# HEK293-uGFP (unstable GFP) Stable Cell Line

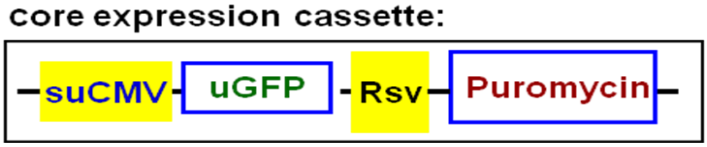
| Catalog Number | Amount | Storage |
|---|---|---|
| SC058 | 1 vial of cells (2 x $10^6$ cells) in 80% DMEM, 10% FBS, 10% DMSO | Liquid nitrogen |

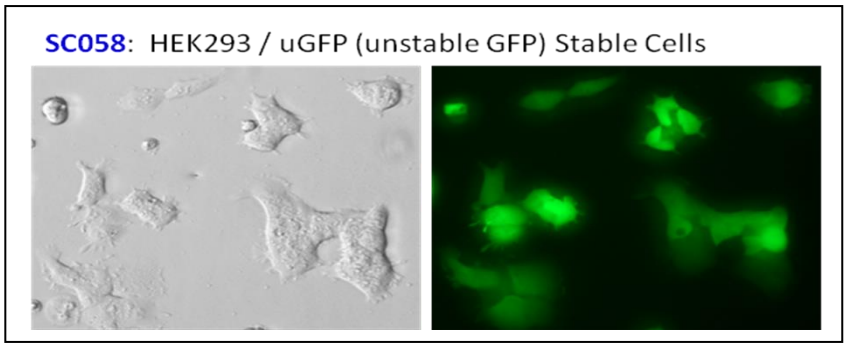
## Product Description

The 293 Cell Line is a permanent line established from primary embryonal human kidney cells transformed with sheared human adenovirus type 5 DNA. The expressed E1A adenovirus gene allows these cells to produce very high levels of protein.

The green fluorescent protein (GFP) is a widely used reporter, provide an easy detection in living cells. However, it is a very stable protein and accumulated in cells with long half-live, which limits its application that requires fast turnover responses in signal pathway assay and in knockdown / knockout detection. Therefore, the unstable GFP (uGFP) was created as the destabilized version reporter. **The uGFP is best used for the time course, dose response kinetics and for the fast responses to knockdown (via siRBA/ shRNA) or knockout (via CRISPR).**

HEK293-uGFP cells were transformed from the HEK293 cell line and stably express an engineered **unstable GFP** marker (click to see sequence). This uGFP shows an shortened in vivo half-life of ~ 2 hours. The Cell Line contains a Puromycin resistance gene. uGFP is constitutively expressed at high-levels under the CMV promoter. The following expression construct was integrated into cell's genome.




**Bright field** **GFP filer (Ex490nm/Em510nm)**

## Culture procedures



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- Thaw the vial of frozen cells quickly in a 37 °C water bath (1-3min); decontaminate the outside of the vial with 70% ethanol.
- Transfer the entire contents of the cryovial into a T-75 $cm^2$ flask containing 15 ml of pre-warmed complete medium. Incubate the cells overnight in a 37 °C incubator, 5% CO2.
- The following day, replace the medium with 15 ml of prewarmed, complete medium (Optional: add final 1 µg/ml of Puromycin in medium).
- Incubate the cells and monitor cell density.
- Pass cells (1:10 dilution) when the culture reaches 80-90% confluence.
- Freeze cells at a density of 3 x $10^6$ cells/ml using 90% complete medium with 10% DMSO.

## Complete medium

D-MEM (high glucose)
2mM L-glutamine
10% Fetal Bovine Serum (FBS)
0.1 mM MEM Non-Essential Amino Acids (NEAA)
1% Pen-strep / Antibiotic-antimycoplasma

## Quality Control

Each vial contains >2 x $10^6$ cells with >95% viability before freezing. Cells are verified to be free of bacteria, viruses, and mycoplasma.

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