Chromatography Processor Operating Instructions

Introduction

The chromatography processor software was written in-house at UW-Madison to provide an easy way to digitally collect chromatograms from our student laboratory HPLC and GC systems. The software has two main components. The acquisition component reads data from the instrument via a National Instruments USB-6008 12-bit analog-to-digital converter (ADC). The data processing component allows the user to visual the chromatogram, measure retention times and peak heights, and calculate integrals of chromatographic peaks.

Hardware

The detector's output is attached to pins 2 and 3 of the ADC. The ADC is then connected to the PC by USB. The signal is read as a differential input between pins, where pin 2 is (+) and pin 3 is (-). The maximum input voltage at either pin is +/- 1.0 volts. The ADC samples at a rate of 104 Hz. When 5000 samples have been collected, this data is transferred to the program, where it is averaged and reduced to 5 data points to be displayed on-screen. This provides a net sampling rate of 10 points per second, with each point being the average of 1000 samples from the instrument.

Acquiring Data

Open the Chromatography Processor program. The green light icon provides a toggle control of the data acquisition. The program will prompt you to save the data after the acquisition is stopped. Start the data acquisition before you inject your sample. Mark the time of your injection using the zero control on the GC or the Mark control on the LC. While the acquisition is running, the arrow keys can be used to expand or contract the y-axis: up increases sensitivity, down decreases sensitivity. Alternatively, you may use the power zoom buttons. When the last peak has eluted, stop the acquisition by toggling the green light icon and save your data.

Preparing the Chromatogram

The following tasks can be done while data is still being acquired or after acquisition has finished. Before processing our data, we must center the baseline and set our reference time. To zero the baseline, activate the Set Baseline tool, then highlight a flat part of the chromatogram with the mouse. To set a reference time, activate the Set Reference tool, then click on the "mark" in the chromatogram you made while injecting.

Finding Peak Height and Integrating Peaks

Integrate peaks by activating the Integration tool, then highlighting each individual peak with the mouse. Do this for each peak of interest. The Integration tool will display, in order, the retention time of the peak, the height of the peak relative to the baseline, and the area under the peak's curve relative to the baseline. You can remove accidental or unwanted integration regions by simply clicking them while the tool is active. Exit integration mode by again clicking the Integration tool.

Saving and Printing

You should save your file by "printing" it to PDF. Do this by clicking "Print" in the File menu. Be sure that "Print to PDF" is selected as the active printer, and then click OK. A dialog will appear allowing you to save a .pdf file with an image of your data.

To be safe, you should also re-save your data by clicking "Save" in the File menu. Note that saving the file preserves the chromatogram, but does not save any processing that you have done yourself. The file may be re-opened in Chromatography Process to re-do this processing.