

Figure 1A. Commonly vector production with serum by state of art processes.

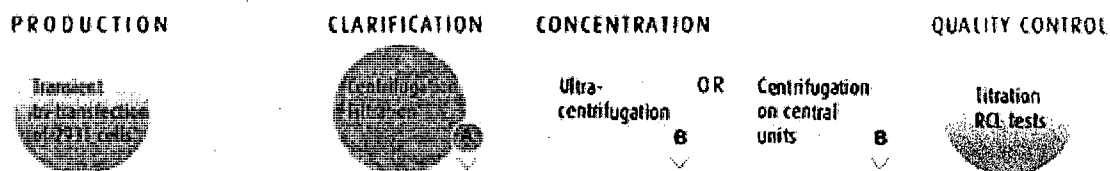


Figure 1B. Viral vector concentration and purification process.

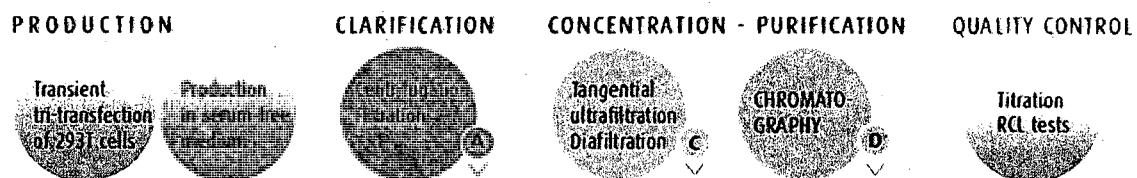
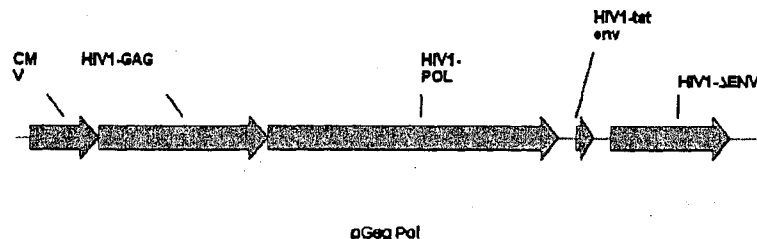
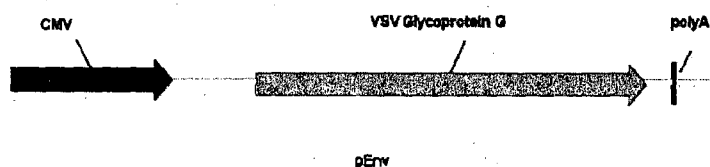


Figure 1C. Plasmid harboring gag and pol genes, Envelope expressing helper plasmid and Transgene expression plasmid without cDNA.



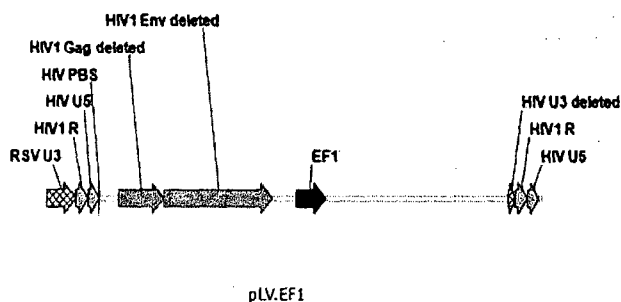
Plasmid harboring gag and pol genes

FEATURES	Length (bp)	
promoter	602	//label=CMV
CDS	1502	//label=HIV1_gag
CDS	2737	//label=HIV1-pol
CDS	1173	//label=HIV1-env deleted
protein_bind	204	//label=HIV1_RRE



Envelope expressing helper plasmid

FEATURES	Length (bp)	
Promoter	677	//label=CMV
CDS	1642	//label=VSV_glycoprotein_G
polyA_signal	768	//label=HBB2



Transgene expression plasmid without cDN

FEATURES	Length (bp)	
repeat_region	230	//label=RSV_U3
Feature	119	//label= packaging signal
CDS	362	//label=HIV1_gag_deleted
Promoter	236	//label=EF1_promoter
Feature	18	//label=Primer Binding Site PBS
repeat_unit	55	//label=HIV_U3 deleted
repeat_unit	95	//label=HIV_R
repeat_unit	85	//label=HIV_U5

Figure 2A. Summary of batch B and C titers used for the transcriptomics study.

	Titer (TU/mL) (1)	PP/TU (2)
rLV-EF1 batch B	4,7E+08	104
rLV-EF1 batch C	8,9E+09	67

Figure 2B. Foreskin cells growth 48h post-transduction with batch B and C vectors.

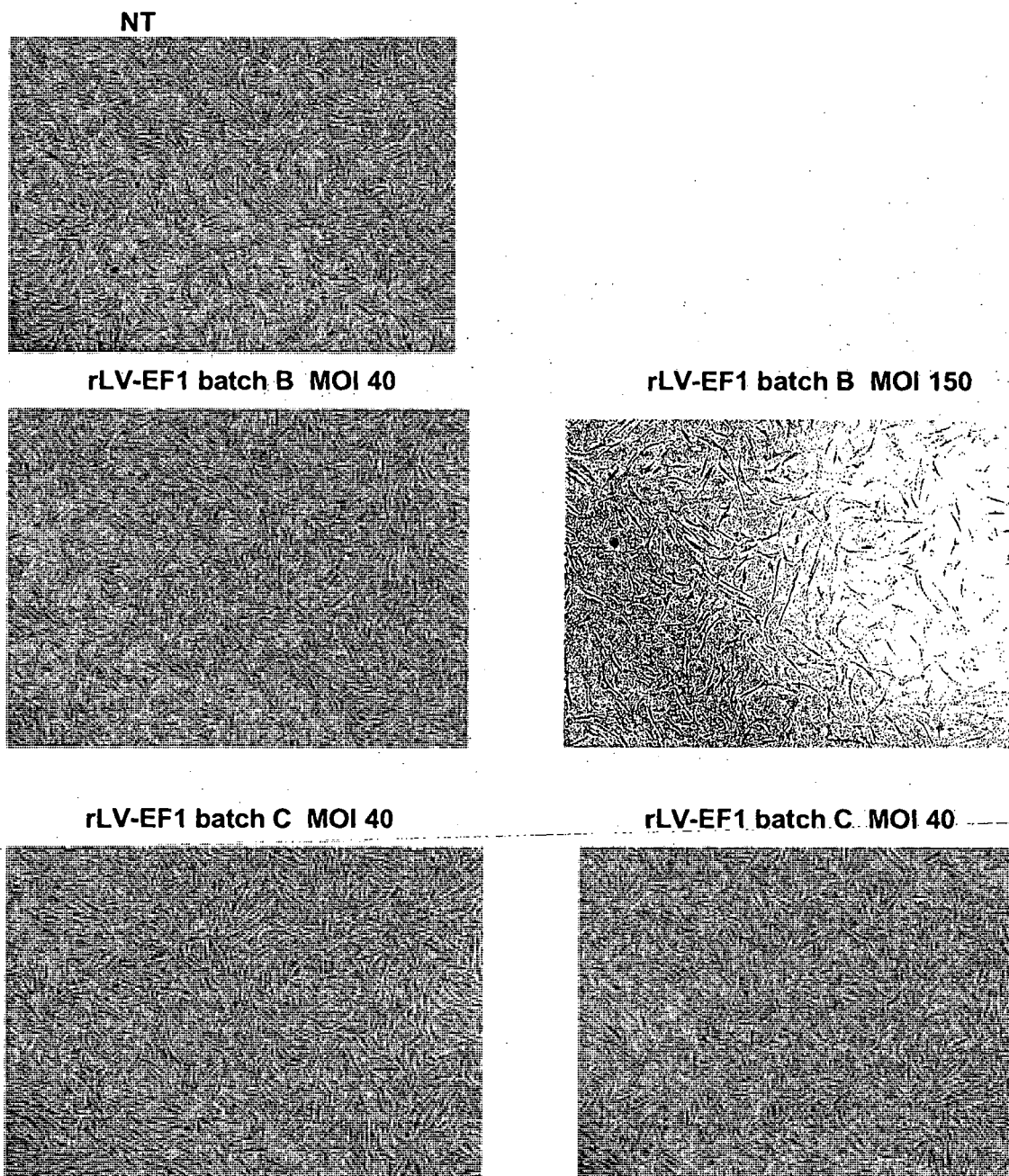
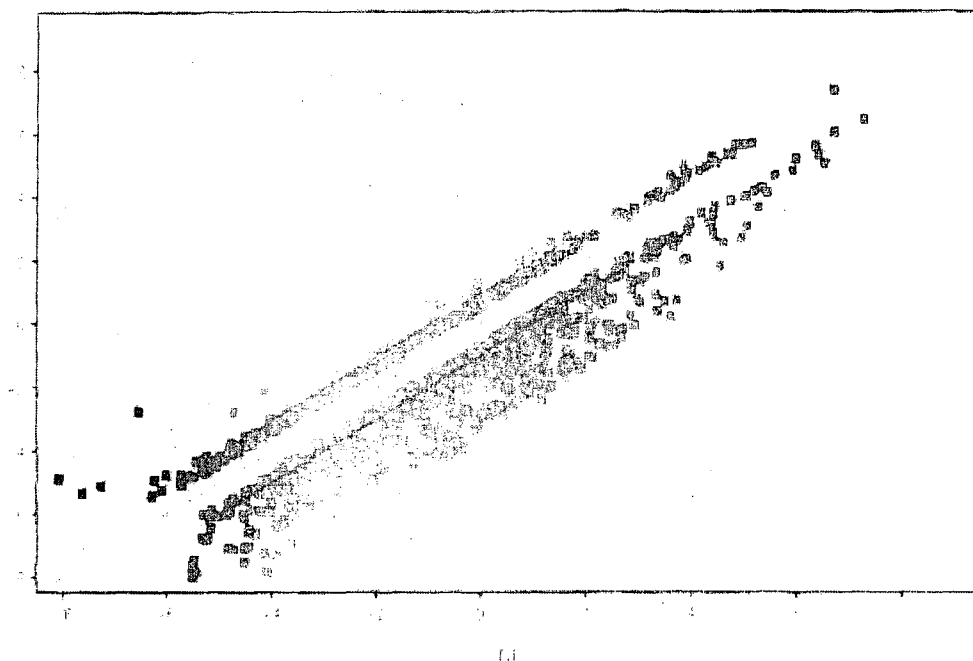
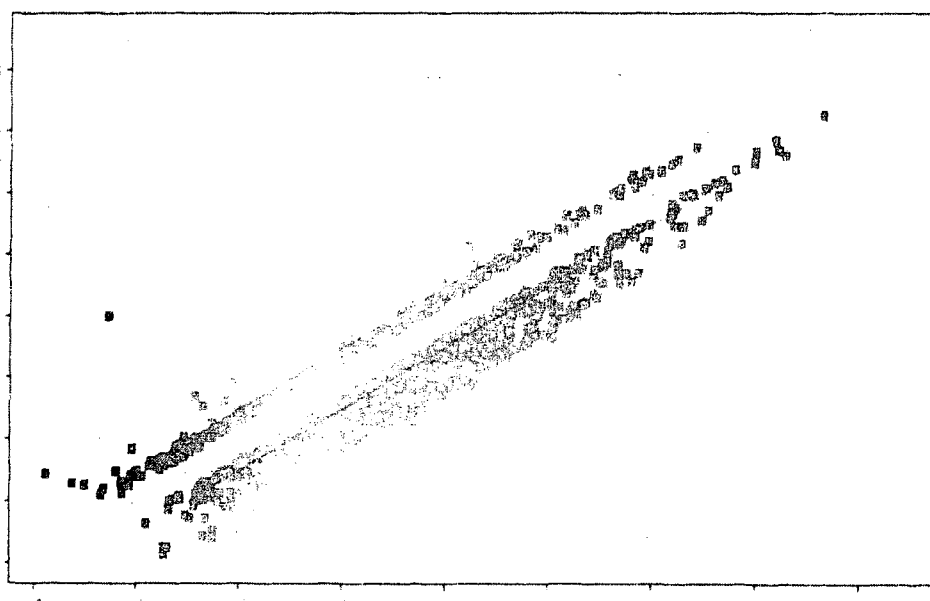


Figure 3A. Scatterplot of differential probes in cells transduced with batch B at MOI 150 vs. non-transduced cells.



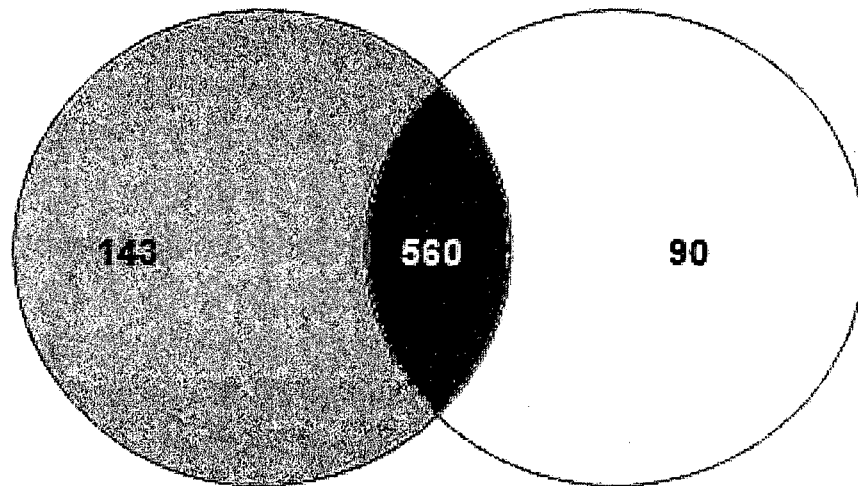
Selection of Fold Change $\geq 1,5$

Figure 3B. Scatterplot of differential probes in cells transduced with batch C at MOI 150 vs non-transduced cells.



Selection of Fold Change $\geq 1,5$

Figure 4. Venn diagram: downregulated probes in cells transduced with batch B vs non-transduced cells and downregulated probes in cells transduced with batch C vs. non-transduced cells MOI 150.



Downregulated probes in cells transduced
with batch B at MOI150
versus non-transduced cells.

Downregulated probes in cells transduced
with batch C at MOI150
versus non-transduced cells

Figure 5. Profile plot of cell cycle probes in cells transduced with batch B versus non-transduced cells at MOI 150 with $FC \leq -3$.

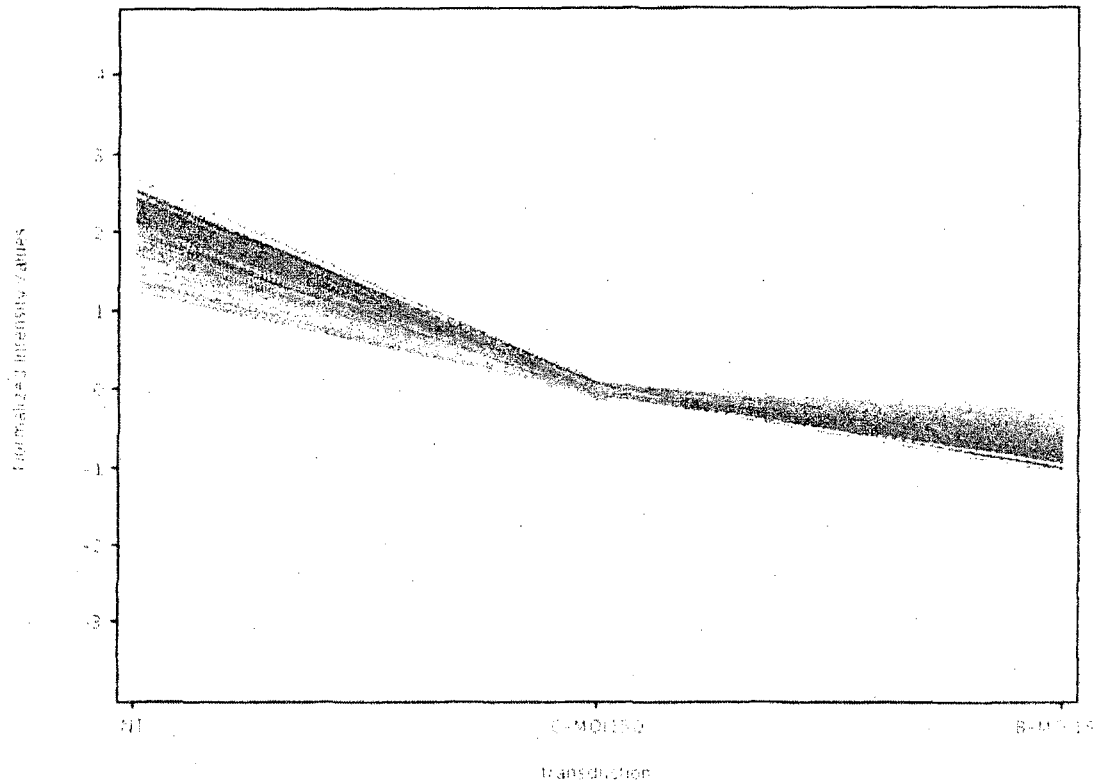


Figure 6A. Technical validation of microarrays results by RT-qPCR results.

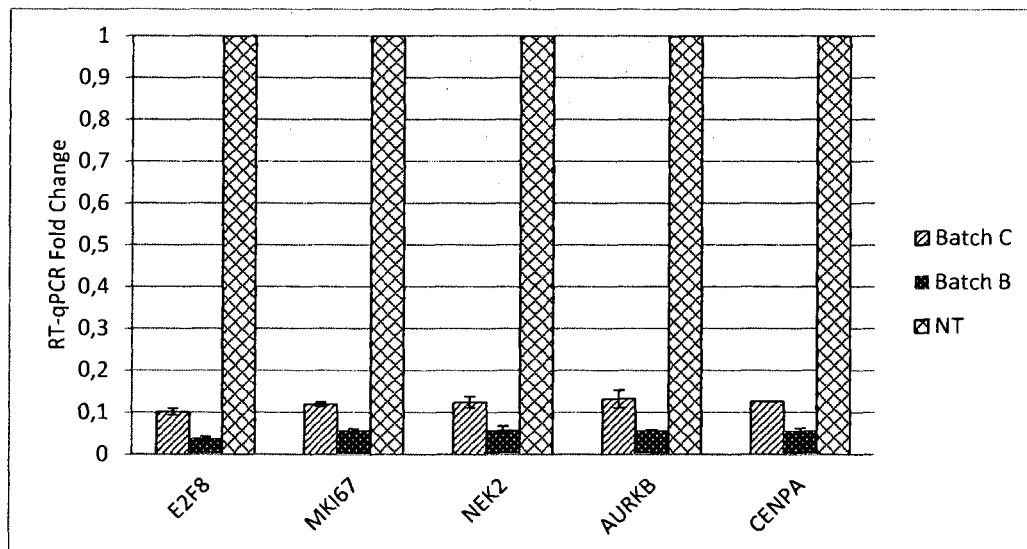
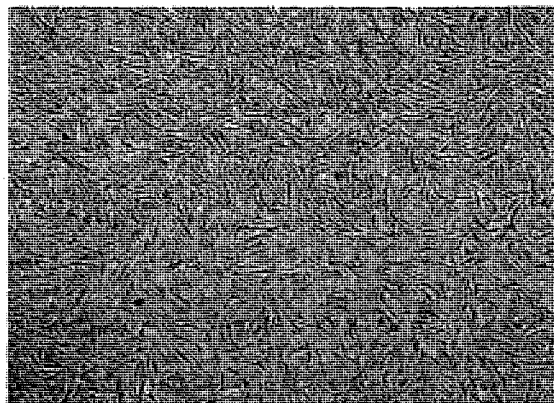


Figure 6B. Values of technical validation of microarrays results by RT-qPCR results.

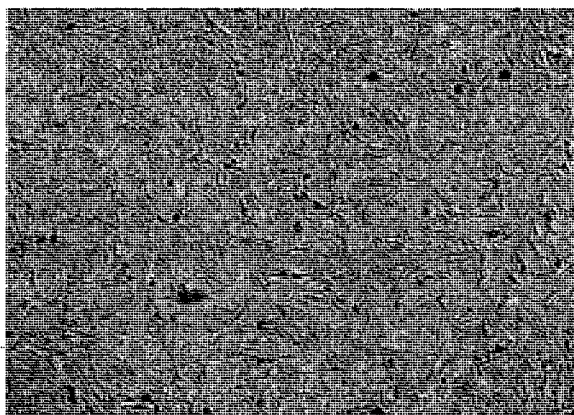
Gene (1)	Batch (2)	RT-qPCR FC versus NT (3)	Microarray FC versus NT (4)
AURKB	MOI 150 batch C	-7,5	-5,2
	MOI 150 batch B	-17,4	-10,1
NEK2	MOI 150 batch C	-8,0	-5,8
	MOI 150 batch B	-16,8	-10,4
E2F8	MOI 150 batch C	-9,8	-12,1
	MOI 150 batch B	-25,9	-19,6
MKI67	MOI 150 batch C	-8,3	-5,6
	MOI 150 batch B	-17,4	-8,6
CENPA	MOI 150 batch C	-7,9	-6,2
	MOI 150 batch B	-17,7	-11,5

Figure 7A. Cells transduced with batch B obtained with serum (batch B-S) compared to non-transduced cells.

NT



rLV-EF1 batch B-S MOI 40



rLV-EF1 batch B-S MOI 150

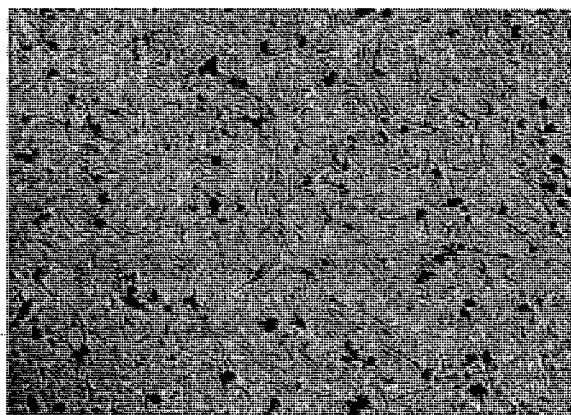


Figure 7B. Characteristics of batch B-S used for the transcriptomics study.

	Titer (TU/mL) (1)	PP/TU (2)
rLV-EF1 batch B-S	6,8E+07	494

Figure 8: Scatterplot of differential probes in cells transduced with batch B obtained with serum (B-S) versus non-transduced cells at MOI 150.

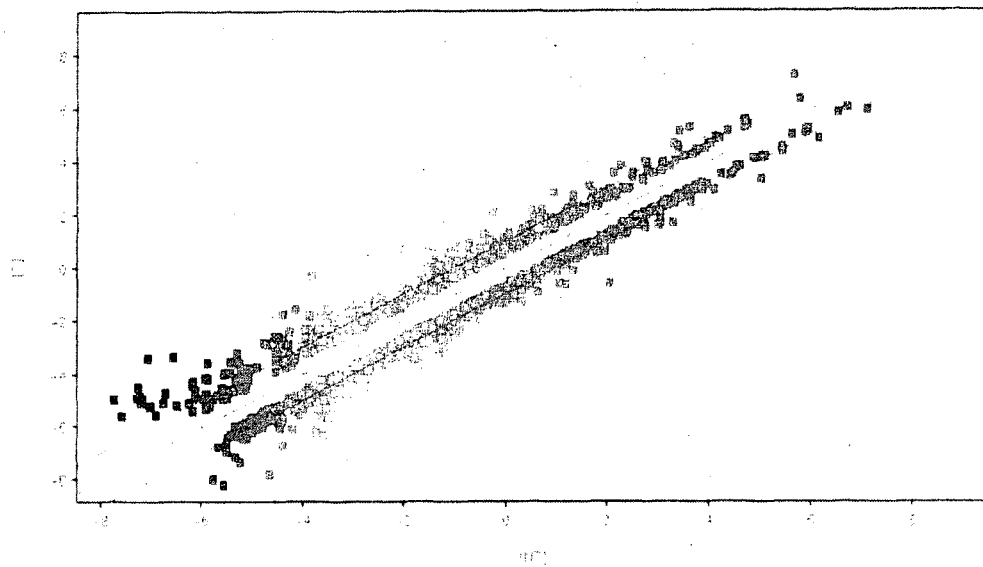


Figure 9. Scatterplot of differential probes in cells transduced with batch B obtained with serum (B-S) versus non-transduced cells at MOI 40.

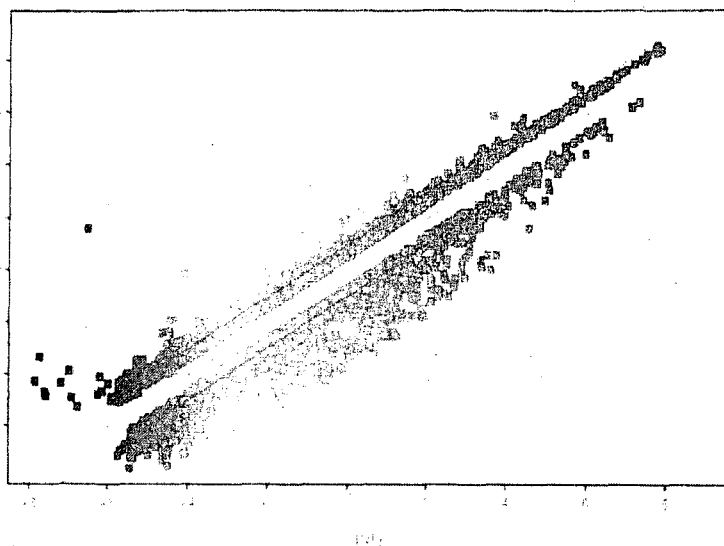


Figure 10. Venn diagram: Modulated probes in cells transduced with batch B obtained with serum (B-S) at MOI 150 vs non-transduced cells and modulated probes in cells transduced with batch B obtained with serum (B-S) at MOI 40 vs non-transduced cells.

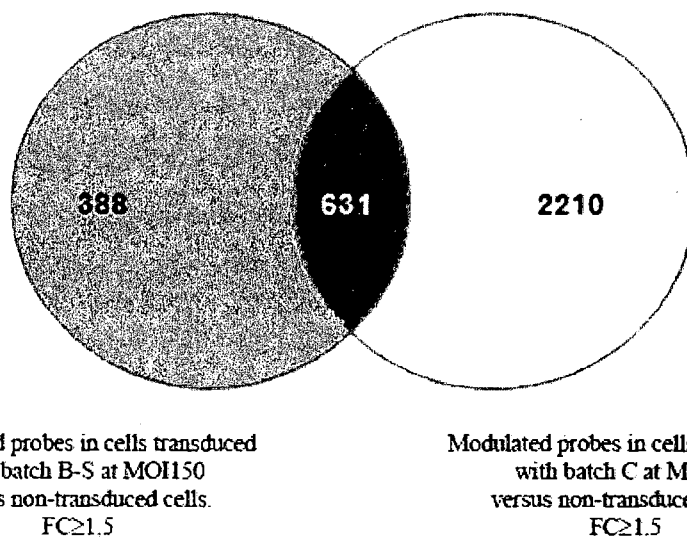


Figure 11. Profile plot representing probes impacted in cells transduced with batch B obtained with serum (B-S) at MOI40 and 150 and not differential in cells transduced with batches B and C, at MOI40 and 150.

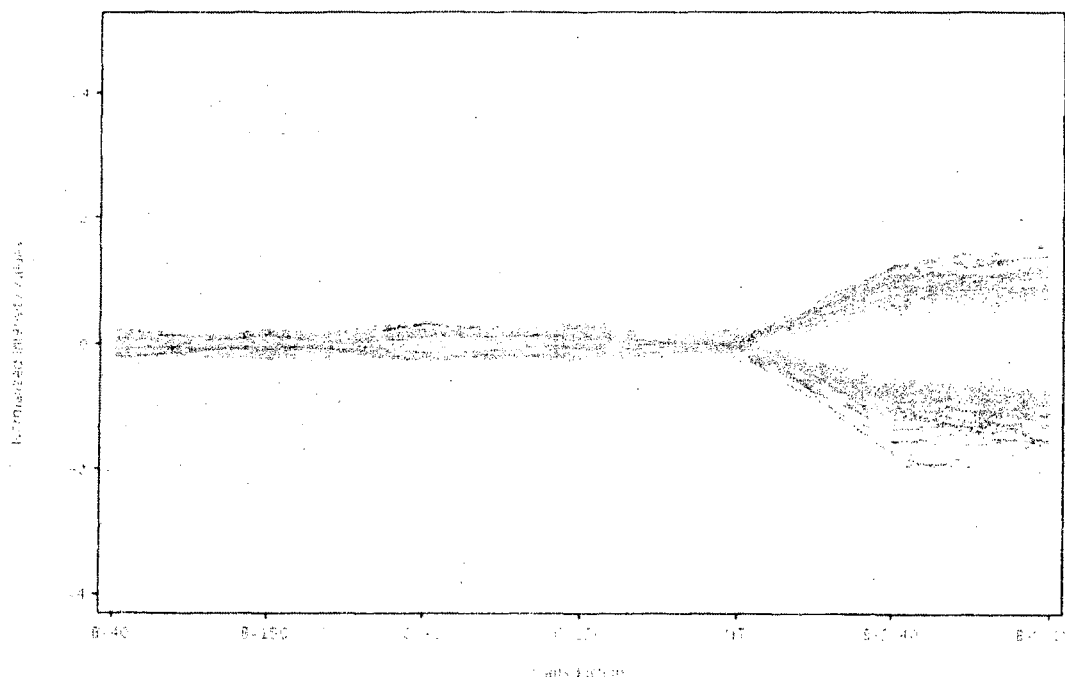
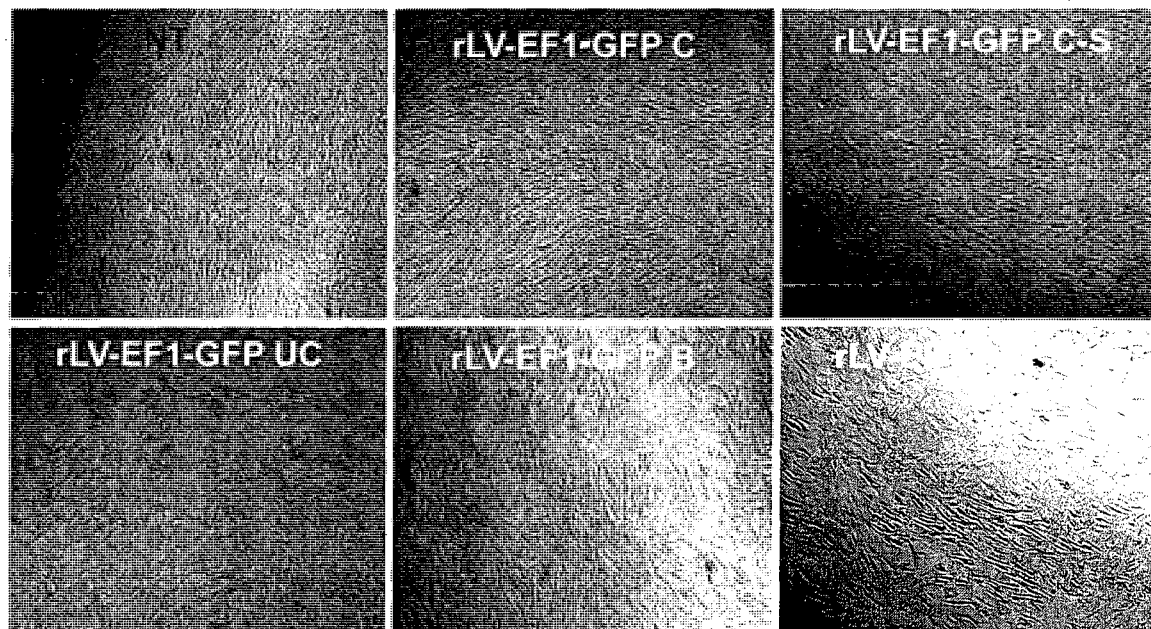


Fig 12 . Characterization of rLV-EF1-GFP vectors batch used (B, C, B-S and UC)

rLV-EF1-GFP Batch	Titer (TU/ml) (1)	PP/TU (2)
C	7,57E+07	195
B	9,26E+07	361
B-S	2,48E+08	293
UC	1,12E+08	348

Fig. 13 : Photos of Foreskin fibroblast cells transduced with rLV-EF1-GFP vectors. Cells were observed 48 hours after transduction.

MOI 40



MOI 150

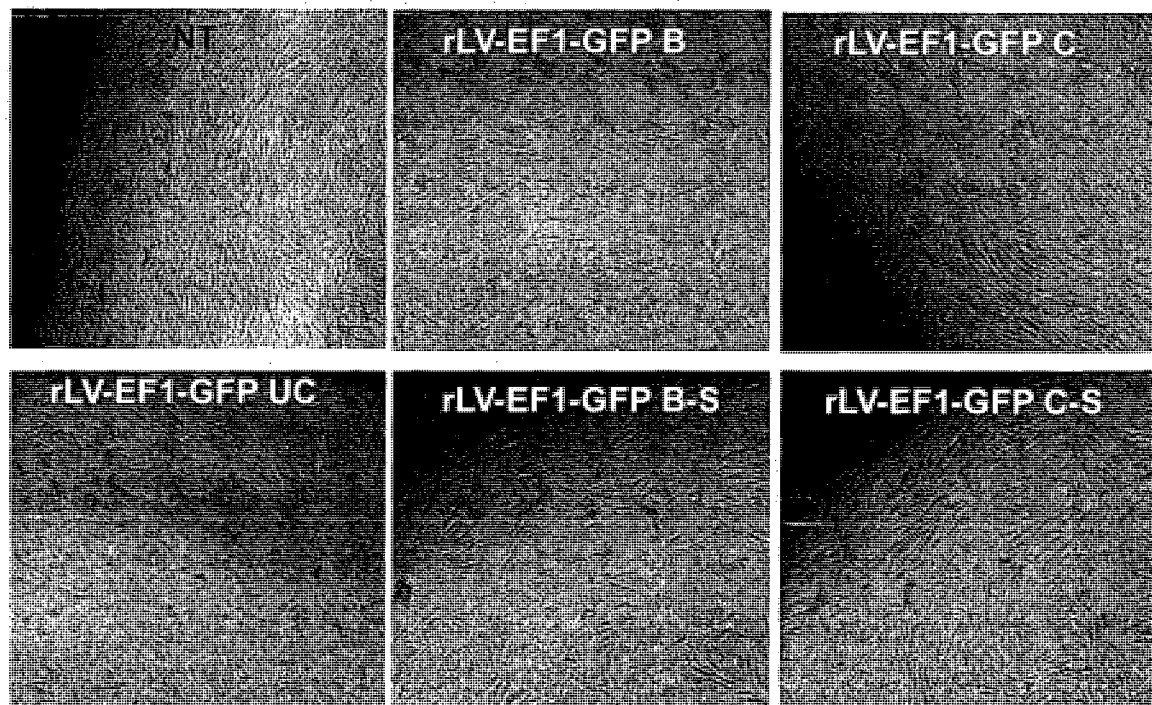


Fig 14: Probe selected in figure 5 (downregulated cell cycle genes with $FC \leq -3$ with B-MCS-MOI150 vs NT) represented in the following conditions: NT, B-GFP-MOI40, C-GFP-MOI40

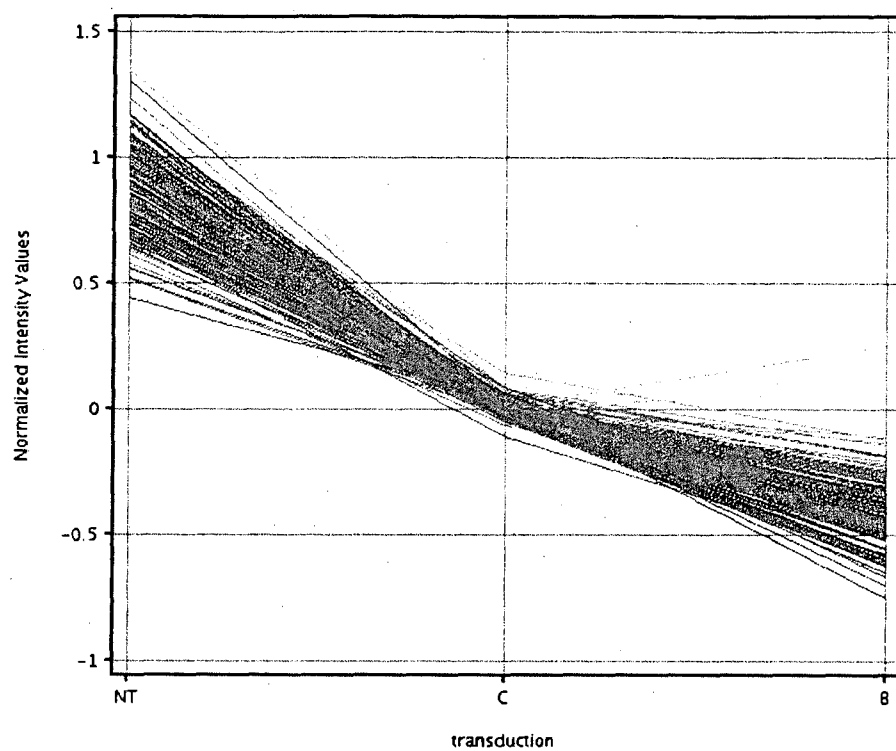


Fig. 15: Profile plot showing behavior of probes corresponding to table 3 after transduction with rLV-EF1-GFP batch B and C at MOI 40 versus Non Transduced (NT) cells.

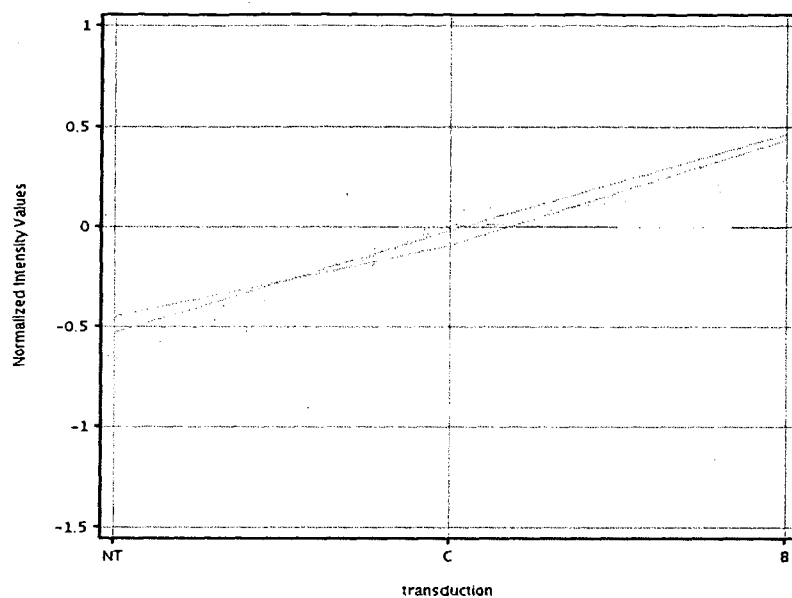


Fig. 16: Profile plot representing the probes corresponding to the 18 genes from table 4 and MKI67 after transduction with rLV-EF1-GFP vector at MOI 40.

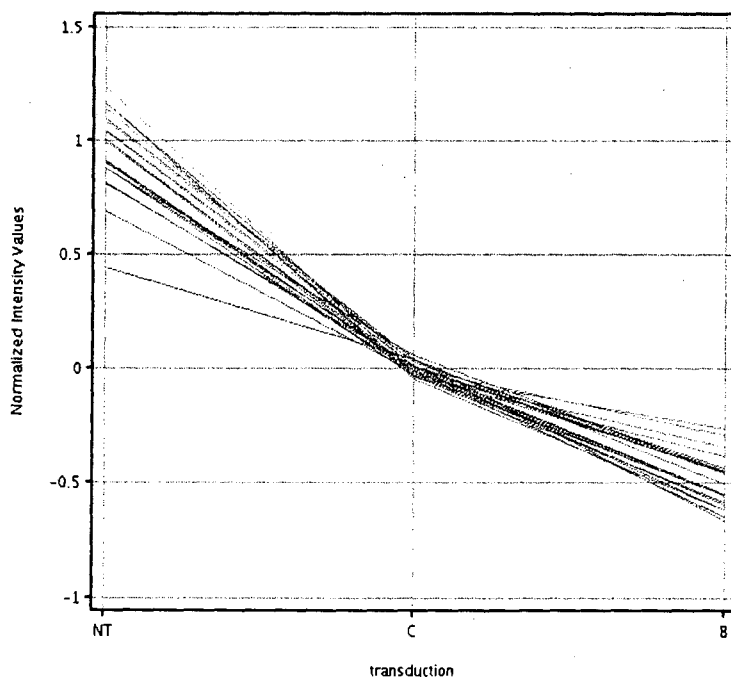


Fig 17. Profile plot representing the 18 cell cycle genes from table 4 and MKI67 after transduction with rLV-EF1-GFP vector (B, B-S and C) at MOI 40.

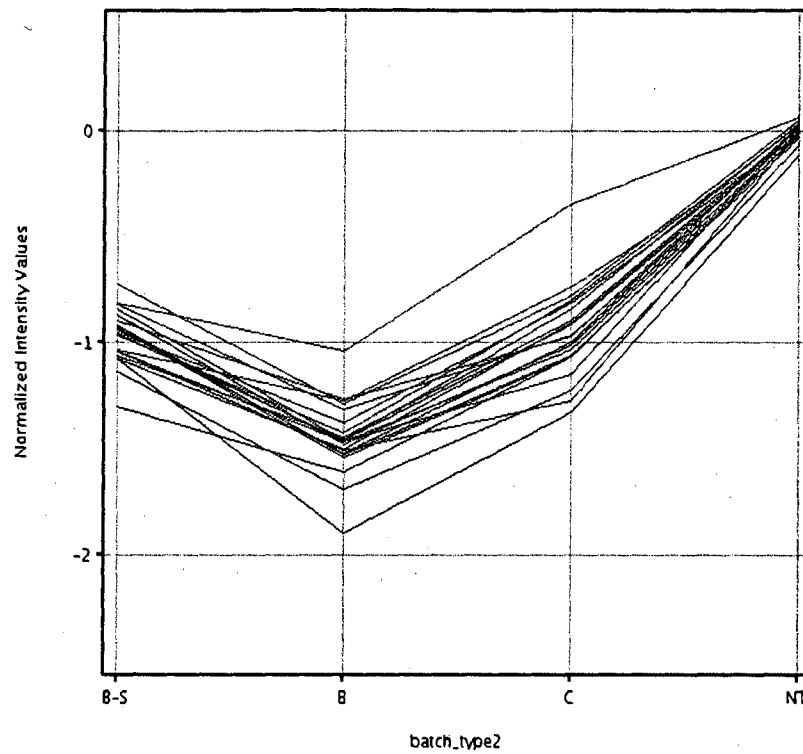
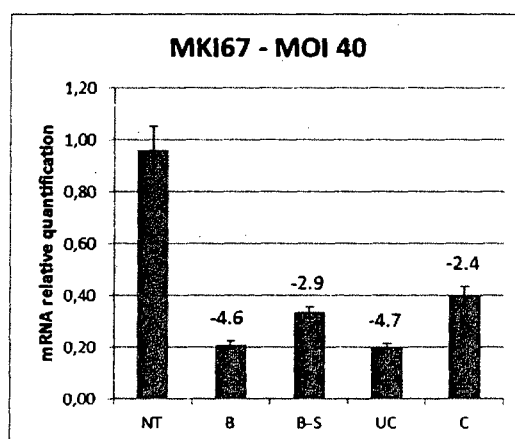


Fig. 18. Validation of some cell cycle genes as biomarkers by RT-qPCR.

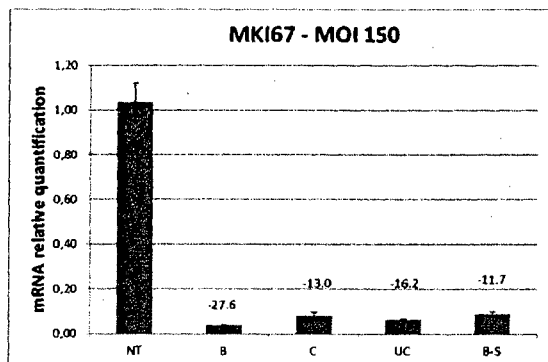
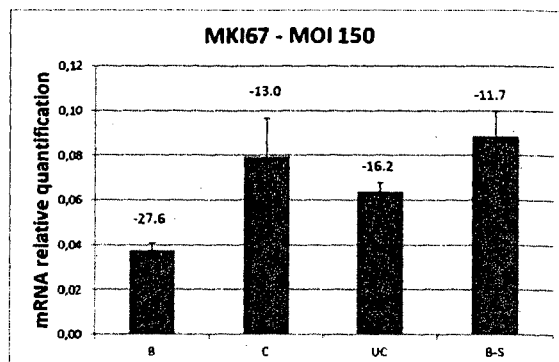
mRNA relative quantification values are represented as mean $\pm$ SD calculated from 3 samples per condition. Fold change values for each condition compared to NT are indicated above each diagram bar when differences are significant (*t*-test unpaired, *p*-value < 0.05).

• **18A. MKI67 – MOI 40**



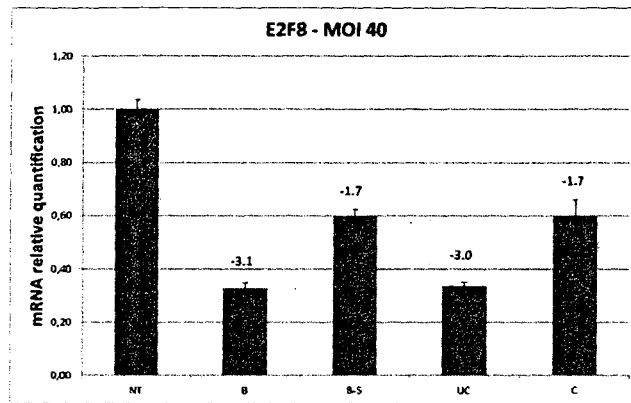
	p-value (t-test) cells vs. cells transduced with batch C on $\Delta\Delta Ct$
NT	0,00
B MOI 40	0,00
B-S MOI 40	0,05
UC MOI 40	0,00

• 18B. MKI67 – MOI 150



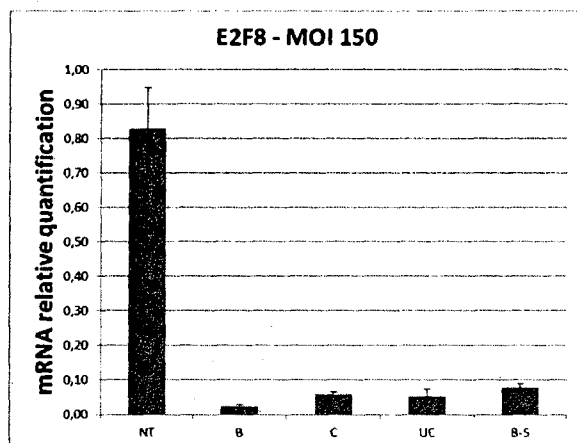
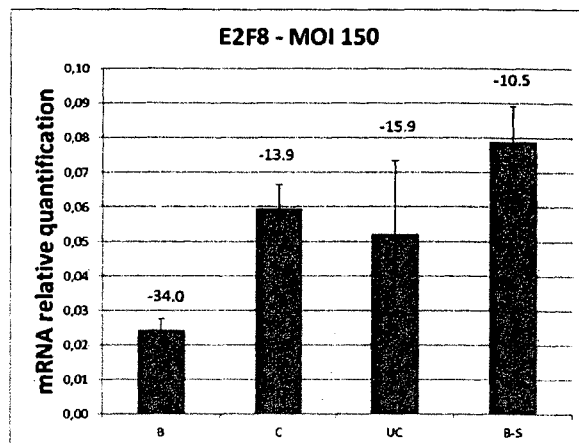
	p-value (t-test) cells vs. cells transduced with batch C on $\Delta\Delta Ct$	p-value (t-test) cells transduced vs. non-transduced cells on $\Delta\Delta Ct$
NT	0	
B MOI 150	0	1,05E-06
C MOI 150		3,48E-05
UC MOI 150	0,18	1,12E-06
B-S MOI 150	0,43	7,92E-06

- 18C. E2F8 – MOI 40



	p-value (t-test) cells vs. cells transduced with batch C on $\Delta\Delta C_t$	p-value (t-test) cells transduced vs. non- transduced cells on $\Delta\Delta C_t$
NT	0,00	
B MOI 40	0,00	0,00
B-S MOI 40	0,95	0,00
UC MOI 40	0,00	0,00
C MOI 40		0,00

- 18D. E2F8 – MOI 150

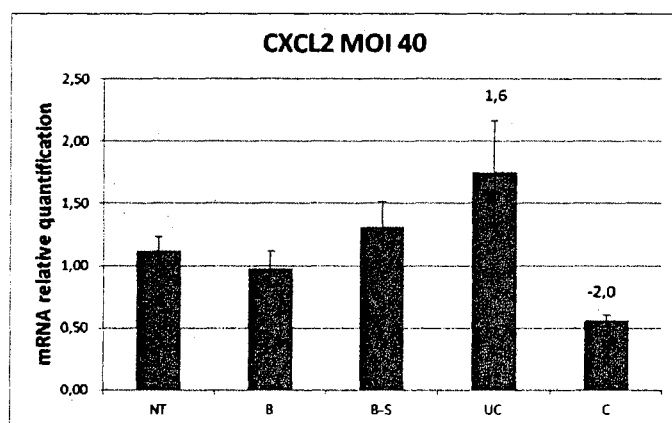


	p-value (t-test) cells vs. cells transduced with batch C on $\Delta\Delta Ct$
NT	0,00
B MOI 150	0,00
C MOI 150	
UC MOI 150	0,30
B-S MOI 150	0,99

Fig. 19. Validation of CXCL2 and EREG genes as biomarkers by RT-qPCR.

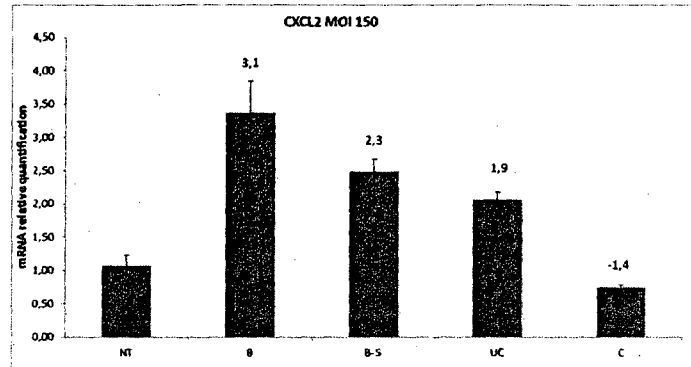
mRNA relative quantification values are represented as mean $\pm$ SD calculated from 3 samples per condition. Fold change values for each condition compared to NT are indicated above each diagram bar when differences are significant (t-test unpaired, p -value < 0.05).

- 19A. CXCL2 MOI 40**



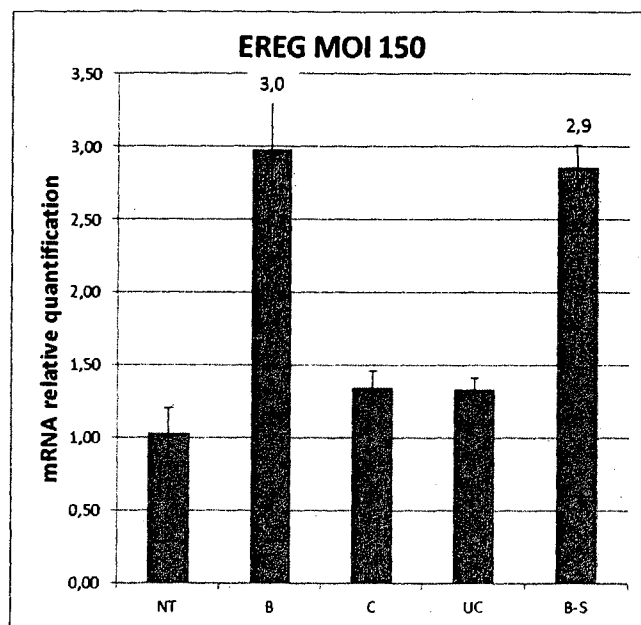
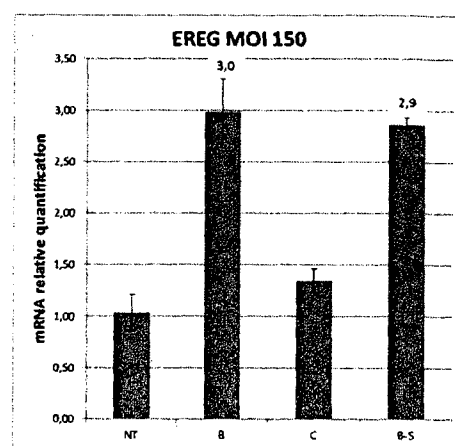
	p-value (t-test) cells vs. non- transduced cells on $\Delta\Delta Ct$	p-value (t-test) cells transduced vs. cells transduced with batch C on $\Delta\Delta Ct$
NT		0,00
B MOI 40	0,24	0,01
B-S MOI 40	0,22	0,00
UC MOI 40	0,04	0,00
C MOI 40	0,00	

- 19B. CXCL2 MOI 150



	p-value (t-test) cells vs. cells transduced with batch C on $\Delta\Delta Ct$	p-value (t-test) cells transduced vs. non- transduced cells on $\Delta\Delta Ct$
NT	0,01	
B MOI 150	0,00	0,00
B-S MOI 150	0,00	0,00
UC MOI 150	0,00	0,00
C MOI 150		

• 19C. EREG MOI 150



	p-value (t-test) cells vs. cells transduced with batch C on $\Delta\Delta Ct$	p-value (t-test) cells transduced vs. non- transduced cells on $\Delta\Delta Ct$
NT	0,07	
B MOI 150	0,00	0,00
C MOI 150		0,07
UC MOI 150	0,93	0,06
B-S MOI 150	0,00	0,00