

TE2000

T-FL Epi-FI Attachment
T-FL-E Motorized Epi-FI Attachment

Instructions



Thank you for purchasing the Nikon products.

This instruction manual is written for the users of the T-FL EPI-FL ATTACHMENT and T-FL-E MOTORIZED EPI-FL ATTACHMENT which are to be used together with Nikon's Inverted Microscope ECLIPSE TE2000. To ensure correct usage, read this manual carefully before operating the instrument.

- It is prohibited to reproduce or transmit this manual in part or whole without Nikon's expressed permission.
- The contents of this manual are subject to change without notice.
- Although every effort has been made to ensure the accuracy of this manual, if you note any points that are unclear or incorrect, contact your nearest Nikon representative.
- Some of the products described in this manual may not be included in the set you have purchased.
- Be sure to read the manuals for any other products that you are using with this attachment (the microscope, power supply, super high-pressure mercury lamp power supply, high-intensity light source, etc.).
- If the T-FL-E motorized Epi-fl attachment is connected to the T-HUBC HUB controller, please refer to the instruction manual supplied with the HUB controller.
- If the T-FLC-E motorized cassette holder is being used, please refer to the instruction manual supplied with the T-HUBC HUB controller.

Warning / Caution Symbols Used in This Manual

Although Nikon products are designed to provide you with the utmost safety during use, incorrect usage or disregard of the instructions can cause personal injury or property damage. For your safety, read the instruction manual carefully and thoroughly before using the instrument. Do not discard this manual but keep it near the product for easy reference. In this manual, safety instructions are indicated with the symbols shown below. Be sure to follow the instructions indicated with these symbols to ensure correct and safe operation.

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



WARNING

1 Intended product use

This system should only be used for microscopic observation. Do not use it for any other purpose.

2 Do not disassemble

Disassembling may cause electrical shock, exposure to ultraviolet light, and/or malfunction. Do not disassemble any parts other than those mentioned in this manual. If you notice any malfunction, contact your nearest Nikon representative.

3 Read the instruction manuals carefully

For your safety, carefully read this manual and the manuals provided with the other products used with the system. Make certain to heed the warnings and cautions at the beginning of each manual in particular.

| | |
|--|--|
| Cautions regarding the power supply | Read the manual for the microscope. |
| Cautions regarding lamp heat | Read the manuals for the microscope and the light source (super high-pressure mercury lamp power supply or high-intensity light source). |
| Cautions regarding ultraviolet light produced by the lamp | Read the manual for the light source (super high-pressure mercury lamp power supply or high-intensity light source). |
| Cautions regarding lamp bursting and the gas sealed inside the lamp | Read the manual for the light source (super high-pressure mercury lamp power supply or high-intensity light source). |
| Cautions regarding the lamp specifications | Read the manual for the microscope and the light source (super high-pressure mercury lamp power supply or high-intensity light source). |

4 Mercury lamps and xenon lamps

The mercury (or xenon) lamp used with this system requires special handling. In order to use this system safely and correctly, carefully read the warnings below and beware of the dangers. Also carefully read the manual for the super high-pressure mercury lamp power supply (or high-intensity light source) and the manual (if provided) by the manufacturer of the lamp and follow their instructions.

The Hazards of Mercury Lamps and Xenon Lamps

- 1 Mercury (and Xenon) lamps radiate ultraviolet light that is harmful to the eyes and skin when they are turned on. Direct viewing of light from these lamps may result in blindness.
- 2 Gas is sealed under very high pressure inside the lamps. The pressure increases when the lamp is on. If the lamp is scratched, dirty, subjected to high external pressure or physical impact, or used beyond its operational life, the sealed gas may escape or the lamp may burst. This can result in someone inhaling the gas, injuring themselves on the glass, or other accidents.
- 3 When the lamp is on, the lamp and its surroundings become extremely hot. Touching the lamp with bare hands could result in burns. Flammable materials placed near the lamp could ignite.
- 4 Using other than the specified type of lamp could result in an accident, such as a burst.

Because safety is a top priority in the design of Nikon products, the hazards described above should not pose any danger as long as you heed all of the warnings and cautions in the manuals and use the system only for its intended purpose. However, the hazards described above could lead to an accident if you fail to heed all of the warnings and cautions in the manuals, if you strike the system, or if you attempt to disassemble the system. Therefore, always be sure to heed all of the warnings and cautions.

Continued on the following page.

Continued from the preceding page.

5 Always turn the lamp off when exchanging filter blocks or cassette holder

When exchanging filter blocks or cassette holder, always be sure to turn off the lamp connected to the Epi-fl attachment. If the lamp is left on during exchange, there is a danger of exposure to ultraviolet light.

6 Use the ultraviolet light shield

Do not directly see the light irradiated from the objective. Harmful excitation light will be given out from the objective depending on the excitation method used. When you see the part around the objective, be sure to see through the ultraviolet light shield.



CAUTION

1 Turn off the power when assembling the equipment, connecting or disconnecting cables, or when replacing the lamp

In order to prevent electrical shock and damage to the equipment, always turn off the power switch on the microscope, power supplies, etc., and unplug the power cord before assembling the equipment, connecting or disconnecting cables or replacing a lamp.

2 Do not wet the equipment

If the microscope, the system or the power supply gets wet, a short circuit may result that could damage it or make it extremely hot. If you accidentally spill a liquid on the equipment, immediately turn the power switch off and unplug the power cord. Then use a dry cloth to wipe away the moisture. If any liquid gets inside the equipment, do not use it; instead, contact your nearest Nikon representative.

3 Caution concerning assembly

Be careful not to pinch your hands or fingers when assembling the equipment.

4 Correctly assemble the equipment and center the lamp

Be sure to correctly assemble the epi-fl attachment and center the lamp. If this is not done correctly, light source image from the lamp may focus on structural parts (such as a filter frame) in the optical path. This may lead to an increase in temperature of the surface of the structural part, which causes deformation and smoldering.

For details on assembly, see the chapter, "V. Assembly" in this manual.

For details on centering the lamp, see the instruction manual with the power supply of the lamp.

Notes on Handling the System

1 Handle the system gently

This system is a precision optical instrument. Handle the system gently, avoiding any physical shocks.

2 Handling of filters

- Interference filters (especially excitation filters, which are exposed to strong light) deteriorate with the passage of time. Replace them according to the number of hours they have been in use.
- Filter characteristics may change if the filter is exposed to high humidity. In order to prevent changes in or deterioration of filter characteristics, avoid using or storing the filters under conditions of high humidity or high temperature, and avoid subjecting them to rapid temperature changes. When a filter is not in use, storage in a desiccator or a hermetically sealed container with a drying agent is recommended.
- The filters in the eight types of filter blocks listed below offer sharp, high-resolution waveform characteristics in comparison with normal filters. However, because they have sophisticated coatings, they must be handled with extra care. Abrasion caused by cleaning is a special concern. (Follow the procedure described in section “1. Filter and lens cleaning” of chapter, “VIII. Care and Maintenance.”)

Single-band filter blocks: DAPI, FITC, TxRed

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

3 Dirty lenses



Do not let dust, fingerprints, etc. get on the lenses. Dirt on the lenses, mirrors, etc. will adversely affect the view of the image. If any of the lenses gets dirty, clean it as described in chapter “VIII. Care and Maintenance.”

4 Installation location

In order to avoid degraded performance and to prevent malfunctions, take the following requirements into consideration when selecting a location to install the system:

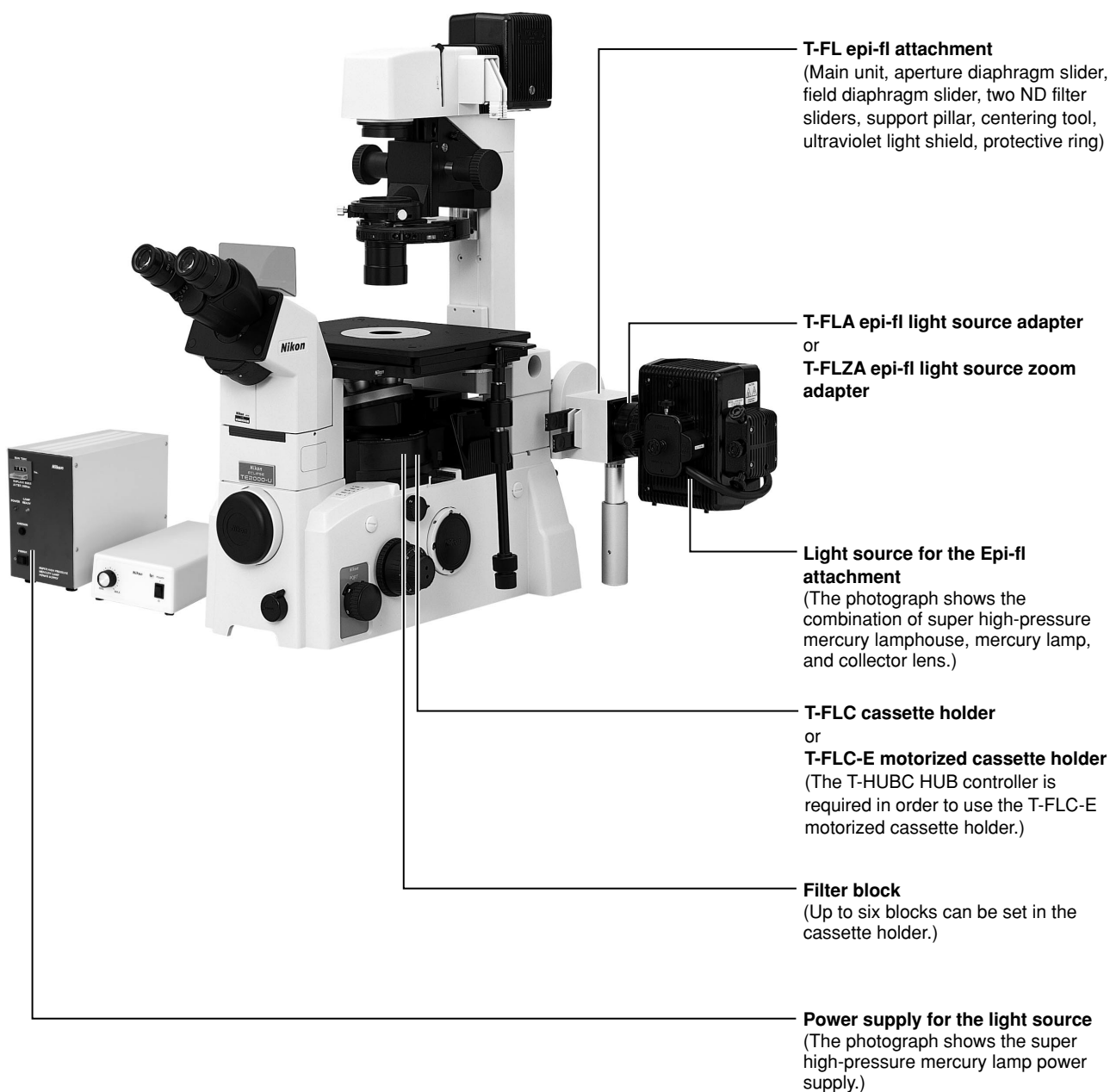
- Install the system in a location with little vibration.
- Avoid installing the system in a location exposed to direct sunlight.
- Avoid installing the system in a dusty location.
- Avoid installing the system in a location subject to high temperatures (40°C or higher) or high humidity (60% or higher). (Such conditions could allow mold or condensation to form on the lenses and filters.)

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I. Names of Component Parts

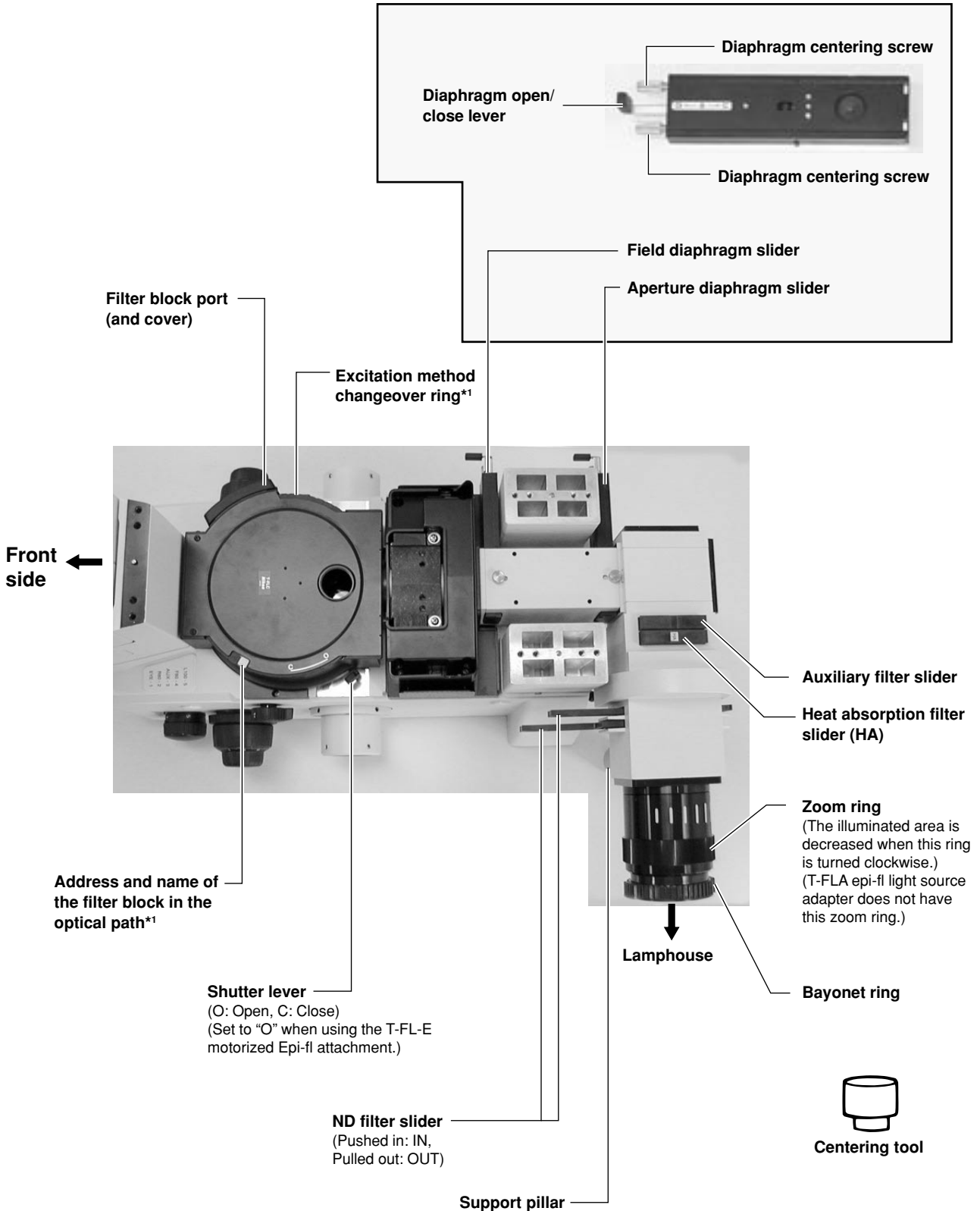
The picture below shows all the parts needed for the Epi-fl microscopy mounted on the TE2000-U which can perform bright-field microscopy.



- For details on the T-FL-E motorized Epi-fl attachment, see page 22.
- For details on how to operate the T-FLC-E motorized cassette holder, please refer to the instruction manual supplied with the T-HUBC HUB controller.

II. Names of Operational Parts

The photograph below shows the T-FLC cassette holder and T-FL Epi-fl attachment mounted on the TE2000-U. For clarity, the stage, revolving nosepiece and dia-illuminator are omitted from the photograph.



*1: Not equipped on the T-FLC-E motorized cassette holder.

III. Microscopy

The general procedure for microscopy is described below.

For details on each step, see the corresponding item in chapter “IV. Operation of Each Part.”

If the system is not yet assembled, see chapter “V. Assembly” first.

For details on the assembly, handling, and use of the microscope, power supply, and light source, see their respective manuals.



WARNING

Before using the system, be sure to read the “⚠ WARNING” and “⚠ CAUTION” at the beginning of this manual, and also the section entitled, “Notes on Handling the System.” Be certain to heed all of the warnings and cautions.

Also be sure to read the manuals for any other products that you are using with this system (the microscope, power supply, super high-pressure mercury lamp power supply, high-intensity light source, etc.), and heed all of the warnings and cautions in those manuals.

Before starting

- 1 Check the cumulative “lit on” time of the lamp. If the time has exceeded the average operation life for lamps of its kind, replace the lamp.
- 2 Use non-fluorescent slide glass.
- 3 Use non-fluorescent immersion oil.
- 4 In order to prevent fading of the specimen, always insert the shutter into the optical path whenever you are not actually looking through the binocular eyepiece.
- 5 When switching the excitation method by rotating the excitation method changeover ring, always be sure the ring is set to a click-stop position. Do not operate the microscope while the ring is set to positions between click-stops. If the microscope is operated while this ring is set between click-stops, some internal parts will increase in temperature.

1 Epi-fl microscopy

- During Epi-fl microscopy, turn off the microscope’s diascope illumination. (Turn off the dia-illumination ON/OFF switch on the microscope main body or turn off the power switch on the power supply.)
- If DIC attachments are attached to your microscope, remove the analyzer and DIC prism for objective out of the optical path.

1

Select the excitation method.

Rotate the excitation method changeover ring to insert the filter block of the desired excitation method into the optical path. (p. 14)

2

Remove the shutter from the optical path.

Move the shutter lever to the “O” (Open) position and remove the shutter from the optical path. (p. 13)

3

Center the episcopic lamp.

Turn the episcopic lamp on and center the lamp.

For details, see the manual for the light source (super high-pressure mercury lamp power supply or high-intensity light source).

Remove an objective and screw on the centering tool in its place. Center the lamp while observing the window of the centering tool.

- The T-DH dia-illuminator 100W can be tilted so that you can completely observe the window of the centering tool.
- Set the aperture diaphragm to fully open.

4 Focus on the specimen.

Place the specimen on the stage and focus on the specimen with the 10x objective. For details, see the manual for the microscope.

5 Adjust the diopter and the interpupillary distance.

For details, see the manual for the microscope.

6 Center the field diaphragm.

(p. 12)

7 Switch to any desired objective.

Switch to any desired objective and view the specimen.

- Readjust the focus.
- Use ND filters to adjust the brightness. (p. 13)
- Close the field diaphragm so that it is just outside the viewfield. (p. 12)
- Usually, the aperture diaphragm is left fully open. (p. 12)
- When using an oil-immersion type objective, apply immersion oil between the specimen and the objective. (p. 16)
- Place the shutter into the optical path whenever you temporarily halt Epi-fl microscopy. (p. 13)

2 General bright-field microscopy

Even when the Epi-fl attachment is installed, general bright-field microscopy can be performed as with a microscope for normal bright-field microscopy.

- 1** Place the shutter into the optical path to block the episcopic optical path.
- 2** Rotate the excitation method changeover ring to move the empty position into the optical path.
- 3** Turn on the power supply, turn on the dia-illumination ON/OFF switch on the microscope main body, and light the diasopic lamp.
- 4** Focus on the specimen.
- 5** Focus and center the condenser. Adjust the condenser aperture diaphragm and the field diaphragm on the microscope main body.
For details, see the manual for the microscope.

3 Combining Epi-fl microscopy with DIC microscopy

Even when the Epi-fl attachment is installed, DIC (differential interference contrast) microscopy can be performed by combining various accessories. DIC microscopy can be used in place of Epi-fl microscopy to find target objects in order to prevent color fading of the specimen, or Epi-fl and DIC microscopy can be used simultaneously to utilize the characteristics of both microscopies.

For details on DIC microscopy procedures, see the manual for the DIC attachments.

Accessories required for DIC microscopy

- T-ND6 sextuple DIC nosepiece
- DIC prism for objective
- DIC objective
- T-A DIC analyzer
- TE2000 system condenser
 - Condenser lens
 - DIC module
 - Condenser turret (or condenser slider)
 - Other required parts
- T-P DIC polarizer

- 1 Install the accessories.
For details, see the manual for the TE2000 DIC attachment.
- 2 Place the shutter into the optical path to block the episcopic optical path.
- 3 Rotate the excitation method changeover ring to move the empty position into the optical path.
- 4 Turn on the power supply, turn on the dia-illumination ON/OFF switch on the microscope main body, and light the diascope lamp.
- 5 Bring the empty hole of the analyzer and that of the polarizer into the optical path.
- 6 Turn the condenser turret so that the “A (open)” indication is in front. (When using a condenser slider, remove the module on the optical path to open the optical path.)
- 7 Move the DIC objective into the optical path.
- 8 Focus on the specimen.
- 9 Center and focus the condenser.
- 10 Place the polarizer and analyzer into the optical path.
- 11 Adjust the polarizer azimuth.
- 12 Operate the condenser to move the module with the same indication (L, M or H) as the objective’s DIC code (DIC L, DIC M, DIC H) into the optical path.
- 13 Adjust the aperture and field diaphragms of the diascope illumination.

When performing Epi-fl and DIC microscopy simultaneously, move the desired filter block into the optical path, and remove the episcopic shutter out of the optical path.

Adjust the ND filters and aperture diaphragm on the Epi-fl attachment, and the ND filters on the microscope to balance the brightness of the fluorescent and differential interference contrast images.

4 Combining Epi-fl microscopy with Ph microscopy

Even when the Epi-fl attachment is installed, Ph (phase contrast) microscopy can be performed by combining various accessories. Ph microscopy can be used in place of Epi-fl microscopy to find target objects in order to prevent color fading of the specimen, or Epi-fl and Ph microscopy can be used simultaneously to utilize the characteristics of both microscopies.

For details on Ph microscopy procedures, see the manual for the microscope.

Accessories required for Ph microscopy

- Ph objective
 - TE2000 system condenser housing a Ph module or ELWD-S/SLWD condenser (SLWD condenser is used only for T-DS dia-illuminator 30W.)
 - Centering telescope
- 1 Install the accessories.
For details, see the manual for the microscope.
 - 2 Place the shutter into the optical path to block the episcopic optical path.
 - 3 Rotate the excitation method changeover ring to move the empty position into the optical path.
 - 4 Turn on the power supply, turn on the dia-illumination ON/OFF switch on the microscope main body, and light the diascopic lamp.
 - 5 Place the Ph objective into the optical path.
 - 6 Set the optical path of the condenser to “A (open).”
 - 7 Focus on the specimen.
 - 8 Center and focus the condenser.
 - 9 Set the optical path of the condenser to the same Ph code (Ph1, Ph2, Ph3) as the objective.
 - 10 Center the annular diaphragm of the condenser.
The centering method differs according to the type of condenser. See the manual for the microscope.
 - 11 Adjust the field diaphragm.

When using Epi-fl and Ph microscopy simultaneously, move the desired filter block into the optical path, and remove the episcopic shutter out of the optical path.

Insert and remove the ND filters on the Epi-fl attachment, and the microscope to balance the brightness of the fluorescent and phase contrast images.

IV. Operation of Each Part

1 Field diaphragm

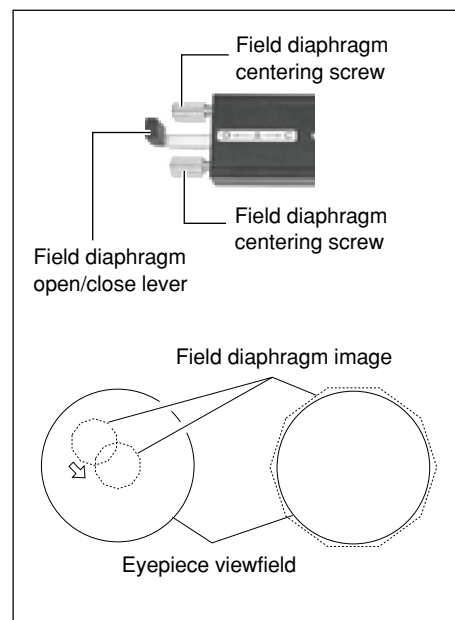
The field diaphragm restricts illumination to the area of the specimen that is being viewed. Operating the field diaphragm open/close lever changes the size of the field diaphragm. For normal observation, the size of the diaphragm should be such that it is just outside (or inside) the edge of the viewfield. If a broader area than necessary is illuminated, stray light will enter the optical system, creating flaring, reducing the contrast of the optical image, and expanding the area of fading of the specimen.

The operation of the field diaphragm is especially important in videomicroscopy or photomicrography; generally, the best results are obtained by stopping down the field diaphragm to just slightly larger than the image area or the area that will be reproduced on the film, i.e., the size of the picture composition frame.

The field diaphragm of this Epi-fl attachment needs to be centered before usage.

Centering the field diaphragm

- 1 Perform steps [1] to [5] in “1. Epi-fl microscopy” of chapter “III. Microscopy.”
- 2 Stop down the field diaphragm. (Push in the field diaphragm open/close lever.)
- 3 Move the center of the field diaphragm image to the center of the viewfield. (Turn the field diaphragm centering screws.)
- 4 Adjust the field diaphragm to roughly the size of the viewfield. (Pull out the field diaphragm open/close lever.)
- 5 Once again, move the center of the field diaphragm image to the center of the viewfield. (Turn the field diaphragm centering screws.)

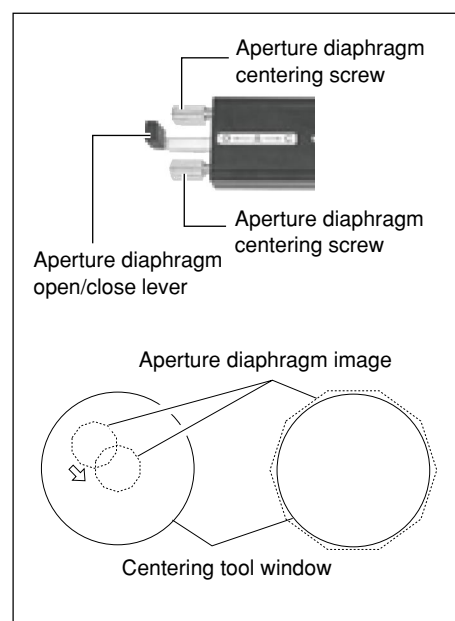


2 Aperture diaphragm

The aperture diaphragm is used to adjust the numerical aperture of an illumination optical system. When using an episcopic-fluorescent illumination optical system, the aperture diaphragm functions to adjust the brightness of the image and the size of the stray light. Although the amount of stray light decreases when the aperture diaphragm is stopped down, the image of the specimen also becomes darker. Although the image becomes brighter when the aperture diaphragm is opened, the amount of stray light may also increase. It is possible to effectively eliminate stray light and secure a bright specimen image by stopping down the aperture diaphragm to a size equivalent to the diameter of the objective pupil.

Centering the aperture diaphragm

- 1 Perform steps [1] to [3] in “1. Epi-fl microscopy” of chapter “III. Microscopy.”
- 2 Stop down the aperture diaphragm. (Push in the aperture diaphragm open/close lever.)



IV. Operation of Each Part

- 3 Move the center of the aperture diaphragm image to the center of the centering tool window. (Turn the aperture diaphragm centering screws.)
- 4 Adjust the aperture diaphragm to roughly the size of the centering tool window. (Pull out the aperture diaphragm open/close lever.)
- 5 Once again, move the center of the aperture diaphragm image to the center of the centering tool window. (Turn the aperture diaphragm centering screws.)

3 Shutter

The shutter blocks the light from the light source. In order to prevent fading of the specimen, always place the shutter in the optical path when not actually observing a specimen through the microscope. (Moving the shutter lever clockwise places the shutter into the optical path, blocking the light.) Make closing the shutter a habit in order to protect important specimens.

In addition, when temporarily halting Epi-fl microscopy in favor of microscopy using diascope light, do not forget to insert the shutter into the optical path and block the episcopic fluorescent light.

Note: If the T-FL-E motorized Epi-fl attachment is used, be sure to set the shutter on the cassette holder to “O” (Open). For details on operation of this attachment, please see the chapter “VII. T-FL-E Motorized Epi-fl Attachment.”

4 T-FLA epi-fl light source adapter / T-FLZA epi-fl light source zoom adapter

The T-FLA epi-fl light source adapter is used to connect a lamphouse to the Epi-fl attachment when using a light source such as a super high-pressure mercury lamp.

T-FLZA epi-fl light source zoom adapter is an optical system used to increase the brightness per unit area of the image surface by decreasing the illumination area versus the standard field number of 22 mm when monitoring using a high sensitivity TV camera. (Note, however, that brightness is increased only when using objectives of 40x or lower.) The field number can be varied from 22 mm to 11 mm. Since 11 mm is equivalent to a diagonal length for the image device of 2/3 of an inch, it is possible to increase the brightness to the most effective level before using a camera that has an image device of this size.

The field becomes smaller and the image becomes brighter when the zoom ring is turned clockwise.

5 ND filters

An ND filter reduces illumination without changing the color balance of the light. When using strong fluorescent light, or when a specimen is badly faded, adjust the brightness by pulling out the ND filter sliders to place the ND filters in the optical path. (If the fluorescent light is too strong, the contrast may worsen.)

The following chart shows how brightness is affected by different combinations of ND filters.

| ND4 | ND8 | Brightness |
|-----|-----|------------|
| – | – | 1 |
| ○ | – | 1/4 |
| – | ○ | 1/8 |
| ○ | ○ | 1/32 |

○: In optical path
–: Not in optical path

Replacing Filters

Pull the tab on the filter frame toward the outside and drop in the ND filter that matches the indication on the filter frame. Use a soft clean cloth to handle filters so that your fingers do not directly touch the filter frame.

6 Ultraviolet light shield

The ultraviolet light shield prevents the ultraviolet light (irradiated from the objective to the specimen) from bouncing back and entering the observer's eye. To remove this shield, loosen the fixing screws and then pull off the shield.

7 Heat absorption filter

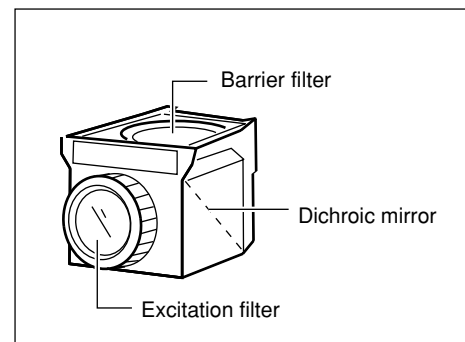
The slider with the indication "HA" at the rear of the Epi-fl attachment is the heat absorption filter slider. For normal Epi-fl microscopy, set the heat absorption filter slider in the optical path.

If the near-infrared excitation light is to be used, pull the HA slider out of the optical path since the HA filter absorbs the near-infrared and infrared light.

8 Filter blocks

Filter blocks consist of three types of optical components: an excitation filter (EX filter), a barrier filter (BA filter), and a dichroic mirror (DM). Using the items below as a guide, select a combination of filters that is suited to your purposes and to the characteristics of the specimen and the fluorescent stain.

- You can select different combinations of excitation and barrier filters for the same excitation method.
- Excitation filters, barrier filters, and dichroic mirrors can all be purchased individually.
- Excitation filters will deteriorate over time since they are exposed to intense light. Replace them as necessary.

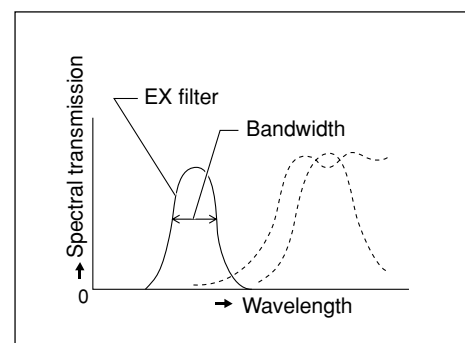


1 Selecting excitation filters (EX filters)

Excitation filters selectively pass the light within a certain range of wavelengths needed to cause the specimen to fluoresce (excitation light) and filter out all other light. The range of wavelengths that a given filter passes is called the "bandwidth" of the filter.

An excitation filter's bandwidth determines the brightness of the fluorescent image the occurrence of self-fluorescence (fluorescence originating from materials other than the fluorescent stain), and the extent of fading. A wide bandwidth allows a high level of excitation light to illuminate the specimen, producing a brighter image. However, a wide bandwidth also leads to a high level of self-fluorescence and severe fading. Conversely, while a narrow bandwidth yields a dark image, since little excitation light reaches the specimen, self-fluorescence and fading are minimal.

When self-fluorescence is pronounced, use an excitation filter with a narrow bandwidth. (The resulting fluorescent image will be darker, however.) Excitation filters are likely to deteriorate the more they are used, since they are exposed to intense light. Replace excitation filters as necessary.



IV. Operation of Each Part

| | Narrow | Excitation filter bandwidth | Wide |
|---------------------------------|---------|-----------------------------|------------|
| Brightness of fluorescent image | Dark | | Bright |
| Occurrence of self-fluorescence | Minimal | Pronounced | Pronounced |
| Extent of fading | Minimal | Pronounced | Pronounced |

2 Selecting barrier filters (BA filters)

A barrier filter allows only fluorescent light generated by the specimen to pass and blocks all other excitation light reflected from the specimen. This filter makes it possible to observe the fluorescent image without unnecessary light (i.e., on a dark background).

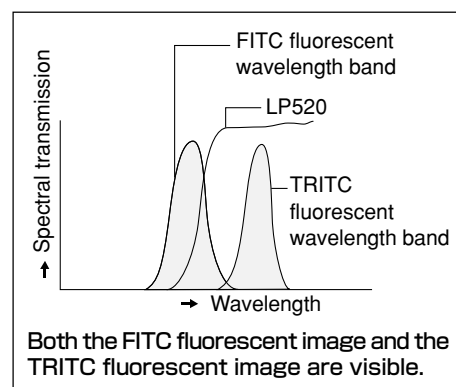
There are two types of barrier filters: LP filters (long-pass filters), which block all wavelengths that are shorter than a certain boundary wavelength and allow to pass all wavelengths that are longer than the boundary wavelength, and BP filters (band-pass filters), which allow only light in a certain range of wavelengths to pass. Use whichever type best suits your purposes.

LP filters (long-pass filters)

LP filters block all wavelengths that are shorter than a certain boundary wavelength and allow to pass all wavelengths that are longer than the boundary wavelength. The boundary wavelength is called the cut-on wavelength.

- ① When the specimen is stained with a fluorescent color for which the fluorescent light wavelength band and the excitation wavelength band (the light that is absorbed by the specimen in order to fluoresce) are extremely close together, the fluorescent light can generally be seen most effectively if a barrier filter with a “cut-on” wavelength that is comparatively short, within the range permitted by performance considerations, is selected. As a general rule, the longer the cut-on wavelength is, the more complete the separation between the excitation light and fluorescent light, and the darker the background of the fluorescent image becomes. Recently, however, because of improved filter performance, it is becoming increasingly common to use barrier filters with shorter cut-on wavelengths.
- ② To view the fluorescent images of all the colors in a specimen stained in multiple colors, use an LP filter.

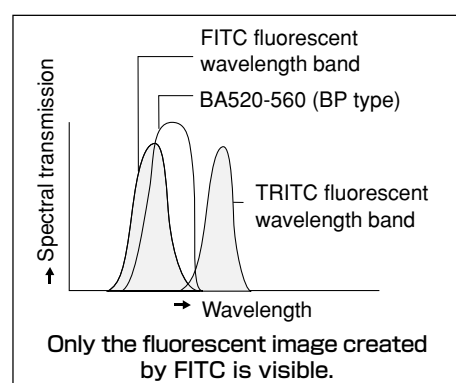
However, when using a normal dichroic mirror, an excitation filter, and an LP filter-type barrier filter in combination, the stain that fluoresces at the longer wavelength (for example, TRITC when the specimen is stained with FITC and TRITC) may not be excited sufficiently, with the result that the fluorescent image created by the stain may appear extremely dark. In this type of situation, the use of a multi-band filter is recommended.



BP filters (band-pass filters)

A BP filter allows only light in a certain range of wavelengths to pass. This type of filter is used to view the fluorescent image created by a specific stain when a given specimen has multiple stains. (For example, in a specimen with two stains, FITC and TRITC, select BA520-560 to observe the fluorescent image created by FITC.)

However, you may not be able to distinguish the self-fluorescence from the other fluorescence in the image created by the BP filter since the image will only be of one color (green, in the above example). It is best to use an LP filter if you wish to distinguish the self-fluorescence from subtle differences in hue.



3 Excitation filter and barrier filter replacement

Excitation and barrier filters can be removed from the filter block and replaced with other filters. (The filters are screwed into the filter block. The dichroic mirror cannot be removed from the filter block.)

9 Oil-immersion type objectives

Objectives marked “oil” are oil-immersion type objectives. These objectives are used with immersion oil applied between the specimen and the tip of the objective. Always use non-fluorescent oil. (For example, Nikon Immersion Oil DF.) If other kinds of oil are used, fluorescent light from the oil may adversely affect the image.

Bubbles in the oil will adversely affect the viewing of the image. Be careful to prevent the formation of air bubbles. To check for air bubbles, look at the exit pupil of the objective. (When using the T-TD eyepiece tube D, place the Bertrand lens in the optical path by rotating the eyepiece tube turret to the position “B”. and focus on the exit pupil of the objective by rotating the Bertrand lens focusing screw. When using the T-TS eyepiece tube S, insert the centering telescope instead of an eyepiece, and rotate the eyepiece part of the centering telescope to focus on the exit pupil of the objective.) If there are bubbles in the oil, remove them by one of the following methods.

- Turn the revolving nosepiece slightly, moving the objective in question back and forth one or two times.
- Add more oil.
- Remove the oil and replace it with new oil.

Use as little oil as possible (just enough to fill the space between the tip of the objective and the specimen). If too much oil is applied, the excess oil will flow onto the stage and around the condenser.

Any oil remaining on oil-immersion type objectives or on the tip of dry-type objectives will have a discernible, negative effect on the image. After using oil, wipe all of it away and also make sure that there is no oil on the tips of the other objectives. Oil on the condenser lens should also be wiped away carefully after use.

Use petroleum benzene to wipe away immersion oil. Then, after the oil has been removed, wipe the lens surfaces, etc., with absolute alcohol (ethyl alcohol or methyl alcohol).

If you cannot obtain petroleum benzene, use methyl alcohol. However, because methyl alcohol does not clean as well as petroleum benzene, it will be necessary to wipe the surfaces repeatedly. (Usually, three or four times is sufficient to clean the lenses.)

Use petroleum benzene only to remove immersion oil from the tips of objectives; do not use it for cleaning the fluorescent filters, etc. Use absolute alcohol and petroleum benzene according to the instructions provided by their manufacturers.

10 Fluorescent photomicrography

For the basic procedures and key points of photomicrography, see the manual provided with the photomicrographic equipment. Please note, however, that when using a fluorescent specimen, the fluorescence may fade during exposure. Take the following countermeasures in order to avoid this problem.

1 Use high-sensitivity film

Use “Tri-X (ISO400)” for monochrome photomicrography. For color shots, use daylight-type high-sensitivity film, such as “Kodak Ektachrome 400 (ISO400)” or “Fujichrome 400 (ISO400).”

2 Creating a bright optical system combination

Even if the total magnification on the film is the same, the exposure time can vary greatly for different combinations of objectives and projection lenses. Rather than increasing the magnification of the projection lens, increasing the magnification of the objective is recommended. (This is because, in general, the numerical aperture of the objective increases as the magnification increases, and the higher the numerical aperture, the brighter the image.)

3 Adjusting the excitation light

If the excitation light is too bright, the specimen will fade quickly, making it impossible to get a good shot of the fluorescent image. Therefore, adjust the brightness by inserting ND filters into the optical path.

4 Specimen

If a faded portion of a specimen is shot, the exposure time increases, the color reproduction is poor, and the resulting photomicrograph will not be satisfactory. Move the specimen and shoot a more vivid portion of the specimen that has not previously been exposed to the excitation light. We recommend using the DIC method or the phase contrast method to select the portion to be shot, and then switching to the Epi-fl method for shooting the actual photomicrograph.

11 TV microscopy

When performing microscopy using a high-sensitivity TV camera, it is sometimes best to insert an infrared (IR) cut filter in front of the camera receptor. Experiment, and use the IR cut filter when needed.

V. Assembly

The procedures for assembling the system are described below.

For details on the assembly, handling, and usage of the microscope, super high-pressure mercury lamp, etc., see their respective manuals.



WARNING

Before using the system, be sure to read the “⚠ WARNING” and “⚠ CAUTION” at the beginning of this manual, and also the section entitled, “Notes on Handling the System.” Be certain to heed all of the warnings and cautions.

Also be sure to read the manuals for any other products that you are using with this system (the microscope, super high-pressure mercury lamp power supply, high-intensity light source, etc.), and heed all of the warnings and cautions in those manuals.

In particular, mishandling a mercury (or xenon) lamp used with this system can lead to a serious accident. Exercise caution.

In order to prevent electric shock fire, accidents involving ultraviolet light, burns, and other injuries, make sure that the power switches for the system and super high-pressure mercury lamp (or high-intensity light source) power supplies are turned off before beginning assembly work.

Required tools

- Hexagonal screwdriver (2 mm) : 1 (provided with the microscope)
- Hexagonal wrench: 1 (provided with the cassette holder)

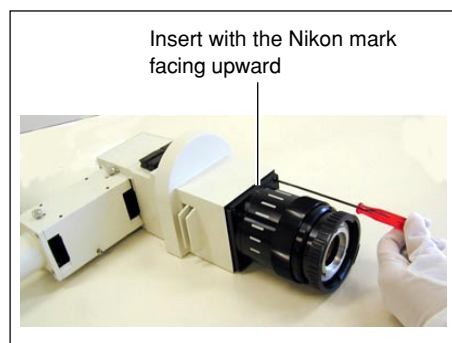
See the illustrations while assembling the system.

If the microscope is already fully assembled, remove the dia-illuminator, stage, objectives, and revolving nosepiece.

Scratches or fingerprints on the lenses and filters will adversely affect the image. Handle these components carefully in order to keep them free from scratches and fingerprints.

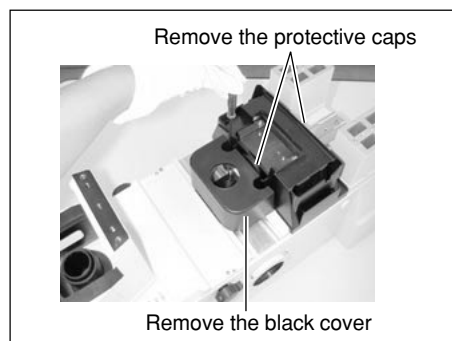
1 Attaching a light source adapter to the Epi-fl attachment

Insert the light source adapter into the Epi-fl attachment with the Nikon mark facing upward and secure using the hexagonal socket head bolt.

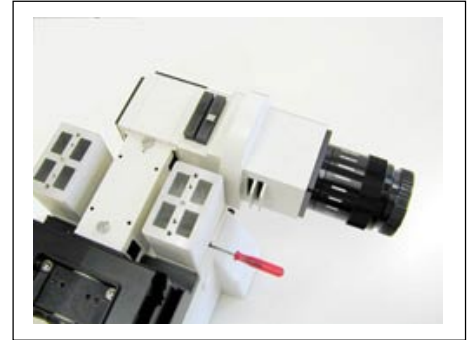


2 Attaching the Epi-fl attachment to the microscope

- 1 Use a hexagonal driver to remove the two screws that secure the black cover in the middle of the microscope.
- 2 Remove the two protective caps.



- 3 Insert the Epi-fl attachment into the guides on the back of the microscope and secure by tightening the clamp screws with a hexagonal driver. Attach the protective ring to the end of the Epi-fl attachment (revolving nosepiece side).



- 4 Loosen the clamp screw securing the support pillar so that the sliding part at the end slides freely. Screw the support pillar into the bottom of the Epi-fl attachment and secure the end to the installation surface by tightening the clamp screw with a hexagonal driver.





- 5 Insert the field diaphragm slider into the front side opening and the aperture diaphragm slider into the rear side opening.

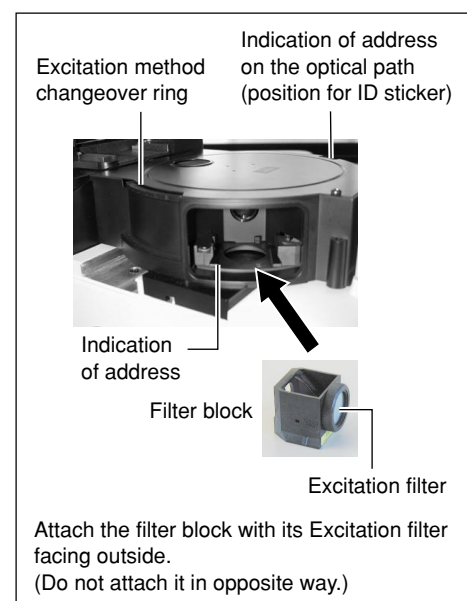
3 Attaching a cassette holder to the microscope

Insert the cassette holder along the attachment groove from the right side of the microscope and secure with screws on the left and right. (Use the provided hexagonal wrench. Note that the screws should be tightened after the filter block port cover is removed.)

4 Attaching a filter block

It is possible to attach up to six filter blocks.

- 1 Remove the filter block port cover, check the turret address, and insert the filter block along the guide groove.
- 2 Rotate the excitation method changeover ring to display the same address and affix an ID sticker that matches the filter block type. Affix “” or “” (a blank sticker that can be written on freely) for spaces in which no filter block is inserted.
- 3 Re-attach the filter block port cover.



5 Installing the ultraviolet light shield

Attach the ultraviolet light shield to the eyepiece tube using the two screws.

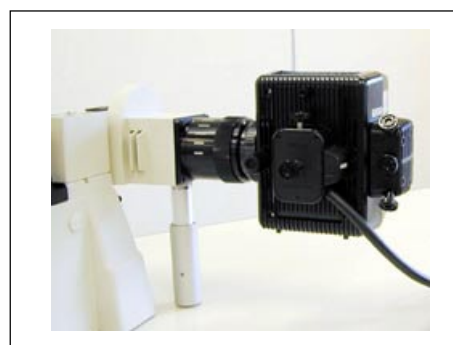
6 Microscope assembly

Follow the instructions in the microscope manual.

7 Installing the light source

Install the light source on the bayonet ring on the back side of the light source adapter. Be sure to attach the collector lens in between.

For details see the manual provided for either the super high-pressure mercury lamp power supply or the high-intensity light source.



Now the assembly is completed.

VI. Troubleshooting Tables

Improper use of the microscope may adversely affect performance, even if the microscope is not damaged. If any of the problems listed in the table below arise, take the countermeasures indicated. Also refer to the instruction manual supplied with the microscope, light source and the DIC attachment (If DIC attachment is also used).

| Problem | Cause | Countermeasure |
|--|---|---|
| Lamp does not light. | The power supply is not plugged in. | Plug the power cord into an outlet. |
| | The lamp connector is not connected to the power supply. | Connect the lamp connector to the power supply. |
| | The lamp has reached the end of its operational life. | Replace the lamp. |
| | The fuse is blown. | If the fuse can be replaced, replace it. Otherwise, contact your nearest Nikon representative. |
| Even though the lamp is on, the image is not visible. | The shutter is in the optical path. | Remove the shutter from the optical path. |
| | The filter block has stopped at an intermediate position. | Rotate the excitation method changeover ring to the clickstop position. |
| | The filter block selection is incorrect. | Select the correct filter block. |
| | The combination of excitation filter, barrier filter and dichroic mirror is inappropriate, or one of these components is missing. | Use a filter block with a correct combination. |
| Even though the lamp is on, the image is extremely dark. | The light source is not centered properly. | Center the lamp. Especially when using a 100x objective, recenter the lamp while observing the fluorescent image. |
| | ND filters are in the optical path. | If necessary, remove the ND filters from the optical path. |
| | The combination of excitation filter, barrier filter and dichroic mirror is inappropriate for the specimen. | Use a filter block with a combination suited for the specimen. |
| | A halogen lamp is being used with a dark specimen. | Change the light source to a mercury lamp. |
| | The designated objective is not being used with UV or V excitation. | Use the designated objective. |
| The illumination light is leaking over to the observation side. | The installed position of a filter block has deviated from the prescribed position. | Insert the filter block until it touches the back of the turret. |
| Contrast is poor. | The objective or cover glass is dirty. | Clean the objective or cover glass. |
| | The immersion oil is fluorescing. | Use non-fluorescent immersion oil (Nikon Immersion Oil DF). |
| | The slide glass is fluorescing. | Use non-fluorescent slide glass. |
| | The room is too bright. | Darken the room. |
| Viewing is poor. | There is no cover glass in place. | Use a cover glass. (However, no cover glass is required when using an NCG objective.) |
| | No immersion oil has been applied to the tip of an immersion-oil type objective. | Apply Nikon Immersion Oil DF. |
| | The specified immersion oil is not being used. | Apply Nikon Immersion Oil DF. |
| | The filter block being used is not suited for the specimen. | Use a filter block suited for the specimen. |
| The viewfield is vignetting. | The field diaphragm has been stopped down too far. | Open the field diaphragm so that it is just outside of the viewfield. |
| | The ND filter slider or shutter has stopped at an intermediate position. | Pull out or push in the slider all of the way. |
| | The installed position of a filter block has deviated from the prescribed position. | Insert the filter block until it touches the back of the turret. |
| | The filter block has stopped at an intermediate position. | Rotate the excitation method changeover ring to the clickstop position. |

VII. T-FL-E Motorized Epi-fl Attachment

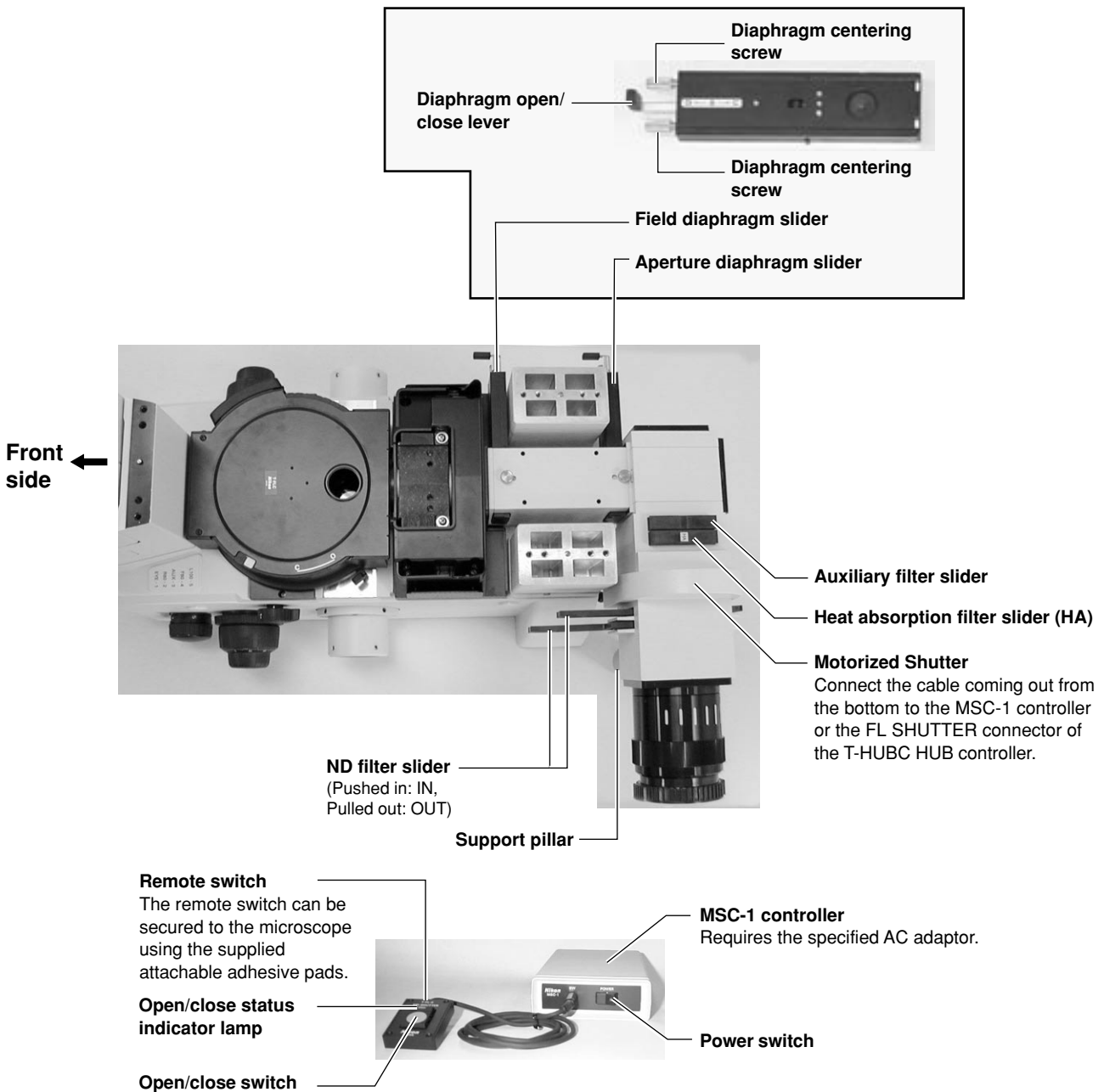
This attachment functions just like the T-FL Epi-fl attachment for the Inverted Microscope ECLIPSE TE2000, but with the addition of a motorized shutter. The shutter can be opened and closed using either a remote switch or an input signal connected to the external input terminal on the attachment.

Be sure to set the shutter on the cassette holder to “O” (Open) when using this attachment.

This attachment also requires a rated 9-V adaptor (Y-THAC adaptor) as a power supply when used other than in conjunction with motorized options for the TE2000.

■ Names of operational parts

For clarity, the stage, revolving nosepiece and dia-illuminator are omitted from the photograph.

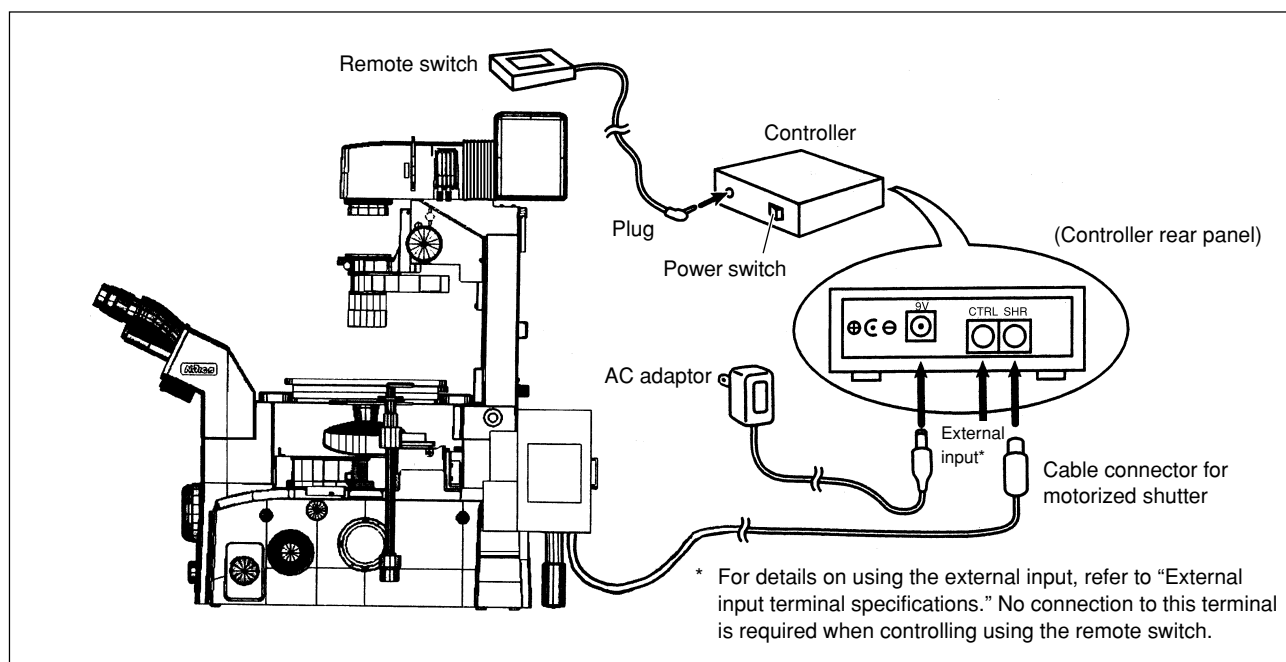


The MSC-1 controller and remote switch are not used if the cable of the motorized shutter is connected to the T-HUBC HUB controller so that the shutter can be opened and closed using a remote control pad or from a personal computer.

VII. T-FL-E Motorized Epi-fl Attachment

■ Assembly

- 1 Attach this equipment to the microscope. For details on how to attach this equipment, see the procedure described in the chapter “V. Assembly.” Similarly refer to each instructions when attaching the lamphouse, cassette holder, filter block, ND filter slider, shutter lever, ultraviolet light shield, etc.
- 2 Connect the cable connector for motorized shutter to the “SHR” connector of the MSC-1 controller or the “FL SHUTTER” connector of the T-HUBC HUB controller.
- 3 When connecting to the MSC-1 controller:
Connect the remote switch plug to the connector on the front of the controller.
The remote switch can be attached to the microscope using the supplied adhesive pads.
Connect the plug of the AC adaptor (sold separately) to the controller.

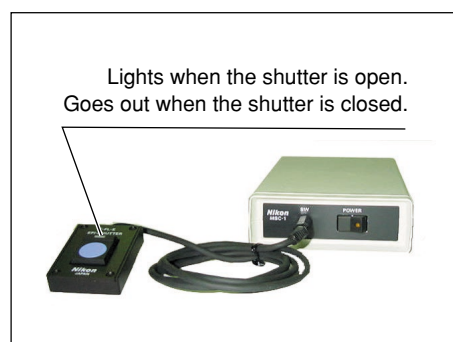


■ Control procedure

The control procedure described here is for when the MSC-1 controller and remote switch are used.

If this equipment is to be connected to the T-HUBC HUB controller, please refer to the instruction manual supplied with the HUB controller.

- In preparation, set the cassette holder shutter to “O” (Open).
- Set the power switch of the controller to ON.
- Each time the button on the remote switch is pressed, the shutter will alternately open and close. The shutter is open when the indicator lamp is on, and closed when it is off. (This is true only while power is being supplied to the motorized shutter.)



VII. T-FL-E Motorized Epi-fl Attachment

External input terminal specifications

The shutter can be opened and closed by supplying a TTL or open collector signal to the external input terminal of the CTRL connector on the rear panel of the controller.

Controller CTRL connector specifications

| CTRL connector | | |
|----------------|-----------------------|----------------|
| 1 | DC5V | Output |
| 2 | DC9V | Input |
| 3 | Shutter closed status | H level output |
| 4 | Shutter open status | H level output |
| 5 | Close shutter command | L level input |
| 6 | Open shutter command | L level input |
| 7 | Not used | – |
| 8 | DC0V | – |

- A DC5V reference signal is output from Pin 1.
- It is possible to supply DC9V±1V power from the external device via Pin 2. (Input from external device: Approx. 1A)
Note, however, that power supplied from the DC IN jack has priority and that the power line is automatically switched if a plug is inserted in the jack.
- Output from the controller is TTL.
- Input to the controller may be either TTL or open collector.
- The open/close shutter signals should be set between 100 to 400 ms.

Troubleshooting

| Problem | Cause | Countermeasure |
|----------------------------------|---|---|
| Shutter does not function | The remote switch plug, motorized shutter cable, and/or AC adaptor connector are not connected, or the AC adaptor is not plugged into an electrical outlet. | Make sure all connections have been made correctly. |

For problems other than the above, please refer to the chapter “VI. Troubleshooting Tables”.

VIII. Care and Maintenance

1 Filter and lens cleaning

Do not let dust, fingerprints, etc. get on the lenses or filters. Dirt on the lenses, filters, etc. will adversely affect the view of the image. If any of the lenses or filters get dirty, clean them as described below.

- Use an air blower to blow away dust. If that does not suffice, brush away the dust with a soft, clean brush, or else wipe it away gently with gauze.
- To remove fingerprints or grease, use a piece of soft, clean cotton cloth, lens tissue, or gauze moistened with absolute alcohol (ethyl alcohol or methyl alcohol). However, do not use the same area of the cloth, etc. to wipe more than once.
- Use petroleum benzine to clean off immersion oil. Wiping with absolute alcohol (ethyl alcohol or methyl alcohol) after the oil has been removed finishes the clean up process.
- If you cannot obtain petroleum benzine, use methyl alcohol. However, because methyl alcohol does not clean as well as petroleum benzine, it will be necessary to wipe the surfaces repeatedly. (Usually, three or four times is sufficient to clean lenses or filters.)
- Use petroleum benzine only to remove immersion oil from objectives; do not use petroleum benzine for cleaning the entrance lens on the eyepiece tube, filters, etc.
- Absolute alcohol and petroleum benzine are both highly flammable. Handle them carefully, especially around open flames and when turning power switches on and off, etc.
- Follow the manufacturer's instructions when handling absolute alcohol and petroleum benzine.

2 Cleaning painted components

Do not use organic solvents such as alcohol, ether, or paint thinner on painted components, plastic components or printed components. Doing so could result in discoloration or in peeling of the printed characters. For persistent dirt, dampen a piece of gauze with neutral detergent and wipe lightly.

3 Storage

Store the system in a dry place where mold is not likely to form.

Store the objectives, eyepieces, filter blocks, etc. in a desiccator or similar container with a drying agent.

Put the vinyl cover over the system after use to protect it from dust. Before putting on the vinyl cover, turn off the power switches for the microscope and the Epi-fl attachment light source, and wait until the lamphouse has cooled.

4 Regular inspection (charged)

Regular inspection (expenses charged) of this system is recommended to maintain peak performance. Contact your nearest Nikon representative for details about regular inspection.