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Guava Flow Cytometry

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ABSTRACT

Please contact Dr. Steven Wilhelm (wilhelm@utk.edu) for additional information regarding this protocol.

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6520

Cleaning the machine

- 1 Click CLEANING from the main menu

Note

Always clean the machine before and after use

- 2 Click START CLEAN
- 3 Load the following:
 - *dH₂O in W1,W2, W3, W4, W5, W6, and position 1
 - *100 µL bleach in position 2
 - *GUAVA ICF in position 3 and 4
 - *dH₂O in capillary shutdown (position 9)

GUAVA InCyte Acquisition Workflow

- 4 To ensure optimal performance, allow the machine to warm up for at least 10 min prior to acquiring samples.
- 5 Select InCyte from the main menu
- 6 Click the EDIT WORKLIST button
- 7 Define the worklist parameters (e.g., wells to analyze, events to acquire, time to acquire, dilution factor, original volume, replicates, etc)
- 8 Click RUN WORKLIST

- 9 Select the folder where you want to save the data file and click SAVE
- 10 Select the Analysis Method that you want to use for your specific sample set. To open a saved method, click RETRIEVE on the dialog box, then OPEN.

Note

If you want to creat a new Analysis Method, click NEW, then OK and set the parameters.
- 11 Place the microtiter plate in the tray
- 12 Load the following cleaning solutions:
 - *dH₂O in W2, W4, W5
 - *Empty tubes in W5, W6
 - *100 µL bleach in W1
 - *ICF in position 10
 - *dH₂O in position 9 (capillary shutdown)
- 13 Click OK to load
- 14 Select the sample for ADJUST SETTINGS in the dialog box
- 15 Select the type of plots and parameters you wish to display
- 16 Use the GAIN CONTROLS to select the Threshold parameter from the drop-down menu and use to adjust the gains (FSC, SSC, GRN, YLW, RED, NIR, RED2, or NIR2)

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- 17** To adjust COMPENSATION, click NEXT STEP, then ADJUST SETTINGS
 - 18** When you are finished adjusting the settings, click NEXT STEP.
 - 19** Click RESUME WORKLIST and it will begin acquiring samples