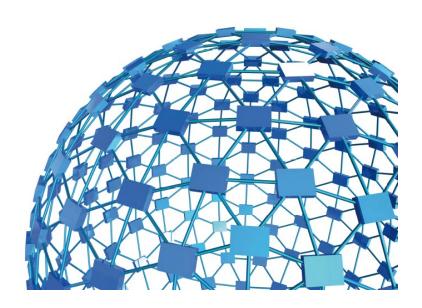


instruction manual

Hot/Cold Plate NG Cat. No. 35150



UGO BASILE S.R.L.

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instruction manual

Hot/Cold Plate NG Cat. No. 35150

Serial No.

SAFETY CONSIDERATIONS

ALTHOUGH THIS INSTRUMENT HAS BEEN DESIGNED WITH INTERNATIONAL SAFE-TY STANDARD, THIS MANUAL CONTAINS INFORMATION, CAUTIONS AND WARN-INGS WHICH MUST BE FOLLOWED TO ENSURE SAFE OPERATION AND TO RETAIN THE INSTRUMENT IN SAFE CONDITIONS.

SERVICE AND ADJUSTMENTS SHOULD BE CARRIED OUT BY QUALIFIED PERSONNEL, AUTHORIZED BY UGO BASILE ORGANIZATION.

ANY ADJUSTMENT, MAINTENANCE AND REPAIR OF THE OPENED INSTRUMENT UNDER VOLTAGE SHOULD BE AVOIDED AS MUCH AS POSSIBLE AND, WHEN INEVITABLE, SHOULD BE CARRIED OUT BY A SKILLED PERSON WHO IS AWARE OF THE HAZARD INVOI VED.

CAPACITORS INSIDE THE INSTRUMENT MAY STILL BE CHARGED EVEN IF THE IN-STRUMENT HAS BEEN DISCONNECTED FROM ITS SOURCE OF SUPPLY.





www.ugobasile.com

Hot / Cold Plate

Cat. No. 35150



General

This new **Hot/Cold Plate NG** offers a wide temperature range, presetable in the range -5°C to 65°C, can be used as:

- A conventional HOT PLATE, to carry out a rapid precise screening of narcotic type analgesic drugs according to the well known Hot Plate Test devised by N.B. Eddy and D. Leinbach.
- As a COLD PLATE; the Cold Plate Test is useful in studying cold receptors and cold allodynia, a phenomenon very frequently observed in chronic pain on humans.

The lid reduces humidity condensation on the plate at low temperatures.

Two working modes allow for testing at fixed temperature or at increasing/decreasing temperature (RAMP).

An optional **auxiliary Plate** (heat only) can be connected to the main unit and will be useful in the habituation phase.

Brand new, user friendly software, to set up the experiment and manage the results.



for Rats for Mice

- IT CAN BE USED AS HOT PLATE OR COLD PLATE
- NEXT GENERATION INSTRUMENT: SAME RELIABILITY, INNOVATIVE TECHNOLOGY!



Main Features

- OPERATING TEMPERATURE:
 -5.0°C to 65.0°C in steps of 0.5°C (0.1°C precision)
- DETECTION: by pedal switch
- OPERATING MODES: fixed or ramping temperature, for dynamic experiments
- X-PAD SOFTWARE: brand new, user friendly software included as standard, to set up the experiment and manage the results
- CONTROLS: 4"3 touch-screen to set and monitor the test
- DATA PORTABILITY: via the USB Memory-Key, included as standard



MOD.04 REV 0

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	35150-320	1	1	1		RESTRAI	NER LID	(for H/C	PLATE only)	
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Hot/Cold Plate NG

Cat. 35150

1 GENERAL

The hot plate test is a test of the pain response in animals, used in basic pain research and in testing the effectiveness of analgesics by observing the reaction to pain caused by heat.

The method, devised by N.B. Eddy and D. Leimbach in 1953 (see paragraph 8.1-Method Papers) evaluates the reaction time of mice dropped on a heated surface and thus confronted with a heat stimulus applied to the plantar surface.

Eddy and Leimbach used a behavioral model of nociception where behavior such as jumping and hind paw-licking are elicited following a noxious thermal stimulus. Licking is a rapid response to painful thermal stimuli that is a direct indicator of nociceptive threshold. Jumping represents a more elaborated response, with a latency, and encompasses an emotional component of escaping.

When a central analgesic agent is administered to the animals, reaction time is markedly increased.

The new Hot/Cold Plate 35100 has been designed to study more recently published cold receptor systems of neurons, which are not activated by cold but rather by chemicals, as well as the traditional cold receptors. These neurons are present in the CNS as well as PNS, but they are different from the neurons which sense pain.

1.1 Principle of Operation

While in the original model, hot plate temperature was obtained by thermoregulated water circulation, commercially available models were later designed, including heating elements, precisely regulated, to perform rapid and precise screening of analgesic drugs on the animal by the hot plate test.

The test is performed by placing an animal on a heated (or cooled) surface; a transparent glass cylinder is used to keep the animal on the heated surface of the plate. After a brief period, typically several seconds, an animal will lift and lick a paw as the heat of the surface becomes uncomfortable: the animal is then immediately removed from the apparatus.

The dependent variable in this test is the latency time, defined as the time period between the zero point, when the animal is placed on the plate surface, and the time when the animal shows stereotypical response to pain/strong discomfort, such as paw licking,



or jumps to avoid thermal pain; the operator stops the counter via the pedal switch, thus recording the current temperature and the reaction time.

2 INSTRUMENT DESCRIPTION

The Instrument features:

- 1) a main unit of original design, see paragraph 2.1, incorporating the plate proper and a touch screen command/display
- 2) a Plexiglas **restrainer**, see paragraph 2.2.

The 35150 Hot/Cold Plate NG combines the same functionality of the previous version, higher performances, greater stability and uniformity, more precise and quicker temperature control, and much easier experimental organization and data management.

Operating modes will allow to work with constant temperature or ramp (timed or exponential), defining the initial and final temperature to set an upward or a downward ramp.

The plate temperature is monitored by a precise temperature sensor embedded in the plate, in 0.1°C steps and reaction time is recorded in 0.1s increments.

2.1 Main Unit

The Plate is cut from a 10mm thick aluminium slab and is mated to high surface heating/cooling elements which provide even temperature to the entire test area.

A solid state temperature regulator senses the plate temperature via a digital thermometer and powers the heating/cooling elements through a proportional circuit to minimize temperature overshoot.

2.1.1 Important Notice on Temperature Range

The plate temperature can be set by the operator in the range -5 to 65°C (in steps of 0.5°C). The extremes of this ample range can be reached, provided the room temperature remains in the interval 18-25°C.

Temperature increase/decrease at different rates is slightly different in the different ramp sections:

Temperature ramp	Range	Max. Rate ¹
▲ Up	-5° to 65°C	+9°C/min
▼Down	65° to 30°C	-7°C/min
▼Down	30° to 5°C	-3°C/min
▼Down	5° to -5°C	-1°C/min

¹ The above measures have been taken at a room temperature of 21°C



2.1.2 Considerations Over Animal Welfare

In any analgesia test, great care must be taken to prevent the animals from inadvertent harm.

When an investigator initially begins using the hot plate test, and when first assessing a new strain of animal, care must be taken to identify the appropriate amount of stimulus (i.e., temperature of surface) that will produce the desired response (i.e., paw lick or jumping).

When operating at constant temperature, a cut-off time, generally 30 sec must be established to minimize risk of an animal sustaining tissue injury from prolonged exposure to the heated surface.

One should also assess the effects of a reference analgesic such as morphine to determine that the planned hot plate temperature will result in the desired sensitivity (J.N. Crawley "Current protocols in Neuroscience" J Wiley, 2001)

The 35150 is very quick in temperature changes: when operating in "ramp" mode, please pay attention when operating at temperature extremes and with extremely quick temperature ramps, to avoid irreversible damages on the animals on test (in particular when testing mice).



2.1.3 Touch Screen Command/Display

The 35150 incorporates a 4"3 touch-screen, for basic setting and monitoring, via an intuitive panel.

The picture shows the main menu.

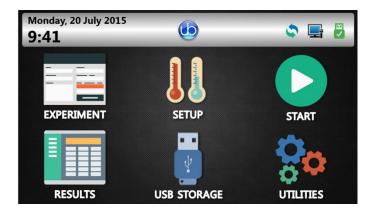


Figure 1, "Main Menu"

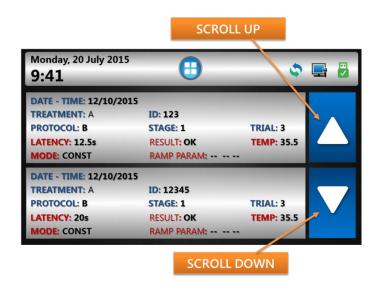
During the test, the touch-screen indicates the actual plate temperature, the operating mode, and the latency time.

this field indicates the environment temperature.





The test results, can be browsed in a condensed view.



The brand new **X-PAD** software, included as standard, provides a user-friendly interface to set the experiment and a powerful tool to manage the results easily on one's PC. Data exchange from the instrument to the PC is managed via the USB flash drive provided.

See also paragraphs 2.5-Experimental Configuration and 2.6-Data Collection and Management

2.1.4 Notes on Resistive Touch-Pads

If you have in mind the sensitivity to touch of your tablet or smart-phone, you might think the Hot/Cold Plate touch-screen does not react promptly, and you have to press down harder, or use a pen.

This is because we employ a resistive touch-screen (while most smart phones use a capacitive screen); the resistive screens are better of our application because of

- High resistance to dust and water
- Better use with gloved hand or stylus (because the conductive technology is dependent on the conductive nature of human body, it doesn't work if the user is wearing gloves).

2.2 Perspex Restrainer

The animals are confined by a clear removable cylinder made of clear Plexiglas, which dimensions have been found to be suitable for both mice or rats.

Cylinders of different heights are available on request.



A **cylinder lid** is provided with the standard package, which purpose is to reduce frost condensation at low temperatures.

To guarantee the rated performances, the lid should always be used under 10°C.



2.3 Additional Plate

An "auxiliary" Hot Plate, Cat. No. 35150-002, can be supplied as an option, useful in the training and acclimation phase, for high-throughput tests.

The 35150-002 is a basic, heating only, additional hot plate and does not record the temperature and reaction time.

As an independent unit, it can be used for basic tests as well, using a stopwatch.

Temperature range is from 20°C (or room temperature, if >20°C) to 65°C.

The 35160 combo-package, which includes a 35150 Hot/Cold Plate and a 35150-002 Hot Plate, can be taken into consideration.

See paragraph 7.2-Optional.

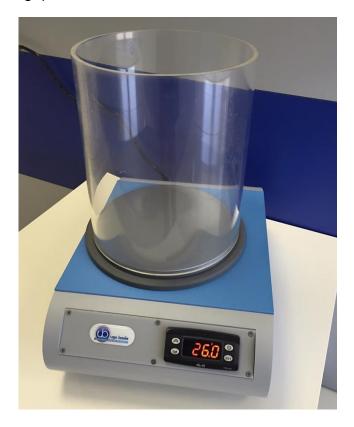


Figure 2, "Additional Hot Plate 35150-002"

2.4 Hot/Cold Plate Features

The main features of the new model are:

- **TEMPERATURE**: adjustable in the range -5.0 to 65.0°C, in steps of 0.55C (see note ²)
- TEMPERATURE ACCURACY: 0.1°C
- MODE: hot plate, cold plate, timed ramp, exponential ramp (see 4.3)
- DETECTION: by pedal switch
- LATENCY TIME: up to 999 seconds, in steps of 0.1
- CONTROLS: 4"3 touch-screen to set and monitor the test
- SOFTWARE: X-PAD brand new, user-friendly version, to set the experiment and manage the results
- DATA PORTABILITY: via the USB flash-drive provided.

² As the temperature is set in 0.5°C, a precision which requires continuous compensation to be maintained, you may notice the decimal figure in the temperature reading changing up and down. This does not mean that the plate temperature is not precise, but rather the contrary. The plate immediately integrates small temperature losses due to room temperature, animal temperature, etc.



2.5 **Experimental Configuration**

Via the new **X-PAD** software, the operator can easily organize the experiment on her/his PC, and upload it to the Hot/Cold Plate via the USB flash drive provided.

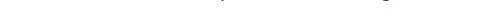
Treatments, protocols, stages, animals, and various test features can be quickly defined and saved for future use.



Configurations are easily:

- exported to Text, Excel or Pdf reports
- saved to cloud to DropBox, OneDrive, GoogleDrive

Data Collection and Management



A basic version of the collected data can be viewed on the touch-screen; when transferred to PC via USB drive, test results appear in full version.

The X-PAD software automatically classifies the data, combining configuration settings with test results. The user can add information, before or after the test.

Results appear in a tree-like structure, where columns can be dragged and dropped to customize the layout.

Data are quickly:

2.6

- exported to Text, Excel or Pdf reports



INSTALLATION 3

3.1 **Unpacking & Preliminary Check**

Check the contents of the shipment for completeness, packing list to hand, and visually inspect the instrument as soon you take it out of the packaging. Use the supplied *Check* List.

If the instrument is damaged, inform the carrier immediately, notifying our company. If after having tested it, the instrument fails to meet rated performances, please contact our after sales service, see paragraph 5.4-Customer Support.





Protect the environment!

Dispose of packaging properly, according to existing and applicable waste management rules and regulations.

3.2 Notes on the Instruction Manual

The 35150 Instruction Manual included in the package (on the USB pen drive) is necessary for the correct installation and operation of the instrument.

We recommend reading the manual with attention, as it is essential for the correct installation and operation of the instrument.

Please save the manual, ready to be consulted by the qualified personnel who use the instrument. Print it, only if necessary.

Our Instruction Manuals are available as free download on our web. For any additional information and/or assistance, you are welcome to contact our Service Department (see paragraph 5.4-Customer Support), specifying the serial number of your instrument.

3.3 General Safety Instructions

The following guidelines must be followed to ensure safe operation.

- ! DO NOT attempt to open or perform any service work
- ! DO NOT connect up human subjects



3.4 Assembling the Instrument

Put the instrument on a stable and flat bench or table surface. Position onto the base plate the animal restrainer supplied with the instrument. Once the Hot/Cold Plate is assembled, do not lift or move the combined unit. In particular, do not incline the base, as the Perspex restrainer may accidentally fall off the plate.

Please <u>do not block the grids</u> which protect the cooling fans on the cabinet side.

To guarantee the rated performances, a free area should be left around the instrument, as indicated in the drawing.

Obstructions which prevent the cooling fans to operate efficiently may degrade the instrument and alter its performance.





3.5 Before Applying Power

Consider the Power Module (see figure 3), positioned left on the back panel, which encompasses – from left to right - the mains switch, the inlet connection of the mains cord, and the fuse holder/voltage selector.



Figure 3, "Power Module"

3.5.1 Mains Switch

This two-pole toggle switch, which complies with international safety standards, is lighted when the instrument is **ON**.

3.5.2 Fuse Holder

The fuse holder comprises two fuses, one on the live, and the other on the neutral. Use (T5A) timed fuses for operation at both 115 or 230 Volts. For fuse replacement, please refer to paragraph 5.1-Electrical.

3.5.3 Mains Cord

It is a standard cable, Cat. # E-WP008. Make sure your power outtake is provided with a reliable ground connection.

3.6 Intended Use

The Hot/Cold Plate is intended for investigation use on **laboratory animals only**.

3.7 Additional Safety Consideration

- Use original accessories and spare parts only, see also paragraph 7-ORDERING INFORMATION.
- 2) immediately disconnect and replace damaged mains cord.
- 3) do not obstruct a comfortable access to the power module.
- 4) do not obstruct the grids of the cooling fans on the sides of the cabinet, see paragraph 3.4
- 5) do not operate in hazardous environments or outside prescribed environmental limitations (i.e. +10c° / +40c°, 95% max. relative humidity, non-condensing)
- 6) do not spray any liquid on the connectors and on the geared motor

UGO BASILE DOES NOT ACCEPT ANY RESPONSIBILITY FOR PROBLEMS OR HARM CAUSED TO THINGS OR PERSONS, ARISING FROM:

incorrect electrical supply;



- incorrect installation procedure;
- incorrect or improper use or, in any case, not in accordance with the purpose for which the instrument has been designed and the warnings stated in the instruction manual supplied with the instrument;
- replacement of original components, accessories or parts with others not approved by the manufacturer;
- servicing carried out by unauthorized personnel.

See also paragraphs 2.1.1 and 2.1.2

3.8 Connections

Connect the mains cord to a power outtake, **provided with a reliable earth connection**.

Now consider the connection module which is positioned on the front panel, for easy and quick access.

It encompasses the following connectors, from left to right:

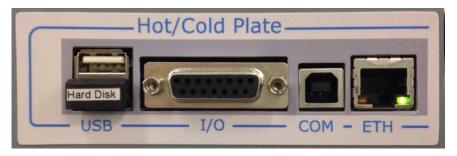


Figure 4, "Connection Module"

- the upper USB enables data exchange (configurations & results) with the PC, and allows firmware upgrades, via the USB flash drive provided, or to connect a keyboard (see paragraph 3.8.2)
- the lower USB accommodates the USB storage key labelled "HARD DISK".
 WARNING: DO NOT REMOVE IT! See paragraph 3.8.1
- I/O this D-SUB 1
- 5 connector provides TTL outputs for latency and temperature, see paragraph
 4.8.1-Pin-Out
- COM this USB-B 2.0 allows communication to the PC (for Ugo Basile use, programming and debug)
- ETH the Ethernet connector will be used for remote diagnosis and Internet access used for standard operation.



3.8.1 USB Hard Disk



The USB storage key must **be ALWAYS connected to the bottom port** (see picture). This USB key works as the inner memory of the system. It contains the collected data and the protocols.

If the instrument does not detect the USB HARD DISK, a message box asks to disconnect and to reconnect the USB Key and restart the device.



If during a normal operation, the user disconnects the USB HARD DISK, the device automatically switch off and restarts.

<u>WARNING</u>: this operation could damage the experiment data and Impair the internal memory.

3.8.2 Keyboard

When setting up the experiment, it is often requested (although not compulsory) to enter data via the touch-screen. For example information about the test, stage, animal, treatment, etc.

To ease the data input, it is possible to connect a <u>standard QWERTY keyboard</u> to the upper USB port. The keyboard will be automatically recognized and <u>alphanumeric</u> data typed on the keyboard will appear on the touch-screen.

Beside all the alphanumeric keys, the PC will accept:

- The backspace, to cancel the entered data
- The ESC key, to leave the menu
- The ENTER key, to confirm the input and move to the following field

3.9 Installation of X-PAD Software

The **X-PAD** software is saved on the USB Flash Drive provided with the instrument: download it on the Hard Disk of your PC.

4 OPERATION

Switch on the Hot/Cold Plate: the instrument will perform a check of the CPU, which might take up to 60 seconds, during which the display will show:



After switching on the instrument, first of all, get familiar with its controls.



4.1 Home Page

The test is managed by the 4"3 touch-screen.

This is how the Home Page looks like.

From the Home Page, the logo button on the tool-bar, opens the "screen-saver" window





This window displays current date and time, and the version of the software on-board.

From any other menu, the logo button always brings back to the Home page.

4.1.1 System Icons



NEW SOFTWARE DETECTED: if this icon is present, a new version of SW has been detected on the USB Key.



ETHERNET DETECTED: an Ethernet connection has been detected



USB KEY DETECTED: if this icon is present, a USB has been detected on the USB port.

4.1.2 Home Page Icons



In the EXPERIMENT page, it is possible to enter information about the test, including treatment, protocol, stage, trial, see paragraph 4.2.



Temperature and Mode are defined in the SET-UP page, see paragraph 4.3.



goes to START Page, see paragraph 4.4





goes to RESULTS Page, see paragraph 4.4.1



goes to USB STORAGE Page, see paragraph 4.6



goes to UTILITIES Page, see paragraph 4.6.1

4.2 Experiment

In the EXPERIMENT page, it is possible to enter information about the test.

Depress the keybord icon activate the virtual keyboard.







Enter a maximum of 12 characters in each field. TREATMENT, PROTOCOL, STAGE and TRIAL are text boxes, where it possible to specify the related information. Repeat the same process for each lane.

In the RESULT table the entered information will be matched with the test results. Of course, the user may choose not to enter any information and leave the fields empty.



The Home Page icon brings back to the home page.





NOTE:

Whenever a virtual keyboard is involved to enter data, it is possible to use an external QWERTY keyboard, connected to the Hot/Cold Plate's upper USB port, see also paragraph 3.8.2.

4.3 Set-Up

In this page it is possible to set-up **Mode** and **Temperature**.

The user may choose among the following modes, by depressing the related button at the page bottom.



CONSTANT: when this mode is selected, the plate maintains the preset temperature

RAMP: the RAMP mode is a timed mode: the user sets the time it take to reach the final temperature

RAMP EXP: with this mode, the plate reaches the preset final temperature starting from the initial one, as quickly as possible, also depending on the room temperature.

Having selected the mode and temperature (see paragraphs 4.3.1 and 4.3.2), press to go back to the Home page.



4.3.1 Constant Temperature

With **Constant Temperature** mode, the user may select the desired temperature, from -5.0°C to 65.5°C, with 0.1°C increments (or decrements).



Depress the icon to activate the numeric keyboard, and enter a temperature value between -5 and 65°C. Confirm by "OK".



Go back to the home page , then select the button to run the test. See paragraph 4.4. The plate maintains the same temperature for the duration of the test.

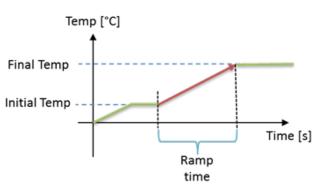


Before proceeding with the test, please read paragraph **4.4.1-Important Notes on Multiple Trial Experiments a**ttentively!

4.3.2 Increasing/Decreasing Temperature Ramp

This mode allows testing the subjects with an **increasing/decreasing** temperature. It is a timed mode, hence it is necessary to set the initial and final temperature, and the ramp (the <u>time the plate takes to go from the initial to the final temperature</u>).





Depress the icon to activate the numeric keyboard to enter:

- Initial temperature: between -5.0 and 65.0°C
- final temperature: between -5.0 and 65.0°C
- if the initial temperature is lower than the final temperature, the ramp will be increasing: on the contrary a decreasing temperature is obtained by setting an initial temperature higher than the final one)
- ramp: from 1s to 999s. Please note that the maximum temperature increasing/decreasing rate is dictated by physical limits. See also paragraph 2.1.1)

After setting temperatures and ramp, press to go back to the home page, then select to run the test, see paragraph 4.4.

4.3.3 Exponential Temperature Ramp

With the RAMP EXP mode, the plate reaches the temperature setpoint, starting from an initial one, **increasing/decreasing** the temperature as quickly as possible.





Depress the icon to activate the numeric keyboard to enter:

- Initial temperature: between -5.0 and 65.0°C
- final temperature: between -5.0 and 65.0°C
- if the initial temperature is lower than the final temperature, the ramp will be increasing: on the contrary a decreasing temperature is obtained by setting an initial temperature higher than the final one)

After setting temperatures, press to go back to the home page, then select to run the test, see paragraph 4.4.



ATTENTION:

In the case the room temperature exceeds the suggested values, the following warning message will appear:



It is still possible to proceed with test (pressing OK), but be aware that the plate performances in reaching the preset temperature are not guaranteed.

It is advisable to change the room temperature or move the device to a room where the suggested tempertatures can be attained.

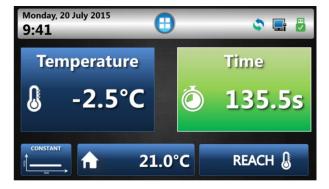
4.4 Starting the Test

Having set the temperature and the ramp (in temperature ramp modes), press





The plate rapidly reaches the preset temperature (initial temperature in ramp mode).



During the process, the temperature becomes yellow



and the right button indicates (read the note "ATTENTION" at the paragraph end).



When the preset temperature is reached, the figure becomes green



the right button becomes

START

The instrument is ready for the test proper: drop the animal on the plate and start the test and the latency timer, by depressing either the START button or the pedal switch.

When the animal shows stereotypical response to pain/strong discomfort, typically paw licking, or jumps to avoid thermal pain, the operator stops the counter by the **pedal switch**, thus recording the current temperature and the latency time.

The data are saved and will appear in the **RESULTS** table, see paragraph 4.4.1.

In the **ramp mode**, if the the final temperature is reached without an animal reaction, the plate maintains it and the latency timer keeps counting, until the pedal switch is depressed: in the result table the record will be marked as **FAIL**.



ATTENTION:

The STOP button should <u>never</u> be used to score the latency time.

This button interrupts the test: the plate goes back to the original temperature, and the datum is **not saved**.

4.4.1 Important Notes on Multiple Trial Experiments

This paragraphs suggests how to use the Hot/Cold Plate in optimum conditions, when working with constant temperature, in order to save time and guarantee high throughput.

Press the "START PAGE" button, then press "REACH"



The device will reach the set point temperature and stabilize around this value.



During this phase, the temperature will be colored in yellow:







If the stopped button is pressed, the temperature reaching procedure will be immediately stopped and temperature will go back to the initial value.

When the set point is reached, the Temperature will change colour becoming green and the value will be kept at the reached one.



To start the first trial of the experiment, press either the pedal switch (<u>highly recommended!</u>) or the start button.



After the first trial has begun, the temperature will be colored in white.





When a trial is over and the user wants to save the data, <u>pressing the pedal</u> is the only option available. In fact this is the only option which maintains the preset temperature.

Pressing the STOP button would - as explained earlier - return the temperature to the original value, thus inevitably requiring more time to increase/decrease again the temperature to the preset value.

Stopping the trial with the pedal switch, will maintain the set point value, waiting for the pedal switch to be pressed again in order to start the following trial: during this phase, the Temperature will always remain colored in white.

The described procedure may be repeated as many times as necessary.

In case the user has loaded a protocol previously created on the X-Pad, containing more than one animal, the software will automatically scroll to the next animal as soon as the pedal is pressed to end the ongoing test.

When all the trials belonging to a same experiment are completed, the user has to press the stop button in order to end the experiment and be able to exit from the Start Page. At this point, it is not necessary to hold the temperature at the preset value anymore.



4.5 Results

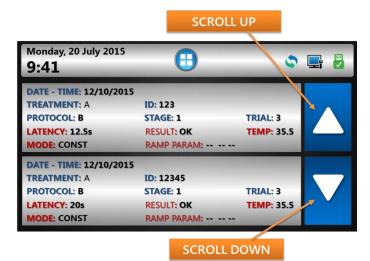
Data are saved in the internal memory and they can be scrolled on the touch-screen. Reach the results section by depressing the "RESULTS" icon from the home page.



The home page is reached from any menu, but depressing the

As you see from the example, all the Experiment-related information entered by the user appear under the blue headings

- DATE-TIME
- TREATMENT
- IF
- PROTOCOL
- STAGE
- TRIAL



The test results appear under the red headings:

- LATENCY
- TEMPERATURE
- MODE
- RESULT
- RAMP PARAM.

Scroll through the results by the UP \triangle and DOWN \bigvee arrows.

Results can then be saved on the USB flash drive, and uploaded on the PC for further processing, see paragraph 4.6.1. When transferred to PC via USB drive, test results appear in extended version.

4.6 USB Storage



By depresing the USB icon, you will reach the USB Storage menu.





4.6.1 Save Data



By pressing the "SAVE DATA" icon, results are uploaded on the USB-flash drive.

Insert in the upper USB port the USB drive provided with standard package. Correct insertion is confirmed by the \Box icon on the toolbar.

If you try to save the data without USB-KEY, the following dialog box will appear.



When the download procedure ends correctly, the following dialog box is displayed.

The file will be saved in .CSV format.



4.6.2 Load Protocol



When a USB flash drive, containing at least one protocol is connected to the device, the "LOAD PROTOCOL" button will open the following window, displaying all the loadable protocols found.

Use the arrows on the right of the screen to scroll the list up and down.



Having selected the desired protocol, if the protocol was properly loaded the confirmation box will show up, while an error box will appear if the file has been found corrupted.

After opening a protocol, the user will be able to test the animals following to the loaded protocol, combined with the information contained therein.

4.7 Utilities

The UTILITIES menu offers the following options:





4.7.1 Software Update



For software update you will receive the "HC_Update.hex" file by E-Mail or download it from our web site.

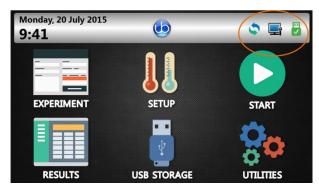
The file is to be saved on a USB key (FAT32 file system formatted), for example the standard Ugo Basile USB key provided with the Hot/Cold Plate.

Proceed as follows:

- A. Switch OFF the Hot/Cold Plate
- B. Copy the "HC_Update.hex" file on USB Key (file system FAT32);
- C. Switch ON the Hot/Cold Plate
- D. Insert the USB KEY containing "HC_Update.hex" file into the UPPER USB port (see picture on the right)



- If the USB key is detected, the
 icon on the system bar will appear on the tool-bar
- If the system recognizes a new SW version, the icon will appear on the toolbar.



Only if the two icons are there, it is possible to proceed with the update.

E. Select in sequence:





F. Confirm by answering OK to the question "Update Hot/Cold Plate application?





G. Wait about 30 seconds for the instrument to reboot.





IMPORTANT:

do not remove the USB Key during the updating procedure!!!

- H. Wait about 30 seconds
- I. Switch OFF the instrument
- J. Remove the USB key from the upper USB port (NB: do NOT remove the USB Hard disk from the lower USB port!)
- K. Switch ON the instrument
- L. Select "USB STORAGE" and then "ERASE MEMORY"
- M. Answer "OK" to the confirmation window



IMPORTANT

Erasing the memory will determine the loss of <u>all the data stored in the hard disk</u>. It's highly recommended to export all meaningful data and results before doing it.

N. Press the HOME button twice to check the new SW version.



4.7.2 Setting Date & Time

Select the "DATE-TIME" icon



Set date and time by using the the UP and DOWN arrows, then confirm by pressing OK.





4.7.3 Diagnostics



By selecting this function, the Hot/Cold Plate will perform a self-diagnostics automatic test.

The device will perform a check of the temperature range, from -5 to 65°C.

If the USB drive is inserted in its slot, data about self-diagnoses can be exported and viewed as a diagram.



4.7.4 Reset HD



This button resets the HD, which contains ALL THE EXPERIMENTAL DATA.

A message box will ask to confirm the action.



ATTENTION:

After confirming the procedure, it will be impossible to recover the data.

4.7.5 Offset



The plate temperature is precisely regulated via an embedded temperature sensor. The "OFFSET" function, which allows modifying the default settings of the reading temperature sensor, is for **factory settings only**.



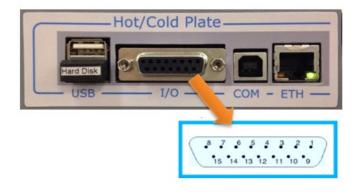
ATTENTION:

Acting on the temperature offset means altering the default settings for the temperature detection, and consequently the reliability of the temperature measurement has to be ascertained.

4.8 Signal Output

For some sophisticated application, and interface with other systems, it might be useful to obtain outputs for temperature, mode, and status.

The 15-pin D-SUB connector (see picture and paragraph 3.8) provides different output trigger, in the form of TTL signals.





4.8.1 Pin-Out

Pin No.	DESCRIPTION	LEVEL LOW	LEVEL HIGH
Pin 1	COUNTER STATUS	TIMER OFF	TIMER ON
Pin 2 to 6	NOT USED		
Pin 7	TEMPERATURE	REACHING	REACHED
Pin 8 to 13	NOT USED		
Pin 14 -15	GROUND		

5 MAINTENANCE

While any service of the instrument ought to be carried out by Ugo Basile personnel or by qualified personnel authorized by UGO BASILE organization, this manual section describes normal maintenance procedures which can be carried out at your facility.



<u>UNPLUG THE MAINS CORD BEFORE CARRYING OUT ANY</u>
<u>MAINTENANCE JOB!</u>

5.1 Electrical

To inspect and/or replace the fuses, **disconnect the mains cable first!** Insert a miniature screwdriver in the slot indentation, see paragraph 3.5.2, and snap out the slide which houses the fuses. Use T5A fuses for operation at both 230 or 115 Volts.

Snap in the fuse slide: the mechanical "click" ensures that it is locked.

5.2 Cleaning

Hot/Cold Plate 35150 does not require any maintenance apart from normal cleaning.



Do not use organic solutions, on the restrainer and cabinet, as they are liable to impair the cylinder transparency and to crack the acrylic components and the touch pad.

Cotton wool and water can be used for cleaning purposes. For disinfection, use a non-alcoholic disinfectant, or H2O2.

5.3 Long Inactivity

The instrument does not require any particular maintenance after long inactivity, except cleaning.



5.4 Customer Support

For any further information you may desire concerning the use and/or maintenance of the Hot/Cold Plate, please do not hesitate to contact our **service department** (or our local distributor) either directly of via our support page http://www.ugobasile.com/support.html:



UGO BASILE s.r.l.

Via G. Di Vittorio 2

21036 GEMONIO - Varese, ITALY



Phone: +39 0332 744574



service@ugobasile.com logistics@ugobasile.com sales@ugobasile.com

<u>Before sending any instrument to our factory for repair</u>, please contact our logistics department to obtain a return authorization number (RMA) and shipping/packing instructions. We may not be held responsible for damages during transport due to poor packing; whenever possible, please use the original packing.

6 35150 SPECIFICATIONS

General	
Commands	4"3 touch-screen (resistive)
Read-out	on the touch-screen
Power Requirement	Universal input 85-264 VAC, 50-60Hz, 40W max.
Sound Level	< 60 dB (A)
Operating Temperature	18° to 25°C see paragraph 2.1.1
Operation	
Temperature	adjustable in the range -5.0 to 65.0°C, with 0.1°C increments
Precision	+/- 0.1°C
Mode	hot plate, cold plate, ramp (increasing or decreasing)
Counting Start	by START button or pedal switch
Counting Stop	by pedal switch
Latency Time Counter	up to 999.9 seconds, in 0.1 steps
Data Acquisition	via X-PAD software, provided
Data Portability	by USB flash drive, provided
TTL Output	for temperature and counter status



Data	exported to Text, Excel or Pdf, or saved to cloud to Drop- Box, OneDrive, GoogleDrive
Configurations	exported to Text, Excel or Pdf, or saved to cloud to Drop- Box, OneDrive, GoogleDrive
Physical	
Total Weight	8Kg
Shipping Weight	12Kg approx.
Dimensions	25(w)x37(d)x47(h)cm – including restrainer
Packing Dimensions	68x34x28cm
Warranty	35150 is covered by a 24-month warranty

7 ORDERING INFORMATION

35150	Hot/Cold Plate, complete with standard accessories			
35150-001	Cabinet (controller with touch-screen and Plate assembly)			
35100-286	Cylindrical Animal Restrainer, for Mice and Rats, 25cm height			
35150- 320pippo	Restrainer Lid M-LM 222			
37215-303	Pedal Switch			
E-AU 041	USB pen drive, including: 35150-302 Instruction Manual X-PAD Software Package			
E-WP008	Mains Cord – Europe (or E-WP008-1 U.S.A. / E-WP008-2 U.K.)			
Set of 2 fuses				

7.1 Thermal-Escape Set-Up

The 35250 **Thermal-Escape Device** includes a complete 35150 Hot/Cold Plate and the following additional parts:

35150-002 Additional Hot Plate

35250-003 Set of Tubes/Catwalk for 35250

7.2 Optional

For high-throughput tests, the combination of a 35150 Hot/Cold Plate with a basic, heating only additional hot plate can be taken into consideration. Temperature range is from 20°C (or room temperature, if >20°C) to 65°C.



The 35150-002 is a basic unit and does not record the temperature and reaction time, which can be noted manually, but it is of great help during the habituation phase before the test, reducing the use of the main unit to the test proper.

As it is an independent unit, it can be used for basic tests as well, using a stop-watch to detect latency time.

Operation is easy and straightforward, as the operator is simply required to enter the temperature by the arrows positioned on the front panel.



Model **35160** includes both a standard 35150 and a 35150-002:

35150-002 Additional Hot Plate (heating only)

35160 Combo Package including Hot/Cold Plate NG, complete with dedicated software, USB output, USB flash drive, and Hot Plate 35150-002. Universal input 85-264 VAC, 50-60Hz

8 BIBLIOGRAPHY

8.1 Method Papers

N.B. Eddy & D. Lembach: "Synthesis, Anti-Inflammatory and Analgesic Activity of Pyrido [2, 1-b] Quinazoline Derivatives" J Pharma Exp 953: 703-385, 1953

8.2 Papers Dealing With Ugo Basile Hot/Cold Plate

- A. Yamamoto et alia: "Two Different Avian Cold-Sensitive Sensory Neurons: Transient Receptor Potential Melastatin 8 (TRPM8)-Dependent and -Independent Activation Mechanisms" Neuropharmacol 111: 130-141, 2016
- Y.H. Huang et alia: "Pulsed Radiofrequency Attenuates Diabetic Neuropathic Pain and Suppresses Formalin-Evoked Spinal Glutamate Release in Rats" Intl. J. Med. Sciences 13 (12): 984-991, 2016
- C.V. Möser: "TANK-Binding Kinase 1 (TBK1) Modulates Inflammatory Hyperalgsia by Regulating MAP Kinases and NF-κB Dependent Genes" J. Neuroinflammation 12:100, 2015



- S.H. Lin et alia: "Genetic Mapping of ASIC4 and Contrasting Phenotype to ASIC1a in Modulating Innate Fear and Anxiety" <u>Eur. J. of Neuroscience</u> 41(12): 1553–1568, 2015
- L. Zvejniece et alia: "R-Phenibut Binds to the α2–δ Subunit of Voltage-Dependent Calcium Channels and Exerts Gabapentin-Like Anti-Nociceptive Effects" Pharmacol. Biochemistry and Behav. 137: 23-29, 2015
- S.J. Wang et alia: "Acupoint Specificity on Acupuncture Regulation of HYPOTHALAMIC- Pituitary-Adrenal Cortex Axis Function" <u>BMC Complem.</u>
 & Alternative Med. 15: 87, 2015
- W. Kallenborn-Gerhardt et alia: "Nox2-Dependent Signaling Between Macrophages and Sensory Neurons Contributes to Neuropathic Pain Hypersensitivity" Pain 55(19): 2161-2160, 2014
- M.W.H. Schaap et alia: "Measuring Pain-Related Behaviour in Four Inbred Rat Strains. Differences in Hot Plate Behaviour" Proceedings of Measuring Behavior 2012, MB 2012
- D. Piomelli et alia: "Anandamide suppresses pain initiation through a peripheral endocannabinoid mechanism" Nature NSC, 2010



We do the search for you: we weekly browse bibliography and link new papers to the bibliography section of each UB device.

Don't forget to check our web page periodically for updated bibliography!

INSTRUCTION MANUAL June 2017

REVISION 2

Notes

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CE CONFORMITY STATEMENT

Manufacturer UGO BASILE srl

Address Via G. di Vittorio, 2 – 21036 Gemonio, VA, ITALY

Phone n. +39 0332 744574

Fax n. +39 0332 745488

We hereby declare that

Instrument. HOT/COLD PLATE

Catalog number 35150 - 35150-002 - 35250

is manufactured in compliance with the following European Union Directives and relevant harmonized standards

- 2006/42/CE on machinery
- 2014/35/UE relating to electrical equipment designed for use within certain voltage limits
- 2014/30/UE relating to electromagnetic compatibility
- 2011/65/UE and 2015/863/UE on the restriction of the use of certain hazardous substances in electrical and electronic equipment

Account *Manager* Mauro Uboldi

Nome / Name

October 2018

Date Firma / Signature

MOD. 13 Rev. 1



Monza, May 24, 2016

Laboratory Test and Certification: 35150 Hot/Cold Plate NG

We hereby certify that the 35150 Hot/Cold Plate NG has been tested in our laboratory for 2 weeks, in order to identify any possible trouble with the use with animals.

Support has been given to Ugo Basile R&D Team to solve those issues and to incorporate improvements based on our experience.

Particular care has been taken in comparing the new NG features with the previous models by Ugo Basile.

Best regards

Guido Cavalettí

Head, Experimental Neurology Unit - Director, PhD Program in Neuroscience

School of Medicine and Surgery - University of Milano-Bicocca

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