Supplemental Results



Figure S3. Transfection of U1snRNA mutants does not reduce cell viability. Cells from a 6-well plate were transfected with 1.5µg of either wildtype U1 snRNA or the four mutants as indicated, harvested 36 hours post transfection, resuspended in 1 x phosphate buffered saline to which 5µl of 7-AAD Viability Staining Solutionwas addded. Fluorescence in the red channel was measured by FACS. Values represent 100 less % of cells above Mock (un-transfected cells) fluorescence.



Figure S5. The heterologous hybrid intron is constitutively spliced. cDNA from the transfection in Figure 4B was amplified using the primers indicated with amplification of a 344bp product being indicative of constitutive splicing.



Figure S4. Secondary RNA structure of U1 snRNA stem loop I. The stem loop sequence of WT and mut was submitted to M-fold RNA software to obtain the structure shown.



Figure S6. Tat enhances expression of the SD4SA7 mutant more than WT.R38. Cells were transfected with 2µg pHIV-gp140uncGFP containing either WT.R38 or SD4SA7 mutations and either 100ng of pCMV-Tat(2stop) (-Tat) or pCMV-Tat (+Tat) with 100ng pCMV-Rev. Fold activation is expressed here as +Tat/-Tat. Transfections were performed on three separate days and the average shown, error bars represent the standard deviation of the mean.