instruction manual

P.A.M.
Pressure Application Measurement
Cat. No. 38500
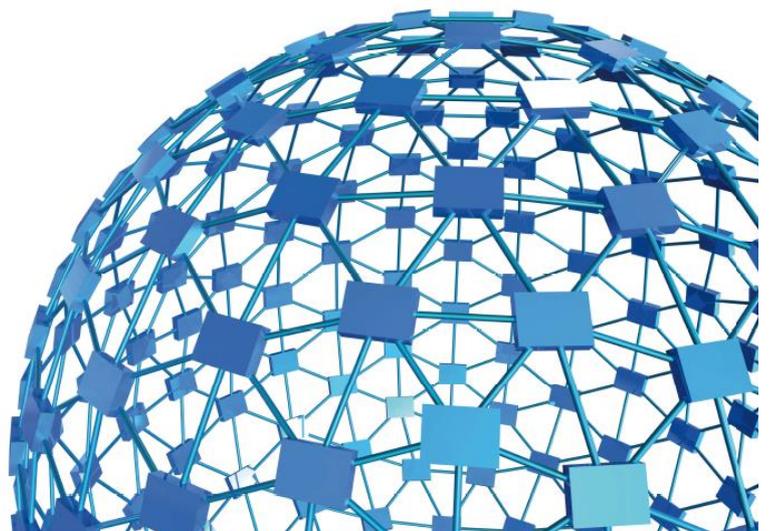
instruction manual

P.A.M.
Cat. No. 38500

Serial No.

SAFETY CONSIDERATIONS

ALTHOUGH THIS INSTRUMENT HAS BEEN DESIGNED WITH INTERNATIONAL SAFETY STANDARD, THIS MANUAL CONTAINS INFORMATION, CAUTIONS AND WARNINGS 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 INVOLVED.

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

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PAM

PRESSURE APPLICATION MEASUREMENT

Cat. No. 38500

General

The new P.A.M. (Pressure Application Measurement) from Ugo Basile is a novel, easy-to-use tool for measuring mechanical pain threshold in experimental **joint hypersensitivity models in rodents.**

The PAM device has been designed and validated specifically for the mechanical stimulation and assessment of **joint pain**, and therefore is especially useful in studying **arthritis**.

The PAM applies a quantifiable force for **direct stimulation of the joint** and automatic readout of the animal response.

The operator simply wears on his/her thumb a special force sensor, specially designed to apply force to **rat and mouse joints,** and measures the force which elicits the animal response (normally, limb withdrawal).

Each PAM device comes standard with two force sensors, a **large one** useful for stimulating rat joints, a **smaller sensor** recommended to test mice; an optional **paw transducer/applicator** is also available, to stimulate the animal paw.

Joint Pain

Arthritis

MECHANICAL PAIN THRESHOLD IN:

- Joint Hypersensitivity
- Chronic Joint Inflammation

Main Features

- Rat and Mouse Transducers included
- Maximum Applicable Force: 1500g
- Resolution: 0.1g

- Automatic recording of Limb Withdrawal
- User-controlled application of pressure directly to the joint
- DCA Software included - **NEW 2014 release**

Ugo Basile: more than 10,000 citations

ugo basile®
TRANSFORMING IDEAS INTO INSTRUMENTS

CLIENTE / CUSTOMER_____

Ordine No. / Order No. _____ Data / Date____/_____/_____

UB code	CAT.No.	√	38500	38550	DESCRIPTION	DESCRIZIONE
	38500-001		1	1	ELECTRONIC UNIT	UNITÀ ELETTRONICA
	38500-002		1		JOINT TRANSDUCER (large)	TRASDUTTORE (large)
	38500-003		1		JOINT TRANSDUCER (small)	TRASDUTTORE (small)
	38500-016			1	APPLICATOR WITH TRANSDUCER (for large animals)	APPLICATORE CON TRASDUTTORE (per animali grandi)
	38500-303		1	1	PEDAL SWITCH	PEDALE
	38450-330			1	FILAMENT WITH MAGNET (long)	FILAMENTO con MAGNETE (lungo)
	38550-320			1	FILAMENT WITH MAGNET (short)	FILAMENTO con MAGNETE (corto)
	38550-321			1	PERSPEX POINTED PUSHER	CONO PRESSIONE IN PERSPEX
52010-325			1	1	USB LEAD	CAVO USB
E-AU 059			1	1	UNIVERSAL POWER SUPPLY	ALIMENTATORE UNIVERSALE
E-AU 041 USB pen-drive	DCA		1	1	SOFTWARE	SOFTWARE
	38500-302				INSTRUCTION MANUAL	MANUALE D'ISTRUZIONE
OPTIONAL						
	38500-006				PAM PAW PRESSURE APPLICATOR	APPLICATORE ZAMPA

DATE / /	Serial No.	IMBALLATO DA / PACKED BY
Universal Input 85-264 VAC, 50-60Hz		

IMPORTANT/IMPORTANTE:

Check the shipment for completeness immediately after receipt: should you find any discrepancy, please fill in the following part and transmit it to our fax no. **+39 0332 745488**

Al ricevimento della merce controllate che la spedizione sia completa: in caso di discrepanza, completate il formulario di seguito riportato ed **inviatelo** al nostro fax no. **0332 745488**

FROM: Name	Company/Institution
DATE	REF.

NOTE

MOD.04 REV 0

C O N T E N T S

FIGURE INDEX

P.A.M. PRESSURE APPLICATION MEASUREMENT

Cat. 3 8 5 0 0

1   GENERAL

The new P.A.M. (Pressure Application Measurement) from Ugo Basile is a novel, easy-to-use tool for measuring mechanical pain threshold in experimental **joint hypersensitivity models and chronic joint inflammation in rodents.**

The PAM device has been designed and validated specifically for the mechanical stimulation and assessment of **joint pain**, and therefore is especially useful in studying **arthritis**.

The PAM applies a quantifiable force for **direct stimulation of the joint** and automatic readout of the animal response.

The operator simply wears on his/her thumb a special force sensor, specially designed to apply force to **rat and mouse joints**, and measures the force which elicits the animal response (normally, limb withdrawal).

Each PAM device comes standard with two force sensors, a large one useful for stimulating rat joints, a smaller sensor recommended to test mice; an optional **paw transducer/applicator is also available**, to stimulate the animal paw.

1.1   Rationale of the technique

Arthritis is associated with chronic, debilitating pain in the joints. Current metrics of arthritic pain in animal models are indirect, by scoring the level of motor activity or the animal weight distribution (Barton et al. 2007); while correlating well with the level of joint pain, their metric is a composite picture of complex pain responses, and provides little direct information about local stimulation and locally-evoked responses.

The quantification of localized joint hypersensitivity is not common in animal experiments; in this sense the PAM device represents a step forward toward multifactorial measurement of pain-related behavior in animal research; the **PAM** is the **first instrument designed specifically to apply force to the joint and automatically detect the animal response.**

1.2   Principle of Operation

The force transducer is mounted on the operator's thumb so that routine procedures may be employed to examine and test the joint concerned.

The completion of each test may be indicated by the operator either by the sudden release of the pressure on the joint.
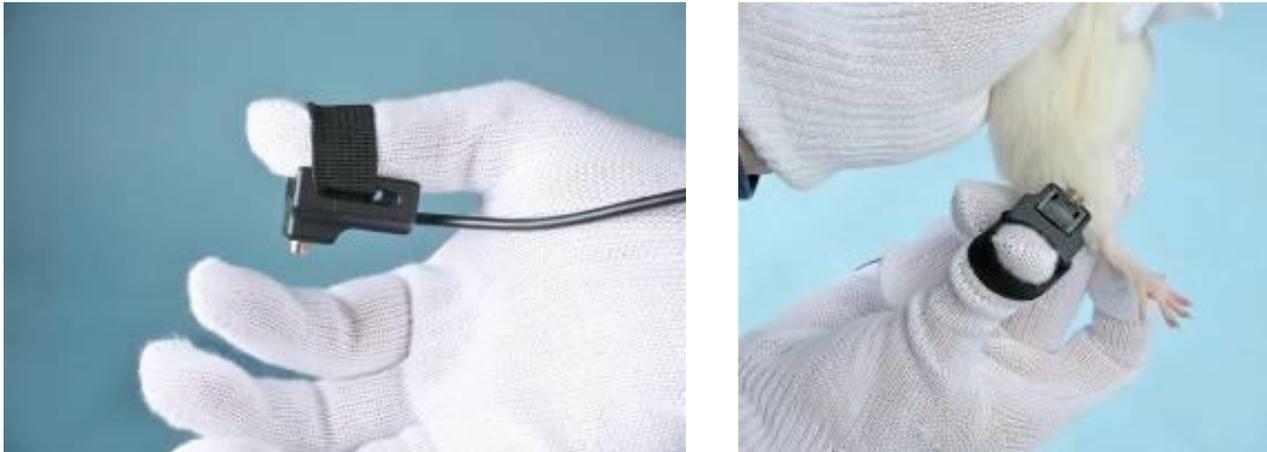
Figure 1: "Joint transducer" (small) mounted on the operator's thumb

The display then gives the operator a summary of the results of the test (*i.e.* force and time corresponding to the animal response) and the operator may choose to reject the results or to accept them, in which case they are recorded in PAM's internal memory.

The rate of application of the force is decided by the operator and the PAM includes software tools that help the operator in consistently applying the force at the desired rate.

The results of, typically, several thousand tests may be stored in PAM for transfer to a PC when convenient.

In addition to the thumb transducers, a hand-held force applicator is also available to measure pain threshold in paws and muscles (see also paragraph 2.3).

Figure 2: "Paw Transducer"

1.2.1   Consistent Results

The PAM device has been designed to make sensitivity experiments easy and consistent, thanks to its:

- Effective **peak detector**, for a reliable and automated detection of the animal response

- **Rate-Meter** (see paragraph 6.4) and Slope features (see paragraph 7.5), ensuring the desired force is applied at a consistent rate

- **Software**, acting as a quality control tool, by showing the applied force (red line) and the desired target force rate (blue line) in real time.

The rate of application of the force (target force rate) is set by the operator from the configuration menu (see section 5.6.1, option 2) and the PAM includes software tools that help the operator in consistently applying the force at the desired rate.

See the below screenshot of the software showing the force trace (**in blue**) and the desired target force rate (**in black**).

The experimenter can consistently apply the force at the desired rate, by simply making sure that the blue trace lays on the black line.

2   INSTRUMENT DESCRIPTION

Figure 3: "P.A.M. Standard Package"

The **38500** PAM device comes as a complete package including:

- an Electronic Unit **38500-001**

- a Large Joint Transducer **38500-002** (for rat)

- a Small Joint Transducer **38500-003** (for mouse)

a USB flash-drive with drivers, DCA software and the instrument's Operating Manual 38500-302The instrument is delivered in a convenient plastic case which should be retained for the safe storage of PAM and its accessories.

Beside the above listed items, the case also contains

- USB Cable **52010-325**

- an External trigger foot pedal **38500-303**

- Power adapter **E-AU 059**

See also paragraph 9-ORDERING INFORMATION.

The paw pressure transducer featured in picture 2 is an optional accessory, see paragraphs 2.3 and 9.1-Optional.

2.1 Electronic Unit

The electronic unit is lodged in a compact box. The three buttons below the display are **soft buttons** whose function changes at different stages of the PAM program.

The PAM is switched **ON** by pressing the **ON/OFF** button.

The lower line of the PAM display, 4 by 20 character, is normally used to indicate the current function of each button.

When the PAM is switched **ON**, the initial menu choice is between making measurements and configuring the instrument to suit the user's specific requirements.

The electronic unit is battery operated: its internal rechargeable battery is charged automatically when external power is supplied to its miniature USB 'B' socket, either from a PC or from a suitable USB power adapter.

A fully charged battery will operate PAM continually with its display at maximum brightness for approximately 16 hours.

When the PAM is switched **OFF**, it goes into a standby mode in which it takes almost no current from the battery although it still updates its internal calendar clock.

Figure 4 "Electronic Unit"

Each PAM transducer has a built-in identification chip, facilitating the PAM adjustment to match that of the transducer. Prior to any measurements, the PAM automatically balances out any offsets using its **Autozero** function.

A relevant feature of the PAM electronics is the **Ratemeter**, see paragraph 6.4, which provides a visual cue to make sure the desired force is applied at a consistent rate.

The electronics stores results of several hundred tests, which can be transfered to a PC when convenient.

The detailed menu options will be described later, see paragraph 5.4, 5.5 and 5.6.

2.2   Joint Transducers (Small and Large)

The Small and Large joint transducers have been designed specifically to apply force to the joints (for example to study animal models of arthritis) of mice and rats but they can be used also for other parts of the animal body, including muscles and paws, or for other joints, such as the ankle.

Each transducer contains a memory chip which identifies the type of transducer and its specific characteristics including all relevant factory calibration information. This feature allows PAM to adjust automatically to each transducer and ensures that transducers and PAM instruments may be interchanged without the need for routine re-calibration.

Figure 5 "Joint Transducers"

Figure 6 shows how to wear the transducer comfortably on the operator's thumb, and how to apply the force

Figure 6 "How to Use Transducers"

2.3 Paw Pressure Applicator

An optional paw transducer/applicator is also available, rapidly transforming the PAM into a Digital Randall-Selitto.

The force sensor is mounted into a hand-held device. The force sensor incorporates a cone-shaped applicator to stimulate the animal's paw with a "Randall-Selitto approach" (as shown is Figure 6).

The same principle can be applied to the animal's tail or muscles.

Figure 6 "Paw Transducer"

3 DATA ACQUISITION

The DCA **software** provided with the instrument (on the USB pen-drive) ensures automatic recording and storing of the animal response.

The software includes the Slope feature, to help the operator via a visual cue in consistently applying the force at the desired rate.

The results of several hundred tests may be stored in PAM for transfer to a PC when convenient, see paragraph 7.6-Downloading Data from PAM to PC.

4 INSTALLATION

4.1 Unpacpippoking & 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 8.2-Customer Support.

Protect the environment!

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

4.2 Notes on the Instruction Manual

The 38500 Instruction Manual included in the package (on the USB 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 page. For any additional information and/or assistance, you are welcome to contact our Service Department (see paragraph 8.2-Customer Support), specifying the serial number of your instrument.

4.3 Charging the Internal Battery

The instrument is dispatched from the factory with a fully charged battery.

However, it is recommended that the battery is charged before initial use by connecting it to a USB socket on a functioning PC or to a suitable power adapter, using the USB cable provided, for at least 3 hours.

4.4 Connections

Plug one the touch-stimulation transducer and the foot pedal into the connector located on the back panel of the PAM electronic unit.

4.5 Installing the DCA Software

ATTENTION:
Before installing the software, please read the INSTALLATION NOTES at the beginning of the manual.

The software provided with the PAM (on the USB pen drive) should be installed on a suitable Windows PC, running Windows Vista, 7, 8 or XP.

First, install the software, by double-clicking on the Start Ugo Basile.exe file; follow the instructions until installation is complete.

Then, connect the PAM via a USB port. The PAM will be automatically recognized and the drivers will install automatically, by simply following the instructions on the screen.

The Ugo Basile DCA Software can now be launched from the desktop icon or from the Program Files folder.

See section 7, for instructions on how to use the Ugo Basile DCA Software.

4.6   Intended Use

The PAM is intended for investigation use on laboratory animals only.

4.7   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

4.8   Additional Safety Consideration

a.   Use original accessories and spare parts only, see also paragraph 9-ORDERING INFORMATION.

b.   Do not operate the instrument in hazardous environments or outside pre-scribed environmental limitations (i.e. +18C°/+24C°, 60% relative humidity, non-condensing);

c.   Do not spray any liquid on the connectors;

d.   Keep inflammables far from the instruments.

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 in-struction manual supplied with the instrument;
- replacement of original components, accessories or parts with others not ap-proved by the manufacturer;
- servicing carried out by unauthorized personnel, see also paragraph 8-MAINTENANCE.

5   OPERATION

5.1   Consideration on the Animal Behavior

The PAM device is a simple and easy-to-use device. As usual in any behavioral meas-urement, care must be taken by the operator to guarantee a consistent performance of the test.

When using the PAM, we can envisage two main potential sources of experimental vari-ability: the first is the animal emotional state, which can be overcome by preliminary ha-bituation procedures and the second is the rate at which the operator applies the force.

The device incorporates several tools to help the operator consistently apply the desired amount and rate of force on the animal, the **ratemeter** on the electronic unit display, and the **slope function**, on the software, see also paragraph 1.2.1. These tools are described in the following paragraphs.

5.2   First Time Operation

After charging the PAM for at least 3 hours, plug in the touch-stimulation transducer and turn the instrument **ON** by pressing the **ON/OFF** button.

ON / OFF

Once the introduction screen is completed, the initial menu will appear as below.

```
         Main Menu
-----------------------------------
Measure              Configure
```
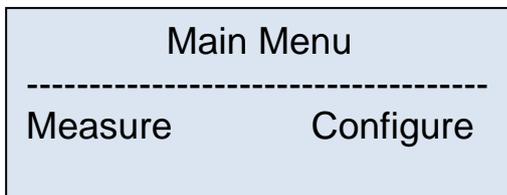
The lower line of the display indicates the current functions of the three **soft buttons** labelled F1, F2 and F3.

In this case, F1 is **Measure**, F2 is not being used and F3 is **Configure**.

Press **Measure**. A screen similar to the following will appear:

```
      PAM Transducer
     Large: 0->1500gf
     Calibrated: 091009
     EVF gain matched
```
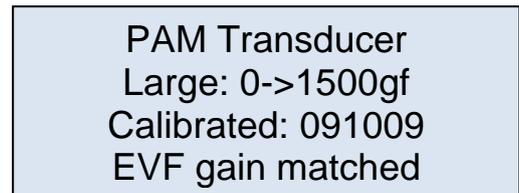
After a few seconds, the instrument completes an **Autozero** operation and then the display shows:
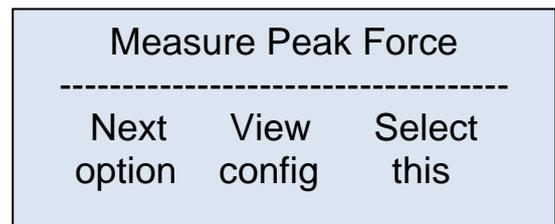
Note that, in this case, all three **soft buttons** are in use.

```
      Measure Peak Force
-----------------------------------
  Next      View     Select
 option     config     this
```

! **IMPORTANT NOTE**:

Do not apply force while the instrument is carrying out the autozero procedure.

Press **Select this**.

```
      Measure Peak Force
-----------------------------------
  Next      View     Select
 Option     config     this
```
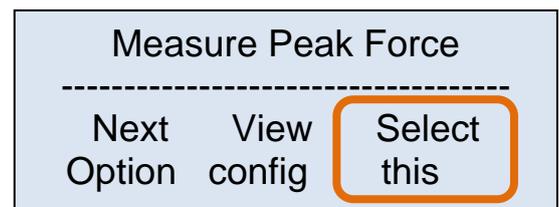
The following display will appear:

```
 23/07/09          14:23:35
         Test 1
   Exit            Test
```
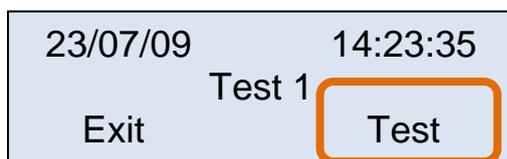
Check that no force is being applied to the connected transducer and then press **Test.**

PAM checks the transducer zero and then displays:
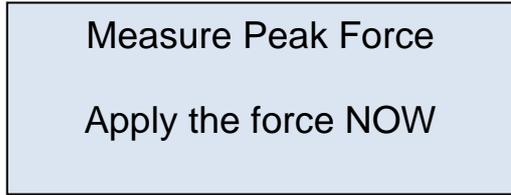
For as long as the force is applied steadily, the **Force** value will indicate the applied force in gf and the **Time remaining** will indicate the number of seconds left to complete the test.

> Measure Peak Force
>
> Apply the force NOW

When the transducer is released, the screen changes, and showing the peak force and the reaction time in seconds (over a total measurement time, in seconds).

> Test 1
> Peak Force 234.5gf
> at 3.55s of 4.30s
> Reject   Accept
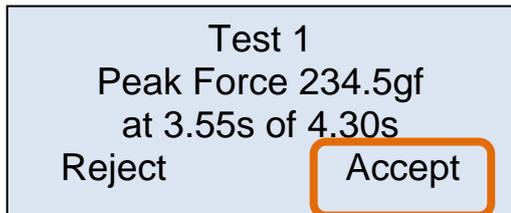
Press **Accept** and the screen changes to:

> 23/07/09   14:25:43
> Test 2
> Exit   Test
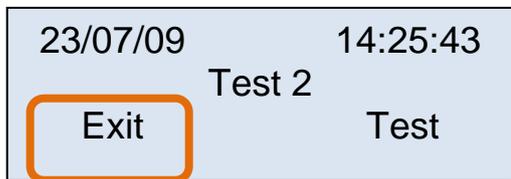
While pressing **Exit** the display returns to:

> Measure Peak Force
> ------------------------------------
> Next   View   Select
> Option   config   this
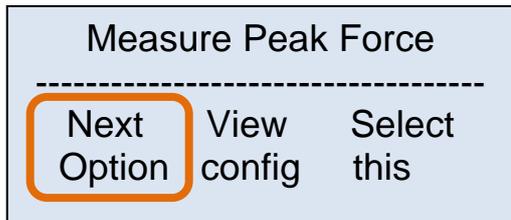
Press **Next option**, then press **Select this.**

> View Acquired Data
> ------------------------------------
> Next   Select
> option   this
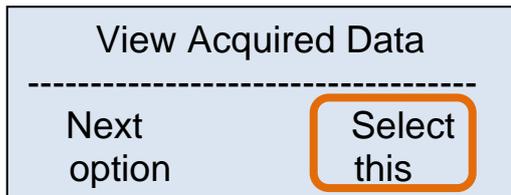
The stored data will be loaded and after 1-2 seconds the display shows:

> 23/07/09   14:23:35
> No 1   Auto
> 234.5gf at 3.55s
> Previous   Exit   Next

The most relevant information of each measurement (Date, Time, Progressive number of the measurement, Automatic or Foot Pedal measurement mode, Peak Force, Peak Time) are shown and the measurements can be browsed by simply pressing **Next**.

Press **Exit** to return to:

> Measure Peak Force
> ------------------------------------
> Next   View   Select
> option   config   this
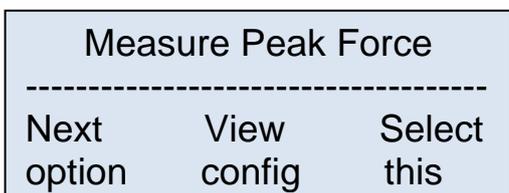
Press the **ON/OFF** button to switch OFF the PAM.

N O T E : If the PAM does not behave as described, please repeat all the steps.

If the problem persists, charge the internal battery and reset the PAM manually by pressing with a needle into the small reset hole located to the right of the display.

5.2.1   Automatic Dimming of the Display

The display backlight may be configured to dim automatically in most menus, if no soft button is pressed during a period preset between 2s and 29s.

By default this feature, which extends the battery life, is OFF and may be turned ON from the Configuration Menu.

5.3   Routine Operation of the Instrument

5.3.1   Turning ON the PAM

The PAM is turned **ON**, by pressing the **ON/OFF** button.

5.3.2   Menu Operation

PAM is operated via a number of menus by pressing the three **soft buttons**, F1, F2 and F3 under the display.

The current functions of F1, F2, F3 are indicated by the lower line/s on the display.

For example, the following is a typical display in the **Measure** menu:

```
      Measure Peak Force
--------------------------------------
Next         View        Select
option       config        this
```

The user steps through the available option (which appear on the upper line) by pressing "**Next option"** and then selects the desired option by pressing "**Select this"**.

5.3.3   Turning OFF the PAM

ON / OFF

The PAM may be turned **OFF** from most display screens, by pressing the **ON/OFF** button.

To preserve battery life, PAM switches **OFF** automatically, after a period of inactivity (if no soft button is pressed), which can be preset from **2 to 29** minutes or **never,** from the **Configure** menu; the factory default setting is **never**.

PAM also switches **OFF** automatically if the battery voltage is below a critical level.

5.4   Main Menu

The **Main Menu** is the first menu which appears when PAM is switched ON.

```
Main Menu
-------------------------------------
Measure          Configure
```
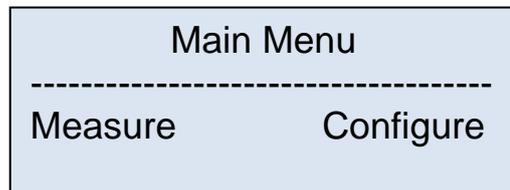
Via the "**Measure**" menu, see paragraph 5.5 it is possible to choose the options relating to measurements and transferring collected data are to the PC.

The "**Configure"** menu allows the user to adjust a number of parameters to suit particular measurement requirements, see paragraph 5.6.

5.5   Measure Menu

When Measure is selected from the Main Menu, a screen appears which is similar to the following, and depends on the force selected.

```
PAM Transducer
Large: 0->1500gf
Calibrated: 091009
EVF gain matched
```
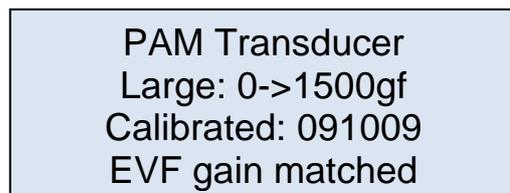
After a few seconds, PAM completes an **Autozero** procedure and the following screen is displayed:

```
Measure Peak Force
-------------------------------------
Next        View        Select
option      config        this
```
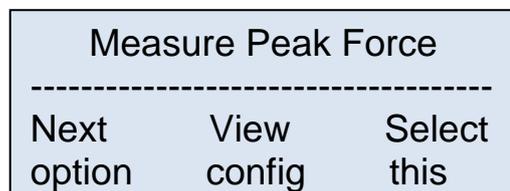
This is the initial screen of the **Measure** menu which consists of the following 6 options, which appear in a loop:

- Measure Peak Force
- View Acquired Data
- Transfer Data to PC
- Memory Status
- Erase Memory

Press **Next option** to go to the following option; press **View config** to view the configuration in use.

These options are discussed in the next sub-headings. The descriptions given refer to factory-set default values for the configuration settings. The effects of altering these settings will be discussed later.

5.5.1   *Measure Peak Force*

Pressing **Select this** at the **Measure Peak Force** screen gives a display similar to:

```
24/07/09              15:46:09
              Test 4

Exit                      Test
```
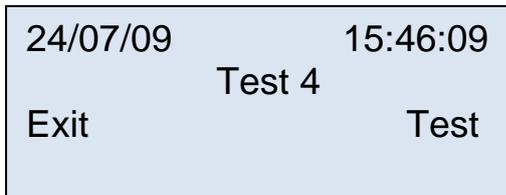
which shows the current date and time, the next test number (in the example the test will be the fourth to be stored since PAM's memory was last erased) and the options "Exit" and "Test".

When **Test** is pressed, a caption will appear, intimating that no force be applied to the transducer during the autozero procedure, after which the following display:

```
Measure Peak Force

Apply the force NOW
```
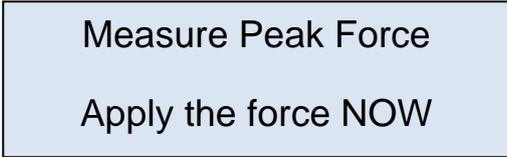
The user should now perform the measurement. As soon as the initial force threshold is reached, the display shows:

Force indicates the currently applied force in gf while **Time remaining** counts down in 0.05s steps from the Maximum measurement time, which may be adjusted up to 30s (default 15s).

```
Acquiring Force Data
Force = 123.4gf
Ends on force level
Time remaining 10.2s
```
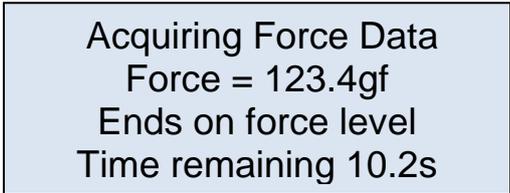
Normally, the test is terminated either by releasing the force on the transducer (default) or (optionally); in alternative, the animal response can be scored visually and by pressing the included foot pedal switch.

A visual score of the sensory or pain threshold can be necessary when there is no clear limb withdrawal, but rather a vocalization or a very small or complex animal movement

In either case, the display shows typically:

```
           Test 4
   Peak Force 293.6gf
     at 7.70s of 8.69s
Reject              Accept
```
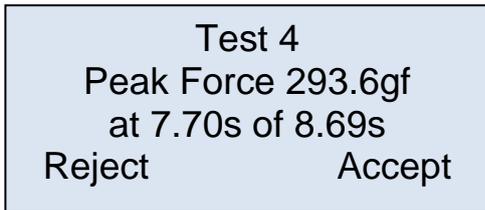
The first figure, 7.70s indicates the peak time (*i.e.* the latency time of the animal response), while the second figure, 8.69s, indicates the total duration of the test. The user must choose whether to **Reject** or **Accept** the results of the test, by pressing the corresponding soft-button.

If the result is rejected, it is possible to stop the measurements or repeat the previous test. If the result is accepted, the data are stored in the PAM memory and the user may stop the measurements or proceed with another test.

If the **Maximum measurement time** is exceeded during the test, the measurement is terminated automatically and the display shows:

Pressing **Begin again** effectively returns the user to the first Measure Peak Force screen, while pressing **Repeat test** allows the user to repeat the current test.

```
Measurement time
  was exceeded
Begin      Repeat
again        test
```

5.5.2   View Acquired Data

Select **View Acquired Data** from the **Measure** menu, followed by **Select this;** the total number of stored measurements will be displayed, and then the following screen will appear, with data related to the last measurement:

```
16/09/09          10:34:15
No 1                  Auto
      234.5gf at 3.55s
Previous     Exit      Next
```

Press **Previous** to view the test before the one being displayed. If the first measurement was displayed, as in this example, pressing **Previous** will display test 70, as the test numbers loop.

Press **Next** to view the next stored measurement, **Exit** to return to option 1 of the Measure menu (i.e. Measure Peak Force).

5.5.3   Transfer Data to PC

Via this option it is possible to transfer the data stored in PAM's memory to the PC, see paragraph 7.6-Downloading Data from PAM to PC for instruction.

The PAM must be connected to the PC via the USB cable supplied and that the PC is running the appropriate DCA software provided with the instrument.

If this option is selected in the absence of a PC, correctly connected and running appropriate software, PAM displays "**PC not available"** for a few seconds and then returns to the first option on the Measure menu.

For detailed software details and instruction, see paragraph 7.

5.5.4   Memory Status

Select the **Memory Status** option from the Measure menu to check the PAM data memory available to store further measurements; in the following example: 75.4%.

```
Memory Status
Available 75.4%
Tests left: >547
Back
```

The estimated value of **>547** for **Tests left** is based on the assumption that future measurements will be similar in length to those already stored.

The number of tests that can be stored, strongly depends on the duration of each test.

Press **Back** to return to the first option of the **Measure** menu.

5.5.5   Erase Memory

When selecting **Erase Memory** from the **Measure** menu, you will be given the option to cancel all the data stored in the memory, by pressing **Confirm**.

The configuration settings will not be changed.

```
Erase memory
All stored data
 will be lost
Cancel        Confirm
```
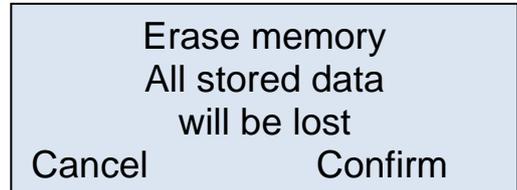
If you don't wish to cancel the data, press **Cancel** to return to the start of the **Measure** menu.

The **Erase Memory** option is offered automatically after transferring the data to the PC, see paragraph 7.6.

5.5.6   Back to Main Menu

To go back to the main menu, press **Exit.** When the Exit choice is not available, press Next Option, until Exit appears over the F2 soft-button.

5.6   Configure Menu

When **Configure** is selected from the **Main Menu**, the following screen appears, which allows the user, by pressing **Set config,** to adjust a **number of configuration settings in PAM**.

```
Configure Menu
------------------------------------
Set              Restore
config          defaults
```
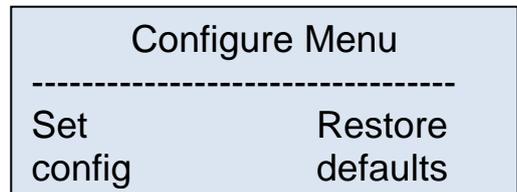
These include **options related to the measurement process**, such as whether the measurement is ended automatically by removing the applied force or manually by pressing the foot trigger switch, but also covers more **general settings**, such as the brightness of the display and the period of inactivity before PAM will switches off.

An example:

by pressing **Set Config**, the first configuration option appears, from which the user chooses between ending the measurement automatically or by pressing the Foot Pedal.

The underscoring under **Force lvl** indicates that the **end on force level** option (factory default) is selected.

```
Configuration Menu
Force lvl        Foot pedal
-----------
Next            Change
```
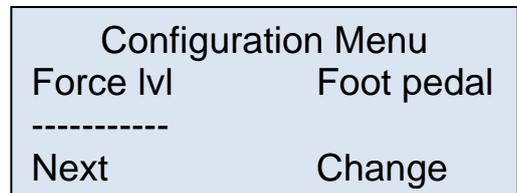
Pressing **Change** moves the selection to **Foot pedal**. There is no need to confirm the choice; press **Next** to see the next configuration option.

```
Configuration Menu
Force lvl        Foot pedal
                 -----------
Next            Change
```
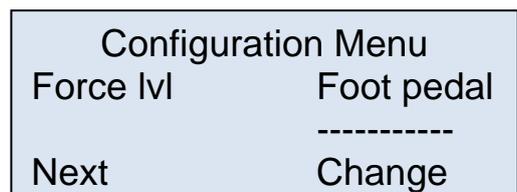
5.6.1   Configuration Options

The **Configure** menu shows 17 screens in a loop, by which the user may adjust the following features of PAM in order to:

1) Adjust the target force rate **(120gf/s),** see also paragraph 6.5

2) Use Ratemeter or Text (**Text**), see also paragraph 6.4

3) Set how each measurement is ended, automatically when reaching the force level, or by pedal switch (**force lvl**)

4) Set a warning sound to inform if a preset force is exceeded (**none**)

5) Adjust the warning sound threshold value (**300gf**)

6) Adjust the force value which triggers the start of the measurement (**2% of full scale**)

7) Adjust the maximum time allowed for a measurement (**15s**)

8) Set if the operator should be asked Left or Right paw at each test (**Do not**)

9) Use Subject Numbers or only Test numbers (**only Test**)

10) Store every measurement regardless or only the good measurements, *i.e.* accepted by the user (**only good)**

11) Store the force waveform or only the peak value (**waveform**)

12) Show abbreviated configuration settings on the Measure menu (**Do not**)

13) Show the offset value before each measurement (**Do not**)

14) Adjust the period of inactivity after which PAM switches OFF, from 2 to 29 minutes, or never (**never**), see also paragraph 5.3.3

15) Adjust the normal (undimmed) display brightness (**10**)

16) Adjust the period of inactivity after which the display dims, from 2s to 29s, or never (**never**) see also paragraph5.2.1

17) Set current date and time: this can be set by the PAM control unit or by the PC, see also paragraph 6.3.

The values in the brackets are the factory default settings. See also 6.5-Available Configuration Settings.

Press **Exit** to return to the Main Menu**: you will be asked to save Changes to Configuration**.

5.6.2   *Restoring Default Values*

From the Configure Menu, it is possible to restore the factory default values; press **Restore defaults** and the following screen will be displayed:
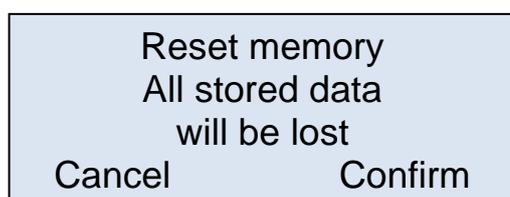
Reset memory All stored data will be lost Cancel Confirm	Pressing **Cancel** returns to the **Main Menu** without altering the configuration settings nor the data memory.
	Press **Confirm** to restore the factory-set configuration values as indicated in paragraph 5.6.1.

Remember that this action will also clear the data memory!

6 OTHER IMPORTANT FEATURES OF THE PAM

6.1 Battery Voltage Warning

The voltage of the internal rechargeable battery used in the PAM is monitored continuously during routine operation. Two warning levels are provided.

If the battery is low but still usable, the dashed line of most menus will include **Battery Low**. For example, the first **Measure** screen will become:

The PAM may still be used, but the USB lead should be connected to an active PC or a USB power adapter to charge the battery <u>as soon as possible</u>.

```
Measure  Peak  Force
  ----Battery Low-----
Next     View    Select
option   config   this
```

If the battery is so low that PAM cannot operate, then the instrument will switch OFF immediately. The data which have been stored will be retained but PAM cannot be used until it is connected to an active PC or a USB power adapter using the USB lead provided.

Clearly, it is not advisable to allow the battery to drop to this critical level.

6.2 Memory Limit Warning

Each time the user decides to make a new measurement, the PAM checks that sufficient data memory is available.

If the available memory is too low, the request is refused and the user is advised to transfer the stored data to a PC and/or to erase the existing data; in practice, this critical situation should never arise as **the user is given a clear warning each time a measurement is made** if the available space is not sufficient for 10 more measurements.

The PAM memory stores more than 1700 tests.

6.3 Calendar Clock

PAM has an internal calendar clock which maintains real time even while PAM is switched off. To set the PAM clock, go to the Configure menu, see prg. 5.6.1, item 17).

When the supplied PC software is used to transfer data or to monitor the force applied to the transducer, the PAM internal clock is synchronised automatically to the PC clock, see also paragraph 7-USING THE PC SOFTWARE FOR PAM.

6.4   Applied Force Ratemeter

The Ratemeter is a useful tool, which helps the operator in consistently applying the force at the desired constantly-increasing rate.

If the **Ratemeter** option is selected from the Configure menu (see paragraph 5.6.1, item 2), during each measurement, the PAM's display will appear as follows:
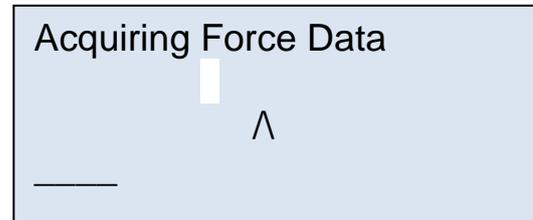
The **white rectangle** on the second line moves left or right of the **centre marker**, when the applied

```
┌─────────────────────────────┐
│ Acquiring Force Data        │
│              ▮              │
│                  ∧          │
│                             │
│ ────                        │
└─────────────────────────────┘
```

force is respectively below or above that required to maintain the selected force rate. In the above example, the applied force is below the ideal level for the specified target force rate.

The software Slope feature (see Slope Function) can be used in alternative, with the same purpose.

The fourth line of the display is a **time progress bar** indicating how much of the available measurement time has been used.

6.5   Available Configuration Settings

A complete list of configuration options is shown below including the adjustment ranges and default values, see also paragraph 5.6.1-Configuration Options.

Configuration flags which may be set or cleared.

- Whether each measurement is ended automatically on the **force level** (default) or by pressing the **foot pedal.**

- Whether **every measurement** is stored regardless (sometimes required by GLP) or the user may **choose which measurements** to store (default).

- Whether **the transducer offset value is shown** before each measurement or **not** (default).

- Whether **a warning sounds** if a preset **force is exceeded during** a measurement or **not** (default).

- Whether **abbreviated configuration settings** are shown on the Measure menu or **not** (default).

- Whether the stored data includes the **force waveform** (default) or **not**.

- Whether the operator should be asked to **indicate Left or Right limb** or **not** (default).

- Whether PAM should use **Subject Numbers** or **not** (default).

- Whether PAM should use the applied **force ratemeter** during tests or display live **force and time in text** (default).

Configuration values which may be adjusted:

- The **force value** (as a percentage of the full scale range) **which triggers the start** of the measurement. It may be adjusted from **0.4% to 5.0% in 0.1% steps** with a default value of 2%.

- The **threshold force** value in gf **which triggers the warning,** if enabled. It may be adjusted from **100gf to 1000gf in 10gf steps** with **a default value of 300gf.**

- The **maximum time** allowed for a measurement. It may be adjusted from **10s to 30s in 1s steps** with **a default value of 15s.**

- The **period of inactivity** after which **PAM switches OFF**. It may be adjusted from **1min to 29min in 1min steps** or **never** with a default value of never.

- The normal (undimmed) **display brightness**. May be adjusted from **1 to 10** with a default value of 10.

- The **period of inactivity** after which the **display dims**. May be adjusted from **1s to 29s in steps of 1s** or **never** with a default value of never.

- The **target force rate** which is the ideal rate at which the stimulus force should be applied, see also paragraph 5.6.1-Configuration Options

- The **date and time settings** of PAM's internal clock may be adjusted manually. The internal clock is normally set by synchronising it with the PC to which it is attached. Synchronising occurs when data is downloaded from PAM to the PC and when force measurements are monitored live on the PC.

- **Return to main menu**. This **menu** option acquires the latest configuration information and returns to the initial menu.

6.6   Updating the PAM firmware

The hardware used in PAM is very flexible and many of PAM features are determined by its internal software program or firmware. When improvements and additions are made to the firmware, existing users may update easily to the new version. The associated PC software includes an option to update the PAM firmware.

6.7   Skipping the introduction

The routine introduction shown when PAM is turned ON may be skipped by holding down the left soft button.

7   USING THE PC SOFTWARE FOR PAM

This section assumes that the **DCA PC** software has been installed successfully as described in paragraph 4.5. The DCA software manages the GSM, the PAM, the Analgesy-Meter DAQ and the eVF instruments by Ugo Basile.

7.1　　Note on the DCA PC Software

The DCA software is a guide: specifically, this software is a **monitoring tool** and it is not intended to drive the device (the software does not substitute for the controller); all operations must therefore be driven from the PAM control unit.

This includes changing the slope of the PAM target rate displayed on the PC monitor, which is in fact done from control unit. In fact the slope function on the PC reflects the Target force rate set in the configuration menu, see paragraph 5.6.1-Configuration Options.

7.2　　Starting the DCA Software

First connect the PAM device to the PC using the supplied USB cord, then run the **Ugo Basile DCA (Data Collection Application) Software** program from the Windows Start Menu or from the DCA icon on the desktop.

In the screen which appears, the Info Panel informs "**Device found**":

To proceed with the experiment press the "**Use Device**" button.

Figure 7 "DCA Software"

Wait until the info Panel confirms "**device connected**".
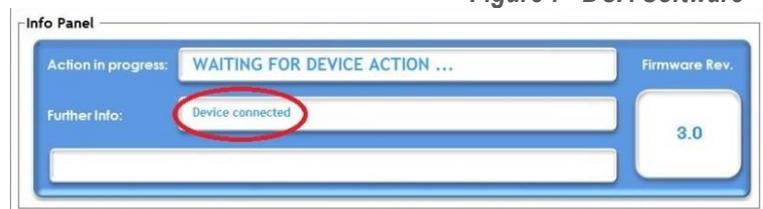
7.3   Starting the Experiment

Now that the software is ready to work, the test-monitoring window will appear:

Figure 8 "Test Monitoring"'

The test-monitoring window includes the following sections:

7.3.1   Main Box

As soon as the experiment is started from the device, the main box on the left will continuously plot the applied force intensity along time.

Note that the applied force appears as a **blue line**, overlapping a straight black line: the latter indicates the desired target force rate, as set via in the instrument configuration menu (slope feature)
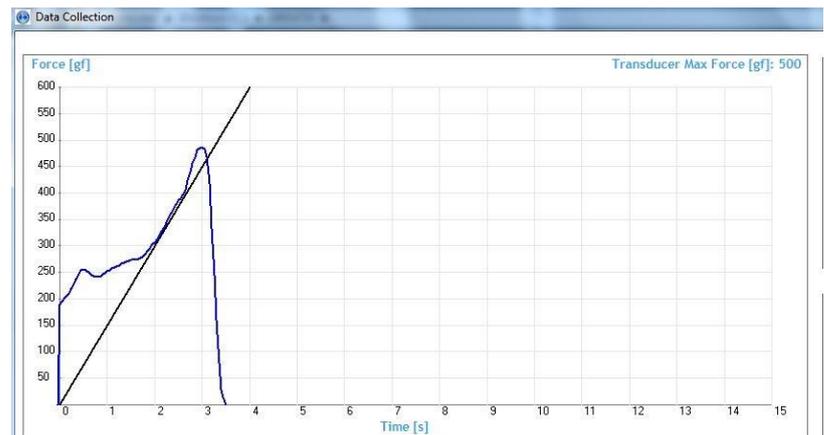
See paragraphs 1.2.1-Consistent Results and 7.5-Slope Function.

This function helps the experiment to apply the force consistently, by simply making sure that the blue trace lays on the black line.

At the beginning of each new trial, the plot window refreshes so that just the current and latest one is visualized.

A caption on the top right of the window reminds the Transducer Max Force, as set in the instrument configuration menu.

7.3.2   Info Panel

The Info Panel indicates the action in progress (in these examples TEST MONITORING, and WAITING) and additional information regarding the action

In addition, the panel shows the current Firmware revision.

During the data Analysis, the panel shows specific data related to the selected file
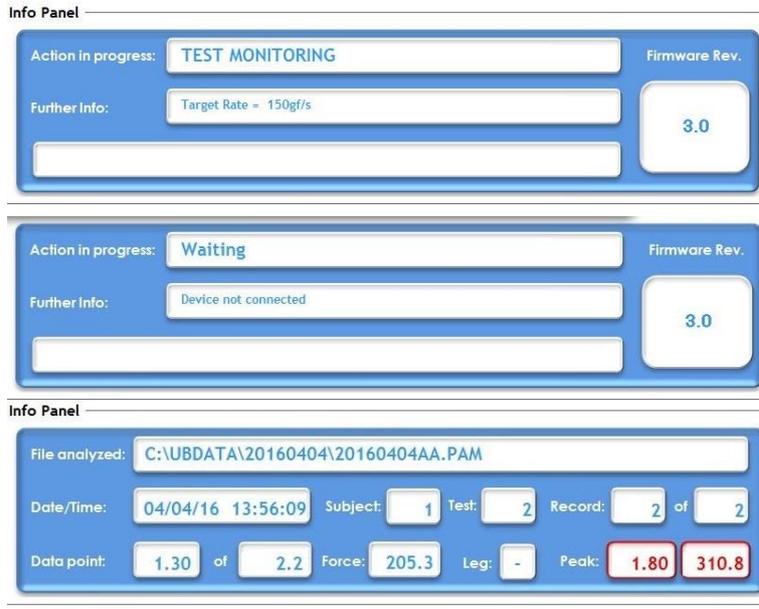
7.3.3   Device Control Panel

From the Control Panel it is possible to start the software (USE DEVICE) or to disconnect it (DISCONNECT).

The current value and the overall peak are displayed in the related boxes, together with the time.

7.3.4   Analysis Control Panel and Notes Tool

All data files are stored in the folder **C:\UB Data** in sub-folders which are named according to the date of data transfer, in the format **YYMMDD**; for example, data transferred on July 23rd, 2013 will be found in the folder **C:\UB Data\130723**. All the trials conducted in the same experiment are saved in the same file.

Individual files, containing the downloaded data, are named automatically according to the date, followed by two letters which progress from **AA, AB, AC**… through to **ZZ**.

For example, the third set of data downloaded on July 23rd, 2013 would be stored in the file:

C:\UB Data13\090723AC.PAM

where "PAM" is the extension of this proprietary software.

During the test, the Analysis panel is not active

Once the experiment is over and the data transmitted from the device to the computer, push the button "**Analyze**" to open the folder where the .PAM files are saved and stored during the test.

Select the file from the window which automatically appears:
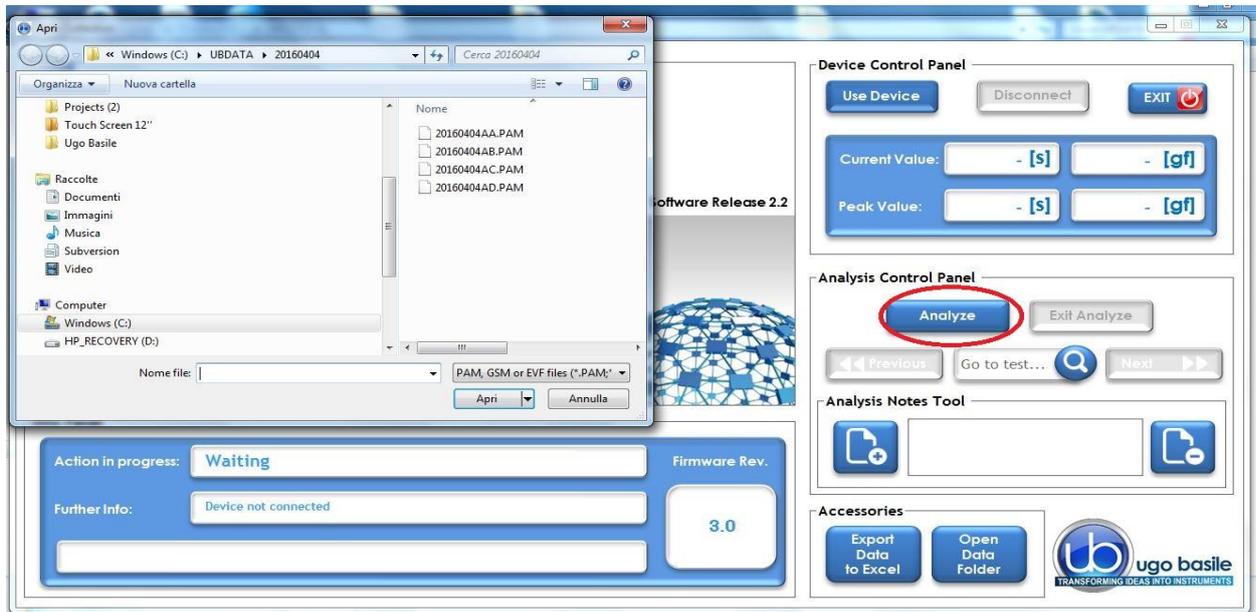
Figure 9 "Analyze Window"

When opening the selected file, all the plots referring to the trials contained in the related experiment will be shown:
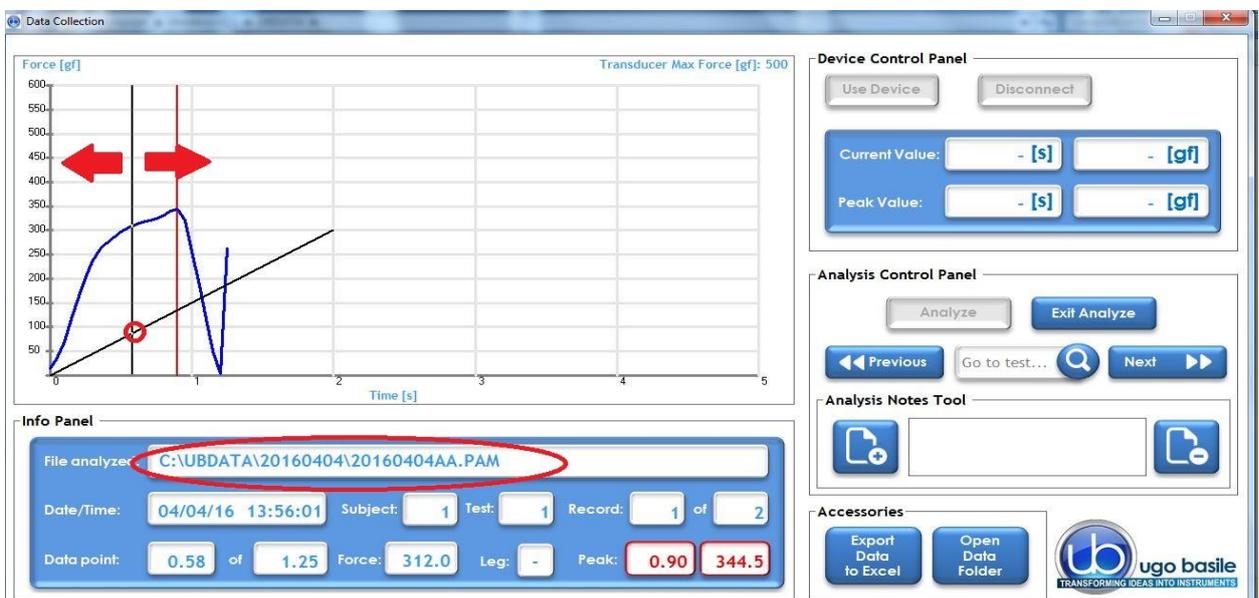
Figure 10 "Test Monitoring"

By moving the **black vertical line** (indicated by the arrows in the above screenshot) left or right along the slope, the user will be able to select a specific moment in the test.

To save it, select the "**Add Note**" button, left on the "Analysis Notes Tool".

The note is remove by the button on the right.
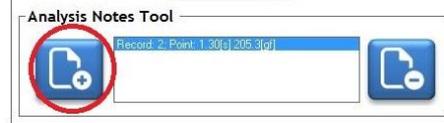
Note that the Info Panel now shows the data relating to the point currently selected.

Moreover, the recorded peak measure is pointed out, associated with time.

7.3.5   *Accessories*

In this section you will find the Export Data to Excel and the Open Data Folder buttons.

7.4   **Exporting and saving data**

Once the analysis of the experiment is accomplished, it is possible to export the data to Excel or to open a data folder.
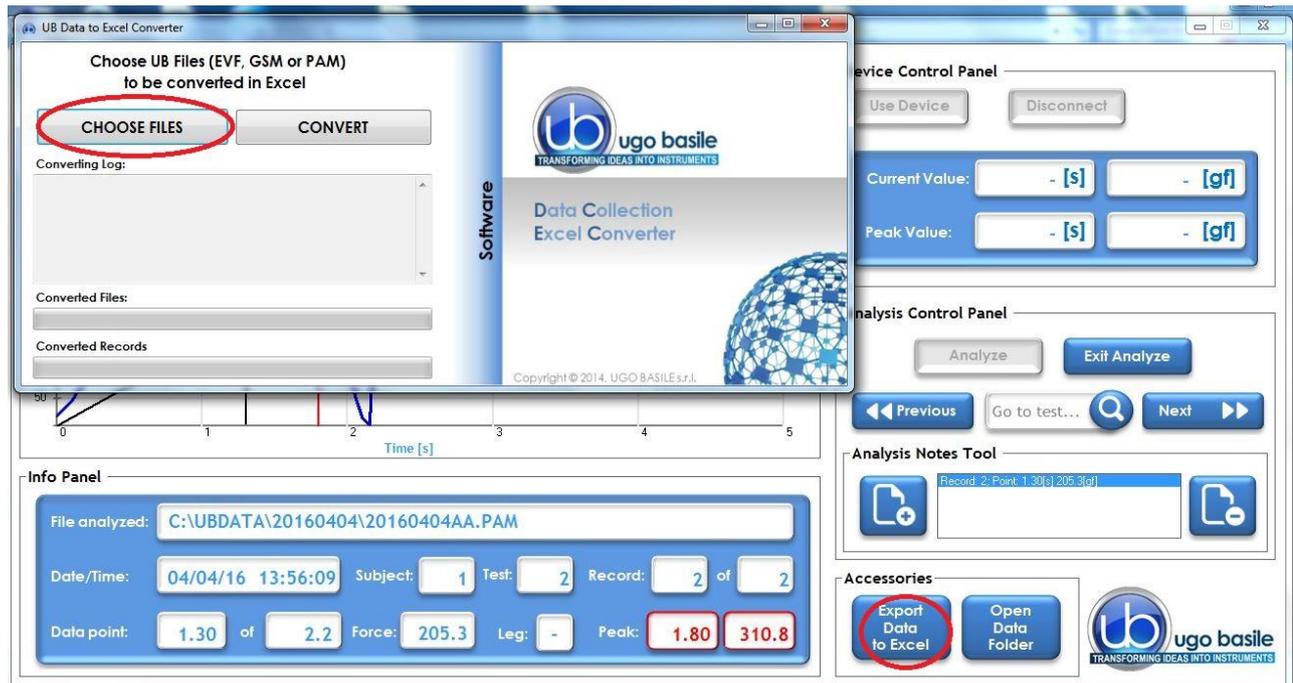
While the DCA PC software is not designed for statistical analysis, exporting the data using the DCA PC software creates a user friendly file compatible with Excel. One will find this automatically generated file to be most useful, because Excel seems to be the most common spreadsheet program which allows a researcher to perform whatever analytical statistics are preferred.

The data in the Excel file consists of columns for measurement number, peak force and peak position, filled with the values for each of the test downloaded.

When all the operations and experiments are completed, it is recommended to click "Disconnect" from the "**Device Control Panel**", in order to close the connection between the device and the software.
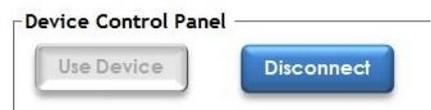
7.5 Slope Function

By the "Slope" function, the experimenter can consistently apply the force (i.er. pull the animal) at the desired rate, by simply making sure that the blue trace lays on the black line, see paragraphs 1.2.1 and 7.3.1.

The graphic screen is a visual guide during the experiments; the applied force is displayed as a blue line, while the straight black line (slope) shows the target force rate set from the device configuration menu, see paragraph 5.6.1, option 1.

The target force is also indicated in the Info Panel, 150gf/s in this example.

The slope represents what it looks like when the force is applied in a linear increasing rate; one should think of the slope as the **ideal**, the "target" that the user should try to match as s/he is applying the force. Matching the two lines will assist an operator in applying the force smoothly and steadily; this is what we call "**slope function**".

With practice, a researcher can become very skilled at steadily applying force, at a steadily increasing rate of application of the force, and really improve results.

The waveform is displayed until the user Accepts or Rejects the test, by using the buttons on the PAM control unit.

7.5.1 Changing the Slope

Remember, the PAM controller is the brain of the instrument and the software is only reporting information in the brain; the DCA PC software displays information provided by the PAM instrument. As the slope is the visual representation of the rate of application of force, to change the slope of the blue line in the software, one must change the rate of application of force.

This rate is stored in the PAM controller, so one must return to the PAM controller, toggle through the selected menus to reach the proper instructions. When the rate of application of force is changed in the internal memory of the PAM controller, it will automatically change the slope of the blue line quiiwhich is displayed in the software.

7.6   Downloading Data from PAM to PC

When working with the PAM not connected to the PC, all the data are stored inside the device memory. Data can be saved on the PC; data download is also controlled by the PAM device.

First, ensure that the controller is properly connected to the PC, and that the software is working properly.

Then, using the menu and buttons on the PAM controller, toggle through selections (**Main Menu > Measure > Next Option**) until the Transfer Data to PC appears on the blue display window of the PAM controller.

```
Transfer Data to PC
---------------------
Next            Select
Option    Exit    this
```
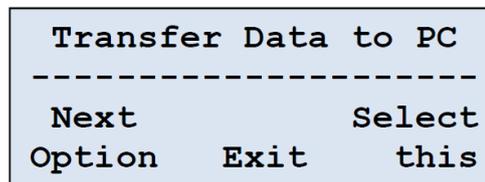
A progress bar in the Info Panel of the DCA software monitors the download: the same panel will confirm when the data download is complete.
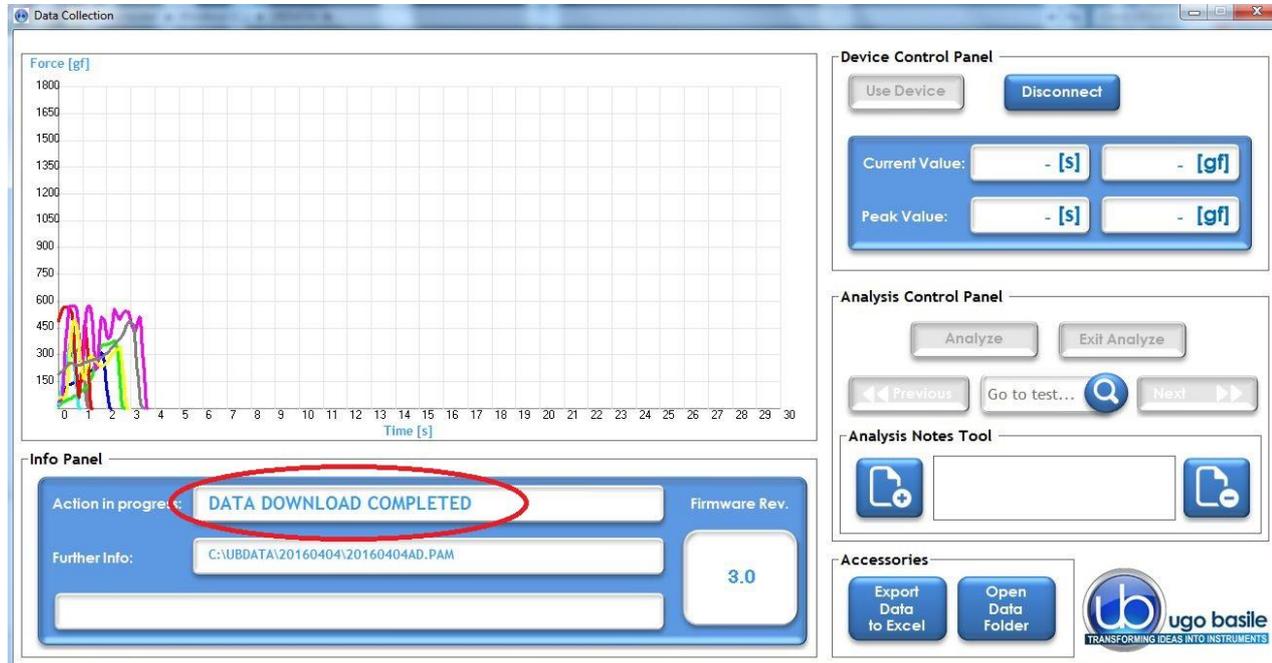
Data can now be analyzed as described in paragraph 7.3.4

The **Transfer Data to PC** operation, also ensures that the PAM clock and the PC clock are synchronized.

If this is performed when the PAM has no stored data, it will only synchronize the PAM and PC clocks.

8 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 section of the instruction manuals describes normal maintenance procedures which can be carried out at your facility.

8.1 Long Inactivity

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

8.2 Customer Support

For any further information you may desire concerning the use and/or maintenance of the PAM, 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 :

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Via G. Di Vittorio 2
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Phone : +39 0332 744574

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

Before sending any instrument to our factory for repair, 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.

9 ORDERING INFORMATION

38500	PAM Pressure Application Measurement, consisting of:
38500-001	Electronic Unit
38500-002	Large Joint Transducer (Rat)
38500-003	Small Joint Transducer (Mouse)
38500-303	External trigger switch (foot pedal)
52010-325	USB Lead
E-AU 059	Universal Power Supply

E-AU 041	USB pen-drive, including	
	38500-302	Instruction Manual
	DCA	Software

9.1 Optional

38500-006 Paw Transducer

9.2 Special High-Pressure Model for Large Animals

To assess mechanical sensitivity in larger animals, a modified version of PAM, the 38550, was designed, consisting of a handheld pressure applicator, to meet the experimental needs of manoeuvrability and resistance. The 38550 includes three different mechanical stimulators:

38550	**PAM, special high-pressure model for large animals, consisting of:**
38500-001	Electronic Unit
38500-016	Applicator with Transducer
38450-330	Filament with Magnet (long)
38550-320	Filament with Magnet (short)
38550-321	Perspex Pointed-Pusher
38500-303	External trigger switch (foot pedal)
52010-325	USB Lead
E-AU 059	Universal Power Supply
E-AU 041	USB pen-drive, including

| | **38500-302** | Instruction Manual |
| | **DCA** | Software |

| *38550* | PAM, special high-pressure model for large animals |

10 Instrument Specifications

Operation	
Commands	via soft-buttons
Read-out	multifunction graphic display
Starting	via keys on the Controller front panel
Force Range	from 1 to 1500 grams (maximum force can be preset)
Force increasing rate	monitored via the Ratemeter on the electronic unit, or via the slope function on the PC
Latency Time	read-out on the graphic display, in 0.1 s steps

Connection to PC	via USB cable (A to mini-B)
Power Requirement	either battery operated, or via USB cable connected to PC or universal converter (85-264 VAC, 50-60Hz)
Operating Temperature	10° to 40° C
Sound Level	negligible
Pollution Degree	≤ 2
Physical	
Total Weight	0.8Kg
Shipping Weight	2.7Kg approx.
Packing Dimensions	46x38x27cm
Warranty	

38500 is covered by a 24-month warranty

11   BIBLIOGRAPHY

12   Method Paper

- N. J. Barton *et alia:* "**A novel behavioural technique for measuring hyper-sensitivity in a rat model of joint pain"** Journal of Neuroscience Methods 163*: 67-75,* **2007**

- J. Leuchtweis et alia: "**Validation of the Digital Pressure Application Measurement (PAM) Device for Detection of Primary Mechanical Hyperalgesia in Rat and Mouse Antigen-Induced Knee Joint Arthritis...**" Methods & Findings in Exp. & Clinical Pharmacol., 32(8): 581-589, *2010*

13   Other Papers Mentioning UB PAM

- B.Y. Cooper et alia: "**Exposure to Gulf War Illness Chemicals Induces Functional Muscarinic Receptor Maladaptations in Muscle Nociceptors**" NeuroToxicology 54: 99-110, *2016*

- A. David-Pereira et alia: "**Metabotropic Glutamate 5 Receptor in the Infralimbic Cortex Contributes to Descending Pain Facilitation in Healthy and Arthritic Animals**" Neuroscience 312:108-119, *2016*

- T.J. Nutter et alia: "**A Delayed Chronic Pain Like Condition with Decreased K_V Channel Activity in a Rat Model of Gulf War Illness Pain Syndrome**" NeuroToxicology 51: 67-69, *2015*

- D. Amorim et alia: "**Amitriptyline reverses hyperalgesia and improves associated mood-like disorders in a model of experimental monoarthritis**" Behav. Brain Res 265: 12-21, *2014*

- S. Mohammadi et alia: "**α9-Nicotinic Acetylcholine Receptors Contribute to the Maintenance of Chronic Mechanical Hyperalgesia, but not Thermal or Mechanical Allodynia**" Molecular Pain 10(64): 1-9, *2014*

- B. Cooper: "**Synergistic Actions of Pyridostigmine Bromide and Insecticides on Muscle and Vascular Nociceptors**" DTIC Document, Jan 2014

- T. Schwagarus et alia: "**A New Method for Measuring CFA-induced Mechanical Hyperalgesia in the Rat**" Evotec *2012*

- **38550:** P.M. Raundal et alia: "**Pre-Test Habituation Improves the Reliability of a Handheld Test of Mechanical Nociceptive Threshold in Dairy Cows**" Res. In Vet. Science 102: 189-195, *2015*

- **38550**: P. Di Giminiani et alia: "**Capsaicin-induced Neurogenic Inflammation in Pig Skin: A Behavioural Study**" Res. In Vet Science 96(3): 447-453, **2014**

- **38550**: P. Di Giminiani et alia: "**Nociceptive responses to thermal and mechanical stimulations in awake pigs**" Eur. J. Pain 17(5): 638-648, **2013**

Visit our web page to check the latest papers mentioning Ugo Basile's devices!

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Notes

Notes

Notes

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.	**PRESSURE APPLICATION MEASUREMENT**
Catalog number	**38500**

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

- *2004/108/CE relating to electromagnetic compatibility*

- *2011/65/UE on the restriction of the use of certain hazardous substances in electrical and electronic equipment*

Account *Manager*

Adriano Basile
Nome / *Name*

April 2014

Date

Firma / *Signature*