

# **Operating Manual**

## **ApoTome.2**

Knowledge of this manual is required for the operation of the instrument. Would you therefore please make yourself familiar with the contents of this manual and pay special attention to hints concerning the safe operation of the instrument.

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

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# 1 INTRODUCTION

## 1.1 Important notes



The ApoTome.2 system is an extension to the Axio Imager, Axio Observer, Axiovert 200 and Axio Zoom.V16 microscopes. The ApoTome.2 system consists of a slider, which is inserted in the plane of the luminous-field diaphragm of the incident-light beam path, and an accompanying control box.

A complete ApoTome.2 imaging system comprises the following components:

- microscope
- anti-vibration table or anti-vibration microscope support
- cooled digital camera with high dynamic range
- PC with at least 1 GB RAM according to Carl Zeiss specifications plus monitor, operating system Windows® XP, Windows® Vista (32 bit version) or Windows® 7 (32 bit version)
- ApoTome.2 slider with ApoTome control box
- Imaging software AxioVision 4.8.2 control software, service pack 1 / ZEN 2011 (blue edition)



This Operating Manual describes the use of the ApoTome.2 hardware. For notes and instructions for the control of the ApoTome.2 through software, please refer to the Operating Manual of the imaging software AxioVision / ZEN 2011 (blue edition).



Prior to the initial start-up of the ApoTome.2, read the notes on instrument safety as well as the chapters Instrument Description (Chapter 2) and Startup (Chapter 3). Especially note that on stands of the Axiovert series delivered before May 1, 2002 the base plate of the fluorescence beam path must be replaced by a new base plate (000000-1197-874).

## 1.2 Notes on device safety

The ApoTome.2 was designed, produced and tested in compliance with DIN EN 61010-1 (IEC 61010-1) "Safety requirements for electrical measuring, control and laboratory instruments" and meets the requirements of the EC directives 2006/95/EC "Low Voltage Directive" and 2004/108/EC "Electromagnetic Compatibility".

The device is to be disposed of according to the WEEE Directive 2002/96/EC.

It is marked with the  mark.

This Operating Manual includes information and warnings that must be observed by the user.

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

**CAUTION**

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

**CAUTION**

*Hot surface!*

**CAUTION**

*Emitted LED radiation!*

**CAUTION**

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

**CAUTION**

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

**NOTE**

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

The ApoTome.2 may only be used for the techniques described in this manual.

Particularly observe the following notes:



The manufacturer cannot assume any liability for any other application, including that of individual modules or single parts. This also applies to all service or repair work that is not carried out by authorized service personnel. Moreover, 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 the Zeiss Service department or Carl Zeiss Microscopy Service to repair the instrument.



The ApoTome.2 control box is equipped with an integrated power supply which permits the use of line voltages in the range between 100 and 240 V  $\pm$ 10 %, 50 / 60 Hz, without having to adjust the line voltage on the instrument.



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



Make sure to use only fuses of the rated power required. The use of makeshift fuses and the short-circuiting of the fuse holders are not permitted.



The ApoTome.2 slider and the ApoTome.2 control box are not equipped with any special devices for the protection from corrosive, potentially infectious, toxic, and radioactive specimens or other specimens that could be hazardous to health. When handling such specimens, observe all legal regulations, particularly the relevant national regulations for the prevention of accidents.



Gas-discharge lamps, e.g. HBO 100, emit ultraviolet radiation that can cause burns to the eyes and skin. Therefore, never look directly into the light of these lamps and avoid direct incidence of their light on your skin. When using the microscope, always use the protective units 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, replace them only when they have cooled down and make sure to wear protective gloves and a face guard. (For details, please refer to Operating Manual HBO 100.)



When using fluorescence filters, do not remove the heat-absorbing filter for the protection against heat emitted by the microscope illuminator 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 replacing the lamp and allow the instrument to cool down for approximately 15 minutes.



Do not operate the device in potentially explosive environments.





Dust and dirt can impair the performance of the instrument. Therefore, protect the instrument from these influences as far as possible and cover it with the dust cover when not in use. Always check whether the instrument is switched off before you cover it.



Clogging or covering ventilation slots can lead to heat accumulation that can damage the instrument and, in extreme cases, cause fire. Always keep the ventilation slots clear and make sure that no objects enter the instrument through these slots.



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 ApoTome.2 is a high-precision instrument that can be impaired in its performance or destroyed when handled improperly.



Do not put defective instruments in household waste. They have to be disposed of in accordance with the WEEE Directive 2002/96/EC.  
Specimens also need to be properly disposed of in conformity with applicable legal provisions and in-house work instructions.

### 1.3 Warning and information labels

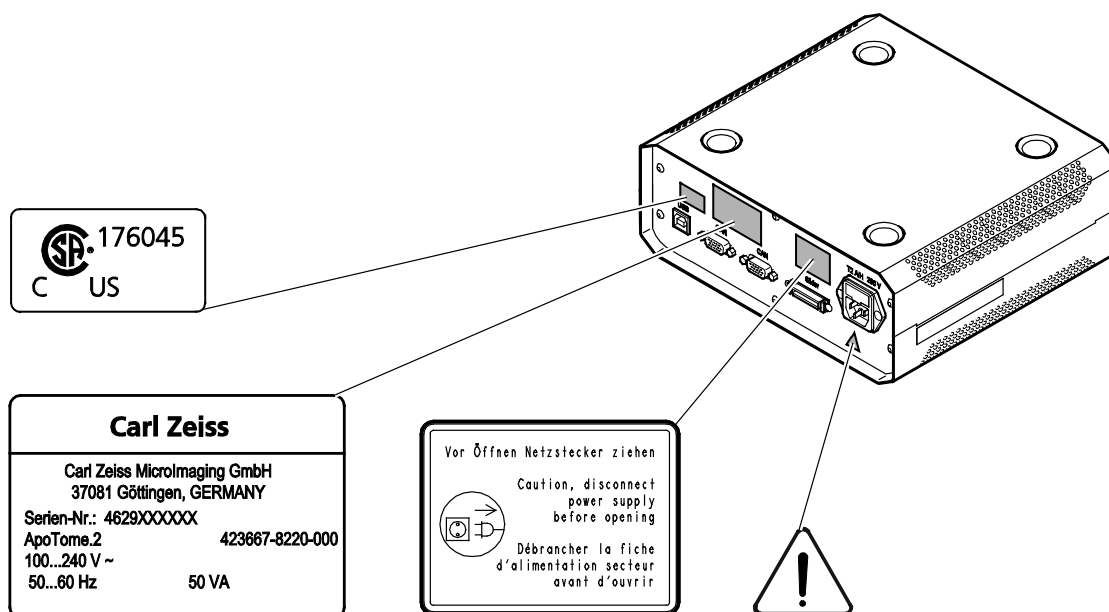


Fig. 1-1 Warning and information labels

### 1.4 Notes on warranty

The manufacturer guarantees that the instrument has no material and production defects when delivered. You must inform us of any defects immediately and minimize any damage. If the manufacturer is informed of such a defect, he is obligated to remove 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 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 ApoTome.2 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 first with the Carl Zeiss Microscopy Service in Germany (see page 30) or with the Carl Zeiss agency in your country.

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## 2 INSTRUMENT DESCRIPTION

### 2.1 Product name and intended use

Manufacturer's product name: **ApoTome.2 for Axio Imager** (423667-9100-000)  
**ApoTome.2 for Axio Observer and Axiovert 200** (423667-9000-000)  
**ApoTome.2 for Axio Zoom.V16** (423667-9200-000)

The ApoTome.2 allows depth-discriminated images (= optical sections) of fluorescence specimens to be produced. Compared to the conventional reflected light fluorescence methods, these optical sections feature increased contrast and enhanced optical resolution in axial direction. Furthermore, optical sections through the specimen are the prerequisite for the three-dimensional reconstruction of structures.

### 2.2 Instrument description and main features

The ApoTome.2 hardware consists of two components:

1. slider with transmission grids changer
2. control box

The slider is directly connected to the control box via a cable. The control box is connected to the PC by a USB cable or, optionally, it can be connected directly to the microscope by a CAN BUS cable. In this case, the ApoTome.2 directly communicates with the PC via the electronic system of the microscope.



If an Axiovert 200 is used, the control box can only be connected to the PC by a USB cable. A connection via CAN BUS is not possible!

Major features of the **ApoTome.2** include:

- ApoTome.2 slider for the plane of the luminous-field diaphragm in the reflected light path
- ApoTome.2 slider with two click-stop positions:

**Click-stop position 1:**

An open passage is provided in the reflected light path. Thus, normal fluorescence observations can be carried out in the wide field, e.g. to find interesting structures and to position the specimens.

**Click-stop position 2:**

In this position, a glass plate with evaporated grid structure is in the light path. The grid structure is laterally moved in the specimen plane by means of a scanner mechanism. By capturing three (or more) images at different grid positions and by subsequent calculation, it is possible to produce an optical section through the specimen. Three different grid frequencies selected via software are available. They allow the user to obtain different section thicknesses and to use different objectives.

2.3 System overview

2.3.1 ApoTome.2 for Axio Imager

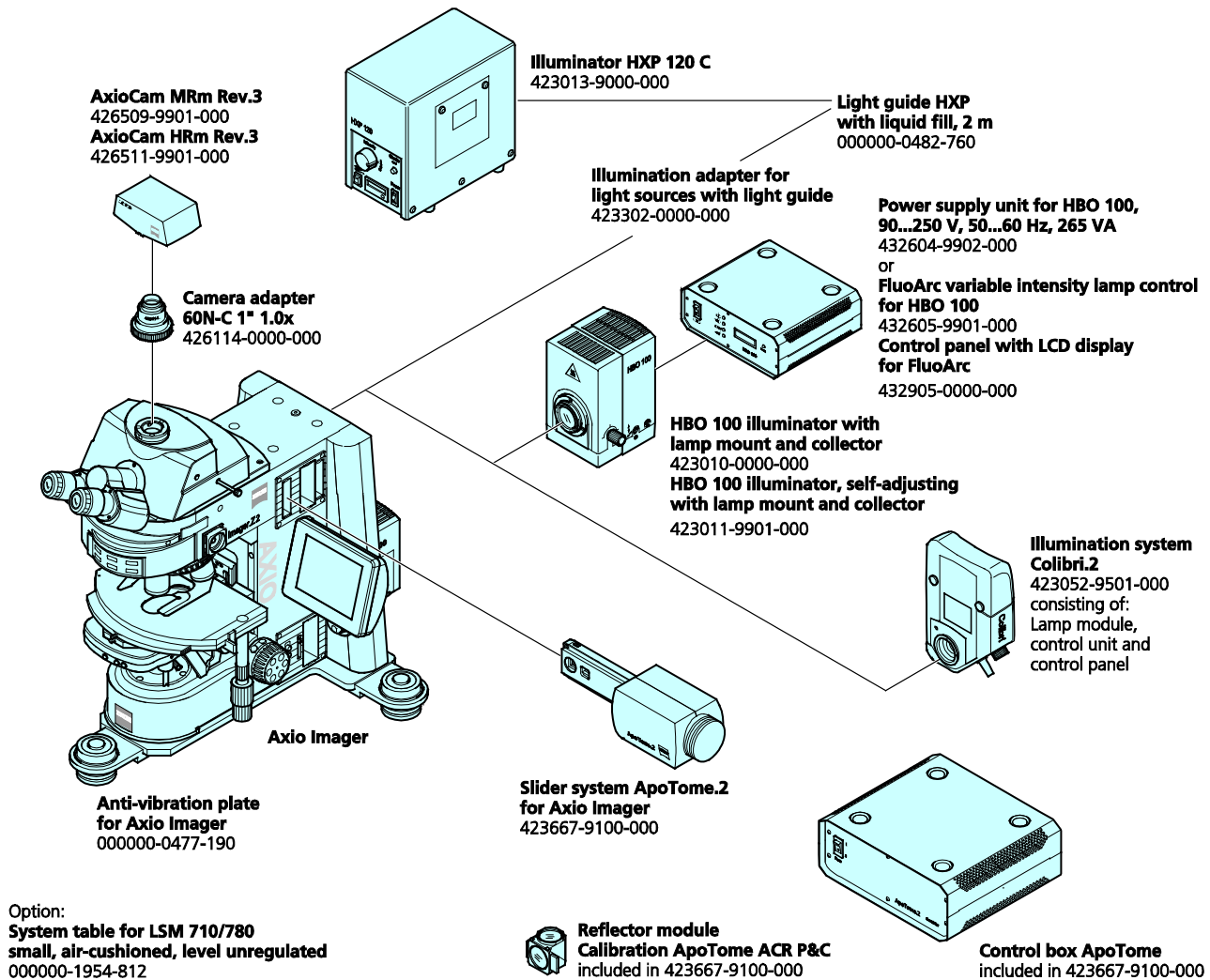


Fig. 2-1 System overview ApoTome.2 for Axio Imager

2.3.2 ApoTome.2 for Axio Observer and Axiovert 200

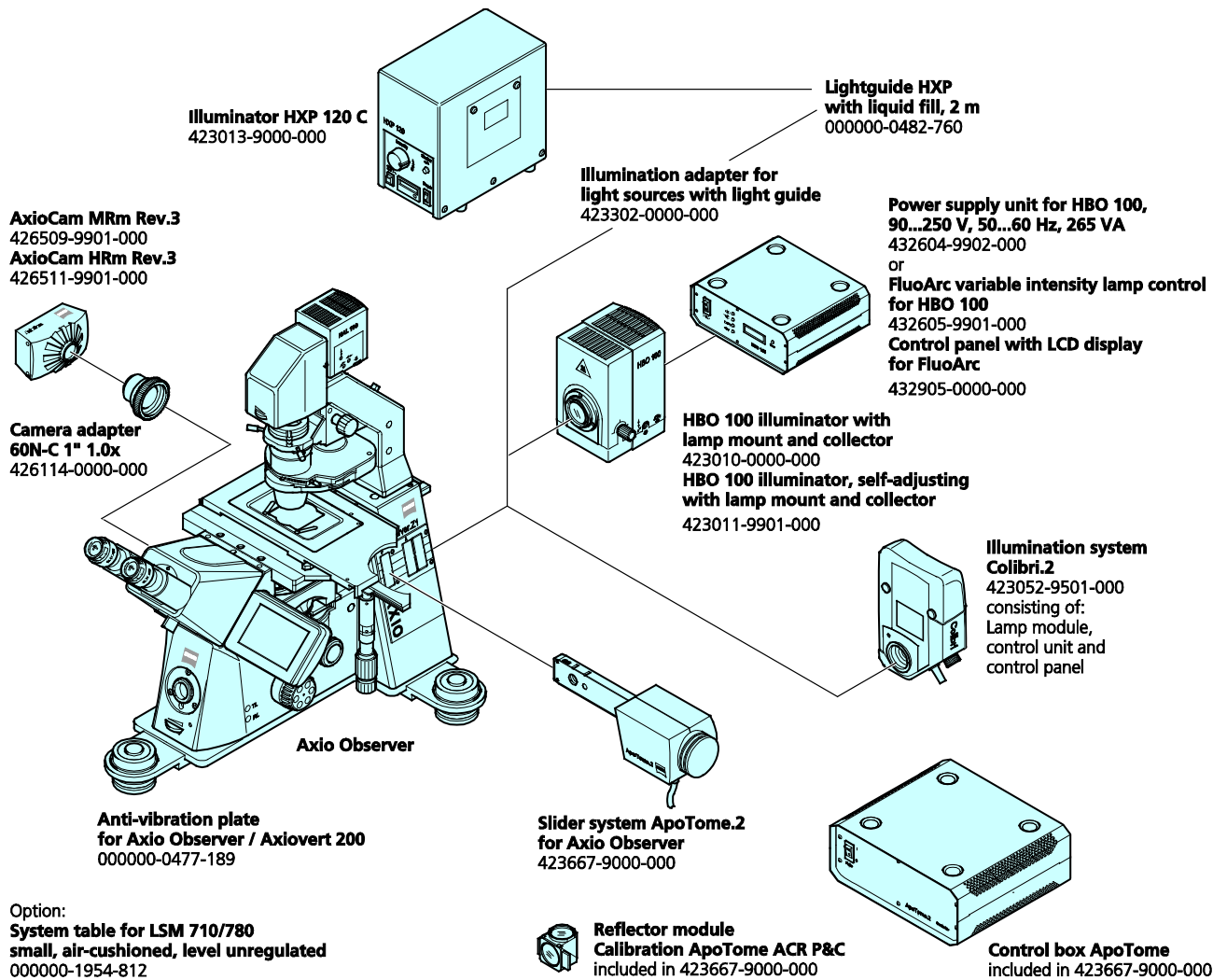


Fig. 2-2 System overview ApoTome.2 for Axio Observer and Axiovert 200

2.3.3 ApoTome.2 for Axio Zoom.V16

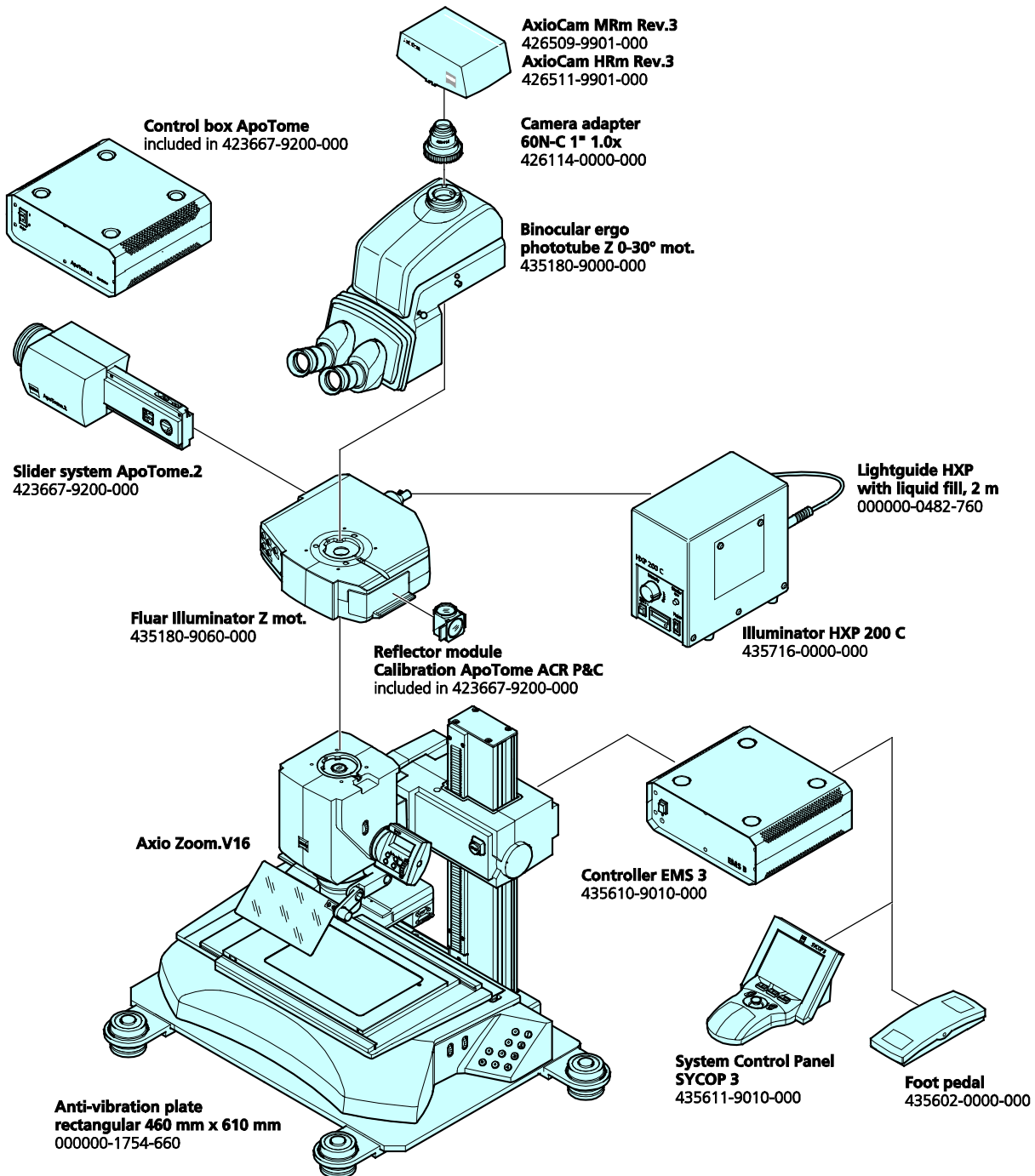
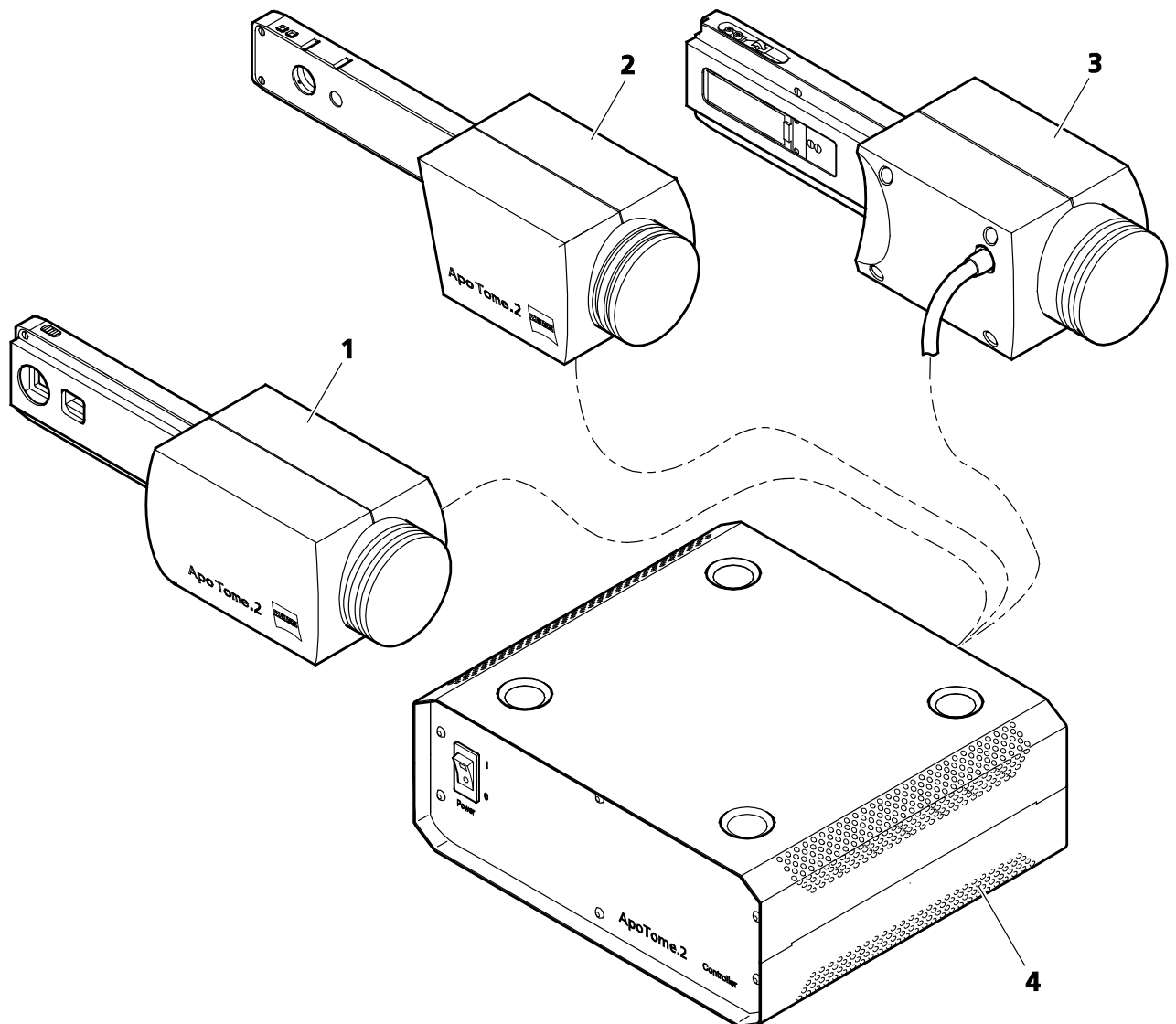


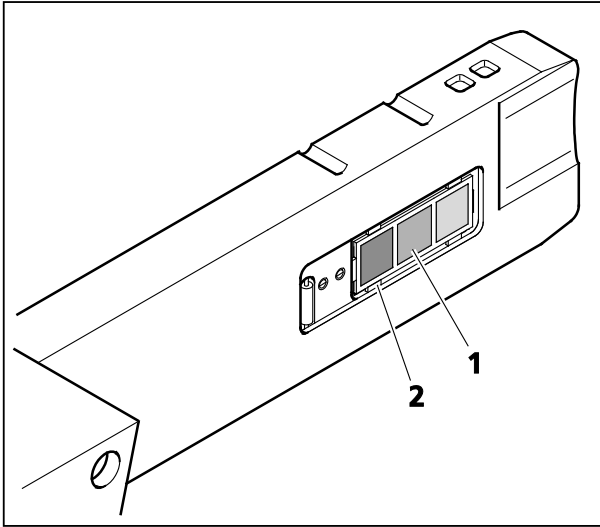
Fig. 2-3 System overview ApoTome.2 for Axio Zoom.V16

## 2.4 General view of ApoTome.2



- 1 Slider ApoTome.2 for Axio Imager
- 2 Slider ApoTome.2 for Axio Observer and Axiovert 200
- 3 Slider ApoTome.2 for Axio Zoom.V16
- 4 Control box ApoTome.2

**Fig. 2-4** Components of ApoTome.2



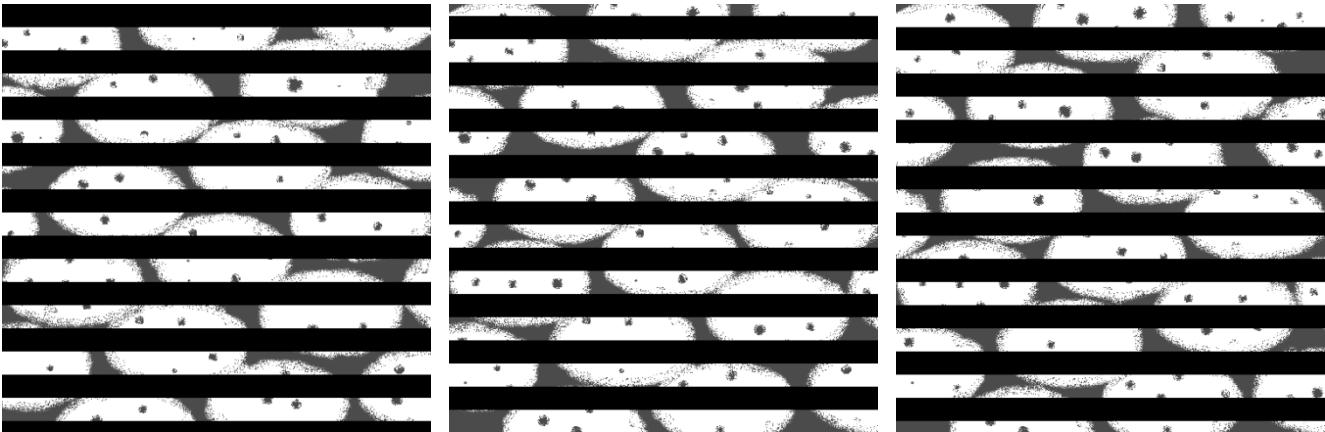
**Fig. 2-5 Changer with transmission grids**

## 2.5 Operating principle

Three different transmission grids (Fig. 2-5/1) are installed in the slider ApoTome.2 on a motorized changer (Fig. 2-5/2). The transmission grids are three metallic grid structures that are evaporated on a glass plate.

If the transmission grid is located in the illumination beam path, the grid structure will be imaged in the specimen plane. For image capture the image of the grid structure is shifted in three defined steps ("phases") in the specimen by means of a scanner mechanism in ApoTome.2. During this procedure a given point of the specimen is illuminated with different intensity depending on the phase position of the transmission grid.

The three images captured (see Fig. 2-6) with the grid structures are calculated, so that an optical section with increased contrast and depth discrimination through the specimen will be obtained. The calculation makes the grid structures of the initial images disappear.



**Fig. 2-6 Image capture in three defined steps (phases)**

The ApoTome.2 slider is provided with three different transmission grids, which cover the total magnification range, as indicated in Chapter 2.6. The software automatically recommends the optimal grid to be used. Optionally, another grid can be selected manually (e.g. to adjust the section thickness to the experimental conditions).

Please refer to Chapter 2.6 for a precise list about the correct transmission grids for the different magnifications.



## 2.6 Suitability of objectives for the use of ApoTome.2

The operating principle described in Chapter 2.5 places specific demands on the correction state of the objectives used.

Especially the correction of the longitudinal color aberration must be of high quality. Shifting the grid structure along the optical axis minimizes the residual error.

The recommended objectives for the Axio Imager or Axio Observer / Axiovert 200 or Axio Zoom.V16 microscopes are listed in the following tables.

### Axio Imager

Objective	M	NA	Immersion	Grid / Section thickness @ 490nm [RU/μm]			DAPI with FS 34	DAPI with FS 49
				High grid	Middle grid	Low grid		
EC Plan-Neofluar	10x	0,3	Air	2,9 / 31,9	1,7 / 18,2	<b>0,9 / 9,9</b>	✓	✓
EC Plan-Neofluar	20x	0,5	Air	2,4 / 9,2	1,4 / 5,3	<b>0,7 / 2,9</b>	✓	✓
EC Plan-Neofluar	40x	0,75	Air	1,6 / 2,8	<b>0,9 / 1,6</b>	0,5 / 0,9	✓	✓
EC Plan-Neofluar	40x	1,3	Oil	2,5 / 2,2	<b>1,4 / 1,2</b>	0,8 / 0,7	✓	✓
EC Plan-Neofluar	63x	0,95	Air	<b>1,0 / 1,1</b>	0,6 / 0,7	0,4 / 0,4	✓	✗
EC Plan-Neofluar	63x	1,25	Oil	1,6 / 1,5	<b>0,9 / 0,9</b>	0,5 / 0,5	✓	✓
EC Plan-Neofluar	100x	1,3	Oil	<b>1,0 / 0,9</b>	0,6 / 0,5	0,4 / 0,3	✓	✓
LCI Plan-Neofluar	25x	0,8	Oil, water or glycerin	2,9 / 6,6	1,7 / 3,7	<b>0,9 / 2,0</b>	✓	✓
LCI Plan-Neofluar	63x	1,3	Water or glycerin	1,5 / 1,3	<b>0,9 / 0,7</b>	0,5 / 0,4	✓	✓
Plan-Apochromat	10x	0,45	Air	4,2 / 20,4	2,4 / 11,5	<b>1,3 / 6,2</b>	✓	✓
Plan-Apochromat	20x	0,8	Air	3,2 / 4,9	1,8 / 2,8	<b>1,0 / 1,5</b>	✓	✓
Plan-Apochromat	40x	0,95	Air	1,6 / 1,7	<b>0,9 / 1,0</b>	0,5 / 0,5	✓	✓
Plan-Apochromat	40x	1,3	Oil	2,5 / 2,2	1,4 / 1,2	<b>0,8 / 0,7</b>	✓	✓
Plan-Apochromat	40x	1,4	Oil	2,4 / 1,8	<b>1,4 / 1,0</b>	0,7 / 0,6	✓	✓
Plan-Apochromat	63x	1,4	Oil	1,6 / 1,2	<b>0,9 / 0,7</b>	0,5 / 0,4	✓	✓
Plan-Apochromat	100x	1,4	Oil	<b>1,0 / 0,8</b>	0,6 / 0,5	0,4 / 0,3	✓	✓
LD LCI Plan-Apochromat	25x	0,8	Oil, water or glycerin	2,9 / 6,6	1,7 / 3,7	<b>0,9 / 2,0</b>	✓	✓
C-Apochromat	10x	0,45	Water	4,2 / 20,4	2,4 / 11,5	<b>1,3 / 6,2</b>	✓	✓
C-Apochromat	40x	1,2	Water	2,2 / 2,0	<b>1,2 / 1,1</b>	0,7 / 0,6	✓	✓
C-Apochromat	63x	1,2	Water	1,4 / 1,3	<b>0,8 / 0,7</b>	0,5 / 0,4	✓	✓
LD C-Apochromat	40x	1,1	Water	2,2 / 2,3	<b>1,2 / 1,3</b>	0,7 / 0,7	✓	✓
α Plan-Apochromat	63x	1,46	Oil	1,5 / 1,0	<b>0,9 / 0,6</b>	0,5 / 0,3	✓	✓
α Plan-Fluar	100x	1,45	Oil	<b>1,0 / 0,7</b>	0,6 / 0,4	0,3 / 0,2	✗	✗
α Plan-Apochromat	100x	1,46	Oil	<b>1,0 / 0,7</b>	0,6 / 0,4	0,3 / 0,2	✓	✗

**Axio Observer / Axiovert 200**

Objective	M	NA	Immersion	Grid / Section thickness @ 490nm [RU/μm]			DAPI with FS 34	DAPI with FS 49
				High grid	Middle grid	Low grid		
EC Plan-Neofluar	10x	0,3	Air	2,9 / 31,5	1,7 / 18,5	<b>0,9 / 9,8</b>	✓	✓
EC Plan-Neofluar	20x	0,5	Air	2,3 / 9,0	<b>1,4 / 5,4</b>	0,7 / 2,9	✓	✓
EC Plan-Neofluar	40x	0,75	Air	1,6 / 2,7	<b>0,9 / 1,6</b>	0,5 / 0,9	✓	✗
EC Plan-Neofluar	40x	1,3	Oil	2,4 / 2,1	1,4 / 1,3	<b>0,8 / 0,7</b>	✓	✓
EC Plan-Neofluar	63x	0,95	Air	<b>1,0 / 1,1</b>	0,6 / 0,7	0,4 / 0,4	✓	✓
EC Plan-Neofluar	63x	1,25	Oil	1,6 / 1,5	<b>0,9 / 0,9</b>	0,5 / 0,5	✓	✗
EC Plan-Neofluar	100x	1,3	Oil	<b>1,0 / 0,9</b>	0,6 / 0,6	0,4 / 0,3	✓	✗
LCI Plan-Neofluar	25x	0,8	Oil, water or glycerin	2,9 / 6,5	1,7 / 3,8	<b>0,9 / 2,0</b>	✓	✓
LCI Plan-Neofluar	63x	1,3	Water or glycerin	1,5 / 1,3	<b>0,9 / 0,8</b>	0,5 / 0,4	✗	✗
Plan-Apochromat	10x	0,45	Air	4,2 / 20,2	2,4 / 11,7	<b>1,3 / 6,1</b>	✓	✓
Plan-Apochromat	20x	0,8	Air	3,1 / 4,8	1,8 / 2,8	<b>1,0 / 1,5</b>	✓	✓
Plan-Apochromat	40x	0,95	Air	1,6 / 1,7	<b>0,9 / 1,0</b>	0,5 / 0,5	✓	✓
Plan-Apochromat	40x	1,3	Oil	2,4 / 2,2	1,4 / 1,3	<b>0,8 / 0,7</b>	✓	✓
Plan-Apochromat	40x	1,4	Oil	2,4 / 1,8	<b>1,4 / 1,1</b>	0,7 / 0,6	✓	✓
Plan-Apochromat	63x	1,4	Oil	1,5 / 1,2	<b>0,9 / 0,7</b>	0,5 / 0,4	✓	✓
Plan-Apochromat	100x	1,4	Oil	<b>1,0 / 0,8</b>	0,6 / 0,5	0,4 / 0,3	✓	✗
LD LCI Plan-Apochromat	25x	0,8	Oil, water or glycerin	2,9 / 6,5	1,7 / 3,8	<b>0,9 / 2,0</b>	✓	✓
C-Apochromat	10x	0,45	Water	4,2 / 20,2	2,4 / 11,7	<b>1,3 / 6,1</b>	✓	✓
C-Apochromat	40x	1,2	Water	2,1 / 1,9	<b>1,3 / 1,1</b>	0,7 / 0,6	✓	✓
C-Apochromat	63x	1,2	Water	1,4 / 1,3	<b>0,8 / 0,7</b>	0,5 / 0,4	✓	✓
LD C-Apochromat	40x	1,1	Water	2,1 / 2,3	<b>1,3 / 1,4</b>	0,7 / 0,7	✓	✓
α Plan-Apochromat	63x	1,46	Oil	1,5 / 1,0	<b>0,9 / 0,6</b>	0,5 / 0,3	✓	✓
α Plan-Fluar	100x	1,45	Oil	<b>1,0 / 0,7</b>	0,6 / 0,4	0,3 / 0,2	✗	✗
α Plan-Apochromat	100x	1,46	Oil	<b>1,0 / 0,7</b>	0,6 / 0,4	0,3 / 0,2	✗	✗

**Axio Zoom.V16**

Objective	M	Zoom (total magnification)	Grid / Section thickness @ 490nm [RU/μm]			DAPI with FS 34	DAPI with FS 49
			High grid	Middle grid	Low grid		
Plan-Apochromat	1x	40	1,8 / 82,7	1,22 / 56,7	0,9 / 43,8	✓	✓
Plan-Apochromat	1x	80	1,5 / 25,2	1,0 / 17,4	0,8 / 13,5	✓	✓
Plan-Apochromat	1x	100	1,2 / 19,7	0,9 / 13,7	0,7 / 10,8	✓	✗
Plan-Apochromat	1x	112	1,1 / 17,7	0,8 / 12,4	0,6 / 9,8	✓	✗
Plan-Neofluar	1x	40	1,8 / 82,7	1,22 / 56,7	0,9 / 43,8	✓	✓
Plan-Neofluar	1x	80	1,5 / 25,2	1,0 / 17,4	0,8 / 13,5	✓	✓
Plan-Neofluar	1x	100	1,2 / 19,7	0,9 / 13,7	0,7 / 10,8	✓	✓
Plan-Neofluar	1x	112	1,1 / 17,7	0,8 / 12,4	0,6 / 9,8	✓	✓
Plan-Neofluar	2,3x	40	2,0 / 72,0	1,4 / 49,3	1,1 / 37,9	✓	✓
Plan-Neofluar	2,3x	120 <sup>1</sup>	1,7 / 9,1	1,1 / 6,3	0,9 / 4,9	✓	✓
Plan-Neofluar	2,3x	150	1,6 / 5,9	1,1 / 4,1	0,8 / 3,1	✓	✓
Plan-Neofluar	2,3x	160	1,5 / 5,3	1,0 / 3,6	0,8 / 2,8	✓	✗
Plan-Neofluar	2,3x	250	1,1 / 3,2	0,7 / 2,2	0,6 / 1,8	✗	✗

<sup>1</sup> In ZEN (blue edition) no preset zoom positions



The following filter sets are recommended if ApoTome.2 shall be used with Axio Zoom.V16: 20, 38, 43.



Axio Zoom.V16 allows the user to select also the zoom position, besides the objective. ApoTome.2 may be used with Axio Zoom.V16 with a total magnification of 40x or higher.

**Further filter sets recommended for ApoTome.2**

Order No.	Name	Excitation / Color splitter / Emission	Fluorophores (examples)
488001-9901-000	FS 01	BP 365/12 FT 395 LP 397	Alexa 405, BFP, DAPI
488802-9901-000	FS 02	G 365 FT 395 LP 420	Alexa 350, BFP, DAPI
488009-9901-000	FS 09	BP 450-490 FT 510 LP 515	Alexa 430, Alexa 488, FITC
488010-9901-000	FS 10	BP 450-490 FT 510 BP 515-565	Alexa 430, Alexa 488, FITC, Cy2
488016-9901-000	FS 16	BP 485/20 FT 510 LP 515	Alexa 488, FITC, MitoTracker Green
488017-9901-000	FS 17	BP 485/20 FT 510 BP 515-565	Alexa 488, FITC, MitoTracker Green, Cy2
488020-9901-000	FS 20	BP 546/12 FT 560 BP 575/640	Alexa 555, Rhodamine, dTomato

Order No.	Name	Excitation / Color splitter / Emission	Fluorophores (examples)
488026-9901-000	FS 26	BP 575-625 FT 645 BP 660-710	mPlum, Nile Red
489038-9901-000	FS 38 HE	BP 470/40 FT 495 BP 525/50	Alexa 430, Alexa 488, FITC
489043-9901-000	FS 43 HE	BP 550/25 (HE) FT 570 (HE) BP 605/70 (HE)	Alexa 546, Alexa 555, Cy3, Dil
489046-9901-000	FS 46 HE	BP 500/25 FT 515 BP 535/30	Alexa 488, FITC, eGFP, eYFP
489047-9901-000	FS 47 HE	BP 436/25 FT 455 HE BP 480/40	CFP
488049-9901-000	FS 49	G 365 FT 395 BP 445/50	DAPI
488050-9901-000	FS 50	BP 640/30 FT 660 BP 690/50	Alexa 647, Alexa 660, Cy5
489059-9901-000	FS 59 HE (Colibri)	BP 445/25 (HE) BP 510/15 (HE) DFT 460 + 520 (HE) DBP 480/22 + LP 530 (HE)	Alexa 433, Alexa 488, CFP, FITC, eYFP
489060-9901-000	FS 60 (Colibri)	BP 445/25 (HE) BP 510/15 (HE) BP 588/27 (HE) TFT 460 + 520 + 605 (HE) DBP 480/22 + 546/32 + LP 615	Alexa 430, Alexa 568, Alexa 594
489061-9901-000	FS 61 (Colibri)	BP 474/28 (HE) BP 585/35 (HE) DFT 495 + 605 (HE) DBP 527/54 + 645/60 (HE)	CFP, YFP, mRFP
489062-9901-000	FS 62 (Colibri)	BP 370/40 (HE) BP 474/28 (HE) BP 585/35 (HE) TFT 395 + 495 + 610 (HE) TBP 425 + 527 + LP615 (HE)	DAPI, CFP, YFP

## 2.7 Technical data

### Dimensions (width x depth x height)

ApoTome.2 slider for Axio Imager .....	approx. 278 mm x 90 mm x 76 mm
ApoTome.2 slider for Axio Observer/Axiovert 200 .....	approx. 295 mm x 90 mm x 78 mm
ApoTome.2 slider for Axio Zoom.V16.....	approx. 278 mm x 90 mm x 76 mm
ApoTome.2 control box .....	approx. 255 mm x 220 mm x 96 mm

### Weight

ApoTome.2 slider .....	approx. 1.1 kg
ApoTome.2 control box .....	approx. 2.9 kg

### Ambient conditions

#### Transport (in packing)

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

#### Storage

Permissible ambient temperature .....	+5 to +40 °C
Permissible relative humidity (without condensation).....	max. 75 % at +35 °C

#### Operation

Permissible ambient temperature .....	+5 to +40 °C
Permissible relative humidity .....	max. 75 % at +35 °C
Air pressure .....	800 hPa to 1060 hPa
Operating altitude .....	max. 2000 m
Pollution degree .....	2

### Functional data

Area of use.....	closed rooms
Protection Class.....	I
Degree of protection .....	IP 20
Electrical safety.....	as per DIN EN 61010-1 (IEC 1010-1) and in compliance with CSA and UL regulations
Overvoltage category .....	II
Radio interference suppression.....	as per EN 55011 Class B
Noise immunity .....	as per DIN EN 61326-1
Line voltage.....	100 to 240 V ±10 %
	Line voltage adjustment is not necessary!
Line frequency.....	50 to 60 Hz
Power consumption of ApoTome.2 .....	max. 50 VA

**Fuses as per IEC 127**

ApoTome.2 control box..... T 2 A/H/250 V, 5x20 mm

**Grid frequencies**

Axio Imager slider (transmission grids 1 / 2 / 3) ..... 5 / 9 / 17.5 lp/mm

Axio Observer / Axiovert 200 slider (transmission grids 1 / 2 / 3) ..... 10 / 17.5 / 35 lp/mm

Axio Zoom.V16 slider (transmission grids 1 / 2 / 3) ..... 10 / 15 / 20 lp/mm

---

## 3 STARTUP

The users can install and start up the ApoTome.2 themselves.

On request, however, Zeiss Service will install or convert the ApoTome.2 with costs at the customer's place.



Before installing and starting up the device carefully read the **Notes on device safety**.

### 3.1 Microscope stand requirements

The grid projection method used for the ApoTome.2 is very sensitive to vibrations that may originate from different sources. Vibrations are visible in the result image as streak artifacts. Therefore, the microscope should be installed as free from vibrations as possible on an anti-vibration table or a suitable microscope support.

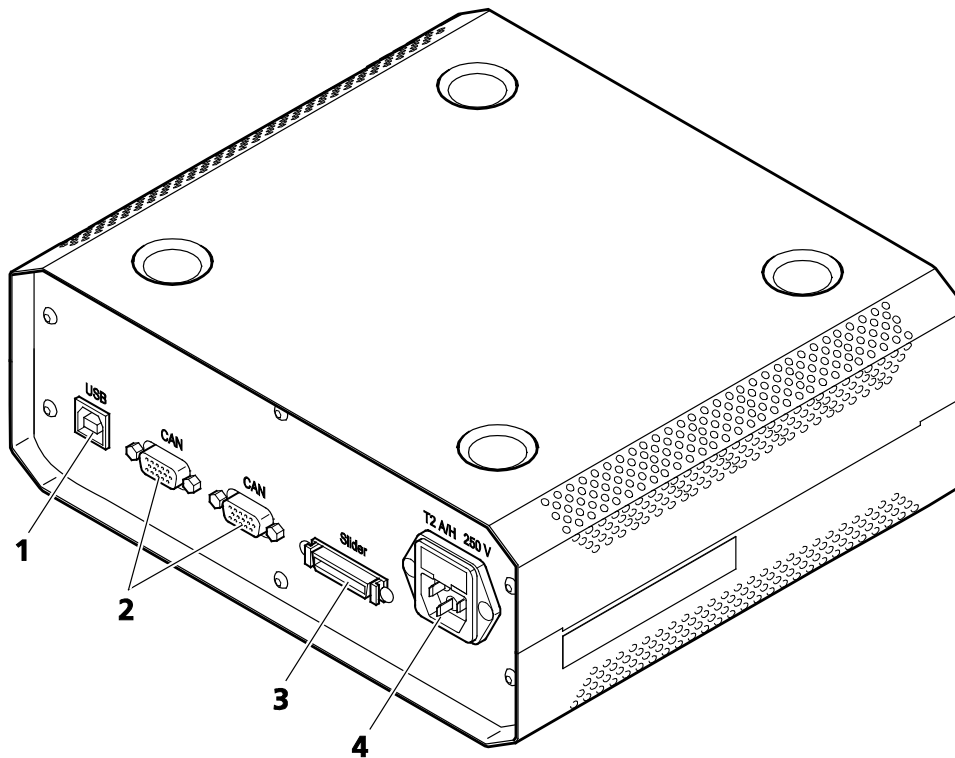
### 3.2 Unpacking ApoTome.2

The ApoTome.2 slider is delivered in a polyethylene box with outer carton package. The ApoTome.2 control box is delivered in an outer carton package.

Besides the slider, the box contains the accessories required for the calibration of ApoTome.2:

- Reflector module Calibration ACR P&C
  - Fluorescence test specimen for grid focus calibration
  - Mirror specimen with cross-line graticule for phase calibration
- 
- Remove all units from the packaging and verify that all components listed on the delivery note are present.
  - Keep the original packaging for a possible longer storage or return of the instrument to the manufacturer or dispose of it properly.

### 3.3 Connecting the control box




- |   |               |   |  |
|---|---------------|---|--|
| 1 | USB port      | 3 | Connector for ApoTome.2 slider ( <b>Slider</b> ) |
| 2 | CAN port (2x) | 4 | Mains connector                                  |


**Fig. 3-1 Connectors on control box (rear panel)**

- Plug the connection cable ApoTome.2 to the power connector **Slider** (3-1/3) at the rear side of the control box.
- Connect the mains connector (3-1/4) of the control box to a power socket via the mains cable.

The control box can be connected in two different ways:

- Via USB cable directly at the PC:
  - Use a USB cable to connect the USB port (3-1/1) on the rear panel of the control box with a free USB port at the PC used.
- Via CAN BUS cable to a free CAN BUS port on the microscope stand:
  - Use the CAN BUS cable (457411-9011-000) to connect one of the CAN ports (3-1/2) at the rear panel of the control box with a free CAN BUS connector on the microscope stand used.

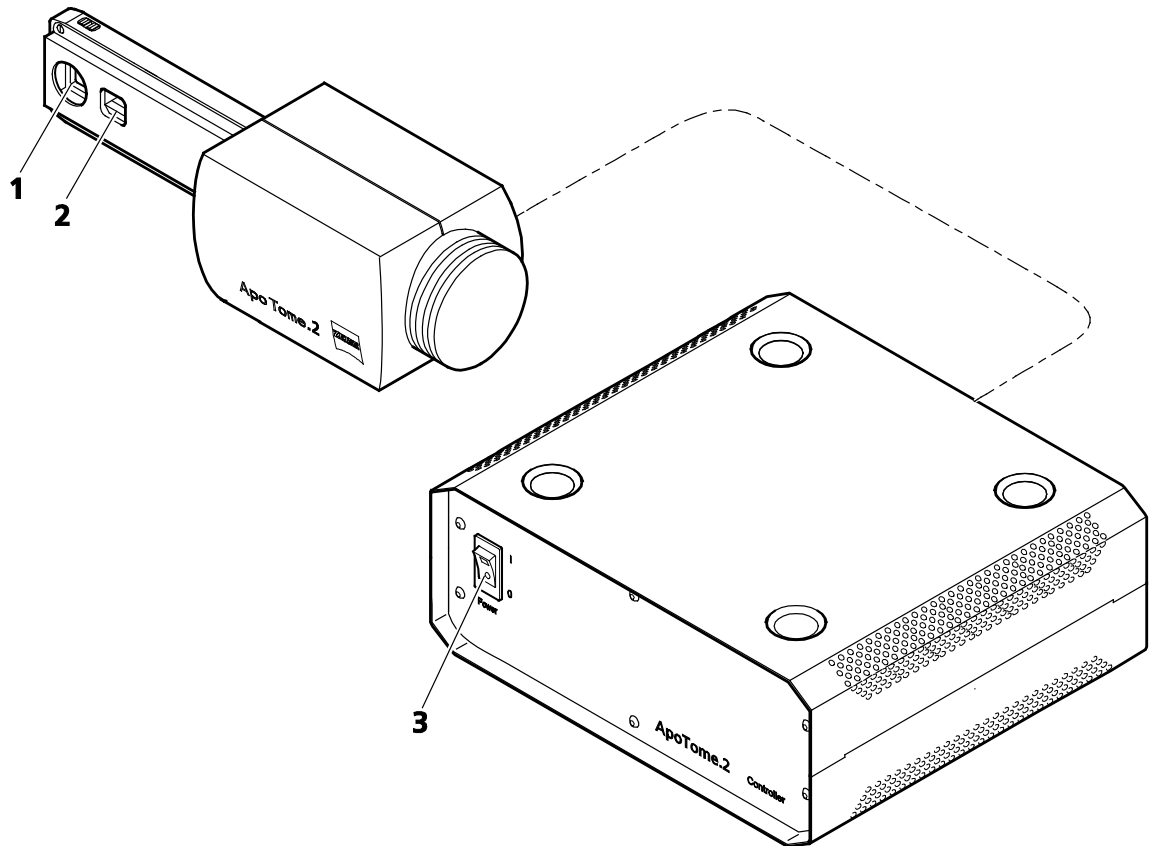
 If an Axiovert 200 is used, the control box can only be connected to the PC via a USB cable. A connection via CAN BUS is not possible!

 Please refer to the Operating Manual AxioVision Rel. 4.8.2 / ZEN 2011 (blue edition) for more detailed information about the configuration.



## 4 OPERATION

### 4.1 Control and functional elements of ApoTome.2



- 1 Click stop position 1 - free passage
- 2 Click-stop position 2 - transmission grid
- 3 On/off switch

**Fig. 4-1** Control and functional elements of ApoTome.2

## 4.2 Switching ApoTome.2 on/off

### 4.2.1 Switching on

- Use the on/off switch (4-1/3) at the front panel of the ApoTome.2 control box to switch the device on.
- The power indicator in the switch lights up.



Switch on the device approximately 15 minutes before the first image capture in order to allow it to warm up to the necessary operating temperature.

The slider must be inserted in the microscope stand to warm up properly.

### 4.2.2 Switching off

- After use, switch the device off by activating the on/off switch (4-1/3) at the front panel of the ApoTome.2 control box.
- The power indicator in the switch turns off.

## 4.3 Operating the slider

### 4.3.1 Inserting the slider

Carefully insert the ApoTome.2 slider in the plane of the luminous-field diaphragm of the reflected light beam path of the microscope used. On insertion, a short beep signal is generated. In the first click-stop position, conventional imaging is possible (free passage). In the second click-stop position, the transmission grid is moved into the beam path.

### 4.3.2 Click-stop position 1

In click-stop position 1 of the ApoTome.2 slider, an open passage is in the reflected light beam path. In this position the microscope can be operated in the conventional way. The slider does not affect any function of the microscope stand used.

### 4.3.3 Click-stop position 2

In click-stop position 2, the transmission grid is positioned in the beam path of rays. All functions for the adjustment and control of the instrument are operated via the software. For additional information, refer to the Operating Manual of the software used.

## 5 CARE, MAINTENANCE, TROUBLESHOOTING AND SERVICE

### 5.1 Cleaning the device



Before starting cleaning works, switch off the device and disconnect it from the mains.



Ensure that no moisture enters the device.

The care of the ApoTome.2 is limited to the works described below:

- After every use, switch off the device and cover it with the protective cover (protection from dust and moisture). If necessary, store the ApoTome.2 slider in the provided case.
- Do not install the device in a damp room; the maximum permissible humidity is 75 %.
- Remove dust and loose contamination from visible optical surfaces with a fine brush, rubber blower, cotton bud, optics-cleaning tissue or a dust-free cotton cloth without exerting pressure.
- Wipe off water-soluble contamination (coffee, cola, etc.) after breathing by using a dust-free cotton cloth or a moistened cloth. The water used may contain a dash of a mild detergent.
- Wipe off stronger oily or fatty contamination (immersion oils, fingerprints) with a cotton bud, lens cleaning paper or a dust-free cotton cloth using optics cleaning agent L.  
This cleaning agent is made of 90 percent by volume gasoline and 10 percent by volume isopropanol (IPA). Its individual constituents are also known under the following synonyms:  
gasoline: surgical spirits, petroleum ether  
isopropanol: 2-propanol,  
dimethyl carbinol,  
2-hydroxypropane

Clean the optical surfaces parallel to the alignment of the grid exerting a slight pressure onto the surface.



Clean the transmission grid with extreme care to avoid the damage of the grid structures!

For the use of the ApoTome.2 in humid climatic zones, observe the following instructions:

- Store the device in a bright, dry and well ventilated room; humidity < 75 %. Store components and accessories that are especially susceptible to humidity, such as objectives and eyepieces, in drying chambers.

Precision-mechanical optical instruments are always endangered by mold attack under the following conditions:

- relative humidity of > 75% longer than three days at temperatures of +15 °C to +35 °C,
- installation in dark rooms without movement of air,
- dust deposits and fingerprints on optical surfaces.

## 5.2 Maintaining the device

### 5.2.1 Performing checks

- Make sure that the available line voltage corresponds to the line voltage values as labeled on the ApoTome.2 control box.
- Check power cable and power plug for damage.
- In the event of perceptible damages, switch off the instrument and safeguard it against inadvertent use. Have damages repaired by appropriately qualified staff.

### 5.2.2 Replacing fuses



Before replacing fuses, make sure to disconnect the power cable.

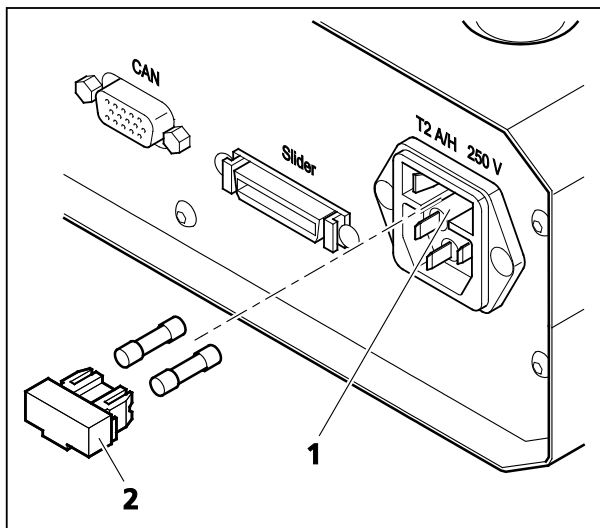


Fig. 5-1 Replacing fuses

The fuse compartment is located at the rear panel of the control box. It is combined with the appliance connector and contains two fuses of the type **T 2 A/H 250 V**.

- Disconnect the power cable.
- Pull out the fuse holder (5-1/2) forward. For doing this, use a small screwdriver, if necessary.
- Remove the defective fuses from the holder and replace them by new fuses.
- Slide the fuse holder back into the fuse compartment (5-1/1) as far as it will go.
- Connect the power cable.

### 5.3 Troubleshooting

Problem	Cause	Troubleshooting
Device does not respond to any control command	no current	Actuate power switch, connect power cable to power outlet.
	no connection to the PC	Establish CAN BUS cable connection to the stand or the USB cable connection to the PC.
	no connection between slider and control box	Plug the cable of the slider into the respective connector on the control box.
	device defect	Call Technical Service.
No transmission grid visible in eyepiece although the slider is in the ApoTome position (click position 2)	transmission grid not focused	To obtain an artifact-free result image, the transmission grid must be imaged with a high quality in the specimen plane. For this purpose, calibrate the system as described in the Operating Manual of the imaging software AxioVision 4.8.2 / ZEN (blue edition).

### 5.4 Wear parts and tools

Designation	Catalogue no.	Purpose
Fuse type <b>T 2 A/H 250 V</b>	000000-0149-688	Function of control box
Reflector module calibration "ApoTome" ACR P&C	424930-9902-000	Calibration of phase position of scanner
Mirror test specimen	000000-1182-440	Calibration of phase position of scanner
Fluorescence test specimen	000000-1213-943	Calibration of the focus position of the transmission grid for various wavelengths
Cover plate	423666-0102-000	Replacement of the original cover plate on Axio Imager

## 5.5 Requesting service

All repairs of mechanical, optical or electronic components inside the instrument and of the electrical components of the ApoTome.2 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 the Carl Zeiss representative responsible for your region.

For additional information, contact us at:

[micro@zeiss.de](mailto:micro@zeiss.de)

or visit us in the Internet at:

<http://www.zeiss.de>

**6 APPENDIX****6.1 List of abbreviations**

CE	Conformité Européenne (Conformity with EC directives)
CSA	Canadian Standards Association
DIN	Deutsches Institut für Normung (German Institute for Standardization)
EC	European Community
EN	European standard
HBO	Mercury vapor short-arc lamp for fluorescence
IEC	International Electrotechnical Commission
IP	International Protection
ISO	International Organization for Standardization
T	time-delay (a type of fuses)
UL	Underwriter Laboratories (USA audit authority)
UV	ultraviolet

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