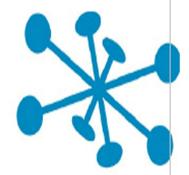


NanoSPR6 User's manual





rev.04-2019

NanoSPR 6

This document is a manual for check-up and adjustment of the apparatus that includes device with program package and it can serve as an operation manual provided that a user is sufficiently trained. While the software operation is expounded in the document **Program package NanoSPR 6**, this document refers to the device operation itself.

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Device description

Delivery set

- 1. Device.
- 2. Power source (220 V AC or 100...220 V AC).
- 3. PC connection USB cable.
- 4. Connection cable for subsidiary channel signal.
- 5. Installation CD on agreement with Customer.
- 6. Spare parts (variable set on customer's demand). Typical set includes:
 - F1-65 prism 1 piece;
 - K8-50 prism 1 piece;
 - Rubber gaskets 5 pieces;
 - Sensor chips 20 pieces;
 - Cell 2 pieces;
 - Immersion liquid 1 ml.

Additional components that can be included into the set:

If the device is completed with plunger pumps, the following components are included in the set:

- Plunger pump 2 pieces;
- Syringe 2 pieces;
 - Cable.

If the device has a sample thermostabilization system, then one of the included cells is thermally stabilized.

Attention: Program components provided on the installation disk are individual for your device exemplar.

Turning on the device and installing program package

To turn on the device, connect it to free USB port of the computer and to the power source. Turn on the computer and the device power source.

To install the program:

Insert the installation CD.

1. Unpack and install bridge drivers by running CP2101_Drivers.exe (for Windows XP) or CP210x_VCP_Win_XP_S2K3_Vista_7 (for Windows 7/8/10).

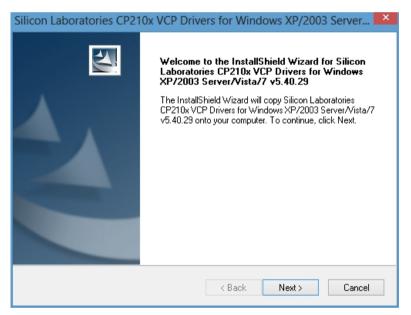


Figure 1USB Driver Installation Wizard

Silicon Laboratories CP210x VCP Drivers for Windows XP/2003 Server	×
License Agreement Please read the following license agreement carefully.	1
END-USER LICENSE AGREEMENT IMPORTANT: READ CAREFULLY BEFORE AGREEING TO TERMS SILICON LABORATORIES INC., SILICON LABORATORIES INTERNATIONAL PTE. LTD., AND THEIR AFFILIATES (COLLECTIVELY, "SILICON LABS") HAVE DEVELOPED CERTAIN MATERIALS (E.G., DEVELOPMENT TOOLS, EXAMPLE CODE, EMBEDDABLE CODE, DLLS, SOFTWARE/COMPUTER PROGRAMS AND OTHER THIRD PARTY PROPRIETARY MATERIALS ("LICENSED MATERIALS") THAT YOU MAY USE IN CONJUNCTION WITH SILICON LABS' MCU PRODUCTS. ANY USE OF THE LICENSED MATERIALS IS SUBJECT TO THIS END-USER LICENSE ACCEPTIVE THE THIRD ANY USE OF THE LICENSED MATERIALS OF OTHER O I accept the terms of the license agreement O I do not accept the terms of the license agreement	*
InstallShield	el

Figure 2Accept License Agreement

Silicon Laboratories CP210x VCP Drivers for Windows XP/2003 Server
Choose Destination Location Select folder where setup will install files.
Setup will install Silicon Laboratories CP210x VCP Drivers for Windows XP/2003 Server/Vista/7 v5.40.29 in the following folder.
To install to this folder, click Next. To install to a different folder, click Browse and select another folder.
Destination Folder c:\\MCU\CP210x\Windows_XP_S2K3_Vista_7 Browse
InstallShield Kack Next > Cancel

Figure 3Select Destination Folder



Figure 4Press Finish to begin Driver Update

🙈 Silico	on Laboratories CP210x USB to UART Bridge Driver Ins 💌
*	Silicon Laboratories Silicon Laboratories CP210x USB to UART Bridge
	Driver Version 6.5
	Install Cancel

Figure 5Press Install

Silicon Laboratories CP210x VCP Drivers for Windows XP/2003 Server/Vist		
	InstallShield Wizard Complete	
	🚜 Silicon Laboratories CP210x USB to UART Bridge Driver Ins 🗙	
	Scanning (Not Responding)	
4	(T Bridge	
	Driver Version 5.4.29	
	Please wait while system is scanned	
	Change Install Location Install Cancel	
	< Back Finish Cancel	

Figure 6Driver Installation may take a while

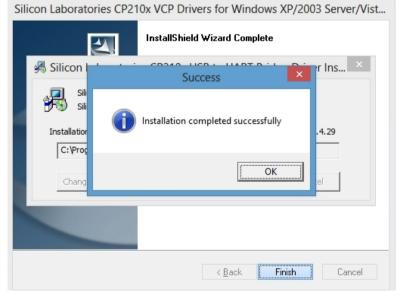


Figure 7Driver Installation is Complete

2. Install the device software by running SetupXXX.exe. During the installation enter the factory device number into the "Serial number" field (factory number is marked on the lower surface of the load-bearing slab of the device and specified on the installation disks).

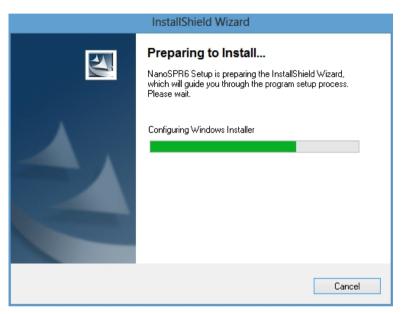


Figure 8NanoSPR Device Software Installation Wizard

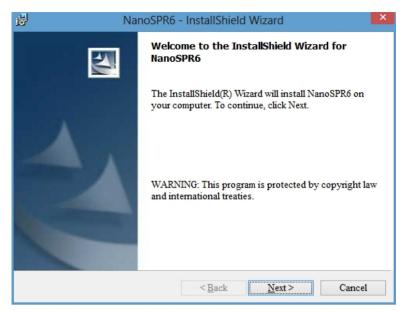


Figure 9Click Next

NanoSPR6	- InstallShield Wizard	×
Customer Information		44
Please enter your information.		
User Name:		
Lab		
Organization:		
Hewlett-Packard		
Serial Number: 055		
Install this application for:		
 Anyone who uses the 	is computer (all users)	
 Only for me (Lab) 		
InstallShield		
	< Back Next >	Cancel

Figure 10Provide User Name and Organization

ц <mark>е</mark>	NanoSPR6 - InstallShield Wizard	X
	tion Folder Text to install to this folder, or click Change to install to a different	N
	Install NanoSPR6 to: C:\Program Files (x86)\NanoSPR\NanoSPR6\ Change	
InstallShield	d <back next=""> Cancel</back>	

Figure 11Select Installation Folder

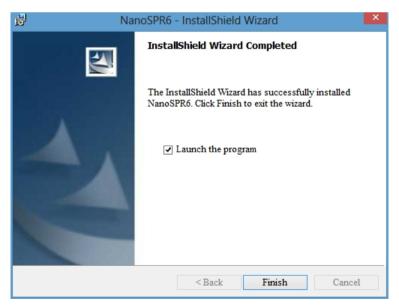


Figure 12Installation is Complete

3. **Start** the Plas06.exe program (start "as Administrator" in Windows 10, do it only once, during the initial setup, this way the program will be able to save communication port settings).

4. If a window "Found New Hardware Wizard" is shown upon the program start, choose

-Yes, this time only/Install from a list or specific location/

and specify CP2101 drivers location. Default location is C:/Cygnal/CP2101/win.

Possibly, it will be necessary to install these drivers more than once!

5. If a diagnostic window "Device not found" is shown, select the following menu items successively

Options/Set Options/Advanced/PC port = Auto/OK

While starting the program first time, the **Set Device** window will appear.

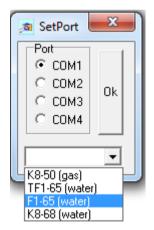


Figure 13Set Device

It is necessary to select the following options in this window:

- The port the device is connected to (usually COM3).
- The prism installed on the device.

The device is ready for operation.

Design

The device is built in Kretschmann geometry. The main element of the device is the retroreflecting measurement prism, which is installed on the rotating table. The rear prism face is made reflecting, and angle between top and rear faces of the prism equals to 90 degrees. The sensor chip (a glass slide with deposited thin Au layer) is installed on the top prism face using the immersion liquid. Properly, the surface plasmon resonance (SPR) phenomenon is observed in the sensor chip. A cell intended for injection of the investigated probe is pressed to the Au layer. To observe the SPR phenomenon, the intensity of the polarized light reflected by the Au layer is investigated as a function of angle of incidence of light on this layer (further – angle of incidence).

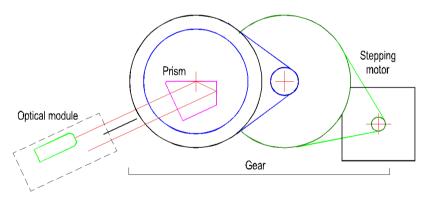
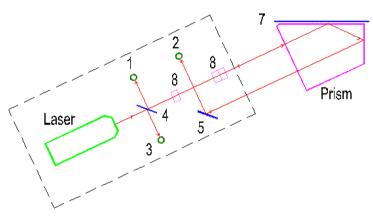


Figure 14Device mechanical diagram

The table with prism is made rotating to change the angle of incidence. The rotation is carried out on the axis, which approximately coincides with the glass slide surface. The rotation can be realized manually using the preset ring or programmatically by the controlled precision rotation system. Precision rotation system consists of stepping motor and doublestage reducer, which outlet axis the table with prism is installed in via the friction clutch.

The maximum operational angle scanning range amounts to 17 degrees.

The operational rotation direction is in the angle of incidence increase direction.



Optical arrangement

Figure 15Device optical block

A semiconductor laser is used as a light source in the device. Polarized light of the laser is split into two beams by beam splitting means 8. These two beams provide two-channel device operation mode. Beam in the first channel falls on the measurement prism and further on the sensor chip 7, passing the

transparent plate 4 that serves for take-off of the part of the beam energy on the photodiode 3 for laser power control. The point of incidence of beam on the chip coincides with the table rotation axis. The beam reflected from the chip falls on the photodiode 2 of the registration system after turning by 90-degree prism angle and turn mirror 5. The accepted optical arrangement provides design compactness and weak dependence of positions of light spot on the chip and on the registration system photodiode what is important for ensuring of accuracy of the SPR curve measurement.

The second channel arrangement differs from the first channel arrangement by absence of plate 4 and photodiodes 1 and 3.

The device allows to calculate absolute values of angle of incidence. Note that under normal incidence of light on the input prism face (the autocollimation position) angle of incidence equals to prism angle. Therefore, it is necessary to know the device autocollimation position for determination of the absolute values of angle of incidence. Photodiode 1, which the light reflected by the front prism face can fall in after reflecting from the glass plate 4, is used for determination of the autocollimation position. Maximum signal of the photodiode 1 (the calibration signal) is observed at the value of the angle near to the autocollimation. Difference of this value from the precise value (the correction) is measured by the manufacturer for every device exemplar and is stored in the file, which is included in the installation system supplied along with the device. Here positive value of the correction, e.g. 15, means that maximum calibration signal is observed when the beam reflected by the front prism face is deflected downward by 15 angular minutes relative to the autocollimation position.

The device can be completed with the following measurement prisms:



Figure 16 K8-50 prism

• K8-50 prism for operation in gaseous environment;



Figure 17F1-65 prism

• F1-65 prism for operation in aqueous environment.

Note: It is possible to find information about prisms by selecting the menu items sequentially: **Options/Advanced/Optics**.

Measurement cells

Two device application modes are provided:

1) Individual and practically independent 2 measurement channels with each of channels having own cell unit. Up to 2 independent probes can be analyzed in this mode.

2) The same 2 channels placed into one common cell. In this mode, one probe can be analyzed into 2 components, i.e. 2 tests of one probe are carried out simultaneously.

Device can be completed with single- or 2-channel cells, correspondingly.

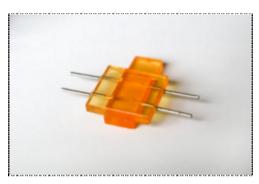
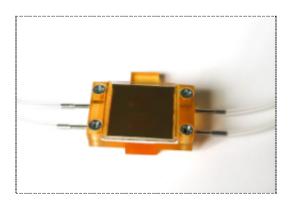
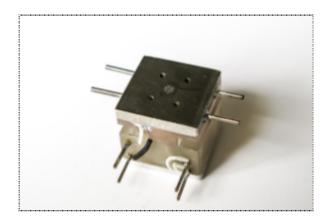


Figure 18Flow mode cuvette



Thermally stabilized cell

There are also two types of thermally stabilized cells: singlechannel and 2-channel. Both cells have a resistive heater and platinum thermal sensor. These cells provide regulation and constant temperature support for investigated liquid in the range from room temperature to 60° C.



Pumps



The device can be completed with NanoSPR-Pump 8-channel peristaltic pump, which provides liquid flow speed of 1-500 μ l/min per one channel.

Device operation

Checking the device

To check the device, install the selected prism and the sensor chip, and do not forget to use immersion liquid. If you work with liquids, install the cell and fill it with the required solution. Start the program and turn on the **Adjustment** mode by pressing the **Adjustment mode** button (or by selecting the **Tools/Adjustment mode** menu item). Laser beam intensity will increase and signals of all device channels will be visualized in the arisen window as horizontal color lines (even signals of the channels unused in the current device realization). Length of the lines is proportional to corresponding signal values.

Observe the light spot on the working layer through the cell while turning table with the prism using the angle preset ring. Brightness of the spot increases noticeably at certain rotation angle that corresponds to the SPR angle. The spot must be situated inside the cell not far from the center and it must not go out of the cell borders during the table rotation. The beam that came out of the prism must fall on the rotary mirror 5. Observe the position of the spot of the beam that came out of the prism on the rotary mirror 5 surface while turning table with the prism using angle preset ring (see <u>Optical arrangement</u>). The spot position has to be changed insignificantly.

If you have found the position corresponding to the SPR angle, decrease the angle of incidence by turning the preset ring clockwise on several degrees. Start the measurement by pressing the **Single measurement** button (or by selecting the **Measurement/Single** menu item). The measurement will start. Measurement results will be represented in the opened window entitled SPR1. You have to see the SPR curves; moreover, maximum signals must be above the window half height, but they must not exceed its boundaries.

If all of the above mentioned is fulfilled, the device is ready for operation.

Adjusting the device

If after Checking the device you are sure that device adjustment is wrong, you can try to rectify the situation. To do this, loosen the left screw of the optical block mount to the device plate and reach the adjustment position described in the Checking the device section by slightly turning the block. If all is set up correctly, but the signal is absent or very weak, it is possible that the working beam does not fall on the photodetector 2. In this case it is necessary to take off the lid by unfastening two screws at the bottom of the optical block and visually check whether the beam falls on the photodetector 2 after reflection from rotary mirror 5. If it is necessary, one can turn the support with the mirror 5 in the required direction.

Note: Displacement of any optical block elements (except the mirror 5) leads to impairment of the measurement of absolute angle values accuracy.

Changing the prism

To change the prism, turn off the fixation screw of the prism table and pull out the prism (along with the table).

Install and fix the table with a new prism. Check the device as described in the <u>Checking and adjusting the device</u> section.

Note: After you change the prism or change its position you have to carry out the device <u>Calibration</u> before starting the measurements in order to obtain accurate results.

Calibration

Calibration is the process of associating the angular sweep of the device to the built-in calibration reference point. Angular position of the reference point is located near from the position of normal incidence of light on the input prism face and it is known precisely. Calibration allows calculating absolute values of angles of incidence to a high accuracy.

Calibration should be carried out after changing the prism or any modification of its position or simply before starting important measurements.

Set the prism in the position required by the conditions of the experiment by turning the angle preset ring. Press the Device Calibration button (or select the Measurement/Calibration menu item). The device will start to operate in the calibration mode (for more details see **Device description. Design**). During the calibration the special mode for measurement of the reference point signal is established and the system position in the moment of the maximum reference point signal is determined and saved. The reference point signal is drawn in the SPR1 window, and the calculated position of its maximum is shown by the vertical line and the text is outputted, e.g. "Zero"=607 a.m., in the same place. It means that the angle of incidence of light on the input prism face equals zero when the prism rotation angle equals to 607 angular minutes. The passport correction for true angular reference point position is taken into account automatically. It should be noted that the maximum marker can visually be apart from the displayed signal maximum. It is because the accuracy of maximum position calculation is significantly higher than that of curve drawing.

Connection and cables

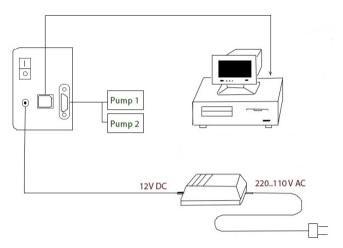


Figure 19 Device connection diagram

Connecting the thermo-cell

Devices equipped with thermo-heater support come with pair of connectors. Both connectors have four pins, note that each connector has only two wires attached. Also note, that wires are connected to different pins on connectors (one connector has wires attached to pins 2-3 and the other one to pins 1-3), see figure below.



Figure 20Thermo-heater connectors

This pin spacing on connectors is important and defines how to make connection to the thermo cell.

See the figure below where the heater and thermo-couple pins (on the thermo cell) are spaced differently.

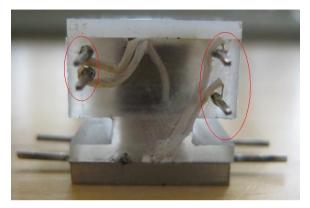


Figure 21Thermo cell pins

To connect thermo cell, match the device connector pins to the cell pins such that the spacing between pins on device side and cell side is the same.

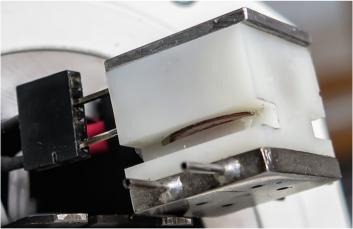


Figure 22Connecting the thermo cell

There is no polarity on heater and thermo-couple pins.

Program package NanoSPR 6

System requirements

Supported operating systems: Windows XP/Vista/7/8/10 (32/64 bit).

PC configuration for normal device and program operation:

CPU frequency – no less than 1000 MHz.

Standard COM port or USB (depends on device modification).

100 MB of free disk space. Mouse.

Note: Device will work at lower CPU frequencies but its operation stability decreases. If it is necessary to work on low-power PC, you can follow these instructions to reduce CPU load:

- Increase curve drawing speed by adjusting the Drawing acceleration element (highest possible value - 9) on the Single page in the Options window of the program.
- Increase measurements Period, s during measurements in Multiple mode.
- Increase Delay, s between measurements during measurements in Slope mode.

Measurement modes

Multiple mode 🕶

Single measurement – single-stage measurement.

Kinetic measurements:

- Multiple measurement reiterated measurement.
- Track mode measurement with tracking of the minimum.
- Slope measurement of signal intensity on the slope of the curve.
- Subsidiary channel subsidiary channel for the external signal measurement.
- Angle calibration.
- Stop measurement termination.
- Z Pause measurement pause/resumption.
- Adjustment adjustment of the instrument.

Single measurement 😾

Single measurement of the SPR curves within the angular range specified by the **Max angle, degree** option and in the angular coordinates specified by the **Absolute angle-Angle in air-Angle in glass** options. The results are processed (see Data processing) and displayed in the window entitled **SPR1**, which is opened automatically if necessary (see Data presentation). Positions of the curves minima are also marked with vertical markers and displayed in the top right corner of the window. If the curve minimum is not found, the corresponding message is displayed instead of the angular value. The results can be saved to file in text or graphic format (see Save file), copied to clipboard (Copy Clipboard) or printed (Print graphics).

Multiple measurement 😾

Periodic measurement of the SPR curves (in the same way as in the Single measurement mode). Data obtained in each measurement are processed and displayed similarly to the Single measurement mode, then for every enabled measurement channel the angular position of the minimum is found (see Data processing). If the minimum is found, its position is displayed on the plot in the window entitled **SPR2** (see Data presentation). If the minimum is not found, a corresponding message is displayed in the **SPR1** window, and a corresponding point is omitted in the **SPR2** window.

If the prescribed value of the **Period**, s element is less than the time required for the single measurement, then the measurements will be performed continuously.

Kinetic data displayed in the **SPR2** window can be processed mathematically (see Data processing).

The results can be saved to file in text or graphic format (see Save file), copied to clipboard (Copy Clipboard) or printed (Print graphics).

Additionally, the SPR curves can be saved in text files directly in the course of the measurement in the Automatic data saving mode. The measurement process can be interrupted by sending the **Pause** command or terminated by pressing the **Stop** button. In the latter case, the last measured point data may be lost.

Maximal continuous measurement time depends on several parameters; first of all, it depends on the angular scan range (**Max angle, degree**). With increase of the angular scan range the measurement time increases. The maximum continuous measurement time for the angular scan range of 5 degrees is 8 hours.

Track mode 🚾

Operation in this mode is similar to the operation in the Multiple measurement mode, but it is much faster because the smaller angular range is scanned. The measurement is carried out in the following way. On starting this mode, the first measurement is performed in the same way as in the Single measurement mode. Then, for the multi-channel instrument. the Managing Channel window is opened for the selection of the managing channel. After the selection of the required channel configuration, the instrument is positioned at the angle of **Deviation, degree/2** (see Set options) to the left of the SPR curve minimum in the managing channel and the scan is performed in the angular range of **Deviation**, degree. For every enabled channel the angular position of the minimum is found (see Data processing) and, if the minimum is found, its position is displayed on the plot in the window entitled SPR2 (see Data presentation). If the minimum is lost in the course of the measurement (for example during fast processes), the program will switch to the mode of full SPR curve measurement and after finding of the minimum it will return to the Track mode.

Note: The minimum in one of the channels cannot be found in some cases for the multi-channel instrument. In this case the correspondent data point will be omitted.

The results can be saved to file in text or graphic format (see Save file), copied to clipboard (Copy Clipboard) or printed (Print graphics).

The measurement process can be interrupted by sending the Pause command or terminated by pressing the Stop button or by sending the corresponding command from the main menu. In the latter case, the last measured point data may be lost.

Maximal continuous measurement time depends on several parameters; first of all, it depends on the angular scan range (**Deviation, degree**). With increase of the angular scan range the measurement time increases. The maximum continuous measurement time for the angular scan range of 2 degrees is 3.5 hours.

Slope 🔛

In this mode measurements are carried out without mechanical rotation of the measurement prism and are characterized by high speed of operation and low noise level.

If the **Rel.units** (see below) representation mode is selected, then the «Set Former angle of measurements? [Yes] [No]» question is displayed after start of the measurement. In the case of the positive answer, measurement prism is set in the position which it was in during the last measurement session and the measurement begins. This allows continuing the measurements, interrupted for some reason. If the other mode is selected or in the case of the negative answer, the first measurement is performed in the same way, as in the Single measurement mode and the Managing Channel window for selection of the managing channel is opened for the multi-channel instrument. Then, if the **50** % element (Options) is selected, the measurement prism is positioned at the angle which the signal corresponds to the half of the height of the SPR curve left slope at, and the measurement begins. If the **Pointer** element is selected, user can select the point on the curve by dragging the marker that appears in the **SPR1** window with the left mouse button pressed. Then the measurement prism is positioned corresponding to selected point and the measurement begins.

The measurement consists in reading the signals in the enabled channels, processing these signals and displaying them in the window entitled **SPR2** (see Data presentation) when the prism position is fixed. Then the instrument waits for the time specified by the **Delay**, **s** element and the process is repeated.

Processing of signals and their representation depends on the representation mode, selected for the **SPR2** window. Three choices are possible; they are selected by the corresponding element on the Slope page in the **Options** window: **Axis in rel.units, Axis in grad.corr** or **Axis in degree**. If the Axis in rel.units element is selected, signal levels (in ADC code units) are used as the kinetics points. If the **Axis in grad.corr** element is selected, values of the signals are multiplied by the gradients of the corresponding SPR curves. This allows comparing signals from the two channels more correctly. If the Axis in degree element is selected, values of the signals are converted into the effective angle change and kinetics is displayed in angular units (degrees). The kinetic data obtained can be additionally processed depending on the **Peak-filter** and **Averaging** options. The results can be saved to file in text or graphic format (see Save file), copied to clipboard (Copy Clipboard) or printed (Print graphics).

The measurement process can be interrupted by sending the Pause command or terminated by pressing the Stop button. In this case, the last measured point data may be lost.

Maximal continuous measurement time depends on the Delay, s option and for delay of 0.1 s it equals 2 hours, and for delay of 10 s - 200 hours. Maximal continuous measurement time is calculated and indicated before the beginning of the measurement in the **Reminder** window.

Angular calibration $\overline{>}$

It is necessary to know absolute values of angle of incidence of light for certain calculations. For absolute angles determination a built-in calibration reference point is present in the device. Angular position of the reference point is located near from the position of normal incidence of light on the prism and it can be determined precisely during device adjustment by the manufacturer as a device calibration correction.

System calibration is the process of associating the angular sweep system to the built-in calibration reference point. Calibration should be carried out each time after changing the prism or any modification of its position (e.g. by preset ring).

Calibration mode can be turned on by selecting the **Measurement/Calibration** menu item or by pressing the **Device calibration** button in the main window.

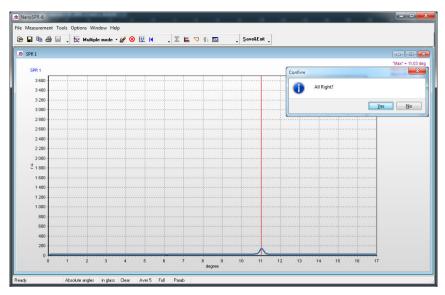


Figure 23Angular calibration

During the calibration, the special mode for measurement of the reference point signal is established and the system position in the moment of the maximum reference point signal is determined. The reference point signal is drawn in the **SPR 1** window, and the calculated position of its maximum is shown by the vertical line and the text is outputted, e.g. "0"=607 a.m., in the same place. It means that the angle of incidence of light on the input prism face equals zero when the device rotation angle equals to 607 angular minutes. The passport correction for true angular reference point position is taken into account automatically.

System stores the determined calibration value and uses it for the calculation of angles of incidence of light in **Absolute angle** modes (see <u>Data processing</u>, <u>Data presentation</u>). If a device calibration was not carried out, the system will use an old calibration value without any warning.

Note: An instrumental calibration correction is saved to the "DeviceDat.dat" file, which the instrument is supplied with, and it is read out automatically on the first program start.

Adjustment mode 🖳

To switch to the adjustment mode, choose the **Tools/Adjustment mode** menu item or press the "**Adjustment mode**" button. In this mode the signal intensities in all device channels are monitored periodically and their values are displayed as a color bar chart diagram in the main window.

This process lasts for several minutes and then switches off automatically. It can be also terminated by pressing the <u>Stop</u> button or by pressing the "**Adjustment**" button again.

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Adjustment				-
		2172		Laser Power Channel 1 Channel 2 Angle Label Subsidiary 1100 Bettery 110 Temper 1 110 Temper 2 110
		2 252		
102				
0,398				
126				
0 500	1 000 1 500	2 000 2 500	3 000 3 500	

Figure 24 Adjustment mode

Note: The indication is carried out for all channels, even for those that are not used in the current device model.

The signals table can be shown to the right of the plot if the Legend element is checked in the pop-up menu of the window.

This mode is convenient for the device adjustment and testing purposes.

Subsidiary channel

Subsidiary channel is intended for measurement and representation of the user's external signal voltage. The measured range is -5V...+5V.



Figure 25Subsidiary channel measurement cable

The subsidiary channel can be operated in two different modes: in the independent mode and in the synchronous mode. During operation in the first mode, the measurement is performed periodically after a period of time specified by the timer. In this mode the measurement starts immediately after the window is opened. In the second mode the measurement in the subsidiary channel is performed simultaneously with the SPR kinetics measurement. It means that the measurement is performed when the next kinetics points series is determined (the minimum positions in the <u>Multiple measurement</u> mode or the signal measurements in the <u>Slope</u> mode).

Note: The independent measurement mode is available only during the stand-alone operation of the subsidiary channel. If any mode of kinetics measurement has been switched on, the synchronous mode will be automatically switched on.

<u>Attention! If the 5 channel is turned off, the external signal</u> measurement mode is unavailable.

It is possible to control the Subsidiary window and mode via the <u>Pop-up menu</u> and from the **Subsidiary** panel on the **Advanced** page in the **Options** window.

These panels contain the following elements:

- Averaging switches on/off the averaging mode.
- Number of points defines the number of points that are used for averaging.
- Independence Period, s specifies the measurement period.

Note: Inaccuracy of the measurement of the internal signal can be reduced by executing the accurate <u>Subsidiary channel</u> <u>calibration</u>.

Subsidiary channel calibration

Calibration of the subsidiary channel consists in determination of precise values of amplification factor and position of the zero line of the channel.

To perform the calibration, proceed as follows:

1. Switch the channel on in the independent mode by pressing the button in the main window.

2. Short-circuit the wires corresponding to the input signal of the subsidiary channel and wait for some time while the zero line is drawn.

3. Connect the constant-current source with known voltage to the channel input and wait for some time while the line

corresponding to this voltage is drawn.

4. Interrupt the measurement by selecting the **Independence** item in the pop-up menu. Changing the <u>curve scale</u>, determine the applied voltage value as a difference between the signal and zero line levels. Divide the real applied voltage by this value and obtain the **k1** coefficient.

5. Open the **Options** window on the **Advanced** page, read the amplification coefficient value \mathbf{k} from the **Amplification** window and replace it with a new value calculated as the $\mathbf{k}*\mathbf{k}\mathbf{1}$.

6. Short-circuit the wires corresponding to the input signal of the subsidiary channel and wait for some time while the zero line is drawn.

7. Determine the precise **p1** value, which corresponds to the "0" input signal.

8. Open the **Options** window on the **Advanced** page, read the pedestal value **p** from the **Pedestal**, **V** window and replace it with a new value calculated as **p-p1**.

Pause X

The pause is switched on/off by pressing the **Pause** button or the <u>"Ctrl" + "**P**" keys in the keyboard</u>. This mode is intended for the temporary measurement interruption. It is useful when the examination of data already obtained is desired, to switch to another application and so on.

Switching on the Pause mode is indicated by change of the button image color.

It is recommended to use this mode also when making some commutations, changing options etc.

Note: If the Pause mode is switched on during the kinetics

measurement, then program operation interrupts but <u>the</u> measurement time count continues.

Stop 😣

Kinetics measurements can be interrupted at any time by pressing the **Stop** button. After pressing the button, the interruption may not occur immediately because the system may need some time to stop the operation correctly.

Information about the last measured point may be lost when the **Stop** command is used.

Restart I

Sets the instrument mechanical system to its initial position. This operation is performed automatically on starting the program and it can be repeated using the **Restart** command at any time when the instrument is not carrying out the measurement. It is recommended to execute this operation if the fault in the mechanical system of the instrument is suspected.

Note: With an **Init Device** option turned on in the Multiple page in Options window, **Restart** command will be executed automatically before every measurement.

Main window

When the program starts, it detects and initializes the instrument and after that opens up the Main window of the program.

The Main window contains main <u>Menu</u>, control <u>Buttons</u> below the menu and <u>Status bar</u> at the bottom of the window.

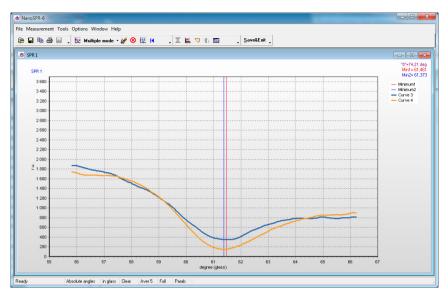


Figure 26Main Window

The middle part of the window is empty just after start of the program. Windows with plots of the measurement results will be located here.

Menu and control buttons have standard appearance, structure and functioning unlike **Status bar** which serves as an active control element.

Most of available functions are duplicated by menu items and control buttons. Some of them are also duplicated by the status bar active fields. Therefore, the purpose and the action of most buttons are described in corresponding sections.

Note: The main window current size and position are stored by the system. You can resize or drag the main window to any desirable position, and its size and position will be reproduced when you run the program next time.

Control Buttons

Control buttons are normally located in 4 customizable toolbars which are situated in the top part of the main window.

Initially, buttons are located in the following order:

Toolbar 1 contains:



Open file – read text files. Save file – save data in a file. AutoSave – enable automatic data saving in a text file. Copy Clipboard – copy data into system Clipboard. Print graphics – copy plot to the printer.

Toolbar 2 contains:

- Single measurement start single-stage measurement.
- Kinetics measurement start kinetics measurement.
- Multiple measurement start reiterated measurement.

Track mode – start measurement with tracking of the minimum.

Slope – start measurement of signal intensity on the slope of the SPR curve.

- ClearSPR1 clear the SPR1 window.
- \otimes Stop terminate the measurement.
- Subsidiary channel subsidiary channel.
- Restart set up initial position.

Toolbar 3 contains:



Set options – set up parameters.

Pause – pause/resume the measurement.

Adjustment – enter the instrument adjustment mode. Calibration – start calibration of absolute angles measurement.

Pumps – control the probe pumping system.
 Open/close the Monitor window intended for the monitoring of temperatures and device power supply voltage.

Toolbar 4 contains:

Save&Exit − finish device operation.
 Ø Help − Open help file.

Clearing the SPR1 window

The SPR1 window can operate in two different modes: all measured curves are plotted in the same window or a new window is opened for every curve. Representation mode can be switched via the <u>pop-up menu</u> (SPR1 in New Window) or via the <u>main menu</u> (Window/SPR1 in New Window). Window clearing is intended for removing the old curves when operating in the first mode.

If the **Auto Clear SPR1** mode is switched off when operating in the first mode, then all measured curves are plotted in the SPR1 window and the last curve number is indicated. If the clearing mode is switched on, then not more than 16 lately measured curves will be displayed in the window and curve number will not be indicated.

It is possible to switch automatic window clearing on/off on the **Service** page in the <u>Options</u> window or via the <u>Status bar</u>.

Note 1: After window clearing curve numeration starts from unity.

Note 2: <u>Attention!</u> Continuous operation without window clearing leads to computer memory overflow and may cause measurement termination.

Save&Exit

To finish the work with the instrument, press the **Save&Exit** button, select the **File/Save&Exit** item from the <u>main</u> <u>menu</u> or press the **F4** key in the keyboard.

The system will request:

• "Save SPR2?", if the SPR2 window was opened but was not saved;

• "Delete AutoConserved files?", if the <u>Automatic data</u> saving mode was switched on;

• "Save SPR1?" and then close the program.

Note: The program can be also closed by pressing the system close button in the top right corner of the window; in this case no data saving requests will be displayed and certain option settings which were defined during the operation will not be stored.

Main menu

Almost all menu functions are duplicated by the main window <u>buttons</u> or by the <u>pop-up menu</u> commands; these functions are described in their descriptions, with the exception of the following ones:

Tools/Set device – records instrument data contained in the "deviceDat.dat" file to the system registry and allows reinstalling

the port the instrument is connected to.

Window/SPR1 in New Windows – if this element is selected, each measured SPR curve is plotted in a separate window.

Window/Notes – calls and opens/closes the <u>Notes</u> window.

Window/View – calls and opens the <u>View</u> review window.

Window/Align – controls the windows arrangement.

Window/Style – controls the display style of buttons, menu, etc.

Help/Help Hardware – calls the instrument (hardware) HELP-file.

Help/Help Software – calls the software HELP-file (it is connected by default).

Toolbars setup

Control buttons are normally located in 4 customizable toolbars in the top part of the main window. Toolbars adjustment is carried out in the **Customize** dialog window, which is opened from the Tools/Customize menu of the main window. Furthermore, it is possible to hide or show buttons that appear in each panel. To do this, click on the right field of the toolbar (down arrow) and check or uncheck the flag opposite to the button of interest in the drop-down window. Customize dialog window contains the following pages: **Toolbars, Actions, Options**.

The **Toolbars** page contains the Toolbars panel, which elements for turning on/off the **File**, **Measurement**, **Tools**, **Exit**, **Adjustment** toolbars in the main window of the program are located on. To turn on/off either toolbar it is necessary to check or uncheck the corresponding flag. This page also contains the **ToolbarOptions** panel which allows turning on captions of correspondent buttons for the selected toolbar or all toolbars if the **Apply caption options to all toolbars** flag is checked.

The Actions page contains the Categories and Actions panels. These panels allow every user of the program to adjust toolbars. Namely, user can pick only the buttons that he needs in the toolbar. To do this, select the category of interest from the Categories panel, and elements related to this category will be shown in the Actions panel at that. Then choose the element from the Actions panel by clicking it with left mouse button and drag it to the desired toolbar of the main window while holding the mouse button.

The **Options** page contains the **Personalized Menus and Toolbars** and **Other** panels. If the **Menu show recently used items first** flag in the **Personalized Menus and Toolbars** panel is checked, then the first button in the toolbar will be the one that was used last in the latter operation session. The Other panel allows switching to large toolbar button icons when the **Large icons** flag is checked, and controlling switching on/off the pop-up prompts that appear when the mouse pointer is pointed at the button symbol in the toolbar.

Status bar

The Status bar is an active control element of the Main window. It is intended not only to inform about the operation modes that are currently set, but also to set up and modify some of them. That partially duplicates controls in the <u>Options</u> window.

Ready	Absolute angles	in glass	n Clear	Non	Full	Parab
		in grace	n_enear		1 411	- arae

Figure 27Status bar

The Status bar consists of 8 fields, first and eighth of which are intended for indication only, while the rest of them are used for both indication and modification of corresponding modes. To switch between the different modes, click on corresponding field in the status bar. At that, the inscription in the string field will change, and the appearance of the field may change. Status bar fields and their destinations are listed below.

1. This field indicates the current operation mode.

2. Indicates and modifies the light angle-of-incidence calculation and presentation mode:

Relative angles – SPR1 window displays the turning angle of the prism table with reference to its initial position as a coordinate;



Absolute angles – SPR1 window displays the absolute angle of incidence of light as a coordinate. In order for this mode to operate correctly, the <u>Angular calibration</u> is to be carried out first (see also <u>Data processing</u>).



3. Specifies absolute angles calculation mode: in glass – angles indicated in the data plots correspond to the angles of incidence of light on the medium under investigation;

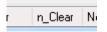
igles	in glass	n_C

in air – angles indicated in the data plots correspond to the angles of incidence of light on the input face of the prism. When the beam reflected from the input prism face appears to lie above the incident beam, the angles are considered negative; in the opposite case they are considered positive. See also <u>Data processing</u>.

ngles	in air	n_Cl

4. Enables/disables automatic erasing of the SPR curves in the SPR1 plot. Available only when the SPR1 in New Windows mode is turned off:

n_Clear – all SPR curve plots are displayed;



Clear – on plotting every sixteenth SPR curve the window is cleared, and then the next 16 curves are plotted and the window is cleared and so on.

 Clear	N

Note 1: The window can be cleared at any time by pressing the **Clear SPR1** button.

Note 2: <u>Attention!</u> Continuous operation without window clearing leads to computer memory overflow and may cause measurement termination.

5. Indicates and controls processing of kinetics curve data (SPR2) for the <u>Multiple measurement</u> and <u>Slope</u> operation modes. Options for data processing modes can be adjusted in the <u>Options</u>**Multiple** and <u>Options</u>**Slope** windows.

None – data processing is switched off;

Peak – single spike filter (peak-filter) mode is switched on;

Peak+Aver – peak-filter and data averaging modes are switched on;



AverX. – data averaging mode is switched on, averaging is executed by X points.

6. Indicates and controls the plot scale of the OY axis for kinetic measurement in the SPR2 window:

Auto – OY axis scale in the SPR 2 window is selected automatically. This is the default mode;

Link – OY axis scale in the SPR 2 window is dependent on the SPR 1 window scale so that with changing of the plot scale in the SPR 1 window the OY axis scale in the SPR 2 window is changed automatically;

Non	Link	Parab

Fix – previously adjusted OY axis scale (e.g. in **Link** mode) is kept.



7. Indicates and controls the method of determination of the SPR curve minimum:

Abs (Absolute minimum) – if this element is turned on, minimum position is calculated as the angular position of the absolute minimum of the curve;

Fix	Abs	

Parab (**Parabolic approximation**) – method of approximating the SPR curve by the second-order polynomial; minimum position is then calculated for this polynomial function. When operating in this mode, SPR curve minimum could be found even if the minimum point is not reached during the measurement. This is the default mode;



Average value – determination of the minimum position by averaging midpoints of several segments connecting two points which are located at grade on different slopes of the SPR curve. This method is implemented in some versions of the program only;

Cubic (Cubical approximation) – method of approximating the SPR curve by the third-order polynomial; minimum position is then calculated for this polynomial function.

This method is implemented in some versions of the program only;

Center of mass – this method is implemented in some versions of the program only.

The minimum determination method can be also selected via the <u>Status bar.</u>

Note: Different methods could be implemented in different versions of the program.

8. This field is used by the <u>automatic data saving</u> to the files. It displays the name of the last data file saved and time when it was saved.

All operation modes displayed and accessible for changing in the status bar can be also switched on/off using control elements in the "Set options" window that can be activated by the <u>Options</u> button or from the **Options** menu item. Some modes can be managed from the <u>Pop-up menus</u>.



Figure 28 AutoSave options

Pop-up menus

The SPR1, SPR2, View, Subsidiary and Adjustment windows possess the pop-up menus. The pop-up menu is called by the mouse click on the correspondent window using the right mouse button.

For the most of windows the pop-up menu contains the following items:

Align – in its turn, contains embedded menu that controls the windows arrangement. All submenu items have standard meaning except the **Normalized** item. When this item is selected, the SPR1 window is placed in the left third part of the screen and has the full screen height, and the SPR2 window is placed on the right from the SPR1 window. If the Subsidiary window is opened, the SPR2 window height is halved and the Subsidiary window is placed underneath the SPR2 window. Such windows arrangement is used as default.

Copy Clipboard – copies the contents of the window, which the pop-up menu was called from, to the system Clipboard.

Save Graphic – saves the information contained in the window, which the pop-up menu was called from, to the file (see <u>Save file</u>).

Print – prints out the plot contained in the window, which the pop-up menu was called from, using the system printer (see <u>Print</u>).

Delete Window – deletes the window, which the pop-up menu was called from.

Note: This is the only method to delete the already open windows.

Pop-up menu in each window contains specific items in

addition to the common items.

SPR1 window pop-up menu

This pop-up menu contains the following items:

Minimum Search – allows finding and displaying the minimum of the last SPR curve measured or opened from the file.

 $\label{eq:Clear SPR1} \begin{array}{c} Clear \ SPR1 - clears \ the \ window \ by \ deleting \ all \ previously \\ plotted \ curves. \ This \ menu \ item \ is \ duplicated \ by \ the \ similar \\ button. \end{array}$

Auto Clear SPR1 – turns on the mode, during which not more than 16 curves are plotted in the window. After that the window is cleared, and the next 16 curves are plotted etc. This mode can be also controlled via the Status bar.

SPR1 in New Windows – when this item is selected, each measured SPR curve is plotted in a separate window. Otherwise, all curves are displayed in the same window. This menu item is duplicated in the main menu.

Legend – controls the representation of the curves marking table that is to the right of the plots.

SPR2 window pop-up menu

This pop-up menu contains the following items:

Difference – when this item is selected, the curve that presents the arithmetic difference of the curves correspondent to the 1 and the 2 channels is plotted in the SPR2 window. Otherwise, kinetic curves are plotted for each channel. This element is available for two-channel instrument only.

Axis-units – submenu which contains elements that define measurement units for outputting the kinetics in the window. During operation in the Slope mode submenu elements are

available, if the Axis in degree mode is turned on. If the measurements are executed in this mode, the measured signal level is converted into the effective change of the angular position.

Angle degree – measurement units – degrees.

Angle minute – measurement units – angular minutes.

Angle second – measurement units – angular seconds.

Refr.Index – the measured kinetics is converted to the effective value of refractive index of the medium.

Tools – submenu which contains elements for turning on the service possibilities for processing of the data represented in the window.

Coordinate – turns on the mode of mouse pointer coordinates determination while it moves over the plot.

Distance – turns on the mode of determination of the distance between two sequentially specified plot points. The distances along the X and Y coordinates are determined separately.

Delta Y Line – turns on the mode of determination of the distance between two sequentially specified levels along the Y coordinate.

Delta Y Region – turns on the mode of determination of the distance (along the Y coordinate) between the average values of points which are included in two selected regions.

Gradient – turns on the mode of determination of the curve gradient between two selected points.

It is also possible to turn on service possibilities from the Status bar.

Link mode – submenu which contains elements for scaling of the vertical axis of the plot displayed in the SPR2 window.

Full Graphic – the full window with the initially set scale.

Link mode – when the plot scale in the SPR1 window is changed, the OY axis scale of the plot in the SPR2 window changes automatically.

Fix Left Axis – breaks the link between the SPR1 and SPR2 windows and stores the OY axis scale previously specified in the Link Graphics mode.

SPR2 marks – when this item is selected, the points on the curves displayed in the SPR2 window are marked with markers.

Legend – controls the representation of the curves marking table that is to the right of the plots.

Show Const - controls the display of constants showing the sign and magnitude of the change in kinetics.

Subsidiary window pop-up menu

This pop-up menu contains the following items:

Independence – when this item is selected, the independent operation mode is switched on. In this mode the measurement starts immediately after the window is opened and it is performed periodically after a period of time specified by the timer. Otherwise, the measurement is performed simultaneously with the SPR kinetics measurement. It means that the measurement is performed when the next kinetics point is determined (the minimum position in the Multiple measurement and Track modes or the signal measurement in the Slope mode).

Tools – submenu which contains elements for turning on the service possibilities for processing of the data represented in the window.

Coordinate – turns on the mode of mouse pointer coordinates determination while it moves over the plot.

Gradient – turns on the mode of determination of the curve gradient between two selected points.

Subsidiary marks – when this item is selected, the points on the curves displayed in the Subsidiary window are marked with markers.

Legend – controls the representation of the curves marking table that is to the right of the plots.

Axis Auto – if this option is set, the scale is selected automatically in such a way that all displayed information is visible.

Averaging – turns on/off the averaging mode. Number of points which are averaged is defined in the Subsidiary panel on the Advanced page in the Options dialog window.

View window pop-up menu

This pop-up menu contains the following items:

Add File – adds kinetic from the new file to the window.

Clear – clears the window.

Axis Auto – if this option is set, the scale is selected automatically in such a way that all displayed information is visible. Otherwise, the maximal value is set equal to the maximal OY axis value of all kinetic curves loaded, and minimal value is set equal to the minimal OY axis value.

Legend – controls the representation of the curves marking table which is to the right of the plots.

Set options

Setting of the options is executed in the "Options" dialog window. This window is activated by the "**Set Options**" button or the "**Options/Set Options**" menu item.

It is possible to set up options related to kinetic measurements by pressing the right field (down arrow) of the

Kinetic measurements button in the second toolbar. At that, a menu will drop down; select the **Options Kinetic** menu item and the **Options** dialog window which contains pages related to kinetic measurements will open.

The "**Options**" window elements are located on seven pages:

- The Multiple page contains device adjustment elements for operation in the multiple SPR-curve measurement Multiple measurement mode.
- The Track page contains device adjustment elements for operation in the Track mode.
- The Slope page contains device adjustment elements for operation in the Slope mode without mechanical rotation of the measurement prism.
- The Single page contains device adjustment elements for operation in the single curve measurement Single measurement mode.
- The Service page contains control elements for data representation windows color appearance and control elements for sound accompaniment of the measurements.
- The Advanced page contains elements that assign parameters of the device optical part and subsidiary channel control elements.
- The Monitor page contains elements for control and management of the temperature and device power supply.

Most of the changes made take effect after pressing the "OK" button. Changes of some options are immediately sent to program; control elements of these options are highlighted with green color.

The "**Default**" button is intended for recovery of the default options values.

It is recommended to change options before beginning of the measurement. Options change during the measurement may lead to unpredictable results. Options which can be changed during the measurement can be managed using the Status bar or Pop-up menu.

23 Options Multiple Track Slope Single Service Advanced Monitor SPB 1 OK Max angle. 15 🚍 Refresh Minimum dearee Leaend 2 🗙 Cancel Period s SPB 2 Peak-filter 1 channel On 2 channel On Averaging 5 8 Number of points 3 🚍 Lines width Difference 1-2 Difference 2-1 Init. device Default 🗸 Const Legend AutoSave 8 SPB 1 File SPR 2 Period min 2

Multiple page

Figure 29Multiple page options

The **Multiple** page is intended to set up main program options for the **Multiple** mode and contains the following control elements:

Max angle, degree – specifies the angular prism rotation range when operating in the <u>Single measurement</u> and <u>Multiple</u> <u>measurement</u> modes and when searching for the operating point before measurement in the <u>Slope</u> mode;

Period, s – specifies the measurement period duration in the <u>Multiple measurement</u> mode. If the period specified is less than the time required for the measurement of one point, the measurement will be executed continuously;

The **Channel On** elements allow switching on (when the flag is set) or off (when the flag is not set) device measurement channels.

The **Lines width** element specifies the width of the lines which are used for plotting.

For **SPR1** window:

Refresh Minimum – if this flag *is set*, the last values of the minima will be marked on the plots and the previous ones will be cleared. If this flag is *not set*, all old values will remain;

SPR1 Legend – controls the representation of the curves marking table that is to the right of the plots.

For **SPR2** window:

Peak-filter – switches on/off the single peak filtration mode for the kinetics curve;

Averaging – switches on/off the data averaging mode. The number of points averaged is determined by the **Number of points** element;

Difference 1-2 - in Difference mode, the kinetic curve will be constructed as the difference channel 1 minus channel 2.

Difference 2-1 - in the Difference mode, the kinetic curve will be constructed as the difference channel 2 minus channel 1.

SPR2 Legend – controls the representation of the curves marking table that is right of the plots.

The AutoSave panel is intended for managing automatic saving of experimental data during the operation. The SPR 1 element switches on/off the <u>Automatic data saving</u> mode for SPR1 window, and the SPR 2 element switches on/off kinetics auto saving (for SPR2 and Subsidiary windows). If one switches the SPR2 auto saving on, then the **Period, min** element is activated. This element specifies the auto save period for the SPR2 and Subsidiary windows. On default the kinetics (the SPR2 window) is saved to the "SPR2.sp2" file. The file name can be modified after pressing the **File** button.

Track page

lultiple Track S	lope Sing	le Service	Advanced	Monitor	
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Deviation, degree		Refresh Minimu Legend	IM		× <u>C</u> ar
5	SP	R 2			
🗸 1 channel On	1	Peak-filter			
🗸 2 channel On	V	Averaging			
ines width 3	•	Number of poi Difference Difference	1-2		
AutoSave		Legend	🔽 Const	2	<u>D</u> efa

Figure 30Track page options

The **Track** page in the **Options** window is intended for the **Track** mode options setup and contains the following control elements:

Deviation, degree – specifies the angular scanning range when operating in the Track mode. Maximum deviation of the

prism from the angular position of the minimum equals to the half of the Deviation, degree value.

The **Channel On** elements allow switching on (when the flag is set) or off (when the flag is not set) device measurement channels.

The **Line width** element specifies the thickness of the lines used for drawing the plots.

The **SPR1 panel** contains the following elements:

Refresh Minimum – if this flag is set, the last values of the minima will be marked on the plots in the SPR1 window and the previous ones will be cleared. If this flag is not set, all old values will remain.

Legend – controls the representation of the curves marking table of the SPR1 window that is to the right of the plots.

The **SPR2 panel** contains the following elements:

Peak-filter – switches on/off the single peak filtration mode for the kinetics curve.

Averaging – switches on/off the data averaging mode. Number of points that are used for averaging is defined by the Number of points element.

Difference 1-2 - in Difference mode, the kinetic curve will be constructed as the difference channel 1 minus channel 2.

Difference 2-1 - in the Difference mode, the kinetic curve will be constructed as the difference channel 2 minus channel 1.

Legend – controls the representation of the curves marking table of the SPR2 window that is to the right of the plots.

The **AutoSave panel** is intended for managing automatic saving of experimental data during the operation. The SPR2 element switches on/off kinetics auto saving (for SPR2 window). On default the kinetics (the SPR2 window) is saved to the "SPR2.sp2" file. The file name can be modified after pressing the File button.

Options					×
Multiple Track Slope	Single	Service	Advanced	Monitor	
	SPR 2	<u>.</u>			🗸 <u>о</u> к
Delay, S x10 🔟 テ	🔘 Axi	s in rel.un s in grad.c s in degre	its OPoi	0% inter nsitivity x S/N	X <u>C</u> ancel
✓ 1 channel On✓ 2 channel On	✓ Av	ak-filter eraging	ma a 3 6	a	
Lines width 3 🕃	🔘 Di	veraging Tir fference 1 fference 2	-2		
AutoSave	🔳 Leg	end	🔽 Const		<u>D</u> efault
File		SPR 2			

Slope page

Figure 31Slope page options

The **Slope** page is intended to set up options for the <u>Slope</u> mode and contains the following control elements:

Delay, Sx10 – specifies the interval between the measurements;

The **Channel On** elements allow switching on (when the flag is set) or off (when the flag is not set) device measurement channels.

The **Line width** element specifies the width of the lines which are used for plotting.

For **SPR2** window:

These three elements determine the method of calculation and presentation of the kinetics:

Axis in rel.units – kinetics is displayed in ADC code units (relative units);

Axis in grad.corr – kinetics is displayed in ADC code units but the values are corrected during the calculation by taking into consideration the SPR curve slope at the measurement point;

Axis in degree – during the calculation ADC code values are recalculated into the effective angle change and kinetics is displayed in angular units.

These two elements determine the point on the SPR curve at which the measurement is executed:

50 % – before the start of the measurement mechanical system of the instrument is automatically set into position corresponding to a half of the altitude of the SPR curve left slope;

Pointer – program allows the user to select the position of the mechanical system by dragging the marker in the SPR1 window to prescribed position (when the left mouse button is pressed).

Sensitivity -This mode allows automatically select the operating point on the SPR curve, corresponding to its maximum slope.

Max S/N - This mode allows automatically select the operating point on the SPR curve, corresponding to the maximum signal / noise ratio.

Peak-filter – switches on/off the single peak filtration mode for the kinetics curve;

Averaging – switches on/off the data averaging mode. The time interval during which the obtained points are averaged is determined by the **Averaging time**, **s** element;

Difference 1-2 - in Difference mode, the kinetic curve will be constructed as the difference channel 1 minus channel 2.

Difference 2-1 - in the Difference mode, the kinetic curve will be constructed as the difference channel 2 minus channel 1.

SPR2 Legend – controls the representation of the curves marking table that is right of the plots.

The AutoSave panel is intended for managing automatic saving of experimental data during the operation. The SPR 1 element switches on/off the <u>Automatic data saving</u> mode for SPR1 window, and the SPR 2 element switches on/off kinetics auto saving (for SPR2 and Subsidiary windows). If one switches the SPR2 auto saving on, then the **Period**, **min** element is activated. This element specifies the auto save period for the SPR2 and Subsidiary windows. On default the kinetics (the SPR2 window) is saved to the "SPR2.sp2" file. The file name can be modified after pressing the **File** button.

Single page

Multiple Track Slo	ope Single	Service	Advanced	Monitor	
Max angle, degree 13	5	Averagi	ng		<u>✓ 0</u> K
☑ 1 channel On ☑ 2 channel On		🗖 Decima	tion		X Cance
V Auto clear SPR1					
Lines width 3		Cut	ing sensitivi	ty 100	
☑ SPR1 Legend		5	Drawing	tion	<u>D</u> efault

Figure 32Single page options

The **Single** page in the **Options** window is intended for SPR1 window operation mode options setup for both single and kinetic measurements and contains the following control elements:

Max angle, degree – specifies the angular prism rotation range when operating in the Single measurement and Multiple measurement modes and when searching for the operating point before measurement in the Track and Slope modes.

The **Channel On** elements allow switching measurement channels of the instrument on (the flag is set) or off (the flag is not set).

Auto Clear SPR1 – allows/forbids automatic curves clearing in the SPR1 window. It works only if the SPR1 in New Windows mode is turned off. Not more than 16 curves will be output into the plot if the Auto Clear SPR1 mode is turned on.

It is also possible to switch this mode on/off via the Status bar.

The **Line width** element specifies the width of the lines which are used for plotting.

SPR1 legend – controls the representation of the curves marking table that is to the right of the plots.

Averaging – switches on/off the SPR curve averaging mode; if the averaging is switched on, then **n** measured points of the SPR curve are averaged into one point (n is the value that is specified in the **Number of points** window which is activated only if the **Averaging** element is marked). The averaging is made immediately during the measurement and all subsequent operations (such as minimum search, saving data to file etc.) are executed with averaged values. Thus number of points of the SPR curve is reduced by a factor of n. This mode is available during the single-channel instrument operation only.

Decimation – switches on/off the mode of the SPR curve "decimation"; if this mode is switched on and a value of n is set in the Number of points window, then saving SPR curve to file results in saving only n points that are uniformly picked from the measured array of points.

Cutting sensitivity - Adjusts the sensitivity of trimming SPR curves.

The **Drawing acceleration** element allows controlling the speed of the SPR1 curve representation. Increase in speed is achieved by plotting not all of the measured points (but all of the measured points are taken into account during the data processing and saving the data to a file).

Service page

Multiple Tra	ck Slope Single Service Advanced Mor	nitor
Color		✓ <u>о</u> к
SPR1	Panel Passive	
	Start Color Active	X Cance
	End Color Active Color Defa	ult
Sound		
Sound On		
📃 Start		
🔲 End		
Error		<u>D</u> efault
🔲 Ring		-

Figure 33Service page options

The Service page contains service control elements:

The **Color** panel contains control elements that allow specifying color appearance of the <u>SPR1</u>, <u>SPR2</u>, <u>Subsidiary</u> and <u>View</u> windows. The window, which colors are being specified for, is selected by the left element.

It is possible to specify the following options for the first three windows:

• The color of the passive window.

• The color of the top part of the active window (Start Color).

• The color of the bottom part of the active window (End Color).

The window will have a gradient color if Start Color and End Color do not coincide.

All changes made are sent to the program immediately.

It is possible to select the color of the passive window only for the **View** window. This window will always have the selected color.

The **Color Default** button allows restoring all colors to default values.

The **Sound** panel contains control elements for managing sound signals that accompany the program operation:

The **Sound On** element switches on/off all sound signals.

The **Start** string contains the following elements:

Control element for switching on/off the sound that accompanies the measurement beginning. If this element is selected, then the button is activated. A sound file can be selected if one clicks on this button. The name of the selected file is displayed in the text field which is located in the middle of the string.

End and **Error** strings are similar and correspond to the end of the measurement and the measurement error, respectively.

The sound that is defined by the **Ring** string is used upon the pumps control.

Advanced page

Hultiple Track Slope Sing	le Service	Advanced	Monitor	
Optics Prism type K8-50 (gas)		evice °C port CO	M3 👻	<u><</u>
Immers. Refr. Index1,6100Glass Refr. Index1,6160	• V] Position contr] Data repair au] Laser control		× <u>C</u> a
Subsidiary Independence Period, S x10	Ampl	ification x10 1	5	
Averaging	Pede	estal, mV 🛛 1	8	
	E Le	gend		<u>D</u> efa
		V Full files	names	

Figure 34Advanced page options

The **Optics** panel contains the following elements:

Prism type – specifies the type of the used prism;

Immers. Refr. Index – specifies the refractive index of the immersion;

Glass Refr. Index – specifies the refractive index of the glass carrying the active layer.

The program contains the built-in database which contains the following preset parameters:

• four prism types with correspondent refractive indices and working angles,

- two refractive indices for immersion,
- two refractive indices for glass.

If user introduces new values of refractive indices for the immersion or the glass material, these values are added to the database. It is possible to add 3 values of each parameter in such a way. The prism types are predefined and cannot be changed by user.

The **Device** panel contains the following elements:

PC port – specifies PC port which the instrument is connected to. If this option is set to **Auto**, then the program will try to determine the port automatically;

Position control – switches on/off the mode of program tracking of allowable position of mechanical system of the instrument. It is <u>not recommended to turn on</u> this option;

Data repair automatically – activates or deactivates the procedure of automatic repair of the data that program receives from the instrument. It is <u>not recommended to turn off</u> this option;

Laser control – activates/deactivates the automatic accounting of laser radiation instability.

The **Subsidiary** panel contains the following elements:

Independence Period – specifies the measurement period during the independent mode of operation;

Averaging – switches on/off the data averaging mode. The number of points averaged is determined by the **Number of points** element;

Pedestal, V and **Amplification, mV/ru** elements are intended for set-up of amplification factor and position of the base line for the subsidiary channel (see <u>Subsidiary channel</u> calibration);

Legend – controls the representation of the curves marking table that is right of the plots.

Full file name – this element is used when reading previously saved files. If this flag *is not set*, then only a filename of read file is displayed; if this flag *is set*, then a full name (i.e. filename + path) is displayed.

Monitor page

Options									х
Multiple	Track	Slope	Single	Service	Advanced	Mo	nitor		
Cell tem Tempera	perature ture, C x			Temper s	et, C x10	370		<u>✓ 0</u> # <u>★ C</u> ane	
Temper Tempera		10 0,0		Temper s	et, C x10	200			
Power V x10	125	,5		Alarm se	et, V x10	90	•		
-100	E Ter	mper. co	rr *1000		Device#	71	02	<u>D</u> efau	lt

Figure 35Monitor page options

The **Monitor** page contains 3 panels with control elements:

The **Cell temperature**, **C** panel contains elements for indication of the measured temperature (at the left) and for specifying the cell temperature (at the right).

The **Temperature**, **C** panel contains elements for indication of the measured temperature (at the left) and for specifying the

laser temperature (at the right). This panel is not used in the current device model.

The Power panel contains elements for indication of the power supply voltage (at the left) and for specifying the limiting power supply voltage Alarm set at which the power supply emergency warning is displayed (at the right).

The Temper.corr.*1000 element, which allows inputting the cell temperature measurement system correction, is located in the bottom left part.

The instrument serial number is displayed in the bottom right part for reference.

Data presentation

Presentation of the data measured or opened from the files takes place in five types of windows, entitled as **SPR1, SPR2, Subsidiary, Notes, View and Monitor**. Many windows of the **SPR1, SPR2** and Subsidiary type can be opened simultaneously, but only one window of each type contains data which can be processed. Window arrangement can be controlled via the main menu or via the pop-up menus of these windows themselves. Only one instance of the Notes and View windows can exist.

The **SPR1 window** opens in all measurement modes except the stand-alone operation with the subsidiary channel. It is intended for direct display of the SPR curves being measured.

The **SPR2 window** opens in all measurement modes except the Single measurement mode and stand-alone operation with the subsidiary channel. It is intended for presentation of kinetics of the processes. The **Subsidiary window** opens when the Subsidiary mode is switched on. It is intended for displaying the signal in the subsidiary measurement channel.

The **Notes window** is intended for the text user's comments to the measurement process.

The **View window** is intended for simultaneous representation of several previously obtained kinetic curves.

The **Monitor window** is intended for representation of measured cell and laser temperatures and source voltage.

The **Timer window** is opened as a separate window if one makes a mouse double-click on the timer indication in the top right corner of the **SPR2** window. If one makes a mouse double-click on the timer window field, the window will disappear and timer indication will return to the **SPR2** window.

Information displayed in the **SPR1**, **SPR2** and **Subsidiary** windows can be saved to file in text and graphic formats (see Data saving). Additionally, the window can be copied to clipboard (Copy Clipboard) or printed (Print graphics).

Information contained in the **Subsidiary** and **Notes** windows is saved automatically when saving information displayed in the **SPR2** window in any text format.

Information displayed in the View windows can be saved to file in graphic format ***.bmp** or ***.wmf**, copied to clipboard or printed.

The **SPR1**, **SPR2**, **Subsidiary**, **View** and **Notes** windows can display the information previously saved in the text format (see Open file and Command Line).

SPR1 window

Windows of this type open automatically when it is necessary.

The **SPR1 window** is used in all measurement modes except the stand-alone operation with the subsidiary channel. It is intended for presentation of measured SPR curves. If the **SPR1 in New Windows** mode is selected (this mode can be selected via the <u>pop-up menu</u> or <u>main menu</u>), then each new curve is plotted in a separate window. At this, the curve number is displayed in the window title. Otherwise, all curves are plotted in the same window. In this case the **Auto Clear SPR1** mode can be switched on.

Data plots read from the file are always displayed in a separate window.

On the OX axis the angle in degrees is laid off:

with the Absolute angle mode of f – the prism turning angle,

with the **Absolute angle** mode on – the angle of incidence of light on the input face of the prism when the **in Air** mode is selected, or the angle of incidence of light on the top face of the prism when the **in Glass** mode is selected. These modes can be set via the <u>Status bar</u>, in the dialog box brought up by pressing the "Set Options" button or from the menu (see <u>Data processing</u>).

Signal magnitudes measured in the enabled channels are laid off along the OY axis in arbitrary units.

In this window the measured SPR curves are plotted and positions of their minima are displayed if they have been found successfully. The minimum of the curve that corresponds to the first channel is displayed in red color, and the minimum of the curve that corresponds to the second channel is displayed in blue color.

If the flag of the **SPR1 legend** element in the pop-up menu is set, then the table with curves marks is displayed to the right of the plots.

The initially specified plot scale can be changed:

- Mark the rectangle with the left mouse button pressed and then release the mouse button. If the rectangle is marked starting from its top left corner, then its contents will "stretch out" into the whole window. This operation can be repeated for many times. If the rectangle is marked starting from any other corner, then the plot will restore its initially specified scale.
- With the right mouse button pressed, the plot can be "dragged" in the window to represent different segments of the curves.

With the cursor positioned within the window, the pop-up menu can be brought up by pressing the right mouse button. This menu contains control elements for the main functions of the window.

The window can be also controlled via the main menu and the Set options window.

In the right top corner of the window the following information is displayed:

- The value of the last angle calibration;
- The last value that corresponds to the minimum of the curve measured in the first channel. It is displayed in red color;
- The last value that corresponds to the minimum of the curve measured in the second channel (for a two-channel device)

and the number of the measured point (when the kinetics measurement is carried out). It is displayed in blue color.

The contents of the SPR1 windows can be saved to file in both text and graphic formats (see Data saving). Additionally, the window can be copied to clipboard (Copy Clipboard) or printed (Print graphics).

Clearing the SPR1 window 🕷

The SPR1 window can operate in two modes: 1) all measured curves are plotted in the same window or 2) a new window is opened for every curve. Representation mode can be switched via the pop-up menu (SPR1 in New Window) or via the main menu (Window/SPR1 in New Window). Window clearing is intended for removing the old curves when operating in the 1 mode.

If the **Auto Clear SPR1** mode is switched off when operating in the 1 mode, then all measured curves are plotted in the SPR1 window. If the clearing mode is switched on, then not more than 16 lately measured curves will be displayed in the window.

SPR2 window

Windows of this type open automatically when it is necessary.

The SPR2 window is used in all measurement modes except the Single measurement mode and stand-alone operation with the subsidiary channel. It is intended for presentation of kinetic plots of the process under study.

Plots displayed in this window represent the kinetics of the angular position of the SPR curves minima (in the Multiple

measurement and Track modes) or the signal magnitudes measured in the enabled channels (in the Slope mode). For the two-channel instrument all information related to the first and to the second channel is displayed in red and blue color, respectively.

It is possible to operate in the mode when the difference of the signals measured in the first and the second channels is displayed. Switching of the presentation modes can be made from the popup menu.

With the cursor positioned within the window, the pop-up menu can be brought up by pressing the right mouse button. This menu contains control elements for the main functions of the window.

The window can be also controlled via the main menu and the Set options window.

The horizontal scale (time scale) is set automatically.

The vertical scale is initially set equal to the horizontal scale of the SPR1 window (in the Multiple and Track modes) and the vertical scale of the SPR1 window in the Slope mode. In the course of the measurements vertical scale of the window can be changed in the following way:

- Switch the Full Graphic mode to the Link Graphics mode. This can be done via the pop-up menu or via the status bar.
- Change the SPR1 window scale to zoom into the region of interest. The SPR2 window scale will be changed automatically.
- Switch the set Link Graphics mode to the Fix Left Axis mode. The chosen vertical scale will be fixed.

The specified plot scale can be changed for the convenience of the work with plot:

- Mark the rectangle with the left mouse button pressed and then release the mouse button. If the rectangle is marked starting from its top left corner, then its contents will "stretch out" into the whole window. This operation can be repeated for many times. If the rectangle is marked starting from any other corner, then the plot will restore its initial scale.
- With the right mouse button pressed, the plot can be "dragged" in the window to represent different segments of the curves.

The scaling operations can be performed directly in the course of measurements. Nevertheless, it is more correct to make them after the measurements are completed or interrupted by the Pause mode.

In the right top corner of the window the following information is displayed:

- Current timer value. Clicking on the timer twice brings up a separate timer window, which can be dragged to any desirable position in the screen. To hide this window, click on it twice.
- The measurement time for the last data point.
- The slope constant of the curve measured in the first channel.
- The slope constant of the curve measured in the second channel (for a two-channel instrument).

It is possible to switch the curve points markers and the curve marking table (legend) on/off from the pop-up menu.

The following determination service possibilities are provided for processing of the data that are represented in the window:

- Mouse pointer coordinates determination.
- Determination of the distance between two points along the X and Y coordinates.
- Determination of the distances along the Y coordinates.
- Curve gradient determination.

These service possibilities are available from the pop-up menu. Operation results are displayed in two panels located in the bottom left corner of the window.

Service possibilities are more fully described in the Data processing section.

The contents of the SPR2 windows can be saved to file in both text and graphic formats (see Data saving). Additionally, the window can be copied to clipboard (Copy Clipboard) or printed (Print graphics).

Subsidiary window

Windows of this type can be opened by pressing the _bm11 Subsidiary Channel button in the main window.

The **Subsidiary window** is intended for displaying the signal measured in the subsidiary measurement channel.

It is possible to work with subsidiary channel only if this channel is enabled. Information displayed in this window can be saved to file in graphic format and is always saved automatically when saving information displayed in the SPR2 window to file in text format (see Data saving). The window can be also copied to clipboard (Copy Clipboard) or printed (Print graphics).

The initially specified plot scale can be changed:

- Mark the rectangle with the left mouse button pressed and then release the mouse button. If the rectangle is marked starting from its top left corner, then its contents will "stretch out" into the whole window. This operation can be repeated for many times. If the rectangle is marked starting from any other corner, then the plot will restore its initially specified scale.
- With the right mouse button pressed, the plot can be "dragged" in the window to represent different segments of the curves.

All scaling operations can be performed directly in the course of measurements. Nevertheless, it is more correct to make them after the measurements are completed or interrupted by the Pause mode.

It is possible to switch the curve points markers and the curve marking table (legend) on/off from the pop-up menu.

The following determination service possibilities are provided for processing of the data that are represented in the window:

- Mouse pointer coordinates determination.
- Curve gradient determination.

These service possibilities are available from the pop-up menu. Operation results are displayed in two panels located in the bottom left corner of the window. Service possibilities are more fully described in the Data processing section.

For more detailed information about this window and the subsidiary channel see Subsidiary Channel section.

Information displayed in this window is saved automatically when saving information displayed in the SPR2 window to file in text format (see Data saving).

The Subsidiary windows can display the information previously saved in the text format (see Open file and Command Line).

Notes window

Windows of this type can be opened from the main menu: **Window/Notes**.

The **Notes window** is intended for typing any user's comments to the measurement process.

Additionally, the following information is put into the window as a separate line:

• Current date and time upon the mouse click on the date or time representation fields in the **Notes** window;

• Results of service measurements (distance between two points, distance along the Y coordinates, curve gradient) in the <u>SPR2 window</u> upon the mouse click on this results representation fields in the **SPR 2** window;

• Current experiment time upon the mouse click on the current time timer.

Information displayed in this window is saved automatically when saving information displayed in the **SPR 2** window in text format.

The **Notes** windows can display the information previously saved from this window.

View window

This window can be opened from the main menu: Window/View.

The **View window** is intended for simultaneous representation of many previously obtained results. This window can represent either SPR curves or kinetic curves. Type of the curves represented is defined by the type of the curve which was loaded first in the window.

The curves for representation can be read from previously saved *.sp1, *.sp2 and *.spr files.

The time axis scale of the image is selected automatically according to the most long-duration kinetic curve loaded when representing kinetic curves. The OY axis scale is selected depending on the **Axis Auto** option, which can be set upon clicking the right mouse button in the pop-up menu of the **View** window. If this option is set, the scale is selected automatically in such a way that all displayed information is visible. Otherwise, the maximal value is set equal to the maximal OY scale value of all kinetic curves loaded, and minimal value is set equal to the minimal OY scale value of all kinetic curves loaded.

The initially specified curves scale can be changed:

• Mark the rectangle with the left mouse button pressed and then release the mouse button. If the rectangle is marked starting from its top left corner, then its contents will "stretch out" into

the whole window. This operation can be repeated for many times. If the rectangle is marked starting from any other corner, then the plot will restore its initially specified scale.

• With the right mouse button pressed, the plot can be "dragged" in the window to represent different segments of the curves.

The contents of this window can be saved to file in graphic format *.bmp or *.wmf, copied to clipboard or printed. To do this, select **Save file**, **Copy** clipboard or **Print** items in the pop-up menu of the window, correspondingly.

It is possible to close the **View** window using the pop-up menu only by selecting the **Delete Window** item.

Monitor window

This window can be opened from the main menu: **Tools/Monitor** or by pressing the **Monitor on/off** button in the main window.

The **Monitor window** is intended for:

• displaying the power supply voltage and warning upon its decrease lower than the specified value.

• displaying and specifying the cell temperature.

• displaying and specifying the temperature of the additional channel of the laser temperature stabilization (not implemented).

The page contains 3 similar strings:

The **Cell t**, **C** string contains elements corresponding to the cell temperature: elements that indicate measured (at the left) and specified (at the right) temperature and an indicator button for switching the temperature stabilization mode on/off.

The **Temp2, C** string is similar to the previous string but it corresponds to the laser temperature. This string is not used in this device model.

The **Battery,V** string contains elements for displaying the power supply voltage (at the left) and specifying the power supply voltage at which the power supply emergency warning window is displayed (at the right).

Data saving

Save file – saving data in a file of controlled format.

Automatic data saving in the Multiple measurement mode.

Data files format -*.SP1 and *.SP2 files format.

Copy to Clipboard – copying information from the window to clipboard.

Print graphic – printing the plot on a printer.

Save File

Data saving to a file.

To save data, press the Save button in the main window or select the Save command from the main menu or from the pop-up menu of any window.

If this command is sent from the main menu or using the button in the main window, then the program will suggest to save data in the currently active window (the active window is indicated by color); if this command is sent from the pop-up menu, then the program will save data contained in that window which pop-up menu the command has been sent from. After sending the command the program requests the file name and opens it for writing. The file format is determined by the file name extension the user has defined in response on the system request.

Information from the **SPR1**, **SPR2** and Subsidiary windows can be saved in text (*.**SP1**, *.**SP2**, *.**SPD** or *.**CHD**) or graphic format (*.**BMP** or *.**WMF**). Information from the View window can be saved in graphic format only (*.**BMP** or *.**WMF**).

Note: Data stored in the **SPR2** and **Subsidiary** windows are saved simultaneously in the same file. Information contained in the Notes window is stored in a separate file, which is saved simultaneously with the **SPR2** window.

It is recommended to keep all data files in the "Data" folder, which is created automatically during installation of the program.

Data saved in the text format can be read using the Open file command or on starting the application using the Command line. Previously saved files can be also loaded into the View review window.

For more detailed information about *.**SP1**, *.**SP2** and *.**SPD** file formats see Data files format.

Note: If some files were not saved, then the program will suggest saving them on exit.

Automatic data saving 🛱

Automatic data saving mode is intended for automatic saving of SPR and kinetics curves to text files in the ***.sp1** and ***.sp2** formats, correspondingly. Automatic data saving of SPR curves is available only in the <u>Multiple measurement</u> mode while automatic saving of kinetics curves is available in any kinetics measurement mode.

To switch the automatic data saving mode on/off, open the options setting window (<u>Set Options</u>) and select the **Multiple** or **Slope** page.

For automatic saving of SPR curves, switch on the **SPR 1** option. If this mode is on, then after beginning of the measurement in the <u>Multiple measurement</u> mode a dialog window **AutoSave data as text** is opened. It is possible to select or introduce the base file name in this window. After the measurement of each SPR curve, the program will save data to the file with a name, received by addition of the file number to the base name.

If the SPR curves saving mode is enabled, the <u>AutoSave</u> button in the main window becomes available. This button can be used for on-the-fly switching curves saving on/off in the course of the measurements.

After each file is saved, its name appears in the rightmost field of the <u>Status bar</u>.

By default automatic saving of SPR curves is disabled, and it can be enabled as described above or by opening the SPR2 file previously saved with the enabled automatic data saving mode.

Note: Up to 999 files can be saved in this way. In case of need to save more data files, switch the automatic data saving mode off, introduce another base file name and switch it on again.

For automatic saving of kinetics curves, switch on the **SPR 2** option. Switching this mode on activates the **Period**, **min** element that allows specifying the period of auto saving for SPR2 and Subsidiary windows data. If this mode is on, SPR2 and Subsidiary windows data are periodically saved in a file named **"SPR2.sp2"**. To change the file name, press the **File** button and introduce a new name.

After each file is saved, its name appears in the rightmost field of the <u>Status bar</u>.

The state of the SPR2 auto saving mode (on/off) is stored when you exit the program and is used as a default setting at the next program starting.

Copy Clipboard 🛱

Copying the data from the **SPR 1**, **SPR 2** or **Subsidiary** window into the system Clipboard for further use in other applications.

If this command is sent using the <u>**Copy to Clipboard**</u> <u>Button</u> or from the main <u>Menu</u>, the data displayed in the currently active window will be copied to Clipboard. The active window is indicated by the change of its color.

If this command is sent from the <u>pop-up Menu</u>, the data displayed in the window, which pop-up menu the command has been sent from, will be copied to Clipboard.

Print graphic 🚔

Printing out the plot displayed in the **SPR1**, **SPR2** or **Subsidiary** window on the printer selected through the print dialog.

If this command is sent using the <u>**Print graphic** Button</u> or from the main <u>Menu</u>, the plot displayed in the currently active window will be printed out. The active window is indicated by the change of its color.

If this command is sent from the <u>pop-up Menu</u>, the plot displayed in the window, which pop-up menu the command has been sent from, will be printed out.

Data files format

Experimental data can be saved to files in the *.sp1, *.sp2, *.spd, *.bmp and *.wmf formats. The old files format *.spr is also supported.

The *.sp1, *.sp2 and *.spd files are text files. Data, corresponding to the SPR1 window, are saved to *.sp1 files, and data, corresponding to the SPR2 window, are saved to *.sp2 files.

The *.SP1 file has the following format:

The first string contains the following service information: window name (spr1), number of the points, initial and final OX axis values (in angular minutes), initial and final OY axis values (in relative units), information about the coordinates, which the data and correction value (in angular minutes) are recorded in.

Information about the coordinates, which the data is recorded in, can have the following values: **degree** or a.m. – information is recorded in the relative angles; **deg.(air)** or

a.m.(air) – absolute values of angles of incidence of light on the prism; **deg.(glass)** or **a.m.(glass)** – absolute values of angles of departure of light from the prism.

The second string consists of the headers of data columns located below.

In the first data column angle values are recorded in <u>tens of</u> <u>angular seconds</u> (e.g. 120 means 1200 angular seconds or 20 angular minutes), in the rest of the columns signal values in the enabled channels are recorded in relative units. If the column is filled with zeros, the channel corresponding to this column is disabled.

The *.SP2 file has the following format:

The first string contains the following service information: window name (spr2 or spr3), number of the points, initial and final OX axis values, initial and final OY axis values, information about the coordinates, which the information is recorded in.

Information about the coordinates, which the information is recorded in, can have the same values as for the **SPR1** window (see above).

The second string consists of the headers of data columns located below.

In the Multiple measurement and Track modes time in seconds is recorded in the first column, angle corresponding to the minimum of the 1 channel in tens of angular seconds is recorded in the second column, angle corresponding to the minimum of the 2 channel in tens of angular seconds is recorded in the third column, subsidiary external channel data is recorded in the fourth column.

In the Slope mode in the first column time is recorded in seconds, if the measurement delay exceeds 1 second (**Period**, **s** exceeds 1); otherwise, time is recorded in 0.1 s. Signal values in the 1 and 2 channels are recorded in the second and third columns, respectively, and subsidiary channel data is recorded in the fourth column.

*.**SPD** file format differs from the *.**SPx** file format in the following way:

- angular values are recorded in degrees (with three symbols after the decimal point), not in the tens of angular seconds,
- every twentieth measured point is recorded to the file corresponding to the **SPR1** window, i.e. a "decimation" procedure of the SPR curve is carried out automatically while recording the file.

The **Subsidiary** window data are saved simultaneously with data contained in the SPR2 window in the same file.

The **Notes** window data are saved simultaneously with data contained in the **SPR2** window in a separate file, which has the same name as the **SPR2** data file but with the *.**txt** extension.

Open File 🖶

Reading data files, previously saved (see Save file) in the text format (*.SP1, *.SP2, *.SPR or *.SPD).

When this command is sent, the program requests a file name, opens corresponding windows for the selected file and loads the data from the file in these windows. Working with these data has no difference from working with the experimentally obtained data (see Data presentation). It is possible to load data files in different ways. If the View window is opened, then the files are loaded into this window. If this window is not opened, then the file is loaded into the SPR1 or the SPR2 window depending on the file type.

The following actions are carried out additionally when loading the SPR2 data file:

- search for the **Subsidiary** window information in the file. When such information is found, then this window is opened.
- search for the file with **Notes** window information. When such file is found, then this window is opened.

If the Multiple measurement mode has been operated with the Automatic data saving mode switched on, then auto saved SPR curve files can be also viewed (see Viewing the automatically saved files).

Viewing the automatically saved files

Reading data files, previously saved automatically in the Automatic data saving mode.

If the <u>Multiple measurement</u> mode has been operated with the **Automatic data saving** mode switched on, then auto saved SPR curve files can be also viewed. To do this, proceed as follows:

• Use the <u>Open file</u> command to open the file containing the kinetics data (sp2). The SPR2 window that contains the kinetics plot will be opened.

- Switch on the <u>SPR2 Marks</u> mode if it has not been set.
- With the CTRL keyboard key pressed, click on the point of interest on the kinetics curve with the left mouse button. If the SPR curve file that corresponds to this point is found, the SPR1 window will open and the curve will be loaded in this window. In case the file is not found, the program will display a corresponding warning message.

Alternatively, the automatically saved SPR curve files can be viewed as <u>usual files</u> in sp1 format.

Data processing

This instrument is constructed according to the Kretschmann configuration with sample illuminated by parallel polarized laser beam. Angular distribution of intensity of the reflected light is determined during the mechanical rotation of the measuring prism. Precision mechanical rotation system and built-in fine angular position determining system allow determining absolute angles of incidence of light.

The following information is used during the calculation of absolute angles of incidence:

- The result of angular positioning of the system with respect to the inner reference point (**Zero position**), which is carried out on submitting the <u>Device calibration</u> command.
- Prism parameters specified by user on the **Advanced** page in the **Device** window.
- Absolute angular position correction of the inner reference point defined by the <u>manufacturer</u> and stored in the Devicedat.dat file.

The angles are recalculated directly in the course of measurements and used for plotting the curves. The following information is used depending on the options setting:

- turning angles of the prism table, if the **Relative angle** mode is set;
- angles of incidence of light on the input prism face in the **Absolute Angle in Air** mode;
- angles of incidence of light on the top prism face in the **Absolute Angle in Glass** mode.

The same angles will be saved in the data file created on submitting the <u>Save file</u> command or during the files auto save.

Data processing is executed both immediately during the measurements and when processing obtained results.

During the device operation the following information processing is executed.

The kinetics is determined as the change in the angular position of the minimum of the SPR curve during the kinetics measurements in the Multiple measurement and Track modes. Several methods are used to determine the angular position of the minimum:

Absolute minimum – position of the minimum is calculated as the angular position of the absolute minimum point of the SPR curve. This method is the most reliable and stable, but not very precise.

Parabolic approximation – the SPR curve is approximated to the second-order polynomial function and the minimum of this polynomial function is calculated. It is possible to determine the SPR curve minimum even if the minimum point is out of the measurement range when operating in this mode. This is the most advanced method; it is characterized by the lowest noise of the kinetics curve. This method is defined as default.

Average value – position of the minimum is calculated as the mean value of the centers of several line segments which connect two points that are located at grade on different slopes of the SPR curve. These points are chosen in the vicinity of the minimum.

Cubical approximation – the SPR curve is approximated to the third-order polynomial and the minimum of this polynomial function is calculated. Unfortunately, calculation errors may be sometimes observed when using this method of the minimum determination.

Some versions of the program also use such a method of the determination of the SPR curve minimum as **Center of Mass**. Stability of this method is also lower than of Parabolic approximation method.

Note: The latter three methods are implemented in some program versions only.

Angular position of the SPR curve minimum can be converted to the equivalent value of refractive index of the medium under investigation during the measurement of absolute angles. Virtually, this is simply another kinetics representation mode. Representation modes can be controlled via the pop-up menu of the SPR2 window.

During the kinetics measurement in the <u>Slope</u> mode the kinetics is determined as a change of a signal value due to the SPR curve shift when the device mechanical system is fixed. The kinetic curve can be plotted either as a direct signals change (with or without taking into account the SPR curve steepness in the measurement point) or as a result of conversion of a signals values to the equivalent values of the SPR curve minimum shift. Kinetics representation modes, which are described in the previous paragraph, are available in the latter case. The kinetics

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presentation method can be controlled via the <u>pop-up menu</u> of the SPR2 window or by the elements in the <u>Options</u> window:

Axis in rel. units – kinetics is determined as the signals values that are changed due to the SPR curve shift.

Axis in grad. Corr. – kinetics is determined as the signals values that are changed due to the SPR curve shift, taking into account the SPR curve steepness in the measurement point.

Axis in degree – signals change is converted to the equivalent shift of the SPR curve minimum.

One method of the SPR curve processing is provided.

Averaging – the SPR curve averaging. After switching this mode on, it is possible to define the number of averaged points n. The averaging is made immediately during the measurement and all subsequent operations (such as minimum search, saving data to file etc.) are executed with the averaged values. Thus number of points of the SPR curve is reduced by a factor of n. This mode is available during the single-channel instrument operation only.

Two methods of the kinetics curve processing are provided.

Peak-filter – allows filtrating sizeable single-point peaks on the kinetics curve.

Averaging – switches on/off the data averaging mode. The number of points averaged is determined by **Averaging time, s** and **Number of points** elements.

Number of points – determines the number of points averaged in the <u>Multiple measurement</u> mode;

Averaging time, s – determines the time interval of the measurement during which the obtained points are averaged. In any case the averaging is executed using the "running window" method.

Switching of the modes on is indicated in the <u>Status bar</u> in the main window and it can be changed via the Status bar or via the **Options** window.

The subsequent information processing includes the following possibilities.

Service possibilities, which are provided for the data in the **SPR2** window, are described below. These possibilities can be turned on by selecting the corresponding **Tools** submenu item in the <u>pop-up menu</u> of the **SPR2** window. Only one possibility can be turned on at the same time. The results are displayed in the bottom left corner of the window. All results are presented in the same units the plot is represented in.

1. If the **Coordinate** element is selected, the mouse pointer coordinates determination takes place while the mouse pointer moves over the plot.

2. If the **Distance** element is selected, the distance between two sequentially specified plot points is determined. The distances along the X and Y coordinates are determined separately.

3. If the **Delta Y Line** element is selected, an **Assign first point** prompt appears in the top left corner of the main window. Following the prompt, it is necessary to press the left mouse button and place the arisen dotted horizontal line at the selected level on the plot. An **Assign second point** prompt will appear. Following this prompt, it is necessary to specify the second level in the same way as the first. The result will contain the distance between the marked levels along the Y coordinate.

4. If the **Delta Y Region** element is selected, the distance between the average values of the points which are included in two selected regions is determined. Each region is defined by two outermost points which are specified by user following the **Assign first point**, **Assign second point**, **Assign third point** and **Assign fourth point** prompts, correspondingly. 5. If the **Gradient** element is selected, the curve gradient between two points, which are specified following the **Assign first point** and **Assign second point** prompts, is determined, correspondingly.

Two service possibilities are provided for the data in the **Subsidiary** window. These possibilities can be turned on by selecting the corresponding **Tools** submenu item in the <u>pop-up</u> <u>menu</u> of the **Subsidiary** window.

1. If the **Coordinate** element is selected, the mouse pointer coordinates determination takes place while the mouse pointer moves over the plot.

2. If the **Gradient** element is selected, the curve gradient between two points, which are specified following the **Assign first point** and **Assign second point** prompts, is determined, correspondingly.

Device information

It is possible to receive information about the available prisms and the device number from the **Options/Set Options/Advanced Menu**.

Upon this the **Advanced** page in the **Options** window allows:

- choosing the working prism from the suggested list and, if necessary, introducing corrections for its parameters,
- choosing (or introducing) information about the immersion liquid and the substrate glass.

The **Monitor** page in the **Options** window allows finding out your device number.

If there is a theoretical limitation on the maximum angle of incidence of light on the active layer with the introduced

parameters, you will receive its value on closing the **Options** window.

Command line

One parameter can be specified in the command line of the program call.

The program accepts two types of the parameters.

1. If the command line parameter represents a file name that has the **.SP1** or **.SP2** extension, then the program will open this file in the same way as by the Open file command.

2. On starting the program, it detects the instrument and checks some instrument parameters. If the instrument is not detected, the error messages are displayed. If the "debug" command line parameter is specified, the program will be executed in a special mode, which allows operation without the instrument with some limitations.

Pump control

The device can be completed with NanoSPR-Pump 8channel peristaltic pump for support of probe pumping through the cells. This pump provides pumping velocity in the range of 1 μ l/min.500 μ l/min per one channel.

Pumps management is realized from the "Pump control" window which can be called from the main menu or by pressing the **Pump control** button in the main window.

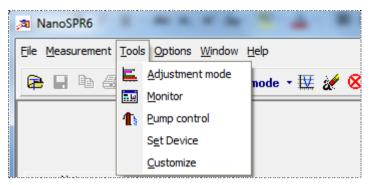


Figure 36Pump control

This window provides the following possibilities:

- 1. Adjustment of the pumping velocity in the range indicated above.
- 2. Measurement of the pumped liquid volume.
- 3. <u>Pumps operation programming</u>.
- 4. Switching on/off the following modes:
 - Probe pump-out (Drain) mode; this is the operational mode.
 - Accelerated pump-out mode.
 - Pump mode (for a short time).
 - Stop mode.
 - Accelerated pump mode.

Note: It is recommended to carry out measurements with the device in the Drain mode only.

There is an element indicating the current probe volume being pumped in the bottom right corner of the window. It is possible to zero the current value by clicking on this element with mouse.

A button that opens the pumps programming panel is located in the top right corner.

A button that switches the "on top of other windows" mode on is located in the top left corner.

As for the rest, the windows interface is intuitively obvious and self-explanatory.

Pumps programming

To program the pumps operation and operate them according to the program it is necessary to call the "Pump control" (or "Rotary Pump control") <u>pump control</u> window from the main menu or by pressing the **Pump control** button. There is a button that opens the pumps programming panels in the top right corner of this window. These panels contain 3 buttons each:

• "**Run script**" – switches on the operation according to the program. Once you start the script, the status with change in left lower corner from "Ready" to currently executed operation. Green bar will present the progress of the current step.

1 Pump Control	
0	Set Port 🛛 🔍
Pump8	Script
O K4 44 ■ >> >>> >> >> >> >>> >>> >>>>>>>>>	🔊 🔇 🚷
— +/- 1	121
Script - Fast_Drain - 1,0 min	

Figure 37Running the script

• **"Stop script"** – stops the operation according to the program.

• "New/Edit script" – allows to develop or edit the program.

The **Edit script** development window is opened upon switching on the development/editing mode.

New program development starts from choosing the required action (Action) by the selection element located in the top service string of the window. The following actions are provided:

- 1. Fast drain rapid pump-out.
- 2. Drain pump-out. This is the main operation mode which is generally used during the measurements.
- 3. Fast pump rapid pump.
- 4. Pump pump.
- 5. Stop.
- 6. Pause.

1 Pump 0 Edit scri	pt			- • ×
Action	10 🕃			<u>N</u> ext line
Fast_Drain A Drain Fast_Pump Pump Stop Pause	Time, min	ul/min	Volume, ul	Save Cancel

Figure 38Pump actions

Elements that allow specifying time of operation (min), pumping velocity (μ l/min), probe volume (μ l) for the selected action are activated in the top service string of the window after selecting an action. After specifying required parameters it is possible to activate the next program string from the pop-up

menu or by pressing the **Next line** button. It is possible to return to previous strings and edit them. To save the created program, choose the **Save** button or corresponding pop-up menu item.

Action Time, min ul/min Volume, ul 1 Drain 5,0 10 50 2 Fast_Drain 500 10 10 3 Drain 10 Image: State S	np 0 Edit so	Edit script			
1 Drain 5,0 10 50 2 Fast_Drain 500 3 Drain 10	Drain	▼ 5 8	10 🛱	92	<u>N</u> ext line
2 Fast_Drain 500 3 Drain 10	tion	Time, min	ul/min	Volume, ul	
3 Drain 10	ain	5,0	10	50	
×	ist_Drain	rain	500		
	ain		10		🗸 Save
					X Cancel
					🗁 Open file

Figure 39Sample pump script

To edit previously created file, call it by pressing the **Open file** button and edit it.

Resetting the pumped volume value.



Figure 40Reset the volume value

The volume window is shown by the arrow. During the operation this window displays an approximate liquid volume that was pumped by one channel. To reset the value click on it with a mouse.

Monitoring and stabilization of the temperature

The following monitoring possibilities are provided in the device:

- Monitoring and stabilization of the cell temperature.
- Monitoring of the power supply voltage.

Switching of the monitoring mode on is indicated by periodical red coloring of buttons/indicators in the "On" column of the window.

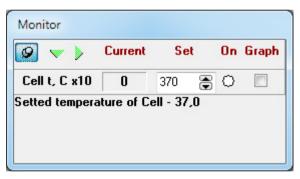


Figure 41Monitoring window

The monitoring mode is switched on upon the <u>Monitor</u> window call that can be called from the <u>main menu</u>: **Tools/Monitor** or by pressing the **Monitor On/Off** button.

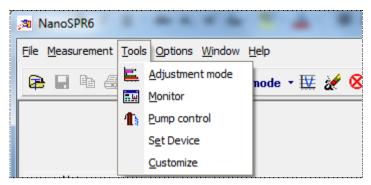


Figure 42Call monitor window

When the monitoring mode is on, the periodical measurements of power supply voltage and temperature of the cell and additional temperature channel are carried out and measured values are displayed in the **Monitor** window. A power supply emergency warning window is displayed upon power voltage decrease to the previously specified value.

The cell temperature stabilization is possible only if the special cell with built-in heater and thermometer is used. Temperature stabilization occurs by means of cell warming so the stabilizable temperature cannot be lower than the ambient temperature.

It is possible to switch the temperature stabilization of the cell and/or additional temperature channel on/off from the **Monitor** window by clicking on the corresponding button/indicator located in the "On" column.

Monitor				
🧟 🔷 🕨	Current	Set	On	Graph
Cell t, C x10	0	370 🕃	0	
Setted tempera	ture of Ce	əll - 37,0		

Figure 43On / Off temperature monitoring

If the **Graph** element has been checked, the **Temperature** window is opened that displays a real-time plot of the investigated liquid temperature.

It is possible to specify the stabilization temperature and power monitoring actuation level from the <u>Monitor</u> page in the **Options** window or directly from the **Monitor** window using elements located in the "Set" column.

Options					×
Multiple Track	Slope Single	Service	Advanced	Monitor	
Cell temperature	e				🗸 <u>о</u> к
Temperature, C	:	Temperat	ure set, C	20.0 🔀	
Temperature 2					X <u>C</u> ancel
Temperature, C	:	Temperat	ure set, C	20.0 🔀	
	I				
Power	1		_		
v		Alarm	set, V 🧕	.0 🔀	
-100 🖨 Te	mper. corr *1000		Device#	030	<u>D</u> efault

Figure 44Monitor options window