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27 SOP 4.

HLA B27 SOP 4: Acquisition of Stained Whole Blood Samples on the BD FACSCanto™ II Flow Cytometer

Purpose

To acquire data on the BD FACSCanto[™] II flow cytometer using stained whole blood specimens.

Scope

This procedure applies to the clinical laboratory environment with the BD FACSCanto II flow cytometer for the purpose of detecting the HLA B27 antigen using whole blood specimens. We recommend that all personnel who operate the instrument be sufficiently trained to fully perform and implement this guideline.

Equipment Required

BD FACSCanto II flow cytometer and workstation Vortex

Materials Required

Biohazard safety manual Biohazard sharps waste container Personal protective equipment (PPE)

- Protective gloves
- Protective eyewear
- Closed-toe shoes
- Lab coat

Prepared immunofluorescent labeled samples for acquisition

Procedure

Before beginning this procedure, start the flow cytometer according to HLA-B27 SOP 1: BD FACSCanto™ II Cytometer Startup Procedure.

Creating a new worklist without importing the BD FACS[™] SPA III worklist

- 1. Open BD FACSCanto[™] clinical software.
- 2. Enter the appropriate user and log in information.
- 3. Enter the word *Control* in the sample name column. This will allow generation of a .csv file to monitor the control results.
- 4. Enter the lot number or other identifier of the HLA-B27 control positive or negative for the control sample in the **Sample ID** field.



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- 5. In the Panel column, select the HLA-B27 panel.
- 6. Repeat steps 3 and 4 for any remaining control tubes.
- 7. Enter the patient information as required for your facility.
 - **Sample ID** is a required field which can be manually entered or read from a barcode reader.
 - The **Sample Name** and **Case Number** fields are optional.
- 8. Continue entering the remaining samples into the worklist.

Importing an HLA-B27 worklist from the SPA III

- 1. Start BD FACSCanto clinical software.
- 2. Enter the appropriate user and login information.
- 3. From the File menu, select Import SPA Worklist.



4. Navigate to the appropriate HLA-B27 worklist and click **OK**.

Acquiring data

- 1. Ensure that the tube guide is in the loader position, surrounding the SIT.
- 2. Vortex each tube for two seconds before installing the carousel on the BD FACSCanto II cytometer.



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- 3. Enter the appropriate carousel number in the worklist if manually creating the worklist.
- 4. Click Run in BD FACSCanto clinical software.
- 5. In the dialog that opens, save changes to the worklist to include today's date and your initials or run number. Click **Save**.

The samples will be aquired according to the specifications in the software.

6. Once the worklist is complete, a dialog opens. Click **OK** and continue to data analysis.

References

- BD FACSCanto[™] II Instructions for Use, document 23-12882-01.
- BD FACSCanto™ Clinical Software Reference Manual, document 23-14529-00.
- BD HLA-B27 Application Guide for BD FACSCanto Flow Cytometers, document 343366 Rev. A.

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