Nikon

Microscope

ECLIPSE 90i

Instructions

<Reference>

Introduction

Thank you for purchasing this Nikon product.
This instruction manual, which describes basic microscope operations, is intended for users of the Nikon ECLIPSE 90i microscope.
To ensure correct use, please read this manual carefully before operating the product.

- This manual may not be reproduced or transmitted in whole or in part without Nikon's express consent.
- The contents of this manual are subject to change without notice.
- Although every effort has been made to ensure the accuracy of this manual, errors or inconsistencies may remain. If you note any points that are unclear or incorrect, please contact your nearest Nikon representative.
- Some of the products described in this manual may not be included in the set you have purchased.
- Make sure you have read the manuals for any other products attached to or to be used with this product (super high-pressure mercury lamp power supply, high-intensity light source, etc.).

Warning/Caution symbols used in this manual

Although Nikon products are designed to provide the utmost safety, ignoring safety precautions or improper use may result in personal injury or property damage, as well as voiding the terms of the warranty. To ensure safe use, please read the instruction manual carefully and thoroughly before trying to operate the instrument. Do not discard this manual. Store in a convenient location near the product for ready reference.
In this manual, safety precautions are indicated by the following symbols. For safe, correct use of the product, always follow the instructions indicated by these symbols.

Symbol	Meaning
 **WARNING**	Disregarding instructions indicated by this symbol may result in death or serious injury.
 **CAUTION**	Disregarding instructions indicated by this symbol may result in injury or property damage.

Meaning of symbols used on the product

When appearing on the product, the symbols below indicate the need for caution at all times during use.
Consult the instruction manual and read the relevant instructions before attempting to use or adjust any part to which the symbol has been affixed.

	**Caution! Biohazard** This symbol found on the stage indicates the following: • WARNING: Contact between sample and the product may result in biohazard risks. • To avoid biohazard contamination, avoid touching the contaminated portion with bare hands. • Decontaminate the contaminated part according to the standard procedure specified for your laboratory.
	**Caution for heat** This symbol found on the lamphouse of the ECLIPSE 90i indicates the following: • The lamp and surrounding areas (including the lamphouse) become very hot during and immediately after a period of illumination. • Risk of burns. Do not touch the lamp or surrounding areas during or immediately after a period of illumination. • Make sure the lamp and surrounding areas have cooled sufficiently before attempting to replace the lamp

Abbreviations used in the manual

The product names and abbreviations used in this manual are given below.

The manual uses the following abbreviations:

Name of device	Abbreviation
Microscope ECLIPSE 90i	90i
C-ER Eye Level Riser	Eye Level Riser
C-TE Ergonomic Binocular Tube	Ergonomic Binocular Tube
C-TEP DSC Port for Ergonomic Binocular Tube	DSC Port
D-DH Digital Imaging Head M	DIH-M
D-DH-E Motorized Digital Imaging Head E	DIH-E
D-PS 90i Power Supply	Power Supply Unit
D-LH Precentered Lamphouse	Lamphouse
DS Camera Head DS-5M	Camera Head
DS Camera Control Unit DS-L1	DS-L1
DS Camera Cable	Camera Cable
Super High-pressure Mercury Lamp Power Supply	Mercury Lamp Power Supply
Super High-pressure Mercury Lamphouse	Mercury Lamphouse
D-CH 90i Condenser Holder	Condenser Holder
D-CUD-E Motorized Universal Condenser Dry	Motorized Universal Condenser
D-CUD Universal Condenser Dry	Universal Condenser
D-N7-E Motorized Septuple Nosepiece	Motorized Nosepiece
D-NF-E 90i Motorized ND Filter Unit	Motorized ND Filter

How to use this instruction manual

This instruction manual is composed of two parts, as below:

Manual 1 "Microscopy" describes basic microscope operations that you must follow. Please read this manual carefully before operating the product.

Manual 2 "Reference" describes the operations of each attachment. Please read an appropriate section as necessary.

	Title	**Importance**	**Content**
Manual 1	Microscopy	Must be read	Safety precautions Microscopy, Assembly, Troubleshooting, Care and maintenance, Technical Specifications
Manual 2	Reference	As necessary	Detailed operations of attachments

Contents

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Individual Operations

Item	Title	Operating sections
1	Power ON/OFF	Power switch
2	Setting the information for mounted components in the 90i	PC
3	Brightness Adjustment	Brightness control knob, preset switch, ND filter, NCB filter, filter holder, DS-L1, PC
4	Optical Path Switching	Optical path switching lever (trinocular eyepiece tube, DIH-M), DIH optical path changeover switch (DIH-E), DS-L1, PC
5	Vertical Stage Motion	Focus knob, coarse focus switch, DS-L1, escape switch, PC
6	XY Stage Motion	X stage knob, Y stage knob, XY knob torque adjustment screws
7	Stage Rotation	Rotation clamp screw
8	Selection of Magnification	Nosepiece, DIH-M zoom knob, DS-L1, PC
9	Nosepiece Rotation	PC
10	Autofocus	DS-L1, PC
11	Diopter Adjustment	Diopter adjustment rings
12	Interpupillary Adjustment	Eyepiece sleeve
13	Adjusting the Observation Position	Ergonomic binocular tube
14	Adjusting the Condenser Position	Condenser focus knob, condenser centering screws
15	Adjusting the Aperture Diaphragm	Condenser aperture diaphragm objectives, aperture diaphragm open/close switch, DS-L1, PC
16	Selecting a Condenser	Condenser
17	Adjusting the Field Diaphragm	Field diaphragm open/close switch, DS-L1, PC
18	Oil Immersion Operation	Oil immersion objectives, oil immersion condensers
19	Water Immersion	Water immersion objectives
20	Fluorescence observation	DIH-M, DIH-E, DS-L1, PC
21	Selecting Fluorescent Filters	Filter cube
22	Excitation Light Balancer	Excitation light balancer
23	Image Capture	Camera, DS-L1, PC
24	Switching of Microscopy Methods	DS-L1, PC
25	Switches	PC
26	Buzzer	PC

For use of the DS-L1 for operation control, refer to the instruction manual provided with the DS-L1.

For use of a PC for operation control, refer to the instruction manual provided with the software.

1 Power ON/OFF

1.1 Turning on/off the microscope

When the power switch of the power supply unit is in the ON position (button depressed), power is supplied to the microscope. (The LED remains lit.)

When the power switch is in the OFF position (button in projected position), there is no power supplied to the microscope.

1.2 Turning on the Epi-fl attachment light source (mercury lamp)

Refer to the operating manual provided with the Mercury lamp power supply.

Always observe all warnings and precautions described in the manual.

2 Setting the information for mounted components in the 90i

2.1 Setting the information of mounted objectives, filter cubes, and condenser modules

When using the 90i for the first time, be sure to set the information for mounted components such as objectives, filter cubes, and condenser modules in the 90i. If this information is not set, the 90i will not operate properly. Do not skip this process.

When using the motorized universal condenser, set the information for the condenser modules.

[Setting method]

The settings can be entered by using iEZSetup of the i Series Support Tools software. The interactive menu of iEZSetup allows you to enter necessary settings in sequence.

For details, refer to Chapter 3 "Using iEZSetup" of the i Series Support Tools software manual.

[Factory settings]

Objectives:	Not mounted
Filter cubes:	Not mounted
Condenser modules:	Not mounted

2.2 Changing the information for mounted objectives, filter cubes, and condenser modules

When objectives, filters cubes, or condenser modules are replaced, the information of the components can be changed individually.

[Setting method]

For this setting, use iSetup of the i Series Support Tools software.

For details, refer to Chapter 5 "Using iSetup" in the i Series Support Tools software manual.

3 Brightness Adjustment

Image brightness can be adjusted by the following methods:

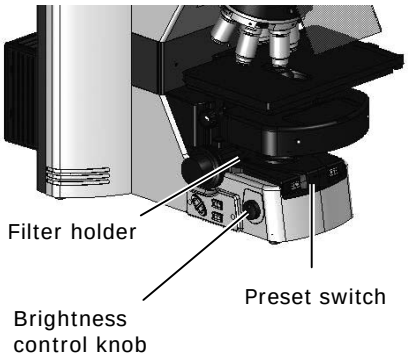
	Method	Operating controls	Description
Transmitted image	Adjusting the lamp voltage (Shifts the color temperature.)	Brightness control knob, DS-L1, PC	3.1
		Preset switch	3.2
	Inserting/removing the ND filter	ND filter IN/OUT switches	3.3
Epi-fl image	ND filter	Inserting/removing ND filter (DIH-M, DIH-E)	3.4
	Adjusting the aperture diaphragm (for Epi-illumination)	Aperture diaphragm lever (DIH-M, DIH-E)	3.5
(Monitor image)	Camera adjustment	Application software for camera control: Display mode, exposure mode, exposure compensation, camera gain adjustment, etc.	3.6

3.1 Adjustment using the brightness control knob

After making sure that the preset switch lamp is not lit, turn the brightness control knob. (If the preset switch lamp is lit, the brightness control is inoperable.)

Adjusting brightness with the brightness control knob will affect the lamp color temperature and alter the color balance of the image. If accurate color reproduction is critical, set a commercially available color compensation filter on the filter holder to make brightness adjustments.

If accurate color reproduction is critical, do not use the brightness control knob to adjust brightness. Instead, press the preset switch and move the NCB11 filter (color temperature adjustment filter) into the optical path. This will provide optimum color accuracy.



Brightness control knob	Image brightness	Image color
Clockwise turn	Becomes brighter.	Color balance shifts to blue.
Counterclockwise turn	Becomes darker.	Color balance shifts to red.

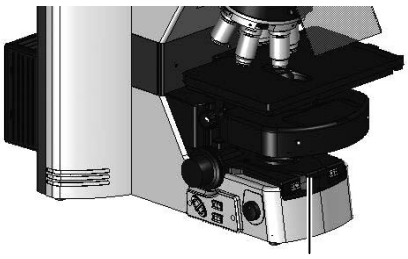
For use of the DS-L1 for operation control, refer to the instruction manual provided with the DS-L1.

For use of a PC for operation control, refer to the instruction manual provided with the software.

3.2 Adjustment using the preset switch

When the preset switch lamp is lit, the lamp voltage is 9 V. The best color reproducibility can be achieved by inserting the NCB11 filter into the optical path in this condition.

If the preset switch lamp is not lit, the brightness returns to the setting made with the brightness control knob.

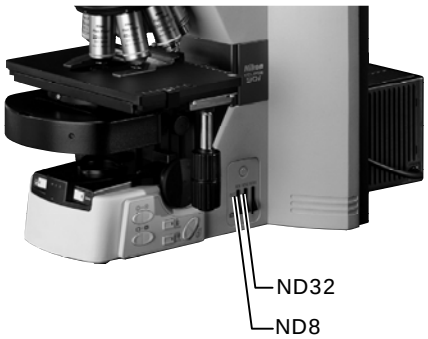


3.3 Adjustment with the ND filter IN/OUT switch

The two forward-located switches of the three closely arranged switches are for ND filters. The ND filters are used to adjust light intensity. Higher ND values correspond to reduced transmissivity and darker images. The color balance is not affected. Push the IN side to move the ND filter into the optical path and darken the image.

ND8: Reduces light intensity to 1/8.

ND32: Reduces light intensity to 1/32.



Brightness	ND8	ND32
1	-	-
1/8	○	-
1/32	-	○
1/256	○	○

3.4 Adjustment with the ND filter of the DIH-M or DIH-E

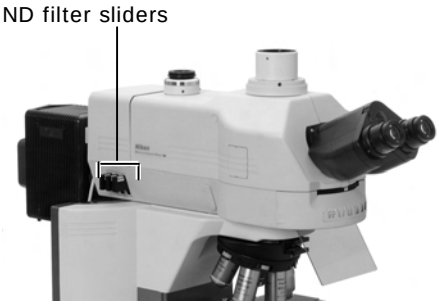
Pushing in the ND filter slider moves the ND filter into the optical path and darkens the fluorescent image.
ND filters are used to adjust light intensity. Higher filter numbers correspond to lower transmission rates (i.e., darker images). ND filters do not affect color balance.

ND4: Reduces light intensity to 1/4.

ND8: Reduces light intensity to 1/8.

ND16: Reduces light intensity to 1/16.

3 Brightness Adjustment



DIH-M or DIH-E

Brightness	ND4	ND8	ND16
1	-	-	-
1/4	○	-	-
1/8	-	○	-
1/16	-	-	○
1/32	○	○	-
1/64	○	-	○
1/128	-	○	○
1/512	○	○	○

3.5 Adjusting the aperture diaphragm for Epi-illumination

In Epi-fluorescence microscopy, image brightness can be adjusted by varying aperture diaphragm size. Stopping down the aperture diaphragm darkens the image, while opening up the aperture diaphragm makes the image brighter.

Use the aperture diaphragm of the DIH-M or DIH-E after performing the centering procedures. For a detailed discussion of this topic, refer to "20.5 Aperture diaphragm for Epi-illumination."

3.6 Camera adjustment (adjusting the brightness of the image on the monitor)

When observing images captured by the camera and displayed on the monitor, you can adjust brightness by varying camera adjustment parameters, such as display mode, exposure mode, metering mode, exposure compensation, and image level adjustment.
For detailed discussion, refer to the operating manual provided with the camera or camera control software.

4 Optical Path Switching

4.1 Optical path distribution

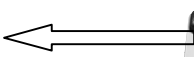
With the ergonomic binocular tube or trinocular eyepiece tube, the optical path switching lever allows distribution of light to the binocular section and camera port.

	Position of optical path switching lever	Optical path distribution (%)	
		Binocular section	Camera port
Ergonomic binocular tube	Pushed in	100	0
	Extended	50	50
Trinocular eyepiece tube T	Pushed in	100	0
	Extended by one notch	20	80
	Extended by two notches	0	100
Trinocular eyepiece tube F	Pushed in	100	0
	Extended	0	100

When the DIH-E is used, the DIH optical path changeover switch allows the distribution of light to the binocular section, front port, or rear port.

Each time the DIH optical path changeover switch is pressed, the full (100%) light path changes in the following sequence.

Binocular section → Rear port → Front port



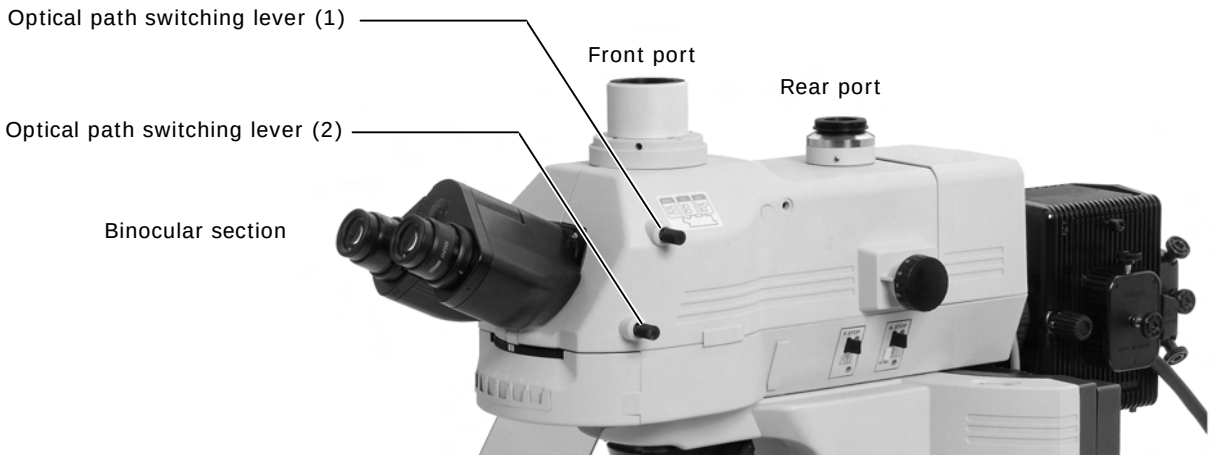
The selection is indicated by the DIH light path position indicator.

4 Optical Path Switching

With the DIH-M, two optical path switching levers allows distribution of light to the binocular section, front port and rear port.

Position of optical path switching lever (1) (top lever)	Position of optical path switching lever (2) (bottom lever)	Optical path distribution (%)		
		Binocular section	Front port	Rear port
Extended	Extended	100	-	-
Pushed in	Pushed in	-	100	-
Extended	Pushed in	-	-	100

It is not possible to have only the optical switching lever (2) extended. Applying excessive force may result in damage.

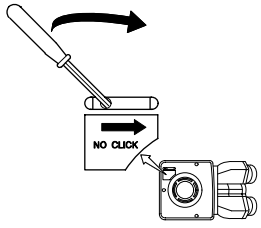


For use of the DS-L1 for operation control, refer to the instruction manual provided with the DS-L1.

For use of a PC for operation control, refer to the instruction manual provided with the software.

4.2 Disabling the clicking of the optical path switching lever

The trinocular eyepiece tubes T and F have a “NO CLICK” switch on their tube attaching surfaces. Slide this switch in the direction of the arrow with the tip of a pointed tool to disable clicking for the optical path switching lever. Set the switch to this position if you need to eliminate the slight vibrations resulting from the clicking action.



5 Vertical Stage Motion

5.1 Prohibited action

Avoid the following action, which can cause equipment malfunctions.

- **Turning the right and left focus knobs in mutually opposing directions.**

5.2 Knob rotation direction and stage motion direction

Turn the focus knob to raise or lower the stage and to adjust image focus.

To lower the stage	Turn the knob toward the front.
To raise the stage	Turn the knob toward the back.

Focus knob

5.3 Number of knob turns and distance of stage travel

In Auto mode, the feed rate of the focus knob automatically changes according to the N.A. of the objective. In Manual mode, it can be set to Coarse, Fine, or Extra Fine.

[Setting method]

The setting can be changed by using iSetup of the i Series Support Tools software.

For details, refer to "5.11.2 Control-Related Setup" in the i Series Support Tools software manual.

[Factory setting]

Manual

[Coarse/Fine/Extra Fine switching in Manual mode]

Each time the coarse focus switch is pressed, the mode changes in the following sequence: Coarse → Fine → Extra Fine → Coarse. At the same time, the buzzer is activated and the coarse focus switch lamp status changes.

	Amount of travel	Lamp indication	Buzzer
Coarse	2500 to 10 μm/sec Varies according to the speed of knob rotation.	ON	Long beep (once)
Fine	100 μm/rotation	Off	Short beep (once)
Extra Fine	25 μm/rotation	Flashing	Short beeps (twice)

Coarse focus switch
(Also located on the other side)

5.4 Changing the vertical stage motion range and its lower limit

The vertical stage motion range (coarse/fine focus stroke) extends from a position 2 mm above the focal point (reference position) to a position 15 mm below. However, when the motorized universal condenser is used, the lower limit of the vertical stage motion can be changed to a position 25 mm below the focal point (reference position).

[Setting method]

For this setting, use iSetup of the i Series Support Tools software.

For details, refer to "5.11.1 Setting Up the Up/Down Focus Motion" in the i Series Support Tools software manual.

[Factory setting]

–15 mm

5.5 Escape

When the escape switch is pressed, the stage lowers (escape operation) 5 mm, and the escape switch lamp lights. When the escape switch is pressed again, the stage rises 5 mm to its original position, and the escape switch lamp turns off.

The escape operation is not activated if the distance between the current position and the lower limit of the vertical stage motion is less than 5.5 mm. Also, focus knob is disabled so that the stage cannot be moved up/down during and after an escape operation. (Note that the stage vertical motion can be operated form iContol or the DS-L1.)

Note:

The nosepiece can be rotated at an escape position. However, it should be noted that doing so may cause the tip of the objective to contact the specimen when the stage returns to the original position if the microscope's adjustments (e.g., the diopter adjustment) have not been properly set.

6 XY Stage Motion

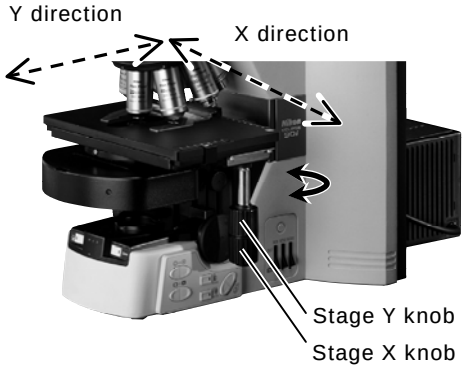
6.1 Prohibited action

Avoid the following action, which can cause equipment malfunctions.

- **Moving the stage or the specimen holder to the left and right by holding stage or holder directly.**

6.2 Knob rotation direction and stage motion direction

To move the stage in the X or Y direction, rotate the stage X knob or stage Y knob.



6.3 Adjusting the knob heights

The heights (positions) of the X knob and Y knob can be changed. Hold the knob and move it along its vertical axis to the desired height. The knobs can be moved up/down by 18 mm.

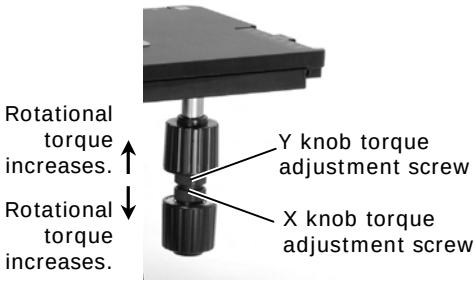
6.4 Adjusting the knob rotation torque

When the X knob and Y knob are moved to the top and bottom positions, the torque adjustment screws can be found between the knobs.

Turning the torque adjustment screw to move them closer towards the respective knobs increases rotational torque.

(To increase rotational torque, turn the adjustment screw counterclockwise and clockwise, as viewed from above, for the Y knob and X knob, respectively.)

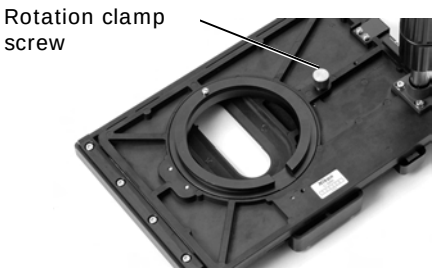
Avoid loosening these screws excessively. If they are too loose, the top surface of the stage may move, even at a very light touch.



7 Stage rotation

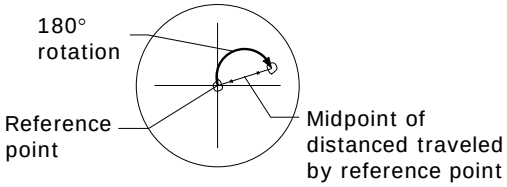
7.1 Stage with rotating mechanism

Loosen the rotation clamp screw on the bottom surface of the stage to allow the stage to swivel.



7.2 Centering the stage

(1) Move the 10x objective into the optical path and focus on the specimen. Select a specific point on the specimen as a reference point, then move the reference point to the center of the field of view using the stage knobs.



(2) Loosen the stage rotation clamp screw and rotate the stage approximately 180°.

(3) If the reference point deviates from the center of the field of view, insert the hex driver into the stage-centering screws* (2 locations) and adjust by moving the stage a distance equal to the distance traveled by the reference point from the center of the field of view.

(4) Using the stage knobs, return the reference point to the center of the field of view.

(5) Move the 40x objective into the optical path and repeat steps 2 through 4.

***** The stage-centering screws (also used as mounting screws) are located at two points on the back of the elevating section.

8 Selection of Magnification

8.1 Changing the magnification using objectives and optical zoom

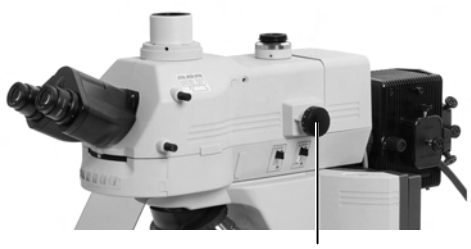
To switch magnifications, press the objective changeover switch to insert an objective with a different magnification into the optical path.

If the DIH-M is attached to the microscope, the zoom knob on the right side allows adjustment of magnification between 0.8x and 2x for rear port observation.

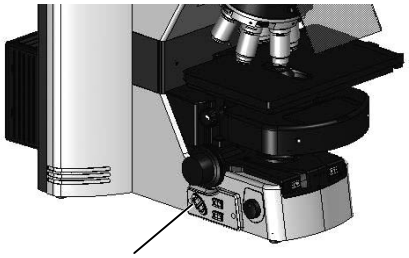
Clockwise rotation of the zoom knob increases magnification. The number at the pointer indicates the effective magnification. The knob clicks at each 0.2x increment.

Total magnification of rear port observation is calculated as follows:

Magnification of objective x Magnification of zoom knob



Zoom knob



Objective changeover switch

For use of the DS-L1 for operation control, refer to the instruction manual provided with the DS-L1.

For use of a PC for operation control, refer to the instruction manual provided with the software.

8.2 Canceling the automatic adjustment of motorized attachments when switching objectives

When objectives are switched, the field diaphragm, vertical stage position, motorized universal condenser (turret, aperture diaphragm), and motorized ND filter are automatically adjusted to optimum conditions. These automatic adjustment operations can be canceled (turned OFF) for individual motorized attachments. The factory settings are recommended for normal use.

[Setting methods]

The setting can be changed by using iSetup of the i Series Support Tools software.

For details, refer to "5.10.2 Objective, Optical Path Switching, and Optical Zoom Interlock Setups" in the i Series Support Tools software manual.

[Factory setting]

ON

8.3 Changing the size of the diascopic field diaphragm when switching objectives

The size of the field diaphragm is automatically adjusted to an optimum value according to the size of the field of view when objectives are switched. The size of the field diaphragm in this adjustment can be changed in the direction of increased stop-down.

[Setting methods]

The setting can be changed by using iSetup of the i Series Support Tools software.

For details, refer to "5.10.3 Compensation Setup during Interlocking" in the i Series Support Tools software manual.

[Factory setting]

1.00
(A value of "1.00" is given to the size of a field diaphragm image that is just outside the field of view. For adjustment, specify the rate in relation to this maximum value. Note that only values less than 1.00 can be set.)

8.4 Changing the size of the aperture diaphragm when switching objectives

When the motorized universal condenser is used, the aperture diaphragm size is automatically adjusted when switching objectives so that the image contrast becomes optimal. The size of the aperture diaphragm in this adjustment can be changed in the direction of increased stop-down.

[Setting methods]

The setting can be changed by using iSetup of the i Series Support Tools software.

For details, refer to "5.10.3 Compensation Setup during Interlocking" in the i Series Support Tools software manual.

[Factory setting]

1.00
(A value of "1.00" is given to 75% of the size of the N.A. of the objective. For adjustment, specify the rate in relation to 1.00. Note that only values less than 1.00 can be set.)

8.5 Changing the amount of adjustment of the motorized ND filter when switching objectives

When the motorized ND filter is used, the motorized ND filter automatically adjusts the brightness in switching of objectives so that the image brightness remains constant. The overall brightness in this automatic adjustment can be changed to a brighter level or a darker level.

[Setting methods]

The setting can be changed by using iSetup of the i Series Support Tools software.

For details, refer to "5.10.3 Compensation Setup during Interlocking" in the i Series Support Tools software manual.

[Factory setting]

0.75
(A value of "0.75" is given to the brightness achieved by the automatic adjustment.)

9 Nosepiece Rotation

9.1 Preventing contact between the objective and specimen when switching objectives

The 1x and 2x objectives have very long focal depths compared to the objectives of other magnifications. Therefore, the objective can be easily brought too close to the specimen during observation. For instance, if the objective is at the position closer than the parfocal distance of 60 mm, switching to the maximum-magnification objective mounted at the adjacent position can cause the tip of the objective to contact the specimen.

To avoid contact between an objective and a specimen, it is possible to activate the following operation for the nosepiece.

1. Disallows the rotation of the nosepiece when switching from a 1x or 2x objective to an objective with a W.D. (working distance) of less than 1 mm.
2. Activates the stage escape operation before rotating the nosepiece when switching from a 1x or 2x objective to an objective with a W.D. (working distance) of less than 1 mm.
   (The stage escape position will be 5 mm lower than the current position. If the stage cannot be lowered by 5 mm, it will descend as far as it can go.)

[Setting method]

For this setting, use iSetup of the i Series Support Tools software.

For details, refer to "5.7.3 Special Control Setup" in the i Series Support Tools software manual.

[Factory setting]

OFF

9.2 Skipping positions not mounted with an objective

Positions not mounted with an objective can be skipped. This function uses the registered information for mounted objectives. Be sure to enter "Not mounted" for positions where no objective is installed.

[Setting method]

For this setting, use iSetup of the i Series Support Tools software.

For details, refer to "5.7.3 Special Control Setup" in the i Series Support Tools software manual.

[Factory setting]

OFF

9.3 Preventing the adhesion of oil/water to dry objectives

When dry objectives and liquid immersion objectives are used together, it is possible to activate the following operation for the nosepiece in order to prevent the adhesion of oil or water to dry objectives.

1. Disallows the rotation of the nosepiece when the objective before or after the switching is a liquid immersion type.
2. Activates the stage escape operation before rotating the nosepiece when the objective before or after the switching is a liquid immersion type.
   (The stage escape position will be 5 mm lower than the current position. If the stage cannot be lowered by 5 mm, it will descend as far as it can go.)

[Setting method]

For this setting, use iSetup of the i Series Support Tools software.

For details, refer to "5.7.3 Special Control Setup" in the i Series Support Tools software manual.

[Factory setting]

OFF

10 Focus Adjustment

10.1 Parfocal compensation

By presetting the focal point for each objective, focus problems resulting from switching objectives can be automatically corrected. This correction function can be turned ON or OFF.

[Setting method]

The focal point can be set by using iControl of the i Series Support Tools software. The correction function can be turned ON/OFF using iSetup of the i Series Support Tools software.

For details, refer to "4.5.3 Setting Up the Focus Position for Up/Down Motion" and "5.11.1 Setting Up the Up/Down Focus Motion" in the i Series Support Tools software manual.

[Factory settings]

Focus position: Not set

Correction: ON

10.2 Autofocus

When a digital camera (L1 or U1) is connected to the 90i, the autofocus function can be used.

The autofocus function can be controlled by using iControl of the i Series Support Tools software and the DS camera control unit DS-L1. For operational details, refer to the instruction manual provided with the respective product.

Observe the following precautions when using autofocus.

CAUTION

Before using autofocus, check your microscope for the following. Disregarding these precautions may result in damage to the objective or sample.

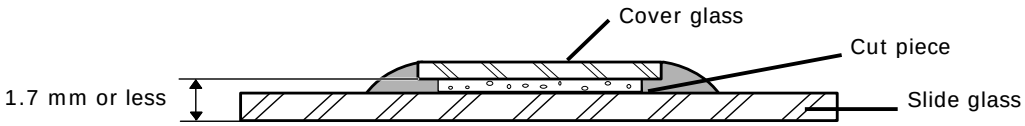
Precautions for samples

1) The autofocus function may be used with samples mounted on specimen slides with cover glass in the sizes indicated below.

- Thickness:
  0.9 mm to 1.7 mm, inclusive [thickness of slide glass + thickness of cut piece (not including the cover glass)]
- Slide glass:
  Complies with JIS R3703 or ISO 8037.
- Cover glass:
  Complies with JIS R3702 or ISO 8255.

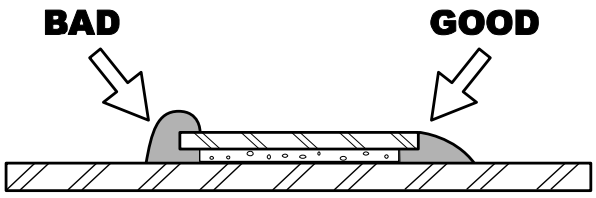
  [JIS R3702]
  Thickness: 0.12–0.17 mm for No. 1; 0.15–0.18 mm for No.1-S

  [ISO 8255]
  Thickness: 0.13–0.17 mm for No. 1; 0.15–0.17 mm for No.1-H



2) The autofocus function may force contact between the sample and the tip of the objective, resulting in damage to both. For samples like the ones shown below, adjust focus with the focus knob rather than relying on autofocus.

- A sample with sealing agent rising excessively above the cover-glass top surface.
- A sample whose sealing agent is damp, not completely dry
- A sample with sealing agent adhering to the surface of the cover glass
- A sample whose cover glass is sticking out from the slide glass



10 Focus Adjustment

Precautions for 90i setup

1) Make sure the observation method is set to bright-field microscopy.

2) Make sure the objective used is

- N.A. 0.04 to 0.95
- x1 to x100.

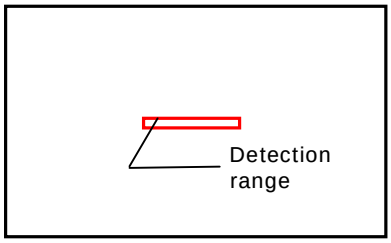
(Phase contrast, liquid immersion, polarization, Hoffman, multi-immersion, and industrial-use objectives may not be used.)

3) Make sure the objective information is set at each address on the nosepiece.

Other precautions

1) Autofocus relies on contrast at the center of the view field. Thus, the area of the target to be examined will not come into focus unless it is at the center of the view field. The microscope system may have difficulty focusing accurately on samples like the ones indicated below.

- Low contrast samples
- Samples densely dyed (dark) over the entire view field
- Samples prepared for methods other than bright-field microscopy
- Samples that lack light and dark patterns running horizontally along the screen (lateral direction in the diagram shown at right)



2) Since autofocus responds to the highest contrast part of the sample, the focused position of the sample may not always be the area you want to observe. In such cases, adjust focus visually by turning the focus knob of your microscope.

3) The autofocus may occasionally misfocus due to external disturbances. If the image fails to come into focus, try autofocusing again.

4) The microscope system may operate erratically if dirt or dust accumulates in the following areas.

- Top or bottom faces of the slide glass
- Tip of the objective
- Surface of the condenser
- In the middle of the optical path

11 Diopter Adjustment

Diopter adjustment compensates for differences in visual acuity between the right and left eyes, improving binocular observation. It also minimizes focal deviations when switching objectives. Adjust diopter settings for both eyepieces.

(1) Turn the diopter adjustment ring of each eyepiece and align the end face of the diopter adjustment ring with the line. (This is the diopter adjustment reference position.)

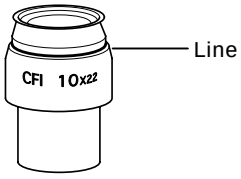
(2) Perform steps 1) to 10) in “Bright-Field Microscopy” in the "Microscopy" instruction manual to focus on the specimen with the 10x objective.

(3) Set the 40x objective in the optical path. Using the focus knobs, focus on the specimen.

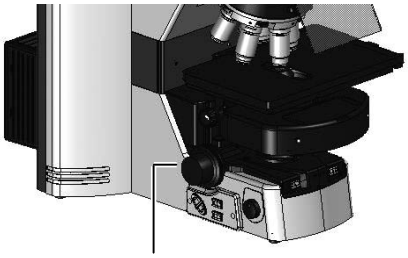
(4) Set the 4x or 10x objective in the optical path.

(5) Focus on the specimen using the diopter adjustment rings instead of the coarse/fine focus knobs. When making focus adjustments, be sure to look through the right eyepiece with your right eye and the left eyepiece with your left eye.

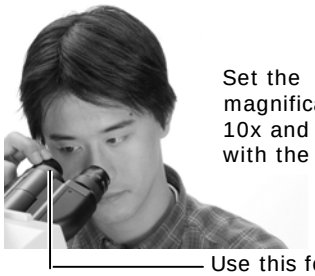
(6) Perform steps (3) through (5) twice.



Reference position for diopter adjustment



Set the 40x objective in the optical path.
Use this for focal adjustment.



Set the magnification to 10x and observe with the right eye.

Use this for focal adjustment.

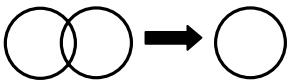
Observe with the left eye.

Use this for focal adjustment.

12 Interpupillary Adjustment

Interpupillary adjustment improves the ease of binocular observation.

Perform steps 1) to 10) in “Bright-Field Microscopy” in the "Microscopy" instruction manual and focus on the specimen using the 10x objective. Then, move the eyepiece sleeve until the fields of view for the right and left eyes coincide.

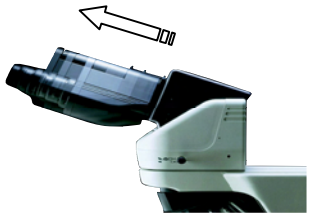


Converge until the right and left fields of view coincide.

Field of view

13 Adjusting the Observation Position

The ergonomic binocular tube makes it possible to extend and tilt the binocular section. Adjust the position of the binocular section for the most comfortable viewing.



14 Adjusting the Condenser Position

Adjust the condenser position (focusing and centering) so that the light passing through the condenser forms an image at the correct location (center of the optical path) on the specimen surface.

(1) Perform steps 1) to 10) in “Bright-Field Microscopy” in the "Microscopy" instruction manual to focus on the specimen using the 10x objective.

(2) Stop down the field diaphragm to the minimum setting.

Turn the field diaphragm knob and stop down the field diaphragm to its minimum setting.

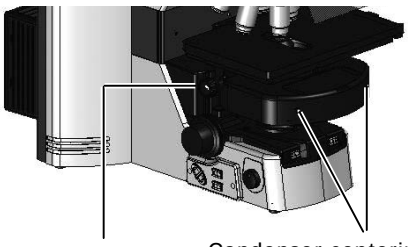
(3) Turn the condenser focus knob to form the field diaphragm image on the specimen surface.

(4) Turn the condenser centering screws so that the field diaphragm image is positioned in the center of the field of view.

(5) Set the 40x objective in the optical path. Turn the coarse/fine focus knobs and focus on the specimen.

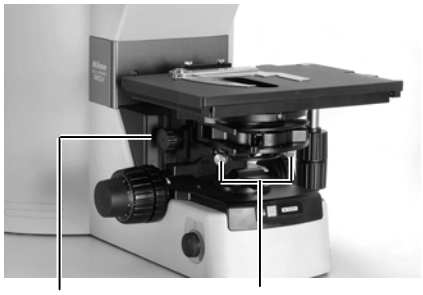
(6) Turn the condenser focus knob to form the field diaphragm image on the specimen surface.

(7) Adjust the condenser centering screws until the field diaphragm is at the center of the eyepiece field of view. This is easiest if you set the field diaphragm aperture to slightly smaller than the eyepiece field of view.



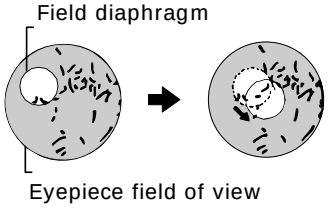
Condenser focus knob

Condenser centering screws



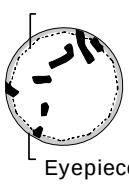
Condenser focus knob

Condenser centering screws (When using a condenser other than the motorized universal condenser)



Field diaphragm

Eyepiece field of view



Field diaphragm

Eyepiece field of view

15 Adjusting the Aperture Diaphragm

The setting of the aperture diaphragm affects optical image resolution, contrast, depth of focus, and brightness. Turning the condenser aperture diaphragm ring (or aperture diaphragm lever) changes the size of the aperture diaphragm. In the case of the motorized universal condenser, use the aperture diaphragm open/close switch.

A small aperture diaphragm opening reduces resolution and brightness but increases contrast and depth of focus. A large aperture diaphragm size increases resolution and brightness but reduces contrast and depth of focus. These characteristics involve inherent tradeoffs and cannot be optimized independently. Generally, aperture settings that are 70 to 80% of the numerical aperture of the objective will provide satisfactory images with suitable contrast.

Since an excessively small aperture diaphragm opening will degrade image resolution, we do not recommend setting the aperture diaphragm to less than 60% of the objective's numerical aperture.

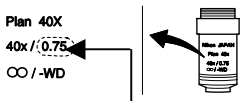
For use of the DS-L1 for operation control, refer to the instruction manual provided with the DS-L1.

For use of a PC for operation control, refer to the instruction manual provided with the software.

Aperture diaphragm open/close switch

Aperture diaphragm lever (When using a condenser other than the motorized universal condenser)

The numerical aperture is indicated on the side of the objective.



Condenser aperture diaphragm scale= 0.75×0.7～0.8 = 0.525～0.6

15.1 Adjusting the aperture diaphragm opening using the condenser scale

Since the condenser scale indicates the numerical aperture, adjust the aperture diaphragm ring according to the scale.
(Normally, the index on the aperture diaphragm ring should align with a scale line that corresponds to 70 to 80% of the numerical aperture of the objective.)

15.2 Adjusting the aperture diaphragm opening using the centering telescope (optional)

Remove one eyepiece and attach the centering telescope (optional) in place using the optional adapter. Turn the aperture diaphragm ring to stop down to the minimum aperture. While holding down the flange of the centering telescope, turn the eyepiece of the centering telescope and focus on the aperture diaphragm.

Turn the aperture diaphragm ring to adjust the aperture. (Normally, the aperture diaphragm should be adjusted to around 70 to 80% of the size of the field of view.)

After the adjustment, remove the centering telescope and adapter and reattach the eyepiece.

16 Selecting a Condenser

Objective magnification	Condenser (◉: Optimum, ○: Suitable, x: Not suitable)			
	Achromatic/aplanat condenser	Achromat condenser	Abbe condenser	1-100x condenser
1x	x	x	x	○Note 2
2x	x	x	x	◉ Note 2
4x	x	○Note 1	○Note 1	
10x to 100x	◉	○	○	◉

Note 1: The entire field of view may not be covered if a UW eyepiece is attached.
Note 2: Swing out the top lens before use.

Depending on the type of objective, the indicated numerical aperture of the objective may not be achieved.

For example, when an objective with an N.A. of 1.4 is used, the maximum aperture of the Abbe condenser will be only about 65% of the objective's N.A., even when the condenser aperture diaphragm is wide open.

Refer to the condenser operating manual for information on universal condensers.

17 Adjusting the Field Diaphragm

The field diaphragm controls the amount of illumination falling on the area of the specimen being viewed. Operating the field diaphragm open/close switch changes the size of the field diaphragm. For normal observations, the size of the diaphragm should be slightly wider than the boundary of the field of view. If light is irradiated over a larger area than necessary, stray light can enter and degrade the contrast of the optical image.
Appropriate field diaphragm settings are particularly important for photomicrography and digital image capturing. In general, good results will be obtained by stopping down the field diaphragm to settings slightly wider than the area to be reproduced within the photo frame or monitor display.

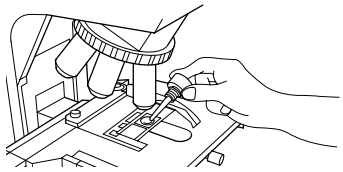
For use of the DS-L1 for operation control, refer to the instruction manual provided with the DS-L1.

For use of a PC for operation control, refer to the instruction manual provided with the software.

Field diaphragm open/close switch

18 Oil Immersion Operation

Objectives marked “Oil” are oil-immersion objectives. Objectives of this type are used with immersion oil applied between the specimen and the tip of the objective.



For maximum performance, oil-immersion objectives with numerical apertures of 1.0 or higher should be combined with oil-immersion achromatic/aplanat condensers. Oil-immersion condensers are used by applying oil between the specimen and the condenser.

Any bubbles in the immersion oil will degrade image quality. Be careful to prevent bubbles from forming. To check for air bubbles, fully open the field diaphragm and aperture diaphragm, remove the eyepiece, and examine the exit pupil (bright round section) of the objective inside the eyepiece tube. If it is difficult to ascertain the presence of bubbles, attach a centering telescope (optional), then look for air bubbles while turning the eyepiece section of the centering telescope to adjust focus. If you detect bubbles, remove them by one of the following methods:

- Turn the revolving nosepiece slightly to move the oil-immersed objective back and forth once or twice. (In the case of the condenser, gently turn the condenser focus knob to move the condenser up and down slightly.)
- Add more oil.
- Remove the oil and apply new oil.

Use as little oil as possible (just enough to fill the space between the tip of the objective and the specimen, or between the tip of the condenser and the specimen). Too much oil will result in excess oil flowing onto the stage and around the condenser.

Any oil remaining on the oil-immersion objective or adhering to the dry-type objective will noticeably degrade image quality. After use, thoroughly wipe off all oil, and make sure that no oil remains on the tips of other objectives. Additionally, carefully wipe off oil from the condenser.

Use petroleum benzine to wipe off immersion oil. For optimum results, we recommend following up petroleum benzine with absolute alcohol (ethyl or methyl alcohol).

If petroleum benzine is unavailable, use methyl alcohol alone. When using just methyl alcohol, note that surfaces will need to be wiped repeatedly to ensure complete removal of immersion oil. Usually, three or four times should be sufficient to clean the lens.

CAUTION

When using petroleum benzine or absolute alcohol, always follow the instructions provided by the manufacturer. These liquids are highly flammable and must be kept away from flames and sparks.

19 Water Immersion

Objectives marked “WI” or “W” are water-immersion objectives. These objectives are used with immersion water (distilled water or physiological saline) applied between the specimen and the tip of the objective. Microscopy procedures are the same as for oil-immersion objectives.

Since water evaporates readily, monitor the immersion water during observation. Avoid using too much water, since excess water will flow onto the stage and around the condenser, promoting corrosion.

After use, wipe off water from the tip of the objective and condenser, then follow up by wiping with absolute alcohol.

If you observe water stains, apply a small amount of neutral detergent and wipe gently, then follow up with absolute alcohol.

20 Fluorescence Observation

20.1 Warning

The mercury lamp (or xenon lamp) used with the DIH-M or DIH-E requires careful handling. Be sure to read the warnings described in the beginning of this manual and in the operating manual provided by the manufacturers of the super high-pressure mercury lamp power supply (or high-intensity light source) and lamp. Observe all the warnings and precautions described in those documents.

20.2 Shutter

The shutter blocks illumination. When suspending microscopy, close the shutter to prevent fading of specimen colors. (Set the shutter lever to the C position to move the shutter into the optical path and block light.) To protect important specimens, make it a habit to use the shutter whenever appropriate.

When pausing Epi-fluorescent microscopy to perform microscopy with diascopic light, move the shutter into the optical path to block the Epi-illumination.

Each time the shutter switch is pressed, the shutter closes or opens. The shutter switch lamp lights when the shutter is in the optical path.

For use of the DS-L1 for operation control, refer to the instruction manual provided with the DS-L1.

For use of a PC for operation control, refer to the instruction manual provided with the software.

Shutter switch

20.3 Light shield

The light shield protects the observer's eyes from ultraviolet light reflected from the specimen. To remove the plate, loosen the clamp screw and pull it forward.

Light shield

20.4 Field diaphragm for Epi-illumination

The field diaphragm is used to restrict the illumination to the area of the specimen being viewed. Operating the field diaphragm lever changes the size of the field diaphragm.

Position of field diaphragm open/close lever	Field diaphragm setting
Pushed in	Open
Pulled	Constricted

For normal observations, stop down the diaphragm so that the aperture boundaries are just outside (or inside) the field of view. Illuminating an area broader than necessary will result in stray light entering the field of view, generating flare, reducing image contrast, and expanding the area of the specimen subject to fading.

Appropriate field diaphragm settings are particularly important for photomicrography and digital image capturing. In general, good results will be obtained by stopping down the field diaphragm to slightly wider than the area to be reproduced within the photo frame or monitor display. In the case of the DIH-E, the size is automatically adjusted to be just outside the field of view.

Adjust the field diaphragm of the DIH-M or DIH-E after performing the following centering procedures:

For use of the DS-L1 for operation control, refer to the instruction manual provided with the DS-L1.

For use of a PC for operation control, refer to the instruction manual provided with the software.

Centering the field diaphragm

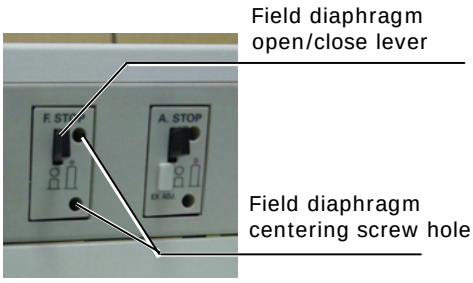
(1) Adjust the diopter and interpupillary distance according to the procedures described in "Epi-fluorescence Microscopy" in the separate Microscopy manual.

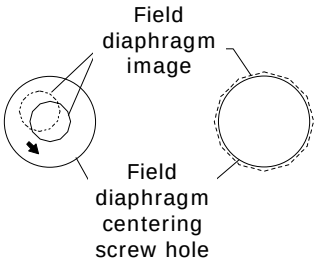
(2) Stop down the field diaphragm. (Pull the field diaphragm open/close lever.)

(3) Move the center of the field diaphragm image to the center of the field of view. (Using the hex driver provided with the microscope, turn the field diaphragm centering screws.)

(4) Open up the field diaphragm to match the field of view. (Push in the field diaphragm open/close lever.)

(5) Again, move the center of the field diaphragm image to the center of the field of view. (Turn the field diaphragm centering screws.)





20.5 Aperture diaphragm for Epi-illumination

The aperture diaphragm determines the numerical aperture for illumination and the optical system. In Epi-fluorescence microscopy, it is used to adjust image brightness. Use the aperture diaphragm open/close lever to adjust the size of the opening of the aperture diaphragm.

Position of aperture diaphragm open/close lever	Size of aperture diaphragm
Pushed in	Open
Pulled	Constricted

A narrow aperture diaphragm setting reduces the amount of backlighting and makes the image darker. In contrast, opening up the aperture diaphragm increases backlighting and image brightness. For ordinary Epi-fluorescence microscopic observations, keep the aperture diaphragm open. Stop down the diaphragm only when image brightness needs to be adjusted.

Use the aperture diaphragm of the DIH-M or DIH-E after performing centering. (The centering tool may not be included in the set you have purchased.)

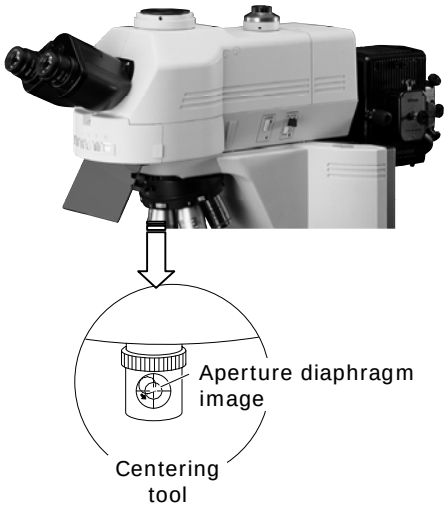
Centering the aperture diaphragm

(1) Close the shutter for the epifluorescent light. (Make sure that the shutter switch lamp is lit.)

(2) Remove one objective from the revolving nosepiece and insert the centering tool in its place.

(3) Move the centering tool into the optical path.

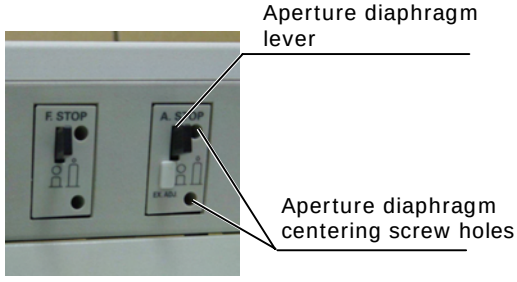
(4) Turn on the Epi-illumination light source and open the shutter.



(5) Stop down the aperture diaphragm. (Pull the aperture diaphragm lever.)

(6) While observing the centering tool screen, move the center of the aperture diaphragm image to the center of the screen. (Using the hex driver provided with the microscope, turn the field diaphragm centering screws.) Be sure to view the centering tool screen squarely, from the front.

(7) After centering is completed, close the shutter, remove the centering tool, and reinstall the objective.



20.6 Switching excitation methods

Up to six filter cubes each can be inserted to the DIH-M and to the DIH-E.

For the DIH-M, rotate the excitation method switchover turret on the front of the equipment to move the desired cube into the optical path.

In the case of the DIH-E, operate the excitation method switch and insert a desired filter cube into the optical path.

The number on the front section of the turret indicates the filter cube set into the optical path.

For use of the DS-L1 for operation control, refer to the instruction manual provided with the DS-L1.

For use of a PC for operation control, refer to the instruction manual provided with the software.

Excitation method switchover turret

Number of the fluorescent cube set into the optical path

Excitation method switch

20.7 ND filters for Epi-illumination

Pushing in an ND filter slider sets the respective ND filter into the optical path and darkens the fluorescent image.

ND filters reduce light intensity without altering the color balance of the illumination. Higher filter numbers correspond to lower transmission rates (i.e., darker images). If fluorescence is too strong or if specimen colors fade too rapidly, insert ND filters into the optical path to adjust brightness. Excessively strong fluorescence may result in poor image contrast.

ND4: Reduces light intensity to 1/4

ND8: Reduces light intensity to 1/8.

ND16: Reduces light intensity to 1/16.

You can combine these three filters to achieve various levels of brightness.

DIH-M or DIH-E

Brightness	ND4	ND8	ND16
1	-	-	-
1/4	○	-	-
1/8	-	○	-
1/16	-	-	○
1/32	○	○	-
1/64	○	-	○
1/128	-	○	○
1/512	○	○	○

(-: Outside optical path, ○: In optical path)

20.8 Prohibiting the automatic adjustment of the episcopic field diaphragm in switching of optical paths

When the DIH-E is used, the episcopic field diaphragm size is automatically adjusted to be just outside the field of view when switching optical paths. This automatic adjustment can be disabled (turned Off). It is recommended to keep the factory setting in normal use.

[Setting method]]

The setting can be changed by using iSetup of the i Series Support Tools software.

For details, refer to "5.10.2 Objective, Optical Path Switching, and Optical Zoom Interlock Setups" in the i Series Support Tools software manual.

[Factory setting]

ON

20.9 Prohibiting the automatic adjustment of the episcopic field diaphragm when switching zooms

When the DIH-E is used and the rear port optical path is selected, the episcopic field diaphragm size is automatically adjusted to be just outside the field of view when switching optical zooms. This automatic adjustment can be disabled (turned OFF).

The factory setting is recommended for normal use.

[Setting method]

The setting can be changed by using iSetup of the i Series Support Tools software.

For details, refer to "5.10.2 Objective, Optical Path Switching, and Optical Zoom Interlock Setups" in the i Series Support Tools software manual.

[Factory setting]

ON

20.10 Changing the size of the episcopic field diaphragm when switching zooms

When the rear port of the DIH-E is used, the episcopic field diaphragm is automatically adjusted to an optimum size when switching zooms according to the size of the field of view. The size of the episcopic field diaphragm in this adjustment can be changed in the direction of increased stop-down.

[Setting method]

The setting can be changed by using iSetup of the i Series Support Tools software.

For details, refer to "5.10.3 Compensation Setup during Interlocking" in the i Series Support Tools software manual.

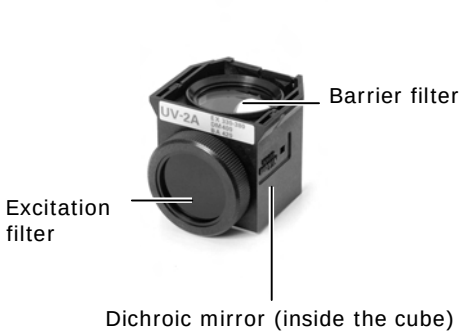
[Factory setting]

1.00
(A value of "1.00" is given to the size of an episcopic field diaphragm image that is just outside the field of view. For adjustment, specify the rate in relation to this maximum value. Note that only values less than 1.00 can be set.)

21 Selecting Fluorescent Filters

A filter cube is comprised of the following three optical components: an excitation filter (EX filter), a barrier filter (BA filter), and a dichroic mirror (DM). Select a filter cube with the appropriate combination of optical components for the specimen characteristics, fluorescent pigment, and the purpose intended. Keep in mind the following:

- Different combinations of excitation and barrier filters may be selected for the same excitation method.
- Other types of excitation filters, barrier filters, and dichroic mirrors can be purchased separately.
- Excitation filters are exposed to strong light during operations and tend to age rapidly. Replace the excitation filter at frequent intervals.

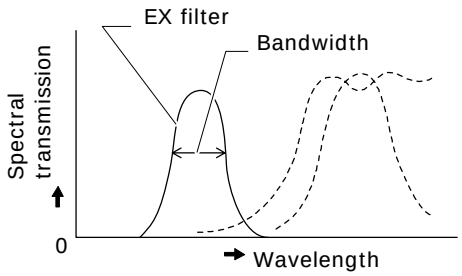


21.1 Selecting excitation filters (EX filters)

Excitation filters allow selective transmission of light (excitation light) in the waveband required for fluorescent light emissions from the specimen, blocking light of all other wavelengths. The range of wavelengths allowed to pass through a filter is referred to as bandwidth.

The range of the bandwidth of the excitation filter determines the brightness of the fluorescent image, the generation of self-fluorescence (fluorescence resulting from substances other than fluorescent pigments), and degree of fading. The broader the bandwidth, the greater the amount of excitation light irradiated onto the specimen, increasing brightness. However, this also increases the amount of self-fluorescence and causes faster color fading. Narrow bandwidth reduces the amount of excitation light striking the specimen and causes the image to appear darker, but reduces self-fluorescence and fading. For specimens with pronounced self-fluorescence, use excitation filters with a narrow bandwidth (note that this will make the fluorescent image darker).

Excitation filters are exposed to strong light during operations and tend to age rapidly. Replace the filter at intervals determined by usage.



	Narrow	EX filter bandwidth	Wide
Brightness of fluorescent image	Dark		Bright
Generation of self-fluorescence	Low		High
Degree of color fading	Low		High

21.2 Selection of barrier filter (BA filter)

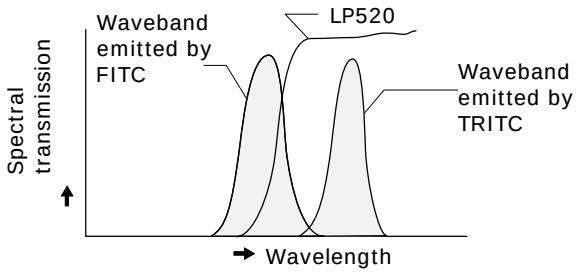
The barrier filter allows only fluorescent light emitted by the specimen to pass, blocking excitation light. This allows viewing of a fluorescent image without excess illumination (dark background).
There are two types of barrier filters: LP filters block all light below a certain wavelength but pass all light of longer wavelengths. BP filters pass only light of a certain waveband, blocking all other light. Use the filter type appropriate for your intended purpose.

LP filter (long-pass filter)

LP filters block all light below a certain wavelength but pass all light of longer wavelengths. This border wavelength is called a cut-on wavelength.

(1) For specimens dyed with a fluorescent pigment in which the fluorescent waveband and excitation waveband (light that the specimen absorbs in order to emit fluorescent light) are very close, select a barrier filter with the shortest cut-on wavelength permitted by performance requirements for efficient fluorescent microscopy.
If the cut-on wavelength is long, excitation light and fluorescent light will be entirely distinct, tending to darken the background of fluorescent images. However, recent advances in filter performance have resulted in increased use of filters of short cut-on wavelengths.

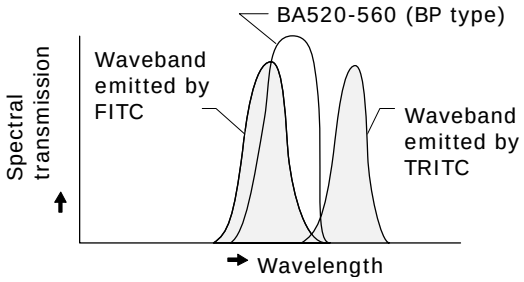
(2) For multiple-dye specimens, use an LP filter for microscopy of fluorescent images of all dyes.
Note that a combination involving an ordinary dichroic mirror, an excitation filter, and an LP-filter-type barrier filter will be incapable of exciting dyes that emit long-wavelength fluorescent light – for example, TRITC in the case of FITC and TRITC. This will result in very dark TRITC fluorescent images. For such cases, we recommend using multiband filters.



Both the FITC and TRITC images are visible.

BP filter (bandpass filter)

The bandpass filter passes only light of a certain wavelength, blocking all other wavelengths.
BP filters are used for microscopy of fluorescent images involving a specific dye in multiple-dye specimens. (For example, in a double-dye specimen of FITC and TRITC, the BA520-560 filter enables microscopy of just the FITC fluorescent image.)
However, BP filters will not indicate self-fluorescence, if any (because the fluorescent image in the above combination is green only). LP filters are better suited for making fine distinctions in self-fluorescence based on slight color differences.



Only the FITC fluorescent image is visible.

21.3 Replacing excitation and barrier filters

Excitation and barrier filters can be removed from the filter cube and replaced. (Dichroic mirrors cannot be dismounted from the filter cube.)
Excitation filters are screw-in filters.

Barrier filters are slide-in filters. Align the projection on the barrier filter with the groove on the filter cube and turn clockwise approximately 30 degrees to secure in place.

22 Excitation Light Balancer

Attach the optional D-FB excitation light balancer to the DIH-M or DIH-E to adjust the wavelength characteristic of the excitation light. The excitation light balancer is used in combination with a dual-band filter cube.

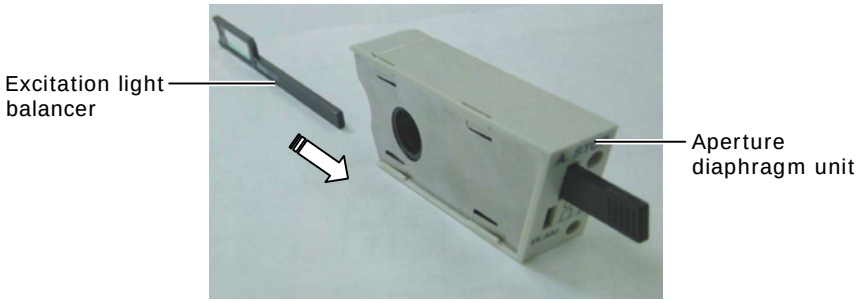
Required products

- D-FB excitation light balancer
- Dual-band filter cube

(1) Switch off the light source for the DIH-M or DIH-E.

(2) Detach the aperture diaphragm unit from the DIH-M or DIH-E.

(3) Attach the D-FB excitation light balancer from the back end of the aperture diaphragm unit to fix it.



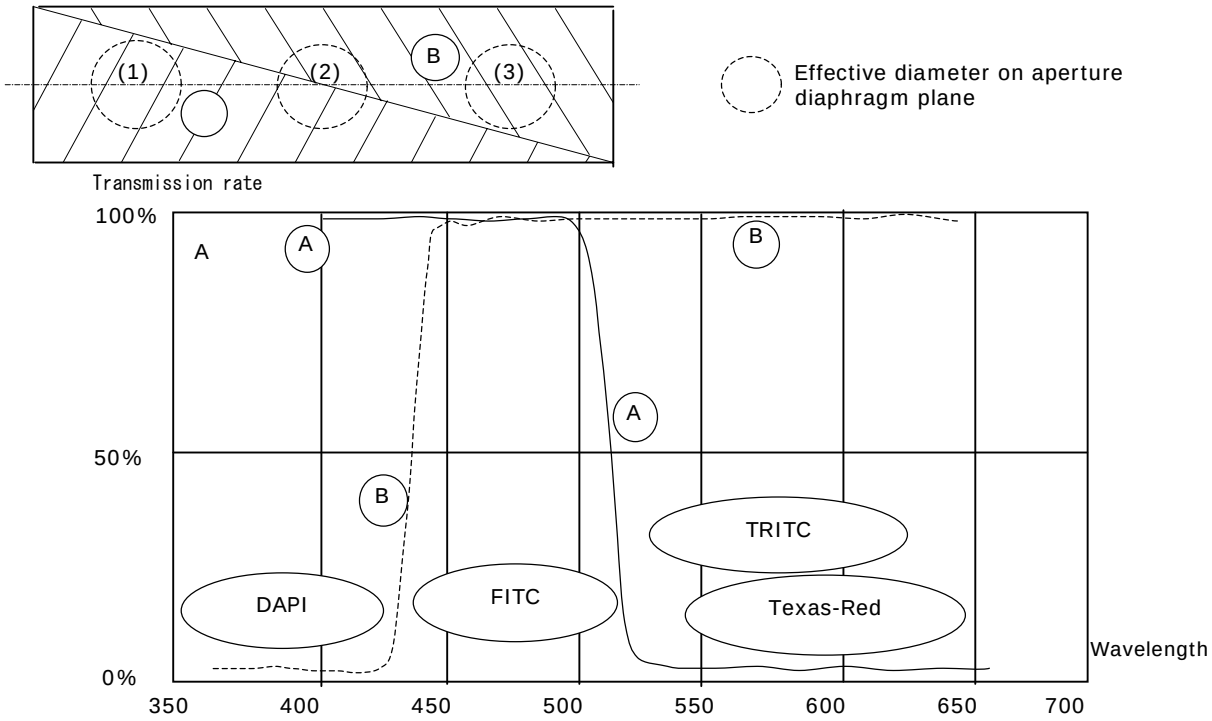
- Take care not to touch the glass surface of the excitation light with your bare hands or fingers.

(4) Reattach the aperture diaphragm unit to the DIH-M or DIH-E.

(5) Mount a dual-band filter cube to the DIH-M or DIH-E.

(6) Move the excitation light balancer laterally to adjust the excitation light.

Detailed view of excitation light balancer



The excitation light balancer is set so that the transmission rate remains at about 100% for typical dark fluorescent FITC.

Position of optical path	DAPI	FITC	TRITC/Texas-Red
(1)	100%	100%	0%
Between (1) and (2)	Variable (100% - 50%)	100%	Variable (0% - 50%)
(2)	50%	100%	50%
Between (2) and (3)	Variable (50% - 0%)	100%	Variable (50% - 100%)
(3)	0%	100%	100%

About objectives

Lamp voltage: If accurate color reproduction is critical, push in the preset switch on the microscope, set the NCB11 filter into the optical path, and use ND filters to make brightness adjustments.

Plan Fluor	40x/0.75	40xH/1.3	100xH/1.3
S Fluor	40x/0.9	40xH/1.3	100xH/1.3
Plan Apo	40x/0.95	60xH/1.4	100xH/1.4

23 Image Capture

Image can be captured by attaching a camera head to the ergonomic binocular tube, trinocular eyepiece tube, DIH-M, or DIH-E.

For more detailed discussion of this topic, refer to the operating manual provided with the camera head or camera control software.

Proper adjustment of light intensity and focus on the microscope side are important for obtaining clear images. Listed below are key considerations in capturing clear images.

For use of the DS-L1 for operation control, refer to the instruction manual provided with the DS-L1.

For use of a PC for operation control, refer to the instruction manual provided with the software.

23.1 Adjusting light intensity

Lamp voltage: When color reproducibility is a high priority, press the preset switch of the microscope to turn on the switch lamp, insert the NCB11 filter into the optical path, and adjust the brightness using the ND filters.

Filter: Place a commercially available color compensation filter on the filter holder at the microscope base, as necessary.

23.2 Adjusting the condenser

- Always focus and center the condenser.
- Center the annular diaphragm for phase contrast microscopy.
- For normal operations, set the diaphragm aperture to 70 to 80% of the N.A. of the objective.

23.3 Confirming the photomicrographic range

The image on the monitor represents the photomicrographic range.

23.4 Confirming focus

Check focus by viewing through the eyepiece and viewing the monitor. If the focal positions for the two images differ, adjust the focal position adjustment screw at the camera port. (When using the ergonomic binocular tube)

23.5 Making adjustments to keep out extraneous light

Field diaphragm: Stop down the diaphragm to a setting just slightly wider than the area shown on the monitor.

Eyepiece section : Mount the binocular cap (sold separately) on the eyepiece section.

23.6 Fluorescence photomicrography

The fluorescence of fluorescent specimens may fade during exposure. To prevent this, do the following:

(1) Select a brighter optical combination.
Even if the overall magnification is the same on the monitor, the combination of objective and camera zoom can result in significant variations in exposure time. We recommend increasing the magnification with the objective rather than the zoom. (Generally, the aperture of the objective increases with magnification. The larger the numerical aperture, the brighter the resulting image.)

(2) Adjusting the excitation light
Excessively bright excitation light will accelerate specimen fading while making it more difficult to acquire suitable fluorescent images. Use ND filters to adjust brightness.

(3) Specimen
Photomicrography of faded specimen sections requires prolonged exposure times and results in poor color reproduction and low-quality images. Move the specimen to obtain images from a fresh section of the specimen previously unexposed to excitation light. For best results, use the differential interference contrast or phase contrast methods to select a specimen section for photomicrography, and then switch to the fluorescent method to capture images.

24 Switching of Microscopy Methods

24.1 Switching microscopy methods

When the 90i is combined with the motorized universal condenser and the DIH-E, microscopy methods can be switched easily by using the DS-L1 or iControl of the i Series Support Tools software. When microscopy methods are switched, the motorized attachments automatically adjust to optimal conditions according to the selected microscopy method, as shown in the table below.

Table Automatic adjustments when switching microscopy methods

	90i			Motorized universal condenser		Motorized nosepiece	DIH-E						Motorized ND filter
	Lamp	Field diaphragm	Vertical stage motion	Turret	Aperture diaphragm		Excitation method switchover turret	Episcopic field diaphragm	Shutter	Analyzer	Optical path changeover	Zoom	
Bright field	ON	*1	-	*2	*3	-	Turret address 6	-	CLOSE	OUT	-	-	-
DIC	ON	*1	-	*2	*3	-	Turret address 6	-	CLOSE	IN	-	-	-
Simul-taneous DIC/FL	ON	*1	-	*2	*3	-	Turret address 1	-	OPEN	IN	-	-	-
FL	OFF	-	-	*2	-	-	Turret address 1	-	OPEN	OUT	-	-	-
Ph	ON	*1	-	*2	Fully open	-	Turret address 6	-	CLOSE	OUT	-	-	-
Dark field	ON	Fully open	-	*2	Fully open	-	Turret address 6	-	CLOSE	OUT	-	-	-

[How to read the table]

- : No operation

*1 : Adjusts the field diaphragm size to be just outside the field of view according to the objective set in the optical path.

*2 : The most suitable condenser module is inserted into the optical path according to the selected microscopy method and the objective set in the optical path.

*3 : Adjusts the aperture diaphragm size to achieve the optimum image contrast according to the objective set in the optical path.

[Supplementary information]

The automatic adjustments performed when switching microscopy methods are not affected by disabling of the adjustment of the field diaphragm and motorized universal condenser (turret, aperture diaphragm) described in "8.2 Canceling the automatic adjustment of motorized attachments in switching of objectives."

[Operating method]

The operation can be performed by using the DS-L1 or iControl of the i Series Support Tools software.

For details, refer to the manuals provided with the respective products.

24.2 Setting a desired microscopy method

In addition to the microscopy methods described in "24.1 Switching microscopy methods," up to six desired microscopy methods can be registered.

[Setting method]

A desired microscopy method can be registered using iControl of the i Series Support Tools software.

For details, refer to "4.5.2 Setting Up Arbitrary Microscopy Method (Setting Up the Profile)" in i Series Support Tools software manual.

[Factory setting]

Not registered

24.3 Canceling the automatic adjustments of motorized attachments

When microscopy methods are switched, the motorized attachments automatically perform adjustments. These automatic adjustment operations can disabled (turned OFF) for individual motorized attachments. The factory settings are recommended for normal use.

[Setting method]

For this setting, use iSetup of the i Series Support Tools software.

For details, refer to "5.10.1 Microscopy Method Interlock Setup" in the i Series Support Tools software manual.

[Factory setting]

ON

25 Switches

25.1 Changing Enable/Disable setting of switches

The switches of the 90i can be enabled or disabled.

[Setting method]

For this setting, use iSetup of the i Series Support Tools software.

For details, refer to "5.10 Control-Related Setup" in the i Series Support Tools software manual.

[Factory setting]

ON (Enabled)

26 Buzzer

26.1 Turning buzzer ON/OFF

The buzzer that is activated when switches are operated can be turned ON or OFF.

[Setting method]

For this setting, use iSetup of the i Series Support Tools software.

For details, refer to "5.10 Control-Related Setup" in the i Series Support Tools software manual.

[Factory setting]

ON