rtTA (reverse tetracycline Transcriptional Activator)
Expression Stable Cell Line

<table>
<thead>
<tr>
<th>Catalog Number</th>
<th>Product name</th>
<th>Amount</th>
</tr>
</thead>
<tbody>
<tr>
<td>SC038-GB</td>
<td>rtTA (GFP-Bsd) / Hela stable cells</td>
<td>1.0 ml / vial</td>
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<tr>
<td>SC038-GP</td>
<td>rtTA (GFP-Puro) / Hela stable cells</td>
<td>(in cell frozen medium)</td>
</tr>
<tr>
<td>SC038-RB</td>
<td>rtTA (RFP-Bsd) / Hela stable cells</td>
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</tbody>
</table>

**Storage:** liquid Nitrogen.

**Product Description**

Hela cell was derived from human cervical cancer cells. The cell line was found to be remarkably durable, can divide an unlimited number of times in a laboratory cell culture plate as long as fundamental cell survival conditions are met.

This stable cell line is derived from HeLa cells, transformed via lentiviral system. It constitutively expresses the **rtTA-m2** (ABC65845.1) (reverse tetracycline Transcriptional Activator). **rtTA-m2** is a mutant of rtTA that has increased stability, reduced background expression in the absence of doxycycline (Dox, tetracycline analog) and improved inducibility in the presence of Dox.

The **rtTA-m2** expressed under the enhanced EF1a promoter. A "fluorescent-antibiotic" fusion dual marker is expressed under the constitutive RSV promoter (Note: the fusion fluorescent marker showed weaker fluorescent intensity than wild-type GFP). The following expression cassette was integrated into the Hela genome in the cell line (see scheme below).
This cell line expressed rtTA protein binds to tetracycline operator elements (TetO) in the presence of doxycycline (Dox). Any promoters that embedded with TetO sequences can binds to the rtTA protein when the inducer (Dox or tetracycline) is present, which activates the transcription of the promoter's downstream gene (So called, Tet-On inducible system). Therefore, this cell line can be used for Tetracycline or Dox inducible target expression when applied with target expression vectors (with TetOn inducible promoter), or for generation of any inducible target stable cell lines (after introduced a TetOn expression vector).

**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 15 ml of pre-warmed complete medium. Incubate the cells overnight in a 37°C incubator, 5% CO2.
3. The following day, replace the medium with 15 ml of pre-warmed, complete medium.

   **Note:** To maintain the cell line genetic stability for long-term culture, you can add the antibiotic into the culture medium. So optionally, add the specific antibiotic (dependent upon cell line product) in the medium at the final concentration as follows:

   - Bsd (Blasticidin): 10 ug/ml
   - Puro (Puromycin): 0.5 ug/ml

4. Incubate the cells and monitor cell density.
5. Pass cells (1:10 dilution) when the culture reaches 80-90% confluent.
6. Freeze cells at a density of 3 x 10^6 cells/ml using 90% complete medium with 10% DMSO.
Complete medium
   MEM / EBSS medium
   2mM L-glutamine
   10% Fetal Bovine Serum (FBS)
   0.1 mM MEM Non-Essential Amino Acids (NEAA)
   1% Pen-strep

Quality Control
   Each vial contains greater than 2 x 10^6 cells with >95% viability before freeze. Cells are tested free of bacteria, viruses, mycoplasma.

Warranty and user terms

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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.
Attachment: GenTarget’s pre-made stable cell line list:

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<tr>
<th>Catalog #</th>
<th>Product Name</th>
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<td>HEK293-TLV lentivirus packing cells</td>
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<tr>
<td>SC001</td>
<td>HEK293-GFP stable cells</td>
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<tr>
<td>SC002-Bsd</td>
<td>luciferase (firefly), HEK293 stable cells (Blasticidin)</td>
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<td>luciferase (firefly), HEK293 stable cells (GFP-Blasticidin)</td>
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<td>luciferase (firefly), HEK293 stable cells (GFP-Puromycin)</td>
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<td>luciferase (firefly), HEK293 stable cells (Neomycin)</td>
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<td>HEK293T / Luciferase stable cells (RFP-Puromycin)</td>
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<td>Color Switch, CRE report cell line: Hela-loxP-GFP-RFP (Neo)</td>
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<td>SC071-Puro</td>
<td>Color Switch, CRE report cell line: Hela-loxP-GFP-RFP (Puro)</td>
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<tr>
<td>SC072-G</td>
<td>Human T47D / GFP Stable Cells</td>
</tr>
<tr>
<td>SC072-LG</td>
<td>Human T47D / Luciferase &amp; GFP Stable Cells</td>
</tr>
<tr>
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<td>Human T47D / Luciferase Stable Cells</td>
</tr>
<tr>
<td>SC073-GB</td>
<td>Human MCF10A / GFP (Bsd) Stable Cells</td>
</tr>
<tr>
<td>SC073-GP</td>
<td>Human MCF10A / GFP (Puro) Stable Cells</td>
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<td>SC073-Luc</td>
<td>Human MCF10A / Luciferase (Puro) Stable Cells</td>
</tr>
<tr>
<td>SC074-GB</td>
<td>Human SW1990 / GFP (Bsd) Stable Cells</td>
</tr>
<tr>
<td>SC074-GP</td>
<td>Human SW1990 / GFP (Puro) Stable Cells</td>
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<td>Code</td>
<td>Description</td>
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<td>SC074-LG</td>
<td>Human SW1990 / Luciferase &amp; GFP (Puro) Stable Cells</td>
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<td>Human SW1990 / Luciferase (Puro) Stable Cells</td>
</tr>
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<td>SC075</td>
<td>Human ACE2 (RFP) Expression in Hela Cell Line</td>
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<td>SC076</td>
<td>Human ACE2 (RFP) Expression in HEK293T Cell Line</td>
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<td>SC076B</td>
<td>Human ACE2 (GFP) Expression in Hela Cell Line</td>
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<td>SC077</td>
<td>COVID-19 Spike (S) Protein / Hela Cell Line</td>
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<td>SC078-G</td>
<td>Mouse Panc02 / GFP Stable Cell Line</td>
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<td>Mouse Panc02 / Luciferase (Firefly) Stable Cell Line</td>
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<td>Human MIA Paca-2 / GFP Stable Cells</td>
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<tr>
<td>SC079-LG</td>
<td>Human MIA Paca-2 / Luciferase &amp; GFP Stable Cells</td>
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<tr>
<td>SC079-LR</td>
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<td>SC079-Luc</td>
<td>Human MIA Paca-2 / Luciferase Stable Cells</td>
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<td>SC079-R</td>
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<td>SC080-G</td>
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<tr>
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<td>Human HT-29 / GFP &amp; Luciferase Stable Cell Line</td>
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<tr>
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<td>Human SHP-77 / <strong>Luciferase</strong> &amp; GFP Stable Cells</td>
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<tr>
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<td>Human MDA-MB-468 / <strong>Luciferase</strong> &amp; <strong>RFP</strong> Stable Cells</td>
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