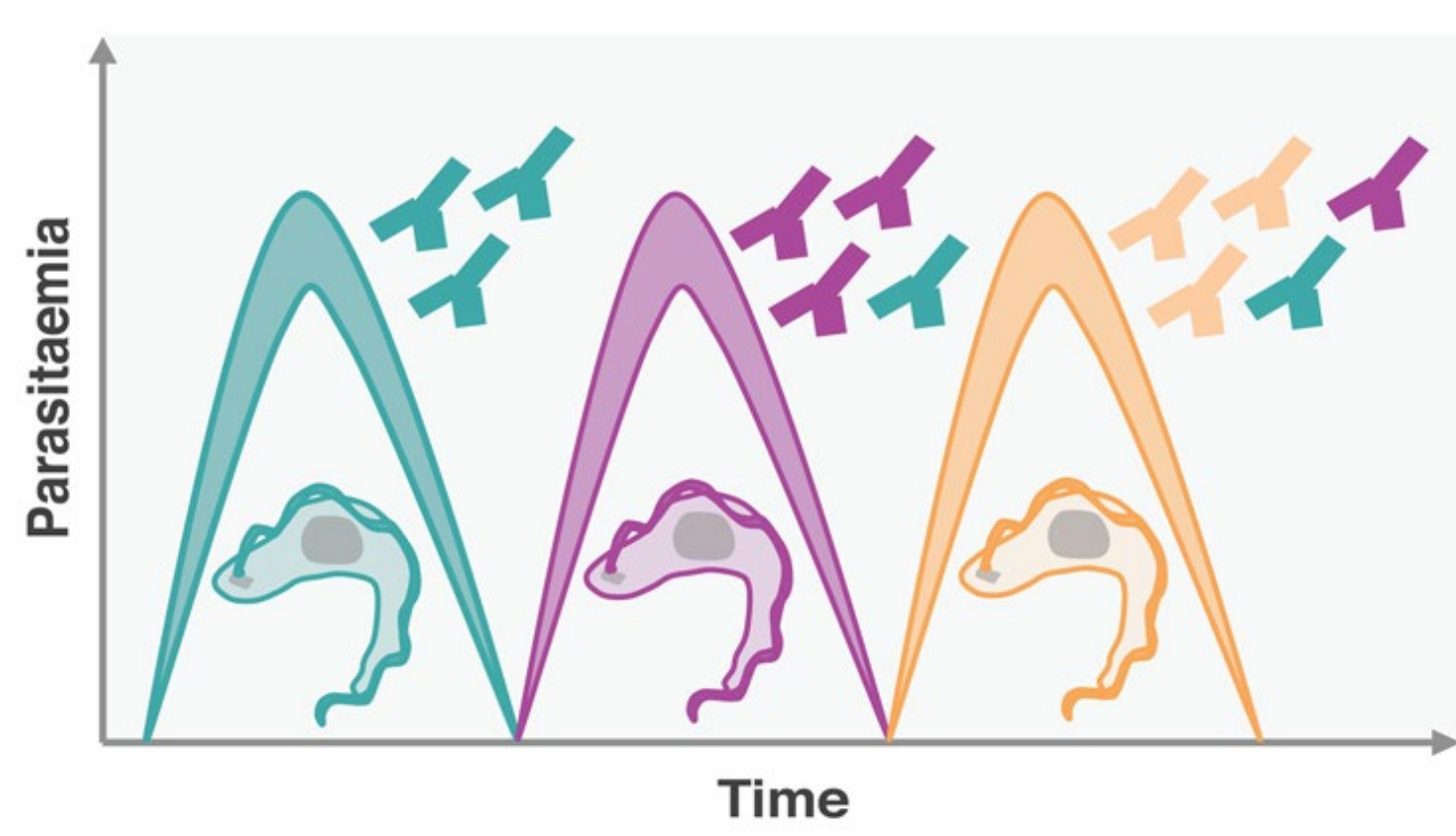


Introduction

Several persistent pathogens employ antigenic variation to continually evade mammalian host adaptive immune responses. African trypanosomes use variant surface glycoproteins (VSGs) for this purpose, transcribing one telomeric VSG expression site (ES) at a time, with strict silencing of other telomeric ESs.

Antigenic variation during infection



BES → Other ES → MC

VSG genes repertoire

Bloodstream expression-site (BES) n=15



Minichromosomes (MC) (app 100 copies)

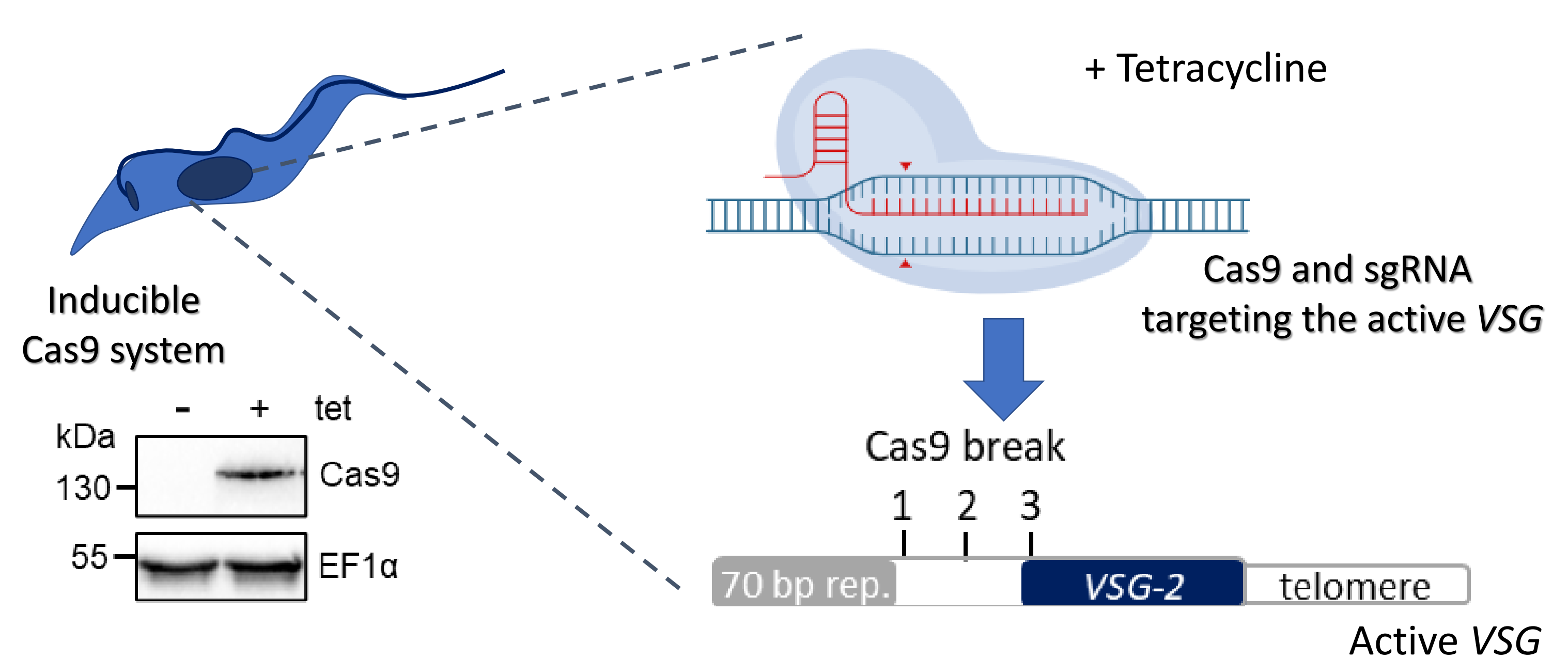


MES (n=5)



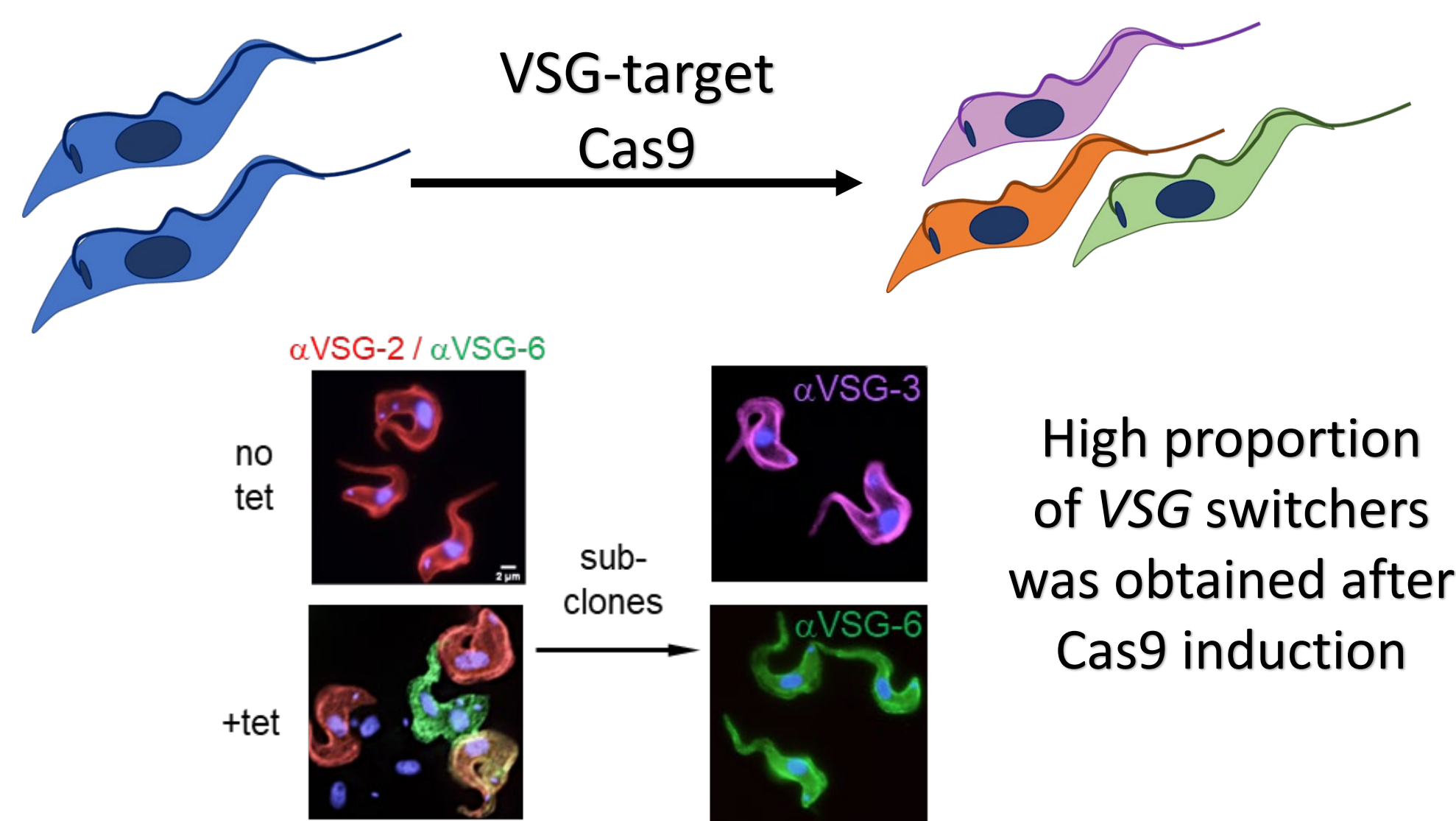
Faria, J. 2021. *Parasitology*. doi:10.1017/S0031182020002437

We have established a highly efficient and conditional CRISPR/Cas9-based system for *Trypanosoma brucei*. Our strategy is to use gRNAs targeting the active VSG for allowing high-frequency VSG gene recombination and switching *in vitro*. Thereby assessing factors that may contribute to this VSG hierarchy without immune system pressure.

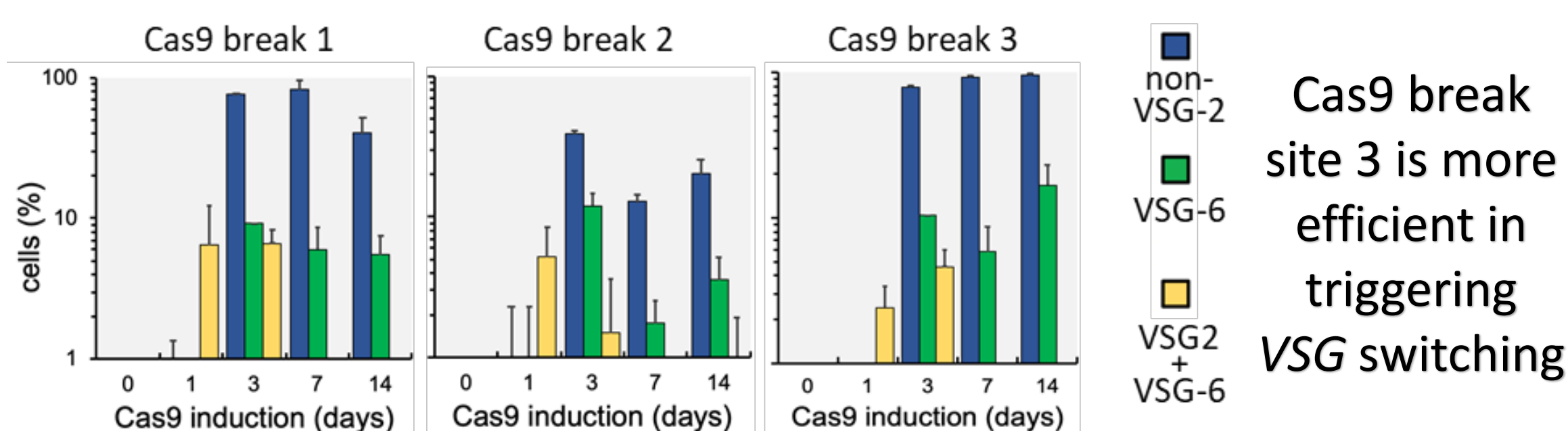


Rico, E., et al., 2018. *Sci Rep*. doi: 10.1038/s41598-018-26303-w

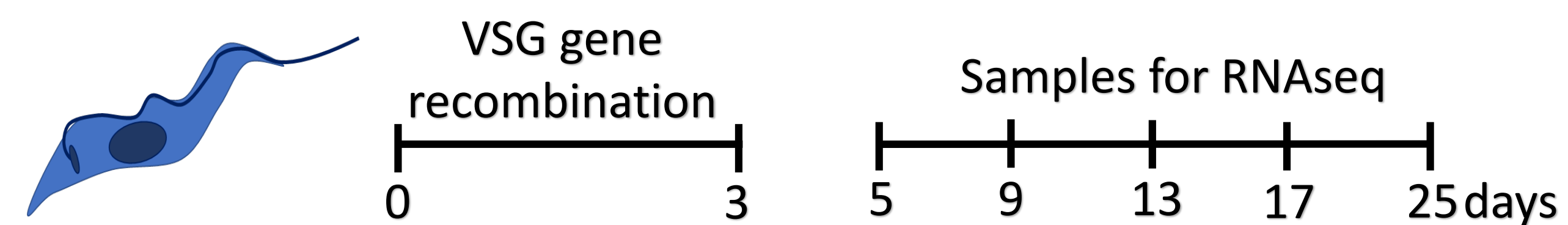
CRISPR-Cas9 efficiently triggers high frequency VSG gene recombination



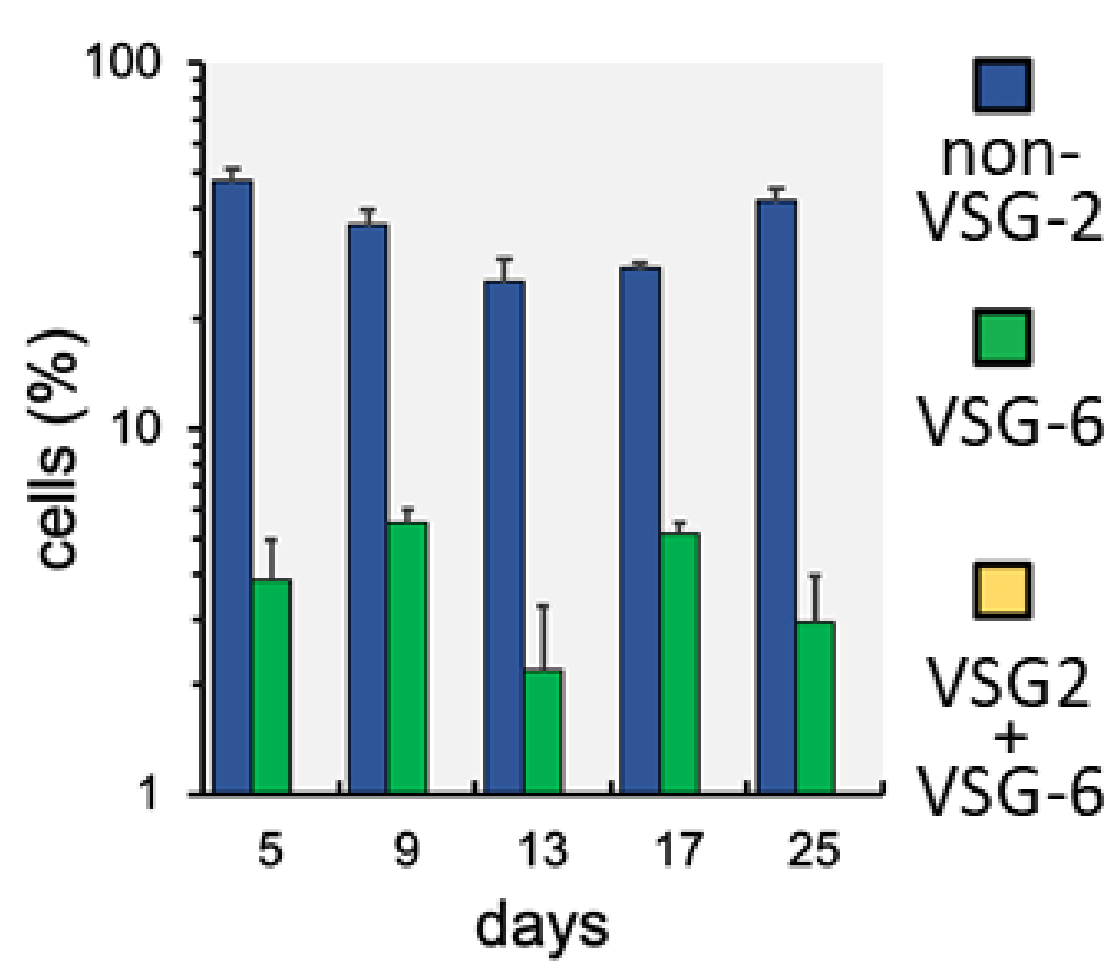
High proportion of VSG switchers was obtained after Cas9 induction



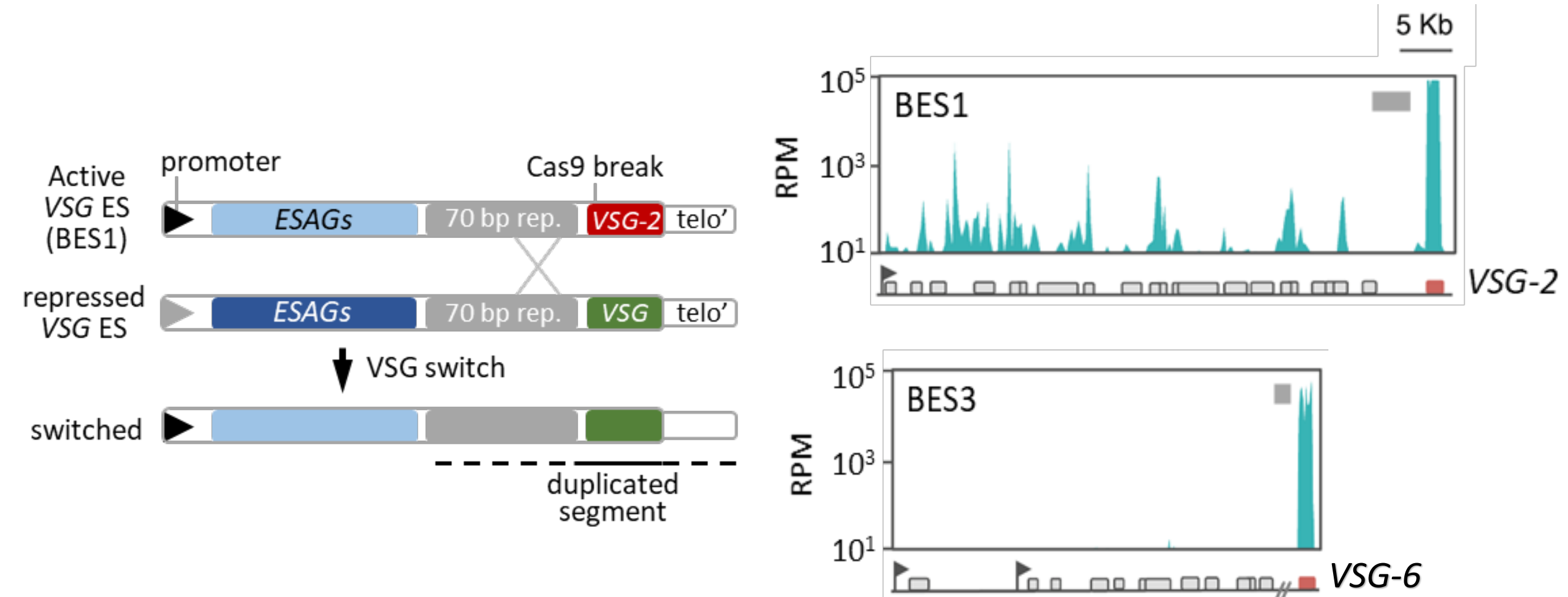
Cas9 break site 3 is more efficient in triggering VSG switching



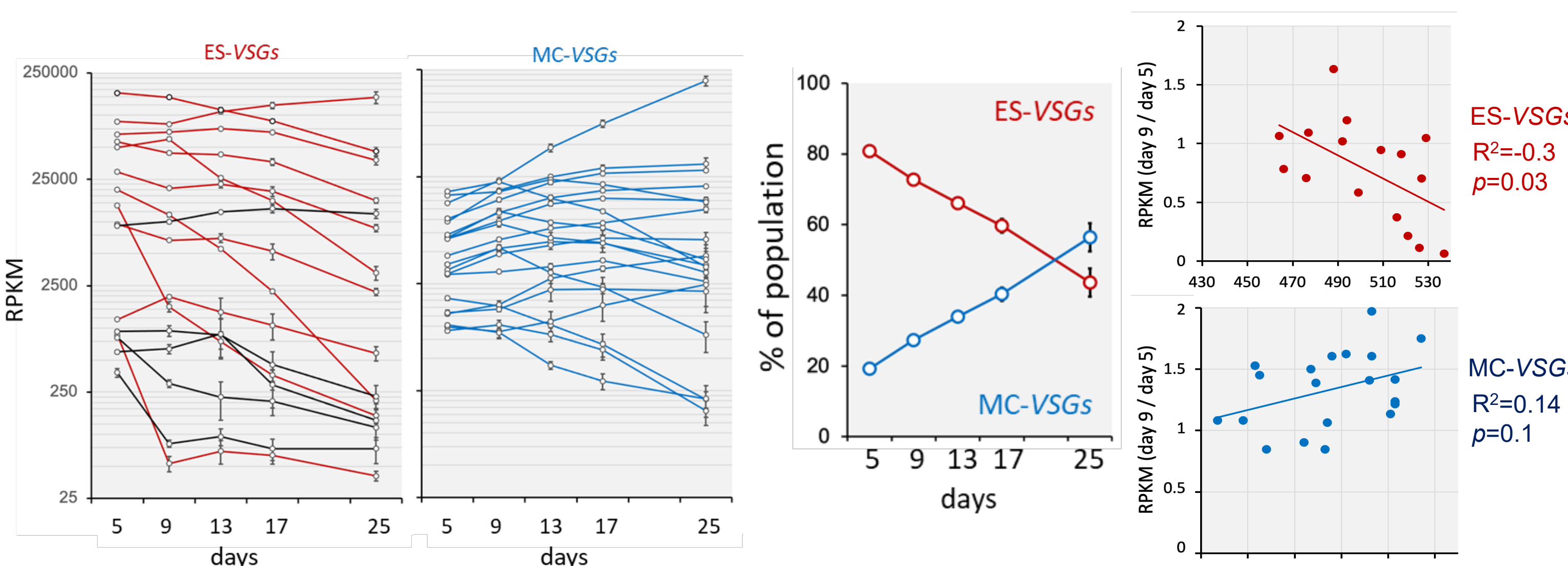
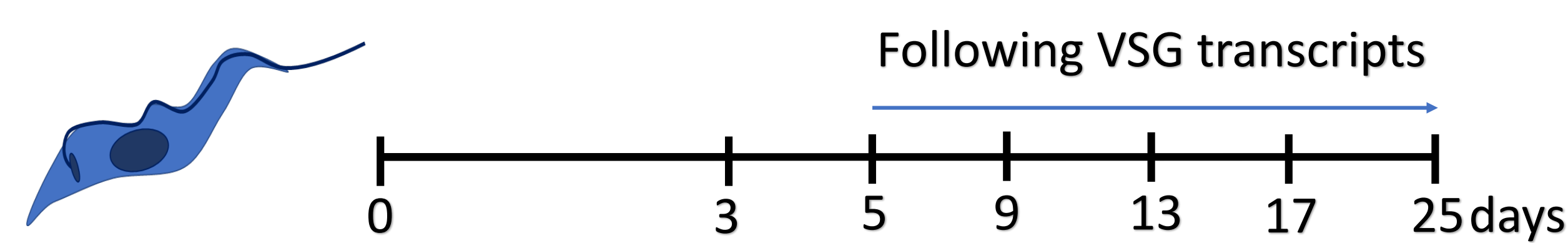
Proportion of VSG switchers over time



VSG-2 disruption by Cas9 triggers VSG-gene conversion



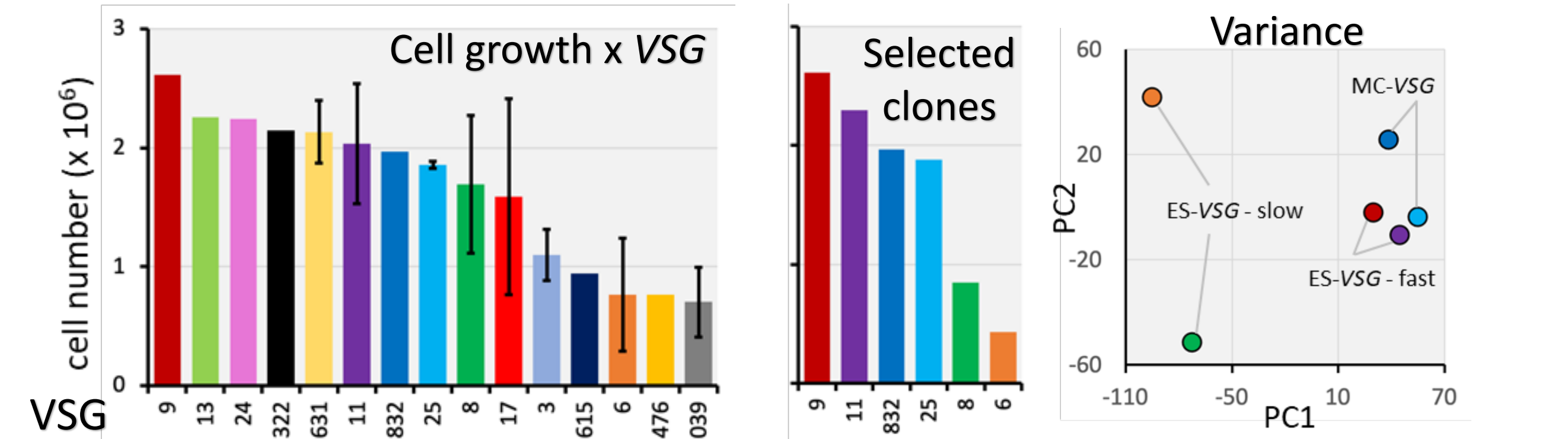
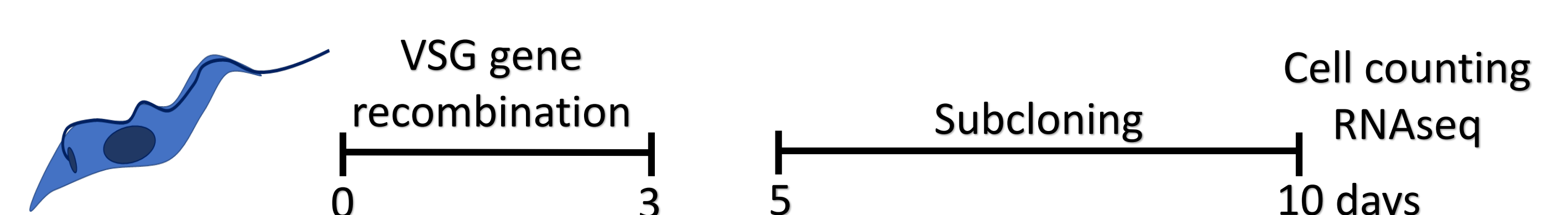
VSG dynamics differs according to their location



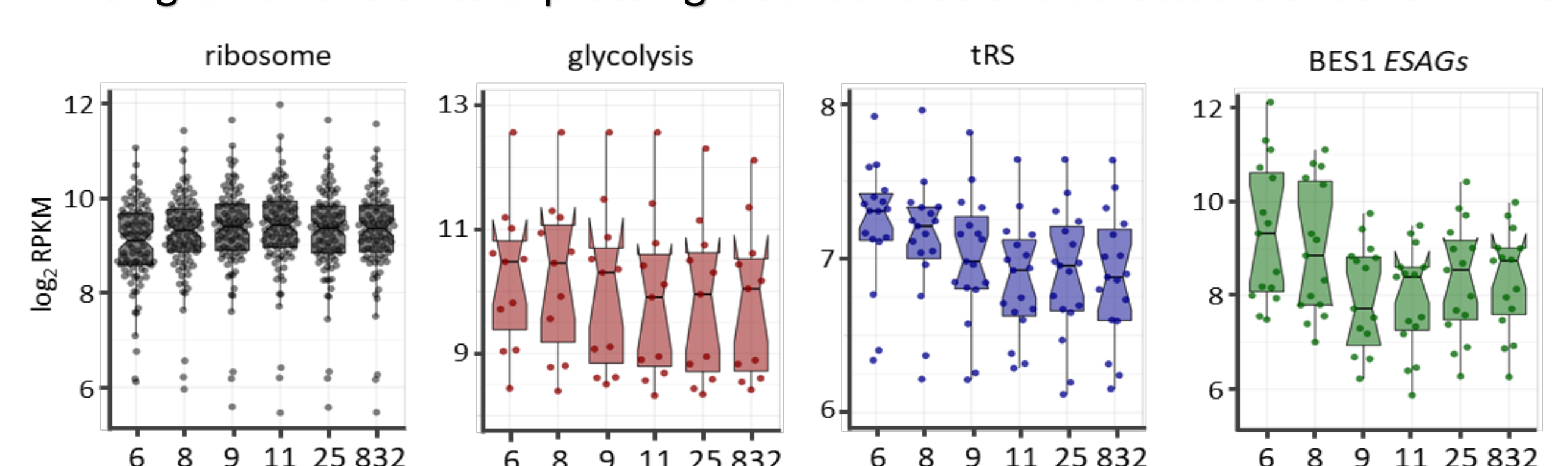
Abundance of VSG transcripts for ES and MC-VSGs

MC-VSGs become predominant in the population over time

Transcriptomic alterations in slow growth clones



Cell growth of clones expressing distinct VSGs and selected clones for RNAseq



Widespread transcriptomic differences observed for slower growth clones.

Conclusions

- Cells expressing different VSGs grow at different rates.
- Cells expressing minichromosomal VSGs display a selective advantage.
- Cells expressing longer VSGs display the greatest competitive differences.
- The transcriptome is altered in slower clones.
- All of the above features operate independent of a host immune response.

Contact

Douglas Escrivani de Oliveira
Email: Descrivanideolive001@dundee.ac.uk
ORCID:0000-0002-4872-8960