



CRISPR ready, Cas9 expression / A549 stable cell line manual

Catalog Number	Product Name	Amount
SC043-Cas9-Puro	A549 / Cas9 (Puro) Stable cell line	1 vial of cells ($>2 \times 10^6$ cells) in 80% medium, 10% FBS, 10% DMSO
SC043-Cas9-GP	A549 / Cas9 (GFP-Puro) Stable cell line	
SC043-Cas9-RP	A549 / Cas9 (RFP-Puro) Stable cell line	

Storage: liquid Nitrogen.

Product Description

The **A549** cell line is originated from human adenocarcinomic, alveolar basal epithelial cells. The A549 cell line was first developed in 1972 by D. J. Giard, et al. through explant culture of lung carcinomatous tissue from a 58-year-old Caucasian male. These cells grow as adhesive monolayer cells in culture flash.

CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats) is the advanced genomic gene editing technology. A target's sequence specific guide RNA molecule (gRNA) directs a cas endonuclease to the genomic DNA target sequence. Then, the Cas enzyme creates a double-strand break at the target sequence that can be repaired by either Non-Homologous End-Joining (NHEJ), which can result in insertion or deletions (InDels), or correction / Homology Directed Repair (HDR). InDels can disrupt expression of the target gene while repair by HDR, which requires the presence of a repair template, allows modification of the gene.

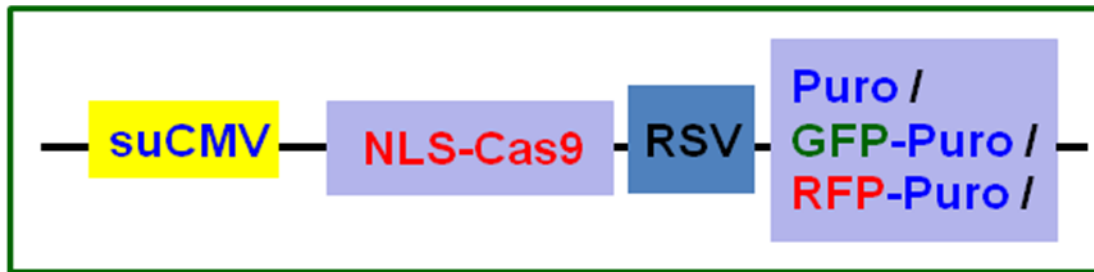
Cas9 is the most frequently used cas endonuclease so far. GenTarget's CRISPR ready cell lines are transformed from lentivirus transduction, expressing a standalone cas9 enzyme. Each cell line expresses the nuclear penetrating humanized wild-type Cas9 endonuclease. The Cas9 enzyme is driven by our engineered super strong CMV promoter (suCMV). Each cell line also contains an **Puromycin** antibiotic selection marker or an **fluorescent-Puromycin** fusion dual marker (**GFP**-puromycin or **RFP**-Puromycin) under the RSV promoter.

The **CRISPR ready (Cas9 expression) stable cell lines** make the genomic editing easier than ever. You simply deliver the target specific gRNA into the CRISPR ready cell line and select the knock-out or knock-in cells for your desired target. No need to do cas9 containing CRISPR cloning, and no worry about the



hard-to-delivered Cas9 constructs. You only need to generate the much easier in cloning, and much smaller in size of the target-gRNA constructs. GenTarget also provide services to generate your target specific, ready-to-use gRNA lentivirus. Please [contact us](#) if you need the CRISPR gRNA lentivirus services.

Please see the structure of the **expression cassette** (below) that integrated into each cell genome (note: the cell line cannot be guaranteed originated from a single cell colony and multiple expression cassettes may exist in some cells).



Culture procedures

1. Thaw the frozen vial of cells quickly in a 37°C water bath (1~3min), decontaminate the outside of the vial with 70% ethanol.
2. Transfer the entire contents of the cryovial into a T-75 cm² flask containing 20 ml of pre-warmed complete medium. Incubate the cells overnight in a 37°C incubator, 5% CO₂.
3. On the following day, replace the medium with 20 ml of prewarmed, complete medium.
4. Incubate the cells and monitor cell density.
5. Pass cells (1:5 to 1:10 dilution) using 0.25% Trypsin-EDTA solution when the culture reaches ~90% confluent.
6. Freeze cells at a density of ~3 x 10⁶ cells/ml using 90% complete medium with 10% DMSO.

Complete medium

F12K medium
2mM L-glutamine
10% Fetal Bovine Serum (FBS)
1% Pen-strep

Option to add: add final concentration of Puromycin at: **0.6 ug/ml** (Note: do not add antibiotic at 1st time cell thaw).



Quality Control

Each vial contains greater than 2×10^6 cells with >95% viability before freezing. Cells are validated as free of bacteria, viruses, and mycoplasma.

Warranty and user terms

1. This product is warranted to perform as described when used in accordance with this manual. GenTarget, Inc. MAKES NO REPRESENTATIONS AND EXTENDS NO WARRANTIES OF ANY KIND, EITHER EXPRESS OR IMPLIED. GenTarget's sole remedy for breach of warranty should be, at the option of GenTarget, to repair or replace the product if this product does not meet the stated quality standard.
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5. GenTarget is not liable, and does not have any responsibility or liability, whatsoever for any direct and indirect, consequential, or other damages resulting from using this Product.
6. Gentarget **do not** provide the protected reporter's sequences information for all our cell line products unless a license was purchased.

Attachment: GenTarget' s pre-made stable cell line list:

Catalog #	Product Name
SC001	HEK293-GFP stable cells
SC002-Bsd	luciferase (firefly), HEK293 stable cells (Blasticidin)
SC002-GB	luciferase (firefly), HEK293 stable cells (GFP-Blasticidin)



SC002-GP	luciferase (firefly), HEK293 stable cells (GFP-Puromycin)
SC002-Neo	luciferase (firefly), HEK293 stable cells (Neomycin)
SC002-Puro	luciferase (firefly), HEK293 stable cells (Puromycin)
SC002-RB	luciferase (firefly), HEK293 stable cells (RFP, Blasticidin)
SC002-RP	luciferase (firefly), HEK293 stable cells (RFP-Puromycin)
SC002T-RP	HEK293T / Luciferase stable cells (RFP-Puromycin)
SC003	LacZ (6His, RFP) / HEK293 Expression stable cell line
SC004-Bsd	CRE Expression / HEK293 Cell Line (Bsd)
SC004-GP	CRE Expression / HEK293 Cell Line (GFP, Puro)
SC004-Neo	CRE Expression / HEK293 Cell Line (Neo)
SC004-Puro	CRE Expression / HEK293 Cell Line (Puro)
SC004-RB	CRE Expression / HEK293 Cell Line (RFP, Bsd)
SC004-RP	CRE Expression / HEK293 Cell Line (RFP, Puro)
SC005-Bsd	HEK293-TetR (Bsd)
SC005-GB	HEK293-TetR (GFP-Bsd)
SC005-Hygro	HEK293-TetR (Hygro)
SC005-Neo	HEK293-TetR (Neo)
SC005-Puro	HEK293-TetR (Puro)
SC005-RB	HEK293-TetR (RFP-Bsd)
SC005-RP	HEK293-TetR (RFP-Puro)
SC006	Flp recombinase Expression HEK293 stable cell
SC007	HEK293-RFP stable cells
SC008	GFP-LacZ & RFP Expression HEK293 Cell Line
SC009	GFP & RFP / HEK293 stable cells
SC010	HEK293-CFP stable cells
SC011	HEK293-YFP stable cells



SC012	TAT Expression / HEK293 Cell Line
SC013	Glutamine Synthetase (6His) Expression HEK293 Cell Line
SC014	human P53 Inducible Expression Cell line
SC015	Human OCT3/4 Expression Stable cells
SC016	Human LIN28 Expression stable cells
SC018-Bsd	Color Switch, CRE report cell line: HEK293-loxP-GFP-RFP (Bsd)
SC018-Neo	Color Switch, CRE report cell line: HEK293-loxP-GFP-RFP (Neo)
SC018-Puro	Color Switch, CRE report cell line: HEK293-loxP-GFP-RFP (Puro)
SC020-Puro	luciferase (Renilla), HEK293 stable cells (Puromycin)
SC020-RP	luciferase (Renilla), HEK293 stable cells (RFP-Puromycin)
SC021-GB	Luciferase (firefly) & CRE Expression cell line (GFP-Bsd)
SC021-Puro	Luciferase (firefly) & CRE Expression stable cell line (Puro)
SC021-RP	Luciferase (firefly) & CRE Expression cell line (RFP-Puro)
SC022-RB	HEK293-CFTR cell line with RFP and Blasticidin dual marker
SC023-RB	HEK293-CLCN2 cell line with RFP and Blasticidin dual marker
SC024-RB	HEK293-TRPC3 cell line with RFP and Puromycin dual marker
SC025-RB	HEK293-KCNN4 cell line with RFP and Puromycin dual marker
SC026-RB	HEK293-ATP2B2 cell line with RFP and Puromycin dual marker
SC027-RB	HEK293-TRPV1 cell line with RFP and Puromycin dual marker
SC028	Inducible RFP HEK293 Expression cell line
SC029	inducible RFP HEK293 stable cell line with GFP marker
SC030	inducible GFP HEK293 stable cell line with RFP marker
SC031-Puro	Hela-RFP Expression Cells
SC032-Bsd	Luciferase (firefly) Expression Hela cells (Bsd)
SC032-GB	Luciferase & GFP Expression Hela cells (Bsd)



SC032-GN	Luciferase & GFP Expression Hela cells (Neo)
SC032-GP	Luciferase & GFP Expression Hela cells (Puro)
SC032-Puro	Luciferase (firefly) Expression Hela cells (Puro)
SC032-RB	Luciferase & RFP Expression Hela cells (Bsd)
SC032-RN	Luciferase & RFP Expression Hela cells (Neo)
SC032-RP	Luciferase & RFP Expression Hela cells (Puro)
SC033	Inducible GFP HEK293 stable cell line
SC034-Bsd	Hela-GFP stable cells (Blasticidin)
SC034-Puro	Hela-GFP stable cells (Puromycin)
SC035-Puro	TetR Expression (Puro) / Hela stable cells
SC036	Inducible GFP Expression Hela cell line
SC037	Inducible RFP Expression Hela cell line
SC038-GB	rtTA (GFP-Bsd) / Hela stable cells
SC038-GP	rtTA (GFP-Puro) / Hela stable cells
SC038-RB	rtTA (RFP-Bsd) / Hela stable cells
SC039-Bsd	CHO-GFP stable cells (Blasticidin)
SC039-Puro	CHO-GFP stable cells (Puromycin)
SC040-Bsd	MDA-MB-231 / GFP (Bsd) Stable Cell Line
SC040-Puro	MDA-MB-231 / GFP (Puro) Stable Cell Line
SC040-TetR	MDA-MB-231 / TetR (Puro) stable cells
SC041	MDA-MB-231 / Luciferase-2A-RFP Stable Cell Line
SC042	SH-SY5Y / GFP (Puromycin) stable cell line
SC043-Bsd	A549 / GFP stable cells (Blasticidin)
SC043-Cas9-GP	A549 / Cas9 (GFP-Puro) Stable Cell Line
SC043-Cas9-Puro	A549 / Cas9 (Puro) Stable Cell Line
SC043-Cas9-RP	A549 / Cas9 (RFP-Puro) Stable Cell Line



SC043-LG	A549 / Luciferase-2A-GFP (Puromycin) stable cell line
SC043-Luc	A549 / Luciferase (Puromycin) stable cell line
SC043-TetR	A549 / TetR (Puro) stable cells
SC044	MDA-MB-231 / Luciferase-2A-GFP Stable Cell Line
SC045-Cas9-Bsd	Hela / Cas9 (Bad) Stable Cell Line
SC046	SH-SY5Y / RFP (Puromycin) stable cell line
SC047-GB	RKO / GFP (Blasticidin) Stable Cell Line
SC047-TetR	TetR Expression (Bsd) / RKO stable cells
SC048	Luciferase (Puro) / Jurkat T Cell line
SC049-1	Jurkat T / GFP Stable Cell (EF1a Promoter)
SC049-2	Jurkat T / GFP Stable Cell (Flt1 Promoter)
SC049-3	Jurkat T / GFP Stable Cell (CD43 Promoter)
SC049-4	Jurkat T / GFP Stable Cell (CD68 Promoter)
SC049-5	Jurkat T / GFP Stable Cell (Survivin Promoter)
SC050-G	MCF7 / GFP (Puromycin) Cell Line
SC050-L	MCF7 / Firefly Luciferase (Puro) Cell Line
SC051-G	ZR-75-1 / GFP (Puromycin) Cell Line
SC051-L	ZR-75-1 / Firefly Luciferase (Puro) Cell Line
SC053-L	NCI-H1299 / Luciferase (Puro) Stable Cells
SC054-L	CFPAC-1 / Luciferase (Puro) Stable Cells
SC055-G	MLLB2 / GFP (Neomycin) stable cell line
SC056-TetR	mouse CT26 / TetR Expression (Bsd) stable cells
SC057-Bsd	MDA-MB-231 / RFP (Bsd) Stable Cell Line
SC058	HEK293 / uGFP (unstable GFP) Stable Cells
SC059-Bsd	MDA-MB-231 / Luciferase (Bsd) Stable Cell Line
SC059-Puro	MDA-MB-231 / Luciferase (Puro) Stable Cell Line



SC060-G	Human B lymphocyte / GFP Stable Cells
SC060-LG	Human B lymphocyte (Luciferase / GFP) Stable Cells
SC060-LR	Human B lymphocyte (Luciferase / RFP) Stable Cells
SC060-R	Human B lymphocyte / RFP Stable Cells
SC061-G	Mouse CT26 / GFP Stable Cells
SC061-LG	Mouse CT26 (Luciferase & GFP) Stable Cells
SC061-LR	Mouse CT26 (Luciferase & RFP) Stable Cells
SC061-PDL1	Mouse CT26 / PDL1 Stable Cells
SC061-R	Mouse CT26 / RFP Stable Cells
SC062-G	Human AsPC1 / GFP Cell Line
SC062-LG	Human AsPC1 / Luciferase and GFP Cell Line
SC062-LR	Human AsPC1 / Luciferase and RFP Cell Line
SC062-Luc	Human AsPC1 / Luciferase Cell Line
SC062-R	Human AsPC1 / RFP Cell Line
SC063-LR	Mouse B lymphocyte (Luciferase & RFP) Stable Cell
SC063-Luc	Mouse B lymphocyte / Luciferase Cell Line
SC063-R	Mouse B lymphocyte / RFP Cell Line
SC064-G	Human HaCAT / GFP (Puro) Cell Line
SC064-TetR	Human HaCAT / TetR (Bsd) Cell Line
SC065-G	Mouse MB49 / GFP Stable Cells
SC065-LG	Mouse MB49 / Luciferase & GFP Stable Cells
SC065-LR	Mouse MB49 / Luciferase & RFP Stable Cells
SC065-R	Mouse MB49 / RFP Stable Cells
SC066-G	Human ES2 / GFP Stable Cells
SC066-LG	Human ES2 / Luciferase & GFP Stable Cells
SC066-LR	Human ES2 / Luciferase & RFP Stable Cells



SC066-R	Human ES2 / RFP Stable Cells
SC066-TetR	Human ES2 / TetR (Puro) Stable Cells
SC067-G	Human SW403 / GFP Stable Cells
SC067-Luc	Human SW403 / Luciferase Stable Cells
SC068-G	Human PANC-1 / GFP (Puro) Cell Line
SC068-LG	Human PANC-1 / Luciferase & GFP (Puro) Cell Line
SC068-Luc	Human PANC-1 / Luciferase (Puro) Cell Line
SC068-R	Human PANC-1 / RFP (Puro) Cell Line
SC069-G	Human 786-O / GFP Cell Line
SC069-LG	Human 786-O / Luciferase & GFP Cell Line
SC069-luc	Human 786-O / Luciferase Cell Line
SC070-G	Hela-nucGFP stable cells
SC070-R	Hela-nucRFP stable cells
SC072-G	Human T47D / GFP Stable Cells
SC072-LG	Human T47D / Luciferase & GFP Stable Cells
SC072-Luc	Human T47D / Luciferase Stable Cells
SC073-GB	Human MCF10A / GFP (Bsd) Stable Cells
SC073-GP	Human MCF10A / GFP (Puro) Stable Cells
SC073-Luc	Human MCF10A / Luciferase (Puro) Stable Cells
SC074-GB	Human SW1990 / GFP (Bsd) Stable Cells
SC074-GP	Human SW1990 / GFP (Puro) Stable Cells
SC074-LG	Human SW1990 / Luciferase & GFP (Puro) Stable Cells
SC074-Luc	Human SW1990 / Luciferase (Puro) Stable Cells
SC017	MDA-MB-231 / niRFP (Puro) Stable Cell Line
SC075	Human ACE2 Expression in Hela Cell Line



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SC076	Human ACE2 Expression in HEK293T Cell Line
SC019	Firefly & Renilla Dual Luciferase Hela Cell Line
SC077	COVID-19 Spike (S) Protein / Hela Cell Line
SC071-Puro	Color Switch, CRE report cell line: Hela-loxP-GFP-RFP (Puro)
SC071-Neo	Color Switch, CRE report cell line: Hela-loxP-GFP-RFP (Neo)