Systems Virology
of Emerging Respiratory Viruses

TCL007.0P - Treatment of Calu-3 cells with Interferon β

(4-12-10, Lynn Law)

This protocol is designed for cell culture using 6-well plates and 2 ml of medium/well.

I. Preparing Interferon β working solution (Sigma I4151)

Vial contains 100KU in solution.

Using sterile technique and assuming the final concentration of 500U/ml and 2 ml medium/well:
To make a 50X solution (25 KU/ml):

1. Bring INFβ up to 4 mls with a buffer that contains at least 0.1 % BSA. Aliquot into microfuge tubes or vials (~500 µl/tube). Label with contents, date, initial and anything else you think is necessary. Aliquots should be stored in refrigerator.
2. Make 20 X working solution just prior to use-
   a. Add 480 µl of 50X solution to 720 µl of cell culture medium.

II. Treating cells with Interferon β

Set up:

- Label tubes with permanent ink.
- Make Interferon β 20 X solution (100 µl /well)
- Cell culture medium
- Cold 1 X PBS
- Dry ice
- TRIZOL Reagent

1. Remove medium from wells. Adding 1.9 ml of culture medium back to each well.
2. Add 100 µl of Interferon β 20X stock solution to all wells that are to be treated.
3. Start timing once treatment is added to the cells.

III. Collecting samples for RNA analysis

- Samples are collected following protocol TCL001.0P

1. Harvest T=0 samples just before proceeding with Step II above.
2. Harvest samples post-treatment according to experimental design.