



## **Genome-TALER™ & Genome-CRISP™ Human AAVS1 Safe Harbor Gene Knock-in Kits**

**Catalog# SH-AVS-K100**

**Catalog# SH-AVS-K000**

**Catalog# SH-AVS-K200**

**Catalog# SH-AVS-K002**

### **User Manual**

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## **USER MANUAL**

### **Genome-TALER™ Human AAVS1 Safe Harbor Gene Knock-in Kit**

### **Genome-CRISP™ Human AAVS1 Safe Harbor Gene Knock-in Kit**

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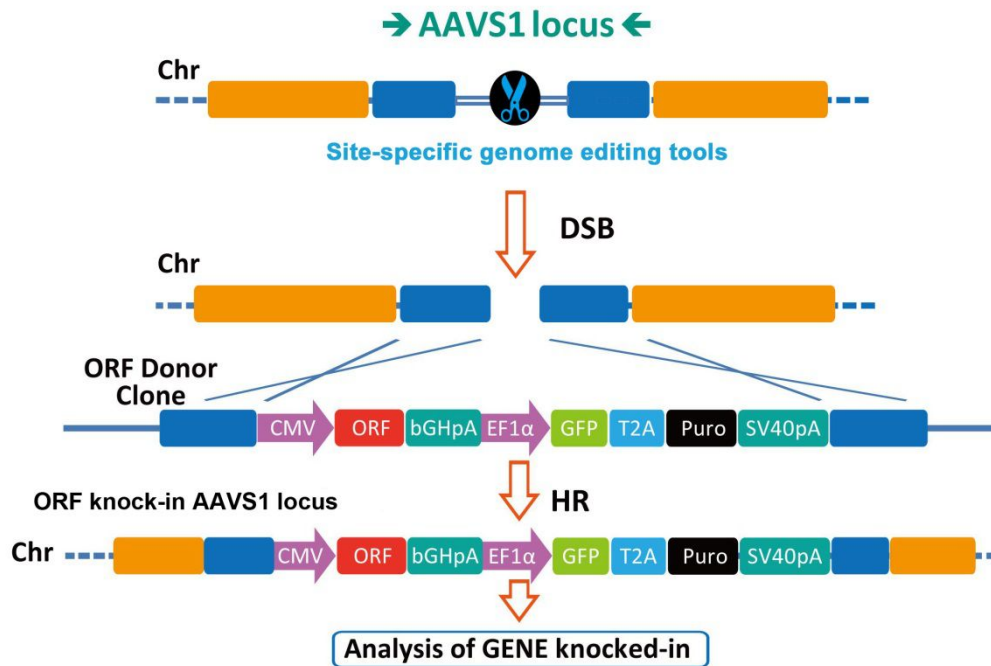
## **I. Introduction**

### **Safe gene targeting**

Genome modification by insertion of genes of interest and other genetic elements in unique site(s) of chromosome(s) is of great value for cell engineering. The genetically modified cells are valuable for therapeutic research, gene function study as well as lineage tracking and analysis. All these applications depend on the reliable and predictable function of the transgene without perturbing any endogenous gene and/or other regulation element. Random integration of the transgene, on the contrary, can present a threat of unpredicted insertion or mutagenesis.

The new approach recently developed is to deliver the transgene to a predetermined and safe site in a genome. AAVS1 (also known as the PPP1R2C locus) on human chromosome 19, is a well-validated “safe harbor” to host a DNA fragment with expected function. It has an open chromatin structure and is transcription-competent. Most importantly, there is no known adverse effect on the cell resulting from the inserted DNA fragment of interest.

The GeneCopia AAVS1-specific TALEN or CRISPR-Cas9 system can generate a DNA double-strand break (DSB) in AAVS1 on human chromosome 19, stimulating natural DNA repair mechanisms. In the presence of AAVS1 ORF knockin clones, homologous recombination (HR) occurs, resulting in integration of the DNA fragment from the ORF knockin clone into the safe harbor locus.



**Figure 1.** Illustration of genome-editing-tool-mediated transgene integration at the human safe harbor AAVS1 site.

### Introduction to TALEN

Transcription activator-like (TAL) effectors can recognize and bind host plant promoter sequences through a central repeat domain consisting of a variable number of ~34 amino acid repeats. The residues at the 12th and 13th positions of each repeat provide a simple one-to-one code for binding to each DNA base in the target sequence, e.g. NI = A, HD = C, NG = T, and NN = G or A.

TAL effectors have been utilized to create site-specific gene-editing tools by fusing target sequence-specific TAL effectors to nucleases (TALENs), transcription factors (TALE-TFs) and other functional domains. These fusion proteins can recognize and bind chromosome target sequences specifically and execute their gene-editing functions, such as gene knockout, knockin (with donor plasmid), modification, activation, repression and more.

### Introduction to CRISPR-Cas9

In the CRISPR-Cas9 system, the complex of a CRISPR RNA (crRNA) annealed to a trans-activating crRNA (tracrRNA) is sufficient to guide the Cas9 endonuclease to a specific genomic sequence to generate a double-strand break (DSB) in the target DNA. This system can be simplified by fusing crRNA and tracrRNA sequences to produce a synthetic chimeric single-guided RNA (sgRNA). The selected target sequence consists of a 20-bp DNA sequence complementary to the crRNA or the chimeric sgRNA, followed by the trinucleotide (5'-NGG-3') protospacer adjacent motif (PAM) which is recognized by the Cas9 and essential for cleavage. This RNA-guided DNA recognition mechanism of CRISPR-Cas9 provides a simple but powerful tool for precise genome engineering.

## Human AAVS1 Safe Harbor Gene knock-in Kits

**The GeneCopoeia Genome-TALER™ human AAVS1 safe harbor gene knock-in kit** is designed to specifically transfer your gene of interest, selection marker or other genetic element from a donor plasmid into the AAVS1 safe harbor site on human chromosome 19 via TALEN-mediated homologous recombination (HR). HR is a natural DNA repair mechanism that occurs in response to DNA double-strand breaks (DSB). This DSB is created by a pair of AAVS1-specific TALENs.

**The GeneCopoeia Genome-CRISP™ human AAVS1 safe harbor gene knock-in kit** is designed to specifically transfer your gene of interest, selection marker or other genetic element from a donor plasmid into the AAVS1 safe harbor site on human chromosome 19 via CRISPR-Cas9-mediated homologous recombination (HR). HR is a natural DNA repair mechanism that occurs in response to DNA double-strand breaks (DSB). This DSB is created by an AAVS1-specific CRISPR-Cas9 system.

### Advantages

#### Safe integration

Designated AAVS1 human genome safe-harbor integration site ensures transcription-competency of the transgenes and presents no known adverse effect on cells

#### Specific targeting

TALEN- or CRISPR-mediated DNA DSBs at the AAVS1 site stimulate homologous recombination dramatically for transgene integration

#### Single copy number

Single copy number of the transgene ensures predictable expression levels, simplifies phenotype interpretation, and prevents transgene silencing.

#### Compatible knock-in ORFs

Over 20,000 sequence-verified human ORFs are compatible for transgene donor DNA design

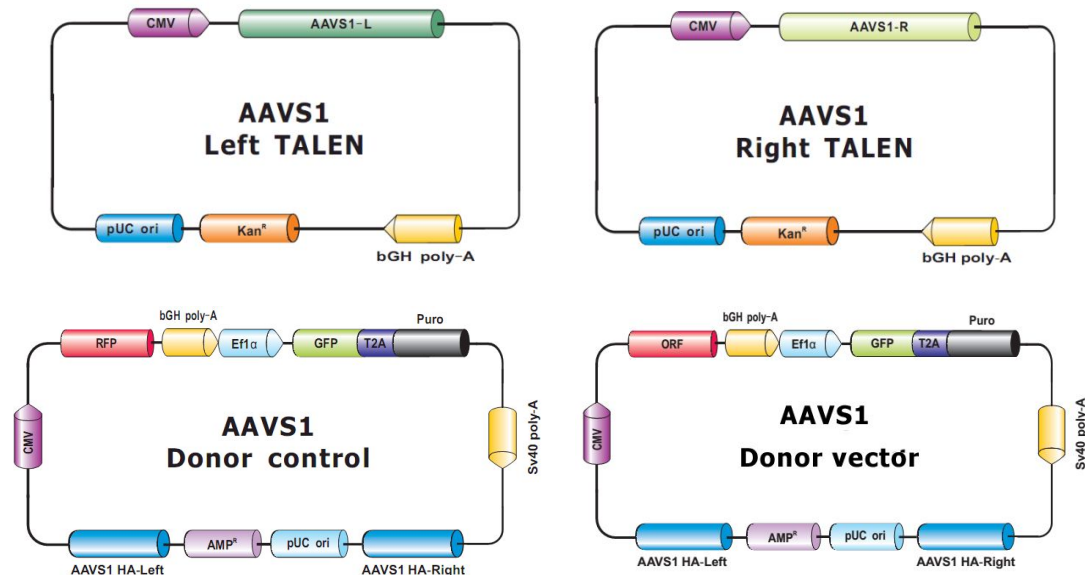
## II. Contents and Storage

**Genome-TALER™ human AAVS1 safe harbor gene knock-in kit (Cat# SH-AVS-K100)**  
**Genome-TALER™ human AAVS1 safe harbor gene knock-in kit (without donor) (Cat# SH-AVS-K000)**

Cat. No.	Product Name	Qty	Concentration	Shipping and Storage
<b>TN-AAVS1-L</b>	AAVS1 left TALEN	10 µg	500 ng/µl	Shipped at room temperature. Stored at -20°C
<b>TN-AAVS1-R</b>	AAVS1 right TALEN	10 µg	500 ng/µl	Shipped at room temperature. Stored at -20°C
<b>DC-DON-SH01*</b>	AAVS1 donor vector	10 µg	500 ng/µl	Shipped at room temperature. Stored at -20°C
<b>DC-RFP-SH01</b>	AAVS1 RFP control	10 µg	500 ng/µl	Shipped at room temperature. Stored at -20°C
<b>HQPAVSHR-5</b>	5' HR primer pair	200 reactions	10 µM	Shipped at room temperature. Stored at -20°C
<b>HQPAVSHR-3</b>	3' HR primer pair	200 reactions	10 µM	Shipped at room temperature. Stored at -20°C

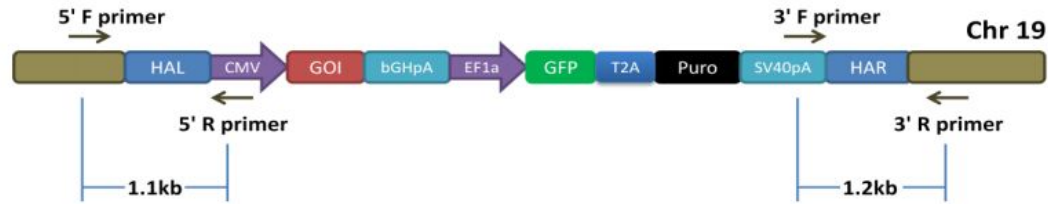
\* DC-DON-SH01 only comes with the SH-AVS-K100 kit. AAVS1 knock-in ORF donor clones can be searched from our collection of 20,000 knock-in clones and purchased separately.

### (A) TALEN and donor plasmids



## Human AAVS1 Safe Harbor Gene knock-in Kits

### (B) Knock-in verification PCR primers



**Figure 2.** AAVS1 safe harbor gene knock-in kit components. **(A)** TALEN and donor plasmids **(B)** Knock-in verification primer pairs

**Acknowledgement:** Design of the AAVS1 left TALEN, AAVS1 right TALEN, and AAVS1 donor control vectors was performed by Dr. Jizhong Zou of the NIH Center for Regenerative Medicine, a Common Fund initiative of the U.S. National Institutes of Health.

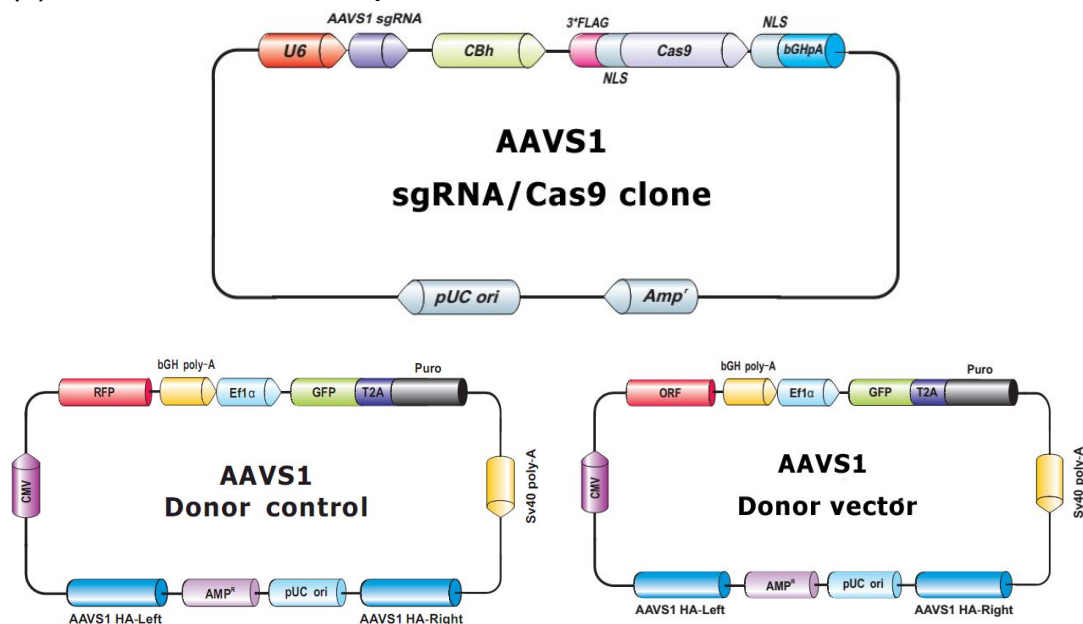
**Genome-CRISP™ human AAVS1 safe harbor gene knock-in kit (Cat# SH-AVS-K200)**  
**Genome-CRISP™ human AAVS1 safe harbor gene knock-in kit (without donor) (Cat# SH-AVS-K002)**

Cat. No.	Product Name	Qty	Concentration	Shipping and Storage
<b>HCP-AAVS1-CG02</b>	All-in-one AAVS1 sgRNA /Cas9 expression clone	10 µg	500 ng/µl	Shipped at room temperature. Stored at -20°C
<b>DC-DON-SH01*</b>	AAVS1 donor vector	10 µg	500 ng/µl	Shipped at room temperature. Stored at -20°C
<b>DC-RFP-SH01</b>	AAVS1 RFP control	10 µg	500 ng/µl	Shipped at room temperature. Stored at -20°C
<b>HQPAVSHR-5</b>	5' HR primer pair	200 reactions	10 µM	Shipped at room temperature. Stored at -20°C
<b>HQPAVSHR-3</b>	3' HR primer pair	200 reactions	10 µM	Shipped at room temperature. Stored at -20°C

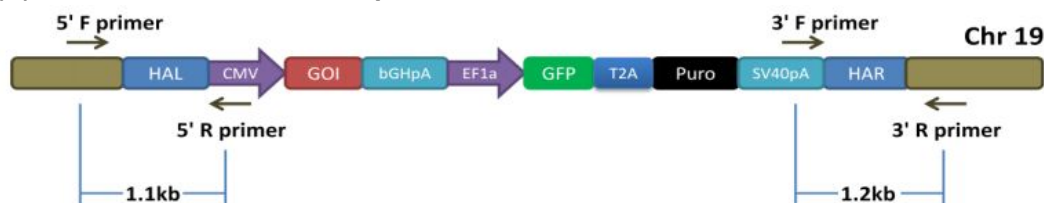
\* DC-DON-SH01 only comes with SH-AVS-K200 kit. AAVS1 knock-in ORF donor clones can be searched from our collection of 20,000 knock-in clones and purchased separately.

## Human AAVS1 Safe Harbor Gene knock-in Kits

### (A) CRISPR-Cas9 and donor plasmids



### (B) Knock-in verification PCR primers



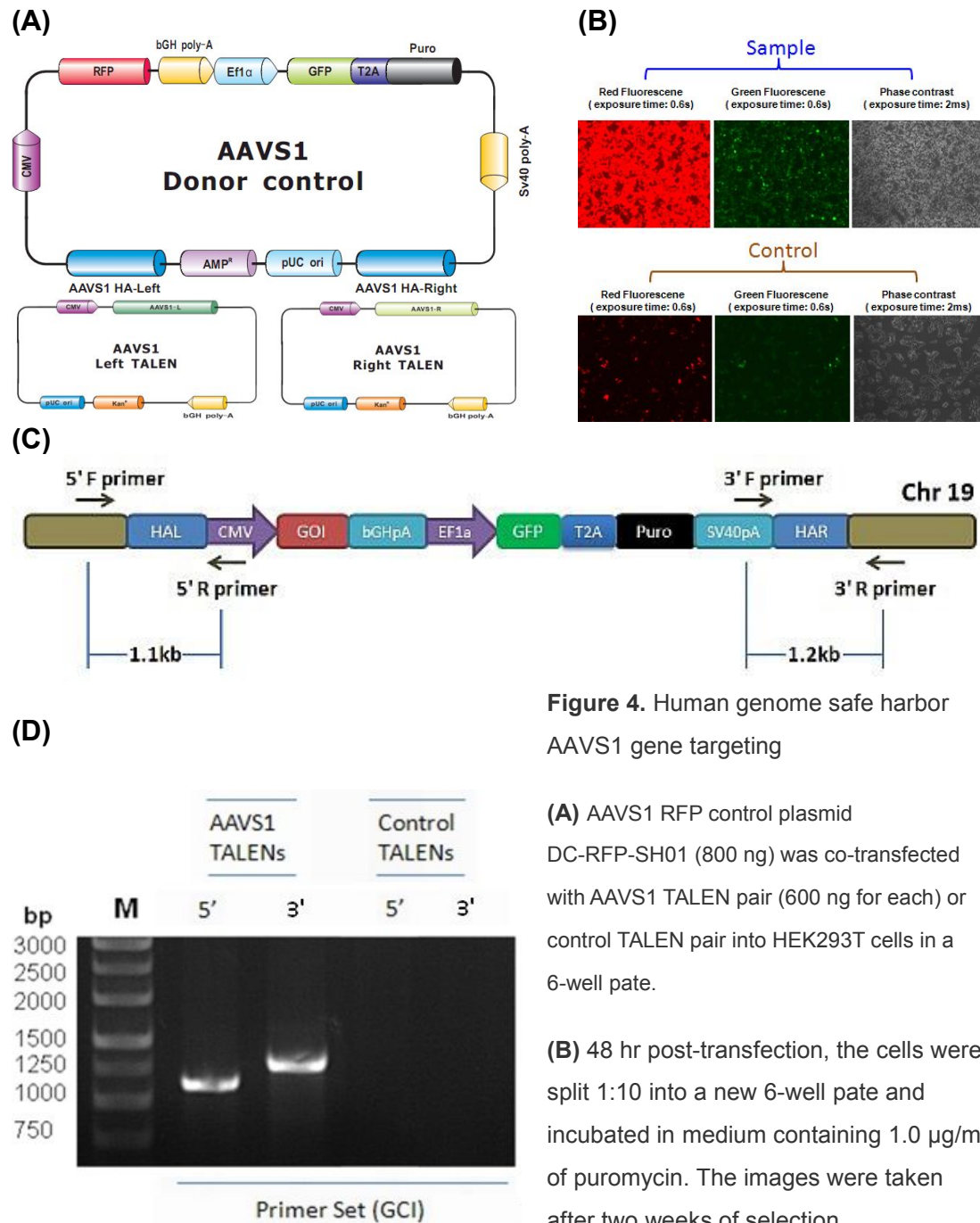
**Figure 3.** AAVS1 safe harbor gene knock-in kit components. **(A)** AAVS1 CRISPR-Cas9 and donor plasmids **(B)** Knock-in verification primer pairs

### Additional materials required

1. LB Agar and broth containing 50 µg/ml kanamycin
2. 6-well tissue culture plates and related tissue culture supplies
3. Other specific media and additives specific for cell type of interest
4. Any high-transformation efficiency RecA- and EndA- E.coli competent cells (GCI-5a chemically competent E. Coli, Cat# STK200-10 or -20)
5. Dulbecco's Modified Eagle's Medium (D-MEM) high glucose with sodium pyruvate and glutamine (Invitrogen, Cat. # 11995073)
6. EndoFectin™ Plus Transfection Reagent (Genecopoeia, Cat. # EFP1003-01/02)
7. Qiagen EndoFree Plasmid Maxi Kit (Qiagen, Cat. # 12362)
8. Qiagen DNeasy Blood and Tissue Kit (Qiagen, Cat. # 69504)
9. iProof High-Fidelity DNA Polymerase (BioRad, Cat. # 172-5301)
10. Fetal Bovine Serum (Invitrogen, Cat. # 16000036)
11. Penicillin/Streptomycin (Invitrogen, Cat. # 15070063)
12. Trypsin-EDTA (Sigma, Cat. # T3924)
13. **\*\*Optional\*\*** - For difficult-to-transfect cells, the use of an electroporation system (e.g. Lonza's NucleoFector or Invitrogen's Neon system) is highly recommended

# Human AAVS1 Safe Harbor Gene knock-in Kits

## III. Example



**Figure 4.** Human genome safe harbor AAVS1 gene targeting

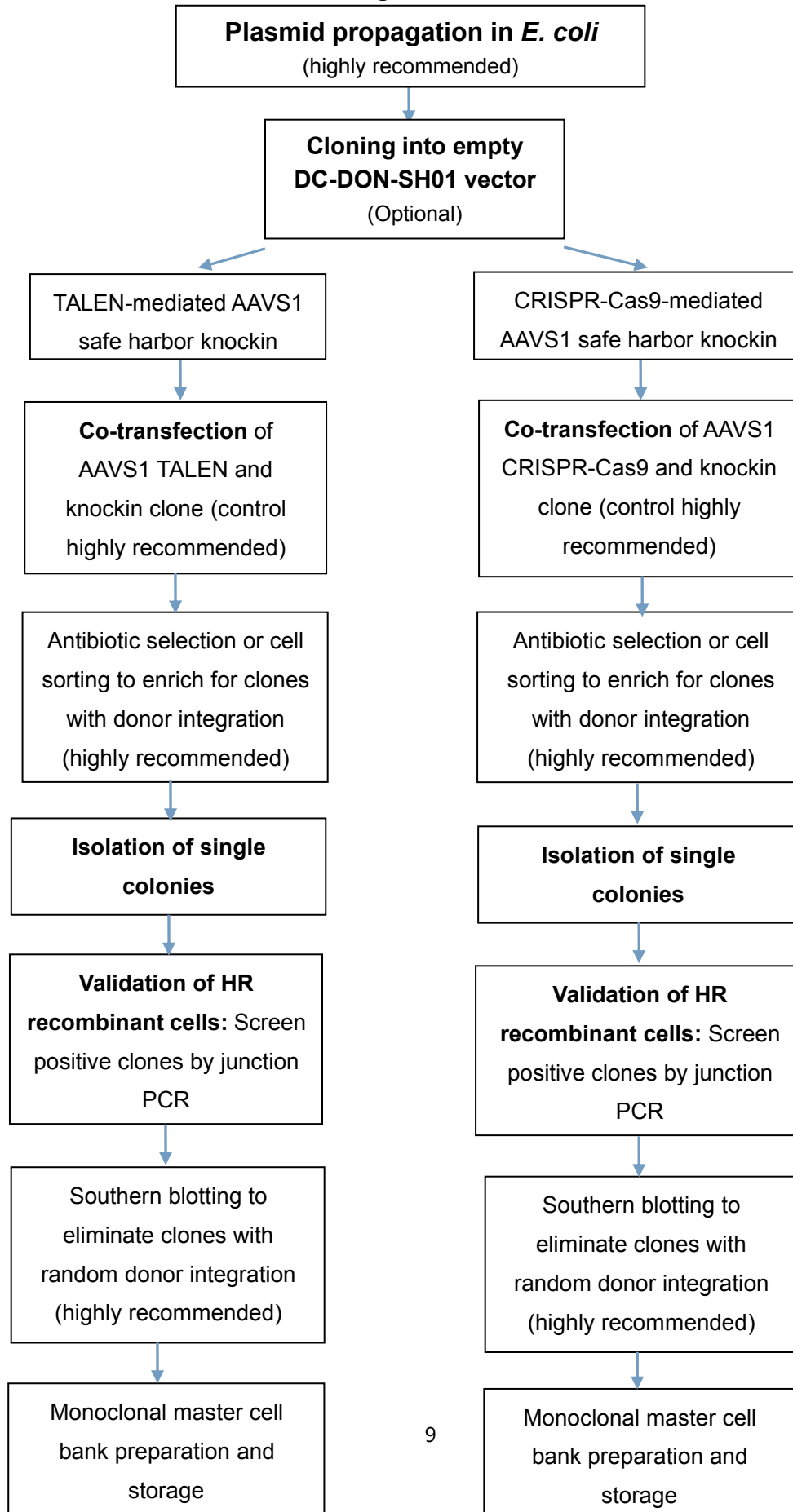
**(A)** AAVS1 RFP control plasmid DC-RFP-SH01 (800 ng) was co-transfected with AAVS1 TALEN pair (600 ng for each) or control TALEN pair into HEK293T cells in a 6-well pate.

**(B)** 48 hr post-transfection, the cells were split 1:10 into a new 6-well pate and incubated in medium containing 1.0  $\mu$ g/ml of puromycin. The images were taken after two weeks of selection.

**(C) (D)** PCR primers designed to amplify the HR junction were used to verify the specific and successful integration.



#### IV. Overview of Safe Harbor Integration



## V. Critical Steps

### A. Plasmid propagation

We recommend propagating the plasmids provided in the safe harbor kit before the gene targeting experiment. Plasmids can be transformed using standard conditions suitable in any RecA- and EndA- E.coli competent cell.

For transformation of kit plasmids, we suggest plating 50-200µl of transformed cells on fresh LB plates with relevant antibiotics. Incubate the plates at 37°C overnight. Inoculate colonies from the transformation and grow them at 37°C overnight in ~200ml of LB media containing relevant antibiotics. Use an endotoxin-free plasmid DNA maxiprep kit to extract plasmid DNA after overnight growth. See the table below for resistance and recommended antibiotic concentration for each plasmid.

To confirm integrity of the amplified plasmids, we recommend restriction digestion analysis or direct sequencing.

Plasmid	Resistance	Recommended antibiotic concentration
TN-AAVS1-L	Kanamycin	50µg/ml
TN-AAVS1-R	Kanamycin	50µg/ml
HCP-AAVS1-CG02	Ampicillin	50µg/ml
DC-DON-SH01	Ampicillin	50µg/ml
DC-RFP-SH01	Ampicillin	50µg/ml
ORF knock-in clones in DC-DON-SH01	Ampicillin	50µg/ml

### B. Cloning into empty DC-DON-SH01 vector

#### 1. Ligation

- 1) Digest and gel-purify the vector plasmid. Dilute it to 10ng/µl.
- 2) Set up 10µl ligation reaction for each control and test sample:

Volume	Item
1.0 µl	Digested DC-DON-SH01 empty vector
7.0 µl	DNA insert (~30-50 ng) or water control
1.0 µl	10X T4 DNA ligase buffer
1.0 µl	T4 DNA Ligase (40 U/µl)
<b>10.0 µl</b>	<b>Total Reaction Volume</b>

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- 3) Incubate reactions at 25°C for 1-2 hours (sticky-end ligation) or O/N at 16°C (for blunt-end ligation)

### 2. Transformation

Transform competent cells (transformation efficiency at least  $1 \times 10^9$  colonies/ $\mu$ g pUC19) with the entire ligation reaction (10 $\mu$ l) following the provided protocol of the competent cells. Plate the transformed competent cells on LB-Ampicillin/Carbencillin agar plates.

### 3. Screening correct clones

- 1) Depending on the ratio of colony numbers for the cDNA sample vs. the negative control sample, randomly mark 5 or more well-isolated colonies.
- 2) Prepare a PCR Master Mix with PCR primers flanking the insert:

1 rxn	10 rxn	Composition
0.1 $\mu$ l	1 $\mu$ l	5' PCR primer (10 $\mu$ M)
0.1 $\mu$ l	1 $\mu$ l	3' PCR primer (10 $\mu$ M)
0.2 $\mu$ l	2 $\mu$ l	50X dNTP mix (10 mM of each)
2.5 $\mu$ l	25 $\mu$ l	10X PCR Reaction Buffer
21.9 $\mu$ l	219 $\mu$ l	Nuclease-free water
0.2 $\mu$ l	2 $\mu$ l	Taq DNA polymerase (approx. 5 U/ $\mu$ l)
<b>25<math>\mu</math>l</b>	<b>250<math>\mu</math>l</b>	<b>Total volume</b>

- 3) Mix the master mix very well and aliquot 24 $\mu$ l into each well of 96-well PCR plate or individual tubes.
- 4) Pick each marked colony from step 1) using sterilized tips and mix in each well (or tube).
- 5) Proceed with PCR using the following program:

94°C, 4 min	1 cycle
94°C, 0.5 min, then 68°C, 1 min/1 kb*	25 cycles
68°C, 3 min	1 cycle

\* Depending on the size of final PCR product, use a shorter or longer time.

- 6) Take 5 $\mu$ l of the PCR reaction and run it on a 1.2% agarose/EtBr gel in 1X TAE buffer to identify clones with correct insert.

4. Inoculate a positive colony containing insert in an appropriate amount of LB-Ampicillin/Carbencillin broth. Incubate at 37°C overnight. Extract and purify the construct using an endotoxin-free plasmid purification kit. Sequence verification of the insert is optional.

### C. Co-transfection of AAVS1 genome editing tools and donor plasmid

1. Plate ~100,000 to 300,000 cells/well in a 6-well plate following the recommended conditions for cell type(s) being transfected. Include wells for the following:
  - a) AAVS1 TALENs (or HCP-AAVS1-CG02) + positive control DC-RFP-SH01
  - b) Positive control DC-RFP-SH01 only
  - c) AAVS1 TALENs (or HCP-AAVS1-CG02) + donor in vector DC-DON-SH01
  - d) Donor in vector DC-DON-SH01 only

On the day before transfection, trypsinize and count the cells. The number of cells plated in each well should be determined so that they are 70-80% confluent at the time of transfection.

2. The next day, prepare transfection complexes of genome editing tool plasmids and donor plasmids using suitable transfection reagents according to the manufacturer's instructions. Leave the transfection complexes on the cells to react for >6 hours.

Example: For HEK293T cells using EndoFectin™ Plus Transfection Reagent, transfect 0.5µg of each TN-AAVS1-L and TN-AAVS1-R (1µg total) and 1µg of donor vector.

#### Tech Notes:

- 1) Since transfection efficiencies vary across different cell lines, we recommend optimizing the input of genome editing tool plasmids to donor vectors for best results. We recommend starting with a 1:1 ratio (e.g. 1µg of donor HR plasmid, 0.5µg of each TALEN plasmid, or 1µg of HCP-AAVS1-CG02 plasmid).
- 2) For optimal results, we recommend complexing DNA with transfection reagent in serum- and antibiotic-free media and cells growing in complete media (e.g. DMEM/F12+10% FBS w/o antibiotics).
- 3) For hard-to-transfect cells (e.g. primary, stem, hematopoietic), it may be advisable to utilize a non-passive transfection method. Please follow recommended guidelines provided by the manufacturer for the specific cell type(s) being transfected.
3. 24 hours post-transfection, remove transfection media and split the cells 1:10 and 1:20 in complete growth media w/antibiotics. Plate cells into 6-well plates and save a set of plate(s) for characterization of samples by junction-PCR assay (see below). Allow cells to recover for 24 hours.
4. Begin puromycin selection 48 hours post-transfection. For 293T cells, the recommended concentration of puromycin is 1 µg/ml.

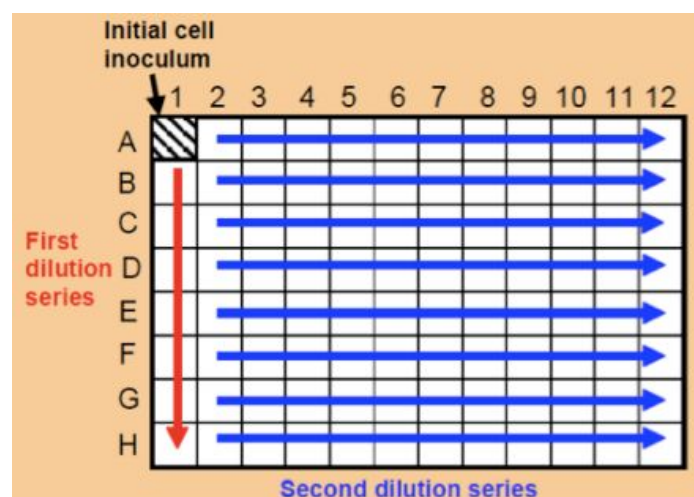
#### Tech Note:

Establishing a kill-curve on untransfected cells can determine the effective working puromycin concentration for a target cell line. The concentration of puromycin (typical working range of 0.5µg-5µg/ml) that kills >90% of cells after 48hours of selection is the correct dose for the cells being selected.

## D. Clonal isolation of cell lines

Serial dilution is widely used to isolate single clones with desired modifications, followed by an expansion period to establish a new clonal cell line. Like most clonal isolation methods, there is no guarantee that the colonies arose from single cells. A second round is advised to increase the likelihood of clonal isolation. Also, it is worth noting that cell types can vary substantially in their responses to single-cell isolation, therefore literature specific to the cell type of interest should be consulted.

1. Fill each well of a sterile 96-well plate with 100µl of medium except for well A1, which should remain empty.



**Figure 5:** Illustration of serial dilution.

2. Add 200µl cell suspension to well A1. Mix 100µl from A1 with the medium in well B1. Avoid bubbles. Continue this 1:2 dilution through column 1. Add 100µl of medium back to column 1 so that wells A1 through H1 contain 200µl.
3. Mix cells and transfer 100µl of cells from column 1 into column 2. Mix by gently pipetting. Avoid bubbles. Repeat these 1:2 dilutions through the entire plate. Bring the final volume to 200µl by adding 100µl of medium to all but the last column of wells.
4. Incubate plates undisturbed at 37°C.
5. Cells will be observable via microscopy over 3 days and be ready to score in 5-8 days, depending on the growth rate of cells. Mark each well on the cover of the plate indicating which well contains a single colony. These colonies can later be subcultured from the well into larger vessels.

### Tech Note:

- 1) Adding 4000 cells in well A1 ( $2 \times 10^4$  cells/ml) is a good starting concentration. Increase the concentration for more difficult to grow cell lines.
- 2) If the reporter gene is fluorescent, determine which of these colonies express it. If the reporter gene is not observable you will have to wait until later in the culture process.
- 3) Label each well with a single colony using a unique identification number and record this number on the plate and in your notebook.

## E. Validation of HR recombinant cells

1. Assay for genome editing tools cutting and HR of donor vectors on samples as follows:
  - 1) AAVS1 TALENs (or HCP-AAVS1-CG02) + positive control DC-RFP-SH01:  
Select cells in Puromycin for 7-10 days. The resulting colonies should be RFP & GFP positive.
  - 2) Positive control DC-RFP-SH01 only:  
  
Select cells in Puromycin for 7-10 days, after which very few colonies (if any) should be seen compared with Sample a. The presence of PuroR, RFP/GFP+ colonies indicates random integration events.
  - 3) AAVS1 TALENs (or HCP-AAVS1-CG02) + donor in vector DC-DON-SH01  
  
Select cells in Puromycin for 7-10 days, after which colonies should be GFP positive. Expression of the insert may be detected by qPCR or Western blot.
  - 4) Donor in vector DC-DON-SH01 only  
  
Select cells in Puromycin for 7-10 days, after which very few colonies (if any) should be seen compared with Sample c. The presence of PuroR, GFP+ colonies indicates random integration events.
2. To confirm donor vector integration specifically at the AAVS1 target locus, junction-PCR can be performed using PCR primer pairs that flank the 5' AAVS1 homology arm (5' AAVS1-HA-L) and 3' AAVS1 homology arm (3' AAVS1-HA-R).
3. Protocol for junction-PCR
  - 1) Primer sequences

Primer description	Primer name	Primer sequence
5' AAVS1 Positive Control Donor - Forward Primer	HQPAVSHR-5F	See datasheet
5' AAVS1 Positive Control Donor - Reverse Primer	HQPAVSHR-5R	See datasheet
3' AAVS1 Positive Control Donor - Forward Primer	HQPAVSHR-3F	See datasheet
3' AAVS1 Positive Control Donor - Reverse Primer	HQPAVSHR-3R	See datasheet

The primers are provided as mixes (F/R primers) at 10 $\mu$ M. Validation of either the 5' or 3' homology arms for donor integration is usually sufficient; however, both arms can be done for additional confirmation.

- 2) Protocol details for junction-PCR assay:
  - a) Isolate genomic DNA from positive control cells or test sample cells using a suitable genomic DNA miniprep kit. Please follow the protocol recommended by the

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manufacturer.

b) Perform junction-PCR (PCR reaction below)

Reagent	TALEN or CRISPR-Cas9 cut + positive control donor	Positive control donor only
Genomic DNA(60~100ng/ $\mu$ l)	1 $\mu$ l	1 $\mu$ l
10 $\mu$ M 5' (or 3') AAVS1 PCR Primer Mix	1 $\mu$ l	1 $\mu$ l
5 $\times$ UltraPF™ Buffer (Mg <sup>2+</sup> free)	5 $\mu$ l	5 $\mu$ l
10 mM dNTPs	0.5 $\mu$ l	0.5 $\mu$ l
20mM MgSO <sub>4</sub>	2.5 $\mu$ l	2.5 $\mu$ l
UltraPF(5U/ $\mu$ l)	0.25 $\mu$ l	0.25 $\mu$ l
PCR-grade distilled water	14.75 $\mu$ l	14.75 $\mu$ l
<b>Total</b>	<b>25<math>\mu</math>l</b>	<b>25<math>\mu</math>l</b>

98°C, 5min

98°C, 20sec

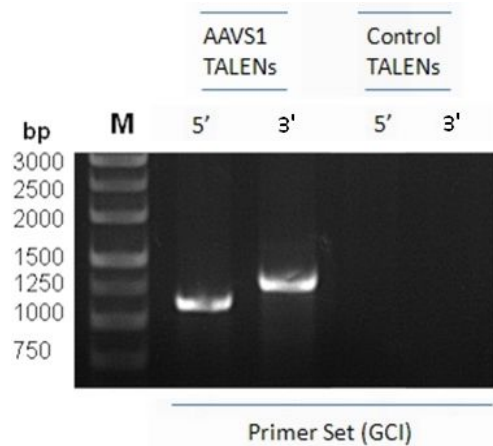
55°C, 30sec

72°C, 1min

72°C, 7min

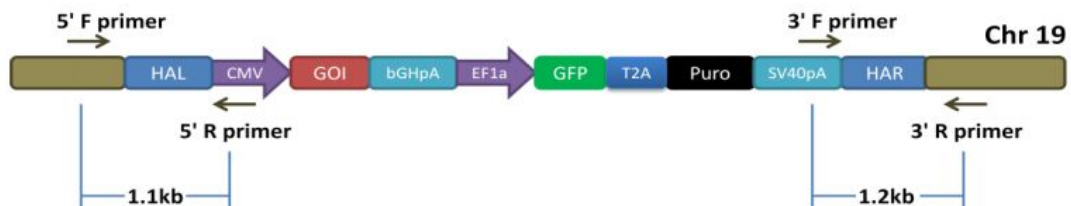
Hold at 4~16°C

35 cycles



Run the PCR reaction on a 1% Agarose/EtBr gel in 1X TAE buffer to confirm the junction-PCR result.

Sample results for 5' and 3' junction-PCR assay shown below:



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### Tech Note:

- 1) If the 3'junction PCR band is weaker than 5' junction PCR band, it is likely that the amplification efficiency for the 3'junction region is lower due to the nature of the chromosomal structure, modification and sequence around that region.
- 2) One positive in junction PCR is sufficient to confirm the integration.
- 3) It is possible that random integration can coexist with AAVS1-specific integration. Southern blotting can be used to detect coexisting random integration. The method is described in: <http://bloodjournal.hematologylibrary.org/content/117/21/5561.full.html>

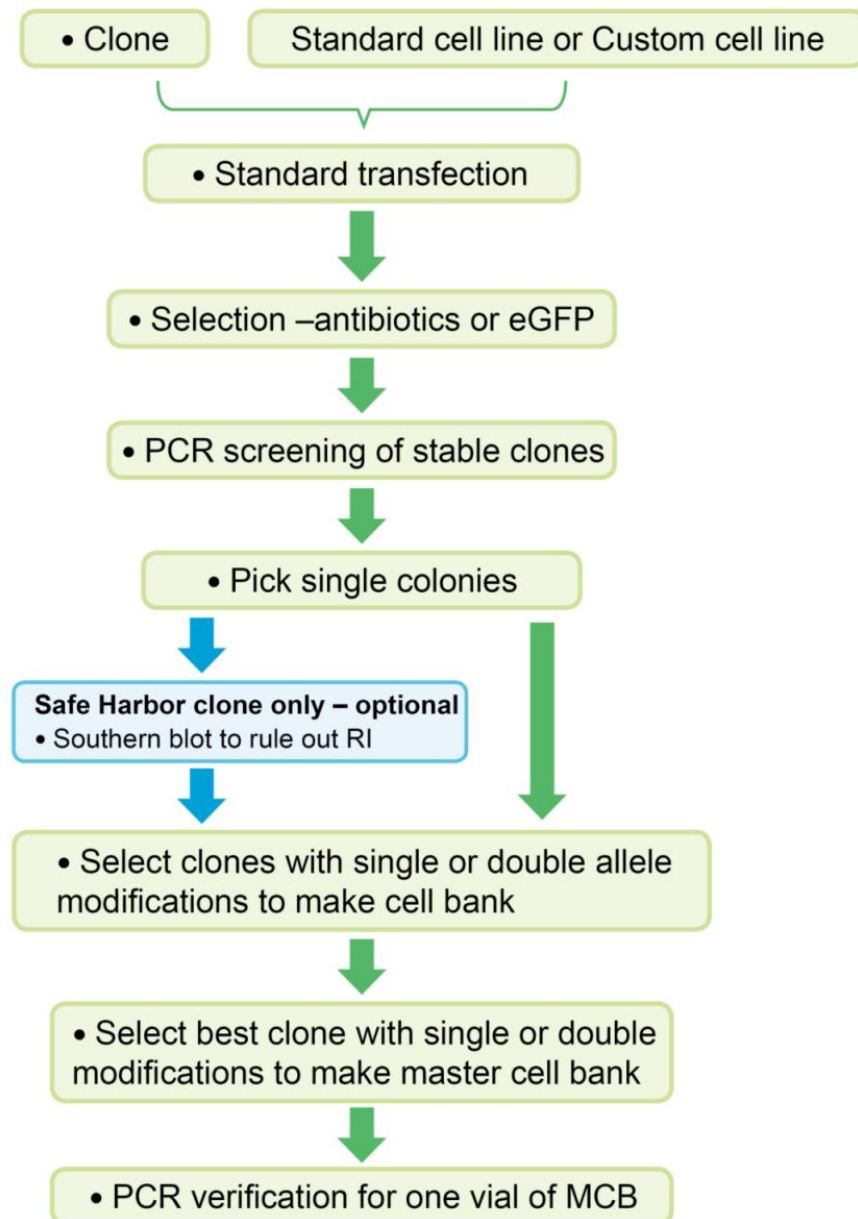


## VII. Related Services

### Stable cell line services

GeneCopoeia offers monoclonal stable cell line service with customized TALEN- or CRISPR-mediated genome modifications. Cell banking service is also available.

### TALEN/CRISPR Stable Cell Line Development Services



## VII. References

1. Zou, J. et al. 2009. Gene targeting of a disease-related gene in human induced pluripotent stem and embryonic stem cells. *Cell Stem Cell*. 2009 Jul 2;5(1):97-110
2. Sadelain, M. et al. 2011. Safe harbours for the integration of new DNA in the human genome. *Nat Rev Cancer*. 2011 Dec 1;12(1):51-8.
3. van Rensburg, R. et al. 2013. Chromatin structure of two genomic sites for targeted transgene integration in induced pluripotent stem cells and hepatopoietic stem cells. *Gene Therapy*. 2013 20(2):201-14.
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## VI. Limited Use License and Warranty

### Limited Use License

Following terms and conditions apply to use of the Genome-TALER™ Human AAVS1 Safe Harbor Gene Knock-in Kit & Genome-CRISP™ Human AAVS1 Safe Harbor Gene Knock-in Kit (the Product). If the terms and conditions are not acceptable, the Product in its entirety must be returned to GeneCopoeia within 5 calendar days. A limited End-User license is granted to the purchaser of the Product. The Product shall be used by the purchaser for internal research purposes only. The Product is expressly not designed, intended, or warranted for use in humans or for therapeutic or diagnostic use. The Product must not be resold, repackaged or modified for resale, or used to manufacture commercial products or deliver information obtained in service without prior written consent from GeneCopoeia. This Product should be used in accordance with the NIH guidelines developed for recombinant DNA and genetic research. Use of any part of the Product constitutes acceptance of the above terms.

### Limited Warranty

GeneCopoeia warrants that the Product meets the specifications described in the accompanying Product Datasheet. If it is proven to the satisfaction of GeneCopoeia that the Product fails to meet these specifications, GeneCopoeia will replace the Product. In the event a replacement cannot be provided, GeneCopoeia will provide the purchaser with a refund. This limited warranty shall not extend to anyone other than the original purchaser of the Product. Notice of nonconforming products must be made to GeneCopoeia within 30 days of receipt of the Product. GeneCopoeia's liability is expressly limited to replacement of Product or a refund limited to the actual purchase price. GeneCopoeia's liability does not extend to any damages arising from use or improper use of the Product, or losses associated with the use of additional materials or reagents. This limited warranty is the sole and exclusive warranty. GeneCopoeia does not provide any other warranties of any kind, expressed or implied, including the merchantability or fitness of the Product for a particular purpose.

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