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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 cm²-2 ml/well)
- 60 mm dish (21 cm²-3.5 ml/dish)
- 100 mm dish (56 cm²-10 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** $\times 10^4$ /ml cells for Biochem, and
 - (ii) **0.5** $\times 10^4$ /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.