

Fluorescence Detector RF-2000

Operating Instructions



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CE

DECLARATION OF CONFORMITY

We, Dionex Softron GmbH, declare that the product Fluorescence Detector

RF-2000

to which this declaration relates, is in conformity with the following European standards:

# EN 50081-1 (1992):	Electromagnetic compatibility - Generic emission standard Part 1: Residential, commercial and light industry
# EN 50082-1 (1992):	Electromagnetic compatibility - Generic immunity standard Part 1: Residential, commercial and light industry

Attention Dionex Softron will not accept any liability for damage, direct or indirect, caused by connecting this instrument to devices which do not meet relevant safety standards.

January 27, 1998

QM-Manager

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1 Precautions

The Dionex **RF-2000** is the fluorescence detector for the high performance liquid chromatograph. In order to operate the unit safely, strictly observe the following points. It is dangerous not to comply with the following points.

- Use the unit for any purpose mentioned above.
- Follow the procedures described in the operating instructions.
- Observe the warnings and cautions.
- Do not disassemble or modify the unit without approval from Dionex. Failing to do so may lead to a dangerous situation or damage to the instrument.
- For internal repair of the product, contact the Dionex Service .

1.1 Warnings in these Operating Instructions

Warning:	Indicates that failure to take note of the accompanying information may result in personal injury.
⚠ Important:	Indicates that failure to take note of the accompanying information may cause wrong results or damage to the instrument
Please note:	Indicates General information to assist you in obtaining optimum performance.

1.2 Warning Labels on the Unit

For safety of operation, this unit is provided with the mark at the portion where special cautions are required. When operating the parts where this mark is indicated, exercise special caution after reading the operating instructions.

Warning:

- The xenon lamp may explode in the course of long use (more than 1000hours). Replace the lamp of expired life (500 hours) with a new one.
- Be sure to wear protective gears, such as protective mask, heavy and longsleeved shirt and gloves, when handling the xenon lamp. Since high pressure gas is charged in the xenon lamp, if the lamp is strongly shocked or the glass surface is scratched, the lamp may explode and pieces of glass may scatter.
- When mounting or replacing the xenon lamp, be sure to turn OFF the power switch and disconnect the power cable. Voltage is applied to the electrode by the electric charge right after the power switch is turned OFF. Therefore, leave the xenon lamp for more than five minutes after turning the power switch OFF before opening the cover of the light source chamber.
- The xenon lamp is very hot when it is lit. Therefore, when replacing the xenon lamp, wait for more than an hour after turning the power OFF to cool the lamp enough for replacement.
- High voltage (approx. 30 kV) is applied to the xenon lamp during ignition. When turning ON the power, be sure to mount the lamp and close the cover of the light source chamber.



▲ Important:

Be sure to read this instruction manual before supplying power to this unit.

- Be sure to mount the xenon lamp correctly before supplying the power to this unit (refer to 4.7 Installing the Xenon Lamp). Failing to do so may damage the main unit because high voltage around 30kV is applied to electrode when turning ON the xenon lamp.
- Use the power supply voltage at the state which conforms to the power supply setting (display of the power supply connector unit at the rear) of this unit. For changing the power supply setting of this unit, refer to 8.4 Changing the Set Power Supply Voltage. However, when the voltage is 220V \pm 10%, set the power supply voltage of this unit at 230Vac.



1.3 Replacement of Fuses

This unit uses the following fuse. Be sure to replace the fuse of the same type and capacity.



⚠ Important:

When the xenon lamp is lit, the flow cell is irradiated by the excitation light beam. Do not look into the excitation light beam, while the xenon lamp is lit.



1.4 Precautions on Installation Site and Handling of the Unit

Generally, a large amount of organic solvents is used in the high-performance liquid chromatograph. Sufficient care should be taken in installation site and handling of the unit. Please take care of the notices in the text, not to mention the precautionary requirements listed below:

1.4.1 Ventilation

Solvents used in the high-performance liquid chromatograph are inflammable and/or toxic. Be sure to ventilate the room well.

1.4.2 Fire

The lighting of fires is prohibited in the room where high-performance liquid chromatograph is installed. Do not install any devices which may emit sparks in the room. It is also necessary to provide a fire extinguisher preparing for the worst.

1.4.3 Safety goggles

Wear safety goggles in handling solvents.

1.4.4 Other equipment

A washstand or the like is necessary to be furnished nearby preparing for the cases such that solvent comes into the personnel's eyes or the personnel touches toxic solvent.

1.4.5 Power source

Working voltage range and power consumption of this system are as follows. Be sure to connect to the power source conforming to them.

Part No. 706.228.35353.38, 220-230V ~ ±10%/240V ~ ±10%, 350VA, 50-60Hz

If working with a different working range, different fuses must be used! Please contact the Dionex Service.

1.4.6 Grounding

To prevent electric shock and to secure safe operation of the system, always make grounding.

1.4.7 Repair and maintenance of the unit

Normal maintenance of this unit can be performed without removing the cover. Do not remove the cover at the normal maintenance. Contact the Dionex Service if repair requires to remove the cover of the main body.

1.5 Precaution in the region where 240V is used as working voltage

1.5.1 Confirming the working voltage

The voltage setting must correspond to the working voltage used. When the working voltage is between 220V and 230V, it is not necessary to change the factory setting (230Vac). However, when the working voltage is 240V, change the voltage setting as follows:

- (1) Turn OFF the power switch.
- (2) Pull out the power supply cable from the power supply connector.
- (3) Open the fuse holder cover at the rear of this unit by a slotted screwdriver.



(4) Change the rotary switch showing the voltage in the fuse holder to display **240Vac**.



(5) Close the fuse holder cover until a clicking sound is heard.

1.6 Precautions on Static Electricity

Liquid chromatography using flammable organic solvents as mobile phase requires proper care against fire, explosion, etc. Particularly, among various possible accidents, those caused by static electricity are difficult to anticipate, and tend to occur only with unexpected conditions which often make countermeasures insufficient. At a site where preparative liquid chromatography is practiced, a large amount of flammable substances may be used. Therefore, once an accident happens, it could lead to tremendous damage. The mechanism of accident caused by static electrical discharge and preventive measures are described below. Take due care in safety measures in handling of equipment.

1.6.1 Mechanism of Static Electrical Discharge Accident (Example)

Accidents caused by static electricity take place through the following processes.

When liquid is fed at high speed through a small-diameter tube like the pipe of a liquid chromatograph, static electrical charge occurs by friction between solid and liquid as shown in the figure below





When the charged liquid is collected in an insulated vessel, the static charge accumulates gradually, and the voltage can easily reach a few kilovolts. If some other conductive object is brought near the vessel, electricity is discharged at a certain distance from the vessel releasing heat energy. If flammable gas of sufficient concentration exists nearby, ignition is caused by this energy.



Fig.: Conditions which may cause accidents

1.6.2 Preventive Measures against Accidents

The principal preventive measure is the prevention of charging and storage of static electricity among those items shown in Mechanism of Static Electrical Discharge Accidents. The preventive measures are shown below. It is recommended to exercise two or more measures simultaneously.

• Particularly when a large quantity of flammable solvent is held in a large vessel, be sure to observe the preventive measures 1, 2, and 3.

Preventive measure 1.

Use metallic (conductive) waste liquid vessel which is well grounded. This releases the charge of the waste liquid and vessel to ground.

- Be sure to ground the vessel properly. Disconnecting of grounding wire or poor grounding defeat the purpose of using a metallic vessel.
- There are some metallic cans which have no conductivity due to an oxidized coating or lacquer on their surface. Be sure to confirm the grounding of vessels by a tester before application.
- When a liquid with almost no conductivity (of 10-10s/m or less) is discharged into the vessel, it is necessary to mix it with another liquid with some conductivity. (The other liquid can be placed in the vessel in advance.)

Preventive measure 2.

Minimize the clearance of both inlet and outlet of vessel to prevent flame from entering the vessel.

• Use a cap with three holes for 18 liter and 4 liter cans.



Fig.: Anti-static electricity measures for vessel

Preventive measure 3.

Do not approach the vessel with charged objects including the human body. Charging prevention measures for human body

- a) Prevention of charging of shoes and clothes
- b) Grounding of human body
- c) Make working floor conductive

Suitable products to be used for those measures a), b), and c) are available on the market.

• When persons who use no charge prevention measures approach dangerous sections, they have to be grounded beforehand. (For example, they should contact grounded metal by hand.)

Preventive measure 4.

Use pipes with inner diameter of 2mm or more for waste liquid line for large flow rates.

• Inclusion of bubbles in the tube may increase the amount of charging by ten times. Check that there is no inclusion of air via tube joints.

Preventive measure 5.

When it is impossible to use a conductive vessel, use caution in the following points.

- a) Set the vessel so that the pipe outlet will be placed below the liquid level in the vessel. Or, dip a grounded metal (ex. pipe connected to the main body of device) in the liquid. Please note: This method is not effective for liquid with small conductivity (10-10s/m or less).
- b) Use a vessel of the smallest possible capacity to minimize the damage by fire if it should occur.
- c) Prevent the room from being dry. Humidity of 65% or more has charge prevention effects.

2 General Information

2.1 Outline

The basic design concept of the Dionex Fluorescence Detector **RF-2000** is to enhance productivity of analysis through its highest performance and stability.

This concept is realized by the highest sensitivity of the detector (higher than 300 signal to noise ratio for Raman spectrum of water), the highest wavelength accuracy (\pm 2nm), the highest wavelength reproducibility (\pm 0.2nm) and the GLP/GMP compliance support and improvement of operational safety.

The Dionex **RF-2000** supports operation via the data system CHROMELEON. Please refer to the sections "7.2 Control from External Equipment via RS-232C" and "7.2.3 Controlling the **RF-2000** via CHROMELEON" for details.

2.2 Features

• Greater sensitivity

The reengineered **RF-2000** optical system provides significantly increased sensitivity (signal-to-noise ratio greater than 300 for Raman spectrum of water). Furthermore, use of appropriate excitation and emission spectral scan conditions together with wavelength time programming allow high sensitivity detection of multiple components at trace level concentrations.

- Enhanced wavelength accuracy The **RF-2000** offers unsurpassed wavelength accuracy (± 2nm) and wavelength reproducibility to further enhance the reliability of wavelength selectivity and unitto-unit performance.
- Support for GLP/GMP compliance
 Daily checks and log-keeping can be automated*. The standard validation
 documents such as Certificate of Compliance and Hardware Validation SOP are
 provided to facilitate compliance with regulatory requirements.
 *With optional System Check software
- Enhanced safety features

Both heat and leak sensors are provided as a standard feature. In addition, the lamp may be turned ON/OFF via time program, minimizing unnecessary lamp consumption during continuous, unattended runs.

3 Component Location and Function

3.1 Front View

RF-2000 EX(nm) EM(nm) Data mill see a normal sem zero run 7 8 9 et ce 4 5 6 shift func 1 2 3 mark shift 0 · Enter	

No. Description

1 Control panel

Function Performs setting and display.

3.2 Top Side and Right Side



No.	Description	Function
1	Light source chamber cover	Remove this cover when replacing the lamp.
2	Cell cover	Remove this cover when replacing the cell.
3	Cell inlet tubing	Marked with blue cover tubing.
4	Cell outlet tubing	

3.3 Bottom Side



3.4 Rear View



No.	Description	Function
1	Horizontal direction adjusting	Adjusts horizontal alignment of the lamp
	screw	on replacement.
2/upper	Vertical direction adjusting screw	Adjusts vertical alignment of the lamp on
		replacement.
2/lower	Lamp fixing screw	Used to fix the lamp.
3	Fan	Removes internal heat.
4	External input and output	Terminals used for connection with
	terminals	external instruments.
5	Grounding terminal	Grounding is made through this terminal.
6	Fuse holder	Fuse is set in this holder.
7	Power supply connector	Connector for power cable
8	RS-232C connector	Connector for control by CHROMELEON

4 Installation

4.1 Basic Installation Requirements

- Warning: To take full advantage of the **RF-2000** performance capabilities and to ensure its operational stability over a long service life, verify that the selected installation site satisfies the following requirements.
- 1. Ventilation Ventilate the room where the high performance liquid chromatograph is located since the solvent used is flammable and/or toxic.
- 2. 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.
- 3. Sink Install a sink nearby for flushing eyes or skin which have been in contact with solvent.
- 4. Corrosive gas and Avoid installation in a place exposed to corrosive gases or dust.
- 5. Electromagnetic noise Avoid locations subject to intense magnetic or electromagnetic fields. Use an additional noise filter if power line noise interferes.
- 6. Space requirements This system is designed to be used on a table or stand, preferably a solid and flat surface with a depth of 70cm or more.
- 7. Others Select an installation site with the following parameters to maintain full performance of the system.
 - (1) Maintain room temperature within 4~35C, without extreme fluctuations.
 - (2) Avoid direct output of a heater or a cooler.
 - (3) Avoid exposure to direct sunlight.
 - (4) Avoid locations subject to strong vibrations or prolonged weak vibrations.
 - (5) Maintain relative humidity within 45~85%.
 (However, humidity should be below 70% when the room temperature is 30C or higher.)

4.2 Electrical Connections

Warning: When the power supply voltage is 220-230V~, operate the unit with the voltage setting of the power supply connector at the rear of this unit at 230Vac. When the power supply voltage is at 240V~, set the power supply connector at 240Vac. For changing the setting of power supply connector, refer to "8.4 Changing the set power supply voltage".

If the power supply is not stable or the capacity insufficient, satisfactory performance is not possible. Verify for the total power supply for the system before preparing power supply.

• Verify that the power switch of the main unit is turned OFF.

4.2.1 Connection to outlet

- (1) Connect the female connector of the power supply cord supplied with the unit to the power supply cord connector at the rear of the unit, and plug the male connector into a power supply outlet.
- Supplied power supply cord is of three-conductor (3P) type.
 When connecting to two-conductor (2P) type power supply outlet, use the provided power supply adaptor.



4.2.2 Grounding

- (1) When three-conductor (3P) type power supply outlet is used, the unit is grounded by the power supply cord.
- When two-conductor (2P) type power supply outlet is used, the unit is not grounded. In this case, carry out grounding from the grounding terminal on the unit rear panel.
 To prevent electric shock and to secure safe operation of the system, always

ground the unit.

4.3 Fluidic Connections

4.3.1 Connecting the Detector and the Column

Typical connection between this equipment and the column is described in this section.

(1) Cut the piping tube (SUS Tube 1.6 x 0.3, standard accessory) at the length necessary for connecting the column outlet and the cell inlet pipe (pipe with blue tape) of this equipment. Cut the tube perpendicular. If the tube is cut diagonally dead volume is

Cut the tube perpendicular. If the tube is cut diagonally, dead volume is generated to deteriorate separation.



- (2) Connect the piping between the column and the cell inlet pipe with the supplied male PEEK nut and the male to male union as shown in the figure below. Tighten the male PEEK nut by the hand firmly. It is not necessary to use a tool for tightening.
- **I** Please note: Insert the piping tube into the column joint and the male to male union securely until it comes to the end and then tighten the male PEEK nut. Similarly, insert the cell inlet pipe until it comes to the end and tighten the male PEEK nut so that dead volume is avoided.



4.3.2 Detector Outlet Side Piping

- (1) Prepare a waste liquid bottle.
- (2) Connect the male to male union and the Tefzel tube (standard accessories of this product) to the cell outlet pipe and put the other end in the waste liquid bottle, as shown in the figure.
- Please note: Pressure tolerance of the cell is 1960 kPa {20kgf/cm²} (Value in {} is for reference). Pressure exceeding this level may cause leak of liquid or damage of the cell. To prevent such troubles, pay attention to the following points:

- Do not apply excessive back pressure to the outlet side of the cell.
- In order to prevent clogging in the flow line, thoroughly clean the flow line before stop delivery of solvent when the analysis is finished. Especially, when the buffer solution is used, salts may be crystallized inside flowline. Be sure to clean the flow line.
- When delivery of the solvent is stopped temporarily, put the tip of the flow line outlet in the waste liquid in the bottle to prevent clogging of the flow line. This is to keep inside the flowline being wet.

▲ Important: When a mobile phase with high insulation, such as hexane, is used, it is dangerous since static electricity is changed at the waste liquid bottle. Use a metal liquid bottle for waste and ground the bottle.

(Refer to section "1.6 Precautions on Static Electricity")

4.4 Connecting the Recorder and the Integrator

4.4.1 Connecting with a Recorder

Connect the RECORDER terminal of this instrument and the recorder using the provided signal cable.

When using a stranded wire, twist the end tightly, or tin it with solder. Using a small screwdriver or other tool, depress the rectangular button adjacent to the appropriate terminal hole. Insert the wire and release the button to clamp the wire in position, as shown in the figure below.



⚠ Important:

ant: Be sure to turn the power of the main unit OFF before connecting the signal cable.

4.4.2 Connecting with an Integrator

Connect the integrator to the INTEGRATOR terminals in the external input/output terminals at the rear of the **RF-2000**. Connect the cable supplied with the integrator. Connect each terminal of the signal cable to the terminal block supplied with the integrator.

4.5 Connecting a Drain Tube for Solvent Leakage

Each component in the **RF-2000** is designed so that liquid leakage in the equipment is discharged from the liquid leak tube connection port at the right side or the lower front side of the equipment. Connect liquid leak tube if necessary.



4.5.1 Connecting a Liquid Leak Tube

Cut the drain tube (standard accessory) for liquid leakage at an appropriate length and connect the waste liquid tube to the drain tube and put the other end in the waste liquid bottle.

⚠ Important: Place the waste liquid bottle lower than the device at the bottom. If the flow line of the liquid leak discharging tube is placed above the bottom face of the **RF-2000**, the waste liquid cannot be drained.

4.6 Installing the Xenon Lamp

Be certain that the AC power cable plug is disconnected from this equipment for prevention of electric shock when installing the xenon lamp.

Warning:

- Be sure to wear protective gears, such as protective mask, heavy and long sleeve shirt and gloves, when handling the xenon lamp.
 - Since high pressure gas in changed in the xenon lamp, if the lamp is strongly shocked or the glass surface is scratched, the lamp may explode and piece of glass may scatter.

- When the light source chamber cover shall be opened, be sure to turn OFF the power switch and disconnect the power cable from power supply.
 - It is very dangerous to touch the lamp by any mistake because high voltage, approx. 30kV, is applied to the + pole of the xenon lamp when the lamp is lit on and 20V is applied while the lamp is lit.
 - The xenon lamp is still charged just after the power switch is turned OFF. Leave more than 5 minutes after the power switch is turned OFF before open the light source chamber cover to avoid electric shocks.
 - It is very high temperature right after the Xenon lamp is turned OFF. Wait until it is cooled down sufficiently for replacement. The time required for cooling down the lamp is an hour when it is left with its power supply turned OFF, while it is 30 minutes when the lamp is turned OFF with "LAMP" of the AUX FUNC set at "0".



4.6.1 Installation procedure

- (1) Remove the two mounting screws of the light source chamber cover with a screwdriver and remove the light source chamber cover.
- (2) Loosen the sliding plate screws and slide the sliding plate so that the inside of the light source chamber case can be seen.
- (3) Insert the supplied screwdriver in the hole for screwdriver for lamp fixing along the guide and turn in counterclockwise to loosen the lamp fixing screw.
- (4) Take the xenon lamp out of the case and remove the knurled screws at the plus pole and the minus pole of the lamp.
- (5) Turn the plus pole of Xenon lamp upward, and insert the xenon lamp into the xenon lamp holder with the gas filling protrusion (tip off) of the xenon lamp faced to the opposite side of the beam-condensing mirror.

- ▲ Important: When doing this, do not touch the valve surface of the lamp. If touched, wipe off the surface with a gauze wet with ethanol (provided with the lamp), etc. If the lamp is lit with fingerprints on it, the fingerprints are burned and causes explosion of the lamp valve.
- (6) Tighten the lamp mounting screw with the screwdriver through the hole for screwdriver for lamp fixing to fix the lamp.





Lamp mounting direction

(7) Close the sliding plate and fix it with the sliding plate screws.



- (8) Set the terminal of cable connected to the plus pole in the screw at the plus pole of the lamp and fix it with a knurled screw securely.
- (9) Remount the light source chamber cover.

Warning: When tightening the terminal at the plus pole with a knurled screw, be sure to allow the cable to the plus pole be bent.

If the plus pole is stretched by the cable, there occurs a danger that the xenon lamp may be destroyed while being lit. Besides, when tightening the knurled screw, be sure to tighten by fingers. It is dangerous to use any tool such as wrench.

4.7 Adjusting the Xenon Lamp Alignment

4.7.1 Adjusting the light source position (coarse adjustment)

Adjust the light source position for maximum equipment performance. Be sure to adjust the light source position during installation or replacement of the lamp. Adjustment consists of coarse and fine adjustment.

Please note: The xenon lamp may not be lit on if the lamp is heated just after lit off after being lit on for a while. In such case, wait for about 10 minutes until the lamp is cooled and turn ON the power switch again.

- (1) Turning power ON, and make sure lamp LED on the front panel is lit.
- (2) Set excitation wavelength to 540nm.
- (3) Remove the flow cell and place a piece of white paper where the flow cell was installed and where green light is observed on the paper.
- (4) Adjust the light source horizontal direction adjusting screw for maximum brightness of the green light on the paper.
- (5) Adjust the light source vertical direction adjusting screw in a same way as above (4).
- (6) Remove the paper.



4.7.2 Adjusting the light source position (fine adjustment)

Important: If a pen recorder is connected, proceed as described below. If not, please contact the Dionex Service for the fine adjustment of the light source position.

- (1) Connect a recorder to the 10mV RECORDER terminal of the **RF-2000** (see section 7.1), and turn the power ON.
- (2) Remove the flow cell from the **RF-2000**. Pour distilled water into the flow cell using the provided syringe. Make sure that no bubbles are present in the flow cell, and install the flow cell in the sample compartment (see "8.1 Maintenance of Flow Cell").
- (3) Set recorder parameter as follows:
 - 1) Range : 10mV/Full Scale
 - 2) Chart Speed : 10mm/min
- (4) Adjust the zero position by pressing the **zero** key of the **RF-2000**.
- (5) Adjust the zero position of the recorder.
- (6) Press **func** key of the **RF-2000** until the fluorescent character displays

LAMP ADJUST

(7) Press ENTER.

The parameters are automatically set as follows (Refer to "5 Operation"):

· · ·		/	
1)	EX	:	350nm
2)	EM	:	450nm
3)	GAIN	:	2 (x 4)
4)	RANGE	:	4 (x 1/8)
5)	RATIO	:	0 (OFF)
6)	sens	:	2 (MED)
7)	rsp	:	1 (0.1s)
8)	Z WAVE	:	1 (OFF)

The above parameters are set and the auto-zero function is activated automatically while the following message appears on the screen.



Then the emission wavelength is changed and set at 397nm automatically and the following message appears.

|--|

Please note: Be sure to obey the order of adjustment of the horizontal direction adjusting screw and the vertical direction adjustment screw as described below. If the order is not correct, the light source position deviates greatly from the slit of the excitation monochromator and re-adjustment cannot be possible. In this case, carry out the adjustment from "4.7.1 Adjusting the light source position (Coarse adjustment)" again.

(8) Adjust the horizontal direction adjusting screw on the rear panel, till the pen position comes to the maximum point. (till the value in the display indicates its maximum.)



Rear View

- (9) Adjust the vertical direction adjusting screw on the rear panel till the pen position comes to the maximum point. (till the value in the display indicates its maximum.)
- (10) Repeat the steps (8) and (9) so that the maximum value at the horizontal direction adjustment and that of the vertical direction adjustment become the same. In many cases, the value indicated on display of **RF-2000** is more than 30.
- (11) Stop recording by the recorder. To stop, press **shift down**, **plot** and **Enter**.
- (12) Pressing the **CE** key resets each parameter at the state before "LAMP ADJUST" is performed.

5 Operation

5.1 Precautions for Operation

- Important:
 1. Be sure the sample compartment cover is closed during measurement. Measurement cannot be made correctly while the cover is opened.
 - 2. Precautions to prevent clogging of the flow cell. Dusty or clogged flow cells are the most frequent causes of trouble in any detector. After analyzing a high-concentration sample, thoroughly flush it from the flow cell, using a large amount of mobile phase. Buffer solution crystallizes upon drying, and can clog the flow cell and tubing. Never leave buffer solution in the unit as mobile phase. Always flush the flow lines prior to shutdown of the instrument.

5.2 Fundamentals of Operation

5.2.1 Turning power ON

- (1) Push the power switch on the front panel to turn the power ON. Push this switch again to turn the power OFF.
- (2) When the power is turned ON, it operates as follows:
 - 1) Turning power ON
 - 2) All of the dots in the display unit and all the indicator lamps light.
 - 3) Control program version No. is displayed.
 - 4) The instrument seeks the home position of the gratings.
 - 5) When initialization and calibration are done normally, the following message is displayed and the instrument is ready for operation.

	EX(nm)	EM(nm)	Data
;	350	400	0.00

- 6) Measuring parameters and time program when the power was turned off previously are saved.
- Please note: If an error message is displayed after turning the power ON, take an appropriate measure as referred in section "9.2 Error Messages".

5.2.2 Display unit

The display unit consists of display screen and indicator lamps. Their descriptions and functions are as follows.



No. Display or description Function

1)	EX (nm)	Displays excitation wavelength (unit: nm)
2)	EM (nm)	Displays emission wavelength (unit: nm)
3)	Data	Displays emission intensity (no unit)
4)	shift	Shift key indicator lamp
5)	prog run	Time program operation indicator lamp;
		Lights when time program is being executed.
6)	remote	Remote mode indicator lamp
7)	lamp	Light source lamp;
		Lights when the Xe light source is lighting.
8)	scan	Scan lamp;
		Lights during wavelength scanning.

5.2.3 Keyboard

19 keys on the front are used for operation and setting. They are classified into the following three types:

1) STD-func key

After pressing this key, the specified operation is immediately executed (**zero** key, etc.).

 shift-func keys Pressing this key after pressing the shift key, the specified operation is executed (9^{scan} key, etc.).

3) edit keys

Used for inputting parameters and editing a time program (ten keys, etc.).



(1) STD- func	keys	
zero	Auto zero key	Pressing this key performs zero adjustment
∆ zero shift ∇	Zero shift key	Zero position on the recorder or integrator can be moved by pressing this key. It is moved upward by pressing the Δ side and downward by pressing the ∇ side.
mark	Mark key	Mark is added to the data being recorded in the recorder and integrator by pressing this key. Mark size: approx. 4mV (INT), approx. 4mV (REC 10mV), approx. 0.4mV (REC 1mV)
run	Run key	It is a switch to start and stop a time program.
(2) shift-func l	keys	
shift + edit	Edit key	Moves to time program edit mode.
shift + range	Range key	Moves to range setting mode.
shift + sens	Sensitivity key	Moves to sensitivity setting mode.
shift + scan	Scan key	Wavelength scanning is performed. The
ohift ± ron	Doononoo kov	scan lamp is lit while scanning.
shint + rsp	Response key	Noves to response setting mode.
(3) edit keys		
0 - 9	Numerical keys	Numerical values are input with these keys.
Enter	Enter key	Set input value with this key.
CE	Clear key	Returns the display screen to the initial
		linguit value is cleared by pressing this key
		when entering a numerical value. Display
		and alarm are cleared by pressing this key
		when an error is displayed. But, errors such
		as equipment failure cannot be cleared with
abiff I dal	Delete kov	this key.
snift + dei	Delete key	the time program
func	Function key	Advances to the next setting item in the
	i anotion noy	display screen.
		AUX. FUNC setting screen is forwarded.
shift + back	Back key	Returns to the previous setting item.
		AUX. FUNC setting screen is scrolled
		backward.
		Returns to the previous setting item by
		program.

5.2.4 Setting excitation wavelength and emission wavelength

EX(nm)	EM(nm)	Data
<<350>> 1000.00	400	

The value for excitation wavelength is blinking in the initial state to indicate that the excitation wavelength can be changed. Pressing the **func** key, emission wavelength becomes possible to be changed and pressing **shift + back** keys in this state, excitation wavelength becomes possible to be changed again. (Basically, any blinking value shows being possible to be changed.) Enter a desired wavelength and press **Enter** to change the measuring wavelength.

[Example 1]

To change the excitation wavelength from 350nm to 360nm:

(1) Ensure that the value for EX (nm) is blinking.

EX(nm)	EM(nm)	Data
<<350>> 1000.00	400	

(2) Press the keys **3 6 0** and **Enter** to enter 360nm.

EX(nm)	EM(nm)	Data
<<360>> 1000.00	400	

[Example 2]

To change the emission wavelength from 400nm to 450nm:

(1) Ensure that the value for EM (nm) is blinking by pressing the **func** key.

EX(nm)	EM(nm)	Data
360 1000.00	<<400>>	

(2) Press the keys **4 5 0** and then **Enter** to enter 450nm.

EX(nm)	EM(nm)	Data
360 1000.00	<<450>>	

- 1. When the display is not in the initial state, press the **CE** key to return to the initial state.
- 2. Any value outside the setting range is not accepted. Setting range is 0nm and from 200nm to 900nm and measuring range is 0nm and from 200nm to 650nm.

5.2.5 Setting sensitivity

Setting procedure for sensitivity is as follows:

(1) Press **shift + sens**. The following is displayed.

SENS		<<2>>
EX(nm)	EM(nm)	Data

Sensitivity can be changed.

(2) Enter the desired set value with numerical key and press Enter.

Set value	Sensitivity	
1	HIGH	
2	MED	
3	LOW	

Default value at the factory is set at 2 (MED).

Sensitivity MED is 32 times and HIGH is 1024 times as high as LOW sensitivity. The measurement range can be set as follows by combining with GAIN (refer to the section "5.5 Additional Functions").

SENS	GAIN	Sensitivity magnifications	
3 (LOW)	1 (x 1)	approx. x 1	
3 (LOW)	2 (x 4)	approx. x 4	
3 (LOW)	3 (x 16)	approx. x 16	
2 (MED)	1 (x 1)	approx. x 32	
2 (MED)	2 (x 4)	approx. x 128	
2 (MED)	3 (x 16)	approx. x 512	
1 (HIGH)	1 (x 1)	approx. x 1024	
1 (HIGH)	2 (x 4)	approx. x 4096	
1 (HIGH)	3 (x 16)	approx. x 16384	

(3) To return the display to the initial state, press **CE**.

Please note: When the values of sensitivity (SENS) and/or Gain (GAIN) are changed, it is necessary to create a new calibration curve.

5.2.6 Setting recorder output range

Procedure to set recorder output range is as follows:

- (1) Press **shift** + **range**.
 - The following is displayed.

Range can be changed.

(2) Enter a desired set value with numerical key and press Enter.

Set value	Range
0	Output : 0V
1	x 1
2	x 1/2
3	x 1/4
4	x 1/8
5	x 1/16
6	x 1/32
7	x 1/64
8	x 1/128
9	x 1/256

When the emission intensity is maximum (about 1000) and the range is x1, signal of about 500mV is output to the 10mVFS terminal and signal of about 50mV is output to the 1mVFS terminal.

For selecting a range, determine the maximum value of emission intensity to be recorded in the recorder and apply the value in the following equation to determine the range.

Range \leq 20 \div (Maximum intensity of emission) (Common to both 10mVFS and 1mVFS)

[Example] To record the emission intensity in the range of 0 \sim 150 in the recorder:

Range $\leq 20 \div 150 = 1/7.5$ In this case, select range x 1/8.

- (3) To return the display to the initial state, press **CE**.
- **Please note:** When the emission intensity is at the maximum value (1000), signal of 1V is output to INTEG.1V terminal regardless of setting of the range.

5.2.7 Zero position adjustment for recorder

Adjust the zero position of recorder according to the following procedure before starting measurement.

- (1) Set the measuring range to a required value (refer to the previous item "Setting recorder output range").
- (2) Press **zero** to set the emission intensity to near 0.
- (3) Move the baseline to a desired position using the pen position adjusting knob of the recorder or pressing the Δ zero shift ∇ key and start measurement. The baseline returns to the position set in (3) thereafter by pressing zero.

Please note: Range variable by pressing the Δ **zero shift** ∇ key is as follows: Approx + 100 ~ approx. - 20 (scale).

5.2.8 Setting response

In this equipment, a digital filter is used to improve S/N ratio.

Responsibility becomes better by setting the filter response fast but noise reduction effect becomes less.

On the contrary, responsibility becomes worse by setting the response slow, but noise reduction effect becomes large.

4 steps from 1 to 4 are available as the response for the digital filter by the setting of parameter "RESPONSE". Responsibilities at each "RESPONSE" value compared to conventional analog filter and equivalent time constants for analog filter are as shown in the table below:

RESPONSE value	Equivalent time constant of analog CR filter	Minimum peak width at half height (Note)
1	0.1s	0.2s or more
2	0.5s	1.1s or more
3	1.5s	3.6s or more
4	3.0s	6.6s or more

Setting procedure for RESPONSE:

Press shift + rsp. The following is displayed and RESPONSE can be changed.

EX(nm)	EM(nm)	Data
RESPONSE		<<3>>

- 2) Enter the desired value with numerical key and press **Enter**.
- 3) Press **CE** to return to the initial state of display.
- Please note: If response is set slower, responsibility becomes poorer and peak height becomes lower, and peaks with less width of half-height show more deterioration of the peak height. Here, the figures of minimum peak width of half-height indicate for reference that the peak height becomes 10% lower at each response setting value. Even when responsibility becomes poorer and the peak is broadened, the peak area is not changed.

5.3 Operating Procedure in Spectrum Scanning Mode

To seek the optimum excitation wavelength and emission wavelength, this unit is equipped with a spectrum scan function which can store two sample spectra and one background spectrum.

This unit can output the spectrum of the excitation light.

- To measure the excitation spectrum or the emission spectrum of the sample, set the "RATIO" parameter of the "Additional functions" to "1".
- To measure the spectrum of the excitation light, set the "RATIO" parameter to "2" and the "SPCTYPE" parameter to "1" (refer to "5.5 Additional functions")

5.3.1 Flow line in spectrum scanning

Liquid in the cell should not be moved while scanning. It is necessary to switch the flow line by the high-pressure 6-way valve as shown in the figure below: The 6-way valve is usually set for the flow line indicated by the solid lines and is switched for the flow line indicated by the broken lines before starting scanning.



As an easier method to stop the flow, the pump may be stopped at the rising of peak. However, it is necessary to take into consideration that the flow cannot stop immediately when the pump is stopped.

Please note:

Do not open the drain valve to stop the flow. The life of the column may be shortened by pressure shock.

5.3.2 Setting parameters

Before starting scanning, set the scanning wavelength and scanning speed by AUX. FUNC.

1) 2) 3) 4) 5) 6)	SCAN FILE SPC TYPE EX SCAN BGN EX SCAN END EM SCAN BGN EM SCAN END	Scan data file Scan type (excitation, emission) Excitation scan start wavelength Excitation scan end wavelength Emission scan start wavelength
6)	EM SCAN END	Emission scan end wavelength
7)	SCAN SPD	Scanning speed

SCAN FILE No. 0 is dedicated for background spectrum and FILES No. 1 and 2 are for samples spectra.

5.3.3 Executing scanning

Press **shift** + **scan** to start scanning. The scan lamp is lit during scanning. To stop scanning, press **shift** + **scan** again.

Please note: If scanning is executed when previous data is stored in the SCAN FILE, the previous data is erased and a new data is stored.

5.3.4 Output of spectrum data

Spectrum data obtained can be output at the recorder or integrator. Before outputting, select output range, file No. and plot speed. Set the output range. Set the file No. and plot speed in the parameters SCAN FILE and PLOT SPD in AUX. FUNC.

Set value for PLOT SPD	Plot speed
1	1nm/s
2	5nm/s
3	10nm/s

To plot, select and display SPC PLOT in AUX. FUNC and press **Enter**. To stop plotting, press **Enter** again. Output example:



A mark is added at the start and end.
Please note: If the output changes abruptly at the beginning or the end of the spectrum plot, the mark may not be recorded.

Mark : approx. 4mV (INT) approx. 4mV (REC 10mV) approx. 0.4mV (REC 1mV)

Output time T is as follows: T = (SCAN END - SCAN BGN) /Plot speed (1, 5, or 10nm/s)

For example, when the range from 200nm to 350nm is to be output at the plot speed of 5nm/s (PLOT SPD = 2):

T = (350 - 200) nm/5nm/s = 30s

Please note: Data in any file which is not scanned cannot be output. If data does not exist in the specified file, "DATA NOT EXIST" message is displayed. Similarly, plotting is not possible when data does not exist in the background file (SCAN FILE = 0).

5.4 Creating and Executing a Time Program

5.4.1 Command list

Commands which can be used for a time program are listed below:

Command	Description	Setting range	Remarks
ΕΧλ	Excitation wavelength	0, 200 ~ 900	
ΕΜλ	Emission wavelength	0, 200 ~ 900	
ZERO	Execution of zero adjustment	None	
MARK	Marking on output for recorder	None	
RANG	Output range for recorder	0~9	Recorder is short- circuited when it is set to 0.
RESP	Response	1 ~ 4	
SENS	Sensitivity	1 HIGH	
		2 MED	
		3 LOW	
GAIN	Gain	1 x 1	
		2 x 4	
		3 x 16	
SCAN	Execution of wavelength	0~2	Specifies file no. where
	scanning		scanned data is to be stored.
EVNT	EVENT output of ON/OFF	0 OFF	
		1 ON	
LOOP	Repeats time program from	0 ~ 255	
	the beginning.	0 means 256	
		times.	
STOP	Stops time program.	None	
LAMP	Turn ON and OFF the	0 OFF	
	lamp.	1 ON	

Example of time program is shown below:

E	EX(nm)	EM(nm)	Data
().01	ZERO	

Emission intensity is set at 0 in 0.01 minute after starting the time program.

EX(nm)	EM(nm)	Data
5.00	ΕΧ λ	300

In 5minutes, EX (Excitation wavelength) is set at 300nm.

5.00	ΕΜ λ	400
EX(nm)	EM(nm)	Data

In 5 minutes, EM (Emission wavelength) is set at 400nm.

EX(nm) EM(nm)	Data
5.10	SENS	3

In 5.1 minutes, sensitivity is set at 3 (Low).

	•		
	EX(nm)	EM(nm)	Data
	5.10	GAIN	3
Right after that, gain is	set at 3 (x 16)		
	EX(nm)	EM(nm)	Data
	5.10	RESP	2
Right after that, response	se is set at 2 (0.5s).	
	EX(nm)	EM(nm)	Data
	5.10	RANG	5
Right after that, range is	s set at 5 (x 16	δ).	
	EX(nm)	EM(nm)	Data
	10.00	MARK	

In 10 minutes, marking on the recorder is performed.

EX(nm)	EM(nm)	Data
20.00	SCAN	0

In 20 minutes, wavelength scanning is executed and data is stored in file 0. (Type, wavelength range and speed of the scan should be set in AUX. FUNC in advance.)

		2
 999.99	LOOP	2

In 999. 99 minutes, the timer is reset to 0 and the time program is repeated from the beginning. The time program is executed twice and stopped.

- Since the above time program is just an example for explanation and may not be adequate for actual measurement.
- The screen display does not change as shown in the above example synchronously with the time program. Check the program in the edit mode.

5.4.2 Explanation of display screen

It is necessary to enter the edit mode to edit a time program.

(1) Press **shift** + **edit** and the following is displayed.

EX(nm)	EM(nm)	Data
10 I FFT	USED	22

- 10: Number of steps already set.
- 22: Number of available steps left.

The above example shows that the time program is set for 10 steps and there are 22 steps available.

(2) Press **Enter**, and the following is displayed.

EX(nm)	EM(nm)	Data
TIME VALUE	FUNC	

Time: Elapsed time from time program start (minute) Func: Command name Value: Set value

(3) If a time program is made, the following is displayed by pressing **Enter** again. The contents in the display are the same as those in (2).

(Example)

<<1.00>>	ΕΧ λ	350
EX(nm)	EM(nm)	Data

The above display indicates that the excitation wavelength becomes 350nm in one minute from the time program start.

(4) When pressing **Enter** repeatedly, the time program is displayed in forward steps.

(5) Pressing **shift** + **back**, the time program is displayed in backward steps.

5.4.3 Flow of setting

Flow in setting a time program is shown below:



Enter a time for the next command.

5.4.4 Scanning by program

Example setting to execute scanning in 5 minutes and store data in file No. 2 using a program is shown below:

Press CE to return to the initial screen. (1)



<<5.00>>	SCAN	2
EX(nm)	EM(nm)	Data

- Enter the second and later steps if necessary by repeating (4) to (6). To finish (7) setting the program, press **CE** twice.
- **I** Please note: • When setting multiple steps, it is not necessary to set the commands in order of time. They are automatically rearranged.
 - Set the STOP command at the end of a program unless the program is to be executed repeatedly.
 - In selecting a function, the previous function is displayed by • pressing **shift** + **back**.

5.4.5 Deleting a step

Display the step desired to be deleted and press **shift** + **del**. (1) Display the step desired to be deleted.



(2) Press **shift** + **del**.



The step displayed in the program is deleted and the next step is displayed if any. When any next step is not set, the following is displayed.

5.4.6 Start and stop

After setting a time program, start and stop the time program according to the following procedure:

- (1) To start a time program:
 - Press **run**.

The LED for **prog run** is lit.

(2) To stop the time program:

When the program is stopped, it returns to the state just before the time program is started.

There are two ways to stop a time program: forced stop of the program being executed, and stop of a time program by STOP command in the program.

Procedure of forced stop of the time program is as follows: Press **run**.

The LED for **prog run** is lit. Now the time program stops.

5.4.7 LOOP command

A program can be repeated for the number of times set by LOOP command. The LOOP is a command to reset TIME at 0.00 and start the program again.

	TIME	FUNC	VALUE
1)	15.00	ΕΧλ	300
2)	20.00	ΕΧλ	350
	30.00	LOOP	3

In the above setting, steps 1) and 2) are repeated 3 times in a cycle of 30 minutes.

Please note: • Steps set after the LOOP command are invalid.

• Up to 255 can be set as the VALUE for LOOP command. When 0 is set for the value, the program is repeated 256 times.

5.5 Additional Functions

AUX. FUNC (AUXILIARY FUNCTION) is equipped in this instrument.

5.5.1 AUX. FUNC List

Туре	Command	Function
4	TIME	Displays the elapsed time while a time program is
		executed (when MONIT-TIME is set)
4	SMPL EN	Displays output from emission measurement sensor
4	REF EN	Displays excitation light beam monitor value
1	GAIN	Sets gain. $(1 = x1, 2 = x4, 3 = x16)$
1	SCAN FILE	Sets file No. Where scan data is to be stored
		(0=Background, 1 and 2=Sample)
1	SPC TYPE	Sets scan type. (1=EX, 2=EM)
1	EX SCAN BGN	Sets excitation scan start wavelength. (200 ~ 900nm)
1	EX SCAN END	Sets excitation scan end wavelength. (200 ~ 900nm)
1	EM SCAN BGN	Sets emission scan start wavelength. (200 ~ 900nm)
1	EM SCAN END	Sets emission scan end wavelength. (200 ~ 900nm)
1	SCAN SPD	Sets scanning speed. (1=SUPER, 2=FAST,
		3=MED,4=SLOW)
1	PLOT SPD	Sets output speed of spectrum data to recorder.
		(1=1nm/sec, 2=5nm/sec, 3=10nm/sec)
3	SPC PLOT	Outputs spectrum data to recorder.
1	EVENT	Sets EVENT OUT terminal status. (0=OFF, 1=ON)
1	EXT-S	Used when EVENT OUT terminal is used for starting
		external equipment, etc.
		(0=EVENT, 1=Time program, 2=Unreleasable error,
		3=Wavelength scan)
1	MONIT-TIME	Sets when elapsed time is desired to be displayed in
		executing a time program. (0=not to display, 1=to display)
1	LAMP	Sets ON/OFF of Xe lamp. (0=OFF, 1=ON)
1	RATIO	Decides operation of light source compensation by
		division.
		(0= without division, 1 = with division, 2 = Excitation light
		energy)
4	XE TIME	Displays accumulated operating time of xenon lamp.
1	Z WAVE	Sets the output holding function when setting of
		wavelength, sensitivity and gain is changed. (0=set, 1=not
		set)
3	RS232C	Sets communication parameters of RS-232C and activates
		remote control mode. Refer to "7.2 Control from external
		system via RS-232C".
3	CLOSE KEY	Refuses any key input.
3	LAMP ADJUST	Enters lamp position adjusting mode.
3	S/N CHECK	Used for Performance Check.

"Type" in the above table indicates a kind of operating procedure.

Type 1: Enter a desired value with numerical keys and press the Enter key.

Type 3: Press the **Enter** key to execute the function.

Type 4: The status is displayed without any key operation.

5.5.2 Setting procedure for AUX. FUNC

TIME

(Display of elapsed time of time program)

EX(nm)	EM(nm)	Data
TIME 100.00		

This function displays the elapsed time from the program start while the time program is executed.

However, this is displayed only when MONIT-TIME is set to 1 in AUX. FUNC.

SMPL EN

(Output from emission measurement sensor)

SMPL EN		XXX.X
EX(nm)	EM(nm)	Data

This function displays output from emission measurement sensor (Photo multiplier tube).

REF EN

(Excitation light beam monitor value)

REF EN		XXX.X
EX(nm)	EM(nm)	Data

This function displays output from excitation light beam monitor (Photodiode).

GAIN

(Setting of gain)



This function sets gain for signal from the detector. Enter a desired value with numeric key and press **Enter**.

Default set value at the factory is "2" (Gain = x 4). For general use, "2" (Gain = x 4) can be applied.

Set value	Gain
1	x1
2	x4
3	x16

Magnification can be set as follows by combining with SENS (refer to section "5.2.5 Setting Sensitivity").

SENS	Sensitivity magnifications	
3 (LOW)	approx. x 1	
3 (LOW)	approx. x 4	
3 (LOW)	approx. x 16	
2 (MED)	approx. x 32	
2 (MED)	approx. x 128	
2 (MED)	approx. x 512	
1 (HIGH)	approx. x 1024	
1 (HIGH)	approx. x 4096	
1 (HIGH)	approx. x 16384	
i Please note:	When values of sensitivity it is necessary to create a r	SENS) and/or Gain (GAIN) are changed, new calibration curve.

SCAN FILE

(Setting of scan file)

EX(nm)	EM(nm)	Data
SCAN F	ILE	

This function sets No. of file where scan data is to be stored. Enter the desired file No. with numerical key and press **Enter**.

Set value	Туре
0	Background
1, 2	Sample

SPC TYPE

(Setting of scan type)

EX(nm)	EM(nm)	Data
SPC TY	'PE	

This function sets spectrum scan type. Input a set value with numeric key and press **Enter**.

Set value	Туре
1	Excitation scanning
2	Emission scanning

EX SCAN BGN

(Setting of excitation scan start wavelength)

_

 EX(nm)
 EM(nm)
 Data

 EX SCAN BGN

This function sets start wavelength for excitation scanning.

Enter the desired start wavelength with a numeric key and press Enter (200 ~ 900).

EX SCAN END

(Setting of excitation scan end wavelength)

EX(nm) EM(nm)

Data



This function sets end wavelength for excitation scanning. Enter the desired end wavelength with a numeric key and press **Enter** (200 ~ 900).

Please note: EX SCAN BGN should be smaller than EX SCAN END.

EM SCAN BGN

(Setting of emission scan start wavelength)

EX(nm) EM(nm) Data



This function sets start wavelength for emission scanning. Enter desired start wavelength with numeric key and press **Enter** (200 ~ 900).

EM SCAN END

(Setting of emission scan end wavelength) This function sets end wavelength for emission scanning.

EX(nm) EM(nm) Data

EM SCAN END	

Enter desired end wavelength with numeric key and press Enter (200 ~ 900).

Please note: EM SCAN BGN should be smaller than EM SCAN END.

SCAN SPD

(Setting of scanning speed)

EX(nm) EM(nm)

This function sets scanning speed.

Enter the desired set value with numeric key and press Enter.

Set value	Scanning speed
1	SUPER (approx. 3000nm/min)
2	FAST (approx. 600nm/min)
3	MEDIUM (approx. 120nm/min)
4	SLOW (approx. 24nm/min)

Data

i Please note:

te: When scanning speed is set to super, spectrum data is stored at an interval of 4nm. When scanning speed is set to the other value, spectrum data is stored at an interval of 1nm.

PLOT SPD

(Setting of plot speed)

EX(nm) EM(nm) Data

This function sets output speed of spectrum data to the recorder or integrator. Enter the desired set value with numeric key and press **Enter**.

Set value	Output speed
1	1 nm/s
2	5 nm/s
3	10 nm/s

Please note: When recording to a recorder or an integrator is performed simultaneously with wavelength scanning, data is kept being output to the recorder or integrator regardless of this setting.

SPC PLOT

(Output of spectrum)

EX(nm)	EM(nm)	Data
SPC PL	ОТ	

This function outputs spectrum data in the file set in SCAN FILE to a recorder or an integrator.

Press Enter to start outputting to the recorder.

EVENT

(Setting of EVENT OUT terminal)

EX(nm) EM(nm) Data

This function sets ON (close) / OFF (open) of EVENT OUT terminal (relay contact) located in the rear side of this equipment.

Enter the desired set value with numeric key and press Enter.

Set value	EVENT OUT terminal
0	OFF (open)
1	ON (close)

EXT-S

(Function setting of EVENT OUT terminal)

EX(nm) EM(nm) Data

This function specifies control mode for external instruments by EVENT OUT terminal.

Enter the desired set value with numeric key and press **Enter**.

Set value	Output speed
0	EVENT output is controlled by EVENT set value.
1	Relay is turned ON for about one second at a time program start.
2	Relay is turned ON at an occurrence of error which cannot be reset.
3	Relay is turned ON during wavelength scanning.

MONIT-TIME

(Setting of display of elapsed time of time program)





This function displays the elapsed time from the time program start. It is displayed on the TIME screen in AUX. FUNC.

Enter the desired set value with numeric key and press Enter.



LAMP

(Setting of xenon lamp ON/OFF)

LAMP		
EX(nm)	EM(nm)	Data

This function controls ON/OFF of xenon lamp.

Set value	Xenon lamp
0	OFF
1	ON

Please note: When the xenon lamp is lit on, this instrument performs initialization. After completion of initialization, all set values are reset to the state before lit on lamp.

RATIO

(Setting of division correction to output signal)

EX(nm)	EM(nm)	Data
RATIO		

This instrument corrects the change of the fluorescence light energy caused by the change of excitation light energy by dividing the monitored excitation light energy. This function sets this correction.

Generally, this division correction is performed (Set value: 1).

Set value	Functions
0	Outputs the fluorescence light energy.
1	Executes the division correction.
	(Outputs the ratio of "fluorescence/ excitation")
2	Outputs the excitation light energy.

Please note: When the xenon lamp is OFF (see item of LAMP), the value is automatically set to "0" (outputs fluorescence light energy). When the value is set to "1" (executes division correction) or "2" (output the excitation light energy.) before the lamp is turned OFF, the value is again set to "1" or "2" after the lamp is turned ON.

During scanning the excitation wavelength, this unit measures the spectrum without the division correction, because the corrected spectrum may not indicate the optimum excitation wavelength for analysis with this unit.

XE TIME

(Display of operating time of xenon lamp)

EX(nm)	EM(nm)	Data
XE TIME		100

This function displays accumulated operating time of xenon lamp (Unit: hour). Press **0** and **Enter** to reset the accumulated time to 0.

Please note: Do not reset the time except for when replacing the lamp.

Z WAVE

(Output holding function at change of wavelength, sensitivity or gain)

EX(nm) EM(nm) Data

This function sets operation/non-operation of output holding function on changing wavelength, sensitivity or gain.

Set value	Correction
0	Operation
1	Non-operation

Please note: If the wavelength is changed, light energy in the background changes and the output value varies. By setting this operation (set value: 0), the output value does not vary when the wavelength is changed. When changing the wavelength during the analysis, set this operation.

This function can not operate correctly when response value is set at "4-(3.0sec)".

RS232C

(Setting of RS-232C and starting communication)

EX(nm)	EM(nm)	Data
RS232C		

Press the **Enter** key to display the commands for starting the communication and setting parameters of RS-232C (see "7.2 Control from External equipment via RS-232C").

CLOSE KEY

(Rejects any key input.)

EX(nm) EM(nm) Data

CLOSE KEY

This function refuses any key input. After pressing **Enter**, any key input is rejected. To cancel this function, press **shift** and **CE** simultaneously.

LAMP ADJUST

(Lamp position adjustment)

EX(nm) EM(nm) Data

LAMP ADJUST

Press **Enter** to enter the xenon lamp position adjusting mode and the light source intensity is displayed. For adjusting procedure, see section "4.7.1 Adjusting the light source position."

S/N CHECK

(Performance check)



Press **Enter** to go to the performance check mode.

In this mode, you can perform a sensitivity check, noise width measurement and calculation of S/N value. The following message is retained on the screen, since the parameter settings of the integrator must be changed for these measurements.

EX(nm) EM(nm) Data

S/N	CHECK
ENTER	

To continue the measurement, press **Enter**. To exit from this mode and return to the initial screen, press **CE**.

6 Performance Verification

Inspection Environment

Carrying out inspection measurements in an environment where the equipment is exposed to sudden changes of temperature or to air currents may affect the repeatability of the data.

Inspections should be conducted in locations where changes of room temperature are less than 2°C. Places where the equipment is exposed to strong air currents from air conditioners should particularly be avoided.

- **Appropriate Location** For proper and safe use of the equipment, select a site in consideration of the following.
- **1. Corrosive Gas and Dust** To extend the life of your equipment and maintain the proper performance standards, avoid using your equipment at a site exposed to corrosive gas or dust.
- 2. Ventilation and Fire If flammable liquids or toxic solvents are used, be sure to ventilate the room well. Never use fire in the room when flammable solvents are used.
- **3. Electromagnetic Noise** Never use your equipment near devices that generate strong magnetism. If the electrical power line generates electrical noise, connect your equipment to a power line with a commercially available filter.
- 4. Workspace Your equipment is designed to be used on a desk or similar surface. Use only a firmly fixed, flat surface that is at least 70 cm deep. If another piece of equipment is to be placed to the left of the unit, be sure to provide at least 15mm of space between the units.

In order to maintain the performance of your equipment, avoid subjecting it to:

- (1) Direct air currents from air conditioners
- (2) Direct sunlight
- (3) Vibration
- (4) High temperature or humidity. The room temperature should be within the range of 4°C to 35°C, and the humidity between 45% and 85%, with minimum fluctuation in room temperature. (However, humidity should be below 70% when the room temperature is 30°C or higher.)

6.1 Outline of System Validation

6.1.1 Outline

For the holistic validation of HPLC system, chromatographic analysis is performed under the analytical conditions specified by the manufacturer. The system status can be judged based on the obtained results. This is because the failure of the LC system may depend on the analytical conditions. Holistic validation procedure described in this manual is the standard for checking the status of the LC system, serving as the basis of inspection.

In routine operation, the operators must perform a system suitability test under the pre-determined analytical conditions. If any problem occurs under such conditions, perform holistic validation first described in this manual. If the result of holistic validation meets the acceptance criteria, the LC system itself is working properly and the cause of the problem is related to the analytical method itself. On the other hand, if the results do not meet the acceptance criteria for holistic validation, it implies that there is a problem in the LC system itself and modular validation should be performed for diagnosis of each LC module.

The repeatability (relative standard deviation) of retention time, peak area, and peak height is measured to check whether the values meet the acceptance criteria.

6.2 Repeatability Test of Chromatographic Data (Isocratic LC)

6.2.1 Purpose

The purpose of this test is to confirm that the chromatographic data can be obtained with good repeatability for the LC system to be inspected. An HPLC System for this inspection consists of the pump, detector, column oven, autosampler and data system.

6.2.2 Preparation for Inspection

- (1) Prepare the following parts and reagents:
 - (a) Isocratic LC system
 - (b) Mobile phase (acetonitrile/water = 4/1 (v/v)
 - * Use acetonitrile and water of HPLC grade.
 - (c) Column
 - (4.6mm ID X 150mm)
 - (d) Sample
 - * Mixture of two components (acridine and anthracene)

[Method for sample preparation]

Place 10mg of acridine and 10mg of anthracene in the volumetric flask of 100ml capacity. Add 100ml acetonitrile and shake it to dissolve. Transfer 1ml portion of the sample solution into the 10ml volumetric flask and add acetonitrile, resulting in a total volume of 10ml.

- (e) Water (HPLC grade or equivalent)
- (f) Isopropyl alcohol

- (2) Check the connection of the units. Refer to the instruction manuals for details of the connection of each unit.
- (3) Before installing the column, observe tubing connection of the LC system. Use tubing with 0.3mm I.D. or less from the outlet of auto injector to the column inlet and from the column outlet to the detector inlet. Length of the tubing should be 30cm or less to minimize the extra-column band broadening.
- (4) Flush the flow line with appropriate solvents dependent on the operation status of the system. General guideline is shown below. To flush the flow line with solvents, connect the inlet and outlet tubing of the column directly using a proper union.

Column should be connected after flushing.

* Newly installed system

Flush the flow line with isopropyl alcohol, then with water at a flow rate of 2 ml/min for ten minutes.

* When low polarity solvent (such as hexane) is used as the mobile phase in the system:

Flush the flow line with isopropyl alcohol, then with water at a flow rate of 2 ml/min for ten minutes.

* When mixture of water and organic solvent, or organic solvent miscible with water (methanol, acetonitrile, etc.), or buffer solution is used as the mobile phase in the system:

Flush the flow line with water at a flow rate of 2 ml/min for ten minutes.

(5) Set the mobile phase (b) and flush the flow line, then connect the column (c) to the LC system.

6.2.3 Procedure

The outline of the procedure is described below:

- (1) Set the flow rate of the pump to 1ml/min and column oven temperature to 40°C. Start pump flow and temperature control. Then, confirm that the solvent comes out from the outlet tubing of the detector and no leak of solvent is observed at the connection.
- (2) Set the parameters of detector.

EX (nm)	:	360	GAIN	:	1
EM (nm)	:	450	LAMP	:	1
Response	:	2	RATIO	:	1
Sens	:	2			

- (3) Monitor the baseline. When a stable baseline is obtained, press the zero point adjustment key of the detector. Then, inject 10µl of mobile phase and confirm no peak is observed.
- (4) Next, inject 10µl of test sample solution. Repeat measurement at least five times.
- (5) Obtain the chromatographic data of retention time, peak area, and peak height from five successive analyses.
- (6) Calculate the average (X) of the data and %RSD using the equations as shown below.

$$SD = \sqrt{\frac{\sum_{i=1}^{5} (XI - XA)^2}{4}}$$

Acceptance Criteria

The acceptance criteria for the %RSD are shown below.Retention time :0.5% or lessPeak area :Acridine 5.0% or less, Anthracene 2.0% or lessPeak height :Acridine 5.0% or less, Anthracene 2.0% or less(It is necessary that changes of room temperature are less than 2°C)

Because the fluorescence intensity of Acridine changes more than that of Anthracene by the fluctuation of the room temperature, the %RSD of Acridine is larger than that of Anthracene.

6.3 Repeatability Test of Chromatographic Data (Gradient LC)

6.3.1 Purpose

The purpose of this test is to confirm that the chromatographic data can be obtained with good repeatability for the LC system to be inspected.

An HPLC System for this inspection consists of the pump, detector, column oven, auto injector, and data system.

6.3.2 Preparation for Inspection

(1) Prepare the following parts and reagents.

- (a) Gradient LC system
- (b) Mobile phases
 Solvent A: acetonitrile (HPLC grade or equivalent)
 Solvent B: water (HPLC grade or equivalent)
- (c) Column (4.6mm ID X 150mm)
- (d) Sample
 - * Mixture of two components (acridine and anthracene)

[Method for sample preparation]

Place 10mg of acridine and 10mg of anthracene in the volumetric flask of 100ml capacity. Add 100ml acetonitrile and shake it to dissolve. Transfer 1ml portion of the sample solution into the 10ml volumetric flask and add acetonitrile, resulting in a total volume of 10ml.

- (e) Water (HPLC grade or equivalent)
- (f) Isopropyl alcohol
- (2) Check connection of the units. Refer to the instruction manuals for details of connection of each unit.
- (3) Before installing the column, check tubing connection of the LC system. Use tubing with 0.3mm I.D. or less from the outlet of auto injector to the column inlet and from the column outlet to the detector inlet. Length of the tubing should be 30cm or less to minimize the extra-column band broadening.
- (4) Flush the flow line with appropriate solvents dependent on the operation status of the system. General guideline is shown below. To flush the flow line with solvents, connect the inlet and outlet tubing of the column directly using a proper union.

The column should be connected after flushing.

* Newly installed system

Flush the flow line with isopropyl alcohol, then with water at a flow rate of 2 ml/min for ten minutes.

* When low polarity solvent (such as hexane) is used as the mobile phase in the system

Flush the flow line with isopropyl alcohol, then with water at a flow rate of 2 ml/min for ten minutes.

* When mixture of water and organic solvent, or organic solvent miscible with water (methanol, acetonitrile, etc.), or buffer solution is used as the mobile phase in the system

Flush the flow line with water at a flow rate of 2 ml/min for ten minutes.

(5) Set the mobile phase (b) and flush the flow line, then connect the column (c) to the LC system.

6.3.3 Procedure

Outline of procedure is described below:

- (1) Set the flow rate of the pump to 1ml/min, initial concentration of solvent B to 50%, and column oven temperature to 40°C. Then start pump flow and temperature control. Then, confirm that the solvent comes out from the outlet tubing of the detector and no leak of solvent is observed at the connection.
- (2) Set the parameters of **RF-2000**.

EX (nm)	:	360	GAIN	:	1
EM (nm)	:	450	LAMP	:	1
Response	:	2	RATIO	:	1
Sens	:	2			

- (3) Monitor the baseline. When a stable baseline is obtained, press the zero point adjustment key of the detector. Then, inject 10µl of mobile phase and confirm no peak is observed.
- (4) Next, inject 10µl of test sample solution. Repeat measurement at least five times.
- (5) Obtain the chromatographic data of retention time, peak area, and peak height from five successive analyses.

(6) Calculate the average (X) of the data and %RSD using the equations as shown below.

$$SD = \sqrt{\frac{\sum_{i=1}^{5} (XI - XA)^2}{4}}$$

XA = (X1 + X2 + + X4 + X5)/5 %RSD = (SD/XA) *100 X1....X5 : Data XA : Average SD : Standard deviation %RSD : Relative standard deviation

Acceptance Criteria

The acceptance criteria for the %RSD are shown below.Retention time:0.5% or lessPeak area:Acridine 5.0% or less, Anthracene 2.0% or lessPeak height:Acridine 5.0% or less, Anthracene 2.0% or less(It is necessary that changes of room temperature are less than 2°C)

* **Reference:** Ternary or quaternary gradient system is inspected in basically the same way.

Because the fluorescence intensity of Acridine changes more than that of Anthracene by the fluctuation of the room temperature, the %RSD of Acridine is larger than that of Anthracene.

7 Control from External Equipment

7.1 Connecting External Input and Output Terminals



Terminal name	Explanation
EVENT OUT Note)	Outputs signals for relay contact (maximum rating: 50V, 0.1A).
	It is turned ON/OFF according to the program or the set value
074D7	of EVENT parameter in AUX. FUNC.
START	Starts this instrument by external contact signals. Time
CTOD	Ctope, this instrument, by systemal context simple. Time
510P	Stops this instrument by external contact signals. Time
	terminal
COM	COMMON terminal for START and STOR signal
	Signal output terminal connected to integrater (1) full coole)
(+) INTEG. IV	Signal output terminal connected to integrator (1V full scale).
(+) REC. 10IIIV	Signal output terminal connected to recorder.
(+) REC. IIIIV	(10mV Iuli scale, 1mV Iuli scale)
GND	Grounding terminal
	EVENT OUT to make a line to make a long for a superal second size the
Please note:	EVENT OUT terminal is turned ON for several seconds in the
	period from powering up until the detector initialization is
	malfunction.

Wiring

(1) Strip the sheath of the connecting cable for approx. 10mm in length at the end.

(2) If the core wire is solid, just insert it into the hole of a terminal while pressing the button above the hole using a small screwdriver or similar tool.

If it is stranded, twist the end well, then insert it into the hole while pressing the button above the hole using a small screwdriver or similar tool. To disconnect the cable, also press the button and pull out the cable.



 Please note: A signal cable is provided with the RF-2000 as a standard accessory. In case two or more circuits or terminals shall be used, prepare cable separately. To protect the cable from breaking of wire, it is recommended to use stranded cables. Cables having a core wire of diameter within the range shown below may be used.
 Solid wire: Ø 0.4-1.0 (AWG 26-18) Stranded wire: Ø 0.3mm² to 0.75mm² (AWG 22-20)

Element wire diameter: Ø 0.18 at the minimum

7.2 Control from External Equipment via RS-232C

An external device or data system such as CHROMELEON can control the **RF-2000** using the RS-232C interface. This section explains how to connect the **RF-2000** to an external system and how to operate the **RF-2000** using external control.

7.2.1 Connecting external equipment

The pins of RS-232C terminal at the rear panel of the **RF-2000** are defined as shown in the figure below. **Pin No. Signal name Function**



Use the optional RS-232C cable (part no. 8914.0115) for the connection between the **RF-2000** and a PC.

The figure below shows the connection in this cable.



7.2.2 Setting parameters and activating the remote control mode

Press the **func** key (or **shift** + **func** key) repeatedly, until the display indicates the command:

EX(nm) EM(nm) Data

Press the **Enter** key to display the commands for setting the parameters for RS-232C.

The commands are shown as follows.

When pressing the **func** key, the display successively displays the commands in the following order.

When pressing the **shift** + **func** key, the commands are indicated in the reverse order.

Activating the Remote control mode:

	EX(nm)	EM(nm)	Data
	CONNE	СТ	
Then press the func	key to set the ba	aud rate:	
	EX(nm)	EM(nm)	Data
	BAUD		
Then press the func	key to set the da	ata word size	:
	EX(nm)	EM(nm)	Data
	DATA E	BITS	
Then press the func	key to set the pa	arity:	
	EX(nm)	EM(nm)	Data
	PARITY	•	
Then press the func	key to set the st	op bit:	
	EX(nm)	EM(nm)	Data
	STOP E	BIT	
(1) Activating rer	note control moc	le	
	EX(nm)	EM(nm)	Data

CONNECT

Press the **Enter** key. The **RF-2000** is is now controlled from the external equipment via the RS-232C interface.

In the remote control mode, the display of the **RF-2000** indicates the excitation wavelength, the emission wavelength and the emission intensity, and **RF-2000** outputs the analog signals of the emission intensity. In this mode, any key input is disabled.

Please note: When turning the power ON again, the remote mode is activated. To release this mode, press the **shift** key and the **CE** key simultaneously.

(2) Setting baud rate

EX(nm)	EM(nm)	Data	
BAUD			
Set va	lue	Baud rate	
1		300 bps	
2		600 bps	
3		1200 bps	
4		2400 bps	
5		4800 bps	
6		9600 bps	

Enter the desired set value with the numeric key and press the Enter key.

(3) Setting data word size

EX(nm)	EM(nm)	Data
DATA E	BITS	

Set value	Data word size
7	7 bit communication
8	8 bit communication

Enter the desired set value with the numeric key and press the **Enter** key.

(4) Setting parity

EX(nm)	EM(nm)	Data	
PARITY			

Set value	Parity
0	No parity
1	Odd parity
2	Even parity

Enter the desired set value with the numeric key and press the Enter key.

(5) Setting stop bit

EX(nm)	EM(nm)	Data	
STOP BIT			
• • •			
Set val	ue	Stop bit	
 Set val	ue	Stop bit No stop bit	
 Set val 0 1	ue	Stop bit No stop bit 1 stop bit	

Enter the desired set value with the numeric key and press the Enter key.

7.2.3 Controlling the RF-2000 via CHROMELEON

⚠ Important:	Requirements:	
	 The communications parameters must be correctly set on the instrument: 	
	Baudrate: 9600	
	Databits: 7	
	Parity: None	
	Stopbits: 1	
	To enter the parameters, proceed as described in the previous section.	

• The detector must be switched to the remote mode. The keyboard will then be deactivated. To release the keyboard lock, simultaneously press the <SHIFT> and <CE> keys for at least 3 seconds. See the previous section.

After the **RF-2000** device driver is correctly activated in the CHROMELEON Installation Program (usually by Dionex Service), you can control the **RF-2000** from your control panel.

Choose "Command..." from the "Control" menu. Click "Emission" to view the available commands and variables for the **RF-2000**. Under "Help", you will see a description of the currently selected command. If you need further assistance on specific commands, please press F1 in CHROMELEON to open the CHROMELEON online Help system. Find the required term via the Index.

Recording Spectra

Spectra cannot be recorded simultaneously with the emission signal. Single spectra scans are performed instead. During this time, the pump flow should be stopped to guarantee a stable status in the flow cell of the **RF-2000**.

The duration of the scans depends on the set scan speed and the scan range. A scan from 200 to 900 nm with the scan speed "Fast" takes approx. 85 seconds. The result of a scan is either an emission spectrum or an excitation spectrum. These spectra are automatically saved in a spectra library. A library by the name **RF-2000.LIB** is created in the timebase directory of the local datasource. All **RF-2000** spectra are added to this library.

Proceed as follows to scan an emission spectrum:

- Stop the pump flow via the Flow=0 command.
- Then perform the "ScanEmission" command. Depending on when the pump flow was stopped and when the scan procedure was started, either a background spectrum or a peak spectrum is saved.
- Perform the analog procedure to determine an optimum excitation value.

Perform the following steps to record a difference spectrum:

- Record a background spectrum as described above.
- Then choose the command "Save Background Spectrum" to save this spectrum separately in the detector.
- Reset the pump flow to the original value via the command "Flow= ... ". Continue until the maximum of the peak to detect is reached.
- Stop the pump flow and record a new spectrum.

The two spectra now exist in the RF-2000 spectra library as well as in individual spectra in two different storage locations within the detector.

• Perform the command "GetSpectraDifference" to receive a difference spectrum.

The detector reacts as if a Scan procedure was started. The result of the difference formation is saved to the RF-2000 spectra library again. This spectrum indicates the optimum emission or excitation value.

▲ Important: The basic requirement for forming difference spectra is the correspondence between the wavelength range defined by the start wavelength and the end wavelength as well as spectra type (Excitation or Emission).

PGM-File: Scan Procedure

The following PGM-file allows you to find optimum values for the "Emission" parameter. Proceed in the same way for the "Excitation" parameter.

-0.1	Pressure.LowerLimit =	0
	Pressure.UpperLimit =	400
	%A.Equate =	"%A"
	%A.Type =	Automatic
	%B.Equate =	"%B"
	%B.Type =	Automatic
	%C.Equate =	"H2O/MeOH 80/20"
	%C.Type =	Automatic
	%D.Equate =	"%D"
	%D.Type =	Automatic
	Flow =	1.000
	%B =	0.0
	%C =	100.0
	%D =	0.0
	Emission.ExWavelength =	275
	Emission.EmWavelength =	350
	Emission.Gain =	1.0
	Emission.Response =	0.5
	Emission.Sensitivity =	Low
	Emission.Step =	Auto
	Emission.Average =	On
0.000	Emission.Autozero	
	Inject	
	Emission.AcqOn	
6.000	; stop flow	
	Flow =	1.000
	Flow =	0.000
	; start scanning	
	Emission.ScanEmission	
	; wait for start of scan	
	Wait	Emission.Scanning
	; wait for end of scan	
	Wait	Not emission.Scanning
	; continue flow	
	Flow =	0.000
	Flow =	1.000
10.000	Emission.AcqOff	
	End	

7.2.4 Controlling the RF-2000 via GynkoSoft

Controlling the **RF-2000** via the GynkoSoft data system is also possible. The device driver for the **RF-2000** must be installed. The sensitivity can be set to "Low" (default) and "High" in GynkoSoft. The setting "Medium" is not supported by GynkoSoft. GynkoSoft supports **RF-2000** functions with the exception of the spectra scan commands, this type of data cannot be recorded or output via GynkoSoft.

8 Maintenance

8.1 Maintenance of Flow Cell

M Important: When the xenon lamp is lit, the flow cell is irradiated by the excitation light beam. Do not stave into the excitation light beam, while the xenon lamp is lit.

8.1.1 Mounting and dismounting the flow cell

- Loosen the screw and take the tubes out of the tube holder.
 Loosen the cell mounting screw and pull out the flow cell upward.
- To mount the flow cell, pushing the flow cell, set the pins in the pin holes to fix the position, and tighten the cell mounting screw.
 Fasten the tubes with the tube holder and tighten the screw to fix the tubes.



8.1.2 Cleaning the flow cell

Dirty cells and piping may cause phenomena such as an unstable baseline or a large baseline noise due to pulsation of the solvent delivery pump. In this case, clean the flow lines with the following procedures.

Item to be prepared	Qty
Syringe	1
Adapter	1
Water	100mL
Acetone	50mL
0.1N nitric acid	50mL

Warning:

Mixing nitric acid with organic solvent may form an explosive substance. Be extremely careful when handling nitric acid.

- (1) Deliver acetone from the cell inlet pipe using the syringe and adapter.
- (2) Use distilled water to flush the flow lines as in step 1.
- (3) Flush the flow lines with 0.1N nitric acid.

- (4) Flush out the nitric acid with distilled water.
- (5) Replace with mobile phase. When the mobile phase used for analysis is not miscible with water, replace the flow lines with acetone and replace it with the mobile phase.



8.1.3 Cell disassembling and cleaning

Please note:

- Carefully handle the flow cell and Teflon packing so as not to stain or damage them. Careless handling will result in deterioration of sensitivity or leakage of liquid. For handling, tweezers coated with resin tube at tips must be used.
- (1) Loosen the M3 screw using a screwdriver. Remove the M3 screws and the flow cell guide.
- (2) Loosen the knurled setscrew, then remove the inlet side Teflon packings, the outlet side Teflon packing and the flow cell.



- (3) Soak the flow cell new inlet side Teflon packing and outlet side Teflon packing in acetone for cleaning and then clean them again with isopropyl alcohol. If the flow cell is still stained, use other organic solvents or detergent (Clean 99, Contaminon, or Exstran etc.) to clean the flow cell. Be sure to remove any such solvent completely before the installation described below.
- (4) Screw in the inlet side knurled setscrew to the bottom.

- (5) Insert the inlet side Teflon packing (4 holes), flow cell, and outlet side Teflon packing (1 hole) into the flow cell holder.
 - Be careful not to set the Teflon packings to wrong position.
- (6) Screw in the outlet side knurled setscrew with the degree that no force is applied to the flow cell, the inlet side Teflon packing and the outlet side Teflon packing.
- (7) Mount the flow cell guide. Tighten the M3 screw using the screwdriver and fix the flow cell guide.
- (8) Tighten the outlet side knurled setscrew by fingers.
- (9) Mount the flow cell unit on the instrument main body and fix it by tightening the cell mounting screw.
- (10) Fasten the tubes with the tube holder and tighten the screw to fix the tubes.



- I Please note:
 - 1. Do not disassemble components other than the cell, Teflon packings, cell retaining ASSY, and knurled setscrews.
 - 2. Be careful not to tighten the knurled setscrews without setting Teflon packings at both sides of the cell as the cell may be destroyed.
 - 3. Be sure to use new Teflon packings to reassemble the unit.

8.1.4 Liquid leakage from flow cell

A flow chart to find probable causes and counter measures for liquid leakage and clogging of pipes is shown in the following.



8.1.5 Countermeasure to LEAK error

If the detector of the leak sensor is wet, even though there is no liquid leakage, LEAK error may be indicated.

To recover this, open the sample compartment, suck the liquids in the drain pan by syringe and wipe the liquids on the leakage detector by cotton stick.



8.1.6 Structure of flow cell

Pressure tolerance of the cell is 1960 kPa {20kgf/cm²} (value in { } is reference.) If the pressure exceeds this value, the cell may be broken. Be careful when applying the back pressure etc.

Structure of the flow cell unit:



8.1.7 Troubles caused by bubbles in the flow cell

(1) Symptoms caused by bubblesWhen bubbles are present in the flow cell, the following symptoms appear.

- 1) Spiky noises are observed.
- 2) The base line drifts in one direction and abruptly changes its drift direction. This symptom appears repeatedly.

3) The base line is not stabilized. Excessive drifts or noises are observed.

When the end of the flow cell outlet pipe is blocked, the pen moves greatly.

(2) Checking the flow cell whether bubbles are present inside or not.

Remove the flow cell from the sample compartment, and hold the cell holder toward a light source (light bulb, fluorescent lamp, etc.) Look into the flow cell located in the cell holder and check the upper and lower side of the cell whether any bubbles are present inside or not.



Example : Bubbles in the flow cell

8.1.8 Driving bubbles out and liquid leakage test

- (1) Remove the flow cell from the instrument.
- (2) Flow methanol or acetonitrile into the flow cell using a syringe or a pump. The flow rate should be as high as possible.
- (3) Ensure that no liquid leaks from the flow cell. If leakage is observed, stop the liquid flow, and make an appropriate measure according to section "8.1.4 Liquid leakage from flow cell".
- (4) Visually check the flow cell light path, and check whether bubbles are present or not. If any bubbles be trapped inside the cell, stop and open the outlet tubing lightly and repeatedly with finger to flush away the bubbles. If bubbles cannot be driven out of the flow cell, drain the liquid from the flow cell once and fill the liquid into the flow cell again, and check the cell for bubbles. If bubbles are still present in the flow cell, repeat these procedures several times.
- (5) When no more bubbles are present, mount the flow cell to the equipment.
- (6) If bubbles cannot be removed by above procedures, the inner surface of the flow cell may be stained. Drain the liquid from the flow cell, and then clean the flow cell according to Sections "8.1.2 Simple Cleaning of flow cell" and "8.1.3 Cell disassembling and cleaning".
- Please note: When a mobile phase which is immiscible with methanol or acetonitrile is used, replace inside the flow paths with a liquid which is miscible first. When a buffer is used, flow distilled

water before replacing the liquid.

8.2 Checking the Xenon Lamp

8.2.1 Note in moving this instrument

▲ Important: When moving this instrument, be sure to remove the xenon lamp and put it in the protective case for the xenon lamp. Pay special attention in handling a xenon lamp since a small explosion is occurred by high pressure charged in the xenon lamp when it is destroyed by an impact. If you touched the xenon lamp valve by hand, clean it with ethyl alcohol before lighting.

8.2.2 Service life of xenon lamp

A xenon lamp is consumable. Luminescence point of the lamp may move around or the lamp may flicker in the course of time. When these phenomena occur, noises in the baseline become extremely large and measurement is not performed accurately.

▲ Important: Xenon lamp used dangerous since it

Xenon lamp used for a long time (1000 hours or more) is dangerous since it may explode. Replace the lamp after using for the ensured hours (500 hours).

Check the flickering of the lamp according to the following procedure:

- (1) Connect a pen recorder to the recorder terminal at the rear of this unit.
- (2) Fill distilled water in the flow cell using the provided syringe. Make sure that no air bubble is there in the flow cell.
- (3) Turn on the power of the recorder, and adjust the zero point to zero accurately.
- (4) Set the parameters as follows.

EX	: 350nm
EM	: 500nm
RANGE	: 7 (x64)
SENS	: 3 (LOW)
RESPONSE	: 2 (0. 5s)
GAIN	: 3 (x16)
RATIO	: 1 (fixed)
Z WAVE	: 1

- (5) Press the **ZERO** key of the **RF-2000** to adjust the zero point.
- (6) Move the emission wavelength at around 350nm so that the displayed value for data becomes around $700 \sim 800$.
- (7) Set the range so that the recorder pen varies in the range of $50 \sim 100$ scales.
- (8) Set the recorder chart speed to about 10mm/min and record for about 30 minutes.
- (9) After completing recording, calculate the noise ratio as shown in the figure. If the noise ratio is 5% or more, the lamp is at the end of its service life. Replace it with a new one.


8.2.3 Operating time of xenon lamp

Operating time of the xenon lamp is displayed in XE TIME in AUX. FUNC. The value is automatically accumulated while the lamp is lit on. Enter **0** and **Enter** to reset the time when replacing the lamp. Operating time of lamp is indicated in the unit of hour.

Please note: Reset the time only when replacing the lamp.

8.2.4 Replacing procedure for xenon lamp

Replace the xenon lamp following the same procedure as described in section "4.6 Installing the Xenon Lamp" and "4.7 Adjusting the Xenon Lamp Alignment".

⚠ Important:

Xenon lamp is extremely hot right after turning the power switch OFF. Wait for more than an hour after turning the power switch OFF and be certain that the xenon lamp is no longer hot by opening the light source chamber cover before replacement.

When LAMP of AUX FUNC is set to "0", xenon lamp is turned off with the power still supplied. (Refer to "5.5.1 AUX FUNC.") The lamp is cooled down in about 30 min. under this condition. After cooling down of the lamp, turn the power switch OFF, pull out the power cable and wait for 5 min before replacing the lamp.

8.3 Replacing the Fuse

Warning:

This instrument uses two of the following fuses. Be sure to replace the fuses of the same type and capacity.

for power supply: 220-240 VAC Part no. 706.072.0165221 250V 3.15AT

Replace the blown fuse according to the following procedure. Be sure to use a fuse specified in the Accessories and Spare Parts list.

- (1) Turn the power switch OFF.
- (2) Disconnect the power cable from the power supply connector.
- (3) Open the fuse holder cover with a regular screwdriver.



(4) Take out the fuse holder to this side.



- (5) After replacing the fuse, push the fuse holder in with the arrow side faced down.
- (6) Close the fuse holder cover until it clicks.

8.4 Changing the Set Power Supply Voltage

To change the power voltage setting of this unit, it is necessary to change the setting of the power supply connector unit, of the Xenon lamp power supply and the fuse.

Warning: Setting of the power supply voltage for this unit should conform to the operating power supply voltage.

8.4.1 Setting of the power supply connector

(1) Turn OFF the power switch.

STOP

- (2) Pull out the power supply cable from the power supply connector.
- (3) Open the fuse holder cover at the rear of this unit by a slotted screwdriver (see illustration in the previous section).
- (4) Change the rotary switch showing the voltage in the fuse holder as follows.



Fuse holder cover

Power supply voltage

100V±10% 120V±10% 220V±10% 230V±10% 240V±10%

Setting of rotary switch 100Va.c 120Va.c

230Va.c 230Va.c 230Va.c 240Va.c

(5) Close the fuse holder cover until a clicking sound is heard.

Warning: If the power supply voltage is 100 or 120 Va.c, a different fuse type must be used! Please contact Dionex Service.

8.4.2 Setting of the Xenon lamp power supply

- (1) Turn OFF the power supply switch.
- (2) Remove the rubber lid of the power supply voltage switch of the xenon lamp located on the right side of this unit.
- (3) Conform the selection switch of the xenon lamp power supply to the power supply voltage as the following table.



Warning: If the power supply voltage is 100 or 120 Va.c, a different fuse type must be used! Please contact Dionex Service.

(4) Push in the rubber rid into the hole to seal the switch.

8.5 Periodical Cleaning

STOP

If the cover or the panel of the unit is stained, wipe out dirt or dust with soft cloth or a piece of paper. If necessary, use a non-ionic detergent.

9 Troubleshooting

9.1 Summary of the wost Frequently Problem
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Symptom	Probable cause	Remedy
 Power is not supplied even by turning the power switch ON. 	Fuse for power supply is blown.	Check the fuse in the fuse holder at the rear of this instrument and replace if it is blown.
	Causes other than above cause.	Contact the Dionex Service department.
 Xenon lamp is not lin on. 	AUX. FUNC. LAMP is "0".	Set the AUX. FUNC LAMP to "1" (light ON). (See "5.5 AUX. FUNC.")
	Failure of wiring contact	Check the connection of high voltage cable connected to the + pole of the xenon lamp. Be sure to disconnect the AC cable from the plug socket before this check. It is very dangerous if the power is accidentally turned ON.
	Lamp is not cooled.	Leave the lamp for about 10 minutes to allow it cool.
	Power supply for xenon lamp is defective.	Contact the Dionex Service department.
	An overheat error occurs.	Contact our service sales office or representatives. (See "9.2 List of Error messages".)
 No signal comes out. 	Lamp is not lit.	Referring to item 2, check the lamp.
	Adjustment of light source position is considerably bad.	Readjust the light source position.
	Measuring conditions are not set properly.	Check the settings of wavelength and sensitivity.
	Causes other than above causes.	Contact the Dionex Service department.
4. Improper S/N value (Signal level is low.) 4. Improper S/N value	Improper installation of the flow cell	Check that the flow cell is properly installed in the sample compartment. (See "8.1.1 Mounting and dismounting the flow cell")
	Improper light source position	Readjust the light source position. (See "4.7 Adjusting the Xenon Lamp Alignment")

Symptom	Probable cause	Remedy
(cont.:) 4. Improper S/N value (Signal level is low.)	Bubbles in the flow cell	Drive bubbles out of the flow cell. (See "8.1.8 Driving Bubbles out of Flow Cell and Liquid Leakage Test")
		Check that the knurled setscrews are tightened. (See "8.1 Maintenance of Flow Cell")
		Replace the packings to new ones. (See "8.1 Maintenance Flow Cell")
	Improper wavelength	Check wavelength.
		If the wavelength is out of the specified range, contact our service department for wavelength adjustment.
	Improper sensitivity adjustment	Contact our service department for sensitivity adjustment.
	Defective-HV board (preamplifier circuit)	Contact our service department.
	Causes other than above causes	Contact our service department.
5. Improper S/N value (Noise level is excessive.)	Improper installation of flow cell	Check that the flow cell is properly installed in the sample compartment. (See "8.1.1 Mounting and dismounting the flow cell")
	Improper light source position	Readjust the light source position. (See "4.7 Adjustment of Xenon Lamp Alignment")
	Flickering of the xenon lamp	Check the xenon lamp for flickering. (See "8.2.2 Service life of xenon lamp") Replace the xenon lamp. (See "4.6
	Bubbles in the flow cell	Drive bubbles out of the flow cell. (See "8.1.8 Driving Bubbles out of the Flow Cell and Liquid Leakage Test") Check that the knurled setscrews are tightened (See "8.1 Maintenance of Flow Cell") Replace the packings to new ones (See "8.1 Maintenance of Flow Cell")
	Causes other than above causes	Contact our service department.

Symptom	Probable cause	Remedy
6. Raman peak of water is not displayed.	Impurities in water	Use new distilled water.
	Stains of the flow cell	Clean the cell (See "8.1 Maintenance of Flow Cell") Replace the cell (See "8.1 Maintenance
	Bubbles in the flow cell	Drive bubbles out of the flow cell. (See "8.1.8 Driving Bubbles out of the Flow Cell and Liquid Leakage Test") Check that the knurled setscrews are tightened (See "8.1 Maintenance of Flow Cell")
	Improper wavelength	Replace the packing (See "8.1 Maintenance of Flow Cell") Check wavelength. If the wavelength is out of the specified range, contact our service department for wavelength adjustment.
	Defective M2 mirror Causes other than above causes	Contact our service department for cleaning of the mirror. Contact our service department.
7. There is no output at the recorder terminal.	Range is set to 0.	Set the range to an appropriate value.
	Causes other than above cause.	Contact the Dionex Service department.
8. "OVER" is displayed	Integrator output is over to + side. Sensitivity setting (SENS) is too high. ("OVER" is still displayed even if ZERO key is pressed.) Causes other than above	Press the ZERO key to adjust zero point. Set sensitivity lower (see "5.2.5 Setting Sensitivity")
	causes	department.

Symptom	Probable cause	Confirmation of cause	Countermeasures
Transient spiking	Bubbles flowing through the cell	With the flow cell tubing connected, remove the cell, and check it under flow, check for bubbles or cell fouling.	 Apply pressure to the cell outlet side lightly. Stop or open the end of the outlet pipe with a finger lightly.
Sawtooth baseline	Bubbles flowing through the cell	n	 Degas the mobile phase.
Continuous spiking	Bubbles flowing through the cell/ Bubbles trapped in the cell	Π	3) Clean the cell interior with isopropyl alcohol. Remove the cell inlet tubing connector and force isopropyl alcohol from the outlet side with a syringe. Take care not to allow air come in and connect the inlet side connector and flow mobile phase.
Spiking occurring at every stroke of the pump. No equilibration of baseline.	Bubbles trapped in the cell	n	4) Cell rinsing
	Cell window is		
Drift Noise is excessive	stained.		
Drift	Impurities in the mobile phase	The drift stops after stopping liquid supply.	5) Check the pump, column, reservoir and mobile phase for dust or impurities.
Noise is excessive.	Light source lamp degradation	Check the operating time of the lamp.	6) Replace the lamp with a new one.

Symptom	Probable cause	Confirmation of cause	Countermeasures
Baseline wanders.	The unit is in the presence of a strong air current.		7) Change the location of the instrument or place it in a protective environment.
Noise occurs corresponding to the pump stroke (when using a small plunger pump)	Mobile phase pulsation		8) Eliminate pulsation from the pump. Use a damper.

9.2 List of Error Messages

9.2.1 Errors which cannot be reset

Turn the power of the instrument OFF and turn it ON again. If the error message persists, turn OFF the power and contact your Dionex Service Representative.

(1) RAM error

It is displayed when something is wrong with the RAM.

EX(nm)	EM(nm)	Data
RAM	FAILURE	

(2) ROM error

It is displayed when something is wrong with the ROM.

EX(nm)	EM(nm)	Data
ROM F	AILURE	

(3) OVERHEAT error

It is displayed when the temperature in the instrument is abnormally high. If this error occurred, the Xenon lamp is not lit even if the power is turned ON again.

To repair this instrument and to cancel this error, contact our sales office or representatives.

9.2.2 Errors which can be reset

Alarm sound stops by pressing **CE** and the next step is proceeded.

(1) Set value loss error

It is displayed if the parameters or the time program set previously are lost at power on. Press **CE** and the instrument becomes operational. Set parameters again.



(2) File type error

It is displayed when the spectrum type or wavelength range for the file 0 and those for the file 1 or 2 which is to be output in SPC PLOT are different.

EX(nm)	EM(nm)	Data
ERR F	ILE TYPE	

(3) No file error It is displayed when there is no data in the file in SPC PLOT or there is no data in the file 0 when the file 1 or 2 is specified (subtracted spectrum output).



Emission motor origin (fine)

(7) Wavelength calibration data error It is displayed when the wavelength calibration data entered in the unit is abnormal.



(8) LAMP error

It is displayed when the xenon lamp is not lit.

- To lit the xenon Lamp again, operate the following procedure. Turn the power OFF and then turn the power ON.
- Set the "LAMP" parameter in additional functions to "1".
- If this error message is displayed again, refer to "8.2 Checking the Xenon Lamp" and "9 Troubleshooting".

EX(nm)	EM(nm)	Data
ERR L	AMP	

(9) LEAK error

It is displayed when leak of liquid from the flow cell is detected. See "8.1 Maintenance of the Flow Cell".

EX(nm)	EM(nm)	Data

ERR LEAK

Please note: If the detector of the leak sensor is wet, even though there is no liquid leakage, LEAK error may be indicated. To recover this, refer to "8.1.5 Countermeasure to LEAK error".

9.2.3 Error which can be reset with the shift + CE key.

 Rejecting key input
 When "CLOSE KEY" of the additional function is set or **RF-2000** is controlled from a external equipment via RS-232C, any key input is rejected. To release these settings, press the **CE** key and the **shift** key simultaneously. (Refer to "5.5 Additional Functions" and "7.2 Control from external equipment via RS-232C".)

EX(nm) EM(nm) Data

10 Specifications

Light source:	150W xenon lamp	
Light source chamber:	Ozone self dissolving type lamp house	
Monochromators:	Ion brazed holographic concave diffraction grating monochromators F/2. 4 for both excitation and emission sides	
Number of grooves:	900 lines/mm	
Wavelength range:	200~650nm and 0 order	
Bandwidth:	15nm for both excitation and emission sides	
Wavelength accuracy:	±2nm	
Wavelength repeatability:	±0.2nm	
Wavelength scanning speed:	SUPER, FAST, MEDIUM and SLOW, 4 steps SUPER: Approx. 3,000nm/min FAST: Approx. 600nm/min MEDIUM: Approx. 120nm/min SLOW: Approx. 24nm/min	
Wavelength moving speed:	Approx. 15,000nm/min	
Detector:	Signal side: Photomultiplier tube Monitor side: Photodiode	
Measuring method:	Ratio compensation system with monochromatic light monitoring.	
Time constant:	0.1, 0.5, 1.5and 3s, 4 steps.	
Sensitivity:	HIGH, MED and LOW, 3 steps (sensitivity difference is approx. 32 times)	
Gain:	x1, x4, and x16, 3 steps (approx. 1 time, 4 times, and 16 times)	
Range:	1/∞ (0mV), x1, x1/2, x1/4, x1/8, x1/16, x1/32, x1/64, x1/128, x1/256	
Output terminals:	Recorder: 1mV, 10mV Integrator: 1V/FS Contact signal output (EVENT OUT output)	
Input terminals:	Contact signal input (START input, STOP input)	
Cell:	12µl square quarts flow cell, pressure tolerance 1.96Mpa {20kgf/cm ² } (figure inside { } is reference.)	
Communication:	Controllable by CHROMELEON via RS-232C	
Display unit:	Fluorescent display tube	
Measuring function:	 * Monitor function Auto zero, Base line shift * Time program function Up to 32 steps can be programmed. * Wavelength scanning function Excitation and Emission wavelength scanning Differential spectrum Plot output 	

Filing:	 * Measurement condition One measurement condition can be stored. * Time program One time program can be stored. * Spectrum Three spectra can be stored.
	(However, they are not backed up when the power is turned OFF.)
Dimensions:	260 (width) x 520 (depth) x 205 (height) mm
Weight:	Approx. 17kg
Power supply:	AC 100V±10%, 120V±10%, 220V ±10%, 230V±10%, 240V±10% 50, 60Hz
Power consumption:	350VA
Operating temperature range:	4 ~ 35°C
Operating humidity range:	$45 \sim 80\%$ (However, humidity should be 70% or below when the temperature is 30°C or higher.)
S/N:	S/N at the Raman peak of distilled water is 300 or higher. (when EX = 350nm and RESPONSE = 1.5s.)

11 Standard Accessories

Category	Description	Part No.	Qty
Parts	Signal cable	706.228.3411091	1
	Power cable (for 220 ~ 240V)	706.071.6081406	1
	RS-cable	8914.0115	1
	RS-adapter	8914.0110	1
	Syringe	706.046.00001	1
	Syringe adapter	706.204.01647	1
	Male nut PEEK	706.228.18565	4
	Male to male union 1.6C	706.228.1600403	2
	Tefzel tube (1.6 x 0.3, 2m)	706.228.1849506	1
	SUS tube (1.6 x 0.3, 0.5m)	706.228.2230500	1
	Drain tube	706.016.3135002	1
Consumable parts	150W xenon lamp	5057.1000	1
	Fuse3.15A (for 220 ~ 240V)	706.072.0165221	2
	Teflon packing inlet (3 pcs)	706.228.3413781	2
	Teflon packing outlet (3 pcs)	706.228.3413681	2
Tools	Screwdriver		1
Other	RF-2000 Operating Instructions	5820.5750	1
	Certificate of compliance		1
	Label Ver.3		1

⚠ Important:

When carrying out maintenance, be sure to use the parts described in the sections 10 and 11. Normal function of the system is not guaranteed when other parts are used.

12 Replacement Parts

Description	Part No.	Remarks
150W xenon lamp	5057.1000	Light source
Cell for LC fluorescence monitor	706.200.66504	Part for flow cell
Inlet side Teflon packing	706.228.3413781	Part for flow cell (3 pieces)
Outlet side Teflon packing	706.228.3413681	Part for flow cell (3 pieces)