

3 replicates

A Barcoded pool
Log phase promastigotes
~8·10⁶ cells/ml



B Sand fly infections
150 sand flies per replicate




C DNA isolation

Start pool (1·10⁷ cells)

2 days PBM (50 flies)

6 days PB (50 flies)

9 days PB (50 flies)



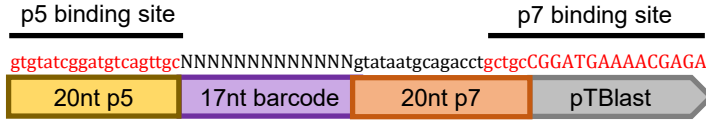
E Binding sites for library amplification from barcode regions

p5 binding site

p7 binding site

gtgtatcggatgtcagttgcNNNNNNNNNNNNgtataatgcagacctgctgcCGGATGAAAACGAGA

20nt p5 17nt barcode 20nt p7 pTBlast

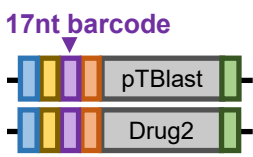


D Barcode read-out

17nt barcode

pTBlast

Drug2

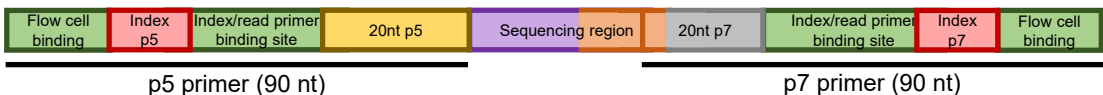


F Amplicon library for Illumina sequencing

Barcode + constant region (32 nt)

Flow cell binding Index p5 Index/read primer binding site 20nt p5 Sequencing region 20nt p7 Index/read primer binding site Index p7 Flow cell binding

p5 primer (90 nt) p7 primer (90 nt)



G Barcode location in SSU locus

SSU locus Calmodulin IR T7 RNAP Calmodulin IR 17nt barcode pTBlast SSU locus

pVY087 cassette

