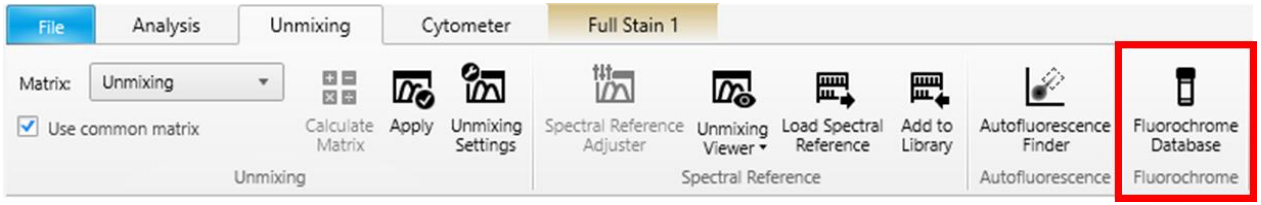
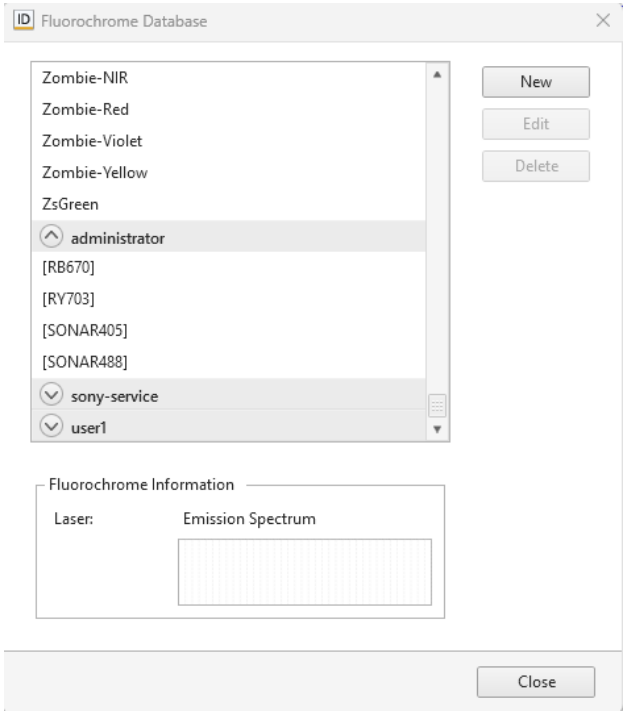
Fluorochrome Database Workflow Guide

The purpose of this workflow guide is to learn how to add new fluorochromes to the software and how to edit pre-existing fluorochromes. You will need to add a new fluorochrome if you cannot find it in the software.

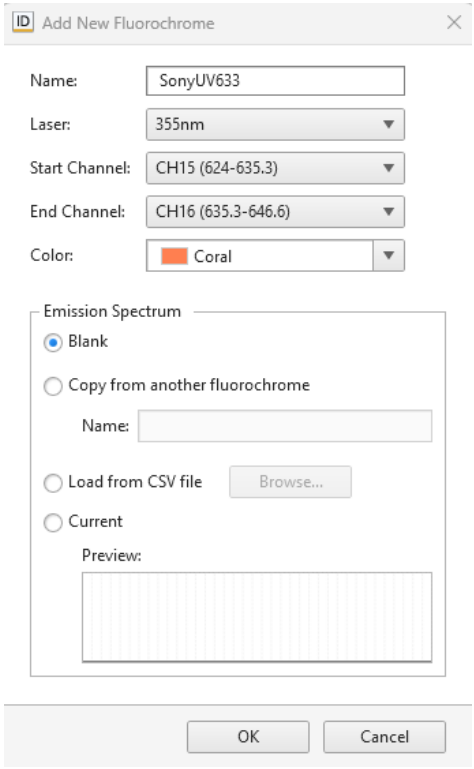
1. The Fluorochrome Database is located under the Unmixing tab all the way to the right of the menu at the top of the software.



2. The Fluorochrome Database has the Preset Fluorochromes as well as any that have been added by users. Those that have been added to the software have square brackets around the name, for example [SONAR488]. Fluorochromes that have been added to the software are categorized by the user that added them in the Fluorochrome Database.



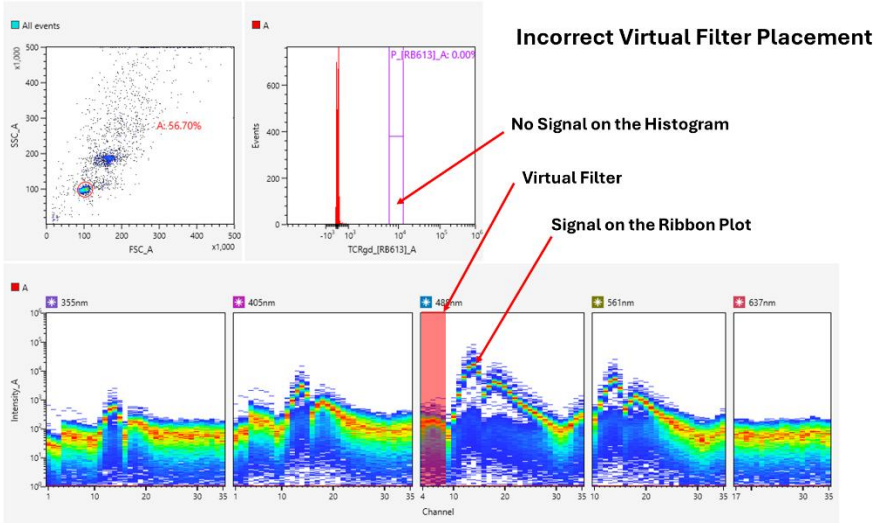
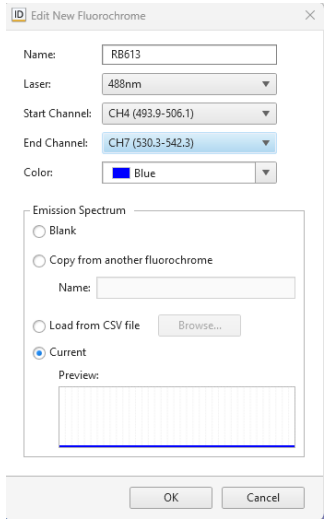
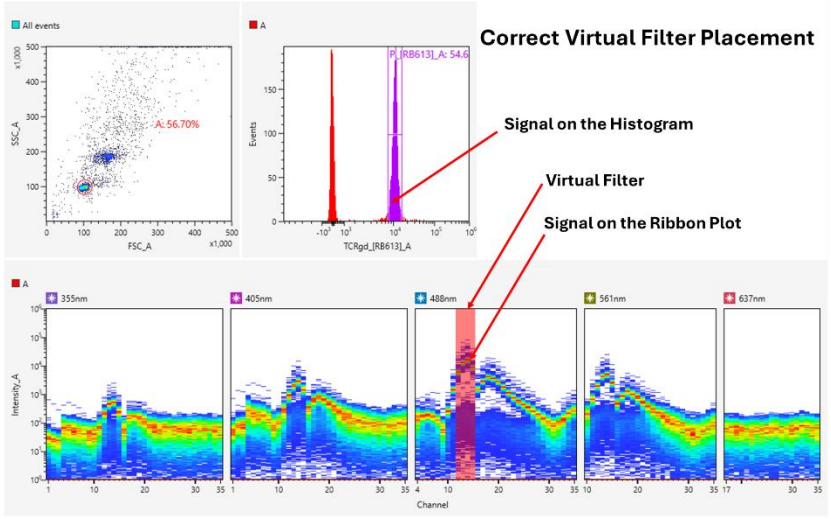
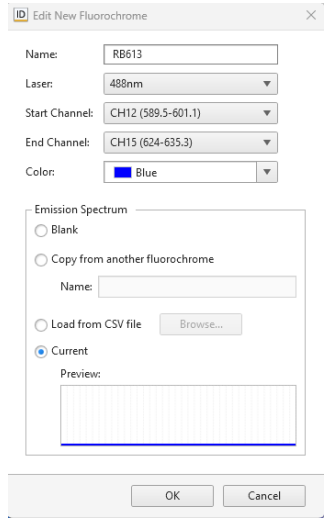
3. To add a new fluorochrome, select “New” on the top right of the Fluorochrome Database window and enter the name of the fluorochrome. Select the main excitation laser and select the 2-3 detectors beginning at the peak emission. The 2-3 detectors will form the Virtual Filter (VF), which is where the instrument will look for the peak signal from the fluorochrome. For example, if a new fluorochrome is called SonyUV633 and has an excitation/emission of 355/633, see how it would be entered as a new fluorochrome to the right. You can also select the color that the Spectral Reference will display in the Unmixing Settings and Library. You should choose a dark color as this will be displayed against a white background. You do not need to enter any information for the Emission Spectrum (this is only a preview).



4. If you enter a fluorochrome name when using the Experiment Designer that is not recognized by the software, you will be prompted with this same window from Step 3 to add a new fluorochrome. **NOTE: It is very important that you match the exact name of the fluorochrome to what is already in the software. For example, “Brilliant Violet 605” will not be recognized but “BV605” will be.**

Troubleshooting an Incorrect User Created Fluorochrome

If you do not see positive signal in the histogram of a single color control, it is possible that the Virtual Filter (VF) of that fluorochrome is incorrect, especially if it is a Custom Fluorochrome.

To edit the VF of a Custom Fluorochrome, you can use the Fluorochrome Database. Open the Fluorochrome Database, select the fluorochrome you wish to edit, and click “Edit” in the upper right corner of the window.

You can then enter the correct information for the VF by following the instructions in Step 3.