PlantPen/N-Pen N 110

Manual and User Guide

Please read this manual before operating this product

Photon Systems Instruments

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The contents of this manual have been verified to correspond to the specifications of the device. However, deviations cannot be ruled out. Therefore, a complete correspondence between the manual and the real device cannot be guaranteed. The information in this manual is regularly checked, and corrections may be made in subsequent versions.

The visualizations shown in this manual are only illustrative.

This manual is an integral part of the purchase and delivery of equipment and its accessories and both Parties must abide by it.

TABLE OF CONTENTS

1 INFORMATION BEFORE USING N-PEN DEVICE

Read this manual carefully before operating the device. If you are not sure about anything in the manual, contact the manufacturer for clarification.

	By accepting the device, the customer agrees to follow the instructions in this guide.

Always follow corresponding manuals while working with the N-Pen device or doing the maintenance.

It is forbidden to interfere with the hardware or software of the N-Pen device in any way without previous agreement with the manufacturer.

The following table presents basic highlight symbols used in this manual:

Symbol	Description
	Important information, read carefully.
	Complementary and additional information.

Tab. 1 Used symbols.

2 TECHNICAL SPECIFICATION

Protocols	
Normalized Difference Greenness Index	NDGI = (R780 – R560)/(R780 + R560)
Nitrogen content	- Correlated with NDGI - Measured in percentage - Calibration for maize, wheat and barley
LED lighting	
Dual wavelength light source	565 nm and 760 nm
Detector	
Type	PIN photodiode with bandpass filters
Wavelength range	From 500 to 800 nm
Data storage and transfer	
Internal memory capacity	Up to 16 Mb
Internal data logging	Up to 100,000 measurements
Data transfer	USB cable Bluetooth (transfer up to 3Mbps for distance up to 20m)
PC software	FluorPen 1.1 (Windows 7 and higher)
Battery	
Type	Li-Ion rechargeable battery
Capacity	2000 mAh
Max. charging current	0.5 A
Charging	Via USB port - PC, power bank, USB charger, etc.
Battery life	70 hours typical with full operation Low battery indicator
Other	
Sample holder	Mechanical leaf-clip
Display	Graphical display
Keypad	Sealed, 2-key tactile response Turns off after 5 minutes of no use
Built in GPS module	Ultra-high sensitivity down to -165dBm High accuracy of <1.5 m in 50% of trials
Size	135 x 65 x 33 mm
Weight	188 g
Operating conditions	Temperature: 0 to +55 °C Relative humidity: 0 to 95 % (non-condensing)
Storage conditions	Temperature: -10 to +60 °C Relative humidity: 0 to 95 % (non-condensing)
Warranty	1 year parts and labor

3 GENERAL INFORMATION

N-Pen N 110 is a light-weight, battery-powered reflectance-based instrument that provides a convenient, cost-effective method for effective nitrogen management in plants throughout their growing season. Essentially, the N-Pen characterizes nitrogen amount by means of reflectance and by the concept of a close link between chlorophyll content and nitrogen content in plants.

Rugged and compact N-Pen can be used for plant biology research or for education. Due to an inbuilt GPS module and splash-proof cover is N-Pen suitable for field experiments. The measurement is non-destructive and permits quick, repeated monitoring throughout the growing season. The device is equipped with rechargeable Li-ion battery.

Measured data are sequentially stored in the internal N-Pen memory. Data transfer is via USB or Bluetooth communication. Comprehensive FluorPen 1.1 software provides data transfer routines and many additional features for data presentation in tables and graphs.

Fig. 1 Device description.

4 LIST OF EQUIPMENT AND CUSTOMER INFORMATION

Standard version of the N-Pen device package consists:

- **N-Pen N 110**
- **Carrying Case**
- **FluorPen software and driver** (on a USB flash disc)
- **Operation Manual** (PDF on a USB flash disc)
- **USB Cable**
- **Other Accessories or Optional Features** (according to your specific order)

	For USB connection you need to have the USB driver installed in your PC. You find the driver on the installation disk (USB driver folder). If any item is missing, please, contact the manufacturer. Also check the carton for any visible external damage. If you find any damage, notify the carrier and the manufacturer immediately. The carton and all packing materials should be retained for inspection by the carrier or insurer. For customer support, please write to: **support@psi.cz**

5 CARE AND MAINTENANCE

N-Pen N 110

- Never submerge the device in water!
- Keep the optical part clean and dry! Inspect visually the optical window and the white pad on the inner side of the leaf clip after each deployment. Use soft, non-abrasive tissue for cleaning.
- The device should not come in contact with any organic solvents, strong acids or bases.

6 PRINCIPLE OF MEASUREMENT

Based on reflectance measurement at 565 nm and 760 nm, the N-Pen calculates NDGI (normalized difference greeness index) and predicts relative nitrogen content (N-content) in dry matter. As leaf structure specifically influences plant reflectance profile, N-content quantification was calibrated for three separate crops: wheat, barley and corn.

6.1. LEAF OPTICAL PROPERTIES AND NITROGEN

The N-Pen assesses N amount by means of spectral reflectance and by the concept of a close link between chlorophyll content and N content in soil and plants (Evans, 1983, 1989; Penuelas et al., 1994; Schlemmer et al., 2005). Since the presence of chlorophyll affects reflectance properties of leaves, optical methods based on spectral reflectance have been suggested to detect chlorophyll concentration (Yoder and Pettigrew-Crosby, 1995; Richardson et al., 2002; Gitelson et al., 2003).

Spectral reflectance is one of the optical methods widely used for indirect quantification of crop physiological status, which can be influenced by various factors, such as plant nutrients or pathological status. Chlorophyll absorption spectrum contains two absorption bands, one in the red and one in the blue region of visible spectrum (Fig. 2). Reflectance spectrum of green leaves roughly complements the absorption spectrum (compare A and B in Fig. 2), indicating that the presence of chlorophyll is critical for optical properties of leaves (Thomas and Gausman, 1977; Gitelson Merzlyak, 1994).

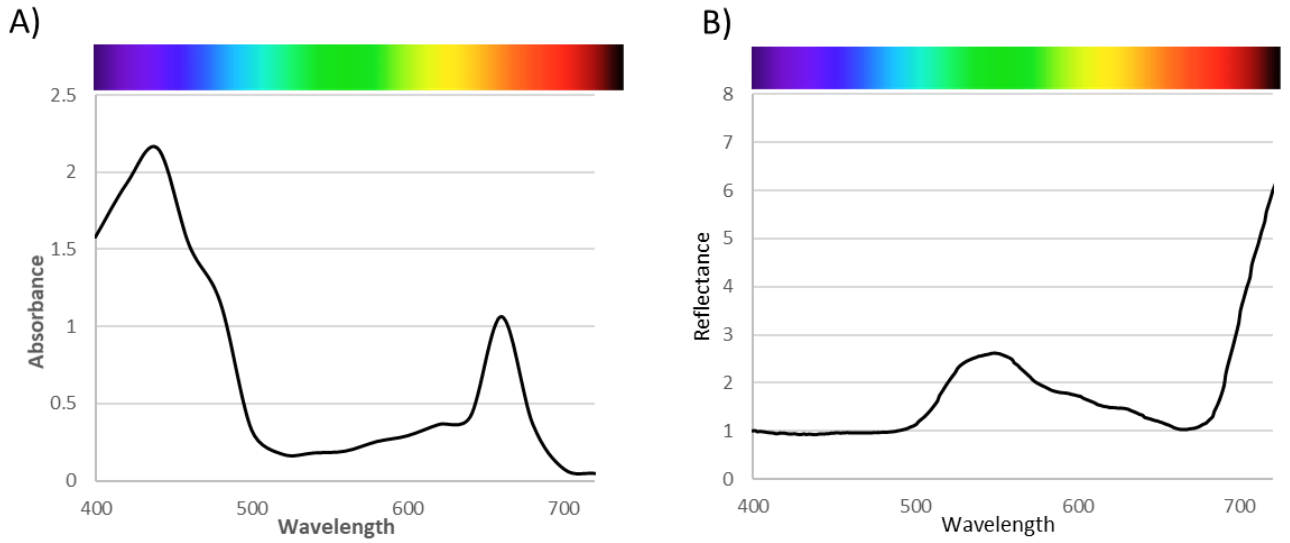


Fig. 2 Absorption and reflectance specrtra (A) leaf pigment extract (B) reflectance of leaf surface

To determine optimum wavelength for N-content prediction, several experimental plant groups with different levels of N nutrition were examined by measurement of spectral reflectance (Klem 2008). The reflectance values at each wavelength along reflectance spectrum were correlated with N-content in experimental plant groups. Pearson's correlation coefficient was used as a statistical measure of the strength of linear relationship between the paired data (Fig. 3A). The highest correlation was found for reflectance in the ranges 530 – 630 nm and 700 – 720 nm (negative correllation) and in the NIR region 750 – 900 nm (positive correlation). Indices based on reflectance in the green region (around 560 nm) were reported to be more sensitive to N and chlorophyll content (Gitelson *et al.* 1997) than indices based on reflectance in chlorophyll absorption maxima (e.g. NDVI).

Normalized Difference Greenness Index (NDGI) is calculated from leaf reflectance at wavelength bands 565 nm and 760 nm (equation 1),

$$NDGI = (R_{760} - R_{565}) / (R_{760} + R_{565}) \qquad \textit{(Equation 1)},$$

where R_{565} and R_{760} are reflectance values at indicated wavelengths. Based on a close relationship between NDGI and N-content in barley leaves at mid-tillering stage (Fig. 3B), the NDGI index was proposed to be a convenient tool for determination of N-content and N-nutrition state in barley (Klem, 2008).

Determination of NDGI index is strongly dependent on plant and leaf developmental stage as well as on environmental variability of field cultivation. Especially the first rapidly expanding leaf and the older leaves at the onset of senescence exhibit high variability of NDGI results. The most evident relationship between NDGI and N-content was reported in the second and the third youngest leaf (Klem 2008).

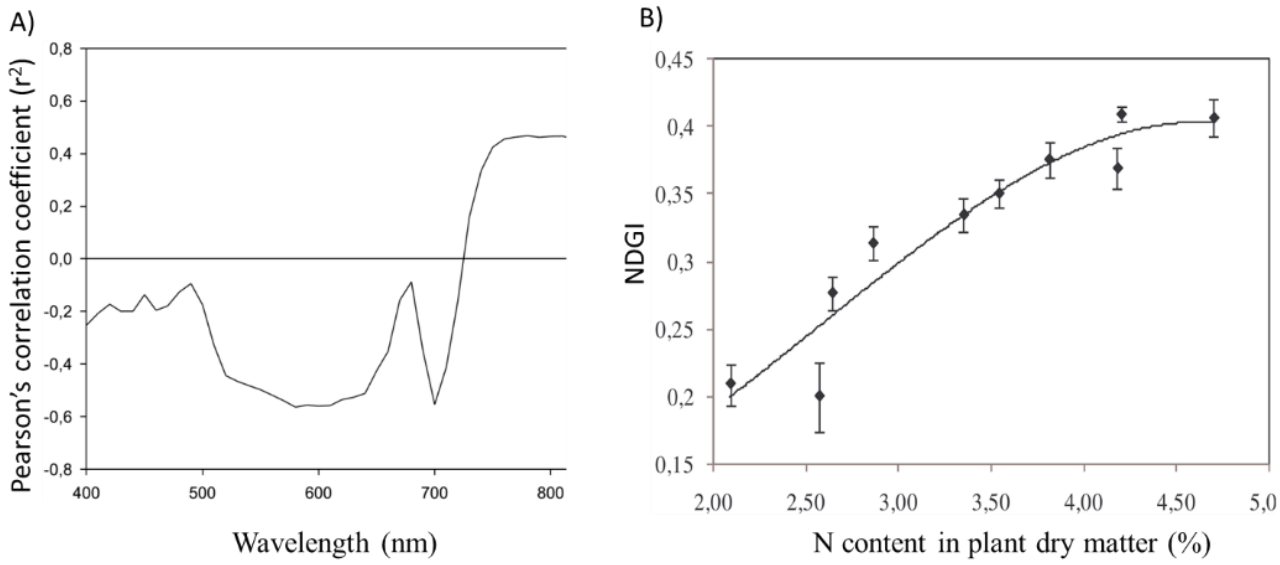


Fig. 3 Correlation between N content in barley dry matter and reflectance at particular wavelengths indicated as Pearsons correlation coefficient along the reflectance spectrum (A). Relationship between NDGI index and N content in barley dry matter (B) can be interpreted as a sigmoid curve. Vertical bars represent 95 % confidence intervals.

6.2. N-PEN CONFIGURATION

The N-pen N 110 is configured to quantify reflectance of the leaf illuminated with two LED light sources (565 nm or 760 nm, Fig. 4), calculate the NDGI index from average reflectance values and to predict the relative N-content in dry matter of plants or the postharvest grain N- content. Due to differences in leaf structure of various crops, formulas for calculation of N content were developed specifically for wheat, barley and corn. Each crop was tested on several varieties with similar results.

The N-Pen is pre-set by the manufacturer for a series of 10 measurements. After completing the series, the instrument calculates average NDGI value and the corresponding nitrogen content in plant dry matter (%). The average value is then calculated as follows: the maximum and minimum values are extracted and the average is calculated of the remaining eight values. If one (or more) of the 8 values significantly differ from the rest (outliers), the device does not accept it (them) and asks you to repeat one or more measurements.

The outliers typically arise with high variability of measured values, eventual leaf damage, disease infection or inproper fixing of leaf to measuring chamber. To exclude the outliers from the measurement the instrument uses Grubbs's test for outliers, where a *t* value is calculated as the largest absolute deviation from the sample mean. The *t* value is then compared with critical values stated in the table of Grubbs' critical values. If the calculated *t* value were higher than the critical value, the value is excluded due to high probability that the value is an outlier.

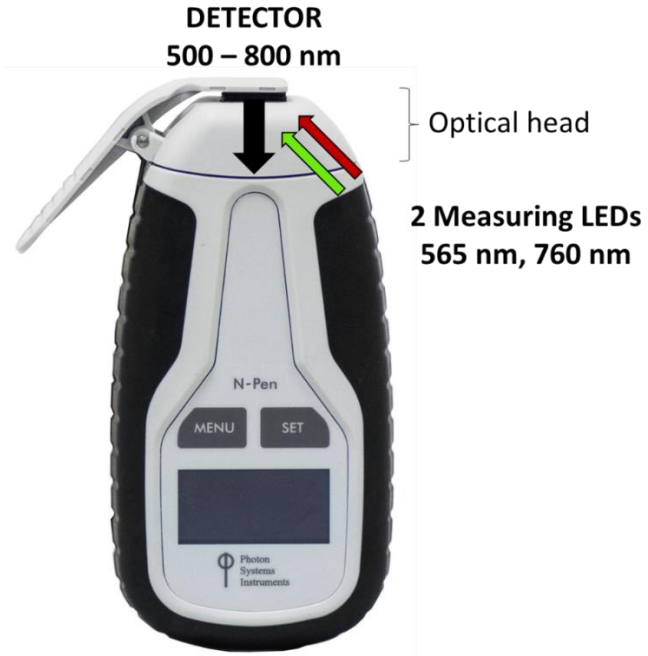


Fig. 4 N-Pen configuration

6.3. NITROGEN NUTRITION

The outcome of the measurement with N-pen is formulated as relative N-content in plant dry matter (%). Another practical interpration of the NDGI value, is the N nutrition state (Fig. 5). The maximum NDGI value is set to 100 % and the minimum reflectance is set equal to 0 % as follows:

1. NDGI > 0.65 indicates 100% N nutrition state (N = 100 %)
2. NDGI < 0.25 indicates 0 % N (N = 0 %)
3. If 0.65 > NDGI > 0.25, then nutrition state is calculated as N (%) = (NDGI - 0.25) * 250

As a general rule, nutrition state at early stages of development characterized by onset of tillering should be maintained at higher levels. Later, during shoot elongation and emerging flag leaf, (Novoa and Loomis 1981).

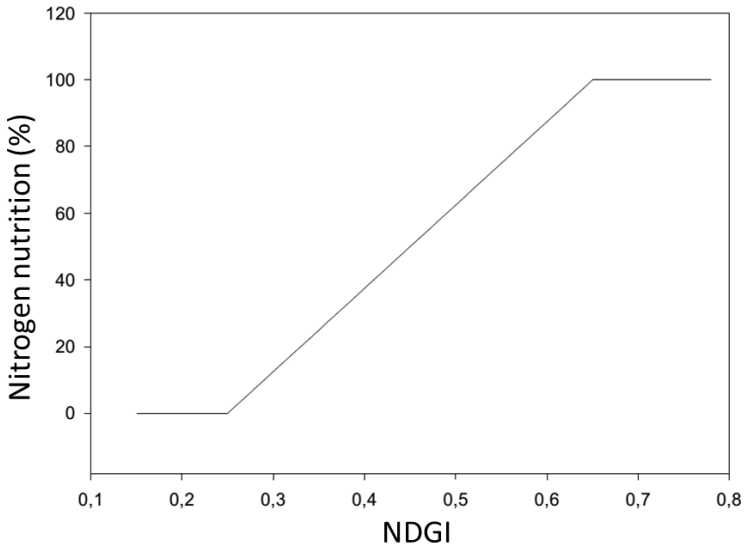


Fig. 5 Prediction of relative N nutrition from NDGI

7 HOW TO GET STARTED

Define the number of samples per each experimental group. The measurement is performed as a series of measurements, where the number of measurements can be set in range from 6 to 20. The pre-set number 10 can be used, for example, to measure 10 plants (each plant is measured once) or to measure 5 plants (each plant is measured twice). In case of high variability, the user is advised to reconsider the sampling method or to increase the sampling scale.

The N-Pen is controlled using two buttons:

- Use the **MENU** key to scroll through sequential menu options on the digital display.
- Use the **SET** key to select a menu option based on cursor (>) position.

	Before calibration, make sure that the optical part of the device, including the inner part of the sample holder, is clean. The device turns off automatically after 8 minutes of no use.

1. Switch on the device by holding ***SET*** button for 1 second.
2. Calibrate the N-Pen. Close the leaf clip with empty chamber and in the main menu select ***Calibration.*** The message "**Insert white sample"** appears on the display.
3. Press **SET** to perform and store the calibration to the device memory.
4. Set the device for measurement. Select ***Measurement*** in the main menu and select the type of crop (i.e. wheat, barley or corn). Then select the type of prediction, which is either N-content in dry matter of ***plant***, or the postharvest ***grain*** N-content. When ready, the display shows the number of remaining measurements, for instance *"Meas. left: 10"*. You can change the pre-set number in ***Settings > Readings***.
5. Select the second or the third youngest leaf and clip the leaf in the mid part along the leaf axis to the detector chamber.
6. Start the measurement by pressing ***SET*** to obtain and store the readings. The display shows the number of remaining measurements when ready for the next measurement.
7. Repeat the previous step to complete the whole series of measurement.
8. When complete, the instrument displays NDGI index value and the nitrogen prediction.
9. Download the readings from the device to computer. Press ***SET*** to exit the course of measurement and proceed to chapters 9 and 10.
10. Turn off the device by selecting ***Turn Off*** in the main menu.

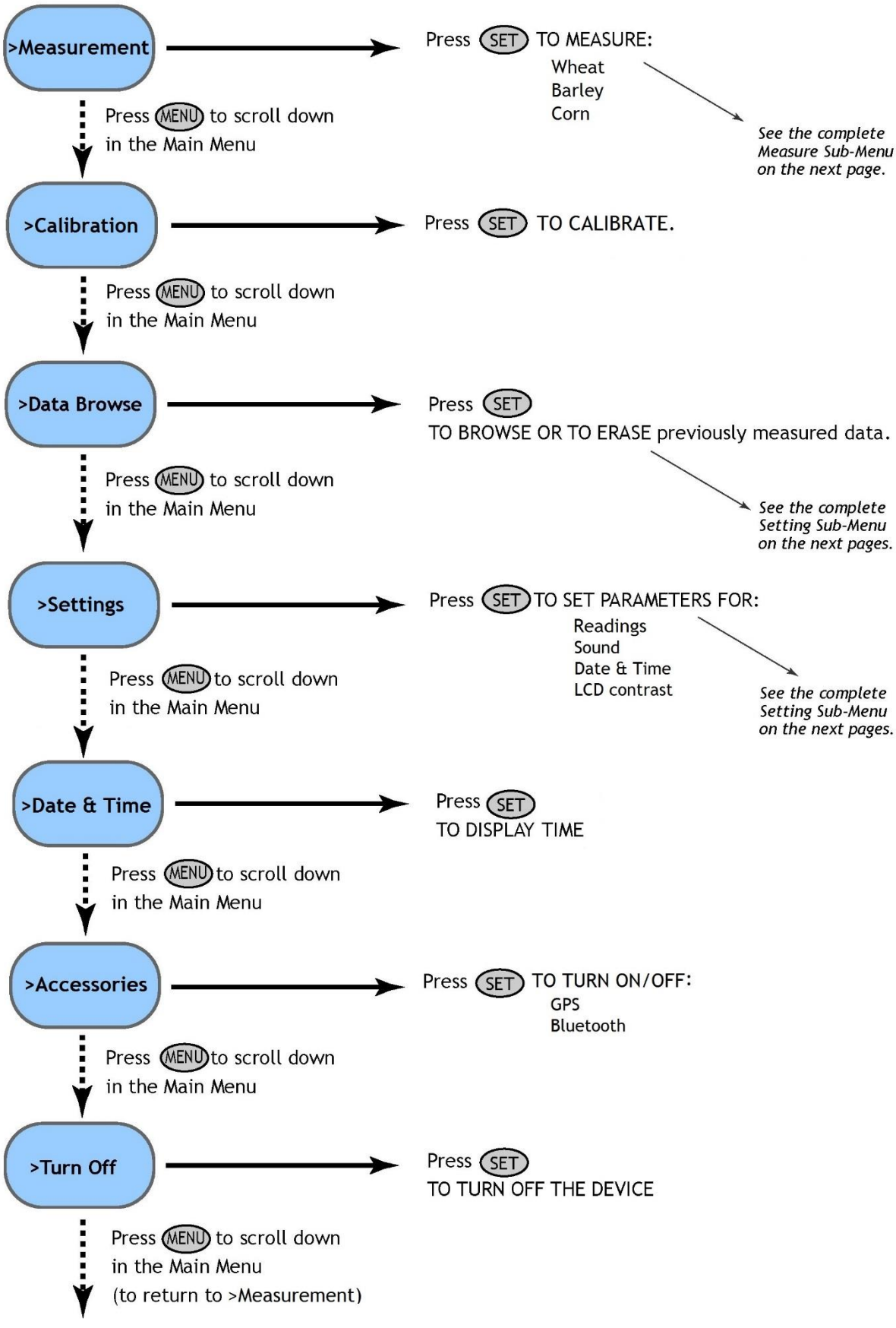
8 CONTROL MENU TREE

The next pages show the structure of the operation scheme, which includes the Main Menu and first-level Sub-Menus.

- The blue color represents the Main Menu and its Options.
- The yellow color represents the first-level Sub-Menus and their Options.
- Full-line arrows are used for the **SET** key.
- Dashed-line arrows are used for the **MENU** key.

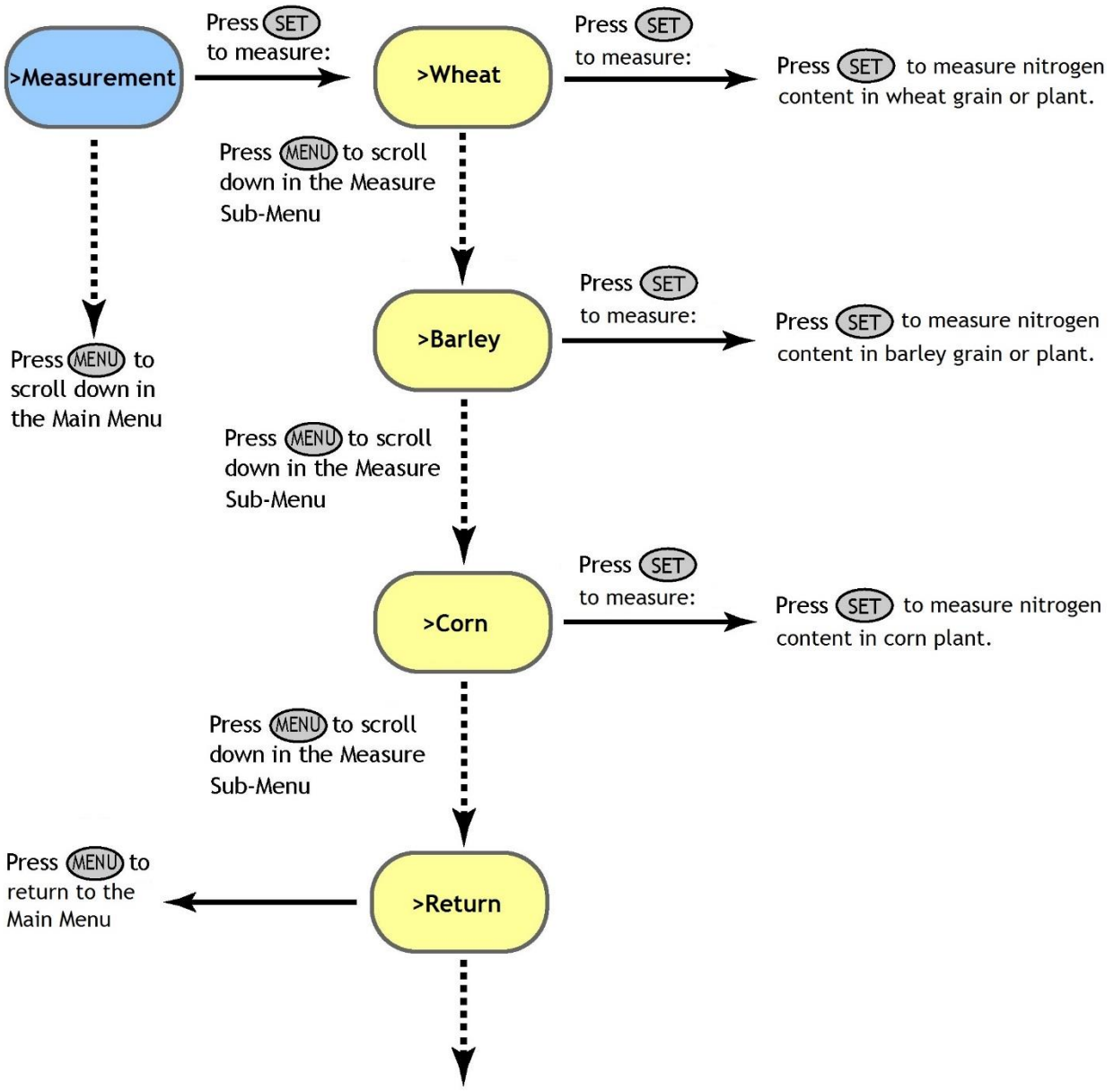
Main Menu

To start hold the SET key for 1 second.



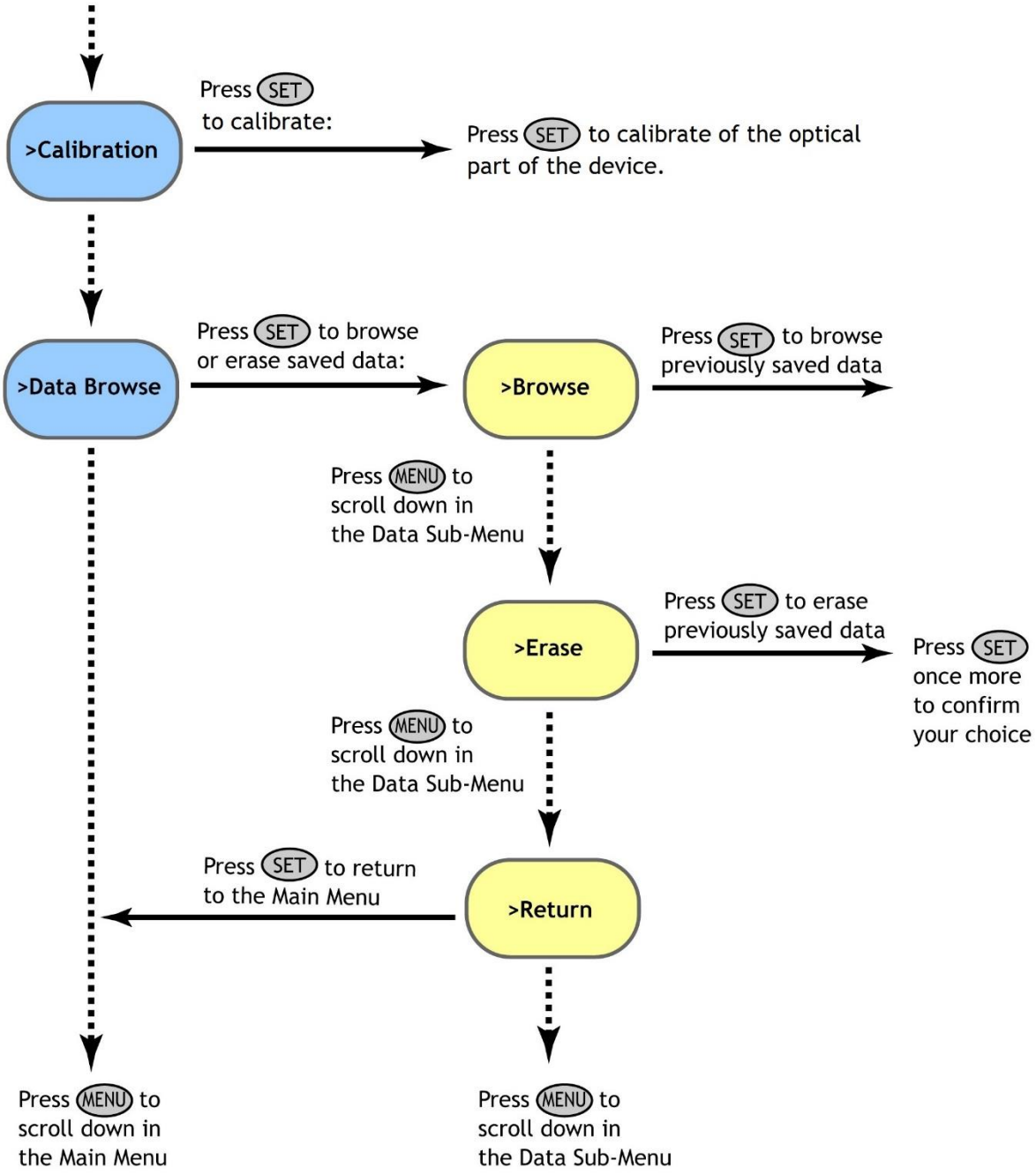
Measurement Sub-Menu

Use the Measurement Sub-Menu when measuring selected parameters.



Calibration and Data Sub-Menu

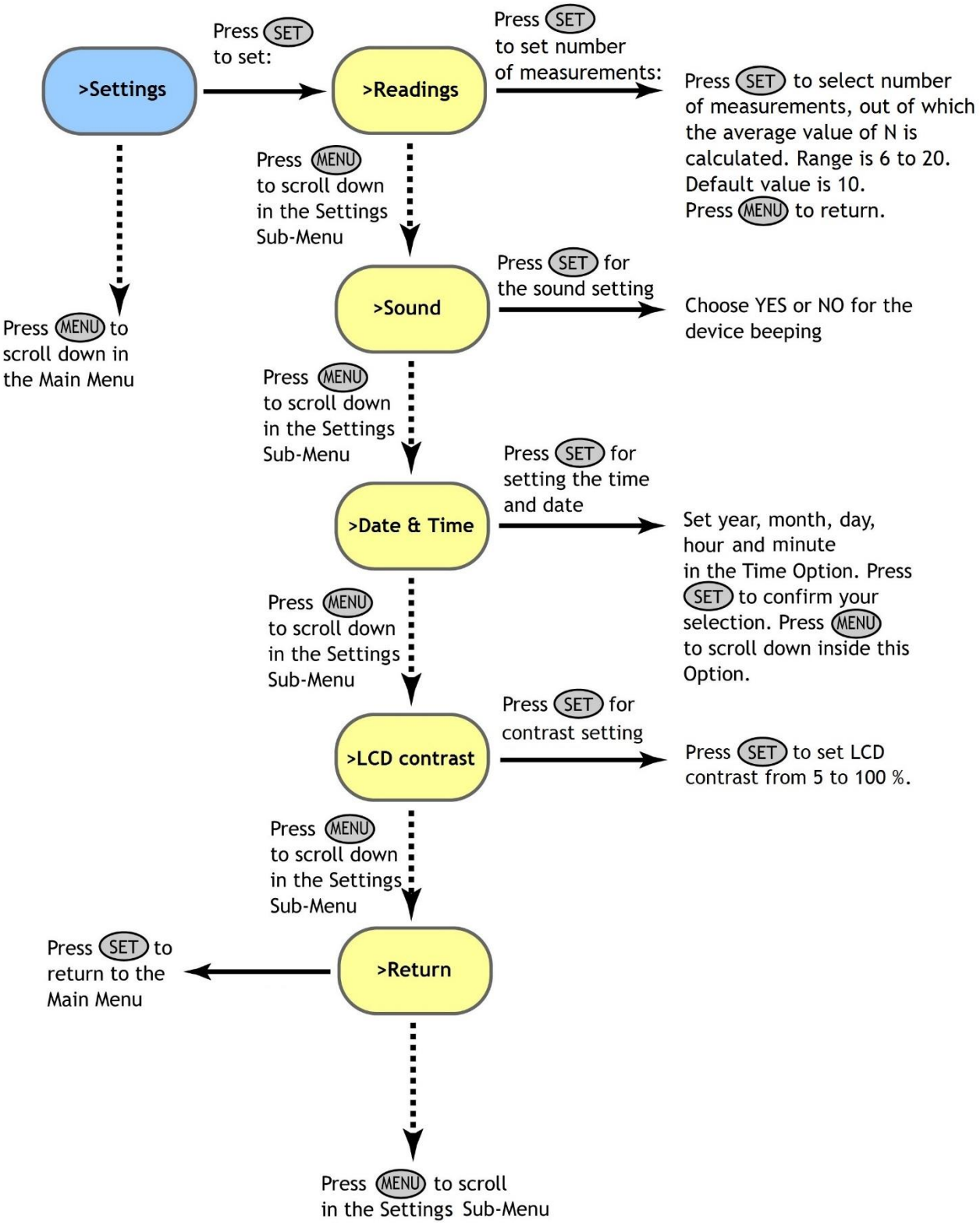
Use the Data Sub-Menu when browsing or erasing previously measured data.



IMPORTANT NOTE: Be aware that it is not possible to erase single data.
All stored data are erased!

Settings Sub-Menu

Use the Settings Sub-Menu to set the number of measurements, date, time, or the sound mode.



9 USB AND BLUETOOTH CONNECTION

9.1 USB CONNECTION

Connect the USB cable with the N-Pen device. Please note that lock in system is used.

⚠	Pay attention when connecting the USB cable not to damage the outlet connector on the Pen device. Make sure that you orient the cable correctly prior connecting the inlet with the outlet and the cable is upright towards the device.

To connect N-Pen with your computer please follow steps below in Fig. 6:

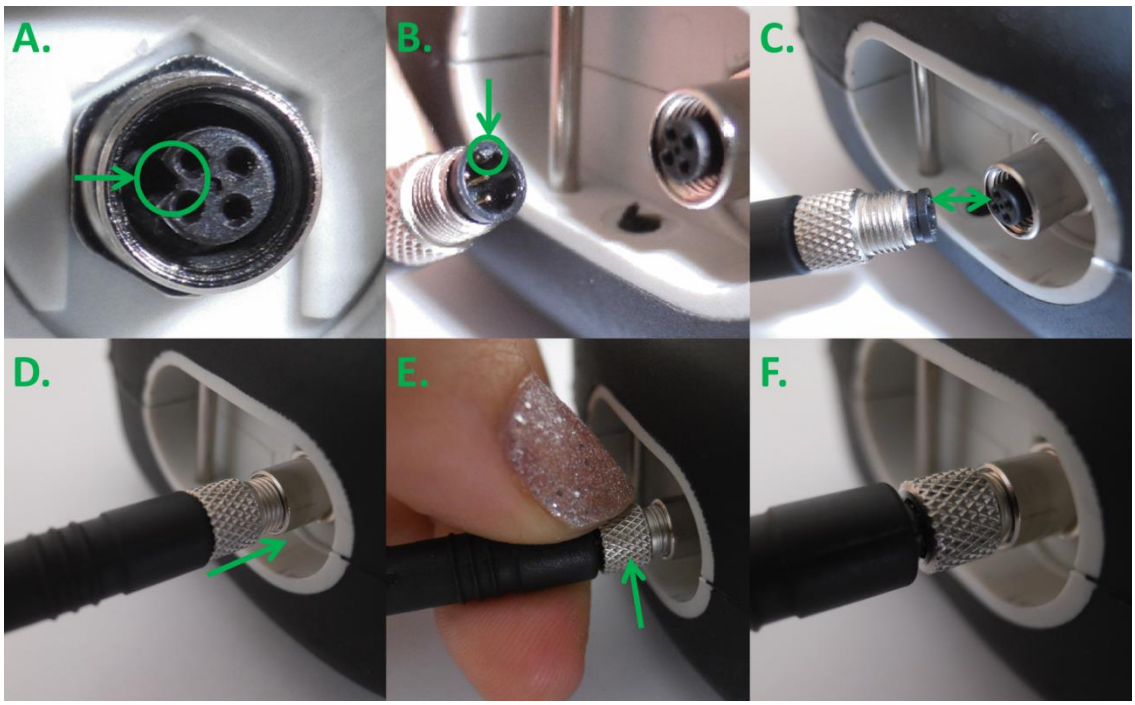


Fig. 6 How to connect N-Pen with PC.

A) Outlet connector on N-Pen device. B) Inlet part on the USB cable. C – E) Position the cable horizontally, plug in the inlet and screw the securing screw. F) Correct connection of the USB cable and Pen device.

Connect the USB cable to a computer. The N-Pen **switches ON** automatically after connecting the cable to the PC. For USB connection you need to have the USB driver installed in your PC. You find the driver on the installation disk (USB driver folder). If you check the Device Manager in Windows you should see the USB serial port in the device tree. In case of missing driver, you may download it from PSI websites. When the driver is installed correctly you should be able to connect to the device in the FluorPen software menu **Setup > Device ID**.

For more information about FluorPen software see chapter 10.

9.2 BLUETOOTH CONNECTION

Before you set up the Bluetooth connection between the N-Pen and your PC, make sure you have these components:

1. Bluetooth enabled PC

The PC with which you connect must have Bluetooth wireless technology, either built-in or through a Bluetooth card. Make sure that the PC's Bluetooth setting is "discoverable" (meaning that it shows up when other devices search for nearby Bluetooth connections). Consult the user guide for your PC or Bluetooth card to learn how to do this.

2. Bluetooth configuration software properly set up on PC

Before you can download files to your PC, you will need to set up the Bluetooth software that came with your PC, or your PC's Bluetooth card. This software varies by manufacturer. Please consult your PC's Bluetooth documentation for more information.

3. Bluetooth must be switched on visible on both devices

To pair the N-Pen with another Bluetooth device, such as a computer, you will need to ensure that Bluetooth is switched on visible on both devices.

9.3 BLUETOOTH PAIRING

1. Enabling Bluetooth in the N-Pen

- Switch ON the N-Pen (press and hold the **SET** key).
- Scroll to the **Accessories** menu (press the **MENU** key, then press the **SET** key).
- Select **Bluetooth On** (press the **MENU** key, then press the **SET** key) to enable Bluetooth.

	Keep in mind that the N-Pen turns off automatically after about 8 minutes of no action. Turning off the N-Pen always turns off Bluetooth.

2. Starting Bluetooth Application on Your PC

Be aware that this description is working on Windows 7; some of the steps may be different on your PC.

- Select: **Start > Devices and Printers** (Fig. 7).

  You may also start your Bluetooth application via the Control Panel: **Start > Control Panel > Hardware and Sound > Devices and Printers**.

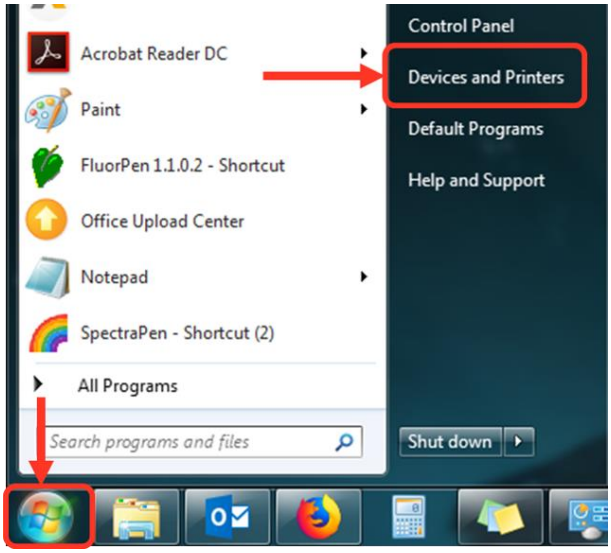


Fig. 7 Start Bluetooth Application.

3. Opening the Add Bluetooth Device Application

- Select: "Add a device" to start searching new Bluetooth device. Be sure that the N-Pen is in discoverable mode (see step 1).

Fig. 8 Add a device.

4. Selecting the N-Pen

- Select: PSI Pen icon.
- Select: Next (Fig. 9).

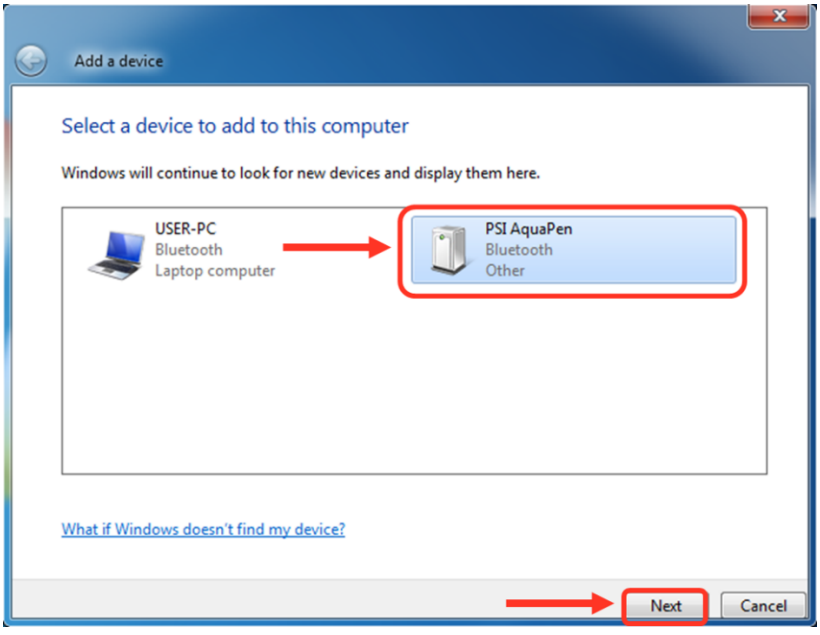


Fig. 9 Select the PSI Pen.

5. Starting the Pairing Process

Your Bluetooth Pairing Code is: 0000

- Select: “Enter the device’s pairing code”.
- Enter: **0000** (four digits).
- Select: Next (Fig. 10).

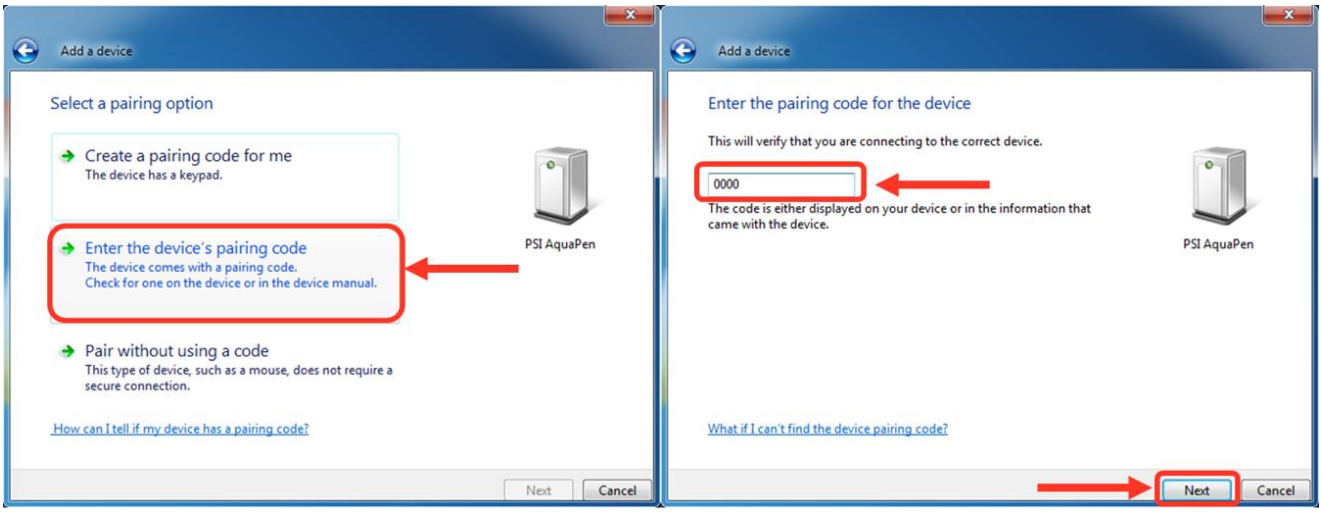


Fig. 10 Pairing process.

New version:

- Select: Yes (Fig. 11). **Please note that the N-Pen device does not display the verification number. The verification code is not important for the BT connection.**
- Select: Next.

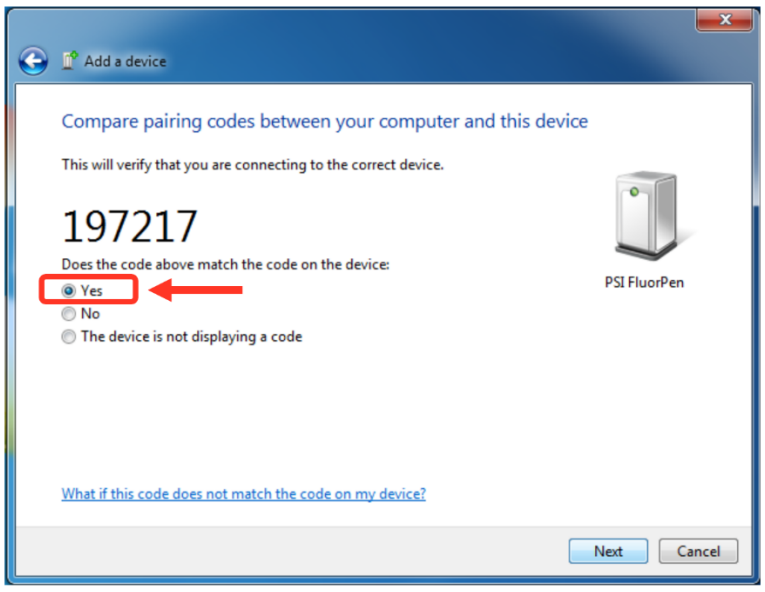


Fig. 11 Verifying of the BT pairing.

6. Completing the N-Pen Pairing

- Select: Close (Fig. 12).

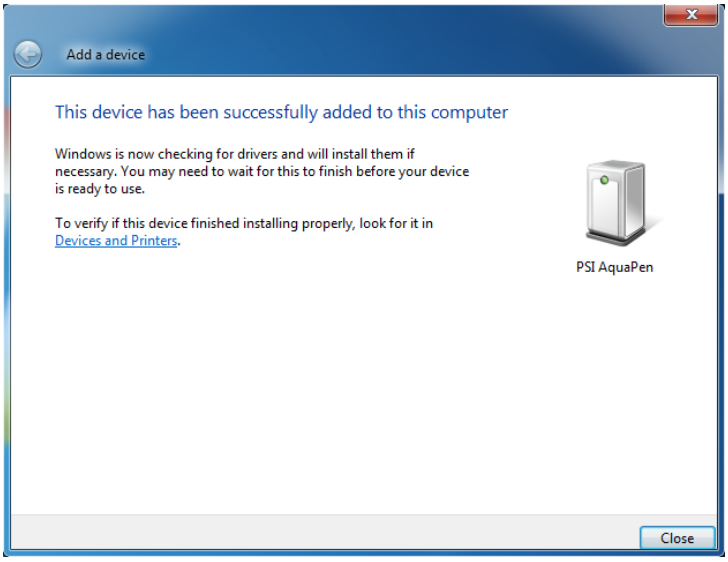


Fig. 12 Finishing.

On computer run the program FluorPen 1.1. For more information about FluorPen software see chapter 10.

10 FLUORPEN SOFTWARE

10.1 SOFTWARE INSTALLATION

1. Save the FluorPen software provided on the USB flash disk to your computer and launch the FluorPen program.
2. To connect and recognize your N-Pen device in the FluorPen software proceeds first with the registration of your FluorPen software (Fig. 13).
   - Select: Help > Register
   - Enter: your serial registration number.
   - Select: OK

Fig. 13 Software registration.

	Please note that you will find your serial (registration) number in the file **SN.txt**, which is included on the enclosed USB flash disk. Please remember: it is not possible to download data from the N-Pen device without software registration.

3. Switch on the N-Pen and enable Bluetooth or connect USB cable to the PC.
4. Make sure that your PC and the N-Pen are properly paired (see chapter 9 for complete information on USB and Bluetooth pairing).
5. Select: **Setup > Device ID (Ctrl+I)**. If properly connected, the message “Device: NPen” appears in the bottom part of the screen (Fig. 14).

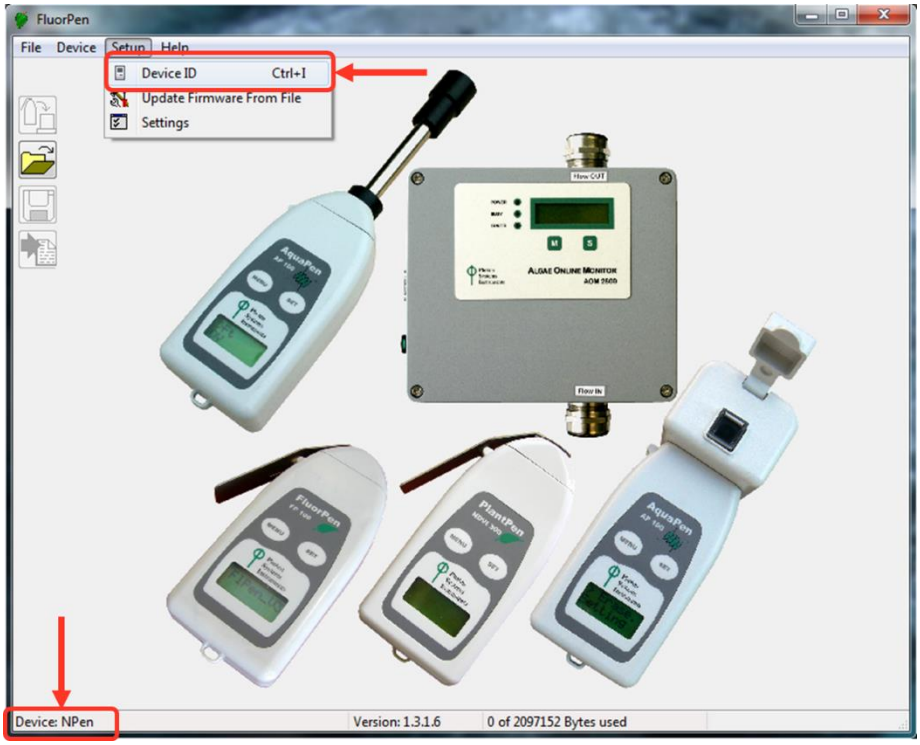


Fig. 14 Connecting N-Pen with Software.

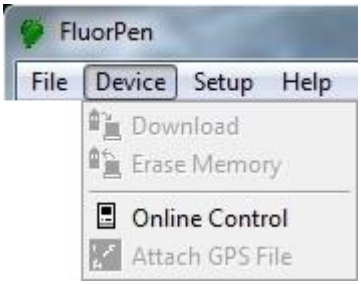
10.2 MENU AND ICON EXPLANATION

10.2.1 MAIN MENU

MENU: File

Load	Loads previously saved data files.
Save	Saves data to hard disc.
Export	Exports data in .txt format.
Export to JSON	Exports data in JavaScript Object Notation.
Close	Closes the current experiment.
Close All	Closes all running experiments.
Exit	Exits the program.

MENU: Device



Download	Downloads data from the N-Pen to your PC.
Erase Memory	Erases data from the N-Pen memory.
Online Control	Online control of N-Pen device.
Attach GPS File	Used for download data from GPS module (active only in older N-Pen version N 100).

MENU: Setup

Device ID Detects the connected device.

Update Firmware Used for firmware updates.

Settings Used for modification of the program settings.

MENU: Help

About Offers basic information about the program.

Register Used for the FluorPen software registration.

Icon Explanation:

Download Downloads data from the N-Pen to PC.

Load Loads (opens) previously saved data files.

Save Saves data to hard disc.

Export Exports data in .txt format.

10.2.2 MENU SETTINGS

MENU > Setup > Settings

After Download – Memory Erase

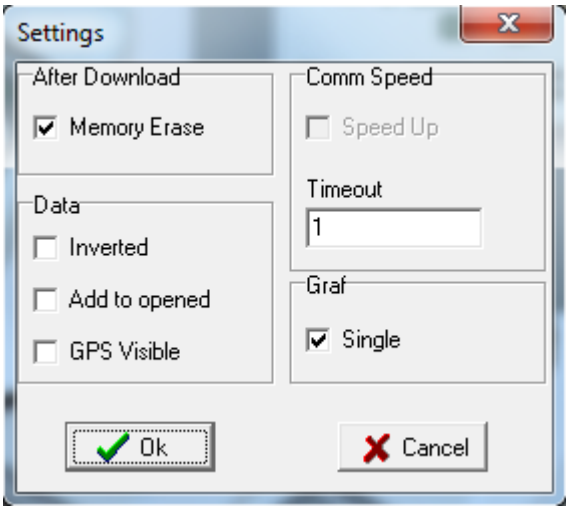
If the box is checked the N-Pen memory is erased after each data download.

Data – Inverted

If the box is checked the polarity of data is inverted, e.g., multiplied by -1. This feature can be helpful for a certain type of experiment when the measured data are undesirably interpreted as negative values.

Data – Add to opened

If the box is checked the downloaded data are added to that of the current opened experiment.



Data – GPS Visible

This option is active only in older N-Pen version N 100. In new version N 110 the GPS data are automatically downloaded and paired with protocol measurements.

Graf – Single

This function is not available for N-Pen device.

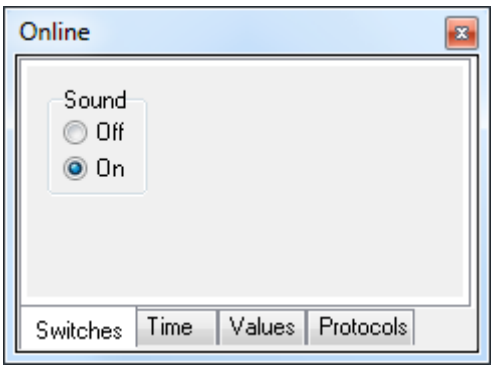
10.2.3 MENU ONLINE CONTROL

This function can be used for Online Control your N-Pen device after connection with your PC.

- Select: **Menu > Device > Online Control**

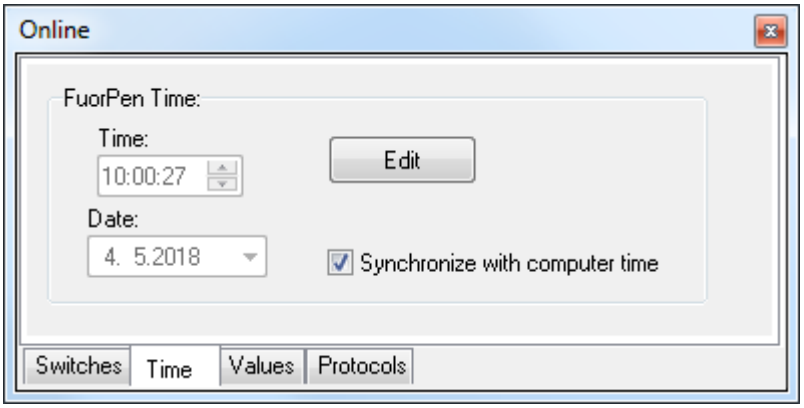
Online Control – Switches

Sound On/Off - choose On/Off for device beeping.



Online Control – Time

Set the N-Pen time a date. You can also synchronize time of N-Pen device with computer time.



Online Control – Values, Online Control – Protocols

These functions are not available for N-Pen device.

10.3 DATA TRANSFER

1. Perform a measurement with your N-Pen.
2. Click the **Download** icon or select **Device > Download**.
3. The Data table appears - Fig. 15.

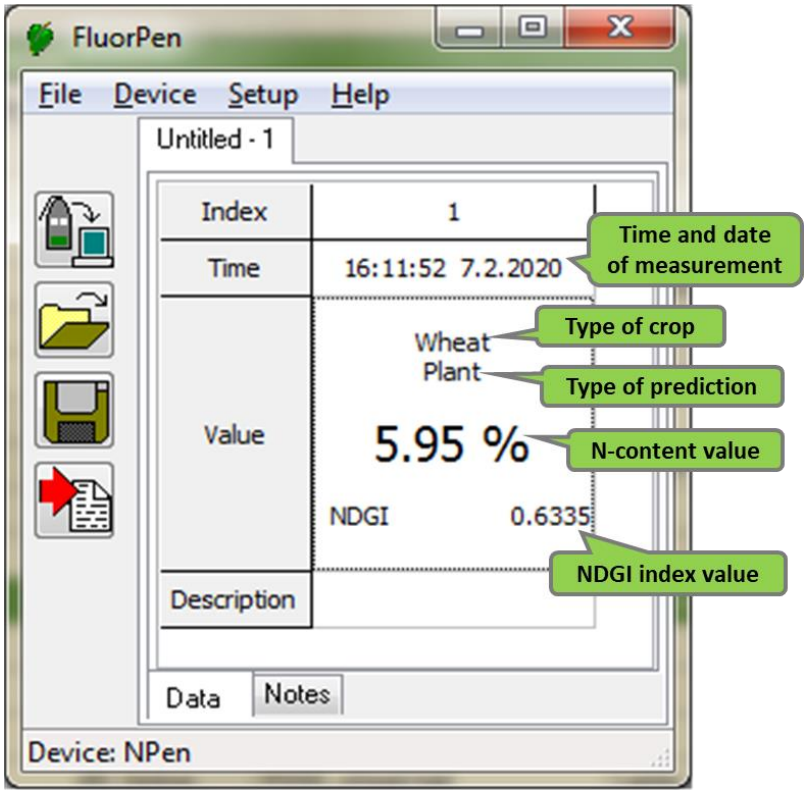


Fig. 15 Example of Data Transfer

4. For **export** press **File > Export** or **Export** icon (See chapter 10.2.1.). Select the data you want to export.

   Selected only – exports only one measurement that is selected by mouse, otherwise it will export everything.

   Source data – exports raw data, in case of nitrogen content: NDGI.

   Description – exports the data description if any.

   Computed values – export calculated data, in case of nitrogen content: N (%).

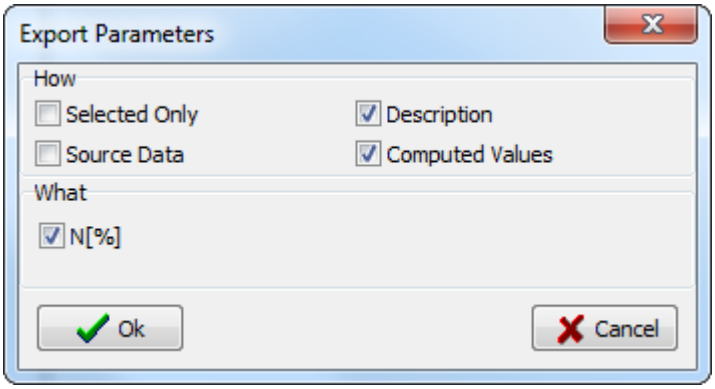


Fig. 16 Export data.

10.4 FIRMWARE UPDATE

⚠	All data in the N-Pen memory are erased during the firmware update! Before starting any firmware update, export all your data from the N-Pen memory into your computer!

1. Starting Update

- Select: **Setup > Update Firmware From File** (Fig. 17).

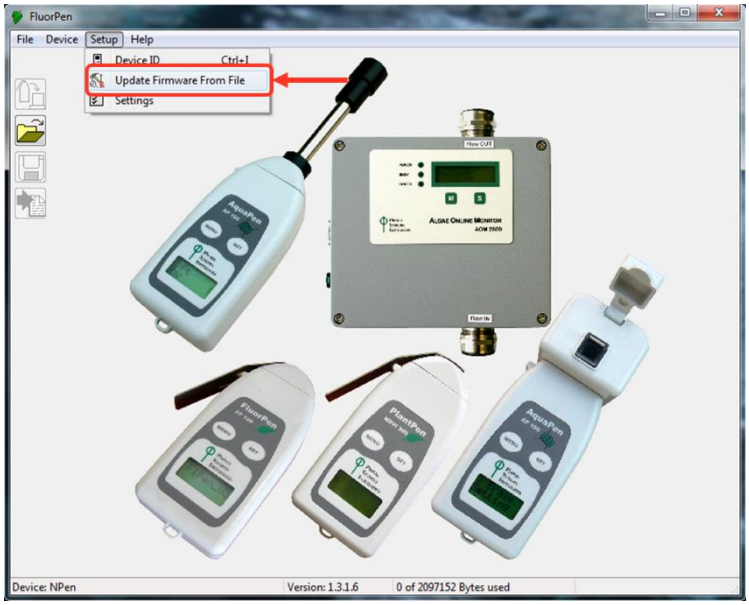


Fig. 17 Update Firmware.

2. Warning

- Select: OK to start update (Fig. 18).

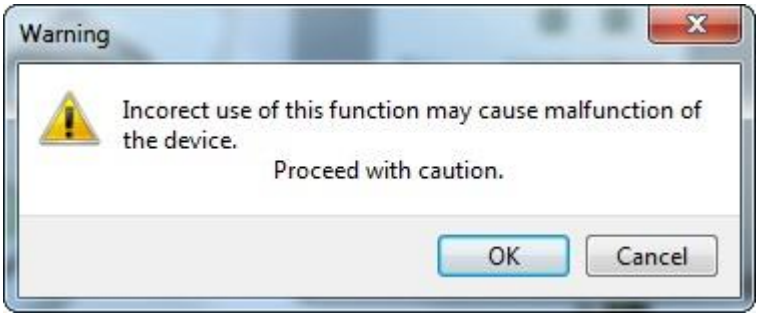


Fig. 18 Warning.

3. Selecting .bxn file

- Find: Binary file (with the extension .bxn) (Fig. 19).
- Select: Open.

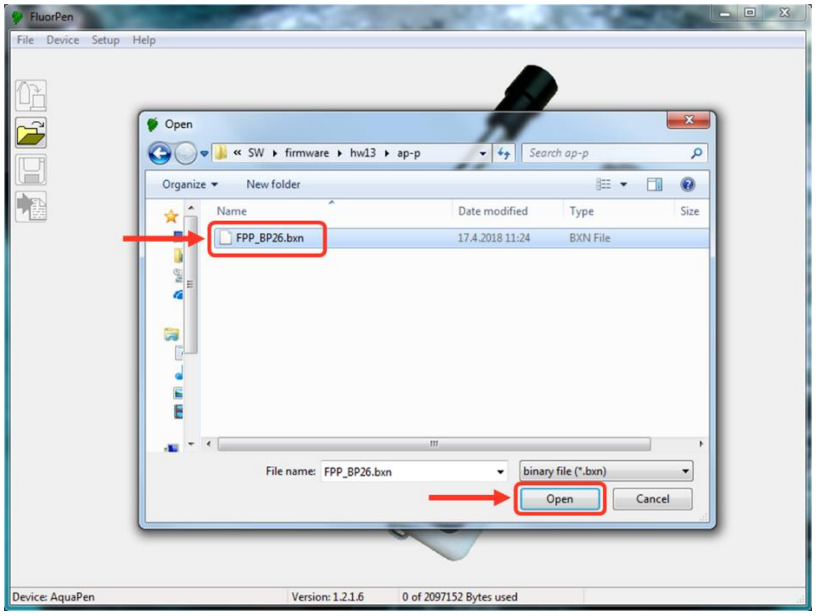


Fig. 19 Select .bxn file.

4. Finishing Upload

- Select: **OK** to start uploading of the update (Fig. 20).

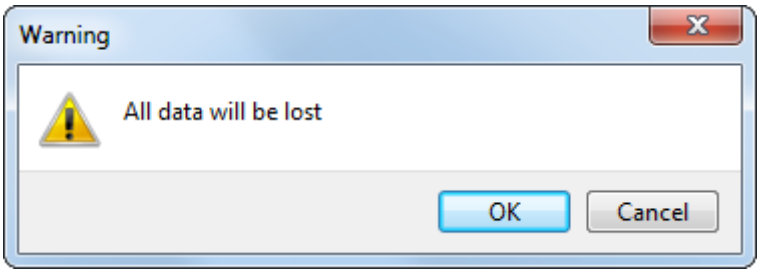


Fig. 20 Finishing upload.

- The bottom bar indicates the upload progress (Fig. 21).

Fig. 21 Upload progress.

- Press: **OK** to finish upload (Fig. 22).

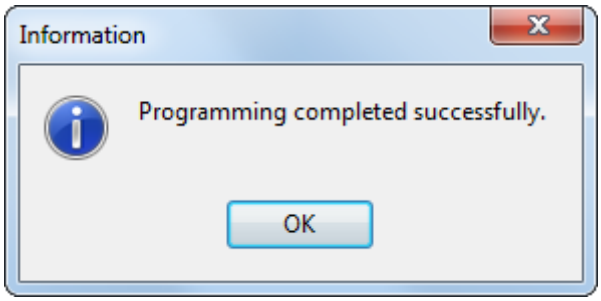


Fig. 22 Finish upload.

11 GPS MODULE

N-Pen device has integrated GPS module which can be turned on during the measurement and the GPS coordinated will be added to the downloaded data.

	For proper GPS reading, the time in your N-Pen and in your computer must be synchronized. Preset time and time zone must correspond to GPS time (time zone) in your location.

11.1 GPS / N-PEN OPERATION

1. Check the time setting in N-Pen device: **Settings > Date & Time**
2. Switch the GPS module on:
   - Select: **Accessories > GPS**
   - Press **SET** to confirm.
   - Wait until the GPS position is found – "**Starting GPS**".
   - The GPS module is ready when the icon in upper panel change – see on Fig. 23.

Fig. 23 GPS icons.

3. Go to **Measurement** and choose required protocol.

	For prompt determination of the coordinates use the option **Accessories > GPS > Location.**

	The device may need a clear view of the sky to acquire satellite signal. Keep in mind that the N-Pen turns off automatically after about 8 minutes of no action. Turning off the N-Pen always turns off GPS module.

11.2 DATA DOWNLOAD

1. Enabling Communication:
   - Switch on the computer and the N-Pen. Set your computer to N-Pen communication: enable Bluetooth or connect to USB port.
2. Downloading Data from the N-Pen
   - Start FluorPen program.
   - Connect N-Pen device: **Setup > Device ID (Ctrl+I)**
   - Download measured data from the N-Pen to your PC. Data measured with activated GPS module are downloaded with GPS coordinates (Fig. 24).

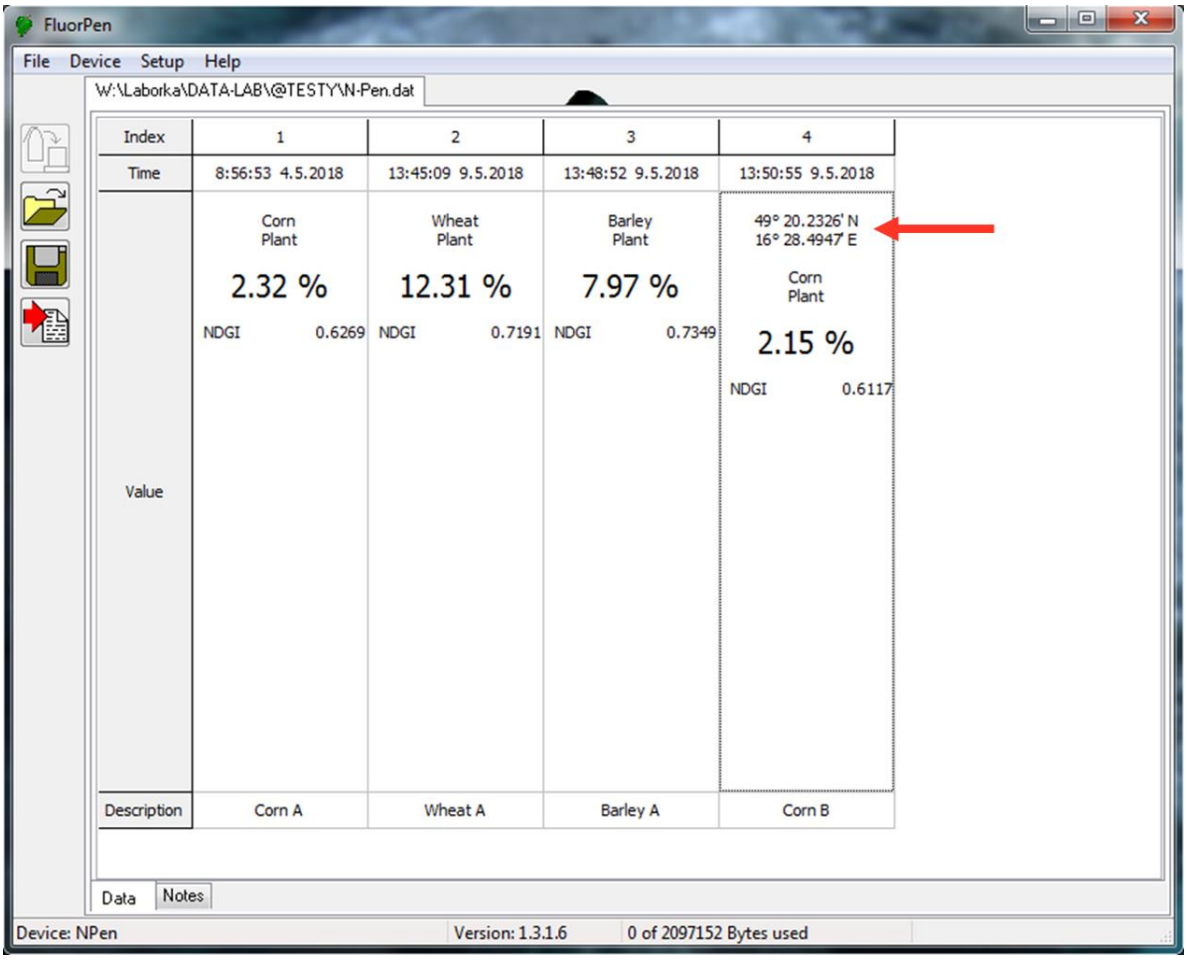


Fig. 24 GPS coordinates.

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13 WARRANTY TERMS AND CONDITIONS

- This Limited Warranty applies only to the N-Pen device. It is valid for one year from the date of shipment.
- If at any time within this warranty period the instrument does not function as warranted, return it and the manufacturer will repair or replace it at no charge. The customer is responsible for shipping and insurance charges (for the full product value) to PSI. The manufacturer is responsible for shipping and insurance on return of the instrument to the customer.
- No warranty will apply to any instrument that has been (i) modified, altered, or repaired by persons unauthorized by the manufacturer; (ii) subjected to misuse, negligence, or accident; (iii) connected, installed, adjusted, or used otherwise than in accordance with the instructions supplied by the manufacturer.
- The warranty is return-to-base only and does not include on-site repair charges such as labor, travel, or other expenses associated with the repair or installation of replacement parts at the customer's site.
- The manufacturer repairs or replaces faulty instruments as quickly as possible; the maximum time is one month.
- The manufacturer will keep spare parts or their adequate substitutes for a period of at least five years.
- Returned instruments must be packaged sufficiently so as not to assume any transit damage. If damage is caused due to insufficient packaging, the instrument will be treated as an out-of-warranty repair and charged as such.
- PSI also offers out-of-warranty repairs. These are usually returned to the customer on a cash-on-delivery basis.
- *Wear & Tear Items* (such as sealing, tubing, padding, etc.) are excluded from this warranty. The term *Wear & Tear* denotes the damage that naturally and inevitably occurs as a result of normal use or aging even when an item is used competently and with care and proper maintenance.

14 TROUBLESHOOTING AND CUSTOMER SUPPORT

In case of troubles and for customer support, please, visit **FAQ** on our websites, write to **support@psi.cz** or contact your local distributor.