Scratch Wound Assay General Protocol

Stack Lab

1. Seed the cells in 8 well plate (Nunc # 174934), culture until confluent.

2. Wash the cells with PBS 3 times, add SFM (2 ml/ well), incubate in 37°C over night. Pretreat the cells if desired.

3. Scratch the cells: Hold a 200 ul tip, tip point against bottom of the culture dish, gently but quickly scratch a straight line through the cells. Two or three lines per well is recommended. Check the scratch under microscope to see if you get a well cut line. Wash out floating cells with PBS.

4. Add fresh SFM and treatment (if desired).

5. Take 0 hour photo, make a dot on bottom of plate for later reference. Common end points are 24-48 h. For better visualization, the cells can be fixed and stained with Diff-Quick.

6. Analysis of migration distance using at least 10 points along the wound will provide comparison of the ‘% wound closure’ relative to the initial wound width.

Revised 09/2008 (YL)