DataApex
Clarity™
CHROMATOGRAPHY SOFTWARE

USER GUIDE

Clarity Software ENG

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To facilitate the orientation in the **User Guide** manual and **Clarity** chromatography station, different fonts are used throughout the manual. Meanings of these fonts are:

Open File (italics) describes the commands and names of fields in **Clarity**, parameters that can be entered into them or a window or dialog name.
WORK1 (capitals) indicates the name of the file and/or directory.
ACTIVE (capital italics) marks the state of the station or its part.
Chromatogram (blue underlined) marks clickable links referring to related chapters.

The bold text is sometimes also used for important parts of the text and the name of the **Clarity** station. Moreover, some sections are written in format other than normal text. These sections are formatted as follows:

Note: Notifies the reader of relevant information.

Caution: Warns the user of possibly dangerous or very important information.

Marks the problem statement or trouble question.

Description: Presents more detailed information on the problem, describes its causes, etc.

Solution: Marks the response to the question, presents a procedure how to remove it.

1 Installation

Topics covering installation of **Clarity** software, **Colibrick**, **Multicom**, etc. Also the connection between **Clarity** and chromatograph is explained.

1.1 Installing Clarity Software

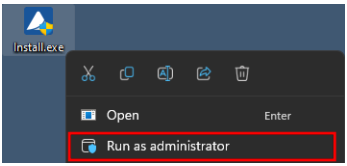
These are the basic steps you have to follow for the First installation of Clarity.

First installation of the software (since version 9.0)

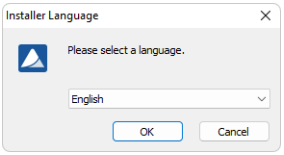
1. Install the software BEFORE connecting any hardware.

   More Info:

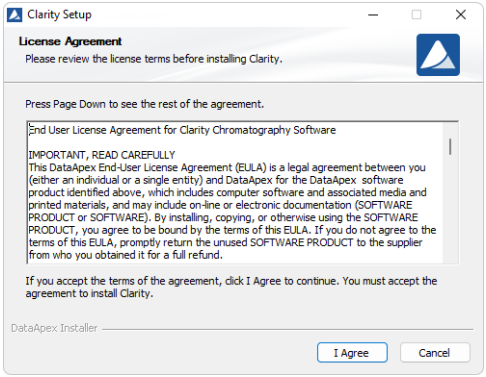
   - The software can be installed by inserting the installation USB and running install.exe or by downloading the installation from the Downloads at our website.
   - On administered systems use „*Run as Administrator*“ from intended User account. Administered systems are managed by an administrator and users using the PC may not have administrator privileges. Insufficient privileges may result in:
     - Clarity not being installed.
     - When installed from Administrator account, the installation directory may be read only for users with limited privileges and the station will be inoperable.



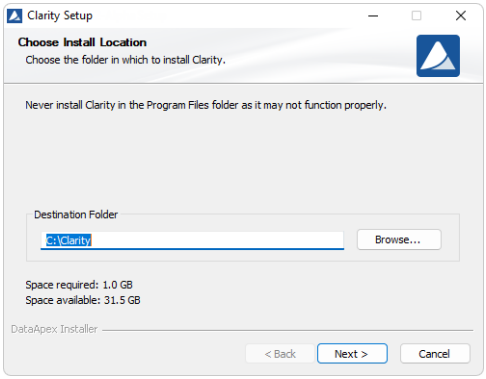
2. Select the language.



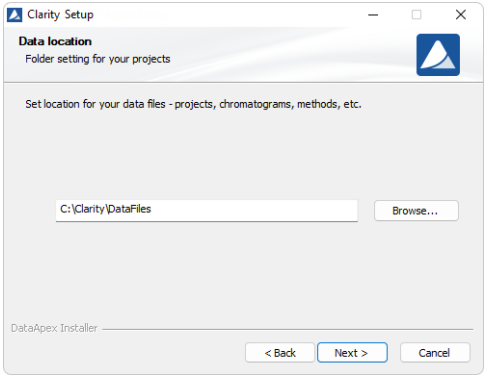
3. Confirm the License Agreement. It is possible to continue only in case you agree with the statement.



4. Choose the destination folder. The user must have Read/Write/Modify access to the installation directory.

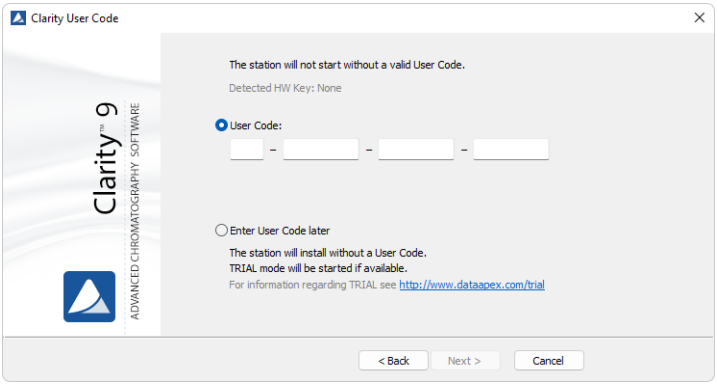


5. Set location for your data files. C:\CLARITY\DataFiles is set by default. Notice that Data location folder name cannot contain following characters / : * ? " < > | and also cannot start or end with a space and cannot end with a dot.



6. Enter the User code corresponding to your hardware key or select *Enter User Code later* to start 30-day Trial.

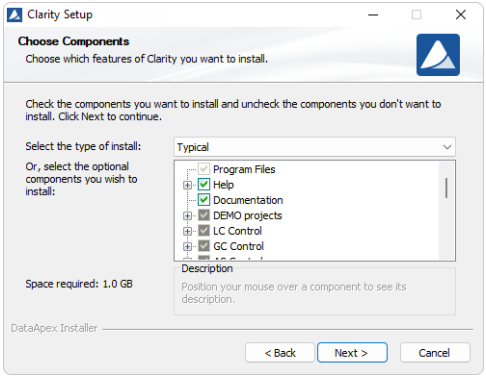
Note: The user code can be found on the back side of the card provided with the Installation USB. Alternatively, you can contact DataApex support to obtain one.



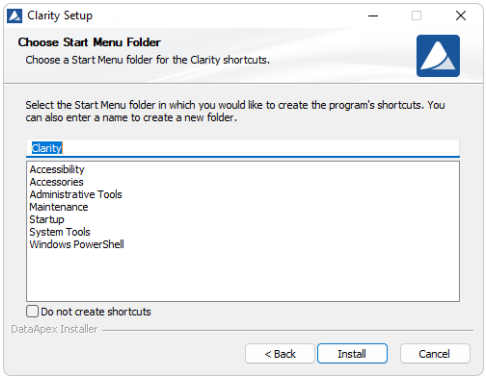
7. Select the type of installation. Make sure that control module for your device is selected to be installed.

Note: In most cases "Typical" should be selected. "Custom"/"Full" installation is necessary e.g., for Agilent and other devices controlled via ICF, DANI devices and few others.

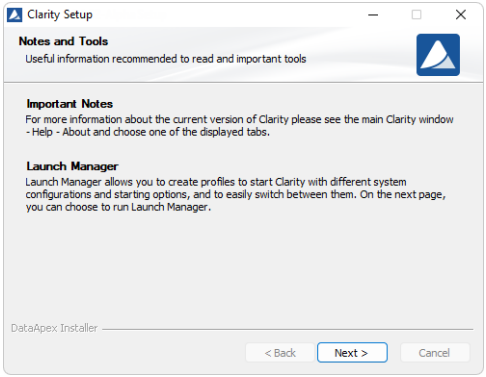
Note: List only contains names of original devices. If you can't find your device, even though it is stated as controlled, it is most likely an OEM version of different device.



8. Select the Start Menu folder for the shortcut or create a new one. After clicking *Install* installation process will start.



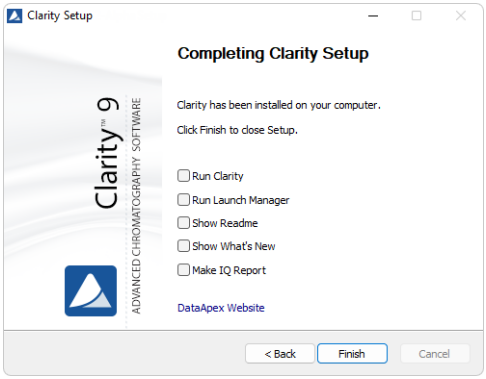
9. Wait for installation to finish. In the end you might be prompted to confirm installation of several hardware drivers.
10. When installation is finished *Notes and Tools window* will be opened.



11. Finally the last window will offer you several actions which may be executed after the installation is completed.

More Info:

- The *Launch Manager* allows you to start Clarity using different profiles that correspond to different configurations and combinations of instruments, projects, methods, etc. For example, different users or groups of users can have the chromatography station configured on the same computer.
- The *Installation Qualification - IQ* (Make IQ report option) is a procedure that confirms that the software has been installed successfully and that the files are in the correct version.



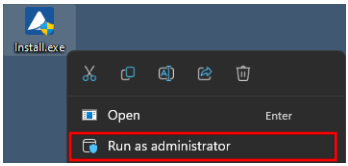
1.2 Updating Clarity Software

These are the basic steps you have to follow to update Clarity.

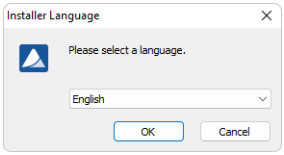
1. Check for the updates: click *Help - Check for Updates...* or download new version of the software from the Downloads at our website.
2. Run the installer.

More Info:

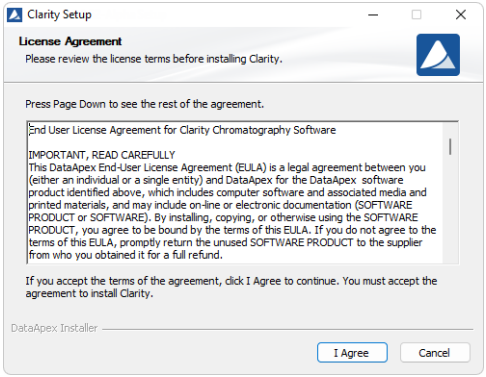
- On administered systems use „*Run as Administrator*" from intended User account. Administered systems are managed by an administrator and users using the PC may not have administrator privileges. Insufficient privileges may result in:
  - Clarity not being installed.
  - When installed from Administrator account, the installation directory may be read only for users with limited privileges and the station will be inoperable.



3. Select the language.



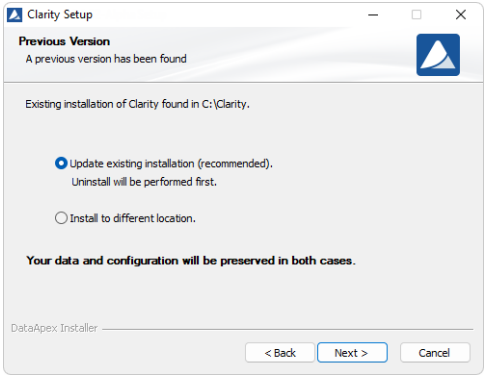
4. Confirm the License Agreement. It is possible to continue only in case you agree with the statement.



5. Previous version of Clarity will be detected. Decide whether to update or preserve this version.

Note: In both cases your data and configuration will be preserved.

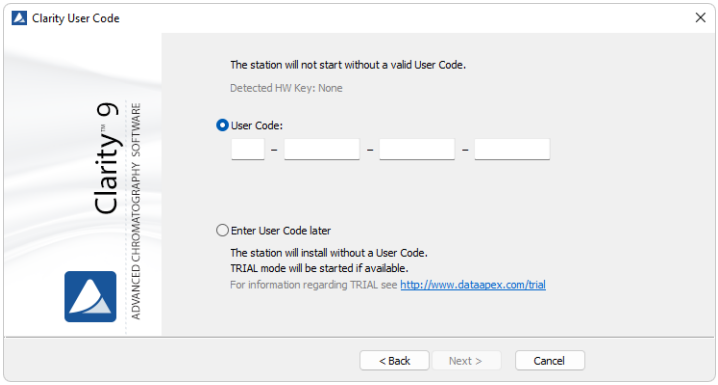
Note: Selecting *Install to different location* will result in presence of two different Clarity version at once (current version will be left as it is and new version will be installed elsewhere).



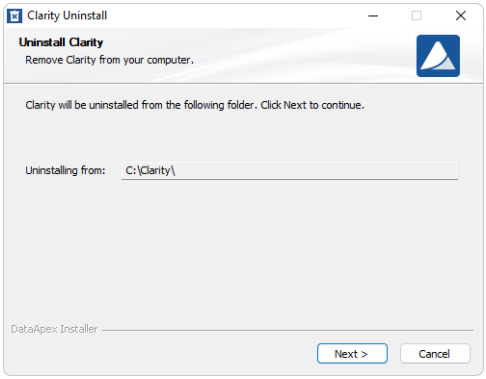
6. If you select to *Update existing installation* it is necessary to confirm the User code (code from current version will be pre-filled).

Caution: Each major version (change of the first version number) since version 9.0 requires new User Code. Make sure that you have a valid User Code for newly installed version before proceeding further. For more information see https://www.dataapex.com/upgrade.

Caution: Downgrading is not supported and might be problematic in some cases.

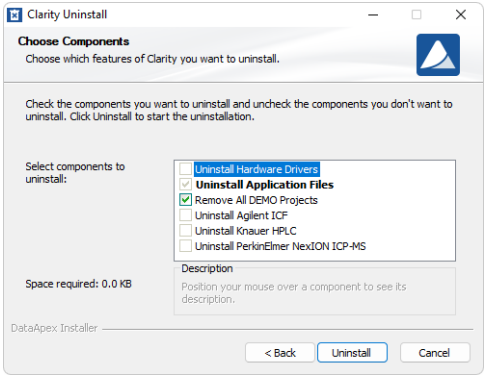


7. Clarity will proceed with uninstallation, following window appears.

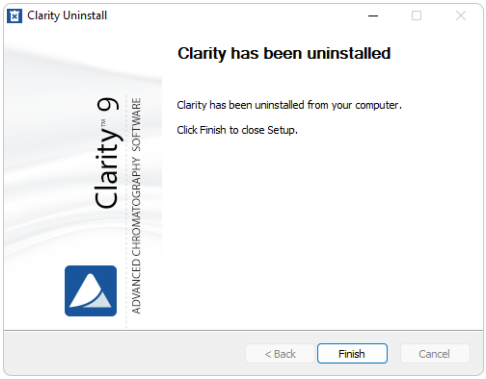


8. Continue to *Choose Components* step and click *Uninstall*.

Caution: When *Remove All DEMO Projects* is selected all files within them will be lost, including any data you saved into their folders. DEMO projects should never be used to store your data.

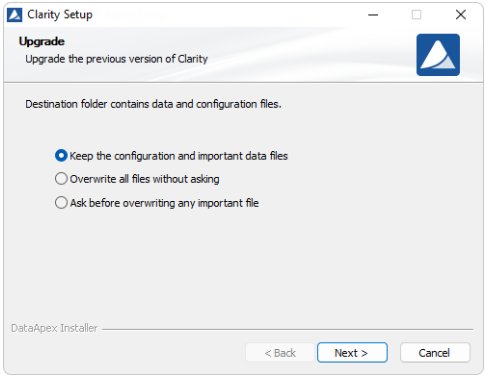


9. Wait for uninstall to finish and click *Finish* in the following window.

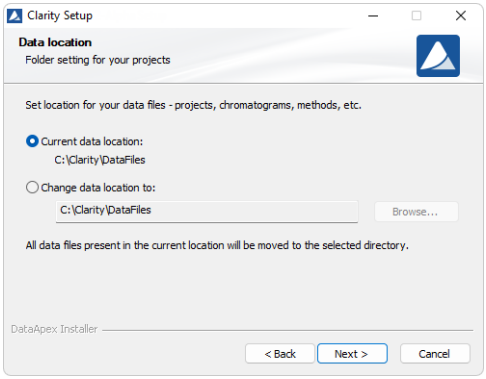


10. Choose what to do with existing data and configuration files.

Caution: It is recommended to select *Keep the configuration and important data files* to preserve all of your settings and data. Other two options might lead to losing important files and should be only used under special circumstances.



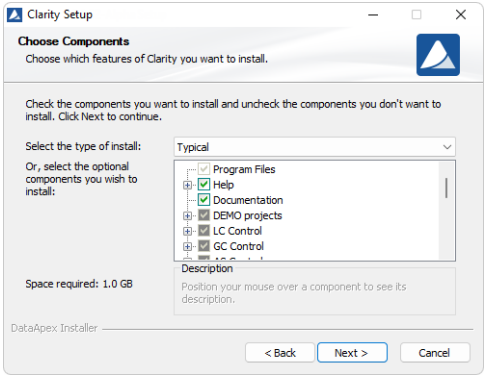
11. Set destination folder for your data files. If data location is changed all data from current version will be moved accordingly.



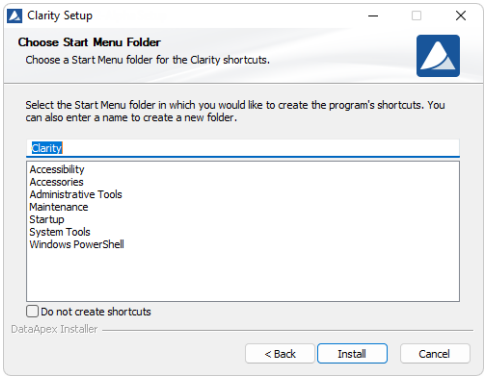
12. Select type of installation. Installation type which was used for current version will be preselected. Make sure that control module for your device is selected to be installed

Note: In most cases "Typical" should be selected. "Custom"/"Full" installation is necessary e.g., for Agilent and other devices controlled via ICF, DANI devices and few others.

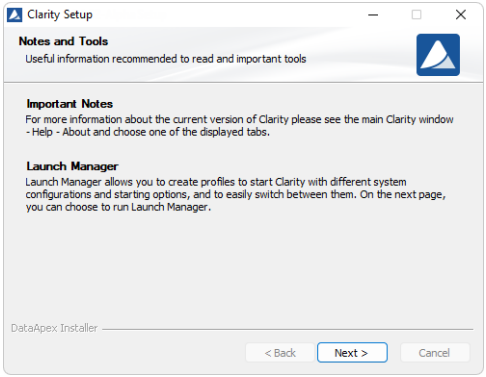
Note: List only contains names of original devices. If you can't find your device, even though it is stated as controlled, it is most likely an OEM version of different device.



13. Select the Start Menu folder for the shortcut or create a new one. After clicking *Install* installation process will start.



14. Wait for installation to finish.
15. When installation is finished *Notes and Tools window* will be opened.



16. Finally the last window will offer you several actions which may be executed after the installation is completed.

More Info:

- The *Launch Manager* allows you to start Clarity using different profiles that correspond to different configurations and combinations of instruments, projects, methods, etc. For example, different users or groups of users can have the chromatography station configured on the same computer.
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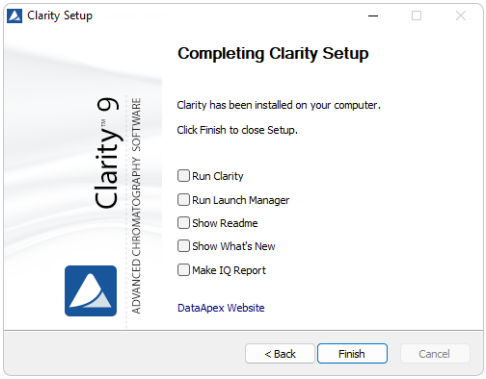


Caution: When upgrading from Clarity 6.2 or older to Clarity 7.0 or newer be aware that there is change in the installation structure - the content of the original installation folder is separated to three new subfolders BIN, CFG and DataFiles. The respective files are moved automatically during update to the new locations if you selected *Update existing installation*. In the rare cases this fails, some files may need to be moved manually.

1.3 Installing Colibrick

Colibrick is an external 24-bit A/D converter designed for acquisition of data from any chromatograph. It uses the USB communication channel and it is powered from the PC.

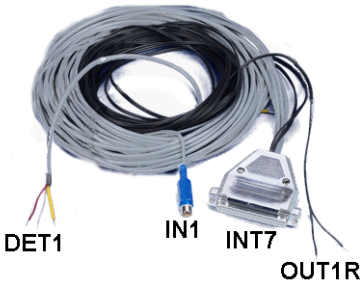
1. First install Clarity.

   More Info:

   - The driver is by default installed in *Typical* installation of Clarity. It can be found in *Hardware* section of *Choose Components* step.
   - The *Colibrick* device is identified by its S/N. If you exchange it by another one later, it will be also necessary to reconfigure it in the *Clarity - System Configuration* dialog.

2. Connect *Colibrick* to a USB port in your computer. It will be detected automatically.

3. Connect the CANNON SUB D 27-pin connector on the (INT7) cable to *Colibrick* back panel.



4. Connect the cables (Starting, Digital output and Analog Signal) to the chromatograph as explained in *Connecting a chromatograph analogue output to Clarity*.
5. Start Clarity and then add the *Colibrick* channels to specific Clarity instruments as explained in *Adding a new device*.
6. Check the LED's on the front panel to find out about the status of *Colibrick* and whether it has been installed properly.

More Info:

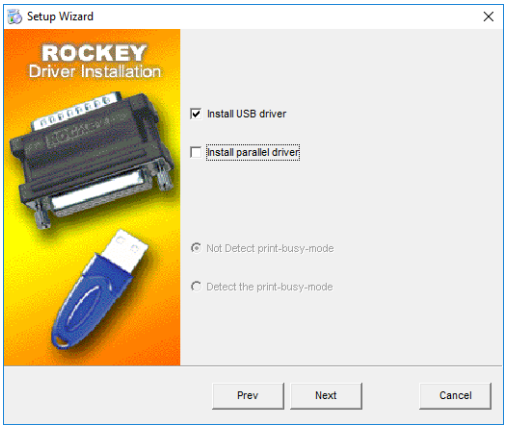
- Ready (orange) LED status Indicates correct installation.
- Data (blue) LED status Indicates connection to the chromatography data station.
- Digital Input (green) LEDs status
  - LED ON - the input status is High (logical "1") or not connected.
  - LED OFF - the input status is Low (logical "0") or connected to the ground (GND).
- Digital Output (red) LEDs status
  - LED ON - the output status is High (logical "1"), the relay contact is opened.
  - LED OFF - the output status is Low (logical "0"), the relay contact is closed.

1.4 Installing a Rockey USB dongle

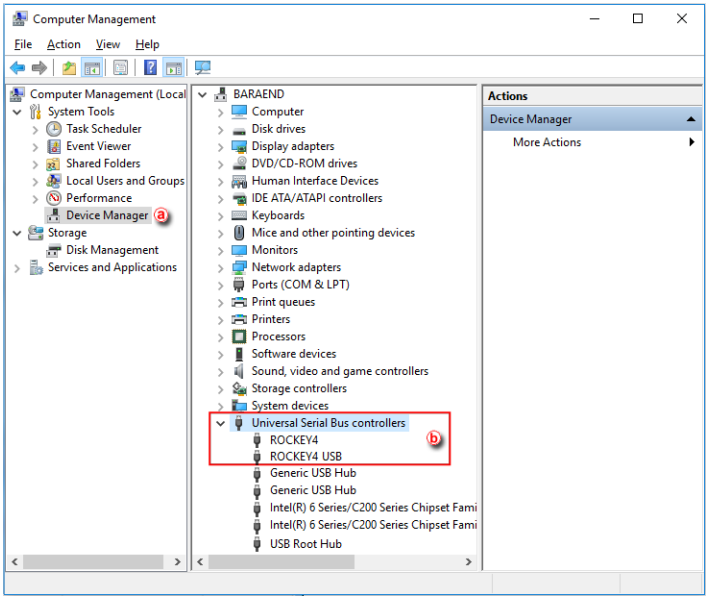
Currently supplied RkNDUSB HW keys use the HID (Human interface device) technology and do not require any drivers.

For old RkUSB keys the drivers will be installed automatically during the installation of Clarity. For this reason it is important to install Clarity before plugging the key. If the installation did not proceed as expected or you may have an old version of Windows, follow the procedure below.

1. First install Clarity.
2. Connect the USB dongle to a USB port on the computer.
3. Install the Rockey drivers by running INSTDRV.EXE in C:\CLARITY\BIN\HW_DRIVERS\ROCKEY\. The following window will appear.
4. Select the Install USB driver option and click on Next to finish the installation.



5. Verify that the driver has been installed correctly. Meaning that the *Device Manager* ⓐ has the item "*Universal Serial Bus Controllers*" - "*Rockey4 USB*" ⓑ .



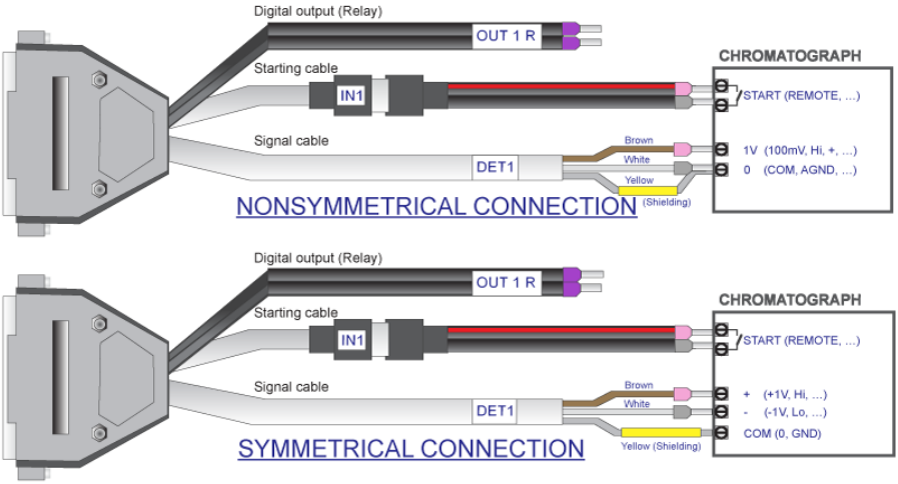
If this does not work, try the following procedure after the installation of Clarity: After connecting the dongle, Windows will detect a new Plug and Play device and the *Found New Hardware Wizard* will appear.

1. Select "*Search for a suitable driver for my device.*"
2. Select "*Specify a location*" and then select the C:\CLARITY \BIN\HW_DRIVERS\ROCKEY\folder. The rest of the installation will be carried out automatically.

Note: On Windows 10 and Windows 11, the driver for older HW keys (Rockey4USB.sys) can only be installed if Memory Integrity/Core isolation function of Windows Security is turned off. This function is turned on by default. If an incompatible driver is already installed on the computer, it cannot be turned on. But if it is enabled, the driver installation fails with Error code 39.

1.5 Connecting a chromatograph analogue output to Clarity

The Clarity Station cable (INT7) connects the station to the chromatograph and it is a set of Starting, Digital output and Analog Signal cables connected to a CANNON SUB D 27-pin connector.



1. Install your external A/D converter like *Colibrick* (refer to ***Installing Colibrick***) or an A/D converter card (refer to the specific HW manual).
2. Switch off your chromatograph.
3. Connect the bare wires to the chromatograph depending on your equipment, configuration and the following guidelines.

More Info:

- The **Signal cables** "DET 1" to "DET 4" carry the main signal from the chromatograph to the computer. The connection can be asymmetrical or symmetrical.
- The **Starting cables** "IN1" to "IN4" come in pairs, one part connected to the 27-pin connector and ending on a female RCA connector and the other with a male RCA connector and free leads for connection to a starting contact or a button for a manual start.
- The **Digital Output cables** "OUT 1R" to "OUT 4R" end on free leads and they can be used for synchronizing autosamplers.

Caution: The shielding must be connected. It works not only as the shielding, but also as the analogue ground against which measurement takes place. In the case of asymmetrical output of a detector (only two leads/terminals/pins/screws) the shielding must be connected to the white lead! No lead of the signal cable may remain unconnected.

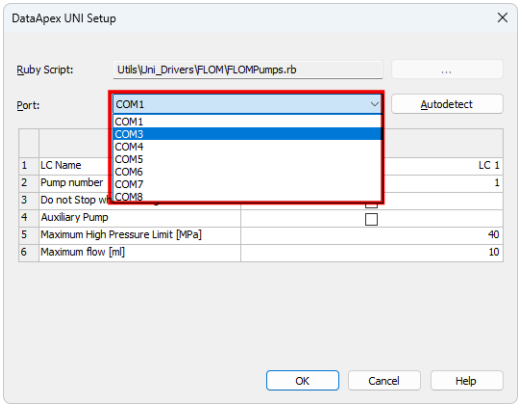
4. Connect the CANNON SUB D 27-pin connector to the A/D convertor.
5. Switch on your computer and your chromatograph.

1.6 Installing Multicom

MultiCOM is a USB to RS232 converter developed for controlling via the RS232 serial interface. It is connected to the PC via the USB port and has 6 serial 9-pin ports. It also has a free USB port for the connection of the ***USB hardware key***.

1. First install Clarity. *Multicom* driver is by default installed in *Typical* installation of Clarity. It can be found in *Utils* section of *Choose Components* step.
2. Connect *Multicom* to a USB port in your computer. It will be detected automatically. LED diodes will be turned on one by one.
3. Connect your devices to the *Multicom* RS232 ports.
4. Start Clarity and then add each device to a specific Clarity instrument as explained in ***Adding a new device***.
5. Select the appropriate port from the list during the device setup.

Note: When using functions like *Autodetect* and *Check* LED diode of selected port will blink. This can be used to find the desired port number on *Multicom*.



6. Check the LEDs on the top panel to find out about the status of *Multicom* and whether it has been installed properly.

More Info:

Green LED status:

- OFF – not connected to USB, the driver is not installed or *Multicom* is in suspend mode.
- ON (Constant) – idle state, no communication.
- BLINKING:
  - Two short consecutive blinks – only sending data from USB to the COM port.
  - Turned off twice consecutively – only receiving data from the COM port into USB.
  - Constant blinking – both sides are receiving and sending data.

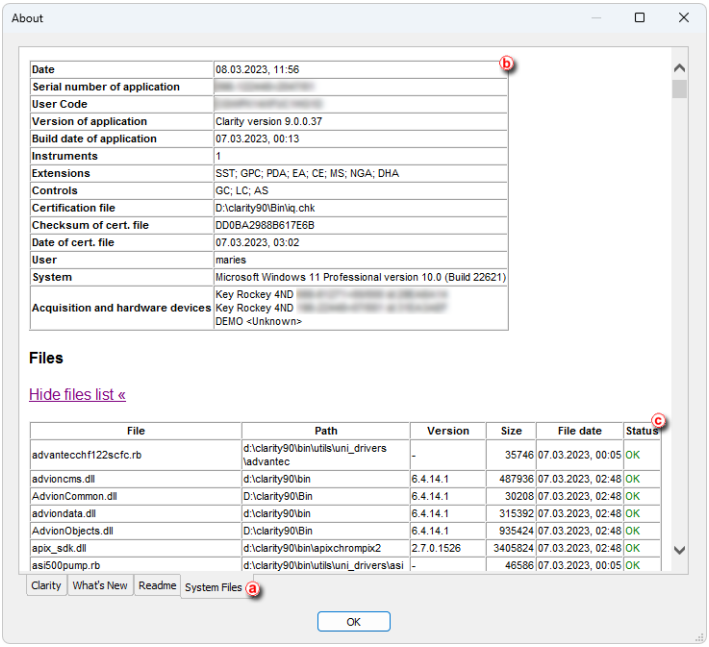
2 Configuring the Chromatography Station

Chapters covering settings in the *System Configuration* dialog.

2.1 Obtaining information about Clarity configuration

To find out information on the supported control modules and Extensions, used A/D converters and purchased Instrument licenses follow this procedure:

1. Open the *About* box: Select *Help - About…* on the *Main* window.
2. Switch to the *System Files* tab ⓐ . Note that it may take a while for Clarity to generate the report.
3. In the first table ⓑ there is info about:
    - Clarity SW version
    - Number of purchased instrument licenses
    - Extensions available
    - The allowed control modules
    - Acquisition and hardware devices
4. Go to the files table ⓒ to find information about the drivers and its status.
   If the status is other than *OK* there may be an issue with the driver. The version of the drivers developed by DataApex should be the same as that of Clarity.



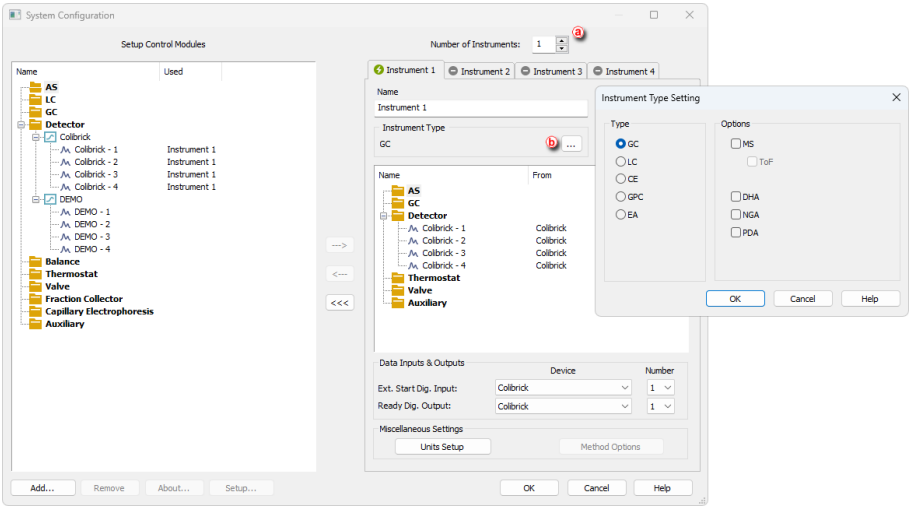
2.2 Setting the number and type of instruments

1. Enter the *System Configuration* dialog: select *System - Configuration…* on the *Main* window.
2. In the *Number of Instruments* field ⓐ you can set a number of instruments, based on bought licenses or you can use non-bought Instrument as the Offline one.

Note: The Offline instruments can be used in the same way as your standard ones except for data acquisition.

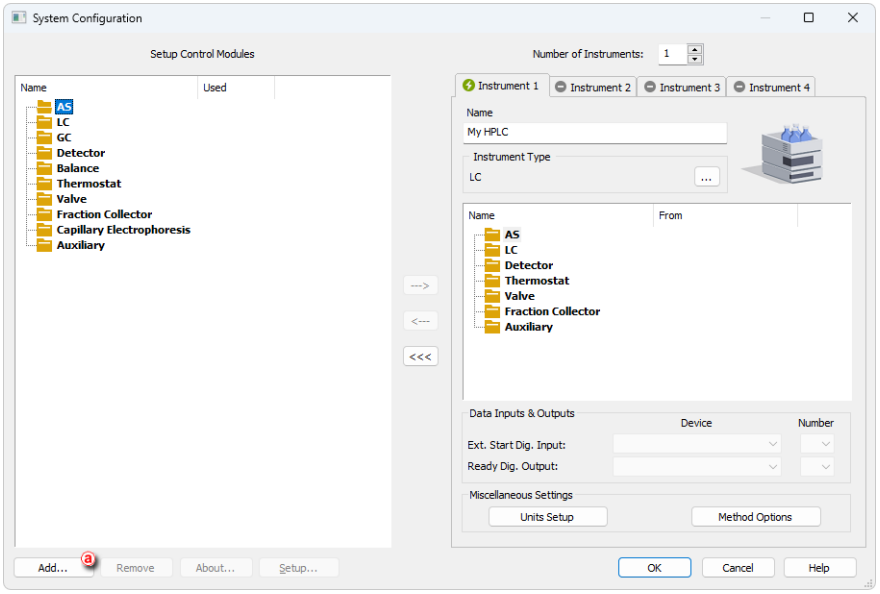
3. Set the *Instrument Type* field ⓑ according to the type of instrument you are using e.g., GC or LC.

Note: The rest of the instrument types - GPC, PDA, EA, NGA, etc., need to have the particular Extension license purchased.



2.3 Adding a new device

1. Enter the *System Configuration* dialog: select *System - Configuration…* on the *Main* window.

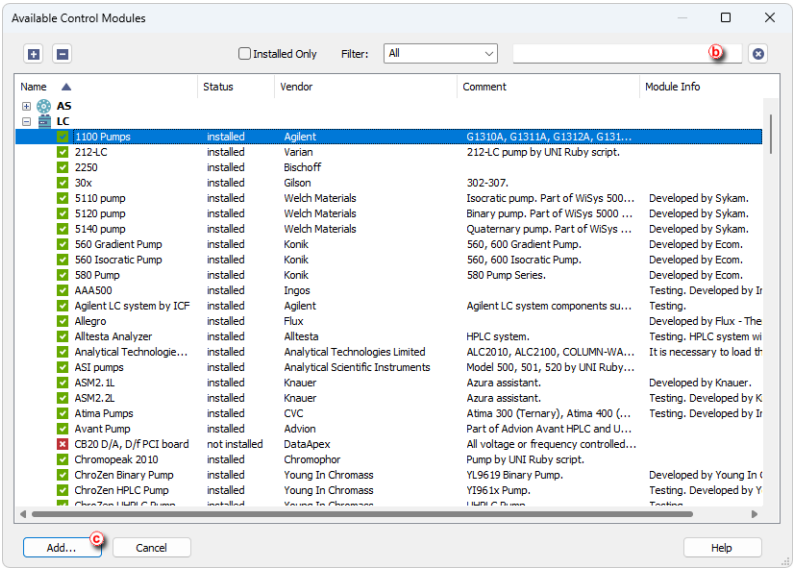


2. If the device you want to add is not in the list, click on *Add* ⓐ and the following window will open. Here you can filter the list by typing some text in the filter field

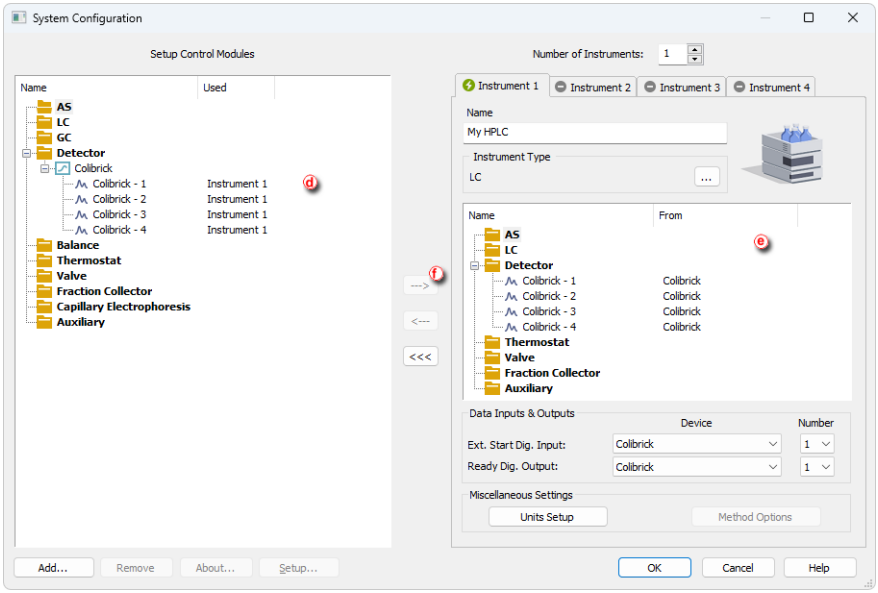
or filter by Name, Vendor, etc ⓑ .

Note: If the status of the module is *not installed*, double-click the line with this device to see why the module is not installed and how to remedy the situation.

3. Select the device and click on *Add* ⓒ or double-click the line.



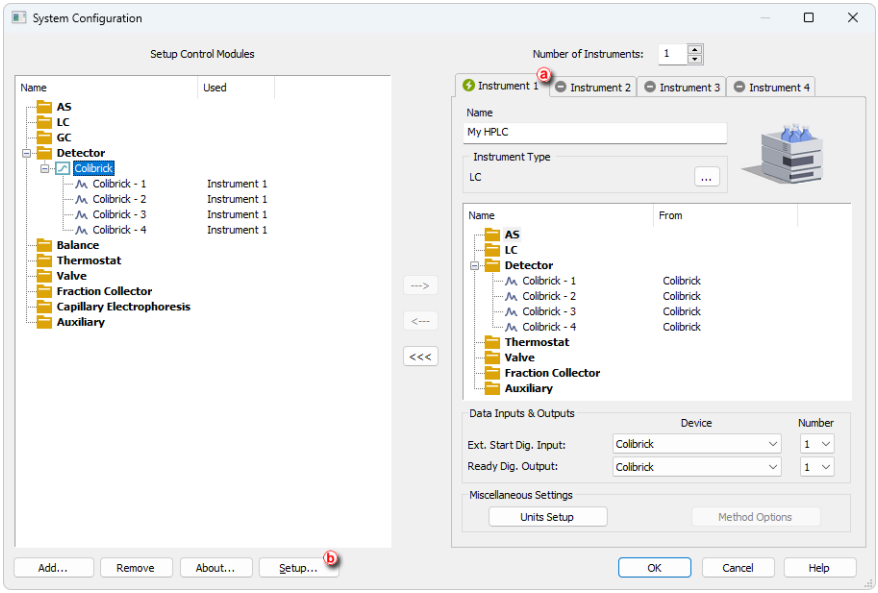
4. Add new devices to the appropriate instrument: drag and drop the device from the left pane ⓓ to the *Instrument* pane on the right ⓔ or select the device and click on ---> ⓕ .



5. If you need to help with configuring a device, go to the topic ***Configuring a device***.

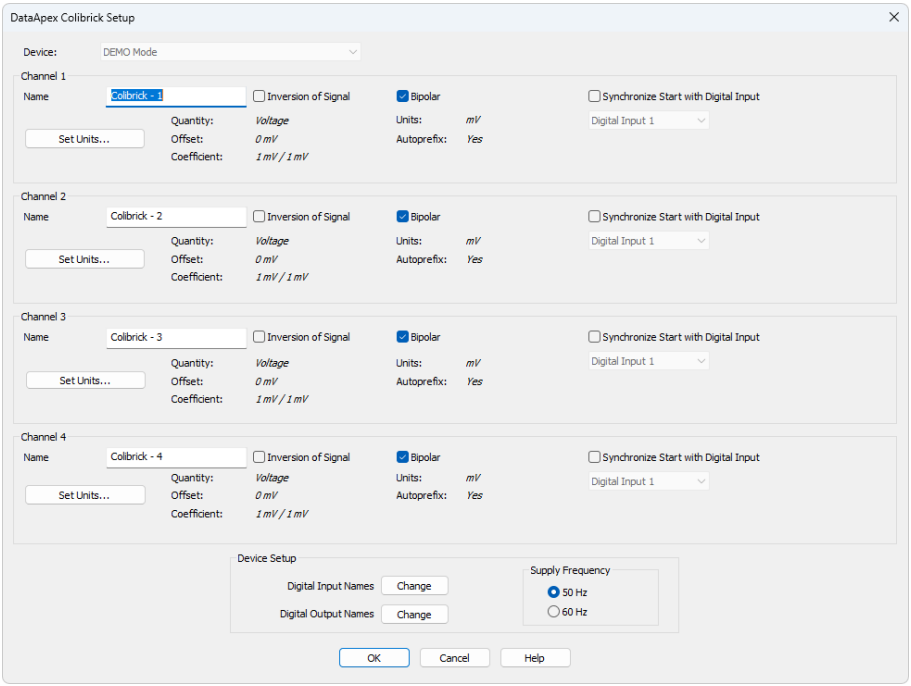
2.4 Configuring a device

1. Enter the *System Configuration* dialog: select *System - Configuration…* on the *Main* window.
2. Select the Instrument 1..4: click on the corresponding tab ⓐ .



3. To configure a device, double-click it or click the *Setup...* button ⓑ after selecting it. A device setup dialog will appear. Setup dialogs are specific to each device.

For more information on how to configure a specific device, go to the specific manual or use the *Help* button.

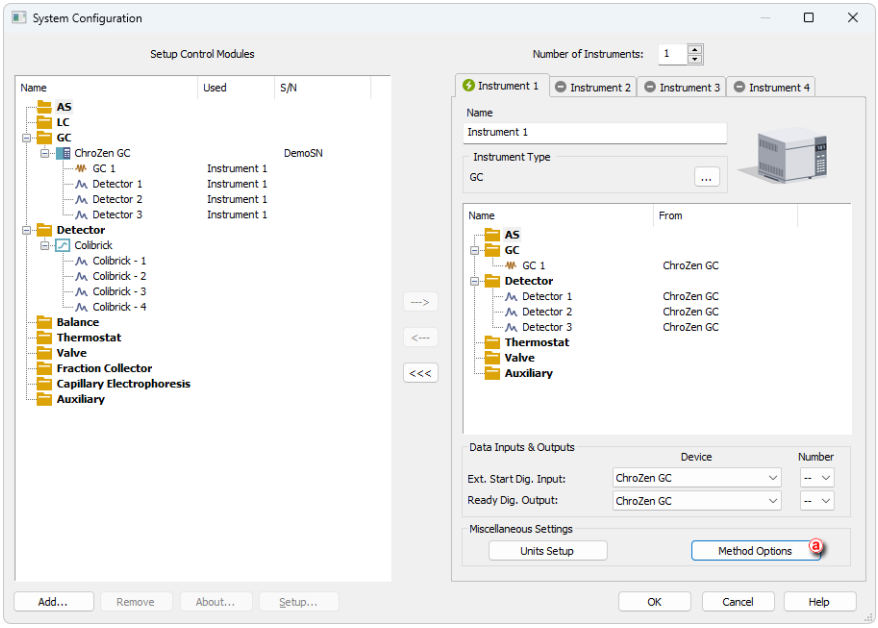


2.5 Setting the options for sending the method to the instrument

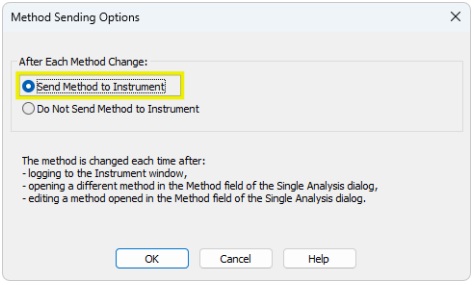
In a default Clarity installation, each time you change the method on Instrument, you need to send it manually to configured devices. Let's describe how to change that behavior so the method will be automatically sent to devices after each time you change the method.

Caution: We do not recommend to set the automatic sending of method on HPLC instruments since you may mistakenly send a method to configured pumps with wrong flow or pressure limits!

1. Enter the *System Configuration* dialog: select *System - Configuration…* on the *Main* window.
2. In the group *Miscellaneous Settings* click on *Method Options* button ⓐ .



3. Choose one of the option of the method sending.



If you select the *Send Method to Instrument* option, the method selected in Single analysis window will be sent every time you log into the instrument or this method is modified, or when the sequence is finished.

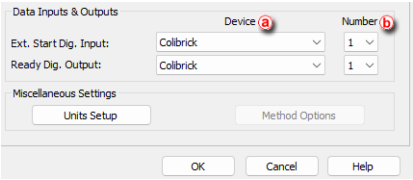
2.6 Assigning digital input and output to start acquisition

There are basically three means when Clarity decides to start the acquisition:

1. User presses Run button in Single Analysis (user probably did manual injection and notifies Clarity that its time to start)
2. Clarity is outside of run, but one of the controlled detectors starts to provide data marked as run data.

3. Clarity gets the digital input marked as START from device over:
    a. Communication line (LAN, serial (RS-232), USB/GPIB)
    b. Wire as TTL signal (this connection requires an external A/D converter or other device offering digital input to Clarity)

The last case (3) is most typical way and this chapter describes how to assign digital input and output in Clarity for the most common wiring of devices (typically an autosampler). The setting is performed in System Configuration dialog through *Ext. Start Dig. Input* and *Ready Dig. Output* functions.



2.6.1 Data Inputs & Outputs

- Settings are specific for each particular instrument.
- ⓐ : Only devices configured on the Instrument are offered in device list.
- ⓑ : Input number list allows to select input number. Available inputs are specific for particular instrument and correct number depends on actual wiring.

2.6.1.1 External Start Digital Input

The device (in most cases the sampler) provides the injection state to Clarity over its digital output. This may be a real digital output on the device or just a simulated one (Clarity does not distinguish between the two). Selecting a right *Ext. Start Dig. Input* is necessary, because Clarity watches the state of the selected input all the time and reacts on its change.

More Info:

Various devices are providing a lot of different inputs, and these may be used for other reasons, not only for analysis starting. The user needs to select the proper input which will allow the starting. For devices with virtual inputs, and in most cases also samplers with real outputs on the hardware, Start is signaled on output number 1.

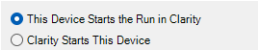
After receiving START signal from a sampler, that notifies Clarity about performed injection, Clarity sends instruction to Start Acquisition to other modules (detector, column oven etc.) of the system .

2.6.1.1.1 Possible synchronization issues

Missing start of analysis

Situation could happen when there is configured compact HPLC (without sampler) and external sampler or controlled sampling valve. The sampler or valve lets Clarity know, broadcasts the analysis start and the HPLC needs to get the information from Clarity, otherwise it will never start running.

For some chromatographs, the settings of the module and its behavior are set in the module's configuration based on the actual wiring, and it is upon the person installing Clarity and setting up the configuration to set the whole system correctly.



For correct option always consider the source of the analysis start first (sampler, sampling valve, Start button on a instrument) and ask a question "What will happen if Clarity does not signal anything? Will the instrument run anyway?"

- If **Yes** - option should be set to *This Device Starts the Run in Clarity*
- If **No** - option should be set to *Clarity Starts This Device*

2.6.1.2 Ready Digital Output

Note: Settings *Ready Dig. Output* in *System Configuration* dialog is relevant only for Active Sequence using sampler without **AS Control** module. Conversely is **not relevant** for controlled samplers used with **AS Control** module (signal over communication line), neither for Passive Sequence.

Ready Dig. Output defines the device and its specific pin through which Clarity informs other parts of the system that injection can be performed. After starting a sequence, controlled modules READY states are verified, Clarity triggers the *Ready Dig. Output* and changes its state, thus broadcasting to other modules of the system that sequence can be run. READY signal is received by the sampler that performs the injection and sends START signal. Then Clarity detects START signal and triggers again *Ready Dig. Output* and change its state, thus preventing the sampler to perform another injection.

Caution: Using the Event Table in Method Setup other actions can be configured changing the parameters of Digital Output. Note, that if the same Digital Output is modified, it may cause conflicts in synchronization. Avoid using these inputs and outputs in the Event Table.

2.6.1.3 Settings Examples

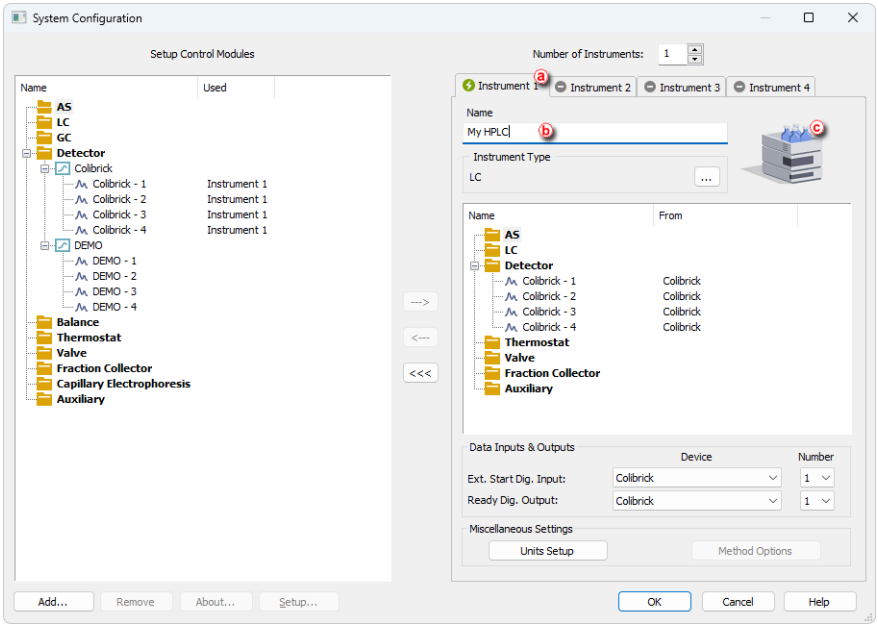
Default assignment of the *Ext. Start Dig. Input* and *Ready Dig. Output* functions can be found in the manual for the corresponding hardware.

For most common wiring of autosampler see chapter Connecting Autosamplers (AS) or respective subsections in **Getting Started manual**.

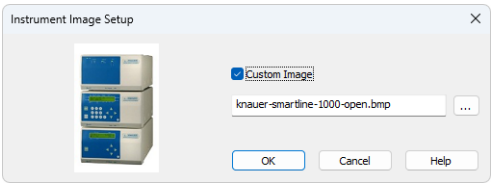
- AS + GC set - Active Sequence
- AS + LC set - Active Sequence
- AS + GC set - Passive Sequence
- AS with Clarity control module - Active Sequence + A/D converter
- AS with Clarity control module - Active Sequence + digital acquisition

2.7 Setting a custom image and a name for an instrument

1. Enter the *System Configuration* dialog: select *System - Configuration…*on the *Main* window.



2. Click on the *Instrument 1..4* tab to select the instrument ⓐ .
3. Type the name of the instrument in the *Name* field ⓑ .
4. Click on the image of the Instrument ⓒ to invoke *Instrument Image Setup* dialog.



5. Check the *Custom Image* check-box and then on ... to select your image. Click *OK* to save your changes.
6. Repeat for every Instrument you wish to change the name or the pictures.

2.8 Tablet mode

This procedure shows how to activate and use a Tablet mode. The Tablet mode represents a specific windows layout that should simulate a single-window

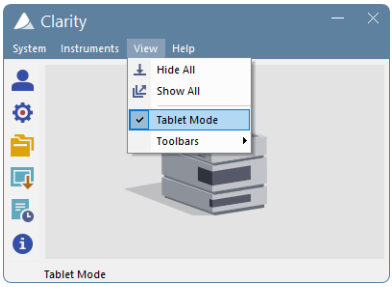
application. It is designed for devices with small displays. The optimum resolution is 1920 x 1080.

In Tablet mode, *Instrument window* is narrower than in standard layout and is positioned on the left side of the monitor, other windows open on top of each other and fill the remaining space on the monitor. Tablet mode allows the use of higher scale in Windows (up to 200 %) which improves work with the software and readability of parameters.

Enabling Tablet mode

1. In *Clarity Main* window menu, click *View* and select *Tablet Mode* item.

   Note: Switching to Tablet mode is possible only when instruments are closed.



2. After logging in to the Instrument, new windows open in tablet layout. *Instrument window* is positioned to the left side of the monitor, other windows open on top of each other and fill the remaining space on the monitor.

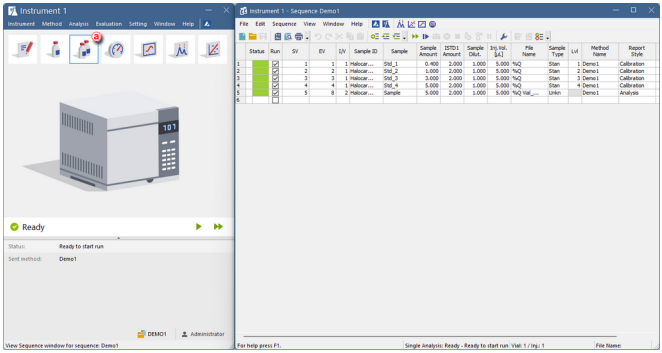
   More Info:

   Instrument window cannot be fully maximized. Its width can be expanded up to a maximum of 50% of the monitor width and is stored.

   Other windows always fill the remaining space of the display.

   Method Setup and *Single Analysis* windows are opened maximized.

   If the scale in Windows is set to more than 100%, some of the icons and analysis status line can break into a new line.



3. Icon of an active window is highlighted by a blue frame in the *Instrument window*ⓐ .

Disabling Tablet mode

1. In Clarity window menu, click *View* and select *Tablet Mode* item (active tablet mode is highlighted by a tick icon).
2. After logging in to the Clarity Instrument, new windows will open in layout used before activating the Tablet mode.

3 Configuring the User Accounts

Chapters describing how to use User Accounts in **Clarity**, how to restrict some operations for defined users and how to set shared settings for a group of users.

3.1 Configuring the User Accounts

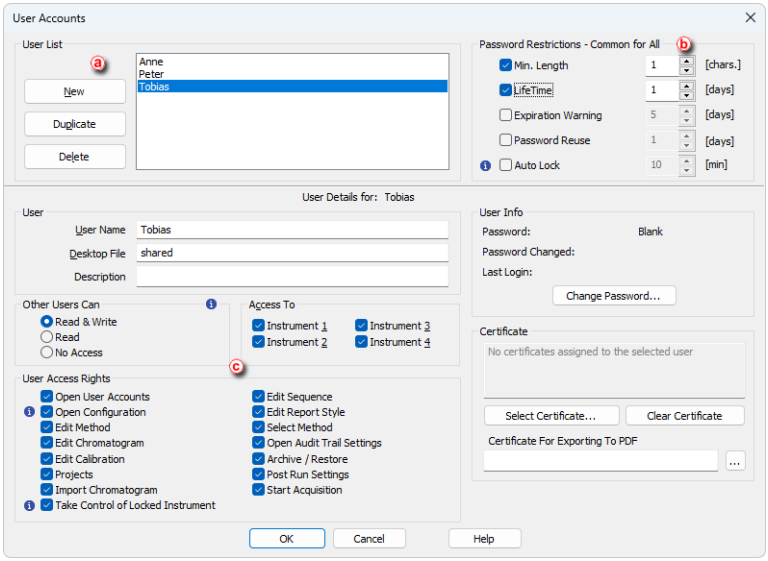
The User Accounts can be configured from the *User Accounts* dialog. It allows you to configure the settings for each user (Name, Password, Access Rights and Digital Certificates).

1. Open the *User Accounts* dialog: click on or choose *System - User Accounts*.
2. To create a new user, follow the procedure explain in Create a new user account

More Info:

- Leave blank (do not create any user) for unprotected mode - everyone working with station will share a common desktop file, and actions will be logged to audit trail under 'administator' name.
- The desktop file (user profile) stores information about the last used project, table and graph settings, etc.

3. Set the minimum password length and life time ⓑ .
4. Fill in the User Access Rights section ⓒ . More is explained in Restricting access chapter.

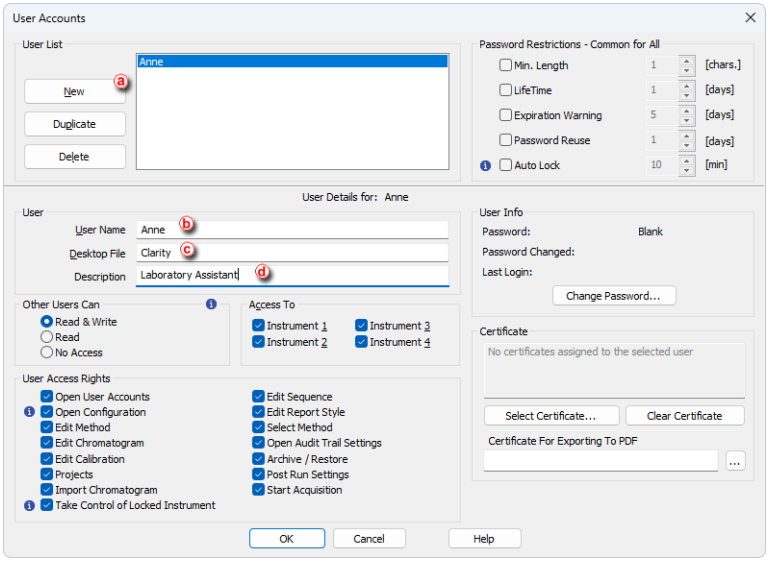


3.2 Creating a new user account

1. Open the *User Accounts* window: click on or choose *System - User Accounts*.
2. To create a new user, click the button *New* ⓐ and fill in the new User Name ⓑ .
3. Enter the desktop file name ⓒ . If you left the Desktop file field empty, the desktop file [USERNAME].DSK will be automatically created.

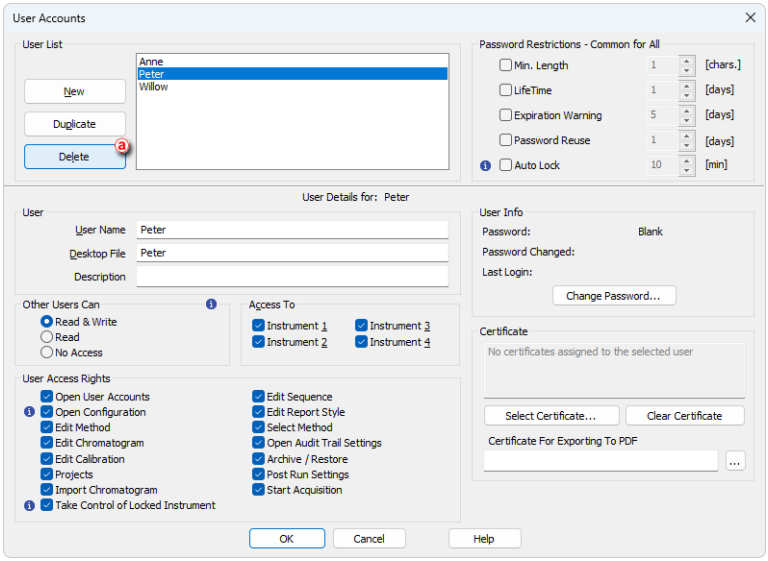
   Note: This file contains settings about the size, location and visibility of the windows as well as all the amendable Instrument parameters which are not part of system files.

4. Fill in a user description if you want to. ⓓ
5. Click *OK* to accept the changes.



3.3 Deleting a user account

1. Open the *User Accounts* window: click on or choose *System - User Accounts*.
2. To delete a user, select the user in the *User List* and then click the *Delete* button ⓐ .
3. Click the *OK* button to accept the changes.



3.4 Sharing user settings among users

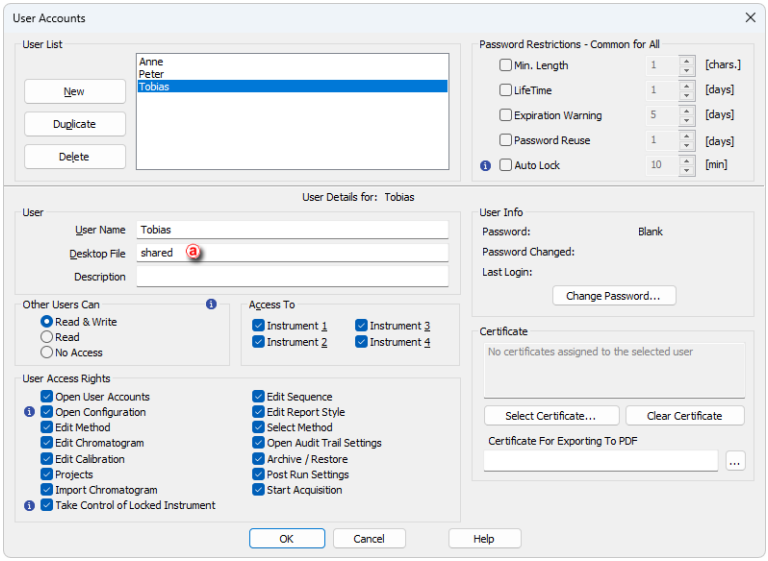
In Clarity user settings e.g., setting of *User Columns*, the width of the columns in tables, customization of the toolbars are saved in the desktop files *.DSK.

1. Open the *User Accounts* dialog: click on or choose *System - User Accounts*.
2. Select the user to have a shared desktop file.
3. Then type the name of the desktop file to be shared to the User Details section ⓐ .

More Info:

This file contains settings about the size, location and visibility of the Instrument windows as well as all the amendable *Instrument* parameters which are not part of system files (shown columns in tables, etc.).

4. Repeat for every user you want to have the shared desktop setting.
5. Click *OK* to accept the changes.

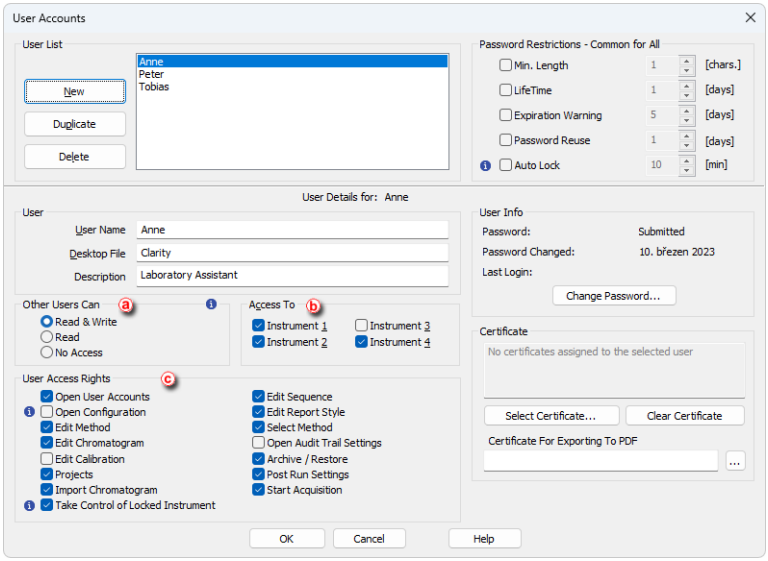


3.5 Restricting access

In Clarity it is possible to:

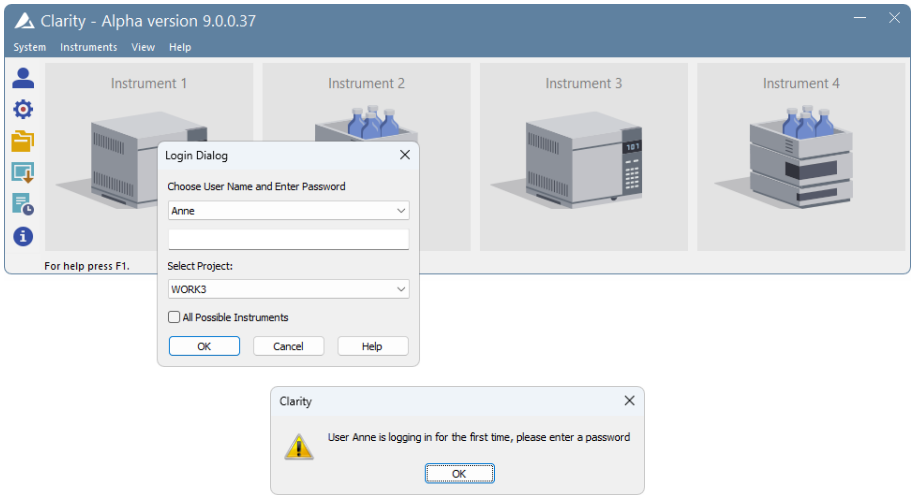
- Restrict other users' access to your files
- Restrict user's access to instruments
- Restrict user's access to Clarity procedures

1. Open the *User Accounts* dialog: click on or choose *System - User Accounts*.
2. Configure the file access rights for other users ⓐ .
3. Check the instruments the user will have access to ⓑ .
4. Check the Clarity procedures the user will have access to ⓒ .
5. Click *OK* to accept the changes.

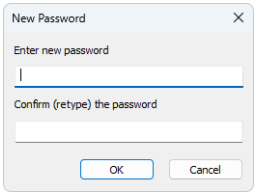


3.6 Setting a password for the first time

1. If the user has been created already and the password was left blank, then the first time you click on any of the instruments in the *Main* window, after selecting the user and clicking *OK* in the *Login Dialog*, you will be asked to enter a new password.



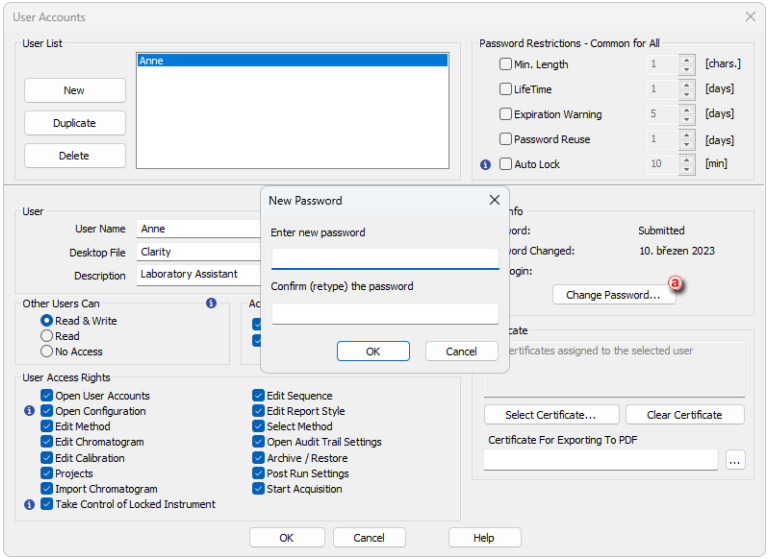
2. Type in and confirm the new password and then click *OK*.



3. Alternatively or if you are creating a new user, you can follow the same procedure as explained in section *Changing a user password* and create a new one.

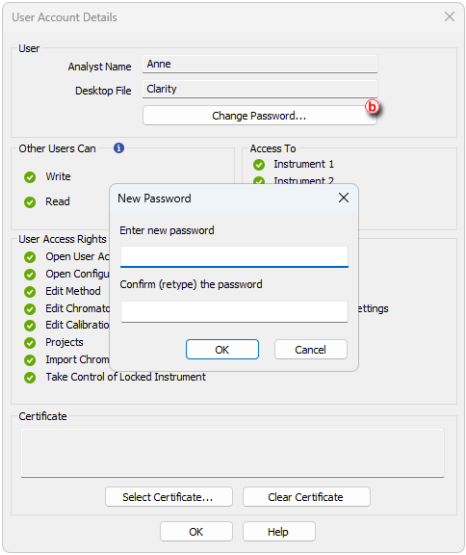
3.7 Changing a user password

1. Open the *User Accounts* window: click on or choose *System - User Accounts*.
2. Select the user in the *Users List* and then click *Change password* ⓐ .
3. Type the new password, confirm it and then click *OK*.



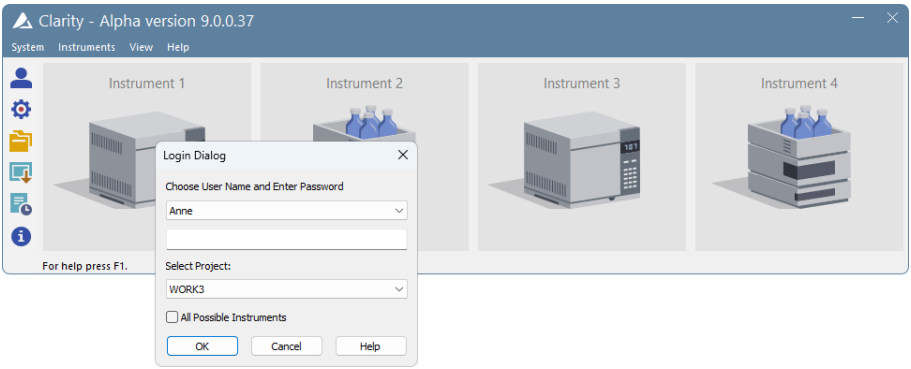
Alternatively you can change password by following these steps:

1. Open the *User Accounts Details* window by choosing *System - User Details*, select the user and enter current password.
2. Click on *Change Password* ⓑ and enter and confirm the new password.



3.8 Logging in without a password

1. Create a new user account without setting up a password as explained in section ***Creating a new user account*** or remove the password, if it was previously set, in an previously created account as explained in ***Changing a user password***.
2. Click on the *Instrument* you wish to open in the *Main* window.
3. Select the user name and click *OK* while leaving a blank password.



4 Proposed workflow for routine analysis

Below there is description of a procedure that can be used for performing routine analysis of uniform samples. Steps are described in more details in linked topics.

1. Measure typical sample.
2. Adjust the integration of the chromatogram as needed. Details are described in How to integrate chromatogram topic.
3. Save the method including this adjusted integration. Details are described in Saving the chromatogram method as a template method topic.
4. Create model calibration that will be used in calibration cloning. Details are described in Creating model calibration to use in calibration cloning topic.
5. Set the calibration cloning in the sequence, method (and potentially in the calibration). Details are described in Calibrating using clone on first recalibration or Compensating for response drift using bracketing, Improving quantification with the standard addition method.
6. Create a sequence. Do not fill the levels of standard samples and post run actions.
7. Run the sequence
8. Review the data. In case some minor adjustments are needed adjust the integration.
9. Fill in the Levels and Post Run actions to the sequence file.

   Note: If you need to sign the documents and print the report with the signature do not fill the Print actions in the sequence now. After the recalibration is performed, manually sign every chromatogram and then use Batch dialog - Post run options to print the reports with signatures.

10. In *Batch* dialog perform *Complete Processing* of the sequence. Details are described in Reprocessing whole sequence topic.
11. Now you have the sequence recalibrated, reports printed and possibly other Post Run actions performed.

5 Method Setup

Following chapters include useful tips regarding the *Method Setup* and working with method.

5.1 Setting up a method

The method essentially contains information on how the analysis will be performed, how the resulting signal will be processed and what events will be triggered and when.

Any method can be created or edited in the *Method Setup* window. To use the method for measurement it has to be sent to the instrument and for sequence measurement method has to be set at each row.

Template Method vs. Chromatogram Method

- The sent method is used as a template for new chromatograms and its contents are copied into the chromatogram file after acquisition or batch file reprocessing. You can open the different sections of the sent method setup from the icons in the *Instrument* window or with the command *Method - .....* on the toolbar.
- The changes to the chromatogram method do not affect the template method and they are performed in the lower pane of the *Chromatogram* window.
- The calibration file is linked to the template method and to the chromatogram method by its name.

Method Setup window

- The title of the *Method Setup* dialog displays a method that is currently opened. If you make any changes, the method becomes (MODIFIED).
- The upper part of the Method Setup dialog displays a set of icons with which it's possible to create a new method, open existing method, save method, save method as, open *Report Setup*, open method audit trail, send method by e-mail or open help.
- Upon pressing the OK button it will be automatically saved and *Method Setup* dialog will close.
- Tabs shown in Method Setup window are dependent on devices actually configured on the Instrument, e.g. MS Method, PDA Method, LC Gradient can be only visible when proper instrument type and device is set.

Send Method

- Only a method that is saved can be sent.
- Pressing the Send Method button will result in two actions:
  - Method will be sent to all connected hardware and thus displayed in the information table of the Instrument window - just as before.
  - Method will be set as Method for Single Analysis - you can start the single analysis form the instrument window using the ▶ icon.
- When measuring sequence, each row can have different method. It is set while creating or editing the sequence tab in the *Method Name* column.

Method Development

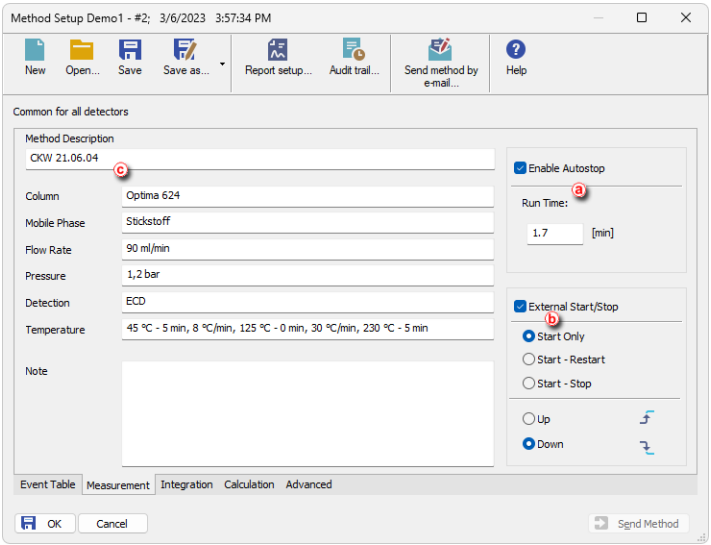
Method currently used for acquisition or present on already locked row of the sequence can't be edited, and can be viewed in read-only mode only.

When developing a method, you can configure the following settings among others:

1. **Set the measurement conditions.**

   Here you can disable or enable *Autostop* and set the run time for the analysis ⓐ as well as configure the external signal start and stop settings ⓑ (default setting *Start Only* should be used in majority of applications). *Autostop* can be set within interval 0.2 - 9999 min, such value is not influenced and does not influence any device specific time program settings on different tabs (like *LC Gradient* or *GC*). When time defined in *Autostop* passes, acquisition is finished, chromatogram is created, remaining time from time programs will be spent in so called *Control* state.

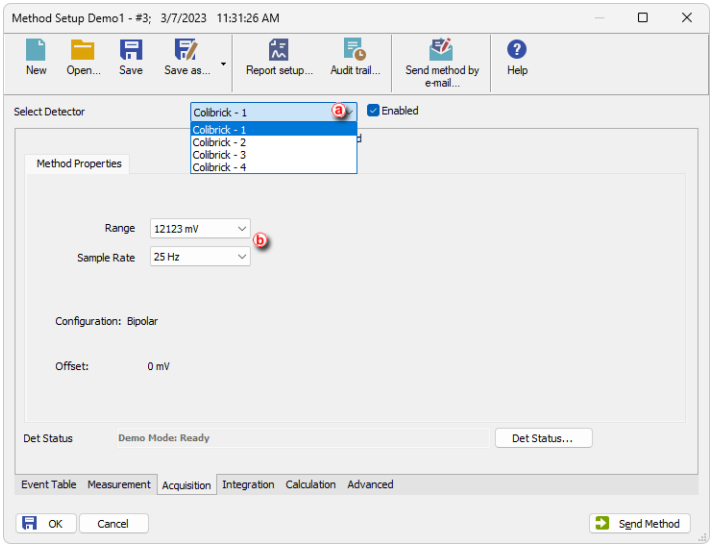
   You can also describe some parameters of the method and add a note ⓒ . Such information is purely informative and do not influence analysis in any way. They are saved into the resulting chromatogram and are showed in the *Chromatogram* window.



2. **Set the Data Acquisition parameters.**

   It allows you to select, enable/disable the detector signals ⓐ and to set measurement parameters e.g., the signal range and sample rate ⓑ . At any time at least one detector has to be *Enabled*.

   Layout of this tab, together with possible parameters to be set, are dependent on the configured devices (control modules) on the Instrument.

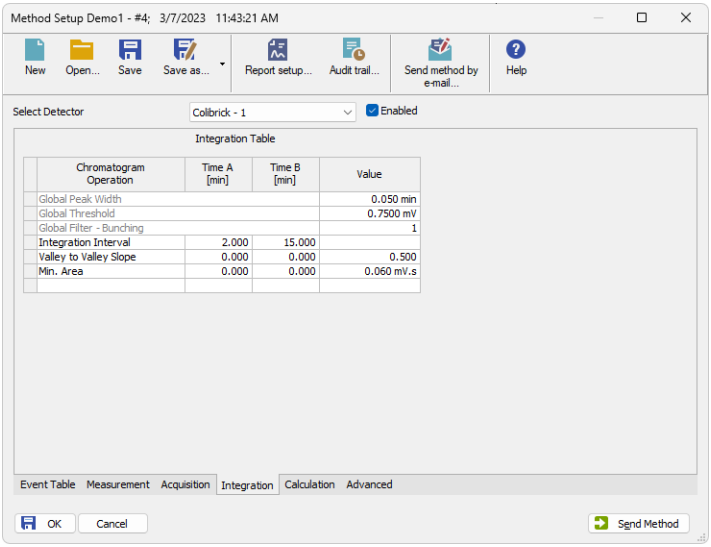


3. **Set the Signal Integration parameters.**

   When using a multi-detector configuration, there is an integration table for each detector (i.e. signal), thus every resulting signal can be integrated in a different way.

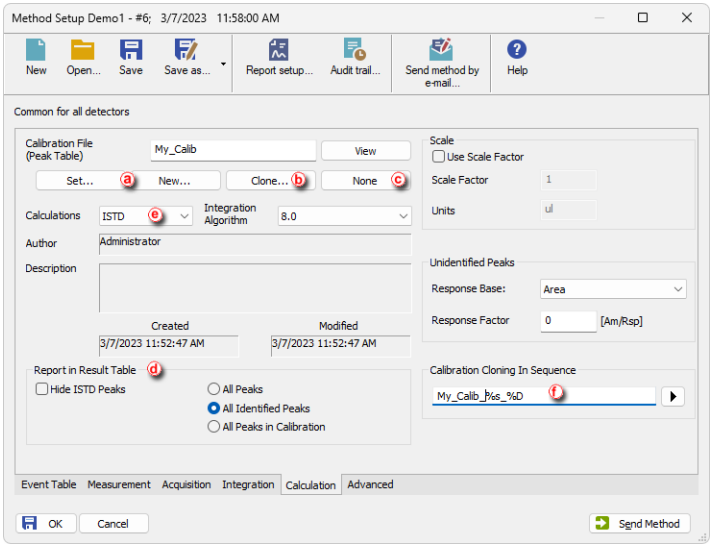
   Every resulting chromatogram acquired with such method will be integrated automatically accordingly the integration tab.

   If you aren't able to edit tab parameters the current detector probably isn't *Enabled*.



Note: Integration parameters are often modified manually after the first acquired chromatogram directly in the Chromatogram window. Then you can manually rewrite them into any method in the Method Setup dialog or you can use the *Method - Save as Template* menu command in the *Chromatogram* window to easily copy them into your method. For more details see the chapter **Saving the chromatogram method as a template method** on pg. **73**..

4. **Set the calculation options.**
   - You can create *New...* calibration file or *Set...* the one ***created previously*** ⓐ and configure the different settings related to it.
   - When calibration file is set, resulting chromatograms will be calibrated according to it and calibration standards will be used to automatically re-calibrate the attached calibration file.
   - *Clone...* ⓑ button will create a copy of a current calibration file. In the following *Save As* dialog you can select the name and location of the new calibration file. The copy will then be linked to the method.
   - If it is desired to have no calibration in the method you can use the *None* button ⓒ .
   - In this tab you can also choose how you want the results to appear in the results ⓓ and set calculation type ⓔ .
   - Name of calibration clone ⓕ must be set here to use calibration cloning in sequences, refer to chapter "Calibrating using clone on first recalibration" on page 95.



5. **Configure the settings in the additional tabs.**

   Method Setup also has tabs specific to currently configured devices and *Event Table* tab, used for configuring what events will be triggered and when e.g., shutting down Clarity after the sequence is finished.

   However, there are some limitations:

   - Event Table is for specific purposes, not needed for standard analyses.
   - Does not have control against infinite loops.
   - Must be filled per row, Fill Down/Fill Series can't be used in Event Table.

Note: For more information on any of the settings above, press F1 in such Clarity window to go the corresponding section of the Help or use the **Clarity Reference Guide**.

5.2 Saving the chromatogram method as a template method

Each chromatogram contains saved copy of the method used for acquisition of such chromatogram. However, changes done in *Chromatogram* window (e.g. *Description, Integration Parameters, linked Calibration File*...) are not updated in the original method used for acquisition nor processing. To amend your method accordingly, you can create template method from the method in chromatogram, which is useful after optimizing integration parameters.

1. Open the chromatogram with optimized method.
2. Select the *Method - Save as Template…* to save the method (including the changes made by you) from the current chromatogram as a new method, or for example overwrite (update) your acquisition method.

Note: It is not possible to overwrite any method that is currently in use, for example a method opened on any Instrument.

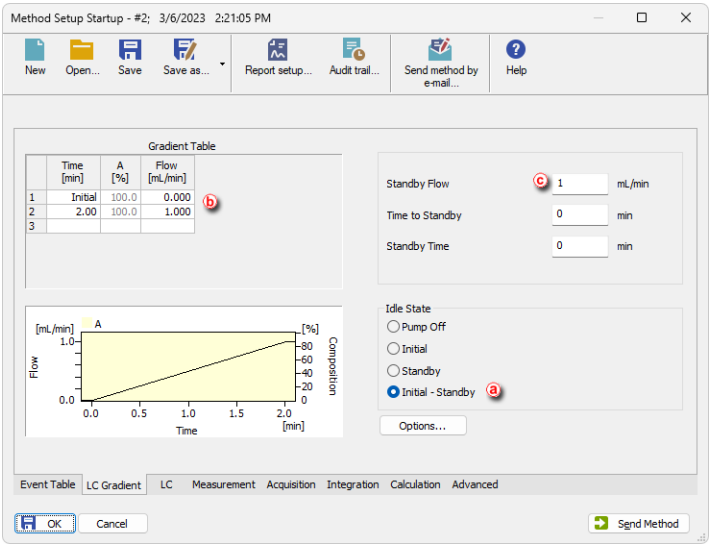
5.3 Setting up a slow flow rate increase and decrease on your LC pump

You can protect your chromatograph column from sudden pressure changes by setting up a *Sequence* together with three different methods, *Startup*, *Shutdown* and *Analysis* to ensure a slow flow rate increase and decrease on your LC pump. This procedure is mainly recommended for pumps controlled by Zebrick D/A converter or UNI Ruby scripts due to less strict pressure limit controls in these control types.

- Set the *Instrument Method Sending* option to "Do not send Method to Instrument", opposite to the topic ***Automatic sending of a method to an instrument after each change***.

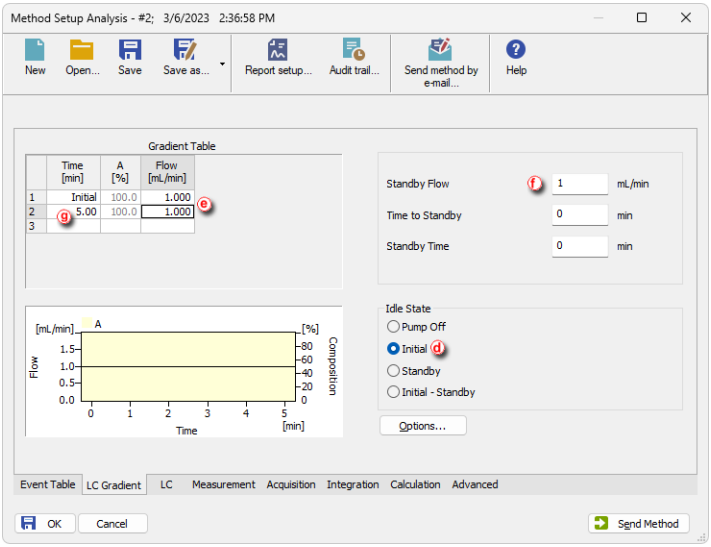
Startup Method

1. From the *Instrument* window open the *Method Setup* window using the *Method - Method Setup* command or the *Method Setup*  icon.
2. Create *New Method* and *Save* it under the name *Startup*.
3. Navigate to the *Method Setup - LC Gradient* tab.
4. Set the *Idle State* to *Initial - Standby* ⓐ .
5. In the *Gradient Table*, ⓑ set the *Initial Flow* to 0 and on the second row set the *Flow* to its *Standby Flow* value ⓒ and the *Time* to obtain the appropriate flow rate increase for your column.
6. Click OK to accept the changes.



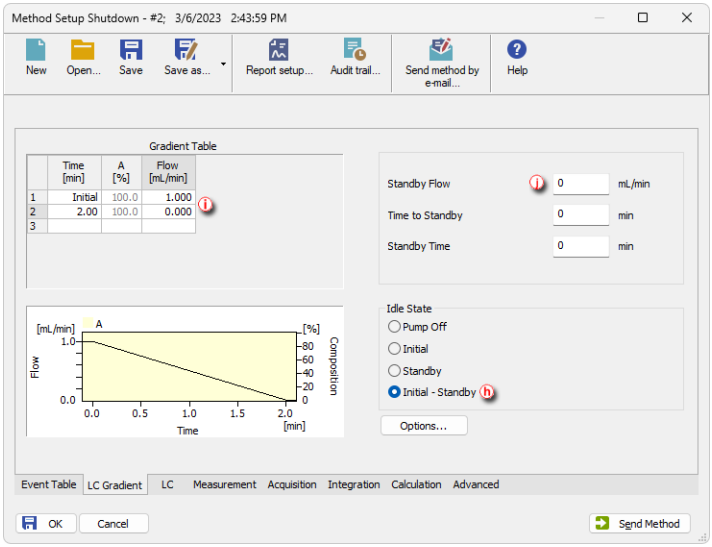
Analysis Method

7. Repeat steps 2 to 6 but this time:
    - Save the *Method* under the name *Analysis*.
    - Set the *Idle State* to *Initial* ⓓ .
    - Set the *Initial Flow*, *Flow* on the second row ⓔ and *Standby Flow* value ⓕ to the value of *Standby Flow* from *Startup* method.
    - Set the *Time* to the total duration of your analysis ⓖ .



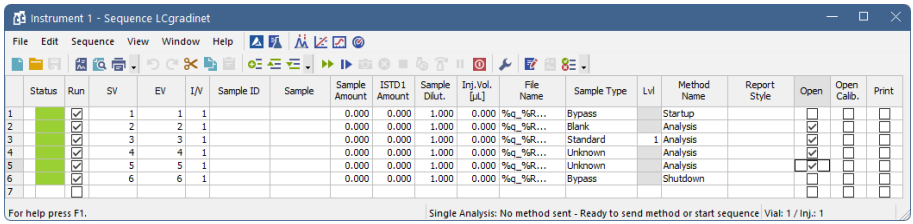
Shutdown Method

8. Repeat steps 2 to 6 but this time:
   - Save the *Method* under the name *Shutdown*.
   - Set the *Idle State* to *Initial - Standby* ⓗ .
   - On the second row, set the *Flow* to 0 and the *Time* to obtain the appropriate flow rate decrease for your column ⓘ .
   - Set the *Standby Flow* to 0 ⓙ .



Sequence

9. Set up a *Sequence* as explained in ***Running a Sequence*** and:
   - On the first row set the *Sample Type* column as *Bypass* and the *Method Name* to *Startup*.
   - On the last row set the *Sample Type* column as *Bypass* and the *Method Name* to *Shutdown*.
   - Set as many rows as you need in between according to the conditions of your analysis or sequence and set the *Method Name* to *Analysis*.

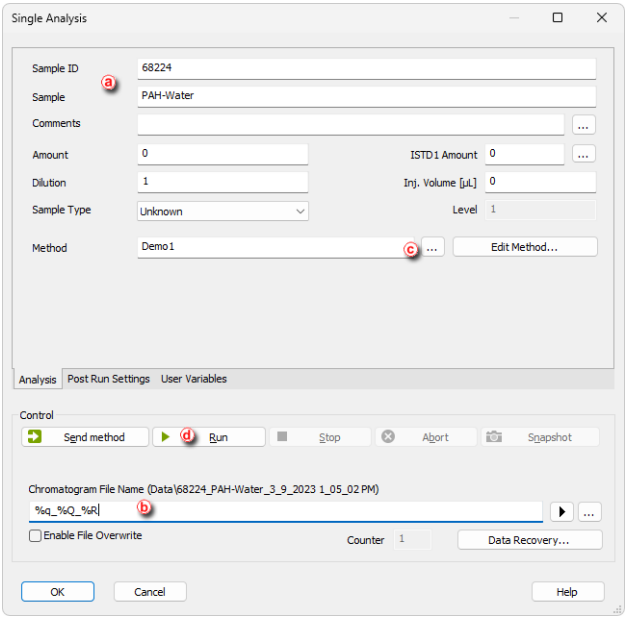


6 Data Acquisition

Chapters describing how to perform a measurement using *Single Analysis* or *Sequence* and how to evaluate chromatograms during run.

6.1 Running a single analysis

1. Open the *Single Analysis* dialog by selecting *Analysis - Single* or clicking on icon in the *Instrument* window.
2. Fill in the sample information into the *Sample ID* and/or *Sample* fields ⓐ .
3. Fill in the *Chromatogram File Name* ⓑ . It is recommended to use variables to do so, for example, *%q* (*Sample ID*) and *%n* (counter) or *%R* (date and time) to prevent file name conflicts. Name preview is displayed above the field. For more info regarding variables see "Chromatogram File Name" in the Clarity Reference Guide.
4. Select the *Method* ⓒ which should be used for measurement. It is also possible to edit it by clicking *Edit Method*.
5. What should happen after analysis like automatic report printing can be set on the *Post Run Settings* tab.



6. Start the analysis by clicking the *Run* button ⓓ . Analysis can also be triggered by *External Start* from the chromatograph. The Instrument will get to the

RUNNING state and the *Single Analysis* dialog will close. The state is visible in the status line in the *Instrument* window.

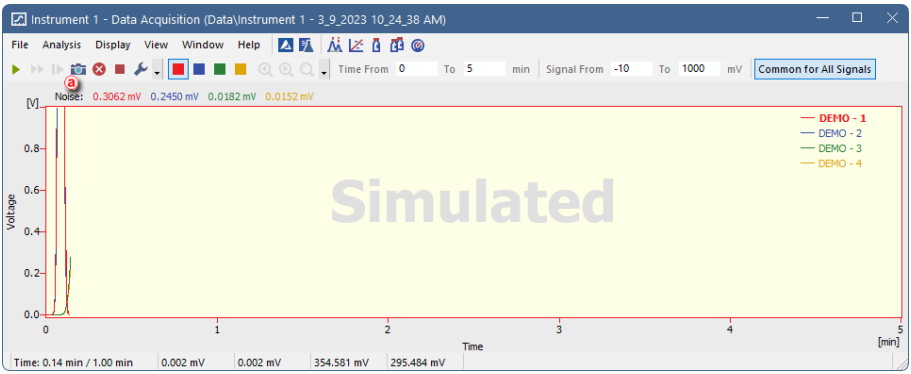
7. It is possible to monitor the analysis from *Data Acquisition* window, see Pre-evaluating a chromatogram during acquisition for more.

6.2 Pre-evaluating a chromatogram during acquisition

1. Open the *Data Acquisition* window by select *Window - Data Acquisition* or click on in the *Instrument window*.

2. To create a temporary chromatogram, select *Analysis - Snapshot* or click on ⓐ . Every time you take a snapshot, a temporary chromatogram will be created again from the beginning up to the point where you clicked.

Caution: To preserve a snapshot it has to be saved under new name, otherwise it will be overwritten once analysis is finished and final chromatogram is created.



6.3 Creating and running a sequence

1. Open the *Sequence* window by selecting *Analysis - Sequence* or click on the icon in the *Instrument* window.
2. Create a new sequence file by click ⓐ . or open an already created sequence ⓑ . If you already have a sequence ready skip to step 11.
3. Save the new sequence by selecting *File - Save As…* and give it a name in the saving dialog.
4. Check the checkbox in the first (empty) row in the *Run* column ⓒ . The row will be pre-filled with default basic information on the analysis.
5. Fill in the *Sample ID* and/or *Sample* columns ⓓ .
6. Fill in the *File Name* ⓔ . It is recommended to use variables to do so, for example, *%q* (*Sample ID*) and *%n* (counter) or *%R* (date and time) to prevent file name conflicts. Hold the mouse pointer over the file name field to see

resulting name. For more info regarding variables see "Chromatogram File Name" in the Clarity Reference Guide.

7. The *SV* (Starting Vial) and *EV* (End Vial) rows ⓕ will be pre-filled with numbers corresponding to the sample position in the autosampler tray, if you are using an autosampler. It is possible to change these as needed, injection from multiple vials can be made on one sequence row.
8. Fill in volume of the injection in the *Inj. Vol.* column ⓖ .
9. Select the method to be used in the *Method Name* column.
10. Tick any of the *Open, Open Calib., Print* etc., columns ⓗ in case you wish to open measured chromatogram in chromatogram or calibration window or print the results after each measurement of a sample.

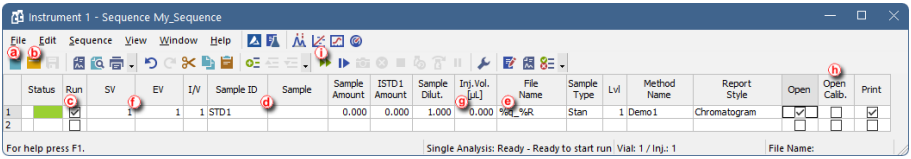
    Caution: To correctly include chromatograph graphs in the reports it is necessary to check both *Open* and *Print* checkboxes. It is also necessary to fill in *Report Style* when *Print* or *Print to PDF* is used.

11. Repeat the steps 4-10 for rows you need to add to the Sequence Table.

    Note: It is possible to display additional columns by right-clicking the sequence table, selecting *Setup Columns* and moving items from *Hide Columns* to *Show Columns* list by *Show* button.

12. Check the validity of the sequence by selecting *Sequence - Check Sequence* or clicking on icon. In valid sequences, all rows will show the symbol in the *Status* column. Invalid sequences will issue a warning message with the cause of the problem.
13. Save the sequence by selecting *File - Save* or clicking on icon.
14. Run the prepared sequence by selecting *Sequence - Run* or clicking on icon ⓘ .

    Note: The sequence state will change to *WAITING FOR INJECTION* or *INJECTING* state, depending on *READY* state of other controlled modules. The state is visible at the bottom of the Sequence window.



6.4 Shutdown after sequence is finished

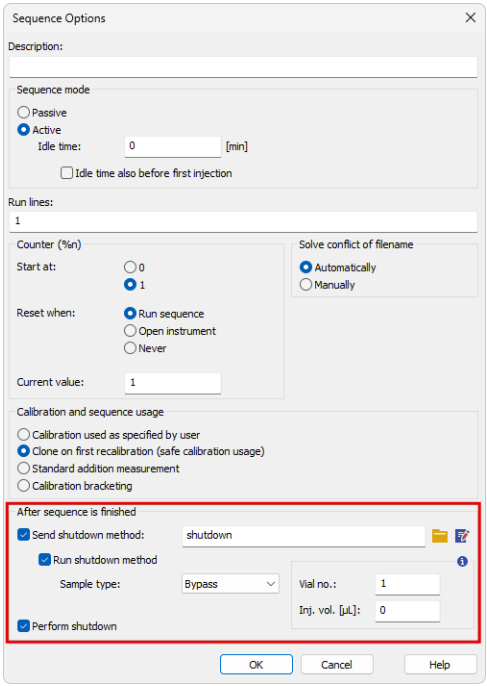
After the sequence is finished, it might be sometimes convenient to send (and run) shutdown method and/or perform shutdown. As the behavior of used controlled devices may be quite specific, there are multiple options how to get them to a desired standby state. For example some detectors may be able to perform run with lamps off or switch them off after end of a run, while others will not get to ready state

without lamp on. Proper combination of the possible actions needs to be set accordingly.

1. To setup shutdown open the *Sequence Options* in your opened sequence by selecting *Sequence - Options* or clicking on icon.
2. Now, you need to decide whether you only need to send shutdown method to controlled devices to prepare them for shutdown (step 3), or if you need also to run such method, e.g. for lowering the gradient (steps 3 and 4). As mentioned in the beginning of chapter this setting is highly depended on individual control module capabilities.
3. *Send shutdown method:* checkbox: Check this checkbox to be able to select and send shutdown method to controlled devices. You can also edit such method, if needed. This option will only send the selected method to devices.
4. *Run shutdown method* checkbox: Use this option to run selected method. Resulting chromatogram will be saved to the current project as [SEQUENCENAME - SHUTDOWN - METHODNAME - %R.PRM], where %R stands for current date and time.

   Note: Use *Bypass* to omit injection from the vial, or *Unknown* for injection from specified vial. Valid Vial position and Injection volume may be required by some autosamplers, even if the injection will not be performed (i.e. when method is not run or Bypass injection is selected).

5. *Perform shutdown* checkbox: Use this option to shutdown all controlled devices. Shutdown command is sent after sending and completed run of the shutdown method. Note that the reaction of some devices to the Shutdown may be amended in their respective Setup window in System Configuration.



Note: The parameters of the *After sequence is finished* section are also executed when the user stops the sequence by *Stop* command or the sequence is stopped by an error. If it is necessary to stop the running sequence without sending (and running) the shutdown method, use the *Abort* command.

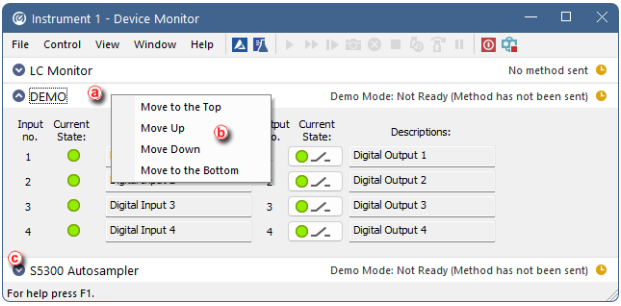
Note: When the sequence is already finished and the shutdown is also already performed, if new lines are added to the finished sequence and such sequence is then resumed, the whole *After sequence is finished* will be applied again.

7 Device Monitor

The *Device Monitor* window serves firstly to show the current status of each control module and secondly for direct control of the device. The layout and possible control options greatly vary based on the used control modules.

The window can be personalized. To change the order of individual monitors right click on the module's name ⓐ and choose desired action in the *context menu* ⓑ .

The individual monitor panel can be collapsed or expanded using the arrow icon ⓒ in the header of the panel.

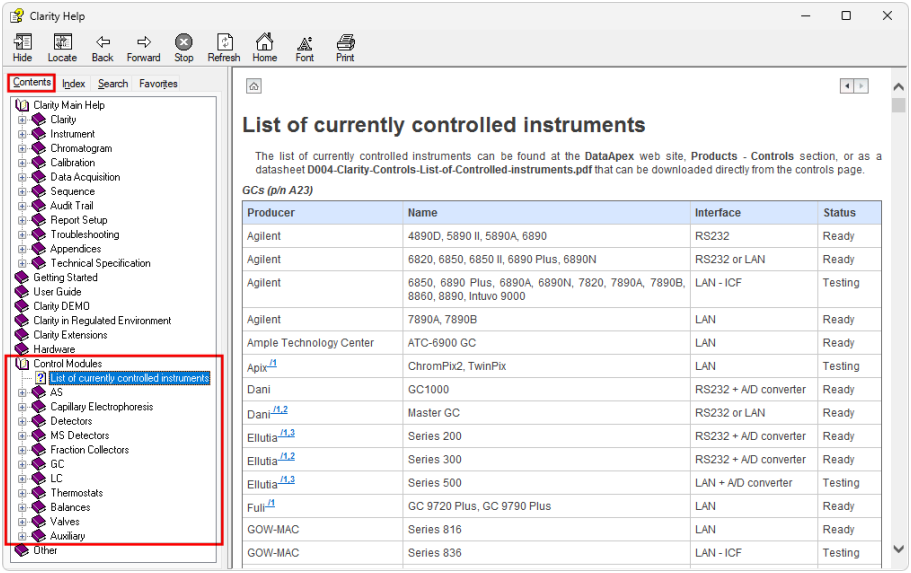


7.1 How to set parameters during run

Besides performing some service commands outside run, in some cases it is possible to perform some actions also during run. This is individual for specific control modules and could be found in the help or manual of respective control module. Control module helps are located at the bottom of the *Contents tab* of Clarity help which can be invoked by pressing *F1*. Manuals are available at www.dataapex.com/downloads.

Caution: Manuals are not available for all controlled devices. Control modules developed by third parties are only supplied with help file.

Common actions available for the pumps are described in next chapter.



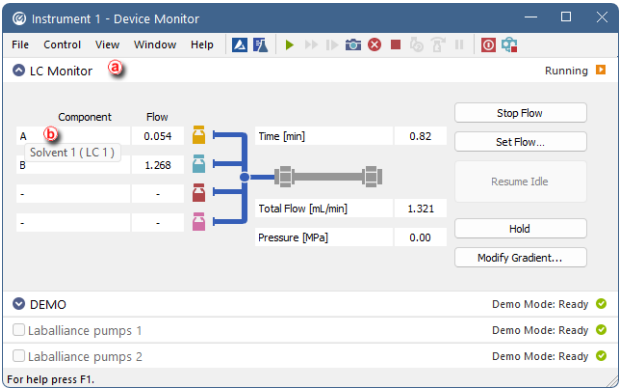
7.2 How to directly control LC gradient pumps during run

For gradient pumps, the monitor consists of the *LC Monitor* ⓐ section common to all pumps present in the gradient. Maximum number of solvents configured to LC Gradient is 4 - it could be any combination of isocratic and gradient pumps. If another pump is needed, it has to be added as an auxiliary pump.

Note: Tooltip ⓑ over each component shows name of the pump.

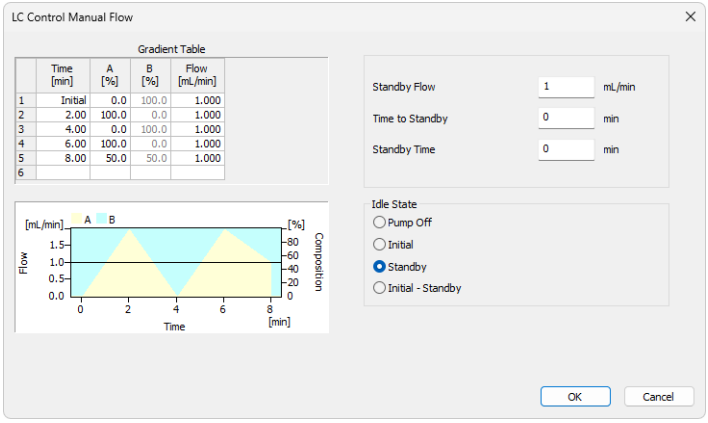
It is possible to:

- *Stop Flow* (pump is stopped without stopping analysis) and *Set Flow* (custom flow rate and composition can be set) both can be done outside or during analysis. Beware that when used during analysis the gradient cannot be restored and the set flow will be used for the rest of acquisition.
- *Resume Idle* switches pump to the idle state as defined in *Method Setup - LC Gradient*.



Further commands are available for selected pumps (typically those controlled by Clarity in real time). They are enabled only during the run where gradient is in progress.

- *Hold* halts the gradient at its current state and the button changes to *Resume* which can be used to resume the gradient from the point where it was halted.
- *Modify Gradient* invokes *LC Control Manual Flow* dialog (similar to the *Method Setup - LC Gradient*) which can be used to adjust the gradient for the current analysis. This does not influence the method used for analysis in any way.



8 Chromatogram Editing

This chapter focuses mainly on adjusting integration. For proposed general workflow see chapter How to integrate chromatogram

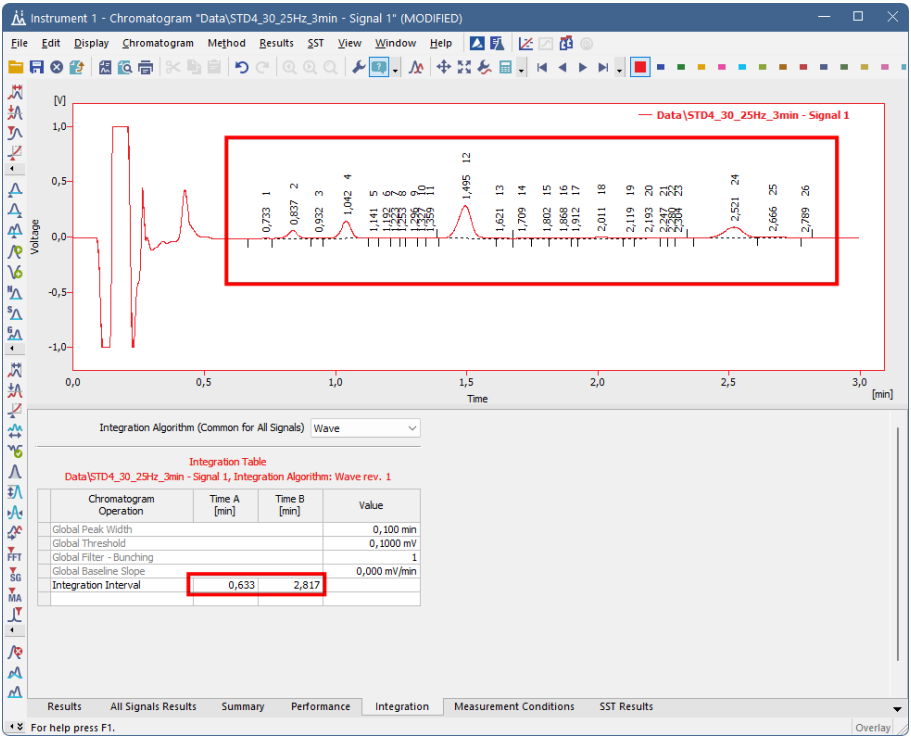
Clarity Tips&Tricks videos covering Integration topics can be found in Clarity Integration playlist.

8.1 How to integrate chromatogram

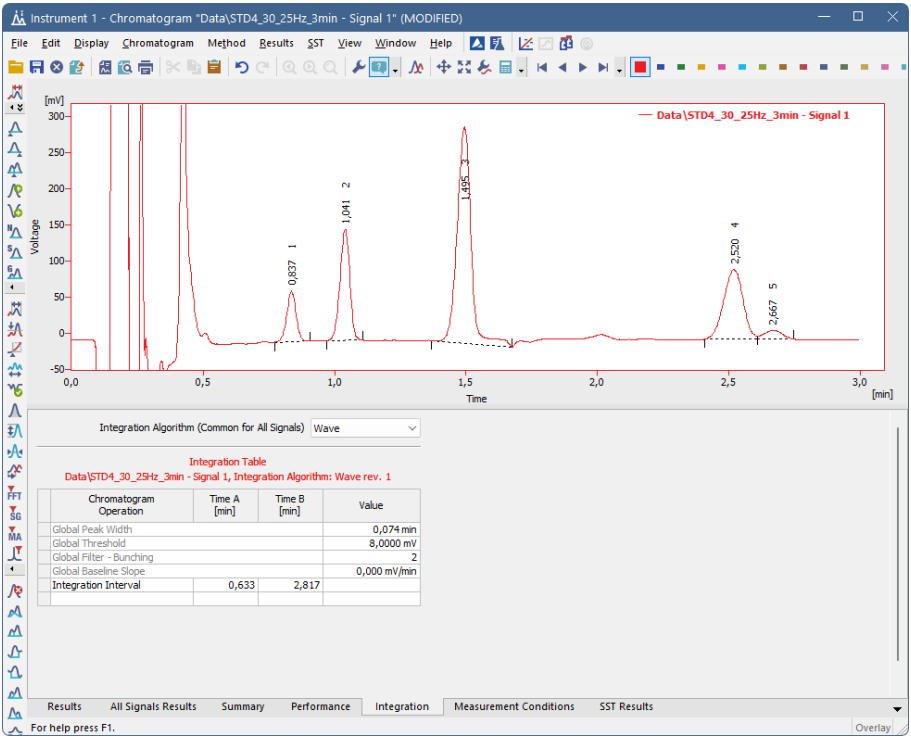
In order to have the integration of routine samples automated as much as possible it is necessary to optimize integration parameters and then save them to the method that is used for measurements. Two integrations algorithms (Wave and Legacy) with slightly different approach are available, if you are not able to achieve satisfactory integration in one of them try using the other one. Integration Algorithm can be changed on Integration tab in Method Setup/Chromatogram.

1. Open one typical chromatogram for your analysis.

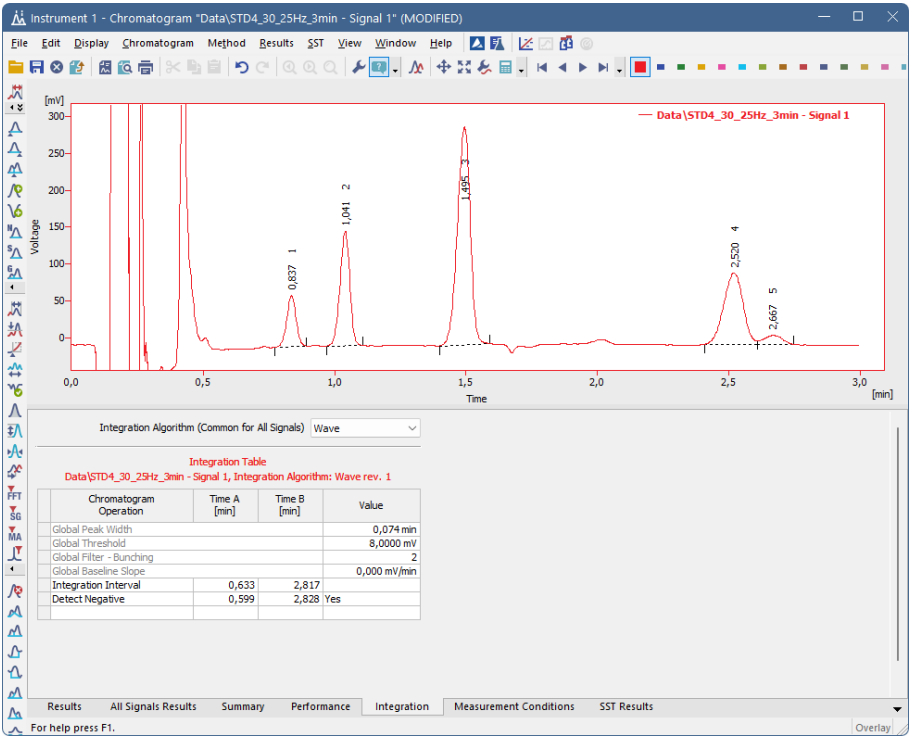
2. Set *Integration Interval* as needed. More details are in Setting Integration Interval topic.



3. Set *Global Peak Width* parameter. Choose the narrowest peak which should be integrated in your chromatogram and select his start and end. This setting is used for calculation of *Global Bunching*.
4. Set *Global Bunching* parameter. This is a filter which is calculated based on *Global Peak Width* and sample rate.
    a. Alternatively, for chromatogram with high noise, it is possible to use other filters - *FFT Filter*, *Savitzky-Golay Filter* or *Moving Average Filter*.
5. Set *Global Threshold* parameter. This is influenced by the *Global Bunching* or other filter set previously. Select an interval containing only noise, not peaks. A more detailed description of the global parameters is covered by topic Modifying Global parameters.

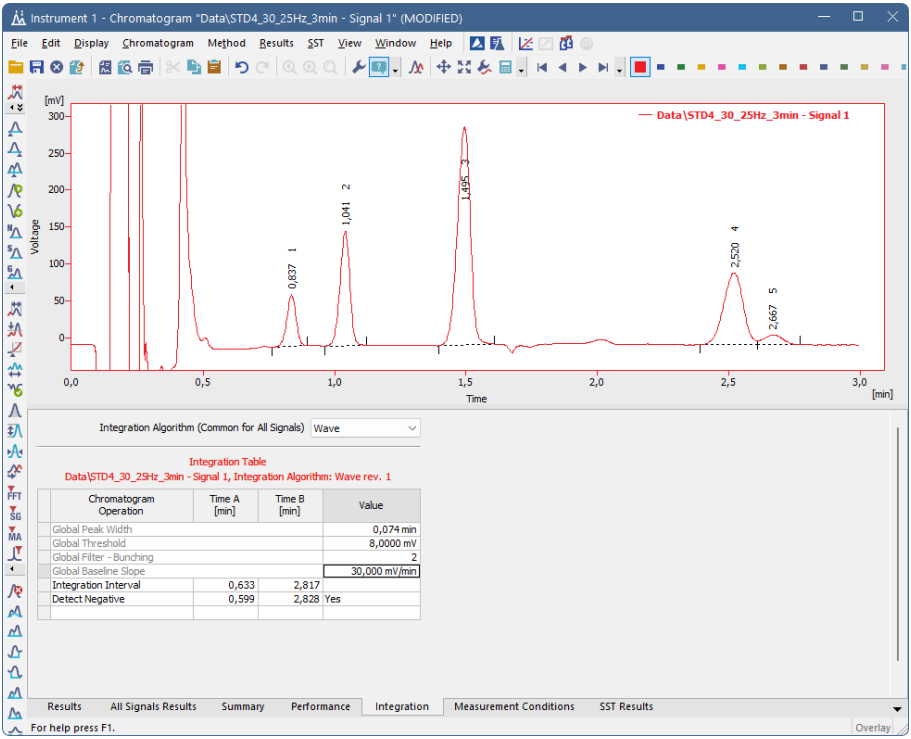


6. (Optional) Set *Detect Negative* parameter. This has two potential uses, firstly when you want to integrate also negative peaks. Secondly it may help when signal has inconsistencies such as dips. This will prevent the integration algorithm from placing peak starts/ends into them.



7. Set *Global Baseline Slope* parameter. Change this parameter after you have all the peaks integrated, it servers to optimize their start/end positions.

Note: Global Slope is available only in Wave Integration Algorithm.



8. If you still aren't satisfied with the integration there are another possibilities for manual changes.
    a. Try to use Interval parameters such as *Baseline Together*, *Baseline Valley*, *Forward Horizontal* etc..
    b. Or try to use Peak parameters such as *Peak Start*, *Peak End*, *Peak Both*, *Add Positive* etc..

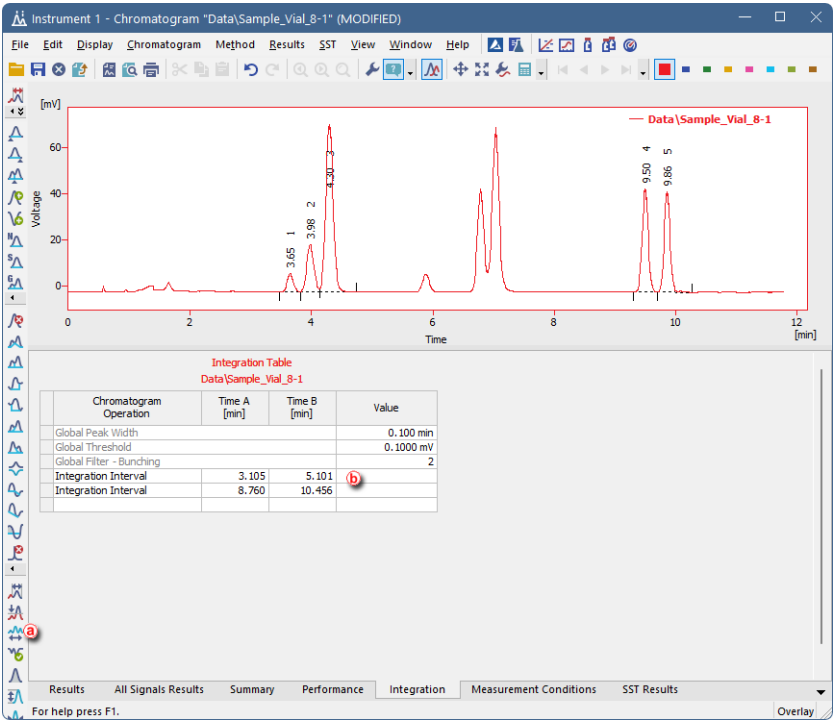
    Note: *Add Positive* operation should be used as a last resort because is prone to every Retention time shifts. Peak Start and Peak End should be preferred parameters as they are defined relative to peak apex.

9. When you are happy with the integration. Save method as Template and use it for further measurements of these sample types.
10. If you have already measured some chromatograms and want to use this integration in them you can do it using Batch - Reprocess by Method.

8.1.1 Setting the integration interval

1. Signal outside of the interval is not integrated, this function is mainly useful for omitting solvent peaks etc.

2. To set the interval on which to apply the integration use *Chromatogram - Integration - Integration Interval* or icon ⓐ . It is possible to set multiple intervals within a single chromatogram.
3. You can check, modify the interval or delete this operations in the *Integration Table* ⓑ .



Note: If you want to exclude specific interval from the integration refer to ***Remove peak from integration***.

8.1.2 Modifying Global parameters

Peaks are only integrated if they exceed a minimum width and height. Slope parameter influences position of starts and ends of peaks. These optimizations can be achieved both globally or locally in a specified interval. It is always recommended to maximally utilize global parameters before using any local one. Global Bunching works as a smoothing filter, it has to be used only globally to whole chromatogram.

Global Peak Width:

- To adjust minimum width use *Chromatogram - Global Peak Width* or use  icon ⓐ and select the narrowest you want to integrate.

Global Threshold:

- To adjust minimum height use *Chromatogram - Global Threshold* or use  icon ⓐ and select an interval containing only noise, not peaks.

Global Bunching:

- Works as a smoothing filter that automatically averages number of data points in order to preserve at least 30 data points per narrowest peak. It is dependent on Global Peak Width value. To use it select *Chromatogram - Global Bunching* or use the  icon ⓐ after you done adjusting the Global Peak Width parameter.

Global Slope:

- To adjust minimum slope, where the peak should start/end use *Chromatogram - Global Slope* or use  icon ⓐ and select the point where the peak should start/end.

Note: Available only in Wave Integration Algorithm.

If you are still not happy with the integration suggested by selection from the graph you can modify the parameter values in the *Integration Table* ⓑ .

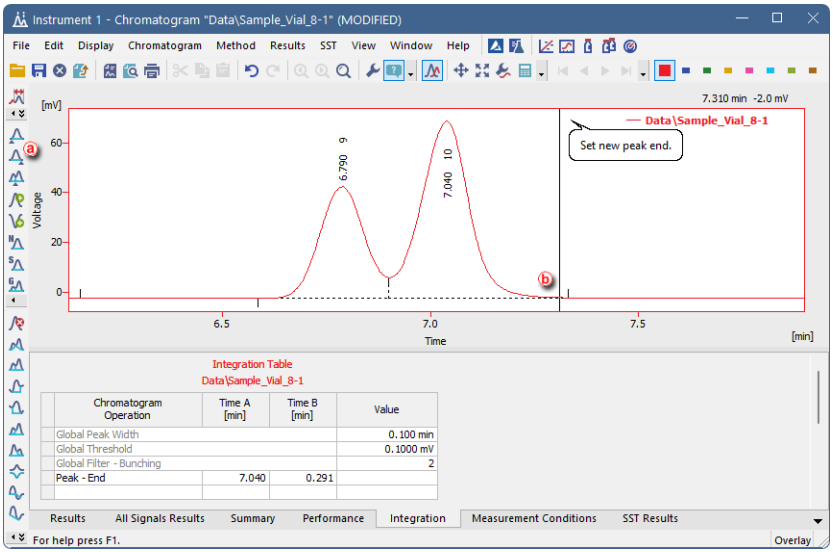
For setting the Local variants of Global parameters:

1. Setting local parameters is similar to the global ones, the only difference is that they are only applied on selected time interval rather than entire chromatogram.
2. To set local parameter use context menu *Chromatogram - Integration* and select parameter you need. Similarly you can find the icon in the bottom part of the vertical toolbar ⓒ . Select the time interval in the graph. Small dialog appears ⓓ , here you can further specify the time interval and mainly select the value of the parameter. You can use *Suggest Value* button ⓔ to select the value from the graph same as for global parameter.
3. Again, all values can be further modified in the *Integration Table* if needed.

Note: Global bunching is a filter that is always applied to the whole chromatogram. To filter data locally use other filers from Chromatogram - Integration menu.

8.1.3 Modifying the beginning and the end of a peak

1. To change the position of peak beginning and/or end use *Chromatogram - Peak - Start* and/or *Chromatogram - Peak - End* alternatively use and/or icons ⓐ .
2. Set the new beginning ⓑ and/or end by clicking the new position in chromatogram graph.



8.1.4 Removing a peak from integration manually

With the introduction of the WAVE integration algorithm, there are now 2 ways to remove the peak from the integration. First one is present also in a Legacy Integration algorithm, the second is exclusive for the Wave IA.

1. Use *Chromatogram - Baseline - Lock* (or icon ⓐ) or *Chromatogram - Peak - Hide*.
2. Click once to mark the start of the interval that should be omitted from integration and click second time to mark its end ⓑ .

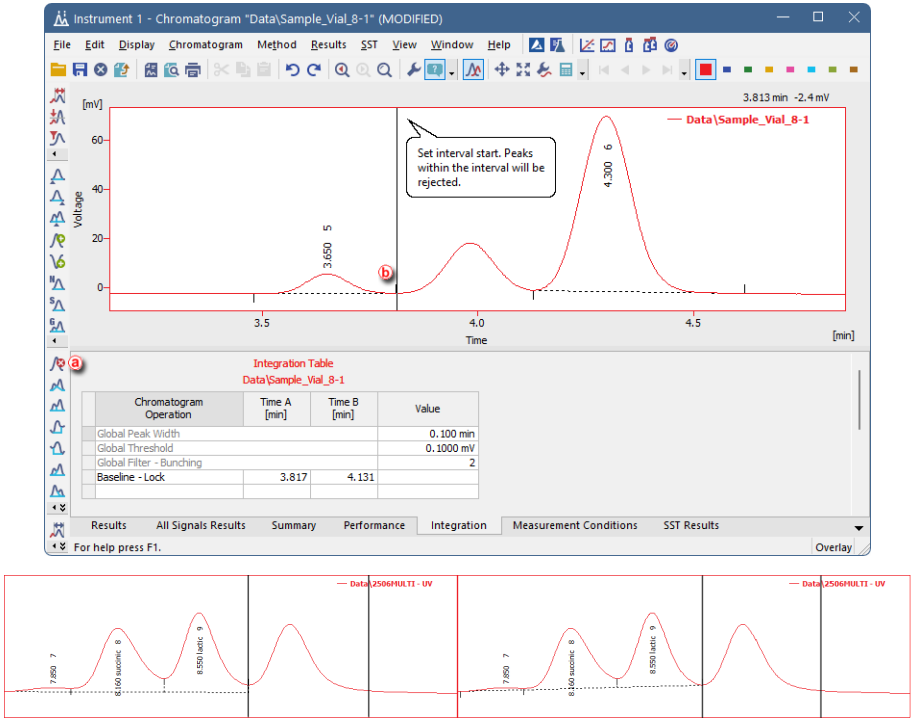


Fig. 1: Difference between Peak Hide (left) and Baseline Lock (right).

Note the two vertical guidelines marking the beginning and end of the peak. More than one peak can be removed from integration at once.

8.1.4.1 Baseline - Lock

This tool completely removes all peaks that have their maxima within a set time interval. This could affect other peaks with maxima outside the specified window, particularly if they share the same baseline.

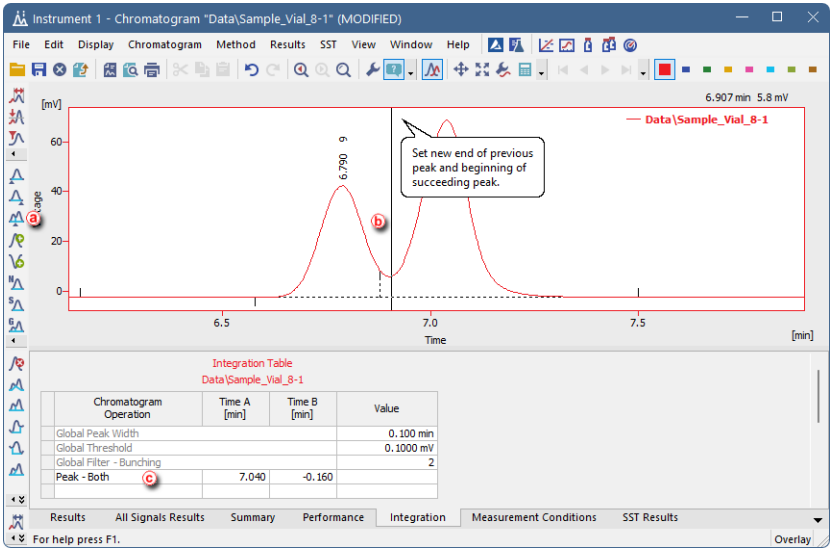
8.1.4.2 Peak - Hide (and Show)

By default, the Wave algorithm might automatically ignore some of the minor peaks identified during integration. However, completely eliminating these peaks could potentially affect the baseline's trajectory. Therefore, these functions offer a flexible way to handle unwanted peaks without completely removing them.

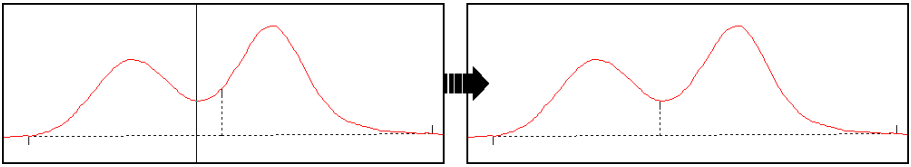
8.1.5 Changing the position of a peak separating line

1. Select *Chromatogram - Peak - Both* or click in the *Peak* toolbar ⓐ .
2. Set the new position of peak-separating line by clicking in the chromatogram graph ⓑ .

3. You can check, modify the position or delete this operation in the *Integration Table* ⓒ .

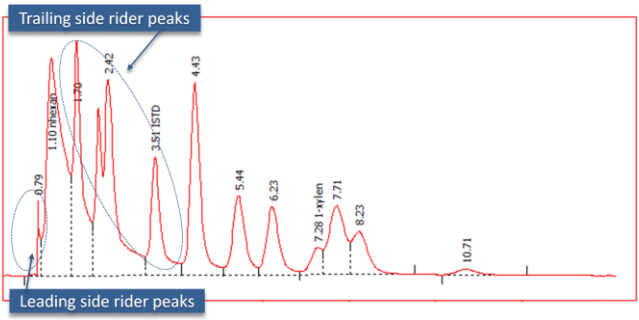


Note the vertical guideline indicating the currently selected peak separator position.



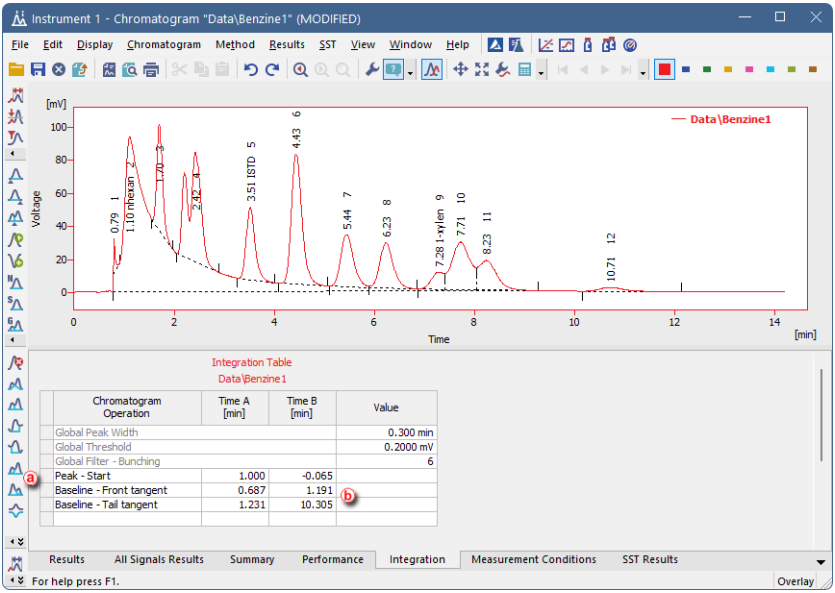
8.1.6 Separating rider peaks by tangent

Rider peaks are small peaks which are not well resolved from a large and asymmetrical neighbor but sit on its leading or trailing side.



1. Select the rider peaks you want to separate on the leading side of the mother peak: use *Chromatogram - Baseline - Front Tangent* or icon ⓐ .
2. Select the rider peaks you want to separate on the trailing side of the mother peak: use *Chromatogram - Baseline - Tail Tangent* or icon ⓐ .
3. You can check, modify the interval or delete this operations in the *Integration Table* (*Baseline - Front tangent* and *Baseline - Tail tangent* rows) ⓑ .

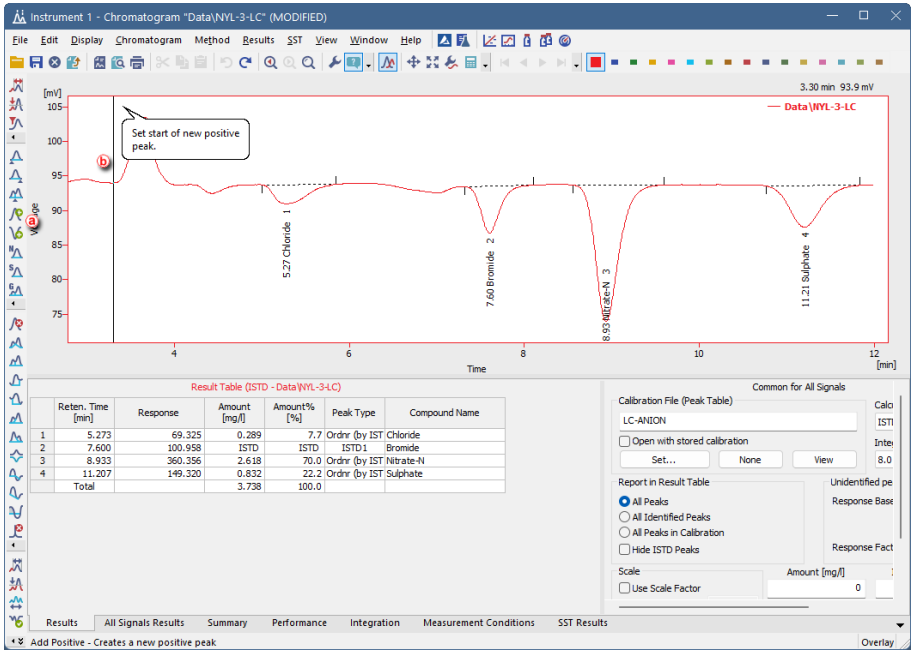
Note: You can also use the *Tangent Slope Ratio* and *Tangent Area Ratio* from *Chromatogram - Separation* menu to set a threshold for the separation of rider peaks based on those two parameters.



8.1.7 Adding a new peak manually

Caution: Manual addition of a peak should be used as a last resort to finish integration of individual chromatograms. Since the function is based on absolute time, when used in template method results might be inaccurate if retention time shift occurs.

1. Select *Chromatogram - Peak - Add Positive/Negative* (also accessible from context menu) or click and in the *Peak* toolbar ⓐ . Holding Ctrl key while selecting the function will allow you to use it repeatedly.
2. Click in the chromatogram and set the beginning ⓑ and the end of a new peak.



Note the two vertical guidelines marking the beginning and end of the peak. After finishing the operation new peak is added ⓒ .

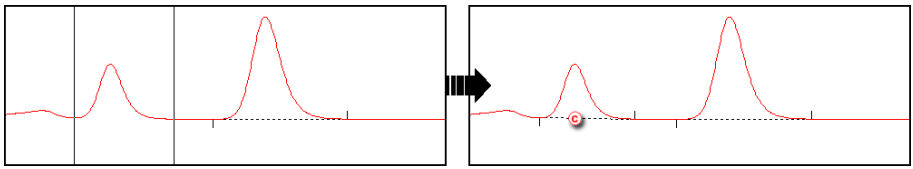


Fig. 2: Add Positive

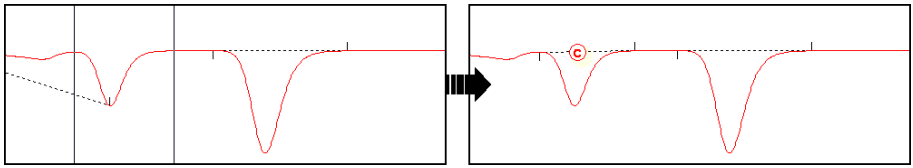


Fig. 3: Add Negative

8.2 Saving the chromatogram method as a template method

Each chromatogram contains saved copy of the method used for acquisition of such chromatogram. However, changes done in *Chromatogram* window (e.g. *Description, Integration Parameters, linked Calibration File*...) are not updated in the original method used for acquisition nor processing. To amend your method

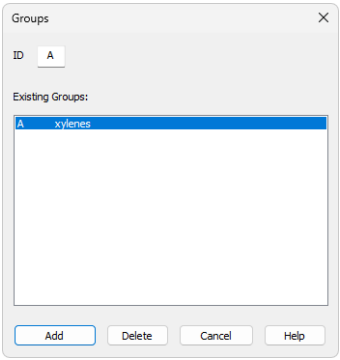
accordingly, you can create template method from the method in chromatogram, which is useful after optimizing integration parameters.

1. Open the chromatogram with optimized method.
2. Select the *Method - Save as Template…* to save the method (including the changes made by you) from the current chromatogram as a new method, or for example overwrite (update) your acquisition method.

Note: It is not possible to overwrite any method that is currently in use, for example a method opened on any Instrument.

8.3 Adding a peak to a group

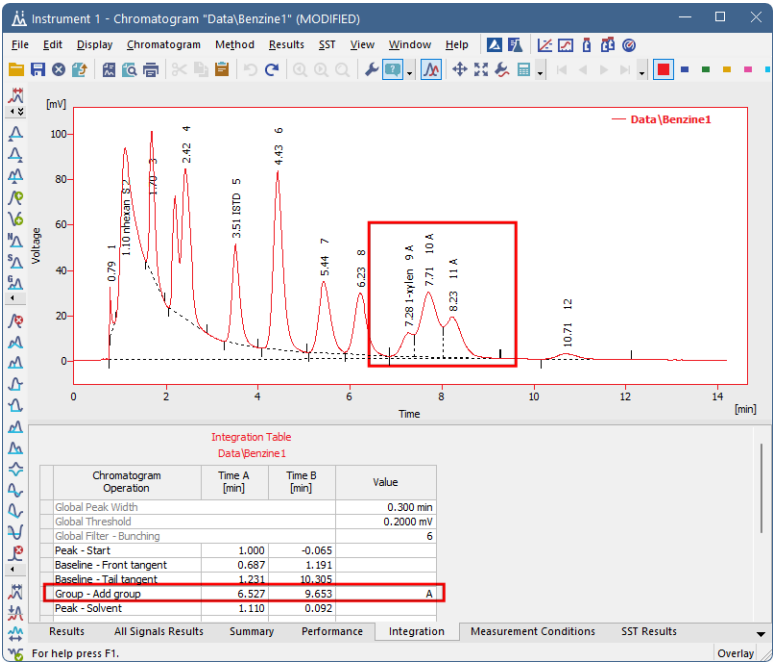
1. Use menu item *Chromatogram - Peak - Peak Groups…* to invoke *Groups* dialog.



2. Either select an existing group from the list or create a new one by inserting a letter to *ID* (groups are defined by a single letter) field and click the *Add* button.

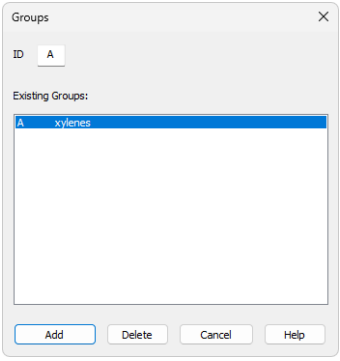
   Note: Group Name is based on the calibration file, similarly to the Compound Name.

3. Click in the chromatogram for the first time to select the start point and second time to select the end point of the group interval. Peaks with apexes found in the interval will be added to the new group.

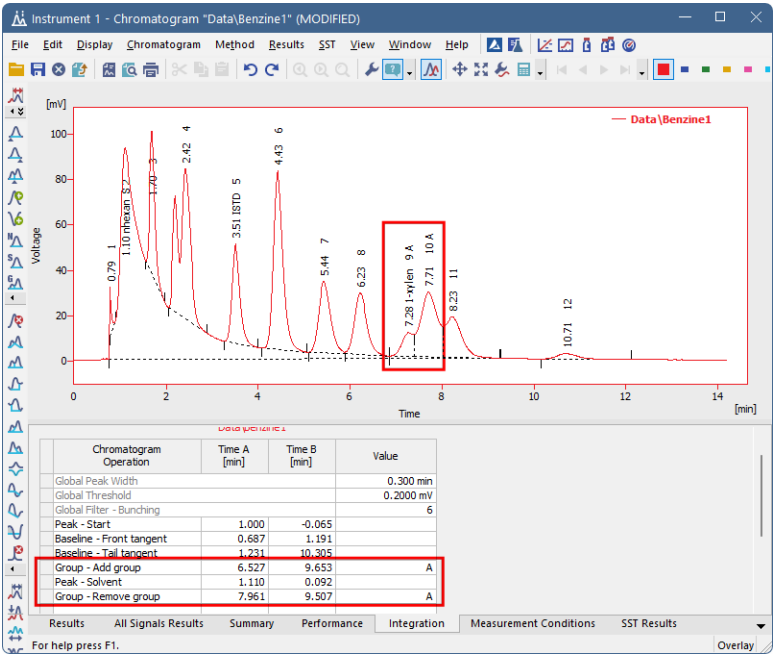


8.4 Removing a peak from a group

1. Use menu item *Chromatogram - Peak - Peak Groups…* to invoke *Groups* dialog.



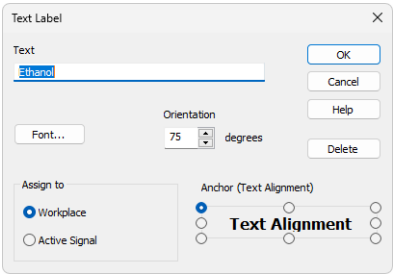
2. Select an existing group from the list and click *Delete*.
3. Click in the chromatogram to select the start and again to select the end of the interval which should be removed from the group.



8.5 Adding text and lines to a chromatogram

Adding text label

1. Right-click in the chromatogram graph to invoke context menu and select *Create Label - Text*.
2. "T" will appear next to the cursor, click wherever you would like to place the label. *Text Label* dialog will open.
3. Write the text in the *Text* field.
4. Select the font by clicking the *Font* button.
5. Enter the *Orientation* of the text (0 degrees equals horizontal position).
6. Select the *Anchor point* for the text.
7. Use *Assign to - Workplace*, if you want the text to stay in the same location regardless of the opened chromatogram (labels stored in the desktop file). Or use *Assign to - Active Signal*, if you want the text to shift as the chromatogram signal moves, zooms in and out (labels stored in the chromatogram file). The text will be displayed only when the respective chromatogram signal is active.
8. Click the *OK* button to accept the settings.

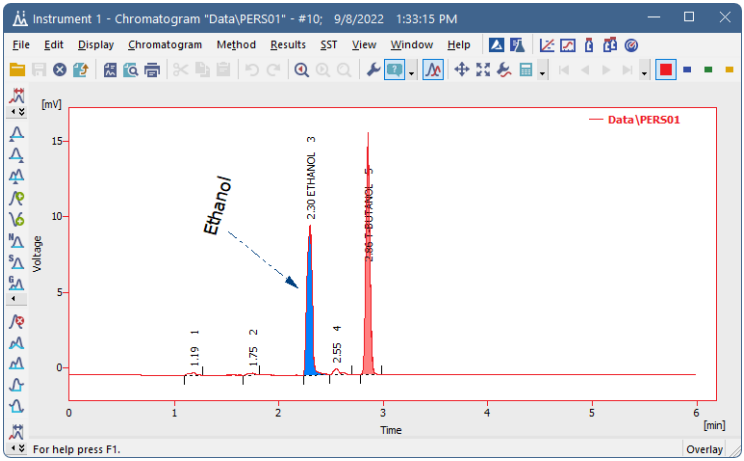


9. Click and drag the text if you wish to move it to a different location.
10. Double-click the text to open *Text Label* dialog to adjust the label settings or to delete it.

Adding line label

1. Right-click in the chromatogram graph to invoke context menu and select *Create Label - Text*.
2. "L" will appear next to the cursor, click and drag to place the line label. *Line Label* dialog will open.
3. Select whether you want to add an arrow tip at the beginning, end or at both ends of the line.
4. Select the color by clicking the *Color* button.
5. Enter the *Line Width*.
6. Select the *Line Style*.
7. Use *Assign to - Workplace*, if you want the text to stay in the same location regardless of the opened chromatogram (labels stored in the desktop file). Or use *Assign to - Active Signal*, if you want the text to shift as the chromatogram signal moves, zooms in and out (labels stored in the chromatogram file). The text will be displayed only when the respective chromatogram signal is active.
8. Click the *OK* button to accept the settings.

9. Click and drag the line if you wish to move it to a different location. Alternatively drag one of the ends to change its position.
10. Double-click the line to open *Line Label* dialog to adjust the label settings or to delete it.


Instrument 1 - Chromatogram "Data\PERS01" - #10; 9/8/2022 1:33:15 PM
File Edit Display Chromatogram Method Results SST View Window Help
[mV]
Voltage
Data\PERS01
Ethanol
1.19 1
1.75 2
2.30 ETHANOL 3
2.55 4
2.86 1-BUTANOL 5
Time
[min]
For help press F1.
Overlay

9 Calibration

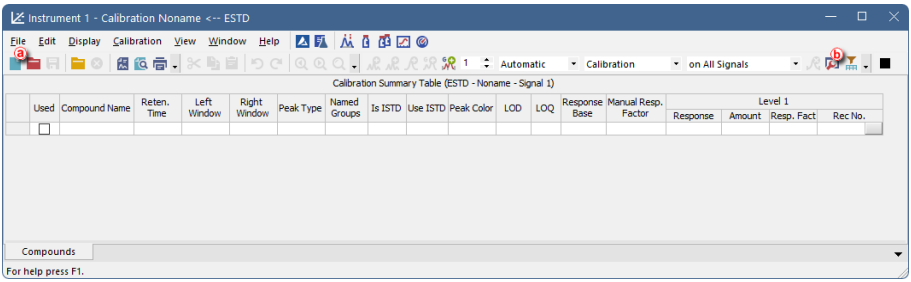
Following chapters contains multiple topics which will guide you through the basic principles of calibrating in **Clarity** and also introduces you to advanced solution, for example using the *Bracketing*.

Clarity Tips&Tricks videos covering Calibration topics can be found in Clarity Calibration playlist.

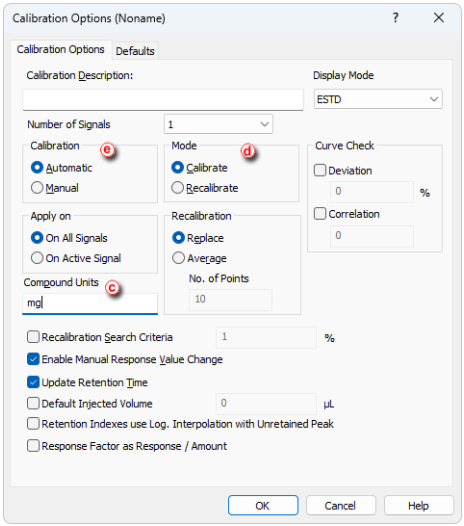
9.1 Creating a new calibration

This chapter covers creating a calibration file. You should have at least one integrated measured standard to be able to fill in the desired peaks into the newly created calibration.

1. Open the Calibration window: choose *Window - Calibration* in the *Instrument* window or click .

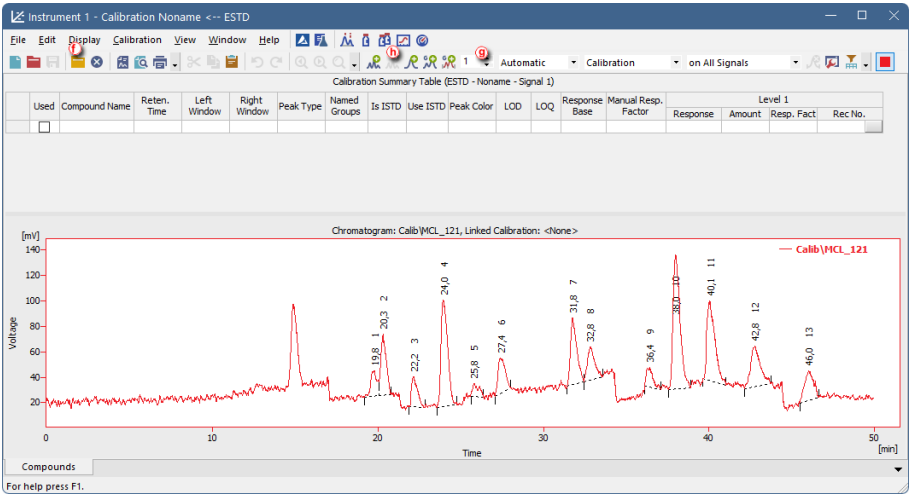


2. Create a new calibration file: select *File - New* or click ⓐ .
3. Open the *Calibration Options* dialog: choose *Calibration - Options…* or click ⓑ .

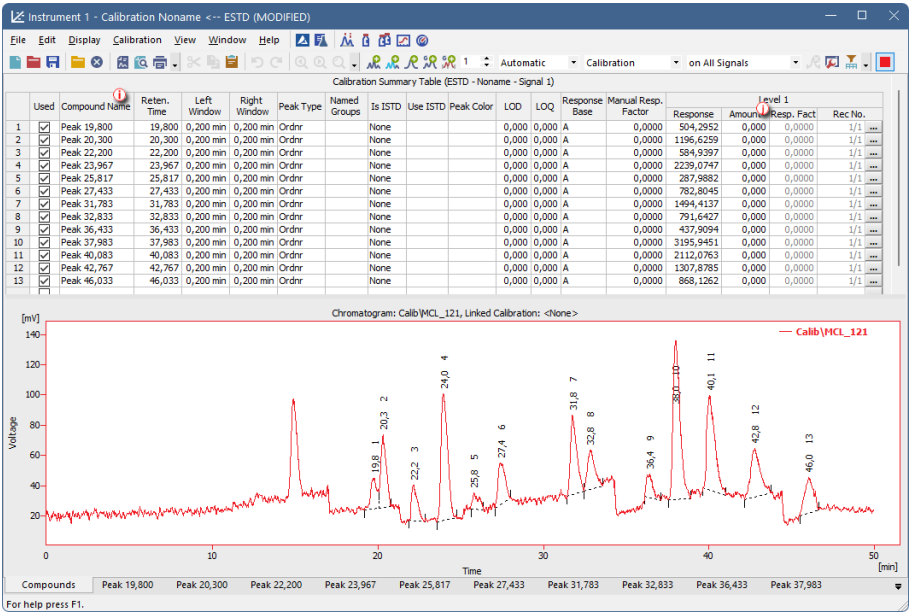


4. Fill in the units in the *Compound Units* section ⓒ to suit your analysis conditions.
5. Set the *Mode* to *Calibrate* ⓓ .
6. Set the *Calibration* option to *Automatic* to add the peaks without modification or *Manual* to modify them one by one ⓔ .
7. Open integrated chromatogram of a standard: choose *File - Open Standard…* or click on ⓕ in the *Calibration* window.
8. Add peaks belonging to the compounds of interest from the chromatogram of the standard to the calibration file.

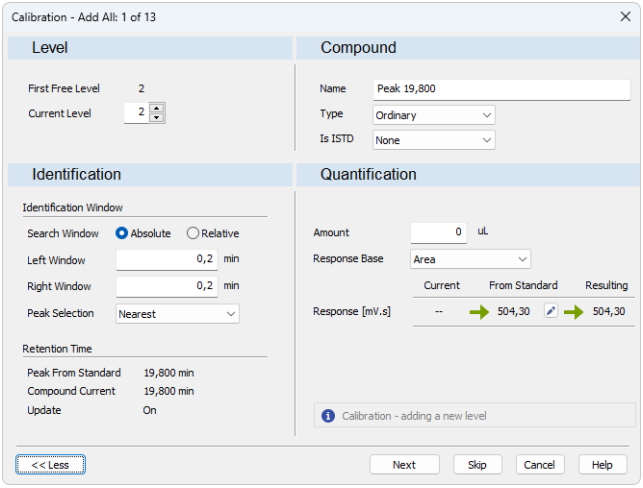
Note: Select *Calibration - Add All* or click on to add all integrated peaks or the Add Peak / Add Group icons to add specific peaks ⓗ . Regardless of the set Current Level ⓖ the peaks will be added to the first free level.



9. If you selected *Automatic* calibration:
   - Name the peaks identified in the *Calibration Summary Table* ⓘ by their retention times by typing the *Compound Name* for each peak. No compound name may be used more than once.
   - Fill in the *Amount* ⓘ for each compound into the *Calibration Summary Table*.



10. If you selected *Manual* calibration: Fill in the Amount, the Compound Name and set any other parameters related to the peak on the *Calibration - Add Peak* window. This window will open once for each one of the peaks processed.

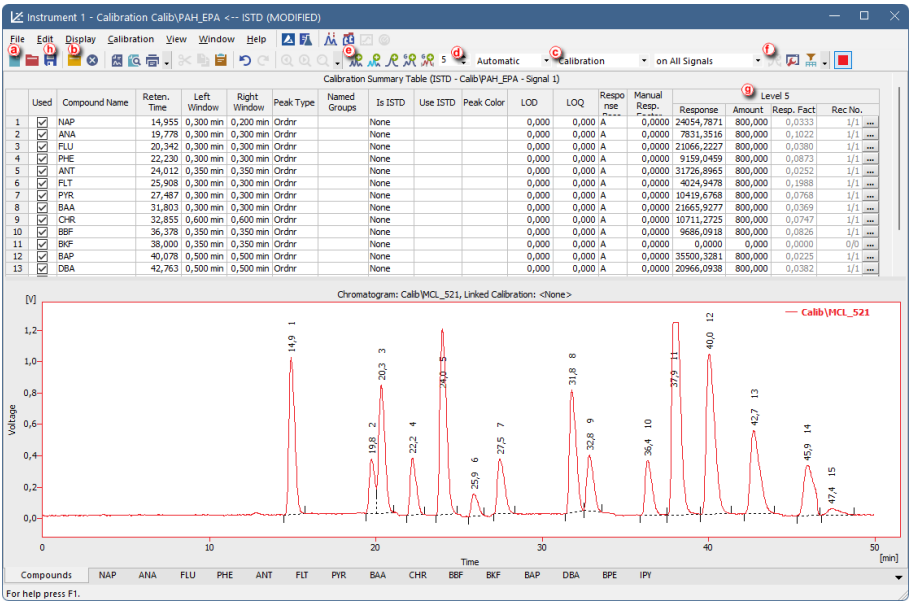


11. Save the calibration file - *File - Save* or click .

9.2 Adding a new calibration level

Here we describe how to add concentration levels to the calibration file to obtain the calibration curve of all compounds. This procedure has to be repeated several times, once for each calibration level to be added.

1. Open the Calibration window: choose *Window - Calibration* on the Instrument window or click icon.



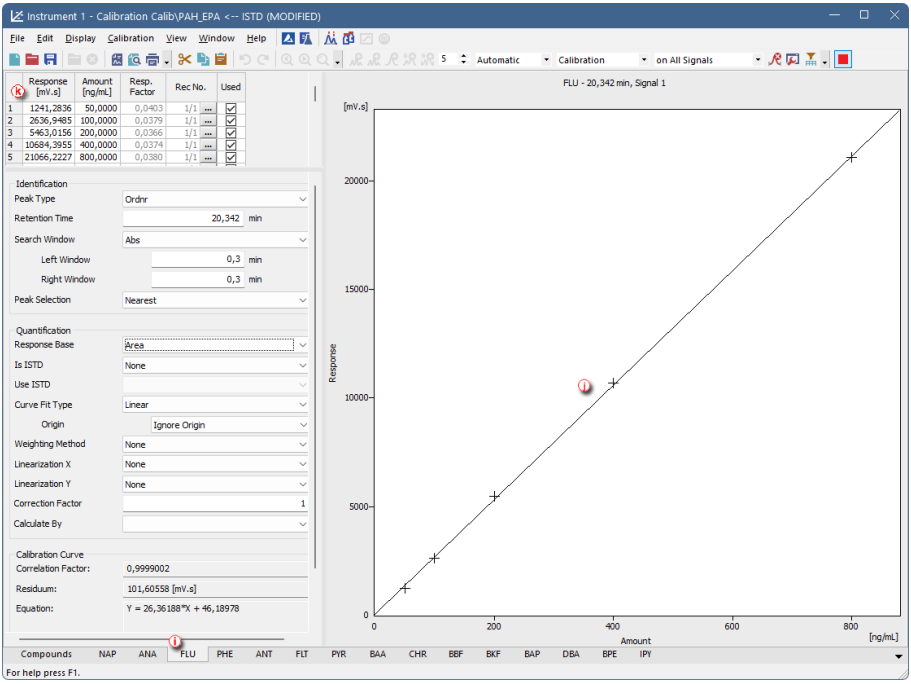
2. Open the calibration file: choose *File - Open…* or click ⓐ .
3. Open calibration standard: select *File - Open Standard…* or click ⓑ .

   Note: Select a measured and qualitatively evaluated chromatogram where all peaks are available, if possible.

4. Check that the *Automatic* option is selected in the first field and the *Calibration* option in the second field of the calibration mode settings ⓒ .
5. The calibration level number is in the *Current Level* field ⓓ set automatically to the first free level.
6. Add all peaks in the chromatogram of the calibration standard to the calibration file: select *Calibration - Add All* or click ⓔ .

Note: In case more peaks than expected emerge in the calibration, the surplus peaks can be deleted by selecting them in the *Calibration Summary Table* and deleting them using *Calibration - Delete Compound* or clicking on ⓕ .

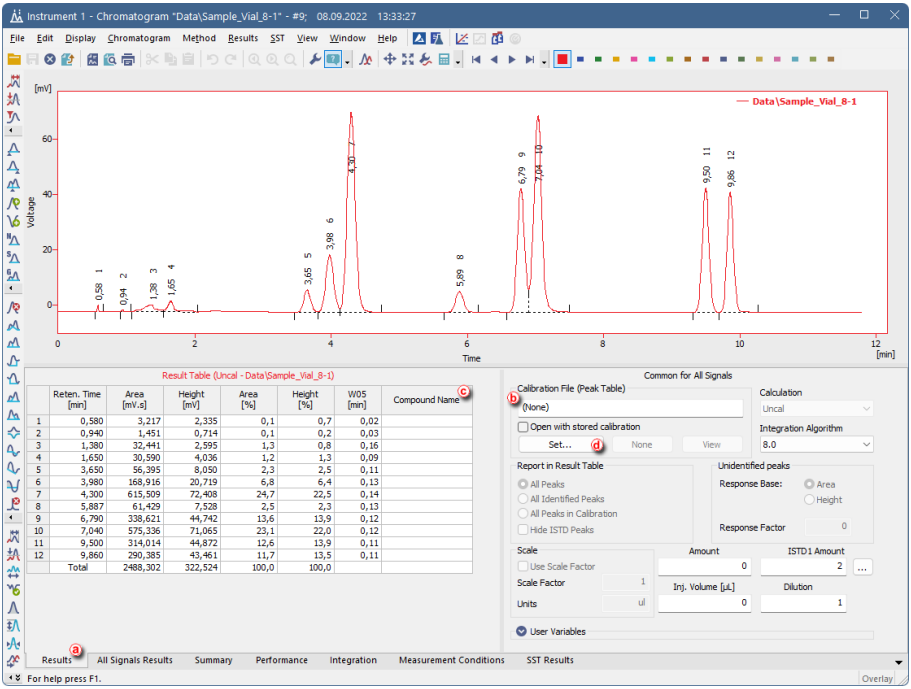
7. Set the amounts of the particular compounds into the *Calibration Summary Table*, into the *Amount* column ⓖ of the respective calibration level.
8. Save the calibration file: choose *File - Save* or click ⓗ
9. Click any tab below ⓘ and you will be able to see the calibration curve ⓙ with all the levels added for one specific compound ⓚ .



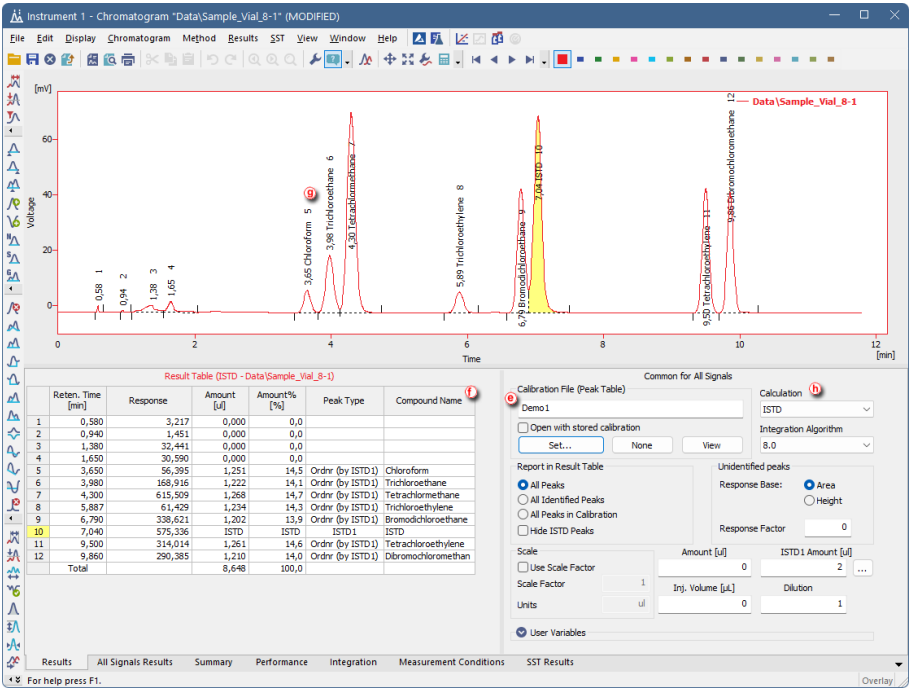
9.3 Applying a calibration to a chromatogram

If the calibration file was not assigned in the template method, the measured chromatogram will not have it linked either. Here you will learn how to link a calibration file to a chromatogram.

1. Switch to the *Results tab* ⓐ at the bottom part of the *Chromatogram window*.



2. Check whether the *Calibration File (Peak Table)* field ⓑ is set to *(None)*. If that is the case, then the chromatogram does not have a calibration file linked to it.
3. Also check the *Compound Names* ⓒ in the Result Table section. This column must be empty.
4. To link the calibration file to the Chromatogram, click the *Set…* button ⓓ in the right section of the *Results tab*. You will get a list of all calibrations available in the present project.
    5. Select the correct calibration file from the list and click *OK*. The content of the Chromatogram window will change.

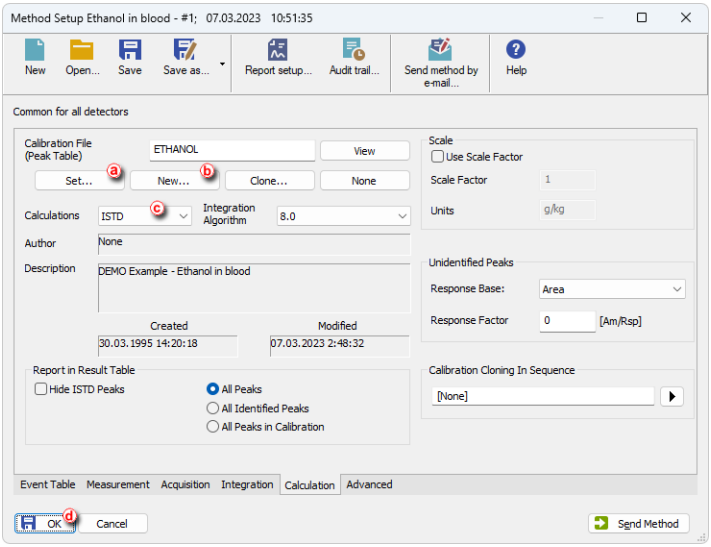


6. Check that the *Calibration File (Peak Table)* field contains the name of the calibration file. ⓔ
7. The *Compound Name* column ⓕ in the *Result Table*, as well as the identified peaks in the graph ⓖ , will now have the names of the identified peaks from the calibration file.
8. Check the *Calculation* field ⓗ to see the type of calculation performed on the chromatogram.
9. Save the chromatogram: select *File - Save* or click .

9.4 Setting the calibration in the method

After the acquisition is performed according to a method, the resulting chromatogram files may be calibrated using a specific calibration. If you need to measure a large number of similar samples, it would be advisable to define a calibration file prior to the acquisition.

1. Open the template method from the Instrument window by using the *Method - Method Setup* command.
2. Go to the *Calculation* tab.

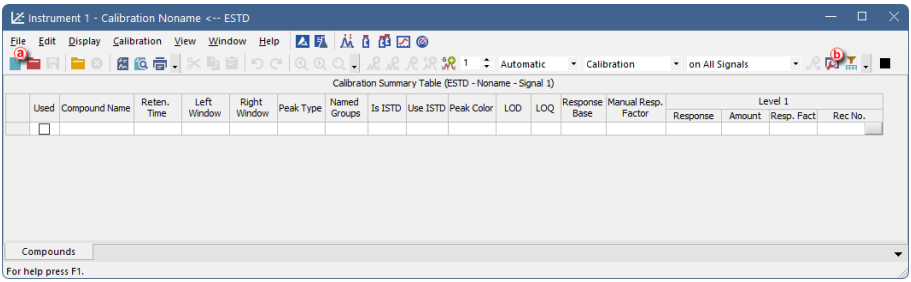


3. Click the *Set…* button ⓐ to select a calibration file for the method, or create a new calibration file by clicking the *New…* button ⓑ .
4. Change the default calibration type in the *Calculations* field ⓒ .
5. Click *OK* ⓓ to save the changes to the opened method.
6. You can modify the calibration file after acquisition. For more info go to ***Apply the calibration to a Chromatogram***.

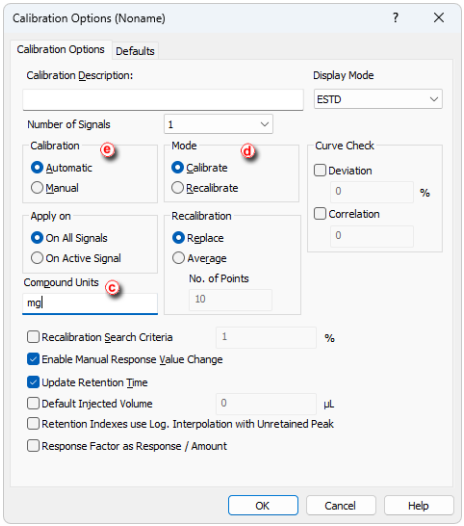
9.5 Calibrating with manually entered Response Factors

When using a free calibration, the amounts for each component are calculated using the *Response Factor* instead of a calibration curve.

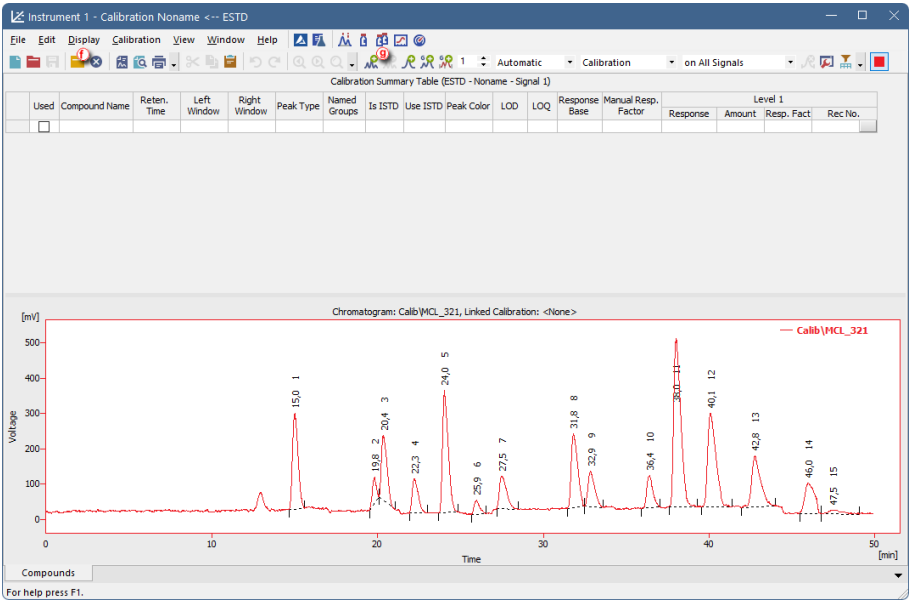
1. Create a new calibration file: select *File - New* or click ⓐ .



2. Open the Calibration Options dialog: choose *Calibration - Options…* or click . ⓑ



3. Check that the *Compound Units* ⓒ are set correctly, *Mode* to *Calibrate* ⓓ and *Calibration* option is set to *Automatic* ⓔ .
4. Open an integrated chromatogram of a standard (containing peaks of compounds of interest with a known concentration): select *File - Open Standard…* or click ⓕ on the *Calibration* window.
5. Add all peaks in the chromatogram of the calibration standard to the calibration file. Choose *Calibration - Add All* or click on ⓖ .

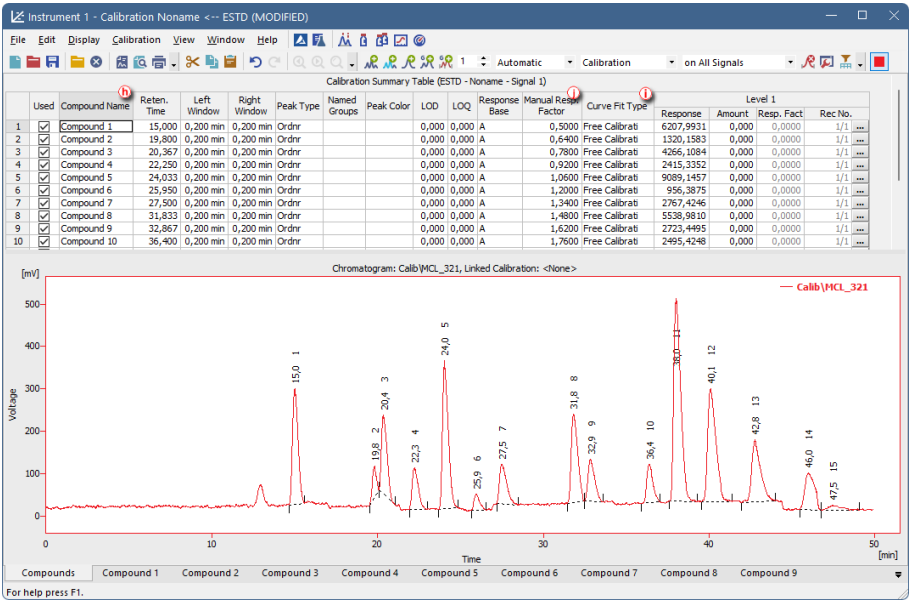


6. Name the peaks identified in the *Calibration Summary Table* ⓗ by their retention times by typing the *Compound Name* for each peak. No peak name may be used more than once.
7. Set the *Calibration Fit Type* to *Free Calibration* ⓘ for each one of the compounds in the *Calibration Summary Table*.

More Info:

Right click on the table and select the option *Set Columns...* to add the *Calibration Fit Type* column to the table. Alternatively, you can click on each compound tab at the bottom of the *Calibration* window and set the *Calibration Fit Type* from there.

8. Type in the *Manual Response Factor* for each one of the compounds ⓙ .

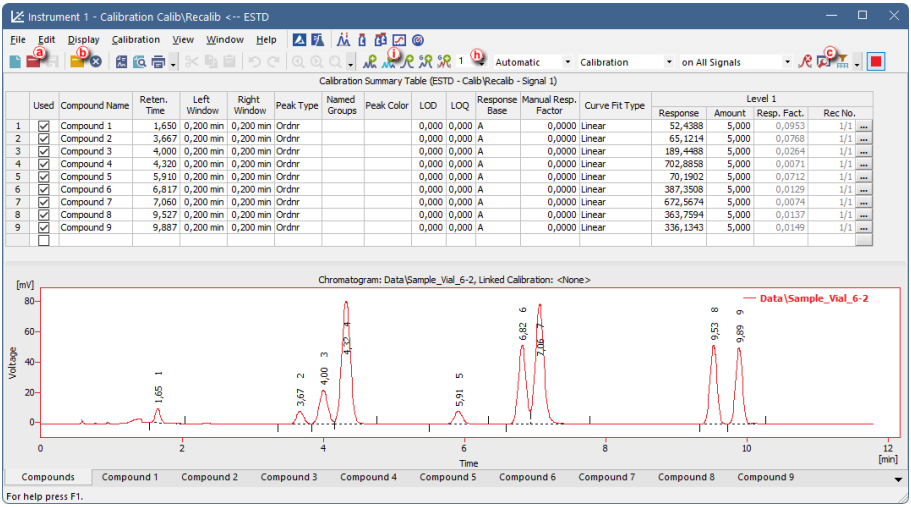


Caution: Calibration curve cannot be constructed when amount is set to zero for all levels. In case calibration curve could not be constructed for one of the compounds, no results are calculated for all of the compounds (as the total amount could be wrong). If you do not want to calculate amounts for all identified compounds, you could use free calibration with zero response factor.

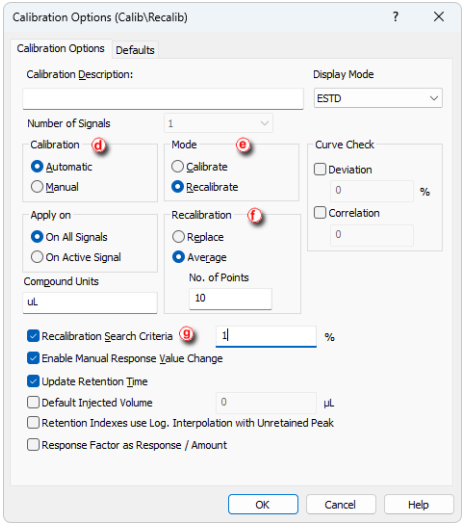
9.6 Recalibrating a calibration

You can modify an existing calibration by reloading peaks within one specific level using the option *Recalibrate*.

1. Open the Calibration window: choose *Window - Calibration* on the *Instrument* window or click .

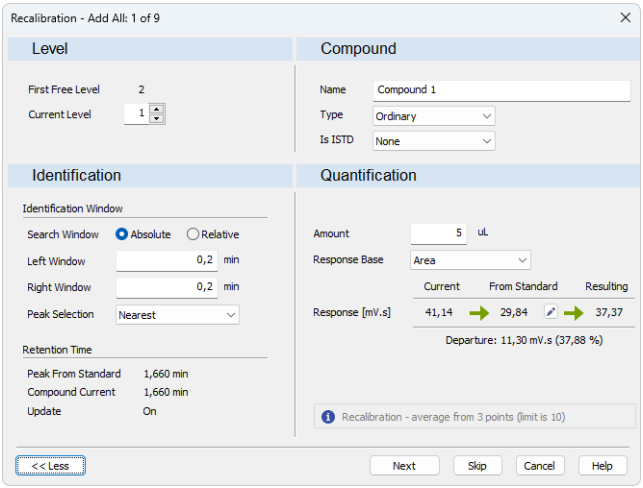


2. Open the calibration file: choose *File - Open…* or click ⓐ .
3. Open the calibration standard with which you want to recalibrate: select *File - Open Standard…* or click ⓑ .
4. Open the *Calibration Options* dialog: choose *Calibration - Options…* or click ⓒ .



5. Set the *Calibration* option to *Automatic* to add the peaks without modification or to *Manual* to modify them one by one ⓓ .

6. Set the *Mode* to *Recalibration* ⓔ .
7. Select how and whether the new values will be added in the *Calibration Options* dialog.
    - Choose *Replace* or *Average* option to decide what to do with new response values ⓕ .
    - *Recalibration Search Criteria* defines how much the original and new values can differ for the recalibration to be performed ⓖ .
8. Select the *Level* you wish to recalibrate ⓗ .
9. Add peaks to be recalibrated from the calibration standard to the calibration file on the *Calibration* window using *Calibration - Add Existing* or clicking on ⓘ .
10. If you selected *Manual* calibration:
    - Fill in any parameters related to the peak in the *Calibration - Add All* window. This window will open once for each one of the peaks processed.

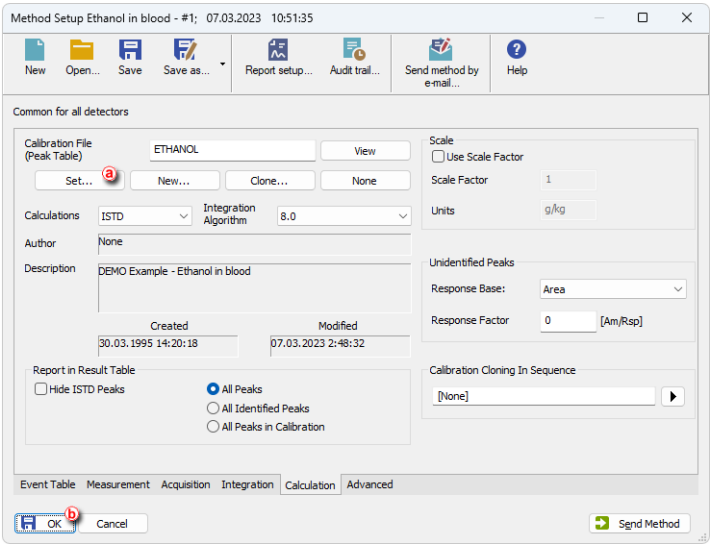


11. Save the calibration file: choose *File - Save* or click on .

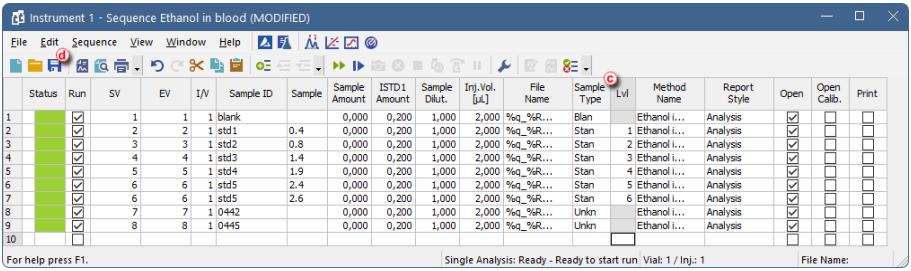
9.7 Automatic recalibration using a sequence

Here we describe how to add more data to a calibration point from more than one calibration standard chromatogram using a sequence.

1. Open the method file: use *File - Open Method…* on the Instrument window or click in the *Method Setup* dialog.
2. Go to the *Calculation* tab.



3. Connect the calibration file to the method by using the *Set…* ⓐ button.
4. Save the method file - click *OK* ⓑ or select *File - Save Method* or click .
5. Open the Sequence window: select *Analysis - Sequence* or click in the *Instrument* window.



6. Fill in the Sequence Table as described in the section ***Run a sequence***.
7. For the calibration standards, fill in the *Sample Type* and *Lvl* columns ⓒ in the *Sequence Table*.

More Info:

The *Sample Type* column for the calibration standard should be set to the *Standard* value, the *Lvl* column for each standard must have a value between *1* and *20*. For a blank sample, select *Blank* in the *Sample Type* column.

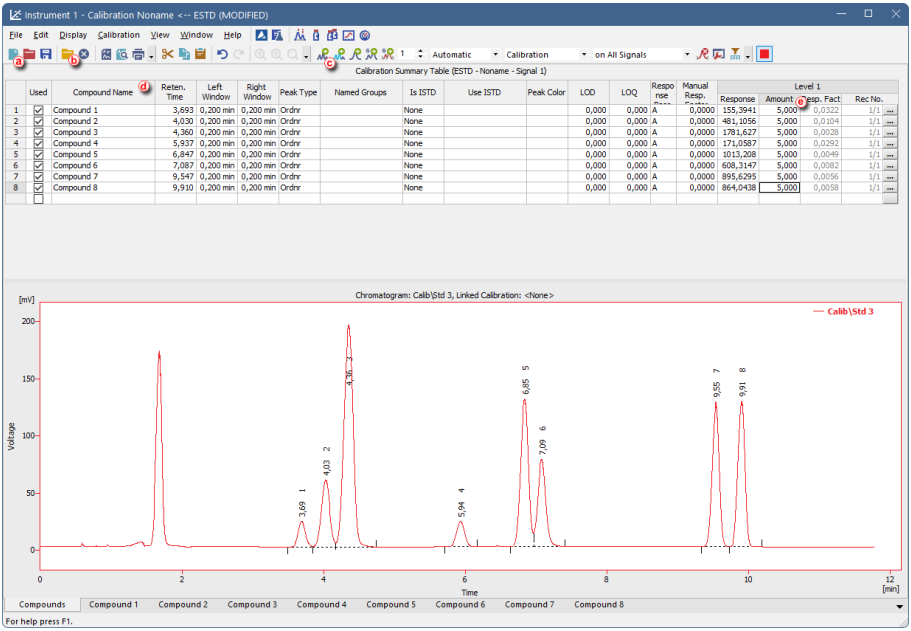
8. Save the sequence file: select *File - Save* or click . ⓓ

The file is now prepared so that the calibration standards measured according to their sequence rows will automatically recalibrate the calibration file.

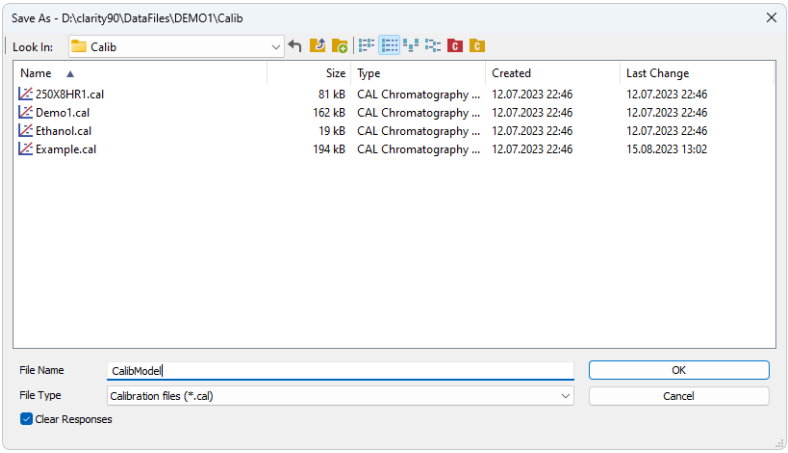
9.8 Creating model calibration to use in calibration cloning

This topic will show you how to prepare model calibration to be used during calibration cloning. If you have calibration already created from the previous measurement skip to step 4.

1. In the *Calibration* window, select *New* ⓐ calibration and *Open* ⓑ typical chromatogram as standard.
2. Use *Add All* ⓒ to add the integrated peaks to calibration.
3. Fill in the *Compound Names* ⓓ and the *Amounts* ⓔ for every level that will be used.



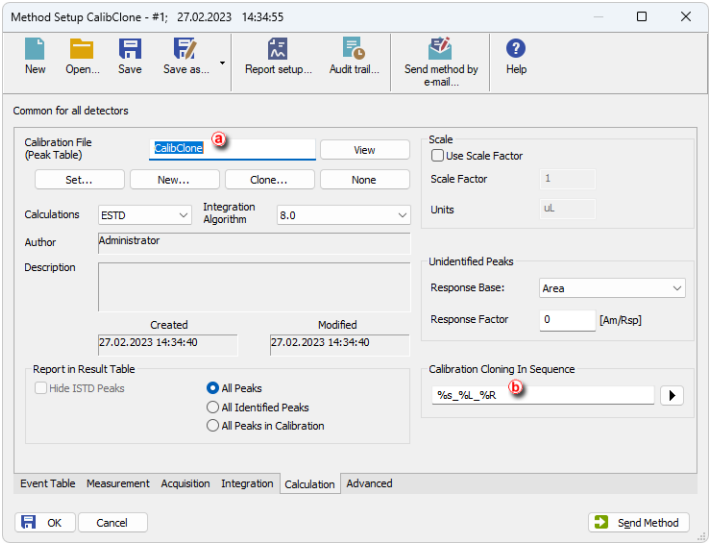
4. Select the *Save As* option. In the *Save As* dialog check the *Clear Responses* checkbox and save the calibration under a new name.



9.9 Calibrating using clone on first recalibration

Option *Clone on first recalibration* sets the sequence and calibration to the Safe Calibration Usage mode. This option will create a clone (copy) of calibration defined in the method upon completing the first row of the sequence. Cloned calibration is attached to each new chromatogram produced by the given sequence.

1. Open the *Method Setup* dialog - Calculation tab. Select *Method - Calculation* from the *Instrument* window.



2. Click on *Set...* button and select a calibration file ⓐ to be used during cloning at first recalibration.

   Note: This calibration will remain unchanged as newly created clone of the calibration will be used with new responses.

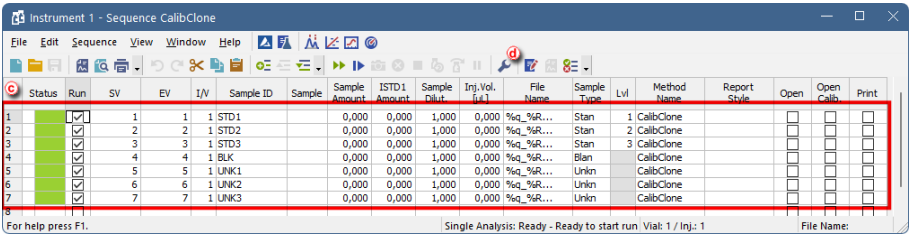
3. Create a custom name for the calibration files in *Calibration Cloning in Sequence* ⓑ as explained in ***Creating customized file names automatically***.

   Note: The name of the final calibration file will match just the content of this field - if you wish to include the name of the template calibration, include the name in this field again (e.g. "test - %s %L %R").

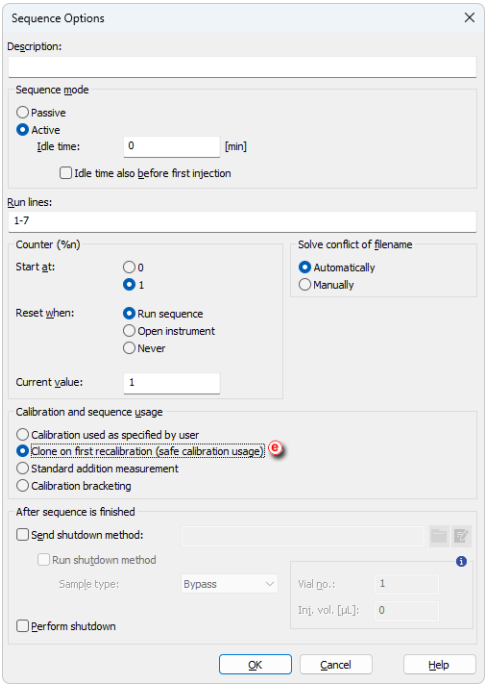
4. Follow the steps in ***Creating and running a sequence*** to create your sequence based on the example below.

   More Info:

   - Set the row/s for the standard/s at the beginning. ⓒ
   - Add a row for a blank, if you wish to.
   - Set the row/s for the unknown samples.



5. Click to open the *Sequence Options* Dialog ⓓ .
6. Select *Clone on first recalibration* ⓔ and click *OK*.



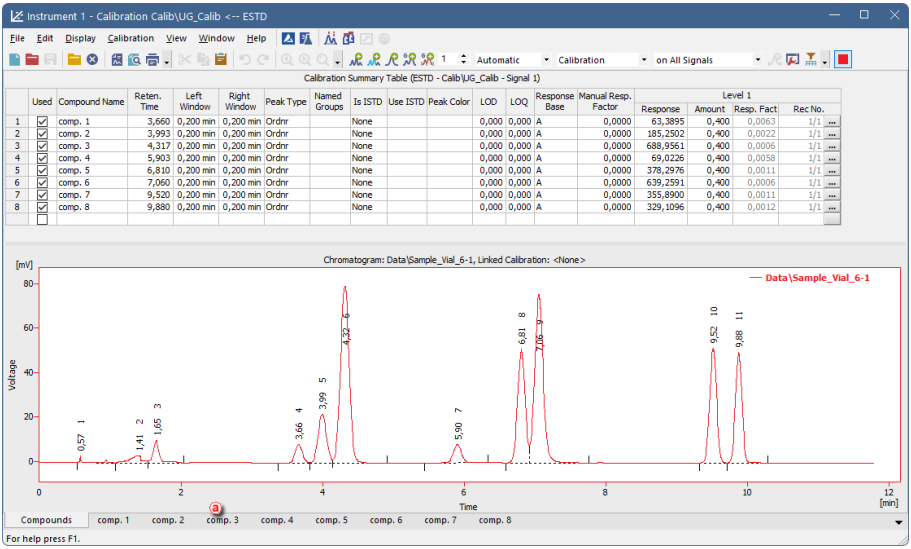
7. Run the sequence as explained in ***Creating and running a sequence***.

Note: The measured sequence can be reprocessed using Batch. For more info see the topic Reprocessing whole sequence while using calibration cloning.

9.10 Calibration adjustments

Following articles will describe how to perform some of the most common calibration modifications that can improve your calibration. Using this guide you manage to fit your calibration closer to your analytical application.

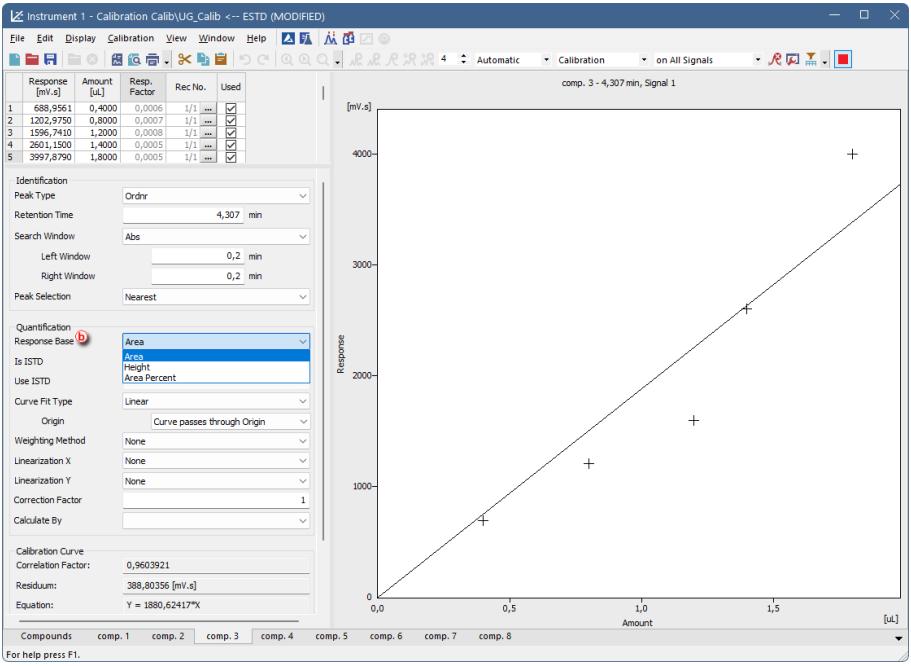
Once you added all chromatograms to your calibration and filled ***Amount*** values for all compounds and all levels, you can start to adjust it. Note that the ***Amount*** values are the values you know, because they originate from the concentration levels of your calibration solutions. Further adjustments of the calibration are available on the corresponding ***Compound Tabs*** at the bottom of the window. In our case, the Comp No. 3 ⓐ is edited.



	Used	Compound Name	Reten. Time	Left Window	Right Window	Peak Type	Named Groups	Is ISTD	Use ISTD	Peak Color	LOD	LOQ	Response Base	Manual Resp. Factor	Level 1 Response	Level 1 Amount	Level 1 Resp. Fact	Level 1 Rec No.
1	☑	comp. 1	3,660	0,200 min	0,200 min	Ordnr		None			0,000	0,000	A	0,0000	63,3895	0,400	0,0063	1/1
2	☑	comp. 2	3,993	0,200 min	0,200 min	Ordnr		None			0,000	0,000	A	0,0000	185,2502	0,400	0,0022	1/1
3	☑	comp. 3	4,317	0,200 min	0,200 min	Ordnr		None			0,000	0,000	A	0,0000	688,9561	0,400	0,0006	1/1
4	☑	comp. 4	5,903	0,200 min	0,200 min	Ordnr		None			0,000	0,000	A	0,0000	69,0226	0,400	0,0058	1/1
5	☑	comp. 5	6,810	0,200 min	0,200 min	Ordnr		None			0,000	0,000	A	0,0000	378,2976	0,400	0,0011	1/1
6	☑	comp. 6	7,060	0,200 min	0,200 min	Ordnr		None			0,000	0,000	A	0,0000	639,2591	0,400	0,0006	1/1
7	☑	comp. 7	9,520	0,200 min	0,200 min	Ordnr		None			0,000	0,000	A	0,0000	355,8900	0,400	0,0011	1/1
8	☑	comp. 8	9,880	0,200 min	0,200 min	Ordnr		None			0,000	0,000	A	0,0000	329,1096	0,400	0,0012	1/1

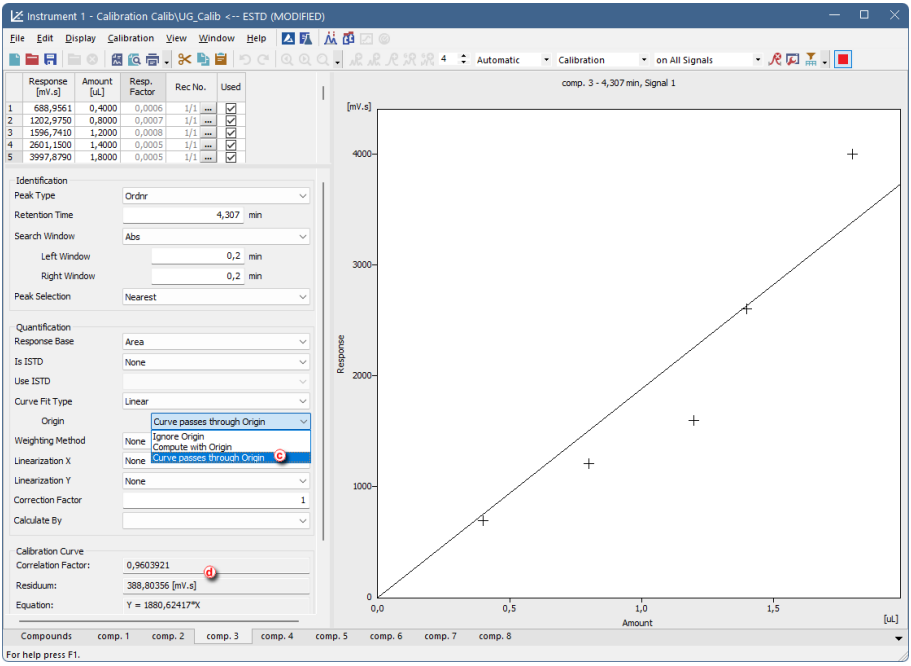
Modifying Response Base

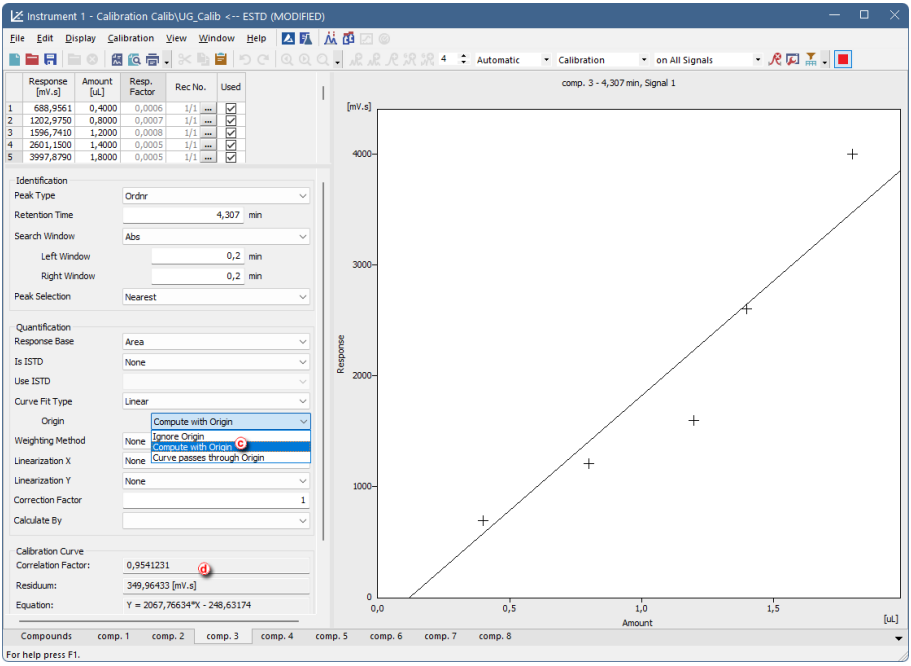
Set the *Response Base* ⓑ to modify whether the calibration curve will be calculated using *Area* or *Height* of the corresponding peak belonging to the specific compound. Changing the *Response Base* can help to create better fitting of the calibration to not well resolved peaks.

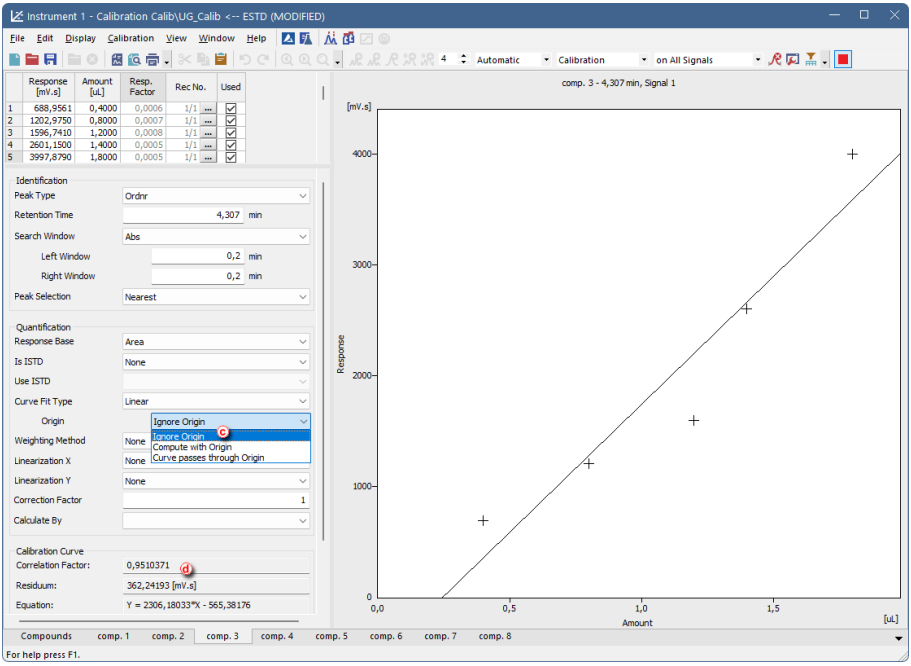


Modifying Origin

Another option how to improve your calibration is setting of *Origin* ⓒ and its incorporation or exclusion in calibration calculations. In **Clarity** there are three available setting options, as described on the following images. There is also demonstrated an effect of the *Origin* setting on calculated *Equation* and *Correlation factor* ⓓ .


Instrument 1 - Calibration Calib\UG_Calib <-- ESTD (MODIFIED)
File Edit Display Calibration View Window Help
Automatic
Calibration
on All Signals
Response [mV.s]
Amount [uL]
Resp. Factor
Rec No.
Used
1 688,9561 0,4000 0,0006 1/1
2 1202,9750 0,8000 0,0007 1/1
3 1596,7410 1,2000 0,0008 1/1
4 2601,1500 1,4000 0,0005 1/1
5 3997,8790 1,8000 0,0005 1/1
Identification
Peak Type Ordnr
Retention Time 4,307 min
Search Window Abs
Left Window 0,2 min
Right Window 0,2 min
Peak Selection Nearest
Quantification
Response Base Area
Is ISTD None
Use ISTD
Curve Fit Type Linear
Origin Curve passes through Origin
Ignore Origin
Compute with Origin
Curve passes through Origin
Weighting Method None
Linearization X None
Linearization Y None
Correction Factor 1
Calculate By
Calibration Curve
Correlation Factor: 0,9603921
Residuum: 388,80356 [mV.s]
Equation: Y = 1880,62417*X
comp. 3 - 4,307 min, Signal 1
[mV.s]
Response
Amount
[uL]
Compounds comp. 1 comp. 2 comp. 3 comp. 4 comp. 5 comp. 6 comp. 7 comp. 8
For help press F1.


Instrument 1 - Calibration Calib\UG_Calib <-- ESTD (MODIFIED)
File Edit Display Calibration View Window Help
Automatic
Calibration
on All Signals
Response [mV.s]
Amount [uL]
Resp. Factor
Rec No.
Used
1 688,9561 0,4000 0,0006 1/1
2 1202,9750 0,8000 0,0007 1/1
3 1596,7410 1,2000 0,0008 1/1
4 2601,1500 1,4000 0,0005 1/1
5 3997,8790 1,8000 0,0005 1/1
Identification
Peak Type Ordnr
Retention Time 4,307 min
Search Window Abs
Left Window 0,2 min
Right Window 0,2 min
Peak Selection Nearest
Quantification
Response Base Area
Is ISTD None
Use ISTD
Curve Fit Type Linear
Origin Compute with Origin
Ignore Origin
Compute with Origin
Curve passes through Origin
Weighting Method None
Linearization X None
Linearization Y None
Correction Factor 1
Calculate By
Calibration Curve
Correlation Factor: 0,9541231
Residuum: 349,96433 [mV.s]
Equation: Y = 2067,76634*X - 248,63174
comp. 3 - 4,307 min, Signal 1
[mV.s]
Response
Amount
[uL]
Compounds comp. 1 comp. 2 comp. 3 comp. 4 comp. 5 comp. 6 comp. 7 comp. 8
For help press F1.



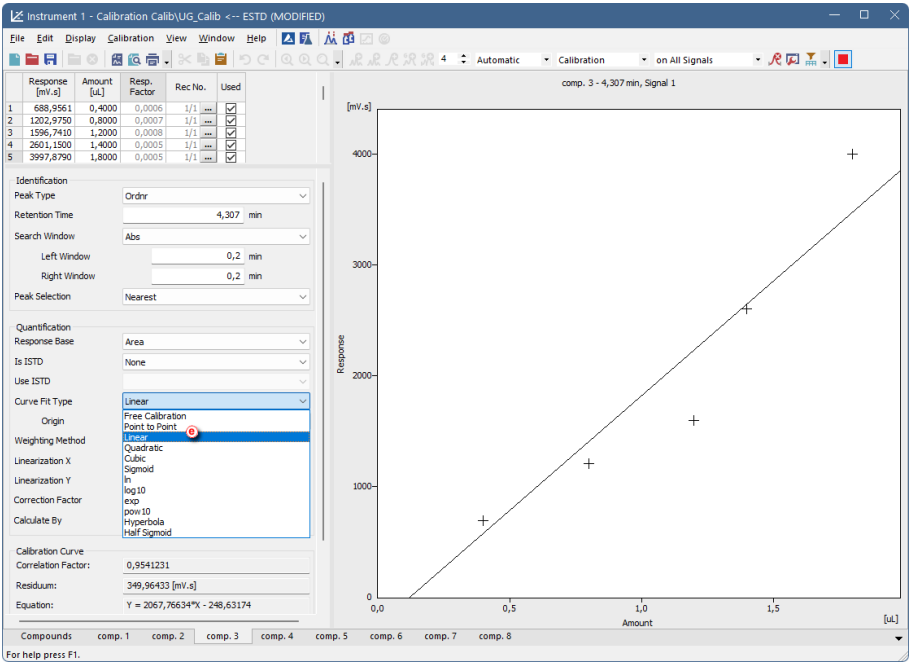
As you can see from the *Correlation factor* ⓓ values, changing the *Origin* setting from the *Curve Passes through Origin* to *Compute with Origin* improved the curve fit. However, the option *Ignore Origin* has proved to be the best match for the used detector.

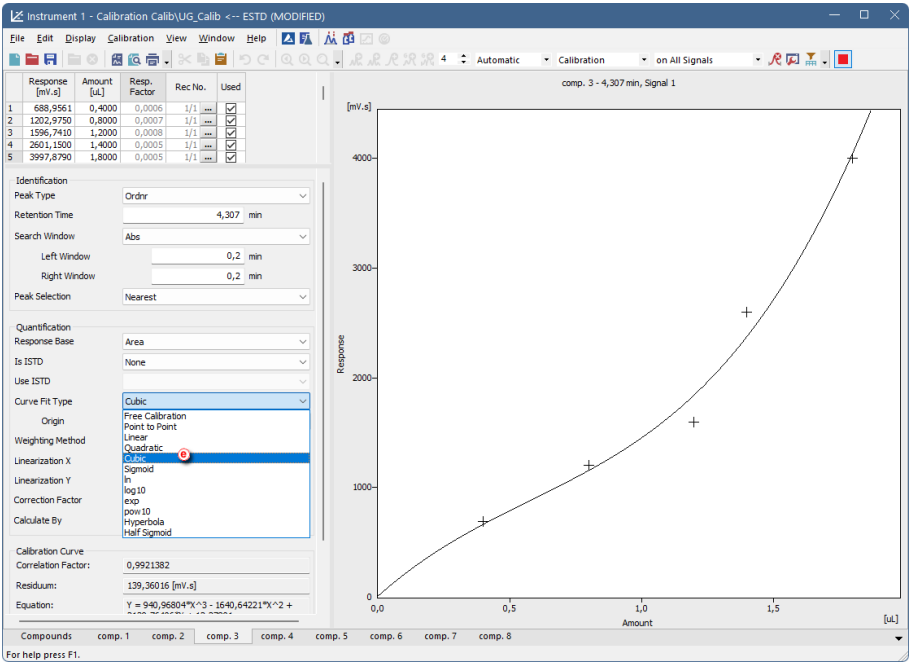
Modifying Curve Fit Type

Following images demonstrate how to modify *Curve Fit Type* ⓔ from *Linear* to *Cubic*. *Cubic Curve Fit Type* is used as an example of non-linear *Curve Fit Types*.

- Linear calibration curve is commonly used for detectors with linear response, such as Flame Ionization Detector (FID) or Refractive Index Detector (RID).
- Non-linear calibration curve is typical for detectors such as Electron Capture Detector (ECD) or Evaporative Light Scattering Detector (ELSD).

When changing *Curve Fit Type* ⓔ pay also attention to the values of the calculated *Equation* and *Correlation Factor*. Increasing value of the *Correlation Factor* indicates better selected *Curve Fit Type* for the measured data.

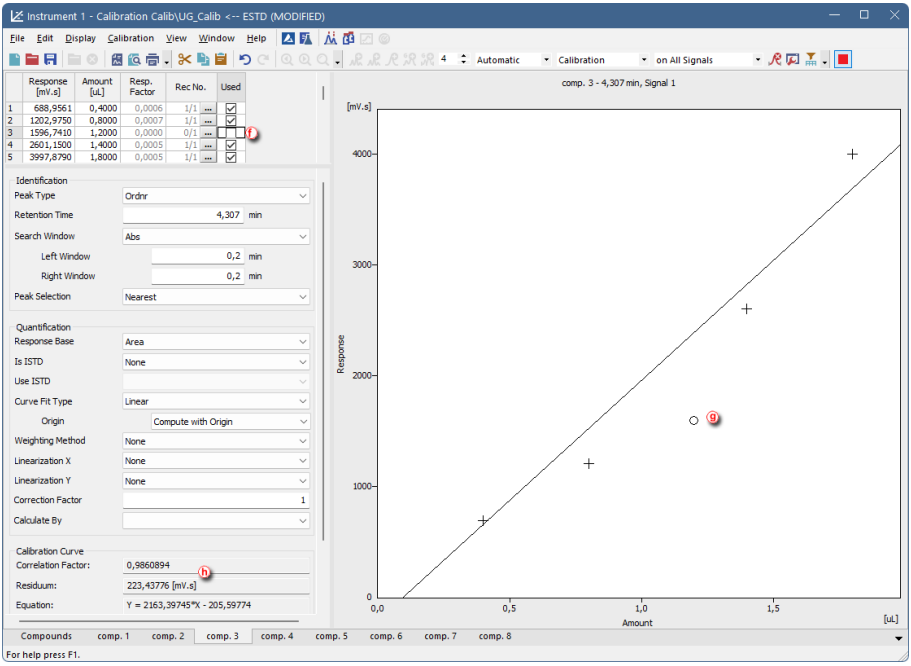

Instrument 1 - Calibration Calib\UG_Calib <-- ESTD (MODIFIED)
File Edit Display Calibration View Window Help
Automatic
Calibration
on All Signals
Response [mV.s]
Amount [uL]
Resp. Factor
Rec No.
Used
1 688,9561 0,4000 0,0006 1/1
2 1202,9750 0,8000 0,0007 1/1
3 1596,7410 1,2000 0,0008 1/1
4 2601,1500 1,4000 0,0005 1/1
5 3997,8790 1,8000 0,0005 1/1
Identification
Peak Type Ordnr
Retention Time 4,307 min
Search Window Abs
Left Window 0,2 min
Right Window 0,2 min
Peak Selection Nearest
Quantification
Response Base Area
Is ISTD None
Use ISTD
Curve Fit Type Linear
Origin
Weighting Method
Linearization X
Linearization Y
Correction Factor
Calculate By
Free Calibration
Point to Point
Linear
Quadratic
Cubic
Sigmoid
ln
log10
exp
pow10
Hyperbola
Half Sigmoid
Calibration Curve
Correlation Factor: 0,9541231
Residuum: 349,96433 [mV.s]
Equation: Y = 2067,76634*X - 248,63174
comp. 3 - 4,307 min, Signal 1
[mV.s]
Response
Amount
[uL]
Compounds comp. 1 comp. 2 comp. 3 comp. 4 comp. 5 comp. 6 comp. 7 comp. 8
For help press F1.



As you can see, changing the *Curve Fit Type* from *Linear* to *Cubic* increased the *Correlation Factor* from 0.954 to 0.992.

Excluding of Measured Point from Calibration

Clarity also allows excluding any point from calibration. This is helpful in case that the specific measurement went wrong. Following images describe how to do that. Please keep in mind that the excluded point isn't deleted, the point is still part of the calibration, however, it is omitted from calculation of calibration curve. To exclude the selected measurement point from the calibration simply uncheck that measurement in the *Used* ⓕ column. The excluded point in the graph of calibration curve will be changed from cross to empty circle ⓖ and calibration curve *Equation* and *Correlation Factor* ⓗ will be recalculated.



As you can see, excluding the incorrectly measured calibration point resulted in more accurate calibration.

9.11 Calibration - Advanced Topics

This chapter contains collection of more advanced SOPs topics covering calibration usage in Clarity.

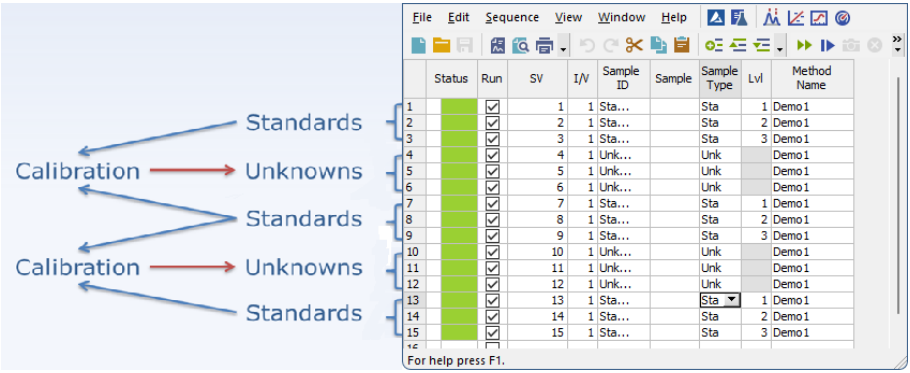
9.11.1 Compensating for response drift using bracketing

Bracketing is a direct calibration method used to compensate for the variation in instrument response with time. Typical use is for detector response deteriorating or sample containing compounds staining or interacting with the column. Bracketing is not helpful for random variations. Bracketing may be in place when 2 calibration curves measured on the same series of standards have a stable trend and good correlation, but they are not the same.

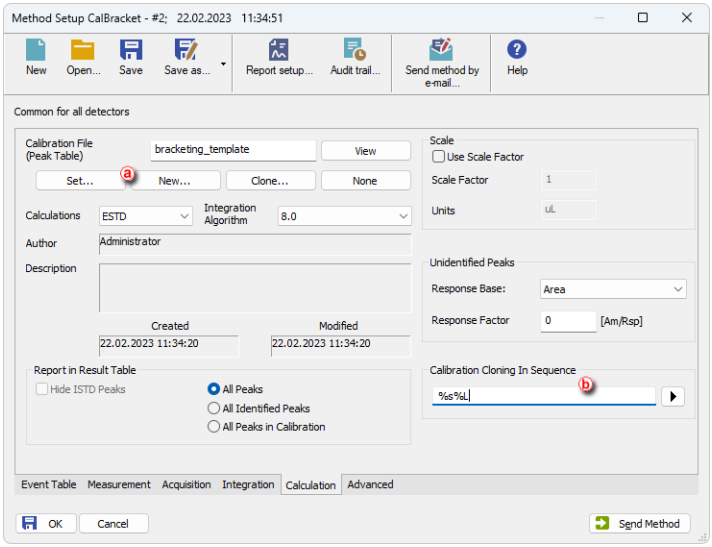
To use bracketing in sequence, the order of the rows must be standards, then unknowns, then standards again. Usually, two standards are used; more standards could be used if the measuring instrument has a non-linear response. Samples are evaluated by a calibration which is created by averaging the standards before and after the unknown. As every unknown sample series is demarcated by calibration standards it uses a single calibration. Calibration will be cloned from the previous calibration clone whenever an unknown sample or blank follows the calibration standard. The newly cloned calibration file has all responses cleared - apart from

the responses from the last series of calibration standards (immediately preceding the current unknown samples).

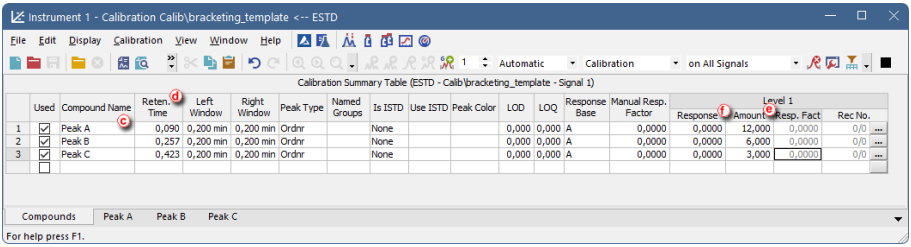
Bracketing in Clarity

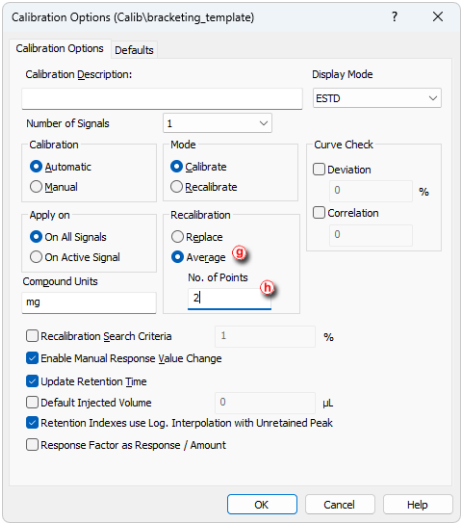


1. Open the method that will be used in the sequence: click *File - Open Method…* from the *Instrument* window.
2. Navigate to *Calculation* tab.
3. Set the template calibration as Calibration File: click *Set...* ⓐ and select the calibration. Note that this calibration will remain unchanged, newly created clone of the calibration will be used with new responses.
4. Set the name of the cloned calibration: set the name in *Calibration Cloning In Sequence* ⓑ. You can use predefined parameters (refer to ***Creating customized file names automatically***).



5. Click *OK* and save the modified method.
6. Open Calibration window and the template calibration: click *Window - Calibration* from the *Instrument* window and *File - Open...* in the *Calibration* window, select the template calibration (in this case bracketing_ template.cal).
7. Set *Compound Names* ⓒ , *Retention Time* ⓓ , *Amounts* ⓔ , etc., but no *Responses* ⓕ (you can use previously measured standard to get the Retention Times).
8. Update *Calibration Options*: Set *Recalibration* to *Average*ⓖ and *No. of Points* to 2ⓗ (no more points needed as they would not be applied anyway).
9. Save changes and close the calibration.

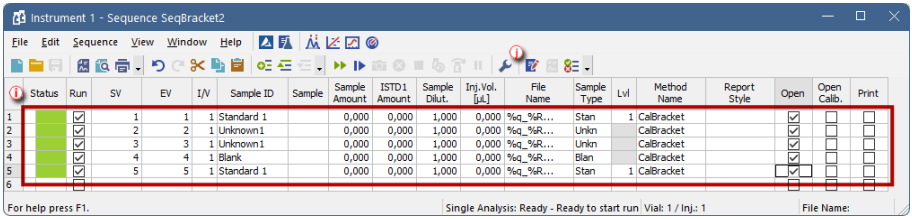




10. Open *Sequence* window and create a new sequence: click *Analysis - Sequence* and then icon.
11. Set the sequence according to the following steps: (for more details about creating the sequence refer to ***Creating and running a sequence***)
    - Set the row/s for the standard/s. ⓘ
    - Add a row for a blank, if you wish to.
    - Set the row/s for the unknown samples.
    - Repeat the row/s for the standard/s.

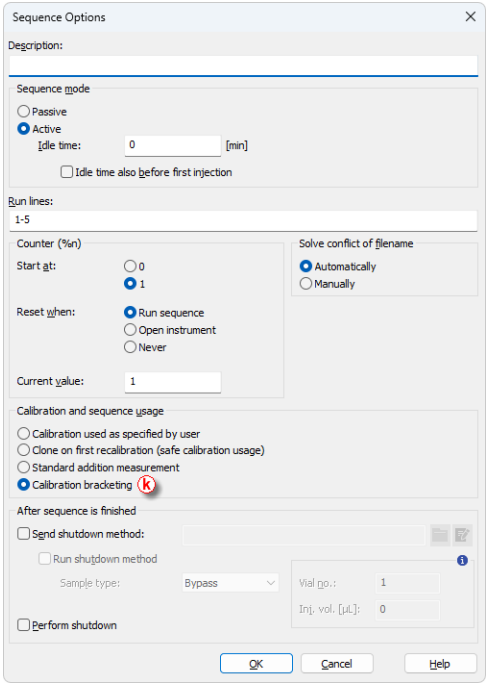
Note: The sequence must start and end with a row with *Standard Sample Type*.

- Repeat the previous four steps for every "bracket" of unknown samples you wish to add.



12. Set the sequence to operate in the calibration bracketing mode:
    - Click icon to open the *Sequence Options* dialog. ⓙ
    - Check *Calibration Bracketing*. ⓚ
    - Click *OK*.

Note: If your sequence is using multiple methods, calibration using bracketing is still possible to use but make sure that the Calculation tab is exactly the same for all the methods used in the sequence.



13. Run the sequence (for more details about running the sequence refer to *Creating and running a sequence)* .
14. The results shown while the sequence is running are recalculated at the end of each bracket, when the standard after the unknown sample is acquired.

The calibration used is an average of the two calibrations, before and after the unknown.

Note: The measured sequence can be reprocessed using Batch. For more info see the topic Reprocessing whole sequence while using calibration cloning.

9.11.2 Creating a Multisignal Calibration

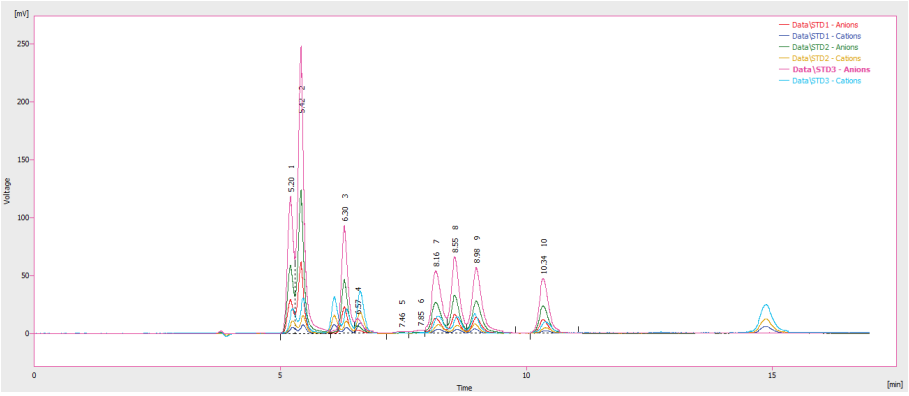
This topic describes how to construct a multisignal calibration in Clarity. Below is an example of creating and constructing a three (concentration) level calibration which provides calculation parameters for calculating results in two- signal chromatograms. This procedure should be utilized when different compounds are detected on each signal. In case all signals detect the same compounds there is no need to use *On Active Signal* option, unless you don't want to quantify all of the compounds on some signals.

Note: For cases there is need to create and construct more than a two-signal calibration, the applied approach remains the same. It varies from the below procedure slightly. These aspects are reflected in the respective steps of this topic.

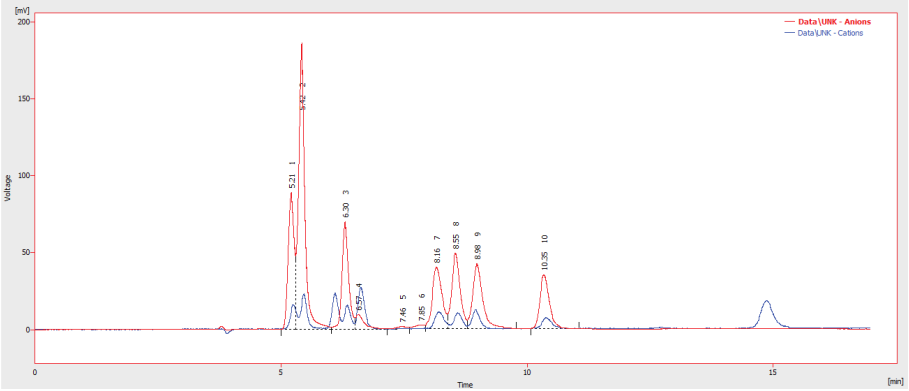
This guide is based on two-signal chromatograms of standards with simulated data (for demonstration purposes it can be said that first signal detects anions and second detects cations) on three concentration levels (5 000 ppm, 10 000 ppm and 20 000 ppm) and one sample chromatogram. All chromatograms used in this guide have been integrated in manner to fit the demonstration purpose.

Prerequisites:

- Integrated chromatograms of standards.

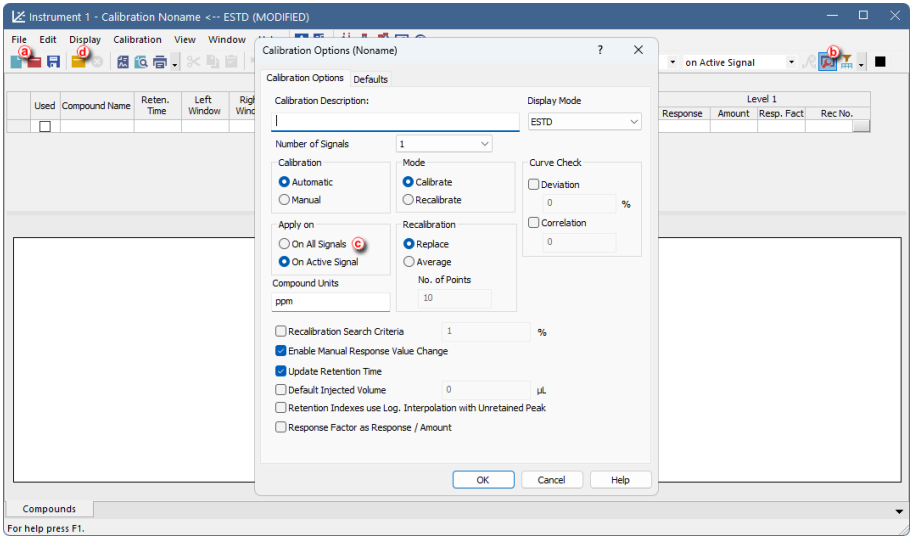


- Integrated chromatogram of sample.



1. Open the Calibration window: select *Window - Calibration* in the *Instrument* window or click .
2. Create a new calibration file: select *File - New* or click ⓐ .
3. Open the Calibration Options dialog: select *Calibration - Options…* or click ⓑ .
4. Set section *Apply on* to option *On Active Signal* field ⓒ .

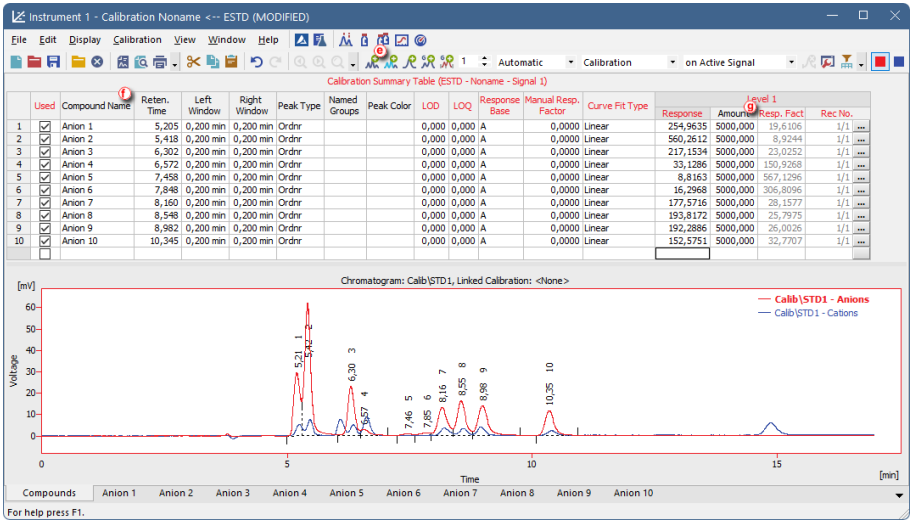
Note: As mentioned in the beginning. If all peaks in the standard chromatogram represent the same compound across all signals it is viable to change the option to *On All Signals*. In such case there is no difference in working with multisignal and single signal calibration.



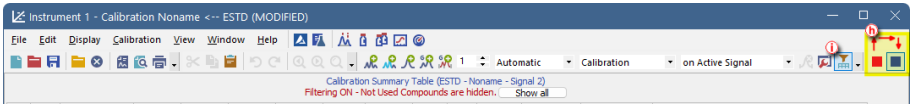
5. Open calibration standard: select *File - Open Standard…* or click ⓓ to open measured and integrated chromatogram with the lowest concentration level where all peaks are available.
6. Make sure the upper toolbar displays *on Active Signal*. It is also recommended to make sure you are currently on the first signal as well (focused red square in right part of the upper toolbar).
7. Fill the *Calibration Summary Table* with peaks from the currently selected signal in chromatogram using the *Add All* icon ⓔ .

   Note: If only some peaks should evaluated it can be done by using *Add Peak* on peaks that need to be included in calibration.

8. Rename automatically pre-filled names of peaks ⓕ .
9. Enter values for given standard into the *Amount* column ⓖ .

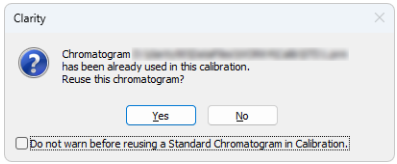


10. Switch to the second signal in chromatogram to make it active ⓗ .
11. Optional: Switch the table filter on using *Filter Not Used Compounds* icon ⓘ . With this option enabled, rows with not *Used* compounds on given signal are hidden, which makes it easier to navigate *Calibration Summary Table*.



12. Fill the *Calibration Summary Table* with peaks from the currently selected (second) signal in chromatogram using *Add All* icon. Dialog will be invoked after clicking *Add All* icon questioning if an already used chromatogram should be reused for this calibration. It is necessary to confirm the reuse by clicking *Yes* button.

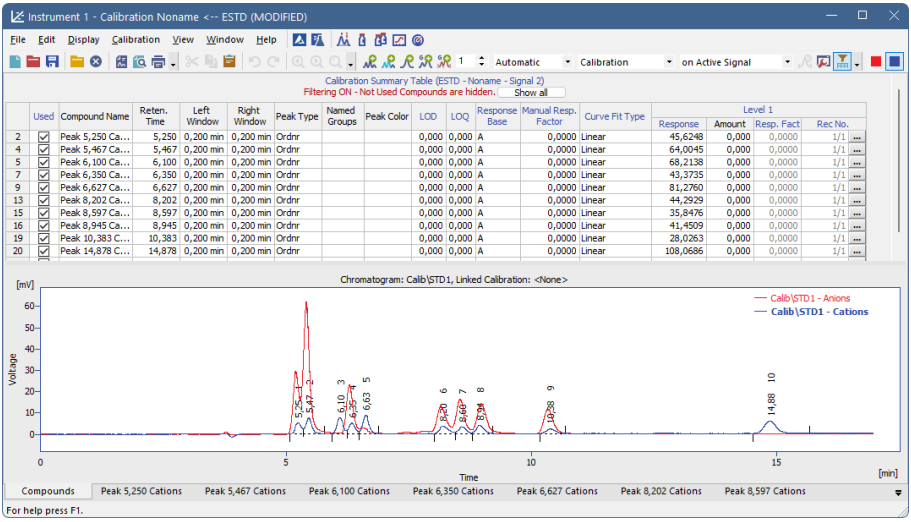
Note: In case of constructing calibration with more than two signals, steps 12 and 14 have to repeated as many times as necessary in order to fill in peaks from all signals to *Calibration Summary Table*.



Note:

This dialog will be invoked multiple times based on number of signals in calibration for calibration with more than two signals. It is possible to switch off invoking of the dialog using the checkbox in the bottom of the dialog.

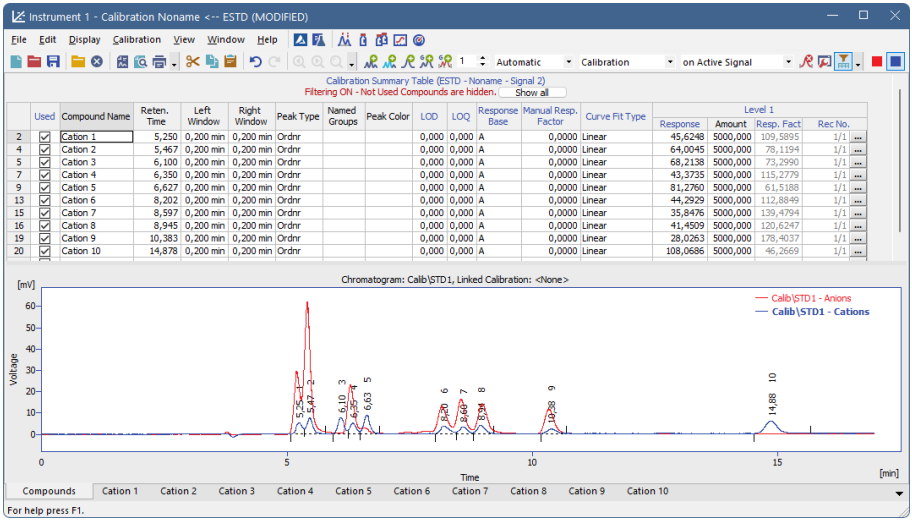
Note: Which signal of chromatogram is active and is being worked with is given by the *Calibration Summary Table* title's color and displayed number of currently active signal.



	Used	Compound Name	Reten. Time	Left Window	Right Window	Peak Type	Named Groups	Peak Color	LOD	LOQ	Response Base	Manual Resp. Factor	Curve Fit Type	Level 1 Response	Level 1 Amount	Level 1 Resp. Fact	Level 1 Rec No.
2	☑	Peak 5,250 Ca…	5,250	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	45,6248	0,000	0,0000	1/1
4	☑	Peak 5,467 Ca…	5,467	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	64,0045	0,000	0,0000	1/1
5	☑	Peak 6,100 Ca…	6,100	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	68,2138	0,000	0,0000	1/1
7	☑	Peak 6,350 Ca…	6,350	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	43,3735	0,000	0,0000	1/1
9	☑	Peak 6,627 Ca…	6,627	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	81,2760	0,000	0,0000	1/1
13	☑	Peak 8,202 Ca…	8,202	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	44,2929	0,000	0,0000	1/1
15	☑	Peak 8,597 Ca…	8,597	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	35,8476	0,000	0,0000	1/1
16	☑	Peak 8,945 Ca…	8,945	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	41,4509	0,000	0,0000	1/1
19	☑	Peak 10,383 C…	10,383	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	28,0263	0,000	0,0000	1/1
20	☑	Peak 14,878 C…	14,878	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	108,0686	0,000	0,0000	1/1

13. Rename the automatically pre-filled names in the *Calibration Summary Table* column.
14. Enter values into *Amount* column. The first concentration level is finished now. It is possible to proceed building up to next levels of calibration.

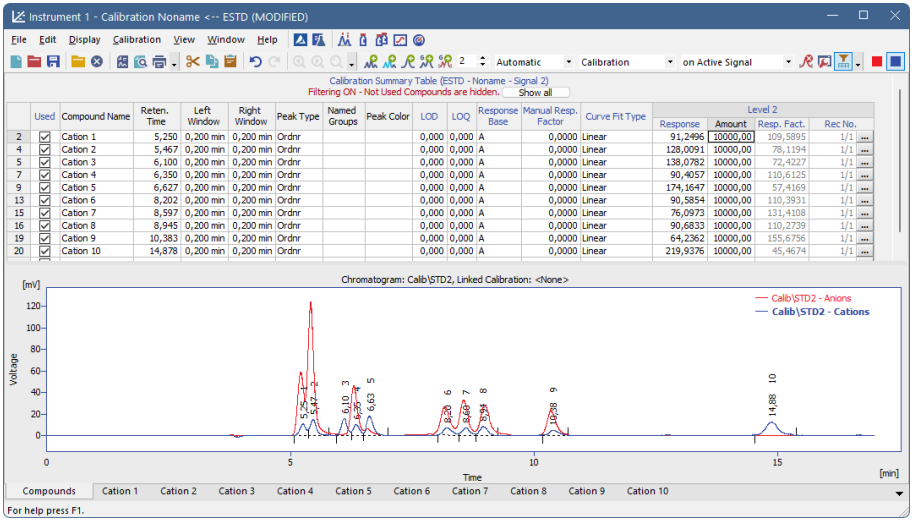
Note: All rows of *Amount* column have to be filled in.



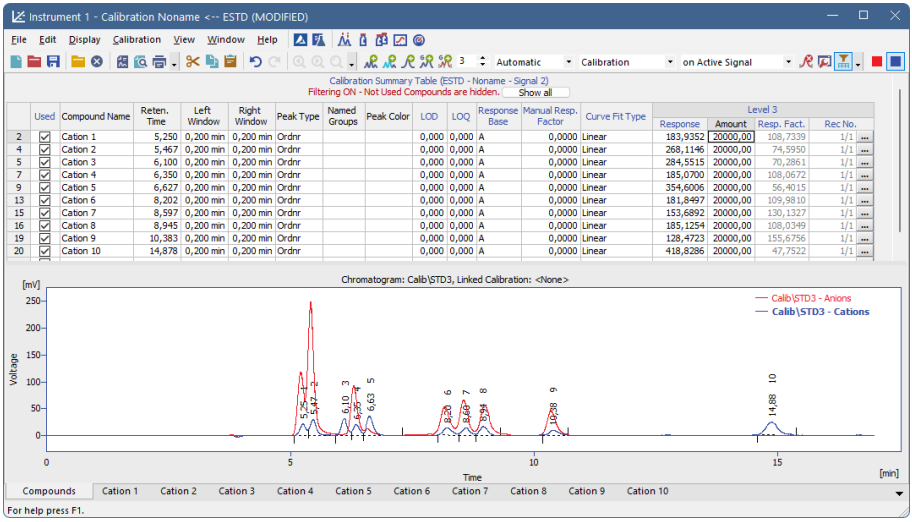
	Used	Compound Name	Reten. Time	Left Window	Right Window	Peak Type	Named Groups	Peak Color	LOD	LOQ	Response Base	Manual Resp. Factor	Curve Fit Type	Level 1 Response	Level 1 Amount	Level 1 Resp. Fact	Level 1 Rec No.
2	☑	Cation 1	5,250	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	45,6248	5000,000	109,5895	1/1
4	☑	Cation 2	5,467	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	64,0045	5000,000	78,1194	1/1
5	☑	Cation 3	6,100	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	68,2138	5000,000	73,2990	1/1
7	☑	Cation 4	6,350	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	43,3735	5000,000	115,2779	1/1
9	☑	Cation 5	6,627	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	81,2760	5000,000	61,5188	1/1
13	☑	Cation 6	8,202	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	44,2929	5000,000	112,8849	1/1
15	☑	Cation 7	8,597	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	35,8476	5000,000	139,4794	1/1
16	☑	Cation 8	8,945	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	41,4509	5000,000	120,6247	1/1
19	☑	Cation 9	10,383	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	28,0263	5000,000	178,4037	1/1
20	☑	Cation 10	14,878	0,200 min	0,200 min	Ordnr			0,000	0,000	A	0,0000	Linear	108,0686	5000,000	46,2669	1/1

15. Open chromatogram of standard of the next (second) concentration level.
16. Fill the next level of *Calibration Summary Table* with Responses for each signal following the same steps as for Level 1 (Steps 8-16, renaming the compounds is omitted as the names carry over from previous levels).

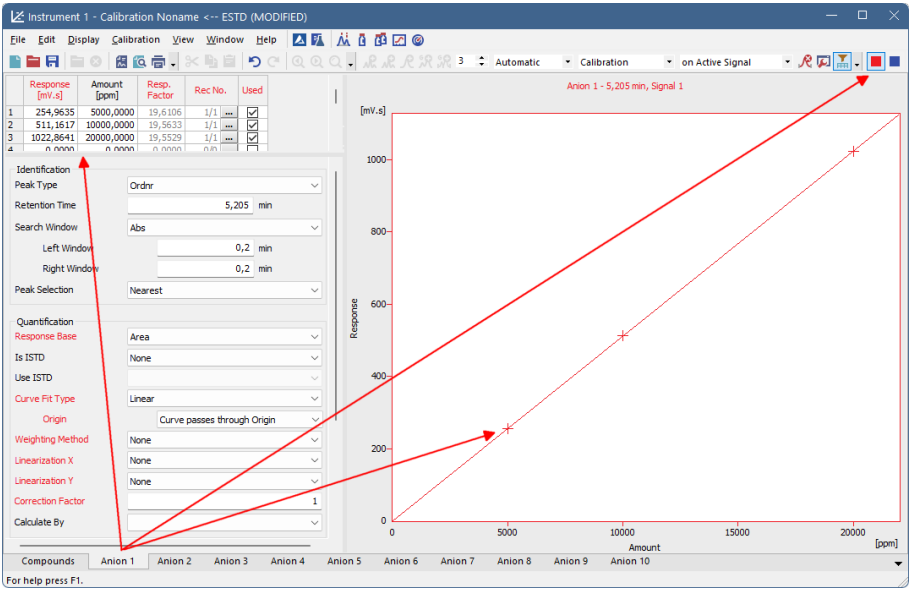
Note: *Add Existing* can be used instead of *Add All*. This command only adds response for peaks that are preset on first level. This means that extra peaks (or peaks chosen not to be evaluated) that are present in the second (and later) standards, but not in the first one will not be added.

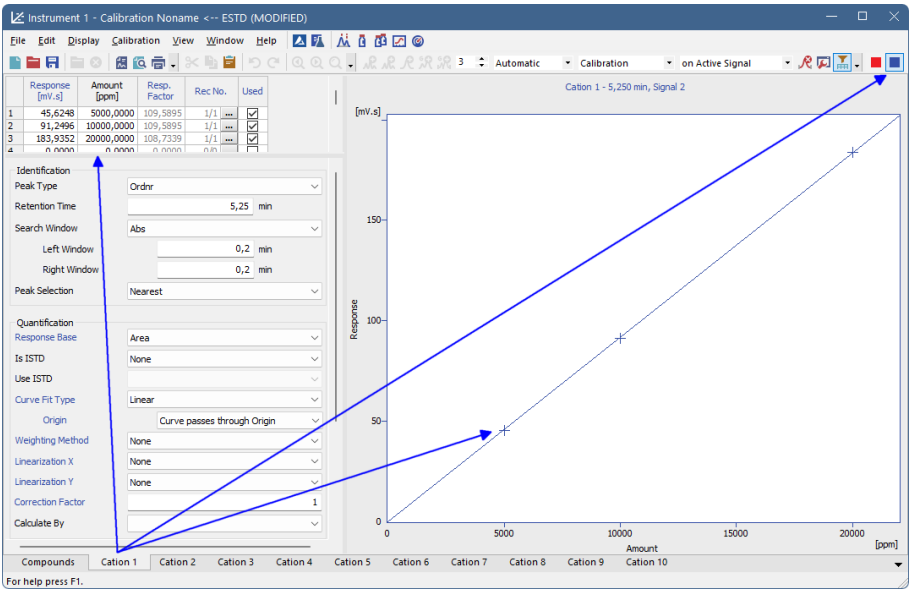


17. Open chromatogram of standard of the next (third) concentration level.
18. Fill the *Calibration Summary Table* in the same manner as for second level (step 18).



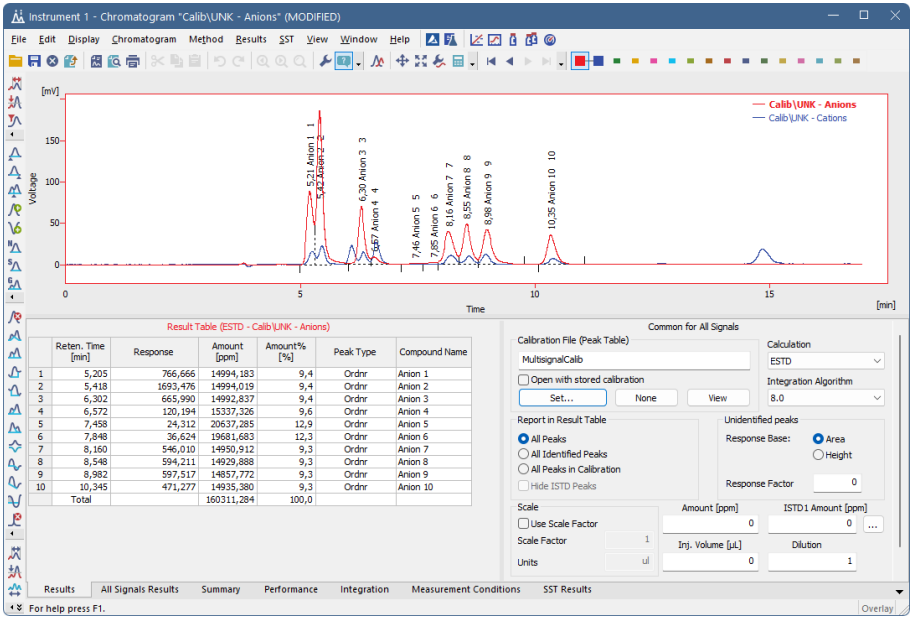
Note: It is possible to review calibration curves for each compound on the respective tabs of individual compounds. Notice that calibration curve are displayed only for valid signal (When filtering is enabled tabs of individual compounds are displayed only on signal where they are used). Signal selection is located in right upper corner of the *Calibration Window*.

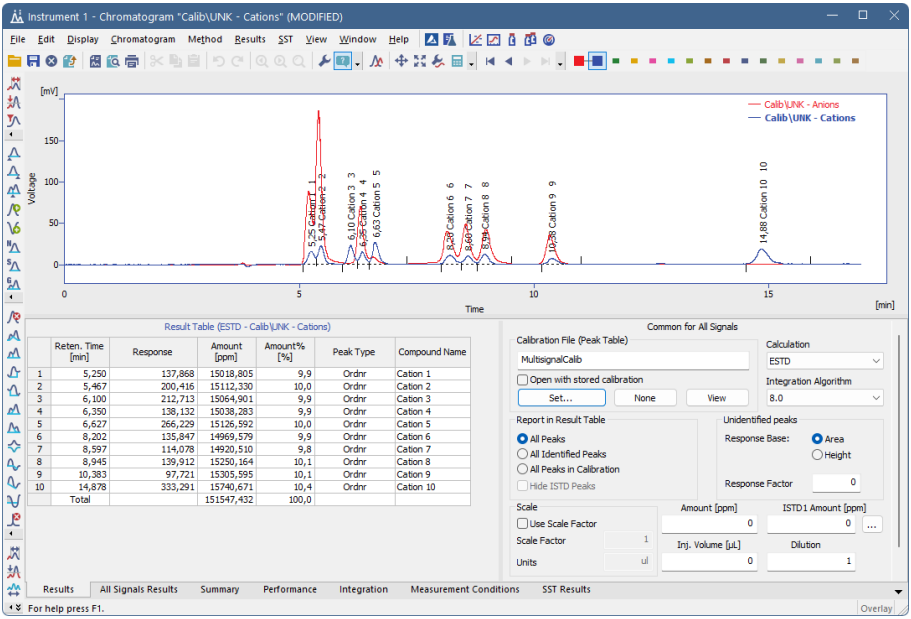


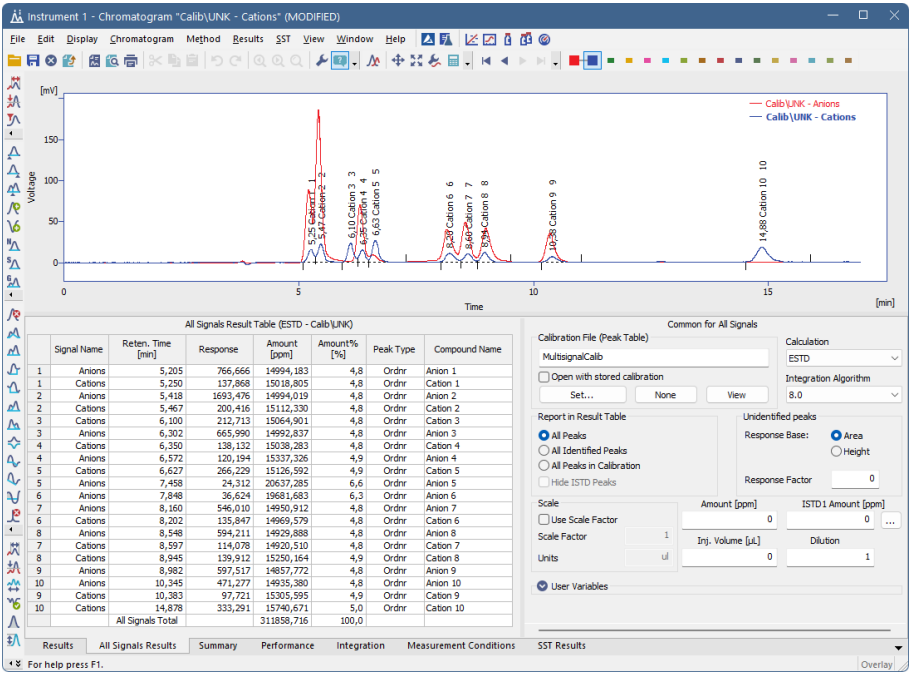


19. When calibration is finished do not forget to save it using select *File - Save* or click on .

20. For calculating result in sample open the **Chromatogram window** select *Window - Chromatogram* on the *Instrument* window or click on  and open chromatogram of sample and link the calibration to chromatogram.
21. Review results on *Results Table* of each individual signal or review results for all signals on *All Signals Results* Table.

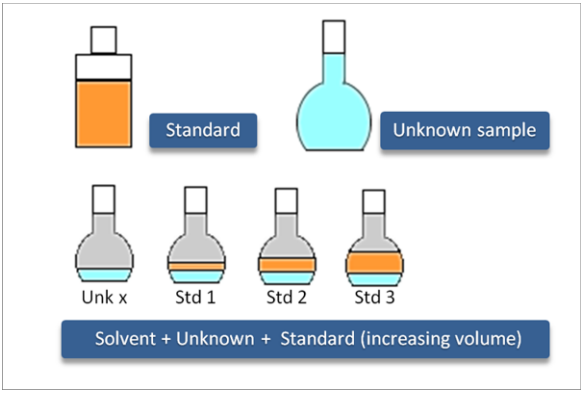



Instrument 1 - Chromatogram "Calib\UNK - Cations" (MODIFIED)
File Edit Display Chromatogram Method Results SST View Window Help
[mV]
Voltage
Time
[min]
Calib\UNK - Anions
Calib\UNK - Cations
5,25 Cation 1
5,47 Cation 2
6,10 Cation 3
6,35 Cation 4
6,63 Cation 5
8,20 Cation 6
8,40 Cation 7
8,94 Cation 8
10,38 Cation 9
14,88 Cation 10
Result Table (ESTD - Calib\UNK - Cations)
Reten. Time [min] | Response | Amount [ppm] | Amount% [%] | Peak Type | Compound Name
1 5,250 137,868 15018,805 9,9 Ordnr Cation 1
2 5,467 200,416 15112,330 10,0 Ordnr Cation 2
3 6,100 212,713 15064,901 9,9 Ordnr Cation 3
4 6,350 138,132 15038,283 9,9 Ordnr Cation 4
5 6,627 266,229 15126,592 10,0 Ordnr Cation 5
6 8,202 135,847 14969,579 9,9 Ordnr Cation 6
7 8,597 114,078 14920,510 9,8 Ordnr Cation 7
8 8,945 139,912 15250,164 10,1 Ordnr Cation 8
9 10,383 97,721 15305,595 10,1 Ordnr Cation 9
10 14,878 333,291 15740,671 10,4 Ordnr Cation 10
Total 151547,432 100,0
Common for All Signals
Calibration File (Peak Table)
MultisignalCalib
Open with stored calibration
Set... None View
Calculation
ESTD
Integration Algorithm
8.0
Report in Result Table
All Peaks
All Identified Peaks
All Peaks in Calibration
Hide ISTD Peaks
Unidentified peaks
Response Base: Area Height
Response Factor 0
Scale
Use Scale Factor
Scale Factor 1
Units ul
Amount [ppm] 0
Inj. Volume [µL] 0
ISTD1 Amount [ppm] 0
Dilution 1
Results All Signals Results Summary Performance Integration Measurement Conditions SST Results
For help press F1.
Overlay

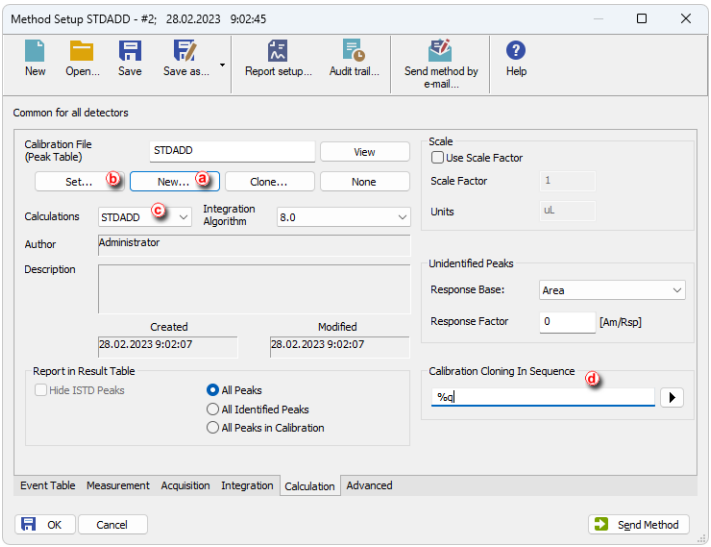


9.11.3 Improving quantification with the standard addition method

Standard addition is a quantification approach (similar to ESTD or ISTD) useful in case the sample matrix is complex and when it influences responses of analytes. By spiking samples with a series of increasing amounts of the analytes, standard addition calibration curves for each sample are obtained from which the concentrations of unknown samples can be calculated.



1. Create a new method or open your already prepared method. Go to *Method Setup - Calculation* tab.
2. Create *New* calibration file ⓐ or *Set* your already created Calibration file ⓑ .

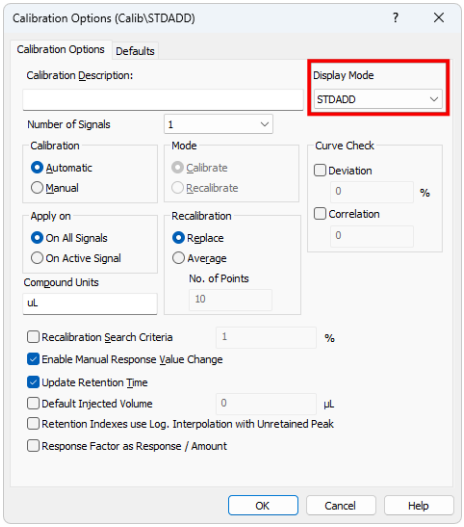


3. Select *STDADD* in the *Calculations* drop-down list ⓒ .
4. Create a custom name for the cloned calibration files in *Calibration Cloning in Sequence* ⓓ and click *OK*.

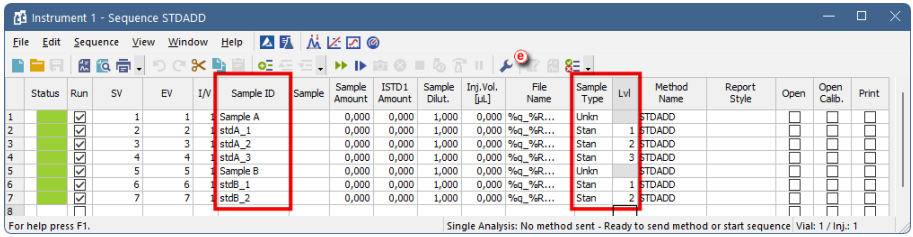
> *Note:* Make sure that each of the measured samples will have a unique calibration name; use predefined parameters to achieve that.

5. Open the *Calibration* window by selecting *Window - Calibration* in the *Instrument* window.

6. Open the *Calibration Options* dialog. Choose *Calibration - Options…* or click on . Select the *STDADD* in the *Display Mode* drop-down list and click ***OK***.

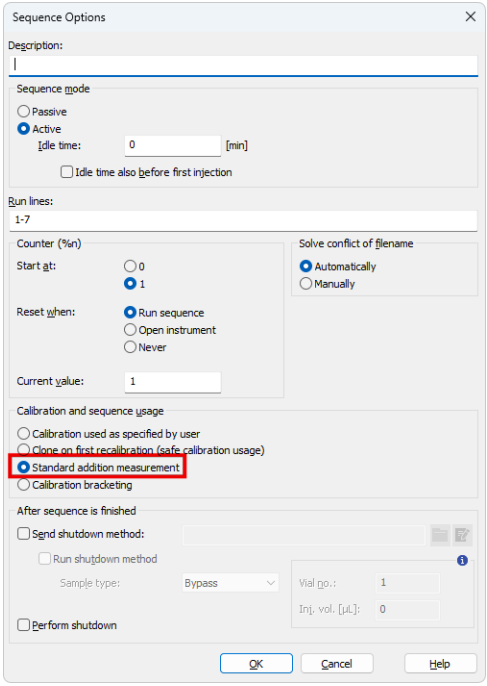


7. Create a new sequence with the given order of the lines:
   - Sample A
   - Sample A with Standard level 1
   - Sample A with Standard level 2
   - Sample A with Standard level X
   - Sample B
   - Sample B with Standard level 1
   - Sample B with Standard level X
   - *continue with the given pattern until desired state*
8. Set the columns as follows: for Unknown samples set *Sample Type* column to *Unknown* and for Standard Addition samples, set *Sample Type* column to *Standard* and *Lvl* column to level corresponding to the added amount in given sample.

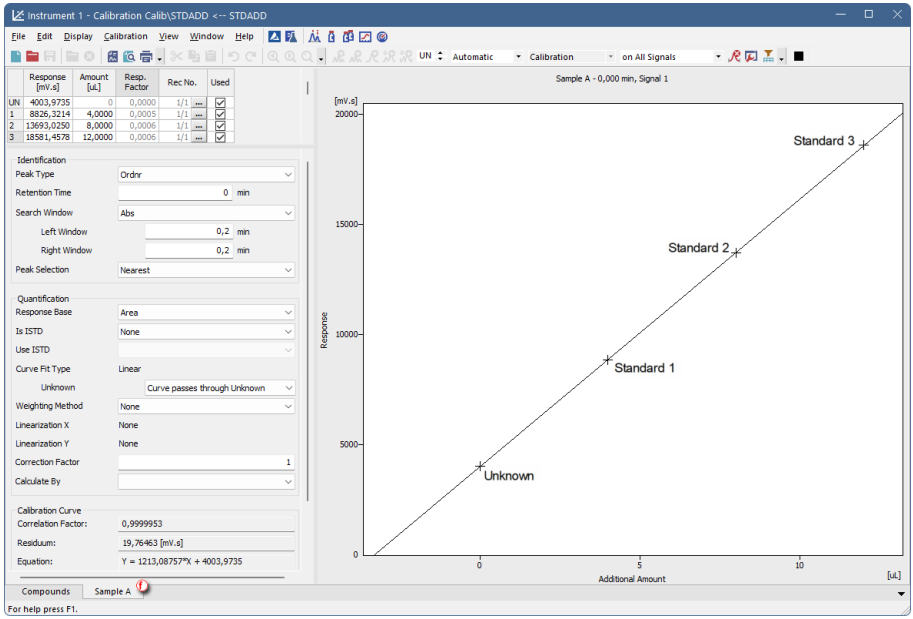


Note: In case you want to use a blank sample too, such sample shall be always put in the sequence before the unknown sample.

9. Click to open the *Sequence Options* dialog ⓔ and select *Standard Addition Measurement* and click *OK*.



10. Run the sequence and wait until the sequence is finished.
11. Open the *Calibration* window: choose *Window - Calibration* in the *Instrument* window or click on .
12. Go to *File - Open* and open the cloned calibration file for the desired sample.
13. Click on the *Compound* tab ⓕ to see the calibration curve. Fill in the amounts of the standard samples.



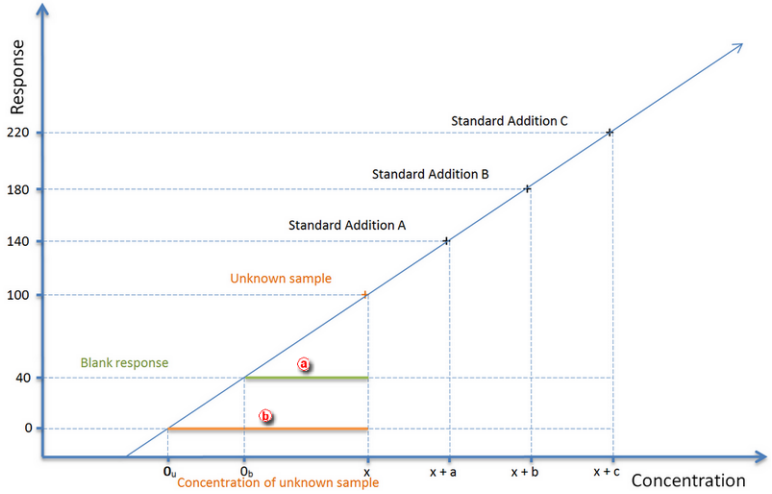
14. Open chromatogram of the desired unknown sample. The Result Table now contains amount of the unknown sample, calculated using Standard Addition.

Note: The measured sequence can be reprocessed using Batch. For more info see the topic Reprocessing whole sequence while using calibration cloning.

9.11.3.1 How the concentration is calculated

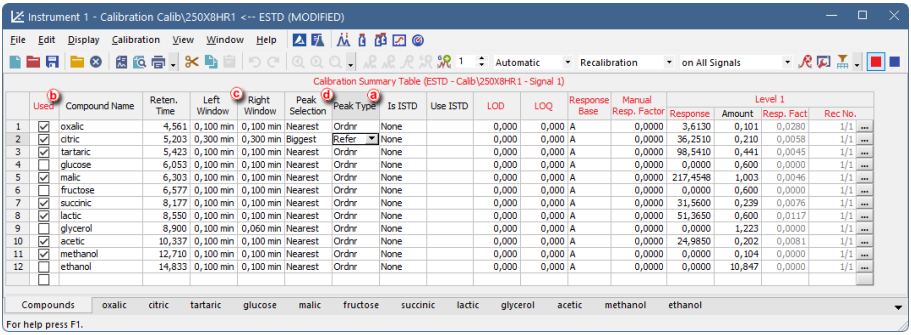
The concentration of an unknown sample is calculated using a calibration curve, which intersects in the point [0,0].

As shown in the picture below, when not using Blank samples the the concentration of an unknown sample equals the orange line (0 response corresponds to 0 unknown sample concentration - 0u), whereas the concentration of the UNKNOWN SAMPLE when using Blank equals the green line (Blank response corresponds to 0 unknown sample concentration - 0b).



9.11.4 Using a reference peak to improve compound identification

A reference peak is a peak used as a reference for recalculating the retention times for the rest of the peaks in a chromatogram. This method allows a better compound identification in those cases where there might be a drift in the retention times in repeated analyses. It is possible to set multiple reference peaks. For ordinary peaks, the expected retention times will be adjusted by linear interpolation between the nearest reference peaks.



1. Open the calibration file: choose *File - Open…* or click on .
2. Select *Refer* on the *Peak Type* column for an easily identifiable compound (i.e. main compound, ISTD standard, any well resolved peak of the matrix) that you will use as a reference ⓐ . To apply the reference peak, *Used* check-box must be checked ⓑ .

3. Edit the Left and Right window values to define the range within which the peak should appear ⓒ. This window may include other peaks if the selected reference peak meets the *Peak Selection* criteria.
4. Select the Peak Selection criteria: biggest, nearest, first or last peak ⓓ.

More Info:

Ordinary peaks are identified by the nearest option by default. For reference peaks the biggest will be set as default. Note that the biggest refers to the selected Response Base, i.e. if the Area is the Response Base, the detected peak may not be the highest one. In specific cases selecting First or Last may be advantageous.

To add the *Peak Selection* column:

- Right click anywhere on the table
- Select *Set Up Columns* to open the relevant window.
- Select *Peak Type* on the right and click on *Show* and then *Ok*.

5. Repeat steps 3 to 5 to add more reference peaks.

Note: In case of multisignal calibration, where compound used as reference does not match the selection criteria on some signal(s), create a variant of the compound only for that signal(s). The name must be different - e.g. Oxalic - UV, Oxalic - RI), copy the rest of the original row, check the used checkbox only for one variant on each signal. Now change the Peak Selection criteria value, or if suitable, change the Peak type to Ordinary, and select other Reference peak on that signal.

6. Save the calibration file: choose *File - Save* or click  .
7. Link the calibration to your chromatogram as explained in *Applying a calibration to a chromatogram*.

9.11.5 Normalized Area % Calculation

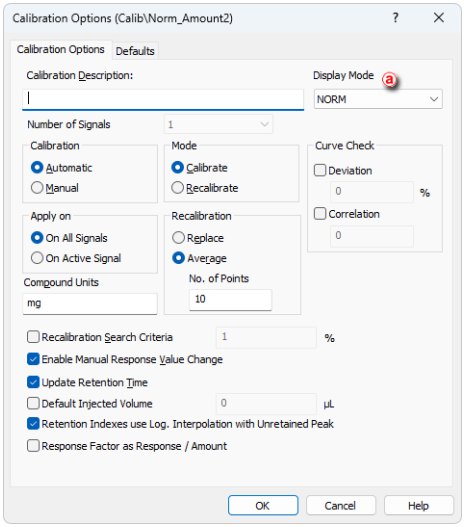
For Normalized Area % calculation no specific settings are necessary, the calculation results are always present in the *Area%* column in the Result table.

9.11.6 Normalized Amount % Calculation

This topic describes how to use Clarity, in order to obtain correct results calculated according to normalization calibration method. The normalization can be achieved by two means. First option is applicable to known response factors specific for each analyte. Second option is to use calibration based on standard sample with known fraction composition. In the latter case, Clarity automatically calculates response factors for all analytes that are subsequently applied in evaluation of the unknown sample. The results are displayed in *Amount%* column in the *Summary Table* and represent percent fraction of each analyte present in the unknown sample.

For both options set the calibration to the NORM Display mode, this ensures that Clarity checks that all the peaks are used for calculations and 100% Amount really corresponds to the whole sample.

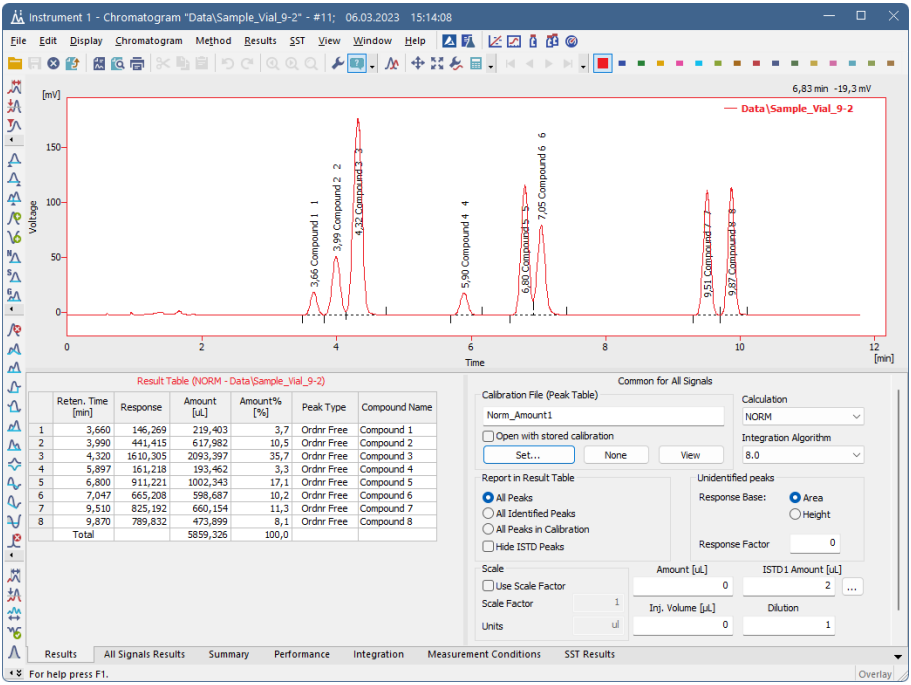
- Set *Display Mode* to *NORM* ⓐ in *Calibration Options* dialog.



A) Application of specific Response Factors

- Use procedure explained in the chapter Calibrating with manually entered Response Factors.

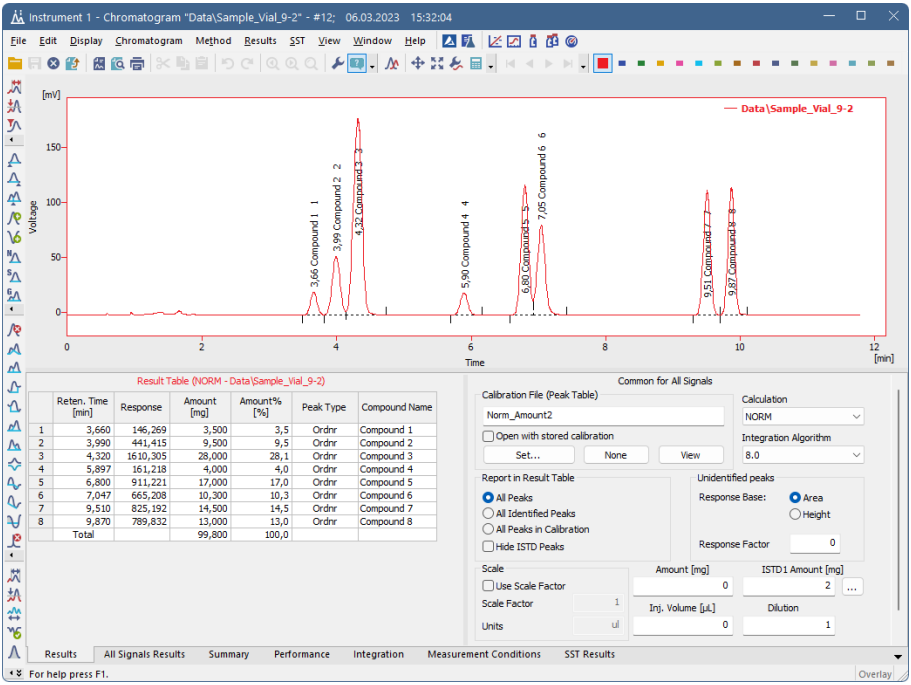
Figure below displays *Result Table* of a chromatogram with linked calibration that was created using the above mentioned method. Note different values in columns *Area [%]* and *Amount%* are caused by various values in *Manual Response Factor* column.



B) Application of automatically calculated Response Factors based on calibration with standard sample with known composition

- Use procedure explained in the chapter Creating a new calibration.

Figure below displays *Result Table* of a chromatogram with linked calibration that was created using the above mentioned method. Note different values in columns *Area [%]* and *Amount%* are caused by calculated values in *Response Factor* column.



9.11.7 Using Calculate By to determine the amount of a compound with no standard available

When no standard of a compound is available and a compound with known response ratio to the unavailable one is present within the sample (this information can be found in respective norm etc.) the *Calculate By* and *Correction factor* can be used to determine its amount in the sample.

1. Open the calibration file.
2. The *Calculate By* and *Correction Factor* are hidden by default. To display them right-click the table, select *Setup Columns...*and in the following dialog move the respective items from *Hide Columns* list to *Show Columns* list.
3. For compound (Chloroform in the example) ⓐ that should be evaulated by another one, select its *Calculate By* field ⓑ and pick which compound to use for the calculation (Trichloroethane in the emxaple).

Note: Only compounds (peaks) already added to the calibration can be selected.

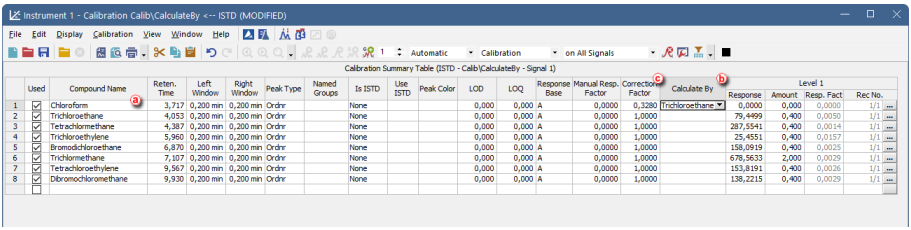
4. Fill the known response ratio to the *Correction Factor* column ⓒ .

Note: The *Amount* filled in the row that will be calculated using calibration curve of another compound will be ignored.

5. The resulting amount in *Chromatogram Results Table* will be calculated according to the calibration curve of compound selected to *Calculate By*, and multiplied by the *Correction Factor*.

Note: A compound marked as *ISTD* should neither be used as a compound that is used to calculate another compound nor as a compound that is calculated using another compound.

In the example Chloroform is calculated based on the calibration curve of Trichlorethane, their response ratio is 0.328.



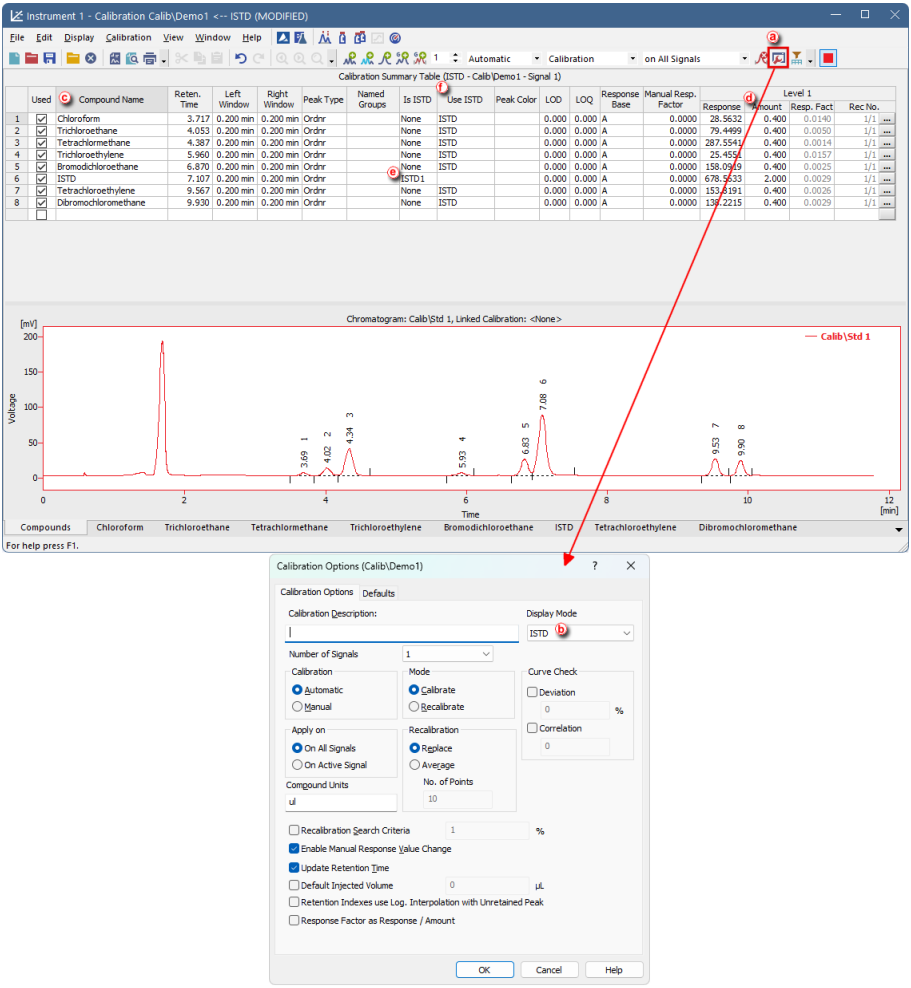
9.11.8 Calibration with ISTD

The use of internal standard (ISTD) helps to compensate for non-reproducible injected volume or for analyte losses during sample preparation. A known amount of internal standard compound (which should have similar properties as the analyte, but should not be present in the samples) is added to the standards and samples. The determined analyte amounts are then corrected based on the ratio of the added and detected amounts of the ISTD compound(s).

This chapter provides detailed insights into the differences when creating a calibration with ISTD compared to a standard calibration, as described in Creating a new calibration.

1. In the *Calibration Options* window (accessible via the icon ⓐ in the *Calibration* window), change the *Display Mode* to *ISTD* ⓑ .
2. Open your first calibration standard and add peaks to the Calibration Summary Table as usual.
3. Label the peaks by the compound names in the corresponding column ⓒ and fill in the *Amount* column ⓓ with the amounts of the present compounds including ISTD.
4. In the row corresponding to ISTD, change the type of the cell in the *Is ISTD* column to *ISTD1* ⓔ . The *Use ISTD* column ⓕ now contains the name of the selected ISTD compound in all the other rows. In compounds tabs, the axes are labeled according to the selected ISTD.

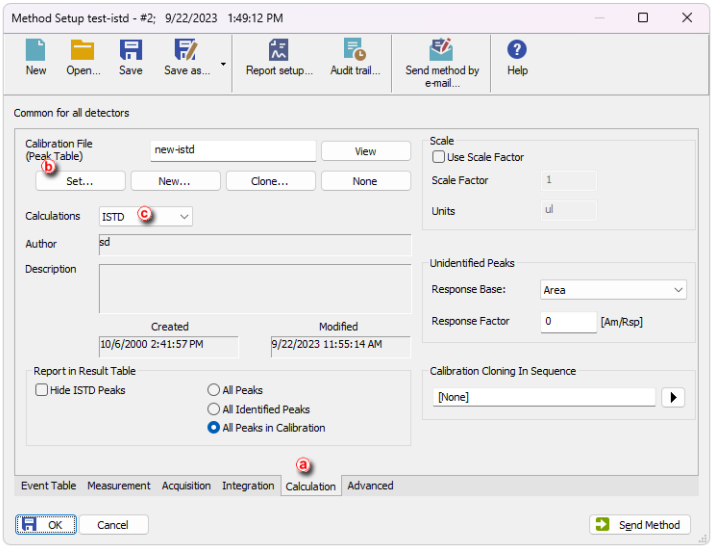
Note: You can set up to 10 compounds as ISTDs in the *Is ISTD* column. When more ISTDs are present, you can specify which one to use for quantification of which compound in the *Use ISTD* column.



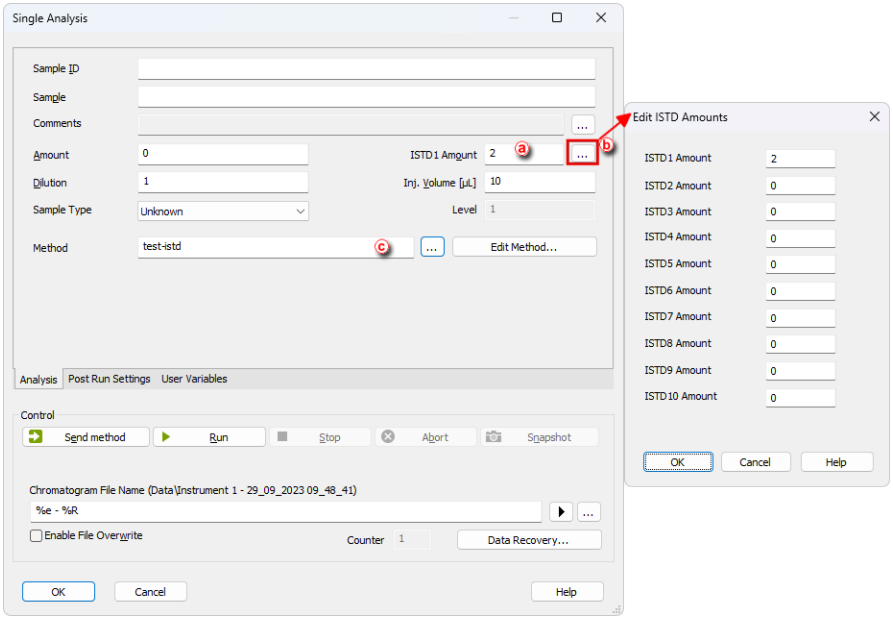
5. Now, the first calibration level is set. Continue as usual to set the remaining calibration levels using the measured calibration standards (as shown in Adding a new calibration level). Enter the ISTD amounts according to the mode you plan to work in.

The created ISTD calibration may now be linked to an already measured chromatogram for direct results assessment (see Applying calibration to a chromatogram), and it may be also linked to a method. This causes that the chromatograms measured using this method will be automatically evaluated using the pre-selected ISTD calibration.

6. To link the ISTD calibration to a method, in *Method Setup*, open the *Calculation* tab ⓐ and set the created ISTD calibration file ⓑ. The *ISTD* parameter ⓒ should be selected in the *Calculations* option. Press the *OK* button to save the Method and close the dialog.



7. To use the ISTD method in single analysis, set the *ISTD1 Amount* ⓐ in the *Single Analysis* window to the desired value. In the case of more than one ISTD compound present, set the amounts in the dialog which opens by clicking the triple dot ... menu ⓑ.
8. Select the previously created method ⓒ. You can *Run Acquisition* directly from the *Single Analysis* window.



The sequence analysis setup is analogous to the single analysis. The ISTD2–ISTD10 amount columns are not shown by default and can be added via the *Edit - Setup columns...* dialog.

Note: In the case the amount of internal standard is the same for all standards and samples, it is possible to enter zero instead of the amount (which may not be known). In such case, the correction will be based on the ratio of the ISTD peak response in the standards and unknowns. Note that the ISTD amounts must be entered or set to zero both in Calibration and in the Sample Header. A mismatch will be detected and reported as error.

9 Batch Processing

This chapter focuses on processing more chromatograms using *Batch* dialog. There are 3 main ways of workflow Reprocessing whole sequence, Reprocessing by Method and Post Run options, all will be explained in more detail in this chapter.

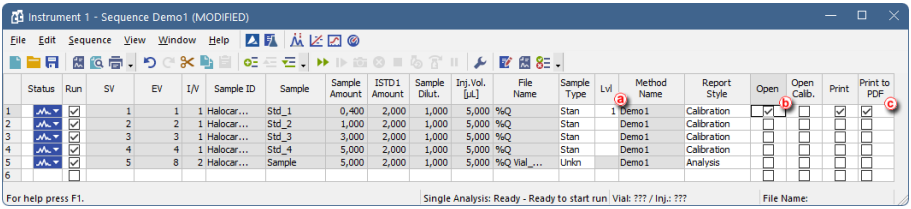
Clarity Tips&Tricks videos covering Batch topics can be found in Clarity Batch playlist.

9.12 Reprocessing whole sequence

Reprocessing whole sequence can be convenient if performing reintegration, recalibration, or changing types of samples is needed. It can also be used with calibration cloning, more in the topic Complete processing with calibration cloning. Steps below described process where sequeunce is already measured by template method with optimized integration parameters, for more see Save Method as a Template.

1. Measure the sequence with no post run options set and/or no *Levels* for standards filled in (Typically, you want to perform the recalibration/export/print of the results after reviewing the chromatograms).
2. Review the chromatograms: check the integration, etc.
3. Go to the measured sequence, set *Levels* ⓐ for calibration standards and enable Post Run options you wish to perform, e.g., *Print To PDF* ⓒ . It is also possible to uncheck (check) the Run checkbox at rows you don't want to reprocess.

Note: To print or export the data the chromatogram has to be opened in the *Chromatogram* window - this is achieved by checkbox in the *Open* ⓑ column on a given row.

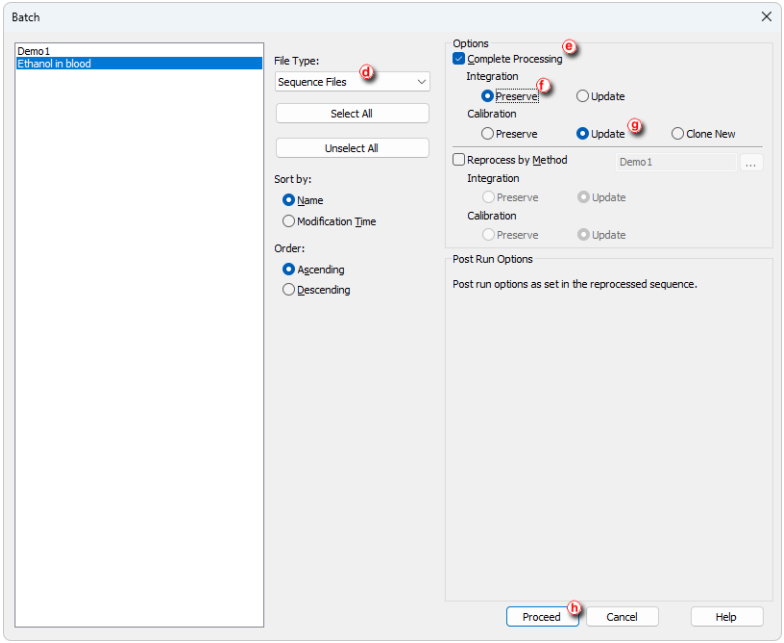


4. Open the *Batch* dialog by using *Analysis - Batch* in the *Instrument* window.
5. Switch *File Type* to *Sequence Files* ⓓ .
6. Select the Sequence you wish to reprocess.
7. Check the *Complete Processing* ⓔ checkbox.
8. To preserve manually updated integration (this mainly considers small individual adjustments where optimized parameters weren't satisfactory) select the *Preserve* ⓕ in the *Integration*part of *Options* section.
9. To perform recalibration based on updated integration select *Update* ⓖ in

the *Calibration* part of *Options* section.

10. *Proceed* ⓗ with the batch processing.

The recalibration will be performed and Post Run options defined in the sequence will be performed.



Caution: The operations during Batch reprocessing are done row after row, injection after injection. In some situations it is necessary to perform the Complete Processing in two steps - first just the recalibrations, second the post-run actions (either using the Complete Processing again with the Integration a Calibration settings in the Options section to Preserve, or through running the post-process on selected chromatograms only through procedure described in Performing Post Run Options from Batch dialog section). For example, if the sequence is using calibration bracketing the unknowns are measured before second standards set and if the unknowns were reported during the recalibration step, the responses from the second standards set would be missing in the report.

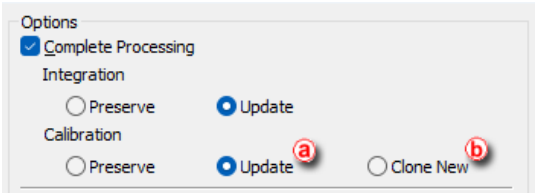
9.13 Reprocessing whole sequence while using calibration cloning

If you are using any kind of the calibration cloning in the sequence the reprocessing whole sequence is similar to regular reprocessing described in the previous topic.

Please refer to the Calibrating using clone on first recalibration, Compensating for response drift using bracketing or Improving quantification with the standard addition method topics to learn more about Calibration Cloning in Clarity.

The difference from the standard Reprocessing procedure described in Reprocessing whole sequence topic is shown below.

- In the *Batch* dialog you have additional option for calibration behavior, the meanings of options are:
  - If you select Update ⓐ in Calibration part of Options section the calibration(s) that are currently linked to measured chromatogram(s) will be recalibrated (new cloning is NOT performed).
  - If you select Clone New ⓑ in Calibration part of Options section new calibration(s) will be created as if the sequence was run again (new clone (s) are created and linked to chromatograms).



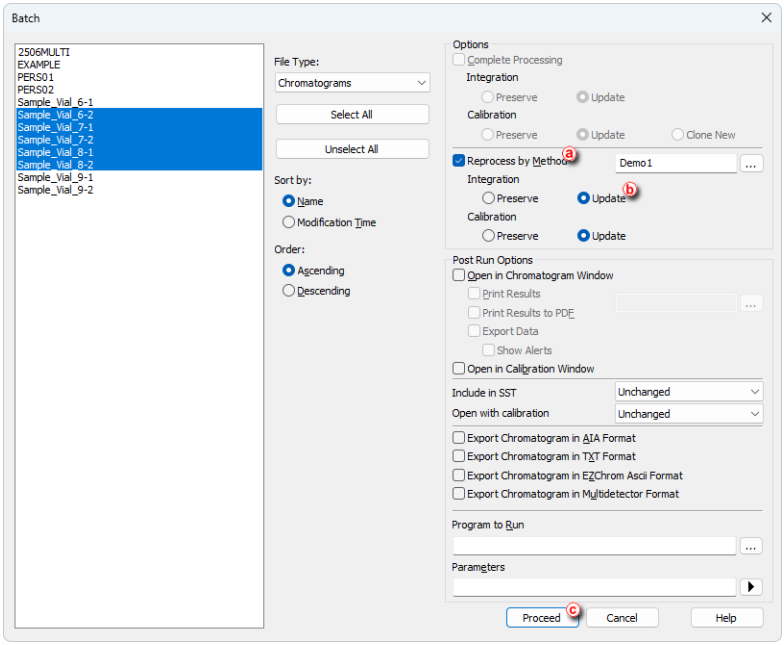
9.14 Reprocessing by selected method

Reprocessing by method is suitable for applying Integration parameters to previously measured chromatograms after these parameters were saved to the method by Save Method as a Template. You can also changed the link to the calibration file without the calibration being reprocessed.

Note: If the Sequence file is selected to be reprocessed by method, all currently linked chromatograms will be reprocessed. No recalibration is carried out, when *Calibration - Update* is selected only links to calibration are changed without any changes to the calibration file itself.

1. Prepare your method with desired integration and linked calibration.
2. Open *Batch* dialog by menu *Analysis - Batch...* from the Instrument window.
3. Select chromatograms you want to reprocess (Alternatively, select sequence(s), depending on your *File Type*).
4. Select *Reprocess by Method* ⓐ and define your created method.
5. Select *Update* ⓑ Integration in case you want to change integration parameters as set in the defined method.
6. Click on Proceed ⓒ .

Note: *Reprocess by Method* can be combined with *Post Run Options* if you are sure that changes in chromatograms do not have to be reviewed before exporting/printing.



Now your chromatograms are reprocessed, with linked calibration file and amended integration parameters from the used method.

9.15 Performing Post Run Options from Batch dialog

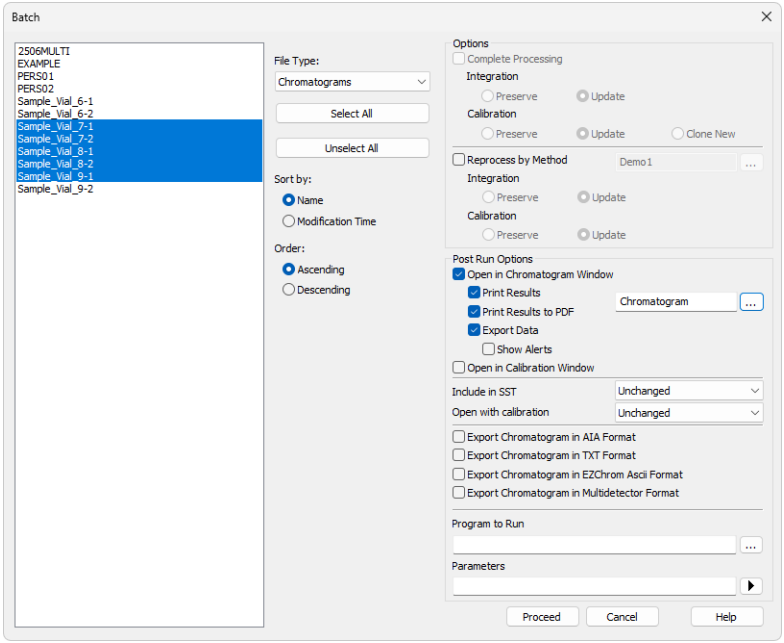
This procedure is useful if you need to *Print* or *Export* more chromatograms at once, e.g. after all of them were reviewed and signed.

1. Navigate to *Batch* dialog by clicking *Analysis - Batch* in *Instrument* window.
2. Select chromatogram(s)/sequence(s) that you want to print reports for (when sequence file is selected, all chromatograms that are currently linked in it will be reported). Which files are displayed is based on *File Type* drop-down menu.
3. Select *Open in Chromatogram Window* to be able to select *Print Results* and/or *Print Results to PDF* and/or the *Export Data* (or select any other option).
4. Select report style you would like to use by clicking ... button.
5. (Optional) change *Open with Calibration* to *Stored*, this will ensure that data will be printed in the state they were saved in last time.

   Note: By default chromatograms are opened with *Linked* calibration. If the calibration was changed after saving chromatograms the results might be influenced.

6. Click *Proceed*.

Note: It is also possible to process data before printing them by using *Complete Processing* or *Reprocess by Method* options. When *Complete Processing* is selected Post Run options are governed by settings in reprocessed sequence. For more info see chapter regarding Batch dialog for more info.



10 Results and Calculations

Advanced topics describing how to use *User Columns* to calculate custom results, etc.

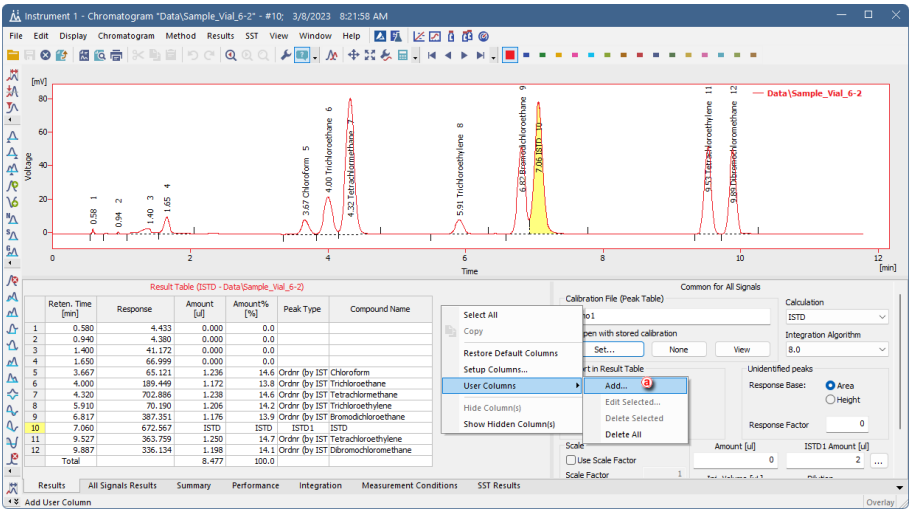
10.1 User Columns

This article describes how to append a new column with user-defined calculation to the *Chromatogram - Result Table*. For more details about User Columns refer to **Reference Guide Manual**.

Note that majority of calculations require chromatogram to be calibrated. Without it the necessary values are not available.

Caution: *User Columns* are stored in .DSK files. If the station is used by more users and *User Columns* are needed it is recommended to set shared .DSK file as described in chapter "Sharing settings among users".

1. Open a chromatogram you want to work with and switch to *Results* tab.
2. Right click on the *Result Table* and select *User Columns - Add...* ⓐ , the *Add User Column dialog* will be opened.

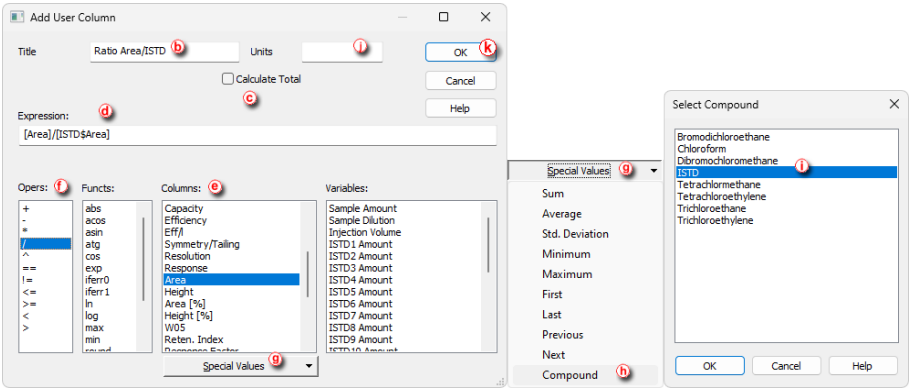


3. Fill in the *Title* ⓑ of the new column. In our case *Ratio Area/ISTD*.
4. Check/Uncheck *Calculate Total* ⓒ (depends on whether sum of calculated values has a meaning).
5. Fill in *Expression* ⓓ line , which presents the user's defined calculation. In our case:

- In *Columns:* ⓔ list double-click *Area*.
- In *Opers:* ⓕ list double-click */*.
- In *Columns:*list select *Area*, then click on *Special Values* ⓖ , select *Compound* ⓗ , choose *ISTD* ⓘ and click *OK*.

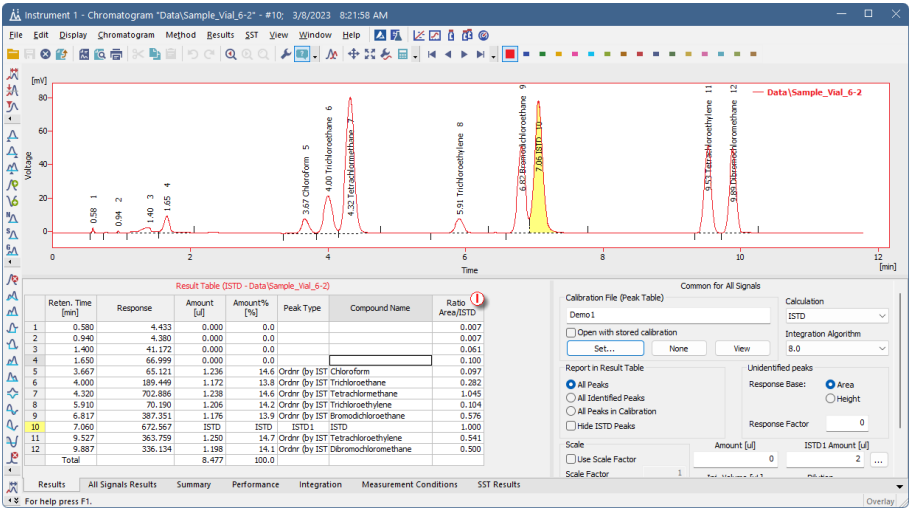
6. Fill in appropriate *Units* ⓙ based on the formula.
7. Click *OK* ⓚ in the *Add User Column* dialog.

Note: It is possible to fill *Expression* by typing if you know the correct syntax.



8. New *User Column* is added to the Result Table ⓛ .

Note: You can edit existing *User Columns* by right-clicking into them and selecting *User Columns - Edit Selected...*.

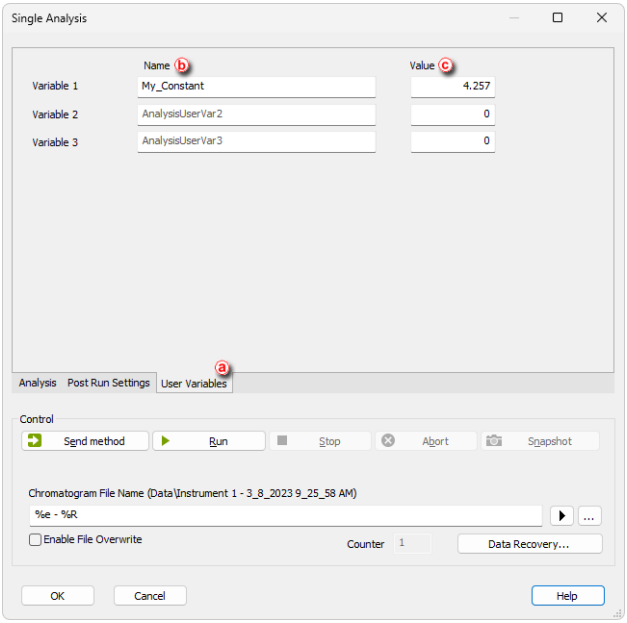


10.2 User Variables

This article describes how to set *Analysis* and *Method User Variables* which can be subsequently used in *User Columns* calculations. *Analysis User Variables* can be defined in *Single Analysis* or *Sequence* window. *Method User Variables* can be defined in *Method Setup* window.

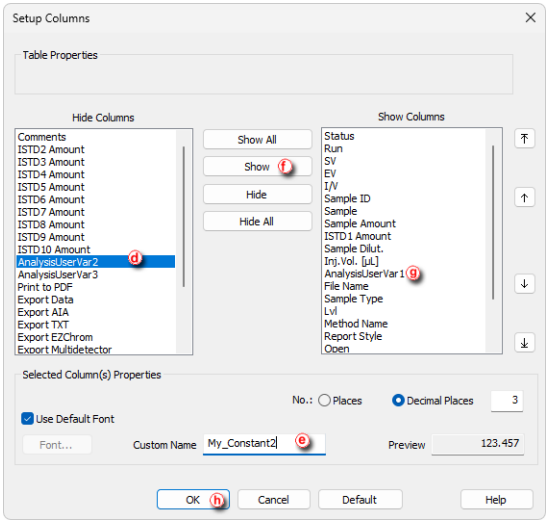
A) Setting of Analysis User Variables in Single Analysis window

1. In *Single Analysis* dialog navigate to the *User Variables* tab ⓐ .
2. Define *Name* of the variable ⓑ . If the field is left empty, default name *AnalysisUserVar1-AnalysisUserVar3* will be used.
3. Define numerical *Value* of the variable ⓒ .

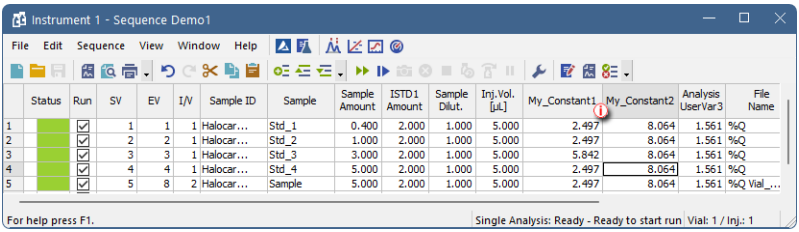


B) Setting of Analysis User Variables in Sequence window

1. In *Sequence* window the *AnalysisUserVar1 - AnalysisUserVar3* columns are hidden by default.
2. To display them right-click on the *Seqeunce Table* and use *Setup Columns...* command.
3. Select the variable in the *Hide Columns list* ⓓ , fill its name ⓔ and click *Show* ⓕ (item will be move to the *Show Columns* list ⓖ).
4. Repeat the previous step if more variables are needed then click *OK* ⓗ .

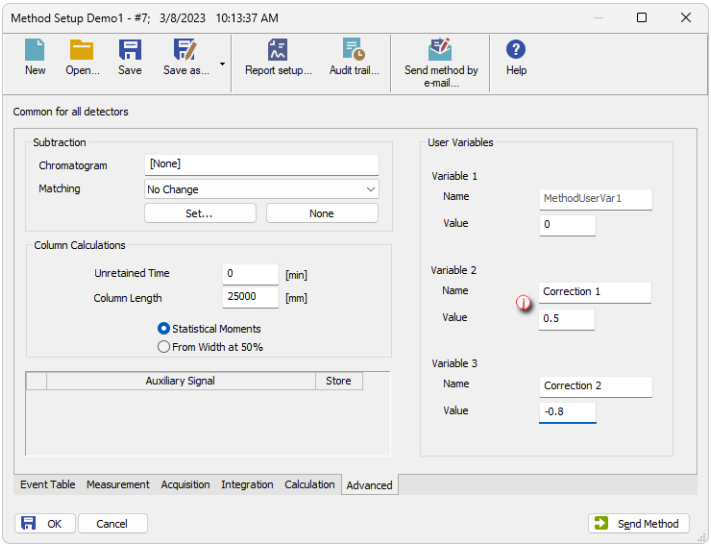


5. Defined *Custom Name* of the variable is used directly in the column header ⓘ . Fill in the values for each row (different values can be used).



C) Setting of Method User Variables in Method Setup window

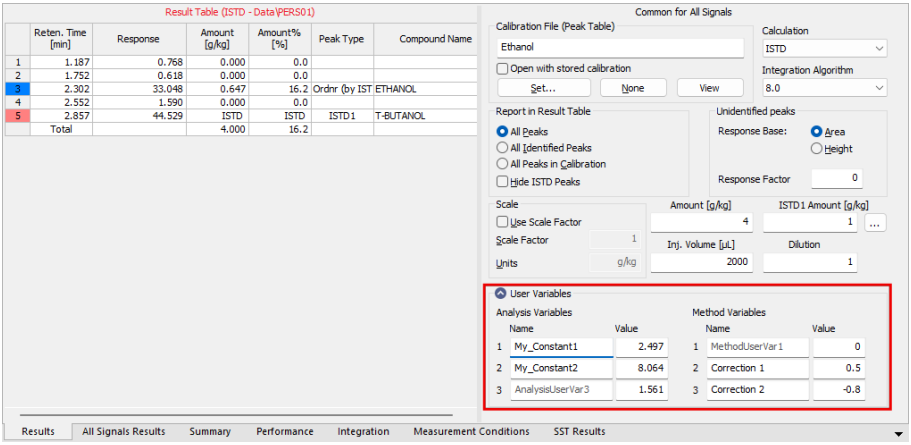
1. In the *Method Setup* dialog navigate to *Advanced* tab
2. Define *Name*, numerical and *Value* of the variables that should be used ⓘ . Default name *MethodUserVar1-MethodUserVar3* is used when the *Name* is empty.



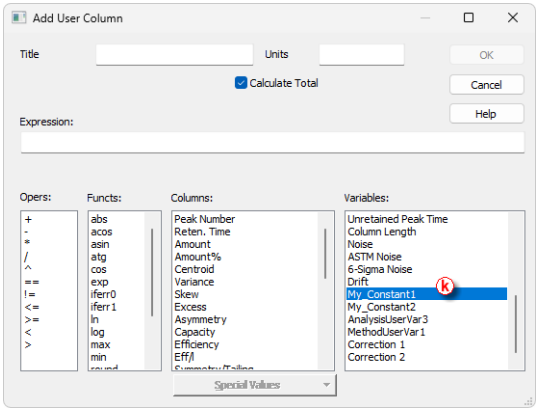
D) Setting of User Columns with User Variables

Analysis and *Method User Variables* are also editable directly from *Chromatogram* window on *Results* tab in *User Variables* section (it is collapsed by default and must be expanded by clicking the arrow symbol).

Variables can be modified in the same manner as in the previous cases.



Note: To use variables in custom calculations add *User Column* as described in chapter "User Columns" and while formulating the *Expression* select the desired variable from *Variables* list ⓚ .



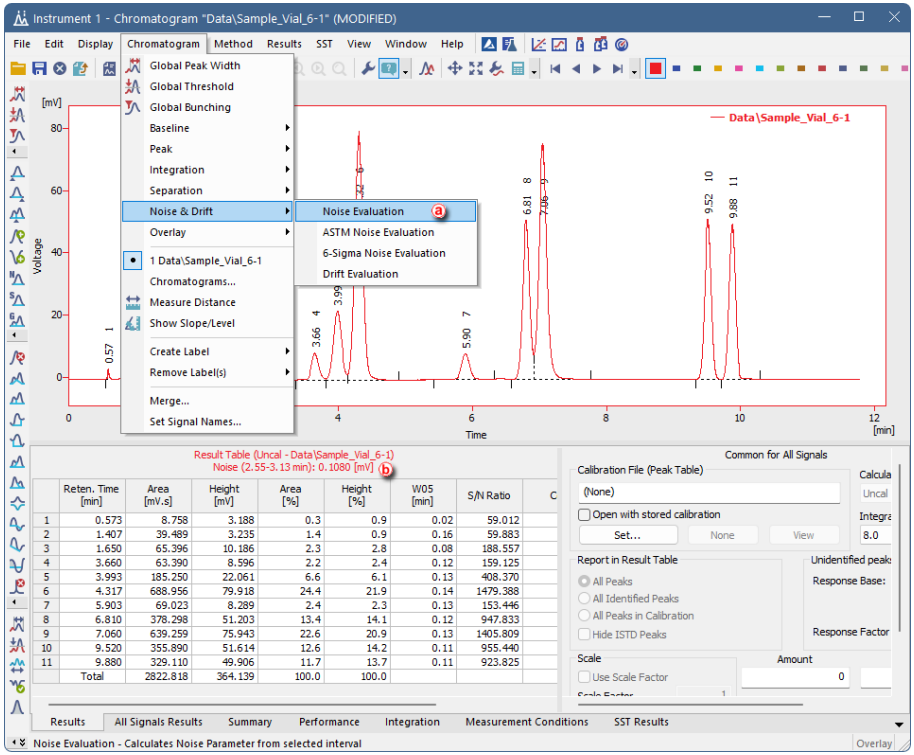
10.3 Signal to Noise Ratio Calculation

This article describes how to calculate Signal to Noise Ratio using *User Columns* calculations with *Variables*.

10.3.1 Noise Parameter Evaluation

At first *Noise* value has to be evaluated. This value is subsequently used to calculate the *Signal to Noise Ratio*.

1. Open measured chromatogram in the *Chromatogram* window
2. Use *Chromatogram - Noise & Drift - Noise Evaluation* ⓐ and select the interval for noise calculation. The same result can be obtained from the *Integration Table* after selecting the *Evaluation - Noise* operation and inputting the desired interval manually.
3. The value of the evaluated Noise Parameter is shown in the *Result Table* header ⓑ .
4. For the same chromatogram such value is stored as a *Noise* variable in the *Edit User Columns - Variables* ⓔ .



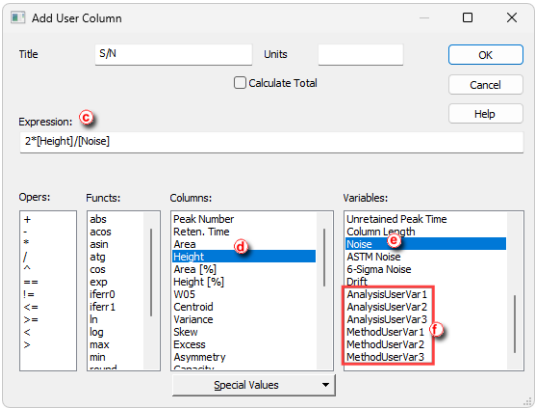
10.3.2 Calculating Signal to Noise Ratio

Signal to Noise Ratio is important parameter used for method validation. Most commonly used generic formula for Signal to Noise = 2*Peak Height/Noise.

For such calculation, two possible approaches exist:

- The *Noise* is determined from the same chromatogram within area with no peaks. The *Noise* variable ⓔ can be used directly in the formula entered in the *Expression* ⓒ edit box. In that case the resulting formula will be for example 2*[Height]/ [Noise], where both parameters are determined from the same chromatogram.
- The *Noise* is determined from other chromatograms (e.g., by measuring blanks and evaluating the *Noise* in the area of expected peak). In the *Edit User Column* dialog in the *Expression* ⓒ edit box use the [Height] ⓓ from the Columns section for peak height and enter the calculated *Noise* value as a constant. The resulting formula will be then e.g. 2*[Height]/0.1080, where the number 0.1080 is the calculated *Noise* parameter. Alternatively, the calculated *Noise* parameter could be stored as a *Method User Variable* or as an *Analysis User Variable* ⓕ to

simplify changes of the value - this variable can be then used instead of entering the numerical value in the *Expression* edit box.



Note: In the *Expression* edit box, variables are treated as numbers, therefore operators and numbers can be used to modify the formula. The resulting formula can be e. g. 2* [Height]/ [Noise] or 2* [Height]/ [UserVariableNoise] (where the [UserVariableNoise] represents user variable to which the Noise value determined from chromatogram blank is stored) or 2*[Height]/0.1080 (where the 0.1080 represents the actual Noise value determined from chromatogram blank).

Note: Setting the *Analysis User Variable* or the *Method User Variable* is described in the section **User Variables on pg. 141**.

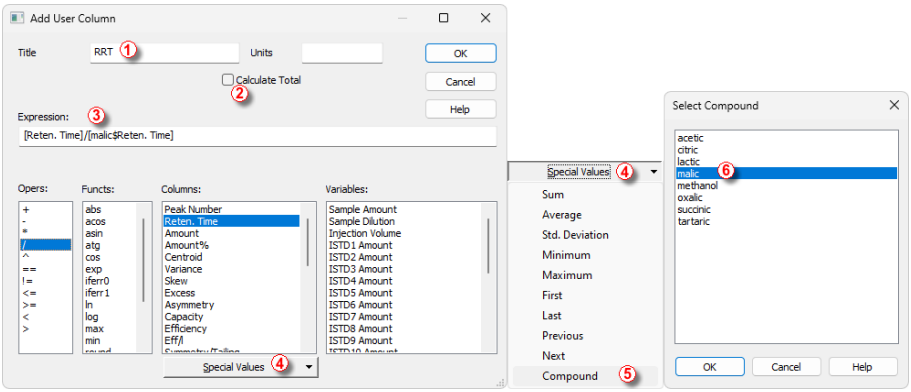
10.4 How to calculate Relative Retention Time or Retention Time Ratio

This article describes how to calculate Relative Retention Time which is a comparison of Retention Time of one compound to another.

1. Open a chromatogram you want to work with and switch to *Results* tab.
2. Add a *User Column* as described in chapter "User Columns".
3. Fill the *Title* for the new column - e.g. *RRT* ① .
4. Uncheck *Calculate Total* ② .
5. Enter "*[Reten. Time]/[malic$Reten. Time]*" formula to the *Expression* field ③ , where "malic" represents your compound which should be used for calculation of relative retention time. You can either use items from *Columns:* list or type it manually.

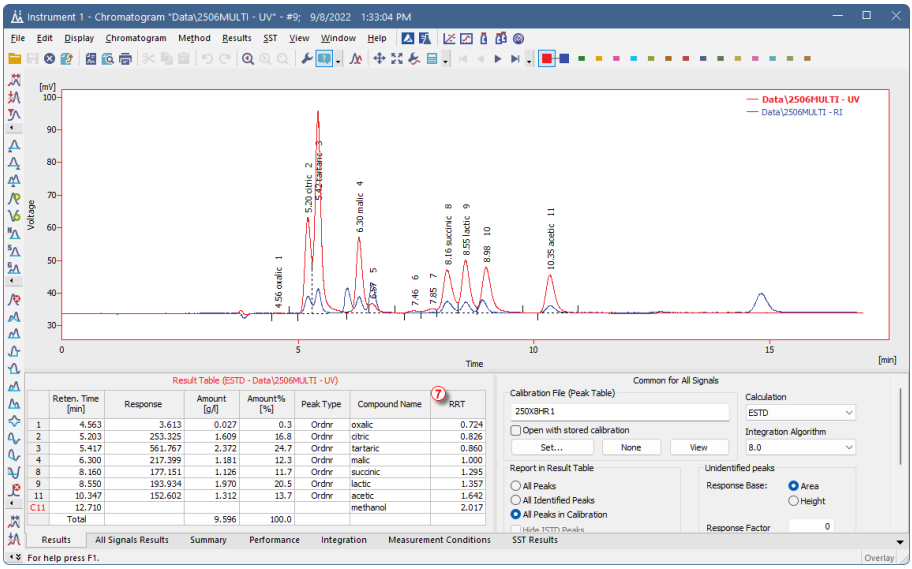
Note: To insert *[malic$Reten. Time]* select *Reten. Time* in *Columns:* list, click *Special Values* ④ , in the drop-down list select *Compound* ⑤ and in the following dialog select compound ⑥ you want to use for calculation of RRT (the list is based on calibration file).

6. Close the *Add User Column* by clicking *OK*



7. New *RRT* column ⑦ is displayed in the *Result Table*.

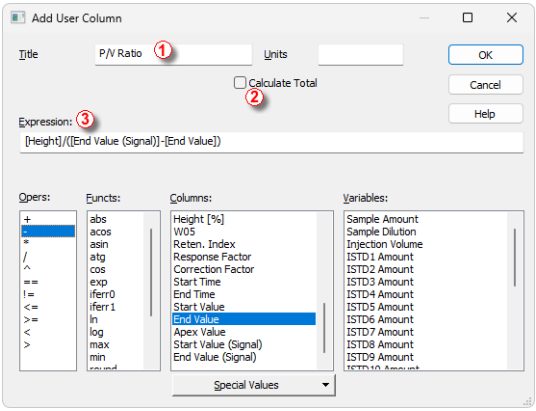
Note: Quick check that it is calculated correctly is to look at the *malic* compound. There should be value of *1*.



10.5 How to calculate Peak to Valley Ratio

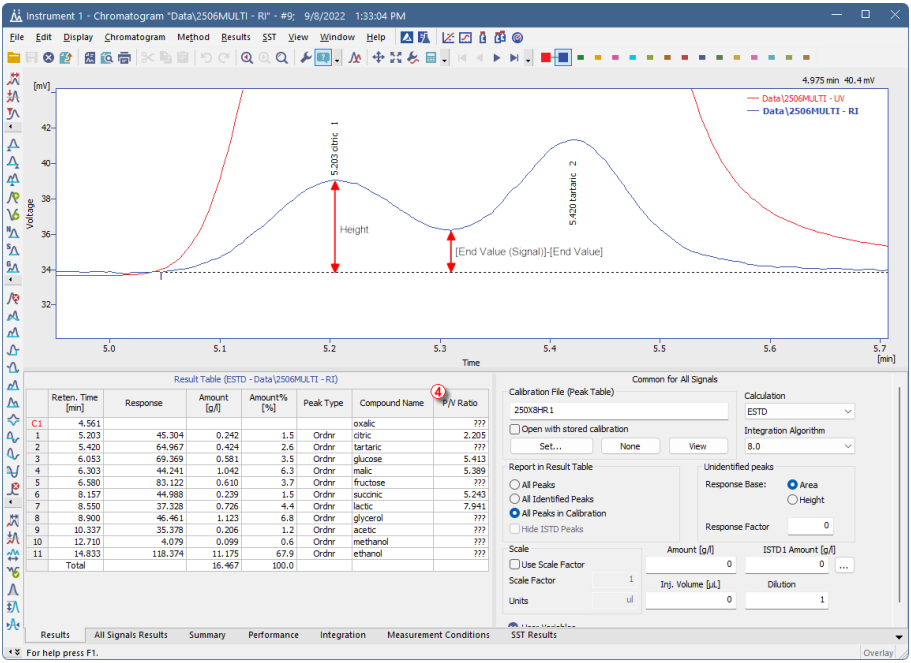
This chapter describes how to calculate Peak to Valley, which is a ratio of signal height in peak apex and signal height in the end of peak. This ratio is used for e.g., impurity determination.

1. Open a chromatogram you want to work with and switch to *Results* tab.
2. Add a *User Column* as described in chapter "User Columns".
3. Fill the *Title* for the new column - e.g. *P/V Ratio* ① .
4. Uncheck *Calculate Total* ② .
5. Enter "*[Height]/([End Value (Signal)]-[End Value])*" formula to the *Expression* field ③ . You can either use items from *Columns:* list or type it manually.
6. Close the dialog by clicking *OK*.



7. New *P/V Ratio* column ④ is displayed in the *Result Table*.

Note: Value is only valid for peaks, where peak end is not on the baseline.



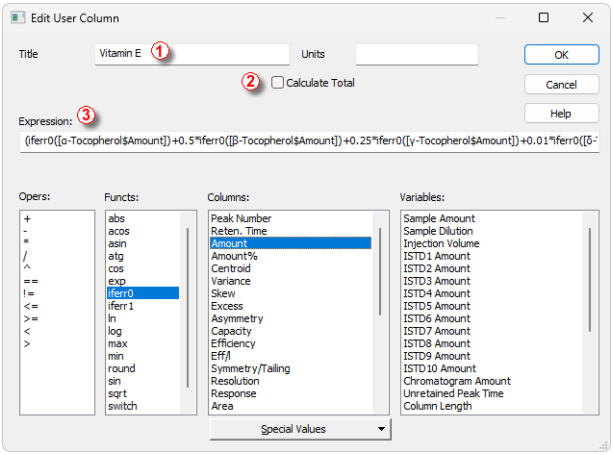
10.6 How to handle not-found compounds in your calculations

If any compound is missing from a chromatogram, its amount is not calculated. In any equations within *User Columns*, where this amount would be used, the calculations would produce ??? (invalid result). In such cases, the *iferr0* function can replace the missing value with 0 (analogically, the *iferr1* function replaces the missing value with 1). This functionality can be used in situations, when a substance is present in multiple chromatogram peaks, possibly even in different ratios. *User Columns* can be used to determine its total content. The *Expression* (equation) would then contain the Amounts of compounds in the chromatogram. However, some of the compounds may not be present in the evaluated chromatogram. The following case scenario shows how the *iferr0* function can be used. The goal is to calculate the content of Vitamin E in different Tocopherols.

1. Open a chromatogram you want to work with and switch to *Results* tab.
2. Add a *User Column* as described in chapter "User Columns".
3. Fill the *Title* for the new column - e.g. *Vitamin E* ① .
4. Uncheck *Calculate Total* ② .
5. Enter the formula for calculating the vitamin E content, e.g., "*([α-Tocopherol$Amount]+0.5* [β-Tocopherol$Amount]+0.25* [γ-Tocopherol$Amount]+0.01*[δ-Tocopherol$Amount])/10*" to the *Expression* field.

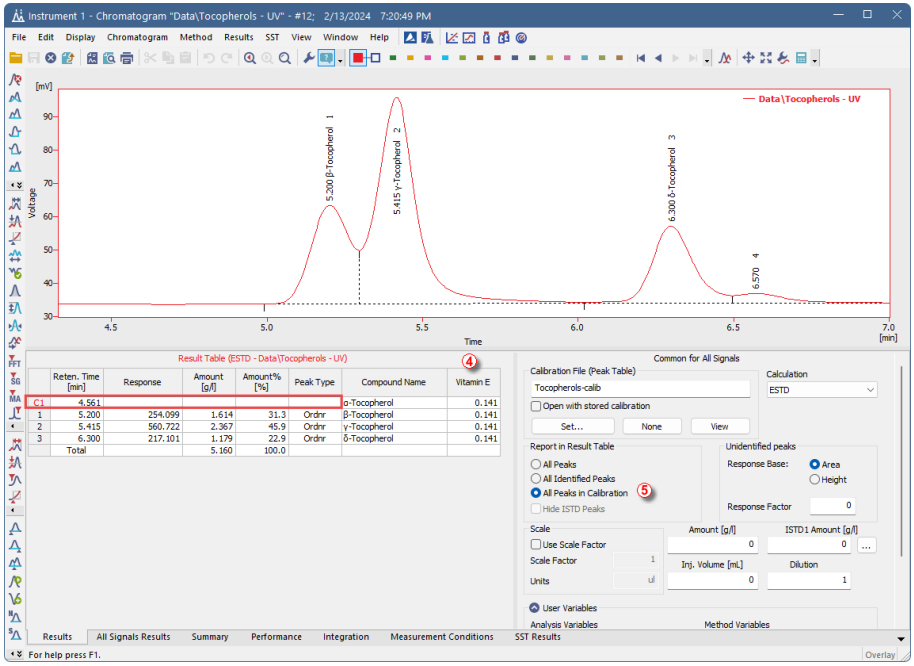
You can either use items from the *Columns:* list with *Special Values* or type it manually.

6. Apply the *iferr0* function on the items that may be missing from the result table. The resulting function will look like the following: *(iferr0 ([α-Tocopherol$Amount])+0.5*iferr0 ([β- Tocopherol$Amount])+0.25*iferr0 ([γ-Tocopherol$Amount])+0.01*iferr0([δ-Tocopherol$Amount]))/10* ③ .
7. Close the dialog by clicking *OK*.



8. The new *Vitamin E* column ④ is displayed in the *Result Table*.
9. In the case that any of the compounds is not found in the chromatogram (such as here, in the case of α-Tocopherol), the Amount is not calculated. The *iferr0* function replaces the not-found Amount with zero in further calculations.

Note that the *All Peaks in Calibration* ⑤ option is selected. Otherwise, the missing compound would not be present in the *Result Table*, and the calculation would not be possible.



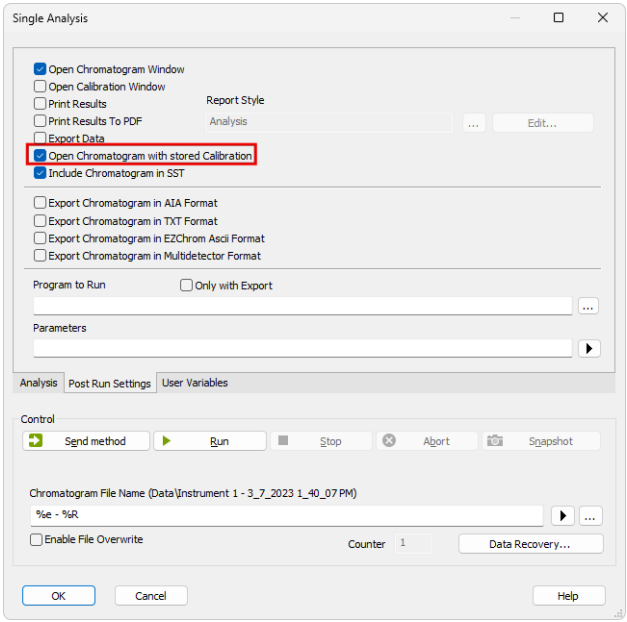
10.7 How to Display Older Results when Linked Calibration is Modified

Clarity does not store any results in the chromatogram, the result table is always calculated from the actual state of the calibration file referenced in the *Calibration Table (Peak Table)*, i.e. *Linked* calibration. Any changes in this calibration will be immediately reflected in the displayed results. Each time a chromatogram is saved the current state of the linked calibration is stored (i.e. *Stored* calibration) in the chromatogram history (just values needed to calculate the results, not a complete calibration). Chromatogram opened with a version from history (i.e. opened with *Stored* calibration) will display results as they were at that time.

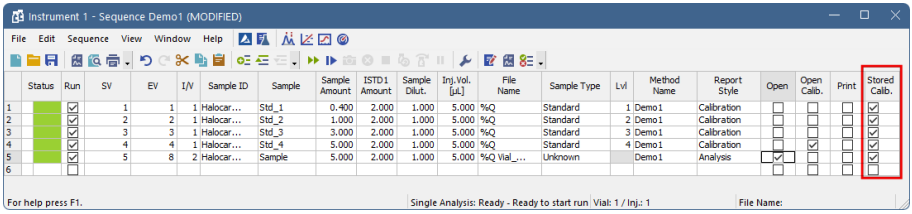
In case the calibration file is reused for some time, opening the chromatogram with linked calibration will show changed results due to changes in the linked calibration. To avoid this, two approaches are possible:

1. Make a copy of the calibration file so each series of measured chromatograms will be linked to a separate calibration file. This has advantage in case some amendments to the calibration will be necessary later, as the amendments will affect only the related chromatograms. Such procedure could be automated from sequence. For more information regarding this procedure, see the chapter **"Calibrating using clone on first recalibration"** on pg. **95**..

2. Using the *Open with stored calibration* option. Such option is accessible from multiple dialogs, depending on whether Single Analysis or Sequence is measured (automated approach) or this option can be selected upon opening the chromatogram.
    - In the *Single Analysis* dialog - *Post Run Settings* tab select *Open with stored calibration* option.



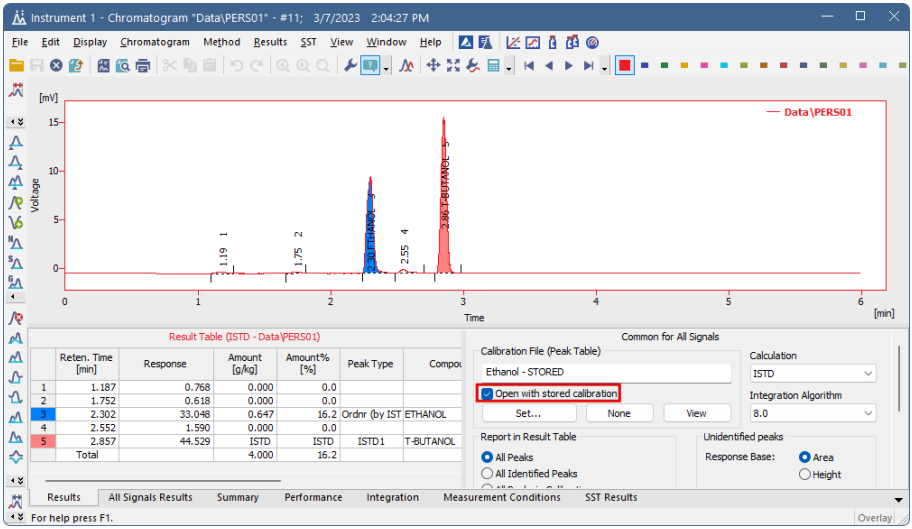
    - In the *Sequence* window select checkbox in the *Stored Calib.* column. By default, *Stored Calib.* column is hidden. To show it, right mouse click in the sequence table and choose *Setup Columns...*. From the *Setup Columns* dialog, choose *Stored Calib.* from the left list and click on the *Show* button - it will be added to the show list. Once you click the *OK* button, you will return to the *Sequence* window and the new *Stored Calib.* column will be added.



    - In the *Chromatogram* window select *Open with stored calibration* option. This will open the chromatogram with the most recent point

from history and show the results according to the stored calibration. Any changes in the linked calibration will not affect those results. To open the chromatogram with the stored calibration this way it is necessary to re- open the chromatogram using the *Open Chromatogram* dialog - re-opening the *Chromatogram* window is not sufficient.

Note: This setting is only applied and saved to the currently opened chromatogram. It will not be transferred to the next (different) opened chromatogram.



10.8 Calculating percentage content of a compound in a solid sample

This is a standard procedure used across various chromatography applications - a known amount of a sample is dissolved in a known volume of a solvent and the goal is to determine the percentage content of the compound in the sample.

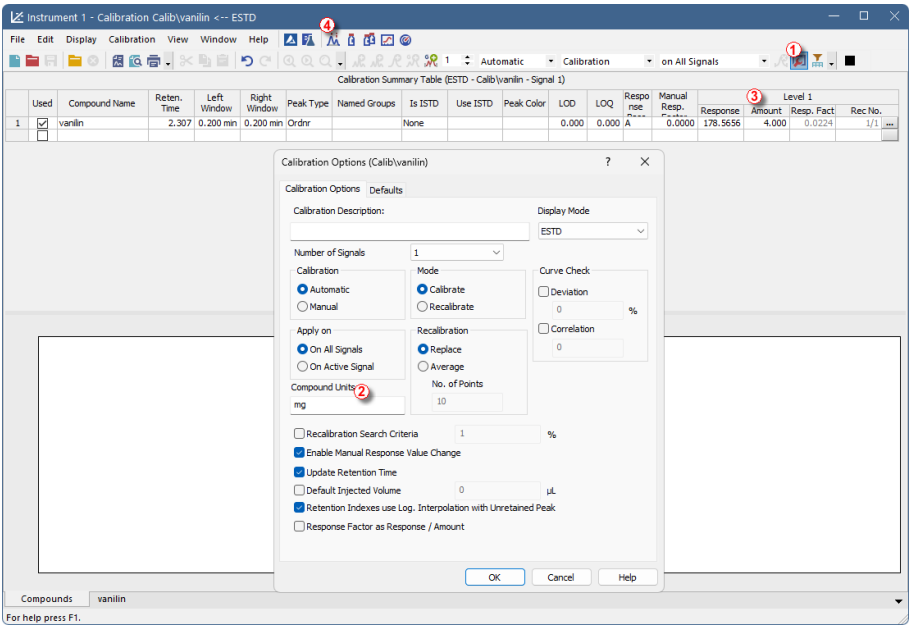
1. Open your calibration/create a new one in the *Calibration* window.
2. Click the icon to open *Calibration Options* dialog ① .
3. Specify used unit for the amount in *Compound Units* field ② . Confirm changes by clicking *OK* button.

Note: Fill the unit used for the total amount of sample. In our specific case sample was weighted and *mg* is used. Set units are used in the *Chromatogram* window for further calculations.

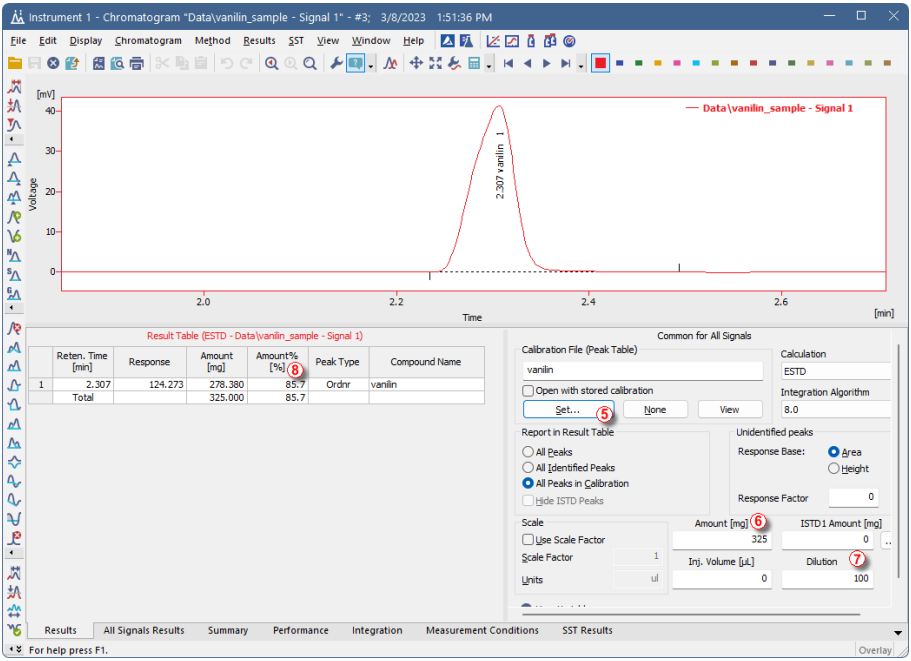
4. Fill corresponding *Amount* for each compound as concentration ③ .

Note: That means amount divided by the volume of solvent used to prepare the standard solution. In our example *400 mg* of **vanilin** was dissolved in *100 ml*, thus the *Response* from the standard chromatogram corresponds to *4 mg/ml* which is the value entered in the *Amount* field in the calibration.

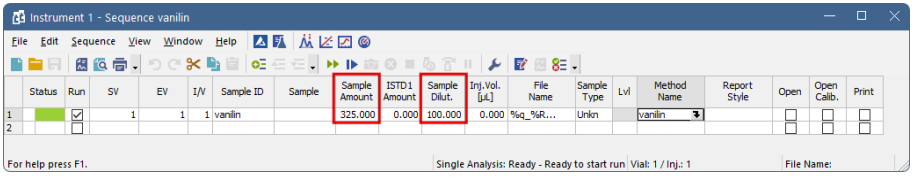
5. Save the calibration and open *Chromatogram* window ④ .



6. Open chromatogram(s), assign each chromatogram prepared calibration using the *Set...* button ⑤ .
7. Fill the amount of the sample that had been used ⑥ . *Amount* refers to the mass of the sample used. Units are automatically copied from the *Calibration Options* (see step 3). Amount replaces the *Total* value in the *Amount* column of the *Result Table*. In our example: *325 mg* of sample had been used.
8. Fill the dilution that had been used ⑦ . *Dilution* refers to the solvent volume that has been used to dilute the sample. Dilution multiplies the values in the *Amount* column of the *Result Table*. In our example: *325 mg* of sample had been diluted by *100 ml* of solvent.
9. Column *Amount%* in the *Result Table* now displays the percentage amount of the compound in the sample ⑧ .

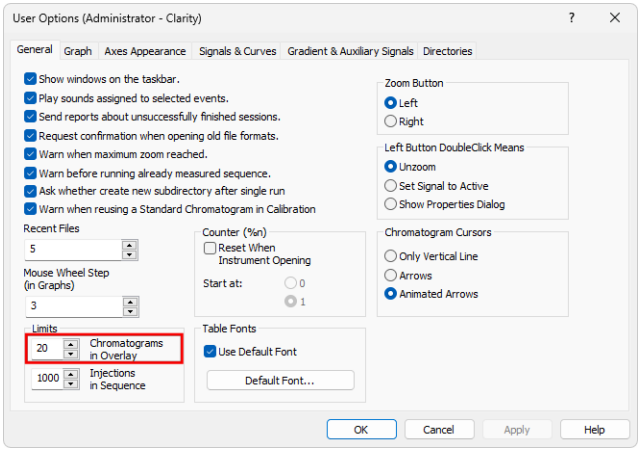


Note: It is possible to automatize this process. Calibration can be linked to the used method so it is automatically linked. *Amount* and *Dilution* can be pre-filled in *Single Analysis* or *Sequence* windows. Beware that columns in *Sequence* have slightly different names - *Sample Amount* and *Sample Dilut.*



10.9 Comparing the results from several chromatograms

The default maximum number of chromatograms in Overlay is set to 20. In case summary report should include more chromatograms, this limit can be changed in the User Options window, which can be invoked from the *Instrument window* by using *Setting - User Options....*

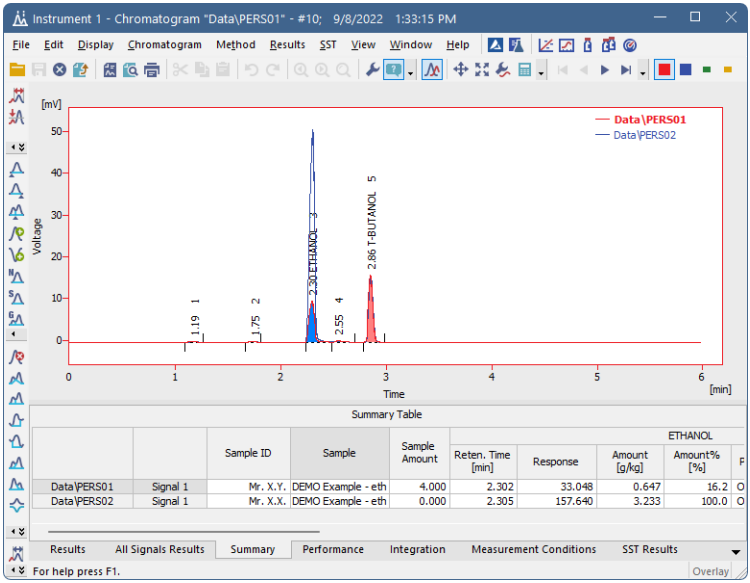


1. Open the chromatograms using *File - Open Chromatogram...*. In *Open Chromatogram* dialog select desired chromatograms and click *Open in Overlay*.

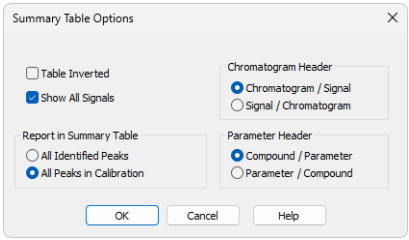
   Note: You can also use *File - Open Chromatograms From Sequence...* command which opens all chromatograms from sequence including Standards.

2. Click on the *Summary* tab in the lower part of the window to display the *Summary Table*. In the rows you can see chromatograms and signals with measured values and in the columns there are identified peaks from all calibrated chromatograms.

   Caution: *Summary Table* is based on calibration meaning only calibrated peaks will be present in it.



3. Right click on the *Summary Table*, if you want to change the visualization of the table or add a custom column, etc.
4. To see all signals, select the *Show All Signals* checkbox in the *Summary Table Options* dialog accessible from the pop-up menu of the *Summary Table*. By default, only signals containing calibrated peaks are visible in the *Summary Table*.



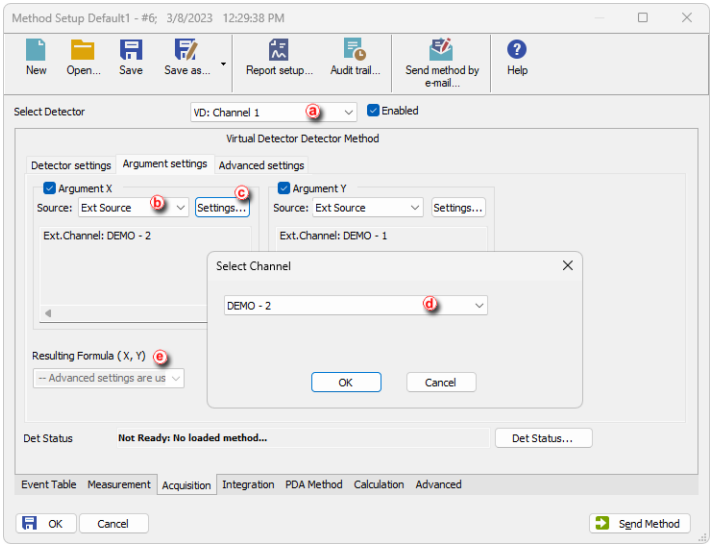
Note: It is also possible to compare parameters from different chromatograms and check if they fall within set limits by using the SST Extension.

10.10 Confirming the identity of a compound by using the signal ratio

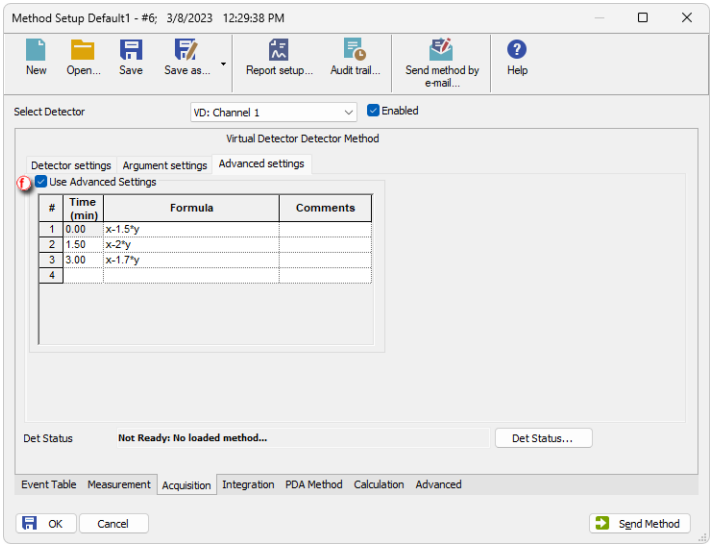
The identity of a compound can be confirmed by using a dual wavelength detector in conjunction with the Virtual Detector and checking if the signals ratio is constant with the following procedure. Prerequisite to this is a know signal ratio for used wavelengths. With correct comparison formula the Virtual Detector signal should

result in flat line when the peak is pure compound, impurities with different signal ratio would be represented as response change on the Virtual Detector signal.

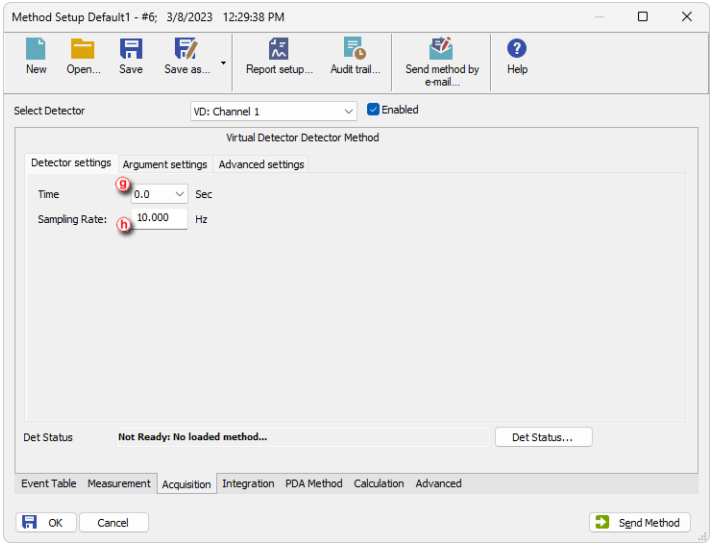
1. Add the Virtual Detector as well as your dual wavelength detector to the instrument, if not already present, based on chapter *Adding a new device*.
2. Open the Method Setup dialog and navigate to *Acquisition* tab.
3. Select *VD: Channel 1* in the *Select Detector* drop-down list ⓐ .
4. On the *Argument settings* tab, tick the *Argument X* checkbox, select an *Ext. Source* ⓑ , then click on the *Settings* button ⓒ , select the appropriate detector *Channel* ⓓ and click *OK* .
5. Repeat the same steps for *Argument Y* and select the second channel from the detector.
6. If want to use the same signal ratio for entire analysis you can fill in the Resulting Formula field ⓔ with the proper formula. If you need multiple formulas for different times skip this step and continue with the next one.



7. To set multiple formulas for different time intervals navigate to the *Advanced settings*, check the *Use Advanced Settings* ⓕ checkbox and fill the time table as needed.



8. On the *Detector settings* tab, set the *Time* to 0 ⓖ to avoid distortion and fill *Sampling Rate* ⓗ equal to the actual detector sample rate.



Method prepared this way can be used to check compound identity as mentioned in the beginning of this chapter.

11 Data Reports

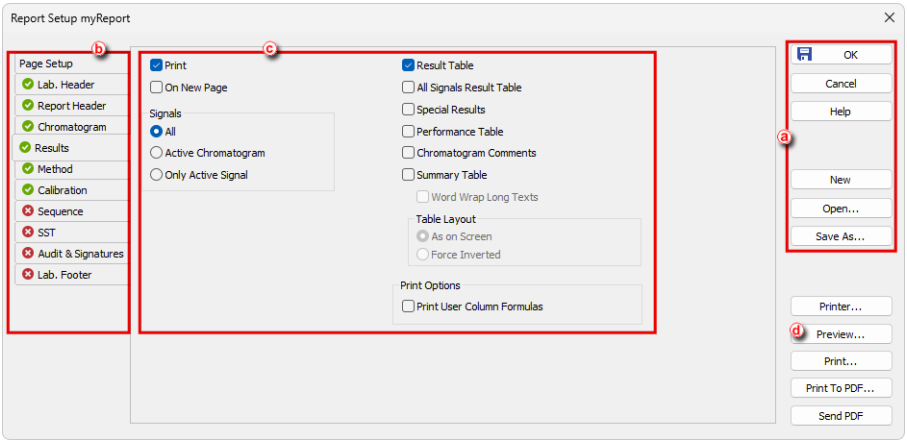
Following chapters describe how to create a report, adjust it and print.

11.1 Setting up a report style for printing

It is possible to select what information and how you want to have printed in the report. You can create different report styles and store them in different *.STY files.

To see an example on how to set up a style to obtain a specific layout see chapter *Creating a report (example)*.

1. From the *Instrument, Calibration, Chromatogram* or *Sequence* windows, select *File - Report Setup*. From the *Method Setup* window click the *Report Setup* button.
2. Click the *New* button if you wish to create a new report style or click the *Open* button ⓐ to open existing report style. Name of currently opened style is included in the window header.
3. Select the tab corresponding to the section you wish to modify ⓑ .
4. Click and drag the tabs to a new position if you wish to change the order in which they will be printed in the report. Tabs can also be moved via context menu which is invoked after right-clicking on them. The first two (*Page Setup, Lab. Header*) and last two (*Audit & Signatures, Lab. Footer*) tabs have fixed position.
5. Select options you would like to include in that section. ⓒ
6. Click *Preview* ⓓ to see the result preview and repeat steps 3.-5. if you wish to modify anything.
7. Click *Save As* to save the report style under a new name or *OK* to accept the changes to the current style. ⓐ



11.2 Printing or previewing a report

The report may include information about methods, calibrations, chromatograms etc. It is possible to setup what information you would like to include in the report as explained in *Setting up a report style*.

Caution: While setting reports it is necessary to pay attention to several crucial settings bellow:

- The information on the report will vary depending on which window we open it from, even if we use the same report style.
  For example, the Chromatogram tab in the Chromatogram Report Setup refers to the chromatograms opened in that specific window. Same tab in the Calibration Report Setup refers to the calibration standard opened in the Calibration window if any. In the Instrument window it will refer to the chromatograms produced after the last run or analysis.
- Only opened chromatograms will be printed. For this reason when setting up automatic printing from *Single Analysis*, *Sequence*, *Batch* windows etc. *Open in Chromatogram Window* option must always be used as well.
- If you set up the report to be printed automatically it is always necessary to specify *Report Style* to be used.

Note: It is also possible to report multiple already measured chromatograms at once in Batch dialog. See Post Run Options from Batch topic for more info.

For more info regarding reports see the **Clarity Reference Guide**.

To *Print* a Report:

1. Select *File - Print* in the *Instrument, Calibration, Chromatogram* or *Sequence* windows. In *Method Setup* click *Report Setup* icon and in the following dialog click *Print...*.

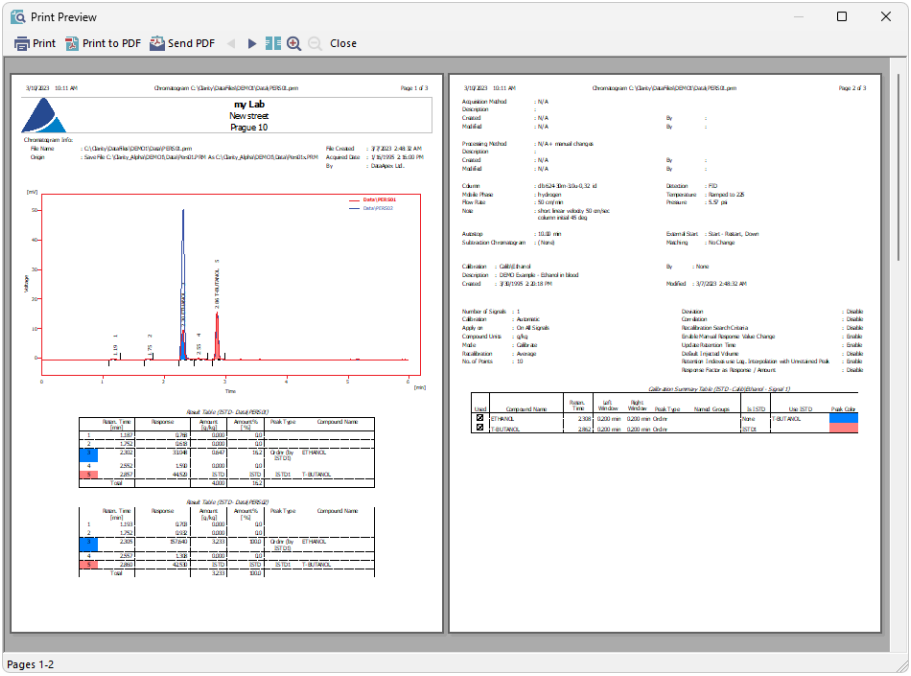
2. *Print* dialog will open for you to setup your printing options and confirm the printout.

To *Print to PDF*:

1. Select *File - Print to PDF* in the *Instrument, Calibration, Chromatogram* or *Sequence* windows. In *Method Setup* click *Report Setup* icon and in the following dialog click *Print to PDF*.
2. *Print to PDF* dialog will open for you to enter file name confirm the saving of the file.

To display *Print Preview*:

1. This feature is useful while preparing your own report style to check whether the outcome is as needed.
2. Select *File - Print Preview* in the *Instrument, Calibration, Chromatogram* or *Sequence* windows. In *Method Setup* click *Report Setup* icon and in the following dialog click *Preview...*.
3. *Print Preview* dialog will open. From there you can browse or print the report.

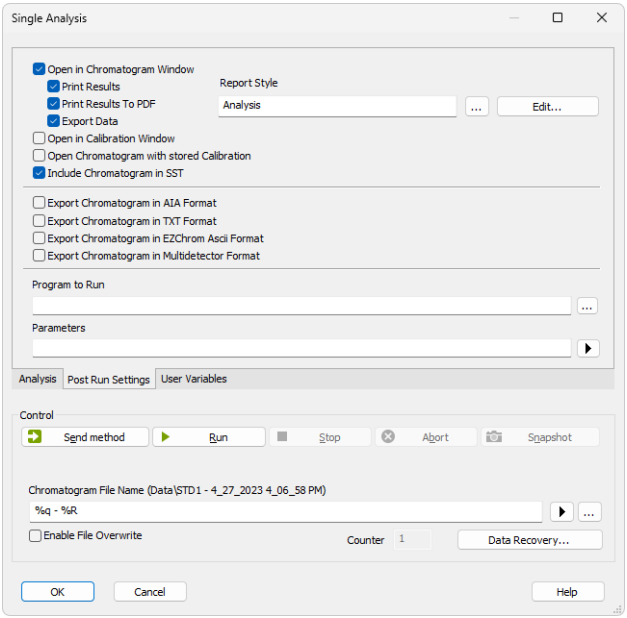


Automatic report generating after analysis is finished:

By default generated reports are placed next to the original files, to change it use *Setting - User Options* in the *Instrument* window. Navigate to *Directories* tab and specify your custom path.

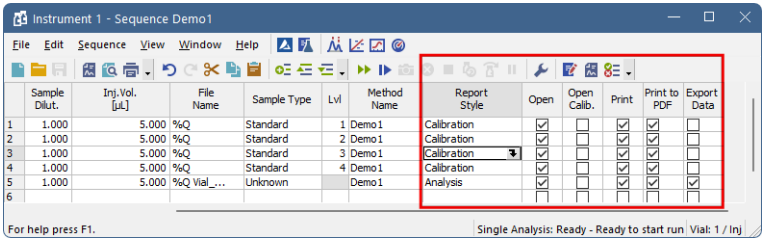
Single Analysis

1. Navigate to *Single Analysis - Post Run Settings* tab.
2. Check options *Open in Chromatogram Window*, *Print Results* and/or *Print Results to PDF*.
3. Select *Report Style* by clicking ... button.
4. It is also possible automatically export data, to do so check *Export Data* option. What should be exported can be set by using *Setting - Export Data* in the *Instrument* window.



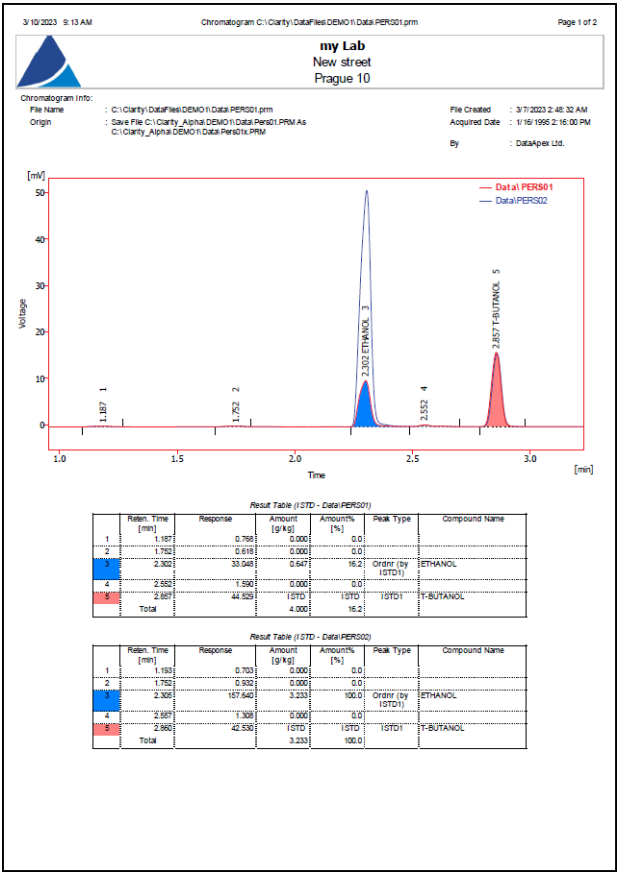
Sequence

1. Procedure is similar to the single analysis. In *Sequence* window some of the required columns are hidden by default, to display them right-click the sequence table and use *Setup Columns* in the following dialog move desired columns to the *Show Columns* list.
2. Check checkboxes in columns *Open*, *Print* and/or *Print to PDF* on rows where report should be generated.
3. Select *Report Style* for each row where report is generated. It is possible to modify specif report style by selecting its cell and clicking .
4. It is also possible automatically export data, to do so check *Export Data* option. What should be exported can be set by using *Setting - Export Data* in the *Instrument* window.



11.3 Creating a report style (example)

This is an example on how to setup a new report style to obtain the results shown in the picture. Beware that tables are printed as on screen, if e.g., calibration table is to wide to fit on one page you can adjust the column width in *Calibration* window.



3/10/2023 9:13 AM Chromatogram C:\Clarity\DataFiles\DEMO1\Data\PERS01.prm Page 1 of 2

my Lab
New street
Prague 10

Chromatogram Info:

File Name : C:\Clarity\DataFiles\DEMO1\Data\PERS01.prm
Origin : Save File C:\Clarity_Alpha\DEMO1\Data\Pers01.PRM As C:\Clarity_Alpha\DEMO1\Data\Pers01x.PRM

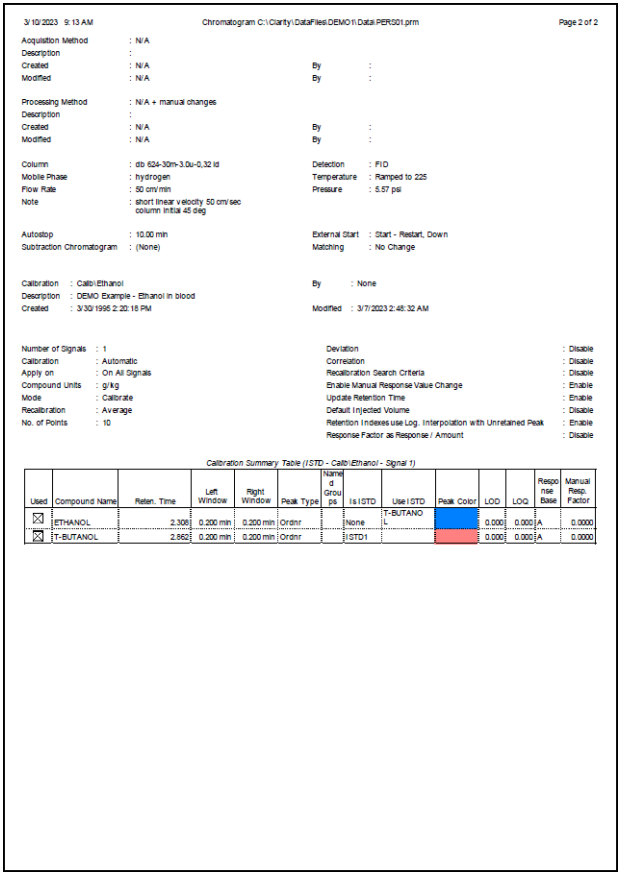
File Created : 3/7/2023 2:48:32 AM
Acquired Date : 1/16/1995 2:16:00 PM
By : DataApex Ltd.

Result Table (ISTD - Data\PERS01)

	Reten. Time [min]	Response	Amount [g/kg]	Amount% [%]	Peak Type	Compound Name
1	1.187	0.768	0.000	0.0		
2	1.752	0.618	0.000	0.0		
3	2.302	33.048	0.647	16.2	Ordnr (by ISTD1)	ETHANOL
4	2.552	1.590	0.000	0.0		
5	2.857	44.529	ISTD	ISTD	ISTD1	T-BUTANOL
	Total		4.000	16.2		

Result Table (ISTD - Data\PERS02)

	Reten. Time [min]	Response	Amount [g/kg]	Amount% [%]	Peak Type	Compound Name
1	1.193	0.703	0.000	0.0		
2	1.752	0.932	0.000	0.0		
3	2.308	157.640	3.233	100.0	Ordnr (by ISTD1)	ETHANOL
4	2.557	1.309	0.000	0.0		
5	2.868	42.530	ISTD	ISTD	ISTD1	T-BUTANOL
	Total		3.233	100.0		



3/10/2023 9:13 AM Chromatogram C:\Clarity\DataFiles\DEMO1\Data\PERS01.prm Page 2 of 2

Acquisition Method : N/A
Description :
Created : N/A By :
Modified : N/A By :

Processing Method : N/A + manual changes
Description :
Created : N/A By :
Modified : N/A By :

Column : db 624-30m-3.0u-0.32 id
Mobile Phase : hydrogen
Flow Rate : 50 cm/min
Note : short linear velocity 50 cm/sec column initial 45 deg

Detection : FID
Temperature : Ramped to 225
Pressure : 5.57 psi

Autostop : 10.00 min
Subtraction Chromatogram : (None)

External Start : Start - Restart, Down
Matching : No Change

Calibration : Calib\Ethanol
Description : DEMO Example - Ethanol in blood
Created : 3/30/1996 2:20:16 PM

By : None
Modified : 3/7/2023 2:48:32 AM

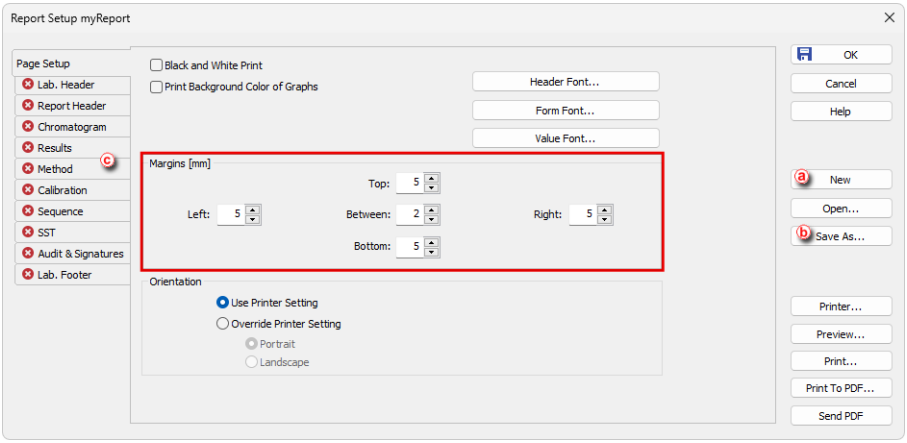
Number of Signals : 1
Calibration : Automatic
Apply on : On All Signals
Compound Units : g/kg
Mode : Calibrate
Recalibration : Average
No. of Points : 10

Deviation : Disable
Correlation : Disable
Recalibration Search Criteria : Disable
Enable Manual Response Value Change : Enable
Update Retention Time : Enable
Default Injected Volume : Disable
Retention Indexes use Log. Interpolation with Unretained Peak : Enable
Response Factor as Response / Amount : Disable

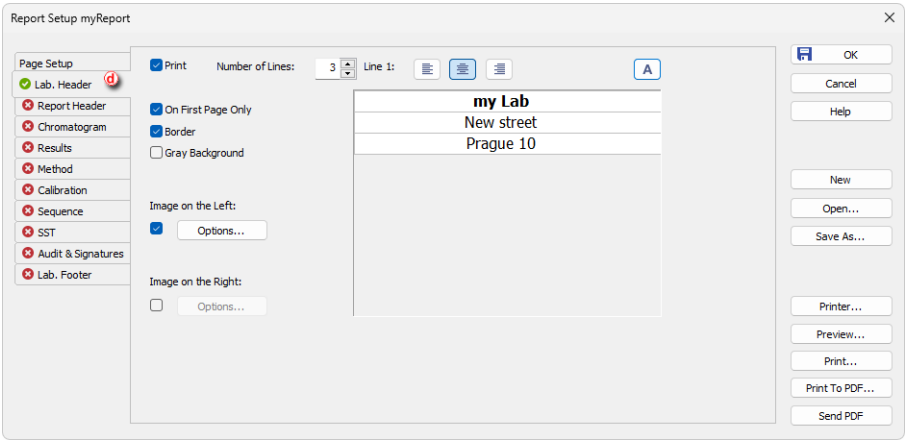
Calibration Summary Table (ISTD - Calib\Ethanol - Signal 1)

Used	Compound Name	Reten. Time	Left Window	Right Window	Peak Type	Named Groups	Is ISTD	Use ISTD	Peak Color	LOD	LOQ	Response Base	Manual Resp. Factor
☒	ETHANOL	2.306	0.200 min	0.200 min	Ordnr		None	T-BUTANOL		0.000	0.000	A	0.0000
☒	T-BUTANOL	2.662	0.200 min	0.200 min	Ordnr		ISTD1			0.000	0.000	A	0.0000

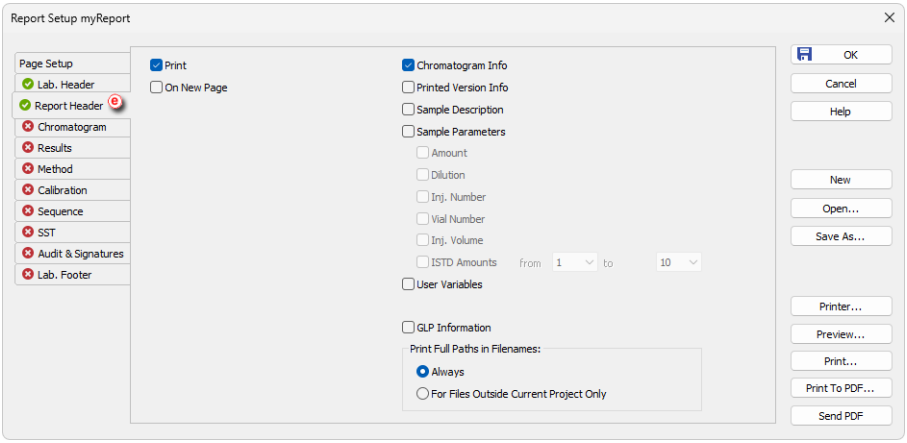
1. Open the chromatograms PERS01 and PERS02 in overlay (located in C:\CLARITY\DATAFILES\DEMO1\DATA).
2. Select the graph area you want to print by clicking and dragging the cursor in the chromatogram graph.
3. Create a new report style.
   - Select the *File - Report Setup*.
   - Click on the *New* button ⓐ to create a new report style.
   - Click on the *Save As...* button ⓑ and save the style under the name (*myReport* in this example).
4. Click and drag the tabs ⓒ to set them in desired order. This will be the order the sections will have in the report.
5. Set the *Margins* as needed.



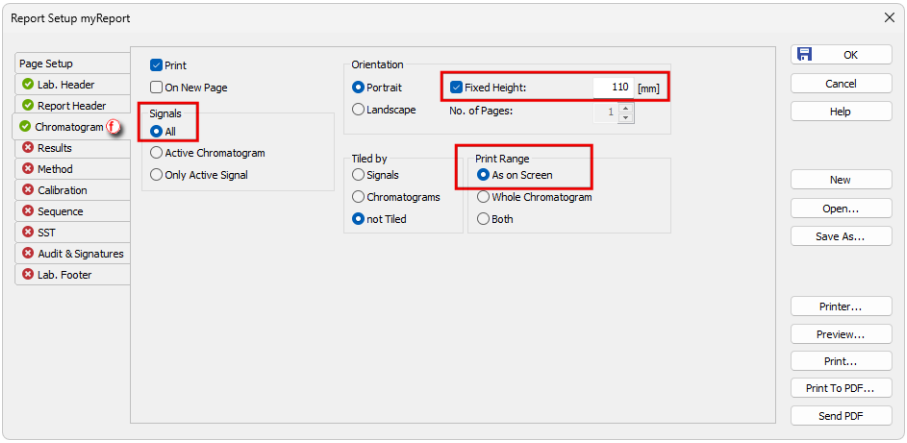
6. Set the *Lab Header*.
    - Click the *Lab Header* tab ⓓ .
    - Check the checkboxes *Print*, *On First Page Only* and *Border*.
    - Check the *Image on the Left* checkbox and click on the *Options...* button to select the logo.
    - Click in each line in the text box and write the you want to have in the header.
    - Click in the first line of text and then on the *Font* icon (icon with "*A*") and select *Bold* as a *Font Style*.



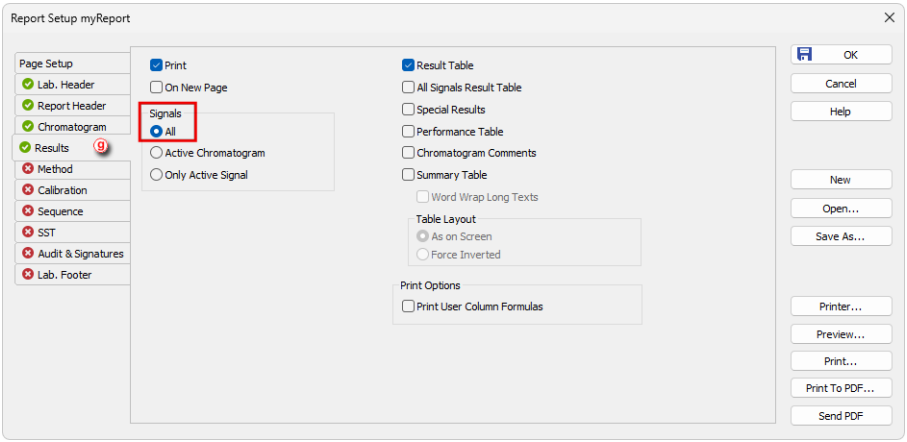
7. Set the *Report Header*.
    - Click the *Report Header* tab ⓔ .
    - Check the options *Print* and *Chromatogram Info*.



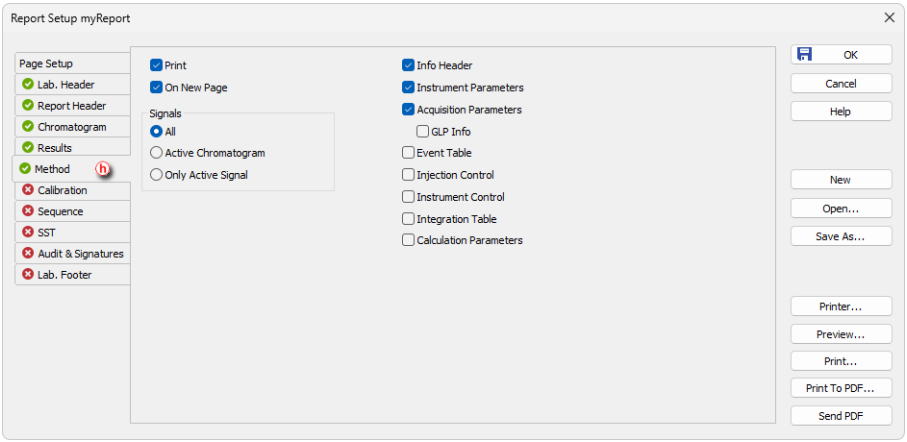
8. Set the *Chromatogram section*. Only the format of printed graphs is set here, result tables are printed based on *Results* tab, see next step.
    - Click the *Chromatogram* tab ⓕ .
    - Check the options *Print* and *Fixed Height* and set the height to 110 mm.
    - Select the options *Signals - All* and *Print range - As On Screen*.



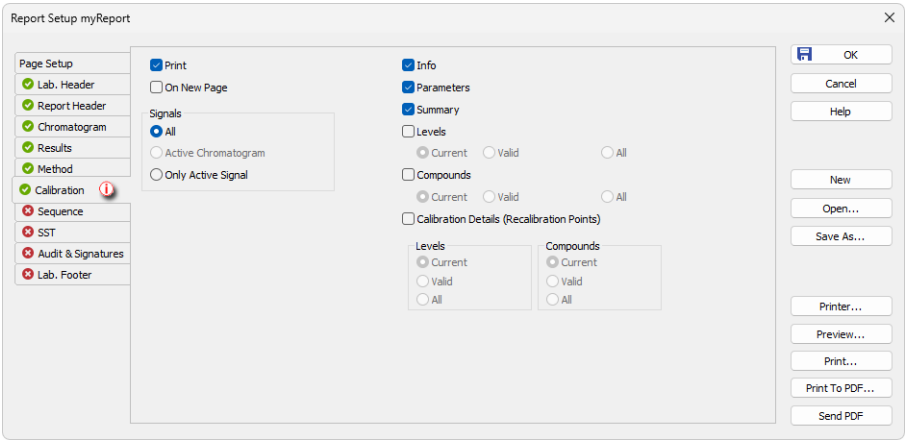
9. Set the *Results* section.
    - Click the *Results* tab ⓖ .
    - Check the options *Print* and *Result Table*.
    - Select the option *Signals - All*.



10. Set the *Method* section.
    - Click the *Results* tab. ⓗ
    - Check the options *Print*, *On New Page*, *Info Header*, *Instrument Parameters* and *Acquisition Parameters*.
    - Select the option *Signals - All*



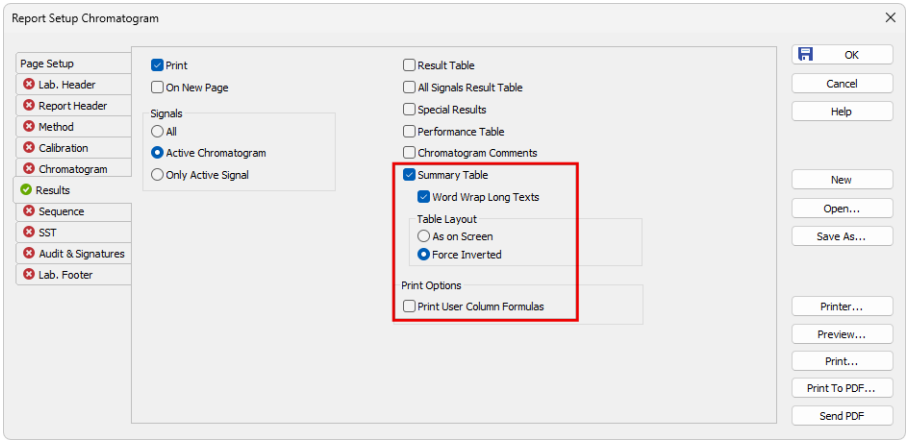
11. Set the *Calibration* section.
    - Click the *Calibration* tab ⓘ .
    - Check the options *Print*, *Info*, *Parameters* and *Summary*.



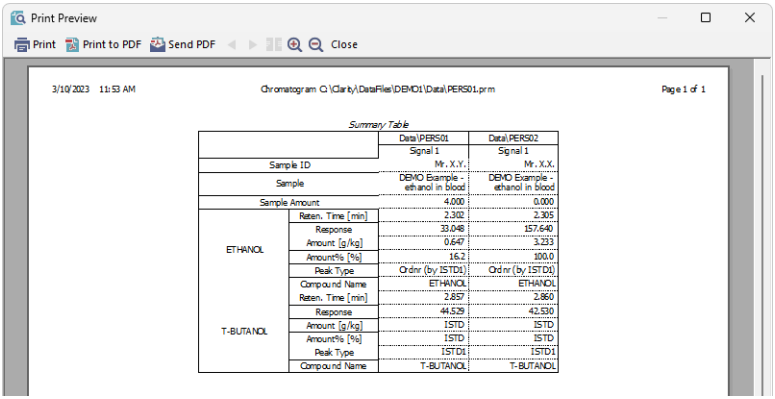
12. You can preview your report by clicking *Preview...*. For more info see ***Printing or previewing a report***.
13. Click the *OK* to save the report style.

11.4 Printing the summary table

1. Setup *Summary* table in *Chromatogram* window. For information on how to create Summary Table, see the chapter **"Comparing the results from several chromatograms"** on pg. **156**.
2. Right-click in the *Summary Table*, if you want to change the visualization of the table, add a custom column, etc.
3. Open the *Report Setup* dialog and check the *Summary Table* checkbox in the *Results* tab.
4. It is also recommended selecting *Word Wrap Long Texts* and *Force Inverted* (table will be printed with inverted layout ignoring the on-screen layout which is more suitable for reports) options. *Print User Column Formulas* can be used when custom columns are included in the table.



5. The results for all opened chromatograms will be printed.



12 File Management

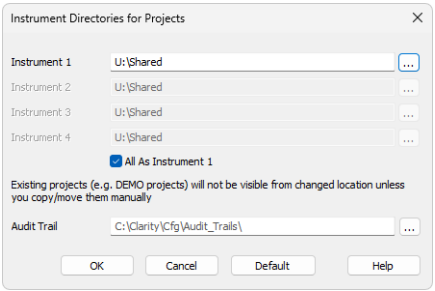
How to set project directories, create new projects, preset file names of measured chromatograms based on variables, store files into subfolders etc.

12.1 Setting up project directories

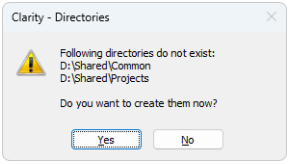
It is possible to set different directories for projects from each instrument and to set Audit Trail directory. It is useful while working with shared disk and it is needed to access measured data from another PC using **Clarity Offline**.

To setup custom project directories do following:

1. Open Main **Clarity** window.
2. Open the *Instrument Directories for Projects* dialog using the *System - Directories...* command or the icon.



3. Set the desired directory e.g., *U:\Shared*.
4. If needed change the *Audit Trail* directory as well.
5. When saving the changes, the following dialog may appear. Confirming it will create the necessary subfolder structure in your target location. COMMON is filled by default files that are necessary for functioning.



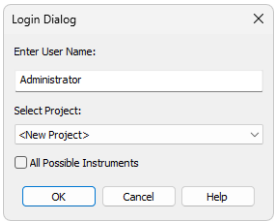
Caution: Be aware that when updating Clarity the COMMON files outside of the installation directory must be updated manually.

12.2 Creating a new project

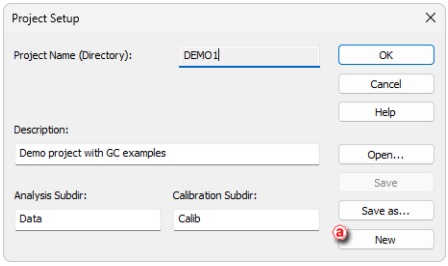
Creating a new project ensures that the measured data will be later easily found. The project itself is a directory in **Clarity's** DATAFILES subfolder or selected directory where all relevant files are saved (methods, calibrations, sequences, chromatograms).

1. To create new project navigate to the *Project Setup* dialog by selecting *Instrument - Project...* in the *Instrument* window.

Note: You can also open a project through the *Login Dialog* opened from the Main Clarity window by selecting the *New Project* option and clicking OK.

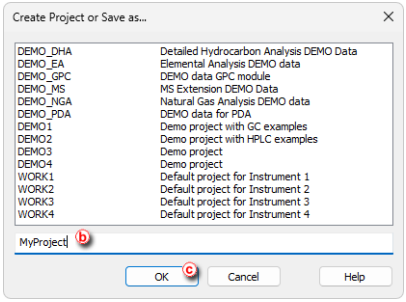


2. In the *Project Setup* dialog click the *New* button ⓐ to create a new project.



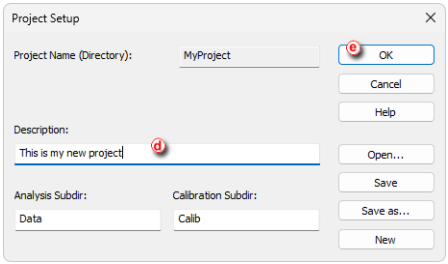
3. Set the name of the new project ⓑ and click *OK* ⓒ .

Note: Entered project name must not contain invalid characters, i. e. \ /:*?"< >|.



4. Fill in the project description in the *Description* field ⓓ and click *OK* ⓔ .

Note: The change of the project will require you to restart the Instrument. If there are any unsaved files opened, you will be prompted to save them. The newly created project does not include any files (for example method, calibration etc.).



12.3 Creating customized file names automatically

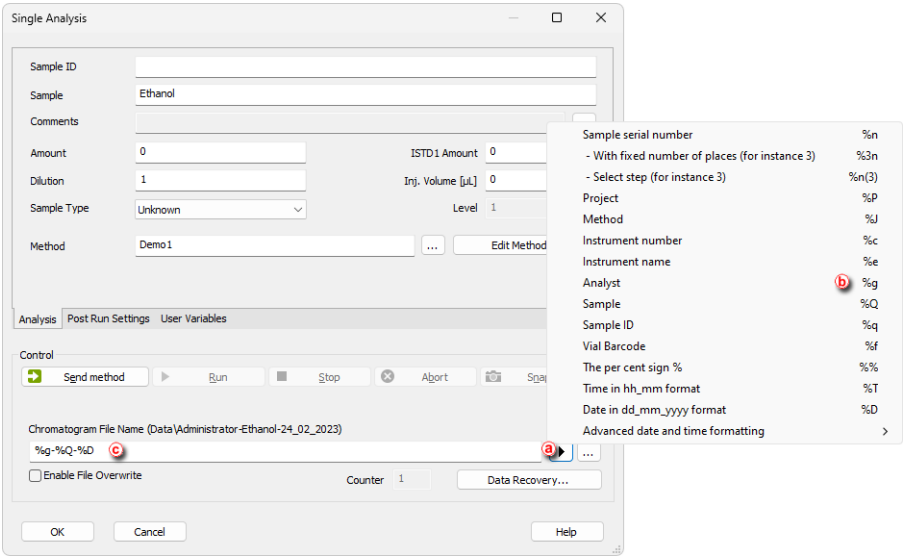
You can create customized file names automatically by appending variables to them.

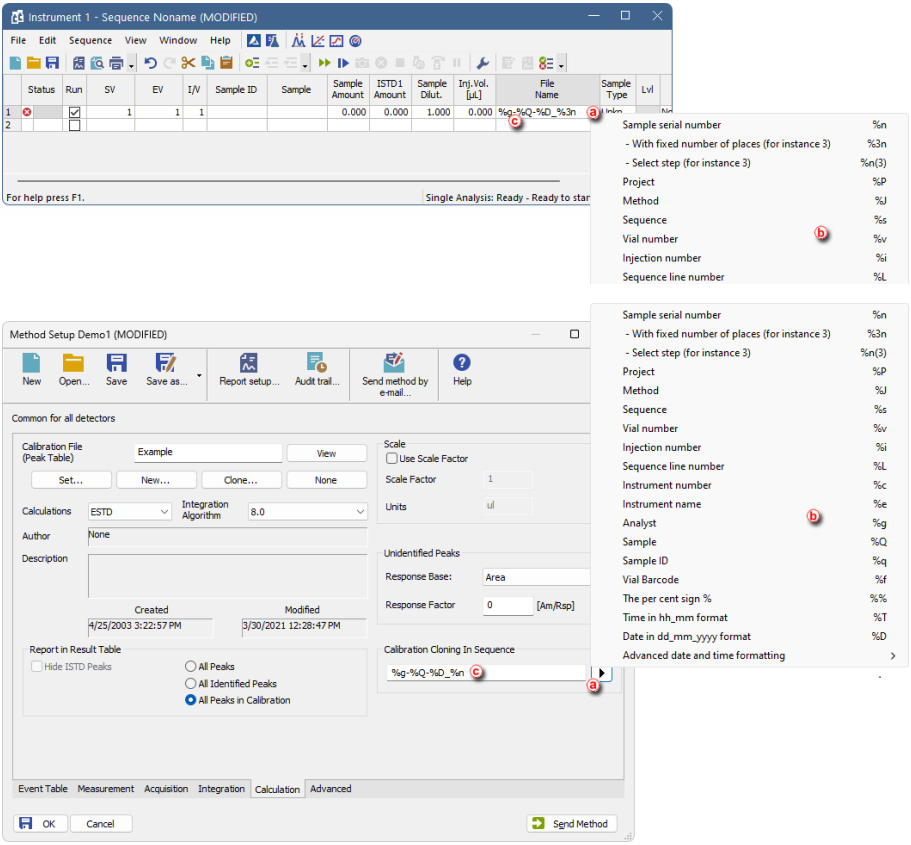
File names can be created in the *Single Analysis* window, in the *Sequence* window and on the *Method Setup - Calculation* tab.

1. Click on the respective icons , , in the *Instrument* window to open the *Single Analysis*, the *Sequence* or the *Method Setup - Calculation* window.
2. Click on to open the variable list. ⓐ
3. Select the variable you want to include in the file name. ⓑ
4. Repeat the previous two steps to add more variables.
5. Insert any allowed characters between the variables to create your file name. ⓒ

More Info:

- Variables are preceded by the "%" character and they are substituted by their value upon file creation. For example "*%g-%Q-%D*" will create a file name with the name of the *Analyst*, the *Sample* and the *date*: "*Administrator-Ethanol-24_02_2023.prm*".
- To prevent an *Unresolved File Name* error you can append the *Sample Serial Number* "%n" or the date and time "%R".


Single Analysis
Sample ID
Sample
Ethanol
Comments
Amount
0
ISTD1 Amount
0
Dilution
1
Inj. Volume [µL]
0
Sample Type
Unknown
Level
1
Method
Demo1
Edit Method
Analysis
Post Run Settings
User Variables
Control
Send method
Run
Stop
Abort
Chromatogram File Name (Data\Administrator-Ethanol-24_02_2023)
%g-%Q-%D
Enable File Overwrite
Counter
1
Data Recovery...
OK
Cancel
Help
Sample serial number %n
- With fixed number of places (for instance 3) %3n
- Select step (for instance 3) %n(3)
Project %P
Method %J
Instrument number %c
Instrument name %e
Analyst %g
Sample %Q
Sample ID %q
Vial Barcode %f
The per cent sign % %%
Time in hh_mm format %T
Date in dd_mm_yyyy format %D
Advanced date and time formatting
a
b
c



12.4 Storing files into project subfolders

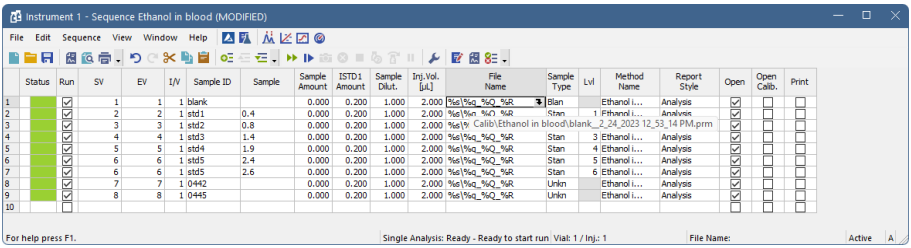
It is possible to include the name of a subfolder in the *File name* to store chromatograms into project's subfolder. If the subfolder doesn't exist, it will be created when the chromatogram is stored. The directory can be set using the **"\"** character. Any string of the variables or characters in front of the **"\"** character is considered as the subfolder name. Any string of variables or characters after the **"\"** character is used as the file name.

Subfolders created this way are present in DATA and CALIB subfolders of used project.

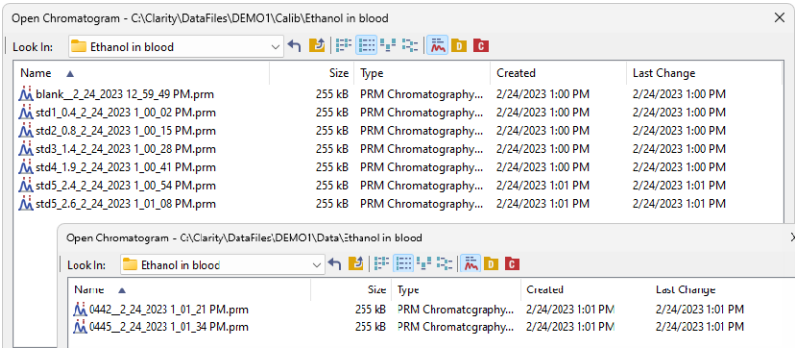
Here we provide an example on how to store chromatograms from one sequence into subfolder named after it.

1. Login to *Instrument* with DEMO1 project.
2. Open the *Sequence* window.
3. Open ETHANOL IN BLOOD.SEQ.

4. Set the File Name of the first row to **%s\%q_%Q_%R**. (To see detailed description on how to create the name using variables see chapter "Creating customized file names automatically" on page 176.)
5. Right click on the *File Name* column and select *Fill down* to apply the name to all rows.



6. The Chromatogram file with a name of SAMPLEID_SAMPLE_DATE-AND-TIME will be stored in *Demo1\Data\Ethanol in blood* folder for unknown samples and in *Demo1\Calib\Ethanol in blood* folder for standards and blank.



- To create subfolder for each month use the %m or %B variables and for each day use the %a or %A variables.
- Same procedure can be also used in *Single Analysis* window.

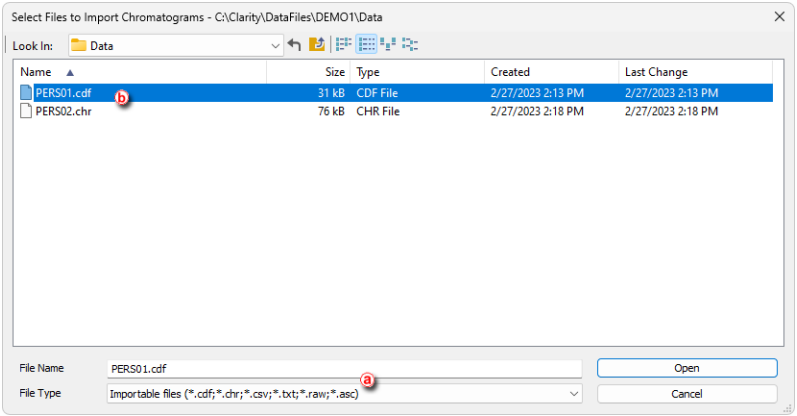
13 Import and Export Data

Clarity allows to import or export chromatograms from/to various formats. Following chapters describe you how to, for example, set exporting chromatograms to a LIMS.

13.1 Importing a chromatogram into Clarity

It is possible to import chromatograms from other chromatography software. Supported formats are : AIA (*.CDF suffix), EZChrom ASCII (*.ASC suffix), Text format (*.TXT), Comma Separated Values format (*.CSV) or Multi-detector format (*.CHR). The particular procedure depends on type of file you want to import.

1. From the *Chromatogram* window open the *Open Files To Import* dialog by selecting *File - Import Chromatogram…*.
2. You can select the *File Type* which you want to import ⓐ . By default you can see all supported formats.
3. Select the file(s) you want to import ⓑ and click on the *Open* button.



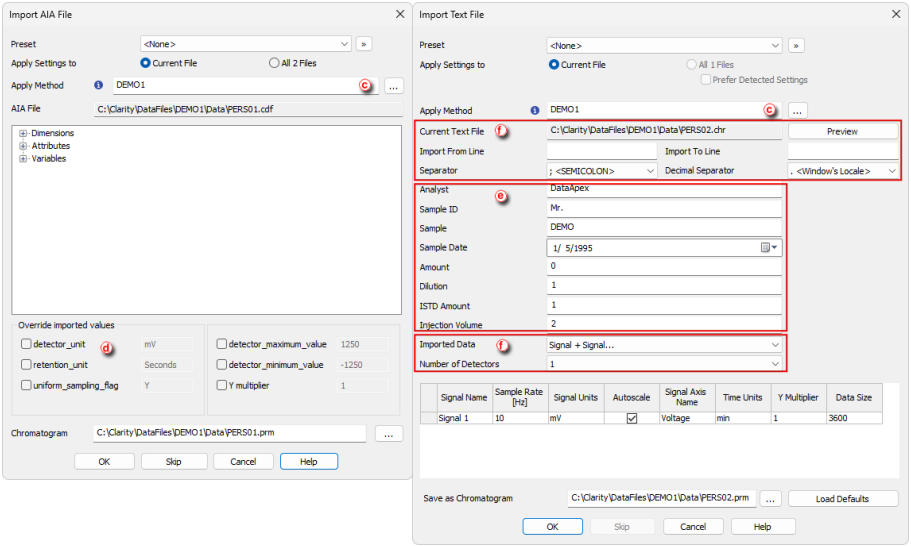
4. Set the parameters as needed in the subsequent dialogs depending on the file format.
5. For all file formats you can select imported file name and which method to apply ⓒ . This will apply integration, calibration etc.

- Import AIA File - when importing AIA file (*.CDF suffix).
  - You have option to inspect data from the source file and override some of them ⓓ e.g, *Detector Unit*.
- Import Text File - when importing Text, Multidetector or EZChrom ASCII file (*.TXT, *.CHR, *.ASC and *.CSV suffixes).
  - You can set information regarding given sample ⓔ like *Dilution* or *Injection Volume* etc. You can also set information regarding source file

format ⓕ .

Note: File format related settings is typically detected automatically and there is no need to change it.

- *Save As* - when importing *.RAW file, as there is no need to set other parameters.



13.2 Exporting a chromatogram from Clarity to a different chromatography data station

It is possible to export chromatograms to other formats used by chromatography data stations. The supported formats are : *AIA* (*.CDF suffix), *EZChrom ASCII* (*.ASC suffix), *Text format* (*.TXT), or *Multi-detector format* (*.CHR).

1. Open the *Chromatogram* window by selecting *Window - Chromatogram* on the *Instrument* window or click on  .
2. Open the chromatogram you want to export, then open the *Export Chromatogram* dialog by selecting *File - Export - Export Chromatogram….*
3. Select the export format ⓐ . Notes:

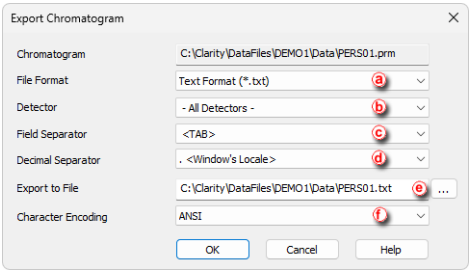
   Note: For PDA export of 3D data only the EZChrom ASCII format is supported.

4. Select the signals to be exported ⓑ .

   Note: You can choose individual signals, *All Detectors*, *3D Data* or *All Detectors + 3D Data* (3D Data variants in EZChrom ASCII only).

5. Select the field separator character ⓒ .

6. Select the decimal separator character ⓓ .
7. Open the *Export Chromatogram As* dialog clicking on ... . Enter the name and location of the file into which you wish to export the chromatogram ⓔ or fill in the file name.
8. Select the character encoding for the exported file (*ANSI* or *Unicode (UTF-8)*) ⓕ .
9. Click *OK* to finish the export.



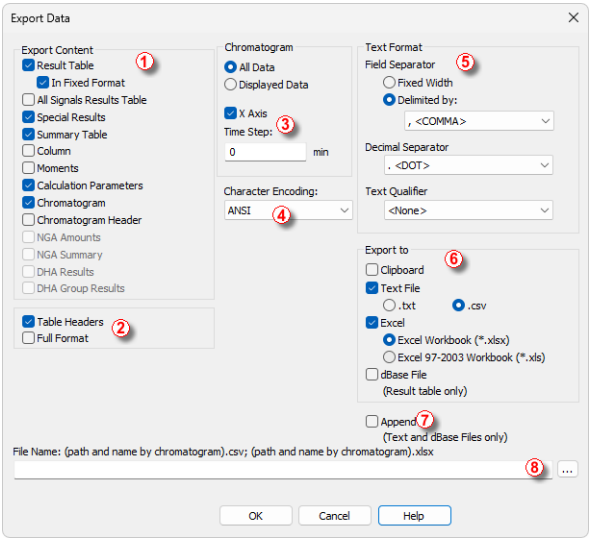
13.3 Exporting data from Clarity

It is possible to export selected parts of the results into external files. All of this can be achieved automatically from *Sequence* or *Single Analysis* window by selecting *Export Data* in *Postrun Options* or manually from the *Chromatogram* window.

1. Open *Export Data* dialog from *Instrument* window by using *Setting - Export Data...*
2. Select which data you want to include in the exported files in the *Export Content* group ① . Detailed description of these fields can be found in the Reference Guide.
3. Select whether you want the tables exported with headers and/or in the full format ② .
4. Select whether the exported chromatogram data will contain time column or not. If you intend to perform bunching for the exported data specify the time period for the bunching ③ .

   Note: The mean value for selected *Time step* period is calculated as a sum of the highest Y_{Max} and lowest Y_{Min} signal curve value found within each *Time Step* period divided by two - $(Y_{Max}+Y_{Min})/2$.

5. Select the character encoding to be used (*ANSI* or *Unicode (UTF-8)*) ④ .
6. Select the appropriate text formatting for the export ⑤ .
7. Select the format or formats to which the data will be exported. You can choose any number of export types at the same time ⑥ .
8. Select whether the exported data should be appended into an existing file ⑦ (in this case file name ⑧ must be filled in) or exported as a new file (leave the file name empty as shown bellow).



- This setting will be used for automated data export from *Sequence* or *Single Analysis*.
- To manually export data you can invoke the same dialog from *Chromatogram* window by using *File - Export - Export Data...* command or icon. In this case dialog also includes *Export* button which can be used the export data from currently opened chromatogram.

13.4 Exporting data for LIMS

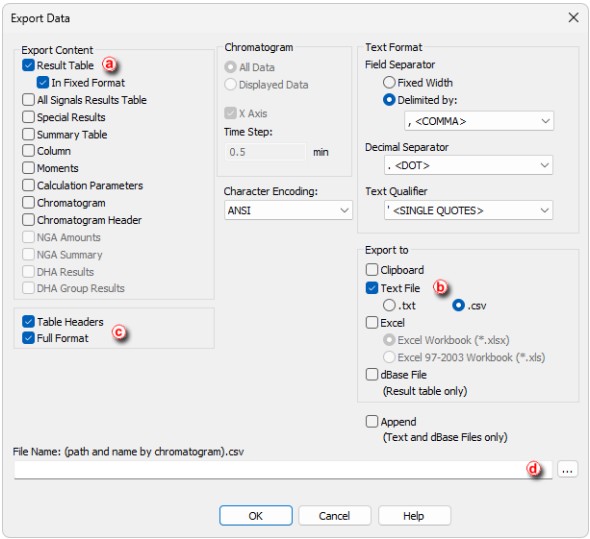
Following chapter describes how to export data from Clarity to be used for LIMS. Each LIMS requires a different approach in importing external data, therefore it may be necessary to adjust your LIMS to be able to correctly process imported data from Clarity.

Export settings

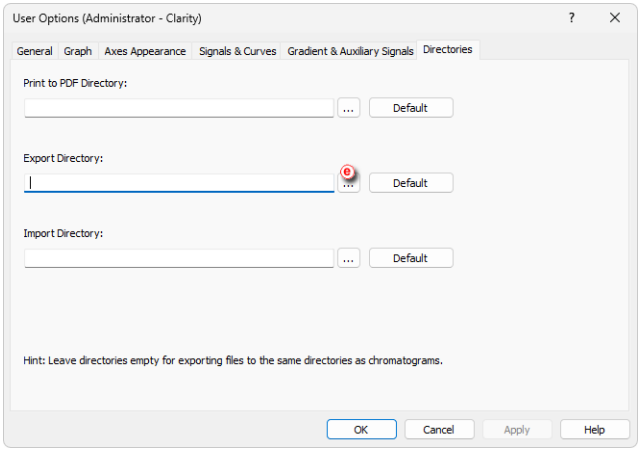
1. Open Clarity *Instrument* window and select - *Setting - Export Data...*.
2. In the *Export Data* dialog set the required *Export Content* ⓐ .

   Note: While exporting *Result Table* it is recommended to check *In Fixed Format*. Otherwise the content will change according to changes on screen. Beware that *In Fixed Format* is only applied to the *In Result Table*.
3. Select desired file formatting in *Text Format* section.
4. Select *Export to - Text File* ⓑ and choose preferred suffix.
5. Select *Full Format* option ⓒ . This will precede each result table row with a file name, date and time, thus allowing easy sorting after import.

6. Set *File Name* ⓓ and check *Append* option, in this way all results will be exported to a single file which can then be simply imported directly to your LIMS. Alternatively, leave the field empty in which case each export will be performed to a separate file. Exported files will have automatically generated file name from the chromatogram name.



- By default, files are exported to the same directory as the original chromatogram.
- To export to a single directory, specify it in the *User Options - Directories* tab ⓔ, accessible from the *Instrument* window by *Setting - User Options...* command.



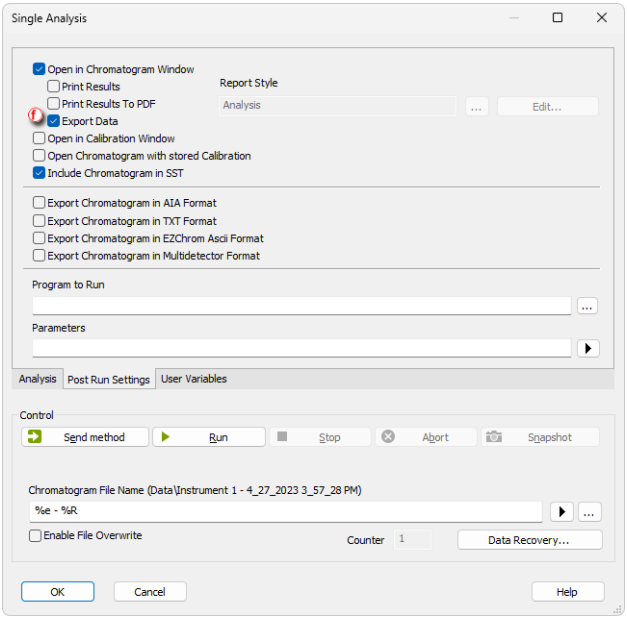
Locations to Export from

Three most common locations in exporting data to LIMS are:

Single Analysis

- In the *Post Run Settings* tab, of the *Single Analysis* dialog, select *Export Data* ⓕ .

Note: Export of data will be performed automatically after a single analysis is finished.



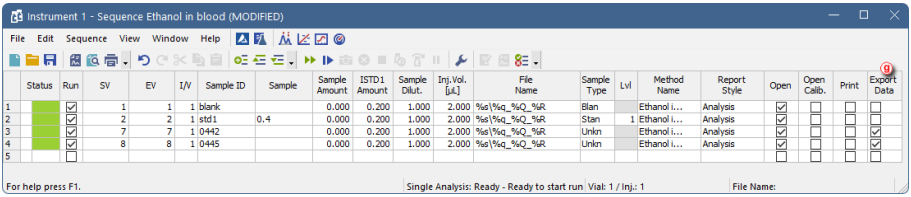
Sequence

- In the *Sequence* window, select checkbox in the *Export Data* ⓖ column for row(s) to be exported.

  Note: Export of data will be performed automatically after the row has been measured.

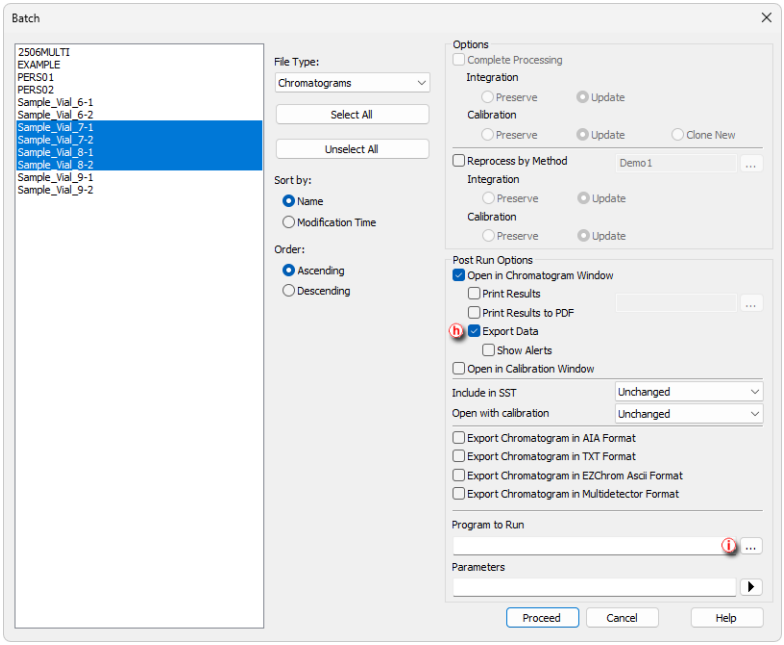
  More Info:

  - By default, *Export Data* column is hidden. Right mouse click in the sequence table and choose *Setup Columns...* to show it.
  - In the *Setup Columns* dialog, choose *Export Data* from the left list and click on the *Show* button - it will be added to the show list. Once you click on the *OK* button, you will return to the *Sequence* window and new *Export Data* column will be present.



Batch

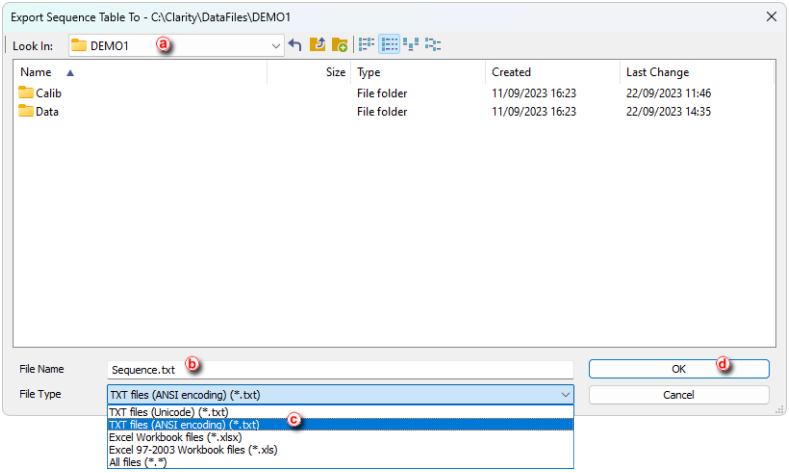
- Export multiple chromatograms at once using the *Batch* dialog, accessible from the *Instrument* window by *Analysis - Batch...* command.
- Select chromatograms to be exported and check the option to *Export Data* ⓗ .
- Clarity is able to start external program with a parameter ⓘ . Specify a program to run using the … and specify a command line parameter - typically the file name of the exported text file *%e*. Once ready to export the data, click the *Proceed* button.



13.5 Exporting sequence

It is possible to export sequence to other more common formats. Supported formats are *Excel Table* (*.XLSX and *.XLS suffixes) or *Text format* (*.TXT).

1. Open the sequence you want to export, then use the *File - Export…* command to open the *Export Sequence Table To* dialog.
2. Select the appropriate folder for saving the exported sequence ⓐ .
3. Fill in the name under which the exported sequence will be saved ⓑ .
4. Choose the format in which you want to export ⓒ . For Text format, the *ANSI* or *Unicode* encoding are available.
5. Press the *OK* button to perform the export ⓓ .

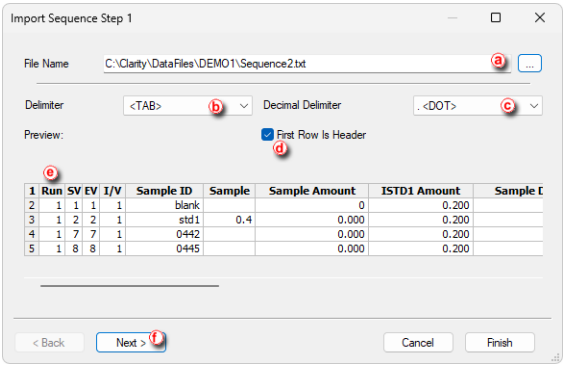


13.6 Importing sequence

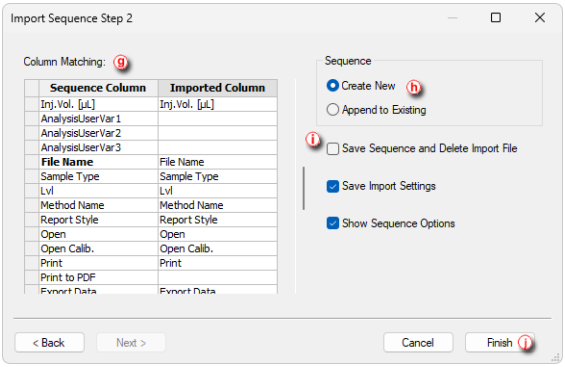
It is possible to import sequence that has been stored in a text file. Values have to be in delimited format and separated by an arbitrary delimiter.

Import of the sequence is one of the functions that enables an interconnection between Clarity and **LIMS** systems.

1. Open an Import dialog (*File - Import…*) from the *Sequence* window.
2. Choose the file which you want to import ⓐ . Supported formats are : .TXT, .CSV and .PRN
3. Select the character used as field delimiter ⓑ . Possible options are <*TAB*>, <*SPACE*>, <*COMMA*> or <*SEMICOLON*>.
4. Select the character used as decimal delimiter ⓒ . Options for the decimal delimiter are <*Window's Locale*> (the station will use the setting specified in the local settings of MS Windows), <*COMMA*>, <*DOT*> or <*SEMICOLON*>.
5. If the text file to be imported contains column descriptions in the first row, use *First Row Is Header* ⓓ checkbox. This row is used for mapping in the *Import Sequence Step 2* dialog.
6. Preview of the first five rows of the imported sequence table is displayed in the bottom part of the dialog ⓔ .
7. Click *Next* to continue ⓕ .



8. Set the mapping so that the columns used in Clarity (*Sequence Column*) will correspond to the columns from the imported file (*Imported Column*) ⓖ . Columns that are highlighted in bold letters are required. When the imported table contains column headers (the *First Row is Header* checkbox from the *Import Sequence Step 1* dialog has been checked), **Clarity** will attempt to automatically map the columns according to their names. Manual mapping will override the default name mapping. **Clarity** will store these names for future imports .
9. Select whether you want to save imported sequence as a new file or just append it to currently opened one ⓗ .
10. Three checkboxes allows you to do following ⓘ :
    - Original file will be erased after successful import.
    - Settings of this dialog will be saved for future imports.
    - Sequence Option will be shown immediately after finishing import.
11. Press the *Finish* button to accomplish import and close the dialog ⓙ .



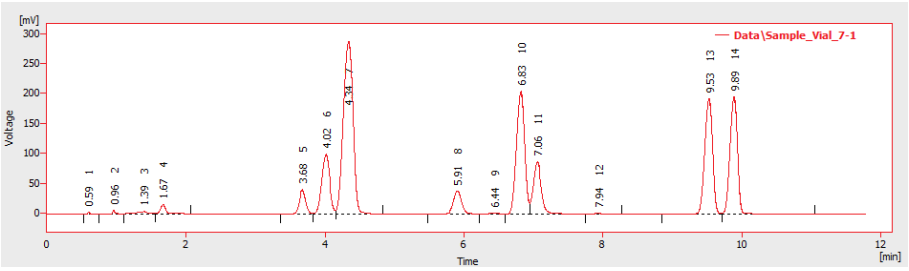
13.7 Exporting a chromatogram as a picture

You can export a chromatogram as a picture to the clipboard or as a file to a folder of your choice. The picture will include the labels and lines included in the chromatogram.

1. Open the *Chromatogram* window by selecting *Window - Chromatogram* from the *Instrument* window or click on .
2. Open the chromatogram you want to export.
3. Select *File - Export - Export as picture to clipboard* and paste the picture to MS Word, MS Powerpoint, Open Office Writer or any other suitable application of your choice.

   or

4. Select *File - Export - Export as picture to file...* and then select the folder where you would like to save the file in *Enhanced Metafile Format*.



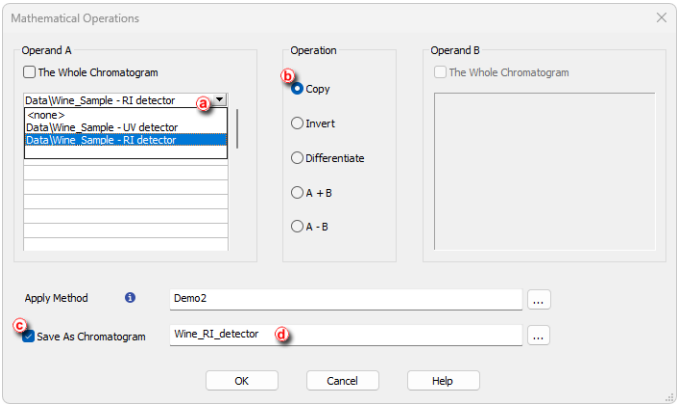
14 Mathematical Operations

This functionality was implemented into Clarity to give users a tool to manipulate already measured data without modifying original raw data. Mathematical operations present a very convenient approach to perform various actions such as extraction of a selected signal(s) from multiple signal chromatogram or subtraction of various chromatograms from each other. You can display three examples of applications of *Mathematical Operation* in following chapters. Many other applications may be developed by Clarity users on their own.

14.1 Extract chromatogram's signal using Mathematical Operations

You can save a particular signal from a chromatogram that contains several of them. This results in a stand-alone chromatogram file containing only the individual signal of choice, not all of the signals from the original chromatogram which might be confusing when working with a larger number of signals.

1. Open your a multi-signal chromatogram that you want to work with.
2. In *Chromatogram* window select *Chromatogram - Overlay - Mathematics* to open *Mathematical Operations* dialog.
3. From the drop-down menu ⓐ in the *Operand A* section select the desired signal of chromatogram you want to save separately. Select the *Copy* operation ⓑ , check *Save As Chromatogram* ⓒ , fill a name of the new chromatogram ⓓ and click *OK* to save the signal as standalone chromatogram.

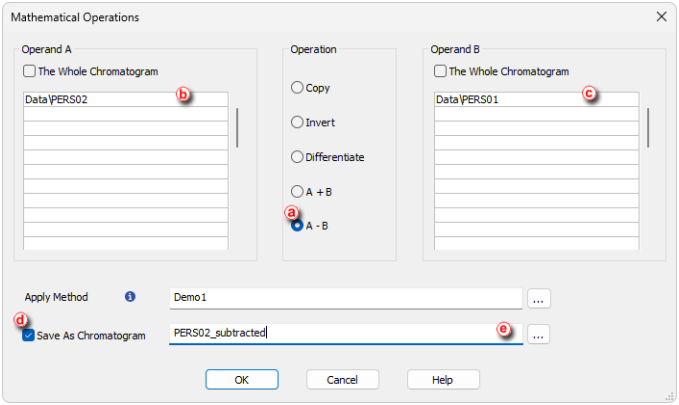


14.2 Subtraction of various chromatograms using Mathematical Operations

If you would like to use one of your chromatograms as a baseline for another chromatogram(s) there is a way to subtract the desired chromatogram from any

other chromatogram. It is possible to set *Subtraction Chromatogram* directly in *Chromatogram* window on *Measurement Conditions*, however, if you want to have both original and subtracted chromatograms as individual files you can use *Mathematical Operations*.

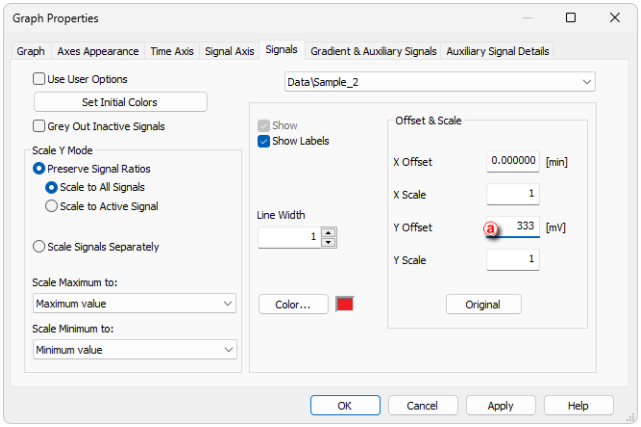
1. Open your chromatograms that you want to work with in *Overlay*.
2. In *Chromatogram* window select *Chromatogram - Overlay - Mathematics* to open *Mathematical Operations* dialog.
3. Select operation *A - B* ⓐ . From the drop-down menu in the *Operand A* section, select the chromatogram you want to subtract from ⓑ .
4. From the drop-down menu in the *Operand B* section, select the chromatogram you want to subtract ⓒ .
5. Check span *Save As Chromatogram* ⓓ , fill a name ⓔ and click *OK* to save the new subtracted chromatogram.



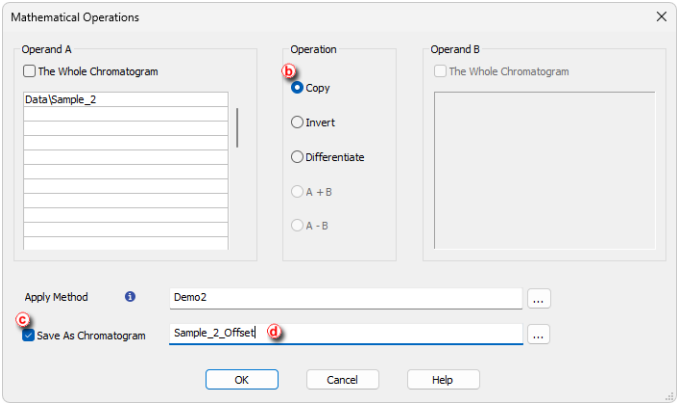
14.3 Copying of chromatogram using Mathematical Operations

Following chapter describes how to copy a modified chromatogram into a new one without changing the original chromatogram. For example we set up new Offset of Y-axis and this modification is about to saved in new chromatogram and want to keeping the original chromatogram unchanged.

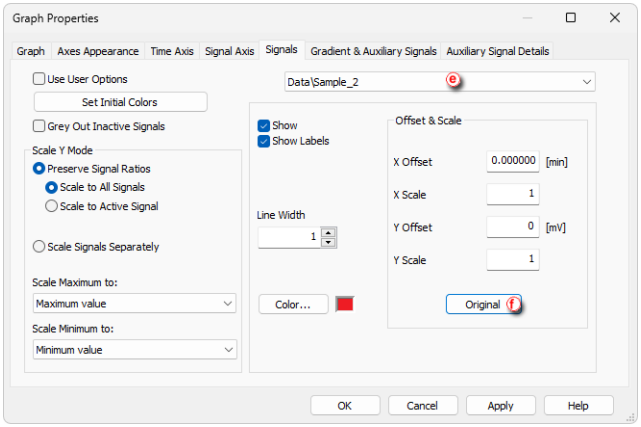
1. Open your chromatogram that you want to work with.
2. In *Chromatogram* window click *Display - Properties* to open *Graph Properties* and go to *Signals* tab.
3. Into Field *Y Offset* ⓐ write down value of desired offset in mV, in this example we choose 333 mV, and click *OK* button.



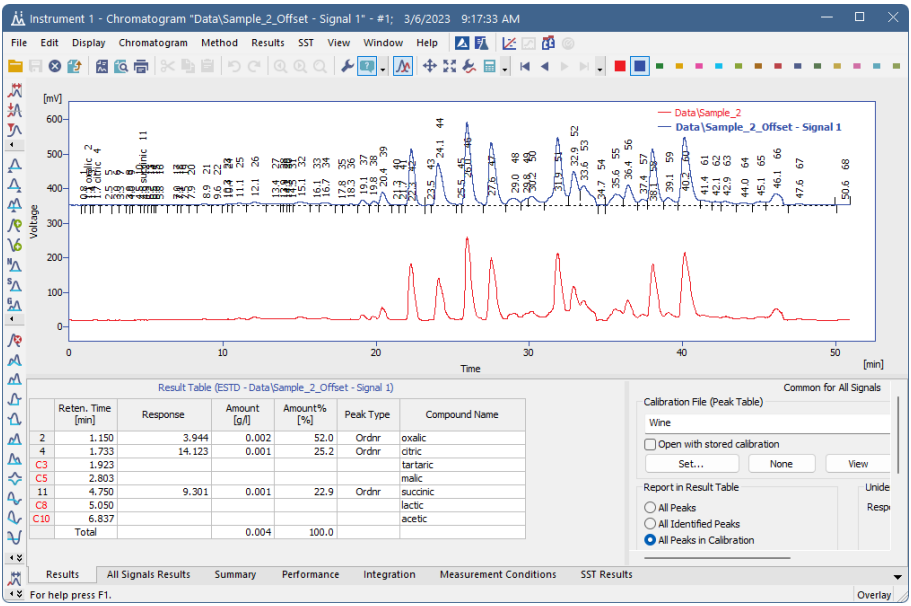
4. Select *Chromatogram - Overlay - Mathematics...* and the *Mathematical Operations* window will open.
5. Select *Copy* option ⓑ. Check *Save as Chromatogram* ⓒ, name the new chromatogram ⓓ and click the *OK* button.



6. Both chromatograms will be displayed in overlay mode.
7. Restore the original chromatogram by navigating to *Graph Properties - Signals* as in step 2.
8. Select the original chromatogram ⓔ, click *Original* button ⓕ and close the dialog by clicking *OK*.



9. Both chromatograms will be displayed in overlay mode as shown in the next image.



15 Archive and Restore

Clarity allows you to archive your PROJECTS (methods, sequences, measured chromatograms) and also the COMMON folder in which the print styles are stored.

15.1 Archiving a project (Creating an Archive)

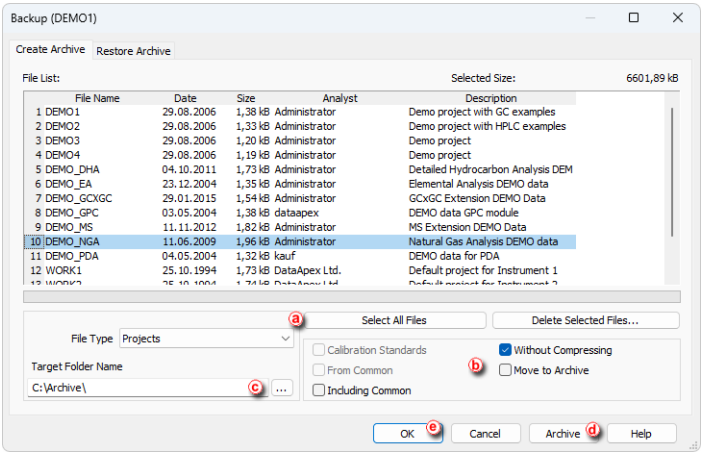
It is strongly recommended to archive the whole project folder after being shelved, but it is also possible to archive specific files only. An archive can be made by simply copying the files or by compressing them into one file (*.DGZ format).

1. Open the *Backup* dialog by selecting *Instrument- Archive...* on the *Instrument* window.
2. Select *Projects* in the *File Type* ⓐ option to archive a complete project directory.
3. Select the project or projects you wish to back up from the list. The *Select All Files* button ⓐ will select all projects.
4. Choose from the following options ⓑ :
    - Uncheck the *Without Compressing* option to archive all files into one compressed file.
    - Check the *Move to Archive* option to have the original files erased after backing them up.
    - Check the *Including Common* option to also back up the COMMON subdirectory.
5. Choose the output directory and name for the archive ⓒ . Compressed files will have the .DGZ extension.

    Caution: If you archive data in compressed format with a previously used name, you can possibly override the former archive and loose all data.

6. Click the *Archive* button ⓓ to back up the project or the *OK* button ⓔ , if you do not need to back up any more files.

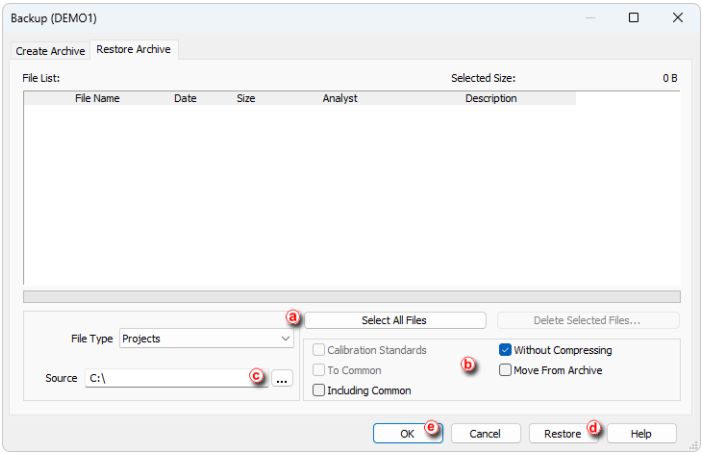
Note: Clicking *OK* will archive current selection.



15.2 Restoring a project from an archive

1. Open the *Backup* dialog by selecting *Instrument - Restore...* in the *Instrument* window.
2. Choose the source directory and select the source file ⓒ . Compressed files will have the .DGZ extension.
3. Select *Projects* in the File Type ⓐ option to restore a complete project directory.
4. Choose from the following options ⓑ :
   - Uncheck the *Without Compressing* option to restore all files from a compressed format.
   - Check the *Move from Archive* option to have the archived files erased after restoring them.
   - Check the *Including Common* option to also restore the COMMON subdirectory.
5. Select the project or projects you wish to restore from the list. The *Select All Files* button ⓐ will select all projects.
6. Click the *Restore button* ⓓ to restore the project or the ***OK*** button ⓔ if you do not need to restore any more files.

Note: Clicking *OK* will restore current selection.

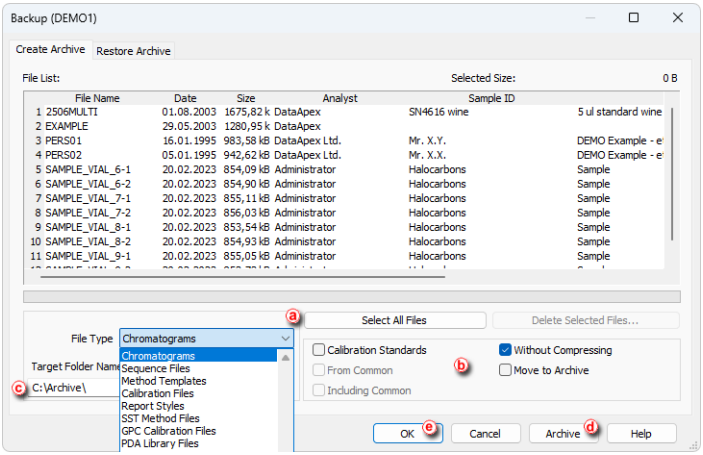


15.3 Archiving specific files (Creating an Archive)

It is strongly recommended to archive the whole project folder after being shelved but it is also possible to archive specific files only. An archive can be made by simply copying the files or by compressing them into one file (*.DGZ format).

1. Open the *Backup* dialog by choosing *Instrument - Archive...* in the *Instrument* window.
2. Select the *File Type* ⓐ option according to the files you wish to archive.
3. Select the files you wish to back up from the list. The *Select All Files* button ⓐ will select them all.
4. Choose from the following options ⓑ :
   - Uncheck the *Without Compressing* option to archive all files into one compressed file.
   - Check the *Move to Archive* option to have the original files erased after backing them up.
   - Check the *Calibration Standards* option when archiving chromatograms. The chromatogram files from the CALIB subdirectory (instead of the DATA subdirectory) should be listed.
   - Check the *From Common* option to display the system files from the COMMON directory.
5. Choose the output directory and name for the archive ⓒ . Compressed files will have the .DGZ extension.
6. Click the *Archive* button ⓓ to back up the file or the *OK* button ⓔ if you do not need to back up any more files.

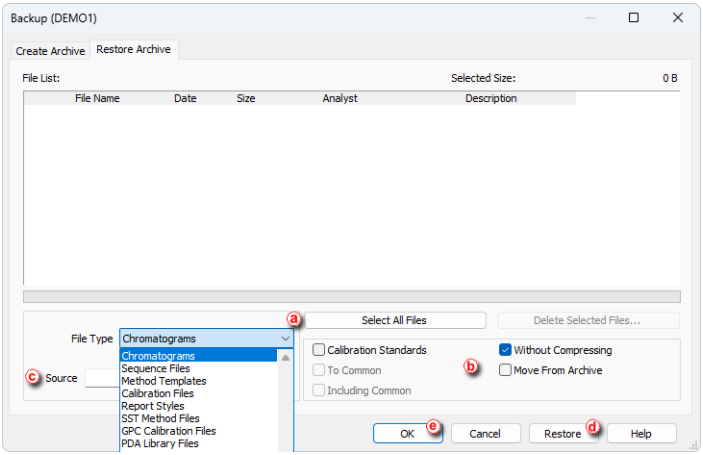
Note: Clicking *OK* will archive current selection.



15.4 Restoring a file from an archive

1. Open the *Backup* dialog by choosing *Instrument - Restore...* on the *Instrument* window.
2. Choose the source directory and select the source file ⓒ .
3. Select the *File Type* ⓐ option according to the files you wish to restore.
4. Choose from the following options ⓑ **:**
   - Uncheck the *Without Compressing* option to restore files from a compressed format.
   - Check the *Move from Archive* option to have the archive files erased after restoring them.
   - Check the *Calibration Standards* option when restoring chromatograms to the CALIB subdirectory (instead of the DATA subdirectory).
5. Select the files you wish to restore from the list. The *Select All Files* button ⓐ will select them all.
6. Click the *Restore* button ⓓ to restore the files or the *OK* button ⓔ if you do not need to restore any more files.

Note: Clicking *OK* will restore current selection.


Backup (DEMO1)
Create Archive
Restore Archive
File List:
Selected Size:
0 B
File Name
Date
Size
Analyst
Description
a
Select All Files
Delete Selected Files...
File Type
Chromatograms
Chromatograms
Sequence Files
Method Templates
Calibration Files
Report Styles
SST Method Files
GPC Calibration Files
PDA Library Files
c
Source
Calibration Standards
To Common
Including Common
b
Without Compressing
Move From Archive
OK
e
Cancel
Restore
d
Help

16 Managing the Chromatography Station

Following chapters contains extended information how to set restrict access to various parts of Clarity to certain users or set up communication with a mobile application.

16.1 Enabling instruments to be used by Clarity2Go application

Clarity enables to send specific parameters over the internet to be monitored via **Clarity2Go** application. In this way you can monitor your analyses while outside the laboratory. Note that both PC with Clarity and smartphone must be connected to internet. For more information regarding Clarity2Go installation refer to www.dataapex.com/product/clarity2go.

The whole solution consists of three independent parts:

- **Clarity** station (at your side) - sends information about state and running analyses to the server.
- **Server** (at **DataApex's** side) - dispatches the information from **Clarity** stations to **Clarity2Go** clients.
- **Client** (at your side) - device (smartphone or tablet) with installed **Clarity2Go** application processes information from the server.

DataApex is providing a free public server for this use.

How to set up Clarity:

1. In the *Clarity* main window, go to *System* menu and click on the command *Clarity2Go...*.
2. In the opened *Clarity2Go Configuration* dialog, check the checkbox of *Instrument* 1 to 4, depending on the instruments you want to monitor. Every checked Instrument will get its unique *Instrument ID*.
3. Click the *OK* button to save the configuration and continue with steps described in **How to set up Clarity2Go** section or refer to *Advanced* options bellow.

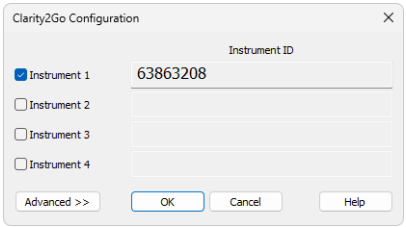


Fig. 4: Clarity2Go Configuration - Basic

Options described below are optional and are not obligatory for correct functionality. They are revealed by clicking the *Advanced* button.

- *Unregister All Instruments* - disables the monitoring in the **Clarity2Go** application. Instruments that have been registered will no longer be available for monitoring. If you will later change your mind, you will have to generate new *Instrument ID*.
- *Web Server Address* - do **not** change this field. It defines the address of **Clarity2Go** web server. Address other than default will result in the monitoring to be not functional! Press the *Default* button to set functional web address of the server.
- *Proxy Server Address* - consult with your local administrator if a proxy server is applied in your local network and then provide the proxy server address.
- *Protect by Password* - provided password will be valid for **all** Instruments. The same password needs to be provided in the **Clarity2Go** application to unlock the monitoring.
- Click the *OK* button to save the configuration and continue with steps described in **How to set up Clarity2Go**.

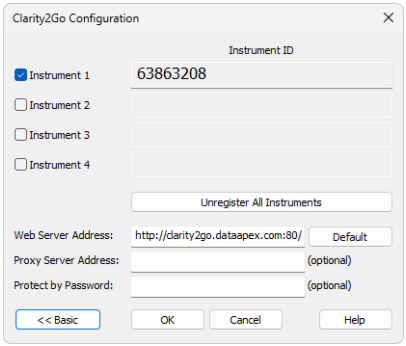


Fig. 5: Clarity2Go Configuration - Advanced

How to set up Clarity2Go:

Once you have configured instrument(s) in **Clarity**, it's time to monitor those instruments using **Clarity2Go** application. This part assumes that you have **Clarity2Go** for Android application installed and running.

1. Make sure that you are not in the **Clarity Demo** mode, indicated by a gray stripe at the bottom of the application with the inscription **DEMO mode**. Tap on the *TURN OFF DEMO*.

Note: **Clarity Demo** mode does not allow to add instruments.

*Fig. 6: **Clarity Demo** mode gray stripe*

2. In the *Settings*, tap on *Demo - Switch to demo mode* which turns the **Clarity Demo** mode *OFF*.
3. Return back. In case you are configuring **Clarity2Go** for the first time, you will see that there is no instrument.

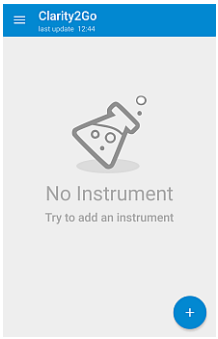


Fig. 7: No Instrument screen

4. Tap on the blue "plus" button at the bottom right corner to add a new instrument.
5. Enter *Instrument ID* that has been generated by **Clarity** and enter password (only if you have set it up in **Clarity**). Tap on the *OK* button to start monitoring this instrument.

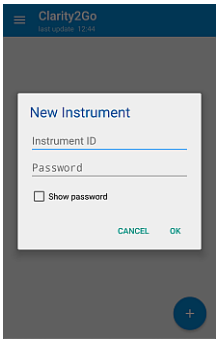


Fig. 8: New Instrument screen

6. The newly configured instrument will be added to the list of instruments that are being monitored.

Note: You can invoke the application menu by tapping on the 3 horizontal lines - the menu contains *Settings*, built-in *Help* and *About* options.

16.2 Locking/Auto Locking a Clarity Instrument

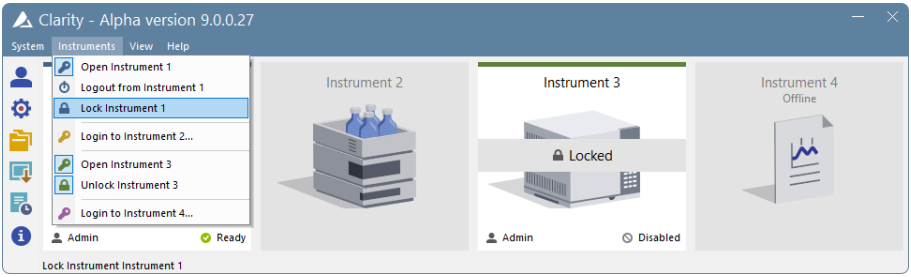
You may lock a Clarity Instrument protected by password if you want to prevent unauthorized access to it, for example, when an analysis is running.

Manual Lock

- Instrument can be manually locked either from *Instrument* window by clicking *Instrument - Lock Instrument_Name*.

  or

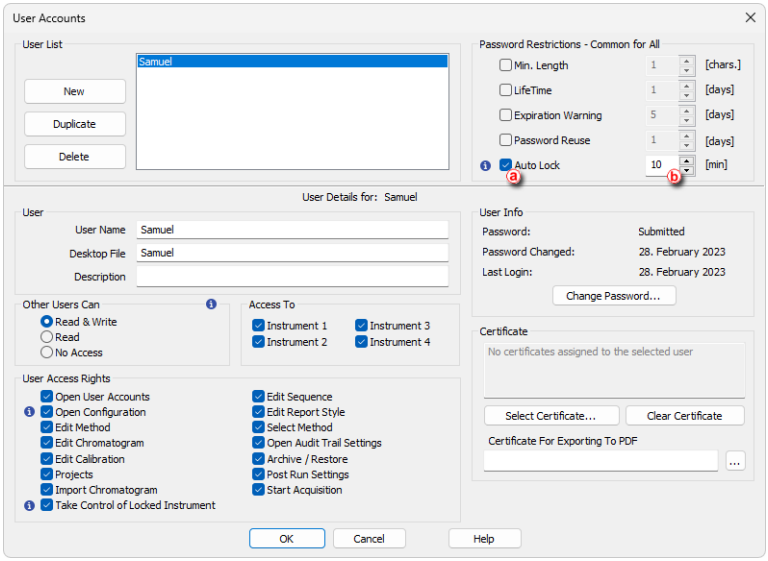
- From the *Main Station* window by clicking *Instruments - Lock Instrument_Name*.



Auto Lock

It is possible to set the automatic lock function so that all opened instruments will be locked after a period of inactivity.

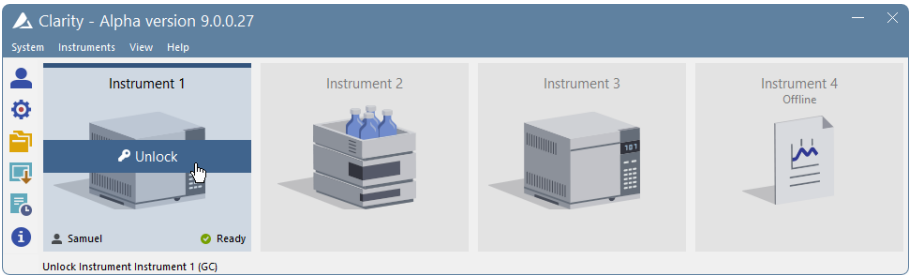
1. Open the *User Accounts* window by clicking on or choose *System - User Accounts*.
2. Check the *Auto Lock* function ⓐ .
3. Set the period of inactivity in minutes after which all opened *Instruments* will be locked ⓑ .



Caution: When an Instrument is Auto Locked, all eventual unsaved changes in modal dialogs (Method Setup, Single Analysis, Report Setup, etc) are discarded and those dialogs are closed.

16.3 Unlocking a Clarity Instrument

1. Select *Instruments - Unlock Instrument 1* from the *Main Clarity* window or click on the *Instrument* image.
2. Enter the password and click OK.



Unlocking Instrument after entering wrong password three or more times:

If you enter the wrong password three times or more, then a message will appear asking you to restart the program.

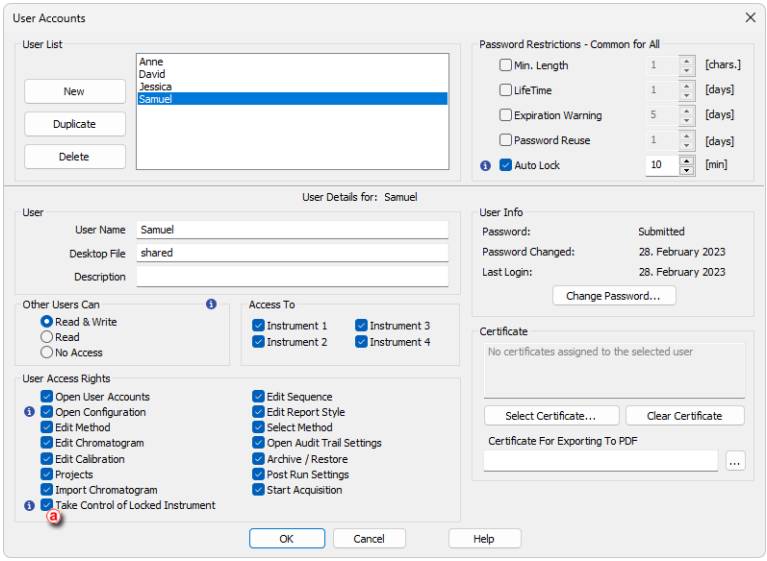
1. Unlock the *Instrument* using credentials for a user with administrator rights over the locked *Instrument* according to chapter *Taking control over locked instrument*.
2. Restart Clarity. *Instrument* can again be opened by the previous user.

16.4 Taking control over Clarity Instrument

You may take control of Instrument where different user is already logged. To be able to do that the option *Take Control of Locked Instrument* has to be enabled in the *User Accounts* settings for the user who wants to take over.

Setting up the User Right

1. Open the *User Accounts* window by clicking on or choosing *System - User Accounts*.
2. Check the *Take Control of Locked Instrument* in *User Access Rights* ⓐ part of the dialog for users who should be able to do so.



Note: It is recommended to set one common *Desktop File* for all users with this option enabled. Otherwise desktop of the first logged user will be used for all users that take control over the Instrument, until the Instrument is closed again.

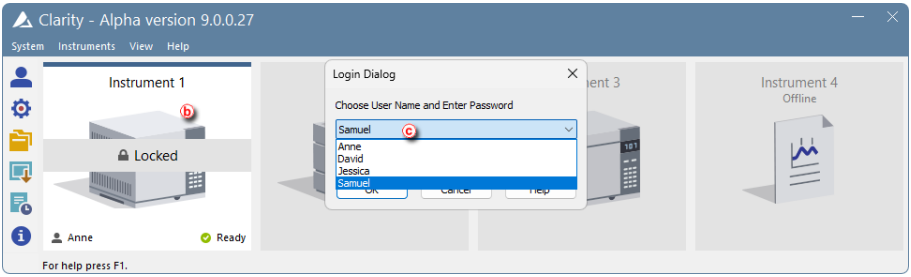
Note: It is also recommended to set the same *Edit...* rights for users that plan to take over Instruments as different rights may limit the options of second user.

Note: It is possible to set the *Auto Lock* function so that all opened instruments will be locked after a period of inactivity.

Taking control of Locked Instrument

Steps are similar to the standard logging to instrument.

1. Select *Instruments - Unlock Instrument 1* from the *Main Clarity* window or click on the *Instrument* image ⓑ .
2. Choose your User Name ⓒ .
3. Enter the password and click OK.



Caution: All unsaved changes in files made by the previous user will be discarded.

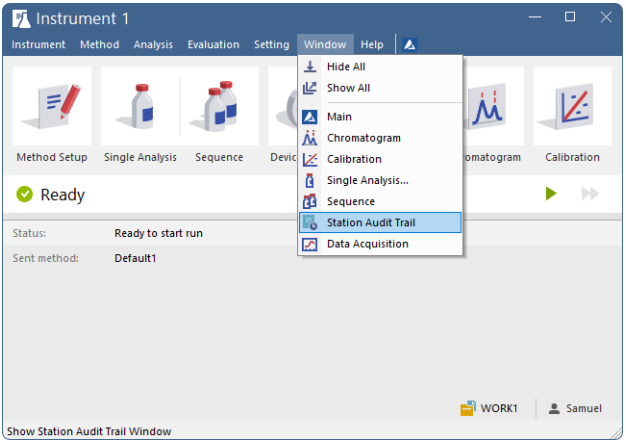
Note: It is possible to take control of locked Instrument using the Command line parameters. Always include the user *(user=…)* and password *(p=…)* parameters for each command, because the instrument is kept locked.

16.5 Monitoring Events and Operations in Clarity

The ***Audit Trail*** can be used for finding out who did what and when, including file operations, changes to settings, events that take place during data acquisition and system messages. Therefore this is an essential tool for troubleshooting and managing Clarity.

To access the ***Station Audit Trail***:

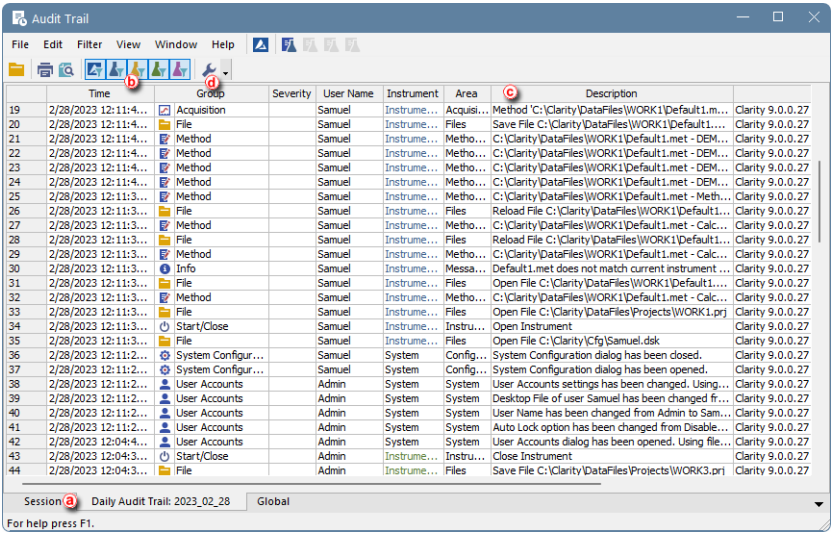
1. Select *Window - Station Audit Trail* from the *Instrument, Chromatogram, Calibration, Sequence* or *Data Acquisition* windows. Alternatively click icon in the *Main Station* window.



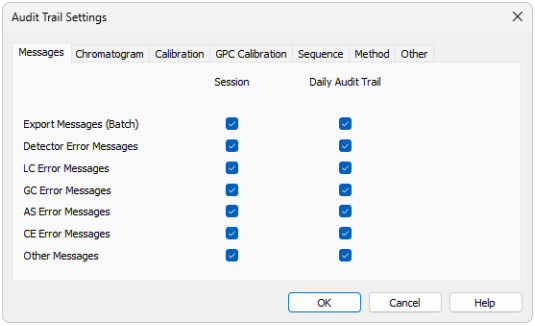
2. Click on the *Session* tab ⓐ if you would like to see the log from the time Clarity was started or on the *Daily Audit Trial* tab for the present day events.

   Note: The *Daily Station Audit Trail* is stored in one separate file every day the station is running. These files can be opened by using the command *File - Open Audit Trail (Append)*.

3. Click on the *Instruments* or *System* icons ⓑ to filter out events and operations based on instrument where they occurred.
4. Inspect the *Description* column to find out about operations and events that have taken place ⓒ .



5. Click on the *Properties* icon ⓓ to set up which events and operations should be recorded in the *Session* or *Daily Audit Trail*.



To access the file-specific *Local Audit Trail* (Chromatogram, Calibration, Sequence and Method):

1. Select *Window - Chromatogram, Calibration or Sequence Audit Trail* from the corresponding window or click on the *Audit Trail* icon in the *Method Setup* window to open corresponding *Audit Trail*.

Note: The *Local Audit Trails* are included in the *Chromatogram, Calibration, Sequence* and *Method* files and contain the whole file history.

16.6 Controlling Clarity from an external application

It is possible to send commands to Clarity using Windows command line parameters and also to read its status through Windows Dynamic Data Exchange (DDE).

- For more information on the commands go to our List of commands in the **Clarity Reference Guide**.
- You can also find the list of variables which will give you information on Clarity status on our DDE datasheet.

17 Clarity in Network

Clarity might be used in network and following chapters describe different approaches of such usage.

17.1 Clarity in network overview

Clarity is not a client-server (C/S) solution, nonetheless it can be configured for use in multi-user and multi-instrument networked environment.

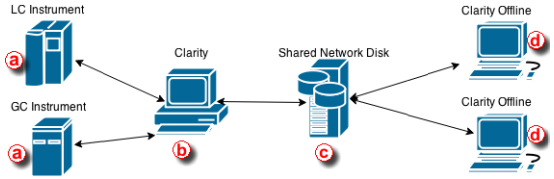
What does the solution Clarity in network offer?

- Instrument control, real-time signal monitoring and run control is possible only trough the local Clarity station, i.e. Clarity must be connected to respective chromatography instrument. Each Clarity needs to be set locally (including .cfg and .psw).
- Using the *System - Directories*, the location of the Clarity data/projects can be set anywhere within the network (typically on a shared server back upped drive). The data can be then accessed from any Clarity station on the network, including the Clarity Offline stations intended for evaluation of data from another computers.
- Clarity Offline allows users to prepare methods and evaluate acquired data.
- With Clarity Offline users are able to work with acquired data on additional computers in the laboratory or at home.
- File access conflicts may occur when accessing the same file (e.g. method) from different Clarity stations.

What does the solution Clarity in network not offer?

- Central management of users.
- Central management of documents such as chromatograms, calibrations and methods.
- Direct control of acquisition, i.e. run/stop/abort from other Clarity stations.
- Direct control of instruments from other Clarity stations.
- Watch real time signal being acquired by detectors from other Clarity stations.

17.2 Multiple Clarity stations in a network



More Info:

- Note that it is possible to have more than one Clarity ⓑ in the network.
- Shared Network Disk ⓒ can be on the same PC as well and not as a separate unit as seen in the picture above.

Clarity in network is a solution that consists of at least one Clarity, at least one Clarity Offline and a reliable computer network as the most basic setup.

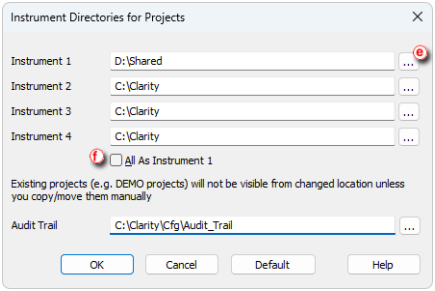
Simplified scheme of the possible configuration is displayed in the diagram above. LC and GC instruments ⓐ are controlled via Clarity ⓑ. Chromatograms, calibrations and methods are all saved (using directory configuration) on a shared network disk ⓒ. Clarity Offline ⓓ could then be used for evaluation of acquired chromatograms and preparation of methods which are saved (using directory configuration) on the shared network disk. Clarity ⓑ is able to send those prepared methods to corresponding instruments.

Note that this shared network disk is accessible to all computers within this computer network therefore a much wider configuration can be implemented than the one described above.

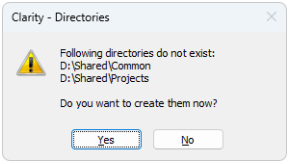
Following step-by-step guide will help you configure the Clarity in network solution.

Procedure A - Firstly we will configure Clarity ⓑ which will acquire data and save them to the shared network disk.

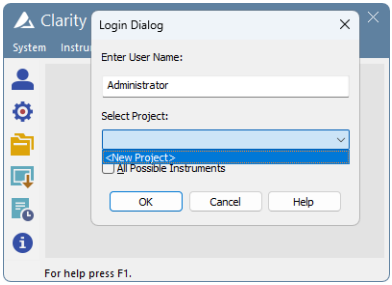
1. In the main *Clarity* window, go to *System - Directories* or use the icon .
2. *Instrument directories for Projects* dialog will open.
3. Choose Instrument that will share all the created files; chromatograms, calibrations, methods, sequences, reports etc..
4. Use the ... ⓔ to browse for shared network disk. Once you locate it, click *OK*. Path to the shared network disk is now filled in the corresponding Instrument.
5. In case you want all Instruments to have the same directory, check the option *All As Instrument 1* ⓕ. This will copy the directory path for the rest of the instruments as it is set for *Instrument 1*.



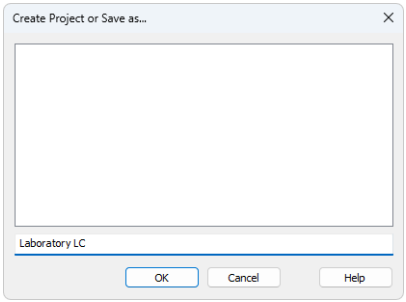
6. To save the configuration click the *OK* button.
7. Configuring the directory for the first time will result in the following message. Click *Yes* to allow the creation of necessary structure. Upon clicking *Yes*, COMMON and PROJECTS folders are created with default documents necessary for correct functionality.



8. When you try to login for the first time with the new directory configuration, you will be asked to create a new project.



9. Fill project name and click *OK*.

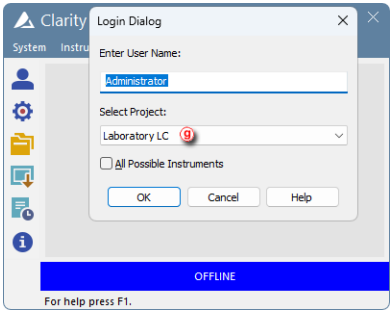


Anything created/measured within this project is saved on the shared network disk and therefore is accessible to other Clarity stations.

Procedure B - Secondly we will configure Clarity Offline ⓓ which will be used for data evaluation from shared network disk.

We need to configure directory from which Clarity Offline will open chromatograms, calibrations and methods saved on the shared network disk.

1. Configure the directory according to step *1-6*, described above.
2. When you login, select the appropriate project (the one you filled in step 9) using the drop-down box ⓖ and click *OK*.



If you have followed the steps correctly, your Clarity in network is configured. If you are not sure, you can test it by measuring some chromatogram and evaluate it on Clarity Offline. Once you see measured chromatogram in the Data directory you know it has been configured correctly.

17.2.1 Migrating Clarity Project into a Network

This step by step guide will help you to move your Clarity project with measured chromatograms, calibrations, prepared methods and other files into a shared network disk or a shared server back upped drive.

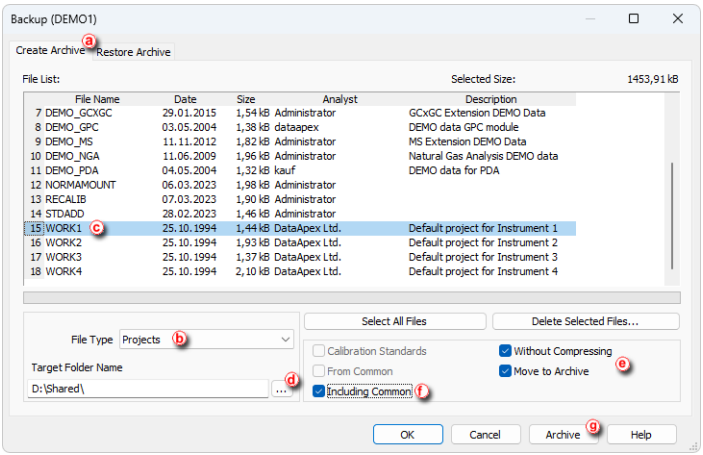
This guide assumes that you have already set up directory for Clarity in network therefore you have all the necessary structure prepared - if not, refer to **Procedure A** described in the chapter **"Multiple Clarity stations in a network"** on pg. **211**.

Principle behind migrating Clarity project into a different location is straightforward. It is necessary to move the whole project directory (e.g. WORK1) as well as the project file itself (e.g. WORK1.PRJ).

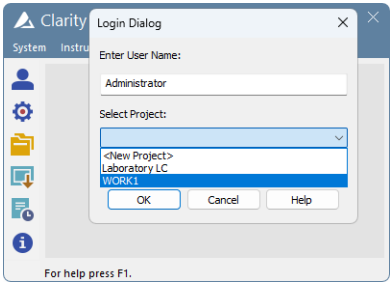
More Info:

- Note that it is possible to move the project directory and the project file also using *File Explorer* when Clarity is turned off.

1. Switch *Directories* back to the previous (default) location, so you can see previously used projects.
2. Log to the project you don't want to migrate, e.g. one of the demo projects.
3. In the *Instrument* window, go to menu *File - Archive...*.
4. *Backup* dialog opens on the *Create Archive* tab ⓐ .
5. Because we want to migrate whole project, change *File Type* using the drop down list ⓑ to *Projects*.
6. Section *File List* now contains all projects in Clarity. Click the project to be migrated (e.g. WORK1) ⓒ .
7. As a *Target* select the destination of the shared network disk using the ... at ⓓ .
8. Check the options *Without Compressing* and *Move to Archive* ⓔ .
9. Check also *Including Common*, if you use customized report style, template sequence and other files stored in Common folder ⓕ (you will be prompted to confirm overwriting the default files created during **Procedure A**).
10. When everything is set as described above, you can click the *Archive* ⓖ button which migrates project WORK1 to the D:\SHARED\.

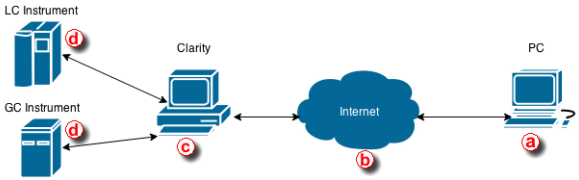


11. Now change the directories back to the network shared drive.
12. *Login* to the Instrument which has set directory in the network. Notice that in your Clarity *Login Dialog* the *Select Project* drop down list offers your migrated project (WORK1) and project created after setting the new directory only.



13. Your Clarity project has been successfully migrated and you can start working.

17.3 Using remote desktop to control Clarity



This step by step guide will help you to connect remotely to a PC with Clarity ⓒ installed from your home or office ⓐ . It will allow you to connect remotely to that PC and take control over the whole computer and thus control Clarity and connected instruments. The connection is realized over the internet or a local computer network. Connection over the local computer network is more secure since the transferred data never enter the internet.

Requirements:

- ⓐ PC needs to have Remote Desktop Connection installed
- ⓑ Internet connection or a reliable computer network
- ⓒ PC with Clarity needs to have Remote Desktop Connection installed

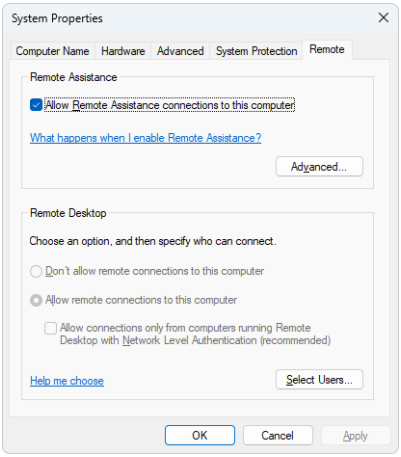
More Info:

- Remote Desktop Connection is a standard application installed in Windows operating systems.

This description is for setting up a remote connection in Windows 11. Dialogs may vary depending upon the Windows version used.

1. To allow remote connections on the PC with Clarity ⓒ you want to connect follow the steps below:
    - Go to the Control Panel - System and Security - System - Allow Remote Access.
    - If you're prompted for an administrator password or confirmation, type the password or provide confirmation.

- Under Remote Desktop, select one of the three options.
- Click Select Users.
- In the Remote Desktop Users dialog box, click Add.
- In the Select Users or Groups dialog box, do the following:
    - To specify the search location, click Locations and then select the location you want to search.
    - In Enter the object names to select, type the name of the user that you want to add and then click Check Names. This will check whether the user exists. If not, it will trigger a not found dialog. Check the name once again. Note that this user must have a profile on this computer. If the user name is correct, click OK.
    - The name will be displayed in the list of users in the Remote Desktop Users dialog box. Click OK and then click OK again.
- In case Remote Desktop options are grayed out your computer is probably in a domain and due to domain policies you may not be able to change the settings. Contact your network administrator to resolve the situation.



2. Look up the name of the remote computer ⓒ . You will to provide this information in step 5.
    - Go to System - About.
    - There is the Device Name, which is the info you need.
    - Alternatively your network administrator might also be able to give you the name of the computer.
3. Set a password for your user account on PC with Clarity ⓒ . Your user account must be password protected before you can use Remote Desktop to connect to

another computer. Password protection is now default setting in Windows, so you may have that already set up.

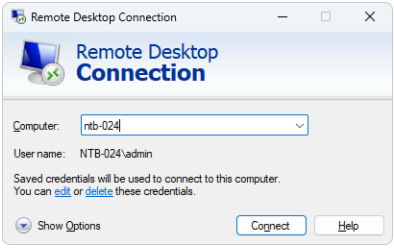
- Go to the User Accounts - Sign In Options.

4. To allow Remote Desktop connections through a Windows Firewall on the remote PC ⓒ follow the steps below:

   If you're having trouble connecting, Remote Desktop connections might be getting blocked by the firewall. Here's how to change that setting on a PC. If you're using another firewall, make sure the port for Remote Desktop (usually 3389) is open.

   - Go to the Control Panel.
   - Click System and Security.
   - Click Allow an app through Windows Firewall under Windows Defender Firewall section.
   - Click Change settings and then check the box next to Remote Desktop.
   - Click OK to save the changes.

5. Start Remote Desktop from the computer you want to work from ⓐ .
   - Open Remote Desktop Connection.
   - In the Computer box, type the name of the computer that you want to connect to, and then click Connect. (You can also type the IP address instead of the computer name.)
   - Note that the remote PC cannot be in sleep mode or hibernating.



More Info:

- This text has been taken from the How-to: "Connect to another computer using Remote Desktop Connection" created by Microsoft Windows.

Once you successfully connect to the remote PC you can work as if you were sitting in the lab and working with Clarity. The remote desktop will be presented in the normal window. To terminate the session, close the window.

This solution then enables you to:

- Control instruments that are directly connected to Clarity.
- Monitor data acquisition.
- Evaluate chromatograms in Clarity.
- Work on other projects and leave the remote session open and check once in a while if everything is running smoothly.

Possible situations that may arise using the Remote Desktop Connection:

- If you remotely connect to a PC where you are currently logged in, you will be automatically put through and you can start working.
- However, if you try to connect to a PC when there is logged in someone else, e.g. another analyst, he will be asked if he allows the remote connection to put through. If he declines the remote connection you will not be able to connect.
- PC that you are connecting to must be turned on, it is not possible to connect to a PC that is off or in sleep mode.

18 Utilities

Clarity installation contains various utilities for validating the installation or predefining various Clarity profiles which helps you in using more configurations of laboratory instruments.

18.1 Checking that the software has been installed correctly (Installation Qualification - IQ)

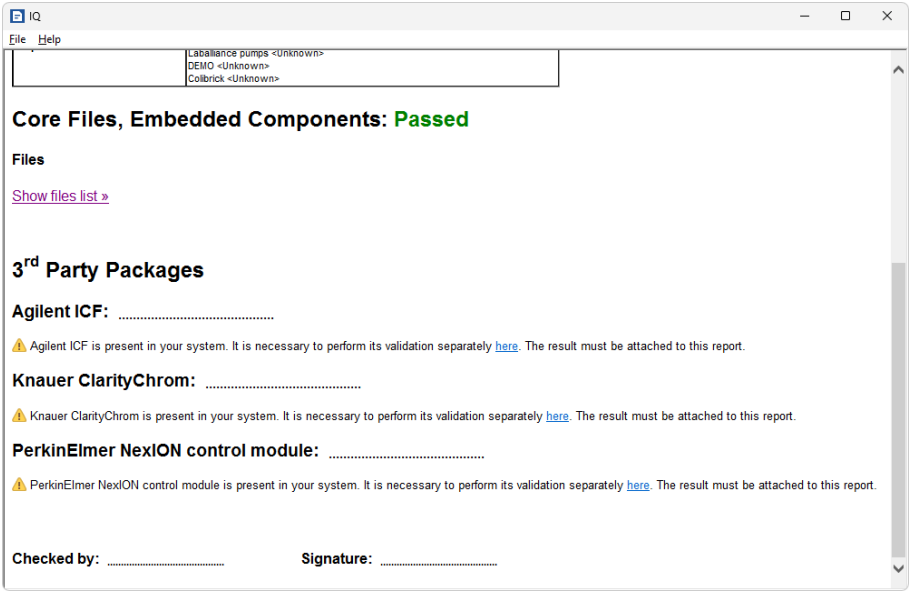
The *Installation Qualification* (**IQ**) is a procedure confirming that the software has been installed successfully and that correct versions of files are present.

1. Install the Clarity station according to the instructions of the *Installation Wizard*.
2. After the installation has been completed, you can open *IQ* by searching for  *IQ Report* in the search field of the Start menu. Alternatively you can start *IQ* by using C:\CLARITY\BIN\IQ.EXE.
3. If the installation has been correctly performed, the status should read: "*Installation Qualification Test: Passed*".
4. If the Installation Qualification fails, it is recommended to uninstall and then re-install Clarity. If it fails again, contact DataApex support.

   Note: You can use *Show files list* to display list of all validated files and search for the ones that are causing issues.

5. The *Installation Qualification* report can then be printed, copied to the Windows Clipboard or sent as an email.

Caution: Some driver packages have standalone IQ that must be performed separately by clicking *here* in given section. Clarity IQ is **NOT** valid unless IQ of all components passed and reports are stored together.

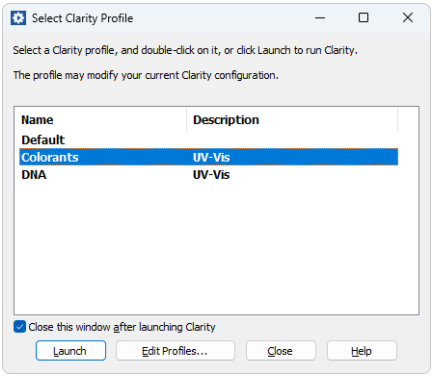


Note: The most common reason for a "Failed" result is the installation of an updated version over an existing installation of Clarity. This itself does not produce any errors but since some of the files are preserved from the original installation, the checksums will not match.

18.2 Editing Clarity profiles for different analyses using the Launch Manager

Launch Manager is a utility that allows you to start the Chromatography Station with specific setting. The collection of these settings is saved as a *Profile* in *Launch Manager*. You can select specific configuration, desktop file containing custom calculations and which instrument and project specifc user will be asked to log in.

1. Start the *Launch Manager* either from the Windows Start menu or using C:\CLARITY\BIN\LAUNCHMANAGER.EXE.
2. Click on the *Edit Profiles* button to create or modify the profiles.



3. Select a profile ⓐ to modify it or click on the *New* button to create a new profile ⓑ .
4. If you created a new profile, fill its name. For easier orientation it is recommended to fill in the *Description* which eases the distinguishing of different profiles.
5. Select the configuration file that will be loaded after the start of Clarity ⓒ .

   Note: The list of configuration files is retrieved from the installation directory (configuration files are located in C:\CLARITY\CFG by default). Clicking on » button opens menu for managing the configuration files, see Creating a duplicate configuration using the Launch Manager for one example of usage.

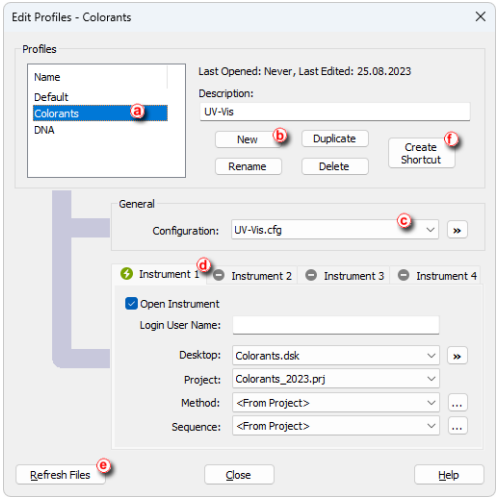
6. If you select the <Last Used> option, Clarity will start with the last configuration it was opened with or if Clarity is running, the present configuration will be preserved.
7. For each Instrument, choose if it should be opened at the Clarity start by *Open Instrument* checkbox. Select the User, Desktop, Project, Method and Sequence files that will be loaded when opening instruments.

   Note: In the desktop file, all layout setting is saved (the width of the table columns, the custom toolbars), but also custom calculations created in *User Columns*.

   Note: If <From Project> is selected, the Method and Sequence that were last opened in selected Project will be used.

8. You can use *Refresh Files* button ⓔ to reload all the files displayed in the *Edit Profiles* dialog.
9. Once the profile is configured, *Close* the *Edit Profiles* dialog and *Launch* the profile either by double clicking on the profile or by selecting a profile and clicking on the *Launch*.

Note: You can use *Create Shortcut* ⓕ to place shortcut on the desktop which can be directly used to launch given profile.

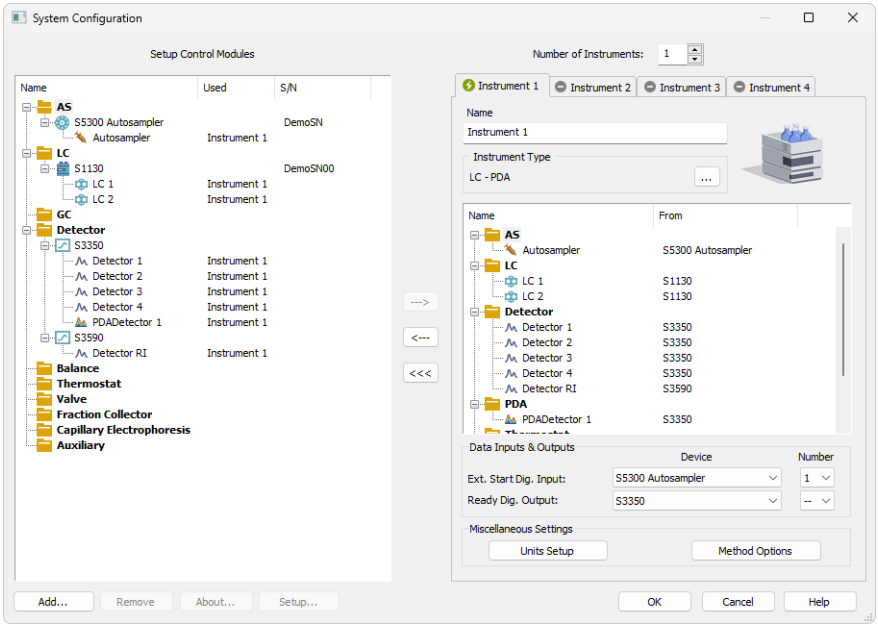


18.3 Creating a duplicate configuration using the Launch Manager

This topic describes how to create a duplicate configuration using the *Launch Manager*. This becomes especially handy when you have system with two detectors (e.g., RI + DAD) but you don't need to run them together every time.

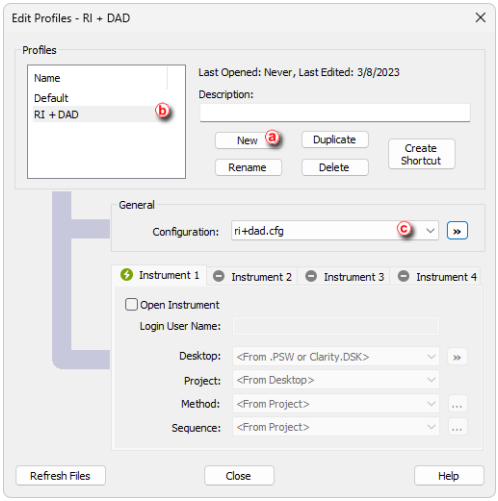
1. If you don't have any configuration yet, start Clarity and configure your device(s) according to the chapter "Adding a new device" and close Clarity. Otherwise start with the next step.

   Note: In this example we start with LC system with both DAD and RI detectors configured.

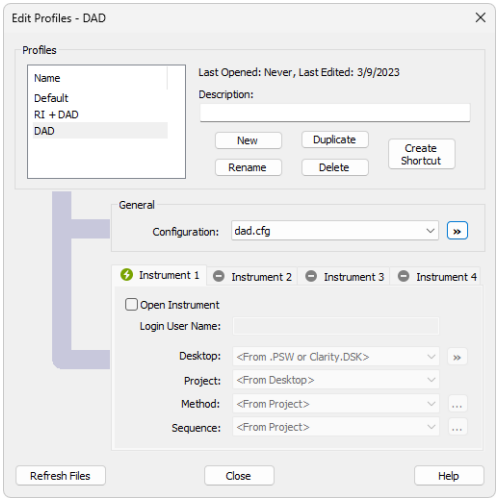


2. Open *Launch Manager* either from the Windows Start menu or by using C:\CLARITY\BIN\LAUNCHMANAGER.EXE.
3. In the *Select Clarity Profile* dialog click *Edit Profiles...*.
4. In the *Edit Profiles* dialog, click the *New* button ⓐ to create a new profile and name your profile (e.g. *RI + DAD*). Click *OK* to save the profile. Newly created profile is displayed in the small table on left ⓑ .
5. From the drop-down list ⓒ , select CLARITY.CFG, click the » button, from the selection choose *Duplicate* and in the following dialog name your configuration (e.g. ri+dad.cfg).

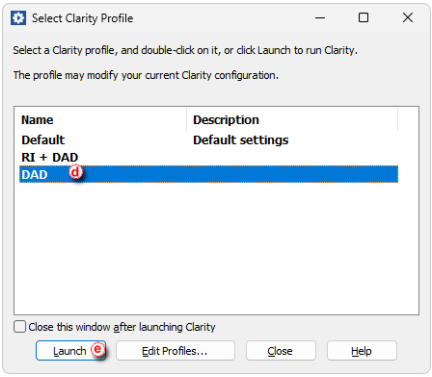
Note: This step will create a duplicate configuration (.cfg file) based on the current Clarity setup (as described in step 1.). It prevents Clarity from overwriting a default configuration.



6. Create a new profile (DAD) with a different configuration (duplicated ri+dad.cfg named as dad.cfg), similarly to steps 4.-5. Once done, your profile should look similar to the image below.

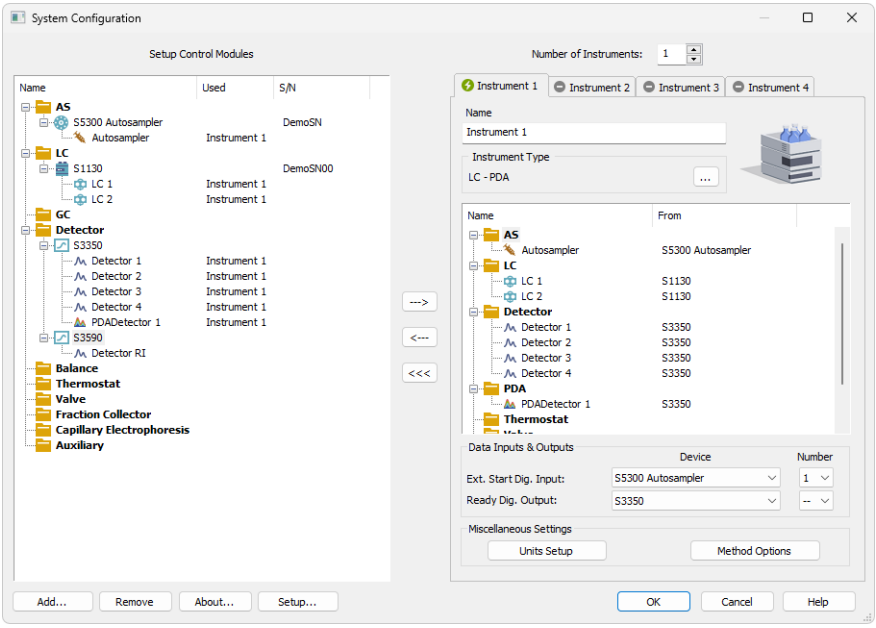


7. Close this dialog to return to the main *Launch Manager* window.
8. In the *Launch Manager* window select the *DAD* profile ⓓ which has not been configured but simply duplicated and click on the *Launch* ⓔ button.



9. Clarity has launched with the selected profile, open *System Configuration*. Since the configuration has been duplicated from the initial configuration, both detectors are present. Remove the RI detector from instrument and save the configuration by click *OK* in the *System Configuration* window.

   Note: This step may differ based on used control modules. In our case simply drag the RI module from right side to the left one. In other modules e.g., when using ICF you have to invoked the control module setup and change it there.



10. Launch each profile using the *Launch Manager* to make sure that correct configuration is loaded. Make necessary changes to the configuration if needed.

19 Extensions

Chapters describing topics related to specific Extensions.

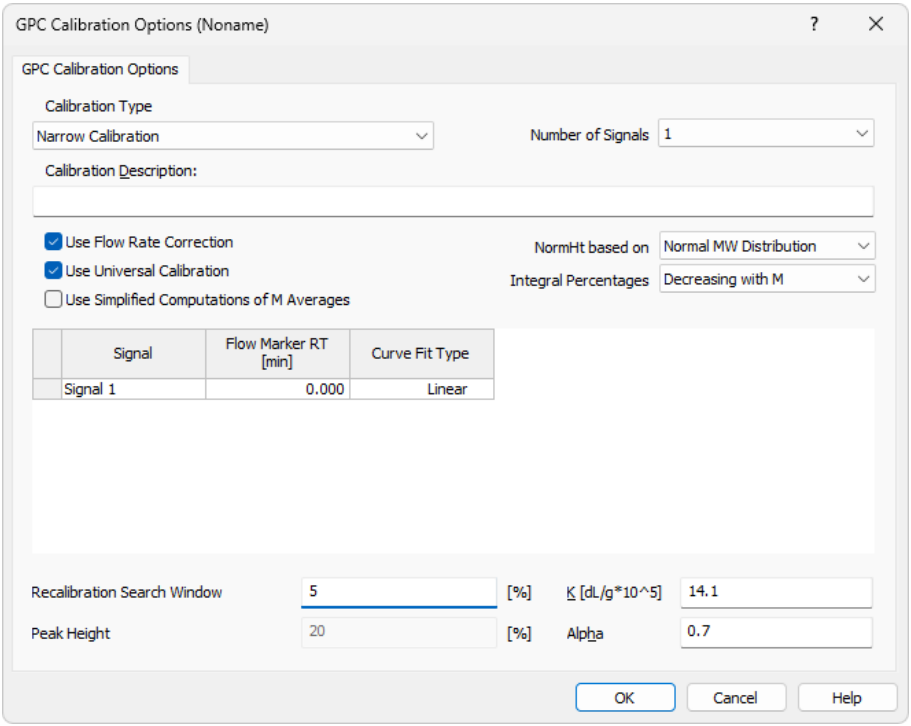
19.1 GPC operations

Following chapters describe specific procedures concerning GPC.

19.1.1 Creating a GPC calibration

To be able to create a GPC calibration, you need to have a measured and integrated GPC chromatogram of a standard sample and the instrument type of Clarity must be set to *GPC*. The *GPC* mode can be toggled on/off by selecting the *Setting - GPC Mode* in the *Instrument* window.

1. Open the Calibration window: select *Window - GPC Calibration* in the *Instrument* window or click on .
2. Create a new calibration file: select *File - New* or click on ⓐ .
3. Following dialog *GPC Calibration Options* will show up. Here you can setup various options and *Calibration Types*.



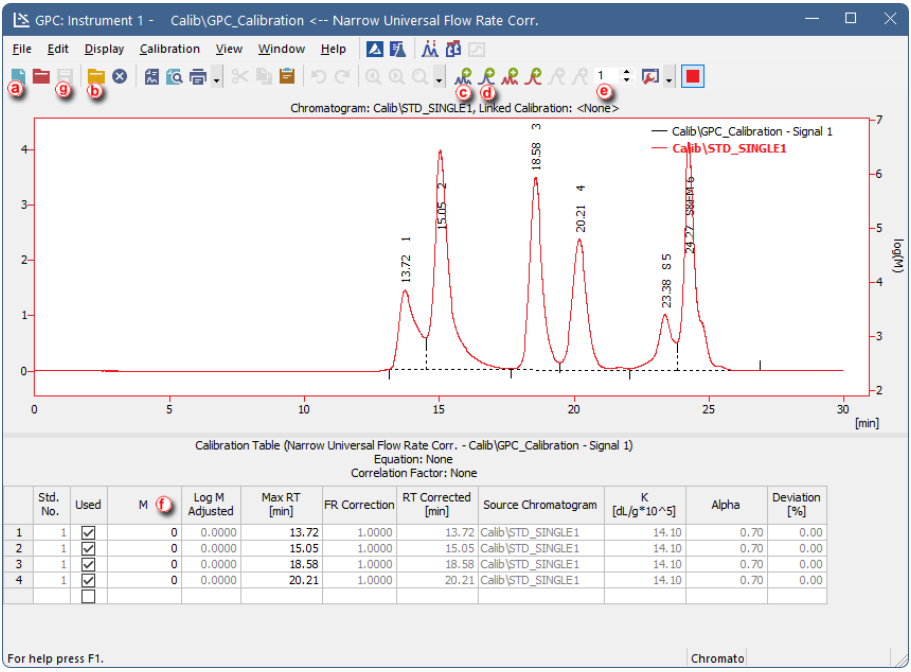
Caution: Once selected, the Calibration Type can't be changed later. More on GPC Calibration Options can be found in the *GPC Extension* manual, accessible on www.dataapex.com.

4. Click *OK* to save the calibration. To fill calibration name open the *File - Save As* dialog window.
5. Open integrated chromatogram of a standard: select *File - Open Standard…* or click on  ⓑ in the *Calibration* window.
6. Add peaks in the chromatogram of the calibration standard to the calibration file: select *Calibration - Add All* or click on  ⓒ (if you have multiple peaks in your chromatogram) or select *Calibration - Add Narrow Peak* or click on  ⓓ to add desired peak from your standard.

Note: If you have multiple chromatograms of standards, you can repeat these steps to add the desired peaks: open the standard and click *Add Narrow Peak*, then open another standard and click Add Narrow Peak, repeat for all standards. The number in field ⓔ is connected to the used standard and is not connected to the concentration level. Setting a peak on already used number will overwrite the values with a newly added one.

Note: When using any type of Broad calibration, *Add Broad Peak* will be enabled instead of *Add Narrow Peak*.

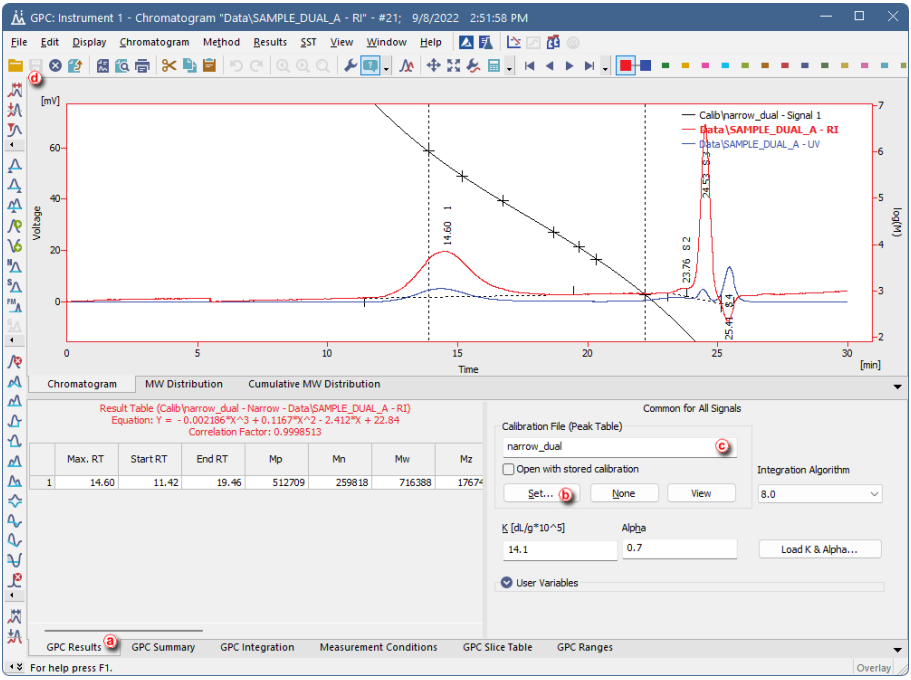
7. Fill in the appropriate molecular weight values for the respected peaks into the *M* column ⓕ .
8. Save the calibration file: from *File - Save* or click on  ⓖ .



19.1.2 Applying a GPC calibration to a chromatogram

If the calibration file is not assigned to the template method, the measured chromatogram will not have it linked either. To link a calibration file to a chromatogram do as follows:

1. Switch to the *GPC Results* tab ⓐ at the bottom part of the *Chromatogram* window.



2. To link the calibration file to the Chromatogram, click the *Set…* button ⓑ , so that the *Open GPC Calibration* dialog window will emerge. You can select calibration file from your current project or you can navigate to other folders.
3. Select the desired calibration file from the list and click *OK*.
4. Check that the *Calibration File (Peak Table)* field contains the name of the calibration file ⓒ .

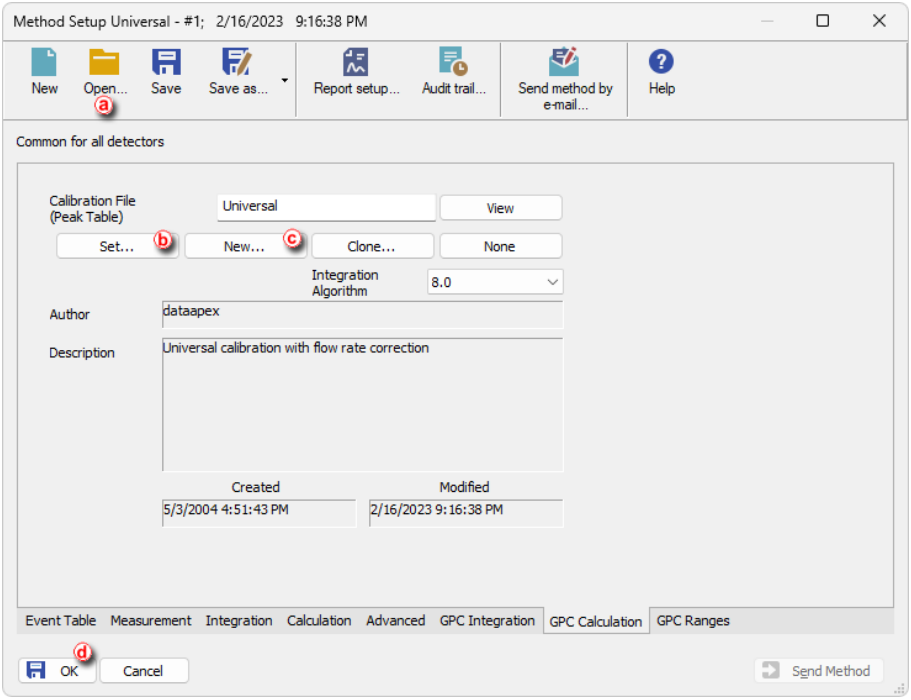
   Note: When no calibration file is linked to a chromatogram, the field *Calibration File (Peak Table)* contains inscription *(None)*.

5. Save the chromatogram by selecting *File - Save* or clicking on ⓓ .

19.1.3 Setting a GPC calibration in the template method

Setting a GPC calibration in the template method allows you to automatically calibrate all measured chromatograms using such method during analyses.

1. Open the *Method Setup - GPC Calculation* dialog by selecting *Method - GPC Calculation...* in the *Instrument* window.
2. Open the template method by selecting the *Open...* icon ⓐ .



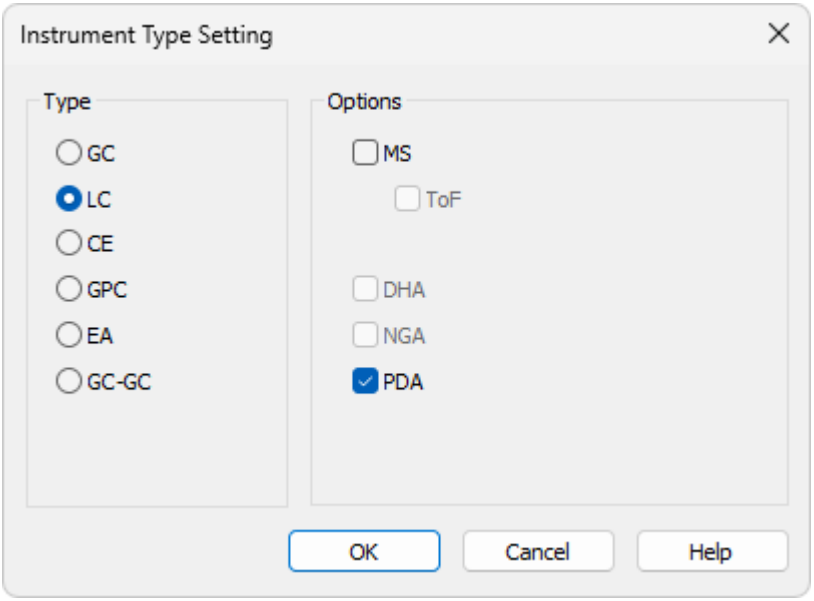
3. Click the *Set…* button ⓑ to set a GPC calibration file for the template method, or create a new one by clicking the *New…* button ⓒ .
4. Click *OK* ⓓ to apply and save the changes made to the template method.

19.2 PDA Operation

Pick the desired topic in the following chapters.

19.2.1 How to set Clarity instrument to display PDA data

- To switch an Instrument to *PDA mode*, select the **LC-PDA**, **GC-PDA**, **CE-PDA** or **GPC-PDA** option from the *Instrument Type Setting* dialog.
- *Instrument Type Setting* dialog is invoked by clicking on the … button in the *System Configuration* dialog.
- Options that are technically possible and have been purchased are enabled by default. Otherwise they are automatically disabled.



19.2.2 How to open PDA chromatogram

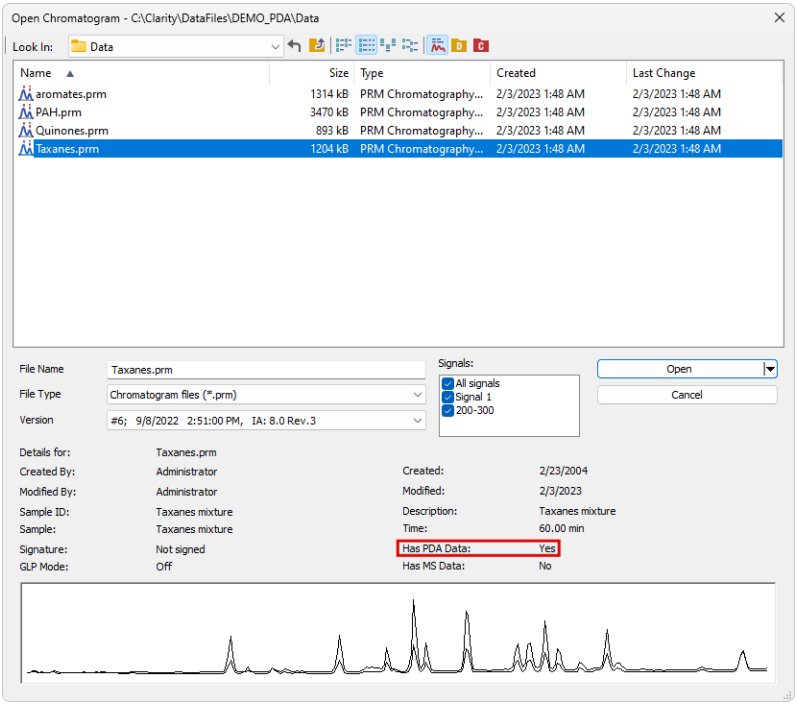
1. In the *Chromatogram* window, click the *Open Chromatogram* icon .
2. Choose a chromatogram that **has PDA data**.

   Note: Notice that the presence of PDA data is noted in the chromatogram details at the bottom part of the dialog.

3. After choosing chromatogram(s) click *Open* (or one of the other options when selecting multiple chromatograms).

   Note: For more information regarding the opening options refer to chapter Open Chromatogram in the Reference Guide.

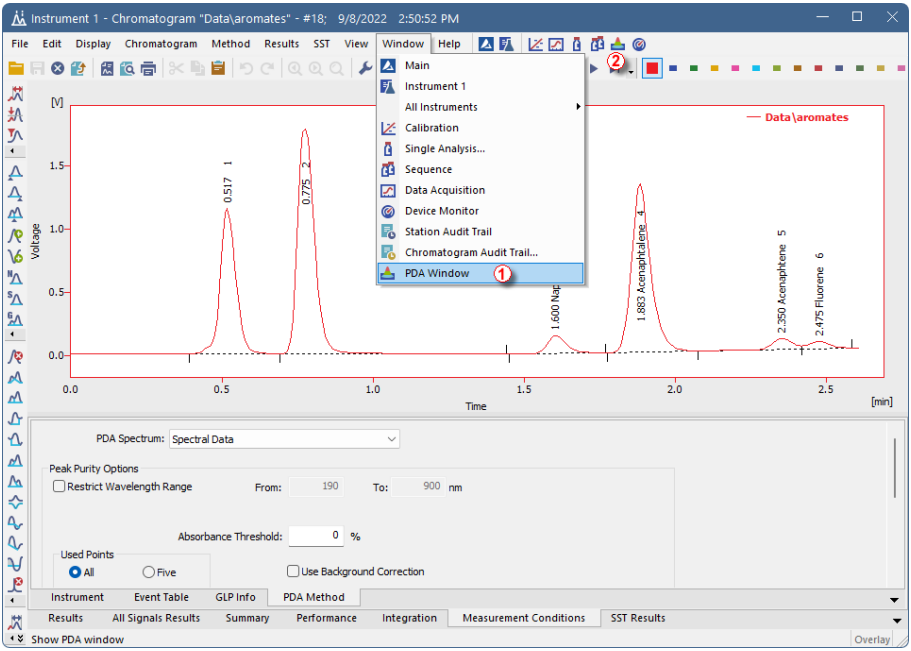
4. Next suggested topic is: "How to work with PDA chromatogram" on the next page.



19.2.3 How to work with PDA chromatogram

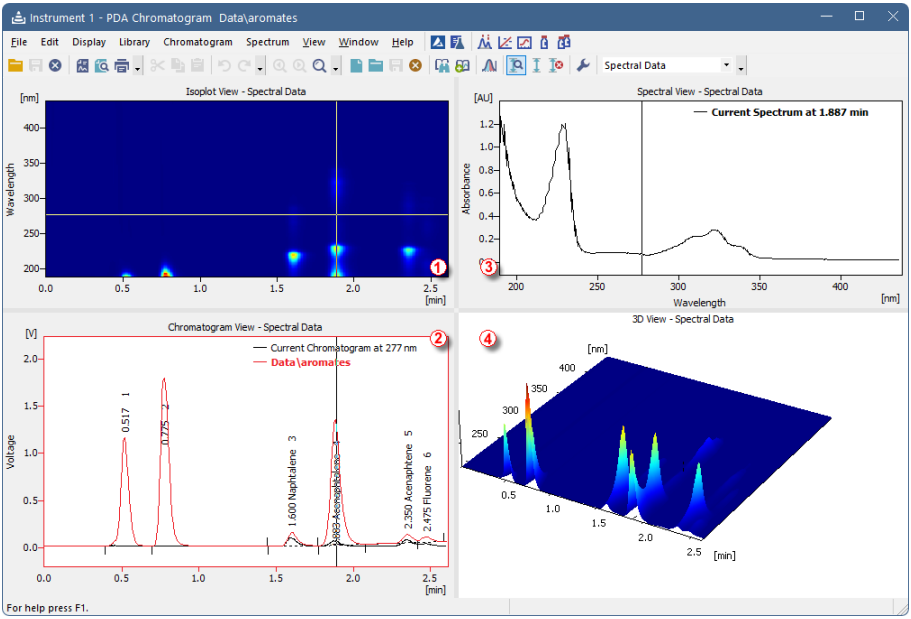
Instrument Type must be set to PDA - see the chapter **"How to set Clarity instrument to display PDA data"** on pg. **232**.

- To open *PDA Chromatogram* window navigate to *Window - PDA Window* ① in the *Chromatogram* window alternatively click the *PDA Window* icon ② .

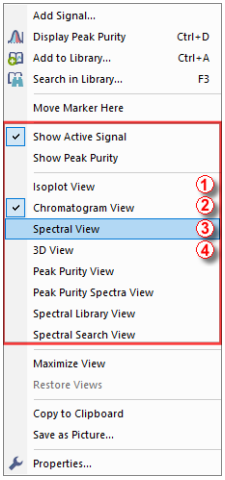


How to display PDA data

1. Spectra are automatically displayed in the *PDA Chromatogram* window upon opening chromatogram that contains **PDA** data.
2. By default, *PDA Chromatogram* is divided into 4 separate panes, each with a different information. You can change number of panes from the *View* menu



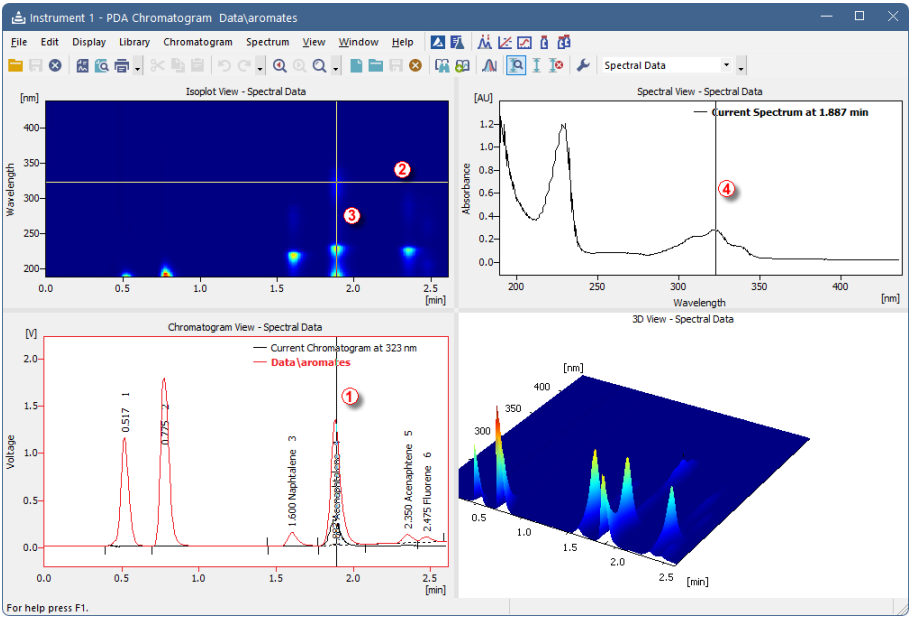
3. To change a view of a pane, right mouse click and select one of the views. For a more specific example, see the chapter **"How to search in PDA library"** on pg. **241**..
4. Numbers ① to ④ correspond to different views in the picture above.



How to use markers in PDA Chromatogram

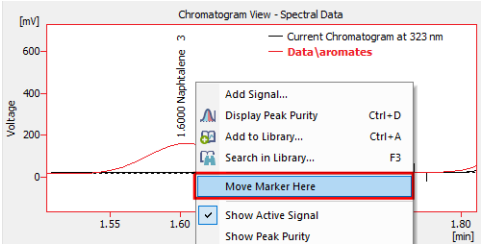
Panes in the *PDA Chromatogram* contain graphs with markers (thin lines of inverse color crossing the data plot ① ② ③ ④). Corresponding data are displayed according to the marker positions.

Note: Moving marker in one pane moves it accordingly in other ones. For example, moving marker ① in *Chromatogram View* will result in corresponding shift of marker ③ in *Isoplot View*.



- **To change spectral data based on time**: drag the vertical marker ① or ③ to the desired time. Note that the cross cursor changes to ⟺ once the move is possible.
- **To change wavelength**: drag the horizontal marker ② or ④ to desired position. Notice that the cross cursor changes to ↕ or ⟺ respectively once the move is possible.
- **To change both**: wavelength and time, move to the junction where the two markers ② and ③ meet. Once the cross symbol changes to ✥, left mouse click + hold + drag to your desired position.
- In the *3D View* you can move the whole graph to your area of interest - simply left mouse click + hold + drag. Note that markers in other panes will change accordingly to the moved area.

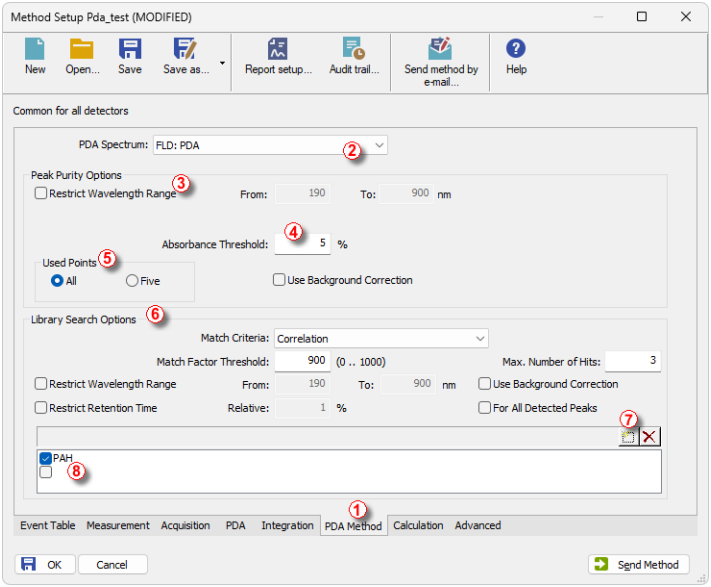
Caution: Increasing the zoom may cause that the markers to be out of the current view. To move them to the current zoom, right mouse click in the pane and use *Move Markers Here* from context menu.



19.2.4 How to set PDA method

This section deals only with method setup related only to **PDA options**. For a method setup see "Setting up a method"

1. *PDA Method* tab ① is available only when **PDA** Extension is configured on the given instrument.
2. From the drop down ② box select *PDA Spectrum* - if chromatogram contains more than one (typically DAD and FLD).
3. For the *Peak Purity Options* there are several settings which can restrict the evaluation of peak purity. It can be restricted based on the *Restrict Wavelength Range* ③ check-box and filling the range of wavelength in which it will be evaluated.
4. Another restriction can be made using the *Absorbance Threshold* ④ . This determines which part of peak will be used. For 0% spectra from entire peak will be included in calculation. For 5%, spectra which has less than 5% of absorbance compared to spectrum in apex will be excluded from calculation etc.
5. *Used Points* ⑤ specifies the number of points peak purity will be evaluated, either from the *All* the points or from the *Five* most significant.
6. In the *Library Search Options* ⑥ select criteria according to which the *Search in Library* command, described in see pg. **241**., will be performed.
7. To add another **PDA library** to search in, click the icon ⑦ and then click ..., from the dialog choose a library.
8. List of libraries that will be searched in is listed ⑧ . Alternatively enable/disable the search for specific library using the check-box.



19.2.5 How to display peak purity

Displaying peak purity is one of the fundamental tasks when ensuring that no co-eluting or co-migrating impurities contribute to the peak's response.

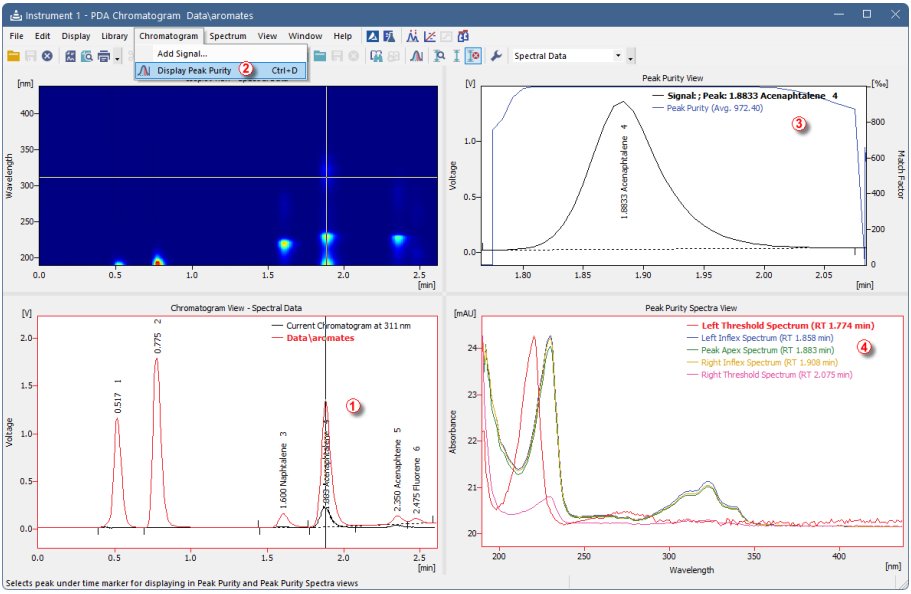
1. Used views: *Chromatogram View*, *Peak Purity Spectra View*, *Peak Purity View* and *Isoplot View*.

   Note: See chapter "How to work with PDA chromatogram" on page 234 for more information about using the *PDA Chromatogram* window functions.

2. Move the marker ① to the peak for which you want to display peak purity.
3. In the menu *Chromatogram* - click the *Display Peak Purity* ② command or click the icon in the toolbar.

   Note: *No peak selected* in the *Peak Purity View* means that the marker from step 2 is not positioned on any peak.

4. *Peak Purity View*: displays signal for selected peak and its calculated *Peak Purity* ③ .
5. *Peak Purity Spectra View*: displays spectra in several significant points ④ of the peak selected in the *Peak Purity View*.



19.2.6 How to work with PDA library

PDA Library serves for storing compounds along with their PDA data such as spectrum. Working with PDA libraries and data is possible from *PDA Chromatogram* window.

Note: It works on a similar basis as calibration. *Search In Library* command, searches the **PDA library** for similar spectrum as defined by your cursor axis in the chromatogram.

PDA library can be managed either from the menu *Library* or using the toolbar which is shown below:

Manage libraries:

- Create a new library: click the *New Library* icon.
- Open existing library: click the *Open Library* button and in the dialog choose your desired library.

Add compounds:

1. Add a spectrum into the library: click the *Add Spectrum* icon which opens *Spectrum Property*. In the dialog you can specify compound name and additional comment.

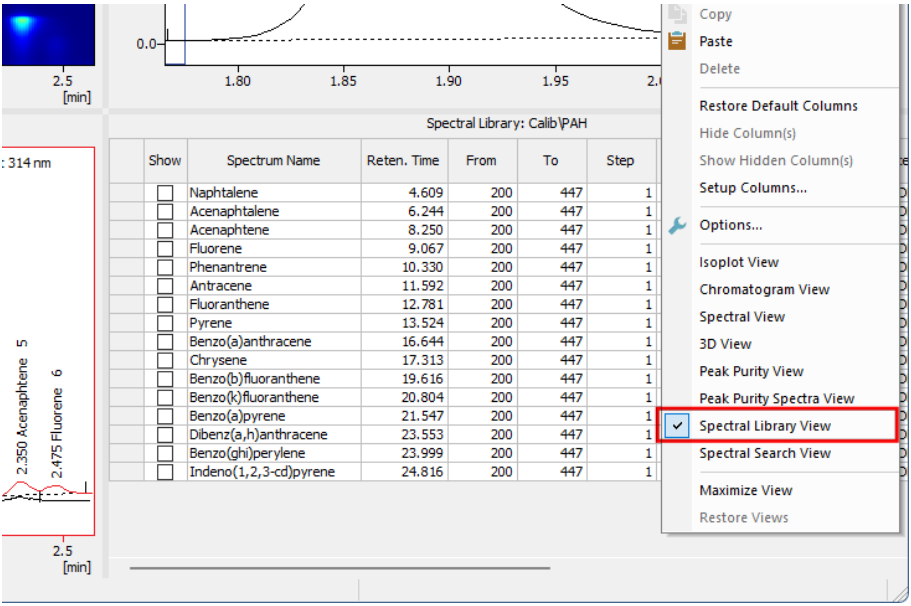
2. Add all identified peaks: click the command from the menu *Spectrum - Add All Identified Peaks*.

Note: Identified peaks are those that correspond to peaks identified and named in the calibration file. Peaks are added into the currently opened library.

3. Save library: to keep any changes made in the library, click the *Save Library* icon.
4. Close library: click the *Close Library* icon. If you have unsaved changes you will be prompted to either save the changes or discard them.

View library:

1. Spectral Library View: to view contents of your library, right mouse click in any pane of *PDA Chromatogram* windo and choose *Spectral Library View*. It contains a table with spectrum name, retention time and other parameters.



19.2.7 How to search in PDA library

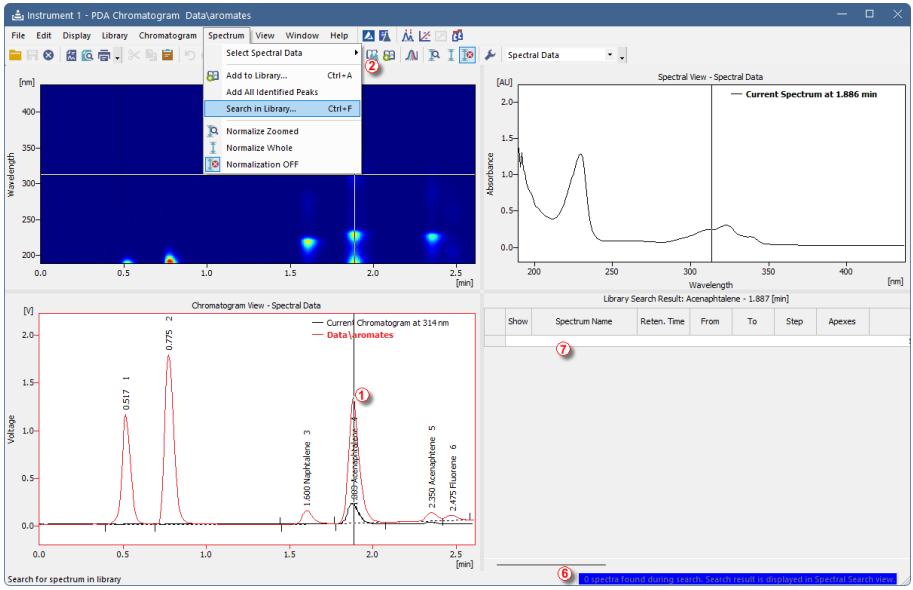
To search for a matching compound in the PDA library, there is number of ways.

Most common way to search in library:

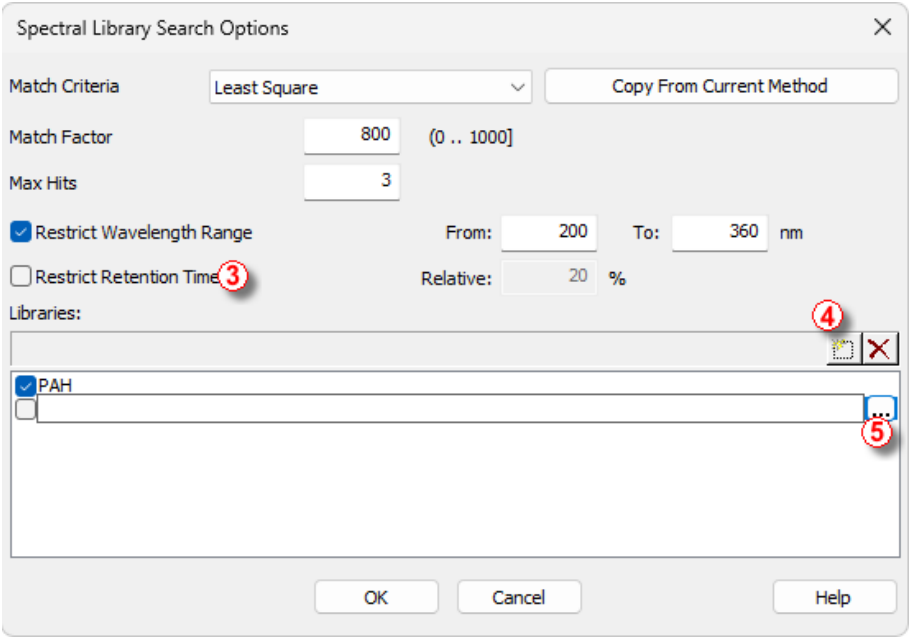
1. Move marker ① to peak for which you want to search in library.
2. Right mouse click to a pane you don't need and select *Spectral Search View* to display pane which shows search results.
3. Use the *Search in Library* ② icon from the toolbar. *Spectral Library Search Options* dialog will pop-up where you can further refine match criteria as well as searching across multiple libraries.

Note: Same can be achieved by pressing *F3* or *CTRL + F* on the keyboard. Alternatively you can use *Spectral Search View*command from context or *Spectrum* menu.

Search in library is based upon multiple requirements (mainly on *Match Factor*), if any of the requirements fails, the compound will not be displayed in the result of the search.



1. Invoke *Spectral Library Search Options* dialog according to steps 1-3 at the beginning of this topic.



2. Set the search parameters in the top half of the dialog.
3. Note that *Restrict Retention Time* check-box ③ should be enabled only when the **PDA library** has been created under same conditions as the measured chromatogram.
4. You can add new library entry using the ④ and use ⑤ to select library which may contaiint.
5. Once criteria are set up, click *OK* to apply them and to perform the search.

Possible problem during library search:

- After performing search, you may notice that the blue ribbon ⑥ at the bottom of the window that states *0 spectra found during the search* and *Library Search View* is also empty ⑦ .
- Possible reason for such behavior can be that the *Library Search Options* are incorrectly set up e.g., selected library does not contain matching spectrum.

19.2.8 How to view specific spectra in overlay

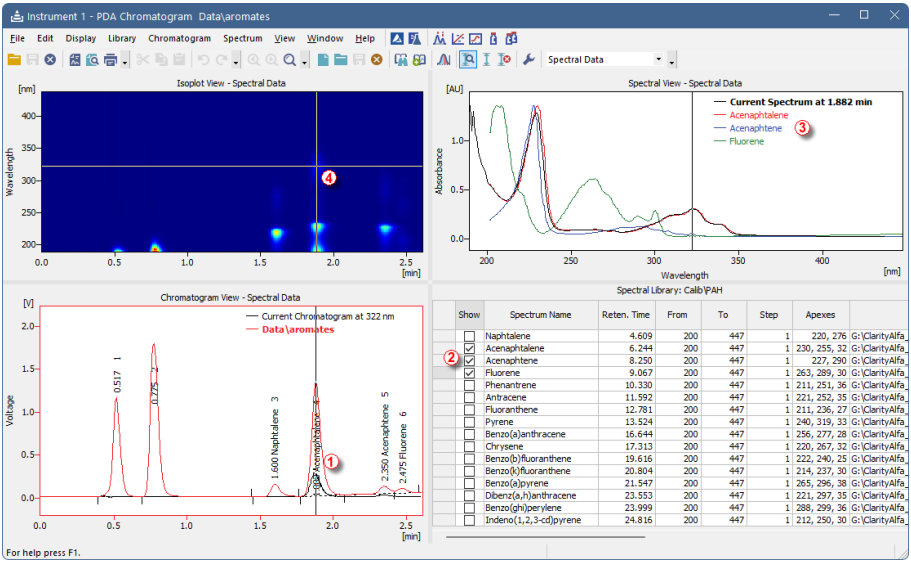
To view specific spectra from **PDA library** in overlay and thus have the opportunity to compare current spectrum against spectra from the **PDA library**, follow the steps below.

1. Used views: *Chromatogram View*, *Spectral Library*, *Spectral View* and *Isoplot View*.

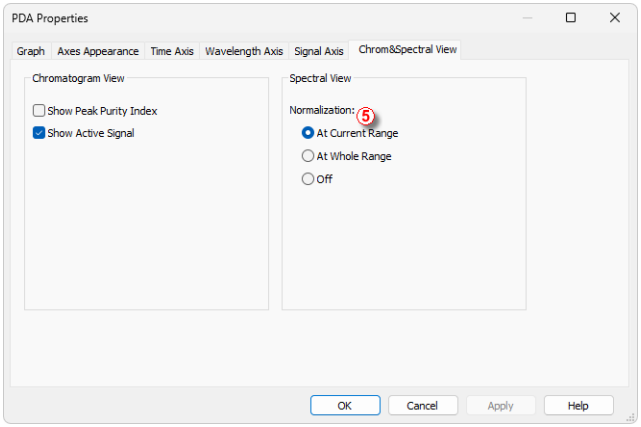
   Note: How to change views is shown in "How to work with PDA chromatogram" on page 234

2. Move the marker to your desired position ① .
3. In the *Spectral Library View* check the *Spectrum Name* for each ② spectrum that you want to show in the *Spectral View*.
4. *Spectral View* displays current spectrum along with other spectra ③ checked in the **PDA library**.
5. You can move the marker ④ in the *Isoplot View* to move alongside both wavelength and retention time axes.

Note that those steps can be also performed on results from the *Search in Library* - thus having spectra from the search in overlay.



Note: To compare spectra it is recommended to normalize their view. Right click into *Spectral View* and select *Properties*. In *PDA Properties* dialog navigate to *Chrom&Spectral View* tab and select *Normalization: At Current Range* ⑤ .



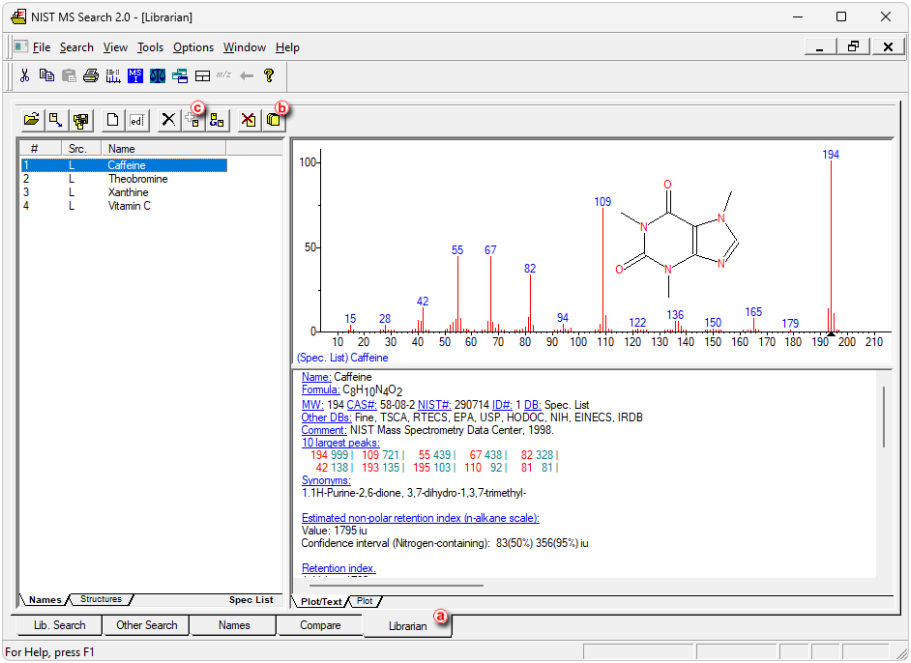
19.3 MS Operation

Pick the desired topic in the following chapters.

19.3.1 Creating and filling your own MS library

Own **MS Library** can be created using external program **NIST MS Search** accessed either from the *MS toolbar* or from the *MS* menu in the *MS Chromatogram* window:

- Use the *Manage Libraries* button to open the **MS Search**.
- Switch to the *Librarian* tab ⓐ .
- Use the *Create Library* button ⓑ to create your own library, and *Add to Library* button ⓒ to add the selected compounds into it.



To be able to add selected compounds (or spectra) to own library, such compounds must be selected in the *Librarian* tab. To do so, you can either perform the *Single Compound Search* or use *Add Spectrum to Library* icon or command from the *MS Chromatogram* window.

Add Spectrum to Library

After clicking the *Add Spectrum to Library* icon or command, the view will lock in the graph and will let you select the spectrum. After selecting the spectrum the *Add MS Spectrum to Library* dialog for inserting that particular **MS Spectra** appears:

- Set the *Averaging Time Range* field to perform averaging and smoothing of the spectra inserted into the library – if not selected, the actual spectra as clicked into the graph will be stored.
- Set a *Compound Name* under which you want to add the spectrum into the library.
- Press the *OK* button.
- The **MS Search** program will open, which allows you to add the spectrum into the library upon switching to the *Librarian* tab and using the *Add to Library* button.

Single Compound Search

After clicking the *Single Spectrum Search* icon or command, the view will lock in the graph and will let you select the spectrum. After setting the desired parameters and clicking the *Search* button the **MS Search** program will open. Switch to the *Librarian* tab and using the *Add to Library* button add the spectrum into the library.

Note: For more details on *Single Search Compound* please see the chapter "MS Libraries" in MS Extension manual.

19.3.2 Integration of signals in MS

In MS Extension, two types of signals can be integrated: Quantification Signals and Result Table Signal.

Result Table Signal

Result Table Signal serves for filling the Result Table with peaks other than those mentioned in MS Method. Any signal from the detectors configured on the particular Instrument can be selected as Result Table Signal.

- Switch to Integration tab in Chromatogram window.
- In the drop-down list select Result Table Signal (as defined on MS Method tab). If the Result Table Signal is not selected, integration table will not be shown at all.
- Apply integration operations as desired.

Quantification Signal

Quantification Signal serves for quantification of compounds mentioned in MS Method. Integration table is unique for each quantification signal. When multiple compounds are quantified on the same quantification signal they will share the same table.

- Switch to MS Integration tab in Chromatogram window.
- In the drop-down list select desired compound (defined by m/z ion and the name). Chromatogram will be focused on the compound's peak.
- Apply integration operations as desired.

Note: Even if you select Result Table Signal, e.g. TIC also as your Quantification Signal for any of your compounds, the integration table from Result Table Signal will not be used for such Quantification Signal. Each quantification signal has its own integration table.

19.3.3 MS Search and improving Match probability

This topic describes how MS Search, a library search is done and how to improve Match probability with library compounds.

19.3.3.1 MS Search - library search for compounds in MS

MS Search - Single Compound Search

Performs a simple search of a single spectrum in one or more spectral libraries.

- Open *MS Search* dialog using the *MS - Single Compound Search* menu or  icon in the *Chromatogram* window.
- Cursor will be focused into the chromatogram.
- Select desired search area, either an exact retention time or an interval (while holding **CTRL** key when clicking).
- MS Search dialog will be opened with filled in the selected search interval.
- In Max Hits select how many results (up to) shall be shown.
- Click on Search.
- Select the desired compound by checking the checkbox and possibly add such compound to your MS Method.

MS Search - Automatic Compound Search

Serves for searching of possible compounds of interest on a defined signal.

- Open *MS Search* dialog using the *MS - Automatic Compound Search* menu or  icon in the *Chromatogram* window.
- In the drop-down list select a signal to be searched and define the search retention time interval.
- Select Libraries to be searched in.
- Optionally set some Search Options and hit *Search* button.
- In the search results, you can select any compound at each peak found at specified retention time, sorted by the best matches.
- After selecting some compounds, you can add the to your MS method by clicking the *Add All Selected to Method* button.
- When selecting any of the compounds, you can compare the spectrum with the spectrum from library in the graph below the search results.

MS Search - Target Compound Search

Performs a simple search of a single spectrum in one or more spectral libraries.

- Open *MS Search* dialog using the *MS - Target Compound Search* menu or  icon in the *Chromatogram* window.
- By clicking the *Select Compound...* button open *Select NIST Compound dialog*.
- Enter the name of the desired compound and select it from the table below. If you use more than one library, select the library to be searched in the combobox on the left side.

- Select retention time or whole chromatogram to be searched in and hit the *Search* button.
- You will get results - exact retention times of peaks with the highest probability.
- You can filter the results by *Filter by Min Match Factor* slider/editbox, based on Match probability.
- Select the desired compound and add such compound to your MS Method.

19.3.3.2 Improving Match probability

Restrict m/z Range

Serves for narrowing the m/z range to be searched. Provided you know the range of compounds' m/z you are interest in, you can omit the remaining m/z from the search, thus raising the Match probability. In case you enter wrong m/z range, a dialog will appear and will tell you what the m/z range of the current chromatogram is.

Use Selected m/z

Defines what m/z should or should not be searched.

- *Search Only Selected* will search only for the defined m/z. Suitable when you know typical m/z for the compounds you are interested in.
- *Search All But Selected* will omit selected m/z from search. Suitable e.g. when you don't want the used solvent to interfere with the search results.

Background subtraction

Defines exact points and/or intervals of retention time to be omitted from the search. Such values can be inserted manually or selected directly in the graph.

Preview Spectrum in Library

Shows the search results, based on the above-mentioned options, directly in the NIST library. You can check here, whether your setting were right, or even find out the typical m/z for selected compounds.

19.4 SST operations

Following chapters describe specific procedures concerning SST.

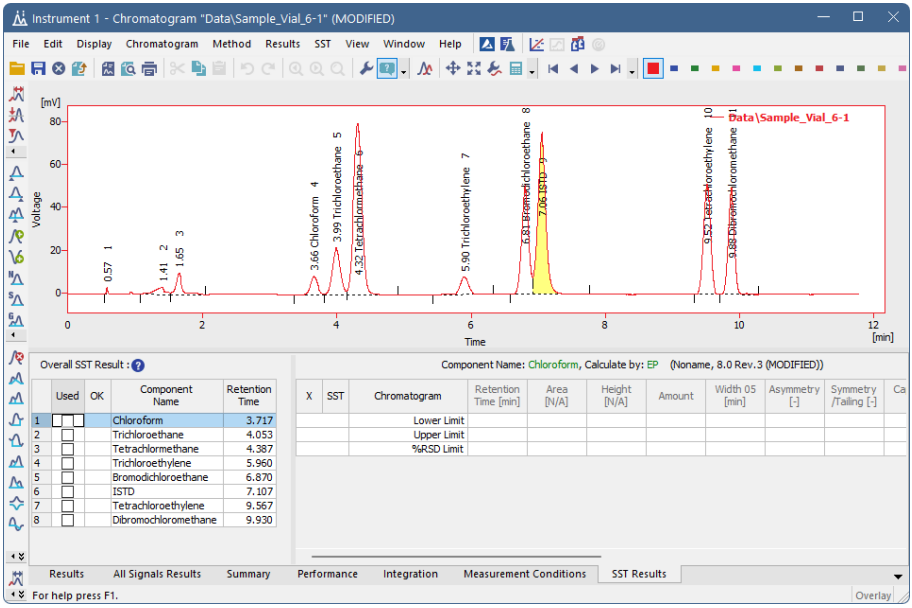
19.4.1 Using SST for quality control

The optional SST Extension allows to set up limits for selected parameter. It compares measured values against those preset limits and perform actions based on the result. The SST works with calibrated chromatograms and the evaluation is based on the compound name in calibration.

Here we provide an example on how to create SST method which can be used to check if the control samples are within expected limits and how to set up sequence to be checked by different SST methods.

Caution: In order to get desired reactions to passed or failed limits for this scenario it is necessary to have chromatogram **Overlay Mode switched off**.

1. Create a SST method in the Chromatogram window using the:
   - *SST - SST Result* to display the SST tab
   - *SST - New* to create a new SST method
   - *SST - Update from Calib* to load the list of peaks from a calibration file (There has to be calibration linked to the chromatogram on the *Results* tab)
   - Than the screen will look similar to this:

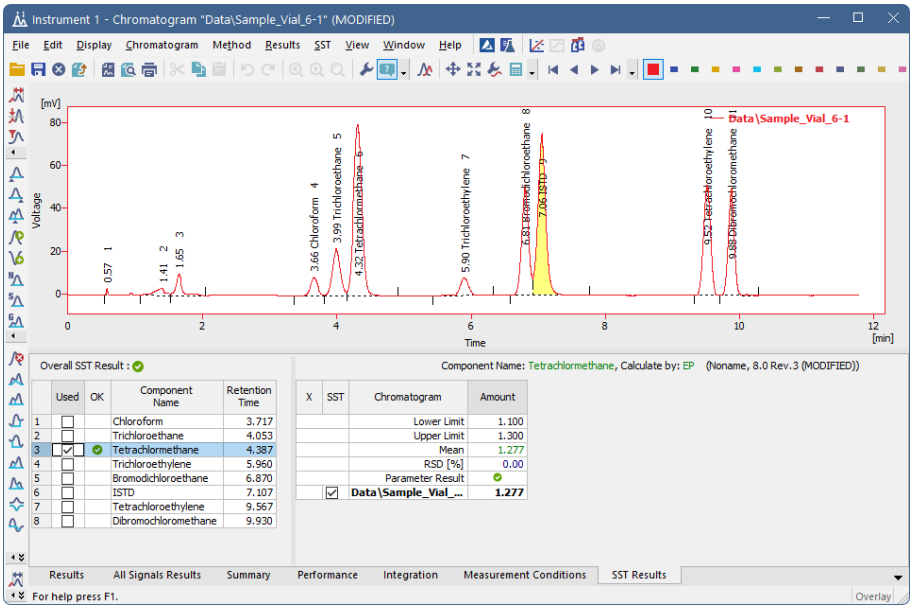


2. As the second step, fill in the necessary parameters for the limits. *(In this case Tetrachlormethane peak is the one used to check actual against expected value in Control sample., Expected Amount is 1.2, lower limit is 1.1, upper limit 1.3.)*
   - Select the checkbox of the *Tetrachlormethane* in the table on the left.
   - Double-click the *Amount* column in the table on the right to activate it.
   - Set the 1.1 value in the *Lower Limit* cell of the column, 1.3 into the *Upper Limit* cell.
   - Right-click the table and select the *SubParameters* item. On it uncheck the *%RSD Limit* item (as you do not need it) and select the *Each Individual Value*.

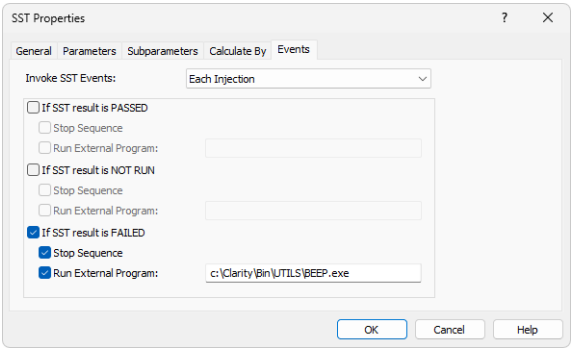
More Info:

- When the SST Result is based on *Each Individual Value* than it compares each evaluated value with the Upper Limit or Lower Limit.
- When the SST Result is based on *Mean of All Values* than it compares average value of all opened chromatograms with linked calibration with the Limits. So pay attention to all opened chromatograms.

- You may hide the inactive rows and columns by using the *SST - Show All Columns* and *SST - Show All Rows* items in the menu.
- The result of the check is displayed by the green tick mark or red cross. You can validate several different parameters at the same time.
- You should see similar result now:



3. In another step you can set reaction to the test result:
   - Use the *SST - Events* command to open the *SST Properties* dialog again.
   - Set the tab as on next picture (sequence will stop and beeping sound will be played if SST fails):

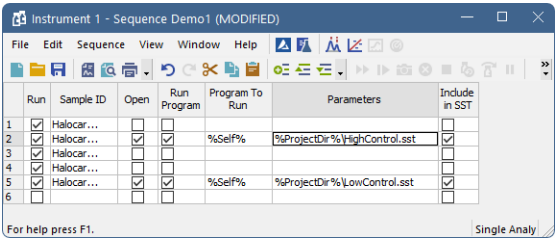


- It is possible to trigger any external program wanted and set different events on failed or passed check etc.

4. Save the created method by using the *SST - Save item* (I used HighControl.sst file name).
5. You can create other SST methods in the same manner. For example if you use multiple calibration levels it is possible to create methods which control each level against its own limits.

How to set up sequence to be checked by different SST methods

- Set the sequence accordingly:
  - Right-click the sequence table and select the *Setup Columns* item.
  - Set the columns *Open*, *Run Program*, *Program to Run*, *Parameters* and *Include in SST* as visible. Any other columns may be displayed as desired.
  - Set the columns for the rows with *High Control* and *Low Control* samples as shown on the next picture:



  - This will force Clarity to open the correct SST method for the row and perform the check.

Caution: Parameters like *%Self%* are case sensitive. All 3 checkboxes *Open*, *Run Program* and *Include in SST* must be checked for SST to work correctly.