Lux2 Your best friend in the lab







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Safety Information

To avoid damage or malfunction of the CytoSMART™ Lux2:

- Be sure to set this device on a firm, stable and horizontal surface. The device may break or cause injury if it falls
- Shut down the device immediately if it produces smoke, a strange odor or unusual noise. Continued use may lead to fire. Unplug the USB cable immediately
- Do not connect cables in ways other than those mentioned in this manual, as this
 may cause damage and lead to fire
- If water or other liquid enters the device, discontinue use immediately. Continued use may lead to fire. Unplug the USB cable immediately
- Do not attempt to repair or remove the outer case of the device. If you open this instrument, you put yourself at risk of bodily harm due to electrical shock in addition to damaging the device.
- Never attempt to repair, disassemble or modify this device yourself, tampering with this product may result in injury or fire
- Do not drop the device or allow objects to fall onto it.
- Do not allow the device to be submerged in liquid
- Do not place heavy objects on top of this device and never stand or lean on this device. Failure to follow this may cause the equipment fall or collapse, which results in possible breakage and/or injury
- Do not autoclave the device
- Do not perform the sterilization cycle of an incubator with the device inside





Live Cell Imaging System Instruction Manual

1. Introduction

Technology to Make Your Work Easier

Thank you for purchasing the CytoSMART™ Lux2. Soon you will be monitoring cell cultures from any location at any time. It is not only an easy way to be more efficient, live-cell monitoring and alerts will also save you thousands of dollars by preventing your precious research cells from dying.

This second generation CytoSMART™ Lux2 allows for time-lapse videos of a cell culture next to capturing single images. Real-time information and notifications regarding the cell culture are available, due to instant analysis of the experiment. All results are accessible from any internet browser, which allows you to check-in from any PC, laptop or smartphone. It really is that simple.

How Easy the CytoSMART™ Lux2 Works

The CytoSMART™ Lux2 system can be installed within several minutes and requires minimal training.

Hardware and software work together like a charm. The system consists of a monitoring device, which operates inside a CO_2 -incubator. The device is linked to a Windows tablet or computer. The Lux2 is delivered with a Windows tablet, so no additional hardware is required. Via the tablet, you can customize the imaging settings and get started. The images are then transferred to the CytoSMART $^{\mathrm{TM}}$ Cloud via an internet connection.

All image files are temporarily stored on the tablet to assure data safety. They are then transferred to the CytoSMART™ Cloud, which is accessible via any web browser. In the cloud, it is possible to view and/or download images, videos and other results regarding the cell culture. All your data is right there when you need it.

With the CytoSMART™ Lux2 comes a two-year subscription to the CytoSMART™ Cloud. This includes 50 Gb image storage, access to the cloud and online image analysis.



Ideal Circumstances for Cell Cultures

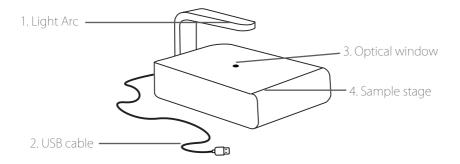
With the CytoSMART™ Lux2, cells can remain in their defined culture conditions inside the incubator while monitoring them online. There is no need to disturb the ideal circumstances of your experiment. The system also provides automatic email alerts regarding the cell culture, e.g. email notifications when a specified cell confluency or temperature is reached.

The CytoSMART™ Lux2 is ideal for monitoring migration assays or hypoxic cell cultures. The system can also be used for cell growth curves and the screening of effects from culture media and growth factors.

System Overview

The hardware of the CytoSMART™ Lux2 is shown below (fig. 1):

- 1. **Light arc** Contains the light source for imaging
- **2. USB cable** For connecting the device via USB, which is also the power source for the device
- **3. Optical window** Cover glass, allowing for image acquisition
- **4. Sample stage** Can hold most commonly used culture vessels



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2. Getting Started

The content of the CytoSMART™ Lux2 package is shown below (fig. 2):

- 1. CytoSMART Lux2
- 2. CytoSMART Lux2 Instruction Manual
- 3. CytoSMART Lux2 Quick Starter Guide
- 4. Cable clips
- 5. License key

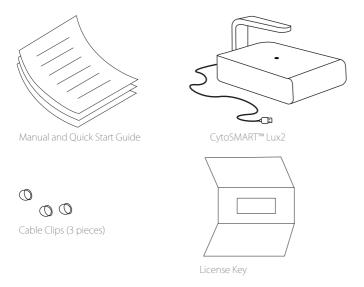


Fig. 2. CytoSMART™ Lux2 package overview.

Remember, if you ever have any questions regarding your purchase, please visit https://www.cytosmart.com/support or contact our support staff via support@cytosmart.com. We are happy to help you out.



3. Setting up the system

It is really easy to set up the CytoSMART™ Lux2 system. All you need are these items:

- An incubator
- The CytoSMART™ Lux2 device
- Cable clips
- A Windows tablet or PC
- An internet connection

Be sure to remove any wrapping before placing the CytoSMART™ Lux2 inside the incubator. Also clean the device with a damp lint free cloth and IPA or EtOH (70%).

Important: Do not use acetone to clean the device and do not autoclave the device.

Cable Placement

Check if the incubator has a cable port in the back.

- If so, guide the USB cable from the CytoSMART™ Lux2 through it (fig. 3a)
- If no port is present, place the cable between the door and frame of the incubator (fig. 3b).

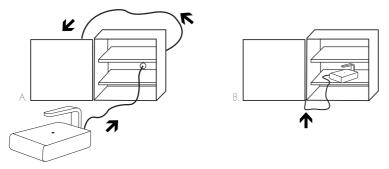


Fig. 3. When a port is present, guide the cable through it (a), otherwise guide it between the door and frame (b).

Important: Be careful when guiding the cable between the door and frame of the incubator. The door hinges can cause a cable breach, which is not covered by warranty.

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Securing the Cable

You can easily secure the USB cable on the outside of the incubator using the three cable clips. We recommend you attach the tablet on the incubator door. When doing so, use one cable clip to secure the USB cable connection near the tablet. For more details, please check the quick start guide of the tablet.

Status Indicator

To check if the device is active, open the incubator to see if the CytoSMART™ Lux2 is illuminated:

- Green light: the device is active and an image is visible on the live screen.
- No light: the device is in-active. The device is powered down, as it is in between imaging or not properly connected.

Vessel Placement

It does not matter which culture vessel you use. The CytoSMART™ Lux2 is compatible with a variety of culture vessels, including T-flasks, well plates, petri dishes and flat tubes. Please place the CytoSMART™ Lux2 in the incubator with the logo facing the operator. This way the image moves in the directions indicated by the arrows when the culture vessel is moved on the CytoSMART™ Lux2 (Fig. 4).

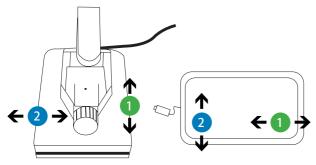


Fig. 4. Movement of the cell culture with the corresponding movement on the live screen.

Starting the Software

You do not have to manually activate the software. The CytoSMART™ software will start automatically when the tablet is activated. To start recording, the CytoSMART™ Lux2 needs to be properly connected via USB.



4. Setup an experiment

This is it, the real magic begins here. With the CytoSMART™ Lux2 properly placed inside the incubator and the software active, you can begin with the imaging of an experiment. Place a culture vessel on the Lux2 to begin. An image of the cells will be visible on the live screen (fig. 5).

Zoom Level

Before you can get your images, you need to select what these images should look like. Select the correct zoom level for the experiment. Just select **'Menu'** (1) on the bottom left corner and select **'Zoom in'** or **'Zoom out'** (2), for a 200x or 100x magnification respectively (Fig. 5).

Focus

The right focus is all you need to get the perfect images. Use the **'Slide bar'** (3) to adjust the focus (Fig. 5). The plus and minus buttons can be used for more precise adjustments.

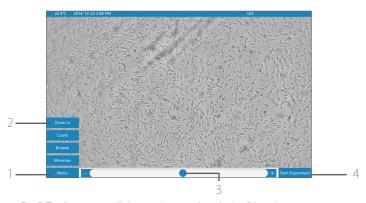


Fig. 5. The live screen will show an image, when the Lux2 is active.

Important: Registration of your device is required before you can start imaging! The next pages will help you with this.

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Registration of the Device

After you press **'Start experiment'** (4) for the first time, the device needs to be registered using the license key provided. By selecting **'Existing organization'** (5) from the menu (Fig. 6), the device can also be added to a pre-existing CytoSMARTTM account. If you are a new user, please select **'New registration'** (6).



Fig. 6. Registration menu.

To register the CytoSMART™ Lux2, enter a name (7) for the device, e.g. company name or research group. Other CytoSMART™ products can be added to this name. Next, create a personal account using an email address (8) and a personal password (9). An on-screen keyboard will appear, allowing you to enter your details. To finish, select **'Sign Up'** (10) from the menu (fig. 7).



Fig. 7. Account details menu.



Enter the provided license key (11) and select "Register" (12) to continue (fig. 8).

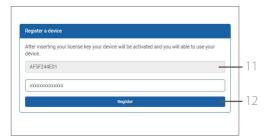


Fig. 8. Registration details menu.

Account Setup

It does not matter whether you want to include one or multiple experiments in one project. The CytoSMART™ Lux2 can do it all. An experiment is one imaging session of a cell culture experiment.

Select "Start Experiment" (4) in the live screen (fig. 5), to start a new experiment simply login. A menu will appear (Fig. 9) asking to provide login details (13). Use the details created previously, and select 'Log in' (14).

Do you want to create new account? Just select **'Create an account with'** (15) from the menu.

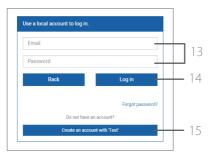


Fig. 9. Account log in menu.

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Start a Project

If you are the creator of a new project, you will be assigned as a project owner or an admin (16) and this cannot be changed (fig. 10). A project can hold multiple experiments. Optionally, you can give additional users access to a project by adding their email address (17). These additional users will receive an invitation email to join the project.

You need to provide a project name to continue (18).

Next, an optional project description and visual representation (Project Icon) can be added to a project (19). Here you can provide additional information regarding the project, for all users with project access.

After entering all details, select 'Create my project' to create a new project (20).

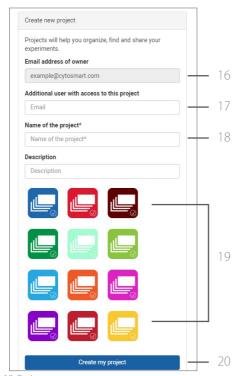


Fig. 10. Project menu.



Start an Experiment

After you have created a project, the new experiment screen (Fig. 11) will appear. By selecting the dropdown menu (21), you can assign the experiment to a different project. When you want to start a new project, select the **'Create a new project'** (22) option. This will return you to the project menu.

An experiment name is required to continue (23). Additionally, an experiment specific description can be added if you wish (24).

Next, set the desired imaging frequency by selecting **"Snapshot interval (in minutes)"** (25). A drop down menu will appear with different intervals to choose from.

Important: Since biological cells can be susceptible to light, it is advised not to use a higher recording frequency than needed.

Now it is time to push the big button. After selecting **'Start my experiment'** (26), the imaging will begin and a confirmation email with a link to the experiment will be sent to the project owner and optional additional users.

When the imaging has started, the system will go in sleep mode. For more details regarding the sleep mode, please check the quick start guide of the tablet.

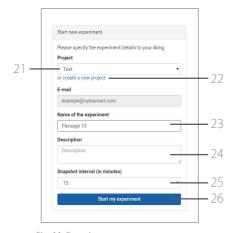


Fig. 11. Experiment menu.

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Ending an Experiment

Stopping is as easy as starting. To end an experiment, select the red **'Stop experiment'** button (27) from the live screen and select 'Yes' to confirm (fig. 12). A confirmation email indicating that the project has ended will be sent.

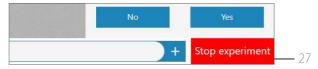


Fig. 12. Ending an experiment.

5. Connecting to the Cloud

One of the coolest features of this hardware is that the experiment results can be accessed through the CytoSMART™ Cloud. If you want to access the cloud, the following are required:

- PC, tablet or another device with a web browser.
- · Active internet connection.
- CytoSMART™ Cloud login details.

A two-year subscription to the CytoSMART™ Cloud is included with the CytoSMART™ Lux2. After starting an experiment, the captured images will be stored in the cloud. To allow for this, make sure the system has an active internet connection. All time logs in the cloud are set to GMT.

Confirmation Email

After starting an experiment, a confirmation email is sent that contains a link to the cloud (fig. 13).

Important: When you did not receive a confirmation email, please check the spam folder.



Fig. 13. Confirmation email.



Alternatively, go to https://cloud.cytosmart.com/ to log in and view your projects and experiments. This link will lead you to the CytoSMART $^{\text{\tiny IM}}$ Cloud login page (Fig. 14). Use your credentials created earlier to log in, see § 4.



Fig. 14. Login menu.

Project Overview

After logging in to the CytoSMART™ Cloud, you will get access to an overview of all projects (fig. 15).

Navigating the portal is pretty simple. If you have multiple projects in an account, use the **'Search Projects'** bar (1) to quickly search for a project. To create a new project, select **'New Project'** (2). This is similar to when beginning a new project, as described in § 4. It is not possible to start an experiment from the cloud.

You can get to your data easily. To access a project and all its assigned experiments, select the projects corresponding **'Project Icon'**, **'Project Title'** or **'Arrow Icon'** (3).

To delete a project, select the 'Waste Bin Icon' (4).

Select the **'Pencil Icon'** (5) to edit project details. This allows you to change the project name and its description.



Fig. 15. Project overview.

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Experiment Overview

After selecting a project (3), you will get access to an overview of all experiments assigned to that project (fig. 16). In the top, you can see an overview of the project data (6). As with the project overview, use the **'Search'** bar (7) to find a specific experiment within a project.

Select a specific experiment by clicking on its **'Experiment Icon'**, **'Experiment Title'** or the **'Arrow Icon'** (8). This will bring up the experiment page of that specific experiment.

To edit details of an experiment, select the **'Pencil Icon'** (9). This allows you to change the name and description of an experiment.

To delete an experiment entirely, select the 'Waste Bin Icon' (10).



Fig. 16. Experiment overview.



Experiment Results

After you select a specific experiment, the data collected during that experiment will be presented in the CytoSMART™ experiment page (fig. 17). In the next few pages, you will find an overview of the panes and buttons in this Experiment Page.

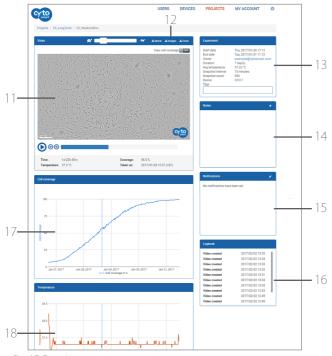


Fig. 17. Experiment page.

Overview:

- 11: Time-lapse movie of the cell culture.
- 12: Download buttons to download movies (.mp4), images (.jpeg) and cell coverage, and temperature values (.csv).
- 13: Experiment Summary Pane.
- 14: Notes Pane.
- 15: Notification Pane.
- 16: Logbook Pane.
- 17: Cell coverage graph.
- 18: Temperature graph.

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Fig. 18. Time-lapse movie pane.

Time-Lapse Movie Pane

This pane shows you a time-lapse movie of the cell culture (fig. 18). You can control the playback of the movie by the buttons below the image, these include play, pause, forward one picture, backward one picture. It also has a slider to control the movie (point in time). The **'View Tracking'** button allows to display the cell coverage in the time-lapse movie. Adjust the playback speed of time-lapse movie with the **'Turtle-Cheetah'** slider

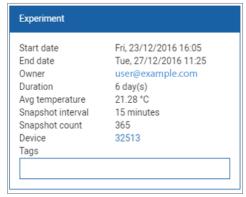


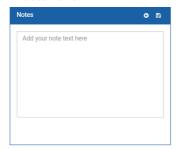
Fig. 19. Experiment summary pane.

Experiment Summary Pane

The summary pane (fig. 19) does exactly what you would expect: it provides an overview of the project i.e. project start date, duration, average temperature and number of images taken (snapshot count).



Notes Pane



You can add additional information (e.g. passage number, experiment conditions, etc.) regarding your project in the notes pane (fig. 20). This information will be stored in the database with your project, including a date and time-stamp.

Fig. 20. Notes pane.

Notification Pane

In the notification pane (fig. 21), you can set thresholds to enable email notification if:

- Cell coverage reaches a specified percentage.
- Temperature crosses a specified limit.



Fig. 21. Notification pane.

To set a notification, select the "+" button. The notification pane will unfold allowing you to specify the Type and Value of the desired notification. You can add multiple upper and lower limits. Furthermore, you can delete or add these notifications at any time throughout the duration of the experiment. When a limit is reached, the project owner receives an email notification.

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Logbook Pane

Note Removed	20-01-2017 12:49
ote Added	20-01-2017 12:49
lotification Added	20-01-2017 12:49
Notification Removed	20-01-2017 12:49
Notification Removed	18-11-2017 13:45
Notification Added	18-11-2017 13:45

The logbook pane shows a log of all the changes made to the experiment (fig. 22).

Fig. 22. Logbook pane.

Cell Coverage Graph

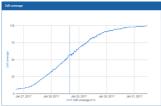


Fig. 23. Cell coverage graph.

In this graph, the cell coverage - in percentage of surface area - is plotted against the duration of the experiment (fig. 23).

Temperature Graph

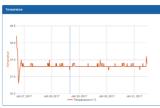


Fig. 24. Temperature graph.

In this graph, the temperature (°C) is plotted against the duration of the experiment (fig. 24).



6. Offline Experiment

Internet access is not required for you to experiment. With the CytoSMART™ Lux2 it is possible to perform an offline experiment when there is no internet access. However, we recommend to only use this for short-term experiments. Furthermore, access to the results will be limited to the system directly connected to the CytoSMART™ Lux2. Also, you cannot create videos, perform analysis or set notification thresholds of the images during an offline experiment.

Zoom and Focus

Before you start an offline experiment, adjust the zoom level and focus setting for the experiment. Please refer to § 4 for more details.

Start an Offline Experiment

When the CytoSMART™ Lux2 is active but without an active internet connection (1), an orange notification will be visible in the live screen (Fig. 25). With selecting **'Start Experiment'** (2), an offline experiment will begin.

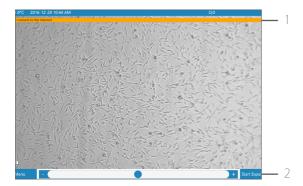


Fig. 25. Offline live screen, note the orange notification bar.

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After selecting **'Start Experiment'**, the offline experiment menu will appear (Fig. 26). Provide an experiment name here (2) to proceed.

Next, select the desired imaging frequency (3). A drop down menu will appear with different intervals to choose from.

Important: Since biological cells can be susceptible to light, it is advised not to use a higher recording frequency than needed.

When all details are set, select the **'Start'** button (4). Now the offline experiment will begin and the system will automatically go into sleep mode in between imaging.

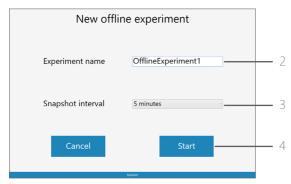


Fig. 26. Offline experiment menu.



Ending an Offline Experiment

Select the red '**Stop experiment**' button (5), to end an experiment and select 'yes' to confirm (Fig. 27).



Fig. 27. Ending an offline experiment.

Accessing Experiment Results

To access the obtained results (Fig. 28), select 'Menu' and then 'Browse' (6) from the live screen. To minimize the CytoSMARTTM software, select 'Menu' and then 'Minimize' (7).

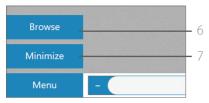


Fig. 28. Accessing results.

This will open a folder containing all the images (.jpg) of the offline experiments (Fig. 29). You can review the image files from the offline experiment or transfer them to a USB flash drive or other data storage device.

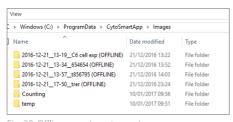


Fig. 29. Offline experiment overview.

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7. Cleaning and Troubleshooting

Cleaning and Maintenance

Lucky for you, the CytoSMART™ Lux2 requires minimal cleaning for reliable operation. Unplug the USB cable before cleaning the device. Use a damp, lint free wipe to clean the outer case with IPA or EtOH (70%).

Removing Dust Particles from Optical Window

If you see dust particles on the optical window, remove them by cleaning with a soft, lint free wipe. Do not use abrasive chemicals or rough materials to clean, as this may scratch or break the cover glass. A scratched or broken cover glass can be repaired but this is not included within the product's warranty.

Important: Do not use acetone to clean the CytoSMART™ Lux2. Also, do not autoclave the CytoSMART™ Lux2, this can result in damage to the electronics.

The CytoSMART™ Lux2 does not need regular maintenance. Do not perform any repairs or service on the device yourself, contact CytoSMART™ if a problem should arise, via support@cytosmart.com.

Troubleshooting

When a problem with the CytoSMART™ Lux2 occurs, an orange notification bar will appear with a message (fig. 30). One or multiple of the following messages can appear:

- Please connect the device
- Please connect the charger
- Please connect to the internet
- Server offline, online functionality unavailable
- Warning! Prolonged live mode can cause increased temperature

Please connect the device Please connect the charger

Fig. 30. An example of notification messages.



Please connect the device

When this error message appears, there is a problem with the CytoSMART™ Lux2 connection with the tablet or other Windows device. This is usually caused by the USB cable not being properly connected. To resolve this problem, make sure the Lux2 is connected properly via USB 3.0.

Please connect the charger

When the tablet is not connected to a functioning power outlet, connectivity problems may occur with the CytoSMART™ Lux2. To resolve this, make sure the tablet is connected to a functional power outlet using the provided charger.

Be sure that the power outlet is functioning (e.g. by plugging in another electrical device). If the message remains, switch off the tablet, unplug and re-connect all cables and reconnect the Lux2. Then start the tablet again.

When using a USB hub, be aware that some older versions – e.g. non-powered USB hubs – might not provide enough power for the Lux2 to operate properly.

Please connect to the internet

When this error message appears, the tablet has no active internet connection (Fig. 25). Without an internet connection, the system cannot connect with the CytoSMART™ Cloud. To resolve this, please try the following:

- 1. Make sure that there is an active internet connection.
- 2. Reboot the tablet.
- 3. Verify an active internet connection by opening a website.
- 4. If no website is loading, verify the internet connection on another device.

If your internet connection is working correctly but the message remains, contact your IT department for further assistance in accessing an internet connection. Otherwise please contact our support, via support@cytosmart.com.

Note, you can still perform an offline experiment in this scenario.



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Server offline, online functionality unavailable

When this error message appears, the CytoSMART™ Lux2 cannot upload to the CytoSMART™ Cloud. You can still perform an offline experiment.

If this error remains, please contact our support, via support@cytosmart.com.

Warning! Prolonged live mode can cause increased temperature

When the CytoSMART™ Lux2 has been active for 3 minutes continuously, due to activity in the live screen, this warning will appear.

It is not recommended to continuously monitor the culture vessel from the live screen, as this may affect cells due to temperature change or prolonged exposure to light.

No Project Email Received

A confirmation email should arrive a few minutes after starting an experiment. If no email was received, please check to see if the SPAM filter blocked the email.

Important: : To send out a confirmation email, the CytoSMART™ Lux2 requires a working internet connection

Video Is Shaking

If the images in your time-lapse imaging appear to be shaking, it is likely that your incubator is shaking due to a problem with the bearings or a nearby shaker (e.g. inside another incubator). To check this, please place the CytoSMART™ Lux2 outside of the incubator and start a project to see if the shaking remains. If the problem persists, please contact our support, via support@cytosmart.com.



8. Technical Specification

AC 100-240V, 2A, 10W, 50/60Hz
Max 2.4 x 1.5 mm
100X & 200X
1280 x 720 pixels
JPEG, CSV, AVI
LED, wavelength 550 nm
5 MP CMOS
10.1-inch tablet
No optical filters
133 x 90 x 100 mm (LxWxH) or
5,2" x 3,5" x 3.9" (LxWxH)
0.5 kg or 1.1 lbs.
Unlimited storage in the CytoSMART™
Cloud with the subscription.
5-40 °C, 20-95% humidity
Windows 7 or higher
1.44 GHz CPU
2 GB of RAM
32 GB of free drive space
DirectX 11 or higher capable graphics
card or integrated chip
USB 3.0 port
Internet connection
Windows 7 or higher
2.4 GHz i5 CPU
4 GB of Ram
128 GB of free drive space
DirectX 12 capable graphics card or
integrated chip
USB 3.0 port
Internet connection

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9. Warranty

The CytoSMARTTM Lux2 is covered by warranty - if the device is properly used and installed - for a period of one year (1 year) from the date of purchase. If any defects occur during this warranty period, CytoSMARTTM will repair or replace the defective parts at its discretion without charge.

The following defects, however, are specifically excluded:

- 1. Defects caused by improper operation.
 - 2. Damage caused by accident or misuse.
 - 3. Damage caused by autoclavation of the device.
 - 4. Damage cause by the sterilization cycle of an incubator.
 - Repair or modification done by anyone other than CytoSMART™ or an authorized agent.
 - 6. Damage caused by substituting alternative parts.
- 7. Use of fittings or spare parts supplied by anyone other than CytoSMART TM .
- 8. Damage caused by disaster.
- 9. Corrosion caused by improper solvent, sample or treatment of the device.

CytoSMARTTM cannot be held responsible for damage incurred during shipment of a defective instrument. It is recommended that you save the original packing material in which the instrument was shipped. This warranty is limited to the replacement of defective products.

For any inquiry or request for repair service, please visit us at www.cytosmart.com/support or contact our support staff via support@cytosmart.com.



10. Ordering Information

Would you like to purchase extended warranty for the CytoSMART™ Lux2? These products are available separately from CytoSMART™ for the duration of 1 year, 2 years or 5 years.

Cat. No.	Description	Contents
HCA-1003	CytoSMART™ Care - Lux2, option 1	Extended warranty for 1 year
HCB-1003	CytoSMART™ Care - Lux2, option 2	Extended warranty for 2 year
HCC-1003	CytoSMART™ Care - Lux2, option 3	Extended warranty for 5 year

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Support

Visit <u>www.cytosmart.com</u> for more information, support or sign-up for our newsletter. If you have any question regarding a CytoSMART™ product, please contact our support staff via <u>support@cytosmart.com</u> or call us at +31 (0)88 20 32 200.

If you require technical assistance, our support staff is available to answer your phone-call, email or live-chat during weekly office hours (Central European Time, UTC/GMT +1 hr.)

Disclaimer:

For research use only. Not for use in diagnostic procedures.

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