Figure 1.

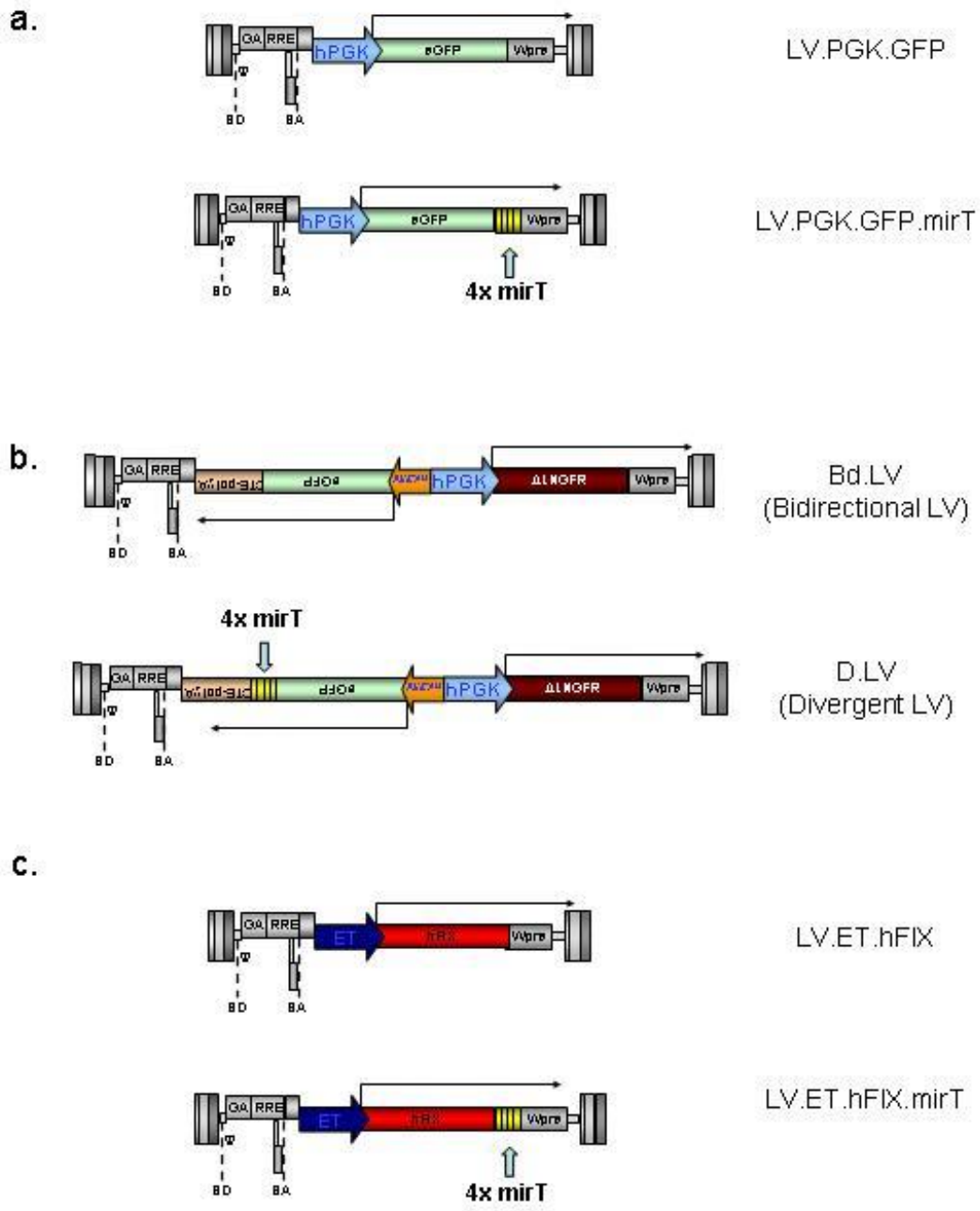
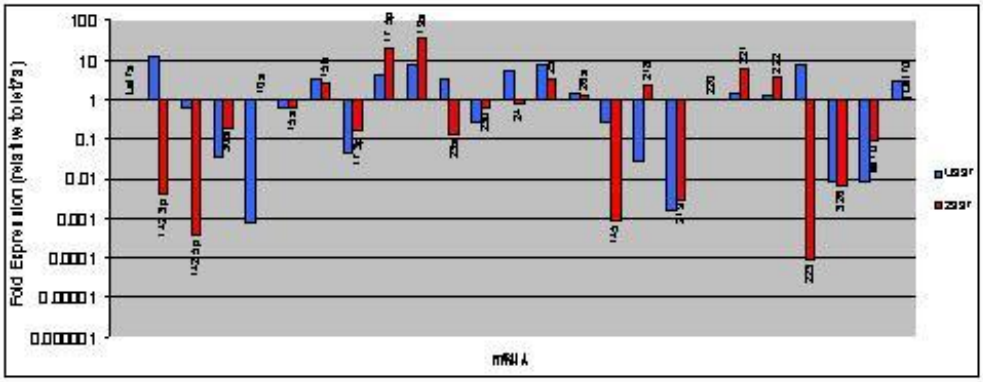
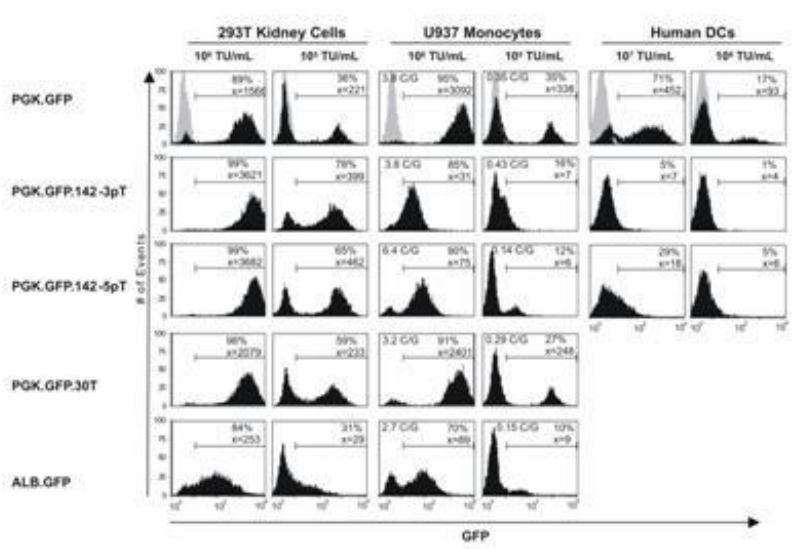

a.
GA
RRE
hPGK
eGFP
Wpre
Ψ
BD
BA
LV.PGK.GFP
LV.PGK.GFP.mirT
4x mirT
b.
ΔLNGFR
Bd.LV
(Bidirectional LV)
D.LV
(Divergent LV)
c.
ET
LV.ET.hFIX
LV.ET.hFIX.mirT

Figure 2.

a.


Fold Expression (relative to let7a)
100
10
1
0.1
0.01
0.001
0.0001
0.00001
0.000001
mRNA

b.


293T Kidney Cells
U937 Monocytes
Human DCs
PGK.GFP
PGK.GFP.142-3pT
PGK.GFP.142-5pT
PGK.GFP.30T
ALB.GFP
# of Events
GFP

c.

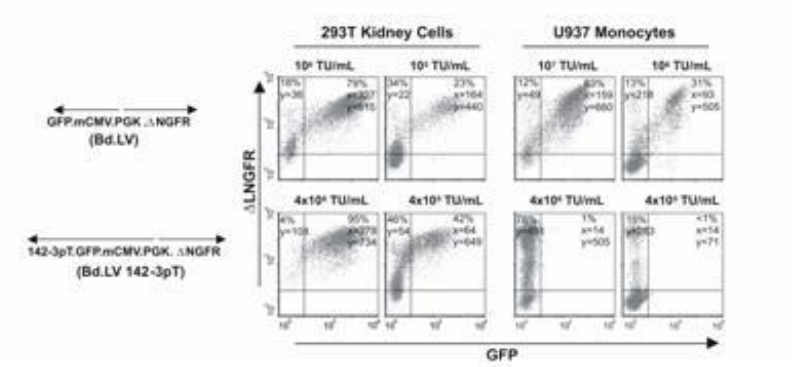

293T Kidney Cells
U937 Monocytes
GFP.mCMV.PGK .ΔNGFR
(Bd.LV)
142-3pT.GFP.mCMV.PGK. .ΔNGFR
(Bd.LV 142-3pT)
ΔLNGFR
GFP

Figure 2.

d.

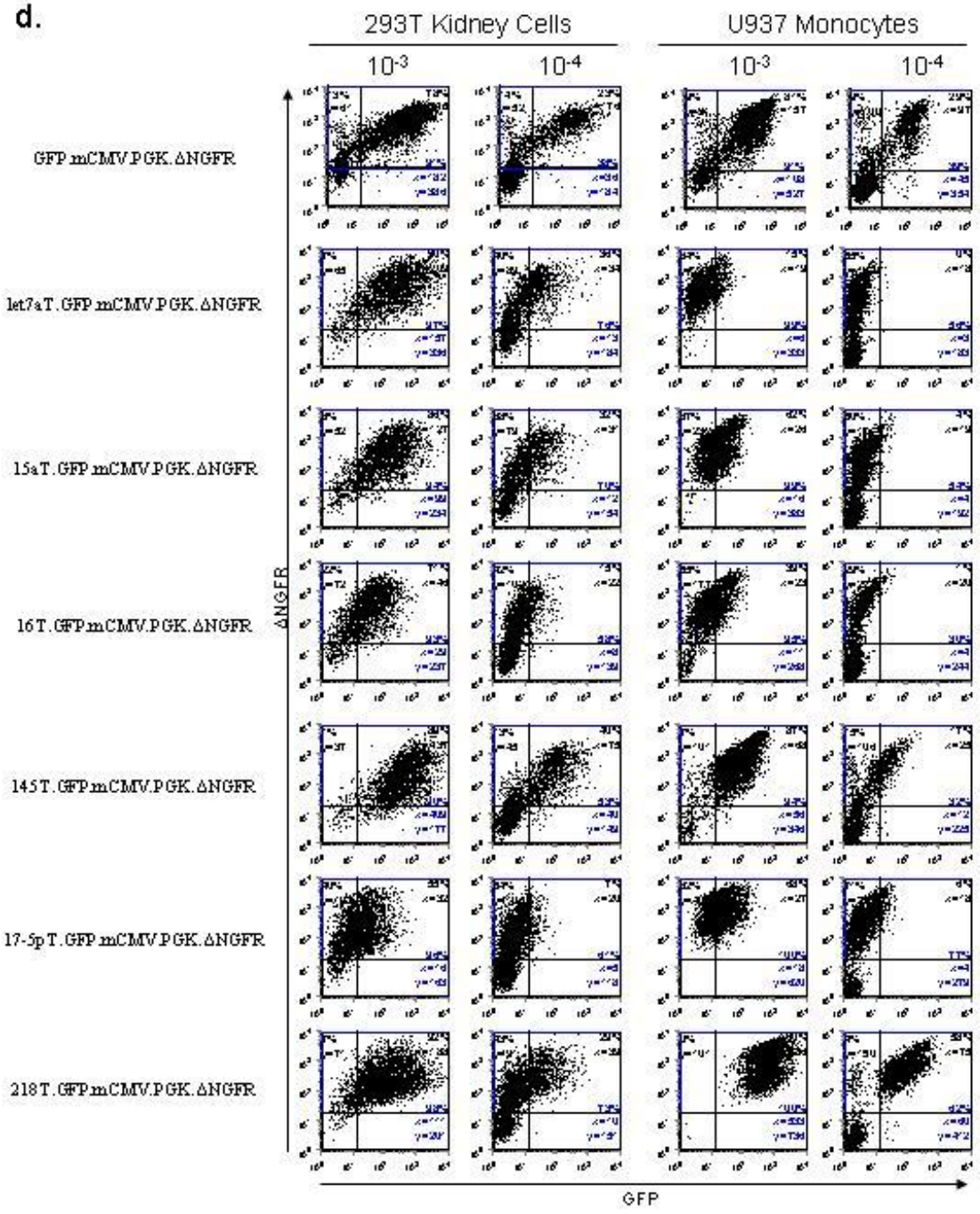

293T Kidney Cells
U937 Monocytes
10-3
10-4
10-3
10-4
GFP.mCMV.PGK.ΔNGFR
let7aT.GFP.mCMV.PGK.ΔNGFR
15aT.GFP.mCMV.PGK.ΔNGFR
16T.GFP.mCMV.PGK.ΔNGFR
145T.GFP.mCMV.PGK.ΔNGFR
17-5pT.GFP.mCMV.PGK.ΔNGFR
218T.GFP.mCMV.PGK.ΔNGFR
ΔNGFR
GFP

Figure 3.

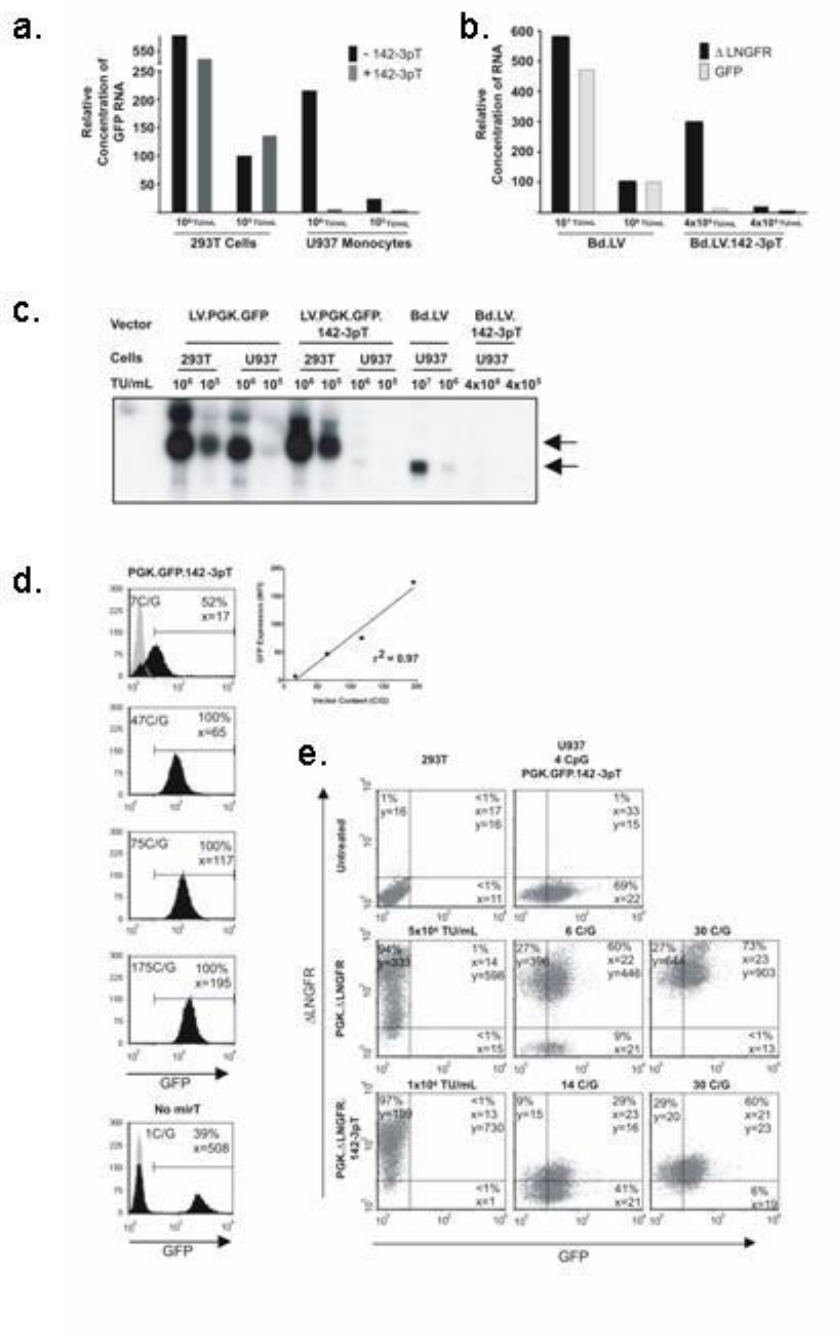

a.
Relative Concentration of GFP RNA
- 142-3pT
+ 142-3pT
293T Cells
U937 Monocytes
b.
Relative Concentration of RNA
ΔLNGFR
GFP
Bd.LV
Bd.LV.142-3pT
c.
Vector
LV.PGK.GFP
LV.PGK.GFP. 142-3pT
Bd.LV
Bd.LV. 142-3pT
Cells
293T
U937
TU/mL
d.
PGK.GFP.142-3pT
7C/G 52% x=17
47C/G 100% x=65
75C/G 100% x=117
175C/G 100% x=195
No mir T
1C/G 39% x=508
GFP
e.
293T
U937 4 CpG PGK.GFP.142-3pT
Untreated
5x10⁵ TU/mL
6 C/G
30 C/G
1x10⁵ TU/mL
14 C/G
30 C/G
PGK-ΔLNGFR
PGK-ΔLNGFR. 142-3pT
ΔLNGFR
GFP

Figure 4.

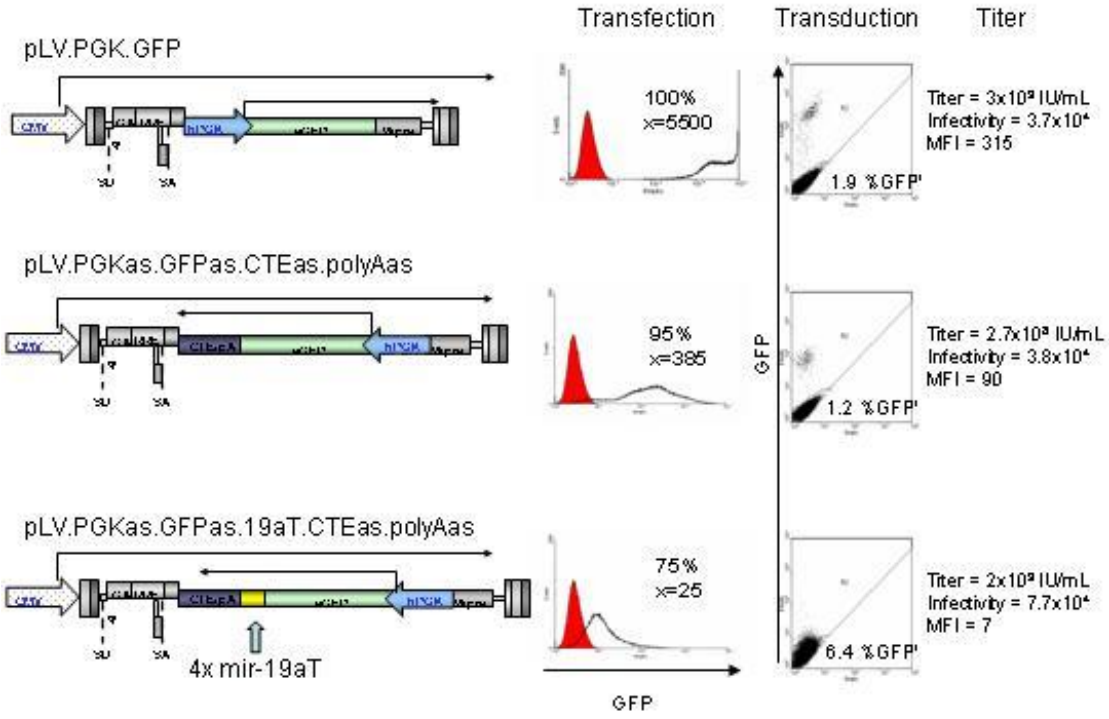

Transfection
Transduction
Titer
pLV.PGK.GFP
100%
x=5500
Titer = 3x10⁸ IU/mL
Infectivity = 3.7x10⁴
MFI = 315
1.9 %GFP
pLV.PGKas.GFPas.CTEas.polyAas
95%
x=385
Titer = 2.7x10⁸ IU/mL
Infectivity = 3.8x10⁴
MFI = 90
1.2 %GFP
pLV.PGKas.GFPas.19aT.CTEas.polyAas
4x mir-19aT
75%
x=25
Titer = 2x10⁸ IU/mL
Infectivity = 7.7x10⁴
MFI = 7
6.4 %GFP
GFP
GFP

Figure 5.

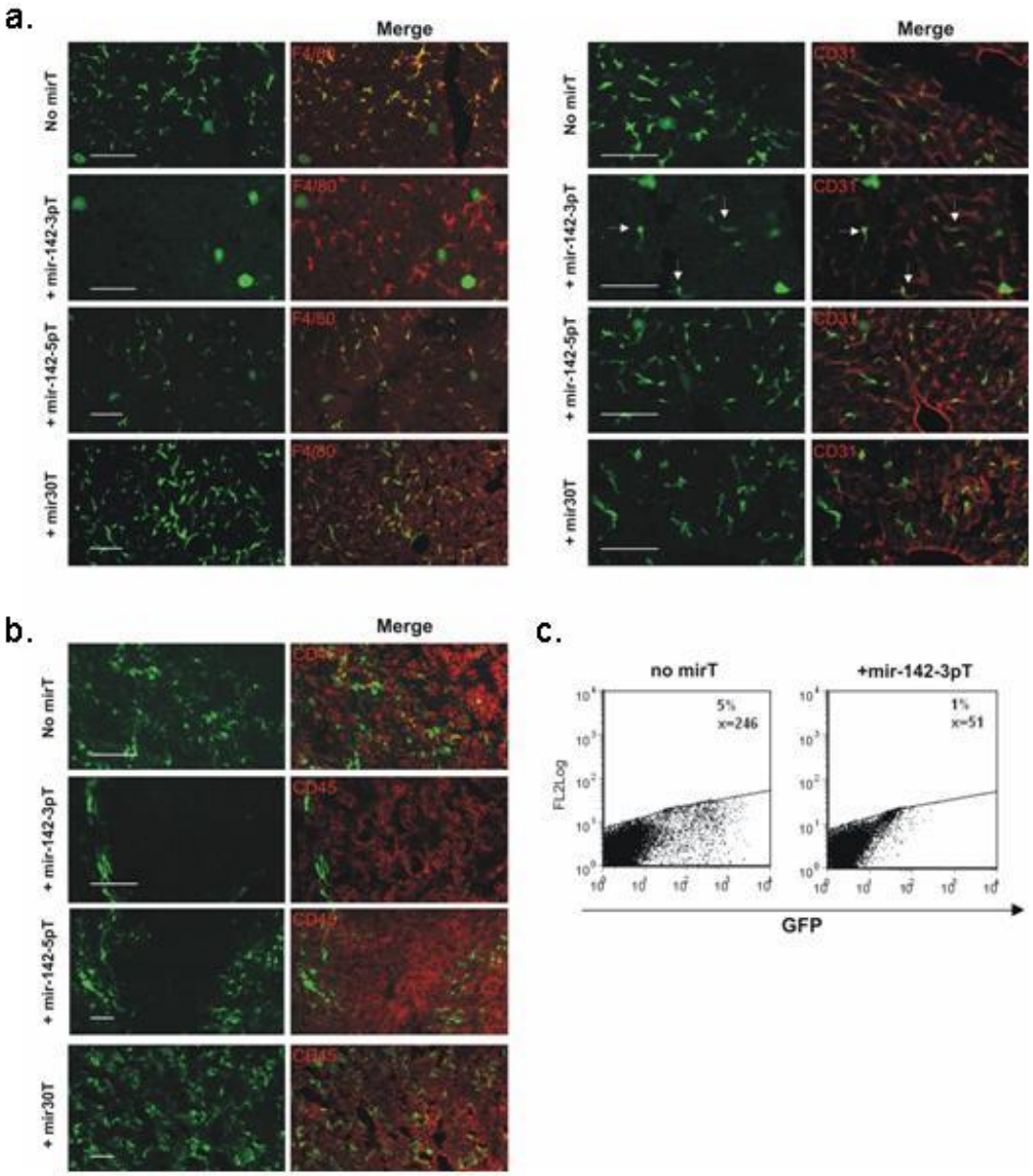

a.
Merge
No mirT
+ mir-142-3pT
+ mir-142-5pT
+ mir30T
F4/80
CD31
b.
CD45
c.
no mirT
+mir-142-3pT
5%
x=246
1%
x=51
FL2Log
GFP

Figure 6

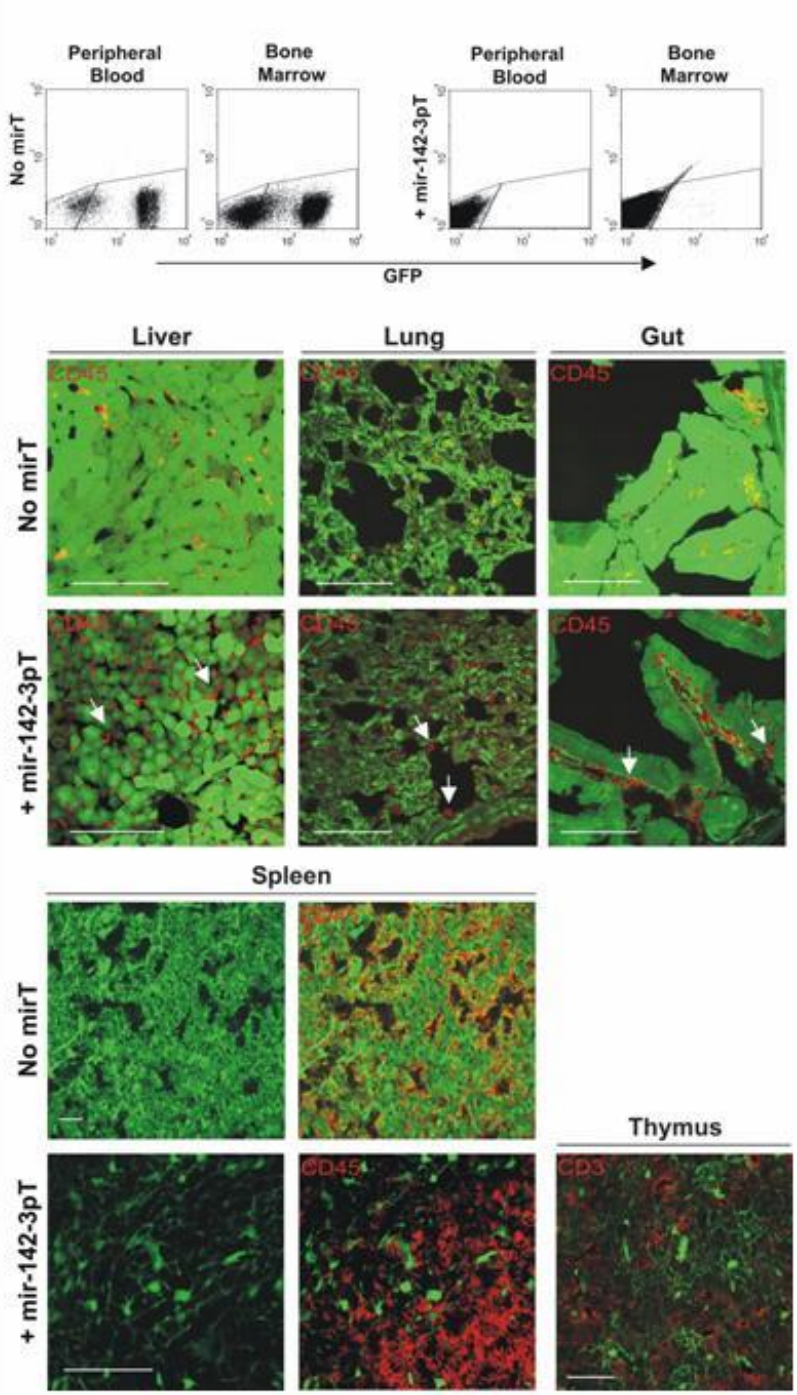
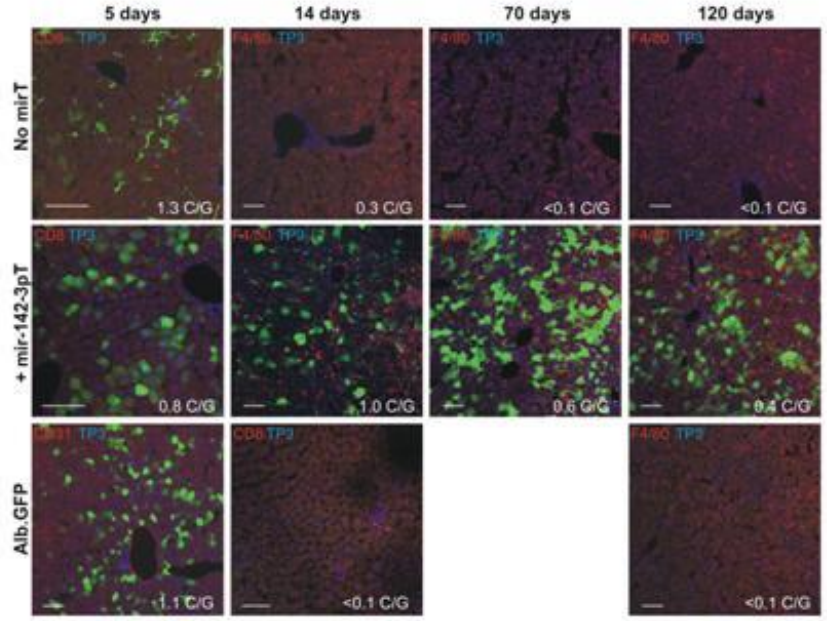
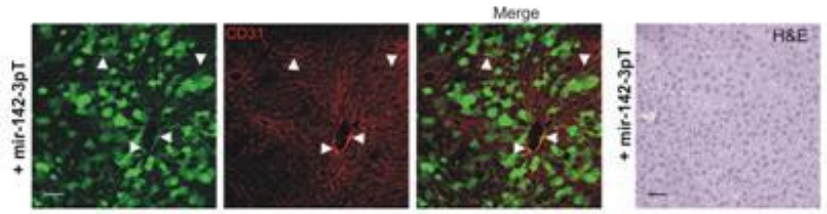

a.
Peripheral Blood
Bone Marrow
Peripheral Blood
Bone Marrow
No mirT
+ mir-142-3pT
GFP
b.
Liver
Lung
Gut
No mirT
+ mir-142-3pT
CD45
Spleen
Thymus
CD3

Figure 7.

a.


5 days
14 days
70 days
120 days
No mirT
+ mir-142-3pT
Alb.GFP
1.3 C/G
0.3 C/G
<0.1 C/G
<0.1 C/G
0.8 C/G
1.0 C/G
0.6 C/G
0.4 C/G
1.1 C/G
<0.1 C/G
<0.1 C/G

b.


Merge
+ mir-142-3pT
CD31
H&E
+ mir-142-3pT

d.

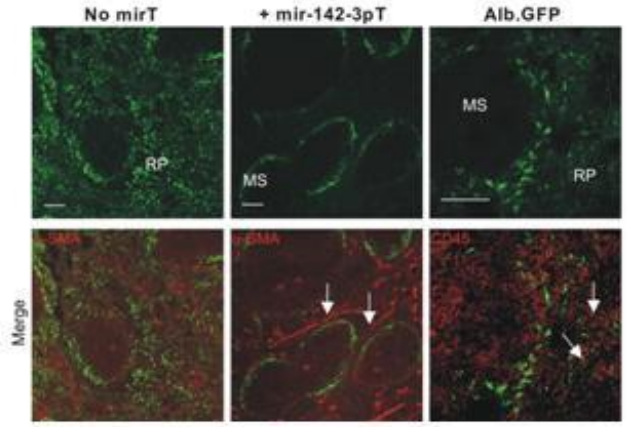
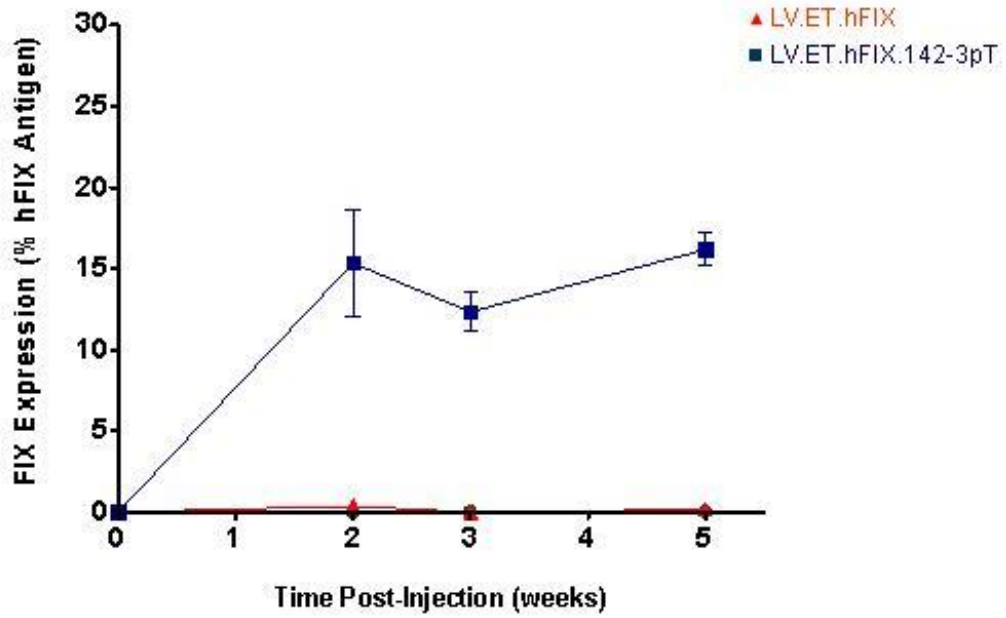

No mirT
+ mir-142-3pT
Alb.GFP
RP
MS
MS
RP
Merge

Figure 8.


LV.ET.hFIX
LV.ET.hFIX.142-3pT
FIX Expression (% hFIX Antigen)
30
25
20
15
10
5
0
0
1
2
3
4
5
Time Post-Injection (weeks)

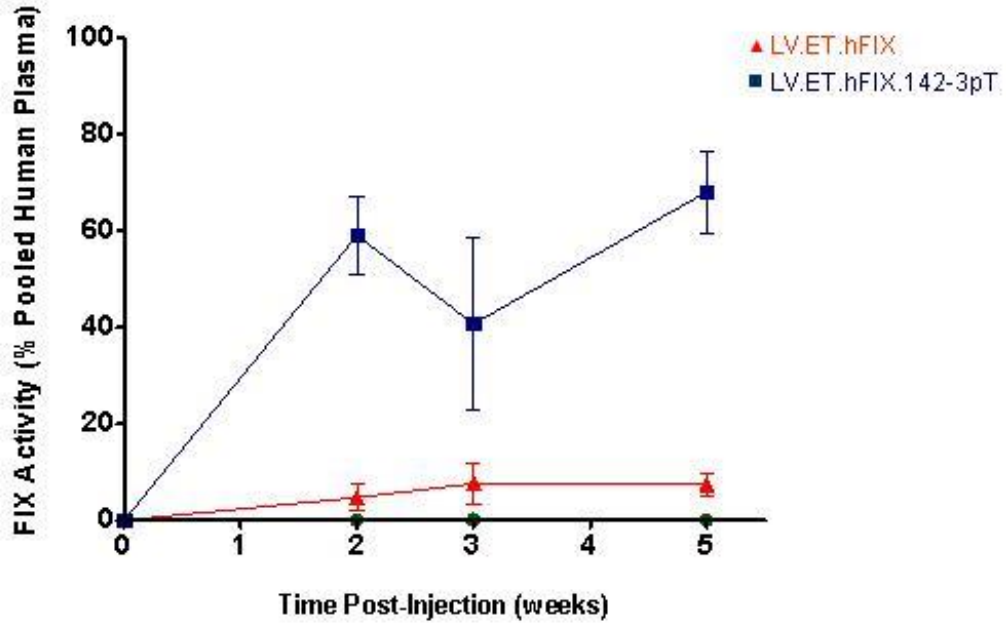
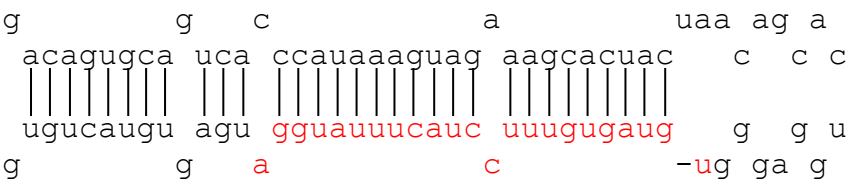

LV.ET.hFIX
LV.ET.hFIX.142-3pT
FIX Activity (% Pooled Human Plasma)
100
80
60
40
20
0
0
1
2
3
4
5
Time Post-Injection (weeks)

Figure 9

A. Mature hsa-mir-142 stem loop sequence.



B. 1x.mir-142as.Target

TCCATAAAGTAGGAAACACTACA