

**Axio Imager**  
**Upright Microscope**  
**Operating Manual**

Knowledge of this manual is required for the operation of the instrument. Therefore it's mandatory to make yourself familiar with the contents of this manual and pay special attention to hints concerning safe operation of the instrument.

The specifications are subject to change; the manual is not covered by an update service.

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Issued by: Carl Zeiss Microscopy GmbH  
Carl-Zeiss-Promenade 10  
07745 Jena, Germany

[microscopy@zeiss.com](mailto:microscopy@zeiss.com)  
[www.zeiss.com/microscopy](http://www.zeiss.com/microscopy)



Carl Zeiss Microscopy GmbH  
Königsallee 9-21  
37081 Göttingen, Germany

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## 1 INTRODUCTION

### 1.1 Notes on instrument safety

The Axio Imager microscopes have been designed, produced and tested in compliance with DIN EN 61010-1 (IEC 61010-1) and IEC 61010-2-101 safety requirements for electrical measuring, control and laboratory instruments.

The instruments meet the requirements of EC Directive IVDD 98/79/EC (In Vitro Diagnostic); they are marked with the **CE** mark.

The instruments are to be disposed of in accordance with EU Directive 2002/96/EC (WEEE).

This operating manual includes information and warnings that must be observed by the user.

The following warning and information symbols are used in this manual:

**NOTE**

*This symbol is a warning, which you must observe under all circumstances.*

**CAUTION**

*This symbol is a warning, which indicates a hazard to the instrument or instrument system.*

**CAUTION**

*This symbol is a warning, which indicates a hazard to the user of the instrument.*

**CAUTION**

*Hot surface!*

**CAUTION**

*UV radiation is emitted!*

**CAUTION**

*LED radiation ranging from 400 nm to 700 nm!  
Do not look into the LED illuminator.*

**CAUTION**

*Disconnect the instrument from line power before opening it!*

**CAUTION**

*Crushing hazard*

The Axio Imager microscopes, including original accessories, may only be used for the microscope techniques described in this manual.

Particular attention must be paid to the following warnings:

 The manufacturer cannot assume any liability for any other applications of the instrument including those of individual modules or single parts. This also applies to all service or repair work that is not carried out by authorized service personnel. In case of non-compliance, all warranty claims shall be forfeited.

 The power plug must be inserted in an outlet featuring a grounding (earth) contact. The grounding effect must not be made ineffective by an extension cable that does not have a protective ground wire.

 If it is determined that protection measures are no longer effective, the instrument must be switched off and safeguarded against inadvertent operation. Please contact a Zeiss service agency or the Carl Zeiss Microscopy Service to repair the instrument.

 On stands with motorized focusing drive, there is the risk of getting one's fingers jammed between stage carrier and the base of the stand, when the stage is moved down. Therefore, do not reach with your hands under the stage carrier.

 The manual microscopes (Axio Imager.A2, .A2 LED, .A2m, .D2 and .D2m) are equipped with a power supply integrated in the stand allowing line voltages to be used in the ranges 100 to 127 V and 200 to 240 V AC  $\pm 10\%$ , 50/60 Hz, without the voltage setting on the instrument having to be changed. The motorized models (Axio Imager.M2, .M2m, .Z2 and .Z2m) are powered through the separate power supply VP232-2 belonging to the stand. On this power supply, too, the voltage setting need not be changed in the line voltage range 100 to 240 V  $\pm 10\%$ , 50/60 Hz.  
The HBO 100 (ebq 100 dc) and XBO 75 (ebx 75 isolated) power supply units are designed for a line voltage range from 100 to 240 V AC, 50/60 Hz, and automatically adapt to the applied line voltage.

 Before switching on the instrument, check whether it is suitable for the available line voltage. Always disconnect the instrument from the power outlet before opening the instrument and before changing the fuses. See Section 5.2.2.

 Take care to ensure you only use fuses according to the information given in the Technical Data. Use of makeshift fuses and short-circuiting of the fuse holders are not permitted.

  The Axio Imager microscopes are not equipped with any special devices for protection from substances that are corrosive, potentially infectious, toxic, radioactive, or other substances that could be hazardous to health. Make sure to observe all legal regulations, particularly the relevant national accident prevention regulations when handling such substances.

  Do not put defective instruments in household waste. They have to be disposed of in accordance with applicable legal provisions. Also, specimens need to be properly disposed of in conformity with applicable legal provisions and in-house work instructions.



Gas-discharge lamps, e.g. HBO 50; HBO 100 or XBO 75, emit ultraviolet radiation, which can cause burns to the eyes and skin. Therefore, never look directly into the light of these lamps and avoid direct, unprotected incidence of their light on your skin. When using the microscope, always use the protective devices belonging to the instrument (e.g. special attenuation filters or the fluorescence shield). When they are hot, gas-discharge lamps are under high internal pressure. Therefore, change them only when they have cooled down, and make sure to wear protective gloves and a face guard.



When fluorescence filters are used, the heat protection filter for heat emitted by the microscope illuminator must not be removed, since fluorescence filters are sensitive to heat, and their performance could be impaired.



Avoid touching the hot lamp housing. Always pull the power plug before changing the lamps and allow the instrument to cool down for some 15 minutes.



Dust and dirt may impair the instrument's performance. Therefore, the instrument must be protected from these influences as far as possible and covered with the dust cover when not in use. Always check whether the instrument is switched off before you cover it.



Clogged or covered ventilation slats may lead to heat build-up that will damage the instrument and, in extreme cases, cause a fire. Always keep the ventilation slats clear and ensure that no objects enter the instrument through the ventilation slats.



The instruments may only be operated by trained personnel who must be aware of the possible dangers involved with microscopy and the particular application concerned. The Axio Imager microscopes are high-precision instruments that can be impaired in their performance or destroyed when handled improperly.



The LED illuminator is a Laser Class 2M device. Do not look directly into the LED light.



Do not put defective instruments in household waste. They have to be disposed of in accordance with applicable legal provisions.



Also, specimens need to be properly disposed of in conformity with applicable legal provisions and in-house work instructions.



For stands equipped with motorized focusing drives, there is a **risk of fingers getting crushed** in the working area when the microscope stage is being lowered.



- Do not reach into the area between the microscope stage / stage carrier / condenser carrier and the stand base while the stage is being lowered.
- The automatic movement can be stopped by pressing the STOP button on the TFT display. (See Fig. 1-2).



Bei Verwendung eines Scanningtisches (ca. 4 kg Gewicht) muss zum Schutz des Fokussiertriebs der Hochleistungsfokus aktiviert werden (siehe Seite 49). Probengewichte bis maximal 5 kg können bei aktiviertem Hochleistungsfokus auf den Tisch aufgelegt werden.



Make sure to read the safety notes provided with Immersol 518 F<sup>®</sup> immersion oil.



Immersol 518 F<sup>®</sup> immersion oil irritates the skin. Avoid any contact with skin, eyes and clothing. After inadvertent skin contact, wash the oil off with plenty of water and soap.

After inadvertent eye contact, immediately rinse the eye with plenty of water for at least five minutes. If the irritation persists, consult a specialist.



Proper disposal of Immersol 518 F<sup>®</sup> immersion oil: Take care to ensure that the immersion oil does not enter surface water or the sewage system.

## 1.2 Warning stickers

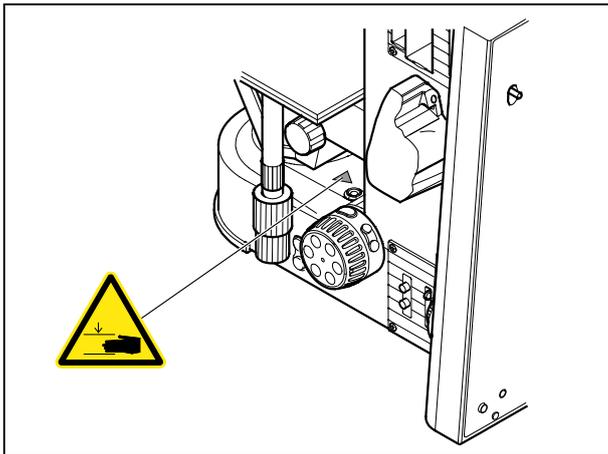


Fig. 1-1 "Crushing Hazard" warning sticker on the base of motorized stands



Fig. 1-2 STOP button

The STOP button on the TFT display serves to switch off the movement of the focusing drive instantaneously, so as to prevent a collision with the stage or the specimen placed on it.

Actuation of the STOP button causes the movement of the focusing drive to be halted immediately.

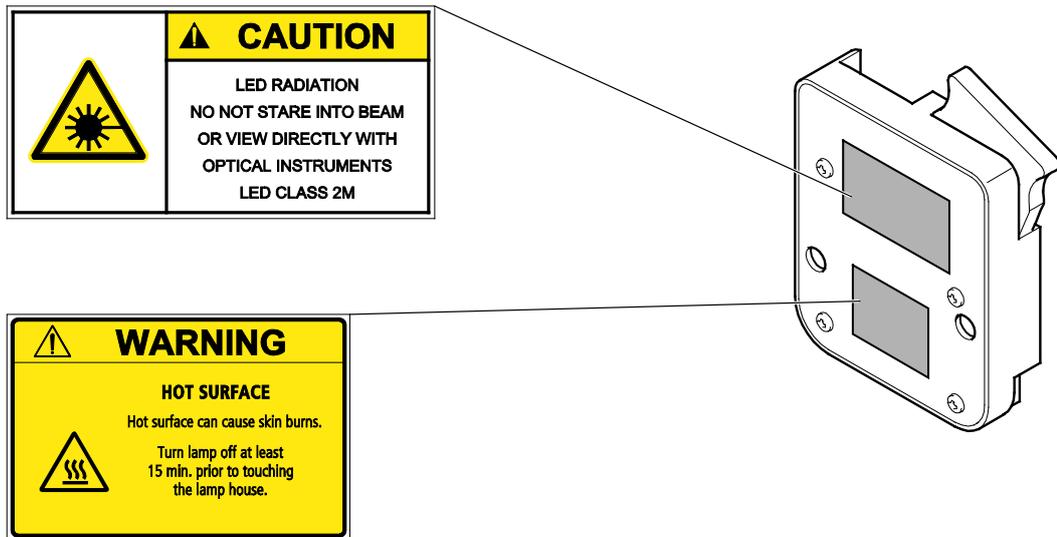


Fig. 1-3 Warning stickers on the underside of the LED illuminator

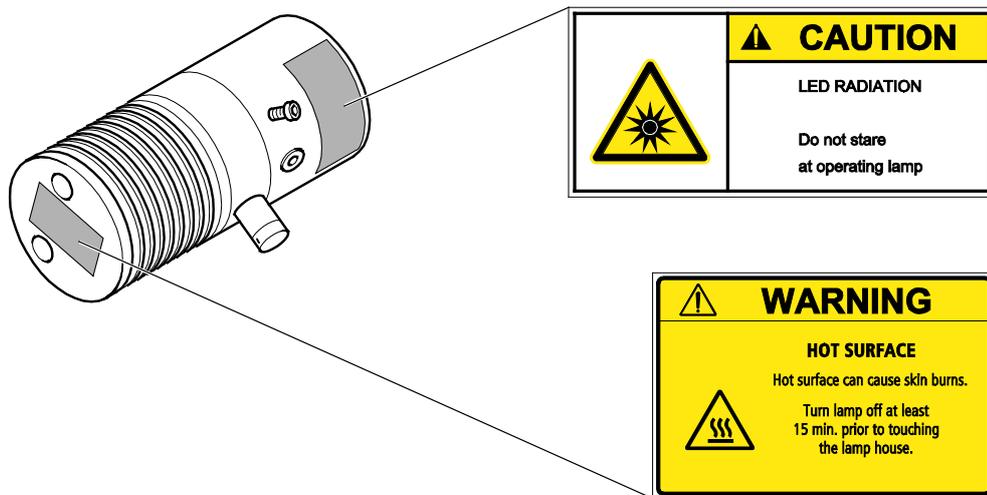


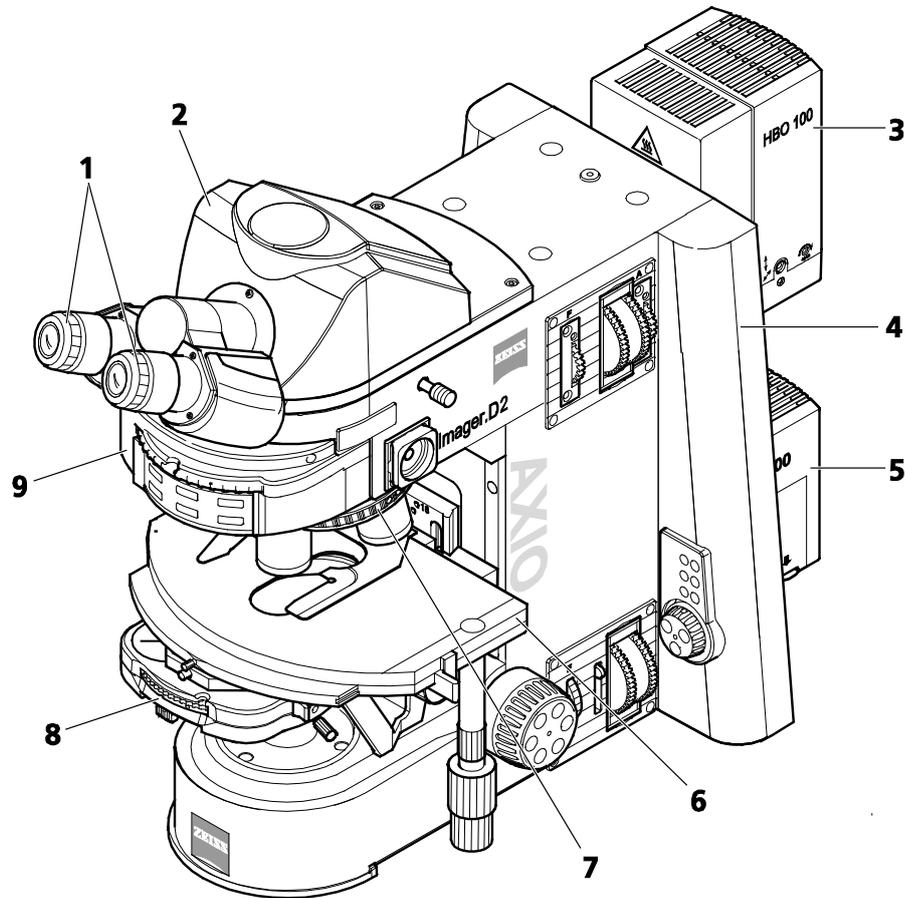
Fig. 1-4 Warning stickers on the attachment lamp VIS-LED

### **1.3 Notes on warranty**

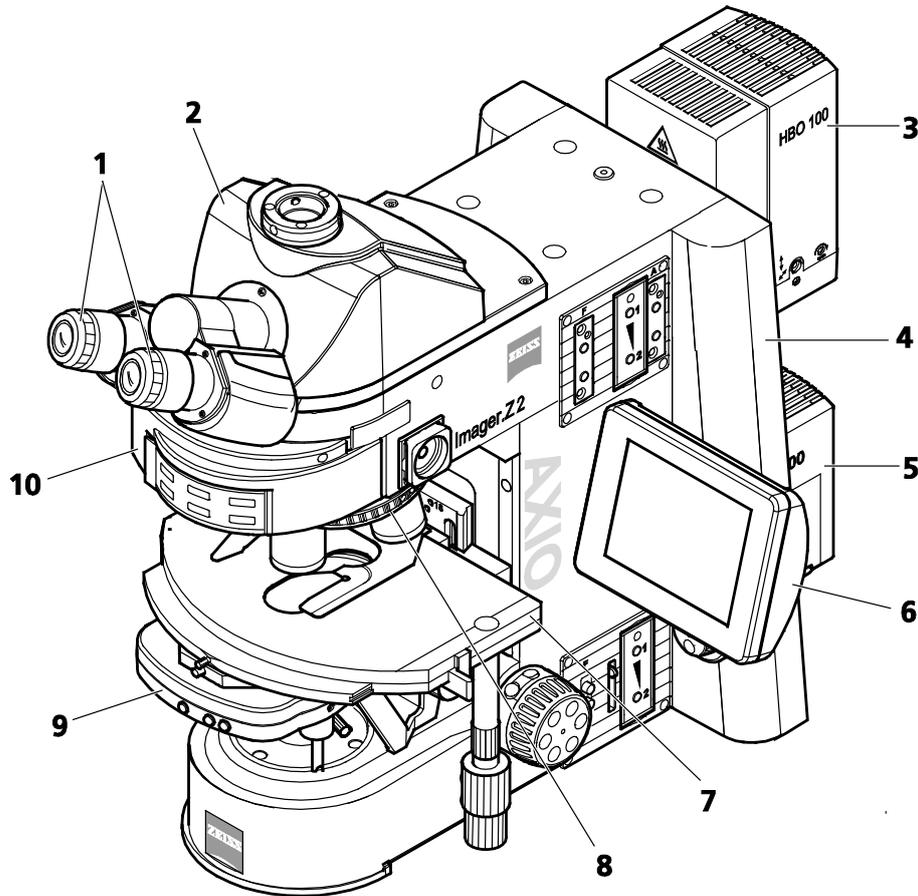
The manufacturer guarantees that the instrument has no material or production defects when delivered. You must inform us of any defects immediately and do everything to minimize any damage. If the manufacturer is informed of such a defect, he is obligated to rectify it; it is his decision whether he does this by repairing the instrument or by delivering an instrument free of any defect. No guarantee is provided for defects caused by natural wear (wearing parts and consumables in particular) and improper use.

The instrument manufacturer is not liable for damage caused by faulty operation, negligence or any other tampering with the instrument, particularly the removal or replacement of instrument components, or the use of accessories from other manufacturers. This forfeits all warranty claims.

With the exception of the work specified in this manual, no maintenance or repair of the Axio Imager may be undertaken. Repairs may only be performed by Zeiss service staff or specially authorized personnel. Should any defect occur with the instrument, please get in touch with your local Zeiss representative.

**1.4 Overall view of Axio Imager - manual**

- 1 Eyepieces
- 2 Binocular tube
- 3 Reflected-light illuminator (HBO 100)
- 4 Microscope stand, manual
- 5 Transmitted-light illuminator (HAL 100)
- 6 Mechanical stage
- 7 Nosepiece
- 8 Condenser
- 9 Reflector turret

**1.5 Overall view of Axio Imager - motorized**

- 1 Eyepieces
- 2 Binocular phototube
- 3 Reflected-light illuminator (HBO 100)
- 4 Microscope stand, motorized
- 5 Transmitted-light illuminator (HAL 100)
- 6 TFT display
- 7 Mechanical stage
- 8 Nosepiece
- 9 Condenser
- 10 Reflector turret

## 2 INSTRUMENT DESCRIPTION

### 2.1 Name and intended use

Manufacturer's designation:	<b>Axio Imager:</b>	<b>Axio Imager MAT:</b>	<b>Axio Imager LED:</b>
	Axio Imager.A2	Axio Imager.A2m	Axio Imager.A2 LED
	Axio Imager.D2	Axio Imager.D2m	
	Axio Imager.M2	Axio Imager.M2m	
	Axio Imager.M2p	Axio Imager.Z2m	
	Axio Imager.Z2		

The Axio Imager / Axio Imager MAT microscopes have been designed as universal microscopes for applications in biology and medicine for the examination of blood and/or tissue specimens from the human body, as well as for materials examinations.

They may also be used as true reflected-light microscopes or, if equipped with a transmitted-light equipment, as combined reflected-light/transmitted-light microscopes.

Typical applications of the Axio Imager microscopes include, for instance:

- Medical examinations in laboratories (research), clinics and medical practices
- Science and research (colleges, universities) in the fields of medicine and biology
- Industrial applications (pharmacology, food technology)

Typical applications of the Axio Imager MAT microscopes include, for instance:

- Metallographic laboratories
- Vehicle industry
- Microsystems engineering

### 2.2 Instrument description and main features

With the advanced pyramid and modular design, the Axio Imager microscope incorporates time-tested principles in microscope construction, thus ideally combining modern requirements made on design, ergonomics, operating convenience and function with technical performance.

Depending on the instrument configuration, the following microscopy and contrasting techniques are possible:

#### Transmitted light:

- Brightfield (H)
- Darkfield (D)
- Phase contrast (Ph)
- Differential Interference Contrast (DIC)
- Polarization contrast (Pol)
- Circular polarization

#### Reflected light:

- Brightfield (H)
- Darkfield (D)
- Differential Interference Contrast (DIC)
- Differential Interference Contrast in circularly polarized light (C-DIC)
- Polarization contrast (Pol)
- Fluorescence

The Axio Imager Microscope is available in ten stand versions. However, the scope of the equipment of these stands is variable and can be customized to the user's requirements within the range of optionally available microscope components.

Coded models:

Axio Imager.A2, A2m and A2 LED

Partially motorized models:

Axio Imager.D2, D2m

Motorized models:

Axio Imager.M2, M2m, M2p

Axio Imager.Z2 and Z2m

The binocular phototubes and suitable adapters permit one microscope camera, one reflex camera or one digital / video camera to be attached for documentation purposes.

**2.3 Equipment and compatibility table**

Component	Option	Type of Stand									
		A2 LED	A2	M2p	M2	D2	Z2	A2 m	M2 m	D2 m	Z2m
Stand	Manual	+	+	-	-	+	-	+	-	+	-
	Motorized	-	-	+	+	O*	+	-	+	O*	+
Coding	PC readable	+	+	+	+	+	+	+	+	+	+
Tube lens turret	Coded	O	O	O	O	O	O	O	O	O	O
	Motorized	-	-	O	O	-	O	-	O	-	O
Reflector turret	6 positions, coded	O	O	O	O	O	O	O	-	O	O
	6 positions, motorized	-	-	O	O	O	O	-	+	O	O
	6 positions, motorized ACR	-	-	-	-	-	O	-	-	-	O
	10 positions, motorized ACR**	-	-	-	-	O	O	-	-	O	O
Objective nosepiece	6 positions, coded POL	O	O	-	O	O	O	O	O	O	O
	6 positions, coded HD DIC	O	O	-	O	O	O	O	O	O	O
	6 positions, motorized HD DIC	-	-	-	O	-	O	-	O	-	O
	6 positions, motorized HD DIC ACR	-	-	-	O	-	O	-	O	-	O
	7 positions, coded HD	O	O	+	O	O	O	O	O	O	O
	7 positions, motorized HD	-	-	-	O	-	O	-	O	-	O
Modulator turret for C-DIC/TIC	Manual	O	O	O	O	O	O	O	O	O	O
	Motorized*****	-	-	-	O	-	O	-	O	-	O
Modulator turret for transmitted-light DIC	Motorized*****	-	-	-	-	-	O	-	-	-	O
Attachable stage carrier with condenser carrier	0 mm – 25 mm	+	+	+	+	+	O	O	O	O	O
Attachable stage carrier for detachable condenser carrier	0 mm – 45 mm	O	O	O	O	O	O	O	O	O	O
Attachable reflected-light stage carrier	0 mm – 63 mm	O	O	O	O	O	O	O	O	O	O
Transmitted-light illumination	Manual	-	+	-	-	+	-	O	O	O	O
	Motorized	-	-	-	+	-	+	-	-	-	O
Transmitted-light LED	-	+	O	+	O	O	O	O	O	O	O
Two-position transmitted-light filter wheel	Manual	-	+	-	O	O	O	O	O	O	O
	Motorized	-	-	-	O	-	O	-	-	-	O
Reflected-light illumination	Manual***	O	O	O	O	O	O	+	-	+	-
	Motorized***	-	-	-	-	-	O	-	+	-	+
Reflected-light luminous-field diaphragm	Manual	O	O	O	O	O	O	+	O	+	O
	Motorized	-	-	-	-	-	O	-	O	-	O

Component	Option	Type of Stand									
		A2 LED	A2	M2p	M2	D2	Z2	A2 m	M2 m	D2 m	Z2m
Reflected-light aperture diaphragm	Manual	○	○	○	○	○	○	○	○	○	○
	Motorized	-	-	-	-	-	○	-	○	-	○
Reflected-light two-position filter wheel	Manual	○	○	○	○	○	○	○	○	○	○
	Motorized	-	-	-	○	-	○	-	○	-	○
FL attenuator	Manual	○	○	○	○	○	○	○	○	○	○
	Motorized	-	-	-	-	-	○	-	○	-	○
Reflected light / transmitted light selection	Manual	+	+	-	-	+	-	+	-	+	-
	Software	-	-	+	+	-	+	-	+	-	+
Mixed light with extra power supply	Manual	+	+	-	-	+	-	+	-	+	-
	Software	-	-	+	+	-	+	-	+	-	+
Focus (z-axis)	Manual	+	+	-	-	+	-	+	-	+	-
	Motorized, 25 nm	-	-	+	+	-	-	-	+	-	-
	High-performance focus (motorized, 10 nm)	-	-	-	-	-	+	-	-	-	+
TFT display	-	-	-	○	+	-	+	-	+	-	+
Apotome	-	○	○	○	○	○	○	○	○	○	○
Power supply	External	-	-	+	+	-	+	-	+	-	+
	Internal	+	+	-	-	+	-	+	-	+	-
Mechanical CAN stages	Motorized****	○	○	○	○	○	○	○	○	○	○
Scanning stages	Piezo	○	○	○	○	○	○	○	○	○	○
	DC / stepper motors	○	○	○	○	○	○	○	○	○	○
Fast z-piezo operation	With manual stage	○	○	○	○	○	○	○	○	○	○
	With scanning stage	○	○	○	○	○	○	○	○	○	○
Motorized 2TV tube	-	-	-	○	○	-	○	-	○	-	○
Condensers	Manual	○	○	○	○	○	○	○	○	○	○
	Motorized	-	-	○	○	-	○	-	○	-	○

+ = Included in stand

○ = Optionally available

○\* = Included only, if equipped with Light Control Mot. 430000-9304-000

- = Not possible

\* = Motorized (6-position and 10-position) reflector turret may be used.

\*\* = ACR function not available with Axio Imager D2 and D2m.

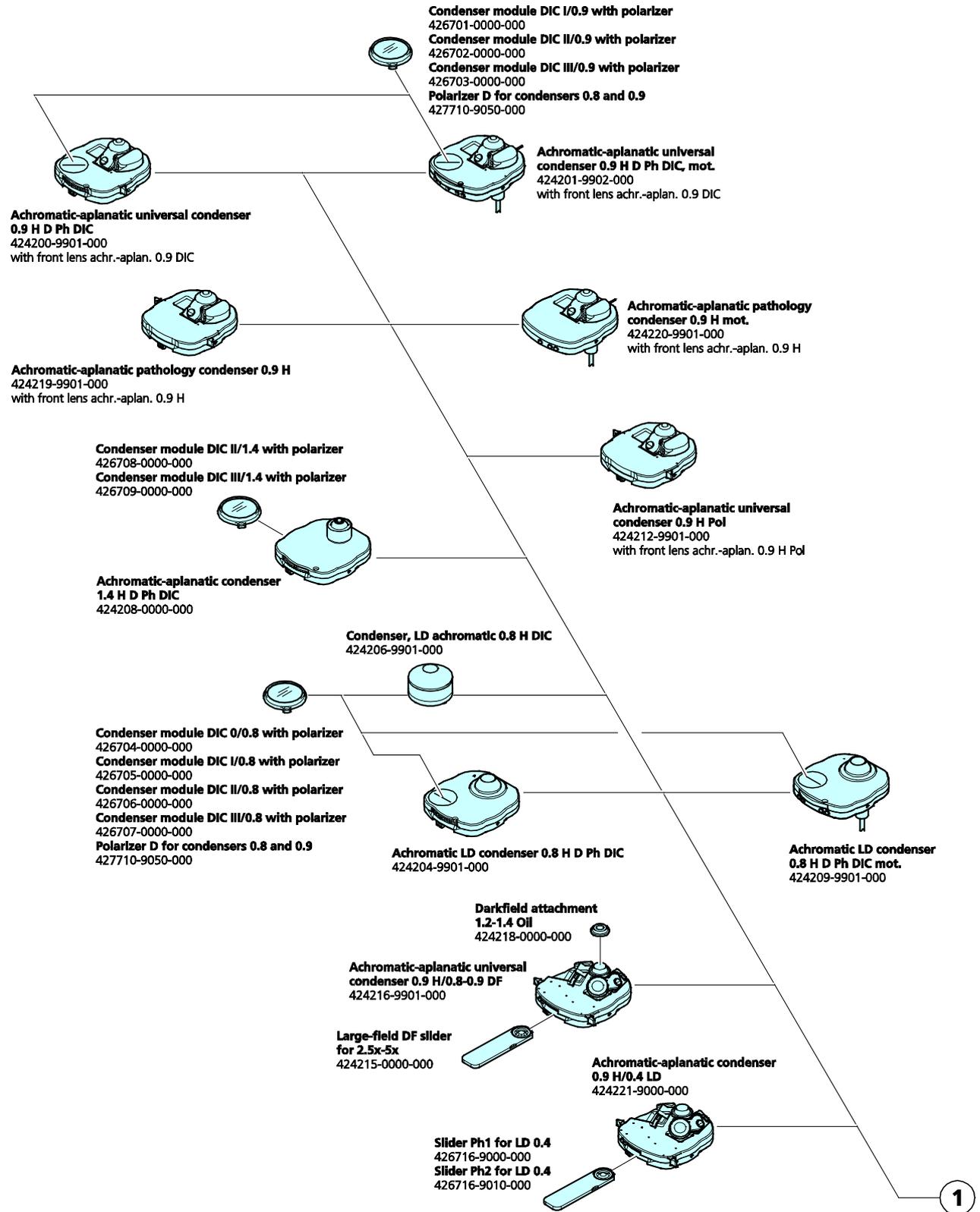
\*\*\* = All reflected-light illumination systems come with a motorized shutter.  
For fluorescence applications, it may be optionally replaced by a high-speed shutter.

\*\*\*\* = USB/CAN converter 432909 is required for use on Axio Imager A2 LED, A2, A2m, D2 and D2m.

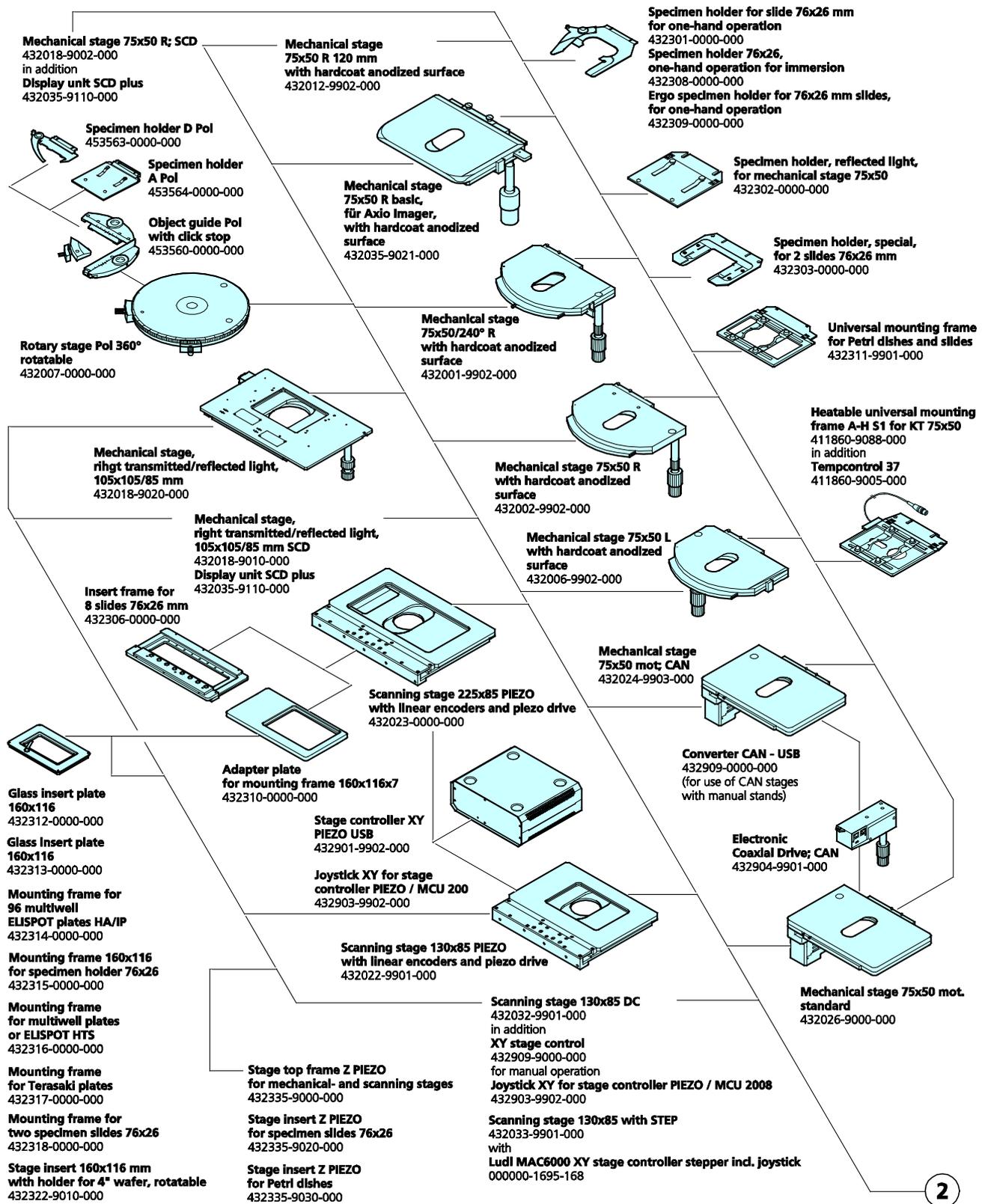
\*\*\*\*\* = Motorized only if used with an objective nosepiece.

2.4 System overview of Axio Imager

Condensers



## Microscope stages



## Objective nosepieces, reflector turrets, filters, prisms

**Reflector module brightfield ACR P&C for reflected light**  
424928-9901-000  
**Reflector module C-DIC/TIC ACR P&C for reflected light**  
424941-9000-000  
**Reflector module darkfield ACR P&C for reflected light**  
424922-9901-000  
**Reflector module Polarizer ACR P&C for reflected light**  
424923-9901-000  
**Reflector module Pol ACR P&C for HBO 100 for reflected light**  
424924-9901-000  
**Reflector module C-DIC/TIC ACR P&C for reflected light**  
424929-9903-000  
**Reflector module DIC/Pol red I ACR P&C for reflected light**  
424938-0000-000  
**Reflector module DIC/Pol ACR P&C for reflected light**  
424939-0000-000  
**Reflector module with analyzer ACR P&C for reflected light**  
424941-9050-000

**Analyzer module DIC ACR P&C shift free for transmitted light for combination with modulator turret**  
424921-9010-000  
**Analyzer module DIC ACR P&C for transmitted light**  
424921-9901-000  
**Analyzer module DIC ACR P&C shift free for transmitted light**  
424932-9901-000  
**Analyzer module Pol ACR P&C for transmitted light**  
424937-9901-000

**Modul Optovar 2.5x P&C (only for transmitted light)**  
424936-0000-000  
**Optovar module 1.6x P&C (only for transmitted light)**  
424935-0000-000  
**Optovar module 1.25x P&C (only for transmitted light)**  
424934-0000-000

**Reflector module FL EC P&C**  
424931-0000-000  
**Reflector module FL ACR P&C**  
424933-0000-000

**6-position reflector turret man. cod. for P&C modules**  
424906-0000-000

**6-position reflector turret mot. ACR, for P&C modules**  
424912-0000-000  
**6-position reflector turret mot. for P&C modules**  
424907-0000-000  
(included in stands M2m)

**10-position reflector turret mot. ACR, for P&C modules**  
424913-0000-000

**Filter sets for reflector modules FL on request**

**Reflector module C-DIC/TIC ACR P&C for reflected light**  
424929-9903-000  
**DIC prism C I for modulator turret for EC EPN 5x-20x**  
426921-0000-000  
**DIC prism C II for modulator turret for EC EPN 50x-100x**  
426922-0000-000  
**TIC prism for modulator turret for EC EPN 5x-100x**  
426923-9901-000

**4-position modulator revolver for circular DIC/TIC**  
424703-0000-000  
**4-position modulator revolver mot. for circular DIC/TIC**  
424704-0000-000

**DIC prism 0, I and II; 5x, 10x, 20x for modulator turret**  
426924-9000-000  
**DIC prism III 40x for modulator turret**  
426924-9010-000  
**DIC prism III HR PA 63x/1.4 for modulator turret**  
426924-9020-000  
**DIC prism III HC 63x/1.4 for modulator turret**  
426924-9030-000  
**DIC prism III HR PA 100x/1.46 for modulator turret**  
426924-9040-000  
**DIC prism III HC PA 100x/1.46 for modulator turret**  
426924-9050-000

for transmitted light:  
**Compensator mount 6x20**  
424705-0000-000

for reflected light:  
**Compensator mount 6x20 with darkfield stop**  
424706-0000-000

**4-position modulator turret mot. for transmitted-light DIC**  
424707-9000-000  
for use with  
**Analyzer module DIC ACR P&C shift free for transmitted light**  
424921-9010-000  
(only with Z2 and Z2m)

**Analyzer slider D, fixed**  
433605-0000-000  
**Compensator lambda, 6x20**  
473704-0000-000  
**Kompensator Lambda/4, 6x20**  
473714-0000-000

**Wedge compensator 0-4 Lambda, 6x20**  
000000-1140-633  
**Compensator lambda, rotary +/-8°, 6x20**  
473710-0000-000

**Tilting compensator K 0-30 Lambda, 6x20**  
000000-1115-698  
**Tilting compensator B 0-5 Lambda, 6x20**  
000000-1115-700

**Rotary compensator Brace-Köhler Lambda/10, 6x20**  
000000-1115-703

**Antiglare screen**  
452163-0000-000

**DIC slider C 6x20 for objectives EC 5x-20x**  
000000-1105-192  
**DIC slider C 6x20 for objectives EC 50x-100x**  
000000-1105-193  
**TIC slider 6x20**  
000000-1105-190  
(to be used with C DIC reflector module  
424941-9000-000)

**Objectives M 27 ICS objectives on request**

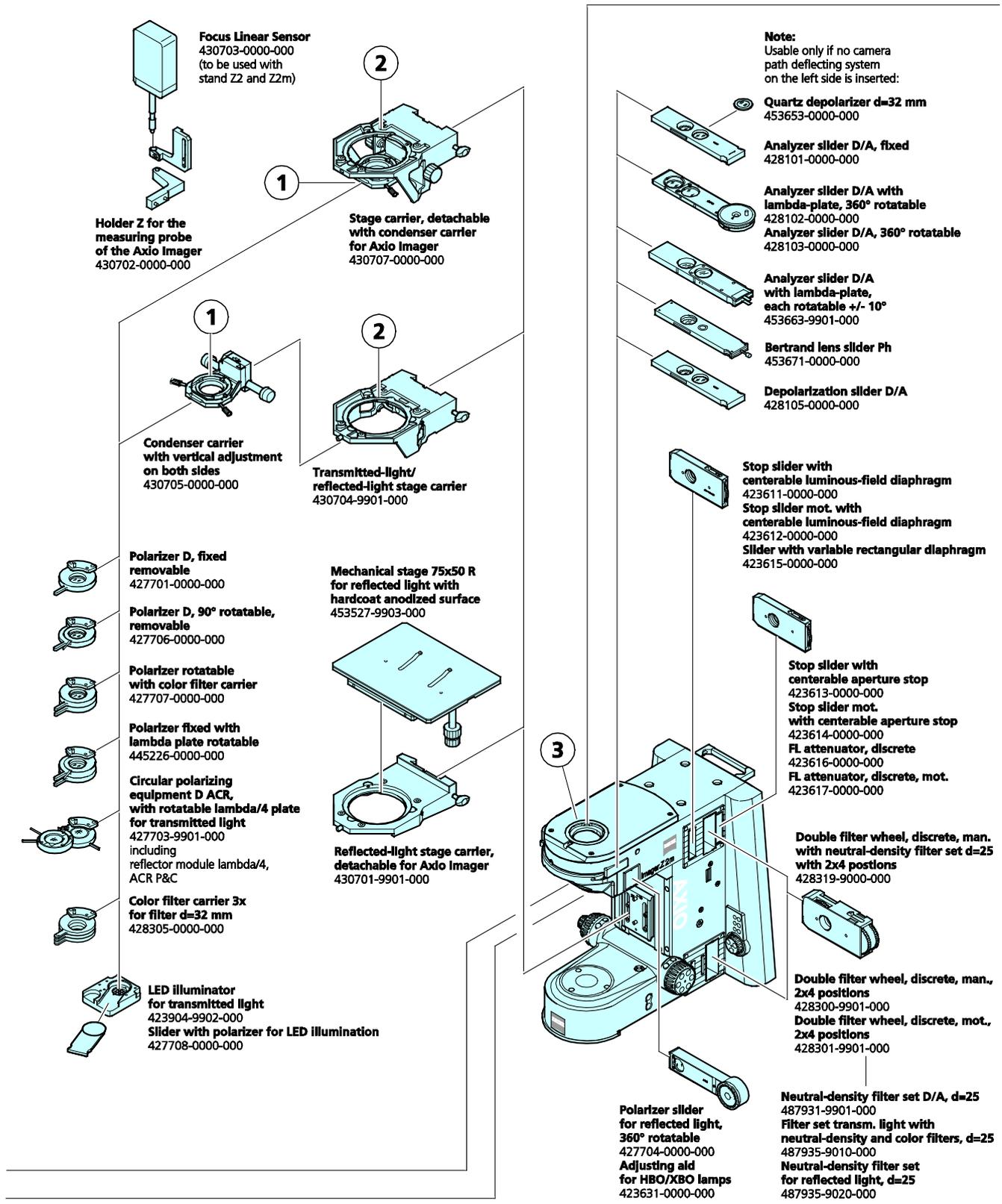
**DIC slider on request**

**6-position objective nosepiece, HD DIC M27 cod.**  
424504-0000-000  
**6-position objective nosepiece, HD DIC M27 mot.**  
424505-0000-000

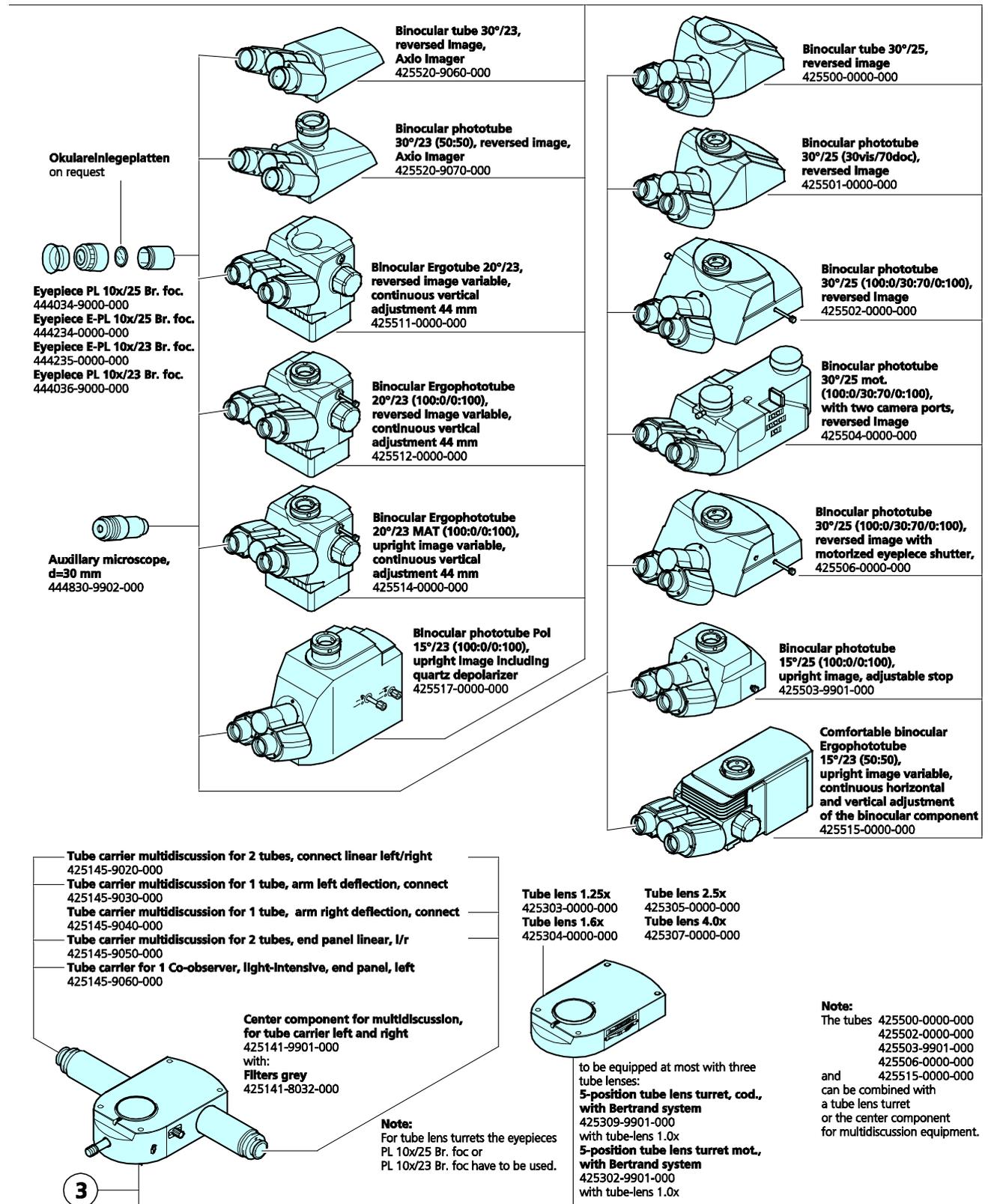
**7-position objective nosepiece, HD M27 cod.**  
424501-0000-000  
**7-position objective nosepiece, HD M27 mot.**  
424502-0000-000  
**6-position objective nosepiece, Pol M27 cod.**  
424503-0000-000  
**6-position objective nosepiece, HD DIC M27 mot. ACR**  
424507-0000-000

**Objective intermediate ring ACR for cylindrical, short objective sleeve**  
424508-0000-000  
**Objective intermediate ring ACR for cylindrical, long objective sleeve**  
424511-0000-000  
**Objective intermediate ring ACR for conic, short objective sleeve**  
424509-0000-000  
**Objective intermediate ring ACR for conic, long objective sleeve**  
424510-0000-000

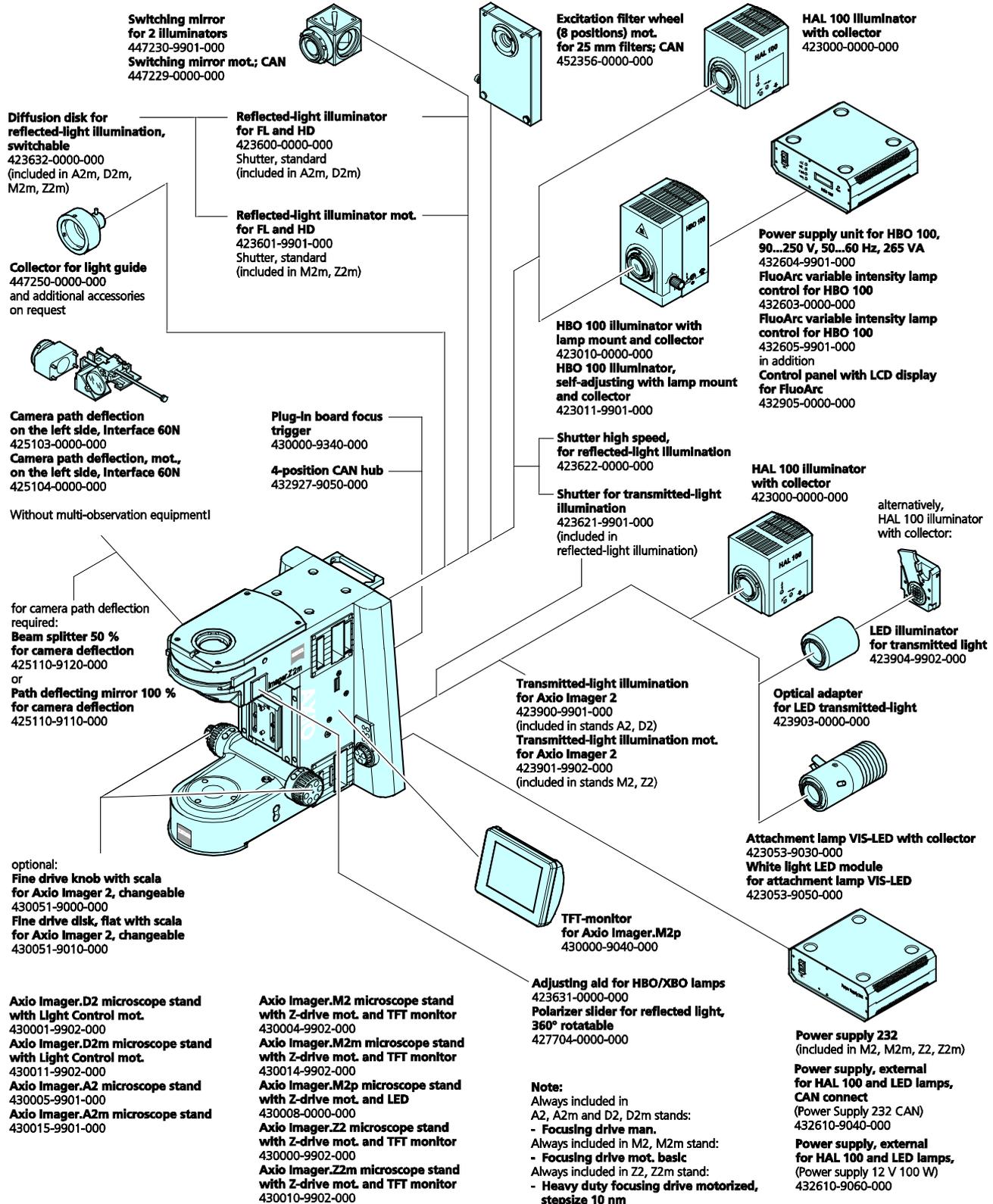
**Stand, stage carriers, polarizers, sliders**



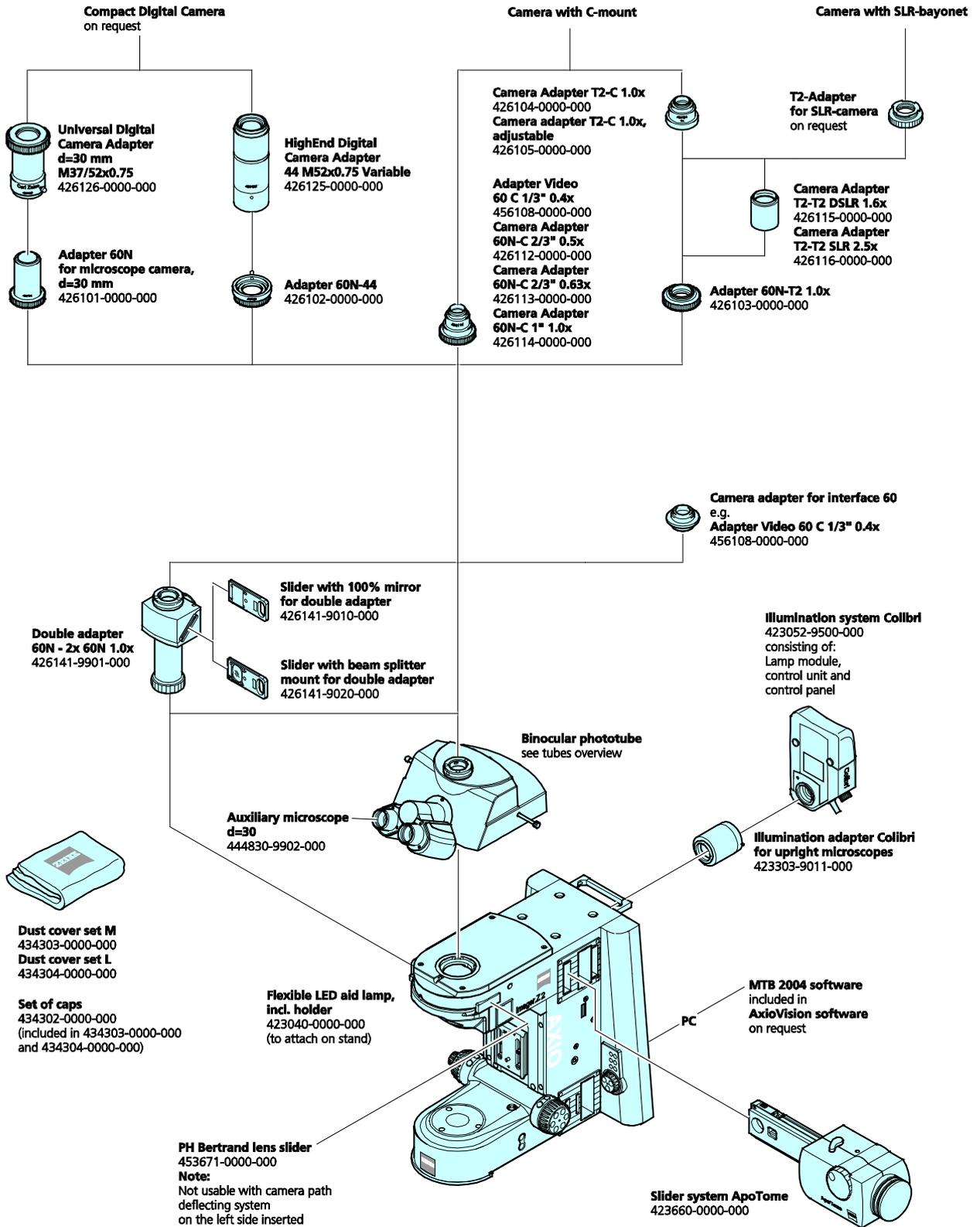
## Tubes, eyepieces, tube lens turrets, multidiscussion



## Stand, illumination



Documentation



## 2.5 Objectives

The objectives are the optical heart of the microscope. The following is an example of how objectives can be labeled:

N-ACHROPLAN 10x/0.25 ∞/0,17.

where

10x : Objective magnification, with a defined color ring on the objective being allocated to each magnification step (Zeiss color code)

0.25 : Numerical aperture

∞ : Infinite image distance: These objectives can only be used with ICS microscopes from Carl Zeiss.

0.17 : Can only be used with cover slip thickness  $D = 0.17$  mm.

or

– : Can be used with cover slip thickness  $D = 0$  or  $0.17$  mm.

Other labels:

Oil : Oil immersion objective

Ph 2 : Phase-contrast objective with green inscription and phase stop Ph 2

The color of the inscription denotes the contrasting method the objective is designed for:

Black: Standard

Green: Phase contrast

Red: Strain-free for polarization (Pol); low-strain for differential interference contrast (DIC)

The color rings indicate the magnification of the objective (color code):

Color ring on objective	Black	Brown	Red	Orange	Yellow	Green	Light blue	Dark blue	White
Magnification factor	1x; 1.25x	2.5x	4x; 5x	6.3x	10x	16x; 20x; 25x; 32x	40x; 50x	63x	100x; 150x

Objective magnification multiplied by eyepiece magnification (usually 10x) results in overall visual magnification: e.g.  $10 \times 10 = 100x$ .

When working with the microscopes, total magnification should not drop below or exceed the range of useful magnification. The range of useful magnification was defined by Ernst ABBE as 500 to 1,000 times the numerical aperture of the objective used. There is no resolution for further details above that limit. Accordingly, the range of useful magnification for an objective with a numerical aperture of 0.3 is between 150x and 300x.

Exact observance of the cover slip thickness of 0.17 mm is all the more necessary the higher the numerical aperture of the objective. For this reason, certain objectives are equipped with a correction mount providing adjustment to different cover slip thicknesses. To this end, a specimen area is searched for, and the position of the correction ring where optimum focus and image contrast are obtained is determined (refocusing is invariably required).

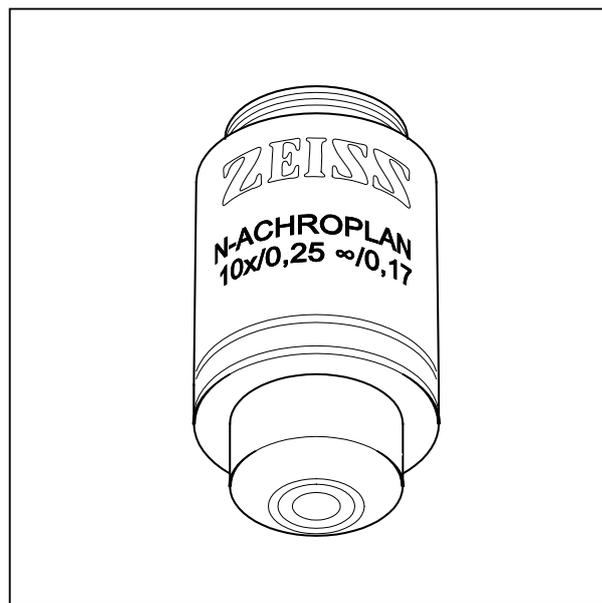


Fig. 2-1 Objective

When immersion objectives are used, the air between the cover slip and the objective is replaced by a liquid, which in most cases is immersion oil. The plastic oiler containing 20 ml of Immersol 581 F<sup>®</sup> immersion oil ( $n_D = 1.518$ ) is particularly suitable for this purpose.

To prevent oil contamination of the specimen when the nosepiece is turned, the resilient mounts of the immersion objectives can be locked in their lifted position by turning them clockwise (do not forget to unlock them again!).

## 2.6 Eyepieces

The field-of-view number of the eyepieces PL 10x/25 Br. foc. and E-PL 10x/25 Br. foc. is 25 mm; that of eyepieces W-PL 10x/23 Br. foc. and E-PL 10x/23 Br. foc. is 23 mm.

W-PL and PL in the eyepiece designation refers to the excellent image flatness up to the edge of the field of view.

If required, eyecups for the eyepieces can be ordered under Cat. No. 444801-0000-000.

## 2.7 Stage micrometers and eyepiece reticles

Measuring and counting using a microscope requires stage micrometers and eyepiece reticles, a selection of which is listed below:

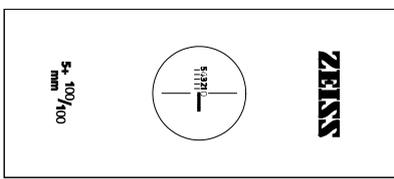
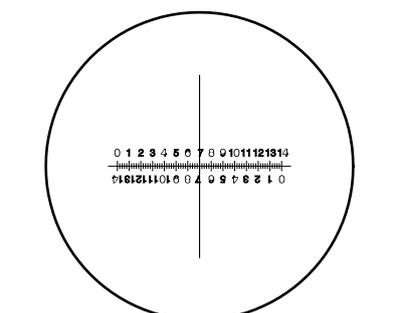
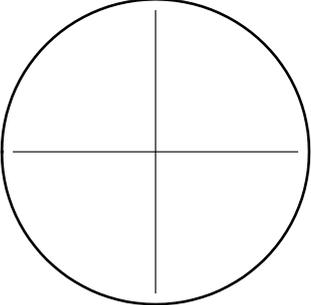
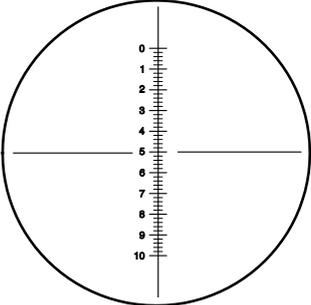
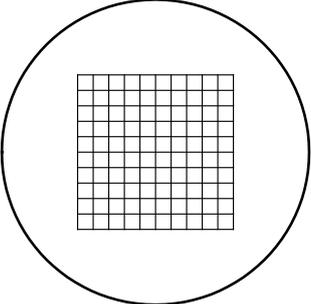
Illustration	Designation, technical data	Cat. No.
	Stage micrometer, positive 5 + 100/100 y <b>D = 0.17 mm</b> Graduation on +y-axis: 5 mm in 5 intervals Graduation on -y-axis: 1 mm in 100 intervals with 2 opposing scales = 10 $\mu$ m, accuracy $\pm 1 \mu$ m	474026-0000-000
	<b>Crossline micrometer 14:140/d = 26 mm</b> Graduation length = 14 mm Increments = 0.1 mm Graduation tolerance $\leq 0.001$ mm	454060-0000-000

Illustration	Designation, technical data	Cat. No.
	<p><b>Eyepiece reticle /d = 26 mm</b> For the alignment of the reticle by means of alignment specimen.</p>	474064-0000-000
	<p><b>Crossline micrometer 10:100/d = 26 mm</b> Graduation length = 10 mm Increments = 0.1mm Graduation tolerance <math>\leq 0.001</math> mm</p>	474066-9901-000
	<p><b>Net micrometer 12.5x12.5/5;10/d = 26 mm</b> Area 12.5x12.5 mm, divided into 10x10 fields</p>	474068-0000-000

 If an eyepiece reticle is used, the binocular tube or the phototube must be equipped with two focusing eyepieces ("foc."), into one of which the eyepiece reticle is to be mounted.

**2.8 Technical data****Dimensions (width x depth x height)**

Axio Imager stand, manual with HBO 100 .....	approx. 300 mm x 721 mm x 505 mm
Axio Imager stand, motorized with HBO 100 and TFT display .....	approx. 390 mm x 721 mm x 505 mm

**Weight**

Axio Imager, manual/motorized (dependent on equipment) .....	approx. 18 to 40 kg
--	---------------------

**Ambient conditions****Transport (in packaging):**

Permissible ambient temperature .....	-40 to +70 °C
---------------------------------------	---------------

**Storage:**

Permissible ambient temperature .....	+10 to +40°C
Permissible relative humidity (no condensation) .....	max. 75% at 35 °C

**Operation:**

Permissible ambient temperature .....	+10 to +40 °C
Permissible relative humidity .....	max. 75% at 35 °C
Atmospheric pressure .....	800 hPa to 1060 hPa
Altitude .....	max. 2000 m
Pollution degree .....	2

**Operating data for coded Axio Imager, equipped with an integrated power supply or motorized Axio Imager using the VP232-2 external power supply**

Operating environment.....	Closed room
Protection Class.....	I
Protection Type .....	IP 20
Electrical safety.....	in compliance with DIN EN 61010-1 (IEC 61010-1) including CSA and UL directives
Overvoltage category .....	II
Radio interference suppression.....	in accordance with EN 55011 Class B
Noise immunity .....	in accordance with DIN EN 61326 /A1
Line voltage for integrated power supply .....	100 to 127 and 200 to 240 V ±10 % Change of line voltage setting is not required!
Line voltage for external power supply VP232-2 .....	100 to 240 V ±10 %
Line frequency.....	50/60 Hz
Power consumption of coded Axio Imager .....	max. 260 VA
Power consumption of Axio Imager, motorized .....	max. 190 VA
LED illuminator .....	400 to 700 nm, peak at 460 nm
Attachment lamp VIS-LED .....	400 to 700 nm, peak at 460 nm

**Transformer HBO 100**

Operating environment .....	Closed room
Protection Class.....	I
Protection Type .....	IP 20
Line voltage .....	100 VAC ... 240 VAC
Line frequency.....	50/60 Hz
Power consumption when HBO 100 is used .....	155 VA

**Fuses in accordance with IEC 127**

Axio Imager microscope stand, manual .....	T 5.0 A/H / 250V, 5x20 mm
Power supply VP232-2 for Axio Imager, mot. ....	T 4.0 A / 250V, 5x20 mm
Transformer HBO 100.....	T 2.0 A/H, 5x20 mm

**Light sources**

Halogen lamp .....	12 V/100 W
Adjustment of light source .....	continuous, approx. 0.7 to 12 V
Mercury vapor short-arc lamp .....	HBO 103 W/2
Power consumption of HBO 103 W/2 .....	100 W

**Axio Imager, coded**

Stand with manual stage focusing	
Coarse drive .....	2 mm/revolution
Fine drive .....	0.2 mm/revolution; 2 µm scale interval
Lifting range .....	max. 25 mm
Height stop .....	mechanically adjustable
Achromatic-aplanatic universal condenser 0.9 H D Ph DIC with swivel-type front lens, achromatic-aplanatic 0.9 DIC, for objective magnifications $V_{obj.} < 10x$ .....	front lens 0.9 swiveled out
for objective magnifications $V_{obj.} \geq 10x$ .....	front lens 0.9 swiveled in
8-position turret disc	
Objective change:	
Manually .....	via 6-position or 7-position nosepiece, HD or HD DIC M27
Change of method modules	
Manually .....	via 6-position reflector turret

**Axio Imager, motorized**

Stand with motorized stage focusing:

Mean step size of stepper motor .....	25 nm (Axio Imager.M2)
	10 nm ±10 (Axio Imager.Z2)
Quick lowering/lifting of stage .....	10 mm
Lifting range.....	25 mm
Height stop .....	electronic
Focusing speed.....	variable

Achromatic-aplanatic universal condenser 0.9 H D Ph DIC, mot. with

swivel-type front lens, achromatic-aplanatic 0.9 DIC,

for objective magnifications  $V_{obj.} < 10x$ ..... front lens 0.9 swiveled outfor objective magnifications  $V_{obj.} \geq 10x$ .....front lens 0.9 swiveled in

8-position turret disc

Objective change:

Manually or motorized..... via 6-position or 7-position nosepiece

Change of method modules

Manually .....

Motorized .....

Manually/motorized.....

High-performance focus for scanning stages ..... Applicable for specimens weighing up to 5 kg

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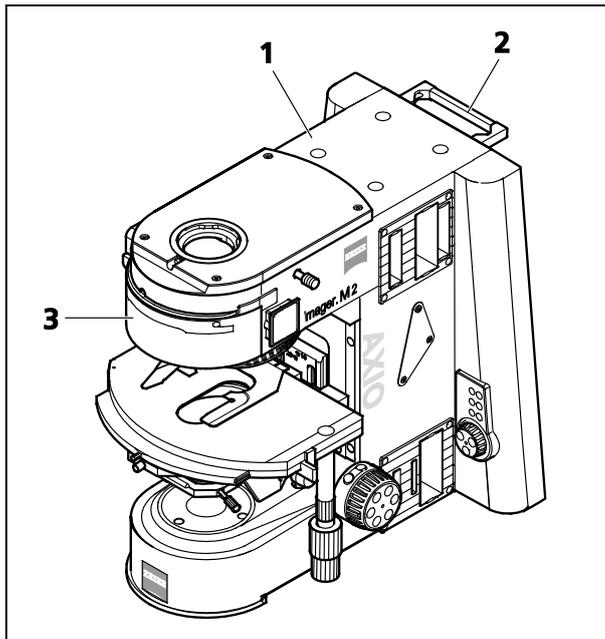
430000-7344-001

31

### 3 START-UP

The Axio Imager microscope can be installed, converted and started up by the customer. On request, the microscope is also installed or converted by Zeiss Service with costs.

 Before installing and starting-up the microscope, be sure to carefully read the **notes on instrument safety** (see Section 1).



**Fig. 3-1** Setting up the microscope

#### 3.1 Unpacking and installing the microscope

The basic instrument is supplied packed to commercial standards in a polyethylene case with cardboard packaging.

It contains the following components:

Stand, binocular tube, objectives, eyepieces, condenser, HAL 100 illuminator, microscope mat and various small parts, such as DIC slider, spare lamp, dust cover, tool bag with tools.

The following components are factory-installed to the microscope stand: Mechanical stage, specimen holder, objectives, polarizer, filter holder, reflected-light illumination and adjusting aid for HBO/XBO lamps.

Additional optional accessories are supplied in a separate box.

- Remove all components from the packaging observing the provided instructions for unpacking the instrument.

 You can carry the stand using the handle (3-1/2) on the rear side of the stand. The second point of engagement must never be located on the microscope stage, as the guide may get damaged otherwise. The grip recess on the cover part 3-1/3) constitutes a safe second point of engagement.

- Use the delivery form to check the delivered items for completeness.
- Place the instrument (3-1/1) onto a vibration-free, flat worktable.
- Keep the original packaging for storage or for returning the instrument to the manufacturer, or dispose of it properly.

### 3.2 Attaching or changing binocular tube or phototube

All binocular tubes listed in the system overview (refer to Section 2.4) can be attached to the manual or the motorized stand as described below.

- Use an SW 3 ball-headed screwdriver to undo clamping screw (3-2/4) and remove the tube (3-2/2) upwards.
- Place dust cap (3-2/3) for tube lens protection on the dovetail of the binocular tube.
- Remove dust cap (3-2/6) from the desired tube.
- Insert tube (3-2/1) with the dovetail into the stand opening (3-2/5) and align the tube.
- Tighten clamping screw (3-2/4).

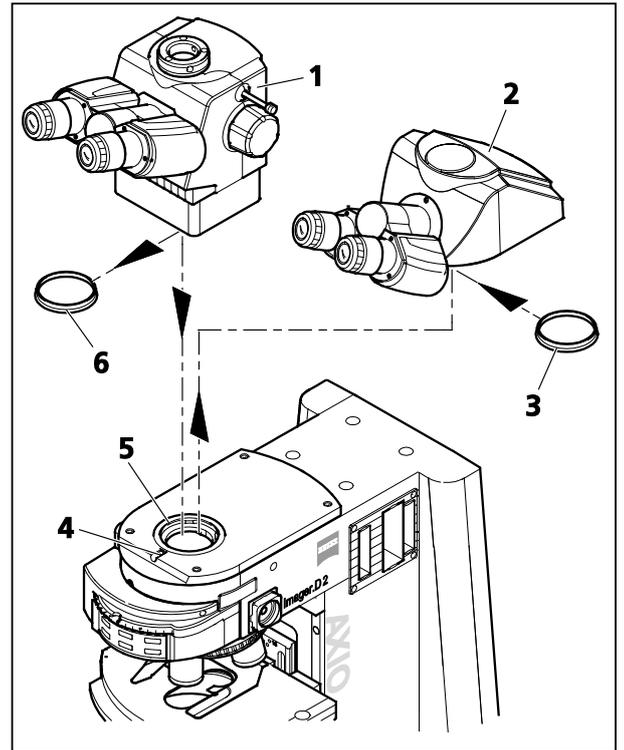


Fig. 3-2 Changing the binocular tube



#### CAUTION

Do not hold the tube by the two eyepiece supports when carrying it.

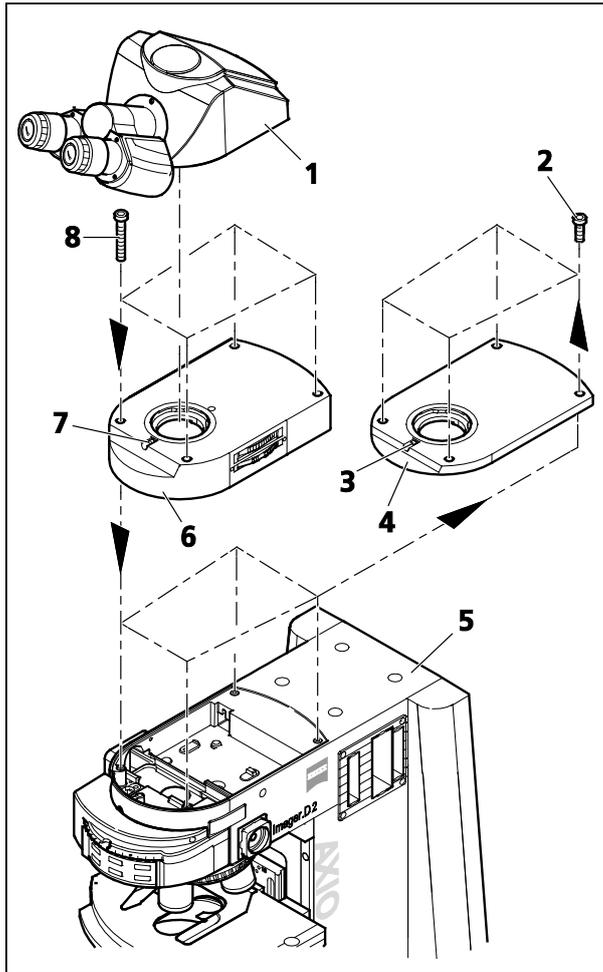


Fig. 3-3 Mounting the tube lens turret

### 3.3 Mounting the tube lens turret

- Use SW 3 ball-headed screwdriver to loosen clamping screw (3-3/3) and remove the tube (3-3/1) upward.
- Unscrew four fastening screws (3-3/2), remove coupler plate (3-3/4) upward and store it for any future use.
- Put tube lens turret (3-3/6) onto stand (3-3/5) and screw it down using the four supplied fastening screws (3-3/8).
- Unscrew the tube lens by hand from the tube to be used (3-3/1). Store it in the storage box.
- Insert tube (3-3/1) without tube lens with its dovetail into the mount of the tube lens turret (3-3/6) and tighten clamping screw (3-3/7).

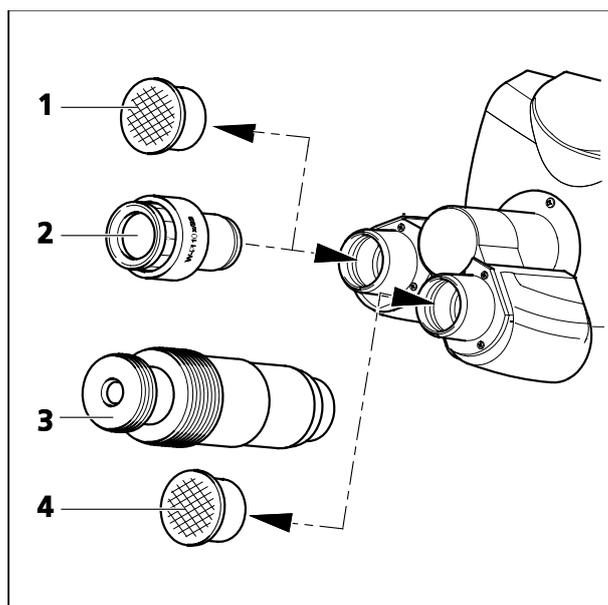


Fig. 3-4 Inserting eyepieces

### 3.4 Inserting eyepieces and auxiliary microscope

- Remove both dust caps (3-4/1 and 4) from the binocular tube.
- Remove both eyepieces (3-4/2) from their cases and insert them into the binocular tube as far as they will go.
- The auxiliary microscope (3-4/3) can be inserted into one of the eyepiece sockets of the binocular tube in place of an eyepiece. It is used to view aperture diaphragm, phase and dark-field stops and to center phase and dark-field stops. Focusing on these diaphragms and stops is possible by means of the adjustable eye lens, which can then be locked by means of the clamp screw.

### 3.4.1 Inserting the eyepiece reticle

Eyepiece reticles (3-5/3) may be inserted into eyepieces marked with a red dot.

The slight image shift caused by the additional path through glass is taken into account on the diopter scale by the fact that the zero point position is indicated not by the white dot, but the red dot.

Make sure the line figure always faces the field stop.



Eyepiece reticles may only be installed by Zeiss service technicians working in a dust-free environment.

### 3.4.2 Compensation of ametropia when eyepiece reticles are used

Correct use of an eyepiece reticle requires two focusing eyepieces, e.g. PL 10x/23 Br. foc, to enable the user to compensate for differences in the visual acuity of his or her eyes.

- Use the focusing lens of the adjustable eyepiece to focus on the line figure of the eyepiece reticle.
- Viewing through the eyepiece, use the focusing drive to focus on the microscope image of a specimen placed on the stage.
- As soon as the microscope image and the eyepiece reticle are focused in the above eyepiece, turn the focusing eye lens of the second eyepiece to focus the microscope image for the second eye.

On having done this, both microscopic images and that of the eyepiece reticle are focused.

From this point in time on, you should focus only with the focusing drive.

### 3.4.3 Inserting the fold-over eyecups

The eyepieces have a rubber eyeglass protection ring to prevent scratches on spectacles. The protection rings can be replaced with fold-over eyecups when required.

- Remove the eyeglass protection rings (3-5/2) from the eyepieces and attach the eyecups (3-5/1).

Sometimes the eyeglass protection rings are seated very tightly in the eyepiece groove, so you may need a blunt object (stick) to prod them off.

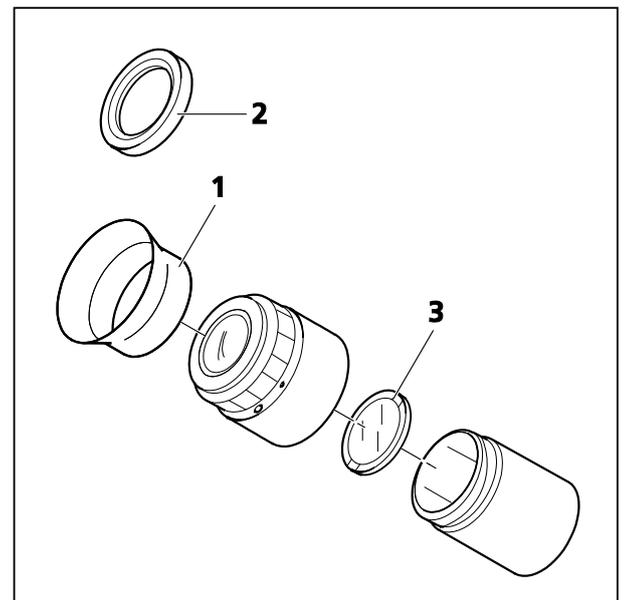
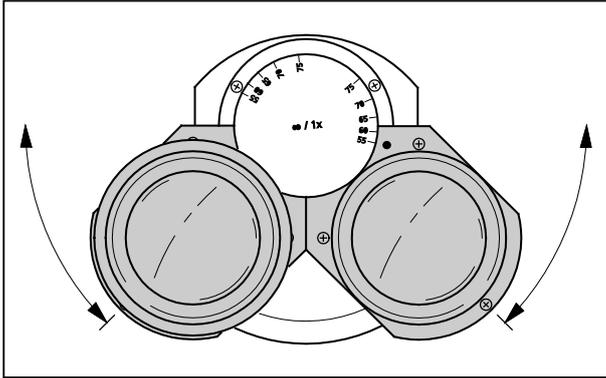


Fig. 3-5 Inserting the fold-over eyecups

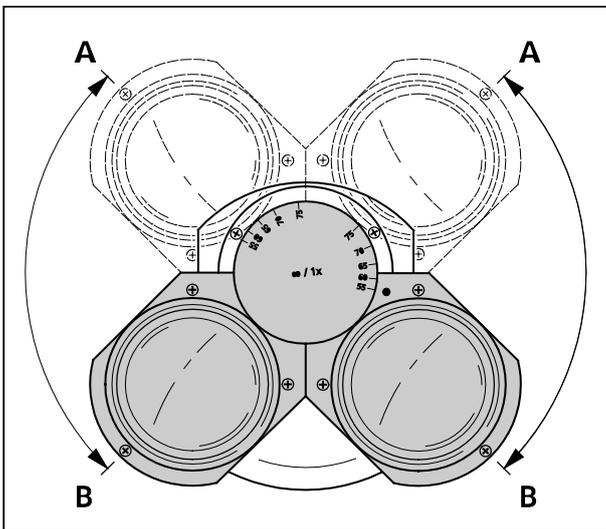


**Fig. 3-6** Setting the interpupillary distance on the binocular tube

### 3.5 Setting the interpupillary distance on the binocular tube

- To adjust the eyepiece distance to your individual interpupillary distance, swing the eyepiece tubes symmetrically toward or away from one another (Fig. 3-6).

The adjustment of the interpupillary distance is correct when you see only **one** round image while looking through the two eyepieces!



**Fig. 3-7** Setting the viewing height on the binocular tube

### 3.6 Setting the viewing height

- The viewing height can be adjusted to individual requirements by swiveling the eyepiece tubes up (3-7/A) or down (3-7/B).

The ergonomic binocular tubes (425511-0000-000, 425512-0000-000 und 425515-0000-000) provide continuous height adjustment over a range of 50 mm. Adjustment is by means of the rotating knob.

### 3.7 Fitting components to the camera port of the binocular phototube

#### Adapter for Interface 60N (external thread M52 x 1)

 The Axio Imager uses a new connector type "Interface 60N" to adapt the camera. The known adapters for "Interface 60" (inside diameter 30 mm), however, can also be used further on.

Microscope cameras (e.g. AxioCam of Carl Zeiss), customary SLR cameras (Single Lens Reflex; 35 mm film or digital) or compact digital cameras may be mounted to the camera port.

 For your work with photomicrographic devices, please pay attention to the corresponding manuals of the cameras as well.

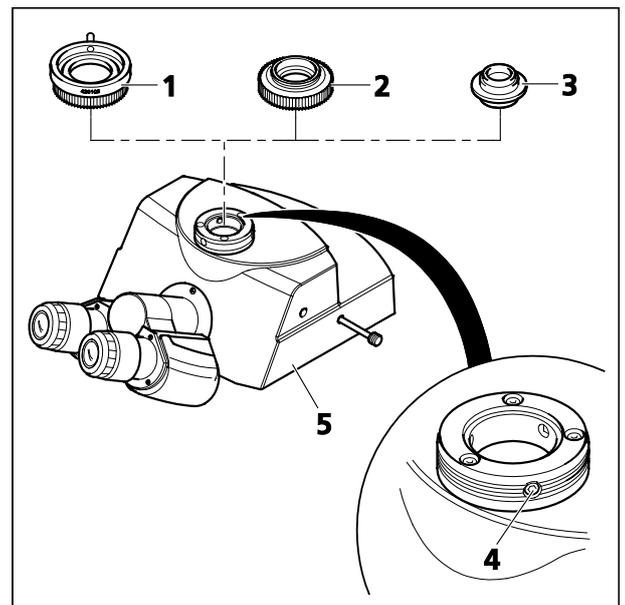
- Fix the camera adapter 60N (3-8/1; **2**) to the camera.
- Remove the dust cap from the camera port.

 Please observe the following: The three set screws (SW 3) (3-8/4) at the camera port must jut out neither to the external thread nor to the internal bore hole.

- Attach the pre-assembled unit to the camera port, adjust it and fasten the union nut of the adapter (3-8/1 or **2**) fingertight.

#### Adapter for Interface 60 (plug-in diameter 30 mm)

- Fix the camera adapter 60 (3-8/3) to the camera.
- Remove the dust cap from the camera port.
- Insert the pre-assembled unit in the camera port (do not screw in set screws too deeply).
- Turn clockwise three set screws (SW 3) at the tube (3-8/4) to fasten adapter.



**Fig. 3-8 Fitting components to the phototube**

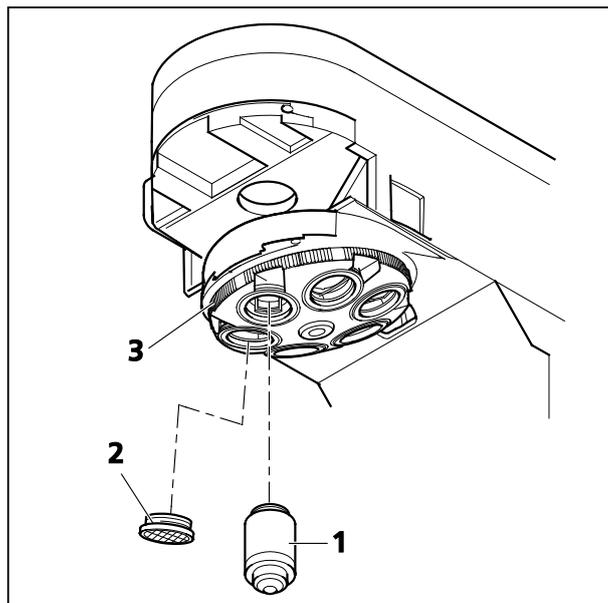


Fig. 3-9 Screwing in objectives

### 3.8 Screwing in objectives

- Move the mechanical stage with stage carrier to the lower stop position.
- Remove dust caps (3-9/2) from the respective openings on the objective nosepiece.
- Remove objectives (3-9/1) from the case and screw them into the nosepiece (3-9/3) starting with the lowest magnification (clockwise rotation).
- Make sure to cover objective mounts of the nosepiece not in use by dust protection caps.



In combination with objectives with W 0.8" thread please use adapter M27x0.75 "0" on W 0.8 (000000-1095-168).

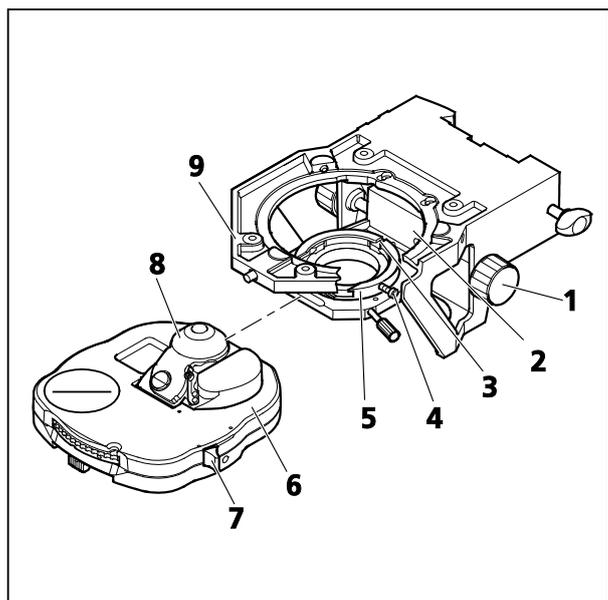


Fig. 3-10 Attaching the achromatic-aplanatic universal condenser

### 3.9 Attaching or changing the condenser

- Turn coarse focusing drive to move the stage carrier to the upper stop position.
- Turn height control (3-10/1) to move condenser carrier (3-10/2) down.
- Slightly unscrew locking screw (3-10/4) (SW 1.5), if necessary.
- If available, swivel out front lens (3-10/8) of condenser by means of lever (3-10/7).
- Directing the locking screw on the bottom of the condenser to the groove (3-10/3), insert the condenser (3-10/6) between condenser carrier (3-10/2) and stage carrier (3-10/9) in the centerable condenser mount (3-10/5) as far as it will go.
- Tighten locking screw (3-10/4) on condenser mount. In doing so, do not apply excessive force to avoid any damage to the condenser mount.

- When using a motorized condenser (3-11/1) (on the motorized stand) thread the connecting cable (3-11/3) through the opening in the stage carrier to the back and plug the plug into socket (3-11/2) on the right of the stand base.

Removal of the condenser is in reverse order.



When using other types of condensers, proceed analogously.

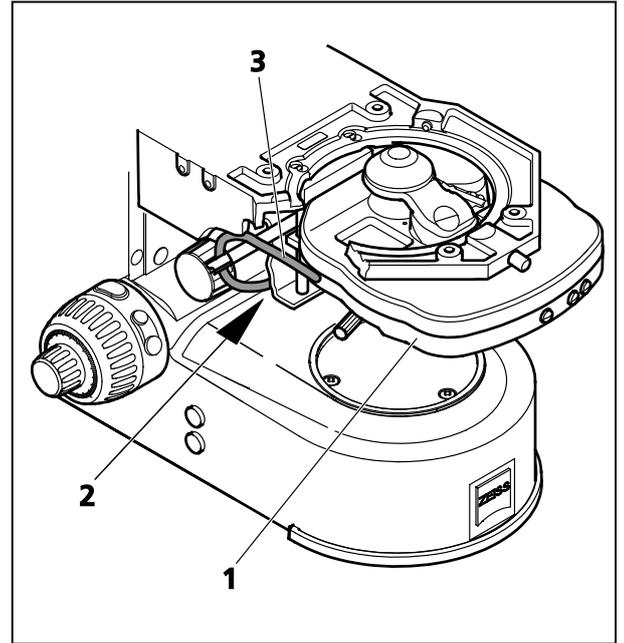


Fig. 3-11 Attaching the achromatic-aplanatic universal condenser, mot.

### 3.10 Replacing the DIC prism on the universal condenser



#### CAUTION

Before replacing the prism on the motorized universal condenser, make sure to open the aperture iris first with the corresponding control button to prevent damage of the iris blades.

- Use the tool to remove cover (3-12/1) or unscrew it (if cover is at the bottom). Turn turret disk until the DIC prism to be replaced appears in the blank aperture (3-12/4). Hold the disk at its knurled ring.
- Unscrew retaining ring by means of the so-called dual-function tool (3-12/2).
- Then, screw the tool (3-12/2) with the threaded side into the prism mount (3-12/3) and pull out the DIC prism (3-12/6).
- Remove DIC prism (3-12/6) from the tool and screw on the new, desired DIC prism.
- Mount the DIC prism in reverse order paying particular attention to the correct orientation of the DIC prism: Tongue 3-12/5 must be in the recess of the mount in the condenser. Take care that the labeling on the knurled ring of the turret disk is correct.

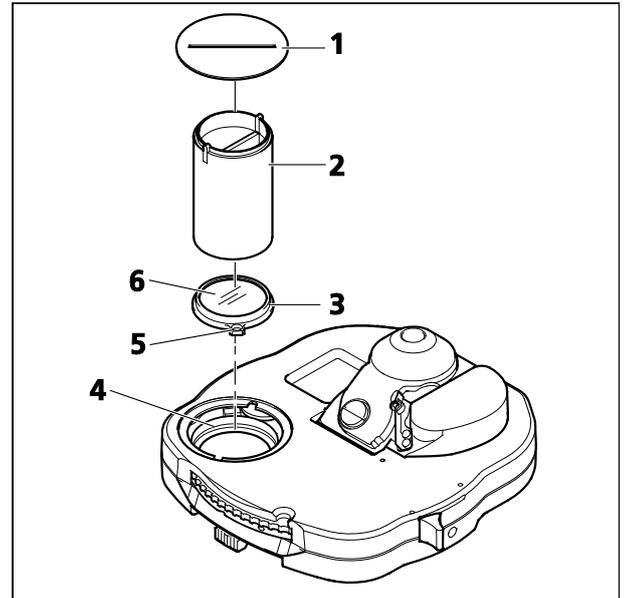
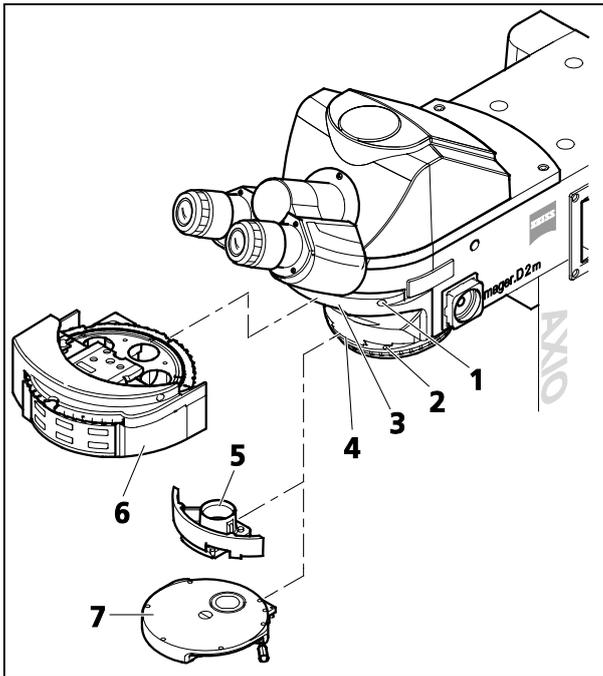


Fig. 3-12 Replacing the DIC prism



**Fig. 3-13** Inserting reflector turret, compensator mount or 4-position modulator

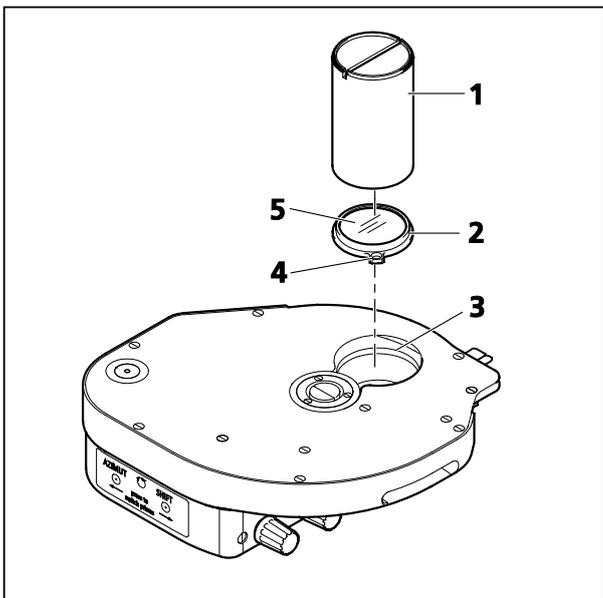
 If the instrument is to be transported to another location, the reflector turret must be replaced by the cover part provided with a grip recess.

### 3.11 Inserting reflector turret, compensator mount 6x20 or 4-position modulator turret



Install or change the motorized reflector turret or modulator turret only when the microscope is switched off.

- Loosen the locking screw (3-13/1). Pull out the reflector turret (3-13/6) or the cover part, using the grip recess (condition as delivered to customer).
- Insert compensator mount (3-13/5) or the 4-position modulator turret (3-13/7) manual or motorized) into guide (3-13/4) on the top part of the stand as far as it will go. Tighten the clamping screw (3-13/2).
- Attach reflector turret (3-13/6) to the guide (3-13/3) in the top part of the stand and push it in until it reliably (audibly) snaps in.
- Tighten clamping screw (3-13/1).
- If the reflector turret was supplied without inserted reflector modules, insert them as described in Section 3.19.



**Fig. 3-14** DIC-Prisma im Modulatorrevolver wechseln

### 3.12 Changing the DIC prism of the modulator turret

- Pull the modulator turret frontward out of the stand and set it down.
- Manually turn the turret disk until the desired position is accessible in the opening (3-14/3) from the top.
- Screw out the retaining ring, using the dual-function tool (3-14/1).
- Now, screw the threaded side of the tool (3-14/1) into the prism mount (3-14/2) and then pull the DIC prism (3-14/5) out.
- Remove the DIC prism from the tool and screw on the new DIC prism desired.
- Proceed in reverse order to install the DIC prism. In so doing, make sure that the DIC prism is correctly oriented. (Tongue 3-14/4 must be located in the recess of the mount provided in the modulator turret.) Make sure the labelling on the knurled ring of the turret disk is correct.

### 3.13 Changing the stage carrier

- To remove the stage carrier (3-15/3), grip it firmly and loosen the clamping lever (3-15/2) until the stage carrier can be taken out of the guide from the right to the left.
- Check the position of the stop screws provided for the adjustment of the dovetail guide and alter it, as required for the stage carrier or microscope stage used. (See Section 3.14.)
- To attach the stage carrier, insert it on the left into the guide and push it horizontally against the supporting surface of the guide and vertically against the upper Stop bolt (3-15/1).
- Tighten clamp screw (3-15/2) well and verify that the stage carrier is exactly seated in the guide.

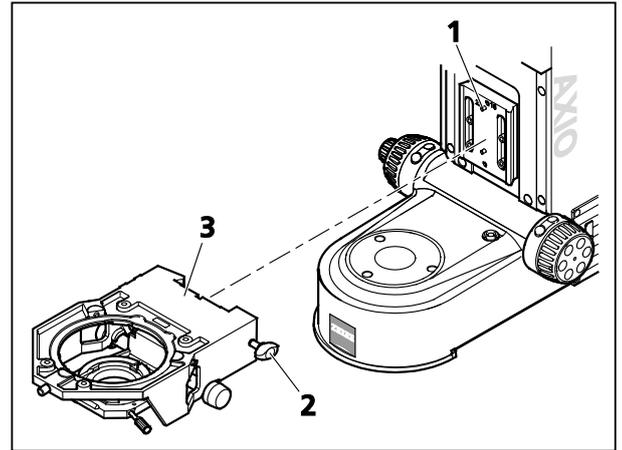


Fig. 3-15 Changing the stage carrier

### 3.14 Setting stops for the adjustment range of the stage carrier dovetail guide

Lower stop screw (3-16/5):

- When employing the reflected-light/ transmitted-light stage carrier with condenser carrier 430707-0000-000, use a slot-head screwdriver to screw the lower stop screw into the upper tapped hole (3-16/3).
- When employing the reflected-light or reflected-light/transmitted-light stage carrier 430704-9901-000 or 430701-9901-000, screw the lower stop screw into the lower tapped hole (3-16/4).

Upper stop screw (3-16/6):

- When an 18-mm thick microscope stage is used, screw the upper stop screw into the tapped hole marked with number "18" (3-16/1).
- When a 20-mm thick microscope stage is used, screw the upper stop screw into the tapped hole marked with number "20" (3-16/2).

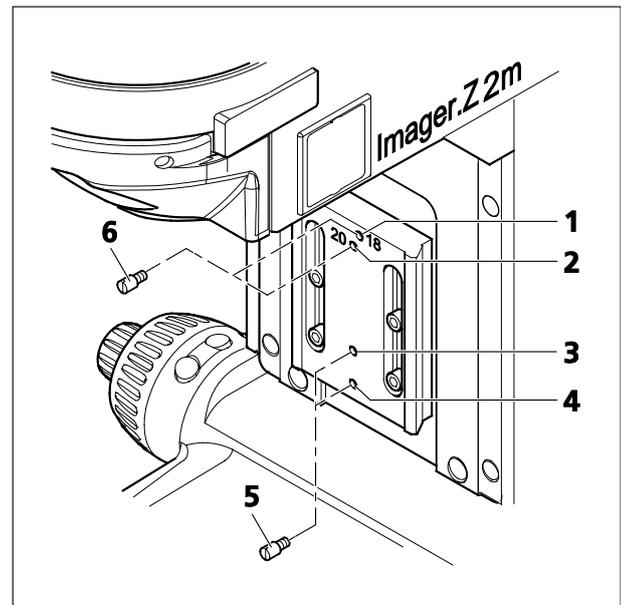
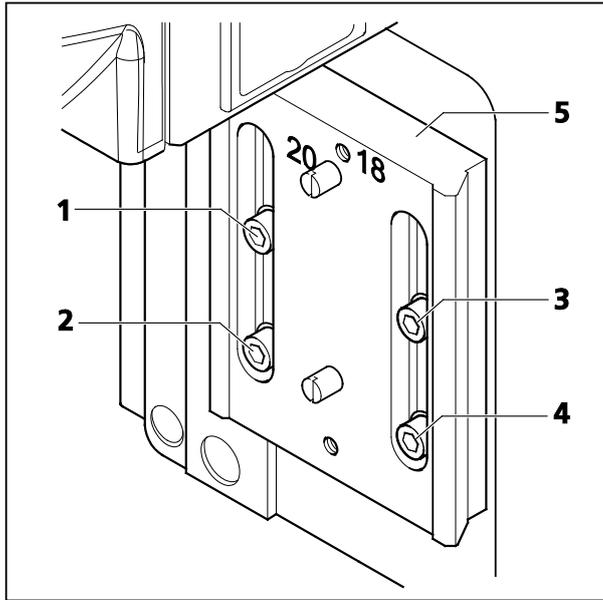


Fig. 3-16 Setting dovetail guide stops



The thickness of the stage is shown right next to the order number on a sticker attached to the microscope stage. Should the sticker be missing on the microscope stage, the stop screw is to be set to 18 mm to maintain focusability.



**Fig. 3-17** Setting the dovetail guide up for specimen space expansion

### 3.15 Setting the dovetail guide of the stage carrier

When stage carrier 430701-9901-000 is used, the specimen space needs to be shifted for specimens of less than 13 mm, so that the specimen can be focused.

To do so, proceed as follows:

- Remove the stage carrier (see Section 3.13).
- **Loosen** the four locking screws (3-17/1 to 4) of the dovetail guide.
- Move the guide plate (3-17/5) as much **upward** in the slots as it will go.
- Then, tighten the four screws. **Always** proceed in the following order:
  - upper left screw (3-17/1),
  - lower left screw (3-17/2),
  - upper right screw (3-17/3),
  - lower right screw (3-17/4),
- Attach the stage carrier (see Section 3.13).

 If stage carrier 430701-9901-000 is no longer used, the guide plate must be set back to the **lower** position (normal position).

### 3.16 Attaching the condenser carrier to the transmitted-light/reflected-light stage carrier

- Use the drive knob (3-18/2) to adjust the guide of the condenser carrier (3-18/3) in such a way that the two screws (3-18/4) are accessible.
- Attach the condenser carrier to the stage carrier (3-18/1). Push it to the upper stop in a firm and straight manner, and tighten the two screws.

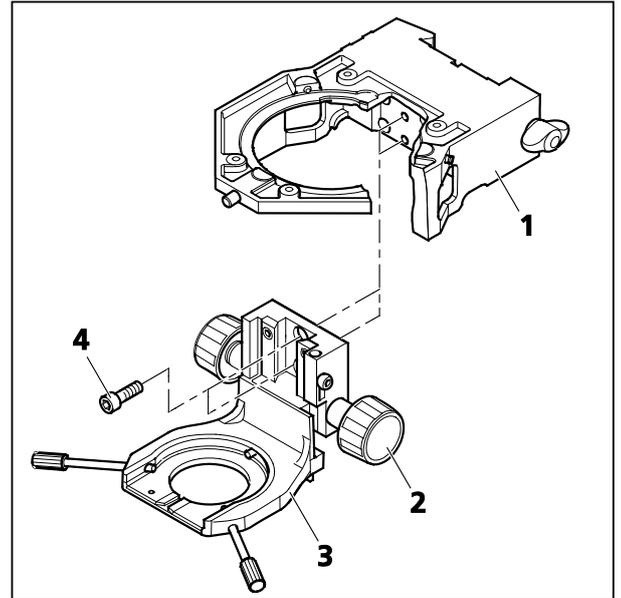


Fig. 3-18 Attaching the condenser carrier to the stage carrier

### 3.17 Changing the fine-drive knob or the fine-drive disk on the stand

The fine-focusing drive to the right and left on the stand can be provided either with a fine-drive knob or a fine-drive disk.

To change the fine-drive knob or the fine-drive disk, proceed as follows:

- Turn the coarse-drive knob on the relevant side of the stand in such a way that the mounting hole (3-19/2) points upward.
- Insert the ball-headed screwdriver SW 1.5 (3-19/1) into the mounting hole and loosen the locking screw of the fine-drive knob or the fine-drive disk.
- Pull the fine-drive knob or the fine-drive disk sideways off the shaft.
- Slide the fine-drive disk or the fine-drive knob onto the shaft, so that the recess (3-19/3 or 4) is located under the mounting hole.
- Tighten the locking screw.

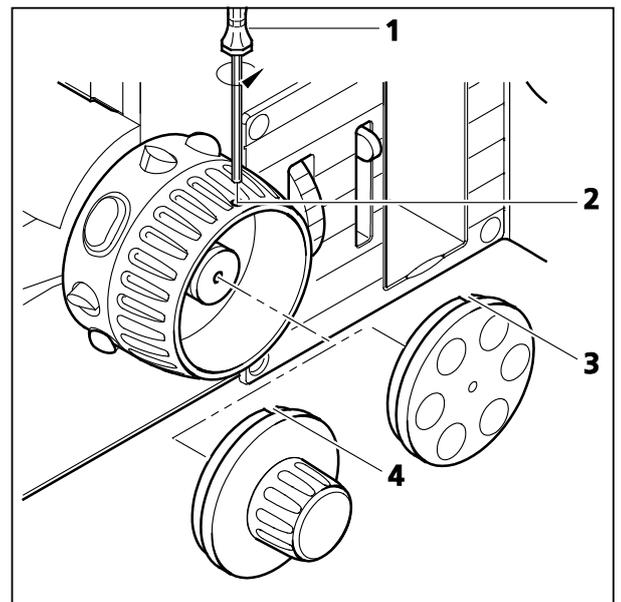
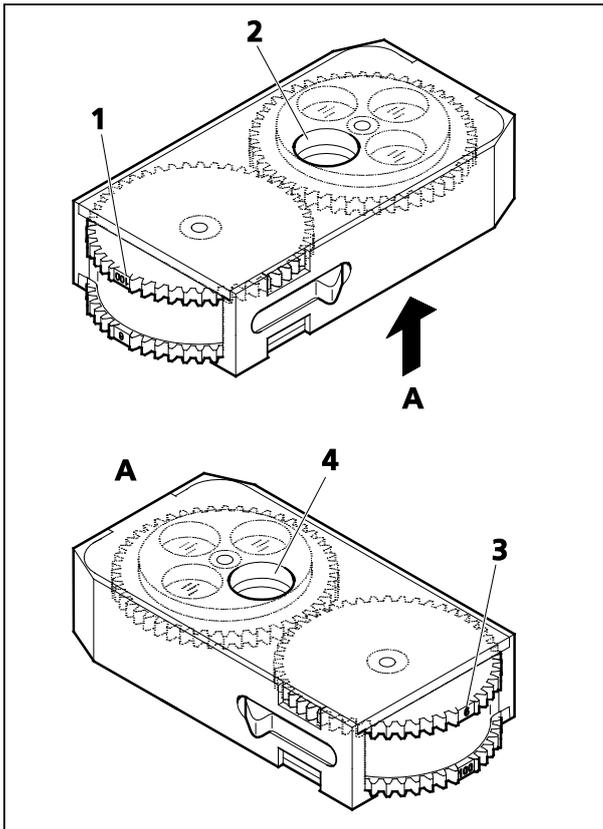


Fig. 3-19 Changing the fine-drive knob or fine-drive disk



**Fig. 3-20** Equipping 2-position filter wheel, manual

### 3.18 Equipping 2-position filter wheels 2x, discrete

#### 3.18.1 Filter wheel, manual

The supplied neutral-density filter set for transmitted light or reflected light consists of:

- 1 filter with 50%
- 2 filters with 25%
- 1 filter with 12%
- 1 filter with 6%
- 1 filter with 1.5%
- 8 retaining rings (3 as spares)

The manual filter wheel 2x need not be opened for inserting the filters in the two filter wheels.

The figures engraved to the filter wheel (3-20/1 or 3) indicate which filter position of the respective filter wheel is in the filter opening (filter wheel 1: 3-20/2 or filter wheel 2: 3-20/4). The figure indicates the transmission (in %) of the set filter position.

- Set the desired position on the filter wheel. Insert the corresponding filter with the reflective side up. Take care that no dirt gets onto the filter surface.
- Screw in the retaining ring.

In filter wheel 1, insert the following filters:

- Position **100**: No filter (100% transmission)
- Position **50**: Neutral-density filter 50%
- Position **25**: Neutral-density filter 25%
- Position **12**: Neutral-density filter 12%

In filter wheel 2, insert the following filters:

- Position **100**: No filter (100% transmission, two positions available)
- Position **6**: Neutral-density filter 6%
- Position **0.4**: Neutral-density filter 1.5% (to be inserted first) and neutral-density filter 25%

### 3.18.2 Filter wheel, 2-position, motorized

The motorized filter wheel 2x is to be equipped with the same filter set as the manual filter wheel 2x.

To insert the filters in the two filter wheels, the motorized filter wheel 2x must be opened on the corresponding side.

The filter mounts are labeled with the corresponding position numbers 1 to 4 (3-21/**1a** to **4a** and **1b** to **4b**).

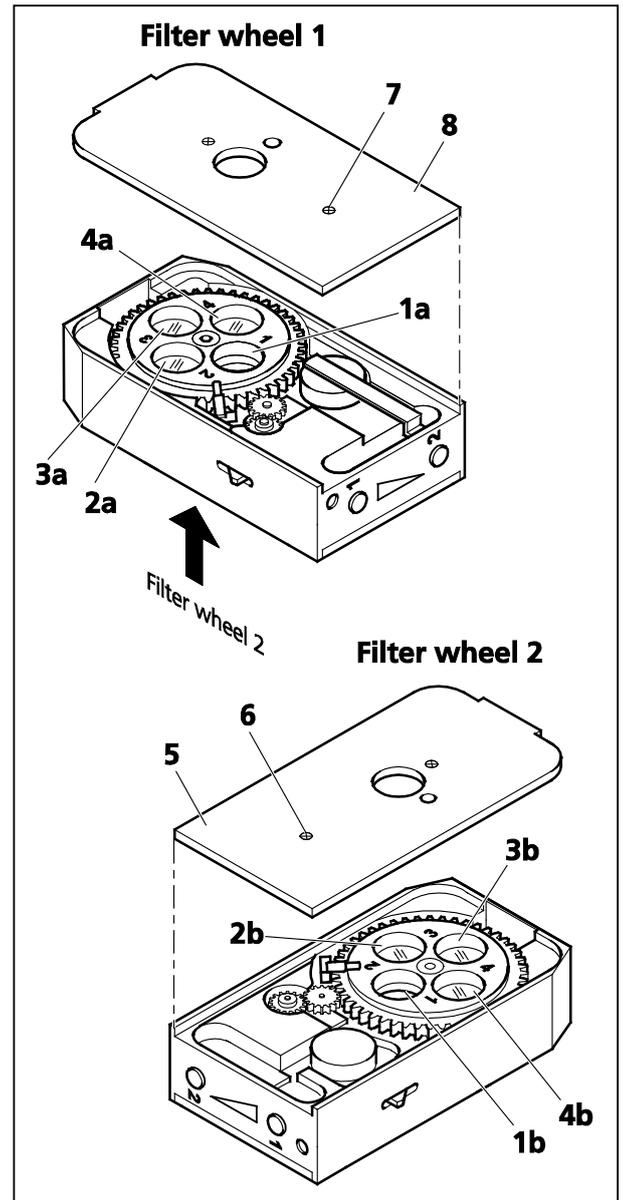
- Loosen both screws (3-21/**6** and **7**) and remove cover plate (3-21/**5** and **6**).
- Take care that no dirt gets onto the filter surface.
- Screw in the retaining ring.
- After all filters have been inserted, put on the cover plate and screw it down.

In filter wheel 1 (428301-9901-000; Fig. 3-21), insert the following filters:

- Position **1a**: No filter (100% transmission)  
 Position **2a**: Neutral-density filter 12%  
 Position **3a**: Neutral-density filter 25%  
 Position **4a**: Neutral-density filter 50%, reflective coating up

In filter wheel 2, insert the following filters:

- Position **1b**: No filter (100% transmission)  
 Position **2b**: Neutral-density filter 6%  
 Position **3b**: No filter (100% transmission)  
 Position **4b**: Neutral-density filter 0.5% and neutral-density filter 25%, reflective coating down



**Fig. 3-21** Equipping filter wheel 2x, motorized (only for 428301-9901-000)

### 3.19 Installing and removing P&C reflector modules

Normally, the reflector turret is factory-equipped with P&C (Push&Click) reflector modules on customer's request. However, the user can also change the equipment of these turrets.

The reflector turret is designed to hold maximally six or ten reflector modules dependent on the model.

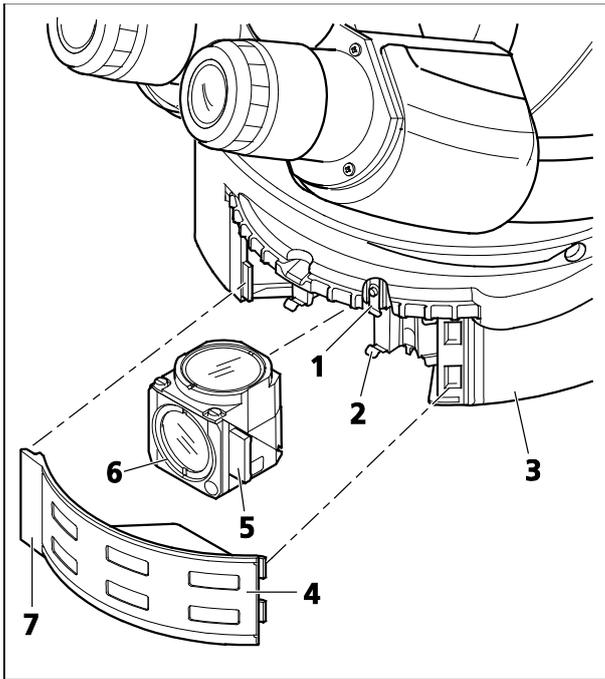


Fig. 3-22 Changing P&C reflector modules

#### 3.19.1 Installing a module

- Fold up cover flap (3-22/4) on reflector turret (3-22/3) to the right and unhinge it using the handle (3-22/7) arranged at the left.
- Turn reflector turret until the desired position (position ID labeled on reflector turret) becomes accessible in the mounting hole.
- Holding it by the handles (3-22/5) arranged right and left on the module, insert the module (3-22/6) obliquely from the bottom into the upper spring clips (3-22/1) on the reflector turret.
- Then press the module at the bottom until it snaps securely into the bottom spring clips (3-22/2) of the reflector turret as well.

#### 3.19.2 Removing a module

- Slightly tilting the module, pull it out first from the bottom spring clips and then from the top spring clips. Remove it completely.

- After having removed and installed the reflector modules, hang in the cover flap and let it snap in.
- Turn on the reflector turret by three (6-position reflector turret) or five positions (10-position reflector turret) to swing the just installed module into the optical path.
- After you equipped the turret as desired, label the adhesive labels with the new filter combination and affix them to the corresponding fields on the cover flap.

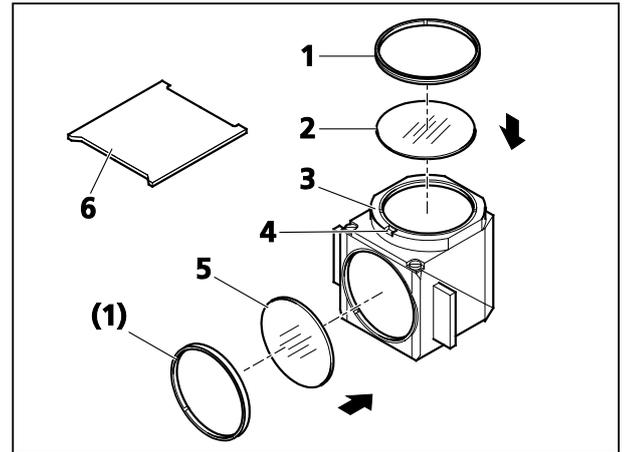
### 3.20 Changing the filter set in the reflector module FL P&C

The filter sets for the FL P&C reflector module can be combined and assembled individually by the customer.

Insert only fluorescence filters having a free aperture of  $\geq 22$  mm, as otherwise the image may be masked. Make sure this requirement is met, when using filters from other manufacturers.

Filter sets or fully assembled FL P&C reflector modules can be ordered from Carl Zeiss.

- Remove reflector module FL P&C (3-23/3) from the reflector turret and put it down (refer to Section 3.19).
- Use mounting plate (3-23/6) of tool kit to unscrew retaining ring (3-23/1).
- Turn the reflector module round and let the filter (3-23/2 or 5) drop out on a soft surface.
- Insert the barrier filter (emission filter) at (3-23/2), the exciter filter at (3-23/5). Secure both filters by means of retaining rings (3-23/1).



**Fig. 3-23** Changing the filter set in reflector module FL P&C

Barrier filter and exciter filter may be provided with a designation and an arrow on their circumference. The arrow indicates the direction the particular filter is to be installed in the reflector module; it must always point inwards (refer to arrows in Fig. 3-23).

To minimize image offset during multiple fluorescence image captures, an additional label can be provided on the barrier filter to indicate the position of the wedge angle.

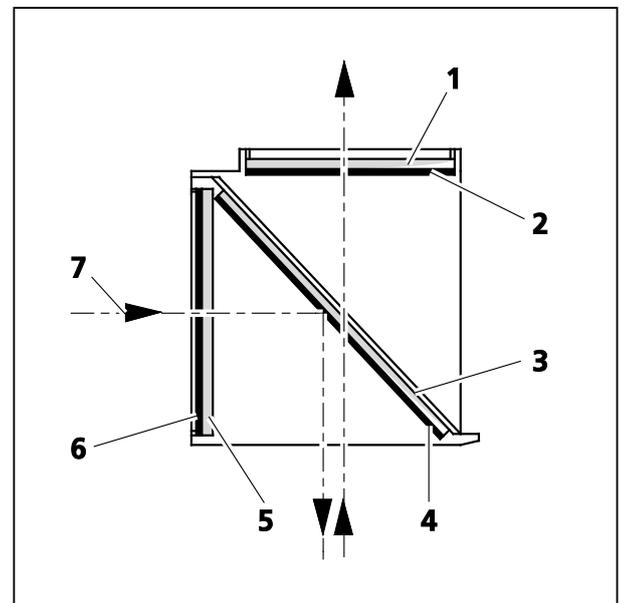
This label should be aligned to the orientation groove (3-23/4) when you insert the barrier filter in the reflector module used. This is to ensure that the wedge angle of the barrier filters is in the same, defined position in the reflector modules used thus compensating or minimizing the already minimal module-to-module image shift when Zeiss filter sets are used.

If it is necessary to mount filters that do not carry any directional mark (arrow), it is advisable to follow this procedure:

Mount the filters with the reflective dielectric layers in such a way that the reflective layer (3-24/6) on the exciter filter (3-24/5) points outwards (relative to the reflector module). On the barrier filter (3-24/1), the reflective layer (3-24/2) points inwards (Fig. 3-24).

The reflective layer (3-24/4) of the beam splitter (3-24/3) should point downward when in its mounting position.

The arrows (3-24/7) mark the illumination and imaging beam path.



**Fig. 3-24** Installing filter and beam splitter

### 3.21 Changing the beam splitter in the reflector module FL P&C

 In mounting filters and beam splitters, take extreme care to prevent damage to and contamination of the optical components.

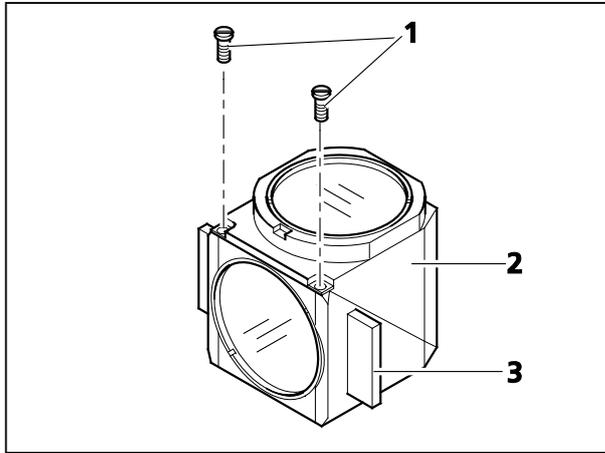


Fig. 3-25 Changing the beam splitter

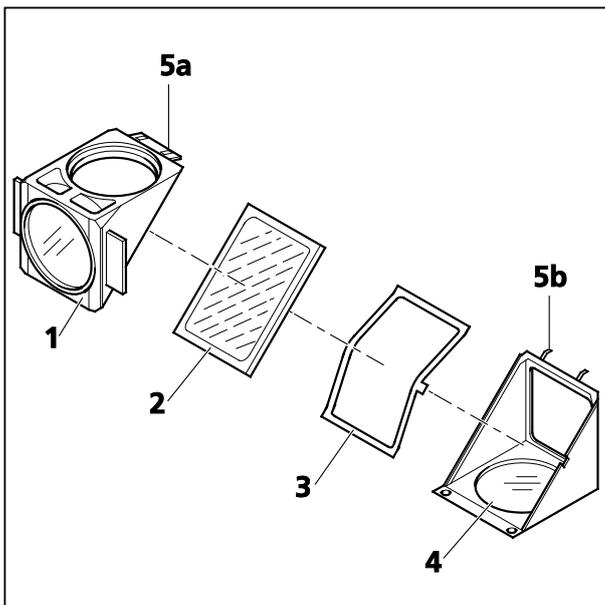


Fig. 3-26 Changing the beam splitter

We recommend ordering completely equipped FL P&C reflector modules, since changing the beam splitter is quite demanding.

However, if you should choose to change the beam splitter, follow this procedure:

- Remove the FL P&C reflector module from the reflector turret (also refer to Section 3.19.2).
- Undo the two slotted screws (3-25/1) with a screwdriver.
- Hold both halves of the reflector module together (**emission** half (3-25/2) and **excitation** half (3-25/3), turn in the position opposite to the installation position and put it down.
- Tip up the **excitation** half of the module (3-26/1), which now is on top, and remove it from the retaining pins (3-26/5b) on the bottom emission half of the module (3-26/4).
- Remove beam splitter (3-26/2) and spring-loaded frame (3-26/3) from the bottom half of the module.
- Remove the old beam splitter and carefully place the new one onto the spring-loaded frame (3-26/3) with the reflective side facing up and place both parts together into the bottom half of the module. Take care to ensure that the side tongue of the spring-loaded frame is in the appropriate recess in the bottom half of the module.

 The reflective (coated) side (3-27/3) of the beam splitter has a beveled edge (3-27/1) or corner (3-27/2).

- Place the **excitation** half of the module (3-26/1) onto the **emission** half (3-26/4) (retaining pins 3-26/5b and eyelets 3-26/5a mesh with one another). Hold both halves together and turn them back into the installation position.
- Re-insert the slotted screws and tighten them up.
- Finally, affix the adhesive label with the name of the filter combination to the side of the module.

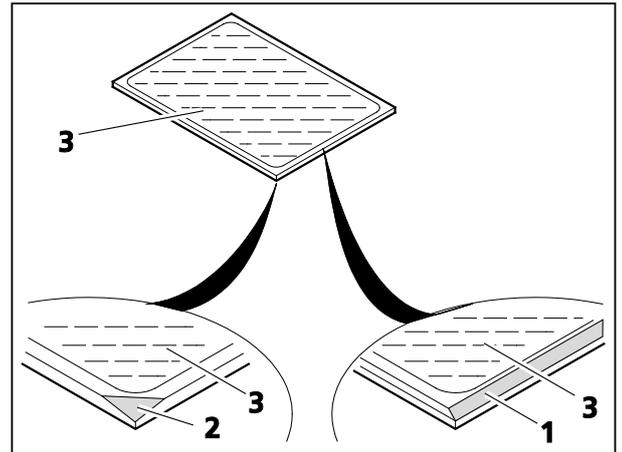


Fig. 3-27 Marks on beam splitter

### 3.22 Activating/deactivating the high-performance focus

When a scanning stage (approx. weight: 4 kg) is used, the high-performance focus must be activated to protect the focusing drive.

Thus, specimens weighing up to 5 kg may be placed on the stage for microscoping.

To activate/deactivate the high-performance focus, proceed as follows:

- Use the Z drive to move the microscope stage into the lowest position possible.
- Use ball-headed screwdriver SW 3 (3-28/2) to screw the two screws (3-28/1), located to the right **and** left on the stand, in or out until they reach their respective end positions.

Screws screwed out: High-performance focus is activated.

Screws screwed in: High-performance focus is deactivated.

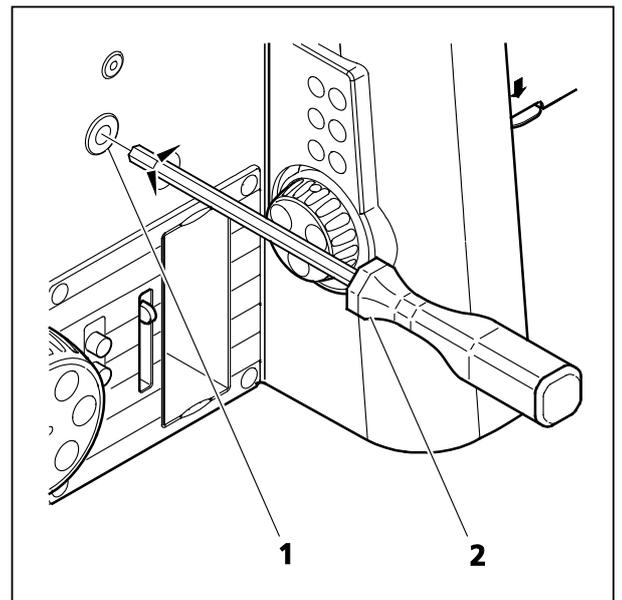


Fig. 3-28 Activating/deactivating the high-performance focus

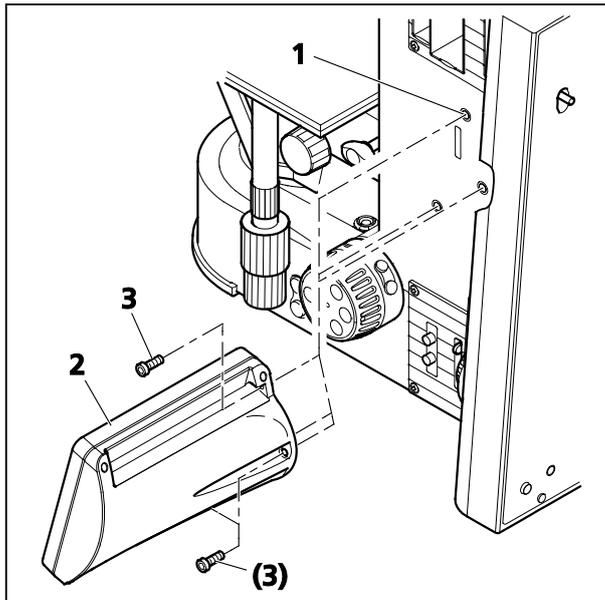


Fig. 3-29 Mounting the TFT display

### 3.23 Mounting the TFT display to the motorized stand



#### CAUTION

When mounting the TFT display, the microscope must be switched off.

- Mount the TFT display (3-29/2) to the right side of the motorized stand (3-29/1) using the three screws (3-29/3).

In doing so, stand and TFT display are automatically connected electrically via the plug contact.

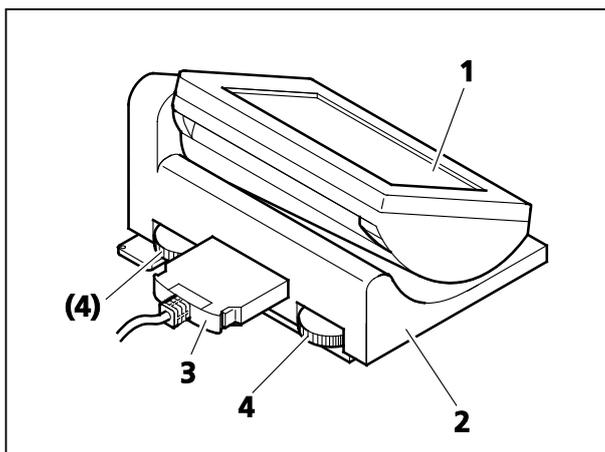


Fig. 3-30 Attaching the TFT display to the docking station

### 3.24 Attaching the TFT display to the docking station



#### CAUTION

To connect the TFT display and the docking station, the microscope must be switched off.

- To hook the docking station up to the motorized stand, the corresponding plug-in module carrying the **DOC** port must be installed on the back of the stand.



The plug-in module is to be installed by the sales technician only.

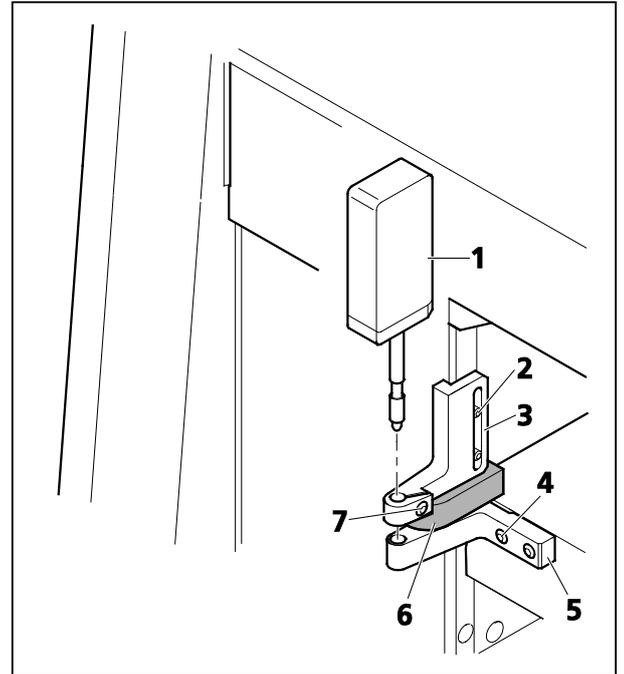
- If the TFT display is already mounted on the motorized stand, it needs to be dismantled first.
- Then, the mounting holes and the opening for the plug-in contact on the stand must be closed with the caps supplied.
- Put the TFT display (3-30/1) on the docking station (3-30/2) and screw it on, using the long Allen screwdriver provided. In so doing, make sure that the plug-in contact is accurately inserted into the opening.
- Use the connection cable (3-30/3) to connect the docking station to the **DOC** port on the back of the stand.
- The angle of the TFT display can be changed with the aid of the two knurled thumb screws (3-30/4) located on the back of the docking station..

### 3.25 Installing the focus linear sensor

 Only in connection with Axio Imager.Z2 / .Z2m stands.

 Use adjusting block 430702-0102-000 for installation.

- Switch on the microscope (see Section 4.3.1.1).
- Screw objective which is in optical path out of nosepiece.
- Move the mechanical stage with stage carrier to the higher stop position.
- Put the adjusting block (3-31/6) against the stage carrier and fix the top part of holder Z (3-31/3) on it.
- Bring the top part of holder Z (3-31/3) into line with the adjusting block and fasten it with both screws (3-31/2) to the stand.
- Press the bottom part of holder Z (3-31/5) against the adjusting block, so that stage carrier and bottom part of holder Z are flush.
- Fasten bottom part of holder Z (3-31/5) with both screws (3-31/4) to the stage carrier.
- Insert the focus linear sensor (3-31/1) in the top part, bring it into line with the lower edge and fasten it by means of clamping screw (3-31/7).
- Move the stage carrier to the lower stop position and remove adjusting block.
- Switch off the microscope (see Section 4.3.1.2).
- Plug the cable of the focus linear sensor into connector (3-50/7) on the rear side of the stand and switch on the microscope (see Section 4.3.1.1).



**Fig. 3-31** Installing the focus linear sensor

 An incorrect installation of the focus linear sensor can cause problems in focusing the object.

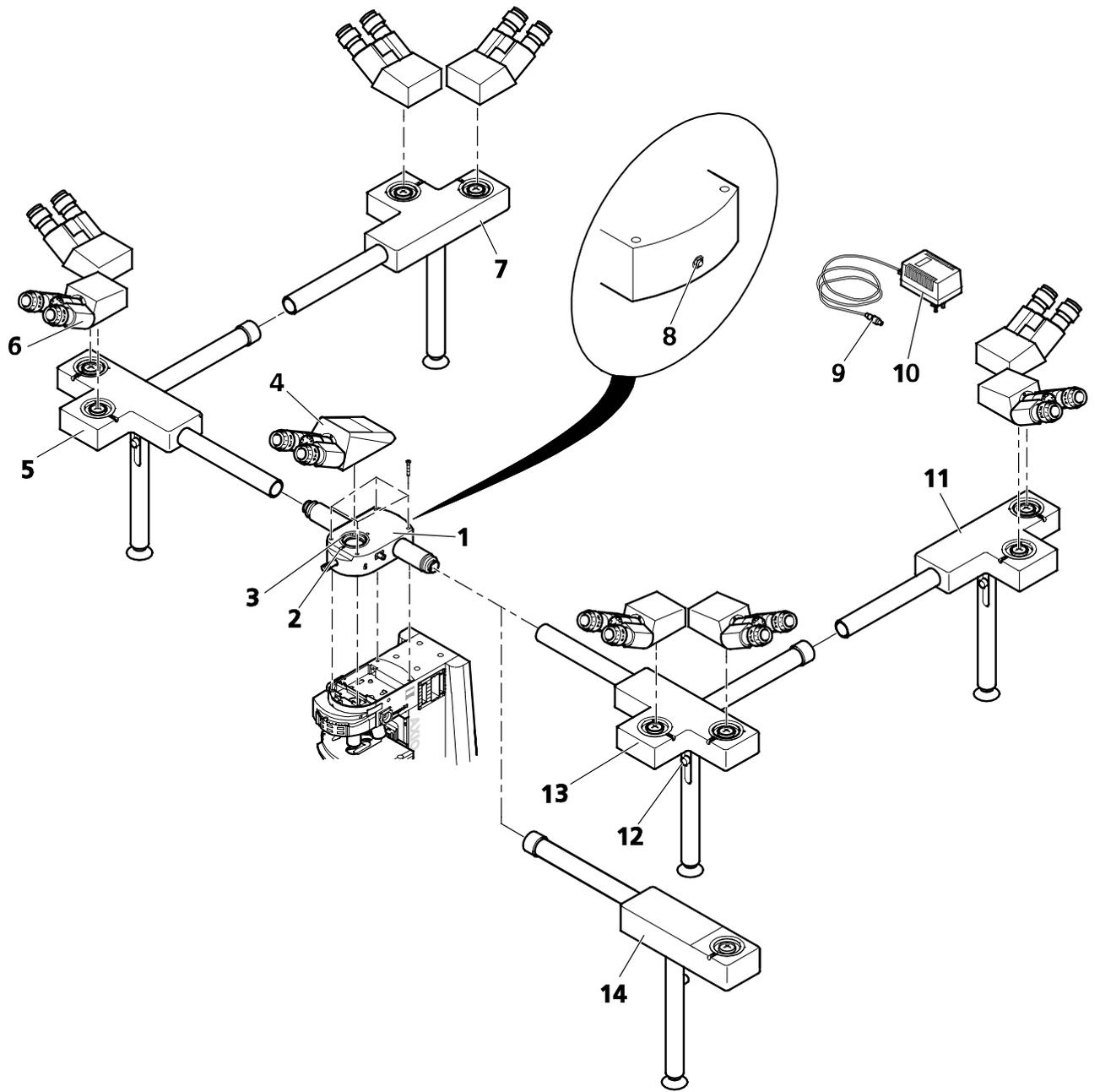
 The focus linear sensor can be activated and deactivated via the TFT display.

### 3.26 Assembling the multidiscussion equipment

For the installation of the multidiscussion equipment, we recommend you call on Zeiss Microscopy Service (see Section 5.5), as it requires careful optical adjustment and centering of the fields of view of the individual co-observer tubes.

If nevertheless you want to install the equipment yourself, follow this procedure:

- Remove binocular tube and coupling plate or intermediate tube (if installed) from the stand (refer also to 3.2 ff.).
- Install the multidiscussion equipment (3-32/1) in the same way as the tube lens turret.
- Remove the tube lens from the tube.
- In place of the tube lens, screw in the gray filter (gray filter must be ordered separately. It is not required when the high light intensive extension arm is used).
- Insert the binocular tube or phototube (3-32/4) in the dovetail mount (3-32/3) of the multidiscussion equipment, align it and fasten it by means of clamping screw (3-32/2).
- To the left side of the multidiscussion equipment, attach connecting piece L (3-32/5), end connector L (3-32/7) or the high light-intensity extension arm (3-32/14). In doing so, be sure to remove the protective caps from the tube sockets. The pin must engage in the opening. Then, screw down the adapters with the sleeve nut.
- To the right side, fasten connecting piece R (3-32/13), end connector R (3-32/11) or the high light-intensity extension arm (3-32/14). In doing so, be sure to remove the protective caps from the tube sockets. The pin must engage in the opening. Then, screw down the adapters with the sleeve nut.
- Install and clamp two binocular tubes each for the multidiscussion equipment (3-32/6) to connecting piece L (3-32/5) and connecting piece R (3-32/13). Install and clamp one binocular tube to the high light-intensity extension arm.
- When aligning the adapters and end pieces mechanically, take care to ensure that the fields of view, starting from the tube of the main observer (3-32/4) keep centered. To check this, select a prominent feature of the specimen and move it to the intersection point of the eyepiece reticle. After you have installed the first adapter, insert an eyepiece with eyepiece reticle in the corresponding co-observer tube and particularly perform vertical adjustment of the adapter tube by means of setscrew (3-32/12) until the selected specimen detail is again in the intersection point of the eyepiece reticle.
- To connecting piece L, add another connecting piece L (3-32/5) or end connector L (3-32/7) and to connecting piece R add another connecting piece R (3-32/13) or end connector R (3-32/11) for multidiscussion equipment and fasten the parts.
- Attach and clamp two binocular tubes for multidiscussion equipment to end connector R and end connector L each. Perform vertical adjustment with the eyepiece reticle.
- Insert plug (3-32/9) of plug-type power supply (3-32/10, 115 V or 230 V) in connector (3-32/8) on the multidiscussion equipment and securing it against slipping off by tightening the screw ring.
- Connect the plug-type power supply for the light pointer (3-32/10) to a power outlet (115 V or 230 V).



- |   |                           |    |                            |    |                                    |
|---|---------------------------|----|----------------------------|----|------------------------------------|
| 1 | Multidiscussion equipment | 6  | Co-observer tube           | 11 | End connector R                    |
| 2 | Clamping screw            | 7  | End connector L            | 12 | Setscrew                           |
| 3 | Dovetail                  | 8  | Connector for power supply | 13 | Connecting piece R                 |
| 4 | Binocular tube            | 9  | Plug                       | 14 | High light-intensity extension arm |
| 5 | Connecting piece L        | 10 | Plug-type power supply     |    |                                    |

**Fig. 3-32 Assembling the multidiscussion equipment**

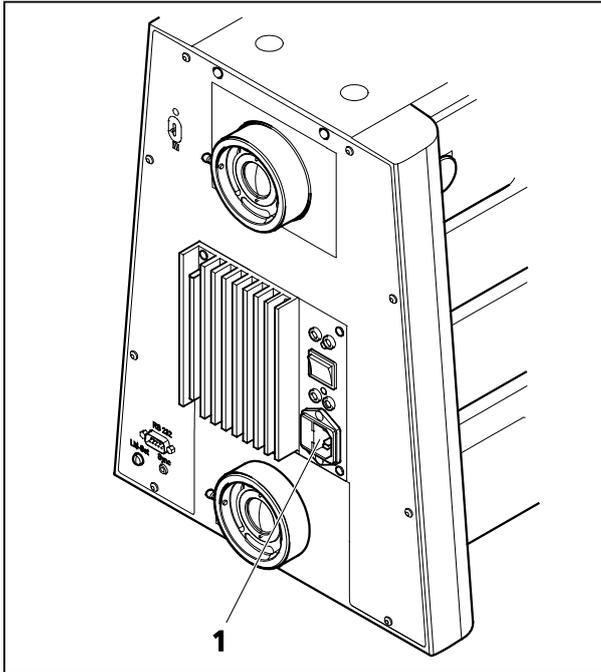


Fig. 3-33 Axio Imager, manual

### 3.27 Connecting to power

#### 3.27.1 Manual stand

- Plug the power cable first into the power connector (3-33/1) of the microscope and then into a power outlet. The microscope can be connected to a line voltage of 100 to 127 V or 200 to 240 VAC, 50/60 Hz. The power unit is set **automatically** to the line voltage available.

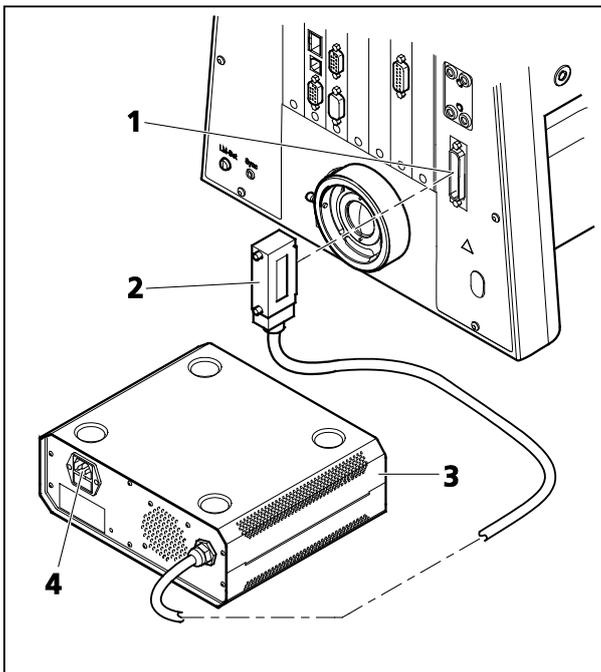


Fig. 3-34 Axio Imager, motorized

#### 3.27.2 Motorized stand

- Plug connecting plug (3-34/2) of power supply VP232-2 (3-34/3) into the corresponding connector (3-34/1) on the rear side of the stand.
- Plug the power cable first into the power connector (3-34/4) of the power supply VP232-2 and then into a power outlet. The power supply can be connected to a line voltage of 100 ... 240 VAC, 50/60 Hz. The power unit is set **automatically** to the line voltage available.

### 3.28 HAL 100 halogen illuminator

The HAL 100 illuminator is used as light source for transmitted-light and reflected-light techniques (except fluorescence) on the Axio Imager. Attachment to the reflected-light or transmitted-light socket is performed analogously.

#### 3.28.1 Attaching the HAL 100 halogen illuminator



Before using the halogen illuminator, make sure to remove the halogen lamp replacement tool from the housing. Otherwise, it might be damaged by heat (refer to Section 3.28.4)

- Remove the protective cap from the reflected-light or transmitted light socket.
- Insert the dovetail mount of the lamp housing (3-35/9) into the corresponding socket (3-35/2 or 3-35/6) and, using the SW 3 ball-headed screwdriver, tighten it with clamping screw (3-35/1 or 3-35/8).
- Insert 3-pole lamp plug (3-35/7) into 3-pole 12 V/100 W connector (3-35/3 – for reflected light or 3-35/5 - for transmitted light) on the back of the instrument.
- Switch the toggle switch for transmitted/ reflected light (3-35/4) to the required position. When it comes to the motorized stand, switching between reflected light and transmitted light is done via the TFT display touch screen.



The halogen illuminators installed for reflected light and transmitted light can only be switched on alternately.



The light manager functionality depends on the position of the toggle switch.

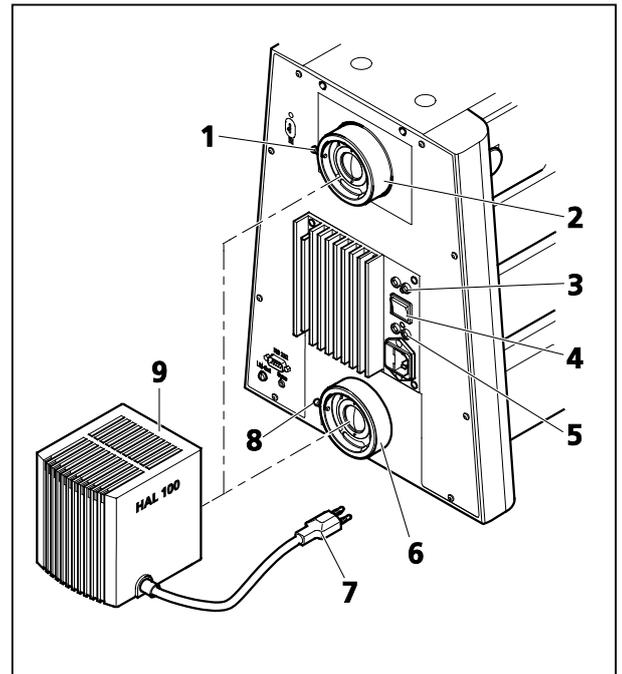


Fig. 3-35 Attaching the HAL 100 halogen illuminator

### 3.28.2 Separate power supply of the HAL 100 halogen illuminator in reflected light

For the combined use of transmitted light and reflected light (mixed light) on the Axio Imager, the halogen illuminator needs to be connected to a separate power supply in reflected light.

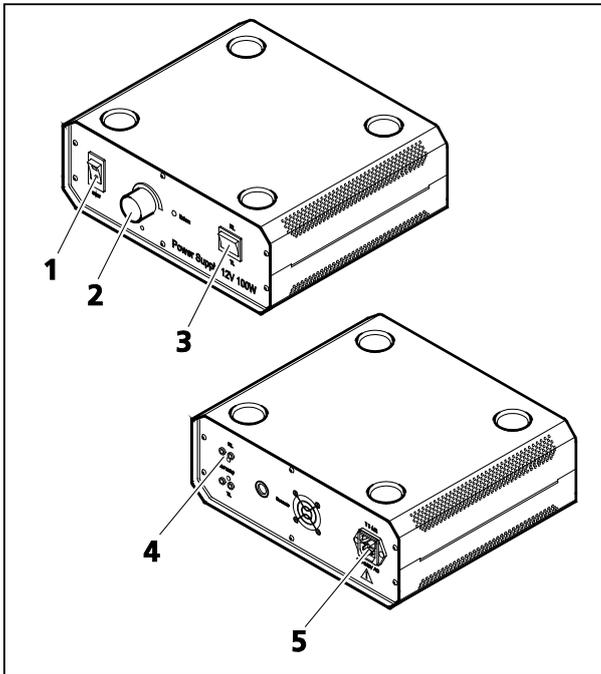


Fig. 3-36 Power supply 12V 100W for HAL 100 in reflected light

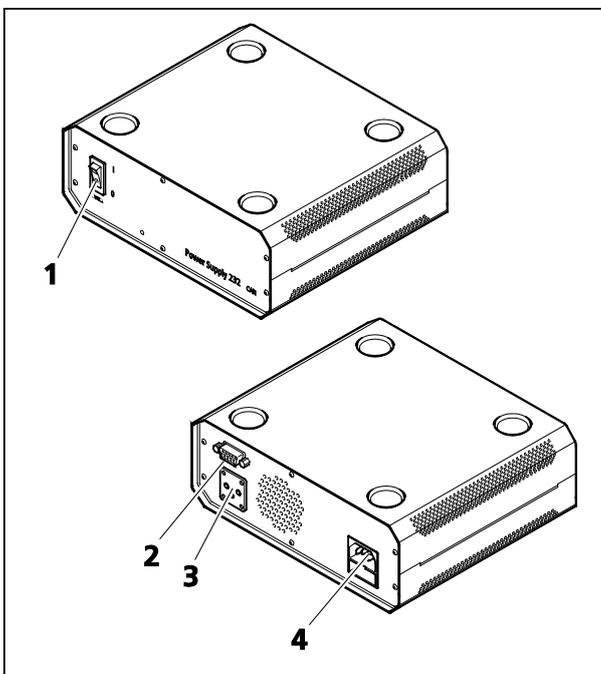


Fig. 3-37 Power Supply 232 CAN for HAL 100 in reflected light

For the coded stand, the external power supply for HAL 100 and LED illuminators (Power Supply 12V 100W) is to be used, while the external power supply for HAL 100 and LED illuminators with CAN connection (Power Supply 232 CAN) is to be employed for the motorized stand.

To connect the HAL 100 to a separate power supply, proceed as follows:

#### Coded stand

- In reflected light, connect the three-pin plug of the HAL 100 to socket **RL** (3-36/4) on the back of Power Supply 12V 100W.
- Set the transmitted-light / reflected-light toggle switch to the position for reflected light (**RL**).
- Use a power cord to connect the power connector (3-36/5) to a power outlet.
- For reflected light, use the ON / OFF switch (3-36/1) on the power supply to turn the HAL 100 on or off, as required.
- Use the rotary knob (3-36/2) on the power supply to adjust illumination intensity.

#### Motorized stand

- In reflected light, connect the three-pin plug of the HAL 100 to the socket (3-37/3) on the back of Power Supply 232 CAN.
- Use a CAN connection cable to connect the CAN connector on the power supply (3-37/2) with a free CAN connector on the back of the stand.
- Use a power cord to connect the power connector (3-37/4) to a power outlet.
- For reflected light, use the ON / OFF switch (3-37/1) on the power supply to turn the HAL 100 on or off, as required.
- Set the illumination intensity of the HAL 100 on the TFT display.
- Set the illumination intensity of the HAL 100 on the TFT display.

### 3.28.3 Adjusting the HAL 100 halogen illuminator

#### (1) Coarse adjustment

- Loosen clamping screw (3-35/1 or 3-35/8) and remove the operational halogen illuminator (3-38/2) from the microscope stand.
- Switch on microscope as described in Section 4.3.
- Direct the light beam to a projection surface (wall) that is at least 3 m away.



Do not look into the light exit aperture of the illuminator.

- Using the SW 3 ball-headed screwdriver turn adjusting screw (3-38/3) until both images of the lamp filament appear as sharp as possible on the projection surface.
- Then, turn adjusting screws (3-38/4 and 5) until the lamp filament of one image exactly fills the gaps of the reflected filament image (3-38/1).

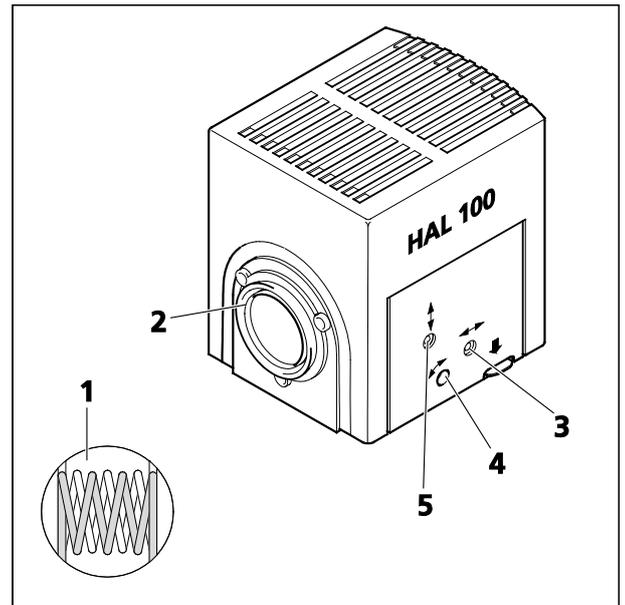


Fig. 3-38 Adjusting the HAL 100 halogen illuminator

#### (2) Fine adjustment

- Reattach the microscope illuminator to the microscope stand and lock it with the clamping screw.
- Move diffusion disk for reflected or transmitted light out and remove the filter wheels from the respective slot.
- With an objective of  $\leq 40x$ , focus on the specimen and search for a free place on the specimen.
- Remove eyepiece and, in the pupil image visible in the eyepiece socket, center the lamp filament and its reflection with adjusting screws (3-38/4 and 5).
- Using adjusting screws (3-38/3) optimize the evenness of illumination of the pupil image.



It is advisable to use the adjusting aid (4-1/5) for fine adjustment of the halogen illuminator mounted to the reflected-light socket. After pulling out the adjusting aid, the lamp filament and its reflection can be viewed directly in its viewing glass.

- Move the diffusion disk in and reinsert the filter wheels.

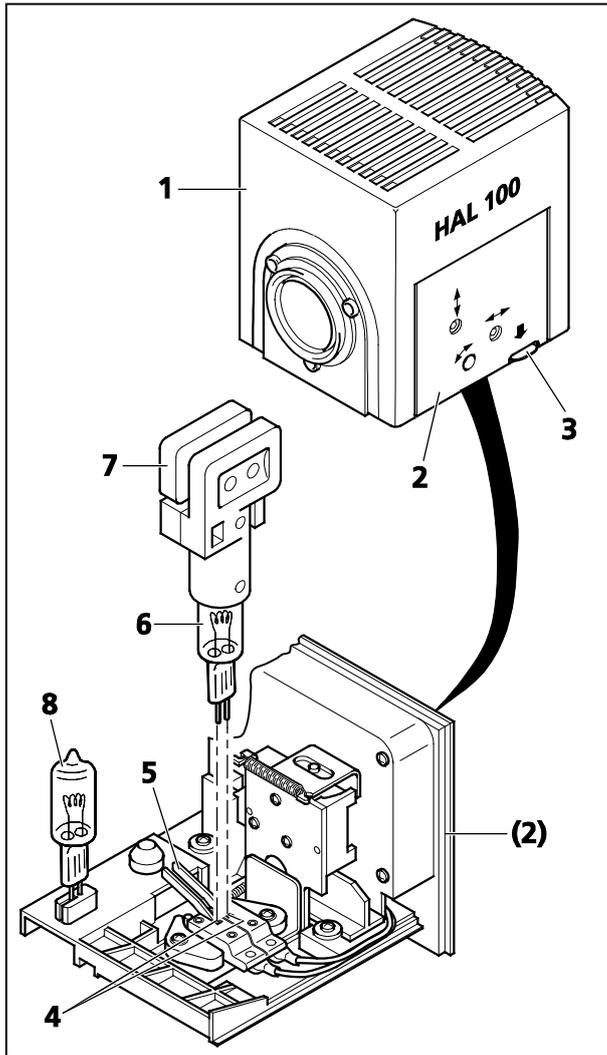


Fig. 3-39 Changing the halogen lamp

### 3.28.4 Replacing the HAL 100 halogen lamp



**CAUTION**  
Hot surface!



You need not remove the lamp housing from the stand to replace the halogen lamp. **Do not** store the supplied replacement tool (3-39/7) for lamp replacement in the lamp housing while the illuminator is operating. The replacement lamp (3-39/8) may remain put on in the lamp housing.

- Switch off the microscope as described in Section 4.3, disconnect plug (3-35/7) from connector 12 V/100 W (3-35/3 – reflected light or 3-35/5 – transmitted light) and allow for a cool down of approximately 15 minutes.
- Depress unlock button (3-39/3) of halogen illuminator HAL 100 (3-39/1), fully pull out lamp carrier (3-39/2) and put it down separately.
- Depress both spring levers (3-39/5) and remove the old halogen lamp (3-39/6) upward.
- Depressing both spring levers insert the new lamp in the lamp socket (3-39/4) and release the spring levers. Always hold/grasp the halogen lamp by means of the replacement tool (3-39/7), as even traces of grease on the lamp may affect its lifetime.
- Shortly depress spring levers once more to center the lamp.
- Insert lamp carrier until it locks.

### 3.29 Installing the LED illuminator for transmitted light

The LED illuminator for transmitted light can be installed optionally on the transmitted light port on the rear side of the instrument or on the bottom of the condenser carrier.

The LED illuminator delivers light of constant color temperature independent of the set light intensity.



#### CAUTION

Avoid looking directly into the LED light.



#### CAUTION

Always make sure that the instrument is switched off when mounting the LED illuminator onto the transmitted-light port or dismounting it from the transmitted-light port.

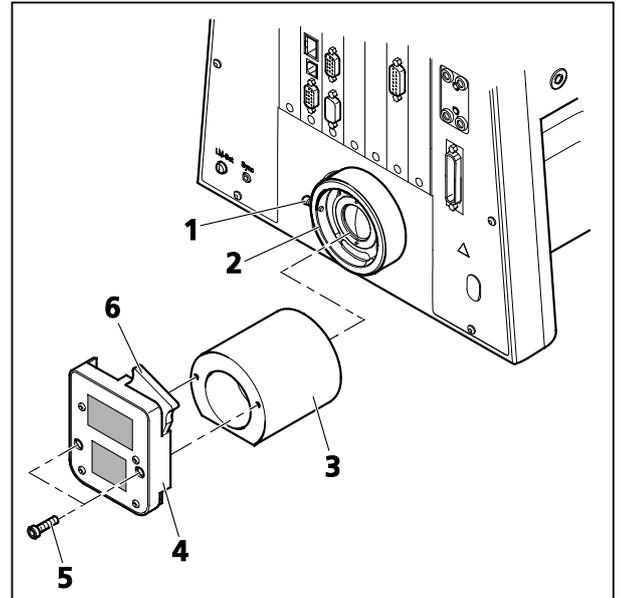


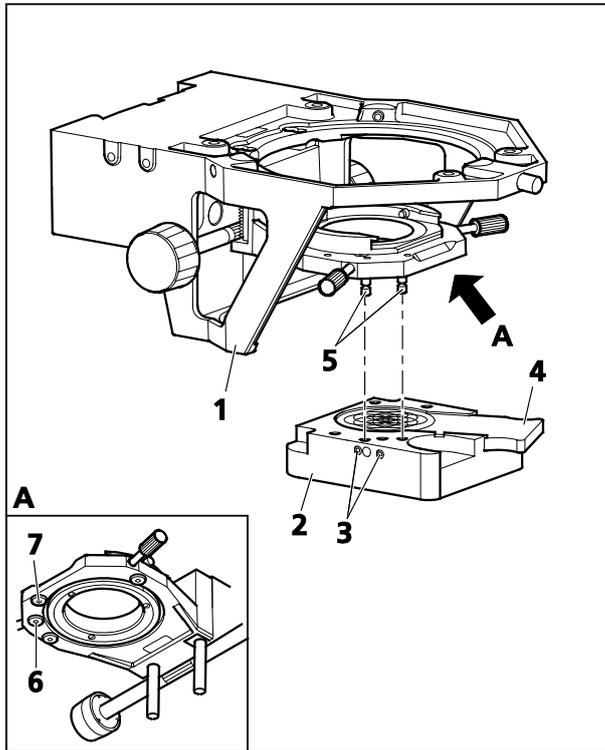
Fig. 3-40 Installing the LED illuminator

#### Installing the LED illuminator on the transmitted light port

- Loosen clamping screw (3-40/1) on transmitted-light port (3-40/2). Remove the halogen lamp.
- Disconnect the illuminator plug from the 12 V/100 W connector (transmitted light) on the back of the instrument.
- Insert adapter optics for LED illuminator (3-40/3) in the transmitted-light port with the slant pointing down and tighten the clamping screw.
- Screw LED illuminator (3-40/4) onto the adapter optics using the provided Allen screws (3-40/5).
- Push slider with diffusion disk 10° (3-40/6) into LED illuminator.
- Plug the plug of the LED illuminator into the 12 V/100 W connector (transmitted light) on the back of the instrument.



You can change the color temperature for specimen observation by inserting color temperature filters in the slider with diffusion disk (to be held by retaining ring).



**Fig. 3-41** Installing the LED illuminator

### Installing the LED illuminator on the condenser carrier

- Remove the halogen illuminator from the transmitted-light port and disconnect the illuminator plug. Close the transmitted-light port with the cover cap.
- Move microscope stage and condenser carrier fully up using focusing drive (stage) and height control (condenser).
- Screw bolts (3-41/5) by hand into the tapped holes (3-41/6 and 7) in the bottom of the condenser carrier.
- Hold the LED illuminator (3-41/2) parallel to the underside of the condenser carrier (3-41/1). Move it upwards so that the bolts (3-41/5) fit into the corresponding holes on the illuminator. Fasten the illuminator by means of the countersunk screws (3-41/3) located at the left side of the LED illuminator.
- Push slider with diffusing glass 80° (3-41/4) into the LED illuminator, or for transmitted-light polarization, push in the polarizer for LED (427708-0000-000).

When using the polarizer, simple polarization contrast is possible. However, conoscopy cannot be carried out.

- Plug the plug of the LED illuminator into the 12 V/100 W connector (transmitted light) on the back of the instrument.

 You can change the color temperature for specimen observation by inserting color temperature filters in the slider with diffusing disk (to be held by retaining ring).

### 3.30 Attachment lamp VIS-LED for transmitted light

#### 3.30.1 Installing the attachment lamp VIS-LED

The attachment lamp VIS-LED is only intended for use with transmitted light.



#### CAUTION

Avoid looking directly into the LED light.



#### CAUTION

Always make sure that the instrument is switched off when mounting the attachment lamp VIS-LED onto the transmitted-light port or dismounting it from the transmitted-light port.

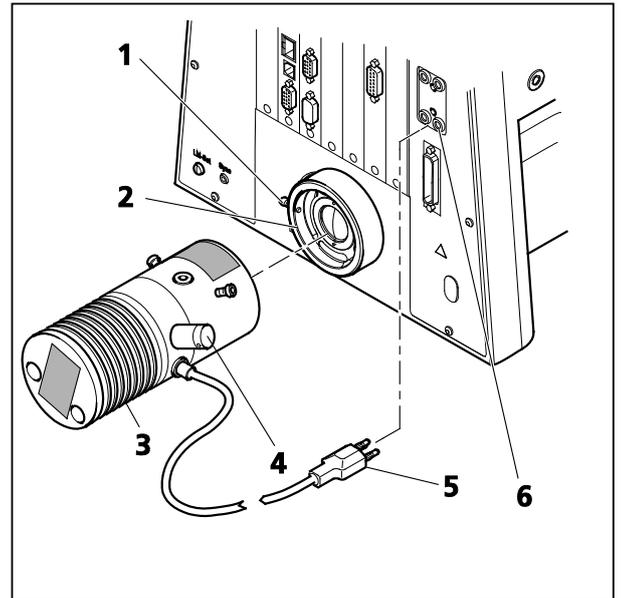


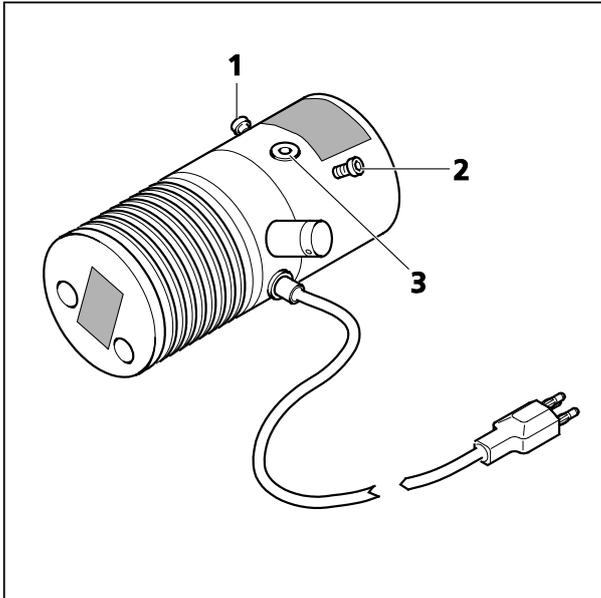
Fig. 3-42 Installing the attachment lamp VIS-LED

- Loosen the locking screw (3-42/1) on the transmitted-light port (3-42/2) and remove the halogen lamp.
- Insert the add-on illuminator (3-42/3) with the dovetail ring into the port (3-42/2). Use ball-headed screwdriver SW 3 and the locking screw (3-42/1) to lock the illuminator firmly into position.
- Connect the three-pin illuminator plug (3-42/5) to the lower three-pole 12 V/100 W socket (3-42/6) for transmitted light (**TL**) on the back of the instrument.
- For the coded stand, set the transmitted-light/reflected-light toggle switch to the position for transmitted light (**TL**). When it comes to the motorized stand, switching between reflected light and transmitted light is done via the TFT display touch screen.



If the AxioCam MRm is used for documentation purposes, the 12-percent gray filter provided must be employed to attenuate light intensity.

The add-on illuminator is equipped with a BNC port (3-35/4) for TTL trigger control.



**Fig. 3-43** Adjusting the attachment lamp VIS-LED

### 3.30.2 Adjusting the attachment lamp VIS-LED

- Switch on the add-on illuminator via the microscope.
- Switch off the diffusion disk for transmitted light and remove the filter wheels for transmitted light.
- Use  $\leq 40\times$  objective to focus on a free area of the specimen.
- Remove the eyepiece. Use the adjusting screws (3-43/**1** and **2**) to center the image of the illuminator in the pupil image.
- Use the focusing screw (3-43/**3**) to optimize the pupil image evenly.
- Switch the diffusion disk for transmitted light on again and insert the filter wheels for transmitted light.

### 3.31 HBO 100 illuminator

#### 3.31.1 Inserting the HBO 103 W/2 mercury vapor short-arc lamp

For safety reasons, the HBO 100 illuminator and the HBO 103 W/2 mercury vapor short-arc lamp are supplied separately packed.

Therefore, inserting the HBO 103 W/2 lamp into the lamp housing is the first step to start up this illuminator.

For a description of how to insert or replace the HBO 103 W/2 lamp, read the instructions for use supplied with it.



#### CAUTION

To reduce the transmission, use an FL attenuator, discrete (423616-0000-000 or 423617-0000-000). The gray filters mounted in the 2-position filter wheels (428300-0000-000 or 428301-0000-000) are not permanently stable.

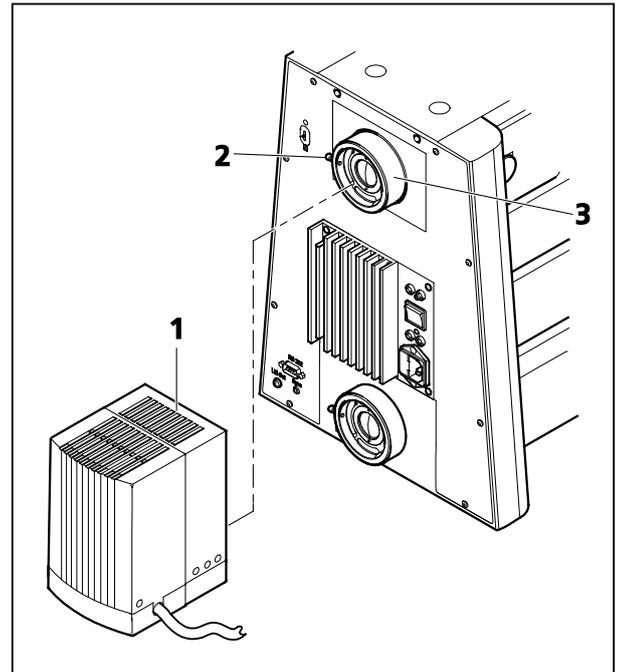


Fig. 3-44 Attaching the HBO 100 illuminator

#### 3.31.2 Attaching the HBO 100 illuminator

- Remove the cover from the reflected-light socket (3-44/3).
- Insert the dovetail of the lamp housing (3-44/1) into the reflected-light tube (3-44/3) on the back of the instrument and tighten the clamping screw (3-44/2) by means of a SW 3 ball-headed screwdriver.
- Insert the multi-pin plug of the HBO 100 illuminator into the device connector (3-45/1) on the HBO 100 transformer and secure it by means of a coupling ring.
- First, connect the power cable to the power socket (3-45/2) on the HBO 100 transformer, and then connect it to a power outlet.

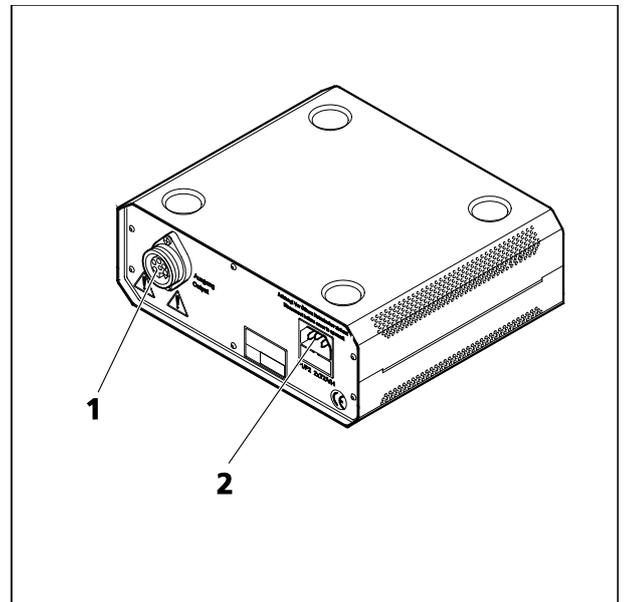


Fig. 3-45 HBO 100 transformer

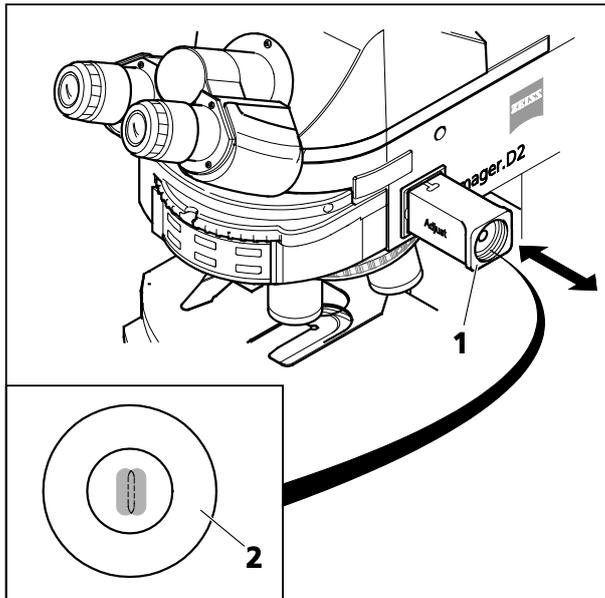


Fig. 3-46 Adjusting aid

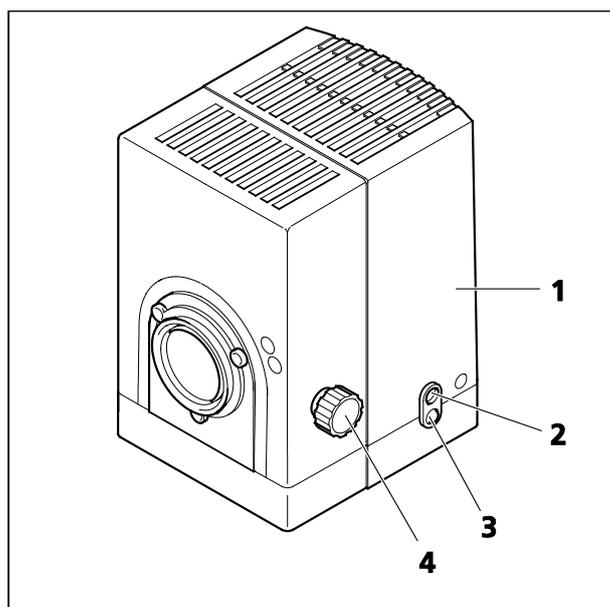


Fig. 3-47 Aligning the HBO 100

### 3.31.3 Aligning the HBO 100 illuminator

The HBO 100 illuminator is available in two versions (manual and automatic alignment).

The self-adjusting HBO 100 (423011-0000-000) will be aligned automatically after switching on the transformer of the illuminator.

The alignment of the HBO 100 illuminator to be aligned manually (423010-0000-000) is described below.

 If the FL attenuator (manual or motorized) is in the reflected-light path, set it to 100% transmission for illuminator alignment.

- Switch on the HBO 100 illuminator (3-47/1) via the HBO 100 transformer (4-10/2) and allow it to warm up to the operating temperature.
- Pull out adjusting aid (3-46/1) on microscope stand. The brighter arc spot of the HBO 103 W/2 lamp and its slightly darker reflected image become visible in the black-glass window of the adjusting aid.
- Turn the knurled knob (3-47/4) for collector adjustment to focus the brighter arc spot.
- Use the adjusting screws (3-47/2 and 3) to adjust the darker arc spot (reflected arc image) according to the arc spot illustration (3-46/2) within the marked alignment circle.
- Push in the adjusting aid again.

 The two arc spots of the HBO 103 W/2 lamp in the alignment circle of the adjusting aid should be closely side by side!

### 3.31.4 Installing the switching mirror for two illuminators (manual or motorized)

With the aid of a switching mirror, two illuminators can be connected to the reflected-light port of the Axio Imager stand simultaneously, e.g., a halogen illuminator (HAL 100) and a fluorescence illuminator (HBO 100).

While the manual switching mirror can be used on all Axio Imager stands, the motorized switching mirror may only be employed on the M1/M2 or Z1/Z2 stands (firmware version 01.318 or higher).

The switching lever (3-48/6) on the upper side of the manual switching mirror serves to switch manually between the two illuminators.

The motorized switching mirror can be controlled either through the operator buttons on the stand, the touch screen on the TFT display or, if a PC is connected, through the AxioVision software (Release 4.6 or higher). Control via the CAN/USB converter is not possible.

To install the switching mirror, proceed as follows:

- If necessary, remove the illuminator already mounted. To do so, grip the illuminator firmly and loosen the locking screw (3-48/2) on the reflected-light port (3-48/1), using ball-headed screwdriver SW 3.
- Insert the switching mirror (3-35/4 or 8) with the dovetail ring into the port (3-48/1). Align it (with the mounts for the two illuminators pointing to the right and left, respectively) and fix it with the locking screw (3-48/2).
- Plug the connecting cable of the motorized switching mirror into a free CAN connector on the back of the stand.
- Loosen the right locking screw and the left locking screw (3-48/5 and 7, respectively) on the switching mirror. Remove the protective caps and attach the two illuminators, e.g., HAL 100 (3-48/3) and HBO 100 (3-48/9), to the switching mirror, in the same way as if they were mounted directly onto the microscope. Then, fasten them with the relevant locking screw (3-48/5 or 7).
- Next, connect the illuminators electrically to the stand and the power supply unit, respectively.

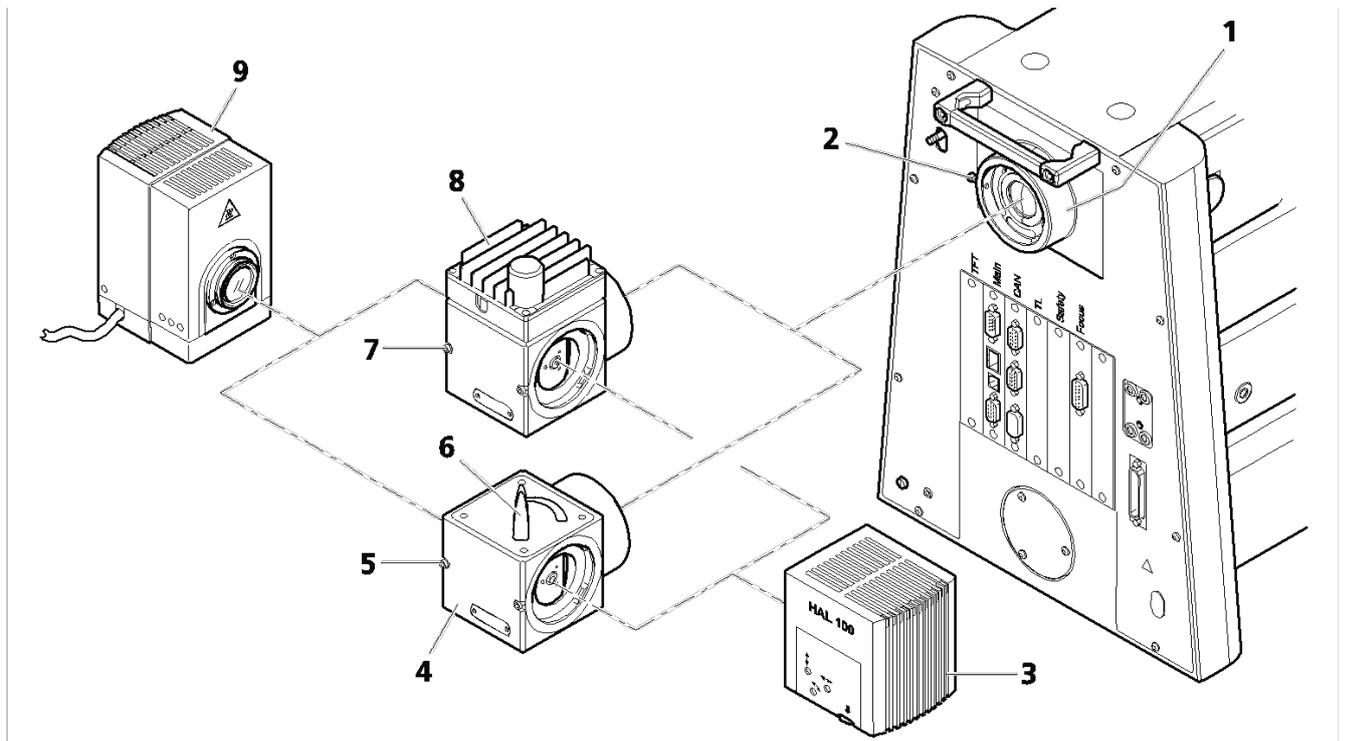


Fig. 3-48 Installing the switching mirror

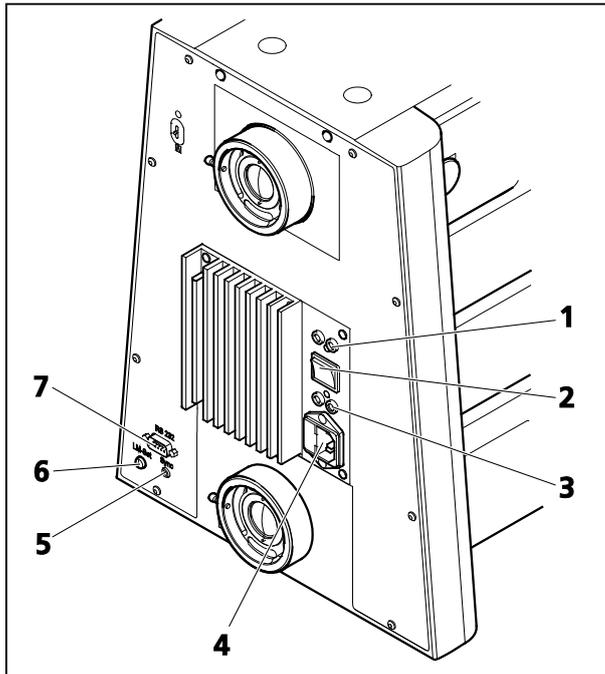


Fig. 3-49 Axio Imager, manual (rear side)

### 3.32 Electrical connections on the rear side of the microscope

#### 3.32.1 Manual stand

##### Legend to Fig. 3-49:

- 1 Connectors for reflected-light halogen illuminator
- 2 Toggle switch reflected/transmitted light (HAL)
- 3 Connectors for transmitted-light halogen illuminator
- 4 Power connector
- 5 Sync connector for camera synchronization
- 6 LM set button for light manager function
- 7 RS 232 port

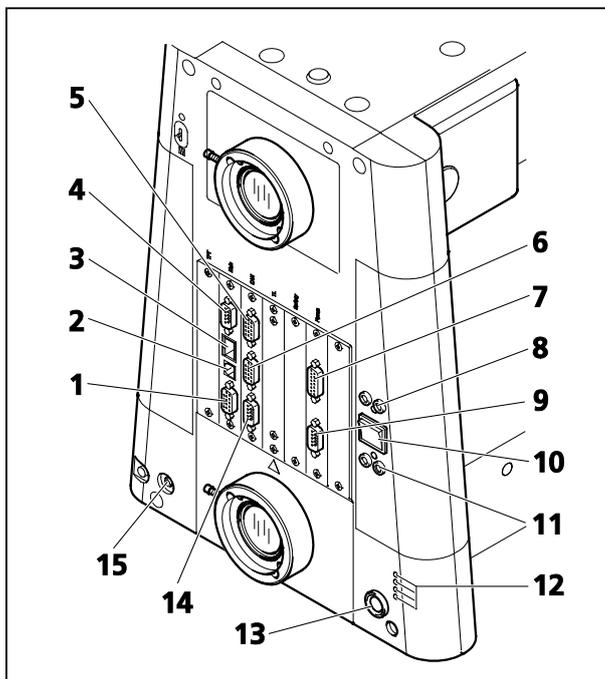


Fig. 3-50 Axio Imager, motorized (rear side)

#### 3.32.2 Motorized stand

##### Legend to Fig. 3-50:

- 1 CAN connector
- 2 USB connector
- 3 Ethernet connector
- 4 RS232 connector
- 5 CAN connector
- 6 CAN connector
- 7 Connector for linear sensor for focus adjustment
- 8 Connectors for reflected-light halogen illuminator (RL)
- 9 Connectors for transmitted-light halogen illuminator
- 10 Connector for power supply VP232-2
- 11 RS232 connector (intended for later applications)
- 12 Sync connector for camera synchronization
- 13 LM set button for light manager function

### 3.33 Changing the mechanical stage

Mechanical stages can be bolted on or turned and centered depending on their type. The range of stage travel is 75 mm in the x direction and 50 mm in the y direction. Fixed stages with drive to right or left are available. The drive is to the right on the rotary mechanical stage.

#### 3.33.1 Fixed mechanical stage 75x50 R

##### (1) Removing the stage

- Unscrew four fastening screws (3-51/3) on stage carrier (3-51/2) using the offset SW 3 Allen key (3-51/4).
- Remove stage (3-51/1) upwards from the stage carrier.

##### (2) Attaching the stage

- Place stage (3-51/1) onto stage carrier (3-51/2) in such a manner that the threaded holes on the bottom of the stage (3-51/6) are positioned above the stage carrier openings (3-51/5).
- Insert four fastening screws (3-51/3) through the stage carrier from below and screw them into the bottom of the stage; use the shorter screws for the front.
- Orient stage in the x-y direction and tighten the fastening screws.

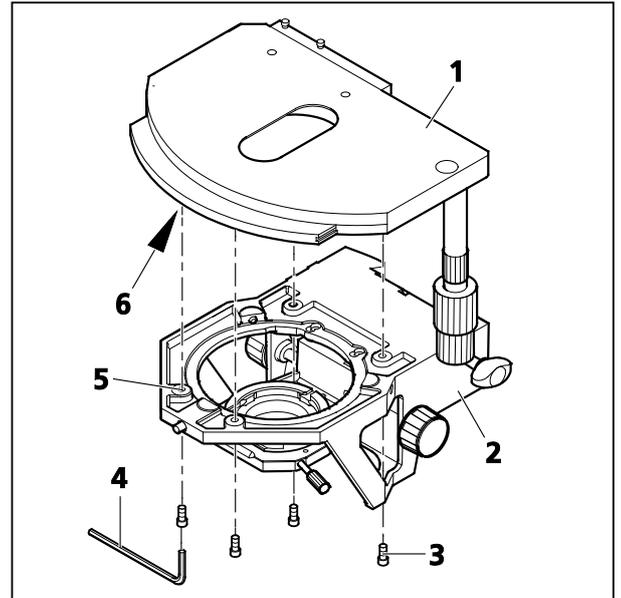


Fig. 3-51 Changing the fixed mechanical stage

#### 3.33.2 Rotary mechanical stage 75x50/240° R

##### (1) Removing the stage

- Loosen screw cap (3-52/3) from the spring housing (about three rotations).
- Turn back centering screws (3-52/1), if necessary.
- Press the stage to the front against spring pin (3-52/4), lift it off the stage carrier (3-52/2) from the back and remove it upwards.
- Retighten screw cap (3-52/3).

##### (2) Attaching the stage

- Loosen screw cap (3-52/3) from the spring housing (about three rotations).
- Place the stage with the dovetail groove on the spring pin (3-52/4).

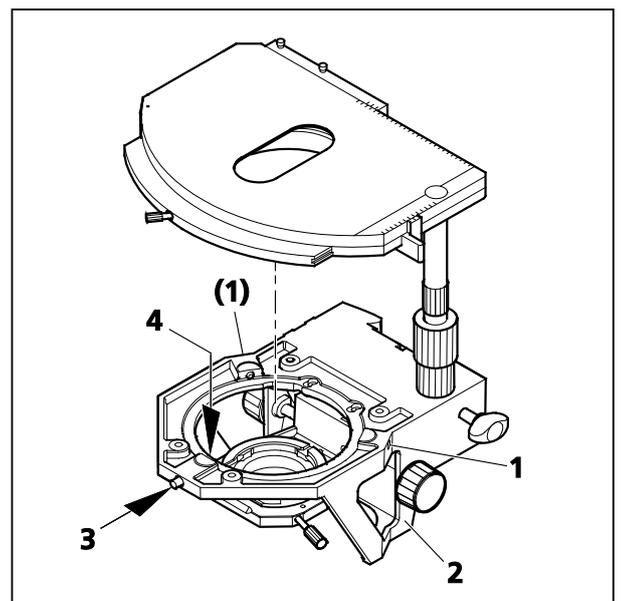
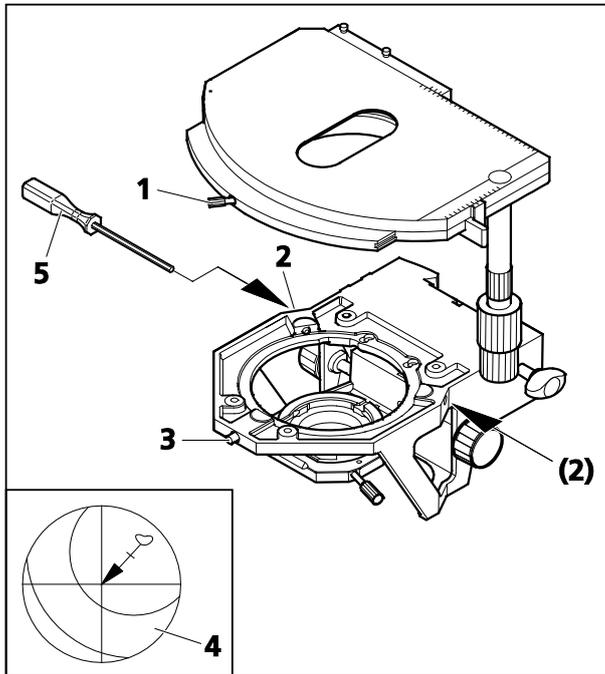


Fig. 3-52 Changing the rotary mechanical stage

- Press the stage to the front against the spring pin and lower it towards the back of stage carrier (3-52/2), and then release it.
- Retighten screw cap (3-52/3).



**Fig. 3-53** Centering the rotary mechanical stage

### (3) Centering the stage

When objectives with high magnification are used, centering can be exact for one selected objective only.

All stages are factory-precentered, i.e. a set specimen detail remains in the image center when the stage is rotated. If the image detail moves from the image center (3-53/4), while you rotate the stage, recenter the stage as follows:

- Loosen stage clamping screw (3-53/1) and the screw cap on the stage carrier (3-53/3).
- Rotate the stage to determine the maximum specimen deflection (3-53/4, tip of arrow) to the eyepiece reticle.
- Reset the two centering screws on the stage carrier (3-53/2) using one SW 1.5 Allen screwdriver (3-53/5) each to move the specimen detail by half the arrow length in the direction of the crossline center. Check whether the specimen detail moves when the stage is rotated again; repeat the procedure, when required.
- When centering is finished, tighten screw cap (3-53/3) again.

The stage can be turned through 240° up to a motion range of  $y \leq 27$  mm. No rotation is possible above this range of movement.

### 3.33.3 Adjusting the travel range on ergonomic drive

On mechanical stages with ergo-drive, the length of the **x** and **y** stage drive can be extended by **max. 15 mm** by axial movement of the drive knobs to further improve operating ease.

### 3.33.4 Removing and attaching additional sleeves

The two drive knobs are fitted with additional sleeves, providing an even finer setting of the specimen position. They can be removed if faster specimen movement is more important.

- First, loosen the two clamping screws (3-54/4) on the bottom additional sleeve (3-54/3) and remove the latter downwards, then loosen the two clamping screws (3-54/2) on the top additional sleeve (3-54/1) and pull it downward as well.
- Mount the additional sleeves back on the drive knobs in reverse order and tighten both clamping screws.

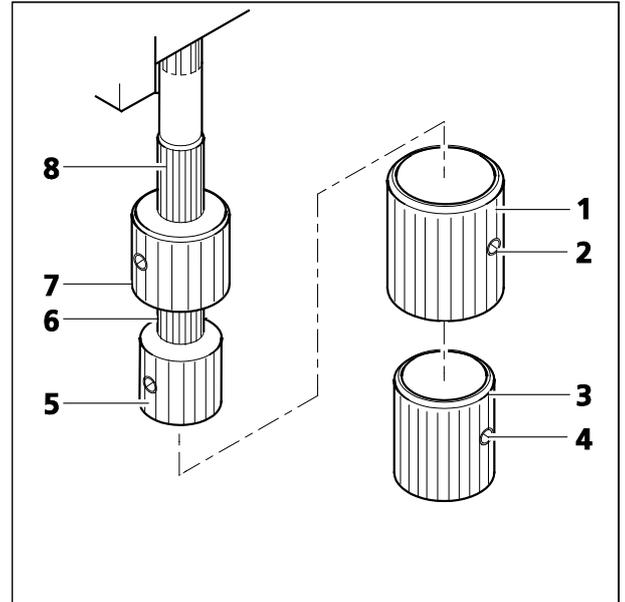


Fig. 3-54 Setting the ergonomic drive

### 3.33.5 Adjusting the smoothness (torque) of the two drive knobs of the ergonomic drive

The ergonomic drive has been factory-set to an average torque. You can change this setting as follows:

#### (1) Adjusting the x direction

- Remove the additional sleeves (3-54/1 and 3) from the drive knobs when necessary. Loosen the clamping screws to do so.
- Push the x drive knob (3-54/5) down and the y drive knob (3-54/7) up.
- Hold the x drive knob (3-54/5) and turn the light knurled ring (3-54/6) above it clockwise (easy action) or counterclockwise (sluggish action) until the desired torque is achieved.

#### (2) Adjusting the y direction

- Hold the y drive knob (3-54/7) and turn the light knurled sleeve (3-54/8) above it clockwise right (sluggish action) or counterclockwise (easy action) until the desired torque is achieved.
- Replace the additional sleeves when necessary and tighten the clamping screws.



To ensure the stage's long service life, remove the wear debris from the specimen slide at regular intervals. Make sure that the debris does not get into the guiding components for x adjustment.

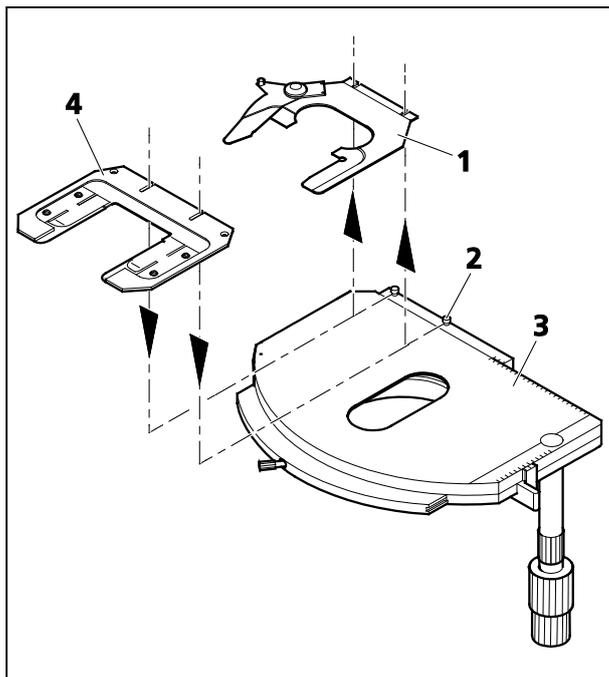


Fig. 3-55 Changing the specimen holder

### 3.33.6 Changing the specimen holder

- Loosen two knurled screws (3-55/2).
- Remove specimen holder (3-55/1) from the mechanical stage (3-55/3) to the front.
- Attach the desired specimen holder (3-55/4) to the mechanical stage with the slots under the heads of the knurled screws and tighten it by means of the knurled screws.

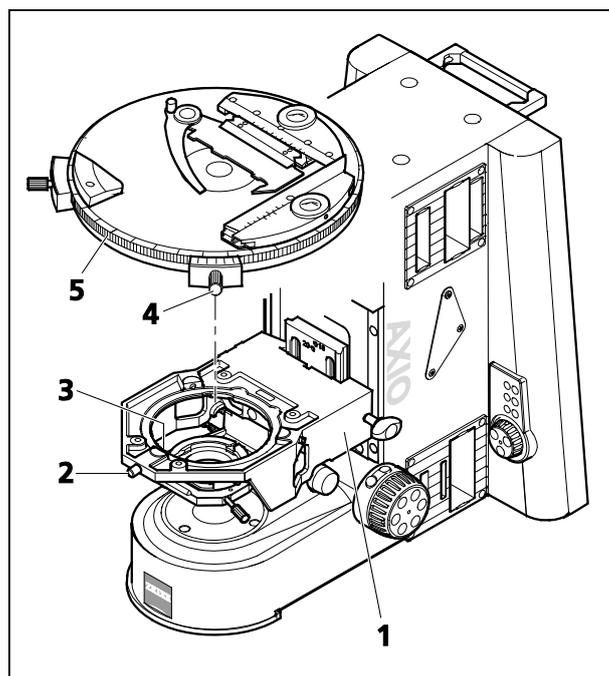


Fig. 3-56 Attaching rotary stage Pol

## 3.34 Rotary stage Pol

### 3.34.1 Removing rotary stage Pol

- Loosen screw cap (3-56/2) of spring housing with about three rotations.
- Press rotary stage Pol (3-56/5) to the front against spring-loaded pin (3-56/3), lift it off the stage carrier (3-56/1) from the back and remove it upwards.
- Retighten the screw cap (3-56/2).

### 3.34.2 Attaching rotary stage Pol

- Where necessary, loosen screw cap (3-56/2) of spring housing with about three rotations.
- Place rotary stage Pol (3-56/5) with the groove of the dovetail (stage bottom) on spring-loaded pin (3-56/3).
- Attach the rotary stage with the clamp screw (3-56/4) pointing to the front right.

- Press the rotary stage Pol to the front against the spring-loaded pin and lower it towards the back into the stage carrier (3-56/1), then release it.
- Retighten the screw cap (3-56/2).

### 3.34.3 Centering rotary stage Pol

With high-power objectives, stage centricity can be exact only for one selected objective.

The centricity will be exact for all objectives, if an objective nosepiece Pol is used.

In this case, swing the objective mount with DIC slot into the operating position.

All stages are factory-precentered, i.e. while rotating the stage the specimen feature set to the center will remain in the center. If the specimen feature moves off the center of the field of view (3-57/5), while rotating the stage, the stage should be recentered by following this procedure:

- Before centering the stage, you must adjust KÖHLER illumination on the microscope (see Section 4.9.1).
- For centering the stage, use a contrasty specimen and an eyepiece with crossline reticle.
- Loosen stage click-stop (3-57/4) or stage clamp (on rotary stage Pol with clamping device) and the screw cap of the stage carrier (3-57/1).
- Rotate the stage to determine the position of maximum offset of the specimen feature (3-57/5, origin of arrow) from the center of the eyepiece reticle.
- Turn the two centering screws on the stage carrier (3-57/2) with an SW 1.5 Allen key (3-57/3) each to move the specimen feature by half of the arrow length towards the reticle center. Rotate the stage again to check if the specimen feature moves off. Repeat the centering procedure, if necessary.
- After centering, retighten the screw cap (3-57/1).

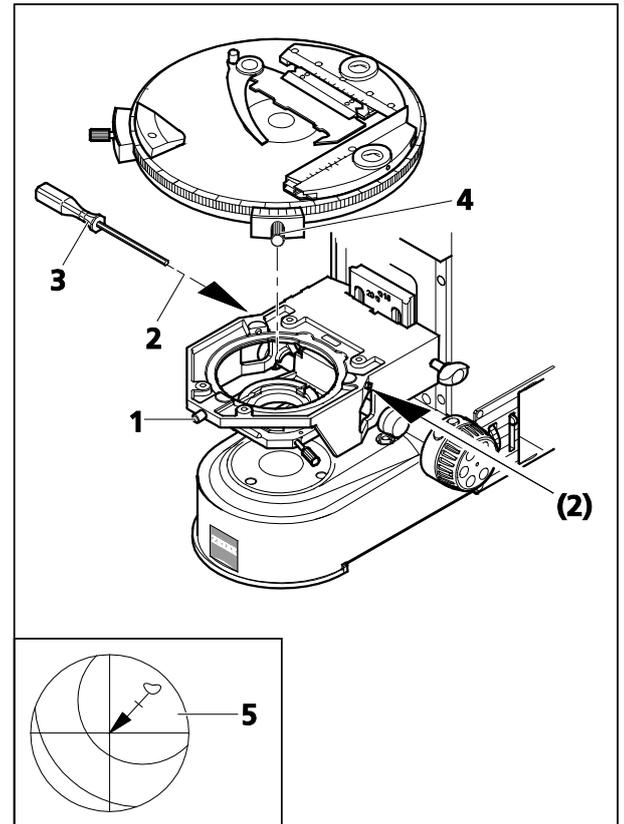
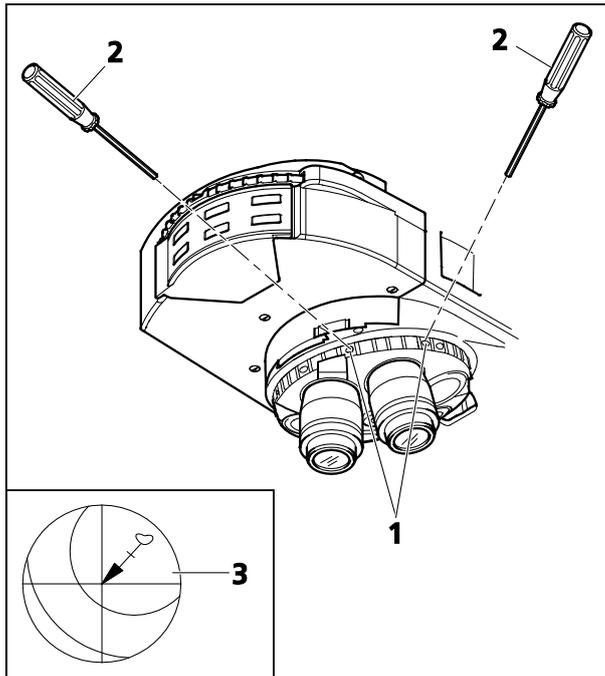


Fig. 3-57 Centering rotary stage Pol



**Fig. 3-58 Centering rotary stage and objectives**

### 3.34.4 Centering objectives

The 6-position objective nosepiece Pol of the Axio Imager is equipped with five centering objective mounts (without DIC slot) and one objective mount with DIC slot (non-centering). This mount is centered relative to the stage. Accordingly, all objectives can be centered relative to the rotary stage.

Stage centering is necessary to ensure that a specimen feature located in the center of the field of view does not move out while rotating the stage. By centering all objectives, the specimen feature remains in the center of the field of view even after changing the objective.

- Before centering the stage, you must adjust KÖHLER illumination on the microscope (see Section 4.9.1).
- To center the stage, use a contrasty specimen and an eyepiece with crossline reticle.
- First turn the nosepiece to swing the non-centering objective mount (mount with DIC slot) into the light path. Center the rotary stage for the non-centering objective mount as described under 3.34.3.

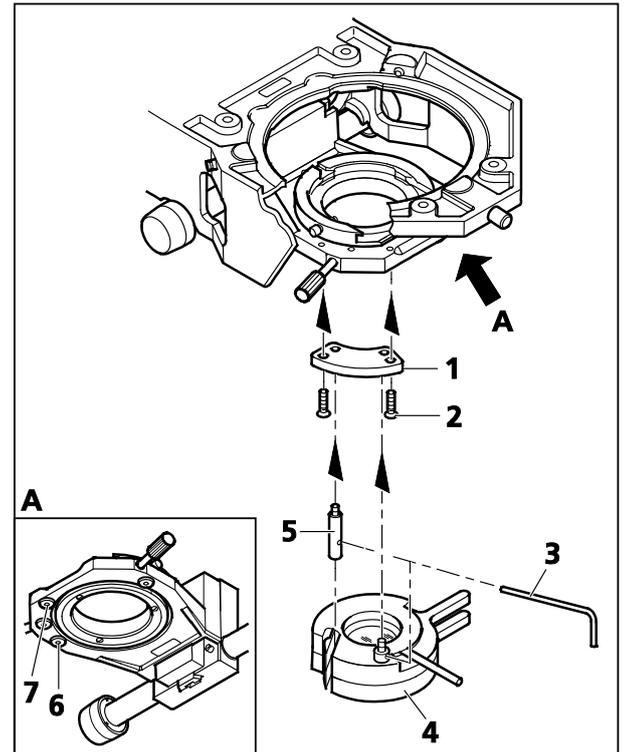
- Turn the nosepiece to move a centering objective mount into the light path.
- Determine the position of the maximum offset of the specimen feature (3-58/3, origin of arrow) from the center of the eyepiece reticle.
- Turn the two centering screws on the objective nosepiece (3-58/1) with an SW 1.5 Allen key (3-58/2) each to move the specimen feature by half of the arrow length towards the reticle center. Rotate the stage again to check if the specimen feature moves off. Repeat the centering procedure, if necessary.
- Center the other five objectives in the same manner.

 To preserve this centered state it is absolutely advisable to change the objectives only by turning the nosepiece by its knurled ring.

### 3.35 Installing Polarizer D, fixed

 If the stand is provided with a detachable stage carrier, remove it (see Section 3.13), put it aside upside down and install the polarizer from top.

- Remove any installed polarizer or color filter carrier from the condenser carrier.
- Fasten adapter plate (3-59/1) with screws (3-59/2) to the condenser carrier making use of the provided holes (3-59/6 and 7).
- Hold the polarizer (3-59/4) parallel to the bottom side of the condenser carrier. Using the offset adjusting lever (3-59/3), screw the retaining bolt of the polarizer (3-59/4) into the rear tapped hole of the adapter plate as far as it will go.
- Using the adjusting lever (3-59/3), screw the detent bolt (3-59/5) into the front tapped hole of the adapter plate as far as it will go.



**Fig. 3-59** Installing Polarizer D, fixed

For the installation of other polarizers or of the color glass holder, proceed analogously.

4 OPERATION

4.1 Coded Axio Imager operation and function controls

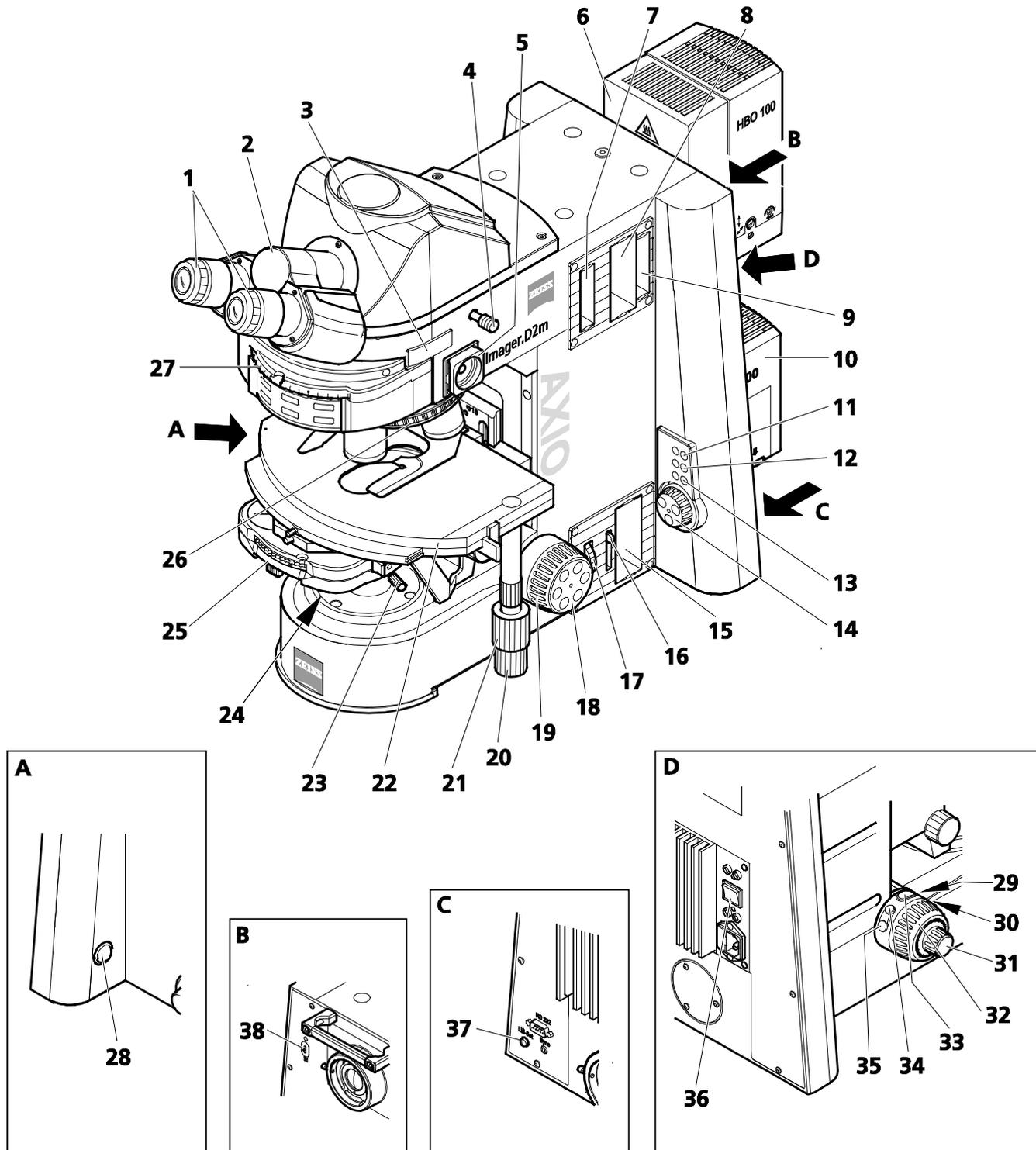


Fig. 4-1 Axio Imager operation and function controls (manual version)

**Legend to Fig. 4-1:**

- 1 Eyepieces
- 2 Binocular tube
- 3 Slot for analyzer slider (usable only, if camera path deflection, left, has not been installed)
- 4 Push-pull rod for camera path deflection, left, with interface 60N
- 5 Adjusting aid for HBO/XBO illuminators
- 6 Illuminator HBO 100 for fluorescence applications
- 7 Slot F for stop slider with centerable luminous-field diaphragm
- 8 Slot for 2-position filter wheels, discrete: The filter wheels are not suitable for fluorescence examinations
- 9 Slot A for FL attenuator, discrete
- 10 Halogen illuminator HAL 100
- 11 RL Button - Reflected-light shutter on/off with indicator LED
- 12 3200K Button - Color temperature of 3200 K on/off with indicator LED
- 13 TL Button - Transmitted-light shutter on/off with indicator LED
- 14 Light intensity control
- 15 Slot for 2-position filter wheels, discrete
- 16 Lever for transmitted-light diffusing glass
- 17 Control wheel for luminous-field diaphragm
- 18 Focusing drive - Fine focusing control, right side
- 19 Focusing drive - Coarse focusing control, right side
- 20 Control knob for X travel of mechanical stage
- 21 Control knob for Y travel of mechanical stage
- 22 Mechanical stage
- 23 Condenser carrier (refer also to Fig. 4-2)
- 24 Polarizer for transmitted light
- 25 Condenser
- 26 Objective nosepiece with objectives
- 27 Reflector turret
- 28 On/off switch
- 29 Button for transmitted-light shutter on/off (see also Section 4.8.7)
- 30 Button for reflected-light shutter on/off (see also Section 4.8.7)
- 31 Focusing drive - Fine focusing control, left side
- 32 Focusing drive - Coarse focusing control, left side
- 33 Button (not used)
- 34 Button for reducing light intensity of Halogen illuminator HAL
- 35 Button for increasing light intensity of Halogen illuminator HAL
- 36 Toggle switch for transmitted/reflected light halogen illuminator
- 37 Button LM-Set (light manager)
- 38 Lever for reflected-light diffusing glass

**Eyepieces (4-1/1)**

- All "Br. foc" eyepiece types are fitted with a control for the compensation of ametropia of the user's eyes (see Section 3.4). Besides, they permit eyepiece reticles to be mounted (see Section 2.7).

**Binocular tubes (4-1/2)**

- The binocular tubes offered permit individual setting of the interpupillary distance and of the viewing height by swiveling the eyepiece sockets (see also Sections 3.5 and 3.6) within set limits. The tubes permit – dependent on the model – the viewing height to be adjusted in a range of 50 mm. Tubes are available with viewing angles of 15°, 20° or 30°.
- Used in combination with corresponding adapters, the binocular phototubes allow the installation of a camera of the user's choice. The optical path is selected by means of a push-pull rod located at the right side of the tube having two or three operating positions. The binocular phototube 30°/25 (425502-0000-000 and 425501-0000-000) additionally features an eyepiece shutter that is operated via a second push-pull rod on the left side or via a button on the right-hand side (425506-0000-000, motorized eyepiece shutter).

**Slot for analyzer slider (4-1/3)**

- For the accommodation of an analyzer slider or Bertrand lens slider PH (453671-0000-000).
- If the camera path selector mirror has been installed, this slot cannot be used.

**Push-pull rod for camera path deflection, left (4-1/4)**

- Switching the optical path to the camera path deflection, left, with interface 60N.
- The camera path selector may optionally be equipped with 50% beam splitter or with 100% reflecting mirror.
- Push-pull rod pushed in: 100% observation through eyepieces
- Push-pull rod pulled out: 50% : 50% Eyepiece/camera path (with beam splitter) or 100% camera path (with reflecting mirror)

**Adjusting aid for HBO/XBO illuminators (4-1/5)**

- Serves for viewing arc spot and reflected arc spot image in lamp alignment.
- Adjusting position (Adjust): Adjusting aid pulled out up to the stop

**Slot F for stop slider (4-1/7) with centerable luminous-field diaphragm**

- With the stop slider with centerable luminous-field diaphragm inserted in slot F, you can center the luminous-field diaphragm and adjust its diameter in the reflected-light path.
- When inserting the stop slider, the clamping spring points upward.
- Adjustment of the diameter is via the knurled wheel, centering is performed by means of the two centering screws of the stop slider.
- To remove the luminous-field diaphragm slider from the slot, put a ball-headed screwdriver into the free hole on the stop slider, slightly cant the screwdriver and pull the stop slider out.

**Slot for 2-position filter wheels, discrete (4-1/8)**

- When using the HAL 100 halogen illuminator in the reflected-light path, the 2-position filter wheels, discrete, can be used for light-intensity adjustment. The unit contains four filters each (neutral-density) mounted on two filter wheels.

**CAUTION**

Neutral-density filters are not suited for fluorescence examinations, as they might be damaged.

- When inserting the filter wheels, the clamping spring points upward.
- To remove the filter wheel, introduce a screwdriver into the top hole, slightly cant it and pull the filter wheel out of its receptacle.
- The positions of the two filter wheels are labeled accordingly and set by turning the knurled wheels. The filters can be combined in any way.

**Slot for FL attenuator, discrete (4-1/9)**

- Use the FL attenuator, discrete, to attenuate the light in the fluorescence path when using the HBO 100.
- When the FL attenuator is used, the clamping spring points upward.
- To remove the FL attenuator, introduce a screwdriver into the top hole, slightly cant it and pull the FL attenuator out of its receptacle.
- The FL attenuator has six labeled positions that are set by turning the knurled wheel.

**RL button – Reflected-light shutter on/off (4-1/11) with indicator LED**

- Alternately opens or blocks the reflected-light path; function corresponds to 4-1/30.
- The indicator LED lights, when the shutter is opened.

**3200K button – Color temperature 3200 K on/off (4-1/12) with indicator LED**

- Controls the connected halogen illuminator to the voltage value that corresponds to a color temperature of 3200 K. This color temperature is needed for taking color photos on the microscope.
- The indicator LED lights, when the color temperature of 3200 K is set.

**TL button – Transmitted-light shutter on/off (4-1/13)**

- Alternately opens or blocks the transmitted-light path; function corresponds to 4-1/29.
- The indicator LED lights, when the shutter is opened.

**Light-intensity control (4-1/14)**

- Used to adjust the d.c. voltage supply of the halogen illuminator in the range of approx. 1.8 to 12 V; function corresponds to 4-1/34 and 35.
- The annularly arranged LED's indicate the set voltage in 15 stages.

**Slot for 2-position filter wheels, discrete (4-1/15)**

- When using the HAL 100 halogen illuminator, the 2-position filter wheels, discrete, can be used for light-intensity adjustment. The unit contains four filters each (neutral-density) mounted on two filter wheels.
- When inserting the filter wheels, the clamping spring points upward.
- To remove the filter wheel, introduce a screwdriver into the top hole, slightly cant it and pull the filter wheel out of its receptacle.
- The positions of the two filter wheels are labeled accordingly and set by turning the knurled wheels. The filters can be combined any way.

**Lever for transmitted-light diffusing glass (4-1/16)**

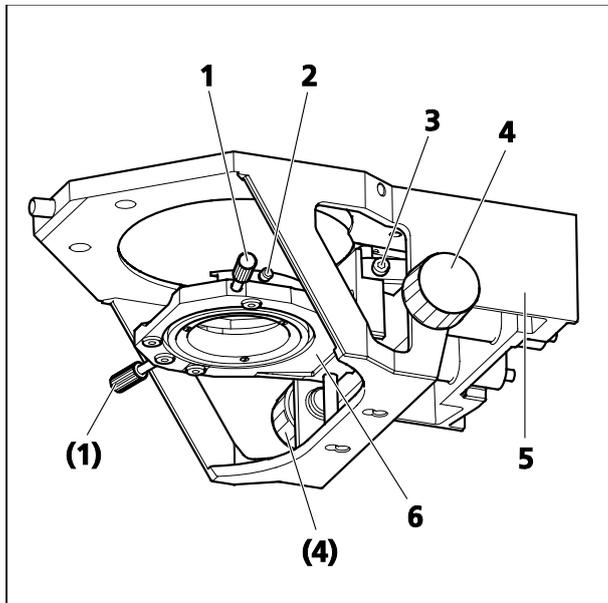
- Lever in top position: Diffusing glass in light path
- Lever in bottom position: Diffusing glass not in light path

**Control wheel for luminous-field diaphragm (4-1/17)**

- Control wheel for continuous adjustment of the luminous-field diaphragm (transmitted light)

**Focusing drive – Fine focusing control (4-1/18), right-hand side**

- Control for fine focusing
- 1 revolution of fine focusing control = 0.1 revolution of coarse-focusing control

**Fig. 4-2 Condenser carrier****Focusing drive – Coarse focusing control (4-1/19), right-hand side**

- Control for coarse focusing
- 1 revolution of coarse focusing control = approx. 2 mm
- Total focusing range: approx. 24 mm

**Condenser carrier (4-1/23)**

- The condenser carrier (4-2/6) is part of the stage carrier (4-2/5).
- Insert the condenser in the mount of the condenser carrier and fasten it by means of clamping screw (4-2/2).
- To center the condenser, turn the two centering screws (4-2/1).
- Turn the vertical control (4-2/4) to adjust the condenser (condenser carrier) in the Z-axis.
- The clamping screw (4-2/3) serves as stop for the vertical adjusting range of the condenser. The stop facilitates finding the KÖHLER illumination position again.

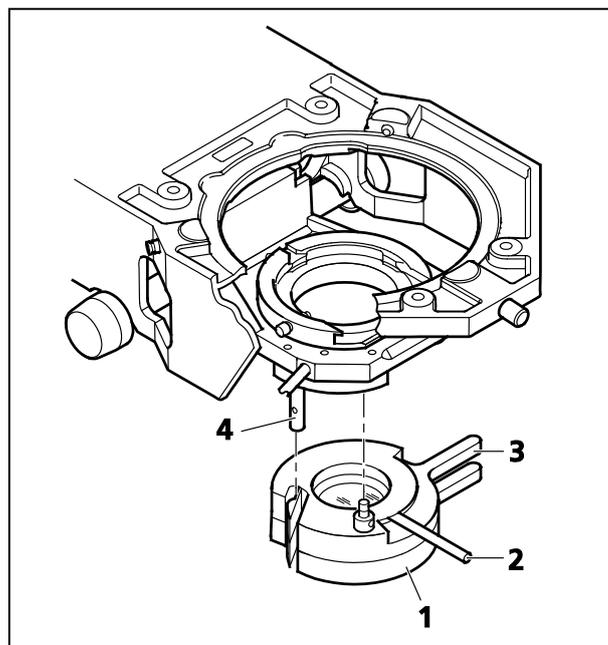
**Polarizer for transmitted light (4-1/24)**

Various types of transmitted-light polarizers are available: with wixed or rotatable polarizer or with additional lambda/4 plate or with additional neutral-density filter position (cf. System Overview of Axio Imager, Section 2.4).

- Using handle (4-3/3) swing the polarizer (4-3/1) (or the neutral-density filter, if necessary) into the light path until it is reliably fixed by the detent bolt (4-3/4).
- If available, rotate the lambda plate or the lambda/4 plate by maximally 45 degrees to the right or left by moving handle (4-3/2).



The color filter carrier is operated analogously to the polarizer.

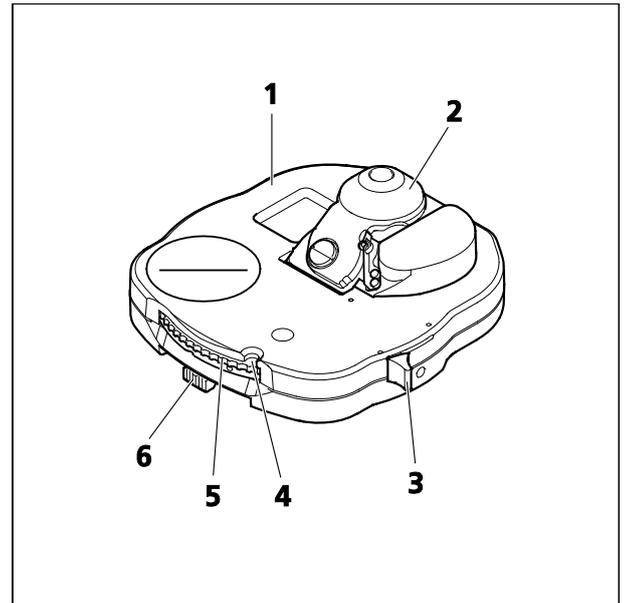
**Fig. 4-3 Polarizer for transmitted light**

**Condenser (4-1/25)**

Dependent on the model, the achromatic-aplanatic universal condenser (4-4/1) is equipped with:

- Swivel-type front lens
- Turret disk for:
  - Brightfield - **H**
  - Darkfield - **D**
  - Phase contrast - Ph **1**, Ph **2**, Ph **3**
  - Interference contrast - DIC **I**, **II**, **III**
- Aperture diaphragm (iris diaphragm)

The front lens (4-4/2) is swiveled in/out with lever (4-4/3). By turning turret disk (4-4/5) the brightfield insert and the contrasting stops are moved into the light path. In window (4-4/4), the ID of the set turret position (e.g. **D**) is visible. Using sliding control (4-4/6), you can open or close the aperture diaphragm.



**Fig. 4-4** Achromatic-aplanatic universal condenser 0.9 H D Ph DIC

**Objective nosepiece (4-1/26)**

- Dependent on the model, the nosepiece is equipped with six or seven M27 objective mounts with slot for DIC slider or seven M27 objective mounts.
- Objectives are quickly changed (objective position) by turning at the knurled ring of the objective nosepiece.

**Reflector turret (4-1/27)**

- With six mounts for replaceable P&C reflector modules or P&C analyzer modules
- Reflector modules (reflector positions) are quickly changed by turning at the knurled ring of the reflector turret.
- Active turret position is indicated in window.

**On/off switch (4-1/28)**

- Position O = Instrument switched off.
- Position I = Instrument switched on.

**On/off button for transmitted-light shutter (4-1/29)**

- Alternately opens or blocks the transmitted-light path.

**On/off button for reflected-light shutter (4-1/30)**

- Alternately opens or blocks the reflected-light path.

**Focusing drive – Fine focusing control (4-1/31), left-hand side**

- Control for fine focusing

**Focusing drive – Coarse focusing control (4-1/32), left-hand side**

- Control for coarse focusing

**Intensity button (4-1/34)**

- Reduces the light intensity of halogen illuminator HAL.

**Intensity button (4-1/35)**

- Increases the light intensity of halogen illuminator HAL.

**Toggle switch for transmitted/reflected-light halogen illuminator (4-1/36)**

- Alternately switches the halogen illuminator for transmitted light or reflected light on/off.
- Toggle switch up: Reflected-light halogen illuminator ON (transmitted-light OFF)
- Toggle switch down: Transmitted-light halogen illuminator ON (reflected-light OFF)

**Button LM-Set (4-1/37)**

- Saves the values of the current light intensity and the optical path nosepiece position (brief beep).
- Adaptation of the luminosity of the ring-like arranged LEDs at the light intensity control:
  - Switch on the illuminator and adjust light intensity at the control so that all LEDs light up.
  - Maintain **LM-Set** key pressed down. You hear a long beep tone.
  - Adjust the desired light intensity using the light intensity control (4-1/14).
  - Release the **LM-Set** key to save the light intensity adjusted.

**Lever for reflected-light diffusing glass (4-1/38)**

- Lever in top position: Diffusing glass in light path
- Lever in bottom position: Diffusing glass removed from light path



## 4.2 Axio Imager operation and function controls (motorized version)

In this section, those operation and function controls of the motorized version of the Axio Imager are described that are different from the manual version. Controls not described here correspond to the manual version.

As a great number of components are offered optionally, the specific design of your microscope may be different from the illustrations below. So it is possible, for instance, to use also manually controlled components (such as reflector turret, condensers, filter wheels) on the motorized stand. In this case, however, the functionality of the microscope, particularly its operation via TFT display (touchscreen) will be restricted.

### Legend to Fig. 4-5:

- 1 Binocular phototube with motorized eyepiece shutter – Button for opening/closing the eyepiece shutter
- 2 Camera path deflection left, with interface 60N (built into stand) - Operation via TFT display (touchscreen)
- 3 Slot F for stop slider with centerable luminous-field diaphragm
- 4 Slot for 2-position filter wheels, discrete mot.: The filter wheels are not suitable for fluorescence applications.
- 5 Slot A for FL attenuator, discrete mot.
- 6 TFT display (touchscreen) for operation and adjustment of microscope (refer to Section 4.8)
- 7 Slot for 2-position filter wheels, discrete mot.
- 8 Motorized focusing drive, coarse/fine
- 9 Motorized objective nosepiece
- 10 Motorized condenser
- 11 Motorized reflector turret - Operation via TFT display (touchscreen)
- 12 Button for opening the luminous-field diaphragm
- 13 Button for closing the luminous-field diaphragm
- 14 Button for quick-action lowering of stage LOAD POSITION
- 15 Button for quick-action lifting of stage OPERATING POSITION
- 16 Button (setting depends on stand type)
- 17 Button (setting depends on stand type)
- 18 Button (setting depends on stand type)
- 19 Button for rotating nosepiece anticlockwise by one position (factory-set)
- 20 Button for rotating nosepiece clockwise by one position (factory-set)
- 21 Button for quick-action stage lifting OPERATING POSITION
- 22 Button for quick-action stage lowering LOAD POSITION
- 23 Button (not used by factory setting)
- 24 Button (not used by factory setting)
- 25 Button (not used by factory setting)
- 26 Button for turning reflector turret clockwise by one position (factory-set)
- 27 Button for turning reflector turret anticlockwise by one position (factory-set)
- 28 Button LM-Set

 The assignment of keys 10 ... 23 can be changed individually on the TFT display (see Section 4.8: Touchscreen - **Settings tab**).

Further motorized components (such as the motorized stop slider, motorized filter wheels, motorized FL attenuator and the motorized tube lens turret) are to be operated via the buttons directly arranged on the respective components.

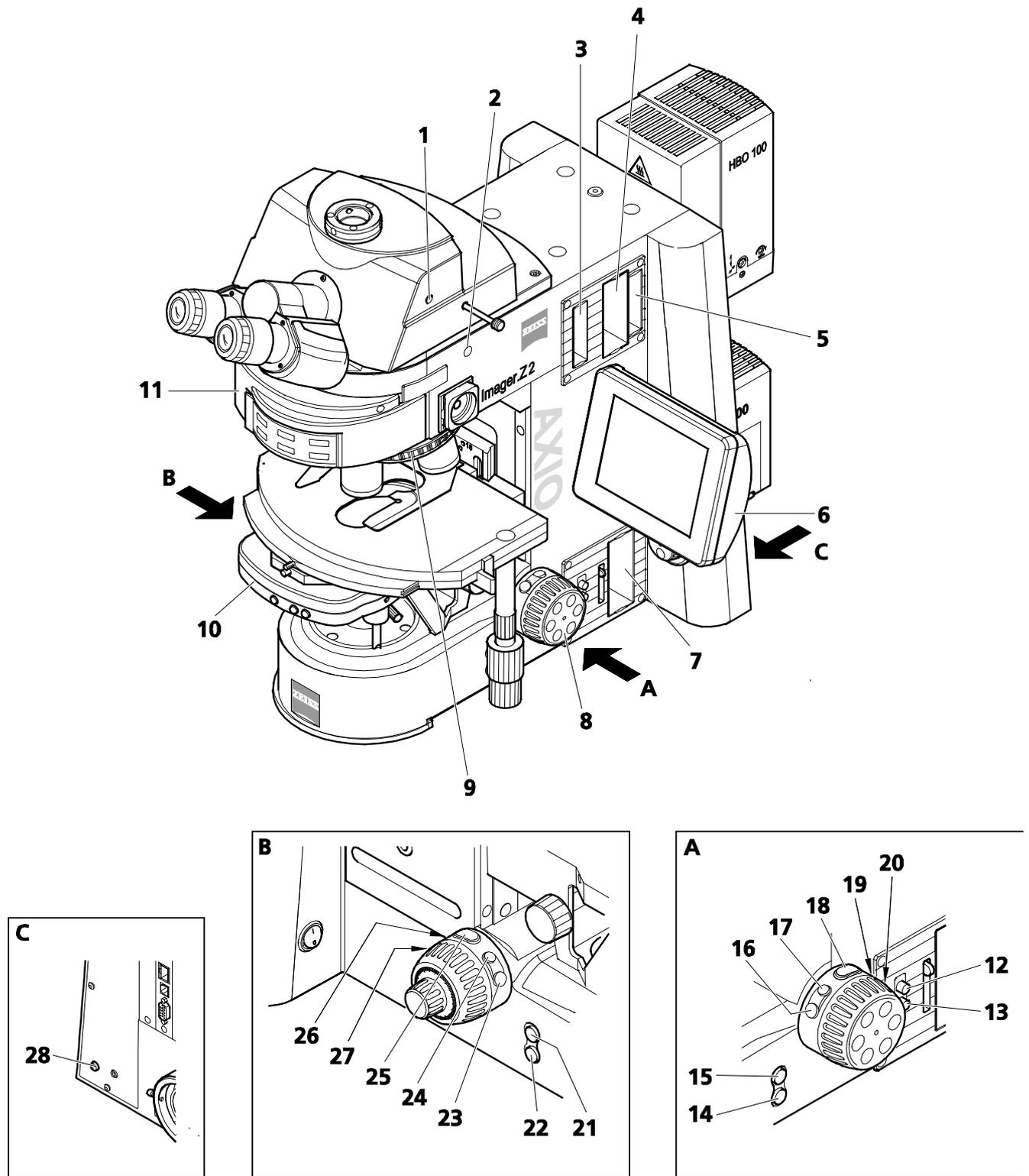


Fig. 4-5 Axio Imager operation and function controls (motorized version)

### Binocular phototube with motorized eyepiece shutter (4-5/1)

- In addition to the manually operated beam splitter, the binocular phototube with motorized eyepiece shutter 30°/25 (425506-0000-000) is equipped with a motorized eyepiece shutter, which is operated via the button on the right side (alternately on/off) or via the TFT display.

### Motorized camera path deflection, left (4-5/2)

- The motorized camera path deflection (100:0/50:50) is exclusively operated via the TFT display.

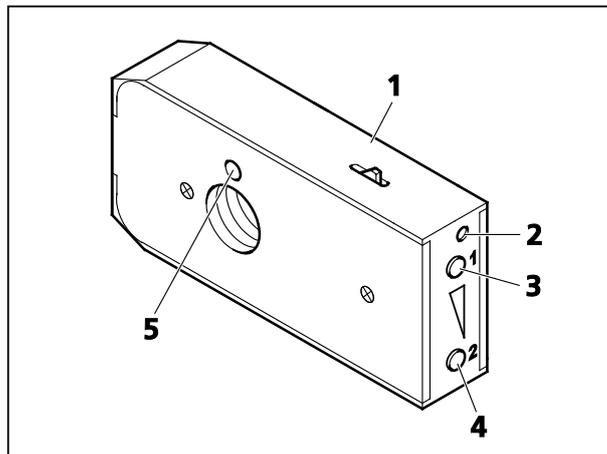


#### CAUTION

Switch off the microscope before inserting stop slider mot., filter wheel 2x mot. and FL attenuator discrete mot. into the corresponding slots.

### Slot F for stop slider mot. (4-5/3) with centerable luminous-field diaphragm

- The motorized stop slider is to be inserted analogously to the motorized filter wheel 2x discrete mot. and the FL attenuator discrete mot.
- The diaphragm is opened or closed by pushing the respective button on the slider.



**Fig. 4-6** Inserting/removing filter wheel 2x discrete mot.

### Slot for 2-position filter wheel, discrete mot. for reflected light (4-5/4) and transmitted light (4-5/7)

- When using the HAL 100 halogen illuminator in the reflected-light and the transmitted-light path, the filter wheel 2x discrete mot. can be used for brightness adjustment. This filter wheel module contains four filters each (neutral-density filters) on two filter wheels.



#### Caution

Neutral-density filters are not suitable for fluorescence applications as they may be damaged by the light used.

- Before inserting the filter wheel module, first activate the click-stop mechanism (4-6/1) by pushing in. At the right-hand front end (4-6/2) a small silver plate becomes visible. Then, push the filter wheel module into the corresponding slot until you hear it click into place.
- To remove the filter wheel module, introduce a screwdriver into the top hole (4-6/2) and push in the silver plate to deactivate the click-stop mechanism. Slightly cant the screwdriver in the hole and pull the filter wheel module out of the slot.
- Adjust the desired positions of the filter wheels by pushing the top (4-6/3) or the bottom button (4-6/4). When the filter wheel module has been removed, the selected transmission can be read from the sight glass (4-6/5). The positions of the two integrated filter wheels can be combined optionally with each another.

### FL attenuator discrete mot. for reflected light (4-5/5)

- The FL attenuator discrete should be used to attenuate the fluorescence path when using the HBO 100.
- Before inserting the FL attenuator, first activate the click-stop mechanism (4-7/1) by pushing in. At the right-hand front end (4-7/2) a small silver plate becomes visible. Then, push the FL attenuator into the corresponding slot until you hear it click into place..
- To remove the FL attenuator, introduce a screwdriver into the top hole (4-7/2) and push in the silver plate to deactivate the click-stop mechanism. Slightly cant the screwdriver in the hole and pull the FL attenuator out of the slot.
- The FL attenuator has six positions that can be set by pushing buttons (4-7/3) or (4-7/4) in forward or return run.

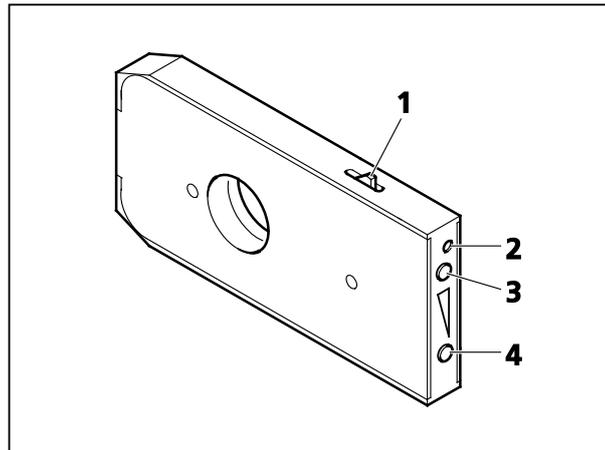


Fig. 4-7 Inserting/removing the FL attenuator mot.

### Motorized focusing drive coarse/fine (4-5/8), on both sides of stand

- Operation is performed manually by turning the coarse or fine focusing controls (on both sides of stand), which act on an electric encoder or by AxioVision 4.3 software.

### Motorized universal condenser (4-5/10)

- Adjustment of aperture diaphragm via buttons (4-8/2 - open) and (4-8/3 - close)
- Swiveling in/out of the front lens (4-8/1) with button (4-8/4)
- Rotating the condenser turret clockwise by means of button (4-8/5), anticlockwise by means of button (4-8/6)

### Button for quick-action stage lowering to LOAD POSITION (4-5/14 or 22)

- Upon actuation of this button, the stage will be lowered somewhat to move it out of the focus position. The current focus position will be saved.
- The specimen can be changed.



As soon as you turn the motorized focusing drive (4-5/4) in the load position, the saved operating position shall be deleted and the current position saved instead.

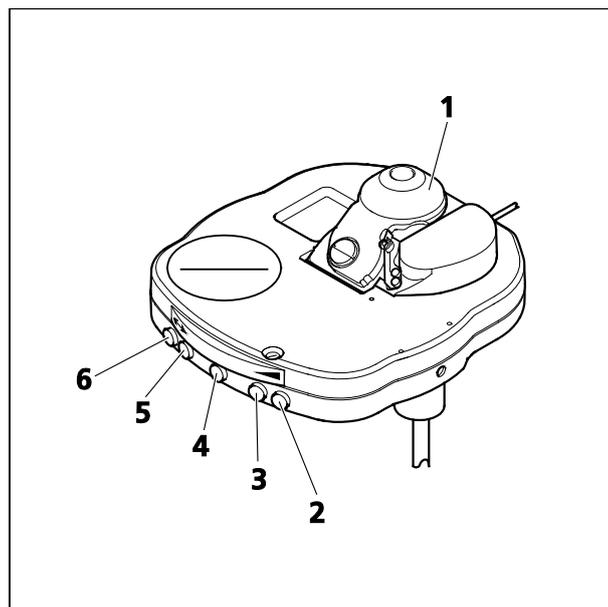
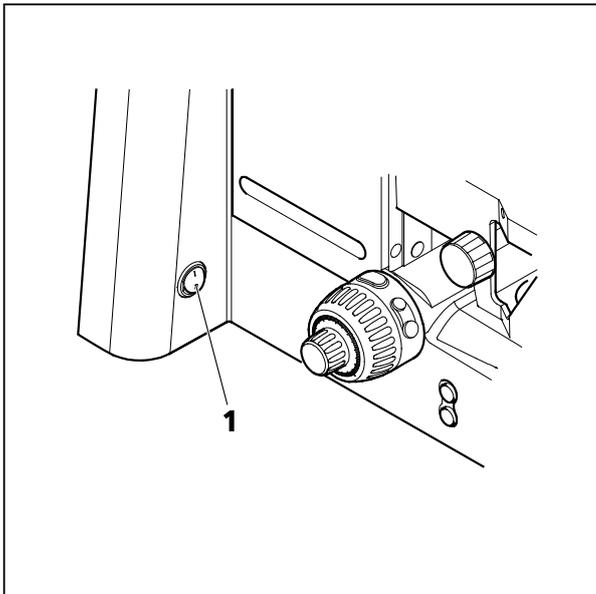


Fig. 4-8 Achromatic-aplanatic universal condenser, mot.

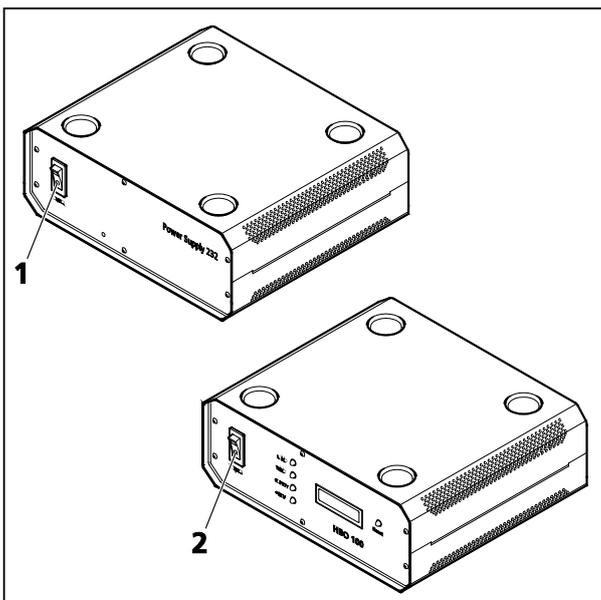
**Button for quick-action stage lifting to OPERATING POSITION (4-5/15 or 21)**

– On activation of this button, the stage will be moved into the focus position saved last.

 You can abort the automatic travel of the stage to the load position/operating position by pressing the button again or by pressing the **STOP** button on the TFT display.



**Fig. 4-9** Switching the manual microscope on/off



**Fig. 4-10** Switching motorized microscope and HBO 100 illuminator on/off

### 4.3 Switching microscope and HAL 100 illuminator on/off

#### 4.3.1.1 Switching on

- Remove the dust covers from instrument.
- Switch on the manual version of the microscope at the on/off (4-9/1) switch located at the left side of the stand. With the motorized microscope, first switch on the power supply VP232-2 at its on/off switch (4-10/1), and then operate the on/off switch on the stand (Position **I**).
- Adjust the desired light intensity using the light intensity control.
- If you installed one HAL 100 halogen illuminator each for reflected and for transmitted light, use toggle switch (4-1/36) to switch to the desired illuminator (reflected light: toggle switch up; transmitted light: toggle switch down).

 The functioning of the light manager depends on the setting of the toggle switch.

#### 4.3.1.2 Switching off

- After use, switch the manual and the motorized microscope off by means of the on/off switch (4-9/1) on the left side of the stand. Then, on the motorized version, switch off the power supply VP232-2 at the on/off switch (4-10/1) (Position **O**).
- Cover the instrument with the dust covers.

#### 4.4 Switching the HBO 100 on/off

- The HBO 100 illuminator, which is used in place of the HAL 100 illuminator for fluorescence contrast examinations, is to be switched on and off at the on/off switch (4-10/2) of the HBO 100 transformer.
- After any illuminator switch off, allow for a cool down of approx. 15 minutes before switching on the illuminator again. Failure to do so will unnecessarily shorten the service life of the mercury vapor short-arc lamp.

#### 4.5 Binocular phototube 30°/25 mot. with two camera ports (2TV tube mot.)

The 2TV tube mot. (4-11) is available for all Axio Imager stands of type .M2 / .M2m and .Z2 / .Z2m.

Like all other binocular tubes this tube is to be mounted to the coupler plate and connected to the stand via the CAN bus. To this end, connect the cable to one of the three available CAN connectors (Fig. 3-50).

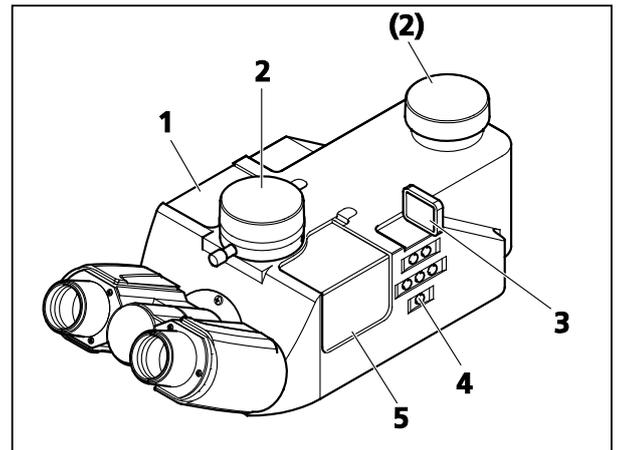
The 2TV tube has two TV ports (4-11/2).

The front TV port is adjustable in the X, Y and Z axis and to the rear (fixed) TV port.

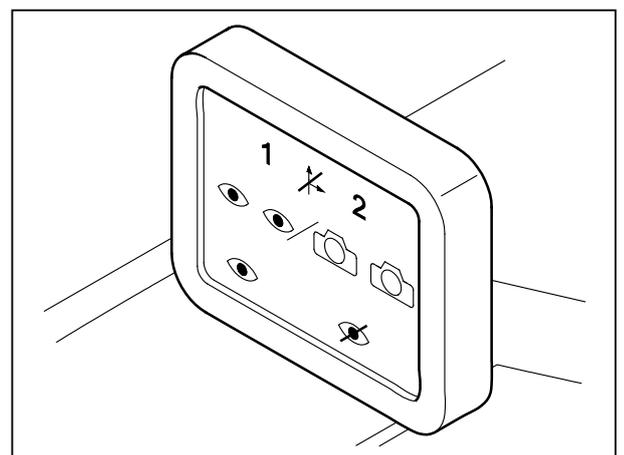
The tube is configured via the TFT display or the MTB 2004.

LEDs on the tube display (4-11/3, 4-12) indicate the selected light path setting.

If you find the illumination of the display disturbing, you can switch it off by pushing the eyepiece shutter button (4-13/1) for three seconds.



**Fig. 4-11** Binocular phototube 30°/25 mot. with two camera ports (2TV tube mot.)



**Fig. 4-12** Tube display

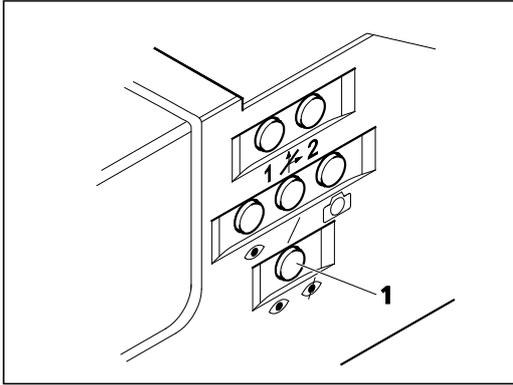


Fig. 4-13 Control panel

You can set the light path and the eyepiece shutter via the buttons of the control panel (4-11/4, 4-13/1) on the right side of the tube.

If you activated the dazzle protection via the TFT display (see Section 4.8.6.2), you can use the eyepiece shutter instead of the light path shutter.

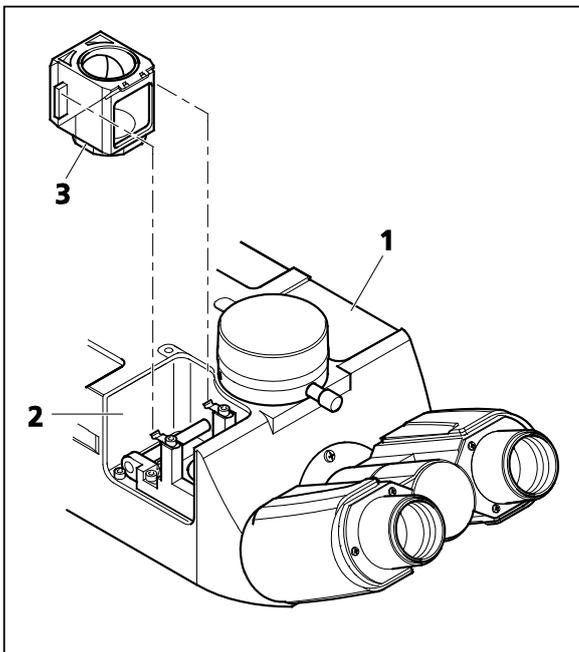


Fig. 4-14 Receptacles for P&C modules

In addition, the 2TV tube has two receptacles (4-14/1 and 2), into which P&C modules (4-14/3) can be inserted.

One of these receptacles is factory-fitted with a 100% mirror, allowing 100% of the light to be directed to either the front or the rear TV port.

Two receptacles for P&C modules are available to the user to install, for instance, an additional beam splitter for dual camera operation.

- To insert P&C modules into the 2TV tube, remove the corresponding cover (4-11/1 and 5) held magnetically by lifting them off.
- Insert the P&C module(s) and reattach the cover(s).

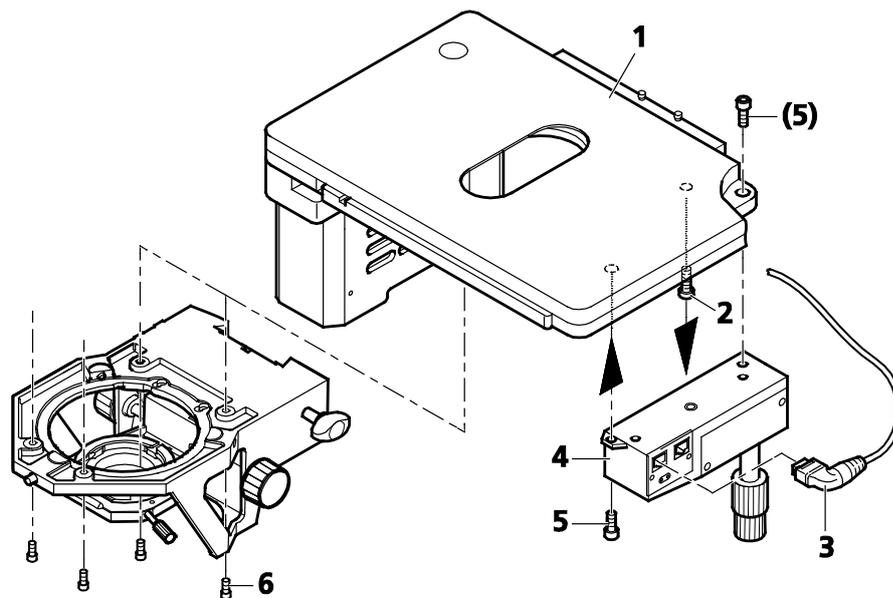
## 4.6 Mechanical Stage 75x50 mot. CAN

### 4.6.1 Assembling Mechanical Stage 75x50 mot. CAN



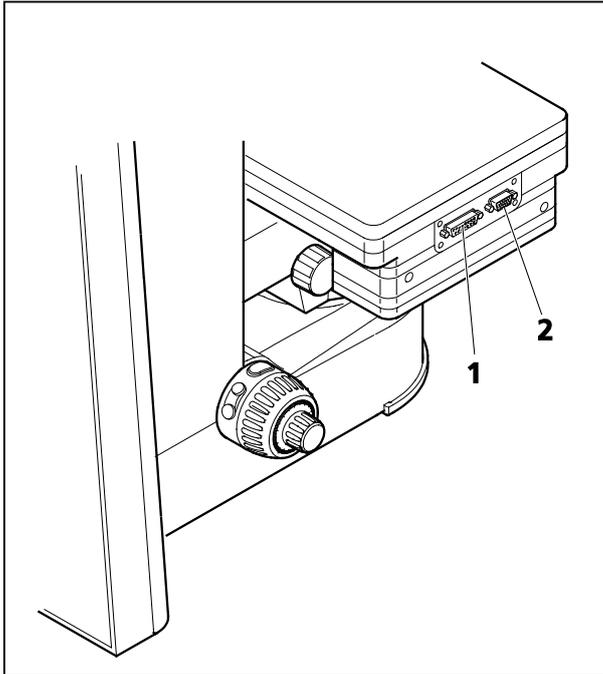
- While assembling the stage, take care that you do not disconnect any existing cable connections from the stage motors.  
Mixing up of plugs at the stage motors may lead to the destruction of the integrated measuring systems!
- After you have unpacked the mechanical stage, **remove the transport lock (4-15/2)** on the underside of the mechanical stage.

- Slide the stage plate (4-15/1) forward and fasten coaxial drive (4-15/4) to the underside of the stage using two screws (4-15/5).
- Plug angled CAN bus connector (4-15/3) into one of the two sockets on the coaxial drive.
- Place mechanical stage (4-15/1) onto stage carrier in a way that the holes in the underside of the mechanical stage coincide with the through holes of the stage carrier.
- Using the angled Allen key (SW 3), screw four screws (4-15/6) from the bottom into the underside of the stage with the shorter screws being inserted in the front holes.



- 1 Mechanical stage 75x 50 mot. CAN
- 2 Transport lock (screw)
- 3 Angled CAN bus connector (100-0600-144)
- 4 Electronic coaxial drive CAN
- 5 Fastening screws on coaxial drive
- 6 Fastening screws on stage carrier

**Fig. 4-15 Assembling Mechanical Stage 75x50 mot. CAN**



**Fig. 4-16 Connecting Mechanical Stage 75x50 mot. CAN (Version 2 and mot. standard)**

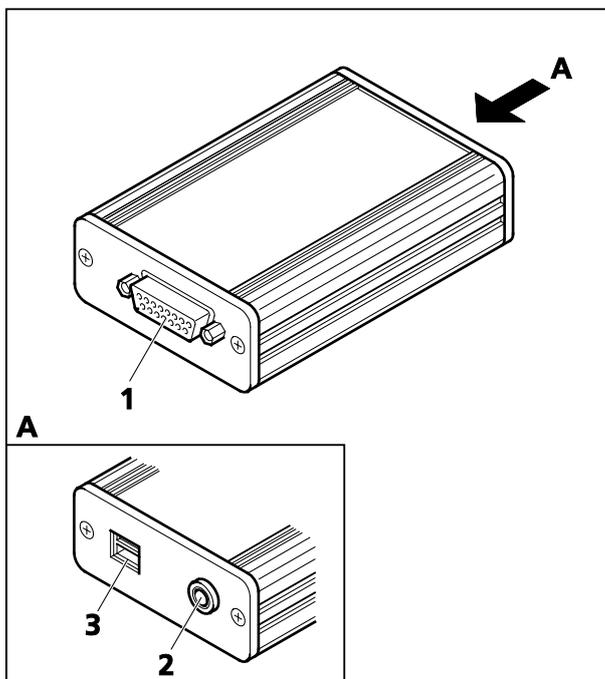
#### 4.6.2 Connecting Mechanical Stage 75x50 mot. CAN

The connector panel of the Mechanical Stage 75x50 mot. CAN (Version 2 and mot. standard) is located on the left-hand side.

- Using the CAN bus cable, connect the CAN connector of the mechanical stage (4-16/2) with the CAN connector on the stand (back panel).
- Plug the connecting plug of the connection cable of the coaxial drive to connector (4-16/1) of the mechanical stage.

The motorized stages can be directly connected to stands .M2 / .M2m / .Z2 / .Z2m through the CAN bus. In this case, the speed of stage travel can be adjusted via the TFT display according to the magnification of the selected objective (see Section 4.8.6.1).

If the objectives are specified when you switch on the stand (.M2 / .M2m / .Z2 / .Z2m), the optimum speed of stage travel is calculated directly. However, you may change (and store) this speed, if necessary.



**Fig. 4-17 CAN/USB converter**

#### 4.6.3 CAN/USB converter

On manual stands, CAN bus stages (432024-9903-000 or 432026-9000-000) can be directly connected to the PC via the CAN/USB converter.

Unlike motorized stands, the speed of stage travel cannot be adjusted depending on magnification.

- Use the CAN bus cable to connect the CAN connector of the mechanical stage (4-16/1) with the CAN connector of the CAN/USB converter (4-17/1).
- Use the USB cable to connect the USB-connector of the PC with the CAN/USB converter (4-17/3).
- Connect the DC voltage input terminal of the CAN / USB converter (4-17/2, 24 V DC / 1.25 A) to the plug-type power supply output (Output 24 V DC, 1.25 A). Then plug that power supply into the power outlet.



The CAN / USB converter may only be operated with the power supply unit provided by the manufacturer.

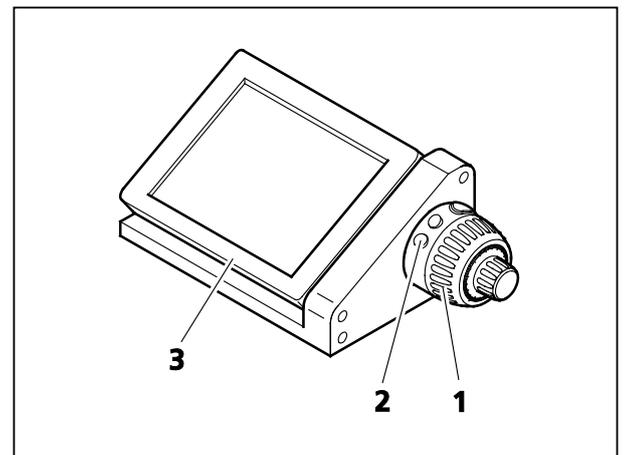
The plug-type power supply is a protection class II device (totally insulated). If its housing is damaged, the power supply unit needs to be put out of operation.

The plug-type power supply and the CAN / USB converter must not come into contact with moisture.

The USB/CAN converter is equipped with a plug-type power supply, which allows line voltages ranging from 100 V to 240 V  $\pm 10\%$  (50 / 60 Hz) to be used, without requiring any change of voltage on the unit.

#### 4.6.4 Docking station

- The docking station can only be used in conjunction with the motorized stand.
- If the microscope stand is inconvenient to operate from the right-hand side, the functions of the touch screen TFT display (4-18/3 on the docking station), the pushbutton ring to the right (corresponds to 4-18/2 on the docking station) and the focusing drive to the right (corresponds to 4-18/1 on the docking station) can be operated via the docking station, detached from the stand.
- Default button allocation can be altered via the TFT display.
- It is possible to change the angle of the TFT display with the aid of the two knurled thumb screws (see also 3-30/4) located on the back of the docking station.
- The coaxial drive (4-15/4) of the motorized mechanical stage may be removed from the latter (see Section 4.6.1). With the help of two screws, it can be vertically fastened to the back of the docking station.



**Fig. 4-18** Docking station with TFT display, pushbutton ring and focusing drive

## 4.7 Functions of the Light Manager

It is the function of the Light Manager to temporarily create optimum illumination settings for the various contrast methods and magnifications used and to make these settings reproducible to the user by providing the possibility to store them permanently.

The Light Manager has three operating modes: OFF, CLASSIC, SMART. The scope of functions available in the individual modes depends on some optional stand components.

The Light Manager is available for the transmitted-light contrast methods (brightfield, phase contrast, DIC, darkfield, polarization), for the reflected-light contrast methods (brightfield, darkfield, DIC, polarization) and for epifluorescence. When working in reflected light, the motorized stop slider, the neutral-density double-filter wheel and the motorized fluorescence attenuator, as far as available, are also included in the Light Manager functionality.

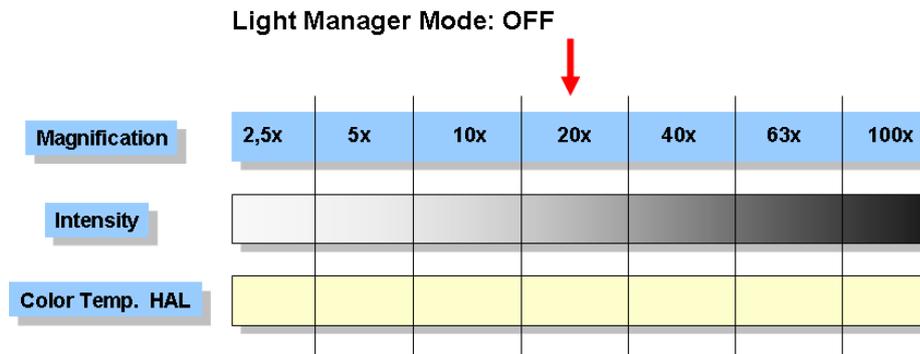
The following table informs you which operating modes of the Light Manager can be used for the individual stand models and the contrast methods mentioned.

<b>AxiImager Stativ</b>		<b>A2 LED</b>	<b>A2</b>	<b>A2m</b>	<b>D2</b>	<b>D2m</b>	<b>M2</b>	<b>M2m</b>	<b>Z2</b>	<b>Z2m</b>
OFF temporarily		+	+	+	+	+	-	-	-	-
OFF permanently		-	-	-	-	-	+	+	+	+
CLASSIC	TL	+	+	+	+	+	+	+	+	+
CLASSIC	RL (MAT)	+	+	+	+	+	+	+	+	+
CLASSIC	FL (BioMed)	-	-	-	-	-	-	-	+	+
SMART	TL	-	-	-	-	-	+	+	+	+
SMART	RL (MAT)	-	-	-	-	-	+	+	+	+
SMART	FL (BioMed)	-	-	-	-	-	-	-	-	-



RL (MAT) / FL (BioMed): Only one brightness value available per objective.

#### 4.7.1 Light Manager Mode: OFF

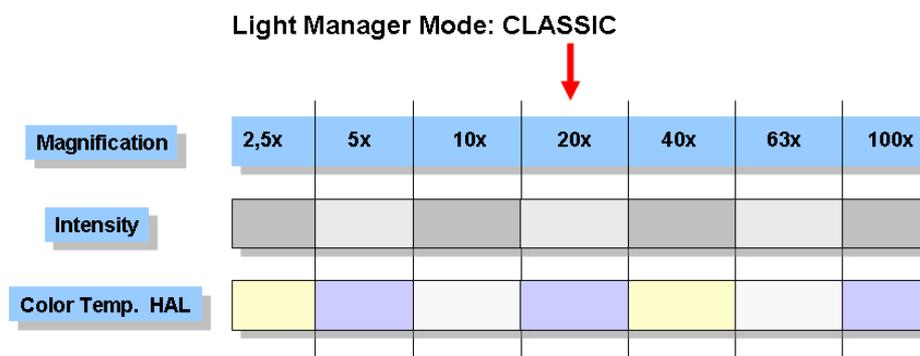


If the Light Manager is (temporarily) switched off, the microscope behaves like a classical light microscope.

Starting from a selected magnification and a corresponding lamp voltage, the operator must readjust the latter manually to get a comparable impression of image brightness when setting higher or lower magnifications.

However, as the color temperature of the halogen lamp varies with lamp voltage, the operator would have to use neutral-density filters additionally to achieve a comparable impression of the specimen.

#### 4.7.2 Light Manager Mode: CLASSIC

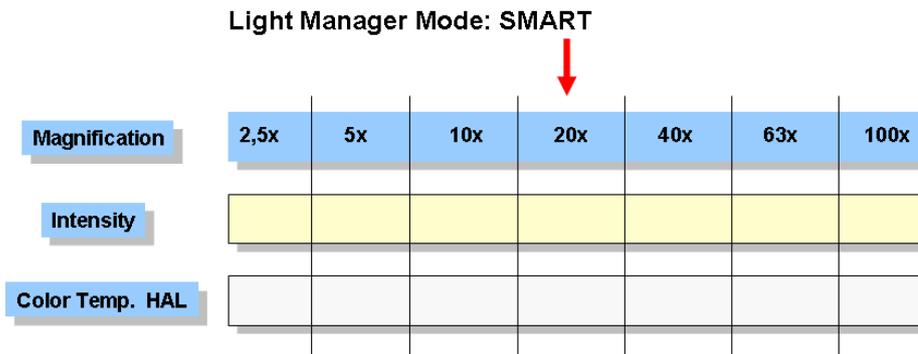


If the Light Manager is operated in CLASSIC mode, the operator can choose his own "optimal" illumination settings for every magnification.

The corresponding values are automatically stored in the temporary memory of the Light Manager when changing the objective.

If you want to keep the settings saved even beyond the switch-off of the microscope, you must once push the **LM-Set** button on the back right of the stand after. After a confirmation beep, all available values will be stored. After a second beep approximately three seconds later, you can switch off the microscope.

### 4.7.3 Light Manager Mode: SMART



In SMART mode, the Light Manager automatically calculates the optimum brightness for a specific contrast method for all objectives specified via the TFT display (or the MTB 2004).

If a motorized filter wheel with neutral-density filters is in the transmitted-light path, the color temperature is kept constant through the available filters.

The lamp voltage will be readjusted only, if it is impossible to achieve the optimum illumination through the neutral-density filters. The computed values are automatically stored in the temporary memory of the microscope.

Like in CLASSIC mode, the values can be stored permanently by pushing the **LM-Set** button on the back right of the stand.

In SMART mode, the operator can vary lamp intensity and attenuation by neutral-density filters by  $\pm 60\%$  for every magnification without recalculation being performed for the complete magnification range.

Any additionally installed coded or motorized microscope components (tube lens turret and Optovar modules in reflector turret) will also be considered for computing the brightness.

#### 4.7.4 Light Manager of the coded microscope

The Light Manager of the manual microscope controls the light intensity for object observation via the lamp voltage. The functioning depends on the used Light Control:

##### Light-Control, manual

Lamp voltage is saved for each

- Objective nosepiece position (coded)

##### Light-Control, motorized

Lamp voltage is saved for each

- Objective nosepiece position (coded)
- Reflector turret position (coded)
- Tube lens turret position (motorized)

On manual stands, always the CLASSIC mode of the Light Manager is active. In this mode, no illumination values are computed for the range of objectives used. The operator adjusts the brightness for every objective individually. Permanent storage is performed by pressing the **LM-Set** button on the rear side of microscope. This must be done for every objective individually.

When an objective is swung into the light path, for which the light intensity had been adjusted before already, the system will automatically set the corresponding stored value.

The light intensity settings of halogen lamp or LED illuminator are stored separately each for transmitted and reflected light. If the Light Manager shall be used for reflected-light contrast methods, make sure to set the voltage selector on the back right of the stand to reflected light before. Otherwise, the Light Manager will interpret the configuration of the stand as if fluorescence illumination has been configured. If a motorized neutral-density double filter wheel has been configured in reflected light, it will be set to 100% transmission and is thus inactive.



On the manual stands Axio Imager.A2, .A2m, .D2, .D2m, the Light Manager can be deactivated temporarily. To this end, you must keep the RL reflected-light shutter on/off button (4-1/11) depressed while switching on the microscope.

#### 4.7.5 Light Manager and Dazzle Protection control on manual stands

- Keeping the **RL** button depressed while switching on the instrument:  
-> **Light Manager (LM) & Dazzle Protection (DP)** are permanently deactivated / activated

#### 4.7.6 Light Manager of motorized microscope

The Light Manager of the motorized microscope (Axio Imager.M2, .M2m, .Z2, .Z2m) controls the light intensity for the observation of specimens in such a way that for all adjusted magnifications the operator gets the same impression of image brightness. The operating mode of the Light Manager can be selected via the Settings menu on the TFT screen (OFF, CLASSIC, SMART). Then, follow this procedure:

- Select the required contrast method.
- Adjust the light intensity for all objective nosepiece positions.
- Press the **LM-Set** button. On doing so, the current settings are transferred to the permanent memory. They will thus be available again when you switch on the microscope the next time.

The light intensity is changed for all others objective, too, depending on the objective and post-magnification. When turning on the nosepiece the light manager sets the light intensity as follows:

- if installed the motorized filter wheels are turned to keep the color temperature constant.
- If this will not do to completely match the calculated light-intensity levels, the intensity of the halogen lamp is varied by changing the lamp voltage. That way, of course, the color temperature for object observation changes as well.
- The luminous-field diaphragm is adapted to the visual field of the eyepieces (see also Section 4.8.6.1 on page 119), the user, however, may readjust it individually.
- The aperture diaphragm is adjusted according to the objective aperture (changes will be saved separately for bright field and DIC).

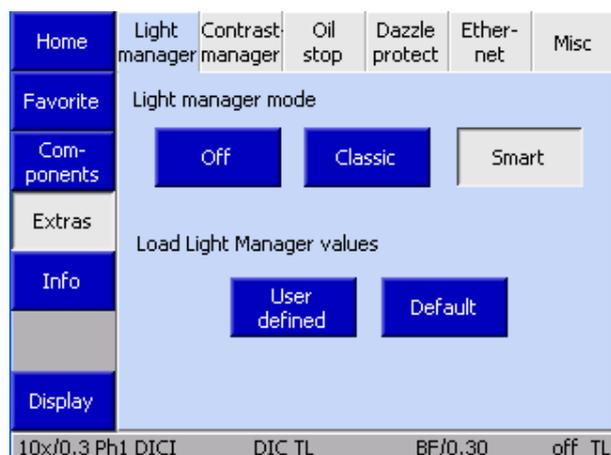


Fig. 4-19 Setting/resetting the Light Manager

- If you want to reset temporary Light Manager values to the last settings saved with the **LM-Set** button, you can do this by pressing the **User defined** button. On doing so, the temporarily stored values are overwritten by the permanently stored ones and activated.
- If the manufacturer's default settings are to be used instead, the **Default** button must be pressed. On doing so, the default values will be loaded, written to the temporary memory and activated. If you want to use the default settings permanently, write them to the permanent memory by pressing the **LM-Set** button. The manufacturer defaults cannot be overwritten.

 The calculated intensities apply only to the contrast technique selected. That means the intensity must be adjusted specifically for each contrast technique.

 The Light Manager works only if all dependent components were configured correctly (see Section 4.8.6).

## 4.8 Operating the motorized microscope via the touchscreen of the TFT display

### 4.8.1 Screen layout

On the motorized Axio Imager, the operator can operate and configure the microscope, set it up for different users and use optional functions via the TFT display. The TFT display is designed as touch-sensitive screen.

The control elements and information displays are contained on various, thematically different pages. Basically, a page on the TFT display is divided into the following main areas (Fig. 4-20).

#### 4.8.1.1 Navigation button bar

The navigation button bar at the left edge of the screen contains buttons via which you can activate all pages. The buttons available depend on the currently displayed page. However, the following buttons are accessible on all pages:

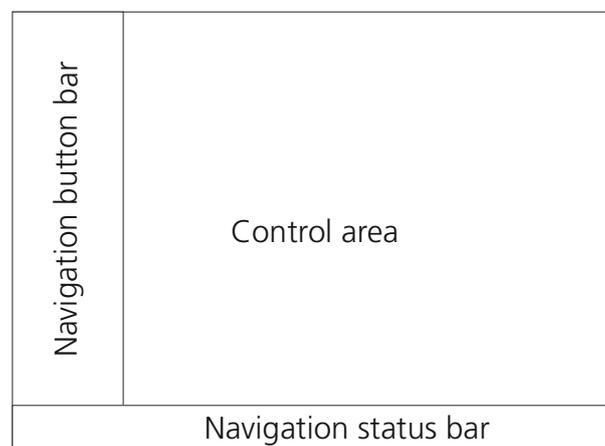
- **Home** Activates the start page
- **Favorites** Calling up the favorites page
- **Display** Activates the display page

#### 4.8.1.2 Status line

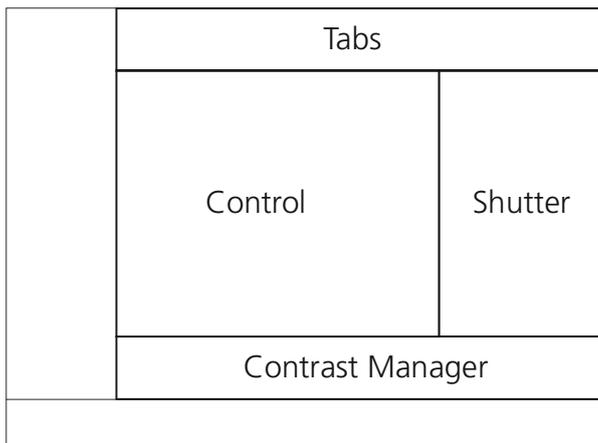
The status line on the lower edge of the screen furnishes information on the current settings of the:

- objective nosepiece,
- reflector turret,
- condenser,
- lamp voltage or intensity,
- switch status of the internal power supply.

Pop-up windows are not shown on the status line



**Fig. 4-20** Main areas of TFT display



**Fig. 4-21** Control area of the TFT display

#### 4.8.1.3 Control area

The control area is where the actual operation of controllable components takes place.

The control area is subdivided into further subareas (see Fig. 4-21):

##### (1) Tabs

The user can call the desired component function up via a *Tab*. Tabs are used to activate the desired function, which is displayed in the control subarea. Maximally six tabs per page are available.

##### (2) Illumination channels

The buttons for reflected light (**RL illumination**) and transmitted light (**TL illumination**) are located on the right edge of the control area. The **Close** and **Open** buttons function like switches, i.e. the shutter in the optical path of the microscope is either open or closed. If there are no shutters, the halogen or LED illuminators configured are controlled directly. When no illuminator has been configured, the control buttons will not appear.

##### (3) Control

The control subarea contains control elements dependent on the button selected on the navigation button bar or the tab.

##### (4) Contrast Manager

The contrast manager constitutes the centerpiece of the operation efficiency concept employed in all motorized Axio Imagers. With its help, the various contrast methods possible are set automatically and reproducibly.

The contrast manager can be switched **ON** and **OFF** globally. The **Contrast** tab is not available when the contrast manager has been switched off. If this is the case, no contrast can be selected and the corresponding components are not set automatically.

If the contrast manager has been switched **ON** globally, it will be **active** whenever a contrast button has been pushed. A transmitted-light contrast method that is dependent both on condenser and reflector modules (e.g., DIC) can be selected both via the transmitted-light contrast button and via the reflector position (by pressing the DIC TL analyzer position in the case of DIC).

If the contrast manager is **not active**, it can be activated both on the **Objectives** tab and the **Reflector** tab. On the *Objectives* page, the transmitted-light contrast methods plus fluorescence are available. On the *Reflector* page, all the reflector-dependent contrast methods can be selected. This is of particular interest when it comes to reflected-light methods.

Even though the **shutters** or illumination switches are integrated in the functionality of the contrast manager, they can be operated independently.

At the bottom edge of the control area, a bar is arranged containing buttons for selecting the contrast method. The Contrast Manager is controlled via the following page: **Microscope** → **Revolver** → **Contrast**. The contrast methods available depend on the current microscope configuration. Altogether, the following contrast methods are available:

Abbr.	Method	Requirements
FL	Fluorescence	Installed reflected light shutter (standard), FL filter set used in reflector turret
BF	Brightfield	Reflected light: Reflector turret with brightfield module available Transmitted light: Configuration of a transmitted-light illuminator
DIC	Differential Interference Contrast	Transmitted light: Installed motorized achromatic-aplanatic condenser 0.9 H D Ph DIC or motorized LD condenser 0.8 H D Ph DIC Reflected light: Motorized reflector turret with C-DIC module available
PH	Phase Contrast	Transmitted light: Installed motorized achromatic-aplanatic condenser 0.9 H D Ph DIC or motorized LD condenser 0.8 H D Ph DIC
DF	Darkfield	Transmitted light: Installed motorized achromatic-aplanatic condenser 0.9 H D Ph DIC or motorized LD condenser 0.8 H D Ph DIC Reflected light: Motorized reflector turret with darkfield module available
C-DIC TIC	Circular DIC, Total Interference Contrast	Reflected light: Motorized modulator and coded or motorized reflector turret with C-DIC module available
POL	Polarization Contrast	Transmitted light: Motorized 0.9 H D Ph DIC achromatic-aplanatic condenser or motorized 0.8 H D Ph DIC LD condenser available, polarizer installed in condenser, analyzer module installed in reflector turret or analyzer slider Reflected light: Polarizer / analyzer module installed in reflector turret



The contrast methods result from the interaction of condenser, reflector, modulator turret and shutter positions as well as other parameters. The current contrast method is automatically recognized and displayed on the TFT display. In the event of non-permitted manual settings (e.g., vacant reflector position or wrong position of the condenser turret), no contrast method can be indicated or the contrast manager gets switched off.

## (5) Popup windows

Popup windows are overlaid to a page for one of the following reasons:

- They prompt the operator for additional entries: The operator must make a choice (adapt the configuration after initialization, enter values, etc.).
- They display error messages or special advice: Possibly such messages must be confirmed with the **Close** button.
- They display the operating state (wait time): Such windows close automatically.



While a popup window is open, you cannot operate the overlaid page.

## 4.8.2 Menu structure

Depending on the microscope configuration, the actual menu structure may deviate from the structure shown below. It shows the total scope inclusive of optional components and menu items that are accessible only if the user has administrator privileges (without administrator login, the user has reading privileges only).

In the second Axio Imager product generation, the stand type (BioMed or Material) does no longer have to be preselected in the user interface!

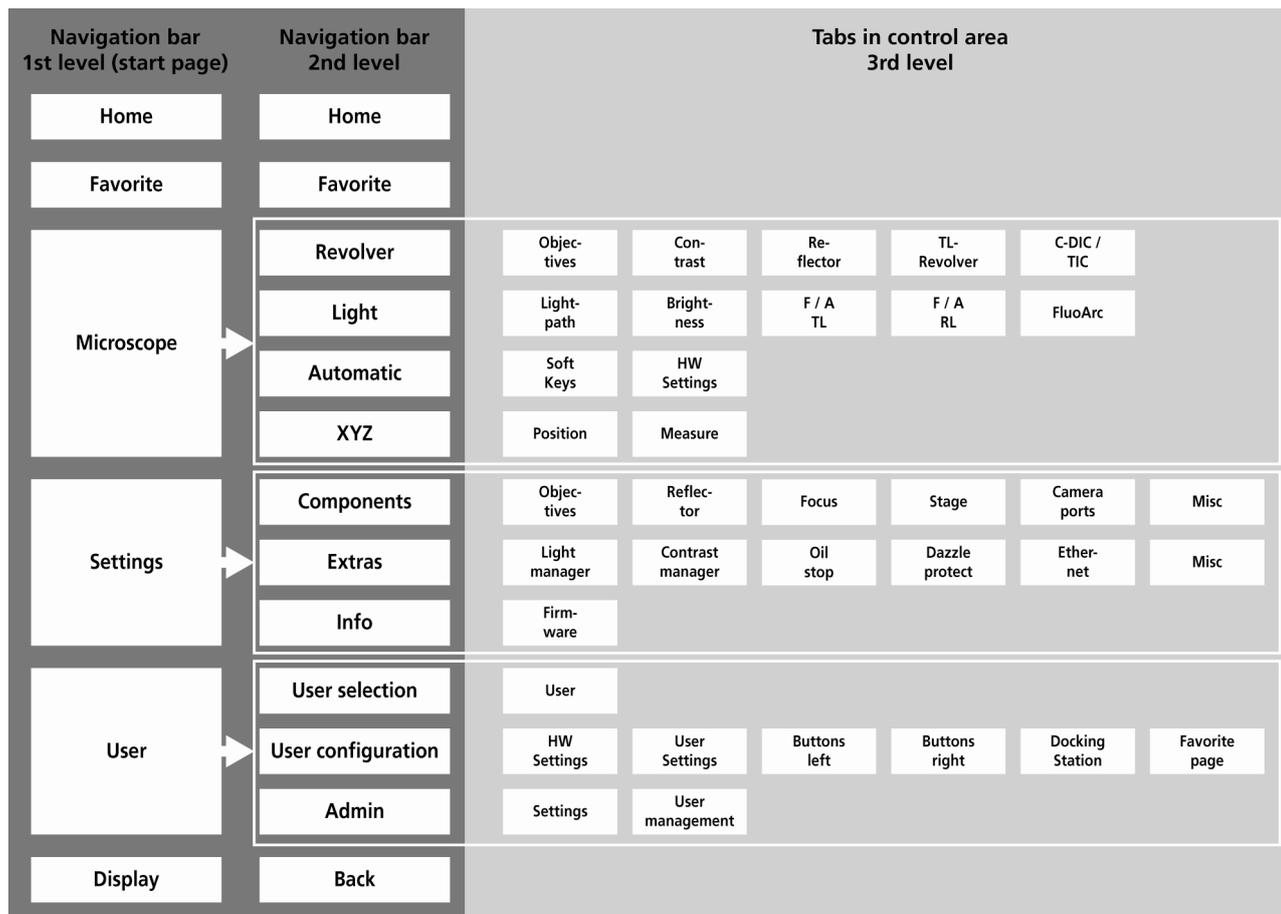


Fig. 4-22 Menu structure

On the navigation button bar, the buttons of the *first level* appear on the very left (Fig. 4-22). By pressing the **Microscope**, **Settings**, **User** and **Display** buttons, the button assignment on the navigation button bar is changing.

The buttons in the *second level* of the navigation button bar activate the correspondingly allocated tabs. By pressing the tabs, further buttons appear in the control area of the screen.

All the operating functions are exclusively displayed in the control area (4-23/2) or in a pop-up window. The instrument status existing at any given time may be conveniently gathered from the navigation bar or the Home page.

### 4.8.3 Home page

After switch on, the microscope is initialized. This process takes a few seconds. The **Home** page (Fig. 4-23) appears when the instrument is first started up.

 If coded / motorized components are exchanged or removed while the instrument is switched off, the operator may possibly have to reconfigure the instrument after it has been switched on again.

You can call all other pages via the buttons on the navigation button bar (4-23/1).

The **control area** (4-23/2) shows the configuration identified, as well as its current status. All coded and motorized control elements detected during initialization appear in the status field, otherwise, the character "-" will be displayed. The control elements are arranged from top to bottom according to their significance.

The following operation controls are located along the right edge (4-23/3):

- **Users** button:  
When this button is pressed, the user selection page will appear (Fig. 4-24). A preconfigured user may be chosen on the **user selection page**. For the configuration of a user, see Section 4.8.6.

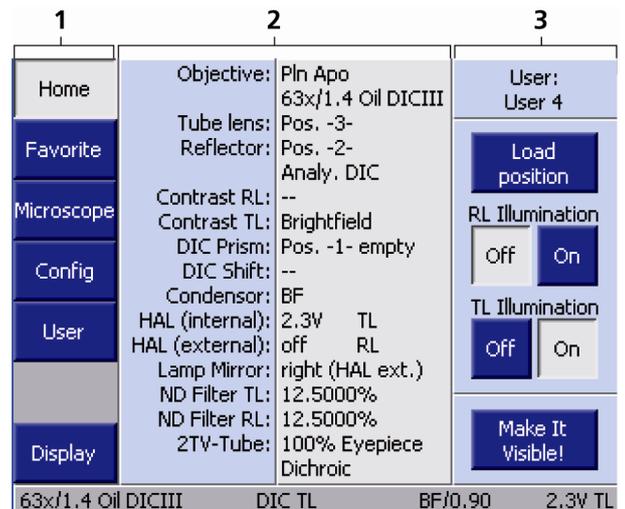


Fig. 4-23 Home page

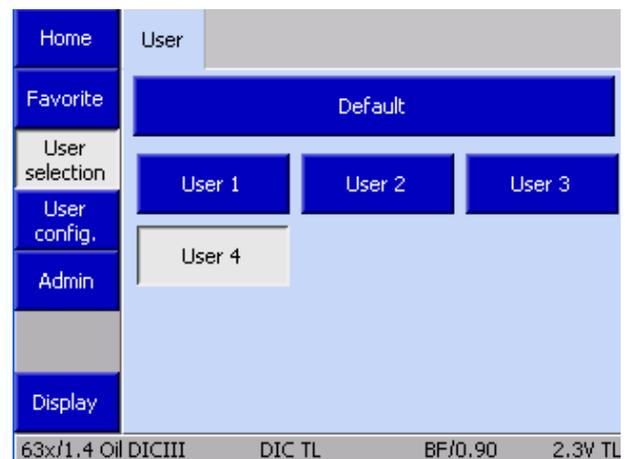


Fig. 4-24 User selection page

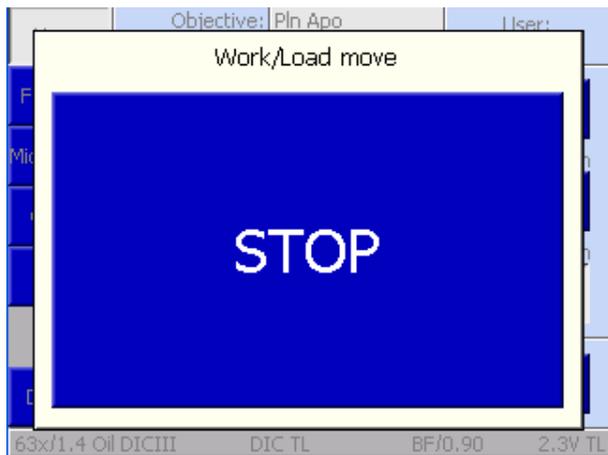


Fig. 4-25 STOP button

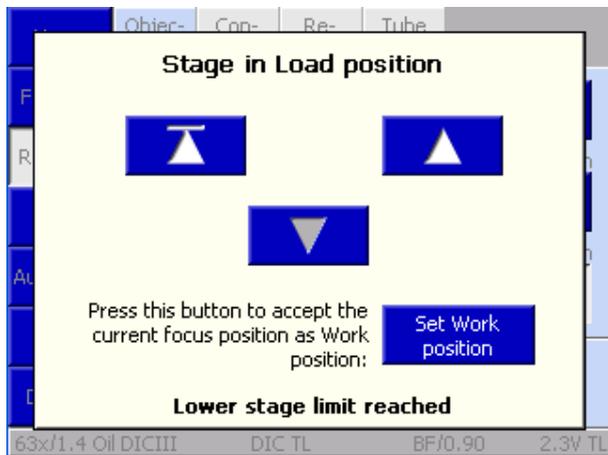


Fig. 4-26 Stage in load position

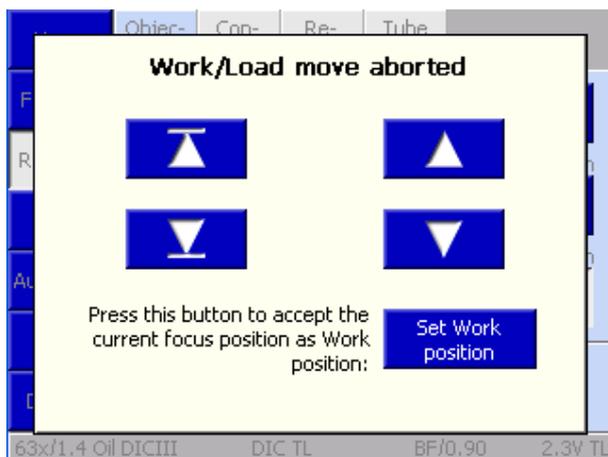


Fig. 4-27 Travel aborted

- **Load Position** button:  
When this button is pressed, the stage travels to the load position. The load position lies approximately 10 mm below the operating position. The load travel of the stage can be interrupted via the **STOP** button (Fig. 4-25) or via any button located on the stand base. When the stage arrived in the load position, the **Load Position** popup window (Fig. 4-26) appears with the following control elements:

 Stage returns to operating position.

 While the button is depressed, the stage will travel to the operating position (to the upper stage stop at the most).

 Stage moves down as long as you press this button (to the lower stage stop at the most).

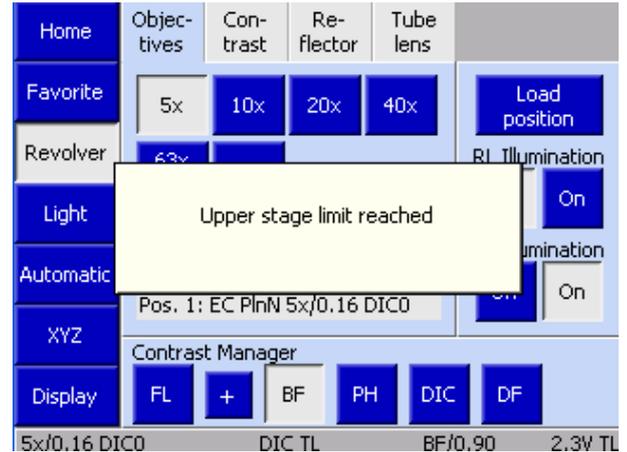
 When this button is pressed, the current focus position is adopted as the operating position.

 **Caution!** If the operating position set is below the current focus position, the focusing drive will move downward when the  button is pressed.

When the **STOP** button is pressed, the load travel is aborted and a pop-up window (Fig. 4-27) is displayed. The additional button appearing here serves to move the stage to the load position originally intended.

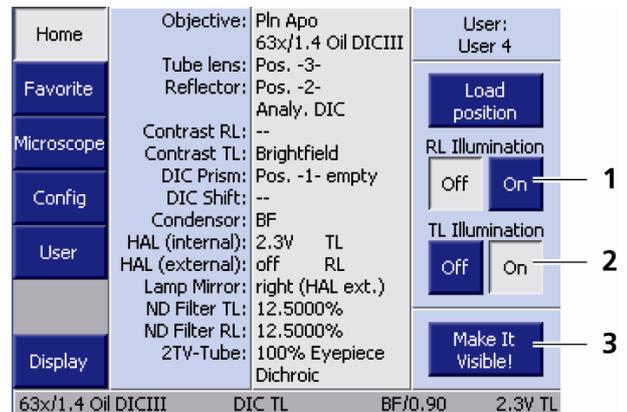
When the lower stage stop is reached, the message "Lower stage stop reached" will appear.

When the focusing drive is used in a regular way, a pop-up window (Fig. 4-28) will appear as soon as the upper or lower limit stop is reached.



**Fig. 4-28** Motorized focusing drive has reached the upper stage limit switch

- **Reflected-light illumination** button (4-29/1)  
The **Close** and **Open** buttons serve to open or close the shutter for reflected light (RL).
- **Transmitted-light illumination** button (4-29/2)  
The **Close** and **Open** buttons serve to open or close the shutter for transmitted light (TL). If there is no shutter in the transmitted-light path, the voltage of the configured lamp will be adjusted instead.
- **Make It Visible!** button (4-29/3)  
This button serves to switch the microscope to a basic state:
  - Transmitted-light lamp adjusted to medium intensity (2.3 V),
  - Luminous-field diaphragm opened,
  - Aperture diaphragm opened
  - TL shutter open,
  - RL shutter closed,
  - All filter wheels in transmitted light switched to blank aperture (100% directed to eyepieces),
  - Condenser switched to brightfield,
  - Reflector turret switched to the nearest HAL position (Halogen = transmitted light),
  - Light path switched to 100% to eyepieces.



**Fig. 4-29** Controls

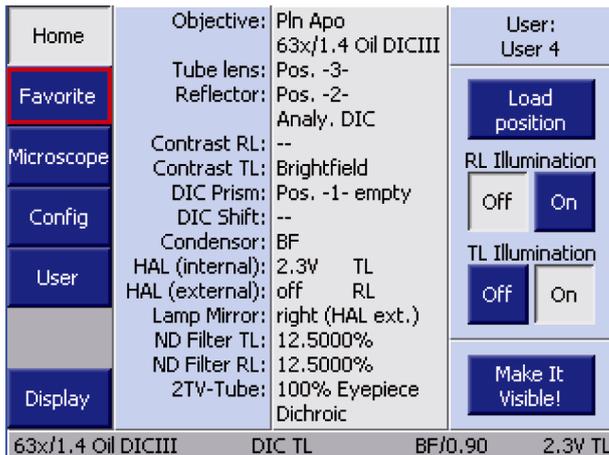


Fig. 4-30 Favorites page

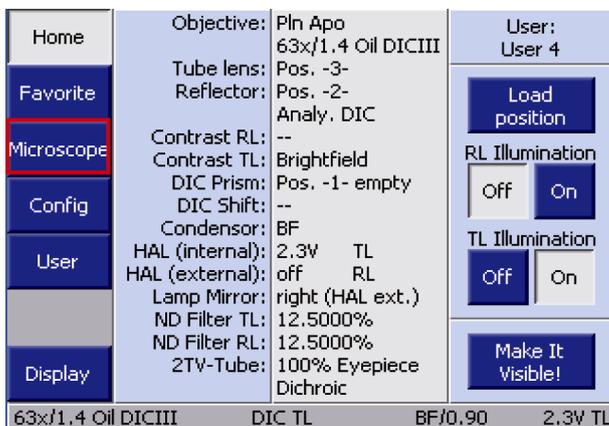


Fig. 4-31 Calling up the Microscope page

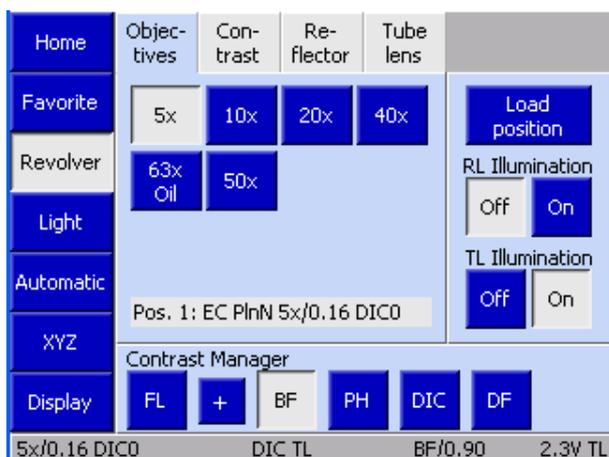


Fig. 4-32 Microscope -&gt; Revolver -&gt; Objectives

#### 4.8.4 Favorites page

In order to streamline the operation of the microscope, a frequently used page can be defined as a favorites page. When it is selected, the system will jump directly to this page. For the procedure of how to define this page, see also Section 4.8.6.

#### 4.8.5 Microscope page

By pressing the **Microscope** button on the navigation bar on the **Home** page, the user gets to the pages from where the operation of the microscope is controlled:

- **Turrets**
- **Illumination**
- **Automatic operation**
- **XYZ**

##### 4.8.5.1 Revolver page

Depending on the configuration carried out under the **Settings** menu item, different tabs and information will appear on the **Revolver** page. These are described in the following.

##### (1) Objectives tab

Depending on the nosepiece installed (all motorized ones), up to seven control elements will appear for the various objective positions (Pos.1 to Pos.7). For objective positions already configured, the system will display the magnification and, where applicable, the following additional information:

- Oil: Oil immersion objective
- W: Water immersion objective
- Imm: Immersions

 The operation page for objectives is not available with coded nosepieces. The active objective is only indicated in the status line.

 If the light manager is active, it is automatically used in the event of a change of objective.

 If a contrast method was set in the contrast manager prior to a change of objective, the contrast manager will automatically try to adapt the method to the objective (i.e., positions on the condenser turret and reflector turret may change). If the contrast method is not available for the objective concerned, the system will switch to brightfield.

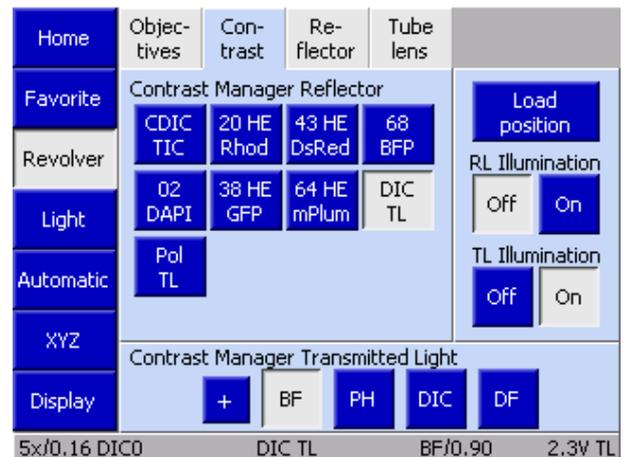
## (2) Contrast tab

On this page, the desired contrast can be selected as a function of the reflector modules, condenser modules and objectives configured. This applies both to the reflected-light and transmitted-light contrast methods.

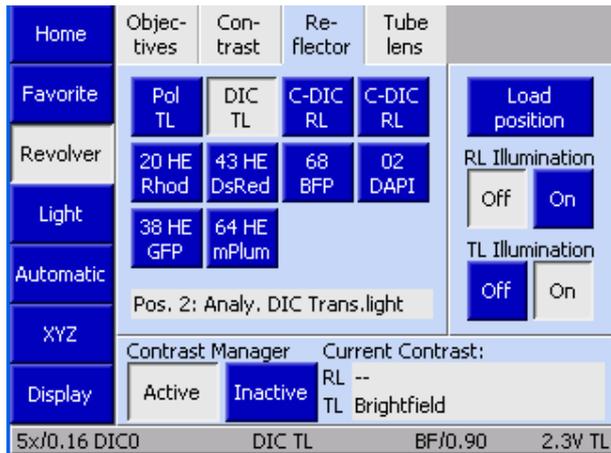
In this context, reflected-light fluorescence counts as a reflected-light method. For this reason, the second-generation Axio Imager does not come with an **FL** button. If motorized components are installed, the corresponding contrast is set at the touch of the button.

The **+** button is provided only if the active light channels (reflected light and transmitted light) are not controlled via the internal power supply.

 The **Contrast** tab will be available only if the contrast manager has been switched **ON** globally. For more information, refer to Section 4.8.6.



**Fig. 4-33** Microscope -> Revolver -> Contrast



**Fig. 4-34** Microscope -> Revolver -> Reflector

### (3) Reflector tab

Depending on the reflector turret installed, 6 or 10 control elements for reflector positions 1 to 6 or 1 to 10 will appear. The reflector modules already configured are recognizable by the text showing on the button.

- To swivel in the reflector module desired, press the relevant button.

If the contrast manager is activated on this page and if components valid for the contrast method determined by the reflector module have been installed, the appropriate method will be set. Should this be impossible, the contrast manager cannot be activated at this reflector position.

The transmitted-light contrast method last chosen will be set at an empty or transmitted-light position.

If an invalid reflector position is selected while the contrast manager is active, the system will travel to the position selected, but without the contrast being adjusted. No contrast method will be displayed.



The tab will not be available if no motorized reflector turret is installed. The active reflector module will only be indicated in the status line (Fig. 4-23).

#### (4) C-DIC/TIC tab

On this page, the desired method can be selected by pressing the relevant button, provided the objective configured and the analyzer installed in the reflector turret are suited for this method.

Depending on the objective used, the settings for **C-DIC** or **TIC** can be made on this tab.

Azimuth and shift (for C-DIC only) are to be set directly via the setscrews provided on the modulator turret. Such data cannot be entered directly on the TFT display.

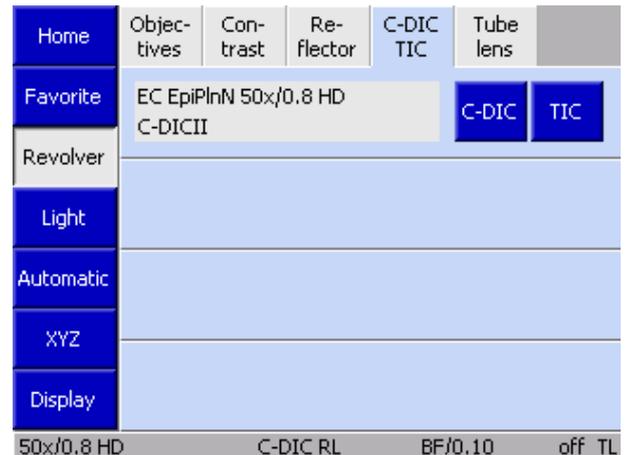


Fig. 4-35 Microscope -> Revolver-> C-DIC/TIC

#### Circular DIC

- To set the azimuth value, turn the front setscrew on the modulator turret. The values concerned are presented directly. Each click-stop position of the setscrew corresponds to a one-degree change. Press the **Zero** button to set the mid-position.
- Turn the rear setscrew to set the shift value. Each click-stop position of the setscrew corresponds to a 0.05-mm change. To reset the value to 0, press the **Zero** button.

**Azimuth** and **shift** values are automatically and temporarily saved for each objective. If no **azimuth** or **shift** has been set yet for a given objective, the settings of the objective last used will be employed. In the event that there are no values to fall back on for any of the objectives, the default values for azimuth and shift will be used.

In addition, it is possible to save up to four pairs of fixed values for azimuth and shift, which you can restore via the **Position** buttons located on the lower edge of the screen.

Assigned buttons are labeled with the azimuth and shift values set. Unallocated buttons are labeled with Pos. 1 to Pos. 4. The **Reset to Zero** button sets the **Azimuth** to mid-position and **Shift** to zero.

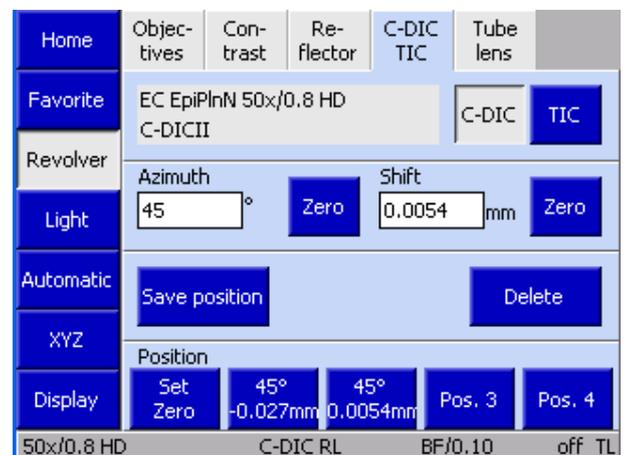


Fig. 4-36 C-DIC settings

the settings of the objective last used will be

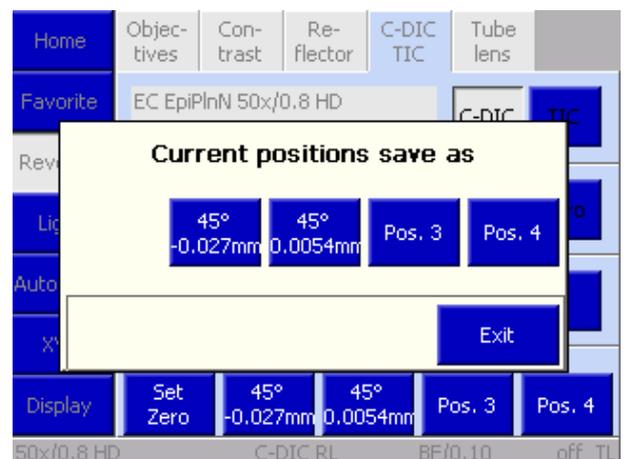


Fig. 4-37 Saving the position

- Before you can save a fixed position, you need to set the desired **Azimuth** and **Shift** values.
- Press **Save position**.  
A pop-up window (Fig. 4-37) with the four position buttons will open.
- Press the desired position.  
A safety query will appear if the position has already been allocated. If you acknowledge with **Yes**, the new position will be stored at the button selected.

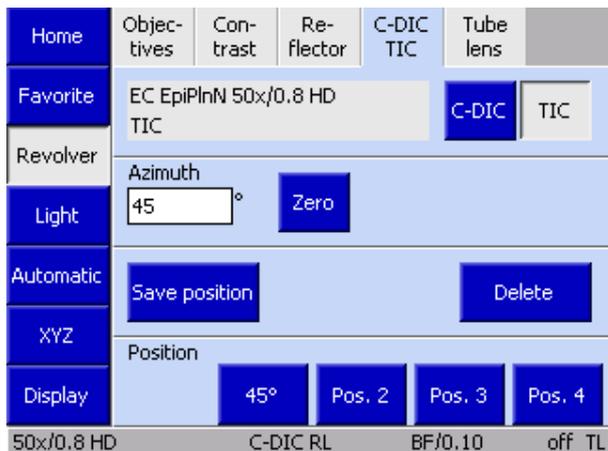


Fig. 4-38 TIC settings

### TIC

The operating procedure for the TIC method corresponds to that for C-DIC. What is different though is that only the azimuth value needs to be set. There is no **Zero** button.



The tab will not be available if no motorized reflector turret is installed.

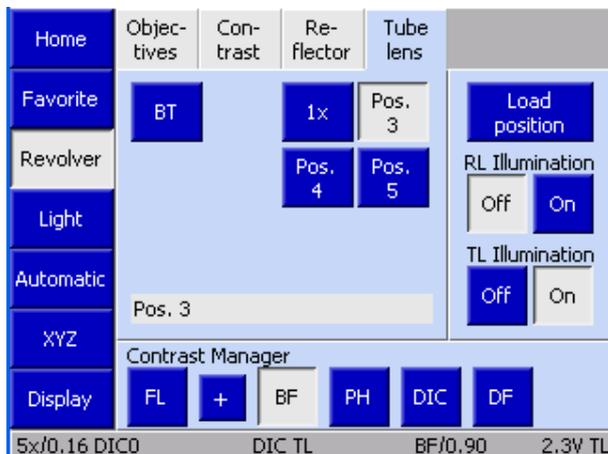


Fig. 4-39 Microscope -&gt; Revolver -&gt; Tube lens

### (5) Tube lens tab

Depending on the type of tube lens turret employed, up to four control elements may be available for tube lens positions 1 to 4. If the tube lenses have been configured, the names will be displayed. The fifth position is always fitted with the Bertrand lens.

The **BT** button serves to swivel the Bertrand lens in and out.

- In order to swivel the desired tube lens into the optical path, the relevant button needs to be pressed.



The tab will not be available if no motorized tube lens turret is installed. The active tube lens will only be indicated in the status line (Fig. 4-23).

#### 4.8.5.2 Light page

Light path and intensity control functionalities are consolidated on the **Light** page.

As many as four tabs are available here, depending on the configuration of the microscope:

- **Light path:** Light path control
- **Light intensity:** Filter wheels and lamp voltage for reflected and transmitted light
- **F/A TL:** Field and aperture diaphragm control in transmitted light
- **F/A RL:** Field and aperture diaphragm control in reflected light

##### (1) Light path tab

Under the **Light path** tab, the light path of the microscope on the imaging side is shown in a schematic diagram.

Color lines symbolize the light path.

- **yellow**  
Active light path, which can be adjusted via mirrors
- **gray**  
Inactive (unknown) light path, which cannot be adjusted

Special case: Dichroic deflection

- **blue**  
"Side Port deflected" deflection plane (first dichroic deflection)
- **green**  
'Side Port in pass-through mode' deflection plane (second dichroic deflection) or deflected for 2TV tube
- **red**  
2TV tube in pass-through mode (dichroic passage)

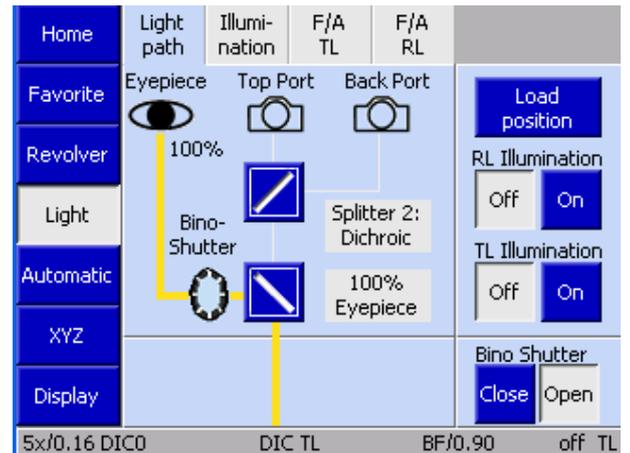


Fig. 4-40 Light page

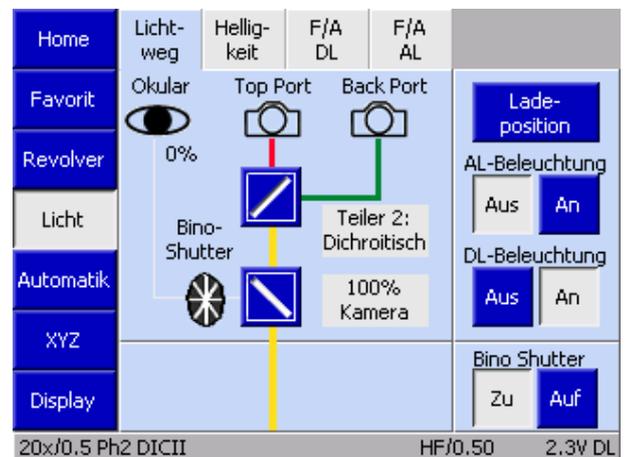


Fig. 4-41 Microscope -> Light -> Light path

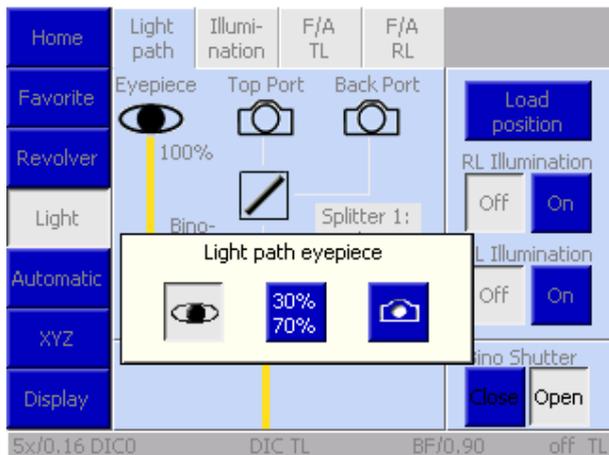
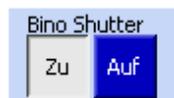


Fig. 4-42 Light path switch 1 in 2TV tube

The light path can be controlled by pressing the  buttons. Various options are available, depending on the type of component.

In Fig. 4-42, you can see how the first light path switch in the 2TV tube is controlled, while Fig. 4-43 shows how the second light path switch in the 2TV tube is controlled.

If an eyepiece shutter is installed, the **Bino Shutter** option is displayed in the **Light path** tab.



The state of the eyepiece shutter is controlled independently of the remaining light path by pressing the **OPEN** and **CLOSE** buttons.



This tab will not be available in the absence of a light switch.

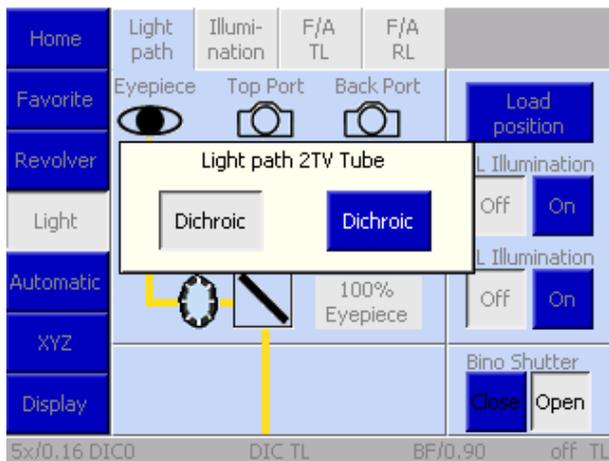
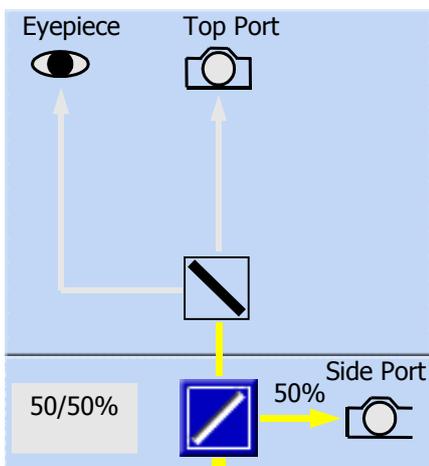
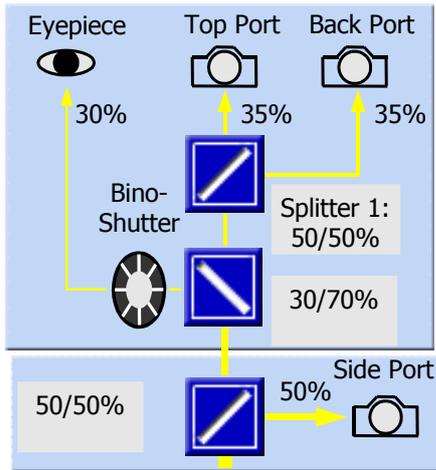


Fig. 4-43 Light path switch 2 in 2TV tube

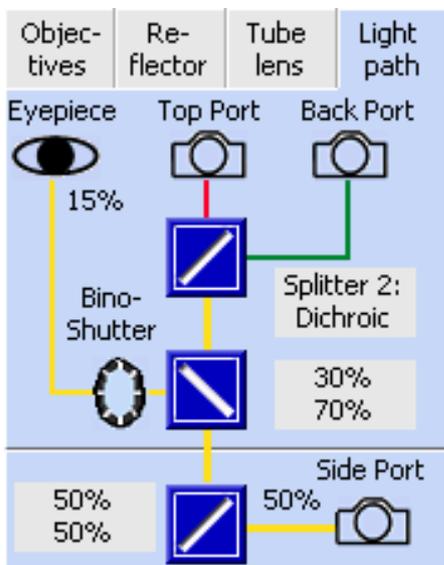
 One of the following schematic diagrams or a similar one is displayed in the control area to show the light path. The configuration is determined during the microscope initialization process. That is why tube changes must take place while the microscope is switched off.



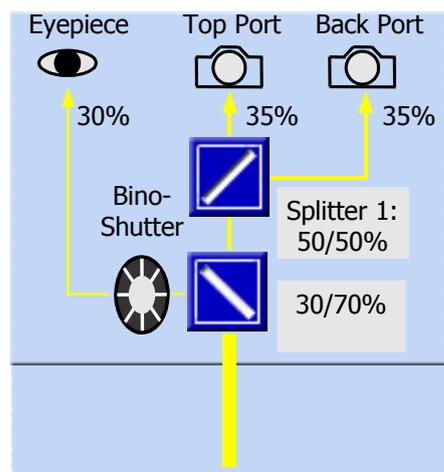
- Only the motorized camera path deflection at the Side Port is available. The deflection plane mirror is active.
- A manual tube (no 2TV tube, no Bino Shutter (eyepiece shutter)) is installed. The "Eyepiece" mirror is inactive and the light path unknown.



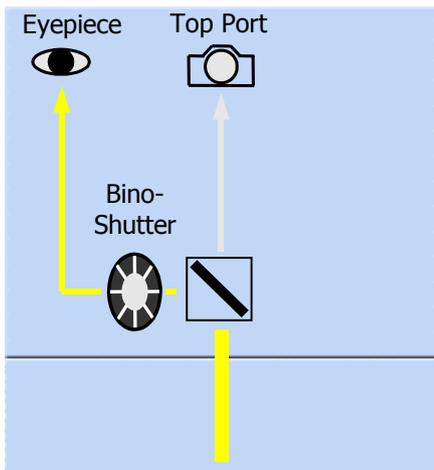
- 2TV tube (Top Port/Back Port) and motorized camera path deflection at Side Port are installed.
- Three mirrors (deflection plane, eyepieces, Top Port/Back Port) are active.



- 2TV tube (Top Port/Back Port) and motorized camera path deflection at Side Port are installed
- Two mirrors (deflection plane, eyepiece, Top Port) and a dichroic beam splitter at the Back Port are active.
- The active light paths are displayed in yellow, inactive light paths appear in extra light gray (line width: 1 pixel).
- The line width for the active light paths depends on the respective quantity of light transmitted:
  - 100% = 4 pixels
  - 50% = 3 pixels
  - 25 - 15% = 2 pixels
  - <15% = 1 pixel
- If a dichroic beam splitter is used, the light path to the top port is shown in red, while the deflected one appears in green. The line width is reduced by one pixel each.



- 2TV tube installed. Mirrors for eyepiece and Top Port/ Back Port deflection are active.
- No motorized camera path deflection at Side Port installed.



- Tube with Bino Shutter (eyepiece shutter) installed.
- Mirror for deflection to eyepieces is unknown.
- Light path for Top Port is unknown.
- No motorized camera path deflection at Side Port installed.

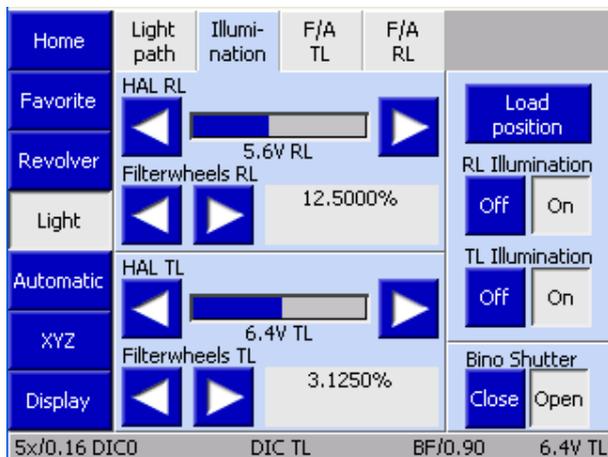


Fig. 4-44 Microscope -> Light -> Brightness

## (2) Brightness tab

This is where the lamp voltage in transmitted and reflected light and the filter wheels can be checked. These functions are available only if motorized filter wheels / attenuators are used.

When the internal power supply is configured both in reflected light and transmitted light, only a bar is displayed, which changes accordingly.

### Working with two power supplies and two illuminators:

Second-generation Axio Imagers can be equipped with an additional external CAN bus enabled power supply. When such a device is detected during the switch-on operation, it will be automatically initialized. In order for the power supply to be used, a light source must be allocated to it under

**Settings -> Components -> Miscellaneous -> TL/RL Illumination.**

Using two power supplies, two halogen or LED illuminators may be operated in reflected and transmitted light at their full power, completely separately or jointly. The desired illuminator is selected via the **On** or **Off** buttons on the TFT, the light control buttons or, if there is no shutter in transmitted light, even directly via the incremental encoder of the light control.

### Generating mixed light:

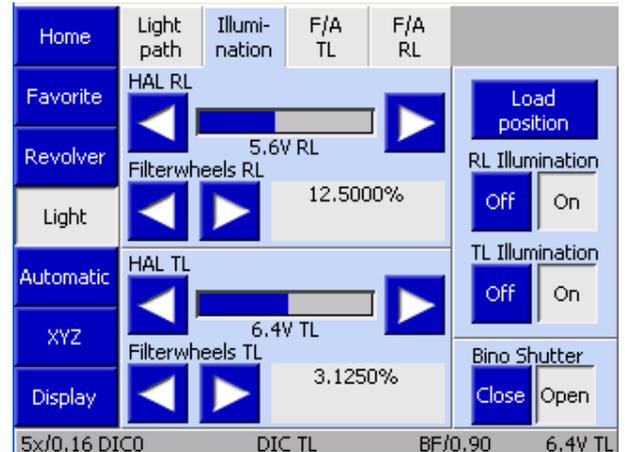
To generate mixed light, the two illuminators need to be switched on. The two power supplies are then controlled synchronously. This means that the brightness of the two illuminators is adjusted up and down in parallel, independently of the intensity regulator.

### (3) F/A TL tab

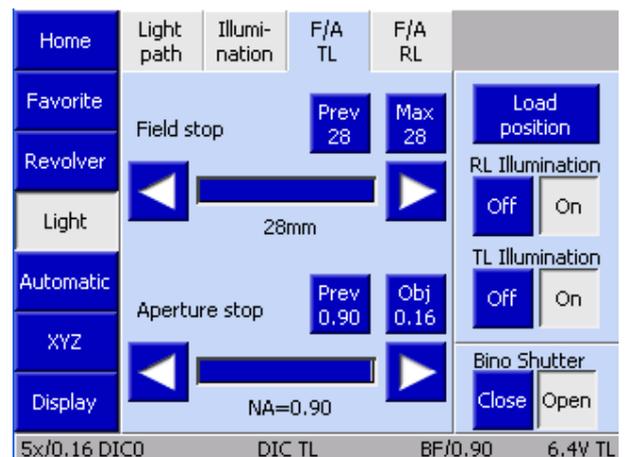
The luminous-field diaphragm diameter and the aperture diaphragm for the transmitted-light path can be set here.



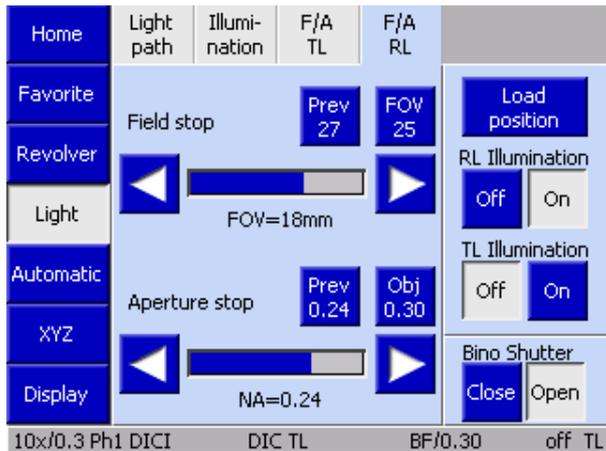
This function will be available only if a motorized field diaphragm or aperture diaphragm is installed.



**Fig. 4-45** Using of two power supplies and illuminators



**Fig. 4-46** Microscope -> Light -> F/A TL



#### (4) F/A RL tab

The luminous-field diaphragm diameter and the aperture diaphragm for the reflected-light path can be set here.



This function will be available only if a motorized field diaphragm or aperture diaphragm is installed.

Fig. 4-47 Mikroskop -> Light -> F/A RL



The following applies to all diaphragms in reflected and transmitted light:

No configuration of objectives and eyepieces available:

Diaphragm aperture data are unscaled. The values presented are absolute in relation to the physical aperture of diaphragms.

Configuration of objectives and eyepieces available:

Diaphragm aperture data are presented in regard to the field-of-view number of the eyepieces and aperture of the objective used.

**Prev** (previous) button:

It always shows the value last set.

**Max/SF** button:

It shows the maximum meaningful (optically scaled) diameter of diaphragms.

When one of the two buttons is pressed, the relevant diaphragm is automatically set to the value displayed.

Whenever the light manager is employed, the optimum diameter is set automatically. Other values, which may range up to the maximum diaphragm diameter physically attainable, can be set via the arrow buttons.

The values last set will be lost when the objective is changed.

### 4.8.5.3 Automatic operation page

This menu item contains two tabs:

- **Soft Keys** and
- **HW Settings**

#### (1) Soft Keys tab

On this page, ten functions or courses of action can be selected under Axio Vision, which may later be called up from here when the program is being executed.

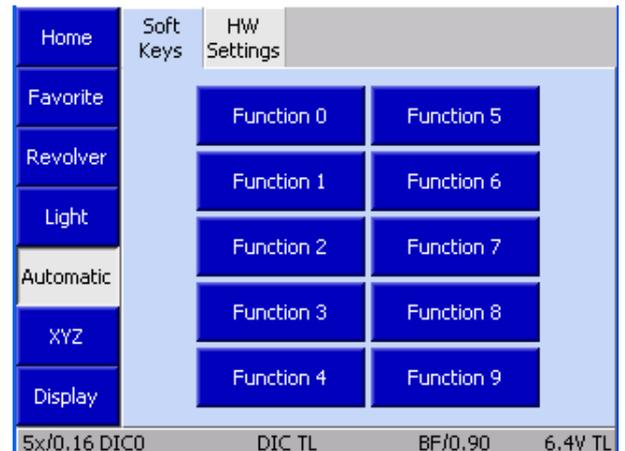
These can be simple settings or a snap with the camera or even a complex experiment. The selection is made within the AxioVision software under the AxioVision Software menu item.

Those functions are only active when the AxioVision software is running and connected to the microscope.

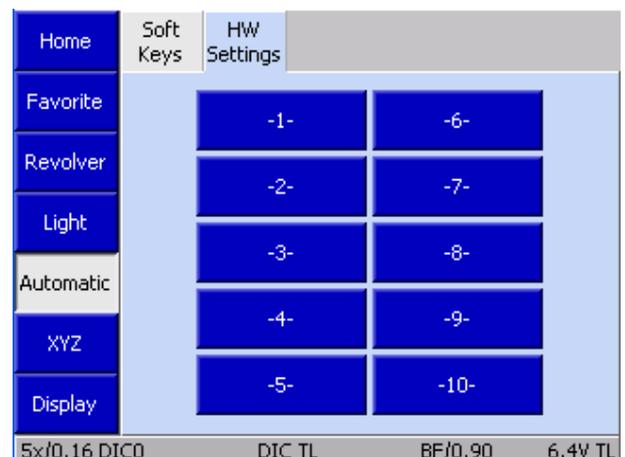
#### (2) HW Settings tab

On this page, hardware settings defined on the microscope can be called up. Complex settings can be saved in an easy and user-specific manner and called up later at the touch of a single button.

For allocation, see also Section 4.8.6.



**Fig. 4-48** Microscope -> Automatic -> Soft Keys



**Fig. 4-49** Microscope -> Automatic -> HW Settings

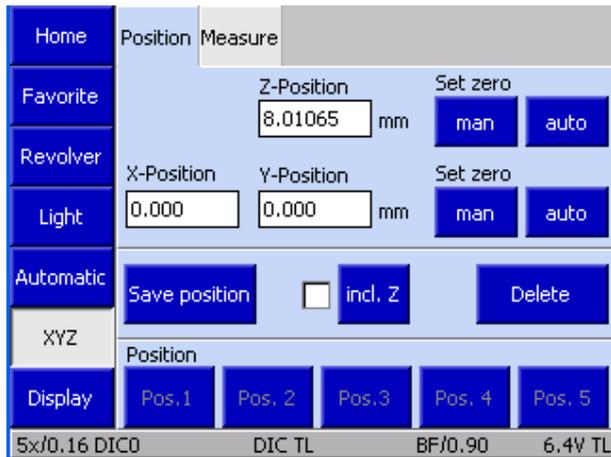


Fig. 4-50 Mikroskope -&gt; XYZ

#### 4.8.5.4 XYZ page

The **Microscope/XYZ** page contains two tabs: **Position** and **Measure**.



The availability of the **XYZ** page depends on the microscope stage used:

- Motorized stages (only CAN bus stages that are directly connected to stands .M2 / .M2p / .M2m or .Z2 / .Z2m:  
Allows settings of XY coordinates and Z focusing drive
- Manual stage:  
Allows settings for Z focusing drive only (all XY controls are absent),  
**Measure** tab not accessible
- Manual stage/manual Z focus drive:  
**XYZ** page is not accessible



During microscope initialization, the system detects whether a stage is installed. Therefore, the stage must be changed only with the microscope switched off.

#### (1) Position tab

The control area under the **Position** tab is subdivided into three functional blocks.



If you do not use a motorized stage, the **Start** button is available in place of the XY controls (refer to (2) Measure) below.

#### Current position display / Set Zero

Displays the current positions of Z, X and Y in millimeters (mm).



If no motorized Z focusing drive is installed, the Z position is not accessible.

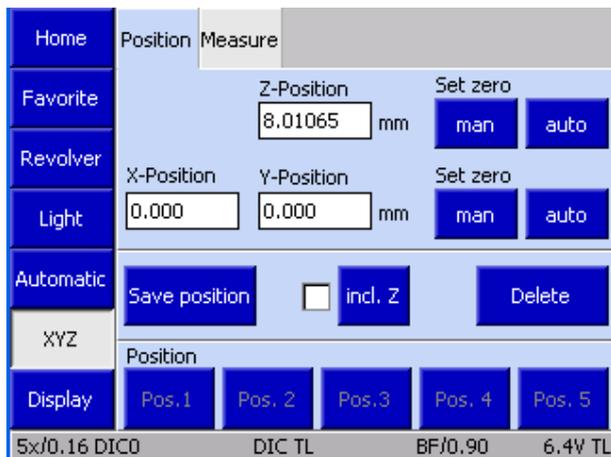


Fig. 4-51 Microscope -&gt; XYZ -&gt; Position

For XY and Z, the two pairs of **Set to Zero** buttons function as follows:

**man** Manual zero setting, i.e. the current position is defined as zero point and the display set to zero.

**auto** Automatic zero setting, i.e. the stage moves into the end position that was defined as zero point. Then, the display will be set to zero.

 Before the stage moves into the lower Z end position, the following popup window appears: "Caution! Remove specimen, before stage moves into end position!" Confirm the message with **OK**, if no specimen is on the stage or with **Cancel** to stop stage movement.

### Save Position

Using the **Save Position** button, the operator can define coordinate positions for the five position buttons at the bottom as follows:

- Move to the desired XYZ position.
- If the Z value shall be stored, activate the **with Z** check box.
- Press the **Save Position** button. This will bring up the **Current positions save as** popup window (Fig. 4-52).

In the window, five buttons, Pos.1 ... Pos.5, are arranged. If a button is used, the XYZ data appear as label, otherwise its number.

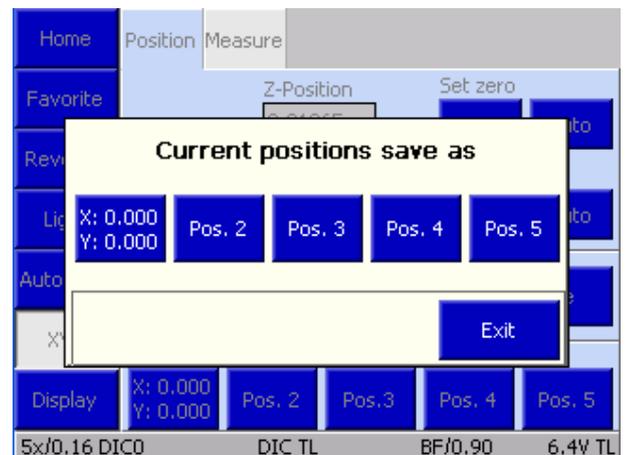
- Save the current position by pressing a position button. If coordinate data have already been assigned to this button, a safety query appears asking you if you want to overwrite the stored data.
- Close the window with the **Cancel** button.

To delete data, press the **Delete** button, select the position button and confirm the safety query with **Yes**.

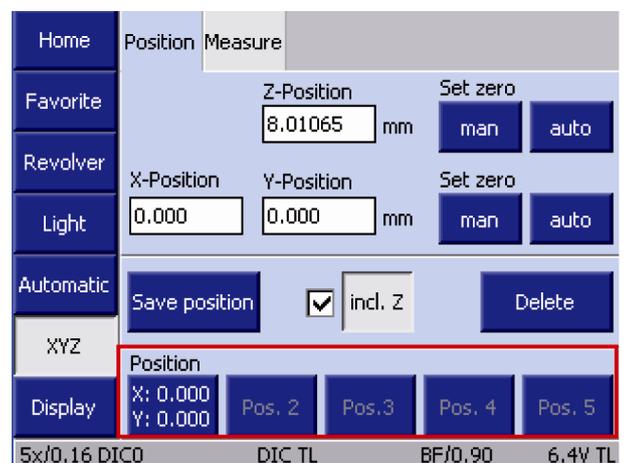
 The positions will only be saved temporarily, i.e., they will be available only as long as the microscope is switched on.

### Driving to stored position

In the bottom Position field, five buttons are arranged. To set the stored coordinate positions assigned to such a position button, press the respective button. The storage of coordinate positions is described above under **Save Position**.



**Fig. 4-52** Microscope -> XYZ-> Save current positions



**Fig. 4-53** Microscope -> XYZ -> Move to Position

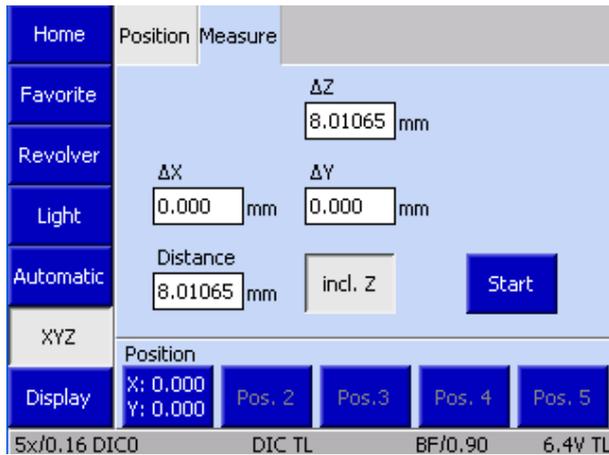


Fig. 4-54 Microscope -> XYZ-> Measure

## (2) Measure tab



This tab will be accessible only if you use a motorized (CAN bus) stage. Otherwise, the **Start** button and a display for the Z-distance  $\Delta Z$  are displayed on the **Position** tab.

Using the controls on the **Measure** tab, the operator can perform simple distance measurements in millimeters (mm). Three options are available for these measurements:

- Distance between two manually set positions
- Distance between a manually set position and a defined position
- Distance between two defined positions



If the Z distance shall be measured, activate the **with Z** button.

- Move to initial position.
- Press the **Start** button. On doing so, the display fields  $\Delta X$ ,  $\Delta Y$  and  $\Delta Z$  will be set to zero.

In the fields,  $\Delta X$ ,  $\Delta Y$  and  $\Delta Z$ , every stage movement is displayed.

The position buttons function as described under **(1) Position tab** above.

## 4.8.6 Settings page

The **Settings** page is accessible from the **Home** page by pressing the **Settings** button on the navigation button bar.

In diesem Teil der Software wird das motorische Stativ konfiguriert. Einige Angaben sind für die automatischen Funktionen Lichtmanager und Kontrastmanager unbedingt erforderlich. Hierzu gehören die Konfiguration der Objektive, Okulare sowie die Leuchtenkonfiguration in Auflicht und Durchlicht (siehe unten). Dies ist zwingend notwendig, da andere Funktionen (Umschaltung DL / AL, die Regelbereiche der Leuchten (HAL, LED und VIS-LED), Lichtmanagerberechnung und Kontrastmanager) hiervon abhängen.

The **Settings** page provides access to the following pages: **Components**, **Extras** and **Info**.

### 4.8.6.1 Components page

#### (1) Objectives

The tab allows the user to configure the objectives.

The tab shows up to seven buttons dependent on the actual number of objective mounts on the nosepiece. As long as you have not configured any objectives yet, the buttons are labeled only with the numbers of the nosepiece positions.

After you configured an objective position, the following objective data will be displayed: Designation of objective, Magnification, Numerical Aperture (NA), Immersion.

- Press the relevant button to configure a turret position.

Home	Objec- tives	Re- flector	Focus	Stage	Camera- ports	Misc
Favorite	1	EC PlnN 5x/0.16 DICO		5	Pln Apo 63x/1.4 Oil DICIII	
Com- ponents						
Extras	2	EC PlnN 10x/0.3 Ph1 DICI		6	EC EpiPlnN 50x/0.8 HD	
Info	3	EC PlnN 20x/0.5 Ph2 DICII				
Display	4	EC PlnN 40x/0.75 DICII			Automatic Detection	
		5x/0.16 DICO	DIC TL	BF/0.90	6.4V TL	

**Fig. 4-55** Microscope -> Settings -> Components -> Objectives

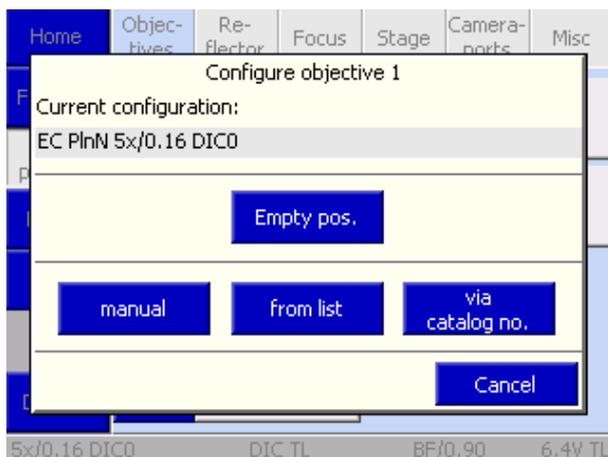


Fig. 4-56 Microscope -> Settings -> Components -> Configure objective

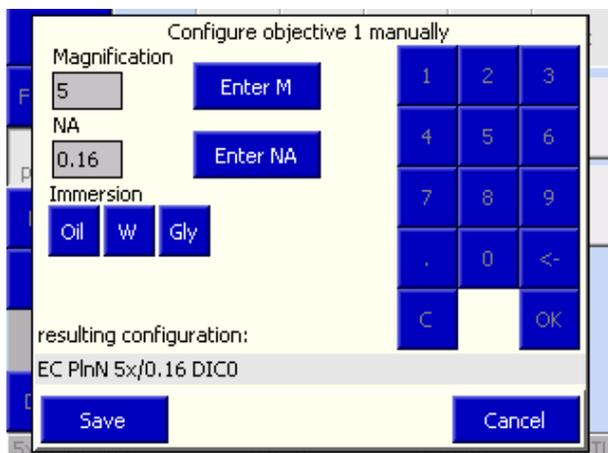


Fig. 4-57 Configuring objective manually

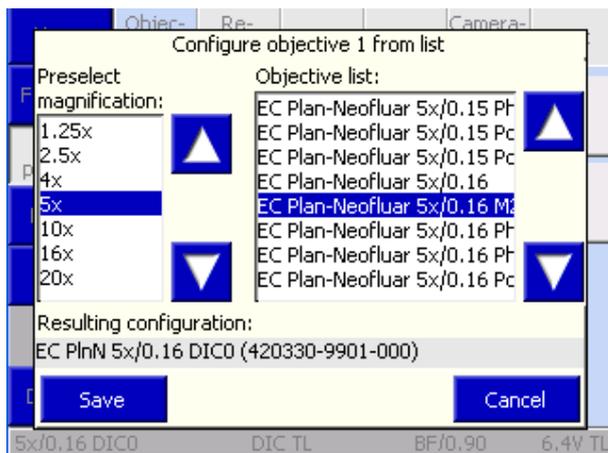


Fig. 4-58 Configuring of objective from list

In the **Configure Objective #** popup window, you can choose among various options:

- **Empty position** button  
Deletes the existing objective configuration at the position currently selected. Acknowledge the safety query with **Yes**.
- **Manual** button  
The user must enter magnification, numerical aperture (NA) and immersion on his / her own. (See Fig. 4-57.)  
To execute the configuration procedure, press the **Save** button. To abort the procedure, press the **Cancel** button.
- **From list** button  
The user chooses an appropriate objective from the **Preselect Magnification** list and the Objective list (Fig. 4-58).  
To execute the configuration procedure, press the **Save** button. To abort the procedure, press the **Cancel** button.

Several DIC prisms / sliders are allocated to some of the objectives suitable for differential interference contrast (DIC). Such allocation is executed when the selection of the objective is acknowledged.

- The DIC slider employed can be chosen in the following pop-up menu. If a motorized condenser is utilized, the condenser disk will automatically move to the correct condenser position as the contrast method is selected.

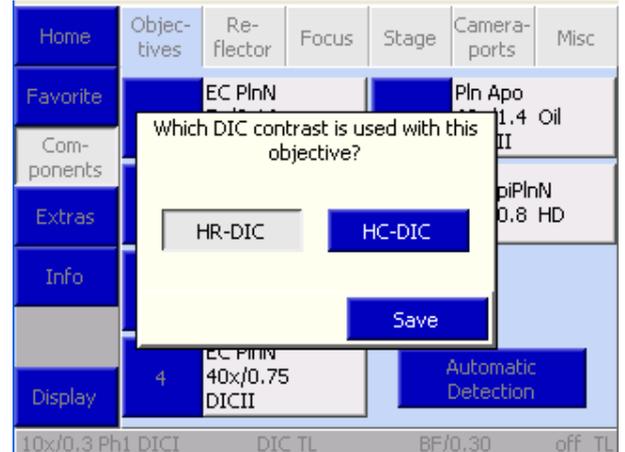


Fig. 4-59 Allocating the DIC slider

- **Via Cat. No. button**  
The user must enter the Zeiss reference number (XXXXXX-XXXX-XXX) to select an objective. To execute the configuration procedure, press the **Save** button. To abort the procedure, press the **Cancel** button.



When entering the 15-digit Zeiss Catalogue No., the leading six zeros or the following seven zeros (after 123456 enter a hyphen (-) or enter 1234-567 and press **OK**). The missing zeros will be added automatically.

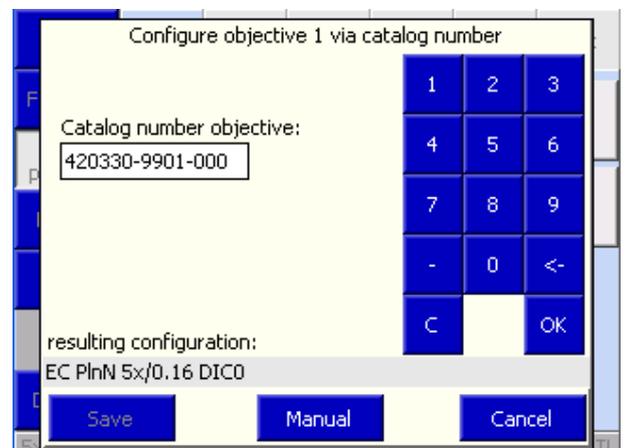


Fig. 4-60 Configuring the objective via reference number

Whenever a nosepiece and objectives with automatic component detection have been selected as stand equipment, the configuration of objectives may be triggered by pressing the **Automatic detection** button. The user does not have to perform any manual settings then.



Manually edited objective positions will not be overwritten by Automatic Component Detection. They must be entered manually as empty positions beforehand.



Once a new objective has been allocated, the relevant objective button on the **Microscope turrets** page will show the magnification and immersion data of that objective.

Home	Objec- tives	Re- flector	Focus	Stage	Camera- ports	Misc
Favorite	1	Pol TL - TL	6	43 HE DsRed RL -		
Com- ponents	2	DIC TL - TL	7	68 BFP RL -		
Extras	3	C-DIC RL RL -	8	02 DAPI RL -		
Info	4	C-DIC RL RL -	9	38 HE GFP RL -		
Display	5	20 HE Rhod RL -	10	64 HE mPlum RL -		

5x/0.16 DIC0      DIC TL      BF/0.90      6.4V TL

**Fig. 4-61** Microscope -> Settings -> Components -> Reflector

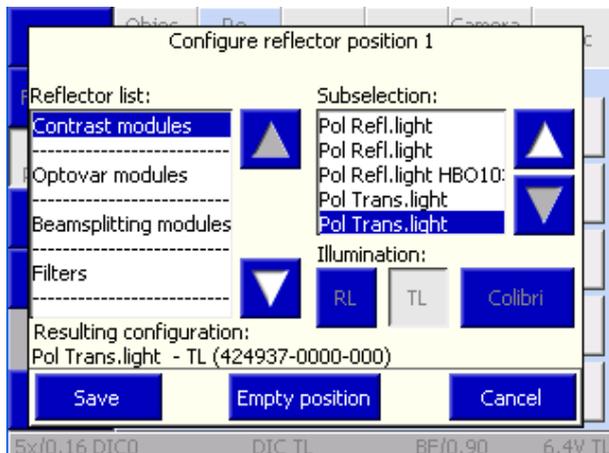
## (2) Reflector tab

With the controls on this tab, you can configure the reflector turret.

The tab shows up to ten buttons dependent on the actual number of positions on the reflector turret, which is detected by the system during initialization (and on activation of the **Settings/Components** page). As long as you have not configured any reflectors yet, the buttons are labeled only with the numbers of the turret positions.

After you configured a reflector position, the following data will be displayed: Designation (Type), Reflected-light module (RL), Transmitted-light position / module (TL).

- To start configuring a reflector turret position, press the corresponding button.
- In the **Configure reflector position #** popup window, you can choose the respective reflector from the list. In the **Resulting Configuration** line, you can read the current selection.
- Press the button for **RL** and/or **TL**, as appropriate.
- Press the **Save** button. If the turret position was already configured, a safety query will appear.



**Fig. 4-62** Configuring the reflector position

The type of module can first be chosen from the reflector list:

- Contrast module
- Optovar
- Beam splitter
- Filter
- Fluorescence filter set

Under **Contrast modules** and **Optovars**, the selection can subsequently be specified via the subselection options provided on the right-hand side.

The same applies to **Beam splitter**. Please note that additional laser safety functions are activated when LSM components are selected. For more details, see the LSM Operator Instructions.

Empty modules and RGB modules can be found in the **Filter** list. RGB filters are used, for example, when color images are to be generated with a monochrome camera via the multi-channel image recording module within the Axio Vision software. This option is used exclusively in transmitted light.

Instead of fluorescence filter sets, just dyes may be selected. If this is the case, the user would choose the relevant dye from the subselection options listed.

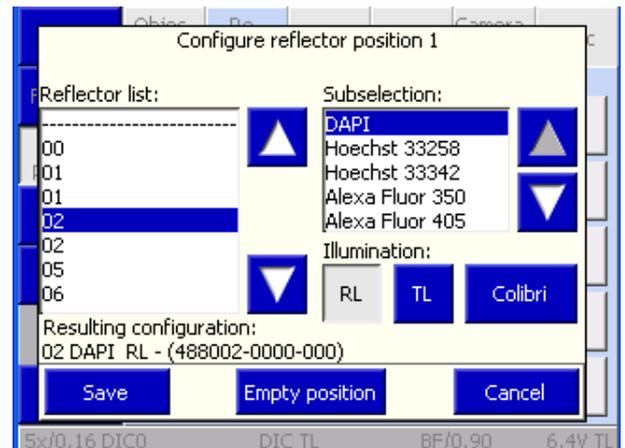
Fluorescence filter sets can be selected by their numbers. Additionally, a suitable dye may be picked for each filter set from the submenu.

 Once a new reflector has been allocated, the relevant reflector button on the **Microscope operation** page will show the corresponding information.

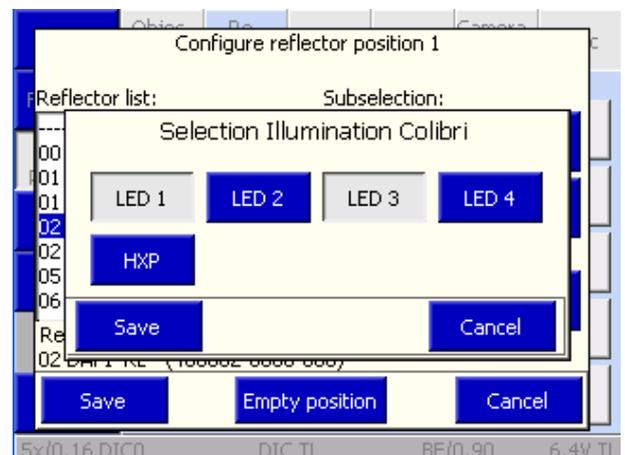
### Configuring a reflector position, using Colibri as a source of illumination

Once Colibri has been recognized and configured as a source of illumination, an appropriate LED can be selected for each filter set or dye.

- To do this, press the **Colibri** button.
- From the following pop-up menu (Fig. 4-64), pick one or several LEDs matching the reflector position selected.
- As an alternative, an optionally installed white-light source (HXP 120) can be picked as well. Note that LED and HXP cannot be chosen at the same time.
- Acknowledge the selection procedure by pressing **Save** or press **Cancel** to discard the selection.
- Acknowledge the selection procedure a second time by pressing **Save** or press **Cancel** to discard the selection.
- Acknowledge the safety query with **OK**.



**Fig. 4-63** Configuring the reflector position using Colibri



**Fig. 4-64** Selecting Colibri / HXP 120 illumination

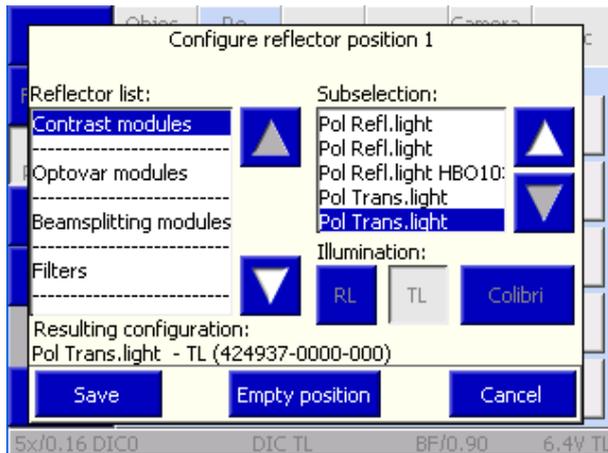


Fig. 4-65 Allocating the lamp port

- If a lamp-switching mirror is employed in the reflected-light path, the **Lamp port** to be used needs to be allocated to that mirror in the last configuration step.
- Press the **Save** button to acknowledge the selection made.



If the contrast manager is activated, the relevant Colibri LEDs will then be switched on when this reflector position is selected.

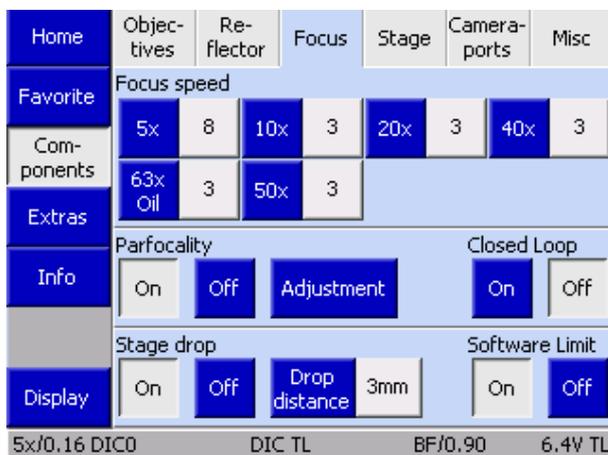


Fig. 4-66 Microscope -&gt; Settings -&gt; Components -&gt; Focus

### (3) Focus tab

On this tab, you can enter the firmware settings for the focusing drive. The speed of the focusing drive is adjustable individually for every objective.

Furthermore, **parfocality**, **linear sensor focus**, **stage lowering** and **electronic limit switches** can be set on this tab.

#### Focus Speed

Up to seven buttons are displayed dependent on the actual number of positions on the nosepiece, which is determined by the system during initialization (and on activation of the **Settings/Components** page). As long as you have not configured any objectives yet, the buttons are labeled only with the numbers of the nosepiece positions.

After you assigned an objective to a specific button, the magnification appears on the left, blue half of the button. The right, gray half of the button is labeled with the focusing speed.

- If you want to change the focusing speed for an objective, press the respective gray part of the button.
- In the **Focus Speed for Objective #** popup window, set the desired speed using the ◀▶ buttons. The higher the numerical value, the higher is the set speed.
- Press the **Save** button.

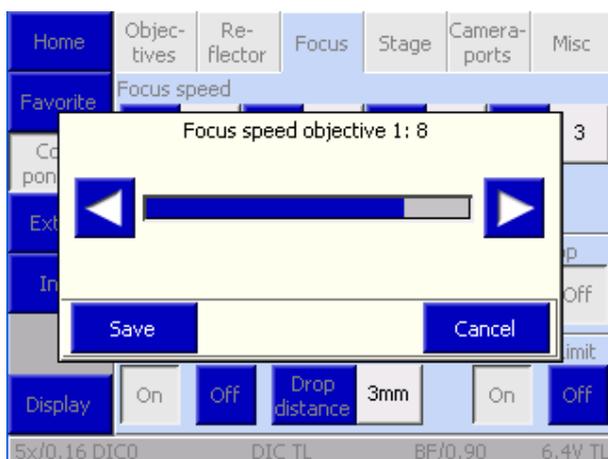


Fig. 4-67 Setting the focusing speed

## Parfocality

The parfocality function is activated or deactivated by means of the **ON** and **OFF** buttons.

The parfocality can be configured via the **Adjustment** button. On pressing this button, a wizard appears guiding you through the configuration procedure.

All objectives must be focused successively. Start with all dry objectives from the highest to the lowest magnification. Then, proceed with all immersion objectives from the highest to the lowest magnification. By pressing the **Next Objective** button, the nosepiece rotates to the next objective. After all objectives were focused, press the **End** button.

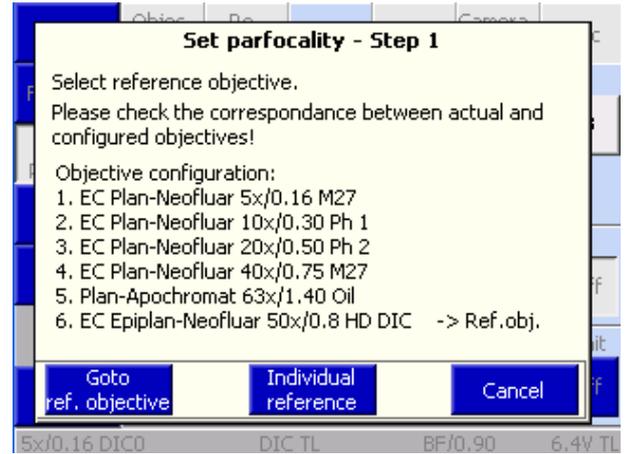


Fig. 4-68 Setting parfocality



If no TFT is installed (Axio Imager M2p), parfocality may also be adjusted manually. For this to be possible, the objective must have been successfully configured with the aid of the MTB 2004 configuration program. Carry out the following steps to this end:

- Activate the programming mode by keeping the **LM Set** button depressed for a while (> 3 s – acknowledgment by a long drawn-out dual beep tone). While the programming mode is active, every second LED on the LED bar graph will be lit up. To start parfocality adjustment, press the **RL** button on the light control.
- Swivel in the reference objective. This is usually the objective with the highest magnification and with air immersion. When it comes to a coded nosepiece, the position to be moved to will be displayed through the bar graph LED. If a motorized objective nosepiece is used though, this objective is swiveled in automatically.
- Focus on your sample and then press one of the buttons located in the middle of the pushbutton ring on the stand.
- Change to the next magnification displayed. This will happen automatically if a motorized nosepiece is used.
- Repeat steps 3 and 4 until all the objectives have been adjusted. A tone will be sounded as acknowledgment.
- Complete the procedure by pressing the LM Set button again. (Acknowledgment by a single long drawn-out beep tone. Bar graph LED returns to standard display.)

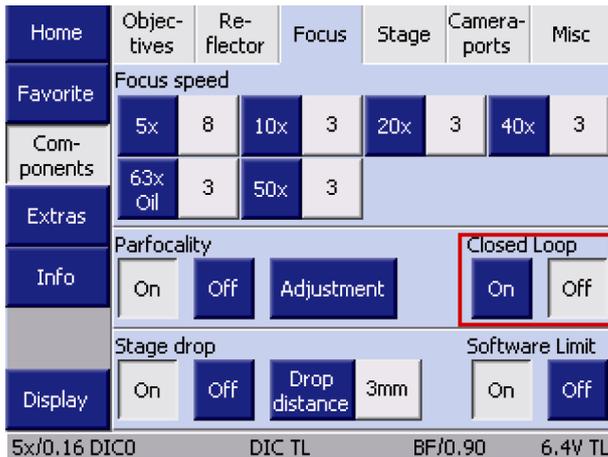


Fig. 4-69 Closed Loop

### Closed Loop

To monitor the stability of the focusing drive, a focus linear sensor can be used for the Axio Imager.Z2 / m. If available, this sensor can be switched on and off here.

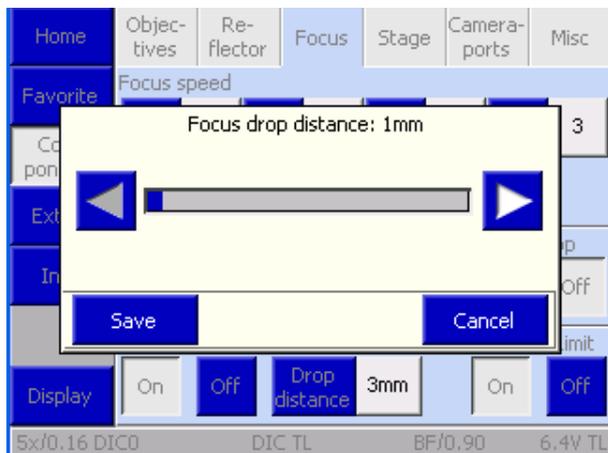


Fig. 4-70 Stage lowering

### Stage lowering

Stage lowering function is activated or deactivated via the **ON** and **OFF** buttons.

The lowering distance can be set by the ◀▶ buttons in 1 mm increments within a range of in 1 mm to 20 mm. To accept the value set, press the **Save** button. To discard it, press the **Cancel** button.

### Software limit (Focus delimiter)

Second-generation motorized Axio Imager stands are provided with software-based focus limiters, which may be set by the users themselves. They can be globally switched **ON** or **OFF** here.

Once the software limit function is active, the focus delimiter can be set and shifted also via the Work / Load buttons on the stand.

- To set the focus limit:  
Keep the upper button depressed for more than 3 seconds. (A beep tone will be sounded as acknowledgment.)
- To shift the focus limit:  
Keep the upper button depressed for more than 3 seconds. (A beep tone will be sounded as acknowledgment.) Simultaneously, move the focus to the new position desired. When the button is released, the current focus position will be saved and acknowledged with a message appearing on the TFT.
- To delete the focus limit:  
Press the **OFF** button on the TFT.

**(4) Stage tab**

If a motorized microscope stage (equipped with a CAN bus controller) is used, this tab will be displayed. Once an objective has been configured, the stage is assigned a defined travel speed for the objective position concerned.

Pressing one of the position buttons allows the travel speed of the motorized stage to be adapted separately to the objective magnification concerned via the ◀▶ buttons.

With the **Stage in load position** function, the user can activate or deactivate the XY stage movement for the load position / operating position. This has an effect on the functionality of the **Load position** button available on the **Microscope operation** page.

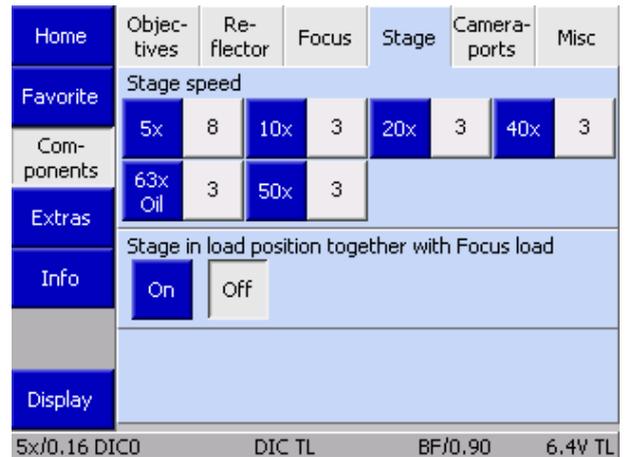
When the function is activated, the focusing drive will first move into the load position, with the stage subsequently traveling forward toward the user. As a result, the specimen can be easily changed.

**(5) Camera ports tab**

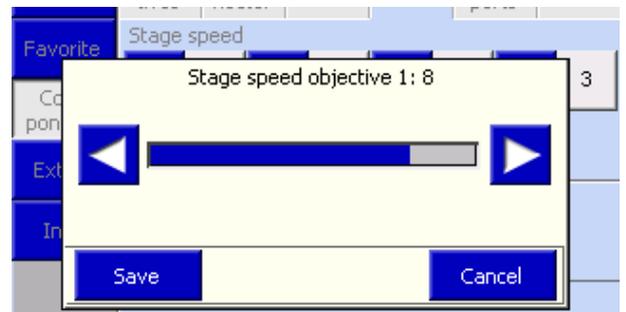
This tab allows you to configure the adapters and beam splitters (beam-splitting mirrors / reflecting mirrors) for the camera ports (Top Port / Back Port / Deflection Plane).



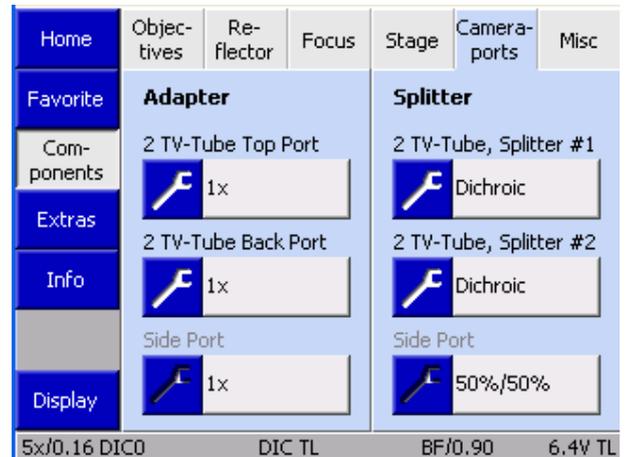
Before attaching/removing the camera path deflection, left (Deflection Plane) or before any tube change, switch off the microscope to ensure that the system detects the correct port status during the next initialization process.



**Fig. 4-71** Mikroskop -> Settings -> Components -> Stage



**Fig. 4-72** Setting the stage speed



**Fig. 4-73** Mikroskop -> Settings -> Components -> Camera ports

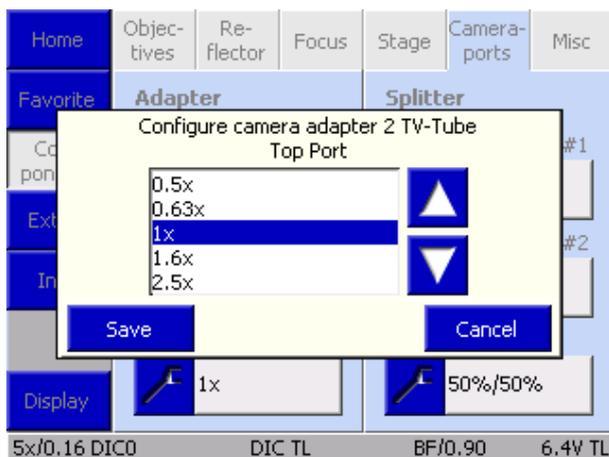


Fig. 4-74 Selecting the camera adapter

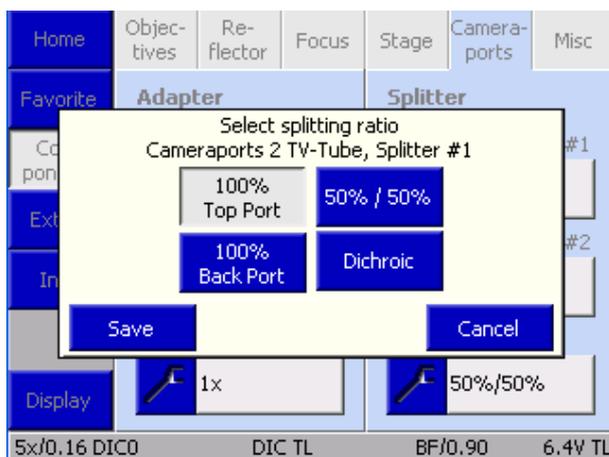


Fig. 4-75 Selecting the splitting ratio

## Adapter

Up to three buttons are displayed here dependent on the equipment of the camera path deflection and the tube used. The status of the ports is detected by the system during initialization (and on activation of the **Settings/Components** page).

- To assign an adapter to a button, press the gray button. This will bring up the **Select Camera Adapter** list (Fig. 4-74).
- Choose the installed adapter from the list using the ▲ ▼ buttons.
- Press the **Save** button, to assign the selected adapter to the port. Press the **Cancel** button to close the window without any selection.

The button is now labeled with the magnification factor. Proceed analogously for the other ports.

## Splitter

Up to three buttons are displayed here dependent on the equipment of the camera path deflection and the tube used. The status of the ports is detected by the system during initialization (and on activation of the **Settings/Components** page).

- To select a splitting ratio, press the gray button. This will bring up the **Select Splitting Ratio** list.
- Press the buttons for the desired splitting ratio; multiple selections are possible.
- Press the **Save** button, to store the selected splitting ratio. Press the **Cancel** button to close the window without any selection.

The button is now labeled with the splitting ratio. Proceed analogously for the other ports.

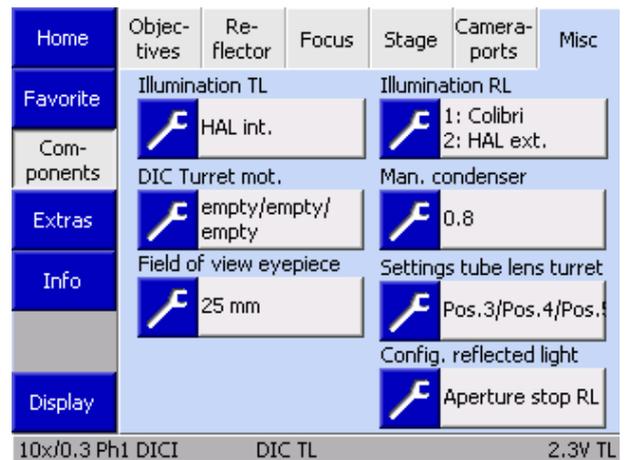


The configured splitting ratios will be offered then for selection on the **Light Path** tab of the **Microscope/Operation** page.

**(6) Miscellaneous tab**

Under this tab, the user can configure other optional components of transmitted-light and reflected-light illumination.

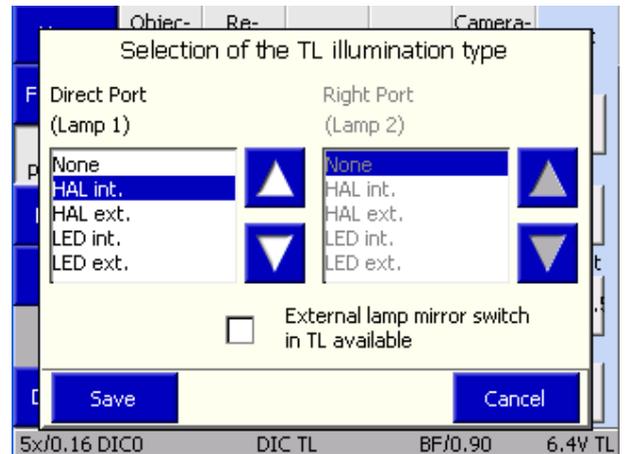
- TL illumination
- RL illumination
- Motorized DIC turret
- Manual condenser
- Field of view of eyepieces
- Tube lens turret
- Reflected-light configuration



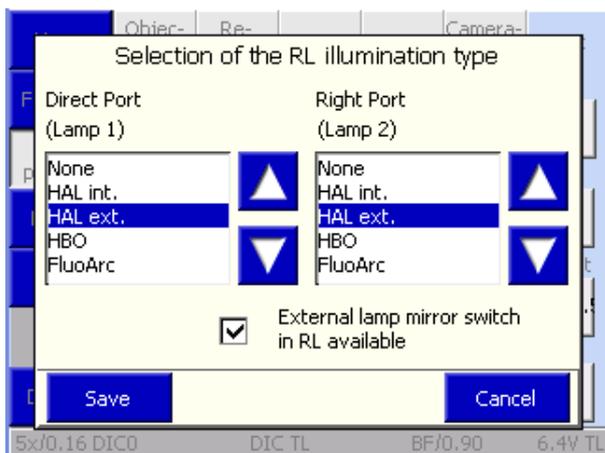
**Fig. 4-76 Microscope -> Settings -> Components -> Misc(ellaneous)**

**TL illumination**

The light source to be employed in transmitted light must be specified here. Optionally, a switching mirror for two illuminators may be used, too. To this end, the appropriate check box must be ticked.



**Fig. 4-77 TL illumination**



**Fig. 4-78 RL illumination**

## RL-illumination

The light source to be employed in reflected light must be indicated here.

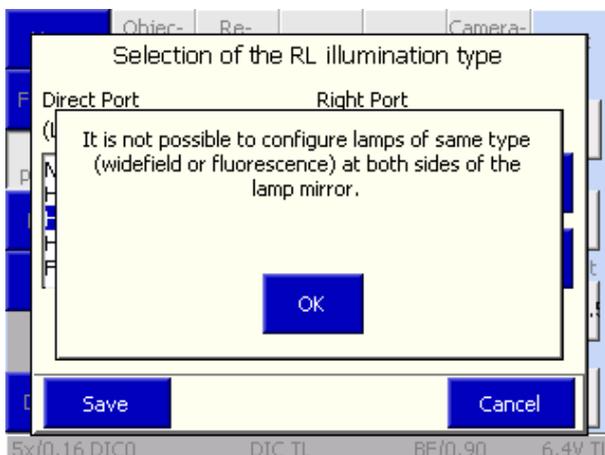
If a switching mirror for two illuminators is used, this mirror will appear in this configuration menu following successful initialization. Operation is effected either via the function buttons (for allocation, see below), or the function concerned is automatically linked with the selection of an allocated reflector module, so that the user does not have to pay attention to any additional components.

Sources of illumination:

- None
- HAL int.
- HAL ext.
- HBO
- FluoArc
- HXP
- Colibri
- LED int.
- LED ext.
- VisLED int.
- VisLED ext.

The addition "internal" or "external" denotes that the component is hooked up either to the power supply incorporated in the stand or to an external CAN bus power supply connected to the stand via CAN bus.

Only types of lamps that are different can be configured. An error message will appear in the event of dual allocation.



**Fig. 4-79 RL illumination error message**

### Motorized DIC turret

Modulator turret allocation for transmitted-light DIC. This is where the user selects the appropriate DIC prisms for turret positions 2, 3 and 4.

**Important:** The contrast manager can only adjust the DIC contrast correctly if the analyzer module specified for the motorized DIC turret has been selected in the reflector turret.



Motorized C-DIC modulator turret for material:

No settings are to be made here, as the positions for C-DIC, TIC and brightfield are always fixed.

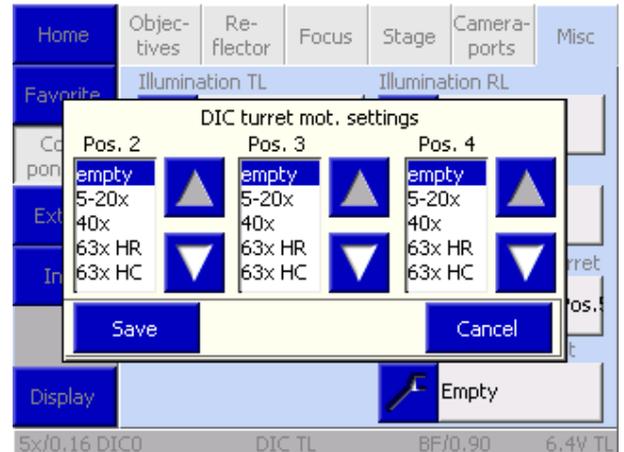


Fig. 4-80 Motorized DIC turret

### Field of view of eyepieces

The user chooses the eyepieces employed via the **Eyepiece field of view selection** pop-up window.

This is important since the correct scaling of the aperture of the motorized stop slider depends on this information.



Fig. 4-81 Field of view of eyepieces

### Manual condenser

If you use a manual condenser, but a motorized luminous-field diaphragm, you should enter the condenser aperture here. This is necessary, as the Light Manager automatically adapts the luminous-field diaphragm to the field of view of the eyepieces, and the various condensers have different magnification factors.

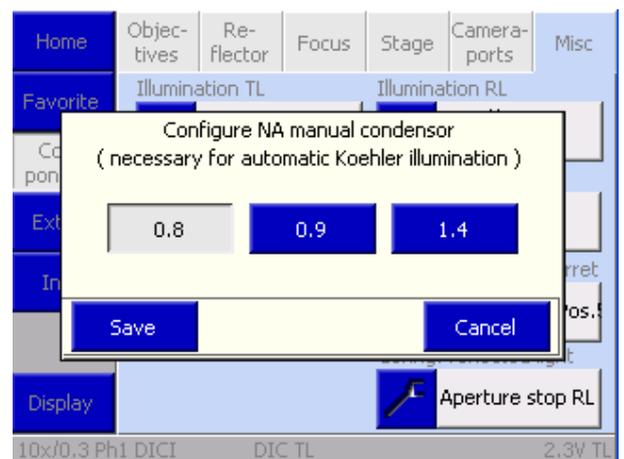


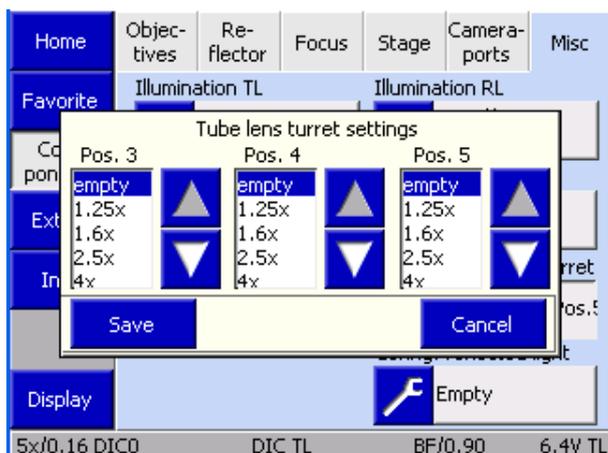
Fig. 4-82 Manual condenser

 With the second-generation Axio Imager, it is possible to configure a number of positions for the motorized condenser. This is done, however, within the MTB 2004 configuration program, with the configuration written directly into the condenser. The following options are available:

- Plate – Serves as a shutter when there is no shutter in the transmitted-light path and an LED light source is used on the lamp socket or when an LED light source is mounted directly under the condenser.
- Polarizer
- DIC prisms

Any condenser configured by the user in this way is fully integrated in the light and contrast manager. This means the following:

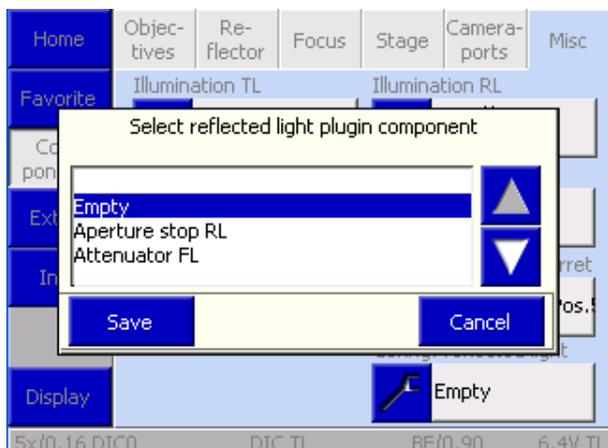
- Plate: When the contrast manager is used, any front lens existing is swiveled out, with the plate moved in front of the transmitted-light path as soon as the fluorescence function is selected.
- Polarizer: When the POL contrast is selected in transmitted light, the polarizer is automatically moved into the light path.
- DIC prisms: The correct DIC prism is swiveled in for the objective chosen.



**Fig. 4-83 Lens turret**

### Lens turret

The tube **Lens turret allocation selection** list allows the user to choose the appropriate magnification factor for turret positions 2, 3 and 4.



**Fig. 4-84 Reflected-light configuration**

### Reflected-light configuration

Whenever a motorized stop slider or an attenuator is used in reflected light, the component in question needs to be specified here. The settings are transferred through a restart of the instrument.

#### 4.8.6.2 Extras page

From the Extras page, you can get to the **Light manager**, **Contrast manager**, **Oil stop**, **Dazzle protection**, **Ethernet** and **Miscellaneous** tabs.

##### (1) Light manager tab

This tab allows the user to switch the light manager on and off.

With the aid of the light manager, light intensity is adjusted automatically. For its uses, see Section 4.7.

Buttons accessible under **Load light manager values**:

The **User-defined** button serves to load that the values last saved via the **LM set** button.

Pressing the **Default** button will load the factory settings back in.

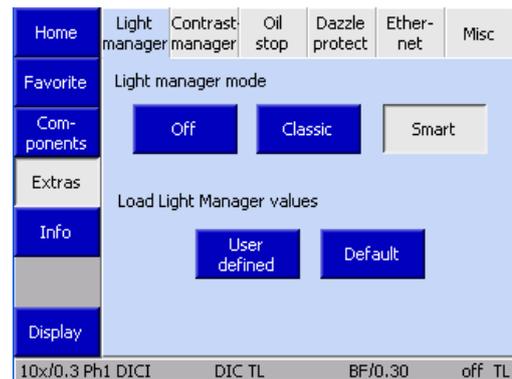


Fig. 4-85 Microscope -> Settings -> Extras -> Light manager



New filter set combinations for transmitted light and reflected light (neutral-density filters, as well as color and neutral-density filters) are offered for second-generation Axio Imagers. Those filters are fully integrated into the light manager functions. For the sake of clarity, the MTB 2004 configuration program writes those filters directly into the filter wheel, without the user having to do so through the TFT. What is more, this component is an ACR-enabled component. This means that the equipment mounted on it is automatically read out by the microscope during initialization. For this reason, a filter wheel of this type can be installed on every stand without having to be reconfigured.

##### (2) Contrast manager tab

This tab serves to switch the contrast manager globally **ON** and **OFF**. These functions come in handy for the user whenever contrast-dependent motorized components are not to be moved automatically.

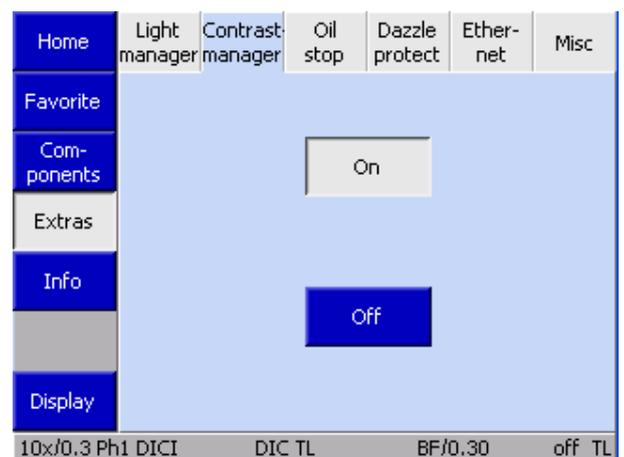
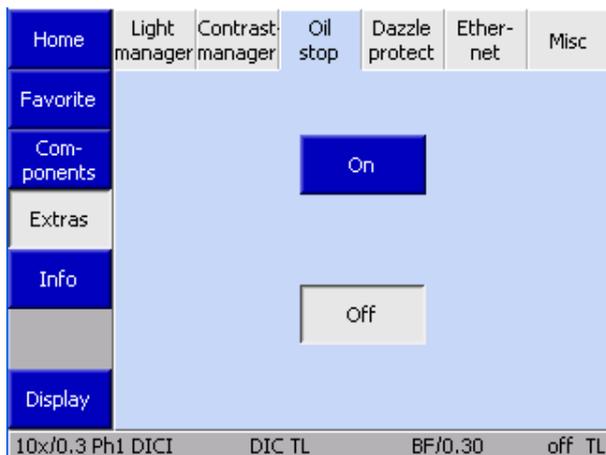


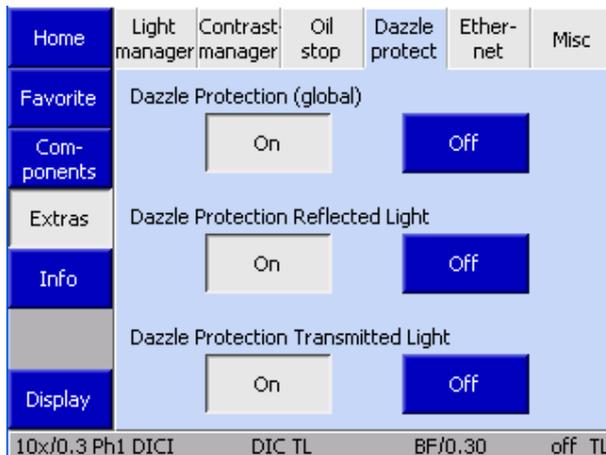
Fig. 4-86 Microscope -> Settings -> Extras -> Contrast manager



**Fig. 4-87** Microscope -> Settings -> Extras -> Oil Stop

### (3) Oil Stop tab

On this tab, you can activate or deactivate the Oil Stop function. This function prevents a dry objective from being swiveled into immersion liquid by lowering the stage whenever you switch from an immersion to a dry objective.



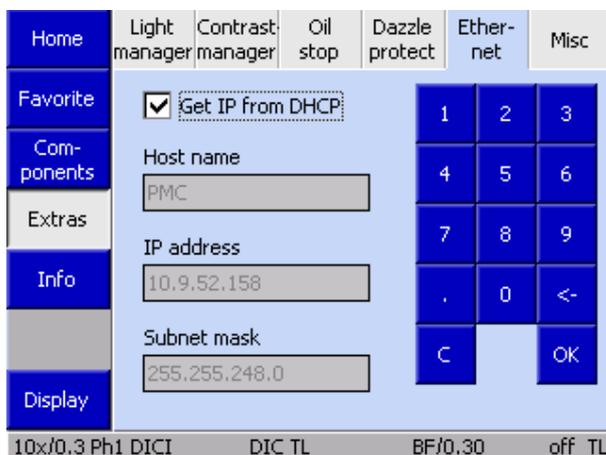
**Fig. 4-88** Microscope -> Settings -> Extras -> Dazzle Protection

### (4) Dazzle protection tab

When you deactivate the Dazzle Protection function globally, all other options on this tab appear grayed out.

If a Bino Shutter is installed and dazzle protection has been activated, the two other dazzle protection fields (TL/RL) are inactive. Dazzle protection is, therefore, deactivated through these components.

If one of the above components is not installed, the corresponding buttons are not available.



**Fig. 4-89** Microscope -> Settings -> Extras -> Ethernet

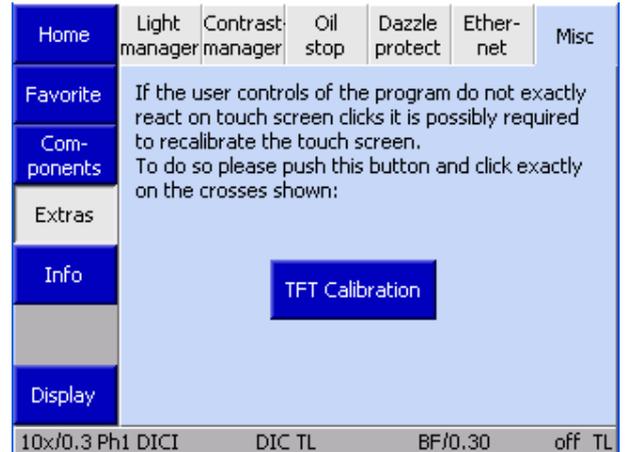
### (5) Ethernet

On this tab, you can choose settings for the connection of the Axio Imager via Ethernet.

For a manually entered IP address to be accepted, acknowledgment with **OK** is required.

**(6) Miscellaneous**

This tab allows you to calibrate the TFT display.

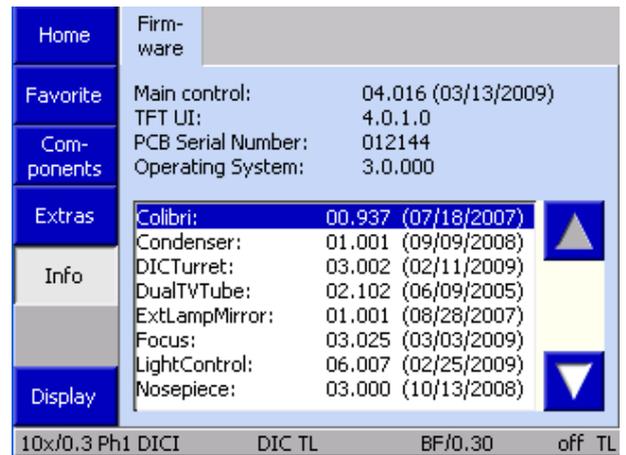


**Fig. 4-90 Microscope -> Settings -> Extras -> Misc(ellaneous)**

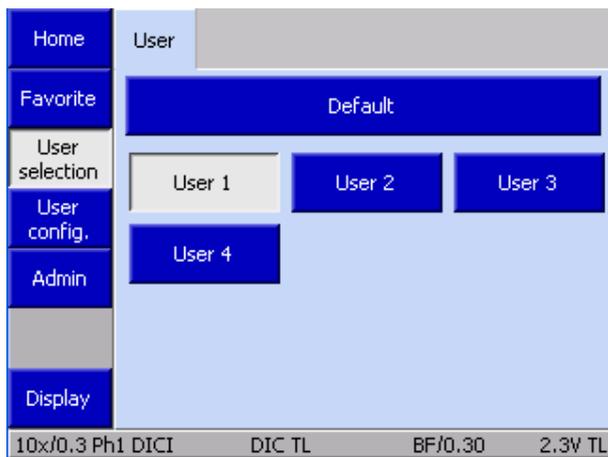
**4.8.6.3 Info page**

The **Settings-Info** page only contains the **Firmware** tab

This tab allows the user to identify the firmware version.



**Fig. 4-91 Microscope -> Settings -> Info -> Firmware**



**Fig. 4-92** Microscope -> User -> user selection

#### 4.8.7 User page

From this page, the user can access three other pages:

- **User selection**
- **User configuration**
- **Admin**

##### 4.8.7.1 User selection page

The **User selection** page contains a tab which serves to select a previously created and configured user.

A default user may be chosen as well. This user contains the Standard settings for the assignment of the buttons located on the instrument and the docking station, based on an automatic assignment plan. Hardware settings may be assigned and are retained after switch-off. Light manager values will be saved.

#### 4.8.7.2 User configuration page

On this page, user-defined configurations can be created and saved. This page is protected by password.

Upon entry of the user password, the configuration for the user chosen on the **User selection** page can be adapted.

The following tabs are available for user configuration:

- **HW Settings** Hardware settings
- **Settings** Change of password
- **Buttons on the left** Left drive knob
- **Buttons on the right** Right drive knob
- **Docking station buttons**
- **Favorites page** Selection of favorites page

##### (1) HW Settings tab

As many as ten hardware settings can be assigned to each user. Each setting may govern a combination of controllable microscope components. The components available appear in a selection list.

- Click on the components desired.  
Clicking on a given component will save its current status for the setting. If a value deviating from that status is to be saved, the value must be changed via the buttons on the stand or via the TFT, before it can be saved.
- To adopt the new value, press the **Update** button afterwards.

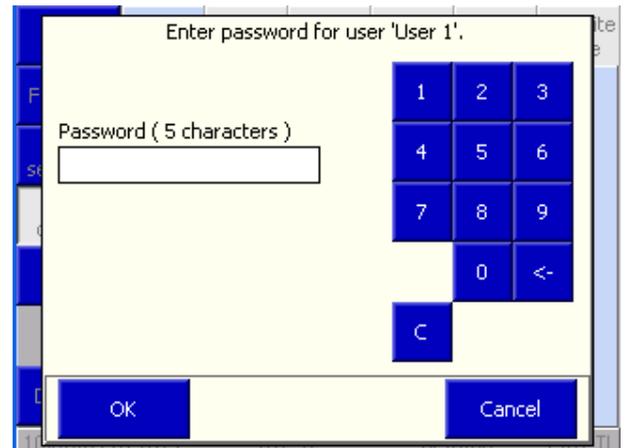


Fig. 4-93 Password query

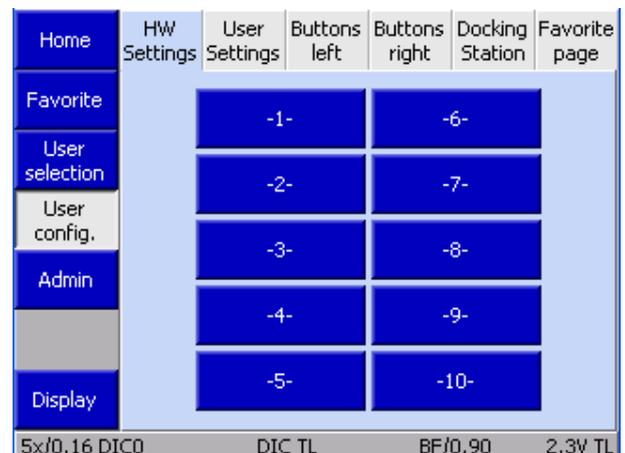


Fig. 4-94 Microscope -> User -> User configuration -> HW Settings

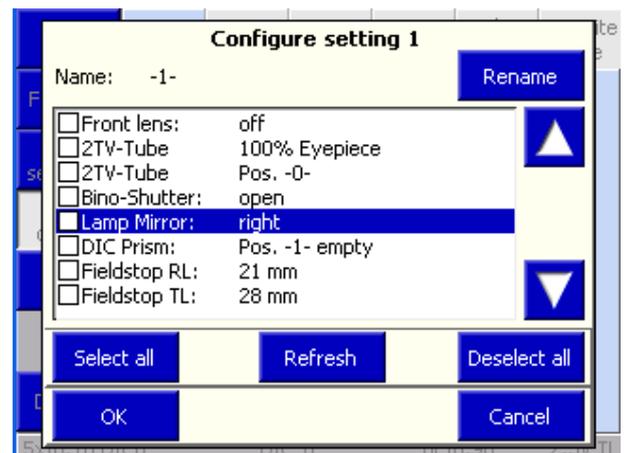


Fig. 4-95 HW Settings list

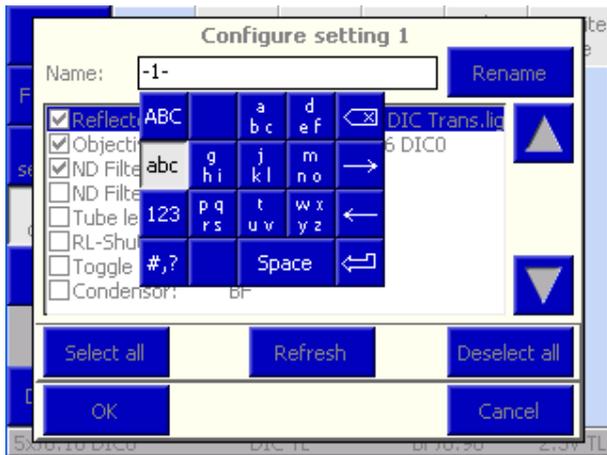


Fig. 4-96 Renaming HW Setting

Each setting can be given an individual name.

- Press the **Rename** button.
- Enter the name and acknowledge by pressing the **ENTER** button on the keyboard.
- Complete this procedure by pressing the **OK** button or discard the entry by pressing the **Cancel** button.

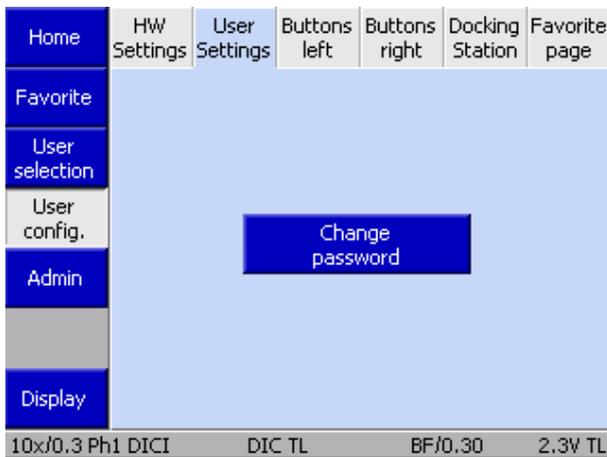


Fig. 4-97 Microscope -&gt; User -&gt; User configuration -&gt; Settings

## (2) Settings tab

This tab allows the password for the user to be changed.

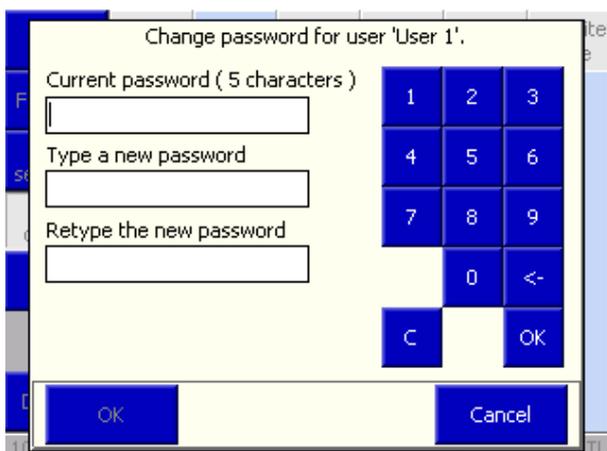


Fig. 4-98 Changing the password

- Press **Change password**.
- Enter the old password.
- Enter the new password (which must be at least five characters long).
- Repeat the new password.
- Acknowledge by pressing the **OK** button on the numeric pad.
- To execute the change, press the **OK** button or discard the change with the **Cancel** button.

### (3) Left buttons tab

On this tab, the control buttons on the left-hand side of the stand can be assigned.

- Click on the **Top left** button pair.
- Use the arrow buttons to select the desired assignment.
- Acknowledge with **OK** or discard with **Cancel**.

Proceed analogously for all other button assignments.

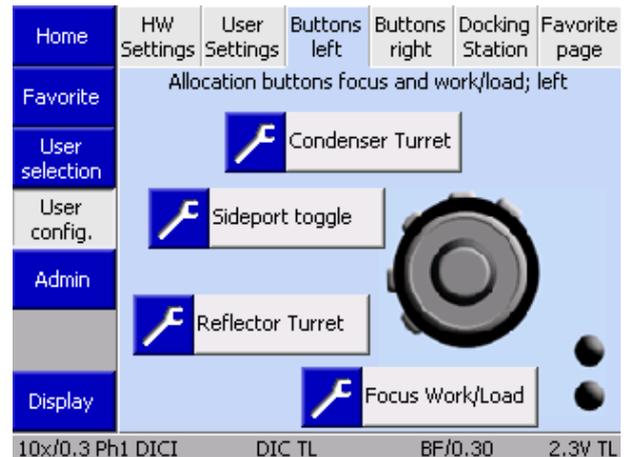


Fig. 4-99 Microscope -> User -> User configuration -> Left buttons

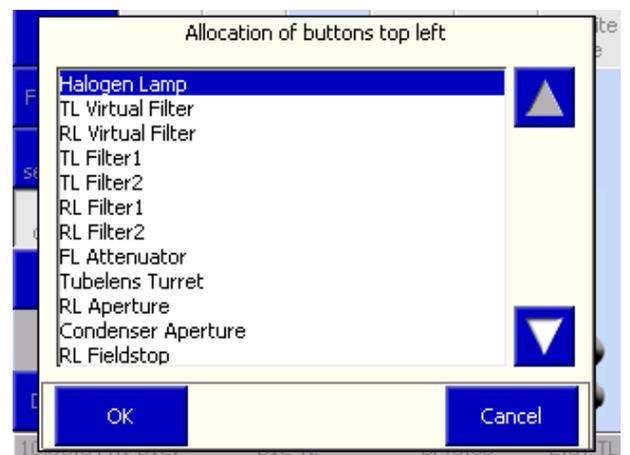
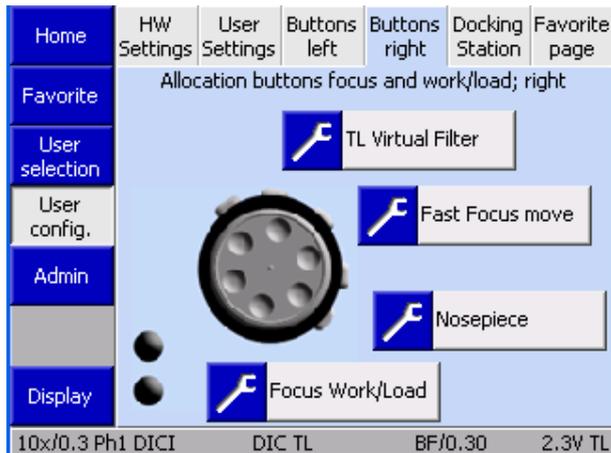
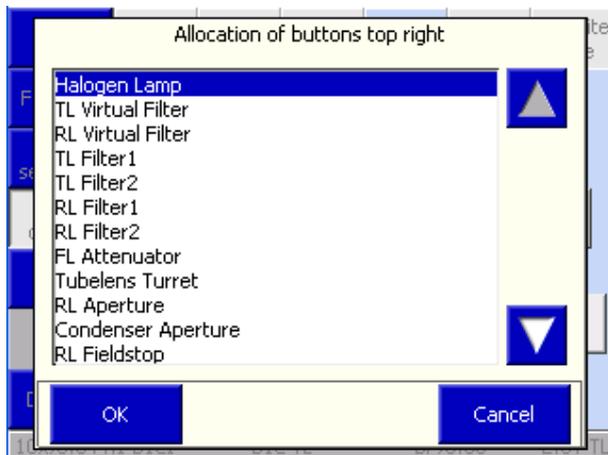


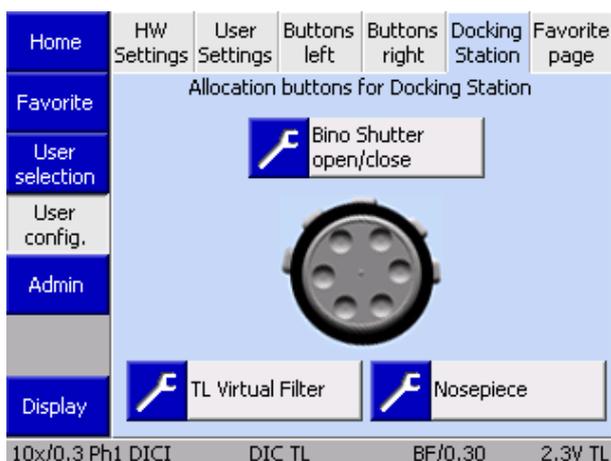
Fig. 4-100 Assignment of top left button pair



**Fig. 4-101** Microscope -> User -> User configuration -> Left buttons



**Fig. 4-102** Assignment of top right button pair



**Fig. 4-103** Microscope -> User -> User configuration -> Docking Station

#### (4) Top right tab

On this tab, the control buttons on the right-hand side of the stand can be assigned.

- Click on the **Top right** button pair.
- Use the arrow buttons to select the desired assignment.
- Acknowledge with **OK** or discard with **Cancel**.

Proceeds analogously for all other button assignments.

#### (5) Docking Station tab

For the assignment of the buttons on the docking station, proceed analogously to Sections (2) of Top left buttons tab and (3) of Top right buttons tab.

## (6) Favorites page tab

- On this page, the user can define one of the tabs listed as a target to be jumped to directly.
- Use the arrow buttons to select the Favorites page.
- To acknowledge, press the **Adopt page as favorites page** button.  
**Important:** This function is executed without any safety query.

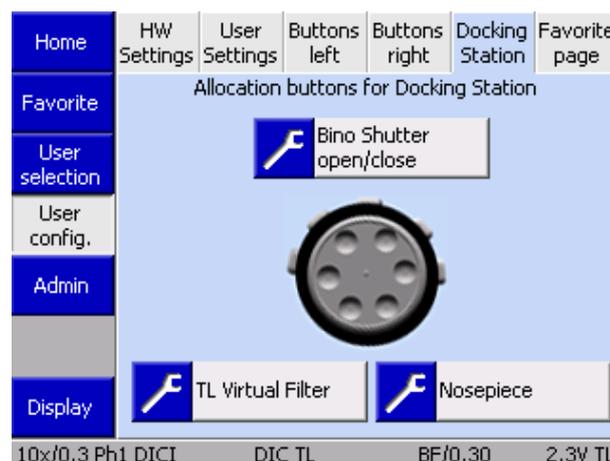


Fig. 4-104 Microscope -> User -> User configuration -> Favorites pages

## 4.8.7.3 Admin page

There are two tabs on the **Admin page**:

- Settings
- User administration

### (1) Settings tab

The following settings can be defined on this tab:

- Language
- Active illumination after switch-on
- Start page after switch-on
- Initialization of ACR objectives after switch-on

### Language

**German** or **English** can be selected here.

Pressing the **Save** button results in the instrument being restarted. After the restart, the microscope is initialized with the language selected.

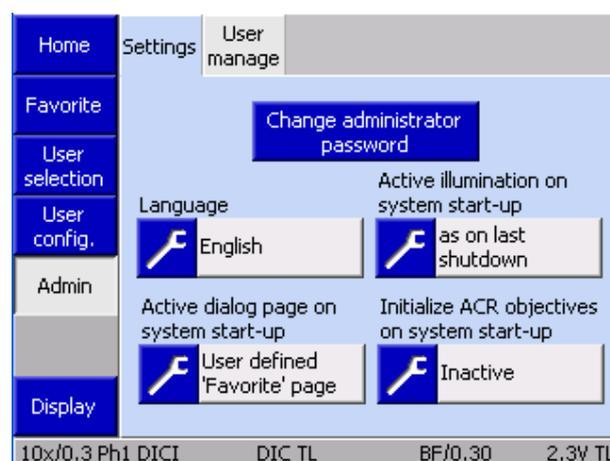


Fig. 4-105 Microscope -> User -> Admin -> Settings

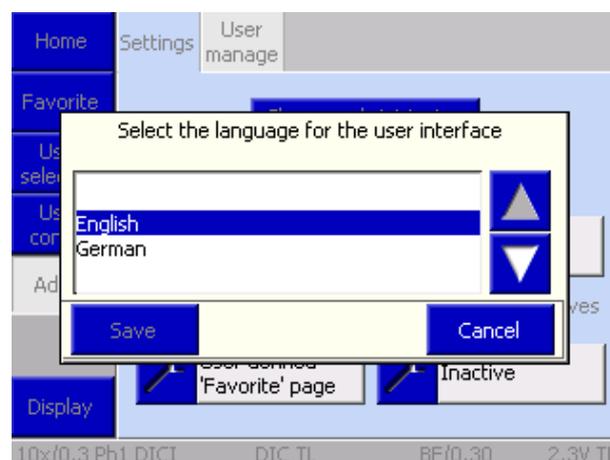


Fig. 4-106 Select language

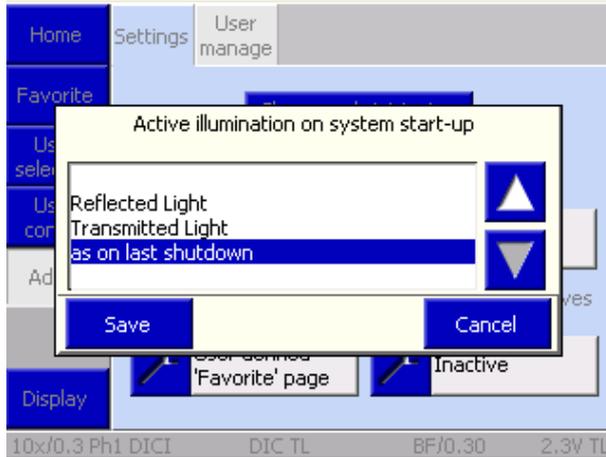


Fig. 4-107 Light path upon switch-on

**Active illumination after switch-on (light path)**

Here, the user can select the light path which is to be active when the microscope is switched on.

- **Reflected light:**  
The microscope starts with the reflected-light illumination previously switched on.
- **Transmitted light:**  
The microscope starts with the transmitted-light illumination previously switched on.
- **Turn-off state:**  
The microscope starts with the state in which it was turned off.

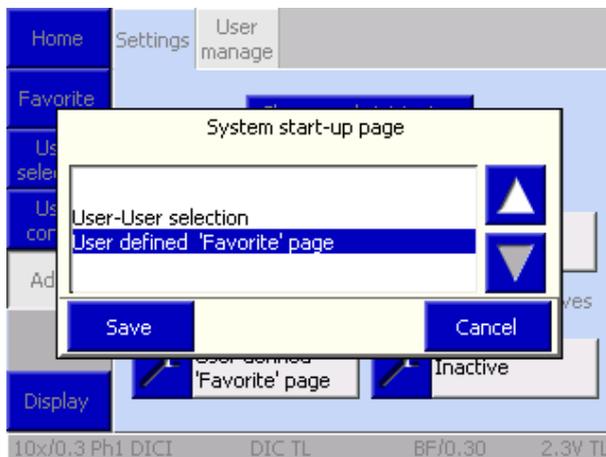


Fig. 4-108 Start page after switch-on

**Start page after switch-on**

The page shown upon the start the microscope can be defined here. There are two options:

- **User selection**  
Primarily of interest for multi-user operation.
- **Favorites page**  
Particularly useful whenever a specific operator's side is preferred.



Fig. 4-109 Initialization of ACR objectives after switch-on

**Initialization of ACR objectives after switch-on**

There are two options:

- **Active**  
When the instrument is started, a recognition run may (!) be triggered by the user. A safety query is displayed prior to the recognition run.
- **Inactive**  
The recognition run is triggered only upon specific instruction given in the settings menu.

## (2) User administration tab

On this tab, the administrator can create, administer and preconfigure users.

Upon pressing the **User #** button, a pop-up menu appears, which offers the following functions:

- User activation / deactivation
- User's name
- Allocation of password
- Adoption of user settings

If user settings are to be transferred from a user already created, the **Adopt user settings** checkbox must be activated.

By pressing on factory settings, you will then get to another pop-up menu. The user, whose settings are to be adopted, can be chosen here.

**Important:** Such adoption will work only from users previously configured.

- After adoption, the new user must be activated by clicking on the checkbox.
- Pressing the Save button completes this operation.

The new user can now be picked from the **User selection** page and be configured further, if necessary.

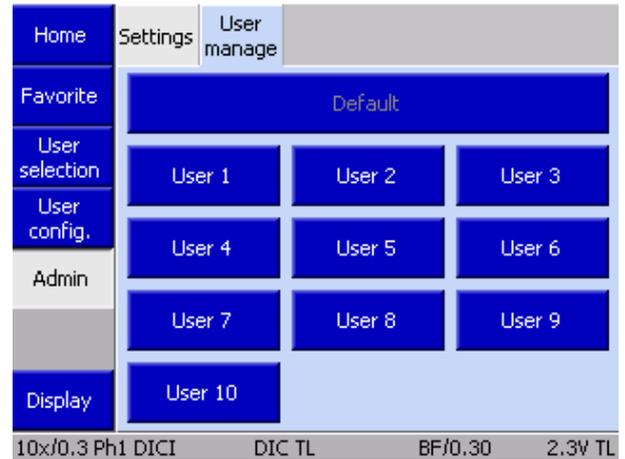


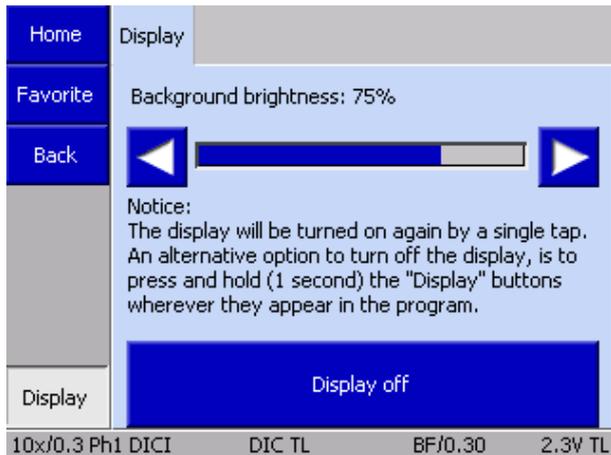
Fig. 4-110 Microscope -> User -> Admin -> User administration



Fig. 4-111 User administration for User 3



Fig. 4-112 Adopting user settings



**Fig. 4-113** Home -> Display

#### 4.8.8 Display page

Using the ◀▶ buttons on the **Display** page, the user can adjust the brightness of the TFT display and of the light control LEDs..

Pressing the **Display off** button turns the TFT display off. Pressing that button a second time switches the TFT display on again.

To dim the TFT display, press the **Display** button on the navigation bar for more than a second. Touching the TFT display again anywhere switches the display on again.

 After switching the TFT display off, the display returns from the **Display** page to the page from which it was activated. This page will be shown after switching the display on again.

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## 4.9 Illumination and contrast methods

### 4.9.1 Setting transmitted-light brightfield according to KÖHLER

#### (1) Application

Transmitted-light brightfield microscopy is the most common of all optical microscopic techniques, as it permits high-contrast or stained specimens (e.g. blood smears) to be viewed easily and quickly.

Beside the so-called direct bundles of rays, the indirect, i.e. the bundles of rays diffracted and scattered at specimen details, are of major importance for imaging true to the object. The higher the portion of indirect bundles of rays (aperture), the more realistic is the microscopic image according to ABBE.

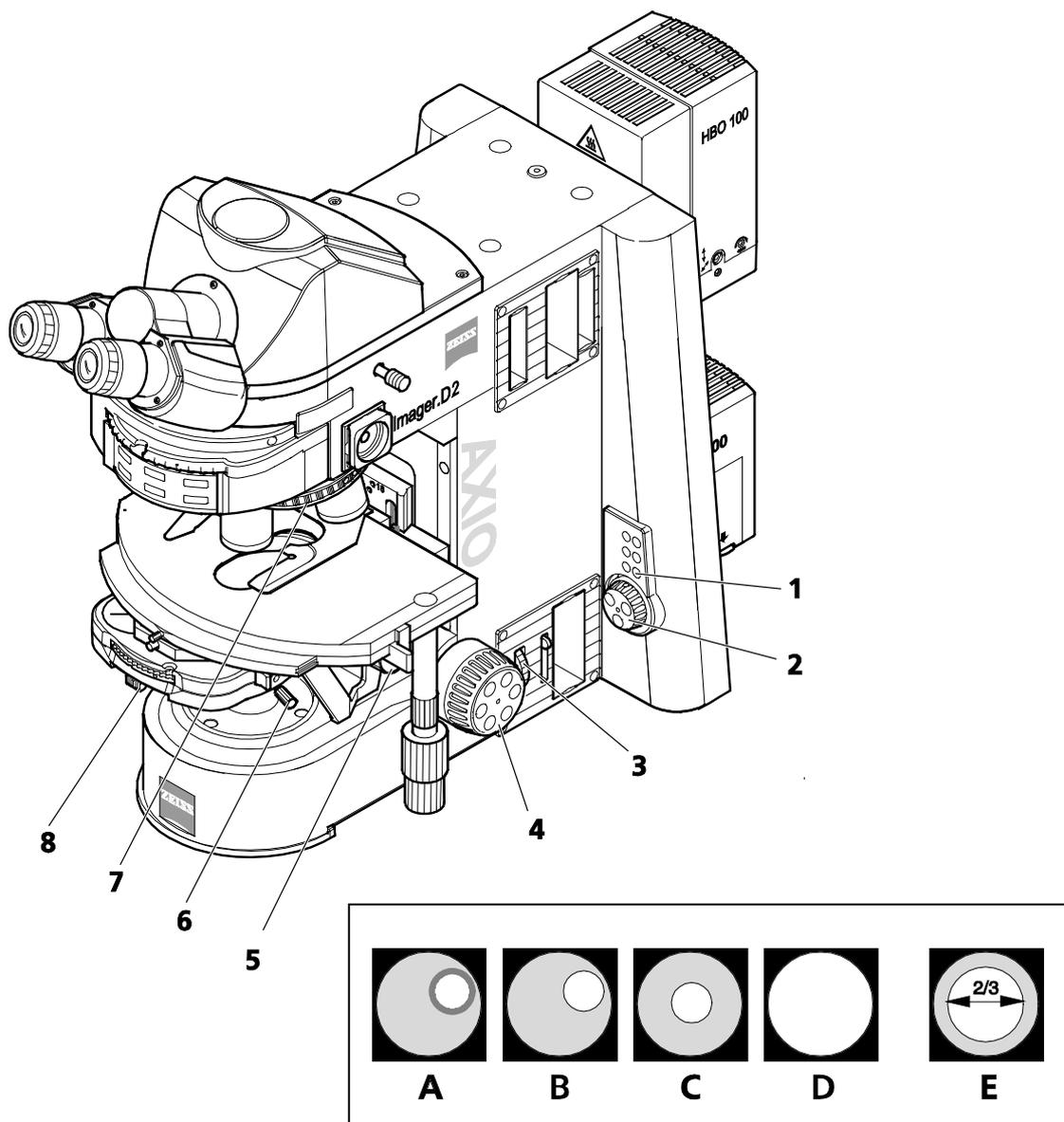
To fully exploit the optical performance of the microscope, particularly that of the objective, condenser, luminous-field diaphragm and aperture diaphragm should be set according to the rules of the KÖHLER illumination principle. These fundamental rules of microscope adjustment are described in detail below in Section 4.9.1 (3) "Transmitted-light brightfield according to KÖHLER" for the Axio Imager.

#### (2) Instrument configuration

- The equipment of all Axio Imager microscopes allows transmitted-light brightfield microscopy.
- For the use of the achromatic-aplanatic universal condenser 0.9 H/0.8-0.9 DF, refer to Section 4.9.2 (4).

#### (3) Setting transmitted-light brightfield according to KÖHLER

- The microscope has been started up correctly as described in Section 3.
- The microscope has been switched on.
- Set the toggle switch for the halogen illuminators on the rear side of the instrument to transmitted light.
- Turn voltage control (4-114/2) on microscope base to adjust image brightness. If the transmitted-light shutter is closed (indicator LED is not lighting), open it by means of button (4-114/1).
- Place a high-contrast specimen on the mechanical stage.
- Swivel in front lens of condenser (for objectives  $\geq 10x$ ) and use the vertical control of the condenser (4-114/5 or 4-115/3) to move it up to the upper stop. The stop must have been set in such a manner that the specimen is not touched by the condenser (for setting the stop of the vertical condenser drive, refer to Section 4.9.1 (4)).
- Swivel in 10x objective (yellow ring, also refer to Section 2.5) on nosepiece (4-114/7) and focus on the specimen using the focusing drive (4-114/4).
- Close luminous-field diaphragm (4-114/3) until it becomes visible (even if not in focus) in the field of view (4-114/A).
- Turn the vertical control of the condenser drive (4-114/5 or 4-115/3) to lower the condenser until the edge of the luminous-field diaphragm appears in focus (4-114/B).



**Fig. 4-114** Microscope settings on Axio Imager for transmitted-light brightfield

- Use both centering screws (4-114/6 or 4-115/1) on the condenser carrier to center the luminous-field diaphragm image (4-114/C). Then, open the diaphragm until its edge just disappears from the field of view (4-114/D).

 When changing the condenser, normally the luminous-field diaphragm remains centered, unless the centering screws were turned.

- To adjust the aperture diaphragm (contrast), remove one eyepiece from the tube and look through the tube with your naked eye. Adjust the aperture diaphragm (4-114/8) to approx.  $\frac{2}{3}$  ...  $\frac{4}{5}$  of the diameter of the exit pupils of the objectives (4-114/E). In most applications, this aperture diaphragm setting provides optimum contrast at almost ideal resolution, and is therefore the best compromise for the human eye.
- Insert the eyepiece back in the tube socket.



Specimen field size and objective aperture change after every objective change as possibly does the centering slightly. Therefore, repeat the adjustment of luminous-field diaphragm and aperture diaphragm to obtain optimum results.

For < 10x objectives, the front lens of the universal condenser (if used) must be folded out and the aperture diaphragm fully opened. In the case of such large fields, the luminous-field diaphragm can also be used for better contrasting by its opening being reduced by a certain range. Closing it too much should be avoided in order not to worsen the evenness of the field of view illumination.

#### (4) Setting the height stop on the condenser carrier

- Loosen the fastening screw of the height stop (4-115/2) using an SW 3 ball-headed screwdriver.
- Use focusing drive to focus on the specimen.
- Close luminous-field diaphragm and focus it by turning the vertical control (4-115/3) of the condenser.
- Carefully raise the condenser slightly without lifting the specimen.
- Tighten fastening screw (4-115/2) of height stop.

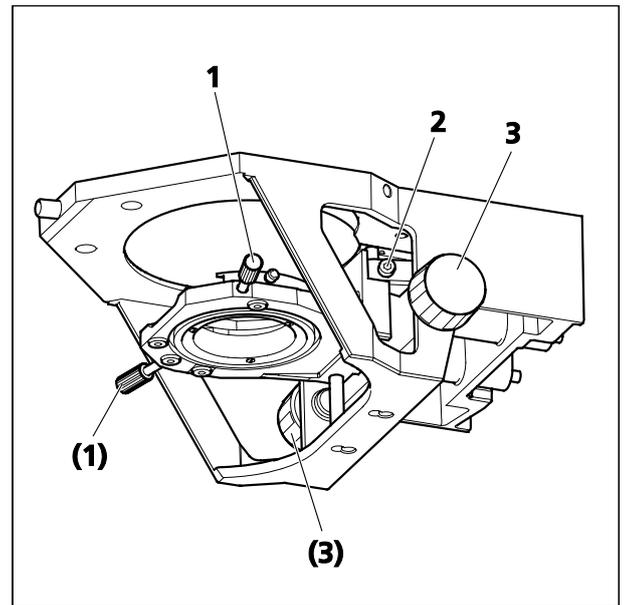


Fig. 4-115 Setting the height stop on the condenser carrier

## 4.9.2 Setting transmitted-light darkfield

### (1) General principle

Due to their transparency, unstained biological specimens, such as bacteria or living cell cultures, are often barely or not at all visible in transmitted-light bright field. This changes markedly if such specimens are viewed in transmitted-light dark field, where the specimen is illuminated using an aperture that is larger than that of the objective used.

In dark field, only the diffracted and scattered light components, which are important for imaging, reach the objective, while the direct unchanged light bundles are directed past the objective. This is one of the reasons why even fine structures can be resolved, which are sometimes below the resolving power of the light microscope and which appear very bright on a dark background.

### (2) Instrument configuration

- Universal condenser with dark-field stop in position D
- Achromatic-aplanatic universal condenser 0.9 H/0.8-0.9 DF (424216-0000-000), for low magnifications with wide-field-DF-slider for 2.5x-5x (424215-0000-000)
- Use of ICS objectives up to the maximum aperture of 0.75. Objectives with a higher aperture can be used in conjunction with the above universal condenser only in the equipment version with an integrated aperture iris stop and/or the screw-on darkfield attachment 1.2–1.4 Oil (424218-0000-000).

### (3) Setting transmitted-light darkfield

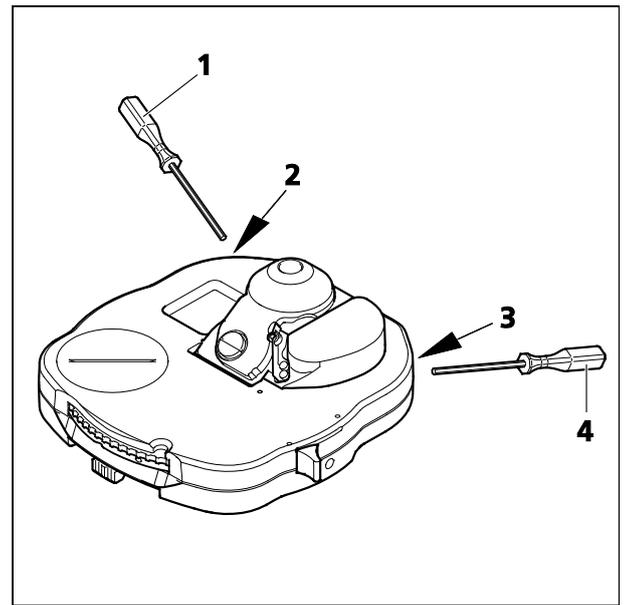
- Set KÖHLER illumination by analogy with transmitted-light brightfield. However, use the objective with the highest aperture instead of the 10x objective.
- Turn the turret disk of the universal condenser to position D and swivel in the condenser front lens.
- Remove the eyepiece from the tube (or replace it with the auxiliary microscope) and check the centering of the darkfield stop in the exit pupil of the objective. If the central darkfield stop D in the universal condenser is outside of or decentered to the exit pupil of the objective, and if the exit pupil is not homogeneously dark, the darkfield stop must be recentered.
- To center the darkfield stop, use the two SW 1.5 Allen screwdrivers (4-116/1 and 4) to turn the two centering screws (4-116/2 and 3) until the exit pupil of the objective appears homogeneously dark. After centering, remove both SW 1.5 screwdrivers from the condenser.

Leaving the two Allen screwdrivers unintentionally in the motorized universal condenser does not pose a risk to the drives of the condenser as these instantly switch off when driving against a mechanical resistance. After having removed the source of the resistance, you can continue using the condenser as usual.

 Since the apertures of objectives with an integrated aperture iris stop are too high for transmitted-light dark field, the aperture iris stop must be closed to the limit aperture of 0.65.

The darkfield method has been perfectly set, when the background of the field of view appears as dark as possible.

- Reinsert the eyepiece in the tube.
- If the height of the dark-field condenser is set correctly and sensitively, it is possible to reduce any brightening in the field of view left, and the luminous-field diaphragm image appears almost perfectly in focus.
- Finally, match the size of the luminous-field diaphragm to the size of the field of view.



**Fig. 4-116 Centering the darkfield stop on the universal condenser**



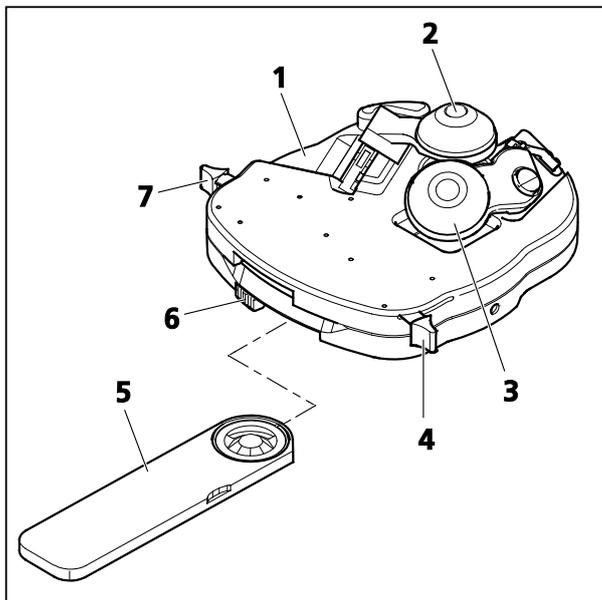
Dark field microscopy requires specimens to be considerably cleaner than in other techniques. Fingerprints, dirt or dust particles, in particular, have negative effects, since they brighten the background of the field of view and decrease the contrast of the object image.

**(4) Use of the achromatic-aplanatic universal condenser 0.9 H/0.8-0.9 DF**

The achromatic-aplanatic universal condenser 0.9 H/0.8-0.9 DF (4-117/1) is equipped with a front lens each for brightfield (4-117/3) and for darkfield (4-117/2), as well as with an aperture diaphragm (4-117/6). For brightfield applications, refer to Section 4.9.1.

The requirements for the use of the universal condenser dependent on the objective used are given in the table below.

Magnification	Brightfield applications	Darkfield applications
2.5x ... 5x	Without brightfield front lens H	Without darkfield front lens D, with wide-field DF slider for 2.5x-5x
10x ... 40x	With brightfield front lens H	With darkfield front lens D
40 x ... 100x	With brightfield front lens H	With darkfield front lens D, with darkfield attachment 1.2-1.4 Oil, Objective with adjustable iris stop



**Fig. 4-117 Achromatic-aplanatic universal condenser 0.9 H/0.8-0.9 DF**

- To swivel the brightfield front lens **H** into the optical path, set lever (4-117/4) in position **I** (darkfield front lens **D** swiveled out). To swivel it out, set lever in position **0**.
- Before swiveling the darkfield front lens **D** in or out, it is necessary to pull out the wide-field DF slider (4-117/5) from the universal condenser (outer click-stop position). To swivel this lens in, set lever (4-117/7) in position **0**. To swivel it out, set the lever in position **I**.
- For immersion applications, screw the darkfield attachment 1.2-1.4 Oil onto the darkfield front lens and apply Immersol 518 F® immersion oil.

**(5) Use of achromatic-aplanatic condenser 0.9 H/0.4 LD**

Achromatic-aplanatic condenser 0.9 H/0.4 LD is equipped with a 0.9 front lens for brightfield use and a second front lens (0.4) for brightfield and phase contrast applications, using slider Ph 1 or Ph 2 at a working distance of approximately 14 mm

- The front lenses are swiveled in and out in the same manner as described for achromatic-aplanatic universal condenser 0.9 H/0.8-0.9 DF.
- Slider Ph must be removed from the condenser when front lens 0.4 is swiveled in or out.

### 4.9.3 Setting transmitted-light phase contrast

#### (1) General principle

The phase contrast technique is ideal for examining thin, unstained specimens, e.g. culture cells. The human eye is unable to see phase differences (differences in refractive index and thickness) between the different cell components.

The phase contrast technique uses the optical modulators "phase stop and phase ring" as well as the interference procedures during formation of the intermediate image in order to change the small phase differences into intensity and color differences that are visible to the eye.

The high-intensity, direct light components are attenuated with the optically defined ring channel "phase stop and phase ring" and given a constant phase shift. The indirect light components diffracted at different cell components, however, bypass this optical channel and are influenced in phase by the refractive index and the thickness differences in the specimen.

In the intermediate image plane, the thus differently influenced partial beams interfere with each other and are amplified or attenuated - depending on the phase position. This interference results in image contents displaying differences in intensity and color that can be perceived by the human eye.

#### (2) Instrument configuration

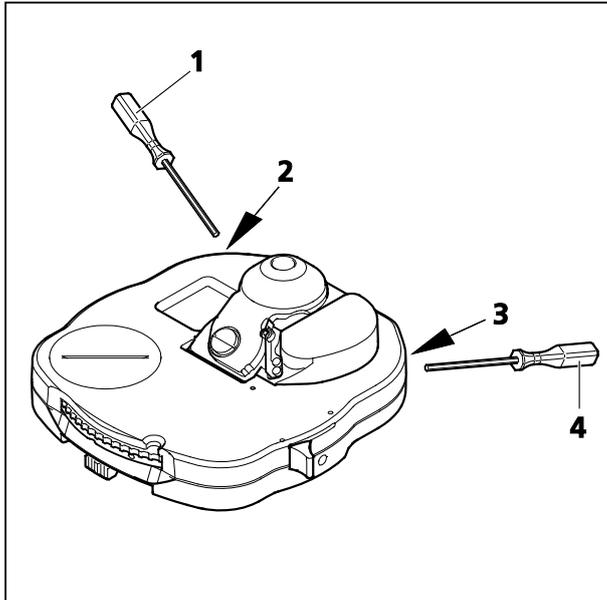
- Phase-contrast objectives with phase rings Ph 1, Ph 2 or Ph 3 for different average numerical apertures that can also be used in brightfield without any restriction.
- Universal condenser with turret disk containing centerable phase stops Ph 1, Ph 2 and Ph 3 for different average numerical apertures.
- The phase stop on the universal condenser swiveled into the light path must agree with the corresponding label on the objective, e.g. Ph 1.

#### (3) Setting transmitted-light phase contrast

- Swivel phase-contrast objective, e.g. the one labeled with Ph 1, into the light path.
- On the turret disk of the universal condenser, swivel in the phase stop labeled like the phase-contrast objective, e.g. Ph 1.
- To check centering and congruence of the bright phase stop (in the condenser) with the dark phase ring (in the objective), remove one eyepiece from the tube and replace it with the auxiliary microscope. Use the correction device on the auxiliary microscope to focus on the phase stop and the phase ring in the exit pupil of the objective.



To check the centration, you can also use the Bertrand lens slider PH. However, this is possible only if there is no camera path deflection installed on the left side of the microscope stand.



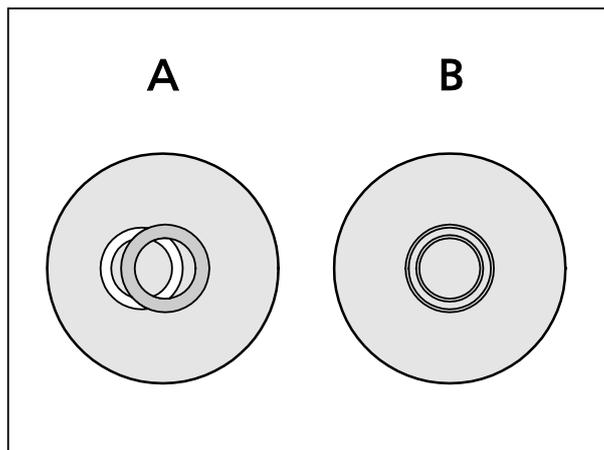
**Fig. 4-118** Centering the phase stop on the universal condenser

- If congruence is not perfect (4-119/**A**), use the two SW 1.5 screwdrivers (4-118/**1** and **4**) for turning the two centering screws (4-118/**2** and **3**) to recenter the bright phase stop until complete congruence with the dark phase ring is achieved (4-119/**B**).
- Finally, remove the auxiliary microscope from the tube and reinsert the eyepiece.

Normally, however, centering is not required, since the phase stops are factory-centered and centering is retained even if the universal condenser is removed from the condenser carrier and attached again.

To enhance the image contrast, an interference wide-band filter, green 32 x 4, can be inserted in the color glass holder (requires color glass holder for 32 mm filter size).

Perfect phase contrast is only achieved if the bright phase stop (in the condenser) and the dark phase ring (in the objective) are exactly congruent in the illumination beam path (4-119/**B**).



**Fig. 4-119** Centering the phase stop (bright ring in condenser) to the phase ring (dark ring in objective)

#### 4.9.4 Setting transmitted-light differential interference contrast (DIC)

##### (1) General principle

The transmitted-light DIC method is a contrast method alternative to polarization allowing contrasty, 3D presentation of transparent specimen details.

The light linearly polarized by a polarizer is split into two partial beams by a birefringent prism. The beams pass two closely adjacent places of the specimen and thus are subject to different path differences caused by differences in refractive index and thickness of the specimen. Afterwards, both partial beams are recombined in a second birefringent prism and, after having passed the analyzer, they have the same vibration direction. Thus, both partial beams can interfere with each other in the intermediate image with the different path differences being converted to different gray values (intensities). A compensator  $\lambda$  (full-wave plate) then converts the gray values to colors.

##### (2) Instrument configuration

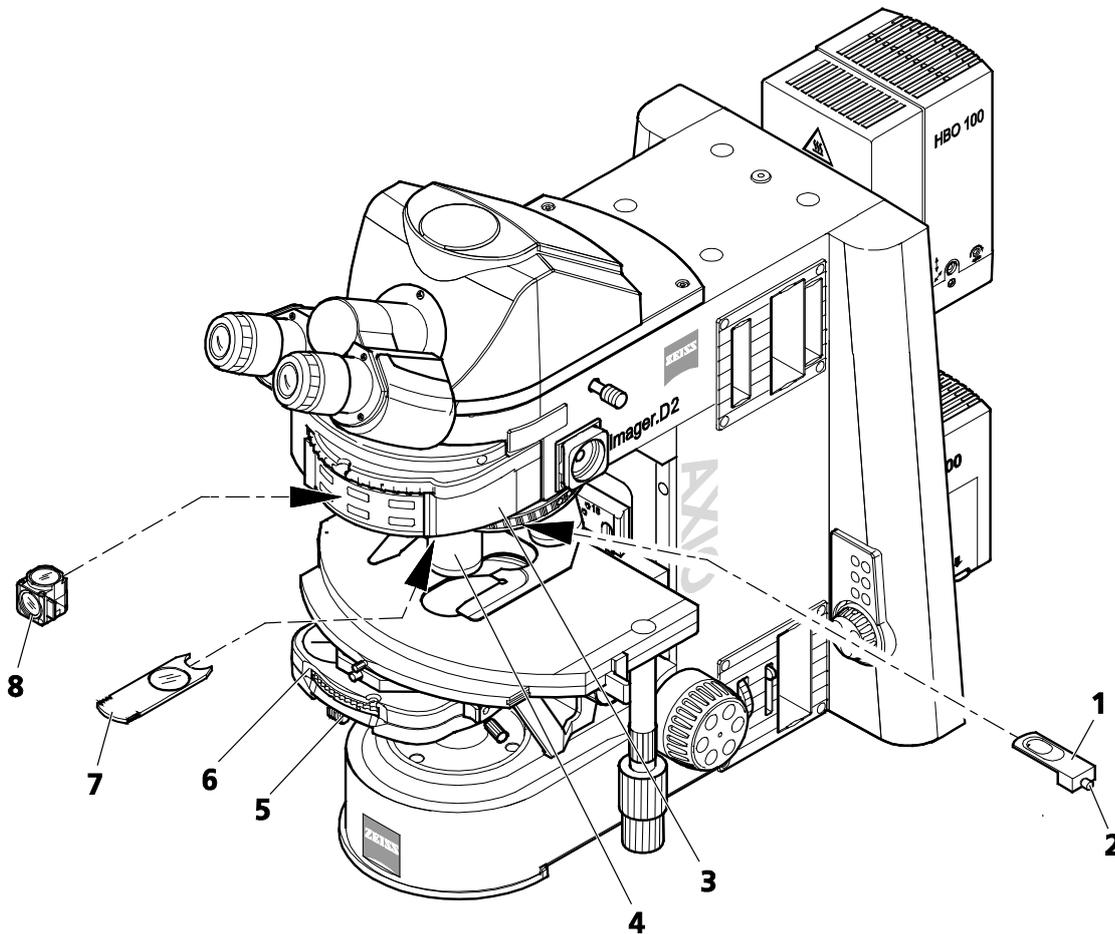
- Objectives offered together with the DIC equipment, e.g. Plan-Neofluar objectives
- Objective nosepiece with slot for DIC slider, matching the objectives used  
or  
motorized four-position modulator turrets in combination with analyzer module DIC ACR shiftfree for transmitted light
- Condenser with turret disk containing DIC prisms (DIC I, DIC II, DIC III)
- Analyzer module D DIC P&C (424921-0000-000) in reflector turret. Only this module may be used on the Axio Imager as it provides a homogeneous field of view in combination with the special condenser modules I, II and III.
- Preferably a rotary mechanical stage

##### (3) Adjusting the transmitted-light DIC with DIC slider and DIC prism in the condenser turret

- On the nosepiece, swivel the objective (4-120/4) suitable for DIC into the light path. Push the corresponding DIC slider (4-120/1) into the slot of the respective objective position. Make sure the DIC slider snaps in reliably.
- On the reflector turret (4-120/3), swivel in the analyzer module D DIC (4-120/8).
- Swivel in the suitable DIC prism I, II or III (digit label on turret disk of condenser (4-120/6)).
- Adjust luminous-field diaphragm and aperture diaphragm (4-120/5) in accordance with the rules of KÖHLER illumination.
- Turn knurled screw (4-120/2) on DIC slider (4-120/1) to adjust optimum contrast. When adjusting the DIC slider symmetrically around its mid-position, the specimen details can be imaged in such a way that they appear three-dimensionally as if they were raised or dented.
- If desired, you can push the compensator  $\lambda$  (4-120/7) into the slot for the analyzer slider above the nosepiece to obtain colored DIC contrast.

For 63x objectives, in addition to the DIC sliders HR (High Resolution), another type, HC (High Contrast), is available, which is to be used depending on the type of specimen and examination to be performed.

 The DIC method employs polarized light and therefore is disturbed by birefringent objects, such as films, in the light path between polarizer and analyzer, as are occasionally used in conjunction with histological sections. The same applies to acrylic glass culture chambers, if the chamber bottom is made of plastic. In these cases, it is advisable to use chambers with bottom plates of glass to avoid loss of optical performance.

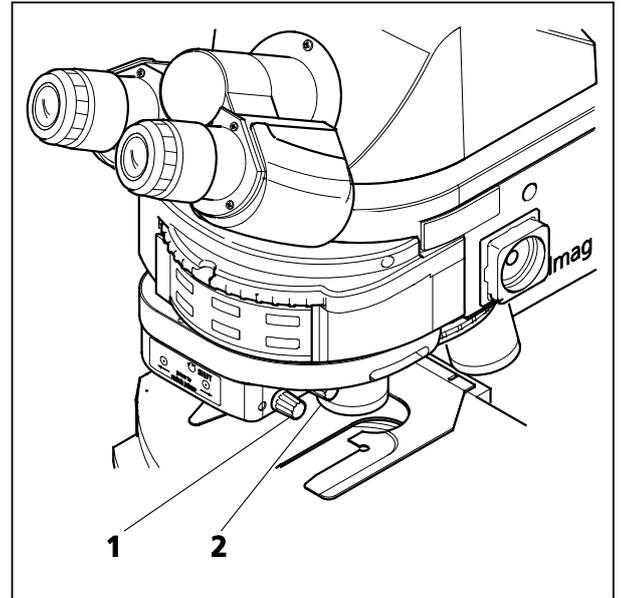


- 1 DIC slider
- 2 Knurled screw
- 3 Reflector turret
- 4 Objective on nosepiece
- 5 Sliding button for aperture diaphragm
- 6 Condenser with DIC prism and polarizer
- 7 Compensator  $\lambda$
- 8 Analyzer module

**Fig. 4-120** Components required for transmitted-light DIC

**(4) Adjusting the transmitted-light DIC with motorized four-position modulator turret for transmitted light**

- On the nosepiece, swivel in the objective suitable for DIC.
- On the reflector turret, swivel in analyzer module DIC ACR shiftfree for transmitted light.
- On the modulator turret, swivel in the appropriate DIC prism by **pressing** on knob (4-121/1) for forward rotation or on knob (4-121/2) for backward rotation of the turret.
- Adjust the luminous-field diaphragm and aperture diaphragm according to KÖHLER.
- Set optimum contrast by turning knob (4-121/2).
- If desired, compensator  $\lambda$  can be inserted into the slot for the analyzer slider located above the nosepiece, so as to create a differential interference contrast (DIC) in colours.

**Fig. 4-121 Motorized four-position modulator turret for transmitted-light DIC**

## 4.9.5 Setting transmitted-light polarization for orthoscopic observation

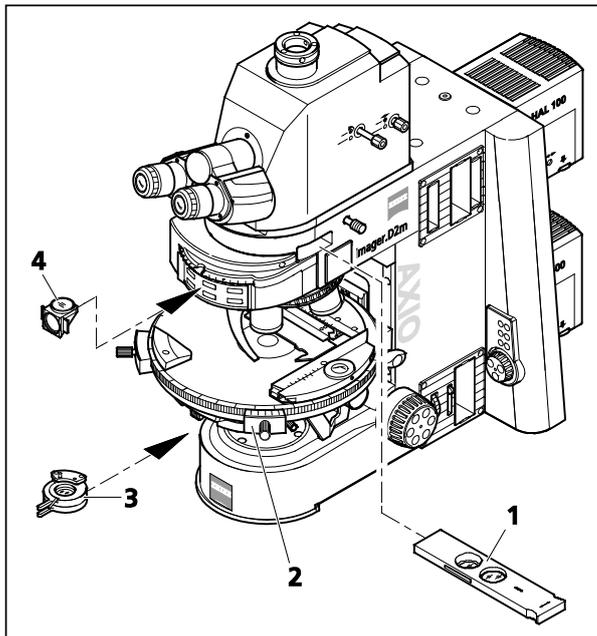
Magnified viewing, e.g. of a thin section, in polarized light is called orthoscopy (Greek: orthos = straight; skopein = seeing), because illumination is by "straight" light rays, which – with the aperture diaphragm largely closed – are parallel to the microscope axis.

### 4.9.5.1 Detecting birefringence

#### (1) Use

The technique of transmitted-light polarization is used for specimens that change the state of polarization of light. These specimens, such as crystals, minerals or polymers, are called birefringent. When these birefringent substances are viewed between crossed polarizers (polarizer  $\perp$  Analyzer), they appear brightened, while their surroundings remains dark.

Birefringent substances are identified by the fact that they show four bright and four dark positions while being rotated through  $360^\circ$  between crossed polarizers. Dependent on birefringence, thickness as well as orientation of the specimen, interference colors ranging from gray (mostly with biological specimens) to white, yellow, red and blue appear in this process. These interference colors can be of the first or any higher order.



**Fig. 4-122** Components for transmitted-light polarization

#### (2) Microscope configuration

- Phototube Pol
- Achromatic-aplanatic universal condenser Pol
- Strain-free objectives
- Rotary stage Pol (4-122/2)
- Polarizer D (rotatable or fixed) (4-122/3) oder polarizer integrated in the turret disk of the condenser
- Analyzer slider (4-122/1) or analyzer module D (4-122/4) in reflector turret (only on microscopes equipped with a reflector turret)

#### (3) Setting the microscope

- Set the microscope as for transmitted-light bright field according to KÖHLER (see Section 4.9.1 (3)).
- Center rotary stage Pol (4-122/2) and objectives (if not done yet – see Section 3.34.3).
- Swivel polarizer (4-122/3) into the light path and, if it is rotatable, turn it to  $0^\circ$ .

- Push the analyzer slider (4-122/1) into the respective slot or swivel in the analyzer module (4-122/4) on the reflector turret. The field of view appears dark due to the polarizers being crossed.



If you work with the analyzer slider on the Axio Imager, swivel the reflector turret to a blank turret position.

- Move the specimen feature you want to examine into the field of view and rotate it with the rotary stage Pol through 360°. As a rule, while being rotated between crossed polarizers, birefringent (anisotropic) specimens will now show the variations in color and intensity described above. However, optically anisotropic substances may also remain dark, if an isotropic direction, e.g. of optically uniaxial or biaxial crystals, is oriented parallel to the direction of observation.

#### 4.9.5.2 Determining the vibration direction $n_{\gamma}$

##### (1) Use

The determination of the vibration directions of  $n_{\gamma}$  and  $n_{\gamma'}$  (vibration direction with the absolutely or relatively highest refractive index) and  $n_{\alpha}$  and  $n_{\alpha'}$  (vibration direction with the absolutely or relatively lowest refractive index) in relation to the morphological directions, e.g. of crystal surfaces, crystal needles or fibers, provides an important criterion for recognition. It is also used for the diagnosis of biocrystals (e.g. gout, pseudo-gout).

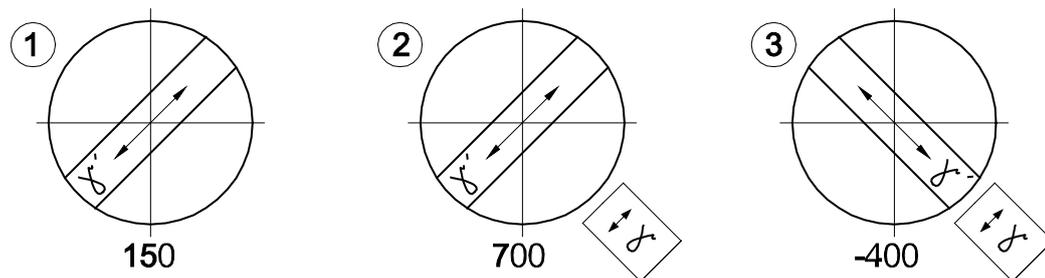


Fig. 4-123 Determining the vibration direction  $n_{\gamma}$  by the example of a synthetic fiber

##### (2) Microscope configuration

- Phototube Pol
- Achromatic-aplanatic universal condenser Pol
- Strain-free objectives
- Rotary stage Pol
- Polarizer D (rotatable or fixed)
- Analyzer slider or analyzer module D in reflector turret (only on microscopes equipped with a reflector turret)

### (3) Setting the microscope

The microscope has been prepared as described in Section 4.9.5.1 (3).

- Rotate the rotary stage Pol with the specimen, e.g. a synthetic fiber, until the specimen appears as dark as possible. In this position, the fiber extends parallel to one of the two directions of the crosslines reticle.
- Next, turn on the rotary stage Pol by 45°, until the longitudinal axis of the fiber is oriented NORTH-EAST (NE) – SOUTH-WEST (SW) (Fig. 4-123). In this position, the specimen shows the maximum brightness (diagonal position) and may appear in any color.
- Push in the full-wave compensator  $\lambda$ .

Like the specimen, the compensator  $\lambda$  is a birefringent object, though one with a defined path difference of 550 nm and the principal vibration direction  $n_\gamma$  definitely oriented in NE-SW direction

By moving compensator  $\lambda$  into the light path, the specimen changes its color. The kind of color change depends on the orientation of the specimen (NE-SW or NW-SE).

The changes in color are based on optical interference. The interference colors (path differences) in both diagonal positions (NE-SW and NW-SE) of the specimen must be compared in this connection.

The path difference results from the superposition (interference) of the vibration direction of the specimen with the vibration direction of the compensator  $\lambda$ .

The greater path difference occurs, if the vibration direction of the specimen with the absolutely or relatively highest refractive index ( $n_\gamma$  or  $n_{\gamma'}$ ) is parallel to the principal vibration direction of compensator  $\lambda$ . The specimen will then appear, for instance, greenish-blue (4-123/2).

The smallest path difference occurs, if the vibration direction of the specimen with the absolutely or relatively lowest refractive index ( $n_\alpha$  or  $n_{\alpha'}$ ) is perpendicular to the vibration direction of the compensator  $\lambda$ . The specimen will then appear, for instance, in yellow (4-123/3).

#### (4) Conclusions

The grayish-white color appearing first in the bright position in the above example (4-123/1) corresponds to a path difference of 150 nm according to the Michel-Lévy color chart (Fig. 4-124).

When the compensator  $\lambda$  is brought into the light path, the non-birefringent "surroundings" of the synthetic fiber appear in a dark red color, which corresponds to the path difference of the compensator of 550 nm (1<sup>st</sup> order interference color for the path difference of 550 nm corresponds to  $1 \lambda$ ).

If the vibration direction ( $n_\gamma$  or  $n_{\gamma'}$ ) of the birefringent specimen to be examined is parallel to the principal vibration direction ( $n_\gamma$ ) of the compensator  $\lambda$ , i.e. in NE-SW direction, the path difference of the specimen (e.g. grayish-white: 150 nm) and the path difference of the compensator  $\lambda$  (red: 550 nm) add up. This results in a color change of the specimen from grayish white to greenish-blue (resulting path difference = 700 nm).

If the vibration direction of the specimen to be examined is perpendicular to the principal vibration direction of the compensator  $\lambda$ , i.e. in NW-SE direction, the path difference of the specimen (e.g. grayish-white: 150 nm) is subtracted from the path difference of the compensator  $\lambda$  (red: 550 nm). In this case, the interference color of the specimen visibly changes from grayish-white to orange (resulting path difference = 400 nm).



Color charts according to Michel-Lévy are available under Cat. No. 42-312.

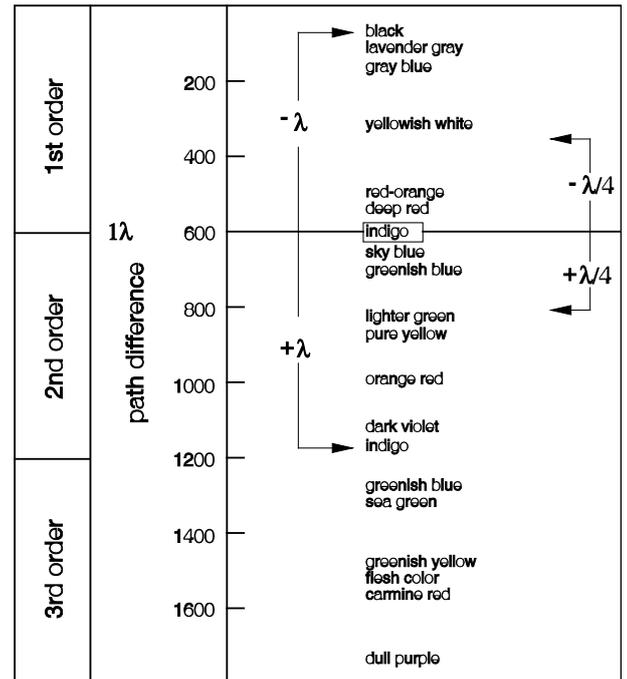


Fig. 4-124 Schematic diagram of the color charts according to Michel-Lévy

### 4.9.5.3 Measuring path differences

For the accurate measurement of path differences, measuring compensators are required. These compensators reset or compensate the path difference produced by the specimen to zero (1<sup>st</sup> order black).

While for the methods described above, the addition position or additionally also the subtraction position is of interest, for measurements **exclusively** the subtraction position is relevant.

Path differences in the specimen can assume very small values ( $1/50 \lambda$  or 10 nm) and very great values (more than  $10 \lambda$  or approx. 5500 nm and more) and with that determine the compensator appropriate for the measurement.

To find the appropriate compensator follow this procedure:

- Set the microscope as for transmitted-light polarization (refer to Section 4.9.5.1 (3)).
- Position the specimen feature to be examined directly under the center of the reticle.
- Restrict the size of the aperture to a value of about 0.2.
- Rotate the rotary stage Pol until the specimen is in extinction position, i.e. it appears **entirely dark**. In this position, activate the 45° click stop.
- Rotate the stage **once** (by 45°), so that the specimen is in diagonal position (lightened up).

The intensity of the interference or color produced by the specimen allows the following conclusion to be drawn:

- If more or less strong interference colors appear on the object, the path difference ranges approximately between  $1/2 \lambda$  and about  $5 \lambda$ .

The suitable compensator is:

**Tilting compensator B 0-5  $\lambda$**  (Accessory 000000-1115-700).

- If the object-side color changes from light gray/white to a strong interference color, when a compensator  $\lambda$  (473704-0000-000) is inserted in the compensator slot, the path difference is ( $1/4 \dots 1/2$ )  $\lambda$ .



The occurrence of the color change effect possibly requires the assessment in two specimen positions lying 90° apart. For this purpose, rotate the centered stage (by 2 click stops).

The suitable compensator is:

**Tilting compensator B 0-5  $\lambda$**  (Accessory 000000-1115-700) or the compensation method according to DE SENARMONT up to  $1 \lambda$  using the **Senarmont compensator 546/4 nm** (Accessory 000000-1115-695).



The compensation method according to DE SENARMONT requires the use of the rotatable analyzer (000000-1076-680).

- After insertion of the compensator  $\lambda$  and rotation of the specimen by  $90^\circ$  (2 click stops), the interference color remains to be white, in this case, however, it is a "higher-order white" and thus the path difference is  $> 5 \lambda$ .  
The compensator appropriate for this application:  
**Tilting compensator K 0-30  $\lambda$**  (Accessory 000000-1115-698)
- A dark gray as appearing interference color suggests very small path differences ( $\lambda/10$  or 54.6 nm).  
The compensator appropriate for this application:  
**Rotary compensator Brace-Köhler  $\lambda/10$**  (Accessory 000000-1115-703).
  
- Push the compensator into the slot as far as it will go.

For measurement preparation and measurement procedure, use the enclosed operating instructions.

#### 4.9.5.4 Transmitted-light circular polarization contrast

##### (1) Use

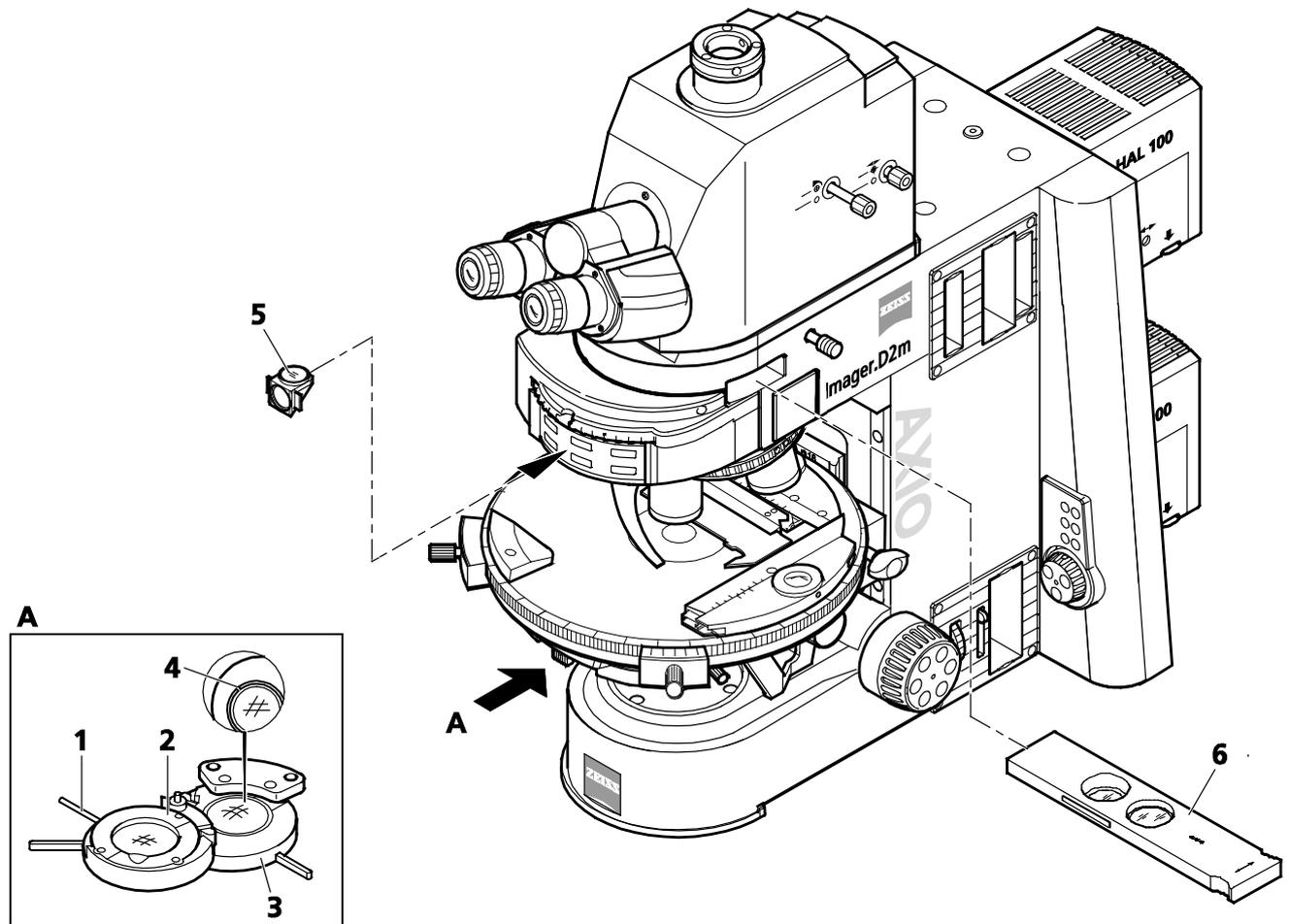
Unlike standard polarization contrast, circular polarization contrast does not show any dark (extinction) positions that depend on the angle of rotation (azimuth) of the specimen relative to polarizer or analyzer. This means, that the image will always look the same while you rotate the stage, as there are no bright and dark positions. With optical anisotropy, all transparent specimens show the interference colors that are characteristic to them.

##### (2) Microscope configuration

- Phototube Pol
- Achromatic-aplanatic universal condenser Pol
- Strain-free objectives
- Rotary stage Pol
- Polarizer D (rotatable or fixed)
- Circular polarization equipment D, ACR, with  $\lambda/4$  plate, rotatable for transmitted light, including reflector module  $\lambda/4$ , ACR, P&C (427703-9901-000)
- Analyzer slider

##### (3) Setting the microscope

- Set the microscope as for transmitted-light bright field according to KÖHLER (see Section 4.9.1 (3)).
- Center rotary stage Pol and objectives (if not done yet - see Sections 3.34.3, 3.34.4).
- For the further settings, for the time being **do not** use a specimen.
- Push the analyzer slider (4-125/6) into the light path.
- Install the circularpolarizer D (see Section 3.35), and insert reflector module  $\lambda/4$ , ACR, P&C (4-125/5) in the reflector turret.
- Swivel the bottom part of the circular polarizer D (4-125/3) into the light path up to the click stop and, at full light intensity, assess the extinction (darkening) of the field of view without specimen. If the extinction is not optimal, correct the position of the vibration direction by slightly and sensibly turning on the polarizer mount (for this, use the adjusting slots (4-125/4) on the bottom of the polarizer), until the extinction is maximum. Normally, however, this will not be necessary, as the position of the vibration direction of the polarizer is factory-adjusted.
- Then, swivel the top part of the circular polarizer D (4-125/2) into the light path.
- Rotate the lever of the  $\lambda/4$  plate of the circular polarizer D (4-125/1) until the extinction is maximum (dark-gray field of view) (lever points 45° to the right).



**Fig. 4-125 Components for circular polarization contrast**

- Do not view an (anisotropic) specimen unless the above adjustment has been done.
- Place the specimen to be examined onto the stage.

The specimens appear constantly and independently of the stage rotation in their specific interference color, which depends on the material, specimen thickness and orientation.



To obtain a contrasty image at high objective magnifications (from approx. 20x), reduce the illumination aperture to a value of between 0.15 and 0.20, i.e. close the aperture diaphragm accordingly.

The effect of the  $\lambda/4$  plate (4-125/2) can be undone by either swiveling it out of the light path or turning it with lever (4-125/1) into one of its two click-stop positions.

#### **4.9.6 Setting transmitted-light polarization for conoscopic observation – determining the optical character of crystals**

For the classification (and thus identification) of crystalline matter, the examination of the interference image in the objective pupil delivers more valuable information than that obtained by viewing the specimen itself. The interference image becomes visible in the eyepiece, if an additional optical system (fixed or focusing Bertrand lens or, on the basic version, the auxiliary microscope or diopter) is used.

In contrast to orthoscopy, this technique is called conoscopy, because here ideally the specimen is illuminated through a widely open cone. In practical microscopic work, this means that the condenser front lens (0.9 or 1.4) must be in the light path, the aperture diaphragm fully open, and the objective, too, should be a high-aperture type.

##### **(1) Use**

The determination of the optical character of transparent and weakly absorbing crystals serves to diagnose crystals. This method is also termed conoscopy. Its main application is the classical mineral microscopy. It allows, however, also synthetic crystals, industrial minerals and plastics (e.g. films) to be identified and characterized.

##### **(2) Microscope configuration**

- Stand with installed phototube Pol or with Bertrand lens slider or with the tube lens turret with integrated Bertrand lens optics
- Strain-free objectives
- Achromatic-aplanatic universal condenser 0.9 H
- Rotary stage Pol
- Polarizer D (rotatable or fixed)
- Analyzer slider or analyzer module D in reflector turret



The phototube Pol can be installed on all Axio Imager stand types.

### (3) Setting the microscope for conoscopy with the phototube Pol

In the case of uniaxial crystals, the most favorable orientation for conoscopic viewing is obtained with those specimen features (e.g. of a thin section) that in orthoscopic viewing change the brightness as little as possible. In this case, the direction of viewing and the optical axis are  $\pm$ parallel. The same refers also to biaxial crystals, if they are viewed along or approximately in the direction of one of the two optical axes.

- Set the microscope as for transmitted-light polarization (see Section 4.9.5.1 (3)).
- Put the specimen onto the stage and focus on it.
- Switch phototube Pol to visual observation, if necessary. To this end, pull out the push-pull rod on the left side (4-126/3).
- On phototube Pol, push in the front push-pull rod (4-126/2) on the right side to move the reticle into the light path.
- Move a selected crystal to the center of the reticle.
- Swivel in the 40x, 50x or 100x objective and, if necessary, refocus the specimen using the focusing drive.
- Check the centering of the objective by rotating the microscope stage. Recenter it, if necessary.
- Turn the front push-pull rod (4-126/2) to close the luminous-field stop so far that only the selected specimen feature remains visible. This is to prevent that the axial figure of the crystal to be examined is not overlapped by axial figures of adjacent crystals. This way, it is possible to view specimen features of up to 10  $\mu\text{m}$  diameter in conoscopic illumination.
- Move the Bertrand lens on the phototube Pol into the light path. To this end, push in the rear push-pull rod (4-126/1) on the right side. On doing so, the axial figure appears in the field of view. To focus the axial figure, turn this push-pull rod.

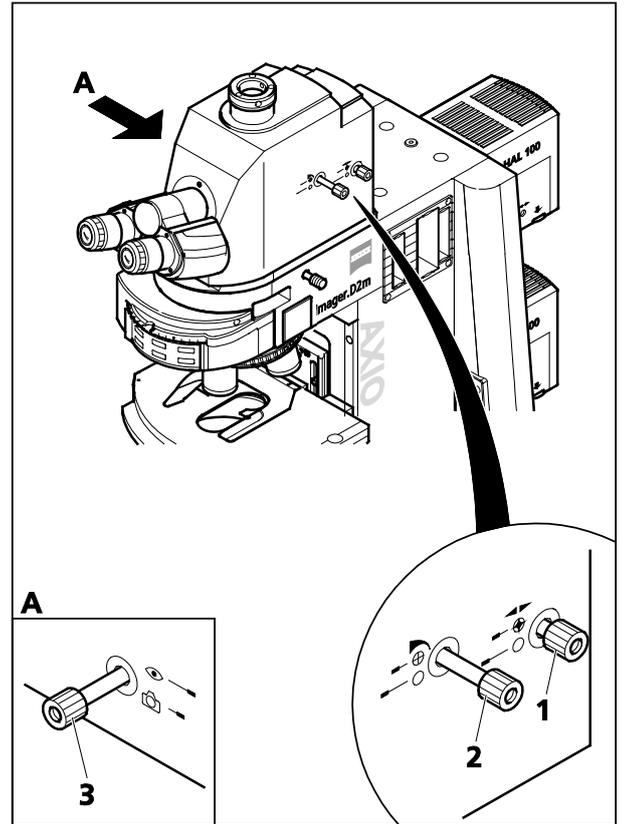


Fig. 4-126 Axio Imager mit montiertem Fototubus Pol

### (4) Setting the microscope with Bertrand lens slider or tube lens turret with Bertrand lens optics for conoscopy of large-size specimens

- Set the microscope as for transmitted-light polarization (see Section 4.9.5.1 (3)).
- Put the specimen onto the stage and focus on it.
- Swivel in the 40x, 50x or 100x objective and, if necessary, refocus the specimen using the focusing drive.
- Check the centering of the objective by rotating the microscope stage. Recenter it, if necessary.
- Close the luminous-field diaphragm so far that only the selected specimen feature remains visible.

- Push the Bertrand lens slider into the light path. Focus the axial figure by means of the lever of the slider,  
or,  
on the tube lens turret, swivel the position with the Bertrand lens into the light path, and focus the axial image by turning the focusing wheel.

**(5) Evaluation**

Crystalline anisotropic specimens can be classified in optically uniaxial and biaxial specimens, each with "optically positive" or "optically negative" character.

**Uniaxial** crystals show a **black cross**, if the optical axis is oriented parallel to the viewing direction. **Dependent on the degree of birefringence and the specimen thickness** concentrically arranged colored **interference fringes (the so-called isochromats)** might appear (see Fig. 4-127, second row).

This cross remains closed, when you rotate the stage. It can be located inside or outside the imaged objective pupil, depending on the position of the section.

With **optically biaxial** crystals, the cross resolves into two **dark hyperbola branches (the so-called isogyres)** depending on **stage rotation**, which are surrounded by colored interference patterns depending on the amount of birefringence and specimen thickness (suggestive of the figure "8").

Inserting a compensator  $\lambda$  (473704-0000-000) or  $\lambda/4$  (473714-0000-000) or a wedge compensator 0-4  $\lambda$  (000000-1140-663) in the compensator slot with the initial state of the axial figure being as illustrated in Fig. 4-127 results in the following changes in color shown schematically (blue and yellow areas) to the axial figure, thus allowing differentiation in "optically positive" and "optically negative".

	Optically uniaxial		Optically biaxial		
	Positive	Negative	Positive	Negative	
$\lambda$ plate (white → blue → yellow)					+ = blue - = yellow
Quartz wedge (Direction of motion at insertion)					↗ Direction of movement ↙
$\lambda/4$ plate (position of black spots)					

**Fig. 4-127 Determining the optical character**

If the positions of the section are more unfavorable, i.e. the center of the cross of optically uniaxial or the isogyre of optically biaxial specimens is outside the objective pupil, the optical character can be assessed as follows:

- The specimen is **optically uniaxial** if the isogyres are **straight** and run (related to the reticle) parallel through the pupil.
- The specimen is **optically biaxial** if the black isogyres are **curved lines** traveling across the pupil on a circular path.

Paying appropriate attention, also such axial figures not illustrated in Fig. 4-127 can be interpreted.

 Axial figures can often be better represented by circular polarization. Especially the determination of the axial angle of optically biaxial specimens (more or less the space between isogyres) succeeds with a much greater unambiguity. The optical character can also be determined by inserting the compensator  $\lambda$  (6 x 20) into the compensator slot.

#### 4.9.7 Setting reflected-light brightfield

 For all reflected-light contrasting techniques, the compensators 6x20 must be removed from the light path (slot for compensators).

##### (1) Application

The reflected-light brightfield microscopy is the simplest and most widely used microscopy technique which is used to examine opaque samples or specimens e.g. polished sections or wafers.

For a true-to-object imaging, indirect ray bundles, i.e. ray bundles diffracted and scattered on the specimen details, are of major importance in addition to the so-called direct ray bundles. The higher this portion of indirect rays (aperture), the more realistic the microscope image will be, according to ABBE's rule.

The incoming, bundled light from the reflected-light illuminator is reflected by a neutral-colored beam splitter. Then it passes to the objective which focuses the beams onto the specimen surface (so-called condenser function). The objective collects the light reflected by the object and generates the intermediate image of the microscope together with the tube lens, which is then observed visually or can be documented objectively.

##### (2) Instrument equipment

- Axio Imager MAT with connected and adjusted HAL 100 halogen illuminator.
- H P&C reflector module in reflector turret, 6x20 compensator mount with darkfield stop for reflected light (424706-0000-000) or 4-position modulator turret.

### (3) Setting reflected-light brightfield according to KÖHLER

- The microscope has been started properly as described in Section 3.
- The microscope has been switched on.
- Switch on the halogen lamp for reflected light using the reflected-light/transmitted-light toggle switch (4-1/36) on the microscope stand.

Depending on the existing equipment, the microscope contains a 6x20 compensator mount or a 4-position modulator turret for setting the contrasting techniques. The 6x20 compensator mount can be used for both brightfield and darkfield. For C-DIC and TIC examinations, additionally the corresponding 6x20 slider is required. Refer also to Section 4.9.9.

The 4-position modulator turret has a combined brightfield/darkfield position (**H/D**) as well as three additional positions for C-DIC (**C1**, **C2**) and TIC (**TIC**). Refer also to Section 4.9.10.

- When using the 6x20 compensator mount, remove the 6x20 slider, if necessary. When using the 4-position modulator turret, set the **H/D** position.
- Swivel the reflector turret into brightfield position **H**.
- Adjust light-intensity control (4-128/5) on microscope stand.
- Place a contrasty reflected-light specimen on the stage.
- Turn nosepiece (4-128/7) to swing in 10x objective (yellow ring, see also Section 2.5).
- Use focusing drive (4-128/6) to focus on the specimen. In doing so, always focus away from the specimen if possible to avoid any collision between objective and specimen.
- Remove the reflected-light diffusion disk. Turn adjusting screws of HAL 100 halogen illuminator to focus and center the image of the lamp filament in the exit pupil of the objective. For that, either pull out the adjusting aid or remove one eyepiece from the binocular tube. Afterwards, push the adjusting aid in again or reinsert the eyepiece. Move the reflected-light diffusion disk into the light path again.
- Set the aperture diaphragm (4-128/2) in mid-position (about half open or closed) by turning its knurled wheel.
- Reduce the size of the luminous-field diaphragm (4-128/4) by turning its knurled wheel until it becomes visible in the field of view (4-128/A).
- Turn the focusing drive (4-128/6) to refocus on the edge of the luminous-field diaphragm (4-128/B) and (using the SW 3 ball-headed screwdriver) turn the centering screws (4-128/3) until the luminous-field diaphragm is concentric with the edge of the field of view (4-128/C).
- Then, open the luminous-field diaphragm (4-128/4) so that it just disappears from the field of view (4-128/D).
- To set the aperture diaphragm (image contrast), remove one eyepiece from the binocular tube and look into the tube with your naked eye or insert the auxiliary microscope in place of the eyepiece.
- Center the aperture diaphragm with the centering screws (4-128/1) and, for specimens with average contrast, adjust the size of the aperture diaphragm to about 2/3 to 4/5 of the exit pupil diameter of the objective (4-128/E) by means of knurled wheel (4-128/2).

In most applications, this aperture diaphragm setting provides optimum contrast at almost ideal resolution, and is therefore the best compromise for the human eye.

- Finally, reinsert the eyepiece, refocus with the coaxial coarse and fine focusing drive (4-128/6) and adapt the image brightness to the specimen to be examined.

 Never use the aperture diaphragm to control the image brightness, but use the light intensity control (4-128/5), or swivel attenuation filters of the 2-position filter wheels into the light path!

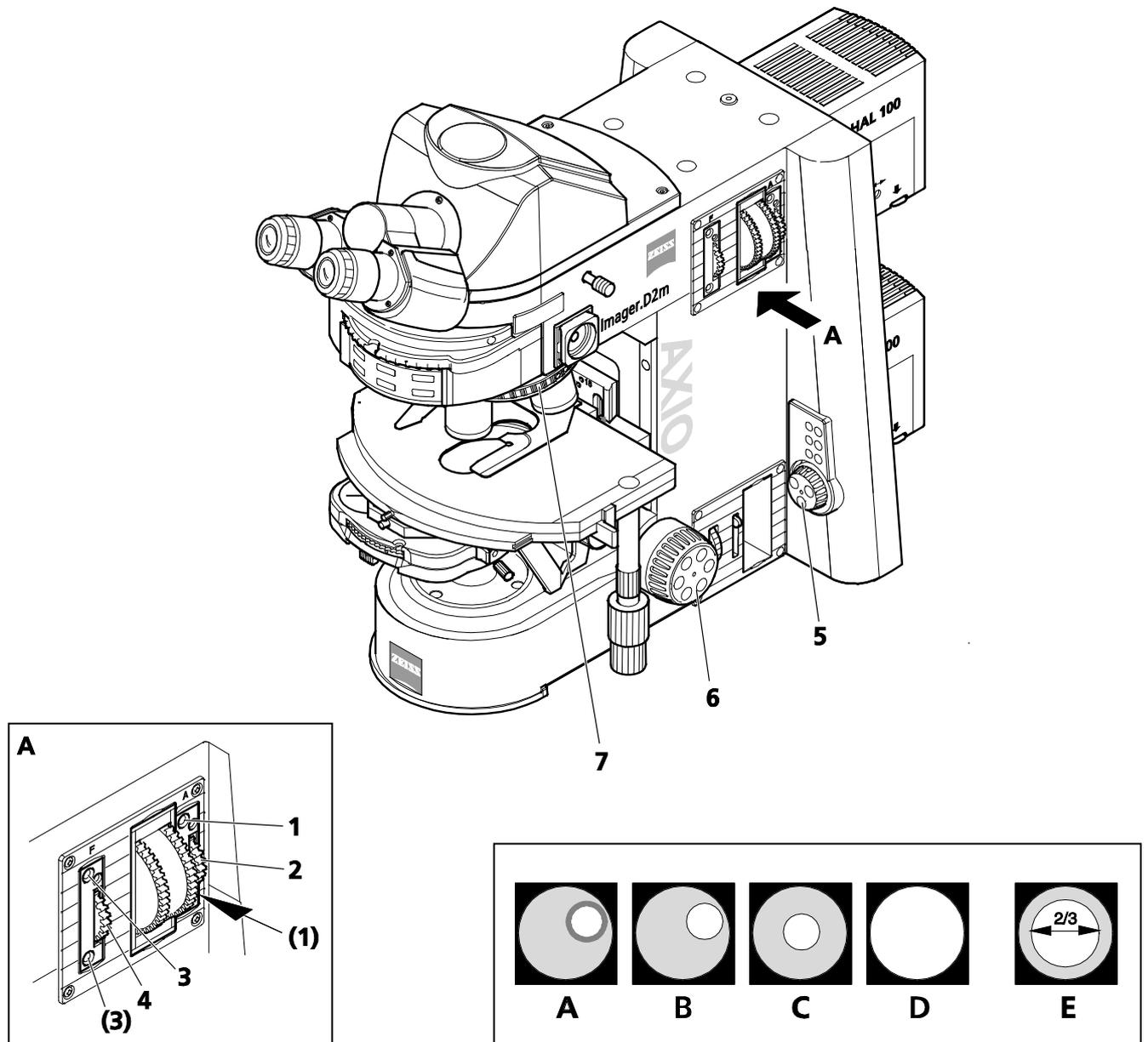


Fig. 4-128 Microscope settings on Axio Imager MAT in reflected-light brightfield

#### 4.9.8 Setting reflected-light darkfield

##### (1) Application

The incident light darkfield technique is used to examine specimens that do not only have reflective surfaces of different reflectivity (ideal brightfield objects), but feature scratches, cracks, pores, or in a nutshell, deviations in plane surfaces. All these light-scattering details shine brightly in the darkfield whilst the reflective plane surfaces stay dark.

##### (2) Instrument equipment

- Axio Imager MAT with connected and adjusted HAL 100 halogen illuminator.
- Epiplan-Neofluar, EC Epiplan-Neofluar, Epiplan objectives additionally labeled with "HD".
- Reflector module DF P&C, 6x20 compensator mount with darkfield stop for reflected light (424706-0000-000) or 4-position modulator turret

##### (3) Setting reflected-light darkfield

- Prepare the microscope as described in Section 4.9.7 for reflected-light brightfield. The luminous-field diaphragm should just disappear from the field of view to avoid reflections.
- Rotate the reflector turret to move the DF P&C reflector module into the light path.
- When using the 6x20 compensator mount, remove the 6x20 slider, if necessary. When using the 4-position modulator turret, set position **H/D**.
- Rotate the nosepiece to swing in the darkfield objective (HD).
- Fully open the aperture diaphragm and remove any neutral-density filter from the light path.
- Place the specimen on the stage and focus on it.

#### 4.9.9 Setting reflected-light DIC and reflected-light C-DIC

##### (1) Application

The reflected-light DIC and reflected-light C-DIC technique (DIC = Differential Interference Contrast, C-DIC = Differential Interference Contrast in Circularly polarized light) serves for producing high-contrast images of phase specimens, i.e. those specimens which only change the phase of the light in contrast to amplitude specimens.

##### (2) Instrument equipment

- Axio Imager MAT with connected and adjusted HAL 100 halogen illuminator.
- Rotary mechanical stage 75x50/240°
- EC Epiplan-Neofluar, Epiplan objectives additionally labeled with "DIC" or "Pol".
- DIC slider, that specially fits the objective that is engraved with magnification and aperture on the top of the slider or 6x20 C-DIC slider (in connection with the reflector module C DIC P&C), 6x20 compensator mount or 4-position modulator turret).

##### (3) Reflected-light DIC, preferably for Epiplan 5x - 100x and LD-Epiplan 10x - 100x objectives

- Prepare the microscope as described in Section 4.9.7 for reflected-light brightfield. The luminous-field diaphragm should just disappear from the field of view to avoid reflections.
- Rotate the reflector turret to move the C DIC P&C reflector module into the light path.
- Rotate the nosepiece to swing in the objective position with DIC slider slot.
- Push the DIC slider into the slot on the nosepiece (above the objective).
- Place the specimen on the stage, focus on it and rotate the mechanical stage until the specimen structure of interest appears at maximum contrast.
- To optimize the contrast, you can turn the knurled screw on the DIC slider.

##### (4) Reflected-light C-DIC

- Prepare the microscope for reflected-light brightfield.
- On the nosepiece, swivel in the objective suitable for DIC.
- Switch the C DIC P&C reflector module into the light path.
- Push the C-DIC slider 6x20 (4-129/3) into the 6x20 compensator mount (4-129/4) or rotate the 4-position modulator turret (4-130/6) at turret wheel (4-130/5) to swing in the desired C-DIC position (**C1** or **C2**).



C1 for 5x ... 20x objectives;  
C2 for 50x ... 100x objectives.

When using objectives of 50x or higher, it is advisable to use the reflected-light diffusion disk.

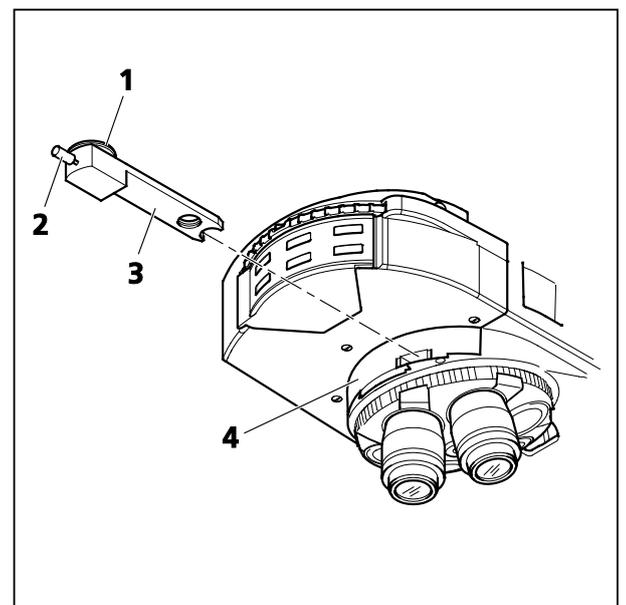


Fig. 4-129 6x20 compensator mount with 6x20 C-DIC slider

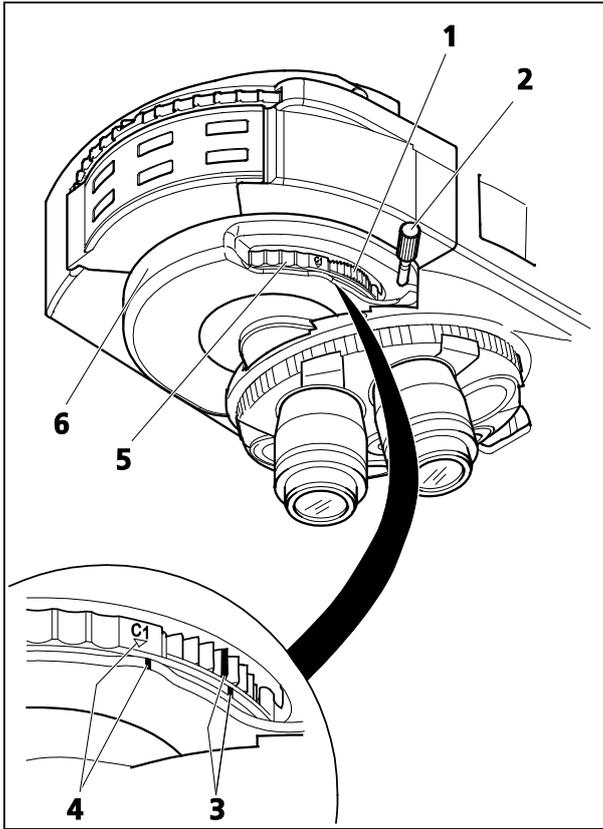


Fig. 4-130 4-position modulator turret

- Place the specimen on the stage, focus on it and rotate the control wheel (4-1291) on the C-DIC slider 6x20 or the modulator turret (4-130/1) until the specimen detail of interest is visible at maximum contrast. Rotating the stage is no longer necessary. If both line marks are facing each other (4-130/3), this corresponds to the mid-position. Rotation by 45° in both directions is possible).
- The contrast can be optimized by turning setscrew (4-129/2) on the C-DIC slider or on the modulator turret (4-130/2). If triangular and line mark are facing each other (4-130/3), this corresponds approximately to the extinction position (best contrast).

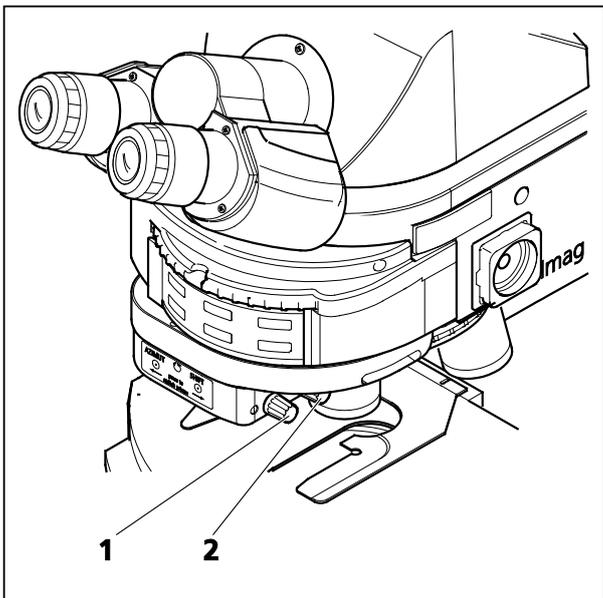


Fig. 4-131 Motorized four-position modulator turret for circular DIC/TIC

#### (5) Motorized four-position modulator turret for circular DIC/TIC

- Set the microscope up for reflected-light brightfield.
- Put reflector module C DIC P&C in the light path.
- On the nosepiece, swivel in the objective suitable for DIC.
- On the modulator turret, swivel in the appropriate DIC prism by **pressing** on knob
- (4-131/1) for forward rotation or on knob (4-131/2) for backward rotation of the turret.
- Put on the specimen, focus and turn knob (4-131/2) (**SHIFT**) in such a way that the structure of interest can be seen in maximum contrast.
- Contrast can be optimized by turning knob (4-131/1) (**AZIMUT**) on the modulator turret.

#### 4.9.10 Setting reflected-light TIC

##### (1) Application

The reflected-light TIC technique (microinterferometry; TIC = Total Interference Contrast in circularly polarized light) can be used to image and measure object structures available in different azimuths.

##### (2) Instrument equipment

- Axio Imager MAT with connected and adjusted HAL 100 halogen illuminator.
- EC Epiplan-Neofluar, Epiplan objectives additionally labeled with "DIC" or "Pol".
- 6x20 compensator mount or 4-position modulator turret
- 6x20 TIC slider with accompanying C DIC P&C reflector module.

##### (3) Setting reflected-light TIC

- Place the specimen (e.g. a step-shaped object) on the stage and prepare the microscope as described in Section 4.9.7 for reflected-light brightfield.
- Rotate reflector turret to swing C DIC P&C reflector module into the light path.
- Push 6x20 TIC slider into 6x20 compensator mount (4-129/4) or rotate turret wheel (4-130/5) of 4-position modulator turret (4-130/6) into TIC-Position (**TIC**). In the field of view, colored interference fringes appear. Turn setscrew (4-132/2) of the TIC slider or the modulator turret to shift the black interference fringe until it appears to be in the center of the field of view.
- To select the structure to be measured, turn control wheel (4-132/1) of TIC slider or modulator turret until the interference fringe system is vertical to the splitting direction of the specimen (see Fig 4-133). The interference fringes can be shifted by means of setscrew (4-132/2) of the TIC slider or the modulator turret.



Please refer to Section 4.9.9 (5) for the use of the motorized four-position modulator turret for the reflected-light TIC technique.

The step height is determined subsequently according to the following formula:

$$d = \frac{n\Delta}{2} = \frac{\lambda b}{2a}$$

where: d = step height in nm

n = refractive index of the environment, usually air (n = 1)

Δ = path difference

a = spacing of interference fringes

b = offset of interference fringes at the step

λ = wavelength of the illumination in nm

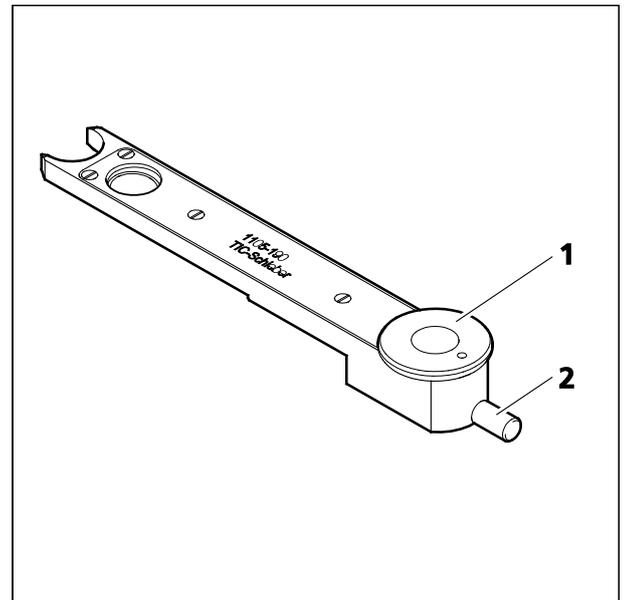
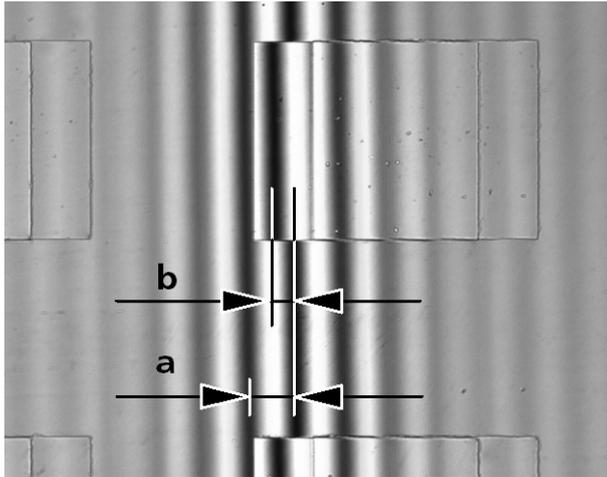


Fig. 4-132 6x20 TIC slider



**Fig. 4-133 Interference fringe pattern**

The values for  $a$  and  $b$  (see Fig. 4-133) are determined using the eyepiece reticle or the micrometer eyepiece.

If you are working with white light (without interference filter),  $\lambda = 550 \text{ nm}$  must be used. When using interference filters, their center wavelength applies.

The measured path difference is aperture-dependent and decreases with increasing illumination aperture.

Accordingly, the following correction values must be taken into account depending on the objective used:

Objective	Correction factor $k$
5x/0.15	1.0057
10x/0.25	1.0161
10x/0.30	1.0236
20x/0.4	1.0436
20x/0.50 and 50x/0.75	1.0718
50x/0.60	1.1111
50x/0.75 and 100x/0.75	1.2038
50x/0.80	1.2500
50x/0.90 and 100x/0.90	1.3929
10x/0.95	1.5241

**Table 1: Aperture-dependent correction**

**Example:**

$a = 11 \text{ mm}$

$b = 5 \text{ mm}$

$\lambda = 550 \text{ nm}$

Objective 20x/0.50

$$d = \frac{\lambda \cdot b \cdot k}{2a} = \frac{550 \text{ nm} \cdot 5 \text{ mm} \cdot 1.0718}{22 \text{ mm}} = 134 \text{ nm}$$

**Caution:**

- If the step and the environment are of different materials, the phase jumps inherent to the material must be taken into account. Whilst the phase jump for all non-conductors is 180° and for semiconductors only deviates slightly from 180°, i.e. the measuring error is negligible, the measured values may be falsified for metals on glass for example. The phase jumps in table 2 calculated for vertically incident light and compact material are to serve as recommended values because it can be assumed that the phase jumps depend on the layer thickness and the angle of incidence of the light. Accurate determination of the thickness is only possible by coating the entire object with a homogeneous layer and then measuring the path difference.
- If the layers or steps are transparent such as silicon dioxide on silicon, the interference fringes may change their color and the determination of the interference order then become problematical. This can also be remedied by additionally coating the surface with a homogenous layer.

Material	Phase jump $\phi$
Copper	140.0°
Gold	142.5°
Silver	151.0°
Bismuth	151.0°
Nickel	157.0°
Iron	157.5°
Zinc	159.0°
Platinum	160.0°
Aluminum	160.0°
Tin	160.5°
Chromium	165.0°
Carbon	160.0°
Graphite	165.0°
Silicon	177.0°
Glass	180.0°

**Table 2: Calculated phase jumps for compact material and vertically incident light**

Half the difference of the phase jumps is included in the determination of the thickness:

$$d = \frac{\Delta}{2} - \frac{\delta\phi}{2}$$

**Example: Extreme case of copper on glass**

$\Phi_{\text{copper}} = 140^\circ$ ,  $\Phi_{\text{glass}} = 180^\circ$  therefore part of the phase jump

$$\frac{\delta\phi}{2} = 20^\circ \text{ or } \frac{\lambda}{18} = 30 \text{ nm}$$

without taking the phase jump inherent in the material into account the measured value would be 30 nm too large.

### 4.9.11 Setting epi-fluorescence



#### CAUTION

To reduce the transmission, use an FL attenuator, discrete (423616-0000-000 or 423617-0000-000). The gray filters mounted in the 2-position filter wheels (428300-0000-000 or 428301-0000-000) are not permanently stable.

#### (1) General principle

The epi-fluorescence technique enables high-contrast images of fluorescent substances to be displayed in typical fluorescence colors. In the epi-fluorescence microscope, light generated by a high-performance illuminator reaches the exciter filter (band pass) through a heat-absorbing filter. The filtered, short-wave excitation light is reflected by a dichroic beam splitter and focused on the specimen via the objective. The specimen absorbs the short-wave light and then emits the long-wave fluorescence light (Stoke's law), which is now gathered by the objective and transmitted by the dichroic beam splitter. Finally, the rays pass a barrier filter (long pass/band pass), which only allows the long-wave light from the specimen to be transmitted.

Exciter and barrier filters must be perfectly matched. They are arranged in a reflector module FL P&C together with the corresponding dichroic beam splitter.

#### (2) Instrument configuration

- Recommended objectives: Plan-Neofluar or Fluar objectives (UV excitation)
- Reflector module FL P&C in reflector turret
- Mercury vapor short-arc lamp HBO 100 for reflected-light illumination
- Halogen illuminator HAL 100 for transmitted-light illumination



Before using the epi-fluorescence technique, make sure to align the mercury vapor short-arc lamp by means of the adjusting aid as described in Section 3.31.3. Re-alignment may be necessary depending on the operating time.

#### (3) Setting epi-fluorescence

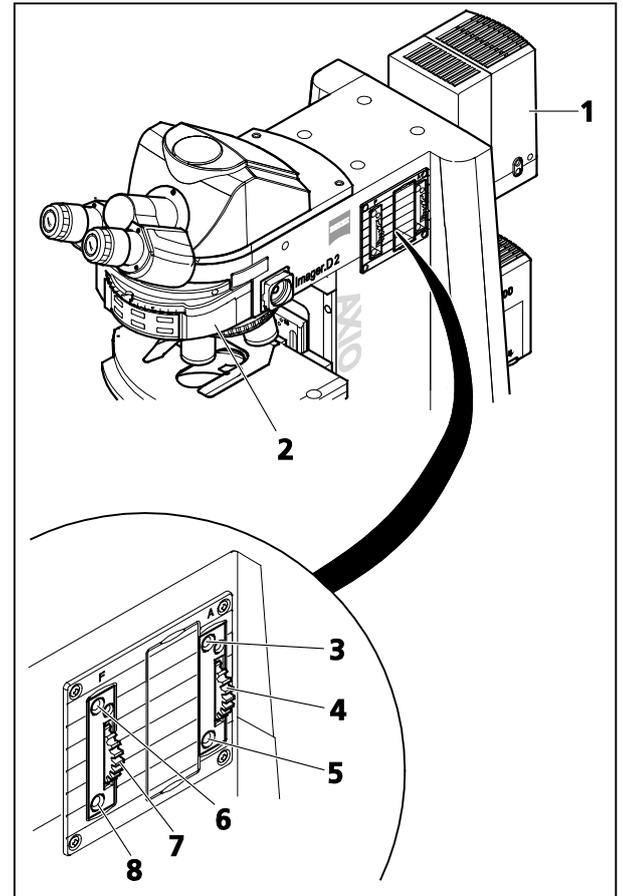
The first epi-fluorescence setting is considerably simplified if you begin with the Plan-Neofluar objective 20x/0.50 and a strongly fluorescing specimen. You may also use demonstration specimens first.



Before setting epi-fluorescence, make sure to remove compensator  $\lambda$  (4-120/7) from the slot above the nosepiece, which may have been left there from a previously performed transmitted-light DIC examination.

- Switch on halogen illuminator HAL100.
- Swivel in Plan-Neofluar objective 20x/0.50.
- First, swivel condenser turret to brightfield position H (or phase contrast Ph) and set the specimen feature to be examined.
- For the time being, keep the light path in the reflected-light illuminator blocked by reflected-light shutter RL (rear right on microscope stand) (indicator LED is lighting).

- Switch on the HBO 100 mercury vapor short-arc lamp (4-134/1) and allow it to warm up to operating temperature for about 15 minutes.
- On the reflector turret (4-134/2), select the reflector module FL P&C containing the desired fluorescence filter combination (depending on the desired kind of excitation) and swivel it in.
- Remove the reflected-light shutter RL from the light path in the reflected-light illuminator and close the transmitted-light shutter TL.
- Remove one eyepiece from the tube and set the aperture diaphragm with your eye. For that, open the aperture diaphragm (4-134/4) until it clears the full exit pupil of the objective. Center the aperture diaphragm to the exit pupil using the two centering screws (4-134/3 and 5), if necessary.
- Reinsert the eyepiece in the tube and close the luminous-field diaphragm (4-134/7) until it is visible in the field of view.
- Use the two centering screws (4-134/6 and 8) to center the luminous-field diaphragm to the edge of the field of view.
- Open the luminous-field diaphragm until it just disappears from the field of view or, if there is a risk of specimen bleaching, close it so far that it is visible in the field of view.
- Finally, refocus on the specimen and optimize the position of the HBO 100 collector as described in Section 3.31.3. Adjust the collector in such a manner that the field of view is illuminated as evenly as possible when using the short-wave excitation reflector module. When you use long-wave excitation modules, correction of the collector position is not required.



**Fig. 4-134** Components for epi-fluorescence on Axio Imager

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#### 4.9.12 Setting reflected-light polarization – Detection of bireflection and reflection pleochroism

##### (1) Use

Incident-light polarization presents another contrasting option for polished sections of ore minerals, coals, ceramic products, certain metals and metal alloys, as these specimens often show a varied reflection behavior in linearly polarized light depending on the orientation of the crystals and specimen features.

The illumination light is linearly polarized by the polarizer and directed through the objective onto the specimen surface, where it is reflected. Here, the light rays experience structure-dependent path differences or polarization-optical rotations, which on passing the analyzer appear as different gray-scale values. A compensator with lambda plate enables the conversion from gray to color contrast. **Important! However, you must not use a 6x20 compensator for this purpose!**

With objectives of very low magnification, a rotatable  $\lambda/4$  plate arranged in front of the objective (Antiflex cap) permits the reflections to be eliminated even with "dark" specimen surfaces, which otherwise would be unavoidable.

##### (2) Microscope configuration

- Stand with installed and aligned HAL 100 halogen illuminator.
- Epiplan-Neofluar Pol, EC Epiplan-Neofluar Pol, Epiplan Pol objectives.
- Reflector module DIC P&C or DIC Rot I P&C in reflector turret;  
or reflector module Pol P&C plus analyzer slider;  
or analyzer slider plus polarizer slider.

##### (3) Setting reflected-light polarization

- Set the microscope for reflected-light brightfield as described in Section 4.9.7.
- If you use the objective position with DIC slider slot, remove the possibly inserted DIC slider.
- On reflector turret (4-135/3), swing the reflector module DIC P&C (4-135/4) into the light path. You may also swing in reflector module Pol P&C and insert the analyzer slider in the corresponding slot. When using the combination of analyzer slider (4-135/1) and polarizer slider (4-135/2), you may also push these into the corresponding slots. If you use the fixed versions of these sliders, the polarizer is oriented in EAST-WEST direction and the analyzer in NORTH-SOUTH direction.
- Put specimen onto stage, set the desired magnification, focus and observe the specimen in the now adjusted polarization contrast while rotating the stage.

If the specimen features show variations in brightness and color, when the stage is rotated, the specimen is said to have bireflection.

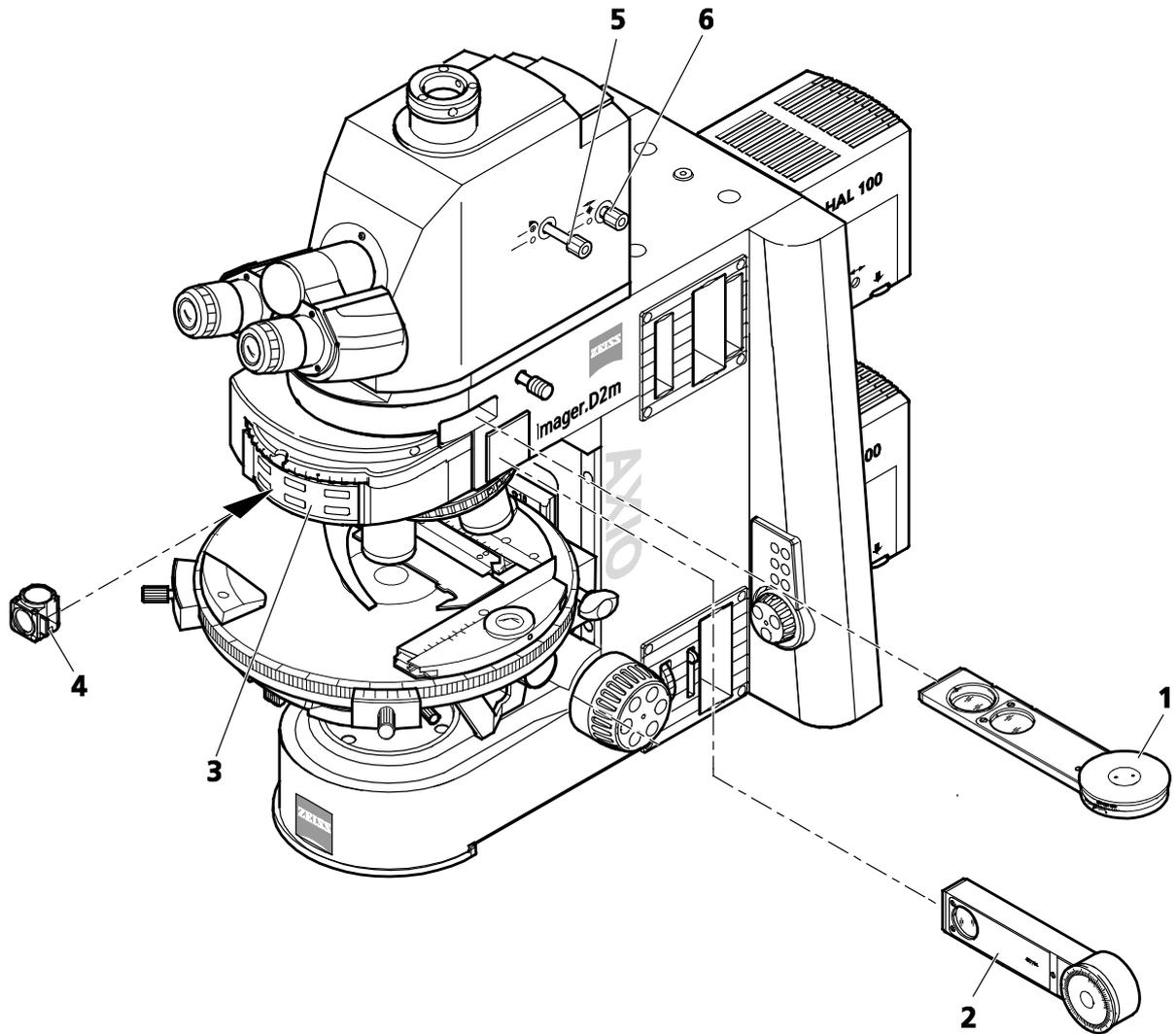
If the specimen has a weak bireflection only, it is advisable to use the analyzer with rotatable lambda plate.

Pleochroism can be made out by color variations occurring in the specimen while rotating the stage (with the reflected-light polarizer moved into the light path and the analyzer moved out).



If the microscope is equipped with the phototube Pol, the following settings are required for this contrast method:

- The Bertrand lens must be inactive (rear push-pull rod (4-135/6) on right side pulled out).
- The luminous-field diaphragm must be open (front push-pull rod (4-135/5) turned counterclockwise as far as it will go).
- The crossline reticle must be inactive (front push-pull rod (4-135/5) pulled out).



**Fig. 4-135** Components for reflected-light polarization

## 5 CARE, MAINTENANCE, TROUBLESHOOTING AND SERVICE

### 5.1 Instrument care

Care of the Axio Imager is restricted to the following operations:

- Eliminate contaminations on the instrument according to the rules for prevention of accidents.
- Switch off the instrument each time after use and place the instrument cover on it to protect it from dust and humidity.
- Do not set up the instrument in a humid room; maximum humidity < 75%.
- Remove dust and loose dirt on visible optical surfaces with a brush, blower brush, cotton bud, optics cleaning tissue or cotton cloth.
- Remove water-soluble dirt (coffee, cola, etc.) by blowing on it and wiping it off with a lint-free cotton cloth or a cloth moistened with water to which you may also add a mild detergent.
- Wipe off stubborn, oily or fatty dirt, e.g. fingerprints and immersion oil, with a cotton swab or lint-free cotton cloth and the optics cleaning solution L.

The cleaning solution consists of 90 vol% gasoline and 10 vol% isopropanol (IPA). The individual components are also known as:

Gasoline:	Medical alcohol, petrolether
Isopropanol:	2-Propanol, Dimethylcarbinol, 2-Hydroxypropane

Clean optical surfaces by polishing in circles, starting in the middle and moving to the edges (only use slight pressure).

When using the microscope in humid climatic zones, proceed as follows:

- Store the instrument in bright, dry and well-ventilated rooms with a humidity of < 75 %. Optical components and accessories that are particularly susceptible to fungus growth, e.g. objectives and eyepieces, should be stored in a dry closet.

The risk of fungus growth on opto-mechanical instruments invariably exists in the following conditions:

- Relative humidity > 75% and temperatures between +15 °C and +35 °C for more than three days.
- Installation in dark rooms without air ventilation.
- Dust deposits and fingerprints on optical surfaces.

## 5.2 Instrument maintenance

### 5.2.1 Performing checks

- Makes sure the line voltage available agrees with the operating voltages required.
- Check the power cable and plug for defects.
- If any damage is visible, switch off the instrument. Have the instrument repaired only by a qualified technician.
- Check the reading of the operating hour meter of the power supply to ensure that the maximum operation time of the mercury vapor short-arc lamp is not exceeded.

### 5.2.2 Changing fuses on the manual stand



Always pull the power plug before changing the fuses.

Should any fuse fail, the cause of such failure must first be determined, with any technical defect that may exist to be properly repaired by trained personnel

On the manual Axio Imager model, the fuse compartment is on the back of the microscope and contains two **T 5.0 A/H / 250 V, 5x20 mm** fuses.

- Pull the power plug.
- Pull out fuse holder (5-1/2) frontward. Use a small screwdriver, if necessary.
- Remove the fuses from the holder and insert new fuses.
- Push the fuse holder back into the fuse compartment (5-1/1) as far as it will go.
- Connect the power plug.

The motorized Axio Imager model is powered through the power supply VP232-2. (For fuse changes, see Section 5.2.3).

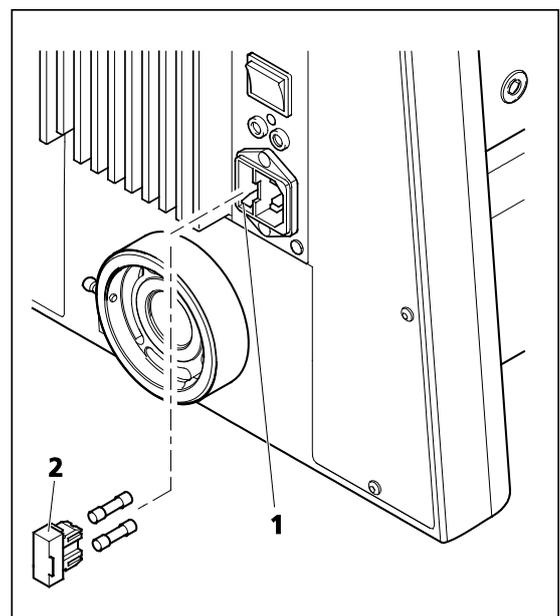
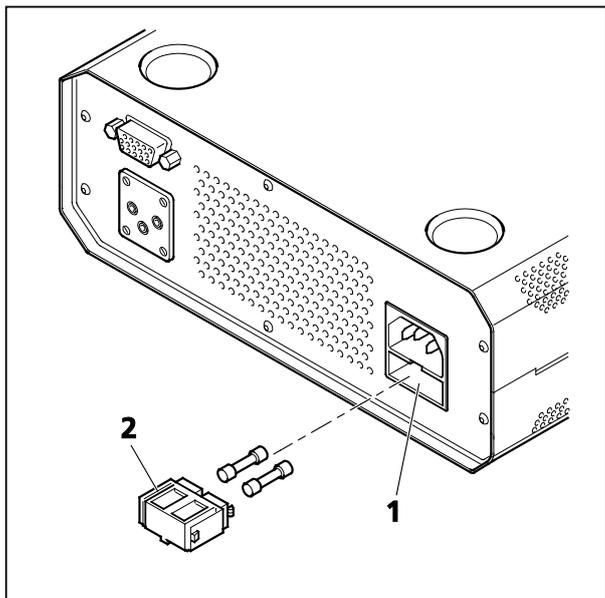


Fig. 5-1 Changing fuses on the stand



**Fig. 5-2** Changing fuses on the power supply

### 5.2.3 Changing fuses on the VP232-2 CAN power supply



Be sure to pull the power plug before changing fuses.

Should any fuse fail, the cause of such failure must first be determined, with any technical defect that may exist to be properly repaired by trained personnel.

The fuse compartment is located on the back of the power supply and contains two type **T 4.0 A / 250 V, 5x20 mm fuses**.

- Pull the power plug.
- Pull out the fuse holder (5-2/2) frontward. Use a small screwdriver, if necessary.
- Remove the fuses from the fuse holder and replace them with new ones.
- Push the fuse holder into the fuse compartment (5-2/1) as far as it will go.
- Plug in the power plug.

### 5.3 Troubleshooting

Problem	Cause	Remedy
Shadows or inhomogeneous image brightness in the field of view; the field is not entirely visible.	The vis/doc push-pull rod on the phototube is not in the correct position (intermediate position).	Move the vis/doc push-pull rod to the correct position (end position).
	The nosepiece with objective has not clicked into place.	Turn on the nosepiece with the objective until it clicks into place.
	The condenser has not been set correctly.	Set the condenser correctly (adjustment, centering); see p. 145 ff.
	The aperture diaphragm has not been set correctly.	Set the aperture diaphragm correctly (centering, aperture); see p. 145 ff.
	The luminous-field diaphragm has not been set correctly.	Set the luminous field-diaphragm correctly (centering, opening); see p. 145 ff.
	The filter has not been inserted correctly in the filter mount.	Insert the filter correctly in the filter mount.
No DIC effect	The polarizing elements are not in the light path.	Move the polarizing elements into the light path and set them to crossed position.
Double images in bright-field examinations	DIC slider in the light path.	DIC slider in the light path.
Low resolving power and poor image contrast.	The aperture diaphragm has not been opened to the correct size.	Set the aperture diaphragm as per the 2/3 rule or the specimen features; see p. 146 ff.
	The condenser has not been focused correctly and front lens 0.9 not swiveled in / out correctly.	Focus the condenser and swivel front lens 0.9 correctly in or out; see p. 146 ff.
	Wrong cover slip thickness for transmitted-light objectives corrected for 0.17 mm cover slip thickness.	Use standardized 0.17 mm cover slips.
	Specimen slide placed upside down.	Turn over the specimen slide; the specimen side should be on top.
	Use of no or non-specified immersion oil with immersion objectives.	Use immersion oil 518 F <sup>®</sup> from Carl Zeiss; see p. 186.
	Air bubbles in the immersion oil.	Remove the bubbles by applying new oil.

<b>Problem</b>	<b>Cause</b>	<b>Remedy</b>
Low resolving power and poor image contrast.	Immersion oil on the front lens of a dry objective.	Clean the front lens of the dry objective; see p. 180.
	The cover-slip correction ring has not been set to the correct cover-slip thickness.	Set the correction ring to the correct thickness; see p. 26.
	Dirt or dust on the optical surfaces of objectives, eyepieces, condensers or filters.	Clean the respective optical components; see p. 180.
Asymmetrically blurred images, e.g. one side in focus, one side not in focus.	The condenser has not been set correctly.	Set the condenser correctly; see p. 146 ff.
	The nosepiece has not clicked into place.	Turn on nosepiece until it clicks into place (click-stop).
	The specimen has not been clamped in position on the stage.	Correctly insert the specimen in the specimen holder and clamp it.
Greater focus differences after objective change.	The focusing eyepieces have not been set correctly.	Set the focusing eyepieces to the appropriate ametropia; see p. 35.
	The objective has not been screwed in completely.	Screw objective fully in as far as it will go.
	The tube lens has either not been inserted or inserted superfluously.	Insert the tube lens or remove the superfluous tube lens.
Left and right fields of view cannot be combined to one image.	The interpupillary distance of the binocular tube has not been set correctly.	Set the interpupillary distance correctly; see p. 36.
	The focusing eyepieces have not been set correctly.	Set the focusing eyepieces to the appropriate ametropia; see p. 35.
Eye-fatiguing microscopy.	The interpupillary distance of the binocular tube has not been set correctly.	Set the interpupillary distance correctly; see p. 36.
	The focusing eyepieces have not been set correctly.	Set the focusing eyepieces to the appropriate ametropia; see p. 35.
	The image brightness is not acceptable.	Adjust the lamp voltage or insert the conversion filter.
	Binocular tube optically / mechanically out of alignment.	Have it checked / repaired by Microscopy Service.

<b>Problem</b>	<b>Cause</b>	<b>Remedy</b>
Dirt or dust in the field of view.	The condenser has not been focused correctly and front lens 0.9 not swiveled in/out correctly.	Focus the condenser and swivel front lens 0.9 correctly in or out; see p. 146 ff.
	The aperture diaphragm opening is too small.	Set the aperture diaphragm as per the 2/3 rule or the specimen features; see p. 146 ff.
	Dirt or dust on the optical surfaces of the objectives, eyepieces, condensers, filters or specimens.	Clean the optical surfaces of the affected components; see p. 180.
The 12V 100W halogen lamp does not function although the on / off switch is set to "on".	The power plug has not been connected to the power outlet.	Connect the power plug to the power outlet considering the power requirements of the instrument.
	12V 100W halogen lamp has not been installed.	Insert 12V 100W halogen lamp; see p. 58.
	12V 100W halogen lamp is defective.	Replace 12V 100W halogen lamp; see p. 58.
	Failure to use the specified 12V 100W halogen lamp.	Use the specified 12V, 100W halogen lamp; see p. 186.
	The fuses are defective.	Replace defective fuses; see p. 181.
	The electronic module is possibly defective.	Have electronic module checked by service and replaced, if required; see p. 187.
	The power outlet does not supply line voltage.	Use another power outlet.
The 12V 100W halogen lamp is flickering, the light intensity is unstable.	End of average service life of 12V 100W halogen lamp.	Replace 12V, 100W halogen lamp; see p. 58.
	Incorrectly installed or broken power cable.	Connect the power cable correctly or replace it.
	The pins of the 12V 100W halogen lamp haven't been inserted correctly in the receptacle.	Insert the pins of the 12V 100W halogen lamp correctly in the receptacle; see p. 58.

**5.4 Spares, consumables and tools**

<b>Designation</b>	<b>Cat. No.</b>	<b>Application</b>
12V 100W halogen lamp	380079-9540-000	For HAL 100 illuminator
HBO 103 W/2 mercury vapor short-arc lamp	380301-9350-000	For HBO 100 illuminator
SW 1.5 ball-headed screwdriver *	000000-0460-470	For changing condensers and stages
SW 3 ball-headed screwdriver *	000000-0069-551	For changing tubes and illuminators
SW 4 offset screwdriver *	000000-0015-278	For removing the carrying handle
Eyepiece eyecup	444801-0000-000	Recommended for low-light techniques to suppress reflections
Dust cover for nosepiece Dust cover for eyepiece sockets	462981-0000-000 000000-0168-373	For closing instrument openings not in use
Immersion medium Immersol 518 F <sup>®</sup> ; Oiler, 20 ml Bottle, 100 ml Bottle, 250 ml	444960-0000-000 444962-0000-000 444963-0000-000	For oil immersion applications $n_D = 1.518$
Cleaning tissue, 300 sheets	462975-0000-000	For cleaning optical surfaces
G-fuse inserts (5 x 20 mm); T 5.0 A/H / 250 V; 2x (Stand, manual) G-fuse inserts (5 x 20 mm); T 4.0 A / 250 V; 2x (Power supply VP232-2 for motorized stand)		Protects the integrated power supply from excessive load  Protects the power supply from excessive load
Dust cover set M Dust cover set L Set of cover caps (included in 434303-0000-000 and 434304-0000-000)	434303-0000-000 434304-0000-000 434302-0000-000	For covering the instrument when it is not in use. For covering eyepieces

\* Included in the tool kit that is supplied with the instrument in tool bag (451892-0000-000).

## 5.5 Requesting service

All repairs of mechanical, optical or electronic components inside the instrument and of the electrical components of the Axio Imager microscopes may only be performed by Carl Zeiss service staff or specially **authorized** personnel.

To ensure optimum setting and trouble-free function of your microscope over a longer period of time, we recommend that you enter into a service/maintenance agreement with Carl Zeiss.

For subsequent orders or when service is required, please get in touch with your local Carl Zeiss representative.

For additional information, contact us at

[micro@zeiss.de](mailto:micro@zeiss.de)

or visit us on the Internet at

<http://www.zeiss.de/Axiolmager>

<http://www.zeiss.de/Axiolmagermat>

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**6 APPENDIX****6.1 List of abbreviations**

AC	Alternating Current
ACR	Automatic Component Recognition
Br.	Suitable for eyeglass wearers
cod.	Coded
CSA	Canadian Standards Association
D	Cover slip thickness
D / DF	Darkfield
d	Diameter (e.g. of filters)
DIC	Differential Interference Contrast
DIN	Deutsches Institut für Normung (German Standards Institute)
doc	Documentation
EC	European Community
EN	European standard
EMC	Electromagnetic Compatibility
FL	Fluorescence
foc.	focusing
H	Brightfield
HAL	Halogen lamp
HBO	Mercury vapor short-arc lamp for fluorescence
ICS	Infinity Color-Corrected System
IEC	International Electrotechnical Commission
IP	Internal Protection (protection by instrument casing)
ISO	International Organization for Standardization
LED	Light Emitting Diode
man.	manual
MC	Microscope Camera
mot.	motorized
$n_D$	Refractive index for D line (sodium)
Ph	Phase contrast
PL	Plan
Pol	Polarization
P&C	Push&Click
R	Right (drive knob to the right of the mechanical stage)
RL	Reflected light

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SW	Wrench size across flats
T	Slow-blow (fuse type)
TL	Transmitted light
TV	Television
UL	Underwriter Laboratories
UV	ultraviolet
VDE	Verband Deutscher Elektrotechniker (Association of German Electricians)
vis	visual
V <sub>obj</sub>	Magnification of objective
W 0.8"	Whitworth-type thread 0.8"
W-PL	Wide-field eyepiece
XBO	Xenon short-arc lamp

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### **6.3 Industrial property rights**

Instruments, instrument components or methods described in this manual are protected by patents and registered utility models:

**US6276804**

**US6392796**

**US5015082**

**CH691699**

**GB2306585**

**US6123459**

**DE29821694**