Instruction for thawing and sub-culturing the HIV reporter cell lines:

Medium used:

Cell Name	101 Bio Cat.#	DNA construct	Medium used
Rev-A3R5-GFP	HRC-1	pNL-GFP-RRE-SA-Puro	**RPMI + 10% FCS (Puro + G418)**
Rev-A3R5-GFP/Luc	HRC-2	pNL-GFP/Luc-RRE-SA-Puro	**RPMI + 10% FCS (Puro + G418)**
Rev-A3-GFP/Luc	HRC-3	pNL-GFP/Luc-RRE-SA-Puro	**RPMI + 10% FCS (Puro)**
Rev-CEM-GFP	HRC-4	pNL-GFP-RRE-SA	**RPMI + 10% FCS**
Rev-CEM-GFP/Luc	HRC-5	pNL-GFP/Luc-RRE	**RPMI + 10% FCS**
Rev-CEM-Luc	HRC-6	pNL-Luc-RRE-SA	**RPMI + 10% FCS**

(**RPMI + 10% FCS :** RPMI 1640 + 10% Fetal calf serum.)

Initiation of cells:

1. Remove vial of frozen cells from -80°C freezer and thaw as quick as possible by putting in 37°C water bath or holding in hands.

2. Transfer the cell suspension to a sterile 15 ml tube. Add 5ml fresh medium dropwise, mix gently after each addition.

3. Collect the cells by centrifuge at 1,000 rpm for 5 minutes.

4. Aspirate off the supernatant and resuspend the cell pellet in 15 ml of media. Plate out the cells on a T75 flask.

For culturing suspension cells:

1. Count cells daily and keep at a density below 1 x 10^6 cells /ml.

2. Add fresh medium when cell density reaches 1 x 10^6 cells /ml.

Q & A

1) Does this cell line grow in suspension? Yes, it is a suspension cell line.

2) When culture HRC-1 and HRC-2 cell lines, can you specify the concentration of puromycin and G418 used?

Puromycin is used at a final concentration of 1 ug/ml. Geneticin (G418) is used at a final concentration of 1 mg/mL.

3) For maintenance, what is the suggested sub-cultivation ratio?

Please keep the cell concentration to below 1 million cells per ml. You can do a 1 to 5-10 folds dilution, the cell will duplicate daily.

P.S.

Use our HIV Infection Enhancer, enhance by 5-20 folds.

Exosome isolation customers also find our Enhancer so helpful.

We also provide 90% viability Human Primary Cells.