Supplementary Figure 10 **LTED** b Positive Controls Scoring sgRNAs 4 Scoring CREs 2 Scoring CREs (>1 guide) replicate 1 2 Scoring Split 0 0.25 0.50 0.75 0.00 Fraction Scoring -2 Total CREs: 1,289 -No -4 7 21 14 C Non-targeting Expanded in LTED 22 days Exhausted in LTED 22 days log2-FC vs Library log2-FC vs Library 2 2 0 -2 7 21 14 7 14 21 7 14 21 d Day Day Day MYD88 -E2>>+E2 Log2-folc-change +E2 d21 -E2 d14 -E2 d60 LTED 14d +3 LTED 21d TLR5 +3 "+E2 d14 -E2>>+E2 "+E2 d21 Log2-folc-change -E2 d14 +3 -E2 d21 -E2 d60 LTED 14d +3 LTED 21d USP8 +E2 d14 +3 -E2>>+E2 Log2-folc-change +E2 d21 -E2 d14 -E2 d21 -E2 d60 LTED 14d

LTED 21d

+3

-3

Supplementary Figure 10. SIDP identifies regulatory regions with context-dependent activity in LTED cells. (a) Bar plots showing the relative fraction of scoring sgRNAs and CREs bearing scoring sgRNAs, upon perturbation of LTED cells via SIDP. Scoring sgRNAs showing a significantly decreased frequency at 21 days post-infection are referred to as DF, while those with a significantly higher frequency as IF. Scoring CRE>1 refers to loci with multiple sgRNA scoring in the same direction (i.e. DF or IF. (b) Box plots overlaid to violin plots showing the log2-fold-change of positive controls and non-targeting sgRNAs in two replicates of LTED cells, at 7, 14 and 21 days, as compared to the initial library. (c) Box plots overlaid to violin plots showing the log2-fold-change of both scoring (either blue or yellow) and non-scoring (white) sgRNAs at 21 days post-infection in LTED cells, at 7, 14 and 21 days, as compared to the initial library. (d) SIDP results at the indicated loci are shown as IGV genome browser screenshots. For each of the indicated condition, the log2-fold change for each sgRNA is indicated, with bars proportional to the effect size, and colour reflecting the sign (blue = IF; yellow = DF).