INSTRUCTIONS

AX REFLECTED LIGHT MODULE

This instruction manual is for the Olympus AX Reflected Light Module. To obtain optimum performance and to familiarize yourself fully with the use of this attachment, we recommend you study this manual together with the instruction manual for the AX70/80 microscopes before operating the attachment.
This unit employs a UIS (universal infinity system) optical design, and should be used only with UIS microscope frames, eyepieces, objectives, condensers, etc. Less than optimum performance may result if inappropriate accessories are used.

- When attached to the AX70 or AX80 microscope frame, the combination allows reflected light fluorescence observation as well as other methods of reflected light observation.

This instruction manual only covers the observation methods made possible by the reflected light module. For transmitted light and combined observation modes, also study carefully the instruction manual for the AX70 or AX80 microscopes to obtain a thorough understanding of the operation of the combined units.

The universal reflected light vertical illuminator features interchangeable excitation cubes to employ excitation light of different wavelengths. It also allows combined or alternating reflected light fluorescence and transmitted white light observations.

1. Reflected light fluorescence + transmitted light phase contrast.
2. Reflected light fluorescence + transmitted light Nomarski differential interference contrast.
3. Reflected light fluorescence + transmitted light brightfield or darkfield observation.

By mounting a white light cube for reflected light observation, the following observation methods become possible:
1. Reflected light brightfield/darkfield observation.
2. Reflected light Nomarski differential interference contrast.

This instruction manual consist of two parts: I. Reflected Light Fluorescence Observation and II. Reflected Light Observation Modes. Use these headings to find the relevant page with the instructions for the particular observation mode.

1 Getting Ready

1. This attachment is a precision instrument. Handle it with care and avoid subjecting it to sudden or severe impact.
2. When installing the microscope, ensure that there is ample free space around the lamp housing, in particular above and below, as the lamp housing will become very hot during operation.
3. When installing the power supply unit (BH2-RFL-T3), to prevent overheating, it is important to make sure to leave at least 5 cm (2") of free space between the wall, or other solid objects, and the rear and right and left sides of the power supply unit.
4. The maximum pressure mercury burner (mercury arc lamp) used should be a USH102D burner (made by Ushio Electric).

   Use the Olympus BH2-RFL-T3 power supply unit for the mercury burner.
5. Make certain that the burner is installed correctly and that all cords are correctly connected.
6. Be sure to use the UV protective shield with the unit. (See page 9.)
7. Do not open the lamp housing while it is turned on or for at least 15 minutes after it is turned off. Lamp housing parts will be extremely hot and will cause burns if touched. (See page 19.)
8. Do not apply excessive force to the limiting mechanisms provided with all accessories.
9. The power supply unit contains high voltage components. Never attempt to disassemble the unit.
10. To avoid potential shock hazard, be sure to ground the power cord wire.
11. Before opening the lamp housing for replacement of the burner or any other internal part, turn OFF the main switch and unplug the lamp housing's connecting cord plug from the output connector on the power supply unit. Wait for 10 minutes or more until the lamp housing cools down.
12. Make sure that the power supply unit's main switch is turned OFF before connecting the power cord to the AC mains.
2 Maintenance and Storage

1. Be careful and avoid leaving dirt or fingerprints on the lenses, filters, or the high pressure mercury burner. If contaminated, clean by wiping gently with a piece of gauze. To remove fingerprints or oil stains, wipe with gauze slightly moistened with xylene or a mixture of ether (70%) and alcohol (30%). Never attempt to use organic solvents to clean the microscope components. To clean plastic parts, use a clean, lint-free cloth lightly moistened with a diluted neutral detergent.

* Since ether and alcohol are highly flammable, be careful to keep these chemicals away from open fire and potential sources of electrical sparks, such as main switches.

2. Do not disassemble any part of the attachment.

3. The mercury burner has a service life of approx. 200 hours. When the hour counter on the power supply unit indicates 200 hours, replace the burner with a new one. (See page 8.)

4. When not using the attachment, keep it covered with the provided dust cover and store it at a dry, clean place.

5. If a dichroic mirror cube is not going to be used for a while, place it in its container and store it in a safe place.

3 Possible Intermediate Attachment Combinations

![Diagram of attachment combinations]

- **U-SWETR**
  - Super Widefield Erect Image
  - Trinocular Observation Tube

- **U-SWTR-2**
  - Super Widefield
  - Trinocular Observation Tube

Max. 1 Intermediate Attachment

AX70
- Microscope Frame

<table>
<thead>
<tr>
<th>Attachment Code</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>U-CPA</td>
<td>Polarized light intermediate attachment</td>
</tr>
<tr>
<td>U-CA</td>
<td>Magnification changer</td>
</tr>
<tr>
<td>U-MDO</td>
<td>Multi-viewing attachment</td>
</tr>
<tr>
<td>U-DA</td>
<td>Drawing attachment</td>
</tr>
</tbody>
</table>
4 Safety Symbols on the Microscope Frame

The following symbols are found on the microscope. Study the meaning of the symbols, and always use the microscope in the safest possible manner.

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Explanation</th>
</tr>
</thead>
<tbody>
<tr>
<td>☠️</td>
<td>Indicates that the surface becomes hot, and should not be touched with bare hands.</td>
</tr>
<tr>
<td>🔥scape</td>
<td>Before use, carefully read the instruction manual.</td>
</tr>
<tr>
<td>⚡️</td>
<td>Indicates a potential fire hazard; when replacing fuses, be sure replacement fuse is of the specified rating.</td>
</tr>
<tr>
<td>🔥</td>
<td>Indicates that the main switch is ON.</td>
</tr>
<tr>
<td>🌋</td>
<td>Indicates that the main switch is OFF.</td>
</tr>
<tr>
<td>🔴</td>
<td>Indicates the Standby-Remote ON/OFF function of the sub-main switch.</td>
</tr>
</tbody>
</table>

5 Caution

If the equipment is used in a manner not specified by this manual, the safety of the equipment may be impaired. In addition, the equipment may also be damaged. Always use the equipment as outlined in this instruction manual.
1. REFLECTED LIGHT FLUORESCENCE OBSERVATION

1 PRINCIPLE

The design of the reflected light fluorescence microscope features a dichroic mirror which directs the excitation light through the objective to illuminate the specimen and provide efficient fluorescence light observation.

The spectral characteristics of the dichroic mirror when it is positioned at an inclination of 45° to the optical axis of incident light is shown in the figure on the left. Because a crossover exists between transmittance and reflectance, it is necessary to employ an appropriate combination of exciter and barrier filters in conjunction with the dichroic mirror to obtain an image with good contrast.

When the dichroic mirror is inclined at 45° to the optical axis of the incident excitation light, the excitation light is reflected towards the objective and other unwanted wavelengths are practically all passed through the mirror.

When the specimen is irradiated by the excitation wavelength, it emits a visible, longer wavelength in accordance with Stoke's law. The dichroic mirror passes almost all of this light to the eyepiece. The barrier filter mounted between the dichroic mirror and the eyepiece blocks out unwanted wavelengths to provide a black background.
**NOMENCLATURE**

* Modules for both reflected light fluorescence observation methods and reflected light observation methods (marked with an * asterisk) are presented here.

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Power supply for halogen lamp

TH3

... Not required except for simultaneous transmitted light and reflected light observation.
3 Assembly Diagram

The diagram below shows how to assemble the various components for reflected light fluorescence observation. For assembly details other than those shown in the diagram below, refer to the instruction manual for the AX70 or AX80 microscope frame.

* When assembling the microscope, make sure that all parts are free of dust and dirt, and avoid scratching any parts or touching the glass surfaces. Confirm that all components are disconnected from the AC mains before assembling.
3-2 Detailed Assembly Procedure

1. Mounting the Cubes (Figs. 1, 2)

1. Using the Allen screwdriver provided with the microscope, loosen the mirror cube housing clamping screw ① on the vertical illuminator and remove the cover ②.

2. Invert the mirror cube housing.
3. Using the Allen screwdriver, loosen the cube clamping screw ③.
4. Hold the cube to be mounted with its index side ④ facing upward and slide it all the way onto the dovetail mount ⑤. Next, be sure to tighten the cube clamping screw ③ immediately.
5. Using a sharp object such as the tip of a ballpoint pen or mechanical pencil, lift the cube’s magnetic index sticker ④ and affix it to the turret at the front.

2. Mounting the Mirror Cube Housing (Fig. 3)

Invert the mirror cube housing ① with the mounted mirror cube(s) again and insert it into the vertical illuminator housing. While pushing the mirror cube housing in the direction of insertion, tighten the clamping screw ②.

★ When performing reflected light fluorescence observation, make sure to tighten the cube clamping screws ③. If a screw is left loose, the protruding screw head may impede turret rotation. (Fig. 2)

3. Attaching the Cube Seals (Fig. 4)

1. Affix stickers corresponding to the mounted mirror cubes at the mirror cube index positions ① on the hard switch.
2. At positions where no corresponding cube is mounted, affix neutral stickers to prevent glare from the indicator lamp.

★ The adhesive strength of the stickers is designed so that the stickers can be easily moved by sliding them sideways.
4 Mounting the Top Lens  
(Fig. 5)

1. Using the Allen screwdriver, loosen the top lens clamping screw ①.
2. Insert the top lens ② into its place inside the arm and tighten the clamping screw ① again.

**Improving the UV340nm Transmission Ratio**

① By removing the lens inside the top lens the UV340nm transmission ratio can be improved.
However, the field iris diaphragm image will become blurred.

**How to Remove the Lens**

1. Insert the shank of the Allen screwdriver into the notches in the spacer ①, as shown in the figure, and rotate counterclockwise to remove.

2. Remove the lens insert from the inner side of the spacer in the same manner.
   ★ The lens insert is necessary for restoring the top lens to its original state. Make sure not to mislay or lose the lens insert.

3. Invert the direction (so that the stepped side will be facing forward) of the first removed spacer ① and attach to the top lens. Tighten with the Allen screwdriver.

5 Mounting the Universal Vertical Illuminator Collector Lens Unit  
(Figs. 6,7,8)

1. Detach the halogen lamp housing for transmitted light.
2. Using the Allen screwdriver, remove the three screws ① securing the lamp housing bracket. Pull out the lamp housing bracket horizontally.
3. Using the Allen screwdriver or Allen wrench, remove the four screws ② securing the rear cover of the microscope frame and detach the cover.
4. From the rear of the arm, carefully insert the lens unit ③ until it comes up against the stop.

5. Insert the provided clamping screws into the three mounting holes ④ on the lens unit and tighten using the Allen screwdriver.

6. Plug the shutter connector plug ⑤ into the connectors on the board at the rear inside the arm. The 8-pin connector plug should be firmly connected to J15, and the 3-pin connector plug should be firmly connected to J14.

7. Return the lamp housing bracket and the rear cover to their original positions.

6 Mounting the Iris Diaphragm Units

- First mount the field iris diaphragm unit at the left hand side.

Mounting the Field Iris Diaphragm Unit

1. Insert the reflected light field iris diaphragm unit ① into the reflected light field iris diaphragm unit mounting slot ② on the microscope frame by sliding the unit onto the dovetail guide.

2. Insert clamping screws into the two mounting holes ③. While pressing the unit towards the left, tighten the screws with the Allen screwdriver.

7 Mounting the AX Reflected Light Conversion Lens or AX Reflected Light Filter Changer

1. Insert gradually the AX reflected light conversion lens ① into the reflected light conversion lens unit mounting slot ② on the microscope frame by sliding the unit onto the dovetail guide.

2. Insert clamping screws into the four mounting holes ③ and tighten the screws with the Allen screwdriver.

★ When using the AX reflected light filter changer, mount it in the same manner as the reflected light conversion lens.
8. Assembling the Lamp Housing for the Mercury Burner

**Mounting the Mercury Burner**

1. Using the Allen screwdriver provided with the microscope frame, loosen the burner socket clamping screw ①.
2. Detach the socket from lamp housing by pulling in the direction indicated by the arrow. (Fig. 11)
3. Loosen the burner clamping screws ③ and ④ (Fig. 12) and remove the transport post. (For burner replacement, remove the used burner.)
4. Insert the + pole of the mercury burner ② into the + terminal and tighten the + clamping screw ③. Then loosen the – clamping screw ④ (marked UP). Insert the – pole of the burner into the – terminal and tighten the – clamping screw ④. (Fig. 12)

   * Only use a USH102D burner (mfd. by Ushio Electric).
   * Be careful and avoid leaving fingerprints or dirt on mercury burner. If contaminated, clean by wiping gently with gauze slightly moistened with xylene or a mixture of ether (70%) and alcohol (30%).
   * To prevent possible damage to the burner, the collector lens can only be installed or removed while the socket and lamp housing are separated.

**Mounting the Collector Lens**

1. Aligning the collector lens positioning groove ① with the pin inside the lamp housing, pull up the collector lens focusing knob ②, and slide the collector lens into the lamp housing as far as it will go. Then release and return the collector lens focusing knob ② to its original position. At this point, confirm that the collector lens can be moved back and forth by turning the focusing knob ②. If not, adjust the position of the collector lens by hand so that it will click into its proper position.
2. Firmly tighten the collector lens clamping screw ③. (Fig. 13)

   **NOTE:** If this clamping screw is not tightened firmly, optimum illumination performance will not be obtainable.

3. Reattach the socket to the lamp housing by reversing the procedure described in 8.1 and 8.2 above. (Fig. 14)
4. Firmly tighten the socket clamping screw ① with the Allen screwdriver. (Fig. 11)

   * If the clamping screw ① is accidentally loosened while the burner is operating, the interlock switch turns off the burner. To restart the burner, first turn OFF the main switch on the power supply. Then disconnect the connecting cord plug from the output connector on the power supply unit and wait for about 10 minutes. Then after firmly tightening the clamping screw ①, reconnect the connecting cord plug and turn ON the main switch again. (Fig. 11)
Resetting the Burner Life Time Hour Counter

1. To reset the burner life time counter to "000.0", press the center portion ① (Fig. 16) of the reset button ① (Fig. 15) on the power supply unit's front panel.
2. The counter shows elapsed time in hours. In order not to impair the safety of the equipment, replace the burner when the counter indicates "200.0" hours.

Mercury Burner Replacement

1. In order not to impair the safety of the equipment, replace the burner when it has been used for 200.0 hours. The burner may crack if used beyond the specified lifetime.
2. Before replacing the burner, wait at least 10 minutes after turning the burner off. Before removing the burner, confirm that the main switch on the power supply unit is OFF, and unplug the connecting cord plug from the output connector on the power supply unit. Refer to page 7 for details on replacement procedure.
3. After replacing the burner, reset the burner life time hour counter to "000.0" as outlined above.

Mounting the Lamp Housing

1. Insert the mercury burner lamp housing collector unit ① into the vertical illuminator until it reaches the click-stop.
2. Tighten the collector unit clamping screw ② with the Allen screwdriver.
10 Connecting the Mercury Burner Power Supply Unit (Fig. 18)

1. Verify that the voltage and frequency of the AC mains outlet match the requirements indicated on the rating plate ① on the power supply unit. (100V models can be used with voltages in the 100-120V, 50-60 Hz range. 200V models can be used with voltages in the 220-240V, 50-60 Hz range.)

2. Plug the connecting cord plug firmly into the output connector ② on the power supply unit.

3. Connect the power cord to the AC receptacle ③ on the power supply unit. Then plug the power cord plug into a wall outlet.
   ★ If provided with a ground wire, make sure to ground the wire.

11 Mounting the UV Protective Shield (Fig. 19)

Insert the grooves in the UV protective shield ② into the flat springs on the underside of the mirror cube housing ①.
★ Align the pin of the flat springs and the grooves in the UV protective shield.

12 Mounting the Light Excluding Slide (Fig. 20)

① When performing fluorescence observation with a low-power objective, image clarity may be reduced by light reflected from the vicinity of the condenser. In this event, use the light excluding slide.
② To mount the light excluding slide ①, lower the condenser and insert the slide into the space underneath the stage. When switching between reflected light and transmitted light observation methods (phase contrast observation, Nomarski, etc.), place the slide ① on top of the light exit window ② during fluorescence observation.
13 Optional Cubes

Optional mirror cubes can be assembled using commercially available barrier filters, exciter filters, and dichroic mirrors.

**Required Dimensions of Optical Components for Cubes**

- **Barrier filter**
  - 24.8 - 24.9 mm diameter, max. 6 mm thickness

- **Exciter filter**

- **Dichroic mirror**
  - 25.7 - 25.9 X 37.7 - 37.9 mm
  - 0.95 - 1.05 mm thickness

*When replacing dichroic mirrors, make sure not to contaminate the mirror in the form of fingerprints, etc.*
The shutter opening screw is provided to accommodate special observation methods. For normal use, do not turn the shutter opening screw but leave it as when shipped from the factory. It is very dangerous if the shutter does not function properly.

To open the shutter, insert the Allen screwdriver into the shutter opening screw hole and rotate it clockwise. Caution is necessary because excessive force will be applied and result in destruction of the shutter if the Allen screwdriver is used in a normal manner. Therefore, it is recommended to rotate the screwdriver by holding the hexa-bar portion of the driver by thumb and index finger. Remove the lamp housing and mirror cube housing and confirm that the shutter is completely opened.

To return the shutter to the original position, carry out the above procedure in reverse order, and then confirm that the shutter is closed.
B. Universal Vertical Illuminator Mirror Cube Housing (AX-URBC)

C. Universal Vertical Illuminator Collector Lens Unit (AX-URBL)

* To improve the UV340nm transmission ratio, remove the lens inside the top lens by referring to page 5.
D. Cube

- Dichroic mirror, barrier filter, and exciter filter combination as appropriate for the desired excitation method. Never disassemble the cube.

E. Reflected Light Conversion Lens (AX-UCV)

- Filter insertion slot
  - Used with the following filters: CM, commercially available filters (with a diameter of 25 mm and a thickness of 6 mm), inserted into the filter slider.
  - (N:O:ED, 25:O:5)

- Conversion lens control knob

- Reflected light filter sliders

AX-RFSL1

AX-RFSL2
F. Hand Switch for Transmitted/Reflected Light

- Shutter button
- Cube selector buttons
- Locating pin for finding objective buttons
- Reflected light illumination ON/OFF button
- Hand switch connector to HSTR/IS connector on the microscope frame
- RECALL button
- Objective selection buttons
G. Light Source for Fluorescent Light

- Mercury Burner Lamp Housing

- Power Supply Unit for Mercury Burner
5 SUMMARY OF R.L.F. OBSERVATION PROCEDURES

Turn ON the main switch

- Wait 5 to 10 minutes for the arc to stabilize after igniting the burner.
- Power supply unit’s main switch
- Submain switch on microscope frame
- Power supply unit for mercury burner

Engage the mirror cube into the light path

- Select a mirror cube suitable for the observation.
- Hand switch

Select the light path for the observation

- Light path selector knob

Place the specimen

- Specimen holder

Engage the objective

- Hand switch
- Select an objective suitable for the observation.

Open the shutter and bring the specimen into focus

- Hand switch
- Coarse focus/Fine focus adjustment knob
- Binocular tube
- Dipter adjustment
- Eyepiece
- Optical axis adjustment
- Field iris diaphragm knob

Using the collector lens focusing knob, brighten the overall field of view

- Collector lens

Adjust the aperture iris diaphragm and field iris diaphragm

- Aperture iris diaphragm knob
- Field iris diaphragm knob

Observation
USING THE CONTROLS

General Precautions for Observation

1. Confirm that the power supply voltage and frequency match the requirements indicated on the rating plate.
2. Make sure that the power cord and connecting cords are firmly and correctly plugged in.
3. If it is required to perform transmitted light phase contrast or transmitted light differential interference contrast observation alone, leave one cube position on the turret empty. This allows for transmission of white light.
4. During use, the field iris diaphragm should be stopped down so that it is circumscribed by the field of view. If the diaphragm is not centered precisely adjust the control with the centering knobs until so.
5. Always use low fluorescence immersion oil with all immersion objectives.
6. Use of an objective with correction collar, such as UPlanApo40X or PlanApo40X, allows compensation for decreased resolution due to dispersion by the thickness of the cover glass. Correction procedure: While rotating the correction collar, focus with the fine adjustment knob until the position with the best resolution is found. Cover glass thicknesses of 0.11 to 0.23 mm can be corrected for.
7. Use the shutter if observation is interrupted for a short time during the procedure. (Turning the mercury burner on and off repeatedly will significantly shorten the service life of the burner.)
8. Always turn OFF the microscope frame's power supply and the lamp power supply when mounting the reflected light units. When using the mercury burner, if the mirror cube housing is removed without the microscope frame's power supply and the lamp power supply being turned OFF, the operator or other persons may accidentally be exposed to harmful ultraviolet light.
9. The shutter of the light path for reflected light is always open to allow observation, if the microscope frame's power supply is turned ON. It is also possible to force the shutter open even in the condition where the microscope frame's power supply is turned OFF. However, in the condition where the shutter is forced open, NEVER attempt to remove the mirror cube housing, or stop the mirror cube turret at any other position than at a click-stop, while the mercury burner's power supply is turned ON.
10. Never attempt to insert anything but the filter slide into the filter insertion slot. When the lamp is lit, the temperature inside the slot will be very high. If the microscope frame's power supply and the burner's power supply are ON when the reflected light field iris diaphragm unit is removed from the slot, inserting a hand or objects into the empty slot may result in burns or fire.

Turning On the Power

Turn ON the main switch on the power supply unit, the sub-main switch on the microscope frame, and the main switch on the power supply unit for the mercury burner. Between 5 to 10 minutes are required for the arc to stabilize after the burner is ignited.

★ Some mercury burners may not ignite the first time the power is turned ON. If the burner does not ignite, turn the main switch OFF once, then wait 5 to 10 minutes before turning it ON again.
★ To avoid shortening the life of the burner, do not turn the burner off within 15 minutes after ignition.
★ After turning the burner off, it cannot be re-ignited before the mercury vapor cools and condenses to a liquid. Wait for about 10 minutes before restarting the burner.
★ When the lamp housing has to be opened for burner replacement, etc., turn the main switch OFF, and disconnect the connecting cord plug from the output connector on the power supply unit and wait for about 10 minutes until the burner is sufficiently cooled down. (If the lamp housing is accidentally opened while the burner is ignited, the safety interlock will activate and switch off the power automatically. However, avoid this kind of improper operation.)
2 Using the Hand Switch for Transmitted/Reflected Light (Fig. 21)

○ For objective change and operation of the RECALL button, see the AX70/80 instruction manual.
1. To open or close the shutter, press the [SHUT] button ③. When the lamp is lit, the shutter will close. During objective change or mirror cube change, the shutter will automatically close to prevent specimen fading and maintain the safety of the operation.
2. Press the transmitted light illumination ON/OFF switch ② to turn ON/OFF the halogen lamp.
3. Press a mirror cube button ③ to engage the corresponding mirror cube into the light path.
○ If an error occurs during the change operation, the error code display on the microscope frame will show a blinking error code number (and a buzzer will sound). In this event, consult Section V, ERROR CODE CHART and take remedial action in accordance with the instructions given.

3 Centering the Field Iris Diaphragm (Fig. 22)

1. Press the hand switch [SHUT] button to place the shutter in the light path and block the light path.
2. Using the hand switch, engage the B or IB excitation mirror cube into the light path. (If neither of these cubes is available, engage some other mirror cube for fluorescence light use.)
3. Press the hand switch [SHUT] button to remove the shutter from the light path and leave the light path open.
4. Press the button corresponding to the 10X objective on the hand switch to engage this objective.
5. Place a specimen on the stage and bring it into approximate focus.
6. Turn the field iris diaphragm knob ① counterclockwise to stop down the diaphragm diameter to its minimum size.
7. Manipulate the two field iris diaphragm centering knobs ② to adjust the image of the diaphragm to be centered in the field of view.
8. To check centration, open the diaphragm by rotating the field iris diaphragm knob ③ clockwise until the diaphragm image touches the periphery of the field of view. If the image is not centered precisely, center again.
9. Further enlarge the field iris diaphragm diameter to a size where the diaphragm image size is a little larger than when it just circumscribes the field of view.

4 Adjusting the Field Iris Diaphragm (Fig. 22)

To obtain good image contrast, adjust the diameter of the illuminating beam in accordance with the objective in use. The field iris diaphragm may also be used to prevent fading of specimen areas outside the observed field.
Using the field iris diaphragm knob ① on the vertical illuminator, adjust the diaphragm image so that the field of view is circumscribed by the field iris diaphragm in order to exclude stray light.
5 Centering the Mercury Burner
(Fig. 23)

- Place the conversion lens in the light path and center the lens.
- Before attempting to center the burner, wait for the arc to stabilize.
  (5-10 minutes after burner is ignited.)
- Perform the following procedure after centering the field iris diaphragm:
  1. Press the hand switch [SHUT] button to engage the shutter.
  2. Bring the B or T6 excitation mirror cube into the light path. (If neither of these cubes is available, engage some other mirror cube for fluorescence light into the light path.)
  - Do not use the U excitation mirror cube for this adjustment. If it is inevitable to use the U excitation mirror cube, be sure to observe the light through the UV protective shield.
  3. Using the hand switch or U-MCB, engage the 10X objective and remove the shutter from the light path.
  4. While observing through the eyepieces, place the centering target for fluorescence light on the stage. Coincide the (+) mark with the center of the field of view.
  5. Remove the 10X objective.
  6. Rotate the field iris diaphragm knob ① clockwise to open the diaphragm.
  7. Screw the burner focusing knob ② all the way in by turning it clockwise.
  8. Use the centering knob ② to bring the brightest part to the center of the (+) mark.
9. Using the collector lens focusing knob ③, narrow the light beam. (Fig. 24-A)

10. Using the centering knobs ④, bring the bright image to the approximate center position. (Fig. 24-A)

11. Rotate the burner focusing knob ② counterclockwise until the size of the bright spot is minimized. (Fig. 24-B)

12. Using the centering knobs ④, split the bright spot into two arc images. Move the two arc images to the positions shown in Fig. 24-C.

13. Using the burner focusing knob ②, adjust the sizes of the two arc images so that they will be almost identical. (Fig. 24-D)

14. Using the centering knobs ④, superimpose the two arc images. Then turn the collector lens focusing knob ③ so that the arc images will appear as shown in Fig. 24-E. This completes the burner centering adjustment.

Note that even if the arc images deviate a little from the center position, this will not pose any trouble during observation.

15. Before proceeding to the observation, adjust the collector lens focusing knob ③ so that illumination in the field of view is even.

★ To avoid serious injury, never open the lamp housing while the burner is turned on or immediately after it is turned off.

② Recenter the burner each time it is replaced.
6 Reflected Light Conversion Lens (AX-UCV)  (Fig. 26)

When the conversion lens IN/OUT knob is pushed in, the lens is engaged into the light path. When pulled out to the stop position, the lens is removed from the light path.

<table>
<thead>
<tr>
<th>Knob</th>
<th>Observation purpose</th>
</tr>
</thead>
<tbody>
<tr>
<td>IN</td>
<td>Mainly used to emphasize the strength of the central excitation light during fluorescence observation, with auto-fluorescence or fluorochrome, fluorescence antibody work, etc. (Used in combination with the collector lens focusing knob on the lamp housing).</td>
</tr>
<tr>
<td>OUT</td>
<td>Fluorescence observation with uniform field of view. (The illuminated area cannot be stopped down.)</td>
</tr>
<tr>
<td></td>
<td>For reflected light observation methods other than fluorescence light.</td>
</tr>
</tbody>
</table>

★ Note that if a filter is inserted with the mercury burner on for a long time, the filter’s metal frame will become very hot and should not be touched with bare hands.

7 Objectives for Various Observation Methods

<table>
<thead>
<tr>
<th>Objective</th>
<th>Reflected light fluorescence</th>
<th>Phase contrast difference</th>
<th>Transmitted light Nomarski DIC</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>U, V, BV</td>
<td>B, IB, G, IY</td>
<td></td>
</tr>
<tr>
<td>UPlanApo</td>
<td>4 X</td>
<td>O</td>
<td>O**</td>
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<td></td>
<td>10 X</td>
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<td>O**</td>
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<td></td>
<td>20 X</td>
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<td>O**</td>
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<td></td>
<td>40 X</td>
<td>O</td>
<td>O**</td>
</tr>
<tr>
<td></td>
<td>40 X O.I.</td>
<td>O</td>
<td>O**</td>
</tr>
<tr>
<td></td>
<td>100 X O.I.</td>
<td>O</td>
<td>O**</td>
</tr>
<tr>
<td>PlanApo</td>
<td>1.25 X</td>
<td>O</td>
<td>O**</td>
</tr>
<tr>
<td></td>
<td>2 X</td>
<td>O</td>
<td>O**</td>
</tr>
<tr>
<td></td>
<td>40 X</td>
<td>O</td>
<td>O**</td>
</tr>
<tr>
<td></td>
<td>60X O</td>
<td>O</td>
<td>O**</td>
</tr>
<tr>
<td></td>
<td>100 X O</td>
<td>O</td>
<td>O**</td>
</tr>
<tr>
<td>UPlanFl</td>
<td>4 X</td>
<td>O*</td>
<td>O**</td>
</tr>
<tr>
<td></td>
<td>10 X</td>
<td>O*</td>
<td>O**</td>
</tr>
<tr>
<td></td>
<td>20 X</td>
<td>O*</td>
<td>O**</td>
</tr>
<tr>
<td></td>
<td>40 X</td>
<td>O*</td>
<td>O**</td>
</tr>
<tr>
<td></td>
<td>100 X O, O.I.</td>
<td>O*</td>
<td>O**</td>
</tr>
<tr>
<td>Uapo</td>
<td>20 X</td>
<td>O</td>
<td>O**</td>
</tr>
<tr>
<td></td>
<td>40 X</td>
<td>O</td>
<td>O**</td>
</tr>
<tr>
<td></td>
<td>40 X O.I.</td>
<td>O</td>
<td>O**</td>
</tr>
</tbody>
</table>

○: Possible combination
○*: Usable, but image may be dark depending on the NA of the objective.
○**: A phase contrast objective (Phl) is required for phase contrast observation.
UPlan F1100X O.I phase contrast objective (Phl) is not available.
—: Not usable, or combination with no corresponding objective.
8 Selecting a Mirror Cube

Select the mirror cube which matches the fluorochrome in use.

Use in accordance with the band width of the excitation wavelength.

Several excitation filter combinations with different band widths are available. Wide band sets (designated by W) are normally used.

In the following cases, however, other sets will give a better result:

1. Weak fluorescence (only B, G excitations) → Use superwide band (SW)
   (With SWB, strong auto-fluorescence may reduce image contrast.)
2. Samples emitting strong auto-fluorescence → Narrow band (N)
   (Fluorescence brightness is somewhat reduced.)

Cube Dichroic Mirror/Filter Combinations

<table>
<thead>
<tr>
<th>Excitation</th>
<th>Mirror cube</th>
<th>Dichroic mirror</th>
<th>Exciter filter</th>
<th>Barrier filter</th>
<th>Application</th>
</tr>
</thead>
<tbody>
<tr>
<td>U</td>
<td>U-MWU</td>
<td>DM400</td>
<td>BP330-385</td>
<td>BA420</td>
<td>• Auto-fluorescence observation</td>
</tr>
<tr>
<td></td>
<td>U-MNU</td>
<td></td>
<td>BP360-370</td>
<td></td>
<td>• DAPI staining: DNA</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>• Hoechst 33258, 33342 chromosome</td>
</tr>
<tr>
<td>V</td>
<td>U-MNV</td>
<td>DM455</td>
<td>BP400-410</td>
<td>BA455</td>
<td>• Catecholamine observation</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>• Serotonin observation</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>• Tetracyline staining: bone, tooth</td>
</tr>
<tr>
<td>BV</td>
<td>U-MWVB</td>
<td>DM455</td>
<td>BP400-440</td>
<td>BA475</td>
<td>• Quinacrine, quinacrine mustard staining: chromosome</td>
</tr>
<tr>
<td></td>
<td>U-MNBV</td>
<td></td>
<td>BP420-440</td>
<td></td>
<td>• Thioflavine S staining: lymphocyte</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>• Acriflavine staining: nucleic acid</td>
</tr>
<tr>
<td>B</td>
<td>U-MNB</td>
<td>DM500</td>
<td>BP450-480</td>
<td>BA515</td>
<td>• FITC staining: fluorescence antibody</td>
</tr>
<tr>
<td></td>
<td>U-MNB</td>
<td></td>
<td>BP470-480</td>
<td></td>
<td>• Acidine orange staining: DNA, RNA</td>
</tr>
<tr>
<td></td>
<td>U-MSWB</td>
<td></td>
<td></td>
<td></td>
<td>• Auramine staining: tuberculous gran</td>
</tr>
<tr>
<td>IB</td>
<td>U-MWIB</td>
<td>DM505</td>
<td>BP460-490</td>
<td>BA515F</td>
<td>• Texas red staining: fluorescence antibody</td>
</tr>
<tr>
<td></td>
<td>U-MNB</td>
<td></td>
<td>BP470-490</td>
<td></td>
<td></td>
</tr>
<tr>
<td>G</td>
<td>U-MWG</td>
<td>DM570</td>
<td>BP510-550</td>
<td>BA600</td>
<td>• Rhodamine, TRITC staining: fluorescence antibody</td>
</tr>
<tr>
<td></td>
<td>U-MNG</td>
<td></td>
<td>BP530-580</td>
<td></td>
<td>• Propidium iodide staining: DNA</td>
</tr>
<tr>
<td></td>
<td>U-MSWG</td>
<td></td>
<td>BP480-550</td>
<td></td>
<td></td>
</tr>
<tr>
<td>IB</td>
<td>U-MWIG</td>
<td>DM565</td>
<td>BP520-550</td>
<td>BA580F</td>
<td></td>
</tr>
<tr>
<td></td>
<td>U-MAIIY</td>
<td></td>
<td>BP545-580</td>
<td>BA610F</td>
<td></td>
</tr>
</tbody>
</table>

For Pigment, Disjunction Applications

<table>
<thead>
<tr>
<th>Excitation</th>
<th>Mirror cube</th>
<th>Exciter filter</th>
<th>Barrier filter</th>
<th>Application</th>
</tr>
</thead>
<tbody>
<tr>
<td>U</td>
<td>U-MNUA</td>
<td>BP360-370</td>
<td>BA420-460</td>
<td>For observing only the U excitation fluorochrome, when using U excitation together with FITC.</td>
</tr>
<tr>
<td>IB</td>
<td>U-MWIBA</td>
<td>BP460-490</td>
<td>BA515F-550</td>
<td>For observing only FITC, when using FITC and TRITC or Texas red for double staining</td>
</tr>
</tbody>
</table>
### Simultaneous Dual Excitation Cube
(Dual Band Filter Cube)

<table>
<thead>
<tr>
<th>Corresponding pigment</th>
<th>Cube</th>
</tr>
</thead>
<tbody>
<tr>
<td>DAPI/ FITC</td>
<td>U-MDA/ FI</td>
</tr>
<tr>
<td>DAPI/ TRITC</td>
<td>U-MDA/ TRITC</td>
</tr>
<tr>
<td>DAPI/ Texas red, DAPI/ propidium iodide</td>
<td>U-MDA/ TX</td>
</tr>
<tr>
<td>FITC/ TRITC</td>
<td>U-MFI/ TRITC</td>
</tr>
<tr>
<td>FITC/ Texas red, FITC/ propidium iodide</td>
<td>U-MFI/ TX</td>
</tr>
</tbody>
</table>

Made to Order

### Simultaneous Triple Excitation Cube
(Triple Band Filter Cube)

<table>
<thead>
<tr>
<th>Corresponding pigment</th>
<th>Cube</th>
</tr>
</thead>
<tbody>
<tr>
<td>DAPI/ FITC/ TRITC</td>
<td>U-MDA/ F/ TRI</td>
</tr>
<tr>
<td>DAPI/ FOTC/ Texas red, DAPI/ FITC/ propidium iodide</td>
<td>U-MDA/ F/ TX</td>
</tr>
</tbody>
</table>

Made to Order

---

**Meaning of Mirror Cube Name**

### U - M N I B A

- **Common to all cubes**
- **A**: For pigment disjunction applications
- **Excitation**: (U, V, BV, B, IB, G, IG, IY)
- **Bandwidth** (SW: Superwide range, W: Wide range, N: Narrow range)
7-1 Reflected Light Fluorescence Observation

© Also refer to SUMMARY OF REFLECTED LIGHT FLUORESCENCE OBSERVATION PROCEDURES on page 16.
1. Engage the desired mirror cube into the light path.
2. Engage the desired objective into the light path.
3. Open the shutter and focus on the specimen.
4. Adjust the collector lens focusing knob so that both brightness and evenness of illumination in the field of view are optimal.

This unit provides maximum excitation intensity to enable image observation even for specimens with low fluorescence. Therefore, during observation with a high-power objective, fading may occur resulting in loss of image brightness and contrast.
To prevent this, reduce the intensity of the excitation light to some extent to reduce the speed of fading. It is advisable to reduce the intensity of the excitation light by using an ND filter whenever applicable. Also, use the shutter so that the specimen will not be exposed to excitation light for longer than necessary.
By using a commercially available anti-facing agent (e.g. DABCO), fading of the specimen can be slowed. For observation at high magnification, in particular, it is recommended to use an anti-facing agent.
★ Note that anti-facing agents are not applicable to some kinds of specimens.

★ This unit can be used in combination with transmitted light brightfield observation, transmitted light phase contrast observation, and transmitted light differential interference contrast observation, as well as reflected light fluorescence observation.
With specimens that fade rapidly, fading can be minimized by initially using transmitted light phase contrast observation or transmitted light differential interference contrast observation for positioning. Reflected light fluorescence can also be used in combination with phase contrast or differential interference contrast observation, making it easy to detect which portion of the specimen is fluorescing.

7-2 Simultaneous R.L. Fluorescence and T.L. Phase Contrast Observation

★ If the differential interference contrast slider U-DICR and analyser AX-AN/AX-AN380 are attached, remove them from the light path by pulling both out to the click stop.
1. Bring an empty position on the cube turret into the light path.
2. Rotate the phase contrast condenser turret to show the same Ph number as the Ph number of the objective.
   If, for instance, the UPlan20X-Ph1 objective lens is used, the ▲ symbol on the phase contrast condenser turret should also be aligned with "Ph1".
3. Center the condenser ring slit and the objective phase annulus.
4. Engage the mirror cube corresponding to the desired excitation into the light path and open the shutter.
5. Adjust the transmitted light intensity for best balance of fluorescence and phase contrast brightness. Then perform the observation.
★ To adjust the intensity of the transmitted light, use the light intensity dial on the microscope frame or an ND filter.
Simultaneous R.L. Fluorescence and T.L. Nomarski DIC Observation

1. In order to accomplish simultaneous reflected light fluorescence and transmitted light differential interference contrast (DIC) observation, insert the analyzer (AX-AN) into the analyzer insertion slot.
2. Bring an empty position on the cube turret into the light path.
3. Adjust the polarizer on the universal condenser until complete extinction is obtained ("crossed Nicol" position).
4. Insert the transmitted light DIC slider into the slot provided the revolving nosepiece.
5. Rotate the universal condenser turret to select the Nomarski prism matching the objective to be used.
6. Engage the objective to be used.
7. Place the specimen on the stage and bring the specimen into focus.
8. Adjust the field iris diaphragm of the transmitted light illumination unit (built into the microscope base) and the aperture iris diaphragm of the universal condenser.
9. Turn the prism shift knob on the transmitted light DIC slider to adjust for best contrast of the differential interference contrast image.
10. Engage the mirror cube corresponding to the desired excitation into the light path and open the shutter.
11. Adjust the transmitted light intensity for optimal fluorescence and differential interference contrast image brightness.
1. U excitation cube (Wide band)
   **U-MWU**

2. U excitation cube (Narrow band)
   **U-MNU**

3. V excitation cube (Narrow band)
   **U-MNV**

4. BV excitation cube (Wide band)
   **U-MWBV**

5. BV excitation cube (Narrow band)
   **U-MNBV**

6. B excitation cube (Wide band)
   **U-MWB**
For fluorochrome emission, a light beam having a specific wavelength is selected from a wide spectrum of wavelengths. The five major peaks of luminance are at wavelengths of 365/366, 434.7, 546.1, and 577.0/579.1 nm. In addition, light beams having wavelengths of 334.2 and 490 nm (with rather low luminance) are also applicable to fluorochrome emission.
II. REFLECTED LIGHT OBSERVATION MODES

By equipping the AX70 or AX80 (transmitted light observation configuration) with UIS objectives for opaque objects, mirror cubes for reflected light (UMBF, U-MDF, U-MDIC), a differential interference contrast slider (UDICR), an aperture iris diaphragm unit for reflected light, etc., these microscopes can accommodate various reflected light observation methods.

1 CONTROLS

A. Light Source for Reflected Light

- 100W halogen lamp housing (AX-LH100)
  - Similar to the halogen lamp housing for transmitted light.

- Power supply unit for halogen lamp (TH3)
  - Not required except for simultaneous transmitted light and reflected light observation.
B. AX Reflected Light Filter Changer (AX-RFCH)

To produce better color reproduction and remain unaffected by other filters, 25FR is the AX-RFSL1, and 26LBD is placed in the AC-RFSL2.

Reflected light filter sliders

AX-RFSL1
AX-RFSL2
AX-RFSL3

Filter insertion slot
Used with the following filter, or commercially available. Filters with a diameter of 25 mm and a thickness of 8 mm, inserted into the filter slider:
25ND6, 25ND25, 25LBD, 25LFS50-25FR

C. Aperture Iris Diaphragm Unit for Reflected Light (AX-RAS)

Aperture iris diaphragm knob

Aperture iris diaphragm centering knob
2-1 Assembly Diaphragm

The diagram below shows how to assemble the various components for reflected light observation methods. For assembly details other than those shown in the diagram below, refer to the instructions for assembly of the various components for reflected light fluorescence observation (page 3) of the instruction manual for the AX70 or AX80 microscope frame.

★ When assembling the microscope, make sure that all parts are free of dust and dirt, and avoid scratching any parts or touching the glass surfaces. Confirm that all components are disconnected from the AC mains before assembling.
2-2 Detailed Assembly Procedure

1 Mounting the Mirror Cube

1. Corresponding to the observation methods, mount the correct mirror cube on the mirror cube turret. (For details on mounting, see page 4, section 1.)

<table>
<thead>
<tr>
<th>Observation method</th>
<th>Cube name</th>
<th>Cube code</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reflected light brightfield observation</td>
<td>BF</td>
<td>U-M/BF</td>
</tr>
<tr>
<td>Reflected light darkfield observation</td>
<td>DF</td>
<td>U-M/DF</td>
</tr>
<tr>
<td>Reflected light Nomarski differential interference contrast observation</td>
<td>BF (or DIC)</td>
<td>U-M/BF (or DIC)</td>
</tr>
<tr>
<td>Reflected light simple polarized light observation</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

2. Affix the cube sticker to the hand switch.

2 Mounting the Light Excluding Adapter (DF Insert) (Fig. 26)

1. Remove the mirror cube housing.
2. Aligning the positioning groove of the DF (darkfield) insert and the pin on the microscope frame, mount the DF insert.
3. Return the mirror cube housing to its original position.

Fig. 26

3 Mounting the Aperture Iris Daphragm Unit (Fig. 27)

1. Insert gradually the aperture iris daphragm unit ① into the aperture iris daphragm unit mounting slot ② on the microscope frame (right side) by sliding the unit onto the guide dovetail.
2. Insert clamping screws into the two mounting holes ③ and tighten the screws with the Allen screwdriver.

Fig. 27

4 Installing the Halogen Lamp

Refer to the explanation on page (Section 5) in the AX70 instruction manual, or page 6 (Section 5) in the AX80T instruction manual No. 1 “ASSEMBLY”.

34
5 Attaching the Lamp Housing

Refer to the explanation on page 5 (Section 6) in the AX70 instruction manual, or page 8 (Section 6) in the AX30T instruction manual No. 1 "ASSEMBLY". However, the lamp housing should be mounted on the upper mounting collar so the connector should be plugged into the power outlet marked.

★ If the power supply unit TH3 is used for the halogen lamp, plug the connector into the power outlet on the rear of the TH3.

6 Others

How to mount the analyzer and the polarizer, etc. is explained during the outline of the observation methods on the following pages.

7 Centering and Adjusting the Aperture Iris Diaphragm  (Fig. 28)

1. Using the hand switch, engage the BF mirror tube into the light path.
2. Press the hand switch SHUT button to remove the shutter from the light path and leave the light path open.
3. Press the hand switch button to engage the 10X objective. Place a highly reflective specimen on the stage and bring it into approximate focus.
4. Remove the eyepieces. Looking at the objective pupil inside the observation tube, turn the aperture iris diaphragm knob ① counterclockwise to leave the diaphragm stopped down to approximately 70 – 80% of the objective pupil diameter.
5. At this point, if the diaphragm is not centered precisely, center it again by manipulating the two aperture iris diaphragm centering knobs ②. Adjust in the same manner as for the field iris diaphragm. (The focusing magnifier UCT30 facilitates this adjustment.)

Adjusting the Aperture Iris Diaphragm

Adjust the numerical aperture of the illumination system to control the brightness of the observed image.

② Depending on the specimen, an image with good contrast and little flare may sometimes be obtained by keeping the aperture iris diaphragm stopped down a little. Please experiment with this to see if it works with the particular specimen.
The following is a summary of basic reflected light brightfield observation.

1. Turn ON the main switch
2. Engage the LBD filter
3. Select the brightfield light path
4. Select the light path for observation
5. Place specimen
6. Engage the 10X objective
7. Interpupillary distance adjustment
8. Diopter adjustment
9. Optical axis adjustment
10. Focus on the specimen
11. Engage the desired objective
12. Adjust aperture iris diaphragm and field iris diaphragm

Observation

Main switch
Submain switch on microscope frame
Switch to reflected light, or Main switch on p.18
Reflected light filter changer on p.35
Hand switch on p.19
Light path selector knob
Specimen clamp
Hand switch on p.19
Binocular tube
Eye piece
Aperture iris diaphragm knob on p.20
Field iris diaphragm knob on p.20
Coarse focus/Fine focus adjustment knob
Coarse focus/Fine focus adjustment knob
Aperture iris diaphragm knob on p.20
Field iris diaphragm knob on p.19
4 REFLECTED LIGHT OBSERVATION METHODS

4-1 Reflected Light Brightfield/Darkfield Observation

1. Selecting the Light Path for Observation (Fig. 29)

   Use the hand switch to select and engage the mirror cube (BF or DF) corresponding to the desired observation method.

<table>
<thead>
<tr>
<th>Cube code</th>
<th>Field iris diaphragm</th>
<th>Aperture iris diaphragm</th>
<th>Antiglare ND filter</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reflected light brightfield</td>
<td>BF</td>
<td>Adjustable as necessary</td>
<td>IN</td>
</tr>
<tr>
<td>Reflected light darkfield</td>
<td>DF</td>
<td>Open</td>
<td></td>
</tr>
</tbody>
</table>

2. Anti-Glare ND Filter (AX-DND) (Fig. 30)

   1. Insert the antiglare ND filter (3) into the filter slot on the right side of the vertical illuminator.
   2. As the filter is inserted, two clicks will be heard. At the first, the filter is in the empty position; at the second, the ND is engaged in the light path.
   3. Ordinarily, if this ND filter is in the light path, it will prevent the glare effect otherwise noticeable when switching from darkfield to brightfield.
   4. When the illumination intensity is too low during brightfield observation or to shorten the exposure time during photomicrography, or to brighten the field of view during darkfield observation, remove the filter from the light path.

3. Installing the Filters (Fig. 31)

   1. Mount one or more of the following filters, or commercially available filters (25 mm diameter), on the filter slider:
   2. Drop the filter (2) into the mount on the filter slider (1). Insert the retaining ring (3) to hold the filter in place.
   3. To replace a filter, push it out of the slider from below.
   * Only the frosted filter 25FR should be mounted on the AX-RFSL1 slider closest to the objective side.
   4. At the first click-stop, the filter is in the empty position, and at the second and third click-stop, the required filter is engaged into the light path.

<table>
<thead>
<tr>
<th>Usable filters</th>
<th>Applications</th>
</tr>
</thead>
<tbody>
<tr>
<td>25FR Frosted filter</td>
<td>To ensure even illumination.</td>
</tr>
<tr>
<td>25LBD Color temperature conversion filter</td>
<td>To convert the color temperature of the halogen source to the color temperature of daylight. Used for observation and when taking color photographs.</td>
</tr>
<tr>
<td>25IF550 Green filter</td>
<td>To enhance contrast during B&amp;W observation. Used when taking B&amp;W photographs.</td>
</tr>
<tr>
<td>25ND25 Neutral density filter</td>
<td>To attenuate light intensity. (Transmission ratio 25%)</td>
</tr>
<tr>
<td>25ND8 Neutral density filter</td>
<td>To attenuate light intensity. (Transmission ratio 8%)</td>
</tr>
</tbody>
</table>
4-2 R.L. Nomarski Differential Interference Contrast Observation

1. Selecting the Light Path for Observation (Fig. 33)

1. Using the hand switch, press the BF or DIC button to engage the required mirror cube.
2. When the U-MDIC differential interference contrast mirror cube is inserted in the mirror cube housing, engage this mirror cube into the light path. With the U-MDIC mirror cube inserted in the cube cassette, it is not necessary to mount and adjust the analyzer and polarizer.
3. Insert the analyzer (AX-AN360) 1 and the polarizer (AX-PC) 2 to engage them both into the light path.
4. Rotate the analyzer polarizer dial 3 until complete extinction is obtained ("crossed Nicols" position).

2. Installing the Nomarski Prism (Fig. 34)

1. Loosen the clamping screw 1 at the front of the revolving nosepiece, and insert the U-DICR differential interference contrast slider 2 with the slide with the inscription facing upward. Tighten the clamping screw to secure the prism.
2. If a LMPPlan objective is used, push in the selector lever 3. If an LMPon- objective is used, pull out the selector lever.

3. Observation Method (Fig. 34)

1. Place the specimen on the stage and move the stage to bring the specimen into focus.
2. Adjust the field iris diaphragm until the diaphragm opening circumscribes the field of view.
3. Stop down the aperture iris diaphragm somewhat may increase the contrast.
4. Rotate the prism control knob 4 of the DIC prism slider to adjust the interference color of the background, and to achieve maximum contrast depending on the specimen under observation, as outlined below:
   a. Rotating the prism control knob of the slider will continuously change the interference color of the background from gray to magenta (100 - 600 nm).
      • If the background color is black (0-order fringe), central field-like observation is possible.
      • If the background color is gray, a pseudo relief image with maximum contrast with gray sensitivity can be obtained.
      • If the background color is magenta, even a minor optical retardation can be observed as a color change.
   b. Care should be taken to keep the specimen surface clean, as any small amount of contamination on the surface may show up due to the exceptionally high sensitivity of the differential interference contrast method.
   c. As differential interference contrast exhibits directional sensitivity, rotate the stage or the specimen to obtain best contrast.

II-4 REFLECTED LIGHT OBSERVATION METHODS
Switching Between Brightfield and Darkfield Observation

1. Loosen the clamping screw (1) at the front of the revolving nosepiece, and gently pull the U-DICR differential interference contrast slider (2) outward until a click is heard. Tighten the clamping screw again.
2. Press BF or DF on the hand switch to disengage the U-MDIC mirror cube. If used, disengage both the analyzer and the polarizer from the light path.

4-3 Reflected Light Simple Polarized Light Observation

To prepare for simple polarized light observation using the vertical illuminator, perform step 1 in Section 4-2, Reflected Light Nomarski Differential Interference Contrast Observation, outlined on page 36.

Observation

1. Place the specimen on the stage and use the coarse and fine adjustment knobs to move the stage up and down to bring the specimen into focus. Simple polarized light observation is now possible.
2. Adjust the field iris diaphragm until the diaphragm opening circumscribes the field of view.
3. Stopping down the aperture iris diaphragm may increase the contrast somewhat.
## III. SPECIFICATIONS

<table>
<thead>
<tr>
<th>Item</th>
<th>Specification</th>
</tr>
</thead>
</table>
| Optical system | UIS (Universal Infinity System) optical system  
Compatible microscope frame: AX70/AX80 |
| Reflected light illumination | Type with reflected light illumination system installed in the microscope arm  
Observation tube magnification: 1X (super widefield applicable: Field number 26.5)  
Observation mode selection: Turret system (max: four cubes)  
Possible observation modes:  
- Reflected light fluorescence*  
- Reflected light brightfield  
- Reflected light darkfield  
- Reflected light Nomarski differential interference contrast  
- Reflected light simple polarized light  
* Using the AX70/AX80 transmitted light illuminated, simultaneous transmitted light Nomarski differential interference contrast observation or transmitted light phase contrast observation becomes possible. |
| Mercury burner lamp housing  
U-ULH  
U-ULS100HG  
U-UCLHG/EXEB | DC 100W high pressure mercury burner USH102D (mfd. Ushio Electric)  
Interlock mechanism (microswitch)  
Burner centering: Up/down, left/right centering knob system and focusing knob system |
| Power supply unit for mercury burner BH2-RFL-T3 | Auto ignition system  
Life time hour counter (displays accumulated hours of operation)  
Ratings: 100 – 120 VAC  50 – 60 Hz  
220 – 240 VAC  50 – 60 Hz, 2.8A |
| 100W halogen lamp housing AX-LH100 | DC 12V 100W halogen bulb (pre-centered)  
12V 100WHAL-L halogen bulb (Philips 7724) |
| Power supply for halogen lamp TH3 | 100 – 120V AC, 50 – 60 Hz, 2.5A |
| Transmitted/reflected light hand switch | Push button type  
- Objective selection buttons (six) and recall button  
- Cube selection buttons (four)  
- Shutter button, reflected light illumination ON/OFF switch (one each) |
| Operating environment | Temperature: 0°C – 40°C (32°F – 104°F). Relative humidity: 30 – 90%.  
Pollution degree II (In accordance with IEC664)  
Installation category (overvoltage category) II (In accordance with IEC664) |
### IV. OPTICAL CHARACTERISTICS

<table>
<thead>
<tr>
<th>Objective designation</th>
<th>Magnification</th>
<th>N.A.</th>
<th>W.D. (mm)</th>
<th>Cover glass thickness</th>
<th>Resolution (μm)</th>
<th>Eyepiece</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>SW-H10X (26.5)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>UV/Plan F1</td>
<td>5X</td>
<td>0.15</td>
<td>20.0</td>
<td>—</td>
<td>2.24</td>
<td>50X</td>
<td>59.9</td>
</tr>
<tr>
<td>Universal Plan</td>
<td>10X</td>
<td>0.30</td>
<td>10.1</td>
<td>0</td>
<td>1.12</td>
<td>100X</td>
<td>14.7</td>
</tr>
<tr>
<td>Semi-Apochromat</td>
<td>20X</td>
<td>0.46</td>
<td>3.1</td>
<td>0</td>
<td>0.73</td>
<td>200X</td>
<td>5.10</td>
</tr>
<tr>
<td>(FN26.5)</td>
<td>40X</td>
<td>0.75</td>
<td>0.63</td>
<td>0</td>
<td>0.45</td>
<td>400X</td>
<td>1.68</td>
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<tr>
<td></td>
<td>50X</td>
<td>1.60</td>
<td>0.66</td>
<td>0</td>
<td>0.42</td>
<td>500X</td>
<td>1.30</td>
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<tr>
<td></td>
<td>100X</td>
<td>3.60</td>
<td>0.31</td>
<td>0</td>
<td>0.36</td>
<td>1000X</td>
<td>0.67</td>
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<tr>
<td>UV/Plan FL-BD</td>
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<td>0.15</td>
<td>12.0</td>
<td>—</td>
<td>2.24</td>
<td>50X</td>
<td>59.9</td>
</tr>
<tr>
<td>Universal Plan</td>
<td>10X</td>
<td>0.30</td>
<td>6.5</td>
<td>—</td>
<td>1.12</td>
<td>100X</td>
<td>14.7</td>
</tr>
<tr>
<td>Semi-Apochromat</td>
<td>20X</td>
<td>0.46</td>
<td>3.0</td>
<td>0</td>
<td>0.73</td>
<td>200X</td>
<td>5.10</td>
</tr>
<tr>
<td>for Bright/Darkfield</td>
<td>50X</td>
<td>0.83</td>
<td>0.66</td>
<td>0</td>
<td>0.45</td>
<td>500X</td>
<td>1.30</td>
</tr>
<tr>
<td>(FN26.5)</td>
<td>100X</td>
<td>3.60</td>
<td>0.31</td>
<td>0</td>
<td>0.37</td>
<td>1000X</td>
<td>0.73</td>
</tr>
<tr>
<td>UV/Plan FL-BDP</td>
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<td>0.15</td>
<td>12.0</td>
<td>—</td>
<td>2.24</td>
<td>50X</td>
<td>59.9</td>
</tr>
<tr>
<td>Universal Plan</td>
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<td>0.25</td>
<td>6.5</td>
<td>—</td>
<td>1.24</td>
<td>100X</td>
<td>18.4</td>
</tr>
<tr>
<td>Semi-Apochromat</td>
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<td>3.0</td>
<td>0</td>
<td>0.84</td>
<td>200X</td>
<td>6.09</td>
</tr>
<tr>
<td>for Reflected light</td>
<td>50X</td>
<td>0.76</td>
<td>0.66</td>
<td>0</td>
<td>0.45</td>
<td>500X</td>
<td>1.42</td>
</tr>
<tr>
<td>Polarized light</td>
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<td>3.60</td>
<td>0.31</td>
<td>0</td>
<td>0.37</td>
<td>1000X</td>
<td>0.73</td>
</tr>
<tr>
<td>UV/Plan F1</td>
<td>20X</td>
<td>0.40</td>
<td>12.0</td>
<td>0</td>
<td>0.84</td>
<td>200X</td>
<td>6.09</td>
</tr>
<tr>
<td>Long Working Distance</td>
<td>50X</td>
<td>0.80</td>
<td>10.6</td>
<td>0</td>
<td>0.47</td>
<td>500X</td>
<td>2.50</td>
</tr>
<tr>
<td>Plan Apochromat</td>
<td>100X</td>
<td>3.40</td>
<td>3.4</td>
<td>0</td>
<td>0.37</td>
<td>1000X</td>
<td>0.87</td>
</tr>
<tr>
<td>(FN26.5)</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>UV/Plan FL-BD</td>
<td>20X</td>
<td>0.40</td>
<td>12.0</td>
<td>0</td>
<td>0.84</td>
<td>200X</td>
<td>6.09</td>
</tr>
<tr>
<td>Long Working Distance</td>
<td>50X</td>
<td>0.80</td>
<td>10.6</td>
<td>0</td>
<td>0.42</td>
<td>500X</td>
<td>2.50</td>
</tr>
<tr>
<td>Plan Apochromat</td>
<td>100X</td>
<td>3.30</td>
<td>3.3</td>
<td>0</td>
<td>0.35</td>
<td>1000X</td>
<td>1.04</td>
</tr>
<tr>
<td>for Bright/Darkfield</td>
<td>(FN26.5)</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>M/Plan Apo Long</td>
<td>50X</td>
<td>0.95</td>
<td>0.3</td>
<td>0</td>
<td>0.35</td>
<td>500X</td>
<td>1.04</td>
</tr>
<tr>
<td>Working Distance Plan</td>
<td>100X</td>
<td>1.40</td>
<td>0.08</td>
<td>0</td>
<td>0.24</td>
<td>1000X</td>
<td>0.59</td>
</tr>
<tr>
<td>Apochromat (FN26.5)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LM/Plan Apo Long</td>
<td>250X</td>
<td>0.9</td>
<td>0.80</td>
<td>0</td>
<td>0.37</td>
<td>2500X</td>
<td>0.50</td>
</tr>
<tr>
<td>Working Distance Plan</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Apochromat for Bright</td>
<td>250X</td>
<td>0.9</td>
<td>0.80</td>
<td>0</td>
<td>0.37</td>
<td>2500X</td>
<td>0.50</td>
</tr>
<tr>
<td>field/26.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
## V. ERROR CODE CHART

If an anomalous condition is present, a blinking error code number will be shown on the AX70 microscope frame’s voltage indicator display and a buzzer will sound. Consult the following error code chart and take remedial action in accordance with the instructions given.

<table>
<thead>
<tr>
<th>Code</th>
<th>Error condition</th>
<th>Remedy</th>
</tr>
</thead>
<tbody>
<tr>
<td>01·02</td>
<td>Improperly revolving nosepiece rotation. (Occurs if anything impedes the rotation of the revolving nosepiece.)</td>
<td>Remove the obstruction. Switch the power OFF and then ON again before attempting the operation again. If the same error occurs again, something is wrong with the equipment. In this event, contact your local Olympus representative for assistance.</td>
</tr>
<tr>
<td>11</td>
<td>Mirror cube failing to stop correctly at click-stop.</td>
<td></td>
</tr>
<tr>
<td>12·13</td>
<td>Sensor failing to detect mirror cube at click-stop.</td>
<td></td>
</tr>
<tr>
<td>20·21</td>
<td>Shutter IN/OUT operation error.</td>
<td></td>
</tr>
</tbody>
</table>
| 22    | Shutter clamped in OUT condition.                    | 1. When the shutter opening screw is loosened, the error code disappears and the shutter IN/OUT operation can be performed.  
2. Neglect the buzzer if the shutter is deliberately opened to accommodate a particular observation mode. To silence the buzzer, turn the microscope frame’s main switch OFF. However, this will render the hand switch inoperative so that the operation should be performed manually. |
| 23·24 | Abnormal condition of the OPTION connector.         | If the OPTION connector is in use, turn OFF the power and then check the seat of the connection cable. Try to operate the microscope again. If the same error occurs again, or if the error code appears when the connector is not in use, something is wrong with the equipment. In this event, contact your local Olympus representative for assistance. |
| 30    | Internal circuit malfunction.                        | Something is wrong with the equipment. Contact your local Olympus representative for assistance. |
## VI. TROUBLESHOOTING GUIDE

Under certain conditions, performance of this unit may be adversely affected by factors other than defects. If problems occur, please review the following list and take remedial action as appropriate. If you cannot solve the problem after checking the entire list, please contact your local Olympus representative for assistance.

### Fluorescence Light Observation

<table>
<thead>
<tr>
<th>Problem</th>
<th>Cause</th>
<th>Remedy</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Optical System</td>
<td></td>
<td></td>
</tr>
<tr>
<td>a. Lamp lights, but field of view remains dark.</td>
<td>The shutter is closed.</td>
<td>Open the shutter.</td>
</tr>
<tr>
<td></td>
<td>ND filter is engaged.</td>
<td>Remove the ND filter from the light path.</td>
</tr>
<tr>
<td></td>
<td>The aperture and/or field iris diaphragm is not correctly opened.</td>
<td>The aperture iris diaphragm should be completely opened. The field iris diaphragm should be opened to the extent that the diaphragm image circumscribes the field of view.</td>
</tr>
<tr>
<td></td>
<td>The mirror cube is not suitable for the specimen.</td>
<td>Change to a suitable mirror cube.</td>
</tr>
<tr>
<td>b. Image is unclear, blurred or has insufficient contrast.</td>
<td>Objectives and/or filters are dirty.</td>
<td>Clean.</td>
</tr>
<tr>
<td></td>
<td>The aperture and/or field iris diaphragm is not correctly opened.</td>
<td>The aperture iris diaphragm should be completely opened. The field iris diaphragm should be opened to the extent that the diaphragm image circumscribes the field of view.</td>
</tr>
<tr>
<td></td>
<td>The mirror cube is not suitable for the specimen.</td>
<td>Change to a suitable mirror cube.</td>
</tr>
<tr>
<td>c. Field of view is obscured, or field of view is not evenly illuminated.</td>
<td>The field iris diaphragm is stopped down too far.</td>
<td>Open the field iris diaphragm sufficiently.</td>
</tr>
<tr>
<td></td>
<td>ND filter is not at click-stop.</td>
<td>Insert the ND filter until it stops at the click-stop.</td>
</tr>
<tr>
<td></td>
<td>Improper centering or focusing of the mercury burner.</td>
<td>Center or focus the mercury burner correctly.</td>
</tr>
<tr>
<td>d. Dark spots visible in the field of view.</td>
<td>Dust/dirt adhering to the burner or the side of the collector lens facing the lamp.</td>
<td>Clean.</td>
</tr>
<tr>
<td>e. The real bulb image and the mirror image cannot be focused.</td>
<td>The collector lens clamping screw is loose.</td>
<td>Tighten firmly.</td>
</tr>
<tr>
<td>f. A tingling sensation is felt when the microscope frame is touched.</td>
<td>The microscope is not correctly grounded.</td>
<td>Make sure the power cord (or ground wire) is correctly grounded.</td>
</tr>
<tr>
<td>2. Electrical System</td>
<td></td>
<td></td>
</tr>
<tr>
<td>a. The main switch indicator does not light up.</td>
<td>Improper connection of the power cord.</td>
<td>Connect the power cord correctly.</td>
</tr>
<tr>
<td>b. Power switch indicator lights, but the mercury burner does not ignite.</td>
<td>Improperly connected connectors.</td>
<td>Connect correctly.</td>
</tr>
<tr>
<td></td>
<td>The burner is not installed.</td>
<td>Install the burner.</td>
</tr>
<tr>
<td></td>
<td>The lamp housing interlock mechanism is activated.</td>
<td>Tighten the burner socket clamping screw.</td>
</tr>
<tr>
<td></td>
<td>Auto ignition is malfunctioning.</td>
<td>Turn OFF the main switch on the power supply. Turn ON again. (Repeat as necessary.)</td>
</tr>
<tr>
<td>c. The burner flickers or is dark.</td>
<td>Insufficient time has elapsed since the burner was turned on.</td>
<td>Wait for at least 10 minutes after igniting the burner.</td>
</tr>
<tr>
<td></td>
<td>The burner life has expired.</td>
<td>Replace the mercury burner when the hour counter reading exceeds 200 hours. (Burner service life is 200 hours.)</td>
</tr>
<tr>
<td>Problem</td>
<td>Cause</td>
<td>Remedy</td>
</tr>
<tr>
<td>---------</td>
<td>-------</td>
<td>--------</td>
</tr>
<tr>
<td>a. Lamp is turned on, but field of view remains dark.</td>
<td>Lamp for reflected light system is not turned on.</td>
<td>Turn on the lamp.</td>
</tr>
<tr>
<td></td>
<td>When in reflected light, darkfield observation, the aperture and field iris diaphragms are stopped down.</td>
<td>Open the aperture and field iris diaphragm.</td>
</tr>
<tr>
<td></td>
<td>Mirror cube is not installed.</td>
<td>Install mirror cube suitable for the observation.</td>
</tr>
<tr>
<td>b. Field of view is obscured, or field of view is not evenly illuminated.</td>
<td>The field iris diaphragm is not properly centered.</td>
<td>Center the field iris diaphragm correctly.</td>
</tr>
<tr>
<td></td>
<td>The field iris diaphragm is stopped down too far.</td>
<td>Open the field iris diaphragm sufficiently.</td>
</tr>
<tr>
<td></td>
<td>The bulb is not centered correctly.</td>
<td>Center the bulb correctly.</td>
</tr>
<tr>
<td></td>
<td>Frosted filter is engaged into the light path.</td>
<td>Remove the frosted filter from the light path.</td>
</tr>
<tr>
<td></td>
<td>Filters are not engaged correctly.</td>
<td>Engage the filters correctly in the light path.</td>
</tr>
<tr>
<td>c. The image shows diffraction.</td>
<td>The aperture iris diaphragm is stopped down too far.</td>
<td>Open the aperture iris diaphragm to the correct diameter.</td>
</tr>
<tr>
<td>d. Visibility is poor.</td>
<td>Non-US series objective is used.</td>
<td>Use only US-series objectives with this microscope.</td>
</tr>
<tr>
<td></td>
<td>Image is not sharp.</td>
<td>Clean the objective.</td>
</tr>
<tr>
<td></td>
<td>Contrast is poor.</td>
<td>Use immersion oil.</td>
</tr>
<tr>
<td></td>
<td>Details are indistinct.</td>
<td>Use recommended immersion oil.</td>
</tr>
<tr>
<td>e. Part of the image is blurred.</td>
<td>The specimen is not mounted correctly on the stage.</td>
<td>Place the specimen correctly on top of the stage and secure it with the specimen holder.</td>
</tr>
</tbody>
</table>