PULSE V2 system

User's manual, V2.12, updated on 22 July 2022

QUICK SETUP

1. Connect the 12 V power adapter to a power outlet and to the 12V DC red socket on top of the unit.
2. Connect up to 10 sensors to the blue connectors on top of the unit.
3. Glue the sensor tips to the shell of the animals to be monitored, above the heart. Attaching the sensor right on top of the heart is probably the most important single aspect in getting a good signal.
4. Turn the PULSE V2 "ON" using the green button on the top right.
5. Cycle through the 10 available channels using the forward and backward black buttons.
6. If necessary, adjust the amplification on each channel using the corresponding black knobs numbered 1-10 at the front of the device.
7. Long-press the backward button to connect to a WiFi network and use FileZilla on your PC to edit the settings file and download logged data.

AVAILABILITY

PULSE V2, as well as the latest user's manual and firmware, are available at https://electricblue.eu/pulse.

BASIS OF OPERATION, ORIGINS, AND IMPROVEMENTS

Over the last decades, Infra-Red (IR) emitters and receivers have been widely used to monitor the cardiac frequency of a large variety of hard-shell molluscs or crustaceans. The typical design involves a small sensor combining an IR emitter and receiver, connected through a cable to an amplification and processing unit, whose output is then analysed in real-time in an oscilloscope or in a PC, or stored in memory for later processing.

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Although there are various IR sensors available, the most commonly employed is the Vishay CNY70 (see the sensor section further below). The sensor is fixed to the exoskeleton of an animal, above its heart. Calcium carbonate and chitin shells allow the IR light from the sensor to pass through, and it illuminates the heart and nearby circulatory vessels. A portion of that light bounces back and is detected by the sensor. Changes in the shape or volume of the circulatory structures during a heart contraction, or heartbeat, change the amount of IR light that bounces back from the animal's internal anatomy to the IR detector. These changes in reflected IR light, transduced to changes in electrical current, are then processed by the electronic circuit and by the software, and plotted or stored in a log file.

Regarding the amplification circuitry, the most widely used design is based on the CAPMON system originally developed by Depledge & Andersen (1990) in the late 1980s, and updated by us in the early 2010s (Burnett et al., 2013). This design has been successfully applied to limpets (Chapperon et al., 2016; Seabra et al., 2016), mussels (Collins et al., 2020; Moyen et al., 2019; Olabarria et al., 2016; Seo et al., 2016), scallops (Gurr et al., 2018), or fiddler crabs (Levinton et al., 2020), to name just a few. During the last decade, we made several improvements to the electronics in order to increase the number of channels simultaneously monitored, or to add data-logging capacity to the electronics package (thus avoiding the need for a dedicated oscilloscope or PC just to record the data). These efforts resulted in a compact, self-contained eight-channel heart frequency logger which we called PULSE v1. As much useful as it was, however, this design still had some limitations:

1. The eight channels were not sampled simultaneously, but instead sequentially. Although this did not matter for most applications, it could be limiting in others.
2. The system lacked any form of wireless connectivity.
3. It was based on Arduino IDE, which was too complicated and cumbersome for many users. Firmware updates, in particular, were quite difficult.
4. Although the system could run independently from an oscilloscope or PC, a PC was still needed for plotting heart beat signals during the experimental set-up, and whenever a real-time check of signals was needed.
5. The extreme signal amplification employed could, in specific circumstances, originate false oscillations from sensor noise. Although not common, this was particularly problematic as it could introduce artifacts in the logged data.

Consequently, we redesigned the whole system from ground up, and called it **PULSE V2**. The main improvements already implemented are:

1. Totally new and simplified amplification circuit, more responsive and virtually immune to positive feedbacks.
2. Individually regulated amplification for each channel.
3. Permanent and simultaneous reading/logging of all channels.
4. Splash-proof (IP 64) enclosure, as well as splash-proof potentiometers, buttons, switches and cable connections.
5. WiFi connectivity for easy data transfer over FTP. Ability to join a pre-existing WiFi or to create its own.

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6. 3.5" LCD screen.
7. 10 independent channels.
8. Easy OTA (over the air) firmware updates.

Apart from these improvements, there are others that we plan implementing **in the future**. These include:

1. A smartphone app that connects to the device via Bluetooth (thus leaving the smartphone's WiFi connection available for other apps), and that plots, on the Smartphone's screen, the 10 channels side by side, allowing for an overall inspection of the PULSE performance at any moment.

2. The automatic detection of cardiac frequency on each channel, which is periodically saved together with the raw data. This will dramatically decrease the post-processing time, especially considering that the device has 10 channels which are permanently logging. At this moment, frequencies have to be manually calculated from the raw data files, although we will be working on an R package to ease this task.

3. Development of a splashproof, induced-charged battery pack to easily allow for using in the field.

Fig. 1: PULSE V2, overall description.

SYSTEM DESCRIPTION

The **PULSE V2** (**Fig. 1**) is composed of:

- A splash-proof box (IP 64) with 10 knobs (potentiometers) in the front, and 3 buttons and 11 connectors on top.
- Up to 10 cables with CNY70 heartbeat sensors at their end, attached to the blue connectors.
- A 12 V AC/DC wall adapter attached to the red connector.

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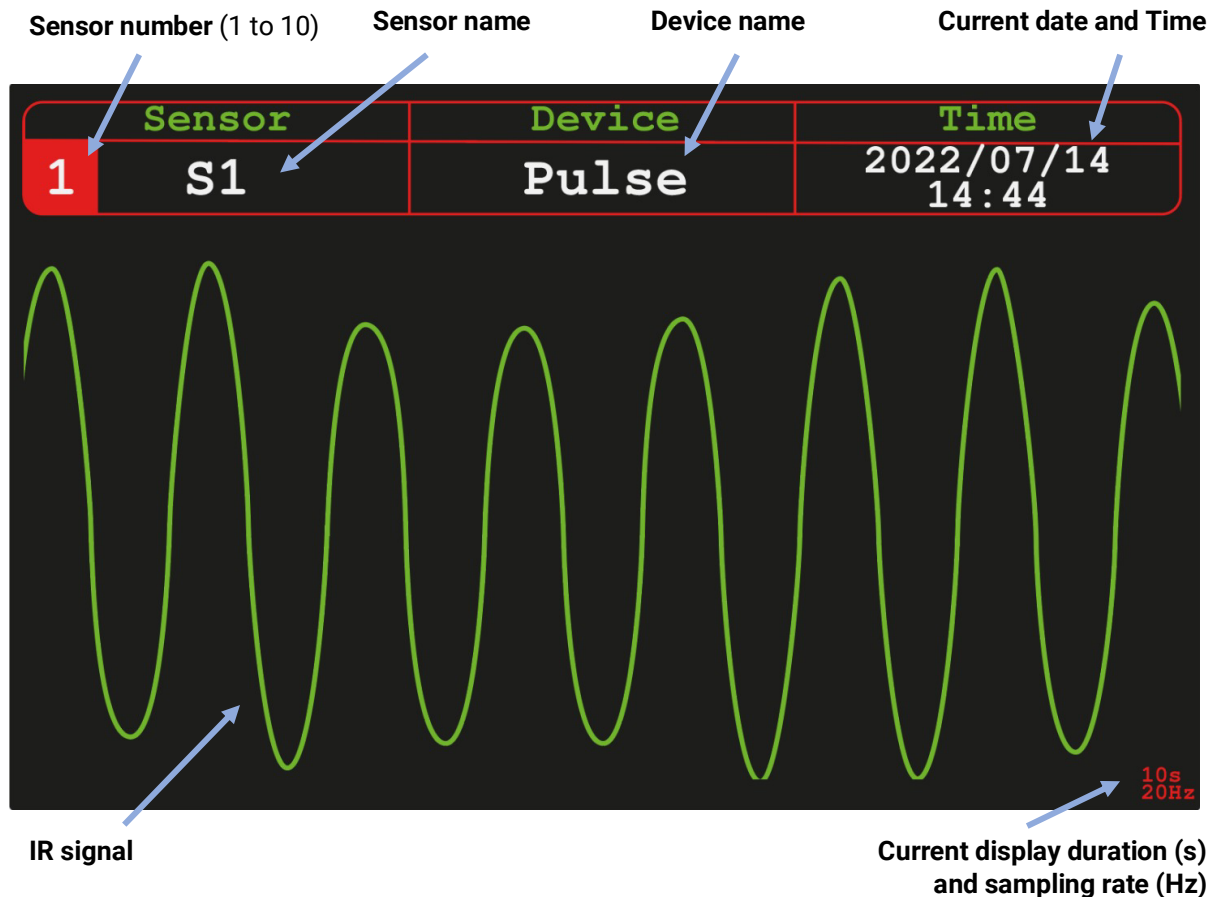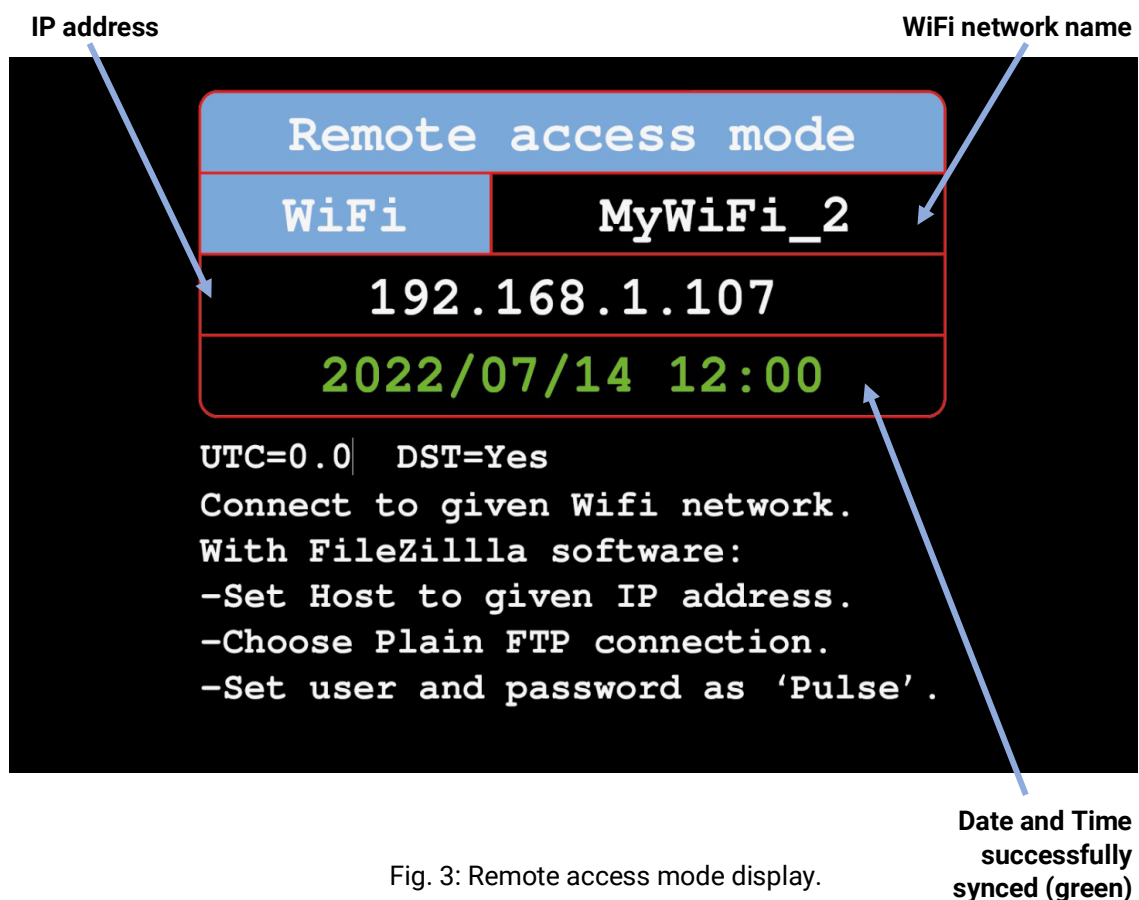
Fig. 2: Readings display.

REGULAR OPERATION

1. First ensure the 12 V power wall adapter is connected to the mains electricity and to the PULSE V2.

2. To power the device, you must press the **Power Button**. If power is available, a red LED will light up inside the **Power Button**.

3. Once powered, the PULSE V2 will read the settings file (**Pulse_settings.txt**, see below) and immediately start collecting readings from all 10 sensors and storing them in its internal memory as CSV files.

4. CSV files will be automatically created hourly, and each CSV file will contain all the readings from the 10 sensors that were collected during that hour. The frequency at which the sensors are being sampled depends on the **Rate_Hz** field set on the configuration file (see the **SETTINGS** section below).

5. Initially, the display will present the readings for sensor 1 continuously (**Fig. 2**). To change the sensor being plotted in the LCD display, you can press the **Forward Button** or the **Backward Button**. To set automatic rotation of the channel being plotted, press **Forward Button** together with **Backward Button** for a moment. A warning "Autoscroll ON" should show on the screen for a few seconds, and a letter 'A' will remain in the top left corner of the plotted graph as a reminder. To turn off automatic channel rotation, just long-press **Forward Button** and **Backward Button** again. The "Autoscroll OFF" warning will show in the screen for a few seconds, and the letter 'A' on the top left corner of the display will disappear.

6. It is important to remind that all 10 channels are constantly being recorded, regardless of the channel that is being plotted at any given moment.

7. The amplification level can be adjusted independently for each channel, turning the corresponding potentiometer (black knobs) numbered 1-10 in the front of the device. This may be useful to ensure an adequate level of amplification to each monitored animal. Too little amplification and the heart beat may be indistinguishable from the background noise. Too much amplification and the signal may become clipped, making it hard to individualize and identify heart beats. Importantly, the amplification level not only changes the shape of the plot, but also the saved data (i.e., permanently affects the data collected).

8. To retrieve the CSV files saved in memory, to edit the Pulse_settings.txt file or to sync the unit's time, you must leave the data plotting mode and enter the '**REMOTE ACCESS MODE**' (see below).

IP address

WiFi network name

Date and Time successfully synced (green)

Fig. 3: Remote access mode display.

REMOTE ACCESS MODE

This mode is used to connect the computer to the PULSE V2 in order to either change its settings, update firmware, or to retrieve logged data. **While in this mode, the system will not collect sensor data**.

1. To enter this mode press and hold the **Backward Button** for 5 seconds. To exit, briefly press again the **Backward Button,** and the PULSE V2 will restart, re-read its **settings file** and resume data collection.

2. In the remote access mode, you should be presented with a different display in the LCD (**Fig. 3**). Upon entering this mode, PULSE V2 will get the WiFi credentials from the **settings file** and attempt to connect to that network.

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NOTE 1: the first time this is done, because there are yet no WiFi credentials in the settings file, the system will automatically create its own WiFi network, with name and password '**Pulse_AP**'. It will also define its IP address as **10.10.10.10**. At this point you can use your computer to edit the settings file, configuring the PULSE V2 to connect to an external network (see the **FTP CONNECTION** and **SETTINGS** sections below).

NOTE 2: Strictly speaking, however, the device does not need to be connected to an external WiFi network. If no WiFi credentials are ever given in the settings file, the device will always create its own WiFi network, with name and password '**Pulse_AP**', and with **10.10.10.10** as IP address. This is enough to enable editing other parameters in the settings file and to enable transferring data files from PULSE to a computer through an FTP client (see more details below). The downside is that the device will not sync its clock because it cannot reach an external network. Also, the computer that will be connected to the PULSE V2 will have to disconnect from its usual WiFi network to connect to the '**Pulse_AP**' network, and therefore will not have online access.

3. The device will attempt to connect to the given external WiFi network for 25 seconds.

   a. If it fails to connect, it will go back to the default mode and create its own WiFi network, with name and password '**Pulse_AP**' and IP address as **10.10.10.10**, as described above.

   b. In case it succeeds, the external WiFi name and the new IP will be shown in the LCD (**Fig. 3**) and you can now proceed to edit the settings file or to download data (see the following sections below). Importantly, if PULSE V2 gets connected to a WiFi network with internet access, it will attempt to sync its clock using the pool.ntp.org service. If this succeeds, the colour of the time being plotted in the '**REMOTE ACCESS MODE**' will change from white to green (**Fig. 3**), indicating that the time has been updated. The sync process will take approx. 10 sec.

FTP CONNECTION

Once PULSE V2 is in '**REMOTE ACCESS MODE**' (either connected to an external network or creating its own network, '**Pulse_AP**'), and an IP address is shown in the display, it starts acting as an FTP server, accepting the connection of an FTP client (a computer with an FTP program installed).

To do this:

1. On your computer, install an FTP client of your preference. We recommend the freely available software **FileZilla**, from https://filezilla-project.org/download.php?type=client#close. There are versions for Linux, Windows and Mac. The following instructions apply to FileZilla, but shouldn't be too different for other software packages.

2. Once FileZilla is installed, you must connect the computer to the same WiFi as the PULSE V2.

3. On FileZilla, click on the '**Open the Site Manager**' button on the top left (**Fig. 4**).

4. On the site manager window, click '**New site**'.

5. On the site's hierarchy tree displayed, the new entry is initially called 'New site', but you can edit its name to something more informative of your choosing.

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6. Still within the site manager window, in the **Host** field of the **General** tab, you must insert the **IP address** displayed in the PULSE V2 screen (**Fig. 3**).

7. In the **Encryption** drop down menu, you must choose '**Only use plain FTP (insecure)**'. This is because the microcontroller in charge of the PULSE V2 functioning does not work with encryption data. This will not expose your computer to any harm, as it only pretrains to the connection between the computer and PULSE V2.

8. Type in '**Pulse**' in both **User** and **Password** fields. These credentials are hardcoded in the PULSE V2 firmware and cannot be changed.

9. In the '**Transfer settings**' tab, select the '**Limit number of simultaneous connections**' box.

10. You can now click '**Connect**'. Once the connection is established, you'll be presented with the file storage tree held in your PULSE V2's memory (**Fig. 5**).

11. You can now right click on any file and choose to download or edit. Pay attention to the '**Local site**' window, as it shows where your downloaded files are being saved to (i.e., on your computer side). You can change this simply by selecting any folder on your computer in the file hierarchy tree below this field.

12. Data files are named **YYYY_mm_dd_XXhXXmin.csv**, for example 2022_7_15_13h41min.csv. You can see an example of a data file at the end of this manual.

13. To edit the settings file (Pulse_settings.txt), you can drag and drop a new file, or right click on it and select '**View/Edit**', and save it after editing. You can see an example of a settings file at the end of this manual.
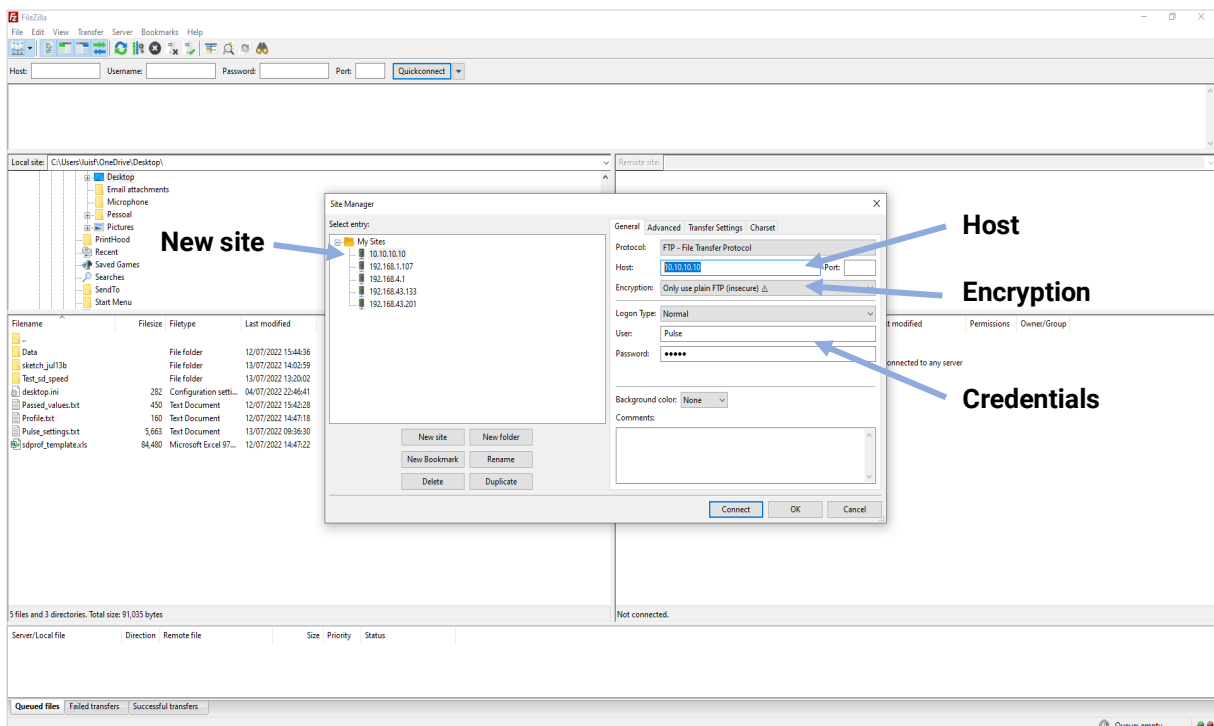
Fig. 4: Creating a new connection in FileZilla interface.

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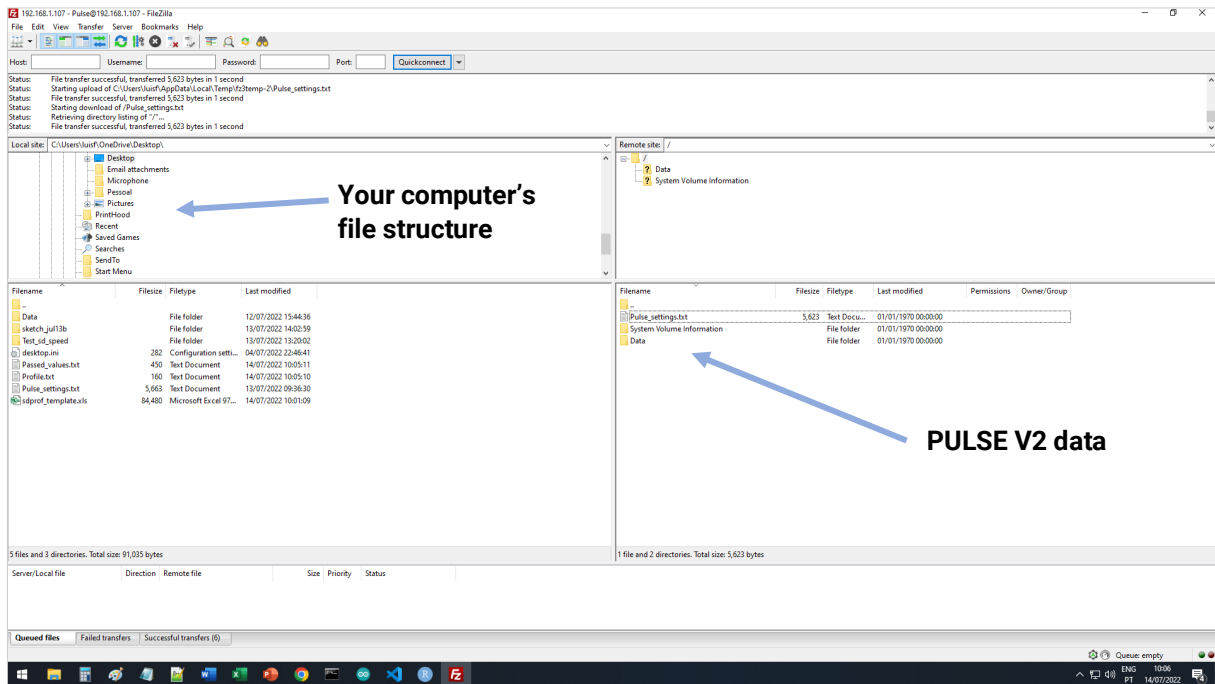
Fig. 5: Accessing your data in FileZilla interface.

SETTINGS

In the file 'Pulse_settings.txt', available through FTP (see above), you can define several variables that control the system behaviour. The currently available options are:

Device_name = Pulse_7

This field defines the name given to this particular device. The name will be shown in the LCD display during data logging and will be added to the data CSV header, so you can always track which device produced each file. This field is limited to 12 characters. If no name is provided, the default name, **'Pulse'**, will be used. This is best used to distinguish between different units assigned to different experimental treatments.

Rate_Hz = 20

Here you can define the frequency at which the sensors will be sampled, in Hz. For example, 20 Hz means that 20 samples are taken per second, or one sample every 50 milliseconds (1000 ms/20 Hz = 50 ms). This value must be an integer (no decimals) and can vary from 5 to 25 Hz. If no value is given or if it is outside the range, it will be set to **10 Hz**. Higher numbers result in larger file sizes, while lower values can lead to fast heart rates to be hard to analyze. We recommend the default 10 Hz for most uses, but an initial assessment of the appropriate rate should always be performed before important data is collected.

Wifi = My_wifi

The Wifi field sets the name of a known WiFi network to which PULSE V2 will connect to. Connecting to this external network means that your computer will retain internet access even when connected to PULSE V2. This

field is limited to a length of 14 characters. In case you don't provide a WiFi name here, PULSE V2 will not attempt to connect to any WiFi network and will instead create its own WiFi network, called '**Pulse_AP'**.

Wifi_pass = Wifi_password

The **Wifi_pass** field provides the password required for the WiFI network defined in the **Wifi** field. Password is limited to a length of 30 characters. If no password is given, the system will assume that the WiFi network provided has no password.

Local_time_zone = 0

This value represents the deviation (positive or negative) from UTC (UTC means Universal Time Coordinated). In other words, it represents the local time zone. When connected to the internet, PULSE will sync its clock and use this value to adjust itself to local time. It accepts values from -12 to 12 with half unit increases. For example, someone in the UK should use '0', in Germany should use '1', in Brazil should use '-3', and in South Australia '9.5'. If the given value is outside these limits, local time will be set to UTC+0. The time shown in the display and the logged files stored in the internal memory will use local time. Because this information is printed in the header of the CSV file, any future analysis will know under which time zone data were collected.

DST = Yes

DST means Daylight Saving Time. This can be set to 'Yes', 'yes', 'Y', or 'y' to consider DST adjustment (+1 hour). Any other text will be interpreted as a negative choice, meaning that no extra hour will be added. For clarity it is recommended typing 'Yes' or 'No'. The time shown in the display and in the logged data files will incorporate the DST setting. It will also be printed in the header of the CSV data files, and thus that information will be available for posterior data verification and processing. **This option will not adjust itself automatically at the end of Summer**. Rather, you must edit again when appropriate to turn it off.

Display_duration = 10

This variable defines the number of seconds displayed in the plot window. It does not affect data collection or their storage in the internal memory, only the length of the display window in the LCD screen. It is limited to a minimum of 1 sec and maximum of 60 secs. Values that surpass limits will be capped to the nearest limit. Only integer values are accepted (no decimals; e.g., '10' and not '10.2'). If left blank or if the value is not recognised, it will be set to **10 seconds**.

NOTE: both the fields **Display_duration** and **Rate_Hz** are shown in the bottom right corner of the LCD during regular data logging (Fig 3), so that you can better interpret what is being plotted at any moment.

S1, S2, S3, ... S10

This is an identifier for each PULSE sensor. You can only use letters and numbers, and each name is limited to 12 characters. Names can be, for example, 'Limpet1' or 'mussel78'. These names will also be carried out to the data logs, so you know which data belongs to each monitored animal. If left blank, sensors will be named after their number, i.e., sensor 1 will be named 'S1', sensor 2 will be named 'S2', and so forth.

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Pulse settings	
Device_name	Pulse
Rate_Hz	20
Display_duration	10
Local_time_zone	0.0
DST	Yes
Access Point	Pulse_AP
WiFi	MyWiFi_2
CSV files	5
SD storage	<0.1%

Fig. 6: Settings display mode

SETTINGS DISPLAY MODE

Current settings can always be checked by long-pressing the **Forward Button** for 5 seconds (**Fig. 6**). The LCD will show the **Device_name**, **Rate_Hz**, **Display_duration, Local_time_zone**, **DST**, the **Access Point** name set by PULSE V2 in case no WiFi connection is possible, and the **WiFi** that the system will try to connect to. It will also show the number of CSV files currently stored in memory and the percentage of memory they occupy. **While in this mode, the system will not collect sensor data**. To exit, briefly press again the **Forward Button,** and the PULSE V2 will restart and resume data collection.

FIRMWARE UPDATE

It is advisable to regularly update the system, as new firmware releases correct bugs and bring new functionalities. To do it:

1. Go to the bottom of the PULSE's support page at www.electricblue.eu/pulse and download the latest firmware.
2. The firmware package is in a compressed format (zip). Unzip it.
3. Connect the computer to the same network used by PULSE V2.
4. Press and hold the **Backward Button** for 5 seconds to bring PULSE into the '**REMOTE ACCESS MODE**'. As explained in detail above, PULSE V2 will become connected to an external network or generate its own network. In any case, an IP address will be shown in the display (**Fig. 3**).
5. At this moment, PULSE V2 will host a webpage under the address http://IP/update, where the IP is the one shown in the Remote Access Mode display (**Fig. 3**). For example, if the IP shown in the display is

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10.10.10.10, the new firmware must be uploaded at the address: 'http://10.10.10.1/update'. If the IP is 192.168.1.107, the firmware upload address will be: 'http://192.168.1.107/update'.

6. In a browser, go to that page (http://IP/update), choose the firmware file you just downloaded and unzipped to your computer, and upload it.

7. A progress bar will be shown. After about 30 seconds, once the upload is complete, the PULSE V2 will reboot automatically and the firmware update will be finalized.

SENSOR PLACEMENT

1. The sensors should be glued to the shell or carapace of the target animal. Normally, we use regular liquid superglue (cyanoacrylate). Liquid superglue tends to be clearer once dry than gel superglue, but requires extra care to avoid run-off that could come in contact with the animal's soft tissues and harm it.

2. Attaching the sensor right on top of the heart is probably the most important single aspect in getting a good signal (**Fig. 7**).

3. Study the anatomy of your target species and identify the approximate position of the heart.

4. Clean the shell or carapace with a cloth. Superglue does not work so well if moisture or a biofilm is present.

5. Without adding glue to the sensor, place it in several locations in the animal's shell and choose the exact position to be used. Be aware that signal quality is almost always inferior while the animal is being manipulated.

6. Place a thin layer of superglue on the sensor and reattach it to the target position in the shell. Be careful not to use too much glue, or it may fall on the animal while still liquid.

7. Hold it in place for some seconds while the glue cures. The attachment should be strong and the head of the sensor should lay flat against the specimen.

8. Sensors can be detached by breaking the superglue bind, typically by exerting force in a direction perpendicular to the point of attachment. If a sensor is going to be reattached, remove the superglue from the surface of the sensor and repeat the previous steps.

Fig. 7: A CNY70 sensor glued to a *Mytilus galloprovicialis* (left), and another glued to a *Patella vulgata* (right).

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TIPS AND TRICKS

1. A good and quick way of testing if the system is working as it should is to place one sensor between the indicator and middle fingers, facing the thumb (**Fig. 8**) and press gently. Your own heartbeat should show up in the screen if that sensor is being plotted and if its amplification is adequate (i.e. the corresponding potentiometer should be half-turned).

2. IR sensors do not work well under direct sunlight. That happens because the Infra-Red radiation from the sun is so high that it saturates readings, and therefore the relatively small signal oscillations from heart beats become imperceptible, even with the potentiometers turned all the way up. To overcome this issue and still be able to work in the field, it is advisable to cast shadow on the sensors and animals, or to wrap them in aluminium foil (a small "umbrella" made out of aluminium foil is usually enough).

3. If you are working with the tip of the sensors submerged in water, **ALWAYS** place the system above the water level to prevent water from infiltrating the sensor cables and flooding the entire system. Cables and connectors are waterproof when used correctly, but persistent water ingress via sensor cables can (and will) overcome the waterproofing barriers.

4. If you are using the system on a new species for the first time, it may be hard to find the heart position. Take time to improve your ability to get good readings.

5. It is quite common for the signal to be sub-optimal or even poor after animals have been disturbed. This very much includes the process of gluing the sensor. In many cases, a proper signal can only be discerned after at least 15-30 minutes since they have last been manipulated.

6. If you are using a brand-new sensor in an animal with a thick shell, you may consider shaving off the outer plastic edge of the sensor with sand paper or a Dremell abrasive tool in order to obtain a flat surface (**Fig. 11**), or to shave part of the shell wall to make it thinner.

7. Abrasive tools can also be used to clean used sensors. However, sanding must be done with care to avoid sanding too deep – as a rule-of-thumb a thin layer of old superglue should always be left in the sensor head. Sanding typically leaves the surface looking opaque, but crucially not only it is not opaque to IR, as a new layer of superglue is added the surface will look transparent once more. If done correctly sensors can often be re-used multiple times, however this is not guaranteed.
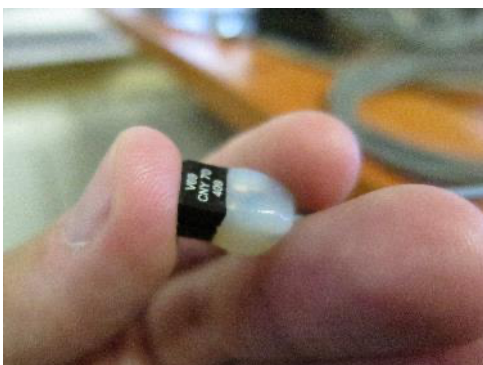
Fig. 8. Testing the system by directing the CNY70 towards the thumb to capture the operator's heartbeat. Caution must be taken not to press too lightly (which will break the close contact between the sensor and the finger), nor too hard (which will decrease circulation and make it harder to see the heartbeat).

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SENSOR REPLACEMENT

The infrared sensor most regularly used for monitoring the heart rate of invertebrates is the Vishay CNY70, but there are a variety of equivalent sensors that can be used. There are many shapes and sizes. Some are cubic, some flat, some have the IR emitter and the receiver together while others have them separated. The CNY70 should be appropriate for most applications, but you can try others. The sensor is connected to a shielded cable that has two wires + shield (Pro Power 3027442 or equivalent) and is waterproofed with acrylic resin. The wire connections are as follows: (i) woven shield wires to ground, (ii) red wire to power and (iii) blue wire for signal.

If handled with caution, each sensor can be glued to an animal, detached and re-used multiple times. Each time a sensor is detached, it is advisable to shave off the remnants of glue, keeping the surface clean and flat. Due to variations in assembling quality (especially due to variations in soldering quality) it is likely that within the same batch of sensors some will last a long time while others may produce sub-optimal readings sooner.

To assemble a new sensor cable follow these steps:
1. Cut the end of the cable, strip the wire ends and tin them (i.e., apply a small amount of solder to the tip of each wire).
2. Take the CNY70 sensor and trim the 4 legs (only a few mm are needed) to reduce the overall size (**Fig. 9**).
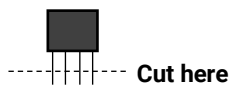
Cut here   Fig.9. Schematics showing how to trim the CNY70 legs.

3. Following the schematic below, and having the sensor with the legs pointing to you and with the marked side up, bend the top right leg towards the low left leg and solder them together (**Fig. 10**, black line).
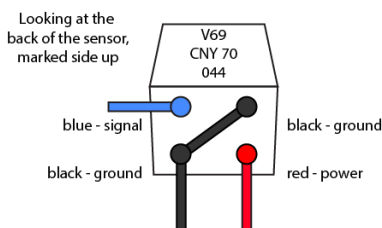
Fig. 10. Wiring the CNY70.

4. Solder the blue wire to the top left leg and the red wire to the bottom right leg.
5. Twist the wire shield and solder it to the bottom left leg (the one that is already connected to the top right leg, shown in black in the figure).
6. Make sure the legs do not touch in order to not short circuit.
7. Cover the legs and the back of the sensor with hot glue or acrylic resin to waterproof everything.
8. (Optional) Shave the outer plastic lip of the sensor with a Dremell abrasive rotary tool to flatten the face of the sensor (**Fig. 11**).
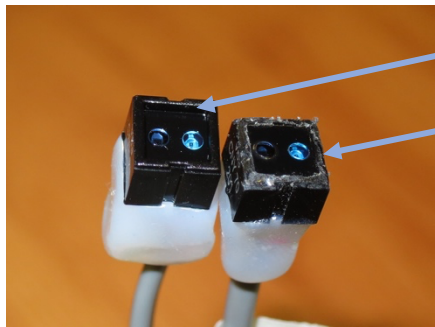
Intact CNY70 sensor (with outer lip).

CNY70 sensor without the outer lip.

Fig. 11. Two CNY70 sensors side by side. The one on the left is intact, but the other is missing its outer lip, which was removed with an abrasive tool.

TECHNICAL CHARACTERISTICS

Waterproof rating	IP 64
Number of independent channels	10
Power input	12V DC
Available memory	32 GB
Sampling frequency	5 to 25 Hz
RTC drift	Up to 10 seconds/month
Connectivity	WiFi and Bluetooth (available in the next firmware update)
Weight	650 g, 1400 g with 10 sensors
Dimensions	160.0 mm wide x 100.6 mm deep x 133.6mm tall (180 mm tall with attached sensors)
Operational temperature	-10 to +60 ºC
Vishay CNY70 sensors	5 V, 50 mA, 950 nm

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REFERENCES

Burnett, N. P., Seabra, R., de Pirro, M., Wethey, D. S., Woodin, S. A., Helmuth, B., Zippay, M. L., Sarà, G., Monaco, C., & Lima, F. P. (2013). An improved noninvasive method for measuring heartbeat of intertidal animals. *Limnology and Oceanography: Methods*, *11*(FEB). https://doi.org/10.4319/lom.2013.11.91

Chapperon, C., Volkenborn, N., Clavier, J., Séité, S., Seabra, R., & Lima, F. P. (2016). Exposure to solar radiation drives organismal vulnerability to climate: Evidence from an intertidal limpet. *Journal of Thermal Biology*, *57*. https://doi.org/10.1016/j.jtherbio.2016.03.002

Collins, C. L., Burnett, N. P., Ramsey, M. J., Wagner, K., & Zippay, M. L. (2020). Physiological responses to heat stress in an invasive mussel Mytilus galloprovincialis depend on tidal habitat. *Marine Environmental Research*, *154*, 104849. https://doi.org/10.1016/j.marenvres.2019.104849

Depledge, M. H., & Andersen, B. B. (1990). A computer-aided physiological monitoring system for continuous, long-term recording of cardiac activity in selected invertebrates. *Comparative Biochemistry and Physiology Part A: Physiology*, *96*(4), 473–477. https://doi.org/10.1016/0300-9629(90)90664-E

Gurr, S. J., Goleski, J., Lima, F. P., Seabra, R., Gobler, C. J., & Volkenborn, N. (2018). Cardiac responses of the bay scallop Argopecten irradians to diel-cycling hypoxia. *Journal of Experimental Marine Biology and Ecology*, *500*. https://doi.org/10.1016/j.jembe.2017.12.011

Levinton, J. S., Volkenborn, N., Gurr, S., Correal, K., Villacres, S., Seabra, R., & Lima, F. P. (2020). Temperature-related heart rate in water and air and a comparison to other temperature-related measures of performance in the fiddler crab Leptuca pugilator (Bosc 1802). *Journal of Thermal Biology*, *88*. https://doi.org/10.1016/j.jtherbio.2019.102502

Moyen, N. E., Somero, G. N., & Denny, M. W. (2019). Impact of heating rate on cardiac thermal tolerance in the California mussel, Mytilus californianus. *Journal of Experimental Biology*. https://doi.org/10.1242/jeb.203166

Olabarria, C., Gestoso, I., Lima, F. P., Vázquez, E., Comeau, L. A., Gomes, F., Seabra, R., & Babarro, J. M. F. (2016). Response of two mytilids to a heatwave: The complex interplay of physiology, behaviour and ecological interactions. *PLoS ONE*, *11*(10). https://doi.org/10.1371/journal.pone.0164330

Seabra, R., Wethey, D. S., Santos, A. M., Gomes, F., & Lima, F. P. (2016). Equatorial range limits of an intertidal ectotherm are more linked to water than air temperature. *Global Change Biology*, *22*(10). https://doi.org/10.1111/gcb.13321

Seo, E., Sazi, T., Togawa, M., Nagata, O., Murakami, M., Kojima, S., & Seo, Y. (2016). A portable infrared photoplethysmograph: heartbeat of Mytilus galloprovincialis analyzed by MRI and application to Bathymodiolus septemdierum. *Biology Open*, *5*(11), 1752–1757. https://doi.org/10.1242/bio.020909

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EXAMPLE OF SETTINGS FILE

```
Device_name=Pulse
Rate_Hz=10
Wifi=MyWify2
Wifi_pass=ThisIsMyPass
Local_time_zone=0
DST=Yes
Display_duration=10
S1=Limpet_1
S2=Limpet_2
S3=Limpet_3
S4=Mussel_1
S5=Limpet_4
S6=Mussel_2
S7=Limpet_5
S8=Mussel_3
S9=Not_used1
S10=Not_used2


//-----------------------------------------------------------//
// For use with firmware version Pulse_v2.3:                 //
// PLEASE refer to the instructions manual for more info.    //
//-----------------------------------------------------------//
//--||--||--||--||--||--||--||--||--||--||--||--||--||--||--//
//-----------------------------------------------------------//
// Device_name                                               //
//   an identifier for the PULSE logger                      //
//   DO NOT use spaces ('Pulse_xyz', not 'logger xyz').      //
//   DO NOT use '=' or '.'                                   //
//   preferably stick to only "abc..z", "0-9", "_" and "-"   . //
//   preferably use an device name with 12 characters or less . //
//   If left blank, Device will be named 'Pulse'.            //
//-----------------------------------------------------------//
// Rate_Hz                                                   //
//   Rate at which voltage values are read in integer Hz     //
//   , no decimals.                                          //
//   Use the lowest possible Rate_Hz to avoid oversampling.  //
//   Typically 20 works well (readings every 50 milliseconds). //
//   A Rate_Hz that is too high may degrade system performance. //
//   A Rate_Hz that is too low results in ladder-like data.  //
//   Rate_Hz is limited between 5Hz and 25Hz.                //
//   If left blank, Rate_Hz will be set to 10Hz.             //
//-----------------------------------------------------------//
// Wifi                                                      //
//   Name of Wifi network used to communicate via FTP.       //
//   Limited to 14 characters.                               //
//   If left blank, there will not be an attempt to connect to //
```

EXAMPLE OF A LOG FILE

```
————————————————————————————,————————————————————,,,,,,,,,
www.electricblue.eu, Portugal,,,,,,,,,
————————————————————————————,————————————————————,,,,,,,,,
Pulse version,V2.9,,,,,,,,,
————————————————————————————,————————————————————,,,,,,,,,
Device,Pulse,,,,,,,,,
Rate_Hz,25,,,,,,,,,
UTC:,0,,,,,,,,,
Day light saving time:,Yes,,,,,,,,,
Window_Sec:,60,,,,,,,,,
S1,S1,,,,,,,,,
S2,S2,,,,,,,,,
S3,Limpet_3,,,,,,,,,
S4,Mussel_4,,,,,,,,,
S5,Limpet_5,,,,,,,,,
S6,S6,,,,,,,,,
S7,Limpet_7,,,,,,,,,
S8,Rock,,,,,,,,,
S9,S9,,,,,,,,,
S10,S10,,,,,,,,,
————————————————————————————,————————————————————,,,,,,,,,
Time,S1,S2,Limpet_3,Mussel_4,Limpet_5,S6,Limpet_7,Rock,S9,S10
2022-07-18 13:55:43.243,1168,1495,1487,1463,1467,1516,1475,1548,1420,1493
2022-07-18 13:55:43.282,1045,1510,1507,1483,1493,1522,1488,1564,1436,1520
2022-07-18 13:55:43.322,1215,1528,1533,1507,1534,1543,1515,1599,1472,1576
2022-07-18 13:55:43.362,1264,1539,1554,1520,1557,1555,1527,1614,1482,1599
2022-07-18 13:55:43.402,654,1552,1565,1530,1568,1565,1531,1619,1493,1606
2022-07-18 13:55:43.442,0,1556,1568,1534,1574,1573,1542,1623,1497,1612
2022-07-18 13:55:43.482,0,1559,1575,1535,1575,1573,1546,1632,1501,1615
2022-07-18 13:55:43.522,84,1552,1558,1527,1568,1564,1541,1623,1493,1603
2022-07-18 13:55:43.562,785,1551,1560,1524,1566,1563,1529,1618,1488,1598
2022-07-18 13:55:43.603,1360,1547,1559,1529,1563,1567,1535,1616,1490,1599
2022-07-18 13:55:43.642,1683,1549,1559,1526,1558,1563,1526,1612,1490,1589
2022-07-18 13:55:43.682,1885,1535,1554,1521,1552,1559,1524,1610,1483,1584
2022-07-18 13:55:43.722,1890,1535,1552,1523,1551,1559,1523,1610,1484,1585
2022-07-18 13:55:43.762,1605,1533,1551,1519,1545,1554,1523,1608,1478,1583
2022-07-18 13:55:43.802,1345,1531,1546,1520,1535,1552,1520,1605,1478,1578
2022-07-18 13:55:43.842,1631,1534,1549,1518,1535,1553,1522,1603,1480,1579
2022-07-18 13:55:43.882,1611,1533,1546,1513,1534,1554,1520,1600,1472,1572
2022-07-18 13:55:43.922,1214,1528,1535,1507,1526,1552,1518,1594,1472,1571
2022-07-18 13:55:43.962,1085,1525,1545,1514,1530,1552,1520,1599,1469,1568
2022-07-18 13:55:44.000,1441,1526,1533,1509,1522,1545,1515,1598,1468,1563
2022-07-18 13:55:44.040,1772,1520,1522,1499,1521,1535,1511,1587,1463,1553
2022-07-18 13:55:44.080,2115,1522,1527,1501,1517,1544,1504,1586,1458,1552
```