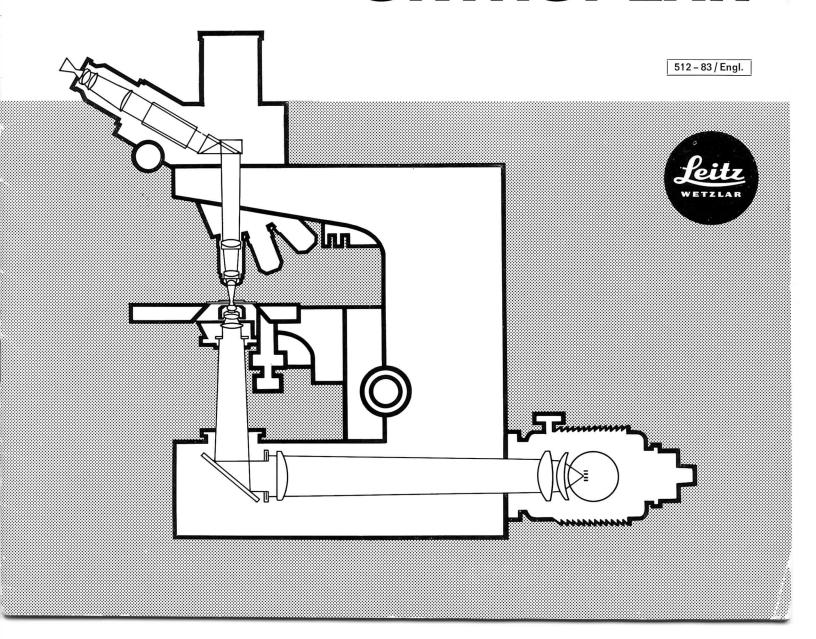
# ORTHOPLAN

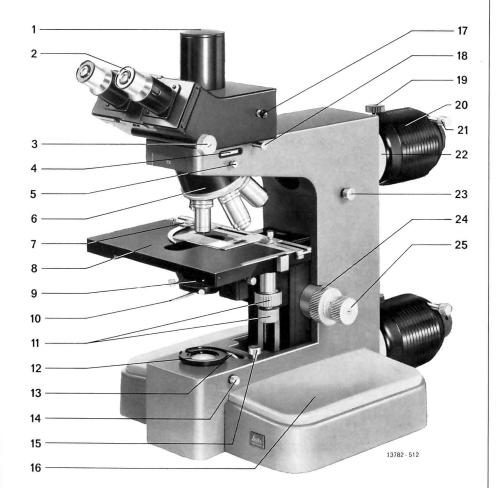


# Instructions ORTHOPLAN®

1)	Unpacking the microscope	3
2)	Assembling the microscope	3-5
3)	Technical details	5-9
	3.1 microscope tube	5
	3.2 objective revolving nosepiece	5
	3.3 microscope illuminators	6
	3.4 objectives	7
4)	Operating the microscope	10-11
	4.1 focusing the specimen	10
	4.2 centring the swing-out condenser	10
	4.3 change of magnification	11
	4.4 use of oil immersion objectives	11
5)	Transmitted-light darkfield	12
6)	Transmitted polarized light	13
7)	Phase contrast	13
8)	Fluorescence	14
9)	Incident-light darkfield	14
10)	Microscopic measurements	15
11)	Photomicrography	16–19
	11.1 with the 4x5" bellows camera	16
	11.2 attaching the bellows camera	17
	11.3 taking photomicrographs	17
	11.4 light sources for photomicrography	18
12)	Care and maintenance	19



1



# ORTHOPLAN

#### Fig. 1

- 1 Removable cover in the photo tube
- 2 GW/GG eyepiece in the eyepiece tubes
- 3 Knob for adjusting the interpupillary distance
- 4 Filter slide
- 5 Screw for clamping the revolving nosepiece
- 6 Revolving nosepiece, horizontally interchangeable
- 7 Object guide; traversing range of the specimen 76x52mm
- 8 Large square mechanical stage No 660
- 9 Swing-out condenser No 602
- 10 Aperture iris lever
- 11 Coaxial drives for the mechanical stage adjustment
- 12 Filter space
- 13 Field iris
- 14 Clamping screw for accessories
- 15 Adjustment knob of the swing-out lens
- 16 Plastic hand rests
- 17 Lever for operating the beam splitter; fully pulled out: all the light in the eyepieces, fully pushed in: 20% light in the eyepieces, 80% in the photo tube
- 18 Locking lever for tube change
- 19 Knurled knob for adjusting the condenser of the low-voltage lamp
- 20 12v 60W low-voltage lamp for incident light (the lamps are identical for incident and transmitted light)
- 21 Lamp socket clamping screw
- 22 Knurled ring for securing the low-voltage lamp
- 23 Adjustment knob for the upper swing-out lens (incident light)
- 24 Coarse adjustment
- 25 Fine adjustment

## 1) Unpacking the microscope

The following parts are packed in a special container:

- 1 microscope stand with changing devices for the attachable components
- 2 microscope tube
- 3 object stage with dovetail changer and condenser holder
- 4 one or two 12v 60W lamps
- 5 small parts such as objectives, eyepieces, condenser. dust-cover, etc.

Transformers and other heavy accessories are packed separately.

Check contents during unpacking carefully with the packing note and examine the packing material for small parts.

All mechanical and optical components are thoroughly cleaned before they leave the factory; any contact with dirt or dust should therefore be strictly avoided; on no account should the lenses of the objectives and eyepieces be touched by hand; any fingermarks on glass surfaces should be at once removed with a piece of soft leather or well-washed piece of lint. Even minute traces of finger perspiration may rapidly attack the surface of high-quality optical glasses.

# Workroom and -place:

The workroom has to meet some basic requirements. It should be as free as possible from dust, and oil- or chemical vapours likely to attack optical and mechanical parts of the microscope. It should be protected from major temperature fluctuations and from vibrations.

The mains socket for the built-in illuminators should include a 10 amp fuse.

# Ascertain correct type of current and voltage!

## 2) Assembling the microscope

- 1) Loosen clamping screw 26 (Fig. 5), insert and lower the object stage in the changing slide 27 until the top of the stage angle is flush with the top of the housing of the drive mechanism. Clamp the stage with screw 26. Fig. 2.
- 2) Lower the stage by means of the coarse adjustment and loosen clamping screw 5 so that the revolving nosepiece or the ULTROPAK® can be inserted fully in the horizontal slide changer 28. Retighten clamping srew 5. Fig. 3.



Fig. 2 Inserting the mechanical stage 27 dovetail changer

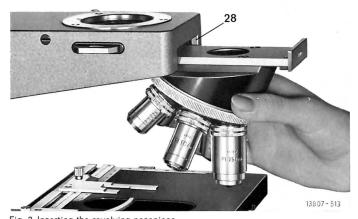


Fig. 3 Inserting the revolving nosepiece 28 Horizontal dovetail changer

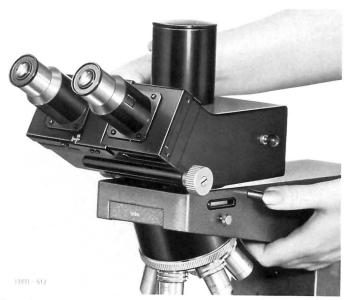
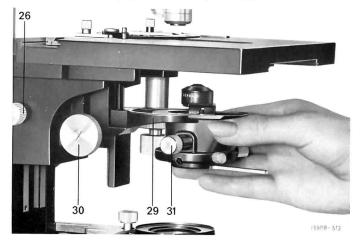


Fig. 4 Inserting the FSA-GW binocular tube

Fig. 5 Inserting the swing-out condenser No 602

- 26 Object stage clamping screw
- 29 Dovetail changer for the condenser
- 30 Knob for the vertical condenser adjustment
- 31 Rotating knob for swinging the condenser top in and out



3) Depress locking lever 18, mount the tube, which can be rotated at any angle on the bayonet changer. Release locking lever 18. After mounting it must be possible to rotate the body tube through  $360^{\circ}$  without difficulty. Fig. 4.

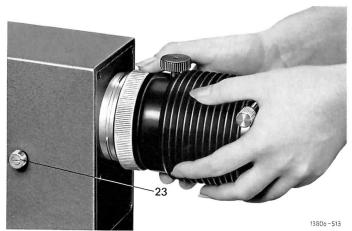
# 4) Inserting the condenser.

Lower dovetail slide 29 by means of knob 30 sufficiently for the swing-out condenser to be easily and fully inserted in the dovetail slide. The two centring screws must face the operator. Rotating knob 31 controls the position (in-out) of the condenser top. Fig. 5.

# 5) Lamp attachments

Identical 12v 60W lamp attachments are used for transmitted and incident light. They are pushed into the bottom and top aperture of the stand respectively and securely attached to the stand by means of the knurled ring 22 (knob 19 must be on top). The lamp attachments must be connected to the mains through their transformers (a. c.) only. Figs. 6 and 7.

Fig. 6 Inserting the low-voltage lamp for incident light 23 Adjustment knob for the upper swing-out lens

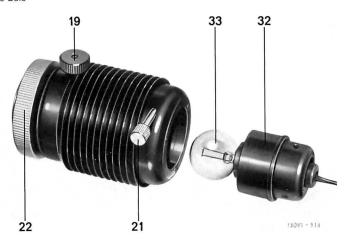


#### Inserting the lamp

Loosen clamping screw 21, pull out lamp socket 32 from the lamp housing. Insert the bulb in the socket so that the small projection on its base engages in the smaller recess of the lamp socket. This is the only position in which the bulb can be pushed into the socket and locked by a clockwise turn. Wipe any fingermarks off the bulb. Replace lamp socket in the lamp housing and clamp with screw 21. Fig. 7.

Fig. 7 12v 60W low-voltage lamp

- 19 Knurled knob for adjusting the lamp condenser
- 21 Lamp socket clamping screw
- 22 Knurled ring for securing the low-voltage lamp
- 32 Lamp socket
- 33 Bulb



## 3) Technical details

The various structural elements such as object stages, tubes, revolving nosepieces etc., are described in detail in our ORTHOPLAN list. A special description of various elements necessary for their efficient use will be found in the following paragraphs.

#### 3.1 Tube

The FSA-GW binocular tube is designed for eyepieces of 30mm diameter. It includes an adjustable beam splitter dividing the light intensity at a ratio of 80:20 (80% for photography, 20% for visual observation) or directing the entire light into the eyepiece tubes. The interpupillary distance is adjusted with knob 3 (Fig. 1). Here the optical tube length compensation maintains full image sharpness in the eyepiece and in the film plane for any interpupillary distance; if this is unknown, the binocular tube is adjusted during observation until a single, circular field of view appears, which can be easily surveyed. The GW and GG eyepieces have focusing eyelenses.

Eyepieces of 23.2mm diameter are used with sleeve adapters.

#### 3.2 Revolving nosepiece

Fig. 8 Revolving nosepiece with 5 plano objectives



The revolving nosepiece (Fig. 8) slides out horizontally and can be replaced by other optical elements, such as the ULTROPAK® illuminator. It accepts five objectives; the threads are numbered. Each outfit is accompanied by an objective/eyepiece chart (magnification table) which states, among other details, the nosepiece threads with which the individual objectives are matched. According to the optical systems used the nosepiece can be supplied with the 1x or the 1.25x tube lens. It must, however, be borne in mind that with the 1.25x tube factor the diameter of the field of view must be divided by 1.25; the 1x tube lens should therefore be used with plano objectives. The table below provides further details.

The tube lens can be exchanged in our factory or by our service agents only, as this requires a readjustment of the nosepiece.

Method of investigation	Recommended eyepieces and objectives	Preferably for revolving nosepiece with tube lens
Transmitted-light brightfield	Plano objectives and GW- or GG eyepieces	1x
Transmitted-light darkfield	Standard objectives or special objectives with iris diaphragm etc. and GF eyepieces	1.25x
Fluorescence	Generally standard ob- jectives and GF eyepieces	1.25x
Phase contrast and phase contrast fluorescence	Special phase contrast objectives and GF eyepieces	1.25x
Incident-light darkfield with ULTROPAK	Special UO objectives and GF eyepieces	Intermediate image system 1.25 with ULTROPAK
Polarized light	at the design stage	_

# 3.3 The microscope illumination

The sharpness and general character of the image are decisively affected by the microscope illumination. In addition to the correct choice of objective and magnification careful adjustment of the illumination, which must be selected to suit the microscope specimen, is one of the basic requirements of

making the best possible use of the microscope. The microscope illumination, i. e. the beam from the light source to the object must meet the following conditions:

- 1) The cross section of the beam in the microscope must be adjusted to the given objective/eyepiece combination.
- It must be possible to limit the cross section of this beam at will.

The first requirement is fulfilled when the microscopical field of view is completely and evenly illuminated. The exit pupil (usually the rear lens of the objective) must also appear evenly illuminated when observed in the tube after removal of the eyepiece.

However, the various objective/eyepiece combinations require different maximum cross sections of the illuminating beam; contrary to expectations it is the lowest-power objective with its wider field coverage which calls for the maximum cross section. The condenser must therefore be computed to produce a beam whose cross section is large enough for the lowest-power objectives. However, this leaves a surplus for higher-power objectives which cannot be fully utilized and may indeed have the adverse effects of reducing contrast, producing reflections and an unnecessary heating of the specimen. The cross section of the beam must therefore be reduced to the appropriate and useful value for all combinations. This facility is provided by two diaphragms suitably situated in the illuminating beam path.

The image of one of the diaphragms limits the illumination in the specimen. When this diaphragm is closed it will be visible in the microscope as a field diaphragm. It thus meets the second requirement; by opening or closing it, the diameter of the beam for illuminating the field of view can be adjusted as necessary.

The purpose of the second diaphragm is to control the cross section of the beam in the exit pupil (or rear lens of the objective). It must therefore appear as an aperture diaphragm, i.e. as a circular boundary of the portion of the rear lens of the objective filled with light when the diaphragm is closed after removing the eyepiece. Fig. 9.

In the exact Köhler's illumination realized in the ORTHOPLAN the field diaphragm is situated between the lamp condenser and the deflecting mirror, and the aperture diaphragm in the substage condenser. The section "Centring the swing-out



Fig. 9 Image of the aperture iris in the objective (eyepiece removed)



Fig. 10 Condenser bottom No 600 with the two condenser tops Achr. 0.90 and Apl. OI 1.25. 10 Aperture iris lever 31 Rotating knob for swinging the condenser top in and out 34 Centring screws 35a Condenser top No 002 35b Condenser top No 003

#### Swing-out condensers for slide change

# Transmitted-light brightfield

	0 0	
Code No	Description	Use
600	Bottom part of condenser, N. A. 0.25 with aperture stop, condenser for low magnifications and	With objectives up to N. A. 0.25. It illuminates even large fields at low magnifications
002	three-lens condenser top Achr. 0.90	
602	= achromatic condenser Achr. 0.90 with good spherical and colour correction. This condenser produces an almost colourless image of the field diaphragm or	With more highly corrected objectives such as fluorite systems, apochromats, plano objectives, and in photomicrography. Limited suitability for fluorescence.
003	4-lens condenser top Apl. Oil 1.25	
603	= achromatic-aplanatic condenser. State of correction similar to that of Condenser 602 at high aperture	Mainly for work with highly corrected oil immersion objectives and for photomicrography in colour with large condenser aperture. In order to make full use of the large aperture of this condenser, immersion oil must be introduced between the front lens of the condenser and the underside of the microscope slide when oil immersion objectives are used. Unsuitable for fluorescence.

condenser" contains instructions about the use of these two diaphragms, p. 10.

The standard outfit of the ORTHOPLAN includes the achromatic swing-out condenser No. 602, N. A. 0.90, Fig. 10. Its aperture is large enough to fill practically all dry objectives with light. Oil immersion objectives, too, can be used with this condenser; only where the recognition of the most minute structures is essential is a condenser aperture larger than 0.90 required; our aplanatic-achromatic swing-out condenser No 603 meets this need with an aperture of 1.25. The table above on the right gives details about the properties of these condensers.

The 12v 60W low-voltage lamp is described on p. 18. Special instructions are available or being prepared for the lamp housings 250 and 500 respectively.

#### 3.4 Objectives

In addition to our firm's emblem, a number of data are engraved on the mount of every objective; it is important for the user to know them. Fig. 11.

#### 170

represents the distance in mm from the shoulder of the objective to the rim of the microscope tube. This distance is also

called the mechanical tube length. LEITZ objectives for transmitted light are corrected exclusively for a mechanical tube length of 170mm. With our binocular tubes it is not possible to maintain the correct mechanical tube length because the optical components such as prisms, beam splitters etc. are moved when adjusting the interpupillary distance. Nevertheless the objectives are correctly used on such tubes, as a tube lens projects the image into the new intermediate image plane without impairing the image quality.

By a suitable choice of tube lenses it is possible to have the image formed in the intermediate plane either at the same ratio of magnification or magnified by a certain factor, which is engraved on the nosepiece. Fig. 12. It must be considered in the calculation of the final magnification (final magnification = objective magnification x eyepiece magnification x tube factor). For reasons of convenience the factors 1 or 1.25 are chosen in this case. See also table p. 6.

#### 0.17

denotes the coverglass thickness for which our transmitted-light objectives are as a rule computed. The table on p. 9 indicates any permissible deviations (without effect on the image quality) and those objectives which can be used either with or without coverglass.

Below the figures indicating the tube lens and coverglass thickness the reproduction ratio (= ratio of intermediate image and object, e.g. 40:1) and the numerical aperture of the objective are given in an abbreviated form, in this case 40/0.65. The term "magnification" is usually given in tables instead of "reproduction ratio" (it is also called primary magnification of the objective). In addition, the state of correction is indicated in the case of Fluorite systems, Apochromats, and Plano objectives. Also, the type of immersion media to be used is engraved on the mount, together with a black ring. Highpower objectives have a spring-loaded front lens mount for the protection of specimen and objective.

Fig. 11 5 plano objectives, with cross sections



Fig. 12 Revolving nosepiece with tube factor engraved



Description of objective  Magnification/Aperture		Focal length mm	Free working distance mm	Cover- glass correc- tion 1)	Type of eye- piece <sup>3</sup> )
Plano objectives	PI 1/0.04 PI 2.5/0.09 PI FI 4/0.10 PI FI 10/0.30 PI 25/0.50 PI 40/0.65 PI Apo OI 100/1.32	33 56 40 18 7.6 4.6 2.4	30 12 14 7.1 0.90 0.58 0.27	D O D O D O D O D O D O D O D O D O D O	GW/GG GW/GG GW/GG GW/GG GW/GG GW/GG
Achromatic dry systems	2.5/0.07 3.2/0.12 3.5/0.10 6/0.18 10/0.25 25/0.50 40/0.65 63/0.85 Iris 63/0.85	57 40 32 23 16 7.1 4.5 2.9 2.9	14 35 23 17.5 5.7 0.92 0.67 0.29 0.29	D O D O D O D D D D D D D D D D D D D D	P
Achromatic Immersion (W = Water Imm.) objectives	OI+W 22/0.65 W 90/1.20 OI 100/1.30 Iris OI 100/1.30-1.10	8.1 2.1 1.9 1.9	0.32 0.09 0.13 0.13	D O D D <sup>4</sup> ) D	P P P
Fluorite dry systems	FI 40/0.85	4.3	0.38	D!	Р
Fluorite oil immersion objectives	FI ÖI 54/0.95 FI ÖI 95/1.32 Iris FI ÖI 95/1.32-1.10	3.4 2.0 2.0	0.22 0.15 0.15	D D <sup>4</sup> ) D	P P P
Apochromatic dry systems	Apo 12.5/0.30 Apo 25/0.65 Apo 40/0.95 Apo 63/0.95	13 7.3 4.5 3.0	2.5 0.86 0.12 0.12	D O D D! <sup>2</sup> ) D! <sup>2</sup> )	P P P
Apochromatic oil immersion objectives	Apo OI 90/1.32 Apo OI 90/1.40	2.0 2.0	0.12 0.06	D D	P P

- with coverglass, thickness 0.17mm (adhere to coverglass thickness

D: — with coverglass, thickness u.1/mm (agnere to coverglass unickness within  $\pm$  0.05mm). O: — without coverglass DO: — with or without coverglass. D l: — Adhere to 0.17mm coverglass thickness within  $\pm$  0.01mm; where the objective has a correction mount set it at the real coverglass thickness within this table. this tolerance.

These objectives are fitted in a correction mount with automatic sharpness compensation. During adjustment image sharpness is almost completely maintained. Ideal possibility of optimum focusing where the thickness of the coverging indicates.

glass is unknown.

3) GW/GG = to be used preferably with GW or GG PERIPLAN widefield eye-

= use with GF PERIPLAN widefield eyepieces

H = use with Huygens eyepieces

4) These oil immersion objectives can be used also for uncovered specimens (smear preparations without coverglass); the negligible reduction in image quality does not prove disturbing.

#### PERIPLAN® widefield eyepieces GW/GG (30mm diameter)

Designation	Field of view index		
GG 8x M	24		
GW 6.3x	28		
GW 10x	24		

#### PERIPLAN widefield eyepieces GF (23.2mm diameter)

Designation	Field of view index	
GF 10x	18	
GF 10x M Graticule 10mm = 100 intervals	18	
GF 16x	15	
GF 25x	10	
GF 25x M Graticule $10mm = 100$ intervals	10	

\* suitable for use only in sleeve adapters

#### Field of view of the eyepiece

The field of view of an eyepiece is the area of the intermediate image in the tube which can be surveyed with it. It appears magnified at the magnification of the eyepiece. Thus, the image diameter of a GW 6.3x eyepiece with 28mm fieldof-view diameter (field-of-view index 28) appears as large as an area of  $6.3 \times 28 = 176$ mm diameter located 250mm in front of the observer. It is also possible to intercept the image at this size with a groundglass screen 250mm above the eyepiece.

### Object field

By dividing the diameter of the field of view by the objective magnification and the tube factor the diameter of the observable object area is obtained.

With the eyepiece mentioned above, a 25/0.50 objective and tube factor 1, the diameter of the observed object field is therefore  $\frac{28}{25 \times 1} = 1.12$ mm

# Operating the microscope

#### 4.1 Focusing the specimen

Clamp microscope slide on the object stage; the object holders can be adjusted for any size of slide up to 130mm.

Choose a medium-power objective for the first observation; preferably 10/0.25 combined with eyepiece GG8xM. Raise the swingout condenser to its highest position, and swing condenser head into the beam path.

Open aperture iris 10 and field iris 13.

Tilt swing-out lens in the foot of the stand into the beam path with knob 15. The lens remains in this position for all examinations in bright- and darkfield.

Turn out focusing eyelenses on GW or GG eyepieces to the black ring.

Focus the specimen with coarse- and fine adjustment. 1 interval of the fine adjustment  $= 1 \mu$ .

Carry out, if necessary, corrections for faulty eyesight: look through the right eyepiece with the right eye, and focus the specimen with the fine adjustment. Look at the same area of the specimen with your left eye adjusting the eyelens on the left eyepiece until the same area appears sharp in the left eyepiece. The fine adjustment must not be altered during this operation. This setting must be accurately repeated after the condenser has been centred, and should be checked from time to time.

# 4.2 Centring the swing-out condenser

Focus specimen by means of the coarse- and fine adjustment. Close field iris completely and focus by vertical adjustment of the condenser 30 (Fig. 5).

Close aperture iris completely.

Align field iris in the centre of the field of view by means of the two centring screws 34 (Fig. 10) of the condenser (centring).

Open aperture and field iris to suit the objective in use and the object examined.

The following rules should be observed for the use of these diaphragms:

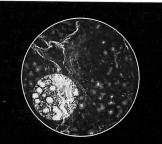
The field iris protects the specimen from unnecessary heat and prevents glare. It is therefore opened far enough only to be clear of the field of view of the microscope.

The aperture iris — provided it is smaller than that of the objective — determines resolution and contrast of the microscope image. Whereas for preparations of normal contrast range the aperture iris is closed so that it keeps only  $\frac{2}{3}$  of the objective aperture open, Fig. 9, the following procedure should be adopted for objects of poor contrast range: At first, the aperture iris is opened far enough to be just visible in the back lens of the objective (remove the eyepiece). The aperture of the condenser and that of the objective are now of the same diameter. If at this position of the aperture iris all object detail is adequately represented, the condenser iris is gradually closed until the less differentiated structural elements, too, become visible. In most cases it is advisable to close the aperture iris no further than  $^2/_3$  diameter of the full objective aperture (i.e. 2/3 of the objective aperture are transmitted). Closing it beyond this point rapidly reduces the resolving power of the objective and therefore the performance of the microscope.

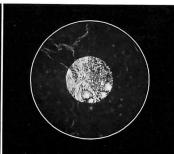
Fig. 13 Centring the field diaphragm a) field iris not yet in focus



b) field iris after focusing



c) field iris centred



d) field iris opened



10

The field iris must not be used for the control of image brightness; this is the exclusive task of the transformer or, in the case of colour photomicrography, of grey screens.

After the centration of the condenser the specimen must be illuminated. Since the lamp socket is already precentred in the 12v 60W lamp attachment, only the lamp condenser has to be adjusted for optimum illumination. Knurled screw 19, Fig. 7.

# 4.3 Changing the magnification

All LEITZ plano objectives have an adjustment length of 45mm and are parfocal on the nosepiece. As a result, only negligible refocusing with the fine adjustment is required during any change in the magnification. The table below contains instructions about the use of the swing-out condenser No. 602.

Objective aperture	Condenser top	Vertical adjustment of the condenser
≧0.25	remains swung in	Approximately in top position. A sharp image must be seen of the field diaphragm
<0.25	swing out	For visual observation approximately in top position; the image of the field diaphragm can be disregarded. For photomicrography lower the condenser until the field diaphragm appears sharp

# 4.4 The use of oil immersion objectives

Oil immersion objectives have the word "Oil" and a black ring at the lower rim engraved on the mount. It is a characteristic feature of the oil immersion objective that the refraction of the rays emerging from the coverglass is reduced if not altogether eliminated; at large angles of aperture total reflection from the surface of the coverglass is also absent. This enables rays of larger angles of aperture to enter the microscope objective which means an increase in its numerical aperture and therefore in its resolving power. The refractive index n=1.515 of the immersion oil is approximately the same as that of the coverglass and of the front lens of the microscope objective.

Focal length and working distance of immersion objectives are usually very short which indicates great care during work with such objectives. The coarse adjustment should be used only until the immersion objective has made contact with the immersion oil. This should be ascertained by inspecting it from the side. Focusing should be continuously controlled in the microscope and the fine adjustment should be used for it exclusively. Care should be taken that the immersion oil is free from air bubbles. LEITZ immersion oil, and, for fluorescence observations, non-fluorescing LEITZ immersion oil should be used.

Generally the condenser No 602 will be adequate also for oil immersion objectives; however, if the full aperture of an oil immersion objective is to be used, e.g. for extremely fine structures, the aplanatic achromatic condenser No 603, N.A. 1.25 is required, and immersion oil must be introduced between condenser head and the underside of the microscope slide. After the end of the examination all optical surfaces in contact with immersion oil must be carefully cleaned. A soft rag dipped in xylene is suitable for this purpose. The surface should now be polished with a dry rag. Alcohol or spirit must never be used for cleaning the objectives and no pressure should be applied.

# 5) Darkfield in transmitted light

For examinations in transmitted-light darkfield with the immersion darkfield condenser D 1.20 mainly the special darkfield objectives ÖI 100/1.30–1.10 and FI ÖI 95/1.32–1.10 with built-in iris diaphragm are used. These immersion objectives are available also without built-in iris diaphragm; for darkfield they must then be used with drop-in funnel stops in order to reduce the objective aperture, which is too high for the darkfield condenser D 1.20, below the limiting aperture of the condenser (here 1.20). Otherwise part of the illuminating beam would enter the objective, and the darkfield image would not be perfect. Large-aperture dry objectives can of course also be used with the darkfield condenser D 1.20.

For darkfield examinations with medium-power dry objectives, especially for serial examinations, the dry darkfield condenser D 0.80, which is simpler to use, is recommended. For dry objectives of apertures larger than about 0.70 a central stop should be inserted in the condenser D 0.80; it does not alter the limiting aperture of the condenser; it merely absorbs stray light.

# Focusing darkfield image with condensers D 1.20 and D 0.80

- Before inserting the darkfield condenser its centring mount should be adjusted to approximately its middle position by means of its centring screws. The darkfield condenser is now pushed fully into its holder on the microscope; however, it is not yet raised to its topmost position at this stage.
- 2) A drop of immersion oil of adequate size is placed on the surface of the condenser.
- 3) The darkfield preparation is placed in position and sharply focused with the objective 10/0.25. If the brightness is insufficient, lower the condenser.
- 4) The condenser is now raised by means of the rack-andpinion movement, and the moment observed when the drop of oil makes contact with the underside of the object slide (momentary light flash in the slide).
- 5) Looking into the tube, with the specimen in sharp focus, the condenser is further raised until the originally observed light ring contracts to the smallest possible light

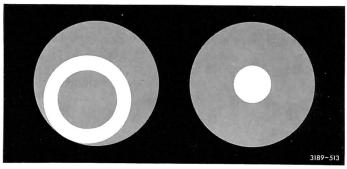


Fig. 14 Light ring and light spot during centration of the darkfield condenser

spot; Fig. 14. This spot is now centred in the field of view by means of the two centring screws of the darkfield condenser. The accuracy of centration can be further increased by making the original light ring (Fig. 14) concentric with the edge of the field of view. The specimen must be critically focused.

- 6) A higher-power objective on the revolving nosepiece is now turned in. Objectives of apertures larger than 1.15 must first be fitted with the appropriate drop-in funnel stop.
- 7) When an oil immersion objective is used a drop of immersion oil is placed also on the surface of the coverglass. See also use of oil immersion objectives in transmitted-light, brightfield. When special darkfield objectives are used maximum brightness in the field of view is obtained by operating the fine adjustment screw; at the same time, the structure of the specimen must become just visible; the diaphragm is now closed until a perfect darkfield is obtained. The object structures appear fringed with light. The various points about setting up the darkfield image apply analogously to the dry darkfield condenser D 0.80. Immersion oil is used neither between condenser and microscope slide nor between coverglass and objective. A preparation of buccal spirillae, easily produced at any time, is a suitable test object for setting up darkfield.

#### 8) Fluorescence

The ORTHOPLAN is suitable for fluorescence microscopy in transmitted and incident light, for which the following items are required: —

Achromatic objectives (if necessary more highly corrected systems) and revolving nosepiece, or ULTROPAK vertical illuminator for incident light; GF eyepieces. For plano objectives, GG or GW eyepieces, tube factor 1x.

Lamp housing 500 or 250 for the 200W high-pressure mercury lamp.

Exciting and absorbing filters (exciting filter in the lamp housing, absorbing filter in the filter slide).

Brightfield, darkfield or phase-contrast fluorescence condenser.

The following lists and instructions provide detailed information: —

List 52–20 "Equipment for fluorescence microscopy" Instructions 513–72 "Lamp housing 250" Instructions "Lamp housing 500"

(in preparation).

# 9) Incident-light darkfield

Use upper lamp attachment.

Swing out upper swing-out lens with knob 23 (Figs. 1 and 6). Insert light tube 45 (Figs. 18 and 19) in the upper lamp aperture.

Insert the ULTROPAK® incident-light illuminator. Fig. 18. The lamp condenser serves for the perfectly even illumination of the object field.

For details of the ULTROPAK incident-light illuminator see List 513-36.

The ULTROPAK is suitable also for orientating investigations in polarized light, when the polarizer is inserted in the front slot of the ULTROPAK; it can be rotated through 90°. The rear slot of the ULTROPAK is provided for sector stops. Two additional slots in the light tube 45 are used for colour filters if required. The analyser is pushed into the filter slide of the stand. Fig. 19.

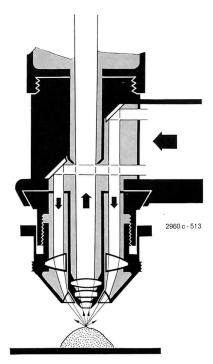


Fig. 17 Optical path in the ULTROPAK incident-light illuminator

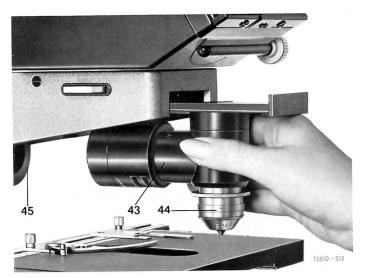
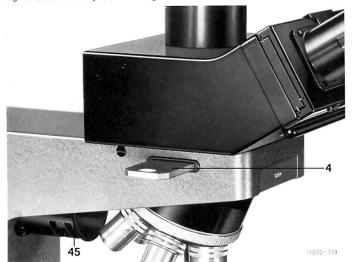


Fig. 18 Inserting the ULTROPAK 43 ULTROPAK housing 44 UO objective 45 light tube

Fig. 19 Slide for analyser, absorbing filters etc.



# 10) Microscopic measurements

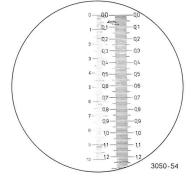
Linear measurements on the object are normally carried out with a micrometer eyepiece  $^*$  (usual graduation 10 mm = 100 intervals), either during visual observation through the inclined binocular tubes or on the focusing screen of the mirror reflex attachment.

Before measurements are begun the micrometer value of the objective in use must be known. This is the distance in the object plane which is just reproduced across a graduation interval in the micrometer eyepiece by the objective. Although these micrometer values are tabulated, the user is advised to determine them himself with the aid of a stage micrometer, since the optical constants of the objectives are subject to slight variations. The micrometer value remains unchanged for exposures with the bellows camera, since the microscopic image and the graduation of the measuring eyepiece change at the same rate when the bellows extension is changed.

Example: —

Determination of the micrometer value with the aid of a stage micrometer 2mm = 200 intervals and of a micrometer eyepiece with graticule 10mm = 100 intervals (visual observation): Fig. 20.

Fig. 20 Graduation of the graticule in the eyepiece and image of the stage micrometer (right)



Zero lines of measuring eyepiece and stage micrometer should be brought to coincidence. The micrometer value is read, with the setting unchanged, off the end of the micrometer eyepiece scale. Examples: If 1.220mm of the stage micrometer occupy 100 intervals of the micrometer eyepiece,

<sup>\*</sup> an eyepiece with a graduated glass plate (graticule)

the micrometer value will be 1.220:100 = 0.01220mm = 12.20  $\mu$ . For low-power objectives which do not form an image of the stage micrometer along the entire scale of the micrometer eyepiece, only 10 intervals of the micrometer eyepiece are used for comparison. If, for instance, 0.36mm of the stage micrometer take up 10 intervals of the micrometer eyepiece, the micrometer value will be 0.36:10 = 0.036mm = 36  $\mu$ .

The screw micrometer eyepiece is used for the most delicate measurements under the microscope. List 513–17 offers detailed information about this instrument.

# 11) Photomicrography

The ORTHOPLAN research microscope can be used with the following photographic attachments:—

ORTHOPLAN with camera carrier and 4x5" bellows camera ORTHOPLAN with ORTHOMAT® fully automatic microscope camera

ORTHOPLAN with micro-attachment for the LEICA®.

#### 11.1 ORTHOPLAN with 4x5" bellows camera

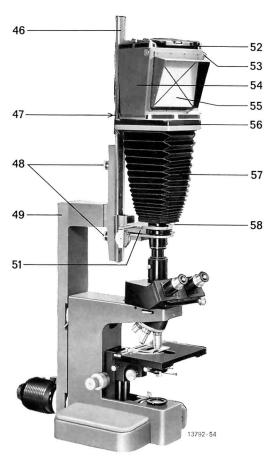


Fig. 21
ORTHOPLAN with 4x5" bellows camera
46 guide bar
47 clamping screw for the mirror reflex attachment (not visible in the
illustration)
48 guide bar clamping screws
49 camera holder
51 bellows support with shutter (see also Fig. 22)
52 darkslide
53 lever for swinging in the mirror
54 mirror reflex attachment
55 groundglass screen
56 bellows camera with rotating darkslide frame
57 bellows
58 bellows locking lever (see also Fig. 21a)

The following items are required:—
sleeve adapter 64 for the photo-tube for 30mm eyepieces
GW or GG eyepieces 62
bellows support 51 and upper light screening collar 66
camera holder 49
guide bar 46
9x12cm or 4x5" bellows camera 57
darkslides 52
cable release 61
lower light screening collar 65



Fig. 21a Synchronized time- and instantaneous shutter 58 bellows locking lever

59 tape measure

focusing magnifier 63

60 marking on the lower light screening collar



Fig. 22 Various parts for photomicrography with the bellows camera 46 quide bar

- 50 knurled screw for adjusting the bellows support
- 51 bellows support with shutter
- 61 cable release
- 61a shutter speed dial
- 62 GW/GG eveniece
- 63 focusing magnifier
- 64 sleeve adapter for GW/GG eyepiece and photo tube
- 65 lower light screening collar
- 66 upper light screening collar screwed into the bellows support 51

#### 11.2 Attaching the bellows camera

Remove the 12v 60W lamp attachment from the stand. Fix camera holder 49 to the stand by means of the two bayonet attachments; press locks downwards. Screw the guide bar 46 to the camera holder 49 with the two screws 48. Place lower light screening collar 65 on the photo tube.

Insert eyepiece.

13792a-54

Turn out eyelens of the eyepiece as far as the black marking ring.

Push bellows support with shutter 51 into the guide bar (upper light screening collar 66 facing downwards) and lower by hand or with drive 50 until the lower rim of the upper light screening collar is flush with the appropriate marking on the lower light screening collar. For the GW 10x eyepiece, for instance, it is the marking GW 10x. See Fig. 21 a.

Mount shutter on the guide bar.

Push bellows camera onto the guide bar and clamp with screw 47.

Lock bellows camera with shutter.

Lever 38 to the left. Replace and secure 12v 60W lamp attachment.

#### 11.3 Taking photomicrographs

After focusing the microscopic image in the observation tube and selecting the portion of the specimen to be photographed, final adjustments are carried out on the groundglass screen of the mirror reflex attachment:—

- 1) Set the speed dial 61 a at "T", and open the central shutter with cable release 61.
  - Turn in deflecting mirror of the mirror reflex attachment (lever 53 to the rear). If necessary adjust lamp brightness\*.
- 2) Loosen clamping screw 47, position the desired picture area by vertically adjusting the mirror reflex attachment. If necessary rotate the attachment until the optimum picture area has been obtained. Retighten the clamping screw after adjustment of the mirror reflex attachment.
  - \* During colour photography the colour temperature of the filament lamp must be matched to the colour film used, see also 11.4 light sources for photomicrography.

3) Refocus the image on the groundglass screen 55 with the fine adjustment of the microscope.

Use the focusing magnifier.

First adjust magnifier for your own eyesight, placing the magnifier on the centre of the groundglass screen and focusing it on the crosslines engraved on it; do not keep eye too close to the magnifier.

- 4) Set the field iris of the microscope so that it just disappears beyond the edge of the groundglass screen image. The setting of the aperture iris depends on the object and its structure.
- 5) The magnification on the groundglass screen can be determined as follows:— Measure the bellows extension from the white ring on the light screening collar to the top of the darkslide frame with the tape measure 59. The magnification V is obtained from the formula:

V = objective magnification x eyepiece magnification x tube factor x bellows extension (cm)

7	

Example:— Objective magnification eyepiece magnification tube factor bellows extension			10x 8x 1x 35cm
	distance of clear vision to which all the values are referred $V = 10 \times 8 \times 1 \times \frac{35}{25}$	=	25cm 112x

The magnification can be found more simply and accurately by using a stage micrometer 2mm=200 intervals in place of the specimen on the object stage. The graduation projected on the groundglass screen is measured with a ruler.

Example:— Let 1 interval on the stage micrometer  $\binom{1}{1_{00}}$ mm) on the groundglass screen be 10mm Image size/object size =  $10:\frac{1}{1_{00}}$  = 1000. The magnification is 1000x.

- 6) Insert photographic filters if required.
- 7) Insert loaded darkslide between pressure frame and darkslide frame.
- 8) Measure the exposure time with the micro exposure meter either on the groundglass or on the clear glass screen. See Instructions MICROSIX-L 54-21.

- 9) Swing in deflecting mirror with lever 53.
- Close camera shutter with cable release, set shutter speed.
- 11) Pull out slide of darkslide so that it is just held in its grooves.
- 12) Expose.
- 13) Immediately return slide into the darkslide which is now removed or replaced (interchangeable darkslide).

Photomicrography with the ORTHOMAT see Instructions 54–19b, with the micro-attachment for the LEICA see Instructions 54–24.

# 11.4 Light sources for photomicrography

# 1) 12v 60W low-voltage lamp

The 12v 60W low-voltage filament lamp used in the ORTHO-PLAN is a special bulb with a flat-core filament. Its light is very bright and of a spectral composition favourable to microscopy and photomicrography. Its load can be controlled with a regulating transformer (normal load 5 amp; maximum lamp current 6 amp.).

Colour temperatur 2850° K at 5 amp.

3200° K at 5.7 amp.

 $3400^{\circ}$  K at 6 amp.

Intermediate values can be obtained from the graph, Fig. 23. A green filter, a diffusion disc, and a conversion filter CB 12 are included with the low-voltage lamp.

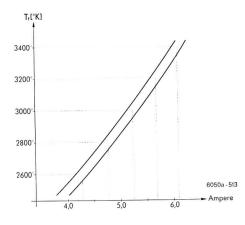


Fig. 23
Dependance of the colour temperature on the current intensitiy for the 12v 60W filament lamp

# 2) High-pressure xenon lamp

High-pressure xenon lamps 150W can be used in our lamp housings 250 or 500.

#### Technical data of the high-pressure xenon lamp

Practically continuous spectrum in the visible range and in the medium and long-wave ultra-violet range;  $T_f=6000^\circ$  K, so that daylight colour films can be used without a filter (Fig. 24).

Luminous density of cathode spot above 100,000 sb of arc above 8,000 sb effective luminous density 15,000–40,000 sb depending on optical arrangement

mean light intensity 350cd. Light flux 3200lm.

Average life 1,200 hours (These particulars apply to about 2 switching operations per operating hour. Frequent switching on and off reduces the life of the lamp).

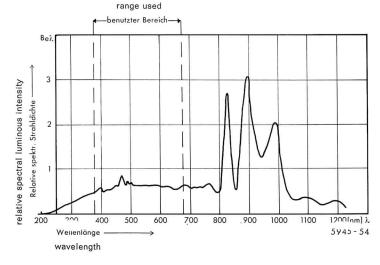
Lamp current 7.5 amps, positive pole top.

lamp voltage approx. 20v,

i.e. rated load 150W.

Further details will be found in the Instructions for the lamp housing 250 or 500.

Fig. 24 Dependence of the relative spectral luminous density on wave length for the high-pressure xenon lamp  $150\mathrm{W}$ 



#### 12) Care and maintenance

For protection against dust the microscope should always be covered with the flexible dust cover after use. From time to time the stand should be cleaned with a piece of linen or chamois leather. Spirit should not be used for this purpose on any account as it attacks the varnish. Benzene, on the other hand, is eminently suitable for the cleaning of varnished parts.

Light spots on the object stage caused by benzene can be removed by rubbing over with liquid paraffin or acid-free vaseline.

Work with acids (above all acetic acid) or corrosive chemicals requires special care. Direct contact of these chemicals with the optical equipment and the stand must be strictly avoided; all parts should be thoroughly cleaned after use.

The optical parts of the microscope should be kept minutely clean. Dust on glass surfaces is removed with a fine, dry sable brush, blowing gently across the glass surface while using the brush. If the dirt resists this treatment, a well-washed piece of lint or chamois leather moistened with a little benzene or xylene is recommended. On no account should spirit or alcohol be used.

Objectives must not be dismantled for cleaning. If internal damage appears in an objective the objective should be sent to our factory for overhaul.

Particular care is recommended during the cleaning of the antireflex coating. The external surface of the eyepieces and front lenses of the objectives are coated with films of approximately the hardness of glass. They are cleaned as carefully as uncoated class surfaces. However, internal surfaces of objectives and eyepieces are sometimes coated with extremely soft films which must be cleaned by very gentle blowing only. For the reason given above, it is not recommended to clean internal surfaces of eyepieces.

Correct treatment maintains the performance of a LEITZ microscope for many years. Any examination or repair which might become necessary should be entrusted to our factory or to one of our official agencies.

Design subject to alterations without notice.

# Weight and dimensions

Foot: length x width Height without tube Height with tube Eye level Total weight approx. Instrument table Working top	300x295mm 340.5mm 517mm 413mm 17 kg 1500x600mm 1200x630mm	12x11.8" 13.6" 20.7" 16.5" 37 lbs. 60x24" 48x25.2"
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