Job Aid

BD FACSymphony™ A1 System: Performing daily small particle detector QC

This job aid contains instructions for daily setup of the small particle detector when using the BD FACSymphony™ A1 Flow Cytometer. See the Small particle detector chapter in the *BD FACSymphony™ A1 Flow Cytometer user guide* for more information.

Before you begin

- Create the SPD QC Experiment template described in the BD FACSymphony™ A1 System: Creating the small particle detector QC experiment template job aid.
- Create the SPD setup application settings described in BD FACSymphony™ A1 System: Performing the initial small particle detector setup procedures job aid.
- Start the cytometer and run a performance check.

Preparing the Samples

Preparing the Megamix bead tube:

- Label a clean, capped 12x75 tube *Megamix beads*.
- Mix the Megamix-Plus SSC beads by gently pipetting the mixture.
- Pipet 500 µL of the Megamix-Plus SSC beads into the Megamix beads tube.

Preparing the diluent tube:

- Label a clean, capped 12x75 tube *Diluent*.
- Pipet 3 mL of the 0.2 µm-filtered 1X PBS into the Diluent tube.

Preparing the cleaning tube:

- Label a clean, capped 12x75 tube *Detergent solution*.
- Prepare a tube with 3 mL of a 1.5% dilution of BD® Detergent Solution Concentrate.

NOTE: Never mix BD® Detergent Solution Concentrate and bleach because they create chlorine gas.

Performing small particle detector QC

Setting up the experiment

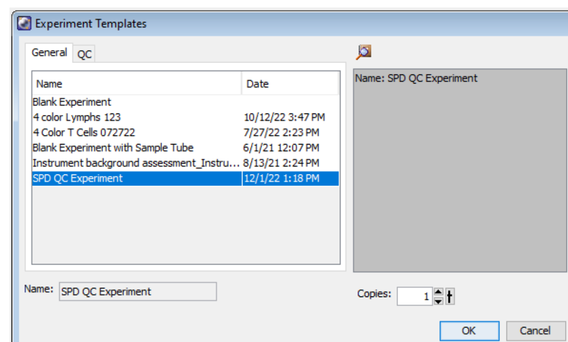
① Verify that your cytometer configuration is appropriate.

Your current configuration will need the same as the configuration used to create the SPD QC Experiment template.

② Open the SPD QC Experiment template.

a. Select a folder in the Browser, then select **Experiment > New Experiment.**

b. Select the SPD QC Experiment template and click **OK.**



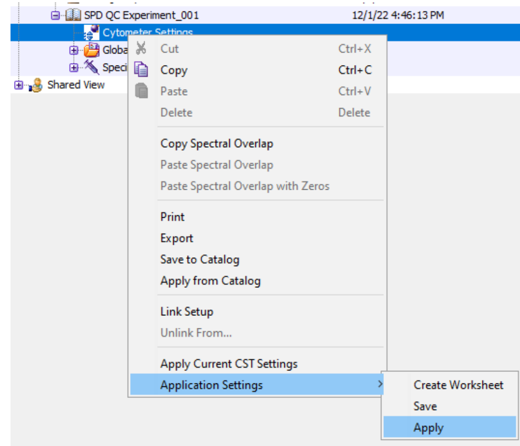
Applying application settings

① Apply the SPD setup application settings.

a. Right-click the **Cytometer Settings** icon and select **Application Settings > Apply.**

b. Select the SPD setup settings you created previously and click **Apply**.

② If the Cytometer Settings Mismatch dialog opens, click **Overwrite**.

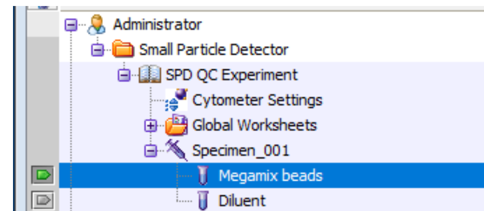


Verifying the application settings

① On the cytometer control panel, press the RUN and LOW buttons.

NOTE: For best results when working with small particle detection, the cytometer should stay in run mode with a low flow rate.

② Set the current tube pointer to the Megamix beads tube.



③ Install the Megamix beads tube onto the cytometer.

a. Remove the DI water tube from the SIP.

b. Wipe the SIP with a delicate task wipe.

c. Install the Megamix beads tube on the SIP. Place the tube support arm under the tube.

④ Click **Acquire Data** in the Acquisition Dashboard.

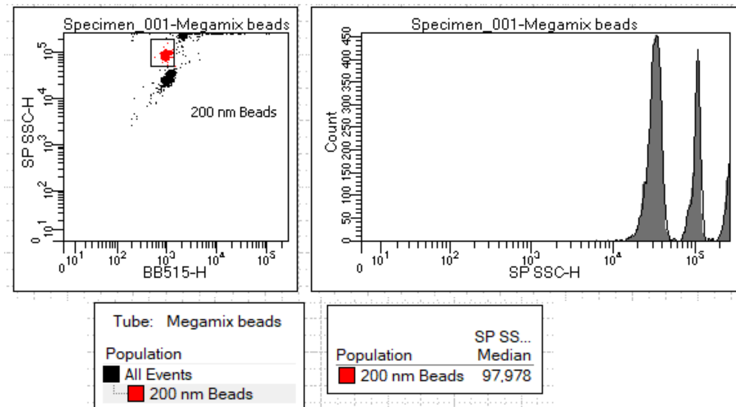
NOTE: Leave the bead tube on the cytometer until the directions tell you to remove the tube. If you must remove the tube, follow the SPD tube removal procedure.

⑤ In the Cytometer window, adjust the SP SSC voltage to place the 200 nm SP SSC-H median at around 100,000, if needed.

⑥ Adjust the 200 nm bead gate, if needed.

Verify that the bead peaks are tight and well separated. If not, realign the sample particle detector.

See the BD FACSymphony™ A1 System: Performing the initial small particle detector setup procedures job aid for more information.



Recording bead data

① Click **Record Data**.

② When recording is complete, install the Detergent solution tube onto the SIP.

a. Remove the Megamix beads tube from the SIP.

b. Wipe the SIP with a delicate task wipe.

c. Install the Detergent solution tube on the SIP. Place the tube support arm under the tube.

③ Run the detergent solution for 15 minutes.

Recording background data

① Install the Diluent tube onto the SIP.

a. Remove the Detergent Solution tube from the SIP.

b. Wipe the SIP with a delicate task wipe.

c. Install the Diluent tube on the SIP. Place the tube support arm under the tube.

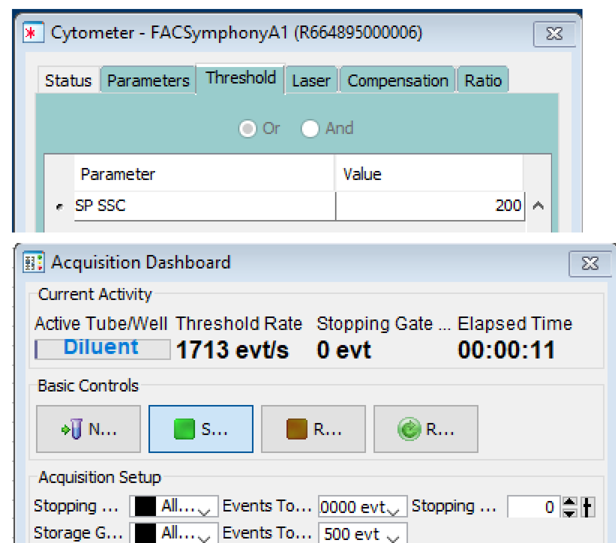
② Set the current tube pointer to the Diluent tube.

③ In the Threshold tab, select **SP SSC** as the parameter and verify that the threshold value is set to 200.

④ Click **Acquire Data.**

⑤ In the Acquisition Dashboard, observe the threshold rate for approximately 5 minutes.

- The threshold rate should slowly decrease and then plateau.
- If the threshold rate has not plateaued after 5 minutes, see the Small particle detector troubleshooting section in the BD FACSymphony™ A1 Flow Cytometer user's guide.



⑥ Click **Record Data.**

⑦ Record for 1 minute, then click **Stop Recording.**

Running your samples

The cytometer is now ready to run your samples.

Tips for running samples:

- Leave the instrument in run mode and low flow rate after starting a small particle experiment, even when waiting between samples.
- Wipe the SIP between samples and avoid leaving the tube support arm to the side for longer than 10 seconds.
- If high background noise is noticed when running samples, clean the fluidics by running a fresh Detergent solution tube for 15 minutes between samples.

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