

## Inoculation of the Tuning Fork Assembly

**1.0 Purpose:** To inoculate Tuning Fork Assembly(s) with adherent cells for use in the Cytomatrix™ SW-Spinner System.

### **2.0 Materials:**

- 2.1 One p100 (55 cm<sup>2</sup>) tissue culture dish per 2 tuning fork assemblies
- 2.2 Media
- 2.3 P200 Pipetman with tips
- 2.4 Razor blade/scissors
- 2.5 Tuning Fork Assemblies

### **3.0 Procedure:**

- **Completely read through the procedure before starting to familiarize yourself with the new terminology for the parts you will be assembling.**

### **4.0 Matrix Seeding**

- 4.1 Outside of hood, cut open outer bag containing the tuning fork assembly (Fig. 1).
- 4.2 Remove inner bag and place into hood.
- 4.3 Cut open inner bag at end closest to black handle.
- 4.4 Remove assembly by grasping tab of black handle and place into a sterile tissue culture plate.

*NOTE: Care should be taken so that black handle is the only portion touched. Do not touch the clear portion of the tuning fork nor the matrices.*

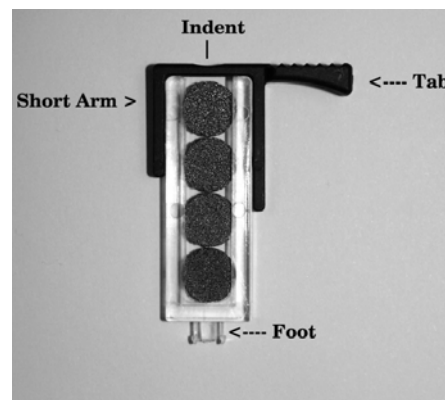
- 4.5 Seed matrix units by drop-wise addition of cell suspension to each individual matrix (see note below). A seeding volume of 100 µl per matrix is recommended.

*NOTE: It is recommended to start with a seeding number of 250,000 cells per unit (inoculum concentration =  $2.5e^6$  cells/ml). This results in a seeding concentration of approximately 4000 cells/cm<sup>2</sup>. Based on your needs/experience this concentration may be adjusted higher or lower.*

*HINT: To ensure complete inoculation, gently touch pipet tip to surface of the matrix to absorb any excess liquid remaining on tip.*

- 4.6 Incubate cells for 2-4 hours in a humidified incubator to allow for cell attachment.

*NOTE: This is sufficient for most adherent cell types, but individual experience may vary. Plan incubation time accordingly.*



**Figure 1. Tuning Fork Assembly**