poly-ICLC
Lyovec (Poly I:C)

B)


140%
120%
100%
80%
60%
40%
20%
0%
Viability (100%)
Not Treated
BO-112
HMW
LMW
Poly-ICLC
Lyovec (Poly I:C)

C)

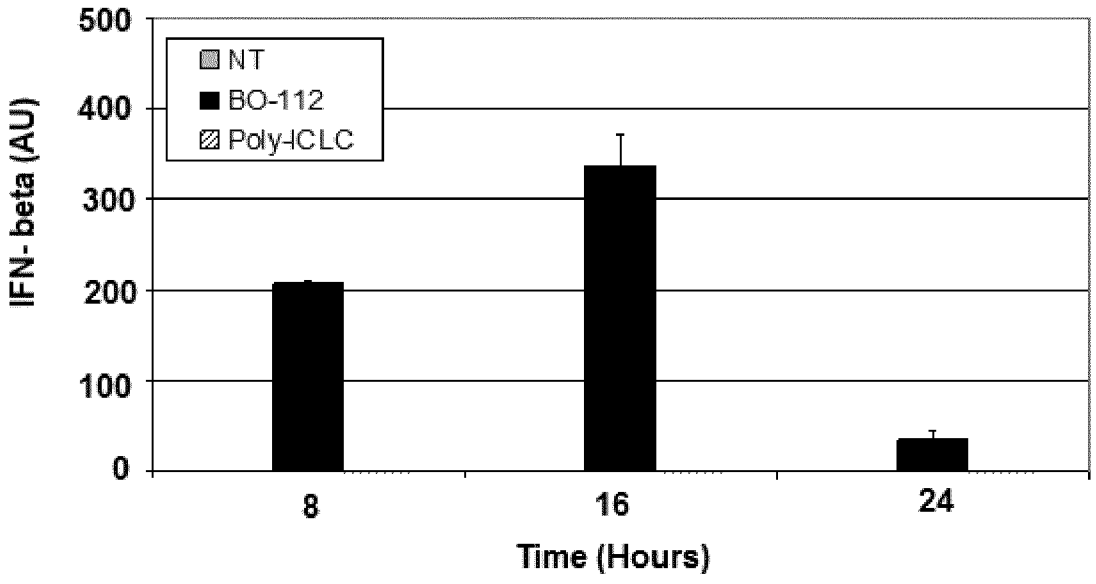

500
400
300
200
100
0
IFN- beta (AU)
NT
BO-112
Poly-ICLC
8
16
24
Time (Hours)

Figure 9

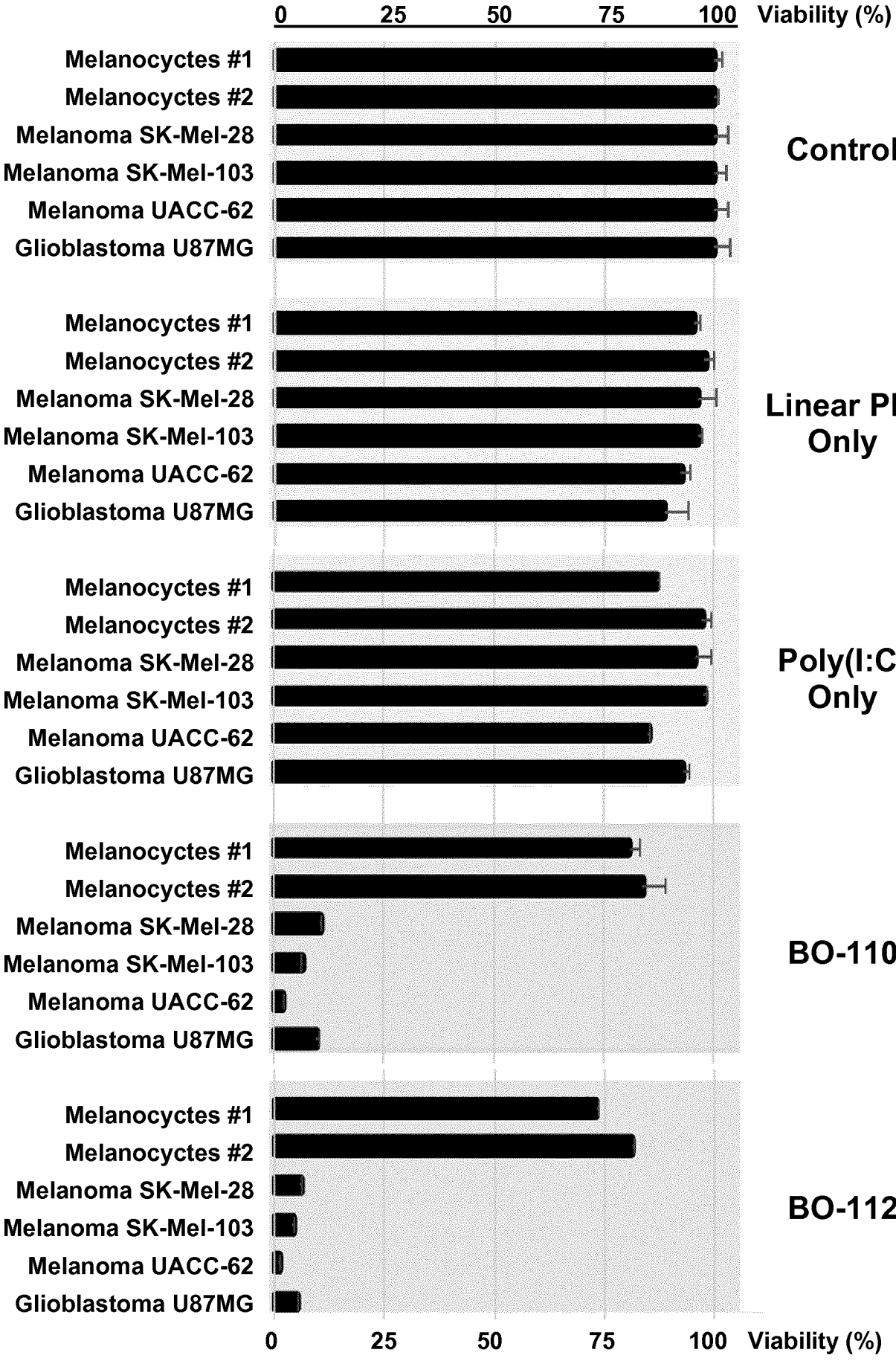
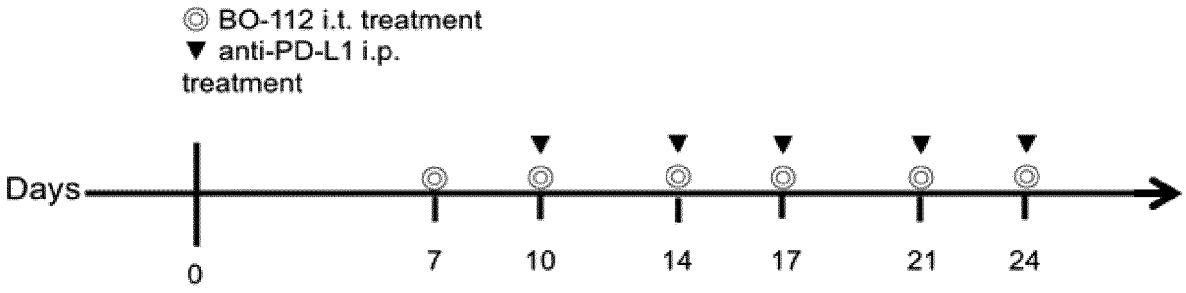
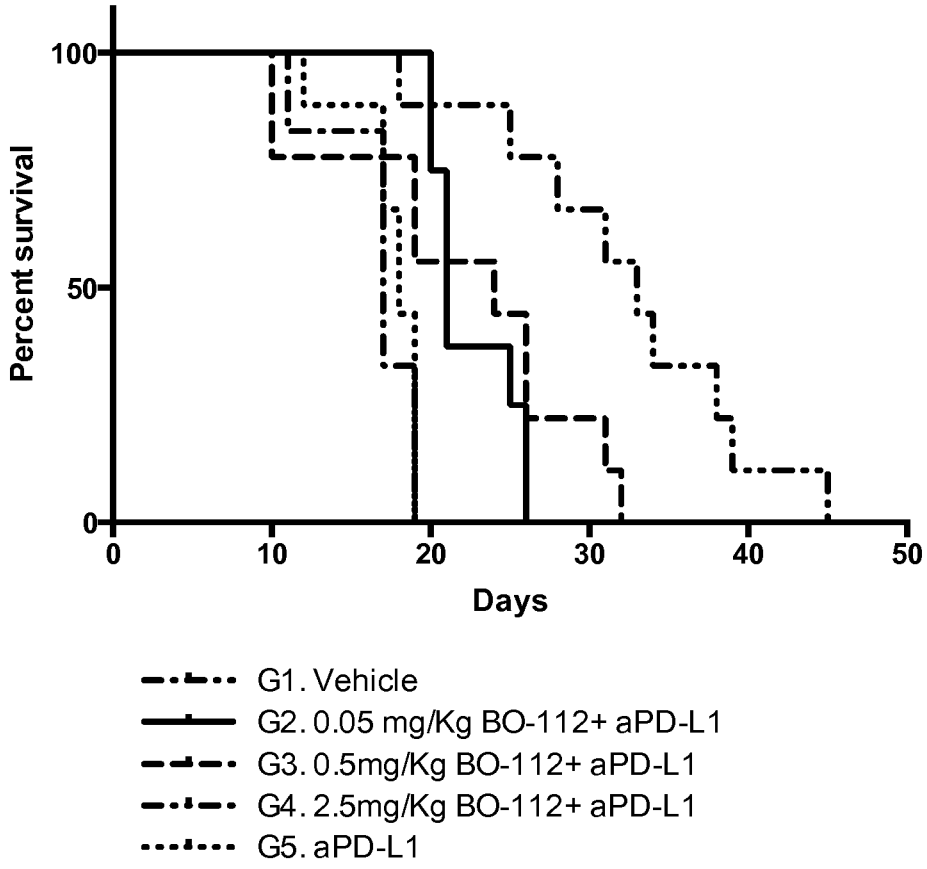

0
25
50
75
100
Viability (%)
Melanocytes #1
Melanocytes #2
Melanoma SK-Mel-28
Melanoma SK-Mel-103
Melanoma UACC-62
Glioblastoma U87MG
Control
Linear PEI Only
Poly(I:C) Only
BO-110
BO-112

Figure 10

A)


◎ BO-112 i.t. treatment
▼ anti-PD-L1 i.p. treatment
Days
0
7
10
14
17
21
24

B)


Percent survival
100
50
0
0
10
20
30
40
50
Days
G1. Vehicle
G2. 0.05 mg/Kg BO-112+ aPD-L1
G3. 0.5mg/Kg BO-112+ aPD-L1
G4. 2.5mg/Kg BO-112+ aPD-L1
G5. aPD-L1