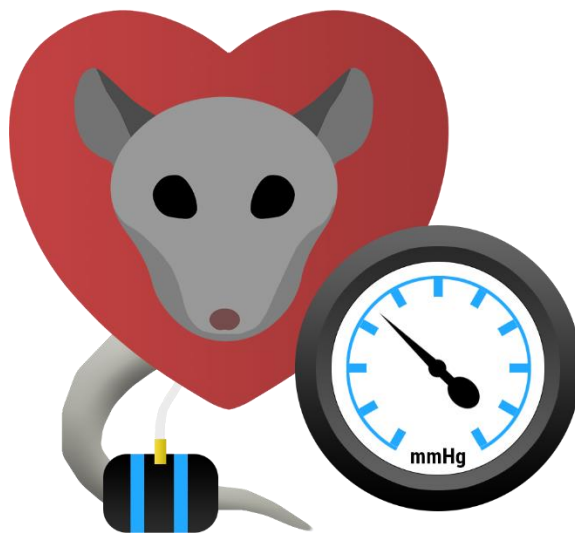


# Systole Phlogiston



Non-invasive blood pressure  
measurement system for rodents

v2.0

Moscow, 2023

# Before getting started

This document is an operation and maintenance manual for the “Systole” non-invasive blood pressure measurement system and the “Phlogiston” heating platform.

We recommend that you carefully read and understand this manual before using the product. This manual contains detailed information and instructions necessary for the proper and safe operation of the product.

Due to continuous product improvement, specifications are subject to change without notice.

Please inform the manufacturer of any errors or malfunctions you have encountered while using our products.

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# 1. Systole

## 1.1. Intended use

The “Systole” is a non-invasive blood pressure measurement system intended for use with rats or mice. The system uses a built-in electric air pump to pressurize the tail cuff until blood flow pulsations stop. While slowly reducing the pressure the system subsequently performs an automatic measurement of systolic pressure and calculates diastolic pressure based on the readings of an infrared pulse sensor worn on the tail of the animal following the cuff.

## 1.2. Specifications

Max pressure in the cuff	up to 300 mmHg (programmable)
Pressure measurement error	up to 3 mmHg (1% of range)
Types of cuffs	<ul style="list-style-type: none"><li>• Ø5-10 mm tail (rats)</li><li>• Ø3-6 mm tail (mice)</li></ul>
Pulse sensor	infrared
Pulse rate	40-600 bpm
Measuring cycle time	30-50 s (depends on the pressure measuring range)
Emergency stop	Software and hardware
Data logging frequency	1000 Hz
Supported OS	<ul style="list-style-type: none"><li>• Windows 10+</li><li>• Ubuntu 22+</li></ul>
Connection interface	USB 2.0
Power supply	USB (+5V, 400 mA)
Dimensions	190 x 100 x 40 mm
Weight	350 g
Warranty period	1 year

## 1.3. Device composition

### Main unit

The front panel of the main unit contains the following elements (listed from left to right):  $\varnothing 3$  mm quick coupling for pressure cuff tube, 3.5 mm connector for infrared pulse sensor, LED indicator, control button



The rear part of the main unit contains the following elements (listed left to right): USB cable connector,  $\varnothing 4$  mm quick coupling for device calibration and testing.

### Pressure cuff

The pressure cuff is designed to compress the blood vessels in the animal's tail.

During pressure measurement the air is rapidly inflated and slowly deflated, allowing the systolic and diastolic pressures to be determined by means of the IR pulse sensor. The pressure cuff consists of a main part, two side inserts, a latex diaphragm and a  $\varnothing 3$  mm transparent tube (1 meter long) for connection to the main unit.



## Infrared pulse sensor

The infrared pulse sensor (IR-sensor) is designed to measure the amplitude of the photoplethysmogram (PPG) when measuring the blood pressure.

It consists of a housing with a changeable silicone insert to fit the required diameter of the rat's (mouse's) tail and a cable.

The IR pulse sensor is plugged into the connector on the front panel of the device. The pressure measurement function is not available unless the sensor is connected.



## 1.4. Preparing the device for operation

**WARNING:** The device is equipped with quick-release couplings for a  $\varnothing 3$  mm transparent cuff tube at the front panel and a special  $\varnothing 4$  mm plug at the rear. The tube or the plug should be inserted by about 1 cm inside the coupling. Please note that you might have to apply a slight additional force to do that.

To disconnect (remove) the tube (or plug), press the ring on the end of the quick-release coupling and gently pull the tube (plug).

Do not apply excessive force when pulling out. In case the tube (plug) seems to be stuck check if the ring on the quick-release coupling is pressed all the way in.

1) Assemble the pressure cuff and the pulse sensor. To do this, insert the cuff into the fitting on the pulse sensor as shown in the figure.

2) Insert the 3.5 mm plug of the pulse sensor into the coupling on the front panel of the device.

3) Insert the 3 mm transparent pressure cuff tubing into the quick-release coupling on the front panel of the device.

4) Connect the USB cable to the device and the computer.



## 1.5. Preparation for blood pressure measurement

### 1) Place the animal in a restraining container

This is only necessary for conscious animals. Place the animal in the restraining container and adjust the stopper on the front to restrict the animal's forward and backward movements. The container should prevent the animal from rotating around its axis. The animal's tail should protrude freely out of the slot on the rear latch of the container.

### 2) Remove external stimuli

Abrupt movements and loud noises should be limited as much as possible, as they cause animals to move during pressure measurement. Sometimes covering the top of the restraining container with a cloth helps to reduce exposure to external stimuli.

### 3) Position the sensor

The pressure cuff should be placed on the proximal end of the tail, with the pulse sensor right behind it. Sometimes the pressure cuff cannot completely compress the tail vessels. In such case move the pressure cuff with the pulse sensor a few millimeters along the tail.

### 4) Ensure that there are no mechanical vibrations

The IR sensor used to measure the pulse is a highly-sensitive device and might respond to any movement of the animal's tail relative to the sensor due to mechanical vibrations.

### 5) Preheat the animal

Warming of rats and mice is necessary to perform pressure measurements. Warming ensures stable (with the necessary volume) blood circulation in the tail. Normally the animals should be pre-warmed up to 28-32 °C for 10-15 minutes. The temperature should also be maintained during the whole pressure measurement process.

### 6) Animal acclimatization

Most animals require some preparation and careful handling to get used to the restraining container, long-term heating and subsequent pressure measurement to allow you to obtain reproducible results. Unlike mice, rats are easier to get used to.



Several full cycles of training may be required for the animal to become accustomed. Despite all the measures taken, some pressure measurements may be unreliable due to the animal's body and/or animal's tail movements.

## 1.6. Software

The latest version is available at <https://rat-house.ru/systole> and <https://neurobotics.ru/downloads>

## 1.7. Performing pressure measurements

Launch the *Systole* software and press the *Enable* button to display the pressure curves and PPG obtained from the device. The connection indicator will turn green if a device is connected to the PC.

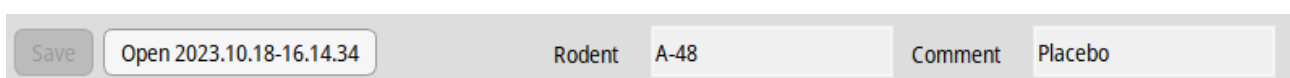
Press the *Measurement* button and the device will rapidly inflate the cuff to the pre-selected pressure. The cuff will compress the vessels in the animal's tail so that the pulse will not be observed on the PPG (in this case the only signal observed are the sensor's noise and tail movement artifacts).

Thereon the pressured air in the cuff will start to slowly drain through a special valve inside the device. Once the pressure in the cuff equals the systolic pressure pulsations become observed on the PPG. The amplitude of the pulsations increases as the pressure in the cuff decreases. The registration finishes automatically once the pressure reaches the minimum value set. Save the data to a file using the dialog box.

After the measurement is finished an analysis procedure automatically starts searching for the systolic and diastolic pressure values shown as vertical marks (SBP and DBP respectively). The key value of the SBP is determined as the first pulsations coordinate, whereas for the DBP the key value is determined as the coordinate where the pulse amplitude stops rapidly increasing as the pressure in the cuff decreases (i.e. the amplitude increases rapidly to the left of this point and increases slowly or slightly decreases to the right of the point).



The measurement can be saved to records (bottom left button) for further viewing at the *Records* tab. After the measurement is saved it can be open by clicking the *Open* button near the saving button.



For convenience every record has additional *Rodent* and *Comment* text fields. These values can be changed afterwards at the *Records* tab.

## 1.8. Manual edit of pressure marks

SBP and DBP marks can be edited manually to achieve more accurate systolic and diastolic pressures values.

Move the mouse pointer to the vertical marker, hold down the left mouse button and move the mark horizontally to the desired location:

- SBP – the onset of pulsations on the PPG during the pressure release in the cuff
- DBP – the endpoint of increase in the amplitude of PPG pulsations

Use the dark red dots displayed above the curve to facilitate the visual detection of PPG pulsation amplitude – these dots correspond to the moments of blood flow pulsations.

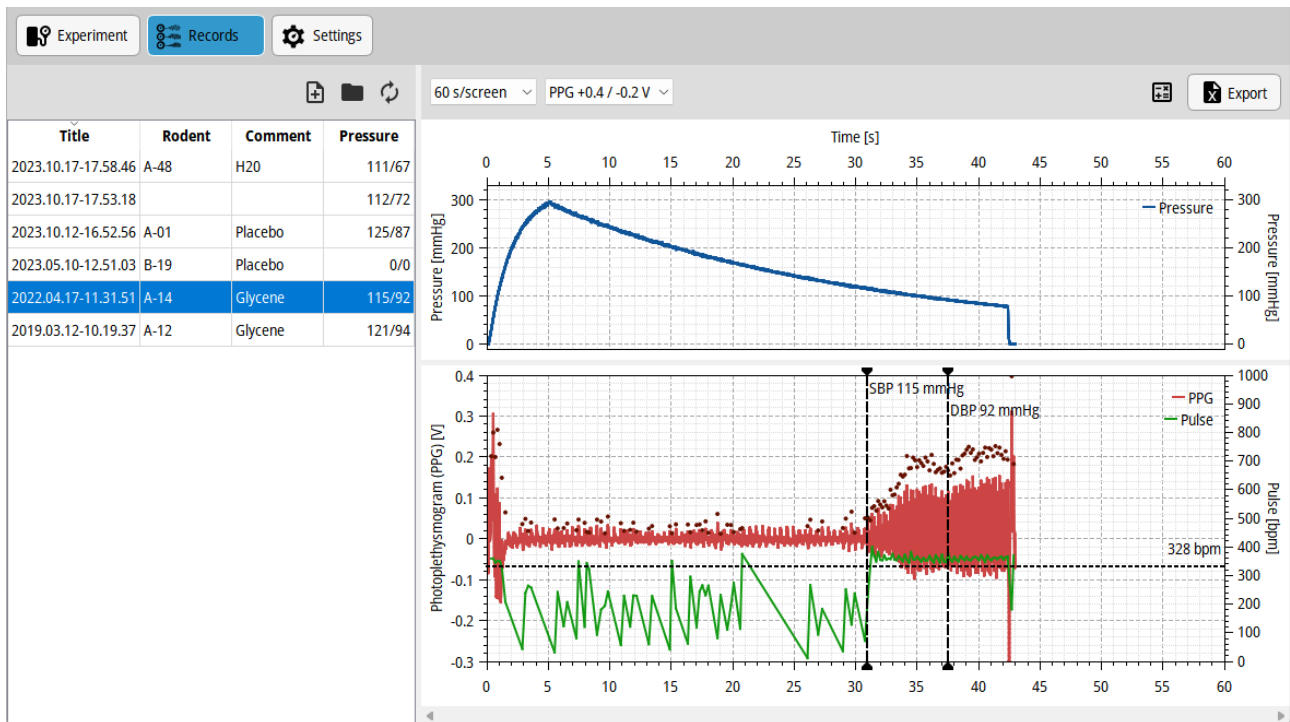
We recommend being guided by the lower boundary of the pulse curve to determine diastolic pressure. As compared to the upper boundary, it is better to see the moment of pulsation amplitude increase cessation.

## 1.9. Records database

All recorded experiments are placed in a single folder and are available at the *Records* tab.

### Single experiment view

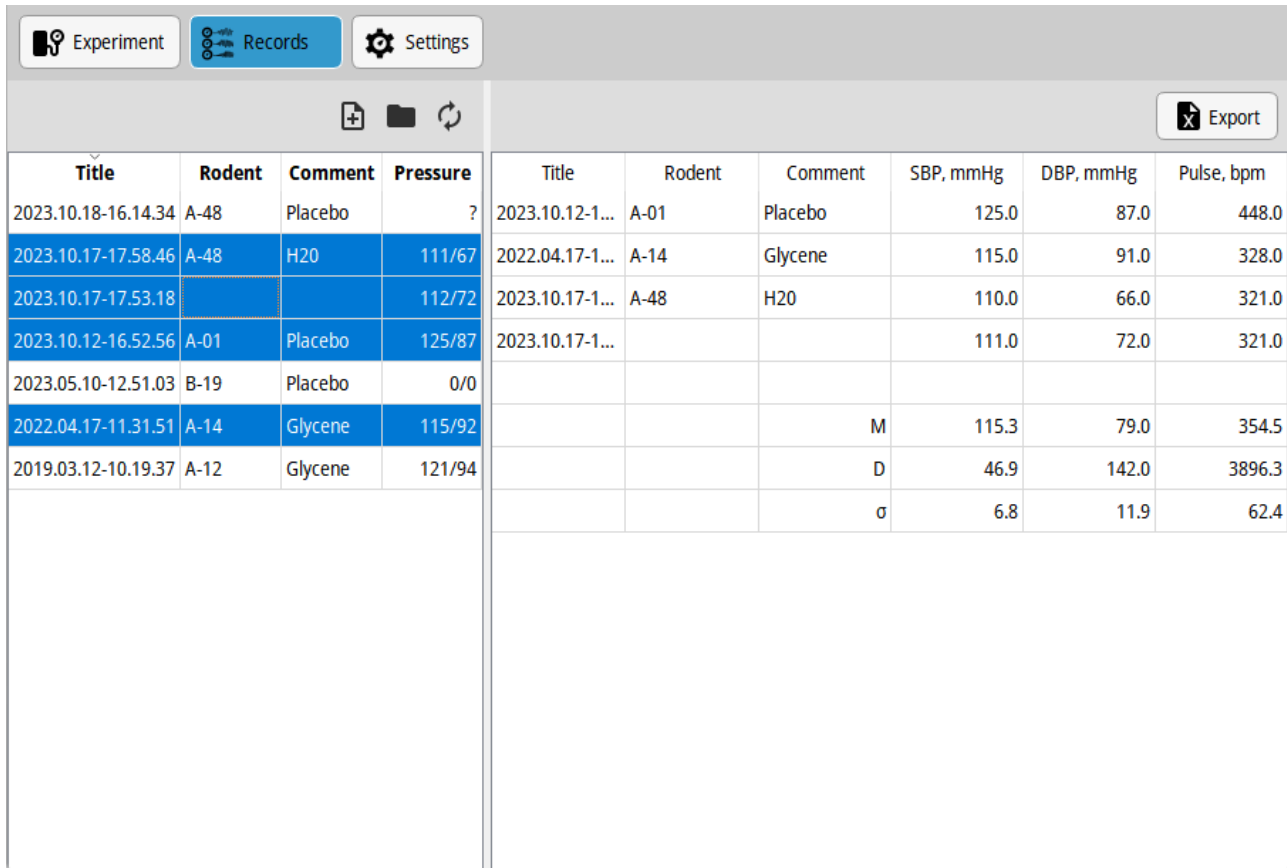
To view an experiment select a row in the records table. The experiment is viewed similarly to the measurement layout. SBP and DBP markers are available for edit.



The *Rodent* and *Comment* can be edited directly in the table cells.

## Summarized experiment table

Overall table of multiple experiments is calculated if several experiment rows are selected. The overall summary table shows SBP, DBP, pulse and respective statistical values: average (M), dispersion (D), standard deviation ( $\sigma$ ).



Title	Rodent	Comment	Pressure	Title	Rodent	Comment	SBP, mmHg	DBP, mmHg	Pulse, bpm
2023.10.18-16.14.34	A-48	Placebo	?	2023.10.12-1...	A-01	Placebo	125.0	87.0	448.0
2023.10.17-17.58.46	A-48	H2O	111/67	2022.04.17-1...	A-14	Glycene	115.0	91.0	328.0
2023.10.17-17.53.18			112/72	2023.10.17-1...	A-48	H2O	110.0	66.0	321.0
2023.10.12-16.52.56	A-01	Placebo	125/87	2023.10.17-1...			111.0	72.0	321.0
2023.05.10-12.51.03	B-19	Placebo	0/0						
2022.04.17-11.31.51	A-14	Glycene	115/92			M	115.3	79.0	354.5
2019.03.12-10.19.37	A-12	Glycene	121/94			D	46.9	142.0	3896.3
						$\sigma$	6.8	11.9	62.4

## Export to spreadsheet

To export experiments or summarized tables press the *Export* button (top right).

Available export formats:

- XSLX – a common spreadsheets format
- CSV – a simple text table format

Available data export types:

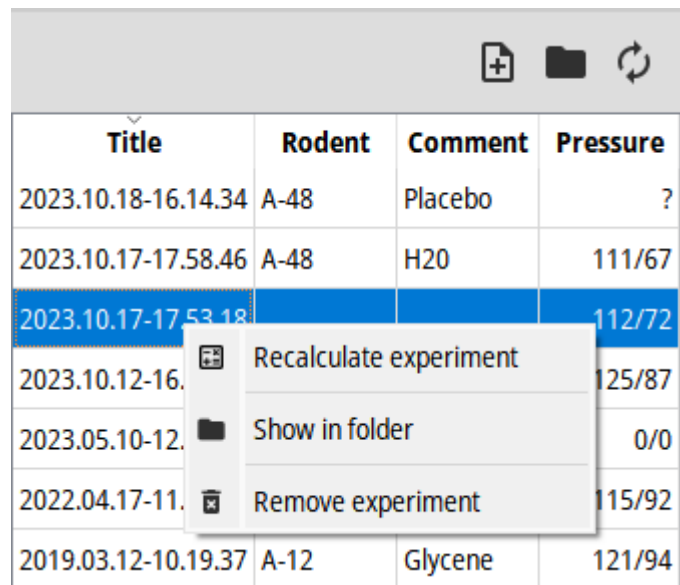
- Essential data – SBP, DBP, pulse, title, rodent, comment
- Full data – essential data plus all measured pressure and PPG signals

The exported summary table spreadsheet fill contains all single experiments in separate sheets.

## Import record

To import a record (.edf) press the button at the upper right corner of the records table.

Records from all versions of Systole software are available for import.



The screenshot shows a table with four columns: Title, Rodent, Comment, and Pressure. The table contains several rows of data. A context menu is open over the row with Title '2023.10.17-17.53.18', showing options: 'Recalculate experiment', 'Show in folder', and 'Remove experiment'. The table header has icons for adding, folder, and refresh.

Title	Rodent	Comment	Pressure
2023.10.18-16.14.34	A-48	Placebo	?
2023.10.17-17.58.46	A-48	H2O	111/67
2023.10.17-17.53.18			112/72
2023.10.12-16.			125/87
2023.05.10-12.			0/0
2022.04.17-11.			115/92
2019.03.12-10.19.37	A-12	Glycene	121/94

## Other operations with records

To view the actual folder, containing record files:

1. Press the folder button at the upper right corner of the records table  
*or*
2. Call the context menu on any record (right click) and choose *Show in folder*

To remove records, select one or more rows in the records table, call context menu (using right mouse click) and press *Remove*. Removed files are placed in the system's *Trash bin*.

## 1.10. Service procedures

### Check leakproofness of cuff and internal parts

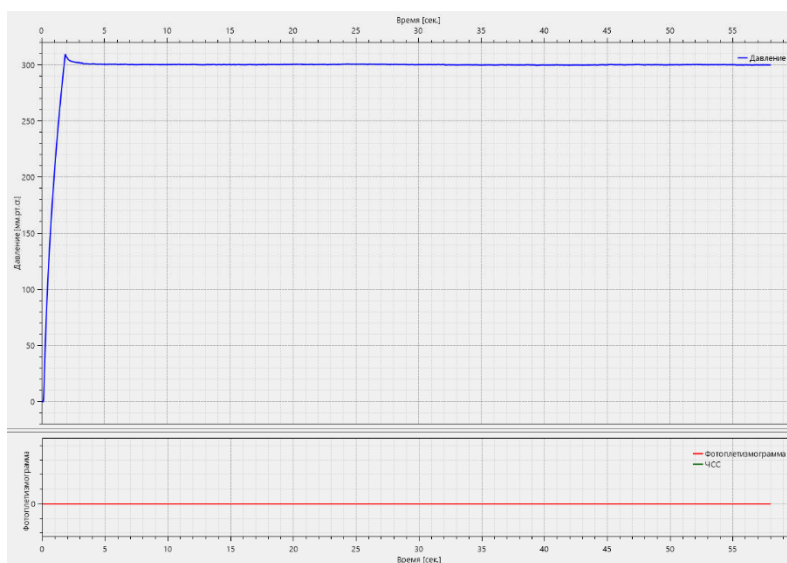
Note: Insert a special  $\varnothing 4$  mm plug (included in the accessory kit) 1-1.5 cm into the quick-release fitting located on the rear panel of the Systole.

Insert the  $\varnothing 3$  mm transparent tube of the pressure cuff into the quick-release coupling on the front of the device. Insert a thin and round object (such as a pen or pencil) inside the pressure cuff. Note that the object should not contain any sharp parts to avoid damaging the diaphragm.

Start the *Systole* software, switch on the recording by pressing the *Enable* button. Perform test inflation to 200 or 300 mmHg by pressing the *Measure* button. The pressure curve should not decrease in a common way (at a rate of 3-6 mmHg per second) and the pressure should decrease very slightly (due to small leaks inside the device). The typical value is less than 10 mmHg per 40-50 seconds, i.e. not more than 0.25 mmHg (0.033 kPa) per second.

Otherwise, make sure you have inserted the plug into the fitting on the rear side of the device and check if you have inserted the  $\varnothing 3$  mm transparent tube all the way into the device and into the cuff.

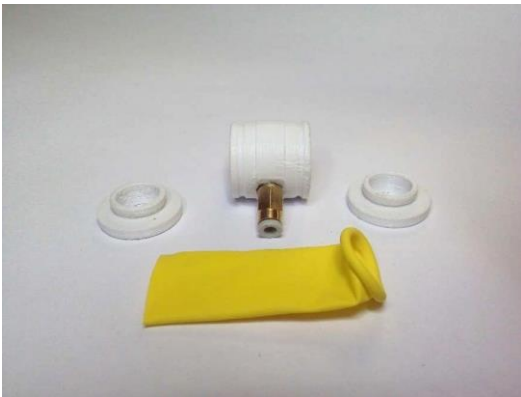
*Note: once finished, remove the  $\varnothing 4$  mm plug from the quick-release fitting at the back of Systole device.*



## Replacing cuff's latex diaphragm

A latex cuff membrane might need to be replaced in case it is leaking due to damage or normal wear and tear.

Pressure cuff consists of the main part, two side inserts, latex diaphragm and the 1 meter long Ø3 mm transparent tube for connection to the main unit.



1. Remove the two side inserts from the cuff. You may need to apply some force to do that. If necessary, use a thin object of some kind for removal.
2. Remove the latex membrane from the main part
3. Prepare a spare latex membrane for replacement
4. Insert the spare latex membrane inside the main part. Then, slide the edges of the diaphragm onto the main part while turning the membrane outward
5. Make sure that no folds were formed on the membrane inside the main part. The inner part should stay flat and loose enough – it should not be stretched out
6. Place the side inserts into the main part by clamping them to the latex membrane. The inserts have to be inserted tightly, so a certain amount of force must be applied
7. The side inserts must be pressed all the way into the main body
8. Check the tightness of the reassembled cuff

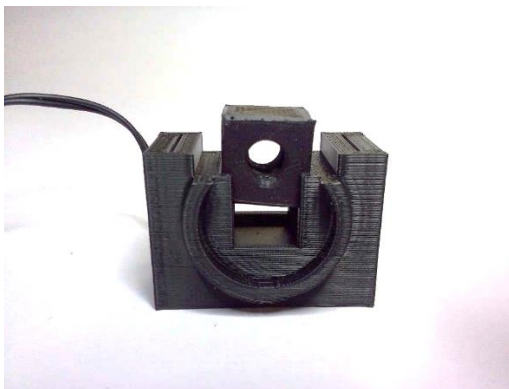


## Replacing PPG sensor's silicone insert

Replacement of the silicone insert of the PPG sensor may be required to adjust the sensor according to the different diameters of rodent's tail.

Rats:  $\varnothing 6$ ,  $\varnothing 8$ ,  $\varnothing 10$  mm

Mice:  $\varnothing 3$ ,  $\varnothing 4$ ,  $\varnothing 5$  mm



1. Disconnect the pressure cuff from the housing on the heart rate sensor
2. Remove the manufacturer's insert by pulling it up out of the sensor and gripping it along the sensor axis
3. Note: The silicone insert has holes for the infrared sensor on its side (besides the tail holes mentioned before). These holes are  $\varnothing 5$  mm for rats and  $\varnothing 3$  mm for mice respectively. Place the inserts with these holes to the side (the holes should be placed opposite to the IR receiver and transmitter) on the pulse sensor
4. The tail holes must be positioned along the sensor axis
5. The insert should be pressed between the positioning notches as shown on the picture to the left
6. Gently install the silicone insert of the desired hole size (the insert slides in with slight friction). It must be plugged all the way down so that the top edge of the insert becomes aligned with the top of the housing

## 2. Phlogiston

### 2.1. Intended use

The Phlogiston is intended to maintain a set temperature while heating small laboratory animals.

### 2.2. Specifications

Maximum heating temperature	+50°C
Temperature control accuracy	±0.5°C
Temperature sensor resolution	12-bit (0.0625°C)
Platform heating time	Up to +32°C – 5 minutes Up to +45°C – 15 minutes
Control source	Built-in digital sensor
External sensor of the animal's surface temperature	Yes (Ø5 mm, measurement rate 3 s)
Type of temperature control unit	Microprocessor
Display	12 x 2 symbols
Control buttons	3 (Up, Down, Enter)
Power input	Medical use certified external power supply unit (220V->12V; current leakage <50 µA, insulation resistance 100 MOhm at 500 V)
USB interface	Yes (galvanic isolation up to 2500 V, leakage current <10 µA)
Usage conditions	Indoors; +5°C to +40°C, humidity ≤80%
Heating platform dimensions (other dimensions are available on the request)	Size: 244 x 123 x 17 mm Material: aluminum top, plastic bottom
Weight	≤ 1 kg

*Note: The maximum temperature and heating rates are given for an ambient temperature of +22°C and the absence of strong air currents. Performance may be reduced at low ambient temperatures or in strong airflows.*

## 2.3. Device composition

- Control unit with built-in display and control buttons
- Heating platform with built-in temperature sensor
- External temperature sensor
- 12V power supply unit (for 220 V main power supply)

## 2.4. Getting started

1. Connect the heating platform to the control unit
2. Connect the external temperature sensor to the control unit
3. Connect the power supply unit to the control unit and the mains supply

*Attention: It is allowed to connect (or disconnect) an external temperature sensor and USB cable to the controller once the power supply is connected. Do not connect (or disconnect) the heating platform from the control unit once the power supply unit is connected.*

## 2.5. Heating modes

1. The external temperature sensor is connected – sensor readings are used to adjust the heating temperature to the local value setpoint.
2. The external temperature sensor is not connected – the heating platform maintains the pre-set temperature using a built-in sensor.  
The external sensor is an optional input device.

## 2.6. Operating instructions

### Turning on and off

To turn on, press and hold the "Enter" button for 1.5 seconds or longer. To turn off, press and hold the "Enter" button for 3 seconds or longer.

### Temperature indication

The control unit display shows the external sensor temperature (top left) and the current platform temperatures (bottom left), as well as the setpoint temperature (right-top or bottom, depending on the presence of the external sensor).

### Setting up the temperature

Change the maintained platform temperature by pressing *Up* and *Down* buttons located to the right of the display. The temperature step is 0.5°C.

The last set temperature is automatically applied after switching on of the device.

Heating stops once the set temperature is reached and reactivates as the animal or platform cools down.

## 2.7. Rodent heating procedure

1. Place the rodent inside a restrainer on the platform, the tail shouldn't contact the platform
2. Put the thermal sensor inside the restrainer through a hole near the rodent's body but not contacting it
3. Place the cuff and sensor on the tail
4. The rodent can be covered with a soft cloth to stabilize heating, minimize external stimuli and lower stress factors
5. Turn heating to 32°C
6. Press *Enable* in the software and observe PPG amplitude. The heating usually takes 10-15 minutes in accordance to room's temperature and rodent's size. PPG should become stable in amplitude and without skips

*Attention: to acquire correct experiment results rodents should be trained for the pressure pumping procedure to minimize movements and recording artifacts.*

### 3. Operating, safety and storage requirements

Do not use the product under conditions of high relative humidity (over 80%), in water or in the rain. Do not allow moisture to enter the interior of the product.

Forbidden to use the device for purposes other than those for which it is intended, in violation of the rules and conditions of use.

### 4. Operating environment

After storage in a cold place (less than +5°C) and after transportation at negative temperatures the product should be kept unpacked for several hours at  $\geq +10^{\circ}\text{C}$  to avoid moisture condensation inside the product.

Operate at +5°C to +40°C and relative humidity noy more than 80%.

Do not use in conditions of precipitation, condensation, salt spray and ozone, in direct sunlight, in explosive environments, in environments with conductive dust, corrosive gases and vapors, and other conditions that do not provide adequate protection against adverse effects.

### 5. Transportation

It is allowed to carry the product by any means of transport, except for non-heated compartments of aircraft and sea transport.

## 6. Storage

Store indoors in the transport package of the manufacturer in the warehouses of the supplier and the consumer, except for warehouses of railway stations, at +5°C to +40°C and relative humidity of no more than 80%.

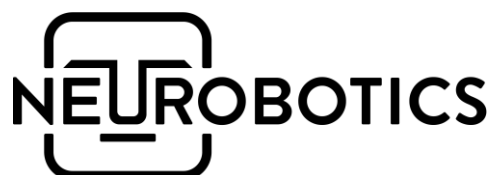
## 7. Recycling

Do not dispose with other household waste. The device should be disposed separately from other waste and properly recycled for reuse to avoid harm to the environment or human health from uncontrolled waste disposal.

For more detailed information on disposal of this product in an environmentally friendly manner contact either the retailer or manufacturer of the product.

# Abbreviations and terms

- DBP – diastolic blood pressure
- IR - infrared
- OS – operational system
- PC – personal computer (desktop computer)
- PPG – photoplethysmogram
- SBP – systolic blood pressure



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