



SPECTROPHOTOMETER PE-5400UV

OPERATING MANUAL DATA SHEET

BKRE.941412.001-03OM

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The present Data Sheet and Operating Manual certify the parameters and technical specification of PE-5400UV spectrophotometer guaranteed by the manufacturer.

The Data Sheet and Operating Manual establish the rules of operation of the spectrophotometer, observance of which ensures trouble-free operation of the instrument.

Prior to switching on the spectrophotometer, study carefully the present Operating Manual and safety precautions.

The construction of the device and its internal software are being continuously improved; the actual version of this document is on the CD with the software.

Attention! The manufacturer provides the warranty maintenance of the spectrophotometer in its service centre with the original packing only.

1. Purpose

PE-5400UV spectrophotometer is intended for measuring the transmittance factor and absorbancy of liquids (including the biological ones) for the purpose of determining the components dissolved in them as well as for measuring the transmittance factor and absorbancy of solid and liquid samples of various origins.

The scope of application of spectrophotometers comprises the environmental analytical and sanitation and epidemiological laboratories of medical institutions as well as chemical, optical and biological laboratories of industrial enterprises, research and development establishment and educational institutions.

2. General Information and Technical Specification

2.1. General Information

PE-5400UV spectrophotometer is manufactured by Ecohim Co. Ltd.

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For correspondence: 22 17th Line, building I, Suite 406, Vasilyevsky Island, Saint Petersburg 199178

Phone/fax: (812) 322-9600, 449-3122, 449-3123

E-mail: info@ecohim.ru, URL: <http://www.ecohim.ru>

PE-5400UV spectrophotometer is a stationary table-top instrument consisting of the optomechanical and electronic units installed in the cabinet. PE-5400UV spectrophotometer is constructed according to a single-beam scheme. The instrument uses a grating monochromator. A halogen and deuterium lamps are used as a radiation source and a silicon photodiode – as a receiver. The results of measurements are output to the liquid-crystal graphical indicator.

The manufacturer equips PE-5400UV spectrophotometer with a four-position or six-position cell holder for European cells with the width of 12 mm; the holders for vials (test tubes) and microcells are also available.

2.2. Certification Information

PE-5400UV spectrophotometer is accompanied by Certificate of Measuring Equipment Type Approval No. 40437 issued by the State Committee for the Russian Federation for Standardisation and Metrology on August 10, 2015 and registered with the State Register of Measuring Equipment under No. 44866-10. The Certificate is valid until July 30, 2020.

Registration Certificate of the Medical Product No. FSR 2010/07089.



2.3. Basic Technical Data

Table 1

Parameter description	Value
Spectral range, nm	190 to 1,000
Range of measurement of spectral factors of direct transmission, %T	0.0 to 100.0
Range of readings of spectral factors of direct transmission, %T	0.0 to 200.0
Range of measurement of absorbancy, B	3.000 to 0.000
Range of readings of absorbancy, B	3.000 to - 0.300
Allowable absolute error limits when measuring the spectral factors of direct transmission, %T	
- 190 to 315 nm	± 1.0
- 315 to 1,000 nm	± 0.5
Re-calculation of the error when measuring the absorbancy	$\Delta A = 0.43 \cdot \Delta T \cdot 10^{A-2}$
Separable spectral range, nm	4
Diffused light level	$\leq 0.3\%T$ at 340 nm
Optical arrangement	single-beam
Overall dimensions, (L x W x H), not more than, mm	465x395x235
Weight: not more than, kg	12.5
Service life, years	8

2.4. Information on the Content of Precious Materials

Table 2

Material:	Content
Gold	none
Silver	none
Platinum	none
Iridium	none
Rhodium	none
Palladium	none
Ruthenium	none
Osmium	none
Diamond	none

3. Standard Equipment

The standard equipment of the spectrophotometer includes:

- PE-5400UV spectrophotometer with the following cell holders installed:
(standard equipment 1)
four-position holder for the cells with the width of 24 mm and length of up to 100 mm 1
(standard equipment 2)
four-position holder for the cells with the width of 12 mm and length of 10 mm..... 1
- *glass cell:
(standard equipment 1)
KFK (L10x24 mm) 4
(standard equipment 2)
L10x12 mm 4
- *quartz cell:
(standard equipment 1)
KFK (L10x24 mm) 2
(standard equipment 2)
L10x12 mm 2
- *complete set of reference light filters (4 filters) 1
- *spare halogen bulb (12 V, 20 W) 1
- *power cord..... 1
- USB-A - USB-B cable for connection to a PC..... 1
- dust cover 1
- Operating manual and data sheet..... 1

Note:

1. The position marked with an asterisk are located in the three recesses under the instrument.
2. The additional accessories (bulbs, cells, light filters) are delivered to separate order.

4. Construction and Principle of Operation

4.1. Component Parts

The spectrophotometer consists of the following major component parts (Figure 1):

1. halogen lamp as a light source;
2. monochromator for extracting the spectral range of the required wavelengths;
3. cell compartment serving for containing the samples and calibration solutions;
4. detector for registration of the light and conversion of it into electrical signal;
5. electronics providing for performance of the measurements and control of the instrument operation;
6. digital indicator (display) for displaying the measurement result and auxiliary information.

4.2. Principle of Operation

The principle of operation of the photometer is based on comparison of the luminous flux Φ_0 having passed through the blank solution (reference solution, with reference to which the measurement is to be performed) and luminous flux Φ having passed through the test medium.

The luminous fluxes Φ_0 and Φ are converted into the electrical signals I_0 and I by the photoelectric receiver. Also, the signal from a non-illuminated receiver I_r is measured. According to the values of these signals, the spectrophotometer microprocessor calculates and displays the measurement result in terms of transmittance, absorbancy or concentration depending on the measurement mode chosen.

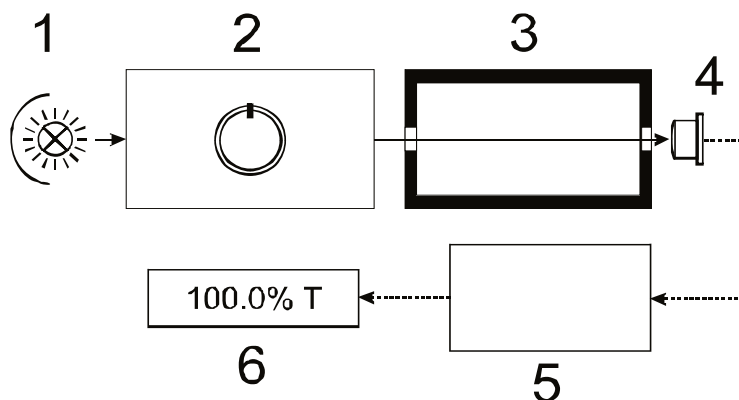


Figure 1 - Functional diagram of the spectrophotometer

4.3. Formulae to be Used for Calculating and Processing the Measurement Results

The transmittance τ of the test solution is determined as ratios of fluxes or signals from the formulae:

$$\tau = \frac{\Phi}{\Phi_0} = \frac{I - I_T}{I_0 - I_T}.$$

Transmittance in per cent **T**:

$$T = \tau * 100\%.$$

Absorbancy **A**:

$$A = \lg \frac{1}{\tau} = \lg \frac{100\%}{T}.$$

Concentration **C** according to the coefficient **F** to be entered:

$$Cx = Ax * K + B.$$

The calculation of the concentration according to the quadratic dependence in the PC software delivered together with the instrument.

5. Operating Conditions

- Ambient air temperature, °C 10 to 35
- Relative air humidity at a temperature of 25°C, % 20 to 80;
- Atmosphere pressure, kPa 84 to 106;
- Supply voltage, V 220±22, at a frequency of 50 Hz;
- The room shall be provided with the protective earthing (neutralling) system;
- The content of aggressive gases, vapours of acids, alkalis and dust in the room air shall be within the limits of sanitary norms established by the regulations in force;
- In the room, there shall be no equipment creating the vibration at the place of installation of the spectrophotometer as well as sources of electric and magnetic fields.

6. Safety Precautions

This spectrophotometer conforms to GOST R 51350-99 "Safety requirements for electrical equipment for measurement, control and laboratory use. General requirements" and GOST R 52319-05 "Safety requirements for electrical equipment for measurement, control and laboratory use" standards. As regards the electromagnetic compatibility, the instrument comply with the requirements GOST R 51522 "Electromagnetic compatibility of technical equipment. Electrical equipment for measurement, control and laboratory use. Requirements and test methods"

To ensure the safe working conditions for the servicing personnel, the following guidelines shall be adhered to:

- The persons allowed to work with the spectrophotometer shall pass the examination in electric safety and know the spectrophotometer operating instructions within the scope of this Operating Manual;
- Prior to starting the work, the spectrophotometer shall be earthed (connected to the neutral);
- All the installation works and replacement of bulbs shall be performed by a specialist on the spectrophotometer disconnected from the electric mains;
- Some chemical reagents used in the spectrophotometry are caustic and/or flammable and the samples can be radioactive, toxic or potentially contagious. Care shall be taken when performing the laboratory procedures with these chemical reagents.

7. Pre-Operation

- After unpacking the spectrophotometer, check the completeness of its delivery according to the list.
- Install the spectrophotometer in a convenient place protected against direct sunlight.
- To achieve the best metrological characteristics of the spectrophotometer, it shall be installed as far from any magnetic and electric fields or electric appliances generating high-frequency fields as possible.

8. Working Procedure

8.1. General Provisions concerning the Measurements

- The cells of the same working length to be used for measurements, shall have the same absorbancy and the same transmittance when being filled with the same solution;
- Every time prior to measurement, the working surfaces of the cells shall be wiped thoroughly with alcohol-ether mixture or other liquids leaving no traces on the glass;
- When placing the cells into the cell holder, never touch the working surfaces (below the liquid level in the cell) with fingers;
- Presence of contaminants or solution drops on the working surfaces of the cell entails obtaining of incorrect measurement results;
- The liquid shall be poured into the cells to approximately 3/4 of the cell height; otherwise the liquid will streak at the corners;
- It is recommended to close the cells with lids.

Attention!

1. All the measurements shall be performed on the spectrophotometer with the cell compartment closed with the cover.
2. When working in the UV area ($\lambda < 340$ nm), use quartz cells.

8.2. Preparation of the Cells

8.2.1. Preparation of the cell with the blank solution

The blank solution (blank, reference solution) is the solution, with reference to which the measurements are performed.

Wash the cell with distilled water or solvent. Having filled the clean cell with distilled water or other solvent being blank solution, wipe the cell from outside with a piece of cloth to remove the fingerprints or drops of liquid. To remove dust, it is recommended to use a squirrel brush.

8.2.2. Preparation of the cell with the test solution

Wash the second clean cell from inside with a small quantity of the test solution for the analysis. Fill the cell with the test solution and wipe the cell with a piece of cloth from outside.

8.3. General Description of the Instrument

The general view of the spectrophotometer is presented in Figure below.



Figure 2 – Spectrophotometer PE-5400UV.

Key: 1 – cell movement knob; 2 – cell compartment cover; 3 – control panel.

8.4. Description of the Buttons and Operating Modes of the Spectrophotometer

8.4.1. Control panel buttons

The control panel of the spectrophotometer comprises the liquid-crystal graphic indicator and buttons, by means of which the instrument is controlled (Figure 3).

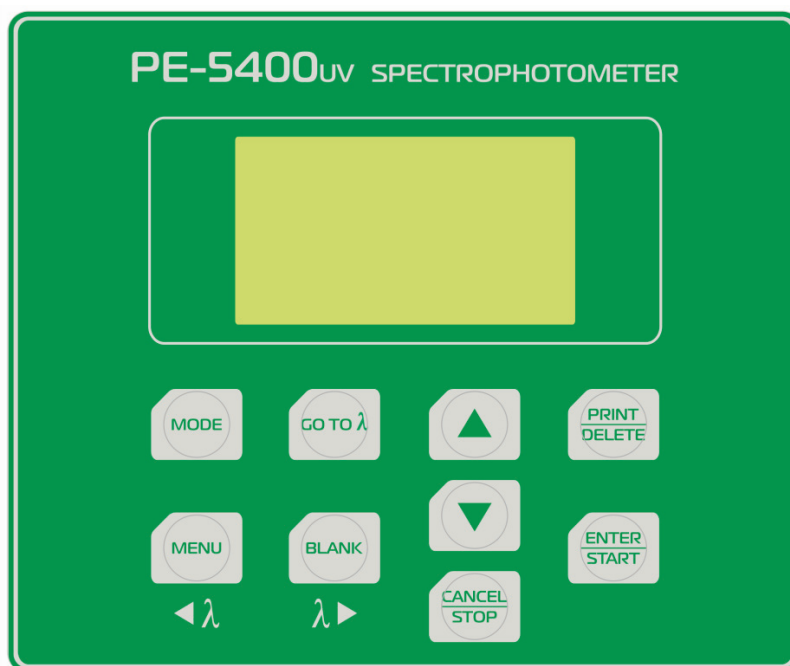


Figure 3 – Control panel of the spectrophotometer PE-5400UV.

Functions of the control buttons

- **MODE:** choosing the operating mode;
- **MENU:** calling the menu of auxiliary functions;
- **GO TO λ :** calling the wavelength setting procedure;
- **BLANK:** zero calibration (setting 0.000 A/100.0%T);
- **PRINT/DELETE:** sending the current values λ , A and T to the PC through the serial port/operation with the table of measurement results;
- **ENTER/START:** confirmation of the choice/writing of the measurement result into the memory;
- **CANCEL/STOP:** cancellation of the selection/stopping the writing of the measurement result into

the memory;

- ▲, ▼ – increase (decrease)/choosing the values, functions.

There are four operating modes of the instrument:

- **A**: measuring the absorbancy;
- **T**: measuring the transmittance, %;
- **C**: determining the concentration C;
- **F**: entering the coefficients of the coefficients of the calculation equation (K and B).

The required mode is chosen by pressing successively the **MODE** button. The current mode is indicated by illuminating the corresponding lettering in the bottom line of the display (Figure 5).

8.5. Switching on the Spectrophotometer

Make sure that there is nothing obstructing the light beam in the cell compartment and the cell compartment cover is closed. Switch on the spectrophotometer by means of the main switch located on the rear panel of the instrument. The audible signal will be heard and the self-diagnostics progress will be indicated on the display (Figure 5). The procedure lasts about 50 seconds. When it is finished, the wording “Warming Up... Any key to Skip” appears on the display and the time remaining to the completion of the warming-up (Figure 6). On expiration of the warming-up time or on pressing any button, the wording “Wait...” appears on the display; during this time the instrument restores the settings of the wavelength and measurement mode, which were effective at the moment of switching-off. Then the spectrophotometer goes to the measurement mode and performs automatically the zeroing procedure (0,000 A/100,0 %T).



Figure 4 – Transmittance measurement mode



Figure 5 – Self-diagnostics procedure progress

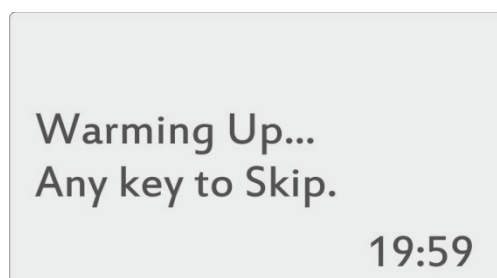


Figure 6 – Waiting for warming-up of the instrument

Note:

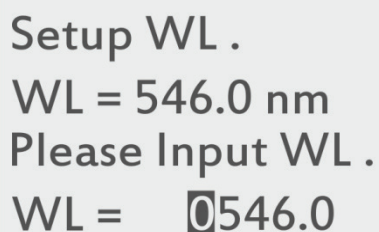
- The instrument requires 20 minutes from the moment of switching on to warm up. Should it be necessary to start the work quickly, the waiting for warming-up can be interrupted by pressing any button, but it should be noted that the insufficiently warmed up instrument may not ensure completely the declared metrological characteristics.
- During the self-diagnostics, the cell compartment of the instrument shall be empty. During this time, the cell compartment cover should not be opened.

8.6. Setting the Wavelength

To set the working wavelength, press the **GO TO λ** button. The window for entering a new wavelength value (Figure 8) will appear on the display.

The value shall be entered by changing the value of each character location of the number indicated in the bottom line of the display. The character location to be changed shall be highlighted by a white marker. To move the marker, use the **◀ λ (MENU)** and **λ ▶ (BLANK)** buttons. The value of the selected character location is changed by scrolling through the values by means of the **▲** and **▼** buttons.

To set the entered wavelength value, press the **ENTER/START** button. Once the wavelength has been changed, the instrument returns automatically to the measurement mode. To cancel the modifications and go back to the measurement mode, press the **CANCEL/STOP** button.



Setup WL .
WL = 546.0 nm
Please Input WL .
WL = 0546.0

Figure 7 – Setting the wavelength

Attention!

- After changing the wavelength, the instrument performs automatically the zeroing procedure; therefore it is recommended to place previously a cell with the blank solution to the working zone and close the cell compartment cover. Otherwise, it will be necessary to perform the zeroing later by means of the **BLANK** button.
- All measurements shall be performed after setting the wavelength and calibration of the instrument, with the cell compartment cover closed.

8.7. Calibration of the Zero Absorbancy

If the absorbancy zero value has not be calibrated at the set wavelength for any reasons , place a cell with the blank solution on the way of the light beam with the cell compartment cover closed and press the **BLANK** to set 0A/100%T. The value 0.000 shall be indicated on the display (Figure 9). If not so, repeat this step once again.

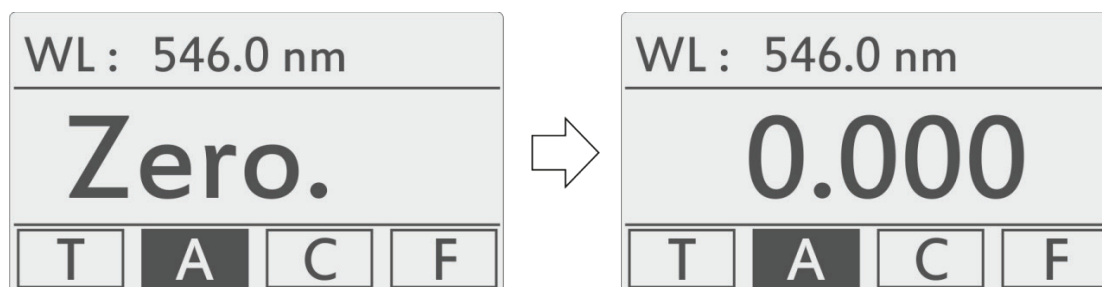


Figure 8 – Calibration of zero (0A/100%T).

8.8. Absorbancy Measurement Mode – A

Switch the instrument to the absorbancy measurement mode A using the **MODE** button.

In this mode, the display indicates the set wavelength value and the measured absorbancy value.

In this mode, the following functions are available:

- Measuring the absorbancy of the object placed in the cell compartment at the current wavelength;
- Changing the wavelength (item 8.6);
- Calibrating (zeroing) the absorbance (item 8.7);
- Writing the measured absorbancy values into the instrument memory in the form of a table of consecutive measurements (item 8.8.2);
- Output of the values of the measured absorbancy through the serial port of the computer;
- Switching to the instrument setting menu, in particular, to perform the compensation for the dark current (item 8.12.1).

8.8.1. Measuring the absorbancy of the object placed in the cell compartment

1. Place the cells with the test solution and the cell with the blank solution into the cell holder nests. Close the cell compartment cover.
2. Without opening the cell compartment, move the cell with the blank solution to the working zone using a knob for moving the cell holder.
3. If necessary, set the wavelength. In this case, the following operation can be skipped, because the zeroing is performed automatically when setting the wavelength.
4. Calibrate the zero absorbancy value to the blank solution using the **BLANK** button. If the displayed value differs from 0.000 by more than 0.001, repeat the zeroing.
5. Without opening the cell compartment, move the cell with the test solution to the working zone using a knob for moving the cell holder. Fix the displayed absorbancy value.

Note: The cell compartment has four nests that make it possible to perform simultaneously the measurement of one cell with the blank solution and up to three cells with the test solutions.

6. Open the cell compartment cover and remove the cells with the sample and the cell with the blank solution.
7. Should it be necessary to test the same sample, but at another wavelength, repeat items 2-5 for each required wavelength.

Note:

- When performing the measurements, it is possible to switch the instrument to the measurement of the transmittance **T** by means of the **MODE** button. In this case, the instrument calibration will be preserved.
- When pressing the **PRINT/DELETE** buttons in the measurement mode, the current value of the quantity being measured is output to the personal computer through serial port.

8.8.2. Writing the absorbancy measurement values to the instrument memory.

The results of the performed measurements can be saved in the instrument non-volatile memory in the table form in the format: <measurement number>/<wavelength>/<absorbancy> (Figure 10).

1. Press the **ENTER/START** button in the measurement mode. The table in the format <measurement number>/<wavelength>/<absorbancy>.
2. Each next time the **ENTER/START** is pressed, the line with the current value of the quantity being measured will be written into the instrument memory and entered into the table. Once the screen is filled completely, the table is scrolled automatically to the last written value. To scroll the screen manually, use the **▲** and **▼** buttons.

3. When pressing the **PRINT/DELETE** button, the menu of operations with the table is displayed (Figure 11).

546.0 nm		0.000 Abs
No.	WL.	Abs
1	546.0	0.000
2	546.0	0.010
3	546.0	0.150
4	546.0	0.235

Figure 9 – Measurement table

In this menu, the following commands are available:

- **PRINT:** output of the whole table to the personal computer through the serial port.
Attention! After being output, all the data are deleted completely from the table and instrument memory.
- **DELETE:** complete deletion of data from the table and instrument memory.
- **CANCEL:** close the menu for working with the table.

Print/Delete?

☒ Print

☐ Delete

☐ Cancel

Figure 10 – Menu of operations with the data table

Choose the required command using the ▲ and ▼ buttons and press the **ENTER/START** button.

4. To go back to the measurement mode, press the **CANCEL/STOP** button.

Note:

- The measurement results are saved in the instrument non-volatile memory and will be available upon switching on the instrument next time.
- To write the results in each mode, a separate table is used. The total data memory capacity for all the tables is 200 records.

8.9. Transmittance measurement mode – T

Switch the instrument to the transmittance measurement mode T using the **MODE** button.

The work in this mode is fully similar to that in the absorbancy measurement mode (item 8.8), but the indication 100.0%T will appear instead of the zero value 0.000 A.

8.10. Measurement Mode with Calculating the Concentration according to Reference Standard – C

Attention! The calibration equation used in this instrument and similar ones is a concentration vs absorbancy dependence C(A) instead of the absorbancy vs concentration dependence A(C) adopted in normative documents.

1. Set the required wavelength (item 8.6).
2. If necessary, calibrate the instrument with the cell containing the blank solution along the beam (item 8.7).
3. Switch the instrument to the mode C using the **MODE** button. The display will contain the menu for working with graduations (Figure 12).

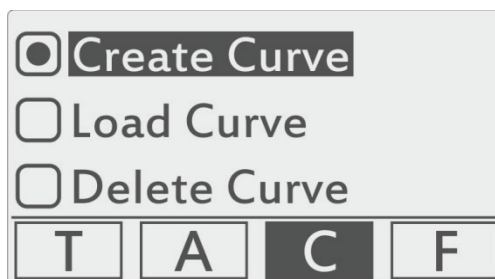


Figure 11 – Menu for working with curves

In this menu, the following functions are available:

- Create Curve (item 8.10.1);
- Load Curve (item 8.10.2);
- Delete Curve (item 8.10.3);

Attention! The work in this mode is performed at the previously set wavelength; the instrument shall be calibrated. In the working zone of the instrument, there shall be a cell with the blank solution.

To exit this mode, use the **MODE** button.

8.10.1. Create Curve

This mode makes it possible to construct a calibration curve using the reference solutions (from two to nine reference standards).

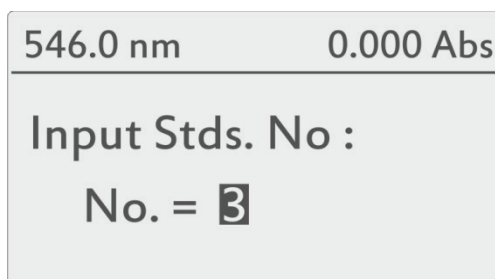


Figure 12 – Entering the number of reference standards

1. Place the marker to the menu item “Create Curve” using the ▲ and ▼ buttons and press the **ENTER/START** button; the field for entering the number of reference standards to be used for graduation will appear on the display.
2. Set the required number of standards for graduation using the ▲ and ▼ buttons and press the **ENTER/START** button.
3. The field for entering the concentration of the consecutive standard (Figure 14) will appear on the display. The cursor is on the first zero of the numerical value of the concentration consisting of six character locations.
4. Use the ▲ and ▼ scroll buttons for setting the first digit (from 0 to 9 or decimal separator) and press the **ENTER/START** button to confirm the selection, following which the cursor will move automatically to the next position.
5. The values of positions from two up to six can be also set in the range from 0 to 9 and include a decimal separator. Assign the values to these positions in the same way as for the first one.
6. Upon entering the values of all the six character locations press the **ENTER/START** button. The instrument will measure the current sample and go to the procedure of entering the values of concentrations of the next reference standard.
7. **Note:** At this moment, the spike solution shall be fed to the working zone and the cell compartment cover shall be closed. Before this, the blank solution shall be in the working zone. At the top right corner of the display, the current value of the absorbancy is indicated; if it differs from 0.000, per-

form the zeroing by pressing the **BLANK** button without exiting the current mode.

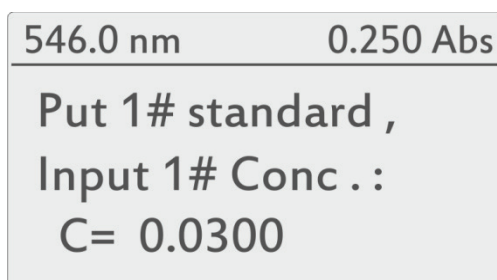


Figure 13 – Entering the reference standard concentration

8. Repeat the operations 3-6 for all the reference standards taken for performing the graduation.

Attention! Should different concentration values be entered when the solution remains the same, the instrument will give an audible error signal and go back to the menu for working with curves.

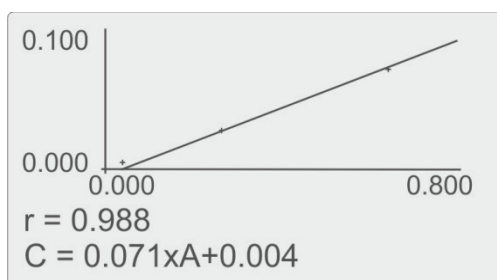


Figure 14 – Construction of the curve

9. After entering the concentrations and measuring the entered number of samples, the calibration curve (Figure 15) will be constructed, the coefficient of correlation r and calibration equation, which is saved in the instrument memory and will be available in the mode "Load Curve" (item 8.10.2).
10. When pressing the **ENTER/START** button again, the instrument starts measurements with calculation of the concentration on the basis of the graduation performed. The table in the format <measurement number>/<wavelength>/<concentration>. Now place the blank solution and working samples into the cell compartment and proceed to the measurement of them. The current absorbancy value is indicated in the top right corner of the display. If necessary, perform the zeroing of the instrument by pressing the **BLANK** button with the blank solution placed without exiting the current mode.
11. When placing the sample to be measured into the working zone and following pressing of the **ENTER/START** button, the calculated concentration value is added to the table and written to the instrument memory.
12. To close the table and go back to the measurement mode C, press the **CANCEL/STOP** button.

Note: The functions of working with the table are the identical for all the measurement modes and described in detail in item 8.8.2.

8.10.2. Load Curve

The non-volatile memory of the instrument cab contain up to 200 created graduations.

Attention! the work in this mode shall be performed after setting the wavelength and calibration of the instrument, with the cell compartment cover closed.

To work with the previously created curve switch the instrument to the mode C by pressing the **MODE** button and select the item "Load Curve".

1. Place the marker to the menu item "Load Curve" using the **▲** and **▼** buttons and press **ENTER/START** button; the table of equations of the curves stored in the instrument memory will ap-

pear on the display.

2. Choose the required curve by means of the ▲ and ▼ buttons and press the **ENTER/START** button.
3. The calibration curve (Figure 15) will be constructed on the screen; the coefficient of correlation r and calibration equation will be indicated.
4. The further work is fully similar to that described in the previous item (8.10.1) starting from the operation 9.

8.10.3. Delete Curve

1. Place the marker to the menu item "Delete Curve" using the ▲ and ▼ buttons and press **ENTER/START** button; the table of equations of the curves stored in the instrument memory will appear on the display.
2. Choose the curve to be deleted using the ▲ and ▼ buttons and press the **ENTER/START** button. The request for confirmation of the deletion will appear on the display.
3. Choose the confirmation of the deletion using the ▲ and ▼ buttons and press the **ENTER/START** button. The curve will be deleted and the table of equations of the curves remaining in the instrument memory will appear on the display.
4. To close the table and go back to the measurement mode C, press the **CANCEL/STOP** button.

8.11. Measurement Mode with Calculating the Concentration according to Entered Coefficients – F

Switch the instrument to the measurement mode F using the **MODE** button. The menu for working with the coefficients will appear on the display (Figure 16).

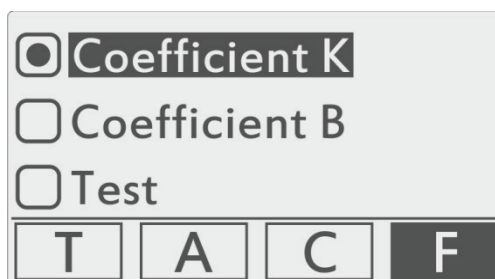


Figure 15 – Menu for working with coefficients

In this mode, the following functions are available:

- coefficient K;
- coefficient B;
- measurement.

8.11.1. Coefficient K;

The coefficient K determines the slope of the calibration curve.

1. Place the marker to the menu item Coefficient K using the ▲ and ▼ buttons and press the **ENTER/START** button; the field for entering the values of the coefficient K (Figure 17) will appear on the display.

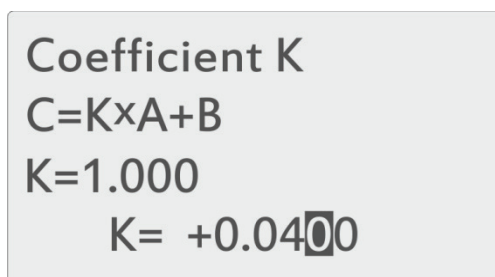


Figure 16 – Entering the coefficient K

2. The value consists of seven character locations, the first of which determines the sign of the coefficient. Use the ▲ and ▼ scrolling buttons to set the values and press the **ENTER/START** button to confirm the selection and move the cursor to the next position.
3. Upon entering all the character places press the **ENTER/START** button once again. The instrument will write the entered value into the memory and go back to the menu for working with coefficients. If necessary, correct the value of the coefficient K and repeat the operations 1-2.
4. To interrupt the operation of entering the coefficient value can be interrupted with cancelling the modifications made and go back to the menu for working with coefficients, press the **CANCEL/STOP** button.

8.11.2. Coefficient B

The coefficient B determines the displacement of the calibration curve relatively to zero. The coefficient B should be entered similarly to that of the coefficient K (item 8.11.1) by means of the menu item "Coefficient B".

8.11.3. Measuring

The measurements with calculating the concentrations according to the set coefficients shall be performed as follows.

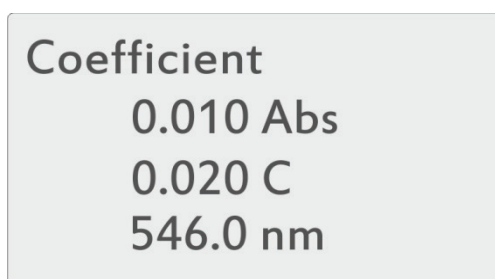


Figure 17 – Measurement with calculating the concentrations according to the coefficients

Place the marker to the menu item "Measurement" by means of the ▲ and ▼ buttons and press the **ENTER/START** button; the current value of the absorbancy, calculated concentration value and set wavelength value are indicated on the display (Figure 18). As the absorbancy of the sample in the cell compartment varies, the concentration value is recalculated automatically. The obtained curve can be used for determining the concentration of unknown samples. To do this, it is necessary to move the cell with the test solution to the working zone.

The measured concentration values can be written into the instrument memory with the use of the measurement table.

1. To output the measured concentration values in the table form, press the **ENTER/START** button; the measurement table will appear on the display.
2. Each next time the **ENTER/START** button is pressed, the current sample concentration will be written into the instrument memory and entered into the measurement table.

Note: The functions of working with the table are the identical for all the measurement modes and described in detail in item 8.8.2.

3. To exit the measurement table measurement mode and go back to the menu for working with coefficients, press consecutively the **CANCEL/STOP** button.

Note: To control the correctness of setting of the 0.000A/100.0%T parameter, place the cell with the blank solution on the way of the light beam and make sure that the absorbancy of the blank solution is equal to zero. Otherwise, press the **BLANK** button. The instrument will be calibrated without exiting the tabular mode.

8.12. Menu of Auxiliary Functions

This menu ensures the access to a number of service functions and system information (Figure 19). To

call the menu of auxiliary functions in the mode T or A, press the **MENU** button.

The menu contains the following items:

- The energy is a system-related diagnostic function intended for using by the service personnel.
- UV-lamp: manual switching-on/off of the deuterium lamp.
- Dark current: dark current compensation procedure (item 8.12.1 below).
- Calibrating WL: procedure of the wavelength scale calibration (item 8.12.2 below).
- Reset of the settings: procedure of restoration of the factory system settings (item 8.12.4 below).
- Version: displaying the numbers of the instrument soft- and hardware (item 8.12.4 below).



Figure 18 – Menu of auxiliary functions

8.12.1. Dark current

Although the instrument provides for automatic dark current compensation when setting the wavelength, it is recommended to perform the dark current measurement and compensation for it after warming-up completely the instrument is case of change of the environment conditions and prior to performing the most important measurements.

Make sure that the cell compartment cover is closed. Place the marker to the menu item “Dark Current” using the ▲ and ▼ buttons and press the **ENTER/START** buttons, the wording “Dark Current...” and then the message “Dark Current OK! ESC to Return” appears on the display (Figure 20). Press the **CANCEL/STOP** button to go back to the menu of auxiliary functions. Press the **CANCEL/STOP** button once again to go back to the measurement mode. After returning to the measurement mode, the zero absorbancy calibration will be performed automatically; therefore the blank solution must be present in the cell compartment.

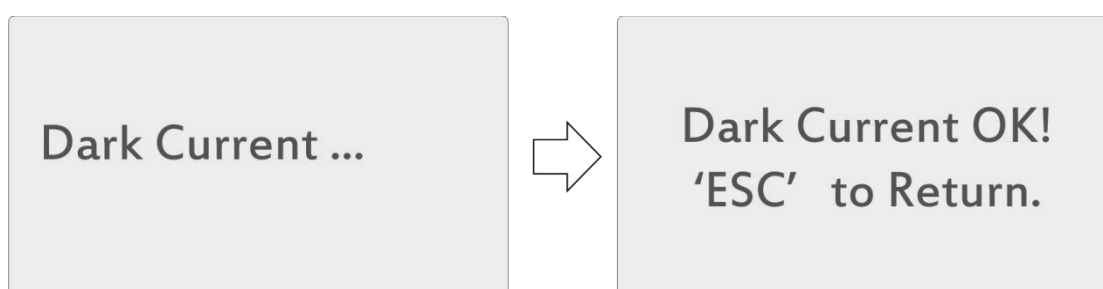


Figure 19 – Dark current compensation

8.12.2. Calibrating WL

This wavelength scale calibration procedure sets automatically the wavelength reference point on the basis of the autocollimation effect.

Make sure that the cell compartment cover is closed. Place the marker to the menu item “Calibrating WL” using the ▲ and ▼ buttons and press the **ENTER/START** button; the message “Calibrating WL...” and then, after expiration of some period of time, the message WL. OK! ESC to Return” appears on the display (Figure 20). Press the **CANCEL/STOP** button again to go back to the menu of auxiliary functions. Press the **CANCEL/STOP** button once again to go back to the measurement mode.

Note: after performing the calibration, the wavelength of 546.0 nm is set automatically.

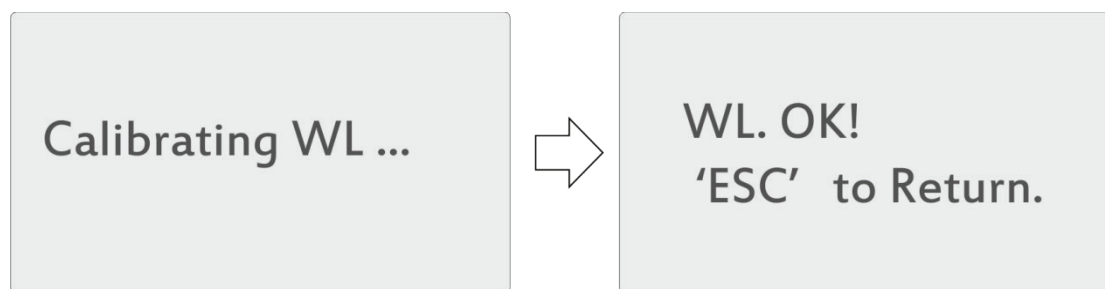


Figure 20 – Calibration of wavelengths

8.12.3. Resetting the Settings

Place the marker to the menu item “Resetting the Settings” using the ▲ and ▼ buttons and press the **ENTER/START** button; the menu for confirmation of resetting the settings will appear on the display.

Place the marker to the menu item “Yes” using the ▲ and ▼ buttons and press the **ENTER/START** button. The spectrophotometer will restore the factory settings; then it will be restarted and switched to the self-diagnostics mode. After completion of the self-diagnostics, the instrument will be switched to the measurement mode.

8.12.4. Version

Place the marker to the menu item “Version” using the ▲ and ▼ buttons and press the **ENTER/START** button; the numbers of versions of the instrument soft- and hardware will be indicated on the display. To go back to the previous menu, press any button.

8.13. Data Output and Processing

8.13.1. Personal computer and software

The spectrophotometer can be connected to the USB port of the personal computer to work with the special software. This software runs under the control of the Windows XP/Vista/7/8/10 operating systems and implements various additional functions.

The XL5x00 Program provides for the input of the results of the measurements from the spectrophotometer into the cells of the open Microsoft Excel™ book after pressing the **PRINT/DELETE** button on the instrument. Thus, the user can program the implementation of the own algorithms of processing the measurement results and form of their representation by means of the Excel™ tools.

QA5400 is the quantitative analysis program. It provides for controlling the spectrophotometer, obtaining the data from the instrument and their further processing. The program makes it possible to:

- create the graduations according to several parallel measurements of a series of reference standards with automatic calculation of the calibration equation coefficients by means of one of the three kinds of approximation: linear passing through the origin, linear and quadratic (parabolic);
- use the direct input of the known values of the calibration equation coefficients obtained previously;
- when constructing the curve and performing the analysis take into account the results of the check experiment where the value of its absorbancy is subtracted from every value of the absorbancy of the reference standards;
- calculate automatically the following calibration parameters: square of the coefficient correlation of the calibration equation, maximum value of the mean square deviation of the calculated value in per cent and maximum error of the calculated value for all the reference standards;
- perform the analysis on one specified wavelength on the basis of the previously performed graduation; here one file can contain the results of analysis of up to 20 samples for up to 10 parallel measurements for every sample;
- calculate automatically the concentration of every sample as well as average concentration value

and convergence in per cent for parallel measurements when performing the analysis of respective samples;

- print out the calibration and analysis reports;
- save the curves created previously to a file and load them for performing the analysis;
- save the analysis results to the file and load them from the file;
- export the tables of the calibration and analysis data to the Microsoft Excel™ format.

Kin5400 is the kinetic analysis program. Measuring the sample at a single specified wavelength with the specified period within the specified time interval. The delay of the beginning of the measurement for certain time can be set. When specifying the measurement parameters, the coefficients for conversion of absorbancy to concentration can be entered.

SC5400 is the scanning program delivered as an option. It provides for control of the spectrophotometer, enables scanning of absorbancy or transmittance of the samples by the wavelength within the wavelength range to be specified with the specified scanning pitch, finding of the peaks on the spectra obtained, saving and loading of the table of peaks and table of scanning results as well as printing of the calibration reports.

The program is protected by means of the Guardant Sign hardware key included in the scope of delivery. Without using the key, the program works in the reduced functionality mode (demo mode).



Figure 21 – Rear panel of the instrument

Key: 1 – main switch; 2 – receptacle for connection the power cord; 3 – interface for connecting the printer; 4 – USB-port connector

9. Maintenance and Checking the Technical Conditions

9.1. Replacement of the Halogen Bulb

1. Switch off the spectrophotometer and disconnect the power cord from the electric mains.
2. Unscrew and remove the knob for switching over the cells in the cell holder.
3. Unscrew four screws fastening the spectrophotometer cabinet and remove the plastic cabinet. Hold back the cell compartment cover.
4. Disconnect the flat connector from the system board on the instrument.
5. Unscrew four screws and remove the protective housing of the illuminator.
6. Replace the halogen bulb: Remove the bulb from the holder and insert a new bulb into the holder nests (Figure 23).
7. Switch on the power supply of the instrument and make sure that the image of the bulb filament is projected to the input slit of the monochromator. If not so, adjust the bulb position.

Attention!

- Never touch the bulb surface with hands; the bulb should be held using a napkin or a piece of cloth.
 - The halogen bulb leads have no polarity.
8. Reassemble the instrument by proceeding in the reverse order (items 5-1). When re-installing the protective housing of the illuminator, make sure that the wires pass through a special recess and are not pressed down by the housing.

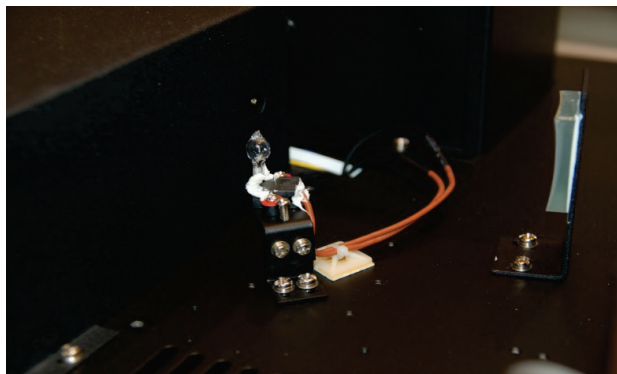


Figure 22 – Fixing the halogen bulb

9.2. Checking the Accuracy of the Wavelength Setting

Usually, the spectrophotometer retains the wavelength setting; however, provided spectrophotometer is exposed to strong shock or abuse, its calibration according of the wavelength shall be checked.

Should the spectrophotometer calibration be accurate, the minimum transmittance (maximum absorbancy) shall be located in the neighbourhood of ± 1 nm from the certified ratings of the filters. It should be noted that the transmittance value itself is not significant, because you are only finding the wavelength, onto which the minimum transmittance (maximum absorbancy) falls.

The PS-7 filter included in the scope of delivery has two clear absorbancy peaks near 431 nm and 585.5 nm.

1. Switch on the spectrophotometer and let it warm up for 20 minutes.
2. Choose the transmittance measurement mode T using the **MODE** button.
3. Set the wavelength of 428 nm.
4. Place an adapter plug for the 10 mm cell and reference filter included in the scope of delivery of the spectrophotometer PE-5400UV into the cell holder. Insert the PS-7 filter into the adapter plug. Close the cell compartment cover.
5. Move the empty nest of the cell holder into the working zone by means of the knob for moving the cell holder without opening the cell compartment.
6. Set 100.0%T by pressing the **BLANK** button. Wait for a few seconds until the transmittance value appears on the indicator. The reading shall be $100.0 \pm 0.2\%$ T.
7. Move the nest of the cell holder with the inserted PS-7 filter into the working zone by means of the knob for moving the cell holder without opening the cell compartment. Write down the transmittance value displayed on the indicator.
8. Increase the set wavelength value by 1 nm and repeat items 5-7 until the wavelength reaches 436 nm.
9. Check the transmittance values fixed. The minimum value shall differ from the certified rating by not more than 1 nm. The accuracy of the wavelength setting for PE-5400UV model is 1 nm.
10. If you want to check additionally the accuracy of the wavelength setting in the middle of the range, set the wavelength of 582 nm.
11. Set the wavelength with increment of 1 nm and repeat items 5-7 until the wavelength reaches 589 nm.
12. Check the transmittance values fixed. The minimum value shall correspond to the interval between 583.5 and 587.5 nm.
13. The positions of maxima of the absorption bands of the PS-7 filter included in the complete set of light filters delivered together with this instrument are given in the report attached.

Attention! If the error of the wavelength setting exceeds 1 nm, it is recommended to contact the manufacturer's service centre.

9.3. Checking the Photometric Accuracy

The photometric characteristics of the spectrophotometer can be checked by means of the reference light filters on the indicated wavelengths (the values are given in the attached report) as well as by means of the KC-105 complete set of light filters or similar verified sets.

1. Switch on the spectrophotometer by pressing the (I/O) button located on the rear panel of the spectrophotometer. Let the spectrophotometer warm up for 20 minutes.
2. Choose the operating mode T (determining the transmittance) by pressing the MODE selection buttons.
3. Place the adapter plug with the inserted reference light filter in one of the nests of the cell holder. The other nests of the cell holders remain empty. Close the cell compartment cover.
4. Set the wavelength corresponding to the light filter characteristics.
5. Move the empty nest of the cell holder to the working zone by means of knob for moving the cell holder. Set 100.0% T by means of the **BLANK** button. Wait for a few seconds until the transmittance value %T appears on the indicator. The reading shall be $100 \pm 0.2\%$. If not so, repeat this step once again.
6. Move the adapter plug with the reference light filter by means of the knob for moving the cell holder without opening the cell compartment. Write down the transmittance value displayed on the indicator.
7. Repeat items 5-6 at least three times. Determine the average value. The error of determining the transmittance in the visible range shall not exceed $\pm 0.5\%$.
8. Repeat items 5-7 for the light filters included in the set with other transmittance values.
9. If necessary, check the photometric accuracy at other wavelength, repeat items 3-8 while setting other values of the wavelengths.
10. If the absolute error of the spectrophotometer exceeds 0.5% when measuring the direct transmittance, it is recommended to perform the following operations:
 - Check the settings of 0.000A and 100.0%T;
 - Wipe the test filters clean;
 - Replace the halogen bulb.

Note: The reference light filters “7%”, “50%” and “90%” are included in the scope of delivery of the spectrophotometer. The values of light filter parameters written in the report attached to the instrument, they are not real values of the light filter parameters, but the values taken on this spectrophotometer prior to performing the primary verification; they are valid for this spectrophotometer only.

- **Attention!** The symbols “7%”, “50%” and “90%” applied to the frames of the light filters are not measured values of their transmittance. The measurement results obtained on the filters shall be compared with the values given in the report.

10. Troubleshooting

Table 3

Trouble	Possible problem	Remedy
The spectrophotometer cannot be switched on	The power cord is not connected to the mains or the contact in the receptacle for connecting the power cord to the instrument is poor	Connect the spectrophotometer to the electric mains; check the reliability of the connections
	The fuse has blown out or the electric element is faulty	Call a qualified engineer
The value of 100%T (0.000 A) is not set Drift of zero and increased dispersion in the readings	The light beam is shielded	Check the cell position in the cell compartment
	The halogen bulb is faulty or installed inaccurately	Check the installation of the bulb or replace it. See item 9.1 of this Manual
	The instrument is faulty	Contact the service department
Drift of zero and increased dispersion in the readings in the UV range only	The deuterium bulb is faulty	Replace the deuterium bulb
	The glass cells are used	Use the quartz cells
The message "Dark Current Error" appears	The compartment for samples is open	Close the cell compartment cover
Incorrect readings	Insufficient sample volume	Fill the cell with larger sample volume
	The cells are prepared poorly	See item 8.2 of this Manual
	Sample evaporation	Prepare the samples away from the spectrophotometer, use ventilation Close the cells with covers
	Bubbles or particles in the solution	Check the solution preparation and analysis procedure
	The wavelength is set incorrectly	Check the procedure of analysis and wavelength setting Proceed according to the methods described in this Manual
	Displacement of the cell holder relatively of the optical axis of the instrument	Correct the cell holder position in the cell compartment
	The instrument setting is distorted	Measure the reference light filters; should there be discrepancies, call the service department

11. Warranty

Ecohim Co. Ltd. guarantees the compliance of the spectrophotometer with the requirements stipulated in item 2.3 hereof provided the consumer adheres to the operation, transportation and storage conditions.

The guaranteed service life of the spectrophotometer is 36 months from the date of shipment to the consumer as determined by the date of the bill of lading or, in the absence of the latter, from the date of verification.

The guaranteed service life of the halogen and deuterium bulbs is 6 months

12. Claims Information

In case of revealing any faults within the guaranteed service life or incompleteness when unpacking the spectrophotometer, the consumer shall send the claim report to the manufacturer's address (item 2.1).

No claims for spectrophotometer may be submitted:

- on expiration of the warranty period;
- if the consumer has broken the operation, storage and transportation rules provided in the operating documentation.

13. Metrological Supervision

PE-5400UV spectrophotometer is subject to regular verification in accordance with document "PE Series Spectrophotometers (models PE-5300VI, PE-5300UV, PE-5400VI, and PE-5400UV). Verification methods VM-242-1033 -2010".

The main verifying instrument is the KS-105 complete set of light filters, Technical Specifications TU 4434-138-07502348-2001. The verification interval is 1 year.

14. Certificate of Acceptance

PE-5400UV spectrophotometer, serial number **54K4UV**_____ has been verified in accordance with the Technical Specifications TU 9443-001-5627822-2009, statutory requirements of national standards and current technical documentation, and recognised to be ready for service.

The acceptance has been performed by _____

Stamp of the Technical
Control Department

Production date _____

Notes

Notes