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Supplemental Information

Generation of High-Titer Self-Inactivated

γ -Retroviral Vector Producer Cells

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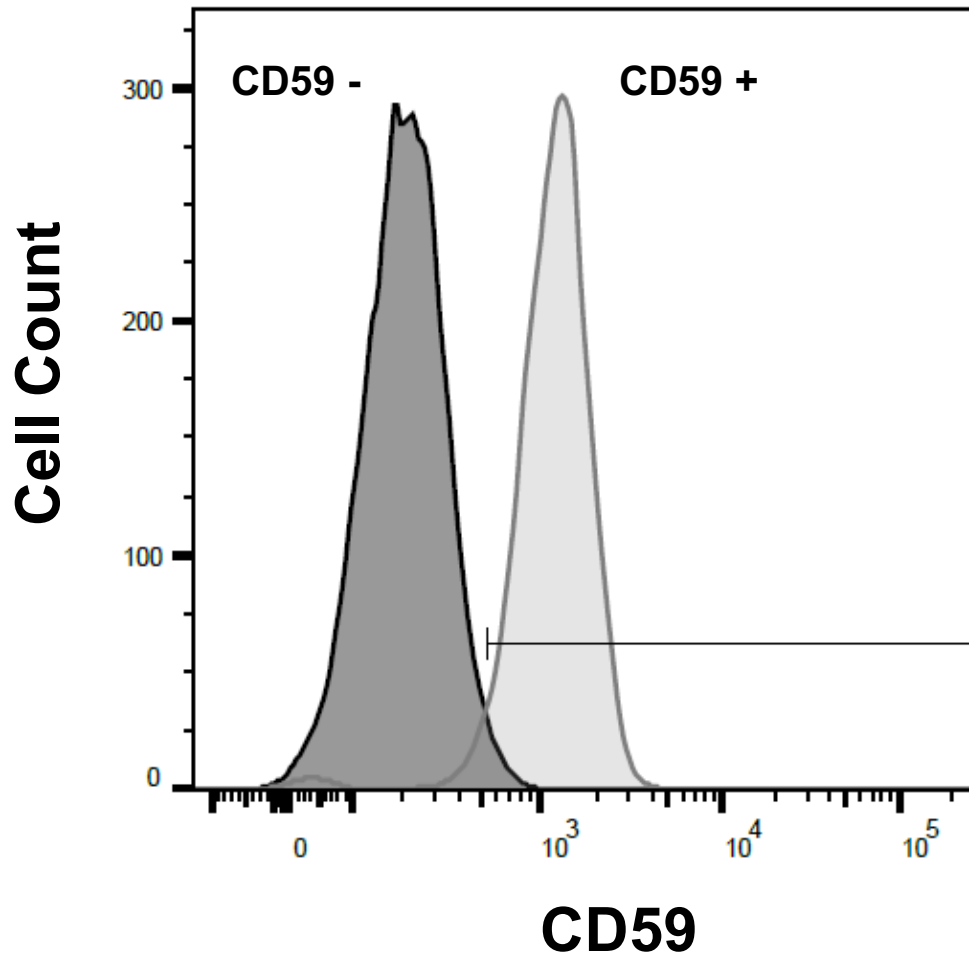


Figure S1. CD59 expression loss in K562 cells after *PIGA* gene inactivation. FACS analysis of K562 cells (light grey) and K562(*PIG*⁻) cells (dark grey) stained with an anti-CD59 antibody FITC-labeled.

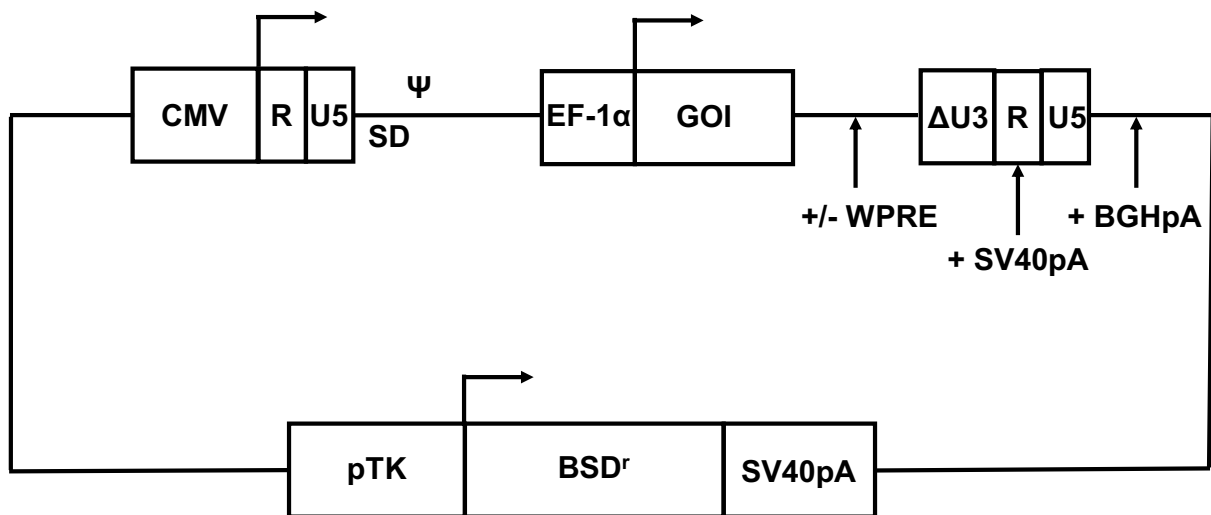


Figure S2. Design of the final SIN vector containing the blasticidin resistant cassette.

SIN vectors contain a gene of interest (*GOI*; *GFP*, *COL7A1* and *PIGA*) under the control of the EF-1 α promoter, the CMV promoter/enhancer sequence and the two poly(A) sequences (SV40pa and BGHpA). WPRE was present only in the GFP and PIGA vectors. SD and Ψ are splice donor site and packaging signal sequence, respectively. In the resistant cassette, the *BSD^r* gene was under the control of the herpes simplex virus thymidine kinase promoter (HSV-TK). A SV40pa was located downstream the *BSD^r* gene.

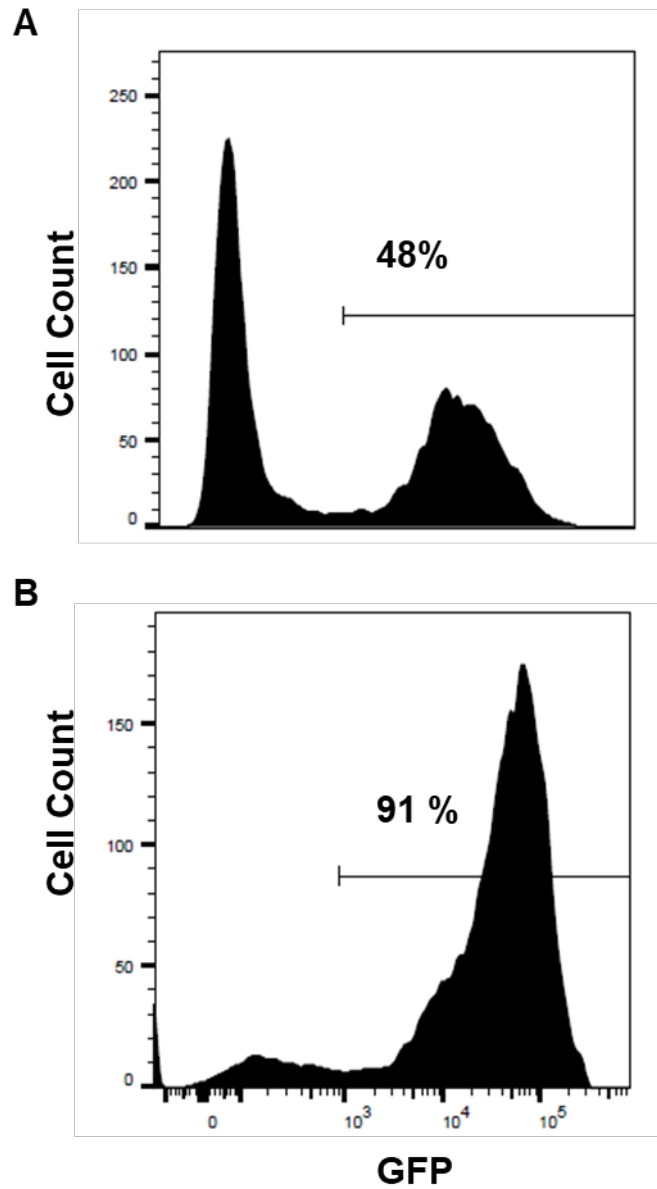


Figure S3. GFP fluorescence of 293Vec-RD114 transfected with the SINVec.GFP plasmid. (A) Cells selected in hygromycin and (B) in blasticidin were analyzed by FACS for GFP fluorescence. Displayed is the result of one representative experiment done three times.

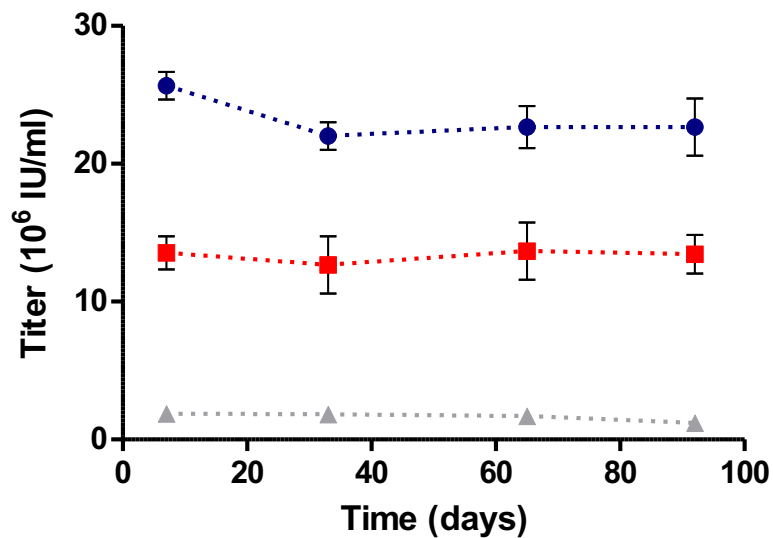


Figure S4. Long-term stability of SIN vector production from stable vector producer clones. Titers were measured by FACS analysis as described in the Materials and Methods section after infection of HT-1080 cells with the SINVec.GFP vector (blue) and the SINVec.COL7A1 (-W) (grey). The SINVec.PIGA vector (red) was titered on K562(PIG-). The SIN GFP, COL7A1 and PIGA vector producer clones had 8, 7 and 4 plasmid copies/cell integrated, respectively. Values presented are the mean of one experiment done in triplicate.