PROTOCOL OF USING LUMI-SCINT FOR LUCIFERASE REPORTER ASSAY

Lan Zhou 11/01/00, Edited by JYP 8/12/02

Cell Extract Preparation

- 1. Make 1X lysis buffer by diluting 5X lysis buffer with water after 24hrs of Txn.
- 2. Add 200ul for each well (12-well plate) and incubate cells @ RT 10min.
- 3. Collect the cell lysate in 1.5ml tube and spin down for 3min.
- 4. The supernatants (cell extracts) should be used immediately or transferred to a new tube and frozen @ -80°C.

Luciferase Assay

- 1. Thaw constituted luciferase reagent (Don't thaw it @ temperature above 25°C).
- 2. Add 50ul of the substrate and 10-20ul cell extract into a tube; mix them.
- 3. Place the tube into the chamber of machine and close the lid immediately.
- 4. Press the key "next" and then "start."
- 5. Read the number.

Set up the Program

- 1. Function 3
- 2. Delay time 5
- 3. Count time 10
- 4. Sample ID
- 5. Sample 1 and press Start/Stop.
- 6. Get its number.
- 7. Change sample and press "Next" and "Start/Stop."
- 8. Get another number.