

SpectroFlo[®] VERSION 3.2.1 RELEASE NOTES

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OVERVIEW

SpectroFlo® 3.2.1 is a patch release to fix a couple of critical defects for multiple autofluorescence extraction and fluidics shutdown using tube rack. In addition, corrections of several other minor defects for multiple autofluorescence extraction are also included.

SpectroFlo® 3.2.1 should only be installed on instruments with the following versions of firmware (or higher). If these versions are not present, the firmware must be updated at the time of software installation:

Paired Loader	Fluidic Motherboard Hardware Revision	SpectroFlo® 3.X
Loader 1.5 'AMS'	Rev 0C and Newer	FMB Firmware: 0.9.70 Aurora AFPGA: v1.0.2 Aurora Firmware (MFPGA): v1.0.1 LMB Firmware: v1.0.73
	Rev 0B	FMB Firmware: 0.9.63.20200312 Aurora AFPGA: v1.0.2 Aurora Firmware (MFPGA): v1.0.1 LMB Firmware: v1.0.73
Loader 2.0 'ASL'	Rev 0C and Newer	FMB Firmware: 0.9.70 Aurora AFPGA: v1.0.2 Aurora Firmware (MFPGA): v1.0.1 LMB Firmware: v2.0.04
	Rev 0B	FMB Firmware: 0.9.63.20200312 Aurora AFPGA: v1.0.2 Aurora Firmware (MFPGA): v1.0.1 LMB Firmware: v2.0.04

Software installation instructions are included at the end of this document.

DEFECT FIXES

Unmixed Result for Multiple Autofluorescence Extraction Incorrect Sometimes: In certain special scenarios, it was discovered that the unmixed result for multiple autofluorescence was incorrect. This defect is corrected in this new version.

Incorrect SIT Position after Fluidics Shutdown Using Tube Rack: It was found that the SIT position was not maintained as intended after fluidics shutdown using tube rack. Instead of being kept down and immersed in the DI water tube to prevent drying out, the SIT was lifted. This issue is corrected.

Edited Multiple Autofluorescence Extraction Not Applied: After initial creation of the multiple AF controls, when Edit Multiple AF to modify some of the gates, only the changes made to the leaf gate were applied, while parent gates were not updated. This issue has been resolved. Now any modifications to both leaf and parent gates will be properly applied to the already generated AF controls.

Unable to Extract AF from Gate If No Area Parameters are available: Exception occurs when attempting to extract AF from a gate if the unstained control tube did not have area signal parameters. This defect is fixed.

Exceptions When Esc Key is Pressed in Some Specific Rare Scenarios: In some specific rare scenarios, pressing the Esc key during loading of the Autofluorescence Explorer or the unmixing wizard could result in exceptions. These issues have been addressed in this version.

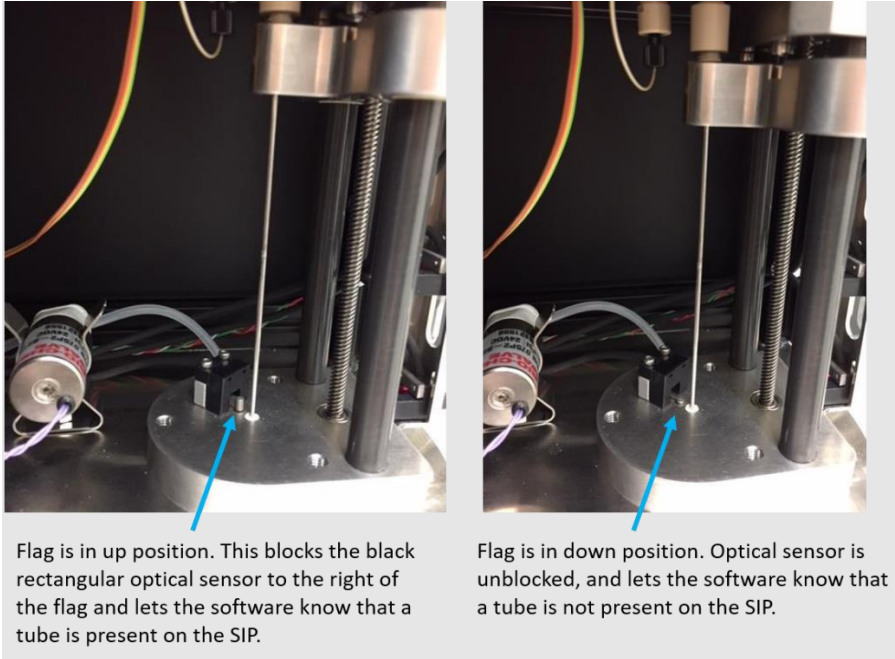
Drag and Drop Autofluorescence Control: In version 3.2.0, it was possible to drag and drop the generated AF controls to a sample group. In this new version, the drag and drop of the AF controls has been locked to prevent possible operator errors.

KNOWN SOFTWARE DEFECTS

- **Post-Acquisition Unmixing File Size Constraint:** We recommend only unmixing multicolor FCS files that contain fewer than 10,000,000 total events. Files that are bigger than 10,000,000 events may take a long time to unmix.
- **Events to Display & Live Unmixing:** If you want to display a lot of events (e.g., 1,000,000) while live unmixing a tube, the software will become very slow. We recommend reducing your events to display to 50,000 events or less when live unmixing your data. After the acquisition is finished, you can display 5-100% of your recorded events without any negative performance effects.
- **Auto-Scaling of Axes for Unmixed Files:** Auto-scaling of plot axes (found in Plot Properties) does not always apply properly for an unmixed tube. You may need to toggle to Manual Scale and then back to Auto Axis Scale to get the scaling to update and apply correctly.
- **Limitation for the Name of User Defined Keyword:** "Name" cannot be used as the key of a user defined keyword. The software will not prevent users from doing so. However, if users define such a keyword and this keyword is added in the statistics table, batch analysis cannot be completed due to a conflict with a reserved string used in the statistics table of SpectroFlo®.
- **Inconsistent Format of Displayed Week in Exported Use Time File:** When exporting the use time log by week, the week number for weeks 1-12 is displayed with an incorrect format in Excel by default.
- **Disabled Laser Delay UI:** In some rare scenarios the Laser Delay fields are not selectable from the Instrument Control Window. They are always accessible if the Instrument Control Window is floated.
- **Data Maintenance App and SpectroFlo® can be Opened at the Same Time:** In some rare scenarios, both the Data Maintenance App and SpectroFlo® can be opened at the same time. When this happens, please close one program before proceeding.

KNOWN HARDWARE DEFECTS

- **Automatic SIT Flush:** SIT flush does not always occur automatically between sample tubes. Occasionally, the mechanical flag that blocks and unblocks the tube sensor gets stuck in the tube present position. If you do not hear a SIT flush after unloading your tube, open the SIT door and check the position of the flag. If it is stuck in the up position when no tube is present, press the flag down. The SIT flush will then execute. If you notice a lot of salt buildup around the flag, let your Cytek Field Service Engineer know so they can clean it out for you.

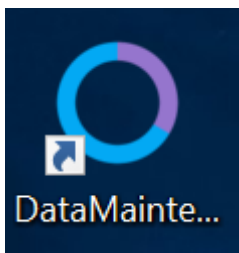


DATA BACKUP AND SOFTWARE INSTALLATION INSTRUCTIONS

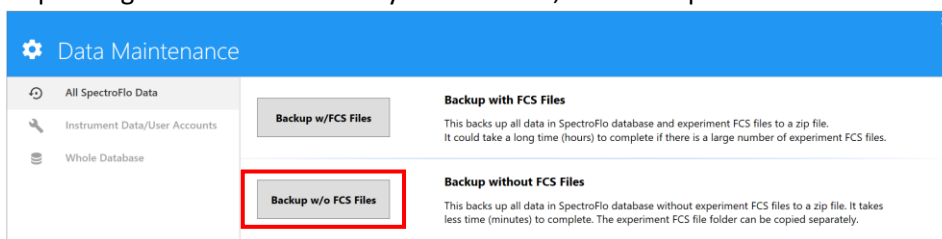
Before upgrading your system to SpectroFlo[®] v3.2.1, we recommend that you back up your data. If anything goes wrong during the installation process, you can always revert to the backup files.

Backing Up Your Data

1. Open the SpectroFlo[®] Data Maintenance tool.

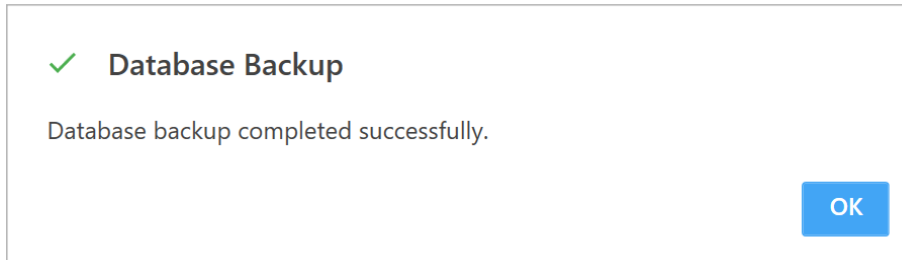


2. Click **Backup w/o FCS Files** button. Select a folder to export the backup ZIP file to and click OK. Depending how much data is in your software, this backup could take some time.



Depending how much data is in your software, this backup could take some time.

3. A message appears when the backup has completed. Click **OK**.



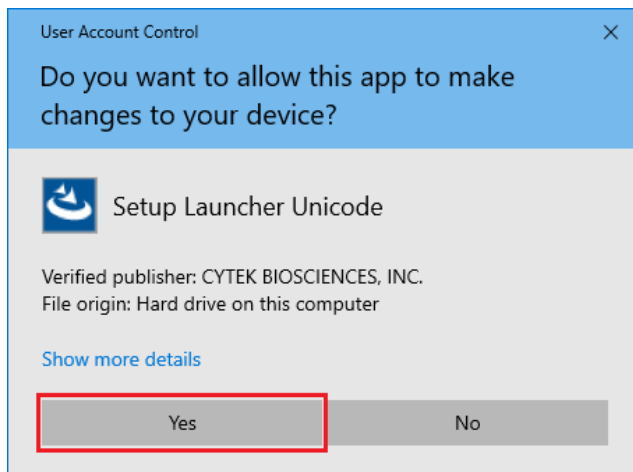
Now that you've backed up your system, continue with the following software upgrade instructions.

Upgrading SpectroFlo[®] from 3.1.x or 3.2.0 to 3.2.1

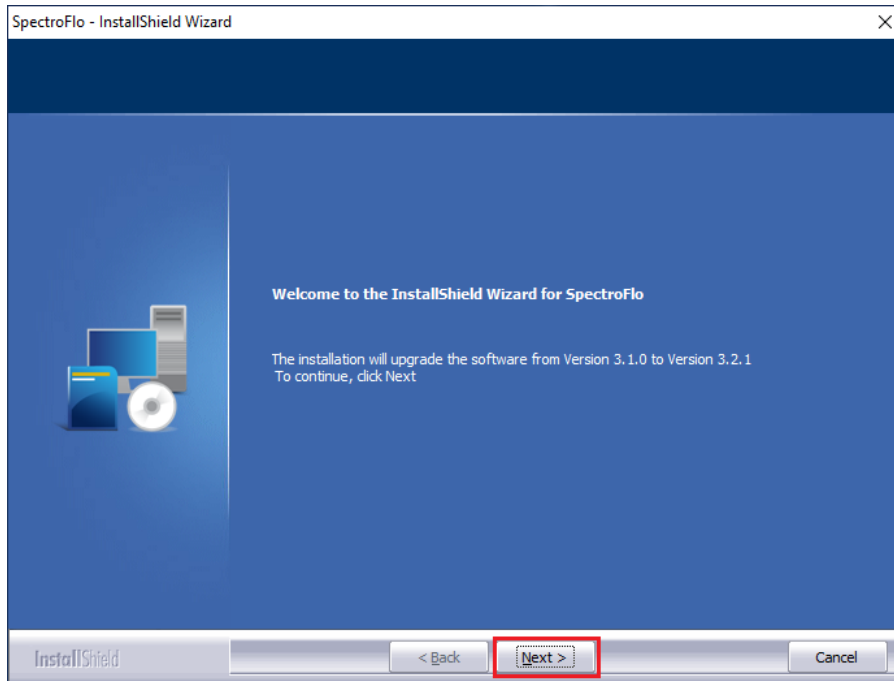
The upgrading procedure is as follows:

1. Download the setup package. Log into the host computer using an account with administrative rights.
2. Start the installer.

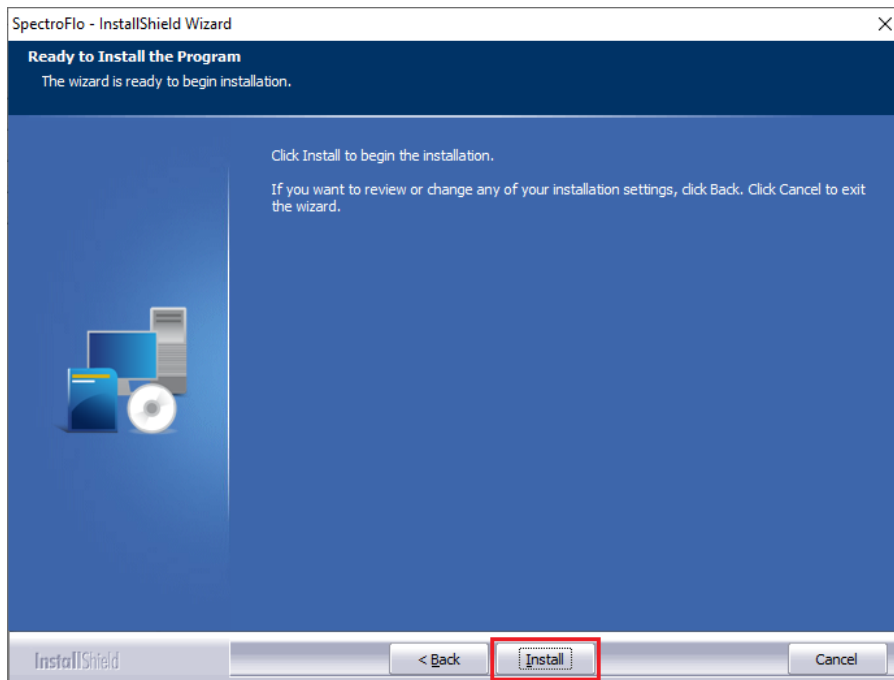
Note: this screen may not show if your PC's user account control setting is set to "Never notify". If the following message appears, click **Yes**.



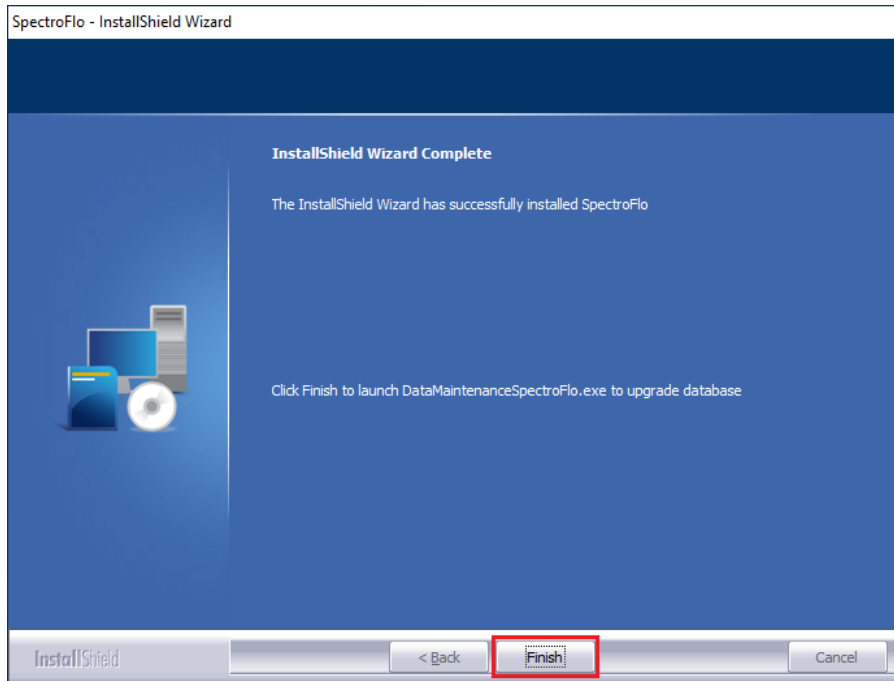
3. Confirm the version and then click **Next** to continue.



4. Click **Install** to begin the installation.



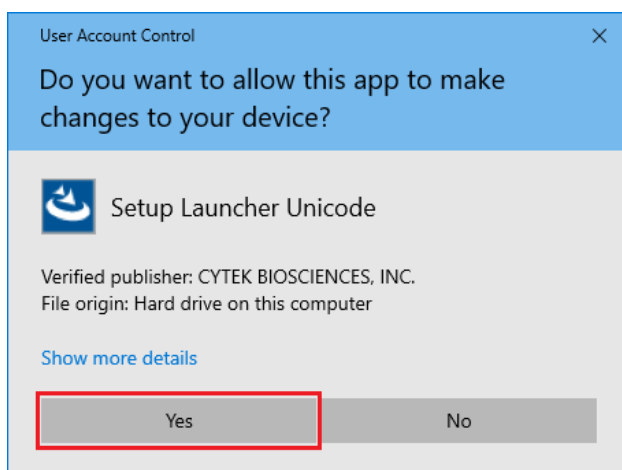
5. Click **Finish** to finish the installation.



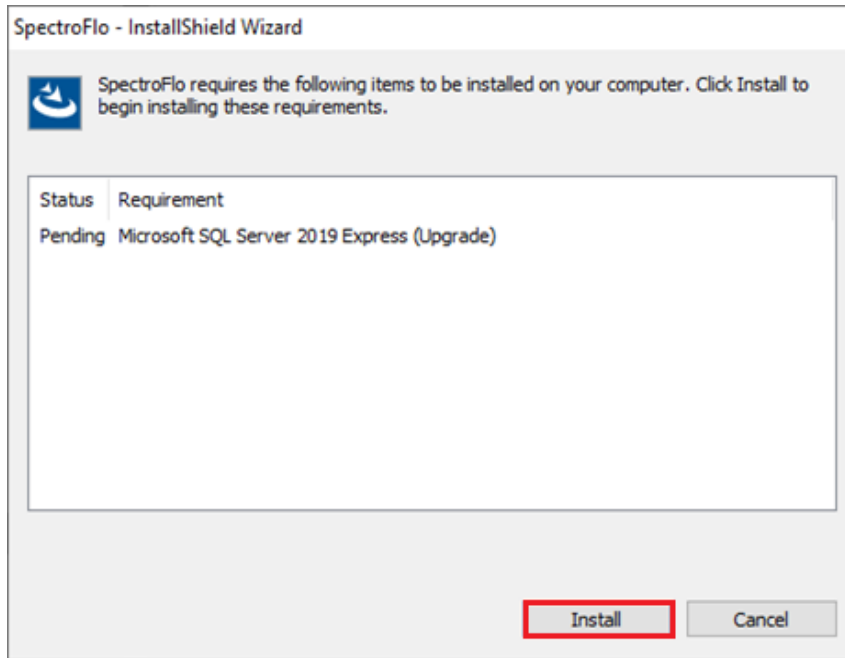
Installing SpectroFlo® 3.2.1 on a New PC

1. Download the setup package. Log into the host computer using an account with administrative rights.
2. Start the installer.

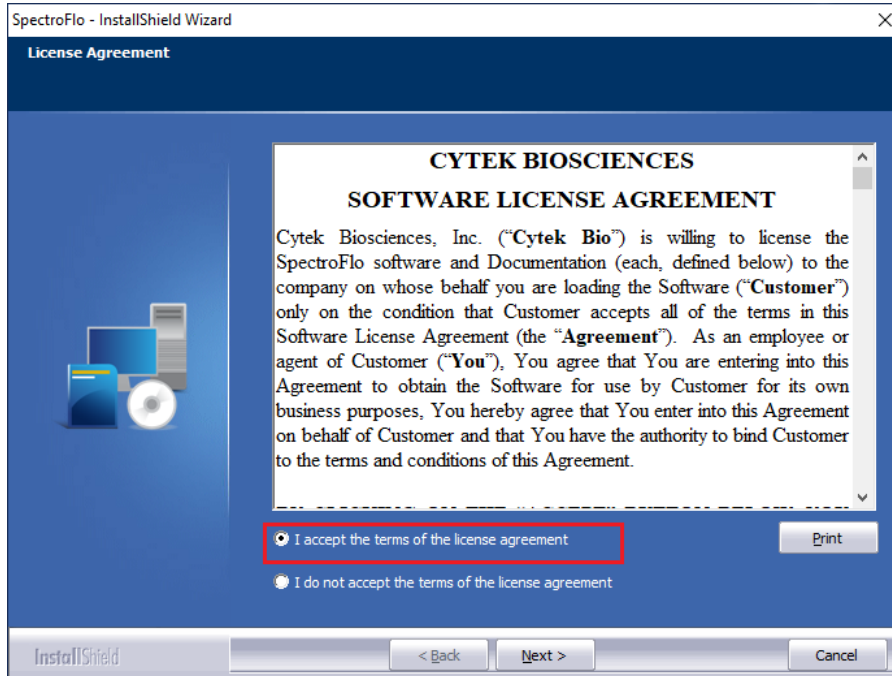
Note: this screen may not show if your PC's user account control setting is set to "Never notify".
If the following appears, click **Yes**.



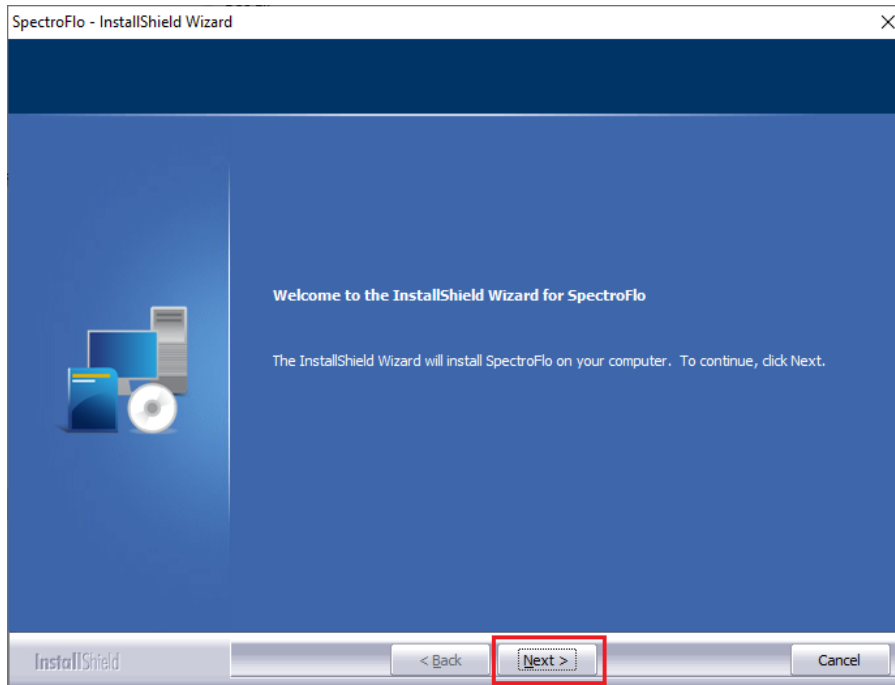
3. Click **Install** to install Microsoft SQL Server 2019 Express RTM.



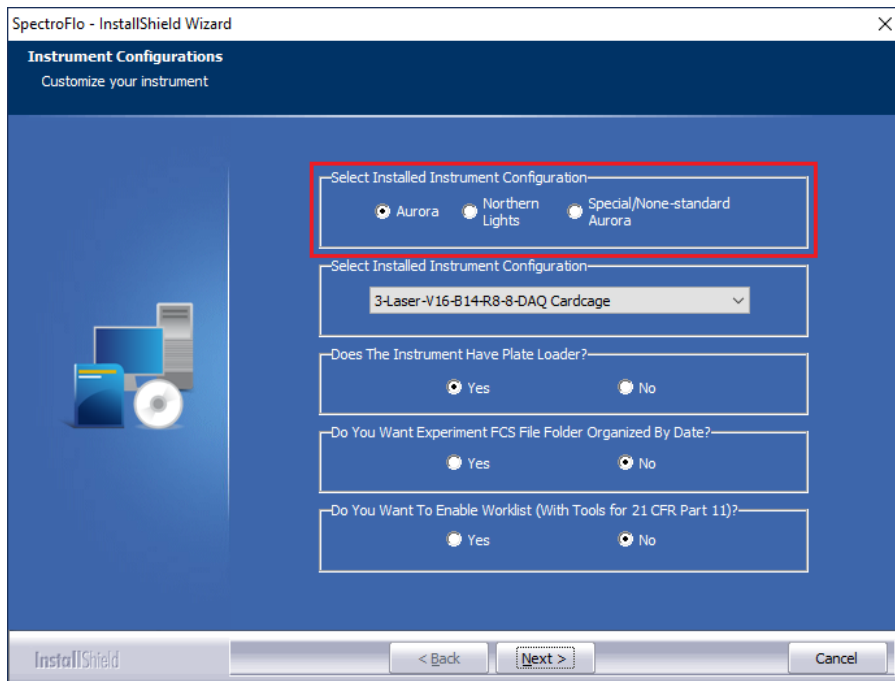
4. Select **I accept the terms of the license agreement**, and then click **Next**.



5. Click **Next** to continue.

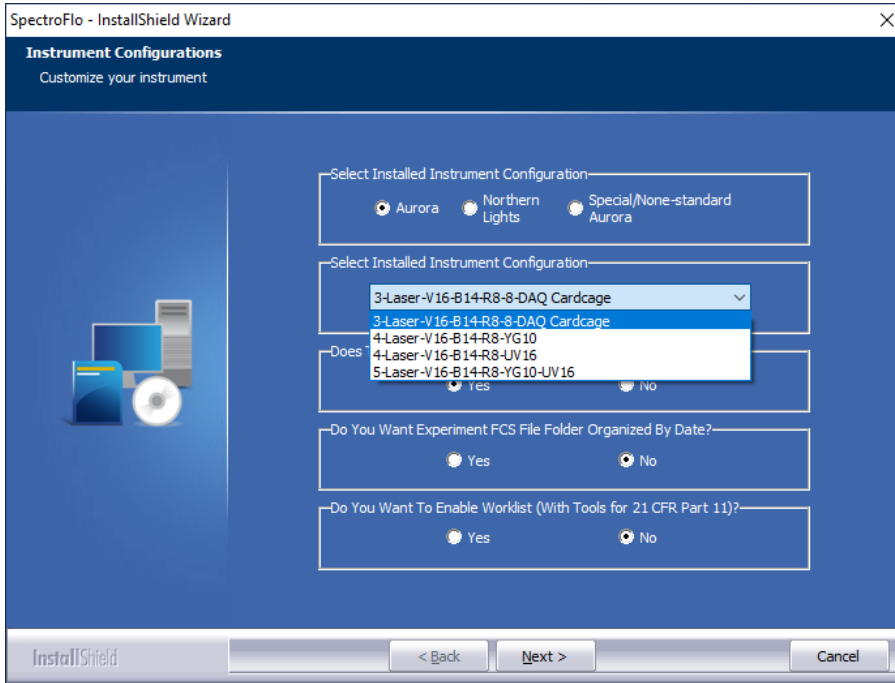


6. Select instrument type that is “Northern Lights” or “Aurora” or “Special/None-standard Aurora”.

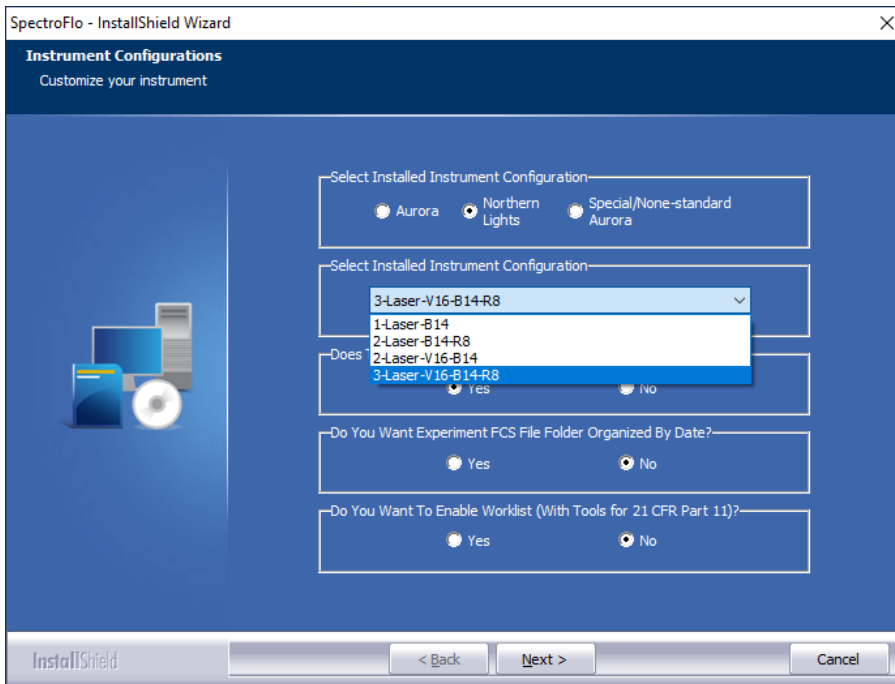


7. Select the configuration for your installed instrument.

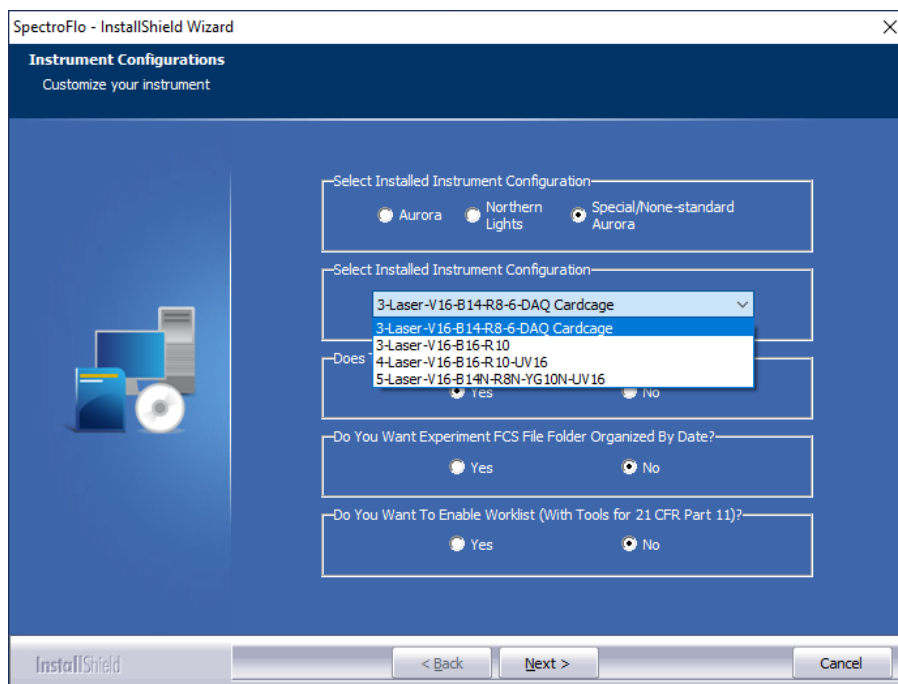
For Aurora:



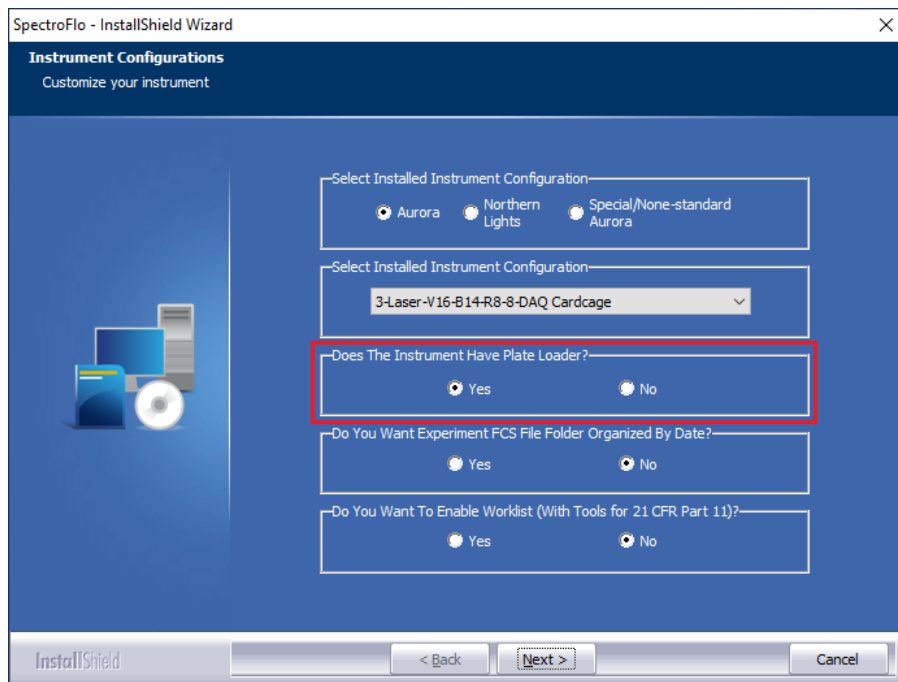
For Northern Lights:



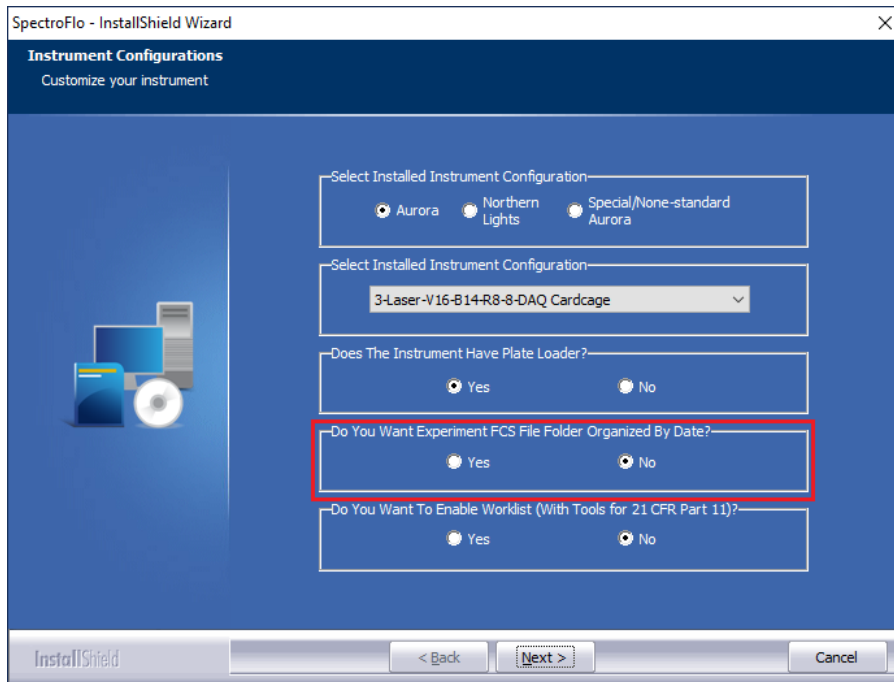
For Special/None-standard Aurora:



8. Choose “Yes” if there is a plate loader. Otherwise select “No”. Then click **Next**.



9. Choose “No” if you don’t want experiment FCS file folders automatically organized by date. Otherwise select “Yes”. Then click **Next**.



SpectroFlo - InstallShield Wizard

Instrument Configurations
Customize your instrument

Select Installed Instrument Configuration:

Aurora Northern Lights Special/None-standard Aurora

Select Installed Instrument Configuration:

3-Laser-V16-B14-R8-8-DAQ Cardcage

Does The Instrument Have Plate Loader?

Yes No

Do You Want Experiment FCS File Folder Organized By Date?

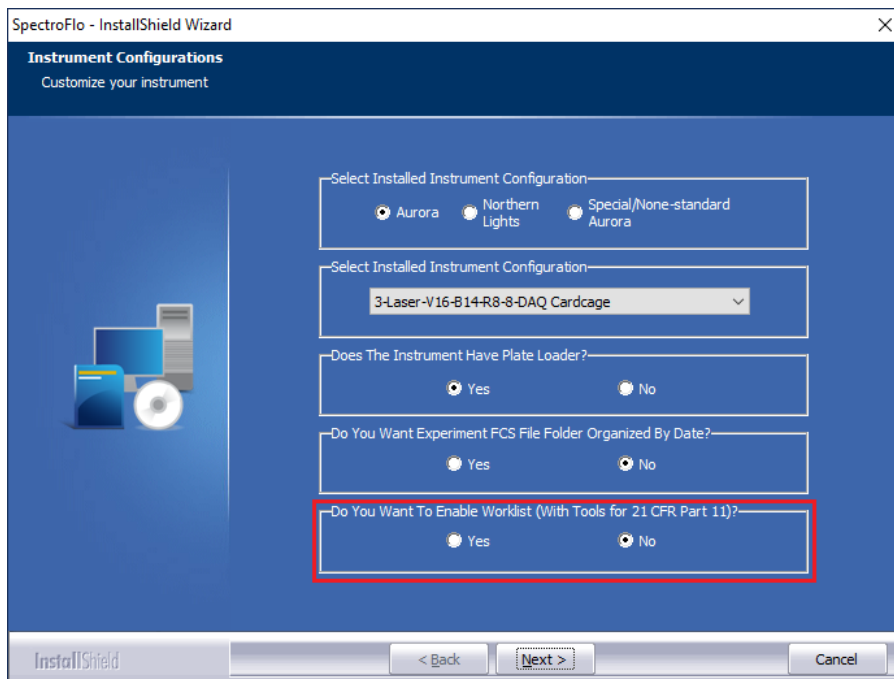
Yes No

Do You Want To Enable Worklist (With Tools for 21 CFR Part 11)?

Yes No

InstallShield < Back Next > Cancel

10. Choose “No” if you don’t want to include the worklist module. Otherwise select “Yes”. Then click **Next**.



SpectroFlo - InstallShield Wizard

Instrument Configurations
Customize your instrument

Select Installed Instrument Configuration:

Aurora Northern Lights Special/None-standard Aurora

Select Installed Instrument Configuration:

3-Laser-V16-B14-R8-8-DAQ Cardcage

Does The Instrument Have Plate Loader?

Yes No

Do You Want Experiment FCS File Folder Organized By Date?

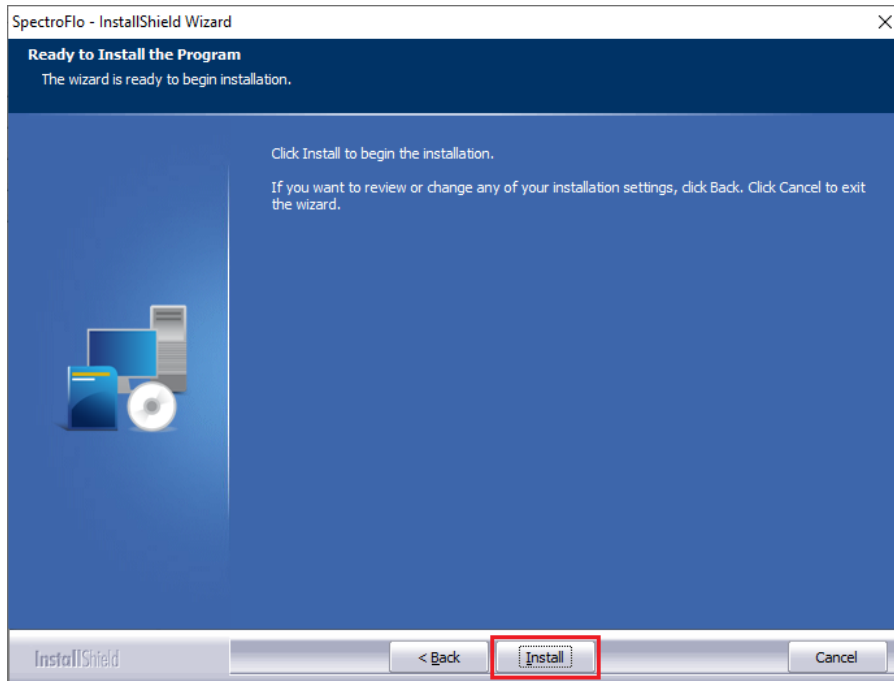
Yes No

Do You Want To Enable Worklist (With Tools for 21 CFR Part 11)?

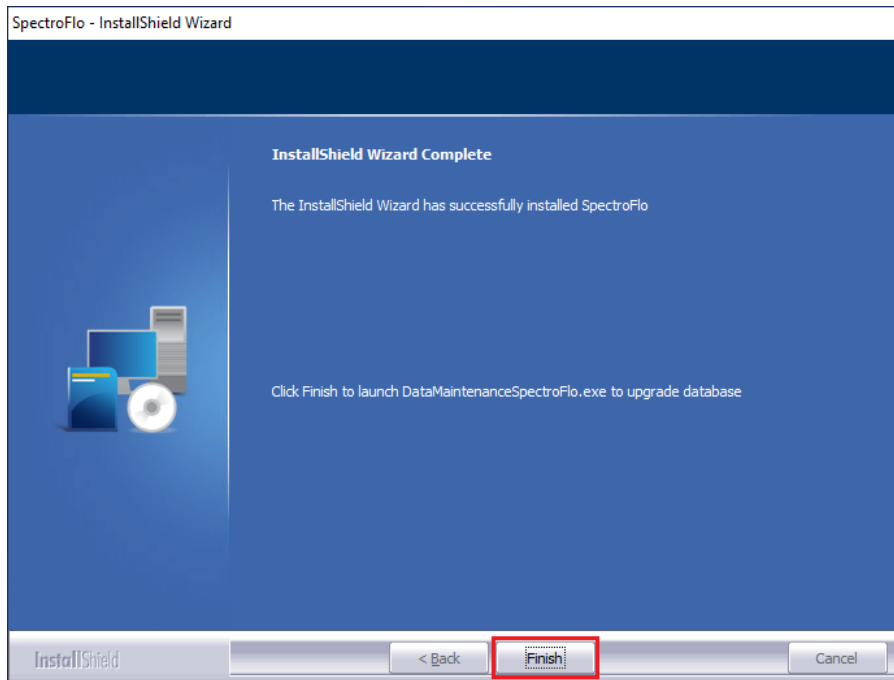
Yes No

InstallShield < Back Next > Cancel

11. Click **Install** to begin the installation.



12. Click **Finish** to finish the installation.



Installing SpectroFlo[®] on a PC Inside a Network

Step 1: Remove security policies around the devices needed to run the cytometer

Two USB ports are required. One is the port that controls the fluidics, and the other is a USB needed for data acquisition (DAQ).

1. Find the fluidics port:
 - a. Open Windows 10 or 11 Device Manager:
 - b. Open Ports: see list of Ports
 - c. Disconnect the USB cable that goes from the instrument to the computer

There is only one cable going from the instrument to the computer. That is the one you need to disconnect.

- d. As soon as you disconnect, one of the ports listed initially disappears. The port that disappears varies across systems, so, for example, it can be COM 3 or COM 6.
2. Identify the DAQ port:
 - a. In Windows 10 or 11 Device Manager, go to Universal Serial Bus Controller. Find the port labeled **Cytek Aurora DAQ**.

This is the other device you need to allow user access.

- b. Write down the port.
3. Allow user access for both devices.
4. Open SpectroFlo[®] software. Log in as Admin.
5. Turn on the instrument and wait for it to connect.

Step 2: If step 1 fails, and you still cannot connect the cytometer, try removing the computer from the network and running it as a local machine. Ideally, we would like to log into the computer as we originally did. User Name: Aurora User, Password: Welcome#1. Make sure that you log into SpectroFlo[®] as Admin.

Step 3: If step 2 fails, download the log file. Go to C:\CytekbioExport\. The file name is ApplicationLog_MM YYYY.txt. If there are several log files, please copy the latest one and send it to Cytek for assistance.

Have Questions? Comments? Want to work with us? Visit us at www.cytekbio.com.