

Standard Operating Procedure

(Laboratory Practices)

Infectious Agent and Strain: Unfixed Human Primary Cells**Lab/Location(s): Jimmy Fund 415****Principle Investigator: John Daley****Protocol Name & Number: Analysis Unfixed Human Primary Cells****Current Revision Date 07/23/2015****Unfixed Human Primary Cells usage practice:**

1. Unfixed Human Primary Cells will be analyzed on the designated instruments:
 - a. BD LSR analyzer

Researchers must run their **Unfixed Human Primary Cells** samples in 96 or 384-well micro titer plates on the HTS (High Throughput Sampler) attached to the above stated analyzers **ONLY**. You may not run Unfixed Human Primary Cells on the BD LSRFortessa. Usage of the HTS system allows for fully automated sample acquisition.

2. Researcher must use universal safety precautions; wear disposable lab coats and gloves.

96 Well Plate Procedure

Analyzer (BD LSR analyzer) Disinfectant protocol for 96well micro titer plate is as follows:

- a) Last row on EACH 96 well plate must be treated as a disinfectant row
(**no exceptions**). This will sanitize the HTS tubing as well as the sample tubing on the analyzer.

Plate Loader Settings for Row: H Wells: 1-12

Sample Flow Rate: 3.0
Sample Volume: 175ul
Mixing Volume: 100
Mixing Speed: 200
Number of Mixes: 0
Wash Volume: 200ul

- Wells 1-6 will contain 200ul (10x) Bleach
- Wells 7-9 will contain 200ul BD FACSRinse
- Wells 10-12 200ul ddH₂O

384 Well Plate Procedure

Analyzer (BD LSR analyzer) Disinfectant protocol for 384well micro titer plate is as follows.

- a) The last row on EACH 384 well plate must be treated as a disinfectant row

(No exceptions). This will sanitize the HTS tubing as well as the sample tubing on the analyzer.

Plate Loader Settings for Row: P Wells: 1-24
Sample Flow Rate: 3.0
Sample Volume: 90ul
Mixing Volume: 45
Mixing Speed: 200
Number of Mixes: 0
Wash Volume: 200ul

- Wells 1-12 will contain 100ul (10x) Bleach
- Wells 13-17 will contain 200ul BD FACSRinse
- Wells 18-24 200ul ddH₂O

Post-analysis procedure:

After each analysis the researcher is responsible for disinfecting the areas used for **Unfixed Human Primary Cells** analysis work.

Please spray the following with **10% Bleach** and use the **Super Sani-cloth** wipes to clean accordingly

- Analyzer
- Analyzer Sample Station
- All affected areas
- The researcher is responsible for removal and disposal of their gloves; disposable lab coat and all wipes used to disinfect the analyzer and affected areas.

- Contact the Flow Cytometry Facility (617-632-3179) or Suzan Lazo (617-632-4571) for more information.