

Guava Flow Cytometry

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Abstract

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

Citation: Dr. Steven Wilhelm, Alyssa Alsante Guava Flow Cytometry. **protocols.io**

dx.doi.org/10.17504/protocols.io.ikyccxw

Published: 14 Jul 2017

Protocol

Cleaning the machine

Step 1.

Click CLEANING from the main menu

NOTES

Alyssa Alsante 29 Jun 2017

Always clean the machine before and after use

Cleaning the machine

Step 2.

Click START CLEAN

Cleaning the machine

Step 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

Step 4.

To ensure optimal performance, allow the machine to warm up for at least 10 min prior to acquiring

samples.

GUAVA InCyte Acquisition Workflow

Step 5.

Select InCyte from the main menu

GUAVA InCyte Acquisition Workflow

Step 6.

Click the EDIT WORKLIST button

GUAVA InCyte Acquisition Workflow

Step 7.

Define the worklist parameters (e.g., wells to analyze, events to acquire, time to acquire, dilution factor, original volume, replicates, etc)

GUAVA InCyte Acquisition Workflow

Step 8.

Click RUN WORKLIST

GUAVA InCyte Acquisition Workflow

Step 9.

Select the folder where you want to save the data file and click SAVE

GUAVA InCyte Acquisition Workflow

Step 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.

🗨 NOTES

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If you want to creat a new Analysis Method, click NEW, then OK and set the parameters.

GUAVA InCyte Acquisition Workflow

Step 11.

Place the microtiter plate in the tray

GUAVA InCyte Acquisition Workflow

Step 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)

GUAVA InCyte Acquisition Workflow

Step 13.

Click OK to load

GUAVA InCyte Acquisition Workflow

Step 14.

Select the sample for ADJUST SETTINGS in the dialog box

GUAVA InCyte Acquisition Workflow

Step 15.

Select the type of plots and parameters you wish to display

GUAVA InCyte Acquisition Workflow

Step 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)

GUAVA InCyte Acquisition Workflow

Step 17.

To adjust COMPENSATION, click NEXT STEP, then ADJUST SETTINGS

GUAVA InCyte Acquisition Workflow

Step 18.

When you are finished adjusting the settings, click NEXT STEP.

GUAVA InCyte Acquisition Workflow

Step 19.

Click RESUME WORKLIST and it will begin acquiring samples