

SPD-500[®]

VARIABLE-WAVELENGTH ULTRAVIOLET/VISIBLE DETECTOR

Installation and Operation Guide



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SPD-500 UV/VIS DETECTOR

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Weight	10Kg	27
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1 General information

1.1 Outline

The SPD-500 variable-wavelength ultraviolet/visible detector is a part of entire HPLC system. By combining with MODEL 501/500 HPLC pump(s) or pump(s) manufactured by other producers, it can be used for routine laboratory's analysis and method development. The SPD-500 detector is designed by means of up-to-date technology as digital data processing and controlling, the baseline noise and baseline draft reduce to a new limit range. Due to the digital data output function, the data can direct transmit to computer via RS232 without any acquisition unit.



Fig1. SPD-500 variable-wavelength ultraviolet/visible detector

1.2 Features

- Advanced optical unit design
The SPD-500 detector introduces a new idea in to its optical unit design. As we known, the principle of this kind of detector is based on Lambert-Beer law as the formula described below so the most important thing we want to do is to improve the light path, and get the biggest light source energy both in sample and reference photo-diodes.
Emphasis improvement:
 - 1) Light source:
 - 2) Light path
 - 3) New design filter
 - 4) Concave photographical grating
- Advanced flow cell designed
- All-out digital signal processing and controlling
- New integrated Power-source module, ensure the system working more reliable.

1.3 The basic principle of the UV Detector SPD-500

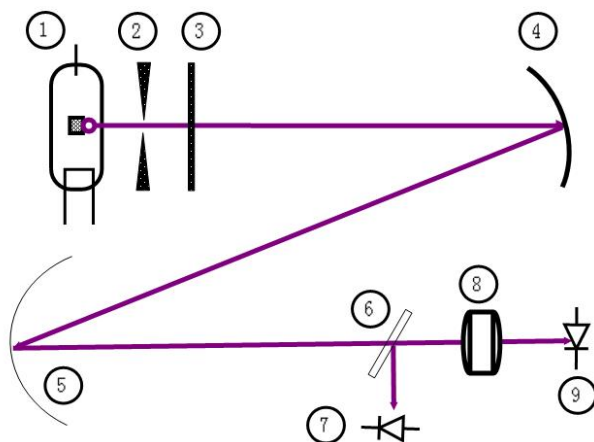


Fig2. SPD-500 Optical path

- 1- Deuterium lamp (or Halogen lamp)
- 2- Slit
- 3- Filter
- 4- Concave mirror
- 5- Concave grating
- 6- Half transparent mirror
- 7- Reference photo diode
- 8- Flow cell
- 9- Sample photo diode

The light beam of the lamp (1) is fading out by slit (2) and then goes through the filter (3). After them passing it, the concave mirror (5) focuses the beam from the slit and reflects the beams to a concave grating (6). The reference photodiode receive half energy from the reflect beams of the half transport mirror and the sample photodiode receive the other half energy go through flow cell and then received by sample photodiode.

General information

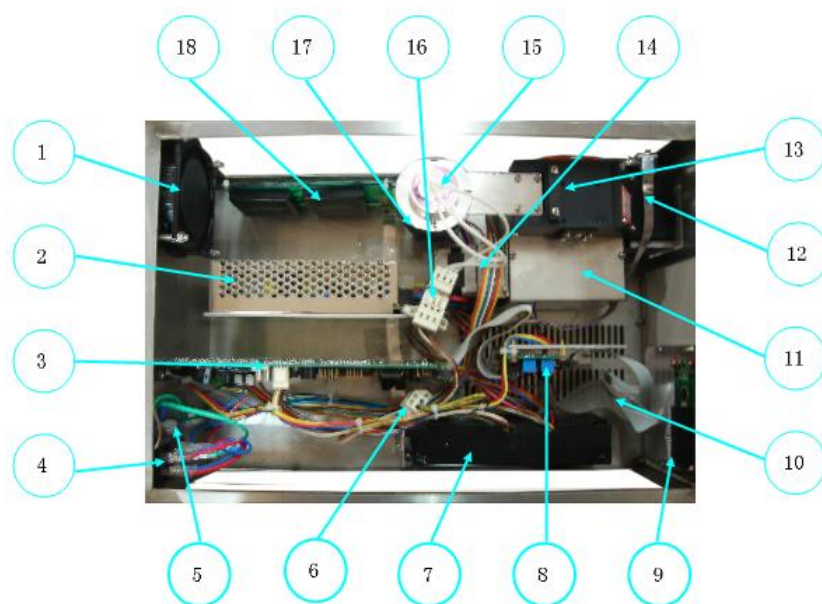



Fig3. Top View of SPD-500 variable-wavelength ultraviolet/visible detector

Note:

- 1—Radiator fan
- 2—Main power source
- 3—Main board
- 4—AC Power plug
- 5—Ground terminal
- 6—Halogen lamp connect plug
- 7—Lamp power source
- 8—LCD(12265B) display driver
- 9—LCD(12265B)
- 10—Cable connect LCD & Keyboard & main board
- 11—Monochromator (include grating and Sine mechanical assembly)
- 12—Grounding strip
- 13—Optical unit
- 14—Step motor
- 15—Lamp(replacement Halogen lamp or Halogen lamp)
- 16—Halogen lamp connector
- 17—Lamp radiator
- 18—DC-DC convertor (regulator)

The entire detector was show above. Usually, it can be described as control and data process unit, optical unit, power source unit and display unit.

Detector control and data process unit is designed for detector's controlling such as keyboard input, RS-232 communication, photoelectric conversion, and data calculation etc. It allows signal transmission from the two









photodiodes to the instrument out put terminals with digital operation. As matter of fact, the detector can out put digital signals directly to PC without any A/D interface. At the mean time, it also supply analog out put for traditional users.

The optical unit consist of lamps, Monochromater, flow cell as it's shown in the figure above.

The integrated main power module designed with explosion protection supply power used for microprocessor (+5V,2A) and the analog chips in the main board and pre-amp boards($\pm 15V, 0.5A$).

2 To use this manual

Symbols in this manual	Implication
	Applied in a case that could result in slight injury or machine damage.
	Applied for improvement of operating efficiency or help in understanding.
	Special indicate the arrow key "LEFT" in the front panel.
	Special indicate the arrow key "RIGHT" in the front panel.
	Refer to

-  Read the instruction manual thoroughly before you use the product first time. If you have an experience of using HPLC pump, the Chapter 1~4 can be omitted.

3 Installation Precautions

3.1 Installation environment

ⓘWarning

To take advantage of the SPD-500 performance capabilities and to ensure its operational stability over a long service life, check that the selected installation site satisfies the following requirements.

- a. Ventilation
Ventilate the room where the high performance liquid chromatograph is located since the solvent used is flammable and/or toxic.
- b. Fire
Never use fire in the same room where the high performance liquid chromatograph is installed. Also, avoid installation in the same room of other devices which may spark. Always keep a fire extinguisher nearby in case of accident.
- c. Sink
Install a sink nearby for flushing eyes or skin which have been in contact with solvent.
- d. Corrosive gas and dust
Avoid installation in a place exposed to corrosive gases or dust.
- e. Electromagnetic noise
Avoid locations subject to intense magnetic or electromagnetic fields. Use an additional noise filter if power line noise interferes.
- f. Space requirements
This system is designed to be used on table or stand, preferably a solid and flat surface with depth of 100cm or more.
- g. Others
Select an installation site with the following parameters to maintain full performance of the system.
 - A) Maintain room temperature within 4~35°C, without extreme fluctuations.
 - B) Avoid direct output of a heater or a cooler.
 - C) Avoid exposure to direct sunlight.
 - D) Avoid locations subject to strong vibrations or prolonged weak vibrations.
 - E) Maintain relative humidity with 45~85%.

3.2 Unpacking

After the instrument unpacking, check the device integrality according to

Installation Precautions

the Standard delivery packing list thoroughly. and then check if there is any damage during transport. If necessary, put forward any claim for damages to the carrier to ask for compensating.

Standard delivery packing list for SPD-500

1. SPD-500 variable-wavelength ultraviolet/visible detector
2. User's manual
3. Power supply cable
4. RS-232 Cable (Special made for SPD-500)
5. Analog connection cable

4 Operation

4.1 Power on SPD-500 detector

Use AC power supply cable in the STD packing list to connect the detector and AC power socket. Be sure that there must be good grounding.

ⓘWarning

The requirement of AC power can be shown on the rear panel of the detector. Incorrect AC input may lead to the instrument damage or injury of human body.

Switch the power on (position 1), The display then light and execute Self check. It will last for one more minute.

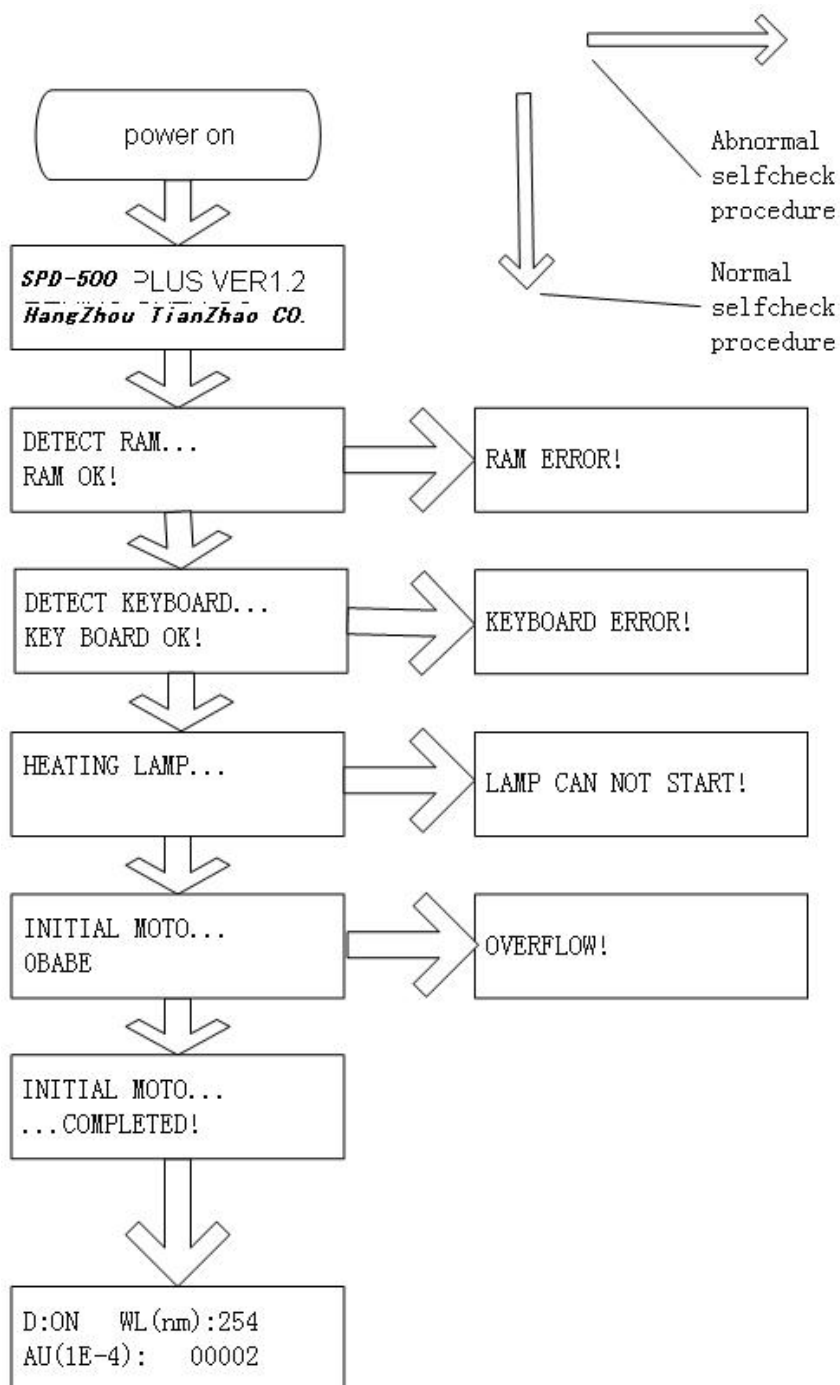


Fig.4 SPD-500 boot procedure

After the instrument self check, the main menu will be shown. At this time, you can input the system working parameter such as wavelength, time constant, signal output range and etc.



Refer to 4.2 “Menu” .

The instrument need approximate 15~30min for additional stabilization and lamp warm up. In very common situation, longer time for instrument warm up, more stable baseline you will get.

Up to now, the detector is ready for use.

☞ In case of error appears in the screen, please check the instrument according to “Troubleshooting” in this manual or contact with the factory.

4.2 Basic Operation-keyboard and LCD display

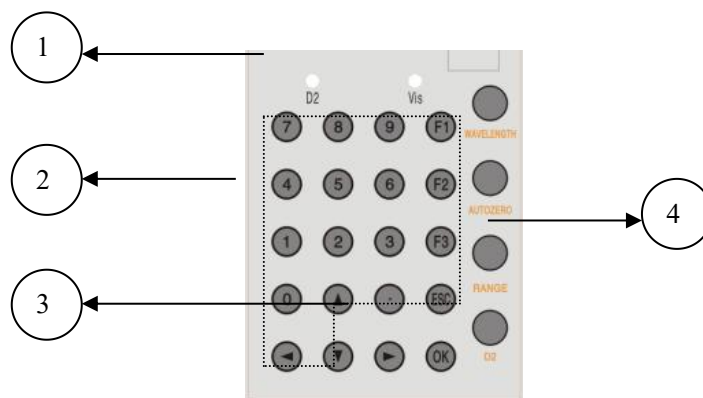


Fig.5 SPD-500 keys and screen

Note:

- 1—LCD display(Model:16265B),.
- 2—Numeral Keys area.
- 3—Arrow Keys area.
- 4—Function keys area.

In order to operate The SPD-500 smoothly and correctly, the operator must have a basic comprehension to the 4 area described above. Here is a brief explanation:

LCD display is a 16 characters, 2lines display module. Menus can be changed by press left key or right key. Every click of arrow keys can move the cursor to previous position or next position. There are six menus included in the SPD-500 design corresponding to “main menu”, “Range & TC menu”, “Initial WL and Gain menu”, “Sample & Reference intensity menu”, “GLP-s/n and CMC permission Number”, “GLP-System operating time & Lamp time”. The menus can be active circulatory by press direction arrow keys continually. Numeral Keys indicate from 0 to 9. They are used to input numerical value where necessary. Such as wavelength value, Range value, serial number and etc.

Arrow keys are used to change displayed menus. For example, if users want to see what the situation now about the sample intensity and reference intensity, he can click the right arrow key again and again, till the menu appears.

There are four functional keys list on the right side of the front panel. They are wavelength input key λ , full scale automatic zero key A/Z, full scale analog and digital output setting key RNG and confirmed key Enter.

These functional keys are shortcut keys. For example, no matter where is the cursor located at, the cursor will jump to wavelength input area when simple click λ key. Specially, the A/Z key can be used for zero the signal at any time when the detector is running. On the other hand, this key can be used for

Operation

shortly returning the main menu.

4.3 Menus

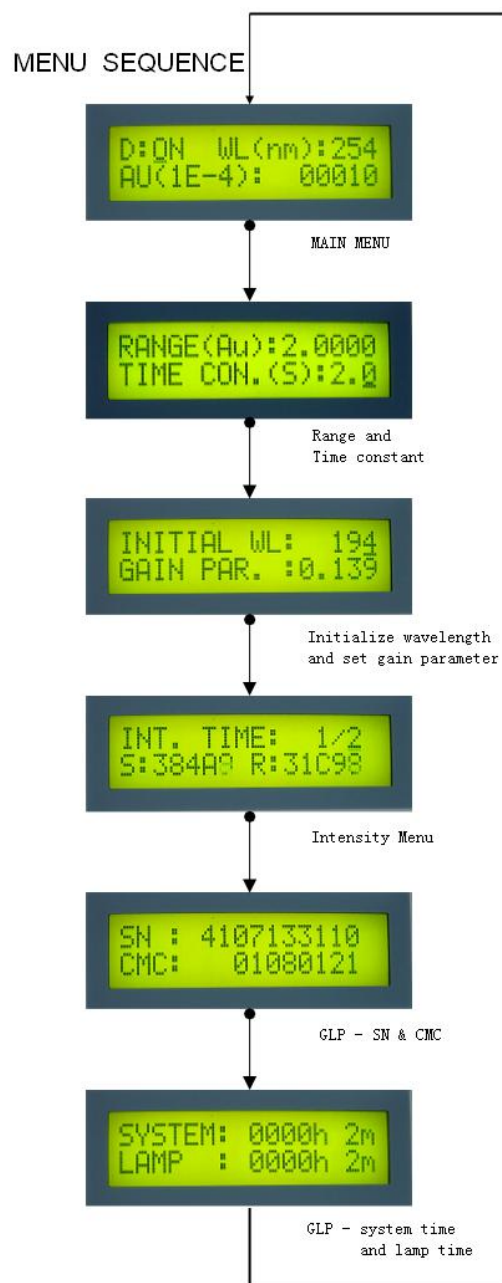


Fig 6 Menu Sequence

4.3.1 Main menu



Operation

Fig.7 Menu "Choose lamps"

Refer to above screen capture, the cursor can be located at three positions:

1) Choose lamps

When the cursor stay at "D:" or "H:" and blinking, it prompt you that you can change lamps from deuterium lamp to halogen lamp or from halogen lamp to deuterium lamp by means of numeral key "1" or "0". The indicated character "D" or "H" is the ab. of current lamps and individually corresponding to "0" or "1". Do remember, you must press enter key to confirm changes

① Be sure that the chosen lamp on the screen must correspond to the actual lamp used in this detector. If not, the lamp may not be lighted up or may cause damage to the lamp power unit.

2) Turn on or turn off lamps



Fig.8 Menu "Turn on or turn off lamps"

The deuterium lamp has its lift time. For more long lift time, we recommend users turn off lamps after analysis.

When the cursor stay at "ON" or "OFF" position, if you press numeral key "0" or "1", the lamps will be turned off or turned on correspondingly. Do remember, you must press enter key to confirm after above processing.

① Frequently turn on or turn off the lamps may reduce the lifetime of the lamps. So, in situation of continuously analysis, this function does not recommend.

3) Change wavelength

When the cursor stay at the position behind "WL(nm):" and blinking, you can input a new wavelength value you desired and then press enter key to confirm finishing new wavelength execution.

The wavelength can be selected within the range from 190 to 700 nm in 1nm steps.

① The wavelength change was carried out by the step motor. So, for larger scale wavelength shift, you should wait for some seconds.

4.3.2 Range and TC menu



Fig.9 Menu "Range and TC"

Operation

When the cursor stay at RANGE(AU) value position and blinking, you can input a new RANGE parameter by means of the key pad 0~9.

The concept of Range is a setting full scale analog or digital output range. E.g. 1Au/full scale means 1Au corresponding to full scale 2000mV.

You can change the Range parameter by input a new numeral value.

Detailed relevant input please refers to table below:

10.00	5.000	2.000	1.000	0.500	0.200	0.100	0.050
15	14	13	12	11	10	9	8
0.020	0.010	0.005	0.002	0.001	0.0005	0.0002	0.0001
7	6	5	4	3	2	1	0

Table1: Initial wavelength calibration and Gain setting menu

① The RANGE parameters take effect to the signal out put either digital or analog. Too small value of this parameter you set may cause to large baseline draft or baseline noise. Sometimes it may lead overflow of the Data acquisition system.

TIME CON. Is the ab. of time constant. This parameter can smooth the signal. The T value can be set to 0.1, 0.2, 0.5, 1, 2, 5, and 10 seconds. The large T value you set the more smoothed signal you will get. In most common analysis, the T value 1 or 2 is recommended.

① Too large of T value may cause the reduction of the peak height and enlargement of the peak width.

4.3.3 Initial wavelength menu



Fig.10 Menu "Initialize wavelength"

This menu is designed for instrument adjustment in house or for salted service engineers.

The value of initial is password protected.

The purpose of this parameter is designed to amend the wavelength accuracy. If you want to change the initial wavelength, you must recalibrate the wavelength by Holmium glass filter, and get the scan curve and then input a new value by press password "246805" first.



Detailed procedure please refer to SOP.1

4.3.4 Gain parameter menu



Fig.11 Menu "The Gain parameter"

The GAIN parameter is designed for instrument adjustment in house or for salted service engineers. The concept of this parameter is creating a relevant value from absorption value Au to the signal read out value mV.

This parameter is password protected.

Gain parameter is the gain calibration parameter. To calibrate this parameter, 0.13% acetone and water solvent is needed.

The value of initial is password protected.



Detailed procedure please refer to SOP.2

4.3.5 Intensity Menu



Fig.12 Menu "Intensity"

The INT. TIME is the integration time in the AD converter between light intensity and signal converted. The larger INT. TIME parameter you set, the stronger signal you will obtain. Usually, this parameter is pre-set in factory and it's also being interest in case of servicing.

The INT.TIME parameter is password protected.

The presented value S and R indicate the light intensity of the Signal and Reference channel in HEX value respectively.

In case of problem happens, we can diagnose where is the trouble in by means of the value of R & S.



Detailed procedures please refer to Chapter 6 Troubleshooting.

4.3.6 GLP menu

The GLP menu includes 2 pages.

First, the instrument series number SN and China metrology Certification

Operation

number of current product SPD-500 (CMC). They are only for view and not allow modifying in any case by the end user.

The second page show you the System total operating time (SYSTEM) and the current lamp operating time (LAMP).

They are only for view and can not allow modifying in any case by the end user.

Once there are some problems occurring, the information in above 2 pages will be interested in.



Fig.13 Menu "GLP"

① After changing lamp, the lamp time can be reset by our servicing engineer.

5 Standard Operation Procedure

5.1 SOP1. Initialize the wavelength

- a) Change the flow cell with a Holmium glass filter cell and then turn on the detector.
Open the data acquiring system (N2000) to get a stable baseline with wavelength 254nm.
- b) Set the detector parameter as below:
Time constant: 1s
Range: 1AuFS
- c) Run scan program by input conceal keys 012345 and then enter.
- d) The scan will automatically trigger a start signal in the data acquisition in N2000.
- e) Save the curve as "C_{holmium}" in a predefined folder.
- f) Pull the Holmium glass out of the cell and repeat the procedure c) ~e), save the curve as "C_{blank}" in the same folder as described above.
- g) Use C_{holmium} minute C_{blank}, and get a curve as described below.
And then determine the wavelength for Peak1, Peak2, Peak3.

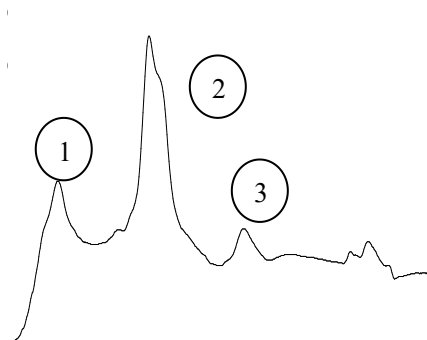


Fig.14 Scan curve for Standard Holmium glass

- h) The three wavelength Holmium characteristic wavelength respectively should be 361nm, 446nm and 536nm.
- i) Determine the maximum differences and adjust the initial wavelength input value.
- j) After finish the value input and press enter, the detector will re-calibrate the wavelength automatically.

Standard Operation Procedure

5.2 SOP2 Build up gain parameter

- a) Connect system with a pump and a detector by capillaries (without injection valve and column) as figure below.

Fig.15 System connection for determining gain parameter

- b) Prepare 200ml 0.13% degassed acetone water marked as A and 200ml pure water marked as B.
- c) After system warm up, build up a stable baseline at least 2 minutes with flow rate 3ml/min by solvent B and use A/Z key to force the baseline nearby zero position.
- d) Change the solvent by solvent A, the absorbance value displayed in the main menu will increase, modify the gain parameter according to the read data. If the read data higher than 112mAu, then decrease the gain value. The new gain value approximately to the function below:

$$\text{gain_value} = \text{old_gain_value} \times \frac{\text{read_data}}{112}$$

Here: gain_value is the gain parameter you have to input

Old_gain_value is the gain parameter you have preset.

The read_data is the absorbance value read out from the screen in mAu.

- e) Repeat d) tow times, you will get the end exact gain parameter value. The system store this data automatically after the main power down.

Standard Operation Procedure

5.3 SOP3. Change the Deuterium lamp



Fig.16 Deuterium lamp



Fig.17 Deuterium lamp assembly

- a) Power down the detector and remove the power plug. Let the lamp cool down at least 15minutes.
- b) Open the cover by loosening double side screws using a cross screwdriver.
- c) To find where is Lamp and lamp holder. Loosening the two screws on the lamp holder. Disconnect the three-pole lamp cable by hand.
- d) Remove the old (or failed) lamp carefully direct from the lamp holder.
- e) Put a new deuterium lamp direct to lamp holder, justified the two screw holes and tighten the two screws by cross screwdriver.
- f) Plug the three-pole cable adapter of the new lamp in to the specified three-pole socket by hand.
- g) Put the instrument cover over the machine and tighten the 4 screws by cross screwdriver.
- h) Connect the main power cable to the detector socket, and then power up.
- i) Reset the Lamp time in the GLP menu. (Please ask the password from the original inspection record in the factory)

① The radiation rays from the Deuterium lamps are harmful to human eyes.
Do not look at the light directly without use any UV preventable glasses.

① If the change is from original halogen lamp to deuterium lamp, don't forget to Change the H to D sign from the main menu.



Please refer to 4.2.1

Standard Operation Procedure

5.4 SOP4. Change the Halogen Lamp

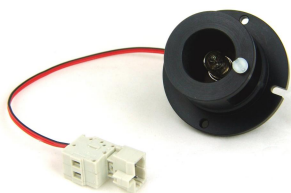


Fig.18 Halogen lamp

- a) Power down the detector and remove the power plug. Let the lamp cool down at least 15 minutes.
- b) Open the cover by loosening double side screws using a cross screwdriver.
- c) To find where is Lamp and lamp holder. Loosening the two screws on the lamp holder. Disconnect the two-pole lamp cable by hand.
- d) Remove the old (or failed) lamp carefully direct from the lamp holder.
- e) Put a new halogen lamp direct to lamp holder, justified the two screw holes and tighten the two screws by cross screwdriver.
- f) Plug the two-pole cable adapter of the new lamp in to the specified two-pole socket by hand.
- g) Put the instrument cover over the machine and tighten the 4 screws by cross screwdriver.
- h) Connect the main power cable to the detector socket, and then power up.
- i) Reset the Lamp time in the GLP menu. (Please ask the password from the original inspection record in the factory)
- j) Check the sample and reference channel intensity in the INT. menu.

① If the change is from original deuterium lamp to halogen lamp, don't forget to Change the sign from D to H in the main menu.

☞ Please refer to 4.2.1

5.5 SOP5. Disconnection & Installation of flow cell

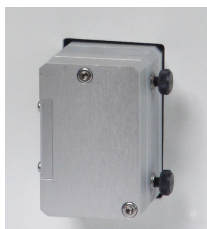


Fig.19 Flow Cell assembly

To disconnect the flow cell from the SPD-500:

- a) Power off the main power switch.
- b) Loosening the front two hex bolts by a special hex wrench (s=3) attached in

Standard Operation Procedure

- the STD packing kit.
- c) Loosing the two fluid connectors in the flow cell up side to the column and the under side to the waste.
 - d) Pull out the flow cell from the Cell assembly. Be careful, don't blur the photo-diode in the front part of the cell assembly.

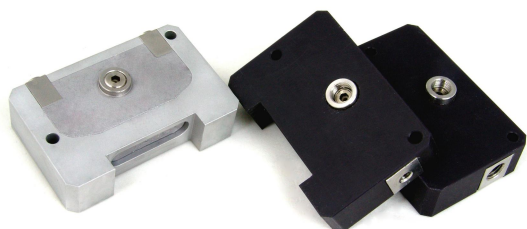


Fig.20 Flow Cells

To install a flow cell to the SPD-500:

- a) Put the flow cell assembly to the cell hopper. Justify the orient by tow mounted holes.
- b) Put the two hex bolts in to the cell assembly and then tighten them by the special hex wrench.
- c) Connect the fluid connector to column and fluid connector to waste.
- d) Power on the detector, and purge the flow cell by a pump using Methanol+water (80:20) to remove the air bubbles inside the flow cell at least 15minites till the stable baseline can be obtained in the data acquisition system.

① If it remains some air bubbles inside the flow cell, the baseline should be fluctuation all the time when delivering of solvent. Be sure that there is no air bubbles inside the flow cell. If necessary, please refer to "Cleaning flow cell".

5.6 SOP6. Cleaning flow cell

- a) Connect the pump and flow cell directly by a bypass capillary.
- b) Enter the detector's INT menu, and write down the intensity value of the sample channel.
- c) Prepare at least 100ml 1Mol NaOH as cleaning solvent.
- d) Turn on the pump and start solvent delivery with flow rate 3ml/min at least 15minutes.
- e) Change the solvent to pure water, then start pump, rinse the flow cell and liquid line till the pH value to 7 by the pH test paper.
- f) Change the solvent to methanol, then start pump, rinse the flow cell and liquid line for 15minute.
- g) Remove the bypass capillary instead of valve, column and so on.
- h) Enter the detector's INT menu again, and to see the situation in the sample intensity. In most common case, the intensity will be increased after the cell cleaned.

6 Troubleshooting

Malfunction	Cause	Remedy
1. No display	Main power fuse burn out	Change new main power fuse
	Check +5V power out put	Change main switch power module
	Check if there is no beep while the instrument power on	Check the main board if the CPU or CPLD module failed otherwise change the main board
	Display module failed	Change new display module
2. Characters in the screen can not be identification	EEPROM witch used to store data may be confusion by transient voltage	Initialize the EEPROM by input password (ask for it from factory)
3. Keyboard detect error	Keyboard Interface abnormal Or keyboard control chip initialize error	Check the connector & cable from main board to keyboard And then restart the instrument If the trouble still occur, please contact the local representation of the factory
4. RAM detector error	RAM chip on the instrument main board work abnormal or power up failed.	Restart the detector, if the problem still occur, change the RAM chip on the main board. Otherwise, please contact with factory
5. Deuterium lamp not start	Inspect the detector parameter where in the main menu if the lamp is set to "D"-deuterium Refer to	Change the lamp choosing position "H" to "D"
	The power of lamp is failed	Inspect the lamp power, if the filament voltage, the anode voltage and trigger voltage working properly
6. Halogen lamp not start	Inspect the detector parameter where in the main menu if the lamp is set to "H" Refer to	Change the lamp choosing position "D" to "H"
	The power of lamp is failed	Inspect the lamp power if the filament voltage working properly.
	LCD display module is failed	Change LCD display module
7. "OVERFLOW" occurs while instrument self check	The light intensities on the reference and sample photo-diodes are too strong	Check if the filter work properly while instrument self check
	One of Pre-amplifier is failed	Change the pre-amplifier board by means of the situation on the menu "INT."

Troubleshooting

8. No signal comes out	Lamp is not lit. The two channel light intensity are very weak	☞ Refer to 5,6
	If use digital out put, Check the communication from instrument digital signal out put to computer	If necessary change the cable or change the communication chip(Max203) otherwise contact to the factory
	Either the reference or the sample pre-amplifiers defect	Change with new one
9. Noise too much big	The range setting is too much small	Find out a properly range parameter, and re-input it. Refer to "menu range"
	The time constant parameter set too small	Change the parameter to a receivability(1 or 2)
	The lamp exceed or near its lifetime	Change new lamp
	The flow cell is too dirty One should see the R & S	Cleaning the flow cell
	Air bubble in mobile phase	Degas the mobile phase Or increase the backpressure at the fluid out port.
	Instrument not grounding well	Reconnect the ground strip
	Abnormal wavelength setting Too low wavelength may cause too much noise	Properly set the correct wavelength
10. High draft	Mobil phase alternate	If the system works in a gradient situation, the draft can be acceptable otherwise rinse the capillaries and cell thoroughly
	System need too much time for warm up	Wait for the lamp and electrical elements stable.
	Column does not balance well	Spend more time balancing columns
	Strong air current lead to temperature does not stable	Inspect the installation site, put the detector in a properly place.
	The fluid has slight leak Inspect the difference after stop delivery	Try to find where is the source of leak and tighten the screws.
	Impurities in the mobile phase Inspect the difference after delivery stop.	Check the pump, column, reserver and mobile phase for dust or impurities
	Dead volume exist in the fluid line may cause the components effluence slowly Then the baseline wave	Check the connector locate at valve, the input port of the cell if the capillaries correct butt joined

7 Integrate the SPD-500 detector in to a LC system.

The minimum integrated HPLC system as we know must include 5 individual modules such as HPLC pump(s), a injector (manual or auto sampler), a column(column oven), a detector and a chromatographic workstation(PC & software). Some other additional article as capillaries, mobile phase, solvent reservoir, waste reservoir and so on.

8 Controlling and acquiring by software N2000

9 Specifications

Flow Cell volume	10 μ l
Wavelength Range	190-700nm
Light Source	Deuterium Lamp and Halogen Lamp(alternatively)
Range of measurement	0-2Abs
Time Constant	0.1/0.2/0.5/1.0/2.0/5.0/10.0s
Wavelengths accuracy	± 2 nm
Baseline noise	1.5×10^{-5} Au (Methanol/water 80:20 FlowRate 1ml/min $\lambda = 254$ nm)
Baseline draft	4×10^{-4} Au/h (Methanol/water 80:20 FlowRate 1ml/min $\lambda = 254$ nm)
Signal out range	16 steps selected
AutoZero	Full-scale Autozero
Display	2x16 Digital
Control	Key pad RS232 interface Remote connector (trigger of start acquisition)
Main power	150W
Weight	10Kg
Dimension	398 mm \times 149 mm \times 267mm
GLP	Record system operation time & lamp operation time automatically