OLYMPUS®

User's Manual
[Operation/Maintenance Manual]

FLUOVIEW FV1200

BIOLOGICAL CONFOCAL LASER SCANNING MICROSCOPE

FV10-ASW

Notice

Thank you for your purchase of Olympus microscope at this time.

Retain this manual in an easily accessible place near a system for future reference.

AX8266

CONTENTS

IMPORTANT

CAUTION

FV1200 is a CLASS 3B laser product.
The procedures for using this system are classified as follows:

- Service
  "Service" means any adjustment or repair performed by service personnels who are provided the service training following to the service manual for this system.
  The performance has influence on the feature of this system, and there is a risk which unintended CLASS 3B laser light is emitted.
- Maintenance
  "Maintenance" means adjustment or other procedures performed by customers to maintain that this system functions properly.
- Operation
  "Operation" means all performance described in the user's manuals in this system.
  CLASS 3B laser light is only emitted from the objective lens during the actual execution.

The User's Manuals of this system consist of the following:
In order to maintain the full performance of this system and ensure your safety, be sure to read these user's manuals and the operating instructions for the laser unit and light source unit before use.

	User's Manual Sets	
	FV1200MPE	FV1200
FV1200MPE / FV1200 User's Manual [Laser Safety Guide]	○	○
FV1200MPE User's Manual [Safety Manual]	○	
FV1200 User's Manual [Safety Manual]	○	○
FV1200MPE User's Manual [Operation / Maintenance Manual]	○	
FV1200 User's Manual [Operation / Maintenance Manual]	○	○
FV1200 FV10-ASW User's Manual [Quick Start]	○	○

Also, we have prepared one service manual for this system as below. Technical personnels who perform the service are required to take the service training.

- FV1200MPE / FV1200 Service Manual

For customers using FV1000MPE or FV1000:
This manual contains the instructions for FV1200 and FV-10ASW (Ver.4.0 and later version).
If the instructions for FV1000MPE / FV1000 and (former version than Ver.4.0) are not covered by this manual,refer to the manual for FV1000MPE / FV1000 on your hand.

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Other brand names and product names are trademarks or registered trademarks of their respective owners.

Caution

If the system is used in a manner not specified by this manual, the safety of the user may be imperiled. In addition, the system may also be damaged. Always use the system as outlined in this instruction manual.

The following symbols are used to set off text in this instruction manual.

CAUTION : Indicates a potentially hazardous situation which, if not avoided, may result in minor or moderate injury or damage to the equipment or other property. It may also be used to alert against unsafe practices.

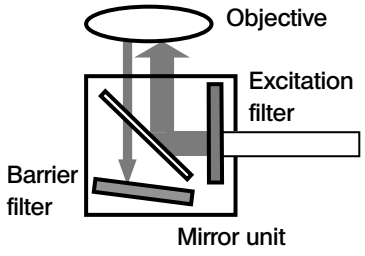
◎ : Indicates commentary (for ease of operation and maintenance).

Precautions and Notes for Glare Prevention

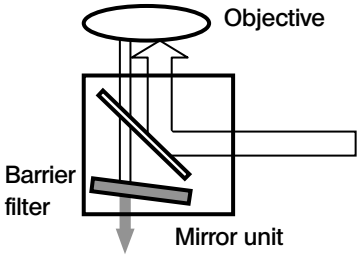
When excitation filter is removed from the fluorescence mirror unit

When the excitation filter is removed from the fluorescence mirror unit and attached on the excitation filter slider or excitation filter wheel on the side of the white lamp, **very glaring light (*1) may enter the eyepieces** in the following cases.

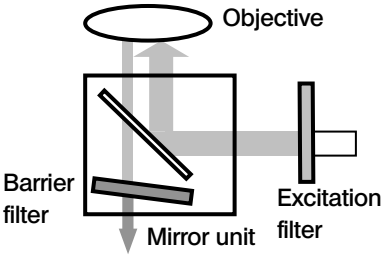
① When the illumination light from the white light lamp (*2) is input **without passing through an excitation filter.**
② When the illumination light from the white light lamp is input through **an excitation filter that does not match** the mirror unit type.



Ordinary mirror unit
Light transmitted through the excitation filter is blocked completely by the barrier filter.



Case of ① above
Among the white light, only the light transmitted through the barrier filter enters the observation light path.



Case of ② above
Light transmitted through the excitation filter is sometimes blocked incompletely by the barrier filter.

To avoid this problem, be sure to observe the following points.

- Before switching the mirror unit and excitation filter, be sure to close the shutter of the mirror unit turret.
- Before opening the shutter of the mirror unit turret, be sure to check the position index of the mirror unit to confirm that it matches the type of the excitation filter engaged in the illumination light path.

(*1) The light will not injure your eyes even if it enters your eyes. However, be sure to stop observation through the eyepieces, and engage the mirror unit and the excitation filter of appropriate combination in the light path to restart observation.
(*2) The white light lamp refers to all lamps for reflected light illumination including mercury and xenon burners.

CAUTION :Never use the above-mentioned mirror units ① and ② for the LSM observation.

I. SYSTEM OVERVIEW

On This Volume

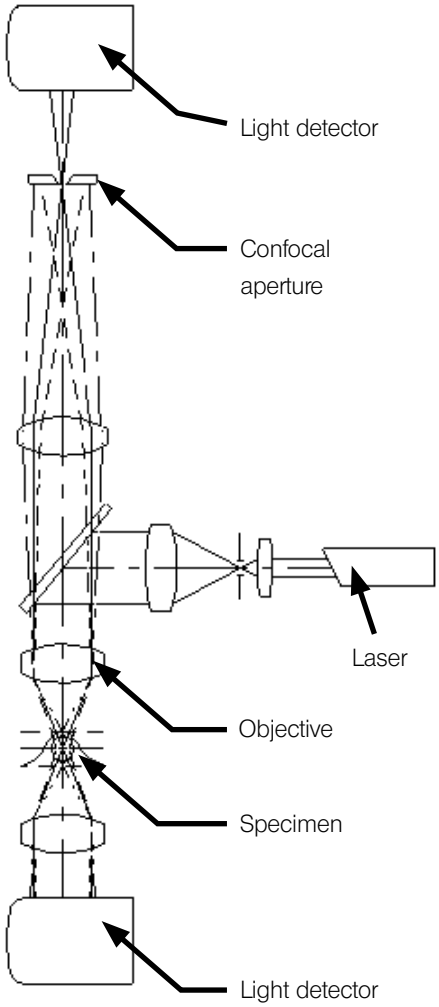
This volume describes the overview of the FLUOVIEW FV1200 system.
Please read this volume so that you can understand the system before use.

1 System Overview

OLYMPUS FV1200 is a confocal laser scanning biological microscope system featuring improved basic performances (sensor system, scanning system and illumination system performances) by considering the “live cell observations”, with which long hours of stable measurement of weak fluorescence is required.
This microscope is equipped with 3 fluorescence channels, 3 lasers and AOTF to meet various applications in a wide range of advanced research fields.

1-1 Principles

A laser scanning microscope converges the laser beam into a small spot using an objective and scans the specimen in the X-Y direction using the laser beam. The microscope then captures the fluorescent light and reflected light from the specimen using light detectors and outputs the specimen image on an image monitor.



As shown in this figure, the confocal optics incorporates a confocal aperture on the optically conjugate position (confocal plane) with the focus position to eliminate light from other part than the focus position. This causes the extraneous light to be viewed as darkness in the observation image, it is possible to slice optically a tissue specimen that has thickness.
On the other hand, an ordinary optical microscope, the light from other part than the focus position is overlapped with the imaging light of the focus position so the image is blurred in overall.
The laser beam that has transmitted through the specimen is detected by the transmitted light detector and provides the transmitted image, which is not a confocal image.
However, when the fluorescence images of the transmitted and confocal images are combined, it is possible to obtain very important information on the specimen.

1-2 Features of the FV1200

1. The photon counting mode is provided to improve the sensitivity and S/N and to enable quantitative optical intensity measurement. Photon counting makes possible long hours of quantitative observation by completely eliminating analog-derived drift. The dynamic range in which photon counting is possible is expanded using a newly designed wideband head amplifier and processing circuitry.

2. High-speed imaging at 8 frames per sec. is made possible by fast galvano mirror. In addition, high-speed image acquisition is possible without stopping the Z-series motors used in the XYZ and XZ observations.

3. During long hours of time-lapse observation, a stable supply of excitation light is made possible thanks to the feedback control of the intensity of each laser. Together with the photon counting function, this function ensures the stability and quantitative nature of long-hour observations.

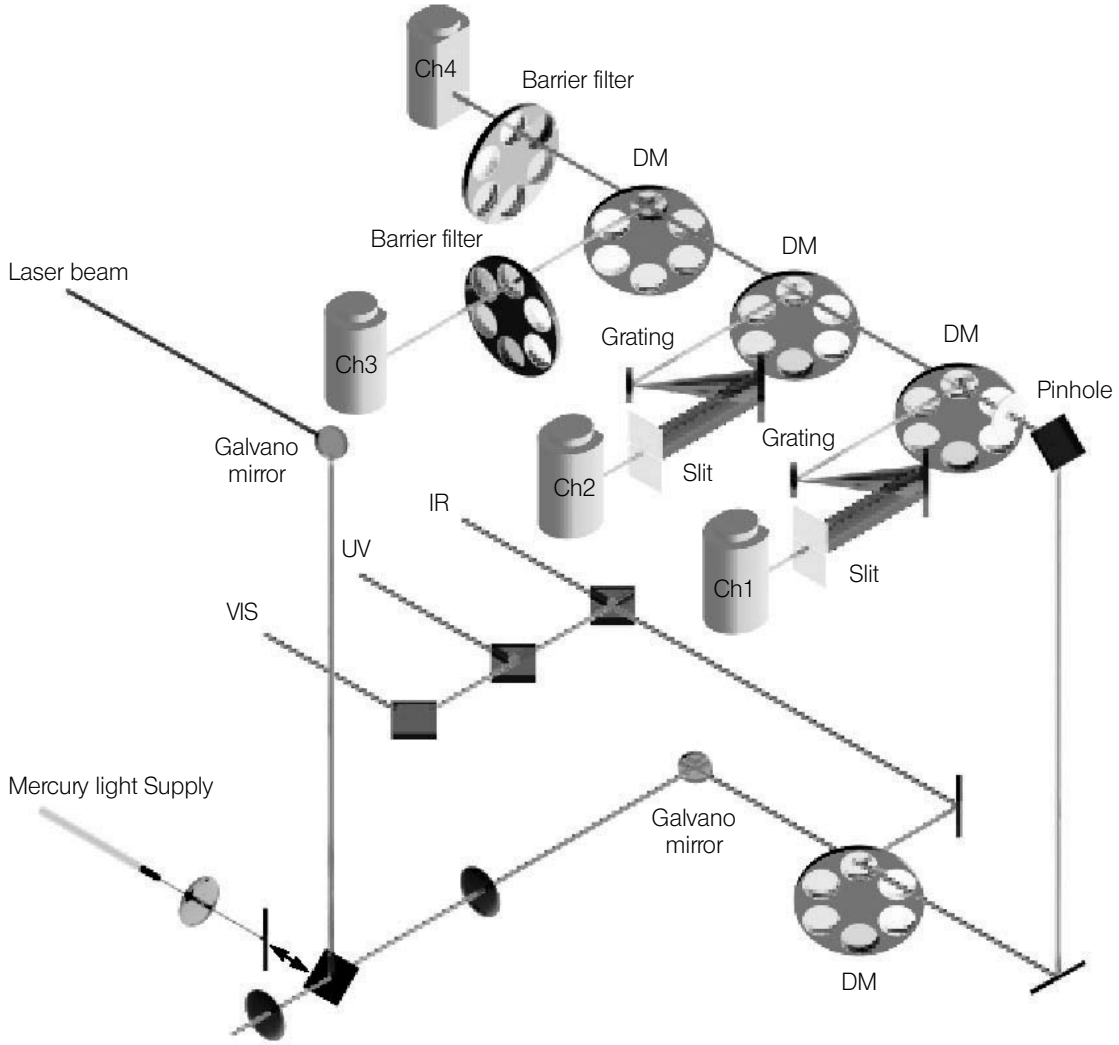
4. Three fluorescence channels, three lasers and AOTF are provided as standard to meet a large variety of applications.

5. With a fully-motorized scan unit and motorized microscope, the entire system is motorized so the scanning conditions including those of the optics can be saved and reproduced.

6. When an extension laser irradiation unit is used for photon activation aiming at causing discoloration, optical simulation or uncaging of the specimen, a system optimized for cell function analysis experiments can be built.

7. When the system incorporates the spectral detector unit that is composed of a 2-channel spectral detector and 1-channel filter, it is possible to set the detection conditions more flexibly, acquire the fluorescence spectral data and use the fluorescence isolation function.

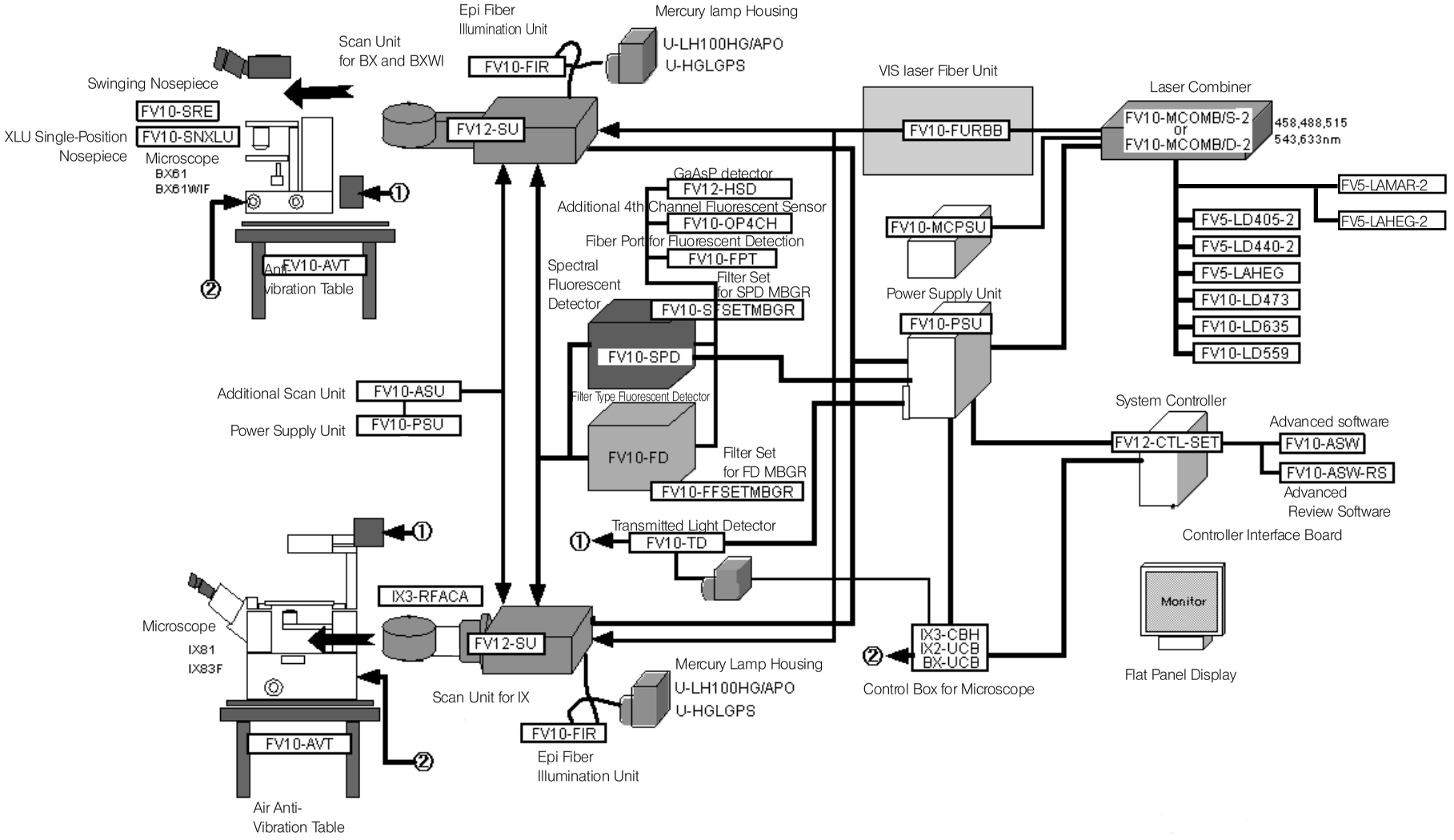
1-3 Optical Path Diagram



◎ This Optical Path Diagram present the system configuration where Additional 4th Channel Fluorescent sensor (FV10-OP4CH) is attached with Spectral Fluorescent Detector (FV10-SPD).

- When the Filter Type Fluorescent Detector (FV10-FD) is attached, instead of the Spectral Fluorescent Detector (FV10-SPD), the Grating and Slit engaged in the Ch1 and Ch2 light path is changed into the Barrier filter in this Diagram.
- When the GaAsP detector (FV12-HSD) is attached, instead of the Additional 4th Channel Fluorescent sensor (FV10-OP4CH), the Ch4 is changed into the HSD1 and HSD2 in this Diagram.

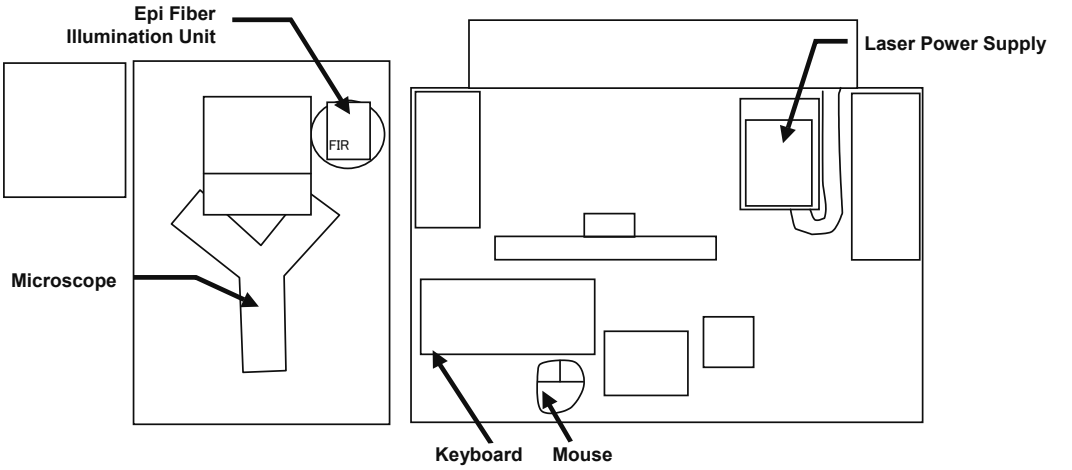
1 System Diagram



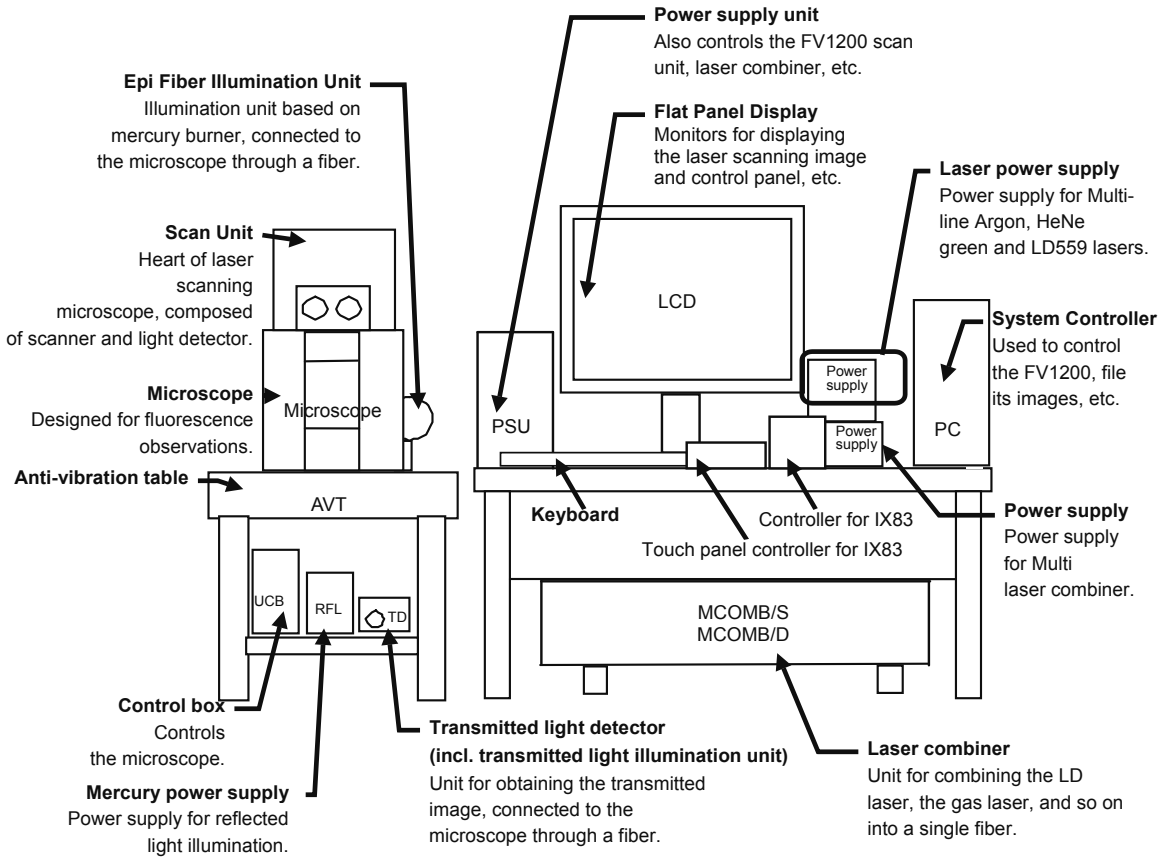
◎ When additional microscope units are required to the system, Olympus service engineer will perform the installation.

2 System Appearance and Functions

The applicable microscopes are the BX61/62TRF, BX61WIF, IX83P2ZF and IX81F.



Top View



Front View

II. PREPARATION For OPERATION

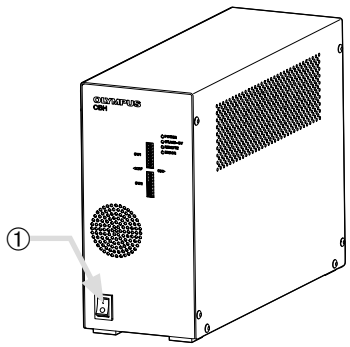
On This Volume

This volume describes the methods for preparation for operation of the FLUOVIEW FV1200 system.
After completing the preparations, activate the software and start observation by controlling the display on the monitor screen.
Please read this volume so that you can understand the system before use.

1 Preparation for Operation

1-1 Turning the Power On

IX3-CBH
Main switch

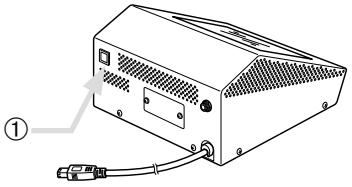


1.Set the power switches of the controller and the monitor to I (ON).

2.Set the power switches of the units of microscope to I (ON).
When using the IX83P2ZF microscope
◎ Set the main switches of the units to O (OFF) in the following order:
1) Main switch ① of the IX3-CBH
2) Main switch ① of the touch panel controller

3. Set the power switches of the following FV1200 units to I (ON).
   - Power Supply Unit FV10-PSU
   - Mercury Burner Power Supply Unit
   - Microscope Control Box BX-UCB , IX3-CBH or IX2-UCB

Touch panel controller
Main switch



4. Set the power switches of the lasers to I (ON).

FV5-LAMAR-2 Power Supply

4.1 Multi-line Argon laser: FV5-LAMAR-2
- Set the power switch ① to ON. (This starts the fan of the laser.)
- Turn the key ② to the ON position.
  It takes a few tens of seconds after the key ② is set to ON till the laser oscillation begins.

FV5-LAHEG-2 Power Supply

4.2 Helium-Neon Green Laser: FV5-LAHEG-2 or FV10-LAHEG230-2
- Turn the key ① to the I (ON) position.
  It takes a few tens of seconds after the key is set to ON till the laser oscillation begins.

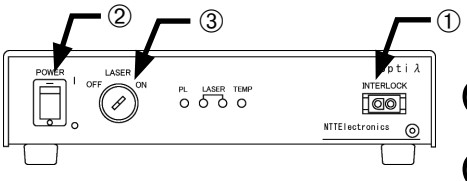
CAUTION **To ensure stable laser light output, it is recommended to warm up the laser power supply after turning it on. The warm-up period should be 10 minutes or more when using the Argon laser power supply and 30 minutes or more when using the Helium-Neon Green or Red laser power supply.**

LD405/440 laser power supply

4.4 LD405/440 laser: FV10-LD405/440

- Make sure that the provided shorting plug is attached to the remote interlock ① or that is connected to your equipment and the interlock is released.
- Set the power switch ② to ON.
- Turn the key ③ to the ON position.
- Set the shutter switch ④ OPEN.

CAUTION **The red lighting of the LASER EMISSION LED of the LD405/440 laser power supply indicates that the laser is oscillating. With a certain setup, the laser beam is output by simply setting the shutter switch ④ to ON.**



FV10-LD559 laser power supply

4.5 LD559 laser: FV10-LD559

CAUTION **Be careful as a stronger laser beam that the rated power may be emitted during calibration.**

CAUTION **Temperature error would occur if LASER key switch is turned OFF during calibration. In such a case, turn POWER switch to OFF and start the system again.**

CAUTION **In an event that a power control error (PL lamp lit in orange) occurred while the system is in use, turn both LASER key switch and POWER switch to OFF position and start the system again.**

- Verify that the remote interlock ① is set properly.
- Connect the power cord with power receptacle.
- Turn POWER switch ② to | - (ON). PL lamp will turn to lighting.
- Insert the provided key into LASER key switch ③ and turn it ON. LASER lamp will blink in red and calibration[1] will take place. When the calibration is completed, the blinking of LASER lamp will turn to lighting and a laser beam will be emitted in stable output condition. (When LASER key is turned ON, first time, after POWER switch is turned ON, the calibration will be executed.)

[1] About calibration

Calibration is performed to adjust the setting temperature of optical element inside the laser head to optimal condition.

1-2 Starting the Software

CAUTION **Turn on the microscope and power supply units before starting this software.**

1. Enter the user name and password to log in the Windows.

CAUTION **Log in using the user name given the Administrator's authority.**

OS	User name	Password
Windows7 (64bit)	olympus	olympus
Others	fluoview	fluoview

FV10-ASW

FV10-ASW 4.0

[FLUOVIEW] icon

2. Double-click the [FLUOVIEW] icon on the desktop.

◎ If more than one user uses the FV1200, each user should personally log in personally. For details, refer to Appendix E, "USER REGISTRATION OF FV1200" in Volume [OPERATION INSTRUCTIONS].

◎ It takes 20 to 30 seconds after the [FLUOVIEW] icon is double-clicked till the software starts up.

CAUTION
- **Images cannot be observed if the manual shutter of the fluorescence mirror unit is close. In this case, slide the shutter to the open position.**
- **When you lower the objective lens, please pay a careful attention so that the objective lens does not touch the specimen.**

1-3 Exiting from the Software

Exit from the application software and shut down Windows.

CAUTION **After exiting the application software, the light of mercury burner power supply unit may exposure to specimen. To avoid this, perform either of the followings,**

- **Close the manual shutter of the mercury burner power supply unit.**
- **Turn off the mercury burner power supply unit.**
- **Close the manual shutter of the fluorescence mirror unit (BX61WI or IX81).**

1-4 Turning the Power Off

Order to set the power switches of the units to O (OFF) is opposite to the order of turning the power switches on.

1.Set the power switch of the laser to O (OFF).

Power turning OFF when using of Multi-line Argon laser (FV5-LAMAR-2)
Turn the key to OFF position. Set the power switch to OFF.

In case of LD559 laser
Turn LASER key switch to OFF position and then, turn POWER switch to O – (OFF) position.

2.Set the power switches of the units of FV1200 to O (OFF).

3.Set the power switches of the units of microscope to O (OFF).
When using the IX83P2ZF microscope
◎ Set the main switches of the units to O (OFF) in the following order:
1) Main switch of the touch panel controller
2) Main switch of the IX3-CBH

4.Set the power switches of the controller and the monitor to O (OFF).

2 USING THE CONTROLS

◎ Only IX83 microscope combination is described.

2-1 Microscope Frame

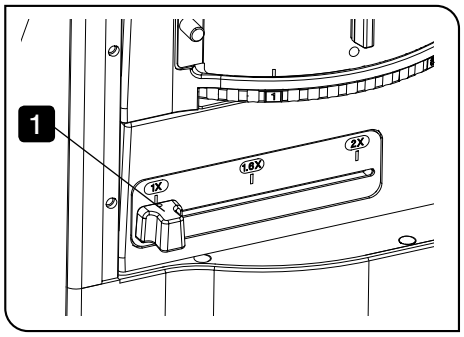
1 Motorized Revolving Nosepiece

The motorized revolving nosepiece can be rotated to switch the objective by controller U-MCZ, touch panel controller or controller.

2 Coded intermediate magnification changer IX3-CAS

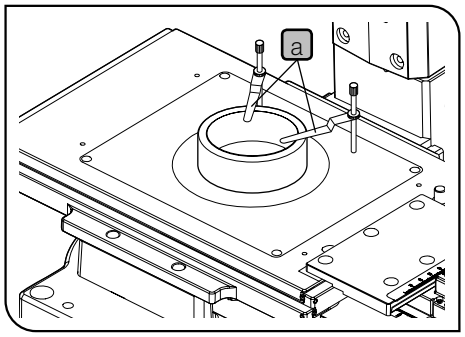
Use the coded intermediate magnification changer IX3-CAS to switch the observation magnification in the following 3 levels according to the objective lens magnification.

- 1X
- 1.6X
- 2X



1 Change the magnification by operating the changing slider.

2-2 Stage



1 Placing the Specimen

Place the specimen on the center of the stage.

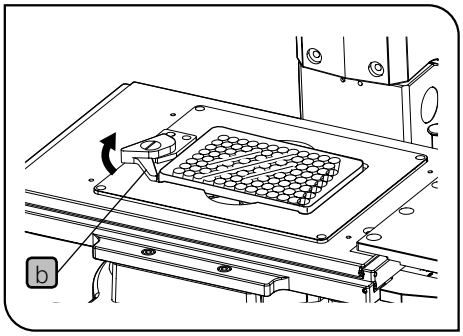
◎ If the specimen is prone to slide on the stage, attach the stage clips (IX-SCL) a and clamp the specimen down with the clips.

With the mechanical stage with right handle IX3-SVR

For IX3-SVR, in addition to the holder for the round stage center plate, following sample holders corresponding to each sample can be attached.

- IX3-HOW : Microplate holder
- IX3-HOS : Slide holder
- IX3-HO35D : Dish holder

CAUTION **The sample holder fixes the specimen to reproduce the specimen position. Do not push up the specimen by the objective lens. The specimen may be popped out.**



IX3-HOW

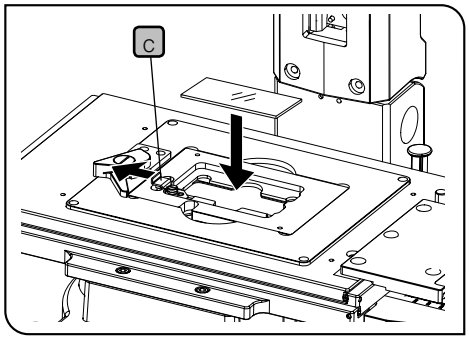
Open the specimen holder b of IX3-HOW, set the microplate in the center, push it toward the right diagonal direction, and return the specimen holder back to the original position.

«Mountable Microplate»

Microplate compliant with SLAS (ANSI/SBS Microplate Standards issued on Jan. 9, 2004.)

Size: 127.76 (plus or minus 0.5) x 85.48 (plus or minus 0.5) mm

Specimen holder: IX3-HOS, IX3-HO35D

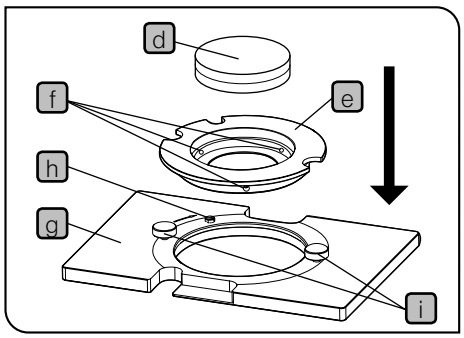


IX3-HOS

Open the specimen fixing part c of IX3-HOS outward, set the specimen in the center, push it toward the right diagonal direction, and return the specimen fixing part back to the original position.

«Chamber Slide Recommended»

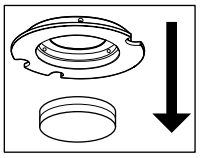
- IWAKI Chamber Slide II (76 x 26 x 0.8 to 1.0 mm)
- Nunc Lab-Tek II Chamber Slide system (25 x 75 x 1.2 mm)
- BD Falcon CultureSlide (25 x 75 x 1.2 mm)



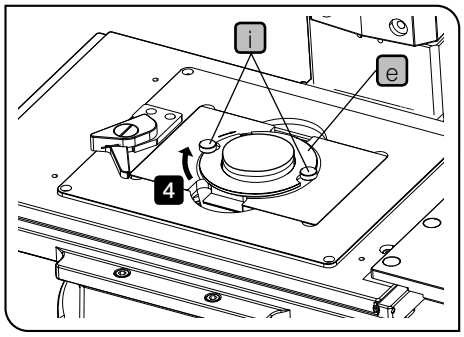
IX3-HO35D

1 Place the 35 mm dish [d] on the 35 mm dish fixing holder [e]. Tighten the fixing screws [f] (3 screws) placed on the side with the Allen screwdriver provided with IX3-HO35D to secure the 35 mm dish.

◎ The 35 mm dish can be secured easily by tightening the fixing screws after flipping over the 35 mm dish in advance.



CAUTION **Do not tighten the fixing screws too firmly. The dish may be damaged.**



2 Set the fixing holder [e] in the center of IX3-HO35D [g] so that the cut-out meets the holder fixing knob [i].

3 Loosen the holder fixing knobs [i].

4 Rotate the fixing holder [e] clockwise to push it to the rotation stopper [h].

5 Tighten the holder fixing knobs [i].

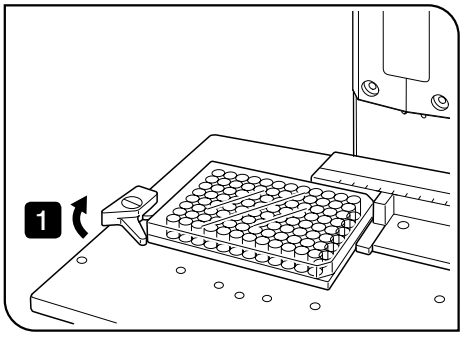
◎ The 35 mm dish fixing holder [e] can be sterilized by using the autoclave.

«35 mm Glass Bottom Dish Recommended»

- Matsunami Glass D111310
- MatTek P35GC-1.5-14-C

«35 mm Dish Recommended»

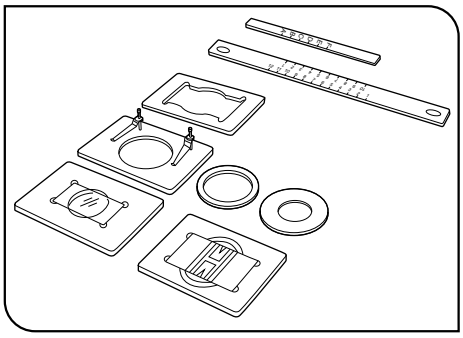
- BD Falcon 351008



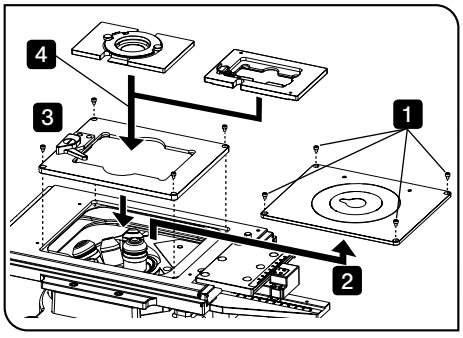
With the mechanical stage IX-MVR + plain stage IX2-SP

◎ 96-well or 24-well microplates, etc. are held in place by the specimen holder.
Microplates with dimensions of max. 136 mm x 92 mm can be accommodated in this way.

1 Open the spring-loaded finger of the specimen holder 1 and slide the microplate into the holder frame. Gently release the curved finger to clamp.



◎ To secure other vessels than microplates, various optional holders are available. A Terasaki plate holder is available for holding Terasaki plates (72-well, 60-well). When using this, it is necessary to replace the stage scales with those provided with the plate holder. Dish holders are available for 35 mm, 54 mm and 65 mm diameter dishes, a slide glass holder is available for holding slide glass, and the IX2-BCTP* is available for a blood cell test plate holder.
* A blood cell test plate or other calculating chamber for bacteria and eosinophil with mounting section dimensions corresponding to H 77 x V 35 x D 2 mm can be used. A 60 mm diameter dish can also be used.



Using the sample holder IX3-SVR

1 Remove the holder fixing screws (4 screws) by using the Allen wrench provided with IX3-SVR.

2 Remove the round stage center plate holder.

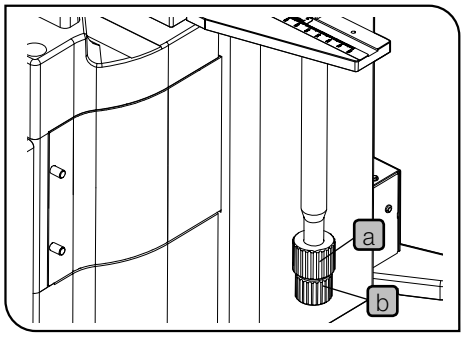
3 Set IX3-HOW in the center of the stage, and attach the holder fixing screws.

4 If you are using IX3-HOS or IX3-HO35D, set the sample holder in the center of IX3-HOW.

2 Moving the Specimen

CAUTION

- **Do not attempt to rotate the stage handle forcedly exceeding the stage movable range. The stage may be damaged.**
- **As the objective may interfere with the stage depending on the focus position, be sure to operate carefully.**



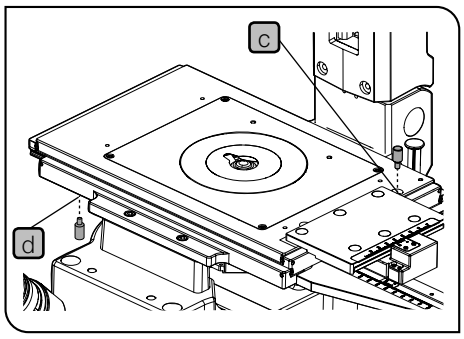
Stage (BX3-SSU)

The stage can be operated by the touch panel controller or the XY-Controller. As to the operation by the touch panel controller and the controller, refer to the Help of the touch panel controller and the controller (cellSens).

Mechanical stage with right handle (IX3-SVR)

To move the specimen to a desired position, rotate the X-axis knob a and Y-axis knob b.

◎ If the stage is used for a long time period, the stage movement range may be narrower rarely. In this case, move the stage several times within the full movement range toward the front/back or right/left direction while holding the top surface of the stage with both hands.



Movement control knob

Attaching the movement control knob provided with IX3-SVR makes it difficult to move the stage in the blocked direction. Even though you may touch the stage accidentally during observation, the observation position can be secured.

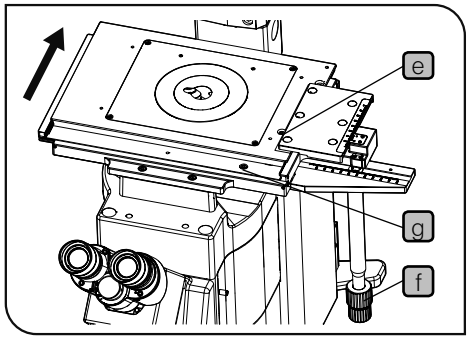
If the movement control knob is attached to the hole of c, the Y-axis movement will be blocked. If it is attached to the hole of d (backside of the stage), the X-axis movement will be blocked.

CAUTION **Do not tighten the movement control knob too firmly. The stage may be damaged.**

◎ If you tighten the movement control knob while focusing on the specimen, it will be defocused.

Movement range limit screw

As a factory default, IX3-SVR, is equipped with the movement range limit screw which limits the stage movable range in the vertical or horizontal direction.
To enlarge the movable range to observe microplates, etc., remove the movement range limit screw.



1 Remove the movement range limit screw e in the vertical direction with the Allen screwdriver provided with the microscope.

2 Rotate the longitudinal handle f to move the stage inward.

3 Remove the movement range limit screw g in the horizontal direction with the Allen screwdriver provided with the microscope.

Stage movable range

With movement range limit screw : 50 mm in vertical direction, 50 mm in horizontal direction
Without movement range limit screw: 75 mm in vertical direction, 114 mm in horizontal direction

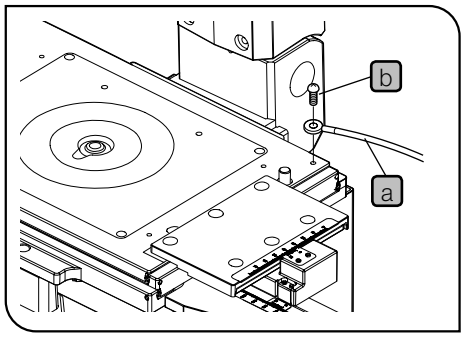
CAUTION **When using the center plate provided with IX3-SVR, attach the movement range limit screw.**

If the movement range limit screw is not attached, the objective may hit the stage.

With the Mechanical Stage IX-MVR

To move the specimen to a desired position, rotate the X-axis knob and Y-axis knob in the same manner as IX3-SVR.

◎ The stage travel area is 130 mm (X-axis) x 85 mm (Y-axis).



3 Connecting the Grounding Wire

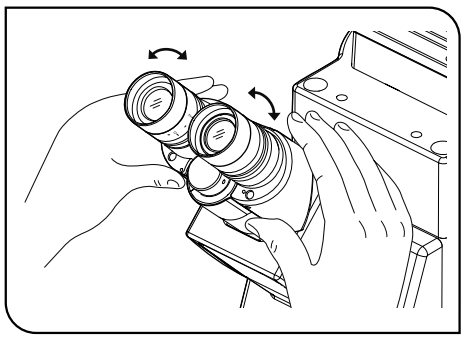
With the Stage BX3-SSU or IX3-SVR

◎ A grounding wire can be attached to the stage for electrophysiological experiments, etc.

Prepare a grounding wire a and one M4 screw b and attach the grounding wire to a screw hole on the stage surface.

CAUTION **The screw hole may sometimes be stuck by paint, etc. In such a case, screw in the M4 screw a few times to expose the metallic thread inside the screw hole and improve the contact before attaching the grounding wire firmly.**

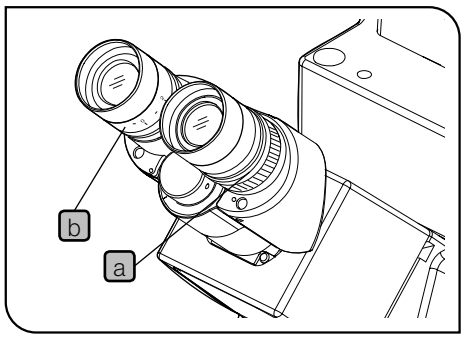
2-3 Observation Tube



1 Adjusting the Interpupillary Distance

While looking through the eyepieces, adjust the binocular vision until the left and right fields of view coincide completely. The index dot • indicates the interpupillary distance.

◎ Note your interpupillary distance so that it can be quickly duplicated



2 Adjusting the Diopter

◎ The diopter adjustment makes it possible to reduce the specimen focusing error even after the objective is switched. As the diopter varies between individuals, the diopter adjustment is required for each person.

CAUTION **The eyepiece with diopter adjustment ring should always be inserted into the observation tube without the diopter adjustment ring.**

1. Set the diopter adjustment rings on both sides to scale "0".
2. Engage a high-power objective (e.g. 40X) in the light path, look into the right eyepiece with your right eye, and bring the sample into focus using the FOCUS button/slider of the touch panel controller.

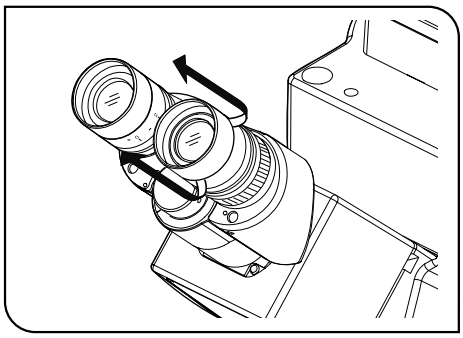
◎ Do not use an immersion objective.

3. Engage a low-power objective (e.g. 10X) in the light path, rotate only the right diopter adjustment ring [a] to bring the sample into focus. At this time do not touch the FOCUS button/slider.
4. Looking into the left eyepiece with your left eye, rotate only the left diopter adjustment ring [b] to bring the sample into focus.

◎ The above procedure adjusts the diopter with reference to the right eye, but it is also possible to adjust with reference to the left eye. In this case, read the above procedure by inverting "right" and "left."

Using an eyepiece including a micrometer disk

1. Looking through the eyepiece with micrometer disk, turn the diopter adjustment ring [b] so that the micrometer in the field of view is sharply visible.
2. Looking through the eyepiece with micrometer disk, focus on the sample using the FOCUS buttons/slider of the touch panel controller so that both the micrometer and sample are sharply visible.
3. Looking through the other eyepiece, turn only the diopter adjustment ring [a] to focus on the sample.



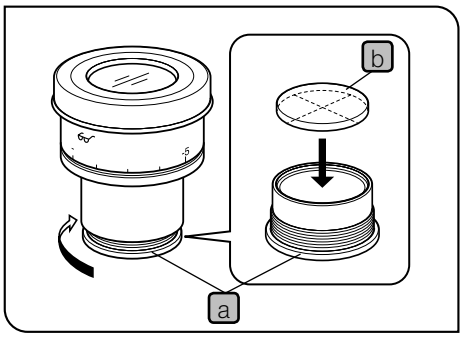
3 Using the Eye Shades

When wearing eyeglasses

Use the eye shades in the normal, folded-down position. This will prevent the eyeglasses from being scratched.

When not wearing eyeglasses

Extend the folded eye shades in the direction of the arrow to prevent extraneous light from entering between the eyepieces and eyes.



4 Mounting the Eyepiece Micrometer Disk

When the WHN10X-H eyepieces are used, an eyepiece micrometer disk can be mounted.
Use 24 mm dia. x 1.5 mm thick micrometer disks.
Turn the built-in micrometer-mounting frame [a] to the arrow direction (see figure) to remove it from the eyepiece and place a micrometer disk [b] into the mounting frame so that the surface with the model indication faces downward.

◎ The micrometer-mounting frame may be to tight for certain micrometer disks.
In this case, turn the frame by holding the circumference with a light, uniform force or by applying the frame against a rubber sheet. Do not grasp the frame with a strong force, as this may deform the frame and make it harder to remove it.
Re-attach the micrometer mounting frame in the original position.

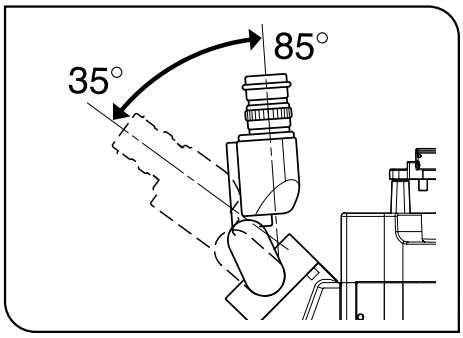
◎ Be careful not to touch the lens or micrometer surface with your finger.

5 Selecting the Light Path of the Trinocular Tube

Slide the light path selector knob to select the desired light path.

(ex.) U-TR30-2, U-TR30H-2

Light path selector knob position		
Pushed in	Middle position	Pulled out
Observation 100%	Observation 20% Camera 80%	Camera 100%



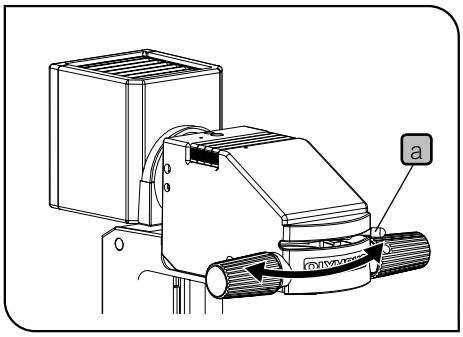
6 Adjusting the Tilt (U-TBI90)

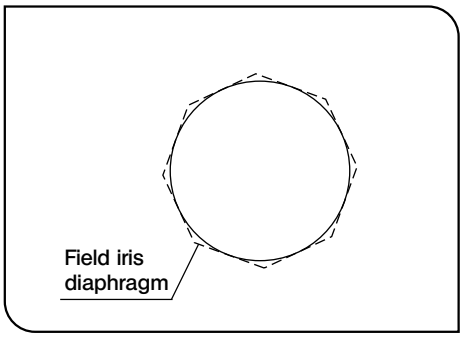
◎ Adjust the height and tilt of the eyepieces to obtain the most comfortable viewing position.
Holding the binocular section with both hands, adjust it to the desired position.

CAUTION **Never attempt to force the binocular section past the upper or lower stop position. Applying excessive force could destroy the limiting mechanism.**

◎ If the U-TBI90 is used together with any type of Intermediate tubes attachment, the image may be cut off or obscured.

2-4 Illumination Column (IX3-ILL)





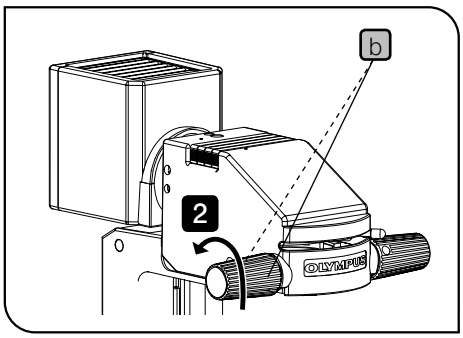
1 Using the Field Iris Diaphragm

◎ The field iris diaphragm lever is used to adjust the diameter of the illumination beam in accordance with the objective in use. Adjust the diaphragm so that the field of view is circumscribed by the field iris diaphragm to cut extra light and improve the contrast of images.

1 Move the field iris diaphragm lever [a] to the left or right to close or open the diaphragm.

○ : Direction for opening the diaphragm

◎ : Direction for closing the diaphragm

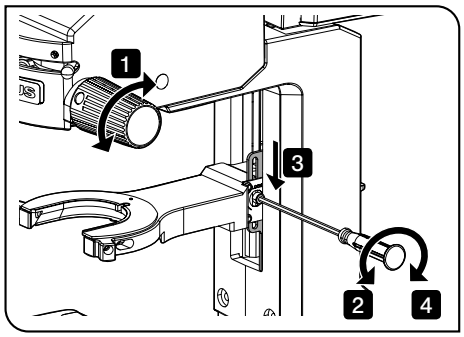


2 Adjusting the Condenser Height Adjustment Knob Tension

1 Loosen the two knob clamping screws [b] on the left adjustment knob using the Allen screwdriver.

2 While holding the right adjustment knob not to rotate it, turning the left adjustment knob counterclockwise (in the direction of the arrow) decreases the rotation tension and clockwise increases it.
Rotating the right adjustment knob allows adjusting the tension of the condenser height adjustment knob while checking it.

3 After adjustment, tighten the two knob clamping screws [b] securely.



3 Condenser refocusing stopper

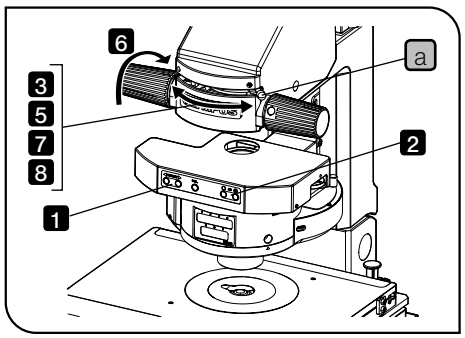
A mechanism returns the condenser back to the original position easily after moving the condenser.

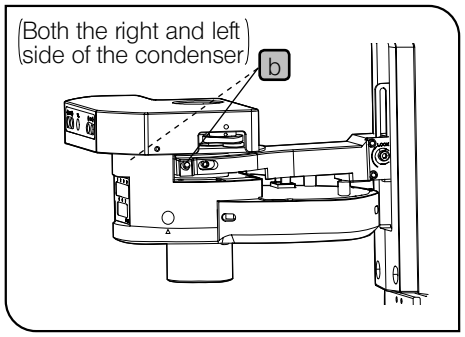
1. Bring the field diaphragm image into focus by rotating the condenser height adjustment knob.
2. Loosen the clamping screws of the stopper using the Allen screwdriver provided with the microscope.
3. Push the top of the stopper downward so that the stopper contacts the column securely.
4. Tighten the clamping screws of the stopper using the Allen screwdriver provided with the microscope.

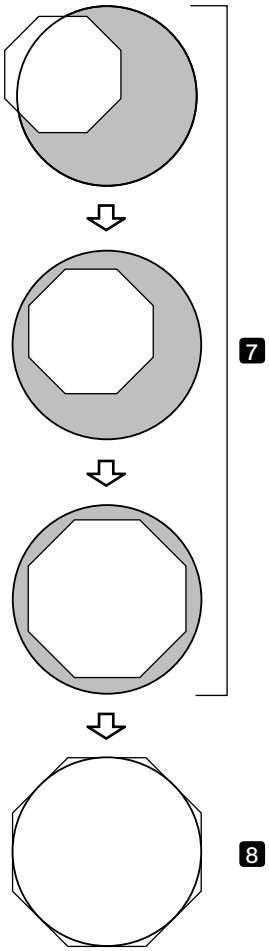
CAUTION **Rotating the condenser height adjustment knob beyond the condenser height adjustment area with an excessive force could damage the microscope. Pay careful attention when rotating it.**

◎ If the manipulator is assembled to the column, the Allen screwdriver provided with the microscope may not be used in some cases.

2-5 Condenser







1 Centering the Condenser

With the Condenser (IX3-LWUCDA, IX2-LWUCD, IX-ULWCD or IX2-MLWCD)

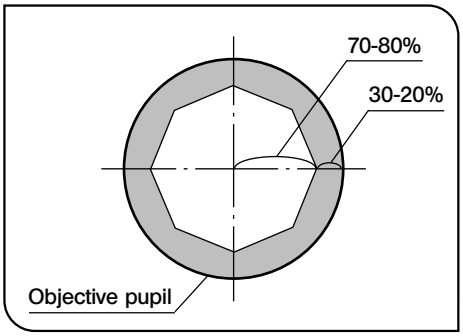
1. Rotate the turret (either manually or electrically) to select the "BF" brightfield observation (with which no optical element is engaged in the light path).
2. Move the aperture iris diaphragm lever to open the diaphragm.
3. Move the field iris diaphragm lever [a] to the fully open position (◎ → ○).
4. Engage the 10X objective and bring the specimen into focus.
5. Using the field iris diaphragm lever [a], stop down the field iris diaphragm until its image is just inside the field of view.
6. Rotate the condenser height adjustment knob to bring the field iris diaphragm image into focus.
7. While gradually opening the field iris diaphragm lever [a], install the Allen screwdriver provided with the microscope in the adjustment hole [b] and rotate it so that the field iris diaphragm image is centered in the field of view of the eyepieces.
8. To check centration, open the field iris diaphragm lever [a] until its image inscribes the field of view. Now the condenser is centered.

   In actual observation, open the field iris diaphragm until its image circumscribes the field of view.

Effect of Field Iris Diaphragm

This is the iris diaphragm to adjust the area to be illuminated. By narrowing down to the level circumscribing the field of view depending on objectives, the excess light can be shielded to acquire the image with good contrast.

◎ Depending on the condenser to be used, the field iris diaphragm may not be viewed by the objective of 40X or higher magnification.

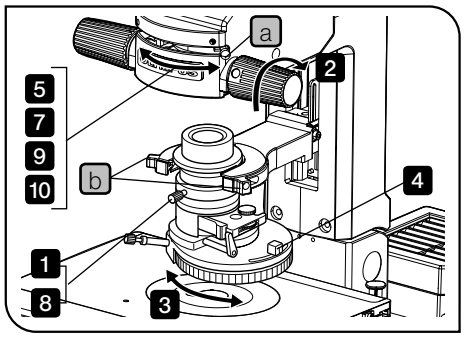


2 Using the Aperture Iris Diaphragm

◎ In general, the potential resolving power of an objective is fully utilized if the diaphragm is stopped down to correspond with the numerical aperture (N.A.) of the objective.

◎ Depending on the specimen, image contrast or focal depth in observation or acquisition may be improved by keeping the aperture iris diaphragm stopped down a little. In general, a good image is obtained if the diaphragm is stopped down to between 70% and 80% of the N.A. of the objective. Stop further down for less contrasty specimens.

◎ To check the position of the perimeter of the aperture iris diaphragm, remove the eyepieces and look into the eyepiece sleeves to view the aperture iris diaphragm image and the objective's exit pupil.



With the U-UCD8 Condenser

1. Rotate the condenser height fine adjustment knob counterclockwise to loosen it, then push the knob all the way toward the rear.
2. Rotate the condenser height adjustment knob in the direction of the arrow to lower the condenser to the level not to hit the specimen.
3. Rotate the turret to select the “BF” brightfield observation (with which no optical element is engaged in the light path).
4. Move the aperture iris diaphragm lever to open the diaphragm.
5. Move the field iris diaphragm lever [a] to the fully open position (◎ → ○).
6. Engage the 10X objective and bring the specimen into focus.
7. While gradually opening the field iris diaphragm lever [a], install the Allen screwdriver provided with the microscope in the adjustment hole and rotate it so that the field iris diaphragm image is centered in the field of view of the eyepieces.
8. Slowly move the condenser height fine adjustment knob toward the front to bring the field iris diaphragm image into focus. When focusing is obtained, rotate the knob clockwise to clamp it.
9. While gradually opening the field iris diaphragm lever [a], install the Allen screwdriver provided with the microscope in the adjustment hole [b] and rotate it so that the field iris diaphragm image is centered in the field of view of the eyepieces.
10. To check centration, open the field iris diaphragm lever [a] until its image inscribes the field of view. Now the condenser is centered.

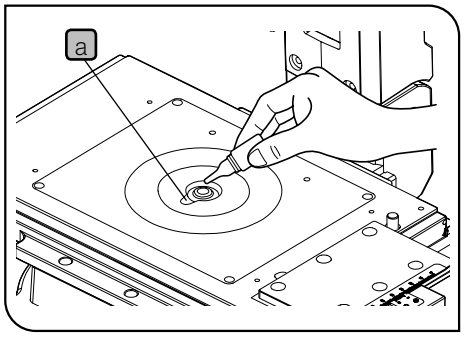
◎ In actual observation, open the field iris diaphragm until its image circumscribes the field of view.

◎ When replacing the specimen or spreading immersion oil, use the condenser height adjustment knob to raise the condenser first. After finishing the procedure, lower the condenser to its lowest position.

2-6 Oil- or Water-Immersion Objective

1 Using Oil- or Water-Immersion Objective

◎ If you use an oil-immersion objective, use immersion oil as described below.



CAUTION **Always use immersion oil supplied by Olympus.**

1 Using a low-power objective, bring the specimen into focus.

2 Rotate the revolving nosepiece to engage the oil immersion objective.

3 Remove the specimen and move the stage insert cut-out a close to the objective front lens. Apply a drop of the immersion oil to the objective front lens. Place the specimen and rotate the fine adjustment knob to bring the specimen into focus.

CAUTION
- **Use as little oil as possible.**
- **If the oil contains air bubbles, the image will be degraded. Make sure the oil is free of air bubbles.**

4 After use, remove immersion oil from the objective front lens by wiping with gauze slightly moistened with absolute alcohol.

◎ The presence of air bubbles can be checked by viewing the pupil of the objective (viewed as a bright circular shape) in the tube after removing the eyepiece and opening the field iris diaphragm and the aperture iris diaphragm completely.

CAUTION **Caution in use of immersion oil:**

If immersion oil enters your eyes or contacts your skin, immediately take the following treatment.

Eyes: Rinse with fresh water (for 15 minutes or more).
Skin : Rinse with water and soap.

If the appearance of the eyes or skin is altered or pain persists, immediately see your doctor.

3 OTHER OBSERVATION METHODS

3-1 Reflected Fluorescence Observation

1 REFLECTED FLUORESCENCE OBSERVATION PROCEDURE

◎ If you need simultaneous observations of reflected fluorescence observation with the phase contrast observation or transmitted light Differential Interference Contrast (DIC) observation, please read Chapter 5, "SIMULTANEOUS FLUORESCENCE OBSERVATION" (page 44).

Preparation (Page)

- Attach the fluorescence mirror unit and objective matching the observation method.
- Set the main switch of related control boxes to " I " (ON).

Set the main switch of the power supply unit to " I " (ON) and wait for the lamp brightness to stabilize (5 to 10 minutes after ignition). ---------- (P.12)

↓

Place the specimen on the stage.

↓

Start up the FV10-ASW software.

↓

Click the ‹EPI› button  of the FV10-ASW software.

↓

Engage the fluorescence mirror unit matching the specimen in the light path. ---------- (P.36)

↓

Engage the objective in the light path, open the shutter and focus on the specimen. ---------- (P.35)

↓

Start observation.

◎ Engage the shutter if you take a short break during the observation. ---------- (P.35)

2 General Precautions for Observation

1. Make sure that the power cord and connecting cables are plugged in securely.
2. If you perform only transmitted phase contrast or transmitted DIC observations*, leave one position on the turret empty.
   This allows for transmission of white light and reproduction of original colors.
   * This care is not required when the transmitted DIC mirror unit IX3-FDICT with built-in analyzer is used.
3. Always use Olympus immersion oil for oil immersion objectives.
4. If you use an objective with correction collar, you can correct contrast degradation due to variation in cover glass thickness by adjusting the correction collar.

Correction procedure

If the cover glass thickness is known, match the correction collar to the cover glass thickness using the collar scale provided. If the thickness is not known, turn the collection collar and adjust the fine adjustment knob to where the image contrast is best.

5. Engage the shutter if you take a short break during the observation.
6. Photobleaching of specimens
   This system features high excitation light intensity to ensure bright observation of dark fluorescence specimens. In consequence, after long period of observations using high-power objectives, the fluorescence of specimens will bleach quicker than usual, causing the view (contrast) of fluorescent images to deteriorate.
   In such a case, slightly reduce the excitation light intensity to slow photobleaching down and to improve the fluorescence images.
   To reduce the excitation light intensity, use ND filters or aperture iris diaphragm as far as the observation is not affected or use the shutter to limit the exposure of specimen to more than necessary light.
   Commercially-marketed photobleaching protection agent (DABCO, etc.) can also delay photobleaching of specimen. The use of photobleaching protection agent is recommended especially when you perform high-magnification observations frequently.
   ◎ Remember that the photobleaching protection agents cannot be used with certain kinds of specimens.

CAUTION **When the excitation filter is removed from the fluorescence mirror unit and attached on the excitation filter slider on the side of the white lamp, very glaring light may enter the eyepieces.**

CAUTION **The mirror units provided at the No. 1 and No. 2 positions in the mirror unit cassette are used for the LSM observation. Do not use them for the other purposes than the LSM observation. Also, do not remove them from the cassette.**

3 Selecting the Fluorescence Mirror Unit

Select the fluorescence mirror unit which matches the fluorochrome in use.

◎ Usage according to the excitation light bandwidth:
Depending on the bandwidth of excitation wavelength, several combinations of the excitation filters and barrier filters are available. The wide-band (W) set is normally used. However, when the fluorescence emitted from substances other than the fluorescent stain is strong, the narrow-band (N) set is recommendable (though the fluorescence becomes slightly darker).

Dichroic mirror and filter configurations of fluorescence mirror units

Excitation method	Mirror unit	Excitation filter	Barrier filter	Dichroic mirror	Applications
U	U-FUW IX3-FUWXL	BP340-390	BA420IF	DM410	• Autofluorescence observation • DAPI: DNA staining • Hoechst 33258, 33342: Chromosome
	U-FUN	BP360-370			
V	U-FVN	BP400-410	BA460IF	DM455	• Catecholamine observation • Serotonin observation • Tetracyline: Bones, teeth
BV	U-FBVW IX3-FBVWXL	BP400-440	BA460IF	DM455	• Quinacrine, quinacrine mustard: Chromosome • Thioflavine S: Lymphocyte • Acriflavine: Nucleic acid • ECFP
B	U-FBW	BP460-495	BA510IF	DM505	• FITC: Fluorescent antibody • Acridine orange: DNA, RNA • Auramine: Tubercle bacillus • EGFP, S65T, RSGFP
	U-FBN	BP470-495			
G	U-FGW IX3-FGWXL	BP530-550	BA575IF	DM570	• Rhodamine, TRITC: Fluorescent antibody • Propidium iodide: DNA • RFP
Y	U-FYW	BP540-585	BA600IF	DM595	Texas Red: Fluorescent antibody

Combinations for color separation

U	U-FUNA	BP360-370	BA420-460	DM410	For observing only the U-excitation stain when using U-excitation stain together with FITC.
B	U-FBWA	BP460-495	BA510-550	DM505	For observing only the B-excitation stain when using B-excitation stain with TRITC or Texas Red.
	U-FBNA	BP470-495			
G	U-FGWA	BP530-550	BA575-625	DM570	For observing only the G-excitation stain when using G-excitation stain together with Cy5.
	U-FGNA	BP540-550			

Exclusive combinations for fluorescent proteins

CFP	U-FCFP IX3-FCFPXL	BP425-445CFP	BA460-510CFP	DM455CFP	For ECFP.
GFP	U-FGFP IX3-FGFPXL	BP460-480GFP	BA495-540GFP	DM490GFP	For EGFP
YFP	U-FYFP IX3-FYFPXL	BP490-500YFP	BP515-560YFP	DM515YFP	For EYFP
RFP	U-FRFP IX3-FRFPXL	BP535-555	BA570-625	DM565	For RFP
mCherry	U-FMCHE IX3-FMCHEXL	BP565-585	BA600-690	DM595	For mCherry

4 Objectives for Various Observation Methods

Objective		Reflected fluorescence						Phase contrast (Phase contrast ring)	DIC		
									U-UCD8		IX3-LWUCDA
		340	U	V	BV	B	G		U-TLO	U-TLD	IX2-LWUCD
UPLSAPO	4X	△	○	○	○	○	○	—	—	—	—
	10X2	△	○	○	○	○	○	—	—	○	○
	20X	△	○	○	○	○	○	—	○	○	○
	20XO	△	○	○	○	○	○	—	○	○	○
	30XS	△	○	○	○	○	○	—	—	○	○
	40X2	△	○	○	○	○	○	—	○	○	○
	60XO	△	○	○	○	○	○	—	○	○	○
	60XW	△	○	○	○	○	○	—	○	○	○
	60XS	△	○	○	○	○	○	—	○	○	○
	100XO	△	○	○	○	○	○	—	○	○	○
UPLFLN	4X	△	○	○	○	○	○	○*(PhL)	—	—	—
	10X2	△	○	○	○	○	○	○*(Ph1)	—	○	○
	20X	△	○	○	○	○	○	○*(Ph1)	—	○	○
	40X	△	○	○	○	○	○	○*(Ph2)	○	○	○
	40XO	△	○	○	○	○	○	—	○	○	○
	60X	△	○	○	○	○	○	—	○	○	○
	60XOI	△	○	○	○	○	○	○*(Ph3)	○	○	○
	100XO2	△	○	○	○	○	○	○*(Ph3)	○	○	○
	100XOI2	△	○	○	○	○	○	—	○	○	○
PLAPON	60XO	—	△	○	○	○	○	—	○	○	○
	60OSC	—	△	○	○	○	○	—	○	○	○
CPLFLN	10XPH	△	△	△	△	△	△	○*(PhC)	—	—	○
LCACHN	20X	—	△	△	△	○	○	○*(PhC)	—	—	—
	40X	—	—	—	—	○	○	○*(Ph2)	—	—	—
LUCPLFLN	20X	○	○	○	○	○	○	○*(Ph1)	○	○	○
	40X	○	○	○	○	○	○	○*(Ph2)	○	○	○
	60X	△	○	○	○	○	○	○*(Ph2)	○	○	○
UAPON	20XW340	○	○	○	○	○	○	—	○	○	○
	40XW340	○	○	○	○	○	○	—	○	○	○
	40XO340	○	○	○	○	○	○	—	○	○	○

○ : Recommended combination.

○* : A phase contrast objective (Ph) is required for phase contrast observation.

△ : Usable but image may be dark.

– : Not available.

5 Turning the Lamp On

Set the main switch to " I " (ON). The illumination light will stop flickering and stabilize in 5 to 10 minutes after ignition.

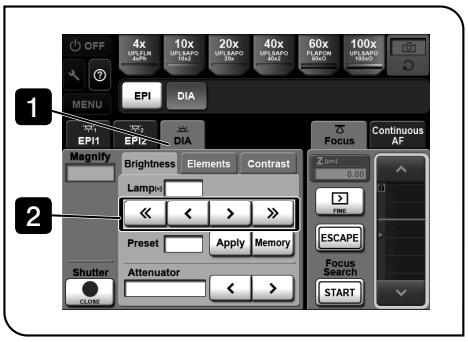
CAUTION

- **To extend the mercury burner life, do not turn it on and off at short interval. If you want to take a short break during the observation for a while in less than 2 hours after ignition, do not switch the power supply unit to off but simply close the shutter of the illuminator during a break.**
- **The mercury burner cannot be reignited until the mercury vapor has cooled down and liquefied. Before reigniting a mercury burner, wait for about 10 minutes after the last time it was turned off.**

◎ For details, see the instruction manual provided with the power supply unit.

◎ The discharge type mercury burner may not be ignited from the beginning on rare occasions due to its characteristics. In this case, set the main switch to " O " (OFF), wait for 5 to 10 seconds, then set it again to " I " (ON).

◎ After replacing the mercury burner, reset the hour counter by holding its reset button till "0.0" is displayed.



Brightness control by using touch panel controller

1 Tap the DIA .
DIA: Transmitted light observation

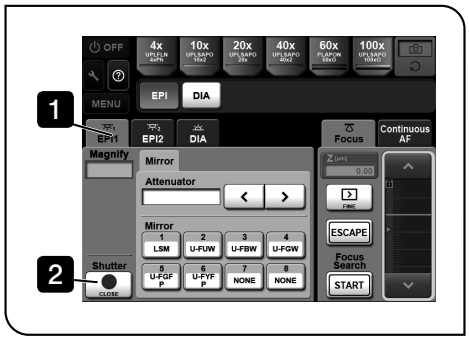
2 Tap the brightness adjustment buttons.

6 Opening/closing the shutter

When using the IX3-RFACA with IX83

The shutter can be opened and closed by the FL SHUTTER button of the touch panel controller and the U-MCZ. The open/close switching condition of the shutter can be checked by the touch panel controller

◎ For details, see the Help provided with touch panel controller.



When using the touch panel controller

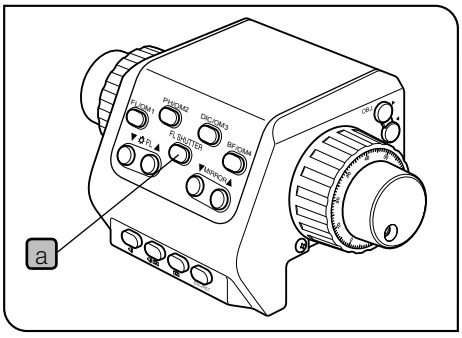
1 Tap the EPI1 .

FL1: Observation by using 1st deck

2 Tap the Shutter .

Blue: The shutter is closed.

White: The shutter is opened.



When the U-MCZ is in use

Press the FL SHUTTER button a of the U-MCZ.

◎ The FL SHUTTER button a opens/closes the reflected shutter only. The transmitted shutter opens/closes automatically depending on the observation modes.

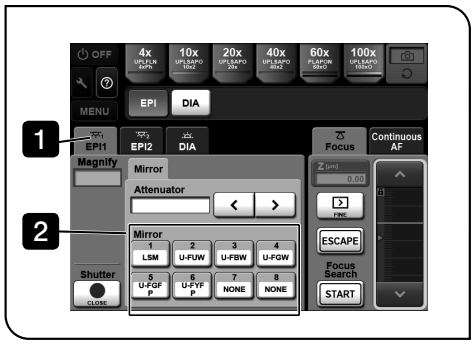
7 Switching over the mirror units in the mirror turret

When using the IX3-RFACA with IX83

The mirror units can be switched over and engaged in the light path by the touch panel controller and the U-MCZ.

◎ The display panel shows the turret number engaged in the light path of either 1st deck. You can select which turret number of the deck is displayed by the touch panel controller. The factory default is set to 1st deck.

◎ For details, see the instruction manual provided with the IX3-CBH, or Help provided with touch panel controller.

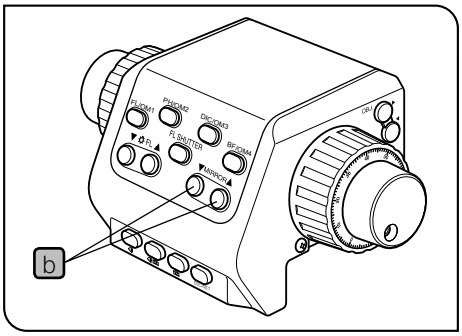


When using the touch panel controller

1 Tap the EPI1.

FL1: Observation by using 1st deck

2 Tap the number of turret to be used.



When using the U-MCZ

Press the MIRROR button b of the U-MCZ.

◎ U-MCZ can switch over the turret of either 1st deck.

3-2 Phase Contrast Observation

◎ A phase contrast objective, phase contrast optical element, and the U-CT30-2 centering telescope are required for phase contrast observation.

◎ If a DIC slider, analyzer or polarizer is engaged in the light path, disengage it.

1 Phase Contrast Optical Elements and Applicable Objectives

With the IX3-LWUCDA or IX2-LWUCD

◎ Insert the optical element (small) in the 30 mm position and the optical element (large) in the 38 mm position. When observing the specimens in wells, it is recommended to use the IX-PHC to obtain the phase contrast effect in a wide range of field of view.

Optical Element	Indication	Applicable Objectives
IX-PHL (small)	PhL	UPLFLN4XPH
IX-PHC (small)	PhC	CPLN10XPH, LCACHN20XPH, CPLFLN10XPH
IX-PH1 (small)	Ph1	UPLFLN10X2PH, UPLFLN20XPH, LUCPLFLN20XPH
IX-PH2 (small)	Ph2	UCPLFLN20XPH, UPLFLN40XPH, LUCPLFLN40XPH, LUCPLFLN60XPH, LCACHN40XPH
IX-PH3 (large)	Ph3	PLAPON60XOPH, UPLFLN60XOIPH, UPLSAPO100XOPH, UPLFLN100XO2PH

Example of attaching optical elements (IX3-LWUCDA)

Small Diameter: PH1, PHL, free or PH2, C, free

Large Diameter: PH3, DIC40, DIC60, DIC100

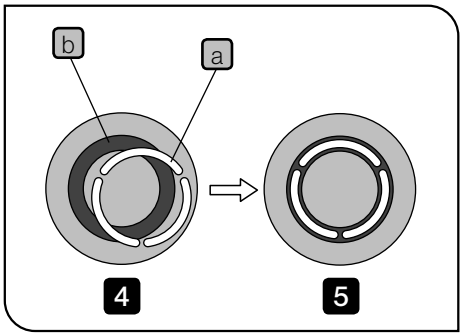
With the IX-ULWCD

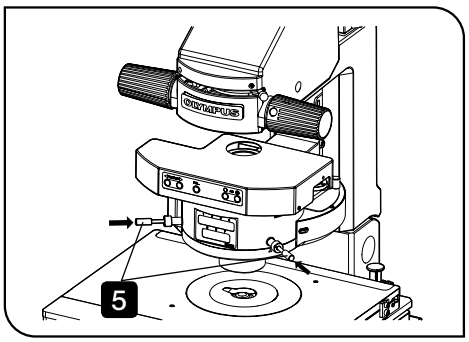
◎ The IX-PHCU or IX-PH1U can be attached only in the Ph1 and PhC. (Do not remove the built-in elements.)

Optical Element	Indication	Applicable Objectives
PHL (built-in)	PhL	UPLFLN4XPH
IX-PHCU	PhC	CPLN10XPH, LCACHN20XPH, CPLFLN10XPH
IX-PH1U	Ph1	UPLFLN10XPH, UPLFLN20XPH, LUCPLFLN20XPH,
PH2 (built-in)	Ph2	UCPLFLN20XPH, UPLFLN40XPH, LUCPLFLN40XPH, LUCPLFLN60XPH, LCACHN40XPH

◎ When using the U-UCD8 or IX2-MLWCD, refer to the provided instructions.

2 Centering the Phase Contrast Ring Slit





◎ Open the aperture iris diaphragm during phase contrast observation.

1 Engage the phase contrast objective in the light path and bring the specimen into focus.

2 Remove an eyepiece and attach the U-CT30-2 centering telescope in place.

3 Engage the ring slit of the condenser matching the phase contrast objective in the light path.

4 Rotate the knurled section of the centering telescope to focus on the ring slit [a] and the phase plate [b] of the objective.

5 Pushing the optical element centering knobs, turn the phase contrast ring slit centering screws (in positions marked) so that the ring slit image overlaps with the phase plate of the objective.

◎ Do not release the hand suddenly while the optical element centering knobs are being pushed in. The optical element centering knobs may be popped out.

6 Remove the U-CT30-2 centering telescope and attach an eyepiece in place.

◎ If the vessel is not completely flat, it may become necessary to adjust the centering again to obtain the optimum contrast. Adjust the centering in each objective power.

7 Adjust the field iris diaphragm so that its image circumscribes the field of view and observe the phase contrast.

◎ Engaging the green filter in the light path will improve the contrast.

◎ If you are using IX83P2ZF, a thin shading may occur in the periphery of the field of view during visual observation.

3-3 Differential Interference Contrast Observation

◎ If a plastic dish is used, the normal optical performance of DIC observation cannot be manifested due to the polarization characteristic of the dish. Use a glass dish.

◎ DIC optical elements, a DIC slider, analyzer, and polarizer are required for DIC observation.

1 DIC Optical Elements, Applicable Objectives and DIC Sliders

With the IX3-LWUCDA or IX2-LWUCD

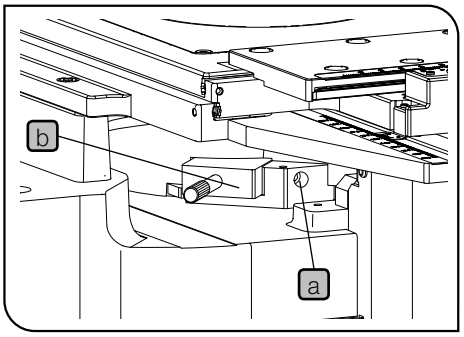
◎ Insert a small optical element (one of the optical elements inside ( ) in the following table) in the 30 mm position and other optical element (large) in the 38 mm position.

UIS2 Series

Applicable Objective \ DIC Slider		U-DICT	U-DICTS Shift Type	U-DICTHC High Contrast Type	U-DICTHR High Resolution Type
UPLSAPO	10X2	(IX2-DIC10)	(IX2-DIC10)	–	–
	20X	(IX2-DIC20)	(IX2-DIC20)	(IX2-DIC20HC)	(IX2-DIC20HR)
	20XO	(IX2-DIC20)	(IX2-DIC20)	(IX2-DIC20HC)	(IX2-DIC20HR)
	30XS	IX2-DIC30	IX2-DIC30	–	–
	40X2	IX2-DIC40	IX2-DIC40	IX2-DIC40HC	IX2-DIC40HR
	60XO	–	IX2-DIC60	–	–
	60XW	IX2-DIC60	IX2-DIC60	–	–
	60XS	IX2-DIC60	IX2-DIC60	–	–
	100XO	IX2-DIC100	IX2-DIC100	–	–
	100XOPH	IX2-DIC100	IX2-DIC100	–	–
PLAPON	60XO	–	IX2-DIC60	–	–
	60XOPH	–	IX2-DIC60	–	–
UPLFLN	10X2	(IX2-DIC10)	(IX2-DIC10)	–	–
	20X	(IX2-DIC20)	(IX2-DIC20)	(IX2-DIC20)	(IX2-DIC20HR)
	40X	IX2-DIC40	IX2-DIC40	IX2-DIC40HC	IX2-DIC40HR
	40XO	–	IX2-DIC40	IX2-DIC40HC	IX2-DIC40HR
	60X	IX2-DIC60	IX2-DIC60	–	–
	60XOI	IX2-DIC60	IX2-DIC60	–	–
	100XO2	IX2-DIC100	IX2-DIC100	–	–
	100XOI2	IX2-DIC100	IX2-DIC100	–	–
LUCPLFLN	20X	(IX2-DIC20)	(IX2-DIC20)	(IX2-DIC20HC)	(IX2-DIC20HR)
	40X	IX2-DIC40	IX2-DIC40	IX2-DIC40HC	IX2-DIC40HR
	60X	IX2-DIC60	IX2-DIC60	–	–
UAPON	20XW340	(IX2-DIC20)	(IX2-DIC20)	(IX2-DIC20HC)	(IX2-DIC20HR)
	40XW340	–	IX2-DIC40	IX2-DIC40HC	IX2-DIC40HR
	40XO340	–	IX2-DIC40	IX2-DIC40HC	IX2-DIC40HR
	100XOTIRF	IX2-DIC100	IX2-DIC100	–	–
	150XOTIRF	IX2-DIC100	IX2-DIC100	–	–
UCPLFLN	20X	(IX2-DIC20)	(IX2-DIC20)	(IX2-DIC20HC)	(IX2-DIC20HR)
	20XPH	(IX2-DIC20)	(IX2-DIC20)	(IX2-DIC20HC)	(IX2-DIC20HR)
APON	60XOTIRF	–	IX2-DIC60	–	–
	100XHOTIRF	–	IX2-DIC100	–	–

◎ When using the U-UCD8, refer to the U-UCD8 instructions.

2 Attaching the Analyzer and DIC Slider



With the DIC Slider U-DICTS/U-DICTHC/U-DICTHR

◎ If you use these sliders, the analyzer U-ANT cannot be used. The transmitted DIC mirror unit IX3-FDICT is required.

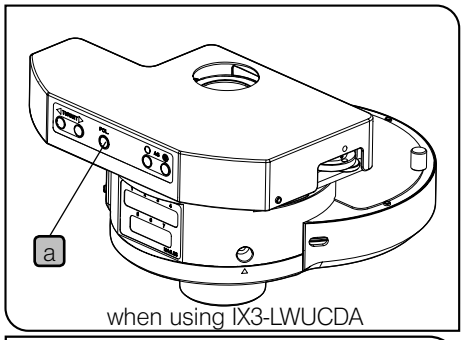
1 Loosen the fixing screw a by using the Allen screwdriver provided with the microscope to remove the dummy slider from the revolving nosepiece.

2 Insert the DIC slider b to be used into the revolving nosepiece by facing its display surface down.

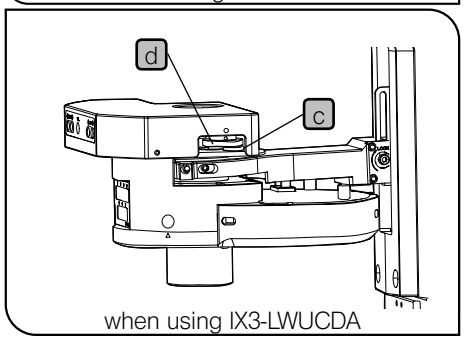
With the Transmitted DIC Mirror Unit IX3-FDICT

IX3-FDICT is the mirror unit equipped with the analyzer. This is attached to the mirror unit cassette and engaged into the light path during the transmitted differential interference observation.

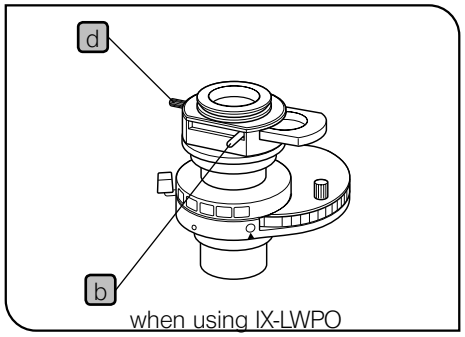
In this case, it is not necessary to attach the analyzer U-ANT to U-DICT.



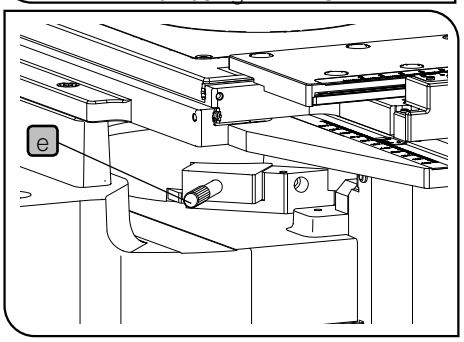
when using IX3-LWUCDA

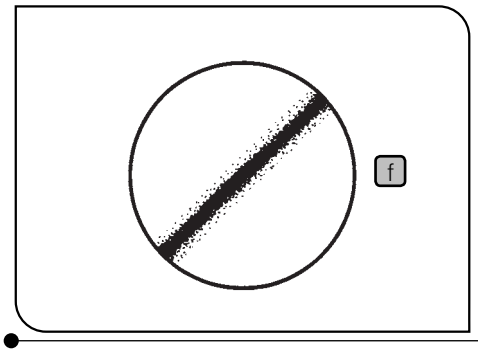


when using IX3-LWUCDA



when using IX-LWPO





3 Cross-Nicol Adjustment

1. Rotate the condenser's turret for the BF (brightfield) light path (with no optical element engaged in the light path).
2. When IX3-LWUCDA is used, push the button [a] of IX3-LWUCDA to engage the polarizer in the light path.
   When IX-LWPO is used, move the polarizer detaching lever [b] on the IX-LWPO polarizer to engage the polarizer in the light path.
3. Engage the 10X objective in the light path, place an optimum specimen for brightfield observation on the stage, bring the specimen into approximate and remove the specimen out of the light path.
4. Refer to page 40 engage the DIC slider and the analyzer in the light path.
5. Remove the eyepieces from the observation tube, look into the observation tube.
6. Move the prism movement knob [e] of the DIC slider in the clockwise direction around the axis until the knob is stopped. First a black interference stripe then a rainbow-colored interference stripe will be observed. Stop the knob at the position in which the black interference stripe can be seen. (State of [f])
7. When IX3-LWUCDA is used, loosen the polarizer rotation knob [d] by rotating the clamping knob [c] in the clockwise direction until the knob stops.

   When IX-LWPO is used, loosen the clamping of the polarizer rotation/clamping knob by rotating slightly the polarizer rotation/clamping knob [d] in a counter-clockwise direction.
8. While looking into the observation tube, rotate the polarizer rotation/clamping knob [d] on the polarizer unit horizontally until the black interference stripe becomes darkest. This is the cross-nicol position.
9. After determining the position, clamp the polarizer.

   When the IX3-LWUCDA is used, rotate the clamping knob [c] in the counterclockwise direction around the axis until the knob is stopped.

   When the IX-LWPO is used, rotate the polarizer rotation/clamping knob [d] in a clockwise direction until the knob stops.

4 Observation Method

1. Rotate the condenser turret to engage the suitable optical element for the objective in use in the light path.
2. Engage the objective to be used in the light path.
3. Place the specimen on the stage and bring the specimen into focus by moving the objective up or down.
4. Adjust the field iris diaphragm so that its image circumscribes the field of view.
5. Adjust the aperture iris diaphragm to enhance the contrast.
6. Engage the DIC slider in the light path.
7. Move the prism movement knob of the DIC slider to select the interference color that can provide the optimum contrast in accordance with the specimen.

U-DICTS :
U-DICTHC :
U-DICTHR :
} The background interference color is continuously variable from black to light gray.

◎ With sensitive color observation using the U-UCD8, engage the UUCDTP530 1 plate (sensitive color plate) in the light path.

- Setting the background color to dark enables an observation like darkfield observation.
- Setting the background color to gray provides observation with high contrast and 3D feeling with the gray sensitive color with which the sensitivity is highest.
- Setting the background color to gray allows very small change in phase to be observed as a change in color.
- There is a directional characteristic with the detection sensitivity because of the configuration of the DIC prism. As a result, the contrast may sometimes be improved by rotating the specimen on the stage.

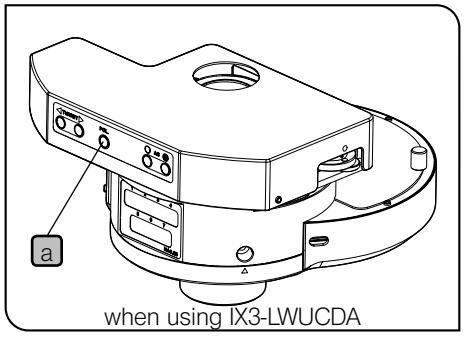
3-4 Simplified Polarized Light Observation

1 Attaching the Analyzer and Polarizer

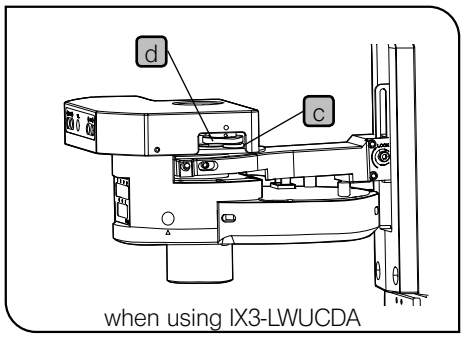
◎ Use the same procedure as that for attaching the analyzer and polarizer for DIC observation.

◎ The simplified polarized light observation is enabled also by attaching the analyzer U-ANT to the dummy slider.

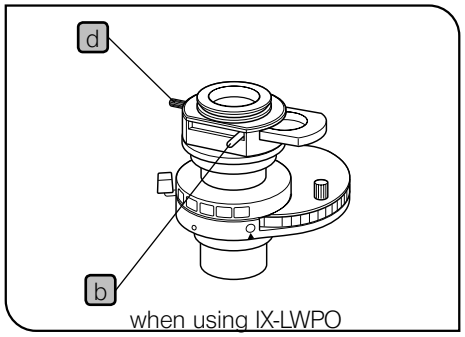
2 Observation Method



when using IX3-LWUCDA



when using IX3-LWUCDA



when using IX-LWPO

1. Rotate the turret for the BF (brightfield) light path (with no optical element engaged in the light path).
2. When IX3-LWUCDA is used, push the button [a] of IX3-LWUCDA to engage the polarizer in the light path.
   When IX-LWPO is used, move the polarizer detaching lever [b] on the IX-LWPO polarizer to engage the polarizer in the light path.
3. Rotate the revolving nosepiece to engage the objective to be used in the light path.
4. Refer to page 40 and engage the analyzer in the light path.
5. When IX3-LWUCDA is used, loosen the polarizer rotation knob [d] by rotating the clamping knob [c] in the clockwise direction until the knob stops.

   When IX-LWPO is used, loosen the clamping of the polarizer rotation/clamping knob by rotating slightly the polarizer rotation/clamping knob [d] in a counter-clockwise direction.
6. Move the polarizer rotation/clamping knob [d] on the polarizer unit horizontally in the counterclockwise direction around the axis until the position in which the field of view is darkest.
7. After determining the position, clamp the polarizer.

   When the IX3-LWUCDA is used, rotate the clamping knob [c] in the counterclockwise direction around the axis until the knob is stopped.

   When the IX-LWPO is used, rotate the polarizer rotation/clamping knob [d] in a clockwise direction until the knob stops.
8. Place a specimen on the stage and bring the specimen into focus. Now the specimen can be observed with simplified polarized light observation.
9. Adjust the field iris diaphragm so that its image circumscribes the field of view.
10. Adjust the aperture iris diaphragm to enhance the contrast.

3-5 Simultaneous fluorescence observations

◎ In case of FV10-ASW opening, Simultaneous fluorescence observations is not available.

◎ By properly combining equipment, this system can be used in transmitted light brightfield observation, transmitted light phase contrast observation and transmitted light DIC (Differential Interference Contrast) observation in addition to the reflected fluorescence observation. With specimens which bleach quickly, photobleaching can be minimized by initially using transmitted light phase contrast or transmitted light DIC observation to determine the area of the specimen to be observed. Reflected fluorescence observation can also be executed simultaneously with phase contrast or DIC observation.

1 Simultaneous reflected fluorescence/phase contrast observations

◎ The phase contrast observation requires the IX3-LWUCDA, IX2-LWUCD, IX-ULWCD, IX2-MLWCD or U-UCD8 condenser and a Ph objective.

1. Rotate the turret of the fluorescence mirror turret and engage the position without fluorescence mirror unit in the light path.
2. Rotate the phase contrast turret to show the same number as the Ph number shown on the objective.
3. Adjust the optical axis between the ring slit and phase plate by centering them.
4. Engage the mirror unit corresponding to the desired excitation in the light path and release the shutter.
5. Adjust the transmitted light for the best balance of fluorescence and phase contrast brightness.

◎ To adjust the transmitted light intensity, use the light intensity control knob of the microscope and ND filters in combination.
Remember that the service life of a halogen bulb tends to decrease when the bulb is kept on continuously at a extremely low voltage, so be careful.

2 Simultaneous reflected fluorescence/DIC (transmitted) observations

◎ The DIC observation requires the following modules.

- Condenser (IX3-LWUCDA, IX2-LWUCD, IX2-MLWCD or U-UCD8)
- DIC slider (U-DICT, U-DICTS, U-DICTHC or U-DICTHR)
- Analyzer (U-ANT, IX3-AN, IX3-FDICT) *U-ANT can be mounted only on the U-DICT.

CAUTION **Do not insert the U-ANT analyzer in the DIC slider, for this will dim the fluorescence observation image and cause the analyzer to be burnt.**

1. Engage the position where the fluorescence mirror unit is not attached in the light path.
2. Adjust the polarizer on the DIC condenser to the “Crossed Nicol” (complete extinction) position.
3. Insert the DIC slider into the position provided on the revolving nosepiece.
4. Rotate the turret on the condenser to select the Nomarski prism matching the objective to be used for observation.
5. Engage the objective to be used in the light path.
6. Place the specimen on the stage and focus on the specimen.
7. Adjust the field iris diaphragm and aperture iris diaphragm.
8. Turn the prism movement knob on the DIC slider to adjust contrast of the DIC image.
9. Engage the mirror unit corresponding to the desired excitation and release the shutter.
10. Adjust the transmitted light for optimum fluorescence and DIC image brightness.

◎ If you use IX3-RFACA, each motorized module can be switched to the simultaneous observation setting status by operating the button on touch panel controller (combined with IX83).
For details, refer to Help of touch panel controller.

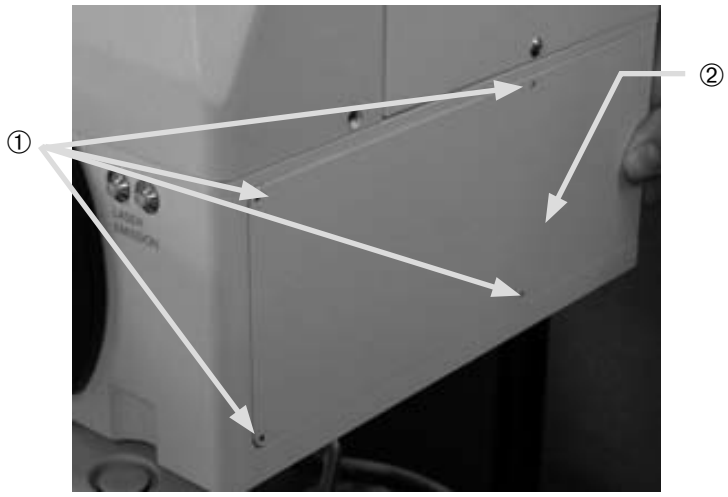
4 Replacement of Cubes

The DM (Dichroic Mirror) on the cubes is used to connect the light path of the optional FV10-ASU Auxiliary Scan Unit or FV12-HSD Non-confocal Point Detector with that of the scan unit. It should be selected according to the observation method.

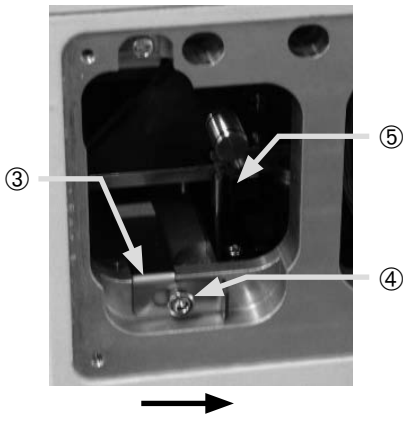
4-1 FV10-ASU

1 Taken out from FV10-ASU

1. Set the light path of the scan unit to the LSM light path. (This can be done with the FLUOVIEW software. For details, refer to the User's Manual for the FLUOVIEW software.)
2. Loosen the four cover clamping knobs ① on the lower part of the right side panel of the scan unit, and remove the cover ②.



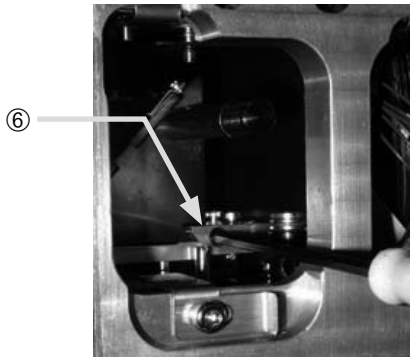
3. Using an Allen screwdriver, loosen the screw ④ retaining the guide lock plate ③, move the guide lock plate in the direction of the arrow, engage it with the pin ⑤ below the guide, and tighten the screw ④ again to lock the guide.



Before moving

After moving

4. Using the Allen screwdriver, loosen the clamping screw ⑥ retaining the DM cube.

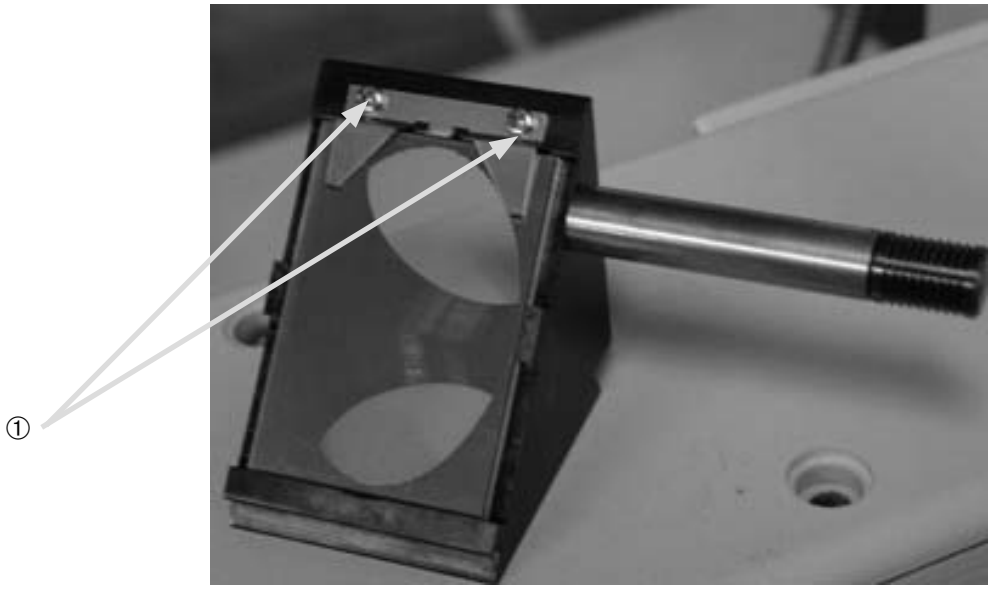


5. Pull out the DM cube insertion knob ⑦ toward you and take out the DM cube from the light path selector mechanism.

2 Fabricating the DM cube of FV10-ASU

1. Using a precision Phillips screwdriver, loosen two screws ① clamping the DM holder plate and take out the DM and DM holder plate.
2. Insert the desired DM and tighten two screws ① to clamp the DM holder plate.

◎ The applicable DM diameter is $26^{-0.1}_{-0.3} \times 38^{-0.1}_{-0.3}$ mm , with thickness of 1±0.05mm.



◎ The DM should be inserted by distinguishing the face and back. Make sure that the reflective surface (interference film surface) of the DM comes as the face.

◎ When replacing the DM cube, take care not to contaminate them with fingerprints, etc.

3 Attaching the DM cube on FV10-ASU

Attach the DM cube by reversing the removing procedure.

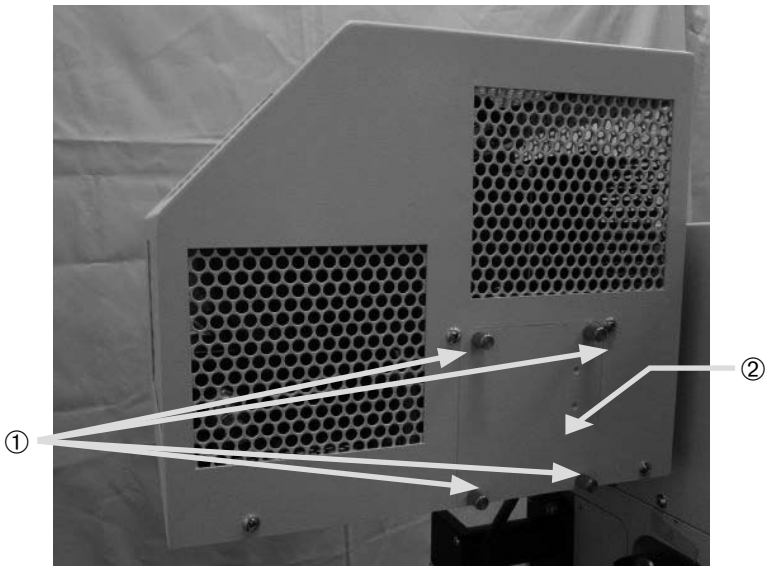
◎ To use DM cube replaced, it requires the following preparations.

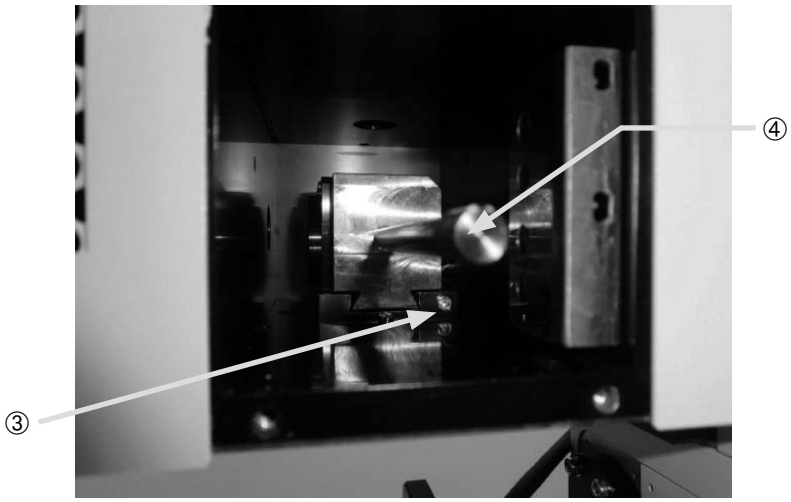
1. Log-in FV10-ASW software with the administrative right.
2. Select [Tools] – [Maintenance] and bring [FV1200 Setup] window to appear.
3. Click on ‹System Settings› button and select [Filter2] tab.
4. Verify that [Nonconfocal Detector] group box is set to " FV10-ASU".
5. Enter information of the spectral cube that was replaced in [Cube] drop down list, [Emission DM] text box, [BF CH1] text box and [BF CH2] text box respectively.
6. Click on ‹Save and Close› button and close [FV1200 Setup] window.
7. Re-boot FV10-ASW software.

4-2 FV12-HSD

1 Taken out from FV12-HSD

1. Using an Allen screwdriver, loosen the four cover clamping screws (provided with slip-off prevention mechanisms) ① on the side panel of the FV12-HSD and remove the cover ②.





2. Loosen the cube cover clamping screw ③ inside the cover.
3. Remove the cube cover by holding the cover knob ④.

2 Fabricating the spectral cube of FV12-HSD

A desired spectral cube can be fabricated by attaching a commercially available barrier filter and DM to the spectral cube frame.

Dimensional conditions for the optical components

Barrier filter $\varnothing 25^{-0.1}_{-0.3}$ mm, max. thickness 6mm
or
$\varnothing 12.7^{-0.1}_{-0.3}$ mm, Thickness 2±0.05mm

Dichroic mirror $26^{-0.1}_{-0.3}$ x $38^{-0.1}_{-0.3}$ mm, max. thickness 1.4mm
or
$\varnothing 12.7^{-0.1}_{-0.3}$ mm, Thickness 2±0.05mm

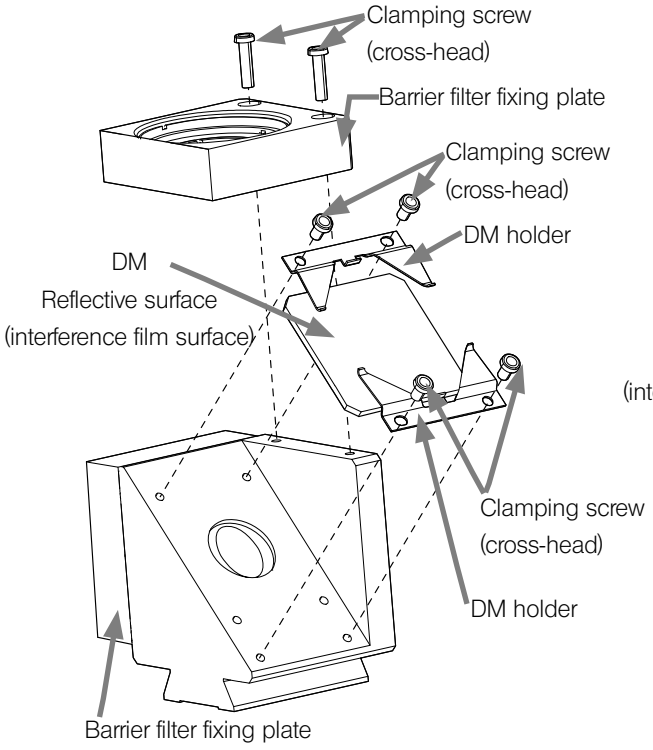
◎ When replacing the DM and barrier filter, take care not to contaminate them with fingerprints, etc.

Attaching a DM

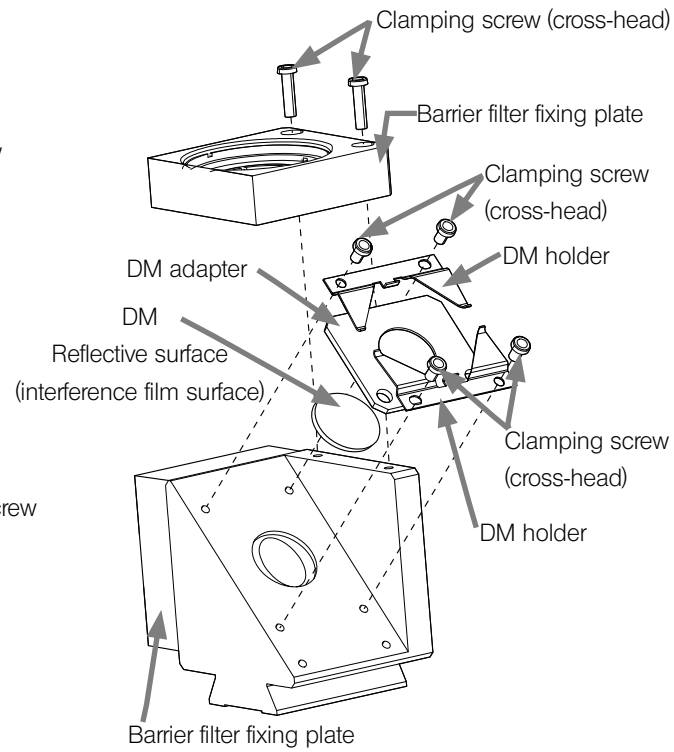
◎ Before attaching DM, detach barrier filter fixing plate on the side of DM relective surface from the spectral cube.

◎ The DM should be inserted by distinguishing the face and back. Make sure that the reflective surface (interference film surface) of the DM comes as the face.

◎ If you are using the DM Ø12.7mm, fix it with DM adapter.



DM 26×38 mm

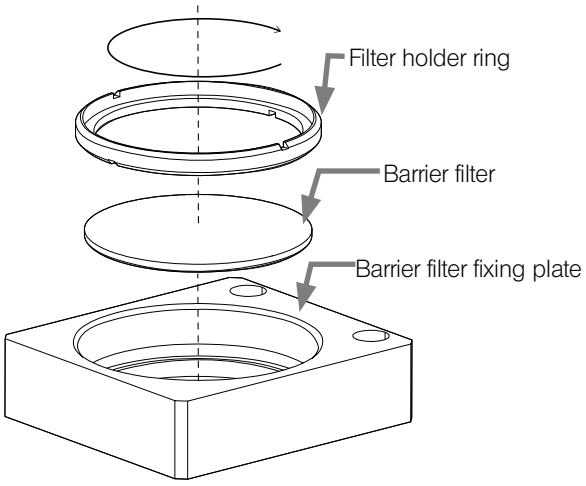


DM Ø12.7 mm

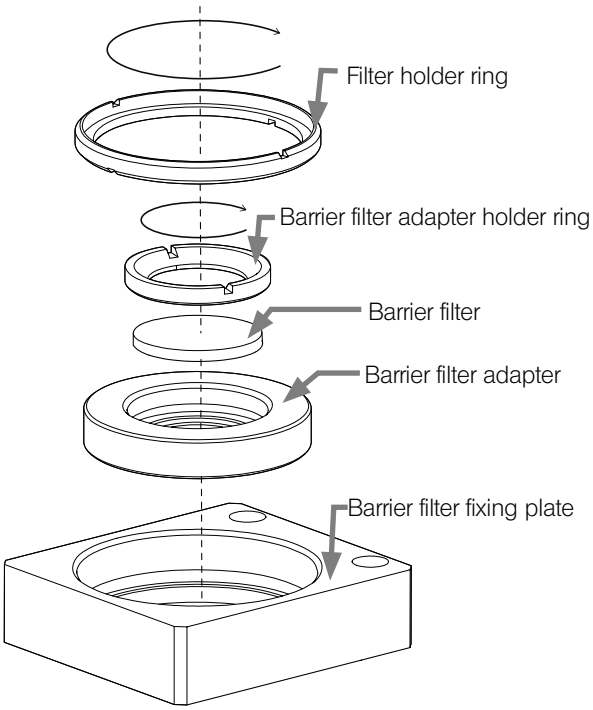
Attaching a barrier filter

◎ Insert the tip of a precision flat-blade screwdriver into the notch on the ring and turn the ring taking care not to scratch the filter to lock it.

◎ If you are using the barrier filter Ø12.7 mm, fix it with barrier filter adapter.



Barrier filter Ø25 mm



Barrier filter Ø12.7 mm

3 Attaching the spectral cube on FV12-HSD

Attach the DM cube by reversing the removing procedure.

FV10-ASW

[LightPath & Dyes] button

◎ Input the information of installed DM into the FV10-ASW software.

1. Click the [LightPath & Dyes] button in the [Image Acquisition Control] window of the FV10-ASW software.
2. Input the information of installed DM using the [LightPath & Dyes] window.

5 LAMP HOUSING INSPECTION SHEET

CAUTION **The standard service life of the lamp houses are eight (8) years of use or 20,000 hours of total power ON period, whichever is the shorter period.**

- Study the instruction manual for the lamp housing before inspection.
- For safe use of the lamp housing, we recommend performing the following inspection periodically (every time you replace the lamp and at least every 6 months).
- The table below identifies the check items to be observed. Put (X) if not applicable or (✓) if applicable.
- If there is any (✓) mark noted, immediately stop use of the product, and contact Olympus for detailed inspections or replace the lamp housing.
- If you detect an abnormality other than that listed below or with other Olympus product, also stop the use of the product and contact Olympus for detailed inspections.
- Note that service, replacement and detailed inspections are charged after expiration of the warranty period.

If you have any questions, please contact Olympus.

	Check results (Date)			
Check items	/	/	/	/
1. More than 8 years have passed since original purchase or the total power ON time has exceeded 20,000 hours.				
2. Illumination flickers when you move the lamp cable or lamp housing.				
3. Lamp cable is unusually hot to the touch.				
4. Scorching or burning odor is produced during use.				
5. Deformation, backlash, or looseness, etc. when you assemble the lamp housing. (Impossibility of removing the top section of lamp housing when you attempt to replace the lamp, etc.)				
6. Discoloration, deformation or cracking of the lamp housing.				
7. Melting, crack, deformation or solidification of the lamp cable or a wiring part.				
8. Increased frequency of servicing compared to similar devices put into use at the same time as the lamp housing.				

* When the Check Result columns become insufficient, copy this sheet.

6 SPECIFICATIONS

The specifications of Microscope, IX83P2ZF, are listed in this manual.
The specifications of FV1200 systems are listed in "FV1200 User's Manual [Safety Manual]".

Item	Specification				
Optical system	UIS2 optical system				
Microscope Frame IX83P2ZF	Motorized light path selection 1 100% for observation light path 2 50% for observation light path, 50% for left side port 3 100% for left side port Motorized 6-position revolving nosepiece (DIC slider attachable) Focusing movable range Upper side: 6.5 mm or more from the original position Lower side: 3 mm or more from the original position Original position: 1 mm above the stage surface Left side port image magnification: 1X				
Illumination Column	Column for installing the lamp housing, with 30 ° tilting mechanism. Condenser holder up/down movement range: 88 mm Condenser refocusing mechanism is available. Designated bulb 12 V, 100 W long-life halogen bulb 12V100WHAL-L (PHILIPS 7724) Power supply: Control box IX3-CBH				
Observation Tube	Type	U-BI90	U-TBI90	U-TR30-2/U-TR30H-2	U-TR30NIR
		Binocular	Tilting binocular	Trinocular	Trinocular
	Field No.	22			
	Tube inclination	45 °	35 ° to 85 °	30 °	30 °
	Interpupillary distance adjustment	50 to 76 mm			
	Diopter adjustment range	- 5 to + 5 diop			
	Intermediate tube	U-EPA2, GX-SPU, U-CA, etc.		IX-ATU	
	Light path selection	None		3 steps: ① BI 100% ② BI 20%, Camera 80% ③ Camera 100%	3 steps: ① BI 100% ② BI 50%, Camera 50% ③ Camera 100%

Stage	Type	IX3-SVR	IX2-SP
	Size	240 mm (D) x 444.5 mm (W)	240 mm (D) x 232 mm (W)
	Movement mechanism	· X- and Y-axis knob with adjustable tension. · Right long axis handle (can be mounted by reversing the right and left)	Not available
		Movement range: 75 mm vertical (Y), 114 mm horizontal (X).	Movement range: (Combined with IX-MVR) 85 mm vertical (Y), 130 mm horizontal (X).
	Option	Replaceable center plate (Ø110 mm)	Replaceable center plate (Ø110 mm)

Condenser	Type	IX3-LWUCDA	IX2-LWUCD	IX2-MLWCD	IX-ULWCD	U-UCD8 (Used together with IX-ADUCD)
	NA	0.55	0.55	0.5	0.3	When a dry top lens is used : 0.9 When an oil top lens is used : 1.4
	WD	27 mm	27 mm	45 mm	73 mm	When a dry top lens is used : 1.5 mm When an oil top lens is used : 0.6 mm
	Turret	small hole 3, large hole 4	small hole 3, large hole 2	4	4	small hole 3, large hole 5
Transportation/ storage environment	• Temperature: Min. -25 °C, Max. 65 °C • Humidity: Min. 10%, Max. 90% (without condensation)					

Item	Specification	
Fluorescence illuminator IX3-RFA L-shape fluorescence illuminator IX3-RFAL IX3-RFALFE	• UIS2 optical system • Projection magnification of the field iris diaphragm: 2X (FN 22) • Aperture iris diaphragm (IX3-RFAL only)	
	• Slider insertion slots ND filter slider	
	• Applicable microscope: IX83P2ZF	
Motorized fluorescent mirror turret IX3-RFACA Coded fluorescent mirror turret IX3-RFACS	IX3-RFACA • Observation switching: Motorized mirror unit turret (switching time approx. 0.5 sec.). • Number of mirror unit positions: 8. • Motorized shutter (Switching time: 0.2 sec.)	IX3-RFACS • Observation switching: Manual mirror unit turret • Number of mirror unit positions: 8. • Manual shutter
	• Applicable microscope: IX83P2ZF,	
Light source unit	Light source : U-HGLGPS	
Mercury lamp housing	• 100 W Mercury lamp housing : U-LH100HG • 100 W Mercury apo lamp housing : U-LH100HGAPO • Designated bulb : 100 W Mercury burner USH-103OL Power supply: Power supply unit U-RFL-T	
Xenon apo lamp housing	• 75 W Xenon apo lamp housing : U-LH75XEAPO • Designated bulb : 75 W Xenon burner UXL-75XB Power supply: Power supply unit U-RX-T	

Item	Specification
Z drift compensating microscope IX3-ZDC	• Applicable microscope frame IX83P2ZF • Applicable observation tubes U-BI90, U-TBI90, U-TR30H-2*, U-TR30NIR*, U-TR30-2* * : in combination with the IX-ATU • Applicable control box IX3-CBH • Controller U-MCZ Touch panel controller PC (software “cellSens” is required to be installed in the PC.) • Laser Laser diode Laser wavelength: 790 nm, (Class 1 IEC60825-1:2007) Laser pulse duration: 2.5 ms, Repetition rate: 100 Hz Accessible laser emission (Maximum instantaneous power): 40 μW Output from laser diode: Beam divergence: 0.1 to 0.49 rad Maximum power: 20 mW • Observation method Reflected light fluorescence observation, phase contrast observation, TIRF, DIC (applicable only with focus search) • Field number 22 • Dimensions & weight 266(W) x 63(H) x 243(D) mm, approx. 2.1 kg
Operating environment	• Indoor use. • Altitude: Max. 2,000 meters • Ambient temperature: 5 to 40 °C (41 to 104° F) • Maximum relative humidity: 80 % for temperatures up to 31 °C (88 °F) (without condensation) In case of over 31 °C (88 °F), the relative humidity is decreased linearly through 70 % at 34 °C (93 °F), 60 % at 37 °C (99 °F), and to 50 % at 40 °C (104 °F). • Supply voltage fluctuations: Not to exceed ±10% of the normal voltage. • Pollution degree: 2 (in accordance with IEC60664-1) • Installation/Overvoltage category: II (in accordance with IEC60664-1)

III. TROUBLE Q&A

On This Volume

This volume describes how to deal with troubles with the FLUOVIEW FV1200 system. If any irregularity is observed, read this volume before calling for service. If the irregularity cannot be resolved by the described remedial action, please contact Olympus for repair.

1 Troubleshooting Guide

1-1 FV1200

The system may be unable to manifest its full performance due to its usage as well as malfunction. In case a problem occurs with the system please check the following list to find appropriate countermeasures.
If the problem cannot be resolved by the described remedial action, please contact Olympus for repair.

Irregularity	Cause	Remedy
1. Laser is not output from the extremity of the objective.	The laser unit is not turned ON.	Turn on the laser unit. Make sure that the emission key is set to ON.
	The laser wavelength is not selected.	Check the laser wavelength to be used.
	The manual shutter of the fluorescence mirror unit is closed. (Manual system only)	Open the manual shutter.
	The reflective mirror inside the fluorescence mirror unit is not in the light path. (Manual system only)	Engage the reflective mirror in the light path.
	The objective is not in the light path.	Engage the desired objective in the light path. When using a manual revolving nosepiece, be sure to stop the objective in the click position.
	The laser beam is too weak.	Increase the laser intensity.
	The properties of the combined cube unit used for the ASU (auxiliary scan unit) do not match the selected laser wavelength.	Engage a DM cube unit matching the selected laser wavelength in the light path.
2. Fluorescence image cannot be observed.	The confocal pinhole diameter is too small.	Increase the pinhole diameter.
	The excitation Dichroic Mirror selection does not match the observed fluorescence wavelength and excitation laser wavelength.	Engage a DM optimum for the observed fluorescence and excitation laser wavelengths.
	The spectral dichroic mirror and barrier filter selections do not match the observed fluorescence wavelength.	Engage a spectral DM and barrier filter matching the observed fluorescence in the light path.
	The acquisition wavelength region setting is not suitable for the observed fluorescence wavelength. (Spectral detection system only)	Set an acquisition wavelength region matching the observed fluorescence.
	The fluorescent dyeing method and excitation wavelength do not match each other.	Select a laser optimum for the fluorescent dyeing method.
	Focus is not adjusted.	Adjust the focus.
	The PMT voltage of the detector is too low.	Increase the PMT voltage.
2. Fluorescence image cannot be observed.	The offset value is too large.	Decrease the offset value.
	The detector for the channel to be detected is not selected.	Select the detector.

Irregularity	Cause	Remedy
3. Transmitted image cannot be observed.	The transmitted light detection channel is not selected.	Select the transmitted light detection channel.
	The transmitted light filter for the microscope is in the light path.	Disengage the filter from the light path.
	The PMT voltage of the transmitted light detection channel is too low.	Increase the PMT voltage.
	The offset value for the transmitted light detection channel is too large.	Decrease the offset value.
4. Image is disturbed.	The system installation location is subject to excessive vibrations.	Contact Olympus.
	Extraneous light such as the light of a fluorescent lamp is detected.	Turn the room light low before acquiring image.
5. Reflected light (laser light) enters the fluorescence image.	The barrier filter is set erroneously or absent.	Engage a barrier filter that can cut the excitation laser wavelength in the light path.
	The set acquisition wavelength is overlapped with or too close to the excitation laser wavelength. (Spectral detection system only)	Select an acquisition wavelength that is not interfered with by the laser wavelength. (Note that, when the confocal pinhole is large and the BS20/80 excitation DM is used, penetration of laser light may become large.)
	The barrier filter that can cut the wavelength of the laser light irradiated from the ASU (auxiliary scan unit) is not selected. In the case of a spectral detection system, the acquisition wavelength setting may be inappropriate.	Engage a barrier filter that can cut the laser wavelength from the ASU in the light path. With a spectral detection system, change the acquisition wavelength setting.
6. Fluorescence image is poor.	The front lens of the objective is dirty.	Clean the objective front lens by wiping it with a piece of gauze.
	When an objective with correction collar is in use, the correction collar is adjusted improperly.	Adjust the correction collar properly.
	The cover glass thickness is inappropriate.	Use a cover glass with thickness of 0.17 mm.
7. Fluorescence image is dark and noisy.	The laser beam is too weak.	Increase the laser intensity.
	The fluorescent dyeing method and excitation wavelength do not match each other.	Select a laser optimum for the fluorescent dyeing method.
7. Fluorescence image is dark and noisy.	The excitation Dichroic Mirror selection does not match the observed fluorescence wavelength and excitation laser wavelength.	Engage a DM optimum for the observed fluorescence and excitation laser wavelengths.

Irregularity	Cause	Remedy
	The spectral dichroic mirror and barrier filter selections do not match the observed fluorescence wavelength.	Engage a spectral DM and barrier filter matching the observed fluorescence in the light path.
	The acquisition wavelength region setting is not suitable for the observed fluorescence wavelength. (Spectral detection system only)	Set an acquisition wavelength region matching the observed fluorescence.
	The confocal pinhole diameter is too small.	Increase the pinhole diameter.
	The scanning rate is too high.	Decrease the scanning rate.
	The HV setting is too high.	Decrease the HV and increase the gain. An alternative remedy is to decrease the scanning rate and decrease the HV.
	The width of the acquisition wavelength region is too small.	Increase the width of the acquisition wavelength region.
	Dyeing is too pale.	Perform optimum fluorescent dyeing.
8. Image is irregularly blurred or the brightness is uneven.	The specimen or stage is tilted.	Install the specimen and stage properly.
9. Observed image is out of focus.	The focus is adjusted improperly.	Adjust the focus in visual observation.
10. The intensity of part of the wavelength region of the spectral characteristic data of fluorescence is dropped.	The spectral characteristics of the fluorescence are affected by those of the excitation dichroic mirror used in double excitation.	Use an excitation DM that does not affect the spectral characteristic data of fluorescence.
11. Flare is observed.	The glass in use is not fluorescence-free glass.	Use fluorescence-free glass.
	The specimen is overstained.	Perform optimum dyeing again or increase the offset value.
12. Visual fluorescent light observation is impossible.	The light path selector in the SU is not set for the visual observation light path.	Select the visual observation light path
	The light path selector in the SU is not set for the visual observation light path	Open the shutter for the mercury burner.
	The mirror unit incorporating a dichroic mirror is not present in the turret of the illuminator	Engage a mirror unit containing DM in the light path.
13. The light from the laser for the ASU (additional scan unit) is not output.	The properties of the combined DM cube unit for ASU do not match the irradiated laser wavelength	Engage a DM cube unit matching the laser wavelength in the light path
	The combined DM cube unit for ASU is not in the light path.	Engage the DM cube unit in the light path.

1-2 IX83P2ZF

Problem	Cause	Remedy
a) The bulb does not light.	Power cord of the IX3-CBH is unplugged.	Plug the power cord into a power outlet.
	Main switch of the IX3-CBH is not " I " ON.	Set the main switch to " I " (ON).
	Bulb is burnt out.	Replace the bulb.
b) The bulb lights but the field of view is dark.	Lamp voltage is too low.	Increase the light intensity to an optimum voltage.
	Condenser is not well positioned.	Adjust the condenser height until the field iris diaphragm image is formed in the specimen plane.
	Light path selector knob is set for the left side port light path.	Change the light path.
	Too many filters are used.	Reduce the number of filters to the minimum required.
	Stage center plate is engaged in the light path.	Move the stage and place the specimen again.
	Field iris diaphragm is not opened wide enough.	Open the field iris diaphragm sufficiently.
c) Field of view is obscured or not evenly illuminated.	An objective that falls outside the condenser's illumination range is used.	Use a condenser that matches the objective.
	Field iris diaphragm is not properly centered.	Center the field iris diaphragm correctly
	Field iris diaphragm is stopped down too far.	Open the field iris diaphragm sufficiently.
	A filter is stopped in an intermediate position.	Set the filter at the appropriate position.
d) Dirt or dust is visible in the field of view.	Dirt/dust on the specimen.	Clean thoroughly.
	Dirt/dust on the eyepieces.	
	Dirt/dust on a mirror unit.	
	Dirt/dust on the optical element.	
	Condenser is not correctly positioned and the frost filter Ø45 mm (45FR) or filter is focused.	Adjust the condenser height until the field iris diaphragm image is formed in the specimen plane.
e) Image glares.	Condenser is raised too high.	Lower to the proper position
	Aperture iris diaphragm is stopped down too far.	Open the aperture iris diaphragm.

Problem	Cause	Remedy
f) Error happens at the time of system starting-up.	There are some mistakes in the order of turning on the power switches.	Turn off all of the power switches, then turn on the switches again in the correct order. As to the correct order in turning on the power switches, refer to the page 12.
g) Visibility of observed image is poor. • Image is not sharp. • Contrast is poor. • Details are poorly visible.	Objective in use is not designed for UIS2 series.	Replace with an objective designed for UIS2 optics.
	Correction collar on the objective equipped with correction collar is not adjusted.	Adjust the correction collar to acquire the best contrast.
	Front lens of the objective is dirty.	Clean the objective.
	The immersion oil appropriate with an oil immersion objective is not used.	Use Olympus immersion oil with the oil immersion objective, Olympus silicone oil with the silicone immersion objective and water with the water immersion objective.
	Immersion oil contains bubbles.	Remove bubbles.
	Inappropriate slide or cover glass thickness.	Replace with glass of appropriate thickness.
	Glass components (condenser, objective, eyepieces, culture vessels, etc.) are dirty.	Clean thoroughly.
	Ring slit and phase plate are not centered.	Center them correctly.
	A plastic culture vessel is used.	Replace the plastic culture vessel with a glass vessel.
h) A part or one side of the field of view is blurred.	Specimen is tilted with respect to the stage.	Place the specimen correctly on the stage and secure it with the stage clip.
i) Field of view of one eye does not match that of the other.	The interpupillary distance is incorrect.	Adjust the interpupillary distance.
	Incorrect diopter adjustment.	Adjust the diopter.
	You are not accustomed to parallel optical axis.	When looking into the eyepieces, do not stare at image from the beginning but see the overall field of view. It is sometimes recommended to turn your eyes away from the eyepieces, look far off and look into the eyepieces again.

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