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Supplemental Information

High-Throughput CRISPR Screening Identifies Genes Involved in Macrophage Viability and Inflammatory Pathways

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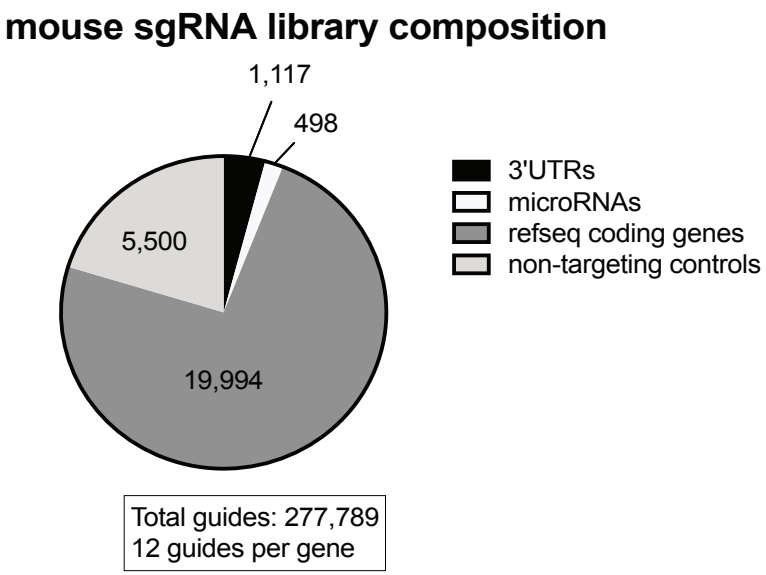
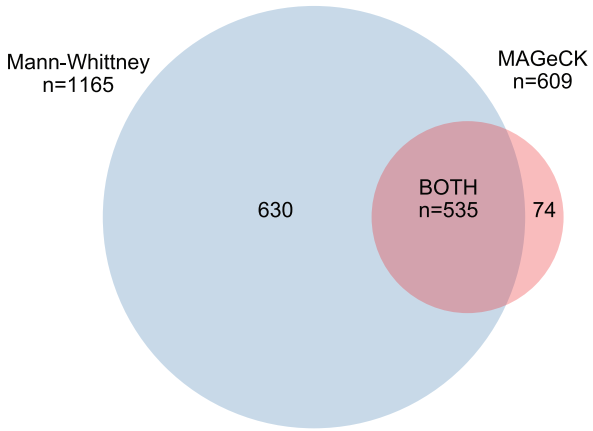
Figure S1.

A.



B.



C.

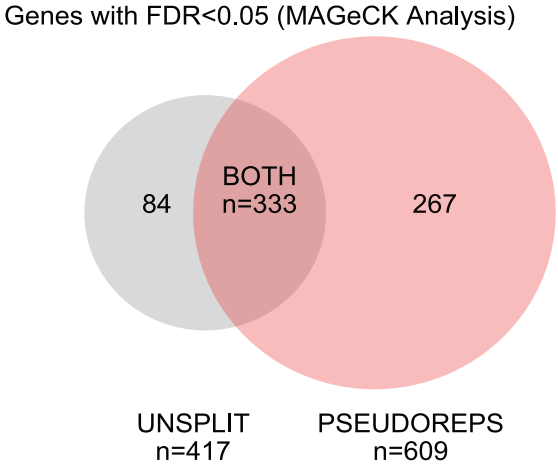
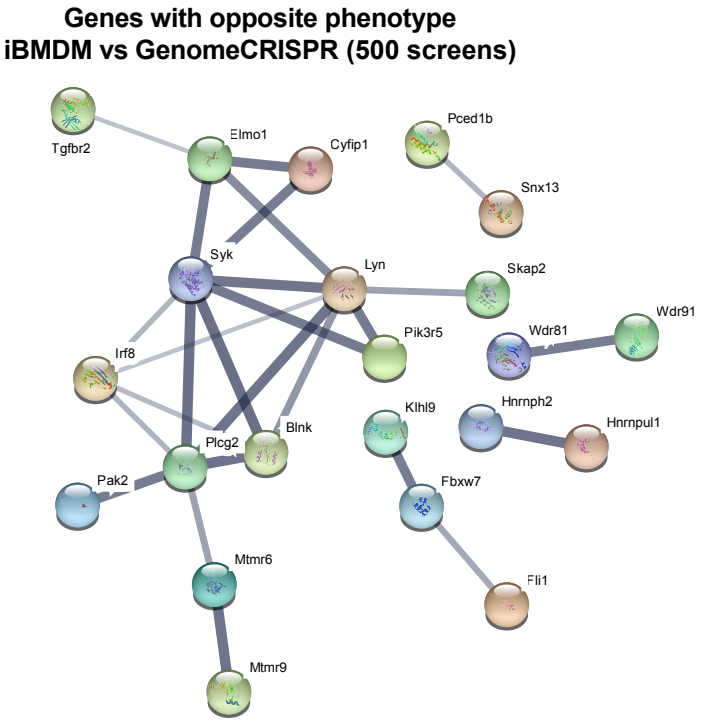


Figure S1: Comparing analysis screening tools, related to Figure 1. A. Breakdown of all genes targeted by our custom mouse sgRNA library is displayed.. B. Venn diagram. Significant genes were determined by Mann-Whittney U-test and MAGeCK analysis (FDR<0.05). 88% of significant genes identified in the MAGeCK analysis were also identified in the Mann-Whittney U-Test. C. Venn diagram. Significant genes were determined on unsplit and insamples replicates by MAGeCK analysis. 80% of the significant genes in the unsplit samples were also identified in the replicate samples.

Figure S2



pathway	description	
mmu04662	B-cell receptor signaling pathway	0.00023
mmu04664	Fc epsilon RI signaling pathway	0.0025
mmu04064	NF-kappa B signaling pathway	0.006
mmu04066	HIF-1 signaling pathway	0.0064

Figure S2: Comparison of current screen to CRISPR screen database, related to Figure 1. Genes with opposite phenotypes in our screen compared to the GenomeCRISPR database are displayed visually using String-DB. KEGG pathway Go-term enrichment is also shown.

Figure S3

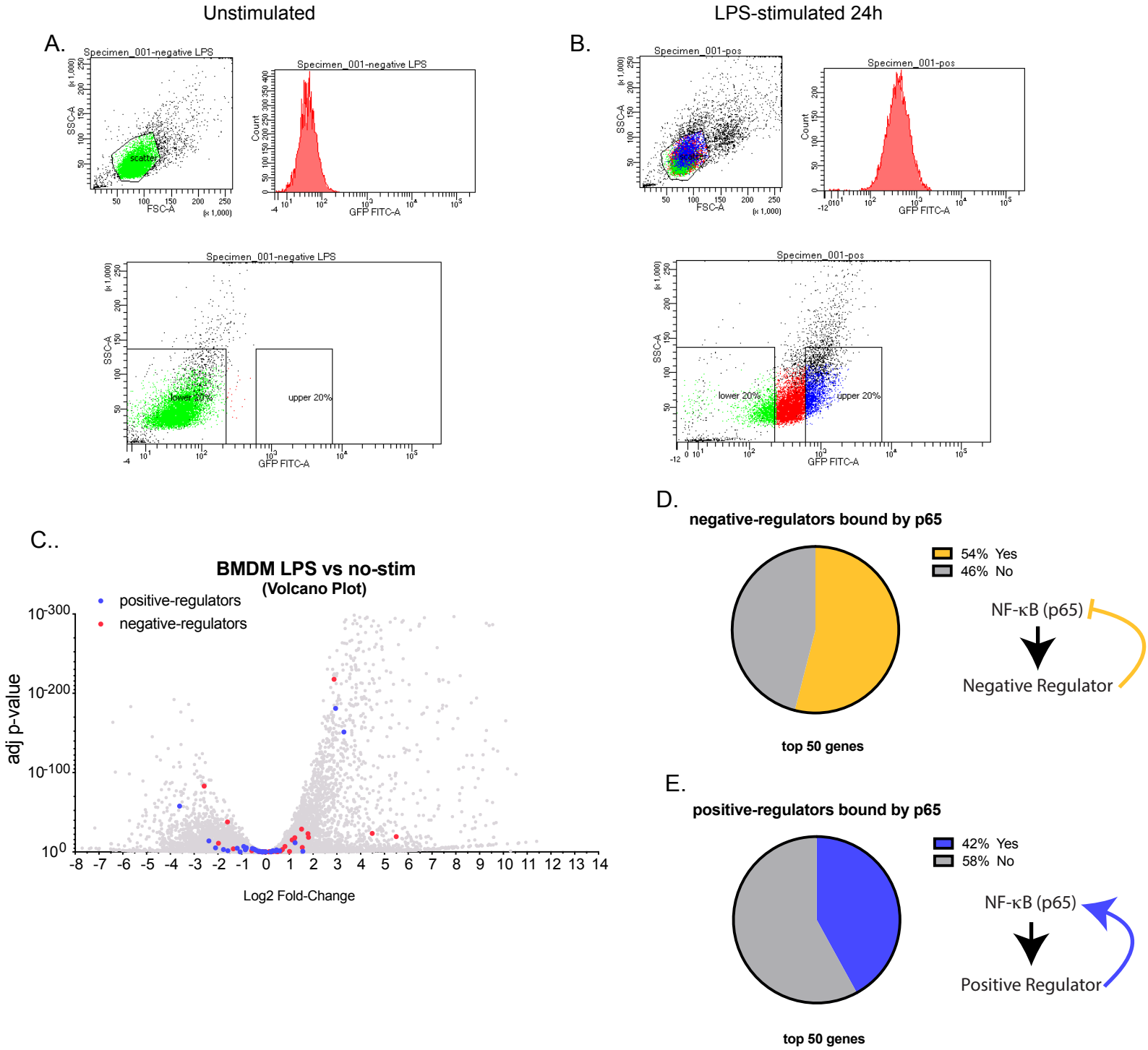


Figure S3: Expression and p65-binding of NF-kB screen hits, related to Figure 3. A-B. NF-κB FACS screen gating strategy for unstimulated cells (A) or 24 h LPS stimulated cells (B). C. Differentially expressed genes in 6 h LPS vs unstimulated BMDMs are displayed as log2 fold-change vs. adjusted p-value volcano plot from previously published data (Zhang et al., 2017). Expression of top 50 positive regulators (blue) and top 50 negative regulators (yellow) is shown. D-E. All p65 targets were determined using the ChIP-seq data from (Lam et al., 2013). Positive p65 binding was called if a p65 peak was greater than 10 and was within 1kb of the annotated transcription start site (TSS). P65 promoter binding was then assessed for the top negative (D) and positive (E) regulators.

Figure S4

A.

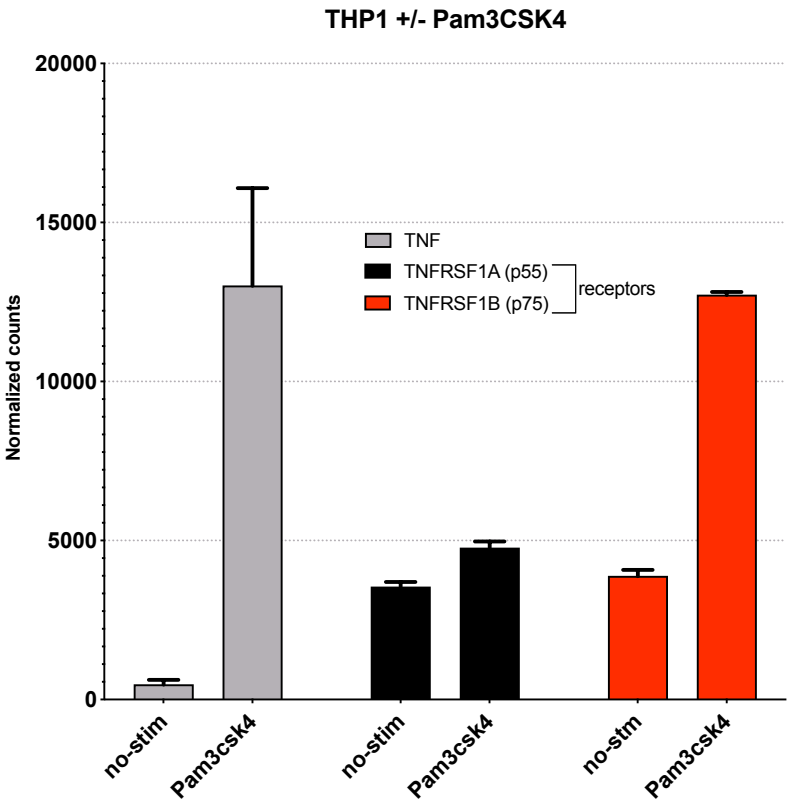


Figure S4: Expression of TNF and TNF receptors in human cells, related to Figure 4. A. Differentially expressed genes were determined for 6h Pam3CSK4-stimulated or unstimulated THP1 (ATCC). Normalized counts +/- SD are displayed for TNF, TNFRSF1A and TNFRSF1B.