# INSTRUCTION MANUAL

# HITACHI F-7000 FLUORESCENCE SPECTROPHOTOMETER FL SOLUTIONS PROGRAM

(OPERATION EDITION)

# Please read through this manual carefully and keep it in a safe place.

- Before using the instrument, read the safety instructions and precautions carefully.
- Keep this manual in a safe place nearby so it can be referred to whenever needed.

## **INSTRUCTION MANUAL**

# HITACHI F-7000 FLUORESCENCE SPECTROPHOTOMETER FL SOLUTIONS PROGRAM (OPERATION EDITION)

Hitachi High-Technologies Corporation

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- 3. Hitachi High-Technologies Corporation assumes no liability for any direct, indirect, or consequential damages arising from use not described in this manual. Utmost care must be exercised when using the instrument.
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#### **PREFACE**

We thank you for purchasing the Hitachi High-Technologies Corporation fluorescence spectrophotometer.

The instrument is intended for fluorescence analysis and should not be used for other purposes.

The fluorescence spectrophotometer is designed for use by persons having a basic knowledge of chemical analysis. Keep in mind that improper use of analytical instruments, chemicals or samples would result not only in wrong analytical results but also in consequences adverse to safety and possibly personal injury.

Be sure to read this manual thoroughly before use to ensure correct use of the instrument.

Carefully read and understand all of the safety instructions prior to operation or maintenance of the instrument.

This manual should be kept at hand so it can be referred to whenever needed.

The contents of this manual are subject to change without prior notice for improvement.

#### **ABOUT THIS MANUAL**

The Hitachi High-Technologies Corporation fluorescence spectrophotometer is accompanied by the following two instruction manuals.

- F-7000 Fluorescence Spectrophotometer Instruction Manual –
   Maintenance Edition
- F-7000 Fluorescence Spectrophotometer Instruction Manual FL Solutions Program - Operation Edition

The cautionary instructions concerning safety are also contained in the maintenance edition of the instruction manual. Be sure to read them before attempting use.

This manual describes the operational procedures for the FL Solutions program formulated for the Hitachi High-Technologies Corporation fluorescence spectrophotometer.

Be sure to read through this manual before attempting operation.

#### **IMPORTANT**

#### **Precautions on CE Conformity Marking**

#### 1. Electromagnetic Compatibility Requirement

This equipment conforms to the EMC Directive 89/336/EEC and has the CE conformity marking. It is designed to satisfy the European Norm EN61326 (1997) and its amendment 1 (1998) & amendment 2 (2000).

This equipment is classified as Class A of EN61326. So, this equipment must not be used in domestic establishments nor in establishments directly connected to a low voltage power supply network which supplies buildings used for domestic purpose.

And this equipment is also designed to comply with table 1 "Minimum immunity test requirements" and with evaluation table 2 "Continuous unmonitored operation". If the equipment is used near an intense electromagnetic source, however, interfering noise may be given to the instrument to cause an adverse effect on its performance or functionality.

#### 2. Safety Requirement

This equipment also conforms to the LVD Directive 73/23/EEC and has the CE conformity marking. Its is designed to satisfy the European Norm EN61010-1: 2001.

Since this instrument is classified in class 1, pollution degree 2 of EN61010-1, be sure to use it in a suitable environment and ground it appropriately.

#### Information for Users on WEEE (only for EU Countries)



This symbol is in compliance with the Waste Electrical and Electronic equipment directive 2002/96/EC (WEEE).

This symbol on the product indicates the requirement NOT to dispose the equipment as unsorted municipal waste, but use the return and collection systems available.

#### Information on Disposal for Users

#### 1. In the European Union

If you need to discard this product or discard user serviceable parts:

Please contact your local sales representative or distributor who will inform you of the recycle of the product.

You might be charged for the costs arising from take-back and recycling.

#### 2. In other Countries outside the EU

If you wish to discard this product, please contact your local authorities and ask for the correct method of disposal.

#### **Warranty on Product**

The Hitachi fluorescence spectrophotometer is warranted to be free from defects in material or workmanship under normal use within the product specifications indicated in this manual and under conditions given below. This warranty is void if the instrument is not used in accordance with the instruction manual.

#### (1) Scope of Warranty

This instrument will be repaired without charge if it malfunctions due to a defect in workmanship during the warranty period.

Note that a substitute part may be used for repair, or replacement with an equivalent product may be made instead of repair.

Such system components as a personal computer and printer to be updated frequently for improvement may not be available in original versions at the time of replacement.

In addition, the instrument resold without consulting Hitachi, consumables and failure of parts that have reached the end of specified useful life are excluded from the warranty.

#### (2) Warranty Period

One year from the date of initial installation (Excluding consumables such as a xenon lamp)

#### (3) Limitations and Exclusions on Warranty

Note that the following cases are excluded from the scope of this warranty.

- (a) Failure due to operation at a place not meeting the installation requirements specified by us.
- (b) Failure due to power supply voltage/frequency other than specified by us or due to abnormality in power supply.
- (c) Corrosion or deterioration of the tubing due to impurities contained in gas, air or cooling water supplied by the user.
- (d) Corrosion of the electric circuits or deterioration of the optical elements due to highly corrosive atmospheric gas.
- (e) Failure due to use of hardware, software or spare parts other than specified by us.
- (f) Failure due to improper handling or maintenance by the user.
- (g) Failure due to maintenance or repair by a service agent not approved or authorized by us.

- (h) Failure due to relocation or transport after initial installation.
- (i) Failure due to disassembly, modification or relocation not approved by us.
- (j) Failure due to acts of God, including fire, earthquake, storm, flood, lightning, social disturbance, riot, crime, insurrection, war (declared or undeclared), radioactive pollution, contamination with harmful substances, etc.
- (k) Failure due to computer virus infection.

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#### (4) About the Warranty Card

Be sure to keep the warranty card in a safe place, since it will not be re-issued.

#### Installation, Relocation and After-sale Technical Service

Installation of the instrument shall be carried out by or under supervision of our qualified service personnel or our authorized service agent. Before installation of the instrument, the customer shall make preparations for satisfying the installation requirements in accordance with this manual.

When relocation of the instrument becomes necessary after initial installation (delivery), please notify our local sales representative or service office nearest you.

#### **Technical Seminars and Training for Customers**

For the customers to acquire in-depth understanding of the analytical instruments, technical seminars and customer training courses are available at our or your site. For further information, contact our local sales representative.

(The technical seminars and customer training courses are available at charge.)

#### **Precautions on Disposal of Instrument**

Although at present this instrument does not use materials that would directly cause environmental disruption, the environmental protection laws and regulations may be revised or amended. When disposing of this instrument, be sure to check the latest issues of the relevant laws and regulations or consult your local Hitachi High-Technologies Corporation service office.

#### Other Precautions

#### **Handling of Chemicals and Samples**

- (1) The user is responsible for following relevant legal standards and regulations in the handling, storage and discarding of chemicals and samples used in analytical operations of the instrument.
- (2) Reagents, standard solutions and accuracy-control samples shall be handled, stored and discarded as instructed by the respective suppliers.

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# SAFETY SUMMARY

#### PRECAUTIONS ON SAFETY

Before using the F-7000 fluorescence spectrophotometer, be sure to read the following safety instructions carefully.



## General Safety Guidelines

- Follow all the operating procedures provided in this manual.
- Installation and maintenance of this instrument shall be carried out by service personnel qualified for this purpose.
- Be sure to observe the warnings indicated on the product and in the instruction manual. Failure to do so could result in personal injury or damage to the product.
- The hazard warnings which appear on the warning labels on the product or in the manual have one of the following alert headings consisting of an alert symbol and signal word DANGER, WARNING, or CAUTION.

4	À

**DANGER**: Indicates an imminently hazardous situation

which, if not avoided, will result in death or  $% \left\{ 1,2,\ldots ,2\right\}$ 

serious injury.

(This warning does not apply to this product.)

**WARNING**: Indicates a potentially hazardous situation which, if not avoided, can result in death or

serious injury.

**CAUTION**: Indicates a hazardous situation which, if not

avoided, will or can result in minor or moderate injury, or serious damage to the

product.

: The alert symbol shown at left precedes

every signal word for hazard warnings, and appears in safety-related descriptions in the

manual.

"NOTE" and "NOTICE" are heading words which do not concern personal safety directly.



# **A** SAFETY SUMMARY



## General Safety Guidelines (Continued)

**NOTICE**: Used to indicate an instruction for preventing damage

to the product.

NOTE : Used to indicate an instruction for ensuring correct

use of the product and accurate analysis therewith.

 Do not modify the instrument, replace parts that are not userserviceable, use non-specified parts, nor remove safety devices, as it could be hazardous.

- Installation at delivery, maintenance and relocation should be referred to service personnel qualified by us.
- Do not perform any operation or action other than described in this manual. When in doubt, please contact our local sales representative or service office nearest you.
- When using a chemical for analytical operation, be sure to provide proper ventilation in the laboratory room as per local requirements.
   Inadequate ventilation could endanger your health.
- Keep in mind that the hazard warnings in this manual or on the product cannot cover every possible case, as it is impossible to predict and evaluate all circumstances beforehand.
   Be alert and use your common sense.



#### **Safety Notices on Fluorescence Spectrophotometer**

#### **Electricity**

- Make sure that the fluorescence spectrophotometer is supplied with power of 100 V AC, 700 VA or more (50 Hz or 60 Hz). If the power voltage fluctuates or noise is applied to the power line, the spectrophotometer may be adversely affected, resulting in any trouble in it.
- Be sure to provide grounding connection together with power line connection. The grounding resistance should be 100 ohms or less.
   Check that the grounding connection is proper. If the grounding connection is improper, the spectrophotometer may become vulnerable to external noise or stray voltage may be produced in it, resulting in an electric shock hazard.
- The high voltage circuit is used inside the spectrophotometer.
   To prevent electric shock, do not open covers during operation unless otherwise necessary.

#### Source of Heat/Flame

Do not smoke or hold a flame near the fluorescence spectrophotometer.

#### **Data Backup**

Data stored on the hard disk may become unusable due to a system failure, wrong operation, computer virus infection, etc. To ensure data integrity in case of accidental damage to the hard disk, **periodically make backup copies of hard disk files onto floppy disks**.

To prevent an erroneous operation on the hard disk, always reserve approx. 100 MB of free space as a working area for application software.



#### **Protection against Computer Viruses**

If any program/data is damaged suddenly or an unexpected operation/screen is encountered, the personal computer is suspected of being infected by a computer virus.

Computer viruses are malicious programs that sneak into personal computers to cause misbehavior or damage to data. And, a program designed to offer protection against computer viruses is called a vaccine program.

Possible causes of virus infection are:

- Downloading a virus-laden program through communication.
- Using a floppy disk or other storage medium infected by a virus.

Note also that once any personal computer is infected by a virus, it may spread to other computers via communication or storage medium.

Never use a program or storage medium that is suspected of containing a virus.

If there is a possibility of virus infection, check for a virus using a vaccine program. Note, however, that some kinds of vaccine programs cannot eradicate particular viruses. So, be sure to make a backup of hard disk files beforehand.

The user is requested to prepare a vaccine program and carry out virus removal on his or her own responsibility.

#### **Power Interruption**

On occurrence of momentary power voltage drop due to power interruption or lightning, the personal computer may become faulty or the system software, application software or data may be damaged. For protection against momentary power voltage drop, it is advisable to use an AC uninterruptible power supply unit (stated according to the Japanese Electronic Industry Development Association guidelines for protection against momentary power voltage drop in personal computers).



#### **Turning On/Off Personal Computer**

Do not turn off power to the personal computer while the hard disk or floppy disk drive is active. If power to the personal computer is turned off while the hard disk or floppy disk is being accessed, the personal computer may become faulty or data/software stored in it may be damaged.

Before turning off power to the personal computer, be sure to quit the fluorescence spectrophotometer control/data processing program (FL Solutions program) first and then take the shut-down procedure using the system software.

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## 1. FUNCTIONS AND SCREEN COMPOSITION

### 1.1 Functions

In this program, the following measurement modes are available:

- Wavelength scan
- Time scan
- Photometry
- 3-D scan
- 3-D time scan

#### **Wavelength Scan**

- (1) Photometric value data can be printed at the specified wavelength intervals or at up to 12 specified wavelengths.
- (2) The function keys provide functions unique to the wavelength scan mode.

  Measurement: [F4], Stop: [F6]
- (3) Wavelength scan measurement can be repeated max. 99 times. And, a repetition period is settable.

#### **Time Scan**

- (1) Kinetics, or rate calculation will be performed.
- (2) Photometric value data can be printed at the specified time intervals or at up to 12 specified time points.

#### **Photometry**

- (1) Quantification method is selectable among specified wavelength, peak area, peak height, peak ratio and derivative methods.
  - Up to 20 standards can be measured. A calibration curve is prepared by drawing polygonal lines through each point of standard concentrations or a regression calibration curve is prepared by least squares. It is also possible to measure up to 20 standards and prepare a calibration curve based on an average value of standard concentrations.
- (2) A calibration curve can be redrawn by re-measuring a specific standard. In a calibration curve graph on the screen window, a value at each point can be read out using the cursor.
- (3) A calibration curve can be printed.A calibration curve can be drawn by inputting its factor value.

#### 1.1 Functions

- (4) Measurement data and a calibration curve are displayed on the same screen window. It is therefore possible to carry out measurement while observing the calibration curve.
- (5) According to the coefficient of determination calculated automatically, judgment can be made on a degree of matching between standard values and a calibration curve. Two/three-wavelength calculation is permitted in preparation of a calibration curve.
- (6) Statistical calculation is possible.Mean value of measured data and standard deviation (SD) can be calculated.
- (7) Upper/lower limit judgment will be performed.
- (8) Spectrum display under photometry
  In the data measured by a quatification method; peak height, peak area, peak ratio or derivative,
  a spectrum due to wavelength scan is displayable.

#### 3-D Scan

- (1) Spectral calculation can be performed using the measured data.
- (2) A peak table can be displayed.
- (3) A bird's-eye view/synchronous spectrum can be displayed and printed.
- (4) Printing conditions are selectable.
  In use of the DDE function, the wavelength interval and start/end wavelengths of data list can be selected.

#### 3-D Time Scan

- (1) A peak table can be displayed.
- (2) A bird's eye view/contour lines can be displayed and printed.
- (3) Printing conditions are selectable.

#### **Secondary Processing of Data**

Measured data can be saved, and a variety of processings can be carried out on the saved data.

(1) Automatic reading of data (spectrum tracing)

By placing the cursor on the currently displayed spectral graph, a photometric value at the cursor position can be read out.

#### (2) Peak detection

Peaks and valleys of the currently displayed spectrum are detected automatically.

#### (3) Change of scale

A measured spectrum can be enlarged or reduced by changing the ordinate and abscissa scales.

#### (4) Smoothing

A measured spectrum can be smoothed for elimination of noise components.

#### (5) Derivative

Spectral derivative is obtainable. It is permitted to specify an order of derivative, that of smoothing and a count value of data.

#### (6) Spectrum calculation

Fundamental arithmetic operations can be made between spectra or a spectrum can be multiplied with a factor.

#### (7) Area

The area value of the specified peak will be calculated.

The average area value is also calculable among peaks.

#### (8) File conversion

Measurement data can be converted into an ASCII text file (\*.txt) or JCAMP-DX file (\*.dx). It is also possible to save a spectrum and calibration curve graph in a Windows meta file (\*.wmf).

#### (9) Display/print font setting

The font, style and size of characters to be displayed and printed can be changed as desired.

#### (10) Data acquisition

Photometric value data on the data processing window and monitor window can be acquired. Acquisition is allowed within 100 data.

#### (11) Printing items collectively specifiable

Printing items on overlaid spectra can be specified collectively.

#### 1.1 Functions

- (12) Specification of printing itemsItems in a peak table can be selected and printed.
- (13) Portrait or Landscape printing settable
  Printing can be made Portrait or Landscape.
- (14) Collective spectrum printingPrinting can be carried out just by specifying a file.
- (15) Selection of spectrum to be displayedThe spectrum to be displayed for overlaying is selectable.
- (16) Automatic saving mode ON/OFF settable ON/OFF can be set in the sample name/comment input window. Automatic saving is effective in all the modes that use a sample table.
- (17) Data in each mode can be transferred to Microsoft® Excel.(This is valid when Microsoft® Excel 2003 has been installed in PC.)
- (18) After copying spectral data into commercial software such as Microsoft® Word, its scale, line type, color, etc. are changeable on the software.
- (19) The report generator (separately available, P/N 250-0455) can be used.

# 1.2 Installation of FL Solutions Program

#### 1.2.1 Specifications of Personal Computer

For running the FL Solutions 2.1 program on Microsoft Windows XP Professional, the following specifications must be met.

OS : Microsoft Windows XP Professional

CPU : CPU that allows the above OS to run in your computer

Main memory : Minimum 256 MB

Hard disk : Minimum free space 5 GB FD drive : One 3.5-inch FD drive

CD-ROM drive : 24× or faster

Interface : USB1.1 or USB2.0  $\times$  1 or more, RS-232C  $\times$  1 or more

#### 1.2.2 Setup of FL Solutions Program

For installing this program, implement the following procedure.

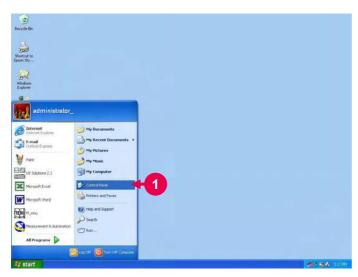
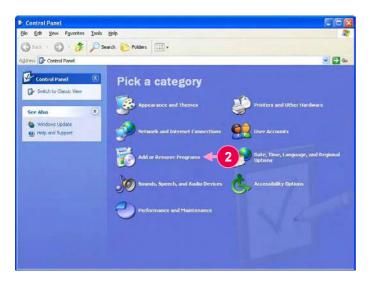


Fig. 1-1

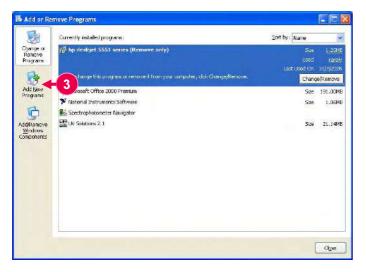
- 1 Click the **start** button of Windows XP and select "Control Panel".
  - A window as shown in Fig.
     1-2 will appear.



Select "Add or Remove Programs."

A window as shown in Fig.
 1-3 will appear.

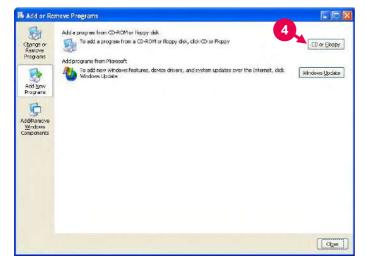
Fig. 1-2



3 Select "Add New Programs."

A window as shown in Fig.
 1-4 will appear.

Fig. 1-3



4 Set the furnished CD in the CD-ROM drive and click CD or Floppy.

A window as shown in Fig.
 1-5 will appear.

Fig. 1-4

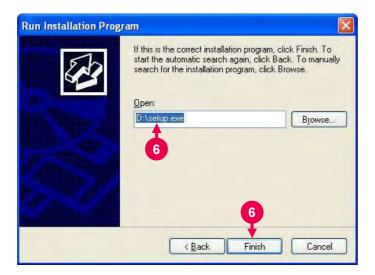


A ........

Click Next.

A window as shown in Fig.
 1-6 will appear.

Fig. 1-5



6 Make sure 'D:\setup.exe' is displayed, and then click Finish.

 A window as shown in Fig. 1-7 will appear.

Fig. 1-6

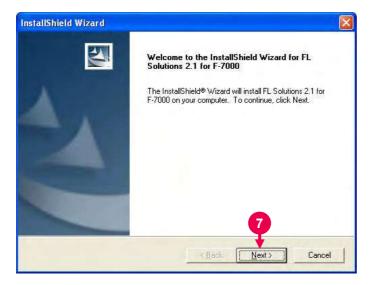


Fig. 1-7

#### 7 Click Next.

A window as shown in Fig.
 1-8 will appear.

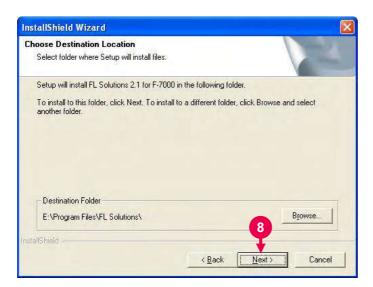


Fig. 1-8

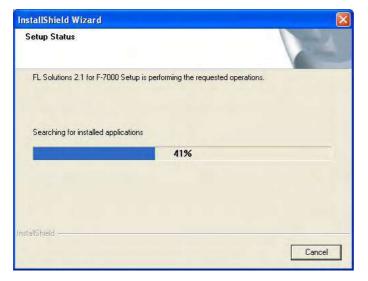


Fig. 1-9



Fig. 1-10

- 8 Click Next.
  - A window as shown in Fig.
     1-9 will appear.

- 9 Installation starts.
  - Upon completion of installation, a window as shown in Fig. 1-10 appears.

- 10 For placing the shortcut of this program on the desktop, put the check mark in this check box. If you do not want to place it, remove the check mark.
- 11 Click Finish.
- 12 Take out the CD from the CD-ROM drive.

#### 1.2.3 Reinstallation of this Program

Before reinstallation, the existing program needs to be deleted. Carry out the following procedure for deleting the program.

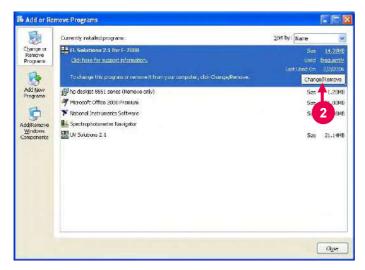


Fig. 1-11

- Select [Start]-[Control Panel] and double-click the "Add or Remove Programs" icon.
  - A window as shown in Fig. 1-11 will open.
- 2 Among the currently installed programs, select "FL Solutions for F-7000" and click **Change/Remove**. After removal, click **Finish**. Then, install the FL Solutions 2.1 program.

#### 1.2.4 Setup of USB Driver

After system setup for FL Solutions, the USB driver needs to be set up for communication with the Model F-7000.



Fig. 1-12



Fig. 1-13

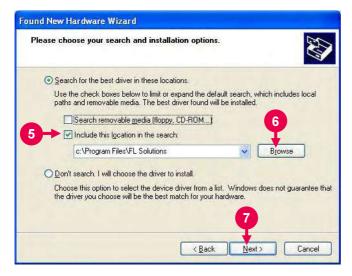


Fig. 1-14

- 1 Connect a USB communication cable and turn on the power switch of F-7000.
- 2 "Found New Hardware" appears at the bottom right of screen, and then a wizard begins.
- 3 Select 'Install from a list or specific location (Advanced).'
- 4 Click Next.

- 5 Put check mark at [Include this location in the search].
- 6 Click Browse and specify the directory in which FL Solutions is set up (usually C:\Program Files\FL Solutions).
- 7 Click Next.



A window for indicating the installing status will appear.
Wait until the installation is completed. When the wizard end window appears, click
Finish.

Fig. 1-15

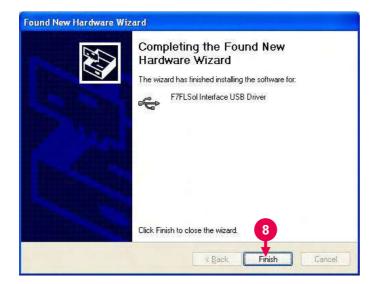


Fig. 1-16

# 1.3 Startup of FL Solutions Program

To start up the FL Solutions 2.1 program, carry out the following procedure.



Fig. 1-17

- 1 Make sure the standardequipped communication cable (USB cable) is connected between the fluorescence spectrophotometer and the personal computer (PC).
- 2 Turn on the PC power supply and wait until Windows XP starts.
- After completion of PC start, turn on power supply to the spectrophotometer main unit.

NOTE: If power supply to the spectrophotometer main unit is turned on before PC start, USB connection cannot be recognized, so communication will be disabled. If this occurs, you should restart the PC with the spectrophotometer power supply turned off, and then turn on that power supply.

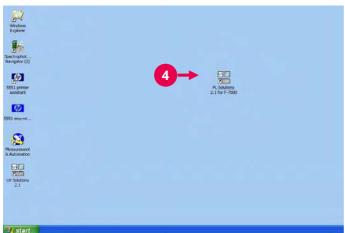
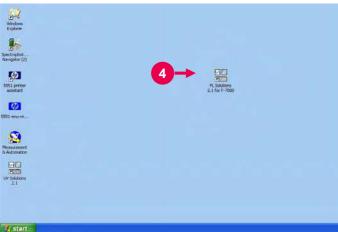


Fig. 1-18



(2) The other method begins with clicking the start button at the bottom left corner of desktop. Next, point to All Programs and Hitachi FL Spectrophotometer, and then select FL Solutions 2.1 for F-7000.

The FL Solutions 2.1 program can be started by either method

Use a desired method.

(1) Usually, this program is

started by double-clicking the "FL Solutions 2.1 for F-7000" icon on the desktop.

below.

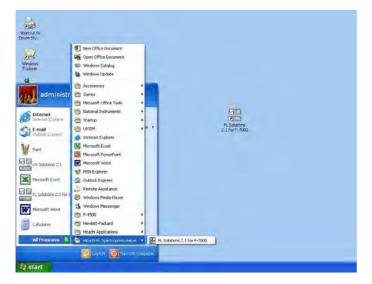


Fig. 1-19

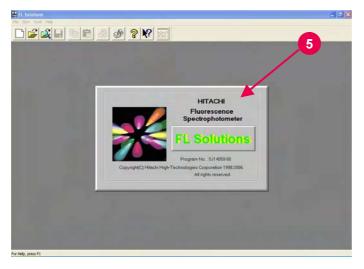


Fig. 1-20

5 The computer now recognizes the fluorescence spectrophotometer connected to it and displays a window as shown in Fig. 1-20. Then, the FL Solutions 2.1 program is launched automatically.

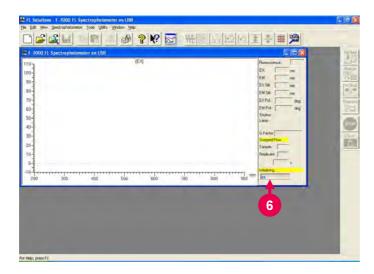


Fig. 1-21

NOTE: If the spectrophotometer is not connected with the communication port or its power is not turned on, the monitor window ("F-7000 Fluorescence Spectrophotometer on USB" indicated on its title bar) is not presented. In this status, functions for other than measurement are usable.

For measurement, first confirm connection of the cable, and then turn on power supply of the spectrophotometer. Next, select the Instrument command from the Tools menu. When selecting and connecting the

6 A gauge of initialization is displayed as shown in Fig. 1-21, so its progress can be checked visually.

In execution of initialization, selfdiagnosis is performed on the following items.

- ROM
- RAM
- Wavelength drive system
- · Xenon lamp turn-on
- Slit mechanism
- Chopper
- Contents of wavelength and slit calibration values

If an abnormality is found in the selfdiagnosis, the relevant error message will be displayed (Fig. 1-22). In this case, a corrective measure should be taken, referring to Appendix

If necessary, notify our local service office.



Fig. 1-21 is presented.

spectrophotometer, the window shown in

Fig. 1-22

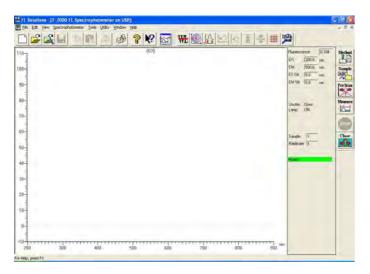


Fig. 1-23

On completion of initialization, the window shown in Fig. 1-23 is displayed.

When "Ready" appears, measurement can be started.

**NOTE**: A sample should be set after "Ready" appears.

If the lid of sample compartment is opened or closed in the "Initializing" or "Parameter" status, zero point may not be set exactly. In this case, perform zero-point adjustment again.

# 1.4 Screen Composition

# 1.4.1 Basics of Window Operation

This manual contains multiple methods for executing a function. For implementing each method, explanation is given using the naming described below.

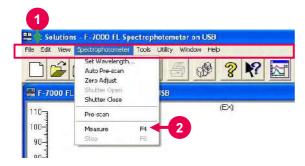


Fig. 1-24 Menu Bar and Command (step ①)



Fig. 1-25 Example Help Display for Measure Command (step ②)

#### 1 Menu bar

 Menu bar is a bar on which menus such as "File" and "Edit" are arranged.

#### 2 Command

- Commands are contained in each menu.
   In the case shown in Fig. 1-24, an expression of "Measure command of Spectrophotometer menu" is used.
- A menu or command is selectable by 2 methods.
  - Move the mouse pointer to a menu and click it.
  - Key in the alphabetic letter after a menu name with the Alt key held down. Then, key in the alphabetic letter after a command name.
- Explanation of a command function can be displayed in the following procedure.
  - ① Click the (Help) button.

    The mouse pointer is now turned into ?.
  - ② Move the mouse pointer to a menu and click the left mouse button. (See Fig. 1-25.)



Fig. 1-26 Example Help Display for Measure Command (step ③)



Fig. 1-27 Example Help Display for Measure Command (step 4)



Fig. 1-28 Toolbar

- When the menu is displayed, move the mouse pointer to the target command and click the left mouse button. (See Fig. 1-26.)
- The help message for the selected command is now displayed. (See Fig. 1-27.)

# 3 Toolbar

Toolbar is a row of tool buttons such as

# 4 Tool button

- Tool buttons are provided for executing the functions of the respective commands.
- In using the button, it is expressed as button of Measurement toolbar" or (analytical condition) button."
- For details of each button, refer to 1.4.4.

## 1.4 Screen Composition

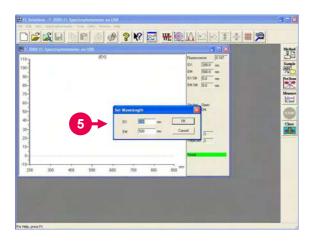


Fig. 1-29 Dialog Box

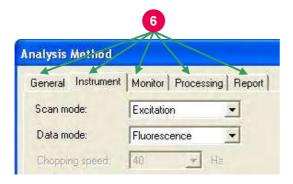


Fig. 1-30 Tab

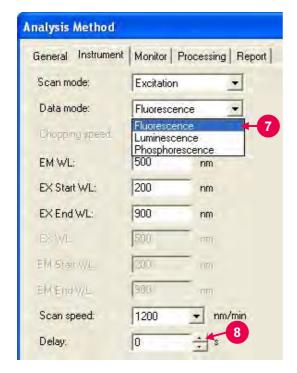


Fig. 1-31 Combo Box and Spin Button

# 5 Dialog box

When you click the button of Measurement toolbar, a small window as shown in Fig. 1-29 is presented. This window is called a "dialog box."

## 6 Tab

- Tabs are used in the Analysis Method and Properties windows.
- Click a tab for changing display.
   Each tab contains setting items.
   The display shown in Fig. 1-30 is expressed as "Instrument tab."

## 7 Combo box

Selection is allowed among the predetermined conditions.
 Click for selecting the desired condition.

## 8 Spin button

 This button is used for inputting a numeral. Click or for setting a numeral. However, a value below decimal point cannot be set. Such a value must be input with the keyboard.

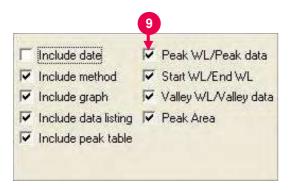


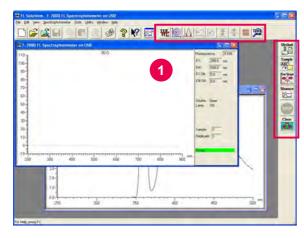
Fig. 1-32 Check Box

# 9 Check box

- For turning on a check box, put a check mark in it by clicking.
- The check box is then set in the status.
- For turning off the check box, click it again to remove the check mark.

# 1.4.2 Kinds of Windows

The FL Solutions 2.1 program has two kinds of windows: monitor window and data processing window.



- Monitor window Set the monitor window active for measurement.
  - The toolbar is now turned into the one for this window.

Fig. 1-33 Monitor Window

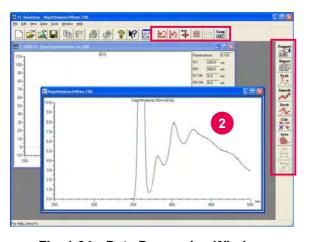


Fig. 1-34 Data Processing Window

- 2 Data processing window Set the data processing window active for data processing.
  - The toolbar is now turned into the one for this window.

# 1.4.3 Data Processing Window

The items displayable in each measurement mode are shown below.

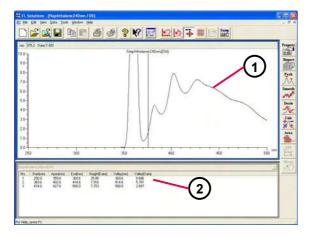


Fig. 1-35 Wavelength Scan

# 1 Wavelength scan

- A spectrum alone can be displayed or both spectrum (1) and peak table (2) can be displayed as shown in Fig. 1-35.
- Display of the peak table can be turned ON/OFF by the "Peak Table" command in the View menu.

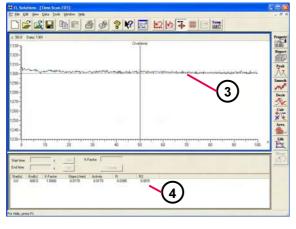


Fig. 1-36 Time Scan (kinetics result table)

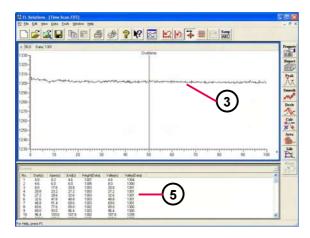


Fig. 1-37 Time Scan (peak table)

## 2 Time scan

 A spectrum alone can be displayed or both spectrum (③) and kinetics result table (④) can be displayed as shown in Fig. 1-36.

- Both spectrum (③) and peak table (⑤) can be displayed as shown in Fig. 1-37.
- Display of the kinetics result table or peak table can be turned ON/OFF by the "Kinetics" or "Peak Table" command in the View menu.

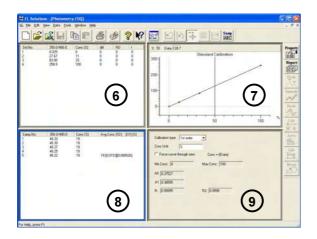


Fig. 1-38

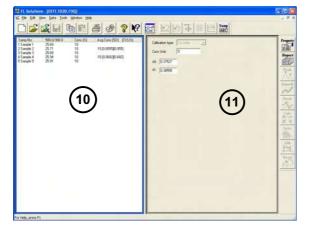


Fig. 1-39

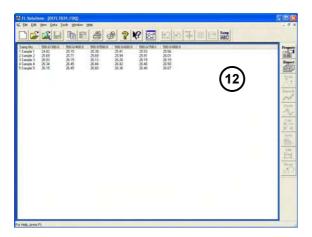


Fig. 1-40

# 3 Photometry

 On start from measurement for a calibration curve, a window as shown in Fig. 1-38 will appear.

Standard data (⑥)
Calibration curve (⑦)
Sample data (⑧)
Calibration curve data (⑨)

 On sample measurement with a calibration curve generated through factor input, a window as shown in Fig. 1-39 will appear.

Sample data (10)
Calibration curve factor (11)

 On measurement of the photometric value at each wavelength in multi-wavelength mode, the sample data is displayed as listed in Fig. 1-40.

Sample data (12)

# 1.4.4 Explanation of Tool Buttons

The FL Solutions 2.1 program provides five kinds of toolbars as shown below. You can also select a command by pulling down each menu from the menu bar. But for quick and easy operation, it is advisable to use these tool buttons.

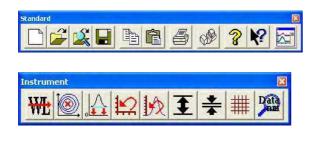








Fig. 1-41 Toolbars

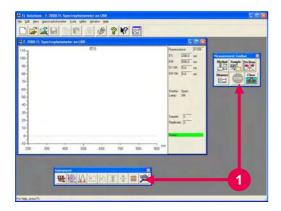


Fig. 1-42 An Example of Toolbar Arrangement

Standard toolbar

Has buttons related to printing and file handling.

Instrument toolbar (displayed in monitor window)

Has buttons related to spectrophotometer control such as wavelength drive and automatic prescanning.

Measurement toolbar (displayed in monitor window)

Has buttons related to measurement such as analysis method setting and sample name input.

Spectrum toolbar (displayed in data processing window)

Has buttons related to spectral display such as auto scaling, ruled line indication and spectrum tracing.

Processing toolbar (displayed in data processing window)

Has buttons related to data processing such as smoothing and differentiation.

These five toolbars are independent and each of them can be located at a desired position as shown in Fig. 1-42 (monitor window).

# 1.4 Screen Composition

The following tables present the functional descriptions of the buttons on each toolbar. The function keys on the keyboard can also be utilized for the same purpose. For the functional assignment to each function key, refer to 1.5.

**Table 1-1 Standard Toolbar** 

Button	Function	Button	Function	
	Creates a new analysis method/sample table.		Opens the print window.	
<b>=</b>	Opens data files.		Indicates the present status of the connected spectrophotometer.	
	Opens a data file in the print preview mode.	<b>?</b>	Indicates the information (version number, etc.) on the FL Solutions program.	
	Saves the current file.	<b>N?</b>	Calls up the help window.	
	Stores a selected character string or graphic item into the clipboard.	<b>₹</b>	Brings the monitor window to the forefront.	
	Pastes the contents of the clipboard.			

**Table 1-2 Measurement Toolbar** 

Button	Function	Button	Function	
Method	Opens the Analysis Method window.	Measure	Starts measurement.	
Sample ABC	Displays the same name/comment entry window.		Stops measurement.	
Sample	Displays the sample table.	Close	Closes the shutter.	
STD 123	For manual input of standard sample values.	Орен	Opens the shutter.	
Pre Scan	Executes pre-scanning.			

Table 1-3 Spectrum Toolbar

Button	Function	Button	Function	
	Restores the scale to its original condition.		For display in a contour map.	
#	Indicates ruled lines.		For display from the front.	
	Indicates a bird's-eye view.		For display from the left.	
	For display from the rear.	1/2	Automatically sets the scale.	
	For display from the right side.	Sанф (ABC)	Displays a dialog box to allow change of sample name/comment.	
	Selects a spectrum to be displayed.	Q	Sets a mode for expansion of contour scale. For this expansion, click this icon and drag the area to be enlarged with the mouse.	
4 4	Indicates the trace cursor.			

Table 1-4 Instrument Toolbar

Button	Function	Button	Function	
₩Ŀ	Performs wavelength drive.		Restores the scale to its original condition.	
	Performs automatic pre-scanning. This is a function for automatic search of the optimum excitation and emission wavelengths for a sample.	₩	Adopts auto scale for vertical- axis display so that you can view the entire spectral graph.	
	Conducts zero-point adjustment. Zero point with the shutter closed is adjustable. Effective for higher-accuracy measurement.	#	Indicates ruled lines.	
1	Enlarges the vertical axis double.	Data	Magnifies the present data and status.	
*	Reduces the vertical axis to half.			

Table 1-5 Processing Toolbar

Button	Function	Button	Function	
Property	Opens the Spectrum Properties window.	Calc	Performs fundamental arithmetic calculations between spectra.	
Report	Outputs a report of data file.	Life	Calculates the life of phosphorescence.	
Peak	Performs peak detection.	Smooth	Performs smoothing.	
Deriv	Performs differentiation.	Reset	Resets data.	
Peak	Performs peak detection in 3- dimensional measured data.	Calc	Performs fundamental arithmetic calculations of 3-dimensional spectra.	
Area	Performs area calculation.			

# 1.5 Functions of Function Keys

The function keys on the keyboard provides the following functions. Each function can be executed directly.

Key No.	Wavelength Scan	Time Scan	Photometry 3-D Scan		3-D Time Scan	
[F4]	Measurement	Measurement	Sample measurement	Measurement	Measurement	
[F5]			Blank measurement			
[F6]	Stop	Stop	Stop	Stop	Stop	
[F7]			Interrupting measurement			
[F8]			Re- measurement			
[F9]			End			

# 1.6 Explanation of Monitor Window

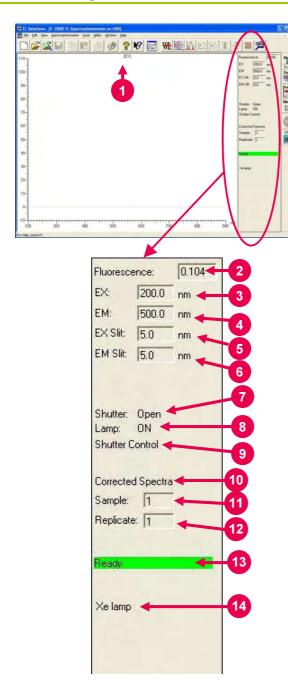


Fig. 1-43

- 1 Sample name indication
  A sample name is indicated. For its input,
  use the sample table or sample
  name/comment input window.
- 2 Current photometric value indication The current photometric value is indicated.
- 3 EX The current excitation wavelength is indicated.
- 4 EM The current emission wavelength is indicated.
- 5 EX slit The current excitation slit width is indicated.
- 6 EM slit
  The current emission slit width is indicated.
- 7 Shutter
  Open or closed state of the shutter is indicated.
- 8 Lamp ON/OFF state of the light source lamp is indicated.
- 9 Shutter control The active state of shutter control is indicated.
- 10 Corrected spectra Setting of spectrum correction is indicated.
- 11 Sample

  The number assigned to the sample under measurement is indicated.
- 12 Replicate count
  A count in repeated measurement is indicated.

- 13 Message indication
  A status message such as "Ready,"
  "Scanning" or "Parameter" is presented.
  In the "Ready" status, the
  (measurement) button can be clicked.
- 14 Error message indication
  An error message "Xe lamp" is presented.
  If any error message appears, take a
  proper countermeasure, referring to 4.7 of
  the maintenance edition.

# 1.7 Input of Spectrophotometer Serial No.

The serial No. of the presently connected F-7000 fluorescence spectrophotometer can be displayed for its confirmation.

Select the Instrument command from the Tools menu or click the button. The dialog box shown in Fig. 1-44 opens. The serial No. of the connected F-7000 fluorescence spectrophotometer can be checked here.

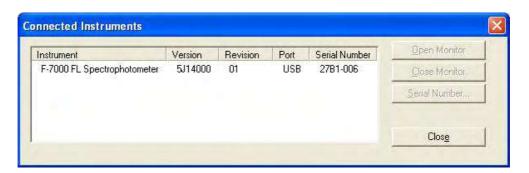


Fig. 1-44

# 1.8 Window Setting

The information to be displayed upon starting the FL Solutions 2.1 program or opening the data processing window can be selected here by excluding unnecessary one from display.

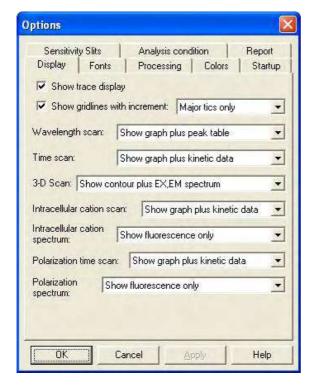


Fig. 1-45 Options Setting Window

- 1 Select Options command from the Tools menu.
- The Options setting window is now displayed. In this window, the following settings can be made. After your setting, click **OK**.
  - The set functions are validated when starting the monitor window again or opening the data processing window.

The Options setting window has the following tabs.

Display tab

Fonts tab

Processing tab

Colors tab

Startup tab

Sensitivity Slits tab

Analysis condition tab

Report tab

# 1.8.1 Display Tab

Items to be displayed when opening a saved data file or data immediately after measurement can be set here. Those items can also be set by tool buttons or View menu after measurement. For the settable range of each parameter, refer to Appendix I.

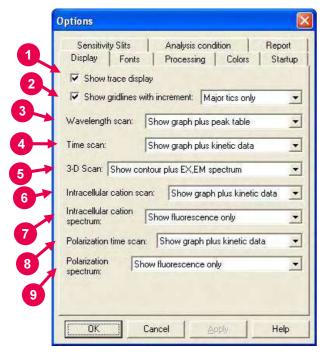


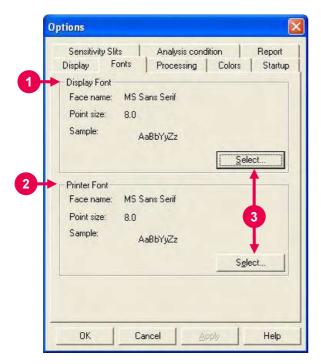
Fig. 1-46 Display Tab

- 1 Show trace display
  Put a check mark in this box, and the trace
  cursor appears to permit spectrum tracing
  when the data processing window is open.
- 2 Show gridlines with increment
  When a check mark is put in this box, the
  contents of monitor and data processing
  windows will be displayed with ruled lines.
  The width of ruled line is selectable.
- Wavelength scan
  Items to be displayed on the data
  processing window open in the wavelength
  scan mode can be set.
- 4 Time scan
  Items to be displayed on the data
  processing window open in the time scan
  mode can be set.
- 5 3-D Scan Items to be displayed on the data processing window open in the 3-D scan mode can be set.
- 6 Intracellular cation scan
  Items to be displayed on the data
  processing window open in the intercellular
  cation scan mode can be set.
- 7 Intracellular cation spectrum
  Items to be displayed on the data
  processing window open in the intercellular
  cation spectrum mode can be set.

- 8 Polarization time scan
  Items to be displayed on the data
  processing window open in the polarization
  time scan mode can be set.
- 9 Polarization spectrum Items to be displayed on the data processing window open in the polarization spectrum mode can be set.

# 1.8.2 Fonts Tab

Display font, character size, etc. can be set. Printer font, character size, etc. are also settable.



Display Font Font, character size, etc. in the spectrum area under display can be set.

- Printer Font Printer font is settable.
- 3 Clicking Select presents a window as shown in Fig. 1-48.

Fig. 1-47 Fonts Tab

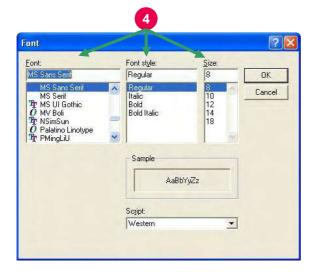


Fig. 1-48 Font Specification

4 Set the desired font, style and size.

# 1.8.3 Processing Tab

Initial parameters of data processing can be set. For the settable range of each parameter, refer to Appendix I.

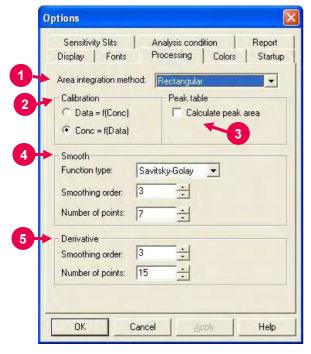


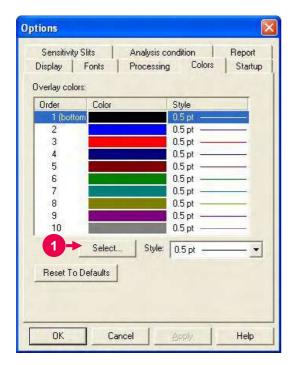
Fig. 1-49 Processing Tab

- Area integration method
   An area integration method can be selected.
- Calibration

  A calibration curve formula to be used for the Photometry mode can be set.
- 3 Peak table/Calculate peak area
  By means of a check mark, inclusion
  of the area calculation item in the
  peak table can be selected or avoided.
- 4 Smooth
   The initial values of smoothing
   parameters can be set. However,
   this setting will not be reflected on the
   parameters of Analysis Method
   window.
- Derivative
  With regard to derivative operation,
  the initial parameters of smoothing in
  an analysis method or to be carried
  out after measurement can be set.

# 1.8.4 Colors Tab

The color of the spectrum to be overlaid is settable here. Color 1 (bottom) is the color of the initial display.



1 Click Select....

 A window as shown in Fig. 1-51 is presented.

Fig. 1-50 Colors Tab

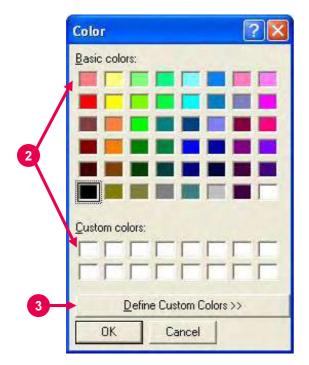


Fig. 1-51 Color Setting

- 2 Select a basic color or custom color.
- 3 If there is no basic color you want, click Define Custom Colors.
  - A window as shown in Fig. 1-52 is presented.

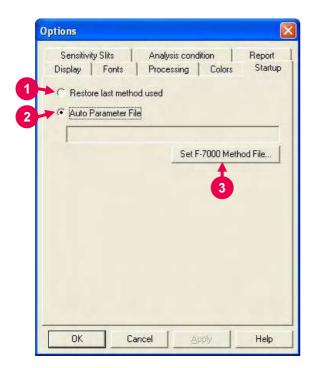


Fig. 1-52 Addition of Color

- 4 Select hue, saturation, luminance, etc. and click **Add to Custom Colors**.
- 5 Your color is then added to the custom colors. So select it and click **OK**.

# 1.8.5 Startup Tab

Analysis method to be taken upon start of the FL Solutions 2.1 program is settable here.



1 Restore last method used The method used last will be brought in.

- 2 Auto Parameter File The specified method will be brought in.
- **3** For specifying a method, click this button.
  - A window as shown in Fig. 1-54 is then presented.

Fig. 1-53 Startup Tab

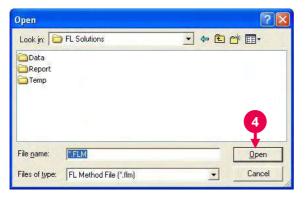


Fig. 1-54

4 Specify a method file and click **Open**. The specified method file appears in Fig. 1-53.

# 1.8.6 Sensitivity Slits Tab

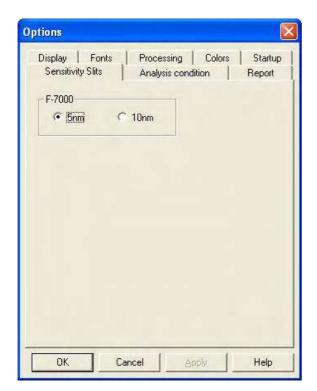


Fig. 1-55 Sensitivity Slits Tab

# Sensitivity Slits

A slit width is settable for sensitivity check.

For sensitivity check, select the Sensitivity Check command from the Utility menu.

# 1.8.7 Analysis Condition Tab

For the settable range of each parameter, refer to Appendix I.

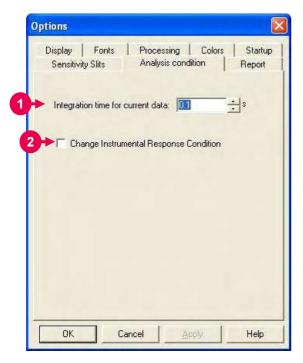


Fig. 1-56 Analysis Condition Tab

- 1 Integration time for current data An integration time for reading the current data by the Read Data command of Tools menu can be set.
- Change Instrumental Response Condition When this check box is turned on, the excitation slit, emission slit and photomultiplier voltage can be set on the instrument function measurement window

This window appears when clicking the Instrument Response command of Utility menu.

# 1.8.8 Report Tab

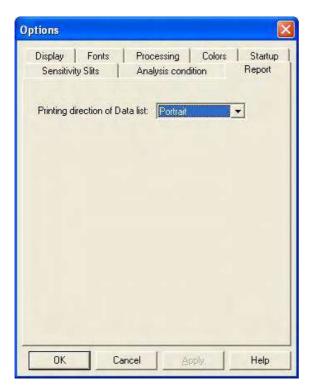


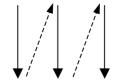
Fig. 1-57 Report Tab

**Data List Printing Direction** 

The direction of data list printing (with constant line spacing) can be set.

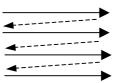
In case of vertical printing:

Wavelength or time and photometric values will be printed vertically in 3 rows.



In case of horizontal printing:

Wavelength or time and photometric values will be printed horizontally in 3 rows. That is, 3 data will be printed on each line.



# 1.9 Saving a Data File

Data can or cannot be saved depending on setting. So perform setting, referring to the table below. Setting in any of patterns ① to ⑤ is recommended.

Setting		"Use sample table in analysis method"					
		ON		OFF			
Sample name/c dialog	omment input	_		ON		OFF	
"Save file automatically after data acquisition"							
Analysis Method - Monitor tab		ON	OFF	ON	OFF	ON	OFF
<ul> <li>"Open data processing window after data acquisition"</li> </ul>							
Result	Display	0	×	0	×	0	×
	Saving	Automatic	Automatic	Automatic	Automatic	Manual	Impossible
Pattern		1	2	3	4	5	6

Read  $\bigcirc$  and  $\times$  symbols in the Result - Display row as follows.

O: Icons or data processing window is open immediately after measurement.

 $\times$  : The data processing window is not open immediately after measurement.

# 1.10 Photometer Menu Commands

The Spectrophotometer menu contains the following commands.

- · Set Wavelength
- Auto Pre-scan
- Zero Adjust
- Shutter Open/Close

## **Set Wavelength**

This command sets the excitation/emission wavelength to a specified wavelength.

Click the Set Wavelength command or button, and the dialog box shown below will come up.

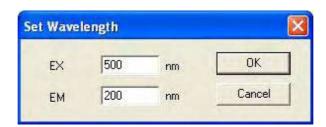


Fig. 1-58

## Zero Adjust

This command automatically closes the shutter and measures a zero point in detection. When the zero point is changed, e.g., after variation of the photomultiplier voltage, automatic zero-point adjustment will be carried out.

**NOTE**: If the lid of sample compartment is opened or closed during zeropoint adjustment, zero point may not be set exactly. In this case, perform zero-point adjustment again.

#### Auto Pre-scan

This command automatically searches for the optimum excitation/emission wavelength for the sample under measurement. Other peaks than the fluorescence peak (such as peaks of excitation scattered light and Raman spectrum) are ignored. On completion of automatic pre-scanning, the optimum excitation/emission wavelength is indicated on the monitor window (in the current wavelength indication field detailed in 1.6).

#### **Shutter Open/Close**

This command is available for opening/closing the excitation beam shutter. The current shutter state is indicated on the right side of the monitor window (③ in Fig. 1-43).

# 1.11 Utility Menu Commands

The Utility menu contains the following command.

- Instrument Response
- · Analog Output Gain
- EX Power Monitor
- · Sensitivity Check
- · Lamp Usage
- Lamp Off

# **Instrument Response**

This command is used for spectral correction. The spectral correction accessory kit (P/N 251-0126) needs to be prepared.

## **Analog Output Gain**

Use this command for setting a data value corresponding to 1 V of analog output. For details, refer to 9.3.

#### **EX Power Monitor**

Use this command for checking output of the monitor detector. When the EX Power Monitor is turned on (clicked), monitor value data is displayed regardless of measurement condition.

## **Sensitivity Check**

Use this command for checking sensitivity. You can check sensitivity/drift with this command. For details, refer to the maintenance edition.

## **Lamp Usage**

Use this command for checking/resetting the usage period of the Xenon lamp. A cumulative turn-on time of the Xenon lamp is indicated.



Fig. 1-59

When the limit use time is exceeded, the following message appears.



Fig. 1-60

After lamp replacement, reset this indication. For details, refer to the maintenance edition.

**Note**: If the Model F-7000 power supply is turned on without starting up the PC, indication of the Xenon lamp turn-on time may not be updated correctly.

# Lamp Off

Use this command for turning off the Xenon lamp.

For turning on the Xenon lamp again, restarting the spectrophotometer is necessary.

# 1.12 Termination of FL Solutions Program

After completion of measurement, shut down the instrument in the following procedure.

1 From the File menu, select the Exit command. A window appears as shown below.

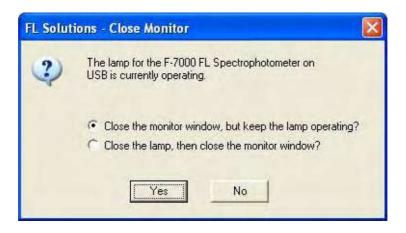


Fig. 1-61

- 2 For termination without turning off the light source lamp, select "Close the monitor window, but keep the lamp operating?"
  - For termination after turning off the light source lamp, select "Close (Turn off) the lamp, then close the monitor window?"
  - Click Yes, and the FL Solutions program will then be terminated.
- 3 Turn off the power switch of the spectrophotometer main unit.
- 4 Click the **start** button of Windows XP, select "Turn Off Computer" and click **OK**.
- Turn off power to the personal computer and the CRT display monitor.(At step 4, the personal computer may be turned off depending on system settings.)
- 6 Turn off power to the printer.

# 2. WAVELENGTH SCAN

# 2.1 Flow of Operation

This section explains the condition setting and operating method for wavelength scan.

Measurement proceeds along the following flow.

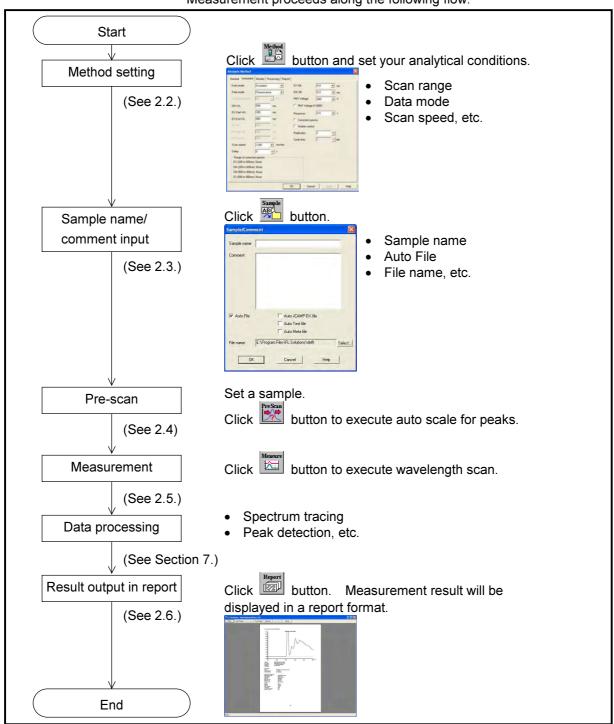


Fig. 2-1 Operational Flow (wavelength scan)

# **Creating an Analysis Method**

Set the analytical conditions (method) prior to measurement.



Select the Method command from the Edit menu or click the button. A window shown in Fig. 2-2 will appear.

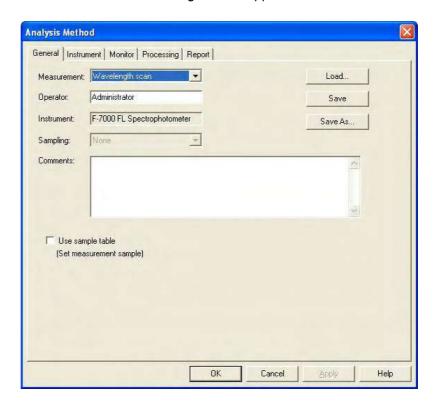


Fig. 2-2 Analysis Method

The following tabs are provided on the Analysis Method window for the wavelength scan mode. These tabs are detailed on the subsequent pages.

- General
- Instrument
- Monitor
- Processing
- Report

## 2.2.1 General Tab

The parameters shown in Fig. 2-3 are provided at the General tab. For the settable range of each parameter, refer to Appendix G.

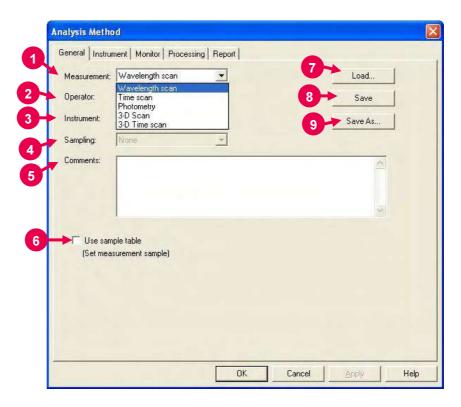


Fig. 2-3 General Tab

# 1 Measurement (mode)

A measurement mode is selectable. Here, select "Wavelength scan."

For Time scan, Photometry, 3-D Scan and 3-D Time scan, refer to Sections 3, 4, 5 and 6, respectively.

# 2 Operator

Enter an operator name.

#### 3 Instrument

The name of the connected instrument is indicated.

## 4 Sampling

- None

Indicates that samples are to be exchanged manually.

## 5 Comments

Enter a description or notes on measuring conditions.

6 Use sample table (Set measurement sample)
Apply a check mark here when a sample table is used.

# 2.2 Creating an Analysis Method

## 7 Load

Click this button to read out saved analytical conditions.

Upon clicking, the file opening window appears. So, select an analytical condition file.

## 8 Save

When you click this button after setting the parameters, the analytical conditions can be saved by overwriting. In case of a new analysis method, clicking this button displays the "Save As" window. You must enter a file name for saving in this window.

## 9 Save As

Click this button for renaming the set conditions for saving. The "Save As" window appears, so enter a file name. **2.2.2 Instrument Tab** When you click the Instrument tab, the window in Fig. 2-4 appears. For the settable range of each parameter, refer to Appendix G.

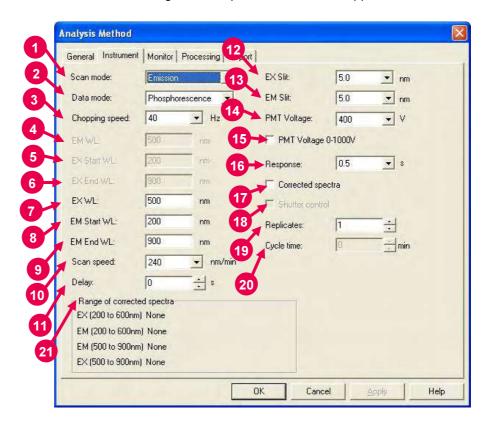


Fig. 2-4 Instrument Tab

## 1 Scan mode

Select a scan mode here. In case of Synchronous, the abscissa of the graph stands for excitation wavelength.

The unit indication of the abscissa becomes 'EX/EM(nm).'

## 2 Data mode

Select a data mode. The Luminescence mode is for measuring luminescence in the biological/chemical field.

In this mode, only the sample side signal is acquired without performing ratio photometry. The Phosphorescence mode is for acquiring only phosphorescence after extinction of excitation beam while turning a chopper on the excitation side.

#### 3 Chopping speed

Select a chopping speed. Setting is allowed when Phosphorescence is selected for Data mode.

The Phosphorescence mode is for acquiring only phosphorescence after extinction of excitation beam while turning a chopper on the excitation side. An increase in chopping speed makes the chopper turns faster.

#### 4 EM WL

For the Excitation scan mode, enter the fixed wavelength on the emission side . For the Synchronous scan mode, enter the start wavelength on the emission side.

#### 5 EX Start WL

This is the start wavelength on the excitation side in wavelength scan. Enter the shorter wavelength side of measuring wavelength range.

#### 6 EX End WL

This is the end wavelength on the excitation side in wavelength scan. Enter the longer wavelength side of measuring wavelength range.

#### 7 EX WL

For the Emission scan mode, enter the fixed wavelength on the excitation side.

#### 8 EM Start WL

This is the start wavelength on the emission side in wavelength scan. Enter the shorter wavelength side of measuring wavelength range.

#### 9 EM End WL

This is the end wavelength on the emission side in wavelength scan. Enter the longer wavelength side of measuring wavelength range.

#### 10 Scan speed

Set a wavelength scan speed.

#### 11 Delay

After pressing the Measure button, measurement is started following the delay time set here. It is used for temperature stabilization, etc. In repeat measurement, it is the time until the start of the first measurement. The delay time is invalid for the second and subsequent measurements.

#### 12 EX Slit

Select a slit width on the excitation side.

#### 13 EM Slit

Select a slit width on the emission side.

#### 14 PMT Voltage

A function for controlling the voltage of the photomultiplier detector (photomultiplier tube). As a reference, changing from 400 V to 700 V increases the data value about two digits and changing from 700 V to 950 V increases it about 1 digit.

When a check mark is put for PMT Voltage 0-1000V, the photomultiplier voltage is settable within a range from 0 to 1000 V.

#### 15 PMT Voltage 0-1000V

When a check mark is put in this box, the photomultiplier voltage is settable within a range from 0 to 1000 V.

#### 16 Response

When "Auto" is selected, response will be set automatically depending on scan speed. Select "Auto" usually.

#### 17 Corrected spectra

A function for determining the spectrum inherent to a sample by correcting the photometer wavelength characteristic using the saved instrument parameters, following measurement with the instrument parameters for photometer control. If there are no instrument parameters for the wavelength range to be measured, only the wavelength range that allows correction is scanned. And if there is no wavelength area measurable, an error will occur. The Instrument Response command is provided in the Utility menu for operating this function.

When this setting is at ON, "Corrected spectra" appears on the monitor window. (See Fig. 1-43 in 1.6.)

#### 18 Shutter control

The shutter can automatically be closed in other than measurement for slowing down sample deterioration due to the energy of excitation beam and opened when measurement starts.

When you put a check mark at the head, the shutter will close, and open at start of measurement. The shutter will close again when measuring wavelength begins returning to the start wavelength

after measurement in the wavelength scan mode.

#### 2.2 Creating an Analysis Method

19 Replicates Set the number of repeat measurements. An average spectrum can also be determined in repeat measurement. Setting is available on the Processing tab (shown in Fig. 2-6).

- 20 Cycle time
  Set a repetition interval.
- 21 Range of corrected spectra
  Spectrum correction range is indicated.

#### 2.2.3 Monitor Tab

When selecting the Monitor tab, the window in Fig. 2-5 appears. For the settable range of each parameter, refer to Appendix G.

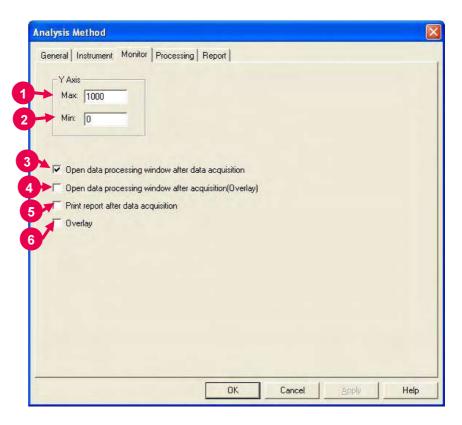


Fig. 2-5 Monitor Tab

Assign the following parameters.

- Y-Axis Max (maximum)
  Enter a maximum value for Y-axis on the monitor window.
- Y-Axis Min (minimum)
  Enter a minimum value for Y-axis on the monitor window.

NOTE: When the remaining memory capacity of PC in operation is very small (system resource is less than 20%), the data processing window will not open even if this item is turned ON. Either close the presently opened data processing window or restart the FL Solutions program.

- 4 Open data processing window after acquisition (Overlay) Select whether or not to conduct data processing with spectra overlaid after sample measurement. This setting is unavailable in repeat measurement.
- Print report after data acquisition
  Select whether or not to print a report after sample measurement.
  When selected (check mark is applied), printing will be made automatically after measurement.
  The items set at the Report tab will be printed. When this item is not selected, data processing is allowed by reading out a file via [File]-[Open].
- 6 Overlay Spectra can be overlaid in the monitor window. This setting cannot be selected for repeat measurement.

**2.2.4 Processing Tab** On selection of the Processing tab, the window in Fig. 2-6 will be displayed.

For the settable range of each parameter, refer to Appendix G.

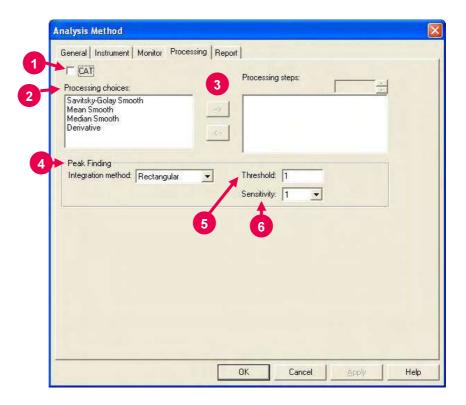


Fig. 2-6 Processing Tab

1 CAT (Computing for Averaging Transient)
Valid when 2 or larger value is entered for Replicates at the Instrument tab.

When a check mark is put at the head, an average spectrum is determined upon carrying out repeat measurement.

This is especially effective for measuring samples with which the emission intensity varies with time.

#### 2 Processing choices

A list of data processing is displayed. Select a data processing item, and click the Right arrow button ( \_\_\_\_\_). Then, the selected method appears in the Processing steps field. For example, select 'Derivative' and click \_\_\_\_\_, then 'Derivative' is displayed in the Processing steps field as shown in Fig. 2-7.

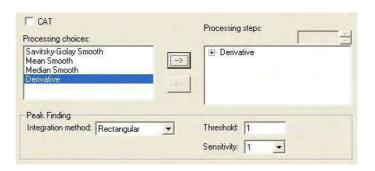


Fig. 2-7

#### 3 Processing steps

The processing sequence is displayed. After measurement, data processing will be carried out in the order set here.

#### How to Delete a Processing Method

Select a processing method to be deleted and click the Left arrow button ( <-- ). Then, the selected method disappears from the Processing steps field.

#### Display and Change of Parameter

When you click [+] in the Processing steps field, a parameter list is displayed. Click a parameter name to be changed, and it becomes changeable. For changing a 'Derivative' parameter, click [+] at the left of 'Derivative' in Fig. 2-7. The present derivative parameters (Derivative order, Smoothing order, Number of points) are displayed. For changing the derivative order, click 'Derivative order," and it is highlighted as shown in Fig. 2-8 to allow your change.

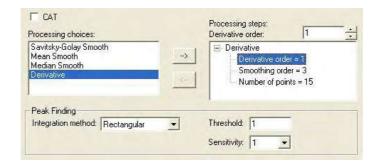
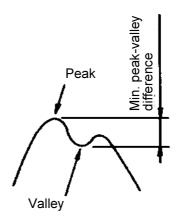


Fig. 2-8

- 4 Peak Finding: Integration method
  Select an integration method for the peaks to be displayed in the
  data processing window.
- Peak Finding: Threshold Enter the minimum photometric value difference (between peak and valley) for definition as a peak.



6 Peak Finding: Sensitivity
Select the number of data points in the horizontal axis direction
(abscissa). For detecting sharp peaks, select a sensitivity value of
"1." Or select "8" for detecting broad peaks.

#### 2.2.5 Report Tab

Select the Report tab, and the window in Fig. 2-9 will appear. For the settable range of each parameter, refer to Appendix G.

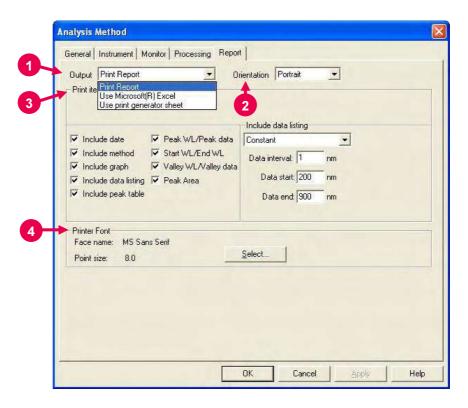


Fig. 2-9 Report Tab

This window is used to select a printout method and items of measurement results.

#### 1 Output

- Print Report
   Data of the specified items will be printed in a report.

   For printing method, refer to 2.6.
- Use Microsoft(R) Excel
   Data of the specified items will be transferred to Microsoft(R)
   Excel. For transferring method, refer to 7.13.
- Use print generator sheet
   Select this method for using the report generator (option).
   For usage, refer to the instruction manual of the report generator.

#### 2 Orientation

Selectable when Print Report is chosen for Output.

Select a printing orientation. Either Portrait or Landscape orientation is selectable. When the Landscape orientation is selected, date and method can be printed.

#### 3 Print items (printable/transferable items)

- Include date
- Include method
- Include graph
- · Include data listing
  - ♦ Constant

Data will be printed at fixed intervals between 2 wavelength points. Make entries for Data interval, Data start and Data end.

♦ Select data

Data at the specified wavelength will be printed. Up to 12 wavelengths can be specified.

When clicking a wavelength value, it allows editing. Select a wavelength within the measurable range set in the Instrument tab.

Include peak table
 Select output items in the peak table.

#### 4 Printer Font

Click the **Select** button for changing the font to be used in the report.

## 2.3 Defining Your Samples

- **2.3.1 Without Using** When "Use sample table" is not selected in Analysis Method, a sample **Sample Table** must be defined in the following procedure before measurement.
  - (1) Click the button. The window in Fig. 2-10 will appear.

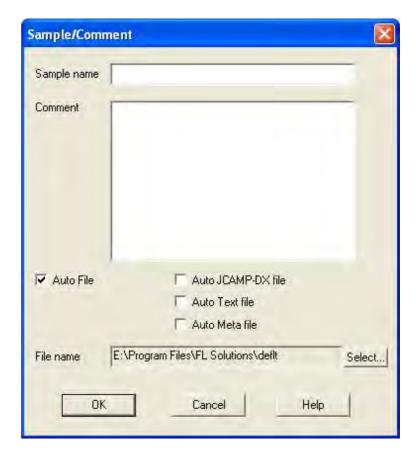


Fig. 2-10

(2) Make entries for Sample name and Comment.Maximum number of enterable characters (half-size)

Sample name: 40 characters

Comment: 255 characters

For automatic saving, put a check mark in the "Auto File" box to specify a file name. First click the **Select** button, and then specify a saving location and enter a file name.

When you put a check mark at the head of "Auto JCAMP-DX file," "Auto Text file" and "Auto Meta file" after turning on "Auto File," wavelength scan data can be saved automatically in the following file formats as soon as this data is automatically saved.

- JCAMP-DX file (\*.dx)
- ASCII text file (\*.txt)
- Graph meta file (\*.wmf)

The data will be saved in the file name entered for "File name."

(3) After completion of setting, click **OK**.

# 2.3.2 With Sample Table

When "Use sample table" is selected in Analysis Method, samples must be defined in the following procedure before measurement.

Open the monitor window and click the button.

When the Sample Table window in Fig. 2-11 appears, enter the sample name and comment for each sample.

1 For creating a sample table newly, make entry for Number of Samples and click the **Update** button. For instance, if there are five samples, enter "5" for Number of Samples and click the **Update** button.



Fig. 2-11 Sample Table

#### 2.3 Defining Your Samples

- 2 For adding a row at the end of sample table, click the **Insert** button.
- 3 To delete an unnecessary row, click its row No. to make it active as shown in Fig. 2-12. Then, click the **Delete** button.



Fig. 2-12

4 Make entries for Sample and Comments of each sample.
The sample names entered here appear on the monitor window.

Maximum number of characters to be entered (half-size)

Sample : 40 characters Comment : 255 characters

Next, you must enter a file name for automatic saving after measurement.

When you click a File Name column, the [...] button appears as shown in Fig. 2-13. Click this button. Upon opening of the window shown in Fig. 2-14, specify a saving location and enter a file name.

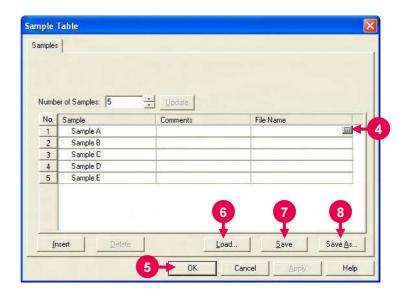


Fig. 2-13

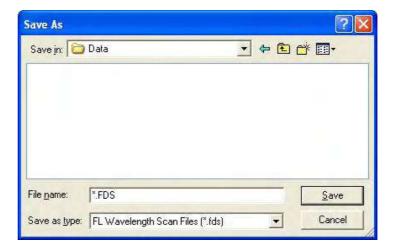


Fig. 2-14

- **5** After entries, check them and then click the **OK** button.
- Load button
   Used for reading out the existing sample table.
   Clicking the Load button displays the file opening dialog.

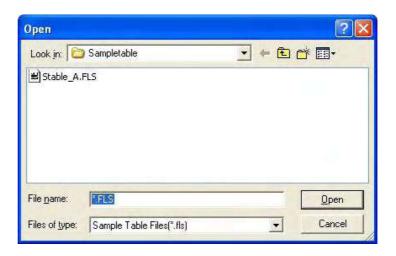


Fig. 2-15 File Opening Dialog

Select a sample table and click the **Open** button. The specified sample table will be read out.

#### 7 Save button

The contents of change can be saved by overwriting. In case of a new generation, the Save As dialog will open.

#### 8 Save As button

Used for saving the sample table with a name newly given. Click this button, and the Save As dialog will open.

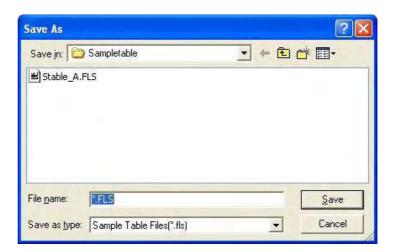


Fig. 2-16 Save As Dialog

Select a saving location and enter a file name. Then, click the **Save** button.

#### 2.4 Prescan

Select the Auto Pre-scan command from the Spectrophotometer menu or click the (pre-scan) button.

This function executes high-speed scan under the set measuring conditions, detects the peak wavelength and moves the system to that wavelength. Also, the upper limit photometric value on the Y-axis scale is automatically set so that the peak-wavelength value becomes approximately 70% of that upper limit value. In this case, the lower limit photometric value on the Y-axis scale is set at 0. Scan rages are as follows.

#### Excitation spectra:

Start wavelength on excitation side to (fixed wavelength on emission side - 20 nm) or (end wavelength on excitation side) whichever shorter

#### Emission spectra:

(Start wavelength on emission side) or (fixed wavelength on excitation side + 20 nm) whichever longer to end wavelength on emission side

#### Synchronous spectra:

Excitation wavelength; Start wavelength on excitation side to end wavelength on excitation side

Emission wavelength; Fixed wavelength on emission side to [fixed wavelength on emission side + (end wavelength on excitation side - start wavelength on excitation side)]

However, when 900 nm is reached, scan is terminated.

## 2.5 Conducting Measurement

# 2.5.1 Without Using Sample Table

- (1) Click the button and make sure a sample has already been defined.
- (2) Set a sample and click (pre-scan) button
- (3) After completion of pre-scan, click the button.

  Measurement starts and a spectrum is displayed.

  For halting the measurement midway, click the (stop) button
- (4) Upon completion of the measurement, the measured data will be saved automatically at the specified saving location using the specified file name when auto saving is selected. If auto saving is not selected, select the "Save As" command from the File menu and save the data. Opening the saved data file will display the data processing window as shown below.

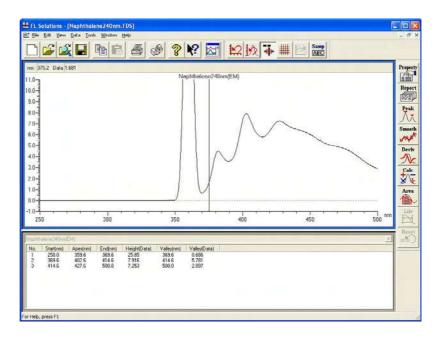


Fig. 2-17 Data Processing Window (wavelength scan)

# 2.5.2 With Sample Table

- (1) Click the button and make sure a sample has already been defined.
- (2) Set a sample and click (pre-scan) button.
- (3) After completion of pre-scan, click the button. The window shown in Fig. 2-18 appears. Click **Yes**.

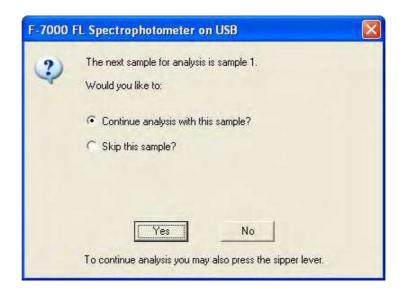


Fig. 2-18

(4) A dialog box opens as shown below.



Fig. 2-19

Click **OK**. Measurement starts.

For halting the measurement midway, click the (stop)

Measure all the samples set in the sample table according to the message guide.

(5) Upon completion of the measurement, the measured data will be saved automatically at the specified saving location using the specified file name. Opening the saved data file will display the data processing window as shown below.

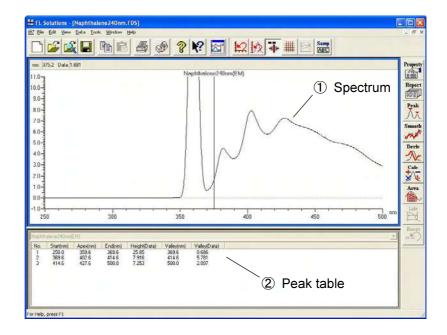


Fig. 2-20 Data Processing Window (wavelength scan)

- Spectrum
   The spectrum of a measured sample is displayed.
- Peak table Peak wavelength, peak height and valley wavelength are listed.

- NOTICE: 1. The data measured under the following conditions will be saved automatically in Program Files\FL Solutions\Data. (Example: C:Program Files\FL Solutions\Data)
  - Measurement was carried out using a sample table with no file name specified in the File Name column.
  - (2) Measurement was carried out with "Auto File" specified and a file name unspecified, avoiding use of a sample table.
  - The data measured under the following conditions will be saved automatically in Program Files\FL Solutions.
     (Example: C:Program Files\FL Solutions)
    - (3) There is not an adequate free area in the specified storage medium (such as floppy disk or MO).
    - (4) Writing is inhibited.

A file automatically saved under such a condition as above is named according to the following rule.

- Rule 1: When a file name is given to analysis method: File name of analysis method + serial No. (001, 002, ...)
- Rule 2: When a file name is not given to analysis method: DEFLT + serial No. (001, 002, ...)

## 2.6 Data Printout

Printout can be provided even when the function for automatic printout after measurement is not used. The following two procedures are available.

(1) Click on the spectrum window, and the circumference turns blue. In this status, select the Print command from the File menu.

Or click the (print) button on the toolbar. The field active on the data processing window (e.g., 1) in Fig. 2-20) will be printed. What is displayed by clicking the Print Preview command in the File menu will be printed.

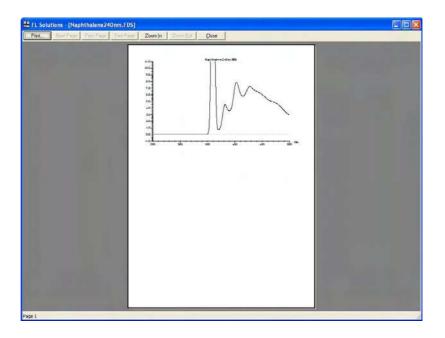


Fig. 2-21

(2) Select the Report command in the Data menu. Or click the button on the toolbar. The Print Preview window will appear as shown below, so click **Print**.

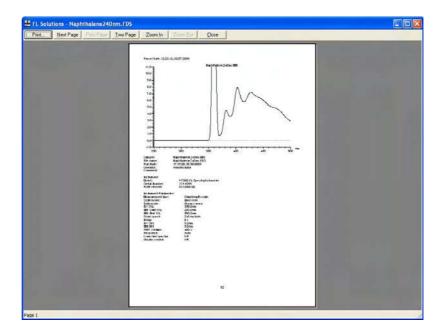


Fig. 2-22

**NOTE**: If characters overflow from the print frame in repeat measurement or in overlaying, then change the printing orientation from vertical (Portrait) to horizontal (Landscape).

Select the items to be printed in the following procedure.

Click the button, and the Spectrum Properties window will appear.

Select the Report tab, and the window in Fig. 2-23 will open.

 $\downarrow$ 

In this window, select desired print items.

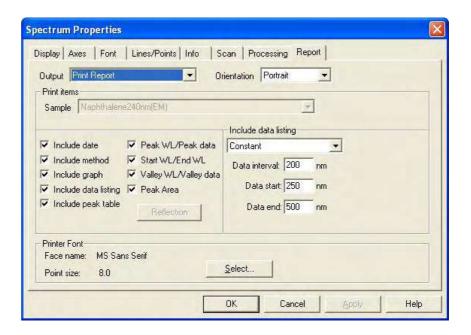


Fig. 2-23

## 3. TIME SCAN

## 3.1 Flow of Operation

This section explains the parameter setting and operating method for time scan. Measurement proceeds along the flow shown below.

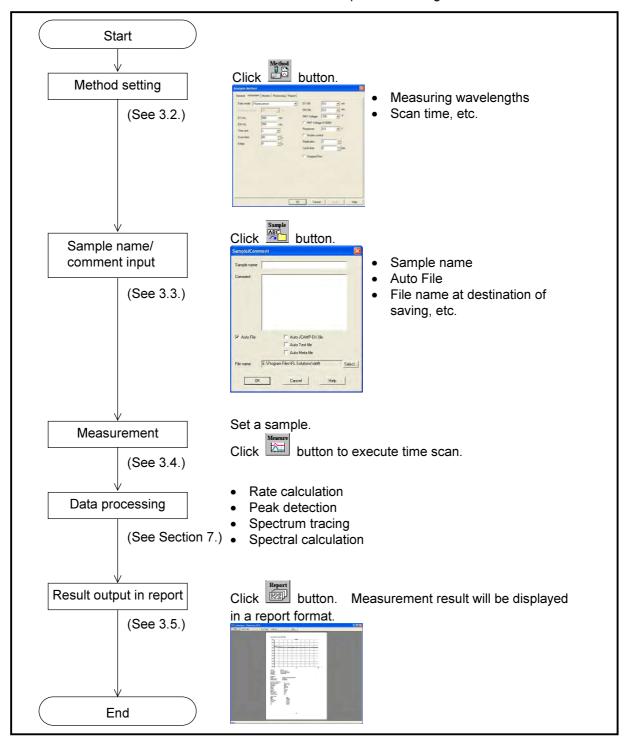


Fig. 3-1 Operational Flow (time scan)

#### **Creating an Analysis Method** 3.2

Set the analytical conditions (method) prior to measurement.



From the Edit menu, select the Method command or click the button. A window shown in Fig. 3-2 will appear.

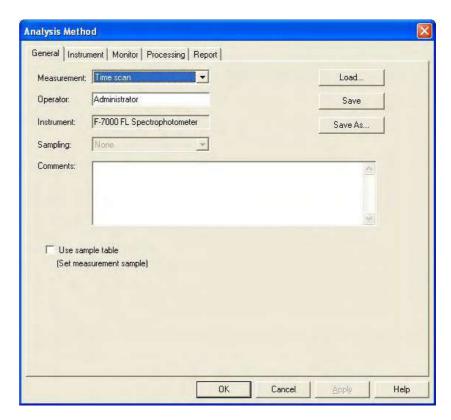


Fig. 3-2 Analysis Method

The following tabs are provided on the Analysis Method window for the time scan mode. These tabs are detailed on the subsequent pages.

- General
- Instrument
- Monitor
- Processing
- Report

#### 3.2.1 General Tab

The parameters shown in Fig. 3-3 are provided at the General tab. For the settable range of each parameter, refer to Appendix G.

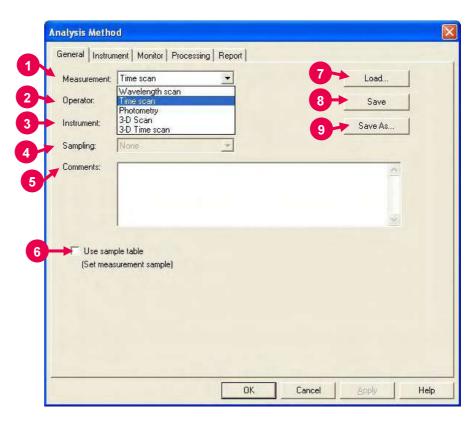


Fig. 3-3 General Tab

#### 1 Measurement (mode)

A measurement mode is selectable. Here, select "Time scan." For Wavelength scan, Photometry, 3-D Scan and 3-D Time scan, refer to Sections 2, 4, 5 and 6, respectively.

OperatorEnter an operator name.

#### 3 Instrument

The name of the connected instrument is indicated.

#### 4 Sampling

- Standard

Indicates that samples are to be exchanged manually.

#### 5 Comments

Enter a description or notes on measuring conditions.

6 Use sample table (Set measurement sample)
Apply a check mark here when a sample table is used.

#### 3.2 Creating an Analysis Method

#### 7 Load

Click this button to read out saved analytical conditions.

Upon clicking, the file opening window appears. So, select an analytical condition file.

#### 8 Save

When you click this button after setting the parameters, the analytical conditions can be saved by overwriting. In case of a new analysis method, clicking this button displays the "Save As" window. You must enter a file name for saving in this window.

#### 9 Save As

Click this button for renaming the set conditions for saving. The "Save As" window appears, so enter a file name.

# **3.2.2 Instrument Tab** This window appears when Time scan is specified for Measurement at the General tab on the Analysis Method window. When you click the Instrument tab, the window in Fig. 3-4 appears.

For the settable range of each parameter, refer to Appendix G.

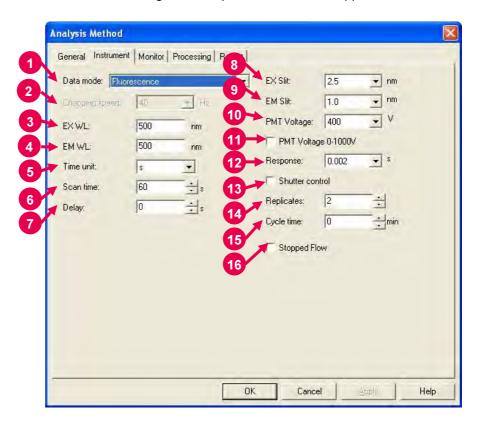


Fig. 3-4 Instrument Tab

#### 1 Data mode

Select a data mode. The Luminescence mode is for measuring luminescence in the biological/chemical field. In this mode, only the sample side signal is acquired without performing ratio photometry. If phosphorescence life is shorter than 20 ms, "Phosphorescence short life" should be selected. In this case, unit time and measuring time are automatically set to ms and 20 ms.

#### 2 Chopping speed

Select a chopping speed. Setting is allowed when Phosphorescence is selected for Data mode.

The Phosphorescence mode is for acquiring only phosphorescence after extinction of excitation beam while turning a chopper on the excitation side. An increase in chopping speed makes the chopper turns faster.

#### 3 EX WL

Enter an excitation wavelength.

#### 4 EM WL

Enter an emission wavelength.

#### 5 Time unit

Set a time unit.

#### 6 Scan time

Set a wavelength scan speed.

For the relationship between scan time and sampling interval, refer to Appendix H.

#### 7 Delay

After pressing the Measure button, measurement is started following the delay time set here. It is used for temperature stabilization, etc. In repeat measurement, it is the time until the start of the first measurement. The delay time is invalid for the second and subsequent measurements.

#### 8 EX Slit

Select a slit width on the excitation side.

#### 9 EM Slit

Select a slit width on the emission side.

#### 10 PMT Voltage

A function for controlling the voltage of the photomultiplier detector (photomultiplier tube). As a reference, changing from 400 V to 700 V increases the data value about two digits and changing from 700 V to 950 V increases it about 1 digit.

When a check mark is put for PMT Voltage 0-1000V, setting is allowed within a range from 0 to 1,000 V.

#### **11** PMT Voltage 0-1000V

When a check mark is put in this box, the photomultiplier voltage is settable within a range from 0 to 1000 V.

#### 12 Response

Set a response time.

#### 13 Shutter control

The shutter can automatically be closed in other than measurement for slowing down sample deterioration due to the energy of excitation beam and opened when measurement starts.

# 14 ReplicatesSet the number of repeat measurements.

**15** Cycle time Set a repetition interval.

#### 16 Stopped Flow

When a check mark is put in this box, the stopped flow mode is set. In this case, when you click the measurement start button, the system waits until an external start signal comes in.

Then, measurement starts.

#### 3.2.3 Monitor Tab

When selecting the Monitor tab, the window in Fig. 3-5 appears. For the settable range of each parameter, refer to Appendix G.

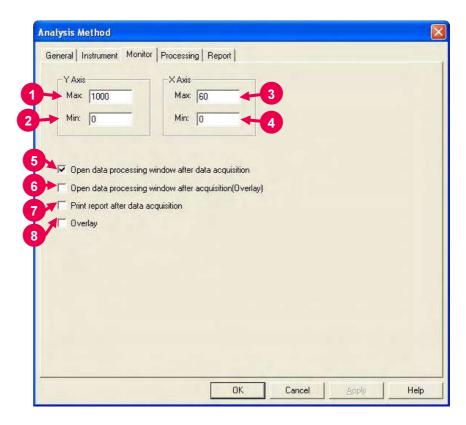


Fig. 3-5 Monitor Tab

Assign the following parameters.

- 1 Y-Axis Max (maximum)
  Enter a maximum value for Y-axis on the monitor window.
- Y-Axis Min (minimum)
  Enter a minimum value for Y-axis on the monitor window.
- 3 X-Axis Max (maximum)
  Enter a maximum value for X-axis on the monitor window.
- 4 X-Axis Min (minimum)
  Enter a minimum value for X-axis on the monitor window.

NOTE: X-Axis Max and Min correspond to a display interval on the monitor window. For example, when Max and Min are 60 and 30, display on the window is made within 30 to 60 s, 60 to 90 s, 90 to 120 s and so on.

Open data processing window after data acquisition
Select whether or not to conduct data processing after sample
measurement. When selected (check mark is applied), an icon
for this function (as shown in

NOTE: When the remaining memory capacity of PC in operation is very small (system resource is less than 20%), the data processing window will not open even if this item is turned ON. Either close the presently opened data processing window or restart the FL Solutions program.

- Open data processing window after acquisition (Overlay) Select whether or not to conduct data processing with spectra overlaid after sample measurement. This setting is unavailable in repeat measurement.
- 7 Print report after data acquisition Select whether or not to print a report after sample measurement. When selected (check mark is applied), printing will be made automatically after measurement. The items set at the Report tab will be printed.
- 8 Overlay Spectra can be overlaid in the monitor window. This setting cannot be selected for repeat measurement.

# 3.2.4 Processing Tab

On selection of the Processing tab, the window in Fig. 3-6 will be displayed.

For the settable range of each parameter, refer to Appendix G.

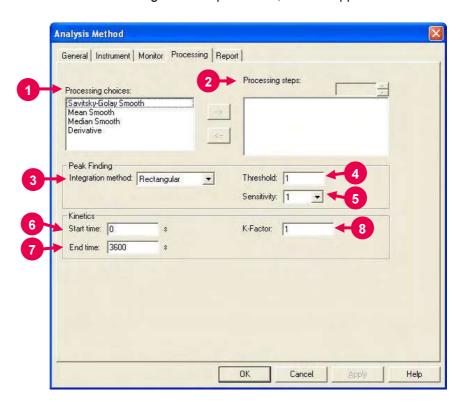


Fig. 3-6 Processing Tab

#### 1 Processing choices

A list of data processing is displayed. Select a data processing item, and click the Right arrow button ( \_--> ). Then, the selected method appears in the Processing steps field. For example, select 'Derivative' and click \_--> , then 'Derivative' is displayed in the Processing steps field as shown in Fig. 3-7.

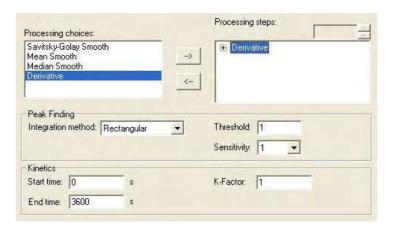


Fig. 3-7

#### 2 Processing steps

The processing sequence is displayed. After measurement, data processing will be carried out in the order set here.

#### How to Delete a Processing Method

Select a processing method to be deleted and click the Left arrow button ( -- ). Then, the selected method disappears from the Processing steps field.

#### Display and Change of Parameter

When you click [+] in the Processing steps field, a parameter list is displayed. Click a parameter name to be changed, and it becomes changeable. For changing a 'Derivative' parameter, click [+] at the left of 'Derivative' in Fig. 3-7. The present derivative parameters (Derivative order, Smoothing order, Number of points) are displayed. For changing the derivative order, click 'Derivative order," and it is highlighted as shown in Fig. 3-8 to allow your change.

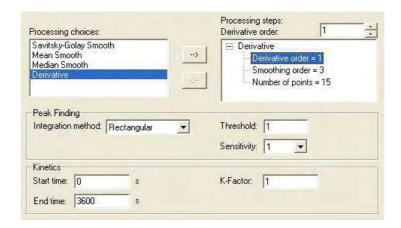


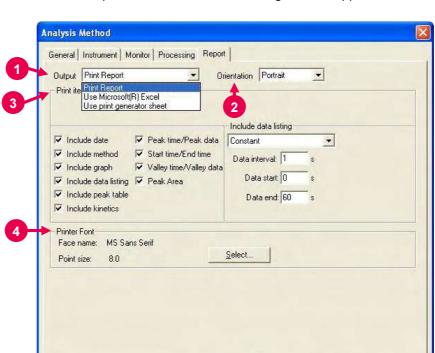
Fig. 3-8

- 3 Peak Finding: Integration method Select an integration method for the peaks to be displayed in the data processing window.
- 4 Peak Finding: Threshold
  Set a detection limit for the photometric value axis of peak and valley.

Peaks or valleys below the set value will not be detected.

#### 3.2 Creating an Analysis Method

- Peak Finding: Sensitivity Select the number of data points in the horizontal axis direction (abscissa). For detecting sharp peaks, select a sensitivity value of "1." Or select "8" for detecting broad peaks.
- 6 Kinetics: Start time Enter a start time for kinetics (rate calculation).
- 7 Kinetics: End time Enter an end time for kinetics.
- 8 Kinetics: K-Factor Enter a K-factor for kinetics.



**3.2.5** Report Tab Select the Report tab, and the window in Fig. 3-9 will appear.

Fig. 3-9 Report Tab

Cancel

Help

This window is used to select a printout method and items of measurement results.

#### 1 Output

- Print Report
   A report will be printed. For printing method, refer to 3.5.
- Use Microsoft(R) Excel
   Data will be transferred to Microsoft(R) Excel. For transferring method, refer to 7.13.
- Use print generator sheet
   Select this method for using the report generator (option).
   For usage, refer to the instruction manual of the report generator.

#### 2 Orientation

Selectable when Print Report is chosen for Output.

Select a printing orientation. Either Portrait or Landscape orientation is selectable. When the Landscape orientation is selected, date and method can be printed.

#### 3 Print items (printable/transferable items)

• Include date

The date and time of analysis can be printed. Printing time will always be printed.

- Include method
- Include graph
- · Include data listing
  - ♦ Constant

Data will be printed at fixed intervals between 2 wavelength points. Make entries for Data interval, Data start and Data end.

♦ Select data

Data at the specified wavelength will be printed. Up to 12 time points can be specified.

When clicking a time value, it allows editing. Select a time within the measurable range set in the Instrument tab.

• Include peak table

Select output items in the peak table.

When a check mark is put in this box, the following output items are selectable.

- ♦ Peak time/Peak data
- ♦ Start time/End time
- ♦ Valley time/Valley data
- ♦ Peak Area
- Include kinetics
- 4 Printer Font

Click the **Select** button for changing the front to be used in the report.

### 3.3 Defining Your Samples

- **3.3.1 Without Using** When "Use sample table" is not selected in Analysis Method, a sample **Sample Table** must be defined in the following procedure before measurement.
  - (1) Click the button. The window in Fig. 3-10 will appear.



Fig. 3-10

(2) Make entries for Sample name and Comment.Maximum number of enterable characters (half-size)

Sample name: 40 characters

Comment: 255 characters

For automatic saving, put a check mark in the "Auto File" box to specify a file name. First click the **Select** button, and then specify a saving location and enter a file name.

When you put a check mark at the head of "Auto JCAMP-DX file," "Auto Text file" and "Auto Meta file" after turning on "Auto File," time scan data can be saved automatically in the following file formats as soon as this data is automatically saved.

- JCAMP-DX file (\*.dx)
- ASCII text file (\*.txt)
- Graph meta file (\*.wmf)

The data will be saved in the file name entered for "File name."

(3) After completion of setting, click **OK**.

# 3.3.2 With Sample Table

When "Use sample table" is selected in Analysis Method, samples must be defined in the following procedure before measurement.

Open the monitor window and click the button.

When the Sample Table window in Fig. 3-11 appears, enter the sample name and comment for each sample.

1 For creating a sample table newly, make entry for Number of Samples and click the **Update** button. For instance, if there are five samples, enter "5" for Number of Samples and click the **Update** button.

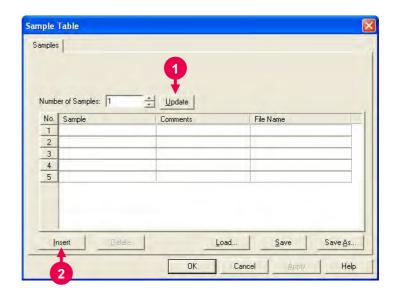


Fig. 3-11 Sample Table

- **2** For adding a row at the end of sample table, click the **Insert** button.
- 3 To delete an unnecessary row, click its row No. to make it active as shown in Fig. 3-12. Then, click the **Delete** button.

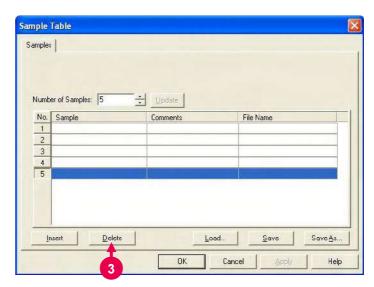


Fig. 3-12

4 Make entries for Sample and Comments of each sample.
The sample names entered here appear on the monitor window.

Maximum number of characters to be entered (half-size)

Sample : 40 characters Comment : 255 characters

Next, you must enter a file name for automatic saving after measurement.

When you click a File Name column, the [...] button appears as shown in Fig. 3-13. Click this button. Upon opening of the window shown in Fig. 3-14, specify a saving location and enter a file name.

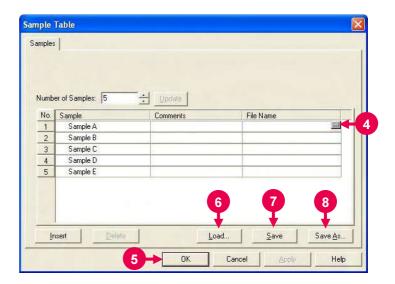


Fig. 3-13



Fig. 3-14

- 5 After entries, check them and then click the **OK** button.
- Load button
   Used for reading out the existing sample table.
   Clicking the Load button displays the file opening dialog.

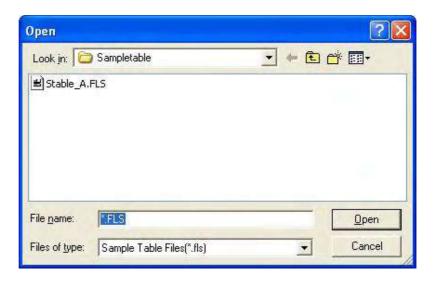


Fig. 3-15 File Opening Dialog

Select a sample table and click the **Open** button. The specified sample table will be read out.

#### 7 Save button

The contents of change can be saved by overwriting. In case of a new generation, the Save As dialog will open.

#### 8 Save As button

Used for saving the sample table with a name newly given. Click this button, and the Save As dialog will open.

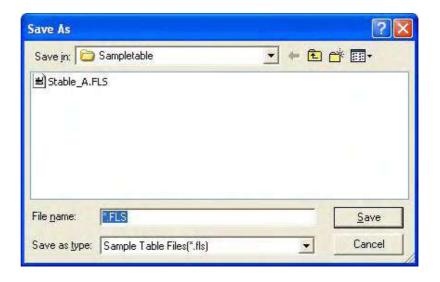


Fig. 3-16 Save As Dialog

Select a saving location and enter a file name. Then, click the **Save** button.

### 3.4 Conducting Measurement

# 3.4.1 Without Using Sample Table

- (1) When "Use sample table" is not selected in Analysis Method, you should click the button and make sure that a sample has already been defined.
- (2) Set a sample and click (measurement) button.

  Measurement starts and a spectrum is displayed.

  For halting the measurement midway, click the button. (stop)
- (3) Upon completion of the measurement, the measured data will be saved automatically at the specified saving location using the specified file name when auto saving is selected. If auto saving is not selected, select the "Save As" command from the File menu and save the data. Opening the saved data file will display the data processing window as shown in Fig. 3-17. This figure shows a spectrum and the result of rate calculation. Peak table can be displayed, but both the result of rate calculation and the peak table cannot be displayed simultaneously. Simultaneous display is selectable from the View menu.

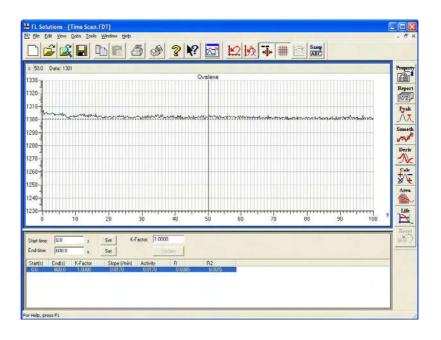


Fig. 3-17 Data Processing Window (time scan)

# 3.4.2 With Sample Table

- (1) Click the button and make sure a sample has already been defined.
- (2) Set a sample and click the button to start measurement
- (3) Upon start of measurement, the window shown in Fig. 3-18 appears. Click **Yes**.

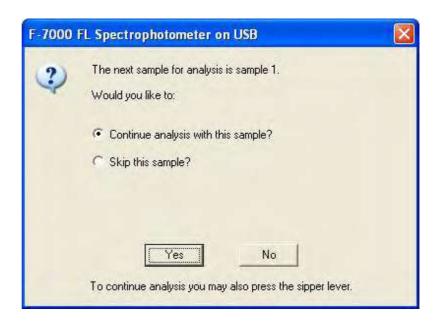


Fig. 3-18

(4) A dialog box opens as shown below.

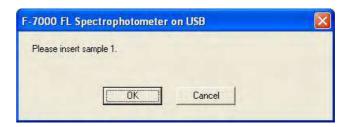


Fig. 3-19

Click **OK**. Measurement starts and a spectrum is displayed.

For halting the measurement midway, click the (stop)

Measure all the samples set in the sample table according to the message guide.

#### 3.4 Conducting Measurement

(5) Upon completion of the measurement, the measured data will be saved automatically at the specified saving location using the specified file name. Opening the saved data file will display the data processing window as shown in Fig. 3-20. This figure shows a spectrum and the result of rate calculation. Peak table can be displayed, but both the result of rate calculation and the peak table cannot be displayed simultaneously. Simultaneous display is selectable from the View menu.

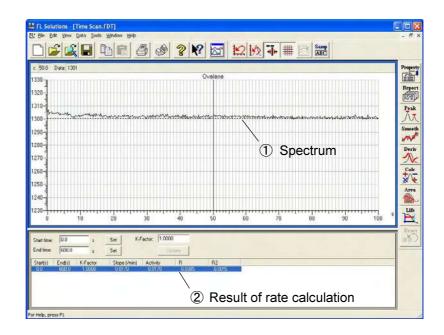


Fig. 3-20 Data Processing Window (wavelength scan)

- Spectrum
   The spectrum of a measured sample is displayed.
- ② Result of rate calculation The result of rate calculation (slope, activity, R, R2) are displayed. Start time and end time are settable by numeric input or clicking Set with the line cursor aligned. After setting, click Update for recalculation. For rate calculation, refer to Appendix B.

When peak table is selected, peak time, valley time, peak start/end times and peak area will be displayed.

# 3.4.3 Measurement in Stopped Flow Mode

- (1) Put a check mark for 'Stopped Flow' at the Instrument tab in the Analysis Method window. At the General tab in this window, remove a check mark from "Use sample table."
- (2) Click the button to make sure a sample has already been defined.
- (3) Click the (measurement) button to start measurement.
- (4) When measurement starts, the window shown in 3-21 appears to await an external start signal.

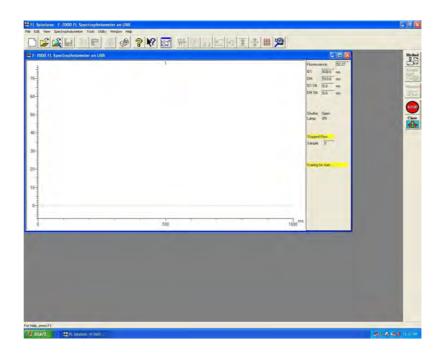


Fig. 3-21 Stopped Flow Measurement Window (external signal wait status)

#### 3.4 Conducting Measurement

(5) On arrival of the external signal, measurement starts and a spectrum is drawn in the window.

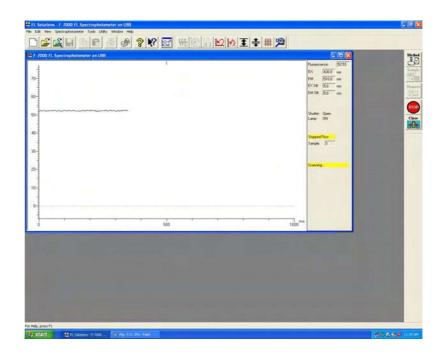


Fig. 3-22 Stopped Flow Measurement Window

For stopping the measurement halfway, click the (stop) button.

(6) Upon completion of the measurement, the measured data will be saved automatically at the specified saving location using the specified file name when auto saving is selected.
If auto saving is not selected, select the "Save As" command from the File menu and save the data.

- NOTICE: 1. The data measured under the following conditions will be saved automatically in Program Files\FL Solutions\Data. (Example: C:Program Files\FL Solutions\Data)
  - Measurement was carried out using a sample table with no file name specified in the File Name column.
  - (2) Measurement was carried out with "Auto File" specified and a file name unspecified, avoiding use of a sample table.
  - The data measured under the following conditions will be saved automatically in Program Files\FL Solutions.
     (Example: C:Program Files\FL Solutions)
    - (3) There is not an adequate free area in the specified storage medium (such as floppy disk or MO).
    - (4) Writing is inhibited.

A file automatically saved under such a condition as above is named according to the following rule.

- Rule 1: When a file name is given to analysis method: File name of analysis method + serial No. (001, 002, ...)
- Rule 2: When a file name is not given to analysis method: DEFLT + serial No. (001, 002, ...)

### 3.5 Data Printout

Printout can be provided even when the function for automatic printout after measurement is not used. The following two procedures are available.

(1) Click on the spectrum window, and the circumference turns blue. In this status, select the Print command from the File menu.

Or click the (print) button on the toolbar. The field active on the data processing window (e.g., 1) in Fig. 3-20) will be printed. What is displayed by clicking the Print Preview command in the File menu will be printed.

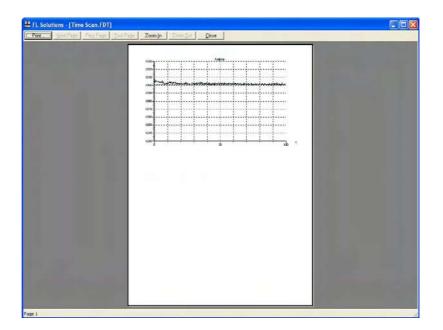


Fig. 3-23

(2) Select the Report command in the Data menu. Or click the (report) button on the toolbar. The Print Preview window will appear as shown below, so click **Print**.

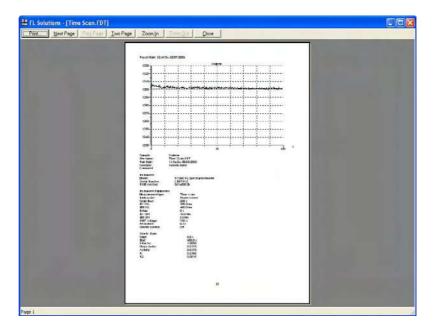


Fig. 3-24

NOTE: If characters overflow from the print frame in repeat measurement or in overlaying, then change the printing orientation at the Report tab shown in Fig. 3-25 from vertical (Portrait) to horizontal (Landscape).

Select the items to be printed in the following procedure.

Click the button, and the Spectrum Properties window will appear.

Select the Report tab, and the window in Fig. 3-25 will open.  $\label{eq:control}$ 

In this window, select desired print items.

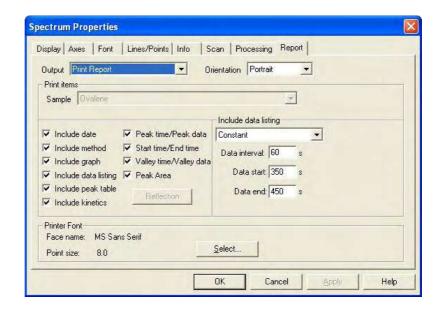


Fig. 3-25

### 3.6 Calculating the Phosphorescence Life

This is the function for calculating phosphorescence life from the measurement result (phosphorescence attenuation curve).

Replacing data value at T1 with  $I_0$ ,  $I_{0/e}$  time (T2) is found, and T1, T2 and  $\tau$  (phosphorescence life) = T2 - T1 are displayed.

Select the phosphorescence life command from the Data menu or click the button on the Processing toolbar. A window as shown in Fig. 3-26 will appear.

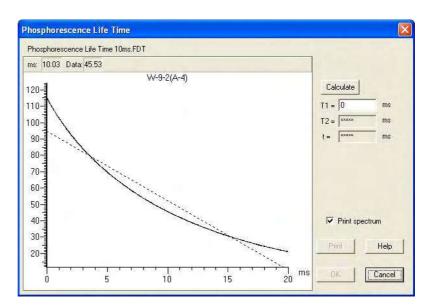


Fig. 3-26

Move the line cursor to the start point (T1) of calculation by the mouse or arrow keys  $[\leftarrow]$  and  $[\rightarrow]$  on the keyboard. Right-click the mouse at the start point. Or enter a numeral in the T1 field and click **Calculate**. The result of calculation will be displayed.

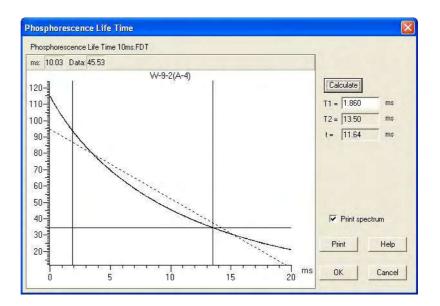


Fig. 3-27

For printout of the calculation result, click **Print**. For printout of the spectrum and calculation result, apply a check mark to "Print spectrum" and click **Print**.

For scale expansion, specify the area to be enlarged with the mouse. Draw a rectangle with the mouse button held down as shown in Fig. 3-28.

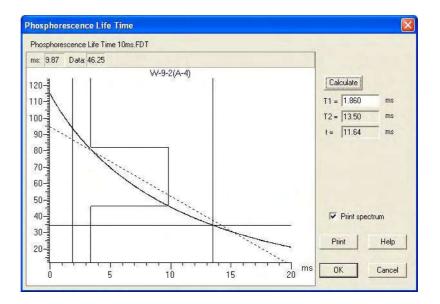


Fig. 3-28

When releasing the mouse button, the specified area will be enlarged as shown in Fig. 3-29.

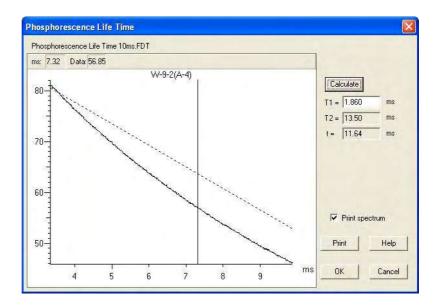


Fig. 3-29

Double-clicking on the spectrum selects auto scale.

### 4. PHOTOMETRY

### 4.1 Flow of Operation

This section explains the method of determining sample concentration with a calibration curve prepared. Measurement proceeds along the flow shown below.

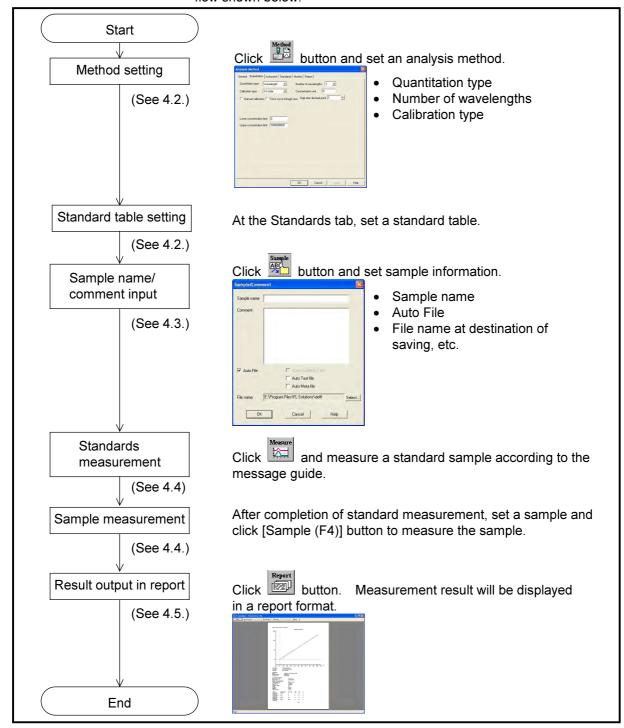


Fig. 4-1 Operational Flow (photometry)

#### **Creating an Analysis Method** 4.2

Set the analytical conditions (method) prior to measurement.



From the Edit menu, select the Method command or click the button. A window shown in Fig. 4-2 will appear.

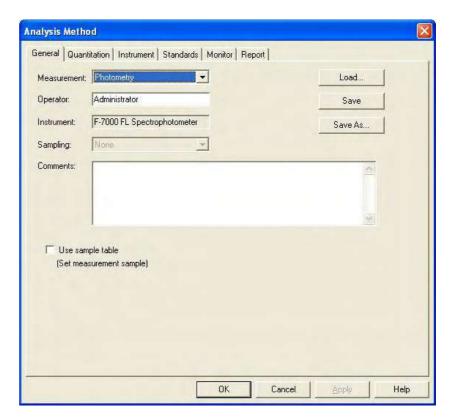


Fig. 4-2 Analysis Method

The following tabs are provided on the Analysis Method window for the photometry mode. These tabs are detailed on the subsequent pages.

- General
- Quantitation
- Instrument
- Standards
- Monitor
- Report

#### 4.2.1 General Tab

The parameters shown in Fig. 4-3 are provided at the General tab. For the settable range of each parameter, refer to Appendix G.

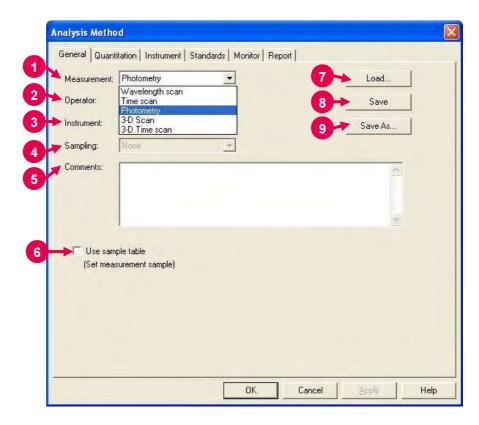


Fig. 4-3 General Tab

#### 1 Measurement (mode)

A measurement mode is selectable. Here, select "Photometry." For Wavelength scan, Time scan, 3-D Scan and 3-D Time scan, refer to Sections 2, 3, 5 and 6, respectively.

OperatorEnter an operator name.

#### 3 Instrument

The name of the connected instrument is indicated.

#### 4 Sampling

- Standard

Indicates that samples are to be exchanged manually.

#### 5 Comments

Enter a description or notes on measuring conditions.

6 Use sample table (Set measurement sample)
Apply a check mark here when a sample table is used.

#### 4.2 Creating an Analysis Method

#### 7 Load

Click this button to read out saved analytical conditions.

Upon clicking, the file opening window appears. So, select an analytical condition file.

#### 8 Save

When you click this button after setting the parameters, the analytical conditions can be saved by overwriting. In case of a new analysis method, clicking this button displays the "Save As" window. You must enter a file name for saving in this window.

#### 9 Save As

Click this button for renaming the set conditions for saving. The "Save As" window appears, so enter a file name.

## 4.2.2 Quantitation Tab

Upon selection of the Quantitation tab, the window shown in Fig. 4-4 appears.

For the settable range of each parameter, refer to Appendix G.

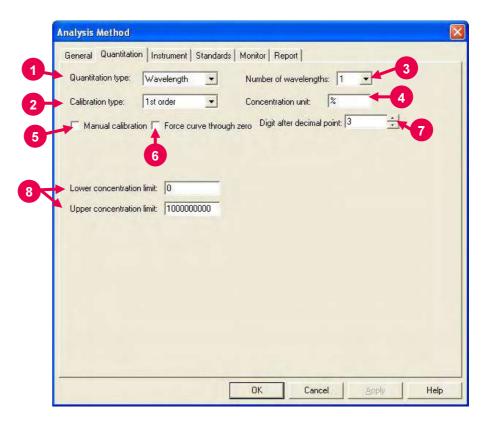


Fig. 4-4 Quantitation Tab

#### 1 Quantitation type

Quantitation type is selectable among Wavelength, Peak area, Peak height, Derivative and Ratio. Spectra measured by the Peak area, Peak height and Derivative methods can be checked, referring to 7.19.

#### (a) Wavelength

Upon selecting 'None' for Calibration type in this quantitation method, photometric values can be obtained at the specified number of wavelengths for Number of wavelengths.

A calibration curve will not be prepared.

Upon selecting 1st order, 2nd order, 3rd order or Segmented in this quantitation method, calibration curve data and sample data will be obtained by one, two or three-wavelength calculation depending on your entry for Number of wavelengths.

### (i) One wavelength calculation

This is the most generally used method. When you specify one wavelength and enter it at the Instrument tab, a calibration curve and concentration are determined from the data  $I_1$  at the specified wavelength as shown in Fig. 4-5 (a).

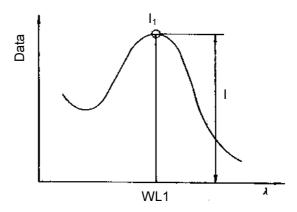


Fig. 4-5 (a)

#### (ii) Two wavelength calculation

When you specify two wavelengths and enter them at the Instrument tab, a calibration curve and concentration are calculated through determination of I by the equation below assuming  $I_1$  and  $I_2$  are the photometric values at two different wavelengths as shown in Fig. 4-5 (b).

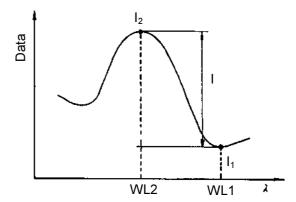


Fig. 4-5 (b)

(iii) Three wavelength calculation

As shown in Fig. 4-5 (c), assuming  $I_1$ ,  $I_2$  and  $I_3$  are the data values at three different wavelengths, a calibration curve and concentration are calculated through determination of I by the equation below. Photometry is made in the order of WL1, WL2 and WL3.

$$I = I_2 - \frac{I_1 (WL3 - WL2) + I_3 (WL2 - WL1)}{WL3 - WL1}$$

But the following relation must hold: WL1 < WL2 < WL3 or WL1 > WL2 > WL3

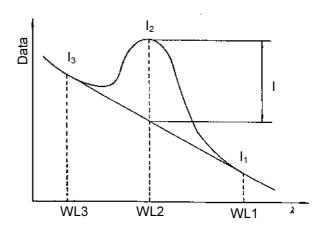


Fig. 4-5 (c)

#### (b) Peak area

Enter the peak wavelength and its accuracy for Peak apex. Integration method (area detection method) is selectable among Rectangular, Trapezoid and Romberg. (Refer to "integration method" in Appendix D.2.) Then, set a threshold value and sensitivity for defining the peak, referring to 7.6.

Measurement will be carried out within the wavelength range set at the Instrument tab. According to the specified threshold level and sensitivity, peak wavelength is sought. The area of the peak found within a range of the entered Peak apex  $\pm$  wavelength accuracy is calculated, thereby preparing a calibration curve.

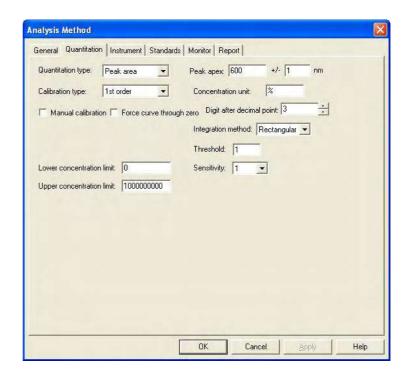


Fig. 4-6 Quantitation Tab (peak area)

#### (c) Peak height

Enter the peak wavelength and its accuracy for Peak apex. Measurement will be carried out within the wavelength range set at the Instrument tab. According to the threshold level and sensitivity specified for the "Peak area" quantitation method, peak wavelength is sought. A calibration curve is prepared using the height of the peak found within a range of the entered Peak apex  $\pm$  wavelength accuracy.

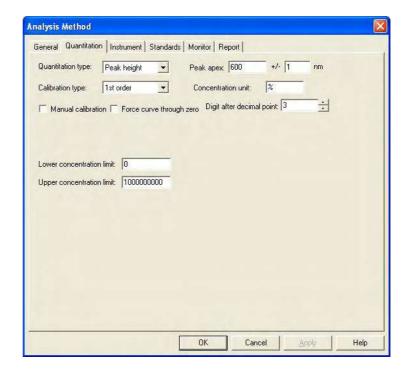


Fig. 4-7 Quantitation Tab (peak height)

#### (d) Derivative

Enter peak wavelength (or wavelength to be used for calibration curve preparation) for Peak apex.

Measurement will be carried out within the wavelength range set at the Instrument tab. A calibration curve is created using the derivative value at the peak wavelength.

Assign your values for derivative parameters; Derivative order, Smoothing order and Number of points.

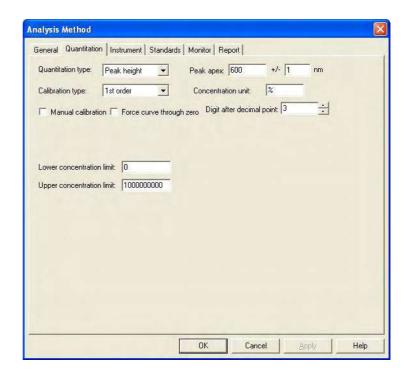


Fig. 4-8 Quantitation Tab (derivative)

#### (e) Ratio

Upon selecting Ratio, an input column for three factors (K-factor 0, K-factor 1, K-factor 2) and a setting column for calculation method (+, -, \*) are displayed.

**NOTE**: By this quantitation method, a calibration curve cannot be created.

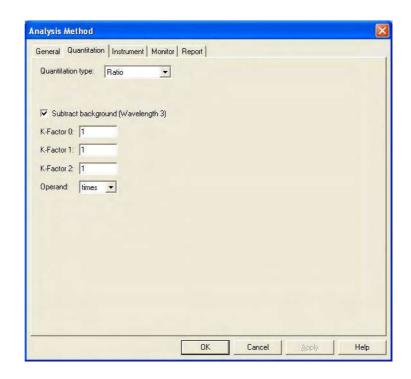


Fig. 4-9 Quantitation Tab (peak ratio)

Calculation is made by the following equation.

RATIO = 
$$\frac{\text{K1 (A (WL1) - A (WL3))}}{\text{K2 (A (WL2) - A (WL3))}}$$
 #K0

# (calculation method): +, -, \*

WL1, 2, 3 : Specified wavelengths

WL3 : Background correcting wavelength

A ( ) : Measured value at specified wavelength

#### 2 Calibration type

Set a calibration type. For selecting factor input (when the formula of standard calibration curve and its factors are already known), put a mark in the check box for Manual calibration and enter values for A0, A1, A2 and A3.

#### (a) None

Mode in which calibration curve is not used. When this mode is selected with Wavelength set for Quantitation type, multi-wavelength measurement at up to six wavelengths is possible. However, no spectrum can be displayed.

#### (b) 1st order

This specifies the most generally used calibration curve. A linear calibration curve is to be created. The formula of calibration curve is as follows.

$$x = A_1y + A_0$$

Here, x is sample concentration and y is sample data.

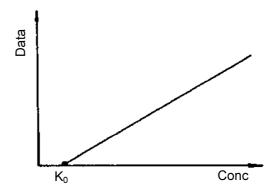


Fig. 4-10 1st Order

#### (c) 2nd order

A calibration curve is created using a quadratic curve. The formula of calibration curve is as follows.

$$x = A_2y^2 + A_1y + A_0$$

Here, x is sample concentration and y is sample data.

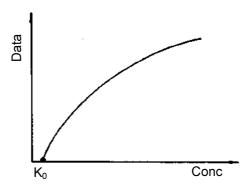


Fig. 4-11 2nd Order

#### (d) 3rd order

A calibration curve is created using a cubic curve. The formula of calibration curve is as follows.

$$x = A_3y^3 + A_2y^2 + A_1y + A_0$$

Here, x is sample concentration and y is sample data.

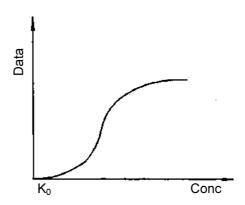


Fig. 4-12 3rd Order

#### (e) Segmented

A calibration curve created by connecting standard samples linearly according to the measured values of respective samples or input data.

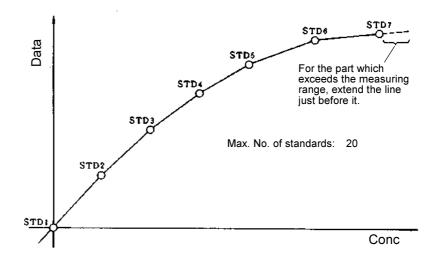
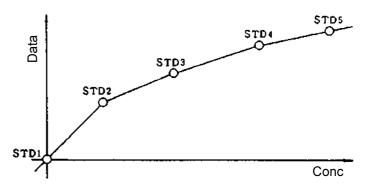
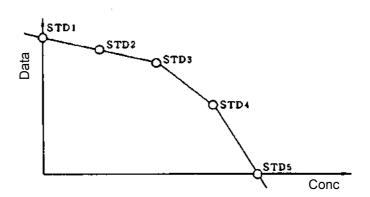


Fig. 4-13 Segmented

NOTE: A proper calibration curve can be created when the photometric value increases or decreases monotonously versus the concentration value. With a monotonic increase, the calibration curve is created with standard 1 (STD1) as origin. And with a monotonic decrease, the calibration curve is created with STD1 plotted on the photometric value axis and STD5 on the concentration axis. In these examples, the number of standards is set at 5.



Calibration curve with monotone increase



Calibration curve with monotone decrease

- Number of wavelengths

  Enter the desired number of wavelengths to be used for measurement.
- 4 Concentration unit Enter a concentration (in mg/mL for example) from the keyboard.

5 Manual calibration

Put a check mark in this box for creating a calibration curve using factors. Enter factors in response to the selected calibration type.

- 6 Force curve through zero
  By putting a check mark in this box, a calibration curve is created so that its factor A<sub>0</sub> takes 0 automatically.
- 7 Digit after decimal point Set the number of digits below decimal point for indicating the calculated concentration of the measured sample. The concentration value is rounded off to the set number of digits.
- Lower concentration limit/Upper concentration limit

  This is used to set a normal value range for the concentration of the measured sample. If the concentration is higher than the upper limit set here, "HIGH" will appear in the Conc column, and if lower than the lower limit set here, "LOW" will appear in the Conc column.

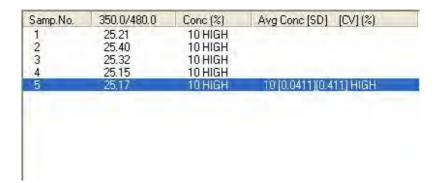


Fig. 4-14 (measurement example with upper limit set at 5)

# 4.2.3 Instrument Tab

When you click the Instrument tab, the window in Fig. 4-15 appears. For the settable range of each parameter, refer to Appendix G.

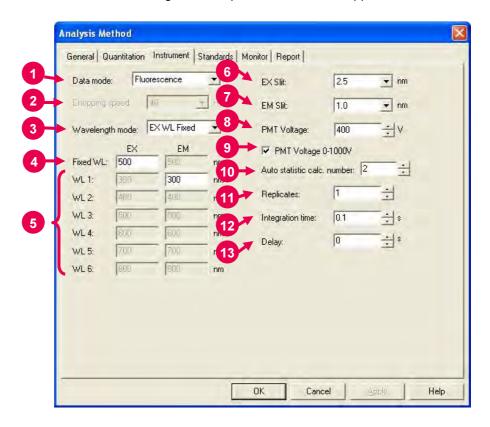


Fig. 4-15 Instrument Tab

#### 1 Data mode

Select one of the three data modes. The Luminescence mode is for measuring luminescence in the biological/chemical field. In this mode, only the sample side signal is acquired without performing ratio photometry.

#### 2 Chopping speed

Select a chopping speed. Setting is allowed when Phosphorescence is selected for Data mode.

The Phosphorescence mode is for acquiring only phosphorescence after extinction of excitation beam while turning a chopper on the excitation side. An increase in chopping speed makes the chopper turns faster.

Wavelength mode Selects a wavelength setting for creation of a calibration curve.

#### 4 Fixed WL

When 'EX WL Fixed' is selected in Wavelength mode, set the fixed wavelength on the excitation side.

When 'EM WL Fixed' is selected in Wavelength mode, set the fixed wavelength on the emission side.

#### 5 WL (1 to 6)

Make your entry for the number of wavelengths specified at the Quantitation tab.

#### 6 EX Slit

Select a slit width on the excitation side.

#### 7 FM Slit

Select a slit width on the emission side.

#### 8 PMT Voltage

A function for controlling the voltage of the photomultiplier detector (photomultiplier tube). As a reference, changing from 400 V to 700 V increases the data value about two digits and changing from 700 V to 950 V increases it about 1 digit.

When a check mark is put for PMT Voltage 0-1000V, setting is allowed within a range from 0 to 1,000 V.

#### 9 PMT Voltage 0-1000V

When a check mark is put in this box, the photomultiplier voltage is settable within a range from 0 to 1000 V.

#### 10 Auto statistic cal. number

Calculation is made for mean value, SD (standard deviation) and CV (coefficient of variation). Set the number of samples for this purpose.

#### 11 Replicates

Set the number of measurements to be repeated.

#### 12 Integration time

A function for obtaining data averaged over the specified time for acquisition of stabilized data.

#### **13** Delay

After pressing the Measure button, measurement is started following the delay time set here. It is used for stabilization after setting an unknown sample, etc. In repeat measurement, it is the time until the start of the first measurement. The delay time is invalid for the second and subsequent measurements.

**4.2.4 Standards Tab** Clicking the Standards tab opens the window shown in Fig. 4-16. For the settable range of each parameter, refer to Appendix G.

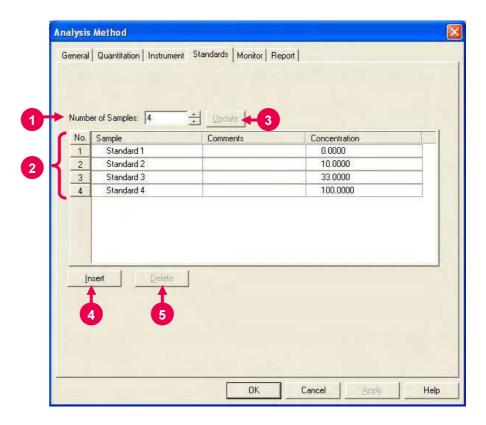


Fig. 4-16 Standards Tab

Number of Samples
For generating a sample table newly, enter the number of samples

here, and then click the **Update** button.

2 Standard Table
 This table gives a list of standards for sample measurement or
 calibration curve preparation. The table contains the items listed
 below.

NOTE: The size of the standard table columns (cells) is changeable.

Bring the cursor to a boundary line between the left and right table headers, and it changes into one shown below.

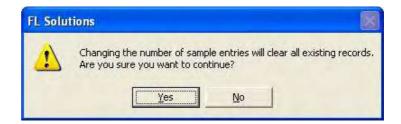


Now click the mouse button on the line and drag it to the left or right to obtain a suitable column size.

- No.
  - This is the number given to each sample.
- Sample
  - Enter the name of each sample.
- Comment
  - Enter comments on each sample.
- Concentration
  - Enter the concentration of each sample.

### 3 Update

When clicking this button, a confirmation message appears.



Clicking **Yes** will clear the presently entered sample names and comments, and table rows are formed by the entered number of samples.

For adding a row below the final one in the sample list, click the **Insert** button.

#### 4 Insert

When you click the **Insert** button, data will be added below the final row in the standard list.

### 5 Delete

Click the column of the sample No. to be deleted, and it becomes active. Then, click the **Delete** button, and the relevant sample data will be deleted.

### 4.2.5 Monitor Tab

When selecting the Monitor tab, the window in Fig. 4-17 appears. For the settable range of each parameter, refer to Appendix G.

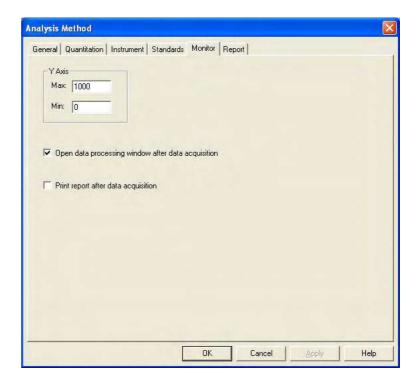


Fig. 4-17 Monitor Tab

Assign the following parameters.

- 1 Y-Axis Max (maximum)

  Enter a maximum value for Y-axis on the monitor window.
- Y-Axis Min (minimum)
  Enter a minimum value for Y-axis on the monitor window.
- Open data processing window after data acquisition
  Select whether or not to conduct data processing after sample
  measurement. When selected (check mark is applied), an icon
  for this function (as shown in
  Opening the saved file allows data processing such as peak
  detection. When this function is not selected, data processing can
  be achieved by reading out the file via [File]-[Open].
- 4 Print report after data acquisition Select whether or not to print a report after sample measurement. When selected (check mark is applied), printing will be made automatically after measurement. The items set at the Report tab will be printed.

### 4.2.6 Report Tab

Select the Report tab, and the window in Fig. 4-18 will appear.

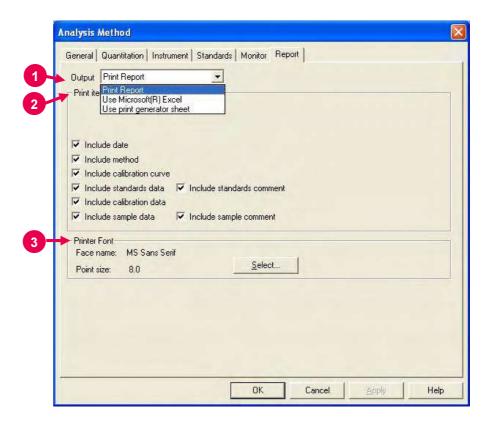


Fig. 4-18 Report Tab

On this window, select the items to be contained in an analysis report.

### 1 Output

- Print Report
   A report will be printed. For printing method, refer to 4.5.
- Use Microsoft(R) Excel
   Data will be transferred to Microsoft(R) Excel. For transferring method, refer to 7.13.
- Use print generator sheet
   Select this method for using the report generator (option).
   For usage, refer to the instruction manual of the report generator.

### 4.2 Creating an Analysis Method

- 2 Print items (printable/transferable items)
  - Include date
  - Include method
  - Include calibration curve
  - Include standards data
  - Include calibration data
  - Include sample data
- 3 Printer Font Click the Select button for changing the front to be used in the report.

## 4.3 Defining Your Samples

# 4.3.1 Without Using Sample Table

When "Use sample table" is not selected in Analysis Method, a sample must be defined in the following procedure before measurement.

(1) Click the button. The window in Fig. 4-19 will appear.

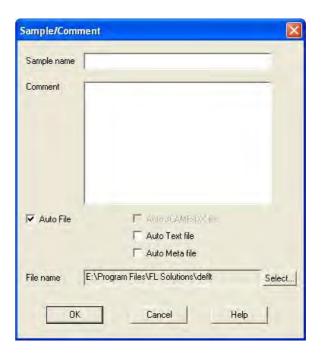


Fig. 4-19

Make entries for Sample name and Comment.Maximum number of enterable characters (half-size)

Sample name: 40 characters

Comment: 255 characters

For automatic saving, put a check mark in the "Auto File" box to specify a file name. First click the **Select** button, and then specify a saving location and enter a file name.

When you put a check mark at the head of "Auto Text file" and "Auto Meta file" after turning on "Auto File," photometry data can be saved automatically in the following file formats as soon as this data is automatically saved.

- ASCII text file (\*.txt)
- Graph meta file (\*.wmf)

The data will be saved in the file name entered for "File name."

(3) After completion of setting, click **OK**.

# 4.3.2 With Sample Table

When "Use sample table" is selected in Analysis Method, samples must be defined in the following procedure before measurement.

Open the monitor window and click the button.

When the Sample Table window in Fig. 4-20 appears, enter the sample name and comment for each sample.

1 For creating a sample table newly, make entry for Number of Samples and click the **Update** button. For instance, if there are five samples, enter "5" for Number of Samples and click the **Update** button.

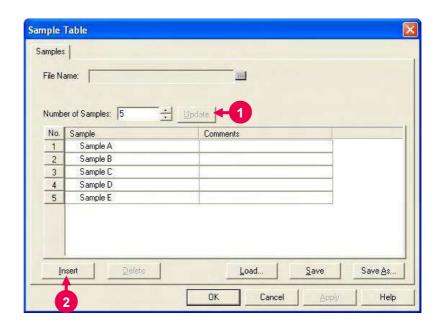


Fig. 4-20 Sample Table

- **2** For adding a row at the end of sample table, click the **Insert** button.
- 3 To delete an unnecessary row, click its row No. to make it active as shown in Fig. 4-21. Then, click the **Delete** button.

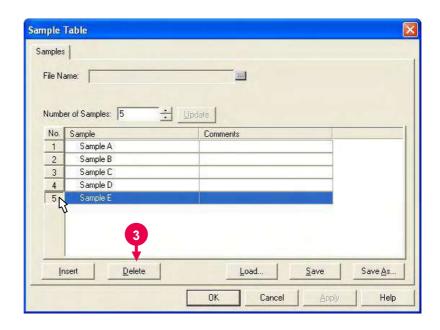


Fig. 4-21

Make entries for Sample and Comments of each sample.
Maximum number of characters to be entered (half-size)

Sample : 40 characters Comment : 255 characters

Next, you must enter a file name for automatic saving after measurement. Click the [...] button, and specify a saving location and enter a file name.

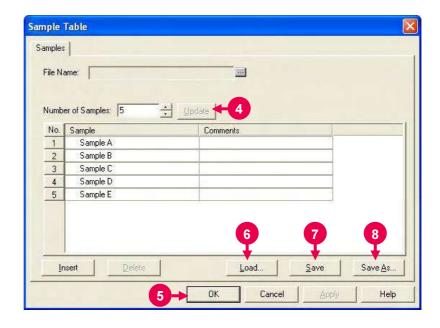


Fig. 4-22

### 4.3 Defining Your Samples

5 After entries, check them and then click the **OK** button.

### 6 Load button

Used for reading out the existing sample table.

Clicking the **Load** button displays the file opening dialog.

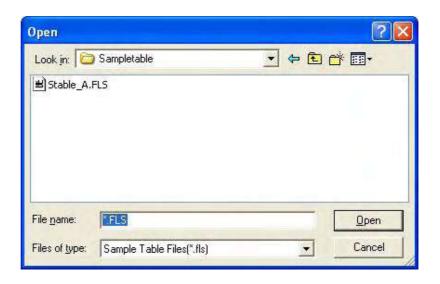


Fig. 4-23 File Opening Dialog

Select a sample table and click the **Open** button.

The specified sample table will be read out.

### 7 Save button

The contents of change can be saved by overwriting. In case of a new generation, the Save As dialog will open.

### 8 Save As button

Used for saving the sample table with a name newly given. Click this button, and the Save As dialog will open.

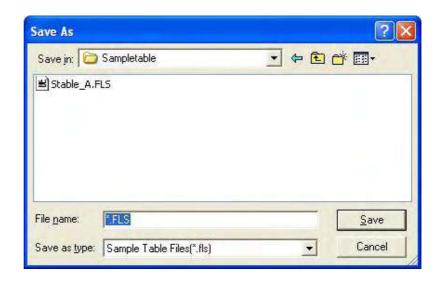


Fig. 4-24 Save As Dialog

Select a saving location and enter a file name. Then, click the **Save** button.

## 4.4 Conducting Measurement

# 4.4.1 Standards Measurement

First create a calibration curve, and then proceed to sample measurement.

- (1) Click , select the Standards tab and make sure standards have already been defined. (Refer to 4.2.4.)

  Click the button or button to also confirm that the samples have already been defined. (Refer to 4.3.)
- (2) Click the (measurement) button to start measurement. When measurement starts, the window in Fig. 4-25 appears.

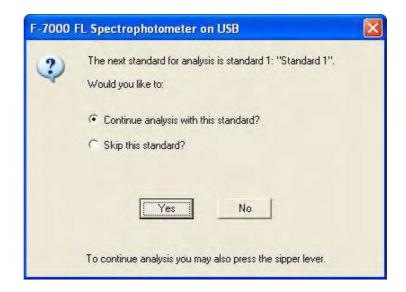


Fig. 4-25

(3) When you click **Yes**, and the window in Fig. 4-26 will appear.



Fig. 4-26

Set standard sample 1 and click OK.

This standard sample is measured and advance to the next standard measurement is awaited. Since a window as shown in Fig. 4-25 appears, you should click **Yes**.

Then, a window as shown in Fig. 4-26 appears. Set the next standard sample and click **OK**. Continue standard measurement until the standards set at the Standards tab in Analysis Method are all measured.

(4) When measurement of all standards is finished, a window as shown in Fig. 4-27 appears. Clicking of the **OK** button under selection of "Go to measure samples" takes you to measurement of unknown samples.



Fig. 4-27

# 4.4.2 Sample Measurement

Now let's measure samples.

Operation differs between when a sample table is used and when it is not used.

(1) Without using a sample table(when "Use sample table" is not selected in Analysis Method)

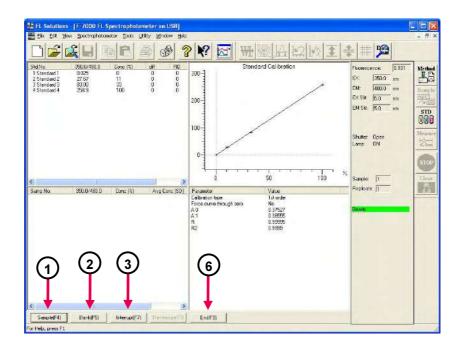


Fig. 4-28

- Set a sample and click the Sample(F4) button.
   Measurement is carried out and the result is displayed.
- ② For blank measurement at the mid-point of sample measurement, click **Blank(F5)**.
- To interrupt the measurement for other samples, click the Interrupt(F7) button.

The window in Fig. 4-29 will appear. Enter a number for Interrupt's Sample No.

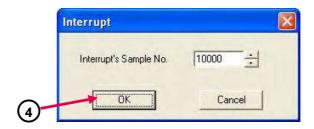


Fig. 4-29

4 Click the **OK** button, and the window in Fig. 4-30 will appear.

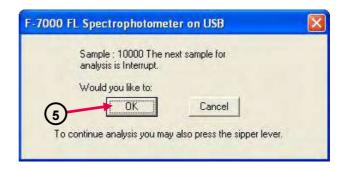


Fig. 4-30

- ⑤ Click the **OK** button, and measurement of the interruption sample starts.
- When sample measurement is finished, click the End(F9) button.

When auto saving is selected, the measured data will be saved automatically at the specified saving location using the specified file name.

When auto saving is not selected, you must select the "Save As" command from the File menu to save the measured data.

### **Entry and Change of Sample Name**

After completion of sample measurement, sample names can be entered on the monitor window (Fig. 4-31).

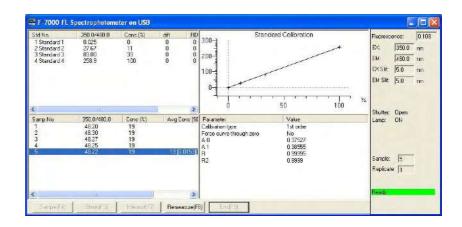


Fig. 4-31

Follow steps (a) to (d).

### 4.4 Conducting Measurement

(a) Select Edit Sample Name from the Edit menu as shown in Fig. 4-32.



Fig. 4-32

(b) The Edit Sample Name dialog opens as shown in Fig. 4-33.

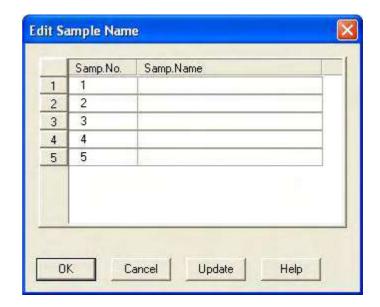


Fig. 4-33

(c) Enter sample names as exemplified in Fig. 4-34 and click the **Update** or **OK** button.

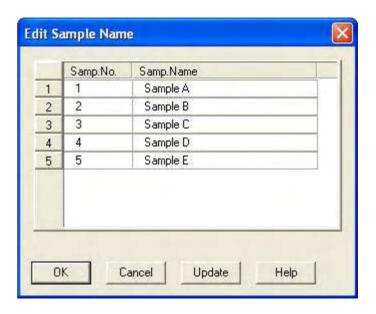


Fig. 4-34

(d) The monitor window is redisplayed with the newly entered sample names as shown in Fig. 4-35.

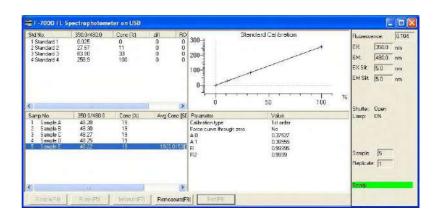


Fig. 4-35

For saving the data after entry of sample names, click the

button. The data processing window appears.

Select the Save As command from the File menu and save the data.

For changing the sample names, follow the same procedure as for entry of sample names.

(2) With sample table (when "Use sample table" is selected in Analysis Method)

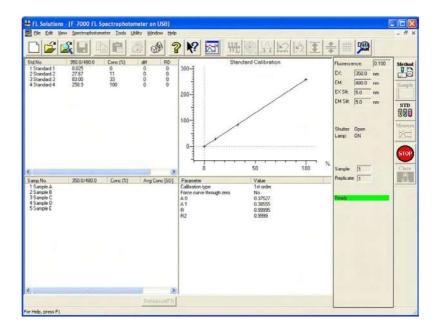


Fig. 4-36

The message box shown in Fig. 4-37 appears. Carry out operation according to the messages. Measurement can be continued by clicking Yes and OK alternately.

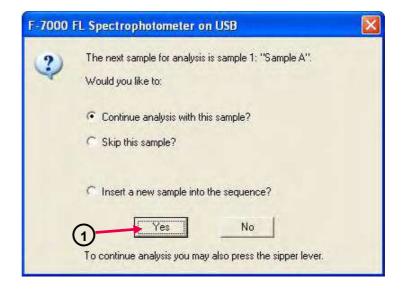


Fig. 4-37

② To interrupt the measurement for other samples, put a check mark for "Insert a new sample into the sequence?" as shown in Fig. 4-38. Then, click **Yes**.

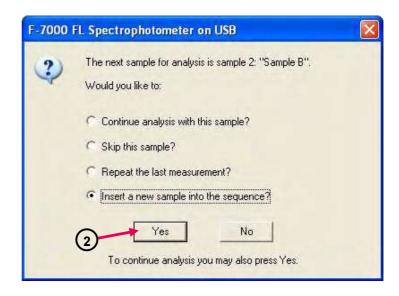


Fig. 4-38

③ The window in Fig. 4-39 appears. Enter the sample name and comment, and then click **OK**.



Fig. 4-39

### 4.4 Conducting Measurement

**When you click OK**, measurement of the interruption sample starts.



Fig. 4-40

When measurement of all samples is over, the data processing window shown in Fig. 4-41 will be displayed automatically (only when "Open data processing window after data acquisition" is selected at the Monitor tab in Analysis Method).

After measurement of all the samples entered for Sample Table, the photometric data will be saved automatically at the specified saving location using the specified file name.

### 4.4.3 Explanation of Measurement Result Window

**Explanation of** Upon completion of photometry or when opening the data file, the data **Measurement** processing window is displayed as shown in Fig. 4-41.

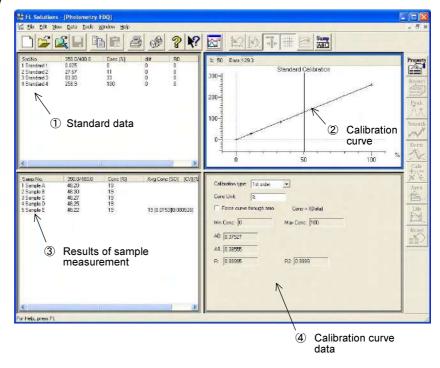


Fig. 4-41 Data Processing Window (photometry)

### 1 Standard data

The measurement results of standards used for preparing a calibration curve are displayed.

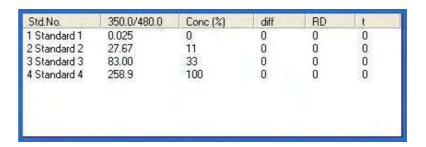


Fig. 4-41-1 Standard Data

### 2 Calibration curve

A calibration curve obtained from standard data is displayed.

The curve can also be traced.

Photometric value and Concentration are indicated at the top left of this field.

### 4.4 Conducting Measurement

Results of sample measurement Sample data such as photometric value (Data), concentration (Conc), mean value (Avg Conc), standard deviation (SD), coefficient of variation (CV) and upper/lower limit judgment results (HIGH/LOW) are displayed.

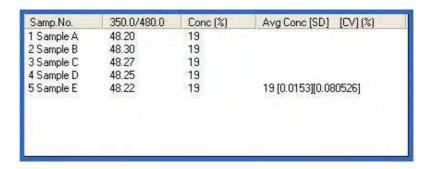


Fig. 4-41-2 Results of Sample Measurement

4 Calibration curve data Data such as determination coefficient and factors (A0, A1, A2, A3) of calibration curve are displayed. And the calibration type can be changed.

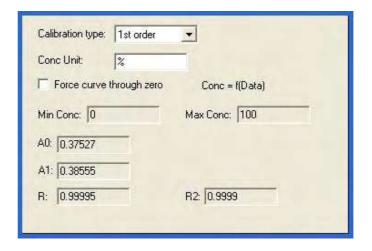


Fig. 4-41-3 Calibration Curve Data

- NOTICE: 1. The data measured under the following conditions will be saved automatically in Program Files\FL Solutions\Data. (Example: C:Program Files\FL Solutions\Data)
  - Measurement was carried out using a sample table with no file name specified in the File Name column.
  - (2) Measurement was carried out with "Auto File" specified and a file name unspecified, avoiding use of a sample table.
  - The data measured under the following conditions will be saved automatically in Program Files\FL Solutions.
     (Example: C:Program Files\FL Solutions)
    - (3) There is not an adequate free area in the specified storage medium (such as floppy disk or MO).
    - (4) Writing is inhibited.

A file automatically saved under such a condition as above is named according to the following rule.

- Rule 1: When a file name is given to analysis method: File name of analysis method + serial No. (001, 002, ...)
- Rule 2: When a file name is not given to analysis method: DEFLT + serial No. (001, 002, ...)

### 4.4.4 Remeasurement

If remeasurement becomes necessary after completing a series of measurements, both standard and sample data accept remeasurement.

① Click the data to be remeasured for making it active as shown in Fig. 4-42.

NOTE: Figure 4-42 shows the window for the case where the sample table is not used. Five buttons including the Remeasure(F8) are displayed. On the window for the case where the sample table is used, only the Remeasure(F8) button is displayed.

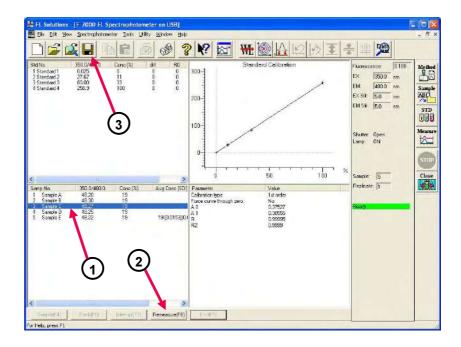


Fig. 4-42 Measurement End Window (without using sample table)

② Click the **Remeasure(F8)** button. A window as shown in Fig. 4-43 will appear.

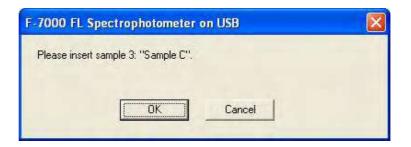


Fig. 4-43

- 3 Click **OK**, and measurement will start. When standards are remeasured, a calibration curve will be displayed through recalculation of the data.
- 4 For saving data after remeasurement, click the button. The data processing window appears. From the File menu, select "Save As" and save the data.

Remeasurement is possible for both standard and sample data.

# 4.4.5 Statistical Calculation

Statistical calculation ensues from sample measurement. The results are displayed in the "Avg Conc" column of the data processing window as shown in Fig. 4-41-2.

$$MEAN = \frac{\sum X_i}{N}$$

$$SD = \sqrt{\frac{\sum X_i^2 - (\sum X_i)^2 / N}{N - 1}}$$

$$CV = \frac{SD}{MEAN} \times 100$$

Here, N = Number of standards

MEAN = Mean value

SD = Standard deviation

CV = Coefficient of variation (%)

**NOTE**: [\*\*\*\*\*] is indicated for SD and CV values when they are nearly "0."

### 4.5 Data Printout

Printout can be provided even when the function for automatic printout after measurement is not used. The following two procedures are available.

(1) Click on the spectrum window, and the circumference turns blue. In this status, select the Print command from the File menu.

Or click the (print) button on the toolbar. The field active on the data processing window (e.g., ② in Fig. 4-41) will be printed. What is displayed by clicking the Print Preview command in the File menu will be printed.

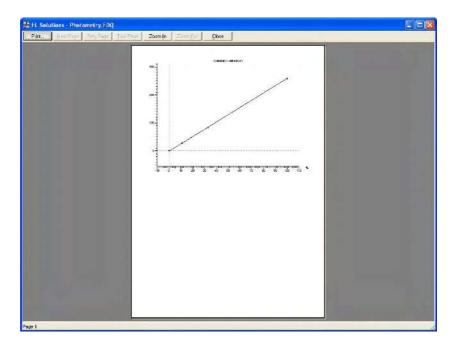


Fig. 4-44

(2) Select the Report command in the Data menu. Or click the report) button on the toolbar. The Print Preview window will appear as shown below. For printout, click the **Print** button. Measurement conditions will be printed together.

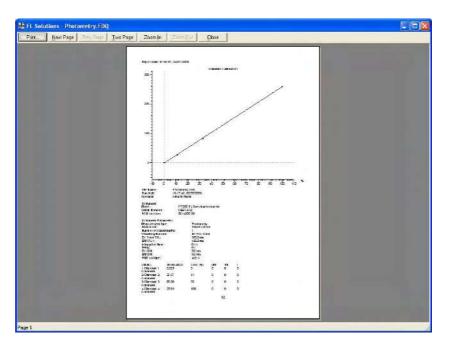


Fig. 4-45

Select the items to be printed at the Report tab with the Photometry Properties command chosen in the Edit menu.

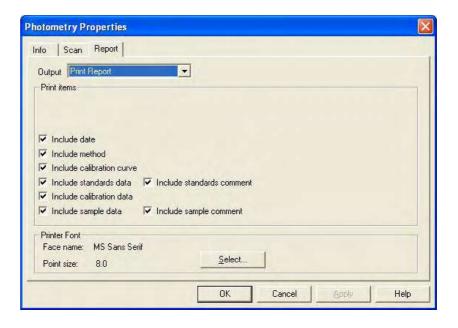


Fig. 4-46

## 4.6 Manual Input of Standard Data

The standard data can be entered manually. This function is usable after measurement of standards. Either select Standard Data from the Edit menu on the monitor window or click the button. A window as shown in Fig. 4-47 will appear. So enter values in the Data column. After entry, click the **OK** or **Update** button. Then, the calibration curve is recalculated and displayed.

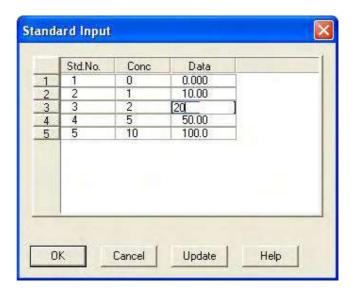


Fig. 4-47

## 4.7 Measurement Using Saved Calibration Curve

Call a calibration curve in the following way.

- (1) Set measurement mode in Photometry.
- (2) From the File menu, select the calibration curve load command. The following message is reported in a dialog box. This message does not appear immediately after turning on power supply or when measurement mode is other than Photometry.

Click the **OK** button.



Fig. 4-48

(3) A window as shown below will appear. Select the file of the calibration curve to be used.
After file selection, click the **Open** button.

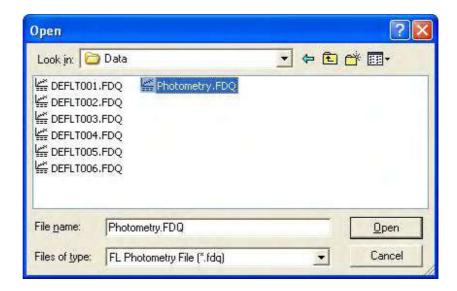


Fig. 4-49

(4) The window in Fig. 4-50 is now displayed. The sample data section is blank.

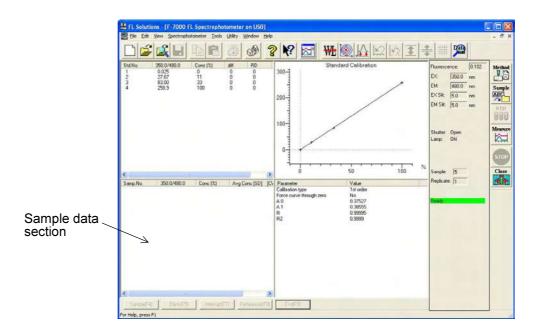


Fig. 4-50

(5) Click the button, and the window in Fig. 4-51 will appear. Enter a sample name and comment. Click the **OK** button.



Fig. 4-51

- (6) Set a sample to be measured. Click the button. (Or select the Scan command from the Spectrophotometer menu.)
- (7) The following message box appears. Click the **OK** button.

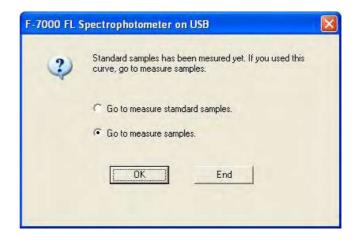


Fig. 4-52

(8) The **Sample** button is now active. Click this button. Measurement starts.

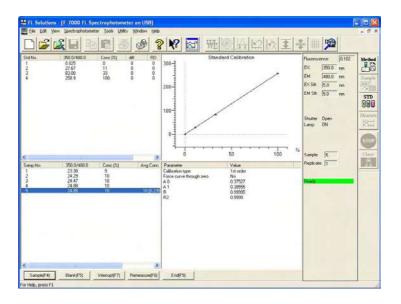


Fig. 4-53

(9) After measurement, click the **End(F9)** button for termination.

### 5. 3-D SCAN

## 5.1 Flow of Operation

This section explains the parameter setting and operating method for 3-D scan. Measurement proceeds along the following flow.

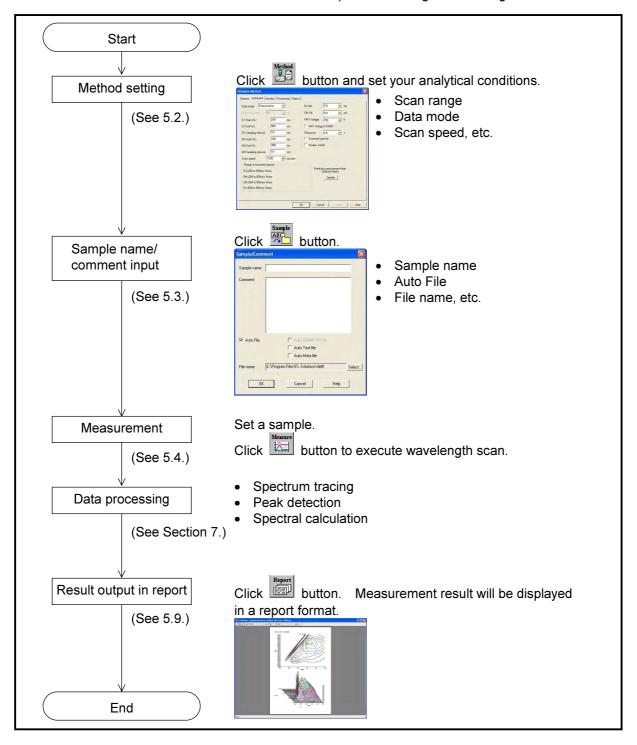


Fig. 5-1 Operational Flow (3-D scan)

## 5.2 Creating an Analysis Method

Set the analytical conditions (method) prior to measurement.

(1) Click the button on the Measurement toolbar. The window in Fig. 5-2 appears.

(Or select the Method command from the Edit menu.)

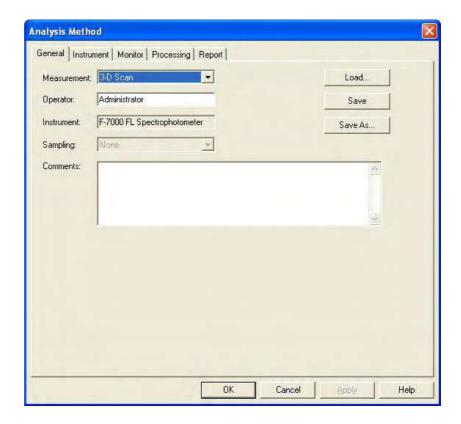


Fig. 5-2 Analysis Method

- (2) The following tabs are provided on the Analysis Method window for the 3-D scan mode. These tabs are detailed on the subsequent pages.
  - General
  - Instrument
  - Monitor
  - Processing
  - Report

### 5.2.1 General Tab

The parameters shown in Fig. 5-3 are provided at the General tab. For the settable range of each parameter, refer to Appendix G.

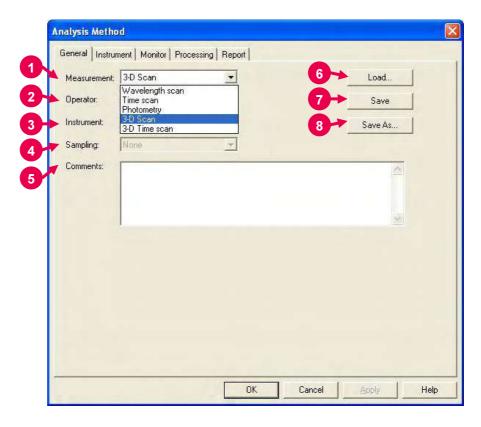


Fig. 5-3 General Tab

### 1 Measurement (mode)

A measurement mode is selectable. Here, select "3-D Scan." For Wavelength scan, Time scan, Photometry and 3-D Time scan, refer to Sections 2, 3, 4 and 6, respectively.

2 Operator

Enter an operator name.

3 Instrument

The name of the connected instrument is indicated.

- 4 Sampling
  - Standard

Indicates that samples are to be exchanged manually.

5 Comments

Enter a description or notes on measuring conditions.

### 5.2 Creating an Analysis Method

### 6 Load

Click this button to read out saved analytical conditions.

Upon clicking, the file opening window appears. So, select an analytical condition file.

### 7 Save

When you click this button after setting the parameters, the analytical conditions can be saved by overwriting. In case of a new analysis method, clicking this button displays the "Save As" window. You must enter a file name for saving in this window.

### 8 Save As

Click this button for renaming the set conditions for saving. The "Save As" window appears, so enter a file name.

# 5.2.2 Instrument Tab

The window shown below is based on that 3-D Scan is specified for Measurement at the General tab. When you click the Instrument tab, the window in Fig. 5-4 appears.

For the settable range of each parameter, refer to Appendix G.

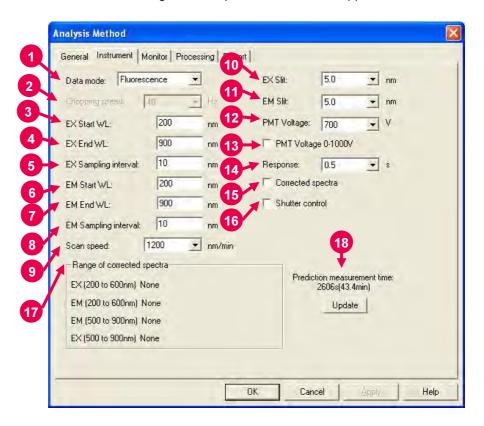


Fig. 5-4 Instrument Tab

### 1 Data mode

Select a data mode. The Phosphorescence mode is for acquiring only phosphorescence after extinction of excitation beam while turning a chopper on the excitation side.

### 2 Chopping speed

Select a chopping speed. Setting is allowed when Phosphorescence is selected for Data mode.

The Phosphorescence mode is for acquiring only phosphorescence after extinction of excitation beam while turning a chopper on the excitation side. An increase in chopping speed makes the chopper turns faster.

### 3 EX Start WL

This is the start wavelength on the excitation side in wavelength scan. Enter the shorter wavelength side of measuring wavelength range.

#### 4 EX End WL

This is the end wavelength on the excitation side in wavelength scan. Enter the longer wavelength side of measuring wavelength range.

#### 5 EX Sampling interval

In the 3-dimensional scan mode, emission spectrum measurement is repeated while shifting the excitation wavelength. Therefore, a shorter sampling interval on the excitation side will result in a longer measurement time.

#### 6 EM Start WL

This is the start wavelength on the emission side in wavelength scan. Enter the shorter wavelength side of measuring wavelength range.

#### 7 EM End WL

This is the end wavelength on the emission side in wavelength scan. Enter the longer wavelength side of measuring wavelength range.

### 8 EM Sampling interval

In the 3-dimensional scan mode, emission spectrum measurement is repeated while shifting the excitation wavelength. Therefore, a shorter sampling interval on the emission side will result in a longer measurement time.

### 9 Scan speed

Set a wavelength scan speed.

### 10 EX Slit

Select a slit width on the excitation side.

#### 11 EM Slit

Select a slit width on the emission side.

#### 12 PMT Voltage

A function for controlling the voltage of the photomultiplier detector (photomultiplier tube). As a reference, changing from 400 V to 700 V increases the data value about two digits and changing from 700 V to 950 V increases it about 1 digit.

When a check mark is put for PMT Voltage 0-1000V, the photomultiplier voltage is settable within a range from 0 to 1000 V.

#### 13 PMT Voltage 0-1000V

When a check mark is put in this box, the photomultiplier voltage is settable within a range from 0 to 1000 V.

### 14 Response

When "Auto" is selected, response will be set automatically depending on scan speed. Select "Auto" usually.

### 15 Corrected spectra

A function for determining the spectrum inherent to a sample by correcting the photometer wavelength characteristic using the saved instrument parameters, following measurement with the instrument parameters for photometer control. If there are no instrument parameters for the wavelength range to be measured, only the wavelength range that allows correction is scanned. And if there is no wavelength area measurable, an error will occur. The Instrument Response command is provided in the Utility menu for operating this function.

When this setting is at ON, "Corrected spectra" appears on the monitor window. (See Fig. 1-43 in 1.6.)

#### 16 Shutter control

The shutter can automatically be closed in other than measurement for slowing down sample deterioration due to the energy of excitation beam and opened when measurement starts.

17 Range of corrected spectra Spectrum correction range is indicated.

#### 18 Prediction measurement time

The predicted measurement time is indicated. After change of a parameter such as scan speed, click the **Update** button. The predicted value will then be updated.

#### 5.2.3 Monitor Tab

When selecting the Monitor tab, the window in Fig. 5-5 appears. For the settable range of each parameter, refer to Appendix G.

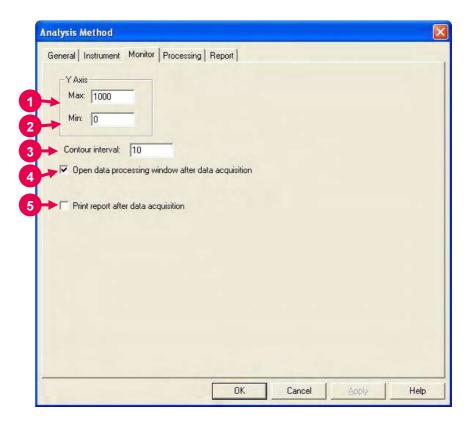


Fig. 5-5 Monitor Tab

Assign the following parameters.

- 1 Y-Axis Max (maximum)

  Enter a maximum value for Y-axis on the monitor window.
- Y-Axis Min (minimum)
  Enter a minimum value for Y-axis on the monitor window.
- 3 Contour interval Function for setting intervals of drawing contour lines.
- Open data processing window after data acquisition
  Select whether or not to conduct data processing after sample
  measurement. When selected (check mark is applied), an icon
  (as shown in Naphth... Did is displayed for the data
  processing window at the end of measurement. And by opening
  this icon, data processing such as peak detection is available.

NOTE: When the remaining memory capacity of PC in operation is very small (system resource is less than 20%), the data processing window will not open even if this item is turned ON. Either close the presently opened data processing window or restart the FL Solutions program.

Print report after data acquisition
Select whether or not to print a report after sample measurement.
When selected (check mark is applied), printing will be made automatically after measurement. The items set at the Report tab will be printed. When this item is not selected, data processing is allowed by reading out a file via [File] - [Open].

# 5.2.4 Processing Tab

On selection of the Processing tab, the window in Fig. 5-6 will be displayed.

For the settable range of each parameter, refer to Appendix G.

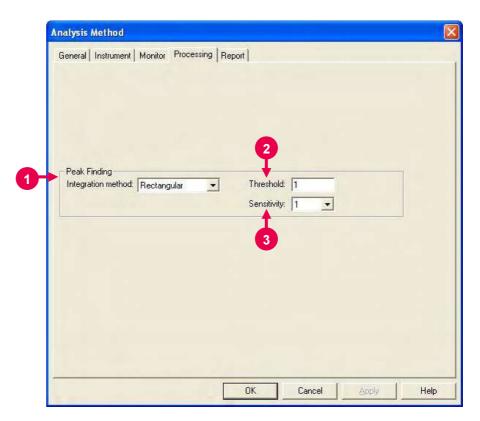
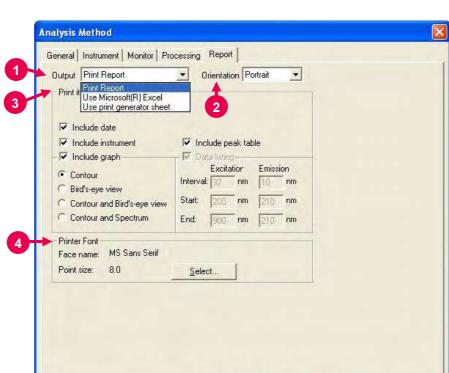


Fig. 5-6 Processing Tab

- 1 Peak Finding: Integration method
  Select an integration method for the peaks to be displayed in the data processing window.
- Threshold Set a detection limit for the photometric value axis of peak and valley. Peaks or valleys below the setting will not be detected.
- 3 Sensitivity Select the number of data points in the horizontal axis direction (abscissa). For detecting sharp peaks, select a sensitivity value of "1." Or select "8" for detecting broad peaks.



**5.2.5** Report Tab Select the Report tab, and the window in Fig. 5-7 will appear.

Fig. 5-7 Report Tab

OK

Cancel

Help

# 1 Output

- Print Report
   A report will be printed. For printing method, refer to 5.9.
- Use Microsoft(R) Excel
   Data will be transferred to Microsoft(R) Excel. For transferring method, refer to 7.13.
- Use print generator sheet
   Select this method for using the report generator (option).
   For usage, refer to the instruction manual of the report generator.

## 2 Orientation

Selectable when Print Report is chosen for Output.

Select a printing orientation. Either Portrait or Landscape orientation is selectable. When the Landscape orientation is selected, date and method can be printed.

## 3 Print items (printable/transferable items)

- Include date
- Include instrument
- Include graph

When you put a check mark for this item, any of the graphs listed below is selectable.

- ♦ Contour
- ♦ Bird's-eye view
- ♦ Contour and Bird's-eye view
- ♦ Contour and Spectrum
- Include peak table
- Data listing
  - ◊ Interval
  - ♦ Start
  - ♦ End

These parameters are settable when "Use Microsoft(R) Excel" or "Use print generator sheet" is specified for Output. For time scan, make entries for Interval, Start and End. For wavelength scan, make entries for Interval, Start and End.

## 4 Printer Font

Click the **Select** button for changing the font to be used in the report.

# 5.3 Defining Your Samples

A sample must be defined in the following procedure before measurement.

(1) Click the (sample name/comment input) button on the Measurement toolbar. The window in Fig. 5-8 will appear.



Fig. 5-8

(2) Make entries for Sample name and Comment.Maximum number of enterable characters (half-size)

Sample name: 40 characters

Comment: 255 characters

For automatic saving, put a check mark in the "Auto File" box to specify a file name. First click the **Select** button, and then specify a saving location and enter a file name.

When you put a check mark at each head of "Auto Text file" and "Auto Meta file" after turning on "Auto File," 3-D scan data can be saved automatically in the following file formats as soon as this data is automatically saved.

- ASCII text file (\*.txt)
- Graph meta file (\*.wmf)

The data will be saved in the file name entered for "File name."

(3) After completion of setting, click **OK**.

# 5.4 Conducting Measurement

- (1) Make sure an analysis method has been set and a sample has been defined. (Refer to "5.2 Creating an Analysis Method" and "5.3 Defining Your Samples.")
- (2) Set a sample in the sample compartment and click the button on the Measurement toolbar to start measurement. (See ① in Fig. 5-9.)

  On this monitor window, the X and Y axes stand for the EM and EX-side wavelengths, respectively.

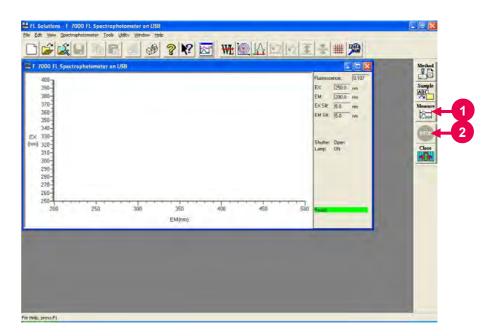


Fig. 5-9 Monitor Window

- (3) For halting the measurement midway, click the button on the toolbar. (See ② in Fig. 5-9.)
- (4) The information to be displayed upon opening the data processing window can be selected here by excluding unnecessary one from display.

  Select the Options command from the Tools menu and click the 'Display' tab in the Options dialog box. Then, the window in Fig. 5-10 appears. For example, select "Show contour plus EX,EM spectrum plus peak table" and click the **OK** button.

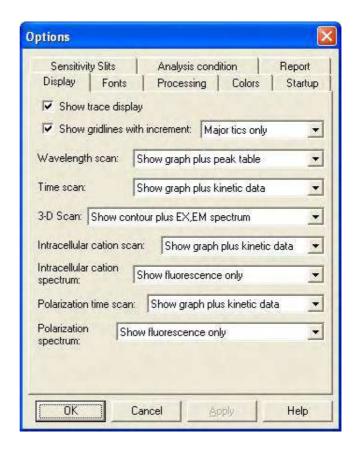


Fig. 5-10

(5) When measurement is completed, the data processing window appears as shown in Fig. 5-11. This is available only when a check mark is put for "Open data processing window after data acquisition" at the 'Monitor' tab of Analysis Method. (See ④ in Fig. 5-5.)

The measured data will be saved automatically when auto saving is selected.

If auto saving is not selected, select the "Save As" command from the File menu and save the data. Opening the saved data file will display the data processing window as shown below.

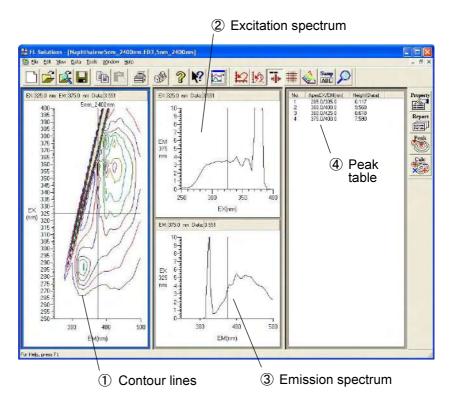


Fig. 5-11 Data Processing Window (3-D scan)

#### Contour lines

From these contour lines, excitation and emission spectra can be read out.

As the cursor is moved, the cursor-specified excitation and emission spectra are displayed in ② and ③.

# 2 Excitation spectrum

An excitation spectrum at the cursor position in ① is displayed. Wavelength and photometric value can be read by selecting the "trace" command from the View menu or clicking the button.

# 3 Emission spectrum

An emission spectrum at the cursor position in ① is displayed. Wavelength and photometric value can be read by selecting the "trace" command from the View menu or clicking the button.

### 4 Peak table

Peak wavelength and peak height are indicated. For setting threshold and sensitivity values, click the button.

- NOTICE: 1. The data measured under the following condition will be saved automatically in Program Files\FL Solutions\Data.

  (Example: C:Program Files\FL Solutions\Data)
  - (1) Measurement was carried out with "Auto File" specified and a file name unspecified.
  - The data measured under the following conditions will be saved automatically in Program Files\FL Solutions.
     (Example: C:Program Files\FL Solutions)
    - (2) There is not an adequate free area in the specified storage medium (such as floppy disk or MO).
    - (3) Writing is inhibited.

A file automatically saved under such a condition as above is named according to the following rule.

- Rule 1: When a file name is given to analysis method: File name of analysis method + serial No. (001, 002, ...)
- Rule 2: When a file name is not given to analysis method: DEFLT + serial No. (001, 002, ...)

# 5.5 Displaying Data in a Bird's-Eye View

3-D Scan data can be displayed in a bird's-eye view.

Select the (bird's-eye view) button on the Spectrum toolbar.

(Or select the bird's-eye view command from the View menu.)

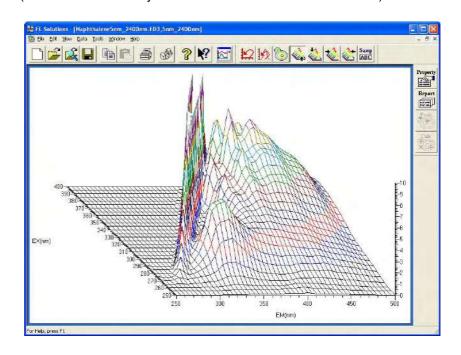


Fig. 5-12

Viewing direction is changeable by clicking (from front), (from rear), (from left) or (from right).

Clicking (contour line diagram) displays a contour line diagram.

# 5.6 Contour Scale Expansion with Mouse

In the contour display of 3-D scan data, display scale can be expanded with the mouse. On the Spectrum toolbar, click the contour scale expansion) button. The button is set in the clicked status.

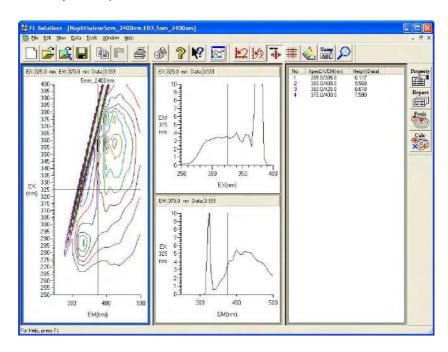


Fig. 5-13

Move the mouse cursor onto the contour as shown in Fig. 5-14 and drag the mouse along an area to be expanded with the left mouse button held down.

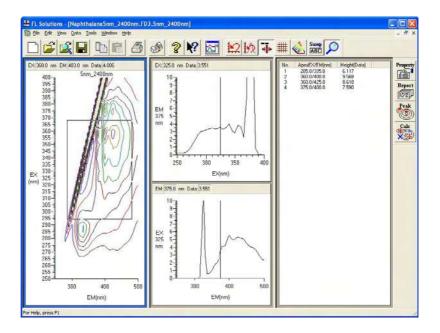


Fig. 5-14

The area enclosed with the mouse cursor is now expanded as shown in Fig. 5-15. For restoring the original display, click the reset) button. (scale

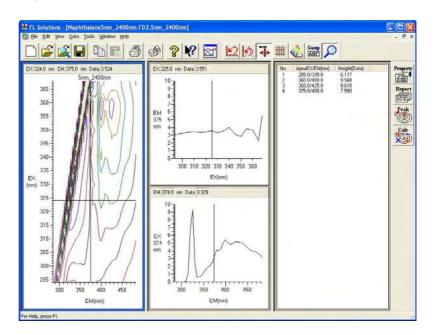


Fig. 5-15

For using the trace cursor on the contour window, click again for returning to the non-clicked status.

# 5.7 Auto Scale Display of Data

When you click the (auto scale) button on the data processing window after completion of measurement or from which the 3-D scan data file has been read out, the contour window, excitation spectrum window and emission spectrum window are displayed in the auto scale mode as shown in Fig. 5-16.

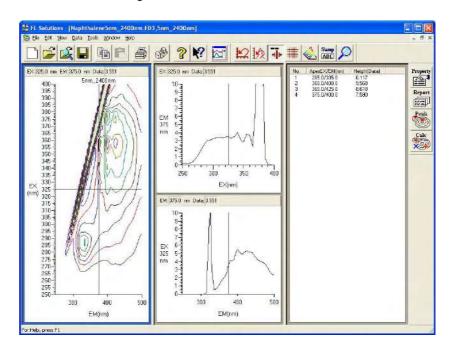


Fig. 5-16 (Before execution of auto scale)

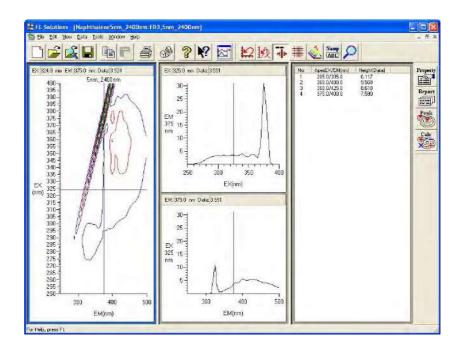


Fig. 5-17 (After execution of auto scale)

For restoring the original display, click the (scale reset) button.

# 5.8 Linking Trace Cursors from Peak Table

On the data processing window where a contour and peak table are displayed, you may move the mouse cursor onto the peak table and click the mouse. As a result, each trace cursor on the contour window, excitation spectrum window and emission spectrum window moves to the peak wavelength position.

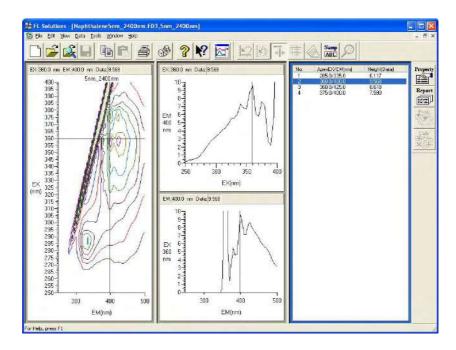


Fig. 5-18

# 5.9 Data Printout

Printout can be provided even when the function for automatic printout after measurement is not used. The following two procedures are available.

(1) Click on the spectrum window, and the circumference turns blue. In this status, select the Print command from the File menu.

Or click the (print) button on the toolbar. The field active on the data processing window (e.g., 1) in Fig. 5-11) will be printed. What is displayed by clicking the Print Preview command in the File menu will be printed.

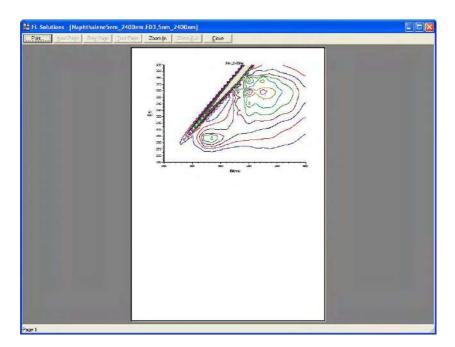


Fig. 5-19

(2) Select the Report command in the Data menu. Or click the button on the toolbar. The Print Preview window will appear as shown below, so click **Print**. Printout items should be selected at the Report tab in Spectrum Properties beforehand.

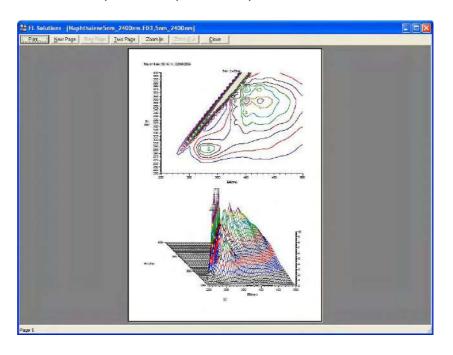


Fig. 5-20

**NOTE**: If characters overflow from the print frame in repeat measurement or in overlaying, then change the printing orientation from vertical (Portrait) to horizontal (Landscape) at the Report tab in Fig. 5-21.

Select the items to be printed in the following procedure.

Click the button, and the Spectrum Properties window will appear.

Select the Report tab, and the window in Fig. 5-21 will open.

 $\downarrow$ 

In this window, select desired print items.

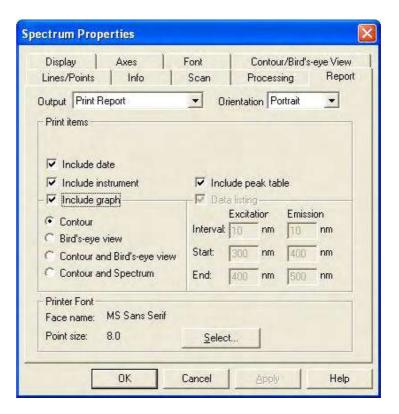


Fig. 5-21

# 6. 3-D TIME SCAN

# 6.1 Flow of Operation

This section explains the parameter setting and operating method for 3-D time scan. Measurement proceeds along the following flow.

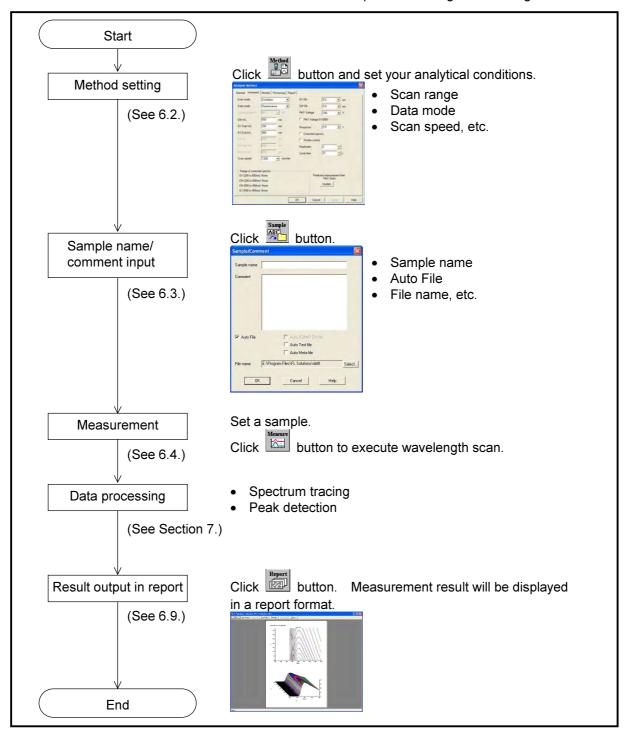


Fig. 6-1 Operational Flow (3-D time scan)

# 6.2 Creating an Analysis Method

Set the analytical conditions (method) prior to measurement.

(1) Click the button on the Measurement toolbar. The window in Fig. 6-2 appears.

(Or select the Method command from the Edit menu.)

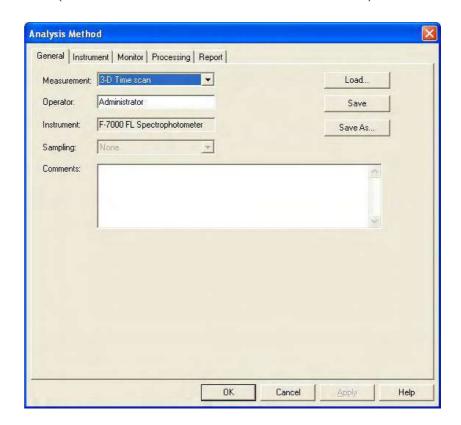


Fig. 6-2 Analysis Method

- (2) On the Analysis Method window, analytical conditions for the 3-D time scan mode can be set. The following tabs are provided on this window. These tabs are detailed on the subsequent pages.
  - General
  - Instrument
  - Monitor
  - Processing
  - Report

### 6.2.1 General Tab

The parameters shown in Fig. 6-3 are provided at the General tab. For the settable range of each parameter, refer to Appendix G.

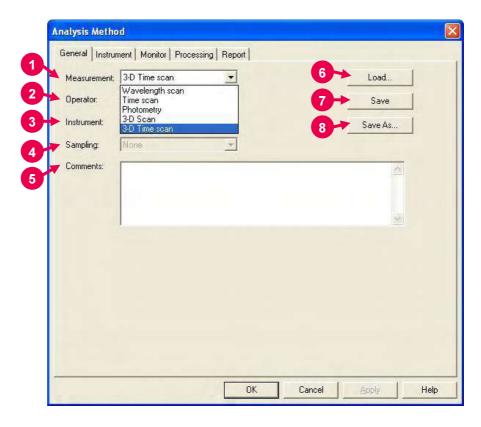


Fig. 6-3 General Tab

## 1 Measurement (mode)

A measurement mode is selectable. Here, select "3-D Time scan."

For Wavelength scan, Time scan, Photometry and 3-D Scan, refer to Sections 2, 3, 4 and 5, respectively.

# 2 Operator

Enter an operator name.

#### 3 Instrument

The name of the connected instrument is indicated.

### 4 Sampling

- Standard

Indicates that samples are to be exchanged manually.

### 5 Comments

Enter a description or notes on measuring conditions.

## 6.2 Creating an Analysis Method

#### 6 Load

Click this button to read out saved analytical conditions.

Upon clicking, the file opening window appears. So, select an analytical condition file.

## 7 Save

When you click this button after setting the parameters, the analytical conditions can be saved by overwriting. In case of a new analysis method, clicking this button displays the "Save As" window. You must enter a file name for saving in this window.

#### 8 Save As

Click this button for renaming the set conditions for saving. The "Save As" window appears, so enter a file name.

# 6.2.2 Instrument

The window shown below is based on that 3-D Time scan is specified for Measurement at the General tab. When you click the Instrument tab, the window in Fig. 6-4 appears.

For the settable range of each parameter, refer to Appendix G.

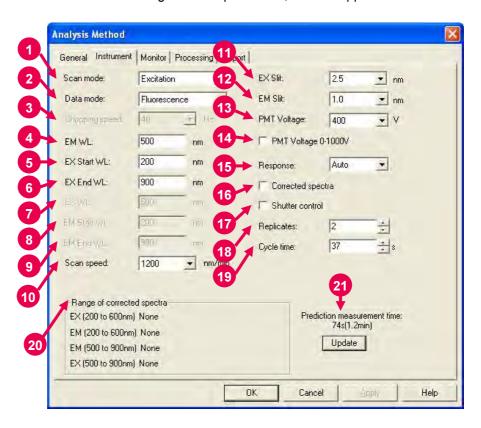


Fig. 6-4 Instrument Tab

#### 1 Scan mode

Select a scan mode here. In case of Synchronous, the abscissa of the graph stands for excitation wavelength. The unit indication of the abscissa becomes 'EX/EM (nm).'

#### 2 Data mode

Select a data mode. The Phosphorescence mode is for acquiring only phosphorescence after extinction of excitation beam while turning a chopper on the excitation side.

#### 3 Chopping speed

Select a chopping speed. Setting is allowed when Phosphorescence is selected for Data mode.

The Phosphorescence mode is for acquiring only phosphorescence after extinction of excitation beam while turning a chopper on the excitation side. An increase in chopping speed makes the chopper turns faster.

#### 4 EM WL

For the Excitation scan mode, enter the fixed wavelength on the emission side . For the Synchronous scan mode, enter the start wavelength on the emission side.

#### 5 EX Start WL

This is the start wavelength on the excitation side in wavelength scan. Enter the shorter wavelength side of measuring wavelength range.

#### 6 EX End WL

This is the end wavelength on the excitation side in wavelength scan. Enter the longer wavelength side of measuring wavelength range.

#### 7 EX WL

For the Emission scan mode, enter the fixed wavelength on the excitation side.

#### 8 EM Start WL

This is the start wavelength on the emission side in wavelength scan. Enter the shorter wavelength side of measuring wavelength range.

#### 9 EM End WL

This is the end wavelength on the emission side in wavelength scan. Enter the longer wavelength side of measuring wavelength range.

# 10 Scan speed

Set a scan speed.

# 11 EX Slit

Select a slit width on the excitation side.

#### 12 EM Slit

Select a slit width on the emission side.

#### 13 PMT Voltage

A function for controlling the voltage of the photomultiplier detector (photomultiplier tube). As a reference, changing from 400 V to 700 V increases the data value about two digits and changing from 700 V to 950 V increases it about 1 digit.

When a check mark is put for PMT Voltage 0-1000V, the photomultiplier voltage is settable within a range from 0 to 1000 V.

#### 14 PMT Voltage 0-1000V

When a check mark is put in this box, the photomultiplier voltage is settable within a range from 0 to 1000 V.

# 15 Response

When "Auto" is selected, response will be set automatically depending on scan speed. Select "Auto" usually.

#### 16 Corrected spectra

A function for determining the spectrum inherent to a sample by correcting the photometer wavelength characteristic using the saved instrument parameters, following measurement with the instrument parameters for photometer control. If there are no instrument parameters for the wavelength range to be measured, only the wavelength range that allows correction is scanned. And if there is no wavelength area measurable, an error will occur. The Instrument Response command is provided in the Utility menu for operating this function.

When this setting is at ON, "Corrected spectra" appears on the monitor window. (See Fig. 1-43 in 1.6.)

#### 17 Shutter control

The shutter can automatically be closed in other than measurement for slowing down sample deterioration due to the energy of excitation beam and opened when measurement starts.

# 18 Replicates

Set the number of repeat measurements.

#### 19 Cycle time

Set a repetition interval.

#### 20 Range of corrected spectra

Spectrum correction range is indicated.

#### 21 Prediction measurement time

The predicted measurement time is indicated. After change of a parameter such as scan speed, click the **Update** button.

The predicted value will then be updated.

#### 6.2.3 Monitor Tab

When selecting the Monitor tab, the window in Fig. 6-5 appears. For the settable range of each parameter, refer to Appendix G.

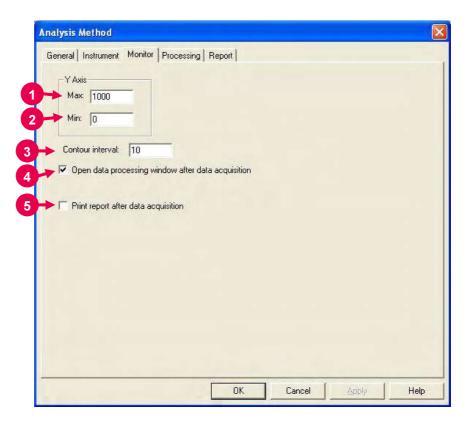


Fig. 6-5 Monitor Tab

Assign the following parameters.

- 1 Y-Axis Max (maximum)

  Enter a maximum value for Y-axis on the monitor window.
- Y-Axis Min (minimum)
  Enter a minimum value for Y-axis on the monitor window.
- 3 Contour interval Function for setting intervals of drawing contour lines.
- Open data processing window after data acquisition
  Select whether or not to conduct data processing after sample
  measurement. When selected (check mark is applied), an icon
  (as shown in Shutte... Similar is displayed for the data
  processing window at the end of measurement. And by opening
  this icon, data processing such as peak detection is available.

NOTE: When the remaining memory capacity of PC in operation is very small (system resource is less than 20%), the data processing window will not open even if this item is turned ON. Either close the presently opened data processing window or restart the FL Solutions program.

Print report after data acquisition
Select whether or not to print a report after sample measurement.
When selected (check mark is applied), printing will be made automatically after measurement. The items set at the Report tab will be printed. When this item is not selected, data processing is allowed by reading out a file via [File] - [Open].

# 6.2.4 Processing Tab

On selection of the Processing tab, the window in Fig. 6-6 will be displayed.

For the settable range of each parameter, refer to Appendix G.

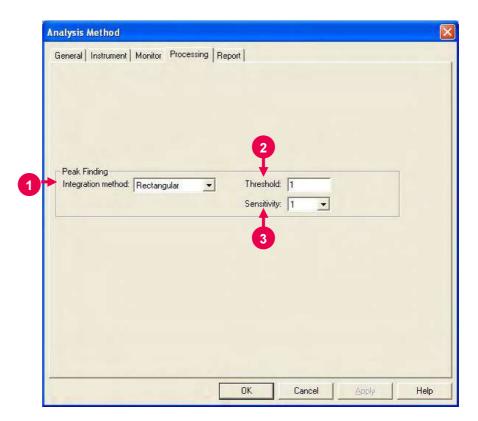


Fig. 6-6 Processing Tab

- 1 Peak Finding: Integration method
  Select an integration method for the peaks to be displayed in the data processing window.
- Threshold Set a detection limit for the photometric value axis of peak and valley. Peaks or valleys below the setting will not be detected.
- 3 Sensitivity Select the number of data points in the horizontal axis direction (abscissa). For detecting sharp peaks, select a sensitivity value of "1." Or select "8" for detecting broad peaks.

# 6.2.5 Report Tab

Select the Report tab, and the window in Fig. 6-7 will appear. For the settable range of each parameter, refer to Appendix G.

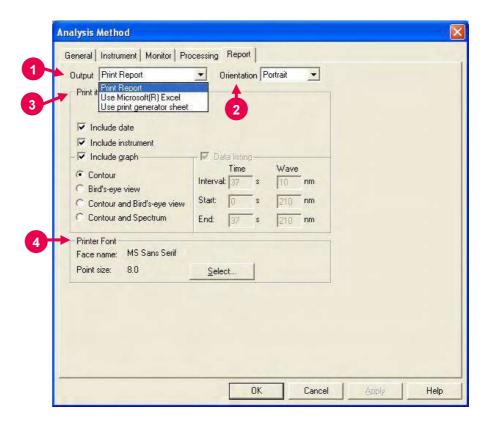


Fig. 6-7 Report Tab

## 1 Output

- Print Report
   A report will be printed. For printing method, refer to 6.9.
- Use Microsoft(R) Excel
   Data will be transferred to Microsoft<sup>(R)</sup> Excel. For transferring method, refer to 7.13.
- Use print generator sheet
   Select this method for using the report generator (option).
   For usage, refer to the instruction manual of the report generator.

## 2 Orientation

Selectable when Print Report is chosen for Output.

Select a printing orientation. Either Portrait or Landscape orientation is selectable. When the Landscape orientation is selected, date and method can be printed.

# 3 Print items (printable/transferable items)

- Include date
- Include instrument
- Include graph

When you put a check mark for this item, any of the graphs listed below is selectable.

- ♦ Contour
- ♦ Bird's-eye view
- ♦ Contour and Bird's-eye view
- ♦ Contour and Spectrum
- Data listing
  - ♦ Interval
  - ♦ Start
  - ♦ End

These parameters are settable when "Use Microsoft(R) Excel" or "Use print generator sheet" is specified for Output. For time scan, make entries for Interval, Start and End. For wavelength scan, make entries for Interval, Start and End.

#### 4 Printer Font

Click the **Select** button for changing the font to be used in the report.

# 6.3 Defining Your Samples

A sample must be defined in the following procedure before measurement.

(1) Click the (sample name/comment input) button on the Measurement toolbar. The window in Fig. 6-8 will appear.

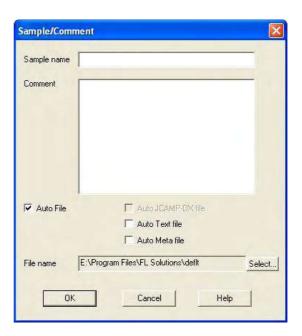


Fig. 6-8

(2) Make entries for Sample name and Comment.Maximum number of enterable characters (half-size)

Sample name: 40 characters

Comment: 255 characters

For automatic saving, put a check mark in the "Auto File" box to specify a file name. First click the **Select** button, and then specify a saving location and enter a file name.

When you put a check mark at each head of "Auto Text file" and "Auto Meta file" after turning on "Auto File," 3-D time scan data can be saved automatically in the following file formats as soon as this data is automatically saved.

- ASCII text file (\*.txt)
- Graph meta file (\*.wmf)

The data will be saved in the file name entered for "File name."

(3) After completion of setting, click **OK**.

# 6.4 Conducting Measurement

- (1) Make sure an analysis method has been set and a sample has been defined. (Refer to "6.2 Creating an Analysis Method" and "6.3 Defining Your Samples.")
- (2) Set a sample in the sample compartment and click the button on the Measurement toolbar to start measurement. (See ① in Fig. 6-9.)

  On this monitor window, the X and Y axes stand for the EM-side wavelength and time (s), respectively.

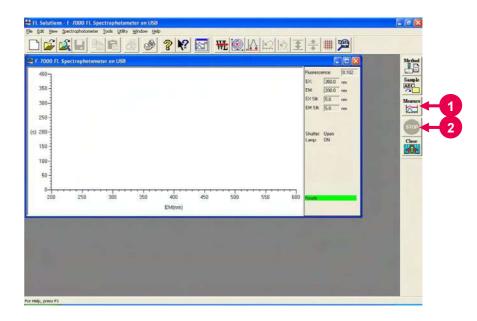


Fig. 6-9 Monitor Window

- (3) For halting the measurement midway, click the button on the toolbar. (See ② in Fig. 6-9.)
- (4) When measurement is completed, the data processing window appears as shown in Fig. 6-10. This is available only when a check mark is put for "Open data processing window after data acquisition" at the 'Monitor' tab of Analysis Method. (See ④ in Fig. 6-5.)

The measured data will be saved automatically when auto saving is selected.

If auto saving is not selected, select the "Save As" command from the File menu and save the data. Opening the saved data file will display the data processing window as shown below.

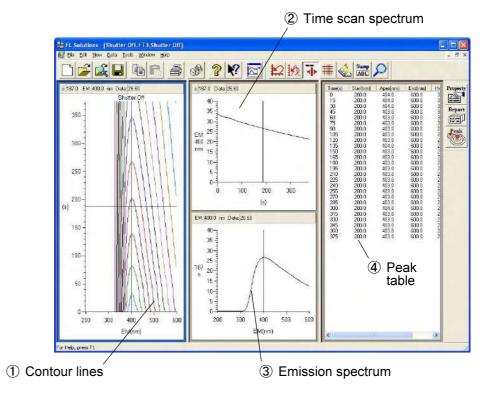


Fig. 6-10 Data Processing Window (3-D time scan)

### Contour lines

From these contour lines, the time-scan spectrum and emission, excitation or synchronous spectrum can be read out.

As the cursor is moved, the cursor-specified time scan spectrum and emission, excitation or synchronous spectrum are displayed in ② and ③.

# 2 Time scan spectrum

A time scan spectrum at the cursor position in ① is displayed.

Wavelength and photometric value can be read by selecting the "trace" command from the View menu or clicking the button

3 Emission spectrum (excitation spectrum, synchronous spectrum) An emission spectrum (excitation spectrum, synchronous spectrum) at the cursor position in ① is displayed. Wavelength and photometric value can be read by selecting the "trace" command from the View menu or clicking the

#### (4) Peak table

Peak wavelength and peak height are indicated. For setting threshold and sensitivity values, click the button.

- NOTICE: 1. The data measured under the following condition will be saved automatically in Program Files\FL Solutions\Data. (Example: C:Program Files\FL Solutions\Data)
  - (1) Measurement was carried out with "Auto File" specified and a file name unspecified.
  - The data measured under the following conditions will be saved automatically in Program Files\FL Solutions. (Example: C:Program Files\FL Solutions)
    - (2) There is not an adequate free area in the specified storage medium (such as floppy disk or MO).
    - (3) Writing is inhibited.

A file automatically saved under such a condition as above is named according to the following rule.

- Rule 1: When a file name is given to analysis method: File name of analysis method + serial No. (001, 002, ...)
- Rule 2: When a file name is not given to analysis method: DEFLT + serial No. (001, 002, ...)

# 6.5 Displaying Data in a Bird's-Eye View

"3-D Time scan" data can be displayed in a bird's-eye view.

Select the (bird's-eye view) button on the Spectrum toolbar.

(Or select the bird's-eye view command from the View menu.)

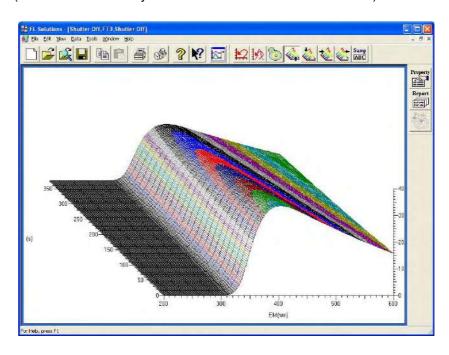


Fig. 6-11

Viewing direction is changeable by clicking (from front), (from rear), (from left) or (from right).

Clicking (contour line diagram) displays a contour line diagram.

# 6.6 Contour Scale Expansion with Mouse

In the contour display of 3-D time scan data, display scale can be expanded with the mouse. On the Spectrum toolbar, click the (contour scale expansion) button. The button is set in the clicked status.

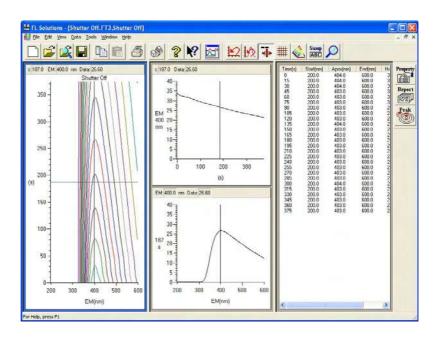


Fig. 6-12

Move the mouse cursor onto the contour as shown in Fig. 6-13 and drag the mouse along an area to be expanded with the left mouse button held down.

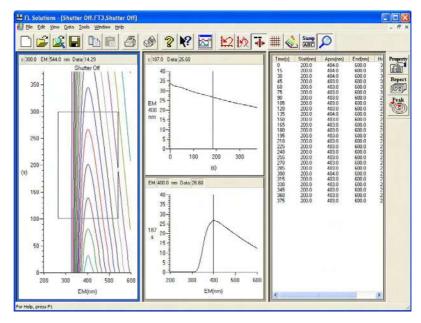


Fig. 6-13

The area enclosed with the mouse cursor is now expanded as shown in Fig. 6-14. For restoring the original display, click the reset) button. (scale

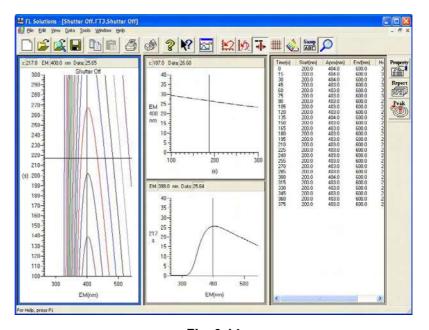


Fig. 6-14

For using the trace cursor on the contour window, click again for returning to the non-clicked status.

# 6.7 Auto Scale Display of Data

When you click the (auto scale) button on the data processing window after completion of measurement or from which the 3-D time scan data file has been read out, the contour window, excitation spectrum window and emission spectrum window are displayed in the auto scale mode as shown in Fig. 6-16.

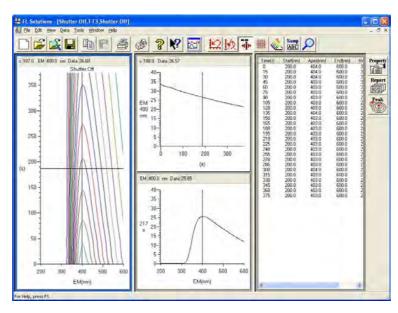


Fig. 6-15 (Before execution of auto scale)

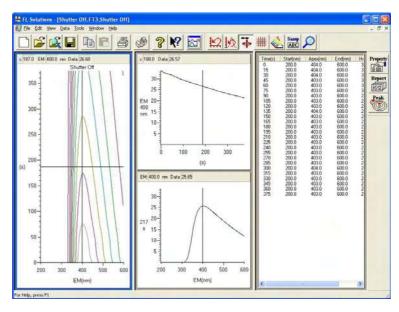


Fig. 6-16 (After execution of auto scale)

For restoring the original display, click the (scale reset) button.

# 6.8 Linking Trace Cursors from Peak Table

On the data processing window where a contour and peak table are displayed, you may move the mouse cursor onto the peak table and click the mouse. As a result, each trace cursor on the contour window, excitation spectrum window and emission spectrum window moves to the peak wavelength position.

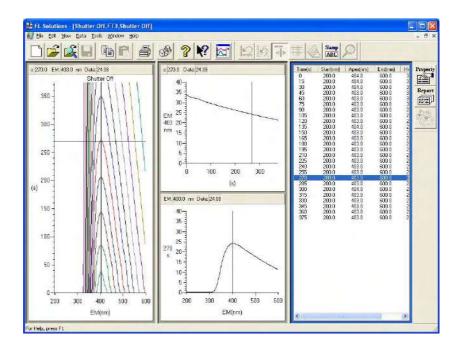


Fig. 6-17

#### 6.9 Data Printout

Printout can be provided even when the function for automatic printout after measurement is not used. The following two procedures are available.

(1) Click on the spectrum window, and the circumference turns blue. In this status, select the Print command from the File menu.

Or click the (print) button on the toolbar. The field active on the data processing window (e.g., 1) in Fig. 6-10) will be printed. What is displayed by clicking the Print Preview command in the File menu will be printed.

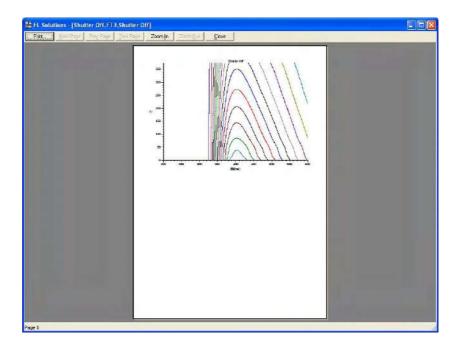


Fig. 6-18

(2) Select the Report command in the Data menu. Or click the button on the toolbar. The Print Preview window will appear as shown below, so click **Print**. Printout items should be selected at the Report tab in Spectrum Properties beforehand.

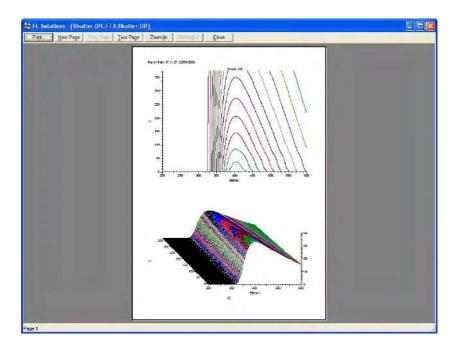


Fig. 6-19

NOTE: If characters overflow from the print frame in repeat measurement or in overlaying, then change the printing orientation from vertical (Portrait) to horizontal (Landscape) at the Report tab in Fig. 6-20.

Select the items to be printed in the following procedure.

Click the button, and the Spectrum Properties window will appear.

Select the Report tab, and the window in Fig. 6-20 will open.

In this window, select desired print items.

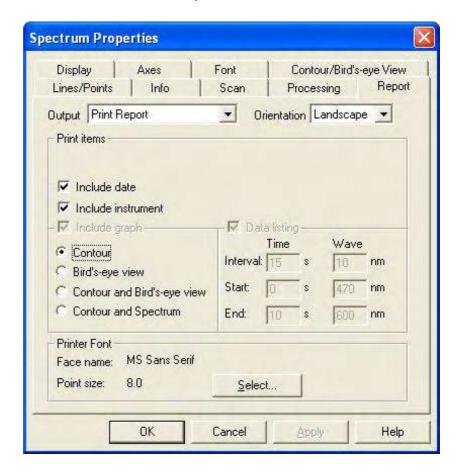


Fig. 6-20

## 7. DATA PROCESSING

This section explains the operation procedures for data processing after measurement.

# 7.1 Loading of Data

Load saved data in the following procedure.

(1) Click the (open) button on the Standard toolbar. A window as in Fig. 7-1 will appear.

(Or select the Open command from the File menu.)

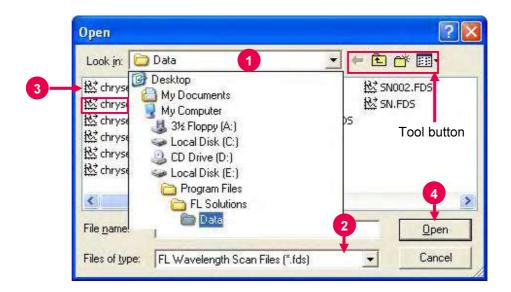


Fig. 7-1

(2) The window (Fig. 7-1) has four tool buttons. Table 7-1 lists the tool buttons and their functions.

Table 7-1 Tool Buttons in Open Window

Tool Button	Function	Tool Button	Function
<b>—</b>	Moves to the previously displayed folder.		Creates a new folder.
£	Displays a hierarchical folder at the next higher level.	<b>III</b> ▼	Lists the following display menu items: Thumbnails, Tiles, Icons, List and Details.

NOTE: A file cannot be opened newly when the remaining memory capacity of PC in operation is very small (the system resource is less than 20%). Either close the presently opened data processing window or restart the FL Solutions program.

- (3) Specify a folder containing a file to be loaded in "Look in."

  For example, specify Program Files ⇒ FL Solutions ⇒ **Data** in the local disk. (Refer to ① in Fig. 7-1.)
- (4) Select a file type in "Files of type."

The following file types are selectable.

- Wavelength scan file (\*.fds)
- Time scan file (\*.fdt)
- Photometry file (\*.fdq)
- 3-D scan file (\*.fd3)
- 3-D time scan file (\*.ft3)
- Analysis method file (\*.flm)
- Sample table file (\*.fls)
- Report file (\*.flr)
- All files (\*.\*)

The files of the selected type will be listed. (Refer to ② in Fig. 7-1.)

(5) Select a file. The selected file is highlighted in blue. (Refer to③ in Fig. 7-1.)

The following two methods are available for selecting multiple files.

Method 1: Click the first file and select the subsequent ones with the [↓] button while holding down the Shift key.

Method 2: Click each file while holding down the Ctrl key.

(6) After completion of selection, click **Open**. (Refer to ④ in Fig. 7-1.) The selected file will open as shown in Fig. 7-2.

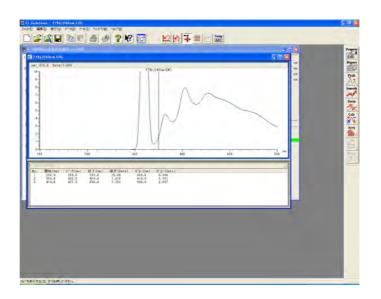


Fig. 7-2

# 7.2 Spectrum Tracing

This function is used to read the photometric value at a certain wavelength on the measured spectrum.

For spectrum tracing, it is required to bring the trace cursor onto the displayed spectrum in the following two ways.

(1) Click the (trace cursor) button on the Spectrum toolbar.
A trace cursor appears on the spectrum. The wavelength and photometric value at the cursor position are indicated at the upper left in Fig. 7-3.

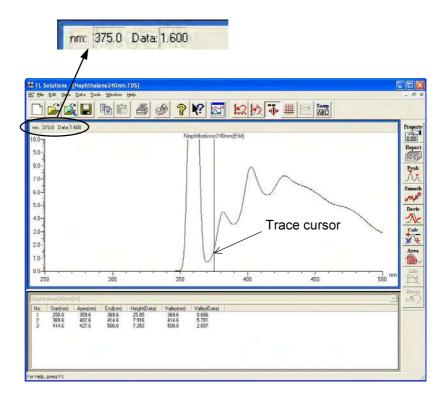


Fig. 7-3

(2) Bring the mouse pointer onto the displayed spectrum and rightclick the mouse. A menu box as in Fig. 7-4 will appear. Select "Trace" from the menu box.

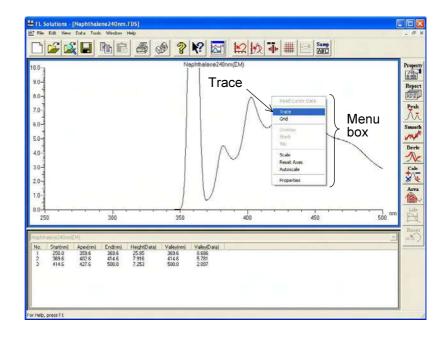


Fig. 7-4

- (3) The trace cursor can be moved in the following two ways.
  - Moving by mouse Bring the mouse pointer to the trace cursor position and move the cursor to a position for readout with the mouse while holding down its left button.
  - ② Moving by arrow keys on keyboard By clicking the [←] or [→] key, the trace cursor moves in the corresponding direction.

# 7.3 Scale Change

You can change the X and Y-axis scales of data.

For scale change, the following three methods are available.

#### 7.3.1 Change by Using Axes Tab

Click the (properties) button on the Processing toolbar.

The Spectrum Properties window will open.

Click the Axes tab. A tab page as in Fig. 7-5 will appear.

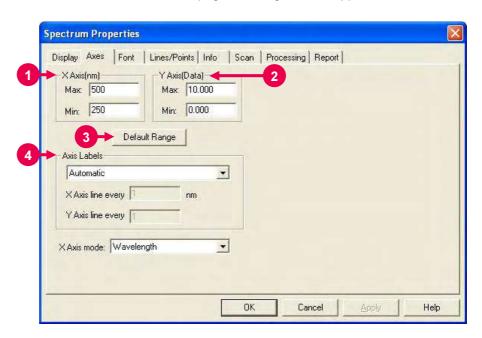


Fig. 7-5 Axes Tab

Enter numerical values in the Maximum and Minimum input boxes for the X and Y axes. The default values are those saved most recently. The above page contains the following setting items.

- 1 X Axis Enter minimum and maximum values for the X axis.
- Y Axis
  Enter minimum and maximum values for the Y axis.
- 3 Default Range This button returns the X and Y axes to the default values.
- 4 Axis Labels Make your selection for the X and Y axis labels from the following three: Automatic, Label high, low, and medium values and Use fixed increment.

# 7.3.2 Change by Right-clicking Mouse

Right-click the mouse on the spectrum window. A menu box as in Fig. 7-6 will appear. Select "Scale" from the menu box.

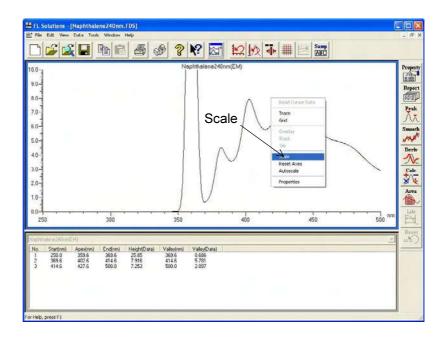


Fig. 7-6

A window as in Fig. 7-7 will appear. The window contains the same setting items as in 7.3.1.

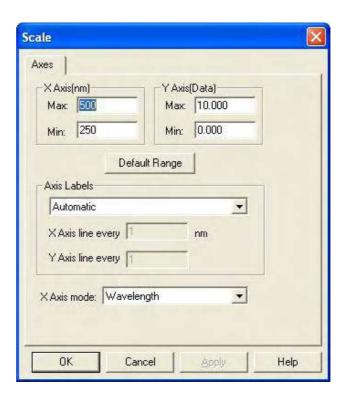


Fig. 7-7

# 7.3.3 Change by Using Left Mouse Button

- (1) The scale can be expanded with the left button of the mouse. Bring the trace cursor onto the spectrum window (Fig. 7-3).
- (2) Specify an area to be expanded. Draw a rectangle while holding down the left mouse button, and then release the button. The area will be expanded.

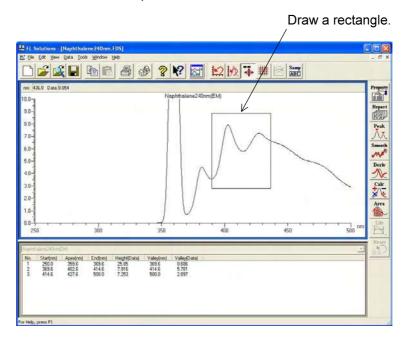


Fig. 7-8

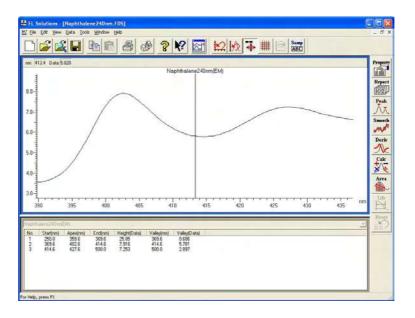


Fig. 7-9

(3) For returning to the original scale, click the button.

# 7.4 Change of Graph Axis

The X axis of a graph can be changed on a spectrum.

Click the (properties) button and select the Axes tab.

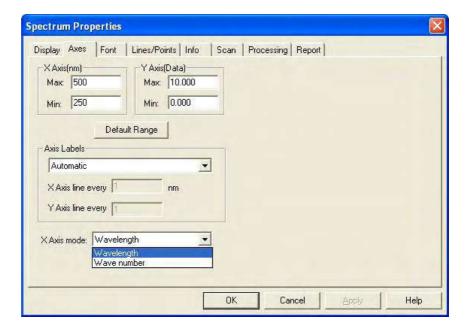


Fig. 7-10 Axes Tab

- X Axis mode
   The X axis is indicated in either unit below.
  - Wavelength Indicated in wavelength (nm).
  - Wave number
     Indicated in wavenumber (Kcm<sup>-1</sup>, K = 1000)
     x·λ = 10000 (x: Kcm<sup>-1</sup>, λ: nm)

# 7.5 Change of Spectral Line

You can change the line pattern and color of a spectrum. Select the Lines/Points tab in the Spectrum Properties window. A tab page as in Fig. 7-11 will appear.

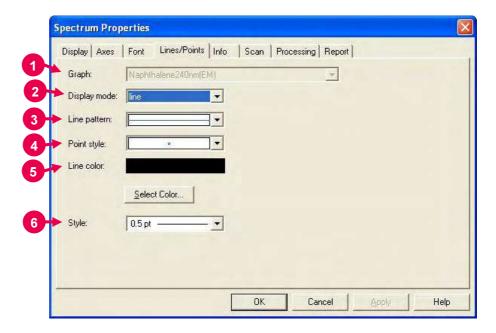


Fig. 7-11 Lines/Points Tab

The above page contains the following setting items.

- 1 Graph
  - Select a spectrum for which you want to set a line pattern.
- 2 Display mode

Data can be displayed in the following modes.

- line
- point
- line & point
- 3 Line pattern

Specify a line pattern such as dotted line or solid line.

4 Point style

Select a point style to be used for printing or displaying a graph.

#### 5 Line color

Upon clicking **Select Color**, the Color window appears.

This window allows you to change the color of spectral line and point. There are 48 basic colors. By clicking **Define Custom Colors**, a smaller window is added on the right side of the Color window as shown in Fig. 7-12. You can now add 16 colors.

These colors are usable for the spectral lines and points.

For defining a color, you can enter values for Hue, Sat (saturation), Lum (luminance), Red, Green and Blue.

Upon completion of the definition, click **Add to Custom Colors**.

Then select a desired color from the defined ones.

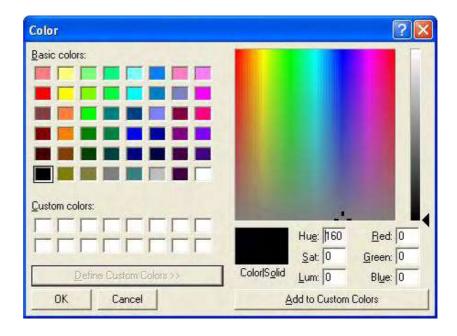


Fig. 7-12

### 6 Style

Specify a line thickness.

# 7.6 Detection of Spectral Peaks

The spectral peaks and valleys are detectable automatically.

Click the (peak detection) button on the Processing toolbar. A window as in Fig. 7-13 will appear. Set an integration method, threshold and sensitivity.

(Or select the Peak Finding Parameters command from the Data menu.)

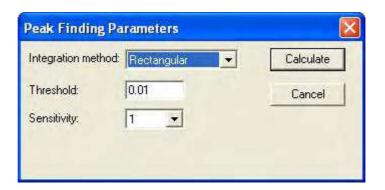


Fig. 7-13

#### 1 Integration method

The following three methods are available and any one may be selected for the peak wavelength. But the area calculation result indicated in the peak table will vary with the selected method.

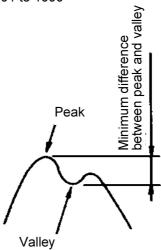
- Rectangular
- Trapezoid
- Romberg

For details of each method, refer to APPENDIX D.

#### 2 Threshold

Set a detection limit of peak and valley (a difference between the peak and valley) on the photometric value axis. Peaks and valleys below the set value will not be detected.

Input range: 0.001 to 1000



#### 3 Sensitivity

Select the number of data points in the X-axis direction. For detection of sharp peaks, select a sensitivity level of "1," and for broad peaks, select "8."

- 1
- 2
- 4
- 8

# 7.7 Indication of Peak Information on Spectrum

Click the (properties) button on the Processing toolbar and select the Display tab.

(Or select the Spectrum Properties command from the Edit menu.)

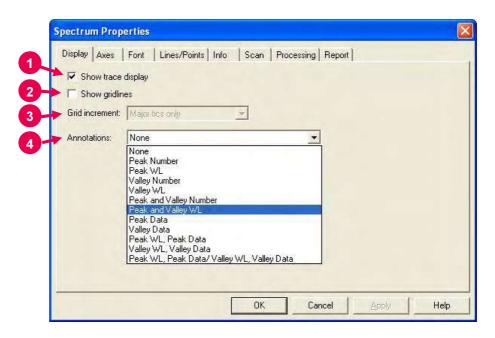


Fig. 7-14 Display Tab

#### 1 Show trace display

By putting a check mark here, the trace cursor is displayed to enable tracing when the data processing window is opened.

#### 2 Show gridlines

By putting a check mark here, the monitor window and data processing window can be displayed with gridlines.

#### 3 Grid increment

Specify a gridline width. This parameter becomes valid when a check mark is put at "Show gridlines."

#### 4 Annotations

Peak information can be indicated on a spectrum. Select items to be displayed on a spectrum and click **OK** or **Apply**.

- None
  - Only a spectrum is displayed.
- Peak Number

Peak number is indicated on a spectrum.

Peak WL

Peak wavelength is indicated on a spectrum.

Valley Number

Valley number is indicated on a spectrum.

Valley WL

Valley wavelength is indicated on a spectrum.

• Peak and Valley Number

Peak number and valley number are indicated on a spectrum.

• Peak and Valley WL

Peak wavelength and valley wavelength are indicated on a spectrum.

Peak Data

Peak data is indicated on a spectrum.

Valley Data

Valley data is indicated on a spectrum.

• Peak WL, Peak Data

Peak wavelength and peak data are indicated on a spectrum.

• Valley WL, Valley Data

Valley wavelength and valley data are indicated on a spectrum.

Peak WL, Peak Data/Valley WL, Valley Data
 Peak wavelength, peak data, valley wavelength and valley data are indicated on a spectrum.

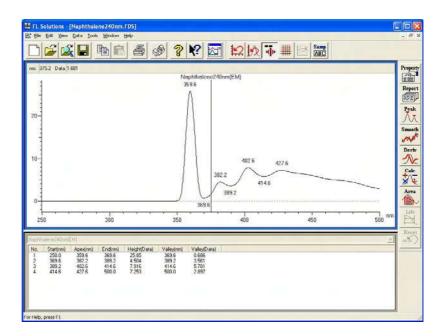


Fig. 7-15 Example of Indication of Peak Wavelength

#### 7.8 **Smoothing of Spectrum**

(smoothing) button on the Processing toolbar.

A window as in Fig. 7-16 will open.

(Or select the Smoothing Parameters command from the Data menu.)



Fig. 7-16

Set the following parameters in the above window.

#### 1 Function type

Select a smoothing method from the following three. For details, refer to APPENDIX D.

- Savitgky-Golay
- Mean
- Median

#### Smoothing order

Set a smoothing order.

Input range: 2 to 4

#### Number of points

Set the number of points to be used in calculation.

Input range: 5 to 101, Initial value: 7

#### Number of times

Set the number of smoothing operations.

Input range: 1 to 100

#### Calculate

This button executes smoothing.

For returning to the original status (before calculation), click the (reset) button.



#### 7.9 **Derivative Operation on Spectrum**

(derivative) button on the Processing toolbar. A window as in Fig. 7-17 will open.

(Or select the Derivative Parameters command from the Data menu.)

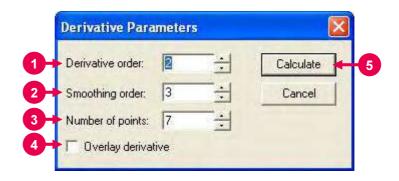


Fig. 7-17

Derivative order Set a derivative order. Input range: 1 to 4

Smoothing order Set a smoothing order. Input range: 2 to 4

Number of points Set the number of data points to be used in calculation. Input range: 5 to 101, Initial value: 15

- Overlay derivative Spectra before and after derivative operation are overlaid for display. Note that the value on the Y axis is not the one after derivative operation.
- Calculate This button executes derivative operation.

Fro returning to the original status (before calculation), click the (reset) button.



# 7.10 Overlaid Display of Spectra

This function is usable for comparison of two or more spectra and other purposes.

The following three methods are available.

# 7.10.1 Insertion from Window

This method is available when two or more spectrum windows are already displayed.

- (1) Select the Overlay command from the Data menu.
- (2) A dialog box as in Fig. 7-18 will appear. The dialog box indicates a sample name for the presently displayed spectrum.



Fig. 7-18

(3) Click **From the windows**. A window as in Fig. 7-19 will appear. (Refer to ① in Fig. 7-18.)

A list of the sample names for the presently displayed spectra is indicated in the left box, and a list of the overlaid sample names in the right box.



Fig. 7-19

- (4) Select a sample name from the left box. The selected sample name will be highlighted in blue. (Refer to ② in Fig. 7-20.)
- (5) Click the **Insert >>** button. (Refer to ③ in Fig. 7-20.)

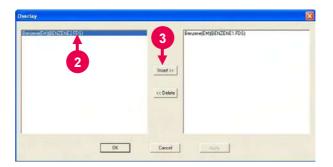


Fig. 7-20

- (6) The selected sample name will be added as shown in Fig. 7-21. (Refer to ④ in Fig. 7-21.)
- (7) Click **OK**. (Refer to ⑤ in Fig. 7-21.)

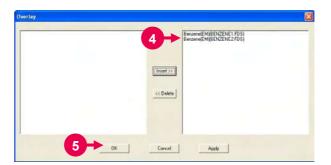


Fig. 7-21

(8) Two spectra will be overlaid on the data processing window. At the same time, a dialog box as in Fig. 7-22 will open. Click Close. (Refer to ⑥ in Fig. 7-22.)



Fig. 7-22

(9) To delete a spectrum, click the corresponding sample name in the above Spectra list. (Refer to ⑦ in Fig. 7-22.) When the clicked sample name is highlighted in blue, the **Delete** button becomes active. Click the **Delete** button. (Refer to ⑧ in Fig. 7-22.)

# 7.10.2 Insertion from File

This method overlays spectra by reading them from the already saved files.

- (1) Select the Overlay command from the Data menu.
- (2) A dialog box as in Fig. 7-23 will appear.

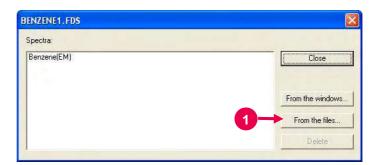


Fig. 7-23

(3) Click **From the files** in Fig. 7-23. (Refer to ① in Fig. 7-23). A window as in Fig. 7-24 will open.

(4) Select a file name for overlaid display and click **Open**. (Refer to ② in Fig. 7-24.)



Fig. 7-24

(5) Two spectra will be overlaid on the data processing window. At the same time, a dialog box as in Fig. 7-25 will appear. Click the **Close** button. (Refer to ③ in Fig. 7-25.)



Fig. 7-25

(6) To delete a spectrum, click the corresponding sample name in the Spectra list. (Refer to ④ in Fig. 7-25.) When the clicked sample name is highlighted in blue, the **Delete** button becomes active. Click the **Delete** button. (Refer to ⑤ in Fig. 7-25.)

#### 7.10.3 Direct Dragand-Drop Operation

This method is available when two kinds of spectrum windows (already processed windows) are displayed on the screen.

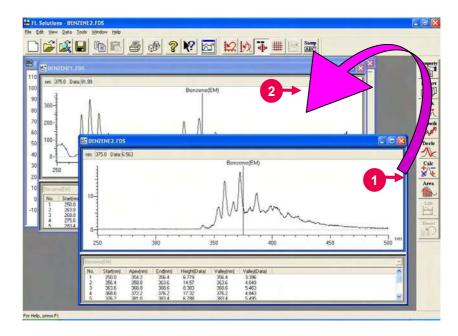


Fig. 7-26

- (1) Drag the blue frame of a spectrum in the following procedure.(Refer to ① in Fig. 7-26.)Move the arrow mouse pointer to the blue frame.
  - Click the left button of the mouse.
  - The mouse pointer will change to  $\mathbf{O}$ .
- (2) While holding down the left mouse button, bring the mouse pointer onto a spectrum to be overlaid. (Refer to ② in Fig. 7-26). When the mouse pointer changes as shown in Fig. 7-27, drop the mouse (release the left mouse button).



Fig. 7-27

Open files as in 7.10.3, click a spectrum to be overlaid and press **Ctrl** + **C** on the keyboard or select the Copy command from the Edit menu. Click the spectrum on which overlaying is desired and paste the first spectrum by **Ctrl** + **V** to complete overlaying.

# 7.10.4 Selection of Spectra from Overlay Window

This method allows you to display only the spectra selected from overlaid ones.

- (1) Click the (display spectra) button on the Spectrum toolbar. A window as in Fig. 7-28 will appear.
- (2) Select a desired sample name from the Visible Spectra list. The selected sample name will be highlighted in blue. (Refer to ① in Fig. 7-28.) (Multiple spectra are selectable by clicking the left button of the mouse on the desired sample names while holding down the Shift key.)
- (3) Click the **OK** button. (Refer to ② in Fig. 7-28.)
- (4) Only the selected spectra will be displayed on the spectrum window. In this example, spectrum No. 4 will be displayed. This spectrum is not printed, but the sample name, analysis method, etc. are printed.

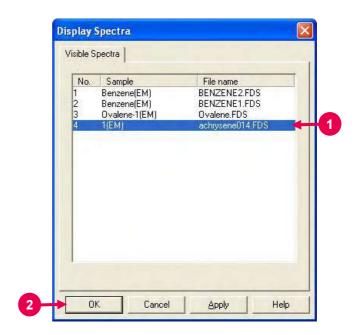


Fig. 7-28

# 7.11 Arithmetic Operation between Spectra

Shown here is an example of obtaining the difference between two overlaid spectra.

Click the (spectral calculation) button on the Processing toolbar.
The Spectral Arithmetic window will open as shown in Fig. 7-29.
(Or select the Spectral Arithmetic command from the Data menu.)

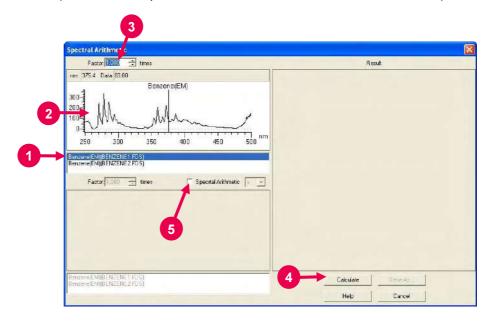


Fig. 7-29

- (1) The sample names for the files opened in the window (file names in parentheses) are listed. From this list, select a file for arithmetic operation. (Refer to ① in Fig. 7-29.)
- (2) The spectrum selected in (1) is displayed. (Refer to ② in Fig. 7-29.)
- (3) Set a factor for the spectrum selected in (1). (Refer to ③ in Fig. 7-29.)
- (4) For multiplying the spectrum by the factor, click **Calculate**. (Refer to ④ in Fig. 7-29.)
- (5) For arithmetic operation between two spectra, put a check mark in the Spectra Arithmetic check box. (Refer to ⑤ in Fig. 7-29.)

- (6) The sample names for the files opened in the window (file names in parentheses) are listed. From this list, select another file for arithmetic operation. (Refer to ⑥ in Fig. 7-30.)
- (7) The spectrum selected in (6) is displayed. (Refer to  $\widehat{\mathcal{T}}$  in Fig. 7-30.)

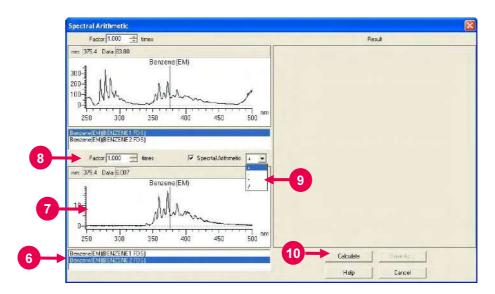


Fig. 7-30

- (8) Set a factor for the spectrum selected in (6). (Refer to ® in Fig. 7-30.)
- (9) Select a mathematical operator for the two spectra. (Refer to 9 in Fig. 7-30.)
  - +: Addition, -: Subtraction, \*: Multiplication, /: Division
- (10) After setting of a mathematical operator, click **Calculate**. (Refer to ① in Fig. 7-30.)

(11) The result of arithmetic operation is displayed as shown in Fig. 7-31. (Refer to ① in Fig. 7-31.)

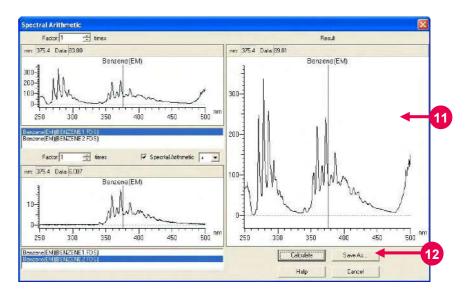


Fig. 7-31 After Arithmetic Operation

(12) Click **Save As**. The result of calculation can be saved in file. (Refer to ① in Fig. 7-31.)

## 7.12 Area Calculation

An area value is obtainable by integration in the specified range of the presently displayed spectrum.

Click the (area calculation) button on the Processing toolbar.

A window as in Fig. 7-32 will appear.

(Or select the Area command from the Data menu.)

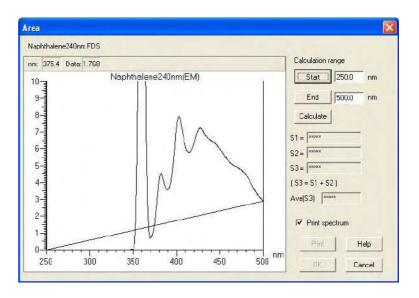


Fig. 7-32

# 7.12.1 Setting of Calculation Range

The range for area calculation is settable in the following two ways.

- (1) Setting by Moving Cursor
  - ① Click the **Start** button. (Refer to ① in Fig. 7-33.)

    Move the trace cursor to a desired start wavelength on the spectrum (refer to ② in Fig. 7-33), and then click the right button of the mouse.
  - ② Click the **End** button. (Refer to ③ in Fig. 7-33.) Move the trace cursor to a desired end wavelength on the spectrum (refer to ④ in Fig. 7-33), and then click the right mouse button.

③ Click the **Calculate** button. The result of calculation will be indicated. (Refer to ⑤ in Fig. 7-33.)

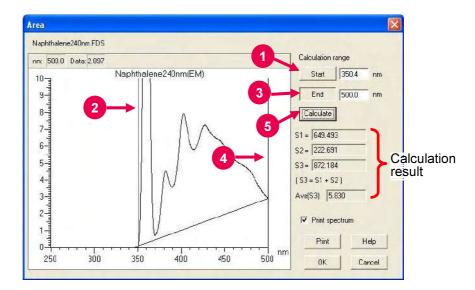


Fig. 7-33

- (2) Setting by Entering Numerical Value
  - ① Enter a desired start wavelength in the Start input box from the keyboard.
  - ② Enter a desired end wavelength in the End input box from the keyboard.
  - 3 Click the Calculate button. The result of calculation will be indicated.

(3) The principle of area calculation is shown below.

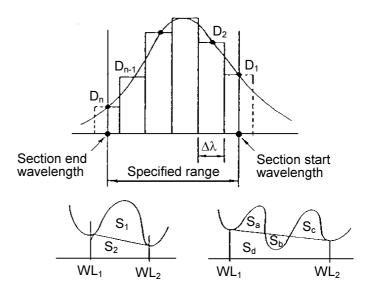


Fig. 7-34 Principle of Calculation

The elements  $(D_1 \text{ to } D_n)$  of a spectrum are subjected to the following calculation for obtaining an area value.

$$\begin{split} S_3 &= S_1 + S_2 = S_a + S_c + S_d \\ &= \{D_2 + D_3 + \cdots D_{n-2} + D_{n-1} + (D_1 + D_n)/2*\Delta\lambda \\ S_2 &= S_b + S_d \\ &= [(D_1 + D_n)*\{\Delta\lambda* (n-1)\}]/2 \\ &= [(D_1 + D_n)*\{section \ start \ wavelength \ (WL_1) - section \ end \ wavelength \ (WL_2)\}]/2 \\ S_1 &= S_3 - S_2 \end{split}$$

 $D_1$ : Spectral data at section start wavelength  $D_n$ : Spectral data at section end wavelength

 $\Delta\lambda$ : Sampling interval

In the time scan mode, change as follows. Start wavelength  $(WL_1) \rightarrow End$  time  $(T_2)$  End wavelength  $(WL_2) \rightarrow Start$  time  $(T_1)$ 

## 7.12.2 Printing of Calculation Result

The result of area calculation can be printed out.

- (1) To print a spectrum and the result of area calculation, put a check mark at "Print spectrum." (Refer to ① in Fig. 7-35.) To avoid printing a spectrum, remove the check mark.
- (2) Click **Print** to start printing. (Refer to ② in Fig. 7-35.)

  Upon execution of printing, the Area window will close.

  The same window can be called up by clicking the calculation) button on the Processing toolbar.

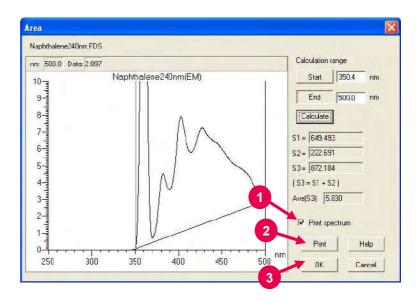


Fig. 7-35

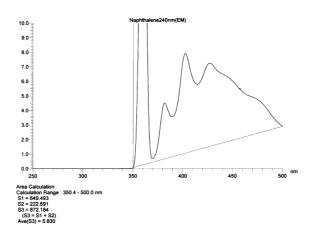


Fig. 7-36 Example of Printout (with spectrum record)

- (3) Upon clicking the **OK** button after calculation (refer to ③ in Fig. 7-35), its result is saved although the Area window is closed.
- (4) The result of calculation is reset by clicking the button on the Processing toolbar with the data processing window displayed. Upon calling up the Area window again, therefore, "\*\*\*\*\*\* will be indicated in each result box.

NOTE: Upon execution of file saving after area calculation, its result is also saved in a file. So a numerical value is indicated in each result box when the Area window is called up. These values cannot be erased by clicking the reset button because they are saved in the computer.

### 7.12.3 Scale Expansion

For expanding the scale in the Area window, use the left button of the mouse.

(1) As shown in Fig. 7-37, specify an area to be expanded by drawing a rectangle with the mouse while holding down its left button on a spectrum.

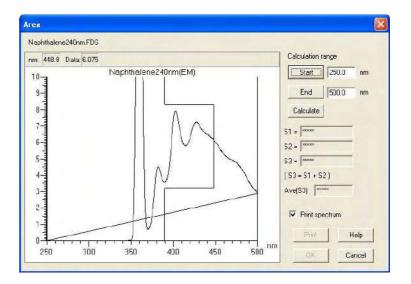


Fig. 7-37

(2) Then release the left button. The specified area will be expanded as shown in Fig. 7-38.

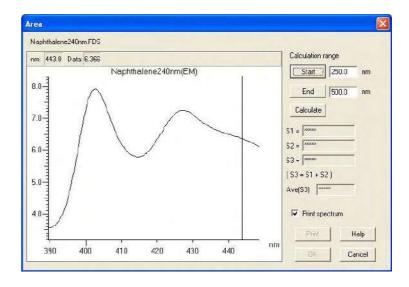


Fig. 7-38

(3) Double clicking the left mouse button on the spectrum returns the scale to the original one.

### 7.13 Transfer of Data to Microsoft(R) Excel

Measured data can be transferred to a worksheet of Microsoft(R) Excel in the following procedure.

(1) Click the (properties) button and select the Report tab.

A tab page as in Fig. 7-39 will open. Select "Use Microsoft(R)

Excel" for Output. Also select data to be transferred (peak table, data listing, etc. in case of wavelength scan). Then click **OK**.

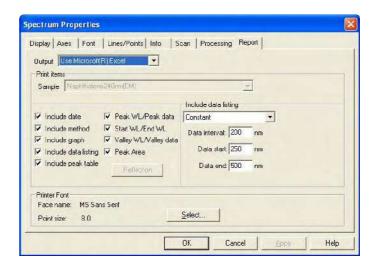


Fig. 7-39

(2) Click the (report) button. The Microsoft(R) Excel will automatically start to read data. The subsequent procedure concerns the Microsoft(R) Excel operation.

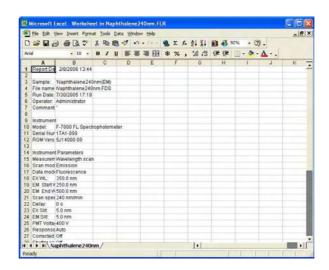


Fig. 7-40

(3) Data can be saved with extension code "xls" of Microsoft(R) Excel. Select **File – Save Copy As** and enter a file name.

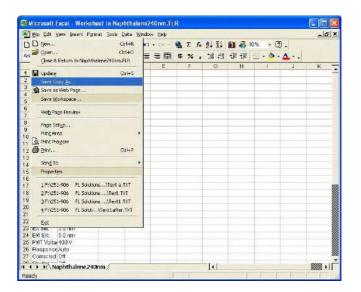


Fig. 7-41

(4) Upon closing the Microsoft(R) Excel, a window as in Fig. 7-42 appears. Now close this window. The original data processing window will appear again.

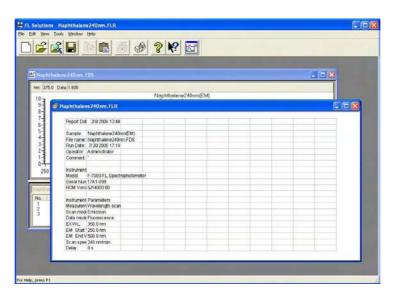


Fig. 7-42

### 7.14 Pasting of Spectrum to Microsoft(R) Word

A spectrum can be pasted to a Microsoft(R) Word text as it is. Take the following procedure.

- (1) Make spectral data active by clicking the relevant spectrum in the data processing window. The thick frame will change to blue.
- (2) Select the Copy command from the Edit menu.
- (3) Open a Microsoft(R) Word text and click a position for pasting. By clicking the Paste command from the Edit menu, the spectrum can be pasted to the Microsoft(R) Word text.

Peak tables and/or kinetics data can also be pasted to a Microsoft(R) Word text in the same way. In this case, they are pasted as a text. By double-clicking the pasted spectrum, functions such as scale change and gridline indication are applicable.

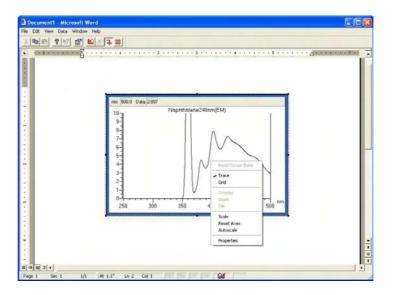


Fig. 7-43 Spectrum Pasted to Microsoft(R) Word

Upon right-clicking the pasted spectrum, a menu box as in Fig. 7-48 will appear to enable scale change, etc. Any change made here is brought into a Microsoft(R) Word text.

**NOTE**: Editing in the Microsoft(R) Word can be made with a PC in which the Microsoft(R) Word and FL Solutions program are installed.

### 7.15 Pasting of Spectrum to Microsoft(R) Excel

A spectrum can be pasted to the Microsoft(R) Excel as it is. Take the following procedure.

- (1) Make spectral data active by clicking the relevant spectrum on the data processing window. The thick frame will change to blue.
- (2) Select the Copy command from the Edit menu.
- (3) Open the Microsoft(R) Excel and select "Paste Special" from its Edit menu. A dialog box as in Fig. 7-44 will open. Make sure that "FL Wavelength Scan Object" is indicated in "As."

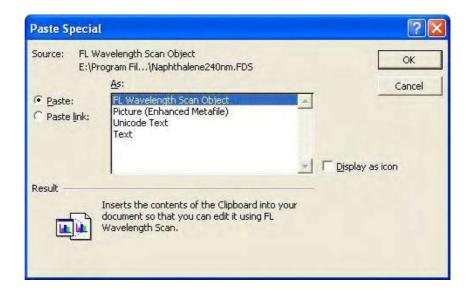


Fig. 7-44

(4) Click **OK**. The spectrum will be pasted to the Microsoft(R) Excel.

Peak tables and kinetics data can also be pasted to the Microsoft(R) Excel in the same way. In this case, they are pasted as data for each cell.

### 7.16 Conversion of Data into ASCII Text File

A file conversion function is provided for data processing with other spreadsheet software. Conversion can be made to the following three data files.

- JCAMP-DX file (\*.dx)
- ASCII text file (\*.txt)
- Windows metafile (\*.wmf)

### **NOTES**: 1. The date/time, analysis method and data list (raw data) is converted into a JCAMP-DX file.

- The items selected on the Report tab page of the Spectrum Properties window are converted into an ASCII text file.
- (1) Load a data file for text conversion and open the data processing window. (Refer to 7.1.)
- (2) Click the Save As command in the File menu.
- (3) A window as in Fig. 7-45 will open.

  Select "ASCII Text Files (\*.txt)" in "Save as type."

For converting a file containing overlaid spectra into an ASCII text, select "ASCII Type 1 files (\*.txt)" or "ASCII Type 2 files (\*.txt)." (For details, refer to 7.17.)

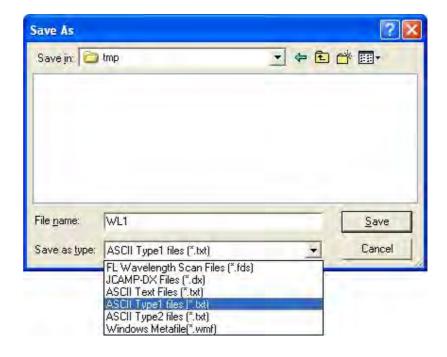


Fig. 7-45

(4) Specify a folder for saving, enter a file name and click **Save**.

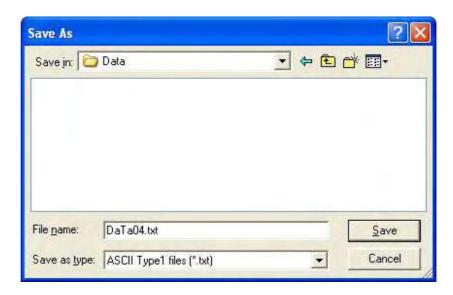


Fig. 7-46

File conversion is effective for a data file in any measurement mode. The selected data is separated by tab.

### 7.17 Conversion of Overlaid Spectra into ASCII File

A file conversion function is provided for processing of overlaid spectral data with other spreadsheet software. Conversion can be made to the following two ASCII formats.

- ASCII type 1 file (\*.txt)
- ASCII type 2 file (\*.txt)

NOTE: The items selected on the Report tab page of the Spectrum
Properties window are converted into an ASCII type 1 file or
ASCII type 2 file. Note that the instrument conditions are not output when the ASCII type 2 file is selected.

- (1) Load a data file for text conversion and open the data processing window. (Refer to 7.1.)
- (2) Click the [Save As command] in the File menu.
- (3) A window as in Fig. 7-47 will open. Select either file below in "Save as type."
  - ASCII Type 1 files (\*.txt)
  - ASCII Type 2 files (\*.txt)

For a single spectrum, select "ASCII type 1 files (\*.txt)."

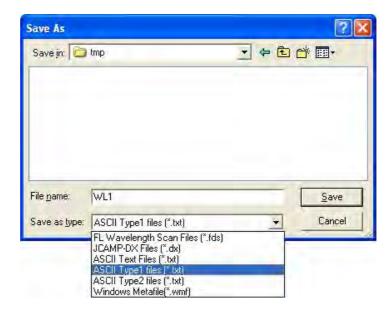


Fig. 7-47

#### **Output Format of ASCII Type 1 file**

For a file containing overlaid spectra, a set of item name and data will be output in two columns.

In reading into the Microsoft(R) Excel, setting is made for each cell in the following format.

15	1st spectral data		2nd spectral data		3rd spectral data	
Ite	em name	Data	Item name	Data	Item name	Data

Figure 7-48 shows an example of output into the Microsoft(R) Excel.

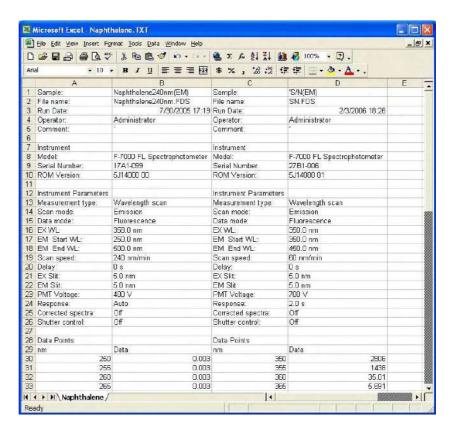


Fig. 7-48

#### **Output Format of ASCII Type 2 file**

For a file containing overlaid spectra, the item name will be output in the leftmost column and spectral data in each column. In reading into the Microsoft(R) Excel, setting is made for each cell in the following format.

Item name	1st spectral data	2nd spectral data	3rd spectral data
-----------	-------------------	-------------------	-------------------

Figure 7-49 shows an example of output into the Microsoft(R) Excel.

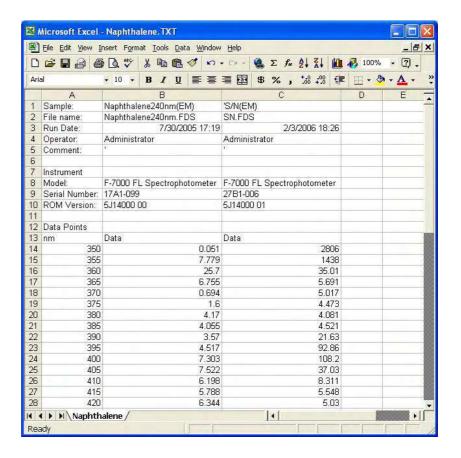


Fig. 7-49

### 7.18 Renaming Your Sample

After measurement, you can change the sample name of saved data. Call up the Info tab page and rename your sample in the following procedure.

- (1) Open a data file for renaming. (Refer to 7.1.)
- (2) Click the ABC (information tab) button on the Spectrum toolbar.
  A dialog box for input of a sample name/comment will appear.
  The dialog box includes the Info tab.
- (3) Or click the (properties) button on the Processing toolbar. The Spectrum Properties window will open. Click the Info tab. A tab page as in Fig. 7-50 will appear.

(You can also open the Spectrum Properties window by selecting the Spectrum Properties command from the Edit menu.)

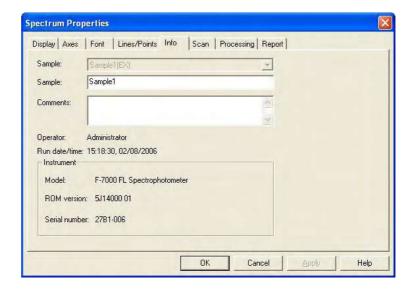


Fig. 7-50

- (4) Input a new sample name in "Sample" and click **OK**.
- (5) To save data under the new sample name, click the button on the Standard toolbar for updating, and select the Save As command from the File menu for saving under a new file name.

### 7.19 Printing of Wavelength and Photometric Values on Display

This program provides a function for reading data such as presently displayed wavelength and photometric values, traced value on the data processing window, etc. The data can also be printed.

## 7.19.1 Reading and Printing of Presently Displayed Wavelength and Photometric Values

Take the following procedure with the monitor window displayed.

(1) Select the Read Data command from the Tools menu. (Refer to ① in Fig. 7-51.)



Fig. 7-51

(2) The Read Data box will appear as shown in Fig. 7-52.Drag the title area (blue) of the box to the lower right. (Refer to② in Fig. 7-52.)

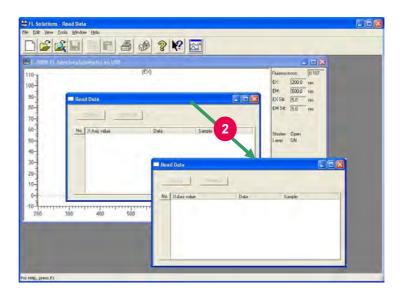


Fig. 7-52

(3) Bring the mouse pointer onto the monitor window and click the right button of the mouse. A menu box will appear. Select "Read Current Data" from the menu box (refer to ③ in Fig. 7-53), and the photometric value and data value will be read into the Read Data box.

Upon start of data reading, Parameter  $\rightarrow$  Scanning  $\rightarrow$  Ready are displayed sequentially in the message display area (refer to ④ in Fig. 7-53).

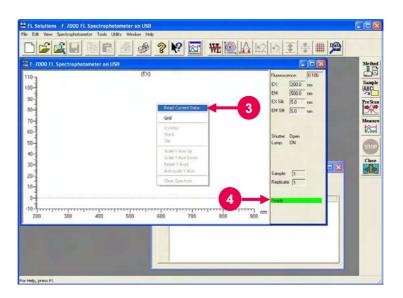


Fig. 7-53

(4) The integration time for data reading is usually set at 0.1 s. To change the integration time, select the Options command from the Tool menu. Then select the Analysis condition tab (refer to ⑤ in Fig. 7-54). A window as in Fig. 7-54 will appear to enable setting change.

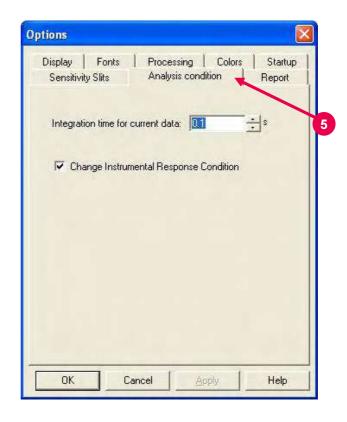


Fig. 7-54

(5) In data reading, sample name information is also indicated.

Sample names can be input. For input, click the sample name column. (Refer to ⑥ in Fig. 7-55.)

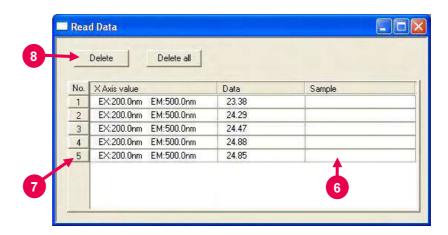


Fig. 7-55

(6) Read data can be deleted. To delete a data value from the table, click the relevant No. (refer to ⑦ in Fig. 7-55). Then click the **Delete** button (refer to ⑧ in Fig. 7-55). A dialog box as in Fig. 7-56 will appear. Click the **Yes** button (refer to ⑨ in Fig. 7-56), and the data value will be deleted.

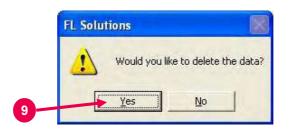


Fig. 7-56

- (7) To delete all data values, click the **Delete all** button.
- (8) For printing the relevant data, click the (print) button on the Standard toolbar.
   (Or select the Print command from the File menu.)

# 7.19.2 Reading and Printing of Wavelength and Photometric Values from Data Processing Window

The wavelength and photometric values can be read and printed from the data processing window.

(1) Open a data file. (Refer to 7.1.)

**NOTE**: Avoid maximizing the opened data file. With this function, two windows are displayed at the same time. If one window is maximized, the other cannot be displayed.

- (2) Select the Read Data command from the Tools menu.
- (3) The Read Data box will appear as shown in Fig. 7-57. Drag the title area (blue) of the box to the lower right. (Refer to ① in Fig. 7-57.)

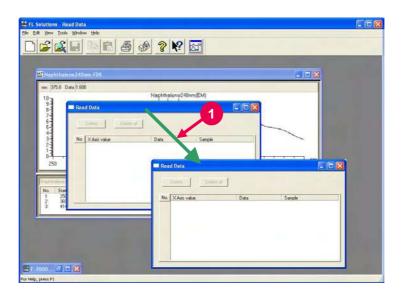


Fig. 7-57

- (4) Click the spectrum display window. (Refer to ② in Fig. 7-58.)
- (5) Click the (trace cursor) button on the Spectrum toolbar. (Refer to ③ in Fig. 7-58.) A trace cursor will appear on the spectrum. (Refer to ④ in Fig. 7-58.)

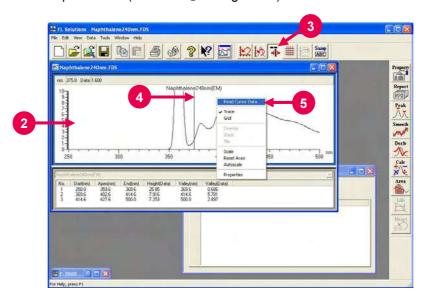


Fig. 7-58

- (6) Move the trace cursor onto a spectrum to be traced and click the right button of the mouse. A menu box will appear. Select "Read Cursor Data" from the menu box (refer to ⑤ in Fig. 7-58), and the wavelength and photometric values will be read into the Read Data box.
- (7) Upon selection of "Read Cursor Data," display becomes as shown in Fig. 7-59.
- (8) In data reading from the data processing window, sample name information is also indicated. Sample names can be edited. For input, click the sample name column. (Refer to ⑥ in Fig. 7-59.)

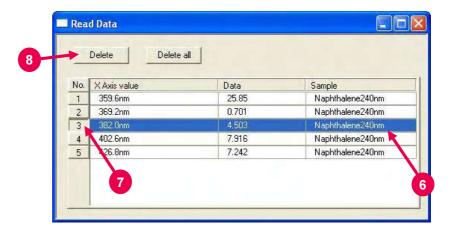


Fig. 7-59

(9) The read data can be deleted. To delete a data value from the table, click the relevant No. (refer to ⑦ in Fig. 7-59). Then click the **Delete** button (refer to ⑧ in Fig. 7-59). A dialog box as in Fig. 7-60 will appear. Click the **Yes** button (refer to ⑨ in Fig. 7-60), and the data value will be deleted.



Fig. 7-60

(10) To delete all data values, click the **Delete all** button.

(11) For printing the relevant data, click the (print) button on the Standard toolbar.

(Or select the Print command from the File menu.)

### 7.20 Reference to Spectrum Measured in Photometry Mode

When the quantification method is peak area, peak height or derivative in the photometry mode, wavelength scan is carried out and according to its result, a calibration curve is prepared. The spectrum measured in this case can be displayed and printed.

(1) Open a data file (photometry file (\*.fdq)). (Refer to 7.1.) A window as in Fig. 7-61 will open.

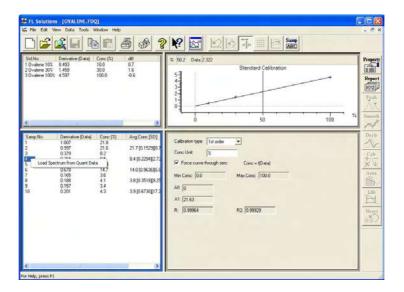


Fig. 7-61

(2) Right-click the numerical part in "Samp. No." in the sample measurement result indication area. (Refer to ① in Fig. 7-62.) The **Load Spectrum from Quant Data** button will appear. (Refer to ② in Fig. 7-62.)

The same operation is possible in the area for standards.

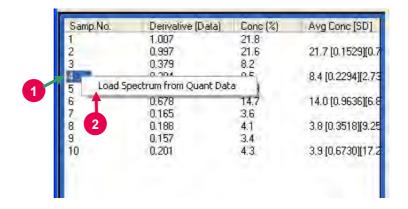


Fig. 7-62

(3) Click the Load Spectrum from Quant Data button. A spectrum window as in Fig. 7-63 will appear.
 (Or click a numeral and select the Load Spectrum from Quant Data command from the Data menu.)

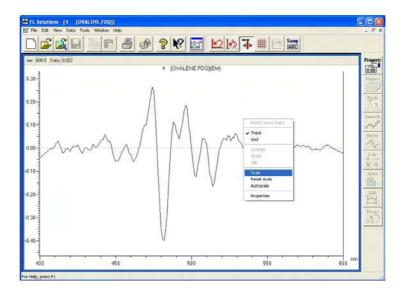


Fig. 7-63

- (4) Upon right-clicking on the displayed spectrum, a menu box will appear. You can also carry out spectrum tracing and scale change.
- (5) The spectrum can be printed by clicking the button (or by selecting the Print command from the File menu).

## 8. MORE CONVENIENT OPERATION (APPLICATION)

### 8.1 Always Starting with Same Analysis Method

The FL Solutions program can be started under the specified analysis method. Note, however, that this function cannot be used unless any analysis method is saved in 8.4.

(1) Select Tools – Options as shown in Fig. 8-1. (Refer to ① in Fig. 8-1.)



Fig. 8-1 Tools Menu

(2) Select the Startup tab (refer to ② in Fig. 8-2), and a tab page as in Fig. 8-2 will appear.

(3) Select "Auto Parameter File" (refer to ③ in Fig. 8-2) and click the **Set F-7000 Method File** button (refer to ④ in Fig. 8-2).

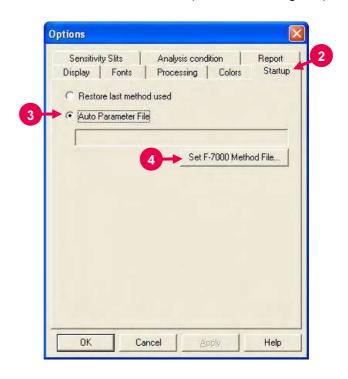


Fig. 8-2 Startup Tab

(4) A window as in Fig. 8-3 will open. Select a desired method file. (Refer to ⑤ in Fig. 8-3.) The extension code of method file is "\*.flm."

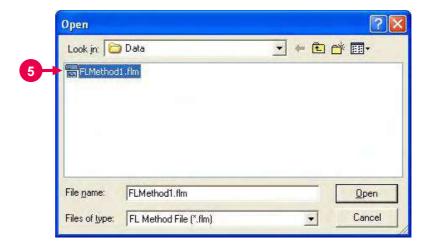


Fig. 8-3

This function is validated from the next startup of the FL Solutions program.

### 8.2 Online/Offline Setting of Spectrophotometer

The spectrophotometer can be set offline. And it can be returned to the online status.

(1) Click the button on the Standard toolbar.
(Or select the Instruments command from the Tools menu.)
The mouse pointer will turn into an hourglass and about 10 seconds later, a window as in Fig. 8-4 will appear. Under this display, the spectrophotometer is connected.

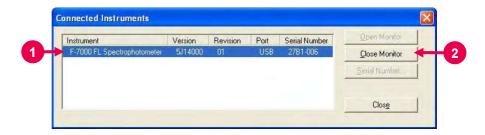


Fig. 8-4

- (2) Click the instrument name on the Connected Instruments window (refer to ① in Fig. 8-4), and the selected line will be highlighted in blue. At the same time, the Close Monitor, Serial Number and Close buttons will become active. Click Serial Number. A serial number will be indicated.
- (3) Upon clicking the **Close Monitor** button (refer to ② in Fig. 8-4), a window as in Fig. 8-5 will open. When you click the **Yes** button (refer to ③ in Fig. 8-5), the offline status is set.



Fig. 8-5

(4) For online setting, click the **Open Monitor** button.Initialization will be executed and the monitor window will appear.

### 8.3 Check of Data File Information

8.3.1 Check of Information on Saved Data

You can check the conditions or date of measurement for saved data.

- (1) Click the (open) button on the Standard toolbar. (Or select the Open command from the File menu.)
  A window as in Fig. 8-6 will appear.
- (2) Specify and right-click a desired file from the displayed file list. (Refer to ① in Fig. 8-6.) A menu box will appear. Select "Properties" from the menu box. (Refer to ② in Fig. 8-6.)

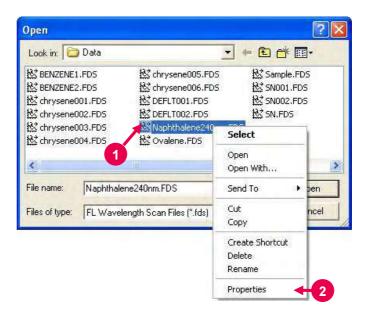


Fig. 8-6

(3) A window as in Fig. 8-7 will appear. Necessary information can be checked by clicking the Info and Scan tabs.

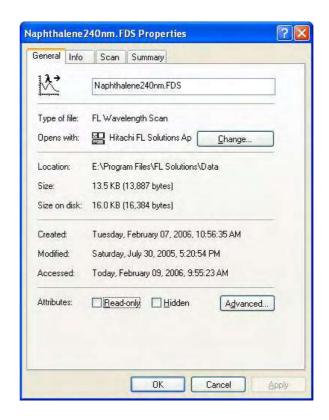


Fig. 8-7

## 8.3.2 Check of Information on Displayed Data

You can check information on presently displayed data.

- (1) Click the (properties) button on the Processing toolbar. (Or select the Spectrum Properties command from the Edit menu.)
- (2) Upon clicking the Info tab (refer to ① in Fig. 8-8), a tab page as in Fig. 8-8 will appear. When you click the Scan tab (refer to ② in Fig. 8-9), a tab page as in Fig. 8-9 will appear.

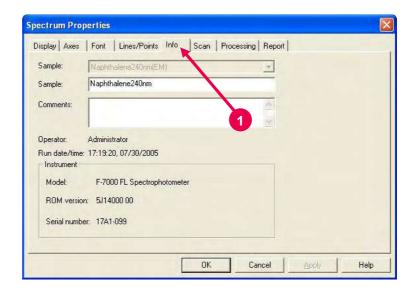


Fig. 8-8 Info Tab

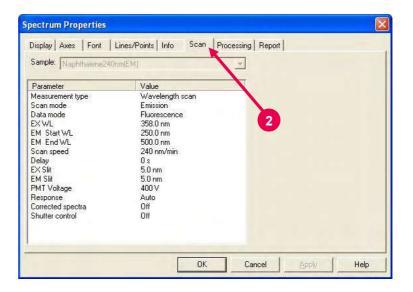


Fig. 8-9 Scan Tab

(3) When multiple spectra are displayed on the presently opened window (recording in overlay mode for example), multiple sample names are indicated by clicking the drop-down list box (refer to 3 in Fig. 8-10). Select a sample name for the data to be checked, and you can check the corresponding information.

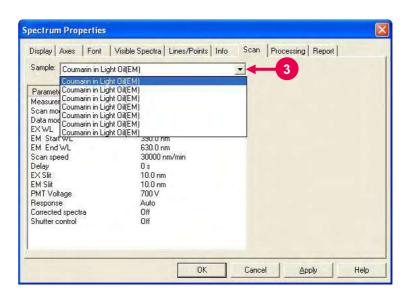


Fig. 8-10

### 8.4 Saving of Analysis Method

The set analysis method can be saved as a file. So instead of setting the method each time, you can just call up the file for setting.

(1) Click the (analysis method) button on the Measurement toolbar.

(Or select the Method command from the Edit menu on the monitor window.)

A window as in Fig. 8-11 will appear.

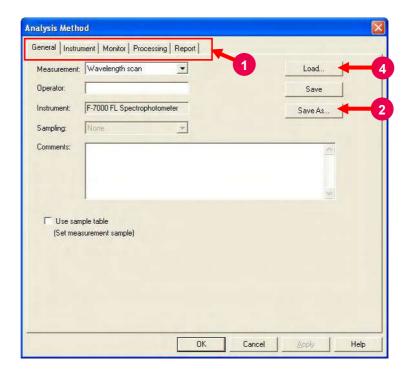


Fig. 8-11

- (2) Select the Instrument tab, Monitor tab, etc. (refer to ① in Fig. 8-11) and set an analysis method, sample name, etc.
- (3) Upon completion of method setting, click the **Save As** button. (Refer to ② in Fig. 8-11.)
- (4) A window as in Fig. 8-12 will appear. Input a file name. (Refer to ③ in Fig. 8-12.) The extension code of method file is "\*.flm."

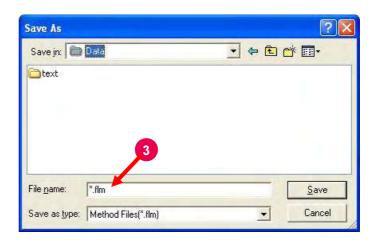


Fig. 8-12

(5) For calling up the saved file, click the **Load** button (refer to ④ in Fig. 8-11) and select a file.

### 8.5 Event Log

If an error occurs in use of the instrument, the details of the error is automatically recorded in an event log. You can also record the date/time of lamp exchange and the occurrence of trouble.

(1) Select the Event Log command from the Tools menu as shown in Fig. 8-13. (Refer to ① in Fig. 8-13.)



Fig. 8-13

- (2) The Event Log tab page will appear as shown in Fig. 8-14. It displays the occurrence time, user, description and computer for communication errors, etc.
- (3) By putting a check mark in the Event or User Notes check box in "View," you can display the corresponding details. (Refer to ② in Fig. 8-14.)

• Event : Display of events and errors

• User Notes : Display of user notes

For displaying only events and errors or only user notes, put a check mark in either check box.

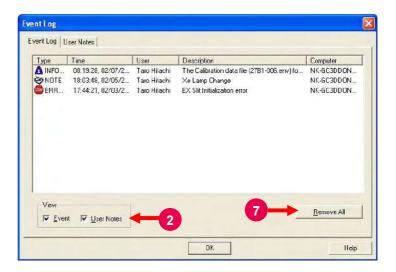


Fig. 8-14 Event Log Tab

(4) For adding a user note, click the User Notes tab. (Refer to ③ in Fig. 8-15.) A tag page as in Fig. 8-15 will appear.

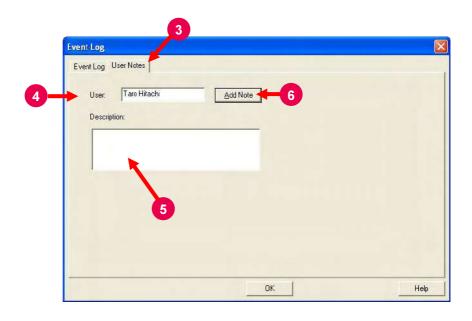


Fig. 8-15 User Notes Tab

- (5) Input a user name. (Refer to ④ in Fig. 8-15.) Enter a note in the Description input box. (Refer to ⑤ in Fig. 8-15.)

  Number of input characters: 127 characters max.
- (6) Click the **Add Note** button. (Refer to **6** in Fig. 8-15.)
- (7) Upon clicking the **Remove All** button, the confirmation message show below will appear.

"Event log will be cleared. Are you sure to execute it?"

By clicking **Yes**, the event log and all of the user information will be cleared. (Refer to ⑦ in Fig. 8-14.)

### 8.6 Check of Program Version

You can check the version of the currently used program. If the instrument develops trouble, inform the service office of the phenomenon and program code.

As shown in Fig. 8-16, select the About FL Solutions command from the Help menu.



Fig. 8-16

A window as in Fig. 8-17 will open. Check the program code.

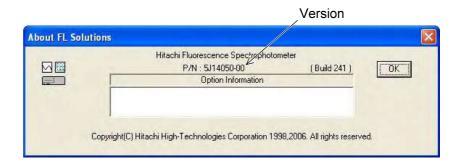


Fig. 8-17

### 8.7 Change of Window Display for Multiple Spectra

Upon clicking the View menu on the menu bar, you can select a method of displaying multiple spectra.





Monitor window

Data processing window

Fig. 8-18 Commands in View Menu

Overlay : Two or more spectra are overlaid for

comparison, etc. (Refer to Fig. 8-19.)

Stack : Spectral are displayed separately in the

vertical direction. (Refer to Fig. 8-20.)

• Tile : Spectra are displayed separately in the vertical

and horizontal direction. For example, 16 spectra are displayed in 4 rows  $\times$  4 columns and 15 spectra in 5 rows  $\times$  3 columns.

(Refer to Fig. 8-21.)

Scale Y Axis Up : Y axis scale is doubled.
Scale Y Axis Down: Y axis scale is halved.

• Reset Y Axes : The scale set under the Monitor tab is taken.

Autoscale Y Axis : The scale is matched with the size of

spectrum.

Reset Axes : The scale set under the Monitor tab is taken.

Autoscale : The scale is matched with the size of

spectrum.

#### Overlay

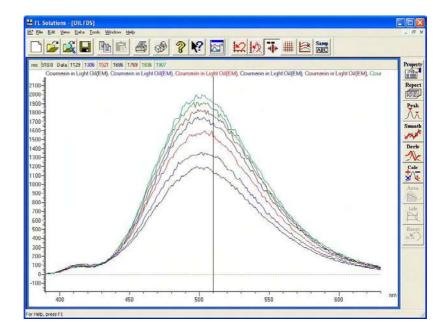


Fig. 8-19

#### Stack

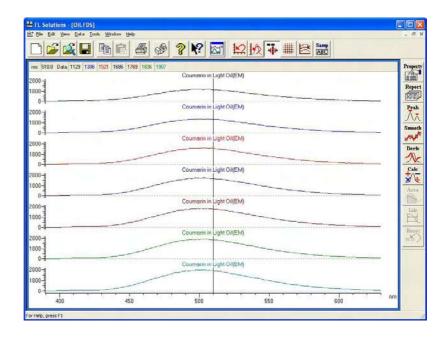


Fig. 8-20

#### Tile

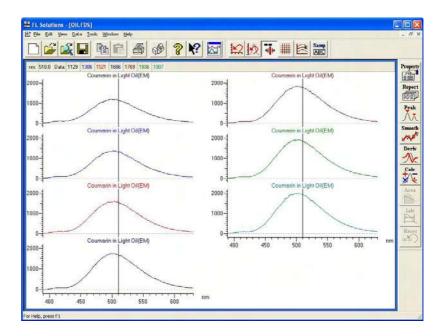


Fig. 8-21

NOTE: When six, seven or more spectra are displayed simultaneously, the size of each spectrum may become excessively small or all the traced values cannot be displayed. In such a case, select the Overlay command from the Data menu to delete unnecessary spectra.

## 8.8 Check of Photometric Value at Specified Wavelength

This is a function for shifting the excitation and emission-side monochromators to the specified wavelengths. It is also usable for checking if the Xe lamp is adjusted properly.

(1) Click the (wavelength drive) button on the Spectrophotometer toolbar.
 (Or open the monitor window and select the Set Wavelength command from the Spectrophotometer menu.)
 A window as in Fig. 8-22 will appear.



Fig. 8-22

(2) Input target wavelength values and click **OK**. Each monochromator will be shifted to the specified wavelength and a photometric value is indicated on the monitor window. (Refer to ② in 1.6.)

#### [Wavelength drive to zero-order light]

For checking if the Xe lamp is adjusted properly or relocating the instrument, the wavelength needs to be driven to the zero-order light. Open the window shown in Fig. 8-22 and set the wavelength at 0 nm. By clicking **OK**, the wavelength is adjusted to the zero-order light.

## 8.9 Collective Printing of Files

Wavelength scan data files (\*.fds), time scan data files (\*.fdt), 3-D scan files (\*.fd3) or 3-D time scan file (\*.ft3) can be collectively printed.

(1) Select the Plot Spectra command from the File menu. A window as in Fig. 8-23 will appear.

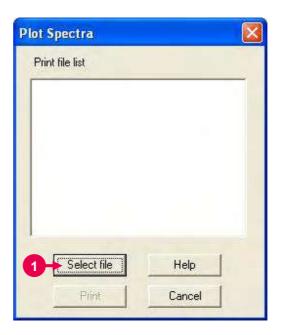


Fig. 8-23

(2) Click Select file. (Refer to ① in Fig. 8-23.)

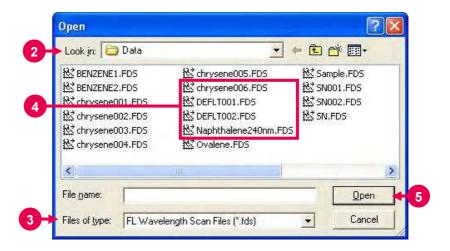


Fig. 8-24

- (3) Specify the folder of files to be printed in "Look in." (Refer to ② in Fig. 8-24.)
- (4) Select a file type in "Files of type." (Refer to ③ in Fig. 8-24.)
- (5) Select files. (Refer to ④ in Fig. 8-24.)

  The following two methods are available for selecting multiple files.
  - Method 1: Click the first file and select the subsequent ones with the ↓ key while holding down the Shift key.
  - Method 2: Click each file while holding down the Ctrl key.
- (6) After completion of selection, click Open. (Refer to ⑤ in Fig. 8-24.) The Plot Spectra dialog box will appear. The selected files are set in the Print file list as shown in Fig. 8-25.

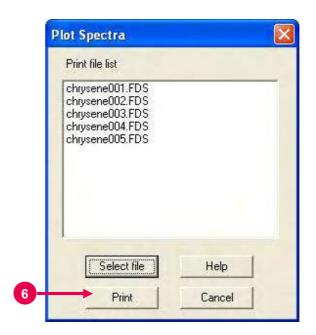


Fig. 8-25

(7) After check, click **Print**. Printing will start. (Refer to ⑥ in Fig. 8-25.)

Printing items are those selected for saving. Up to 100 files can be specified.

#### 8.10 Collective Conversion of Files

Data files can be collectively converted in the following procedure.

(1) Select the Files Conversion command from the File menu.A window as in Fig. 8-26 will appear.

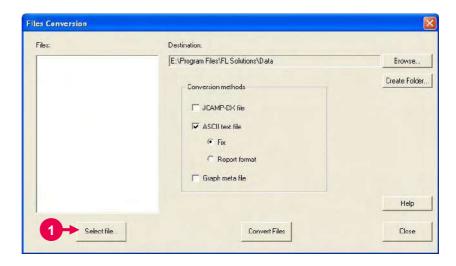


Fig. 8-26

(2) Click the **Select file** button. (Refer to ① in Fig. 8-26.) A window as in Fig. 8-27 will appear.

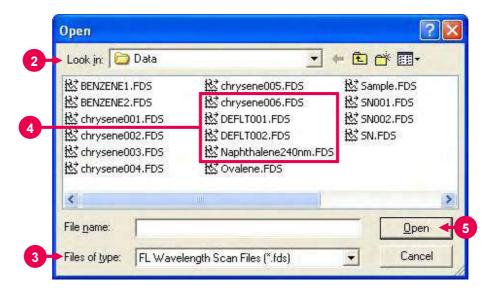


Fig. 8-27

- (3) Specify the folder of files to be converted in "Look in." (Refer to ② in Fig. 8-27.)
- (4) Select a file type in "Files of type." (Refer to ③ in Fig. 8-27.)
- (5) Select files. (Refer to ④ in Fig. 8-27.)

  The following two methods are available for selecting multiple files
  - Method 1: Click the first file and select the subsequent ones with the ↓ key while holding down the Shift key.
  - Method 2: Click each file while holding down the Ctrl key.
- (6) After completion of selection, click **Open**. (Refer to ⑤ in Fig. 8-27.) The Convert Files window will appear. The selected files are set in the Source file list as shown in Fig. 8-28.

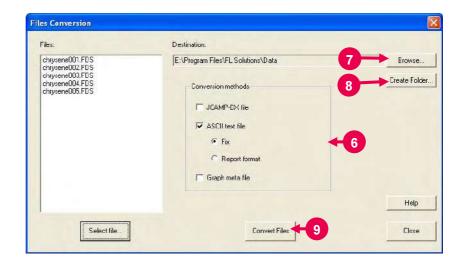


Fig. 8-28

(7) Select a file format after conversion in "Conversion methods." (Refer to ⑥ in Fig. 8-28.) You can select two or more formats for simultaneous conversion.

- JCAMP-DX file
- ASCII text file

For this file format, select information to be converted.

- Fix In any other than the photometry mode, the date, analysis method, and data list are output. In the photometry mode, all information excluding calibration curve graphs.
- Report format
   Output is provided according to the settings on the
   Report tab page of the Spectrum Properties window.
- · Graph meta file
- (8) Specify a folder for saving the converted files.

  When folders exist, the Browse for Folder dialog box appears as shown in Fig. 8-29 by clicking the **Browse** button. (Refer to ⑦ in Fig. 8-28.) Specify a folder in this dialog box and click the **OK** button.



Fig. 8-29

When there is no folder, the Create New Folder dialog box appears as shown in Fig. 8-30 by clicking the **Create Folder** button (refer to (a) in Fig. 8-28). Enter a new folder name in "Name" and click the **OK** button. A new folder will be created.

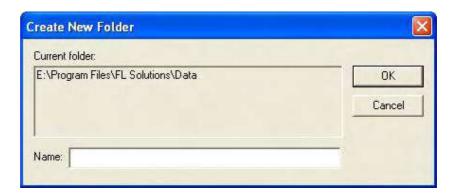


Fig. 8-30

(9) Click the **Convert Files** button (refer to ⑨ in Fig. 8-28) for file conversion.

## 8.11 File Loading with Preview

You can load a spectral data file with preview.

(1) Click the (preview file) button from the Standard toolbar.
 A window as in Fig. 8-31 will open.
 (Or select the Load Spectrum command from the File menu.)

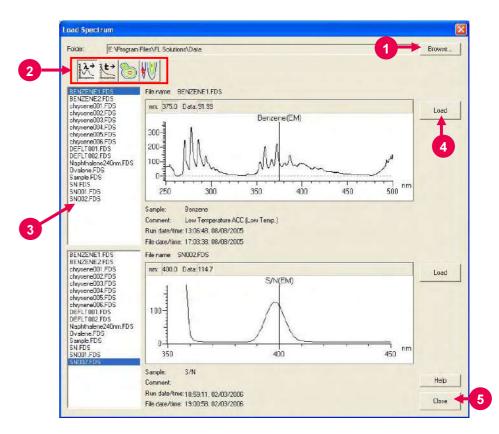


Fig. 8-31

- (2) Click the **Browse** button. (Refer to ① in Fig. 8-31.) A window as in Fig. 8-29 will appear. Specify a folder for spectrum loading.
- (3) Select a file type button. (Refer to ② in Fig. 8-31.)

  The corresponding data files in the folder are indicated in the list box.

: Wavelength scan data file

: Time scan data file

: 3-D scan data file

: 3-D time scan data file

(4) Select a desired file from the data file list. (Refer to ③ in Fig. 8-31.)

The searched data file list is displayed at the left of the window. Select a file from the list, and the relevant spectrum will be displayed. The same two data file lists are displayed. For two files, the preview can be displayed simultaneously. Upon selection of 3-D scan or 3-D time scan data file, the contours for one data element are displayed. A single data file list is available.

- (5) Click the **Load** button. (Refer to ④ in Fig. 8-31.) A spectrum displayed on the preview window will be loaded and displayed on the data processing window.
- (6) To exit the file loading, click the **Close** button. (Refer to ⑤ in Fig. 8-31.)

## 8.12 Enlargement of Values on Monitor

You can enlarge the current values on the monitor.

Click the button on the Spectrophotometer toolbar.

The instrument status window will open as shown in Fig. 8-32.

This window can be enlarged or contracted with the mouse.

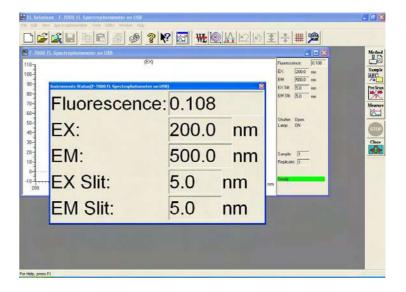


Fig. 8-32

## 8.13 Display of Monitor Window at Forefront

In repeat measurement or use of the autosampler, the monitor window may not be located because some spectral data processing windows are overlaid as shown in Fig. 8-33.

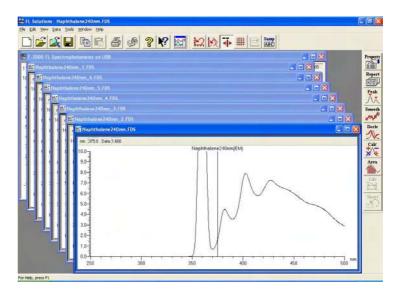


Fig. 8-33

Click the button on the Standard toolbar. The monitor window will be displayed at the forefront.

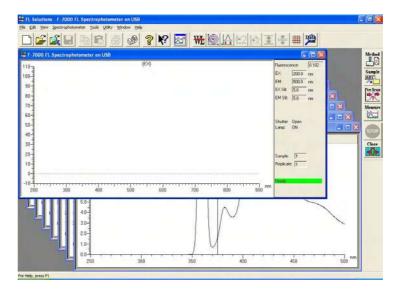


Fig. 8-34

## **MEASUREMENT USING OPTIONAL ACCESSORIES**

For how to connect the cable for each accessory, refer to the instruction manual attached to the accessory.

Given below is the description of operations after setting of your analysis method, which is required as instructed in Sections 2 to 6.

#### **Use of Sample Sipper** 9.1

Hook up the sample sipper to the spectrophotometer.

The parameters for the sample sipper are settable on its front panel. Explained here is the procedure for creating a calibration curve and measuring an unknown sample in the photometry mode.

#### 9.1.1 When Not **Using Sample Table**

A message box appears in the standard measurement session, so operate in accordance with the messages. A message box does not appear in the sample measurement session. Measurement can be started just by pressing the sipping lever of the sample sipper.

- **NOTES**: 1. In use of the sample sipper, remeasurement, interrupt measurement and blank measurement cannot be made. But remeasurement of standards is possible.
  - 2. When repeat measurement is set, the standards are measured by the set number of times. Samples are measured only once.

#### 9.1 Use of Sample Sipper

Select the Measure command from the Spectrophotometer menu or click the (measurement) button on the toolbar. A window as in Fig. 9-1 will open.

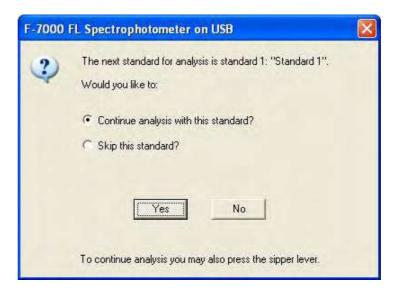


Fig. 9-1

Press the sipping lever. Measurement will start. Upon completion of the 1st standard measurement, a message box for the 2nd standard measurement will appear.

For remeasurement or skipping, select the relevant item with the mouse (by placing a bullet in its radio button) and click **Yes**. When you click **No**, the current measurement sequence is terminated.

For restarting measurement, click the button. Measurement then starts from standard 1.

Upon completion of standard measurement, a window as in Fig. 9-2 will appear.



Fig. 9-2

By clicking **OK**, a calibration curve is drawn and then samples can be measured. When a window as in Fig. 9-3 appears, press the sipping lever. The result of measurement will appear in the area indicated by ① in Fig. 9-4.

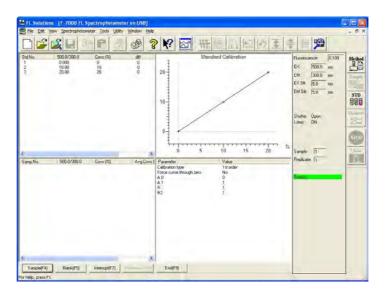


Fig. 9-3

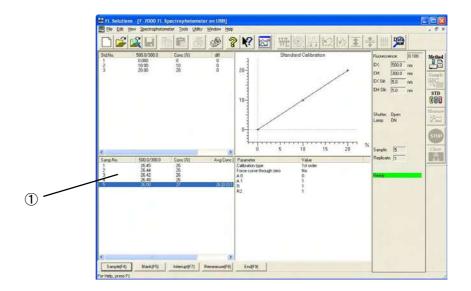


Fig. 9-4

Upon completion of measurement, click the **End** button. The data will be saved.

For remeasuring any of the standards after their measurements, click its data as shown in Fig. 9-5. Then click the **Remeasure** button.

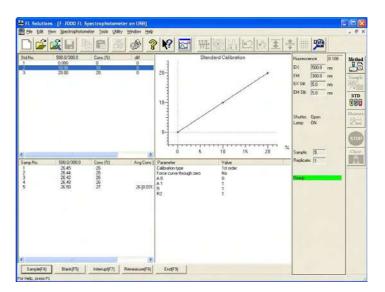


Fig. 9-5

A message box as in Fig. 9-6 will open. Press the sipping lever.



Fig. 9-6

When a standard is remeasured, the calibration curve is recalculated and sample data is also recalculated based on the curve and then displayed. To save the recalculated data, click the **End** button or the (save) button on the toolbar.

## 9.1.2 When Using Sample Table

A message box is displayed in both standard and sample measurement sessions. So operate according to the messages. When the message box appears, press the sipping lever of the sample sipper.

**NOTE**: When the sample sipper is used, blank measurement cannot be made.

Select the Measure command from the Spectrophotometer menu or click the (measurement) button on the toolbar. A window as in Fig. 9-7 will open.

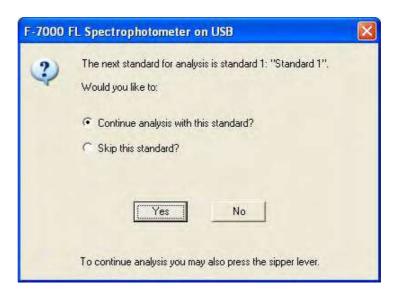


Fig. 9-7

#### 9.1 Use of Sample Sipper

Press the sipping lever. Measurement will start. Upon completion of the 1st standard measurement, a message box for the 2nd standard measurement will appear. For remeasurement or skipping, select the relevant item with the mouse (by placing a bullet in its radio button), and then click **Yes**. When you click **No**, the current measurement sequence is terminated.

For restarting measurement, click the button. Measurement then starts from standard 1.

Upon completion of standard measurement, a window as in Fig. 9-8 will open.

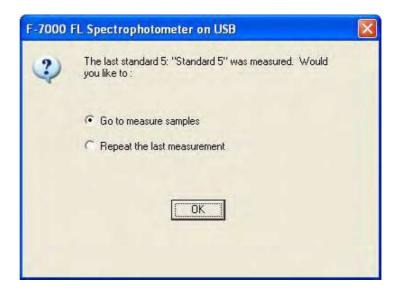


Fig. 9-8

By clicking **OK** with the mouse, a calibration curve is drawn and then samples can be measured.

In case of sample measurement also, a message box as in Fig. 9-9 will appear. Press the sipping lever. Measurement of sample 1 will start.

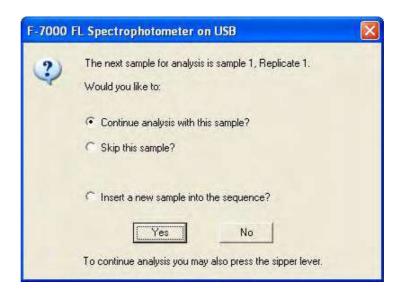


Fig. 9-9

For measurement of sample 2, a message box as in Fig. 9-10 will open.

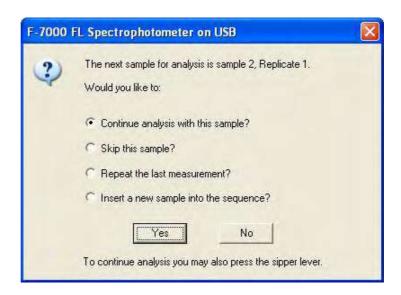


Fig. 9-10

For measurement of sample 2, press the sipping lever.

For skipping this sample (measurement of sample 3), select "Skip this sample?" and click the **Yes** button with the mouse.

For remeasurement, select "Repeat the last measurement?" and click the **Yes** button with the mouse. When repeat measurement has been set, measurement starts from the 1st one (Repl. No. 1) for the sample.

#### 9.1 Use of Sample Sipper

In case of interrupt measurement, select "Insert a new sample into the sequence?" and click the **Yes** button with the mouse. A message box as in Fig. 9-11 will appear. Input a sample name and comment, and then click the **OK** button.



Fig. 9-11

For remeasurement after a series of measurements, click the data to be remeasured as shown in Fig. 9-12. Then lick the **Remeasure** button.

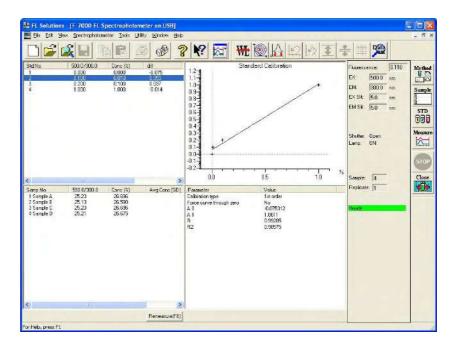


Fig. 9-12

A window as in Fig. 9-13 will open. Press the sipping lever, and measurement will start. When a standard is remeasured, the calibration curve is recalculated and sample data is also recalculated based on the curve and then displayed. To save the recalculated data, click the (save) button.



Fig. 9-13

## 9.1.3 In Wavelength (1) Scan or Time Scan Mode

When Not Using Sample Table

Set your analysis method. When the monitor window appears, press the sipping lever. Measurement will start.

(2) When Using Sample Table

Select the Measure command from the Spectrophotometer menu or click the (measurement) button on the toolbar.

A message box will appear. Follow the messages given.

## 9.2 Use of AS-3000 Intelligent Autosampler

Set the rack parameters, injection volume and other items with the AS-3000 Intelligent Autosampler. For details, refer to the instruction manual attached to the AS-3000.

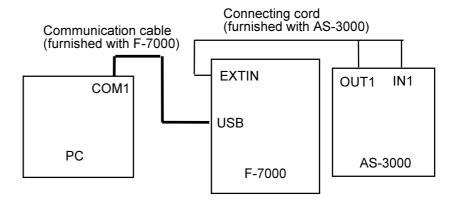


Fig. 9-14 Cord Connection

Parameter setting: Set all parameters in the AS-3000.

**NOTE**: When using a sample table, match the number of samples set in the AS-3000 with that set in the sample table. If the latter is less than the former, some samples cannot be measured.

#### <Measurement>

of the FL Solutions program or click the (measurement) button on the toolbar. A window as in Fig. 9-15 will open.

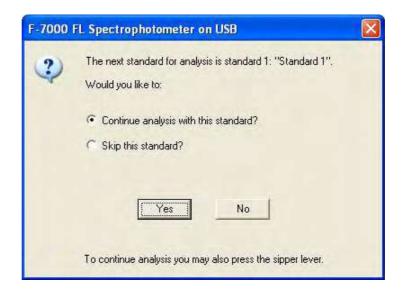


Fig. 9-15 For Start from Creation of Calibration curve

- (2) Press the **START** key on the AS-3000.
- (3) Measurement will start.

## 9.3 Analog Output

For connection, use the analog output terminal. Select the Analog Output command from the Utility menu. (The analog output I/F (option) is required.)

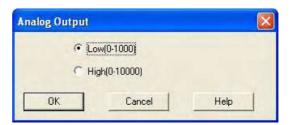


Fig. 9-16

This is a setting of data value corresponding to analog output of 1 V. Analog output value (V) becomes as follows.

Low: (data value)/1000 High: (data value)/10000

### APPENDIX A DETAILS OF QUANTIFICATION

#### A.1 Foreword

In the Photometry mode of the F-7000 spectrophotometer, the following four calibration curve types are available.

- · Linear working curve
- · Quadratic working curve
- · Cubic working curve
- Multiple segment working curve

These are explained in detail below.

## A.2 Linear Working Curve (1st order)

A regression line is determined via the least squares method from a maximum of 20 data. The calculation formula is as follows:

$$x = A1 \cdot y + A0$$

A1 = 
$$\frac{\sum y_i x_i - \frac{1}{n} \sum y_i \cdot \sum x_i}{\sum y_i^2 - \frac{1}{n} (\sum y_i)^2}$$
, A0 =  $\frac{\sum x_i}{n} - A1 \cdot \frac{\sum y_i}{n}$ 

where, x: Sample concentration (input value)

y: Sample data (measured value)

n: Number of samples

### A.3 Quadratic Working Curve (2nd order)

A quadratic curve is determined via the least squares method from a maximum of 20 data. The calculation formula is as follows:

$$x = A2 \cdot y^{2} + A1 \cdot y + A0$$

$$A2 = \frac{S(y^{2}x)S(yy) - S(yx)S(yy^{2})}{S(yy)S(y^{2}y^{2}) - \left\{S(yy^{2})\right\}^{2}}$$

$$S(yy) = \sum y_{i}^{2} - \frac{\left(\sum y_{i}\right)^{2}}{n}$$

$$A1 = \frac{S(yx)S(y^{2}y^{2}) - S(y^{2}x)S(yy^{2})}{S(yy)S(y^{2}y^{2}) - \left\{S(yy^{2})\right\}^{2}}$$

$$S(yx) = \sum y_{i}x_{i} - \frac{\sum y_{i} \cdot \sum x_{i}}{n}$$

$$S(yx) = \sum y_{i}^{3} - \frac{\sum y_{i} \cdot \sum y_{i}^{2}}{n}$$

$$S(yy^{2}) = \sum y_{i}^{3} - \frac{\sum y_{i} \cdot \sum y_{i}^{2}}{n}$$

$$S(y^{2}x) = \sum y_{i}^{2}x_{i} - \frac{\sum y_{i}^{2} \cdot \sum x_{i}}{n}$$

$$S(y^{2}y^{2}) = \sum y_{i}^{3} - \frac{\sum y_{i}^{2} \cdot \sum x_{i}}{n}$$

$$S(y^{2}y^{2}) = \sum y_{i}^{3} - \frac{\sum y_{i}^{2} \cdot \sum x_{i}}{n}$$

where, x: Sample concentration (input value)

y: Sample data (measured value)

n: Number of samples

### A.4 Cubic Working Curve (3rd order)

A cubic curve is determined via the least squares method from a maximum of 20 data. The calculation formula is as follows:

$$x = A3 \cdot y^3 + A2 \cdot y^2 + A1 \cdot y + A0$$

where, x: Sample concentration (input value)

y: Sample data (measured value)

n: Number of samples

## A.5 Multiple Segment Working Curve (Segmented)

The calibration curve is apt to bend when measuring turbid samples or the like. Use of a quantification program allows correcting the calibration curve by using up to 20 standards. Figure A-1 shows an example of curve correction. For the part which exceeds the measuring range of the standards, simply extend the line as it is.

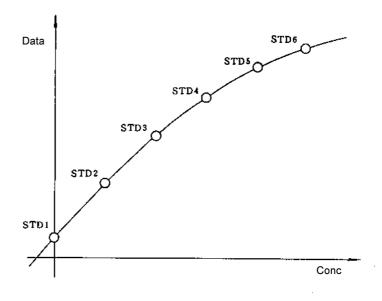


Fig. A-1 Correction of Curve Bending

< Notes on Preparation of Multiple Segment Working Curve>

(1) A correct calibration curve can be created only when the measured value increases or decreases monotonically versus the concentration value. Especially when the inclination is negativegoing, be sure to measure a blank having a data value larger than the other standards. A curve not showing a monotonic increase will appear as in Fig. A-2. And when the data value of the blank is small, regardless of a negative-going inclination, the curve will appear as in Fig. A-3.

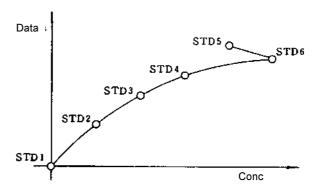


Fig. A-2 Example of Curve without Monotonic Increase (unsuitable curve)

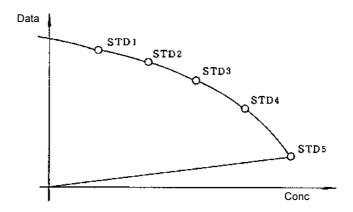


Fig. A-3 Example of Negative-going Curve where Measured Value of Blank is Small (unsuitable curve)

#### (2) Remeasurement of Standards

The standards can be remeasured after once preparing a calibration curve. The curve prepared in such a case will appear as in Fig. A-4.

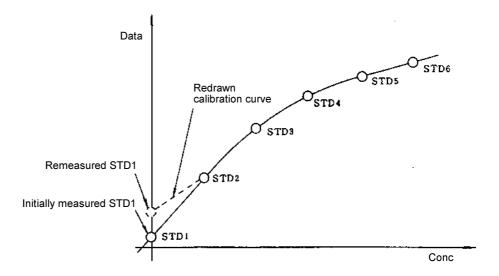


Fig. A-4 Calibration Curve when Standards are Remeasured

## APPENDIX B DETAILS OF RATE ANALYSIS FUNCTION

#### **B.1** Foreword

Rate analysis is used in the analysis of enzyme reactions. It is utilized for clinical and biochemical tests by reagent manufacturers, hospitals and so on. A computer is used to calculate the concentration from the variation in data per unit time, and the result is displayed and printed out.

#### **B.2** Calculation Method

A timing chart for rate analysis is shown in Fig. B-1. Data is acquired when the initial delay time has elapsed after pressing the Measure button. A regression line is determined from this data via the least squares method, and the gradient and activity value are calculated. The calculation formula is as follows.

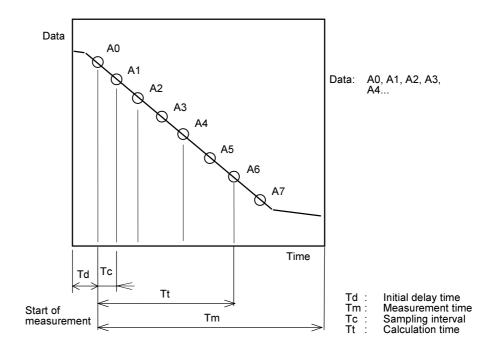


Fig. B-1

Prepare a regression line via the least squares method from the measured data, and obtain a determination coefficient.

$$y = ax + b$$
 where,

$$a = \frac{\sum_{i} x_{i} y_{i} - \sum_{i} x_{i} \sum_{i} y_{i} / n}{\sum_{i} x_{i}^{2} - \left(\sum_{i} x_{i}\right)^{2} / n} \quad b = \sum_{i} y_{i} - \frac{a * \left(\sum_{i} x_{i}\right) / n}{\sum_{i} x_{i}^{2} - \left(\sum_{i} x_{i}\right)^{2} / n}$$

x<sub>i</sub>: Time (s) of each data

y<sub>i</sub>: Value of each data

n: Number of samples

The determination coefficient CD becomes as follows:

$$CD = \frac{\left(n\sum x_{i}y_{i} - \sum x_{i}\sum y_{i}\right)^{2}}{\left(n\sum x_{i}^{2} - \left(\sum x_{i}\right)^{2}\right)\left(n\sum y_{i}^{2} - \left(\sum y_{i}\right)^{2}\right)}$$

• Gradient (variation per minute)

$$D_i = \frac{a}{Tk} = 60a(/min)$$

Activity

$$C_i = k \cdot D_i$$

• R (relative coefficient)

$$R = CD = \sqrt{\frac{\left(n\sum x^{i}y^{i} - \sum x^{i}\sum y^{i}\right)^{2}}{\left(n\sum x^{i} - \left(\sum x^{i}\right)^{2}\right)\left(n\sum y^{i} - \left(\sum y^{i}\right)^{2}\right)}}$$

• R2 (determination coefficient)

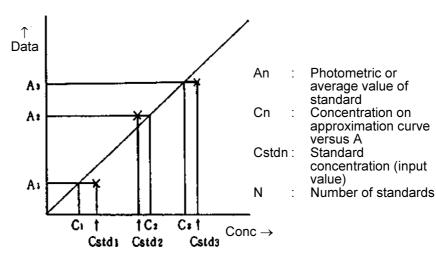
$$R = (CD)^{2} = \frac{\left(n\sum_{i}x y - \sum_{i}x \sum_{i}y\right)^{2}}{\left(n\sum_{i}x^{2} - \left(\sum_{i}x\right)^{2}\right)\left(n\sum_{i}y^{2} - \left(\sum_{i}y\right)^{2}\right)}$$

**NOTE**: If the range for rate calculation does not coincide with the actual measured data range, then use only the measured data within the range for the calculation.

#### APPENDIX C **DETERMINATION COEFFICIENT OF CALIBRATION CURVE**

#### **Calculation of Determination Coefficient C.1**

The determination coefficient and other factors are calculated via the following formula.



Difference DIFF : DIFF<sub>n</sub> =  $C_n - C_{stdn}$ 

 $: RD_n = \frac{DIFF}{\overline{A}} \times 100$ Relative difference RD

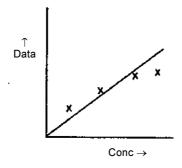
 $\vdots t_n = \frac{DIFF_n}{\sqrt{\frac{\sum DIFF^2}{N-1}}}$ Student's t-test

 $R = \sqrt{\frac{\sum (C_n - \overline{C})^2 - \sum (C_n - C_{stdn})^2}{\sum (C_n - \overline{C})^2}}$ Relative coefficient

Determination coefficient :  $R_2 = (R)^2$ 

## C.2 Usage of Determination Coefficient

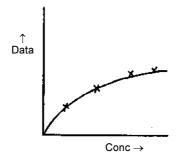
The determination coefficient indicates the goodness of fit of the measured standards and the prepared calibration curve. The closer this value is to "1," the better the fit of the measured value and calibration curve. If the value is far from "1," then the standards must be measured or the calibration curve mode must be changed. Examples of determination coefficients upon changing the calibration curve mode are given below.



x: Standard measurement points

When calibration curve type is set to linear:

Determination coefficient < 1



When calibration curve type is set to quadratic:

Determination coefficient ≠ 1

With the above standard data, it can be seen that a quadratic curve provides a better result. A calibration curve, which was formerly judged through skill or experience, can thus be judged more easily through numerical means.

# APPENDIX D INTEGRATION METHOD AND SMOOTHING

#### **D.1** Foreword

The integration methods and smoothing used in the FL Solutions program are explained here.

### **D.2 Integration Methods**

The FL Solution program comes with the following three integration methods.

- Rectangular
- Trapezoid
- Romberg

## D.2.1 Rectangular Method

The Rectangular method is the simplest calculation method among the above three. Since one sampling interval is equal to the width of each sectional area, the total value of all data points covering a peak is approximated as a target area.

As shown in Fig. D-1, the target area is a total of the rectangular parts obtained from linear approximation of the curve drawn by following the data points. If the number of peak data points is small, approximation will become rough.

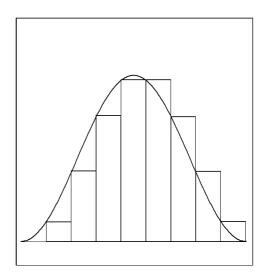


Fig. D-1

## D.2.2 Trapezoid Method

The Trapezoid method is an improved method of peak area calculation. Each section having a width equal to one sampling interval is indicated by a rectangle and a triangle on it. And the target area is given by totaling the area values of all sections.

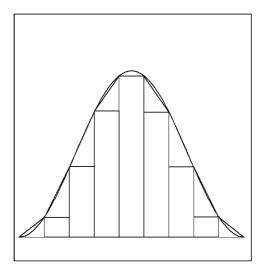


Fig. D-2

When focusing attention on one section, the area  $I_{\text{r}}$  of its rectangular part can be expressed as follows.

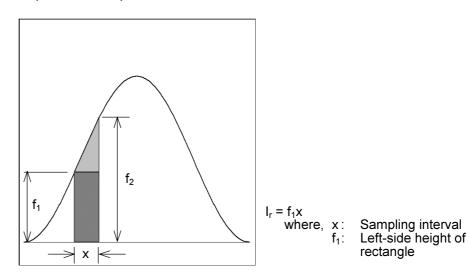


Fig. D-3

To this rectangle, a triangular part is to be added.

When replacing the sectional area with  $I_T$ , and the triangular area with  $I_t$ ,  $I_T$  is given by the following formula.

$$I_T = I_t + I_r = \frac{f_2 - f_1}{2}x + f_1x = \left(\frac{f_1 + f_2}{2}\right)x$$

The above formula can be transformed to cover the entire peak as follows.

$$I_T = \left(\frac{f_1}{2} + f_2 + ... + f_{n-1} + \frac{f_n}{2}\right) x$$

## D.2.3 Romberg Method

The Romberg method is the most exact integration used in this program. The aforementioned Trapezoid method has different step sizes (sampling intervals) for determining a peak area more exactly. This method capable of using a total of inherent errors can use both of two step sizes individually conceivable. However, unlike a classic method of continuous approximation, the step size cannot be reduced freely in the same way as an increase in the X-axis direction for exact integration. This is because a spectrum is handled as average data of the data points which define the spectrum.

Yet, the Romberg method allows an increase in step size by a factor of 2 or 4 when necessary. This method is implemented as follows.

- (1) At every data point, data is integrated by the Trapezoid method.
- (2) At every two data points, data is integrated by the Trapezoid method.
- (3) The results of the above (1) and (2) are combined with each other to obtain the following.

$$I_R = I_n + \frac{I_n - I_{2n}}{3}$$

where, I<sub>R</sub>: Integration by Romberg method

In : Trapezoid method at every data point

I<sub>2n</sub>: Trapezoid integration at every two data point

## D.3 SMOOTHING

#### D.3.1 Foreword

The FL Solutions program comes with the following three smoothing methods.

- Savitsky-Golay smoothing
- · Mean smoothing
- Median smoothing

These are explained in detail below.

#### D.3.2 Savitsky-Golay Smoothing

**D.3.2** Savitsky-Golay For this method, refer to the following literature.

Gorry, P.A.

"General Least-Squares Smoothing and Differentiation by the Convolution (Savitsky -Golay) Method"
Anal.Chem. **1990**, 62, 570-573.

# D.3.3 Mean Smoothing

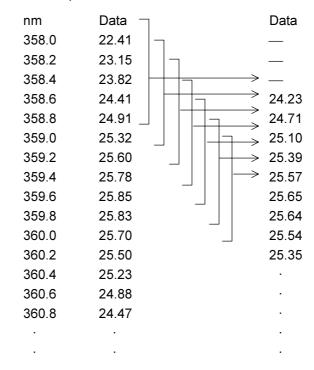
Average data is obtained from the numerical values at the specified data points and set for the center wavelength.

#### Example)

Number of data points: 7 Number of times : 1

In case of 7 data points starting from 358.0 nm, the average value is set for the center wavelength of 358.6 nm. At this time, data disappears at 3 points at the both sides on the spectrum.

#### How to acquire data



When an even number (2n) of data points are specified,

"2n" is replaced with "2n+1" for calculation. This means that the same result is obtained when the number of data points is set at "8" and at "9."

## D.3.4 Median Smoothing

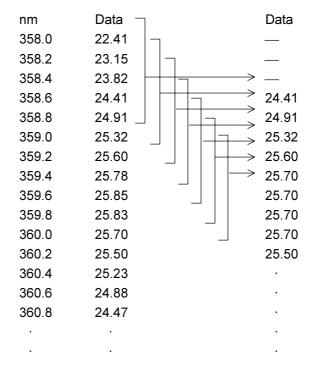
Middle data is obtained from the numerical values at the specified data points and set for the center wavelength.

#### Example)

Number of data points: 7 Number of times : 1

In case of 7 data points starting from 358.0 nm, the 4th smallest value is set for the center wavelength of 358.6 nm. At this time, data disappears at 3 points at the both sides on the spectrum.

#### How to acquire data



When an even number (2n) of data points are specified,

"2n" is replaced with "2n+1" for calculation. This means that the same result is obtained when the number of data points is set at "8" and at "9."

# APPENDIX E DESCRIPTION OF FLUOROMETRY

## **E.1** Description of Fluorescence

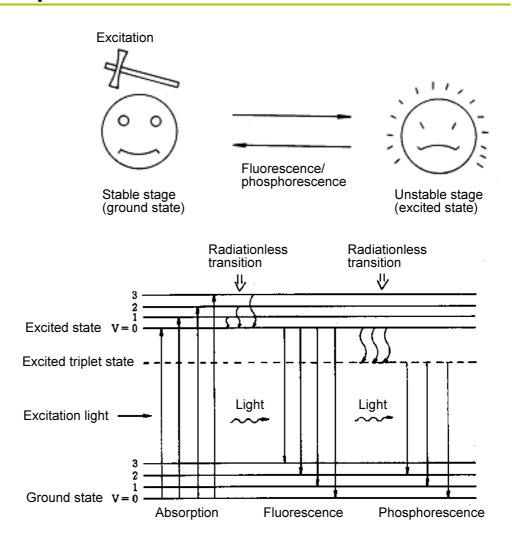


Fig. E-1 Typical Organic Molecular Energy Level

Figure E-1 illustrates the energy level transitions in an organic molecule in processes of light absorption and emission.

When light strike an organic molecule in the ground state, it absorbs radiation of certain specific wavelengths to jump to an excited state. A part of the excitation (absorbed) energy is lost on vibration relaxation, i.e., radiationless transition to the lowest vibrational level takes place in the excited state. And, eventually the molecule returns to the ground state while emitting fluorescent radiation.

Also, if radiationless transition to the triplet state takes place, then phosphorescence is emitted during triplet-to-singlet transition (from the excited triplet state to the ground singlet state).

Generally phosphorescence persists for 10<sup>-4</sup> sec or longer due to the selection rule imposed on the triplet-to-singlet transition. In contrast, fluorescence persists for a period of 10<sup>-8</sup> to 10<sup>-9</sup> sec in most cases.

As mentioned above, a part of the radiation absorbed by a substance is lost as vibration energy, etc.; therefore, the fluorescence emitted from the substance has a longer wavelength than the excitation light (Stokes' law).

The ratio of the number of photons emitted during fluorescence to the number of photons absorbed is called the quantum efficiency of fluorescence. The larger the quantum efficiency a substance has, the more fluorescence it emits. Also, the intensity of fluorescence emitted from a substance is proportional to the quantity of light absorbed by it. When a dilute solution sample is measured, the fluorescence intensity is expressed by the following formula.

 $F = KI_0cle\epsilon\phi$ 

where, F: Intensity of fluorescence

K: Instrumental constant

 $I_0$ : Intensity of exciting radiation c: Concentration of substance

1 : Cell path length

ε : Absorptivity of substance

 $\boldsymbol{\phi} \;\; : \;\;$  Quantum efficiency of substance

## **E.2** Advantages of Fluorometry

As contrasted with fluorometry, absorptiometry for a low-concentration sample is explained in the following:

A sample having 99% transmittance to blank is taken as an example. In the absorbance measurement of such a substance, inaccuracies must always be taken into consideration. Here, the inaccuracy is assumed to be 0.1%. Since it has an effect on both the blank and sample.

Percent transmittance of blank	100.0 ± 0.1%
Percent transmittance of sample	99.0 ± 0.1%
Difference (proportional to sample concentration)	1.0 ± 0.2%

In this example, the uncertainty in concentration measurement is ±20%. While in the fluorometry, a difference from zero level corresponds to the concentration of sample. Accordingly, the measurement accuracy is as follows:

Output signal level at sample measurement	100 ± 0.1
Value corresponding to blank	0 ± 0.1
Difference (proportional to sample concentration)	100 ± 0.2

As can be seen from the above, the fluorometry is very advantageous for analyzing a low-concentration sample since its uncertainty is in most cases theoretically independent of the sample concentration.

Although actual practice may involve some error factors which increase as the sample concentration becomes extremely low, the fluorometry is capable of measuring low concentrations with an accuracy 100 times higher than in absorptiometry.

Figure E-2 is an explanatory illustration of the foregoing description. In absorptiometry, a difference between the quantity of incident radiation  $I_{\rm o}$  and the quantity of transmitted radiation  $I_{\rm t}$  is represented by signal  $I_{\rm s}$ . A level at which the signal  $I_{\rm s}$  becomes almost equal to a noise level is used as a detection limit.

In fluorometry, however, since the quantity of fluorescence  $I_F$  itself is represented as a signal, just a small amount of fluorescence can be amplified electrically for enabling detection.

Still more, since a fluorescence wavelength of a substance is different from its excitation wavelength (incident light wavelength), the fluorescence wavelength is not readily affected by the exciting radiation, thereby contributing to ensuring high sensitivity.

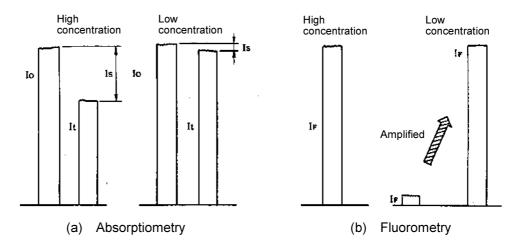


Fig. E-2 Comparison between Absorptiometry and Fluorometry

In addition to high sensitivity, the fluorometry is advantageous in that more information is attainable. An emission spectrum is also available besides an excitation spectrum which corresponds to an absorption spectrum in absorptiometry.

The two kinds of wavelengths can be selected as desired, and a fluorescence spectrum can be recorded using a properly selected excitation wavelength (or vice versa). Thus, quantitative and qualitative analyses can be made for a sample containing plural components.

Figure E-3 shows a simplified spectral graph of measurement of a sample containing multiple components. In absorptiometry, since only the absorption spectrum can be measured, two or more component wavelengths are presented. If the absorption wavelengths are similar to each other, each component cannot be separated in measurement. In fluorometry, even if the absorption wavelengths are similar, a difference in fluorescence makes it possible to select each fluorescence wavelength properly. Thus, each component can be separated in measurement.

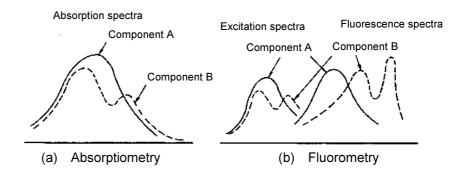


Fig. E-3 Measurement of Multi-component Sample

Table E-1 compares information attainable in absorptiometry and that in fluorometry.

Table E-1 Comparison of Information Attainable in Absorptiometry and Fluorometry

Absorptiometry	Fluorometry
Absorption spectrum only (corresponding to excitation spectrum in fluorometry)	<ul><li>Excitation spectrum</li><li>Fluorescence spectrum</li></ul>

# E.3 Remarks on Measurement in Fluorescence Analysis

# E.3.1 Temperature Dependency of Fluorescence Intensity

**Temperature** For most kinds of samples, an increase of 1 °C in sample temperature **Dependency of** causes the fluorescence intensity to decrease by 1 to 2%.

It is also reported that for some kinds of biochemical samples, the fluorescence intensity decreases as much as 10% as the temperature increases by 1 °C. When analyzing a sample having a temperature-dependent property, it is advisable to use the constant-temperature cell holder (P/N 650-0150). Constant-temperature measurement can be carried out by circulating constant-temperature water through this cell holder.

# E.3.2 Chemical Change in Sample due to Radiation

Some kinds of samples may be susceptible to a chemical change due to exciting radiation. In analysis of such a sample, keep the shutter closed to cut off an excitation beam until measurement is started, and then open the shutter immediately before measurement. If any chemical change due to exciting radiation is observed still, determine a signal level at the start time through extrapolation according to variation in signal level.

# E.3.3 Raman Scattering

In fluorescence measurement, spectra having different natures from that of fluorescence may be observed. These are called Rayleigh scattering spectrum and Raman scattering spectra; the former appearing at the same wavelength position as the excitation spectrum, and the latter appearing at the longer-wavelength side near Rayleigh scattering.

In a fluorescence spectrum, when the excitation wavelength is shifted, only the peak height is changed while the peak wavelength position remains intact. In a Raman scattering spectrum, when the excitation wavelength is shifted, the peak wavelength position is also changed accordingly. Both the Rayleigh scattering and Raman scattering are caused by a solvent which may be contained in the sample. When examining the spectral plot, be careful not to mistake these scattering effects for the fluorescence peak of interest. Table E-2 presents the Raman spectral peak position at each excitation wavelength for the purpose of reference.

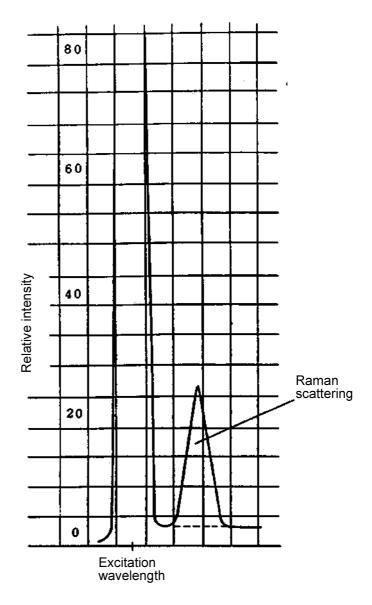


Fig. E-4 Raman Spectrum of Water

Table E-2 Raman Peak Positions at Respective Excitation Wavelengths

	(excitation wavelength)	Water	Ethanol	Cyclohexane	Carbon Tetrachloride	Chloroform
Excitation	248	271	267	267		
wavelength and Raman	313	350	344	344	320	346
peak	365	416	405	408	375	410
position (nm)	405	469	459	458	418	461
	436	511	500	499	450	502

# E.3.4 Handling of High-concentration Samples

In measurement of a high-concentration sample, a variety of error factors may be involved.

The most significant error factor consists in that an excitation beam is absorbed at the entrance of a cell to prevent a sufficient level of excitation at the center of the cell.

Figure E-5 illustrates an extreme case of this condition.

Although fluorescence is emitted in the vicinity of the entrance for the excitation beam, it is not taken into the emission monochromator.

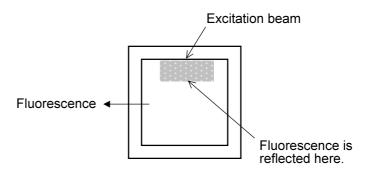


Fig. E-5 Sample Having an Extremely High Concentration

If only the incident point of excitation beam is bright, it is necessary to dilute the sample properly for measurement.

The second significant error factor consists in extinction due to concentration. This condition is caused by preventing activation through interaction of molecules.

The third significant error factor consists in re-absorption of fluorescence. As shown in Fig. E-6, this condition occurs due to overlapping between the short-wavelength side of fluorescence spectrum and the long-wavelength side of excitation spectrum.

Therefore, it seems that the fluorescence spectrum has been shifted toward the long-wavelength side to some extent. In measurement of an ordinary kind of sample, however, this condition will not impede quantitative determination significantly

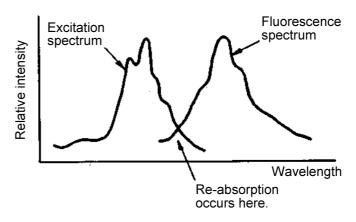


Fig. E-6 Explanatory Illustration of Re-absorption

In any case, if there is a possibility of a measurement error due to high concentration of a sample, dilute the sample properly or carry out surface fluorescence measurement using a solid sample holder.

#### F.3.5 Second-order Scattered Radiation

Where the excitation and emission wavelengths are plotted near each other, care should be exercised not to mistake the Raman and Rayleigh scattering for the fluorescence spectrum as mentioned in E.3.3. Where the excitation and emission wavelengths are plotted apart from each other, care should be exercised not to mistake the second-order and third-order scattered radiation for the fluorescence spectrum. The second-order scattered radiation appears at a wavelength two times longer than the excitation wavelength, and the third-order scattered radiation occurs at a wavelength three times longer. For instance, if the excitation wavelength is 240 nm, the second-order and third-order radiation take place at 480 and 720 nm, respectively. For eliminating these scattered radiations, insert a short-wavelength cutoff filter in the path of fluorescing radiation (before emission monochromator). It is advisable to use the filter set (P/N 650-0157) which is available as an optional accessory.

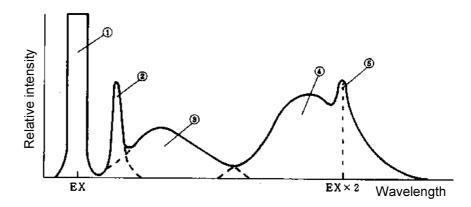
#### F.3.6 of Cell

Contamination Since the fluorescence spectrophotometer provides high sensitivity, just a slight amount of contamination on a cell may have an adverse effect on results of measurement. To prevent this, treat the cell properly after its use. Do not leave the cell containing sample. In evaporation of a solvent, a residue of sample may adhere to the wall of the cell to cause contamination.

> In measurement of a very dilute sample, contamination on the inner and outer walls of the cell may cause a problem. If a droplet of sample solution is put on the outer wall of the cell in sample injection into it, wipe the cell with tissue paper and then set it into the cell holder.

#### E.3.7 Measurement Example of Fluorescence Spectrum

Figure E-7 shows a measurement example of fluorescence spectrum.



- ① Scattering of exciting radiation
- 2 Raman spectrum of solvent
- 3 Fluorescence of impurities, solvent, etc.
- 4 Fluorescence of sample
- 5 Second-order spectrum of exciting radiation

Fig. E-7 Measurement Example of Fluorescence Spectrum

As shown in Fig. E-7, other peaks than a fluorescence peak of sample appear in measurement of fluorescence spectrum. With reference to this example, it is necessary to identify a fluorescence peak of sample.

# APPENDIX F MEASUREMENT OF INSTRUMENTAL RESPONSE (CORRECTED SPECTRA)

Spectrum correction is performed to enable measuring a true spectrum by eliminating instrumental response such as wavelength characteristics of the monochromator or detector (photomultiplier). The measurement of instrumental response is needed to perform spectrum correction. "Instrumental Response" is the function to measure and save the instrumental response.

# F.1 Measurement of Instrumental Response on Excitation Side

## F.1.1 Handling of Rhodamine B

This is the function to obtain the instrumental response on the excitation side such as wavelength characteristics of the excitation monochromator using Rhodamine B as a standard (quantum counter). The instrumental response is automatically read with a single wavelength scan operation. A spectrum is correctable within a range of 200 to 600 nm.



Rhodamine B can cause injury if directly touched or accidentally ingested. When handling it, be sure to wear proper protective gear such as safety gloves and safety mask. If Rhodamine B adheres to the skin, wash it off with soap and plenty of water. Consult a physician when needed. If accidentally ingested, immediately consult physician.

Pour Rhodamine B into a triangular cell in the procedure illustrated in Fig. F-1. The triangular cell filled with Rhodamine B should be in principle stored at a dark place.

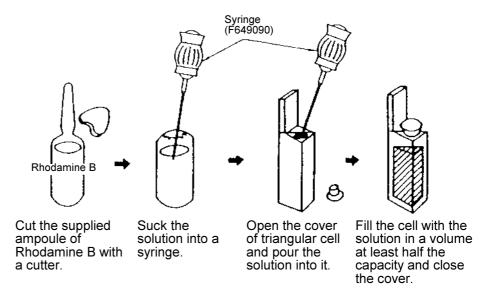


Fig. F-1 Handling of Rhodamine B

# F.1.2 Operating Procedures

- (1) Click the (analysis method) button on the Measurement toolbar. A box for setting your analysis method will appear.
- (2) Select the General tab. On the General tab page, specify "Wavelength scan" for the measurement mode.
- (3) Select the Instrument tab.
- (4) Set "Fluorescence" for the data mode, "400 V" for the photomultiplier voltage and "Excitation" for the scan mode.
- (5) Execute the Zero Adjust command from the Spectrophotometer menu to calibrate the zero point.

(6) As shown in Fig. F-2, set the triangular cell filled with Rhodamine B into the cell holder in the sample compartment, the filter holder to the sample compartment and the red filter (single-side frosted) in the filter holder. In so doing, pay attention to the direction of the triangular cell and filter. (Refer to Fig. F-3)

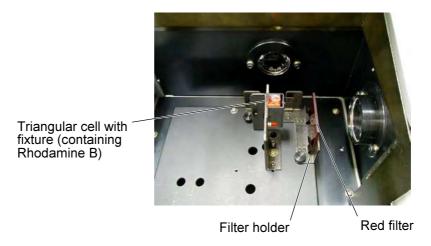


Fig. F-2 Setting of Triangular Cell Containing Rhodamine B

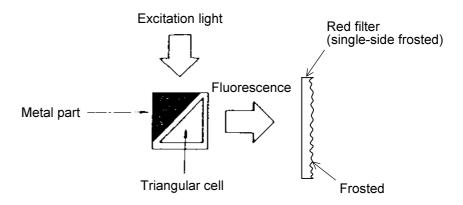


Fig. F-3 Setting Direction of Triangular Cell (top view)

NOTE: It may take time to obtain stable data due to characteristics of the detector immediately after the photomultiplier voltage has been remarkably changed (from 700 V to 250 V for example). When more accurately calibrating the zero point, wait until stable data is obtained and then proceed to step (5).

(7) Execute the Instrumental Response command from the Utility menu. The Instrumental Response dialog box will open as shown in Fig. F-4.

Select "EX [200 to 600 nm]." (Refer to ① in Fig. F-4.)

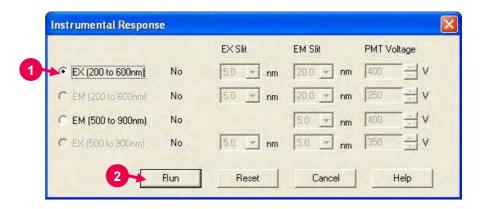


Fig. F-4

(8) Click the **Run** button. (Refer to ② in Fig. F-4.)
A message box "Set the triangular cell with Rhodamine B to cell holder and set the red filter." will appear. After setting the Rhodamine B and red filter, click the **OK** button. (Refer to ③ in Fig. F-5.)

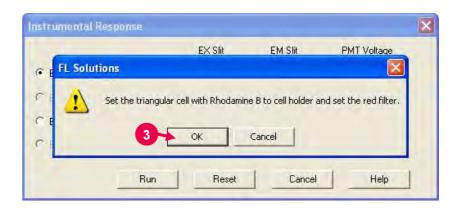


Fig. F-5

(9) The EX wavelength is set at 640 nm and the instrument starts EX wavelength scan (from 198.6 to 601.4 nm).

At this stage, the following measuring conditions are automatically set.

Data mode : Fluorescence
EX slit : 5.0 nm
EM slit : 20.0 nm
Scan speed : 60 nm/min

• Scan mode : Excitation

PMT voltage: 400 VResponse: Auto

(10) Upon completion of measurement, the conditions set before measurement are retrieved.

Now, the instrument response on the excitation side has been measured and stored. Take out the rectangular cell.

### F.2 Measurement of Instrumental Response on Emission Side

#### F.2.1 Overview

This is the function to obtain the instrumental response on the emission side such as wavelength characteristics of the emission monochromator or detector (photomultiplier).

The instrumental response on the emission side is determined by measuring a combination of instrumental response on both the excitation and emission sides by synchronous wavelength scan, and dividing it by the instrumental response on the excitation side preliminarily measured. An emission spectrum is correctable within a range of 200 to 600 nm.

## F.2.2 Operating Procedures

- (1) Click the (analysis method) button on the Measurement toolbar. A box for setting your analysis method will appear.
- (2) Select the General tab. On the General tab page, specify "Wavelength scan" for the measurement mode.
- (3) Select the Instrument tab.
- (4) Set "Fluorescence" for the data mode and "250 V" for the photomultiplier voltage.
- (5) Execute the Zero Adjust command from the Spectrophotometer menu to calibrate the zero point.
- (6) As shown in Fig. F-6, set the diffuser into the cell holder in the sample compartment.

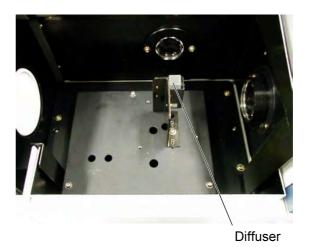


Fig. F-6 Setting of Diffuser

- NOTES: 1. Measurement of the instrumental response on the excitation side must be completed before executing measurement of the instrumental response on the emission side.
  - 2. It may take time to obtain stable data due to characteristics of the detector immediately after the photomultiplier voltage has been remarkably changed (from 700 V to 250 V for example). When more accurately calibrating the zero point, wait until stable data is obtained and then proceed to step (5).
- (7) Execute the Instrumental Response command from the Utility menu. The Instrumental Response dialog box will open as shown in Fig. F-7.
  - Select "EM [200 to 600 nm]." (Refer to ① in Fig. F-7.)

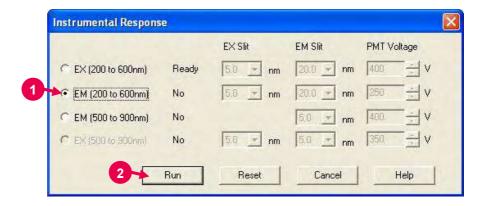


Fig. F-7

(8) Click the **Run** button. (Refer to ② in Fig. F-7.) A message box "Set diffuser to cell holder." will appear. Click the **OK** button. (Refer to ③ in Fig. F-8.)

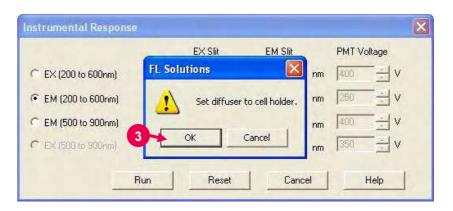


Fig. F-8

(9) The EX and EM wavelengths are set at 198.6 nm and the instrument starts synchronous wavelength scan (from 198.6 to 601.4 nm).

At this stage, the following measuring conditions are automatically set.

Scan mode: Synchronous
Data mode: Fluorescence
EX slit: 5.0 nm
EM slit: 20.0 nm
Scan speed: 60 nm/min
PMT voltage: 250 V
Response: Auto

(10) Upon completion of measurement, the conditions set before measurement are retrieved.

Now, the instrument response on the excitation side has been measured and stored. Take out the diffuser.

## F.3 Measurement of Instrumental Response in Long Wavelength Range on Emission Side

#### F.3.1 Overview

When a corrected spectrum in the long wavelength area of 600 nm or more is required on the emission side, the beam of a lamp whose wavelength characteristics are known is allowed to be irradiated to the emission monochromator, and the instrumental response in the long wavelength range on the emission side is read for correction.

As a light source, a sub standard light source (P/N 250-0123) is used. The correction range by it is 500 to 900 nm and, when combined with an instrumental response in the short wavelength range, spectrum correction is available over the entire wavelength range (200 to 900 nm) on the emission side.

## F.3.2 Operating Procedures

<Mounting and Positioning Sub Standard Light Source>Refer to the instruction manual for the sub standard light source (optional accessory).

- (1) Close the cover of the sample compartment.
- (2) Click the (analysis method) button on the Measurement toolbar. A box for setting your analysis method will appear.
- (3) Select the General tab. On the General tab page, specify "Wavelength scan" for the measurement mode.
- (4) Select the Instrument tab.
- (5) Set "Fluorescence" for the data mode and "400 V" for the photomultiplier voltage.
- (6) Execute the Zero Adjust command from the Spectrophotometer menu to calibrate the zero point.
- (7) Set the sub standard light source in the sample compartment with reference to its instruction manual. (Refer to Fig. F-9.) Turn on the power switch of the sub standard light source and wait for approx. 5 minutes until the lamp stabilizes.



Fig. F-9

(8) Execute the Instrumental Response command from the Utility menu. The Instrumental Response dialog box will open as shown in Fig. F-10.

Select "EM [500 to 900 nm]." (Refer to ① in Fig. F-10.)

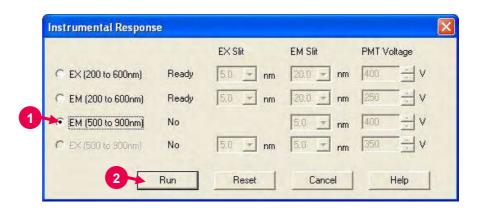


Fig. F-10

(9) Click the **Run** button. (Refer to ② in Fig. F-10.) A message box "Set the sub standard light source." will appear. Click the **OK** button. (Refer to ③ in Fig. F-11.)

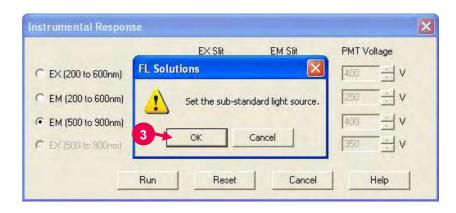


Fig. F-11

(10) The instrument starts EM wavelength scan (from 498.6 to 901.4 nm).

At this stage, the following measuring conditions are automatically set.

• Scan mode : Emission

• Data mode : Luminescence

EM slit : 5.0 nm
Scan speed : 60 nm/min
PMT voltage: 400 V
Response : Auto

(11) Upon completion of measurement, the conditions set before measurement are retrieved.

Now, the instrument response on the emission side has been measured and stored. Take out the sub standard light source and diffuser.

## F.4 Measurement of Instrumental Response in Long Wavelength Range on Excitation Side

#### F.4.1 Overview

When a corrected spectrum in the long wavelength area of 600 nm or more is required on the excitation side, the instrumental response in the long wavelength range on the excitation side is read for the correction with reference to the instrumental response obtained at the long wavelength on the emission side.

This measurement is enabled only when the instrumental response at the long wavelength on the emission side is available. The correction range by it is 500 to 900 nm and, when combined with an instrumental response in the short wavelength range, spectrum correction is available over the entire wavelength range (200 to 900 nm) on the excitation side.

## F.4.2 Operating Procedures

- (1) Close the cover of the sample compartment.
- (2) Click the (analysis method) button on the Measurement toolbar. A box for setting your analysis method will appear.
- (3) Select the General tab. On the General tab page, specify "Wavelength scan" for the measurement mode.
- (4) Select the Instrument tab.
- (5) Set "Fluorescence" for the data mode and "350 V" for the photomultiplier voltage.
- (6) Execute the Zero Adjust command from the Spectrophotometer menu to calibrate the zero point.

(7) Set the diffuser into the cell holder in the sample compartment. (Refer to Fig. F-12.)

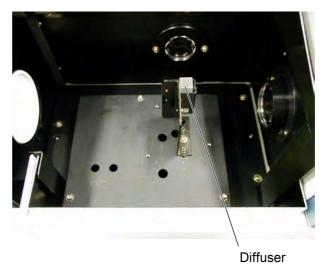


Fig. F-12

(8) Execute the Instrumental Response command from the Utility menu. The Instrumental Response dialog box will open as shown in Fig. F-13.

Select "EX [500 to 900 nm]." (Refer to ① in Fig. F-13.)

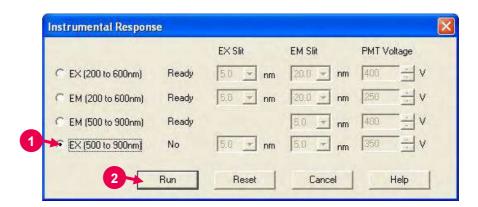


Fig. F-13

(9) Click the **Run** button. (Refer to ② in Fig. F-13.) A message box "Set diffuser to cell holder." will appear. Click the **OK** button. (Refer to ③ in Fig. F-14.)

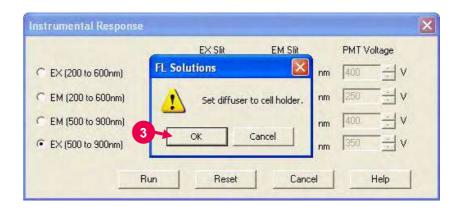


Fig. F-14

(10) The EX and EM wavelengths are set at 498.6 nm and the instrument starts synchronous wavelength scan (from 498.6 to 901.4 nm).

At this stage, the following measuring conditions are automatically set.

• Scan mode : Emission

• Data mode : Luminescence

EX slit : 5.0 nm
EM slit : 5.0 nm
Scan speed : 60 nm/min
PMT voltage: 350 V
Response : Auto

(11) Upon completion of measurement, the conditions set before measurement are retrieved.

Now, the instrument response on the emission side has been measured and stored. Take out the diffuser.

## F.5 Resetting of Instrumental Response

(1) Click the **Reset** button in the Instrumental Response dialog box. (Refer to ① in Fig. F-15.)

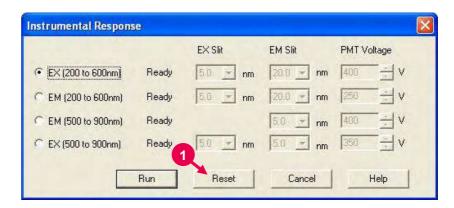


Fig. F-15

(2) A message box "Erase instrumental response on excitation/emission side. OK?" will appear. Click the **OK** button. (Refer to ② in Fig. F-16.)

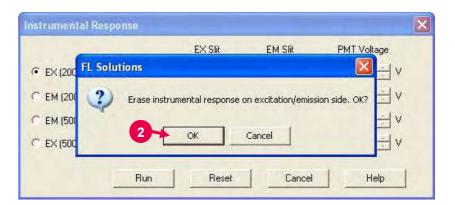


Fig. F-16

(3) Now, the instrumental response has been erased.

# APPENDIX G PARAMETER SETTING RANGE FOR ANALYSIS METHOD

# G.1 General Tab: Common to all measurement modes

Parameter Name	Setting Range
Measurement	Wavelength scan, Time scan, Photometry, 3-D scan, 3-D Time scan
Operator	Up to 20 characters
Comments	Up to 255 characters
Use sample table	ON, OFF

ON: Checked OFF: Unchecked

#### G.1.1 Wavelength Scan: Instrument Tab

Parameter Name	Setting Range			
Scan mode	Excitation, Emissi	ion, Synchronous		
Data mode	Fluorescence, Lu	minescence, Phospho	prescence	
Chopping speed	10 Hz, 20 Hz, 40	Hz		
EX WL			-	
	Scan Mode	Input Range	_	
	Excitation	0, 200 to 900 nm		
	Synchronous	200 to 890 nm	-	
EX Start WL	Input Range: 200 to 890 nm			
EX End WL	Input Range: 210 to 900 nm			
EM WL	Input Range: 0, 200 to 900 nm			
EM Start WL	Input Range: 200 to 890 nm			
EM End WL	Input Range: 21	0 to 900 nm		

(cont'd)

	I	(cont d)
Parameter Name		Setting Range
Scan speed (unit: nm/min)	is indicated. When Phosphoresconfollowing. Chopping speed is in	
	F-7000 (40 Hz)	F-7000 (20 Hz/10 Hz)
	240 60 30	60 30
Delay	0 to 9999 s	
EX Slit (unit: nm)	1.0 2.5 5.0 10.0 20.0	
EM Slit (unit: nm)	1.0 2.5 5.0 10.0 20.0	
PMT Voltage	Setting range: 250	, 400, 700, 950 V
	0 to 1000 V when "P	MT Voltage 0-1000V" is turned ON (checked).
PMT Voltage 0 - 1000 V	ON, OFF	
Response (unit: s)	0.002 0.004 0.01 0.05 0.1 0.5 2.0 4.0 8.0 Auto	
Corrected spectra	ON, OFF	
Shutter control	ON, OFF	
Replicates	1 to 99	
Cycle time	0.0 to 180.0 min	

ON : Checked OFF: Unchecked

## G.1.2 Wavelength Scan: Monitor Tab

Parameter Name	Setting Range
Y Axis: Max	-1199999999999999999999999999999999999
Y Axis: Min	-120000000.0 to 119999999.9
Open data processing window after data acquisition	ON, OFF
Open data processing window after acquisition (Overlay)	ON, OFF
Print report after data acquisition	ON, OFF
Overlay	ON, OFF

### G.1.3 Wavelength Scan: Processing Tab

Parameter Name	Setting Range
CAT	ON, OFF
Processing choices	Savitsky-Golay, Smooth, Mean Smooth, Median Smooth, Derivative
Peak Finding: Integration method	Rectangular, Trapezoid, Romberg
Peak Finding: Threshold	0.001 to 1000
Peak Finding: Sensitivity	1, 2, 4, 8

## G.1.4 Wavelength Scan: Report Tab

Parameter Name	Setting Range			
Output	Print Report, Use Microsoft(R) Excel, Use print generator sheet			
Orientation	Portrait, Landsc	ape		
Print items: Include date	ON, OFF			
Print items: Include method	ON, OFF			
Print items: Include graph	ON, OFF			
Print items: Include data listing	ON, OFF			
	ON: "Constant	" or "Selec	ct data" is selectable	
	When "Constan	t" is specif	ïed:	
	Parameter	S	Setting Range	
	Data interval	Minimum sampling interval to measuring wavelength interval		
	Data start Wavelength range on Instrument tab page			
	Data end Wavelength range on Instrument tab page			
	When "Select data" is specified: Selectable from 12 wavelengths.			
Print items: Include peak table	ON, OFF			
	ON: Items to b	e output p	eak table are selecta	able.
	ltem		Setting Range	
	Peak WL/Peak data ON, OFF Start WL/End WL ON, OFF Valley WL/Valley data ON, OFF Peak Area ON, OFF		ON, OFF	
			ON, OFF	
			ON, OFF	•
			ON, OFF	•
				-

### G.2.1 Time Scan: Instrument Tab

Parameter Name	Setting Range				
Data mode	Fluorescence, Luminescence, Phosphorescence, Phosphorescence Life Time, Phosphorescence Life Time (short)				
Chopping speed	10 Hz, 20 Hz,	10 Hz, 20 Hz, 40 Hz			
EX WL	Input Range:	0, 200	to 900 nm		
EM WL	Input Range:	0, 200	to 900 nm		
Time unit	s, ms				
Scan time	Time Unit	Inp	ut Range		
	S	10 to 9	9000 s		
	ms	100 to	9999 ms		
	When Phosph mode: Chopping S		scan T		s specified for data
	40 Hz	<b>P</b>	Fixed at 20		_
	20 Hz		Fixed at 40		_
	10 Hz		Fixed at 80		- -
Delay	0 to 9999 s				
EX Slit (unit: nm)	1.0 2.5 5.0 10.0 20.0				
EM Slit (unit: nm)	1.0 2.5 5.0 10.0 20.0				
PMT Voltage	Setting range:	250, 4	100, 700, 950	V	
	0 to 1000 V wh	nen "PM	IT Voltage 0-1	1000V" is	turned ON (checked).
PMT Voltage 0 - 1000 V	ON, OFF				
Response (unit: s)	0.002 0.004 0.01 0.05 0.1 0.5 2.0 4.0 8.0				
Shutter control	ON, OFF				
Replicates	1 to 99				
Cycle time	0.0 to 180.0 m	in			

### G.2.2 Time Scan: Monitor Tab

Parameter Name	Setting Range
Y Axis: Max	-119999999.9 to 120000000.0
Y Axis: Min	-120000000.0 to 119999999.9
X Axis: Max (unit: s)	0.1 to 9999.0
X Axis: Min (unit: s)	0.0 to 9998.9
Open data processing window after data acquisition	ON, OFF
Open data processing window after acquisition (Overlay)	ON, OFF
Print report after data acquisition	ON, OFF
Overlay	ON, OFF

## G.2.3 Time Scan: Processing Tab

Parameter Name	Setting Range		
Processing choices	Savitsky-Golay Smooth, Mean Smooth, Median Smooth, Derivative		
Peak Finding: Integration method	Rectangular, Trapezoid, Romberg		
Peak Finding: Threshold	0.001 to 1000		
Peak Finding: Sensitivity	1, 2, 4, 8		
Kinetics: Start time	0 to 3599 s, 0 to 9998 ms		
Kinetics: End time	0 to 3600 s, 1 to 9999 ms		
Kinetics: K-Factor	-9999 to -0.001, 0.001 to 9999		

## G.2.4 Time Scan: Report Tab

Parameter Name	Setting Range				
Output	Print Report, Use Microsoft(R) Excel, Use print generator sheet				
Orientation	Portrait, Landscape				
Print items: Include date	ON, OFF				
Print items: Include method	ON, OFF				
Print items: Include graph	ON, OFF				
Print items: Include data listing	ON, OFF				
	ON: "Constant" o	or "Select o	lata" is selectable.		
	When "Constant" is specified:				
	Parameter	S	Setting Range		
	Data interval	Minimum sampling interval to measuring scan time  Scan time range on Instrument tab page  Scan time range on Instrument tab page		_	
	Data start				
	Data end				
	When "Select data" is specified: Selectable from 12 time points (in measurement range).				
Print items: Include peak table	ON, OFF				
	ON: Items to be output onto peak table are selectable.				
	Item		Setting Range		
	Peak time/Peak data		ON, OFF		
	Start time/End	time	ON, OFF		
	Valley time/Valley data		ON, OFF		
	Peak Area	ON, OFF			
Print items: Include kinetics	ON, OFF				

#### G.3.1A Photometry: Quantification Tab (Quantification type: Wavelength)

Parameter Name	Setting Range			
Quantification type	Wavelength, Peak area, Peak height, Derivative, Ratio			
Calibration type	None, 1st ord	ler, 2nd o	rder, 3rd order, Seg	gmented
Number of wavelengths	Calibration	п Туре	Setting Range	-
	None		1 to 6	_
	1st order	•	1 to 3	_
	2nd orde	r		
	3rd orde	٢		
	Segmen	ted		_
Concentration unit	Up to 8 characters			
Manual calibration	ON, OFF			
	ON: Factors are specifiable.			
	Factor Setting Range			
	A0 to A3	-100000	to 100000	
Force curve through zero	ON, OFF			
Digit after decimal point	0 to 3			
Lower concentration limit	0 to 1e+009			
Upper concentration limit	0 to 1e+009			

# G.3.1B Photometry: Quantification Tab (Quantification type: Peak area, Peak height)

Parameter Name	Setting Range		
Quantification type	Wavelength, Peak area, Peak height, Derivative, Ratio		
Calibration type	None, 1st ord	der, 2nd order, 3rd order	r, Segmented
Peak apex	Wavelength i	range on Instrument tab	page ± 0.1 to 700.0 nm
Concentration unit	Up to 8 chara	acters	
Manual calibration	ON, OFF		
	ON: Factors	are specifiable.	
	Factor	Setting Range	-
	A0 to A3	-100000 to 100000	<u>.</u>
Force curve through zero	ON, OFF		
Digit after decimal point	0 to 3		
Lower concentration limit	0 to 1e+009		
Upper concentration limit	0 to 1e+009		
Integration method	Rectangular, Trapezoid, Romberg		
Threshold	0.001 to 1000		
Sensitivity	1, 2, 4, 8		

#### G.3.1C Photometry: Quantification Tab (Quantification type: Derivative)

Parameter Name	Setting Range		
Quantification type	Wavelength, Peak area, Peak height, Derivative, Ratio		
Calibration type	None, 1st ord	der, 2nd order, 3rd order	, Segmented
Peak apex	Wavelength	range on Instrument tab	page
Concentration unit	Up to 8 chara	acters	
Manual calibration	ON, OFF		
	ON: Factors	are specifiable.	
	Factor	Setting Range	
	A0 to A3	-100000 to 100000	
Force curve through zero	ON, OFF		
Digit after decimal point	0 to 3		
Lower concentration limit	0 to 1e+009 (1000000000)		
Upper concentration limit	0 to 1e+009 (1000000000)		
Derivative order	2, 4		
Smoothing order	2 to 4		
Number of points	5 to 101		

#### G.3.1D Photometry: Quantification Tab (Quantification type: Ratio)

Parameter Name	Setting Range
Quantification type	Wavelength, Peak area, Peak height, Derivative, Ratio
Subtract background [Wavelength 3]	ON, OFF
K-Factor 0	-9999 to -0.001, 0.001 to 9999
K-Factor 1	
K-Factor 2	
Operand	+ (plus), - (minus), * (times)

# G.3.2A Quantification: Instrument Tab (Quantification type: Wavelength, Ratio)

Parameter Name	Setting Range
Data mode	Fluorescence, Luminescence, Phosphorescence
Chopping speed	10 Hz, 20 Hz, 40 Hz
Wavelength mode	EX WL Fixed, EM WL Fixed, Both WL Fixed
Fixed WL	Input Range: 0, 200 to 900 nm
WL 1 to WL 6	Input Range: 0, 200 to 900 nm
EX Slit (unit: nm)	1.0 2.5 5.0 10.0 20.0
EM Slit (unit: nm)	1.0 2.5 5.0 10.0 20.0
PMT Voltage	Setting range: 250, 400, 700, 950 V
	0 to 1000 V when "PMT Voltage 0-1000V" is turned ON (checked).
PMT Voltage 0 - 1000 V	ON, OFF
Auto statistic calc. number	2 to 4000
Replicates	1 to 20
Integration time	0.1 to 10.0 s
Delay	0 to 9999 s

# G.3.2B Photometry: Instrument Tab (Quantification type: Peak area, Peak height, Derivative)

Scan mode I	Setting Range				
	Excitation, Emission, Synchronous				
Data mode	Fluorescence, Luminescence, Phosphorescence				
Chopping speed	10 Hz, 20 Hz, 40 Hz				
EX WL	Scan Mode		Input Ran	ge	•
-	Excitation		0, 200 to 900	nm	-
-	Synchronous		200 to 890 nr	n	- -
EX Start WL	Input Range:	200	to 890 nm		
EX End WL	Input Range:	210	to 900 nm		
EM WL I	Input Range:	0, 2	.00 to 900 nm		
EM Start WL	Input Range:	200	to 890 nm		
EM End WL	Input Range:	210	to 900 nm		
	60000 30000 12000 2400 1200 240 60 30 When Synchrol				n mode,
\ \frac{1}{2}	When Phosphorescence is specified for data mode, select from the following. Chopping speed is in parentheses.				
-	(40 Hz) (20 Hz/10 Hz)				
	240 60 30	·	60 30		
Delay (	0 to 9999 s				
EX Slit (unit: nm)	1.0 2.5 5.0 10.0 20.0				
EM Slit (unit: nm)	1.0 2.5 5.0 10.0 20.0				
	Setting range: 250, 400, 700, 950 V 0 to 1000 V when "PMT Voltage 0-1000V" is turned ON (checked).				
	ON, OFF				

(cont'd)

Parameter Name	Setting Range
Response (unit: s)	0.002 0.004
	0.004
	0.05
	0.1
	0.5
	2.0
	4.0
	8.0
	Auto
Corrected spectra	ON, OFF
Shutter control	ON, OFF
Replicates	1 to 20
Auto statistic calc. number	2 to 4000

#### G.3.3 Photometry: Standard Tab

Parameter Name	Setting Range
Number of Samples	1 to 20
Sample	Up to 40 characters
Comment	Up to 255 characters
Concentration	0 to 100000000

#### G.3.4 Photometry: Monitor Tab

Parameter Name	Setting Range
Y Axis: Max.	-119999999.9 to 120000000.0
Y Axis: Min.	-120000000.0 to 119999999.9
Open data processing window after data acquisition	ON, OFF
Print report after data acquisition	ON, OFF

#### G.3.5 Photometry: Report Tab

Parameter Name	Setting Range
Output	Print Report, Use Microsoft(R) Excel, Use print generator sheet
Print items: Include date	ON, OFF
Print items: Include method	ON, OFF
Print items: Include calibration curve	ON, OFF
Print items: Include standard data	ON, OFF
Print items: Include standard comment	ON, OFF
Print items: Include calibration data	ON, OFF
Print items: Include sample data	ON, OFF
Print items: Include sample comment	ON, OFF

#### G.4.1 3-D Scan: Instrument Tab

Parameter Name		Setting Range	
Data mode	Fluorescence, Phosphorescence		
Chopping speed	10 Hz, 20 Hz,	, 40 Hz	
EX Start WL	Input Range:	200 to 850 nm	
EX End WL	Input Range:	250 to 900 nm	
EX Sampling interval	Input Range:	1 to 50 nm	
EM Start WL	Input Range:	200 to 850 nm	
EM End WL	Input Range:	250 to 900 nm	
EM Sampling interval	Input Range:	1 to 10 nm	
Scan speed (unit: nm/min)	1200 nm/min When Phosp	onous is specified for scan mode, or less is indicated. horescence is specified for data mode, select wing. Chopping speed is in parentheses.	
	(40 Hz)	(20 Hz/10 Hz)	
	240 60 30	60 30	
EX Slit (unit: nm)	1.0 2.5 5.0 10.0 20.0		
EM Slit (unit: nm)	1.0 2.5 5.0 10.0 20.0		
PMT Voltage		: 250, 400, 700, 950 V when "PMT Voltage 0-1000V" is turned ON	
PMT Voltage 0 - 1000 V	ON, OFF		
Response (unit: s)	0.002 0.004 0.01 0.05 0.1 0.5 2.0 4.0 8.0 Auto		

Parameter Name	Setting Range
Corrected spectra	ON, OFF
Shutter control	ON, OFF

ON : Checked OFF: Unchecked

#### G.4.2 3-D Scan: Monitor Tab

Parameter Name	Setting Range		
Y Axis: Max	-119999999.9 to 120000000.0		
Y Axis: Min	-120000000.0 to 119999999.9		
Contour interval	0.001 to 1000.000		
Open data processing window after data acquisition	ON, OFF		
Print report after data acquisition	ON, OFF		

#### G.4.3 3-D Scan: Processing Tab

Parameter Name	Setting Range	
Peak Finding: Integration method	Rectangular	
Peak Finding: Threshold	0.001 to 1000	
Peak Finding: Sensitivity	1, 2, 4, 8	

#### G.4.4 3-D Scan: Report Tab

Parameter Name	Setting Range			
Output	Print Report, Use Microsoft(R) Excel, Use print generator sheet			
Orientation	Portrait, Lands	cape		
Print items: Include date	ON, OFF			
Print items: Include instrument	ON, OFF			
Print items: Include graph	ON, OFF			
	ON: Any of the	following items is selectable.		
	<ul> <li>Contour</li> </ul>			
	Bird's-eye view			
	Contour and Bird's-eye view			
	Contour and Spectrum			
Print items: Include peak table	ON, OFF	ON, OFF		
Print items: Data listing* <sup>1</sup>	ON, OFF			
	ON: Setting can be made for excitation and emission spectra.			
	Parameter	Setting Range		
	Interval	Minimum data interval to measuring wavelength interval		
	Start	Wavelength range on Instrument tab page		
	End	Wavelength range on Instrument tab page		

<sup>\*1:</sup> Valid when "Use Microsoft(R) Excel" or "Use print generator sheet" is specified for Output. Always turned ON when "Use print generator sheet" is specified.

#### G.5.1 3-D Time Scan: Instrument Tab

Parameter Name	Setting Range		
Scan mode	Excitation, Emission, Synchronous		
Data mode	Fluorescence, Luminescence, Phosphorescence		
Chopping speed	10 Hz, 20 Hz, 40 Hz		
EX WL	Scan Mode	Input Range	
	Excitation	0, 200 to 900 nm	
	Synchronous	200 to 890 nm	
EX Start WL	Input Range:	200 to 890 nm	
EX End WL	Input Range:	210 to 900 nm	
EM WL	Input Range:	0, 200 to 900 nm	
EM Start WL	Input Range:	200 to 890 nm	
EM End WL	Input Range:	210 to 900 nm	
Scan speed (unit: nm/min)	60000 30000 12000 2400 1200 240 60 30 When Synchronous is specified for scan mode,		
	1200 nm/min or less is indicated.  When Phosphorescence is specified for data mode, select		
	from the following	ng. Chopping speed is in parentheses.	
	(40 Hz)	(20 Hz/10 Hz)	
	240 60 30	60 30	
EX Slit (unit: nm)	1.0 2.5 5.0 10.0 20.0		
EM Slit (unit: nm)	1.0 2.5 5.0 10.0 20.0		
PMT Voltage	250, 400, 700, 950 V 0 to 1000 V when "PMT Voltage 0-1000V" is turned ON (checked).		
PMT Voltage 0 - 1000 V	ON, OFF		

(cont'd)

Parameter Name	Setting Range
Response (unit: s)	0.002 0.004 0.01 0.05 0.1 0.5 2.0 4.0 8.0 Auto
Corrected spectra	ON, OFF
Shutter control	ON, OFF
Replicates	2 to 99
Cycle time	2 to 3600 s

ON: Checked OFF: Unchecked

#### G.5.2 3-D Time Scan: Monitor Tab

Parameter Name	Setting Range		
Y Axis: Max	-119999999.9 to 120000000.0		
Y Axis: Min -120000000.0 to 1199			
Contour interval	0.001 to 1000		
Open data processing window after data acquisition	ON, OFF		
Print report after data acquisition	ON, OFF		

#### G.5.3 3-D Time Scan: Processing Tab

Parameter Name	Setting Range
Peak Finding: Integration method	Rectangular
Peak Finding: Threshold	0.001 to 1000
Peak Finding: Sensitivity	1, 2, 4, 8

#### G.5.4 3D Time Scan: Report Tab

Parameter Name	Setting Range			
Output	Print Report, Use Microsoft(R) Excel, Use print generator sheet			
Orientation	Portrait, Land	scape		
Print items: Include date	ON, OFF			
Print items: Include instrument	ON, OFF			
Print items: Include graph	ON, OFF			
	ON: Any of the following items are selectable.			
	Contour			
	Bird's-eye view			
	Contour and Bird's-eye view			
	Contour and Spectrum			
Print items: Data listing*1	ON, OFF			
	ON: Setting can be made for time scan and wavelength scan spectra.			
	Parameter	Setting Range		
	Interval	Minimum data interval to measuring wavelength interval		
	Start	Wavelength range on Instrument tab page		
	End	Wavelength range on Instrument tab page.		

<sup>\*1:</sup> Valid when "Use Microsoft(R) Excel" or "Use print generator sheet" is specified for Output. Always turned ON when "Use print generator sheet" is specified.

## APPENDIX H SAMPLING INTERVAL

## H.1 Wavelength Scan

The data sampling interval is set as follows according to the scan speed.

[Data mode = Fluorescence/Luminescence]

Scan Speed (nm/min)	Sampling Interval (nm)		
60000	2.0		
30000	1.0		
12000	0.2		
2400	0.2		
1200	0.2		
240	0.2		
60	0.2		
30	0.2		

[Data mode = Phosphorescence, Chopping speed = 40 Hz]

Scan Speed (nm/min)	Sampling Interval (nm)		
240	0.2		
60	0.2		
30	0.2		

[Data mode = Phosphorescence, Chopping speed = 20/10 Hz]

Scan Speed (nm/min)	Sampling Interval (nm)	
60	0.2	
30	0.2	

## H.2 Time Scan

The data sampling interval is set as follows according to the scan time.

[Data mode = Fluorescence/Luminescence/Phosphorescence/ Phosphorescence Life Time]

	Sampling Interval [ms]			
Scan Time [s]	Fluorescence/Luminescence/ Phosphorescence	Phosphorescence Mode/Chopping Speed		
	Life Time Mode	40 Hz	20 Hz	10 Hz
0.1 ~ 1	1	50	100	200
~ 3	1	50	100	200
~ 7	2	50	100	200
~ 18	5	50	100	200
~ 36	10	50	100	200
~ 72	20	50	100	200
~ 180	50	50	100	200
~ 360	100	100	100	200
~ 720	200	200	200	200
~ 1800	500	500	500	1000
~ 3600	1000	1000	1000	1000
~ 7200	2000	2000	2000	2000
~ 9000	5000	5000	5000	5000

[Data mode = Phosphorescence Life Time (short)]

Chopping Speed [Hz]	Scan Time [ms]	Sampling Interval [µs]
40	20	10
20	40	20
10	80	40

### H.3 3-D Time Scan

The data sampling interval is set as follows according to the scan speed.

[Data mode = Fluorescence/Luminescence]

Scan Speed (nm/min)	Sampling Interval (nm)
60000	2.0
30000	1.0
12000	1.0
2400	1.0
1200	1.0
240	1.0
60	1.0
30	1.0

[Data mode = Phosphorescence, Chopping speed = 40 Hz]

Scan Speed (nm/min)	Sampling Interval (nm)
240	1.0
60	1.0
30	1.0

[Data mode = Phosphorescence, Chopping speed = 20/10 Hz]

Scan Speed (nm/min)	Sampling Interval (nm)
60	1.0
30	1.0

# APPENDIX I PARAMETER SETTING RANGE ON OPTIONS WINDOW

# I.1 Display Tab

Parameter Name	Setting Range
Show trace display	ON, OFF
Show gridlines with increment	ON, OFF ON: Gridline width is selectable from Wide, Middle and Narrow.
Display items: Wavelength scan	Show graph only, Show graph plus peak table
Display items: Time scan	Show graph only, Show graph plus kinetic data, Show graph plus peak table
Display items: 3-D Scan	Show contour plus EX, EM spectrum, Show contour plus EX, EM spectrum plus peak table
Display items: Intracellular cation scan	Show graph only, Show graph plus kinetic data, Show graph plus mean, Show graph plus point
Display items: Intracellular cation spectrum	Show fluorescence only, Show fluorescence plus ratio, Show fluorescence plus concentration, Show fluorescence plus ratio plus concentration
Display items: Polarization time scan	Show Graph only, Show graph plus kinetic data, Show graph plus point
Display items: Polarization spectrum	Show fluorescence only, Show fluorescence plus polarization degree, Show fluorescence plus anisotropy, Show fluorescence plus polarization degree plus anisotropy

# I.2 Processing Tab

Parameter Name	Setting Range
Integration method	Rectangular, Trapezoid, Romberg
Calibration formula	Data = f(Conc), Conc = f(Data)
Data output to peak table	ON, OFF
Smoothing: Processing	Savitsky-Golay, Mean, Median
Smoothing: Order	2 to 4
Smoothing: Number of points	5 to 101
Derivative: Smoothing order	2 to 4
Derivative: Smoothing order	5 to 101

# I.3 Analysis Condition Tab

Parameter Name	Setting Range
Integration time for current data	0.1 to 10.0 s
Change Instrumental Response Condition	ON, OFF

# APPENDIX J ERROR MESSAGE LIST

Error Message	Probable Cause	Remedy
ROM Error	ROM error has occurred.	Contact our local service office.
RAM Error	RAM error has occurred.	Contact our local service office.
Lamp is not lighted	Lamp extinguished during initialization.	Replace the lamp.
Power interruption	Momentary power failure detected at instrument.	Check the connection of USB cable, and start up PC and spectrophotometer again. If error recurs, contact our local service office.
Zero adjust error!	Faulty chopper operation	Contact our local service office.
Auto Pre-scan error! Peak not Found!	Peak was not detected in auto pre-scan.	Peak may be detected by widening the slit or increasing photomultiplier voltage.
Pre-scan error! Peak not Found!	Peak was not detected in normal pre-scan.	Peak may be detected by widening the slit or increasing photomultiplier voltage.
Wavelength Initialization error (EX)!	Initialize position of EX side wavelength feed screw cannot be detected.	Contact our local service office.
Wavelength Initialization error (EM)!	Initialize position of EM side wavelength feed screw cannot be detected.	Contact our local service office.
Shutter Initialization error	Initialize position of shutter cannot be detected.	Contact our local service office.
EX Slit initialization error!	Initialize position of EX side slit cannot be detected.	Contact our local service office.
EM Slit initialization error!	Initialize position of EM side slit cannot be detected.	Contact our local service office.
Chopper error!	Sync signal from chopper cannot be detected.	Contact our local service office.
Progam status error	Abnormality detected in spectrophotometer main unit program.	Contact our local service office.
The Xe lamp was turned off suddenly. Turn off the main switch of the spectrophotometer unit.	Xe lamp has burned out.	Replace with a new Xe lamp.

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