TECAN.

Instructions for Use for

i-control

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Prior to contacting Tecan for product support, prepare the following information for the best possible technical support (see name plate):

- Model name of your product
- Serial number (SN) of your product
- Software and software version (if applicable)
- Description of the problem and contact person
- Date and time when the problem occurred
- Steps that you have already taken to correct the problem
- Your contact information (phone number, fax number, e-mail address, etc.)

Warnings, Cautions, and Notes

The following types of notices are used in this publication to highlight important information or to warn the user of a potentially dangerous situation:

Note
Gives helpful information.

CAUTION
INDICATES A POSSIBILITY OF INSTRUMENT DAMAGE OR DATA LOSS IF INSTRUCTIONS ARE NOT FOLLOWED.

WARNING
INDICATES THE POSSIBILITY OF SEVERE PERSONAL INJURY, LOSS OF LIFE OR EQUIPMENT DAMAGE IF THE INSTRUCTIONS ARE NOT FOLLOWED.

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WARNING

CAREFULLY READ AND FOLLOW THE INSTRUCTIONS PROVIDED IN THIS MANUAL BEFORE OPERATING THE INSTRUMENT.

Notice

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We appreciate any comments on this publication.

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Copyright Information

About the Instructions for Use

This document describes i-control, which is a software to control **Infinite Series** Tecan microplate readers. It is intended as a reference and instruction for the user.

This manual instructs how to:

- Install the software
- Operate the software

Remarks on Screenshots

Data and parameters displayed in screenshots vary depending on the instrument connected. Details and examples are described in the respective Instructions for Use of the connected instrument.

Table of Contents

1. Introduction

1.1 Area of Application

i-control is an easy-to-use and flexible tool, which gives the user complete control over Tecan readers.

i-control presents the raw data for further use in Excel, offering excellent features for research purposes.

Note
Depending on the instrument connected and the modules equipped, certain i-control features may be disabled or invisible.

1.1.1 i-control Intended Use

The **i-control software** is a reader control and data presentation software that comes with Tecan **Infinite Series readers**. i-control is intended for professional use with these readers according to the instrument's instructions.
i-control is designed for use with Microsoft Excel for data presentation.

1.2 User Profile

1.2.1 Professional User – Administrator Level

The administrator is a person who has suitable technical training and corresponding skills and experiences. If the product is used as intended, the person is able to recognize and avoid dangers.

The administrator has extensive skills and is able to instruct the end user or the routine user in assay protocols in connection with a Tecan product within the bounds of the intended use.

Computer application skills and good English skills are required.

1.2.2 End User or Routine User

The end user or routine user is a person who has suitable technical training and corresponding skills and experiences. If the product is used as intended, the person is able to recognize and avoid dangers.

Computer application skills and good language skills for the respective national language at the installation site and English are required.

1.2.3 Service Technician

The service technician is a person who has suitable technical training and corresponding skills and experiences. If the product needs to be serviced or maintained, the person is able to recognize and avoid dangers.

Computer application skills and good English skills are required

Note
Training dates, their duration and frequency are available at your customer support.

Address and phone number can be found in the Internet:
http://www.tecan.com/customersupport

1.3 Specifications

1.3.1 Hardware Requirements

The following hardware requirements and operating system requirements have to be met to use the i-control software:

	Minimum	Recommended
PC	Windows XP (32-bit): Windows compatible PC with a Pentium compatible processor running at 1 GHz	2 GHz (Dual Core)
	Windows 7 (32- or 64-bit): Windows compatible PC with a Pentium compatible processor running at 1 GHz	2 GHz (Dual Core)
	Windows 8.1 (32- or 64-bit): Windows compatible PC with a Pentium compatible processor running at 1 GHz	2 GHz (Dual Core)
	Windows 10 (32- or 64-bit): Windows compatible PC with a Pentium compatible processor running at 1 GHz	2 GHz (Dual Core)
Operating System	Windows XP (32-bit) – SP3 Edition: Professional	Windows 7 (64-bit) – SP1 Edition: Professional
	Windows 7 (32-bit) – SP1 Windows 7 (64-bit) – SP1 Editions: Professional, Ultimate, Enterprise	
	Windows 8.1 (32-bit) Windows 8.1 (64-bit) Editions: Pro, Enterprise **Windows RT NOT supported!**	
	Windows 10 (32-bit) Windows 10 (64-bit) Editions: Pro, Enterprise	
Memory	Windows XP: 512 MB RAM	1 GB RAM
	Windows 7 (32-bit): 1 GB RAM	2 GB RAM
	Windows 7 (64-bit): 2 GB RAM	4 GB RAM
	Windows 8.1 (32-bit): 1 GB RAM	2 GB RAM
	Windows 8.1 (64-bit): 2 GB RAM	4 GB RAM
	Windows 10 (32-bit): 1 GB RAM	2 GB RAM
	Windows 10 (64-bit): 2 GB RAM	4 GB RAM
Space Requirements	2 GB	6 GB
Monitor	Super VGA Graphics	
Resolution	1024 x 768	1280 x 1024

	Minimum	Recommended
Color Depth	256	
Mouse	Microsoft mouse or compatible pointing device	
Communication	1 x USB 2.0	2 x USB 2.0 1 x RS232 (Serial)
Devices	1 x CD-ROM drive	
	Windows 7: DirectX 9 graphics device with WDDM 1.0 or higher driver	
	Windows 8.1: DirectX 9 graphics device with WDDM driver	
	Windows 10: DirectX 9 graphics device with WDDM driver	
.NET	Microsoft .NET Framework 2.0 **Windows Versions Prior to Windows 8:** The required .NET version is installed automatically alongside any existing versions (2.0 is part of Windows 7 SP1, which is required). **Windows 8.1, Windows 10:** Microsoft.NET Framework 3.5.1 (including 2.0) must first be installed before installing i-control! The user will be prompted to install the required .NET framework, if it is not already present	
Microsoft Excel	2002 2003 2007 2010: Only 32-bit editions supported! Starter editions NOT supported! 2013: Only 32-bit editions supported! Starter editions NOT supported! 2016: Only 32-bit editions supported! Starter editions NOT supported!	2010

1.3.2 Reader Compatibility

The following Tecan readers can be used with i-control:

Instrument Types	Measurement Mode
Infinite M200 Infinite M200 PRO	Fluorescence / Absorbance / Luminescence
Infinite F200	Fluorescence / Absorbance / Luminescence / Fluorescence Polarization
Infinite F200 PRO	Fluorescence / Absorbance / Luminescence / Fluorescence Polarization / AlphaScreen/AlphaLISA
Infinite F500	Fluorescence / Absorbance / Luminescence / Fluorescence Polarization
Infinite M1000	Fluorescence / Absorbance / Luminescence / Fluorescence Polarization
Infinite M1000 PRO	Fluorescence / Absorbance / Luminescence / Fluorescence Polarization / AlphaScreen/AlphaLISA

Note
The Connect stacker can be used together with several instruments in order to measure batches of plates. Please refer to the Connect Instructions for Use for more information.

With the Infinite M1000 and Infinite M1000 PRO instruments, only the built-in stacker can be used.

1.4 Software Installation

CAUTION
YOU MUST HAVE ADMINISTRATIVE RIGHTS TO INSTALL THE SOFTWARE.

CAUTION
INSTALL THE SOFTWARE BEFORE PLUGGING THE INSTRUMENT INTO THE COMPUTER.

The i-control software is installed using the following procedure:

1. Insert the installation CD into the appropriate disk drive or CD ROM drive.
2. Open the Windows Explorer and browse to folder **Software** on the installation CD. Double-click **i-Control <version>.exe** to start the installation procedure.
3. Install all **Setup Prerequisites.**

   Depending on your operating system different prerequisites have to be installed. Click **Next** to continue.
4. A message box indicates that the prerequisites have been installed successfully. Click **OK** to continue.

5 In the course of the installation a series of dialog boxes will appear. Read each one, enter any necessary information and click **Next** to continue. The files are installed and the program icon is created.

6. When the **Installation Complete** dialog box appears, click **Finish** and the i-control program is ready to be used.

1.4.1 Instrument Driver Installation

When connecting a real instrument for the first time to i-control, the corresponding instrument driver has to be installed. Please note that a successful installation of the instrument driver is the prerequisite for working with the connected instrument.

1.5 Starting i-control

i-control can be used either with a connected instrument or in simulation mode.

1.5.1 Connected Instrument

CAUTION
INSTALL THE SOFTWARE BEFORE CONNECTING THE INSTRUMENT TO THE COMPUTER.

Connect the instrument to your computer and switch the instrument on. Start the program by selecting **Programs/Tecan/i-control** from the **Windows Start** menu.

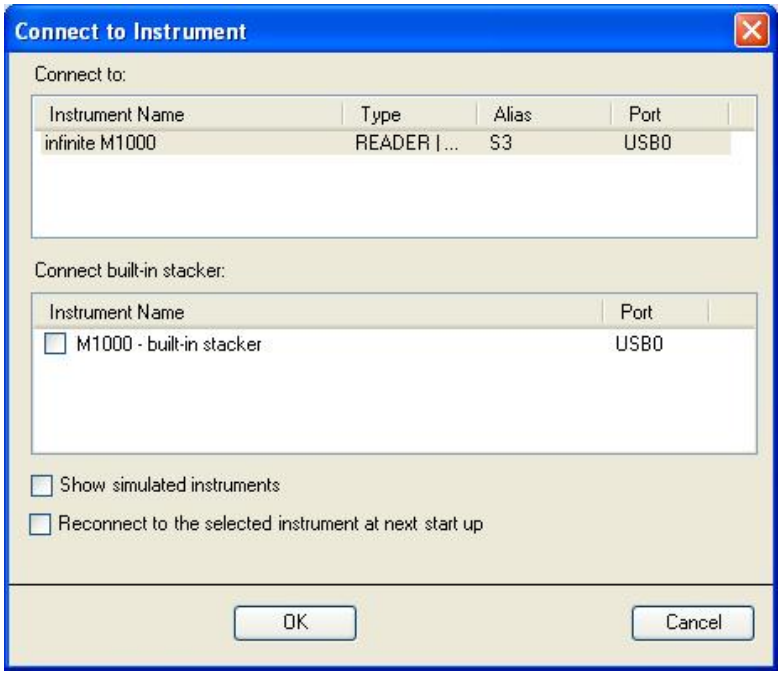
Select **Connect** from the **Instrument** menu or click the connect button and the following dialog box appears:

Example for the **Infinite 200** instrument:

In the **Connect to:** dialog box select the instrument name.

In the **Additionally connect to:** field, select **Connect**, if a **Connect stacker** is connected (for batch processing).

Example for the **Infinite M1000** instrument:



In the **Connect to:** dialog box select the instrument name.

Connect built-in stacker:

With the **Infinite M1000** and **Infinite M1000 PRO** instruments, only the built-in stacker can be used (see screenshot).

Click **OK** to start i-control.

1.5.2 Simulated Instrument

Start the program by selecting **Programs/Tecan/i-control** from the **Windows Start** menu. In the **Connect to Instrument** dialog box, select **Show simulated instruments**; from the **Instrument Name** list, select the demo instrument to connect to.

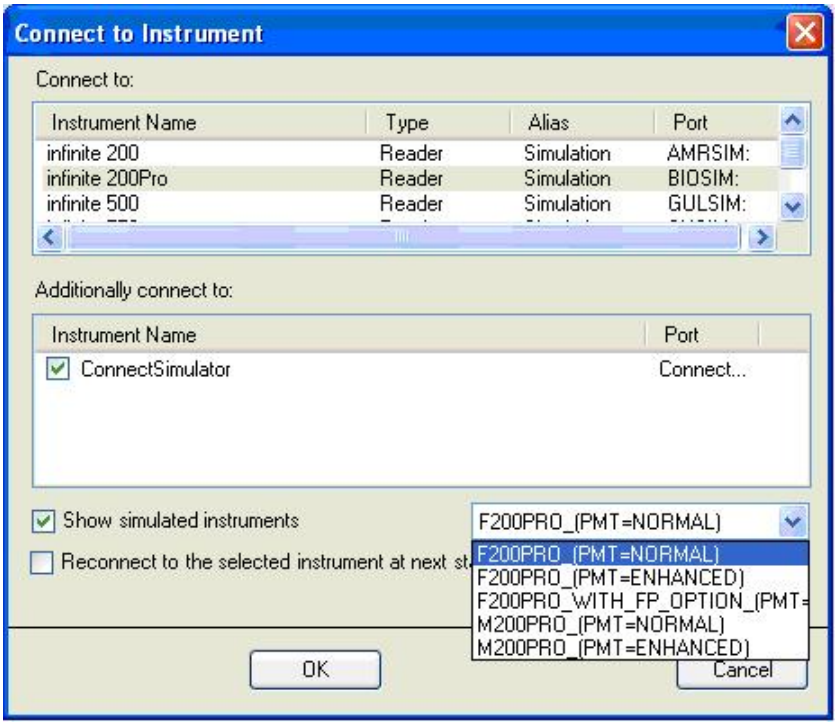
After selecting the simulated instrument, a drop-down list appears offering several options, depending on the instrument selected above.

For the **Infinite 200**, for example, these options are:

- Filter: F200 normal or F200 enhanced or F200 with FP Option
- Monochromator: M200 normal or M200 enhanced

For the **Infinite 200 PRO**, for example, these options are:

- F200PRO_(PMT=NORMAL)
- F200PRO_(PMT=ENHANCED)
- F200PRO_ALPHA
- F200PRO_WITH_FP_OPTION_(PMT=NORMAL)
- M200PRO_(PMT=NORMAL)
- M200PRO_(PMT= ENHANCED)



For the **Infinite F500**, for example, these options are:

- GF500_(PMT=NORMAL)_384
- GF500_(PMT=ENHANCED)_1536/384
- FI.TOP/ABS/HEA/SHK_ONLY_(PMT=Normal)_1536/384
- GF500_WITH_FP_(PMT=NORMAL)_384

For the **Infinite M1000**, for example, these options are:

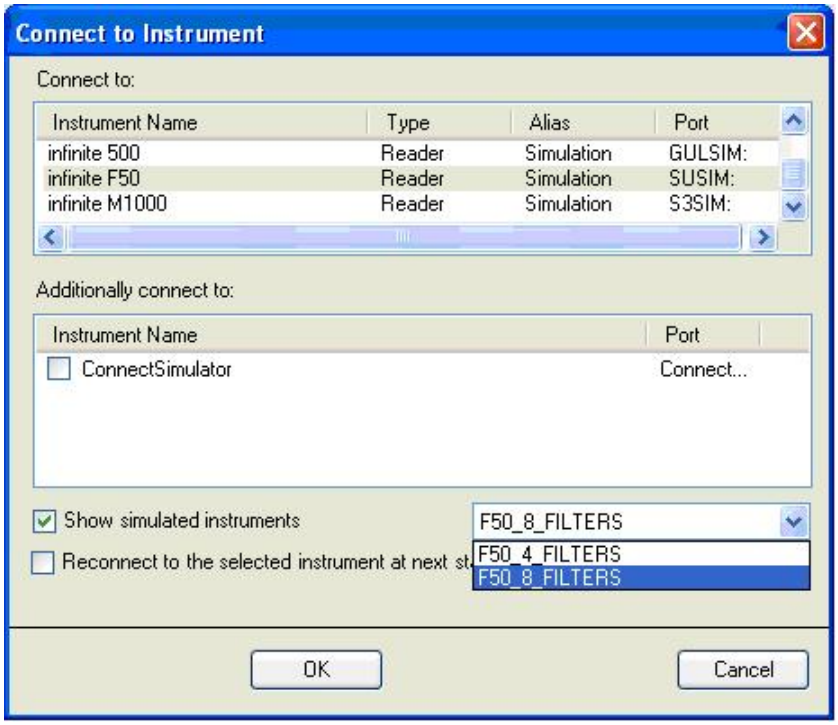
- M1000_384/1536
- M1000_FP_INJ_STACKER
- M1000_384/1536 LCE
- M1000_FP_INJ_STACKER LCE

For the **Infinite M1000 PRO**, for example, these options are:

- M1000PRO_384/1536
- M1000PRO_FP_INJ_STACKER

For the **Infinite F50**, for example, these options are:

- F50PRO_4_FILTERS
- F50PRO_8_FILTERS



Connect built-in stacker:

With the **Infinite M1000** and **Infinite M1000 PRO**, the built-in stacker can be simulated. See selections as shown in the screenshot above.

For a more detailed description on defining parameters for the respective instrument, please refer to the instructions for use for the connected or simulated instruments.

Select **Reconnect to the selected instrument at next start up** in case the same instrument remains attached to one and the same computer.

Click **OK** to start i-control.

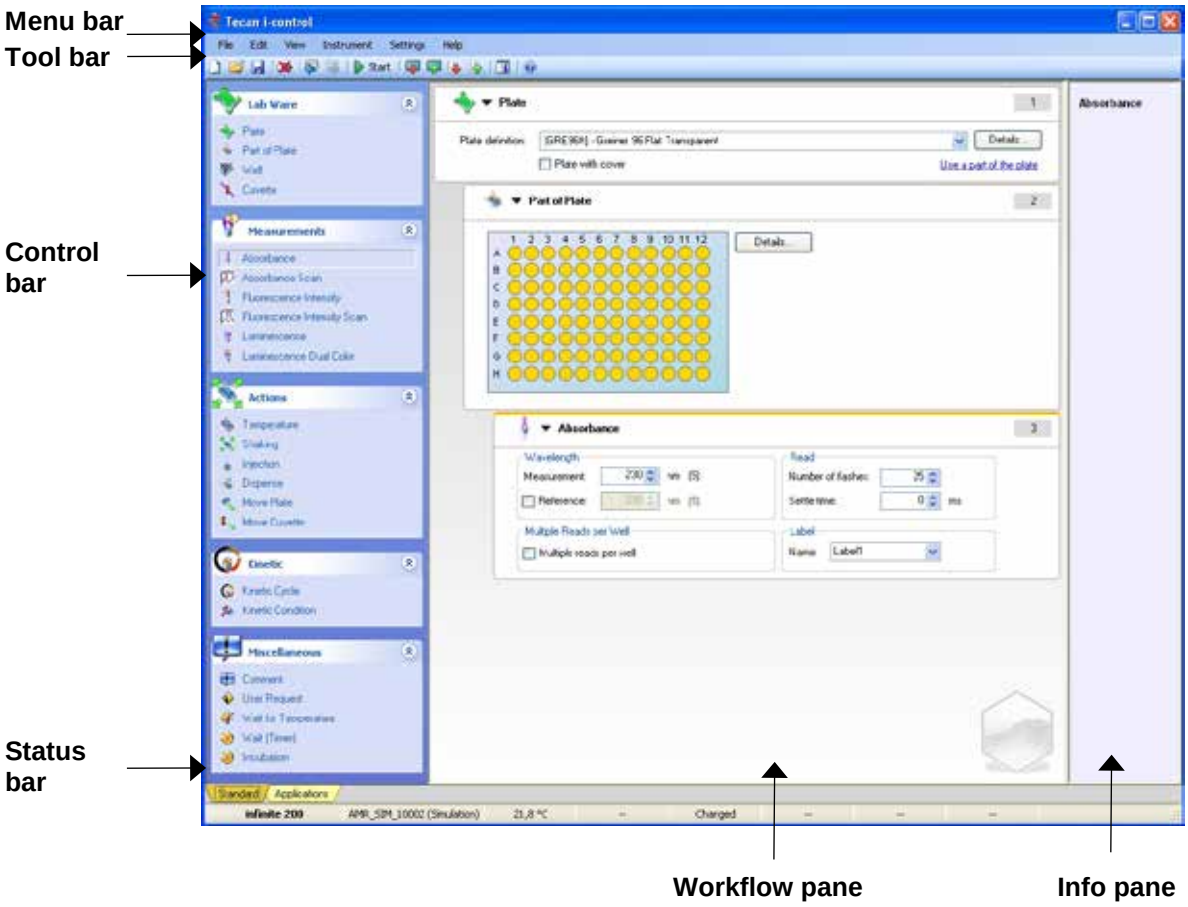
2. Measurement Parameter Editor

2.1 Introduction

The main window of the i-control software is the **Measurement Parameter Editor**, which is used to set up workflows. Each workflow is easily created by dragging and dropping the process steps into a sequence according to the application. The application workflow is then visible to the user in the workflow pane and can be saved for future use. Each process step, that is each program element, can be copied and pasted (menu **Edit – Copy – Paste** or using the Windows standard shortcuts **Ctrl-C, Ctrl-V**) and moved to the desired position in the workflow.

Data can be exported easily to Windows compatible formats (Excel).

Start the software and connect an instrument as described in the previous chapter or select the simulation mode. The i-control main window appears displaying the **Measurement Parameter Editor**:



The **Measurement Parameter Editor** consists of the following items which are described in detail in the subsequent chapters:

· Menu bar	· Status bar
· Tool bar	· Workflow pane
· Control bar	· Info pane

In the left bottom corner of the main window, two tabs appear:

Standard: is displayed for standard applications

Application: is displayed for applications with NanoQuant plates which are currently only available with the **Infinite 200, Infinite 200 PRO, Infinite F500** (DNA quantification only), **Infinite M1000 available for FW 2.0 and higher (Ref 30061442)** and **Infinite M1000 PRO**.

Please consult the Quick Guide for NanoQuant Plates and the respective Instructions for Use of the instrument connected.

2.2 Control Bar

The **Control bar** is divided into five sections. Each section contains program elements used to create an individual workflow. Depending on the instrument connected and the modules installed, these available program elements may vary; e.g. if the instrument is not equipped with an FP module, the FP element is not visible in the measurement section.

Create a workflow either by double-clicking the selected program element or by dragging and dropping it into the workflow pane.

The following program elements are available:

Lab Ware	Plate Part of Plate Well Cuvette (M200 and M200 PRO)
Measurements	Absorbance Absorbance Scan (M200, M200 PRO, M1000 and M1000 PRO) Fluorescence Intensity Fluorescence Intensity Scan (M200, M200 PRO, M1000 and M1000 PRO) Fluorescence Polarization (F200, F200 PRO, F500, M1000 and M1000 PRO) Luminescence Luminescence Dual Color Luminescence Scan (M1000 – available for FW 2.0 and higher - Ref 30061442 and M1000 PRO) AlphaScreen / AlphaLISA (F200 PRO, M1000 PRO)
Actions	Temperature Shaking Injection Dispense Move Plate Move Cuvette (M200, M200 PRO)
Kinetic	Kinetic Cycle Kinetic Condition

Miscellaneous	Comment User Request Wait for Temperature Wait (Time) Incubation

2.2.1 Lab Ware

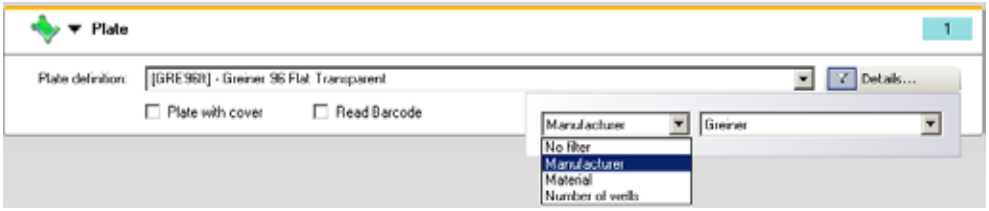
Plate

The **Plate** program element is used to select a plate format from the **Plate definition** drop-down list. Click **Details**... to see further information on the selected plate.

If a plate with cover is used, select the **Plate with cover** checkbox.

The measurement will automatically measure all selected wells of the plate. If you want to measure a specific well or a range of wells, click the link Use a part of the plate in the lower right corner. See **Part of Plate** below.

Under **Details…** it is possible to apply a filter so that only certain plate definition files are shown.

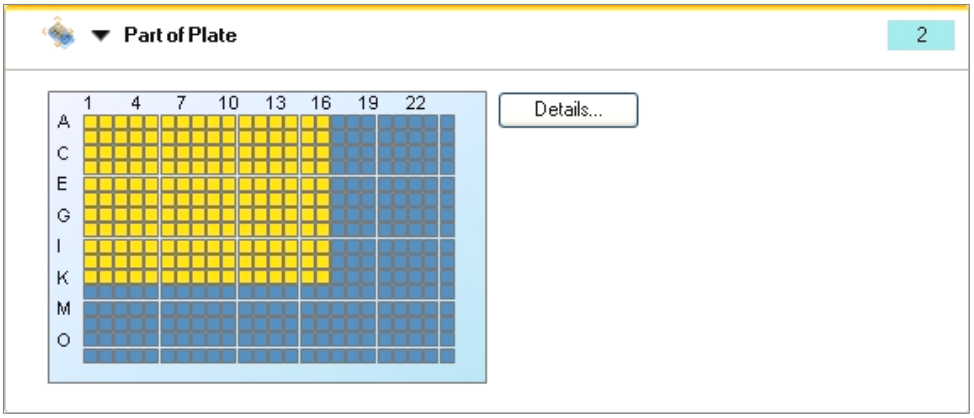


The **Infinite F500**, **M1000** and **M1000 PRO** may optionally be equipped with a barcode scanner. Select the checkbox **Read Barcode** to have the barcode read.

The **Read Barcode** checkbox appears only if the instrument has a barcode reader or if a stacker is connected and has a barcode reader. For further details on the Barcode Scanner option refer to the Instructions for Use of the respective instrument manual.

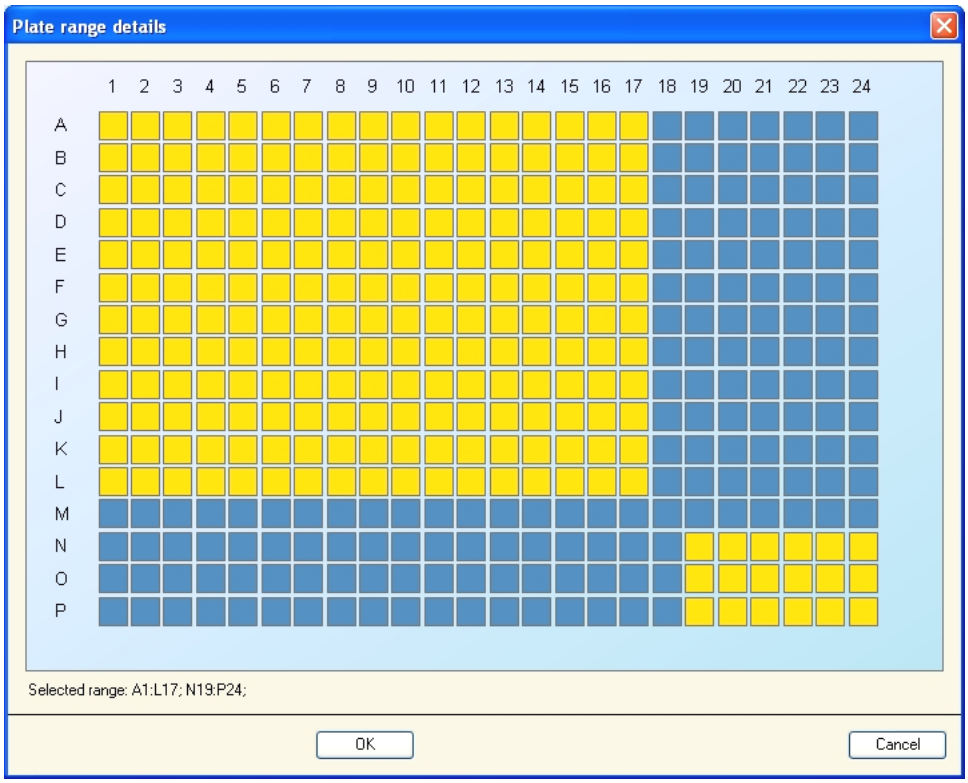
Part of Plate

The **Part of Plate** program element is displayed according to the selected plate format (number of wells). To measure individual wells, click the desired well or to measure a range of wells drag a frame around the desired range.



Independent Parts of Plate

Clicking on **Details…**, the plate preview can be zoomed and independent parts of the plate can be selected:



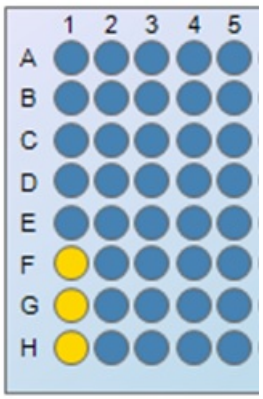
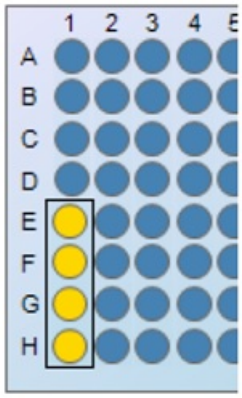
A second range of wells can be selected by pressing the **Control key** on the keyboard and dragging a frame over the wells to be selected.

Infinite M1000 and M1000PRO 4-Channel Absorbance Measurements

The 4-channel absorbance optics of the M1000 and M1000 PRO use all 4 channels whenever possible for the selected plate definition. The topmost well of a plate selection is always measured with the first measurement channel, the wells below are measured with the second, third and fourth channel, accordingly.

Well selections in the lower half of a plate, where one or more measurement channels move outside the well area of the plate, may not be used to avoid erroneous data due to the design of certain plates to avoid reflections. The well selection is therefore limited to patterns that do not encounter this problem.

Examples:

Not allowed plate range	Allowed plate range
	

Well

Use the **Well** program element to perform measurements well by well. Without this program element, all measurement steps are done plate-wise.

Cuvette

The **Cuvette** program element allows performing absorbance measurement in fixed wavelength and scan mode. This option is only available for the **Infinite M200** and **M200 PRO**.

2.2.2 Measurements

For detailed information on measurement methods, refer to the respective Instructions for Use of the instrument connected.

Absorbance

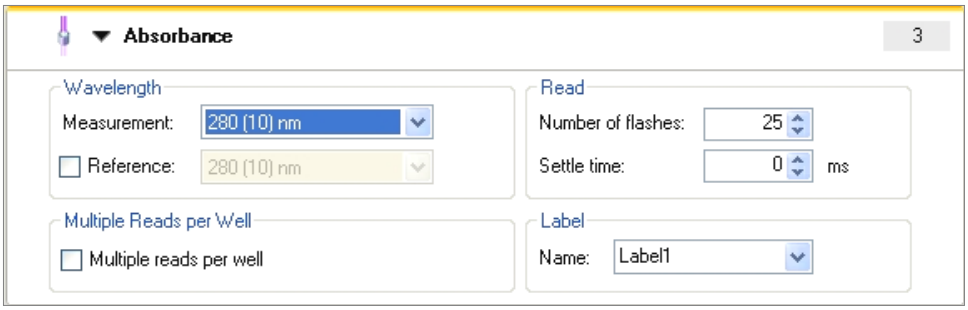
The **Absorbance** program element is used to perform absorbance measurements. Enter or select the respective parameters:

- Wavelength
- Reference
- Read/Flash
- Multiple Reads per Well
- Pathlength Correction (**Infinite 200** and **200PRO M** configurations only)
- Label

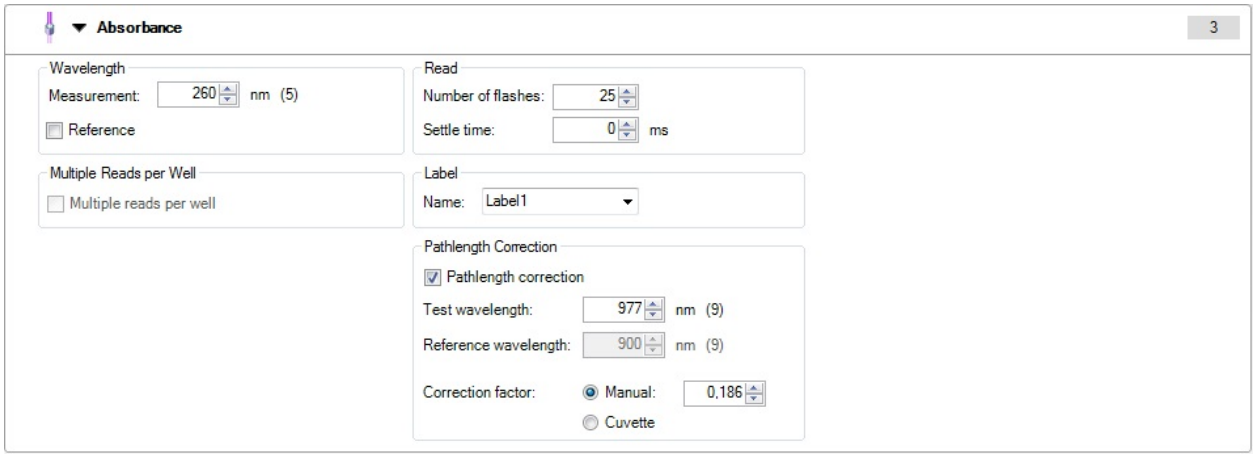
The **Reference** wavelength may be selected to correct for flash variations.

For filter instruments, two drop-down lists display the available measurement and reference filter wavelengths, according to the inserted absorbance filter slide. If the drop-down lists are empty, the absorbance filter either has not been inserted into the reader or has not been defined.

Example for **Infinite 200/200PRO F** Configurations



Example for **Infinite 200/200 PRO M** Configurations



Example for the **Infinite F50**

Pathlength Correction

The **Pathlength Correction,** for Infinite 200 readers with monochromator configuration, can be used to correct the measured absorbance values of samples in microplates to 1 cm pathlength, in order to compare the measurement results to those read with cuvettes or to perform quantitative analysis of samples based on their extinction coefficient.

According to the Lambert-Beer's law, the amount of absorbed light is proportional to the concentration of the sample and to the pathlength of the light passing the sample. Unlike a standard cuvette with a pathlength of 1 cm, the path of the light in a microplate is unknown and depends on the filling volume of wells. For aqueous solutions, the pathlength can be calculated from the absorbance values for water recorded in the near infrared wavelength range (900 nm to 1000 nm) by using a cuvette and the respective microplate.

Note
The absorption of water is temperature dependent. Please make sure that all measurements are performed at exactly the same temperature.

Note
Any light absorption of assay components between 900 and 1000 nm will interfere with pathlength correction.

Note
Please be aware that buffer (salt concentration), organic solvents, meniscus and plate characteristics can affect the pathlength correction measurement.

CAUTION
TURBID SAMPLES CAN LEAD TO A FALSE ESTIMATION OF PATHLENGTH DUE TO SCATTERING OF LIGHT. THE PATHLENGTH CORRECTION WITH CUVETTE WILL NOT COMPENSATE FOR THIS EFFECT.

If **Pathlength Correction** is selected, define the following parameters:

Test Wavelength	Define a wavelength within the available wavelength range to get the specific absorption of water in the aqueous sample. Recommended wavelengths: 977, 997, 998 and 1000 nm
Reference Wavelength	The background of water in the aqueous sample is measured at a **Reference wavelength** of 900 nm.
Correction factor	The **Correction factor** is defined as the absorbance value of water at **Test wavelength** corrected by the absorbance value of water at **Reference wavelength** for the pathlength of 1 cm. **Manual**: Define a manual value that has been previously calculated via water or sample buffer measurements at the **Test** and **Reference wavelengths** with a cuvette. **Cuvette**: If selected, the measurement with a cuvette filled with water or sample buffer in a cuvette port will be executed at defined **Test** and **Reference wavelengths** for the aqueous sample.

Note
Please make sure that the manual correction factor matches the selected Test and Reference wavelengths of your aqueous sample and was determined with the corresponding sample buffer.

The **Pathlength** calculation of the sample is performed as follows:

$Pathlength_{Sample} = (A_{TW} – A_{RW})/(A_{Water}) * 1\ cm$

A_{TW} = Absorption of aqueous sample at Test wavelength

A_{RW} = Absorption of aqueous sample at Reference wavelength

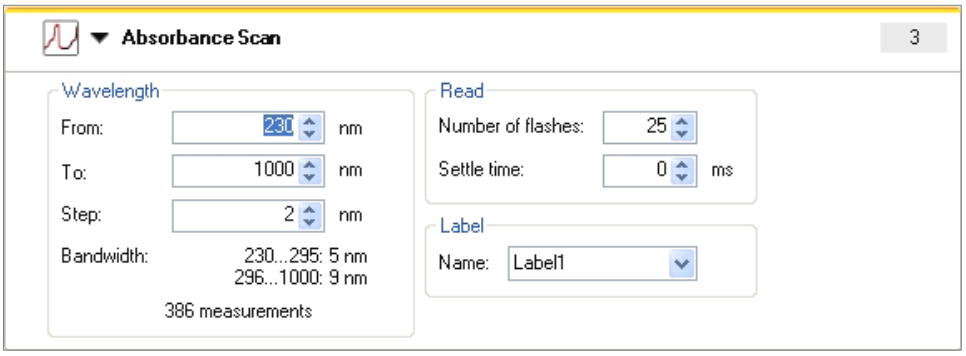
A_{Water}= A_{TW} minus A_{RW} of water in a 1 cm cuvette (= Correction factor)

The calculated pathlength is finally used to correct the absorbance of sample (A_{Sample}) at specific dye wavelength to 1 cm ($A_{SampleCorrected}$):

$A_{SampleCorrected} = A_{Sample}/Pathlength_{Sample}$

Absorbance Scan

The **Absorbance Scan** program element is available with the **Infinite M200**, **M200 PRO**, **M1000** and **M1000 PRO**.

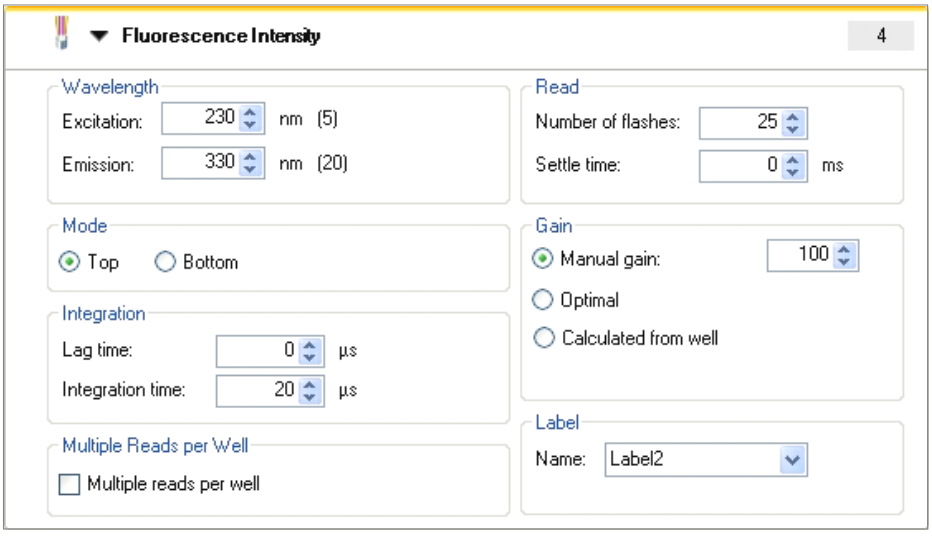


Enter or select the respective parameters:

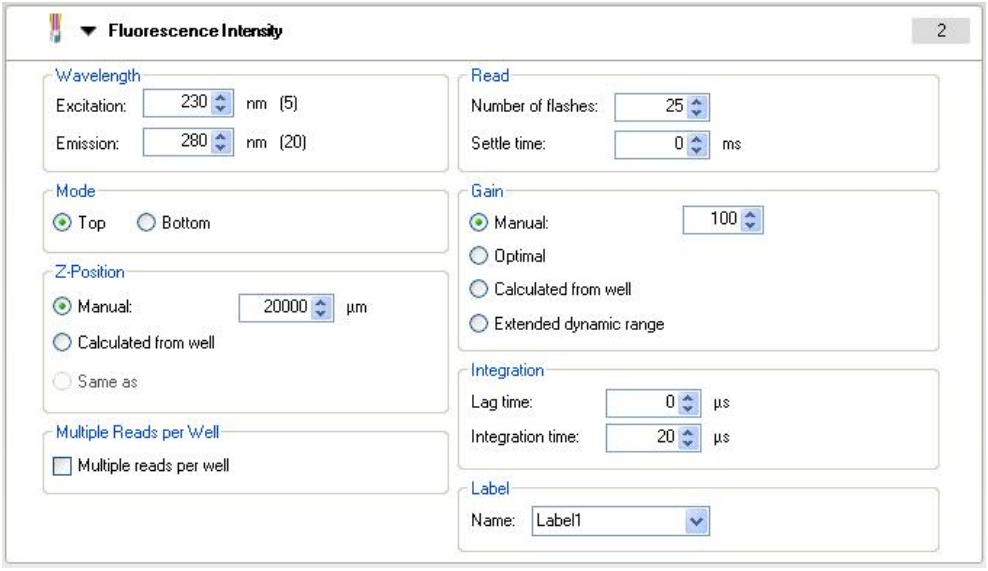
Wavelength	**From**: The lower wavelength limit **To**: The upper wavelength limit **Step**: Enter a valid value.
Read	**Number of flashes**: Indicates the number of flashes (select a number between 0 – 100). **Settle time**: The time between movement of the plate and starting of the read (selectable from 0 – 1000 ms).
Label	**Name**: Enter a label name.

Fluorescence Intensity

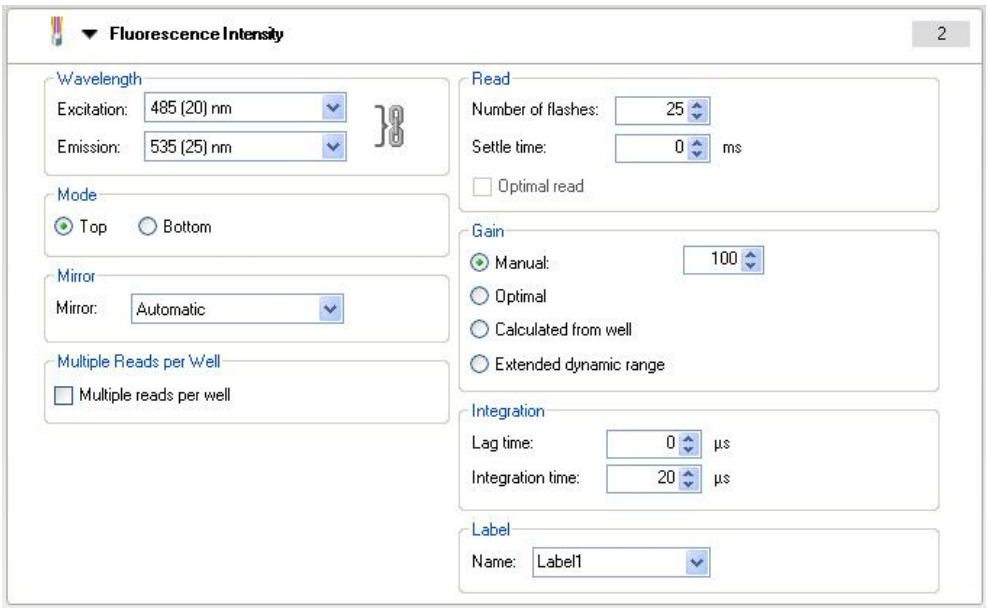
The **Fluorescence Intensity** program element contains fields for the selection of excitation and emission wavelength, top or bottom reading mode, integration and lag time, flash number and gain settings. A checkbox for multiple reads per well gives access to additional function.



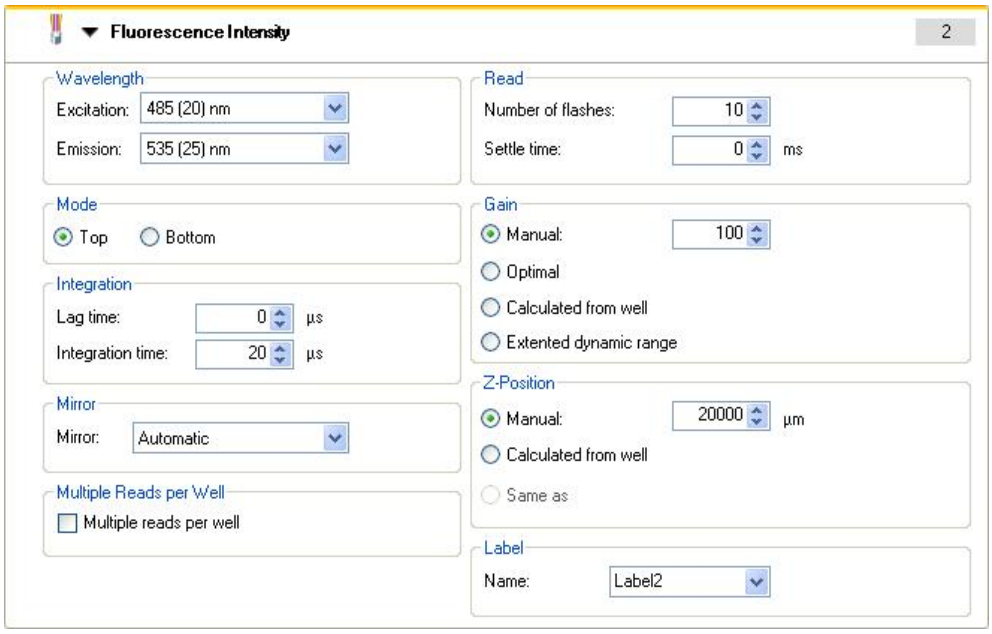
Example when connected to an **Infinite M200 PRO**:



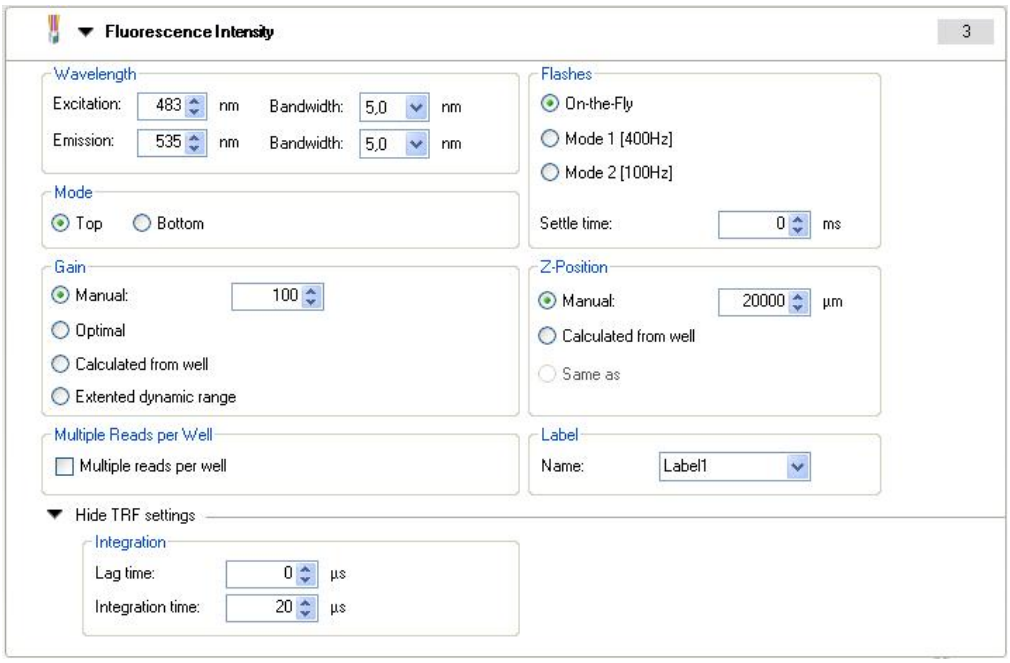
Example when connected to an **Infinite F200 PRO**:



When connected to an **Infinite F500**, this program element looks different: parameter fields for **Mirror** and **Z-Position** are added:



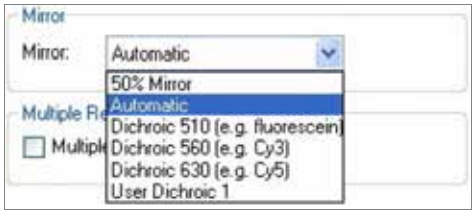
When connected to an **Infinite M1000** or **M1000 PRO**, this program element looks different: parameter fields for **Bandwidth, Show/Hide TRF settings** and additional flash modes are available.



The following are the **Fluorescence Intensity** parameters:

Wavelength	Specify an **Excitation** and an **Emission wavelength**. For filter instruments, two drop-down lists display the available measurement filter wavelengths. If the spin box of fixed values is empty, the excitation and emission filters have not been inserted into the reader or have not been defined. In the **Infinite M200, M200 PRO**, **M1000** and **M1000 PRO** both wavelengths can be entered as fixed values or selected by clicking the up or down buttons.

Bandwidth	For the **Infinite M1000** and **M1000 PRO** instruments, the bandwidth for **excitation** and **emission** can be selected.
Read	Specify a certain **Number of flashes** and, if required, **Settle time** before the next measurement. The number of flashes is selectable from 1 – 100 (up to 200 for **Infinite M1000** and **M1000 PRO**). **Settle time**: Enter a value to specify the time before the start of the measurement.
Flashes	When connected to an **Infinite M1000** or **M1000 PRO** instrument, select one of the following options and, optionally, enter a **Settle Time**: · On-the-fly · Mode 1 (400 Hz) · Mode 2 (100 Hz) For plates up to 384 wells: On-the-fly measurements are performed with one flash per well at 100 Hz. For 1536-well plates: On-the-fly measurements are performed with one flash per well at 400 Hz. In order to obtain a good measurement precision it is recommended to perform fluorescence measurements with the number of flashes that is set as a default value for the respective instrument. **Infinite M1000** and **M1000 PRO** allow switching between two flash frequencies for the Fluorescence Intensity and Fluorescence Intensity Scan mode: 100 or 400 Hz (corresponding to 100 or 400 flashes per second, respectively). The energy of one flash is app. 0.1 Joule for the 400 Hz mode and app. 0.4 Joule for the 100 Hz mode. For standard fluorescence measurements it is recommended to use the 400 Hz mode. For TRF (time resolved fluorescence) measurements the 100 Hz mode is recommended to improve the results.
Mode	Select **Top** or **Bottom**.
Label	Enter a label name.

Gain	The gain is an amplification factor for the photomultiplier tube (PMT) and may be set by selecting one of the following modes: **Manual gain**: user-defined gain value (valid range: 1-255) **Optimal gain**: calculated automatically by the instrument according to the highest signal within the selected well range in order to avoid OVER. Optimal gain determination is performed in a pre-measurement. It is recommended to use the optimal gain function for all applications that produce results with unknown RFU values. **Calculated from well**: determines the optimal gain for the selected well. The resulting gain value is applied to all other wells within the selected well range. **Extended dynamic range**: (available for all Infinite readers) The extended dynamic range option is an automatic gain function that serves to optimally adjust the gain setting for both very high and very low signals on a microplate within one single measurement. By selecting “extended dynamic range”, the measurement is done in two consecutive parts, one with a high and one with a low gain. The results of both measurements are automatically correlated and displayed within one single data set.
Hide/Show TRF settings: Integration/ Lag time	**Integration time:** duration of signal recording per well (valid range: 20-2000 µs). **Lag time**: time between flash and the start of signal integration. While lag time is an optional function, the integration time is a mandatory parameter determining the duration of signal recording. The default values for standard fluorescence intensity measurements are 0 µs lag time and 20 µs integration time. TRF measurements typically require a lag time according to the respective application.
Mirror	Mirror (available for **Infinite F200 PRO** and **F500**) The availability of mirrors depends on the selected plate format and on the types of dichroic mirrors that are installed. The mirror selection for **Infinite F500** looks as follows:  The mirror selection for **Infinite F200 PRO** is limited to the options 50% mirror, Automatic and Dichroic 510 (e.g. fluorescein). If an **Infinite F200 PRO** is configured with an AlphaScreen/AlphaLISA module, the Dichroic 510 mirror is replaced by a specific dichroic mirror, which can be used for AlphaScreen/AlphaLISA measurements only. In that case, only the **50% Mirror** is available in the mirror list. According to the selected filter wavelengths the appropriate mirror may be set by the instrument (selection “automatic”) or manually. Custom dichroic mirrors may be installed and defined by the user (**Infinite F500 only**). For further details on mirrors and mirror selection refer to the Instructions for Use of the **Infinite F200 PRO** and **F500** instrument, respectively.

Z-Position	**Z-Position** (available for **Infinite M200 PRO**, **F500**, **M1000** and **M1000 PRO**) The Z-position represents the height of the measurement head above the microplate. It can be determined as follows: **Manual** (default value: 20000 µm) **Calculated from well**: the instrument automatically calculates the optimal Z-position for one selected well and applies this value to all other wells within the selected well range. **Same as**: may be used for measurements with more than one measurement label. The Z-position is equal to that of the previous label. **Instrument / Z-position control**: may be used to determine the appropriate Z-position using a graphical scheme. The resulting value is applied to all further measurements until a different Z-position is entered. The Z-position control in the Infinite M200 PRO, Infinite M1000 and Infinite M1000 PRO also allows for defining two wells as Blank and Signal, respectively, and setting the Z-position to the value that gives the best Signal-to-Blank (S/B) ratio. For more detailed information on Z-positioning refer to the Instructions for Use of the **Infinite M200 PRO**, **F500**, **M1000** and **M1000 PRO** instrument, respectively.

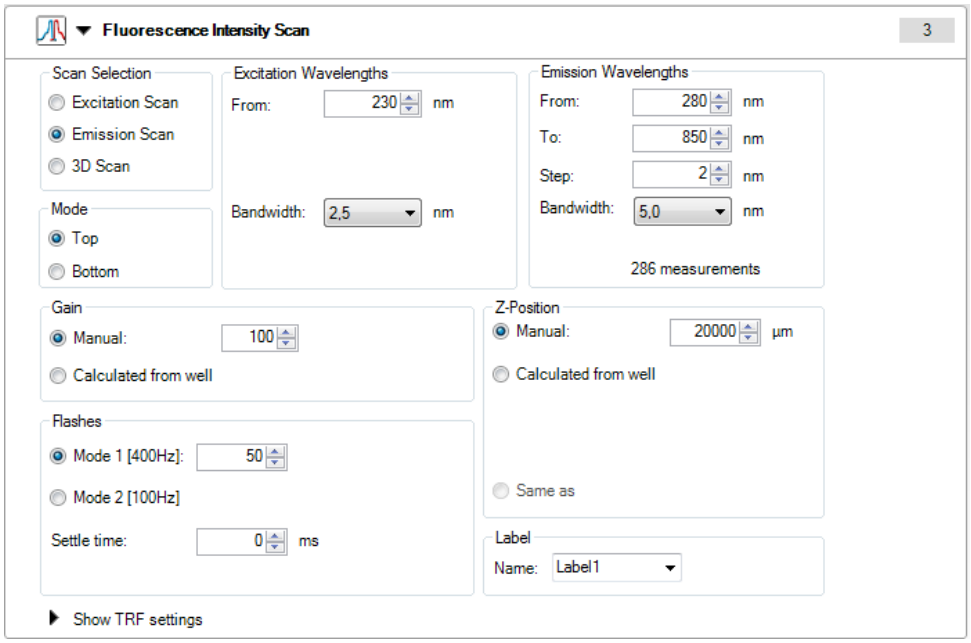
Fluorescence Intensity Scan

The **Fluorescence Intensity Scan** program element is available with the **Infinite M200, Infinite M200 PRO**, **Infinite M1000** and the **Infinite M1000 PRO**.

Example **Infinite M200**

Example **Infinite M200 PRO**

Example **Infinite M1000** and **Infinite M1000 PRO**



Enter or select the respective parameters:

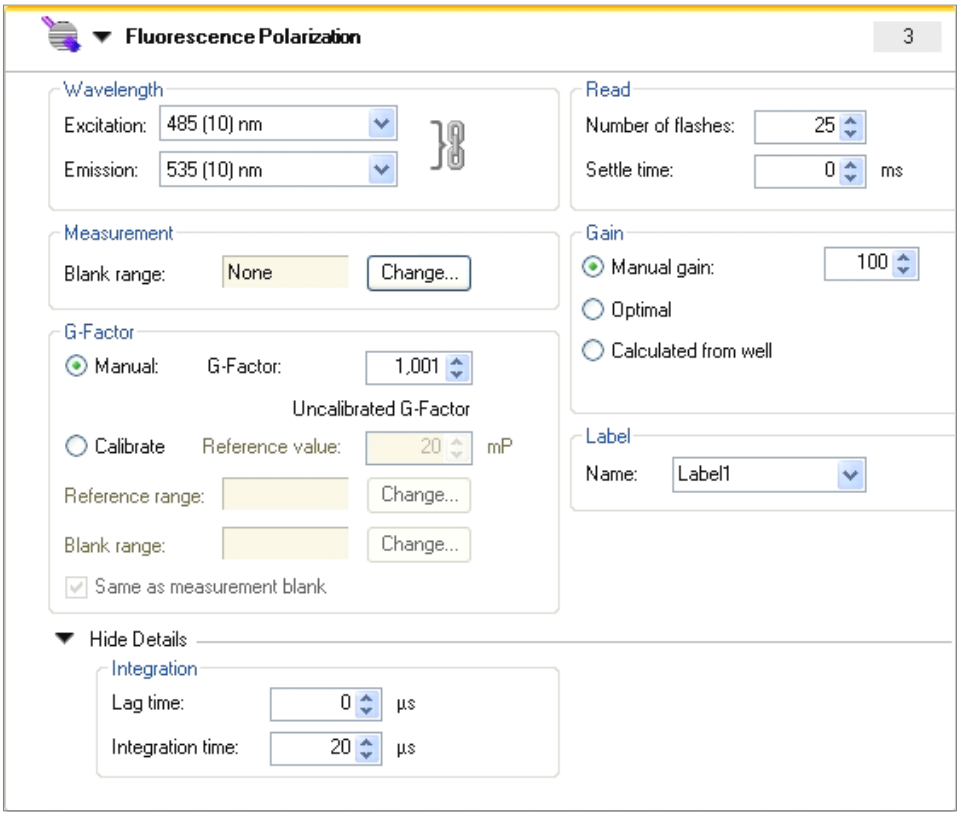
Scan Selection	Select either **Excitation Scan** or **Emission Scan**. With the **Infinite M1000** and **Infinite M1000 PRO** instruments, also the option **3D Scan** can be selected.
Excitation Wavelength	Values can only be entered if **Excitation Scan** is selected. **From**: Specify the range of the excitation by entering a value. **To**: Specify the range of the excitation by entering a value. **Step**: Enter a valid value.
Emission Wavelength	Values can only be entered if **Emission Scan** is selected. **From**: Specify the range of emission by entering a value. **To**: Specify the range of emission by entering a value. **Step**: Enter a valid value.

Bandwidth	For the **Infinite M1000** and **Infinite M1000 PRO** instruments, the bandwidth for excitation and emission can be selected. **Excitation bandwidth** in excitation scan/3D scan: The selection of the excitation bandwidth is limited to 5, 6, 7, 8, 9 or 10 nm (**Merged bandwidths)**, i.e. only the bandwidths that are valid within the whole excitation range of 230 – 850 nm.
Mode	Select **Top** or **Bottom**.
Hide/Show TRF Settings: Integration/Lag time	**Integration time**: duration of signal recording per well (valid range: 20-2000 µs). **Lag time**: time between flash and the start of signal integration. While lag time is an optional function, the integration time is a mandatory parameter determining the duration of signal recording. The default values for standard fluorescence intensity measurements are 0 µs lag time and 20 µs integration time. TRF measurements typically require a lag time according to the respective application.
Gain	The gain is an amplification factor for the photomultiplier tube (PMT) and may be set by selecting one of the following modes: **Manual gain**: user-defined gain value (valid range: 1-255) **Calculated from well**: determines the optimal gain for the selected well. The resulting gain value is applied to all other wells within the selected well range.
Read	Specify a certain **Number of flashes** and, if required, a **Settle time** before the measurement.
Flashes	When connected to an **Infinite M1000 or Infinite M1000 PRO** instrument, select one of the following options and, optionally, enter a **Settle Time**: Mode 1 (400 Hz) Mode 2 (100 Hz) In order to obtain a good measurement precision it is recommended to perform fluorescence measurements with the number of flashes that is set as a default value for the respective instrument. **Infinite M1000** and **Infinite M1000 PRO** allow switching between two flash frequencies for the Fluorescence Intensity and Fluorescence Intensity Scan mode: 100 or 400 Hz (corresponding to 100 or 400 flashes per second, respectively). The energy of one flash is app. 0.1 Joule for the 400 Hz mode and app. 0.4 Joule for the 100 Hz mode. For standard fluorescence measurements it is recommended to use the 400 Hz mode. For TRF measurements the 100 Hz mode is recommended to improve the results.
Label	Type in a label name.

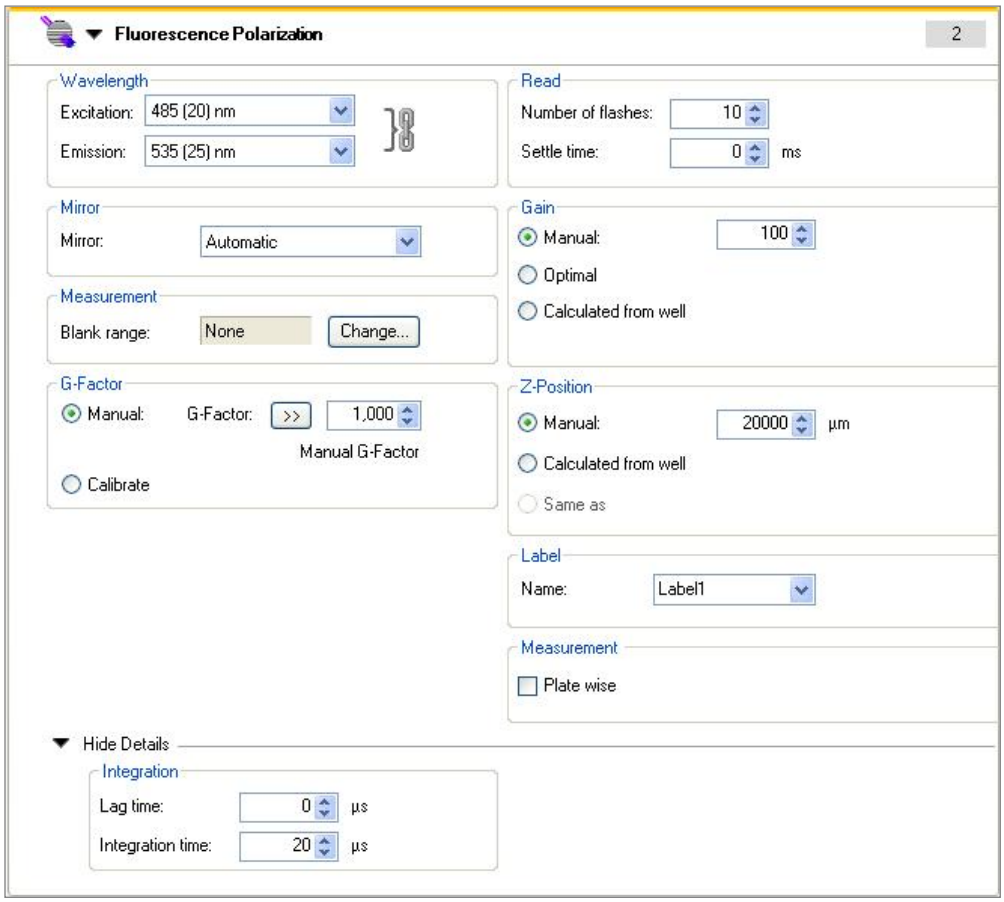
Fluorescence Polarization (available for F200, F200 PRO, F500, M1000 and M1000 PRO)

The **Fluorescence Polarization** (FP) program element is used to measure the rotational mobility of a fluorescent compound. Fluorescence polarization measurements are based on the detection of the extent of depolarization of fluorescence emission light after excitation of a fluorescent molecule by polarized light.

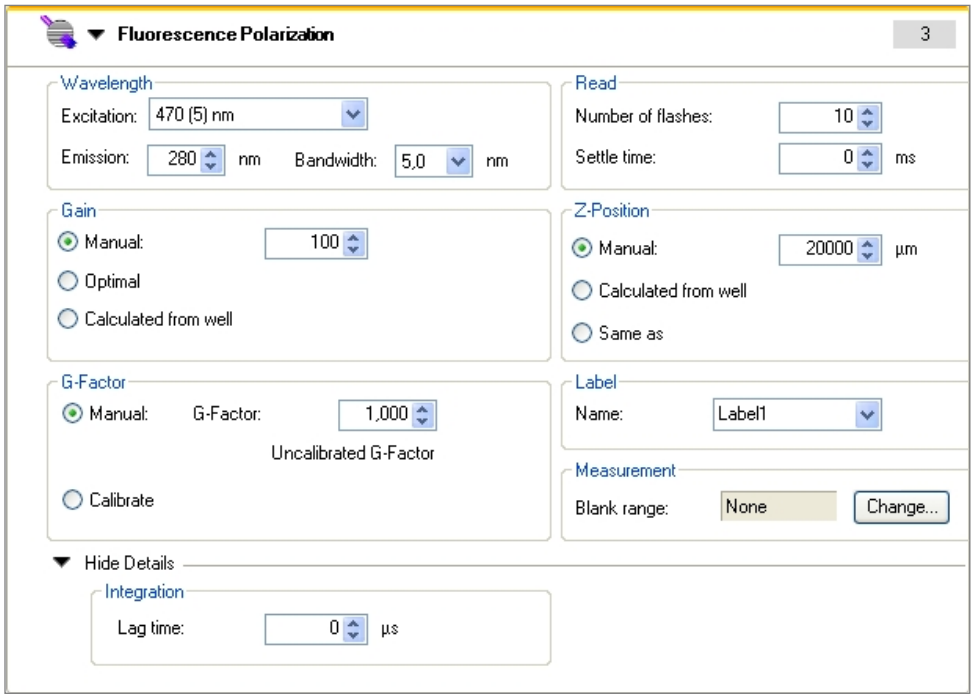
Example when connected to an **Infinite F200** instrument:



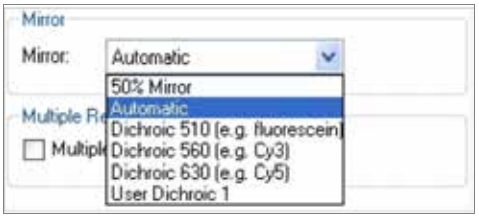
When connected to an **Infinite F500**, this program element looks different: parameter fields for **Mirror**, **Z-Position** and **Plate-wise** are added:



Example when connected to an **Infinite M1000** or **Infinite M1000 PRO** instrument:



Enter or select the respective parameters:

Wavelength	Filter instruments configured for Fluorescence Polarization (FP) measurements are delivered with a standard FP filter slide. The filter slide is equipped with filters and polarizers for excitation and emission, at **485** and **535** nm respectively, and can be applied, for example, for fluorescein-based FP applications.
Bandwidth	For the **Infinite M1000** and **Infinite M1000 PRO** instruments, the emission bandwidth can be entered.
Hide/Show Details: Integration	**Integration time**: duration of signal recording per well (valid range: 20-2000 µs). For Infinite M1000 and **Infinite M1000 PRO** instruments the integration time is defined by the number of flashes. 1 to 1000 flashes can be selected (1 flash is 10 ms integration time). **Lag time**: time between flash and the start of signal integration. While lag time is an optional function, the integration time is a mandatory parameter determining the duration of signal recording. The default values for standard fluorescence intensity measurements are 0 µs lag time and 20 µs integration time. TRF measurements typically require a lag time according to the respective application.
Mirror	**Mirror** (available for **Infinite F500**) The availability of mirrors depends on the selected plate format and on the types of dichroic mirrors that are installed.  According to the selected filter wavelengths the appropriate mirror may be set by the instrument (selection “automatic”) or manually. Custom dichroic mirrors may be installed and defined by the user. For further details on mirrors and mirror selection refer to the Instructions for Use of the **Infinite F500** instrument.
Z-Position	**Z-position** (available for **Infinite F500**, **M1000** and **M1000 PRO**) The Z-position represents the height of the measurement head above the microplate. It can be determined as follows: **Manual** (default value: 20000 µm) **Calculated from well**: the instrument automatically calculates the optimal Z-position for to one selected well and applies this value to all other wells within the selected well range. **Same as**: may be used for measurements with more than one measurement label. The Z-position is equal to that of the previous label. **Instrument / Z-position control**: may be used to determine the appropriate Z-position from a graphical scheme. The resulting value is applied to all further measurements until a different Z-position is entered. For more detailed information on Z-positioning refer to the Instructions for Use of the Infinite F500, M1000 or M1000 PRO instrument.

Measurement	If the Measurement Blank range should be defined, click **Change**.
G-Factor	The G-Factor compensates for differences in optical components between the parallel and perpendicular measurement. The G-Factor is the correction factor that can be determined for the wavelengths of the fluorophore by measuring a sample with a known polarization value. **Uncalibrated G-Factor**: If no calibrated G-factor is available, the default value of 1 will be displayed and marked as **Uncalibrated G-Factor.** In order to enable the measurement, confirm this value or select a new one by either clicking the up and down arrows or by entering a value manually. **Calibrate**: When selecting Calibrate, the G-factor is determined for the current measurement parameters and used for the following FP measurement. In order to perform the G-Factor calibration, please define: **Reference value**: Select a polarization value that shall be used for reference e.g. 20 mP. **Reference range**: Click on **Change** and select the wells filled with the reference fluid, e.g. 1 nM fluorescein. **Blank range**: Click on **Change** and select the wells filled with the reference blank. Select **Same as measurement blank** if the reference blank is the same as the measurement blank. For further details see the respective Instructions for Use of the instrument connected.
Read	Specify a certain **Number of flashes** and, if required a **Settle time** before the measurement.
Gain	The gain is an amplification factor for the photomultiplier tube (PMT) and may be set by selecting one of the following modes: **Manual gain**: user-defined gain value (valid range: 1-255) **Optimal gain**: calculated automatically by the instrument according to the highest signal within the selected well range in order to avoid OVER. Optimal gain determination is performed in a pre-measurement. It is recommended to use the optimal gain function for all applications that produce results with unknown RFU values. **Calculated from well**: determines the optimal gain for the selected well. The resulting gain value is applied to all other wells within the selected well range. **Extended dynamic range**: (available for all Infinite readers) The extended dynamic range option is an automatic gain function that serves to optimally adjust the gain setting for both very high and very low signals on a microplate within one single measurement. By selecting “extended dynamic range”, the measurement is done in two consecutive parts, one with a high and one with a low gain. The results of both measurements are automatically correlated and displayed within one single data set.
Label	Enter a label name.
Plate-wise	If **Plate-wise** is selected, all selected wells will be measured with the parallel emission filter and subsequently with the perpendicular filter. In contrast, of plate-wise is not selected, each well will be measured with the parallel and perpendicular filter before continuing to the next well.

Multiple Reads per Well

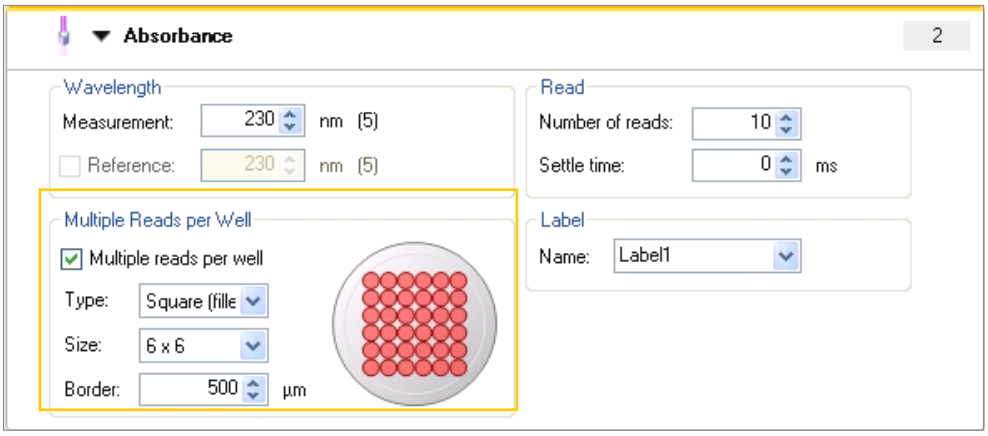
The i-control software allows the user to define multiple reads per well (MRW) in **Absorbance**, **Fluorescence top** and **Fluorescence bottom mode**.

The MRW feature is not available for well wise measurements.

The **Reference wavelength** on the absorbance program element is not selectable in combination with multiple reads per well.

For Infinite M1000 and Infinite M1000 PRO instruments there is a minimum **Settle time** of 10 msec required as soon as **Multiple reads per well** is selected.

The multiple reads per well function can be activated on an absorbance or fluorescence intensity program element by selecting the **Multiple reads per well** check box:



Note
The function Multiple reads per well is only available for the fixed wavelength reading modes Absorbance, Fluorescence intensity top and Fluorescence intensity bottom. The function is not available for scan measurements.

More details on defining parameters for multiple reads per well, are available in the respective Instructions for Use of the instrument connected.

The multiple reads per well function is available for plate formats with up to 384 wells. 1536 well plates are not supported.

Optimal Read

Similar to the MRW read mode, the "Optimal Read" function is a measurement on multiple, spatially separated spots inside the well. The spots are arrayed to cover the whole well area in order to achieve maximal well illumination. The total number of individual measurement spots per well is reflected by the size of the Fluorescence Intensity Bottom fiber and is optimized for plate formats from 6 to 96 wells (see Table: “Optimal Read” spot patterns in different plate formats). 384-well plates are optimally illuminated by a single-spot read.

Example for Infinite 200 PRO, Infinite F500:

Plate	Pattern	Size	Spots
384-well	**Optimal Read** function not available		
96-well	Circle	2x2	4
48-well	Circle (filled)	4x4	12
24-well	Circle (filled)	5x5	21
12-well	Circle (filled)	7x7	37
6-well	Circle (filled)	10x10	76

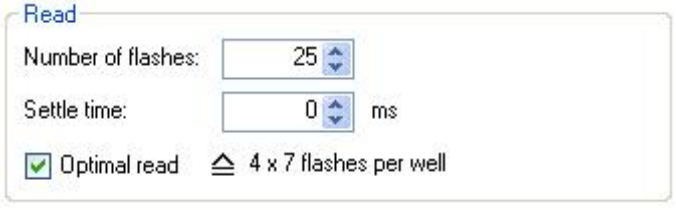
Table: **Optimal Read** spot patterns in different plate formats

Example for Infinite M1000, Infinite M1000 PRO:

Plate	Pattern	Size	Spots
384-well	**Optimal Read** function not available		
96 wells	Circle (filled)	3x3	5
48 wells	Circle (filled)	5x5	21
24 wells	Circle (filled)	7x7	37
12 wells	Circle (filled)	9x9	61
6-well	**Optimal Read** function not available		

Changing the total number of flashes per well (1-100) will result in the automatic adjustment of the number of flashes per spot, giving the user the possibility to obtain representative results in each well.

The total number of flashes is automatically distributed over all measured spots. A minor imprecision occurs if an entered flash number is not divisible without a remainder by the default number of spots for the used plate format. In this case the next possible flash distribution that is integrally divisible by the number of spots per well is calculated, e.g. a measurement with a total of 25-28 flashes in a 96-well plate is performed with 7 flashes per spot, whereas a total flash number of 29 results in 8 flashes per spot.

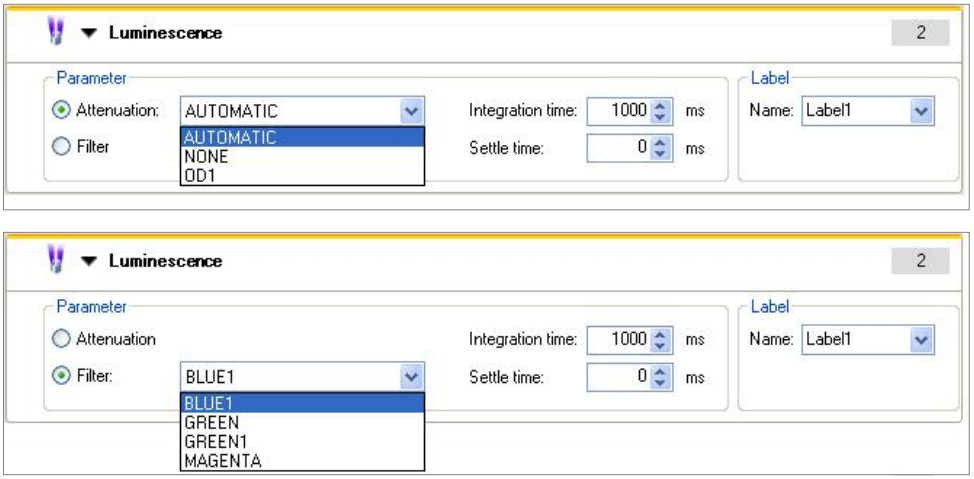


The standard MRW function for Fluorescence Intensity Bottom reads is disabled when “Optimal Read” is activated and vice versa.

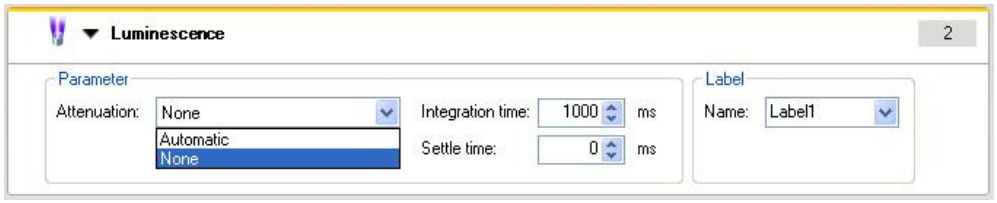
Luminescence

The **Luminescence** program element is used to determine the activity of a luminescent compound.

Example for the **Infinite 200**



Example for the **Infinite 200 PRO**



Enter or select the respective parameters:

Attenuation

For strongly luminescent samples it may be necessary to apply neutral density filters to reduce the luminescent signal.

Select the desired attenuation option. The options depend on the instrument connected:

	F/M200	F/M200 PRO	F500	M1000	M1000 FW 2.0 and higher (Ref 30061442) and M1000 PRO
None	ü	ü	ü	ü	ü
OD1	ü		ü	ü	
Automatic (OD1)			ü	ü	
Automatic (OD2)		ü			ü

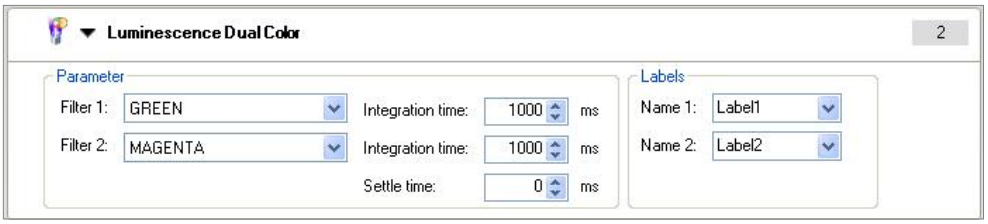
By selecting **Automatic**, only those wells that require attenuation are attenuated by a factor of 10 using an OD1 filter or by a factor of 100 using an OD2 filter depending on the instrument connected.

Filters	**Use of Color Filters for Single Luminescence:** (available for Infinite F500, M1000 and M1000 PRO) All filters that are available for dual color luminescence may be used in single luminescence measurements as well. Besides the attenuation functions an additional dropdown list in the attenuation field displays the filters for GREEN, GREEN1, BLUE1 and MAGENTA to be selected individually for single luminescence applications. Infinite M1000 PRO instruments additionally offer filters for BLUE and ORANGE.
Integration time	Enter a value to specify the duration of integration. All wells will be measured with this fixed user-defined integration time.
Settle time	Enter a value to specify the time delay between a plate transport movement and the start of the measurement.

Luminescence Dual Color

The **Luminescence Dual Color** program element is used to discriminate different wavelengths within the luminescence signal (for assays that are based on 2 distinct signals).

This dual filter system permits independent measurement by detecting two different wavelengths within one well. The available filters depend on the connected reader.

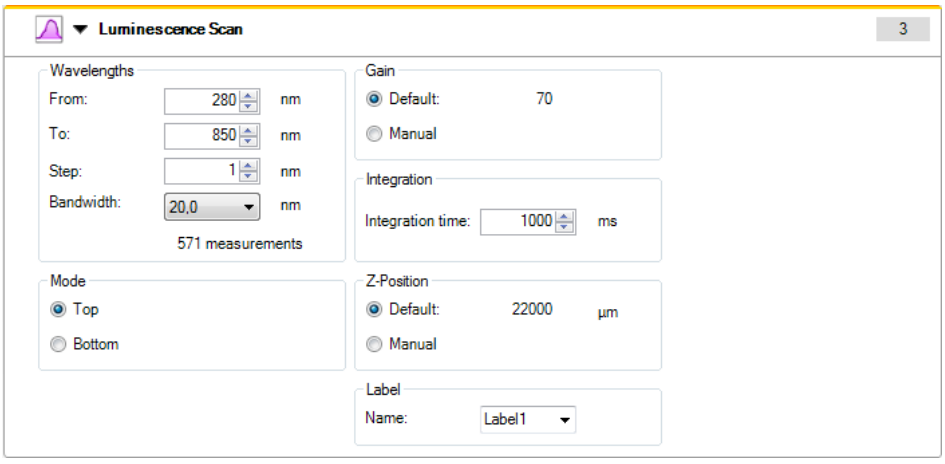


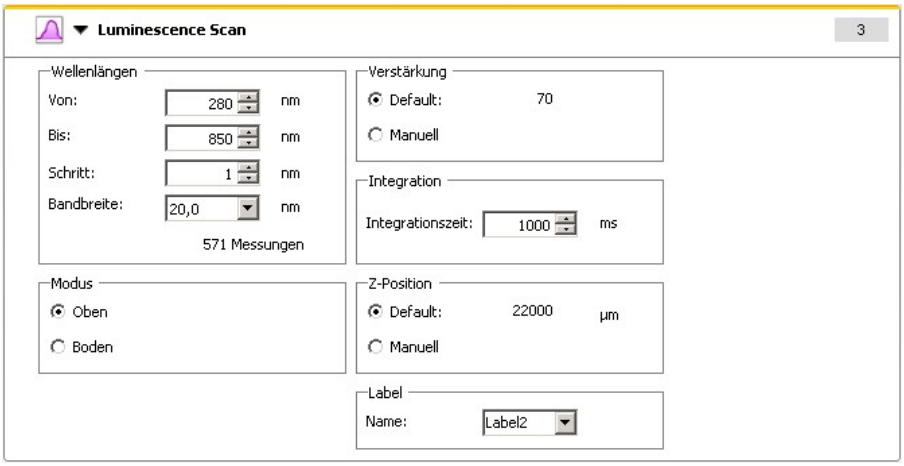
The following are the **Luminescence Dual Color** parameters:

Parameter	Select the appropriate color filters and define an **Integration time** for each label. If required, enter a **Settle time** before the measurement.
Label	Enter different **Label Names**.

Luminescence Scan

The **Luminescence Scan** function is available with the **Infinite M1000** with main firmware V 2.0 or higher (Ref 30061442) and with **Infinite M1000 PRO**.





Wavelengths	**From:** Select the starting wavelength for the scan. **To:** Select the endpoint wavelength for the scan. **Step:** enter a valid value **Bandwidth:** Select a value from the drop down list.
Mode	Select **Top** or **Bottom.**
Gain	**Default:** this value is instrument specific (see also 30036266_IFU_InfiniteM1000) **Manual:** User-defined gain value (valid range 1-255)
Integration	**Integration time:** enter a value to specify the duration of integration.
Z-Position	**Default:** 22000 µm **Manual:** The Z-position represents the height of the measurement head above the microplate.
Label	Type in a label name.

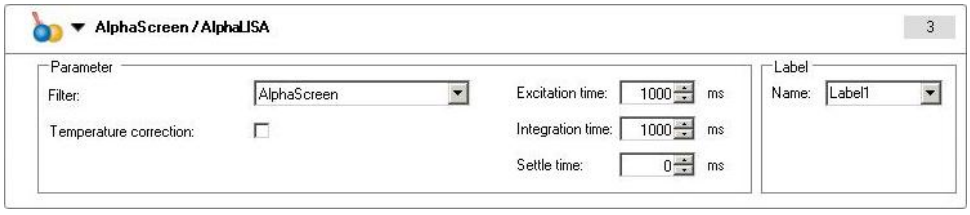
AlphaScreen / AlphaLISA

The **AlphaScreen / AlphaLISA** function is available with the Infinite M1000 PRO and Infinite F200 PRO. It is a measurement designed specifically for AlphaScreen / AlphaLISA assays.

In Infinite M1000 PRO, AlphaScreen / AlphaLISA is based on a luminescence measurement.

In Infinite F200 PRO, AlphaScreen / AlphaLISA is based on a fluorescence intensity measurement.

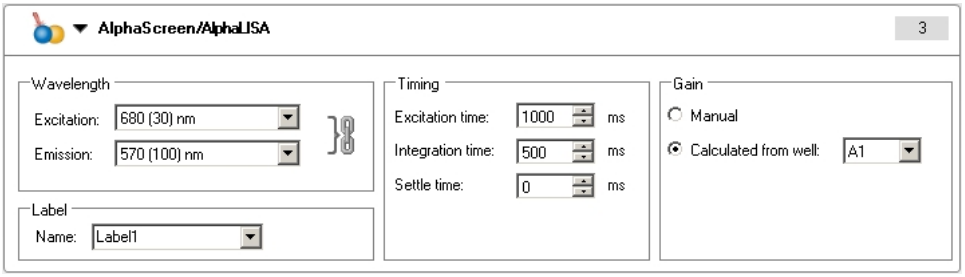
AlphaScreen / AlphaLISA in Infinite M1000 PRO:



Enter or select the respective parameters:

Filter	Select an emission filter for **AlphaScreen** or **AlphaLISA.**
Temperature correction	Check this box to activate the temperature correction function. The temperature correction function is recommended for samples in a temperature range of 20-25°C with a heterogeneous temperature distribution across the plate.
Excitation time	Enter a value to specify the duration of excitation. All wells will be measured with this user-defined excitation time.
Integration time	Enter a value to specify the duration of signal integration. All wells will be measured with this user-defined integration time.
Settle time	Enter a value to specify the time delay between a plate transport movement and the start of excitation.
Label	Type in a label name.

AlphaScreen / AlphaLISA in Infinite F200 PRO:



Enter or select the respective parameters:

Wavelength	Specify an **Excitation** and an **Emission wavelength**. For filter instruments, two drop-down lists display the available measurement filter wavelengths. If the spin box of fixed values is empty, the excitation and emission filters have not been inserted into the reader or have not been defined.
Excitation time	Enter a value to specify the duration of excitation. All wells will be measured with this user-defined excitation time.
Integration time	Enter a value to specify the duration of signal integration. All wells will be measured with this user-defined integration time.
Settle time	Enter a value to specify the time delay between a plate transport movement and the start of excitation.

Gain	The gain is an amplification factor for the photomultiplier tube (PMT) and may be set by selecting one of the following modes: **Manual**: user-defined gain value (valid range: 1-255) **Calculated from well**: determines the optimal gain for the selected well. The resulting gain value is applied to all other wells within the selected well range. The gain calculation for AlphaScreen/AlphaLISA measurements includes a waiting time in between each optimization run to avoid erroneously increased signals due to the previous excitation. This will result in a slightly increased measurement duration. If the fastest-possible reading time is desired, the gain may be optimized in a pre-measurement of a well containing the highest signals/concentrations. The pre-determined gain value can then be set manually for the measurement of the whole plate.
Label	Type in a label name.

Note
AlphaScreen/AlphaLISA measurements are only possible as endpoint measurements and cannot be performed in combination with the injector system and the heating system.

2.2.3 Actions

Temperature

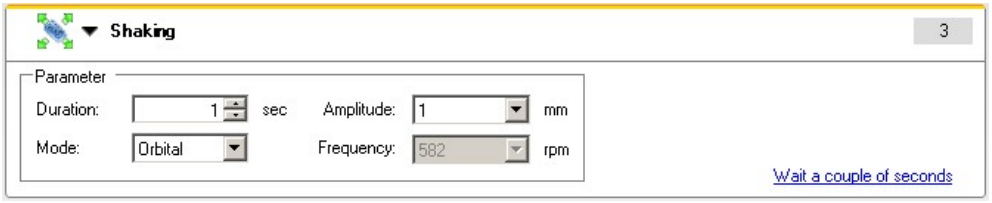
Select the **Temperature** program element to enter a certain target temperature.

Select **On** to enter a target temperature value. Click on the link Wait until temperature is reached to define the **Minimum** and/or **Maximum** temperature values. The heating of the instrument starts when clicking the **Start** button. For pre-heating the instrument, select **Heating…** in the **Instrument** menu and click the **On** button.

The measurement only starts if the current instrument temperature is within the specified range. See 2.2.5 Miscellaneous, Wait for Temperature .

Shaking

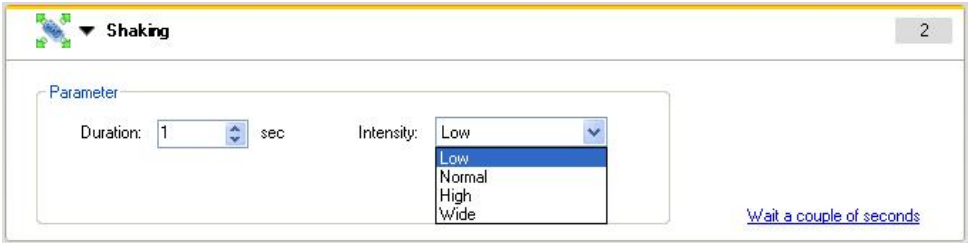
Select the **Shaking** program element if the plate is to be shaken, either before the measurement or between kinetic cycles.



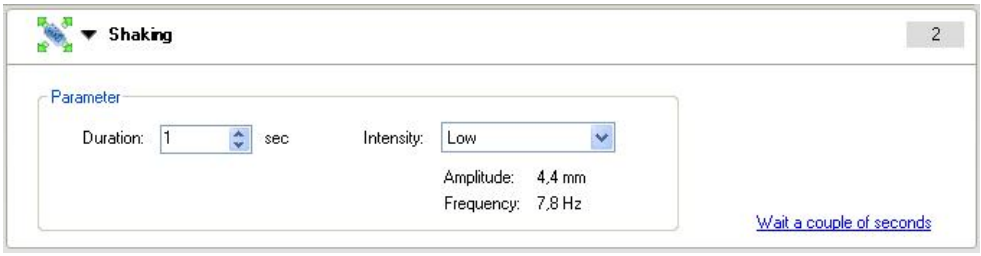
Enter the respective parameters:

Duration	Enter the duration of the shaking process.
Mode	Select between the options **Linear, Orbital** and **Double Orbital** from the drop-down list. The Mode **Double Orbital** is available for **Infinite M1000** for FW 2.0 and higher (Ref 30061442) and **Infinite M1000 PRO**.
Amplitude	Enter the required Amplitude value from the drop-down list.
Intensity	The **Infinite F50** offers the possibility to use pre-defined shaking modes by selecting a shaking **Intensity** from the drop-down list. The corresponding shaking frequency and amplitude are displayed automatically with the selected Intensity mode.

Shaking Modes; Example for the **Infinite F50**



Shaking Modes; Example for the **Infinite F50**

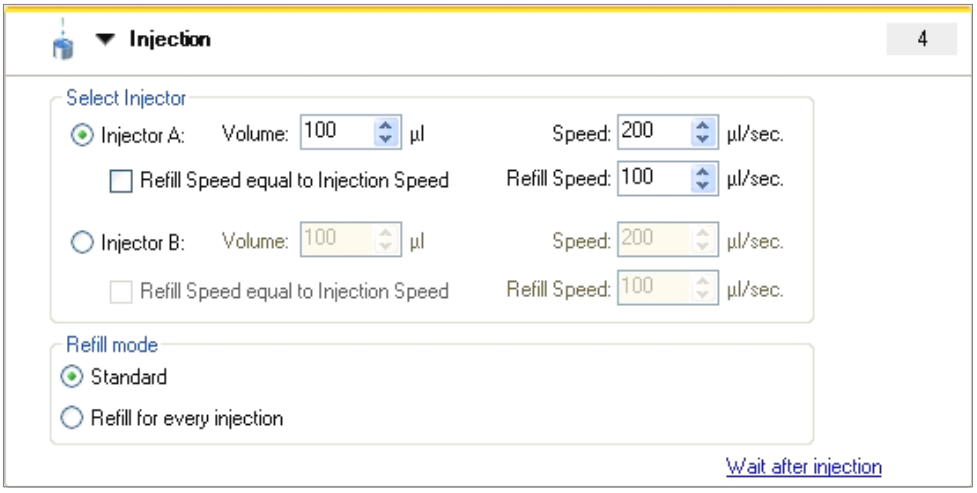


Clicking the link Wait a couple of seconds inserts a new program element. See 2.2.5 Miscellaneous, Wait (Timer).

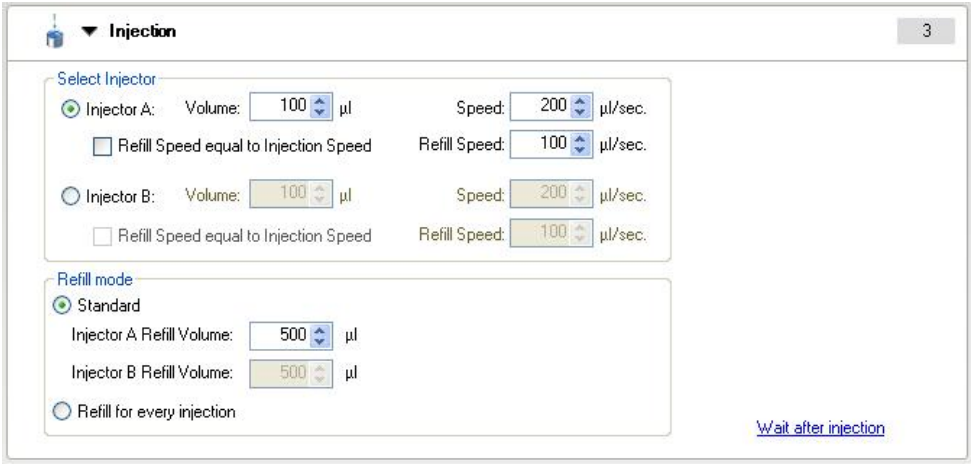
Injection

The **Injection** program element is dependent on a precedent well strip to inject liquid into one well after the other.

See also 3.3.2 The Difference between Inject and Dispense.



Example for the **Infinite M200 PRO**



The following are the **Injection** parameters:

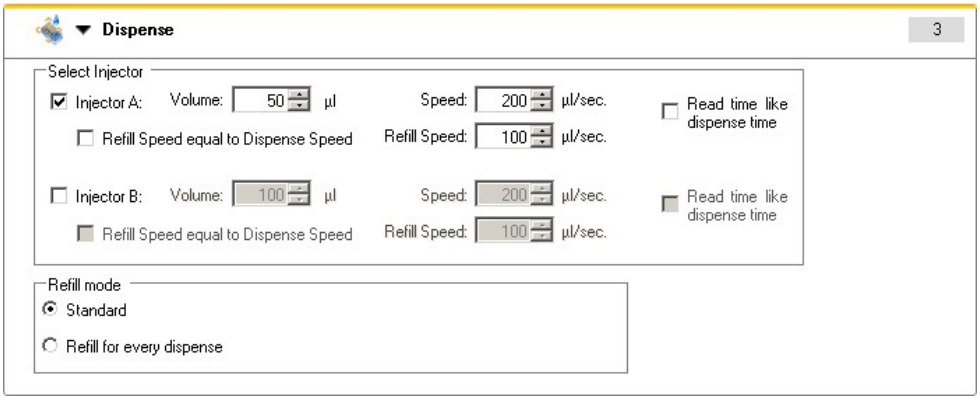
Select Injector	Select either **Injector A** or **B** if the instrument is equipped with two injectors.
	Volume: Specifies the volume to inject into a single well.
	Speed: Specifies the speed of liquid flow during injection.

	Refill Speed equal to Injection Speed: Clear the check box to enter the refill speed which may be different than the injection speed. The syringe can be filled faster, even if the injection speed is low.
Refill Mode	Select either **Standard** or **Refill for every injection**.
	Standard: Injection occurs as long as the syringe contains enough liquid. As soon as the liquid in the syringe is used up, the syringe is refilled with the entered refill volume (200 PRO, M1000 – for FW 2.0 and higher - Ref 30061442, M1000 PRO).
	Refill for every injection: Refilling of the syringe occurs for each injection step.

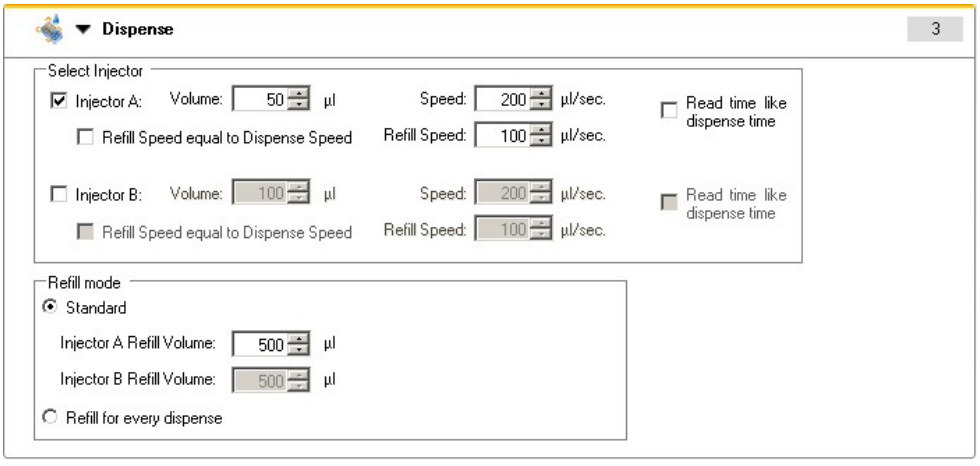
Click the link Wait after injection to define the time for starting the next workflow.
See 2.2.5 Miscellaneous - Wait (Timer).

Dispense

The **Dispense** program element is always used plate-wise to fill the plate (or part of plate) with liquid.
See 3.3.2 The Difference between Inject and Dispense.



Example for the **Infinite 200 PRO**



The following are the **Dispense** parameters:

Select Injector	Select either **Injector A** or **B** if the instrument is equipped with two injectors.
	Volume: Specifies the volume to inject into a single well. **Speed:** Specifies the speed of liquid flow while dispensing. **Refill Speed equal to Dispense Speed:** Clear the check box to enter the refill speed which may be different than the injection speed. The syringe can be filled faster, even if the dispensing speed is low. **Read time like dispense time:** By selecting this check box, the dispense function and the timing of the measurement is linked. Usually, the measurement is performed much faster than dispensing a reagent. Therefore, the time interval differs considerably between dispensing and measuring from the first to the last wells. The overall dispense time is divided by the number of wells to be processed to calculate the measurement delay for every well. However, there is no delay in dispense if the dispense time is shorter than the measurement time.
Refill Mode	Select either **Standard** or **Refill for every injection**. **Standard**: Dispensing occurs as long as the syringe contains enough liquid. As soon as the liquid in the syringe is used up, the syringe is refilled with the entered refill volume (200 PRO and M1000 – for FW 2.0 and higher - Ref 30061442, M1000 PRO). **Refill for every dispense**: Refilling of the syringe occurs for each dispense step.

Move Plate/Cuvette

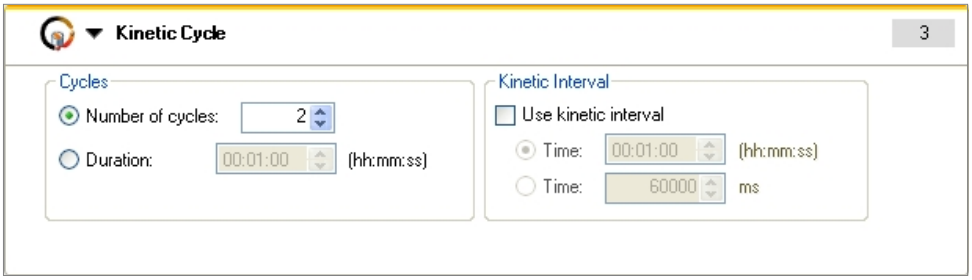
Select the program element **Move Plate/Cuvette** to move the plate/cuvette out of or into the instrument at a certain moment during the workflow.

If the plate/cuvette is moved out of the reader during a workflow (e.g. to pipet some liquid into the wells of the microplate), it must be followed by a subsequent **Move in** step, so that the measurement can be finished.

2.2.4 Kinetic

Kinetic Cycle

Use the program element **Kinetic Cycle** to perform several consecutive measurements, which may be executed in certain intervals.



Enter the respective parameters:

Cycles	**Number of cycles:** Enter a number or click the up or down arrows for the number of actual measurement steps (2 – 1000 cycles) **Duration:** Enter the duration, format hh:mm:ss.
Kinetic Interval	**Use kinetic interval:** Enter the time interval (hh:mm:ss or ms).

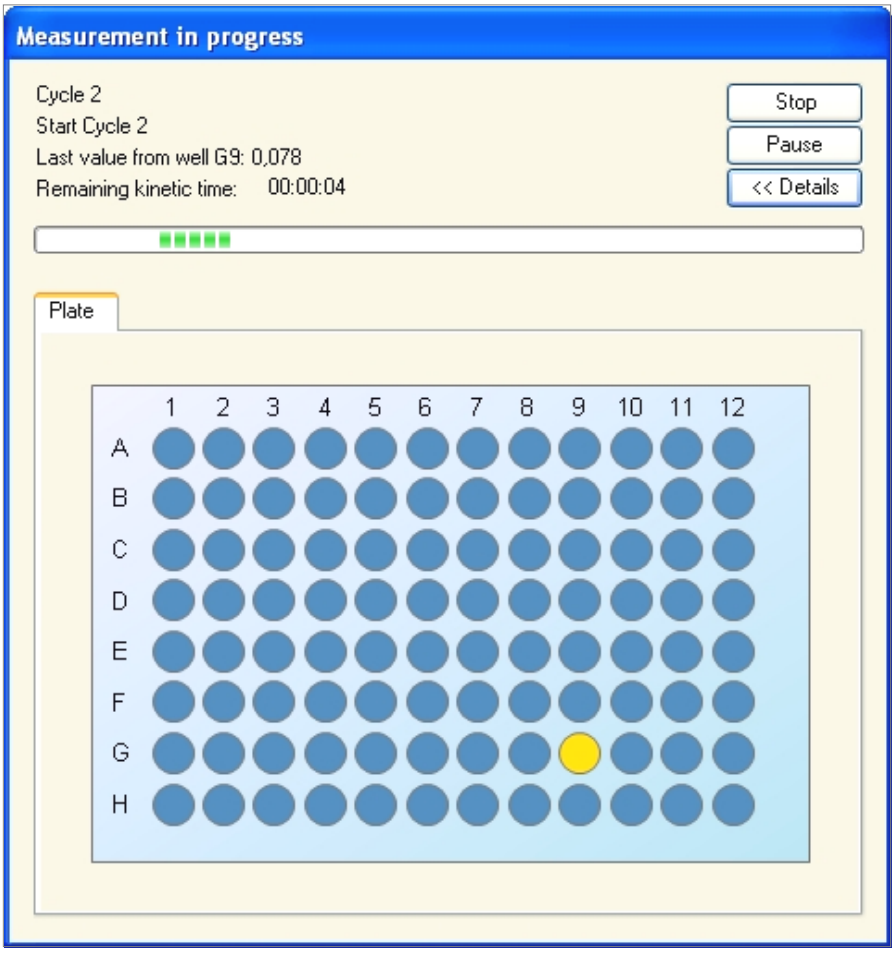
Plate-wise kinetic measurements

Each cycle of the kinetic measurement is performed on all selected wells. Plate-wise kinetic measurements may contain a maximum of ten independent measurement stripes that do not need to be of the same measurement type.

Well-wise kinetic measurements

All cycles of the kinetic measurement are first performed in one well before continuing to the next well. Well-wise kinetic measurements may be composed of a maximum of four measurement stripes of the same type, e.g., four absorbance stripes. The Infinite M1000 and Infinite M1000 PRO allow five measurement stripes of the same type within well-wise kinetic measurements.

After having started the measurement, it is possible to interrupt a plate-wise kinetic measurement clicking the **Pause** button and to continue:



Kinetic Condition

Use the **Kinetic Condition** program element to define which actions should be executed at a certain cycle.

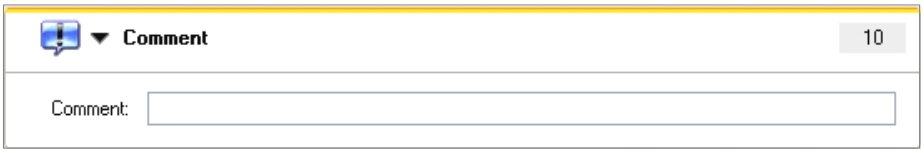
If **3** is entered for **Execute command at cycle** within a kinetic measurement containing, e.g. a **Shake** step, shaking is performed only at cycle 3.

Note
Kinetic conditions such as Shake, Inject and Dispense should be inserted right after a Kinetic Cycle program element in order to ensure optimal result reproducibility.
Users are advised to set up suitable scripts prior to the measurements and to use the same script for all similar kinetic measurements in order to obtain comparable results.

2.2.5 Miscellaneous

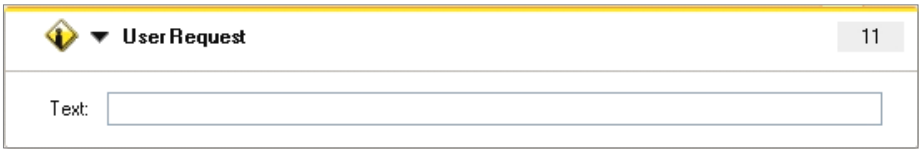
Comment

Use the program element **Comment** to enter a remark or statement for the current measurement in the text field. This text is shown together with the measurement in the Excel output sheet.



User Request

The **User Request** program element informs the operator of the instrument to execute a definite action during the workflow at a certain time.

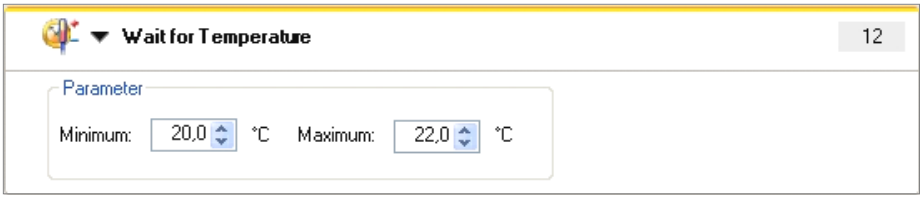


If for example the **Move Plate** program element is used to move the plate out to perform a certain action, then the entered text should inform the operator to perform these actions. A dialog box shows the message and the measurement process stops until **OK** is clicked.

If the plate should be moved in after pipetting for example, then the text **Move Plate In** informs the operator to move the plate in after pipetting to continue the workflow.

Wait for Temperature

Use the program element **Wait for Temperature** to define a valid temperature range for the assay.

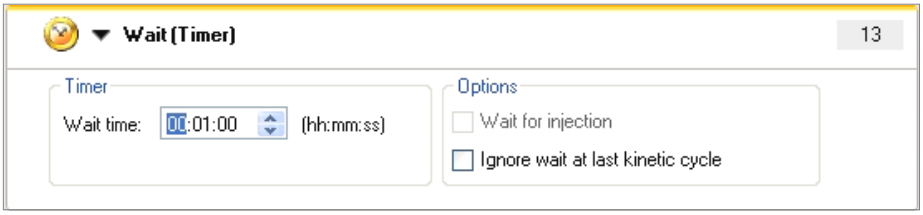


This is typically used after a **Temperature** program element.

Wait (Timer)

Use the **Wait (Timer)** program element to define a certain waiting period before the next step within a workflow is executed.

In the **Wait time** field enter the required time.



Enter the respective parameters:

Timer	Enter the **Wait time** (hh:mm:ss)
Options	**Wait for injection:** The time for injection is included in the wait time. **Ignore wait at last kinetic cycle:** When the program step **Wait (Timer)** is the last action within a kinetic run, the wait time will be ignored in the last cycle.

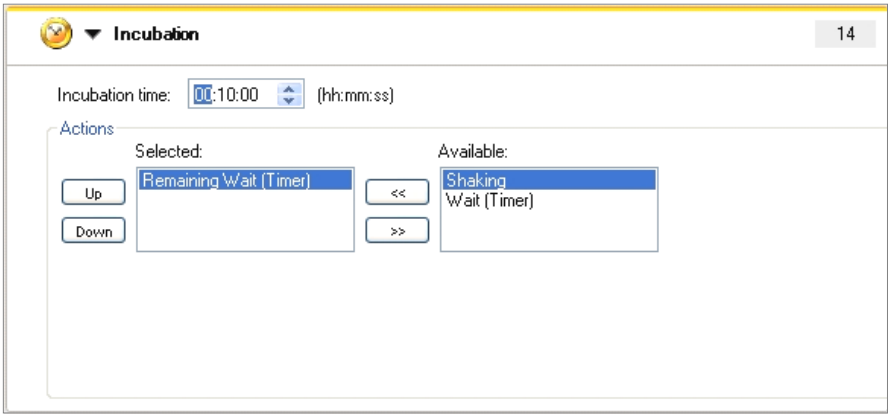
Incubation

Incubation is always done at the heating position to ensure inside the instrument proper temperature distribution.

Incubation can consist of shaking and waiting steps (up to 2 shaking steps and up to 2 waiting steps are allowed in any combination).

The **Remaining Wait** step waits until the overall incubation time is over (including shaking and waiting times).

The incubation program element is typically used to perform shaking and waiting at a certain temperature for a certain time.



The incubation stripe of the **Infinite F50** contains only an input field for the **Incubation time**.

Example for the **Infinite F50**

Enter the appropriate parameters for incubation:

Incubation time	Enter the total time (min. 5 s)
Actions	**Available** actions: **Shaking, Wait (Timer)** 2 wait and 2 shaking actions are allowed. Select actions by double-clicking or use the arrow keys. Organize actions by using the up/down keys. **Remaining Wait (Timer):** mandatory, cannot be deleted or edited (duration 3 s)

2.3 Workflow Pane

The main window in i-control is the **Workflow pane**, where the measurement script is visible and where parameters are defined and edited.

There are two ways to insert a program element from the **Control bar** into the **Workflow pane**:

- Select a program element from the **Control bar**; by double-clicking it, it is inserted into the **Workflow pane** directly after the previous program element.
- Click the program element in the **Control bar** and drag it into the **Workflow pane** to the respective position.

The program elements are numbered according to their sequence.

Once a program element has been inserted into the **Workflow pane**, settings and parameters for this element can be entered or edited.

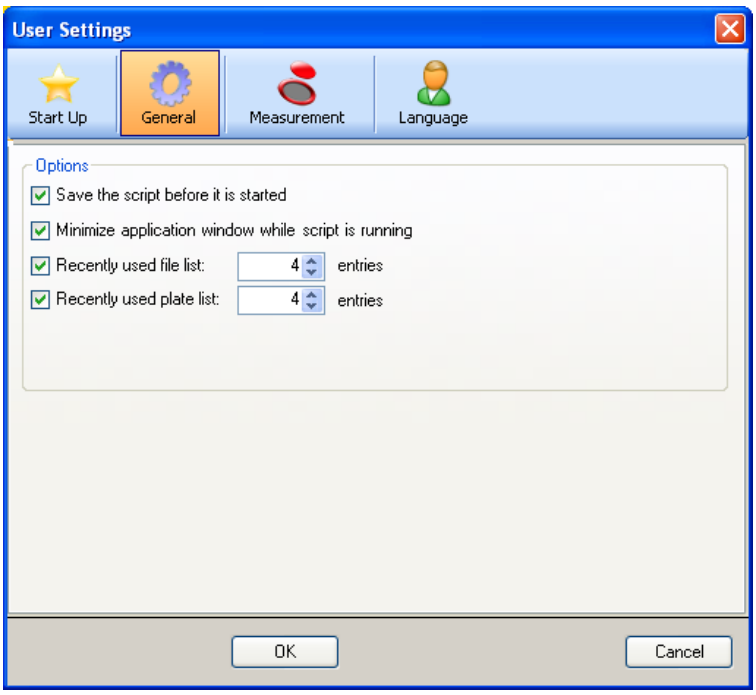
Single program elements inside the **Workflow pane** can be collapsed to display the most important information or expanded to access all editable functions. Click one of the triangles next to the title of the program element, ▼ or ▶, to switch between the two view modes.

By default, i-control starts with the **Plate** element and the **Part of Plate** element in the **Workflow pane**. This can be modified in the **Settings** menu – **User Settings** (see 4.1.5 Settings Menu - User Settings...).

Currently selected program elements within the **Workflow pane** are displayed with a yellow line on the upper border.

If a program element contains errors or is invalid within the current workflow, the element will be flagged with an error mark and the number of the element is highlighted in red. In the **Status bar,** the number of **Errors** appears in red. If the **Info pane** is active, detailed information on the error is displayed. If the workflow contains errors, the measurement script can neither be saved nor started.

It is recommended to always save the workflow before starting a measurement. You can define this feature as default in the **Settings** menu – **User Settings…** – **Options** (Select **Save the script before it is started**).



2.3.1 Hierarchy of Elements

The hierarchy of elements in the **Workflow pane** is as follows:

1. Plate
2. Part of Plate (Range)
3. Well

Any desired measurement step can be inserted directly after a plate, range or well element. Use the **Release** and **Indent** options in the **Edit menu** to modify the sequence of execution of the single strip component. Select an element in the **Workflow pane,** click the right mouse button and select **Release** or **Indent**.

Other elements from the **Control bar** can be inserted into the hierarchy of a workflow as follows:

The first **Range** element is inserted directly after the **Plate** element; then all subsequent **Range** elements can be inserted.

Well elements can only be inserted directly after a **Range** or a **Plate** element.

Only measurement steps of the same mode (e.g. absorbance only with different wavelengths) are allowed within one well element.

Kinetic steps are possible within a **Plate**, **Range** or **Well** element.

Dispense steps are possible within a **Plate** or **Range** element.

Injections steps are possible within a **Well** element.

User Request, Comment, Wait and Wait until temperature is reached steps are possible within a **Plate**, **Range** or **Well** element.

2.4 Info Pane

The **Info pane** on the right side of the screen displays information that is relevant for the currently selected program element. Any warnings and errors are shown.

3. Defining Measurements

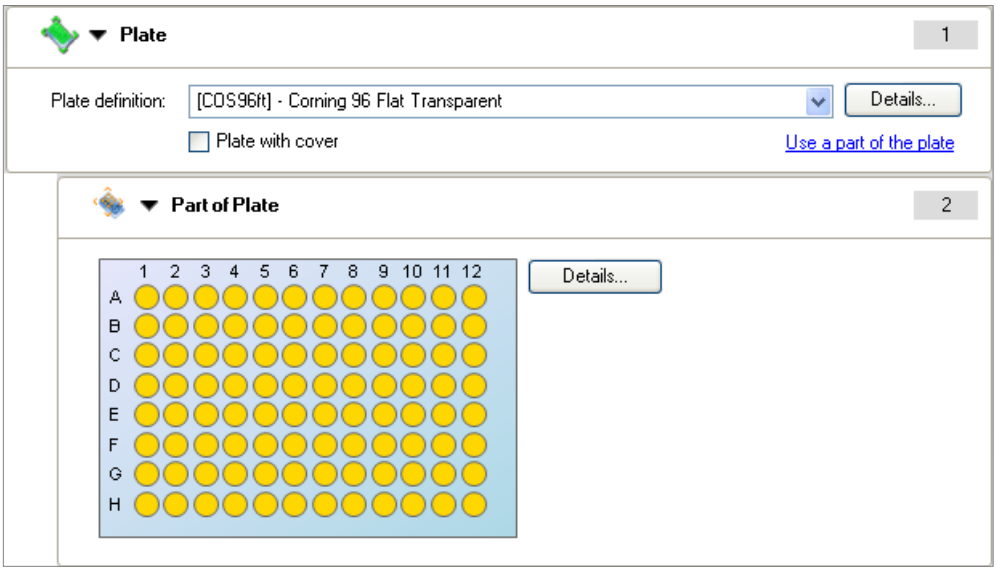
The following chapter describes some examples to illustrate the definition of different measurements.

The **Infinite M1000** and **Infinite M1000 PRO** offer the **Quick-Start-Script** button in the front right corner on the top cover of the instrument. It may be used to start favorite measurement scripts directly from the instrument.

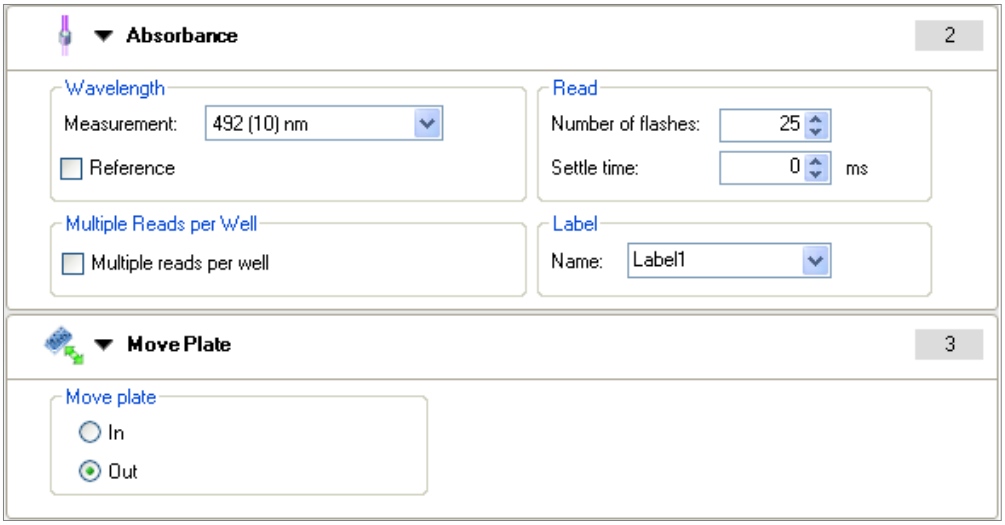
3.1 Defining End Point Measurements

The following example describes an **Absorbance End Point Measurement** in all wells of a 96 well plate:

1. Select a 96 well plate (e.g. Greiner 96 Flat Transparent) from the **Plate definition** drop-down list. If the **Part of Plate** program element is not visible, click the link Use a part of the plate. It is recommended to use the **Part of Plate** program element in every workflow, even if all wells are measured.
2. Double-click the **Absorbance** program element from the **Control bar**, and define the **Workflow** as follows:
3. **Wavelength/Measurement: 492 nm**
4. **Read/Number of reads/flashes: 25 (per well)**
5. **Settle time** (time between moving the plate and starting the measurement): **0 ms**:



If the plate shall be moved out of the instrument after measurement, insert a **Move Plate** program element and select the **Out** radio button.



If a **Move Plate** program element is not defined after the measurement, the plate will stay inside the instrument until **Move Plate Out** is clicked.

After finishing the definition as described above start the measurement by clicking the 

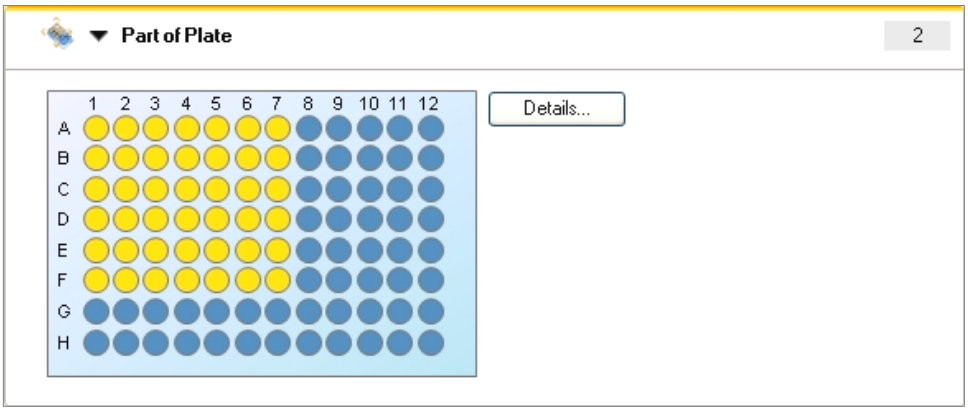
button on the toolbar.

When clicking the **Start** button, Excel opens automatically and the results are displayed in a worksheet.

3.1.1 Plate Size – Part of the Plate

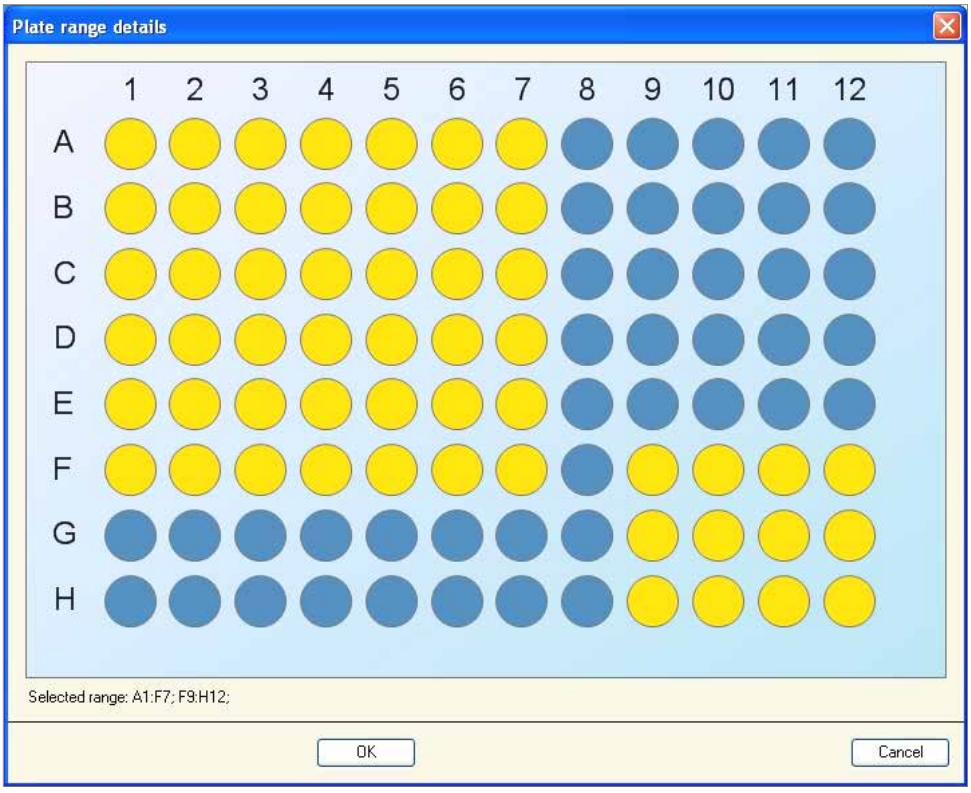
Use the **Plate** program element in the workflow pane to choose a plate format. Select the desired plate format from the **Plate definition** drop-down list (e.g. a black Greiner 96 well plate).

To measure a particular well or a range of wells on the plate click the link Use a part of the plate. In the **Part of Plate** program element click the desired well or drag a frame over the range of desired wells (e.g. A1 to F7). The selected wells are displayed in yellow; unselected appear in blue.



Wells can be selected by dragging a frame over the plate. Further ranges can be selected by holding down the Ctrl key on the keyboard and dragging another frame around the wells to be selected.

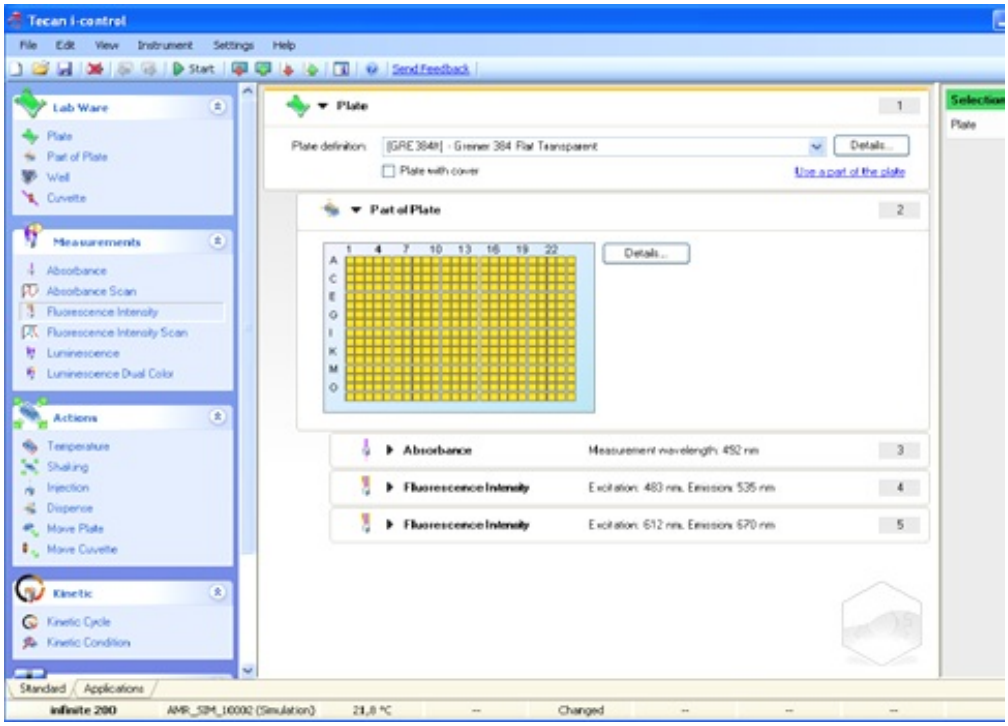
By clicking on **Details...** the plate is zoomed in; well selection can be done also in the zoomed window.



3.2 Defining Multilabel Measurements

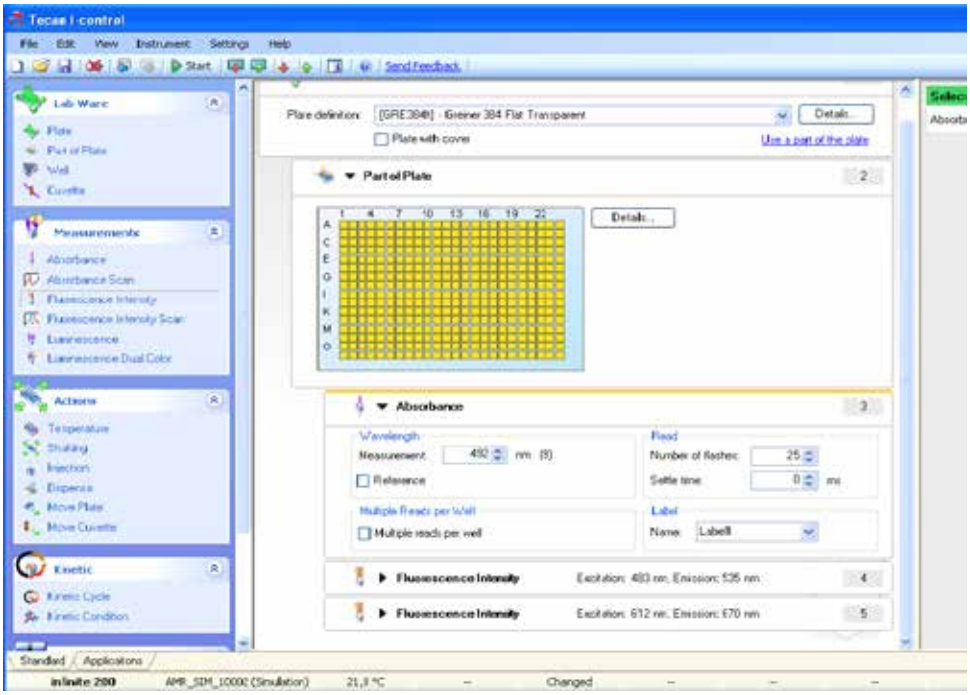
Multilabel measurements are measurements with multiple consecutive reading modes, e.g. with multiple absorbance, fluorescence, luminescence labels or with mixed measurements.

The following example describes the definition of a multilabel measurement in a 384 well plate:



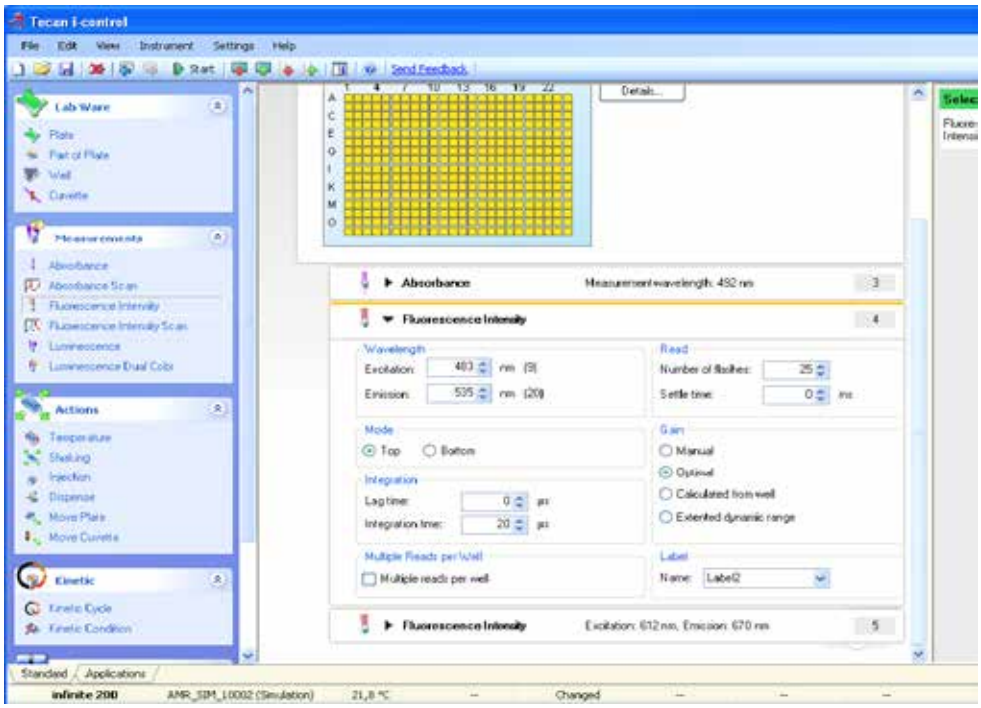
Label 1 – Absorbance Measurement in all wells

1. Select a 384 well plate (e.g. Greiner 384 Flat Transparent) from the **Plate definition** drop-down list; select all wells in the **Part of Plate**.
2. Insert the **Absorbance** program element from the Control bar, and define as follows:
3. **Wavelength/Measurement: 492 nm**
4. **Read/Number of reads: 25**



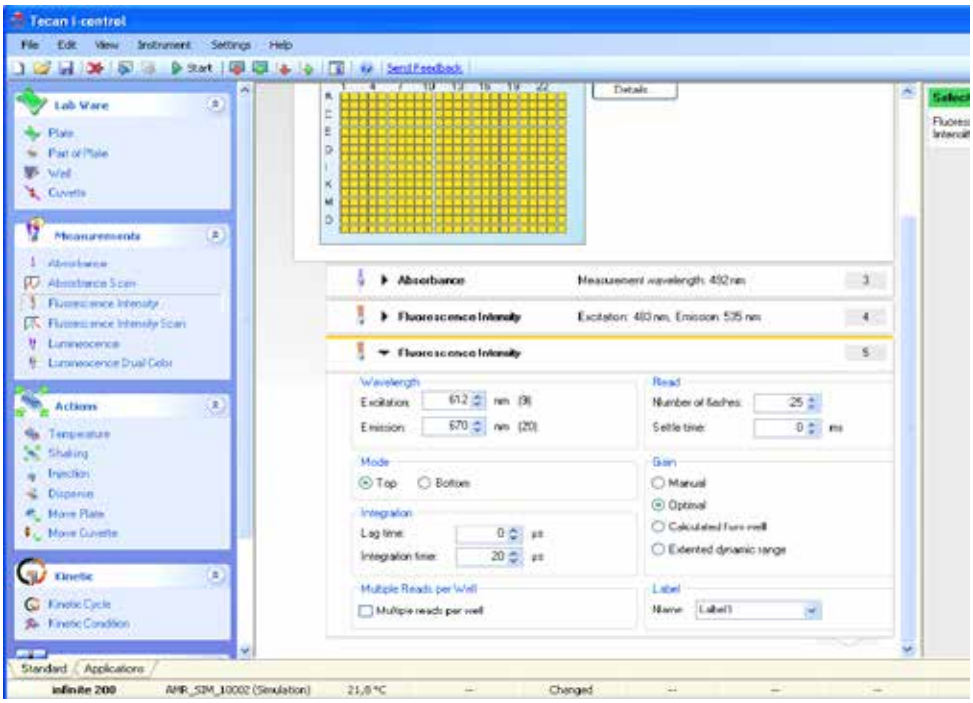
Label 2 - Fluorescence Intensity in all wells

1. Insert the **Fluorescence Intensity** program element from the **Control bar** and define as follows:
2. **Wavelength/Excitation: 483 nm**
3. **Wavelength/Emission: 535 nm**
4. **Read/Number of reads: 25**
5. **Gain: Optimal**



Label 3 – Fluorescence Intensity in all wells

1. Insert a second **Fluorescence Intensity** program element from the **Control bar** and define as follows:
2. **Wavelength/Excitation: 612 nm**
3. **Wavelength/Emission: 670 nm**
4. **Read/Number of reads: 25**
5. **Gain: Optimal**



After finishing the definition as described above start the measurement by clicking the 

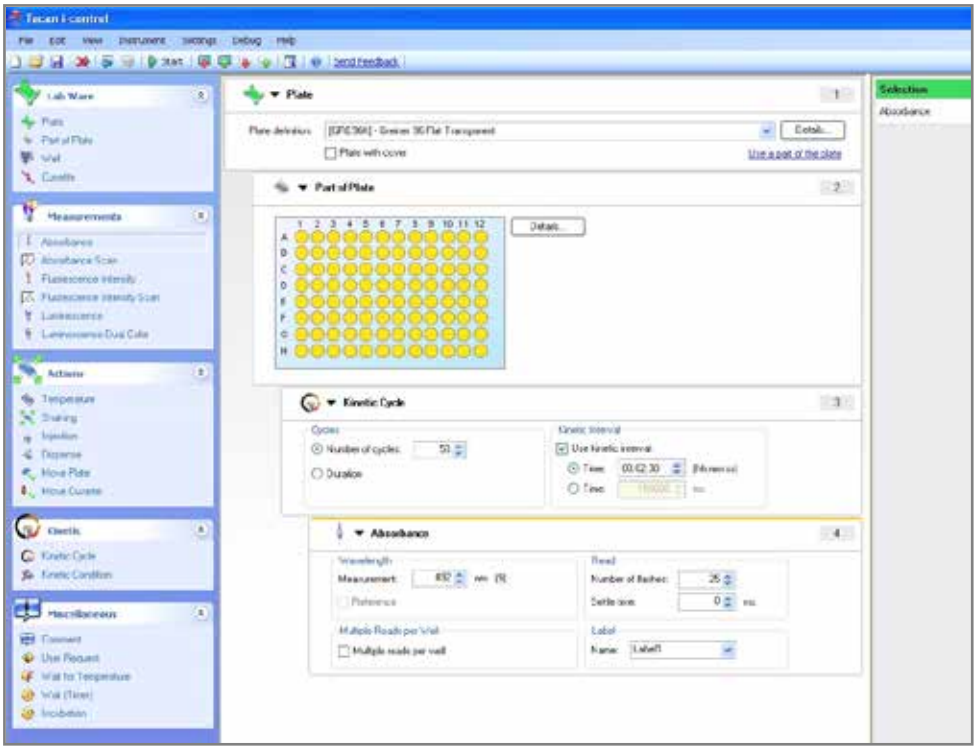
button on the toolbar.

When clicking the **Start** button, Excel opens automatically and the results are displayed in a worksheet.

3.3 Defining Kinetic Measurements

The following example describes a kinetic measurement of a 96 well plate:

1. Select the 96 well plate (e.g. Greiner 96 Flat Transparent) from the **Plate definition** drop-down list, and select all wells in the **Part of Plate** program element.
2. Double-click the **Kinetic Cycle** program element and define as follows:
3. **Cycles/Number of cycles: 50**
4. **Kinetic Interval** (intervals between measurements): select **Use kinetic interval** and enter: **2 minutes 30 seconds**.
5. Double-click the **Absorbance** program element and define as follows
6. **Wavelength/Measurement: 492 nm**
7. **Read/Number of reads: 25**



After having finished the definition as described above start the measurement by clicking the  button on the toolbar.

When clicking the **Start** button, Excel opens automatically and the results are displayed in a worksheet.

Use Gain Regulation (available for all Infinite readers)

The command **Use gain regulation** is only available for plate-wise kinetic measurements in fluorescence top/bottom and fluorescence polarization mode.

Upon activating **Use gain regulation**, fluorescence kinetic measurements with increasing signals are prevented from running into “OVER” once the samples produce too high RFU values. Instead the initially set gain (manual/ optimal/ calculated from well) is automatically reduced in order to permit the measurement of even very high signals.

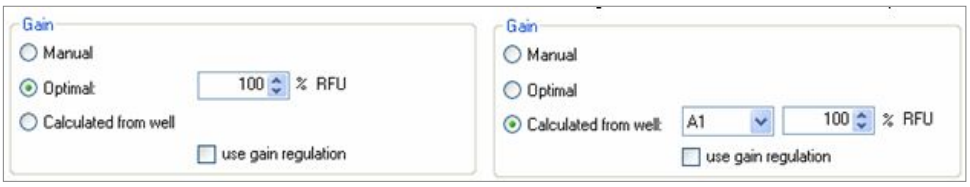
Results that are obtained with different gain settings are highlighted accordingly. All RFU values with different gain settings are automatically correlated, allowing the evaluation of the entire kinetic data within one and the same graph.

Kinetics: x% of Gain (available for all Infinite readers)

The function “x% of … gain” is available for plate-wise kinetic measurements in Fluorescence Top/Bottom and Fluorescence Polarization mode.

The following options are available:

- Start a kinetic measurement with **x% of optimal gain** (optimal gain is calculated in a pre-measurement based on the highest signal within the defined well range on the microplate and set as initial gain for the kinetic measurement)
- Start a kinetic measurement with **x% of calculated from well gain** (the optimal gain setting for one defined well is calculated in a pre-measurement and set as initial gain for the kinetic measurement)



The percentage of the initial gain may be set individually from 20-100%, with 100% being set as default value.

3.3.1 Defining Well Kinetic Measurements with Injections

A **Kinetic Measurement** means that the whole plate is measured in several consecutive cycles with the same settings.

To define a **Well Kinetic**, select **Well** from the **Control bar** by double-clicking or drag the **Well** program element from the **Control bar** into the **Workflow pane** and drop it between **Part of Plate** and **Kinetic Cycles**. If necessary, a **Kinetic interval** can be defined.

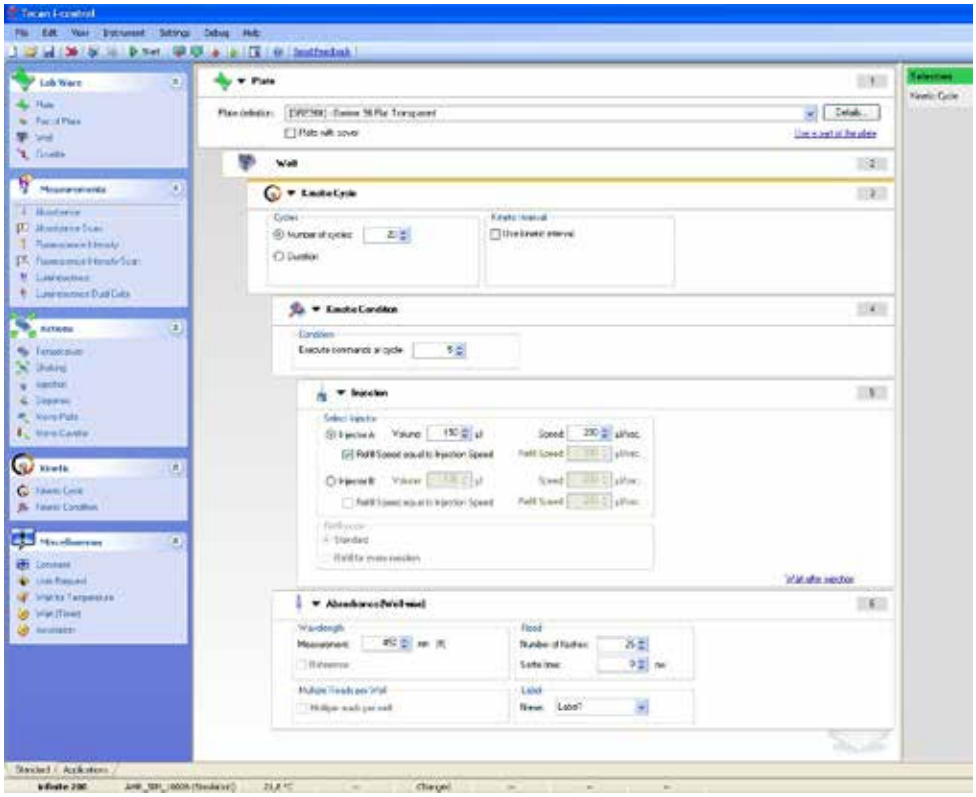
Injectors' parameters can be defined using the **Injection** program element from the **Control bar**. Double-click it or drag and drop it between **Kinetic cycles** and **Absorbance** in the **Workflow pane**. Define volume and speed.

In the **Kinetic Condition** program element, define after which kinetic cycle the injection should be performed. Drag it between **Kinetic Cycle** and **Injection** in the **Workflow pane** and define at which kinetic cycle (e.g. after kinetic cycle 3) the injection (=command) should be executed.

It is very important to **Release** the **Absorbance** program element to the same indentation as the **Kinetic Condition** for kinetic measurements.

See also 3.4 Indenting and Releasing Program Elements and 3.4.1 Ways to Indent or Release Program Elements.

The **Workflow pane** appears as shown in the screenshot:



After having finished the definition as described above start the measurement by clicking the 

button on the toolbar.

When clicking the **Start** button, Excel opens automatically and the results are displayed in a worksheet.

3.3.2 *The Difference between* Inject *and* Dispense

The action which is associated with inserting one of these program elements is identical: a defined volume of a liquid is injected into each selected well. The only difference is the workflow:

Injecting is done well-wise, which means that the liquid is injected into the first well, and then this well is measured as defined, before the liquid is injected into the second well and so on.

Dispensing is done plate-wise, which means the liquid is first dispensed into all wells of the plate, and the whole plate is measured thereafter.

3.4 Indenting and Releasing Program Elements

The decision to indent/ release a program element will modify the workflow of the instrument during measurements.

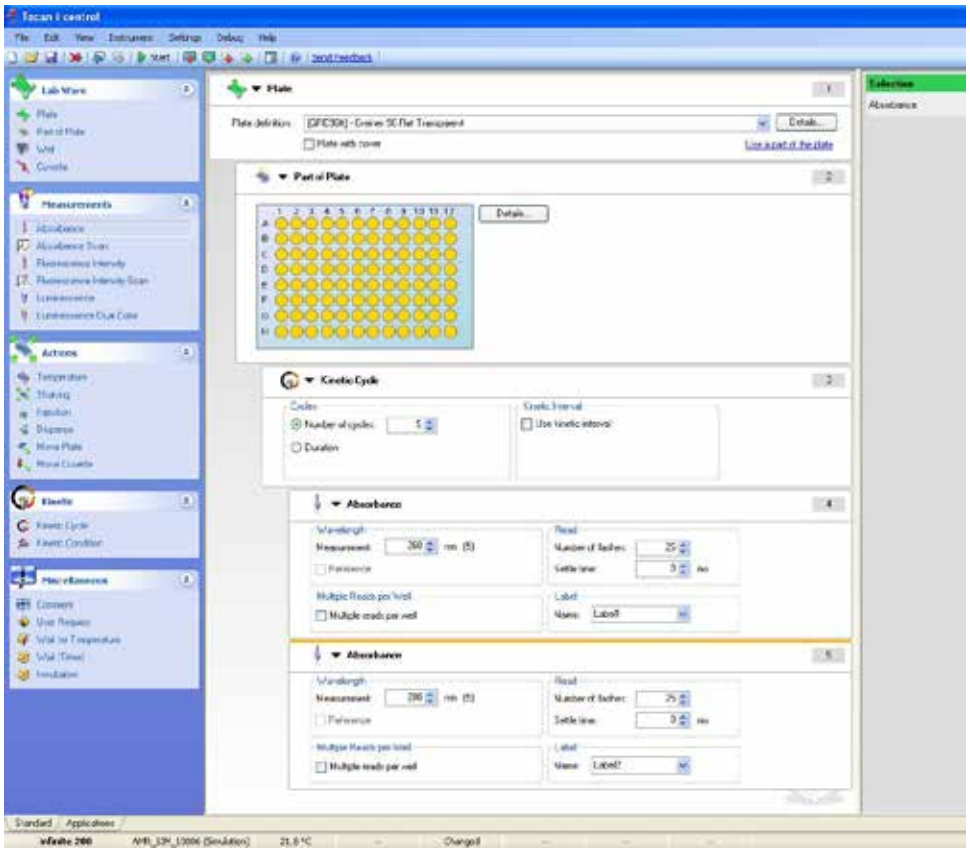
The actions of all program elements with the same indentation are performed sequentially. The only dependence between these program elements is that the next action starts directly after the previous action is finished.

A program element that is indented more than the previous program element shows dependence between the two program elements. This means the parameters defined in the first program element are also active for the second (indented) program element.

The following is an example of how to define a **Multilabel kinetic** with two **Absorbance labels**. The example shows that the two **Absorbance** program elements depend on the **Kinetic Cycle** program element, which depends on the **Part of Plate** program element, which depends on the **Plate** program element. Define the parameters for an example as follows:

1. **Plate**: 96 well plate, e.g. Greiner 96 Flat Transparent
2. **Kinetic Cycle/Number of cycles:** 5
3. **Absorbance/ Wavelength:** 260 nm
4. **Number of reads:** 25
5. **Label Name:** Label1
6. Second **Absorbance/Wavelength:** 280 nm
7. **Number of reads:** 25
8. **Label Name:** Label2

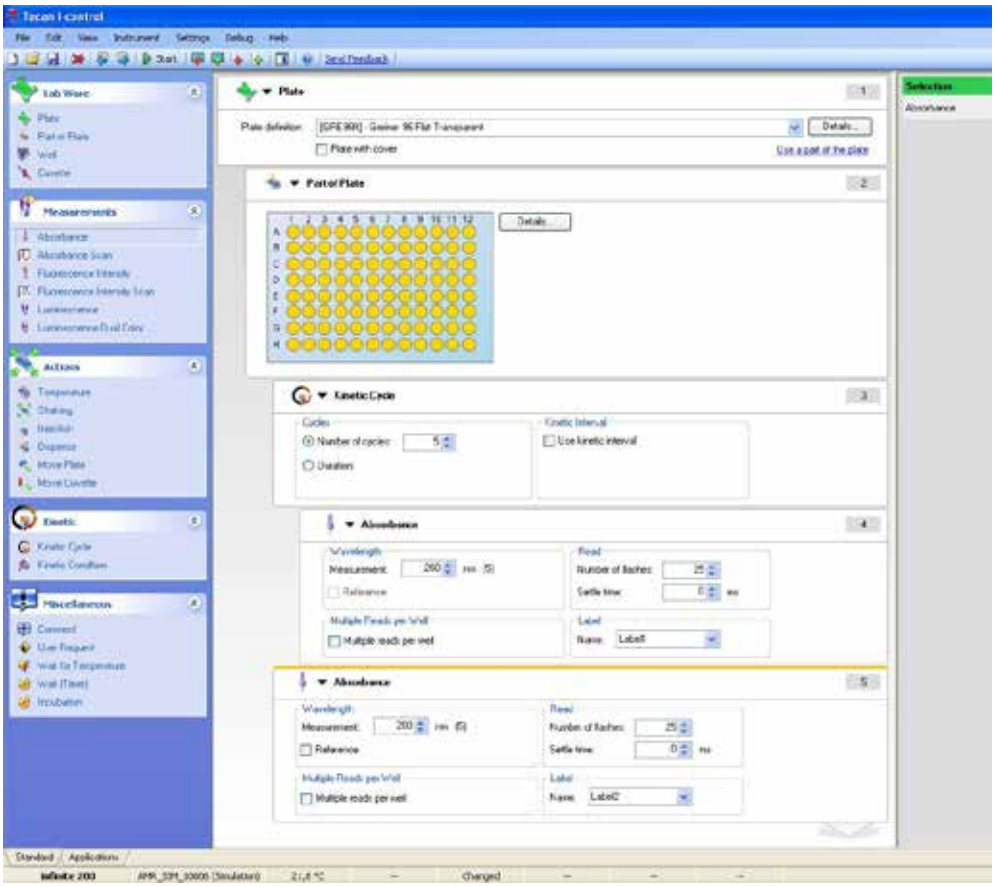
The **Workflow pane** appears as shown in the screenshot:



The above definition results in the following workflow:

The **Absorbance** of all wells of a 96 well plate is first measured at **260 nm** and then at **280 nm**. Both **Absorbance** measurements are performed in 5 kinetic cycles.

Indenting the second **Absorbance** program elements on a level with **Kinetic Cycle** item changes the workflow. Select the second **Absorbance** program element and click the right mouse button. Select **Release Strip** from the context sensitive menu. The **Parameter window** appears as shown in the screenshot:



In this workflow, an **Absorbance Kinetic** measurement with 5 cycles is done first at 260 nm; finished this loop, **Absorbance Endpoint** measurement at 280 nm is performed.

3.4.1 Ways to Indent or Release Program Elements

Select a program element from the Workflow pane.

- Click **Edit** and **Indent/Release**.
- Use the / buttons in the **Tool bar** to release or indent the selected element.
- Click the right mouse button and click **Release** or **Indent**.

4. Menus

4.1 Menu Bar

4.1.1 File Menu

New

This command opens a new measurement workflow. If an empty document is to be opened, you will be asked to save the current workflow.

Click **Yes** to save the current workflow or click **No** to create a new workflow without saving the previous one. Click **Cancel** to leave the dialog box.

Open

This command opens an existing i-control workflow (*.mdfx) from the selected folder. If you want to open an existing workflow while another one is still open, you will be asked if you want to save the workflow. Click **Yes** to save the current workflow to a certain destination or click **No** to create a new workflow without saving the previous one. Click **Cancel** to leave the dialog box.

Save

This command saves the current script.

Save As…

This command saves the current workflow under a different name.

Open from Template (available for all Infinite readers)

Templates are predefined scripts that are similar to common i-control scripts, but contain some additional information, e.g. a short description of the measurement parameters.

Templates may be assigned to distinct groups and may be annotated individually. By default, the **Open from template** dialog opens when i-control is started. The **User settings** dialog contains a checkbox that can be used to hide the **Open from template** dialog by default.

Note
All templates are designed as example scripts for common applications.

It is the responsibility of the user to validate all parameters for the purpose of the particular application before using a template.

All templates are designed as example scripts for common applications.

It is the responsibility of the user to validate all parameters for the purpose of the particular application before using a template.

List of most recently used script files

A list of the most recently saved workflow files is displayed. Define how many files are to be included in this list in the Settings menu à User settings.

Exit

This command exits and closes the program. If you are still connected to an instrument, you will be asked if you want to disconnect and to close the program. Click **Yes** if you want to exit or click **No** if you want to return to the program.

4.1.2 Edit Menu

Cut

This command cuts the selected program element, which can be pasted again.

Copy

This command copies the selected program element.

Paste

This command pastes the selected program element.

Delete

This command deletes the selected program element.

Release Strip

This command releases the selected program element.

Indent Strip

This command indents the selected program element.

Select All

This command selects all program elements in the workflow pane.

4.1.3 View Menu

Info Pane

This command shows or hides the info pane.

Toolbar

This command shows or hides the toolbar.

Status Bar

This command shows or hides the status bar (located at the bottom of the window).

Collapse All

This command collapses all program elements in the workflow pane to view only one line of text.

Expand All

This command expands all program elements in the workflow pane to extended view and shows all visible parameters.

4.1.4 Instrument Menu

Disconnect/Connect

This toggle command connects or disconnects an instrument to or from i-control. To connect to an instrument select the instrument name from the list.

Start

This command starts the measurement process. If the measurement is started, a small window informs that the measurement is in progress. Excel opens automatically and the results are displayed in a worksheet.

Start Stacker Run

If the reader is connected to a **Connect** stacker, it is possible to perform batch processing. Select **Start Stacker Run** and the defined i-control script is performed on all available plates in the input stack.

Movements…

Choose this command to define plate, cuvette and filter movements.

Click **Plate Out** to move the plate carrier out or click **Plate In** to move the plate carrier in. Click **Filter Out** to move the selected filter carrier out.

Click **Cuvette In/ Out** to move the cuvette correspondingly.

When a measurement is started, the plate is moved into the instrument automatically.

Heating…

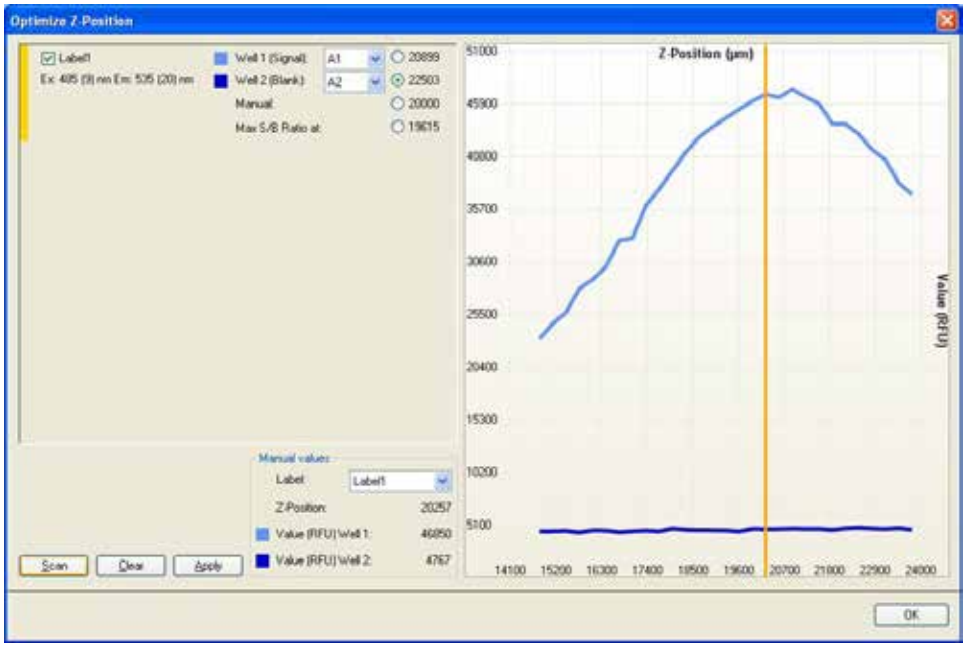
This command is used to set the target temperature of the instrument manually.

Select or enter the **Target temperature** and click **Set and On** to start instrument heating. Click the **Read** button to display the current temperature inside the instrument or click the **Auto** check box to have it read automatically. Click **Off** to stop heating.

Click the down button,  , to display the heating graph and click the up button,  , to hide it. Click the close button,  , to exit the **Heating** dialog box.

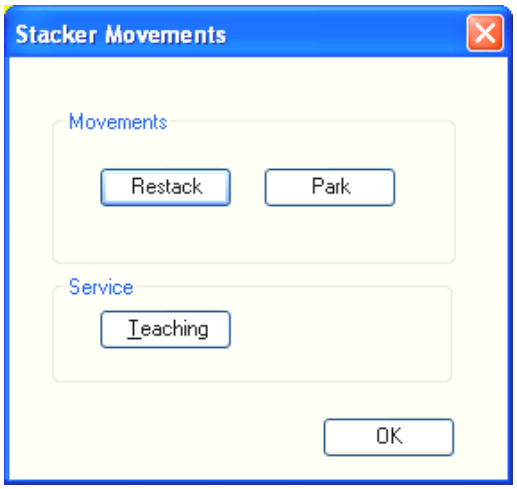
Z-Position

For a detailed description of optimizing the Z-position, refer to the Instructions for Use of the **Infinite F500, M1000**, **M1000 PRO** and **M200 PRO**.



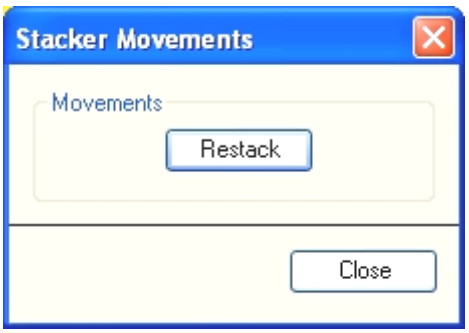
Stacker Control

If the reader is connected to a **Connect** stacker, the **Stacker Control** option appears in the **Instrument** menu.



- Select **Restack** to return the processed plates from the output stack to the input stack in their original order. After **Restack** is selected, a dialog box appears in which the plate type must be selected and confirmed with **OK**, before the restacking procedure is performed.
- Select **Park** to move the gripper into the park position.
- Select **Teaching** to start the Positioning Wizard. For details, see the Instructions for Use for Connect, chapter **5. Positioning Wizard in i-control and magellan**.

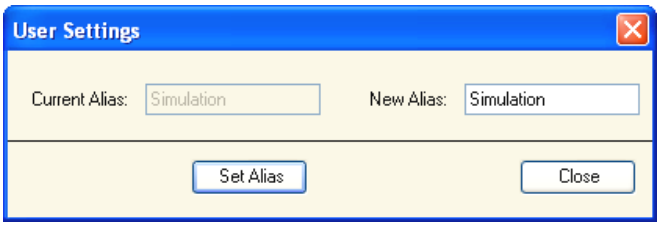
With the **Infinite M1000** and **Infinite M1000 PRO** instruments, only the built-in stacker can be used. If the instrument is connected to a stacker, the **Stacker Control** option appears in the **Instrument** menu:



- Select **Restack** to return the processed plates from the output stack to the input stack in their original order. After **Restack** is selected, a dialog box appears in which the plate type must be selected and confirmed with **OK,** before the restacking procedure is performed.

Properties

Select **Properties** to set a new alias name for the instrument. Enter a new name in the **New Alias** field and click **Set Alias** to confirm.



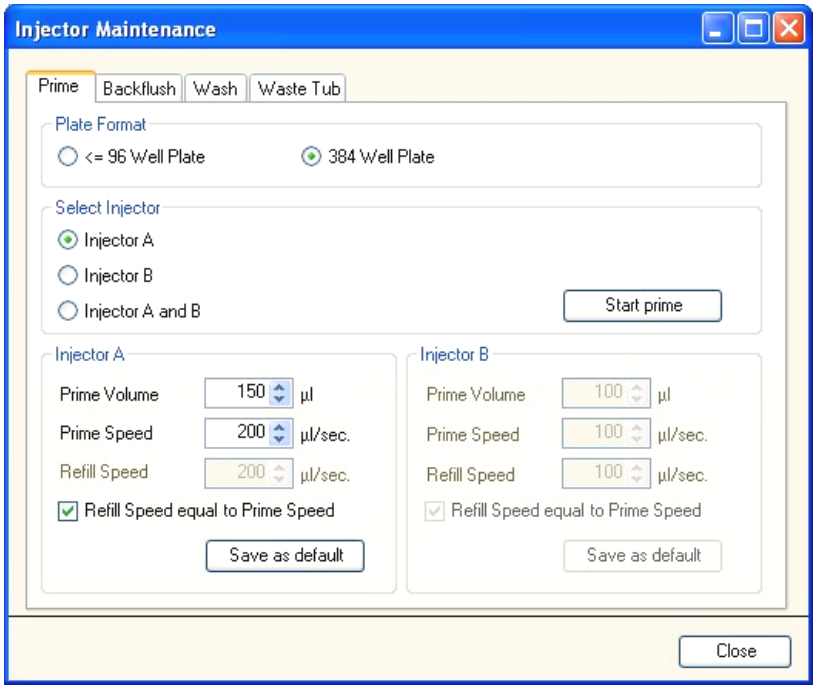
These settings take effect after restarting the software.

4.1.5 Settings Menu

Injectors...

This command opens the injector maintenance dialog box containing the following procedures:

Prime (Example for the Infinite F500)



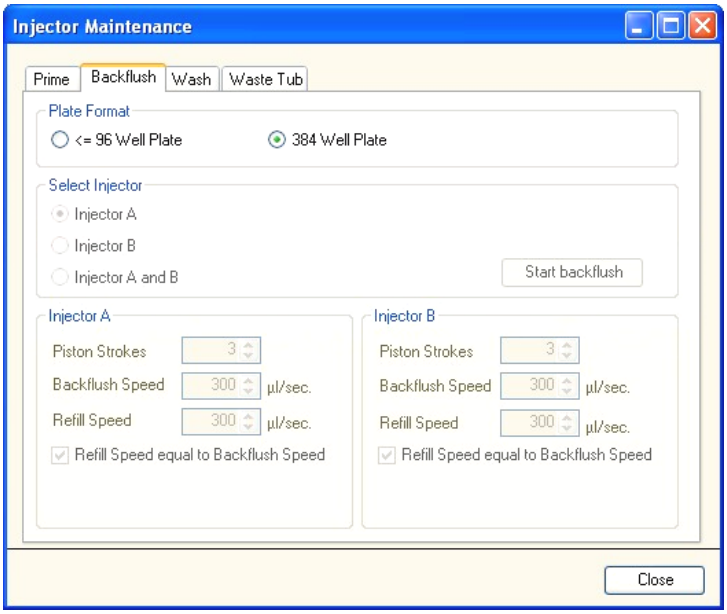
Select injector A, B or both A and B. Depending on which injector is selected the corresponding group box can be edited.

Select the **Prime Volume** and the **Prime Speed** depending on the instrument connected.

Click **Start prime** to start the priming procedure.

Refer to the Instructions of Use of the connected instrument for further details and examples.

Backflush (Example for the Infinite F500)



Select injector A, B or both A and B. Depending on which injector is selected the corresponding group box can be edited.

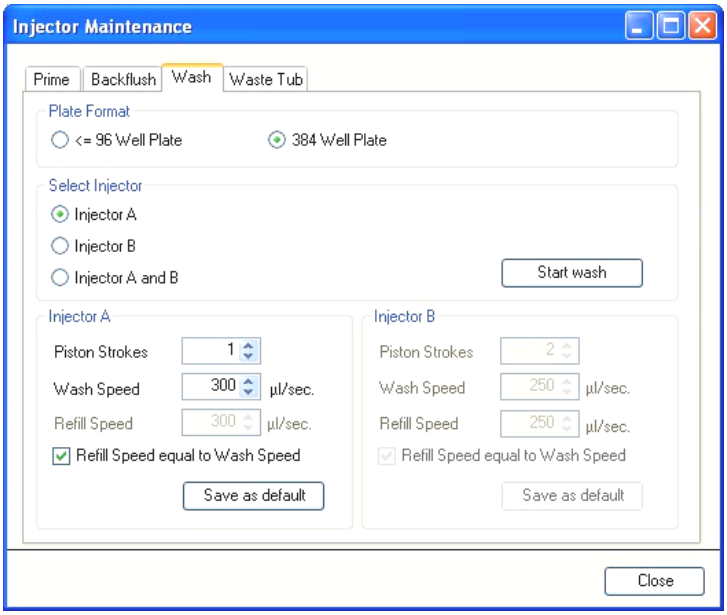
Select the **Piston Strokes** and the **Backflush Speed** depending on the connected instrument.

One piston stroke corresponds to the total volume of the used injector syringe.

Click **Start backflush** to start the reagent backflush procedure.

Refer to the Instructions of Use of the instrument connected for further details and examples.

Wash (Example for the Infinite F500)



Select injector A, B or both A and B. Depending on which injector is selected the corresponding group box can be edited.

Select the **Piston Strokes** and the **Wash Speed** depending on the connected instrument.

Click **Start wash** to start the washing procedure.

Waste Tub

Click **Empty Waste Tub** only when the waste tub has been emptied manually. The software will then alert the user if the waste tub needs to be emptied again.

Refer to the Instructions for Use of the connected instrument for further details and examples.

Filter Definitions (Infinite F200, F200 PRO, F500)

Select the appropriate filter position and enter the new wavelength, bandwidth, and measurement mode for each new filter:

Measurement Mode:	Choose from the dropdown list **FI** for fluorescence intensity, **ABS** for absorbance measurements, **FP** for fluorescence polarization, **ALPHA** for AlphaScreen/AlphaLISA (F200 PRO only), and **Empty** for filter-free positions.
Wavelength:	Enter the filter wavelength. For fluorescence intensity and fluorescence polarization measurements, set the filter wavelength within the allowed range of the connected instrument. Absorbance filters are definable between 230 and 1000 nm (Excitation only).
Bandwidth:	Enter the bandwidth (nm) of the filter.
Description:	This field can be used for individual user remarks about the filter, e.g. filter name, application, etc.
Purchase Date:	This option enables the user to enter the purchase or installation date of the filter.
Flash Counter:	The flash counter monitors the number of flashes through a filter. The flash counter number provides the user only with additional information about the filter in use. For a new filter, set the counter to 0. For a previously used filter, enter the last collected flash number if the number is available. The flash counter number is saved together with other information about the filter on the filter slide microchip. If you replace a filter, this information will be lost unless the last filter flash number is manually documented by the user.

Confirm the new filter values by clicking **Save**. Close the Filter Definition dialog and the system is ready to perform measurements with the new filters.

Refer to the Instructions of Use of the connected instrument for further details and examples.

Plate Definition...

This command allows you to choose a plate file from the drop-down list of available plates. The plate definition files contain all relevant parameters of a specific plate type, e.g. coordinates of measurement points, number of columns, number of rows, well form, well diameter, plate height, plate height with cover…).

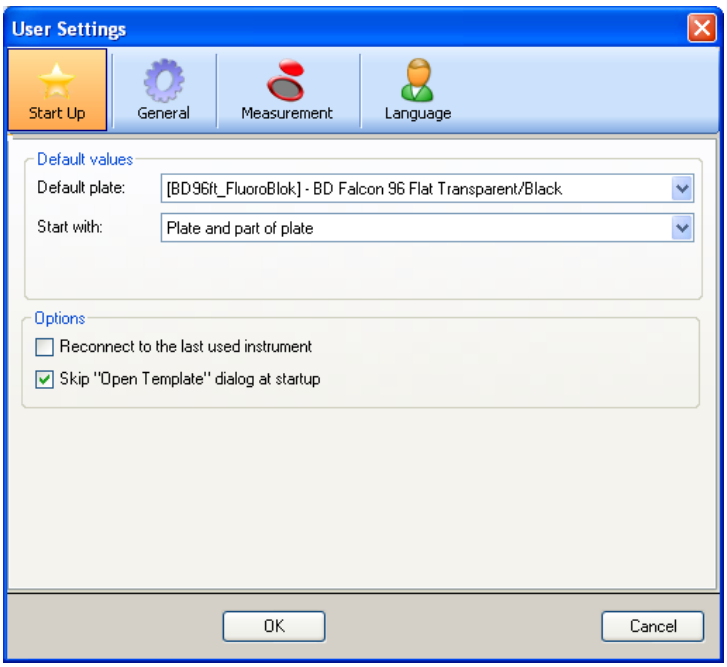
A graphic element at the bottom of the dialog visualizes the parameter which is currently defined.

The available plate types are dependent on the instrument connected.

To make a custom plate definition file, choose one from the list as a template. After the appropriate settings have been defined, save it under a different name.

Click **Save as** to save the selected plate definition as a *.pdfx-file.

User Settings...



Tab Start Up:

Behavior at start up can be set.

1. Select a default plate.
2. Determine if the workflow pane should start with an empty workflow, plate only, or plate and part of plate.
3. Select whether the last used instrument should be reconnected
4. Select whether the ‘Open Template’ dialog at startup should be skipped.

Tab General:

General options can be set.

1. Ask to save the workflow (when changed) before the measurement starts.
2. Determine if i-control window should be minimized while the measurement is performed.
3. Determine the length of the list of recently used plate files (combo box for plate selection in the plate program element).
4. Determine how many recently used workflow files are to be listed in the file menu.

Tab Measurement:

Certain measurement settings can be saved as default settings.

1. Absorbance: Select default number of flashes;
   Pathlength correction: Select default value for test wavelength and default value for manual correction factor.
2. Fluorescence: Select default number of flashes, default value for manual z-position and default value for manual gain.

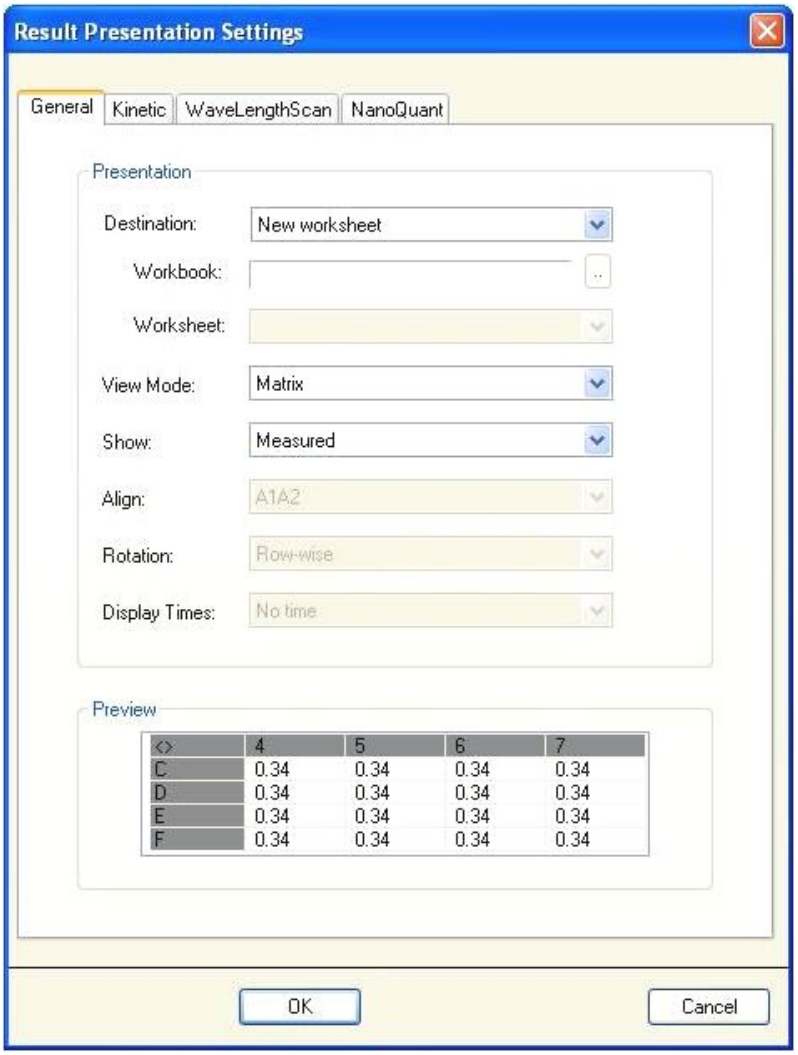
Tab Language:

1. Select the language of the i-control software (English and German are currently available).

Click **OK** to save your settings or click **Cancel** to leave the dialog box without saving any changes.

Result Presentation...

This command offers the following tabs to determine the output settings of the measured results in Excel:



Depending on the connected instrument, different tabs are visible. The **Infinite F500**, **M1000** and **M1000 PRO** have for example an additional tab for fluorescence polarization.

General	Presentation:
	Destination: Select between **New workbook**, **New worksheet, Use previous worksheet** or **Use existing workbook**. If **New workbook** is selected, a new workbook is opened every time a measurement script is performed. If **New worksheet** is selected, a new worksheet of the existing workbook is created. If no workbook is open a new one is created. If **Use existing workbook** is selected, a workbook and a worksheet must be selected. First select the workbook (an Excel file), and then select the sheet the results should be placed into. **View Mode:** Select between **Matrix** and **List**. If **Matrix** is selected, the data alignment corresponds to a microplate; times per well cannot be displayed. Not relevant for kinetic result presentation. If **List** is selected, choose between: Align, Rotation, Display Times.

Note
The option Use previous worksheet must not be used with i-control versions lower than version 1.5.

	Show: Select between **All** and **Measured**. If **All** is selected, the whole plate geometry, including all possible rows and columns, is displayed. If **Measured** is selected, only the results of the measured wells are displayed. **Align:** Select between **A1A2** or **A1B1**. If **A1A2** is selected, the results are arranged in rows (of the microplate). If **A1B1** is selected, the results are arranged in columns (of the microplate). **Rotation:** Select between **Columnwise** or **Rowwise**. If **Columnwise** is selected, the results are displayed in a column (in the Excel sheet). If **Rowwise** is selected, the results are displayed in a row (in the Excel sheet). **Display Times:** Select between **No time** or **Time per well**. If **No Time** is selected, only the values are displayed. If **Time per well** is selected, a timespan for each value is displayed.
Polarization	**Result:**
	Show polarization: Shows polarization data **Show anisotropy:** Shows anisotropy data **Show total intensity:** Shows total intensity data
	Intermediates:
	Show parallel intensity: Shows parallel intensity data **Show perpendicular intensity:** Shows perpendicular intensity data **Show parallel raw data:** Shows parallel raw data **Show perpendicular raw data:** Shows perpendicular raw data

Kinetic	**Result:**
	Rotation: Select between **Columnwise** or **Rowwise**. If **Columnwise** is selected, the results are displayed in a column (in the Excel sheet). If **Rowwise** is selected, the results are displayed in a row (in the Excel sheet). **Align:** Select between **A1A2** and **A1B1**. If **A1A2** is selected, the results are arranged in rows (of the microplate). If **A1B1** is selected, the results are arranged in columns (of the microplate). **Display Times:** Select between **Time per cycle** and **Time per well**. If **Time per cycle** is selected, a timespan per cycle is displayed. If **Time per well** is selected, a timespan for every well is displayed.
	Cycles:
	Range: Select **All** to display all cycles. **Specified range** is currently not available.
Wavelength Scan	**Result:** **Show Wavelength Scan data**
	Wavelength:
	Presentation: Select between **Wavelength over well** or **Wells over wavelength**. If **Wavelength over well** is selected the wells are displayed in a column (in Excel) and the appropriate wavelength data in the row.
	If **Wells over wavelength** is selected the wells are displayed in a row (in Excel) and the appropriate wavelength data in the column below. **Align:** select between **A1A2** and **A1B1**. If **A1A2** is selected the results are arranged by rows. If **A1B1** is selected the results are arranged by columns.
	Show Wavelength chart
	This command appends an Excel chart per well to the worksheet; in this chart, values over wavelength are displayed (X-axis: wavelength, y-axis: values).
NanoQuant	**Show Raw Data** Select the **Show Raw Data** box to display the raw measurement values of Nucleic Acid Quantification and Labeling efficiency measurements.

Exception History…

The **Exception History** dialog box shows a list of exceptions (instrument errors, software failures) with date and time.

Every time an exception occurs and an error box is displayed, all relevant information is collected and saved in a zip-file. Each of these zip-files leads to an entry in this list.

Relevant information is: The error message and number, communication log-files and system information (like operating system version, free amount of disc space).

Every entry (which corresponds with a zip-file) can be saved as a separate file to a user-defined location using the floppy disc symbol at the lower left corner of the dialog box.

This information is helpful to the customer support or help desk to track problems.

4.1.6 Help Menu

Contents

This command opens the online help file and allows you to browse through the different topics.

Index

This command opens the online help file and allows you to enter the first letters of your search query; a selection of help topics will appear.

Search

This command opens the online help file and allows you to enter your search query.

Tecan Homepage

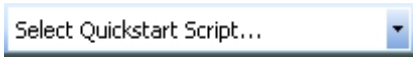
This command opens your favorite browser and navigates to the Tecan homepage.

About…

This command lists the version numbers of the software and hardware components of the currently installed i-control.

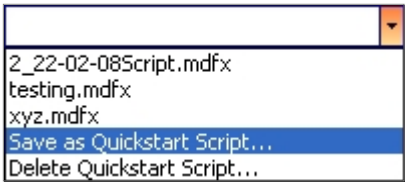
4.2 Toolbar

The following commands are accessible via the toolbar:

	Opens a new measurement workflow
	Opens an existing file
	Saves the current workflow
	Releases the selected program element
	Indents the selected program element
	Starts the measurement
	Starts Stacker Run (only available with stacker)
	Connects or disconnects an instrument
	Moves plate out
	Moves plate in
	Moves cuvette out (M200, M200 PRO)
	Moves cuvette in (M200, M200 PRO)
	Moves filter out (F200, F200 PRO)
	Moves ExFilter out (F500)
	Moves EmFilter out (F500)
	Restacks (only available with stacker)
	Parks gripper (only available with stacker)
	Opens heating dialog
	Shows or hides the info pane
	For starting favorite measurement scripts directly from the instrument (M1000, M1000 PRO)
	Opens the i-control help file
	Opens i-control webpage

Select Quickstart Script (M1000, M1000 PRO)

The currently visible workflow can be saved and started directly from the instrument:



When the favorite script has been saved and is active in the text field, pressing the Quick-Start-Script button on the instrument will start this script.

Saved favorite scripts can also be deleted.

5. Batch Processing

5.1 Introduction

If the reader is connected to a **Connect** stacker, it is possible to perform batch processing. The defined i-control script will be performed on each of the available plates in the input stack.

CAUTION
DO NOT USE MICROPLATES WITH COVERS, WHEN USING THE CONNECT STACKER TO PERFORM BATCH PROCESSING.

Note
The defined script will be performed on each of the available plates in the input stack. It is not possible to run the entire stack through more than once per script.

With the **Infinite M1000** and **Infinite M1000 PRO** instruments, the built-in stacker can be used.

Please refer to the respective Instructions for Use.

5.2 Microplate Requirements for Batch Processing

The use of plate types is limited according to the specifications of the connected instrument; see the corresponding Instructions for Use for details.

Any common microplate ranging from 6 to 1536 well formats conforming to the ANSI/SBS standards (ANSI/SBS 1-2004; ANSI/SBS 2-2004, ANSI/SBS 3-2004 and ANSI/SBS 4-2004) may be used with the **Connect** or built-in stacker for batch processing.

Microplates with covers cannot be used with the stacker.

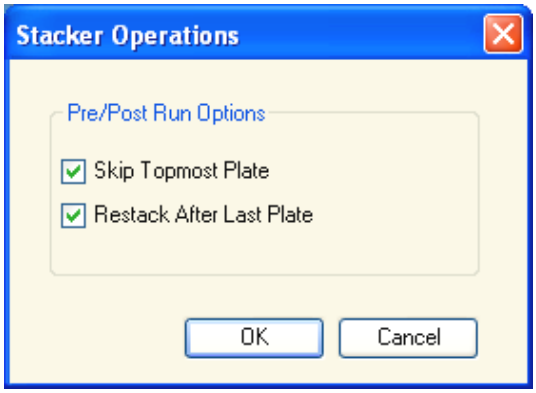
Parameters	Characteristics
Overall plate height	From 7.3 mm to 20 mm **Infinite M1000**, **Infinite M1000 PRO**: from 7.0 mm to 23 mm
Footprint	Length = 127.76 mm ± 0.5 mm Width = 85.48 mm ± 0.5 mm
Minimum difference between plate height and skirt height	≥ 6 mm (only relevant if a Connect stacker is installed)

5.3 Start Stacker Run

Once a script has been defined, batch processing can be started by selecting **Start Stacker Run** from the **Instrument** menu or by clicking the

button on the toolbar. The output stack of the Connect stacker must be empty before starting a run.

The **Stacker Operations** dialog box appears:



- Select **Skip topmost plate** if the plate shall be neglected for measurement. The topmost plate will not be processed and will be moved to the output stack.
- Select **Restack after last plate** to return all plates in their original order to the input stack after all plates have been processed.

Click **OK** to confirm the settings and start batch processing.

Excel opens automatically and the measurement results of each plate measurement will be saved in a separate worksheet. If **Read barcode** has been selected in the **Plate** program element, the worksheets will be named according to the corresponding barcode number; otherwise they will be named **Plate 1, Plate 2** etc.

CAUTION
IF THE READER IS OPERATED WHILE POSITIONED ON THE CONNECT STACKER BUT WITHOUT USING THE CONNECT STACKER, MAKE SURE THAT THE GRIPPER IS IN THE PARK POSITION AND DOES NOT HINDER ANY OF THE READER'S MOVEABLE PARTS (E.G. PLATE CARRIER, CUVETTE CARRIER, FILTER SLIDE, ETC.).

When starting a stacker run with injector the following message appears in the software:

"The software tracks the filling volume of the waste tub and stops the stacker run as soon as the filling volume becomes too high.

To process the maximum number of plates it is recommended to use **Refill for every injection** dispense/injection mode.

For well kinetic measurements it is recommended to use the maximum possible refill volume for **Standard** injection mode."

If the stacker run stops because the waste tub is full, the following message appears:

"Stacker run stopped because waste tub is full."

Empty the waste tub and start the stacker run again with the remaining plates.

5.4 Restacking

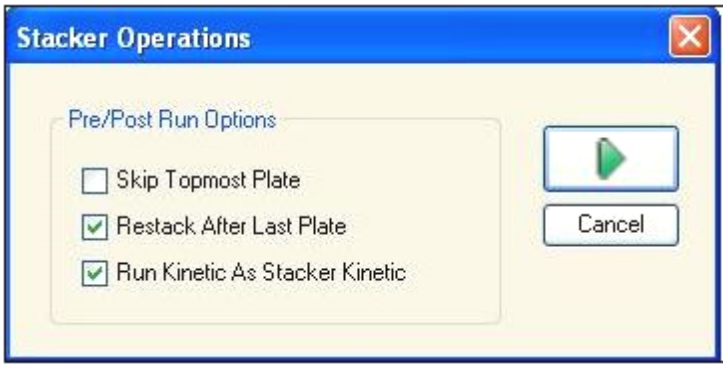
The **Infinite M1000** and **Infinite M1000 PRO** allow restacking of plates without a preceding measurement. Restacking is also possible when the input stack contains plates.

5.5 Stacker Kinetics (available for Infinite 200 PRO, F500, M1000 and M1000 PRO)

In contrast to kinetic measurements on one plate, stacker kinetics allow for the analysis of multiple plates in a time-dependent manner. After all plates in the input stack have been measured (cycle 1), the plates are automatically restacked in their original order and measured again until the user-defined number of cycles is completed on all plates. A maximum of 300 cycles is possible. To facilitate data evaluation, a separate results sheet is generated for each plate and named according to the plate number or barcode (if installed). Results of subsequent cycles are automatically added to the corresponding results sheet.

Stacker kinetics are operable with any plate-wise kinetic measurement script, and combinable with all available kinetic conditions. Note that temperature settings can only be maintained when the plate is located inside the instrument, not in the input/output stack.

In order to perform a stacker kinetic measurement, the workflow / script can be set up in the same way as a usual kinetic measurement and started using the button **Start Stacker Run**. A **Stacker Operations** dialog opens to provide access to additional functions specific for stacker measurements. By selecting the box **Run Kinetic as Stacker Kinetic,** the script is automatically executed as a stacker kinetic measurement.



6. Gas Control Module (GCM) Enhanced Support

6.1 Introduction

The i-control software supports data logging and data display for the **GCM Enhanced**, which is an optional module for **Infinite F200 PRO** and **Infinite M200 PRO** devices.

Note
Data logging and data display
do not work in conjunction with stacker applications.

6.2 Prerequisites

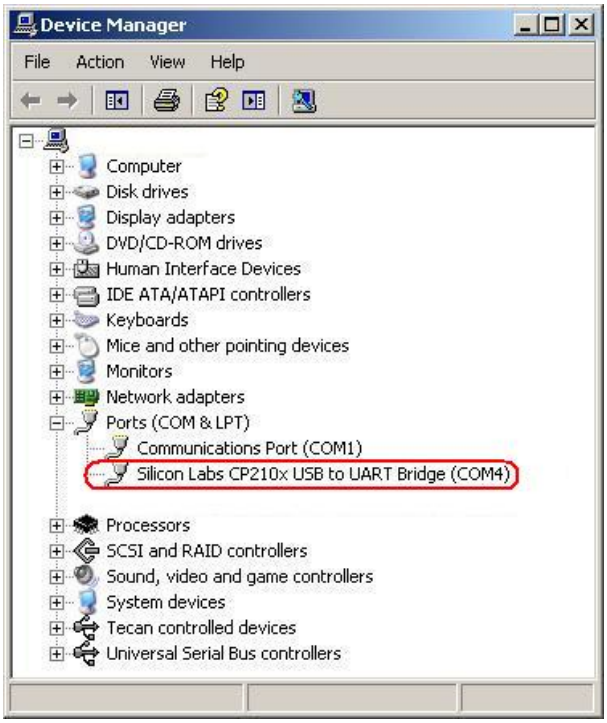
In order to enable communication between the **GCM Enhanced** and i-control, you have to install the Virtual Com Port (VCP) driver from the i-control data carrier (CD-ROM).

Furthermore, you have to connect the **GCM Enhanced** to your PC via the USB cable enclosed with the module.

In order to verify that the **GCM Enhanced** is connected properly, when using Windows 7, navigate to Start > Settings > Control Panel > System and Security > System > Device Manager.

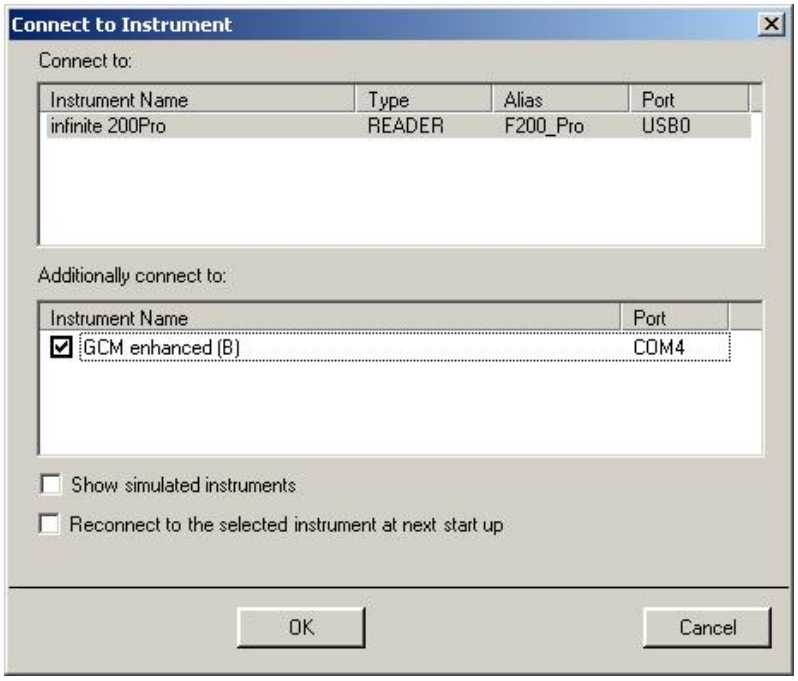
When using Windows 8 go to: Apps > Control Panel > System > Device Manager.

Within the **Device Manager**, navigate to Ports (COM & LPT) and check for an entry similar to “Silicon Labs CP210x USB to UART Bridge”.



6.3 Connecting to GCM Enhanced

Once the VPC driver is installed and the **GCM Enhanced** is connected to the PC, the **GCM Enhanced** appears in the **Additionally connect to:** section of the **Connect to:** dialog box:



6.4 Data Logging

i-control starts logging data provided by the **GCM Enhanced** when a measurement is started (and from then on every 30 seconds), until the measurement is finished.

The data is written into a log file called **GCM-log_YYYY-MM-DDThh-mm-ss.txt.** YYYY-MM-DDThh-mm-ss stands for date and time of log file creation.

Log file name example: GCM-log_2012-01-01T12-34-56.txt

i-control creates a separate log file for each measurement.

The location of the log file depends on the operating system:

- On Windows XP computers, this log file is stored in C:\Documents and Settings\All Users\Documents\Tecan\LogFiles\i-control\ 1.12\ <Instrument_Serial_Number>\.
- On Windows 7, Windows 8.1 and Windows 10 computers, this log file is stored in C:\Users\Public\Documents\Tecan\LogFiles\i-control\1.12\ <Instrument_Serial_Number>\.

A log file line contains the following information, separated by a semicolon:

Name	Description
Date/Time	Date and time when log entry was created
Version	Version of the **GCM Enhanced**
Mode	This entry can have one of the following values: **CO2**, **O2**, **DUAL**, **MANUAL**, **SETTINGS** or **STANDBY**. **SETTINGS:** the alias (device name) of the **GCM Enhanced** and the altitude are displayed. **STANDBY** indicates that no mode is selected or activated. For detailed information about the other modes, refer to **Infinite200 PRO** manual.
Alias	Alias name of the device. Possible values: **A**, **B**, **C** or **D**.
Conc. O2	Current O2 concentration in %
Conc. CO2	Current CO2 concentration in %
Target Conc. O2	Target O2 concentration in %
Target Conc. CO2	Target CO2 concentration in %
Altitude	Altitude in m
Status O2	Possible values for this entry are **ValidData** or **InvalidData**. **ValidData** indicates that the O2 sensor is working **InvalidData** indicates that the O2 sensor might be missing, unplugged or broken.
Status CO2	Possible values for this entry are **ValidData** or **InvalidData**. **ValidData** indicates that the CO2 sensor is working **InvalidData** indicates that the CO2 sensor might be missing, unplugged or broken.
Status Alarm	Possible values for this entry are **Normal** or **Alarm**. **Normal** indicates that the target concentration is normal or the selected mode does not require a target concentration (e.g. mode **Manual**) **Alarm** indicates that the target concentration has not been reached within 20 minutes or deviates for more than 10 minutes during operation.

Log file line example:

2012-01-01 12:34:56;TECAN GCM enh. V1.01;MANUAL;A;20.5;0.1;15;0.5;400;
ValidData;ValidData;Normal

6.4.1 Importing Logged Data into Microsoft Excel

The content of the **GCM Enhanced** log file can be imported into Microsoft Excel for further evaluation.

In order to make sure that the numeric data imported into Microsoft Excel maintains the correct number format, it may be necessary to define the following in Microsoft Excel:

Custom System Separators

- Define "**.**" (period) as the **Decimal separator**.
- Define any other character which is not required as separator, e.g. "**,**" (comma) as the **Thousands separator**.

Delimiters

Import the log file (.txt file) to Excel as a **Delimited** file type.

Select "**;**" (semicolon) as the **Delimiter**. The delimiter, is the character used to separate fields.

Data Format

Select **General** as the **Column data format**. "General" converts numeric values to numbers, date values to dates, and all remaining values to text.

6.5 GCM Enhanced Data Displayed in Status Bar

When the GCM Enhanced is connected via the i-control software, some of the data is displayed in the i-control status bar at the bottom of the application window. This data is updated periodically every 30 seconds.

Depending on the on the **GCM Enhanced** configuration and the selected mode, i-control displays either the current CO2 and O2 concentrations or the current CO2 concentration only.

For further information about **GCM Enhanced** configurations, refer to the Instructions for Use for the **Infinite200 PRO**.

If **GCM Enhanced** is in standby, **GCM Standby** is displayed.

If connection to the **GCM Enhanced** is lost (e.g. because the module has been turned off or unplugged while i-control is running), **GCM Module Error** is displayed.

To remove the error, plug in or turn on **GCM Enhanced**. **Disconnect** the **Infinite 200 PRO** reader and reconnect reader and **GCM Enhanced** with the i-control software via **Connect**.

6.6 GCM Enhanced Data Displayed in Excel

When the **GCM Enhanced** is connected via the i-control software, the current CO2 and O2 concentrations are written to the Excel measurement result sheet.

Depending on the on the **GCM Enhanced** configuration and the selected mode, i-control writes either the current CO2 and O2 concentrations or the current CO2 concentration only.

In endpoint measurements, the concentrations are written into the Excel at the start of the measurement.

In kinetic measurements, the concentrations are written into the Excel at the start of each cycle.

If **GCM Enhanced** is in standby, no concentration values are written into the Excel.

6.7 Precautions before Starting a Measurement

Heating must be activated (at least 30°C, 86°F) during incubation to maintain a stable gas atmosphere.

The plate carrier compartment should be closed until the target concentration is reached.

When reconfiguring the Mode setting of the **GCM Enhanced**, wait at least 30 seconds before starting the measurement, so that i-control can update the **GCM Enhanced** data properly.

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