

# Episomal iPSC Reprogramming Kit

Cat# RF202

## Descriptions

The human episomal iPSC reprogramming kit from ALSTEM is one of the best choices for producing footprint-free and virus-free iPSCs, which provide an ideal source for all stages of your pluripotent stem cell research. Optimized by the ALSTEM team, this kit has proven to be efficient and successful in inducing pluripotency from several different somatic cell types.

## Highlights

- Generate transgene-free, virus-free, and footprint-free iPSC cells
- Flexible in source cell selection
- Enhanced reprogramming efficiency by puromycin selection
- Optimized for feeder-free reprogramming
- Addition of small molecules are not required
- Episomal kit for human induced pluripotent stem cell (iPSC) generation
- Safe for all stages of your research from basic research to pre-clinical research

## Specifications

Product Name	Human iPS Cell Reprogramming Episomal Kit
Catalog #	RF202
Component	hiPSC Reprogramming Episomal Vectors (cat.no. RF202_1) RFP Control Vector (cat.no. RF202_2): Each kit contains sufficient material for 10 reprogramming experiments
Shipping	Ambient temperature
Storage and Stability	Store at -20 °C upon receipt. This product is stable up to 6 months when stored as directed.
Quality Control	Each lot of Human iPS Cell Reprogramming Episomal Kit is tested to ensure human fibroblasts can be reprogrammed to iPSCs.
Restricted Use	For Research Use Only. Not for use in diagnostic or therapeutic procedures.

## Related Products and Services

- Retrovirus iPSC reprogramming kit (Cat# RF101)
- EZStem Gelatin (Cat# M500)
- EZStem stem cell dissociation solution (Cat# M100)
- EZStem stem cell freezing medium (Cat# M050)
- Human iPSC cell line (Cat# iPS11)
- Custom iPSC Generation Service

## Scientific Resources

### Overview of episomal iPSC reprogramming system

Induced pluripotent stem cells (iPSCs) are genetically reprogrammed from adult cells, which are similar to natural pluripotent stem cells, eg., embryonic stem cells (ESCs). iPSCs exhibit a pluripotent stem cell-like state, as the expression of specific stem cell pluripotency markers. While these artificially generated cells are not known to exist in the human body, they show qualities remarkably similar to those of embryonic stem cells. Therefore, iPSCs are an invaluable resource for drug discovery, cell therapy, and basic research, without ESC related ethnic concern.

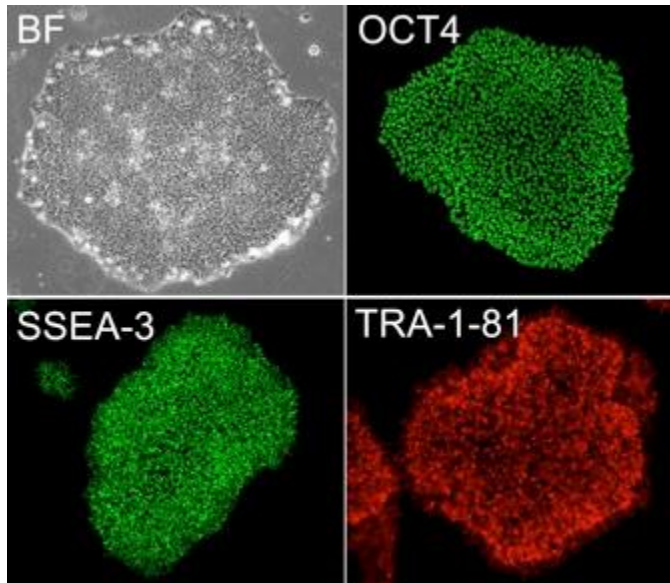
Human iPSCs were first generated in 2007 through retrovirus- or lentivirus-mediated gene transduction. However, retroviral or lentiviral vectors require integration into host chromosomes to express reprogramming genes. Integration-free human iPSCs have been generated using several methods, including adenovirus, Sendai virus, the piggyBac system, minicircle vector, episomal vectors, direct protein delivery, and synthesized mRNA. The reprogramming efficiency of these integration-free methods is impractically low in most cases. Direct delivery of proteins or RNA is labor-intensive, requiring repeated delivery of the reprogramming factors. Modifying Sendai virus vectors or preparing synthesized mRNAs are technically demanding.

The Human iPS Cell Reprogramming Episomal Kit is an optimized mixture of multiple vectors that can reprogram somatic cells to iPSCs without integration. The episomal vectors have the oriP/EBNA-1 (Epstein-Barr nuclear antigen-1) backbone that delivers the reprogramming factors as well as puromycin resistant gene. This system has been successfully demonstrated in the reprogramming of fibroblasts, as well as other adult cells, to iPSCs. High expression of transgenes due to oriP/EBNA-1 mediated nuclear import and retention of vector DNA allows iPSC derivation in a single transfection. Meanwhile, the reprogramming efficiency could be further enhanced by puromycin selection. Besides, the episomes vector usually loss at a rate of ~5% per cell cycle due to defects in vector synthesis and partitioning. And this mechanism allows the removal of episomal vectors from the iPSCs and achieve footprint-free without any additional manipulation.

This episomal mix is able to efficiently generate transgene-free, virus-free, and footprint-free induced pluripotent stem cells (iPSCs) in both feeder and feeder-free conditions. Optimized by ALSTEM team, this episomal mix has proven to be successful in inducing pluripotency for several different somatic cell types.

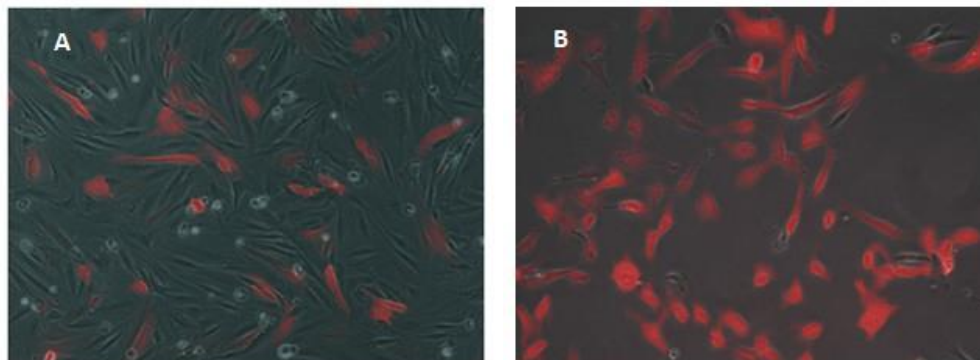
### Generate iPSC Lines from a Wide Variety of Somatic Cell Types

iPS cells have been generated by episomal vectors from a range of somatic cells, including fibroblasts, bone marrow mononuclear cells, PBMCs, lymphoblast B cells, and various disease-type fibroblasts and PBMCs. Each kit provides enough material for 10 reprogramming experiments.

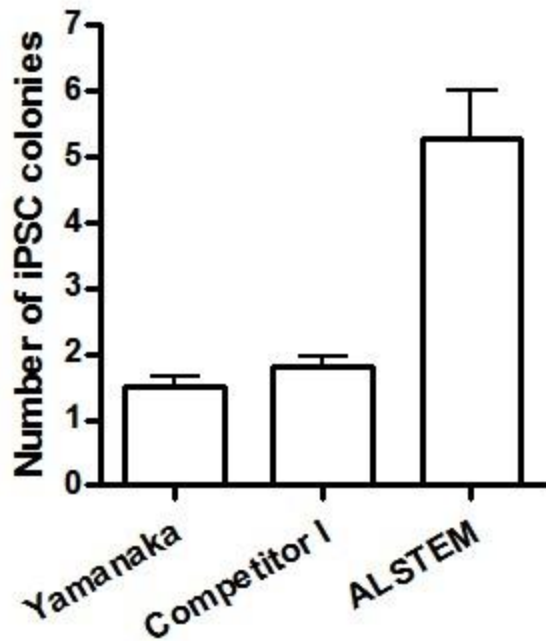


**Figure 1.** Characterization of iPSCs derived from human dermal fibroblasts using ALSTEM episomal vectors. Bright field image of hiPSC colonies (top left), immunostaining of hiPSC colonies expressing ESC specific markers OCT4, SSEA-3, and TRA-1-81.

#### Optimize Reprogramming with Puromycin Selection



**Figure 2.** (A) A typical image of the human dermal fibroblasts transfected with RFP using Neon Transfection Devices (24 hours after electroporation). ~ 30% of fibroblasts are RFP positive. (B) After puromycin selection, more than 90% of fibroblasts are expressing RFP (~5 days after puromycin selection).



**Figure 3.** ALSTEM hiPSC episomal reprogramming kit provides high reprogramming efficiency. Alkaline phosphatase (AP) positive iPSC colonies with typical human ESC morphology were counted on day 24- 27 post-transfection. The number of iPSC colonies was from  $1 \times 10^4$  transfected cells.

## Publications

1. Kondo Y, Steet R, et al. *JCI Insight*. 2018;3
2. Bell S, Silveira H, et al. *Stem cell reports*. 2018;11:183-196
3. Huang CW, Patel S, et al. *Sci Rep*. 2017;7:17401
4. Bell S, Vasuta C, et al. *Stem cells translational medicine*. 2017;6:886-896