Using 2D Blank Subtraction to Remove Baseline Drifts

This procedure explains how to use Empower 2 2D Blank Subtraction to remove baseline drifts caused by gradients.

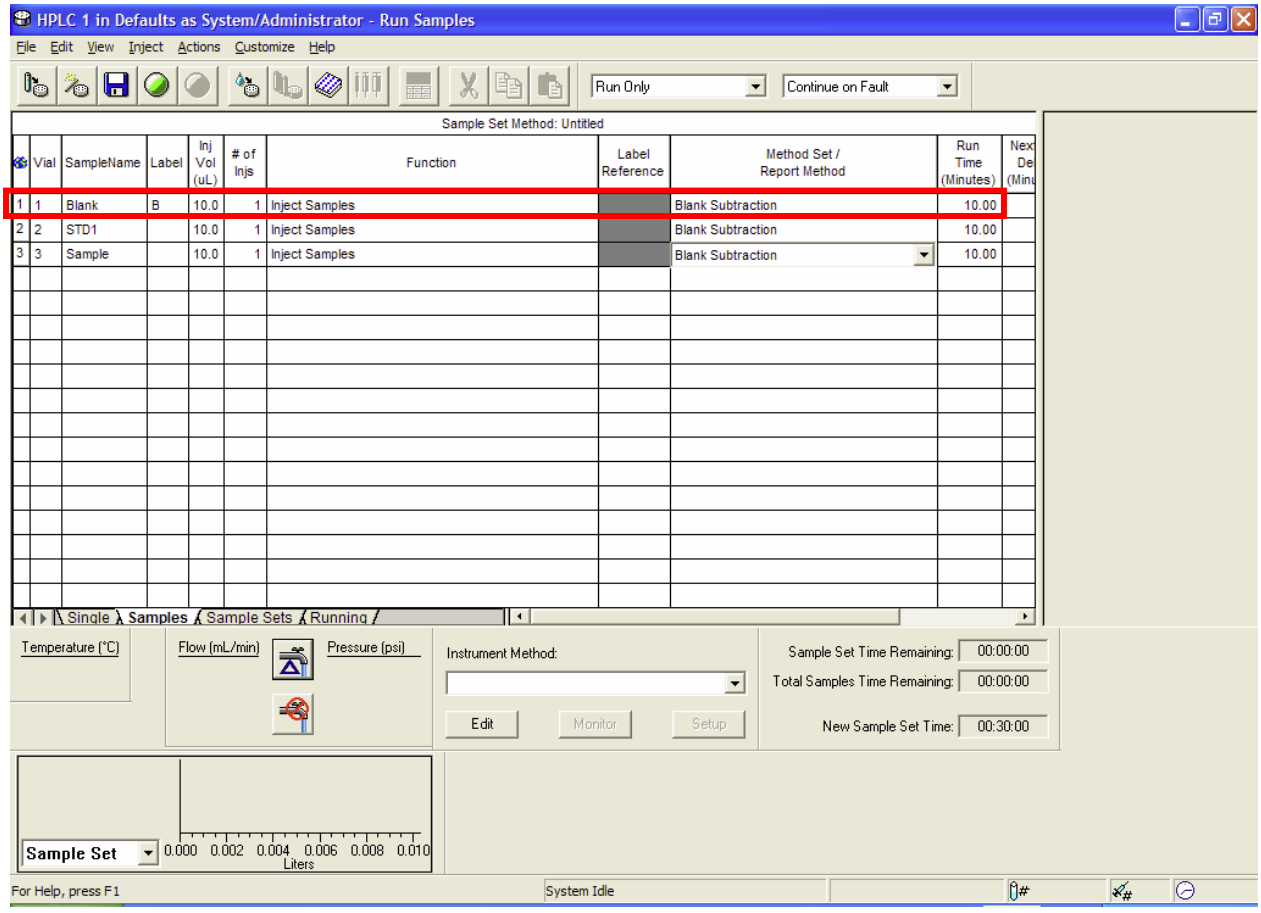
Ensure that you are using the correct recommended HPLC gradient design before beginning this procedure.

Repeat this procedure for each sample set that includes 2D Blank Subtract to account for changes in mobile phase consistency.

Creating a 2D Blank Subtraction Method Set

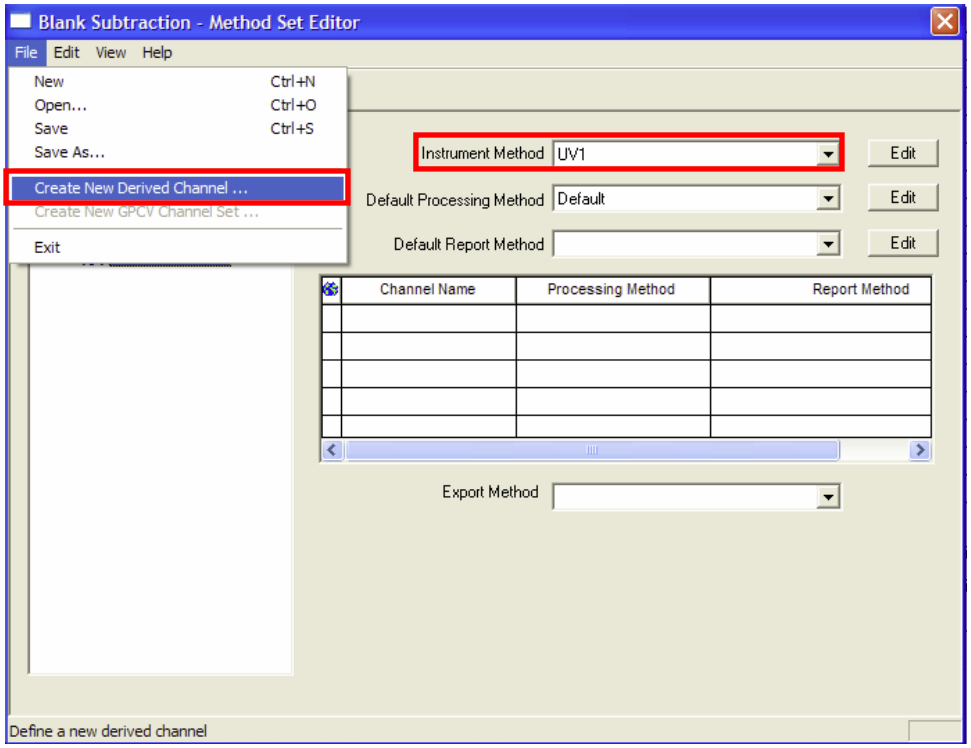
1. Include a blank injection in the acquisition sample set. Be sure to collect this injection using the same conditions (wavelength, Run Time, etc.) used for all standards and samples. Enter **0** as the Inj Vol, and **B** in the Label field to reference the Blank as the 2D reference for Blank Subtraction.

NOTE: *If you have already collected the data, select **Browse Project** and select the **Sample Set** tab. Right-click and select **Alter Sample**.*



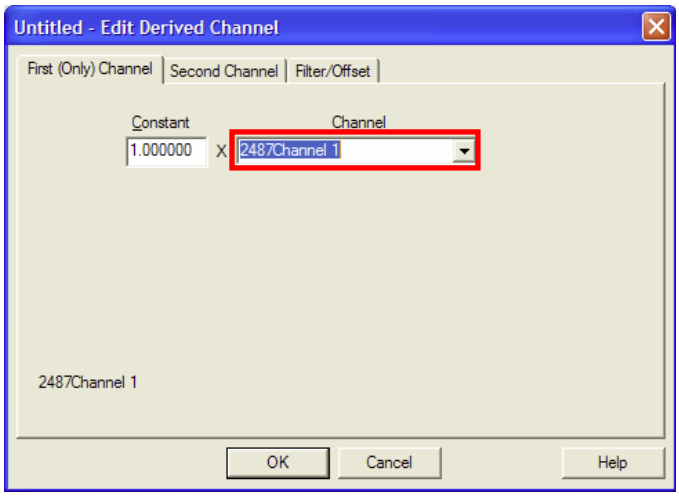
2. In the Run Samples area, select **Edit Method Set**.

3. In the Method Set Editor window, select the desired Instrument Method from the drop-down list, and then select **File > Create New Derived Channel**.

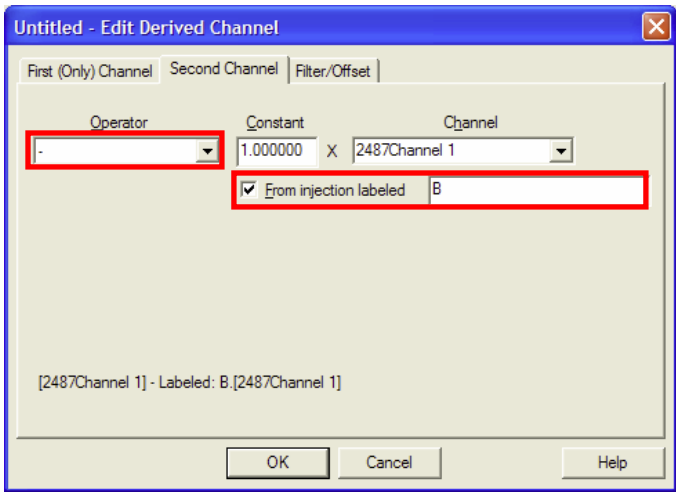


4. Select **Edit** to access the Edit Derived Channel dialog box.

5. In the First Channel tab, select the Channel from the drop-down list.



6. Click the **Second Channel** tab. Select the **Operator**, ensure that the **From injection labeled check box** is selected, enter **B** as the label, and then click **OK** to save the derived channel.



7. In the New Derived Channel dialog box, enter a name for the derived channel, and click **OK**.

8. In the Method Set Editor, select **File > Save As** and save the method set as **Blank subtraction**.

Performing 2D Subtraction of Channels in Review

1. In the Project window, select the **Channel** view.
2. Select the desired Channels, and then click the **Review** tool button.
3. In the Review window, select **File > Open Method Set**, and select the **Blank subtraction** method set.

   The method set performs blank subtraction, extracts the derived channel, and processes the extracted chromatogram.
4. Select **File > Save Result** to save the subtracted channel as a result.

Performing 2D Subtraction of Channels during Acquisition

1. In Quick Set, select the method set that includes the 2D Blank Subtraction.

2. In order for Blank Subtraction to be performed, select either **Run & Process** or **Run & Report** as the Mode.

   All Channels will be collected as raw data without the Blank Subtraction, but calculated results will be saved with the gradient shifts subtracted.

Processing Previously-Acquired Raw Data

1. In the Project window, select the **Sample Set**, **Injection**, or **Channel** view, highlight the desired lines, and click the **Process and Report** tool.

2. In the Processing area, select **Use Specified Method Set** and select the method set that includes the 2D Blank Subtraction.

3. In the Printing area, ensure that the **Print Reports** check box is selected.

4. Click **OK**. The Project window is displayed again, and the method set will perform the blank subtraction and process the derived chromatogram based on the parameters contained in the specified processing method.

NOTE: *When making any changes to a system, you should consider the applicable Standard Operating Procedures (SOPs) and complete the appropriate documentation and validation.*