M568 E 11.8.NF.1 (1/2)
*M568EN01*

Nikon

# Upright Microscope

ECLIPSE

Ci-S / Ci-L

# Instruction Manual

# Introduction

Thank you for purchasing a Nikon product.

This instruction manual is written for users of the Nikon ECLIPSE Ci-S and Ci-L microscopes. To ensure correct usage, read this manual carefully before operating this product.

- No part of this manual may be reproduced or transmitted in any form without prior written permission from Nikon.
- The contents of this manual are subject to change without notice.
- Appearance of the product shown in this manual may be different from that of your product.
- 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 equipment described in this manual may not be included in the set you have purchased.
- If you intend to use any other equipment with this product, read the manual for that equipment too.
- If this equipment is used in a manner not specified by the manufacturer, the protection provided by the equipment may be impaired.
- Training: This product can be used without special training, provided that this manual is read thoroughly before use. Kindly contact your nearest Nikon representative if you have any questions, find any errors, or wish to provide us with your opinion.

## Contents of the Manual

The manual for ECLIPSE Ci-S/Ci-L consists of the following contents. There is also a "Bright-field Microscopy Quick Guide", provided as a spearate document.

◆ **This manual: Instructions**

- Safety Precautions
- Microscopy Procedures
  - Bright-field Microscopy
  - Phase Contrast Microscopy
  - Simple Polarizing Microscopy
  - Sensitive Tint Plate Microscopy
  - Epi-fluorescence Microscopy
- Individual Operations
- Assembly
- Troubleshooting
- Maintenance and Storage
- Specifications and Safety Standards

◆ **Bright-field Microscopy Quick Guide**

## Symbols Used in This Manual

The following symbols are used in this manual.

◆ **Symbols for Safety**

| | |
|---|---|
| ⚠ **WARNING**<br>⚠ **CAUTION** | Highlights important information that should be noted for safety. Read "Safety Precautions" for details. |

◆ **Other Symbols**

| | |
|---|---|
| ❗ | Indicates information you should note or comply with to prevent defects or malfunction of this product. |
| ✔ | Indicates information you should be aware of in using this product, as well as other useful information. |

# Table of Chapters (See next page for the detailed contents.)

# Table of Contents

# Safety Precautions

To ensure correct and safe operation, read this manual before using this product.



## WARNING and CAUTION Symbols used in This Manual

Although this product is designed and manufactured to be completely safe during use, incorrect usage or failure to follow the safety instructions provided may cause personal injury or property damage. To ensure correct usage, read this manual carefully before using this product. Do not discard this manual and keep it handy for easy reference.

Safety instructions in this manual are marked with the following symbols to indicate their importance. For your safety, always follow the instructions marked with these symbols.

| Symbol | Description |
|---|---|
| **WARNING** | Disregarding instructions marked with this symbol may lead to serious injury or death. |
| **CAUTION** | Disregarding instructions marked with this symbol may lead to injury or property damage. |

## Meaning of Symbols Used on the Product

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

| | |
|---|---|
| | **Biohazard**<br>This symbol is affixed to the front of the stand of this product, to call your attention to the following:<br>● WARNING: Using this product may constitute a biohazard risk if a sample comes into contact with this product.<br>● To avoid biohazard contamination, do not touch the contaminated portion with bare hands.<br>● Decontaminate the contaminated part according to the standard procedure specified for your laboratory. |
| | **Precautions against heat**<br>This symbol is affixed to the part near the lamphouse of the ECLIPSE Ci-S to call your attention to the following:<br>● During and immediately after a period of illumination, the lamp and surrounding areas (including the lamphouse) are very hot.<br>● Risk of burns. Do not touch the lamp or surrounding areas during or immediately after a period of illumination.<br>● Make sure the lamp and surrounding areas have cooled sufficiently before attempting to replace the lamp. |

## ⚠ WARNING

**1 Do not disassemble.**

Disassembling this product may result in electric shock or malfunction. Malfunction and damage due to disassembling or modification are unwarranted.

Do not disassemble parts other than those described in this manual. If you experience problems with this product, contact your nearest Nikon representative.

**2 Read the instruction manuals carefully.**

To ensure safety, thoroughly read this manual and the manuals for other equipment to be used with this product. Particularly, all warnings and cautions given at the beginning of each manual must be observed.

Safety is a top design priority for Nikon products. Safety is ensured as long as the user observes all of the warnings and cautions given in the manuals, and uses the system only for its intended purpose. However, failure to heed the warnings and cautions given in the manuals, subjecting the system to shock or impact, or attempting to disassemble the system may result in unexpected accidents and injury.

**Product with CI-FL epi-fluorescence attachment:**

The light source used for Epi-fluorescence microscopy (HG Precentered Fiber Illuminator) requires special care during handling because of its characteristics. Be sure to refer to the manual for the light source being used.

**3 Notes on the power cord**

Be sure to use the specified power cord. Use of other power cords may result in malfunction or fire. This product is classified as having Class I protection against electric shock. Make sure this product is connected to an appropriate protective earth terminal.

Refer to Chapter 6, “2 Performance Properties” for the specified power cords.

- To prevent electric shock, always turn off the power switch (press to the “O” position) for the microscope before connecting or disconnecting the power cord.

**4 Heat from the illuminator (when using ECLIPSE Ci-S)**

During and immediately after a period of illumination, the lamp and surrounding areas (including the lamphouse) are very hot.

- Do not touch the lamp or surrounding areas during or immediately after a period of illumination. There is a risk of burn if you touch the hot area.
- Always attach the lamphouse cover when using this product.
- Make sure the lamp and surrounding areas have cooled sufficiently before attempting to replace the lamp (about thirty minutes).
- To avoid the risk of fire, do not place fabric, paper or highly flammable volatile materials such as gasoline, petroleum benzine, paint thinner, or alcohol near the lamphouse while the lamp is lit or for a period of approximately thirty minutes after the lamp is turned off.

**5 Hazards of mercury lamps (when using the CI-FL Epi-fluorescence attachment)**

The light source used with the epi-fluorescence attachment (HG Precentered Fiber Illuminator) requires special care during handling because of its characteristics. For safe and correct use of this system, carefully read the warnings below. Keep in mind all potential hazards. Additionally, carefully read the manual for the illuminator and the manual from the lamp manufacturer (if provided), then follow the instructions given therein. Failure to heed the warnings and cautions given in the manuals, subjecting the system to shock or impact, or attempting to disassemble the system may result in unexpected accidents and injury.

- **Ultraviolet light**
  When lit, mercury lamps radiate ultraviolet light that can damage the eyes and skin. Direct viewing of the light may result in blindness.
  When changing filter cubes, always turn off the light source of the Epi-fluorescence attachment. Leaving the lamp turned on during filter cube replacement may result in ultraviolet exposure.
- **High-pressure gas**
  The lamps contain sealed gas under very high pressure. And the pressure increases when the lamp is on. If the lamp is scratched, fouled, subjected to high external pressure or physical impact, or used beyond its service life, the sealed gas may leak or the lamp may burst, resulting in gas inhalation, injury from glass, or other accidents.
- **Heat**
  When the lamp is lit, the lamp and surroundings will become extremely hot. Do not touch the lamp with bare hands or place flammable materials near the lamp. Failure to comply may result in burns or fire.
- **Designated lamp**
  Be sure to use the designated lamp. Using other types of lamps may result in accidents, including bursting of the lamp.

| WARNING | |
|---|---|
| **6 Hazardous sample handling**<br>This product is intended primarily for microscopic observations and image capture of cells and tissue set on glass slides.<br>Check to determine whether a sample is hazardous before handling. If sample is hazardous, handle it according to the standard procedure specified for your laboratory. If the sample is potentially infectious, wear rubber gloves and avoid directly touching samples. If such a sample is spilled onto this product, the portion must be decontaminated in a safety manner. Consult your safety supervisor or safety standard of your facility. | |

## ⚠ CAUTION

**1 Power shutdown**

To prevent electric shock and/or malfunction, always turn off the power switch(es) for this product and the peripheral devices (press to the “O” position) and unplug the power cord from the wall outlet before assembling this product, connecting or disconnecting cables, replacing lamps, or cleaning this microscope and the objective.

**2 Lamp replacement precautions (when using ECLIPSE Ci-S)**

- To avoid burns, wait at least 30 minutes after the lamp is turned off to give it sufficient time to cool. To avoid electric shock or malfunctions, never attempt to replace the lamp without first turning off the power switches for this product and the peripheral devices (press to the “O” position) and unplugging the power cord from the wall outlet.
- Make sure the lamphouse cover is securely fitted to the lamphouse after lamp replacement. Never turn on the lamp while the lamphouse cover is open.
- Do not break used lamps. It should be disposed of as industrial waste, in accordance with local regulations and rules.

**3 Designated lamp (when using ECLIPSE Ci-S)**

The product's built-in power source is used for lighting the halogen lamp that is a light source for dia-illumination. Halogen lamps of up to 6V-30W can be lit. Always use the specified halogen lamp. Using an unspecified lamp may cause malfunctions.

Designated lamp: 6V-30W (PHILIPS 5761)

**4 Prevent contact with water.**

Never allow water to come into contact with this product, and avoid using this product in circumstances where it may be splashed with water. Splashing water onto this product may cause a short circuit, resulting in malfunction or abnormal heating. If water is splashed onto this product, immediately turn off the power switches for this product and the peripheral devices (press to the “O” position) and remove the power cord from the receptacle. Then wipe off moisture with a piece of dry cloth or something similar. If water enters, stop the use of the product and contact your nearest Nikon representative.

**5 Do not place any object on top of the product.**

Do not place any object on top of this product.

**6 Cautions on assembling, installing, and carrying the product**

- Take care to avoid pinching your fingers or hands during product assembly and installation.
- Scratches or fouling optical components (such as lens and filters) with fingerprints, etc. will degrade microscope images. Be careful to avoid scratches or direct contact with the lens and filters when assembling.
- The main body weighs approximately 10 kg. When moving it, hold the main body by the grip on the back and the recessed portion at the front bottom side of this product.
- Remove all attachments (if mounted) from the microscope before moving the microscope.
- Do not place this product in a locker or cabinet.

**7 Cautions on use, transportation, and storage**

This product must be operated, transported, or stored in accordance with the following conditions. Installing this product in a hot, humid location may result in the formation of mold or condensation on lenses, impairing performance or causing malfunctions.

- Operating conditions:
  temperature: 0 to +40°C,
  humidity: 60% RH max. (no condensation)
- Transporting/storage conditions:
  temperature: -20 to +60°C,
  humidity: 90% RH max. (no condensation)

**8 Remove any covers from the product before switching on.**

Do not use this product while it is covered with a piece of cloth, etc., This will result in an abnormal heating or a fire hazard. Do not cover this product with a piece of cloth or similar while in use. The system temperature will rise, resulting in a malfunction.

**9 Cautions on sustained observations**

To relieve fatigue resulting from long observation sessions, limit continuous observations to one hour. Take at least 10 to 15 minutes breaks between observation sessions. Adjust the layout of other equipment used and the height of your chair.

**10 Cautions on the disposal of the product**

To avoid biohazard risks, dispose of this product as contaminated equipment in accordance with the standard procedure specified for your facility.

## Notes on Handling the Product

### 1 Handle with care

This product is a precision optical instrument and requires gentle handling. Avoid subjecting it to sudden impacts and shocks.
Even relatively minor impacts are capable of affecting the precision of the objective.

### 2 Weak electromagnetic waves

This product emits weak electromagnetic waves. So as to avoid degrading the performance of precision electronic devices, do not install this product near such devices. If TV or radio reception is affected, move the TV or radio further from this product.

### 3 Dirt on the lens

Scratches or fouling optical components (such as lens and filters) with fingerprints, etc. will degrade microscope images.
If these parts become dirty, clean them as described in Chapter 5, “2.1 Lens Cleaning”.

### 4 Dirt on the lamp (ECLIPSE Ci-S)

Never touch the lamp with bare hands. Dirt or fingerprints on the lamp will result in uneven illumination and reduce the service life of the lamp. Always wear gloves when handling lamps.

### 5 Installation location

This product is a precision instrument. Usage or storage of this product in an inappropriate environment may result in malfunction or a degradation in precision. Consider the following factors when selecting an installation location:

- Select a location free of vibration. Install this product on a level surface.
- Install this product at least 10 cm away from walls.
- Choose a location less exposed to hazards in the event of collisions, earthquakes, or other potential disasters. To keep this product from falling, use a strong rope or other means if necessary to secure it to the working desk or other heavy, stable item.
- Select a layout that allows easy removal of the power cord from the product's AC inlet in the event of an emergency.
- Do not use a desk mat or similar.
- Avoid locations exposed to direct sunlight, locations immediately under room lights, and other bright locations.
- Light from room lights just above this product may enter the objective as extraneous light. If possible, switch off the room lights directly above this product when making observations.
- Select a location with minimal dust.
- To avoid splashes, do not use this product near water.
- Make sure the ambient temperature is 0 to +40°C and humidity is 60% or less. When transporting or storing this product, the ambient temperature must be -20 to +60°C, with the humidity at 90% RH max (with no condensation). Installing this product in a hot, humid location may result in the formation of mold or condensation on lenses, impairing performance or causing malfunctions.
- Do not place this product in a locker or cabinet.

### 6 Handling of focus knob

- Never turn the focus knobs on the right and left sides of the microscope in opposite directions at the same time. Doing so may damage this product.
- Turning the coarse focus knob past its farthest point will damage this product. Never use undue force when turning the knob.

### 7 Protect the ports from dust and extraneous light (when the trinocular tube or the ergonomic tube is attached).

To keep out extraneous light and dust, always attach the supplied cap to any port not currently in use.

### 8 Handling of filters (when using the epi-fluorescence attachment)

- Excitation filters inside a filter cube are exposed to strong light and degrade over time. Replace them after the appropriate number of hours.
- Filter characteristics may alter if the filter is exposed to high humidity. To prevent changes or degradation of filter characteristics, avoid using or storing the filters under conditions of high humidity or high temperature. Avoid subjecting filters to rapid temperature changes. When a filter is not in use, store in a desiccator or hermetically sealed container with a drying agent.
- Especially the filters in the nine types of filter cubes listed below offer sharp, high-resolution waveform characteristics superior to normal filters. However, due to their sophisticated coatings, they must be handled with special care. Take care to avoid abrasion from cleaning. Follow the description in “2.1 Lens Cleaning” in Chapter 5.

  Single-band filter cubes:
  DAPI, FITC, TxRed, GFP

  Multi-band filter cubes:
  F-R, F-T, D-F, D-F-R, D-F-T

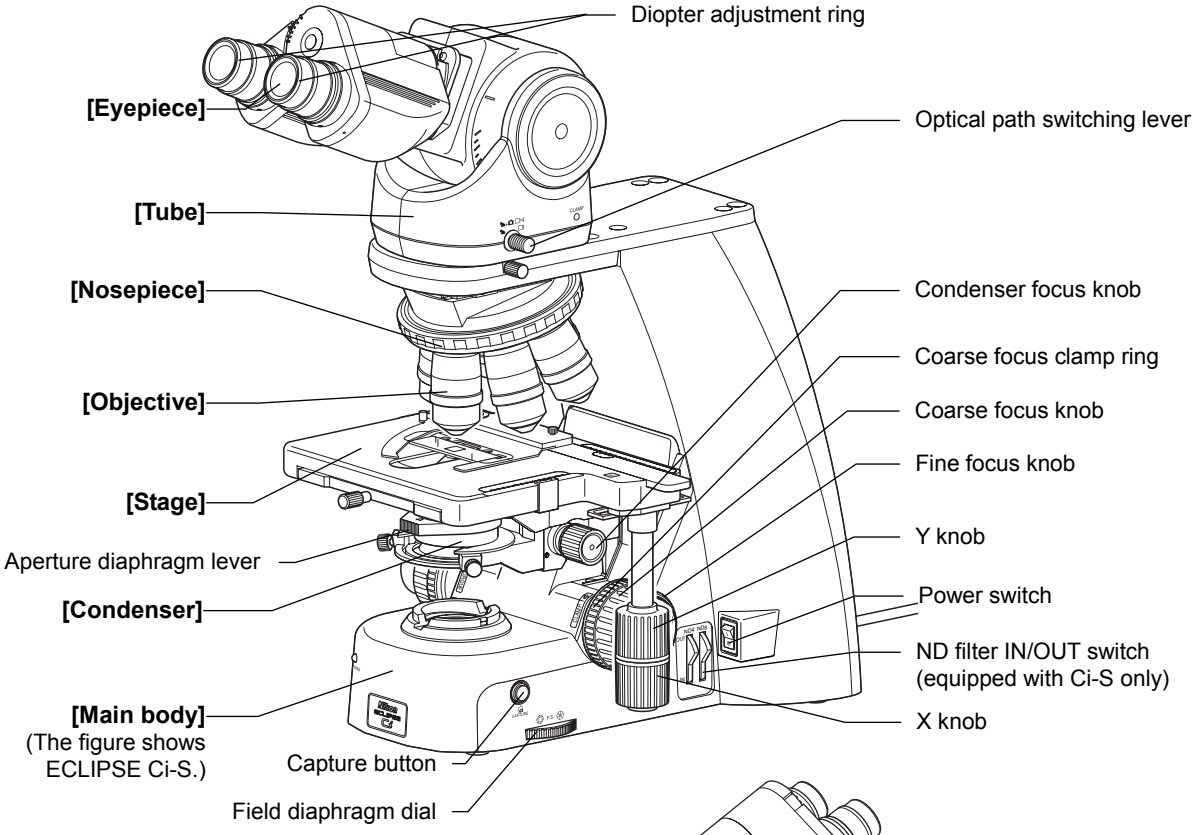
# 1 Microscopy Procedures

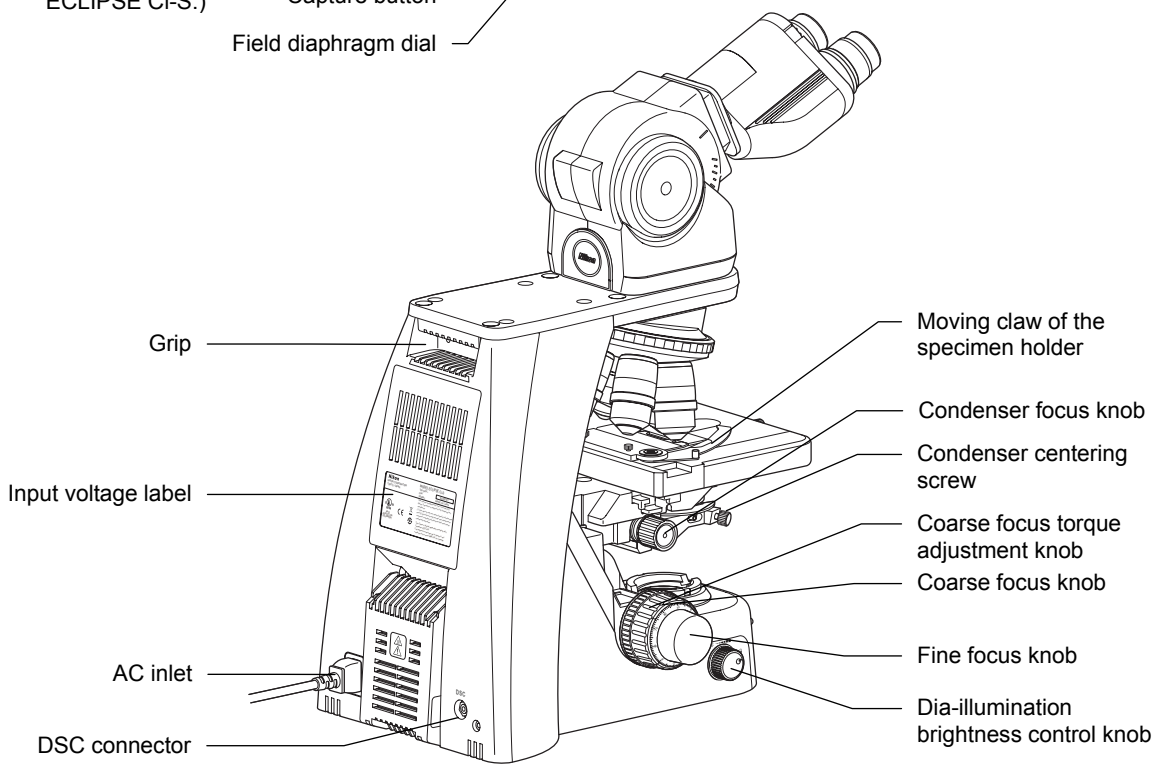
## 1 Bright-field Microscopy

### 1.1 System Configuration and Controls

This section explains an example system configuration and the controls required for bright-field microscopy using the ECLIPSE Ci-S/Ci-L.
Names of components are denoted in the following manner: **[Eyepiece]**.



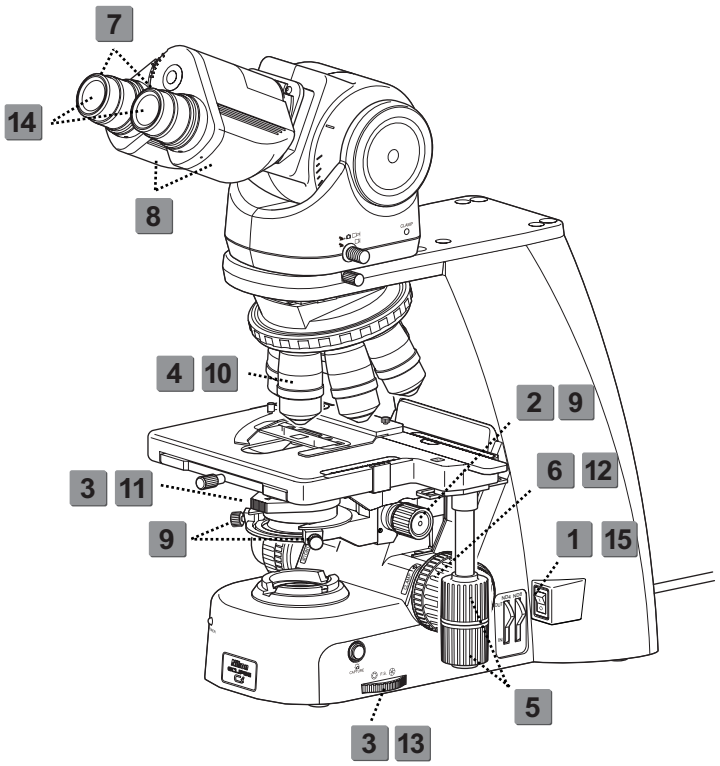


## 1.2 Bright-Field Microscopy Procedure



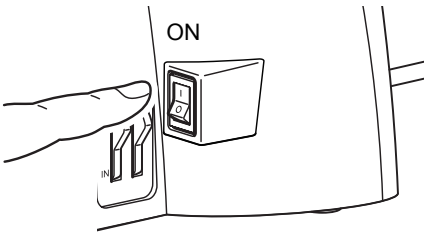
1. **Turn on the power.**
2. **Lower condenser slightly from uppermost position.**
3. **Fully open field and aperture diaphragms.**
4. **Bring the 10x objective into the optical path.**
5. **Bring specimen into optical path.**
6. **Focus on specimen.**
7. **Adjust diopter.**
8. **Adjust interpupillary.**
9. **Focus and center condenser.**
10. **Bring the desired objective into the optical path.**
11. **Adjust the aperture diaphragm.**
12. **Focus on specimen.**
13. **Circumscribe field diaphragm to field of view.**
14. **View specimen.**
15. **Turn off the power.**



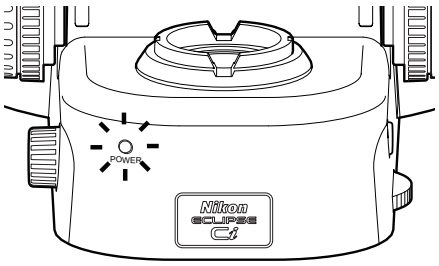

### Preparation for microscopy

**1** **Turn on the power.**

Press the switch to the "|" position to turn on the power to the microscope (the power LED on the front of the main body will light up to indicate that dia-illumination is turned ON).



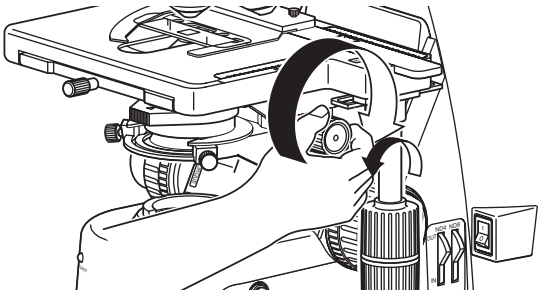

**Power on**




**LED on**

## 2 Lower the condenser slightly from the uppermost position.

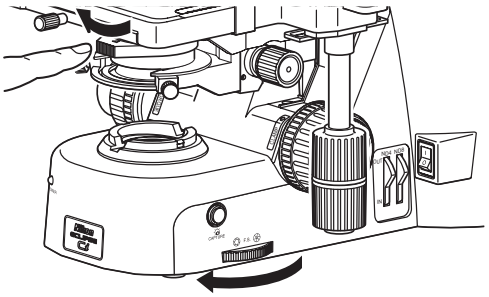
Turn the condenser focus knob until the condenser is positioned at the upper limit (where it clicks to a stop), and then lower it a little.



**Lower the condenser from upper limit**

## 3 Fully open the field diaphragm and aperture diaphragm.

Turn the field diaphragm dial and the aperture diaphragm lever clockwise to open them completely.

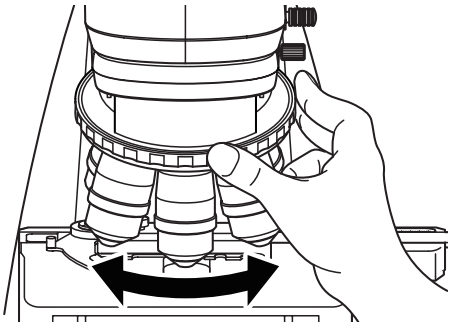


**Fully open field and aperture diaphragms.**

## 4 Bring the 10x objective into the optical path.

Turn the nosepiece to bring the 10x objective into the optical path.

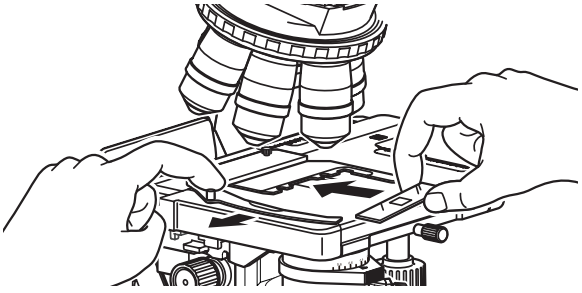
> ✔
> Turn the nosepiece until it clicks.



**Bring the 10x objective into the optical path.**

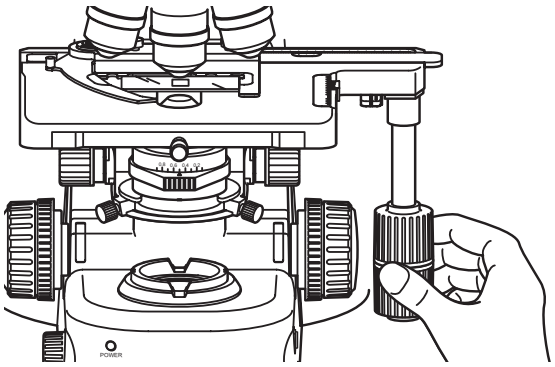
## 5 Place a specimen on the stage, and move the stage to bring the target into view.

(1) Open the claw of the specimen holder's moving part and place the specimen onto the stage, gently stowing the claw back to fix the specimen.



**Setting the specimen**

(2) Rotate the stage knob to move the stage and bring the target into the optical path.
(So that the sample sealed under the cover glass will be lighted.)

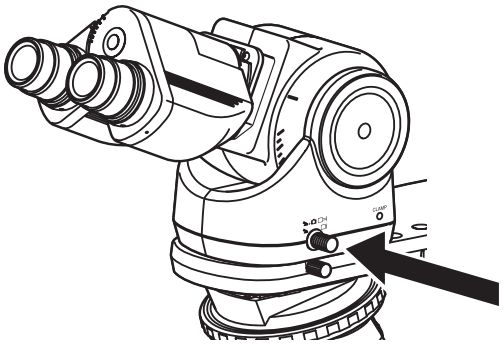


**Bringing the Target into the Optical Path**

## 6 Focus on the specimen. (→See Chapter 2 "2 Focusing on the Specimen (Vertical Stage Movement)" for details)

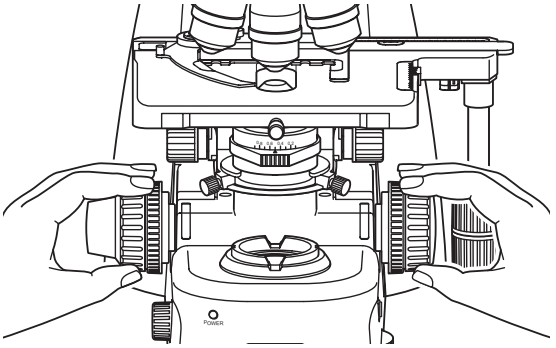
(1) When using the trinocular tube or ergonomic tube, push in the optical path switching lever to distribute 100% light to the binocular section.

(2) Look into the eyepiece and turn the coarse focus knob away to raise the stage to the upper limit. From there, focus on the specimen by lowering the stage.



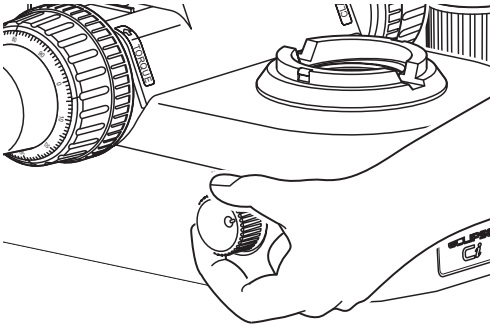
**Switching the Optical Path 100% to the Binocular Part**

(3) When the focus was roughly adjusted using coarse focus knob, turn the fine focus knob to accurately adjust the focus.



**Focusing on the Specimen**

(4) Adjust the brightness of the field of view by turning the dia-illumination brightness control knob.

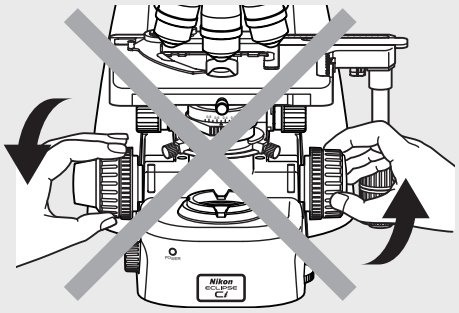


**Brightness Adjustment**

**Notes on controlling the focus knobs**

Avoid the following action, which can cause equipment malfunction.

- Rotating the right and left focus knobs in opposite directions.
- Rotating the coarse focus knob past the limit.



**Don't rotate the knobs in opposite directions!**



## 7 Adjust the diopter. (→See Chapter 2, "4 Adjusting the Diopter" for details)

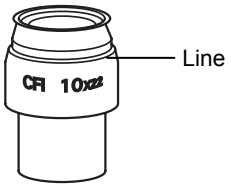
(1) Turn the diopter adjustment ring on the right and left eyepieces to align the end face of the diopter adjustment ring with the line. (This is the diopter adjustment reference position.)

(2) Focus on the specimen using the 40x objective.

(3) Bring the 10x (or 4x) objective into the optical path.

(4) Look into the right eyepiece with your right eye and the left eyepiece with your left eye. Turn the diopter adjustment ring of each eyepiece to focus on the specimen. At this point no focus knobs are used.

(5) Repeat Steps (2) through (4) to make sure the focus has been adjusted properly.



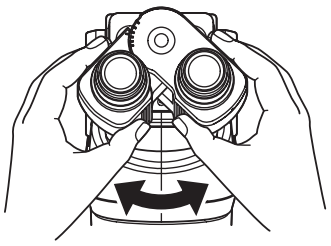

**Reference position for diopter adjustment**

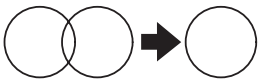
**Adjusting the Diopter**

## 8 Adjust the interpupillary distance.

Look into both eyepieces and rotate the binocular part to adjust the binocular part's opening until the fields of view for the right and left eyes coincide.

For easy adjustment, look into the eyepiece as if you were looking at a distant object.

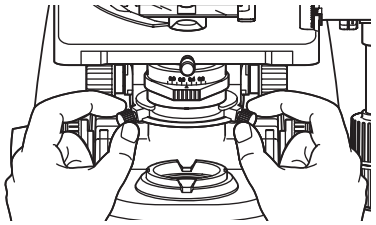




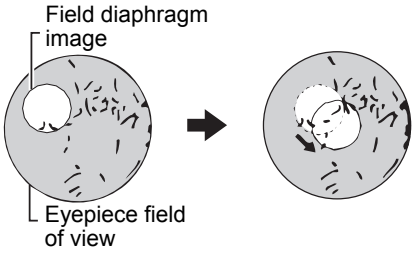
**Adjusting Interpupillary Distance**

## 9 Focus and center the condenser. (See Chapter 2, "5 Focusing and Centering the Condenser" for details)

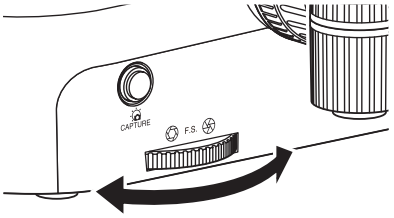
(1) Look into the eyepiece with the field diaphragm stopped down to the minimum. Focus on the field diaphragm image using the condenser focus knob, then adjust the condenser centering screws to center the diaphragm image within the field of view.

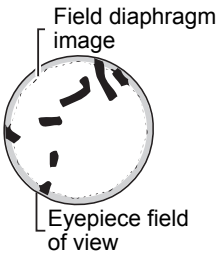


(2) Bring the 40x objective into the optical path to check the focus and centering of the field diaphragm image. Make adjustments in the same way as step (1) as necessary.




(3) Turn the field diaphragm dial and adjust the field diaphragm image so that its size is almost the same as the field of view.



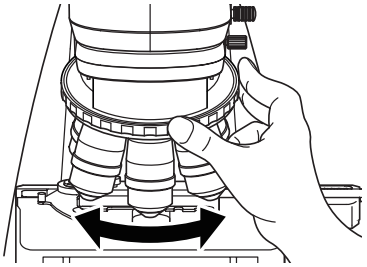

**Adjusting aperture diaphragm**

## Microscopy operation

### 10 Select the desired objective.

Turn the nosepiece to bring the desired objective into the optical path.

> **Swing-out condenser 1-100x**
> Mounting a 1-100x swing-out condenser on the Ci-L main body when using the 1x objective may result in uneven illumination around the field of view.

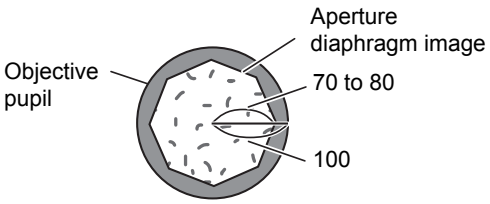


**Bring an arbitrary objective into the optical path.**

### 11 Adjust the aperture diaphragm. (→See Chapter 2, "6 Adjusting the Aperture Diaphragm" for details)

Turn the aperture diaphragm lever on the condenser to adjust the aperture diaphragm so that it is set to 70 to 80% of the numerical aperture of the objective used.

> Be sure to adjust the aperture diaphragm each time you change the objective.
> (You can see the aperture diaphragm image with the centering telescope.)




**Right size of the aperture diaphragm**

### 12 Focus on the specimen.

(1) Look into the eyepiece, and adjust the brightness of the field of view by turning the dia-illumination brightness control knob. You can also adjust the brightness with an ND filter for Ci-S.

(2) Rotate the stage knob to move the stage and bring the target into the optical path.

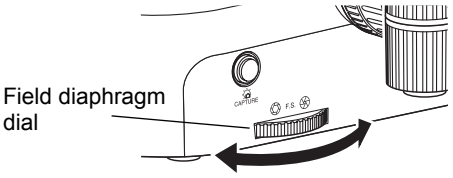
(3) If the specimen is not in focus, turn the focus knob to focus on it.

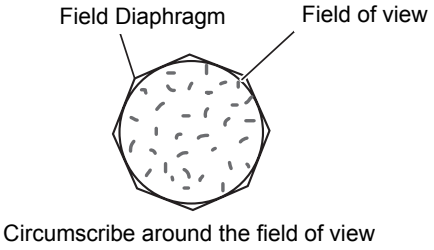
## 13 Adjust the field diaphragm

Turn the field diaphragm dial to adjust the field diaphragm so that it almost circumscribes the field of view.

> **Size of the field diaphragm**
>
> Normally, adjust the field diaphragm so that it almost circumscribes the field of view. Opening the field diaphragm too much results in stray light entering the field of view, generating flare and reducing the image contrast. In addition, the specimen will become decolorized over a wider area.

> **Field diaphragm's adjustment timing**
>
> Be sure to adjust the field diaphragm each time you change the objective.



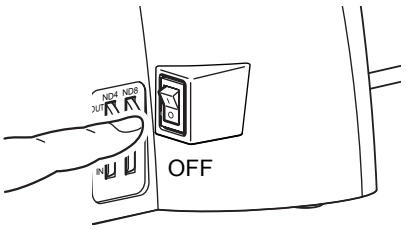

**Adjusting the field diaphragm**

## 14 View the specimen.

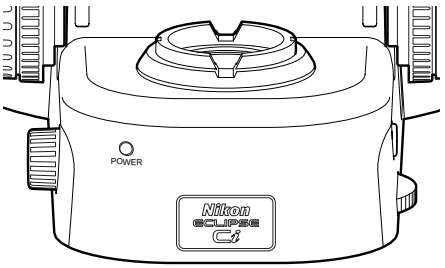
Rotate the stage knob to move the target. If the target is not in focus, use the focus knob to adjust the focus.

## 15 Turn off the power.

Turn off the power switch (press to the “O” position) for the microscope. (The power LED on the front of the main body will turn off.)




**Power off**

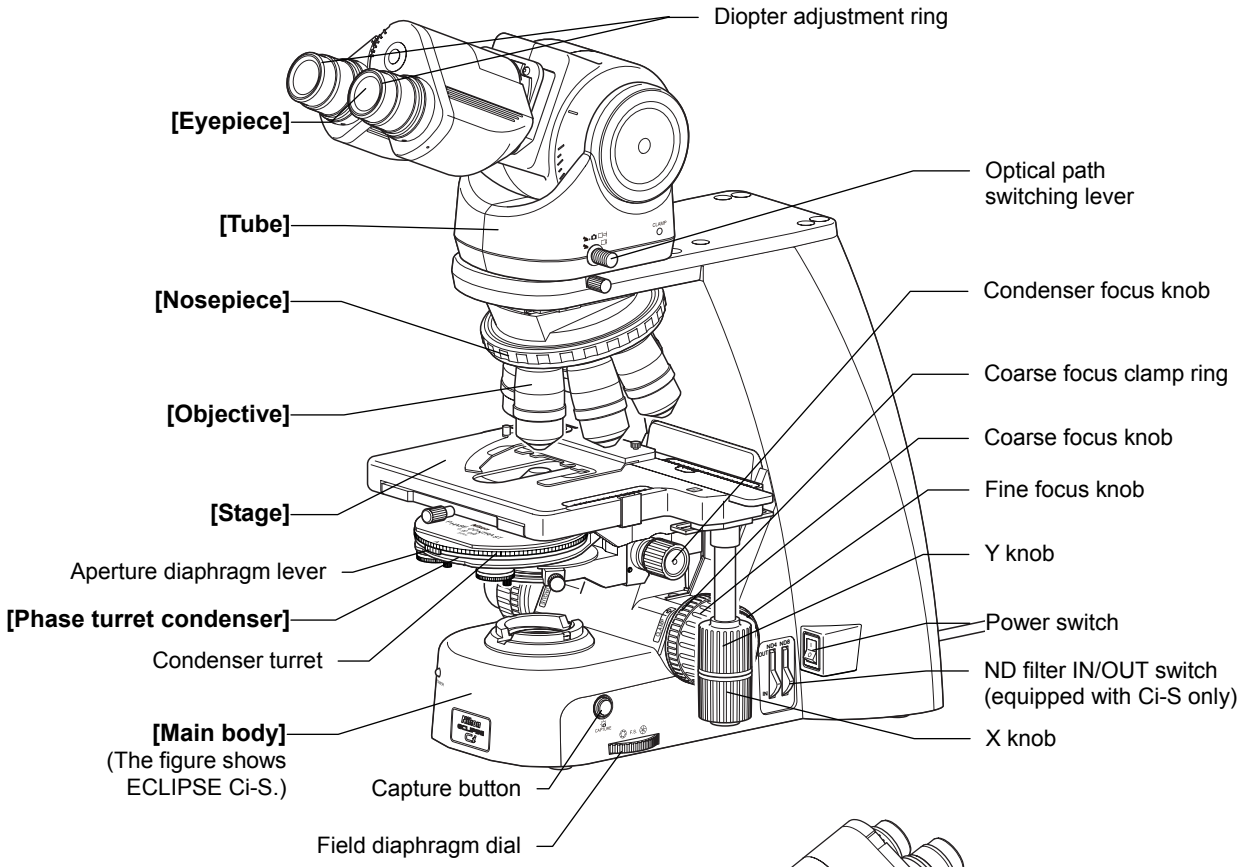


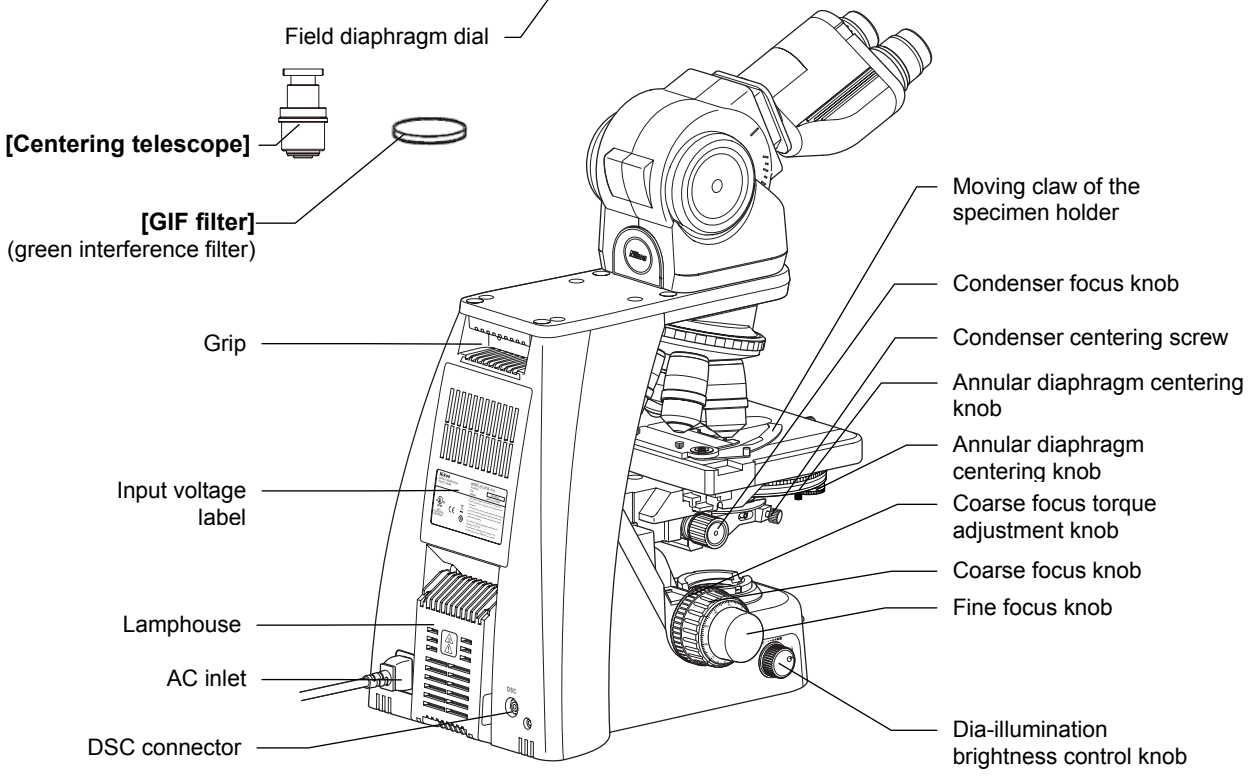

**Power LED off**

# 2 Phase Contrast Microscopy

## 2.1 System Components and Controls

This section explains an example system configuration and the controls required for phase contrast microscopy using the ECLIPSE Ci-S/Ci-L.
Names of components are denoted in the following manner: **[Phase contrast condenser]**.

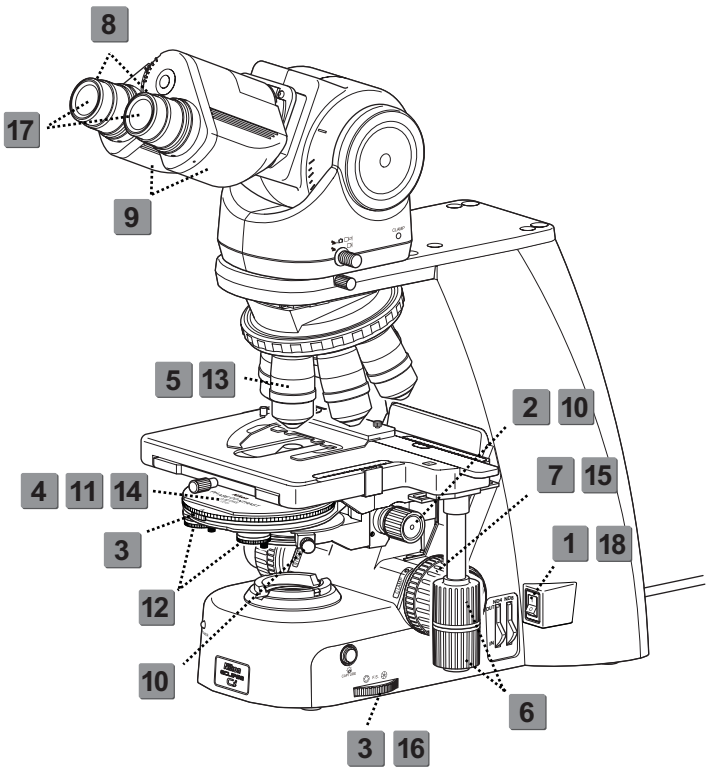


## 2.2 Phase Contrast Microscopy Procedure

In this procedure, only step titles are shown for operations that are the same as those of bright-field microscopy. See “2.1 Bright-field Microscopy” for details.

1. **Turn on the power.**
2. **Lower condenser slightly from uppermost position.**
3. **Fully open field and aperture diaphragms.**
4. **Move the turret to the [A: empty] position.**
5. **Bring the 10x Ph objective into the optical path.**
6. **Bring specimen into optical path.**
7. **Focus on specimen.**
8. **Adjust diopter.**
9. **Adjust interpupillary.**
10. **Focus and center condenser.**
11. **Bring the Ph annular diaphragm [Ph1] position into the optical path.**
12. **Center the Ph annular diaphragm.**
13. **Bring the desired Ph objective into the optical path.**
14. **Match the Ph codes of annular diaphragm and the objective.**
15. **Focus on specimen.**
16. **Circumscribe field diaphragm to field of view.**
17. **View specimen.**
18. **Turn off the power.**




### Preparation for microscopy

**1** **Turn on the power.**

(The power LED on the front of the main body will light up.)

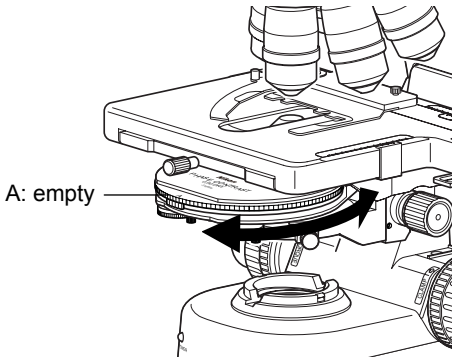
**2** **Lower the condenser slightly from the uppermost position.**

**3** **Fully open the field diaphragm and aperture diaphragm.**

**4** **Move the condenser turret to the [A: empty] position.**

Turn the condenser turret until the [A: empty] symbol comes to the front, and align the hole with the optical path.

> **Aperture diaphragm of the condenser**
>
> Positioning the condenser turret at positions other than [A: empty] deviates the aperture diaphragm from the optical path.




**Move the condenser turret to the [A: empty] position.**

**5** **Bring the 10x Ph objective (Ph1) into the optical path.**

**6** **Place a specimen on the stage, and move the stage to bring the target into view.**

**7** **Focus on the specimen.** **(→See Chapter 2 "2 Focusing on the Specimen" for details)**

**8** **Adjust the diopter.** **(→See Chapter 2, "4 Adjusting the Diopter" for details)**

**9** **Adjust the interpupillary distance.**

**10** **Focus and center the condenser.** **(See Chapter 2, "5 Focusing and Centering the Condenser" for details)**

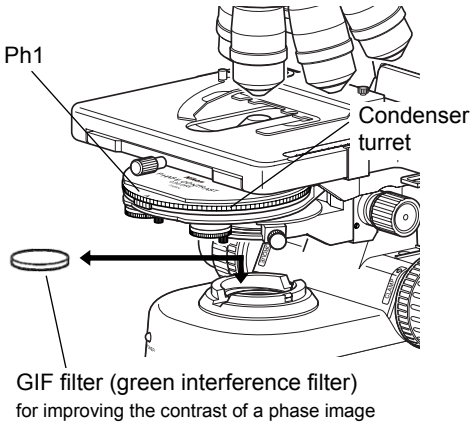
## Microscopy operation (→See also: Chapter 2, Section 14 "Tips for Phase Contrast Microscopy")

### 11 Bring the Ph annular diaphragm (Ph1) in the condenser turret into the optical path.

Turn the condenser turret until the [Ph1] symbol comes to the front.

**GIF filter**

The GIF filter (green interference filter) improves the contrast when placed in the optical path. The filter should be installed on the field lens, or placed inside or on top of the filter cassette holder. Note, however, that it may cause ghosting when mounted inside the filter cassette holder.




**Bring the Ph annular diaphragm into the optical path.**

### 12 Center the Ph annular diaphragm.

To optimize the phase effect, it is important to properly overlap the phase plate of the objective with the Ph annular diaphragm image in the condenser.

(1) Make sure that the 10x objective (Ph1) has been placed into the optical path and that the [Ph1] symbol on the condenser turret is facing the front.

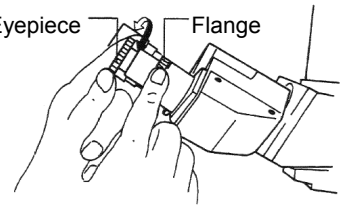
(2) Rotate the stage knob to move the specimen and bring a portion where there is no sample under the cover glass into the optical path.

(3) Remove one eyepiece from the tube, and insert the centering telescope with adapter into the tube.

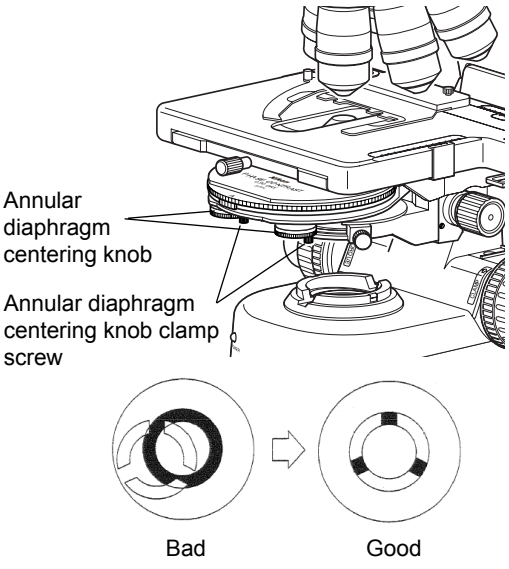
(4) Hold the flange of the centering telescope and rotate the eyepiece to focus on the phase plate of the objective.

(5) If the phase plate of the objective and the annular diaphragm in the condenser are misaligned, loosen the clamp screws of the two annular diaphragm centering knobs before rotating the centering knob to move the entire turret for annular adjustment. Tighten the clamp screws after the completion of the adjustment.

(6) Remove the centering telescope and adapter from the tube, and reattach the eyepiece.




**Focusing on the Phase Ring**




**Centering the Ph annular diaphragm**

## 13 Bring an arbitrary Ph objective into the optical path.

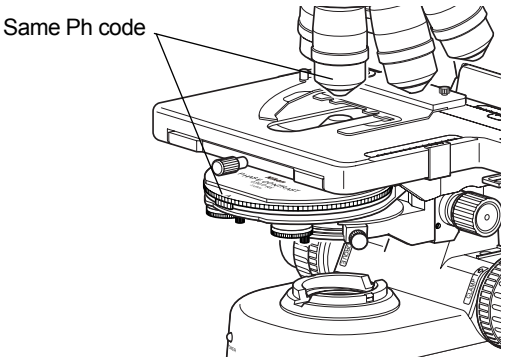
Turn the nosepiece to bring the desired Ph objective into the optical path.

When using an oil immersion type objective, apply immersion oil between the specimen and the objective. (→Chapter 2 “12 Oil Immersion” for details)

> **Phase turret condenser**
>
> The phase turret condenser is intended to be used dry. DO NOT apply immersion oil between the condenser tip's ball and the specimen.

## 14 Adjust the Ph annular diaphragm in the condenser with the Ph objective to be used.

Turn the condenser turret to bring an annular diaphragm with the same Ph code as the objective into the optical path.




**Matching the Ph codes of Ph annular diaphragm and the Ph objective**

> **Ph code**
>
> One of the Ph codes, [Ph1], [Ph2], or [Ph3] is indicated on the Ph objective depending on the size of the phase plate. (Ph codes have nothing to do with the magnification of the objective.) Always use a Ph objective and Ph annular diaphragm with the same Ph code. You cannot experience the phase effect if a different combination of the codes might be used.

> **Centering of the annular diaphragm and the phase plate**
>
> The position of each annular diaphragm in the condenser turret has already been adjusted based on the Ph1 annular diaphragm, but the phase image will differ slightly depending on how the annular diaphragm overlaps the phase plate. For a stricter observation or capturing of still images, check whether the annular diaphragm and the phase plate are concentric at each magnification.

## 15 Focus on the specimen.

(1) Look into the eyepiece, and adjust the brightness of the field of view by turning the dia-illumination brightness control knob. You can also adjust the brightness with an ND filter for Ci-S.

(2) Rotate the stage knob to move the stage and bring the target into the optical path.

(3) If the specimen is not in focus, turn the focus knob to focus on it.

## 16 Adjust the field diaphragm.

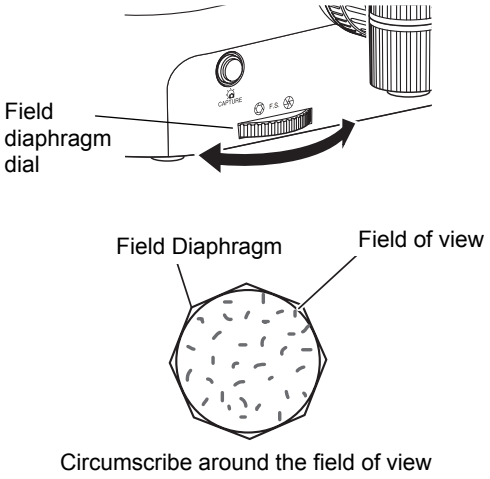
Turn the field diaphragm dial to adjust the field diaphragm so that it almost circumscribes the field of view.

**Size of the field diaphragm**

Normally, adjust the field diaphragm so that it almost circumscribes the field of view. Opening the field diaphragm too much results in stray light entering the field of view, generating flare and reducing the image contrast. In addition, the specimen will become decolorized over a wider area.

**Field diaphragm's adjustment timing**

Be sure to adjust the field diaphragm each time you change the objective.




**Adjusting the field diaphragm**

## 17 View the specimen.

Rotate the stage knob to move the target. If the target is not in focus, use the focus knob to adjust the focus.

**To switch to bright-field microscopy**

- Turn the condenser turret until the [A: empty] symbol comes to the front.
- Microscopy is possible with an objective of 4x or greater, but UW microscopy with the 4x objective will cause vignetting.
- When the condenser is set to [A: empty], its performance is equivalent to that of the Abbe condenser.

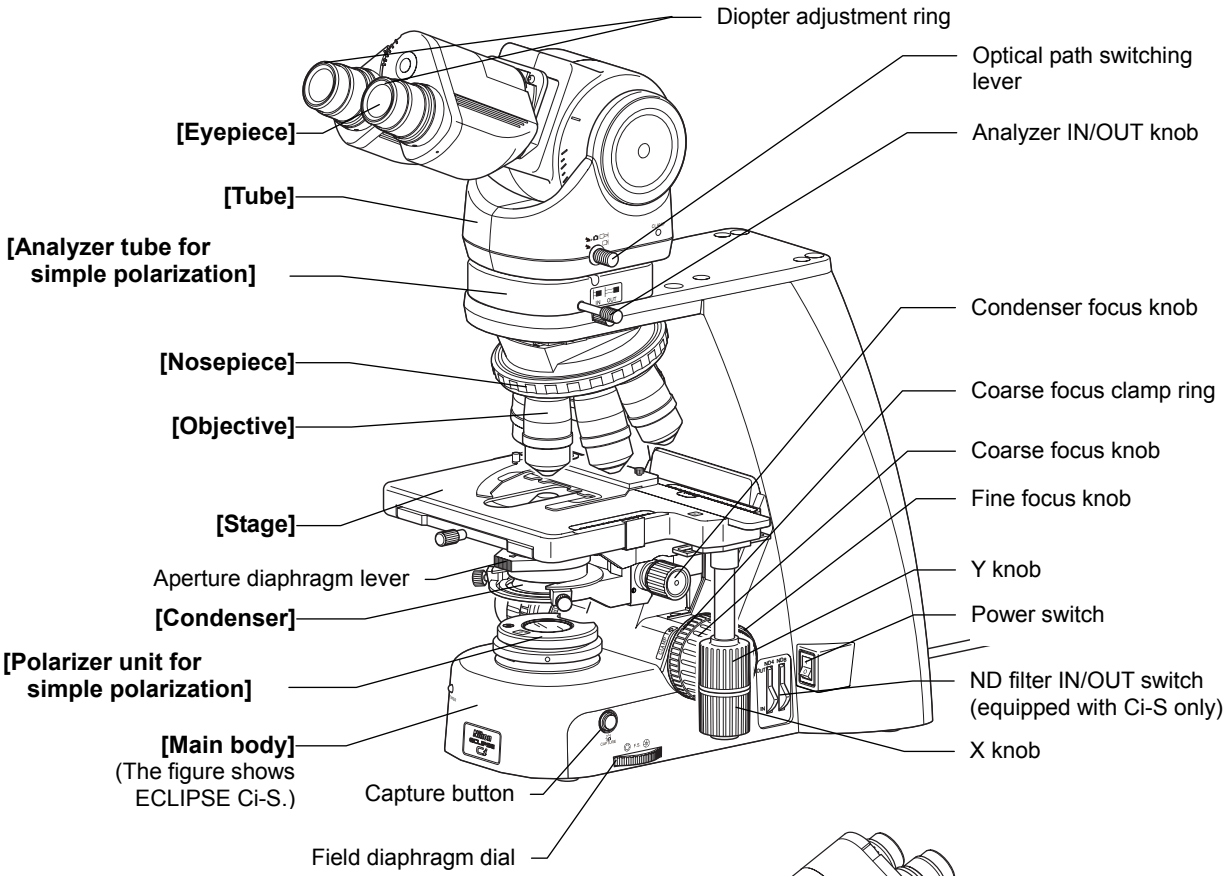
## 18 Turn off the power.

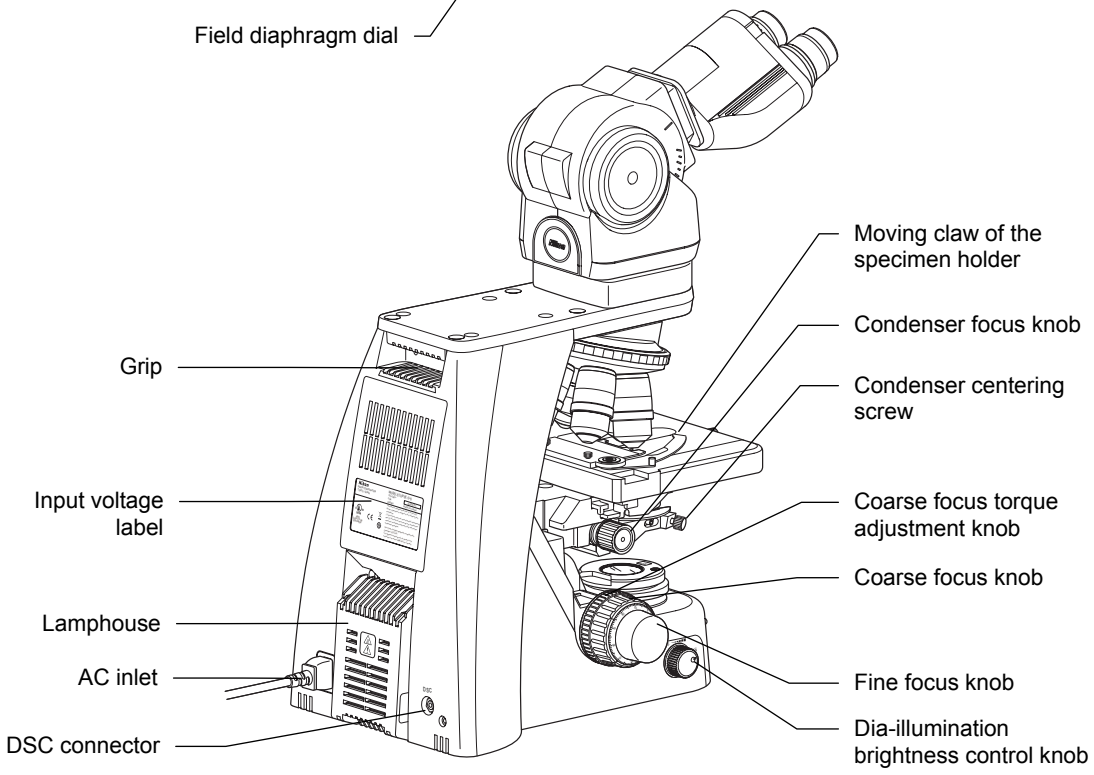
Turn off the power switch (press to the “O” position) for the microscope. (The power LED on the front of the main body will turn off.)

# 3 Simple Polarizing Microscopy

## 3.1 System Configuration and Controls

This section explains an example system configuration and the controls required for simple polarizing microscopy using the ECLIPSE Ci-S/Ci-L.
Names of components are denoted in the following manner: **[Polarizer unit for simple polarization]**.

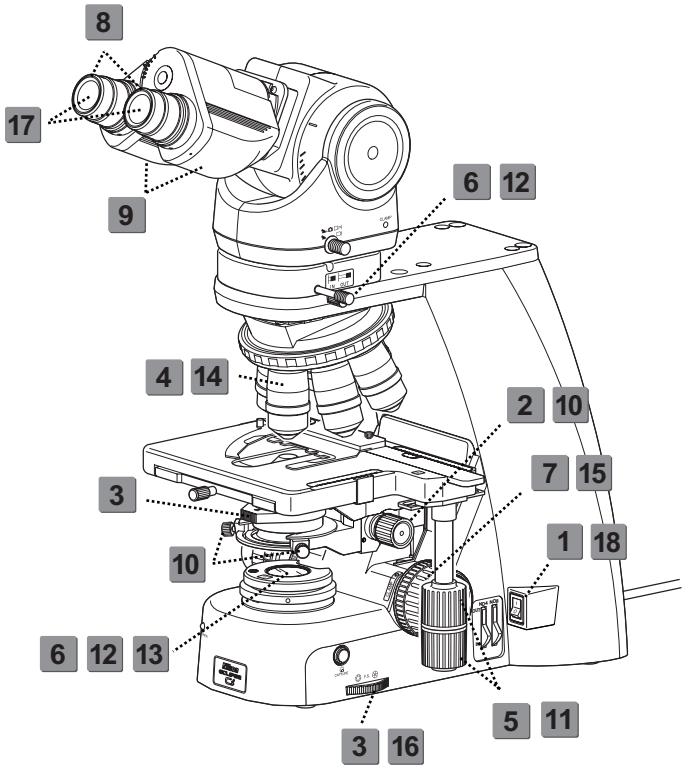


## 3.2 Simple Polarizing Microscopy Procedure

In this procedure, only step titles are shown for operations that are the same as those of bright-field microscopy. See “2.1 Bright-field Microscopy” for details.

1. **Turn on the power.**
2. **Lower condenser slightly from uppermost position.**
3. **Fully open field and aperture diaphragms.**
4. **Bring the 10x objective into the optical path.**
5. **Bring specimen into optical path.**
6. **Remove the analyzer from the optical path.**
7. **Focus on specimen.**
8. **Adjust diopter.**
9. **Adjust interpupillary.**
10. **Focus and center condenser.**
11. **Bring the portion without sample into the optical path.**
12. **Install polarizer unit.**
13. **Adjust orientation of analyzer and polarizer.**
14. **Bring the desired objective into the optical path.**
15. **Focus on specimen.**
16. **Circumscribe field diaphragm to field of view.**
17. **View specimen.**
18. **Turn off the power.**




### Preparation for microscopy

**1** **Turn on the power.**

(The power LED on the front of the main body will light up.)

**2** **Lower the condenser slightly from the uppermost position.**

**3** **Fully open the field diaphragm and aperture diaphragm.**

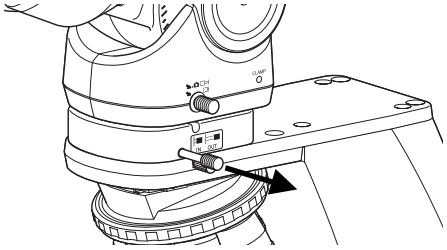
**4** **Bring the 10x objective into the optical path.**

**5** **Place a specimen on the stage, and move the stage to bring the target into view.**

**6** **Remove the analyzer and the polarizer from the optical path.**

Pull out the analyzer IN/OUT knob from the intermediate tube with simple analyzer to remove the analyzer from the optical path.

The polarizer unit for simple polarization has not yet been installed at this point.



**Remove the analyzer from the optical path.**

**7** **Focus on the specimen.** **(→See Chapter 2 "2 Focusing on the Specimen (Vertical Stage Movement)" for details)**

**8** **Adjust the diopter.** **(→See Chapter 2, "4 Adjusting the Diopter" for details)**

**9** **Adjust the interpupillary distance.**

**10** **Focus and center the condenser.** **(→See Chapter 2, "5 Focusing and Centering the Condenser" for details)**

## Microscopy operation

### 11 Bring a portion of the specimen where there is no sample into the optical path.

Rotate the stage knob to move the specimen and bring a portion where there is no sample under the cover glass into the optical path.

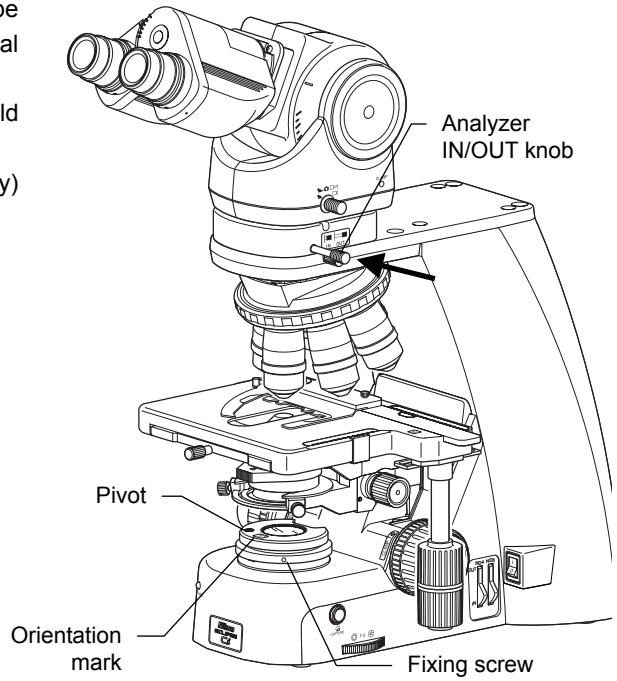
### 12 Bring the analyzer and the polarizer into the optical path.

Push in the analyzer IN/OUT knob into the analyzer tube for simple polarization to bring the analyzer into the optical path.

Set the polarizer unit for simple polarization over the field lens.

Make sure the orientation mark on the polarizer (roughly) comes to the front at this point.

Keep the fixing screw of the polarizer unit loosened.



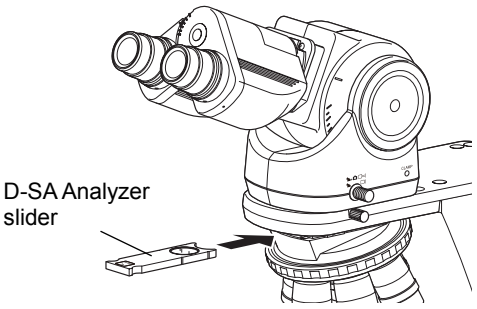

**Bringing the analyzer into the optical path
Installing the polarizer**

**Slider-type analyzer for simple polarization**

Using the D-SA Analyzer Slider for Simple Polarization instead of an analyzer tube for simple polarization can keep the eye point from rising. To use the D-SA Analyzer Slider for Simple Polarization, the C-NA sextuple nosepiece with analyzer slot is required. Use the analyzer slider as follows:

Push in: The analyzer goes into the optical path.

Pull out: The analyzer is removed from and the dummy hole goes into the optical path.




**Using D-SA analyzer slider**

## 13 Adjust the orientation of the analyzer and the polarizer.

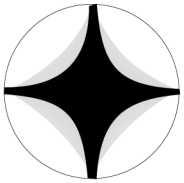
(1) Fully open the aperture diaphragm.

(2) Pull out one eyepiece from the tube.

(3) Look into the eyepiece sleeve and rotate the whole polarizer unit until you can identify a dark cross. (You will see black stripes that change shape as you rotate the polarizer unit.)

(4) Tighten the fixing screw of the polarizer unit to fix the polarizer.

(5) Place the eyepiece back into the tube.



Dark cross

> **Dark cross (crossed Nicols)**
>
> You will see a dark cross when the orientation of the analyzer orthogonally crosses that of the polarizer.

## 14 Bring an arbitrary objective into the optical path.

Turn the nosepiece to bring the desired objective into the optical path.

> **Swing-out condenser 1-100x**
>
> Mounting a 1-100x swing-out condenser on the Ci-L main body when using the 1x objective may result in uneven illumination around the field of view.

## 15 Focus on the specimen.

(1) Look into the eyepiece, and adjust the brightness of the field of view by turning the dia-illumination brightness control knob. You can also adjust the brightness with an ND filter for Ci-S.

(2) Rotate the stage knob to move the stage and bring the target into the optical path.

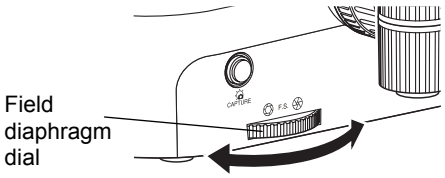
(3) If the specimen is not in focus, turn the focus knob to focus on it.

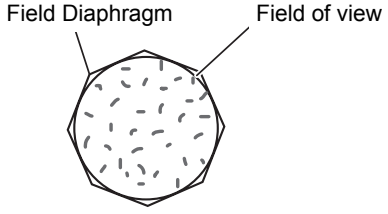
## 16 Adjust the field diaphragm.

Turn the field diaphragm dial to adjust the field diaphragm so that it almost circumscribes the field of view.

> **Size of the field diaphragm**
>
> Normally, adjust the field diaphragm so that it almost circumscribes the field of view. Opening the field diaphragm too much results in stray light entering the field of view, generating flare and reducing the image contrast. In addition, the sample will become decolorized over a wider area.

> **Field diaphragm's adjustment timing**
>
> Be sure to adjust the field diaphragm each time you change the objective.




Circumscribe around the field of view

**Adjusting the field diaphragm**

## 17 View the specimen.

Rotate the stage knob to move the target. If the target is not in focus, use the focus knob to adjust the focus.

> **Strict polarizing microscopy**
>
> If you need a retardation measurement or stricter polarizing observation, use a dedicated polarizing microscope.

> **To switch to bright-field microscopy**
>
> Pull out the analyzer IN/OUT knob to remove the analyzer from the optical path. Remove the polarizer unit from the field lens.

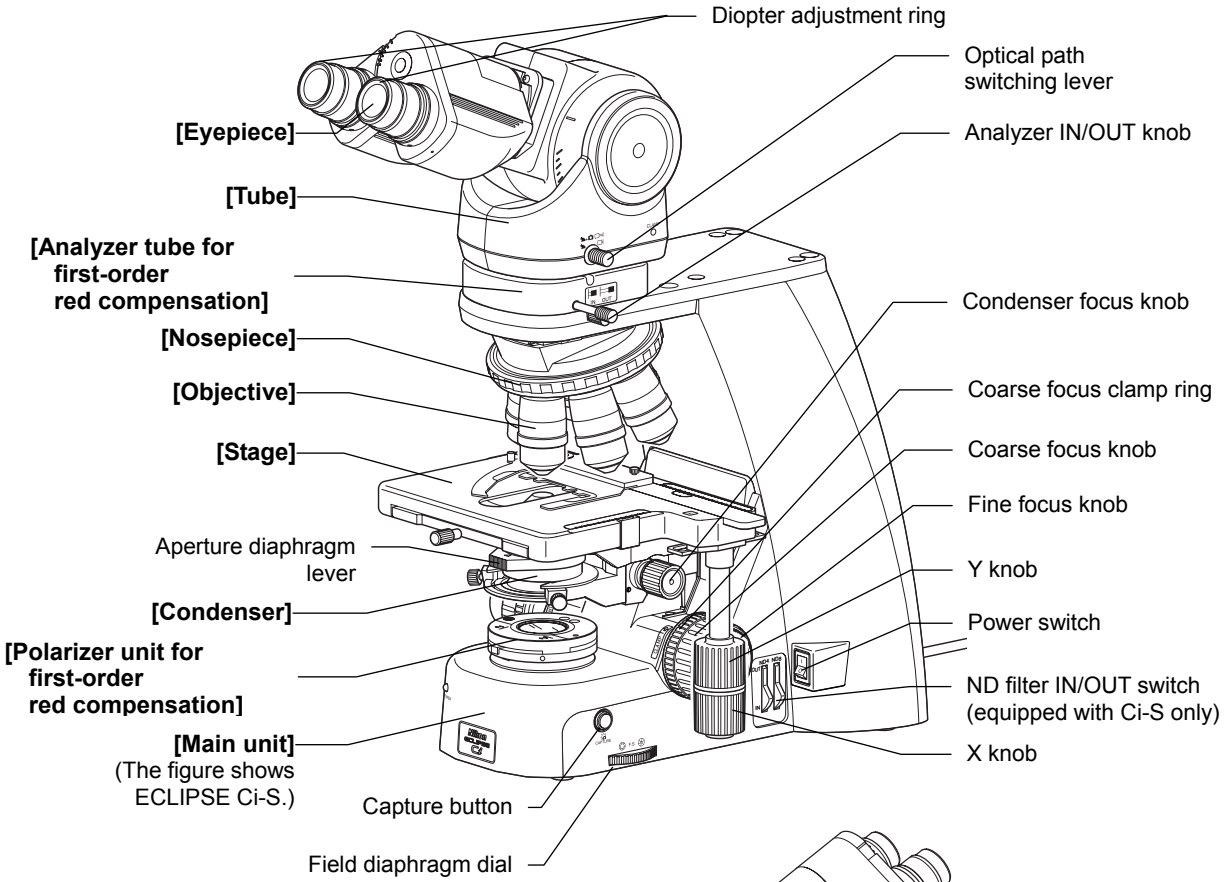
## 18 Turn off the power.

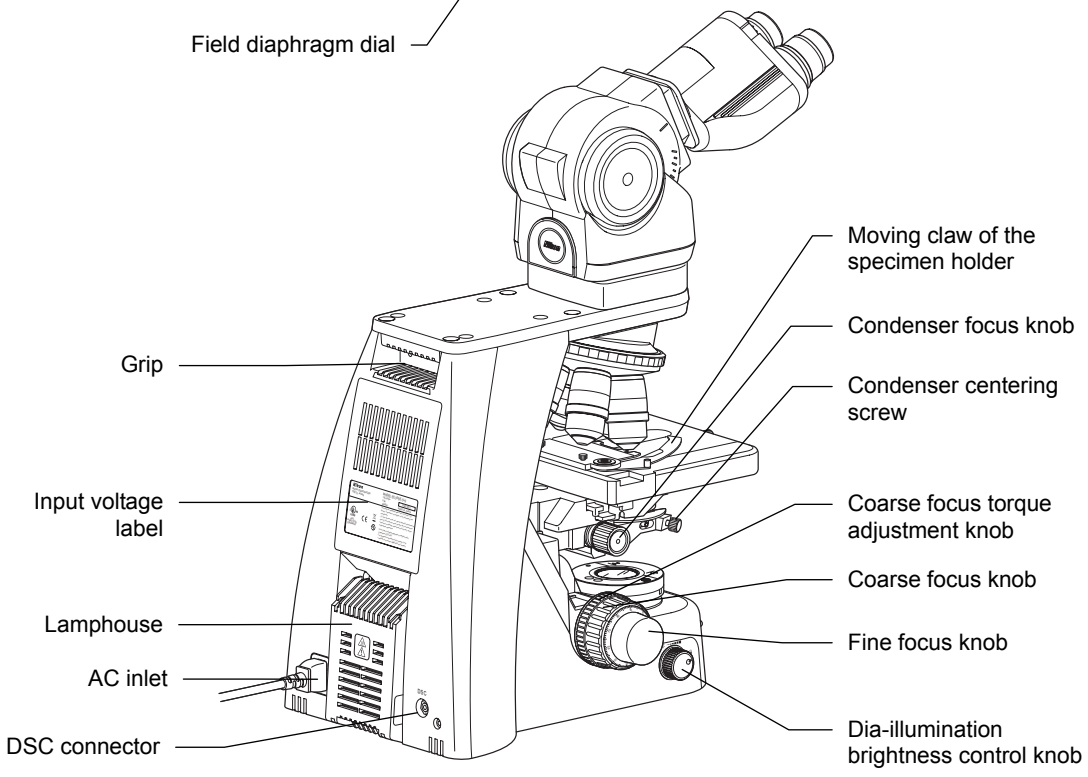
Turn off the power switch (press to the “O” position) for the microscope. (The power LED on the front of the main body will turn off.)

# 4 Sensitive Tint Plate Microscopy

## 4.1 System Configuration and Controls

This section explains an example system configuration and the controls required for sensitive polarization microscopy using the ECLIPSE Ci-S/Ci-L.
Names of components are denoted in the following manner: **[Polarizer unit for first-order red compensation]**.

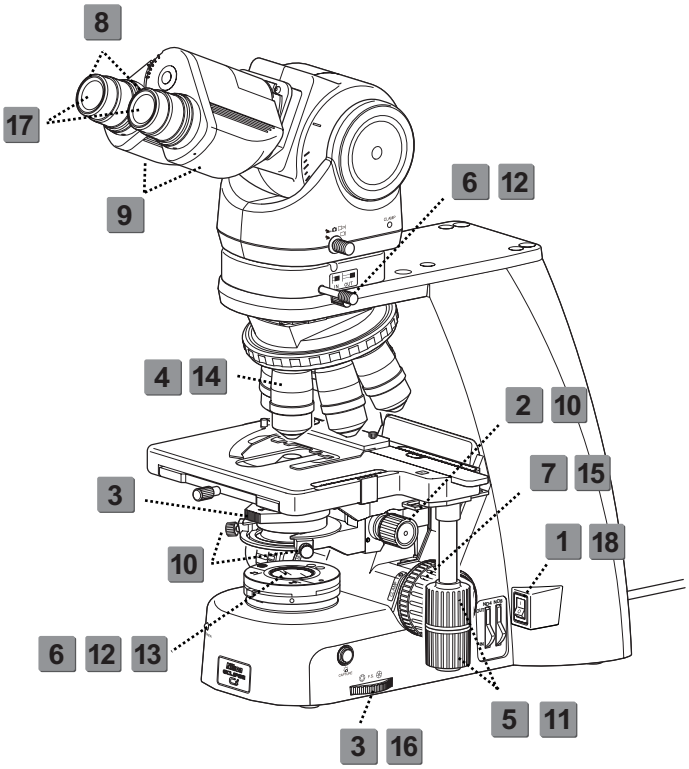


## 4.2 Sensitive Tint Plate Microscopy Procedure

In this procedure, only step titles are shown for operations that are the same as those of bright-field microscopy. See “2.1 Bright-field Microscopy” for details.

1. **Turn on the power.**
2. **Lower condenser slightly from uppermost position.**
3. **Fully open field and aperture diaphragms.**
4. **Bring the 10x objective into the optical path.**
5. **Bring specimen into optical path.**
6. **Remove the analyzer from the optical path.**
7. **Focus on specimen.**
8. **Adjust diopter.**
9. **Adjust interpupillary.**
10. **Focus and center condenser.**
11. **Bring the portion without sample into the optical path.**
12. **Install polarizer unit.**
13. **Adjust orientation of analyzer and polarizer.**
14. **Bring the desired objective into the optical path.**
15. **Focus on specimen.**
16. **Circumscribe field diaphragm to field of view.**
17. **View specimen.**
18. **Turn off the power.**




### Preparation for microscopy

**1** **Turn on the power.**

(The power LED on the front of the main body will light up.)

**2** **Lower the condenser slightly from the uppermost position.**

**3** **Fully open the field diaphragm and aperture diaphragm.**

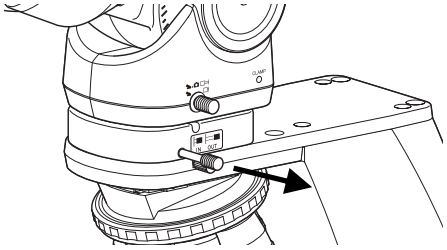
**4** **Bring the 10x objective into the optical path.**

**5** **Place a specimen on the stage, and move the stage to bring the target into view.**

**6** **Remove the analyzer and the polarizer from the optical path.**

Pull out the analyzer IN/OUT knob from the sensitive tint plate analyzer unit to remove the analyzer from the optical path.

The polarizer unit for first-order red compensation has not been installed at this point.



**Removing the analyzer from the optical path**

**7** **Focus on the specimen.** **(→See Chapter 2 "2 Focusing on the Specimen (Vertical Stage Movement)" for details)**

**8** **Adjust the diopter.** **(→See Chapter 2, "4 Adjusting the Diopter" for details)**

**9** **Adjust the interpupillary distance.**

**10** **Focus and center the condenser.** **(→See Chapter 2, "5 Focusing and Centering the Condenser" for details)**

## Microscopy operation

### 11 Bring a portion of the specimen where there is no sample into the optical path.

Rotate the stage knob to move the specimen and bring a portion where there is no sample under the cover glass into the optical path.

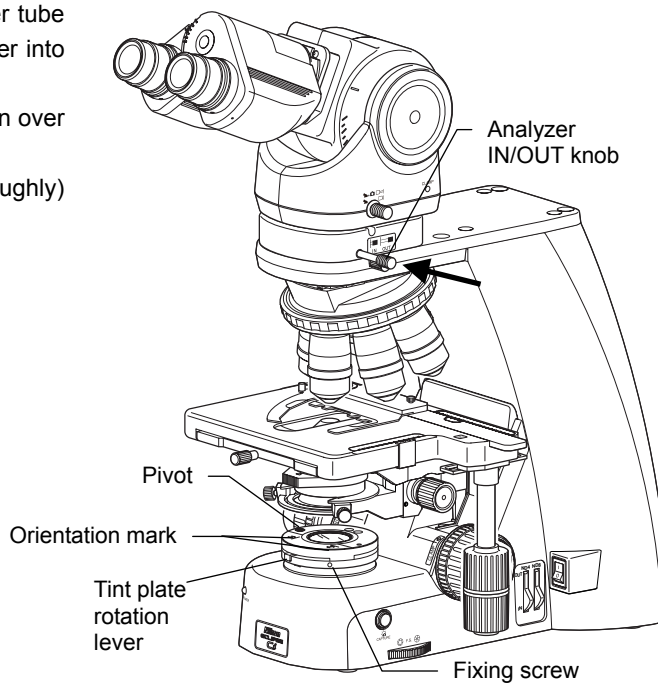
### 12 Bring the analyzer and the polarizer into the optical path.

Push in the analyzer IN/OUT knob into the analyzer tube for first-order red compensation to bring the analyzer into the optical path.

Set the polarizer unit for first-order red compensation over the field lens.

Make sure the orientation mark on the polarizer (roughly) comes to the front at this point.

Keep the fixing screw of the polarizer unit loosened.



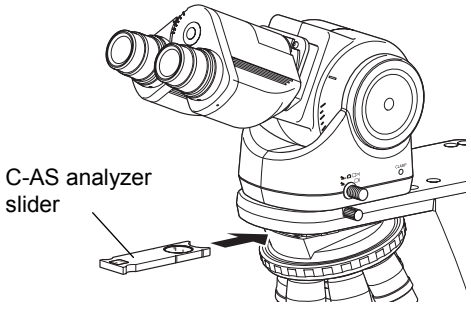

**Bringing the analyzer into the optical path Installing the polarizer**

**Slider-type analyzer for first-order red compensation**

Using the C-AS Analyzer Slider for First-order Red Compensation instead of an analyzer tube for first-order red compensation can keep the eye point from rising. To use the C-AS Analyzer Slider for First-order Red Compensation, the C-NA sextuple nosepiece with analyzer slot is required. Use the analyzer slider as follows:

Push in: The analyzer goes into the optical path.

Pull out: The analyzer is removed from and the dummy hole goes into the optical path.




**Using D-AS analyzer slider**

## 13 Adjust the orientation of the analyzer and the polarizer.

(1) Fully open the aperture diaphragm.

(2) Pull out one eyepiece from the tube.

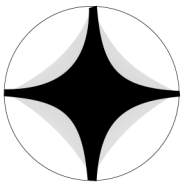
(3) Rotate the lambda plate rotation lever (which makes the upper polarizer move around the pivot) to remove the lambda plate from the optical path. When doing this, support the polarizer unit for first-order red compensation with hands so that it does not move.

(4) Look into the eyepiece sleeve and rotate the whole polarizer unit until you can identify a dark cross. (You will see black stripes that change shape as you rotate the polarizer unit.)

(5) Tighten the fixing screw of the polarizer unit to fix the polarizer.

(6) Put the eyepiece back to the tube and set the lambda plate back into the optical path.

(7) Rotate the lambda plate rotation lever to the left/right limit to make sure that the field of view is colored magenta on both sides.



Dark cross

**Dark cross (crossed Nicols)**

You will see a dark cross when the orientation of the analyzer orthogonally crosses that of the polarizer.

## 14 Bring an arbitrary objective into the optical path.

Turn the nosepiece to bring the desired objective into the optical path.

**Swing-out condenser 1-100x**

Mounting a 1-100x swing-out condenser on the Ci-L main body when using the 1x objective may result in uneven illumination around the field of view.

## 15 Focus on the specimen.

(1) Look into the eyepiece, and adjust the brightness of the field of view by turning the dia-illumination brightness control knob. You can also adjust the brightness with an ND filter for Ci-S.

(2) Rotate the stage knob to move the stage and bring the target into the optical path.

(3) If the specimen is not in focus, turn the focus knob to focus on it.

## 16 Adjust the field diaphragm.

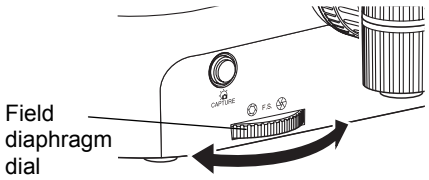
Turn the field diaphragm dial to adjust the field diaphragm so that it almost circumscribes the field of view.

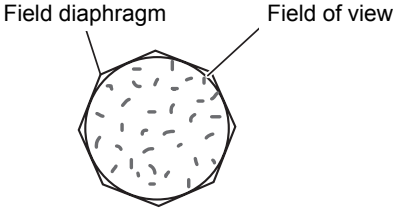
**Size of the field diaphragm**

Normally, adjust the field diaphragm so that it almost circumscribes the field of view. Opening the field diaphragm too much results in stray light entering the field of view, generating flare and reducing the image contrast. In addition, the sample will become decolorized over a wider area.

**Field diaphragm's adjustment timing**

Be sure to adjust the field diaphragm each time you change the objective.




Circumscribe around the field of view

**Adjusting the field diaphragm**

## 17 View the specimen.

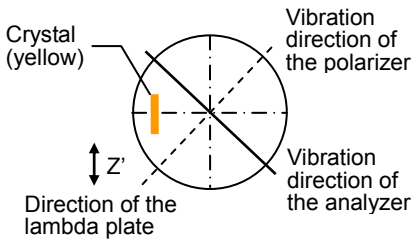
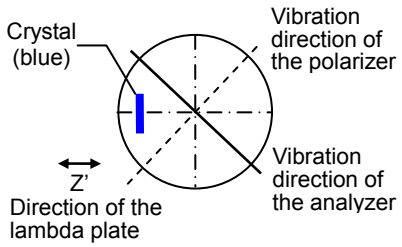
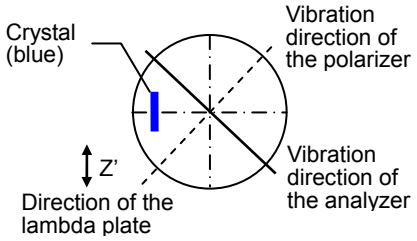
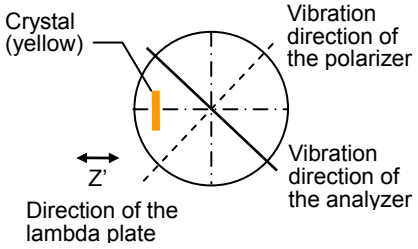
(1) Swing out the lambda plate from the optical path. (The field of view gets darker.)

(2) Rotate the stage knob to move the target. If the target is not in focus, use the focus knob to adjust the focus. (The specimen looks brighter in the dark field of view.)

(3) Bring the lambda plate back into the optical path. (The background of the field of view will be colored magenta.)

(4) Of the needle-like crystals seen in the field of view, check the color of the longitudinal ones.

(5) Turn the lambda plate rotation lever from right to left (clockwise) to check the change of color of the crystal being observed.
Identify the crystal by its change of color. (See the table below.)

| Crystal | Position of the lambda plate rotation lever | |
|---|---|---|
| | Leftmost | Rightmost |
| Urate crystal |  |  |
| Calcium pyrophosphoric acid crystal |  |  |

**Keep the lambda plate clean**

Note that dirt such as dust and fingerprint on the lambda plate can significantly degrade the polarization performance. Keep it clean.

**To switch to bright-field microscopy**

Pull out the analyzer IN/OUT knob to remove the analyzer from the optical path. Remove the polarizer unit from the field lens.

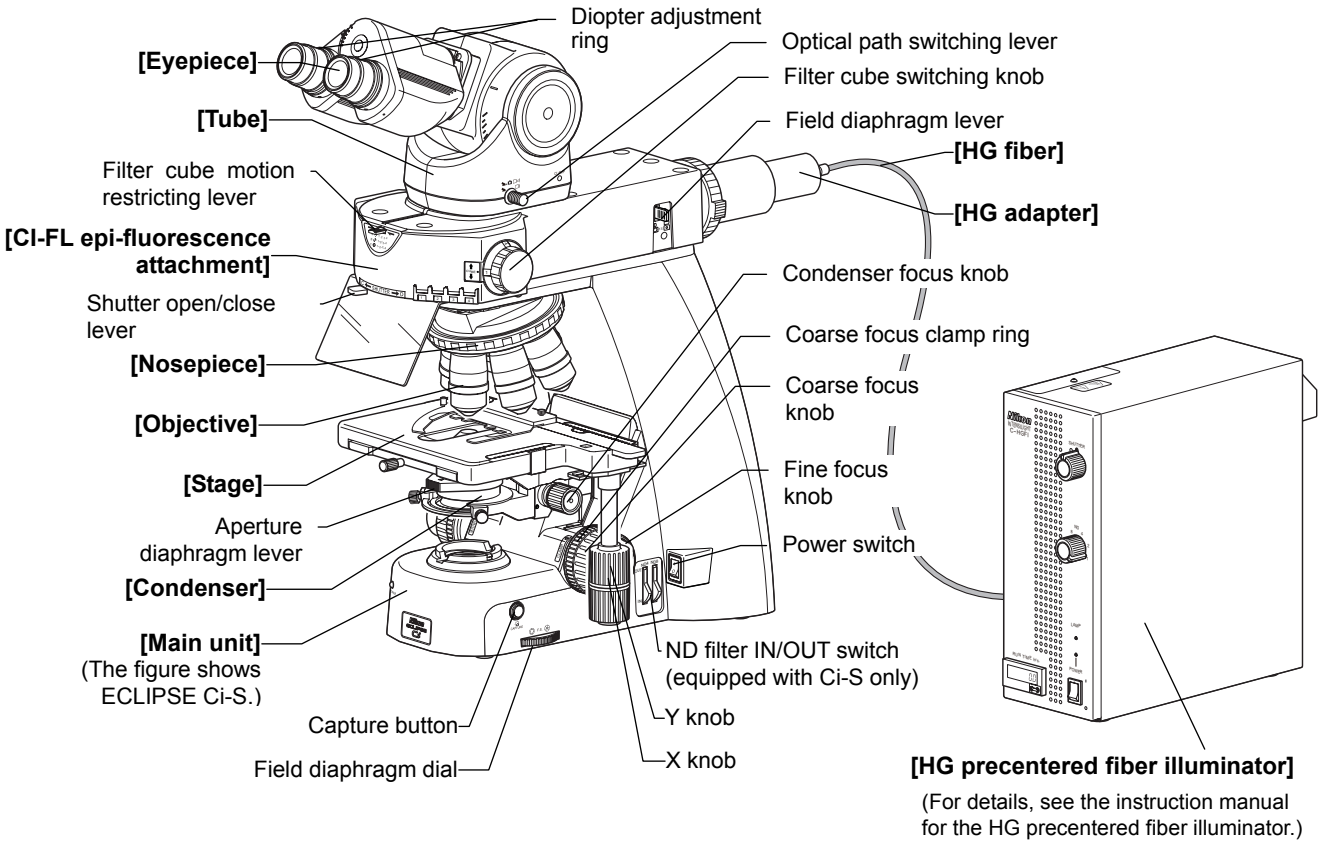
## 18 Turn off the power.

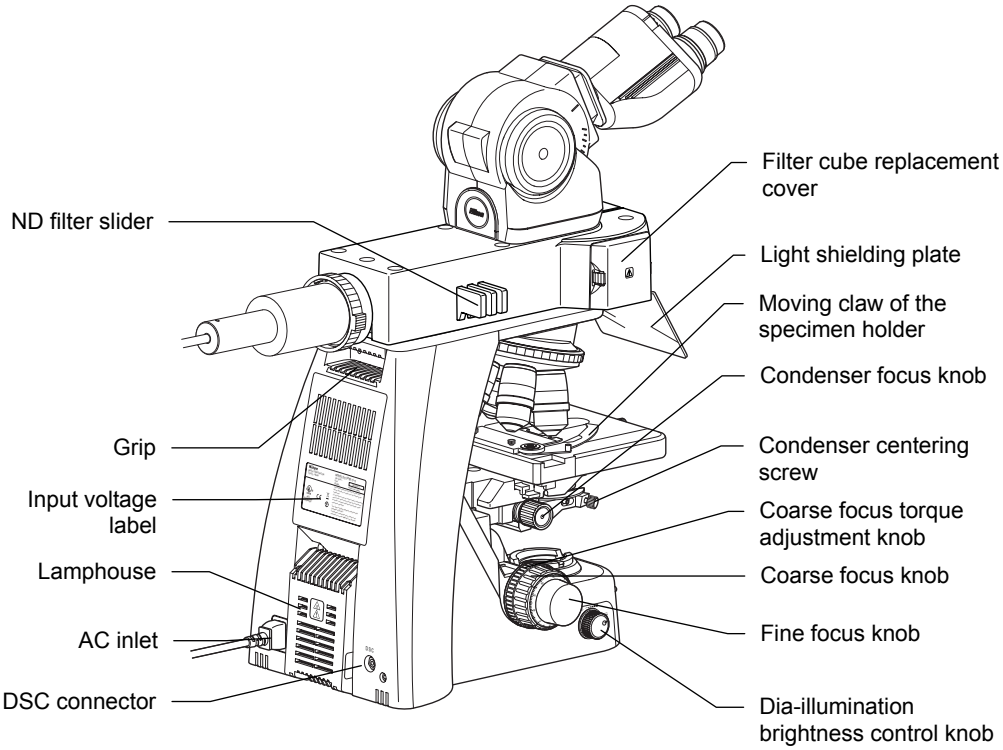
Turn off the power switch (press to the “O” position) for the microscope. (The power LED on the front of the main body will turn off.)

# 5 Epi-fluorescence Microscopy

## 5.1 System Configuration and Controls

This section explains an example system configuration and the controls required for epi-fluorescence microscopy using the ECLIPSE Ci-S/Ci-L.
Names of components are denoted in the following manner: **[CI-FL epi-fluorescence attachment]**.



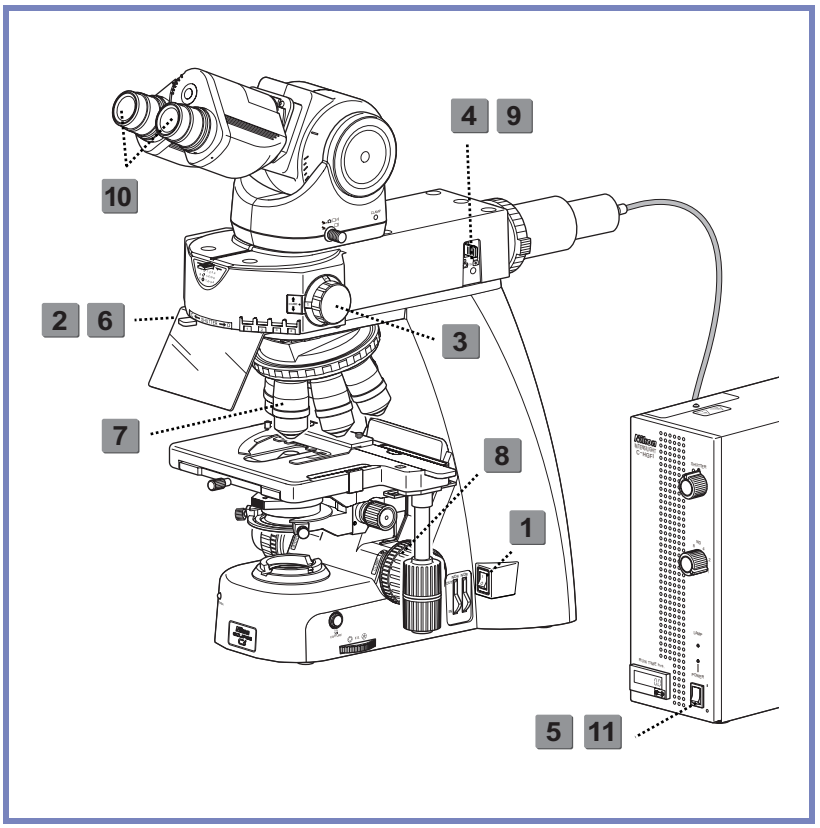
## 5.2 Epi-fluorescence Microscopy Procedure

**⚠ WARNING**

The light source used with the epi-fluorescence attachment (mercury lamp) requires special care during handling because of its characteristics. Make sure you are familiar with and adhere to all warnings and cautions described at the beginning of this instruction manual.

Locate the observation target on the specimen under bright-field microscopy, then proceed to epi-fluorescence microscopy.
(See Chapter 2, “15 Tips for Epi-fluorescence Microscopy” for the tips to locate the observation target on the specimen.)

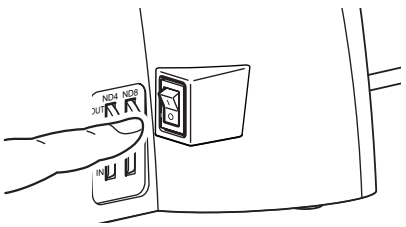
1. **Turn off the dia-illumination power.**
2. **Close the shutter.**
3. **Bring the filter cube into the optical path.**
4. **Fully open the field diaphragm.**
5. **Turn on the mercury lamp.**
6. **Open the shutter.**
7. **Bring the desired objective into the optical path.**
8. **Focus on specimen.**
9. **Circumscribe field diaphragm to field of view.**
10. **View specimen.**
11. **Turn off the mercury lamp.**




### Microscopy operation (→See also: Chapter 2, “15 Tips for Epi-fluorescence Microscopy”)

#### 1 Turn off the microscope power switch (turn off the dia-illumination).

Turn off the power switch (press to the “O” position) for the microscope. (The power LED on the front of the main body will turn off.)

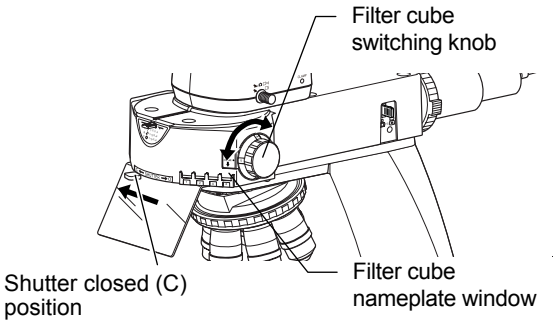


**Turning off the microscope**

## 2 Close the shutter and block the illumination path.

Set the shutter open/close lever of the epi-fluorescence attachment to position “C” to close the shutter and block the optical path.

> **Shutter of the epi-fluorescence attachment**
>
> The shutter blocks illumination.
>
> If the specimen is continuously exposed to the strong light of the mercury lamp, it may become damaged or decolorized.
>
> Be sure to close the shutter when suspending the microscopy or when pausing epi-fluorescence microscopy to perform microscopy with diascopic light. Be sure to get into the habit of performing this operation.




**Closing the shutter**
**Bringing the filter cube into the optical path**

## 3 Bring the filter cube into the optical path.

Bring the desired cube into the optical path by turning the filter cube switching knob.
(Confirm the address of the filter cube to be used, and match the switching knob with the number.)

> **Selecting a filter cube**
>
> A filter cube consists of three types of optical components: an excitation filter (EX filter), a barrier filter (BA filter), and a dichroic mirror (DM). Select the filter cube with the appropriate combination of optical components for the characteristics of the specimen and the fluorescence dye.

## 4 Fully open the field diaphragm of the epi-fluorescence attachment.

Push in the field diaphragm lever of the epi-fluorescence attachment to open the diaphragm fully.

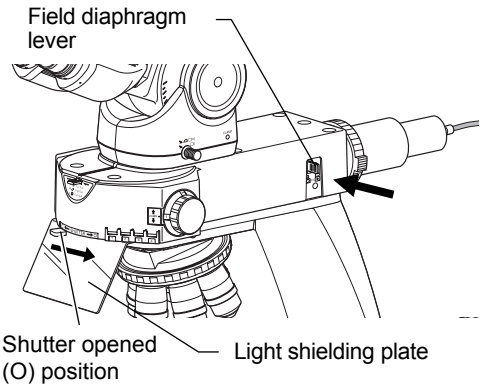
## 5 Turn on the mercury lamp.

See your illuminator's manual for details.

## 6 Open the shutter.

Set the shutter open/close lever of the epi-fluorescence attachment to position “O” to open the shutter.

> **Light shielding plate of the epi-fluorescence attachment**
>
> The light shielding plate protects the observer's eyes from reflected ultraviolet light, which is originally emitted from the objective at the specimen.




**Fully opening the field diaphragm**
**Opening the shutter**

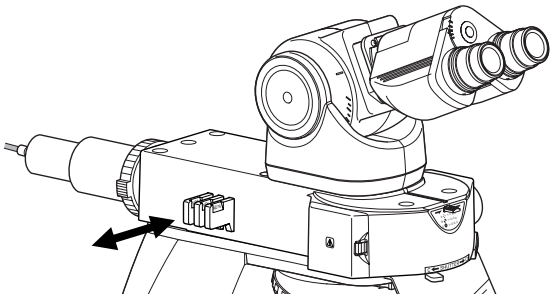
## 7 Bring an arbitrary objective into the optical path.

Turn the nosepiece to bring the desired objective into the optical path.
When using an oil immersion type objective, apply immersion oil between the specimen and the objective. (→Chapter 2 “12 Oil Immersion” for details)

> **Non-fluorescent immersion oil**
> Use Nikon designated non-fluorescent immersion oil.

## 8 Focus on the specimen.

(1) Look into the eyepiece, and adjust the brightness of the field of view with the ND filter of the epi-fluorescence attachment.
(2) Rotate the stage knob to move the stage and bring the target into the optical path.
(3) If the specimen is not in focus, turn the focus knob to focus on it.



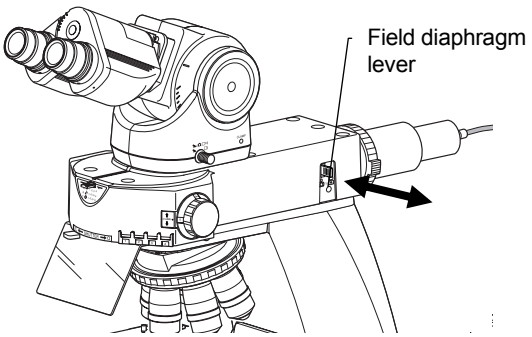
**Adjusting the brightness with ND filters**

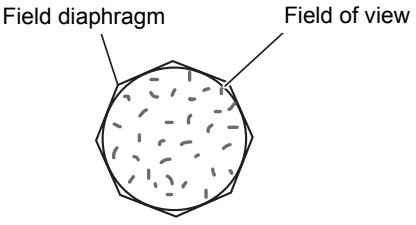
## 9 Adjust the field diaphragm of the epi-fluorescence attachment.

Use the field diaphragm lever of the epi-fluorescence attachment to adjust the field diaphragm so that it almost circumscribes the field of view.

> **Size of the field diaphragm**
> Normally, adjust the field diaphragm so that it almost circumscribes the field of view. Opening the field diaphragm too much results in stray light entering the field of view, generating flare and reducing the image contrast. In addition, the specimen will become decolorized over a wider area.

> **Field diaphragm's adjustment timing**
> Be sure to adjust the field diaphragm each time you change the objective.




Circumscribe around the field of view

**Adjusting the field diaphragm**

## 10 View the specimen.

Rotate the stage knob to move the target. If the target is not in focus, use the focus knob to adjust the focus.

> **Diascopic image in fluorescence observation**
>
> For fluorescence observations, turn off the microscope power switch to make the diascopic image disappear. Bright ambient lights will make it difficult to view the image. Nikon recommends keeping the room dark during fluorescence observations.

> **To return to bright-field microscopy**
>
> - Close the shutter of the epi-fluorescence attachment and block the light emitted by the mercury lamp.
> - Turn on the microscope power switch to turn on the dia-illumination lamp.
> - Turn the filter cube switching knob and bring the position where a filter cube is not located into the optical path.

## 11 Turn off the mercury lamp.

See your illuminator's manual for details.

> **Microscope power switch**
>
> If the microscopy power switch has been turned back ON for bright-field microscopy operation, make sure to turn off the power switch (press to the “O” position). (Make sure that the power LED on the front of the main body is turned off.)

# 2 Individual Operations

## 1 Adjusting the Brightness of a Diascopic Image

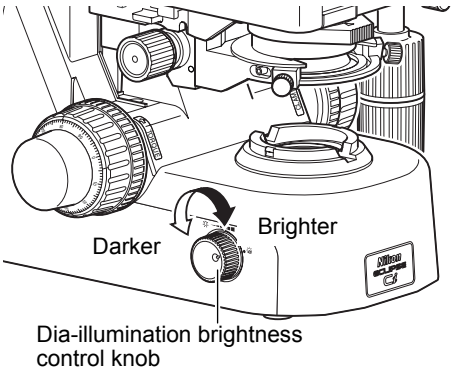
The brightness of a diascopic image can be adjusted by changing the voltage of the lamp with the dia-illumination brightness control knob or by removing/inserting ND filters from or into the optical path.

### 1.1 Adjustment with the Dia-illumination Brightness Control Knob

Turning the dia-illumination brightness control knob changes the voltage of the lamp/LED to change the brightness of the diascopic image. The brightness control knob can be used with click-less operability.

**Brightness Control Knob Rotation and Brightness of the Image**

| Brightness control knob | Image brightness |
|---|---|
| Clockwise rotation | Brighter |
| Counterclockwise rotation | Darker |




**Adjusting the dia-illumination lamp**

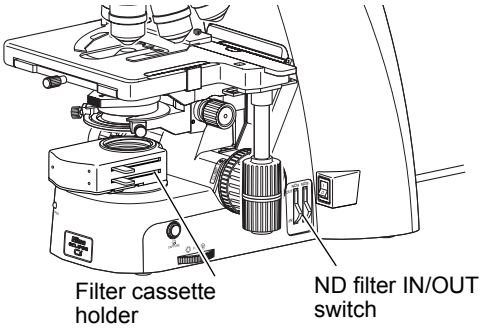
**When using Ci-S (to retain the color balance of the image)**

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 the brightness control knob to the position and use the ND filters for brightness adjustments.

### 1.2 Adjustment with the ND Filter (for Ci-S)

ND filters are used to adjust light intensity. Higher filter numbers correspond to lower transmittance (i.e., darker images). The color balance of the image will not change.
Ci-S has built-in ND filters (ND4 and ND8).
Pushing the each filter IN/OUT switch inserts the corresponding ND filter into the optical path.

ND4: Reduces light intensity to 1/4.
ND8: Reduces light intensity to 1/8.
ND4+ND8: Reduces light intensity to 1/32.




**Adjusting the brightness with ND filters**

**Placing the ND filers over the field lens**

You can add an ND filter by placing the ND filters over the field lens or mounting the filter cassette holder onto the field lens part.

The filter cassette holder can house up to three φ45 mm filters with a thickness of up to 3 mm. Pushing in the filter IN/OUT lever inserts the ND filters into the optical path. In addition, one filter can be placed on the filter cassette holder.

The filter cassette holder cannot be used with a simple polarizer, polarizer unit for first-order red compensation, or the spacer for the nosepiece.

## 1.3 Removing an NCB filter for a brighter image (Ci-S)

Ci-S has an integrated NCB filter (NCB 11) to improve the color reproduction.
If the image still looks dark even after all ND filters are removed from the optical path, removing this NCB filter can make the image brighter.

Follow the procedure below to remove the NCB filter:

(1) Turn off the power switches for the microscope main body and the peripheral devices (press to the “O” position) and remove the power cord from the receptacle.

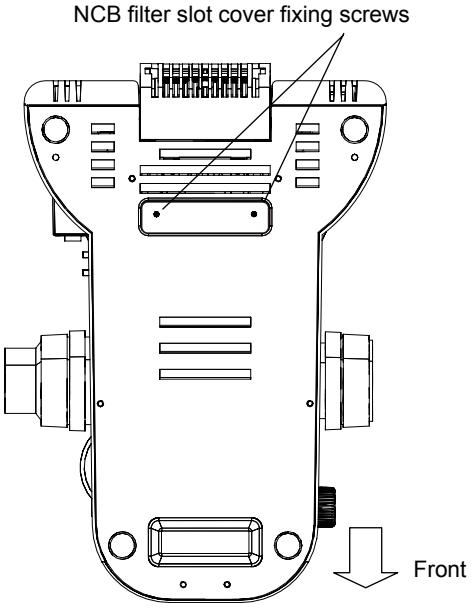
(2) Wait until the lamp and the peripheral devices cool down sufficiently (about thirty minutes).

(3) Remove all accessories such as the camera, objective, nosepiece, eyepiece, tube, condenser, and the stage. See Chapter 3 “Assembly” and use a reverse order of assembling to remove the accessories.

(4) Place the microscope main body upside down with its bottom facing up.

(5) Loosen two NCB filter slot cover fixing screws on the bottom with a tool, remove the cover, and remove the NCB filter along with the filter holder.

(6) Restore the microscope main body in place and follow the procedure in Chapter 3 “Assembly” to attach those accessories back in place.



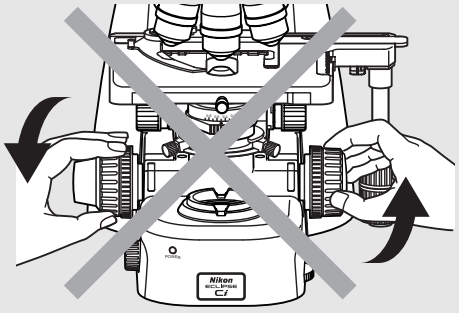

**Remove the NCB filter**

## 2 Focusing on the Specimen (Vertical Stage Movement)

**Note on controlling the focus knobs**

Avoid the following actions, which can cause equipment malfunction.

- Rotating the right and left focus knobs in opposite directions.
- Rotating the coarse focus knob past the limit.



**Don't rotate the knobs in opposite directions!**

Turn the coarse or fine focus knob to raise or lower the stage and shift the focus onto the specimen.

Using an objective of high magnification may cause the specimen to be pushed against the objective, damaging the objective. Follow the procedure below to focus on the specimen to avoid breaking the cover glass or damaging the objective.

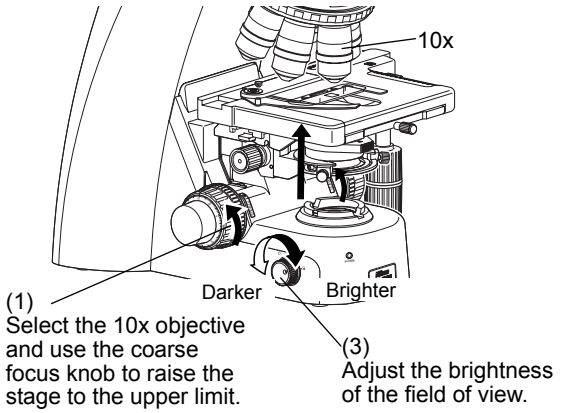
(1) Bring the 10x objective into the optical path and turn the coarse focus knob away to raise the stage to the upper limit.

(2) When using the trinocular tube or ergonomic tube, push in the optical path switching lever to distribute 100% light to the binocular section.

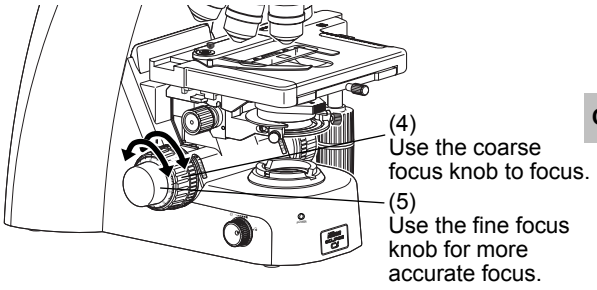
(3) Look into the eyepiece, and adjust the brightness of the field of view by turning the dia-illumination brightness control knob. You can adjust the brightness with an ND filter for Ci-S.

(4) Look into the eyepiece and turn the coarse focus knob slowly to lower the stage and focus on the specimen. Release your hand from the coarse focus knob once it has been focused.

(5) Turn the fine focus knob to adjust the focus more accurately.




**Bringing the 10x objective into the optical path, and raising the stage to the upper limit**




**Using the coarse focus knob to focus→Using the fine focus knob for more accurate focus**

**Raising the stage with the coarse focus knob**

When moving the stage with the coarse focus knob, move your eyes away from the eyepiece and operate the microscope while looking at it from the side.

**Looking into the eyepiece during coarse operation**

When working with the coarse focus knob while looking into the eyepiece, you should only turn the knob in the direction for lowering the stage.

**Switching to an objective of higher magnification**

First use an objective of low magnification to adjust the focus, then switch to an objective of higher magnification.

**Operating distance**

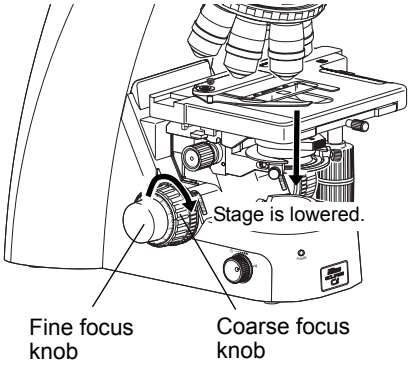
As a 10x or 4x objective has a wider operating distance, the specimen never touches the tip of the objective even if the stage is raised to the upper limit, provided that a slide and a cover glass of standard thickness are used. (Standard thickness being 1.2 mm for the slide glass, and 0.17 mm for the cover glass.)

## 2.1 Focus Knob Rotation and Stage Movement

Both the coarse focus knob and the fine focus knob are located on both right and left sides on the microscope. The table below shows the relationship of the focus knobs' rotation with the stage movement.

**Focus Knob Rotation and Stage Movement**

| Operations | Stage movement |
|---|---|
| Turn the knob toward the front. | Stage is lowered. |
| Turn the knob toward the rear. | Stage is raised. |




**Stage Vertical Movement**

## 2.2 Number of Focus Knob Turns and Distance of Stage Travel

**Number of Focus Knob Turns and Distance of Stage Travel**

| No. of knob turns | Distance of stage travel (vertical direction) |
|---|---|
| One rotation of the coarse focus knob | Approx. 9.33 mm |
| One rotation of the fine focus knob | Approx. 0.1 mm |
| One scale of the fine focus knob | 1 µm |

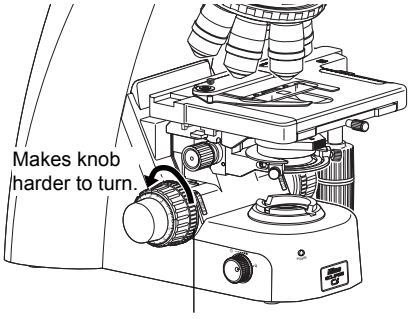
The vertical motion range (coarse/fine focus stroke) of the stage is from approximately 2 mm above the focal point (reference position) to approximately 28 mm below the focal point.

## 2.3 Adjusting the Rotating Torque of the Coarse Focus Knob

Adjust the rotation torque of the coarse focus knob (rotation resistance) by turning the torque adjustment knob (TORQUE) located at the base of the coarse focus knob. If the torque is set too low, the stage may descend under its own weight.

**Adjusting the Rotating Torque of the Coarse Focus Knob**

| Operation of torque adjustment knob | Rotation torque |
|---|---|
| When turned in the direction of the arrow | Rotation torque is increased. |
| When turned in the direction opposite to the arrow | Rotation torque is decreased. |




Coarse focus torque adjustment knob

**Adjusting the torque of the focus knobs**

## 2.4 Refocusing

By turning the coarse focus clamp ring after focusing on the specimen, you can prevent the stage from being raised further with the coarse focus knob. The movement of the stage with the fine focus knob will not be locked.

Using this function, you can refocus with ease by simply turning the coarse focus knob to the limit. This is helpful when switching between similar specimens during the observation.

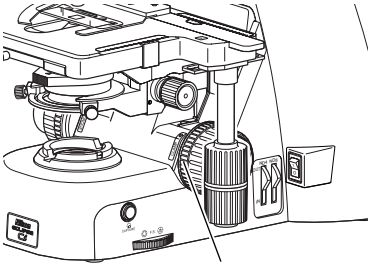
(1) With the focus set on the specimen, tighten the coarse focus clamp ring by turning it approximately 3/4 of a rotation in the direction of the arrow on the base of the microscope. This will clamp the movement of the coarse focus knob.

(2) When replacing the specimen, lower the stage by using only the coarse focus knob.

(3) After replacing the specimen, use only the coarse focus knob to raise the stage slowly until it reaches the upper limit.

At the upper limit, the focus should be more or less on the specimen. Use the fine focus knob for finer adjustment.

If you do not wish to use the refocusing function, be sure to loosen the coarse focus clamp ring to the limit (turn it in the direction opposite to the arrow on the base of the microscope until it hits the limit).



Coarse focus clamp ring

**Refocusing**

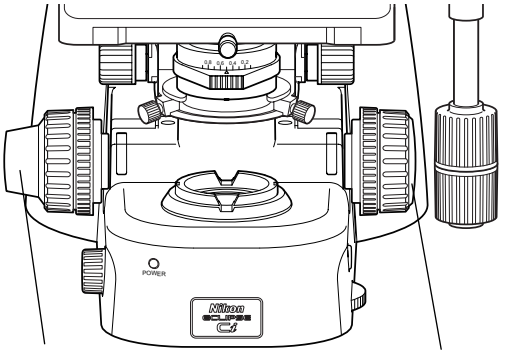
## 2.5 Position Exchange of the Fine Focus Knob

Among left and right fine focus knobs, one is flat and another is convex. Both fine focus knobs are attached to the coarse focus knobs using magnet, so you can detach the left and right knobs from the coarse focus knobs and swap them.

Position them to best suit your usage.

**Removing a flat focus knob**

A flat fine focus knob can be easily removed by inserting a flathead screwdriver or the likes into the notch in the knob.




Convex fine focus knob Flat fine focus knob

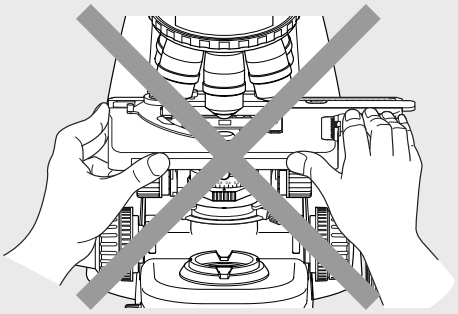
**Position Exchange of the Fine Focus Knob**

## 3 Bringing the specimen into the optical path (Horizontal Stage Movement)

**Note on moving the stage**

Avoid the following action, which can cause equipment malfunction.

- Moving the stage to the right and left by holding the top surface of the stage directly.



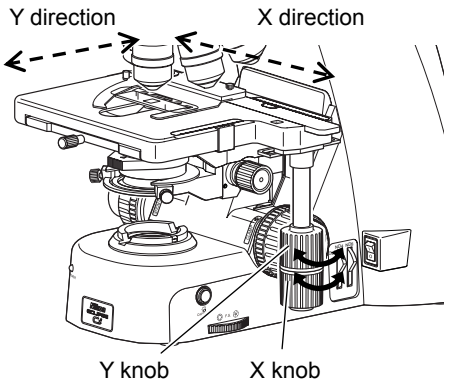
**Do not move the top surface of the stage by hand!**

Turning the stage knob moves the upper plate of the stage in the X and Y directions so that you can move the stage to bring the target into the optical path.

This illuminates the sample sealed under the cover glass.

### 3.1 Knob Rotation and Stage Movement

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




**XY Stage Movement**

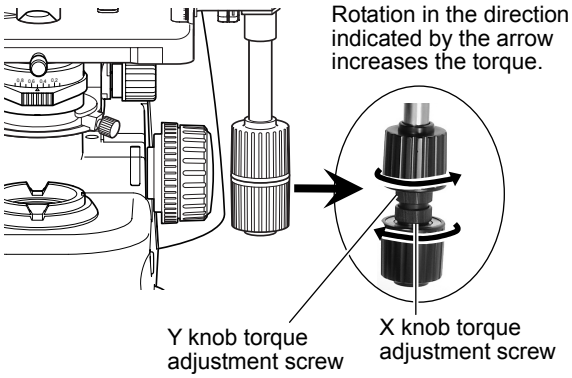
### 3.2 Adjusting the Knob Heights

The heights (positions) of the X knob and Y knob can be changed. Hold the knob and move it along the vertical axis to the desired height.

### 3.3 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 it closer to the respective knob increases rotational torque. (To increase the rotational torque, turn the adjustment screw counterclockwise for the Y knob and clockwise for the X knob, as viewed from above.)

Avoid loosening these screws excessively. If they are too loose, the top surface of the stage may move, even when touched very lightly.




**Adjusting the torque of XY knobs**

## 4 Adjusting the Diopter

The diopter adjustment ring on an eyepiece can be adjusted to match the eyesight of your right and left eyes.

A properly adjusted diopter compensates for differences in visual acuity between the right and left eyes of a person, making binocular observation easier. It also minimizes focal deviations when switching magnification, optimizing the performance of the objective.

Adjust the diopter settings for both eyepieces.

> **Notch on eyepiece**
> The eyepiece has a notch to prevent the rotation. When attaching, match the notch with the protrusion on the eyepiece sleeve. Otherwise, eyepiece is not attached to the correct position.

(1) Turn the diopter adjustment ring on the right and left eyepieces to align the end face of the diopter adjustment ring with the line. (This is the diopter adjustment reference position.)

(2) Follow Steps 1 through 6 in Chapter 1 “1.2 Bright-field Microscopy” to focus on the specimen using the 10x objective.

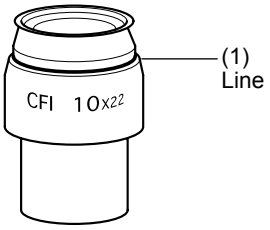
(3) Turn the nosepiece to bring the 40x objective into the optical path, and turn the coarse focus knob and then the fine focus knob to focus on the specimen.

(4) Bring the 10x (or 4x) objective into the optical path.

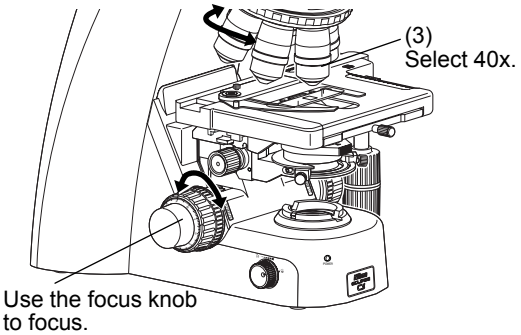
(5) Look into the left eyepiece with your left eye. Without touching the focus knob, focus on the specimen by turning the left diopter adjustment ring.

(6) Look into the right eyepiece with your right eye without touching the focus knob to focus on the specimen by turning the right diopter adjustment ring.

(7) Repeat Steps (3) through (6) to make sure the focus has been adjusted properly.




**Reference position for diopter adjustment**




**Adjusting the Diopter**

## 5 Adjusting aperture diaphragm

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

(1) Follow Steps 1 through 6 in Chapter 1 “1.2 Bright-field Microscopy” to focus on the specimen using the 10x objective.

(2) Turn the field diaphragm dial counterclockwise fully to minimize the field diaphragm.

(3) You can see the diaphragm image in the field of view as you look into the eyepiece. Use the condenser focus knob to adjust so that the field diaphragm image can be outlined clearly.

(4) Adjust the condenser centering screws until the field diaphragm image is at the center of the eyepiece field of view.

(5) Set the 40x objective into the optical path. Bring the 40x objective into the optical path. As you see the field diaphragm image, if the outline of the image is out of focus, use the condenser focus knob to adjust the focus as much as possible.

(6) Turn the field diaphragm dial to adjust so that the field diaphragm image size is almost the same as the field of view.

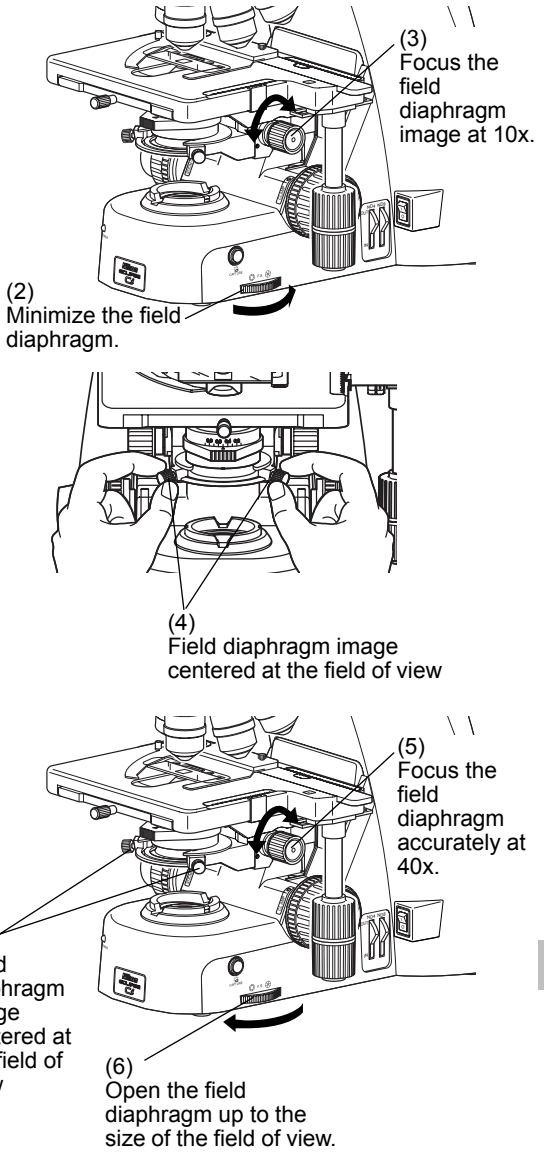
(7) When the center of the field diaphragm image is not centered, turn the condenser centering screws to move the field diaphragm image to the center of the field of view. This is easiest if you adjust the field diaphragm aperture so that it is slightly smaller than the eyepiece field of view.

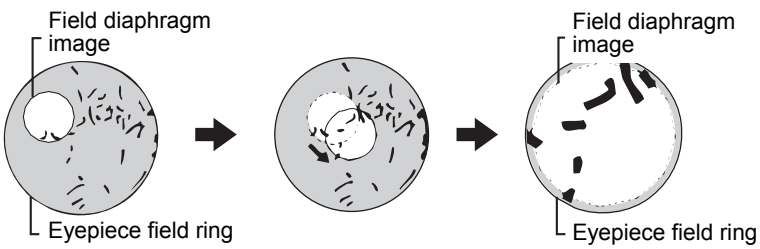
**Correct focusing of the field diaphragm image**

If the outline of the field diaphragm image is reddish or bluish, you have turned the condenser focus knob too much. When the outline is colorless, focusing is correct.

**Field diaphragm image view with the 40x objective**

The field diaphragm image that has been focused with the 40x objective can not be seen so clearly as the one focused with the 10x objective.




**Focusing and centering condenser**

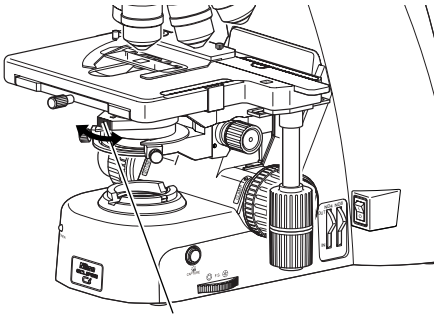
## 6 Adjusting the Aperture Diaphragm

The aperture diaphragm is used to adjust the illumination angular aperture and is important as it affects the resolution, contrast, focal depth, and brightness of an optical image.

Turning the condenser aperture diaphragm lever changes the size of the aperture diaphragm.

Generally, aperture settings at 70 to 80% of the numerical aperture of the objective will provide satisfactory images with suitable contrast.

A small aperture diaphragm opening reduces resolution and brightness but increases contrast and depth of focus. On the contrary, 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.




**Adjusting the aperture diaphragm**

**Relationship of the aperture diaphragm size with the optical image's state**

| Aperture diaphragm | Resolution | Brightness | Contrast | Focal depth |
|---|---|---|---|---|
| Stop down | Lower | Darker | Larger | Deeper |
| Open | Higher | Brighter | Lesser | Shallower |

**Proper size of the aperture diaphragm**

Normally, 70 to 80% of the numerical aperture of the objective is the proper size. 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 numerical aperture of the objective.

**Adjustment timing for the aperture diaphragm**

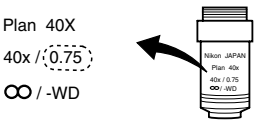
Be sure to adjust the aperture diaphragm each time you change the objective.

## 6.1 Adjusting the Aperture Diaphragm Using the Condenser Scale

The scale on the condenser indicates the numerical aperture. The index on the aperture diaphragm lever should be aligned with the scale line that corresponds to 70 to 80% of the numerical aperture of the objective.

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

For a numerical aperture of 0.75, the index should be aligned with the scale on the condenser at 0.525 to 0.6.




**Indication for 40x magnification / numerical aperture 0.75**

Proper numerical aperture: 0.75 x 0.7 to 0.8=0.525 to 0.6

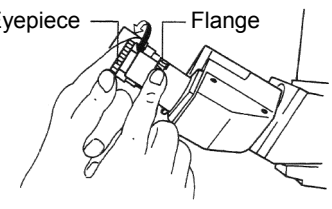
## 6.2 Adjusting the Aperture Diaphragm Using the Centering Telescope

(1) Remove one eyepiece and attach the centering telescope in place, using the adapter.

(2) Turn the aperture diaphragm lever 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.

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

(4) Remove the centering telescope and adapter and reattach the eyepiece.




**Adjustment using the centering telescope**

## 7 Selecting a Condenser

Select a condenser optimal for the magnification of the objective and the microscopy procedure.

**Selecting the Magnification of the Objective and Condenser**

| Objective's magnification | Condenser (⊙: Optimum, ○: Suitable, ×: Not suitable) | | | | | |
|---|---|---|---|---|---|---|
| | Achromat aplanatic condenser | Swing-out condenser | Achromat condenser | Abbe condenser | Swing-out condenser 1-100x | Slide achro condenser 2-100x *4 |
| 1x | × | × | × | × | ⊙*2 | × |
| 2x | × | ○*2 | × | × | ⊙*2 | ⊙*3 |
| 4x | × | | ○*1 | ○*1 | | |
| 10x to 100x | ⊙ | ○ | ○ | ○ | ⊙ | |

*1: The entire field of view may not be covered if a UW eyepiece is attached.

*2: Swing out the top lens before use. Uneven illumination around the field of view may occur.

*3: For objectives of 10x or higher magnification, pull out the slide. For objectives with 2x or 4x magnification, push in the slide to prevent vignetting.

*4: Do not use the 2-100x slide achromat condenser with an achromat objective or a plan achromat objective.

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

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

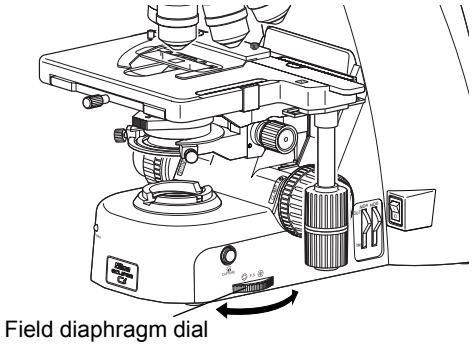
Refer to Section 14 “Tips for the Phase Contrast Microscopy” for the phase contrast condenser.

## 8 Adjusting the field diaphragm

The field diaphragm is used to restrict illumination to the area of the specimen being viewed.

Turning the field diaphragm dial changes the size of the field diaphragm.

For normal observations, the size of the diaphragm should almost circumscribe the field of view.




**Adjusting the field diaphragm**

**Proper size of the field diaphragm**

Usually, the size is optimal when it almost circumscribes the field of view. Opening the field diaphragm too much and illuminating a broader area than necessary will result in stray light entering the field of view, generating flare and reducing the image contrast. In addition, the specimen will become decolorized over a wider area.

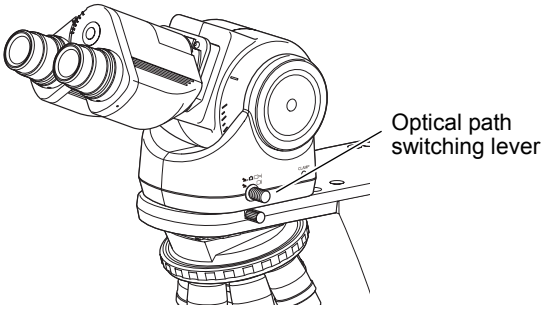
**Field diaphragm's adjustment timing**

Be sure to adjust the aperture diaphragm each time you change the objective.

# 9 Switching the Optical Path of the Tube

## 9.1 Light 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.



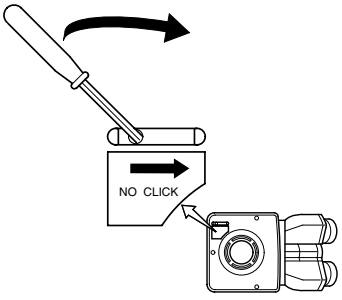

**Switching the Optical Path of the Tube**

**Optical Path Switching Lever and Distribution of Light**

| | Position of the optical path switching lever | Light distribution (%) | |
|---|---|---|---|
| | | Binocular section | Camera port |
| C-TE2 Ergonomic tube | Pushed in | 100 | 0 |
| | Pulled out | 50 | 50 |
| T C-TT trinocular tube | Pushed in | 100 | 0 |
| | Pulled out by one notch | 20 | 80 |
| | Pulled out by two notches | 0 | 100 |
| F C-TF trinocular tube | Pushed in | 100 | 0 |
| | Pulled out | 0 | 100 |

## 9.2 Disabling the Clicking of the Optical Path Switching

C-TT trinocular tube and F C-TF trinocular tube have a “NO CLICK” switch on their tube attaching surface. 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.

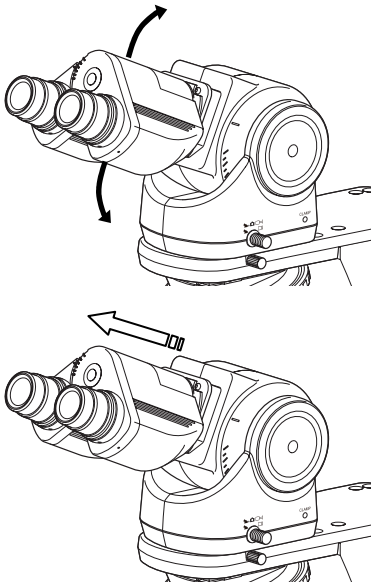



**Disabling the clicking of the optical path switching**

# 10 Adjusting the Binocular Section

## 10.1 Adjusting with the Binocular Section of the Tube

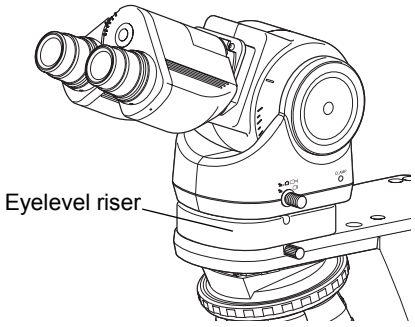
The ergonomic tube makes it possible to tilt and extend the binocular section. Adjust the position of the binocular section for most comfortable viewing.



**Adjusting the binocular section**

## 10.2 Using the Eyelevel Riser

Inserting the eyelevel riser between the tube and the microscope arm allows you to raise the eye point 25 mm higher.




**Using the eyelevel riser**

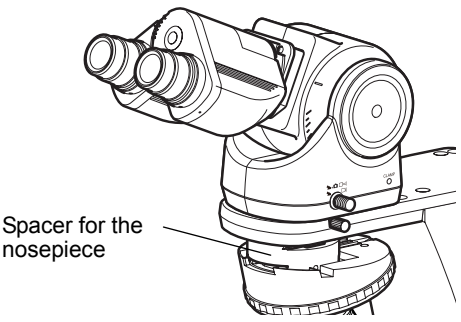
# 11 Using Stage at Lowered Position (Spacer for Nosepiece)

Inserting a spacer for the nosepiece between the arm and the nosepiece allows for operation with the stage set 20 mm lower.

A lowered stage facilitates specimen replacement during a cytodiagnostic examination, etc.

**Using the filter cassette holder not allowed**

The filter cassette holder placed on the field lens is not allowed to use together with the spacer for nosepiece.




**Using the spacer for the nosepiece**

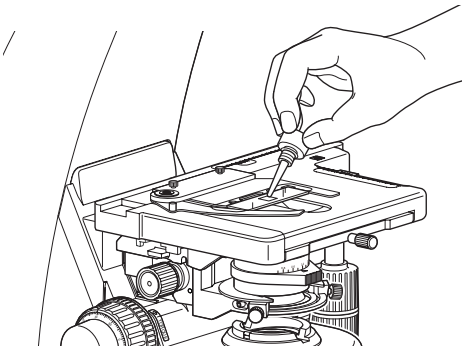
## 12 Oil Immersion

**⚠ CAUTION**

- Petroleum benzine and absolute alcohol are highly flammable. Be careful when handling these materials, especially around open flames and when turning the power switch on or off.
- Follow the instructions provided by the manufacturer when using absolute alcohol.

An objective with an “Oil” marking is an oil-immersion objective. An objective of this type is to be used with designated non-fluorescent immersion oil applied between the sample and the tip of the objective.

For maximum performance, an oil-immersion objective with a numerical aperture of 1.0 or larger should be combined with an oil-immersion achromat aplanatic condenser. The oil-immersion condenser is used by applying a designated non-fluorescent immersion oil between the sample and the condenser.



**Oil immersion**

**Phase turret condenser**

The phase turret condenser is intended to be used dry. Do not apply immersion oil between the condenser and the specimen.

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

**Air bubbles in the oil**

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 with the adapter, then look for air bubbles while turning the eyepiece section of the centering telescope to adjust the 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.

**Cleaning the oil**

Oil remaining on the oil-immersion objective or adhered to the dry-type objective will significantly 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 the oil from the condenser.

Use petroleum benzine to wipe off the immersion oil. For optimum results, Nikon recommends cleaning with absolute alcohol (ethyl or methyl alcohol) after cleaning with petroleum benzine.

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 the immersion oil. (Usually, three or four times should be sufficient to clean the lens.)

## 13 Water Immersion

An objective with a “WI” or “W” marking is a water-immersion objective. Such an objective is 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.

**Cleaning of water**

After use, wipe off the 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.

## 14 Tips for Phase Contrast Microscopy

Phase contrast microscopy is suitable for observation of clear and colorless specimens, undyed or lightly colored specimens, decolored specimens, and ultrathin slices for electron microscopes. The phase contrast method is not suitable for graded or hard dyed specimens.

A phase contrast image appears differently depending on the phase contrast or shape of the specimen, and the properties of the objective. Note the following when you prepare the specimen or select a Ph objective.

### Select a specimen for which the center of the Ph annular diaphragm is not misaligned.

The center of the Ph annular diaphragm will be misaligned when observing a specimen that causes scattered light or generates a lens or prism effect. In particular, with live and thick specimens, oversized specimens, and specimens using a microplate, the center will be misaligned due to the lens or prism effect, making the specimen difficult to observe.

### Ph objective and specimen

Ph objectives include “achromatic”, “plan achromatic”, “plan flour”, and “plan apochromatic” objectives depending on how much the chroma aberration and field curvature are adjusted. These lenses also are further subdivided into several types depending on the properties of the internal phase plate. For favorable microscopy results, the phase contrast amount of the specimen must match the properties of the phase plate. See the table below for the use properties of the Ph objectives.

When using a dark contrast Ph objective, make sure that the phase contrast of the specimen does not exceed the allowed phase contrast amount (latitude). If the phase contrast amount of the specimen is greater than the allowed phase contrast amount, observation is not possible as the image will be illuminated brighter than the background.

You can increase or decrease the phase contrast by the thickness of the specimen, and the refraction index of the mounting agent or culture solution when preparing a phase contrast specimen.

A specimen with weak contrast under a DLL objective may yield better result under a DM objective.

**Use Properties of Ph Objectives**

| Ph contrast objective | | Appearance | Contrast | | Latitude | Usage example |
|---|---|---|---|---|---|---|
| Dark contrast | DLL DL | Generally, an object with larger phase contrast appears darker. Therefore, the image is shown in black in a relatively brighter field of view, similar to the one observed with bright-field microscopy. | Suitable for detailed observation mainly using micro contrast. | Intermediate (with broader usage) | Phase contrast and absorbing object (chromosome) in low and intermediate latitude | Bacteria's spore, general live bacteria, slightly thick specimen, bacteria, dyed specimen, egg, fat particle, crystalline, etc. |
| | DM | | | High (with relatively narrower usage) | Transparent object in low latitude | Bacteria and protozoal flagellum, fibrin basic fiber, fine granule, mounting-agent-selective slice, ultrathin slice, etc. |
| Bright contrast | BM | Generally, an object with larger phase contrast appears brighter. Therefore, the image is shown brighter in a relatively darker field of view, similar to the one observed with dark-field microscopy. | Suitable for morphology, detection, and calculation of fine fiber and granule mainly using macro contrast. | | Almost all areas | Bacteria and protozoal flagellum, fibrin basic fiber, fine granule, blood cell calculation, etc. |

## Using the GIF filter

The GIF filter (green interference filter) improves the contrast when placed in the optical path. The filter should be installed on the field lens, or placed inside or on top of the filter cassette holder. Note, however, that it may cause ghosting when mounted inside the filter cassette holder.

## Phase turret condenser

Phase contrast microscopy requires a phase turret condenser equipped with a Ph annular diaphragm. Matching the Ph annular diaphragm and the phase plate of the Ph objective correctly provides a phase contrast effect.

■ **Ph code**

One of the Ph codes, [Ph1], [Ph2], or [Ph3] is indicated on the Ph objective depending on the size of the phase plate. (Ph codes have nothing to do with the magnification of the objective.) Always use a Ph objective and Ph annular diaphragm with the same Ph code. You cannot experience the phase effect if a different combination of the codes might be used.

■ **Centering of the annular diaphragm and the phase plate**

The position of each condenser diaphragm in the condenser turret have already been adjusted based on the Ph1 annular diaphragm. Usually, once centered with Ph1, further centering is not required upon switching to another magnification. However, the phase contrast image will differ slightly depending on how the annular diaphragm overlaps the phase plate, thus for a stricter observation or capturing of still images, check whether the annular diaphragm and the phase plate are concentric at each magnification. In addition, slightly decentering the annular diaphragm and the phase plate will produce a shadowing effect, resulting in a stereo image. Use this method as appropriate for the specimen.

■ **Infrared ray blocking filter (Ci-S only)**

The lamphouse is equipped with an infrared ray cut filter, which reduces the infrared ray component of the illumination to protect live specimens from being damaged by the heat from the illuminator.

**Notes on using the phase turret condenser**

- Microscopy is possible with an objective of 4x or greater, but UW microscopy with the 4x objective will cause vignetting.
- When the phase turret condenser is set to [A: empty], its performance is equivalent to that of the Abbe condenser.

## Combining with an epi-fluorescence attachment

When the epi-fluorescence and phase contrast attachments are attached to the microscope, it is possible to use epi-fluorescence and phase contrast microscopy in combination. This allows for compensation of each method's shortcomings, for example, by making it possible to find the target using the phase contrast method instead of the epi-fluorescence method that is likely to degrade the color of the specimen or by using both microscopy concurrently.

See Chapter 1 “2.2 Phase Contrast Microscopy Procedure” for the phase contrast microscopy procedure and Chapter 1 “5.2 Epi-fluorescence Microscopy Procedure” for the epi-fluorescence microscopy procedure. When switching between microscopy methods, note the following.

**To switch to epi-fluorescence microscopy**

- Turn the condenser turret until the [C: Shutter] symbol comes to the front.
- Insert the desired excitation filter cube into the optical path.
- Open the shutter for the epi-fluorescence attachment.

**To switch to phase contrast microscopy**

- Close the shutter for the epi-fluorescence attachment and block the excitation light from the epi-fluorescence attachment.
- Turn the filter cube switching knob and move the position without a filter cube into the optical path.
- Bring a Ph objective into the optical path.
- Turn the condenser turret to bring an annular diaphragm with the same Ph code as the objective into the optical path.
- Fully open the aperture diaphragm.

**To use epi-fluorescence and phase contrast microscopy concurrently**

(1) Use phase contrast microscopy to adjust the focus onto the target.
(2) Remove the GIF filter, if installed, on the field lens.
(3) Insert the desired excitation filter cube into the optical path.
(4) Open the shutter for the epi-fluorescence attachment to focus it again.
(5) Adjust the brightness of the fluorescent image using the ND filter of the epi-fluorescence attachment.
(6) Adjust the brightness of the phase contrast image using the ND filter of the microscope. For Ci-L, use the dia-illumination brightness control knob for adjustment.

# 15 Tips for Epi-fluorescence Microscopy

**⚠ WARNING**

The light source used with the epi-fluorescence attachment (mercury lamp) requires special care during handling because of its characteristics. Make sure you are familiar with and adhere to all warnings and cautions described at the beginning of this instruction manual.

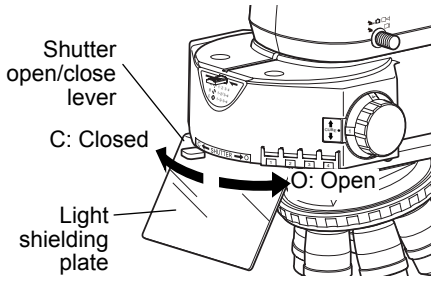
## Protecting the specimen and preventing it from decoloration (shutter for the epi-fluorescence attachment)

The shutter blocks illumination.

Set the shutter open/close lever on the epi-fluorescence attachment to position “C” to close the shutter and block the optical path.

If the sample is continuously exposed to the strong light of the mercury lamp, it may become damaged or decolorized.

Be sure to close the shutter when suspending the microscopy or when pausing epi-fluorescence microscopy to perform microscopy with diascopic light. Be sure to get into the habit of performing this operation.




**Opening/closing the shutter**

## Protecting from ultraviolet light (light shielding plate)

The light shielding plate is used to prevent the reflected ultraviolet light from entering the observer's eyes, which is originally emitted through the objective, from the specimen.

## Using a non-fluorescent slide, cover glass, and immersion oil

For fluorescence observations, be sure to use a non-fluorescent slide, cover glass and our designated immersion oil for an image with better contrast.

## Restricting the illumination to the area of the specimen being viewed (adjusting the field diaphragm)

The field diaphragm is used to restrict illumination to the area of the specimen being viewed.

Turning the field diaphragm lever of the epi-fluorescence attachment changes the size of the field diaphragm.

For normal observations, stop down the diaphragm so that the aperture boundaries circumscribe or inscribe the field of view. Opening the field diaphragm too much and illuminating a broader area than necessary will result in stray light entering the field of view, generating flare and reducing the image contrast. In addition, the specimen will become decolorized over a wider area.

Be sure to adjust the field diaphragm each time you change the objective.

## Adjusting the brightness of the fluorescent image (adjusting the ND)

■ **ND filters in the epi-fluorescence attachment**

ND filters are used to adjust light intensity. Higher filter numbers correspond to lower transmittance (i.e., darker images). ND filters do not affect the color balance.

The epi-fluorescence attachment has three ND filters (ND4, ND8, and ND16) built in.

Push in the ND filter IN/OUT levers to insert ND filters into the optical path and darken the fluorescent image.

As shown below, you can combine these three filters to achieve various levels of brightness.

**Light Reduction by Combined ND Filters of the Epi-fluorescence Attachment**

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

(-: Removed from the optical path, ○: Placed into the optical path)

**Improving the S/N ratio (shielding tube)**

To improve the signal to noise ratio (SNR) during epi-fluorescence observations solely using epi-fluorescence microscopy, we recommend removing the condenser and using the shielding tube provided with the epi-fluorescence attachment.

In particular, when combining Ci-L and the epi-fluorescence attachment, excitation light from the Epi-fl attachment may strike the white LED, making it illuminate and resulting in SNR deterioration. To avoid this situation, make use of the shielding tube provided with the Epi-fl attachment or place a plate on the field lens.

■ **ND on HG precentered fiber illuminator**

You can also adjust the light intensity using the ND on the HG precentered fiber illuminator.
For detailed information, refer to the operating manual provided with the HG precentered fiber illuminator.

## Locating a target on the specimen

The standard procedure for epi-fluorescence microscopy is to first locate the target under differential interference contrast or phase contrast microscopy, and then switch to epi-fluorescence microscopy.
To locate the target under dia-illumination bright-field microscopy, you will need to note the following.

- Under dia-illumination bright-field microscopy, start with a 10x objective, and adequately stop down the condenser.
- Gradually increase the magnification. When the target becomes difficult to locate, switch to epi-fluorescence, and use low excitation light.
- You can also use other techniques, such as using the edge of the cover glass to approximate the position of the target.



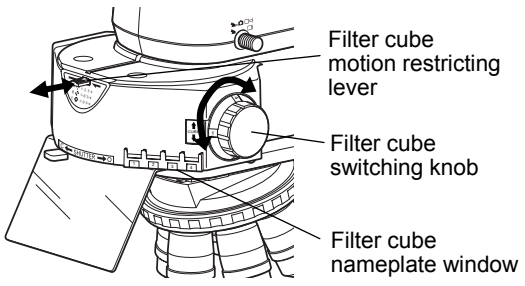
## 15.1 Switching Excitation Methods

Four filter cubes can be inserted into the epi-fluorescence attachment. (→See Chapter 3, “6 Assembly for Epi-fluorescence Microscopy”)

Move the desired cube into the optical path by turning the filter cube switching knob on the right side of the attachment.

For bright-field observations, leave one cube position empty, and move this empty position into the optical path.

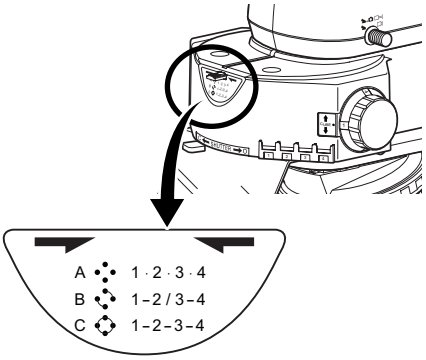
Use the filter cube motion restricting lever located at the upper front section to limit the cube switching operation.




**Switching the filter cube**

**Filter Cube Motion Restricting Lever Functions**

| Lever position | Filter cube switching |
|---|---|
| Position A (push in by two notches) | Locked (filter cubes cannot be switched) |
| Position B (push in by one notch) | Switching between positions 1 and 2, or between positions 3 and 4 only. (The position at which the lever is pushed in determines whether switching is for positions 1 and 2 or positions 3 and 4.)) |
| Position C (first click stop position) | Free (switching possible) |




**Lever position**

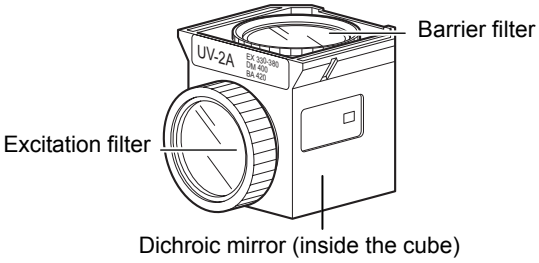
## 15.2 Selecting Filters

A filter cube consists of three types of optical components: an excitation filter (EX filter), a barrier filter (BA filter), and a dichroic mirror (DM). Select the filter cube with the desired combination of optical components to suit the characteristics of the specimen and the fluorophore by referencing the properties of each filter.

You can select a combination of an excitation filter and a barrier filter even if you are using the same excitation method.

Excitation filters, barrier filters, and dichroic mirrors can be purchased separately.

Since excitation filters are exposed to strong light during operations, they will deteriorate over time. Replace the filter at intervals determined by usage.




**Filter cube**

**Spacer inside the filter cube**

Some types of filter cubes cannot be inserted directly into the epi-fluorescence attachment.

Follow the procedure described in Chapter 3, Section 6 “Attaching a filter cube” to remove an internal spacer or reverse the spacer before insertion.

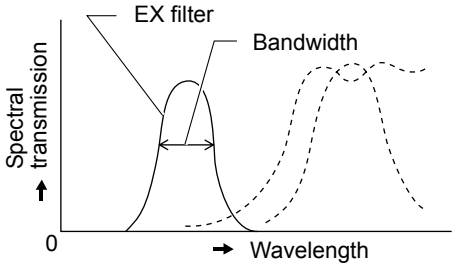


### Excitation filter (EX filter)

Excitation filters allow selective transmission of light (excitation light) in the wavelength range 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 the bandwidth.

The bandwidth range of an excitation filter determines the brightness of the fluorescent image, the generation of autofluorescence (fluorescence resulting from substances other than the fluorophores), and degree of fading. The broader the bandwidth, the greater the amount of excitation light irradiated onto the specimen, thereby increasing the brightness. However, this also increases the amount of autofluorescence 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 autofluorescence and fading. For specimens with pronounced autofluorescence, use excitation filters with a narrow bandwidth. (Note that this will make the fluorescent image darker.)

Since excitation filters are exposed to strong light during operations, they will deteriorate over time. Replace the filter at intervals determined by usage.




**EX filter bandwidth**

**EX Filter Bandwidth and Fluorescent Image**

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

## Barrier filter (BA filter)

Barrier filters allow only fluorescent light emitted by the specimen to pass, blocking excitation light. This allows the fluorescent image to be viewed without excess illumination (dark background).

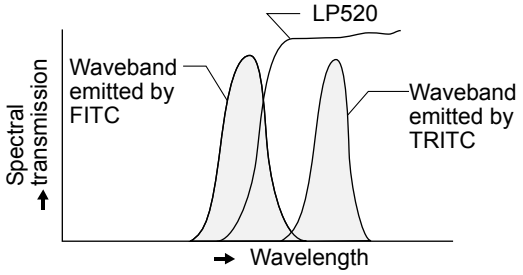
There are two types of barrier filters: LP filters that block all light below a certain wavelength but pass all light of longer wavelengths, and BP filters that pass light of a certain waveband and block all other light. Use the filter type appropriate for your intended purpose.

**LP filter (long-pass filter)**

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

(1) For specimens labeled with a fluorophore in which the fluorescent waveband and excitation waveband (light that the specimen absorbs in order to emit fluorescent light) are very close, selecting a barrier filter with the shortest cut-on wavelength permitted by the performance requirements will result in most 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 developments in filter performance have resulted in increased use of filters of short cut-on wavelengths.

(2) For multiple-labeled specimens, use an LP filter for microscopy of fluorescent images of all fluorophores. Note that a combination involving an ordinary dichroic mirror, an excitation filter, and an LP-filter-type barrier filter will be incapable of excited fluorophores 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.




**Long-pass filter**

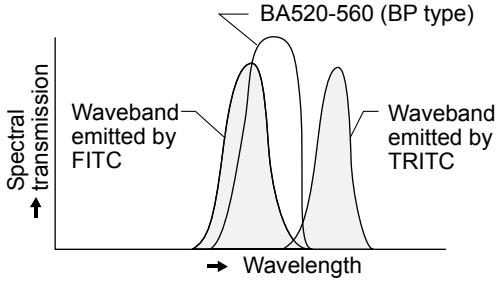
(Both FITC and TRITC images are visible.)

**BP filter (bandpass filter)**

Bandpass filters pass only light of a certain wavelength range, blocking all other light.

BP filters are used for microscopy of fluorescent images involving a specific fluorophore in multiple-labeled specimens. (For example, in a double-labeled specimen of FITC and TRITC, the BA520-560 filter enables microscopy of just the FITC fluorescent image.)

However, BP filters will not separate autofluorescence, if any (because the fluorescent image in the above combination is green only). LP filters are better suited for making fine separation of autofluorescence based on slight color differences.



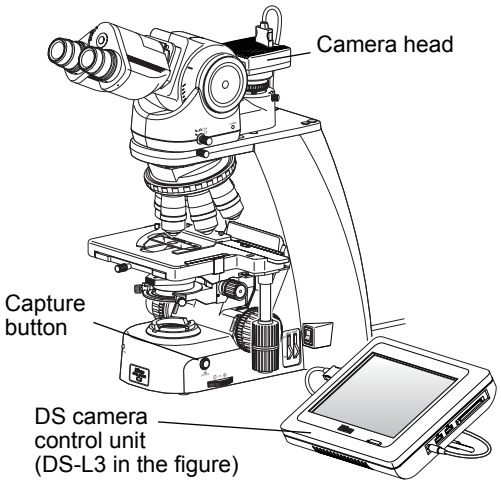

**Band-pass filter**

(Only the FITC fluorescent image is visible.)

# 16 Capturing Images

When the DS-U3 or DS-L3 DS Camera Control Unit is connected to the microscope, you can use the Capture button on the base of the microscope to capture digital images with ease.

When using a tube that allows for light distribution to the binocular section and the camera port, images can also be captured while in an observation posture from the binocular section.




**Photomicroscopy**

## 16.1 Photomicroscopy

The photomicroscopy procedure is described below. Also refer to the instruction manual provided with the DS-U3, DS-L3, or the camera's control software for the details including the camera settings.

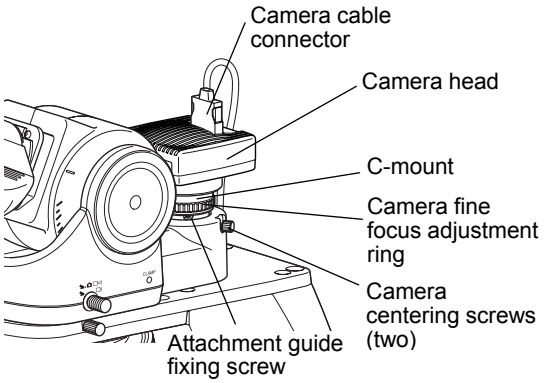
In addition, when using the DS-L3, you must configure at least the following information:

- Folder for data storage.
- File name for saving the file
- File format and file size
- Date and destination of data

**(1) Adjust the illumination of the microscope correctly, and adjust the focus onto the specimen image.**

**(2) Adjust the installation position of the camera head based on the stage movement direction.**

Loosen the attachment guide fixing screw on the C mount, and adjust the camera position and rotation. The movement on the monitor should be in the opposite direction of the stage. (When the stage is moved in the direction from left to right, the image on the monitor should move from right to left.) After making the appropriate adjustments, tighten the screws firmly.




**Adjusting the camera head attachment position**

**(3) Focus the image.**

If the image viewed through the eyepiece appears to be in focus but the image on the monitor is out of focus, turn the camera fine focus adjustment ring on the C mount until the image on the monitor is in focus. Note that such out of focus situations may also indicate incorrect diopter adjustment. Make sure you have made diopter adjustments as well. (→Chapter 2 “4 Adjusting the Diopter”)

**(4) Center the camera.**

Turn the right and left camera centering screws to align the image through the eyepiece with the image on the monitor.

**(5) Select the camera scene mode suitable for the microscopy method.**

**(6) Adjust the camera’s white balance.**

To adjust the white balance, press the WB button while capturing an image of a clear section of the specimen slide. (For fluorescent photomicrography, it is recommended that you adjust the white balance under normal bright-field microscopy conditions before capturing images.)

**NCB filter (for Ci-S only)**
Ci-S has a built-in NCB filter for the color temperature conversion.

**(7) Align the specimen.**

**(8) Readjust the focus onto the target.**

**(9) Adjust the image brightness using the camera exposure compensation function.**

**(10) Check the image using the Freeze button.**

**(11) If the image is acceptable, press the Capture button to save the image.**

(The operating procedure differs if the DF/FL scene mode is selected. For details, refer to the operating manual provided with the camera.)

**Number of images captured**
Seriography is not available because the Capture button is designed to capture only one image.

## 16.2 Tips on Microscope Settings for Photomicroscopy

### Adjusting the light intensity

Lamp/LED: When Ci-S is used in applications for which accurate color reproduction is critical, turn the brightness control knob to the mark and use ND filters for brightness adjustments.

Filter: Place a commercially available color compensation filter on the field lens on the the microscope base, inside or on the filter cassette holder as necessary.

### Adjusting the condenser

- Always focus and center the condenser.
- For phase contrast microscopy, center the annular diaphragm.
- The diaphragm aperture should generally be adjusted to 70 to 80% of the numerical aperture of the objective.

### Confirming the photomicrographic range

The image on the monitor represents the photomicrographic range.

### Confirming the focus

Check the focus both through the eyepiece and on the monitor. If the focal positions for the two images differ, adjust the camera fine focus adjustment ring at the camera port.

### Adjusting 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: Cover the eyepiece with a piece of cloth or similar.

### Protecting fluorescent images from decoloration

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

■ **Use a brighter optical system 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. Nikon recommends increasing the magnification of the objective rather than of the zoom. (Generally, the numerical aperture of the objective increases with magnification. The larger the numerical aperture, the brighter the resulting image.)

■ **Adjusting the excitation light**

Excessively bright excitation light will accelerate the decoloration of the specimen, making it more difficult to acquire suitable fluorescent images. Insert ND filters into the optical path to adjust the brightness.

■ **Specimen**

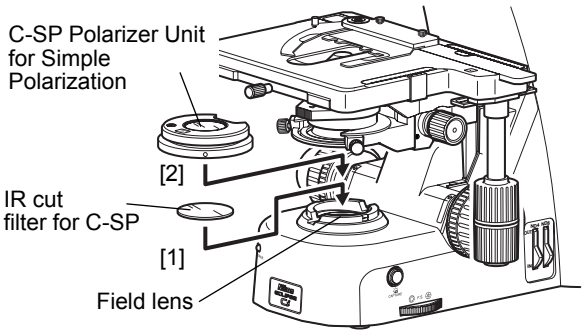
Photomicrography of faded specimen sections requires prolonged exposure time and results in poor color reproduction and low-quality images. Move the specimen to obtain images from a fresh section of the specimen previously unexposed to excitation light. For best results, use the phase contrast method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

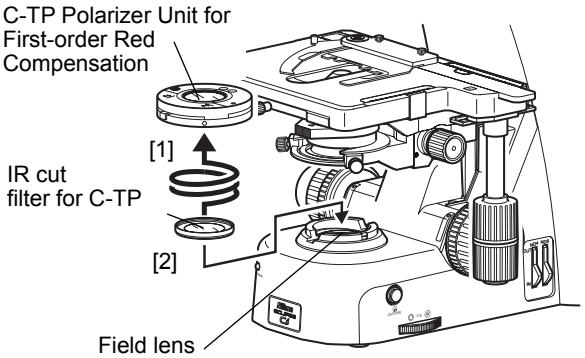
## Enhancing the contrast of a simple or sensitive tint plate image

With Ci-S, since the infrared light from the illuminator reduces the contrast of an image, insert an optional IR cut filter in the field lens before attaching the polarizer unit.

Cover the IR cut filter for the simple polarizer over the field lens.

Screw the IR cut filter for a sensitive tint plate polarizer into the bottom of the polarizer unit for first-order red compensation.




**Attaching an infrared ray blocking filter**

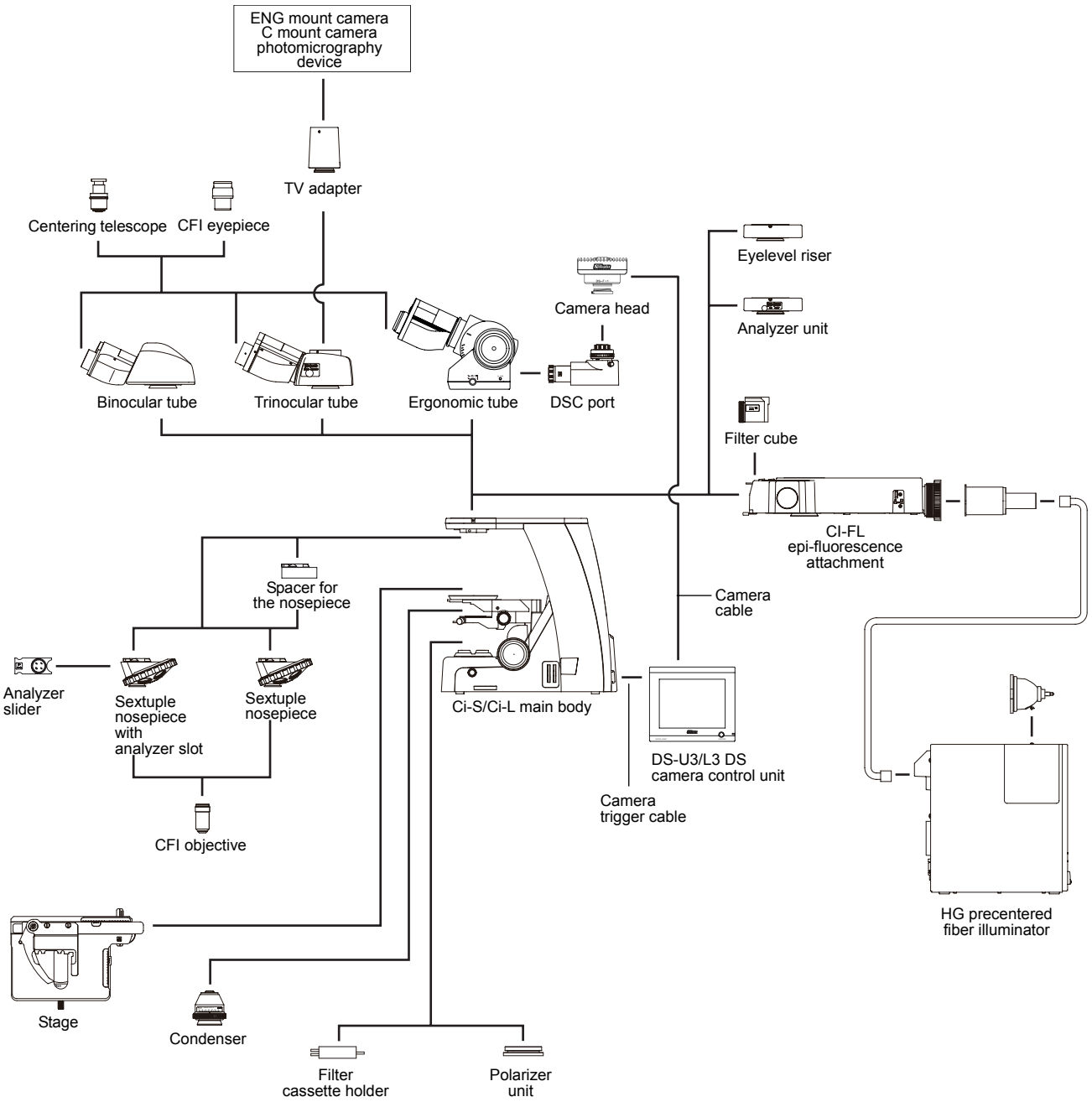
## Adjusting the brightness of the image on the monitor

When observing images captured by the camera and displayed on the monitor, you can adjust the brightness by varying camera adjustment parameters, such as display mode, exposure mode, photometry mode, exposure compensation, and image level adjustment.

See the instruction manual provided with the DS-U3, DS-L3, or the camera's control software for details.

# 3 Assembly

## 1 ECLIPSE Ci-S/Ci-L System Configuration



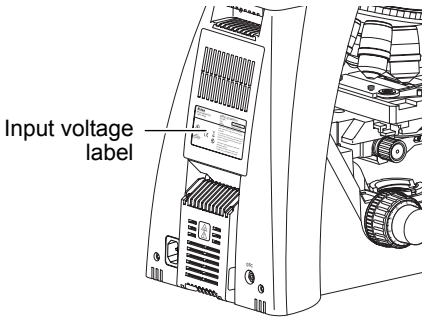
## 2 Assembly for Bright-field Microscopy

[Tool for assembly: hex driver]

### 1 Checking the input voltage

Check the input voltage indicated on the back of the microscope. Use the microscope only if the indicated input voltage matches the power supply voltage for the area in which the microscope will be used.

> **⚠ WARNING**
> If the indicated voltage and the supplied voltage differ, do not attempt to use the microscope. Contact your nearest Nikon representative for advice.



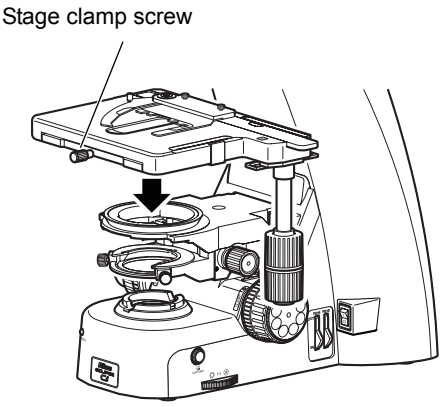

**Checking the input voltage**

### 2 Attaching the stage

(1) Turn the coarse focus knob to remove the cushioning material from the substage section.

(2) Turn the coarse focus knob until the elevating section is brought to the lowermost position.

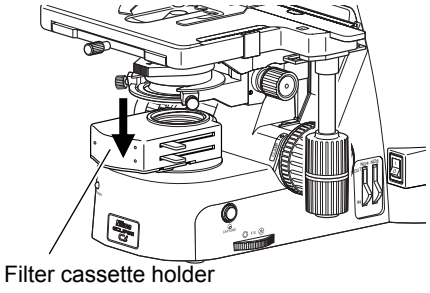
(3) Place the stage on the substage and fix into place with the stage clamp screw.




**Attaching the stage**

**Using the filter cassette holder**

Insert the filter cassette holder on the field lens of the microscope after the stage has been raised up to the upper limit.




**Attaching a filter cassette holder**

**Exclusivity of the filter cassette holder and the spacer for the nosepiece**

The filter cassette holder cannot be used simultaneously with the spacer for the nosepiece.

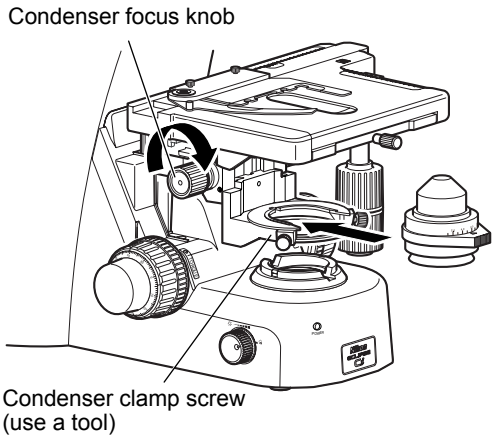
## 3 Attaching the condenser

(1) Turn the coarse focus knob until the elevating section is brought to the upper limit.

(2) Turn the condenser focus knob until the condenser holder reaches the bottom.

(3) Insert the condenser, adjust it to face the front, then secure it in place with the tool.

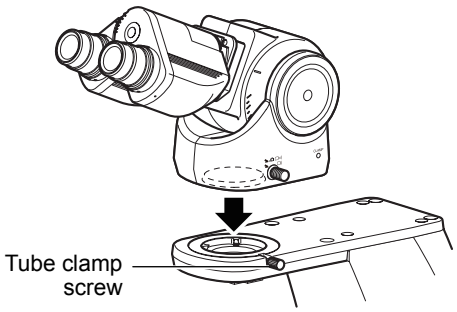
(4) Turn the condenser focus knob until the condenser holder reaches the top.




**Attaching the condenser**

## 4 Attaching the tube

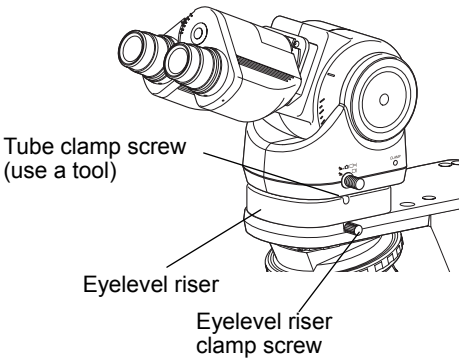
Place the tube on the microscope arm and secure it by tightening the clamp screw on the arm.




**Attaching the tube**

**Using the eyelevel riser**

Place the eyelevel riser on the microscope arm and secure it by tightening the clamp screw on the arm. Attach the tube on the eyelevel riser and secure it by tightening the clamp screw on the eyelevel riser using a tool.



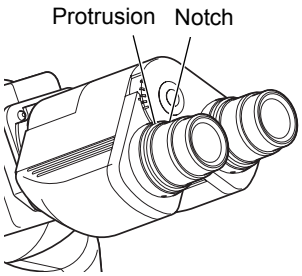

**Attaching the eyelevel riser**

## 5 Attaching eyepieces

Make sure the notch on the eyepiece side and the protrusion of the eyepiece sleeve are aligned, then insert the eyepieces.

**Notch on eyepiece**

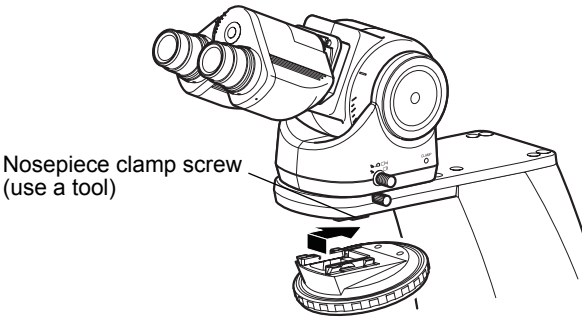
Eyepiece has a notch to prevent rotation. When attaching, match the notch with the protrusion on the eyepiece sleeve. Otherwise, eyepiece is not attached to the correct position.




**Attaching eyepieces**

## 6 Attaching the nosepiece

Place the nosepiece under the fitting part, at a position slightly toward yourself. (Continue sliding the nosepiece until its front position is aligned with that of the fitting part.) Secure it in place with the tool provided with the microscope.




**Attaching the nosepiece**

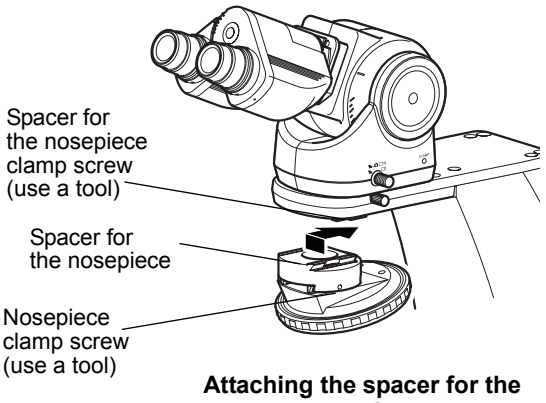
**Using the spacer for the nosepiece**

Follow the procedure below to attach the spacer for the nosepiece.

(1) Slide the spacer for the nosepiece along the groove to attach it to the nosepiece, and secure it in place with the provided tool.

(2) Lower the stage to the limit.

(3) Place the nosepiece with the spacer on the microscope following the procedure described in “Attaching the nosepiece” on the previous page.




**Attaching the spacer for the nosepiece**

**Exclusivity of the spacer for the nosepiece and the filter cassette holder**

The spacer for the nosepiece cannot be used simultaneously with the filter cassette holder placed on the field lens.

## 7 Attaching the objective

Screw the objective into the nosepiece. When attaching the objective in this way, make sure that the magnification of the objective increases when the nosepiece is turned clockwise (clockwise when viewed from above the eyepiece).

## 8 Connecting the power cord

**⚠ WARNING**

Be sure to use the specified power cord. Use of other power cords may result in malfunction or fire. This product is classified as having Class I protection against electric shock. Make sure this product is connected to an appropriate protective earth terminal.

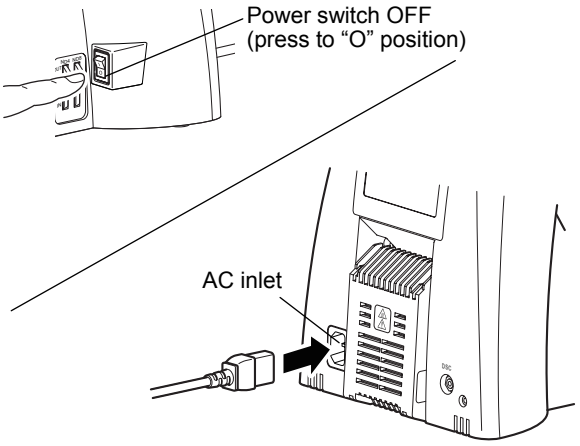
Refer to Chapter 6, “2 Performance Properties” for the specified power cords.

To prevent electric shock, always turn off the power switch (press to the “O” position) for the microscope before connecting or disconnecting the power cord.

(1) Check that the microscope power switch is OFF.

(2) Plug the power cord into the AC inlet at the back of the microscope.

(3) Plug the other end of the power cord into a wall outlet.




**Connecting the Power Cord**

Assembly of the system configuration required for bright-field microscopy is now completed.

## 3 Assembly for Phase Contrast Microscopy

Follow the procedure described in Section 2 “Assembly for Bright-field Microscopy” to perform assembly.

Note the following:

■ **Attaching a condenser for phase contrast microscopy**

Use a condenser for the phase contrast microscopy in the step 3, “Attaching the condenser.”

The procedure is the same.

■ **Attaching a Ph objective**

A Ph objective must be attached in the step 7 “Attaching the objective”.

The procedure is the same.

## 4 Assembly for the Simple Polarizing Microscopy

Follow the procedure described in Section 2 “Assembly for Bright-field Microscopy” to perform assembly.

Note the following:

■ **Attaching an analyzer tube for simple polarization**

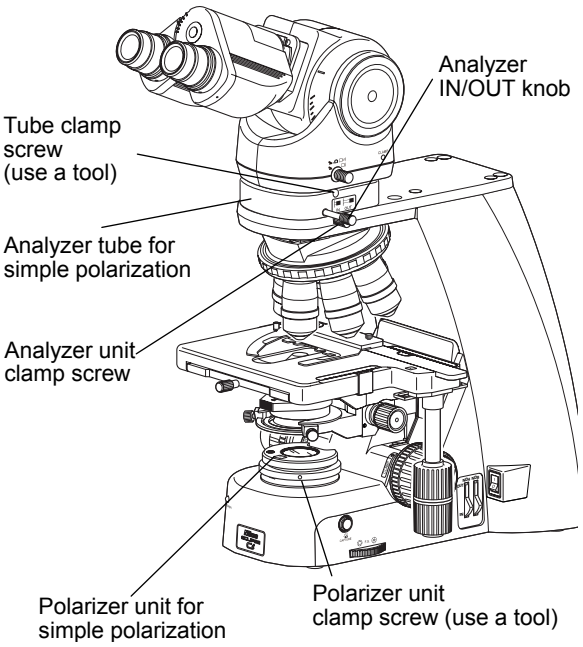
Attach an analyzer tube for simple polarization to the microscope arm prior to the step 4, “Attaching the tube.”

Place the analyzer unit (analyzer tube for simple polarization) on the arm so that the analyzer IN/OUT knob is positioned to the right, and secure the analyzer unit in place with the analyzer unit clamp screw.

Place the tube on the analyzer unit and tighten the tube clamp screws of the analyzer unit using the tool provided with the microscope to secure the tube.

**Slider-type analyzer for simple polarization**

When using the D-SA Analyzer Slider for Simple Polarization instead of an analyzer tube for simple polarization, insert it into the analyzer slot of the nosepiece. (To use the D-SA Analyzer Slider for Simple Polarization, the C-NA sextuple nosepiece with analyzer slot is required.)




**Attaching an analyzer tube for simple polarization, polarizer unit for simple polarization**

■ **Attaching a polarizer unit for simple polarization**

Set the polarizer unit for simple polarization over the field lens on the base of the microscope. Make sure the orientation mark on the polarizer comes to the front at this point and tighten the polarizer unit clamp screw.

The actual securing will take place after the orientation of the analyzer and polarizer has been adjusted in Step 13 of the simple polarizing microscopy procedure.

## 5 Assembly for Sensitive Polarization Microscopy

Follow the procedure described in Section 2 “Assembly for Bright-field Microscopy” to perform assembly.

Note the following:

■ **Attaching an analyzer tube for first-order red compensation**

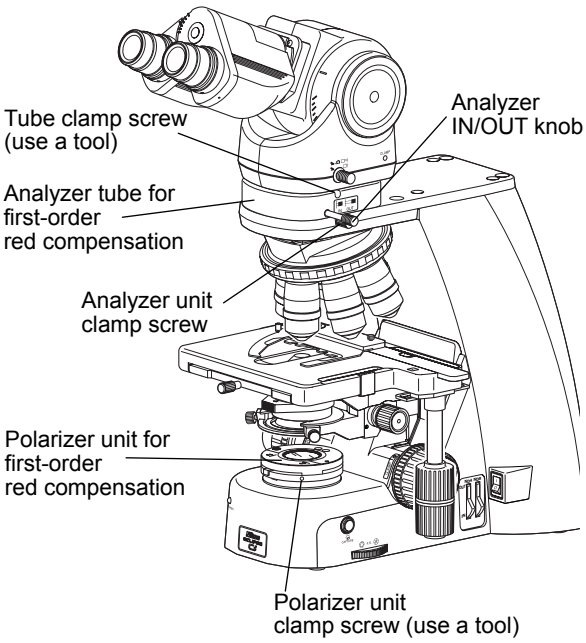
Attach an analyzer tube for first-order red compensation to the microscope arm prior to the step 4, “Attaching the tube.”

Place the analyzer unit (analyzer tube for first-order red compensation) on the arm so that the analyzer IN/OUT knob is positioned to the right, and secure the analyzer unit in place with the analyzer unit clamp screw.

Place the tube on the analyzer unit and tighten the tube clamp screws of the analyzer unit using the tool provided with the microscope to secure the tube.

**Slider-type analyzer for first-order red compensation**

When using the C-AS Analyzer Slider for First-order Red Compensation instead of an analyzer tube for first-order red compensation, insert it into the analyzer slot of the nosepiece. (To use the C-AS Analyzer Slider for First-order Red Compensation, the C-NA sextuple nosepiece with analyzer slot is required.)




**Attaching an analyzer tube for first-order red compensation, polarizer unit for first-order red compensation**

■ **Attaching a polarizer unit for first-order red compensation**

Set the polarizer unit for first-order red compensation over the field lens on the base of the microscope. Make sure the orientation mark on the polarizer comes to the front at this point and tighten the polarizer unit clamp screw.

The actual securing will take place after the orientation of the analyzer and polarizer has been adjusted in Step 13 of the sensitive polarization microscopy procedure.

## 6 Assembly for Epi-fluorescence Microscopy

Follow the procedure described in Section 2 “Assembly for Bright-field Microscopy” to perform assembly.

Note the following:

■ **Attaching the epi-fluorescence attachment**

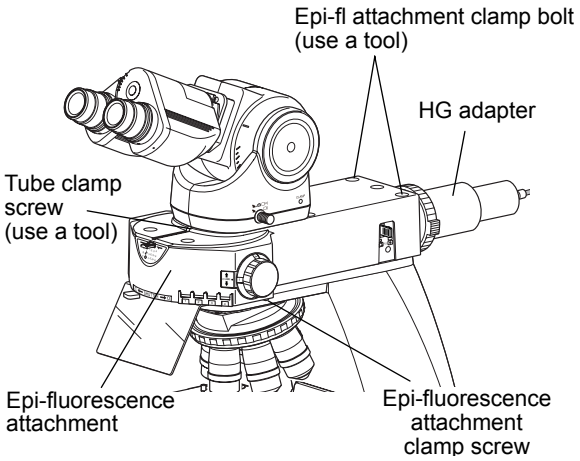
Attach the epi-fluorescence attachment to the microscope arm prior to the step 4, “Attaching the tube.”

(1) Place the epi-fluorescence attachment on the microscope arm.

(2) Secure it by tightening the clamp screw on the arm and two fixing bolts at the rear of the epi-fluorescence attachment. Use a hex wrench provided with the epi-fluorescence attachment to tighten the fixing bolts.

(3) Attach the HG adapter of the mercury lamp illuminator to be used to the bayonet mount on the back. (For detailed information, refer to the instruction manual provided with the mercury lamp illuminator.)

(4) Place the tube on the epi-fluorescence attachment and tighten the tube clamp screws of the epi-fluorescence attachment using the tool provided with the microscope to secure the tube.




**Attaching the epi-fluorescence attachment**

■ **Attaching a filter cube**

> ⚠ **WARNING**
> Always turn off the mercury lamp before inserting or removing a filter cube.

(1) Remove the filter cube replacement cover on the left of the epi-fluorescence attachment.

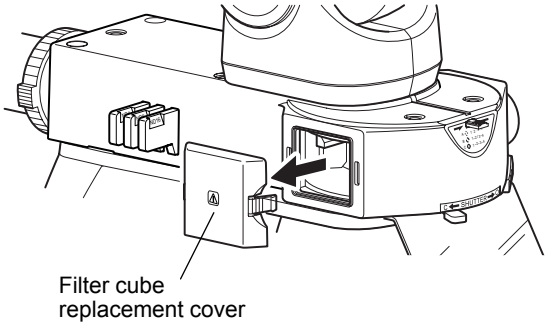
(2) Insert a filter cube.
The filter cubes listed below cannot be inserted directly into the filter bay of the epi-fluorescence attachment. Remove an internal spacer or reverse the spacer before insertion.

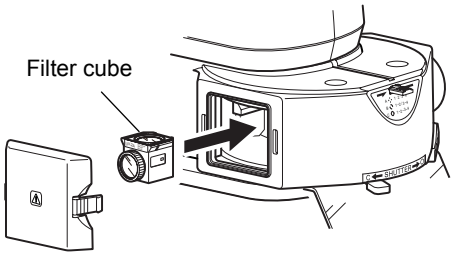
**<Filter cubes requiring spacer removal>**

- UV-2A
- UV-2B

**<Filter cubes requiring spacer reversal>**

- DAPI
- FITC
- GFP-L
- GFP-B
- TRITC
- Tx-Red

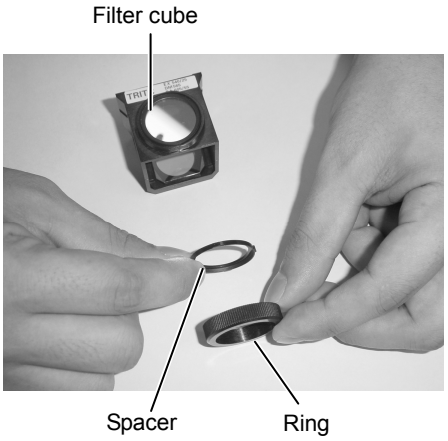



**Attaching a filter cube**

●Removing/attaching the internal spacer●

1) Place the filter cube on a work table with the excitation filter facing up.
2) Unscrew the ring retaining the excitation filter. (Be careful to avoid dropping the filter.)
3) Remove the spacer inside the removed ring.
4) Remove or reverse the spacer as appropriate for the particular filter cube type before its insertion.
5) Reattach the ring.




**Attaching/removing the internal spacer**

(3) Insert a nameplate into the position with the same address as the one indicated on the filter cube switching knob on the right side of the microscope.

(4) Turn the filter cube switching knob and insert other filter cubes into the remaining open bays.

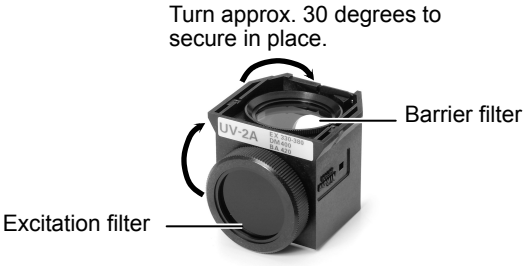
(5) Restore the slot cover back to its original position.

■ **Replacing excitation and barrier filters**

The excitation filter, barrier filter, and dichroic mirror can be removed from the filter cube for replacement.

Excitation filters are screw-in filters, while barrier filters are slide-in filters.

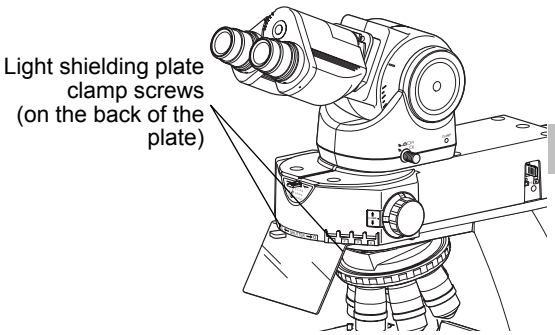
Align the projection on the barrier filter with the groove on the filter cube and turn clockwise by approximately 30 degrees to secure it in place.




**Replacing the excitation and barrier filters**

■ **Attaching a light shielding plate**

Secure a light shielding plate in place with the fixing screws on the lower front of the epi-fluorescence attachment. To remove the plate, loosen the fixing screws and pull it forward.




**Attaching a light shielding plate**

■ **Attaching a shielding tube**

Attach a shielding tube using the same procedure for attaching a condenser to the condenser holder.

## 7 Attaching a Camera

■ **When attaching a camera head to the ergonomic tube**

(1) Screw the camera head into the C mount on the DSC port.

(2) Remove the rear cover of the ergonomic tube and insert the DSC port.

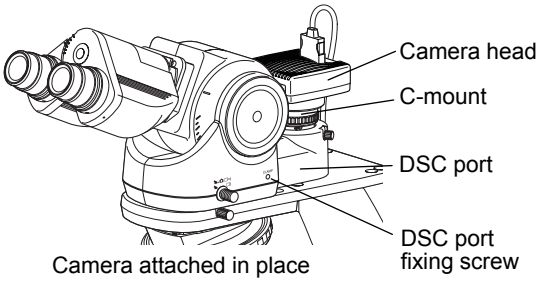
(3) Secure the DSC port in place with the tool provided with the microscope.

(4) Connect the camera head to the DS-U3 or DS-L3 DS Camera Control Unit with a camera cable.

(5) Use a camera trigger cable to connect the DSC connector on the back of the microscope to DS-U3 or DS-L3.

> **Notes when connecting cables**
> When connecting a capture cable to the DSC connector, insert it to the end.

Prior to photomicrography, adjust the camera position as appropriate. (See Chapter 2, “16.1 Photomicroscopy”.)




**Attaching a camera head**

■ **Installing a camera head to the trinocular tube**

Attach a C mount camera, ENG mount camera and a photomicrography device via an adapter to the trinocular tube.

# 4 Troubleshooting

Misuse of this product may adversely affect performance, even if this product is properly functional. If any of the following problems occurs, be sure to check the following table for possible causes before requesting service.

If you detect problems that are not listed below or the problem still persists after measures are taken, turn off the device and contact your nearest Nikon representative.

## 1 Optical System and Operation

### 1.1 General

| Problem | Cause | Measure |
|---|---|---|
| Dirty or dusty field of view when looking into eyepiece. | Dirt or dust rotates when eyepiece is turned.<br>➡ Eyepiece is dirty. | Clean the eyepiece.<br>(→Chapter 5 “2.1 Lens Cleaning”) |
| | Dirt or dust does not rotate when eyepiece is turned ➡ (1) to (5) | |
| | (1) The specimen is dirty if dirt or dust moves when specimen is moved on stage. | (1) Clean the specimen. |
| | (2) The tip of the condenser lens is dirty if dirt or dust goes in and out of view when the condenser is moved up and down while using low magnification objective. | (2) Clean the condenser.<br>(→Chapter 5 “2.1 Lens Cleaning”) |
| | (3) Objective is dirty if dirt or dust disappears when the objective is switched. | (3) Clean the objective.<br>(→Chapter 5 “2.1 Lens Cleaning”) |
| | (4) Field diaphragm image is not focused on the specimen surface. (Condenser is not correctly adjusted.) | (4) Make sure the condenser is focused and centered.<br>(→Chapter 2, “5 Focusing and Centering the Condenser”.) |
| | (5) An aperture diaphragm is stopped down too far. | (5) Open it to proper size.<br>(→Chapter 2 “6 Adjusting the Aperture Diaphragm”) |
| Dirt or dust is displayed on the monitor. | Dirt or dust on the monitor moves when camera is turned<br>➡ Lenses or specimen are dirty or dusty. | Check and clean by following the procedure in “If dirt or dust does not turn by rotating the eyepiece” in “When looking into the eyepiece, dirty or dusty field of view”. |
| | Dirt or dust on the monitor does not move when camera is turned<br>➡ Camera is dirty. | Detach the camera and clean it by following the instruction manual of the camera. |
| Image quality is poor.<br>Contrast is poor.<br>Resolution is poor. | No cover glass is attached. | Attach a cover glass of the specified thickness (0.17 mm). (However, no cover glass is required for an NCG objective.) |
| | The thickness of the cover glass is inadequate. | |
| | Objective correction ring does not match the thickness of the cover glass. (for the objective with a correction ring) | Correct the ring as appropriate. |

| Problem | Cause | Measure |
|---|---|---|
| Image quality is poor.<br>Contrast is poor.<br>Resolution is poor. | Lens and specimen are dirty or dusty. | Check and clean by following the procedure in “If dirt or dust does not turn by rotating the eyepiece” in “When looking into the eyepiece, dirty or dusty field of view”. |
| | An aperture diaphragm is stopped down too far. Otherwise, it is open too much. | Open it to proper size.<br>(→Chapter 2 “6 Adjusting the Aperture Diaphragm”) |
| | Field diaphragm image is not focused on the specimen surface.<br>(Condenser is not correctly adjusted.) | Make sure the condenser is focused and centered.<br>(→Chapter 2, “5 Focusing and Centering the Condenser”.) |
| | No immersion oil is applied to the tip of an oil-immersion objective. | Apply our designated non-fluorescent immersion oil.<br>(→Chapter 2 “12 Oil Immersion”) |
| | The designated immersion oil is not used. | |
| | The immersion oil contains air bubbles. | Remove the air bubbles.<br>(→Chapter 2 “12 Oil Immersion”) |
| | Immersion oil adhering to the tip of the dry-type objective. | Clean it as appropriate.<br>(→Chapter 2 “12 Oil Immersion”) |
| Field of view is too bright. | ND filters are out of the optical path. | Place ND filters in the optical path.<br>(→Chapter 2 “1.2 Adjustment with ND Filters”) |
| | The lamp voltage is too high. | Turn the brightness control knob to the mark position and adjust the brightness with ND filters.<br>(→Chapter 2, “1 Adjusting the Brightness of a Diascopic Image”) |
| Field of view is too dark. | Lamp voltage is too low. | Turn the brightness control knob to the mark position and adjust the brightness with ND filters.<br>(→Chapter 2, “1 Adjusting the Brightness of a Diascopic Image”) |
| | Condenser aperture diaphragm is stopped down too far. | This should normally be adjusted to 70 to 80% of numerical aperture of the objective.<br>(→Chapter 2 “6 Adjusting the Aperture Diaphragm”) |
| | Field diaphragm image is not focused on the specimen surface. | Make sure the condenser is focused and centered.<br>(→Chapter 2, “5 Focusing and Centering the Condenser”.) |
| | Optical path is not switched to binocular 100%. | Set to binocular 100%.<br>(→Chapter 2, “9 Switching the Optical Path of the Tube”) |
| | The lamp has reached the end of its product life (for Ci-S). | Replace the lamp.<br>(→Chapter 5 “1 Replacing the Lamp (for Ci-S)”) |
| Image is yellowish or very bluish. | Lamp voltage is too low or too high. (for Ci-S) | Turn the brightness control knob to the mark position and adjust the brightness with ND filters.<br>(→Chapter 2, “1 Adjusting the Brightness of a Diascopic Image”) |
| Color of the image visible to the naked eye is different from that of the image displayed on the monitor. | White balance of the camera is not set properly. | Set the white balance according to camera's instruction manual. |

| Problem | Cause | Measure |
|---|---|---|
| The entire field of view is bluish or yellowish. | Filter cube is in the optical path when non epi-fluorescence observation. | Remove the filter cube from the optical path. |
| Lack of visibility around periphery of field of view.<br>Illumination is uneven across the field of view.<br>Field of view is not visible. | Parts are attached incorrectly. | Confirm that parts (nosepiece, condenser, etc.) are correctly attached.<br>(→Chapter 3, “2 Assembly for Bright-field Microscopy”) |
| | Movable parts are not seated correctly. | Correctly set the optical path switching lever, nosepiece, filter cube switchover turret, condenser turret, and slider, etc. (Move the part until it clicks.) |
| | Field diaphragm image is not focused on the specimen surface. | Make sure the condenser is focused and centered.<br>(→Chapter 2, “5 Focusing and Centering the Condenser”.) |
| Lack of visibility around periphery of field of view.<br>Illumination is uneven across the field of view.<br>Field of view is not visible. | Field diaphragm is stopped down too far. | Open the field diaphragm slightly wider than the field of view.<br>(→Chapter 2 “8 Adjusting the Field Diaphragm”) |
| | Incorrect combination of the objective with the condenser. | Adopt an appropriate combination.<br>(→Chapter 2 “7 Selecting a Condenser”) |
| | In low magnification microscopy, required condenser operation was not performed. | Perform the operation such as swinging out or sliding the condenser. |
| | A lamp is attached incorrectly. | Attach it correctly.<br>(→Chapter 5 “1 Replacing the Lamp (for Ci-S)”) |
| | Lens and specimen are dirty or dusty. | Clean them as appropriate.<br>(→Chapter 5 “2.1 Lense Cleaning”) |
| Out of focus with an objective of high magnification. | The specimen is upside down. | Turn up the cover glass and attach it to the stage.<br>(→Chapter 2 “2.1 Bright-field Microscopy Procedure - 5 Place a specimen on the stage, and move the stage to bring the target into view”) |
| | The thickness of the cover glass is inadequate. | Attach a cover glass of the specified thickness (0.17 mm). |
| | Fail-safe device for specimen damage protection of the objective is pushed in. | Some objective has a stopper to keep the pushed in state. Turn the tip of the object to release.<br>Tip of the objectives without stopper can not be rotated. Do not use force to draw the stopper. Contact your nearest Nikon representative. |
| A focal deviation is high when switching over objectives. | An objective is attached incorrectly. | Screw the objective all the way in.<br>(→Chapter 3, “2 Assembly for Bright-field Microscopy — 7 Attaching the objective”) |
| | Diopter adjustment has not been performed. | Perform diopter adjustment.<br>(→Chapter 2 “4 Adjusting the Diopter”) |
| Image is not in focus although the stage is raised to the highest position. | The stage is attached incorrectly. | Attach it correctly.<br>(→Chapter 3, “2 Assembly for Bright-field Microscopy — 2 Attaching the stage”) |
| | Refocusing position is set lower than the focusing position. | Check and reset the settings.<br>(→Chapter 2 “2.4 Refocusing”) |

| Problem | Cause | Measure |
|---|---|---|
| One side of the field of view (up, down, right, or left) is not focused. The image flows (i.e. becomes asymmetrically defocused when moving the focal point) | The nosepiece is not attached correctly, or has not been fully rotated to the click stop position. | Attach it correctly and rotate to the click stop position. (→Chapter 3, “2 Assembly for Bright-field Microscopy — 6 Attaching the nosepiece”) |
| | The specimen is tilted relative to the stage surface. | Position the specimen in place on the stage. (→Chapter 2 “2.1 Bright-field Microscopy Procedure - 5 Place a specimen on the stage, and move the stage to bring the target into view”) |
| | The stage is tilted. | Attach the stage correctly. (→Chapter 3, “2 Assembly for Bright-field Microscopy — 2 Attaching the stage”) |
| | The condenser is tilted. | Attach the condenser correctly. (→Chapter 3, “2 Assembly for Bright-field Microscopy — 3 Attaching the condenser”) |
| Images in left and right eyepieces are not coincident. | Interpupillary adjustment has not been performed. | Perform interpupillary adjustment. (→Chapter 2 “2.1 Bright-Field Microscopy Procedure — 8 Adjust the interpupillary distance”) |
| | Diopter adjustment has not been performed. | Perform diopter adjustment. (→See Chapter 2 “4 Adjusting the Diopter” in the “Operation”.) |
| Eyes become fatigued. | Diopter adjustment has not been performed. | Perform diopter adjustment. (→See Chapter 2 “4 Adjusting the Diopter” in the “Operation”.) |
| | Brightness is inadequate. | Adjust the brightness using the brightness control knob or ND filters to attain a suitable brightness. (→Chapter 2, “1 Adjusting the Brightness of a Diascopic Image”) |
| The specimen does not move smoothly. | The specimen holder is not securely-fixed on the stage. | Fix the holder securely. (→Chapter 2 “2.1 Bright-field Microscopy Procedure - 5 Place a specimen on the stage, and move the stage to bring the target into view”) |
| | Rotating torque of the stage knob is set too heavy. | Adjust to appropriate torque weight. (→Chapter 3 “3.3 Adjusting the Knob Rotation Torque”) |

## 1.2 Epi-fluorescence Microscopy

| Problem | Cause | Measure |
|---|---|---|
| Lack of visibility around periphery of field of view.<br>Illumination is uneven across the field of view.<br>Field of view is not visible. | The filter cube is misaligned. | Push the cube in to the limit.<br>(→Chapter 3, “6 Assembly for Epi-fluorescence Microscopy — ■ Attaching a filter cube”) |
| A fluorescent image is not visible (when the lamp is ON). | The shutter is closed. | Open the shutter.<br>(→Chapter 2, “15 Tips for Epi-fluorescence Microscopy — Protecting the specimen and preventing it from decoloration (shutter for the epi-fluorescence attachment”) |
| | The selection of the filter cube is incorrect. | Use a correct filter cube.<br>(→Chapter 2, “15.2 Selecting Filters”) |
| The fluorescent image is very dark (when the lamp is ON). | ND filters of the epi-fluorescence attachment are in the optical path. | Remove the ND filters from the optical path as necessary.<br>(→Chapter 2, “15 Tips for Epi-fluorescence Microscopy — Adjusting the brightness of the fluorescent image (adjusting the ND) — ■ ND filters in the epi-fluorescence attachment”) |
| | Light intensity is set to too low in the setting of the ND on the HG precentered fiber illuminator. | Adjust it.<br>(→Check your illuminator's manual.) |
| | A halogen light source is used for a dark specimen. | Change the light source to a mercury lamp.<br>(→Chapter 2, “15 Tips for Epi-fluorescence Microscopy — Protecting the specimen and preventing it from decoloration (shutter for the epi-fluorescence attachment”) |
| | Mercury lamp on the HG precentered fiber illuminator has reached the end of its product life. | Replace the lamp.<br>(→Check your illuminator's manual.) |
| | A designated objective is not used at UV or V excitation. | Use a designated objective. |
| | The room is bright. | Make it darker. |
| | The optical path switching lever is not set to 100% light distribution for the binocular section. | Switch the lever position to 100% light distribution for the binocular section.<br>(→Chapter 2, “9 Switching the Optical Path of the Tube”) |
| The fluorescent image quality is poor. | The dia-illumination lamp is on. | Turn off the dia-illumination lamp. |
| | The filter cube being used is not suitable for the specimen. | Use a filter cube suitable for the specimen.<br>(→Chapter 2, “15.2 Selecting Filters”) |
| The contrast of the fluorescent image is poor. | The objective or cover glass is dirty. | Clean it as appropriate.<br>(→Chapter 5 “2.1 Lense Cleaning”) |
| | The immersion oil is fluorescent. | Use our designated non-fluorescent immersion oil.<br>(→Chapter 2, “15 Tips for Epi-fluorescence Microscopy”) |
| | The slide is fluorescent. | Use a non-fluorescent slide.<br>(→Chapter 2, “15 Tips for Epi-fluorescence Microscopy”) |
| | Stray light is entering from the condenser. | Lower the condenser, or remove the condenser and attach a shielding tube. |

## 1.3 Phase Contrast Microscopy

| Problem | Cause | Measure |
|---|---|---|
| Poor contrast. | The Ph annular diaphragm of the condenser does not match the phase plate image of the objective. | Adjust so that they match. (→Chapter 1, “2.2 Phase Contrast Microscopy Procedure — 12 Center the Ph annular diaphragm”) |
| | The Ph annular diaphragm of the condenser and the objective selected do not match. | Put the Ph annular diaphragm with the same Ph code as the objective into the optical path. (→Chapter 1, “2.2 Phase Contrast Microscopy Procedure — 14 Adjust the Ph annular diaphragm in the condenser with the Ph objective to be used.”) |
| | The phase contrast of the specimen is too large. | Change the mounting agent or thickness of the specimen when preparing the specimen. (→Chapter 2, “14 Tips for Phase Contrast Microscopy”) |
| | The type of the Ph objective is not suitable for phase contrast of the specimen. | Use a Ph objective suitable for the specimen. (→Chapter 2, “14 Tips for Phase Contrast Microscopy”) |

# 2 Electrical requirements

## 2.1 General

■ **Power supply**

| Problem | Cause | Measure |
|---|---|---|
| There is no power even though the power switch is on. | The power cord is not connected, or is connected improperly. | Connect this product correctly to the network. (→Chapter 3, “2 Assembly for Bright-field Microscopy — 8 Connecting the Power Cord”) |

■ **Illumination**

| Problem | Cause | Measure |
|---|---|---|
| Lamp does not light. | There is no power supplied. | Plug in the power cord. (→Chapter 3, “2 Assembly for Bright-field Microscopy — 8 Connecting the Power Cord”) |
| | The lamp has burned out (for Ci-S) | Replace the lamp with the specified type. (→Chapter 5 “1 Replacing the Lamp (for Ci-S)”) |
| | The lamp is not attached.(for Ci-S) | Attach a designated lamp. (→Chapter 5 “1 Replacing the Lamp (for Ci-S)”) |

■ **Capture button**

| Problem | Cause | Measure |
|---|---|---|
| The capture button does not function. | Camera trigger cable is not properly connected. | Attach it correctly. (→Chapter 3, “7 Attaching a Camera”) |

## 2.2 Epi-fluorescence Microscopy

| Problem | Cause | Measure |
|---|---|---|
| The mercury lamp does not work. | There is no power supplied. | Plug in the power cord. (→Check your illuminator's manual) |
| | Lamp has burned out. | Replace the lamp with the specified type. (→Check your illuminator's manual) |
| | The mercury lamp is not attached. | Attach a designated lamp. (→Check your illuminator's manual) |
| | The mercury lamp's connector is not connected to the illuminator. | Connect it to the illuminator. (→Check your illuminator's manual) |
| The mercury lamp burns out soon after it is turned on. | The lamp type is incorrect.<br>Lamp is at end of its life. | Replace the lamp with the specified type. (→Check your illuminator's manual)<br>If the lamp burns out immediately after the replacement, please contact your nearest Nikon representative. |

# 5 Maintenance and Storage

## 1 Replacing the Lamp (for Ci-S)

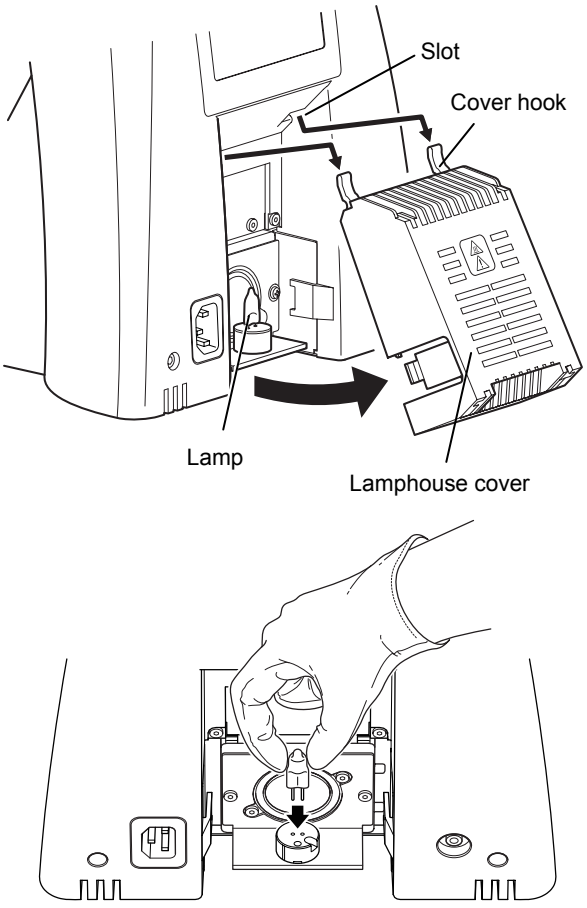
**⚠ CAUTION**

- Beware of burns: Wait until the lamp and nearby parts have cooled before replacing the lamp.
- Beware of electrical shock: Turn off the power switch and unplug the power cord from the wall outlet.
- Beware of abnormal heat generation: Use only the designated lamp.
- Beware of soiling: Avoid touching the glass surface of the lamp with bare hands. Soiling may reduce the service life of the lamp.
- Lamphouse cover: Make sure the lamphouse cover is securely fitted to the lamphouse after lamp replacement.
- Used lamps: Do not break used lamps. It should be disposed of as industrial waste, according to local regulations and rules.

(1) Remove the lamphouse cover on the back of the microscope. Remove the old lamp.

(2) Attach a new lamp. Avoid touching the glass surface of the lamp with your bare hands. Use only the specified lamp (PHILIPS 5761).

(3) Restore the cover back to its original position. Engage the cover hook in the slot on the rear of the unit in the opposite direction indicated by the arrow in the figure.




**Replacing the lamp**

# 2 Cleaning

Follow these procedures when cleaning or decontaminating lenses or other parts.

■ **Tools used for cleaning**

- Blower
- Soft brush
- Soft cotton cloth, lens tissue, or gauze etc.
- Absolute alcohol (ethyl or methyl alcohol), medicinal alcohol
- Petroleum benzine (used only for cleaning immersion oil)

**⚠ CAUTION**

- Petroleum benzine and absolute alcohol used for cleaning are highly flammable. Be careful when handling these materials, particularly around open flames or when turning the power switch on or off.
- Follow the instructions provided by the manufacturer when using petroleum benzine or absolute alcohol.
- When cleaning the product, do not use organic solvents (alcohol, ether, thinner, etc.) for coated, plastic, or printed areas. It will result in discoloration or peeling of printed characters.
- Petroleum benzine should be used only to wipe off immersion oil from the objective, and never to clean the entrance lens at the bottom of the eyepiece tube, prism surface of the eyepiece tube, or the filters.

## 2.1 Cleaning Lenses

Keep the lens free of dust, fingerprints, etc. If there is contamination on the lenses or filters, image quality decreases. If any of the lenses

become dirty, clean them by following the procedure given below.

■ **Cleaning light dirt (such as dust)**

(1) Blow dust off using an air blower.

(2) If this is insufficient, brush away dust with a soft brush or wipe away gently with a piece of gauze.

■ **Cleaning tough dirt (such as fingerprint or grease)**

Moisten lightly a piece of soft, clean cotton cloth, lens tissue, or gauze with absolute alcohol (ethyl or methyl alcohol) and wipe the dirt off.

**Tips on wiping dirt**

Do not reuse cotton cloth, lens tissue, or gauze that have already been used.

## 2.2 Cleaning Parts Other than Lenses

■ **Cleaning light dirt (such as dust)**

Wipe with a silicon cloth.

■ **Cleaning tough dirt (such as fingerprint or grease)**

Dampen a piece of gauze with neutral detergent and wipe the dirt gently.

## 2.3 Cleaning the Immersion Oil

(1) Wipe with petroleum benzine.

(2) Finish off the cleaning with absolute alcohol (ethyl or methyl alcohol) after cleaning with petroleum benzine.

**If petroleum benzine is not available**

If petroleum benzine is unavailable, you may use methyl alcohol. However, typically wipe three or four times because the detergency is weak.

## 2.4 Decontaminating the Product

For routine disinfection of this product, Nikon recommends using 70% medical alcohol.
Use of organic solvents on plastic parts may result in discoloration.

**Note on disposal**

If contact occurs between a sample and this product, determine whether the sample is hazardous. If the sample is hazardous, follow the standard procedures for your facility.

# 3 Storage

- Store this product in a dry location where mold is unlikely to form.
  Storage conditions are as follows: temperature (-20°C to +60°C), humidity (90% RH max., no condensation)
- Store the objectives and eyepieces in a desiccator or similar container with a drying agent.
- Place a cover over this product to protect it from dust.
- Switch off the microscope (press the switch to the “O” position) and wait for the lamphouse to cool before covering this product with a cover.

# 4 Regular Inspections (Charged)

To maintain the performance of this product, Nikon recommends periodic inspections (chargeable service). Contact your nearest Nikon representative for details.

# 6 Specifications and Safety Standards

## 1 Microscopy (Operation Principle)

Use objectives and eyepieces of the microscope to magnify minute cells and tissue optically, and manipulate levers and knobs of the microscope unit to adjust the focus or move the observation point. Then observe or take photographs of the sample fixed on the slide.

■ **Intended use of this product**

This microscope is intended for use in microscopic experiment and diagnostics of cells and tissues at hospitals or by doctors in private practice in the field of pathology, anatomy, and cytology.

The microscopy with diascopic/reflected illuminations is used to observe a sample fixed on the slide (cells and tissue) as the specimen.

The product is classified as an in-vitro diagnostic medical device.

This product is not intended for use for measurement.

The Z axis position display on the display panel at the front of main body, the XY coordinate position display of motorized XY stage, and the scale on the stage is an indicator to reproduce the position and does not guarantee the value of the thickness or length of a sample measured using them.

■ **Intended user**

It is intended for the medical professional and those who work on experimentations in the field of pathology and cytology.

## 2 Performance Properties

■ **Nikon Microscope ECLIPSE Ci-S**

| | |
|---|---|
| Model | ECLIPSE Ci-S |
| Optical system | Infinity-corrected CF optical system<br>Objective CFI60<br>Eyepiece: Field number 22 (with ergonomic tube/binocular tube), 25 (with T/F trinocular tube)<br>Nosepiece: Sextuple |
| Focus up/down motion | Drive system: Manual coarse/fine motion (calibration markings for fine motion: 1 µm/marking)<br>Stroke: 2 mm upward, 28 mm downward<br>With refocusing mechanism |
| Light source for dia-illumination | 6V30W halogen lamp (PHILIPS 5761) |
| Average lamp life | 100 hours |
| Light power supply | 6V30W integrated (100 to 240V) |
| Input ratings | 100-240VAC ±10%, 50/60Hz, 0.8A |
| Power consumption (nominal) | 38W |
| Power cord | • When used in 100-120 V regions outside Japan<br>UL listed detachable power cord set, 3 conductor grounding<br>(3 conductor grounding Type SVT, No.18 AWG, 3 m long maximum, rated at 125 VAC minimum)<br>• When used in 220-240 V regions<br>Detachable power cord set approved in accordance with EU/EN standard, 3 conductor grounding<br>(3 conductor grounding Type H05VV-F, 3 m long maximum, rated at 250 VAC minimum)<br>• When used inside Japan<br>PSE approved detachable power cord set, 3 conductor grounding<br>(3 conductor grounding Type VCTF 3 x 0.75 $mm^2$, 3 m long maximum, rated at 125 VAC minimum) |

■ **Nikon Microscope ECLIPSE Ci-L**

| Model | ECLIPSE Ci-L |
|---|---|
| Optical system | Infinity-corrected CF optical system<br>Objective CFI60<br>Eyepiece: Field number 22 (with ergonomic tube/binocular tube), 25 (with T/F trinocular tube)<br>Nosepiece: Sextuple |
| Focus up/down motion | Drive system: Manual coarse/fine motion (calibration markings for fine motion: 1 µm/marking)<br>Stroke: 2 mm upward, 28 mm downward<br>With refocusing mechanism |
| Light source for dia-illumination | White LED |
| Light power supply | 5V15W integrated (100 to 240V) |
| Input ratings | 100-240VAC ±10%, 50/60Hz, 0.37A |
| Power consumption (nominal) | 6W |
| Power cord | • When used in 100-120 V regions outside Japan<br>UL listed detachable power cord set, 3 conductor grounding<br>(3 conductor grounding Type SVT, No.18 AWG, 3 m long maximum, rated at 125 VAC minimum)<br>• When used in 220-240 V regions<br>Detachable power cord set approved in accordance with EU/EN standard, 3 conductor grounding<br>(3 conductor grounding Type H05VV-F, 3 m long maximum, rated at 250 VAC minimum)<br>• When used inside Japan<br>PSE approved detachable power cord set, 3 conductor grounding<br>(3 conductor grounding Type VCTF 3 x 0.75 $mm^2$, 3 m long maximum, rated at 125 VAC minimum) |

■ **CI-FL Epi-fluorescence Attachment for Nikon Microscope**

| Model | CI-FL epi-fluorescence attachment |
|---|---|
| Optical system | Infinity-corrected CF optical system<br>Variable intermediate magnification: 1x |
| Epi-fl filter turret | Manual quadruple turret |
| Field diaphragm | Manual adjustment |
| ND filter | Manual, 3 filters (slider type; ND4, ND8, ND16) |
| Shutter | Manual (using a lever at the bottom of the turret) |
| Supported illuminator | HG Precentered Fiber Illuminator |

## 3 Physical Properties

■ **Nikon Microscope ECLIPSE Ci-S**

| Model | ECLIPSE Ci-S | |
|---|---|---|
| Operating conditions | Temperature: | 0°C to +40°C |
| | Humidity: | 60% RH max. (no condensation) |
| | Altitude: | 2000 m max. |
| | Pollution degree: | Degree 2 |
| | Installation: | Category II |
| | Electrical shock protection class: | Class I |
| | Indoor use only | |
| Transport/storage conditions | Temperature: | -20°C to +60°C |
| | Humidity: | 90% RH max. (no condensation) |
| External dimensions and weight (Main body only) | External dimensions: | 223 (W) x 331.5 (H) x 331 (D) mm (excluding projections) |
| | Weight: | Approx. 10 kg |

■ **Nikon Microscope ECLIPSE Ci-L**

| Model | ECLIPSE Ci-L | |
|---|---|---|
| Operating conditions | Temperature: | 0°C to +40°C |
| | Humidity: | 60% RH max. (no condensation) |
| | Altitude: | 2000 m max. |
| | Pollution degree: | Degree 2 |
| | Installation: | Category II |
| | Electrical shock protection class: | Class I |
| | Indoor use only | |
| Transport/storage conditions | Temperature: | -20°C to +60°C |
| | Humidity: | 90% RH max. (no condensation) |
| External dimensions and weight (Main body only) | External dimensions: | 223 (W) x 331.5 (H) x 331 (D) mm (excluding projections) |
| | Weight: | Approx. 10 kg |

■ **CI-FL Epi-fluorescence Attachment for Nikon Microscope**

| Model | CI-FL epi-fluorescence attachment | |
|---|---|---|
| Operating conditions | Temperature: | 0°C to +40°C |
| | Humidity: | 60% RH max. (no condensation) |
| | Altitude: | 2000 m max. |
| | Pollution degree: | Degree 2 |
| | Installation: | Category II |
| | Electrical shock protection class: | Class I |
| | Indoor use only | |
| Transport/storage conditions | Temperature: | -20°C to +60°C |
| | Humidity: | 90% RH max. (no condensation) |
| Mass | Approx. 2kg | |