Standard Operating Procedure (Laboratory Practices)

Infectious Agent and Strain: Unfixed Human Primary Cells Lab/Location(s): Jimmy Fund 415, Mayer 583, & Mayer 522a

Principal Investigator: John Daley

Protocol Name & Number: Analysis of Unfixed Human Primary Cells

Current Revision Date: 11/07/2017

Unfixed Human Primary Cells usage practice:

1. Unfixed Human Primary Cells will only be analyzed on the designated instruments:

a. BD LSRFortessa HTS Analyzerb. BD FACSCanto II HTS Analyzer

- Researchers must run their Unfixed Human Primary samples in 96 or 384-well micro titer plates on the HTS (High Throughput Sampler) attached to the above stated analyzers ONLY. 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 Acquisition Procedure

Analyzer Disinfectant protocol for 96 well micro titer plate is as follows:

Last row on EACH 96 well plate must be treated as a disinfectant row.
 This will sanitize acquisition sample tubing.

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

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

- Wells 1-6 will contain 200µl (10%) Bleach
- Wells 7-9 will contain 200µl BD FACSRinse
- Wells 10-12 will contain 200µl ddH₂0

384 Well Plate Acquisition Procedure

Analyzer Disinfectant protocol for 384 well micro titer plate is as follows:

• Last row on EACH 384 well plate must be treated as a disinfectant row. This will sanitize acquisition sample tubing.

Plate Loader Settings for Row: P Wells: 1-24

Sample Flow Rate: 3.0
Sample Volume: 90µl
Mixing Volume: 45
Mixing Speed: 200
Number of Mixes: 0
Wash Volume: 200µl

- Wells 1-12 will contain 100µl (10%) Bleach
- Wells 13-17 will contain 200µl BD FACSRinse
- Wells 18-24 200µl ddH₂0

Post-analysis procedure

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

Please spray the following with 10% Bleach and disinfect with Super Sani-cloth wipes:

- Instrument
- Instrument 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.