GeneCopoeia™
Expressway to Discovery

Premade Cell Line Models

New stable cell lines for your biomedical research

GeneHero™ Cas9 Stable Cell Lines

✓ Study loss or gain of gene function

✓ sgRNA library screening

✓ Compound screening

OncoSpot™ Cancer Biomarker Mutant Cell Lines

✓ Drug discovery

✓ Compound screening

✓ Monitor cancer signaling pathway activity

✓ Reference standard NGS gene mutation detection

Reporter-Gene Labeled Cancer Cell Lines

✓ Cancer cell lines labeled with luciferase and GFP

✓ Monitor tumor cells growth and migration *in vivo*

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GeneHero™ Cas9 Stable Cell Lines

GeneCopoeia's GeneHero™ Cas9 stable cell lines constitutively expressing the CRISPR Cas9 nuclease enable you to carry out CRISPR genome editing applications with high efficiency. The Cas9 stable cell lines are available premade in many human and mouse cell line backgrounds.

Available cell types

More than 70 human, mouse and rat cell lines, including H1299, HEK293, MCF-7 and Neuro2a.

Available cancer types

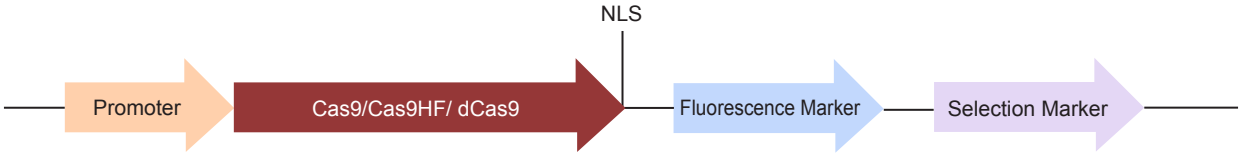
More than 26 different cancers, including lung, cervical, breast, liver, and others.

Advantages:

- ✓ Stable Cas9 at safe-harbor site or randomly integration into cell genome ensures robust and consistent expression of the CRISPR cas9 nuclease
- ✓ Cas9 functionally validated by using the IndelCheck™ T7 Endonuclease I assay
- ✓ Minimizes the need for co-transfection or co-transduction of Cas9 with sgRNAs
- ✓ Validation of sgRNA cleavage activity in a fast-growing, easy-to-transfect or transduce model cell line, either prior to transfection/transduction of your cell line, or to troubleshoot sgRNAs with little or no cleavage activity

Applications:

- ✓ Convenient validation of multiple drug target candidates/discovery
- ✓ Applications requiring genome modification
- ✓ Ideal for high-throughput sgRNA knockout screening application



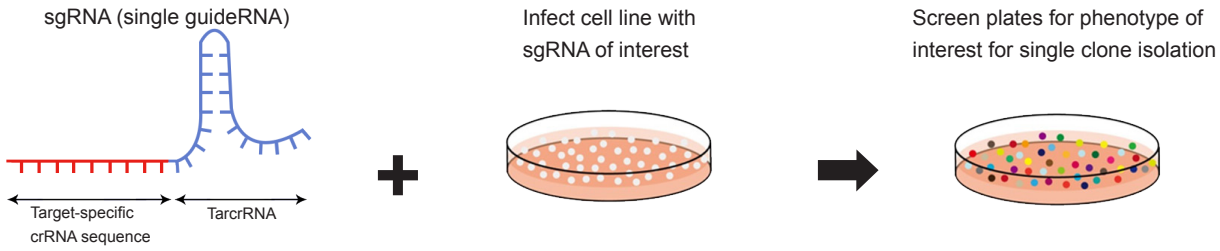
Structure of a stably integrated CRISPR Cas9 nuclease construct. Hygromycin or Puromycin can be used for selection for isolation of single clones. CMV or CBh: Hybrid CBA promoter available. GFP or RFP: Fluorescence Marker available. NLS: Nuclear localization signal.

GeneHero™ Cas9 Stable Cell Lines

The CRISPR Cas9 nuclease, in combination with sgRNAs, is widely used to create targeted genomic modifications, gene knockouts, mutagenesis, fusion tagging, and more, in eukaryotic cells and animal models.

Our CRISPR Cas9 stable cell lines make genome editing faster and easier than before. Simply deliver the target specific gRNA into the GeneHero™ Cas9 stable cell line and select for your desired KO or KI target. Single clone isolation provides consistent, high-level Cas9 expression in a uniform genetic background.

Figure 1.



Testimonial

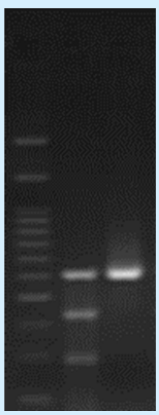
GeneCopoeia's Hela and A549 Cas9 stable cell lines (SL503, SL504) were used in the research listed below for a systematic study of the dispensability and interaction of genes underlying carbohydrate metabolism. Dual-sgRNA libraries were screened in arrays for the desired phenotypes on cellula growth and metabolic fluxes.

"We generated three single guide RNAs (sgRNAs) per gene such that nine unique constructs were present for every gene pair, resulting in a dual-sgRNA library consisting of 459 elements targeting genes individually, as well as 11,475 unique elements targeting two different genes simultaneously. The dual-sgRNA constructs were synthesized from oligonucleotide arrays, cloned into a lentiviral vector, and then transduced into HeLa or A549 cells stably expressing Cas9."

- Molecular Cell 69(4):699-708, 2018

Cas9 expression within these stable cell lilnes exhibits high activity, as shown by the T7 Endonuclease I mismatch cleavage assay.

Figure 2. Human H1299 cells stably expressing Cas9 nuclease from the AAVS1 Safe Harbor site were transfected with a plasmid carrying an sgRNA for the HUWE I gene. Using the GeneCopoeia IndelCheck™ kit, PCR products flanking the sgRNA target site were generated, denatured, reannealed, and treated with (+) or without (-) T7 Endonuclease I, which cleaves mismatched DNA.



GeneHero™ Cas9 Stable Cell Lines

Cell Type	Cell Name	Select Marker
Bladder cancer, Human	T24	Hygro
Blood cancer, Human	Granta-519	Hygro
Bone cancer, Human	U-2 OS	Hygro
Bone marrow cancer, Human	K562	Hygro
Brain astrocytoma, Human	1321N1	Hygro
Breast cancer, Human	AU-565	Hygro
Breast cancer, Human	CAMA-1	Hygro
Breast cancer, Human	DU4475	Hygro
Breast cancer, Human	MCF-7	Hygro
Breast cancer, Human	MDA-MB-231	Hygro
Breast cancer, Human	MDA-MB-468	Hygro
Breast cancer, Human	T47D	Hygro
Breast cancer, Human	SK-BR-3	Hygro
Breast cancer, Human	HCC38	Hygro
Breast cancer, Human	HCC70	Hygro
Breast cancer, Human	HCC1500	Hygro
Breast cancer, Human	HCC1428	Hygro
Caecum cancer, Human	LS411N	Hygro
Cervical cancer, Human	HeLa	Hygro
Colon cancer, Human	COLO 205	Hygro
Colon cancer, Human	HT-29	Hygro
Colon cancer, Human	HCT116	Hygro
Colon cancer, Human	LoVo	Hygro
Colon cancer, Human	RKO	Hygro
Colon cancer, Human	T84	Hygro
Colon cancer, Human	SNU-C1	Hygro
Fibrosarcoma, Human	HT1080	Hygro
Embryonic kidney, Human	HEK293	Hygro
Embryonic kidney, Human	HEK293T	Puro
Immortalized kidney cell line, Human	HK2	Hygro
Kidney cancer, Human	786-O	Hygro
Liver cancer, Human	HepG2	Hygro
Liver cancer, Human	PLC/PRF/5	Hygro
Liver cancer, Human	SNU-387	Hygro

Cell Type	Cell Name	Select Marker
Liver cancer, Human	SNU-423	Hygro
Liver cancer, Human	SNU-449	Hygro
Liver cancer, Human	SNU-475	Hygro
Liver/ascites cancer, Human	SK-HEP-1	Hygro
Liver cancer, Human	A549	Hygro
Liver cancer, Human	H1975	Hygro
Lung cancer, Human	NCI-H1299	Hygro
Lung cancer, Human	NCI-H1299	Puro
Lung cancer, Human	NCI-H1437	Hygro
Lung cancer, Human	NCI-H661	Hygro
Lung cancer, Human	HCC827	Hygro
Neuroblastoma, Human	SH-SY5Y	Hygro
Pancreatic cancer, Human	BXPC-3	Hygro
Pancreatic cancer, Human	CFPAC-1	Hygro
Pancreatic cancer, Human	HPAF-II	Hygro
Acute T cell leukemia, Human	Jurkat	Hygro
Ovary, Human	SK-OV-3	Hygro
Prostate cancer, Human	DU145	Hygro
Pancreatic cancer, Huma	Panc 10.05	Hygro
Stomach/gastric cancer, Human	AGS	Hygro
Stomach/gastric cancer, Human	KATOIII	Hygro
Stomach/gastric cancer, Human	NCI-N87	Hygro
Stomach/gastric cancer, Human	SNU-1	Hygro
Stomach/gastric cancer, Human	SNU-16	Hygro
Colon, Mouse	CT26	Hygro
Embryo fibroblast, Mouse	NIH-3T3	Hygro
Lymph, Mouse	BA/F3	Hygro
Mammary gland, Mouse	4T1	Hygro
Muscle (myoblast), Mouse	C2C12	Hygro
Neuroblastoma, Mouse	Neuro2a	Hygro
Neuroblastoma, Mouse	Neuro2a	Puro
Neuroblastoma, Mouse	Neuro2a	Neo
Glioma, Rat	C6	Hygro

Visit our website
www.genecopoeia.com/product/cas9-cell-line
to see a complete list.

Reporter-Gene Labeled Cancer Cell Lines

GeneCopoeia offers premade single and dual reporter-genes labeled stable cancer cell lines for both *in vivo* and *in vitro* experiments. The dual-labeled cancer cell lines are labeled with luciferase and GFP while the single-labeled cancer cell lines are labeled with GFP only. Choose from among pre-made lines from various tumor types, including breast, liver, pancreas, and colon.

Advantages:

- ✓ Robust luciferase expression permits highly sensitive, non-invasive detection of cancer cell growth *in vivo*
- ✓ Tumor monitoring *in vivo* without the need for substrate perfusion or invasive surgery
- ✓ *In vitro* visualization and immunocytochemistry

Applications:

- ✓ Useful for monitoring various biological functions for sensitive studies *in vivo*
- ✓ Study tumor cell properties by using biochemical biophysical and genetic methods *in vitro*

For more information about GeneCopoeia's Mammalian Stable Cell Line Development Services or premade labeled cancer cell lines, contact us at inquiry@genecopoeia.com.

Selected Cell Lines

Detection Marker: eGFP									
Cat No.	SL101	SL102	SL103	SL106	SL114	SL115	SL116	SL117	SL118
Cancer Cell Lines	NCI-H1299	NCI-H661	NCI-H1975	HT-29	NCI-N87	AGS	HCC70	MCF7	MDA-MB-231
Tissue	Lung	Lung	Lung	Colon	Stomach	Stomach	Breast	Breast	Breast

Detection Marker: Firefly Luciferase and GFP												
Cat No.	SL001	SL002	SL003	SL004	SL005	SL006	SL014	SL015	SL016	SL017	SL018	SL019
Cancer Cell Lines	NCI-H1299	NCI-H661	NCI-H1975	LoVo	COLO 205	HT-29	NCI-N87	AGS	HCC70	MCF7	MDA-MB-231	K562
Tissue	Lung	Lung	Lung	Colon	Colon	Colon	Stomach	Stomach	Breast	Breast	Breast	Bone marrow

Reporter-Gene Labeled Cancer Cell Lines

Luciferase+GFP Dual-Labeled Cancer Cell Lines

Robust luciferase expression permits highly sensitive, non-invasive detection of cancer cell growth and progression *in vivo.*

In addition, GFP expression is useful for tumor monitoring *in vivo* without the need for substrate perfusion, or for visualization and immunocytochemistry *in vitro*.

In a recent paper in Nature Communications, the dual-labeled Luc+ and GFP+ colon cancer cell line from GeneCopoeia was used to explore the effect of physical force on tumor growth, progression and metastasis.

The cells were injected into the tail vein of mice. Upon drug treatment, the dual-labeled cancer cells were isolated from whole blood, and the mode of cell death was assessed using flow cytometry. Whole body bioluminescence imaging (BLI) showed systemic bioluminescence signal distribution throughout mice, and was performed to monitor tumor cell signals *in vivo* at 1 and 2 weeks post injection.

"A dual-labelled luciferase and GFP+ COLO 205 colon cancer cell line was obtained from Genecopoeia (Rockville, MD, USA) and cultured in RPMI-1640 supplemented with 10% (vol/vol) FBS and 1% (vol/vol) PenStrep."

- Nature Communications volume 8, Article number: 14179, 2017

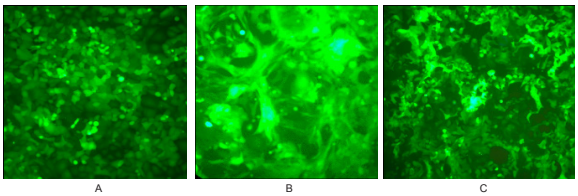


Figure 1. Dual-labeled cancer cell line expressing GFP
GFP fluorescence after exposure for 100 mS at 95% cell confluence using a Nikon Fluorescence Microscope
A: SL001 (NCI-H1299 Lung Cancer Cell Line)
B: SL003 (NCI-H1975 Lung Cancer Cell Line)
C: SL017 (MCF7 Breast Cancer Cell Line)

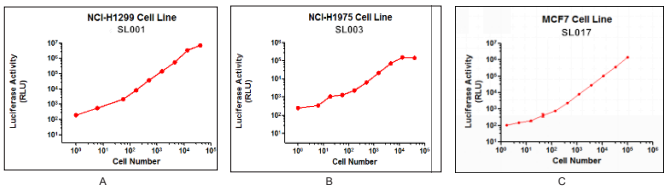


Figure 2. Luciferase activity of dual-labeled cancer cell lines
A: SL001 (NCI-H1299 Lung Cancer Cell Line)
B: SL003 (NCI-H1975 Lung Cancer Cell Line)
C: SL017 (MCF7 Breast Cancer Cell Line)

GFP Labeled Cancer Cell Lines

GeneCopoeia's pre-made GFP-labeled cancer cell lines are useful for *in vivo* tumor monitoring without the need for substrate perfusion, or for *in vitro* visualization and immunocytochemistry.

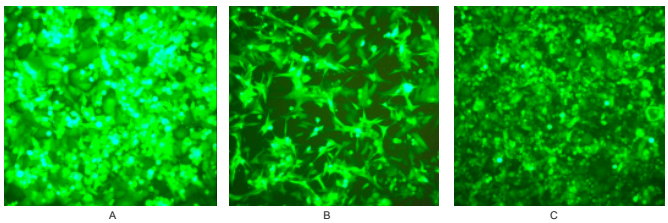


Figure 3. GFP labeled cancer cell lines
GFP fluorescence after exposure for 100 ms
A: SL101 (NCI-H1299 Lung Cancer Cell Line)
B: SL103 (NCI-H1975 Lung Cancer Cell Line)
C: SL116 (HCC70 Breast Cancer Cell Line)

OncoSpot™ Cancer Biomarker Mutant Cell Lines

GeneCopoeia offers genome-edited cell lines that carry diverse hotspot mutations commonly seen in the cancer signaling pathways including EGFR, KRAS and BRAF, in both homozygous and heterozygous forms.

Advantages:

- ✓ Premade and ready to ship
- ✓ Parental cell line included
- ✓ Validated by PCR and Sanger sequencing
- ✓ Precisely gene edited by CRISPR technology
- ✓ Mutations available in both homozygous and heterozygous forms

Custom Cell Line Services

Looking for a different cancer gene of interest?

We can help you design a cutom cell line.

For more information, contact inquiry@genecopoeia.com

Applications:

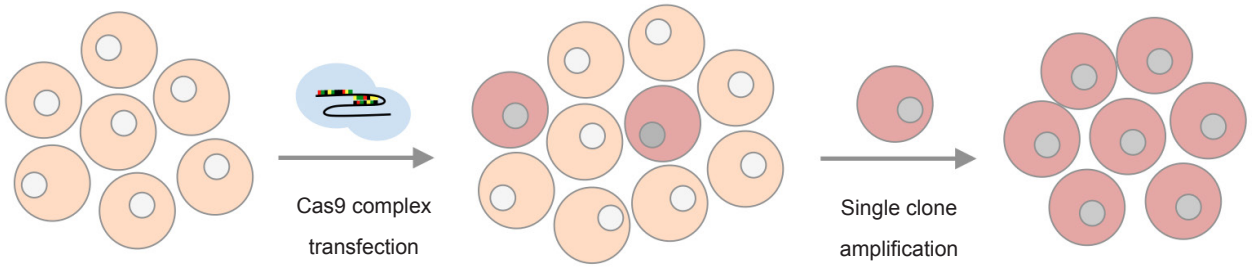
These mutations are highly relevant to diseases and drug targets and suited for drug screening applications.

- ✓ Study molecular and cellular mechanisms
- ✓ Obtain functional characterization
- ✓ As cell line models for the study of metabolic and signaling pathways
- ✓ As *in vitro* models for drug screening and toxicity studies
- ✓ To enhance our understanding of cancer biology and the development of cancer therapies.
- ✓ As reference standard for gene mutation detection by NGS and Fish.

Development of Genome-edited Cell Lines

We started out by using CRISPR to introduce MAPK pathway disease-relevant point mutations or frameshift indel mutations into the HCT116 colon cancer cell line. These mutations are integrated either homozygously or heterozygously.

Figure 1. Cell line construction using CRISPR.



OncoSpot™ Cancer Biomarker Mutant Cell Lines

Single clonal isolation was performed for sequence verification, and then expansion. We provide the original isogenic wild type cell line which minimizes background genetic variation and provides you with greater confidence that any differences observed between the mutant and the parental cell line are due to your specific mutation of interest. All of our cell lines undergo extensive QC and validation.

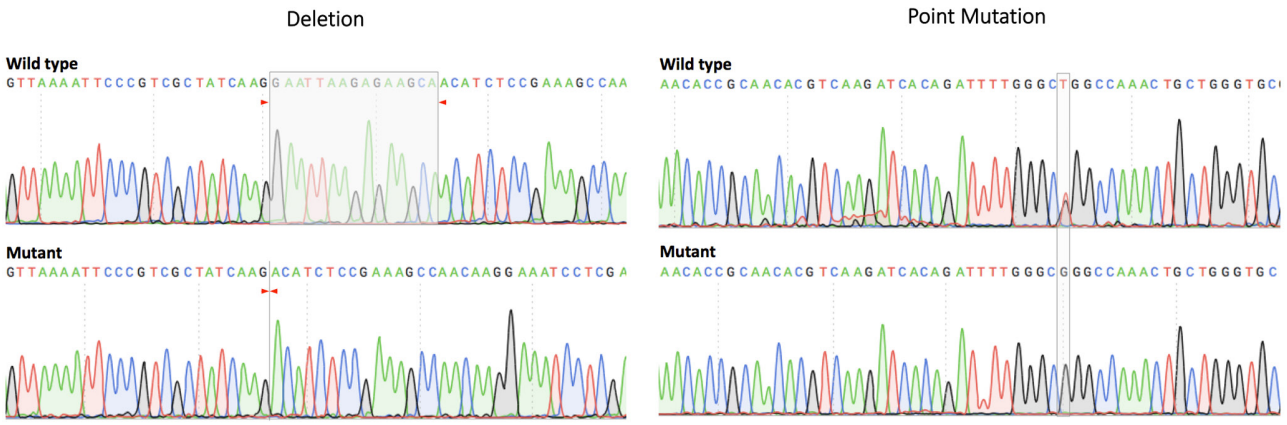


Figure 2. Sequence verification of a single cell clone using PCR and Sanger sequencing. Comparison of the sequencing chromatograms of EGFR ΔE746-A750 cell line (left) and EGFR L858R cell line (right) to wild type. The deleted sequence in EGFR ΔE746-A750 and point mutation in EGFR L858R were marked in grey boxes in the chromatograms.

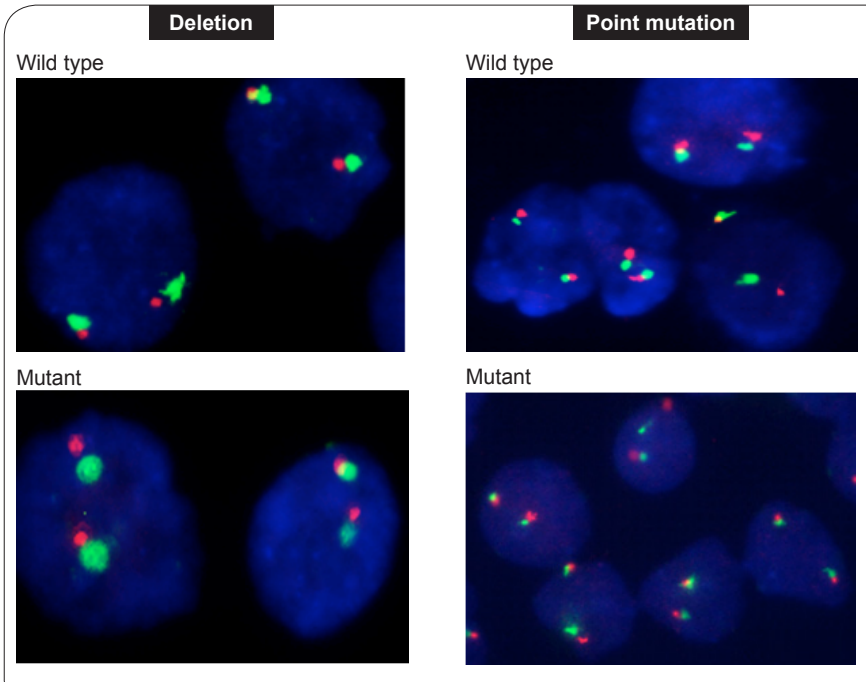


Figure 3. FISH detection of EGFR in wild type (WT) and mutaiton cell lines. GeneCopoeia VividFISH™ FISH probes CEP7/EGFR were hybridized in WT and EGFR ΔE746-A750 cell line (left) and EGFR L858R cell line (right). The green fluorescence indicates centromeres on chromosome 7. Orange fluorescence indicates the EGFR genes on chromosome 7. The cell nuclei are counter-stained with DAPI (blue).