Spectrophotometers

Implen P360 (PSC 533)

Every time you use the Spectrophotometers, you MUST sign the log book.

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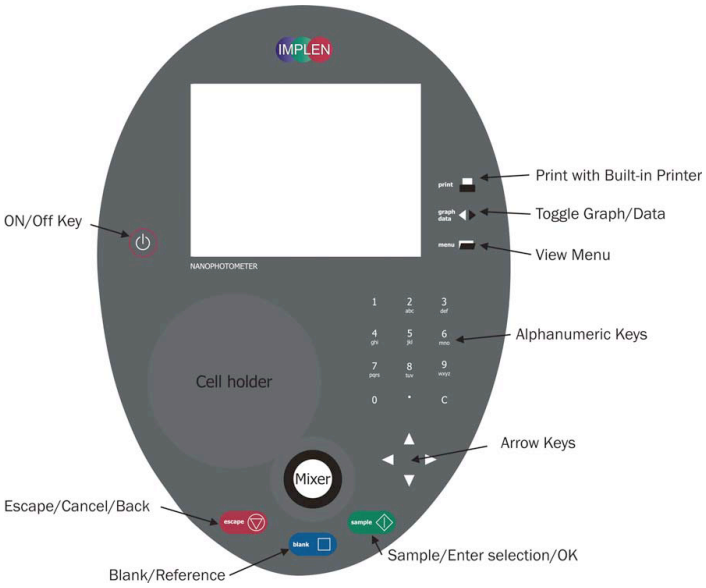
Implen Nanophotometer P-360

1. **Applications and Overview**

Nucleic Acids-	dsDNA, ss DNA, ssRNA, oligonucleotides
Proteins-	Protein UV (A280), Protein Dye, Bradford, Lowry, BCA
Cell Density-	for bacterial cell cultures
General Functions-	Specific wavelengths, wavescan



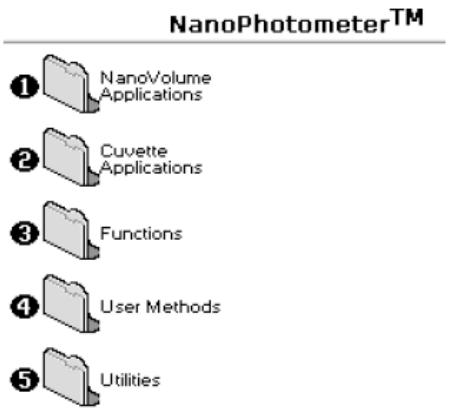
Key	Action
On/Off Key	Turns the instrument on/off.
Arrow Keys	Use the four arrow keys to navigate around the display and select the required setting from the active (highlighted) option.
View Menu	View menu for that application mode. Some of these are common to all applications and described on page 8. Menu unique to an application are described in the relevant section of the NanoPhotometer® P-Class User Manual.
Alphanumeric Keys	Use these to enter parameters and to write text descriptions where appropriate, or required. Use repeated key presses to cycle through lower case, number and upper case. Leave for 1 second before entering next character. Use C button to backspace and 1 to enter a space.
Escape/Cancel/Back:	Escape from a selection and return to the previous folder. Cancel a selection. Stop making measurements.
Blank/Reference	Set reference to 0.000 A or 100%T on a reference solution at the current wavelength in the mode selected. When in scan mode, does a reference scan.
Sample/Enter Selection/OK:	Enter, or confirm a selection. Take a measurement.
Print (P 330 and P 360 only)	Prints the results shown on the screen on the built-in printer, if a built-in printer is connected to the NanoPhotometer®.
Graph/Data (P 330 and P 360 only)	Toggle graph on/off. The graph shows a wavescan plot across the range 220 nm to 400 nm (for Dye methods 220 nm to 750 nm) with cursors denoting 230, 260, 280 and 320 nm (Nucleic Acid methods) and 260, 280 and 320 nm (Protein methods).

2. Start and Menu Options:

Turn power on and wait.

Choose application fold with numeric pad (ex, #1 for Protein)

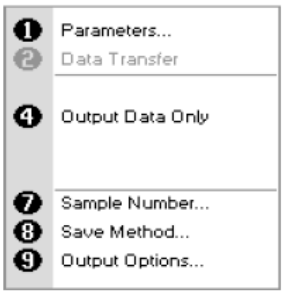
The user interface is built around folders which are displayed on the main screen when the instrument is switched on. Different folders are numbered and opened by using the associated number key on the keypad. After switching on the NanoPhotometer® a self-calibration check is performed and the default main screen "NanoPhotometer®" is offering the choice of:



Keypad number	Description
1	Life Science methods such as nucleic acid assays and protein assays using the NanoPhotometer® P-Class Submicroliter Cell
2	Life Science methods such as nucleic acid assays, protein assays and cell density using cuvettes
3	General spectroscopic methods
4	Contains nine folders that can store user adapted methods (up to 81)
5	Instrument set up (date, time, number format), Output Options and Baseline Compensation set up.

Menu Options:

Menu options for P 330/P 360:



After each measurement the following options are possible in the Menu:

1) Return to parameter screen.
2) Transfer the results via selected Output Option.
4) Toggle on/off the graph in the print-out or saved file.
7) Define the sample number you wish to start from.
8) Save the parameters as a method.
9) Open Output Options settings, possibility to change the Output Options settings within the method as described in 7.3 Output Options / Printer

Exit ***Menu*** by pressing ***Escape*** ▽, **OR** wait

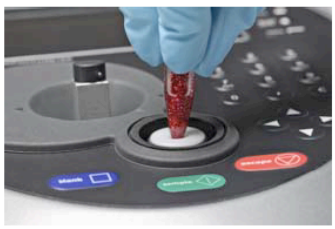
3. THE NANOPHOTOMETER® P-CLASS SUBMICROLITER CELL

With its innovative optical pathway the cell is designed for optimum measurement results with submicroliter samples ranging from 0.3 µl up to 5 µl of undiluted sample. Due to a pathlength of 0.04 mm, 0.1 mm, 0.2 mm, 1 mm and 2 mm the cell is offering an automatic dilution of 1/250, 1/100, 1/50, 1/10 and 1/5 in comparison to a standard cuvette measurement. Because the measurements are processed with undiluted samples, the reproducibility of the results is extremely high. If desired, samples can be retrieved after the measurement for further processing. The NanoPhotometer® P-Class Submicroliter Cell can be used for all UV/Vis analysis utilizing the wavelength range of 190 nm to 1,100 nm.

The NanoPhotometer® P-Class Submicroliter Cell is delivered for version P 300 with one lid with a pathlength of 0.2 mm (Lid 50), for version P 330 with two lids pathlength 0.2 mm (Lid 50) and 1 mm (Lid 10) and for version P 360 with three lids pathlength 0.04 mm (Lid 250), 0.2 mm (Lid 50) and 0.1 mm (Lid 10). Lid 5 (2 mm pathlength), Lid 100 (0.1 mm pathlength) and Lid 250 (0.04 mm) can be ordered optionally. The dilution factor (lid factor) is printed on the lid. Please make sure that you use the appropriate lid for your sample.

3.1 Technical instructions

Step 1 Insert the NanoPhotometer® P-Class Submicroliter Cell into the cell holder with the cell windows facing the light beam. We recommend facing the Implen logo to the front. The light beam is directed from RIGHT to LEFT as indicated with small arrows. Insert the NanoPhotometer® P-Class Submicroliter Cell always in the same direction.



Step 2 Use the integrated vortexer (P 330 / P 360 only) to mix your sample well to achieve an accurate homogeneity of the sample.

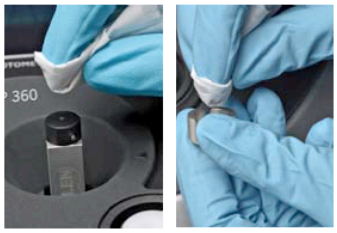
Step 3

Pipette the appropriate sample volume onto the centre of the measuring window. **Warning!!** Do not overfill the well.

Lid	Sample volume	Pathlength	Dilution
5 (optional)	3.5 – 5 µl	2 mm	1:5
10 (optional for P300)	1 – 3 µl	1 mm	1:10
50	0.3 – 2 µl	0.2 mm	1:50
100 (optional)	0.3 – 2 µl	0.1 mm	1:100
250 (optional)	0.3 – 2 µl	0.04 mm	1:250

Step 4

Make sure that for the measurements the lid fits exactly onto the positioning supports mounted to the body of the cell. Take measurement. **Remember** to consider the lid factor in your instrument software. Please refer to the NanoPhotometer® P-Class User Manual for detailed information.



Step 4

Take the lid off and retrieve the sample with a pipette for further applications if desired. Remove sample residues from the measurement window and the mirror in the lid. Clean the measurement window and mirror in the lid well with a slightly wet fluff-free tissue. Use water, 70% ethanol or isopropanol. Do not use aggressive solvents like strong acids or bases or organic solvents at any time.

Important Note: Residual fluffs must be removed for optimum performance

Your cell is ready for the next sample.

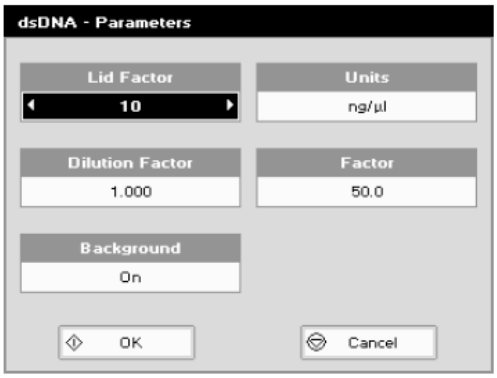
Operation Limitations: Do not autoclave the unit! Do not use an ultrasound bath to clean! Do not drop in water or solvent bath. The unit is water resistant, but not water proof!

3.2 Software instructions

The ***NanoVolume Applications*** and ***Cuvette Applications*** are very similar concerning the analysis of dsDNA, ssDNA, RNA, Oligonucleotides, protein UV and protein dye analysis. This section describes the specific features which have to be considered using the NanoPhotometer® P-Class Submicroliter Cell. For general information please follow the detailed instructions under Nanovolume Applications and Cuvette Applications.

The procedure is as follows:

Exemplary Parameter Screen



Parameter Screen

Step 1 Press 1 to select ***NanoVolume Applications*** folder

Step 2 Press 1 to select ***Nucleic Acids*** folder **OR** 2 to select ***Protein*** folder.

Step 3 Select the method you want to use by pressing the corresponding number.

Step 4 Select the ***Lid Factor*** using the left and right arrows.

Lid	Sample volume	Pathlength	Dilution
5 (optional)	3.5 – 5 µl	2 mm	1:5
10 (optional for P 300)	1 – 3 µl	1 mm	1:10
50	0.3 – 2 µl	0.2 mm	1:50
100 (optional)	0.3 – 2 µl	0.1 mm	1:100
250 (optional)	0.3 – 2 µl	0.04 mm	1:250

Step 5 Select subsequent parameters and specifications as described under **4.** ***Nanovolume Applications and Cuvette Applications.***

After the selections are confirmed the results screen displays in top left corner the chosen Lid and the required sample volume.