MyAssays Desktop

User's Guide

MyAssays

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Introducing MyAssays Desktop

MyAssays Desktop is an easy-to-use data analysis tool for bioassays.

MyAssays Desktop is the only desktop tool with an integrated search-and-download feature providing direct access to the growing online database of protocols from myassays.com. The preconfigured data analysis protocols cover popular applications and support for commercial assay kits (backed by leading kit manufacturers); these are all ready to download and integrate into your lab's workflow.

Beyond this, if your application is not already supported, the software provides a powerful yet easy-to-use system to build and customize your own data analysis protocols.

MyAssays Desktop provides first-class tools for:

- Data Import
- Data Visualization
- Data Analysis (including advanced Curve Fitting)
- Reporting
- Export
- Workflow Integration

How to Use This Guide

We have arranged this guide to help you get started quickly and to simplify how you can find the information and details that you need.

This **Introduction** contains a summary of the key features and editions of the software.

The **Getting Started** section covers installation and general orientation information. It also includes a Quick Start section to show you where the basics are and to walk you through your first data analysis. It also provides links to further information and online video guides.

The **Working with MyAssays Desktop** section dives into further detail, covering each element in more detail with the content arranged into logical sections. This includes:

- getting around the various applications
- protocols
- common tasks (such as importing, exporting, curve fitting, marking outliers to exclude, and much more)

The final **Reference** section provides a more complete description of each facet. Here the content is arranged to simplify quick retrieval.

What Should I Read?

We encourage all new users to read through all of the **Introduction** and **Getting Started** sections.

You will find more information about the topics and get a better understanding from the **Working with MyAssays Desktop** section. Feel free to skip sections here that are not relevant to your applications. If you're working with particular protocols or need to perform specific tasks, be sure to follow the relevant sections under **Protocols** and **Common Tasks**.

We don't expect you to read through the **Reference** section in order; it is more useful to consider it a resource for providing further details as and when you need them.

Features

Supported Measurements

MyAssays Desktop is designed to process measurement data from any microplate or rack-based reader. In addition, it can import and analyze data from any scientific measurement device.

The software supports measurements from any type of detection technology (such as absorbance, luminescence, fluorescence, counts, counts-per-minute), with data collected from each sample as:

- **Endpoint** – A single measurement value
- **Kinetic** – Values measured over time (data can be processed with or without time stamps)
- **Spectral** – Data measured at various multiple wavelengths
- **Multiplex** – Multiple analyte measurements from each sample

Data can be collected and processed together in any combination. For example, dual endpoint measurements read at both 665nm and 620nm can be imported and analyzed (perhaps with a ratio calculation). Even more exotic measurement combinations are possible with features to process multiple endpoint, kinetic, and spectral data all together within the same analysis.

Multiple Plate Measurements

Data can be captured and processed from measurements made from a single microplate or from across multiple plates. Data to process together can be arranged in a single file or in separate files that can be imported and merged together. Curve fitting can be performed with standards defined on some or all plates, with curves computed using averages of standard measurements across plates or from the measurements on the same plate.

Data Import

For maximum flexibility, MyAssays Desktop provides a number of ways to get your data into the system. These are designed to help you get started quickly and to streamline your work-flow:

- In the simplest case, copy and paste your data from any source for quick and convenient analysis.

- The included extensive Import Script library can automatically detect and import data from many proprietary instrument formats. Simply select the file to import; if it's supported, the data and any meta data (data accompanying the read such as measurement date/time, barcode, etc.) are imported automatically. (For the latest list of currently support format, see: **https://www.myassays.com/supported-file-formats.html**.)

- If your data is in an Excel document, the import wizard will guide you through the process of importing the relevant data and meta data from the cells in your workbook. Mark the cells to import the data (with these settings saved to your protocol).

- For other files, use the intuitive and familiar import wizard to specify how to process and import the data from your files.

If your data is spread across multiple files, the import wizard can import your data together for a combined analysis.

Importing Other Types of Data

In addition to first-class support for handling your measurement data, MyAssays Desktop supports importing other types of data, including:

- Measurement meta data (data associated with your readings, such as date/time, plate ID, operator ID, etc.)

- Worklist (e.g., containing sample IDs, dilution factors, standard concentrations)

- Plate name/IDs, barcodes, analyte names, etc.

Beyond this, it is also possible to import any data analysis parameters (including layout, calculation settings, fit method, etc.) from data stored or produced by other systems. In this way, your data analysis process can be fully automated or con-trolled by other software.

Data Visualization

MyAssays Desktop provides a rich modern user interface to clearly present data, configuration, and analysis results, including:

- 2D and 3D interactive views of your measurement data
- Heatmap
- Histogram
- Interactive charts

  - Overlay charts (e.g., by fit, by sample, by replicate) in any combination for convenient visual comparison
  - Zoom in/out
  - Mark outliers (to exclude from analysis) directly from a chart or plate (including outliers in endpoint, spectral, kinetic, and multiplex data)
  - Specify data analysis ranges interactively (e.g., mark subsets of data to process for kinetic or spectral analysis)

- Sample labels for data

  - By name
  - By color
  - By graphical value bars

Data Analysis

Beyond the online database of ready-to-use preconfigured protocols, MyAssays Desktop provides a comprehensive data analysis engine supporting a wide range of applications.

The system provides a toolbox of preconfigured components (including over 30 transforms) ready to bolt together in any combination. Each transform performs a single mathematical task on measurement or calculated data. Transforms include endpoint, kinetic/spectral, and kinetic/spectral reduction.

Transforms include:

- Blank Correction – Subtract a specified background value (or values) from all samples
- %CV – Calculate percentage coefficient of variation of replicates
- Standard Curve Fit – Calculate concentrations from a standard curve fit
- Maximum Slope – Find the maximum slope and report its values (such as slope/time to onset/response)

For a complete list of the 30+ available transforms, see Transforms.

Transforms are linked to your sample layout. If your sample layout changes between assay runs (or even to correct a preparation error), the analysis takes this into account. No reconfiguration of analysis is required to work with different layouts.

The transforms parameters are completely customizable. In addition, a rich expression-based language enables you to specify parameters and methods as mathematical operations using an expressive familiar (Excel-like) syntax.

Results with Labels

Label your results using your own text and highlights. For example, perhaps samples with a response or calculated concentration greater than a specified amount should be clearly indicated as such; use labels to define your own rules accordingly.

Validation

Add automatic checking of measurements and results. For example, check that your samples are within an expected range by defining your own validation rules. These are evaluated with each run of your assay, with the results clearly presented as pass or fail in the report.

Curve Fitting

MyAssays Desktop provides the curve fitting tools you would expect, plus more, including:

- Quantitative tools – Weighted and unweighted 4-parameter logistic, 5-parameter logistic, and linear regression (plus legacy methods such as cubic spline, smoothed spline, cubic regression, point-to-point, and many more)

- Fit data to your own user-defined curve fitting equation

- Best Fit feature automatically compares multiple fit methods and applies the best (with customizable scoring methods)

- Goodness of fit measures reported – R^2, aR^2, SE, F-statistic, P-value, AIC, AICc, BIC and more (and available for further automatic scrutiny such as validation messages or best fit scoring)

- Automatic computation of LLOQ, ULOQ, and LOD based on customizable precision and accuracy thresholds

- View computed curve fit equations in mathematical symbolic notation

Reporting

For some applications, the most convenient type of report to work with is sheet-based; for other scenarios, a page-based structure is more suitable.

- Sheet – A spreadsheet structure comprising of a workbook of worksheets of cells
- Page – Structured ties to a fixed sized printed format (e.g., A4)

For these reasons, MyAssays Desktop supports both. Easily switch between report views to best work with your content as you need to.

Excel Objects

Exported Excel content features native Excel objects (e.g., chart content). This means that you can edit and extend the output using the full power of Excel.

MyAssays Desktop protocols reports can be configured to include power-features such as value bars for enhanced viewing and visualization of content.

Report Templating with Word

For page-based reporting, use the full power of Word to define a reporting template to apply to your MyAssays Desktop output. This can include any content you can create in Word, such as headers, footers, custom images, fonts, macros, signature line, etc.

Analysis results are merged into your Word template for maximum user-customizable reporting output.

Export

MyAssays Desktop provides a wide variety of export options and settings:

- Export to XLS/XLSX/PDF/DOCX/TXT/CSV/HTML/XML.
- XSLT support is provided, enabling custom export definitions to be created for exacting output as required by your existing system.
- Specify the export destination folder.
- Specify the naming convention of the output files, including references to imported meta data (for example, name your output files to match a barcode imported along with your measurement data).
- Configure a protocol to automatically export with these settings so no user intervention is required.

Workflow Integration

MyAssays Desktop has been designed with your workflow in mind and provides a number of features and tools to integrate with other systems and streamline your process.

For routine data analysis, your reports can be generated without any user interaction required at all. For example, MyAssays Desktop data analysis can be performed completely automatically, with data (including any combination of measurement data, sample IDs, parameters, etc.) being imported, analyzed, exported, or printed as soon as a new measurement file is detected from your reader software.

Command Driven

MyAssays Desktop analysis provides a complete command-line interface. Launch the analysis directly from another application or process. Specify measurements files to process, worklists to use, output options. All data analysis parameters can be sent from the command line.

Launcher

To facilitate working with the command line, the user-friendly Launcher utility simplifies building your command line with a point-and-click interface.

Folder Poll

To integrate more effectively with other systems (such as instrument control software), use the integrated Folder Poll tool to automatically launch MyAssays Data analysis as soon as a new data file is detected.

XSLT Export

For integration with other systems, such as LIMS which might have very specific data formatting requirements, utilize the XSLT feature to output data in the exact format required. Example XSLT files are installed to help you get started.

Advanced XML Configuration and Tooling

Inside the system, a well-design XML structure is used to define the data analysis of every assay. Most users will not need know about its existence, but for those specialist scenarios, advanced users can configure data analysis using a powerful built-in XML editor complete with inline help and auto-complete features.

Product Editions

MyAssays Desktop is available in various configurations to suit different budgets and needs:

Basic: Operates with preconfigured protocols (either those included in the installation package or as downloaded from MyAssays.com). The Basic edition allows tweaking limited protocol parameters (such as fit method, sample concentrations, layout, etc.).

Pro: Includes all of the features of the Basic edition plus functionality for creating new custom protocols. This edition provides full access to the Matrix-Transform data analysis model, offering a wealth of easy-to-use tools for building data analysis solutions tailored to your lab.

Pro Add-Ons:

Optional add-on functionality is available for the Pro edition:

Multiplex

Quickly and easily perform multiplex data analysis on your multiplex data:

- Calculate results from multiple analytes within a single analysis step.
- For quantitative analysis, apply the same curve-fitting method to each analyte or use the Best-Fit option to automatically apply the most appropriate fit method for each analyte.
- Specify concentration values as a series (defining the starting concentration of each analyte and the series) or enter specific concentrations for each analyte.
- Specify units per analyte or common units.
- Paste in or import measurement data, analyte names, starting concentration values, or specific concentration values.
- Includes import methods to automatically extract multiplex measurement data and analyte names from Luminex® xPONENT and Meso Scale Discovery Workbench® CSV and TXT formats.

QC

The QC option provides a range of inter-assay analysis features to support the continuous-monitoring automatic validation that your assay remains in control:

- Monitor, track, and chart observations across assay runs.
- Any numerical result can be tracked. For example, monitor raw or calculated control values, %CVs, or curve R^2 values.

- Include Levey-Jennings and CUSUM charts as part of each assay report to compare and visualize the latest results with historical data.

- Generate QC reports on-the-fly as and when needed.

- Automatic validation can ensure that the monitored observations are within defined acceptance parameters. Violations are clearly indicated in the report. For example, an acceptance rule can be defined to test whether a control result of a particular assay run is within a range of +/- 2*SD of the historical observations.

Enhanced Security

For those users working under GxP or 21CFR11, the Enhanced Security edition offers the features necessary to achieve compliance. The software provides protected and tracked access to system features inside a secure environment with:

- Access control – Authentication and authorization are used to permit/deny access to MyAssays Desktop Pro functions.

- Operating-system-level security-data storage protection – Only authorized users can make changes to MyAssays Desktop Pro data files and settings.

- Audit-trail and log-in log – All activity is stored against the logged-in user with a date/time stamp.

- Data certification – Restrict the system to handle only certified data (eliminating the processing of tampered data).

- Password protected and locked outputs (e.g., locked Excel outputs).

Rack Layouts

If your reader measures samples in tubes or vials (rather that microplates), then the Rack Add-On is for you:

- Rack Layout Editor – Define layouts in accordance with racks of tubes or vials.

- Dynamic sample layouts – layouts are automatically adjusted based on the number of samples actually measured.

- Support for microplate layouts included (for the multifunctional lab).

Licensing

Single, multi-seat, and site licenses are available for all editions and add-ons.

Edition Comparison

Feature	Basic	Pro
Downloadable Protocols (from myassays.com)	✔	✔
Installed Protocols:	✔	✔
• 4PL (Constrained by B0 and NSB) Quantification	✔	✔
• 4PL Quantification	✔	✔
• ATP Lite	✔	✔
• Best-Fit Quantification	▬	✔
• DNA 260nm	✔	✔
• DNA Quantification	✔	✔
• Dose–Response Curves for EC50	▬	✔
• ELISA	✔	✔
• ELISA (96 well) - Single Page Report	✔	✔
• LANCE cAMP	✔	✔
• Linearized (Logit-Log) Quantification	✔	✔
• Liquid Handling QC	✔	✔
• OQ Absorbance	✔	✔
• Standards Curve with Titres and Parallelism Plots (Dual)	▬	✔
• Z-Factor	✔	✔
Import Script Support	✔	✔
Excel Import	✔	✔
Worklist Import	✔	✔
Data Visualization (**3D View**/**Heatmap**/**Kinetic Overlay**/**Spectral Plot**)	✔	✔
Versioned Analysis	✔	✔
Export to XLSX/DOCX/PDF	✔	✔
Edit Protocols	Limited	✔
Report Customization	Limited	✔
Custom Export (e.g., for LIS/LIMS)	▬	✔
Multiple Plate Support	▬	✔
Create New Protocols (New Protocol Wizard)	▬	✔
Matrix-Transform Data Analysis	▬	✔
Custom Validation Rules	▬	✔
Custom Evaluation Expressions	▬	✔
Best Fit	▬	✔
Fit to Library Curve	▬	✔

Feature	Basic	Pro
Folder Poll	▬	✔
PLA	▬	✔

Getting Started

Installation

System Requirements

MyAssays Desktop runs successfully on the minimum system configuration listed below. If your computer does not meet the technical requirements, you may be not able to use MyAssays Desktop.

CPU:	Intel/AMD 2 GHz or faster processor (32-bit or 64-bit)
Memory:	2GB RAM
Available disk space:	300MB
Operating systems:	Windows 10, Windows 8.x, Windows 7, Windows Vista, Windows XP SP3
Other software:	Microsoft .NET 4.0.3 or later
Display:	1024 x 768 or higher resolution

Installation Process

The latest version of MyAssays Desktop can be downloaded [here](#).

Upon downloading the installation package:

1. Double-click on the installation package file to launch MyAssays Desktop Setup Wizard and click **Next** to continue.

2. To proceed with installation on your computer, you must accept the terms of End-User License Agreement. Please read the license agreement carefully and then check **I accept the terms of the license agreement** box and press **Next** to continue.
   If you do not agree with terms of the license agreement, click **Cancel** to exit the setup wizard.

3. If you are content with the default path, press **Next** to continue.
   To pick another destination folder, press **Change...** and set preferable root.

4. Check the choices you made and press **Install**. The process will take a few moments.

5. Click **Finish** to complete the installation. Check **Launch MyAssays Explorer on exit** to start working with the software immediately.

6. [Activate MyAssays Desktop](#) with your product serial code and activate your license in order to start working with the application.

> MyAssays Desktop can be installed and run alongside the legacy software WorkOut. Please refer to [Installing Alongside WorkOut](#) for further details.

Launching

After installation, you will see the MyAssays Desktop Explorer icon on your desktop. To launch the application, click on it or:

On Windows 10:

Choose **Start**, then scroll down to and click on the MyAssays Desktop Explorer icon.

For earlier versions of Windows:

- **Windows 8.1 or Windows 8:** Swipe up or choose the arrow at the bottom of the **Start** screen to see a list of all your applications. You might need to scroll left or right to see the MyAssays Desktop Explorer.

- **Window 7:** Choose **Start > All Programs** to see a list of all your applications and click the MyAssays Desktop Explorer icon.

> To access MyAssays Desktop Explorer more conveniently, you can pin it to your Start menu or taskbar. Right-click on the desktop icon and select **Pin to Start** or **Pin to Taskbar**.

Product Licensing

To use MyAssays Desktop, you must provide and activate a valid license.

You can obtain a trial, temporary, or permanent license from MyAssays Ltd.

A serial code represents your license or licenses (for multi-seat site licenses) to use the software. When you first launch the software, you must enter a valid serial code and activate your license.

> A single license can be transferred between systems but can only be active on one system at a time.

License Activation

On launching MyAssays Desktop, if there is no valid license already installed, then the Licensing Activation Wizard will appear.

> ℹ️ An administrator account is required to activate (or deactivate) a license.

If you have a serial code, select the **I have a serial code to activate** button and press **Next**.

> 📝 If you do not yet have a serial code, then you can obtain a trial serial code from https://www.myassays.com/download-and-install-myassays-desktop.html by clicking the **Claim Key** option.

Paste in the serial code along with your email address into the Licensing Activation Wizard and press **Next**.

> 📝 Your email address is required for administrating your license. Access to this email account will be required to transfer your license to another system in the future.

Online Activation

If your PC is connected to internet:

1. Check the **Use internet connection for activation** checkbox and press **Continue** to proceed.

2. Fill in your personal details and press **Activate**.

3. When prompted to grant **User Account Control** permission to access the licensing settings, confirm by selecting **Yes**.

4. If the license activation is accessed, a confirmation window appears.

5. Press **Close** to exit the **License Activation Wizard** and start the application.

Offline Activation

If your PC is not connected to internet, it is still possible to activate MyAssays Desktop using a separate internet-connected device. In this case:

1. Uncheck **Use internet connection for activation** checkbox and press **Continue** to proceed.

2. A QR code will appear; if you have a network-connected device (such as a smartphone or tablet) capable of reading this, then scan the QR code and follow the instructions.

Alternatively, copy and paste the provided link and open it on a system with internet connection (perhaps by transferring via USB memory device).

3. On successful activation of your serial code from your internet-connected device, a **license key** text string will be generated (with a copy sent to the provided email address).

4. Transfer this text string to your PC where MyAssays Desktop is installed and paste in the **License Activation Wizard** under the **license key** setting.

5. When prompted, grant **User Account Control** permission to access the licensing settings and confirm by selecting **Yes**.

6. If the license activation is accessed, a confirmation window appears.

7. Press **Close** to exit **License Activation Wizard** and start the application.

Licensed Features

There are various editions of MyAssays Desktop as well as optional add-ons. The features available to you will depend on your license. To see which features are included in your license:

1. Launch **MyAssays Desktop Explorer**.

2. Select the **File** tab and select the **Information** option:

The availability of licensed features is marked with the following icons:

    for active features.

    for features not supported by your current license.

To learn more about additional features, which are not included in your existing MyAssays Desktop license, click on the **Upgrade License** button.

License Deactivation

If you need to transfer an activated license to another PC, it must first be deactivated from the PC where it is in use.

To deactivate MyAssays Desktop on your PC, launch the Deactivation Wizard as follows:

1. Launch MyAssays Desktop Explorer.

2. Set **File** tab and select **Information** section.

3. Click on the **Deactivate License** button.

Follow the steps of the **License Deactivation Wizard** as described below to deactivate the license:

Online Deactivation

If your PC is connected to internet:

1. Keep the **Use internet connection for deactivation** checkbox checked and press **Continue** to proceed with deactivation.

2. Check the checkbox to confirm deactivation and press **Deactivate**.

3. When prompted to grant **User Account Control** permission to access the licensing settings, confirm by selecting **Yes**.

4. On successful deactivation, a copy of the license serial code will be sent to the email address it is associated with, ready for activation on another system.

5. Press **Finish** to close the **License Deactivation Wizard**.

Offline Deactivation

If your PC is not connected to internet, it is still possible to deactivate MyAssays Desktop using a separate internet-connected device. In this case:

1. Uncheck the **Use internet connection for deactivation** checkbox and press **Continue** to proceed with deactivation.

2. Check the checkbox to confirm deactivation and press **Deactivate**.

3. When prompted to grant **User Account Control** permission to access the licensing settings, confirm by selecting **Yes**.

4. A QR code will appear; if you have a network-connected device (such as a smartphone or tablet) capable of reading this, then scan the QR code and follow the instructions. Alternatively, copy and paste the provided link and open it on a system with internet connection (perhaps by transferring via USB memory device).

5. On successful deactivation of your serial code, a confirmation message will appear in the browser and a copy of the license serial code will be sent to the email address it is associated with, ready for activation on another system.

6. Confirm that the email has been received in the **License Deactivation Wizard** and press **Finish**.

> After successful deactivation, the application will be closed and will not be usable until it is reactivated. The deactivated license will be immediately available for activation on another system.

Activate New License

If you have been provided with a new serial code to replace an existing license (perhaps as the result of a purchased upgrade or support extension), then it will be necessary to activate your new license to replace the existing one.

1. Launch **MyAssays Desktop Explorer**.

2. Select the **File** tab and select the **Information** option.

3. Select **Activate New License**.

4. Press **Yes** to confirm the removal of the existing license.

5. When prompted to grant **User Account Control** permission to access the licensing settings, confirm by selecting **Yes**.

6. Follow the steps of the **License Activation Wizard** as described in the License Activation section to activate new license.

7. On completion of the wizard, any new license updates will be applied to your installation and any new features or support extensions will be available.

Update Existing License

If new features have been added to your existing license (perhaps as the result of a purchased upgrade or support extension), then it will be necessary to update your activated license with the latest information.

1. Launch **MyAssays Desktop Explorer**.

2. Select the **File** tab and select the **Information** option.

3. Select **Update Existing License**.

4. If your PC is connected to the internet, then select **Update my license online in one click**. Otherwise, select **Get a unique link to my activation code** and activate using a separate device that is online (as described in License Activation).

5. When prompted to grant **User Account Control** permission to access the licensing settings, confirm by selecting **Yes**.

6. On completion of the wizard, any new license updates will be applied to your installation and any new features or support extensions will be available.

Quick Start

MyAssays Desktop – The Basics

This section introduces the basics of the MyAssays Desktop system by providing a summary of the key features and functions and where to find them.

This section is intended to be short to help equip you with the basics so that you can start to be productive with the software quickly and feel comfortable to explore further functions yourself.

Firstly, MyAssays Desktop provides two key applications to help organise and work with your data:

- MyAssays Desktop Explorer (MADE) – For arranging, locating, discovering, and launching your assay data
- MyAssays Analysis (MAA) – For performing the assay data analysis

The following subsections provide orientation details for working with both of these applications.

- Introducing MyAssays Desktop Explorer
- Introducing MyAssays Analysis

Introducing MyAssays Desktop Explorer

Orientation

The **Welcome** screen is displayed on launching MADE. Its main area provides short-cut links to:

- Previously used protocol and results files

- Online resources including video tutorials

- The protocols and results folders

- The **New Protocol** wizard (Pro versions only)

Along the left-hand side of the MADE window, you will see tabbed sections to conveniently access the folders that contain the various types of assay data.

Along the top of the MADE window, you will see the Ribbon providing various commands and functions for navigating and managing your data.

Here you can find the **Search MyAssays.com** control, which provides a convenient function to search the online database of preconfigured protocols. Simply enter any appropriate search term such as assay or kit name, product or kit manufacturer name to search for available downloadable content.

Assay Data

If you've just installed MyAssays Desktop, you will find some installed assay protocols and layouts under their respective tabs. The results and reports tabs will be empty until you've ran some analyses and created some reports.

As you run analyses and create various content, MyAssays Desktop will automatically store content in the associated folders.

Where is My Data?

It is worth noting that all of your MyAssays data is stored under a single parent folder. This makes backing up your data a simple process of copying all of the data in this folder. The default location of MyAssays data is the subfolder located under your Windows **Documents** folder.

You may prefer to store your data in a location different to this default location. For example, you may want to:

- Use a common location shared with your colleagues

- Store your data in another location

To access this location setting, select **File | Data**, the **Assay Data Folder Root** section reports the current setting. Use the **Select Folder Root** option to change this as required. This setting is stored with your Windows login account (so will be remembered the next time you log in).

You can of course access all of your data outside of MADE and in Windows Explorer, simply navigate to assay data folder root directory.

Introducing MyAssays Analysis

Orientation

The MAA application is displayed when launching an assay protocol or opening an existing assay results file.

The Overview tab is typically selected when launching MAA. This tab displays the launched assay name and any description. It might also include a link to online content (such as kit manufacturer's product insert).

Along the left-hand side of the MAA window, you will see various tabbed sections for providing and reviewing the data and results for the analysis. All assays will contain the tabs:

- **Overview**
- **Measurements** – From here you can provide and review your measurements (and any meta-data). Various subtabs are provided for viewing your data in different formats.
- **Results** – After performing calculations, the results of the analysis will appear in this tab. This will include the report. If the report is arranged as multiple worksheets, you will see worksheet tabs at the bottom of this area for navigating between worksheets. Also, depending on the analysis performed, you might see various subtabs appearing to the right of a **Report** subtab; these provide interactive controls for working with your data in various ways (such as flagging outliers from a **Standard Curve Fit**).

In addition to the **Overview**, **Measurements** and **Results** tabs, your assay protocol might be configured to include further tabs such as:

- **Microplate** – For defining, reviewing, and managing the sample layout.
- **Sample IDs** – For providing ID values for each unknown sample.
- **Dilution Factors** – For providing dilution factors for each unknown sample. These will be applied to the concentration value computed from the curve.

There might also be further tabs displayed for providing data specific to the assay.

Assay Protocol and Assay Results

When an assay protocol is launched, MAA operates in *protocol mode*. In protocol mode you can provide the data for your analyses and optionally save the settings to the protocol file.

Provide your analysis assay parameters along with your measurement data (or use the provided example data) and then press the **Calculate** button to create the first set of results.

For all assay protocols, typical example data is provided to help you familiarise yourself with the analysis when you don't have any data to hand.

Pressing the **Calculate** button:

- performs the calculations

- generates the report

- and performs any configured export operations

For the first set of calculations, some additional steps are automatically performed as MAA transitions from protocol to *results mode*:

- The results appear in the **Results** tab.

- An assay results file is created. This contains a copy of the assay protocol that was launched and all of the data provided (measurements and analysis parameters).

- The assay results file is automatically named and saved on the first calculation.

The file naming method can be changed under **File | Save | Results Naming**.

Results

The calculated results can be found under the **Results** tab. If the analysis generates charts, then these will also appear in subtabs under **Results**.

When there are multiple subtabs, the report can be found under the Report subtab. If the report is arranged into multiple worksheets (in **Sheet View**), then the sheet can be select using the sheet names appearing at the bottom of the **Report** subtab.

Page View or Sheet View

The report can be displayed as **Sheet View** or **Page View**; these settings are available through the Ribbon's **Report** tab.

When getting started, we suggest using the Sheet View so that your outputs are not confined by the limitations of a printed page. Use the Page View only if and when the layout of the printed page is important (For page-based reporting, you can define a reporting template to apply to your MyAssays Desktop output).

Save the Changes to the Protocol?

Before the first calculation is performed (protocol mode) any interactions performed apply to the launched protocol. If you perform the **Save** operation at this point, then the changes will

be saved, updating the original protocol file. Also, on the first set of calculations the settings are automatically saved, updating the protocol.

This workflow might be useful if you want your changes to apply to future runs of the protocol (so you don't need to repeat the configuration again next time). Alternatively, you might want the changes to only apply to this particular analysis.

For example, perhaps you are running a routine assay but have used a slightly different sample layout than you usually use. You might want to apply this change to the current run only, or you may decide to use this layout in future runs too.

To configure the behaviour that best suits you select **File | Save** and set the **Save changes to the protocol** option as you require. With this option set, any changes you make to the launched protocol are saved to the protocol. The changes are also save to the protocol on the first calculation (as the system transitions from protocol to results mode). Any further changes in results mode will then only apply to the results.

> This **Save changes to the protocol** setting is stored across sessions so it will apply to any other protocols you launch in the future.

Further Save Options

Under the **File | Save** area, you will see further self-explanatory save options:

- **Save**
- **Save Results As**
- **Save Protocol As**

Note that the **Save Protocol As** option can be used when viewing results to extract all of the current configuration settings to a new protocol file. This is useful to package up all of your recent changes made to a results file to a new protocol file.

Results Naming

Before any calculations are performed (protocol mode), the **Results Naming** option is available to enable you to specify how the created assay results file is named and stored. For example, you could name the results file using meta-data imported from the measurements data file and/or customize the subfolder where it is stored.

Save the changes to the protocol so any future runs of the protocol will follow the naming convention you have specified.

Calculate

Press the **Calculate** button when you are ready to calculate the results. In case this is not obvious, if you make any changes to the data or parameters you will be reminded to perform the calculations when you are ready.

> MAA does not perform calculations automatically on every setting change because this could impact on your experience of working with the software. With larger data sets (such as multiplex, kinetic or multiple plate assays) it might take a few seconds to perform the analysis.

Result Versions

Each time the **Calculate** button is pressed a new version of results is created. You can find the version controls including the full version history of the results in the **Home | Versions** Ribbon group.

To view a different version, use the **Current Version** selector. Changing a version will update the whole of the MAA application to reflect the newly selected version; all data views reflect the data and results for the selected version.

If you have made changes but not yet calculated the results of these changes, then the **Current Version** selector will display the new version number with the text **(pending)** following it. This is to indicate that the new results are pending calculation. Press the **Calculate** to create the new results version.

If you change version when results are pending calculation, then those changes will be lost (a warning message will prompt for confirmation before this action is completed).

> You cannot make changes to an earlier previously calculated version; any changes made will apply to a new result versions pending calculation.

In addition, you can remove unwanted versions using:

- **Purge** – Removes all versions except the one currently selected.
- **Rollback** – Removes all versions that follow the selected version.

Video Tutorials

Online videos are provided to help you get started and walk through features in detail:

Getting Started

- https://www.myassays.com/myassays-desktop-quick-start.video
- http://www.myassays.com/myassays-desktop-fundamentals.video
- http://www.myassays.com/download-and-use-protocols-from-myassays.video

Protocols

ELISA

- http://www.myassays.com/elisa-data-analysis-with-myassays-desktop.video
- http://www.myassays.com/multiple-plate-elisa-with-myassays-desktop.video

Dose–Response Curves

- https://www.myassays.com/dose-response-curves-for-ec50-with-myassays-desktop.video

Multiplex

- https://www.myassays.com/myassays-desktop-introducing-the-multiplex-protocol.video
- https://www.myassays.com/myassays-desktop-automatic-launching-of-multiplex-analysis.video

Importing Data

- http://www.myassays.com/import-measurement-data-from-excel-files-with-myassays-desktop.video
- http://www.myassays.com/import-measurement-data-from-multiple-excel-files-with-myassays-desktop.video

Excluding Outliers (Flagging)

- https://www.myassays.com/myassays-desktop-flagging-outliers.video

Worklists

- https://www.myassays.com/myassays-desktop-sample-ids-and-worklists.video

Export

- https://www.myassays.com/myassays-desktop-custom-export-for-lims.video

Folder Poll

- https://www.myassays.com/folder-poll-with-myassays-desktop.video

Inter-Assay Quality Control

- https://www.myassays.com/myassays-desktop-inter-assay-quality-control.video

Rack Layout Editor

- https://www.myassays.com/myassays-desktop-introducing-the-rack-layout-editor.video
- https://www.myassays.com/create-a-quantitative-radioimmunoassay-with-myassays-desktop-pro.video

Library Curve Fit

- https://www.myassays.com/re-use-standards-curve-with-a-library-curve-fit.video

Report Templates

- https://www.myassays.com/myassays-desktop-report-templates.video

Calculating Z Factor/Z Prime (Z')

- https://www.myassays.com/calculating-z-factor-z-prime-using-evaluations.video

Enhanced Security

- https://www.myassays.com/myassays-desktop-enhanced-security-authentication-methods-and-configuration.video
- https://www.myassays.com/myassays-desktop-pro-enhanced-security-with-envision.video

Working with MyAssays Desktop

Key Concepts

Throughout MyAssays Desktop, various concepts and terms are used to communicate and reference the different types of data related to assay data analysis. This section introduces these fundamental concepts.

Measurements

The measurements are the data points acquired from your scientific instrumentation (e.g., microplate reader).

The software supports measurements from any type of detection technology (such as absorbance, luminescence, fluorescence, counts, counts-per-minute), with data collected from each sample as:

- **Endpoint** – A single measurement value
- **Kinetic** – Values measured over time (data can be processed with or without time stamps)
- **Spectral** – Data measured at various multiple wavelengths
- **Multiplex** – Multiple analyte measurements from each sample

Measurement data is typically accompanied with other types of data; examples include date/-time of measurements, plate ID, and user ID. We refer to this data as meta data. The software can import and report this alongside your measurement data.

Import

The import process gets your measurement and any meta data into the software. The software supports a variety of methods including:

- Copy and paste
- Excel selection import
- Automated import for supported third-party proprietary formats

Protocol

An assay protocol defines all of the parameters necessary for performing the data analysis required for any assay. This includes:

- Expected measurement data (e.g., endpoint, kinetic, spectral, multiplex, etc.)
- Layout – How samples are arranged

- Calculations
- Report content and configurations

Also, the protocol can define:

- Import settings (exactly how and what data to import)
- Export settings (how to export the data, e.g., to Excel/PDF or a custom file such as CSV/TXT)
- Quality control settings (intra and inter-assay calculations and settings)

With MyAssays Desktop, the settings for a particular assay protocol are stored in an assay protocol file. These files use the **.assay-protocol** file extension.

> An assay protocol file is a self-contained file with no dependencies on any database, making it easy to transfer protocols between systems and to share protocols with colleagues.

Results

Each time an assay protocol is launched and results are calculated, an assay results file is automatically created. The assay results file contains the results of the analysis including the measurements and meta data and any data specific to the run. This could include:

- Sample IDs
- User-entered notes
- Calculation log
- Any changes to the configuration such as:

  - New sample layout
  - Calculation settings
  - Any flagged data (i.e., marked as outliers)

Assay result files use the **.assay-results** file extension.

> An assay results file also contains its own copy of the assay protocol. This is important so that any changes to the original assay protocol file will not impact the results already calculated. This structure also simplifies sharing data and transferring results between systems.

In addition, an assay results file can contain the whole version history of changes made to the analyses. Each time results are recalculated, a new set of results is created and stored within the same results file. The full-version history is available so you can go back to earlier versions of the results.

Reports

The report is the primary output of interest of the data analysis. A report is generated each time the assay results are calculated.

Typically, the report will contain a table listing the measurement data and results for each sample. The report might also contain charts and other content. MyAssays Desktop Pro provides a wealth of features and functions for tailoring the report content to your exact needs.

Report Views

Depending on your data or preferences there are two types of report, each with their own benefits:

- Sheet view
- Page view

Exports

MyAssays Desktop can export the report content to a variety of formats, including Excel/Word/PDF. In addition, content can be easily output to popular text-based formats such as CSV using preconfigured export methods. Beyond this, XSLT support is provided to enable you to export your content in any format to exactly match you output requirements.

Sample Layout

The sample layout (or simply layout) defines how the samples are arranged on the container or containers from where they were or will be measured.

> Here we use the general term "container" because the samples could be measured in a microplate or a rack of vials; in fact, any vessel for your samples is supported.

Getting Around

MyAssays Desktop provides two key applications to help organize and work with your data:

- MyAssays Desktop Explorer – For arranging, locating, discovering, and launching your assay data
- MyAssays Analysis – For performing the assay data analysis

MyAssays Desktop Explorer (MADE)

MADE is the application launched when you click the MyAssays Desktop Explorer icon installed on your desktop. This application is the starting point for all MyAssays Desktop functionality. From here you can:

- Search and download preconfigured assays from MyAssays.com
- Access useful videos and training guides
- Access technical support
- Manage your product license
- Access your assay data
- Create new assay protocols (requires Pro version)

MyAssays Desktop Analysis (MAA)

If you launch a protocol or open a results file, MAA opens. MAA provides the user interface for working with your assay data analysis. From here you can access all of the various options, including:

- Import/export data
- Provide assay parameters
- Enter sample IDs
- Generate report
- Calculate new results

MyAssays Desktop Explorer

MyAssays Desktop Explorer is the first application of MyAssays Desktop you interact with.

With MADE, you can:

- Find, download, and launch assay protocols from MyAssays.com database
- View existing assay layouts, results, and reports
- Run assay protocols and open results
- Create or request an assay protocol

> ⓘ   Assay protocols and results can also be launched directly from Windows Explorer

MADE Ribbon

Ribbon extends across the top of the main area of the MADE.

Ribbon Tabs:

Home

This tab consists of several groups for operating with data files inside MADE directly.

- **Navigation** includes three main options for moving around tabs, folders, and subfolders inside the application.
  As you continue working on your data analysis, you may prefer to divide and arrange your data in folders by its measurements or features. Therefore, the number of folders and subfolders in tabs will grow. You may use the **Back**, **Forward**, and **Up** navigation commands on **Ribbon** or **Quick Access Toolbar** to change your current location quickly.
- **Clipboard** provides better management of your stored data. You may prefer to rearrange the way your files or folders are stored by using **Paste**, **Cut**, and **Copy** commands. This way you can simply move around data files between various storage folders.

> ⓘ   Clipboard data is automatically deleted when the computer shuts down or restarts, so remember to save important information.

- **File** consists of two commands: **New**, which launches New Protocol Wizard, and **Open**, which launches data from MADE's tabs.
- **Search MyAssays.com** can be of the maximum use for finding and downloading specific preconfigured assays from the MyAssays online database. This tab consists of **Search textbox** and **Search button**.

Tools

This tab embodies **Utilities** you may find useful.

- **Analysis launcher** is a tool for advanced launching of the MAA.
- **Folder Poll Manager** launches MyAssays Folder Poll Manager.

Support

This tab provides connection between MyAssays and the end users.

Assistance group has a Request Protocol command, which you may find useful if the assay template you are looking for is not yet in the MyAssays database.

It also features a backstage view for additional use of application scenarios and configuration.

Backstage Area:

File Tab

File Tab gives you access to MADE's important tasks.

- **Information** contains details about your MyAssays Desktop personal use. This includes product version, current license details, and features.

> You may want to change some features of your MyAssays Desktop (i.e., activate, deactivate, or update license). The Product Licensing describes further details for these options.

- **View** defines whether the Measurements Tab is displayed on the Folder Side Bar and the Launch Assay Protocol with Options setting is displayed on preview pane in the Protocols tab.
- **Data** displays the storage path for persisting your assay data files and Welcome Screen's **Recent** feature options.

- **Drivers**

- **Exit** closes the application.

Quick Access Toolbar

Quick Access Toolbar contains shortcuts to commands and functions to make your work on MyAssays Desktop Explorer easier and personalized.

> You can Customize the Work Area to fully adjust it to your needs.

Application Button

Click on the logo in the upper left corner of the MADE to:

- **Restore** – Return window size to the previous one
- **Move** – Change windows on-screen' locations, if they are not full screen
- **Size** – Widen or narrow the window
- **Minimize** – Put the window in taskbar
- **Maximize** – Make the window full screen
- **Exit** – Close the program

Navigation Shortcuts

The other default commands of Quick Access Toolbar are the ones from **Navigation group**, namely **Back**, **Forward**, and **Up**, which are useful for switching between the tabs you currently work with quickly.

Advanced Options

You can set the Quick Access Toolbar position in this drop-down menu. Click on **Show Quick Access Toolbar Below/Above the Ribbon** (depending on its current location), **Minimize the Ribbon**, and change the Quick Access Toolbars view by adding or excluding some options in **More Commands** pop-up menu.

Folder Side Bar

MADE provides a hierarchical view of the organized data and presents its preview if it is available. MyAssays Desktop stores different types of data in separate subdirectories in the MyAssays **Data Folder Root**. These tabs point to actual Windows folders.

In MADE, you can access information from every subdirectory easily in the appropriate left side bar tab.

> MyAssays Desktop arranges your data automatically in correspondent folders so that you can easily manage, share, and back up your assets. You may prefer to customize how results files are named and where your data is stored.

Folder Side Bar Content

Tab	Contains Files
Search Results	Assay protocol titles that match search query
Protocols	Assay protocol files with .assay-protocol extension
Measurements	Measurement data files in **supported file formats** *(*.TXT, *.CSV, *.XML, *.DAT, *.XLS, *.XLSX, and others)*

Tab	Contains Files
Results	Assay results files with .assay-results extension
Layouts	Layout files with .assay-samplelayout extension
Reports	Report files with supported **report extension** *(*.DOC, *.PPT, *.XLS, *.DOCX, *.XLSX, *.PPTX, *.MHT, *.HTM, *.PDF)*

Selecting a tab displays its content in the main window.

File and Preview Panes

The File and Preview panes are populated with the content of the selected tab.

File Pane

A list of available data files displays on the left side of the active tab. It is a visualization of the folder that persists data, that was used to launch, or is a result of performing a particular assay analysis.

In the File Pane, files can be selected using normal Windows procedures. Right-click to display the default Windows Explorer context-sensitive pop-up menu. Relevant MADE Ribbon options are available when a single file is selected.

Sort and View

The default view of content is **Details**. To change it, right-click on active tab's empty space > **View** > select preferable view from the drop-down list.

Files are sorted by name by default. To change the file sorting order:

- Right-click on the empty space of the active tab > **Arrange Icons By** > choose preferable option form the drop-down list.

OR

- If File List is displayed in default view, click on the column headings **Name**, **Size**, and **Date Modified**.

Subfolders

Tabs may contain subfolders (subdirectories). Creating subfolders in tabs comes in handy when working with big data sets or with multiple versions of analyses. All subfolders will be of the same type as their primary parent. To create a new subfolder, right-click in the File Pane and select **New** > **Folder**.

The File Pane may be navigated using the relevant buttons on the MADE Ribbon.

Preview Pane

Select a single file from the list to see its preview and available actions on the right side of the active window.

The contents of the preview pane depend on the type of file selected:

Folder Tab	Preview Pane Contents
Search Results Protocols	A summary of assay protocols, including the name, the instrument settings, the position used and layout, and any configured transformations
Measurements	Precanned and custom raw data files
Results	Two assets: • **Overview** with the assay protocol summary • **Latest Report** with the latest report of the results
Layouts	A microplate control showing each plate layout in the layout
Reports	The preview of the PDF reports

Welcome Screen

This is the screen that displays as you launch MADE. It contains some useful options for a proficient start. Get acquainted with MyAssays Desktop Explorer's basic functions and begin your work at full speed.

The left pane of Welcome screen consists of:

- **Start** – Shortcuts to basic data analysis options, particularly New Protocol..., Launch Protocol..., and Open Results...

- **Recent.** – A specified number of assay protocols and results you have worked with last (the particular title will be launched in MAA upon selection)

> You can customize a number of entries in the **Recent** list or clear the file history in Data options.

The right pane comprises of two subpanes:

- Upper subpane **Discover MyAssays Desktop** displays links to a detailed review of MyAssays tools and the desktop application specifically. Visit **MyAssays Home Page** or **What's New in MyAssays Desktop?** and get to know latest information about desktop version.

- Lower subpane contains:

  - **Product Videos** – Links to video materials that will help you familiarize yourself with MyAssays Desktop and its components

- **Support** – A link to the **MyAssays Support** contact form and the Request an Assay Protocol dialog box

Recent Used Files Feature

The number of files displayed under the **Recent** subpane of the Welcome Screen may vary from 0 to 20.

To change the number of Recent files, type it in the corresponding box or use the arrows to adjust it.

Press the **Clear Recently Used File list** button if you want to erase the history.

Search Results Tab

This tab displays on the Folder Side Bar after you submit a Search MyAssays.Com.

It displays a File List and Preview of the assay protocols that match search query.

When this tab is active, multiple search results options are available.

Search MyAssays.com

How to: Find an Assay Protocol from MyAssays.com Knowledge Base

First, in MADE, enter an assay title or keyword (i.e., Cortisol, Human, ELISA):

- In the MADE Menu Bar, **Search MyAssays.com** panel

OR

- Use a keyboard shortcut (**CTRL + E**), which automatically redirects you to a search textbox

Next, press the **Search** button or **Enter** on keyboard.

When you have performed a search, the Search Results tab is displayed on Folder Side Bar.

Using the Search Results

Upon performing a search, you will see the list of assay protocols that contain a keyword you have entered.

To specify the search results and ensure finding the protocol you're looking for, each protocol contains a detailed summary. To see further information on a particular position, single-click on its title. Some of the assays contain Kit Booklet from the manufacturer. To read it, click on **View Assay PDF** link under the assay summary.

For every assay protocol in the list, the following actions are available on the Preview pane:

- **Download and Launch** – Persists a single assay protocol to your Protocols tab and automatically launches it in MAA.

> You can also double-click on a file from the list to download it to your PC and launch it in MAA automatically.

- **Download** – Stores a single or multiple assay protocols to your Protocols tab.

> If you select multiple items from the list, only this option is available and displayed. To select multiple protocols from the search results, use the **(Ctrl+Shift+Arrow Keys)** keyboard shortcut and **Ctrl+A** to select all search results.

- **Launch Assay Protocol** – Appears if you have downloaded a particular assay protocol previously. The above actions are then not displayed. To enable re-downloading, click on the **Click here** hyperlink in the message below **Visit on MyAssays.Com**.
- **Visit on MyAssays.Com** – Directs you to the online MyAssays.Com webpage of the selected assay protocol.

No Results Found

If the assay you are looking for is not yet in the MyAssays database, an **Information** dialog box appears.

Click **Yes** to proceed to a Request Protocol form.

Search + Download from MyAssays.com

Download and use preconfigured assay protocols from the MyAssays.com knowledge base on your PC.

Submit a Search and explore search results options of the constantly evolving protocols database.

Protocols Tab

The **Protocols Tab** displays a File List and Preview of the assay protocols that are persisted to your PC.

> Only data files with an .assay-protocol or .apr extension are visible through this folder.

Upon selecting a particular assay title from the list, you will see its summary on the Preview pane and, if it is available, a detailed Booklet Kit from the kit manufacturer.

In the **Protocols Tab**, you can:

How to: Create New Assay Protocol

In the MADE Welcome Screen, click on **New Protocol...**.

OR

In the Folder Side Bar, select the **Protocols** tab and click on the **New** button on the MADE Ribbon.

When this option is selected, a New Protocol Wizard will be displayed.

How to: Launch Assay Protocol File

Navigate to the Protocols tab, select a single assay result file to open, and do one of the following:

- double-click on its title
- right-click on the title and choose **Run with MyAssays Desktop Analysis**
- click on the **Launch Assay Protocol** button in the lower right corner of the Preview pane.

By doing this, you will launch the file in MAA.

> 📝 You can also launch an assay protocol directly from Search Results.

Launch Assay Protocol with Options

Launch an assay protocol with command-line options for more control over the analysis. This feature will open the Launcher tool with the selected protocol.

If you press the **Launch Assay Protocol with Options** button, the dialog window with a visual representation of supported command-line options will appear.

How to: Launch Assay Protocol with Command Line Options

1. Launch MADE.
2. Select the **File** tab of MADE Ribbon.
3. Select the **View** screen.
4. Check the **Show Launch Assay Protocol with Options button** checkbox.

> ℹ️ The **Show Launch Assay Protocol with Options button** state (checked/unchecked) persists between sessions.

If it is checked, a corresponding option appears on the preview pane of the Protocols tab upon protocol file selection.

How to: Request Assay Protocol

Launch MADE and do one of the following:

- Click on the **Request an Assay protocol** on a **Support** subpane of the Welcome Screen.

  OR

- On the MADE Ribbon, select the **Support** tab and press the **Request an Assay protocol** button.

When this option is selected, a **Request Protocol** dialog box will appear. Fill in the fields to specify what kind of an assay you want to be included in the MyAssays knowledge base.

Please mention any information that can be of use in **Other Significant** field.

You can and are welcome to attach any applicable files, such as Example Raw Data, Kit Insert PDF, and Other files (in zipped files).

After you have provided details of an assay, press the **Submit Request** button to send your requirements to MyAssays Support.

Measurements Tab

How to: View the Measurements Tab

1. Launch MADE.
2. Select the **File** tab of the MADE Ribbon.
3. Select the **View** screen.
4. Check the **View Measurements** checkbox.

> The **View Measurements** state (checked/unchecked) persists between sessions.

If it is checked, an additional tab is displayed on the Folder Side Bar.

The measurements tab contains a default **Examples** folder with a 12x8 Endpoint.txt file. For each assay, a text file containing your measurement settings data can be uploaded and pro-cessed for analysis. To upload a file, paste it in the **Measurements** folder in your MyAssays Folder Root.

The New protocol wizard can automatically identify and import the relevant readings from a file saved in many supported file formats, including TXT, CSV, or other proprietary formats.

Visit MyAssays Supported File Formats (http://www.myassays.com/supported-file-format-s.html) to see list of supported measurements data formats.

Results Tab

The **Results Tab** displays a File List and Preview of the results of previously run data analyses. Select a single Assay Results file from the File List to see the Preview of the report.

Results files generate when you have launched a particular assay protocol and performed a data analysis. It is empty on your first time launching MADE and will fill as you work with the application. By default, these files are named after a protocol, but you can change the name (i.e., after incoming measurements, current date, etc.).

> ℹ️ Only assay results data files with an .assay-results or .ars extension are visible through this folder.

When you select a title, two information insets appear on the Preview pane. The **Overview** shows an assay summary and may contain a link to the Booklet Kit from manufacturer. The **Latest Report** contains the report of the selected Assay Results file.

> 📝 To enlarge the Preview view, hold down **Ctrl** button and push the mouse scroll wheel upward.

In the **Results Tab**, you can Open Results in MAA.

Open Existing Results

How to: Launch Assay Results File

Navigate to the Results tab, select a single assay result file to open, and do one of the following:

- double-click on its title
- check the **Open** icon on the MADE Ribbons **File** group
- check the **Open** icon in **Latest Report** inset of the Preview pane

By doing this you will launch the file in MAA.

Layouts Tab

The Layouts tab displays a File List and Preview of the assay layouts persisted to your PC. Vice versa, only files persisted to this tab may be used in running assays by MyAssays Desktop Analysis. Select a single Assay Layout file to view it on the Preview pane.

> **ⓘ** Only data files with an .assay-samplelayout or .mlo extension are visible through this folder.

Assays in MyAssays Desktop Analysis are initially set up to use a default layout for your samples. You will most likely prefer to use your own sample layouts in your data analyses.

Use the Plate Layout Editor to edit existing or create new layouts.

Reports Tab

All assay result data exported into a report file is stored to this folder. Double-click on a title to open it in a relevant application.

Customize the Work Area

You can configure **Work Area** to fully match your preferences and enhance your experience. Learning about its potential may be useful for taking full advantage of the features, tools, and options of the application.

Customizing the Quick Access Toolbar

The Quick Access Toolbar displays tools that you have bookmarked. For making use of its full potential, you may prefer to rearrange and manage these tools.

Location

You may prefer the Quick Access Toolbar to be displayed above or below the Ribbon. Right-click on any spot of the Ribbon and hit on the **Show Quick Access Toolbar Below the Ribbon** command. If the toolbar is already below, you can configure it to be vice versa.

Adding or Excluding Tools

To add a command to the Quick Access Toolbar, choose the one you want from the **Home**, **Tools**, or **Support** tab and right-click on it. In the drop-down menu that will appear, press the **Add to Quick Access Toolbar** command. Upon doing that, the command will be displayed in the Quick Access Toolbar.

To exclude a command from the Quick Access Toolbar, right-click on it and click on **Remove from Quick Access Toolbar**.

Advanced Customizing

You may consider customizing the Quick Access Toolbar to fully match your preferences. To call up a **Customize Quick Access Toolbar** options menu, do one of the following:

• Right-click anywhere on the Ribbon and click on **Customize Quick Access Toolbar** on the context menu.

• Click on the **Advanced** option on the Quick Access Toolbar and click on the **More Commands** command.

Upon doing this, a dialog box will appear. Select tools you prefer and add or remove them from the list.

You can add or remove commands from a list or from a specific tab. You can also determine their on-screen order by pressing the arrows on the right side of the **Quick Access Toolbar** list.

When you have finished customizing toolbar, press the **OK** button to save changes or **Cancel** to undo them.

To return the toolbar to its default configuration, click on **Reset** in the **Customize Quick Access Toolbar** dialog box.

Customizing the Ribbon

The Ribbon is designed to make finding and working with commands convenient and easy. You may consider the capability to personalize and customize the Ribbon in the way you want useful. With this feature, you can change the default Ribbon and create custom tabs and groups with your preferred commands.

To customize the Ribbon, right-click anywhere on the Ribbon and then choose the **Customize Quick Access Toolbar** context menu to bring out the dialog box. Select the **Customize Ribbon** option and personalize your Ribbon view for your convenience.

Minimize Ribbon

This option may be useful to expand your on-screen view of the application. If the **Ribbon** is minimized, tabs appear on click and hide when you click on any other spot of the screen. To do this, right-click on any spot on the Ribbon and check **Minimize Ribbon** in the drop-down list. To undo minimizing, repeat these actions and uncheck the command.

Plate Layout Editor

Most assays allow you to specify your own layout for processing and distinguishing various types of measurement data. The editor is used to create new layouts or modify existing layouts.

You can freely position your samples and change the number of sample groups providing the requirements of the assay are met. Certain assays allow you to provide measurement data ran across multiple plates.

In addition to being used to define a layout before data is collected, the editor can be used after data collection to correct for preparation errors.

> If your samples are measured in tubes or vials then use the Rack Layout Editor to define the sample arrangement.

Using the Plate Layout Editor

The Plate Layout Editor is displayed when you press the **Create** button to create a new layout or **Edit** to edit an existing layout on the Layout Selection pane of MAA **Microplate** tab. You can also create a new layout as a step of the New Protocol Wizard.

Editor Flexibility

The actual flexibility available in your custom layout depends on an assay's requirements. Most assays require a certain number of positions of a particular type to be defined (e.g., **Blank** or **Standard** groups). In most cases, however, you may setup the layout as you prefer providing your layout meets an assay's requirements.

Once your new layout is created, you can select it to be applied to future calculations of an assay. By making calculations or saving settings, your new layout is saved under MyAssays/Layouts in your MyAssays folder root.

Creating Multiple Plate Layouts

A layout can be defined for each real-world physical container. The Plate Layout Editor can be used to define layouts across multiple containers. The group numbering of samples is contiguous and sequential for each type.

There are a number of different ways that standards and samples of an assay might be arranged across multiple plates. Plate Layout Editor allows you to specify for each plate:

1. How samples are arranged

2. Which standards measurements should be used to construct a standard curve

3. Which curve to use to calculate concentrations from

Plate Layout Editor Settings

With the following options, you can specify how samples are arranged on a plate.

There are two modes for specifying an assay layout:

- **Erase** – For removing samples that you are not measuring
- **Fill** – For defining the layout by selecting areas to fill
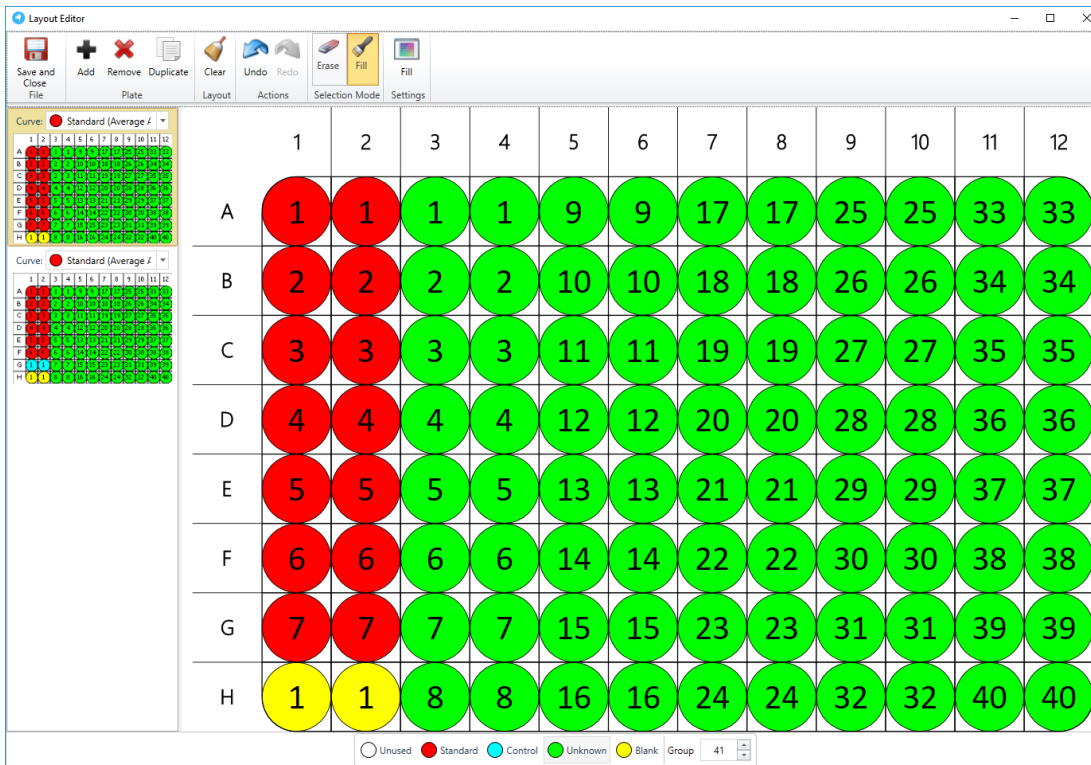
Erase Edit Mode

Erase mode allows you to remove samples that you are not measuring (i.e., mark them **Unused**).

To remove a position:

1. Select the **Erase** icon on the toolbar.
2. Move the mouse pointer over a defined position.
3. Left-click to remove all positions of this sample group.

> If the sample group contains more positions than you want to remove, select **Unused** type on the type and group selector and fill the position(s) with it.

Sample numbering will be adjusted to ensure that the group numbering remains contiguous.

Fill Edit Mode

Fill Edit mode allows greater flexibility in layout arrangement. To enter **Fill Edit** mode, press the **Fill** button. The **Fill Settings** button will appear, enabling you to specify how the positions you select are filled.

Also, the type and group selector can be used to select the next group to fill with your chosen settings.

To change the sample at a position, simply left-click your mouse pointer on the position. To fill an area, left-click at the first position and hold and drag to the last position. The **Fill Settings** dialog window appears, allowing you to change how this fill operates. You can specify the fill direction and number of positions for your selected group. The following fill options are available:

- **By Row**
- **By Column**
- **Rectangle Mode**

If **Rectangle Mode** is selected, you can choose whether the replicates fill **By Row** or **By Column**. You can specify the number of positions per group using the entry box.

> Group numbering must start at 1 and be contiguous for each sample type.

Undo/Redo

Use these buttons to cancel the last change or re-apply it.

Save + Close

When you are satisfied with your changes, press the **Save + Close** button. This will save the new layout to your Layouts folder and make it available to relevant assay analyses.

Multiple Plate Options

If you have selected the Multiple Plate option or are editing a multiple plate layout, the Plate Layout Editor's functionality expands with options applicable only to multiple plate layouts. Also, you can switch between plates and choose curve-fit generation properties.

Options

The toolbar for multiple plate expands with following options:

- **Add Plate** – Adds an empty plate
- **Remove Plate** – Removes a selected plate
- **Duplicate plate** – Copies a selected plate with all defined changes

Curve Selection

Curve selection describes how the multiple layout is arranged and behaves (i.e., defines if and how a curve can be selected).

For multiplex assays, only single layouts can be applied.

Multiple Plate Group Numbering

When defining the layout (in the Plate Layout Editor), the numbering for each sample group on every plate should always start at 1.

This might seem counter-intuitive, however, this is intentional to make things easier when working with multiple plates. With this approach, if you decide to change the number of samples on the first plate you do not need to update the numbering on all plates that follow. For example, being able to refer to, for example, **Control1** to reference all **Control1** replicates is much simpler than having to remember or decode the measurement data.

To handle multiple plate assay operations efficiently, you can refer to sample group either on certain container(s) precisely or with regard to whole measured data. This is described further in the Sample References in Multiple Container Assays.

When the data analysis is performed, the software will apply the group numbers across plates contiguously. This assures the results (for non-calibrator groups types) are presented sequentially and each sample group has a unique number. For example, if the layouts for plate 1 and plate 2 are both defined with **Unknown1-Unknown10**, when the analysis is performed, plate 1 will include **Unknowns1-10** and plate 2 would include **Unknowns11-20**.

Multiple Plate Standard/Calibrator

With **Standard/Calibrator (Control)** sample types, it does not make sense for numbering to be sequential across plates as this would imply that the data for a single curve is spanned across plates. Also, it is possible to configure standard replicates across plates (e.g., every plate has a **Standard1**, etc.). Therefore, for these sample types, numbering always starts at 1 for each plate.

> The number of plates defined in the layout may or may not correspond to the actual number of plates measured. See Trimming and Padding subsection.

Multiple Plate Rules

The number of plates defined in the layout may or may not be equivalent to the number of data containers measured. For example, the layout might only define one container whereas 50 containers' worth of data are provided. Equally, there may be less containers measured than there are defined in the layout. In these cases MAA will rely on the following rules:

Trimming

The layout contains more plates than data containers.

When there are more plates defined in the layout than containers measured, for each data container the layout with the same index will be applied. The extra layouts will be ignored.

Example: *Let's assume that we have a layout made up of 3 plates and 2 containers of data. The analysis will be performed in the following way: **Plate1** on **Container1**, **Plate2** on **Container2**. The **Plate3** will not be used for analysis.*

Padding

The layout contains more plates than data containers.

When there are more containers measured than there are defined in the layout, the last defined layout plate will be reused. For each data container will be applied the layout with the same index. The excessive measurements data will be analyzed using the layout with the last index.

 Example: *Let's assume that we have layout made up of 3 plates and 5 containers of data. The analysis will be performed in the following way: **Plate1** on **Container1**, **Plate2** on **Container2**, **Plate3** on **Container3**, **Plate3** on **Container4**, and **Plate3** on **Container5**.*

> The data analysis evaluation results are presented sequentially, with a unique number for each sample group.

Multiple Plate Analysis

The multiple plate features greatly simplify processing measurement data when your samples span more than one plate.

For multiple plate layouts, the layout can contain one or more plates with different positioning (and curve-fitting options) of the samples on each. Each layout in a collection has the same dimensions (width and height). For example, all containers in a collection might be 12x8.

> While different layouts may be applied on different containers, the same layout is applied on all matrices of a specific container unconditionally.

Plate Layout Editor allows you to freely define how your samples are arranged on each plate. For example, the following multiple plate layout uses a different sample arrangement on each plate:

For quantitative assays, your standards might be arranged on every plate in the batch or you may decide to measure standards only on some of the plates in the batch. You can specify how the standard curve is generated and which curve to use to calculate the concentrations for each plate.

Plate 1

Plate 2

Plate 3

Partial Standards

When running an assay across multiple plates, you might decide that it is not necessary to measure the full range of standards on every plate. Instead you could read the full range of standards on only one plate and measure a subset of the standards on other plates, then visually compare the curve fits to ensure they are usable. This allows you to:

- Save costs on standards
- Save preparation time
- Measure more samples per plate

MyAssays makes this process easy by allowing you to define layouts with partial standard groups. We use this term to define a layout that includes the full range of standards on one or

more plates of the layout with other plates featuring only a subset of the full range of stand-ards.

Multiple Plate Layout Curve Fit Settings

The Plate Layout Editor provides a method for selecting the curve to use for each plate. The lay-out configuration defines how the standard groups are arranged on each plate. This can be the curve generated from either:

- the average (of calibrator groups measured across separate plates)
- a specific plate

Example

With a layout that comprises of 3 plates, the following options for creating a curve are avail-able:

- **Standard** (Average Across Plates)
- **Standard** (Plate 1)
- **Standard** (Plate 2)
- **Standard** (Plate 3)

For more details about curve fitting on multiple plate assays, see Multiple Plate Standard Curve Fit Analysis.

Rack Layout Editor

This section provides information about creating and editing custom rack layouts with the Rack Layout Editor.

The Rack Layout Editor is used to define the sequence of samples when assay data is measured from tubes or vials. The editor is used to create new layouts or modify existing layouts.

Racks are used to represent the sample data on-screen. The editor allows you to define and arrange the sequence of your samples.

In addition to being used to define a layout before data is collected, the editor can be used after data collection to correct for preparation errors.

> 📝 If your samples are measured on a microplate then use the Plate Layout Editor to define the sample arrangement.

Rack Layout

With rack layouts, the number of samples that can be measured in each rack is fixed, this is the rack width and it is the same for all racks measured in an assay. Each rack is measured from left-to-right. For example, the following layout illustrates the racks used to measure 26 samples with each rack containing up to 10 samples.

With rack layouts the actual number of samples may vary at each run. To simplify the configuration of rack layouts, a sequence based editor is used to define the sequence of samples to measured. This is then converted to a layout to apply for samples that are actually read on each run of the assay.

Using the Rack Layout Editor

The Rack Layout Editor is displayed when you press the **Create** button to create a new layout or **Edit** to edit an existing layout on the Layout Selection pane of MAA **Layout (Racks)** tab. You can also create a new layout as a step of the New Protocol Wizard.
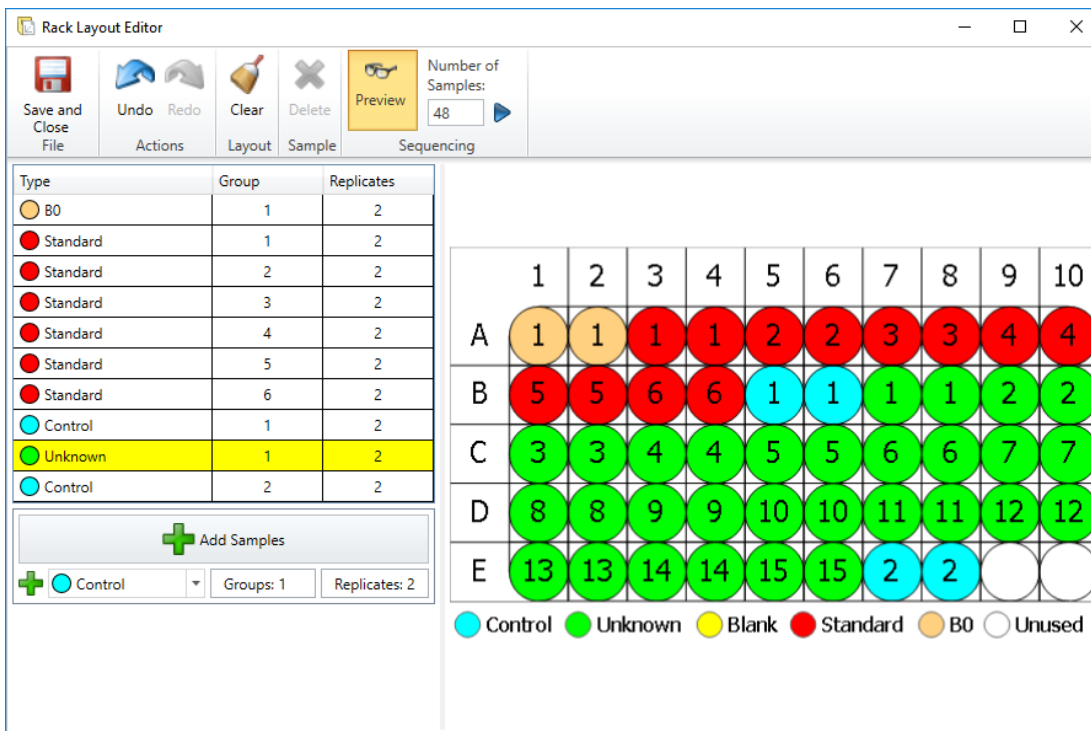
Rack Layout Editor Settings

The Rack Layout Editor is arranged into two panes:

- **Definition** – For defining the sequence of samples to measure and for specifying any dynamic section

- **Preview** – A visual representation of the rack layout using sample sequence with a specified number of samples to measure

Initially the Preview pane will not be displayed. It can be displayed by pressing the **Preview** button.

For example, here we can see the Rack Layout Editor showing a sequence definition of B0, Standards, Controls and Unknowns. The left-hand side shows the definition with the right-hand side showing the preview for 48 samples:

Sequence Definition

A rack assay might be fixed or dynamic:

- **Fixed** – The resulting sequence will contain only the samples specified in the definition pane

- **Dynamic** – The sample layout adjusts in accordance of number of positions measured

A fixed layout is only useful if you plan to run the exact same number of samples each time the assay is used.

A dynamic layout allows you to specify a common layout that can automatically be applied to different runs of the assay where a different number of samples is measured in each case. With a dynamic layout one or more adjacent sample groups are marked as dynamic; these appear highlighted in yellow in the definition pane. The resulting sequence will contain n copies of the groups in the sample (where n depends on the total number of samples). More...

Definition Pane

In this pane, you specify the order in which your samples will be measured.

Add Sample(s) settings

The Add Sample(s) controls are located on the bottom of the Sample Sequence Grid and are used to specify the type and number of samples to add. It comprises of:

- **Type Selector** – Select the sample type from a drop-down list
- **Number of Groups** – The number of sample groups to add
- **Number of Replicates** – The number of replicates in the sample group

After specifying the options, press the **Add Sample(s)** button. The ordering in which samples are added reflects the order in which they will be added to the layout.

In case the grid already contains any samples specified, the **Add Sample(s)** expands with a drop-down menu of insertion settings. Select a sample on the grid and select **Before Selection** or **After Selection** option to specify insertion of a new sample. To add a sample at the end of the list, select the **Append** option.

> Group numbering must start at 1 and be contiguous for each sample type.

Sample Sequence Grid

While the width of the racks is fixed (as defined by protocol settings), the actual number of samples may vary at each run. Thus, you can define the sequence in which the samples will be added by specifying state of sample sections:

- **Fixed** – The resulting sequence will contain these samples in the exact number as they are defined in the section
- **Dynamic** – Adjusts in accordance of number of positions measured. The resulting sequence will contain n copies of the groups in the sample (where n depends on the total number of samples). More...

The defined sequence of groups is listed in the grid, where each row corresponds to a sample. The attributes of each sample are editable.

Set Dynamic

One or more adjacent rows can be set as dynamic. Select one or more adjacent rows in the table and press the **Set Dynamic** button. Rows in the dynamic selection are highlighted with a yellow background. To return the row to the fixed state, select it and press **Remove Dynamic** button.

> To select multiple rows to mark as dynamic, press and hold the Shift key when selecting rows with the mouse.

Undo/Redo

Use these buttons to cancel the last change or re-apply it.

Clear

This button removes all **Sample** configurations (i.e. remove all Fixed and Dynamic sections)

Delete

This button removes selected sample(s) from the table. Sample group numbering may be prompted to be adjusted to ensure that it remains contiguous.

Save + Close

When you are satisfied with your changes, press the **Save + Close** button. This will save the new layout to your Layouts folder and make it available to relevant assay analyses.

Preview Pane

The Preview Pane displays the layout arrangement sequenced using the specified number of samples. To display the preview settings, press the **Preview** button. The **Number of Samples:** edit-box will appear, enabling you to specify how many positions to display on the preview. To apply this setting press the ▶ icon to the right of the edit box.

Racks

When working with assays that measure samples in tubes or vials (rather than microplates), MyAssays Desktop presents the measurement data onscreen using "racks of samples".

Each rack is represented on screen with a fixed number of samples and are filled left-to-right. For example, the following graphic illustrates a rack-layout for 26 measured samples. These samples are represented across 3 racks.

The rack representation provides a convenient method for presenting data measured from multiple samples in a 2D view.

Rack Width

Within an assay a fixed rack width is used. If your reader or workflow works with racks, then use a rack width that corresponds to the actual number of tubes or vials per rack.

> The rack width is the number of samples measured in each rack.

I'm Not Working With Racks

If your reader or workflow does not use racks but instead reads any number of tubes or vials, then adopt a rack width that is a practical size to present your data onscreen. A rack width of 10 is a good choice, but as you work with the software you may prefer a different width.

Dynamic Layouts

When working with rack based assays, the actual number of samples (tubes/vials) to measure may vary considerably between assay runs. Typically, there will be samples that are always read on each assay run (such as standards, controls, blanks) however there will also be sample types (such as unknowns) that will be measured in different volumes on each run of the assay.

For flexibility and ease-of-use, MyAssays Desktop supports the definition of "dynamic" layouts. The main benefit of working with a dynamic layout is that you do not need to redefine the sample layout each time the assay is ran. The sample layout is automatically inferred from the actual number of samples measured and the dynamic sample layout definition.

A dynamic layout defines one or more sample groups as dynamic. Each time the assay is ran, the number of samples allocated for the dynamic section on the layout will be automatically adjusted based on the total number of samples measured.

Example 1:

In this example, the **Unknown** group (in duplicate) is marked as dynamic. This means that the number of **Unknowns** on the layout will depend on the number of samples measured. This particular layout will ensure that a **Control** group is read before and after the **Unknowns**.

Type	Group	Replicates
B0	1	2
Standard	1	2
Standard	2	2
Standard	3	2
Standard	4	2
Standard	5	2
Standard	6	2
Control	1	2
Unknown	1	2
Control	2	2

For example, with 26 samples, the resulting layout comprises of 4 automatically assigned **Unknown** groups:

Using the same layout measured with 40 samples, 11 **Unknown** groups are automatically assigned.

Example 2:

In this example, the dynamic section defines a singlet **Blank** followed by an **Unknown** in duplicate.

Type	Group	Replicates
○ B0	1	2
● Standard	1	2
● Standard	2	2
● Standard	3	2
● Standard	4	2
● Standard	5	2
● Standard	6	2
○ Blank	1	1
● Unknown	1	2

To select multiple rows to mark as dynamic, press and hold the Shift key when selecting rows with the mouse.

For example, with 26 samples, the resulting layout comprises of 4 automatically assigned **Unknown** groups each with an associated blank.

Using the same layout measured with 38 samples, 8 **Unknown** groups each with an associated blank are automatically assigned.

Analysis

Versioned Analysis

Overview

When you launch an assay protocol and press the **Calculate** button, an assay-results file is created with the first calculated version of results (**Version 1**). A result version contains all of the settings and results for a specific **Calculation**. You can select to view a different version of results at any point.

> **ⓘ** Each time the **Calculate** button is pressed, a new version of results is created and stored to the .assay-results.

Changing Versions

When there is more than one version of results available, you can freely switch between them.

How to: Switch Between Versions of the Results

1. Open a results file.

2. Hover to a **Versions Group** on the **Home** tab of the **MAA Ribbon**.

3. Select the Version you need from **Current version** drop-down list.

Changing the version of results updates all settings and results views to display the data relevant for the selected results view. Any changes made to the currently selected version will be saved as a new version.

> **ⓘ** Pressing **Calculate** will create a new version (with version number 1+ the current highest version number).

For example, if there are 3 versions of results and you go back to **Version 1** and make changes, then **Version 4 (pending)** is created. If you then press **Calculate**, the **Version 4** will be stored. This is the new set of results based on **Version 1** with those changes made to **Version 1** (as **Version 4**).

Purge Versions and Rollback

Purge Versions

The Purge operation will delete all version data except the current selected version. This setting is available if your assay contains more than one version.

Rollback

The Rollback operation will delete all data that follows the selected version. It is active if the current version is not the newest version of the results.

> ℹ The **Purge Versions** and **Rollback** operations cannot be undone; however, the changes are not committed to the file until the **Save** operation is performed.

Uncalculated Version

When you make changes to a results file after the last set of calculations and before the **Calculate** button is pressed again, the **Version {n} (pending)** label is displayed in the **Current Version** selector.

Unsaved Changes

If you made changes without calculating them (i.e., a pending version) and want to exit MAA, you will be advised to **Save these settings** on exit. These settings will be saved to the assay-results file as new uncalculated version and also to the assay protocol if that option is selected.

This also applies to assay protocol with no calculated changes.

> 📝 When the assay-results file with uncalculated changes is opened in MAA, the first set of results will be calculated automatically.

Save Options

You can save data you are working with as an:

- **Assay protocol** – To save only the settings representing the assay analysis to be performed
- **Assay results** – To save the settings representing the assay analysis and versioned results of its performance

Save Changes To Protocol

This checkbox appears if you have launched MAA with an assay protocol file. It indicates whether the changes (i.e., layout to use, measurements data, Sample IDs) should be stored in the assay-protocol file, overwriting its default properties.

> ℹ The **Save Changes to Protocol** state (checked/unchecked) persists between sessions.

Save Commands

- **Save**

  To simplify the different types of save options, there is one master **Save** command.

  The master **Save** command is accessible from:

  - Quick Access Toolbar
  - File | Save BSA

  This option saves results of the analysis to the directory it was launched from. It also optionally saves changes to a protocol, if the **Save Changes to Protocol** checkbox is checked.

- **Save Results As**

  With this command you can save the assay results with all changes to a new .assay-results file. When you press the **Save Results As** button, a dialog box appears. You can provide a new file name and a location for it to be stored in.

- **Save Protocol As**

  With this command you can save the modified assay protocol to a new .assay-protocol file. When you press the **Save Protocol As** button, a dialog box appears. You can provide a new file name and a location for it to be stored in.

Naming Results

> This setting is only available if MAA was launched with an assay-protocol file.

With this you can specify a custom target path to save a file to and how save filenames are automatically generated (e.g., to arrange all results into subfolders based on the protocol name).

When the **Results Naming** button is pressed, a dialog window with the following options appears:

1. **Default** (automatically named and stored in Results folder).
   This means a default file naming convention is used and applied by default; this default method is based on the protocol name of the related assay protocol. **Example**: *For "ELISA.assay-protocol," the first time it is ran a results file "ELISA (1).assay-results" is created in the Results folder*.

2. **Custom**
   You can customize the naming method and target path manually, by using macros, or by combining these approaches.

How to: Set Custom Results Naming

If you choose the **Custom** option in the **Results Naming** dialog window, the following text-boxes will appear:

Folder. With this you can provide a custom path to store your files in. The options are:

- Provide a full path to the folder in the textbox (manually or from clipboard).

- Click the **Browse...** button to provide the full path to the folder in **Browse for Folder** tree view. Press **OK** to save the path or **Cancel** to close the dialog box without any updates.

- Click **Manage...** to insert any number of supported macros from the list. Press **Update** to save changes or **Discard** to cancel.

Also, you can combine these options. **Example**: *Provide path to a parent folder via the* **Browse...** *option and use the <ProtocolName><CurrentDateShort> macros from* **Manage...** *to specify folder name.*

Filename. This specifies the title of assay results files. The options are:

- Type a name in the textbox.

- Click **Manage...** to insert any number of supported macros from the list. Press **Update** to save changes or **Discard** to cancel.

Also, you can combine these options. **Example**: *type in "Experimental results for" and use <ProtocolName><CurrentDateShort> macros from* **Manage...** *to specify the analysis type and date it was performed.*

> ℹ️ If the **Test settings when OK is clicked** checkbox is enabled, MAA checks the accessibility of specified target path and verifies if the file can be created.

When these are provided, press **OK** to apply changes or **Cancel** to close the dialog box without any updates.

New Protocol Wizard

How to: Launch New Protocol Wizard

1. Launch the MADE and do one of the following:

2. Select the **New Protocol...** option on MADE Welcome Screen.

3. Select the **Protocols** tab and click on the **New** button of **File** group.

The New Protocol Wizard provides a way to easily create a new assay protocol by entering only the minimum and most important information.

It guides you through the steps you can move between by clicking **Next** and **Back** buttons to configure the following assay parameters:

- Measurement data specifics

- Layout and sample types settings

- Administrative properties (meta data)

- A name and (optionally) a description for the protocol

> At any step, you can click on the cross icon at the upper right of the New Protocol Wizard window to discard all changes and exit.

At the end of the wizard, the .assay-protocol file with defined configuration is added to your default **Protocols** folder.

Analysis Launcher

The Analysis Launcher tool simplifies building up the command line arguments to launch a protocol with a specified set of options in MAA. It provides visual controls and validation to predetermine data analysis with no user intervention required. Also, it comes in handy when configuring multiple runs of a certain data analysis (via assay protocol) with various measurements data.

The Launcher includes:

- Controls for browsing and selecting an assay protocol, measurement data, Sample IDs, and protocol tweaks files

- Various user-friendly controls for selection of command line arguments

- A textbox showing the full command line of the current selection

- The **Copy to clipboard** button, which copies a built command line with selected controls to the clipboard

- The **Launch** button, which launches the command line

The built command line is displayed as changes are made. When valid, the command line can be launched directly from **Launcher**.

How to: Open Analysis Launcher

You can open this tool in one of the following ways:

- Press the **Analysis Launcher** button on the **Tools** tab in the **Utilities** group of the MADE Ribbon.

- Enable the Launch Assay Protocol with Options feature and press the **Launch Assay Protocol with Options** button.

- Launch the **MyAssays.Desktop.Analysis.Launcher** application under the **MyAssays/MyAssays Desktop/** installation root folder.

> If you run the application from the installation root, specify the protocol file path to see all applicable controls.

Installed Protocols

4PL (Constrained by B0 and NSB) Quantification

Constrained [Four Parameter Logistic (4PL)](#) quantification of sample concentrations for enzyme immunoassay (EIA).

With this analysis, the top and bottom asymptotes of the curve are constrained to non-specific and maximal binding as determined by the measurements of **B0** and **NSB** samples.

This protocol is suitable for enzyme immunosorbent assays measured on 96-well (12x8) microplates.

This analysis includes a background correction step. If a blank group is included on the selected layout, the mean of the blank replicates is first subtracted from the raw data measurements (the corrected values are then used in the fit).

B/B0% is calculated for each sample using the measured **B0** and **NSB** samples.

The standard data points (concentration vs. B/B0%) are plotted with a 4PL curve fitted through the standard data points. The curve is constrained so that the upper and lower asymptotes equal **B0** and **NSB** (0 and 100%).

The concentrations of the samples are determined from the fit with any specified dilution factors applied.

4PL Quantification

[Four Parameter Logistic (4PL)](#) quantification of sample concentrations for enzyme immunoassay (EIA).

This protocol is suitable for enzyme immunosorbent assays measured on 96-well (12x8) microplates.

This analysis includes a background correction step. If a blank group is included on the selected layout, the mean of the blank replicates is first subtracted from the raw data measurements (the corrected values are then used in the fit).

B/B0% is calculated for each sample using the measured **B0** and **NSB** samples.

The standard data points (concentration vs. B/B0%) are plotted and the selected curve is fitted through the standard data points. The concentrations of the samples are determined from the fit with any specified dilution factors applied.

ATP Lite Protocol

Quantitative analysis of samples using a standard curve fit to calculate ATP concentrations. This protocol is suitable for ATP Lite 1-step luminescence assay system measured on 96-well (12x8) and 384-well (24x16) microplates.

The standard data points (concentration vs. measurement) are plotted and the selected curve is fitted through the standard data points. The concentrations of the samples are determined from the fit with any specified dilution factors applied.

Samples outside the range of the standards are highlighted in yellow.

Best-Fit Quantification

Best-Fit quantification of sample concentrations for enzyme immunoassay (EIA).

This protocol performs multiple curve-fits with different weighting models and automatically picks the best.

This protocol is suitable for enzyme immunosorbent assays measured on 96-well (12x8) microplates.

This analysis includes a background correction step. If a blank group is included on the selected layout, the mean of the blank replicates is first subtracted from the raw data measurements (the corrected values are then used in the fit).

B/B0% is calculated for each sample using the measured **B0** and **NSB** samples.

The standard data points (concentration vs. B/B0%) are plotted and the determined best-fit curve is fitted through the standard data points. The concentrations of the samples are determined from the fit with any specified dilution factors applied.

DNA 260nm

Quantitative analysis of samples using a standard curve fit to calculate DNA concentrations. This protocol is suitable for assays measured on 96-well (12x8) and 384-well (24x16) microplates.

This analysis optionally includes a background correction step. If a blank group is included on the selected layout, the mean of the blank replicates is first subtracted from the raw data measurements (the corrected values are then used in the fit).

The standard data points (concentration vs. measurement) are plotted and the selected curve is fitted through the standard data points. The concentrations of the samples are determined from the fit with any specified dilution factors applied.

Samples outside the range of the standards are highlighted in yellow.

The %CV of the final concentration replicate values are computed.

DNA Quantification

Quantification of DNA in samples using [linear regression](#).

The standard data points (concentration vs. fluorescence) are plotted and linear regression is used to determine the linear relationship. The DNA concentrations of the samples are determined from the linear model.

The %CV of the final concentration replicate values are computed.

Dose–Response Curves for EC50

Play Video

This assay protocol computes dose–response curves for each sample group using [Four Parameter Logistics (4PL)](#) regression. The fitted curves are plotted and overlaid on a single chart for visual comparison. Also, the fit results values are reported.

This protocol is provided for 96-well (12x8) microplate layout.

For each sample group, the measurement values are plotted against specified dose values. (This assumes that each sample group is dosed in the same way.)

Doses can be specified as:

- A start dose and a common dilution/concentration series
- Unique values for each dose

Assay Protocol

To perform data analysis on this assay:

1. [Launch Dose–Response Curves for EC50 assay protocol](#) or [results file](#).
2. Provide your required measurement data for the assay.
3. [Select or create a layout](#) that matches you assay design.

> All sample groups are assumed to have the same number of positions defined on the layout (e.g., if **Unknown1** has 10 replicates, then all **Unknowns** on the layout must also have 10 replicates).

4. In the **Doses** tab, specify dose values (as described below).
5. Press the **Calculate** button to perform calculations and see the results.

Doses Configuration

The **Doses** editor specifies the doses (*x* values) to plot against the specified responses (measurements). The editor is displayed by clicking on the **Doses** tab. Its display and behaviour depends on the layout design and the mode in which the data is to be specified.

Dose Replicates

Select the number of adjacent positions each dose value applies to. **Dose Replicates** are determined automatically and include all proper divisors (i.e., all divisors excluding itself) of the number of positions in each sample.

For example, for a 12x8 layout with 8 **Unknown** sample groups, each group contains 12 positions. Thus, **Dose Replicates** options include 1, 2, 3, 4, 6.

Dose Concentrations

The left-hand **Doses** grid is used to select and indicate the dose value to edit. Select the dose to edit in a two-column grid.

The **Dose Group** column is numbered from 1 upwards, prefixed with Dose. The number of rows is updated according to the selected **Dose Replicates** option. This is the number of replicates in a sample divided by the selected **Dose Replicates** option.

In the **Dose** column, left-click on a dose to edit its value. There are two modes for specifying dose values:

- **Series** – Provide a **Dose** value for the first dose group and common series to apply to all groups. Use this if all of the doses share a common concentration/dilution series.

- **Non-Serial** – Provide a specific dose value for each dose group. Use this if your doses do not share a common concentration/dilution series or are not in a simple mathematical series.

Use the **Series** button to toggle the concentration mode state.

Paste Option

You can copy and paste the clipboard contents to the **Dose** column. The content is pasted starting from the first row on a line by line basis. This is useful if you have your dose values in another application (such as Excel or notepad).

> ℹ️ Upon pressing the **Paste** button, the mode is automatically switched to **Non-Serial**.

Flashing Replicates

When a row is selected, the positions associated with the selection flash on the Layout Preview pane. With this, you can review to which replicates in each group a provided Dose value is applied. For example, with layout:

	1	2	3	4	5	6	7	8	9	10	11	12
A	1	2	3	4	5	6	7	8	9	10	11	12
B	1	2	3	4	5	6	7	8	9	10	11	12
C	1	2	3	4	5	6	7	8	9	10	11	12
D	1	2	3	4	5	6	7	8	9	10	11	12
E	1	2	3	4	5	6	7	8	9	10	11	12
F	1	2	3	4	5	6	7	8	9	10	11	12
G	1	2	3	4	5	6	7	8	9	10	11	12
H	1	2	3	4	5	6	7	8	9	10	11	12

🟢 Unknown

If **Dose Group** 1 is selected and **Dose Replicates** is 1, then all positions on row A flash. If **Dose Group** 1 is selected and **Dose Replicates** is 2, then all samples on row A and row B flash.

Results

This assay protocol fits a dose–response curve to each sample group of the **Unknown** type. It reports an overlaid chart and a table with fit results values for each sample group.

Dose–Response Curve Chart

The chart contains a curve fit plotted with 4PL for each sample group. It allows you to inspect the fitted curves. For example, here is the chart that shows results for some exemplary data for a layout above:
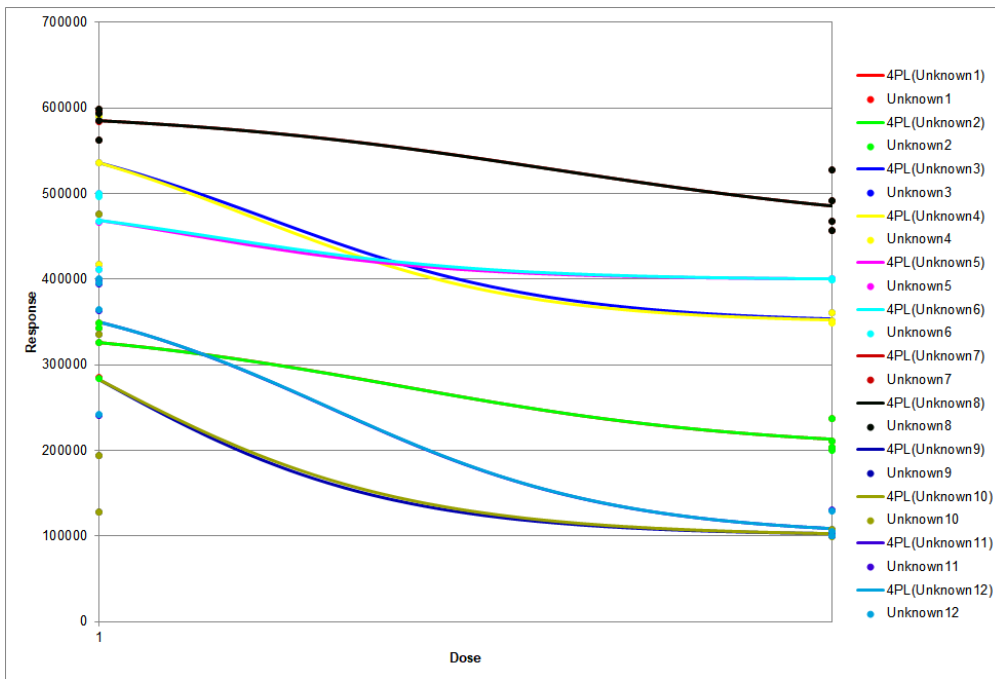
Dose–Response Fit Table

Here is a snippet that shows the Dose–Response Fit Table results for **Unknown1** (with the configuration for **Unknown1** data from the chart above):

Sample	Bottom	Top	Span	Hill Slope	EC50	Log(EC50)	R^2
🟢Unknown1	348207	200753	147454	5.92807	1.33144	0.124322	0.884226

ELISA (96 well) - Single Page Report

Quantitative analysis of samples using a standard curve fit to calculate concentrations. This protocol is suitable for enzyme-linked immunosorbent assays measured on 96-well (12x8) microplates where samples are measured in replicate groups.

The page-based report is designed for printing on a single page.

This analysis optionally includes a background correction step. If a blank group is included on the selected layout, the mean of the blank replicates is first subtracted from the raw data measurements (the corrected values are then used in the fit).

The standard data points (concentration vs. measurement) are plotted and the selected curve is fitted through the standard data points. The concentrations of the samples are determined from the fit with any specified dilution factors applied.

The %CV of the final concentration replicate values are computed.

The report lists the computed concentrations and %CV values. Calculated data points are plotted on the curve. Validation tests check for a good R^2 value (greater than 0.9) and for all samples to be within an acceptable precision threshold (%CV is less than 15).

ELISA

Quantitative analysis of samples using a standard curve fit to calculate concentrations. This protocol is suitable for enzyme-linked immunosorbent assays measured on 96-well (12x8) and 384-well (24x16) microplates.

This analysis optionally includes a background correction step. If a blank group is included on the selected layout, the mean of the blank replicates is first subtracted from the raw data measurements (the corrected values are then used in the fit).

The standard data points (concentration vs. measurement) are plotted and the selected curve is fitted through the standard data points. The concentrations of the samples are determined from the fit with any specified dilution factors applied.

Samples outside the range of the standards are highlighted in yellow.

The %CV of the final concentration replicate values are computed.

LANCE cAMP

Quantitative analysis of samples using a standard curve fit to calculate concentrations for a LANCE assay measured on 96-well (12x8) and 384-well (24x16) microplates.

The ratios of the 615nm and 66nm channels are first calculated for each sample.

The standard data points (concentration vs. ratio) are plotted and the selected curve is fitted through the standard data points. The concentrations of the samples are determined from the fit with any specified dilution factors applied.

Samples outside the range of the standards are highlighted in yellow.

The %CV of the final concentration replicate values are computed.

Linearized (Logit-Log) Quantification

Linearized (Logit-Log) quantification of sample concentrations for enzyme immunoassay (EIA). This uses the linearized method which plots logit B/B0 versus log concentration using a linear fit.

> This protocol is provided to support legacy requirements and methods. In general, the 4PL or 5PL methods are better suited for dose-response curves. The main weakness of Logit(B/B0) is that the accuracy reduces towards the upper and lower limits.

This analysis includes a background correction step. If a blank group is included on the selected layout, the mean of the blank replicates is first subtracted from the raw data measurements (the corrected values are then used in the fit).

B/B0 is calculated for each sample using the measured **B0** and **NSB** samples.

The standard data points (concentration vs. Logit B/B0 with log transform) are plotted and the selected curve is fitted through the standard data points. The concentrations of the samples are determined from the fit with any specified dilution factors applied.

Liquid Handling QC

This report shows the quality control analysis of measurements from a 96-well microplate. The results include:

- The mean and %CV of all measurements on the plate
- The mean and %CV of all measurements by-row and by-column
- The % difference between each position's measurement and the plate mean

Qualitative analysis (reporting pass/fail) using specified limits:

- The %CV of the plate, each row and each column is compared against the %CV limit
- The % difference of each position's measurements and the plate mean is compared against the specified ±% limit

OQ Absorbance

Calculates the accuracy and precision of an absorbance test.

Standards Curve with Titres and Parallelism Plots (Dual)

This protocol:

1. Calculates difference between measurement and reference data.

2. Fits a 4PL standards curve (specified concentrations vs. corrected measurements).

3. Calculates concentrations of all samples (from the standards curve).

4. Calculates titre values for each sample (calculated concentration x specified dilution).

5. Plots overlaid 4PL curves of each sample group for parallel line analysis (specified dilution vs. measurement).

(Sample groups include: Standards, Controls and each Unknown group.)

Z-Factor

Calculates Z-Factor in accordance with *Zhang JH, Chung TD, Oldenburg KR, A Simple Statistical Parameter for Use in Evaluation and Validation of High Throughput Screening Assays. J Biomol Screen. 1999;4(2):67-73.*

Common Tasks

Data Storage

Storing Data

The default path for saving your data files lies in your **Documents** folder, under the MyAssays subfolder. You can open it in MyAssays Desktop Explorer, Windows Explorer, or any other content-managing application.

You may want to change the way your data files are stored. For example, you may prefer your data to be stored in a network location or in a shared folder that is shared with other users of the system.

How to: Set an Assay Data Folder Root

In the MADE Ribbon, select **Backstage File Screen| Data**. Press the **Select Folder Root** button and choose the preferable folder.

> Uninstalling MyAssays Desktop will not remove your data files.

Backing Up Data

Periodical backing up of your assets is beneficial so you will always have a copy of all important data, and is a part of responsible PC administration. To facilitate your backup process, all data files are stored under a single parent data folder that should be backed up.

To determine which folder to backup, launch MyAssays Desktop Explorer and select **File | Data** to obtain the current parent directory. All relevant data is stored in the MyAssays subdirectory of this folder.

Visualizing Data

Heatmap Plate View

The **Heatmap View** displays a plate or rack layout colored in accordance with the meas-urement data numerical values. For example, data with smaller numerical values is depicted in dark green and data with larger numerical values is shown in red.

How to: Access the Heatmap View

1. In MAA, select the **Measurements** or **Microplate** (**Layout (Racks)** for rack assays) tab.

2. Check the **Heatmap View** option in the lower left corner of the **Layout Preview** pane.

> ℹ️ Heatmap view is relevant for endpoint data only.

Structure

Heatmap View comprises:

- Plate preview with the color of each position based on its numerical value converted to a color within a given scale

> 📝 Data for **Unused** positions is not displayed; the wells appear white and empty.

- A heatmap scale (located under plate preview) with clickable minimum and maximum controls
- A scale function menu that determines the heatmap scale type (linear or Log10)
- The auto scale option
- 2D/3D view buttons

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.3640	0.3230	2.1050	2.1240	1.2130	1.2260	1.1590	1.1900	0.1100	0.1160	1.4630	1.4810
B	0.4550	0.4410	1.8690	1.8880	1.2030	1.2180	1.1800	1.2060	0.1320	0.1300	0.7860	0.7680
C	0.5870	0.5750	0.4740	0.5140	1.2060	1.2390	1.2030	1.2100	0.1130	0.1160	0.6130	0.6260
D	0.8060	0.7740	0.3640	0.3780	1.1670	1.1870	1.2110	1.2410	0.1010	0.1060	0.4890	0.4900
E	1.1050	1.0650	0.2790	0.3670	1.2150	1.2250	1.2330	1.2620	0.1110	0.1140	1.5790	1.5490
F	1.4250	1.4450	0.3780	0.4110	1.1960	1.2060	1.2260	1.1990	0.1090	0.1080	0.7920	0.7750
G	1.7910	1.8030	0.5600	0.5970	1.2050	1.2280	1.3080	1.3060	0.1260	0.1270	0.6520	0.6590
H	0.2530	0.2700	0.4630	0.4700	1.2140	1.2610	0.1150	0.1160	0.1150	0.1150	0.4900	0.4770

Heatmap View Settings

Minimum and Maximum Controls

These settings allow you to set the color scheme range displayed on the heatmap plate pre-view. Each position on the layout is colored according to its numerical value and the color scale (using the color range min/max values). Values outside the range are rounded to the nearest min or max color.

How to: Change the Minimum Value

1. Hover and click on the minimum value on the left side of the color bar to edit the heatmap minimum value.

2. Type in a minimum value for the heatmap (e.g., 0.05).

3. Click on the check icon to save your changes or on the cross icon to cancel them.

> If you type in a minimum value that is greater than the maximum value, an error message will be displayed.

How to: Change the Maximum Value

1. Hover and click on the maximum value (right side of the color bar) to edit the max value.

2. Type in a maximum value for the heatmap (e.g., 2.00).

3. Click on the check icon to save your changes or on the cross icon to cancel them.

> For multiple plate multiplex assays, consider all used positions before you type in min and max values for the heatmap.

Scale Function

With the Heatmap View, you can use one of the following scale settings for heatmap generation:

- **Log 10 scale** – Minimum and maximum control values change based on a logarithmic function

- **Linear scale** – Minimum and maximum control values change with a constant speed

How to: Switch Between the Log10 and the Linear Scales

1. With the Heatmap View turned on, click on the **Scale function** button (from the righthand side of the heatmap maximum control) to display the scale settings.

2. Select the scale you want the Heatmap View to display with:

- **Log10 scale** – Click on the **Log10 scale button** (right one). The heatmap is refreshed to display a Log10 color scale.

- **Linear scale** – Click on the **Linear scale button** (left one). The heatmap is refreshed to display a linear color scale.

Auto Scale

The **Auto Scale** setting allows you to automatically scale the plate preview, if the minimum and maximum values are changed for a set of measurement data. For example, if you type a custom maximum value in the heatmap maximum control and then change the measurement data, the Heatmap View will be updated with the original scale.

If you select the Auto Scale and there is a change in the measurement data, the Heatmap View is updated using the new scale. In this case, if the edited measurement data has a new min/max, then the heatmap scale is updated accordingly.

How to: Access the Auto Scale

1. With the Heatmap View turned on, click on the **Auto Scale** button (on the right side of the heatmap maximum control).

2. Click on the **Auto Scale** button.

> You can toggle the Auto Scale setting on and off by clicking on the **Auto Scale** button.

2D/3D View

Similar to the Layout View, you can view the heatmap in a 2D or a 3D format.

How to: View the Heatmap in a 3D Format

1. In MAA, select the **Measurements** or **Microplate** tab.

2. Check the **Heatmap View** option in the lower left corner of right pane.

3. Click the **3D** button.

> You can toggle between the 2D and 3D view using the corresponding buttons.

3D View

The 3D view projects single endpoint measurement data layout in three dimensions for a convenient visual comparison. The 3D view can use Layout or Heatmap colors.

How to: View Endpoint Measurement Data in 3D

1. Launch an assay protocol or open a results file.

2. Select the **Measurements**, **Microplate** (**Layout (Racks)** for rack assays), or **Sample IDs** tab.

3. For assays that contain multiple matrices and/or containers, select the matrix you want to view from the drop-down list.

4. Click the **3D** button in the lower left corner of the Layout Preview pane.

> ⓘ Toggle between 2D and 3D view using the corresponding buttons.

The 3D view is based on the default 2D view comprising of rows and columns corresponding to the x-axis and y-axis of the 3D model; the z-axis represents the measurement at each position.

All other actions applicable to 2D layout are also applicable to the 3D view.

Change 3D View

To manipulate the 3D view:

- Rotate: Hold the left mouse button down and drag in the required direction.
- Zoom In/Out: Use the mouse scroll wheel (or hold down left and right mouse button and move up and down)
- Reset: Press the **r** key

Move

You can move the layout model by pressing the middle mouse button and moving the mouse.

Enable/Disable 3D View

The 3D view is available for layouts up to 384 wells/samples. This setting can be modified under the **3D View** subsection of **View** tab in the MAA's File backstage area.

Standards Chart

For assays with endpoint measurement data, the Standards Chart allows you to inspect your sample data alongside the fitted curve. Also, you can flag standard points directly from the chart; this is useful for excluding outliers from the curve fit.

> The Standard Chart is included in the report as one of Standard Curve Fit transform's report elements.
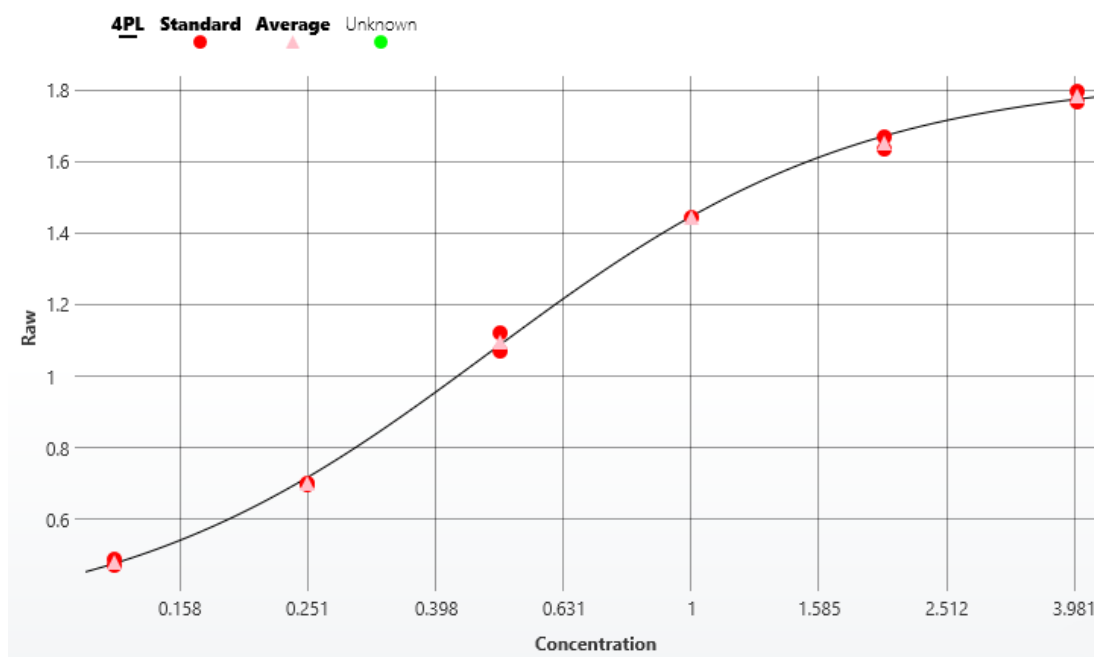
Displaying Samples

Click on the sample name legend above the chart to show/hide the sample groups. For example, here the **Unknown** legend has been clicked to show where the **Unknowns** fall on the curve.

> The **Points on Curve** setting can be used to plot non-calibrator sample types on the fit line.
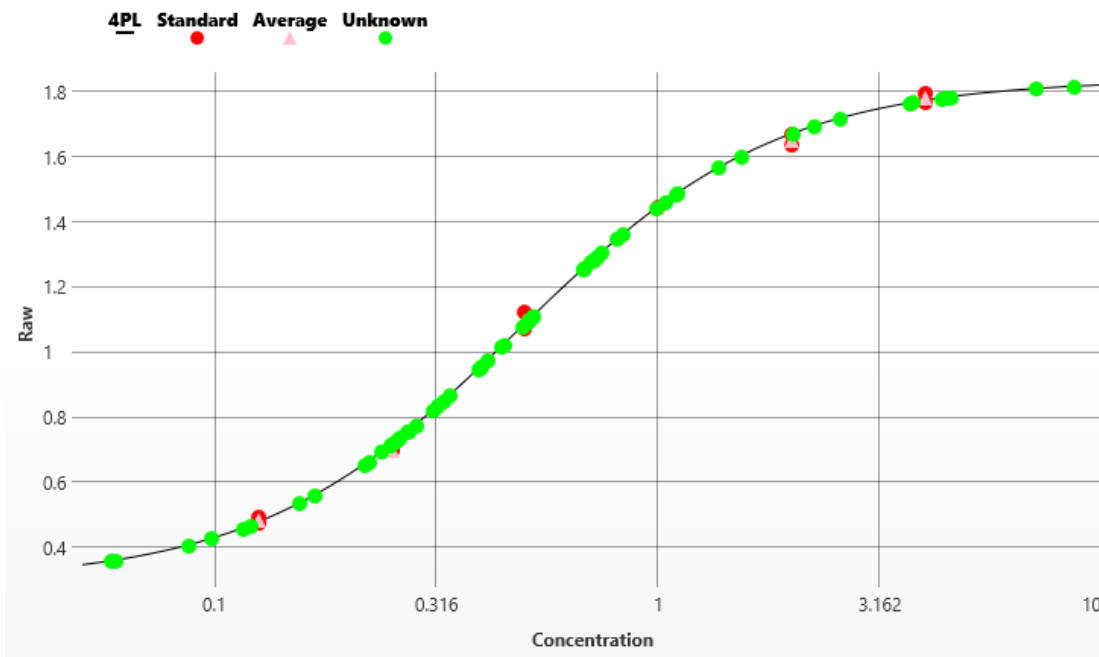
Flag Outliers

Outlier standard points can be removed by simply clicking on the point. After marking the point or points to remove, press the **Calculate** button to generate a new report with the new data set. The flagged points are excluded from the calculations (i.e., not used in the curve fit).
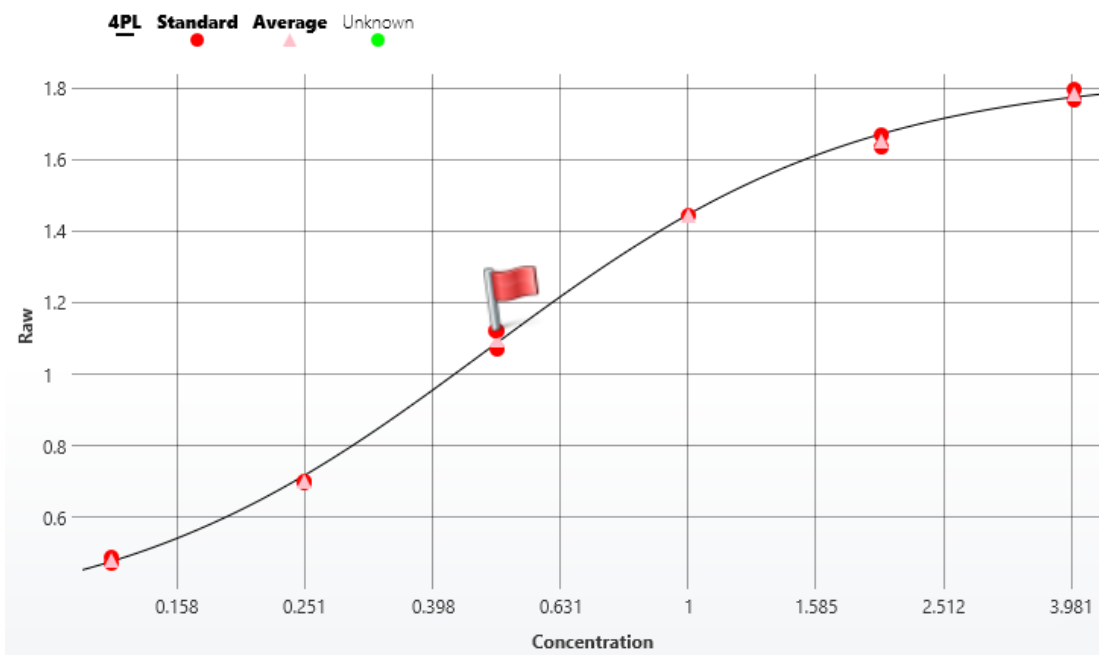
When viewing a Standards Chart for Standard Curve Fit (From Library) transform, flagging **Library Standards** and **Library Average** points in not available.

Chart Zoom

For a closer look at your data points, you can zoom in on the chart by clicking and holding the mouse whilst dragging an area to zoom. Release the mouse button to zoom the chart area. Press the **Reset Zoom** button or double-click on the chart to return to the full view.

Point Details

Hover the mouse over a point to view further information about that point. In the image below, the mouse has been positioned over the point of the third standard to display the point details.
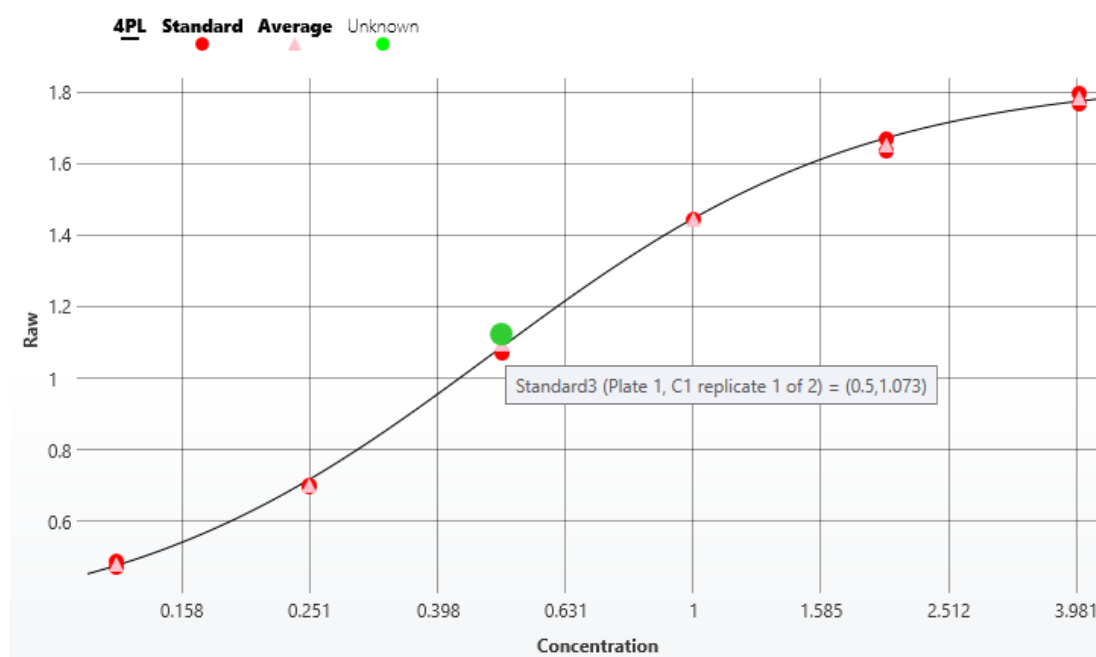
Kinetic/Spectral Overlay

When working with kinetic or spectral data, the Kinetic/Spectral Overlay Chart gives you a clear view of measurements at the position(s) or group(s). The y-axis of the chart corresponds to measurement values; the x-axis represents (depending on the assay) cycle, time, or wavelength values.
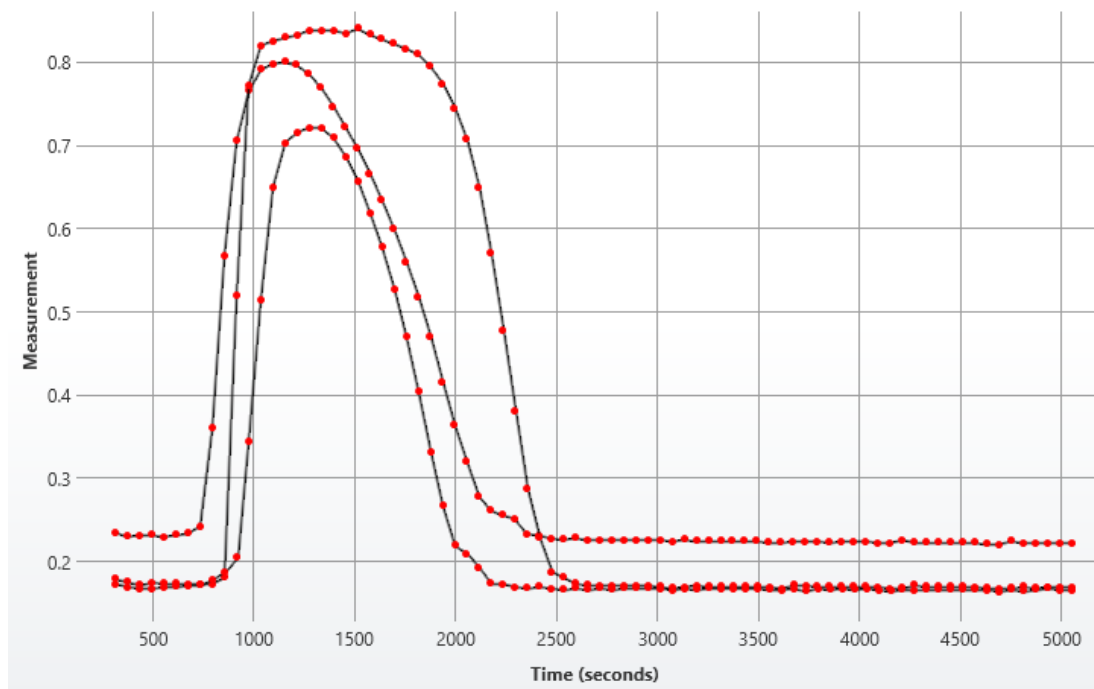
View Selection Modes

Depending on the view selection, you can explore data ranging from a plot that belongs to a single position to the overall plate/analyte values.

How to: View XY Overlay Chart

1. In MAA, select the **Measurements** tab.
2. Select the **Chart** tab.
3. Use the **Select Group**/**Select Position** controls to select the view mode.

There are two modes for Kinetic/Spectral Overlay:

- Position
- Group

For kinetic or spectral data, any specific position can be selected and viewed on a Kinetic/Spectral Overlay chart.
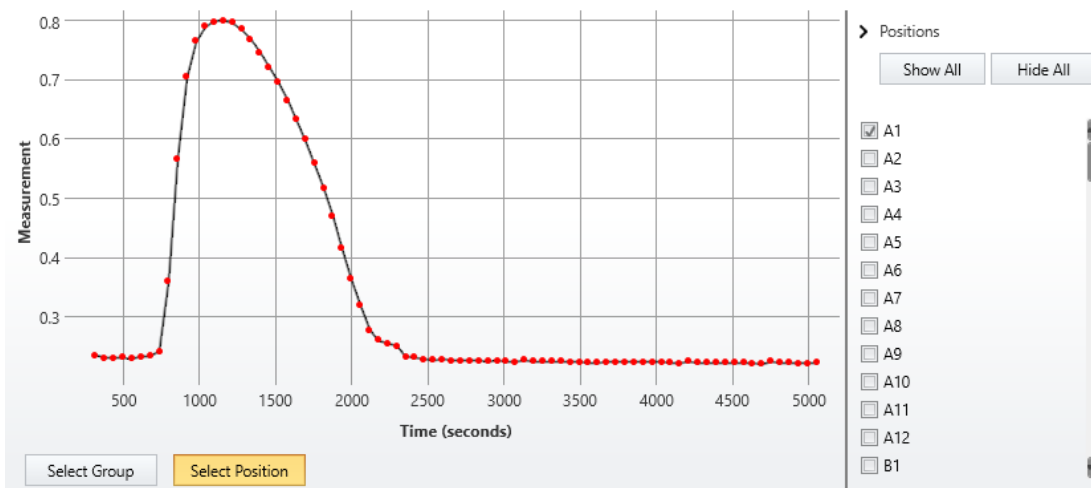
Moreover, any number of positions in any combination can be selected from the right-most View Selection pane. Simply check the checkboxes corresponding to positions you want to view.

Alternatively, any sample group can be also viewed on the Kinetic/Spectral Overlay chart with the same actions as described above.
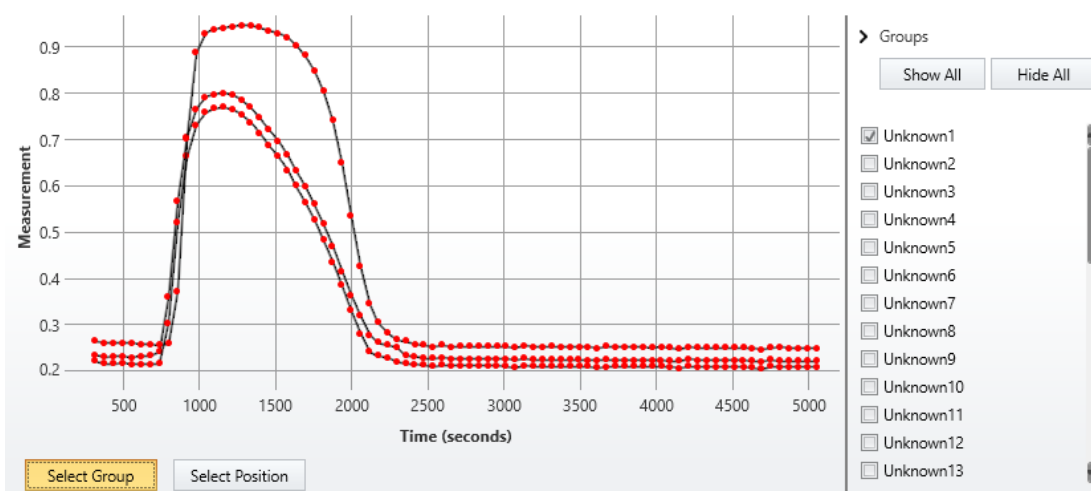
Additionally, you can click on the **Show All** or **Hide All** button to display plots for all of the plate/analyte positions or for none, respectively.

Zoom In/Out

Position the mouse pointer on the area you want to enlarge, press and hold **Ctrl** button, and scroll the middle mouse button up or down to zoom in or out. Alternatively, hold down left mouse button and drag a rectangular area to zoom in.

Double-click the left mouse button or press **Reset zoom** on the upper left of the enlarged area to reset the zoom.

Flagging Outliers

Any data point can be [flagged and unflagged interactively](#) on this view. The chart will update accordingly.
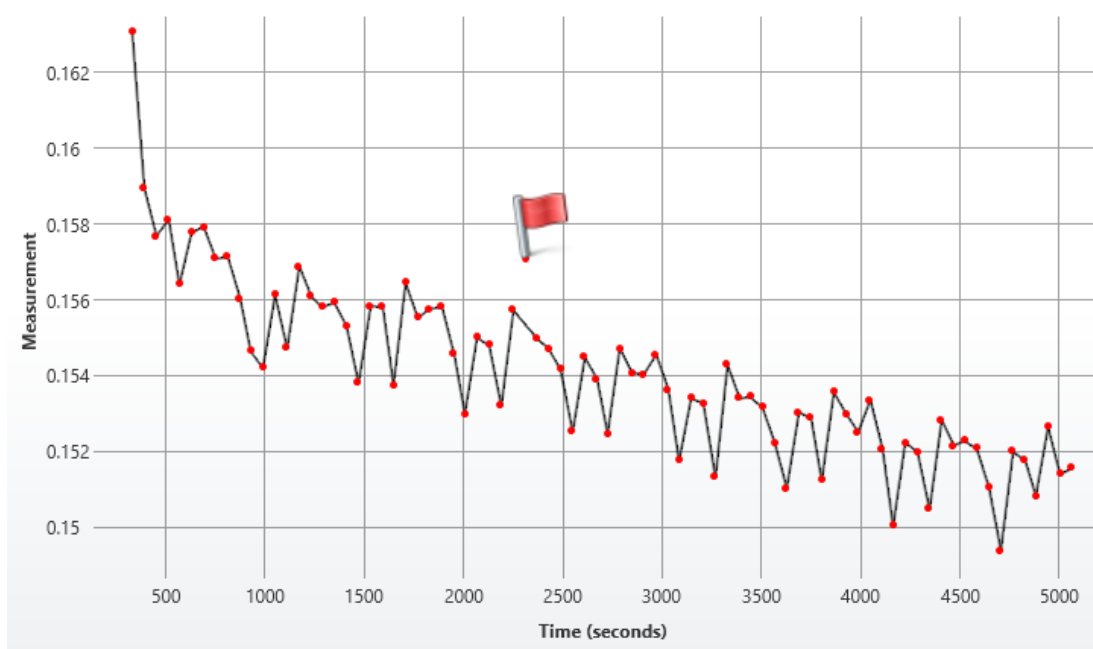
Multiple Plate View

If your measurement data is spread across multiple containers, you can choose which plate/analyte to be displayed. Select the required container from the drop-down list in the left upper corner of the Kinetic/Spectral Overlay Chart.

Kinetic/Spectral Analysis Chart

For kinetic/spectral assays, you can inspect the data analysis results on a Kinetic/Spectral Analysis Chart with respect to the configured XY data settings. Moreover, you can flag points and define subset of data points to consider directly on the chart.
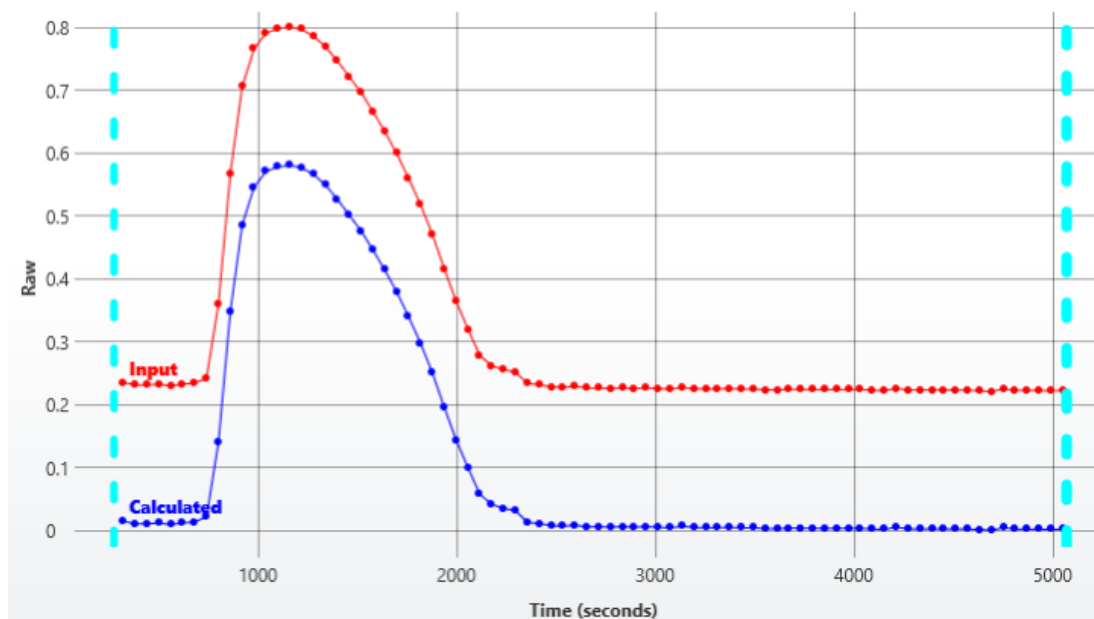
The y-axis of the chart corresponds to the measurement values; the x-axis represents (depending on the assay) cycle, time, or wavelength values.

Displaying Data Plots

In most cases, the results are presented in terms of each position and contain input data plot in red color and calculated result represented in blue.

> For XY Replicates Average and XY Replicates Function, the results are presented for each sample group.

> For XY Blank Correction transform results, the chart is not produced.

Flagging Outliers

To flag an outlier data point, simply click on it. After marking the point or points to remove, press the **Calculate** button to generate a new report with the new data set. The flagged points are excluded from the calculations.
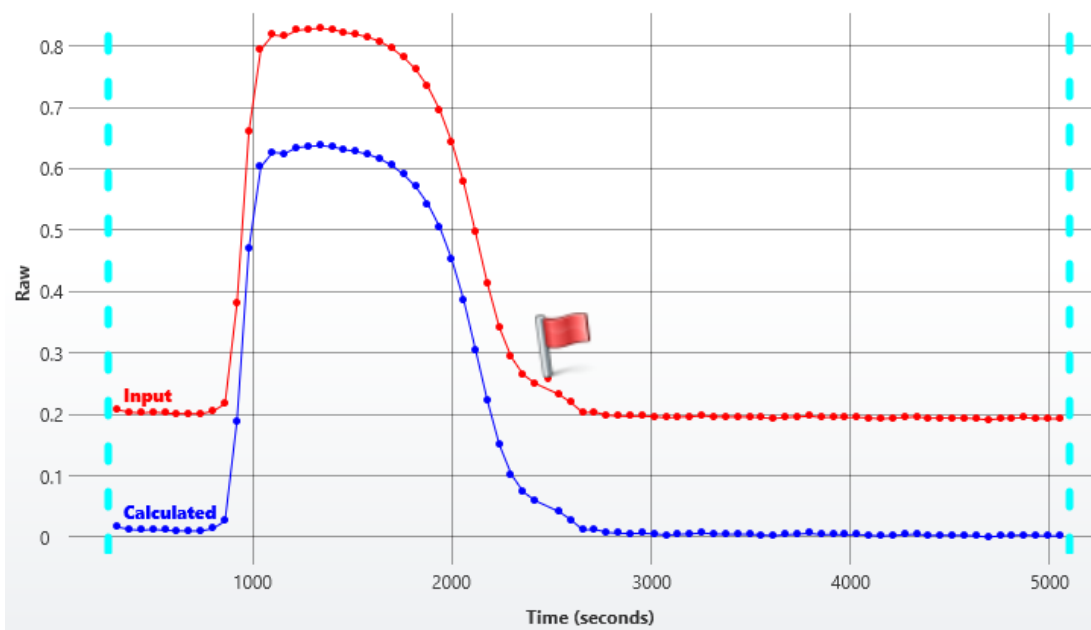
Specifying XY Data Subset

For any position, you can define a subset of kinetic/spectral data points to include in calculations via the positioning of vertical dashed range selectors. The points outside the selected range are displayed as greyed out with no lines between them and are exlcluded from further calculations.

Recalculate to generate a new report with the new data set.
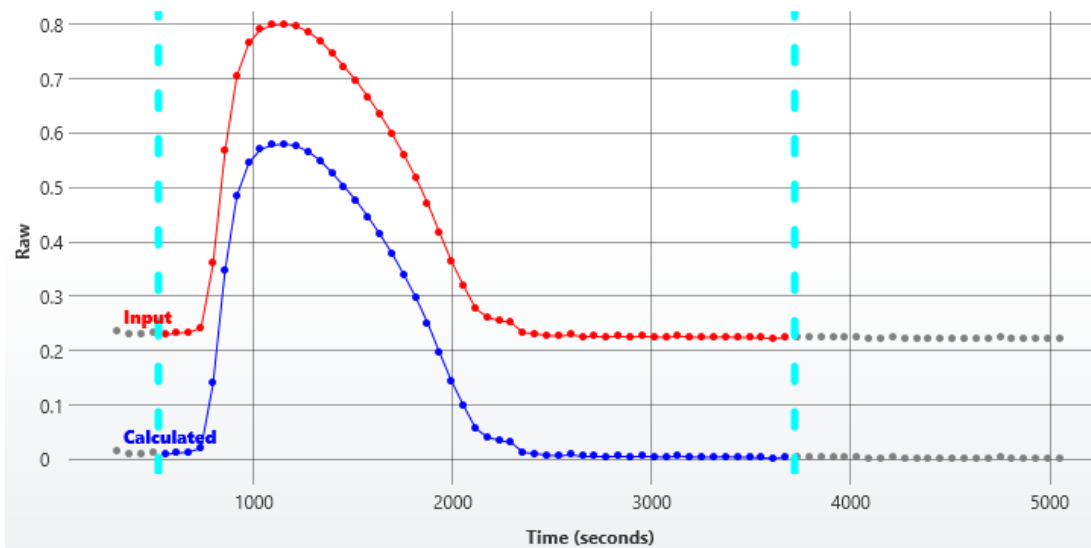
Analysis Description

Also, for all XY Reduction transforms, XY Baseline Correction, XY Fit, and XY Outlier Removal, the description of analysis performed and any non-matrix data for each plot is displayed immediately below the chart.

Fit Parameter

$y = 5.36915x + 0.440824$

Coefficient	Value	+/- Error
m	5.36915	0.221636
c	0.440824	0.118321

Goodness Measure	Value
R²	0.986551
aR²	0.98487
P	0.00000000899332
SE	0.201311
F	586.856
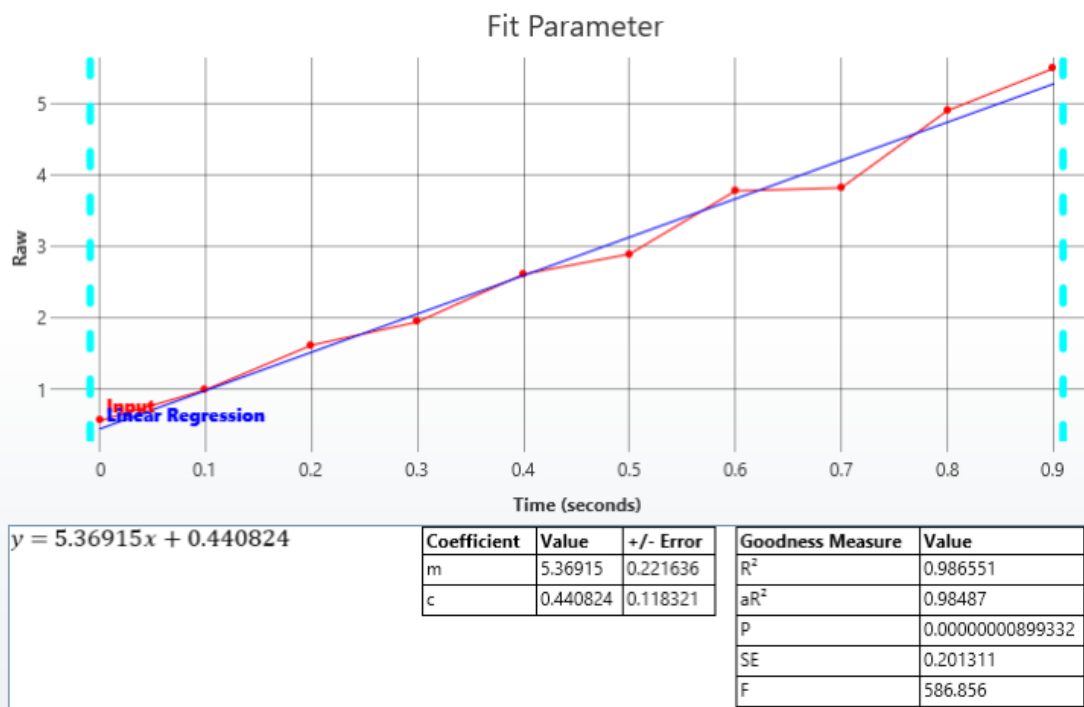
Zoom Options

Position the mouse pointer on the area you want to enlarge, press and hold **Ctrl** button, and scroll the middle mouse button up or down to zoom in or out. Alternatively, hold down left mouse button and drag a rectangular area to zoom in.

Double-click the left mouse button or press **Reset zoom** on the upper left of the enlarged area to reset the zoom.

Multiple Plate View

If your measurement data is spread across multiple containers, each plate/analyte is displayed in a separate Kinetic/Spectral Analysis Chart tab.

Flagging/Outlier Exclusion

Overview

Flagging is used to mark and indicate data to exclude from further analysis. This can be used to identify outliers to ignore issues such as poor measurements or data for incorrectly prepared or contaminated samples.

There are two types of data that can be flagged:

- Position
- Point

For all measurements types, any position can be marked as flagged (e.g., position **A1**). When the data at a position is flagged, any reference to the sample contained at that position will not include the flagged position's data.

For example, if **Standard1** comprises of 3 replicates measured at position **A1**, **A2**, and **A3**, if position **A1** is flagged, **Standard1** would refer to only the non-flagged values (i.e., the average of the data at positions **A2** and **A3**).

For kinetic or spectral data, any specific data point can be marked as flagged too (e.g., the data point for cycle 1 of position **A1**). Any flagged data points are not included in any further analysis. If all points of a position are flagged, then the position is considered to be flagged.

For example, if the kinetic measurement data at position **A1** comprises of 10 cycles, if point 1 is flagged, then a kinetic reduction operation is performed (such as computing the average of kinetic cycles in XY Reduction Average) and the calculations would ignore the data of the flagged point.

Data can be flagged interactively or by calculations. Any flagged data will be indicated in the report and export accordingly. It is also possible to customize the labels for flagged data in the report.

Interactive Flagging

Position or point data can be flagged (and "unflagged") interactively by left-clicking the mouse on the position or data point to flag. This is possible in:

- Measurements View (in the **Measurements** tab)
- Standards Chart (in **Results** tab when working with Standard Curve Fit transform)
- Kinetic/Spectral Measurements Chart (in the **Measurements** tab | **Charts** subtab when working with kinetic/spectral data)

- Kinetic/Spectral Analysis Chart (in the **Results** tab when working with Kinetic/Spectral transforms)

How to: Flag Data Position with Measurements View

1. In MAA, select the **Measurements**, **Microplate**, or **Sample IDs** tab.

2. Hover to the Plate Preview pane on the right of the selected tab.

3. Position the mouse pointer over the point to flag and left-click.

> ⓘ A flagged position appears on the Measurements View with a cross-through it.

How to: Flag Data Position with Standards Chart

1. **Add the Standard Curve Fit Transform:**

   1. Launch an assay protocol or open a results file.

   2. If you cannot see the Assay Properties panel, press the **Properties** button to open it.

   3. Press the **Add** button.

   4. Select the Standard Curve Fit transform.

   5. Press the **Create** button.

   6. Review or edit the transform's input matrix and output matrix name as required.

7. Provide your required settings for the transform (see the **Properties** section above).

8. Press the **Calculate** button.

2. Select the **Standard Curve Fit** tab of the **Results** tab. If your assay comprises multiple plates, select the one that contains the analysis view for your data.

3. Select the sample type on the legend to view how the points are spread on the curve.

4. Position the mouse pointer over the point to flag and left-click.
   1. Press the **Calculate** button to perform the calculations with the new settings and to see the results.

> ℹ Any flagged item can be "unflagged" by left-clicking on the flagged item.
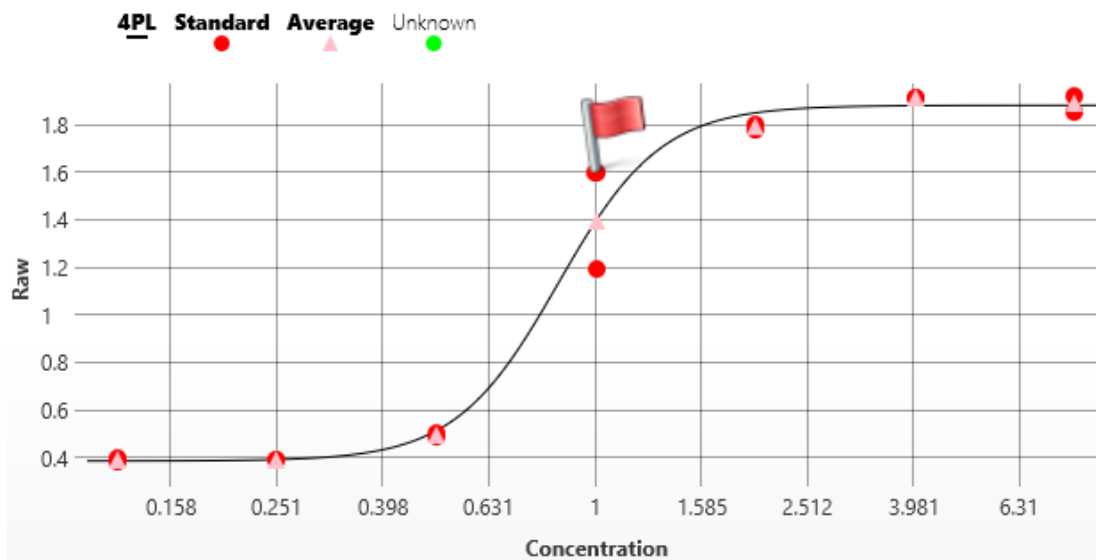
How to: Flag Kinetic/Spectral Data Point with Kinetic/Spectral Measurements Chart

1. In MAA, select the **Measurements** tab.

2. Select the **Chart** tab.

3. Use the **Select Group**/**Select Position** controls to select the required data.

4. Position the mouse pointer over the point to flag and left-click.
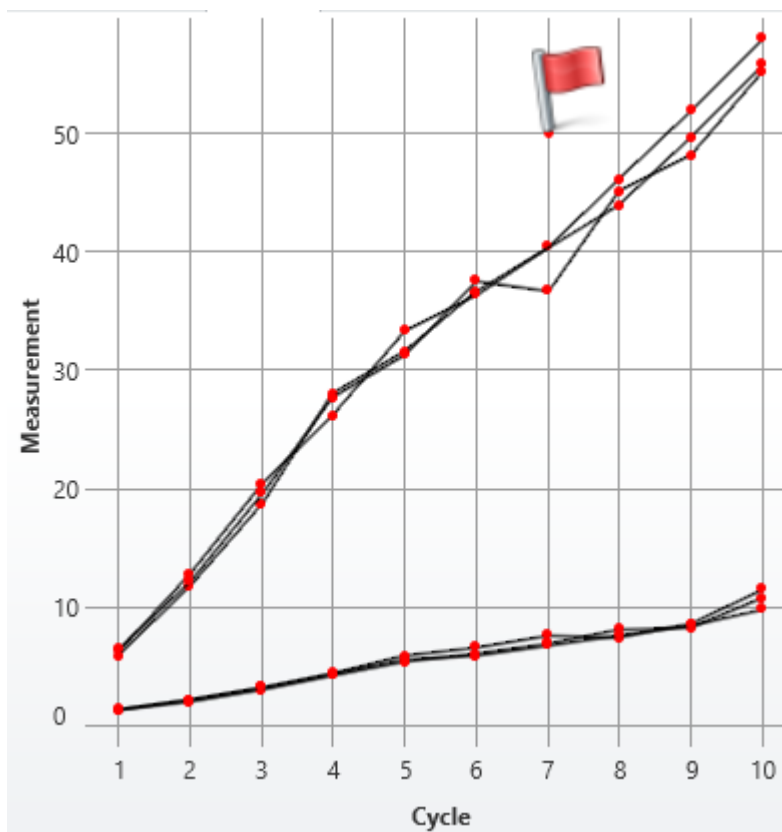
How to: Flag Kinetic/Spectral Data Point with Kinetic/Spectral Analysis Chart

If your data analysis includes an Analysis View (used to display further information about the performed data analysis), then you can flag XY points directly from this view.

1. In MAA, select the **Results** tab.

2. Select the tab that contains the analysis view for your data.

3. Position the mouse pointer over the point to flag and left-click.

> ℹ️ Any flagged item can be "unflagged" by left-clicking on the flagged item.

XY Maximum Slope

Maximum slope is 9.279
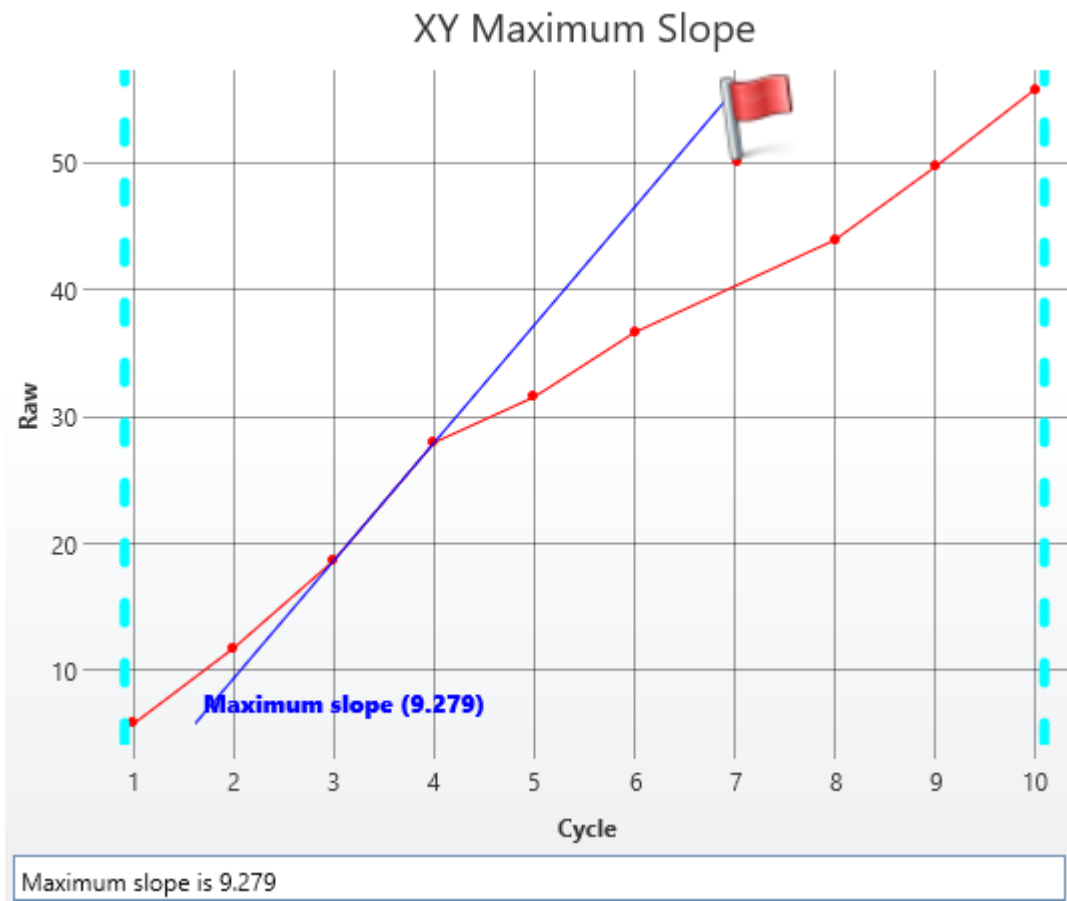
Flags View

If there is any flagged data, then the **Flags** option will appear in the **View** group of the **Home** Ribbon tab. The Flags view control will display a list of the currently flagged items (including both positions and point data if relevant).

Status	Type	Position	Point	Plate	Matrix
☑	Point	A5	7	Plate 1	Raw
☑	Point	A5	6	Plate 1	Raw
☑	Point	A5	5	Plate 1	Raw
☑	Position	A1	-	Plate 1	Raw
☑	Position	A2	-	Plate 1	Raw
☑	Position	A3	-	Plate 1	Raw

Toggle the status of an entry to change its flagged status.

Calculated Flagging

In addition to manually selecting which data to flag, calculations may result in flagged results.

For example, the [Replicate Outlier Removal](#) transform can be used to automatically flag sample replicates that are outside a definable expected range. For more flexibility, the [Auto Flag](#) transform enables you to define conditions to flag endpoint values.

When working with kinetic or spectral data, the [XY Outlier Removal](#) transform can be used to automatically detect specific data points that deviate from an expected pattern beyond an expected threshold.

Also, for certain calculations there might not be a valid result for the calculation. In those cases the result will be marked as flagged (as no further calculation is possible).

Advanced

All transforms include a setting under the **Advanced** properties group for selecting which sample types the transform should calculate results for. For those samples that are not to be calculated, the results can be set to be flagged as either:

- **Flagged with Value**
- **Flagged no Value**

Flagged Data in the Report

The report will indicate flagged data as strikethrough text (such as **~~1.23~~** or **~~Flagged~~**). Where data has been flagged and there is a value associated with it, the original value appears in strikethrough.

Where there is no value associated, the text Flagged appears (also in strikethrough).

For example,

Sample	Positions	Raw	Concentration
🟢 Unknown1	A3	~~1.599~~	1.53791
	A4	1.665	

Here, position **A3** has been flagged. In this case the calculation of the concentration will use only the value from position **A4**.

Here the **Raw** values in positions **A3** and **A4** have been flagged. This means that there is no result for the **Concentration**.

Sample	Positions	Raw	Concentration
🟢 Unknown1	A3	~~1.599~~	~~Flagged~~
	A4	~~1.665~~	

Flagged Data in Text-Based Exports

For text-based export (such as CSV, TXT) where strikethrough is not available, flagged data is identified by asterisk symbols surrounding each flagged data item, for example:

1.23

Flagged

For example, the following export text shows three data values flagged:

```
Sample,Positions,Raw,Concentration
Unknown1,A3 A4,*1.599* *1.665*,*Flagged*
```

Custom Flag Label

If the default strikethrough text "**Flagged**" is not suitable, you can specify your own text to use instead. For example, here the text **No result** is used instead of Flagged.

Sample	Positions	Raw	Concentration
⬤ Unknown1	A3	~~1.599~~	No result
	A4	~~1.665~~	

This is an advanced setting that can only be configured through the XML editor. For more information please refer to Custom Flag Label configuration.

Custom Flag Label Configuration

The Custom Flag Label feature enables specific labels to be defined and used to indicate flagged results instead of the default strikethrough flagged text (such as ~~1.23~~ or ~~Flagged~~).

The settings are available at different levels of the Matrix-Transform Analysis configuration; this provides a fine degree of control over how flagged results are reported. For example, you may prefer to label flagged raw data differently than flagged concentration results.

Additionally, the Standard Curve Fit transform supports conditional flagging and applying labels. This enables the definition of rules to apply that define how to flag and label results in accordance with calculations from the curve. For example, you may want to flag or use a custom label for samples that fall outside a defined concentration range. This is described further in Standard Curve Fit Custom Flag Labels.

> To label sample results that satisfy specified conditions without flagging them, please refer to the Sample Labels topic.

Custom Flag Label Levels

Flag labels can be set at the following levels:

1. By Transform Calculations
2. By Output Matrix
3. General

If there is a combination of custom flag labels defined, then they are handled in the order given above. Custom labels higher in the list take precedence over those further down the list (e.g., any specified in the transform implementation override any defined at both the output matrix and general level).

XML Configuration

These advanced settings are only available through the XML editor. To access the XML editor, select the **XML** tab in the **Assay Properties** panel.

Custom Flag Label: General

The general custom flag label is the label to use in place of the default ~~Flagged~~ label. Note, when specifying custom flag text, strikethrough is not applied.

You can set the general custom flag label at the **Transforms** root level. All flagged data positions on any matrix (raw or calculated) will then use this setting. Enter the flag label with the following syntax:

<Transforms FlagLabel="No result" >

With this setting, the flagged data will appear as follows:

Sample	Positions	Raw	Concentration
⬤ Unknown1	A3	~~1.599~~	No result
	A4	~~1.665~~	

How to: Set General Custom Flag Label

1. Select the **XML** tab in the **Assay Properties** panel (If the **XML** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Locate the **Transforms** element.

3. Add the **FlagLabel** element to the **Transforms** element.

Custom Flag Label: By Output Matrix

A custom flag label can be set on any output matrix of any transform. This defines a common flag label to use for all data positions flagged by the transform.

Provide the label with the following syntax (editable label highlighted in yellow):

<Matrix Id="Calc1"Name ="Calc1" FlagLabel ="Outlier Removed" />

> 📝 This setting only applies to data points flagged by the transform.

How to: Set Custom Flag Label for an Output Matrix

1. Select the **XML** tab in the **Assay Properties** panel (If the **XML** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Locate the **Transforms** element.

3. Locate **Output** element.

4. Add the **FlagLabel** element to the **Matrix** element.

Custom Flag Label: By Transform Calculations

The Standard Curve Fit transform is the only transform that provides custom flag settings by calculation. For more details, see Standard Curve Fit Custom Flag Labels.

Custom Flag Label Reporting

The specified custom flag labels will be used in the report and export; however, the **Sample Table Replicates** setting of matrices configuration also impacts which labels are displayed. This is explained as follows:

- **List**

  For any flagged position that has a custom flag label associated with it, that custom flag label is displayed.

- **Average**

  In this case the flag label is displayed only if all positions in a sample group are flagged; otherwise, average of non-flagged data points is calculated. For the former, the following scenarios apply:

  1. If all positions have no custom flag label then the default flagging settings are used.

  2. If all positions have the same custom flag label, associated with them, then that custom label is displayed.

  3. If positions of a sample group have different flag labels associated with them, then the list of flag labels (custom and/or default) are displayed in the order of the positions. **Example**: *If there are 3 positions in a group and they are all flagged (the first has no custom label, the second has custom label "Label A," and the third has custom label "Label B"), then "Flagged, Label A, Label B" will be displayed*.

- **First**

  Any custom flag label associated with the first position is displayed.

Matrices

Generally, all data in MyAssays is stored in matrices. Each matrix can contain one or more replicates, and for each matrix certain relevant transforms can be applied. In the **Matrices Tab**, the settings for each of these matrices can be configured.

How to: View Matrices Tab

1. Launch an assay protocol or open a results file.

2. If you cannot see the Assay Properties panel, press the **Properties** button to open it.

3. Select the **Matrices** tab (If the **Matrices** tab is not immediately visible, press the **<< Show All Properties** button to expand).

Matrix Selection Pane

This pane includes available matrices in a tree list. This list contains two basic branches: **Raw** and **Calculated**.

Raw

This is input data that is stored in the **Measurements Tab**. The quantity and type of **Raw** matrices depends on the assay type and configuration.

Calculated

This consists of all new matrices that are created and added to output by transforms.

Matrix Configuration Pane

Each input and each output matrix of a newly added transform is automatically included in both Sample and Matrix Tables. With the following settings, you can configure whether and how they are displayed.

General

Name	A title to be used for a matrix in the headers of the Sample Table and Matrix Table. If it is not specified, then the default title is used.

Numerical Formatting

Precision	The number of digits to be displayed for a numerical value. The interpretation of **Precision** depends on the **Type** setting.
Type	Specifies how a numerical value is formatted with regard to precision and rounding options: • **Significant Figures** – The value is reported to the specified number of significant figures. This accepts **Precision** values in range 1 through 14.

	• **Decimal Places** – The number of digits to display to the right of the decimal point. This accepts **Precision** values in range 0 through 14.

Report Contents

Include in Matrix Tables	Defines whether the associated matrix should be included in the Matrix Tables
Include in Sample Tables	Defines whether associated matrix should be included in the Sample Tables
Matrix Table Ids	Defines in which Matrix Table(s) the associated matrix should be included.
Sample Table Ids	Defines in which Sample Table(s) the associated matrix should be included.
Sample Table Replicates	Determines how replicates of each sample group are included in the Sample table: • **Average** – The average value of all replicates is displayed. This applies to Endpoint data only. • **List** – All replicate values are included in the table. • **First** – Only the first value is displayed. This simplifies the view of transforms that result in the same values for all replicates of each sample group (e.g., Average, XY Replicates Average).

Configuration of Endpoint Matrices

For endpoint data you can additionally configure the following settings to apply on the report.

Sample Labels

If this is specified, the **Condition** is evaluated for each position of the associated matrix and **Label** is displayed in place of the value for the position. For further details on this feature, see Sample Labels topic.

Value Bars

Within this category you can configure whether and how to fill replicate values with color for any **Raw** or **Calculated** endpoint matrix.

The value bars are created using the minimum and maximum replicate values of the matrix. These are treated as 0% and 100%, respectively, and filled in accordingly.

Display Value Bars	Defines whether this feature is used in the [Sample Tables](#).
Fill	Defines how to fill in the value bar. Select one of the following types: • **Solid** – To fill value bars self-colored with **Fill Colour**. • **Gradient** – To fill value bars with the gradation from **Fill Colour** in the left to white on the right.
Fill Colour	Click on the drop-down to open a color picker.
Border	It defines how to fill in the value bar's border. Select one of the following types: • **Solid** – To fill value bars self-colored with **Fill Colour**. • **None** – To not display the **Border**.

Sample Labels

The Sample Labels feature can be used add your own text in place of or alongside measurement or computed numerical values. Also, your data or labels can be highlighted to visually enhance the report output; this is useful to draw the reader's eye to specific areas of interest or to color code your report content.

Samples are reported with label and/or color in accordance with the rules that you specify.

> You can define as many rules as you need. If a data point satisfies multiple conditions, then all corresponding labels are displayed in the report.

The rules for sample labels are applied to the corresponding data included in the report. In the Matrix Table report element, the label or coloring would apply to each position. In the Sample Table report element, this might be each individual replicate value or each sample's average; it depends on how the data is configured in the report output.

For example, here is a Matrix Table output highlighting all sample measurements > 1.5 in yellow.

	1	2	3	4	5	6	7	8	9	10	11	12
A	1.586	1.575	1.590	1.556	1.575	1.517	1.741	1.671	1.668	1.778	1.669	1.718
B	0.179	0.186	0.174	0.176	0.181	0.175	2.153	2.067	2.239	2.084	2.177	2.054
C	2.372	2.394	2.306	2.483	2.447	2.267	1.771	1.691	1.711	1.731	1.735	1.843
D	1.408	1.397	1.440	1.424	1.470	1.411	0.589	0.566	0.560	0.565	0.595	0.587
E	1.019	1.047	1.005	0.974	1.014	0.997	2.347	2.336	2.423	2.344	2.315	2.426
F	2.115	2.163	2.069	2.015	2.010	2.017	0.522	0.537	0.528	0.513	0.536	0.505
G	1.377	1.342	1.330	1.360	1.439	1.379	0.082	0.085	0.086	0.084	0.083	0.079
H	1.734	1.760	1.753	1.800	1.705	1.793	0.209	0.201	0.200	0.218	0.215	0.204

With the Sample Table report element, each matrix can be configured to display the average of the sample group or list each replicate. (This can be set with the Sample Table Replicates setting in the Matrix configuration.) In either case the sample label will be applied to the selected data.

For example, the following Sample Table has been configured with the Raw data listing all replicate values. In this case, the label is applied to each replicate that satisfies the rule.

Sample	Positions	Raw
⬤ Unknown1	A1	1.586 > 1.5
	A2	1.575 > 1.5
	A3	1.590 > 1.5
	A4	1.556 > 1.5
	A5	1.575 > 1.5
	A6	1.517 > 1.5

In the following example, the Sample Table has been configured with the Raw data listing the average of the replicates values. In this case, the label is applied to the computed average.

Sample	Positions	Raw
🟢 Unknown1	A1	1.566 > 1.5
	A2	
	A3	
	A4	
	A5	
	A6	

This feature is useful for detecting and reporting samples that are outside expected ranges. For example, you might want to highlight or label samples whose concentration, response, or %CV is outside a specific range.

> ℹ️ Applying label, unlike flagging, does not exclude data from calculations, nor does it change the actual value used in subsequent calculations. It simply replaces their reported values with a specified **Label** and applies any coloring.

How to: Apply a Sample Label

1. Launch an assay protocol or open a results file.

2. If you cannot see the Assay Properties panel, press the **Properties** button to open it.

3. Select the **Matrices** tab (If the **Matrices** tab is not immediately visible, press the **<< Show All Properties** button to expand).

4. Select one of the numbered matrices from under either the **Raw** or **Calculated** branch, which contains the data to label.

5. Click on the drop-down **Sample Labels** property of the Matrix Configuration pane to view **Matrix Sample Label Editor**.

6. Enter the condition (in terms of *x*) along with the label and any highlight or text coloring to apply.

7. Press **Add** to add further rules. Click outside the Editor when you have finished.

8. Press the **Calculate** button to perform calculations with the new settings and to see the results.

Sample Labels Configuration

In the **Matrix Sample Label Editor**, you can add and remove any number of rules and move them up or down with the corresponding arrow buttons.

The rules are specified with the following editable text box properties:

- **Condition** – A Boolean condition to evaluate (typically a <u>conditional expression</u>) in terms of *x*. The *x* value specified in the expression refers to the sample.

- **Label** – The label to be displayed in the <u>Sample Table</u> and <u>Matrix Table</u>, if the **Condition** is satisfied. Use **{x}** to refer to the data value that was used in the condition (i.e., the original data point).

- **Highlight** – This sets the background color for the cell when **Condition** is satisfied. Where there are multiple **Conditions** satisfied, the color of the first satisfied **Condition** is applied.

- **Text** – This sets the text color for the cell when **Condition** is satisfied. Where there are multiple **Conditions** satisfied, the color of the first satisfied **Condition** is applied.

If the **Condition** is satisfied, the replicate will be reported with the corresponding **Label** in place of its value.

Additional Settings

If you click **More>>**, the **Matrix Sample Label Editor** is expanded with two more configuration settings:

- **Sample Types** – The sample types to include when evaluating a **Condition** rule. If this is not specified, then the rule is applied to all sample types.

- **Highlight Row** – This applies a **Highlight** to the whole Sample Table row (and not just the matrix value).

> Where there are multiple Sample Label rules satisfied with **Highlight Row**, the color of the first satisfied rule is applied. This applies to rules satisfied in the associated matrix and rules satisfied in other matrices.

Reporting Data with Labels

The results with labels appear in associated matrix's <u>Sample Table</u> and <u>Matrix Table</u>.

For example,

Sample	Positions	Raw	Concentration
⬤ Unknown1	A3	1.694	>1
	A4	1.766	>1

Here the Condition **x>1** and Label **>1** are specified for **Concentration** matrix. In this case, the calculation of the concentration for positions **A3** and **A4** is reported as **>1**.

Where label has been applied on the data and there is a **{x}** reference associated with the Label, the numeric value appears in the **Label**. For example, with the Condition **x>1** and Label **{x}>1**, the calculation results are reported as:

Sample	Positions	Raw	Concentration
● Unknown1	A3	1.694	2.153>1
	A4	1.766	3.857>1

Sample Labels Examples

Condition	Label Applied If
x<1	The value is greater than 1.
and(x>0, x<1)	The value is in the 0–1 range.
x<Control1	The value is greater than mean of **Control1** sample group.
x>TOV1.d	The value is greater than the d coefficient of the 4PL curve fit (assuming that the 1st transform is a Standard Curve Fit transform using the 4PL curve fit). See Transform Output Variables for more details of how to refer to such results.

Validation

MyAssays Desktop's Validation features can automatically check that measurements and results are within expected ranges. Any configured validation tests are performed as part of each assay run with the results (pass or fail) being clearly presented as part of the output report.

For example, a validation can test that the %CV of the replicates of all sample groups is less than 15.

Validation Definition

A validation is defined using a Boolean validation condition. If the condition evaluates to true, then the validation rule is considered to be satisfied and the result for the validation is **Pass**. If the condition evaluates to false, the validation test result is **Fail**.

The validation condition is specified using an expression along with a matrix reference and type. For example, the validation condition expression:

x < 15

When associated with the %CV matrix and evaluated on all groups will check that the %CV of all sample groups is < 15.

Validation rules are arranged by validations tables. You can add as many validations rules as you need to each validation table. You can create as many validations tables as you need. It is useful to group related validations into validations table accordingly.

> Each validation table is a separate report element so can be configured separately. For example, you might want each validation table to appear on a different sheet in the report.

Working with Validations

The following sections explain how to add, define, and review validation rules. Also, example validation expressions are provided.

How to: Add Validation to an Assay

1. Launch an assay protocol or open a results file.

2. If you cannot see the **Assay Properties** panel, press the **Properties** button to open it.

3. Select the **Validation** tab (If the **Validation** tab is not immediately visible, press the **<< Show All Properties** button to expand.)

4. Press the **Add** button on **Tables** pane.

5. Review and edit the validation's properties as required.

Validation Configuration

The **Validation** tab consists of three panes:

- **Tables** – For managing the validation tables.
- **Conditions** – For managing the validation conditions.
- **Condition Configuration** – For specifying a validation condition.

Tables

The **Tables** pane defines the validation tables that are available for inclusion in the report.

Each validation table is given a unique identifier (e.g., ValidationsTable1).

Use the **Add** and **Remove** buttons to define the number of tables required.

> You may prefer to include all conditions in one single table or arrange conditions between multiple tables.

> The ordering, settings, and inclusion of each Validation Table is controlled via the **Report** contents.

Conditions

The **Conditions** pane defines each validation condition for the selected validations table.

Use the **Add** or **Remove** buttons to add conditions as required. Use the Up and Down buttons to set the order in which the conditions will appear in the table.

For each condition, a description can be specified under the **Description** column. Use this to provide a clear textual description; this will be used in the report to convey what has been validated.

For each condition, select which **Matrix** the condition applies to.

For example, if you plan to validate measurement data (i.e., a Raw matrix), select the Raw matrix to validate. Any of the raw or calculated matrices that contain endpoint data can be validated. Also, it is possible to validate non-matrix data (such as transform output variables).

> If there are no available matrices, then the (NA) label appears. In this case, validation conditions can still be created that do not refer to endpoint matrix data.

> Validation conditions are not applicable to XY data matrices. To validate XY data, use an XY reduction transform and validate the endpoint result.

Conditions Configuration

The **Conditions Configuration** pane is used to specify the selected validation's condition.

The **Expression** setting is used to define the validation expression Boolean condition. This will typically be an expression with a [logical operator](#) and perhaps a [conditional function.](#)

The interpretation of the expression depends on the **Validate** setting:

- **Expression** – The expression is evaluated once (useful for checking specific sample groups).

- **Expression with Every Sample Group (x)** – The expression is evaluated for every sample group on the matrix of every container. (A sample group refers to the average of the replicates that belong to the group that are not flagged).

- **Expression with Every Sample Replicate (x)** – The expression is evaluated for every sample replicate on the matrix of every container.

Expression type expressions are the simplest; they define a single expression that is evaluated once on the specified matrix of each container.

For example, **Standard1 < Standard2** is an example of an **Expression** type expression that validates that the value of Standard1 is less than Standard2 on the specified matrix. (For multiple plate assays, this condition will be evaluated on every plate.)

For **Expression with Every Sample Replicate (x)** or **Expression with Every Sample Group (x)** expressions, the expression will include an x reference, and in these cases the [x variable](#) refers to each position or each group. For example, if you wanted to check that the value at every measured position in a matrix is greater than 0.5, then use the **Expression with Every Sample Replicate (x)** setting with the expression: **x > 0.5**. In this case, every data point (on the selected matrix) is validated.

Similarly, if you wanted to check that the average value of each group in a matrix is less than 3, use the **Expression with Every Sample Group (x)** setting with the expression **x < 3**. In this case, the average of every sample group (on the selected matrix) is validated.

The **Sample Types** setting is used to specify which sample types a **Expression with Every Sample Replicate (x)** or **Expression with Every Sample Group (x)** expression will be evaluated on. By default, these validation expressions are evaluated for all sample types. If you only want to test specific sample types, then use this setting to select which types to validate.

Validating Non-Matrix Data

In addition to validating matrix data, it is also possible to validate other types of numerical data. For example, you may want to verify that the R^2 value of a curve fit is greater than a specific threshold. These types of validations can be defined using the **Expression** type and

referring to the data to validate within the expression. This will typically be using a [Transform Output Variable (TOV)](). The TOVs that are available will depend on your configured transforms.

For example, the expression **TOV1.R2 > 0.9** will check that the R^2 value of the curve fit (as used in the first transform) is greater than 0.9.

Validation Results

Validation conditions are checked when the data analysis is performed (after all transform calculations).

To apply any new or modified settings, press the **Calculate** button and inspect the report to see the results. Each validation test verifies the result in terms of the specified Boolean condition.

> For **Expression with Every Sample Replicate (x)** and **Expression with Every Sample Group (x)** validations, the result is only **Pass** if all evaluations evaluate to true.

The results will appear in the associated [validation table]() in the report and will be marked as **Pass**, **Fail**, or **Inconclusive** accordingly:

Pass

If the validation condition is evaluated as true (the numerical result not equal 0).

Fail

If the validation condition is evaluated as false (the numerical result equals 0). For **Fail**, the whole validation table row is highlighted with yellow background.

Inconclusive

If the validation expression could not be evaluated (e.g., a syntax error in the expression), the result is treated as **Inconclusive** with the error message shown in **Results** column (in red text).

> All validations are evaluated (irrespective of whether they are included in the report). You may [configure how and if the Validation Tables are reported.]() In any case, the results of each validation test are listed in the [Calculation log]().

Examples

Here are examples of validation rules along with descriptions of their behavior. They apply the validation on the selected data matrix.

Type	Condition	Validates
Expression with Every Sample Replicate (x)	**x < 2.5**	All positions (that are not flagged) have a value less than 2.5.
Expression with Every Sample Replicate (x)	**and (x > 0, x < 2.5)**	All positions (that are not flagged) have a value greater than zero and less than 2.5.
Expression with Every Sample Group (x)	**x < 2.5**	The average of the replicates (that are not flagged) of all sample groups is less than 2.5.
Expression with Every Sample Replicate (x)	**and (x > 0, x < 2.5)**	The average of the replicates (that are not flagged) of all sample groups is greater than 0 and less than 2.5.
Expression	**Control1 < 1.5**	The average of Control1 replicates (that are not flagged) is less than 1.5.
Expression	**sd(Control1)<10**	The standard deviation of the Control1 replicates (that are not flagged) is less than 10.
Expression	**pcv(Control1)<15**	The %CV of the Control1 replicates (that are not flagged) is less than 15.
Expression	**median (Control1)>0.75**	The median of the Control1 replicates (that are not flagged) is greater than 0.75.
Expression	**TOV1.R2 > 0.9**	The R^2 value of the curve fit (as used in the first transform) is greater than 0.9. (The R^2 value is referenced using a Transform Output Variable).

Evaluation

MyAssays Desktop's Evaluation feature can be used to calculate and present specific quantities of interest in the report. For example, the limits of detection of an assay could be computed and reported using an evaluation.

Evaluations are expression-based calculations arranged as Evaluations Tables for inclusion in the report. Any value that can be referenced or computed can be included in an Evaluations table.

Unlike the Sample Table or Matrix Table report elements (which display the results of a calculation performed on each sample), an Evaluations Table reports calculations that are not computed for every sample.

> If you want to perform a calculation with a result for every sample, instead of using an evaluation, use a transform with the results for each sample displayed in the Sample Table or Matrix Table.

Evaluation Definition

A single evaluation is defined using an expression and can be given a description to accompany its result.

Evaluations are arranged by Evaluations Tables. You can add as many evaluations as you need to each Evaluations Table. You can create as many tables as you need. It is useful to group related evaluations into Evaluations Tables accordingly.

Working with Evaluations

The following sections explain how to add, define, and review evaluations. Also, example evaluations expressions are provided.

How to: Add an Evaluation to an Assay

1. Launch an assay protocol or open a results file.
2. If you cannot see the **Assay Properties** panel, press the **Properties** button to open it.
3. Select the **Evaluation** tab (If the **Evaluation** tab is not immediately visible, press the **<< Show All Properties** button to expand).
4. Press the **Add** button on **Tables** pane.
5. Review and edit the evaluation's properties as required.

Evaluation Configuration

The **Evaluation** tab consists of three panes:

- **Tables** – For managing the evaluation tables
- **Evaluations** – For managing the evaluations
- **Expression Configuration** – For specifying the evaluation expression

Tables

The **Tables** pane defines the Evaluations Tables that are available for inclusion in the report.

Each table is given a unique identifier (e.g., EvaluationsTable1).

Use the **Add** and **Remove** buttons to define the number of tables required.

> You may prefer to include all evaluations in a single table or arrange evaluations between multiple tables.

> The ordering, settings, and inclusion of each Evaluation Table is controlled via the **Report** contents.

Evaluations

The **Evaluations** pane defines each evaluation for the selected evaluations table.

Use the **Add** or **Remove** buttons to add evaluations as required. Use the **Up** and **Down** buttons to set the order in which the evaluations will appear in the table.

For each evaluation, a description can be specified under the **Description** column. Use this to provide a clear textual description; this will be used in the report to convey what has been evaluated.

For each condition, select which **Matrix** the condition applies to. For example, if you plan to evaluate a quantity using measurement data (i.e., a Raw matrix), select the Raw matrix. Any of the raw or calculated matrices that contain endpoint data can be referenced in an evaluation. Also, it is possible to validate non-matrix data (such as transform output variables).

> If there are no available matrices, then the (NA) label appears. In this case evaluations can still be created that do not refer to endpoint matrix data.

> Evaluations are not applicable to XY data matrices. To perform an evaluation on XY data, first use an XY reduction transform and evaluate the endpoint result.

Expression Configuration

The **Expression Configuration** pane is used to specify the selected evaluation's expression.

The **Expression** setting is used to define the expression to evaluate. This might reference a specific sample group or position on the selected matrix. It might also perform a calculation or function. Refer to the [expression](#) section for a complete description of available expression functionality.

The **Numerical Formatting** options are used to specify how the computed value is presented:

Numerical Formatting

Precision	The number of digits to be displayed for a numerical value. The interpretation of **Precision** depends on the **Type** setting.
Type	Specifies how a numerical value is formatted with regard to precision and rounding options: • **Significant Figures** – The value is reported to the specified number of significant figures. This accepts **Precision** values in range 1 through 14. • **Decimal Places** – The number of digits to display to the right of the decimal point. This accepts **Precision** values in range 0 through 14.

Named Evaluation Variables

When working with evaluations, it can be useful to name the evaluation so that its result can be easily referenced from other calculations. A Named Evaluation Variable (NEV) is simply an evaluation result that has been assigned a name. The NEV can be referenced in:

- Evaluation expressions (that follow its definition)
- [Validation](#) conditions
- [Sample Labels](#) conditions

To define a NEV, set the **Name** attribute on the evaluation configuration.

> This is an advanced setting that can only be configured through the XML editor.

How to: Add a Named Evaluation Variable

This assumes that one or more Evaluations are already configured through Evaluation pane.

1. Select the **XML** tab in the **Assay Properties** panel (If the **XML** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Locate the **EvaluationTables** element and, if there are several **Table** elements configured, locate the one containing required expression.

3. Locate the required **Evaluation** child element and add a **Name** attribute to it.

4. Press **Apply** to save changes.

Referring to Named Evaluation Variables

The syntax for referencing NEVs is as follows:

eval{N}.{Name}

Where:

{N} – 1-based Evaluations Table index

{Name} – The name assigned to the evaluation

When referenced in expression, a syntax is **eval{N}.{Name}**, e.g., **eval1.upper**.

XML Syntax Example

```
<EvaluationTables>

<Table Id="EvaluationsTable1">

    <Evaluation MatrixId="Raw1" Expression="max(tov1.a,tov1.d)" Description="Upper Asymptote" Name="upper" />

    <Evaluation MatrixId="Raw1" Expression="eval1.upper * 2" Description="Upper Asymptote * 2" />

</Table>

</Evaluations>
```

In this example, the first evaluation is named **upper** and referenced in the expression for computing the second evaluation. You can only reference NEVs that originate from evaluations that are defined and evaluated prior to where the reference is being made.

In addition, you can refer to a NEV in other calculations, such as validation and sample label conditions.

> All calculations are computed in the order they are defined in the Report Elements, therefore ensure that Evaluations Tables appear earlier in the sequence of the included Report Elements to where they are referenced. (To change the actual report content ordering use a Report Template.)

For example, to refer to a NEV in the Sample Labels configuration, the Evaluations Table must be above the Sample Table and, if included, Matrix Table in the Report Elements order.

Evaluation Results

Evaluations are computed when the data analysis is performed (after all transform calculations). To apply any new or modified settings, press the **Calculate** button and inspect the report to see the results.

Each evaluated expression results in a single numeric value. If there is an error in evaluating the expression, then the error message is reported (in red) as the result.

> All evaluations are performed (irrespective of whether they are included in the report). You may configure how and if the Evaluation Tables are reported. In any case, the results of each evaluation test are listed in the Calculation log.

Examples

Here are examples of evaluations along with descriptions of their behavior. They apply the evaluation on the selected data matrix.

Evaluates	Computes
Control1	The average of all Control1 replicates (that are not flagged)
Control1/Control2	The ratio of Control1 and Control2
sd(Control1)	The standard deviation of the Control1 replicates (that are not flagged)
pcv(Control1)	The %CV of the Control1 replicates (that are not flagged)
Unknown	The average of all replicates of all Unknown samples (that are not flagged)
median (Unknown)	The median of all Unknown sample measurement (that are not flagged)
TOV1.R2	The R^2 value of the curve fit as used in the first transform (referenced using a Transform Output Variable)

Report Content

Overview

The primary output of the data analysis process is the report. MyAssays Desktop provides comprehensive functionality to configure the report's:

- content

- presentation

- export

This section describes how to configure the content that is included in the report.

> Within the MyAssays Desktop Application, the report can be displayed as **Sheet View** or **Page View**; these settings are available in the **Report** tab. When getting started, we suggest using the Sheet View so that your outputs are not confined by the limitations of a printed page. Use the Page View only if and when the layout of the printed page is important (for page-based reporting, you can define a reporting template to apply to your MyAssays Desktop output).

Report Content Definition

The content available for inclusion in the report depends on the configuration of each of the following settings (these each correspond to a tab in the **Assay Properties** pane):

- **Transforms** – Any configured transforms will result in calculated elements (such as numerical results or charts).

- **Matrices** – The settings of each matrix (raw measurement data and any calculated) can be configured. This includes how numerical data is presented (numerical formatting settings), if it is included, how replicate values are reported, and any optional colored value bars.

- **Evaluation** – Any configured evaluation settings will result in one or more evaluation tables, each listing the results of the computed evaluations.

- **Validation** – Any configured validation settings will result in one or more validation tables, each listing the results (pass or fail) of the computed validations.

Various report elements are available depending on these settings. For example, if a Standard Curve Fit transform is configured, then the calculated concentrations will be available for inclusion in a Sample Table and/or Matrix Table, along with details of the fit (including standards chart and equations), each of which can be further configured.

The configuration and ordering of each report element is defined in the **Report** tab.

How to: View Report Tab

1. Launch an assay protocol or open a results file.

2. If you cannot see the **Assay Properties** panel, press the **Properties** button to open it.

3. Select the **Report** tab (If the **Report** tab is not immediately visible, press the **<< Show All Properties** button to expand).

4. Review and edit the report's elements as required.

Report Element Configuration

The **Report** tab consists of two panes:

- **Report Elements** – This pane manages the report elements and their ordering.
- **Report Configuration** – This displays the available settings for the selected report element.

Report Elements Pane

The **Report Elements** pane lists the elements that will be included in the report. Their ordering reflects the order in which they will be added to the report.

Use the **Add Element** button to add a new report element. A list of available report elements will appear. The selected item will be added to the end of the list.

Use the **Remove** button to remove the selected report element.

To change the ordering of report elements, select an item and use the **Up** or **Down** arrow buttons on the right of this pane.

Report Configuration Pane

The **Report Configuration** pane displays the properties of the selected report element. The actual settings depend on the element; however, there are a number of common elements:

General

Id	Each report element is automatically given a unique identifier (Id). This identifier is used to identify the report element when configuring custom report templates (It is also used for advanced settings such as custom export and XML configuration). For general use, it should not be necessary to change the name of the identifier.
Include	This checkbox can be used to exclude the report element (without deleting the settings).

Title	Optionally provided title for the report element. When specified, the title appears immediately before the element.
	This property can refer to report macros (such as **{PlateNum}** and **{AnalyteName}**) to automatically refer to the data that it is associated with.
	If a title is not specified, then the element is displayed without any title.

Sheet

Sheet Name	This setting defines the name of a sheet to insert the contents into. This is only applicable to sheet-based content such as Excel output or data in the **Sheet View**.
	Each report element can be included on its own separate sheet or elements can be added together on the same sheet. Where a common sheet name is used across report elements, the elements are added in the order as defined in the **Report Elements** pane.
	This property can refer to report macros (such as **{PlateNum}** and **{AnalyteName}**) to automatically group associated content into a sheet of the same name.
	Note, if **Sheet Name** is defined for one report element, then the report content is exported on one sheet with provided **Name**.

> When viewing report content in **Sheet View**, the sheet names appear in the bottom left of the **Report** tab (in the **Results** tab). If there is only one named sheet, then this sheet name will not appear.

Report Elements and Multiple Plate or Multiplex Analyses

If your assay comprises of measurements across multiple plates or measurements for multiple analytes (multiplex assay), then the report will contain an element for each plate or analyte.

To help arrange this type of content into an easy-to-navigate report, it is recommended to use the **Sheet Name** setting; this can be used to arrange content into separate sheets.

For example, with a multiple plate assay, set the **Sheet Name** setting to:

Plate {PlateNum}

With this setting applied to a Sample Table element, the results for each plate will appear in a separate sheet tab named on by the plate number (i.e., Plate 1, Plate 2, Plate 3, etc.).

Report Macros

The following table lists the macros that are available for use in the **Title** and **Sheet Name** properties.

Macro	Expands To
{PlateNum}	1-based plate number
{ContainerNum}	1-based container number (for a multiple plate assay, this is the plate number; for multiplex, this is the analyte number)
{PlateName}	The plate name (as it appears under the **Measurements** | **Plate IDs**)
{AnalyteName}	The analyte name (as it appears under the **Measurements** | **Analytes**)
{ContainerName}	For a multiple plate assay, this is the plate name; for multiplex, this is the analyte name

> These macros can be used in conjunction with other text (e.g., **Plate {PlateNum}** would be expanded to **Plate 1** for the first plate).

Report Elements

To simplify configuration, various report elements are automatically added to the report and configured with default settings as you add new steps to your analysis:

- The Sample Table is the most widely used report element, so it is included by default in all configurations. The Sample Table displays the results of each defined sample group (in accordance with the defined layout).

- If any Evaluation or Validations are configured, an Evaluation or Validation report element will be added automatically.

- If any transform generates additional content, a Transform Content element will automatically be added. For example, the Standard Curve Fit transform produces a standards chart, concentrations table, and equations; this content is automatically added to the report as a Transform Content report element.

You will see these elements appear automatically in the **Report Elements** pane of the **Report** tab. You are free to edit or delete this default content as you require. If analysis steps are removed, then the associated report element will be automatically removed too.

Beyond the automatically added report elements, you can further add your own elements to the report:

How to: Add a Report Element to the Report

1. Open the **Report** tab (If the **Report** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Select the required report element from the **Add Element** drop-down list in the **Report Elements** pane.

3. Review and edit the element's settings as required.

4. Press the **Calculate** button to perform calculations with the new settings.

The following table summarises each available report element. Further details and settings are included in the provided linked topics.

Report Element	Description
Evaluations Table	The details and results of any configured evaluations arranged in a table.
Histogram	The Histogram represents the data by displaying the average of each sample in vertical columns. Each sample is denoted by a bar of the color of its sample type. In addition, to indicate the error or uncertainty in a reported measurement or calculated data, the variability of data can be represented with optional error bars.
Layout	A graphical representation of the sample layout.
Matrix Table	The results displayed in a matrix-table structure. For endpoint data, a numerical value is reported. For XY data, a thumbnail plot is displayed. Each raw or calculated matrix can be marked for inclusion or exclusion in a matrix table using the **Report Contents** section of the Matrices tab.
Matrix View	A layout-based graphical representation of the matrix data. For endpoint data, a numerical value will appear at each position. For XY data, a plot will appear at each position. The color of each position can be colored using the layout color or with a heatmap to represent the endpoint value intensity.
Measurement Meta Data	A table listing the measurements meta data (reported as name value pairs). This corresponds to the data as it appears under the **Measurements** | **Meta** tab.
Notes	Any notes associated with the assay run. Notes can be entered using the **Notes** button in the **View** section of the **Home** tab.
QC Results	Inter-assay quality control results (as defined by Quality Control settings). This could include Observation Table, Levey-Jennings, or CUSUM charts reporting the latest results along with historical results.

Report Element	Description
Sample Table	A table reporting the results of each defined sample group (in accordance with the defined layout). Each row contains the data for a specific sample group.
	For endpoint data, a numerical value is reported. For XY data, a thumbnail plot is displayed.
	Each raw or calculated matrix can be marked for inclusion or exclusion in a sample table using the **Report Contents** section of the Matrices tab.
Text	A textual information with optional inclusion of macros.
Transform Content	Various report elements generated as a consequence of a specific transform.
	For example, a Standard Curve Fit transform adds a concentration table, a chart, and equations to the report as Transform Content report elements.
Validations Table	The details and results of any configured validations arranged in a table. Each validation result is reported as a pass or fail.

Selecting the Data to Include in a Sample Table/Matrix Table

The data to include in a Sample Table or Matrix Table can be configured on a matrix-by-matrix basis. For example, you may want to include only certain matrices in your tables, perhaps excluding the results of intermediary data analysis steps.

The easiest way to specify which data to include in your Sample Tables or Matrix Tables is to use the matrix options: **Include in Matrix Tables** and **Include in Sample Tables**. These settings are available for each matrix. To access these settings, select the **Matrices** tab and select the matrix to edit. These settings apply to all of the Sample Tables or Matrix Tables in your report.

> For each matrix, the **Sample Table Replicates** setting can be used to specify how to report the values for replicates in the Sample Table on a matrix-by-matrix.

Working with Multiple Sample Tables or Multiple Matrix Tables

You may want to include multiple Sample Tables or Matrix Tables in your report with each reporting different aspects of the data.

When working with multiple tables, you might require control over which data to include in which Sample Tables or Matrix Tables. The **Sample Table IDs** and **Matrix Table IDs** settings can be used to select which tables each matrix should be included in. The table Ids correspond

to the report element Ids of the corresponding Sample Table or Matrix Id (as defined in the **Report** tab).

> Each table could be configured to report data for different sample types using the [Sample Table](#)'s **Type** setting.

Report Elements

Evaluations Table

The Evaluations Table report element includes in the report details and results of any configured evaluations arranged in a table, which typically consists of three columns. For example,

Description	Expression	Results
Evaluation #1	Unknown	0.826
Evaluation #2	median(Unknown)	0.649
Median of all Unknown	median(Unknown)	0.649

Here, the **Description** column contains **Expression #{index}** in the first and the second rows and the specified description in the third. The **Expression** column contains the evaluated expression and the **Results** display the computed result of each configured Evaluation.

Settings

Depending on the configuration of the Evaluation settings, the results are arranged in one or more Evaluations Tables. The number of rows in each table corresponds to the number of configured Evaluations.

Evaluations Table

While the **Results** are definitely included, you might prefer to hide other columns by unchecking corresponding checkboxes.

Include Description	Include the description column.
Include Expression	Include the expression column.
Include Header	Determines whether to include the table header.

The Evaluation Table is also configured with common report settings that are covered in the Report Content section.

How to: Add Evaluations Table Element to the Report

1. Open the **Report** tab (If the **Report** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Select the Evaluations Table element from the **Add Element** drop-down list in the **Reports** pane.

3. Review and edit the settings as required.

4. Press the **Calculate** button to perform calculations with the new settings.

Histogram

The Histogram report element includes in the report a chart that graphically presents the spread of measurements or results. This can be useful for spotting outliers.

For example,
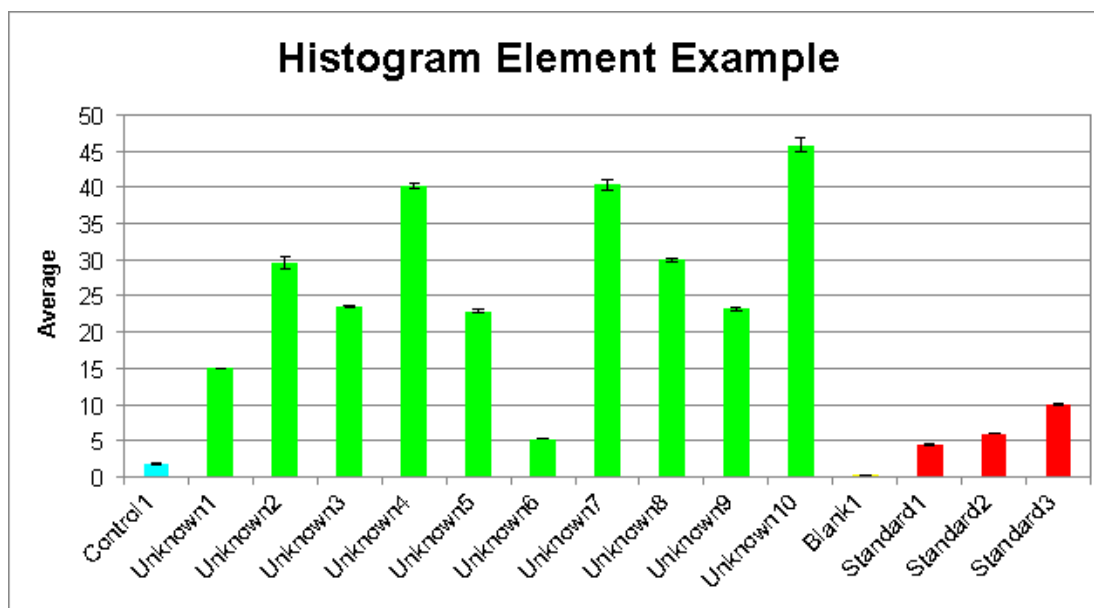
Here the Histogram was generated using a configuration to plot the Average values of **Control**, **Unknowns**, **Blank** and **Standards** samples with error bars indicating the standard deviation and a custom title.

Settings

The Histogram is configured with the common report settings covered in the Report Content section plus the Histogram specific settings described below.

General

Matrix	The matrix of data to plot. Select the relevant matrix name from a drop-down list.

Each histogram can be configured to plot samples of one or more types (e.g., Standards, Unknowns, etc.). The values of each sample group is plotted as a coloured bar on the x-axis. The colour of each sample bar matches its sample type with a label to indicate the sample group name (or any applied Sample ID).

Histogram

Error Bars	Use this setting to select the type of error bars to use (or none).
	Error bars can display the **Standard Error**, **%CV**, or **Standard Deviation** of the (non-flagged) replicates of the group. If the **Replicates** option is selected

	the error bars reflect the minimum and maximum value of the replicates of the sample. With the **None** selection, the error bars are not shown.
Title	The title for a histogram chart.
Sample Types	Which sample types to include in the histogram.

How to: Add Histogram Element to the Report

1. Open the **Report** tab (If the **Report** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Select the Histogram element from the **Add Element** drop-down list in the **Reports** pane.

3. Review and edit the settings as required.

4. Press the **Calculate** button to perform calculations with the new settings.

Layout

The Layout report element includes in the report an image of the sample layout used in the assay. This includes a layout itself and a legend with the sample types that are included in the layout. For example:
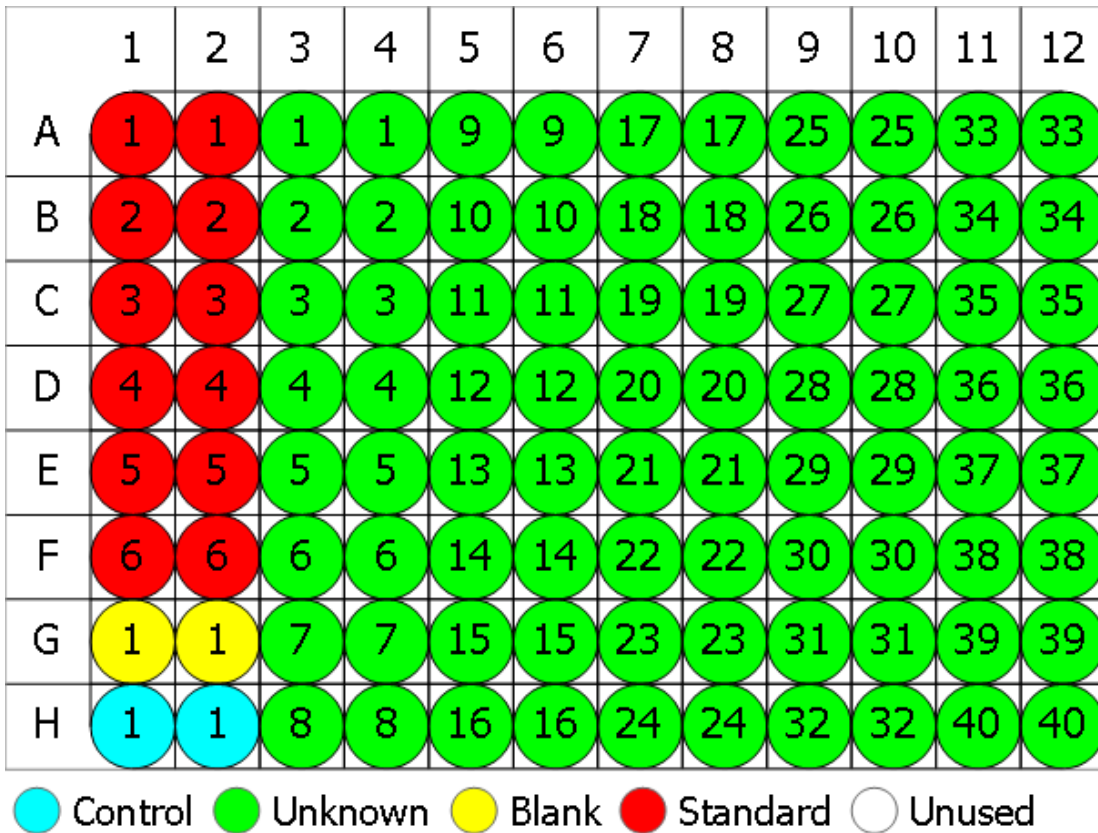
Here is the layout for a 96-well microplate with 6 **Standard**, 1 **Blank**, 1 **Control**, and 40 **Unknown** samples in duplicate. Under the layout image, a sample types collection of the assay is displayed.

Settings

Layout report element includes the following configurable setting:

General

Size	**Small**, **Medium**, or **Large** (medium size will fit on an A4 page-width).

> If multiple layouts are defined in the assay protocol, then all of them will be included in the report.

As with other Report Elements, you can configure how it appears in the report using common report settings.

## How to: Add Layout Element to the Report

1. Open the **Report** tab (If the **Report** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Select the Layout element from the **Add Element** drop-down list in the **Reports** pane.

3. Review and edit the settings as required.

4. Press the **Calculate** button to perform calculations with the new settings.

Matrix Table

The Matrix Table report element includes in the report the results of the performed analysis in a matrix-table structure. This is a table matching the layout dimensions with the data reported in each cell corresponding to the associated position on the matrix. Flagged data is displayed as defined by the associated [transform](#) and [matrix configuration](#). Each cell in the table corresponds to the associated position on the matrix.
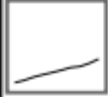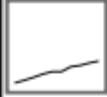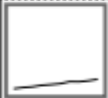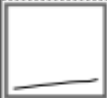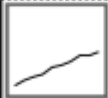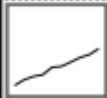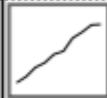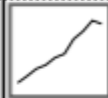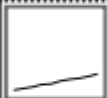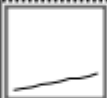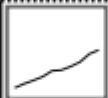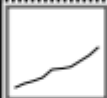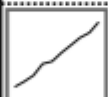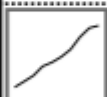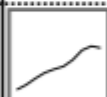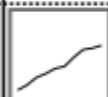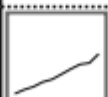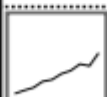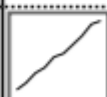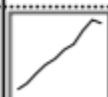
For example,

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

Here is the Matrix Table is reported for an endpoint matrix. In each cell, the value for a corresponding position is displayed. For XY data, each position would contain a thumbnail plot.

All matrices could be reported separately (i.e., a separate Matrix Table for each included matrix) or in a combined arrangement.

For example,

	1	2	3	4	5	6
A	4.318	4.460	15.053	15.003	29.063	30.118
B	6.045	6.003	23.532	23.636	40.364	39.902
C	9.923	10.092	22.744	23.132	5.246	5.273
D	1.809	1.840	40.937	39.919	29.764	30.177
E	0.057	0.057	23.440	22.989	45.226	46.526
Key:	Raw Average					

Here is a snippet of a Matrix Table for the XY Reduction Average raw and calculated data, where each cell contains the thumbnail plot for corresponding input XY position and the calculated numerical result. The last row features the key explaining the arrangement in each position.

Matrix Table with Multiple Plate/Multiplex

If the assay defines multiple plates then matrix tables are included for each of them. Each set of matrices for each plate is prefixed with the plate name and number (e.g., "Plate 1"), then the matrices are included for that matrix.

> With **Combined**, there will be one matrix table for each plate.

> With **Separate**, there might be more than one matrix table for each plate.

Settings

The Matrix Table is configured with specific to this report element and with general report configuration settings.

Matrix Table

The matrix table has 2 configurations that define how the results will be displayed.

Arrangement	Select **Combined** for each cell in the table to list the values of all included

	matrices (per plate/analyte). In this case, the final row of the table will contain a key that explains ordering of the matrices.
	Or select **Separate** for a separate table for each included matrix.

For XY data, there are additional settings for configuring how the thumbnail plots are displayed in the report.

Thumb Plot

For XY data, various options for defining the inclusion and sizing of XY thumb plot charts are available.

Include Thumb Plots	Defines whether thumb plots are included in the report.
Thumb Plot Height	This specifies the height to use for all thumbs in the report.
Thumb Plot Width	This specifies the width to use for all thumbs in the report.

For [XY data](#), various options for defining how XY thumb plot charts are scaled. These can be fixed values (or determined for each position). The plot lines are plotted within this scan.

Thumb Scale

Thumb Auto Scale Max X	Specifies whether the maximum X scale of each plot is: • **Fixed** – The scale limit of the thumb plots used in the table is specified by the associated **Thumb Scale Max X** value. • **Plot** – The scale limit of each thumb plot is automatically determined by the maximum X data in each plot (i.e., each plot may have a different maximum X limit). • **Table** – The scale limit of every thumb plot in the table is the same. This is determined from the maximum X of all plots data points in the table.
Thumb Auto Scale Max Y	Specifies whether the maximum Y scale of each plot is: • **Fixed** – The scale limit of the thumb plots used in the table is specified by the associated **Thumb Scale Max Y** value; • **Plot** – The scale limit of each thumb plot is automatically determined by the maximum Y data in each plot (i.e., each plot may have a different maximum Y limit).

	• **Table** – The scale limit of every thumb plot in the table is the same. This is determined from the maximum *Y* of all plots data points in the table.
Thumb Auto Scale Min X	Specifies whether the minimum *X* scale of each plot is: • **Fixed** – The scale limit of the thumb plots used in the table is specified by the associated **Thumb Scale Min X** value. • **Plot** – The scale limit of each thumb plot is automatically determined by the minimum *X* data in each plot (i.e., each plot may have a different minimum *X* limit). • **Table** – The scale limit of every thumb plot in the table is the same. This is determined from the minimum *X* of all plots data points in the table.
Thumb Auto Scale Min Y	Specifies whether the minimum *Y* scale of each plot is: • **Fixed** – The scale limit of the thumb plots used in the table is specified by the associated **Thumb Scale Min Y** value. • **Plot** – The scale limit of each thumb plot is automatically determined by the minimum *Y* data in each plot (i.e., each plot may have a different minimum *Y* limit). • **Table** – The scale limit of every thumb plot in the table is the same. This is determined from the minimum *Y* of all plots data points in the table.
Thumb Scale Max X	Determines custom maximum *X* scale for thumb plots if **Thumb Auto Scale Max X** is **Fixed**.
Thumb Scale Max Y	Determines custom maximum *Y* scale for thumb plots if **Thumb Auto Scale Max Y** is **Fixed**.
Thumb Scale Min X	Determines custom minimum *X* scale for thumb plots if **Thumb Auto Scale Min X** is **Fixed**.
Thumb Scale Min Y	Determines custom minimum *Y* scale for thumb plots if **Thumb Auto Scale Min Y** is **Fixed**.

How to: Add Matrix Table Element to the Report

1. Open the **Report** tab (If the **Report** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Select the Matrix Table element from the **Add Element** drop-down list in the **Reports** pane.

3. Review and edit the settings as required.

4. Press the **Calculate** button to perform calculations with the new settings.

Matrix View

The Matrix View report element includes in the report a layout-based graphical representation of the matrix data. All corresponding settings of the associated matrix are applied to the view too. Flagged positions are indicated and displayed with a cross-through it.

Matrix view can be output with coloring of :

- **Layout** – Each position shows the numeric value in endpoint matrices or a thumb in XY data according to its sample type. Also, the sample type legend is included.

For example,
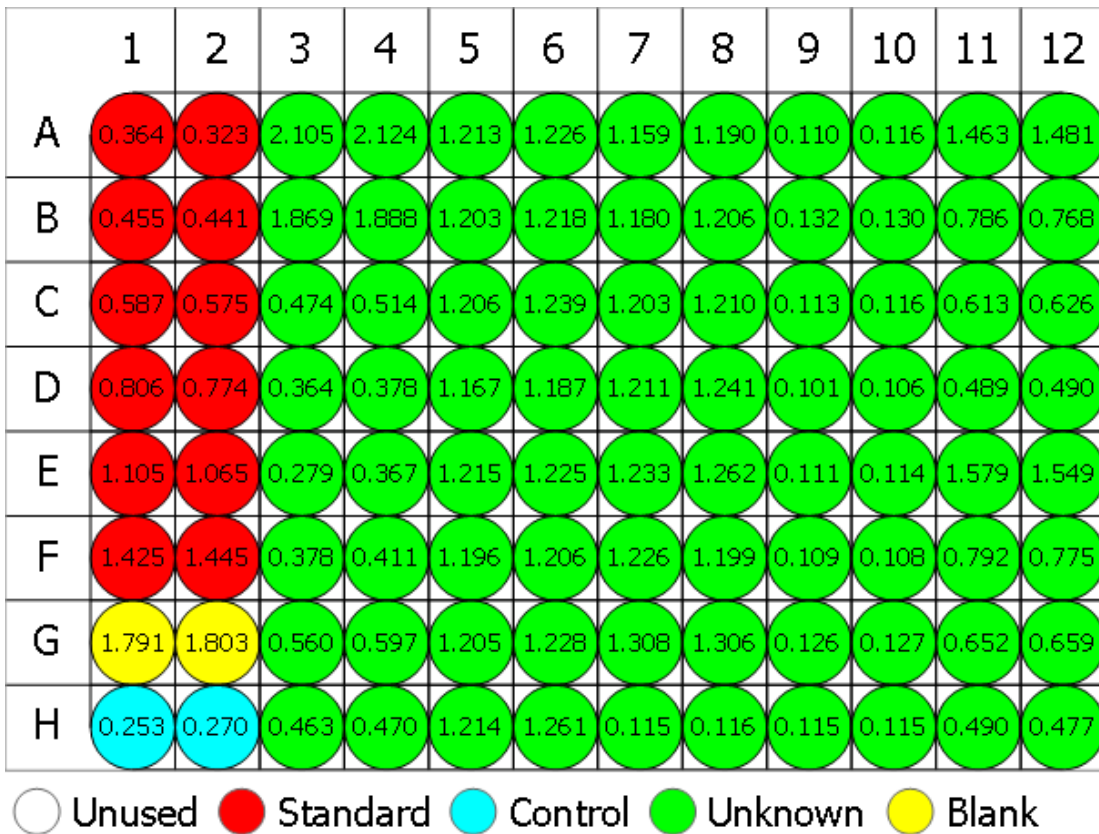
- **Heatmap** – Each position displays the numeric value with respect to the default or custom heatmap scale. The scale legend is also included.

> ℹ️ This setting is relevant for endpoint data only.

For example,

Settings

Matrix View includes the following configurable settings:

General

Matrix	The matrix of data to plot. Select the relevant matrix name from a drop-down list.
Size	**Small**, **Medium**, or **Large** (medium size will fit on an A4 page-width).

Matrix View

Colouring	**Layout** or **Heatmap** (Heatmap setting does not apply to XY data).

The Matrix View is also configured with common report settings that are covered in the Report Content section.

How to: Add Matrix View Element to the Report

1. Open the **Report** tab (If the **Report** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Select the Matrix View element from the **Add Element** drop-down list in the **Reports** pane.

3. Review and edit the settings as required.

4. Press the **Calculate** button to perform calculations with the new settings.

Measurement Meta Data

The Measurement Meta Data report element includes in the report a table listing the measurements meta data (reported as name value pairs). This corresponds to the data as it appears under the **Measurements | Meta** tab.

This data is reported in a two-column table with the name of each entry and its value. For example,

Data Filename	C:\...\Documents\MyAssays\Measurements\ELISA Absorbance.xlsx
Data Main Filename	ELISA Absorbance
Data File Created	7/10/2017
Data File Last Modified	7/10/2017
MEASUREMENT START DATE	Tue Jun 13 2017 13:02:29 GMT+0100 (GMT Summer Time)
MEASUREMENT COMMENT	Comment On Multiple Lines
User interface program version	3.1.1.9
Instrument interface version	0
Instrument serial number	0
PROTOCOL NAME	ELISA Absorbance
PROTOCOL CREATOR DATE	FACTORY PROTOCOL
PLATE TYPE	PerkinElmer OptiPlate
PLATE FORMAT	96 wells (8X12)

Here the meta data table features details about the measurements data file and the details obtained from the VICTOR plate reader.

Settings

The Measurement Meta Data is configured with common report settings that are covered in the Report Content section.

How to: Add Measurement Meta Data Element to the Report

1. Open the **Report** tab (If the **Report** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Select the Measurement Meta Data element from the **Add Element** drop-down list in the **Reports** pane.

3. Review and edit the settings as required.

4. Press the **Calculate** button to perform calculations with the new settings.

Notes

The Notes report element includes in the report any provided notes (e.g., preparation notes, Lot ID, etc.). Notes can be used to reference the peculiarities of this data analysis run.

How to: Add Notes to the Analysis

1. Launch an assay protocol or open a results file.

2. On the **Home** tab's **View** group, click on the **Notes** button.

3. The **Notes** panel will appear down-most of the screen.

4. Enter or paste any Notes as required.

Entered Notes will appear under **Notes:** title. For example,

Notes:

An eyelash contaminated well B4.

Settings

Notes

Notes element includes the following checkbox setting:

Exclude If Empty	Exclude Notes from the results, if it contains no information.

The Notes element is also configured with common report settings that are covered in the Report Content section.

How to: Add Notes Element to the Report

1. Open the **Report** tab (If the **Report** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Select the Notes element from the **Add Element** drop-down list in the **Reports** pane.

3. Review and edit the settings as required.

4. Press the **Calculate** button to perform calculations with the new settings.

QC Results

The QC Results report element includes in the report any inter-assay quality control results (as defined by Quality Control settings) defined for an assay. This could include Observation Table, Levey-Jennings or CUSUM charts, and QC summary reporting the latest results along with historical results.

For example,
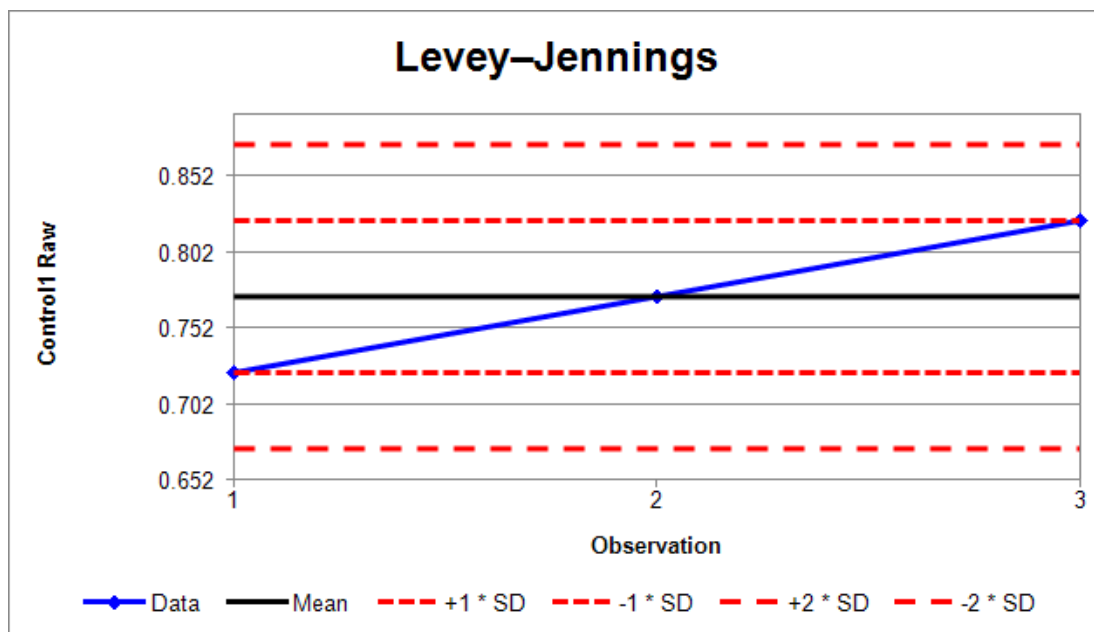
Observation	Control1 Raw	Date	File
3 (Current)	**0.823**	**2017-07-01 / 16:34:07**	**ELISA (3)**
2	0.733	2017-07-01 / 12:32:04	ELISA (2)
1	0.722	2017-07-01 / 11:54:02	ELISA (1)

✔ All observations are within the ±2SD range.

Here the Levey-Jennings chart, Observation Table and Observation summary are reported for current and two previous results of an ELISA assay protocol.

Settings

The QC Results element is configured with common report settings that are covered in the Report Content section.

How to: Add QC Results Element to the Report

1. Open the **Report** tab (If the **Report** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Select the QC Results element from the **Add Element** drop-down list in the **Reports** pane.

3. Review and edit the settings as required.

4. Press the **Calculate** button to perform calculations with the new settings.

Sample Table

The Sample Table report element lists the results of the performed data analysis by sample groups defined on the layout. Flagged data is displayed as defined by the associated [transform](#) and [matrix configuration](#). Each row in the table corresponds to a sample.

Here is an example of a Sample Table:

Sample	Positions	My Raw Data	%CV
🔴 Standard1	A1	0.364	8.440
	A2	0.323	
🔴 Standard2	B1	0.455	2.210
	B2	0.441	
🔴 Standard3	C1	0.587	1.460
	C2	0.575	
🔴 Standard4	D1	0.806	2.864
	D2	0.774	
🔴 Standard5	E1	1.105	2.607
	E2	1.065	

It includes snippet of five Standard samples and the positions they are designated to. Also, the raw values (**My Raw Data** column) for each replicate and the calculated results per sample for the [Percentage Coefficient of Variation](#) transform (**%CV** column) are included. Each row contains replicates of the corresponding sample in the **Vertical** orientation.

For XY data, the Sample Table contains **Thumb Plots** that is simply a thumbnail graphical representation of each kinetic/spectral data plot at a position.

Sample	Positions	Raw	Average
🟢 Unknown1	A1 A2 A3 A4 A5 A6		
🟢 Unknown2	C1 C2 C3 C4 C5 C6		
🟢 Unknown3	E1 E2 E3 E4 E5 E6		
🟢 Unknown4	G1 G2 G3 G4 G5 G6		
🟡 Blank1	B1 B2 B3 B4 B5 B6		
🟡 Blank2	D1 D2 D3 D4 D5 D6		
🟡 Blank3	F1 F2 F3 F4 F5 F6		
🟡 Blank4	H1 H2 H3 H4 H5 H6		

Here the replicates for each sample group, XY Raw data and calculated results for [XY Replicates Average](#) transform are displayed in **Horizontal** orientation.

Settings

The Sample Table is configured with general report and specific to this element settings. The former are described in general report configuration settings and the latter are covered below.

Sample Table

Include Positions	Include the **Positions** column to report the sample positions where the sample was measured. For plate assays, this is checked by default (with **Include Sample Numbers** setting unchecked).
Include Sample Numbers	Include a column titled **Number** to report the 1-based position number for each sample. For rack assays, this is checked by default (with **Include Positions** setting unchecked). ⓘ If both **Include Positions** and **Include Sample Numbers** are checked, the latter appears to the right of the former).
Replicate Orientation	How the replicates appear in the table: • **Vertical** – Each position is listed in a new line of the column. • **Horizontal** – All positions are listed in the same line.
Sample Types	Which sample types to include in the sample table.

For XY data, there are additional settings for configuring how the thumbnail plots are displayed in the report.

Thumb Plot

For XY data, various options for defining the inclusion and sizing of XY thumb plot charts are available.

Include Thumb Plots	Defines whether thumb plots are included in the report.
Thumb Plot Height	This specifies the height to use for all thumbs in the report.
Thumb Plot Width	This specifies the width to use for all thumbs in the report.

For XY data, various options for defining how XY thumb plot charts are scaled. These can be fixed values (or determined for each position). The plot lines are plotted within this scan.

Thumb Scale

Thumb Auto Scale Max X	Specifies whether the maximum *X* scale of each plot is: • **Fixed** – The scale limit of the thumb plots used in the table is specified by the associated **Thumb Scale Max X** value. • **Plot** – The scale limit of each thumb plot is automatically determined by the maximum *X* data in each plot (i.e., each plot may have a different maximum *X* limit). • **Table** – The scale limit of every thumb plot in the table is the same. This is determined from the maximum *X* of all plots data points in the table.
Thumb Auto Scale Max Y	Specifies whether the maximum *Y* scale of each plot is: • **Fixed** – The scale limit of the thumb plots used in the table is specified by the associated **Thumb Scale Max Y** value; • **Plot** – The scale limit of each thumb plot is automatically determined by the maximum *Y* data in each plot (i.e., each plot may have a different maximum *Y* limit). • **Table** – The scale limit of every thumb plot in the table is the same. This is determined from the maximum *Y* of all plots data points in the table.
Thumb Auto Scale Min X	Specifies whether the minimum *X* scale of each plot is: • **Fixed** – The scale limit of the thumb plots used in the table is specified by the associated **Thumb Scale Min X** value. • **Plot** – The scale limit of each thumb plot is automatically determined by the minimum *X* data in each plot (i.e., each plot may have a different minimum *X* limit). • **Table** – The scale limit of every thumb plot in the table is the same. This is determined from the minimum *X* of all plots data points in the table.
Thumb Auto Scale Min Y	Specifies whether the minimum *Y* scale of each plot is: • **Fixed** – The scale limit of the thumb plots used in the table is specified by the associated **Thumb Scale Min Y** value. • **Plot** – The scale limit of each thumb plot is automatically determined by the minimum *Y* data in each plot (i.e., each plot may have a different minimum *Y* limit). • **Table** – The scale limit of every thumb plot in the table is the same. This is determined from the minimum *Y* of all plots data points in the table.

Thumb Scale Max X	Determines custom maximum *X* scale for thumb plots if **Thumb Auto Scale Max X** is **Fixed**.
Thumb Scale Max Y	Determines custom maximum *Y* scale for thumb plots if **Thumb Auto Scale Max Y** is **Fixed**.
Thumb Scale Min X	Determines custom minimum *X* scale for thumb plots if **Thumb Auto Scale Min X** is **Fixed**.
Thumb Scale Min Y	Determines custom minimum *Y* scale for thumb plots if **Thumb Auto Scale Min Y** is **Fixed**.

XY

Data Format	Specifies how XY data is included in the report: • **Thumb** – If **Include Thumb Plots** is checked, a thumb plot is reported for each plot. This is the default option. • **Y Values** – A list of *Y* data point values is reported for each plot. • **X and Y Values** – A list of *X* and *Y* data point values is reported for each plot. • **Index and Y Values** – For each plot, this reports a list where *X* are displayed as 1-based continuous data point numbers and *Y* as data point values.

Advanced Row Ordering Settings

You might prefer to alter the default order in which the samples are presented in the report. For example, in some cases it might be convenient to view the analysis results in descending order, based on results of the Standard Curve Fit transform.

This is an advanced feature that can only be configured through the XML editor.

To set row ordering:

1. Open the XML tab. (If it is not immediately visible, press the **<< Show All Properties** button to expand.)

2. Inside the **Report** element, locate the **SampleTable** child element.

3. Add and configure row ordering attributes as required.

4. Press **Apply** to save changes and then **Calculate** to generate a new report.

Row Ordering Attributes

The following table lists the attributes of the **SampleTable** element that must be specified to define the ordering of rows:

OrderByColumn	Specifies a 1-based number of a column (starting from leftmost) to sort the rows by. If the specified column is displaying **List** of Sample Table Replicates, then the value of the first replicate is used to sort each row by.
OrderBySequence	Specifies the order sequence, based on a **OrderByColumn** setting: ● **Ascending** – For numerical values, the results are listed from the lowest to the highest value. For textual content, the sorting is performed in the alphabetical order. ● **Descending** – For numerical values, the results are listed from the highest to the lowest value. For textual content, the sorting is performed in the reverse alphabetical order. If not specified , the **Ascending** sequence is assumed.

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. For this data, the Blank Correction, Standard Curve Fit, and Percentage Coefficient of Variation transforms are configured to illustrate advanced row ordering.

By default, the Sample Table presents data consequently for each included **Sample Types** , e.g.:

Sample	Positions	Raw	Blank Corrected	Concentration	%CV
● Standard1	A1 A2	0.192 0.199	0.195	~~Flagged~~	~~Flagged~~
● Standard2	B1 B2	0.293 0.300	0.296	182.429	3.173
● Standard3	C1 C2	0.537 0.534	0.535	87.476	0.387
● Standard4	D1 D2	0.858 0.855	0.856	55.188	0.261
● Standard5	E1 E2	1.523 1.527	1.524	20.922	0.700

In this example, we sort rows by Blank Correction results from lowest to highest values. To configure the ordering, enter the **Blank Corrected** column number (4) in **OrderByColumn** and **Ascending** in **OrderBySequence**.

The following XML snippet shows a configuration of a Sample Table with the row ordering attributes:

```
<SampleTable Id="SampleTable1" Types="2,4,5,3" OrderByColumn="4" Order-
BySequence="Ascending" />
```

With the example configuration provided above, the rows are sorted as follows:

Sample	Positions	Raw	Blank Corrected	Concentration	%CV
⬤ Blank1	G1	0.001	0.000	~~Flagged~~	~~Flagged~~
	G2	0.001			
⬤ Standard1	A1	0.192	0.195	~~Flagged~~	~~Flagged~~
	A2	0.199			
⬤ Unknown33	A11	0.241	0.243	327.389	7.615
	A12	0.246			
⬤ Control1	H1	0.297	0.296	181.711	0.000
	H2	0.297			
⬤ Standard2	B1	0.293	0.296	182.429	3.173
	B2	0.300			

How to: Add Sample Table Element to the Report

1. Open the **Report** tab (If the **Report** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Select the Sample Table element from the **Add Element** drop-down list in the **Reports** pane.

3. Review and edit the settings as required.

4. Press the **Calculate** button to perform calculations with the new settings.

Text

The **Text** report element includes any specified text and/or the evaluation of any available text macros.

This element is useful to include custom wording in the report. Also, text macros can be used to include data that may change from run-to-run of the assay. Available text macros include:

- Any imported meta-data
- Filenames (protocol, results, imported data file)
- Current date/time
- Calculation version number
- User name
- PC name

For example, with the following **Text** configuration:

>Protocol: <ProtocolName>
>
>Imported Data: <DataMainFilename>
>
>Results: <ResultsName>
>
>Date: <CurrentDateLong>

In the report, the macros will convert to:

>Protocol: ELISA (96 well)
>
>Imported Data: 96 wells_20170613_152121
>
>Results: ELISA (96 well) (21)
>
>Date: 10 February 2018

Settings

Text

Text	Click on the drop-down to expand the text editor. The editor consists of: - A multiline edit box where you can enter or paste any required information. - A full list of all available macros and their current value. To insert a macro at current cursor position, double-click on it in the list.

The Text element is also configured with common report settings that are covered in the Report Content section.

How to: Add Text Element to the Report

1. Open the **Report** tab (If the **Report** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Select the Text element from the **Add Element** drop-down list in the **Reports** pane.

3. Review and edit the settings as required.

4. Press the **Calculate** button to perform calculations with the new settings.

Transform Content

The Transform Content report element includes additional report content to illustrate settings or results of a specific transform. If the transform applied to the assay possesses any kind of optional items, they are added to the report automatically. Certain transforms comprise a number of optional content.

The additional report elements are:

- **Additional Sample Table Column** – This is an optional additional column that can be included in the sample table. For example,

Sample	Positions	Factor	My Raw Data	Concentration (x Dil. Factor)
● Unknown1	A3	2	2.105	4.210
	A4		2.124	4.248
● Unknown2	B3	5	1.869	9.345
	B4		1.888	9.440
● Unknown3	C3	2	0.474	0.948
	C4		0.514	1.028
● Unknown4	D3	5	0.364	1.820
	D4		0.378	1.890
● Unknown5	E3	2	0.279	0.558
	E4		0.367	0.734

  Here is the Dilution Factors transform outputs optional report content named Factor Column.

- **Chart** – The visual representation of a plotted curve that is included in a report by default. E.g.:
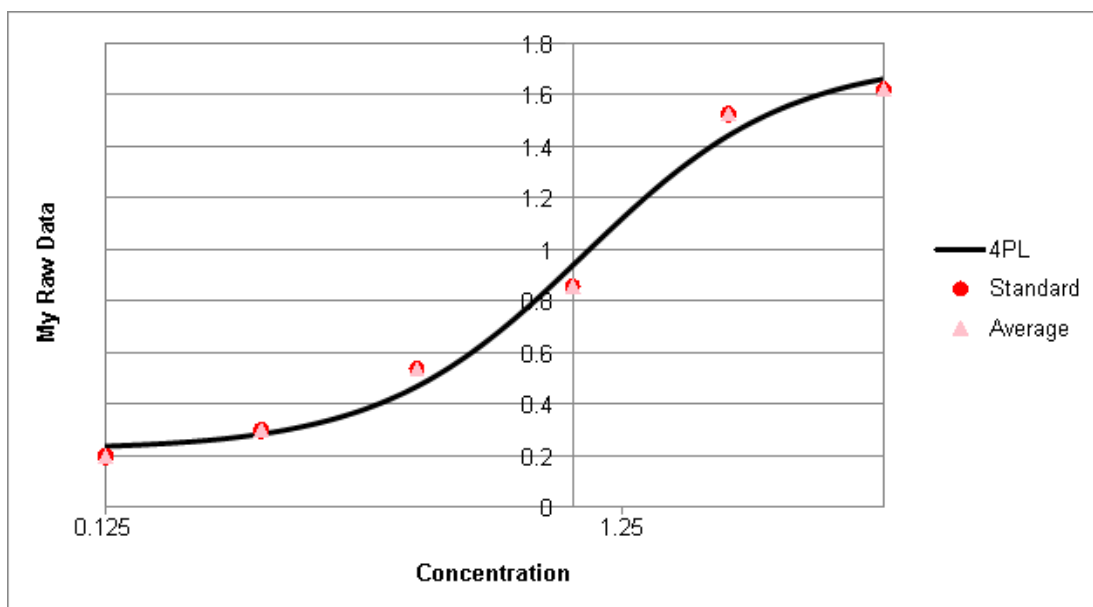
  Here is a chart for Standard Curve Fit transform example.

- **Table** – The table containing additional values computed from a transform. For example, this might refer to Goodness Measures Table, Dose–Response Curves Table, Curve Data Table, etc.

- **Equation** – The formula of a fit method that was used to plot a curve. E.g.:

$$y = mx + c$$

Here is the formula for Linear Regression.

Settings

A report can contain one or more **Transform Content** sub-elements.

The Transform Content is configured with common report settings that are covered in the Report Content section and the setting described below.

Content	Use this setting to specify which sub-elements to include (or exclude) and their ordering. The available sub-elements depend on the associated transform.

You can configure which optional items are actually included in report with the **Content Editor**. Select the sub-element, move in to the Included/Excluded pane with right/left arrow buttons, or change its' positioning with the **Up** and **Down** icon buttons on the right of the Included pane.

To include or exclude all transform content from the report, use the **Add All** or **Remove All** buttons, respectively.

How to: Configure Transform Content in the Report

1. Open the **Report** tab.

2. Select Transform Content element, which contains transform name in the title, e.g., **TransformContentDoseResponseCurvesTable1**.

3. Click on a drop-down of **Content** option of the **Report Configuration** pane.

4. Review and edit content items as required.

5. Press **Calculate** button to perform calculations with the new settings.

Validations Table

The Validations Table report element includes the details and results of any configured validations arranged in a table. This table indicates whether each validation has passed or failed and, when validations are defined, is added to the report automatically.

The Validation Table typically consists of three columns. For example,

Results	Expression	Description
❌ Fail	x < 2.5	All non-flagged positions have a value less than 2.5 (**Expression with Every Sample Replicate (x)** type)
✔ Pass	x < 2.5	The average of the replicates (that are not flagged) of all sample groups is less than 2.5 (**Expression with Every Sample Group (x)** type)
✔ Pass	Unknown1 > 1.5	Condition #3
❓ Syntax Error	Control1 !< 1.5	Condition #4

Here, the **Results** column displays the evaluated result of each configured Validation. For the Fail and Inconclusive (i.e., Syntax Error), the row is highlighted with yellow and red background color, respectively. The **Expression** column contains the verified condition. Note, that the expression for the first and the second validation is the same, but the type of validations is different, as stated in the Description. Finally, the **Description** column contains the description if specified (e.g., on third row) or **Condition#{index}** if it is not.

Settings

Depending on the configuration of the Validation settings, the results are arranged in one or more Validations Tables. By default, the Validation table is displayed as three-column table with the number of rows corresponding to the number of **Conditions** added.

While the **Results** column is definitely included to the report, you might prefer to hide former columns by unchecking corresponding checkboxes.

Description Header	A title for a Validation Table in the report.
Include Description	When checked, the column contains **Description** if specified or **Condition#{index}** if it was not.
Include Expression	If included, this outputs the **Condition** defined for each **Validation**.
Results	This specifies whether **Results** column appears left-most or right-most in the

Column	table.
Results Header	A title for the **Results** column in the report.

The Validation Table is also configured with common report settings that are covered in the Report Content section.

How to: Add Validations Table Element to the Report

1. Open the **Report** tab (If the **Report** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Select the Validations Table element from the **Add Element** drop-down list in the **Reports** pane.

3. Review and edit the settings as required.

4. Press the **Calculate** button to perform calculations with the new settings.

Report Template

Play Video

Overview

The report template enables presenting your analysis results tailored to your laboratory's standards and requirements. This feature merges together report elements with a configured template. Full power of MS Word or WordPad features is available to customise page-based reporting of the results.

For example, you can define custom sequence and/or inclusion of report elements, insert relevant images or textual descriptions for a printed document. Moreover, simply apply styling settings and include the results you require for your current purpose.

> Report template affects Page View and page-based outputs (DOC, DOCX, PDF) of the analysis results only.

Report Template Configuration

How to: Add New Report Template for Page-Based Reporting

1. Launch an assay protocol or open a results file.

2. Select the Ribbon's **Report** tab and switch to the **Page View** of the results (If the **Report** tab is not immediately visible press the **<< Show All Properties** button to expand).

3. Hover to **Template** group and press **New** button. This launches the Template.DOCX file with default application (typically MS Word or WordPad).

4. Review and edit the reporting template content as required.

5. After making and saving changes to the report template press **Calculate** to generate a new report with the new settings.

The application displays a Template.DOCX file for editing – this is the template file. Initially, it contains the MyAssays report element identifiers (**Ids**) in curly braces.

Report templates have no restrictions with regard to the specific information they contain. You can make any changes to the template and save them. For example, reposition, remove or duplicate Ids, change their font/style/etc., add textual information, macros etc. In fact, any change is possible provided it is supported in the editor.

How to: Add Report Element Id to the Report Template

1. Launch an assay protocol or open a results file.

2. Open **Report** tab.

3. Select the Report Element from the list.

4. Copy the element's Id and insert it in the appropriate place of the template in curly brackets (e.g., **{SampleTable1}**).

5. Save changes to the report template and press **Calculate** to generate a new report with the new settings.

When you have made changes to the template file and saved them, you can switch back to MAA and press **Calculate** to generate a new report with the new settings while keeping the editor open. This way you can review modified report in Page View and make further changes if needed.

The defined template will be applied for all page-based reports for this assay results.

> Only one template can be defined for the assay results file at a time. If the previously defined template is no longer useful, edit or delete it.

Edit/Remove Existing Template

After you have created **New** template file, the **Template** group expands with two new options:

- Press **Edit** button to modify you template file's content according to freshly added report elements or updated requirements. It will launch your default .DOCX editor with the existing template file.

- Press **Delete** button to remove the existing template file (a warning message will prompt for confirmation before this action is completed).

Enhanced Security

Introduction

MyAssays Desktop Pro's Enhanced Security mode provides features compatible with working in GxP and 21 CFR Part 11 environments. This includes protected and tracked access to MyAssays Desktop Pro's features and data from inside a secured environment. The system can securely import, process, and output secure and protected content.

> This mode is only available to users with a MyAssays Desktop Pro Enhanced Security license.

Key features include:

- Access control – Authentication and authorization are used to permit/deny access to MyAssays Desktop functions

- Operating-system-level security data storage protection – Only authorized users can make changes to MyAssays Desktop Pro data and configuration files

- Secure hand-off of data – To ensure integrity of imported measurement data

- Secured file protection – To ensure that data files are only accessible by authorized users

- Audit trail and Login log – All activity is logged with date and time stamp against the logged in user

- Password-protected Excel output

The Enhanced Security system shares user management and system security policies with Windows to simplify user management and to eliminate redundant administrator tasks. For example, if your users are already configured on a domain, it is easy to provide those domain users with secured access to MyAssays Desktop Pro.

The Enhanced Security system provides a number of configuration options to support different workflows, system configurations, and data handling methods.

> Use of the Enhanced Security mode alone does not provide compliancy with 21 CFR Part 11. Any facility that requires compliance with 21 CFR Part 11 rules must implement the necessary procedures and controls specified by the regulation throughout the workplace.
> The US FDA does not preapprove or certify systems with respect to 21 CFR Part 11. (It is misleading for any vendor to claim that a system, whether it be an instrument or standalone software, is Part 11 compliant.)

Full compliancy is always a combination of both technological and procedural controls. The compliancy assessment is always made on-site by the FDA.

Video Content

Video tutorials showing walkthroughs of configuration and key features of the Enhanced Security system are available online, including:

- https://www.myassays.com/myassays-desktop-enhanced-security-authentication-methods-and-configuration.video
- https://www.myassays.com/myassays-desktop-pro-enhanced-security-with-envision.video

Authentication and Authorization

Throughout this documentation, the terms **authentication** and **authorization** are used; it is important that these terms be clearly defined and understood:

Authentication – The process by which users are identified by their username and password

Authorization – The actions that an authenticated user is allowed to perform

For example, when a user logs in to Windows by providing her username and password, she is authenticated. An authenticated user is authorized to perform certain operations. For example, an administrator is authorized to perform administrator operations whereas a standard user is not.

Authentication Options

With MyAssays Desktop Pro's Enhanced Security, there are two different authentication systems available:

- **Windows Log In**
- **MyAssays Desktop Log In**

The key difference is how the user must log in, either through the standard Windows Log In method or through a MyAssays Desktop Log In.

With **Windows Log In**, users will log in to Windows as they would do normally. The MyAssays Desktop application can be used without requiring an additional log in prompt.

With **MyAssays Desktop Log In**, when the MyAssays Desktop application is launched, the user must log in to the application to authenticate.

In both cases, user accounts are managed (e.g., adding/creating/disabling/locking user accounts) by the administrator using the standard Windows user management tools (see General User Administration). The user authorizations that are specific to MyAssays Desktop are managed using the MyAssays Desktop Pro **Security Management** tool.

Authentication Context

MyAssays Desktop Pro's Enhanced Security supports these authentication contexts:

- **Local Machine**
- **Domain**

This means that user accounts can be managed either locally (on the local machine itself) or via a domain controller.

Which Authentication Method?

Deciding on the correct authentication method depends on how the workstation will be accessed:

- If each user will log in to Windows using their own Windows account, then use **Windows Log In**.
- If the workstation is configured to use a single common login that is shared by multiple users for general access, then use **MyAssays Desktop Log In**.

In many ways, the **Windows Log In** method is the simplest to work with and administer. End users have a single familiar login method to work with.

However, if you are working with third-party software (such as instrument control software), you may find that it imposes certain requirements on the Windows log in that must be followed. For example, it may be necessary for that software to be operated under a specific restricted Windows login to ensure it is correctly secured. In these cases, it may not be feasible to allow system users' their own Windows log in session; it is for this reason that the MyAssays Desktop Log In authentication option is available.

Windows Log In	MyAssays Desktop Log In
The workstation can only be accessed by a user providing their own personal credentials via the Windows Log In. The credentials provided when the user logs in to Windows are used by MyAssays Desktop. This means that the user does not need to log in to both Windows and MyAssays Desktop separately.	The workstation is routinely used through a single Windows log shared by multiple users. To secure MyAssays Desktop data and features by user, each user has their own user account that MyAssays Desktop uses to authenticate. This means that when MyAssays Desktop is used, the user must log in to the application directly.

Authorization

Any authenticated user will be able to launch the software; however, the operations that they can perform depend on the authorization settings that have been assigned to them.

Each user is assigned to a single authorization group. The system administer can assign authorizations to users with the MyAssays Desktop Pro **Security Management** tool.

For example, a user assigned to the **Run and Review Only** authorization group would only be able to run protocols and review results (he would not be able edit protocol settings), whereas a user assigned to the **Unrestricted** authorization would be able to perform any permitted operation.

If a user successfully logs in to MyAssays Desktop Pro Enhanced Security and that user has not yet been added to the system, the user will be treated as a **Guest** user with no permitted authorizations. (The system advises guest users to contact the administrator to gain access to the software).

With MyAssays Desktop Pro Enhanced Security, any attempt to perform a protected operation is stored in the audit trail (including whether the operation was permitted or denied in accordance with the user's authorization).

Secure Data Hand-Off

When working with measurement data originating from instrumentation (such as microplate readers or gamma counters), it is important to ensure the integrity of the imported data.

In many systems, the hand-off of data between one system and another provides an opportunity for users to tamper with and modify the data either unscrupulously or in error. Such a vulnerable workflow is unlikely to pass regulatory validation or stand up to scrutiny.

For example, after the readings are completed the measurement data is written to a file. This data must then be transferred or imported into the analysis software (MyAssays Desktop Pro). It is at this point that the data could be intentionally (or unintentionally) falsified. MyAssays Desktop Pro Enhanced Security can detect and block such insecure actions.

In addition, in certain scenarios, it is important to ensure that a data file originates from a particular source. For example, in a laboratory with multiple plate readers, it may be important to ensure that measurement data is collected from a specific reader and not transferred to the system from another unexpected or untracked source.

It is for these reasons that MyAssays Desktop Pro Enhanced Security supports restricting the import of measurement in a number of different ways including:

- Requiring a valid data certificate
- Blocking files that were modified after creation
- Restricting data import from only a specific location

These methods can be used alone or in combination to ensure that the data imported into the system meets regulatory requirements for authenticity and traceability.

Data Certification

The most reliable method to secure the hand-off of data is to configure the system so that a valid data certificate is required for all measurement data to import.

A MyAssays data certificate file is generated by the instrument control software. The certificate file includes the information necessary for MyAssays Desktop to validate the authenticity and source of the measurement data file.

If the certificate file is missing, or if the data file has been modified since the measurement data file was certified, then MyAssays Desktop Pro Enhanced Security will block the import (and log the attempt).

The certificate file also stores the origin of the file; this makes it possible for MyAssays Desktop to reject data files that are transferred from another system.

Data Certification with PerkinElmer EnVision

PerkinElmer's EnVision microplate reader supports MyAssays data certification.

Ensure that you are using v14.xxx or later of the manager software.

To export certified measurement from the EnVision:

1. Ensure that you are using v14.xxx or later of the manager software.

2. Configure or perform the export function.

3. Select Export format option as CSV for MyAssays Desktop option:
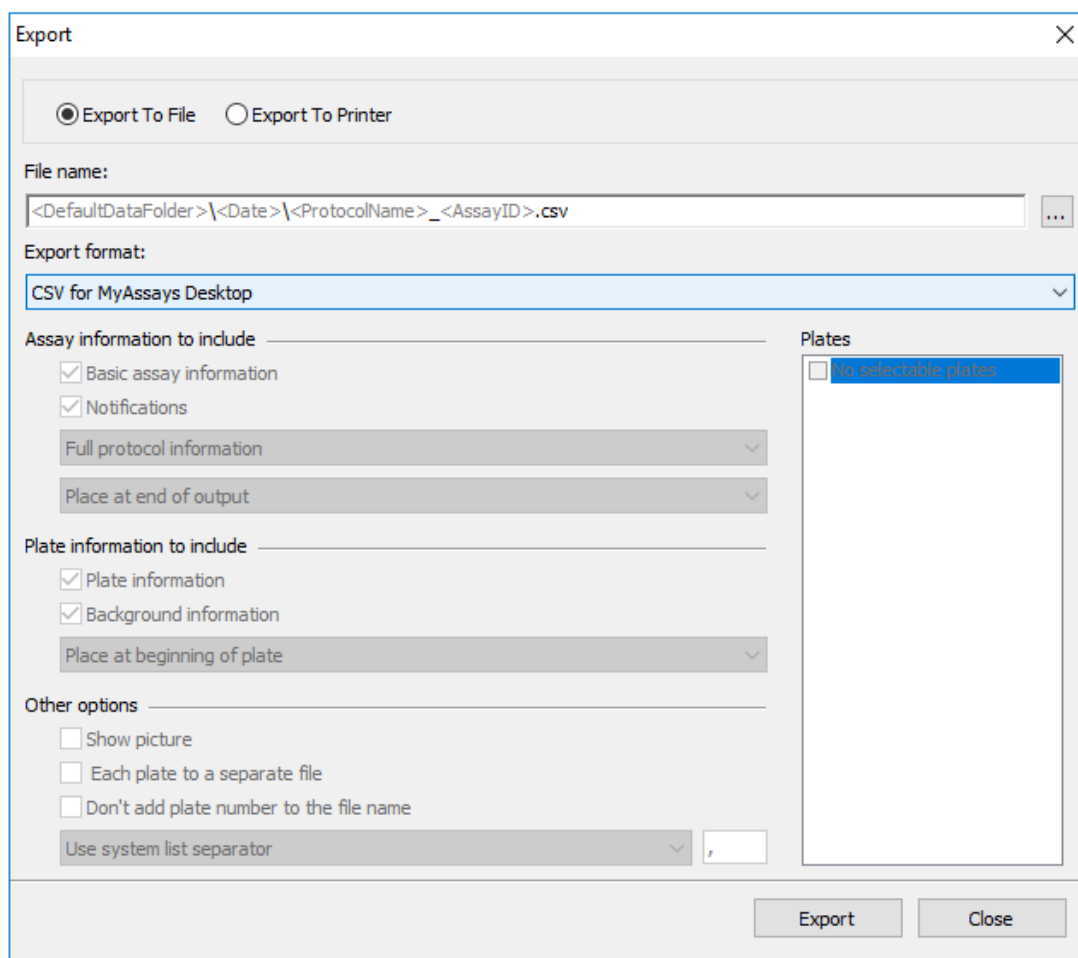
The generated data certificate files are stored alongside the exported file. Data certificate files are hidden by default (so they would not appear in Windows Explorer unless it is configured to show hidden files).

Data Certification with Other Instrumentation

If you are working with other instrumentation, please contact support@myassays.com for advice how to secure the hand-off of data.

MyAssays Ltd. offers solutions for retrofitting secure data certification to legacy systems (without requiring any modification to the third-party software).

Data Protection

One of the advantages of MyAssays Desktop Pro is that it does not require a database to administer. All assay data is stored in standard files, making it easy to manage and transfer data files between users and systems. The Enhanced Security features enable you to configure and restrict access to MyAssays Desktop data files and folders, and to restrict the operations that can be performed on those files.

MyAssays Desktop Pro's Enhanced Security provides various features and options for securely protecting files. The configuration you choose will depend on how your organization prefers to arrange data. For example, you can:

- Partition files by users (**Windows Log In** authentication only)
- Share files between users

What Are the Assay Data Files?

The term **assay data files** refers to all the types of files used by MyAssays Desktop to configure, manage, and store details about the operations that it performs. This includes files for:

- Protocol
- Results
- Layout
- Export method
- Library curves
- Quality control settings
- User preferences
- Export reports (only those exported to the default **Reports** folder)

What about Measurement Data Files?

Measurement data files are generated by instrument manager software and therefore cannot be managed or controlled directly by MyAssays Desktop Pro Enhanced Security. It is the function and responsibility of the instrument manager software to ensure the integrity and security of the measurement data files it generates. To ensure that you have a fully secured system, you will need to ensure that the instrument manager software is sufficiently secured to meet your regulatory requirements.

Where MyAssays Desktop Pro Enhanced Security can help is in the secure hand-off of measurement data from the instrument manager through to data analysis in MyAssays Desktop Pro. In this way, a secure workflow from instrument to exported results can be achieved.

If you can configure the location of where your instrument manager software outputs measurement data files, then it is suggested that you use the **Measurements** subfolder as the default location of the **Assay Data Folder**. In this case, the administrator will need to configure appropriate permissions for the instrument manager software to be able to write to this folder.

Where Are the Assay Data Files Stored?

With MyAssays Desktop, data file management is simplified using the **Assay Data Folder Root**. All assay data is stored under this parent folder (data is arranged under subfolders). Using a single parent folder for all assay data simplifies management of users' assets.

A typical scenario (and the default scenario when Enhanced Security is not used) is for the **Assay Data Folder Root** to be the **MyAssays** folder under each user's **My Documents** folder. Each user has their own protected folder for storing their own data files. In this scenario, each user's data is kept separate from one another. In this case, the operating system provides secure functions for restricting access and permissions to resources.

A common requirement is to share all assay data between users of the system. In this case, the **Assay Data Folder Root** is the same path for all users. This could be a local file path or a network location. This is easy to configure by changing the **Assay Data Folder Root**. However, securing data shared in this way can be a challenge, and it is for this reason that MyAssays Desktop Pro Enhanced Security provides functions to simplify and manage this process.

Traditionally, managing shared access to common assets is difficult in Windows with various authorization settings being spread across different management tools. MyAssays Desktop Pro Enhanced Security simplifies this file management using easy-to-use authorization groups.

For example, if a user should not be permitted to delete or edit files, he can be assigned to the restricted **Run and Review** authorization group. This group does not include the permissions necessary to delete data files.

How Are Files Protected?

Protected data is stored in secure folders protected at the operating-system level. This means that only programs and users with the correct permission to access or modify data can access and make changes to the protected data. Any attempts to manipulate data using tools outside MyAssays Desktop Pro will be denied by the operating system.

There are two types of data protected by MyAssays Desktop Pro's Enhanced Security mode:

Security Data

Only the security system itself has access to the Security Data folder (where all audit trail, login, and user details, and all authorization settings, are stored). In all cases, the security data is protected and cannot be accessed or modified by users without administrator privileges.

Assay Data

Where assay data is shared between more than one user, MyAssays Desktop can provide Secure File Protection. With this feature enabled, MyAssays Desktop Enhanced Security takes control over securing the assay data files between different users.

Only MyAssays Desktop Pro Enhanced Security (and an administrator account) has the ability to make changes to data stored in MyAssays Desktop Pro data folders; in this way, the assay data files are securely protected. Only operations approved by the system (in the form of an authenticated user with the required authorizations) can be performed. Also, secured operations on the data are logged in the audit trail.

Audit Trail and Login Log

The **Audit Trail** keeps track of user activity; user operations are logged by user with a date and time stamp. In addition, the **Login Log** keeps track of log in attempts.

The log data is stored in a protected folder, so it cannot be accessed or modified by standard users.

Log data can be exported to CSV or XML.

Administrator Tasks

This section covers the system administrator tasks necessary to set up and maintain the system:

- Install MyAssays Desktop Pro

- Enable and Configure Enhanced Security

- User Administration (Manage/Add/Authorize Users)

- Monitor Usage

Installation

Installation tasks must be performed by a Windows user with local machine administrator privileges.

Please refer to release notes for latest installation instructions.

Enable and Configure Enhanced Security

After installation and license activation, the Enhanced Security system will not initially be enabled. To enable Enhanced Security, launch the **Security Setup Tool** as follows:

1. Launch **MyAssays Desktop Explorer**.

2. Select **Enable Security** from the **Tools** tab.
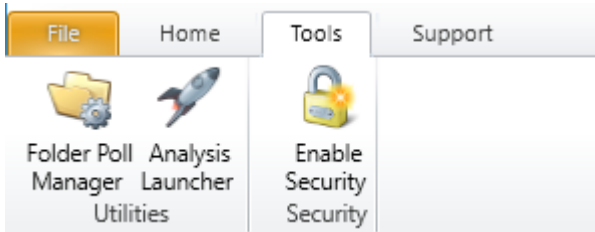
Security Setup Tool

The **Security Setup Tool** guides you through the process of configuring Enhanced Security system with MyAssays Desktop Pro. The tool can be used to set up the initial configuration of the system; you can also rerun the tool to make changes to an existing configuration.

The **Security Setup Tool** makes changes to the system, so it requires local administrator privileges and will display the standard Windows **User Access Control** prompt. Press **Yes** to confirm to start the tool:
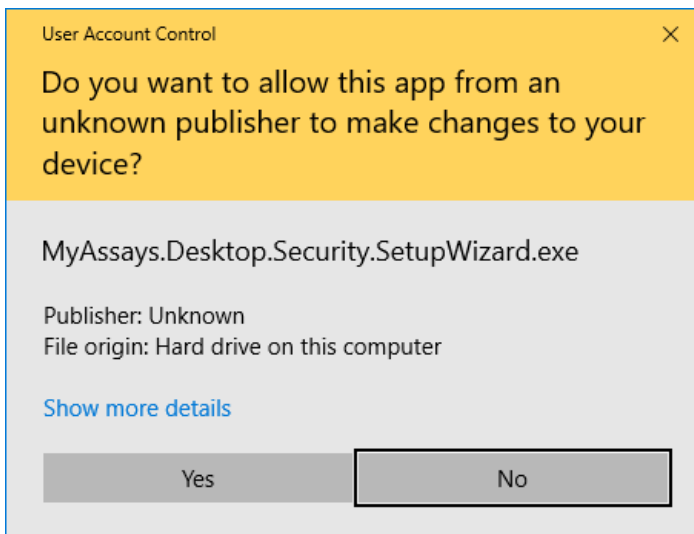
On the first step of the wizard, select **On** to enable the Enhanced Security features:
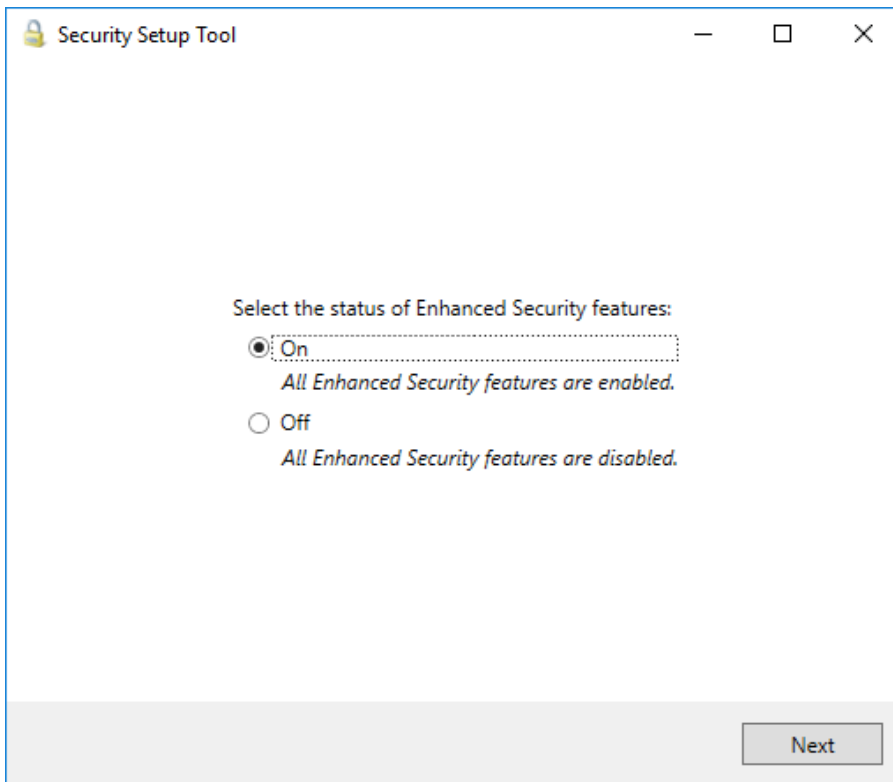
Press **Next** to continue through the wizard.

On the second step, select the authentication method:
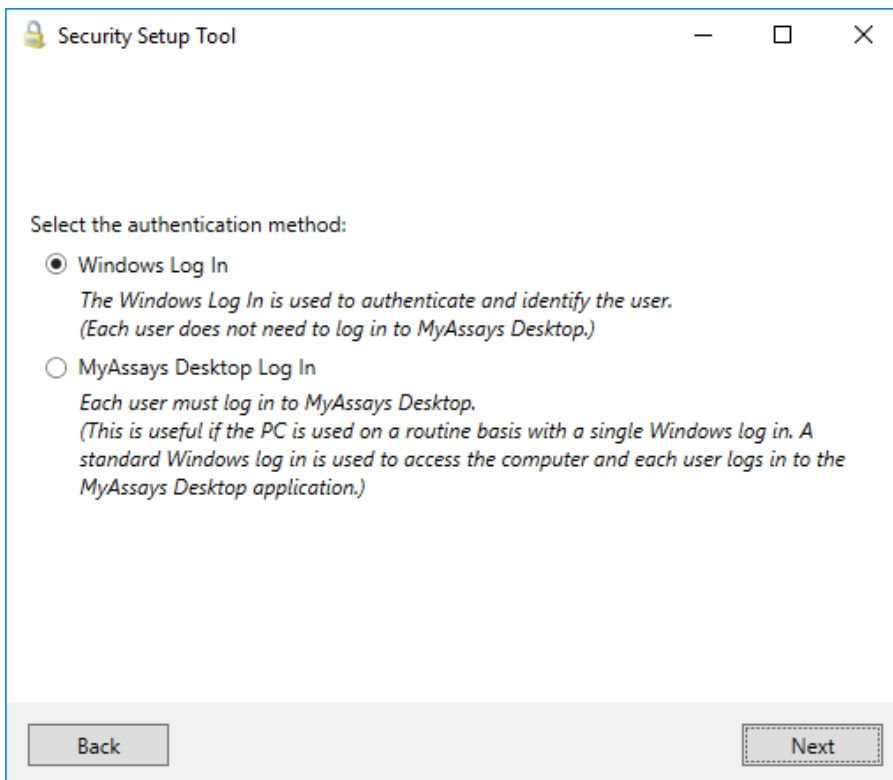
If you select the **MyAssays Desktop Log In** method, then it is also necessary to specify a **Windows Log In** account that is the standard shared Windows log in, i.e.:
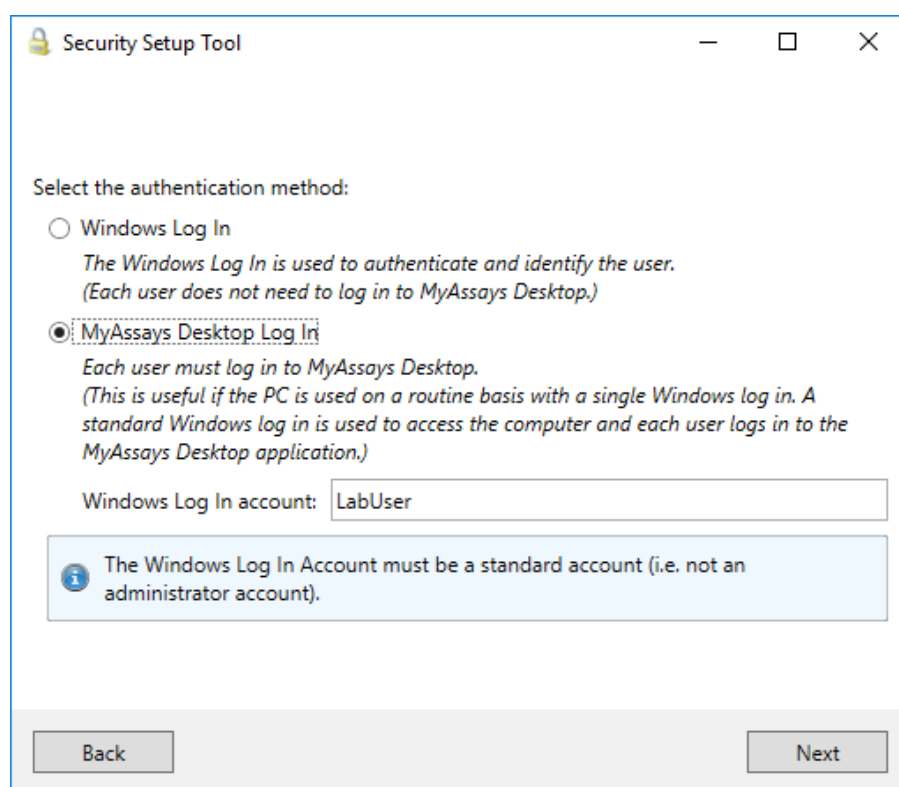
A default account name of **LabUser** is provided. Enter a different account name as required. In any case, please ensure that the account name is a valid local machine account. This **Windows Log In** account is the account from which the system will be used routinely, with the account's access being shared between all users of the system.

Press **Next** to continue through the wizard.

For **Windows Log In** authentication, the next step of the wizard is specifying the location of the **Assays Data Folder Root**:
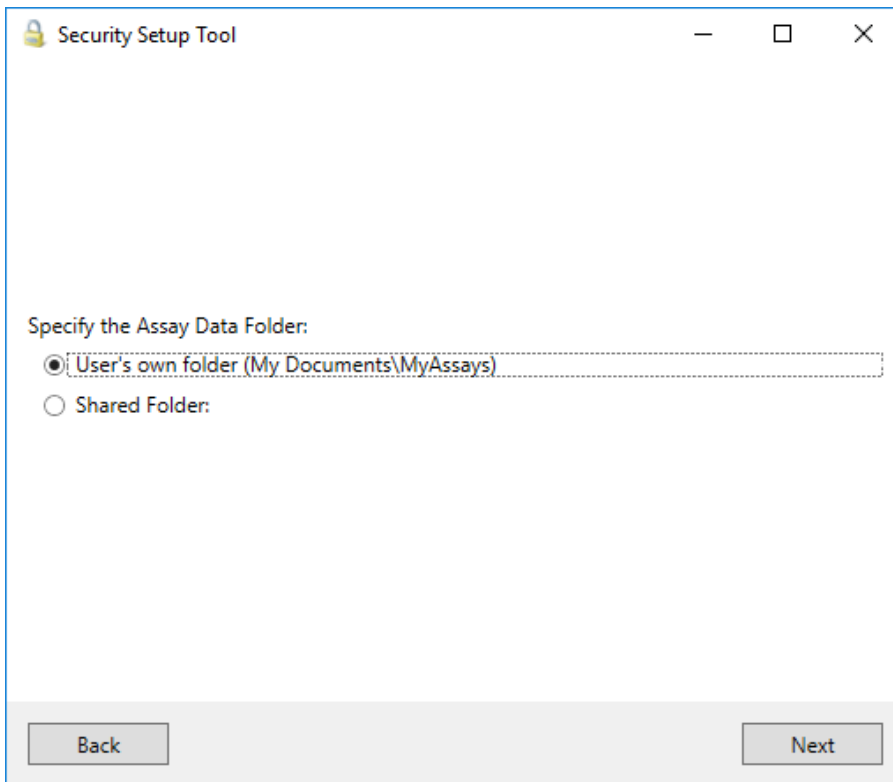
If the **User's Own Folder** option is selected, then each user's assay data is stored under the logged in Windows users account's default Documents location. This is useful is you want to partition each user's data by their own account. In this case, Windows manages the partitioning of data (e.g., one standard user cannot access another user's data).

The **Shared Folder** option configures each user's account to use a single shared assay data repository; all users of the system share the same folder. In this case, MyAssays Desktop Enhanced Security can secure the specified folder so that (when running under a standard Windows account) operations can only be performed on files in that folder by users authorized through the Enhanced Security system.

Secure File Protection
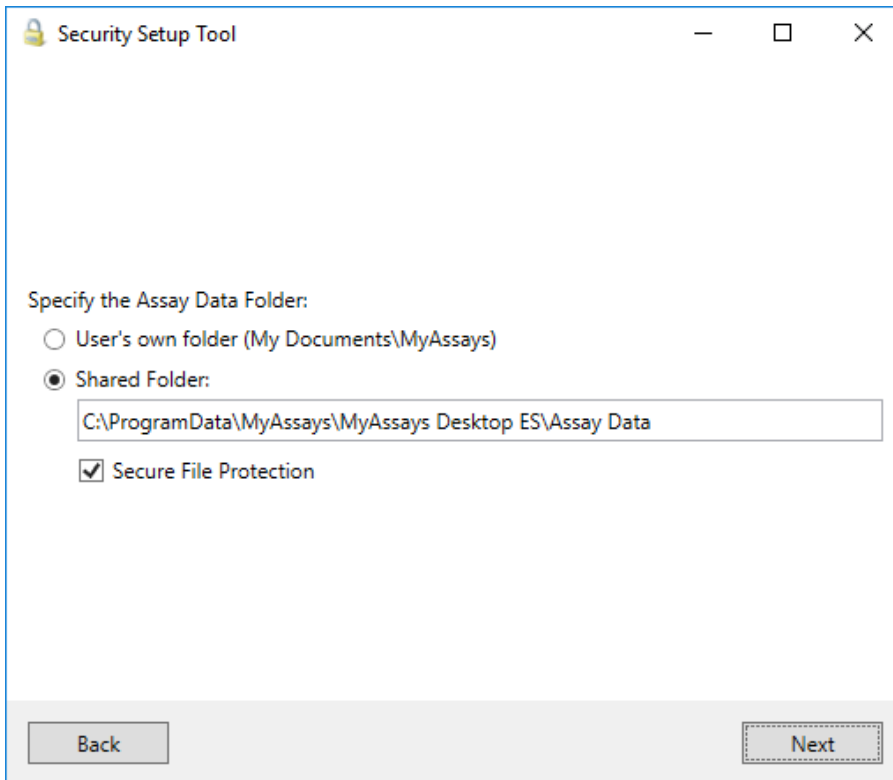
With the **Secure File Protection** option selected, MyAssays Desktop Enhanced Security secures the assay data files stored under the specified **Shared Folder**. Only MyAssays Desktop Pro Enhanced Security (and an administrator account) has the ability to make changes to data stored in MyAssays Desktop Pro data folders; in this way, the assay data files are securely protected. Only operations approved by the system (in the form of an authenticated user with the required authorizations) can be performed. Also, secured operations on the data are logged in the audit trail.

Secure File Protection requires that the system account has permission to take ownership of the specified resource. This is normally the case for the resources of a local machine (such as a local hard drive). If this is not the case (such as with a restricted network resource), then data can still be shared between user's, however the protection of such files should be managed and configured by the resource's administrator.

Press **Next** to continue through the wizard.

For **MyAssays Desktop Log In** authentication, the next step of the wizard is specifying the **User Group Name**:
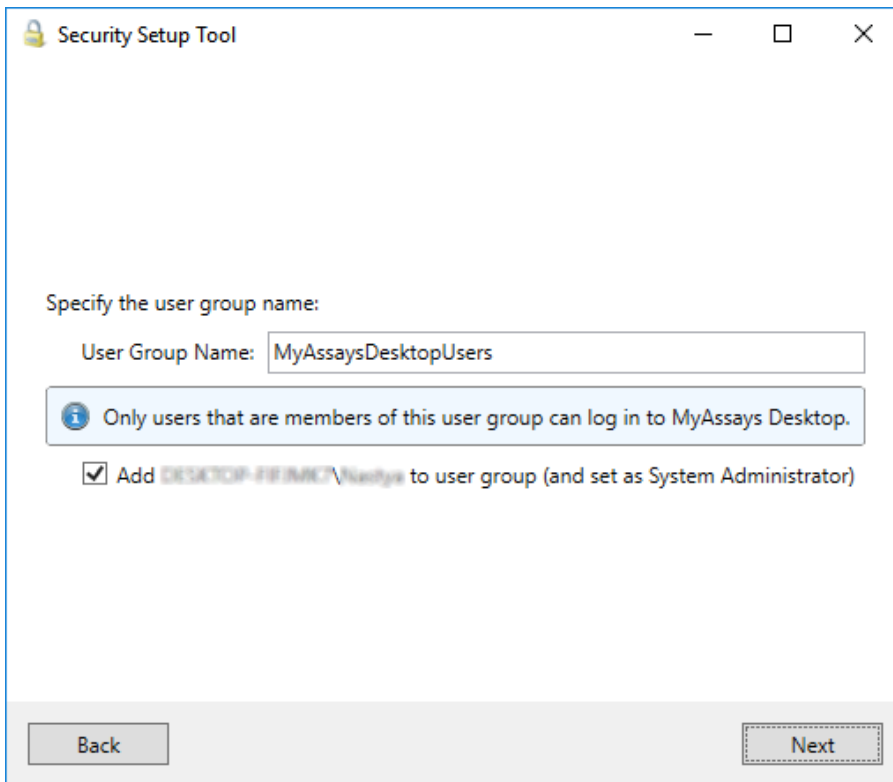
In most cases, you can accept the default settings here to use the specified **User Group Name** and add the current user (the administrator) to the user group and set as an Enhanced Security System Administrator.

With **MyAssays Desktop Log In** authentication, only users that belong to the specified User Group (e.g., **MyAssaysDesktopUsers** will be able to authenticate to access MyAssays Desktop Pro).

Press **Next** to continue through the wizard.

Password Configuration

This step provides various configuration options:
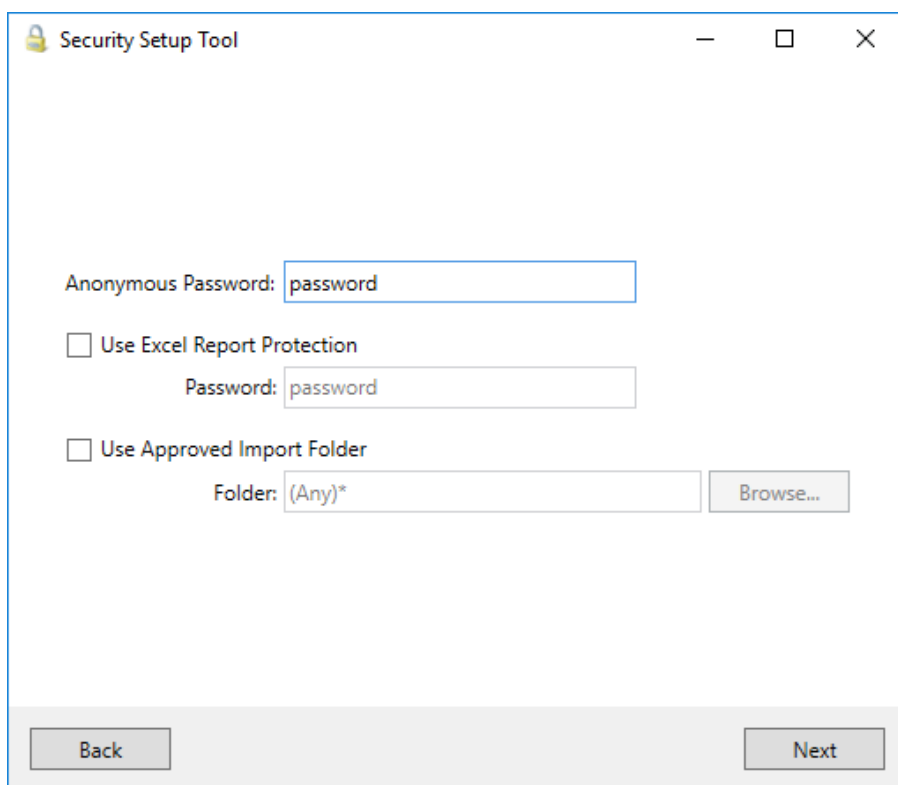
The **Anonymous Password** is used to specify a password for the Anonymous account when working with **MyAssays Desktop Log In** authentication. This account is useful if working with third-party software (such as instrument control software or LIMS) where operations should be performed independently of any specific user account with an automatic log in procedure.

Check **Use Excel Report Protection** to lock all Excel output generated by MyAssays Desktop Pro. This means that modifications to the generated Excel output cannot be made without the password (assuming the **Data Export | Protect XLSX** authorization is set for the user). Enter the password to apply.

The **Use Approved Import Folder** feature enables restricting data import to a specific folder; this means that the system will only allow the import of measurement data files from a specified approved folder. This feature can be used to add a layer of control over the import of data files. For example, the instrument control software might output measurement data to process to a specific folder only. For additional security, the system could be configured so that only the instrument reader software can write to this folder.

Check **Use Approved Import Folder** to restrict import in this way and to specify the folder where data files can be imported from. This restriction applies to all users that belong to an authorization group where the **Data Import | Import From Approved Folder** authorization is set and where the **Data Import | Import from Any Folder** is not set.

The final wizard step displays a summary of the configuration options that will be performed:
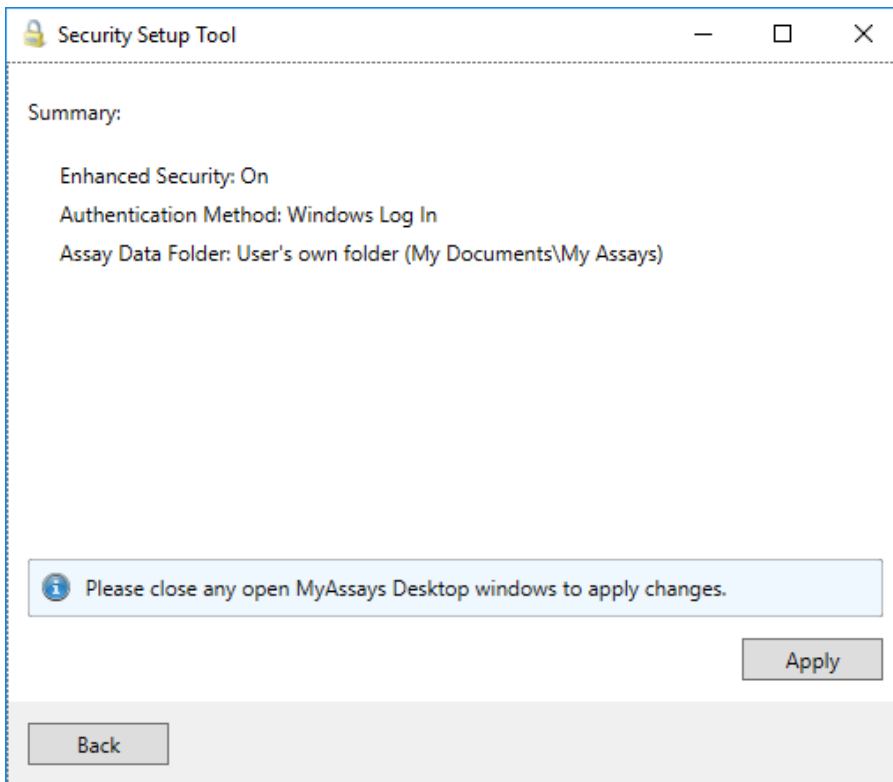
Press **Apply** to apply the specified changes.

On successful completion, press **Finish** to close the setup tool. If there are any MyAssays Desktop windows open, then close them. Changes will be applied the next time MyAssays Desktop is launched.

> When working with a shared assay data folder, it is necessary that MyAssays Desktop Pro is launched at least once with a successful authentication to install the pre-configured protocol and example files under the protected folder.

User Administration

With Enhanced Security enabled and configured using the Security Setup Tool, initially the only user account that will be added to the Enhanced Security system is the user account that was used to perform the setup (i.e., the administrator account). This account is automatically added to the **Unrestricted** authorization group and assigned to the Enhanced Security account type **System Administrator**.

To grant other users permission to use MyAssays Desktop Pro, each user must be added to the Enhanced Security system and assigned to an appropriate authorization group and account type.

Guest Users

When working with **Windows Log In** authentication, users that are not added to the Enhanced Security system can launch MyAssays Desktop Pro but will be treated as a guest user. Guest users are advised to contact the system administrator for access to the software. Guest users cannot perform any operation.

User Accounts

The Enhanced Security system harnesses the Windows user management system for administering user accounts. This means that you do not need to create a whole new set of user accounts; you can use existing accounts that may already be defined on your system (both local or domain accounts).

General User Administration

General user management (such as adding/creating/disabling/locking accounts) can be administered using the standard Windows user management tools. In Windows 10, if using local machine context:

Right-click on **Start** | **Computer Management** and expand **System Tools** | **Local Users** and **Groups** | **Users**.

Password policy rules (password history, minimum length, etc.) can be set in Windows 10 from **Start** | **Run** | **secpol.msc** | **Security Settings** | **Account Policies** | P**assword Policy** and **Account Lockout Policy**. These should be set in accordance with your 21 CFR Part 11 compliance requirements. For more information on configuring Windows password policy, please seehttps://docs.microsoft.com/en-us/windows/device-security/security-policy-settings/password-policy.

Domain

If your network is connected to a domain, then your password policy rules and user management will be administered by the domain controller. Contact your domain administrator for more information.

Domain Users

MyAssays Desktop Enhanced Security can work with your existing domain accounts.

Windows Log In Authentication

For **Windows Log In Authentication**, it is not necessary for a domain administrator to configure the system. The system configuration requires only local administrator privileges. This local administrator can set up the system and assign users (including domain users) access to the software at various authorization levels.

MyAssays Desktop Log In Authentication

For **MyAssays Desktop Log In Authentication**, if Enhanced Security will use domain-based authentication (i.e., users defined in a domain context rather than on the local machine), then the configuration must be performed by a user with domain administrator rights. This is necessary to configure the authorization setting of standard domain users.

In this case, please ensure that the configuration process is performed by a Windows log in with domain administrator rights.

Administering MyAssays Desktop Pro Users

To grant a user account permission to use MyAssays Desktop Pro, the account must be added to the Enhanced Security system. The steps to add a user depend on the authentication method in use, as described in the following sections:

Add Users – Windows Log In Authentication

With **Windows Log In** authentication, the administrator must add each user that requires access using the **Security Management** tool.

With **Windows Log In** authentication, users that attempt to access MyAssays Desktop Pro but have not be added to the Enhanced Security system will be automatically assigned to the **Guest** account type.

The administrator can add users to the Enhanced Security system as follows:

1. Launch **Security Management**
2. Press **Add...**
3. Under the **User Name**, enter the Windows user name (as the user would enter it when logging in).

   If the user is a domain user, then enter the user name including the name (e.g., for a domain "CORP" and username "LabUser" the **User Name** should be: **CORP\LabUser**).
4. Provide a **Full Name** for the user.

Please now proceed to the Configuring a User section.

Add Users – MyAssays Desktop Log In

With **MyAssays Desktop Log In** authentication, the administrator must first add each user that requires access to MyAssays Desktop to the defined MyAssays Desktop user group. This is the group that was specified in the **Security Setup Tool** and defaults to **MyAssaysDesktopUsers**, i.e.:
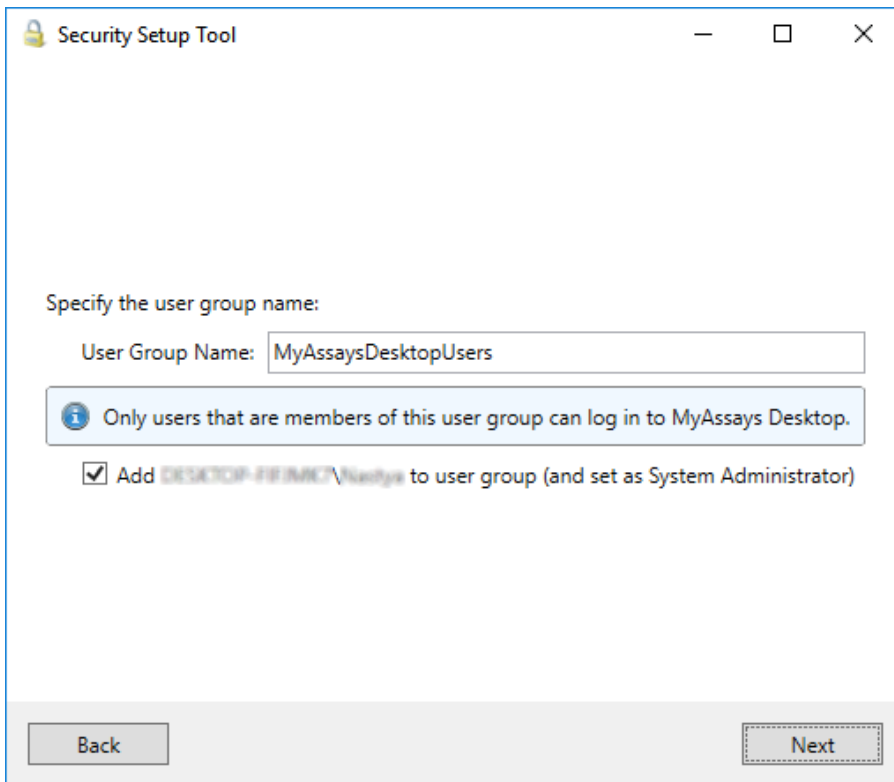
Users can be added to this local machine group using the standard Windows user management tools:

1. In Windows 10, right-click on **Start** | **Computer Management** and expand **System Tools** | **Local Users and Groups** | **Groups**.

2. Locate the user group **MyAssaysDesktopUsers** (or any custom name provided in the configuration) and double-click on it.

3. Press **Add...**

4. Enter the name (or names, each separated by a semicolon) of the user account to add. Press **Check Names** to check that it was entered correctly, and press **OK** to confirm.

Users added to this user group will now be able to authenticate themselves when launching MyAssays Desktop Pro; however, each user must also be assigned to an appropriate authorization group and account type as described in the following section.

Configuring a User

After adding users to the Enhanced Security system (using the method appropriate for the authentication method), each user should be configured as required. This involves setting an appropriate:

- Account type

- Authorization group

Each user's settings are specified through the **User Details** panel, which can be accessed as follows:

1. Launch the **Security Management** tool. This is available from the **Tools** section of MyAssays Desktop Explorer:
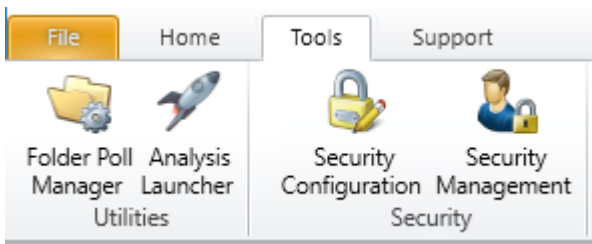
2. Double-click on a user, or select and click **Modify...** to modify the user's details:
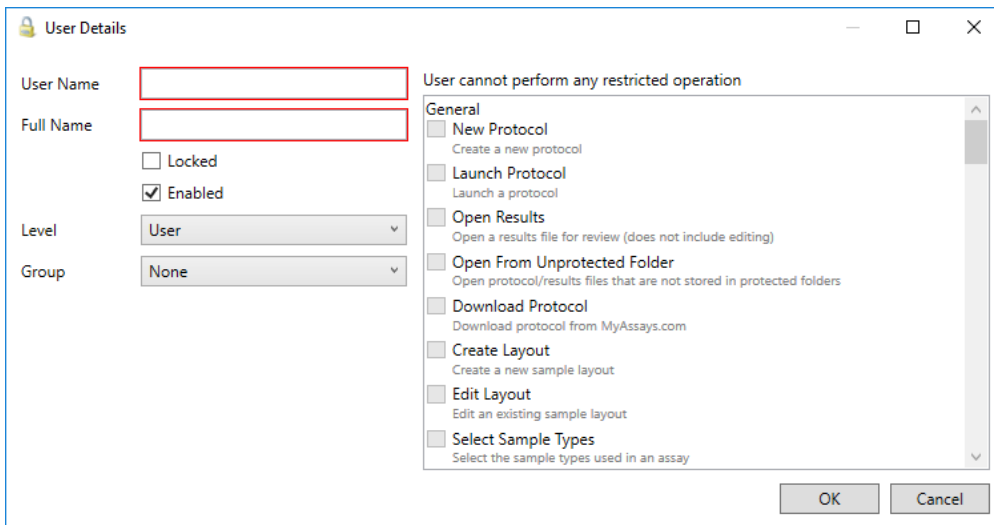
Account Type

The account type or **Level** can be one of the following settings:

- **User** – A standard user (cannot access the Security Management user management console).

- **User Administrator** – An administrator who can assign users to belong to preexisting authorization groups and assign user levels to either User or User Administrator.

- **System Administrator** – A highest level administrator. A system administrator has all functions of the user administrator and can also:

  - Assign user levels to System Administrator

  - Create and edit authorization groups (using **Manage Groups**)

  - Change security options (with **Security Options**)

Only users assigned to the Enhanced Security Account Type Level of **User Administrator** or **System Administrator** can use the **Security Management** tool (also, these users must be Windows administrators).

User Group

The user group (or **Group**) is an authorization group that defines which functions of the software the user can perform. MyAssays Desktop Enhanced Security provides various predefined authorization groups including:

- **None**
- **Unrestricted**
- **Review/Export Only**
- **Run and Review**

For example, users belonging to the **Run and Review** groups are restricted to running protocols and reviewing results. Conversely, an **Unrestricted** user can perform all operations in MyAssays Desktop Pro.

Authorization settings can be configured by the system administrator as described in the following section.

Authorizations

System administrators can manage authorization groups. Click on the **Manage Groups...** button in the Security Management tool to display the User Group Management form:
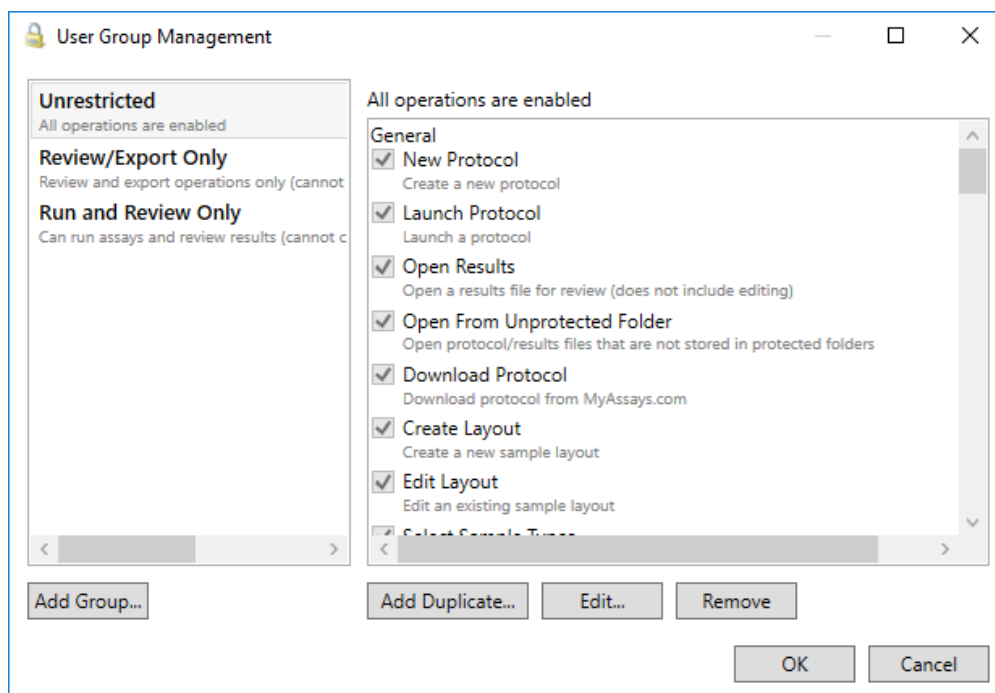
The list on the left shows the existing list of authorization groups. The controls allow a system administrator to edit operations and create new authorization groups.

The **Add Duplicate** button can be used to create a new authorization group, which is a duplicate of the currently selected group. This is useful for quickly creating similar groups.

Security Options

System administrators can also change settings under **Security Options** as follows:
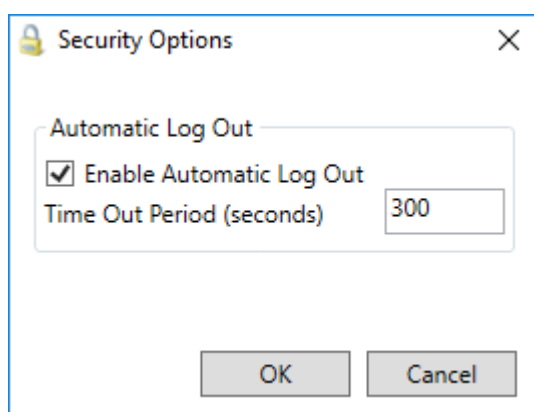
The Automatic Log Out feature is applicable only when working with **MyAssays Desktop Log In** authentication. This feature has two uses:

- Authentication reuse

- Log out inactive users

With **MyAssays Desktop Log In** authentication, multiple users share a single Window session. In this scenario, the operating system cannot provide any automatic log out of the MyAssays Desktop users. It is for this reason that MyAssays Desktop Enhanced Security provides its own automatic log out handling.

In addition, when working with **MyAssays Desktop Log In** authentication, it is likely that a single user will open multiple MyAssays Desktop windows. To avoid requiring the user to log in to each MyAssays Desktop instance, the authentication session is shared between windows within a specified time period. This means that if a user is authenticated in one MyAssays Desktop window, she will be able to open another MyAssays Desktop without being required to log in again, providing that the user has been continuously active on the system. If the user is inactive for the specified **Time Out Period**, then the user is logged out automatically and will need to log in again to use any MyAssays Desktop window.

The Automatic Log Out feature is intended to automatically log out and close any open MyAssays Desktop windows if the user leaves the workstation unattended for the specified period. If any MyAssays Desktop Pro window is open without activity for the specified number of seconds, then the user is automatically logged out.

> For security reasons, if Automatic Log Out is disabled, then the user will need to log in to each and every new MyAssays Desktop Pro window opened (this is because the authentication cannot be shared between MyAssays Desktop Pro windows indefinitely).

Monitor Usage

User administrators and system administrators can use the Audit Trail and Log In Log buttons to view the corresponding logs of information. For example:
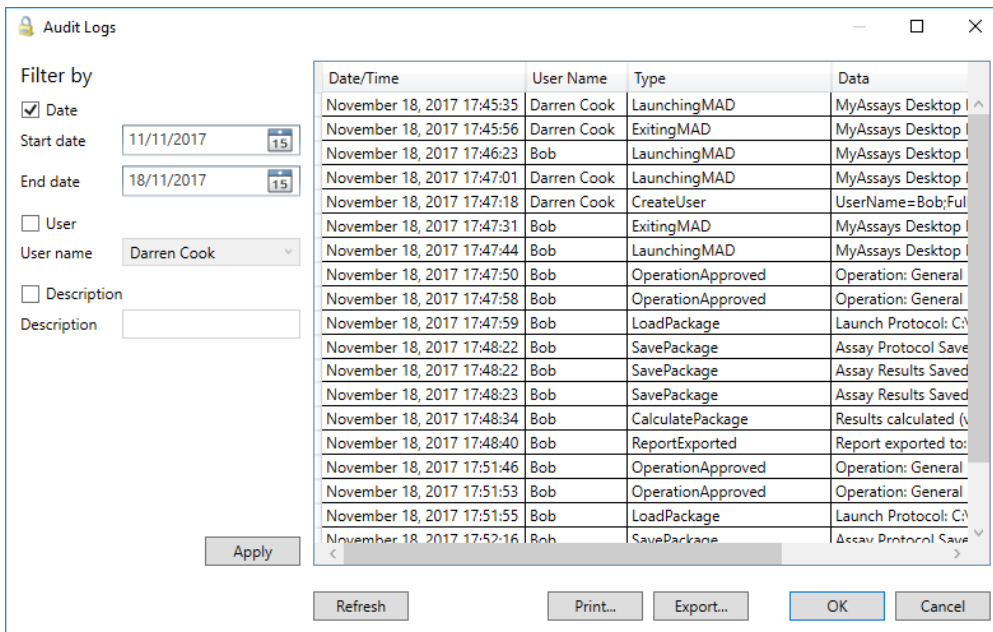
Filter

Tools are provided to filter the log by date, user, and description. Check the appropriate controls, enter the parameters, and click Apply to filter the log.

Export

The Export button displays the Export log tool, which can be used to export the selected log information to an XML or a CSV file. Click Export to export the contents.
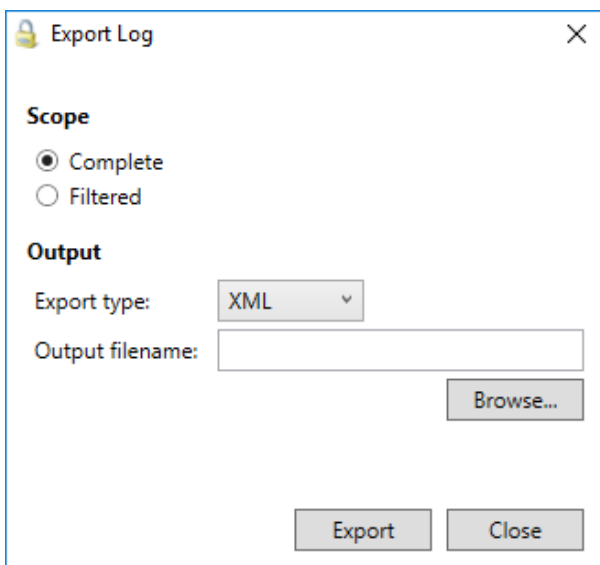
Working with MyAssays Desktop Pro and Enhanced Security

This section explains the functional differences when working with Enhanced Security enabled with MyAssays Desktop Pro; this section provides information for all users of the secured system.

MyAssays Desktop Explorer

The MyAssays Desktop Explorer Welcome screen displays a padlock icon to clearly identify that Enhanced Security is enabled.

Also, the Welcome screen displays the logged in user's Full Name and the details of the last log in attempt (this will include any failed log-in attempts).

The **File | User Accounts** section displays further details about the currently logged-in user including authorization group and account type.

Administrators can access the **Security** group under the **Tools** tab to find the various Enhanced Security configuration tools.

Administrator Security Tools

For administrator users, security tools are available from the **Security** group of the **Tools** tab. This includes the **Security Configuration** and **Security Management** options, i.e.:
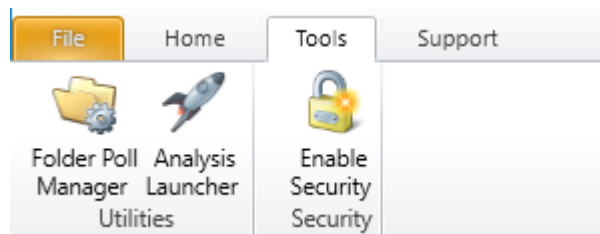
> 📝 These operations are only available for administrator users.

The **Security Configuration** tool can be used to modify the security setup; this launches the Security Setup Tool (only available to system administrators.)

The **Security Management** tool provides control over:

- User configuration
- Defining and setting authorization groups
- Viewing and exporting the Audit Trail and Log In Log
- Automatic Log Out settings

The **Security Management** tool is available for system administrators and user administrators.

File Operations

With Enhanced Security activated and the [Secure File Protection](#) feature enabled, destructive file operations on MyAssays Desktop Pro data stored inside the protected folders are prohibited. This means that the following Windows functions cannot be performed on protected MyAssays Desktop Pro assay data files:

- Delete
- Rename
- Create
- Move

However, authenticated users can be authorized to perform file deletion and file duplication through Enhanced Security by granting those users permission to:

- **Delete Data File**
- **Copy + Paste Data File**

These operations are managed by the Enhanced Security system so they cannot be performed using the standard Windows commands (i.e., Windows delete and copy + paste will not work). Instead, the secure version of these operations can be accessed through the context menu (by right-clicking on a selected single file). These secure file operations are indicated with the security padlock icon.

For example, to duplicate an existing MyAssays Desktop Pro protocol file, right-click on the file to duplicate and select the **Secure Copy + Paste** option, e.g.:
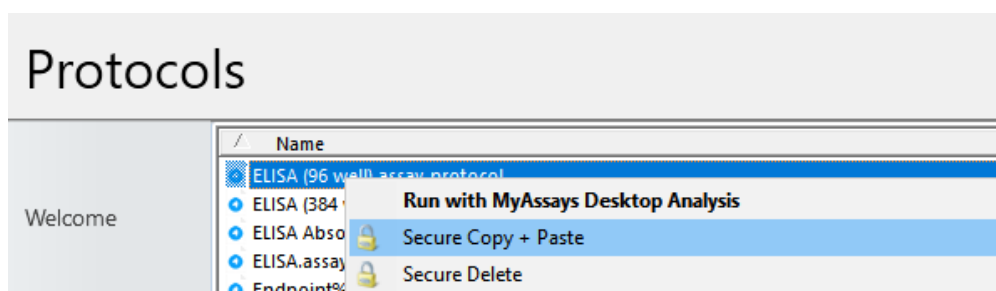
If the authenticated user is authorized to perform the secure file operation, then MyAssays Desktop Pro will prompt for a filename for the new file. Any performed secure file operation is logged in the audit trail alongside the authenticated user.

These secure file operations are available through the right-click context menu in MyAssays Desktop Explorer and in Windows Explorer. Note, the secure file operations can only be

performed on one file at a time (the secure file operations are not available when multiple files are selected).

Log In Options and Password Management

Log In Options

The logging in options are tailored to the selected authentication method. The specifics of how each of them work are different and are described below.

MyAssays Desktop Log In Authentication

With **MyAssays Desktop Log In** authentication, when launching any MyAssays Desktop window, the login prompt will appear:
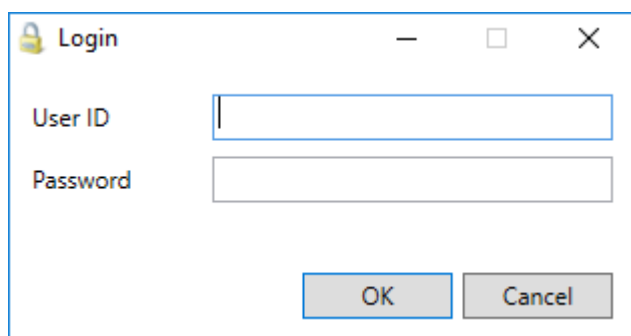
Enter your provided **User ID** (account name) and **Password** to log in.

Logging Out

With **MyAssays Desktop Log In** authentication, users should log out when they have finished working with MyAssays Desktop Pro (otherwise other users can perform unauthorized changes on their behalf). Users can log out from the MyAssays Desktop Explorer under **File | Log Out + Exit**.

Automatic Log Out

If the administrator has enabled the **Automatic Log Out** feature, then the user session is kept active while there is any user activity. If the **Automatic Log Out** period elapses without any user activity, then the user will be logged out of the system with any open windows closed.

In addition, the **Automatic Log Out** feature avoids the need to repeatedly log in to each MyAssays Desktop Pro instance. For example, if another MyAssays Desktop Pro window is opened, the user will not be asked to log in again (providing that the automatic log-out period has not elapsed while there is no user activity).

If the administrator has disabled **Automatic Log Out**, then the user will be prompted to log in to each new MyAssays Desktop Pro window that is opened.

Windows Log In Authentication

With **Windows Log In** authentication, the credentials used to log in to Windows are used to identify the current user; no additional log in step is required when launching any MyAssays Desktop window.

For logging out, the **File** menu includes the **Log Out + Exit** option. This command will exit MyAssays Desktop Explorer and log out of the current Windows session (closing any other open applications).

Password Management

For both authentication methods, password management is handled using the Windows security policy. If the security policy of the system dictates that the user must change his or her password, then after successful login, the user is advised to change the password. The new password must adhere to the system password policy options (such as minimum length and password history).

Quality Control

This section describes the Quality Control (QC) feature of MyAssays Desktop.

> 📝 This feature is only available to users with the QC feature enabled on their license.

Overview

The Quality Control features enable the monitoring of measurements and analysis results across assays runs. This is known as "inter-assay analysis" and can be used to track whether assay performance remains in control by comparing current results with historical observations.

For example, a QC test can check whether the value of the **Control1** in all results files falls in the range of +/- 2*SD and indicate cases where results fall outside this range.

> ℹ️ For intra-assay monitoring you can use the Validation feature.

QC Observations

The general approach is to define specific observations of interest; these are collected and compared across assay runs.

Each observation results in a single numeric value for each assay run. An observation can be a measurement or calculated result (including TOVs). For example, the measurement or calculated results for the sample **Control1** is an observation that is commonly monitored.

The QC process will gather the values of each selected observation across runs of interest. The results can be presented in a table and/or plotted on a QC chart to monitor inter-assay change. If these observations deviate from the specified range, then the assay is "out-of-control" and is highlighted accordingly.

QC Configuration

QC is configured using the intuitive QC Wizard that can be launched from both MADE and MAA.

QC can be configured:

- On demand – Create a QC report as an when you need it
- As part an assay protocol – With the QC results included in each results' report.

Additionally, you can save the defined configuration to a QC settings file and reuse it for future QC operations.

QC Results

In the simplest case, the result for each observation can be displayed in a table reporting observation result for each included assay. More sophisticated outputs can be generated plotting the observation data over time as CUSUM or Levey-Jennings charts.

The following subsections explain how to add, define, and review inter-assay QC analyses.

QC Wizard

Introduction

Inter-assay quality control is configured through the QC Wizard.

> ℹ The QC features can be used in various scenarios to track inter-assay data for chart-ing, analysis, and validation. The wizard provides a range of features and scenarios for convenient and simple inter-assay analysis tailored to your lab's needs and work-flow.

The QC Wizard will walk you through the process of:

1. Selecting the files consider

2. Specifying the observations to monitor

3. Selecting how the results should be presented

The QC Wizard can be launched in two different ways depending on the type of analysis you require:

QC On-Demand

QC On-Demand allows you to generate QC reports as and when you need to, selecting the files to consider. This is useful for the production of an occasional QC report.

To perform QC On-Demand launch the QC Wizard from MADE.

How to: Launch the QC Wizard from MADE

1. Launch MADE.

2. Select the **Results** tab.

3. On the **Ribbon**, hover to the **Quality Control** group.

4. Press the **Create** button to launch QC Wizard or the **Edit** to launch it with an existing configuration file.

> ℹ Select two or more results files and then press **Create** to automatically populate the wizard with these files.

QC as Part of an Assay

QC can be built into an assay so that each time the assays is used the results are compared with historical results of the same protocol.

To add QC to an assay launch the QC Wizard from MAA.

How to: Launch the QC Wizard from MAA

1. Launch an assay protocol or open a results file.

2. On the **Ribbon**, click on the **Add** drop-down button of the **Quality Control** group.

3. Press the **New Configuration** button to launch the QC Wizard or the **Existing Configuration** to specify the configuration file to launch the wizard with.

Selecting Files for Analysis

The Selecting Files for Analysis QC Wizard step specifies the results files which will be included in the inter-assay QC analysis.

The files could be added in the following ways:

Folder

Select this setting if the results you want to compare are stored in the same location and follow the same name convention.

- **Path** – The full path to the results folder location. Click on the **Browse...** button to select a folder in which your results files are stored.

- **Pattern** – Defines the pattern to extract results file with.

By default, the MyAssays Desktop organizes results files in accordance with the protocol name. For example, if a protocol is named "My Endpoint 12x8 Protocol," each run of the protocol will generate a results file that is named after "My Endpoint 12x8 Protocol" convention with a run number in brackets. For example, if the protocol was ran for three times, the newest results file is named "My Endpoint 12x8 Protocol (3)".

This organization of data simplifies the extraction of observation data from previous runs of a particular protocol. With this in place, the results are specified as:

Protocol Name*

> ℹ️ If the QC Wizard is launched in MAA, then the files to monitor are set up to match the configuration of the results naming setting.

For example, for the **My Endpoint 12x8 Protocol** results files, it would be **My Endpoint 12x8 Protocol***.

With this setting, all results for this protocol will be automatically added to QC analysis. In case you want to analyse only some of them, use the Specific Files setting.

Specific Files

This setting is useful if you want to track changes between specific results files or the results stored in different folders. For example, you might need to compare the results from first, fifth, and seventh run of an assay.

To add the files to the list, click on the **Add** icon button and select the required results. For adding multiple files, hold the **Shift** or **Ctrl** key, if the files are or are not sequential.

To remove a file from the list, select it and click on the **Remove** icon button.

Click **Next** to continue with the Wizard, and configure the observations to be tracked between the specified files.

Configuring Observations

This QC Wizard step allows you to add and configure the observations necessary to achieve your requirements of inter-assay quality control. Use the **Add Observation** or **Remove Observation** buttons to add observations as required.

Observations

Name

For each observation, a description can be specified under the **Name** column. Use this to provide a clear textual description for your QC monitoring. This will be used in the header for the **Observation Expression** column in the QC Results table to convey what has been monitored.

Data

For each Matrix-Transform Data Analysis Assays observation, specify which matrix the observation applies to. The numbering is sequential and starts from the index of the first matrix at 1 onward. The list of the available matrices (both raw and calculated) with their indexes is displayed in the Matrices tab. Any of the raw or calculated matrices that contain endpoint data can be referenced in an observation.

Matrix Name

If all selected assays have the same number of matrices and the same matrix names (e.g., Raw, Concentration, and Average), then a list-box with the matrix names will be displayed.

Expression

An expression is used to define the observation data. The type of expression depends on the assay type:

- **Observation** (for Matrix-Transform Data Analysis Assays) – Enter an expression to query the data of the specified matrix. This could simply be the name of a sample group, e.g. **Control1** or a more sophisticated calculation expression. Non-matrix data can be queried by using transform output variables.

- **XPath** (for other assays types) – A **XPath** expression that obtains single numerical value; that expression is then queried for each of the specified results.

> The type of expression to use will be reported in the column name.

Output

The inter-assay observation data can be plotted simply as a table of the values or as a CUSUM or Levey-Jennings chart alongside. For all output types, a text summary is included under each Observation Table.

SD±Limit

For each observation, control limits are expressed in terms of standard deviation range and are calculated automatically for the observation data.

You can define required range for the Table and CUSUM outputs and select the control rule for the Levey-Jennings.

The observation results for an assay run fall outside this range, they are highlighted in the resulting table.

Create QC Report Instantly

You can export a report to an Excel spreadsheet straight-away with the **Create Now...** option. The report will be saved to your MyAssays **Reports** folder.

Advanced XML Settings

The XML editor allows to use both file selection options simultaneously, set numerical formatting for the QC report, and specify how the observation expression is treated.

Set Custom Numerical Formatting

By default, the results in the QC report are displayed to 6 significant figures. To customize formatting settings, add the **NumericalFormatting** element as a child of the **Observation** and set its attributes.

> The **Type** settings are entered into XML without spaces.

For example:

```
<Observation Name="Observation1" Expression="Control1" Matrix="1" SDLimits="2" Out-
put="Table" />
<NumericalFormatting Type="DecimalPlaces"Precision="3" />
</Observation>
```

Here the numerical formatting is set to 3 decimal places.

Advanced File Selection Options

Typically and in most cases, the files are either stored in the same folder and can be conveniently added with **Folder** selection option, or could be selectively picked with **Specific Files** option. However, your files could be stored in several folders, or in the same folder with different naming patterns, or you could simply want to compare a folder with a specific file.

For those cases, you can specify the files to compare in the XML editor.

- **Single Results**

Add a File element as a child to Files and specify the full path to the file:

<File Path="Full file path" />

For example:

```
<File Path="C:\Users\User1\Documents\MyAssays\Results\ELISA\ELISA(1).assay-results"/>
```

- **Folder**

This is specified in the XML inside the **Files** element with following syntax:

<Files Folder="{0}" Search="{1}*"/>

where **{0}** is the full path to the folder and **{1}** is the pattern to select files with, followed by an asterisk *****.

For example, the folder contains five results for an ELISA assay (e.g., ELISA(1).assay-results, ELISA(2).assay-results, etc.) and several results for other assay types. To select only ELISA results, enter:

```
<Files Folder="C:\Users\User1\Documents\MyAssays\Results\" Search="ELISA*" />
```

Optional Inclusion of Subfolders

To include/exclude subfolders from file selection, add the Boolean **IncludeSubFolders** inside the **Files** element.

Setting Observation Type

The optional **IsExpressionXPath** can be added inside the Observation element and set to true/false and the expression is treated accordingly.

QC Results

On this step, if the wizard is launched from MAA (with a protocol or results), generated results can be added to a report (and generated on the next calculation). Otherwise, the results can be saved to the QC Configuration file.

Press **Finish** to exit the wizard.

Observation Evaluation Issues

If one or more observations is not evaluated, they are listed in a separate table to indicate their exclusion from the inter-assay analysis. In this case, the error message describing the issue is displayed instead of result in Result column.

Message	Issue
QC not available for this file. Please open file and recalculate to enable QC	The latest version of results was generated before the QC feature was introduced and needs to be recalculated to enable QC.
Failed to evaluate expression: {expression}	The specified observation expression is not valid for the file.
Failed to parse value:{value}	The specified XPath expression returns non-numeric value and is thus not valid.
Failed to evaluate XPath expression	The specified XPath expression is not valid for the file.

QC Workflows

Here are described the typical workflows when working with the Quality Control. These can be combined to provide accurate and consistent results for all of your research needs.

Generate Report Instantly

In the simplest case, specify the results you want to compare, launch (in either MADE or MAA) and configure observations through the wizard, and press the **Create Now...** button. The .XLSX report with your configured observation results is then generated and stored in your **Reports** folder.

Schedule QC Monitoring for all Future Runs of the Protocol

Typically, a protocol is created for an assay of a particular type (e.g., quantitative measurement of cortisol). This protocol is then run each time the assay is required. Running a protocol acquires the relevant measurement data and processes it in accordance with the calculations configured in the protocol; this generates a results file. If you set the QC to a protocol, it will be applied for all runs of this protocol in the future (so you don't need to repeat the configuration again next time).

Running this workflow is useful for designing systematic QC procedures. With this, the QC analysis is automatically performed as soon as a new results file is generated. For example, perhaps you are running a routine assay and want to monitor if the %CV of the measurements between runs does not go beyond a range of ±2*SD.

In this case, open the protocol and launch the QC Wizard. Check the **Add QC content to report** option, if you want to include the QC Results in the report automatically when new results are calculated.

Compare Results and Add QC Results to Report

Perhaps, you want to determine if the response of control results in today's assay is significantly different from corresponding control's responses in the reference assays. The defined observations you want to include in the report for current assay run only.

In this case, launch today's results file, select the results for comparison, configure required observations in the wizard, and check the **Add QC content to report** option.

> When **Add QC content to report** option is checked, the results of the current assay are always included as the last observation and indicated with **(Current)** after the observation number.

QC Configuration File

For example, your lab has multiple analyses running under the same acceptable limits or works under general quality standards. In this case, you can run and configure typical validating observations and reuse them for every assay.

To create QC configuration file, check the **Save settings to file** checkbox, when finishing working with QC Wizard. This file with all specified settings in place is stored under the QC subfolder in your MyAssays folder or any other specified location.

Note, this configuration may be tweaked depending on the analysis peculiarities (**Edit** or **Add Existing Configuration** commands) and optionally saved to another file, protocol, or results.

Reference

Transforms

Transforms can be described as a set of configurable operations that are performed to generate results. The results might be included in a report or used as inputs to other transforms. You can add multiple transforms in any order to perform a required data analysis with the leverage of transforms performed previously.

Most transforms use one input matrix and produce one output matrix (like Factor). However, this is not always the case. Here are the general rules:

- Each transform can have one or more input matrices.
- Each transform can produce one or more output matrices.
- A particular transform will always have the same number of input and output matrices.
- The input and output matrix types of a transform are fixed.

Matrices and Transforms

Input Matrices

A transform can accept one or more input matrices. Where an input matrix is used, the transform performs calculations using the data on those matrices. For example, the Factor transform multiplies all points on the input matrix by a specific value.

The input matrices available to a transform include the compatible raw data (i.e., of a particular data type) and compatible data that has been calculated prior to this transform.

For example, assuming all data is endpoint, the first transform can only access data on the **Raw** matrices, whereas a second transform can access data on the **Raw** matrices and/or the first **Calculated** matrix.

Output Matrices

Each transform will add one or more calculated matrices to the Matrices at each step. As each transform is evaluated, each container in the results will always contain the same number of matrices (raw and calculated) as all other containers in the results.

As you add transforms, you can configure output settings and display in the report for each of them.

Expressions and Transforms

In addition to the input matrices, any transform that uses MyAssays Analysis Expressions can access data directly from the current transform (including data that is not on the input matrices). Similar to available input matrices, the data that can be accessed depends on the

data that has been calculated up to this point of the evaluation (and data of the compatible type). Thus, the data accessible by an expression includes compatible raw and calculated matrices.

Transform Manager

Transforms are added and configured via the Transform Manager. The Transform Manager occupies the full width and height available for the transform tab's contents.

How to: Add the Transform

1. Launch an assay protocol or open a results file.

2. If you cannot see the Assay Properties panel, press the **Properties** button to open it.

3. Press the **Add** button.

4. Select the transform.

5. Press the **Create** button.

6. Review or edit the transform's input matrix and output matrix name as required.

7. Provide your required settings for the transform (see **Properties** section above).

8. Press the **Calculate** button to perform the calculations with the new settings and to see the results.

Each transform has a set of configurations to specify the parameters of the analysis. Some transforms can also add optional content to the report and be configured with specific calculation settings.

Transform Relevance

A transform may or may not be relevant to the current assay configuration. In this context, the configuration refers to:

• **Matrices (both raw and calculated)** – Certain transforms are required to be executed for a particular data type.

• **Layout** – Some transforms require specific layout rules to be followed.

General Transform Properties

With regard to flexibility of assay setup, each transform can be configured to fully match assay requirements with a number of settings.

Each transform has zero or more specific settings that allow customization of the transform operations. These are covered under each transform section specifically.

On top of that, all transforms share the following common settings:

Advanced

Included Samples	The sample types to be computed by this transform.
Excluded Results	Sample types not selected in the **Included Samples** option will not be computed by this transform; for those cases, this setting specifies what the result will be for those excluded samples: • **Flagged With Value** – The result is flagged with the input value reported. This will appear in the report with the value with strikethrough, e.g., ~~1.23~~. • **Flagged No Value** – The result is flagged with no value reported. This will appear in the report as ~~Flagged~~. • **Equal Input** – The result is set to match the value of the input.

Units

Output Units	The measurement units to be included in the header of the Sample Table. If the output units of the transform are the same as the input, then use "ditto" units to automatically carry these units through (i.e., apply the same units to the transform's output). This shortcut means that it is not necessary to repeat settings across transforms. Ditto units are specified with two dots, i.e.: ..

In addition, the **XY transforms** and **XY Reduction transforms** can be configured with **Data** and **Range** settings.

Endpoint Transforms

Endpoint transforms operate on matrices of data that consist of a single numerical value at each position on the layout. An endpoint transform will compute a single numerical result for each position on the layout.

For example, with a typical ELISA microplate assay, a plate reader will measure and report a single numerical value for each successfully measured well on the plate. Each measurement value is the endpoint of the detection technology for the observed sample. The Standard Curve Fit transform is an endpoint transform that can compute the concentration endpoint from the measurement endpoint.

> For consistency, we refer to all matrices with a single numerical result as an **endpoint** matrix. Strictly speaking, of course an endpoint matrix might not actually be the final conclusion of the analysis. With MyAssays Desktop, you can layer as many transforms as you require to perform the various analysis steps that are required (resulting in multiple "intermediary endpoint" matrices).

Flagged Endpoints

All transforms take into account the flagged status of each input value. Any data point that is flagged is not included in any calculation and ignored in any reference to the sample group that it belongs to.

For example, if a transform subtracts the average of a blank group from all samples, if the blank group is made up of 3 replicates but the 3rd replicate is flagged, then all the samples will be corrected by the average of the 2 non-flagged samples.

In addition, a transform computation for a position might result in a flagged endpoint result. This could be because of a calculation error or because the transform has performed some kind of automatic outlier detection (such Auto Flag and Replicate Outlier Removal).

Sample Types to Process

For all endpoint transforms, by default the calculations are performed on all sample types.

For greater flexibility, the **Advanced** settings of each transform can be used to select to perform the transform's computations on only a subset of the sample types. In this case, it is possible to set what the excluded samples results should be (e.g., flagged or to equal the input endpoint).

For example, it might only make sense to perform a certain calculation on Unknown and Control samples but not with other sample types.

Endpoint Transforms

Transform	Operation
Auto Flag	Flags all data points that satisfy the specified Boolean expression.
Average	Calculates the average of replicates for each sample group.
Blank Correction	Subtracts the specified blank value from each input data point.
Dilution Factors	Multiplies sample groups of the selected type(s) by a specified factor (a unique value can be specified for each group).
Dose–Response Curves	Creates a dose–response chart, fit selected curve fitting method, and calculates the response for the specified dose from the curve for each group of the specified type.
Expression by Matrix (Dual)	Evaluates the expression in terms of x and y for each sample group. The x and y variables refer to the values on the first and on the second input matrix, respectively.
Expression by Matrix	Evaluates the expression in terms of x.
Expression by Position	Evaluates an expression in terms of x for each specified position.
Factor	Multiplies all input data points of the specified sample type(s) by a specified factor.
Matrix Difference	Calculates the difference between values on the first matrix and the second for each sample group.
Median	Calculates the median of the replicates for each sample group.
Percentage Coefficient of Variation	Calculates the %CV of the replicates for each sample group.
Percentage	Calculates a percentage value for each data point with respect to the specified 0% and 100%.
Replicate Outlier Removal	Flags the specified number of replicates for each sample group, in which %CV of its replicates is greater than specified threshold.
Standard Curve Fit	Constructs a standard curve by plotting the standard input concentration values versus data points.
Standard Curve Fit (from Library)	Calculates concentrations of all included samples using a Library Curve.
Standard Deviation	Calculates the standard deviation of the replicates for each sample group.

Transform	Operation
Standard Error of Mean	Calculates the standard error of the mean for replicates of each sample group.
Variance	Calculates the variance of the replicates for each sample group.

Auto Flag

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform flags positions where a Boolean condition is satisfied. This is useful for automatically rejecting and ignoring data points that are outside an expected range.

Properties

Settings

Auto Flag	The expression to evaluate, where a single x variable is set to value of each position of the input matrix

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to flag all positions with a value greater than 1.

Here is some example raw data displayed to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

To flag values in this matrix where the measurement is greater than 1, specify an Auto Flag expression of **$x > 1$**.

Calculated

The transform will generate an output matrix named **Calculated** with the value at each position matching the input data and positions that do not meet the specified condition flagged. With the example data provided above, the results are as follows (also displayed here to 3 decimal places):

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	~~2.105~~	~~2.124~~	~~1.213~~	~~1.226~~	~~1.159~~	~~1.190~~	0.110	0.116	~~1.463~~	~~1.481~~
B	0.455	0.441	~~1.869~~	~~1.888~~	~~1.203~~	~~1.218~~	~~1.180~~	~~1.206~~	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	~~1.206~~	~~1.239~~	~~1.203~~	~~1.210~~	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	~~1.167~~	~~1.187~~	~~1.211~~	~~1.241~~	0.101	0.106	0.489	0.490
E	~~1.105~~	~~1.065~~	0.279	0.367	~~1.215~~	~~1.225~~	~~1.233~~	~~1.262~~	0.111	0.114	~~1.579~~	~~1.549~~
F	~~1.425~~	~~1.445~~	0.378	0.411	~~1.196~~	~~1.206~~	~~1.226~~	~~1.199~~	0.109	0.108	0.792	0.775
G	~~1.791~~	~~1.803~~	0.560	0.597	~~1.205~~	~~1.228~~	~~1.308~~	~~1.306~~	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	~~1.214~~	~~1.261~~	~~0.115~~	0.116	0.115	0.115	0.490	0.477

Average

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform calculates the average of the replicates in each sample group (flagged replicates are not included).

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to find the average position values in each sample group.

> This transform averages the replicates of the sample group and stores the resulting value to each sample group replicate of the output matrix. Therefore, the result will depend on the layout configuration.

Here is some example raw data displayed to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

Layout

The example uses a layout with **Unknowns** in duplicate (e.g., **Unknown1** at **A1** and **A2**, **Unknown2** at **B1** and **B2**, etc.). I.e.:

	1	2	3	4	5	6	7	8	9	10	11	12
A	1	1	2	2	3	3	4	4	5	5	6	6
B	7	7	8	8	9	9	10	10	11	11	12	12
C	13	13	14	14	15	15	16	16	17	17	18	18
D	19	19	20	20	21	21	22	22	23	23	24	24
E	25	25	26	26	27	27	28	28	29	29	30	30
F	31	31	32	32	33	33	34	34	35	35	36	36
G	37	37	38	38	39	39	40	40	41	41	42	42
H	43	43	44	44	45	45	46	46	47	47	48	48

⬤ Unknown

Average

The transform will generate an output matrix named **Average** with the value at each position being the averaged input data for each sample group.

With the example data provided above, the results are as follows (also displayed to 3 decimal places):

- **Sample Table**

Here is a Sample Table snippet, which lists the details of the calculations. For each sample, the following details are reported:

Sample	Positions	My Raw Data	Average
⬤ Unknown1	A1	0.364	0.344
	A2	0.323	
⬤ Unknown2	A3	2.105	2.115
	A4	2.124	
⬤ Unknown3	A5	1.213	1.220
	A6	1.226	
⬤ Unknown4	A7	1.159	1.175
	A8	1.190	
⬤ Unknown5	A9	0.110	0.113
	A10	0.116	

- **Matrix Table**

Here all calculated data is displayed in a Matrix Table.

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.344	0.344	2.115	2.115	1.220	1.220	1.175	1.175	0.113	0.113	1.472	1.472
B	0.448	0.448	1.879	1.879	1.211	1.211	1.193	1.193	0.131	0.131	0.777	0.777
C	0.581	0.581	0.494	0.494	1.223	1.223	1.207	1.207	0.115	0.115	0.620	0.620
D	0.790	0.790	0.371	0.371	1.177	1.177	1.226	1.226	0.104	0.104	0.490	0.490
E	1.085	1.085	0.323	0.323	1.220	1.220	1.248	1.248	0.113	0.113	1.564	1.564
F	1.435	1.435	0.395	0.395	1.201	1.201	1.213	1.213	0.109	0.109	0.784	0.784
G	1.797	1.797	0.579	0.579	1.217	1.217	1.307	1.307	0.127	0.127	0.656	0.656
H	0.262	0.262	0.467	0.467	1.238	1.238	0.116	0.116	0.115	0.115	0.484	0.484

Blank Correction

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform subtracts the average of a specified blank group (or groups) from each sample. This is useful for subtracting background noise from measurements.

The blank value is computed from the average of the (non-flagged) blank replicates.

In the simplest configuration, a single blank value is subtracted from all samples. More sophisticated correction methods are possible, including:

- 1-1 – Where each sample group is corrected by its associated blank group
- By-row – One blank group on each row
- By-column – One blank group on each column

> **i** For more complicated blank correction calculations, an alternative approach is to use the Expression by Position transform.

Properties

Settings

Blank Group	The group number of the sample to use for correction. This can be defined as a number or as an expression. The expression can refer to x, which is the group number of the sample to be corrected. For example, set this to x to perform a 1-to-1 correction where each group is corrected by the sample of the specified blank type with the same group number.
Blank Type	The sample type used as the blank.

Example: Single Blank

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to perform a blank correction using the average of one blank group. This blank group itself is measured in duplicate. The mean of these replicates will be subtracted from all other samples.

Here is some example raw data displayed to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.192	0.199	0.347	0.346	0.325	0.321	0.803	0.797	1.589	1.593	0.241	0.246
B	0.293	0.300	0.586	0.584	0.800	0.793	0.402	0.401	0.839	0.837	0.562	0.564
C	0.537	0.534	0.549	0.547	0.878	0.875	0.471	0.471	0.759	0.756	1.039	1.043
D	0.858	0.855	0.320	0.325	0.531	0.530	1.552	1.551	0.429	0.427	0.424	0.424
E	1.523	1.527	0.486	0.490	1.289	1.283	1.430	1.438	1.494	1.501	1.427	1.429
F	1.622	1.621	0.621	0.626	0.667	0.663	1.350	1.347	0.631	0.634	1.695	1.694
G	0.001	0.001	0.356	0.352	0.418	0.412	1.172	1.174	1.572	1.570	0.595	0.590
H	0.297	0.297	0.796	0.801	1.517	1.515	0.960	0.965	0.534	0.527	0.585	0.584

> This transform requires a layout with a minimum 1 **Blank** group.

Layout

The example uses a layout with **Standards**, **Control**, **Blank**, and **Unknowns** in duplicate (e.g., **Standard1** in **A1** and **A2**, **Unknown1** at **A3** and **A4**, **Unknown2** at **B3** and **B4**, etc.). I.e.:

● Standard ○ Blank ○ Control ○ Unknown

To subtract the mean of the blank positions from the measurements, specify **Blank Group** to **1** and **Blank Type** to **Blank**.

Blank Corrected

The transform will generate an output matrix named **Blank Corrected** with the value at each position being the subtraction of the blank value from the input data. With the example data provided above, the results are as follows (also displayed here to 3 decimal places):

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.191	0.198	0.346	0.345	0.324	0.320	0.802	0.796	1.588	1.592	0.240	0.245
B	0.292	0.299	0.585	0.583	0.799	0.792	0.401	0.400	0.838	0.836	0.561	0.563
C	0.536	0.533	0.548	0.546	0.877	0.874	0.470	0.470	0.758	0.755	1.038	1.042
D	0.857	0.854	0.319	0.324	0.530	0.529	1.551	1.550	0.428	0.426	0.423	0.423
E	1.522	1.526	0.485	0.489	1.288	1.282	1.429	1.437	1.493	1.500	1.426	1.428
F	1.621	1.620	0.620	0.625	0.666	0.662	1.349	1.346	0.630	0.633	1.694	1.693
G	0.000	0.000	0.355	0.351	0.417	0.411	1.171	1.173	1.571	1.569	0.594	0.589
H	0.296	0.296	0.795	0.800	1.516	1.514	0.959	0.964	0.533	0.526	0.584	0.583

Example: 1-1 Blank Correction

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to perform a 1-to-1 correction, where each sample group is corrected by the blank that it is associated with. The blank numbering used in the layout defines the associations.

Here is some example raw data displayed to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	1.962	1.999	2.038	1.922	1.995	0.002	1.628	1.670	1.624	1.554	1.701	0.000
B	0.784	0.760	0.754	0.751	0.797	0.002	1.124	1.104	1.072	1.077	1.106	0.002
C	0.586	0.575	0.582	0.590	0.576	0.001	0.724	0.737	0.690	0.711	0.717	0.002
D	0.441	0.441	0.427	0.419	0.439	0.002	0.095	0.095	0.093	0.096	0.096	0.001
E	1.956	1.970	1.875	2.043	1.959	0.001	1.449	1.479	1.428	1.395	1.385	0.001
F	1.416	1.358	1.407	1.352	1.452	0.001	0.242	0.230	0.249	0.235	0.251	0.000
G	1.438	1.429	1.488	1.401	1.372	0.002	0.943	0.952	0.956	0.912	0.969	0.002
H	0.840	0.827	0.812	0.838	0.860	0.002	1.320	1.260	1.313	1.363	1.312	0.001

Layout

The example uses a layout with 16 **Unknowns** groups (with **Unknown1** in positions from **A1** to **A5**, **Unknown2** in positions from **B1** to **B5**, etc.) and 16 **Blank** singlets I.e.:

◯ Blank ◯ Unknown

To perform 1-1 correction, specify **Blank Group** to *x* and **Blank Type** to **Blank**.

With this specified, the value of each blank will be subtracted from every position of associated group. In other words, **Blank1** (stored in **A6**) value will be subtracted from **A1, A2, A3, A4, A5** (the members of **Unknown1**), and from **A6** itself. Therefore, the calculated matrix will contain blank corrected **Unknown** values and all **Blanks** will be 0.

Blank Corrected

The transform will generate an output matrix named **Blank Corrected** with the value at each position being the subtraction of the associated blank value from the input data. With the example data provided above, the results are as follows (also displayed here to 3 decimal places):

	1	2	3	4	5	6	7	8	9	10	11	12
A	1.960	1.997	2.035	1.919	1.992	0.000	1.628	1.670	1.623	1.554	1.701	0.000
B	0.783	0.758	0.752	0.750	0.796	0.000	1.122	1.102	1.070	1.075	1.104	0.000
C	0.585	0.574	0.581	0.589	0.575	0.000	0.722	0.735	0.688	0.709	0.714	0.000
D	0.439	0.439	0.425	0.417	0.437	0.000	0.094	0.094	0.093	0.095	0.095	0.000
E	1.955	1.969	1.873	2.042	1.958	0.000	1.448	1.478	1.427	1.394	1.384	0.000
F	1.415	1.358	1.406	1.352	1.451	0.000	0.241	0.229	0.249	0.235	0.251	0.000
G	1.436	1.427	1.486	1.399	1.369	0.000	0.941	0.950	0.955	0.910	0.967	0.000
H	0.839	0.825	0.810	0.837	0.858	0.000	1.319	1.260	1.312	1.362	1.311	0.000

Advanced Example: By-Row Blank Correction

In this example, we would like to configure blank correction on a row-by-row basis to blank correct each sample by the average of the two blank samples of the same row.

For better clearness of the process, this example uses simplified endpoint data for a 12x8 microplate:

	1	2	3	4	5	6	7	8	9	10	11	12
A	1	1	2	3	4	5	6	7	8	9	10	1
B	2	11	12	13	14	15	16	17	18	19	20	2
C	3	21	22	23	24	25	26	27	28	29	30	3
D	4	31	32	33	34	35	36	37	38	39	40	4
E	5	41	42	43	44	45	46	47	48	49	50	5
F	6	51	52	53	54	55	56	57	58	59	60	6
G	7	61	62	63	64	65	66	67	68	69	70	7
H	8	71	72	73	74	75	76	77	78	79	80	8

Layout

The example uses a layout with 80 singlet **Unknowns** and 8 **Blank** duplicates for each row (with **Blank1** in positions **A1** and **A12**, **Blank2** in positions **B1** and **B12**, etc.) I.e.:

	1	2	3	4	5	6	7	8	9	10	11	12
A	1	1	9	17	25	33	41	49	57	65	73	1
B	2	2	10	18	26	34	42	50	58	66	74	2
C	3	3	11	19	27	35	43	51	59	67	75	3
D	4	4	12	20	28	36	44	52	60	68	76	4
E	5	5	13	21	29	37	45	53	61	69	77	5
F	6	6	14	22	30	38	46	54	62	70	78	6
G	7	7	15	23	31	39	47	55	63	71	79	7
H	8	8	16	24	32	40	48	56	64	72	80	8

◯ Blank ● Unknown

With the **Blank Group** setting defined as the expression:

1 + floor ((x -1) / (LayoutWidth()-2),1)

This converts the group number of the sample to blank correct (x) to the blank number on its row. In other words, for **x=1**, this means **1+floor((1-1)/10),1)**, **floor=0** and the **Blank Group** is taken as 1. For the first row, x is in range from 1 to 10 and the expression outputs 1, thus for **Unknowns** from 1 to 10 the average of **Blank1** is subtracted.

For the second, x is from 11 to 20, **floor=1**, so the **Blank2** group is subtracted from **Unknowns11-20**, etc.

Blank Corrected

	1	2	3	4	5	6	7	8	9	10	11	12
A	0	0	1	2	3	4	5	6	7	8	9	0
B	1	9	10	11	12	13	14	15	16	17	18	1
C	2	18	19	20	21	22	23	24	25	26	27	2
D	3	27	28	29	30	31	32	33	34	35	36	3
E	4	36	37	38	39	40	41	42	43	44	45	4
F	5	45	46	47	48	49	50	51	52	53	54	5
G	6	54	55	56	57	58	59	60	61	62	63	6
H	7	63	64	65	66	67	68	69	70	71	72	7

Dilution Factors

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform multiplies each selected sample group by a specified factor (a factor can be specified for each sample group).

> ℹ️ A sample prepared with a dilution of 1:2 would use a dilution factor of 2.

Properties

Settings

Factored Samples	The sample types to which the dilution factor should be applied
Factors	The dilution factor to use for each selected **Factored Samples**

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to multiply all odd **Unknown** sample groups by 2 and all even **Unknown** sample groups by 5.

Here is some example raw data displayed to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

Layout

The example uses a layout with **Standards**, **Control**, **Blank**, and **Unknowns** in duplicate (e.g., **Standard1** in **A1** and **A2**, **Unknown1** at **A3** and **A4**, **Unknown2** at **B3** and **B4**, etc.). I.e.:

Standard ● Blank ● Control ● Unknown

To multiply selected sample types in this matrix by specified factors, set **Factored Samples** to **Unknown** and specify **Factors** as **2** for every odd sample group and as **5** for every even sample group.

Concentration (x Dil. Factor)

The transform will generate an output matrix named **Concentration (x Dil. Factor)** with the value at each position of the selected Factored Samples multiplied by specified Factor and sample groups of other types matching the input data.

With the example data provided above, the results are as follows (also displayed to 3 decimal places):

- **Sample Table**

Here is a [Sample Table](#) snippet, which lists the details of the calculations for Unknown samples and the additional report content. For this transform, it is a Factor column displaying specified Factor for each sample group.

For each sample, the following details are reported:

Sample	Positions	Factor	My Raw Data	Concentration (x Dil. Factor)
🟢 Unknown1	A3	2	2.105	4.210
	A4		2.124	4.248
🟢 Unknown2	B3	5	1.869	9.345
	B4		1.888	9.440
🟢 Unknown3	C3	2	0.474	0.948
	C4		0.514	1.028
🟢 Unknown4	D3	5	0.364	1.820
	D4		0.378	1.890
🟢 Unknown5	E3	2	0.279	0.558
	E4		0.367	0.734

The location of Factor column in the Sample table can be customized in the XML tab.

How to: Set Factor Column Position in the Report

1. Select the **XML** tab in the **Assay Properties** panel (If the **XML** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Inside the **ReportContent** element, locate the Factor element.

3. Add the **SampleTableColumnPosition** element to the **Factor** element. Specify the column positioning with one of the following options:

   - **AfterOutputMatrix** – On the right of the output matrix

   - **LeftMost** – Before all matrix data

   - **RightMost** – After all matrix data

- **Matrix Table**

Here all calculated data is displayed in a [Matrix Table](#).

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	4.210	4.248	2.426	2.452	2.318	2.380	0.220	0.232	2.926	2.962
B	0.455	0.441	9.345	9.440	6.015	6.090	5.900	6.030	0.660	0.650	3.930	3.840
C	0.587	0.575	0.948	1.028	2.412	2.478	2.406	2.420	0.226	0.232	1.226	1.252
D	0.806	0.774	1.820	1.890	5.835	5.935	6.055	6.205	0.505	0.530	2.445	2.450
E	1.105	1.065	0.558	0.734	2.430	2.450	2.466	2.524	0.222	0.228	3.158	3.098
F	1.425	1.445	1.890	2.055	5.980	6.030	6.130	5.995	0.545	0.540	3.960	3.875
G	1.791	1.803	1.120	1.194	2.410	2.456	2.616	2.612	0.252	0.254	1.304	1.318
H	0.253	0.270	2.315	2.350	6.070	6.305	0.575	0.580	0.575	0.575	2.450	2.385

Dose–Response Curves

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform fits a dose–response curve for each sample group (with each sample group being dosed in the same way). The resulting dose–reponse curves are overlaid on a single chart for visual comparison.

The transform can also optionally compute and report values from each sample's curve, including:

- ED50/EC50/IC50, etc.
- ED(n)/EC(n)/IC(n), etc.
- Upper and lower bounds
- The dose at a specified response
- Any mathematical function (via an [expression](#))

Properties

Axes

X Axis Title	A name of the **X Axis** on the chart.
X Axis Type	A type to plot an **X Axis** in a chart: • **Logarithmic** • **Linear**
Y Axis Title	A name of the **Y Axis** on the chart.
Y Axis Type	A type to plot an **Y Axis** in a chart: • **Logarithmic** • **Linear**

Chart

Plot Response for Dose Calculation	If checked, then plots the **Response for Dose Calculation** (a black line showing the specified response).

Dose–Response Curves

Doses	This specifies dose concentration values for each group of the specified **Type**.

	Press on the drop-down to display the editor and specify doses settings in one of the following modes: • **Series** – Provide dose for the first sample of the specified **Type** of and common series to apply to all samples. • **Non-Serial** – Provide a specific dose value for each sample separately. Use the **Series** button to toggle the mode state. ℹ️ All sample groups are assumed to have the same number of positions defined on the layout (e.g., if **Unknown1** has 10 replicates, then all **Unknowns** on the layout must also have 10 replicates).
Response for Dose Calculation	A numerical value or an expression representing the response value to calculate the dose of for each sample.
Type	The sample type to use for dose calculation.

Fit

Fit Method	The curve fitting method to construct a dose–response curve for your data based on the specified **Type**.
Weight Method	Specifies the weight function to apply to the curve.
X Transform	Determines how the x values are interpreted in the transform: • **None** – The x values remain as they are. • **Log10** – Each x point is taken as a common logarithm of x. • **Log2** – Each x point is taken as a binary logarithm of x. • **LogN** – Each x point is taken as a natural logarithm of x.
Y Transform	Determines how the y values are interpreted in the transform: • **None** – The y values remain as they are. • **Log10** – Each y point is taken as a common logarithm of y. • **Log2** – Each y point is taken as a binary logarithm of y. • **LogN** – Each y point is taken as a natural logarithm of y.

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in

the **Raw** matrix. In this example, we would like to fit a dose–response curve to each sample group of the **Unknown** type and calculate a dose that has a response of 1.5 from the curve.

Here is some example raw data displayed here to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.000	2.682	2.345	2.612	2.732	2.675	2.664	2.574	2.580	2.361	0.871	2.839
B	0.000	2.577	1.735	2.518	2.701	2.537	2.571	2.450	2.453	2.171	0.437	2.809
C	0.000	2.537	1.290	2.497	2.683	2.447	2.490	2.273	2.344	1.695	0.222	2.783
D	0.000	2.358	0.793	2.318	2.585	2.195	2.205	1.856	2.069	1.070	0.094	2.658
E	0.000	1.958	0.369	1.799	2.224	1.636	1.661	1.192	1.485	0.550	0.037	2.322
F	0.000	1.352	0.151	1.118	1.517	0.909	0.947	0.638	0.868	0.257	0.013	1.495
G	0.000	0.745	0.053	0.540	0.872	0.425	0.485	0.282	0.417	0.094	0.002	0.862
H	0.000	0.312	0.018	0.250	0.398	0.180	0.200	0.113	0.194	0.038	0.001	0.438

Layout

The example uses a layout with one **Unused** column (in positions from **A1** to **H1**) and **Unknowns** in columns (e.g., **Unknown1** at positions from **A2** downward to **H2**, **Unknown2** at **A3-H3**, etc.). I.e.:

○ Unused ● Unknown

To create a curve and calculate concentrations for this matrix, specify the following settings:

1. Set an **X Axis Type** to **Logarithmic**.

2. Set a **X Transform** to **None**.

3. Set an **X Axis Type** to **Linear**.

4. Set a **X Transform** to **None**.

5. Check the **Plot Response for Dose Calculation** checkbox.

6. **Doses** count is equal to maximum number (8) of replicates in sample group of the **Unknown**. Set this parameter to 0.01 and **Series** to divide by 3.

7. Set a **Response for Dose Calculation** to 1.5.

8. **Type** to **Unknown**.

9. Set **Fit Method** of **4PL**.

10. Select a **Weight Method** of **None**.

11. Select a **X Transform** of **None**.

12. Select a **Y Transform** of **None**.

The transform generates a **Dose–Response Curves Table** and a **Dose–Response Curve Chart** by default. Other elements such as Matrix table and Notes can be added to report optionally.

Dose–Response Curves Table

This table lists coefficients calculated from the selected Fit Method and a resulting response value for each sample. With the example data provided above, the results are as follows:

Sample	a	b	c	d	R^2	Result
Unknown1	-0.0584816	0.875198	0.0000376679	2.67099	0.999497	0.0000522186
Unknown2	-0.0901615	0.606878	0.00226574	3.29931	0.997736	0.00184835
Unknown3	0.103291	1.07281	0.0000588689	2.59484	0.999371	0.0000738691
Unknown4	0.176798	1.10415	0.000035979	2.73875	0.999281	0.0000381935
Unknown5	0.0355624	0.998377	0.0000794665	2.64392	0.999147	0.000101774
Unknown6	0.0581275	0.980399	0.000077654	2.66458	0.999619	0.0000965551
Unknown7	0.0190159	0.929062	0.000146444	2.60957	0.999764	0.000199822
Unknown8	0.0468791	0.936045	0.000090142	2.57555	0.999425	0.000124316
Unknown9	0.0138517	0.919968	0.000515222	2.53025	0.999769	0.000767282
Unknown10	-0.00938083	0.681115	0.135122	6.05263	0.999738	0.0267169
Unknown11	0.298789	1.20751	0.0000424681	2.83485	0.999004	0.0000389155

Dose–Response Curve Chart

The chart contains a curve fit plotted with 4PL for dose and for each sample group.
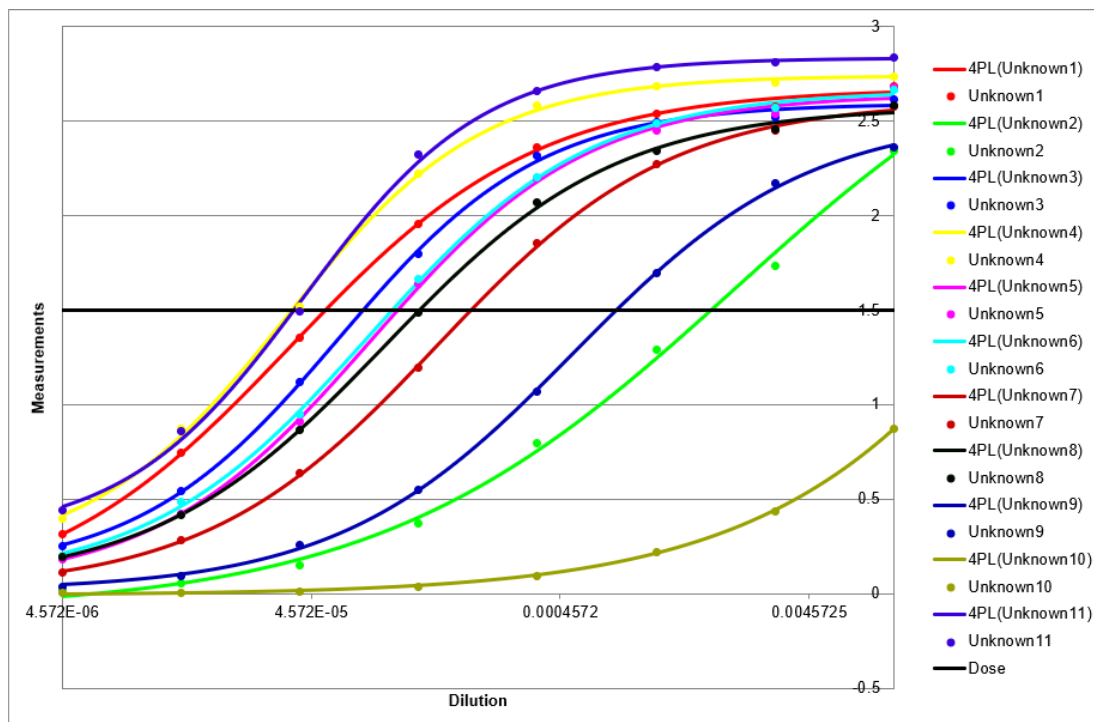
Dose

The output matrix named **Dose** <u>can be optionally added to the report</u>. It contains the calculated dose that has a response value for each data point. With the example data provided above, the results are as follows (also displayed here to 3 decimal places):

	1	2	3	4	5	6	7	8	9	10	11	12
A	~~Flagged~~	0.000	0.002	0.000	0.000	0.000	0.000	0.000	0.000	0.001	0.027	0.000
B	~~Flagged~~	0.000	0.002	0.000	0.000	0.000	0.000	0.000	0.000	0.001	0.027	0.000
C	~~Flagged~~	0.000	0.002	0.000	0.000	0.000	0.000	0.000	0.000	0.001	0.027	0.000
D	~~Flagged~~	0.000	0.002	0.000	0.000	0.000	0.000	0.000	0.000	0.001	0.027	0.000
E	~~Flagged~~	0.000	0.002	0.000	0.000	0.000	0.000	0.000	0.000	0.001	0.027	0.000
F	~~Flagged~~	0.000	0.002	0.000	0.000	0.000	0.000	0.000	0.000	0.001	0.027	0.000
G	~~Flagged~~	0.000	0.002	0.000	0.000	0.000	0.000	0.000	0.000	0.001	0.027	0.000
H	~~Flagged~~	0.000	0.002	0.000	0.000	0.000	0.000	0.000	0.000	0.001	0.027	0.000

How to: Add Custom Calculations to Dose–Response Curves

In addition to computing the dose at a specific response, this transform can compute values from each dose–response curve and report the results in a table. The Dose–Response Curves Fit Table is configured using XML.

Adding Fit Table Configuration

1. Select the **XML** tab in the **Assay Properties** panel. (If the **XML** tab is not immediately visible, press the **<< Show All Properties** button to expand.)

2. Locate the **DoseResponseCurves** element.

3. Add a **FitTable** element (as a child of the **DoseResponseCurves**).

4. Add a child element **Evaluation** for each required evaluation. Set the attributes (in accordance with the following table) and, optionally, add a **NumericalFormatting** child element.

5. Press **Apply** to save changes to the protocol.

Configuring Fit Table Evaluations in XML

The Fit Table can contain one or more **Evaluations**. A single **Evaluation** is defined with the following attributes:

Expression	An expression to evaluate for each sample. The expression can refer to any matrix and/or non-matrix data (i.e., does not reference *x* references) and results a single numerical result.
Description	A header for column with the results of the **Expression** in the report.

For each defined evaluation, you can optionally specify numerical formatting settings under the **NumericalFormatting** child element.

Numerical Formatting

Precision	The number of digits to be displayed for a numerical value. The interpretation of **Precision** depends on the **Type** setting.
Type	Specifies how a numerical value is formatted with regard to precision and rounding options: • **Significant Figures** – The value is reported to the specified number of significant figures. This accepts **Precision** values in range 1 through 14. • **Decimal Places** – The number of digits to display to the right of the decimal point. This accepts **Precision** values in range 0 through 14.

Fit Table Example XML Configuration

<FitTable>

    <Evaluation Description="Bottom" Expression="a">

        <NumericalFormatting Type="DecimalPlaces" Precision="3" />

```
        </Evaluation>
        <Evaluation Description="Top" Expression="d">
                <NumericalFormatting Type="DecimalPlaces" Precision="3" />
        </Evaluation>
        <Evaluation Description="Span" Expression="abs(d-a)">
                <NumericalFormatting Type="DecimalPlaces" Precision="3" />
        </Evaluation>
        <Evaluation Description="Hill Slope" Expression="b" />
        <Evaluation Description="EC50" Expression="c" />
        <Evaluation Description="Log(EC50)" Expression="log(c)" />
        <Evaluation Description="R²" Expression="R2" />
</FitTable>
```

Adding Fit Table to Report

Because this element is an additional advanced configuration, it must be included in the report by adding the following XML elements:

1. Locate the **ReportContent** element and add a child element **Item** with the following attributes:

   - **Id** – A unique identifier that is used to identify the report element in the Report Elements pane and when configuring custom report templates.
   - **Name** – Enter or paste **DoseResponseCurvesFitTable**
   - **Type** – Select **Table** from a drop-down list

   Example in the XML:

   ```
   <Item Id="DoseResponseCurvesFitTable1" Name="DoseResponseCurvesFitTable" Type="Table" />
   ```

2. Locate the **TransformContent Id="TransformContentDoseResponseCurves1"** (inside the **Report** element) and, for each defined Fit Table, add a child element **Content** with the following attributes:

   ItemId – Enter or paste the **Id** from **Item** element

   Include – Select **1** or **true** to include the Fit Table in the report

Example in the XML:

<Content ItemId="DoseResponseCurvesFitTable1" Include="true" />

Here is a snippet that shows how the Fit Table results are computed for **Unknown1** (with the configuration for **Unknown1** data from the example above):

Sample	Bottom	Top	Span	Hill Slope	EC50	Log(EC50)	R^2
● Unknown1	-0.058	2.671	2.729	0.875198	0.0000376679	-4.42403	0.999497

Expression by Matrix (Dual)

Input: **Dual Endpoint** ➡ Output: **Endpoint**

This transform evaluates an expression defined in terms of x and y for all data points (excluding flagged samples). **x** refers to the data for the sample on the first input matrix, and y refers to the data for the same sample on the second input matrix.

Properties

Settings

Dual Matrix Expression	The expression to evaluate, where x and y variables are set to value of each position of the input matrices

> ℹ️ If the expression does not contain the x and y variables, the transform will output the same evaluation result for all positions. For example, if you enter **{2^3+4}**, the resulting matrix will contain 12 in each position.

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw (1)** and **Raw (2)** matrices. In this example, we would like to summarize position values in these matrices.

Here is some example raw data displayed to 3 decimal places:

Raw (1)

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

Raw (2)

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.204	0.201	0.213	0.209	0.206	0.208	0.216	0.217	0.216	0.213	0.205	0.218
B	0.216	0.200	0.214	0.214	0.204	0.201	0.215	0.216	0.203	0.219	0.207	0.204
C	0.206	0.210	0.208	0.203	0.201	0.218	0.212	0.217	0.216	0.213	0.208	0.204
D	0.215	0.215	0.214	0.203	0.214	0.213	0.219	0.219	0.213	0.201	0.202	0.219
E	0.215	0.220	0.219	0.212	0.210	0.211	0.214	0.214	0.201	0.202	0.202	0.207
F	0.214	0.212	0.202	0.200	0.201	0.217	0.213	0.209	0.218	0.207	0.216	0.216
G	0.211	0.211	0.218	0.209	0.214	0.213	0.201	0.211	0.205	0.210	0.201	0.212
H	0.216	0.209	0.215	0.206	0.218	0.206	0.209	0.219	0.215	0.219	0.216	0.217

To add values from **Raw(2)** to corresponding values of **Raw(1)**, specify an expression of $x + y$.

Calculated

The transform will generate an output matrix named **Calculated** with the value at each position being the point by point sum of the **Raw (1)** and **Raw (2)** matrices values. With the example data provided above, the results are as follows (also displayed here to 3 decimal places):

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.568	0.524	2.318	2.333	1.419	1.434	1.375	1.407	0.326	0.329	1.668	1.699
B	0.671	0.641	2.083	2.102	1.407	1.419	1.395	1.422	0.335	0.349	0.993	0.972
C	0.793	0.785	0.682	0.717	1.407	1.457	1.415	1.427	0.329	0.329	0.821	0.830
D	1.021	0.989	0.578	0.581	1.381	1.400	1.430	1.460	0.314	0.307	0.691	0.709
E	1.320	1.285	0.498	0.579	1.425	1.436	1.447	1.476	0.312	0.316	1.781	1.756
F	1.639	1.657	0.580	0.611	1.397	1.423	1.439	1.408	0.327	0.315	1.008	0.991
G	2.002	2.014	0.778	0.806	1.419	1.441	1.509	1.517	0.331	0.337	0.853	0.871
H	0.469	0.479	0.678	0.676	1.432	1.467	0.324	0.335	0.330	0.334	0.706	0.694

Expression by Matrix

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform evaluates an expression defined in terms of *x* for all data points (excluding flagged samples).

Properties

Settings

Matrix Expression	The expression to evaluate, where a single *x* variable is set to value of each position of the input matrix

> ℹ If the expression does not contain the *x* variable, the transform will output the same evaluation result for all positions. For example, if you enter **{2^3+4}**, the resulting matrix will contain 12 in each position.

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to square each measurement.

Here is some example raw data displayed here to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

The **Matrix Expression** setting is specified with expression of ***x*x***.

Calculated

The transform will generate an output matrix named **Calc** with the value at each position being the square of the input data. With the example data provided above, the results are as follows (also displayed here to 3 decimal places):

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.132	0.104	4.431	4.511	1.471	1.503	1.343	1.416	0.012	0.013	2.140	2.193
B	0.207	0.194	3.493	3.565	1.447	1.484	1.392	1.454	0.017	0.017	0.618	0.590
C	0.345	0.331	0.225	0.264	1.454	1.535	1.447	1.464	0.013	0.013	0.376	0.392
D	0.650	0.599	0.132	0.143	1.362	1.409	1.467	1.540	0.010	0.011	0.239	0.240
E	1.221	1.134	0.078	0.135	1.476	1.501	1.520	1.593	0.012	0.013	2.493	2.399
F	2.031	2.088	0.143	0.169	1.430	1.454	1.503	1.438	0.012	0.012	0.627	0.601
G	3.208	3.251	0.314	0.356	1.452	1.508	1.711	1.706	0.016	0.016	0.425	0.434
H	0.064	0.073	0.214	0.221	1.474	1.590	0.013	0.013	0.013	0.013	0.240	0.228

Expression by Position

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform enables an expression to be defined for each position in terms of *x* (where *x* refers to the value of position on the input matrix). This is useful for applying various tests to different samples simultaneously.

Properties

Settings

This allows different expressions to be configured and calculated on the positions of the same index on different plates. Expressions are stored by container (1-based values are used for each container separately).

Click on a drop-down to open a **Position Expressions Editor** with grid-based view of input data. The configuration of the grid depends on the selected layout, thus each position's background is colored to match its sample type.

Position Expressions	If the protocol contains >1 plate/analyte, select the one to specify expressions for from a drop-down list in the upper right corner of the Editor. To maximize its view, click on the arrow button in the lower left corner of the Editor. Within each grid you can select and specify an expression in terms of *x* for: • A single position. • Multiple positions (within a single plate) to set to a same expression. Multiple positions can be selected by: • Dragging the mouse (for rectangular selection) • Using **Ctrl** to select random positions • Using **Shift** to select the end position (rectangular selection) ⓘ **Paste**, **Copy**, and **Clear** commands are supported. For positions with default (simply *x*) or empty expression, the output position is equal to the input position.

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to multiply positions from 1 to 5 by 10, positions 10 and 96 by 100, and position 50 by 2.

Here is some example raw data displayed to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

Provide expressions for positions in Position Expressions Editor:

- **A1**, **A2**, **A3**, **A4**, **A5** with the expression *x*10*
- **A10** and **H12** with the expression *x*100*
- **E2** with the expression *x*2*

Calculated

The transform will generate an output matrix named **Calculated** with the resulting values being the product of input data and specified expressions. With the example data provided above, the results are as follows (also displayed here to 3 decimal places):

	1	2	3	4	5	6	7	8	9	10	11	12
A	3.640	3.230	21.050	21.240	12.130	1.226	1.159	1.190	0.110	11.600	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	2.130	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	47.700

Factor

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform multiplies data points by a common factor. The factor can be specified as a numeric value or as an [expression to evaluate](#).

Properties

Settings

Factor	The value to multiply all data points by. This can be specified as a number or an [expression to evaluate](#).

Example:

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to multiply each measurement by a common factor of 2.

Here is some example raw data displayed here to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

The **Factor** setting for this example is specified as 2.

Factored

The transform will generate an output matrix named **Factored** with the value at each position being the product of the input data and the factor. With the example data provided above, the results are as follows (also displayed here to 3 decimal places):

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.728	0.646	4.210	4.248	2.426	2.452	2.318	2.380	0.220	0.232	2.926	2.962
B	0.910	0.882	3.738	3.776	2.406	2.436	2.360	2.412	0.264	0.260	1.572	1.536
C	1.174	1.150	0.948	1.028	2.412	2.478	2.406	2.420	0.226	0.232	1.226	1.252
D	1.612	1.548	0.728	0.756	2.334	2.374	2.422	2.482	0.202	0.212	0.978	0.980
E	2.210	2.130	0.558	0.734	2.430	2.450	2.466	2.524	0.222	0.228	3.158	3.098
F	2.850	2.890	0.756	0.822	2.392	2.412	2.452	2.398	0.218	0.216	1.584	1.550
G	3.582	3.606	1.120	1.194	2.410	2.456	2.616	2.612	0.252	0.254	1.304	1.318
H	0.506	0.540	0.926	0.940	2.428	2.522	0.230	0.232	0.230	0.230	0.980	0.954

Matrix Difference

Input: **Dual Endpoint** ➡ Output: **Endpoint**

This transform calculates the difference between two input matrices sample-by-sample.

> 📝 If either of the matrix inputs are flagged, the result will be set to ~~Flagged~~.

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw (1)** and **Raw (2)** matrices. In this example, we would like to find the difference between values in these matrices.

Here is some example raw data displayed here to 3 decimal places:

Raw (1)

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

Raw (2)

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.204	0.201	0.213	0.209	0.206	0.208	0.216	0.217	0.216	0.213	0.205	0.218
B	0.216	0.200	0.214	0.214	0.204	0.201	0.215	0.216	0.203	0.219	0.207	0.204
C	0.206	0.210	0.208	0.203	0.201	0.218	0.212	0.217	0.216	0.213	0.208	0.204
D	0.215	0.215	0.214	0.203	0.214	0.213	0.219	0.219	0.213	0.201	0.202	0.219
E	0.215	0.220	0.219	0.212	0.210	0.211	0.214	0.214	0.201	0.202	0.202	0.207
F	0.214	0.212	0.202	0.200	0.201	0.217	0.213	0.209	0.218	0.207	0.216	0.216
G	0.211	0.211	0.218	0.209	0.214	0.213	0.201	0.211	0.205	0.210	0.201	0.212
H	0.216	0.209	0.215	0.206	0.218	0.206	0.209	0.219	0.215	0.219	0.216	0.217

> ℹ️ If the data point values of the **Raw (2)** are greater than the values of **Raw (1)**, the resulting matrix will contain negative values.

Difference

The transform will generate an output matrix named **Difference** with the value at each position being the subtraction between values of the **Raw (1)** and **Raw (2)** matrices. With the example data provided above, the results are as follows (also displayed here to 3 decimal places):

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.160	0.122	1.892	1.915	1.007	1.018	0.944	0.973	-0.106	-0.097	1.258	1.263
B	0.239	0.241	1.656	1.674	0.999	1.017	0.965	0.990	-0.071	-0.089	0.579	0.564
C	0.381	0.365	0.266	0.311	1.005	1.021	0.991	0.993	-0.103	-0.097	0.405	0.422
D	0.591	0.559	0.150	0.175	0.953	0.974	0.992	1.023	-0.112	-0.095	0.287	0.271
E	0.890	0.845	0.060	0.155	1.005	1.014	1.019	1.048	-0.090	-0.088	1.377	1.342
F	1.211	1.234	0.176	0.211	0.995	0.989	1.013	0.990	-0.109	-0.099	0.576	0.559
G	1.580	1.592	0.342	0.389	0.991	1.016	1.107	1.095	-0.079	-0.083	0.451	0.447
H	0.037	0.061	0.248	0.264	0.996	1.055	-0.094	-0.103	-0.100	-0.104	0.274	0.260

Median

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform calculates the median of the replicates in each sample group (flagged replicates are not included).

> ℹ The median is the middle value of the values sorted into order. If there is an even number of values, the result is the average between the two middle values.

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to find median value for each measurement sample group.

Here is some example raw data displayed here to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

Layout

The example uses a layout with **Unknowns** in triplets (e.g., **Unknown1** in positions from **A1** to **A3**, **Unknown2** in from **A4** to **A6**, etc.). I.e.:

	1	2	3	4	5	6	7	8	9	10	11	12
A	1	1	1	2	2	2	3	3	3	4	4	4
B	5	5	5	6	6	6	7	7	7	8	8	8
C	9	9	9	10	10	10	11	11	11	12	12	12
D	13	13	13	14	14	14	15	15	15	16	16	16
E	17	17	17	18	18	18	19	19	19	20	20	20
F	21	21	21	22	22	22	23	23	23	24	24	24
G	25	25	25	26	26	26	27	27	27	28	28	28
H	29	29	29	30	30	30	31	31	31	32	32	32

⬤ Unknown

Median

The transform will generate an output matrix named **Median** with the value at each position being the median of replicates of each sample group. With the example data provided above, the results are as follows (also displayed to 3 decimal places):

- **Sample Table**

Here is a Sample Table snippet, which lists the details of the calculations. For each sample, the following details are reported:

Sample	Positions	My Raw Data	Median
⬤ Unknown1	A1	0.364	0.364
	A2	0.323	
	A3	2.105	
⬤ Unknown2	A4	2.124	1.226
	A5	1.213	
	A6	1.226	
⬤ Unknown3	A7	1.159	1.159
	A8	1.190	
	A9	0.110	
⬤ Unknown4	A10	0.116	1.463
	A11	1.463	
	A12	1.481	
⬤ Unknown5	B1	0.455	0.455
	B2	0.441	
	B3	1.869	

- **Matrix Table**

Here all calculated data is displayed in a [Matrix Table](#).

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.364	0.364	1.226	1.226	1.226	1.159	1.159	1.159	1.463	1.463	1.463
B	0.455	0.455	0.455	1.218	1.218	1.218	1.180	1.180	1.180	0.768	0.768	0.768
C	0.575	0.575	0.575	1.206	1.206	1.206	1.203	1.203	1.203	0.613	0.613	0.613
D	0.774	0.774	0.774	1.167	1.167	1.167	1.211	1.211	1.211	0.489	0.489	0.489
E	1.065	1.065	1.065	1.215	1.215	1.215	1.233	1.233	1.233	1.549	1.549	1.549
F	1.425	1.425	1.425	1.196	1.196	1.196	1.199	1.199	1.199	0.775	0.775	0.775
G	1.791	1.791	1.791	1.205	1.205	1.205	1.306	1.306	1.306	0.652	0.652	0.652
H	0.270	0.270	0.270	1.214	1.214	1.214	0.115	0.115	0.115	0.477	0.477	0.477

Percentage Coefficient of Variation

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform calculates the %CV of the replicates in each sample group (flagged replicates are not included).

> The **%CV** is the Standard Deviation divided by the sample group mean and multiplied by 100. (If the mean or the Standard Deviation is 0, then the result is forced to 0.)

> For groups with less than 2 non-flagged replicates, the result for the group will be set to ~~Flagged~~.

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to find the percentage coefficient of variation for each sample group of the input matrix.

Here is some example raw data displayed here to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

Layout

The example uses a layout with **Standards**, **Control**, **Blank**, and **Unknowns** in duplicate (e.g., **Standard1** in **A1** and **A2**, **Unknown1** at **A3** and **A4**, **Unknown2** at **B3** and **B4**, etc.). I.e.:

	1	2	3	4	5	6	7	8	9	10	11	12
A	1	1	1	1	9	9	17	17	25	25	33	33
B	2	2	2	2	10	10	18	18	26	26	34	34
C	3	3	3	3	11	11	19	19	27	27	35	35
D	4	4	4	4	12	12	20	20	28	28	36	36
E	5	5	5	5	13	13	21	21	29	29	37	37
F	6	6	6	6	14	14	22	22	30	30	38	38
G	1	1	7	7	15	15	23	23	31	31	39	39
H	1	1	8	8	16	16	24	24	32	32	40	40

● Standard   ○ Blank   ○ Control   ● Unknown

%CV

The transform will generate an output matrix named **%CV** with the value at each position being the percentage coefficient of variation in each sample group. The %CV is useful for measuring the deviation between the replicates. The %CV is calculated for each sample group as the % of the standard deviation of the replicate measurements divided by their mean. Typically, for replicates one might expect a %CV value to be < 15. A value greater than this might indicate a preparation or layout error. Acceptable limits depend on the application.

With the example data provided above, the results are as follows (also displayed to 3 decimal places):

- **Sample Table**

Here is a Sample Table snippet, which lists the details of the calculations. For each sample, the following details are reported:

Sample	Positions	My Raw Data	%CV
● Standard1	A1	0.364	8.440
	A2	0.323	
● Standard2	B1	0.455	2.210
	B2	0.441	
● Standard3	C1	0.587	1.460
	C2	0.575	
● Standard4	D1	0.806	2.864
	D2	0.774	
● Standard5	E1	1.105	2.607
	E2	1.065	

- **Matrix Table**

Here all calculated %CV data is displayed in a Matrix Table.

	1	2	3	4	5	6	7	8	9	10	11	12
A	8.440	8.440	0.635	0.635	0.754	0.754	1.866	1.866	3.755	3.755	0.865	0.865
B	2.210	2.210	0.715	0.715	0.876	0.876	1.541	1.541	1.080	1.080	1.638	1.638
C	1.460	1.460	5.726	5.726	1.909	1.909	0.410	0.410	1.853	1.853	1.484	1.484
D	2.864	2.864	2.668	2.668	1.202	1.202	1.730	1.730	3.416	3.416	0.144	0.144
E	2.607	2.607	19.265	19.265	0.580	0.580	1.644	1.644	1.886	1.886	1.356	1.356
F	0.986	0.986	5.915	5.915	0.589	0.589	1.575	1.575	0.652	0.652	1.534	1.534
G	0.472	0.472	4.523	4.523	1.337	1.337	0.108	0.108	0.559	0.559	0.755	0.755
H	4.597	4.597	1.061	1.061	2.686	2.686	0.612	0.612	0.000	0.000	1.901	1.901

Percentage

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform calculates a percentage value for each sample with respect to the specified 0% and 100% values.

Properties

Settings

P0 and **P100** can be defined as sample groups, evaluated in the context of the container. For example, if a multiple container layout contains 2 containers of data, both container 1 and container 2 include 2 **Control** groups (but in different places on each container). If **P0** is defined as **Control1** and **P100** is **Control2**, then when the analysis is performed, the %s for container 1 will be calculated using **Control1** and **Control2** on container 1, whereas the %s for container 2 will calculate %s using the **Control1** and **Control2** from container 2.

P0	The value treated as "0%." This can be defined as a number or as an expression. All values less than **P0** will have negative value in resulting matrix.
P100	The value treated as "100%." This can be defined as a number or as an expression. All values greater than **P100** will have values greater than 100 in resulting matrix.

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to calculate each measurement percentage value with respect to 0-3 range.

Here is some example raw data displayed here to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

To calculate all percentage values in this matrix, specify a **P0** of 0 and **P100** of 3.

%

The transform will generate an output matrix named **%**. The value at each position will equal its percent rank with respect to the specified 0% and 100%. With the example data provided above, the results are as follows (displayed here to 2 decimal places):

	1	2	3	4	5	6	7	8	9	10	11	12
A	12.13	10.77	70.17	70.80	40.43	40.87	38.63	39.67	3.67	3.87	48.77	49.37
B	15.17	14.70	62.30	62.93	40.10	40.60	39.33	40.20	4.40	4.33	26.20	25.60
C	19.57	19.17	15.80	17.13	40.20	41.30	40.10	40.33	3.77	3.87	20.43	20.87
D	26.87	25.80	12.13	12.60	38.90	39.57	40.37	41.37	3.37	3.53	16.30	16.33
E	36.83	35.50	9.30	12.23	40.50	40.83	41.10	42.07	3.70	3.80	52.63	51.63
F	47.50	48.17	12.60	13.70	39.87	40.20	40.87	39.97	3.63	3.60	26.40	25.83
G	59.70	60.10	18.67	19.90	40.17	40.93	43.60	43.53	4.20	4.23	21.73	21.97
H	8.43	9.00	15.43	15.67	40.47	42.03	3.83	3.87	3.83	3.83	16.33	15.90

Replicate Outlier Removal

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform flags up to a specified number of outlier data points in each sample group. The output matrix values match the input matrix values with any [outlier(s) flagged](#). Outliers are determined and removed as follows:

For each sample group, the transform:

1. Calculates the **%CV** of its replicates (including only the replicates not already flagged).

2. If the **%CV** is greater than the specified **%CV Threshold**, then the replicate furthest from the mean is removed.

These steps are repeated until either the new **%CV** of the replicates that are not flagged is below the threshold, or until the specified number of outliers to remove has been removed.

> ℹ️ This transform can be used to perform automatic outlier rejection (useful when samples are measured in replicate groups of 3 or more).

Properties

Settings

Outliers To Remove	Specifies the maximum number of outliers to remove. The minimum and default value for this setting is 1.
%CV Threshold	Defines threshold percentage coefficient of variance. Data point with superior or inferior values will be flagged. The default value is 15%.

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to remove 1 outlier from each sample group.

Here is some example raw data displayed to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

Layout

The example uses a layout with **Unknowns** in columns (e.g., **Unknown1** vertically at positions from **A1** to **H1**, **Unknown2** from **B2** to **H2**, etc.). I.e.:

⬤ Unknown

To flag 1 outlier data point for each sample group, specify an **Outliers To Remove** as 1 and **%CV Threshold** as 15%.

Calculated

The transform will generate an output matrix named **Calculated** with the value at each position matching the input data and outlier data points flagged. With the example data provided above, the results are as follows (also displayed here to 3 decimal places):

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	~~2.105~~	~~2.124~~	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	~~1.579~~	~~1.549~~
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	~~1.791~~	~~1.803~~	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	~~0.115~~	~~0.116~~	0.115	0.115	0.490	0.477

> ℹ If there are only 2 remaining replicates (before the flag), both of them will be flagged and a message will be added to the calculation log.

Standard Curve Fit

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform:

1. Plots the specified concentration values (*X*) against the measurement data (*Y*) for each standard
2. Fits a curve to the standard data points (optionally using weight methods)
3. Calculates concentration values from the curve
4. Determines LLOQ and ULOQ using the specified quantification limits
5. Optionally computes any additional values from the curve (such as LLD)

> ℹ️ To use **Standard Curve Fit** transform, select or create a layout with a minimum of 2 **Standard** sample groups.

> 📋 The **Library Curve** **feature** can be used to save the curve created by this transform for use in other assays (perhaps in assays where standards are not measured)

Properties

Axes

X Axis Title	A name of the **X Axis** on the chart.
X Axis Type	A type to plot an **X Axis** in a chart: • **Logarithmic** • **Linear**
Y Axis Title	A name of the **Y Axis** on the chart.
Y Axis Type	A type to plot an **Y Axis** in a chart: • **Logarithmic** • **Linear**

Calibrators

Concentrations	Defines the *x* value (the concentration) of each calibration point.
	Click on the **Concentrations** setting drop-down to display the editor. Concentration values can be specified using two different modes:

	• **Series** – Provide the concentration value of the first calibrator and define a series to compute the remaining calibrators. This is useful if your samples are in a serial dilution. • **Non-Serial** – Provide concentrations values for every calibrator. Use the **Series** button to toggle the concentration specification mode. 📝 When working with multiple plate assays, it is possible to set different standard concentrations for each plate. 📝 With a multiplex assay, concentration values can be specified for each analyte using the Multiplex Concentration Editor.
Standard Type	Defines a sample type to treat as a standard.

Chart

Points on Curve	In the drop-down editor, check any sample types to plot on the curve; this is useful to show where samples fall on the curve. The computed results of each selected sample will be plotted. In this case, all successfully computed non-flagged sample groups of the selected types will be displayed on the Standards Chart and in the report. (Only non-calibrator sample types are applicable e.g., **Unknown**, **Control**, **Blank**.)

Fit

Calculate From	Defines how the output matrix results are calculated from the computed curve for each sample (i.e., is a result calculated from the curve for each replicate or for the average of the replicates?). The options are: • **Replicates** – Every position will contain the result of *X*, calculated from value of every replicate (*Y*). • **Average** – Replicates from the same sample contain the result of *X*, calculated from averaged value of replicates (*Y*). ℹ️ Any flagged positions on the input are also flagged in the output.
Fit Expression	When using the **Custom Fit** feature, here you enter or paste the equation to fit the data with.
Fit Method	The curve-fitting method to construct a curve for your data based on the specified **Standard Type**.

	If your licence is Pro, you can also use the **Best Fit** feature to plot the most appropriate fit for your data automatically or the **Custom Fit** feature to supply your own equation/model to fit the data with.
Fit Methods	When using the **Best Fit** feature, this editor specifies the **Fit Methods** and **Weight Methods** to process your data with. You can select any combination of items in corresponding checkbox lists. 📋 At least 1 checkbox must be ticked in each listbox.
Fit Score Method	When the **Best Fit** feature is selected, to construct the curve, you can additionally specify the scoring method to be used in evaluating the goodness of applied fit methods. I.e.: • R^2 • R^2 with Low-End Accuracy • R^2 with High-End Accuracy • R^2 with Low-End and High-End Accuracy • 1/Standard-Error • 1/P-Value • 1/AIC • 1/AICc • 1/BIC
Fit To Standard	Defines which points are used as the calibrators to fit the curve to: • **Replicates** – All replicate points from each **Standard** sample group are used as calibrators. • **Average** – Average of the replicates from each **Standard** sample group are used as the calibration points for the curve.
Weight Method	Specifies the weight function for the transform.
X Transform	Determines how the *x* values are interpreted in the transform: • **None** – The *x* values remain as they are. • **Log10** – Each *x* point is taken as a common logarithm of *x*. • **Log2** – Each *x* point is taken as a binary logarithm of *x*.

	• **LogN** – Each *x* point is taken as a natural logarithm of *x*.
Y Trans-form	Determines how the *y* values are interpreted in the transform: • **None** – The *y* values remain as they are. • **Log10** – Each *y* point is taken as a common logarithm of *y*. • **Log2** – Each *y* point is taken as a binary logarithm of *y*. • **LogN** – Each *y* point is taken as a natural logarithm of *Y*.

Quantification Limits

Accuracy (±%)	Defines a percentage limit of accuracy for **Standard Type** sample groups. This is used to verify whether the accuracy of the calibrators' fits in required limits. The default value is 20%.
Precision (%CV)	Defines a %CV precision limit. This is used to verify whether %CV for each **Standard Type** sample group is lower than required limit. The default value is 20%.

The Quantification limits properties are used to determine the Lower Limit Of Quantification (LLOQ) and Upper Limit Of Quantification (ULOQ) from the fit. The LLOQ and ULOQ are computed and added to the report automatically.

The method used to calculate LLOQ and ULOQ can be overridden by setting named evaluations with corresponding **Name** settings. In this case, the custom calculated values will replace the default values.

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to construct a curve fit and calculate concentrations for input matrix.

Here is some example raw data displayed to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.192	0.199	0.347	0.346	0.325	0.321	0.803	0.797	1.589	1.593	0.241	0.246
B	0.293	0.300	0.586	0.584	0.800	0.793	0.402	0.401	0.839	0.837	0.562	0.564
C	0.537	0.534	0.549	0.547	0.878	0.875	0.471	0.471	0.759	0.756	1.039	1.043
D	0.858	0.855	0.320	0.325	0.531	0.530	1.552	1.551	0.429	0.427	0.424	0.424
E	1.523	1.527	0.486	0.490	1.289	1.283	1.430	1.438	1.494	1.501	1.427	1.429
F	1.622	1.621	0.621	0.626	0.667	0.663	1.350	1.347	0.631	0.634	1.695	1.694
G	0.001	0.001	0.356	0.352	0.418	0.412	1.172	1.174	1.572	1.570	0.595	0.590
H	0.297	0.297	0.796	0.801	1.517	1.515	0.960	0.965	0.534	0.527	0.585	0.584

Layout

The example uses a layout with **Standards**, **Control**, **Blank**, and **Unknowns** in duplicate (e.g., **Standard1** in **A1** and **A2**, **Unknown1** at **A3** and **A4**, **Unknown2** at **B3** and **B4**, etc.). I.e.:

● Standard ● Blank ● Control ● Unknown

To configure a **Standard Curve Fit** for this matrix, provide following settings:

1. Set **X Axis Title** to Concentration.

2. Set **X Axis Type** to Logarithmic.

3. Set **Y Axis Title** to Raw.

4. Set **X Axis Type** to Linear.

5. Set **Concentrations** in **Standard1** to 0.125 and a **Series** to multiply by 2.

6. Set **Standard Type** to **Standard**.

7. Set **Calculate From** to **Average**.

8. Select a **Fit Method** of **4PL**.

9. Set **Fit To Standard** to **Average**.

10. Select a **Weight Method** of **None**.

11. Set a **X Transform** to **None**.

12. Set a **X Transform** to **None**.

13. Set **Accuracy (±%)** to 20%.

14. Set **Precision (%CV)** to 20%.

Concentration

The transform will generate an output matrix named **Concentration** with concentrations (x) calculation results for all samples using the curve fit. With the example data provided above, the results are as follows (also displayed here to 3 decimal places):

	1	2	3	4	5	6	7	8	9	10	11	12
A	Flagged	Flagged	0.352	0.352	0.318	0.318	0.844	0.844	2.890	2.890	0.153	0.153
B	0.274	0.274	0.623	0.623	0.841	0.841	0.423	0.423	0.886	0.886	0.600	0.600
C	0.572	0.572	0.585	0.585	0.928	0.928	0.503	0.503	0.799	0.799	1.131	1.131
D	0.906	0.906	0.317	0.317	0.566	0.566	2.561	2.561	0.454	0.454	0.450	0.450
E	2.390	2.390	0.521	0.521	1.550	1.550	1.969	1.969	2.240	2.240	1.947	1.947
F	3.243	3.243	0.662	0.662	0.704	0.704	1.702	1.702	0.671	0.671	5.174	5.174
G	Flagged	Flagged	0.362	0.362	0.439	0.439	1.332	1.332	2.710	2.710	0.630	0.630
H	0.275	0.275	0.843	0.843	2.338	2.338	1.030	1.030	0.566	0.566	0.622	0.622

Click here to learn about Multiple Plate Standard Curve Fit Analysis.

Example Additional XML Evaluation and Flagging Configuration

In addition to plotting a curve fit and calculating concentrations, additional values can be obtained with Standard Curve Evaluations. Moreover, with Standard Curve Custom Flag Labels, you can exclude from analysis and mark with a custom label samples that are, for example, outside the range of the standard calibrator.

In this example (using the data provided above):

- Lower Limit of Detection (LLD) is computed from the curve by calculating the concentration at the average of the blank measurements + 3 * standard deviation of the blank positions.
- The **Name** attribute for LLD is specified and is used as a variable to flag condition x<LLD.

XML Configuration

The set attributes are highlighted in yellow.

\<Settings>

  \<StandardCurveFitStandardType="2" FitMethod="4PL" Concentrations="0.25,0.5,1,2,4,8" XAxisTitle="Concentration" YAxisTitle="OD" FitToStandard="Average" CalculateFrom="Average">

      \<Evaluations>

        \<EvaluationType="CalcY" Expression="Blank + (3 * sd(Blank))" Description="My LLD"Name="LLD" />

      \</Evaluations>

      \<Flags>

        \<FlagType="Input"Condition="x < LLD"Label="Below LLD" />

      \</Flags>

  \</StandardCurveFit>

\</Settings>

Upon calculating data with the aforementioned XML configuration defined, the LLD evaluation is computed and added to the report below an interactive chart. It is displayed in the table downward to LLOQ and ULOQ, which are calculated from the curve automatically.

LLOQ	0.25
ULOQ	2
My LLD	0.222063

With the example data provided above, the results are computed as follows:

	1	2	3	4	5	6	7	8	9	10	11	12
A	Below LLD	Below LLD	0.352	0.352	0.318	0.318	0.844	0.844	2.890	2.890	0.153	0.153
B	0.274	0.274	0.623	0.623	0.841	0.841	0.423	0.423	0.886	0.886	0.600	0.600
C	0.572	0.572	0.585	0.585	0.928	0.928	0.503	0.503	0.799	0.799	1.131	1.131
D	0.906	0.906	0.317	0.317	0.566	0.566	2.561	2.561	0.454	0.454	0.450	0.450
E	2.390	2.390	0.521	0.521	1.550	1.550	1.969	1.969	2.240	2.240	1.947	1.947
F	3.243	3.243	0.662	0.662	0.704	0.704	1.702	1.702	0.671	0.671	5.174	5.174
G	Below LLD	Below LLD	0.362	0.362	0.439	0.439	1.332	1.332	2.710	2.710	0.630	0.630
H	0.275	0.275	0.843	0.843	2.338	2.338	1.030	1.030	0.566	0.566	0.622	0.622

TOVs:

This transform produces the following TOVs.

Here the **{n}** specifies the position of Standard Curve Fit in the sequence of transforms configured via **Transform** tab.

TOV	Refers to
Coefficients of the Fit Method	
The calculated values of the coefficients used in the selected fit method's equation.	
The actual coefficients available will depend on the fit method, please refer to Curve Fitting Methods for more details. For example, with Linear Regression, the coefficients **m** and **c** will be available.	
For example, the Four Parameter Logistic Regression:	
TOV{n}.a	The **a** coefficient value of 4PL
TOV{n}.b	The **b** coefficient value of 4PL
TOV{n}.c	The **c** coefficient value of 4PL
TOV{n}.d	The **d** coefficient value of 4PL
Goodness of Fit Measures	
The values of calculated goodness measures determined from the curve fit.	
TOV{n}.R2	The R^2 value
TOV{n}.AR2	The adjusted R^2 value
TOV{n}.SE	The Standard Error value

TOV	Refers to
TOV{n}.F	The F-Statistic value
TOV{n}.P	The P-value
TOV{n}.AIC	The Akaike's Information Criterion value
TOV{n}.AICc	The Akaike's Information Criterion corrected for small sizes value
TOV{n}.BIC	The Bayesian Information Criterion value
TOV{n}.DWS	The Durbin-Watson Statistic value
TOV{n}.DoF	The number of Degrees of Freedom If there are not enough **Degrees of Freedom**, then values for **P**, **F**, **SE** and **aR^2** cannot be computed.
TOV{n}.RMS	The Root Mean Square of the residuals value (reported for Auto Smoothed Spline only)
Quantification Limits	
The lower and upper concentration values determined from the curve.	
TOV{n}.ULOQ	The determined ULOQ value of the curve fit
TOV{n}.LLOQ	The determined LLOQ value of the curve fit
Sum of Squares	
The sum of squared distances from the fit to the data.	
TOV{n}.SSR	The sum of squares computed from input y replicates and y average (flagged points not included)
TOV{n}.SSC	The sum of squares computed from input y average and y' average (flagged points not included) The y' is computed from curve at x
Named Evaluations	
If there are any Named Evaluations defined for the Standard Curve Fit transform, they can also be referenced in the TOVs. For example:	
TOV{n}.LLD	The determined value of Lower Limit of Detection for the curve fit

Standard Curve Fit (from Library)

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform uses a Library Curve to calculate concentration values.

> 📋 This transform will compute concentrations values using a standards curve that was measured and constructed in a previous assay run. In this way, concentrations can be computed from a library curve without needing to measure new standards as part of this protocol.

Properties

Settings

Library Curve File	Specifies the full file path to the Library Curve file.

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to use a curve created from a previous assay run that used the Standard Curve Fit transform. This library curve has been saved with the name "My Library Curve".

Here is some example raw data displayed to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.269	0.282	2.016	2.063	0.474	0.497	0.403	0.386	0.032	0.031	0.058	0.058
B	2.499	2.607	2.003	2.001	0.298	0.292	2.316	2.250	1.460	1.500	2.206	2.190
C	1.257	1.255	0.351	0.367	2.407	2.514	1.657	1.674	0.502	0.502	0.117	0.122
D	1.211	1.253	2.035	2.079	1.100	1.079	1.382	1.407	2.436	2.487	0.127	0.126
E	1.044	1.067	1.274	1.274	2.002	2.058	1.674	1.646	0.201	0.199	0.581	0.593
F	2.090	2.030	0.304	0.291	0.910	0.903	0.572	0.572	2.217	2.293	0.013	0.013
G	1.228	1.192	1.934	1.890	0.561	0.544	1.133	1.167	1.087	1.118	1.322	1.297
H	1.092	1.056	1.996	1.980	2.061	2.106	2.198	2.111	1.552	1.491	0.002	0.002

Layout

The example uses a layout with **Unknowns** in duplicate (e.g., **Unknown1** in **A1** and **A2**, **Unknown2** at **B1** and **B2**, etc.). I.e.:

⦿ Unknown

To reuse an existing curve, select "My Library Curve" file from Curve Library and apply it to calculate results for an assay.

Concentration

The transform will generate an output matrix named **Concentration** with concentrations (**X**) calculation results for all samples using the curve from the specified library curve file. With the example data provided above, the results are as follows (also displayed here to 3 decimal places):

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.234	0.234	~~Flagged~~	~~Flagged~~	0.519	0.519	0.415	0.415	~~Flagged~~	~~Flagged~~	~~Flagged~~	~~Flagged~~
B	~~Flagged~~	~~Flagged~~	~~Flagged~~	~~Flagged~~	0.272	0.272	~~Flagged~~	~~Flagged~~	2.156	2.156	~~Flagged~~	~~Flagged~~
C	1.486	1.486	0.369	0.369	~~Flagged~~	~~Flagged~~	4.074	4.074	0.536	0.536	~~Flagged~~	~~Flagged~~
D	1.439	1.439	~~Flagged~~	~~Flagged~~	1.200	1.200	1.835	1.835	~~Flagged~~	~~Flagged~~	~~Flagged~~	~~Flagged~~
E	1.151	1.151	1.524	1.524	~~Flagged~~	~~Flagged~~	3.935	3.935	~~Flagged~~	~~Flagged~~	0.625	0.625
F	~~Flagged~~	~~Flagged~~	0.276	0.276	0.963	0.963	0.609	0.609	~~Flagged~~	~~Flagged~~	~~Flagged~~	~~Flagged~~
G	1.397	1.397	~~Flagged~~	~~Flagged~~	0.589	0.589	1.293	1.293	1.219	1.219	1.616	1.616
H	1.178	1.178	~~Flagged~~	~~Flagged~~	~~Flagged~~	~~Flagged~~	~~Flagged~~	~~Flagged~~	2.369	2.369	~~Flagged~~	~~Flagged~~

> All TOVs, produced by the original Standard Curve Fit transform can be referenced from where the Standard Curve Fit (from Library) transforms is used.

Standard Deviation

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform calculates the standard deviation of the replicates in each sample group (flagged replicates are not included). The sample of population method is used.

> The standard deviation is a measure of how widely the replicate values are dispersed from the average of the group.

> For groups with less than 2 non-flagged replicates, the result for the group will be set to ~~Flagged~~.

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to find the deviation between replicates in each sample group.

Here is some example raw data displayed here to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

Layout

The example uses a layout with **Standards**, **Control**, **Blank**, and **Unknowns** in duplicate (e.g., **Standard1** in **A1** and **A2**, **Unknown1** at **A3** and **A4**, **Unknown2** at **B3** and **B4**, etc.). I.e.:

	1	2	3	4	5	6	7	8	9	10	11	12
A	1	1	1	1	9	9	17	17	25	25	33	33
B	2	2	2	2	10	10	18	18	26	26	34	34
C	3	3	3	3	11	11	19	19	27	27	35	35
D	4	4	4	4	12	12	20	20	28	28	36	36
E	5	5	5	5	13	13	21	21	29	29	37	37
F	6	6	6	6	14	14	22	22	30	30	38	38
G	1	1	7	7	15	15	23	23	31	31	39	39
H	1	1	8	8	16	16	24	24	32	32	40	40

● Standard ○ Blank ○ Control ○ Unknown

SD

The transform will generate an output matrix named **SD** with the value at each position being the product of the input data and deviation between positions of each sample group. With the example data provided above, the results are as follows (also displayed to 3 decimal places):

- **Sample Table**

Here is a Sample Table snippet, which lists the details of the calculations. For each sample, the following details are reported:

Sample	Positions	My Raw Data	SD
● Standard1	A1	0.364	0.029
	A2	0.323	
● Standard2	B1	0.455	0.010
	B2	0.441	
● Standard3	C1	0.587	0.008
	C2	0.575	
● Standard4	D1	0.806	0.023
	D2	0.774	
● Standard5	E1	1.105	0.028
	E2	1.065	

- **Matrix Table**

Here all calculated data is displayed in a Matrix Table.

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.029	0.029	0.013	0.013	0.009	0.009	0.022	0.022	0.004	0.004	0.013	0.013
B	0.010	0.010	0.013	0.013	0.011	0.011	0.018	0.018	0.001	0.001	0.013	0.013
C	0.008	0.008	0.028	0.028	0.023	0.023	0.005	0.005	0.002	0.002	0.009	0.009
D	0.023	0.023	0.010	0.010	0.014	0.014	0.021	0.021	0.004	0.004	0.001	0.001
E	0.028	0.028	0.062	0.062	0.007	0.007	0.021	0.021	0.002	0.002	0.021	0.021
F	0.014	0.014	0.023	0.023	0.007	0.007	0.019	0.019	0.001	0.001	0.012	0.012
G	0.008	0.008	0.026	0.026	0.016	0.016	0.001	0.001	0.001	0.001	0.005	0.005
H	0.012	0.012	0.005	0.005	0.033	0.033	0.001	0.001	0.000	0.000	0.009	0.009

Standard Error of Mean

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform calculates the standard error of the mean (SEM) of the replicates in each sample group (flagged replicates are not included). The sample of population method is used.

> The standard error of the mean quantifies how precisely the average of the replicates estimates the true mean of the population. Lower values of SEM indicate more precise estimates of the population mean. Typically, increasing the number of replicates will result in a smaller SEM value and a more precise estimate.

> For groups with less than 2 non-flagged replicates, the result for the group will be set to ~~Flagged~~.

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to find the SEM of each sample group.

Here is some example raw data displayed to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

Layout

The example uses a layout with **Unknowns** in triplets (e.g., **Unknown1** in positions from **A1** to **A3**, **Unknown2** in from **A4** to **A6**, etc.). I.e.:

	1	2	3	4	5	6	7	8	9	10	11	12
A	1	1	1	2	2	2	3	3	3	4	4	4
B	5	5	5	6	6	6	7	7	7	8	8	8
C	9	9	9	10	10	10	11	11	11	12	12	12
D	13	13	13	14	14	14	15	15	15	16	16	16
E	17	17	17	18	18	18	19	19	19	20	20	20
F	21	21	21	22	22	22	23	23	23	24	24	24
G	25	25	25	26	26	26	27	27	27	28	28	28
H	29	29	29	30	30	30	31	31	31	32	32	32

◯ Unknown

SEM

The transform will generate an output matrix named **SEM** with the value at each position being the product of the input data and the standard deviation divided by the square root of the number of non-flagged replicates in each group. With the example data provided above, the results are as follows (also displayed to 3 decimal places):

- **Sample Table**

Here is a Sample Table snippet, which lists the details of the calculations. For each sample, the following details are reported:

Sample	Positions	My Raw Data	SEM
● Unknown1	A1	0.364	0.587
	A2	0.323	
	A3	2.105	
● Unknown2	A4	2.124	0.302
	A5	1.213	
	A6	1.226	
● Unknown3	A7	1.159	0.355
	A8	1.190	
	A9	0.110	
● Unknown4	A10	0.116	0.452
	A11	1.463	
	A12	1.481	
● Unknown5	B1	0.455	0.474
	B2	0.441	
	B3	1.869	

- **Matrix Table**

Here all calculated data is displayed in a [Matrix Table](#).

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.587	0.587	0.587	0.302	0.302	0.302	0.355	0.355	0.355	0.452	0.452	0.452
B	0.474	0.474	0.474	0.226	0.226	0.226	0.354	0.354	0.354	0.216	0.216	0.216
C	0.036	0.036	0.036	0.236	0.236	0.236	0.365	0.365	0.365	0.168	0.168	0.168
D	0.142	0.142	0.142	0.266	0.266	0.266	0.375	0.375	0.375	0.128	0.128	0.128
E	0.269	0.269	0.269	0.284	0.284	0.284	0.379	0.379	0.379	0.483	0.483	0.483
F	0.352	0.352	0.352	0.263	0.263	0.263	0.368	0.368	0.368	0.225	0.225	0.225
G	0.412	0.412	0.412	0.207	0.207	0.207	0.394	0.394	0.394	0.176	0.176	0.176
H	0.067	0.067	0.067	0.256	0.256	0.256	0.000	0.000	0.000	0.123	0.123	0.123

Variance

[Input: **Endpoint** ➡ Output: **Endpoint**]

This transform calculates the variance of the replicates in each sample group ([flagged]() replicates are not included). The sample of population method is used.

> ℹ The variance is the sum of the squares of the difference of each replicate and the mean divided by **n-1** (where **n** is the number of the replicates).

> 📋 For groups with less than 2 non-flagged replicates, the result for the group will be set to ~~Flagged~~.

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to find the variance between measurements in each sample group.

Here is some example raw data displayed to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

Layout

The example uses a layout with **Standards**, **Control**, **Blank**, and **Unknowns** in duplicate (e.g., **Standard1** in **A1** and **A2**, **Unknown1** at **A3** and **A4**, **Unknown2** at **B3** and **B4**, etc.). I.e.:

	1	2	3	4	5	6	7	8	9	10	11	12
A	1	1	1	1	9	9	17	17	25	25	33	33
B	2	2	2	2	10	10	18	18	26	26	34	34
C	3	3	3	3	11	11	19	19	27	27	35	35
D	4	4	4	4	12	12	20	20	28	28	36	36
E	5	5	5	5	13	13	21	21	29	29	37	37
F	6	6	6	6	14	14	22	22	30	30	38	38
G	1	1	7	7	15	15	23	23	31	31	39	39
H	1	1	8	8	16	16	24	24	32	32	40	40

● Standard ○ Blank ○ Control ● Unknown

Variance

The transform will generate an output matrix named **Variance** with the values at each sample group being the product of the input data variance. With the example data provided above, the results are as follows (also displayed to 3 decimal places):

- **Sample Table**

Here is a Sample Table snippet, which lists the details of the calculations. For each sample, the following details are reported:

Sample	Positions	My Raw Data	Variance
● Standard1	A1	0.364	0.001
	A2	0.323	
● Standard2	B1	0.455	0.000
	B2	0.441	
● Standard3	C1	0.587	0.000
	C2	0.575	
● Standard4	D1	0.806	0.001
	D2	0.774	
● Standard5	E1	1.105	0.001
	E2	1.065	

- **Matrix Table**

Here all calculated data is displayed in a Matrix Table.

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.001	0.001	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
B	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
C	0.000	0.000	0.001	0.001	0.001	0.001	0.000	0.000	0.000	0.000	0.000	0.000
D	0.001	0.001	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
E	0.001	0.001	0.004	0.004	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
F	0.000	0.000	0.001	0.001	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
G	0.000	0.000	0.001	0.001	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
H	0.000	0.000	0.000	0.000	0.001	0.001	0.000	0.000	0.000	0.000	0.000	0.000

General Transform Properties for XY Data

The transforms that operate on the XY Data are configured with:

- general settings for all transforms
- general settings for XY and XY Reduction transforms (described below)
- specific settings (if available)

General Settings for XY and XY Reduction Transforms

Data

X Axis	This setting determines which values to use in the **X** chart axis and for evaluations:
	
	- **Index** – The 1-based position number of the data point in the collection.
	- **X** – Depending on the measurements data type, this option is displayed as:
	    - **X (Cycle)** – For kinetic data with no time values, **X** equals the **Index**.
	    - **X (Time)** – For kinetic data, if time values are stored, then this is the time stamp associated with the data point.
	    - **X (Wavelength)** – For spectral data, if wavelength values are stored, then this is the wavelength of the data point.
	- **Y** – Each value is set to a y value of the corresponding input matrix's data point.

Y Axis	This setting determines which values to consider in the **Y** chart axis and for evaluations:
	
	- **Index** – The 1-based position number of the data point in the collection.
	- **X**– Depending on the measurements data type, this option is displayed as:
	    - **X (Cycle)** – For kinetic data with no time values, **X** equals the **Index**.
	    - **X (Time)** – For kinetic data, if time values are stored, then this is the time stamp associated with the data point.
	    - **X (Wavelength)** – For spectral data, if wavelength values are stored, then this is the wavelength of the data point.
	- **Y** – Each value is set to a y value of the corresponding input matrix's data point.

Range

The range settings enable the specification of a subset of the data points to consider; only data points within the specified range will be processed. When a range is set, data points outside the specified range are excluded from the calculation. Thus, only the data points inside the specified minimum and maximum range (that are also not flagged) are processed by the transform.

The range can be defined by setting the **X Minimum** and **X Maximum** properties to a value or expression:

X Minimum	A numerical value or expression that defines the minimum x data point to include within the subset. If this is not specified, then all data points less than **X Maximum** are included.
X Maximum	A numerical value or expression that defines the maximum x data point to include within the subset. If this is not specified, then all data points greater than **X Minimum** are included.

> To refer to the position's corresponding data on an endpoint matrix, use a Matrix Position Reference (e.g., **#2**) to refer to the data on the second matrix. This is useful to set the range of calculations for each sample to be based on the results of previous calculation steps.

In addition, the range can be adjusted interactively by positioning range bars. The range bars are displayed as vertical dashed lines on the chart that you can either:

- Drag with mouse to correct range values
- Double-click on to reset minimum or maximum value

XY Transforms

XY transforms operate on matrices of data that consist of XY data and result in a new XY data matrix.

For example, the XY Baseline Correction transform can be used to remove the inherent baseline from each sample to result in a new XY plot with the measurements shifted down the Y axis.

XY Data

Throughout MyAssays Desktop, we refer to measurements that yield a 2-dimensional data sets for each sample in general terms as "XY Data". This encompasses:

- Kinetic data with measurements made over time (with or without time values).

- Spectral data measured at different wavelengths.

- Scan data measured as a single line (with a distance from an origin).

XY Axes

With certain measurement types, there might be different data available to plot on the X and Y axes. For example, with kinetic data measured over time you might want to use the time values as the X axis or the cycle number instead. The axis settings specify what data should be processed by the transform. For further details you can refer to the General Transform Properties for XY Data subsection.

Flagged XY Data Points

Any data point that is part of an XY data set can be flagged. Flagged data points are excluded from the calculation.

For example, with the XY Baseline Correction transform, if the baseline is computed using the average of the 4 lowest measurements but the lowest measurement is flagged, then the baseline will be computed using the next 4 lowest data points.

In addition to each data point, each position can also be flagged to completely exclude the sample from any further analysis. This follows the same convention as for Flagged Endpoint.

XY Range

By default, XY data transform calculations are performed on all XY data points that are not flagged. However, it is also possible to define a range of points to process. This is useful if the area of interest is only a subset of the measurements. The range minimum and maximum values can be set as an absolute value or as an expression (that can in turn refer to other computed results).

For example, the XY Expression transform could be configured to work with input XY data that has 100 data points and to use only points >=20 and <= 80. In this case, the resulting output XY matrix would contain only this subset.

Refer to the General Transform Properties for XY Data subsection for more details.

XY Transforms

Transform	Operation
Baseline Correction	This transform removes the inherent variable background, or baseline, from each input position.
Blank Correction	Subtracts the average of specified Blank group(s) from each input position.
Expression (Dual)	Evaluates an expression in terms of x and y, where variables are set to the value of each position of the first (x) and second (y) input matrices.
Expression	Evaluates the specified expression for each input position.
Fit	Fits a curve to each used XY plot.
Join	Creates new combined XY plot from two input XY matrices for each position by appending data points from the second matrix to those of the first.
Outlier Removal	Flags data points that are considered to be outliers. The values of the output matrix match the input matrix (the only difference being that outlier(s) might be flagged).
Outlier Removal (for Horizontal)	Flags the specified number of data points for each plot, in which %CV of its points is greater than specified threshold.
Replicates Average	Calculates the XY average plot for each sample group.
Replicates Function	Calculates the new plot for each sample group by using specified function on its replicates.

XY Baseline Correction

[Input: **XY** ➡ Output: **XY**]

This transform removes the inherent variable background (or baseline) from each XY position.

Flagged positions or points are not included in the calculations. The operation is only performed on data within specified range.

Properties

Settings

Method	The method for a baseline correction: • **Average Between X1 And X2** – Correct points by the average *Y* of the non-flagged points between and including points **X Index 1** and **X Index 2**. • **Average Of N Lowest** – Correct points by the average *Y* of the lowest non-flagged **N**. • **Proportional N1 Lowest and N2 Highest** – Proportional percentage between average of the **N1** lowest and **N2** highest non-flagged points.
	The visibility of the following settings depend on the selected **Method**. For example, if you choose the **Average Of N Lowest**, then only the **N** setting will be displayed.
X Index 1 **X Index 2**	These settings are used in the **Average Between X1 And X2** method. These are 1-based index numbers and apply to the data sets with or without time values. Only non-flagged points that are **<= X Index 1** and **>= X Index 2** are used. **X Index 1** and **X Index 2** must be **>=1** and **X Index 2** must be **>= X Index 1**. If there are no included points between **X Index 1** and **X Index 2**, then no correction will be made and the output position will be flagged.
N	This setting is used in the **Average Of N Lowest** method. It is the number of lowest non-flagged data points within a subset. If the number of included points **< N**, then the output position will be flagged. If the number of included points **= N**, then the result is that all points are corrected by the average of all points.
N1	These settings are used in the **Proportional N1 Lowest and N2 Highest** method.
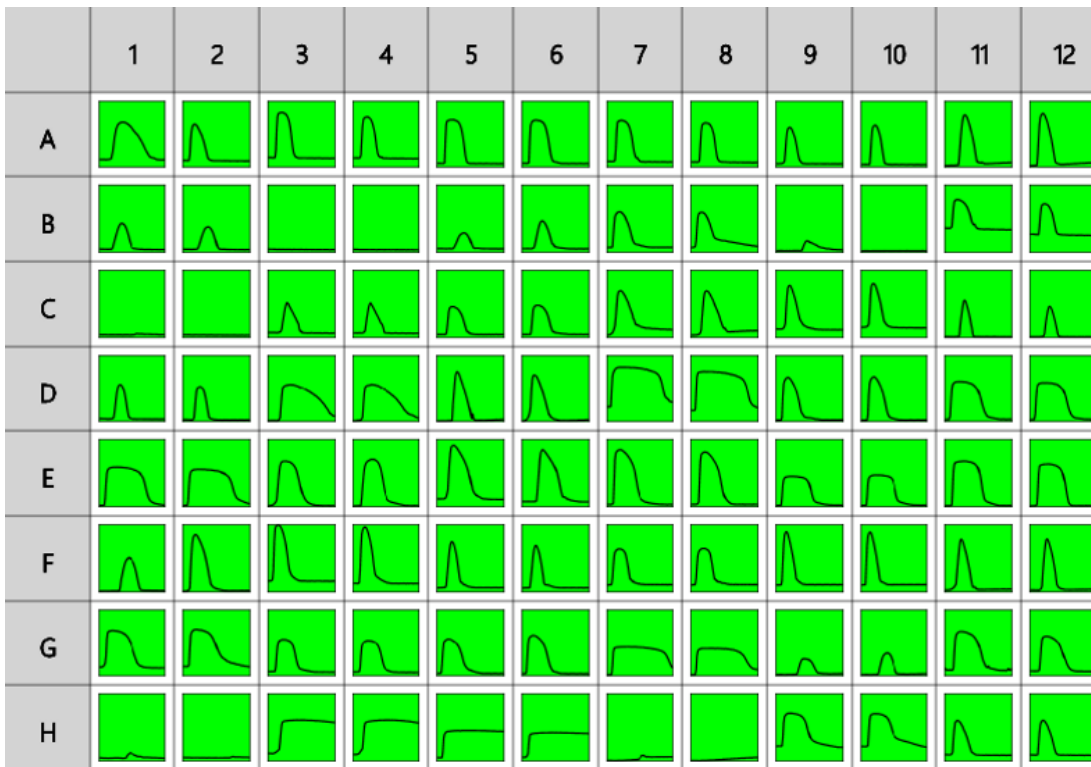
N2	**N1** is the number of the lowest points, **N2** is the number of the highest points to used for correction. **N1** and **N2** must be **>= 1**. The average of each of these is computed. Then each *Y* value is scaled between these points (e.g., if a point equals the average of the lowest, then it is set to 0; or if a point equals the average of the highest, it is set to 100).

Also, note results here are clipped. In other words, all results will fall within the range 0 and 100, so values which are calculated to < 0 or > 100 will be set to 0 and 100, respectively.

If the average of the lowest values is equal to the average of the highest values, then this cannot be calculated and the output position will be flagged. If there are less than **N1** or **N2** valid readings, then the output position will be flagged. |

Example

Let's assume that we have some XY measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to remove the inherent variable background from each point. Here this background is measured as the average of five lowest points for each XY plot and is then subtracted from each point of this plot. The calculations for the **A1** position are shown for illustration of the transform.

Raw

Here is an **XY** matrix with each position containing 80 XY data points. Position data plots of the matrix are displayed in Plate Scale.

The raw data is displayed here to 3 decimal places:

A1 317.780;0.235 377.710;0.230 437.630;0.231 497.530;0.232 557.440;0.229 617.360;0.232 677.280;0.234 737.180;0.242 797.080;0.360 856.980;0.567 916.900;0.705 976.840;0.766 1036.760;0.791 1096.680;0.798 1156.580;0.800 1216.480;0.797 1276.410;0.786 1336.330;0.769 1396.230;0.746 1456.150;0.721 1516.050;0.696 1575.980;0.666 1635.900;0.634 1695.850;0.599 1755.750;0.560 1815.670;0.518 1875.590;0.470 1935.510;0.416 1995.410;0.364 2055.320;0.319 2115.220;0.277 2175.120;0.261 2235.020;0.255 2294.970;0.251 2354.890;0.233 2414.790;0.231 2474.690;0.228 2534.620;0.227 2594.520;0.229 2654.420;0.226 2714.340;0.226 2774.280;0.225 2834.180;0.226 2894.100;0.225 2954.030;0.226 3013.970;0.225 3073.900;0.224 3133.800;0.227 3193.700;0.225 3253.600;0.224 3313.500;0.225 3373.420;0.225 3433.340;0.224 3493.270;0.224 3553.210;0.223 3613.130;0.223 3673.030;0.224 3732.950;0.223 3792.860;0.224 3852.760;0.223 3912.660;0.223 3972.580;0.223 4032.510;0.223 4092.460;0.222 4152.390;0.222 4212.330;0.225 4272.230;0.223 4332.130;0.223 4392.030;0.223 4451.930;0.223 4511.830;0.223 4571.730;0.222 4631.650;0.221 4691.560;0.220 4751.500;0.224 4811.420;0.222 4871.350;0.222 4931.280;0.222 4991.180;0.222 5051.080;0.222

To correct each point with the specified baseline:

1. Specify **Data**: **X Axis** as **X (Time)**, **Y Axis** as **Y**.

2. Leave **X Minimum** and **X Maximum** blank.

3. Specify **Settings**: **Method** as **Average Of N Lowest**, **N** as **5**.

4. Press **Calculate**.

Corrected

The transform will generate an output XY matrix named **Corrected** where each position will contain the plot with the specified baseline subtracted from each input point.

With the example data provided above, the result for the **A1** plot is displayed on the Kinetic/Spectral Analysis Chart as follows:
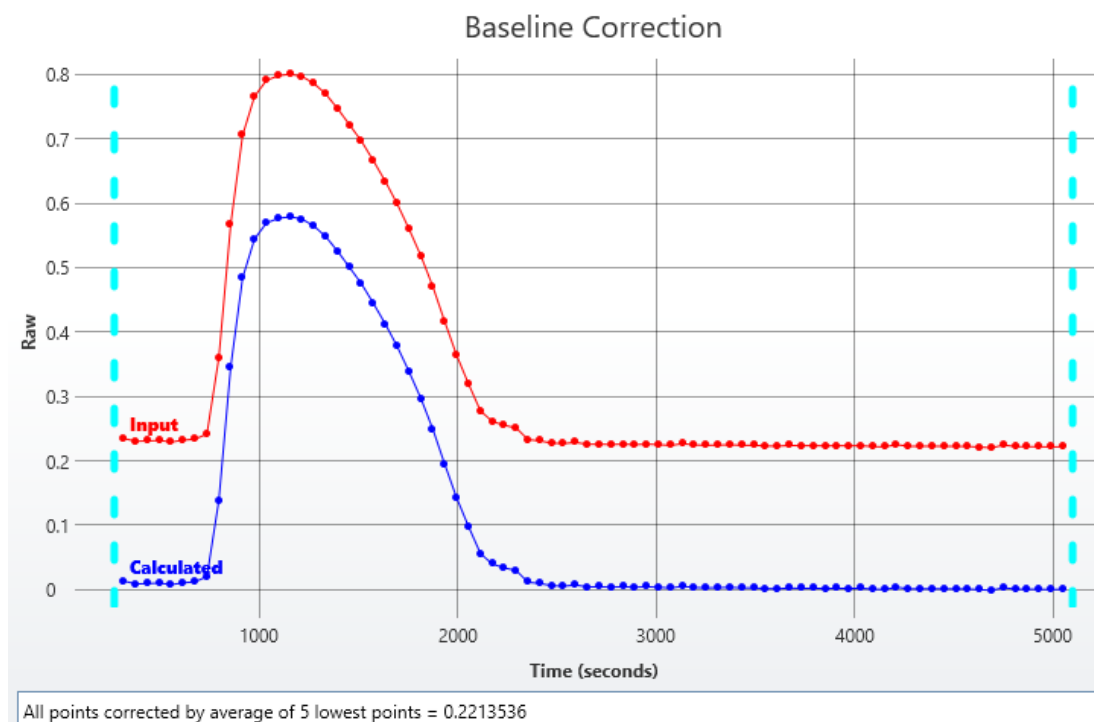
- **Sample Table**

Here is a Sample Table snippet, which shows the difference between input and output plot at the **A1** position. For each sample, the following details are reported:

Sample	Positions	Raw	Corrected
⬤Unknown1	A1		

XY Blank Correction

[Input: **XY** ➡ Output: **XY**]

This transform subtracts the average of a specified blank group (or groups) from each sample on a point-by-point basis. This is useful for subtracting background noise from measurements.

The background plot is first determined by creating an average XY plot of all replicates of the blank group. This background plot is then subtracted from each associated sample on a point-by-point basis.

Flagged positions or points are not included in the calculations. The operation is only performed on data within specified range.

Properties

Settings

> 📝 This transform requires a layout with at least 1 **Blank** group.

Blank Group	The group number of the sample to use for correction. This can be defined as a number or as an expression. The expression can refer to x which is the group number of the sample to be corrected.
	For example, set this to x to perform a 1-to-1 correction where each group is corrected by the sample of the specified blank type with the same group number.
Blank Type	The sample type used as the blank.

Example: Single Blank

Let's assume that we have some XY measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to perform a blank correction using the average of one blank group. This blank group itself is measured in duplicate. The mean of these replicates will be subtracted from all other samples.

Layout

The example uses a layout with **Standards**, **Control**, **Blank**, and **Unknowns** in duplicate (e.g., **Standard1** in **A1** and **A2**, **Unknown1** at **A3** and **A4**, **Unknown2** at **B3** and **B4**, etc.). I.e.:

	1	2	3	4	5	6	7	8	9	10	11	12
A	1	1	1	1	9	9	17	17	25	25	33	33
B	2	2	2	2	10	10	18	18	26	26	34	34
C	3	3	3	3	11	11	19	19	27	27	35	35
D	4	4	4	4	12	12	20	20	28	28	36	36
E	5	5	5	5	13	13	21	21	29	29	37	37
F	6	6	6	6	14	14	22	22	30	30	38	38
G	1	1	7	7	15	15	23	23	31	31	39	39
H	1	1	8	8	16	16	24	24	32	32	40	40

● Standard ● Blank ● Control ● Unknown

To subtract the mean of the blank positions from the measurements, specify **Blank Group** to **1** and **Blank Type** to **Blank**.

Blank Corrected

The transform will generate an output matrix named **Blank Corrected** with the value at each position being the subtraction of the average of **Blank1** sample group from the input data.

Example: 1-1 Blank Correction

Let's assume that we have some XY measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to perform a 1-to-1 correction, where each sample group is corrected by the blank that it is associated with. The blank numbering used in the layout defines the associations.

Layout

The example uses a layout with 16 **Unknowns** groups (with **Unknown1** in positions from **A1** to **A5**, **Unknown2** in positions from **B1** to **B5**, etc.) and 16 **Blank** singlets. I.e.:

○ Blank ● Unknown

Specify **Blank Group** to *x* and **Blank Type** to **Blank** to perform 1-1 correction.

With this specified, the value of each blank will be subtracted from every replicate of associated sample group. In other words, **Blank1** (stored in **A6**) value will be subtracted from **A1, A2, A3, A4, A5** (the members of **Unknown1**), and from **A6** itself.

Blank Corrected

The transform will generate an output matrix named **Blank Corrected** with the value at each position being the subtraction of the associated blank value from the input data. Therefore, calculated matrix will contain blank corrected **Unknown** values and all **Blanks** will be 0.

Advanced Example: By-Row Blank Correction

Let's assume that we have some XY measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to configure blank correction on a row-by-row basis to blank correct each sample by the average of the two blank samples of the same row.

Layout

The example uses a layout with 80 singlet **Unknowns** and 8 **Blank** duplicates for each row (with **Blank1** in positions **A1** and **A12**, **Blank2** in positions **B1** and **B12**, etc.) I.e.:

	1	2	3	4	5	6	7	8	9	10	11	12
A	1	1	9	17	25	33	41	49	57	65	73	1
B	2	2	10	18	26	34	42	50	58	66	74	2
C	3	3	11	19	27	35	43	51	59	67	75	3
D	4	4	12	20	28	36	44	52	60	68	76	4
E	5	5	13	21	29	37	45	53	61	69	77	5
F	6	6	14	22	30	38	46	54	62	70	78	6
G	7	7	15	23	31	39	47	55	63	71	79	7
H	8	8	16	24	32	40	48	56	64	72	80	8

◯ Blank ◯ Unknown

The **Blank Group** setting uses the expression:

1 + floor ((*x* -1) / (LayoutWidth()-2),1)

This converts the group number of the sample to blank correct **(*x*)** to the blank number on its row. In other words, for **x=1** this means **1+floor((1-1)/10),1)**, **floor=0** and the **Blank Group** is taken as 1. For the first row, *x* is in range from 1 to 10 and the expression outputs 1, thus for **Unknowns** from 1 to 10 the average of **Blank1** is subtracted.

For the second, *x* is from 11 to 20, **floor=1**, so the **Blank2** group is subtracted from **Unknowns11-20**, etc.

Blank Corrected

The transform will generate an output matrix named **Blank Corrected** with the value at each position being the subtraction of the average of the associated **Blank** group from the input data.

XY Expression (Dual)

[Input: **Dual XY** ➡ Output: **XY**]

This transform evaluates an expression defined in terms of *x* and *y* for each non-flagged input XY data point within the specified range. In this case *x* refers to the data for the sample on the first input matrix and *y* refers to the data for the same sample on the second input matrix.

> ℹ️ This transform is useful when it is necessary to perform a common operation on two sets of XY data. For example, this transform could be used to create a plot for each sample, that is the difference or ratio of two sets of XY measurements.

Properties

Settings

Expression	The expression to evaluate, where variables are set to the value of each position of the first (*x*) and second (*y*) input matrices. Default expression value is **x - y**. 📝 To refer to the position's corresponding data on an endpoint matrix, use a Matrix Position Reference (e.g., **#2**) to refer to the data on the second matrix.
Plot Input on Chart	When the checkbox is checked, the chart includes both the input and the calculated plot. If it is unchecked, the chart shows the calculated plot only.

Example

Let's assume that we have some XY measurement data for a 12x8 microplate stored in the **Raw** and **Corrected** matrices. In this example, we would like to calculate a new plot by summarizing input plot values in these matrices point-by-point. The calculations for the **A1** position are shown for illustration of the transform.

Here are **XY** matrices with each position containing 80 XY data points. Position data plots of the matrices are displayed in Plate Scale.
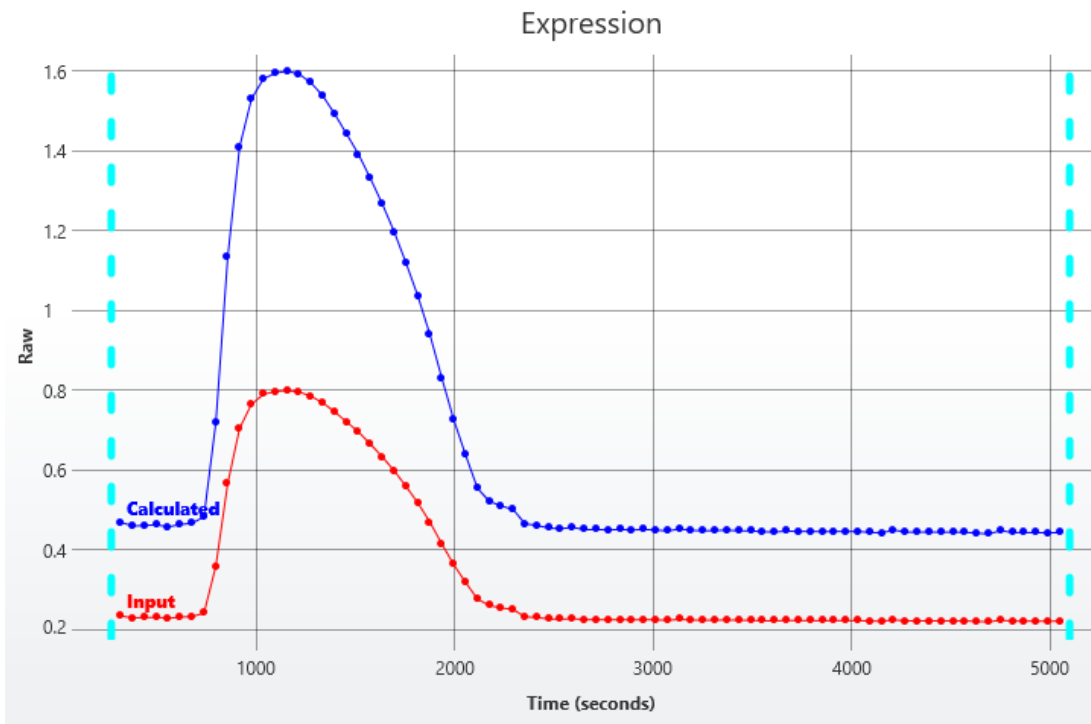
Raw

Corrected
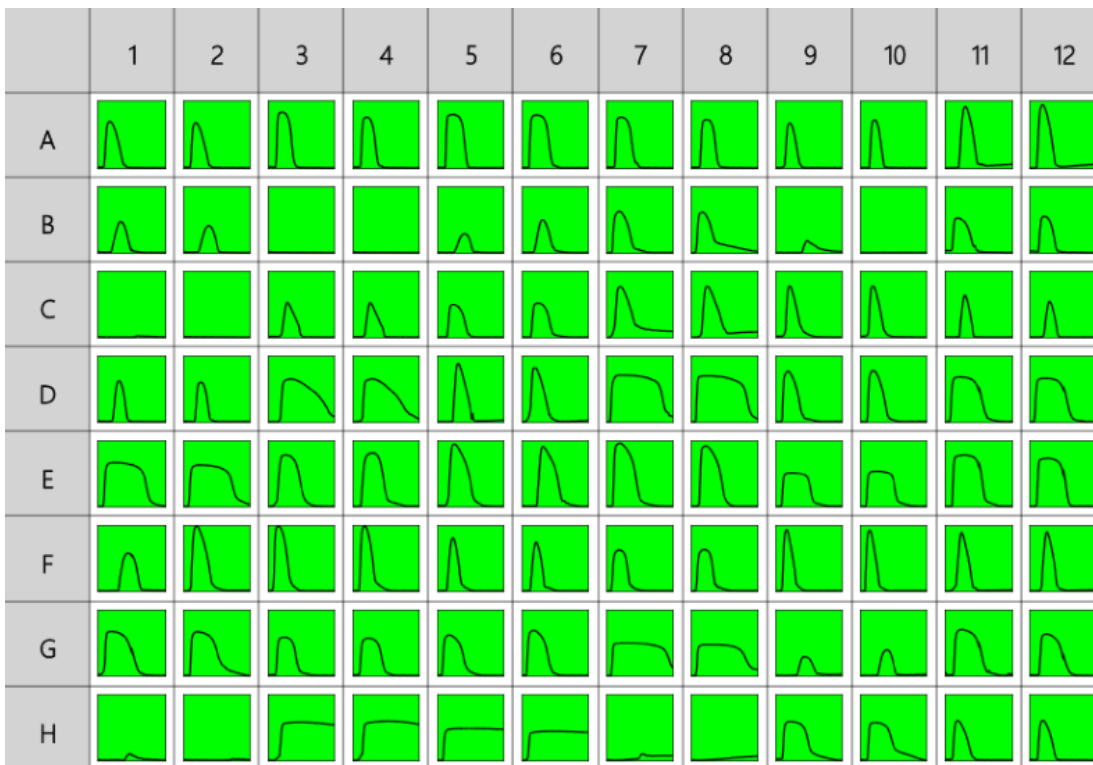
To add plot values of the input matrices and calculate a new plot for each position:

1. Specify output matrix name as **Raw+Corrected**.

2. Specify **Data**: **X Axis** as **X (Time)**, **Y Axis** as **Y**.

3. Leave **X Minimum** and **X Maximum** blank.

4. Specify **Settings**: **Expression** as *x+y*, check the **Plot Inputs on Chart** checkbox.

5. Press **Calculate**.

Raw+Corrected

The transform will generate an output XY matrix named **Raw+Corrected** where each position will contain the plot with each data point being point by point sum of the **Raw** and **Corrected** matrices plots.

With the example data provided above, the result for the **A1** plot is displayed on the Kinetic/Spectral Analysis Chart as follows:
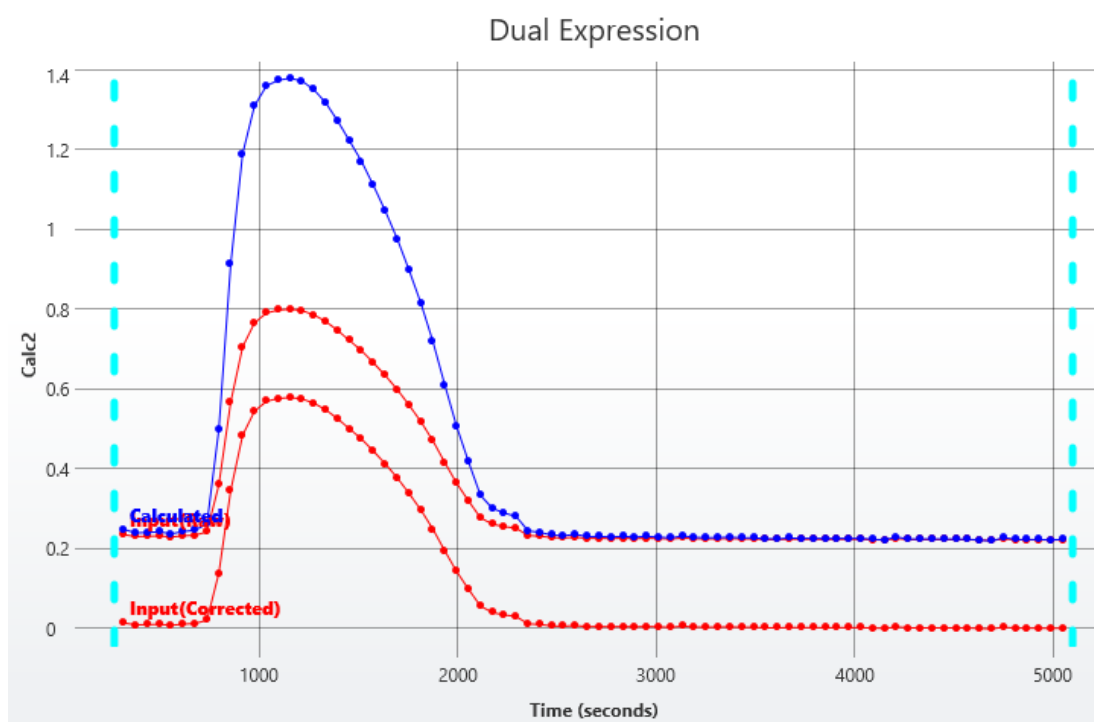
- **Sample Table**

Here is a Sample Table snippet, which shows the input plots and calculated resulting plot at the **A1** position. For each sample, the following details are reported:

Sample	Positions	Raw	Corrected	Raw+Corrected
⬤ Unknown1	A1	⌐_	⌐_	_⌐_

XY Expression

[Input: **XY** ➡ Output: **XY**]

This transform evaluates a specified expression for each input XY data point (only points within the specified range that are not flagged are processed). The resulting XY plot is generated from this expression.

> ℹ This transform is useful when it is necessary to perform an operation on all input points to calculate a new plot for each sample. For example, this transform could be used to subtract, correct, or multiply all plots by a specified value.

Properties

Settings

Expression	The expression to evaluate, where the *y* variable is set to the value of each position of the input matrix. 📝 To refer to the position's corresponding data on an endpoint matrix, use a Matrix Position Reference (e.g., **#2**) to refer to the data on the second matrix.
Plot Input on Chart	When the checkbox is checked, the chart includes both the input and the calculated plot. If it is unchecked, the chart shows the calculated plot only.

Example

Let's assume that we have some XY measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to double data points in each input plot. The calculations for the **A1** position are shown for illustration of the transform.

Raw

Here is an **XY** matrix with each position containing 80 XY data points. Position data plots of the matrix are displayed in Plate Scale.
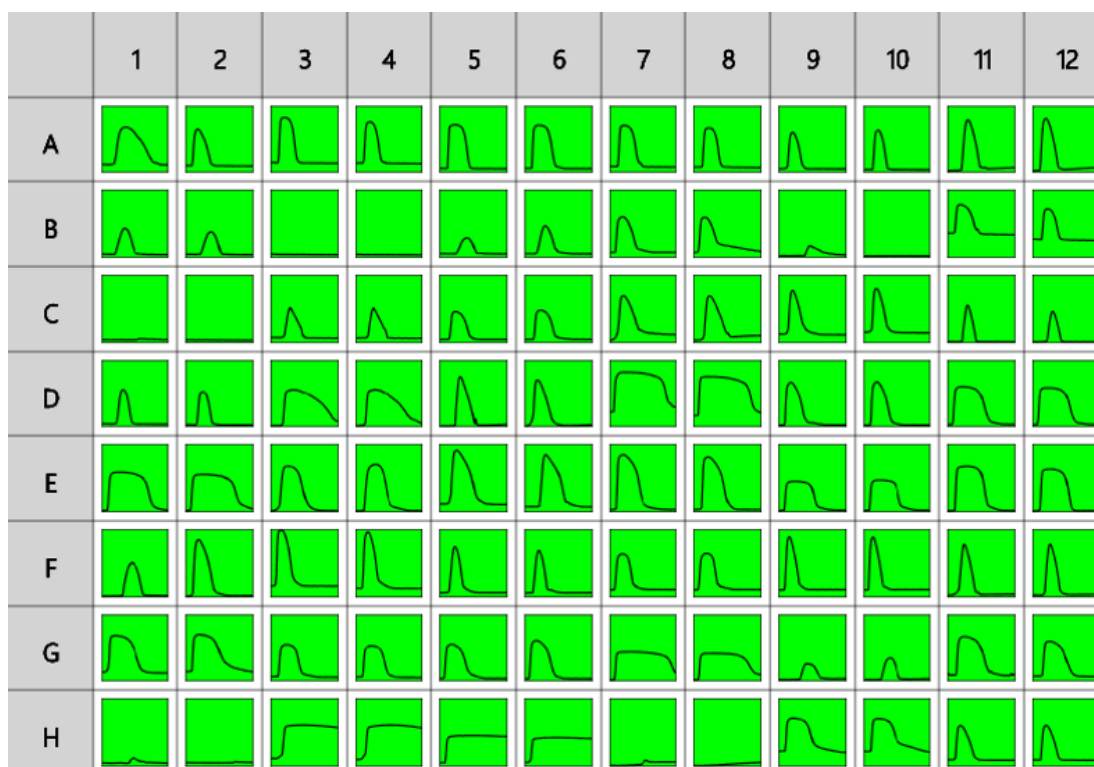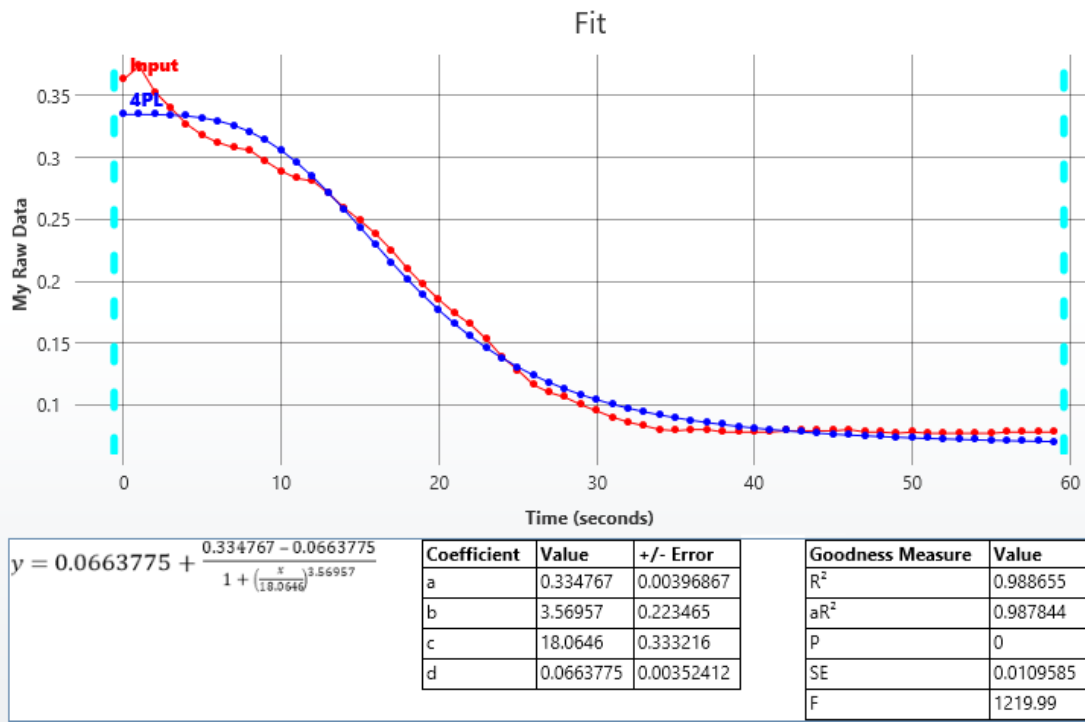
To double each data point in each plot:

1. Specify output matrix name as **Raw *2**.

2. Specify **Data**: **X Axis** as **X (Time)**, **Y Axis** as **Y**.

3. Leave **X Minimum** and **X Maximum** blank.

4. Specify **Settings**: **Expression** as *y*2*, check the **Plot Input on Chart** checkbox.

5. Press **Calculate**.

Raw *2

The transform will generate an output XY matrix named **Raw*2** where each position will contain the plot with each input point multiplied by 2.

With the example data provided above, the result for the **A1** plot is displayed on the Kinetic/Spectral Analysis Chart as follows:

Expression

* **Sample Table**

Here is a Sample Table snippet, which shows the input plot and calculated resulting plot at the **A1** position. For each sample, the following details are reported:

Sample	Positions	Raw	Raw *2
⬤ Unknown1	A1		

XY Fit

[Input: **XY** ➡ Output: **XY**]

This transform fits a curve to each XY plot and generates a new XY plot from the calculated curve.

The resulting plot is computed by calculating the *Y* value of each *X* point using the fitted curve. (Only points within the specified range that are not flagged are processed.)

> ℹ This transform can be used to smooth each XY plot using a particular curve fitting model. If you are interested in a fitting parameter of the curve fit (such as the slope of linear regression or midpoint of 4PL), then use the XY Reduction Fit Parameter transform.

Properties

Settings

Fit Method	The curve fitting method to construct the curve (from each XY plot)

Example

Let's assume that we have some XY measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to fit a 4PL curve for each input plot. The calculations for the **A1** position are shown for illustration of the transform.

Raw

Here is an **XY** matrix with each position containing 80 XY data points. Position data plots of the matrix are displayed in Plate Scale.
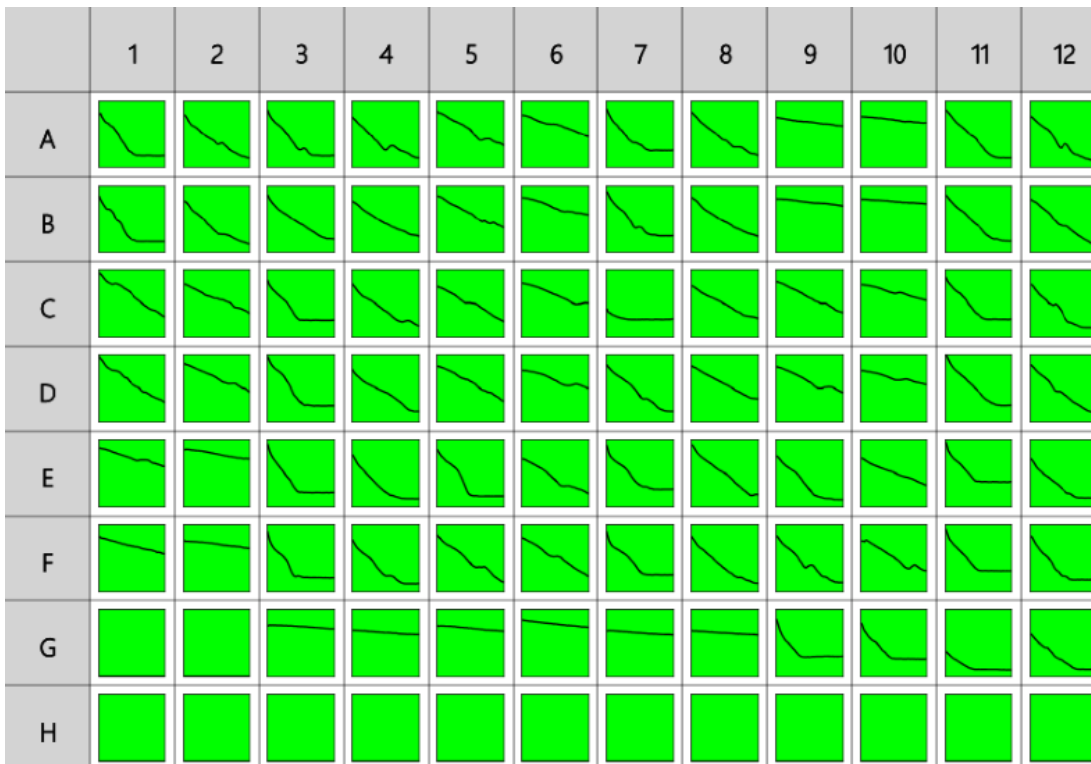
To fit a 4PL curve to each input data plot:

1. Specify output matrix name as **Smoothed**.

2. Specify **Data**: **X Axis** as **X (Time)**, **Y Axis** as **Y**.

3. Leave **X Minimum** and **X Maximum** blank.

4. Specify **Settings**: **Fit** as 4PL.

5. Press **Calculate**.

Smoothed

The transform will generate an output XY matrix named **Smoothed** where each position will contain the 4PL curve fitted to each input plot.

With the example data provided above, the result for the **A1** plot is displayed on the Kinetic/Spectral Analysis Chart as follows:

Fit

$$y = 0.0663775 + \frac{0.334767 - 0.0663775}{1 + \left(\frac{x}{18.0646}\right)^{3.56957}}$$

Coefficient	Value	+/- Error
a	0.334767	0.00396867
b	3.56957	0.223465
c	18.0646	0.333216
d	0.0663775	0.00352412

Goodness Measure	Value
R²	0.988655
aR²	0.987844
P	0
SE	0.0109585
F	1219.99

- **Sample Table**

Here is a Sample Table snippet, which shows the input plot and calculated resulting plot at the **A1** position. For each sample, the following details are reported:

Sample	Positions	My Raw Data	Smoothed
⬤ Unknown1	A1		

XY Join

[Input: **Dual XY** ➡ Output: **XY**]

This transform accepts two XY input matrices and creates new combined XY plot for each position by appending data points from the second matrix to those of the first (Any flagged data points are not included in the output matrix).

> ℹ This transform can be used to combine two separate sets of XY measurements to a single plot (for each sample) for further analysis or reporting.

> 📝 When using this transform, consider the **X Axis Data** setting. The joined data will use the data from both input matrices. To ensure that the resulting joined contents has sequential *x* values, set the **X Axis Data** setting to **Index**.

Properties

> 📝 **Advanced** and **Range** settings do not apply to this transform.

Data

> ℹ If **X** is set as **Index**, then the first index of the joined matrix is 1 plus the last used index.
> If **X** is set as **X**, then the *x* values specified in each input matrix are used (unchanged) in the output matrix.

Units

Output Units	The measurement units to be included in the header of the Sample Table.
	If the output units of the transform are the same as the input, then use "ditto" units to automatically carry these units through (i.e., apply the same units to the transform's output). This shortcut means that it is not necessary to repeat settings across transforms. Ditto units are specified with two dots, i.e.:
	..

Second Matrix X Range Correction

> 📝 This is an advanced setting that can only be configured through the **XML** editor.

CorrectSecondMatrixX	This setting defines how the x values used in the second matrix are corrected if **X Axis** is not **Index**. This can be used to ensure that

	the x values of the joined data are contiguous with a regular interval. • **None** – The results are merged on a point-by-point basis (i.e., first data point from the second matrix is appended to the first point of the first matrix, etc.) • **GapIsAverageInterval** – For each plot, the average interval of the data points in the first input matrix is computed. The first x value of the second input matrix is set to equal the x value of the last data point plus the computed average interval. The subsequent data points are first normalized by subtracting the original value of the first data point. Then, the computed x of the first data point is added to ensure a contiguous range of x values > ℹ In this case, flagged data points are considered and used in these x correction calculations (however, any flagged data points in the input matrices will also be flagged in the output).

Example

Let's assume that we have some XY measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to create a new combined plot for each position. The calculations for the **A1** position are shown for illustration of the transform.

Here are **XY** matrices with each position containing 60 XY data points. Position data plots of the matrices are displayed in Plate Scale.

Read 1
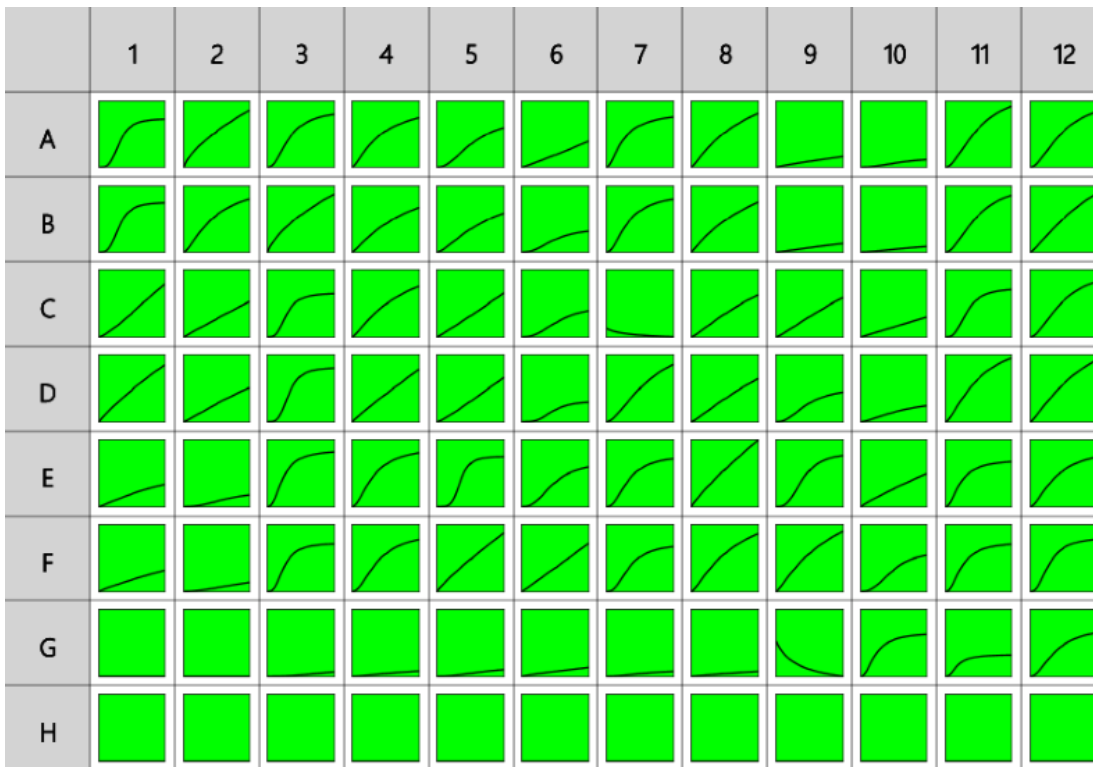
Read 2
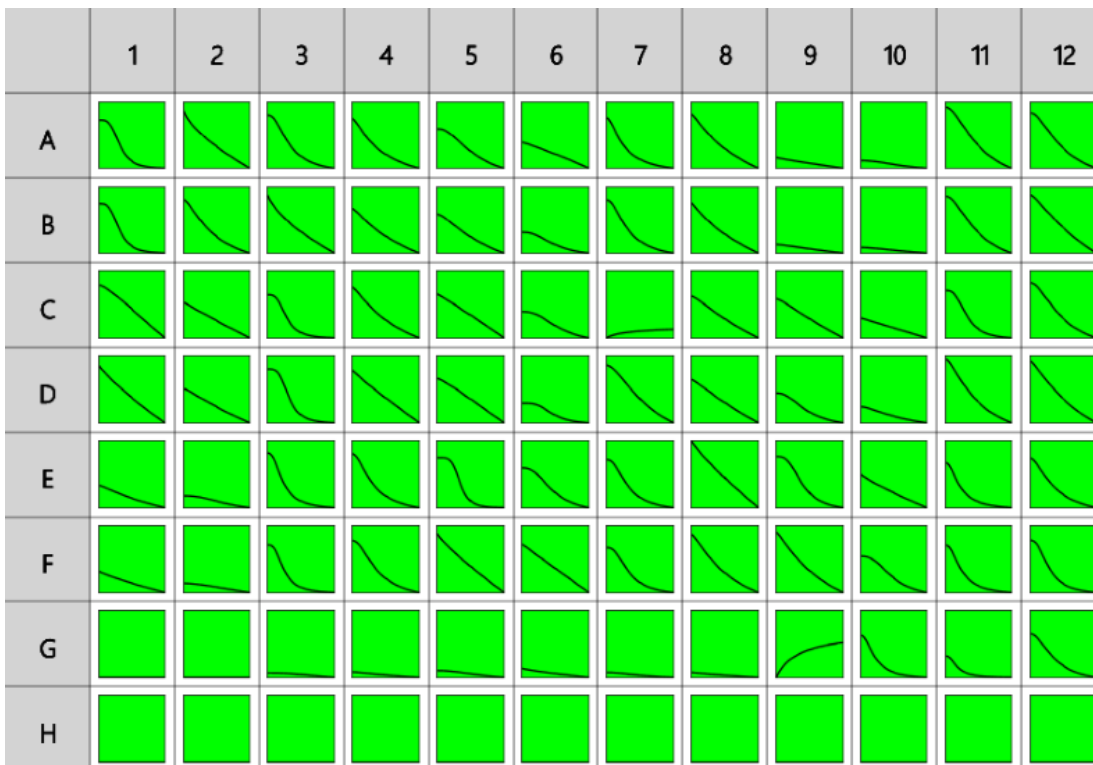
To append the data points from the second matrix to those of the first for each plot:

1. Specify output matrix name as **Joined**.

2. Specify **Data**: **X Axis** as **Index**, **Y Axis** as **Y**.

3. Press **Calculate**.

Joined
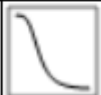
The transform will generate an output XY matrix named **Joined** where each position will contain a new XY plot which is simply the plot from the first matrix with the plot from the second matrix appended to it.

With the example data provided above, the result for the **A1** plot is displayed on the Kinetic/Spectral Analysis Chart as follows:
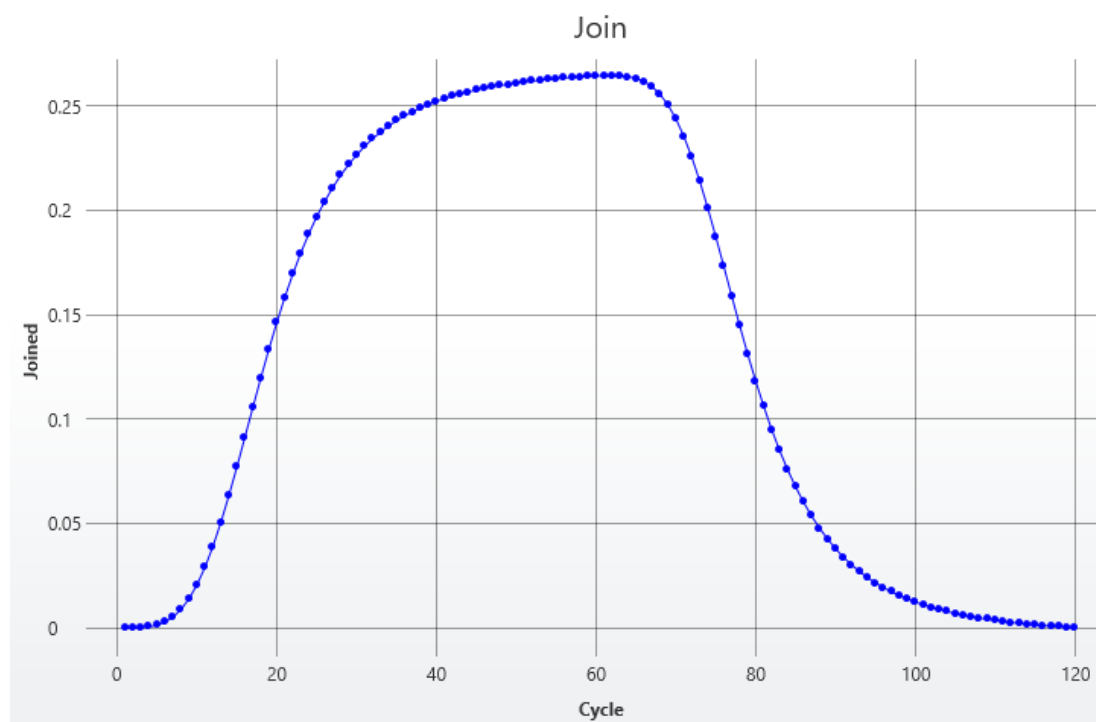
- **Sample Table**

Here is a Sample Table snippet, which shows the input plots and output appended plot at the **A1** position. For each sample, the following details are reported:

Sample	Positions	Read 1	Read 2	Joined
⬤ Unknown1	A1			

XY Outlier Removal

[Input: **XY** ➡ Output: **XY**]

This transform flags data points that are considered to be outliers. The values of the output matrix match those of the input matrix (the only difference being that any specified outlier(s) are flagged).

For each XY plot, the transform:

1. Calculates the linear regression of the specified number of adjacent points (segment)

2. Uses the calculated line to calculate *Y* from each *X* in the segment

3. Compares each calculated *Y* with the original *Y* and flags the points where the % difference is > than the specified threshold

This process is repeated across each sequential segment in the plot for adjacent point sets.

> **i** This transform can be used to automatically remove outlier points in XY data.

Properties

Settings

Num Regression Points	The number of regression points used for each linear regression segment when determining the maximum slope. The default and minimum value is 2.
Threshold	Defines the threshold % accuracy to flag outliers.
	It calculates absolute % difference between back fit value and the original *y* value. The back fit is computed from slope and intercept for each *x* value in the regression set. If this % difference is **>** the specified **Threshold**, then the data point is flagged on the output matrix.
	📝 Points that are flagged in one regression set are not included in calculations of following regression sets.

Example

Let's assume that we have some XY measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to remove up to 3 outliers from each input plot. The calculations for the **A1** position are shown for illustration of the transform.

Raw

Here is an **XY** matrix with each position containing 80 XY data points. Position data plots of the matrix are displayed in Plate Scale.
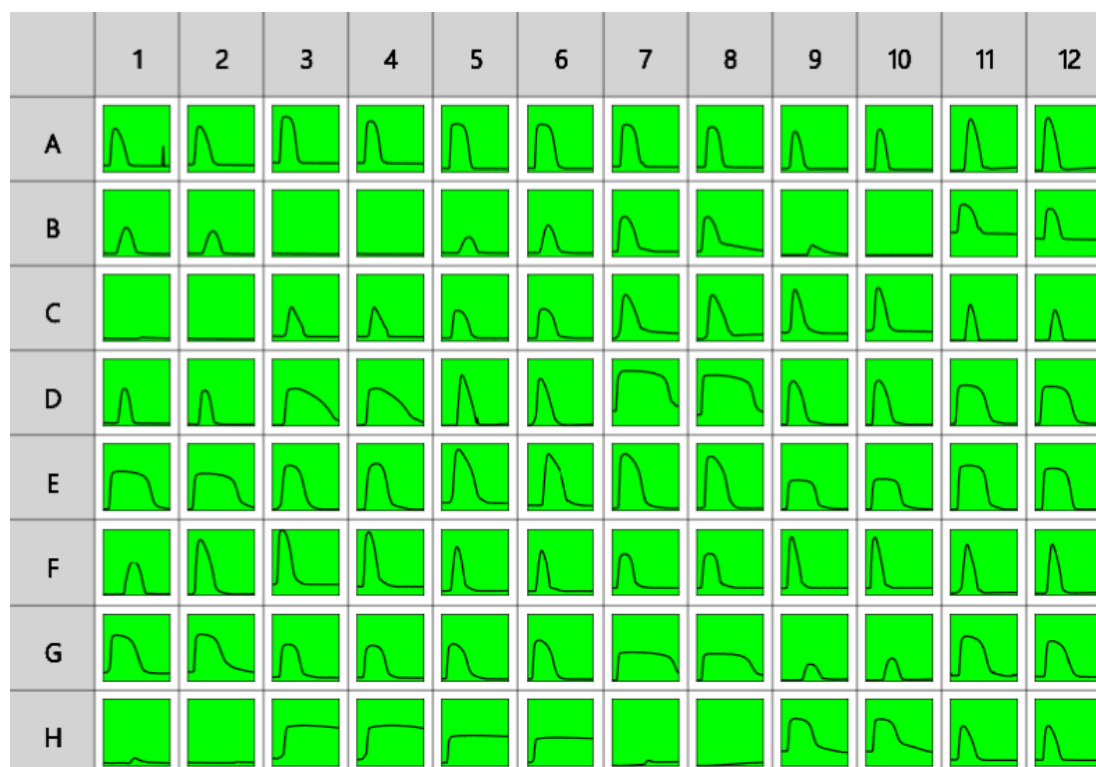

To flag outliers for each plot:

1. Specify output matrix name as **Outliers Removal**.

2. Specify **Data**: **X Axis** as **X (Time)**, **Y Axis** as **Y**.

3. Leave **X Minimum** and **X Maximum** blank.

4. Specify **Settings**: **Number of Regression Points** as **6** and the **Threshold** as **45**.

5. Press **Calculate**.

Outliers Removal

The transform will generate an output XY matrix named **Outliers Removal** where each position will contain the plot matching the input plot and outlier data points flagged.

With the example data provided above, the result for the **A1** plot is displayed on the Kinetic/Spectral Analysis Chart as follows:

Outlier Removal
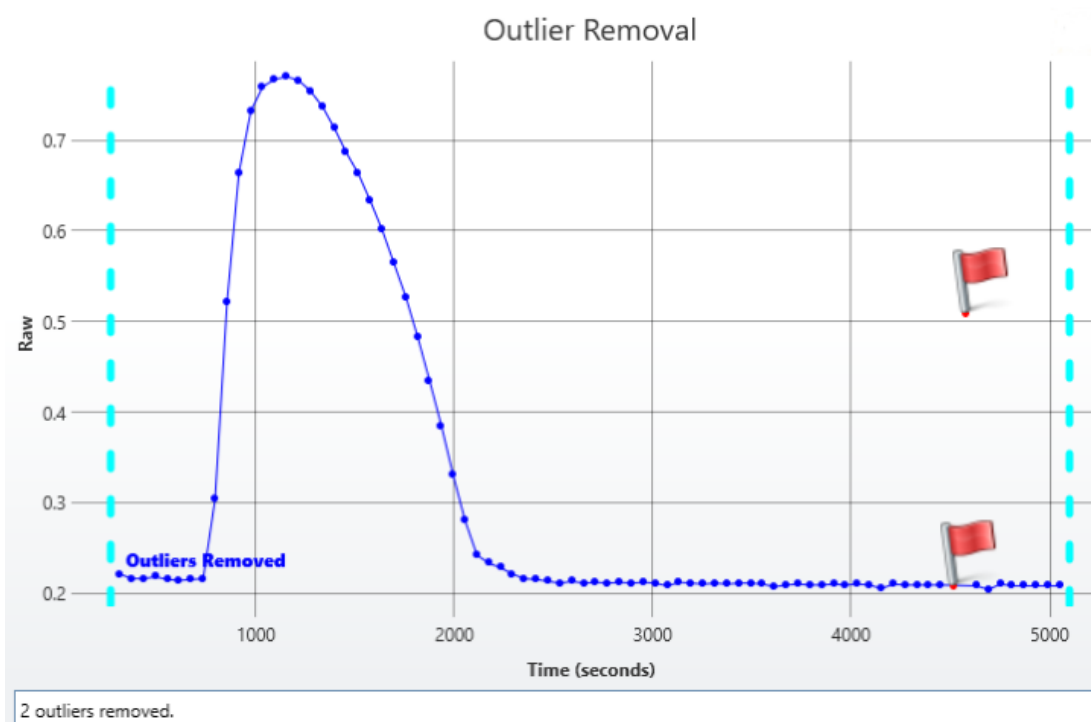
2 outliers removed.

- **Sample Table**

Here is a [Sample Table](#) snippet, which shows the input plot and calculated resulting plot at the **A1** position. For each sample, the following details are reported:

Sample	Positions	Raw	Outliers Removal
⬤ Unknown1	A1		

XY Outlier Removal (for Horizontal)

[Input: **XY** ➡ Output: **XY**]

This transform flags data points that are considered to be outliers. The values of the output matrix match those of the input matrix (the only difference being that any specified outlier(s) are flagged).

For each XY plot, the transform:

1. Calculates the **%CV** of each set of its XY points (including only the points not already flagged).

2. If the **%CV** is greater than the specified **%CV Threshold**, then the point furthest from the mean is removed.

These steps are repeated until either the new **%CV** of the data points that are not flagged is below the threshold, or until the specified number of outliers to remove has been removed. Calculations are performed on the non-flagged replicates within the specified range of each XY plot.

> ℹ This transform can be used to automatically remove outlier points in XY data.

Properties

Settings

% CV Threshold	Defines threshold percentage coefficient of variance. Data point with superior or inferior values will be flagged. The default value is 15%.
Outliers To Remove	Specifies the maximum number of outliers to remove. The minimum and default value for this setting is 1.

XY Replicates Average

[Input: **XY** ➡ Output: **XY**]

This transform calculates the average XY (kinetic/spectral) plot for each sample group. Both the *X* and *Y* values of the replicates in each group are averaged.

> If your XY data features unique *X* values for each point (such as ascending time values that are unique for each sample), you many prefer to set the **X Axis** type to **Index** so that the resulting plot is sequential with an increment of 1. Alternatively, if another **X Axis** setting is used, please note that the resulting XY plot will contain the average of the input *X* values.

Any flagged data points are not included in the analysis.

As with other XY transforms, the calculations are performed on data within the specified ranges only.

> To use this transform, select a layout with at least one sample group that contains more than one replicate.

Example

Let's assume that we have some XY measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like calculate the average plot of the replicate plots for each sample group. The calculations for the **Unknown1** group are shown for illustration of the transform.

Raw

Here is an **XY** matrix with each position containing 80 XY data points. Position data plots of the matrix are displayed in Plate Scale.

Layout

The example uses a layout with 32 **Unknowns** groups in triplicates (with **Unknown1** in positions from **A1** to **A3**, **Unknown2** in positions from **A4** to **A6**, etc.). I.e.:


To calculate averaged data plot for each sample group:

1. Specify **Data**: **X Axis** as **X (Time)**, **Y Axis** as **Y**.
2. Leave **X Minimum** and **X Maximum** blank.
3. Press **Calculate**.

Average

The transform will generate an output XY matrix named **Average** where each position will contain the averaged data plot for each sample group.

With the example data provided above, the result for the **Unknown1** averaged plot is displayed on the Kinetic/Spectral Analysis Chart as follows:
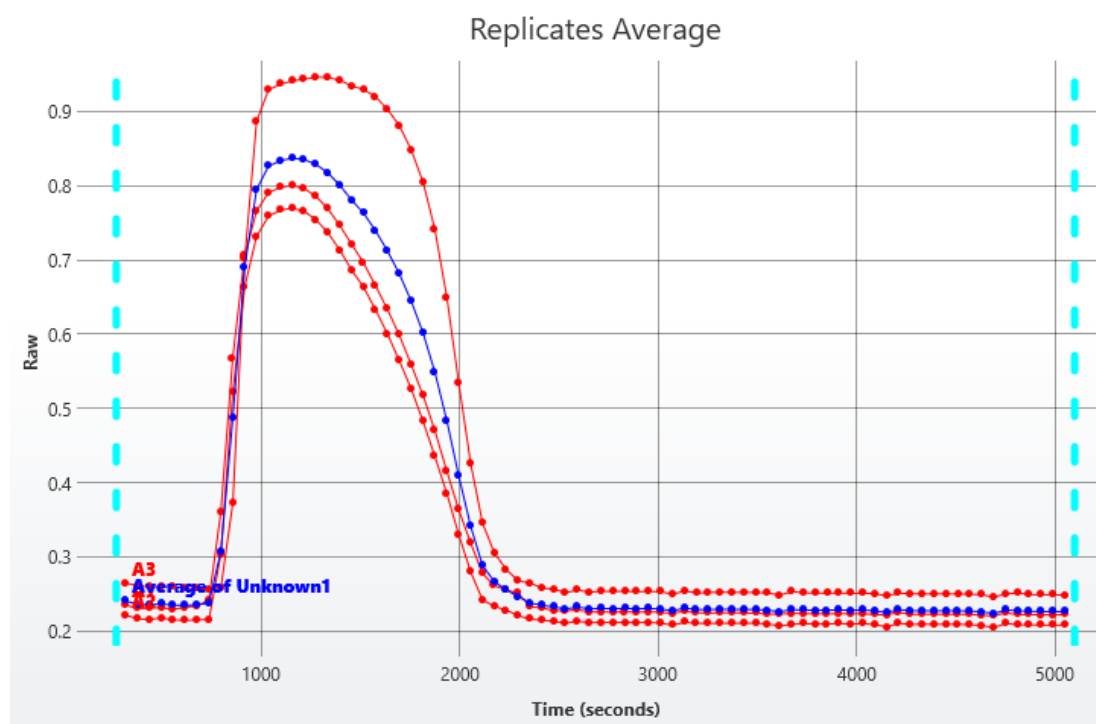
- **Sample Table**

Here is a Sample Table snippet, which shows the input plots and averaged output plot at the **Unknown1** group. For each sample, the following details are reported:

XY Replicates Function

[Input: **XY** ➡ Output: **XY**]

This transform computes a new plot for each XY (kinetic/spectral) replicate group where the *Y* values of the resulting plot are computed from the selected function applied to the corresponding replicate values of the group.

> ℹ Selecting the **Average** function will yield the same results at the XY Replicates Average transform, i.e., an average XY plot of the replicates.

The *Y* value of each point in the plot is the result of the selected function applied to each *Y* value of the point for the replicate. The *X* value of each point is the average of the *X* values of the replicates.

> 📝 If your XY data features unique *X* values for each point (such as ascending time values that are unique for each sample), you many prefer to set the **X Axis** type to **Index** so that the resulting plot is sequential with an increment of 1. Alternatively, if another **X Axis** setting is used, please note that the resulting XY plot will contain the average of the input *X* values.

Any flagged data points are not included in the analysis.

As with other XY transforms, the calculations are performed on data within the specified ranges only.

> ℹ To use this transform, select a layout with at least one sample group that contains more than one replicate.

Example

Let's assume that we have some XY measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like calculate the standard deviation of the replicate plots for each sample group. The calculations for the **Unknown1** group are shown for illustration of the transform.

Raw

Here is an **XY** matrix with each position containing 80 XY data points. Position data plots of the matrix are displayed in Plate Scale.

Layout

The example uses a layout with 32 **Unknowns** groups in triplicates (with **Unknown1** in positions from **A1** to **A3**, **Unknown2** in positions from **A4** to **A6**, etc.). I.e.:


To find the standard deviation between input replicate plots for each sample group:

1. Specify output matrix name as **Replicates SD**.

2. Specify **Data**: **X Axis** as **X (Time)**, **Y Axis** as **Y**.

3. Leave **X Minimum** and **X Maximum** blank.

4. Specify **Settings**: **Function** as **Standard Deviation**.

5. Press **Calculate**.

Replicates SD

The transform will generate an output XY matrix named **Replicates SD** where each position will contain the calculated plot with deviation between input plots of each sample group.

With the example data provided above, the result for the **Unknown1** averaged plot s displayed on the Kinetic/Spectral Analysis Chart as follows:
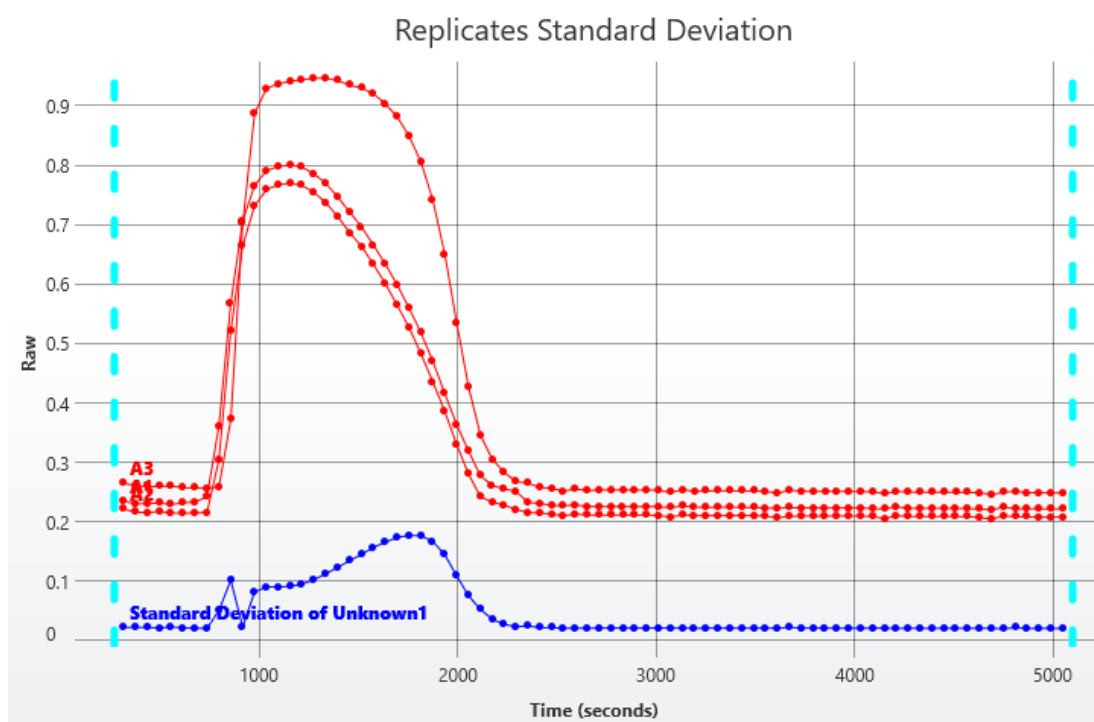
- **Sample Table**

Here is a Sample Table snippet, which shows the input plots and output plot at the **Unknown1** group. For each sample, the following details are reported:

Sample	Positions	Raw	Replicates SD
⬤ Unknown1	A1		
	A2		
	A3		

XY Reduction Transforms

XY transforms operate on matrices of data that consist of XY data and result in a new endpoint data matrix.

For example, the XY Reduction Peak transform will compute the peak value of each XY plot.

XY Data

Throughout MyAssays Desktop, we refer to measurements that yield a 2-dimensional data sets for each sample in general terms as "XY Data". This encompasses:

- Kinetic data with measurements made over time (with or without time values).

- Spectral data measured at different wavelengths.

- Scan data measured as a single line (with a distance from an origin).

XY Axes

With certain measurement types, there might be different data available to plot on the X and Y axes. For example, with kinetic data measured over time you might want to use the time values as the X axis or the cycle number instead. The axis settings specify what data should be processed by the transform. For further details you can refer to the General Transform Properties for XY Data subsection.

Flagged XY Data Points

Any data point that is part of an XY data set can be flagged. Flagged data points are excluded from the calculation.

For example, with the XY Reduction Peak transform, if the peak is set to minimum and is computed using the lowest measurement but the lowest measurement is flagged, then the peak will be computed using the next lowest data point.

In addition to each data point, each position can also be flagged to completely exclude the sample from any further analysis. This follows the same convention as for Flagged Endpoint.

XY Range

By default, XY data transform calculations are performed on all XY data points that are not flagged. However, it is also possible to define a range of points to process. This is useful if the area of interest is only a subset of the measurements. The range minimum and maximum values can be set as an absolute value or as an expression (that can in turn refer to other computed results).

For example, the XY Reduction Average transform could be configured to work with input XY data that has 100 data points and to use only points >=20 and <= 80. In this case, the res-

ulting output endpoint matrix would contain only mean value from this subset at each position.

Refer to the General Transform Properties for XY Data subsection for more details.

XY Reduction

Transform	Reduces Data To
Average	The average of non-flagged data points within a subset at each position.
Fit Parameter	The specified parameter for non-flagged data points within specified range at each position.
Maximum Slope	Determined maximum slope between two adjacent points within specified range at each position.
Peak	Determined peak (minimum or maximum) data point within the specified range of the non-flagged points at each position.
Total	The sum of non-flagged data points within specified range at each position.
X At Y	The X value at which the specified Y value crosses the evaluated plot of non-flagged points within the specified range at each position.
X Between Y	The difference between the first two X points that equal or cross specified Y value at each position.
Y At X	Determined Y value for a specified X value within the specified range of the non-flagged points at each position.

XY Reduction Average

[Input: **XY data** ➡ Output: **Endpoint**]

This reduction transform computes the average *Y* value of each XY plot (within the specified range of points to consider). Flagged positions or points are not included in the calculations.

Example

Let's assume that we have some **XY** measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to reduce data in each position to average of its data points and store resulting value to corresponding position of an endpoint matrix.

Raw

Here is an **XY** matrix with each position containing 10 XY data points. Position data plots of the matrix are displayed in Plate Scale.

Here the reduction of data in the **A1** position is shown for illustration of the transform. The raw data is displayed here to 3 decimal places:

A1 0.000;0.555 0.100;0.988 0.200;1.613 0.300;1.938 0.400;2.608 0.500;2.889 0.600;3.773 0.700;3.816 0.800;4.898 0.900;5.491

To reduce **XY** data to an average endpoint value:

1. Specify **Data**: **X Axis** as **X (Time)**, **Y Axis** as **Y**.

2. Leave **X Minimum** and **X Maximum** blank.

3. Press **Calculate**.

Average

The transform will generate an endpoint matrix named **Average** with the value at each position being the averaged value of the *Y* input data points. With the example data provided above, the resulting averaged value for the **A1** plot is displayed on the Kinetic/Spectral Analysis Chart as follows (also displayed here to 3 decimal places):

2.857
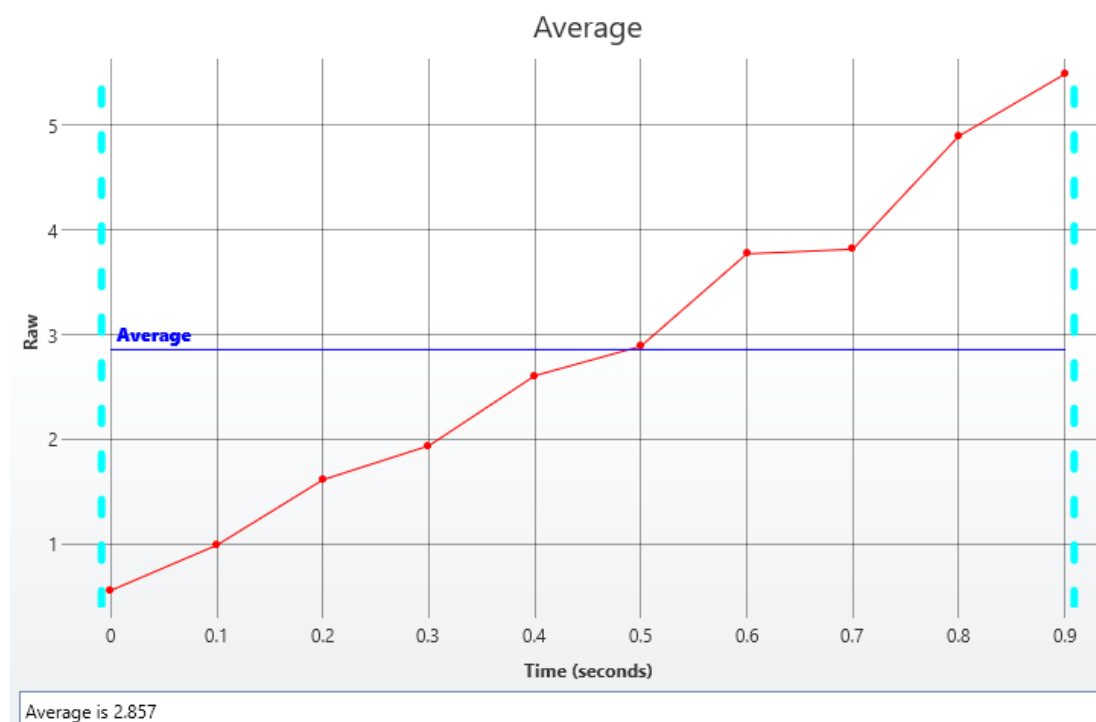
XY Reduction Fit Parameter

[Input: **XY data** ➡ Output: **Endpoint**]

This reduction transform performs the selected curve fit on each XY plot and sets the result to the value of a specified fit parameter. Points outside the specified range and flagged positions/points are excluded from calculations.

> ℹ This transform can be used to obtain the specific fitting parameter of a curve fit to each plot (such as the slope of linear regression or midpoint of 4PL).

Properties

Settings

Fit Method	The curve fitting method to construct a curve for your data
Parameter	Represents the Fit Transform Context Parameter to fit data points by

Example

Let's assume that we have some kinetic/spectral measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to fit the Linear Regression and calculate the R^2 parameter for each position plot.

Raw

Here is an **XY** matrix with each position containing 10 XY data points. Position data plots of the matrix are displayed in Plate Scale.

Here the reduction of data in the **A1** position is shown for illustration of the transform. The raw data is displayed here to 3 decimal places:

A1 0.000;0.555 0.100;0.988 0.200;1.613 0.300;1.938 0.400;2.608 0.500;2.889 0.600;3.773 0.700;3.816 0.800;4.898 0.900;5.491

To reduce the plot at each position to its R^2 parameter:

1. Specify **Data**: **X Axis** as **X (Time)**, **Y Axis** as **Y**.

2. Leave **X Minimum** and **X Maximum** blank.

3. Select Linear Regression **Fit Method** and specify **Parameter** as R2 (with the syntax, provided in Fit Transform Context Parameters).

4. Press **Calculate**.

Calculated

The transform will generate an output endpoint matrix named **Calculated** with the value at each position R^2 measure for each input plot. With the example data provided above, the resulting averaged value for the **A1** plot is displayed on the Kinetic/Spectral Analysis Chart as follows (also displayed here to 3 decimal places):

0.987

Fit Parameter
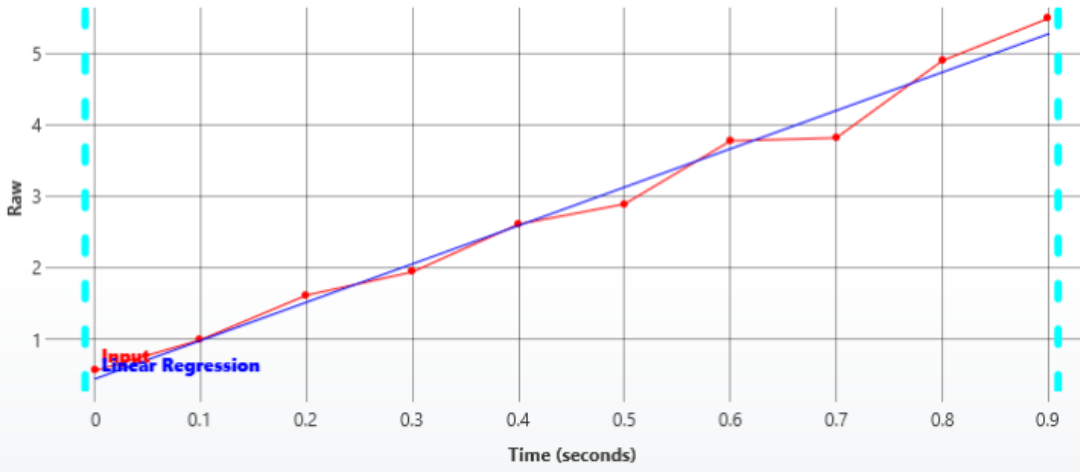
$y = 5.36915x + 0.440824$

Coefficient	Value	+/- Error
m	5.36915	0.221636
c	0.440824	0.118321

Goodness Measure	Value
R^2	0.986551
aR^2	0.98487
P	0.00000000899332
SE	0.201311
F	586.856

XY Reduction Maximum Slope

[Input: **XY data** ➡ Output: **Endpoint**]

This reduction transform finds the maximum slope of each XY plot. The result can be set to the slope, or to the *X* or *Y* value at the maximum slope (absolute or relative to the start *X*).

> ⓘ This transform can be used in kinetic applications to calculate the time to reach a maximum rate or to find the maximum rate in a reaction.

The maximum slope is determined by computing the linear regressions of adjacent points (using the specified number of linear regressions points) and determining the slope with the maximum (upward or downward) value.

Where the result is set to be the *X* or *Y* value of the maximum slope:

- the *X* value is calculated as the centre of the points with the maximum slope note this may not exactly equal an *X* data point).

- the *Y* value is computed from the linear regression of the calculated center *X*.

Flagged positions or points or those outside specified ranges are not included in the calculations.

Properties

Settings

Max is	Determines the type of slope to be found by this transform. The settings are: • **Auto** – The maximum slope is determined from the general direction of each plot by using the linear regression. If the slope of the plot is positive, the **Fastest Increase** is used; if negative, the **Fastest Decrease** is used. • **Fastest Increase** – The maximum slope is the slope with the highest positive value. • **Fastest decrease** – The maximum slope is the slope with the greatest negative value.
Num Regression Points	The number of regression points used for each linear regression segment when determining the maximum slope. The default and minimum value is 2.
Result is	Specifies the result of the analysis. The settings are:

	• **Slope** – The result is the slope of the determined maximum slope. • **X At Centre Absolute** – The result is the *X* at the center of the determined maximum slope. (The center is the midpoint of the data points with the maximum linear regression slope). • **Y At Centre Absolute** – The result is the *Y* at the center of the determined maximum slope. (The center is the *Y* value computed from the *X* midpoint of the data points with the maximum linear regression slope). • **X At Centre Relative** – The result is the *X* at the center of the determined maximum slope relative to the first non-flagged data point that is greater than **X Minimum.** (The center is the midpoint of the data points with the maximum linear regression slope). • **Y At Centre Relative** – The result is the *Y* at the center of the determined maximum slope relative to the first non-flagged data point that is greater than **X Minimum**.(The center is the *Y* value computed from the *X* midpoint of the data points with the maximum linear regression slope). • **Extrapolated Y** – A value of *Y* is calculated from the determined maximum slope using the *X* value specified with **X For Extrapolation**.
X for Extra-polation	When **Extrapolated Y** option in the **Result Is** setting is chosen, then this can be used to specify an expression used to determine the *X* value to calculate *Y* for.

Example

Let's assume that we have some **XY** measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to reduce data for each replicate to maximum slope between its data points and store resulting value to corresponding position of an end-point matrix.

Raw

Here is an **XY** matrix with each position containing 10 XY data points. Position data plots of the matrix are displayed in Plate Scale.

Here the reduction of data in the **A1** position is shown for illustration of the transform. The raw data is displayed here to 3 decimal places:

A1 0.000;0.555 0.100;0.988 0.200;1.613 0.300;1.938 0.400;2.608 0.500;2.889 0.600;3.773 0.700;3.816 0.800;4.898 0.900;5.491

To reduce **XY** plot at a position to a maximum slope to an endpoint value:

1. Specify **Data**: **X Axis** as **X (Time)**, **Y Axis** as **Y**.

2. Leave **X Minimum** and **X Maximum** blank.

3. Define **Max is** as **Auto**, **Num Regression Points** as 2, and **Result is** as **Slope**.

4. Press **Calculate**.

Maximum Slope

The transform generates an endpoint matrix named **Maximum Slope** with the value at each position being the maximum slope between points at each kinetic/spectral plot. With the example data provided above, the result for the **A1** plot is displayed on the Kinetic/Spectral Analysis Chart as follows (also displayed here to 3 decimal places):

10.815

Maximum Slope

Maximum slope (10.8147705928176)

Maximum slope is 10.815
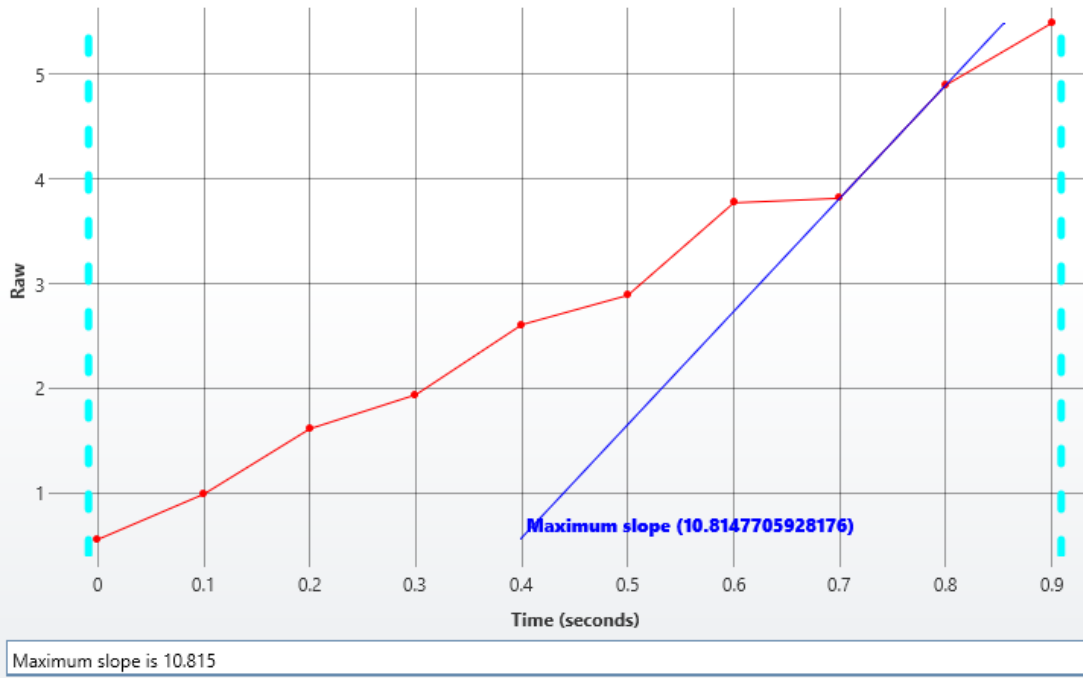
XY Reduction Peak

[Input: **XY data** ➡ Output: **Endpoint**]

This reduction transform computes the peak of all values for each XY plot (within the specified range of points to consider). Flagged positions or points are not included in the calculations.

The result can be set to the *X* or *Y* value at the peak, with the peak being defined as either the minimum or maximum value.

Properties

Settings

Peak	This setting determines which data point is considered as the peak: • **Max** – The point with the maximum *Y* value • **Min** – The point with the minimum *Y* value
Result	This setting determines which axis's coordinate is stored in resulting matrix: • **X** – Projection of **Peak** value on X axis • **Y** – Projection of **Peak** value on Y axis

Example

Let's assume that we have some kinetic/spectral measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to reduce each input plot to the value at its peak point (maximum).

Raw

Here is an **XY** matrix with each position containing 10 XY data points. Position data plots of the matrix are displayed in Plate Scale.

Here the reduction of data in the **A1** position is shown for illustration of the transform. The raw data is displayed here to 3 decimal places:

A1 300.000;0.002 350.000;0.021 400.000;0.106 450.000;0.279 500.000;0.384 550.000;0.279 600.000;0.106 650.000;0.021 700.000;0.002

To reduce kinetic/spectral plot in each position to its maximum endpoint value:

1. Specify **Data**: **X Axis** as **X (Time)**, **Y Axis** as **Y**.

2. Leave **X Minimum** and **X Maximum** blank.

3. Define **Peak** as **Max** and **Result** as **X**.

4. Press **Calculate**.

Peak

The transform will generate an output endpoint matrix named **Peak** with the value at each position being the maximum Y point of each input replicate plot. With the example data provided above, the result for the **A1** plot is displayed on the [Kinetic/Spectral Analysis Chart](#) as follows (also displayed here to 3 decimal places):

500.000

Peak (Max)

Peak (500,0.384)

Maximum point is (500, 0.384), result is 500.000
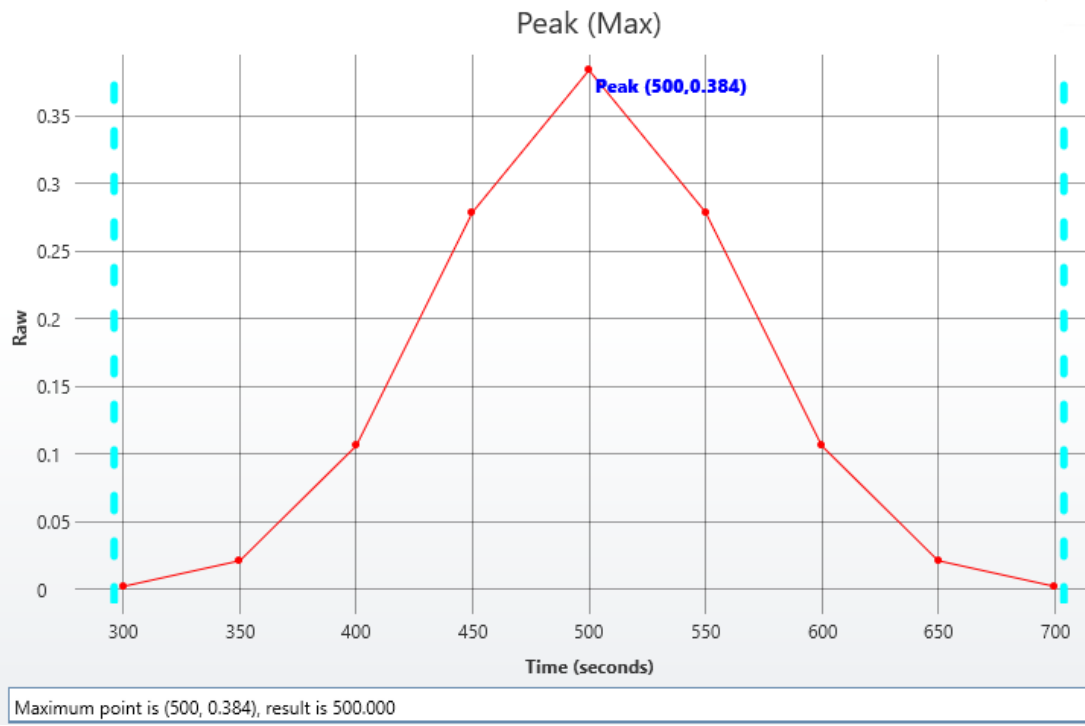
XY Reduction Total

[Input: **XY data** ⇨ Output: **Endpoint**]

This reduction transform computes the sum of all *Y* values for each XY plot (within the specified range of points to consider). Flagged positions or points are not included in the calculations.

Example

Let's assume that we have some kinetic/spectral measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to reduce data in each position to sum of its data points and store the resulting value to the corresponding position of an endpoint matrix.

Raw

Here is an **XY** matrix with each position containing 10 XY data points. Position data plots of the matrix are displayed in Plate Scale.

Here the reduction of data in the **A1** position is shown for illustration of the transform. The raw data is displayed here to 3 decimal places:

A1 0.000;0.555 0.100;0.988 0.200;1.613 0.300;1.938 0.400;2.608 0.500;2.889 0.600;3.773 0.700;3.816 0.800;4.898 0.900;5.491

To reduce **XY** data to a sum of its data points:

1. Specify **Data**: **X Axis** as **X (Time)**, **Y Axis** as **Y**.

2. Leave **X Minimum** and **X Maximum** blank.

3. Press **Calculate**.

Total

The transform will generate an endpoint matrix named **Total** with the value at each position being the sum of *Y* input data point values. With the example data provided above, the result for the **A1** plot is displayed on the Kinetic/Spectral Analysis Chart as follows (also displayed here to 3 decimal places):

28.569
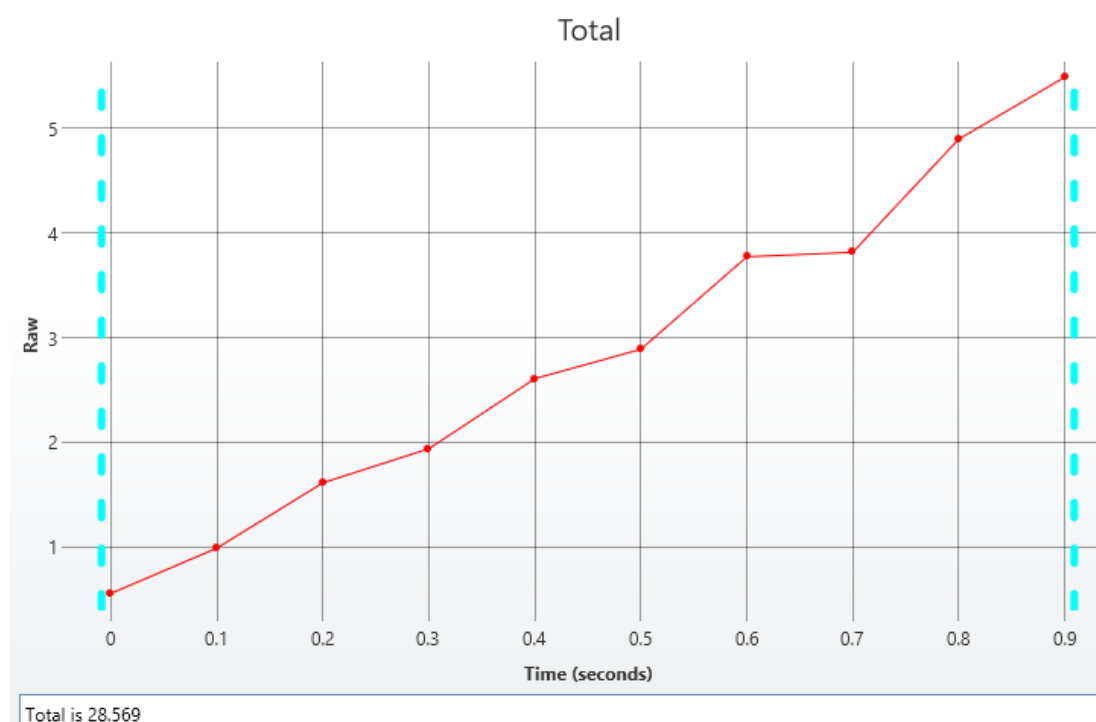
XY Reduction X At Y

[Input: **XY data** ⇨ Output: **Endpoint**]

This reduction transform finds the *X* value at which the specified *Y* value (**Cursor**) crosses the plot (within the specified range). If the *Y* position is in between two adjacent data points, then an interpolated value is calculated from the bounding points.

X and *Y* values can be specified as a relative to the first point or as an absolute value. Flagged positions and points are not included in calculations.

> ⓘ This transform can be used to determine the *X* value at the point where the horizontal (defined by *Y*) value crosses each plot.

Properties

Settings

Cursor	The expression evaluated for each position that represents the target in *Y* value. The default value is 1. 📝 To refer to the position's corresponding data on an endpoint matrix, use a Matrix Position Reference (e.g., **#2**) to refer to the data on the second matrix.
Cursor Type	This defines how the **Cursor** is treated for each position. The settings are: • **Absolute** – The value specified by **Cursor** is the value to use in the calculation of *X*. • **Relative From First Measured** – The value of **Cursor** is added to the *Y* value of the first point for the position (even if the first point is ~~Flagged~~ or outside the range). • **Relative From First In Range** – The value of **Cursor** is added to the *Y* value of the first non-flagged data point within the range.
Result is	Specifies the result of the analysis. The settings are: • **Absolute** – The computed *X* value. • **Relative From First Measured** – The computed *X* value minus the *X* value of the first point for the position (even if the first point is ~~Flagged~~ or out of range). • **Relative From First In Range** – The computed *X* value minus the *X* value

		of the first non-flagged data point within the range.

Example

Let's assume that we have some kinetic/spectral measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to reduce data in each position to the value of *X* at which the specified *Y* value crosses the plot.

Raw

Here is an **XY** matrix with each position containing 10 XY data points. Position data plots of the matrix are displayed in Plate Scale.

Here the reduction of data in the **A1** position is shown for illustration of the transform. The raw data is displayed here to 3 decimal places:

A1 0.000;0.555 0.100;0.988 0.200;1.613 0.300;1.938 0.400;2.608 0.500;2.889 0.600;3.773 0.700;3.816 0.800;4.898 0.900;5.491

To reduce the plot at each position to the value of *X* when crossing/equaling specified **Cursor**:

1. Specify **Data**: **X Axis** as **X (Time)**, **Y Axis** as **Y**.

2. Leave **X Minimum** and **X Maximum** blank.

3. Specify **Settings**: **Cursor** as 1, **Cursor Type** and **Result is** as **Absolute**.

4. Press **Calculate**.

X

The transform will generate an endpoint matrix named **X** with the value at each position being kinetic/spectral plot reduced to resulting numeric value (as defined in specified settings). With the example data provided above, the result for the **A1** plot is displayed on the Kinetic/Spectral Analysis Chart as follows (also displayed here to 3 decimal places):

0.102
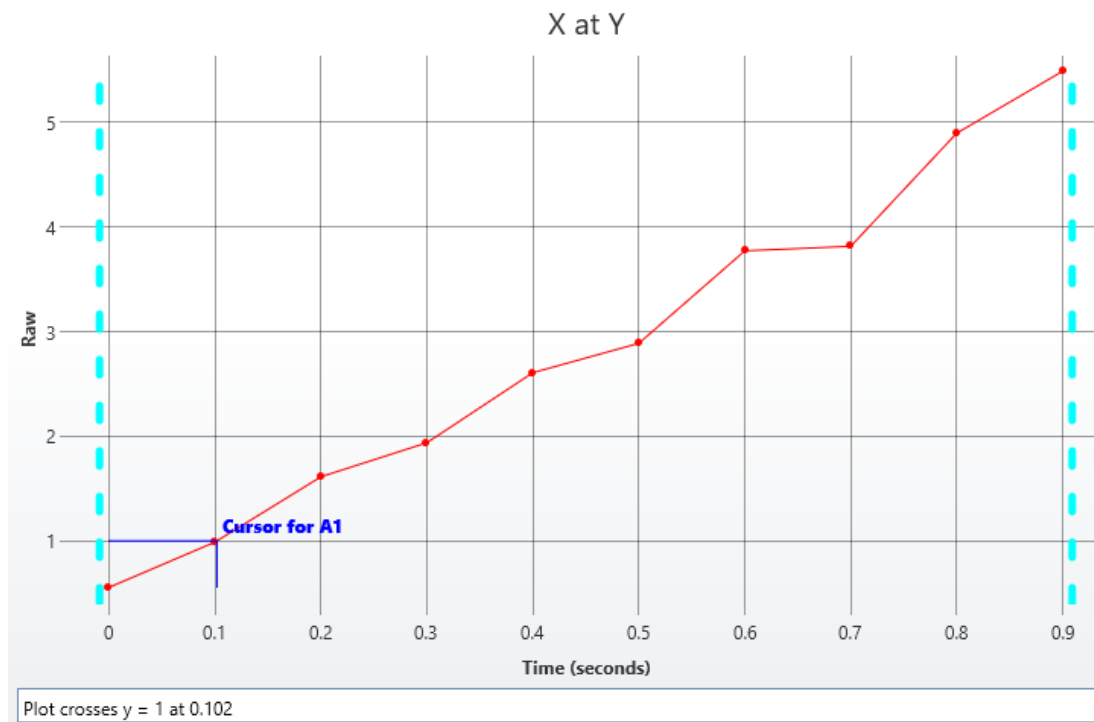
XY Reduction X Between Y

[Input: **XY data** ➡ Output: **Endpoint**]

This reduction transform computes the difference between the first two *X* points at which the specified *Y* value (**Cursor**) crosses or equals the plot.

Y value can be specified as relative to the first point or as an absolute value. Flagged positions and points outside the specified range are not included in calculations.

Properties

Settings

Cursor	The expression evaluated for each position that represents the target in *Y* value. The default value is 1. 📝 To refer to the position's corresponding data on an endpoint matrix, use a Matrix Position Reference (e.g., **#2**) to refer to the data on the second matrix.
Cursor Type	This defines how the **Cursor** is treated for each position. The settings are: • **Absolute** – The value specified by **Cursor** is the value to use in the calculation of *X*. • **Relative From First Measured** – The value of **Cursor** is added to the *Y* value of the first point for the position (even if the first point is ~~Flagged~~ or outside the range). • **Relative From First In Range** – The value of **Cursor** is added to the *Y* value of the first non-flagged data point within the range.

📝 If the plot is not crossed or only crossed once, the result will be flagged.

Example

Let's assume that we have some XY measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to calculate the difference between the first two *X* points that equal or cross the specified **Cursor** (*Y*) and then reduce data to the difference between these *X* points in each position.

Raw

Here is an **XY** matrix with each position containing 40 XY data points. Position data plots of the matrix are displayed in Plate Scale.

Raw data kindly provided by Andreas Engel Krag, Aarhus Universitetshospital.

The reduction of data in the **A1** position is shown for illustration of the transform. The raw data is displayed here to 3 decimal places:

A1 317.780;0.235 377.710;0.230 437.630;0.231 497.530;0.232 557.440;0.229 617.360;0.232 677.280;0.234 737.180;0.242 797.080;0.360 856.980;0.567 916.900;0.705 976.840;0.766 1036.760;0.791 1096.680;0.798 1156.580;0.800 1216.480;0.797 1276.410;0.786 1336.330;0.769 1396.230;0.746 1456.150;0.721 1516.050;0.696 1575.980;0.666 1635.900;0.634 1695.850;0.599 1755.750;0.560 1815.670;0.518 1875.590;0.470 1935.510;0.416 1995.410;0.364 2055.320;0.319 2115.220;0.277 2175.120;0.261 2235.020;0.255 2294.970;0.251 2354.890;0.233 2414.790;0.231 2474.690;0.228 2534.620;0.227 2594.520;0.229 2654.420;0.226

To reduce the plot at each position to the difference between the first and the second *X* value when crossing/equaling specified **Cursor**:

1. Specify **Data**: **X Axis** as **X (Time)**, **Y Axis** as **Y**.

2. Leave **X Minimum** and **X Maximum** blank.

3. Specify **Settings**: **Cursor** as 0.7, **Cursor Type** as **Absolute**.

4. Press **Calculate**.

Difference

The transform will generate an endpoint matrix named **Difference** with the value at each position being the XY plot reduced to resulting numeric value (as defined in specified settings). With the example data provided above, the result for the **A1** plot is as displayed on the Kinetic/Spectral Analysis Chart follows (also displayed here to 3 decimal places):

591.737

Plot crosses y = 0.7 at X1 = 914.729 and X2 = 1506.466, difference = 591.737
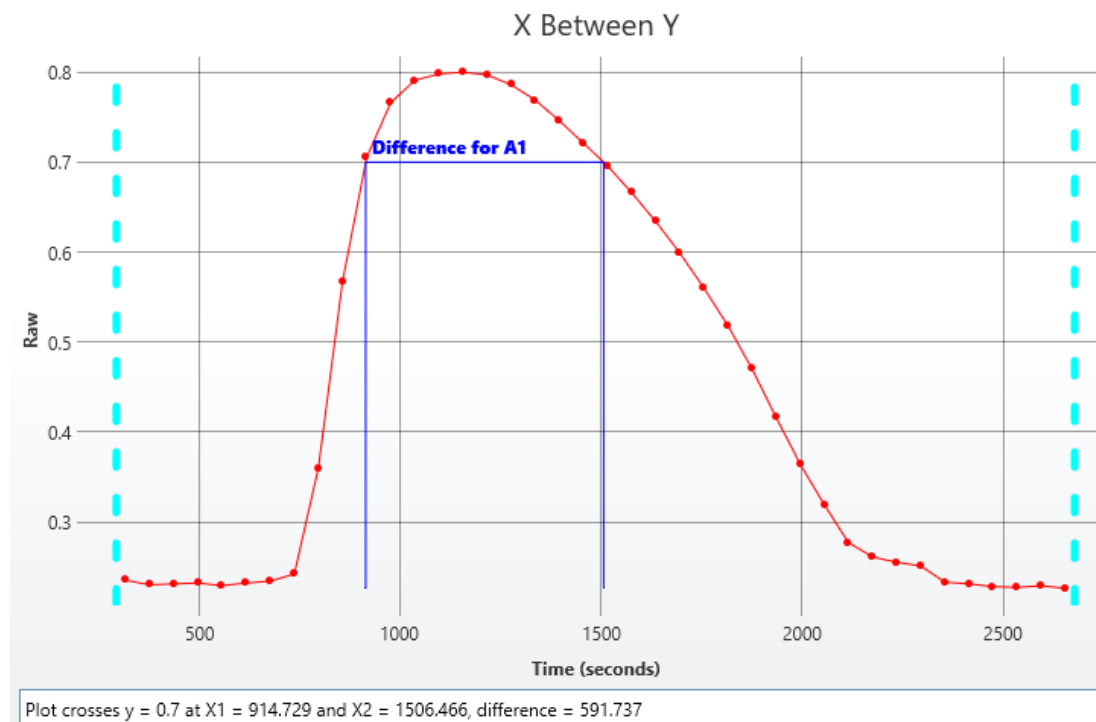
XY Reduction Y At X

[Input: **XY data** ➡ Output: **Endpoint**]

This reduction transform finds the *Y* value at which the specified *X* value (**Cursor**) crosses the plot (within the specified range). If the *X* position is in between two adjacent data points, then an interpolated value is calculated from the bounding points.

The *X* value can be specified as a relative to the first point or as an absolute value. Flagged positions and points are not included in calculations.

> ℹ This transform can be used to determine the *Y* value at the point where the vertical (defined by *X*) value crosses each plot (or more simply the *Y* at a given *X*).

Properties

Settings

Cursor	The expression evaluated for *X* value of each position. 📝 To refer to the position's corresponding data on an endpoint matrix, use a Matrix Position Reference (e.g., **#2**) to refer to the data on the second matrix.
Cursor Type	This defines how the **Cursor** is treated for each position. These settings are used to handle scenarios where *X* values for each position may be out of sync by adding an offset to the cursor. The settings are: • **Absolute** – The value specified by the **Cursor** is the value to use in the calculation of *Y*. • **Relative From First Measured** – The **Cursor** value is added to the *X* value of the first point for the position (even if the first point is ~~Flagged~~ or out of range). • **Relative From First In Range** – The **Cursor** value is added to the *X* value of the first non-flagged data point within the range

Example

Let's assume that we have some kinetic/spectral measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example, we would like to reduce data in each position to the *Y* value at which the specified **Cursor** (*X*) crosses the plot. The resulting value is then stored to corresponding position of an endpoint matrix.

Raw

Here is an **XY** matrix with each position containing 10 XY data points. Position data plots of the matrix are displayed in Plate Scale.

Here the reduction of data in the **A1** position is shown for illustration of the transform. The raw data is displayed here to 3 decimal places:

A1 0.000;0.555 0.100;0.988 0.200;1.613 0.300;1.938 0.400;2.608 0.500;2.889 0.600;3.773 0.700;3.816 0.800;4.898 0.900;5.491

To reduce **XY** data to the defined by the **Cursor** data points in each position:

1. Specify **Data**: **X Axis** as **X (Time)**, **Y Axis** as **Y**.
2. Leave **X Minimum** and **X Maximum** blank.
3. Specify **Settings**: **Cursor** as 0.2, **Result is** as **Absolute**.
4. Press **Calculate**.

Y

The transform will generate an endpoint matrix named **Y** with the value at each position being the *x* value defined by the specified **Cursor**. With the example data provided above, the result for the **A1** plot is displayed on the Kinetic/Spectral Analysis Chart as follows (also displayed here to 3 decimal places):

1.613

Y at X

At x = 0.2 y = 1.613
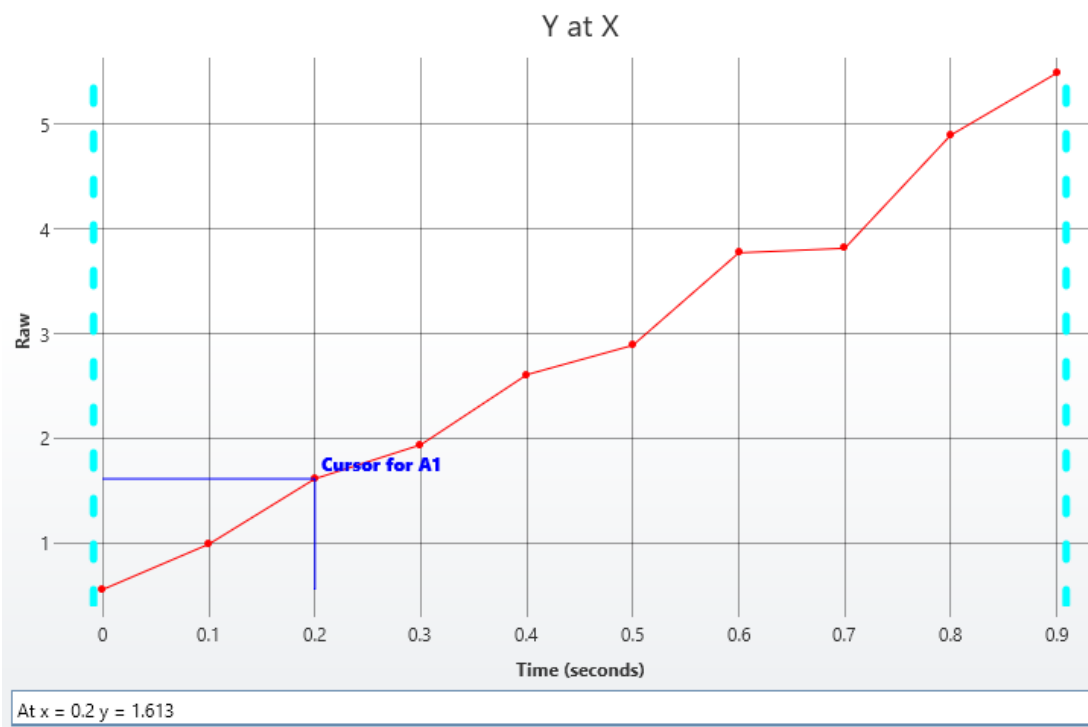
Curve Fitting

Which Fit Method Should I Use?

A single fit method that works perfectly for all assays sadly does not exist. Any curve fitting model is a compromise and simplification of reality in some way or another.

As a first step of evaluating any applied fit, it is important to eyeball the curve to check for sample preparation errors or an erratic fit.

In general, the 4PL or 5PL are the best models for dose–response curves; these model the sigmoidal nature of saturation curves very well. These methods are widely used, cited, and recommended. However, where samples fall outside these saturation limits (the upper and lower asymptotes) no result can be computed and this can be frustrating. Where this occurs, the common advice is to dilute/concentrate the sample and reassay. Alternatively, with weighting applied to these models, the fit can be biased to go through specific points (such as those nearer the *x* or *y* axis). This is useful, if it is important to have good curve accuracy at one end of the curve and to ensure the fit has an expected range of usage.

For certain assays (and especially when working with validated kits), a statistician or manufacturer may recommend a specific 4PL or 5PL model with a particular weighting. In those cases, it is important to use the same method for each run of the assay (and, of course, MyAssays Desktop supports that).

However, without this guidance, it can become tedious to try out different methods and models for each assay run. It is for this reason that MyAssays Desktop Pro includes the Best Fit option. With this option, MyAssays Desktop will perform multiple fits with different weighting options and automatically pick and apply the "best". What is deemed to be the best is open to interpretation and it is for this reason that the software allows you to select different scoring methods. The fit with the highest score is considered to be the best.

After selecting the **Best Fit** fit method, select which fits and weighting methods to consider. For example, with:
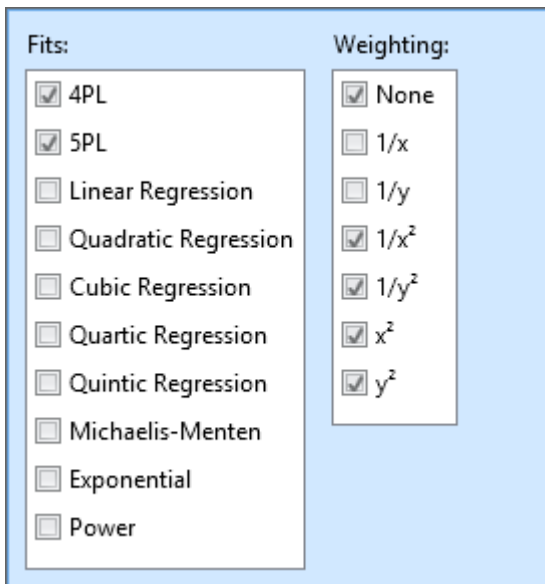
10 methods will be compared.

Next, specify a [fit score method](#) from:

- R^2
- R^2 with Low-End Accuracy
- R^2 with High-End Accuracy
- R^2 with Low-End and High-End Accuracy
- 1/Standard-Error
- 1/P-Value
- 1/AIC
- 1/AICc
- 1/BIC

R^2 is a common method for evaluating a fit, however, care should be taken when using this to compare different fit models (it does not take into account the differences between models). 1/Standard-Error is a good choice for finding the fit with the lowest standard error. The [AIC, AICc, and BIC](#) methods are useful for evaluating different models and these penalize more complicated models (i.e., those with more terms).

For even further flexibility, it is also possible to define your own fit score methods.

Alternatively, MyAssays Desktop includes the Auto Smoothed Spline method in an attempt to provide a "set-it and forget-it" solution. This is sometimes preferable in RIA where a good result is acceptable and better than a more accurate result that requires a lot more effort by the operator. Auto Smoothed Spline is based on cubic spline so the accuracy can be poor and can

lead to unnatural looking curves and erratic behaviour. The pure Cubic Spline method will exactly trace through every data point so this method attempts to smooth this out automatically. The Cubic Spline Smoothed applies a fixed smoothing parameter.

Is My Fit a Good Fit?

The term "good fit" can cover a number of factors. This topic is intended to advise how to evaluate how good your curve-fit is.

Firstly, it is always important to eyeball each curve to check for sample preparation errors or an erratic fit and, of course, verify that the curve's general direction matches the expectations of the biochemistry.

One useful measure that is easy to understand is R^2. If this is > 0.9 then, in general, the model is a good fit to the data (the closer to 1 the better). Other goodness measures are also available to provide further various metrics.

In addition, it is important to consider how good the fit is across the standards by considering the precision and accuracy of the fit at each point. This is why the software computes LLoQ and ULoQ and includes the calibrator table to clearly indicate where the precision (%CV) and accuracy (% Recovery) are within range.

It is quite common to see poorer accuracy at the upper or lower ranges of the standards. In this case, you may want to consider reassaying those samples that are out of this range (by diluting or concentrating and applying a suitable dilution factor).

Where precision is poor an outlier might be the cause. In this case, the software can be used to flag and ignore the outlier, the results can be recalculated without the offending outliers.

To automate the determination of a good fit, controls can be included. These would have a known concentration with validations used to automatically check that the computed concentrations match these expected quantities.

Accuracy and Precision

This section describes how the accuracy and precision of the Standard Curve Fit are defined, used, and computed in a quantitative analysis.

Definition

Accuracy is a measure of how close a measurement matches its known value (standard).

Precision is a measure of how close replicated measurements are to each other.

Limits

You will typically want to define acceptable limits of accuracy and precision for your assays. The acceptable limits depend on the application of the assay. MyAssays Desktop default limits are as follows:

- Accuracy ±20%
- Precision 20%

> These values can be adjusted through the Accuracy and Precision Quantification Limits of the Standard Curve Fit.

With a Standard Curve Fit, the accuracy and precision are likely to vary across the standards. It is for this reason that it is useful to determine the upper and lower limits of quantification (ULoQ and LLoQ) and only use the curve to compute values within a range that has good accuracy and precision.

These quantification limits are determined by computing the accuracy and precision of each standard measurement and computed concentration from the curve.

Computation

Accuracy

To measure the accuracy of the curve at a particular standard, the *x* value of the known concentration (expected) is compared against the *x* value at this point computed from the curve (actual).

So % Recovery or % Accuracy is simply:

= 100 * Actual / Expected

= 100 * (calculated concentration) / (defined concentration)

Precision

To measure the precision of the curve for a particular calibrator (**Standard Type** sample), the %CV is calculated as the % of the standard deviation (SD) of the standard's replicate measurements divided by their mean.

This way % CV Precision is calculated as:

100 * SD / Average

= 100* (standard deviation of calibrator)/(average value of calibrator's replicates)

Evaluation and Results

Accuracy and precision are calculated from the curve with the LLoQ and ULoQ determined from these values; the results for each standard are included in right-most columns of Curve Data Table.

To clearly indicate whether each standard is within the acceptable range or not, each row is highlighted based on whether it satisfies the precision and accuracy rules (rows in light-red fail the test).

Methods

Four Parameter Logistic Regression (4PL)

This nonlinear symmetrical sigmoidal model is known as 4-parameter logistic regression (4PL).

It is quite useful for dose–response and/or receptor–ligand binding assays, or other similar types of assays. As the name implies, it has 4 parameters that need to be determined in order to "fit the curve." The model fits data that makes a sort of S-shaped curve that is symmetrical around its inflection point.

The equation for the model is:

$$y = d + \frac{a-d}{1+\left(\frac{x}{c}\right)^b}$$

Variables

In this model we have the following:

y = the dependent variable (i.e., what you measure as the signal)

x = the independent variable (i.e., what you control, such as dose, concentration, etc.)

Coefficients of the Fit

The 4 parameters consist of the following:

- **a** = the minimum value that can be obtained (i.e., what happens at 0 dose).
- **d** = the maximum value that can be obtained (i.e., what happens at infinite dose).
- **c** = the point of inflection (i.e., the value of x at the point on the S-shaped curve halfway between a and d).
- **b** = Hill's slope of the curve (i.e., this is related to the steepness of the curve at point **c**). It could either be positive or negative.

The rearranged equation to solve x is:

$$x = c \left(\frac{a-d}{y-d} - 1\right)^{\frac{1}{b}}$$

> The **a** and **d** values might be flipped; however, **a** and **d** will always define the upper and lower asymptotes (horizontals) of the curve. **a** and **d** are the same units at y. The curve can only be used to calculate concentrations for signals within **a** and **d**. Samples outside the range of the determined **a** and **d** cannot be calculated.

4PL and IC50

A common requirement is to calculate IC50 from the fit. The IC50 is the concentration of an inhibitor where the response (or binding) is reduced by half. There are actually two ways to do this depending on what you consider IC50 to be:

- Firstly, the midpoint of the sigmoid of the 4PL is equal to the **c** coefficient of the 4PL. In this case you can simply look at the calculated **c** coefficient. Use this method if you consider the midpoint of the sigmoid to be equal to IC50. Mathematically, this is the case as it is the x point at exactly half way between the two horizontal asymptotes.

- Alternatively, if the response is measured between 0 and 100% and you consider IC50 to be where $y = 50$, then you can calculate where $y = 50$ using the equation to solve x (above), substituting in the calculated coefficients.

Linear Regression

It is a basic and easy regression model that fits data in a linear way.

Linear regression sets the linear relationship between the dependent variable (*y*) and a single independent variable (*x*). This model provides starting approximation and, although is seldom used for biological systems, is often used to visualize data relationship at a glance.

The equation for the model is:

$$y = mx + c$$

Variables

In this model we have the following:

y = the dependent variable (i.e., what you measure as the signal)

x = the independent variable (i.e., what you control, such as dose, concentration, etc.)

Coefficients of the Fit

m = the slope of the fitted line

c = the intercept of the dependent axis

The rearranged equation to solve *x* is:

$$x = \frac{y-c}{m}$$

The goal is to determine values of **m** and **c** that minimize the sum of squares (SS) of the residuals between the observed values (i.e., your data) and the predicted values (i.e., the fitted curve). As some observed values will likely be above the fitted curve and some below, you will get positive and negative residuals. SS is used to square each of the residuals, which render all the values positive, then sum them. The smaller the SS, the closer the observed values are to the predicted, the better the model predicts your data.

Fitted plot may show the nonlinear character of data. In this case, you may and are advised to consider setting other methods.

Goodness of Fit Measures

The goodness of fit measures are calculated when each fit method is used. These measures are indicators as to how position the data fits the curve fit model.

The following table describes these measures and how they can be used to assess the fit.

Name	Description
R^2	R^2 is 1 minus the ratio of the sum of the squares of the residuals divided by the sum of the squares of the differences between Y fit and the mean Y value). This will equal 1 for a perfect fit and tend toward 0 for a bad fit. In other words, R^2 is the ratio of variation that is explained by the curve-fitting model to the total variation in the model. In most situations, irreducible errors in measurement will prevent the model from explaining all the variation. Models using a larger set of factors may produce an R^2 value that is closer to 1. However, it may be that the additional factors are essentially modeling noise.
aR^2	aR^2 is the adjusted R^2 value. aR^2 is R^2 adjusted downward to compensate for over-fitting. The larger the number of independent variables is (compared to the number of observations), the lower the adjusted R^2 value will be. When using curve fitting models with a larger number of independent variables, the additional variables may be simply modeling noise.
P	P is the P-value. P is the significance of the model as a probability. It is the P-value of F for the curve-fitting model. This compares the variance attributed to the model with the variance of the residual.
SE	SE is the standard error. The standard error is the root mean square of the residuals.
F	F is the F-statistic. F is the ratio of the variance explained by the curve fitting model to the residual variance.
AIC	AIC is Akaike's Information Criterion; this provides an estimate of the quality of the curve-fitting model for comparison against other models. AIC can be used to compare curve-fitting models and determine which is the best. The model with the lowest AIC value is most likely to be correct. AIC will favour simpler curve-fitting models over those with more parameters (it achieves this by applying a penalty proportional to the number of parameters).
AICc	AICc is Akaike's Information Criterion corrected for small sample sizes. Use this method instead of AIC when comparing curve-fitting models for smaller sample sizes (number of calibrators).

Name	Description
	This method is only applicable when the number of calibrators is at least two greater than the number of parameters.
BIC	BIC is Bayesian Information Criterion; this is very similar to AIC. The key difference is that BIC imposes a higher penalty for the number of parameters in the model.
DWS	DWS is the Durbin-Watson Statistic; this is used to detect the presence of auto-correlation (at lag 1) in the residuals of the regression. DWS values range between 0 and 4. A value close to zero indicates a positive autocorrelation between the residuals. A value close to 4 indicates a negative autocorrelation between the residuals. A value around 2 indicates there is no significant auto-correlation.
DoF	DoF is Degrees of Freedom; this is computed as the number of calibrators minus the number of parameters in the fit. The number of calibrators depends on the selected **Fit To Standard** option; for **Replicates** (if there are replicates) will be more degrees of freedom than for **Average**. If there are not enough **Degrees of Freedom**, then values for **P**, **F**, **SE** and **aR^2** cannot be computed.
RMS	RMS is the Root Mean Square of the residuals (reported for Auto Smoothed Spline only). RMS is the square root of the residual mean squared.

Standard Curve Fit Configuration

Basic Properties

Best Fit

The Best Fit feature will automatically find the most appropriate fit for your data. The feature processes your data with multiple curve fits and multiple weighting methods and automatically picks the best performing fit.

> When working with multiplex data and best fit, a best fit search is performed on each analyte. Thus, the results for each analyte in an assay may use different fit methods — the best for each analyte.

Which Fits?

The default configuration will compare the 4PL and 5PL fit methods each with unweighted and various weighting models. These fit methods are the most widely used fit methods for dose–response curves.

Beyond this default configuration, you can select which fit methods and which weighting models to compare. It is also possible to define your own curve fit equation.

What is Best?

What is deemed to be the best fit can be customized. For example, you might consider the fit with the highest **R^2** to be the best. You also might want to consider the accuracy of the curve at certain regions of the fit (e.g., at the low or high end of your standards data).

To achieve this flexibility, the system uses a fit score method to quantify how good each fit is. The fit with the highest score is considered to be the best fit.

Fit Score Methods

On completion of each fit, the fit is scored using a fit score method equation. The scores of each fit are compared, and the fit with the highest score is considered to be the best. Thus, the score method equation should quantify how good the fit is.

For convenience, the following preset fit score methods are available for selection:

- R^2
- R^2 with Low-End Accuracy
- R^2 with High-End Accuracy
- R^2 with Low-End and High-End Accuracy
- 1/Standard-Error

- 1/P-Value

- 1/AIC

- 1/AICc

- 1/BIC

Beyond these presets, you can define your own fit score methods to evaluate and compare each attempted fit.

Fit Score Method Expressions

This section describes how to define your own fit score method expressions to evaluate a fit. The fit score method equation can reference the goodness measures determined from the fit:

Goodness Measures

Name	Description
R^2	R^2 is 1 minus the ratio of the sum of the squares of the residuals divided by the sum of the squares of the differences between *Y* fit and the mean *Y* value).
	This will equal 1 for a perfect fit and tend toward 0 for a bad fit.
	In other words, R^2 is the ratio of variation that is explained by the curve-fitting model to the total variation in the model.
	In most situations, irreducible errors in measurement will prevent the model from explaining all the variation. Models using a larger set of factors may produce an R^2 value that is closer to 1. However, it may be that the additional factors are essentially modeling noise.
aR^2	aR^2 is the adjusted R^2 value.
	aR^2 is R^2 adjusted downward to compensate for over-fitting. The larger the number of independent variables is (compared to the number of observations), the lower the adjusted R^2 value will be. When using curve fitting models with a larger number of independent variables, the additional variables may be simply modeling noise.
P	P is the P-value. P is the significance of the model as a probability. It is the P-value of F for the curve-fitting model. This compares the variance attributed to the model with the variance of the residual.
SE	SE is the standard error. The standard error is the root mean square of the residuals.
F	F is the F-statistic. F is the ratio of the variance explained by the curve fitting model to the residual variance.
AIC	AIC is Akaike's Information Criterion; this provides an estimate of the quality

Name	Description
	of the curve-fitting model for comparison against other models. AIC can be used to compare curve-fitting models and determine which is the best. The model with the lowest AIC value is most likely to be correct. AIC will favour simpler curve-fitting models over those with more parameters (it achieves this by applying a penalty proportional to the number of parameters).
AICc	AICc is Akaike's Information Criterion corrected for small sample sizes. Use this method instead of AIC when comparing curve-fitting models for smaller sample sizes (number of calibrators). This method is only applicable when the number of calibrators is at least two greater than the number of parameters.
BIC	BIC is Bayesian Information Criterion; this is very similar to AIC. The key difference is that BIC imposes a higher penalty for the number of parameters in the model.
DWS	DWS is the Durbin-Watson Statistic; this is used to detect the presence of auto-correlation (at lag 1) in the residuals of the regression. DWS values range between 0 and 4. A value close to zero indicates a positive autocorrelation between the residuals. A value close to 4 indicates a negative autocorrelation between the residuals. A value around 2 indicates there is no significant auto-correlation.
DoF	DoF is Degrees of Freedom; this is computed as the number of calibrators minus the number of parameters in the fit. The number of calibrators depends on the selected **Fit To Standard** option; for **Replicates** (if there are replicates) will be more degrees of freedom than for **Average**. If there are not enough **Degrees of Freedom**, then values for **P**, **F**, **SE** and **aR^2** cannot be computed.
RMS	RMS is the Root Mean Square of the residuals (reported for Auto Smoothed Spline only). RMS is the square root of the residual mean squared.

Accuracy Functions

In addition to the Goodness Measures, various accuracy functions are available to assess the accuracy of the fit at various points. These accuracy functions enable a comparison to be made between the expected value (the known x of the calibrator) and its actual value calculated from the curve.

The calibration data set is the known concentration values of the standards (x) plotted against their measurements (y). Each curve is fitted to the calibration data set.

To simplify combining accuracy functions together and with other measures (such as **R²**), the accuracy functions return a value between 0 and 1 (where 1 is perfectly accurate).

Function Name	Description
AccuracyRatio	Calculates the accuracy ratio of the expected value and its actual value. This is similar to % recovery; however, here a value between 0 and 1 where 1 is perfect accuracy (i.e., 100% recovery). A % Recovery of 120% or 80% both equal an Accuracy Ratio of 0.8. With AccuracyRatio, if the actual value differs from the expected by a factor of 2 or more, the accuracy ratio is 0.
AccuracyFactor	Calculates an accuracy factor of the expected value and its actual value. This is computed using % Recovery and a Gaussian function such that: 	% Recovery	Accuracy Factor	 	100	1	 	0	0.5	 	200	0.5	 	<0%	Value between 0.5 and 0	 	>200%	Value between 0.5 and 0	 This function avoids reporting a value of 0 for scenarios where the % Recovery is <0% and >200%.
Diff	The absolute difference between the expected value, and is the actual value.

Accuracy Function Forms

Each accuracy function comes in three forms:

- At Specified X Point
- From Low end (ends with Low)
- From High end (ends with High)

The following table provides example expressions calling accuracy functions and their result:

Expression	Result
AccuracyRatio (0.125)	The accuracy ratio for calibrator where x=0.125
AccuracyRatioLow (1)	The accuracy ratio for the calibrator with the lowest x value
AccuracyRatioLow (2)	The accuracy ratio for the calibrator with the second-lowest x value

Expression	Result
AccuracyRatioHigh (1)	The accuracy ratio for the calibrator with the highest x value
AccuracyRatioHigh (2)	The accuracy ratio for the calibrator with the second-highest x value

Combining Measures

A fit score method can reference various goodness measures and accuracy calculations. Remember that the fit with the highest score is considered to be the best. Therefore, if you plan to consider multiple measures, it is recommended that each measure is a value between 0 and 1 and that the various measures are multiplied together.

Score Method Preset Expressions

To illustrate example scoring methods, the following table lists the expressions used for each of the preset scoring methods:

Preset	Expression
R^2	R^2
R^2 with Low-End Accuracy	R^2*AccuracyFactorLow(1)
R^2 with High-End Accuracy	R^2*AccuracyFactorHigh(1)
R^2 with Low-End and High-End Accuracy	R^2*AccuracyFactorLow(1)*AccuracyFactorHigh(1)
1/Standard-Error	1/SE
1/P-Value	1/P
1/Akaike's Information Criterion	1/AIC
1/Akaike's Information Criterion corrected	1/AICc
1/Bayesian Information Criterion	1/BIC

Custom Fit Method

With this feature, you can define your own curve equation to use when fitting a curve to your calibration data. The software will automatically determine the best set of values for each unknown coefficient in your equation. In this way you can model your data using your own method.

Writing a Custom Fit Expression

Your curve equation must be defined as an expression as follows:

- Write the equation in terms of x (i.e., there must be at least one x reference)
- Use mathematical operators
- Use any single letter (excluding x and y) for each coefficient to compute. For longer coefficient names, enclose the name in square brackets (e.g., **[intercept]**)

> At least 1 unknown coefficient must be determined by the fit.

Also, you can optionally:

- Refer to TOVs of previous transforms (e.g., calculated coefficients, goodness measures)
- Insert constants (numerical values)
- Specify a custom name, weighting, inverse equation and define heuristic rules to improve the curve-fitting accuracy (see Advanced topic)

Examples of Custom Fit Expressions

For example, here are the alternatives for custom fit equation of **4PL with r (from expression)**:

- **d+((a-d)/(1+(r*x/c)^b))** – The expression with single-letter coefficients
- **tov1.d+((tov2.a-tov1.d)/(1+(r*x/tov2.c)^tov1.b))** – The expression uses TOVs from previous transforms as coefficients
- **1.23+((a-1.23)/(1+(r*x/c)^b))** – Here the **d** coefficient is a constant (1.23)
- **d+((a-d)/(1+(r*x/[Slope])^b))** – Here the **c** coefficient is named **Slope**

In addition to defining the expression, the Custom Fit Method can be configured further by using advanced custom fit method settings.

Library Curve

The Library Curve feature can be used to save a curve that was measured and computed in one assay for reuse in other assays (perhaps where no standards are measured).

Storing a Curve to the Library

Any curve created with the Standard Curve Fit transform can be extracted and saved to a Curve Library file.

The Curve Library file contains all of the data necessary to reproduce the curve and the Standards chart in future analyses. This includes:

- **Curve Configuration** – All the Standard Curve Fit transform settings (e.g., Fit Method, Calculate From, etc.) specified for the original curve
- **Curve Plot Data** – Calibration data (and average points) used in the curve fit
- **Transform Output Content** – Standard Curve Fit report content (as specified in the assay-results file from which it was created)
- **Origin** – The full path to the assay-results file from which it was created

How to: Save a Curve to the Library:

1. **Add the Standard Curve Fit Transform**

2. Select the **Standard Curve Fit** tab of the **Results** tab. If your assay comprises multiple plates, select the one that contains the analysis view for your data.

3. Press the **Save to Library...** button. If there are changes to the Standard Curve Fit settings that have not yet been calculated, then a message box is displayed advising with from which results version a curve should be saved.

4. Review and edit a name and (optionally) description for your curve library as required.

5. Press the **Finish** button.
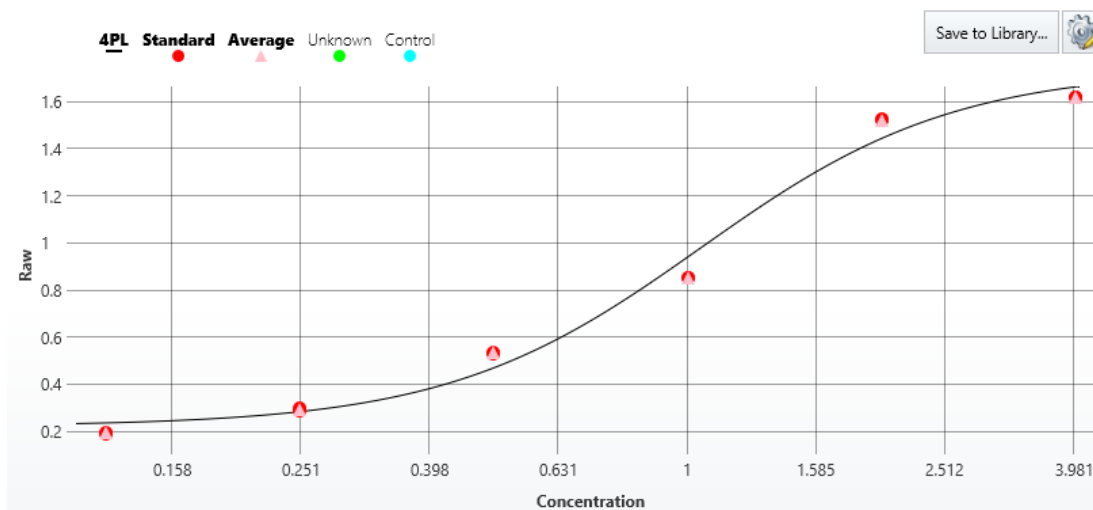
Curve Library files are stored in the **Curves** folder of your MyAssays root folder.

Calculating Results with a Curve from the Library

After the curve is saved, you can apply it by adding a Standard Curve Fit (from Library) transform. The selected curve file is used to recreate the curve for computing the concentrations of all samples measured in the referring assay.

> When using the Standard Curve Fit (from Library) transform on an assay that also includes **Standards** defined on the layout, concentrations for these standards will be computed from the original curve. These standards will not be used to recompute the curve. It can be useful to include controls or a subset of the original standards in subsequent assay runs and use the Validation feature to define expected results to detect sample preparation issues.

Library Curve Details

When reviewing the curve used by the Standard Curve Fit (from Library) transform, click the **Library Curve Details...** button on the Standards Chart to review the name, description, and assay results file, from which the Curve Library file is created. Click on the results hyperlink to open the containing folder and open then original results file from where the curve was created.
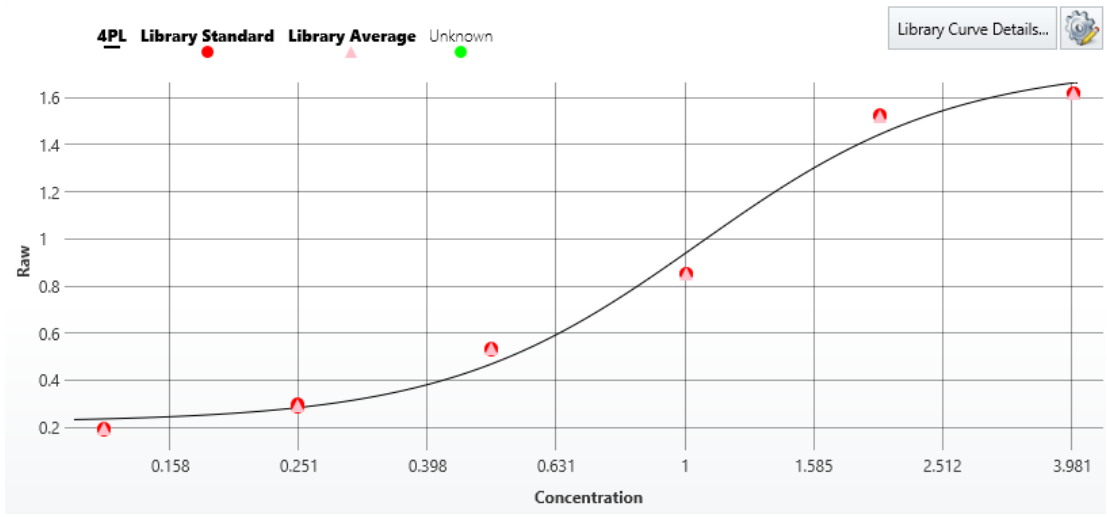
Multiplex Concentration Editor

This editor is used to specify concentration values and units of each analyte for the Standard Curve Fit transform when working with a multiplex assay.

The editor is displayed by clicking on the **Concentrations** setting drop-down.

Multiplex concentrations data can be specified in a number of different ways including:

- A start concentration for each analyte with a common dilution/concentration series
- Unique concentration values for each calibrator (standard) of each analyte
- Common concentration units for all multiplexed analytes
- Unique concentration units for each analyte

The display and behaviour of the editor depends on the way in which the data is to be specified.

For example, here the editor is displaying a start concentration and dilution series for each analyte with the same concentration units applying to all analytes:

Selecting the Analyte to Edit

The **Analyte** column of the left-hand **Analyte** grid is used to select and indicate the analyte to edit. Left-click on an analyte name to select to edit its concentration settings.

Concentration Modes

There are two modes for specifying concentrations:

- **Series** – Provide a **Start Conc.** and common series to apply to all analytes. Use this if all of the analytes share a common concentration/dilution series.
- **Non-Serial** – Provide a specific concentrations value for each calibrator of each analyte.

Use this if your analytes do not share a common concentration/dilution series or if your concentration values are not in a simple mathematical series.

Use the **Series** button to toggle the concentration mode state.

Series Mode

This mode allows you to define a series to calculate the concentrations of each analyte (from a specified starting concentration). The series calculations are specified using the selection of the multiplication or division operator along with a specified factor or divisor.

With the **Series** button toggled on, enter or paste the starting calibrator concentration values for each **Analyte** in the **Start Conc.** column of the left-hand **Analyte** grid. The right-hand grid shows the computed concentrations of the selected analyte.

For example, for the selected Kim1 analyte with a **Start Conc.** specified as 700 and **Series** mode on, with multiplication selected and a factor of 2 specified, the concentrations of the analyte are computed and displayed in the right-hand grid as shown here:

Non-Serial Mode

Non-Serial mode allows the specification of the concentration of each calibrator separately.

With the **Series** button toggled off, select an analyte in the left-hand **Analyte** grid and enter or paste concentration values on the right-hand **Calibrators** grid.

For example, here Kim1 analyte concentration values are specified in non-serial mode.

Units Mode

There are two modes for specifying the concentration units of the analytes:

- The same concentration units applies to all analytes

- Concentration units specified for each analyte

Use the ≡ **Units** button to toggle the concentration mode state.

Identical Units for All Analytes

When the ≡ **Units** button is toggled on, the single specified units of concentrations applies to all analytes. In this case, enter the units below the ≡ **Units** button.

Different Units per Analyte

When the ≡ **Units** button is toggled off, you can enter or paste different output units for each analyte in the **Units** column of the left-hand Analytes grid.

Paste Option

For all columns with a paste icon at the bottom, you can copy and paste the clipboard contents to the corresponding column. The content is pasted starting from the first row on a line

by line basis. This is useful if you have your concentration data in another application (such as Excel or notepad).

Standard Curve Fit Additional Transform Content

The Standard Curve Fit generates Transform Content elements described below. You can include in or exclude them from a report.

These report elements are listed here in alphabetical order.

Best Fit Results Table

This table displays results of Best Fit search. It includes the following columns:

- **Position** – Contains the Score-based rank for the curve fits and weighting methods processed.
- **Score** – Displays the computed result of the Scoring Method.
- **Fit Method** – Displays the curve fit method used for scoring.
- **Weight Method** – Displays the weight method used in combination with each fit method.

Coefficients Table

The three-column table that lists the calculated coefficients of the selected **Fit Method**, their value, and +/- error in correspondent columns.

> The numerical formatting option for result and error can be optionally specified separately. If not specified, default formatting is used.

Curve Data Table

The table that lists the curve-fitting data related to the fit for all of the sample groups of the specified Standard type. The table displays flagged positions using the same convention as the Sample Table.

With default configuration, the table consists of the following columns:

Column	Lists
Calibrator	The sample groups with numbers and colored icons, which contain replicates of each **Standard Type** sample.
Positions	The positions on the layout designated as containing the **Calibrator** sample groups.
Conc. (Defined)	The concentration associated for the sample group (as specified in the Concentrations setting of Standard Curve Fit).
{0}	The input replicate values associated with each **Standard Type** sample, here data is included for each position. > **{0}** implies to name of the input matrix.

Column	Lists
{0} **(Average)**	This column is additionally included if **Fit To Standard** is **Average**. This is the average of the non-flagged values of each Standard Type sample group.
{1}(Calc.)	The concentrations calculated from the curve. If **Calculate From** is: • **Replicates**, then a value is displayed for each non-flagged replicate. • **Average**, then one value is displayed per sample group. ⓘ **{1}** implies to name of the output matrix.
%CV ({1} Calc.)	The percentage coefficient of variation of the calculated concentrations.
% Accuracy	The ratio of the specified concentration (**Conc. (Defined)**) divided by the average of the replicates in the **{1}(Calc.)** column.

Each row is highlighted based on whether it satisfies the precision and accuracy rules. The background color is yellowish for rows that fit and red for those that do not.

Goodness Measures Table

Lists each calculated goodness measure followed by its value in a two-column table.

ⓘ   The numerical formatting option for the value reported for each measure can be set. The default formatting is 6 significant figures.

Fit Equations

Equation

This outputs an equation of the selected **Fit Method**.

ⓘ   Not all fit methods have a fit equation (e.g., Point-to-Point and the Cubic Spline methods). In these cases no content is listed in the report.

Equation with Values

This uses the same equation with the coefficient names replaced by the values determined from the fit. For example, the linear regression equation is:

y=mx+c

With linear regression, a value for **m** and **c** are computed. For example, if **m**=1.23 and **c**=4.56, then the equation would be:

y=1.23x+4.56

Fit Evaluations Table

This table lists Standard Curve Fit Evaluations results in a table. The first entries are the default LLOQ and ULOQ followed by any further Fit Evaluation entries.

The determined LLOQ and ULOQ match one of the specified concentration values.

LLOQ is the first concentration value (**X**) where:

- **% Accuracy** value (from the Curve Data Table) is within 100 +/- **Accuracy (±%)**

- **%CV** value (from the Curve Data Table) is less than **Precision (%CV)**

ULOQ is the highest concentration value (starting from the highest value and going backward) where:

- **% Accuracy** value (from the Curve Data Table) is within 100 +/- **Accuracy (±%)**

- **%CV** value (from the Curve Data Table) is less than **Precision (%CV)**

Standards Chart

The Standard Curve Fit plots the standard data points on axes with the measured values versus the specified concentrations. The chart displays the fit line and calibrators.

> The **Points on Curve** setting can be used to plot non-calibrator sample types on the fit line.

The chart's axes types are set in accordance with **X Axis Type** and **Y Axis Type**. The x-axis is used for the concentrations and the y-axis shows the measurements. The thick black line shows the curve calculated from these standards.

The calibrator replicates are plotted using the color of the specified **Standard Type** with dot. If **Fit To Standard** is **Average**, then the average values are also plotted (in addition to the replicates) using a triangle (pink).

Flagged points are not included in the chart.
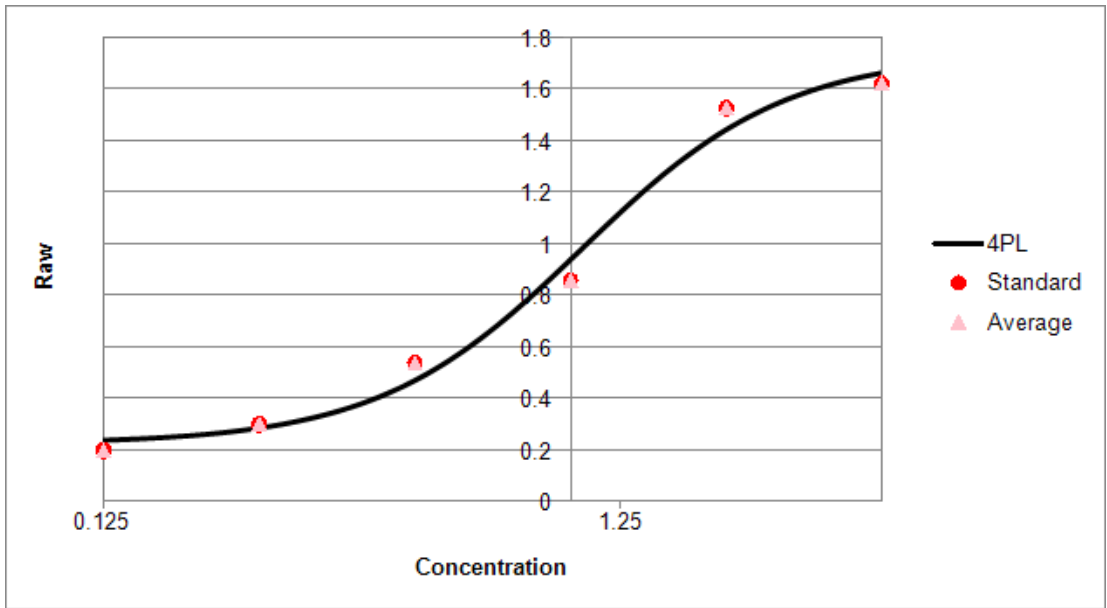
Multiple Plate Standard Curve Fit Analysis

For quantitative assays, you may arrange standards on every plate in the batch or you may decide to measure standards only on some plates in the batch. Also, you can specify how the standard curve is generated and which curve to use to calculate concentrations from for each plate.

Common Scenarios

- Measure standards on every plate in the batch. You may select to calculate concentrations using the standard curve on the same plate as the samples. **Example**: *3 plates are read, each uses the same layout, a curve is created for each plate, and concentrations are calculated using the standard curve on the same plate as each sample*.

- Measure standards on some or all plates and generate a single standard curve using standard replicates from across plates. In this case sample concentrations are calculated using a common curve. **Example**: *3 plates are read, each uses the same layout, and a single curve is created using the standard measurements from each plate. The common standard curve is then used to calculate concentrations for all samples on all plates*.

- Using a different layout on each plate read. **Example**: *3 plates are read, each uses a different layout, and a single curve is created using the standard measurements from each plate*.

- Combine these options and configure a layout that uses a specific curve or common curve for each plate.

Standard Curve Fit Evaluation with Multiple Plate

For the details of the general procedure, please see Multiple Plate Support section.

The additional complexity with evaluating the Standard Curve Fit transform across multiple plates is that for each plate the concentrations must be calculated using the correct curve as specified in the Layout Editor (and taking into account if there are more plates of data than defined in the layout).

For performance reasons, curves should not be unnecessarily recalculated, thus for evaluating the Standard Curve Fit analysis with a multiple plate assay:

1. Identify the curve to use for each plate.

2. Prepare the calibration data for each required curve.

3. Fit each curve once.

Then, after the curve data has been prepared, go through each plate and use the associated fit-ted curve to calculate the concentration values.

> ℹ️ There may be more than one curve generated.

Preparing Calibration Data

1. Where the **Curve selection** for a plate refers to the curve on a specific plate (and not the average), the calibration data is prepared using the calibrator data from the specified plate number.

2. If the curve selection is an **Average Across Plate** selection, then the calibration data for the curve is obtained from all layouts with this setting; to be clear, all plates with this selection will use the same common curve that is computed from points across separate plates.

> 📝 As with all other analyses, flagged points are not included in the fit.

If the Standard Curve Fit transform's **FitToStandard** is **Replicates**, then this is the data that will be used for the fit. If **FitToStandard** is **Average**, then the replicates are averaged (i.e., for each *X* value, there is only 1 *Y* value and that *Y* value is the average of the non-flagged replicates).

Different Concentrations per Container/Plate

When the data for the curve is obtained from the same plate that is being evaluated, you can provide different concentration values per plate/container. With this configuration, the standard concentrations used for the fit are as defined in the Plates section in the XML Editor.

How to: Specify Different Concentrations per Container/Plate

<StandardCurveFitStandardType="2"FitMethod="4PL"
Concentrations="0.125,0.25,0.5,1,2,4"XAxisTitle="Concentration" YAxisTitle="Raw"FitToStandard="Average"CalculateFrom="Replicates">

<Plates>

<Container Concentrations="1,2,4"/>

<Container Concentrations="10,20,40"/>

<Container Concentrations="100,200,400"/>

</Plates>

</StandardCurveFit>

If there are more data containers than Container elements defined, then those additional containers use the primary Concentrations attribute of the Standard Curve Fit properties. If there

are less containers in the data containers than Container elements defined, then those additional Container elements are simply ignored.

Advanced Settings

Custom Fit Advanced Settings

Overview

Advanced settings are available for the [Standard Curve Fit](#)'s **Custom** curve fit option. It addition to providing a custom curve fit expression, it is possible to define further settings including:

- Name – A name for your custom fit (to be displayed on charts and the report)
- Weighting – A weighting model to apply
- Inverse Equation – How to compute *x* values from *y* (i.e., provide an equation in terms of *y*).
- Coefficient Estimate Heuristics – Rules for estimating each coefficient's starting value
- Coefficient Constraints/Ranges – Constraints or acceptable ranges for each coefficient

via the XML editor.

How to: Add Attributes to a Custom Fit Configuration

1. Select the **XML** tab in the **Assay Properties** panel (If the **XML** tab is not immediately visible, press the **<< Show All Properties** button to expand).
2. Locate the **StandardCurveFit** element.
3. Locate the child element **Method**.
4. Provide your required settings for the custom fit in the **Method** element directly.
5. Press **Apply** to save changes.

Attributes and Syntax

You can calibrate Custom fit with the following optional attributes:

DisplayName	A title of the fit for [Standards Chart](#) and [Calculation Log](#).
Inverse	The inverse equation of the fit in terms of *y*. This is used to compute each *x* value from the curve (so ensure this is correct). If this is not provided, then results will be computed using an automated but slower method.
Weight	The weighting method to use.

Configuring Coefficient Estimate Heuristics

Defining a good starting point for calculating initial coefficient estimates improves the accuracy and speed of the fit. For a custom fit, you can provide the starting point estimates for any or all of the coefficients.

In addition, a range can be specified to limit or constrain the fit.

To specify an estimate, add an **Estimate** element (as a child of the **Method**) for each required estimate and set the attributes in accordance with the table below.

Expression	A numeric value or a heuristic expression to calculate.
Name	A name of a coefficient for which the estimate is for.
Min	The minimum value to limit the search range for the coefficient value. If not specified, then the range is -Infinity.
Max	The maximum value to limit the search range for the coefficient value. If not specified, then the range is +Infinity.

Heuristics expressions are used to compute starting estimates. An expression can reference aspects of the calibration data. For example, with 4PL model, the expected result for **a** coefficient might be the y value at the peak of the curve. In this case, you can reference *maxy* variable to define a starting estimate for an upper asymptote (**a**). It will refer to the maximum y value of your calibration data.

The following variables are available for reference.

Variable	Value
averagex	Average of all x values
averagey	Average of all y values
minx	Minimum x
miny	Minimum y
maxx	Maximum x
maxy	Maximum y
yatminx	y at the minimum x
yatmaxx	y at the maximum x
xatminy	x at minimum y
xatmaxy	x at maximum y
medianx	Median of all x values
mediany	Median of all y values
slope	Slope of the data set
intercept	Intercept of the data set

> For more sophisticated estimates, you can use [conditional function](). For example, if the coefficient value depends on whether the y increases or decreases with x, you can compare *yatmaxx* and *yatminx* values and adjust the estimate value accordingly.

Example Configuration

The following XML snippet shows a complete configuration of the **Method** element with

- Custom Display Name
- Inverse Expression
- Estimate Heuristics Expressions for each coefficient

```
<Method DisplayName="4PL (from expression)" Expression="d+((a-d)/(1+(x/c)^b))" Inverse="c*pow(((a-d)/(y-d))-1,1/b)" xmlns="http://www.myassays.com/bestfitsearch">
<Estimate Name="a" Expression="maxx" />
<Estimate Name="d" Expression="minx" />
<Estimate Name="b" Expression="if(yatminx &lt; yatmaxx,3,-3)" />
<Estimate Name="c" Expression="medianx" />
</Method>
```

Standard Curve Fit Custom Flag Labels

When computing concentration values from a curve, there might be samples outside certain ranges that should be flagged and reported with a custom label rather than as a result or error.

The Standard Curve Fit Custom Flag Labels feature enables rules to be defined to specify when a result should be flagged and reported using a label.

Example Uses

A typical use of this feature is to label results for standards where the results cannot or should not be computed from the curve. For example, if a sample is outside the usable range of the curve, then it might be useful to flag and label the result accordingly.

What is considered as the usable range of the curve is open to your own interpretation. For example, the use of the curve could be limited by:

- the determined ULOQ and LLOQ values

- the mathematical limits of the curve

- your own defined limits (defined as absolute values or using expressions with limits computed from the curve fit)

The Standard Curve Fit Custom Flag Label is an advanced option only available through XML configuration. This feature is related to the Custom Flag Label feature, in this case Standard Curve Fit Custom Flag Labels have higher precedence and override any other defined labels.

You may configure custom flagging rules in the Standard Curve Fit transform based on the transform's input or output data (calculated from the curve). If any of these rules are satisfied, then result is flagged with a specified label applied to it.

The rules are defined using expressions written in terms of *x* to define when a result should be flagged.

> The feature only applies to data points specifically flagged by the transform itself. If an input point is already flagged, then any existing custom label (or the absence of the custom label) is copied to the output.

Evaluation and Results

You can define any number of Custom Flag Label rules in any order. If multiple labels are configured, only the first label condition to be evaluated as true is used (in the order they appear in the XML). These specified rules take precedence over the default flagging settings or optionally specified CurveError. Therefore, if any of the rules with **Type="Input"** are evaluated as true, then these will be used in place of any curve error.

For multiple plate assays, the associated curve for the position's plate is used.

If a flag condition is satisfied and a Custom Flag Label is used, then an entry is added to the Calculation Log detailing this application. If there is any error in evaluating a Condition, the details of this is added to the error log.

How to: Add Custom Flag Label Rules

Standard Curve Fit Custom Flag Labels are configured using XML. To add flagging rules to a Standard Curve Fit transform configuration:

1. Select the **XML** tab in the **Assay Properties** panel (If the **XML** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Locate the **StandardCurveFit** element.

3. Add a **Flags** element (as a child of the **StandardCurveFit**) and, optionally, a **CurveError** element.

4. Add a child element **Flag** for each required flagging rule. Set the attributes (in accordance with the following table).

5. Press **Apply** to save changes to the protocol.

CurveError Optional Setting

The optional CurveError setting is used to specify a default custom flag label to use if a value cannot be computed from the curve (i.e., if the result is flagged on the input matrix or while calculating results). It simply replaces the default flagged result in the report with specified CurveError Label.

This is specified in the XML inside the **Flags** element with following syntax:

<Flags CurveError="*My Custom Error Label*" >

Flag Element Attributes

The following table lists the attributes of the Flag element that must be specified to define the flag rule:

Type	The value of x depends on the expression type: • For Type="Input", x is the value of the input position. • For Type="Output", x is the value computed for the output position.
Condition	The condition to evaluate (typically a conditional expression). The flag condition expression can refer to:

	• the value of each position using the reference *x* • the computed <u>coefficients of the curve and goodness measures</u> • <u>Standard Curve Named Evaluations</u> • sample values of the input matrix
Label	If a flag rule is satisfied (i.e., the **Condition** is evaluated to true), then the result of the transform for the position is flagged with the associated **Label**. If you insert **{x}** in the **Label**, it inserts the value of x (e.g., **{x} is Below LLD** might be reported as **0.25 is Below LLD**).

Specifying Rules with XML

When entering conditions as XML there are certain symbols that must be encoded using correct XML syntax. These are listed in the table below.

Operator	Description	XML Encoding
<	Less Than	<
>	Greater Than	>
&	Ampersand	&
"	Double Quote	"
'	Apostrophe/Single Quote	'

For example, if the condition is **x < a** then this would be encoded in XML using **x < a**

Example Configuration

The following XML snippet shows a complete configuration of the StandardCurveFit element with Flag rules. With this configuration, any sample whose measurement response is above or below the computed asymptotes of the <u>4PL</u> (the **a** and **d** coefficients of the fit) will have a label associated with it accordingly.

```
<StandardCurveFit StandardType="2" FitMethod="4PL"
Concentrations="0.125,0.25,0.5,1,2,4" XAxisTitle="Concentration"  YAxisTitle="Raw"
FitToStandard="Average" CalculateFrom="Average">

    <Flags CurveError="Could not calculate from curve" >

        <Flag Type="Input" Condition="x &lt; min(a,d)" Label="Below Lower
        Asymptote" />

        <Flag Type="Input" Condition="x &gt; max(a,d)" Label="Above Upper
        Asymptote" />

    </Flags>
```

</StandardCurveFit>

Example Flag Conditions

The following table lists further example flag conditions.

XML Condition	Description
<Flag Type="Input" Condition="x < Blank1+3*sd(Blank1)" Label="{x} Below LLD" />	If the measurement response is less than Blank1 + 3SD, then the result is flagged and reported with the Label.
<Flag Type="Input" Condition="x < min (a,d)" Label="Below Lower Asymptote" />	If the measurement response is less than the lower value of the a and d coefficients (as computed from the curve), then the result is flagged and reported with the Label.
<Flag Type="Input" Condition="x < 0.125" Label="OD < 0.125" />	If the measurement response is less than 0.125, then the result is flagged and reported with the Label.
<Flag Type="Output" Condition="x < 0.125" Label="Conc. < 0.125" />	If the computed concentration is less than 0.125, then the result is flagged and reported with the Label.

Standard Curve Fit Evaluations

In addition to computing concentrations, ULOQ, and LLOQ from the curve, you can evaluate expressions that:

- Calculate *X* or *Y* values on the curve (using interpolation/extrapolation)
- Reference the coefficients and goodness measures of the curve
- Can be referred to as a variables in other evaluations or Standard Curve Fit custom flag labels
- Can be referenced as the Transform Output Variables

This capability is useful for a range of scenarios, including calculating and reporting limits of detection and quantification.

How to: Add Evaluations to a Standard Curve Fit Transform Configuration

1. Select the **XML** tab in the **Assay Properties** panel (If the **XML** tab is not immediately visible, press the **<< Show All Properties** button to expand).
2. Locate the **StandardCurveFit** element.
3. Add an **Evaluations** element (as a child of the **StandardCurveFit**).
4. Add a child element **Evaluation** element for each required evaluation. Set the evaluation attributes in accordance with the table below.
5. Press **Apply** to save changes to the protocol.

Evaluation Attributes and Syntax

Expression	The expression to evaluate. It can refer to: • The computed coefficients of the curve, Transform Output Variables, and goodness measures • Any defined above in the XML Standard Curve Fit Evaluation with **Name** attribute specified • Sample values of the input matrix Since the result is a single numerical value, the expression cannot refer to *x* or *y* variables.
Type	This setting describes how the expression is treated in report: • **CalcX** – Calculates the *X* value from the fit line using **Expression** as *Y*

	value. The resulting values are reported using numerical formatting of input matrix. • **CalcY** – Calculates the *Y* value from the fit line using **Expression** as *X* value. The resulting values are reported using standard numerical formatting. • **Parameter** – Outputs the computed **Expression** value.
Description	Optionally added textual description of the expression. If this is not specified, the **Expression** itself is used in report.
Name	Optional element that defines a name for evaluation. If it is specified, the **Name** can be used as a variable for other XML configurations (as described below).

\<Settings\>

   \<StandardCurveFitStandardType="2" FitMethod="4PL" Concentrations="0.25,0.5,1,2,4,8" XAxisTitle="Concentration" YAxisTitle="OD" FitToStandard="Average" CalculateFrom="Average"\>

        \<Evaluations\>

          \<EvaluationType=="Type" Expression="Expression" Description="Description"Name="Name" Plot="true" /\>

        \</Evaluations\>

   \</StandardCurveFit\>

\</Settings\>

Named Evaluations

If the **Name** attribute is set, then the evaluation can be referred by this **Name** as a variable in:

- Context variables in evaluations that are defined in XML downward to it
- Standard Curve Flag Label conditions
- Transform Output Variables

> ℹ The **LLOQ** and **ULOQ** are computed automatically and can be used as an evaluation and flag variables too.

Configuring Standard Curve Evaluations

Evaluations are configured using XML and are evaluated using the plate for the associated container. This means that **Blank1** always refers to the first blank sample group on the container. Further details are described in the [Multiple Plate Standard Curve Fit Analysis](#) topic.

For example, the LLD is often computed as the concentration (X) at the average of the blank replicates + 3 * standard deviation of the blank replicates. This can be computed as an expression that calculates X from the curve where Y **= Blank + 3 * sd(Blank)**.

The LLOQ and ULOQ calculations are determined automatically and equal a concentration value (i.e., one of the values defined in Concentrations).

If you create named evaluation LLOQ or ULOQ, then it replaces the default LLOQ and ULOQ calculation, e.g.:

```
<Evaluations>
    <EvaluationType="CalcY" Expression="Control1 + (10 * sd(Control1))"
Description="My LLoQ"Name="LLoQ" />
</Evaluations>
```

Results and Report

The LLOQ and ULOQ are the first entries, and any further evaluations are added below them in the table.

The **Evaluation** data is displayed below the interactive chart and in the report as a simple two-column table. The first column contains **Description** if specified or **Expression** if it was not. The second column contains the numerical value of the evaluated result or any syntax/computation error in red. In other words, report syntax errors in a table (in same way as the [Evaluation feature](#)).

Zero Concentration Calibrator

Using a theoretical zero concentration as a standard curve calibrator can be the source of many mathematical problems when fitting standard curves. Firstly, a zero standard can skew the fitting process with certain fit methods leading to an unexpected curve. Also, with any fit method, a zero value cannot be plotted on a logarithmic X Axis (as is typically used to plot a standard curve). Another issue is that a zero concentration will break fit methods with weighting models such as $1/x^2$. A common workaround is to use a suitably small specified concentration value instead (such as 0.001).

If using a suitably small value is not ideal there are some other options.

Measure Zero Without Using it as a Calibrator

In this scenario the zero standard is measured but not included as a calibrator for the fit; the zero standard is used as a control (and not as a calibrator). To achieve, this add the **Zero** sample type to your layout configuration to denote the location of the zero concentration sample(s).

Including a Zero Standard Calibrator

In this case standards are added to the layout as normal, however, the first standard (**Standard1**) is given a known concentration of 0. In some cases, this might not map directly to your actual standard numberings and you might prefer for **Standard1** to map to the first non-zero standard. In this case, please try the following scenario.

Including a Zero Calibrator

With this scenario, you can specify a **Zero** sample group to be used as a calibrator along with the **Standard** groups. If included, the curve fit will include this sample (i.e., all of its replicates) as a part of the calibrators (with a zero concentration value). The values will be added in the same way as for the standards (i.e., **Average** or **Replicates** using **Calculate From** setting). The *X* value of these points will be zero.

To achieve this, first add the **Zero** sample type to your layout along with your standards. Next, to include the Zero group in the calibrators of the curve, an additional configuration step is required:

How to: Add Zero Concentration to a Standard Curve Fit

1. Select the **XML** tab in the **Assay Properties** panel (If the **XML** tab is not immediately visible, press the **<< Show All Properties** button to expand).
2. Locate the **StandardCurveFit** element.
3. Add an **ZeroType** and optionally **ZeroGroup** attributes (inside the **StandardCurveFit**).
4. Press **Apply** to save changes to the protocol.

Attributes and Syntax

The Zero concentration is calibrated via XML with the following attributes:

ZeroType	The sample type index for a zero sample.
ZeroGroup	The sample group number for a zero sample. If **ZeroType** is specified but this value is omitted, then a default value of 1 is applied.

For example:

<Settings>

   <StandardCurveFitStandardType="2" FitMethod="4PL" Concentrations="0.25,0.5,1,2,4,8" XAxisTitle="Concentration" YAxisTitle="OD" FitToStandard="Average" CalculateFrom="Average" ZeroType="27" ZeroGroup="1">

This will set the zero type to **Zero** sample type and zero group to 1.

> The zero data points are included in Standards Chart in the same way as standards (i.e., with pink average triangle when relevant) but only if the **X Axis Type** is **Linear**. If it is **Logarithmic**, then these values do not appear on the chart as a calibrator.

Weighting Infinity Resolution

Applying weighting on calibration data points can lower the overall error of the method and therefore improve the quality of the results. However, weighting could result in the weight for a zero point to be infinite and thus skew the curve. An infinite weight can occur if the weight method is one of the forms: 1/y or 1/x (or $1/y^2$, etc.) and the calibration point has a 0 value in the axis of the denominator (e.g., (0,0)).

The notification will advise you if this issue has occurred. In this case, to avoid the infinite weights, you can use the following XML attributes:

- **WeightInfinityResolution**

  Specifies how to resolve infinite weights for the curve:

  - **None** – Any infinite weights are used.
  - **MaxFiniteMultipliedByFactor** – Any infinite weight will be replaced by a finite weight which is computed as the maximum finite weight in the calibrators multiplied by the specified factor (**WeightInfinityReplacementFiniteMaxFactor**).

- **WeightInfinityReplacementFiniteMaxFactor**

When **WeightInfinityResolutionWeighting** is **MaxFiniteMultipliedByFactor**, this is the factor to multiply the maximum finite weight by, the result is used in place of any infinite weights.

For example:

<Settings>

    <StandardCurveFitStandardType="2" FitMethod="4PL" Concentrations="0.25,0.5,1,2,4,8" XAxisTitle="Concentration" YAxisTitle="OD" FitToStandard="Average" CalculateFrom="Average" WeightInfinityResolution="MaxFiniteMultipliedByFactor" WeightInfinityReplacementFiniteMaxFactor="1">

With this configuration, the infinite weight will be replaced with the maximum finite weight multiplied by 1.

Standard Curve Fit Coefficient Constraints

Overview

For certain types of curve fitting applications, it is useful to be able to constrain one or more coefficients of the fit to specific values or ranges. With coefficient constraints imposed, the resulting curve fit equation will follow these constraints.

For example, when using the 4PL or 5PL model, it might be useful to constrain the upper and/or lower asymptotes of the curve to equal known saturation points (such as **NSB** or **B0**). These constraints could be based on measurements from specific samples in the assay and used to set limits on the computed curve.

Also, where you expect the resulting fit to fall within certain ranges, you can define constraints to guide the curve fitting process as required. For example, you might be using a curve fit equation that uses a coefficient that must be within a certain range (such as positive), a range constraint defined with a bound can be applied to direct the curve fit accordingly.

Configuration Options

You may configure constraints on one or more the coefficients of the fit method. Each coefficient can be constrained using either:

- a fixed value (specified using an expression that can refer to samples or mathematical operations)

- bound (upper, lower or both)

The value of these constraints can be specified using an expression and may refer to a TOV.

Coefficient Constraints and Parallel Line Analysis

Coefficient Constraints are useful when performing Parallel Line Analysis. In this case, an unconstrained fit can be performed followed by constrained fits using limits defined from the unconstrained fit. The resulting constrained fits can then be compared and considered for parallelism.

> An alternative to using a fixed coefficient constraint is to define a Custom Fit expression with a constant value.

How to: Add Coefficient Constraints to a Standard Curve Fit Transform Configuration

1. Select the **XML** tab in the **Assay Properties** panel (If the **XML** tab is not immediately visible, press the **<< Show All Properties** button to expand).

2. Locate the **StandardCurveFit** element.

3. Add a **FitConstraints** element (as a child of the **StandardCurveFit**).

4. Add a child element **Constraint** for each required operation. Set the attributes in accordance with the syntax below.

5. Press **Apply** to save changes to the protocol.

Attributes and Syntax

For each constraint, with the **Coeff** attribute specify a coefficient on which the constraint will be applied.

The value of these constraints can be specified using an expression (that results in a single numerical value) and may refer to a TOV. The interpretation of the constraint depends on the type of restriction setting:

- **Fixed** – Sets a coefficient to a fixed value

- **LowerBound** – Limits the search range for the coefficient to be greater than specified constraint

- **UpperBound** – Limits the search range for the coefficient to be less than specified constraint

> The **UpperBound** and **LowerBound** can be used separately or in a conjunction. However, **Fixed** type overrides any **UpperBound** or **LowerBound** setting.

Example Configuration

The following XML snippet shows a complete configuration of the StandardCurveFit element with Constraints for 4PL fit method. In this example, the **a** coefficient is constrained to the value of **Blank1** and the **d** coefficient is constrained to the average **d** value as computed from two previous Standard Curve Fit transforms. The result is a curve with constrained upper and lower asymptotes.

```
<Settings>
    <StandardCurveFitStandardType="2" FitMethod="4PL" Concentrations="0.25,0.5,1,2,4,8" XAxisTitle="Concentration" YAxisTitle="OD" FitToStandard="Average" CalculateFrom="Re-eplicates">
        <FitConstraints>
            <ConstraintCoeff="a" Fixed="Blank1" />
            <ConstraintCoeff="b" LowerBound="0" />
            <ConstraintCoeff="c" Upperbound="100" />
```

```
    <ConstraintCoeff="d" Fixed="average(tov2.d, tov1.d)" />

</FitConstraints>
```

With this configuration, the **d** coefficient of the fit is constrained to equal the average of sim-ilar coefficients ([TOV](TOV)s) from previous transforms (. The **a** equals to average of replicates of **Blank1** sample, the **b** must be greater than 0, and the **c** coefficient is configured to be less than 100.

Expressions

Overview

Expressions are available for use throughout MyAssays Desktop to provide flexibility in data-analysis design and configuration.

An expression is simply an operation to be computed. In the simplest case, an expression can refer to a specific numerical value, measurement, or result. For example, the expression **Control1** refers to the average of the Control1 sample replicates (that are not flagged).

> When writing expressions that refer to specific samples, in general, it is preferable to refer to a sample group, such as **Control1**, rather than specific layout positions (such as **A1**). With this approach, if your sample layout changes (perhaps you run the assay with a different sample layout), then you will not need to update your expressions.

An expression might perform mathematical or statistical operations on these values. For example, the expression **pcv(Control1)** computes the %CV of the **Control1** replicates.

An expression can be applied at various levels of the data analysis to define and compute various results. For example, matrix calculations, quantification limits, validation rules, values from fitted curves, and best fit score methods (to decide on the best fit) can all be defined using expressions.

Wherever a numerical value can be entered as a settings parameter, an expression can usually be provided to compute that parameter from other sources.

The Expressions system has been designed to:

- ☑ **Provide custom analysis using a familiar mathematical syntax (similar to Excel)**
- ☑ **Provide an easy way to reference sample data**
- ☑ **Take into account flagged data**

Expressions can refer to measurements or calculated results from specific layout positions (e.g., **A1**), sample groups (e.g., **Control1**), variables, and also the results of other expressions.

Culture Settings

When providing numerical values or expressions through the user interface, enter numerical values and lists using your system's regional settings. There are two settings that can vary between culture settings:

- **Decimal separator** – Typically a period (**.**) on non-European systems and a comma (**,**) on typical European system

- **List separator** – Typically a comma (**,**) on non-European systems and a semi-colon (**;**) on typical European system

For example, depending on your system's culture settings, an example expression could be entered in two forms:

mean(1.5,2.5)

mean(1,5;2,5)

> For advanced users working with data entered directly into the XML editor, it is necessary to follow XML conventions and provide data using invariant settings (that is, period decimal separator for numeric values and a comma list separator).

Syntax and Context

The syntax of expressions is consistent throughout MyAssays Desktop. However, the application of an expression depends on the context it is being used in. For example, expressions are used in the configuration of transforms, validations, and evaluations. In each case, the application of the expression is slightly different.

Where an expression is used as a parameter to another setting, you can provide an expression that can be evaluated to a numerical result, such as **Control1 * 10**.

There are several situations where an expression is entered in terms of x (where x refers to an input value that will be substituted with a value for each evaluation of the expression). For example, the Expression by Matrix transform expects an expression in terms of x. In this context, you can provide an expression such as **x-Blank1** to subtract the mean of **Blank1** from all samples.

Where an expression is entered in terms of x, the expression is evaluated multiple times with x being substituted with different values. In the case of the Expression by Matrix transform, the expression is applied to every position on the input matrix to calculate the result of the output matrix.

There are also scenarios where a Boolean expression is expected. Validations are defined using a Boolean expression to define rules that must be satisfied for the assay to be considered valid.

For example, a validation rule can be defined with the expression **Standard1<Standard2** to check that the standard measurements are in the expected order. Similarly, a validation expression **x < 15** could be used along with %CV data to validate that all samples have a %CV of less than 15.

A typical expression comprises of:

- [operations](#), (e.g., +, *, /, ^)
- [operands](#) (e.g., A1, 1.23, x, Control1, TOV1.a)
- [functions](#) (e.g. pcv, sd, median, floor)

The following table lists some examples of expressions you may use in your assay.

Example Expressions

> ℹ️ All expression examples provided here use a period decimal separator(.) and a comma list separator(,). Use [local settings of your system](#) when entering expressions on your own system.

Expression	Evaluated to
Unknown1	The average of the replicates of **Unknown1** (that are not flagged).
Unknown	The average of all **Unknown** samples (that are not flagged).
A1	The data for position **A1**.
A1+A2	The sum of the data at positions **A1** and **A2**.
mean(A1,A2,A3)	The average of the data at positions **A1**, **A2**, and **A3**.
pcv(Unknown1)	The percentage coefficient of variation of **Unknown1**.
Blank1+(3*sd(Blank1))	The average of the sum of **Blank1** plus 3 times the standard deviation of **Blank1** (this is often used to compute LLD).
pcv ([Unknown1,Unknown2])	The percentage coefficient of variation of all **Unknown1** and **Unknown2** replicates.
median(Standard1)	The median of the replicates of **Standard1** (that are not flagged).
x-Blank1	The value of variable x minus the average of **Blank1** position.
x>15	A Boolean expression that would evaluate to 1 (true) if the value of variable x is greater than 15; otherwise it evaluates to 0 (false).
Ifthenelse(x>15,15,x)	If the value of x is greater than 15, then the result is 15; otherwise the result is x.
x-y	The difference between x and y. Here x and y are the values of corresponding positions from two matrices of data (as used in the [Expression by Matrix (Dual) transform](#)).
median(Unknown.2.All)	The median of all **Unknown** replicates on matrix 2 across all containers.
Unknown1.2.3	The average of **Unknown1** sample group on the 2nd matrix of

Expression	Evaluated to
	3rd container.
TOV1.R2	The R^2 value of the curve fit as used in the first transform (referenced using a <u>Transform Output Variable</u>).
GroupValue("Blank",1)	The value of the group that is **Blank1**. (This is equivalent to the reference **Blank1**, however this method can be useful for applying calculations on group number references.)
x-GroupValue ("Blank",PosGroup())	The value of variable *x* minus the average of the **Blank** sample which has a group number that matches the group number of the sample at the position where the same is being evaluated. This technique can be used to write expressions that apply a calculation in accordance to a layout, such as subtracting Blanks values on 1-to-1 basis. With a layout where each Unknown group has its own **Blank** group (e.g., **Unknown1**, **Blank1**, **Unknown2**, **Blank2**, etc.) this expression will subtract the blank group associated with each sample based on the layout.

Operators

Unary Arithmetic Operator

Operator	Operation	Examples
-	Minus Sign	-1.23 -x

Binary Arithmetic Operators

Operator	Operation	Example
+	Addition	2+1 $x+y$ $x+1$
-	Subtraction	2-1 $x-y$ x-1
*	Multiplication	2*3 $x*y$ $y*2$
/	Division	2/2 x/y $x/2$
^	Raising to a power	2^3 $x^\wedge y$ $x^\wedge 2$

Binary Logical Operators

The numerical evaluation of 1 for true and 0 for false. Thus, 1>0 =1.

Operator	Operation	Examples
<	Less Than	1<2 $x<y$ $x<2$
>	Greater Than	2>1 $x>y$ $x>2$

Operator	Operation	Examples
=	Equal To	1=1 *x=y* *x=2*
<=	Less Than Or Equal	3<=9 3<=3 *x<=y* *x<=x* *x<=3*
>=	Greater Than Or Equal	9>=3 3>=3 *y>=x* *x>=x* *x>=9*
<>	Not Equal	2<>1 *x<>y* *x<>3*

Boolean Operations

The Boolean operations (and, or, not) are implemented as Conditional Functions.

Operands

Numbers

Number operands are simply numbers (e.g., 1.23, -100, 14).

> Ensure that all number values entered into the user interface are provided using the local settings of your system.

Variables

The *x* and *y* variables refer to the value of each position on the associated matrix. All references to these variables in expressions are equated to the associated values.

For example, in the Expression by Matrix transform, you may enter the **x*100** expression, where the *x* variable will refer to the positions of the input matrix. In the resulting matrix, all positions will be multiplied by 100. In Expression by Matrix (Dual), the *x* and *y* variables are set to the position values of the first and second matrices. Thus, when you enter an expression **x+y**, each position on the resulting matrix will be the sum of input matrices values.

Variable	Relevant For
None	Installed Protocols, **Certain Endpoint**: *Auto Flag*, *Average*, *Matrix Difference*, *Median*, *Percentage*, *Percentage Coefficient of Variation*, *Replicate Outlier Removal*, *Standard Curve Fit*, *Standard Deviation*, *Variance*
x	**Certain Endpoint**: *Blank Correction*, *Dose–Response Curves*, *Dilution Factors*, *Expression by Matrix*, *Expression by Position*, *Factor*, XY Blank Correction
x, y	Expression by Matrix (Dual), XY Expression (Dual)
y	XY Expression

Fit Transforms Context Parameters

XY Reduction Fit Parameter and Standard Curve Fit transforms calculate the context parameters of the fit. The former refers to the parameters in configuration settings, and the latter uses them in flagging conditions and/or additional evaluations. These fit transform context parameters are:

- [Goodness of Fit Measures](#) calculated from the fit.

Name	Expression Syntax
R^2	r2
aR^2	ar2
F	f
P	p
SE	se
AIC	aic
AICc	aicc
BIC	bic
DWS	dws
DoF	dof
RMS	rms

- **Coefficients of the Fit** – These vary depending on the selected fit method and are specified within documentation of these methods.

Standard Curve Fit also produces additional values (such as quantification limits) and any defined [named evaluations](#). These can be optionally used for further evaluations of custom flagging within this transform.

> Fit Transforms Context Parameters apply within fit transforms only and cannot be used in general configuration of the assay (e.g., [validation](#) or [evaluation](#) tables) or other transforms.

Named Evaluation Variables

Any [evaluation](#) (including [validation](#) and [sample label](#) conditions) computed after the [Named Evaluation Variable](#) can refer to it.

Transform Output Variables

Any transform evaluated after the transform that produces the [output variable](#) can refer to it. This applies only where the later transform supports parameters with expression.

Layout References

When an evaluation depends on and is associated with certain layout, the expression can include references to positions, sample groups, and/or types defined on the layout. These are defined with case-insensitive input (i.e., uppercase and lowercase letters in expressions are treated as being the same).

For example, **Unknown1** is the same as **unknown1**, **UNKNOWN1**, and **UnKnOWN1**.

General information on referencing the sample layout and its components is provided in the Layout References in Expressions.

Position References

A position ID refers to a specific position on the layout (e.g., **A1**, **H12**). To learn more, see Positioning on the Layout.

The available dimensions depend on the layout read. A position on a particular matrix and plate can be specified with a period, as described in <u>sample references in multiple container assays</u>.

> When writing expressions, it is better to refer to sample groups such as **Blank1** rather than specific positions. If you have to modify your layout (perhaps because of pipetting error), you do not need to update your expressions as the group reference will refer to the modified layout.

Sample References

Sample operand can contain a reference to:

- **Sample group** – It can refer to any group of any type defined on the layout. To use this operand in the expression, type in the sample type name (e.g., **Unknown**, **Blank**, **Standard**) and sample group number. **Example**: *Unknown1 refers to all non-flagged Unknown1 sample group positions on the layout.*

- **Sample type** – If no group number is specified, then it refers to all positions of that type on the layout (e.g., all Unknowns, Blanks, Standards). **Example**: *Unknown refers to all non-flagged data points of Unknown type on the layout.*

The simple sample reference usually refers to the average of non-flagged positions of the type/group. Only for group-enabled functions does it refer to the **list of values of each non-flagged position** that belong to the sample reference. **Example**: *median (Unknown1) calculates the median of all defined on the layout non-flagged Unknown1 position values.*

If a sample reference is enclosed in square brackets (e.g., **[Unknown1]**), it refers to the **list of values of each non-flagged position**. So **[Unknown1]** is actually expanded to the list of results for the non-flagged members of **Unknown1**.

Sample References in Multiple Container Assays

To simplify evaluation and avoid the need to write a different expression for each container, expressions use local group numbering. Thus, the simple **Unknown1** reference always refers to the first **Unknown** sample group on the container being evaluated.

You can refer to a specific position, group, or type defined on specific layout plate with the syntax described below.

Reference	Refers To
A1.2	**A1** position for 2nd container of data
A1.2.3	**A1** position for the 2nd matrix of 3rd container of data
A1..3	**A1** position for 3rd container of data
A1.2.All	The average of all **A1** positions on matrix 2 across all containers
Unknown1.2	**Unknown1** sample group for 2nd container of data
Unknown.2.All	The average of all **Unknown** replicates on matrix 2 across all containers
Unknown1.2.All	The average of all **Unknown1** replicates on matrix 2 across all containers
Unknown1.2.3	**Unknown1** sample group for the 2nd matrix of 3rd container of data
Unknown1..3	**Unknown1** sample data for 3rd container of data

Matrix Position References

This operand comes in handy for accessing the data on the specified matrix at the position the expression is being evaluated for. It is relevant for endpoint data only; however, it is useful for configuring how the XY (e.g., kinetic or spectral) data is treated in analysis. To see further details and configuration examples, please refer to the Matrix Position Reference subsection.

Transform Output Variables

The Transform Output Variable (TOV) feature enables a transform to expose certain calculated numerical results of the analysis as variables. TOVs can be used to reference specific quantities of interest for further analysis.

For example, if you wanted to validate that the calculated R^2 value from a Standard Curve Fit transform was greater than a specific expected value, you could create a Validation condition that references the R^2 value of the Standard Curve Fit transform. Using this feature you could validate that the R^2 value is > 0.9 with the user being alerted if the fit is below an acceptable threshold.

TOV Definition

Each TOV is a named variable with a numerical value that can be referenced in an expression.

The following evaluated values are produced as TOVs by the Standard Curve Fit transform:

- All valid calculated coefficients
- All non-matrix data (quantification limits and sum of squares)
- All valid calculated goodness measures
- All valid calculated Standard Curve Fit Named Evaluations

> Here "valid" means that result was computed successfully and has a value.

Referencing a TOV

Transform Output Variables can be referenced using expressions for:

- Any transform evaluated after the transform that produced the output variable (only where the later transform supports parameters with expression)
- The Validation expression
- The Evaluation expression
- The Sample Label expression
- The Fit Constraint expression
- The Custom fit expression

Identifying Available TOVs

The available TOVs depend on your configured transforms. For each transform that produces TOVs, the available TOVs are added to the calculation log.

How to: Identify Available TOVs

1. [Launch an assay protocol](#) or [open a results file](#).

2. Add and configure the transforms as required.

3. After the results were calculated, open [Calculation log](#) and scroll down to the end.

4. If the transform produces TOVs, review or copy the available TOVs from the list.

The syntax of the TOV as it appears in the [Calculation log](#) is as follows:

TOV added: TOV{N}.{Name}={Value}

Where:

{N} – the number of transform in the sequence configured via **Transform** tab

{Name} – the name of a specific evaluated value

{Value} – the evaluated result of the TOV

When referenced in expression, a syntax is **TOV{N}.{Name}**, e.g. **TOV1.SE**.

> You can only reference TOVs that originate from transforms that are defined and evaluated prior to where the reference is being made. For example, the **TOV2.R2** could be referenced from the third transform onward, but not from the first. For [Validation](#), [Evaluation](#) and [Sample Labels](#), any TOV can be referenced as these elements are prepared after all transforms are computed.

TOVs and Multiple Plate or Multiplex Analyses

For multiple container assays (such as multiple plate or multiplex) the reference to the TOV refers to the TOV output on the container it is associated with.

For example, **TOV1.SE** would refer to the SE value of the 1st transform on the container being evaluated.

Examples:

Here are examples of TOVs along with descriptions of their behaviour.

TOV	Refers to
TOV1.ULOQ	The ULOQ value of the curve-fit as calculated in the first transform.
TOV3.m	The **m** coefficient value of [Linear Regression](#) curve fit method as calculated in the third transform.
TOV1.SE	The Standard Error value of the curve-fit as calculated in the first transform.
TOV1.SSC	The sum of squares of

TOV	Refers to
TOV2.R2	The R^2 value of the curve-fit as calculated in the second transform.

Matrix Position Reference

A Matrix Position Reference is a convenient way to refer to data in a specific matrix. For example, the expression:

#1 - #2

can be used to compute the differences between the values on the first and second matrix (for all positions on the layout).

> A Matrix Position Reference refers to data at the associated position on a specific matrix (and optionally on a specific container/plate/analyte).

When is a Matrix Position Reference Available?

Matrix Position References are available where the expression being evaluated is associated with a position. Examples of transforms with position associated expressions are:

- Expression by Matrix
- Expression by Matrix (Dual)
- XY Expression
- XY Expression (Dual)
- XY Reduction X at Y
- XY Reduction Y at X

Also, the Range parameters (**X Minimum** and **X Maximum**) of XY transforms are position associated.

In these cases, the expression is evaluated at each and every position. The matrix position reference provides a convenient method for accessing the data on the specified matrix at the position the expression is being evaluated for.

Syntax

A Matrix Position Reference is available in two forms:

- With Matrix Number
- With Matrix Number and Container number (e.g., plate/multiplex analyte)

The matrix number is the 1-based number of the matrix. These are listed in the **Matrices** tab.

Example References

Matrix Position Reference	Refers To
#1	Matrix 1
#1.2	Matrix 1 Container 2

> Matrix Position References are only applicable to endpoint matrices. If you mistakenly refer to a matrix that is not endpoint (e.g., XY, kinetic, or spectral) there will be an error evaluating the expression.

XY Data and Matrix Position Reference Examples

When working with XY data (e.g., kinetic or spectral), it is useful to be able to reference calculation results from the previous matrices for further analyses.

Example 1

For a kinetic data analysis, it might be useful to perform calculations only on data after a peak point has occurred. To achieve this, the first step would be to add an XY Reduction Peak transform that will compute the peak for each plot. To do this, use the default settings of this transform and set the result to be the *X* value at the peak for each position.

At this point we have two matrices, one Raw and one Calculated. The matrices (and their numbers) can be found under the **Assay Properties | Matrices** tab, e.g.,

Assay Properties

Transforms   Matrices

⊿····Raw (1)
⋮   ⋮····1. Raw
⊿····Calculated (1)
⋮   ⋮····2. Peak
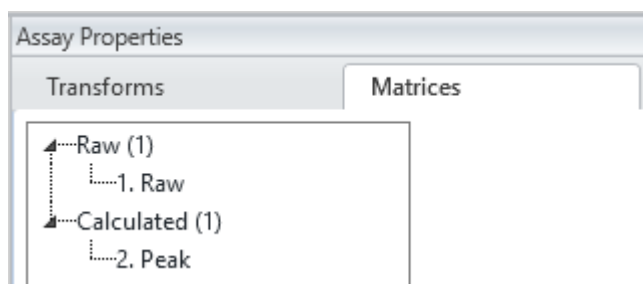
The next step would be to add another transform that uses the computed value of each sample. For example, add the XY Reduction Total transform. This simply calculates the total of the measurements. To configure the transform to only consider measurements from the peak set the **X Minimum** range option to:

#2

I.e.:
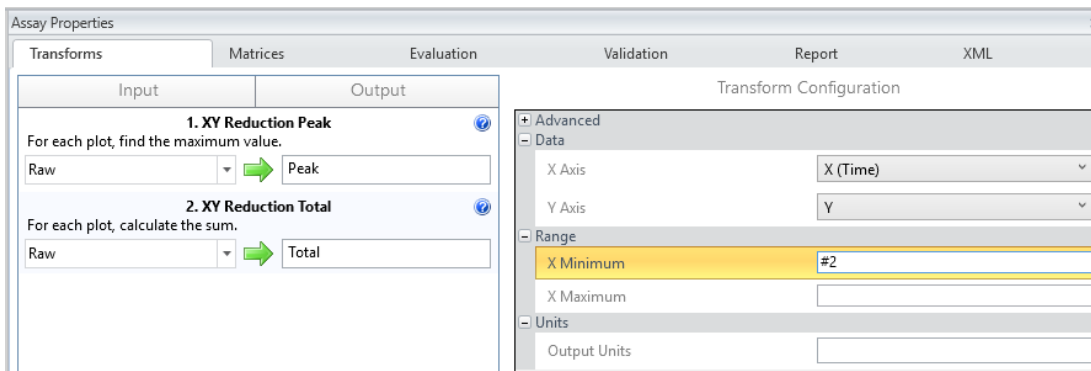
With this setting, the transforms will compute the total of the measurements for each sample but only from the peak point, e.g.:

Total is 21.2183
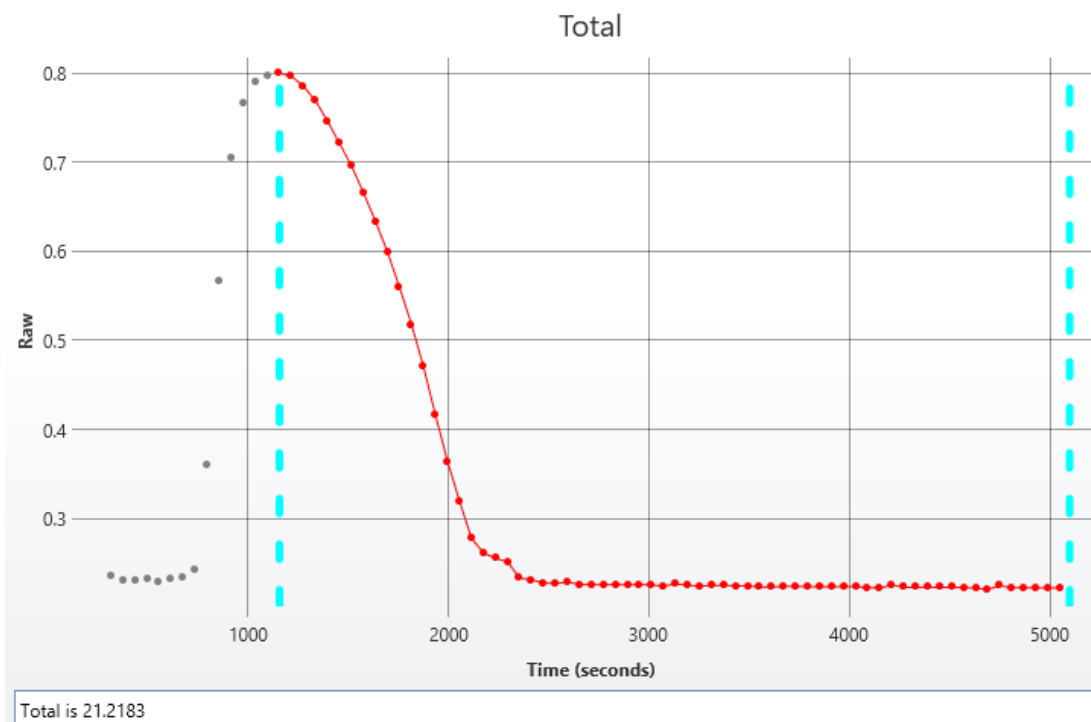
Example 2

In another kinetic example, the time taken to reach half peak can be computed. In this case:

1. The XY Reduction Peak can be used to compute the peak, this time with the result being set to the Y value of the peak.

2. The Expression by Matrix transform is used to compute the Y value at half of the peak.

3. The XY Reduction X at Y transform can be used to calculate the time (X) when the half-peak (Y) is reached. In this case the Matrix Position Reference #3 is used to reference matrix 3 (the half-peak) as the Cursor for the calculation.

I.e.:
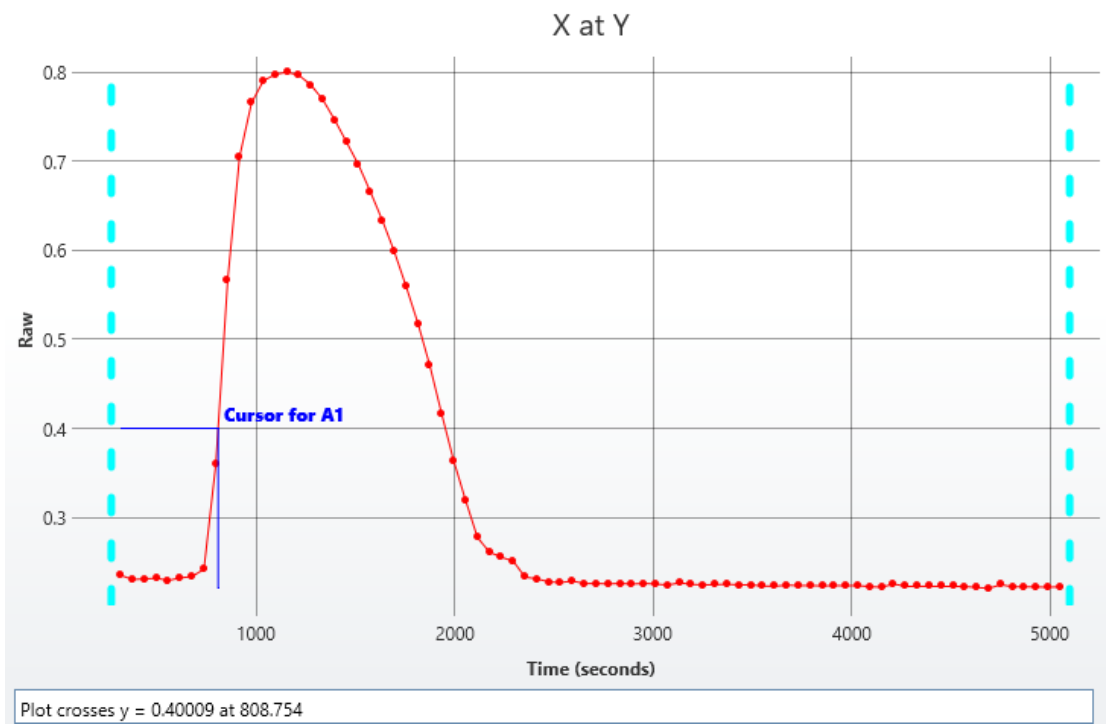
Functions

This section provides reference information for various functions available to perform calculations on data.

Syntax

These take the form of a function name followed by a comma-separated list of values in brackets. The function data can refer to any operand type.

1. If the function refers to a single operand of any type:

   function (operand)

2. If the function refers to a list of operands:

   - If operands do NOT include sample references:

     function (operand1,operand2, ..., operandN)

   - If operands include ONLY sample references:

     function ([operand1,operand2, ..., operandN])

   - If operands include any operand type AND sample references:

     function ([samplereference1,samplereference2, ..., samplereferenceN], operand1, operand2, ..., operandN)

3. If the function requires vector operands:

   - **function ({list of Ys}, {list of Xs})**
   - **function({(samplereference1),(samplereference2), ..., (samplereferenceN)}, {operand1,operand2, ..., operandN})**

Calculation Functions

This section describes functions that calculate the result of mathematical operations on an argument or a list of arguments. Calculation functions use general syntax for functions and perform an operation on a list of 1 or more operands. The evaluation of these results in a single numeric value.

The following table lists calculation functions.

Name	Description	Example
abs	Returns the absolute value of argument. The absolute value of a non-negative argument is the argument itself. The absolute value of a negative argument is -1 times the argument.	**abs(-1.23)** =1.23 **abs(1.23)** =1.23
average	Described in group-enabled functions.	**average(1,2)** =1.5 **average(-3,0.25,5)** =0.75
ceiling	Returns argument (**{0}**) rounded up, away from zero, to the nearest multiple of significance (**{1}**). It requires two input parameters with the following syntax **ceiling({0}, {1})**, where: • **{0}** is the value you want to round off. • **{1}** is the multiple to which you want to round. Regardless of the sign of a **{0}**, a value is rounded up when adjusted away from zero. If the **{0}** is an exact multiple of **{1}**, no rounding occurs.	**ceiling(1.23, 0.05)** =1.25 **ceiling(-1.23, 0.05)** =-1.2
count	Described in group-enabled functions.	**count(1,2,3)** =3 **count(-2.44,2,5,98,6)** =5
exp	Returns **e** raised to the power of the given number. Syntax: **exp({0})** where: **{0}** is the exponent applied to the base **e**. It accepts only a single argument. For example, the system accepts **exp**	**exp(2)** =7.389056... **exp(-1)** =0.367879...

Name	Description	Example
	(A1+A2) and rejects **exp(A1,A2)**.	
floor	Returns argument (**{0}**) rounded down, toward zero, to the nearest multiple of significance (**{1}**). It requires two input parameters with the following syntax **floor({0}, {1})**, where: • **{0}** is the value you want to round off. • **{1}** is the multiple to which you want to round. Regardless of the sign of a **{0}**, a value is rounded down when adjusted away from zero. If the **{0}** is an exact multiple of **{1}**, no rounding occurs.	**floor(1.23, 0.05)** =1.2 **floor(-1.23, 0.05)** =-1.25
intercept	Calculates the point at which the least square's fit line will intersect the Y axis by using existing *x*-values and *y*-values. The intercept point is based on a best fit regression line plotted through the known *x*-values (independent set of arguments) and known *y*-values (dependent set of arguments). ⓘ Requires a vector of arguments as parameters. The number of *Y* and *X* operands is required to be the same and greater than 2. In other words, {*y1,y2,y3*} and {*x1,x2,x3*}; {*y1,y2, ... yN*} and {*x1, x2, ... xN*}.	**intercept({1,2}, {3,2})** =4 **intercept({-2.44,2,5,98,6}, {0.1,0.2,0.3,0.4,0.5})** =-12.152
ln	Returns the natural logarithm of an argument. Natural logarithms are based on the constant e (2.718281828459...). The argument is supposed to be positive. ⓘ **ln** is the inverse of the **exp** function.	**ln(10)** =2.302585... **ln(1.23)** =0.207014...
log	Calculates the logarithm of an argument to the base that you specify. This uses the following syntax with two input parameters: **log({0}, {1})** where: **{0}** is the positive real numerical value or [expression](#) for which you want the logarithm. **{1}** is the base of the logarithm. If the base is omitted, it is assumed to be 10.	**log(2,4)** =0.5 **log(1.23,2)** =0.298658...
log10	Returns the base-10 logarithm of a positive argument.	**log10(1.23)** =0.089905... **log10(0.1)**

Name	Description	Example
		=-1
max	Returns the largest value in a set of values. To determine the maximum of positions in a sample group or sample type, type in the argument it square brackets (i.e., **max ([Unknown1])**). Otherwise the reference is treated as the average of positions.	**max(1,2,3)** =3 **max(-2.44,2,5,98,6)** =98
median	Described in group-enabled functions.	**median(1,2,3,4)** =2.5 **median(-2.44,2,5,98,6)** =5
min	Inversely to **max** function, returns the smallest number in a set of values. All requirements of the **max** apply to **min** too.	**min(1,2,3)** =1 **min(-2.44,2,5,98,6)** =-2.44
mod	Returns the remainder after the argument is divided by a divisor. The result has the same sign as the divisor. Syntax: **mod({0}, {1})** where: **{0}** is the argument for which you want to find the remainder. **{1}** is the value by which you want to divide the **{0}**.	**mod(2,0.3)** =0.2 **mod(1.23,5)** =1.23
pow	Returns the result of a number raised to a power. Syntax: **power({0}, {1})** where: **{0}** is the base argument. **{1}** is the exponent to which the **{0}** is raised.	**power(2,3)** =8 **power(1.23,5)** =2.815305...
slope	Returns the slope of the linear regression line through data points in *X*s and *Y*s. The slope is the vertical distance divided by the horizontal distance between any two points on the line, which is the rate of change along the regression line. Requires a vector of arguments as parameters.	**slope({1,2},{3,2})** =-1 **slope({-2.44,2,5,98,6}, {0.1,0.2,0.3,0.4,0.5})** =112.88

Name	Description	Example
	The number of *Y* and *X* operands is required to be the same and greater than 2 (i.e., {*y1,y2,y3*} and {*x1,x2,x3*}; {*y1,y2, … yN*} and {*x1, x2, … xN*}).	
stdev	The same as **sd** in group-enabled functions.	**stdev(1,2,3)** =1 **stdev(-2.44,2,5,98,6)** =42.772359…
sum	Described in group-enabled functions.	**sum(1,2,3)** =6 **sum(-2.44,2,5,98,6)** =108.56
var	Described in group-enabled functions.	**var(1,2,6)** =7 **var(-2.44,2,5,98,6)** =1829.47472

Conditional Functions

This section describes Boolean logical and conditional operations.

> As with all expression elements, these functions can be used in conjunction with other expression operands, operators and functions. Where a sample reference is used as an input parameter to a conditional function, it refers to the average of the non-flagged data points.

Boolean Logical Functions

These are particularly useful for defining validation, flagging, and sample label conditions.

Name	Description	Example
and	*Syntax:* **and(a,b)** Tests a number of input conditions and returns 1 if all of the conditions evaluate to true, or 0 otherwise.	**and(Unknown>0,Unknown<10)** Tests if the mean of the Unknowns is within the range of 0 to 10.
or	*Syntax:* **or(a,b)** Tests a number of input conditions and returns 1 if any of the conditions evaluate to true, or 0 otherwise.	**or(x<5,x>10)** Tests if the value of x is less than 5 or greater than 10.
not	*Syntax:* **not(a)** Returns a logical value that is the opposite of an input logical value or expression (i.e., returns 0 if the supplied argument is true and returns 1 if the supplied argument is false).	**not(Blank>1)** Tests if the mean of the Blanks is less than or equal to 1.

Conditional Function

Name	Description	Example
ifthenelse	*Syntax:* **ifthenelse(1,2,3)** Requires 3 parameters: 1. The Boolean condition to evaluate (which will evaluate to true or false). 2. The result if the condition is true. 3. The result if the condition is false. If the value of the first condition is true, then the result is the second argument otherwise the third.	**ifthenelse (Unknown1>10,10,Unknown1)** If the value of **Unknown1** is greater than 10 then the result is 10, otherwise the value equals **Unknown1**'s value. **ifthenelse(PosReplicate ()=1,x*0.5,x*0.125)**

Name	Description	Example
	When using the Expression by Matrix transform to define a calculation to perform on all samples, **ifthenelse** can be used to concisely define different calculations to perform on different sample types or replicates by Position Functions to query the sample at each position.	Where the sample is the first replicate the value is multiplied by a factor of 1/2 and other replicates by 1/8. (Here the position function **PosReplicate()** is used to reference the replicate number.)

Group-Enabled Functions

This section describes functions that operate on list of values with an open-ended length.

Where a sample reference is used as a parameter to a group-enabled function, any sample group/type reference inside a Group-Enabled Function's parameter list refers to the non-flagged values of that group/type.

These functions are specified with the following syntaxes:

- **function (Sample type or group reference)** – If there is 1 input sample group/type reference

- **function ([Sample type or group reference, Sample type or group reference, ... , Sample type or group reference])** – If there are 2 or more input sample group/type references.

> When there are 2 or more sample group/type references inside a parameter list, insert each group reference inside square brackets (e.g., **pcv([Standard1, Unknown2]**) to access the values of non-flagged data points.

The following table lists group-enabled functions:

Name	Description	Minimum number of parameters	Example
average	The same as **mean**.	1	**Unknown1** **average ([Unknown, Blank])**
count	Counts the number of non-flagged values.	1	**count(Unknown)**
mean	Calculates the mean of the non-flagged positions.	1	**mean (Unknown1)** **mean ([Unknown, Blank])**
median	Calculates the median of a finite list of non-flagged positions. If there is an even number of arguments in the set, then median calculates the average of the two positions in the middle.	1	**median (Unknown1)** **median ([Standard2, Unknown4])**
pcv	Calculates the percentage coefficient of variance (**%CV**) of a finite list of non-flagged positions.	2	**pcv(Unknown)** **pcv ([Unknown1, Unknown1.2])**

Name	Description	Minimum number of parameters	Example
sd	Calculates the standard deviation of a finite list of non-flagged positions. The standard deviation is a measure of how widely values are dispersed from the average value (the **mean**).	2	**sd (Standard2)** **sd ([Unknown1, Unknown3])**
se	Calculates the standard error of a finite list of non-flagged positions.	2	**se (Unknown1)** Calculates the standard error of the **Unknown1** sample replicates. **se ([Unknown, Control])** Calculates the standard error of all**Unknown** and **Control** sample replicates.
sum	Returns the total of all items in the finite list of non-flagged positions.	1	**sum(Unknown)** **sum([Unknown1, Unknown1.2, Unknown1.3])**
var	Calculates the variance of a finite list of non-flagged positions. Variance is a measurement of the spread between positions in a data set. The variance measures how far each number in the set is from the mean.	2	**var(Unknown4)** **var ([Unknown1, Unknown1.2])**

Example

Let's assume that we have some endpoint measurement data for a 12x8 microplate stored in the **Raw** matrix. In this example we would like to find medium value for **Unknown1** and **Unknown2** positions.

The example uses a layout with 16 **Unknown** sample groups, where **Unknown1** at **A1**, **A2** **A3**, **A4**, **A5**, and **A6**, **Unknown2** at **A7**, **A8**, **A9**, **A10**, **A11**, **A12**, etc.). I.e.:

	1	2	3	4	5	6	7	8	9	10	11	12
A	1	1	1	1	1	1	2	2	2	2	2	2
B	3	3	3	3	3	3	4	4	4	4	4	4
C	5	5	5	5	5	5	6	6	6	6	6	6
D	7	7	7	7	7	7	8	8	8	8	8	8
E	9	9	9	9	9	9	10	10	10	10	10	10
F	11	11	11	11	11	11	12	12	12	12	12	12
G	13	13	13	13	13	13	14	14	14	14	14	14
H	15	15	15	15	15	15	16	16	16	16	16	16

⬤ Unknown

Here is some example raw data displayed to 3 decimal places:

Raw

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.364	0.323	2.105	2.124	1.213	1.226	1.159	1.190	0.110	0.116	1.463	1.481
B	0.455	0.441	1.869	1.888	1.203	1.218	1.180	1.206	0.132	0.130	0.786	0.768
C	0.587	0.575	0.474	0.514	1.206	1.239	1.203	1.210	0.113	0.116	0.613	0.626
D	0.806	0.774	0.364	0.378	1.167	1.187	1.211	1.241	0.101	0.106	0.489	0.490
E	1.105	1.065	0.279	0.367	1.215	1.225	1.233	1.262	0.111	0.114	1.579	1.549
F	1.425	1.445	0.378	0.411	1.196	1.206	1.226	1.199	0.109	0.108	0.792	0.775
G	1.791	1.803	0.560	0.597	1.205	1.228	1.308	1.306	0.126	0.127	0.652	0.659
H	0.253	0.270	0.463	0.470	1.214	1.261	0.115	0.116	0.115	0.115	0.490	0.477

We use the expression:

median ([Unknown1, Unknown2])

Unknown1 and **Unknown2** refer to the lists of non-flagged data points for the **Unknown1** and **Unknown2** sample groups. From this list the median is calculated.

The result will be (also displayed here to 3 decimal places):

1.202

Layout Analysis Functions

Layout Analysis Functions are available only if the evaluation is associated with a layout. Otherwise an error message displays.

Position Functions

Position Functions are only available when there is a position associated with the expression being evaluated. They take no input parameters.

The following table lists the position functions:

Name	Description	Example
PosNum	*Syntax:* **PosNum()** Returns the 1-based position number of each position, which is assigned in accordance with position numbering.	**PosNum()**
PosGroup	*Syntax:* **PosGroup()** Returns the 1-based group number of the sample group at the position. For example, if the position contains **Unknown3**, then the group is 3.	**PosGroup()**
PosType	*Syntax:* **PosType()** Returns the sample type number associated with the position.	**PosType()**
PosReplicate	*Syntax:* **PosReplicate()** Returns the 1-based replicate number of the specified in the argument position. Replicate numbers are assigned in accordance with position numbering. This function is based purely on the layout and does not take flagging into account.	**PosReplicate()**

Layout Functions

These functions require an associated layout.

Layout Functions Parameters

Type

Type can be specified as a sample type number or case-insensitive sample type name in quotes, e.g.:

- **LayoutNumGroups**(3)
- **LayoutNumGroups**("Blank")
- **LayoutNumGroups**("blank")
- **LayoutNumGroups**("bLaNK")

> ℹ️ "**Unused**" type is not supported.

ContainerNum

Where a layout function accepts the **ContainerNum** argument:

- If **ContainerNum** is 0 (or not specified), then the function will be evaluated for all containers in the layout.
- If **ContainerNum** is specified (as a 1-based container number), then only the layout for that container is considered.

GroupNum

This is the 1-based group number of a sample group.

Layout Evaluation Functions

Layout Evaluation functions are provided to enable sample group references to be more flexible (e.g., for applying calculations on group number references).

Name	Description	Example
GroupValue	*Syntax:* **GroupValue(Type,GroupNum)** Returns the value of the sample group with the specified type and group number.	**GroupValue ("Unknown",1)**

> 📝 The **GroupValue** function can be used in conjunction with **PosGroup()** to obtain the group of a specified type that matches the group number of the position being evaluated. For example, if you want to reference the **Unknown** group that has the same number as the current position, use: **GroupValue("Unknown", PosGroup())**

The following table lists the layout functions:

Name	Description	Example
LayoutWidth	*Syntax:* **LayoutWidth()**	**LayoutWidth()**

Name	Description	Example
	The number of columns in the layout (note, this is same value for all containers).	
LayoutHeight	*Syntax:* **LayoutHeight()** The number of rows in the layout (note, this is same value for all containers).	**LayoutHeight()** Is useful for specifying column-based expressions.
LayoutPositions	*Syntax:* **LayoutPositions()** The total number of positions (note, this is same value for all containers).	**LayoutPositions()**
LayoutNumContainers	*Syntax:* **LayoutNumContainers()** The number of containers in the layout.	**LayoutNumContainers()**
LayoutNumGroups	*Syntax:* **LayoutNumGroups (Type,ContainerNum)** The number of groups with the specified type.	**LayoutNumGroups ("Blank", 1)**
LayoutCommonNumReplicatesType	*Syntax:* **LayoutCommonNumReplicatesType (Type)** If all groups of the specified Type on all containers have the same number of replicates, then it returns that number. Otherwise -1 is returned.	**LayoutCommonNumReplicatesType ("Unknown")**
LayoutNumPositions	*Syntax:* **LayoutNumPositions (Type,GroupNum,ContainerNum)** The number of positions with the specified type and group	**LayoutNumPositions ("Standard", 3, 1)**

Name	Description	Example
	number.	
LayoutNumUsedPositions	*Syntax:* **LayoutNumUsedPositions(ContainerNum)** The number of positions that have a type that is not unused (i.e., Type ≠ 1).	**LayoutNumUsedPositions (2)**
LayoutNumGroupsAcrossContainers	*Syntax:* **LayoutNumGroupsAcrossContainers (Type)** Determines the number of groups of the specified Type parameter on each of the containers. If this is the same number on every container (including 0), then this value is returned. Otherwise -1 is returned.	**LayoutNumGroupsAcrossContainers ("Unknown")**

Probability Functions

This section describes syntax and usage of probability functions in MyAssays Desktop.

Overview

The probability functions can be used to examine variability between two sets of data.

Syntax and Parameters

Name	Description	Example
f.dist	Evaluates the value of F probability distribution (degrees of diversity) for two data sets. Requires 3 parameters: 1. **x** – Positive numerical value at which to evaluate the function 2. **df1** – An integer numerical value or an expression (with integer numeric result), that specifies numerator degrees of freedom. 3. **df2** – An integer numerical value or an expression (with integer numeric result), that specifies denominator degrees of freedom.	**f.dist(5.7,5,10)**
f.inv	Returns the inverse of the F probability distribution (the inverse of the **fdist**) for a given probability. Requires 3 parameters: 1. **p** – A probability (between 0 and 1) at which to evaluate the inverse of the **fdist**. 2. **df1** – An integer numerical value or an expression (with integer numeric result), that specifies numerator degrees of freedom. 3. **df2** – An integer numerical value or an expression (with integer numeric result), that specifies denominator degrees of freedom.	**f.inv(0.9,6,96)**

Typically, when specifying degrees of freedom with an expression, you may find the Layout Analysis Functions in conjunction with **count function** useful to refer to the number of non-flagged positions of a certain sample type.

Example Expressions

Expression	Evaluated to
finv(0.95,2,10)	A numerical value for which the F probability equals 0.95 and degrees of freedom specified as 2 and 10.
finv(0.95,LayoutNumGroups("Standard"),count([Standard,Unknown]))	A numerical value for which the F probability equals 0.95 and degrees of freedom specified as the number of **Standards** and the number of all non-flagged **Standard** and **Unknown** samples' replicates.
fdist(0.5667,2,10)	The F distribution value for 0.5667 and degrees of freedom specified as 2 and 10.
fdist(0.5667,count([Standard,Test]),LayoutNumGroups("Standard"))	The F distribution value for 0.5667. The degrees of freedom here are specified as the number of all non-flagged **Standard** and **Unknown** samples' replicates and the number of **Standards**.
fdist(LayoutCommonNumReplicatesType("Standard")*(tov1.ssr),LayoutNumGroups("Standard"),count([Standard,Unknown]))	The F distribution value for the number of **Standard** replicates multiplied by the Sum of Squares TOV from the first configured transform. The degrees of freedom are specified as number of **Standards** and the number of all non-flagged **Standard** and **Unknown** samples' replicates.

Evaluating Z Factor/Z Prime (Z')

Z Factor/Z Prime (Z') (as defined by: *Zhang JH, Chung TD, Oldenburg KR, A Simple Statistical Parameter for Use in Evaluation and Validation of High Throughput Screening Assays. J Biomol Screen. 1999;4(2):67-73*) is used to assess quality of:

- HTS assay

- Population of tested sample compounds

The commonly used equation is

$$Z = 1 - \frac{3SD \ of \ sample + 3SD \ of \ control}{|mean \ of \ sample - mean \ of \ control|}$$

It represents how well separated the positive and negative controls are, and indicates whether an assay is likely to contain false positives/negatives.

This statistic can be added to an assay using the Evaluation option with the expression:

1-((3*sd(Neg Control) + 3*sd(Pos Control))/abs(Neg Control-Pos Control))

Where **Neg Control** and **Pos Control** are the sample/control groups to test.

Integration with Readers

VICTOR Manager Integration with MyAssays Desktop

Overview

This topic describes how to configure the PerkinElmer VICTOR manager software to automatically launch a MyAssays Desktop protocol for analysis of the measured data.

With this configuration in place, data analysis can be performed automatically as soon as VICTOR measurements are completed with little or no user intervention is required. In addition, results can be automatically saved/printed/exported as required to provide a seamless integrated measurement and analysis solution.

Terminology

Between the separate VICTOR manager software and MyAssays Desktop software, there is some overlap in terms used. Both systems use the term "protocol", for clarity in this section we refer to a VICTOR protocol as the *VICTOR measurements protocol* and a MyAssays Desktop protocol as the *MyAssays Desktop protocol.*

Integration Workflow

The general workflow is to:

1. Use the VICTOR manager software to create and configure the required measurement settings (the VICTOR measurements protocol).

2. Export the measurement data to a TXT file.

3. Create a MyAssays Desktop protocol to perform the required analysis.

4. Configure the VICTOR measurements protocol to launch the MyAssays Desktop along with the MyAssays Desktop protocol when the measurements are completed and provide the path to the exported TXT file (using the command line).

The creation of a VICTOR measurement protocol and a MyAssays Desktop protocol are not covered here. It is assumed that you already have these in place and are ready to integrate them.

MyAssays Analysis Launcher

MyAssays Desktop data analysis can be initiated from the command line. MyAssays Desktop offers many options and settings for complete control over the data analysis process.

Writing command line arguments is typically a complicated error-prone process. To simplify this task, MyAssays Desktop includes the MyAssays Analysis Launcher tool. This provides a point-and-click user interface for building a command line to launch MyAssays Desktop analysis. The resulting command line argument is displayed within the tool and can be easily copied and pasted into other applications.

To get started, we recommend familiarising yourself with the MyAssays Analysis Launcher tool. This can be found in MyAssays Desktop Explorer under the **Tools** tab. Select the **Analysis Launcher** option to launch it.

Using Analysis Launcher

To use Analysis Launcher, select a MyAssays Desktop protocol file to launch and use the user interface to configure the operations required.

If you have a compatible VICTOR TXT file to hand, use the **Import Measurements** option and select the file.

If you would like the results to be calculated automatically, check the **Calculate** option.

Notice, in the lower part of the Analysis Launcher the tool that as settings are made the command line argument is updated accordingly.
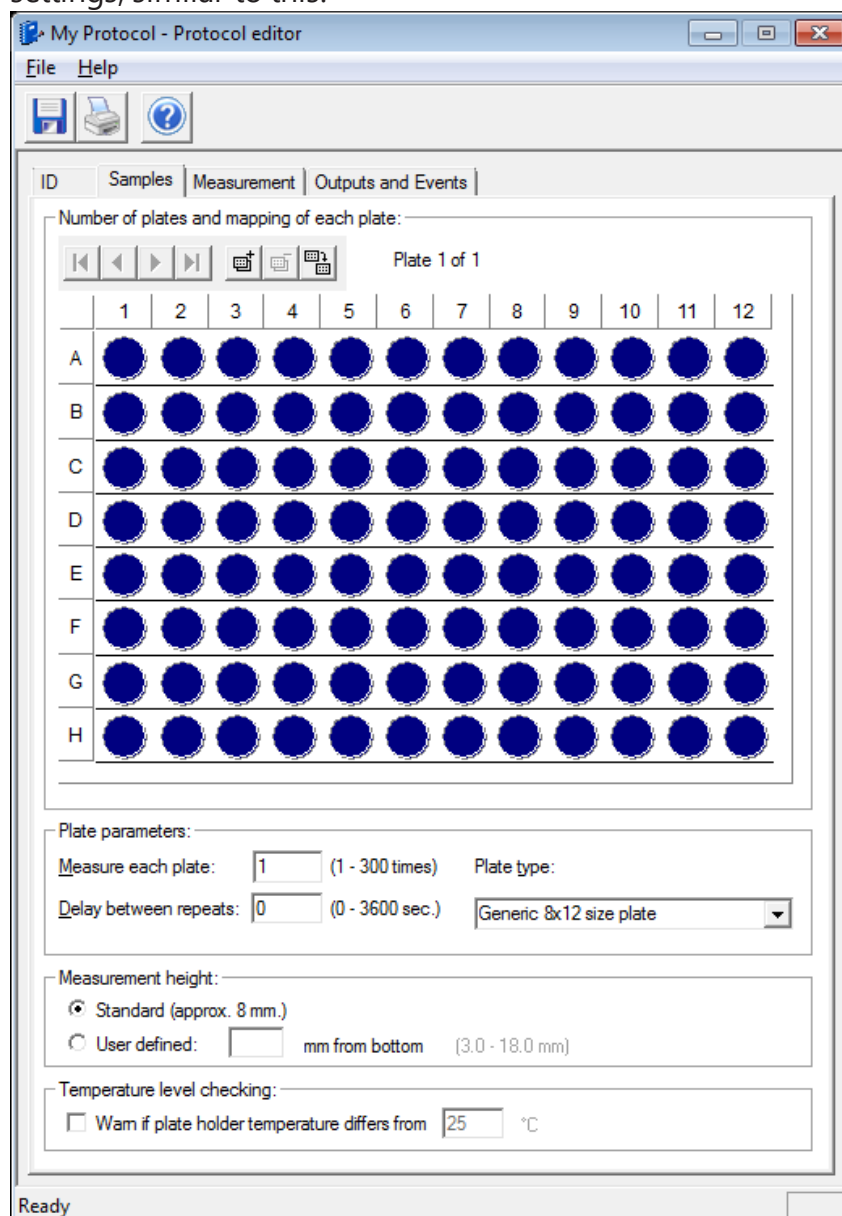
To launch the analysis press **Launch**.

Example Integration

This section describes how to modify a VICTOR measurement protocol to launch MyAssays Desktop:

1. Launch the PerkinElmer VICTOR Manager Explorer.

2. Select and open the VICTOR measurements protocol to edit.

   You should see a window displaying the measurement configuration settings, similar to this:

3. Select the **Outputs and Events** tab

4. Check the **File output** setting

5. Select **File type** as **Text**

6. Specify the **Filename** as required (this could be made up of the various macros)

    For example, in this configuration the file is output to the Documents folder with a name created from the Protocol Name and Run ID:
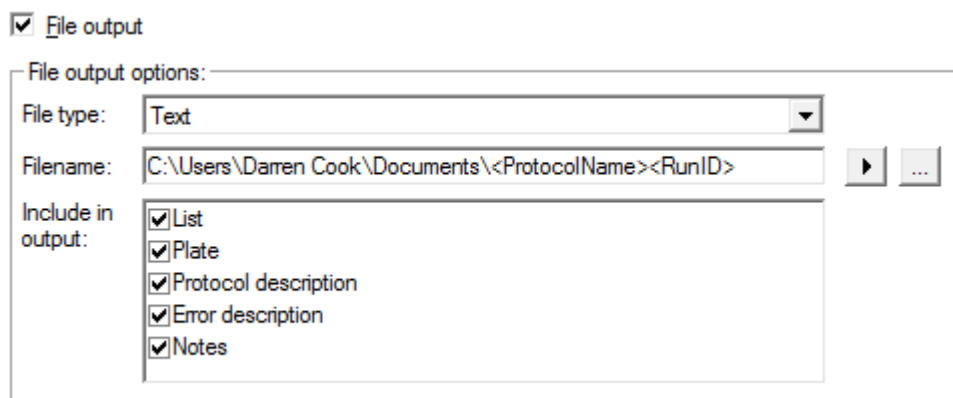
With this setting in place, the results are exported to a text file when measurements are completed. You can try this now (perhaps with an empty plate) to check that the settings are expected.

You can then test importing this data into a MyAssays Desktop protocol "manually" by launching the MyAssays Desktop protocol and using the **Import Wizard** to import the text file.

The final part is to configure the VICTOR measurement protocol's **Assay end system command** setting. This is used to issue the command line.

1. Return to the VICTOR measurements protocol editor.

2. In the first edit box, enter the full path to the MyAssays Desktop Analysis exe, this is typically:

    C:\Program Files\MyAssays\MyAssays Desktop\MyAssays.Desktop.Analysis.exe

    If you have installed MyAssays Desktop to another location, then you will need to enter the correct path. If you are not, use the MyAssays Analysis Launcher application and inspect the command line:
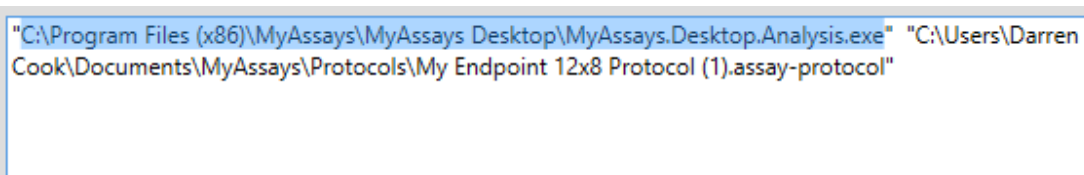
3. In the second edit box, enter the MyAssays Desktop protocol filename to use followed by:

    /import-raw:<OutputFileName>

This setting passes the filename generated by the VICTOR manager software to MyAssays Desktop so that the new measurement data file can be imported. The **<OutputFileName>** is a macro used by VICTOR manager software to refer to the new measurement data file.

For example, to use the MyAssays Desktop protocol: **My Endpoint 12x8 Protocol (1)** in my documents folder I use:

"C:\Users\Darren Cook\Documents\MyAssays\Protocols\My Endpoint 12x8 Protocol (1).assay-protocol" /import-raw:<OutputFileName>

This would appear in the VICTOR measurements protocol editor as follows:
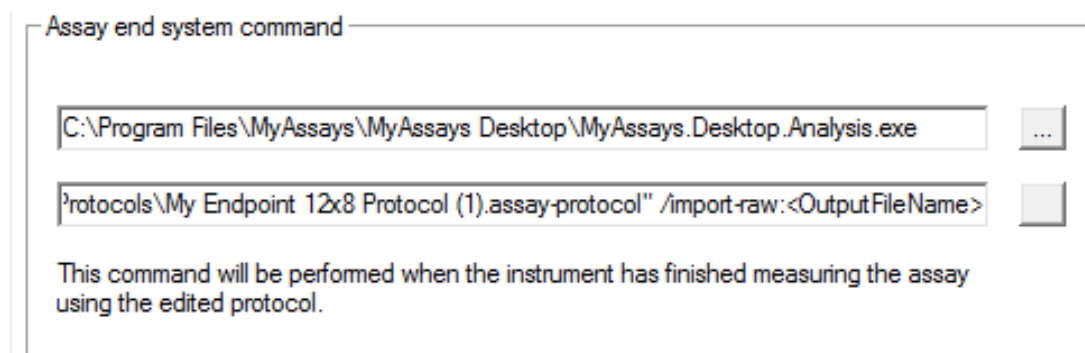
4. Save these changes to the VICTOR measurements protocol.

Similarly, if you are not sure, use the MyAssays Analysis Launcher application and inspect the command line.

With this setting in place, the results will be exported to a text file when measurements are completed and then imported into MyAssays Desktop. You can try this now (perhaps with an empty plate) to check that the integration is working.

With these basic settings, the calculations are automatically performed. This gives the user an opportunity to review the settings and perhaps make some changes (such as a different layout or marking outliers to exclude) before pressing the **Calculate** button.

If you would like calculations to be performed automatically, add:

/calc

To the end of the command line argument in the VICTOR **Assay end system command**, e.g.:

You can also add further command line arguments to automatically print, export, etc. Use the Analysis Launcher tool or refer to the Command Line Arguments section for more information.

EnVision Integration

The EnVision manager software can output data in many possible export configurations.

If using EnVision Manager v14.xxx or later, then select **Export format** option as **CSV for MyAssays Desktop** option:

To configure earlier EnVision Manager versions to output data in a format compatible with MyAssays Desktop EnVision import scripts, use the default CSV **Export format: Plate 2 (with well headers)** option, e.g.:

| File | Print | Event |

☑ File output

☐ Open file in Excel after assay

File name:

```
<DefaultDataFolder>\<Date>\<ProtocolName>_<AssayID>.csv
```
[...]

Export format:

Plate 2 (with well headers) ▼

Assay information to include ─────────────

☑ Basic assay information

☑ Notifications

Full protocol information ▼

Place at end of output ▼

Plate information to include ─────────────

☑ Plate information

☑ Background information

Place at beginning of plate ▼

Other options ─────────────

☐ Show picture

☐ Each plate to a separate file

☐ Don't add plate number to the file name

Use system list separator ▼ [,]

Plates

☐ No selectable plates

Calculation Log

The Calculation Log stores all details of the calculations performed.

How to: Open Calculation Log

1. Launch an assay protocol or open a results file.

2. Press the **Logs** button to open **Logs** panel with **Calculation Log** tab.

3. Review as required.

Logged Calculations

The content included in the Calculation Log depends on the configuration of each of the following settings:

- **Transforms** – Run and results of each transform (in the sequence they are configured).
- **Evaluation** – Run and results of the computed evaluations.
- **Validation** – Run and results (pass or fail) of the computed validations.
- **Transform Output Variables** – Non-matrix calculations produced by the transforms

Additionally, it might also produce fit transform context parameters and a variety of transform-specific calculations. For example, if a Standard Curve Fit transform is configured, then the log also contains coefficient constraints, weighting infinity resolution, custom flag label for Standard Curve Fit and more.

Level of Details

The Calculation Log can be viewed in the following modes:

- **Detailed** – Lists all logged calculations in great depth (e.g., transform calculations for each position)
- **Summary** – Contains overview of performed calculations
- **None** – No data is displayed in the log

By default, the log is displayed in **Detailed** mode.

> ℹ️ To change the level of details, in the XML editor, add the **CalculationLog** element as a child of the **MatrixTransformAnalysis** and select preferable view mode.

Saving Log to a File

Press the **Save Log** button to save the log for the current assay run to your preferred destination in a .TXT file.

Error Log

If any errors occurred during an assay run, they are stored and described in the Error log in similar to the Calculation Log level of details. To view the log, select **Error Log** subpane of **Logs** pane.

Command Line Arguments

MAA can be launched directly from the command line with optional specific arguments.

Syntax

>"C:*MyAssays Installation Root path*\MyAssays Desktop\MyAs-says.Desktop.Analysis.exe" "C:*MyAssays Folder Root path*\MyAssays\Protocols*Protocol File Name*.assay-protocol" /argument1 /argument2 etc.

> 📝 Command line arguments are only relevant when the MAA is opened with an .assay-protocol file and can be specified in any order.

The following table lists optional command line arguments:

Argument	Description	Example
/calc	Performs specified calculations automatically (if **/import-raw** is used, then calculations are performed using the imported raw data).	/calc
/calc-log:{0}	Specifies a file to save calculation progress log.	/calc-log:"Destination file path"
/error-log:{0}	Specifies a file path to save calculation errors log.	/error-log:"Destination file path"
/exit	Closes MAA after calculations are completed/printed.	/exit
/export-report:{0}:{1}	Specifies the export options. {0} is either: • XLSX, PDF, XML (file export type). • XSLT. Requires additional arguments to specify output filename extension and path to XSLT 1.0 file. The exported file will be saved under \MyAssays\Reports\ folder. 📝 If using a Custom Export Plug-In, then configure the protocol to export using	/export-report:pdf /export-report:xslt1:csv:"XSLT file path"

Argument	Description	Example
	the Export Wizard and save the export settings to the protocol (rather than using **/export-report**).	
	The optional {1} specifies whether to launch the exported file with its default associated application. If it is specified, the file will not launch.	**:nolaunch** **/export-report:xml:nolaunch**
/import-con-tainerids:{0}	Specifies a Plate IDs file to use in data analysis. This command-line argument overrides previously configured by **Import Wizard** method.	**/import-con-tainerids:"Data file path"**
/import-raw:{0}	Specifies the raw data file to be used for analysis.	**/import-raw:"Raw data file path"**
/import-sampleids:{0}	Specifies a Sample IDs file to use in data analysis. This command-line argument overrides previously configured by **Import Wizard** method.	**/import-con-tainerids:"Sample IDs file path"**
/import-tweaks:{0}:{1}	Specifies a protocol tweaks file that defines changes to the protocol configuration. • {0} - full path to the protocol tweaks file.	**/import-tweak-s:"Protocol tweak file path"**
	The optional {1} specifies the save setting. If it is specified, the protocol file will be saved with tweaks automatically. If the **Save changes to**	**/import-tweak-s:"Protocol tweak file path":saveprotocol**

Argument	Description	Example
	protocol option is selected (under **File | Save in the Analysis application**), then the changes will be saved to the protocol irrespective of this option setting (e.g., when calculations are performed).	
/noappwin	If this is specified, then the application window is NOT displayed.	**/noappwin**
/print	Automatically prints the report (if the calculations succeeded) with the default printer (if available).	**/print**
/tab:{0}	Allows you to select the tab MAA starts on. {0} is zero-based **tab number**: *Overview = 0; Measurements =1; Microplate = 2; Sample IDs = 3; Results = 4*. If this option is not specified, then the first tab will open.	**/tab:1**

The maximum command line argument is 255 characters. If the command line argument exceeds this length, then a warning message is displayed and the program is exited.

To use long command line arguments, store the command-line argument list in a text file and launch MyAssays.Desktop.Analysis.exe, passing the text file containing the command-line arguments:

MyAssays.Desktop.Analysis.exe @<CommandTxtFile>

Where <**CommandTxtFile**> is the name of the text file to read the command line from.

Examples

>"C:*MyAssays Installation Root path*\\MyAssays Desktop\\MyAssays.Desktop.Analysis.exe" "C:*MyAssays Folder Root path*\\MyAssays\\Protocols*Protocol File Name*.assay-protocol" /import-raw:"Raw data file path" /tab:4 /calc /export-report:xlsx:nolaunch

The selected assay protocol with specified raw data file will be computed. The MAA will start with the **Results tab** and the XLSX report will be saved (but not automatically launched) under \MyAssays\Reports\ folder.

>**"C:*MyAssays Installation Root path*\MyAssays Desktop\MyAssays.Desktop.Analysis.exe" "C:*MyAssays Folder Root path*\MyAssays\Protocols*Protocol File Name*.assay-protocol" /calc /print /noappwin**

The selected assay protocol with specified raw data file will be computed and the results will be printed out.

These settings can be configured using the Analysis Launcher.

Tweak Files

This section provides information about the protocol tweaks feature. This feature enables an external application to change and apply specific settings of the data analysis.

> This is an advanced feature and misuse of it could cause problems.

Overview

Protocol tweaks are used to define changes to the data analysis parameters of an existing protocol. Any of the assay protocol parameters can be tweaked. This is useful to control the analysis process by other software such as instrument-control software or LIMS.

For example, a LIMS could initiate execution of data analysis and specify customizations (such as standard concentrations or layout); the customisations are merged with the existing protocol, the analysis performed, and results generated with data exported as required.

Protocol tweaks are defined in an XML file and can be applied by launching the protocol and specifying the tweak file to apply.

> Any data analysis parameter can be modified.

Tweak Syntax

A protocol-tweak.xml file is an XML with the following syntax:

<?xml version="1.0" encoding="utf-8"?>

<ProtocolTweaks>

<Tweak XPath="*Full assay setting to tweak path*">*Substitute data*</Tweak>

</ProtocolTweaks>

> You can insert any number of tweaks under <ProtocolTweaks> section.

Using the Feature

To use the protocol tweak feature, it is recommend to developing a protocol-tweak file as follows:

1. Build or obtain an assay-protocol file that meets your data analysis requirements.
2. Identify which analysis parameters should be tweaked.
3. Develop a protocol-tweak.xml file.
4. Test the protocol-tweak.xml file.

With your working protocol-tweak.xml file, use your instrument control or LIMS software to insert relevant content into the protocol-tweak.xml file.

How to: Apply Protocol Tweaks

The easiest way to apply a protocol tweak file is to use the Analysis Launcher. Use this tool to select the protocol and tweak file and launch the application.

The Analysis Launcher tool will also generate and display the command line argument to use. You can use this command line setting to launch the operation yourself (perhaps calling it from another software application that prepares the tweak file's contents). More information about command-line arguments is available here.

When the protocol is launched and tweak file applied, the system will perform various checks on the incorporated tweaks to detect any report any possible configuration errors in common usage scenarios.

If the protocol is successfully tweaked the analysis operations will continue as specified. The actual behaviour will depend on any further command line settings (such as automatic calculation, export, etc.)

> Tweak changes are not automatically saved to the incorporated assay file. A tweaked protocol appears as modified and not saved (with ***** in title). If you save the protocol, the tweaks will be saved to the protocol file.

Installing MyAssays Desktop Alongside WorkOut

MyAssays Desktop can be installed and used alongside an existing WorkOut installation (including WorkOut versions 1.x, 2.x, Plus, and Plus MMD). This is referred to as a Side-By-Side installation.

Certain modules (relating to data import and analysis) are shared between the applications. If you plan to use both WorkOut and MyAssays Desktop on the same system, then please follow these points to avoid known conflict issues:

- Always install MyAssays Desktop **<u>after</u>** installing WorkOut (i.e., do not install, update, patch, or repair a WorkOut installation after installing MyAssays Desktop).
- If WorkOut is uninstalled leaving a MyAssays Desktop installation, then it will be necessary to also uninstall MyAssays Desktop and then reinstall MyAssays Desktop.

If MyAssays Desktop has been installed with a Side-By-Side WorkOut installation, then this will be reported in the **File | Information** tab of MyAssays Desktop Explorer.

Known Issues

If the points above are not followed, then you may see the following problems with MyAssays Desktop:

- Unable to import data
- Unable to produce a valid report (on data calculation)

To resolve these issues, please uninstall MyAssays Desktop and then reinstall MyAssays Desktop to update the shared component files to the newest version.

Referencing MyAssays Desktop

You can refer to MyAssays Desktop in your paper, thesis or publication.

To refer to the data analysis tool include the text:

"MyAssays Desktop *Product Version* from MyAssays Ltd., https://www.myassays.com"

> To obtain current *Product Version* open **Information** screen of **File** tab in the
> MADE. You can either click on the **Copy** icon or enter it manually to your pub-
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Version 2.0, January 2004

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