Custom Lentiviral CRISPR, RNAi and cDNA Constructs

CELLECTA

Discovery is yours™

Lentiviral expression vectors are one of the most effective vehicles to introduce and stably express CRISPR sgRNA, shRNA, cDNA, or reporter genes in almost any mammalian cell, including non-dividing cells and whole model organisms. Lentiviral expression constructs packaged into pseudoviral particles can be transduced into cells with very high efficiency--approaching 100% in some cell types--even in those cells most difficult to transfect, such as primary, stem, and differentiated cells.

- Different antibiotic (puro, bleo, neo, blast) and fluorophore markers (GFP, RFP) are available.
- Constructs are provided as plasmid and/or packaged lentiviral particles at a range of titers.

Wide Range of Options for Custom sgRNA Constructs

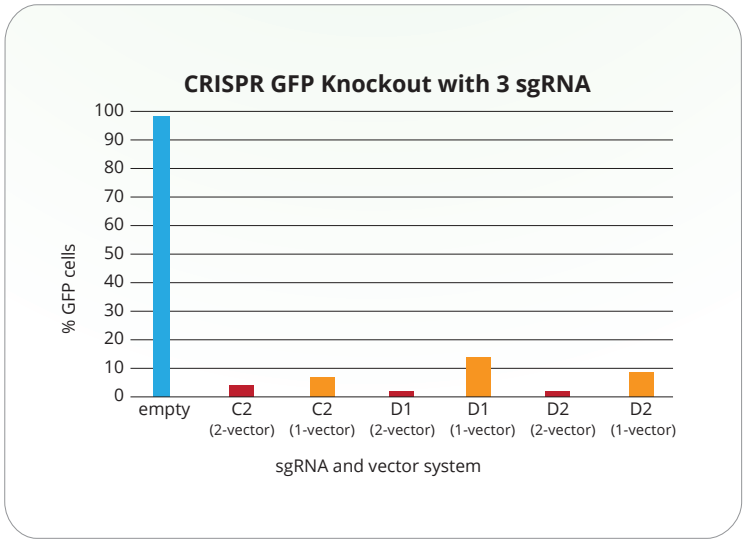
The CRISPR/Cas9 system can be used for gene knockout (KO), knockdown, activation or to initiate knock-ins in vivo or in vitro by using a combination of an sgRNA (single-guide RNA) together with a Cas9 nuclease.

Some examples of custom constructs include

- Constitutive or tet-inducible sgRNA constructs designed for CRISPR KO, CRISPRa, or CRISPRi.
- All-in-one constructs with sgRNA and Cas9 or single-vector sgRNA-only formats
- Complete panels of Cas9 and dCas9-hybrid (e.g., dCas9-KRAB, dCas9-VPH, dCas9-VPR, etc.) expression constructs

Just provide the gene information and we will select the optimal guide designs.

Get the CRISPR KO, CRISPRi, or CRISPRa sgRNA constructs you want, simply and easily.



Construction of shRNA Custom Constructs

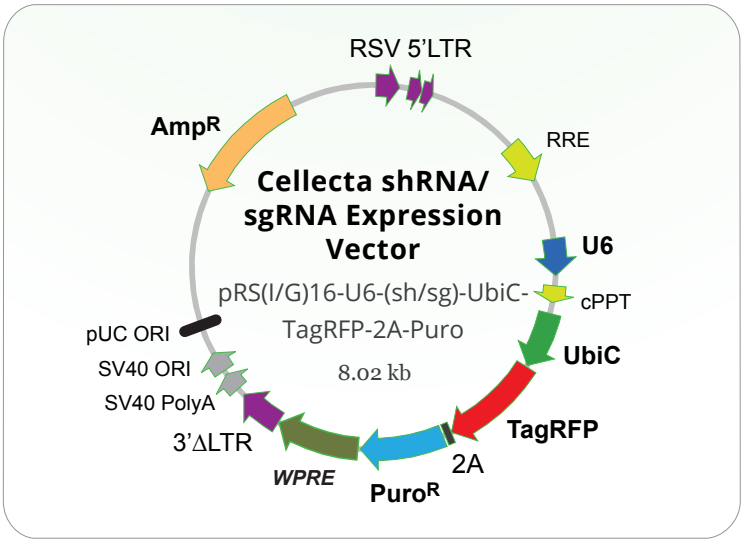
For custom shRNA constructs, we only need to know the RefSeq identification for the gene transcript and we will design any number of shRNA constructs that target it. Choose from a range of vectors with various selections and constitutive or inducible H1 or U6 shRNA promoters.

Stable Expression of Your Gene of Interest

Get a lentiviral construct expressing your gene of interest. Lentiviral vectors stably integrate at high efficiency and are passed onto daughter cells, so it is easy to make clonal expression lines. Even cell pools without clonal selection can typically be treated, grown for several passages, freeze/thawed, without disrupting the construct.

We Also Provide Cell Engineering Services

Want to knock out a gene, knock in a tag to an endogenous gene, get a cell line expressing Cas9, or expressing a different gene of interest? You can just have us make them for you. We offer a complete range of cell engineering services to make Cas9 expression cells, knockout/knockdown genes of interest, or make cells expressing a desired protein. Let us know what you want and we will go through the options with you.



To learn more about these and other Cellecta custom services, visit www.cellecta.com/services or email info@cellecta.com

Custom CRISPR sgRNA Pooled Libraries

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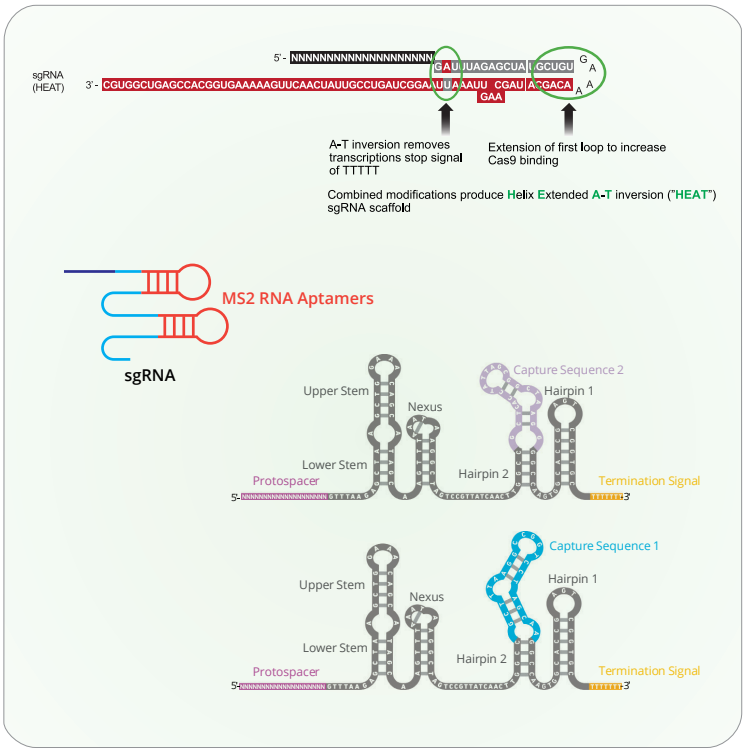
Get the right library for your screen!

In addition to standard CRISPR knockout, specialized applications often require sgRNA libraries with non-standard designs, including:

- sgRNA libraries for CRISPRa and CRISPRi libraries
- Libraries compatible with the CRISPRa SAM complex that requires a modified tracr design
- Single-cell applications that use guides with specific "capture sequences" in the tracrRNA segment (e.g. capture sequences compatible with 10X Chromium Single Cell 3' v3 Gel Beads)
- Studies that combine expression analysis with CRISPR screens and thus need to detect the sgRNA by RNA sequencing
- Designs where barcode or unique molecular identifier (UMI) sequences combined with sgRNAs are required
- sgRNAs that are compatible with other CRISPR Cas proteins (e.g., Cpf1)

Cellecta's **custom sgRNA construct and library service** easily accommodates all the above variations and more. Cellecta has over 15 years of experience making high quality pooled shRNA, barcode, and sgRNA libraries targeting virtually any defined sequences. Just let us know what you need, and we will provide it.



Cellecta Custom CRISPR Libraries

1. Oligo Design and Synthesis

We will design sgRNAs to most genes incorporating our HEAT-modified, improved sgRNA scaffold structure. For more specialized applications, researchers may also provide their own guide sequences.

2. Cloning

After the design step, we synthesize and clone the pool of oligos in any of our standard library vectors or we can make the library in a customer-provided vector. We also have available sets of non-targeting, intron-targeting, and lethal sgRNA controls that can be incorporated into the library.

3. Quality Analysis

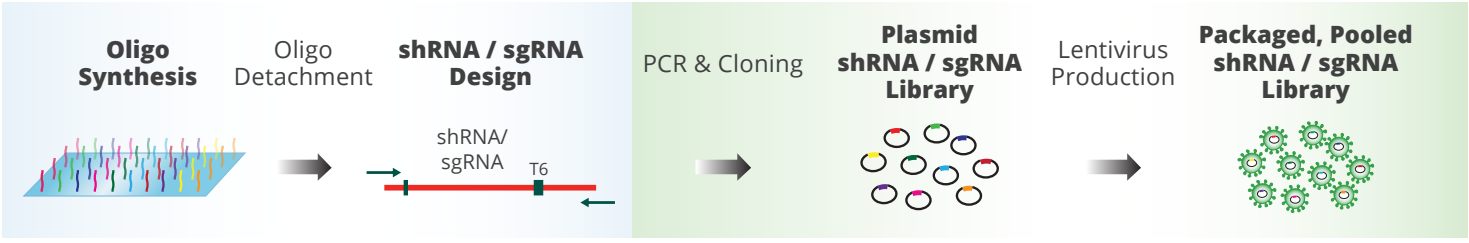
Once the library is made, we isolate a few dozen constructs for full-insert Sanger sequencing to confirm the configuration of the sgRNA expression cassette, and we deep sequence all guide sequences by NGS to confirm full representation of the oligo pool and assess distribution. Libraries that do not meet our standards are remade.

4. Deliverables

On completion, we provide 500 µg of the plasmid library with:

- All sequence information on the sgRNA guides and vector
- The cloning site design
- Primer information for sequencing
- NGS sgRNA distribution data

The whole process takes approximately two months once the gene list is finalized. Additional services to prepare VSV-g pseudotyped viral particles ready to transduce in cells are available.



For more information on Cellecta custom services, visit www.cellecta.com/services or email info@cellecta.com