Nikon

Inverted Microscope

DIAPHOT-TMD

Instructions

NIKON CORPORATION

CAUTIONS

1 Avoid sharp knocks!
Handle the microscope gently, taking care to avoid sharp knocks.

2 When carrying the microscope
When carrying the microscope, support its base inserting both hands into the hollows on the right and left sides of the base.
(The instrument weighs 26 kg.)

3 Place for using
Avoid the use of the microscope in a dusty place, where it is subject to vibrations or exposed to high temperatures, moisture or direct sunlight.

4 Power source voltage
In every case, make sure of the power source voltage by means of the input voltage change-over device (fuse holder) on the rear of the base.
(Refer to P.9, (12).)

5 Light source
Halogen lamp bulb to be used is 12V-50W.
Do not use 12V-100W halogen lamp bulb.
If the lamp bulb of over-rated wattage is used, light adjusting circuit will damage.

6 In lighting the lamp
Take care not to touch the lamp housing being lighted, and don't bring inflammable substances such as gasoline, thinner, and alcohol near to the lamp housing, as some parts of the lamp housing may take a high temperature while the lamp is being lighted.

7 Exchanging the lamp bulb and fuse
Before replacing the lamp bulb or fuse, turn OFF the power switch and disconnect the plug of the power source cord.
In such cases as of replacement, do not touch the lamp bulb with bare hands, immediately after putting out the lamp.

8 Dirt on the lens
Do not leave dust, dirt or finger marks on the lens surfaces.
They will prevent the user from clear observation of the specimen image.

9 Focus knobs
Never attempt to adjust the tightness of the right- and lefthand focus knobs by turning the one, while holding the other in this model microscope, because of causing disorder.

CARE AND MAINTENANCE

1 Cleaning the lenses

To clean the lens surfaces, remove dust using a soft brush or gauze. Only for removing finger marks or grease, should soft cotton cloth, lens tissue or gauze lightly moistened with absolute alcohol (methyl alcohol or ethyl alcohol) be used. For cleaning the objectives use only xylene.

Observe sufficient caution in handling alcohol and xylene.

2 Cleaning the painted surfaces

Avoid the use of any organic solvent (for example, thinner, ether, alcohol, xylene etc.) for cleaning the painted surfaces and plastic parts of the instrument.

3 Never attempt to dismantle!

Never attempt to dismantle the instrument so as to avoid the possibility of impairing the operational efficiency and accuracy.

4 When not in use

When not in use, cover the instrument with the accessory vinyl cover, and store it in a place free from moisture and fungus.

It is especially recommended that the objectives and eyepieces be kept in an airtight container containing desiccant.

5 Periodical checking

To maintain the performance of the instrument, we recommend to check the instruments periodically. (For details of this check, contact our agency.)

CONTENTS

I. NOMENCLATURE

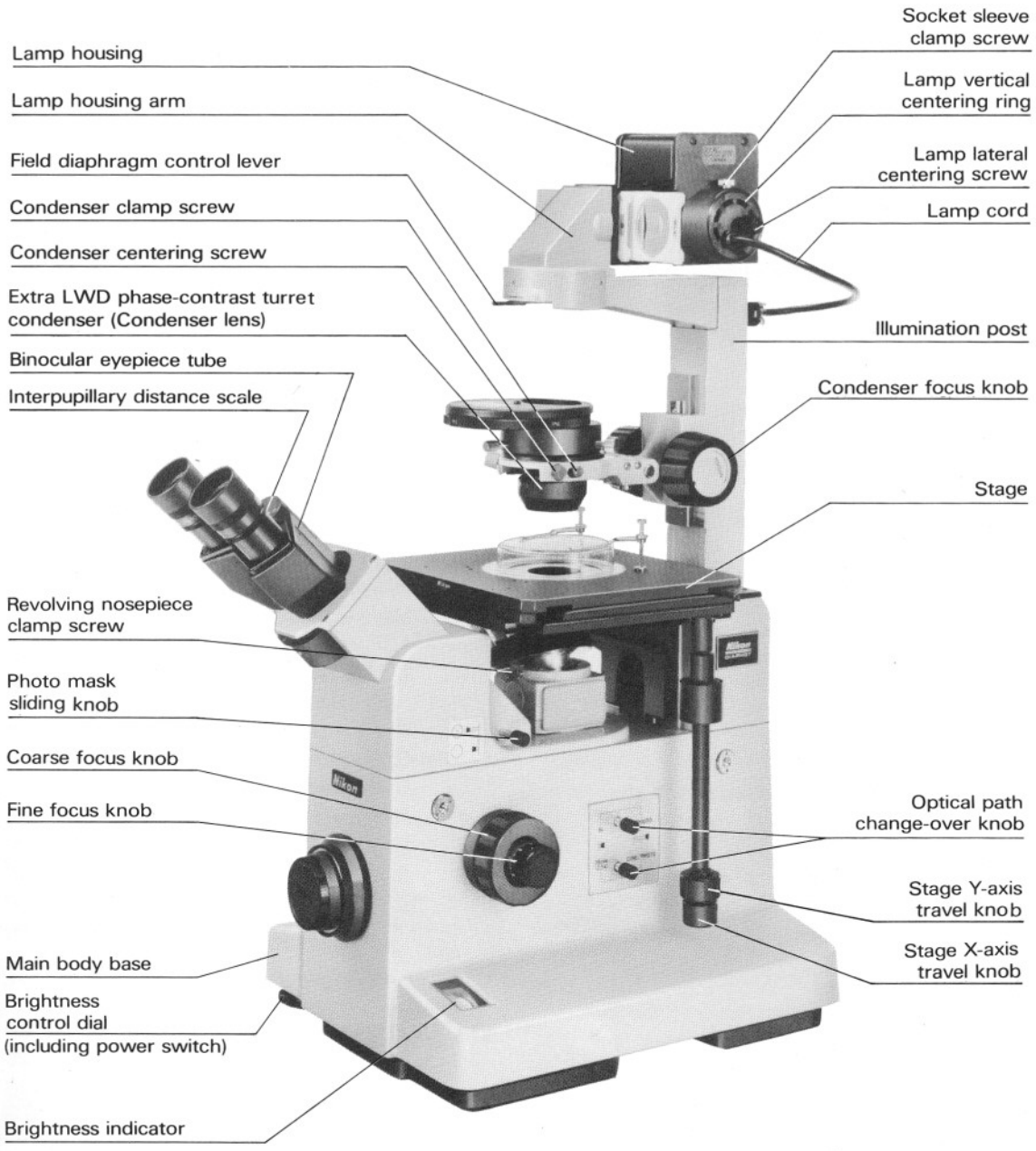


Fig. 1

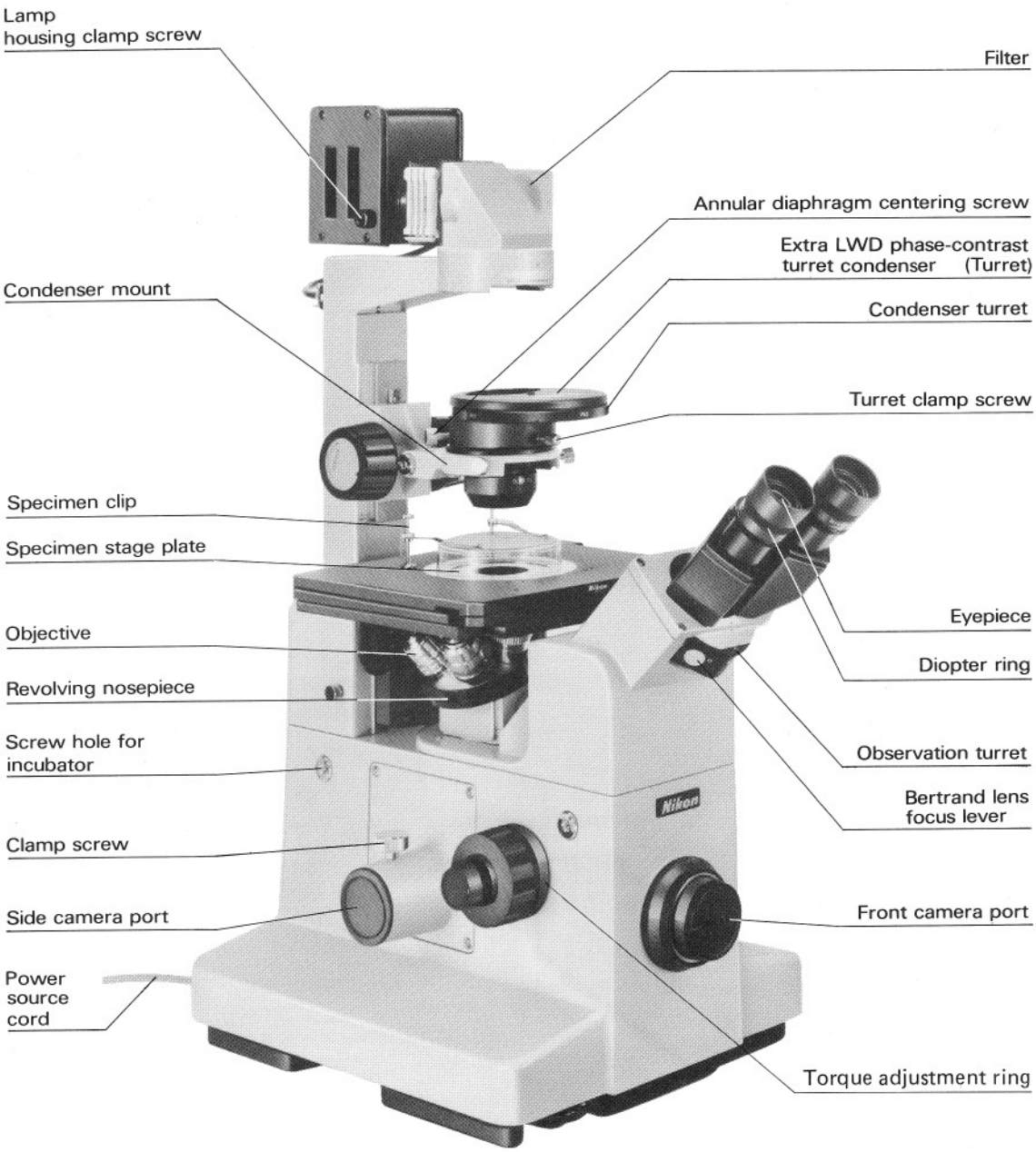

Lamp
housing clamp screw
Filter
Annular diaphragm centering screw
Extra LWD phase-contrast
turret condenser (Turret)
Condenser mount
Condenser turret
Turret clamp screw
Specimen clip
Specimen stage plate
Eyepiece
Objective
Diopter ring
Revolving nosepiece
Observation turret
Screw hole for
incubator
Bertrand lens
focus lever
Clamp screw
Front camera port
Side camera port
Power
source
cord
Torque adjustment ring

Fig. 2

II. ASSEMBLING

Assemble the following units in the order of their numbers given as below:
For the methods of attaching the units, refer to the accounts given in connection with the figures on P. 7 ~ 9.

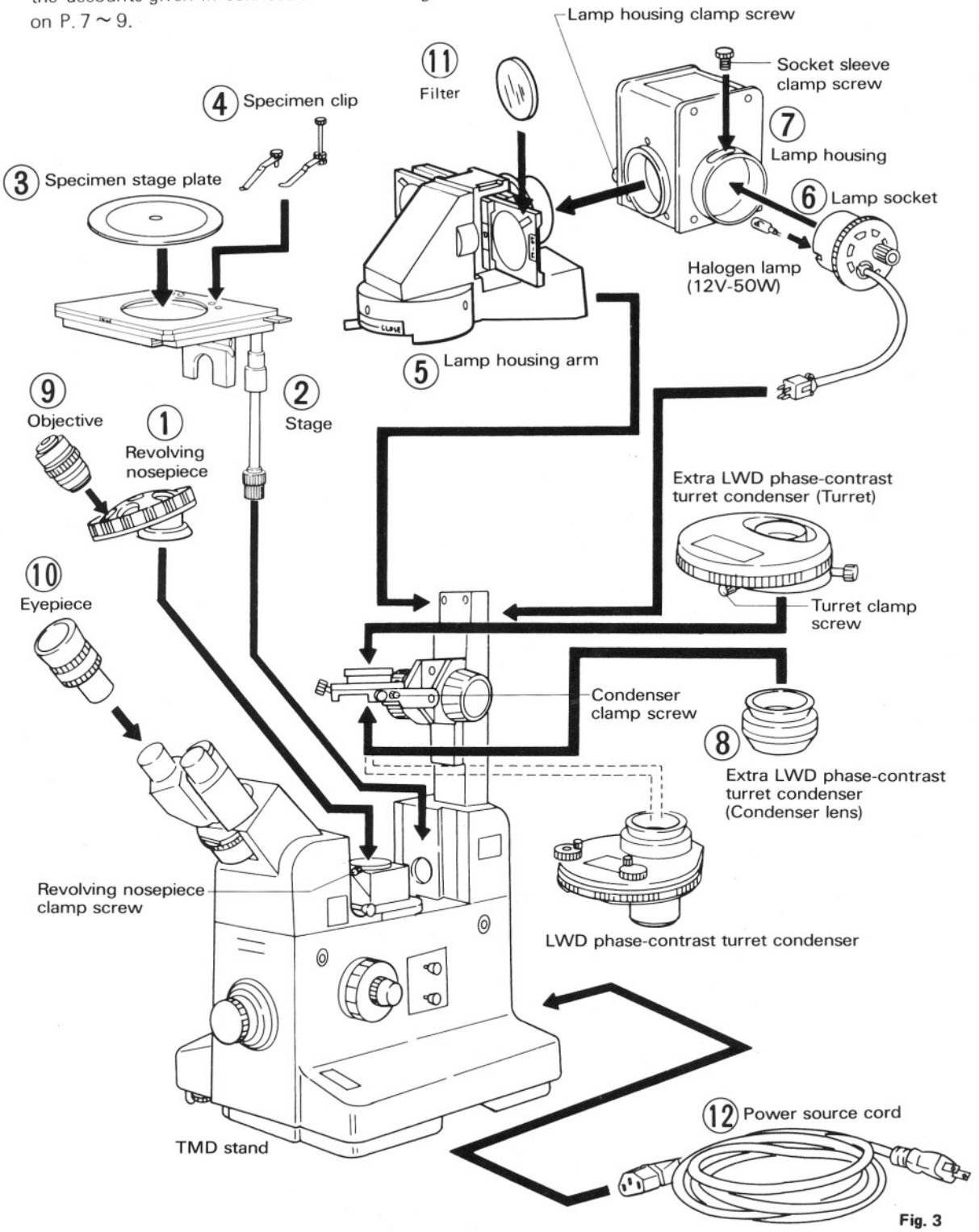


Fig. 3

① Revolving nosepiece

To attach the nosepiece, fitting its attaching groove to the positioning pin, fasten it firmly with the clamp screw. **(Fig. 4)**

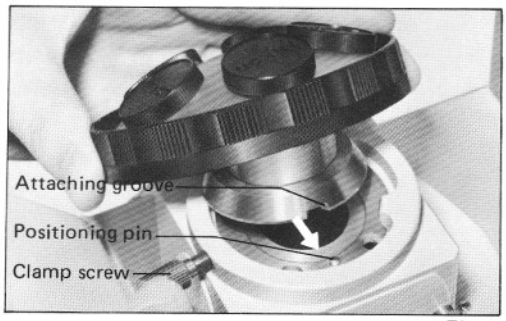


Fig. 4

② Stage

Turning the condenser focus knob, raise the condenser holder up to the highest limit, and swing out the condenser holder.

(Refer to P. 20)

Fitting the dovetail groove on the stage to the dovetail on the microscope body, slide in the stage gently to the limit.

Fasten the clamp screw firmly by means of a screw driver. **(Fig. 5)**

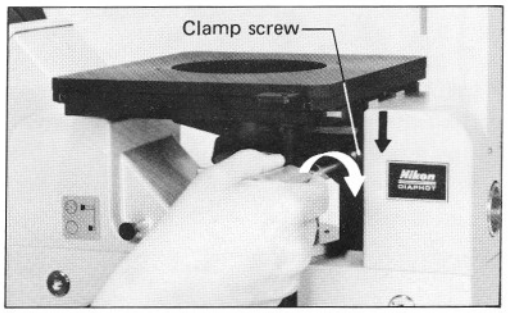


Fig. 5

③ Specimen stage plate

Place the plate into the stage at the center. **(Fig. 6)**

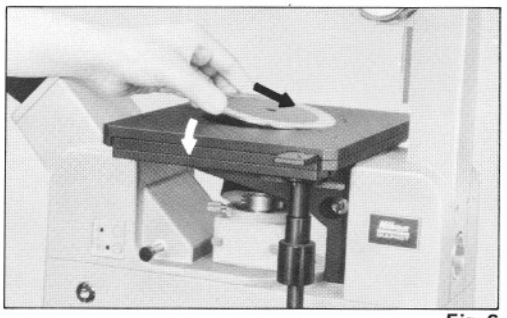


Fig. 6

④ Specimen clips

Attach the clips to the clip holes on the stage. **(Fig. 7)**

Long type clips : Used for thick objects such as a culture dish.

Short type clips : Used for regular glass slides.

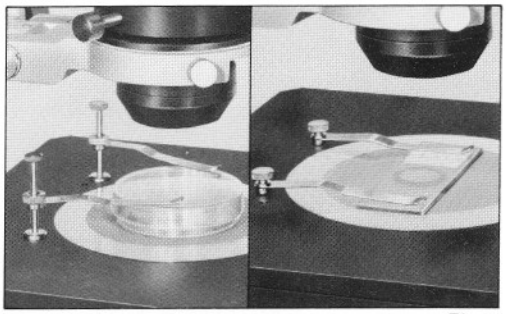


Fig. 7

⑤ Lamp housing arm

Fitting the positioning groove on the arm to the pin on the illumination post, and supporting the arm with the hand, fasten it from the rear side of the post with two hexagonal hole bolts, using a hexagonal wrench. **(Fig. 8)**

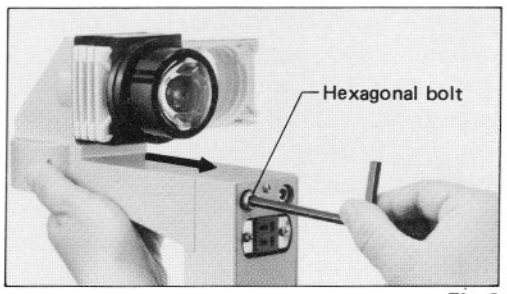


Fig. 8

⑥ Halogen lamp and socket

Insert fully the halogen lamp (12V-50W) with its pins into the holes on the socket. At this time, do not touch the glass portion with the bare hand. Use gloves or cloth.

Then put the socket into the lamp housing, and fasten it in position with the clamp screw. **(Fig. 9)**

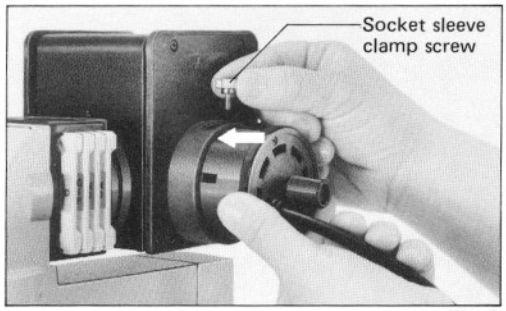


Fig. 9

⑦ Lamp housing

Once release the clamp screw on the lamp housing.

Insert the housing to the arm at the collector lens, and fasten the clamp screw.

Connect the plug for the socket to the receptacle on the rear side of the illumination post. **(Fig. 10)**

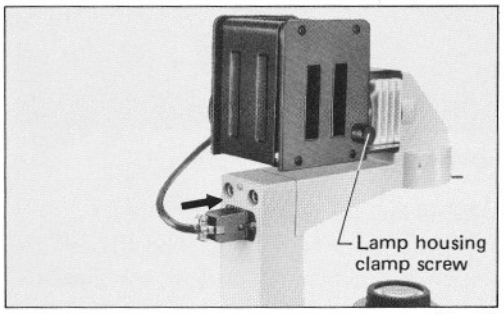


Fig. 10

⑧ Condenser

★ Extra LWD phase-contrast turret condenser

This type condenser consists of a pair of condenser lens and turret.

Attach the condenser lens with its aperture number plate faced toward the user to the bottom of the condenser mount.

Fasten the clamp screw. **(Fig. 11)**

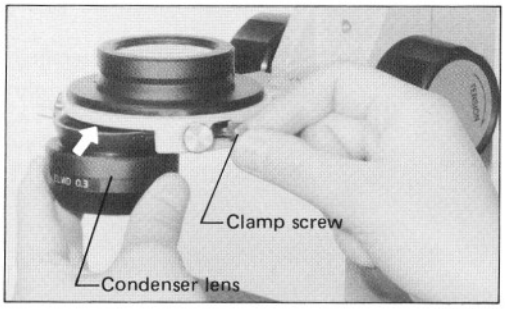


Fig. 11

For attaching the turret, first, release the clamp and two centering screws, and fitting the positioning groove on the turret to the pin on the condenser mount, push in the elastic top of the clamp screw.

Fasten the turret with the clamp screw in position. **(Fig. 12)**

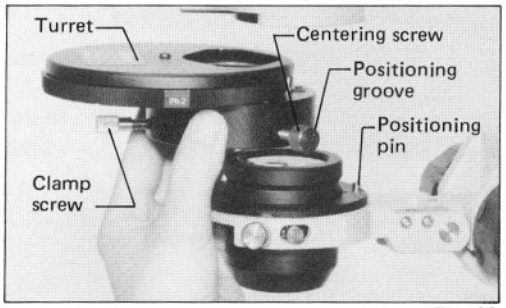


Fig. 12

★ LWD phase-contrast turret condenser

Fitting the positioning pin on the condenser to the groove in the innermost position of the condenser mount, attach the condenser, and fasten it with the clamp screw. **(Fig. 13)**

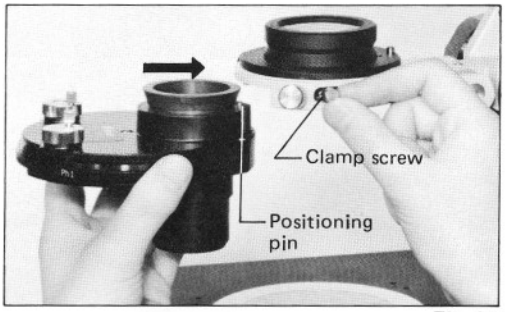


Fig. 13

⑨ Objectives

Beforehand, rotate the coarse focus knob to move the revolving nosepiece to the lowest position.

Attach the objectives to the nosepiece from the left side one after another in such positions that the magnifying power increases, when the nosepiece is revolved clockwise, viewed from above. **(Fig. 14)**

Be careful not to let the tops of objective touch with the stage, etc.

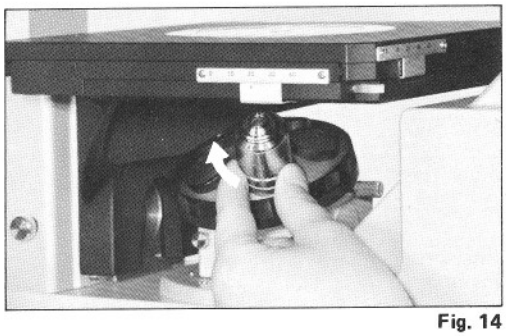


Fig. 14

⑩ Eyepiece

Insert the eyepiece into the eyepiece sleeve of the microscope. **(Fig. 15)**

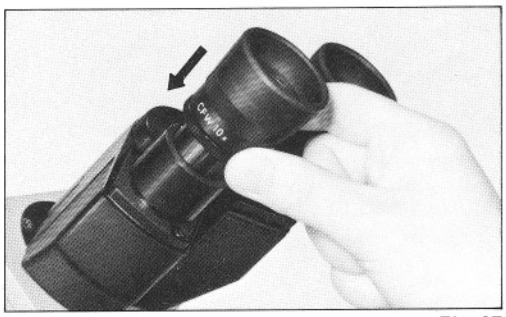


Fig. 15

⑪ Filter

Attach the diffuser with its mat surface turned toward the user **(Fig. 16)** into the receptacle ⌜D⌟ of the filter slider. Attach the desired filter to the filter slider.

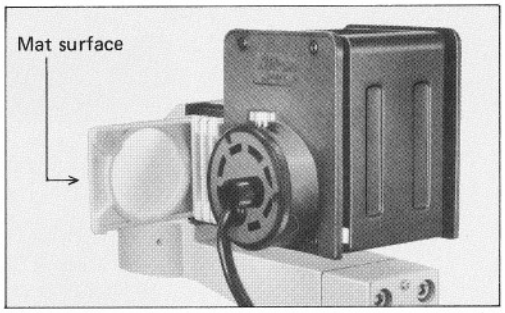


Fig. 16

※Heat absorption filter (optional)

Optional heat absorption filter can be attached as shown in the figure below:

[1] Unscrew the collector lens, [2] insert the heat absorption filter into the filter slider holder, and [3] replace the collector lens. (Fig. 17)

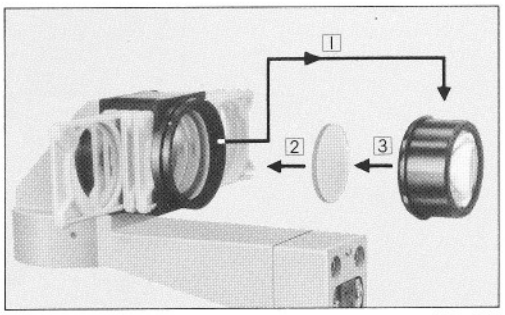


Fig. 17

⑫ Power source cord and fuse

Connect the cord firmly.

The fuse rated 2A/250V or 1A/250V is used. For replacement, remove the fuse cap by turning in the direction of the arrow. The fuse holder, embodying an input voltage changeover device, is to be set so that the power source voltage being used shows up. **(Fig. 18)**

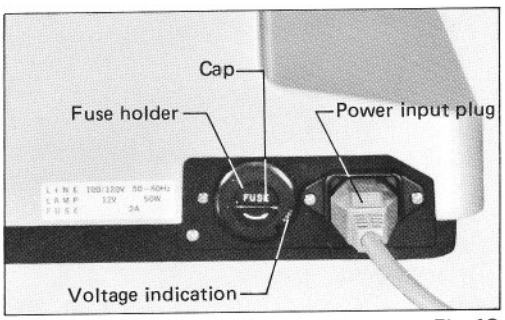


Fig. 18

III. PREPARATION

[In the following, the procedures using the Extra LWD condenser on one hand, and using the LWD condenser on the other, if not exactly the same, will be described to the left and to the right, respectively.]

1. Switching ON the power source, and placing the specimen

1) Connect the power source cord to the socket.
2) Turn ON the power switch, and set the brightness indicator to 6.
3) Place the specimen onto the stage. Fasten it in position using the specimen clips, if necessary.

2. Adjusting the interpupillary distance

1) Set the condenser turret to [PhL], and the observation turret to [O].

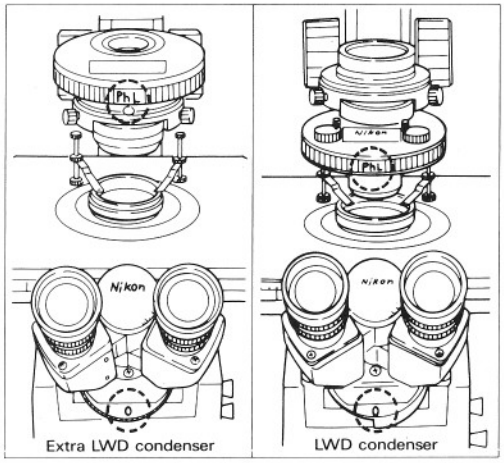


Fig. 19

2) Turn the field diaphragm control lever to [OPEN] to fully open the diaphragm. **(Refer to Fig. 23)**
3) Bring the specimen image into focus, using the 4× objective.
   Adjust the interpupillary distance, as shown in **Fig. 20**, so that the right-and left eye view-fields come together into coincidence.

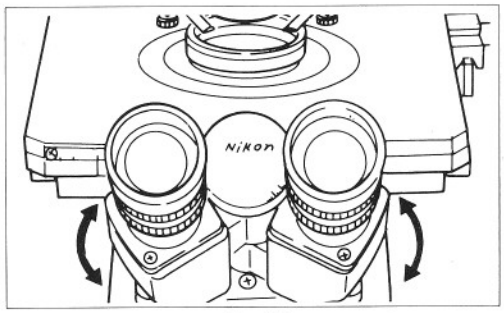


Fig. 20

3. Diopter adjustment

1) Pull the photo mask sliding knob up to the limit to bring the photo mask into the optical path.
2) Turning the diopter ring on each eyepiece, until the crossline image appears sharp. Do this adjustment for right- and lefthand eyepieces. (Fig. 21)

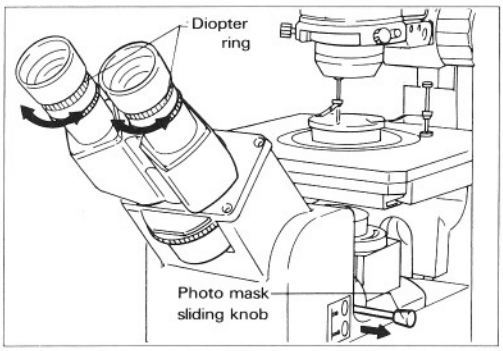


Fig. 21

※ The CF eyepieces being of high eyepoint type, when the observer uses his eyeglasses, it will not be necessary to remove but only to bend the rubber eyeguards. (Fig. 22)

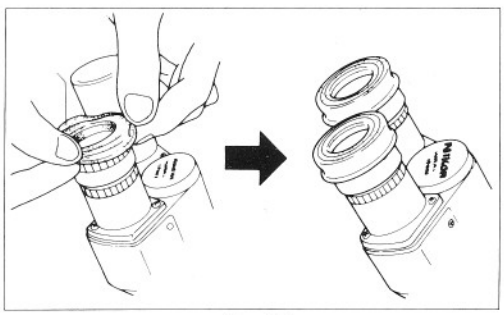


Fig. 22

Extra LWD condenser

LWD condenser

4. Centering the condenser

1) Setting the condenser turret to 〔PhL〕, take out the annular diaphragm from the turret. (Fig. 23)
2) Set the observation turret to 〔O〕.
3) Fully open the viewfield diaphragm. (Fig. 23)

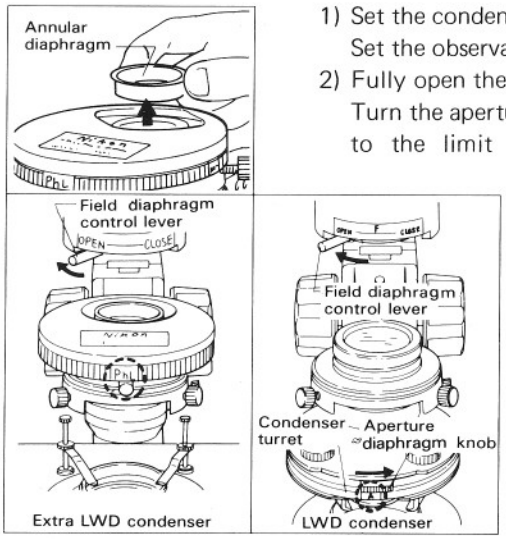


Fig. 23

1) Set the condenser turret to 〔A〕. Set the observation turret to 〔O〕.
2) Fully open the viewfield diaphragm. Turn the aperture diaphragm knob clockwise to the limit to fully open the aperture. (Fig. 23)

4) Bring the specimen image into focus, using the 4× objective.
5) Closing the viewfield to a small area, bring the image of the circumference into sharp focus by means of the condenser focus knob.

3) Bring the specimen image into focus, using the 4× objective.
4) Closing the viewfield to a small area, bring the image of the circumference into sharp focus by means of the condenser focus knob.

6) If the viewfield diaphragm image is found decentered in relation to the eyepiece viewfield, adjust it by means of the condenser centering screws. (Fig. 24 and 25 – 1)
7) Using the 4× objective, adjust the size of viewfield diaphragm so that the viewfield image coincidence with the eyepiece field, as shown in Fig. 25– 2 . Then, if they are found decentered from each other, do centering by means of the condenser centering screws.
8) Reattach the annular diapharagm, once removed, to the condenser turret.

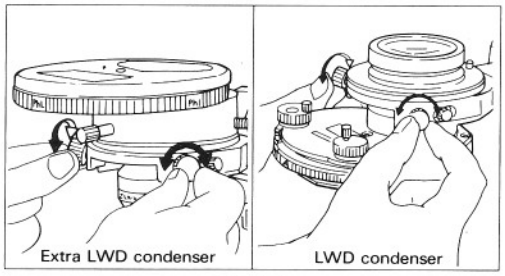


Fig. 24

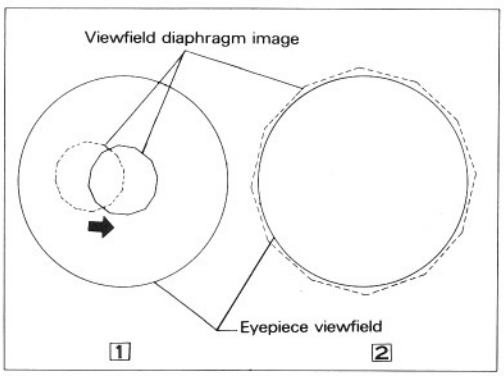


Fig. 25

5) If the viewfield diaphragm image is found decentered in relation to the eyepiece viewfield, adjust it by means of the condenser centering screws. (Fig. 24 and 25– 1)

6) Using the 4× objective, adjust the size of viewfield diaphragm so that the viewfield image coincidence with the eyepiece field, as shown in Fig. 25– 2 . Then, if they are found decentered from each other, do centering by means of the condenser centering screws.

Extra LWD condenser

LWD condenser

5. Centering the lamp

1) Set the condenser turret to [PhL], and remove the annular diaphragm from the turret.
2) Set the observation turret to [O].
3) Fully open the viewfield diaphragm by means of its control lever.
4) **Using the 10× objective,** bring the specimen image into focus.
5) **Change over the observation turret to [B].** Turn the Bertrand lens focus lever to bring the phase-contrast ring inside the objective into focus. **(Fig. 26)**
6) Slide out the diffuser from the optical path. Releasing the lamp housing clamp screw, move the housing back and forth to have the lamp filament image focused on the phase-contrast ring. **(Fig. 27)**
7) After releasing the socket sleeve clamp screw, as shown in **Fig. 28,** manipulate the lateral centering screw and vertical centering ring to bring the filament image to the center, as shown in **Fig. 29.**
8) After finishing the above lamp centering procedure, slide back the diffuser into the optical path.
9) Reattach the annular diaphragm, once removed, to the condenser turret.

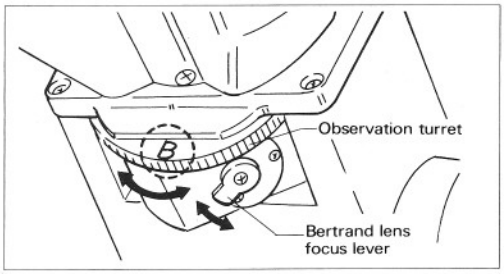


Fig. 26

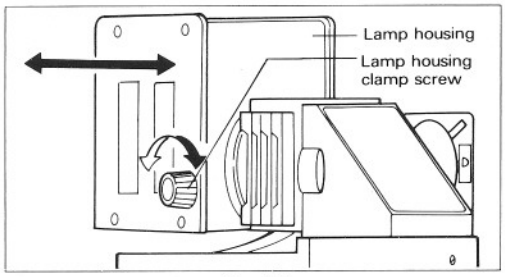


Fig. 27

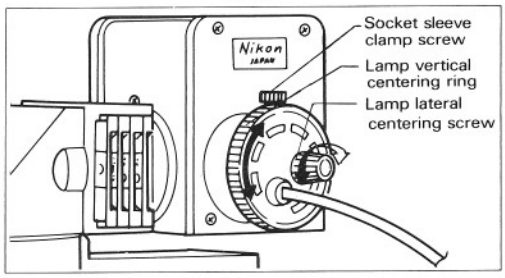


Fig. 28

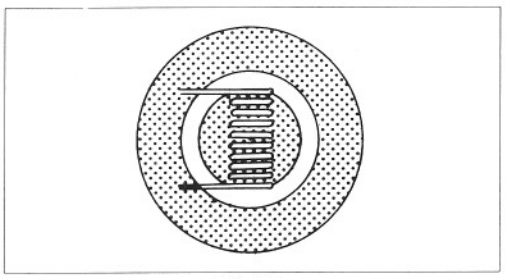


Fig. 29

1) Set the condenser turret to [A]. Set the observation turret to [O].
2) Fully open the viewfield and the aperture diaphragms.
3) **Using the 20× objective,** bring the specimen image into focus.
4) **Change over the observation turret to [B].** Turn the Bertrand lens focus lever to bring the phase-contrast ring inside the objective into focus. **(Fig. 26)**
5) Slide out the diffuser from the optical path. Releasing the lamp housing clamp screw, move the housing back and forth to have the lamp filament image focused on the phase-contrast ring. **(Fig. 27)**
6) After releasing the socket sleeve clamp screw, as shown in **Fig. 28,** manipulate the lateral centering screw and vertical centering ring to bring the filament image to the center, as shown in **Fig. 29.**
7) After finishing the above lamp centering procedure, slide back the diffuser into the optical path.

Extra LWD condenser

LWD condenser

6. Centering the phase-contrast annular diaphragm

1) Set the condenser turret to 〔PhL〕, and the observation turret to 〔O〕.
2) Fully open the viewfield diaphragm.
3) Revolve the 4× objective into the optical path, and bring the specimen image into focus.
4) Change over the observation turret to 〔B〕. Turning the Bertrand lens focus lever, bring the phase-contrast ring inside the objective into focus.

1) Set the condenser turret to 〔PhL〕, and the observation turret to 〔O〕.
2) Fully open the viewfield diaphragm.
3) Revolve the 4× objective into the optical path, and bring the specimen image into focus.
4) Change over the observation turret to 〔B〕. Turning the Bertrand lens focus lever, bring the phase-contrast ring inside the objective into focus.

5) At this time, if the phase-contrast ring in the objective is seen not exactly overlapped on the condenser annular diaphragm image, manipulate the annular diaphragm centering screws, after releasing the clamp screw, as shown in Fig. 30.
After finishing the above centering procedure, refasten the clamp screw. Since any displacement from each other, as shown in Fig. 31, may cause low contrast to the image, it is necessary to bring the annular and ring images into exact coincidence.

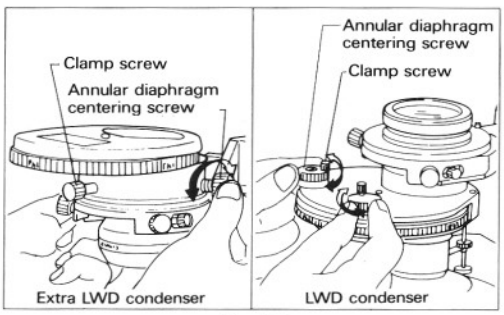


Fig. 30

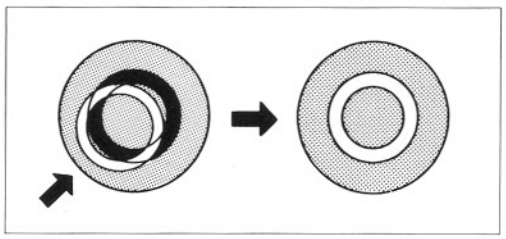


Fig. 31

Note:

If once the centering of the (PhL) annular diaphragm has been accomplished, in general no more centering will be required for other annular diaphragms.
The degree of coincidence, however, having a critical effect on the phase-contrast image, it is recommended for precise observation and for photomicrography to make sure of exact superimposing at every time the magnification is changed over.

5) At this time, if the phase-contrast ring in the objective is seen not exactly overlapped on the condenser annular diaphragm image, manipulate the annular diaphragm centering screws, after releasing the clamp screw, as shown in Fig. 30.
After finishing the above centering procedure, refasten the clamp screw. Since any displacement from each other, as shown in Fig. 31, may cause low contrast to the image, it is necessary to bring the annular and ring images into exact coincidence.

IV. MICROSCOPY

1. Phase-contrast microscopy procedure

1	Turn on the power switch. Light the lamp, and set the brightness indicator to 6.
2	Place the GIF (green interference) filter or NCB10 filter into the optical path.
3	Place the specimen on the stage.
4	Push in the upper knob of the optical path change-over to the limit.
5	Make adjustment of interpupillary distance and diopter. **(Refer to P.10 and 11)**
6	Perform centering of the condenser. **(Refer to P.12)**
7	Perform centering of the lamp. **(Refer to P.13)**
8	Perform centering of the annular diaphragm. **(Refer to P.14)**
9	Revolve the objective (PhL, Ph1, Ph2, Ph3 or Ph4) into the optical path.
10	Set the condenser turret to the same Ph number as that of the objective being used. If precise observation is required, make sure of exact coincidence of the annular diaphragm with the phase-contrast ring, at every time the magnification is changed over.
11	Set the observation turret to [0].
12	Bring the specimen image into focus.
13	Adjust the brightness by selecting the ND filter of lamp voltage (on the brightness indicator 6 ~ 12).
14	Adjust the viewfield diaphragm so that the circumference of the viewfield circumscribes that of the eyepiece viewfield.

2. Brightfield microscopy procedure

★ **Using Extra LWD condenser**

1~7	Refer to the procedure for phase-contrast microscopy 1 ~ 7 P.15.
8	Set the condenser turret to [PhL], and take out the annular diaphragm from the turret. When using the extra LWD condenser, attach an aperture diaphragm (optional) to the extra LWD condenser.
9	Set the observation turret to [O].
10	Change over the objective※ to that to be used.
11	Bring the specimen image into focus.
12	Adjust the brightness by selecting the ND filter or voltage of the lamp (on the brigtness indicator 6 ~ 12).
13	Adjust the opening of the viewfield and aperture diaphragms.

★ **Using LWD condenser**

1~7	Refer to the procedure for phase-contrast microscopy 1 ~ 7 P.15.
8	Set the condenser turret to [A].
9	Set the observation turret to [O].
10	Change over the objective※ to that to be used.
11	Bring the specimen image into focus.
12	Adjust the brightness by selecting the ND filter or voltage of the lamp (on the brightness indicator 6 ~ 12).
13	Adjust the opening of the viewfield and aperture diaphragms.

※ For brightfield microscopy in general the phase-contrast objectives can be used. For critical observation, however, it is recommended to use the brightfield objectives.

3. Manipulation of each part

1) Focusing device

- The arrows in Fig. 32 show the relation between the direction of rotation of the focus knobs and that of vertical movement of the objective nosepiece.

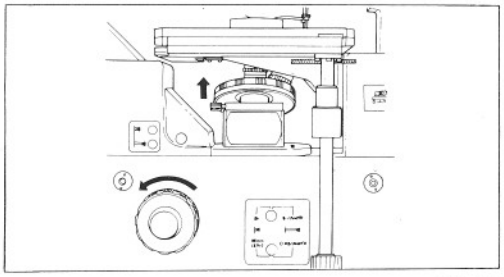


Fig. 32

- One rotation of the fine focus knob moves the objective 0.1mm vertically, the minimum reading of the scale on the knob being 1 μm.
  One rotation of the coarse focus knob moves the objective 4.7mm.
- Tension of the coarse focus knob tightens by turning the torque adjustment ring counterclockwise.
- Never attempt to turn the one knob while holding the other, because of causing disorder.

2) Observation turret

- By turning the turret, the markings [O], [B], [C] and [M] will show up one after another. (Fig. 33)

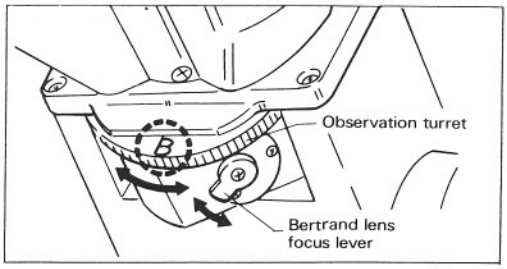


Fig. 33

- When the turret is set to [O], the optical path will be opened.
- When it is set to [B], the Bertrand lens will be put into the optical path, enabling the user to observe the exit pupil of the objective, thus permitting centering the phase-contrast annular diaphragm and the lamp.
  For focusing the Bertrand lens, turn its focus lever, as shown in Fig. 33.
- When the turret is set to [C], the light blocking plate will be inserted into the optical path, which prevents extraneous light from entering the eyepiece, thus being utilized for photography.
- When it is set to [M], the magnifier lens will be put into the optical path to multiply the magnification of eyepiece by 4×.
  This is used for focusing in photomicrography with 4× or 10× objective.

3) Condenser turret

- The condenser turret incorporates the phase-contrast annular diaphragms [PhL], [Ph1], [Ph2], [Ph3] and [Ph4] (except for Extra LWD condenser), each to be used in combination with the objective of the same Ph number.
  Therefore, according to the magnification, they are to be changed over. (Fig. 34)

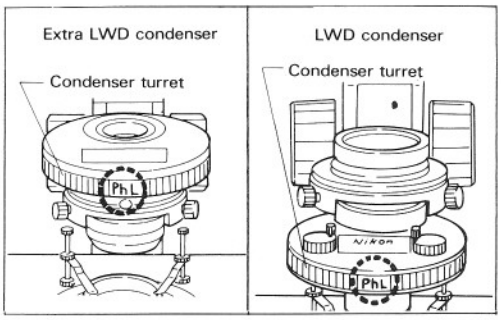


Fig. 34

- The condenser turret of the LWD condenser is equipped with an aperture diaphragm in addition, which, when the turret is set to [A], will be inserted into the optical path.
  Use this facility for brightfield microscopy.
  (Refer to 9) P. 19)
- The annular diapharagm [PhL] on the extra LWD condenser, used in combination with the 4× objective, can be taken away from the turret. (Refer to Fig. 23)
  An optional aperture diaphragm for extra LWD condenser can be attached in place of the annular diaphragm (PhL). (Fig. 35) In this case, condenser turret becomes unrotatable.

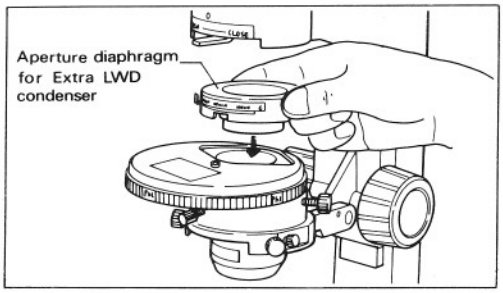


Fig. 35

4) Optical path change-over knob

- For changing over the optical path, push or pull the change-over knob. The distribution of brightness between the observation and photography systems, depending upon the position of the knob is given below:

Table 1

Brightness ratio	For observation	100%	100%	20%	20%
	For photography	0	0	Front camera port 80%	Side camera port 80%

5) Photo mask

- To put the photo mask into the optical path pull the photo mask knob up to the limit. (Fig. 36) It is used in photography, when focusing is to be done with the binocular eyepiece tube of microscope (Refer to P.24), or when diopter adjustment of the eyepiece is to be made.

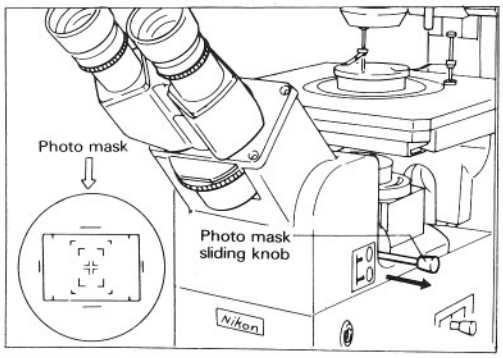


Fig. 36

6) Objectives

- For the DIAPHOT-TMD microscope, in all cases use the CF objectives in combination with the CF eyepieces, both of which have been designed on the basis of our CF (Chromatic Aberration Free) system.
- The observation of specimens under the inverted-type microscope is generally made through the bottom glass (or plastic) of a flat culture stender.
  Since, however, the thickness of such bottom glass (or plastic) differs in all cases from that (about 0.17mm) of the normal coverglass, when using objectives with large numerical aperture (N.A.) (with high magnification) such as CF DL 20X (N.A.0.4) and CF LWD DL 40X (N.A.0.55), the sharpness (resolution) and contrast of the microscope will be affected perceptibly, thus allowing us to take no full advantage of such objectives.
  Therefore, so as not to lower the image quality, such objectives are produced with a correction ring, the rotation of which enables us to use a bottom glass (or plastic) ranging 0～2mm in thickness.
  The use of the correction ring is to proceed as follows:

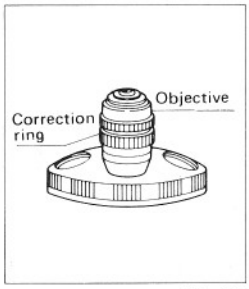


Fig. 37

① Beforehand, measure or estimate the thickness of the glass (or plastic).
Set the correction ring to that value.

② By means of the focus knob on the microscope, bring the specimen image into focus.

③ If no higher resolution and contrast are obtained (only dimmed image), slightly turn the correction ring left by right. The image, thus out-of-focused, is to be refocused by means of the fine focus knob.

④ Now, if the image quality is found better, turn the correction ring slightly further in the same direction. Repeat this procedure, until the best image, corresponding to the thickness, is attained.

If, on the contrary, by the above turning, the quality of image is inferior, turn the correction ring in the opposite direction about two times as far as the previous, and see the image quality. If this is found better, turn the ring slightly further.
Repeat the same procedure to find out the best image.

Take note of the reading at the best position on the scale, for future use, when applying almost the same thickness of glass (or plastic).
The 0-position on the scale is used, when a specimen with no coverglass is to be observed under the ordinary erecting-type microscope.
For general specimens under a standard coverglass of the thickness about 0.17mm, set the correction ring to 0.17.

7) Eyepieces

- **The CF eyepieces will produce the highest quality of image, when used in combination with the CF objectives.**
- The eyepieces are provided with a diopter ring and rubber eyeguard.
- The CF Photo eyepiece lens and CF PL Projection lens are used exclusively for photography and cannot be used for observation.

8) Viewfield diaphragm

- The diaphragm, permitting the user to limit the illuminated area to such an extent as to be observed, is generally closed so that **its circumference circumscribes that of the eyepiece viewfield.**
- To change the opening of the viewfield diaphragm manipulate its control lever. In the position [OPEN] it will be fully opened, and in the position [CLOSE], closed to the smallest opening. **(Fig. 38)**

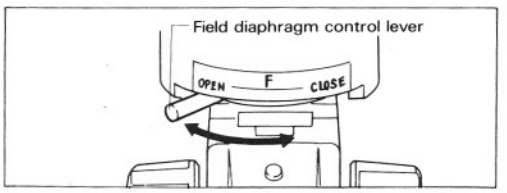


Fig. 38

9) Condenser aperture diaphragm

- For the extra LWD condenser, an aperture diaphragm is optionally available. **(Refer to Fig. 35)**
  For the LWD condenser, turn the condenser turret to the position ⌜A⌟ for the bright-field observation to bring the aperture diaphragm into the optical path.
- The diaphragm, provided for adjusting the numerical aperture of the illumination system, will generally offer a proper contrast to the image, when **closed to 70% ~ 80% of the numerical aperture of the objective being used,** by means of the diaphragm lever or knob. **(Fig. 39 and 40)**

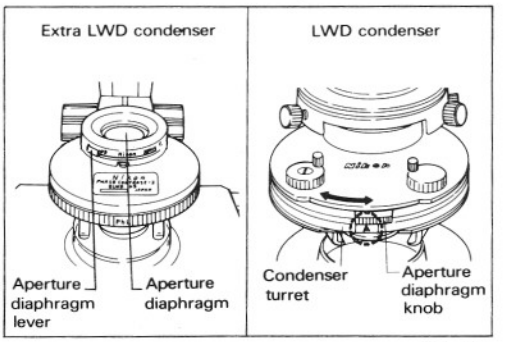


Fig. 39

- The closed diaphragm can be seen on the exit pupil inside the objective with the observation turret set to [B].

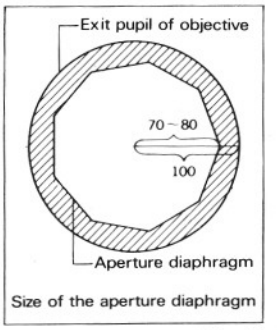


Fig. 40

10) Filters

- Filters are listed in Table 2. Attach each filter into the corresponding slot of the filter slider on the lamp housing arm according to the indication.

Table 2

Type of filter	Use
Diffuser	To be inserted in all cases except for lamp centering
NCB10 color balancing filter	For general purposes and color photography
ND 2 (T=50%) ND16 (T=6.25%)	For adjusting brightness
GIF (green interference)	For monochromatic observation and photography
Heat absorbing filter	For protecting specimens from heating effect of illuminating light

11) Swing-out device of condenser

- Turn the condenser mount upward to swing out the condenser.
  This device facilitates replacement of large specimens, culture dishes, etc. **(Fig. 41)**

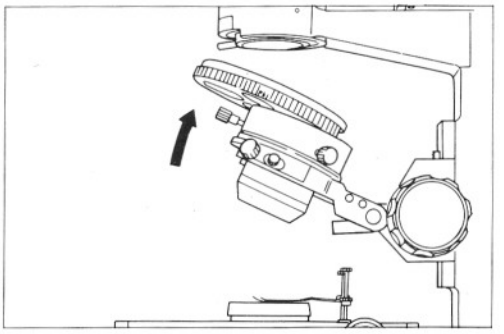


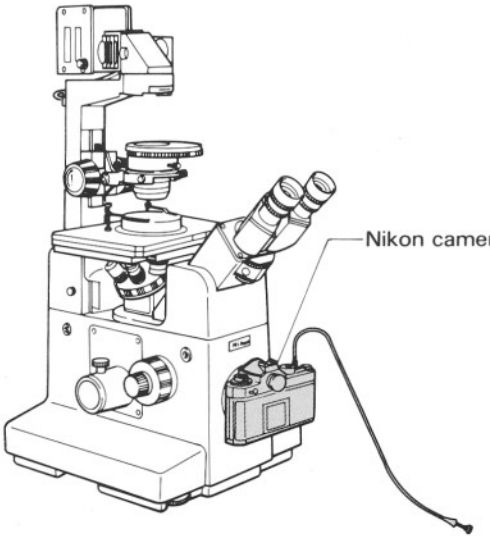
Fig. 41

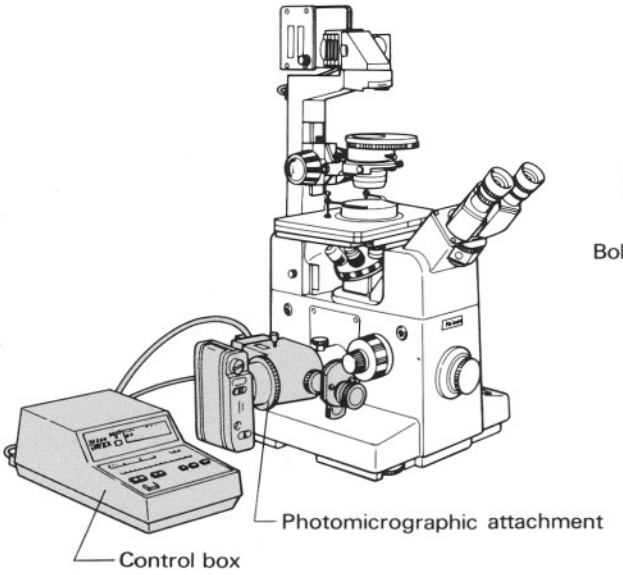
- During observation, swing back the condenser mount to the limit.

V. PHOTOMICROGRAPHY

The microscope DIAPHOT-TMD has two camera attaching ports, one on the front of the microscope stand for accepting directly a 35mm Nikon camera, permitting the use of the exposure and shutter operation of the camera, and the other on the side of the microscope stand for connecting the photomicrographic attachment such as Microflex FX-series, FM-series or cinemicrographic attachment.

1. Nomenclature





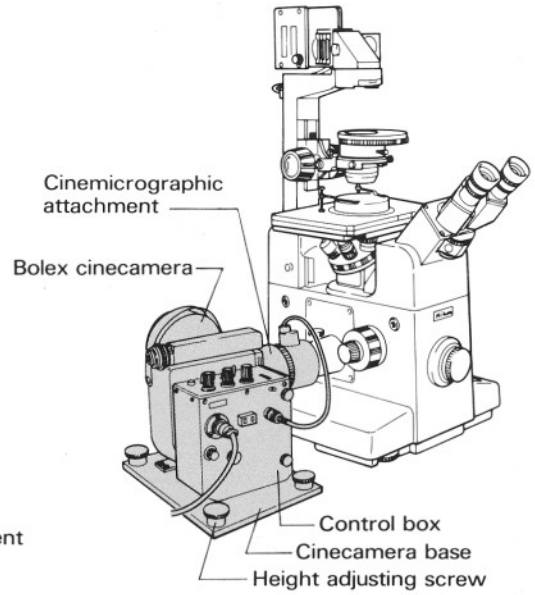


Fig. 42

2. Assembling

1) Attaching directly a 35mm camera to the front camera port

Remove the cap from the front camera port on the TMD microscope.
Lining up the mounting index on the camera to that on the microscope, turn the camera body in the direction of the arrow **(Fig. 43)**, until it click-stops in position.
To detach the camera, depressing the lens mount button, turn the camera body in the opposite direction of the arrow, until it separates from the port.

Caution : **For detaching the camera, never turn it in the direction of the arrow for the possibility of impairing the F-mount.**

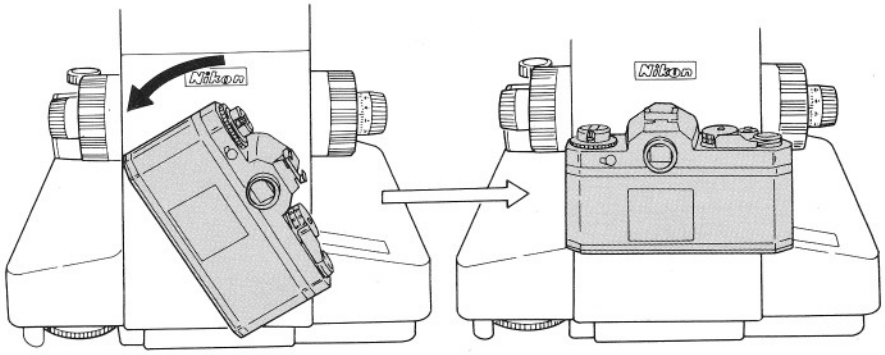


Fig. 43

2) Connecting the photomicrographic or cinemicrographic attachment to the side camera port

① CF PL Projection lens or CF Photo eyepiece

Remove the cap from the sleeve of the side camera port on the TMD microscope.

When using the photomicrographic attachment Microflex FX-series or cinemicrographic attachment CFX, attach the CF PL Projection lens by pushing into the sleeve until it comes into contact with the thrusting surface. (When using the Microflex FM-series or cinemicrographic attachment CFMA, attach the CF Photo eyepiece.)

Caution : Use the CF Photo eyepiece with the magnification and other indications engraved in yellow or with a dot • in white behind the magnification marking.

② Photomicrographic or cinemicrographic attachment

- Photomicrographic attachment

Connect the Microflex main body to the microscope, by pushing the connecting ring ※ into the sleeve to the limit where it touches the thrusting surface, in such a position that the finder attaching part faces toward the user. Fasten the unit by means of the clamp screw.

- Cinemicrographic attachment

Place a cinecamera (Bolex H 16 as standard) on the cinecamera base, and fasten it in position by means of the tripod screw provided on the base. Manipulating the height adjusting screws (×4) on the base, push positively the connecting ring ※ on the attachment main body into the attaching sleeve, and fasten the ring firmly in position by means of the clamp screw.

※ **For connecting the Microflex FX-series, Microflex HFX or cinemicrographic attachment CFX, use the regular connecting ring, but for connecting the Microflex FM-series except HFM or CFMA, the regular connecting ring should be replaced with the 30mm long connecting ring.**

③ Finder cap

Attach the cap, supplied with each photomicrographic or cinemicrographic attachment main body, onto the finder accepting part to prevent extraneous light from entering the attachment main body.

When using the attachment with the finder attached, place the finder cap over the finder.

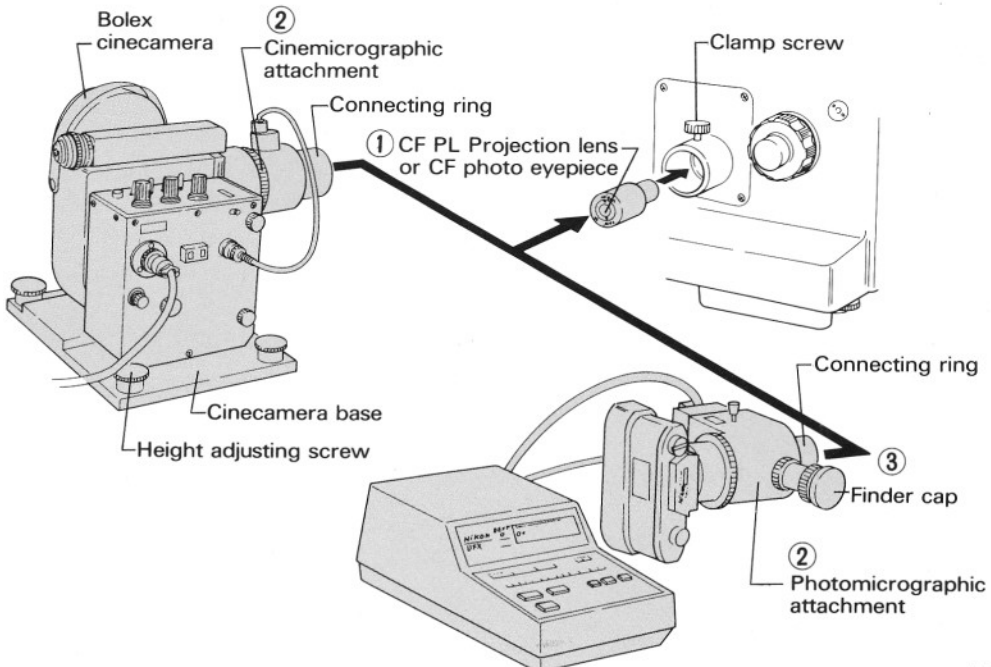


Fig. 44

3. Operating procedure of photography

1) Illumination

(1) Checking the illumination

Unevenness in the illumination will show up more conspicuously in photography than in observation. Consequently, re-check the positioning and centering of the lamp, and for the correct adjustment of the condenser and phase-contrast annuli, beforehand.

(2) Selection of voltage and filter

Since the color temperature of the light source varies with the voltage being used, the selection of voltage and filter is essential in color photography.

Standard combinations of voltage and filter will be given below:

Standard Combination of Voltage and Filter **Table 3**

Film		Voltage	Filter
Color	Daylight type	9	Use NCB10
	Tungsten type	8	Remove NCB10
Monochromatic		6 or higher	Remove NCB10 Contrast filter such as green is usable

The above table gives only the standard combinations.

Depending upon the make of the film, different color renditions may result. In some cases the additional use of a proper color compensation filter (CC filter) may be necessary.

2) Viewfield and aperture diaphragm

The viewfield diaphragm serves to limit excessive light which may produce flare. It should be closed down to an area slightly larger than the picture area.

On the other hand, the condenser aperture diaphragm enables changing the depth of focus, image contrast, resolution of image, etc. in brightfield photography.

Select a proper opening according to the photographic effect desired.

It is a general rule to close the diaphragm to 70% ~ 80% of the numerical aperture of the objective being used.

3) Focusing

Irrespective of the type of the camera attached directly to the front port or the type of the photographic attachment connected to the side port on the microscope, focusing is conducted with the binocular eyepiece tube in the following way:

(1) Holding the photo mask knob, pull it up to the limit to bring the photo mask into the optical path.

(2) Make sure of the diopter adjustment.

(Refer to P.11)

(3) Set the optical path change-over knob.

- **For the 35mm camera on the front port**
  Pull up only the upper knob of optical path change-over to the limit, and push in the lower one to the limit.

- **For the photomicrographic or cinemicrographic attachment on the side port**
  Pull both the upper and lower knobs of the optical path change-over.

(4) Manipulating the coarse and fine focus knobs, bring the specimen image into focus.

Note: For focusing with 4× or 10× objective, set the observation turret to [M] to put the magnifier lens into the optical path, whereby a magnifying device of higher magnification is built, permitting more accurate focusing.

For diopter adjustment of eyepiece, set the observation turret to [O] of empty hole, not to the [M] position.

4) Photo mask and picture area

- **For the 35mm camera on the front port**
  The A-frame determines the picture area as shown in **Fig. 45.**
  The magnification obtained on the film plane will be 2.5 times higher than the power of the objective.

- **For the photomicrographic or cinemicrographic attachment on the side port**
  The A ~ E frames show the largest picture areas obtained on the different film formats, corresponding to the area at the lowest magnifications through the CF PL Projection lens or CF Photo eyepiece.

(Refer to the Table 4.)

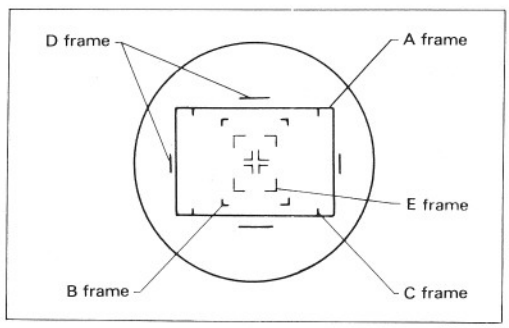


Fig. 45

Table 4

Photomicrographic or cinemicrographic attachment	Type of film	Magnification of CF Photo eyepiece	Magnification of photographic lens	Picture area
HFM AFM EFM PFM	35mm	5X	½X	Frame A
	4"X5"	8X	1.25X	Frame C
	3¼"X4¼" 6X9	5X	1.25X	Frame D
CFMA	16mm	5X	¼X	Frame B
		10X	¼X	Frame E
		Magnification of CF PL Projection lens	Large format adapter (4X)	
UFX HFX AFX PFX	35mm	2.5X	–	Frame A
	4"X5"	2.5X	4X	Frame C
		Magnification of CF PL projection lens	Magnification of photographic lens	
CFX	16mm	2.5X	½X	Frame B
		5X	½X	Frame E

In other cases than indicated in the above table and where the picture area should be determined more precisely, it will be necessary to use the finder system on the photomicrographic or cinemicrographic attachment.

★ Cautions in Photography

- **For the 35mm camera on the front port**

① The exposure and shutter operating mechanism in the camera being used in this case, set the ISO/ASA dial to the film speed to be used, and the speed dial to AUTO.

② Make sure that only the upper knob of optical path change-over is pulled out, and the lower one is pushed in.

③ It is recommended to adjust the brightness by the ND filters to such an extent that a shutter speed lower than 1/8 sec. can be used.
For monochromatic films, however, the adjustment of voltage is possible.

④ Except for making sure of the shutter speed, be careful not to enter stray light into the finder.

⑤ In such a case as requiring a long exposure time, where the eye is kept apart from the eyepiece lens, while the shutter operates, set the observation turret to [C] to prevent extraneous light from entering the eyepiece.

- **For the photomicrographic or cinemicrographic attachment on the side port**

① Make sure that both the upper and lower knobs of optical path change-over are pulled out.

② In such a case as requiring a long exposure time, where the eye is kept apart from the eyepiece lens, while the shutter operates, set the observation turret to [C] to prevent extraneous light from entering the eyepiece.

Note : For the use of the camera, photomicrographic or cinemicrographic attachment, refer to the pertinent instruction manual supplied with the respective attachments.

VI. OPTIONAL ACCESSORIES

1. Incubator

1) Nomenclature

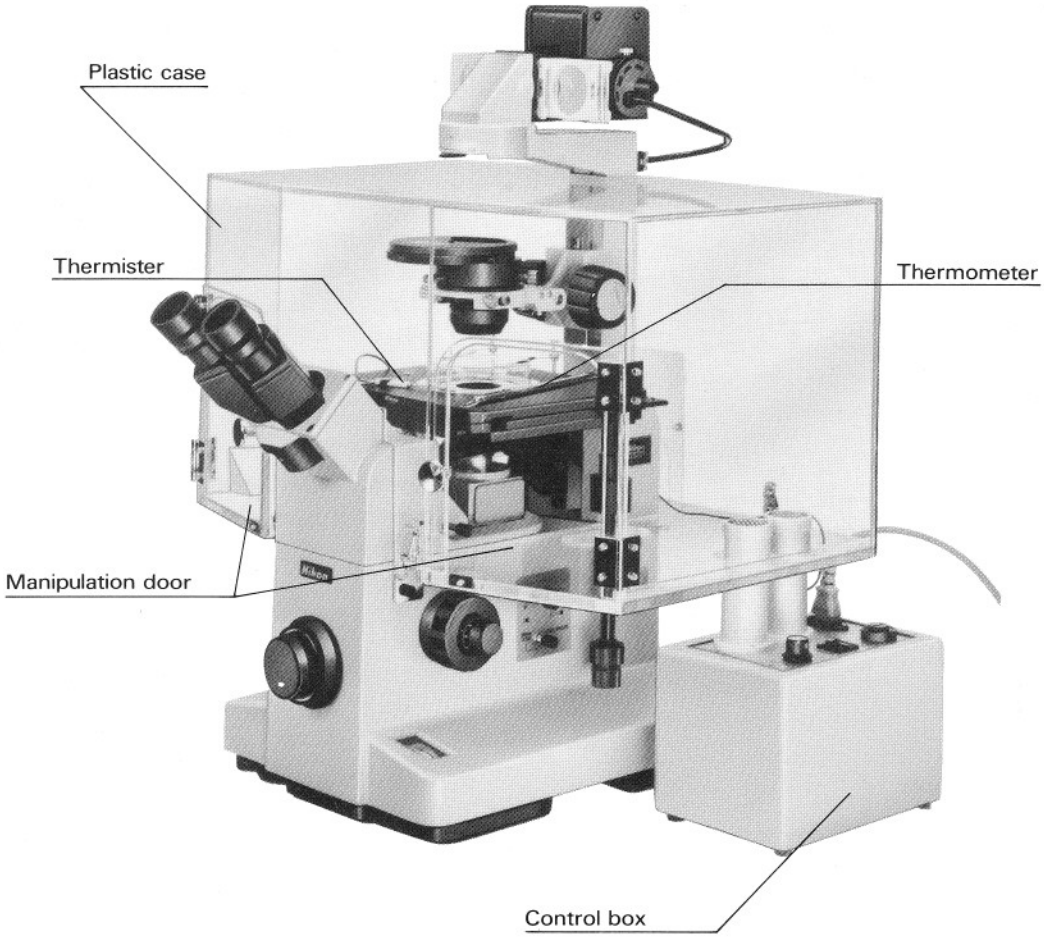


Fig. 46

2) Assembling

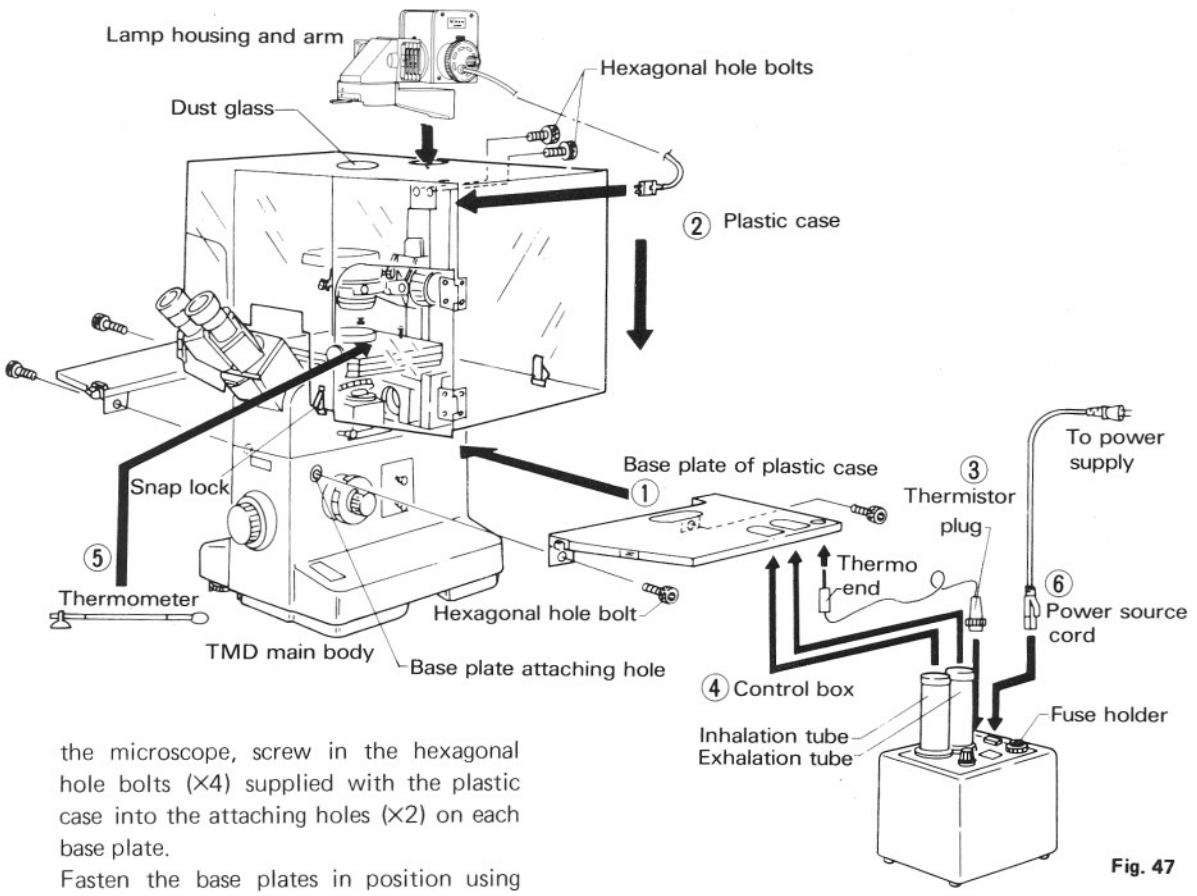


Fig. 47

the microscope, screw in the hexagonal hole bolts (×4) supplied with the plastic case into the attaching holes (×2) on each base plate.
Fasten the base plates in position using hexagonal wrench.

② Plastic case

Once remove the lamp housing arm with the housing from the microscope.
Placing the plastic case gently over the microscope, and fasten it to the base plates by means of the two snap locks at the front and rear positions on either side.
Reattach the lamp housing arm with the housing in its original position.
Place the dust glass into the plastic case top.

③ Thermistor plug

To insert the thermistor plug into the receptacle on the control box, fitting the positioning groove on the plug to the pin inside the receptacle, and the groove on the external tube of the plug to the pin on the outside of the receptacle, turn the milled part of the plug, until the plug is fixed with click. Put the thermo-end of the thermistor through the small hole at the innermost position on the right side base plate of plastic case.

④ Control box

Tilting the control box slightly, set it in such a position that its exhalation and inhalation tubes enter the holes in the base plate, as shown in Fig. 47.

⑤ Thermo-end of the thermistor and the thermometer

Open the manipulation door of plastic case. Putting the hand into the case, place the thermo-end from behind the illuminator post onto the stage at a position as nearest as possible to the specimen. It can be fixed in position by fastening the milled part of the thermo-end, using the specimen clips.
Place the thermometer at an appropriate position on the stage.

⑥ Power source cord and fuse

Connect the one plug of the power source cord to the receptacle on the control box, and the other to the power source socket. The 3A/250V or 1.5A/250V fuse is encased in the fuse holder of the control box.

To take it out for replacement, remove the cap by turning in the direction of the arrow.

The fuse holder is used also as a voltage change-over device.

If it does not show up the voltage being used, pull the holder with its cap removed and set the holder in such a position to show up the correct power source voltage, before using the instrument.

3) Temperature adjustment

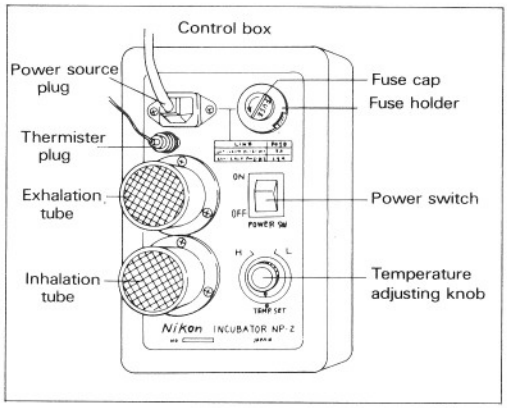


Fig. 48

The incubator starts its function, when the power switch is turned ON.

The standard incubation temperature 37° ±0.5°C at an ambient temperature of 20° ~ 30°C can be changed by setting the position of the adjusting knob.

Refer to the table below:

Table 5

Ambient temperature	Incubation temperature	Position of adjusting knob
20° ~ 30°C	37°C	On the index
	37°C or higher ※	On the [H] side
	37°C or lower ※	On the [L] side
10° ~ 20°C	37°C	On the [H] side

※ The change of temperature caused by turning the adjusting knob to a full extent on the [H] or [L] side is about 4°C.

Note : The table 5 indicates only approximate values.
Depending upon the voltage and frequency of the power source, the incubation temperature will somewhat change.
Be sure of the temperature by means of the thermometer.

4) Cautions

① The incubator controls the temperature of air surrounding the position where the thermo-end is place on the stage. Even though the manipulation door is opened, the air temperature on the stage will hardly change.
As a rule, however, do not open the door except for changing over the objective or replacing the specimen.
Temperature in the neighborhood of the base plates of plastic case will be lower than the incubation temperature on the stage.
Do not place the specimen being cultured on the base plates.

② It takes different times to raise the temperature of air on the stage up to the standard 37°C, depending upon the ambient temperature and the voltage change and frequency of the power source.
At a lower temperature or lower voltage, and when the power source frequency is 60Hz, the rise of temperature will take a longer time.
In this case, turn the temperature adjusting knob to H , and start the operation of the incubator one hour or more eariler than the observation of the specimen.

③ For quick motion cinematography, start the operation of the incubator one hour or longer before picture taking.

2. Mechanical stage

1) Nomenclature

Secure the mechanical stage to the TMD plain stage using the two mounting screws located under its right hand side.

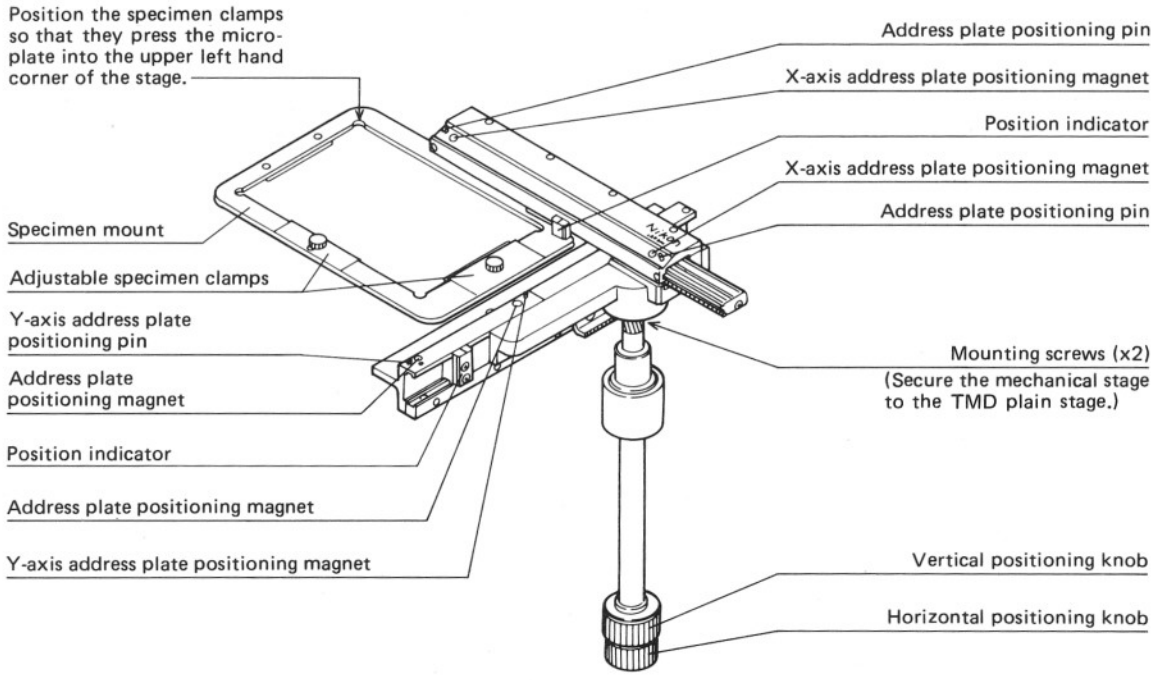


Fig. 49

< Address plates (6 sets) >

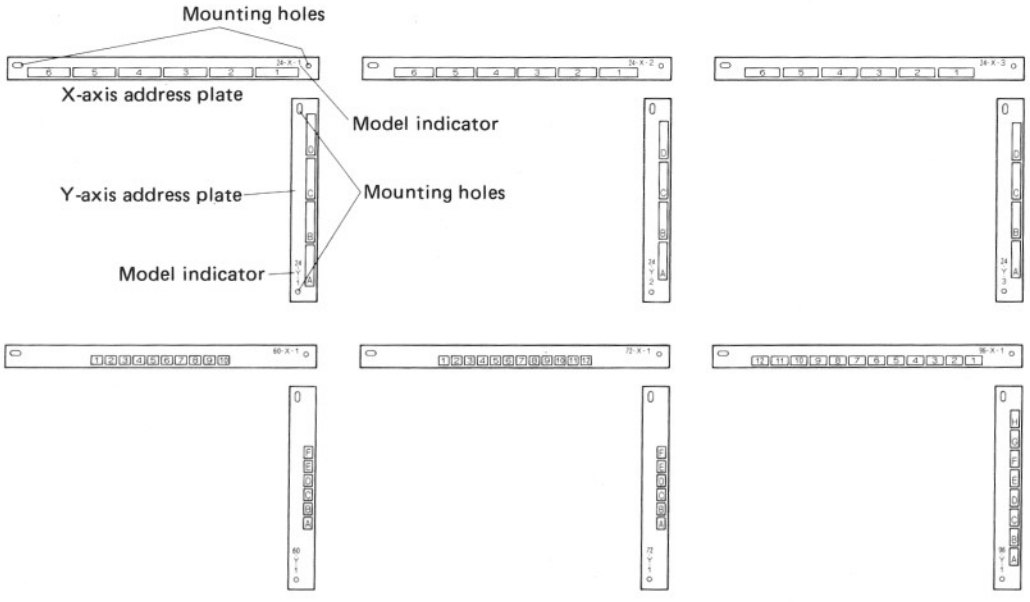


Fig. 50

2) Operation

① Specimen Mounting

Place the specimen holder or microplate in the specimen mount, using the specimen clamps to press it into the upper left hand corner of the mount.

② Specimen Positioning

X-axis (left/right) : Rotate the lower knob. (Stroke: 122mm)

Y-axis (front/back) : Rotate the upper knob. (Stoke: 84mm)

③ Address Plate Use

The optional address plates help determine specimen well positions when performing microplate observation.

- Installation

  Set the address plates in position over their respective positioning pins. The magnets will hold them in place.

- Removal

  The address plates can be easily removed by slipping a fingernail under the edge of the rule and prying it up.

- Well Number Readings

  Well positions can be determined by observing the locations of the index lines along the X (numeric) and Y (alphabetic) address plates. (Well positions correspond to the indicator blocks positioned along the length of the plates.)

Note: The blocks surrounding the alpha-numeric figures describe the perimeters of the well. **(Fig. 51)**

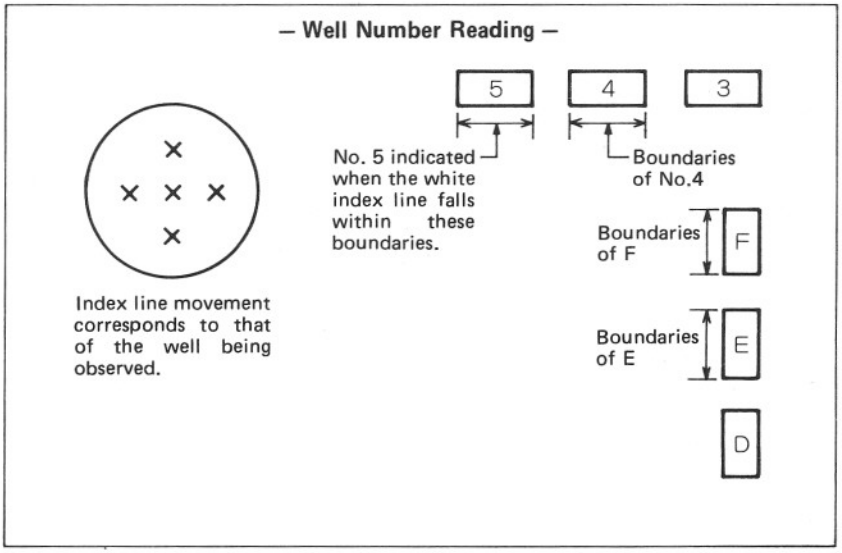


Fig. 51

3) Accesory Combinations

Table 6

Specimen			Specimen Adapter	Address Plate
	Well No.	Manufacturer		
Micro-plate	24	SUMITOMO BAKELITE		24-X-1 (TMD Type) 24-Y-1 (24 Mk. 1)
	24	CORNING		24-X-2 (TMD Type 24-Y-2 (24 Mk. 2
	24	FALCON		24-X-3 (TMD Type) 24-Y-3 (24 Mk. 3)
	60	CALCON NUNC	Microplate holder 60	60-X-1 (TMD Type) 60-Y-1 (60 Mk. 1
	72	COSTAR	Microplate holder 60	72-X-1 (TMD Type) 72-Y-1 (72 Mk. 1
	96	SUMITOMO BAKELITE, FALCON, COOKE LINBRO, NUNC		96-X-1 (TMD Type) 96-Y-1 (96 Mk. 1)
Petri dish			Petri dish holder 60	
Slide glass			Slide glass holder	

VII. TROUBLE SHOOTING TABLE

Although nowhere you can find any disorder or derangement in the instrument, if you encounter some difficulty or dissatisfaction, recheck the use, referring to the table below:

DIAPHOT-TMD

1. Optical

Failures	Causes → Actions
Darkness at the periphery or uneven brightness of viewfield (No appearance of viewfield)	• Revolving nosepiece not in clickstop position (Objective not centered in optical path) → Revolve it click-stop position • Lamp bulb not centered → Centering (Refer to P.13) • Condenser not centered → Centering by using field diaphragm (Refer to P.12) • Field diaphragm too much closed → Open it properly • Dirt or dust on the lens (Condenser, objective, eyepiece, culture dish) → Cleaning • Diffuser not set in or incorrectly positioned → Correct positioning (Refer to P.9) • Revolving nosepiece not correctly attached → Correct attaching (Refer to P.7) • Halfway position of optical path change-over knob. → Bring the knob to a stop (Refer to P.18) • Incorrect setting of condenser turret → Correct the setting position (Refer to P.17) • Incorrect setting of observation turret → Set the turret to [O] • Photo mask knob not fully changed over → Change it over to the limit • Specimen stage plate entering the optical path → Change the position and move the stage
Dirt or dust in the viewfield	• Dirt or dust on the lens (Condenser, objective, eyepiece, field lens) → Cleaning • Dirt or dust on the culture dish → Cleaning • Too low position of condenser → Correct positioning (Refer to P.12)
No good image obtained (low resolution or contrast)	• Brightfield objective used → Use phase-contrast objective • Annular diaphragm of condenser not brought into the optical path → Set the turret to Ph number of objective and put the diaphragm into the optical path (Refer to P.17) • Annular diaphragm in the condenser turret not coincided with phase-contrast ring in the objective → Make exact coincidence (Refer to P.17) • Poor centering of annular diaphragm of condenser → Correct centering (Refer to P.14) • Dirt or dust on the lens surface (condenser, objective, eyepiece or culture dish, etc.) → Cleaning • Coverglass thickness correction ring not set to the thickness of culture dish glass (or plastic) → Make correction (Refer to P.18) • Glass (or plastic) of culture dish thicker than 2mm → Use glass (plastic) not thicker than 2mm

Failures	Causes → Actions
No good image obtained (low resolution or contrast)	• Improper position of condenser → Adjust the position so that view-field diaphragm is imaged in the correct place (Refer to P.12) • Diffuser not inserted → Insert it in correct position (Refer to P.9)
Oneside dimness of image	• Revolving nosepiece not in click-stop position → Revolve it to click-stop position • Revolving nosepiece not correctly attached → Insert it to the limit and clamp it firmly • Revolving nosepiece not clamped → Clamp tightly
Image moves while being focused	• Specimen tilts from the stage surface → Correct the position of specimen on the stage • Revolving nosepiece not in click-stop position → Revolve it to click-stop position • Revolving nosepiece not clamped → Clamp tightly • Condenser not correctly centered → Correct centering (Refer to P.12) • Lamp bulb not correctly centered → Correct centering (Refer to P.13) • Condenser holder tilts → Fasten the condenser in the correct position (Refer to P.19)
Image tinged yellow	• NCB 10 filter not used → Use NCB 10 filter • Too low power source voltage → Raise the voltage over 6 on the indicator
Too bright image	• ND filter not used → Use ND filter

2. Manipulation

Failures	Causes → Actions
No focusing attained, even though the objective moved to the upper limit	• Stage not correctly attached → Correct the attaching (Refer to P.7)
No focusing attaind with 20× and 40× objective	• Glass (or plastic) of culture dish thicker than 2mm → Use glass (or plastic) not thicker than 2mm
No fusion of binocular image	• Interpupillary distance not adjusted → Adjustment (Refer to P.10)
Fatigue of observing eyes	• Incorrect diopter adjustment → Correct adjustment (Refer to P.11) • Inadequate brightness of illumination → Use ND filter or change power voltage

3. Electrical

Failures	Causes → Actions
Lamp does not light even though switched ON	• No electricity obtained → Connect the cord to socket • No lamp bulb attached → Attaching • Lamp bulb blown → Replacement • Fuse blown → Replacement • Input voltage change-over device (fuse holder) not set to house current voltage → Change over the voltage correctly • Input plug for the lamp out of place → Insert the plug into the socket on the rear side of illumination post
Unstable brightness of illumination	• Input voltage not adjusted to house current voltage → Set the indication of input voltage (at fuse holder) on the rear side of microscope base to the power source voltage • House current voltage fluctuates too much → Use transformer or the like (for adequate voltage)
Lamp bulb promptly blown	• Not specified lamp bulb used → Use 12V-50W specified lamp bulb: (Halogen bulb: OSRAM 64610 or PHILIPS 7027) • Too high voltage of house current → Use transformer for adjustment
Insufficient brightness of illumination	• Lamp bulb not centered → Centering (Refer to P.13) • Condenser not centered → Centering (Refer to P.12) • Too low position of condenser → Correct positioning (Refer to P.12) • Not specified lamp bulb used → Use 12V-50W specified Halogen bulb • Dirt on lens (condenser, objective eyepiece, field lens, filter) → Cleaning • Too low voltage → Raise the voltage
Fuse blown	• Not specified fuse used → Use 2A/250V or 1A/250V
Flickering or unstable brightness of lamp bulb	• Lamp bulb going to be blown → Replacement • Connector not connected securely → Secure connection • Fuse holder not firmly fastened → Firm fastening • Irregular change of house current voltage → Use stabilizer • Lamp bulb insufficiently inserted into the socket → Positive connection

INCUBATOR

Failures	Causes → Actions
Pilot lamp does not light even though switched ON	• No current flows → Connect power source cord to the socket • Fuse blown → Replacement • Input voltage indication does not correspond to house current → Coincide the voltage to that of house current
Temperature does not rise	• Incorrect position of temperature adjusting knob → Correct the position (Refer to P.27) • Poor connection of thermistor → Positive connection
Temperature too high	• Incorrect position of temperature adjusting knob → Correct the position (Refer to P.27)
Fuse is likely blown	• Not specified fuse used → Use 3A or 1.5A fuse according to the voltage being used
Unstable incubation temperature	• Poor connection of thermistor → Positive connection • Fuse holder cap not firmly fastened → Fasten firmly the cap • Voltage of house current fluctuates irregularly → Use a stabilizer

ELECTRIC SPECIFICATIONS

DIAPHOT-TMD

Power source	100/120V 220/240V 50/60Hz
Halogen lamp	12V–50W (OSRAM 64610 or PHILIPS 7027)
Fuse	100/120V 2A/250V 220/240V 1A/250V

Incubator for DIAPHOT-TMD

Power source	100/120V 220/240V 50/60Hz
Fuse	100/120V 3A/250V 220/240V 1.5A/250V
Maximum power requirement	250W

Nikon reserves the right to make such alterations in design as may be considered necessary in the light of experience. For this reason, particulars and illustrations in this handbook may not conform in every detail to models in current production.

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