

SigmaAldrich® CRISPRa Synergistic Activation Mediator (SAM)

What is CRISPRa and how can it complement your research?

By activating transcription, CRISPRa screening can reveal genes essential in biological pathways or drug resistance. Commonly used loss-of-function (LOF) screening is a powerful approach for uncovering gene targets to understand the molecular mechanisms behind health and disease. However, screening the over-expression, or activation of genes with CRISPR activators (CRISPRa, Fig. 1), can often reveal additional gene targets for therapeutics and uncover previously unknown molecular mechanisms in disease models, making it an essential tool.

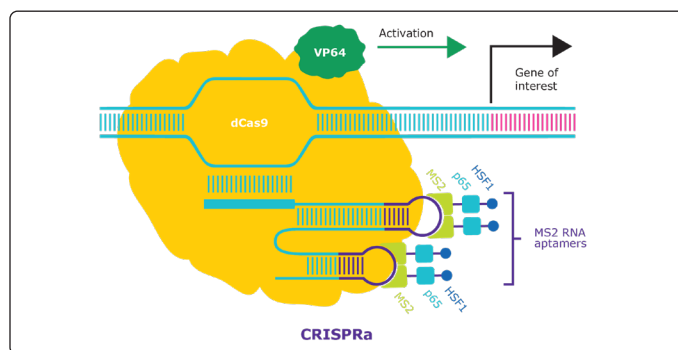


Figure 1. CRISPRa utilizes catalytically inactive dCas9-VP64 and transcriptional activators HSF1, p65, and MS2 to modulate gene expression

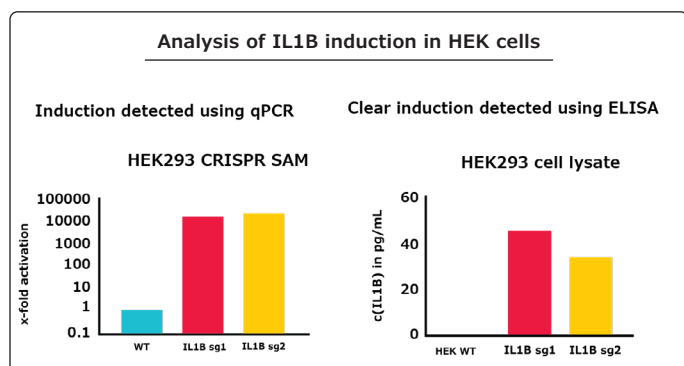


Figure 2. IL1B activation via qPCR and ELISA. In collaboration with Evotec, independent scientists show that the SAM system has consistent activation of IL1B using two different gRNAs targeting upstream of the transcriptional start site (TSS). qPCR reveals a substantial increase of IL1B transcript relative to control. Importantly, increased transcript level correlates with an increase in protein levels as measured by an ELISA assay

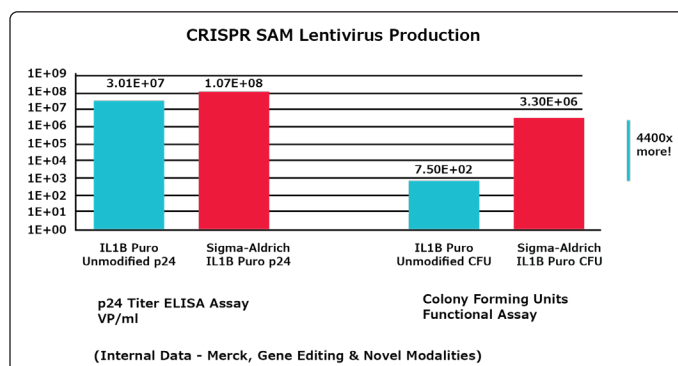


Figure 3. Lentivirus quantification by p24 and CFU assays. Both assays show increased lentiviral titer. Modifications made by our scientists to the vector show significant increase in functional titer when compared to competitors

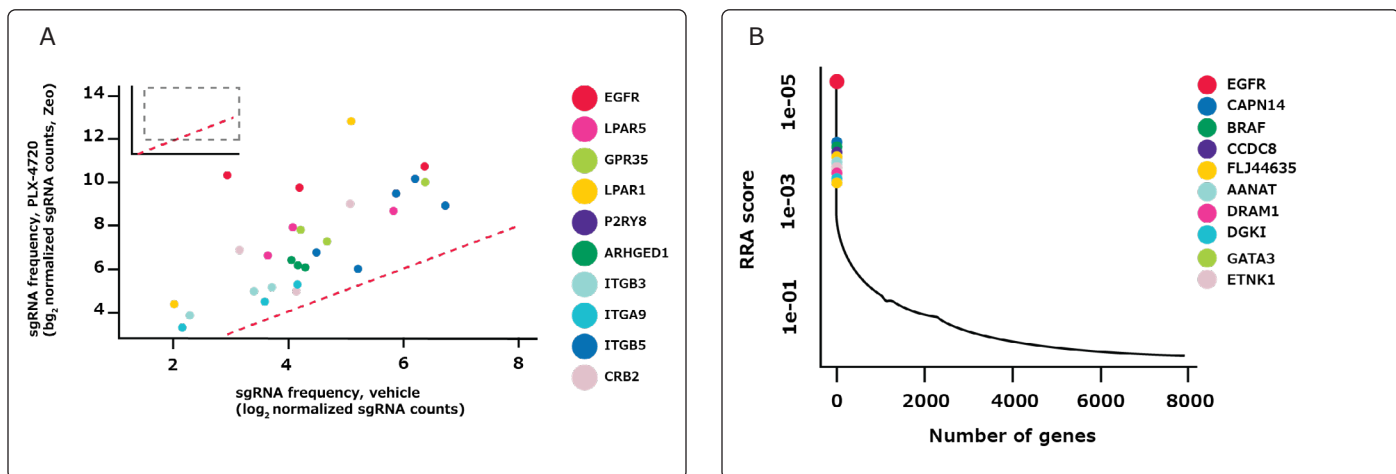


Figure 4. Sigma-Aldrich CRISPR SAM screen recapitulates data showing BRAF inhibitor resistant through EGFR. (A) Scatterplot showing enrichment of multiple sgRNAs per gene after 14 days of PLX-4720 treatment in A375 cells. Non-significant genes not shown. Adapted from Konermann et al., 2015. (B) Plot showing the distribution of RRA scores in treatment vs control for all genes in one subpool of the whole-genome library (alphabetically A thru G). As expected, EGFR was the most significantly enriched target gene. Analysis was performed using the MAGeCK pipeline (Li et al., 2014).

Product Information

Complete Kits					
LentiCRISPR Pools	Species	Description	Total of gRNAs	dCas9-VP64	Selection
HSAMPURO-1KT	Human	Whole Genome gRNA pools	70,771	No	puro
MSAMPURO-1KT	Mouse	Whole Genome gRNA pools	69,716	No	puro
HSAMZEO-1KT	Human	Whole Genome gRNA pools	70,771	No	zeo
MSAMZEO-1KT	Mouse	Whole Genome gRNA pools	69,716	No	zeo
Individual Components	Species	Description	Total # of gRNAs	dCas9-VP64	Selection
SAMVP64BSTV	Any	dCas9-VP64 lentiviral particles	N/A	Yes	blast
SAMVP64BST	Any	dCas9-VP64 plasmid DNA	N/A	Yes	blast
SAMMS2HYGV	Any	MS2-p65-HSF1 lentiviral particles	N/A	No	hygro
SAMMS2HYG	Any	MS2-p65-HSF1 plasmid DNA	N/A	No	hygro
SAMHELPERV	Any	dCas9-VP64 and MS2-p65-HSF1 lentiviral particles	N/A	Yes	blast; hygro
SAMHELPERP	Any	dCas9-VP64 and MS2-p65-HSF1 plasmid DNA	N/A	Yes	blast; hygro

Helper kits are needed to create a dCas9-VP64-MS2 SAM helper cell line prior to transduction of the library or pools

To place an order or receive technical assistance:

To learn more about CRISPR activation and inhibition visit:

SigmaAldrich.com/CRISPRa

To learn about the complete set of products that will take your genome engineering & modulation experiments Beyond the Bench visit :

SigmaAldrich.com/AdvancedGenomics

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