

lumanette USER MANUAL

Incubator microscope for live cell imaging



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Notes on the user manual





Warning of electrical voltage

This symbol indicates that electrical hazards pose a danger to life and health of persons.

Warning

The signal word indicates a hazard with a medium level of risk which, if not avoided, may result in death or serious injury.

Attention

The signal word indicates a hazard with a low level of risk which, if not avoided, may result in minor or moderate injury.

Note

The signal word indicates important information (such as property damage), but not hazards.



Protective gear

Notes with this symbol indicate that your personal protective equipment is to be worn. Wear safety goggles, gloves, protective clothing and safety shoes.

Legal notice

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guarantee the correctness oft he measured values or measurement results. Furthermore, InnoME GmbH assumes no liability for errors or damage resulting from the use oft he measured values.

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Warranty and liability

The device complies with the essential health and safety requirements of the relevant EU regulations and has been repeatedly tested at the factory for proper function. When claiming the warranty, the device number must be stated (see back of the device). Failure to follow manufacturer's instructions, regulatory requirements, or unauthorized modifications to the equipment, or unauthorized replacement of any part of the equipment, may significantly affect the electrical safety of this product and may void the warranty. Any liability, property damage or personal injury resulting from the device being used contrary to the instructions in this operating manual is excluded. Technical design and design changes due to continuous development and product improvement are reserved at any time without prior notice. For damages that result from improper use, no liability is assumed. Warranty claims then expire as well.

Safety

Please read this manual carefully before putting the device into operation and using it and always keep it in the immediate vicinity of the place of installation or on the device!



Warning Read all safety instructions and instructions.

Failure to follow the safety instructions and instructions may result in electric shock, fire and/ or serious injury.

Retain all safety instructions and instructions for the future.

This device may be used by children 14 years old and over, and by persons with reduced physical, sensory or mental capabilities, or lack of experience and knowledge, if they have been supervised or instructed in the safe use of the device and understand the hazards arising therefrom. Children are not allowed to play with the device. Cleaning and user maintenance must not be performed by children without supervision.

- Do not operate the device in potentially explosive areas.
- · Do not operate the device in an aggressive atmosphere.
- The device is not a toy. Keep children and animals away.
- Never immerse the device completely in liquid.
- Protect the device from direct sunlight.
- Do not remove safety signs, stickers or labels from the device. Keep all safety signs, stickers and labels in a readable condition.
- · Do not open the device with a tool.
- Observe the storage and operating instructions (see chapter Technical data).
- If it can be assumed that the device can no longer be operated without danger, it must be taken out of operation and secured against being restarted.
- The customer/ user may not make any changes or repairs. For maintenance or repair, the unit must be returned to the manufacturer or to a location specified by the manufacturer.
- The safety of the user may be impaired by the device if it has visible damage, is no longer working as prescribed or has been stored in unsuitable conditions. In case of doubt, the device should always be sent to the manufacturer or a body appointed by the manufacturer for inspection, maintenance or repair.

Intended use

Only use the device for the microscopy of biological samples and for continuous monitoring of them. Any further use and the utilization of the measurement results of the device are the responsibility of the user. To use the device as intended, use only accessories approved by InnoME or parts tested by InnoME.

Improper use

Do not use the device in potentially explosive atmospheres, with hazardous substances or in contaminated areas. InnoME assumes no liability for damage resulting from improper use. Warranty claims expire in this case. Unauthorized structural changes and attachments or conversions to the device are prohibited.

Staff training

The use of the device is reserved for technically trained personnel. Respective persons must also have read and understood the operating instructions, in particular the Safety chapter.

Residual risks



Protective gear

Wear protective equipment such as gloves, eye protection and protective clothing. Depending on which other products, substances or chemicals you use, further protective measures are necessary. Pay particular attention to the respective safety data sheets for chemicals before using them.



Warning

Do not leave the packaging material careless. It could become a dangerous toy for children.



Warning

The device is not a toy and does not belong in the hands of children.



Warning

This device may pose a hazard if it is used improperly or not as intended by unauthorized persons! Note the personnel qualification.

Note

To avoid damaging the unit, do not expose it to extreme temperatures.

Device Description

The lumanette is a live cell imaging device. It is a real-time incubator microscope system. Inside, 24 mini-microscope enable a new simultaneous observation of your cell cultures. Take advantage of your own incubator and your laboratory computer. Install the free software*, connect the lumanette via USB 3.0 and it is ready for use. There is no need for an additional power supply. Create a new Project in the Software, put the cells in their defined conditions and monitor them live from outside on your computer screen. The lumanette creates images of your cell cultures in a frequency of your choice and analyses them for cell coverage and cell number. Different microscopic techniques, like Brightfield, Darkfield and even Digital Phase Contrast are available. You can sort and graph your acquired dataset in the lumanette's software or export the dataset and use alternative means at any time, easing the workload of organizing your Data for a Scientific study.

The small footprint, about the size of a standard cell culture plate, leaves you enough space in the incubator for your other cell culture ware or more systems. This allows you to use it in parallel with other experiments at the same time. The housing is completely closed and sterilizable with Isopropanol or other common medias for wipe cleaning.

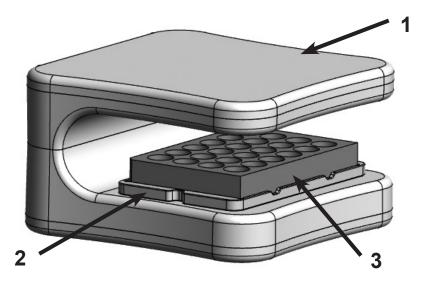
Benefit from a wide range of possible applications and monitor your cells continually.

Application examples:

- · Documentation and analysis of cell growth
- Comparison and analysis of extracellular conditions (e.g. glucose concentration, pH value or different mediums)
- Continiously determining cell confluence in 24 spots
- · Analysing effects of component on cell cultures (e.g. cytotoxicity or biocompatibility)
- Migration or Scratch assays
- Stem cell observation

*You can download the lumanette software from https://www.dropbox.com/s/iiqmqxftl3xaipt/etaluma_lumanettel_0_1.zip?dl=0

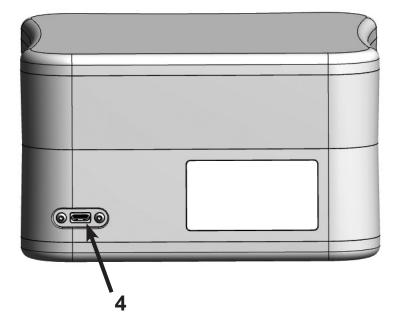
Isometric view



Legend:

- 1. lumanette
- 2. Magnetic adapter frame for fixing cell culture vessels
- 3. Shown: 24-cavity plate, not included
- 4. USB-C socket for data exchange and power supply

Rear view



Connectors & switches

The device can be connected to a computer via the USB-C socket (4) with the supplied USB cable. Only when using a USB cable tested by InnoME, the device is guaranteed to function properly. The device is controlled exclusively via the USB-C interface and a connected computer using the lumanette software.

General technical data

System Requirements	Windows® 7,8 or 10 (32/64 Bit, 2,5 GHz 4 cores, 8 GB RAM)				
Interface (power supply, data exchange)	1x USB 3.0				
Magnification	10 x				
Field of view	1,2 mm x 0,9 mm				
Image Resolution	5 MP				
Display resolution per microscope	2592 x 1944				
Software incl. Algorithm	Cell coverage Cell number (fibroblasts)				
Measuring rate / Adjustable recording interval	5 min.* – 24 h.				
Image capture	30 s for 24 wells				
Export Formats	Picture: PNG, BMP, JPEG/JPG Dataset: CSV				
Light Source	LED				
Camera	5 MP CMOS				
Optical Filters	No Filters				
Operating Temperature	20°C - 45°C				
Humidity -operation	20% - 95% rel.				
Storage Temperature	0°C - 50°C				
Protection	IP62				
Sterilizable	Isopropanol Ethanol				
Dimension (H x W x D)	105 mm x 180 mm x 180 mm				
Weight	1050 g				
Warranty	l year				

* If the minimum system requirement is exceeded, 10 minutes should not be undershot as the smallest adjustable recording interval.

Shipment and Storage

Note: If you store or transport the device improperly, the device may be damaged. Observe the information on transport and storage of the device.

Shipment

When transporting the device, use a suitable container to protect the device against external influences. The original packaging is a suitable transport container. Make sure that the lumanette is not exposed to improper mechanical loads.

Storage

- · Dry, protected from frost and heat
- Not in contact with hazardous substances
- $\boldsymbol{\cdot}$ In a place protected from dust and sunshine
- · If necessary, protect it from dust with a cover
- Storage temperature 0 ... +50 ° C

Delivery

lumanette	x 1
USB-cabel	x 1
Magnetic adapter*	x 1

* among others for Eppendorf 24 cavity plate

Check if the delivery is complete and undamaged. If the shipment shows shipping damage or components are missing, please contact our service (see page 23).



Warning

Do not operate a damaged device as this may cause injury.

Commissioning

The power supply and data exchange take place via the USB-C interface. Only use an InnoME-tested USB cable for commissioning. Only an InnoME-tested USB cable guarantees safe and error-free operation of the device. InnoME assumes no liability for damage resulting from unchecked replacement parts.



Warning

Only connect the device to a terminal that meets the system requirements in the chapter "Technical data". Do not apply overvoltage, otherwise the device may be irreparably damaged.

First use

Please carefully remove the lumanette from the packaging and check that the device and accessories are intact. Connect the lumanette to your computer with the enclosed USB cable. A first initialization of the 24-channel microscope is performed. After successful initialization, the status LEDs will indicate that the device is ready for use by briefly flashing the status LEDs. On the computer, the lumanette is recognized as a camera in the device manager.

Install the lumanette program on your end device that meets the system requirements from the "Technical Data" chapter. This can be downloaded at <u>https://www.dropbox.com/s/iiqmqxftl3xaipt/etaluma_</u> <u>lumanettel_0_1.zip?dl=0</u>. Make sure that the installation directory does not contain special characters except for underscores, hyphens and spaces.

Use

Connect the lumanette to your computer with the enclosed USB cable. After a successful initialization, the green status LED lights to indicate that the device is ready.

When the device is used in an incubator, make sure to place it there at least two hours before the start of an experiment. This serves to compensate differences in temperatures thus ensures stable processes.

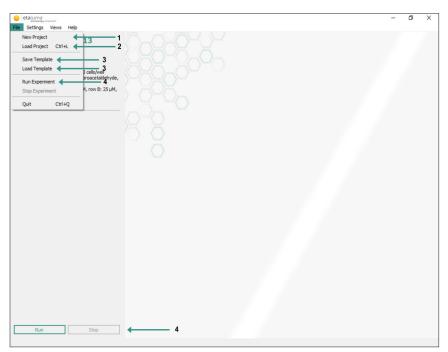
Insert the appropriate magnetic adapter frame with your cell culture vessel and start the software on your computer. Care and cleaning

Care and cleaning

- The device is dust and splash-proofed according to the IP62 standard and can be cleaned and sterilized using a spray-wiping process.
- · Do not use a brush with hard bristles or other hard objects for cleaning.
- Use a soft cloth or a soft sponge to prevent scratches.
- · Do not apply too much pressure and do not use caustic or abrasive cleaners.
- The device is suitable for cleaning with medical cleaning alcohol.
- Apply the disinfectant specifically to the surface to be disinfected.
- · Immediately wipe off after applying the disinfectant.
- Pay attention to a complete wetting of the device.
- Alternatively, the disinfectant can be applied directly to a dry cloth to completely wet the device.

File menu

The File menu item includes actions such as creating or loading projects and templates. Likewise, a long-term observation of a sample can be started and stopped.



1. Create a new project

In order to guarantee the assignability of your recorded and generated data, a project path can be set via the menu item New Project as well as a project name and a description can be added. An assistant will guide you through the most important settings. You can also do this manually at a later time in the **Menu>** Settings> Project Settings.

2. Load project

Experiments already carried out can also be considered retrospectively. For this select the menu item **Load project**. A file dialog will appear, allowing you to navigate to the desired project. Select the **lumanette file** and click **Open**.

3. Save template and load

The menu item **Save Template** allows you to save the settings you have made and thus makes it easier to repeat experiments under the same conditions. The last used camera settings as well as settings for the experiment are saved.

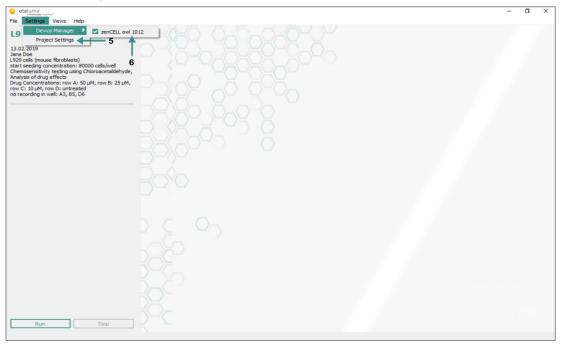
To load the settings, click on **Load Template** and select the desired experiment template in the file dialog box.

4. Start experiment and stop

Run/Stop experiment can be used to start and stop a long-term observation/experiment. While the experiment is running, all captured images are saved in the project folder and the algorithms for the degree of coverage and the cell count are executed. All related information is saved, and a .csv file is created, which stores the percentage of coverage for each image. While the experiment is running, this file must be closed.

If no settings have been made, an image acquisition of all 24 wells will be started at an interval of 10 min.

Menu Settings



5. Device manager

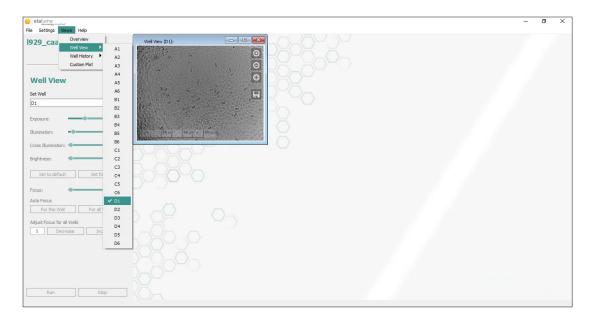
The **device manager** lists all lumanette devices connected to the computer. Click on a lumanette to select it and set it active.

6. Project settings

Project settings allow you to manually change project settings. See also in detail on page 17.

Menu View

In the **View** menu, windows such as **Overview**, **Well View** and **Well History** can be opened and closed. In the **Well View** and **Well History** menu, only the wells set as active are listed. All open windows are checked in the View menu.



Help menu

7. Check for updates

Here a software update can be carried out. You will be automatically redirected to the associated link.

8. Support

If you have additional questions, click **Support**. You will receive the latest version of the operating instructions. The forwarding to the associated link is automatic.

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File Settings Views Help			
L929_caa_20 Support			
13.02.2019 About Jane Doe			
1325 cide (incuse fiteroblast) 1425 cide (incuse fiteroblast) Etat tested) concentration: 80000 cide/vell Chemoentishity testing using Chicroacetaldehyde, Analysis of drug effects Drug Cancentrations: row A: 50 LMP, row B: 25 LM, row C: 13 LMP, row L-stressleet to recording in well A3, 85, DE			
30			
30			
Run Stop			

Project settings

Project settings allow you to make manual settings for your project at any time.

General settings

Project Settings	×
General Settings Experiment Settings	
Project Name: DefaultProject	
Project Directory: Set directory 4 2	
Description:	
	OK Cancel

1. Project name

The **project name** guarantees the assignability of your recorded and generated data by creating a project folder at the specified **project path**.

2. Project path

The **project path** can easily be set via a file dialog box. It may not contain special characters or spaces. Pelase use only characters [a-z] or numbers [0-9].

3. Description

A **description** allows further clarification for the project to be created.

You reach the item **General Settings** via the menu item **Settings > Project Settings** or you follow the Wizard via **File > New Project**.

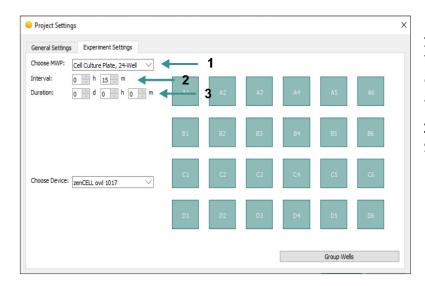
Experiment settings

General Settings	Experiment Settings						
Choose MWP:	Cell Culture Plate, 24-Well 🗸 🗸	←	1				
Interval: Duration:	$\begin{array}{c c} \hline 0 & & h \\ \hline 0 & & v \\ \hline 0 & & v \\ \hline 0 & & v \\ \hline \end{array} \begin{array}{c} h \\ \hline 0 & & v \\ \hline \end{array} \begin{array}{c} h \\ \hline 0 & & v \\ \hline \end{array} \begin{array}{c} h \\ \hline \end{array} \end{array} \begin{array}{c} h \\ \hline \end{array} \begin{array}{c} h \\ \hline \end{array} \end{array} \end{array} \begin{array}{c} h \\ \hline \end{array} \end{array} \end{array} \begin{array}{c} h \\ \end{array} \end{array} \end{array} \begin{array}{c} h \\ \end{array} \end{array} \end{array} \end{array} \begin{array}{c} h \\ \end{array} \end{array} \end{array} \end{array} $	A1	A2	A3	A4	A5	A6
		81	82	B3	B4	B5	B6
Choose Device:	zenCELL owl 1017 \checkmark	C1	C2	C3	C4	C5	C6
		D1	D2	D3	D4	D5	D6
						Group Wel	

1. Camera avtivation

The lumanette is equipped with 24 cameras that can be individually activated and deactivated in the **experiment settings**. Deactivated cameras will not be activated during the experiment. Simply selecting the camera controls their activity.

A preselection is already made by the **setting of the cell culture vessel**, which can continue to be customized individually.

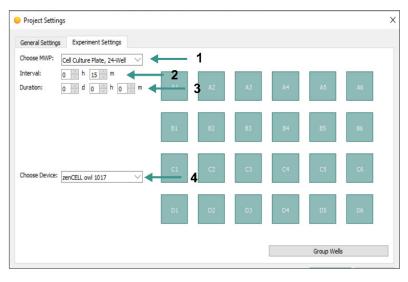


2. Interval

The **interval** is given in minutes and indicates the time span in which a new image acquisition of all activated wells is started.

3. Duration

Set the duration of your experiment.



4. Device selection

Select the device to use the lumanette with which the experiment is to be performed. All lumanette devices connected to the computer are listed in the device selection.

Experiment settings

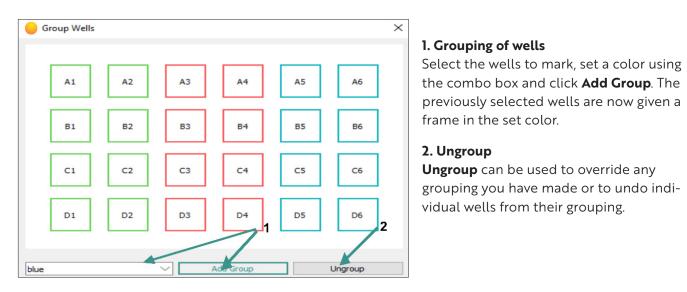
Project Settin	gs							>
General Setting	Experiment Settings							
Choose MWP:	Cell Culture Plate, 24-Well	✓ ←	1					
Interval: Duration:	0 v h 15 v m 0 v d 0 v h 0 v	m 2	3 A2	A3	A4	A5	A6	
		B1	82	B3	B4	BS	B6	
Choose Device:	zenCELL owl 1017	C1	4 C2	C3	C4	C5	C6	
		D1	D2	D3	D4	D5	D6	
			5 -			Group Wel	ls	

5. Grouping of wells

You can colorize Wells and group them together to further illustrate the experiment.

The item **Experiment Settings** can be reached via the menu item **Settings> Project Settings** or you can follow the Wizard via **File > New project**.

Grouping of the wells



The item **Group Wells** can be reached via the menu item **Settings > Project Settings> Experiment Setting** or you can follow the Wizzard via **File > New Project**.

Overview

The **overview** window shows an overview of all available cameras of the selected lumanette device. If groupings have been made via the project settings, the wells are marked in color, as shown in the following figure. Within the well, the last recorded image is displayed.

1. Updating / Save

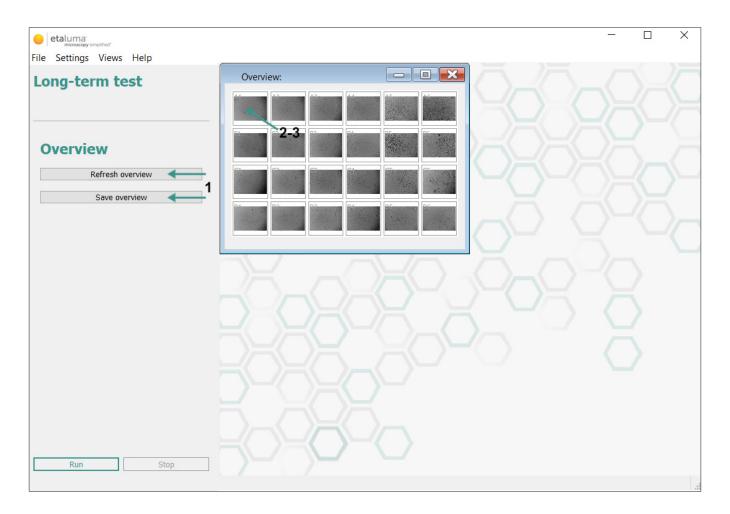
If the view is to be updated, either the **Refresh overview - Button** or the **Save overview - Button** can be made. Refresh will take current pictures of all enabled cameras and refresh the overview. When saving, however, the user will be asked to choose a directory and the recorded images will be archived.

2. Simple click

A simple click in a well opens the corresponding **Well View** window.

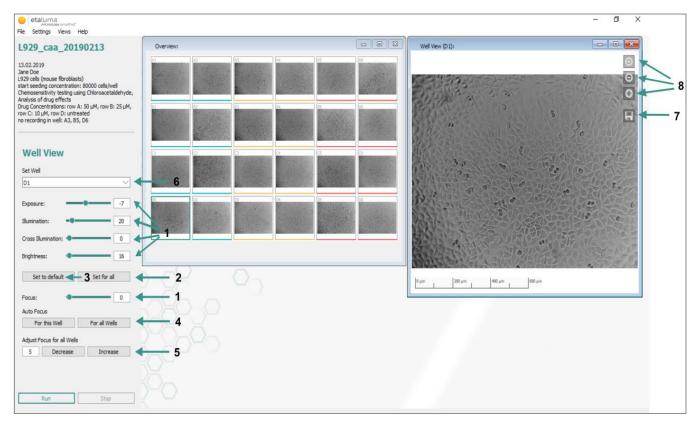
3. Double click

A double-click opens the **Well History** window.



Well View

The Well View window provides a detailed view of the selected well. If the experiment is inactive, the live image of the active camera will be displayed. As soon as the experiment is set active, the setting options in the setup area are deactivated and the detail view shows the last saved image of the corresponding camera. When loading an already completed project while using the same lumanette that was used for this project, the device will be automatically chosen. In this case the Well View window will show the live camera image. If any other or no device is connected, the Well View window shows the last image recorded in this project.



1. Camera settings

Using the setup area, it is possible to flexibly set fine adjustments such as lighting, exposure, brightness and focus via sliders with the mouse. In addition, by left-clicking on the corresponding slider, the value can then be set in steps using the left and right arrows of the keyboard.

2. Save settings

If you have set the camera to the desired configuration, you can transfer it to the other cameras by clicking the **Set for all-Button**. Merely the focus settings will not be applied to the other cameras.

3. Reset to default settings

To reset the configuration to the default, you can click the on **Set to default-Button**.

4. Autofocus

Clicking on the **For this Well-Button** the focus for the selected well will be set automatically. Respectively clicking on the **For all Wells - Button** to all automaticyally focus each well.

5. Focus adjustment

The **Set to default-Button** as well as **Set for all-Button** do not affect the focus settings. If you want to raise or lower the focal plane equally for **all wells**, this can be done by pressing the **Decrease-Button** and **Increase-Button** respectively. Simply enter the desired decrease /increase index and click on the appropriate button. The focal plane can be shifted by 1023 steps.

6. Change well

If the Well View window is to be changed, you can do this by clicking on the desired well in the Overview window or by left-clicking on the Well View window and selecting the desired well via the combo box. The well can also be changed via the **View > Well View** menu. Only one Well View window can be displayed at a time.

7. Image export

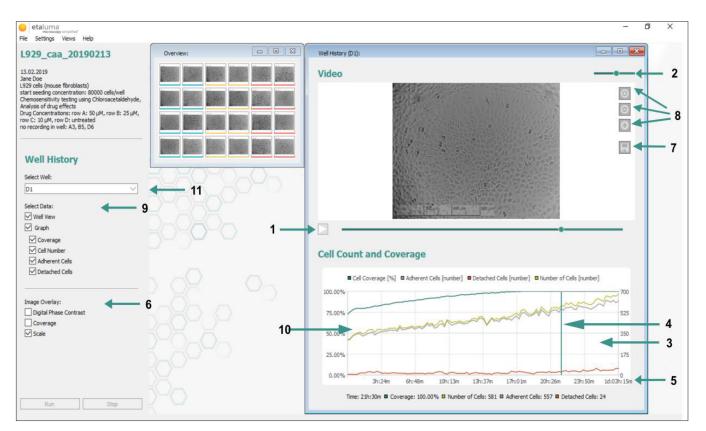
Export possibility of the desired image. A file dialog box sets the location and name.

8. Zoom buttons

Using these buttons, the image can be zoomed in and zoomed out with the digital zoom. Note that this is not an optical zoom. The picture quality is retained.

Well History

The Well History window is the summary of the experiment at the current time.



1. Lapse video

All pictures stored up to this time are put together to a time-lapse video. The experiment can be played in fast motion using the **Play** button.

2. Playback speed

The playback speed can be adjusted via the slider.

3. Graph with the algorithm results

Below the time-lapse video, the results of the selected algorithms are graphically displayed.

4. Data cursor

The marker in the graph indicates the value of the algorithm associated with the displayed image.

5. Data values

The values corresponding to the data cursor are listed below the graph. The point in time is also stated. When the mous pointer is moved over the graph the values correspond to it.

6. Image Overlay

It can be used to control whether the digital phase contrast, the degree of coverage and/or the scale should be shown in the image. A tick activates the corresponding overlay.

7. Image export

Export possibility of the desired image. A file dialog box sets the location and name. The image is saved with the respectively selected overlays.

8. Zoom buttons

Using the buttons, the image can be zoomed in and zoomed out with the digital zoom.

9. Select data

Here the detail view of the Well History window is set. By activating the checkbox the display of images or graphs can be switched on and off. There is the possibility to switch on three different graphs. These include the cell coverage (the corresponding percentage can be read from the left axis), the number of living cells and the number of detached cells (the number can be read off the right axis).

10. Graph export

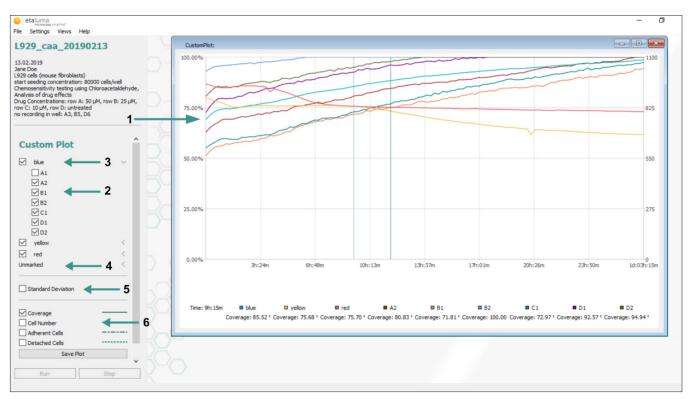
The graph can be exported as a PNG file by right-clicking on it and choosing **Save Plot**. By right-clicking on the graph and choosing **Export Measurement Data** a .csv file will be generated. Both export options only contain the data activated in the graph.

11. Graph export

The desired well can be selected in the current Well History display via the combo box. Further Well History windows can be opened via the **View > Well History** menu or by double-clicking in the desired Well in the Overview window.

Custom Plot

The "Custom Plot" is a simultaneous overview of all graphs, which enables an easy comparison of the grouped measurement curves.



1. Grouped Wells

If groupings were made before, means of the marked wells are calculated and visualized in the "Custom Plot"

2. Selection and deselection of individual measurements

You can select or deselect individual measurements from groupings by setting checkmarks, this way the measurements of their means can be included.

3. Renaming the groupings

By double clicking the groupings' name, you can choose an individual name for this grouping.

4. Ungrouped wells

Wells, which do not belong to any grouping, can be selected under unmarked.

5. Standard deviation

You can select or deselect the standard deviation by setting a checkmark.

6. Selection of graphs

By setting a corresponding checkmark you can choose to visualize between following calculations: cell coverage, cell count, detached and/or attached cells.

7. Save Plot Button

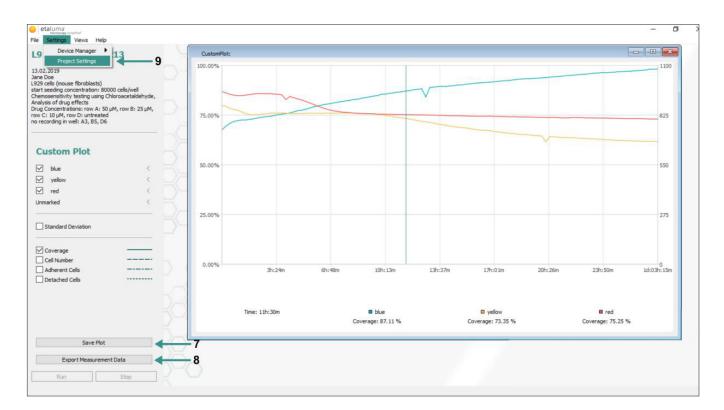
Use the Save Plot button to save the custom plot as an image.

8. Export Measurement Data Button

If a pre-set is chosen for desired wells, their data can be saved as a .csv file by using the "Export Measurement Data" button. This data can be opened with any calculation programme of choice.

9. Change pre-set groupings

You can later change the groupings of the wells under the menu item **Settings >Project Settings> Experiment Settings.**



In case of a defect, please contact info@etaluma.com. www.etaluma.com

Disposal



The crossed-out wheeled bin symbol on a WEEE indicates that it should not be disposed of with household waste at the end of its life. For your free return collection points for WEEE are available near you. The addresses can be obtained from your city or local government. Separate collection of WEEE is designed to enable the reuse and recycling of WEEE, and to avoid adverse effects on the environment and human health from the disposal of potentially hazardous substances contained in the appliances.

The WEEE registration number of InnoME is: DE 43441295.

You are responsible for the deletion of personal data that may be present on the waste equipment to be disposed of.

Pillow Library

The Python Imaging Library (PIL) is

Copyright © 1997-2011 by Secret Labs AB

Copyright © 1995-2011 by Fredrik Lundh

Pillow is the friendly PIL fork. It is

Copyright © 2010-2018 by Alex Clark and contributors

Like PIL, Pillow is licensed under the open source PIL Software License:

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3129 Tiger Run Ct, Ste 218, Carlsbad, CA 92010 Tel. +1 (760) 298-CELL (2355) info@etaluma.com



www.etaluma.com