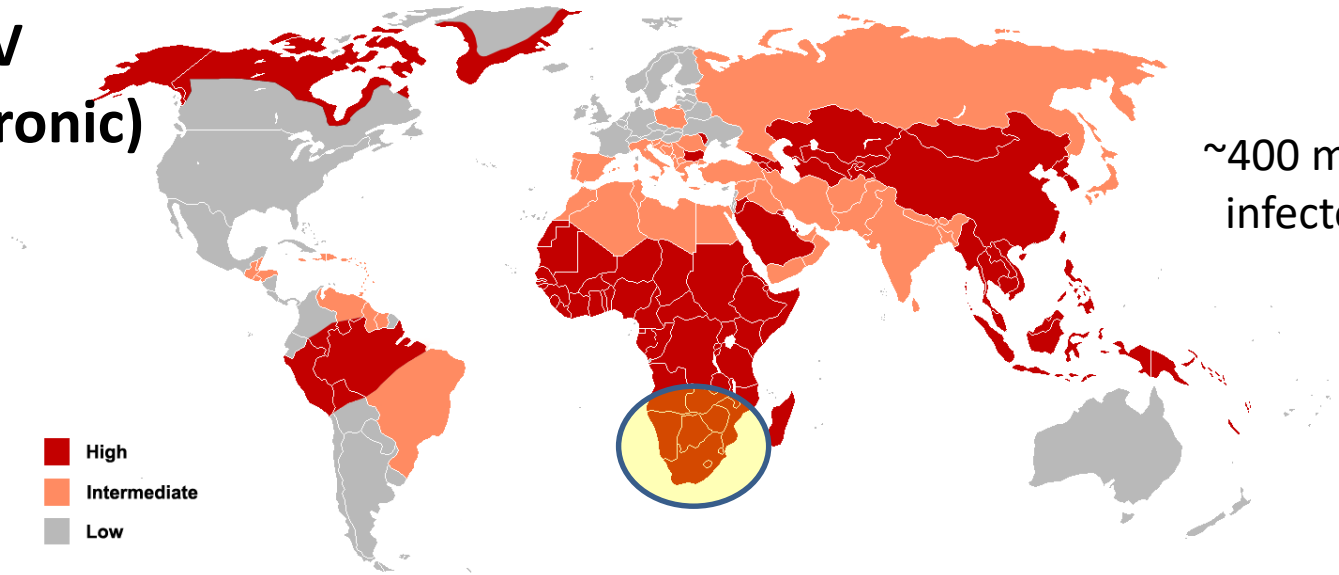
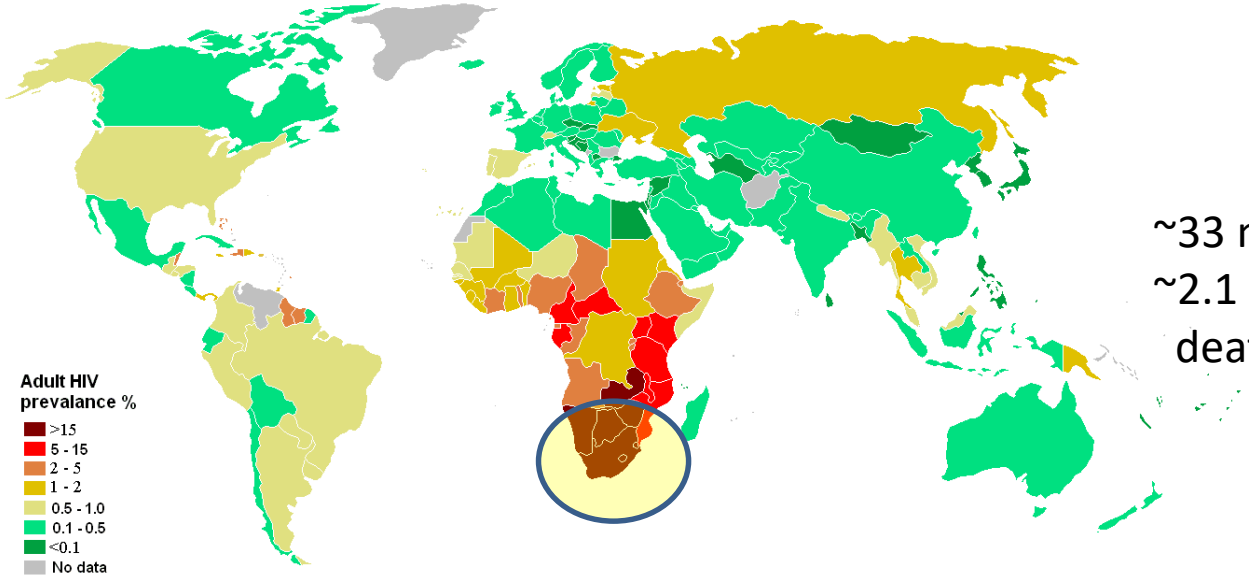
HBV (chronic)



~400 million chronically infected

HIV

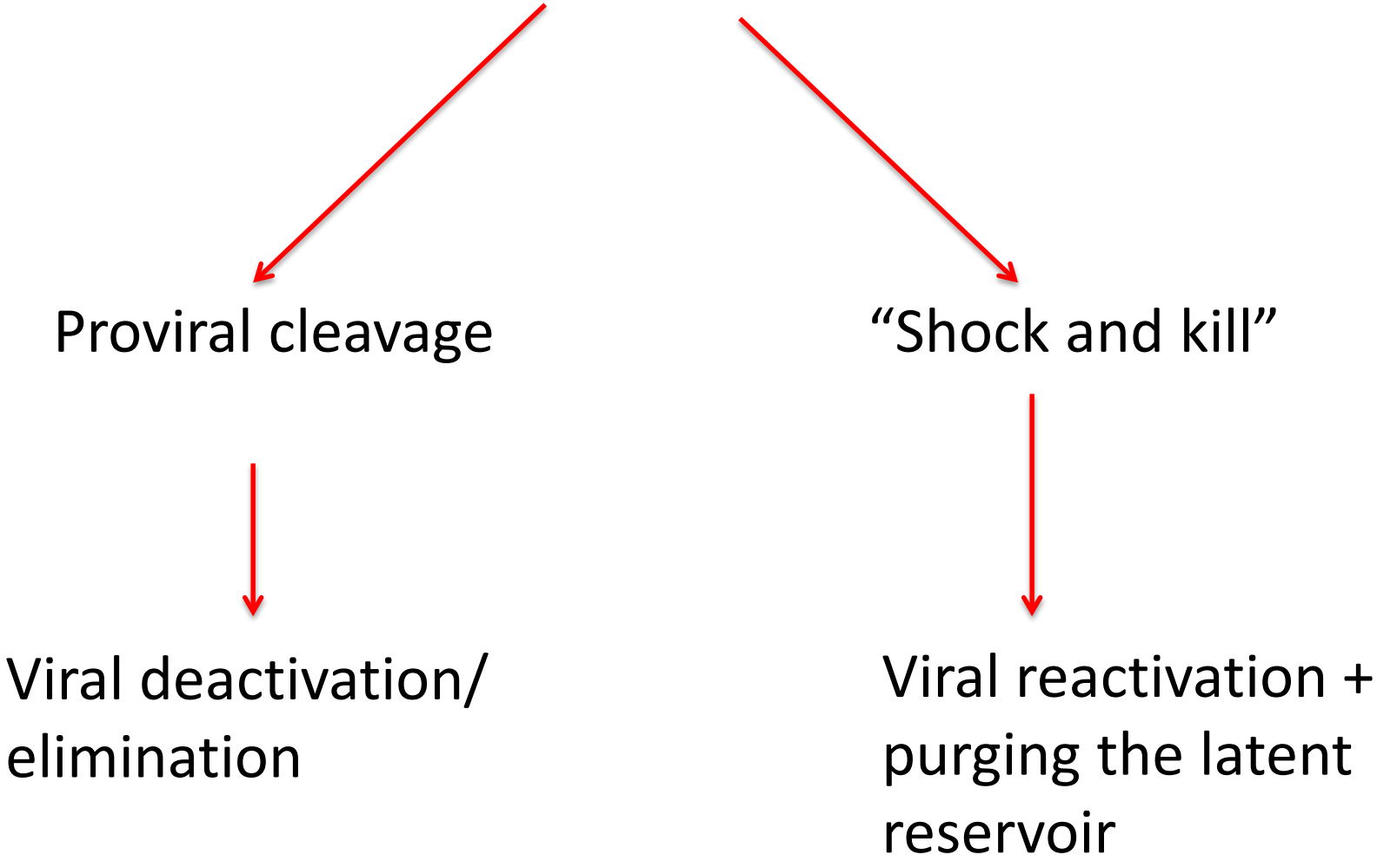


~33 million infected
~2.1 million deaths/year (AIDS)

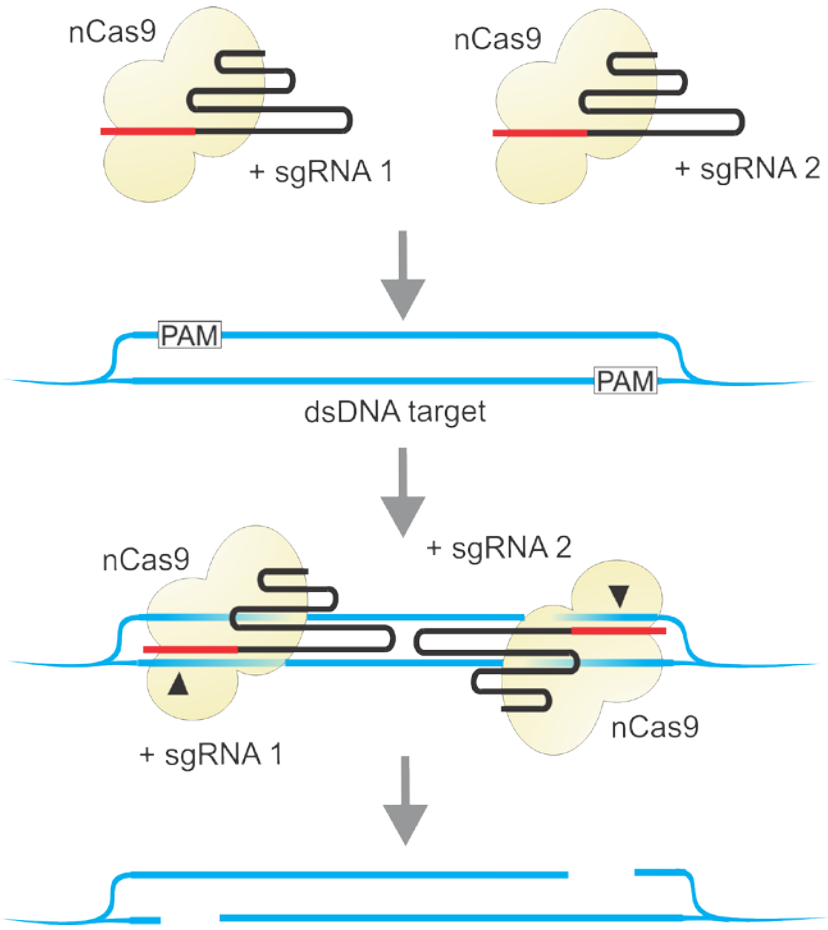
How to eradicate rogue viral DNA?

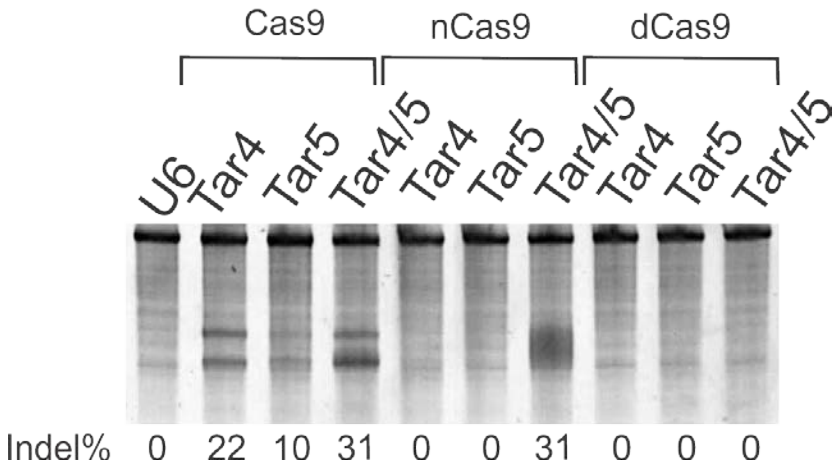
- HBV and HIV are both chronic viral infections. Lifelong treatment is necessary. None of the >25 approved drugs for HIV or HBV can clear the infection.
- HBV replicates via a closed circular DNA (cccDNA) intermediate which persists in the liver.
- HIV forms a dsDNA which integrates into the human genome (provirus) of infected CD4+ cells
- CCR5 editing is a promising approach for HIV. But feasibility/ applicability of *ex vivo* stem cell manipulation is limited

CRISPR “functional cure” paradigms when targeting HIV directly



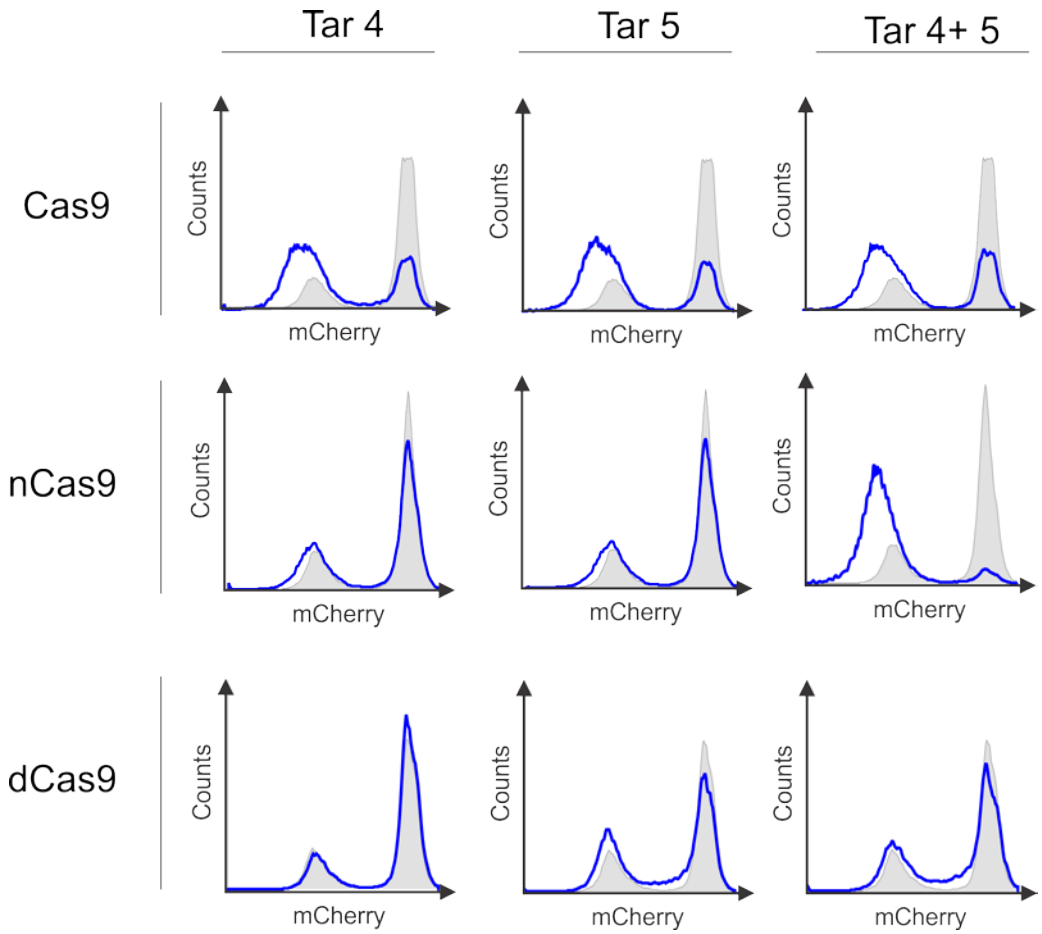
Gene editing using paired sgRNAs and Cas9 “nickase” targeted to HIV



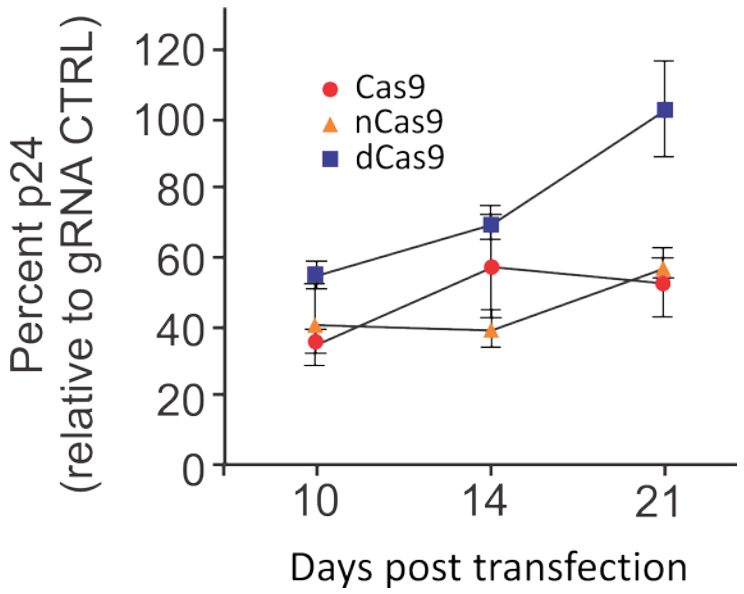


Suppression of HIV proviral expression

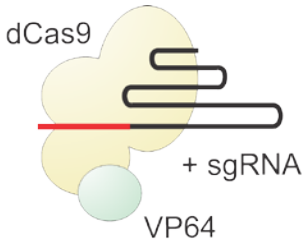
LChIT CEM T cells

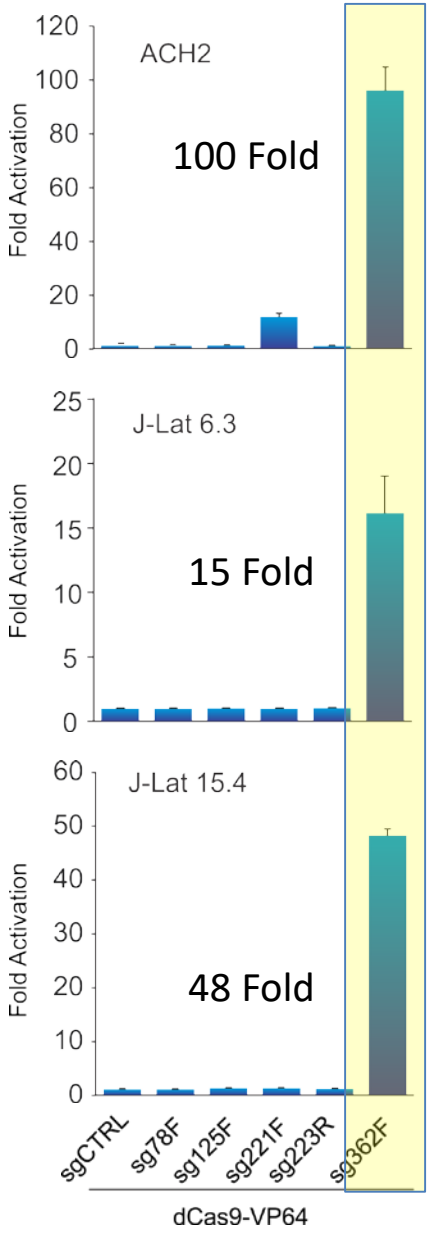


J1.1 cells

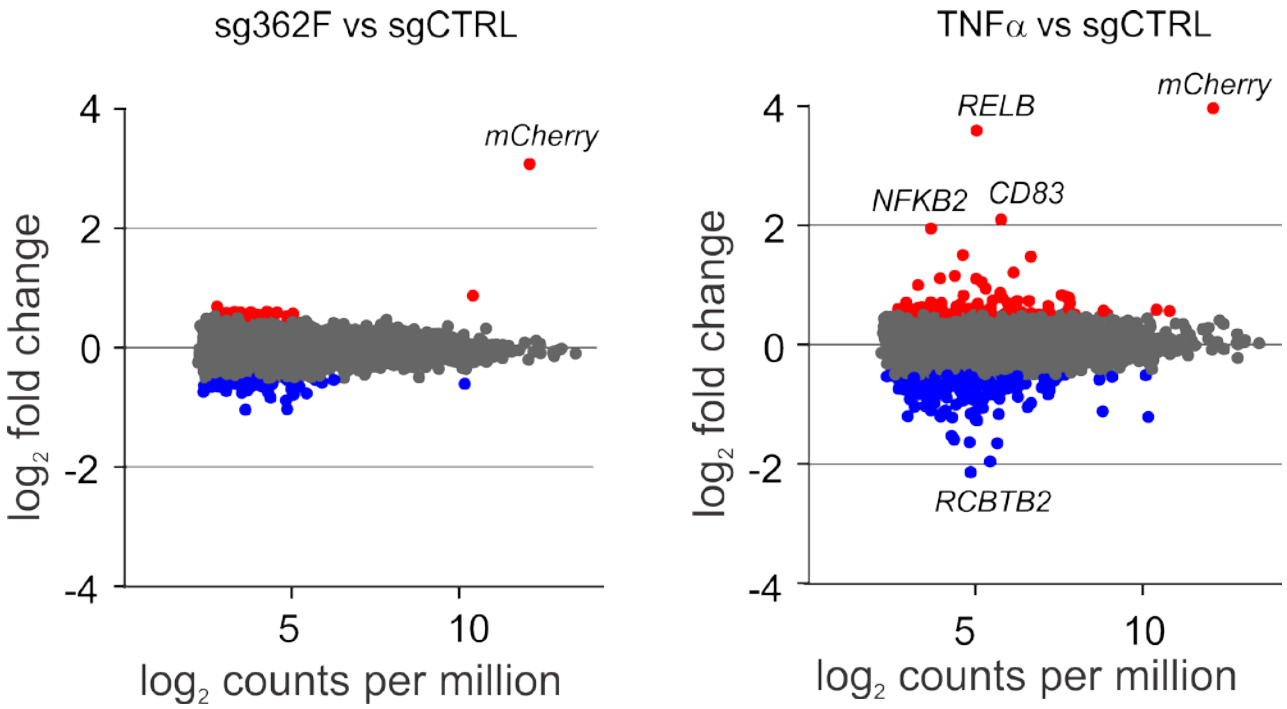


CRIPSR activation (CRISPRa) “shock and kill”





CRISPRa “on-target” specificity



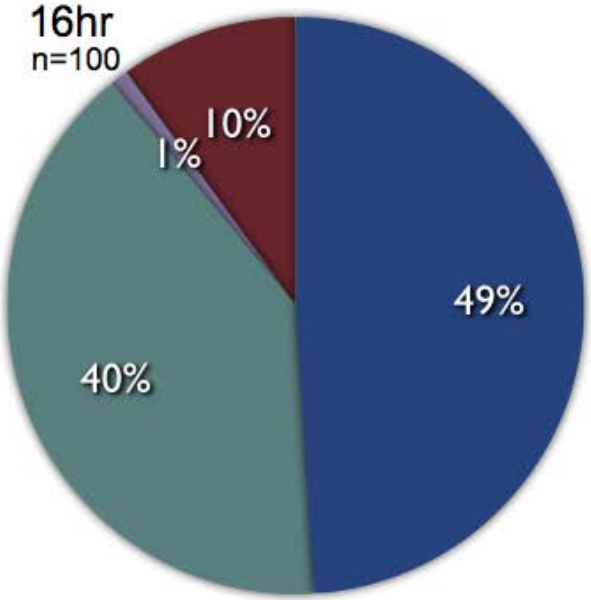
Summary

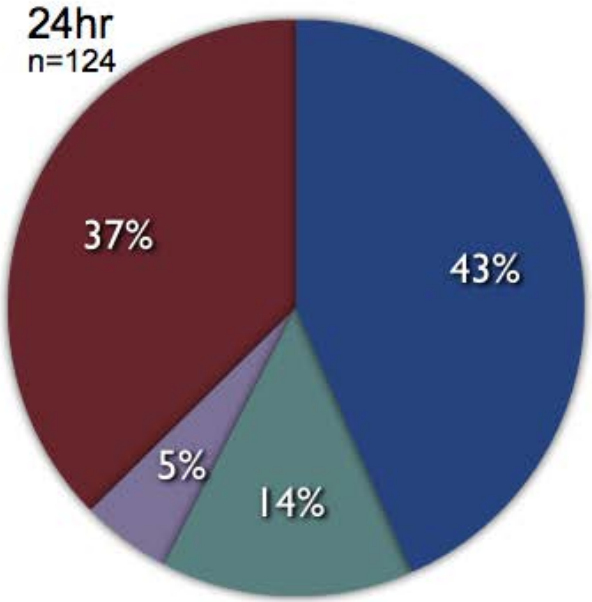
- CRISPR is a powerful tool with the potential to eradicate integrated and/or persistent rogue viral diseases such as HBV and HIV

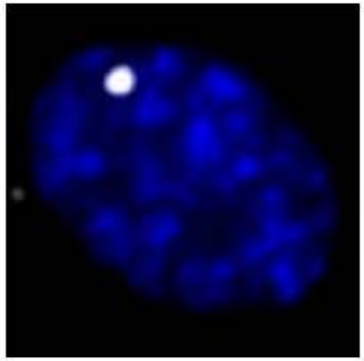
- Some future hurdles exist:
  - Adequate strategy for *in vivo* delivery
  - Need to reach 100% of infected cells/reservoirs
  - Emergence of resistant virus
  - Removing/reducing Cas9 exposure (nuclease activity)

CRSIPR/Cas9

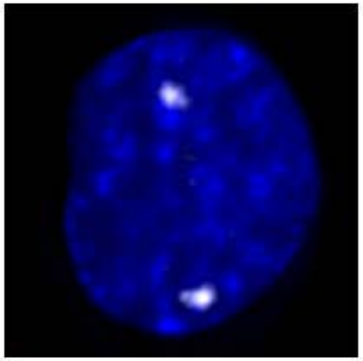
TALENs



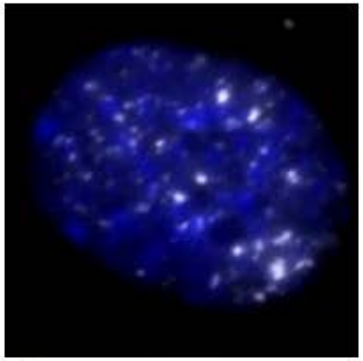




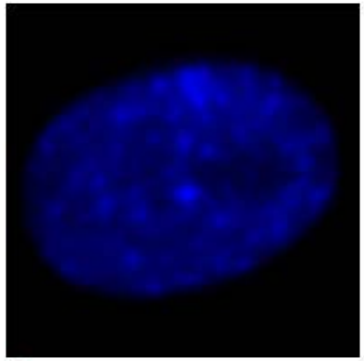
Single allelic break



Dual allelic break



Multiple breaks



No breaks

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