

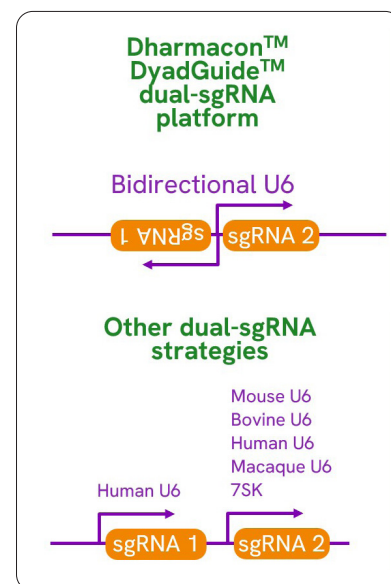


One novel promoter. Two guides. More possibilities.

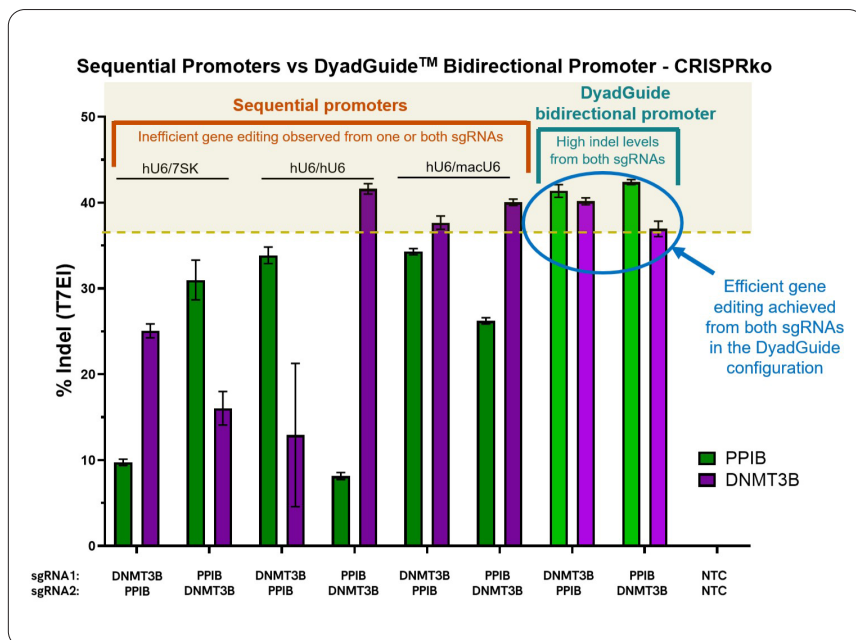
DyadGuide™ CRISPR system

Unmatched consistency in dual-sgRNA expression adds new dimensions to your CRISPR gene editing and modulation capabilities.

- Target the same gene with two sgRNAs for enhanced gene activation, repression, or knockout.
- Target two different genes to enable more complex experimental designs while reducing time, effort, and overall experimental risk.
- Available in sgRNA only and All-in-one sgRNA formats that include the appropriate dCas9-effector or Cas9 expressed within the same lentiviral vector.
- High quality lentiviral and glycerol formats, with Puromycin resistance (Puro^R) or EGFP selection options for additional selection or enrichment following lentiviral transduction.

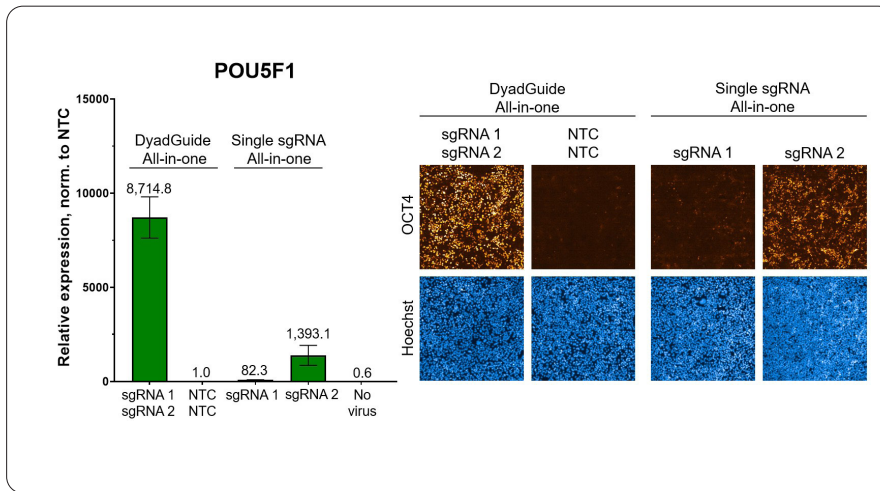


The DyadGuide dual-sgRNA platform utilizes a bidirectional U6 promoter to drive efficient and uniform expression of both sgRNAs, overcoming limitations seen when sgRNAs are expressed by multiple sequential promoters.



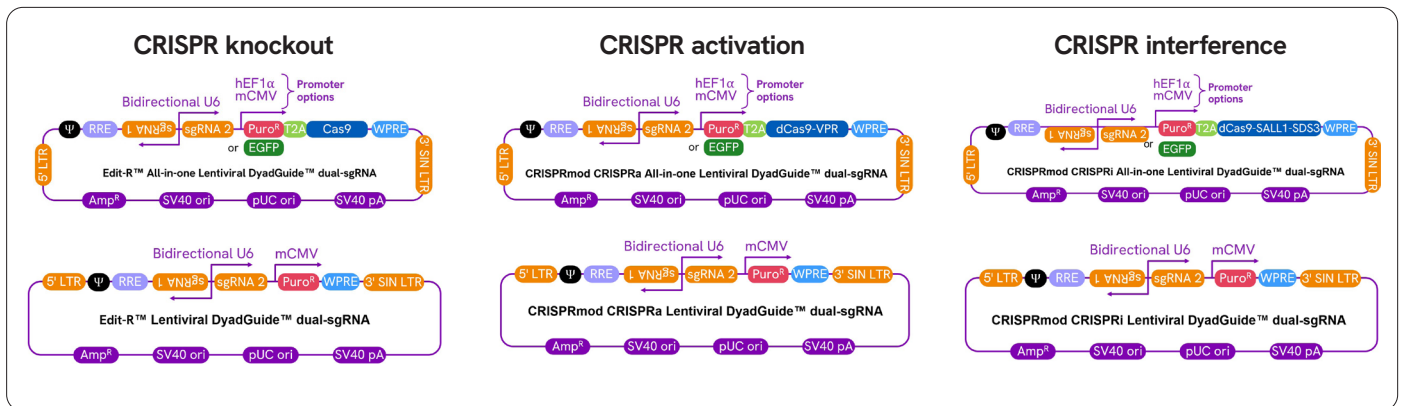
U2OS cells were transduced with Edit-R All-in-one Lentiviral DyadGuide dual-sgRNA particles containing sgRNAs targeting both PPIB and DNMT3B or All-in-one vectors containing sequential U6 promoters (combinations of Human U6 (hU6) and 7SK, hU6 and hU6, or hU6 and macaque U6 (macU6)) driving expression of sgRNAs targeting both PPIB and DNMT3B. Edit-R All-in-one Lentiviral DyadGuide dual-sgRNA particles containing two non-target control (NTC) sgRNAs were used as a control. Cells were selected with 2.5 µg/µL puromycin for 5 days. Genomic DNA was extracted for DNA mismatch detection with T7 Endonuclease I (T7EI) Indel quantification assay.

Synergistic CRISPR activation is achieved with two sgRNAs targeting the same gene when expressed from the bidirectional U6 promoter in the DyadGuide dual-sgRNA expression platform



U2OS cells were transduced with CRISPRmod CRISPRa All-in-one Lentiviral DyadGuide dual-sgRNA particles containing two sgRNAs each targeting POU5F1 or CRISPRmod CRISPRa All-in-one lentiviral particles containing a single sgRNA targeting POU5F1. CRISPRmod CRISPRa All-in-one Lentiviral DyadGuide dual-sgRNA particles containing two non-target control (NTC) sgRNAs were used as a control. Cells were selected with 2.5 µg/µL puromycin for 5 days. (Left) Total RNA was isolated and relative gene expression was measured using RT-qPCR. Robust, synergistic POU5F1 gene activation was observed with the CRISPRmod CRISPRa All-in-one Lentiviral DyadGuide dual-sgRNA system, greatly exceeding the levels achieved by the single sgRNA CRISPRmod CRISPRa All-in-one lentiviral system. (Right) Replicate wells were stained with OCT4 (encoded by POU5F1) antibody and counterstained with Hoechst. Similar to the mRNA levels, OCT4 protein levels were much stronger in wells transduced with the CRISPRmod CRISPRa All-in-one Lentiviral DyadGuide dual-sgRNA system driving two sgRNAs against POU5F1 compared to expression of individual sgRNAs.

The DyadGuide dual-sgRNA expression platform is available in a variety of formats to best suit your experimental needs



Vector diagrams for the DyadGuide dual-sgRNA platform include solutions for CRISPRko, CRISPRa, and CRISPRi

The DyadGuide dual-sgRNAs are available as:

- sgRNA or All-in-one Cas9 + sgRNA formats for CRISPR knockout, CRISPR activation, or CRISPR interference.
- High quality, concentrated, purified lentiviral particles for direct transduction with minimal cytotoxicity; delivered at titers of $\geq 10^8$ TU/mL for dual-sgRNA only and $\geq 10^7$ TU/mL for All-in-one sgRNA formats; glycerol stocks also available.



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