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Human Embryonic Kidney Cells (HEK-293)

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Protocol status: Working
We use this protocol and it's working

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Abstract

This protocol details the Human Embryonic Kidney Cells (HEK-293) culture.

Materials

Media

- DMEM full media containing (DMEM/10% FBS/1% Pen-Strep).

Materials

- 12 Well plate (1 ml/well)
- 6 Well plate/35 mm dish (9.5 cm^2-2 ml/well)
- 60 mm dish (21 cm^2-3.5 ml/dish)
- 100 mm dish (56 cm^2-10 ml/dish)
- 250 ml Flask for sub-culture/maintenance (10 ml) [Tissue culture flask-Greiner bio-one- Cat.No.-658 170]

  CELL CULTURE FLASK, 250 ML, 75 CM², PS, RED STANDARD SCREW CAP, CLEAR, CELLSTAR® TC, STERILE, 5 PCS. **greiner bio-one Catalog #**658170

Day-01 (Mon)

36m

1 Plating with DMEM full media .

2 Warm DMEM full media, PBS, and Trypsin in the 37 °C bead bath for 00:30:00 . Clean the working area by using 70% ethanol.

30m

2.1 Sup out old media without touching cells.

2.2 Wash by adding 5 mL PBS slowly, rinse, and rock back and forth.

2.3 Add 2 mL - 3 mL trypsin (0.25%); keep in incubator for 00:03:00 .

3m

2.4 Check under microscope if cells are detached, add 5 mL media and transfer to a tube.

2.5 Spin 300 x g, 00:03:00 for 00:03:00 .

3m

2.6 Sup out and add 10 mL fresh media & re-suspend cells gently and carefully.

2.7 Count cells density and split accordingly. 15,000 cells/ml for maintenance.

- (i) Usually **1.0-1.5**x10^4/ml cells for Biochem, and
- (ii) **0.5**x10^4/ml cells for IF.

Day-02 (Tue)

3 Rest.

Day-03 (Wed)

4 Replace with DMEM full media/Drug treat.

Day-04 (Thu)

5 Drug treat if necessary /Harvesting.

Day-05 (Fri)

6 Drug treat if necessary /Harvesting.