MIRAX MIDI BF and BF/FL
Digitizer for 12 Slides (V1.12)
Operating Manual

Prior knowledge of this manual is essential for proper MIRAX MIDI operation. You are therefore advised to familiarize yourself with its contents and, equally importantly, to follow special notes and instructions it contains regarding the safe product handling.

We reserve the right to make changes as may be deemed appropriate in the interest of ongoing technical improvement. This manual is not subject to updating or revision services of any kind.

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MIRAX MIDI has been developed, manufactured and licensed exclusively for Zeiss by 3DHISTECH Kft./Hungary.

Further information

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1 INTRODUCTION

1.1 Notes regarding Operational Safety

MIRAX MIDI has been designed, manufactured and tested to comply with the standard requirements of DIN EN 61010-1 (IEC 61010-1) and IEC 61010-2-101, the safety provisions for electrical equipment for measurement, control and laboratory use, with due consideration of applicable requirements under Exhibit 1 of Directive 98/79/EC.

Conformity with Directive 98/79/EC referenced above is documented by the CE label.

This operating manual contains information and warnings that must be followed by owner/operator personnel.

Warning and advisory notes, which are used throughout this operating manual, have the following meanings:

NOTE
This symbol designates a note that should be closely followed.

CAUTION
This symbol designates a potential hazard to the device.

CAUTION
This symbol designates a potential hazard to the user.

CAUTION
Hot surface!

CAUTION
Emergence of UV radiation!

CAUTION
Disconnect mains plug before any kind of intervention into the device!

CAUTION
Crush hazard!

CAUTION
Keep combustible and highly inflammable materials outside the range of the HXP 120 illuminator!

The MIRAX MIDI, including any of its original accessory components, should not be used in any other way than as described in section "Intended Use" of this operating manual.

Any of the following notes should be strictly observed:

In the event of any non-conforming type of use, including non-conforming use of assemblies or single parts, no liability will be assumed by the Manufacturer. This shall also apply to service or repair work of any kind, which is performed by other than duly authorized service personnel. All claims for warranty and warranty services will be null and void in any such case.

The operation of the instrument in explosion-risk environments is not allowed.

Insert the power plugs only into a grounding outlet. The protective action must not be rendered ineffective by extension cables without protective conductor.

If protective devices are found to be ineffective, the device must be shut down and precautions must be taken to prevent unintended usage. For necessary repair work or work to restore proper operating condition, you should contact the Customer Service of Zeiss or the Carl Zeiss Microscopy Service.

Electric power to the MIRAX MIDI is supplied via a special power supply unit. It allows for operation at line voltages in the range of 100 V to 240 V ±10%, 50/60 Hz, without any extra facilities required for voltage transformation.

Check that your local line voltage is compatible with the device specifications before you turn power on.

Remember to pull the power plug before you open the device or replace a fuse!

Use only electric fuses which are rated for the specified nominal current. You are forbidden to use makeshift fuses or short-circuit the fuse holders.

The MIRAX MIDI does not include any special facilities to protect from samples with an etching, potentially infectious, toxic, radioactive or other health-damaging effect. You are under obligation to comply with all legal requirements, notably, national accident prevention rules when handling samples of this kind.

Dirt or dust may adversely impact the device's operating capability. For this reason, measures must be taken to eliminate such influences to the maximum possible degree. During periods of non-use, the device must be protected with a dust cover. Check that power is turned off before you put the cover on.

Avoid touching the lamp bulb. Disconnect power and let the lamp cool down for about 15 minutes before replacing the lamp.

There is crush hazard in the movement area of the slide insert during operation. Please note that the digitizing process has to be interrupted and the insert moved via software to the output position before removing the insert.

Clogged or covered ventilation slots may result in a heat build-up with a damaging effect for the device. In extreme cases, this may also cause fire. Keep the ventilation slots always unobstructed and refrain from inserting or dropping objects into a ventilation slot.

Do not employ the camera adapter or the camera to lift, pull or move the instrument.

A defective device does not classify as domestic waste. It must be properly disposed in accordance with currently valid legal requirements.

The MIRAX MIDI may not be operated by other than duly instructed persons. Operating personnel must be fully aware of the potential dangers which a particular field of application involves. This device is a precision instrument which may suffer a significant reduction in operational functions or even physical destruction following intervention performed in any con-conforming manner.

Keep combustible and highly inflammable materials outside the range of the HXP 120 illuminator!

For operating the UV radiation source HXP 120 observe in any case the health and safety at work provisions. The national legal regulations have to be complied with as well.

- Follow also the operating instructions for the HXP 120 illuminator provided by the Manufacturer.
- Do not expose yourself to the UV radiation of the HXP 120 illuminator.
- Never look directly into the light guide, when the HXP 120 illuminator is switched on. **In case of non-observance your eyes may be damaged.**
- Never remove the light guide from the MIRAX MIDI device when the HXP 120 illuminator is switched on.
- Never connect the light guide to the MIRAX MIDI device when the HXP 120 illuminator is switched on.
- Never remove the housing from the MIRAX MIDI device when the HXP 120 illuminator is connected and switched on.

If an X-Cite 120 illuminator is used, the corresponding warnings included in the manufacturer's operating manual have to be observed.

Carl Zeiss works according to a certified Environmental Management System in compliance with the ISO 14001 international standard. The product has been developed, inspected and manufactured in accordance with the applicable regulations and guidelines of the environmental law of the European Union.

The product and its accessories meet the requirements of the EU Directives 2002/95/EC (RoHS) and 2002/96/EC (WEEE), as far as they are applicable to this product. The product and its accessories were classified as category 8 (medical products).

Carl Zeiss introduced a procedure for taking back and recycling the devices within the member states of the European Union, ensuring the appropriate utilization in accordance with the EU Directives.

For more detailed information about disposal and recycling please contact your competent selling or service organization of Carl Zeiss.

The product must not be disposed of as domestic waste or through municipal waste streams. If resold, the seller must clearly inform the buyer of the product to be disposed of according to the regulations mentioned.

1.2 Notes on Warranty

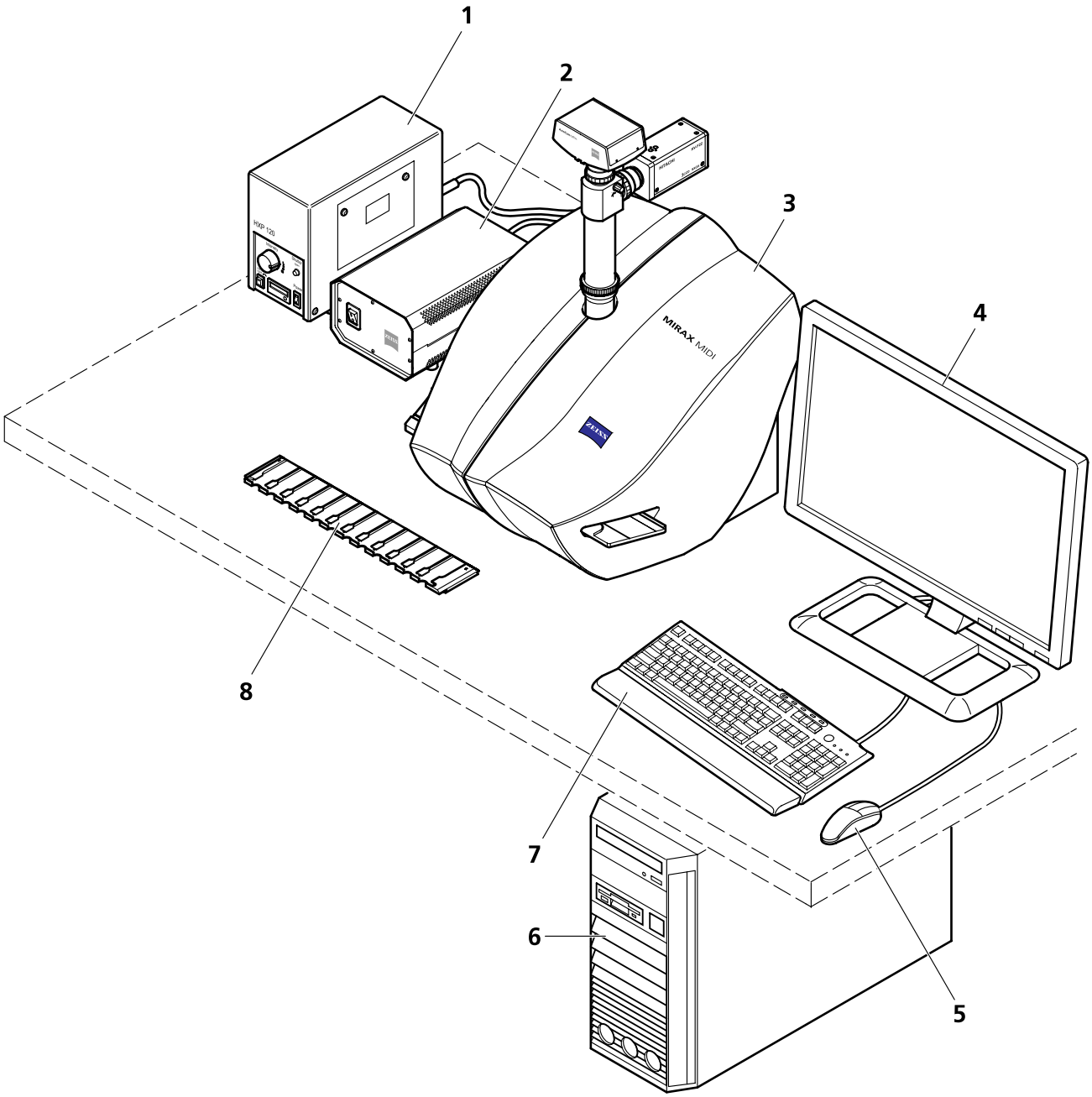
The Manufacturer warrants this device to be free from faults in material and workmanship at the moment of passing into the user's ownership. Defects must be notified immediately on identification and maximum efforts must be undertaken to keep the amount of damage as small as possible. On receiving notice of a fault, the Manufacturer will be under obligation to remove the fault, at his own option, either by providing adequate repair services or by delivering a faultless product for replacement. No warranty will be accepted in cases of natural wear (notably, wear and tear parts) or improper product handling.

The Manufacturer will refuse any liability in the event of damage resulting from faulty operation, negligence or unauthorized intervention into the product, notably, from the removal or replacement of product parts or the usage of accessory parts from other manufacturers. Any such case will void the user's claim for warranty.

Except for action which is explicitly described in this operating manual, you are forbidden to perform any kind of maintenance or repair work on the MIRAX MIDI. Only Zeiss Customer Service personnel or specifically authorized representatives are allowed to handle repair work. On identifying a situation of malfunction, you should, first of all, contact the appropriate Carl Zeiss Representative Office.

2 PRODUCT DESCRIPTION

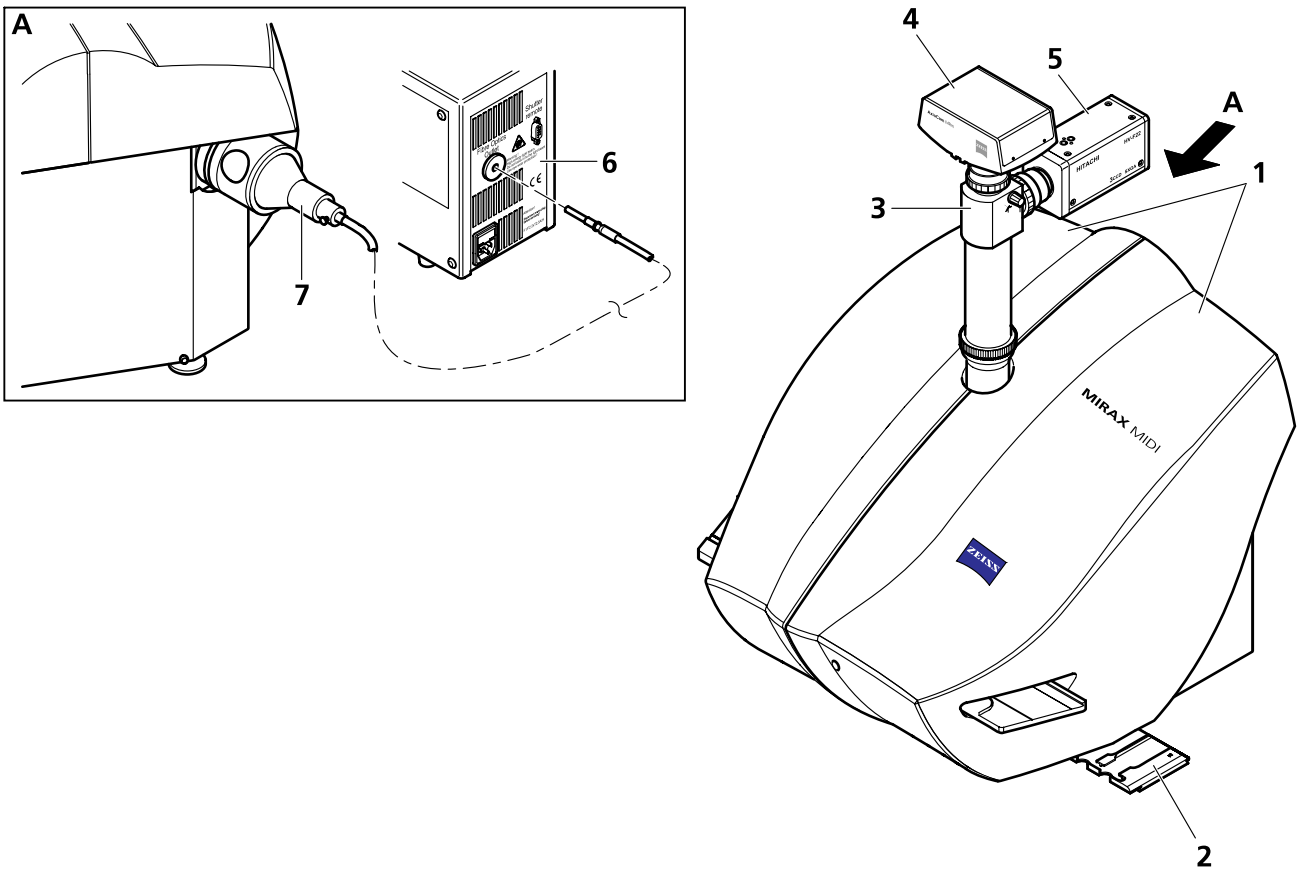
2.1 MIRAX MIDI, General View



1 HXP 120 fiber-coupled illuminator (for MIRAX MIDI BF/FL)
2 Power supply unit (24 V)
3 MIRAX MIDI base unit
4 30" TFT monitor
5 Mouse
6 Image Analysis XEON Workstation
7 Keyboard
8 Slide insert

Fig. 2-1 MIRAX MIDI general view

2.2 MIRAX MIDI Base Unit



1 Protective cover of scanner (made in two parts)
2 Slide insert
3 Double camera adapter (option)
4 Scan camera 1 (in this case: AxioCam MRm)
5 Scan camera 2 (in this case: Hitachi HV-F22 CL)
6 HXP 120 fiber-coupled illuminator (for MIRAX MIDI BF/FL)
7 HXP 120 illumination adapter (for MIRAX MIDI BF/FL)

Fig. 2-2 MIRAX MIDI base unit

2.3 Product Designation and Intended Use

Manufacturer's designation: MIRAX MIDI

The MIRAX MIDI is intended for digitization of biological specimens and samples (above all histological sections) of human or animal bodies on smooth slides in transmitted light mode (MIRAX MIDI BF) and reflected light fluorescence mode (MIRAX MIDI BF/FL).

Typical applications are (but not limited to):

- Hospitals
- Medical practices
- Universities and colleges
- Industry
- Research institutes

2.4 Product Description and Main Features

Essential features of the **MIRAX MIDI** are:

- Robust and stable base unit
- Intuitive operation control via keyboard and mouse
- Insert with a maximum fill capacity of 12 slides
- Option for use of two cameras
- HAL 12 V, 5 W illuminator for transmitted light operation
- Reflected light fluorescence (MIRAX MIDI BF/FL)
- Optional Optovars for flexible, user-selected adaptation of magnification scale (MIRAX MIDI BF/FL)
- High-resolution scanner imaging optics
- Automatic feeding and output of slides
- Automatic identification of slides through barcode recognition
- Automatic sample recognition and digitization of sample detected on slide

2.5 Warning and Information Labels

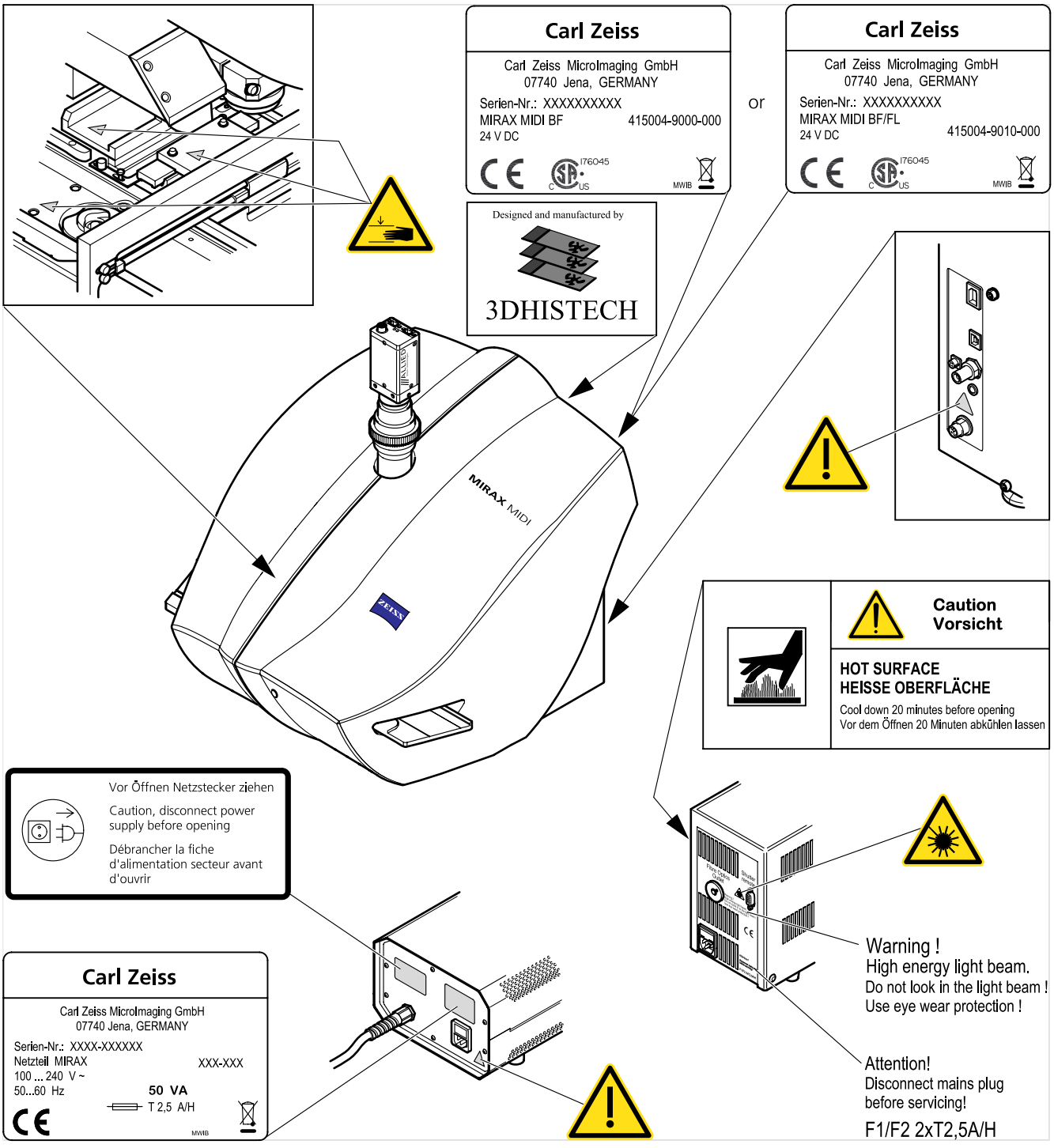
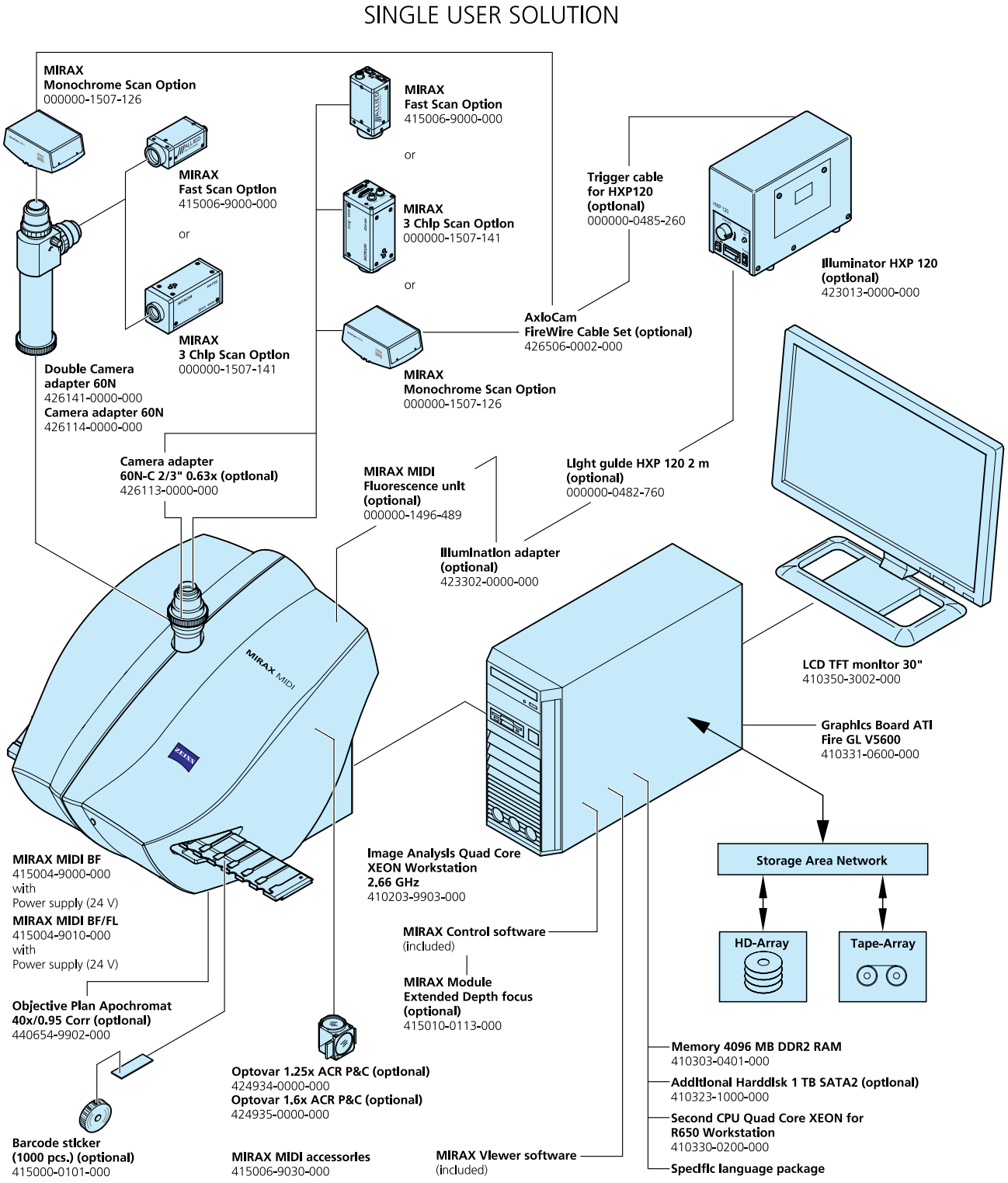
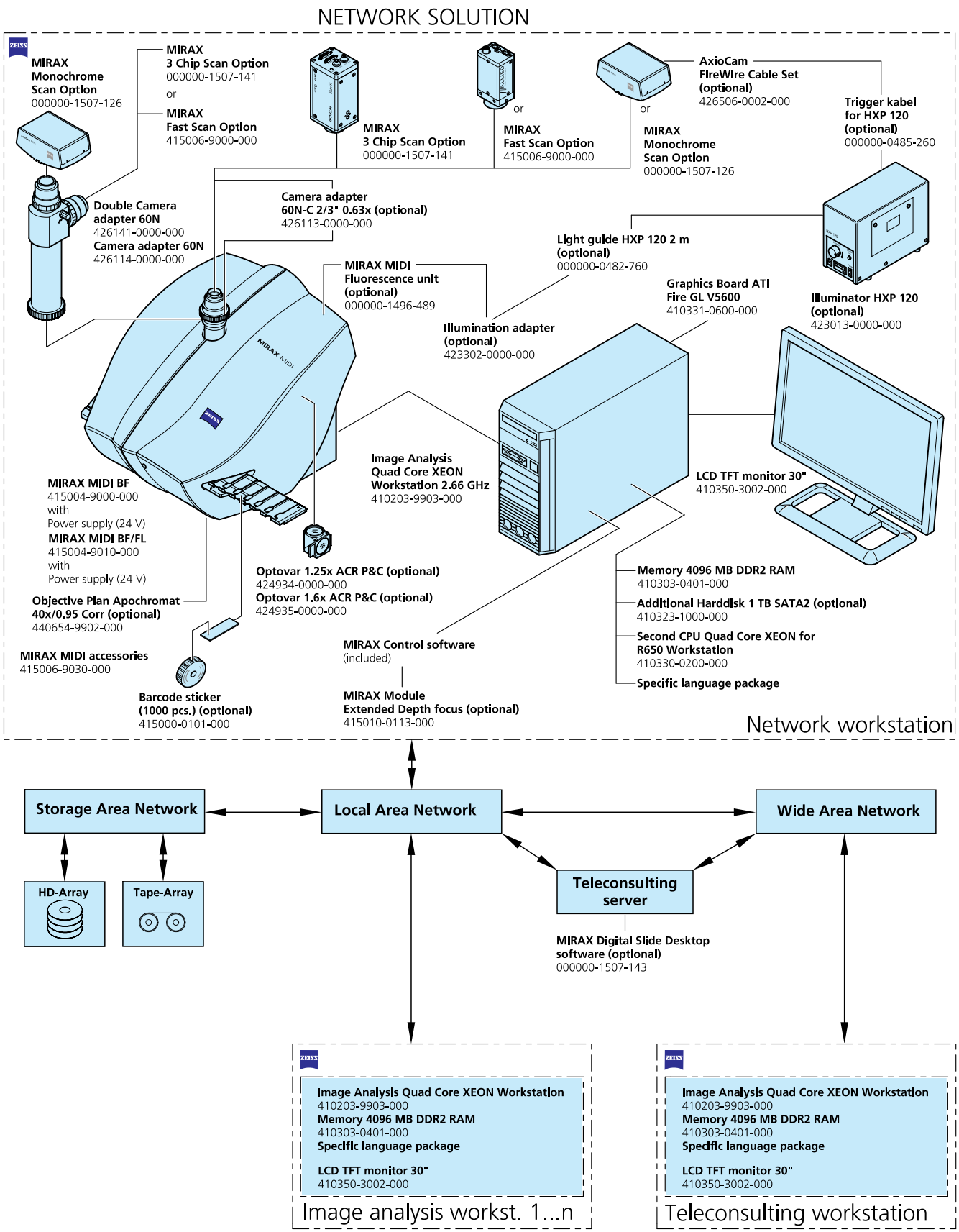


Fig. 2-3 Warning and information labels

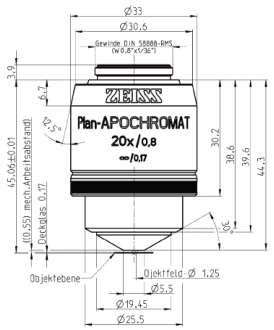
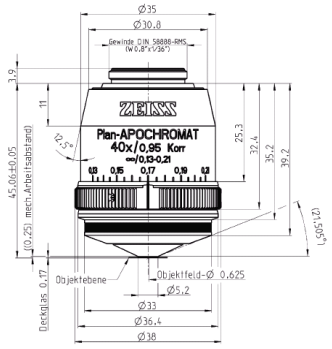
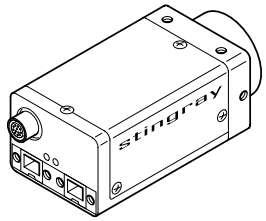
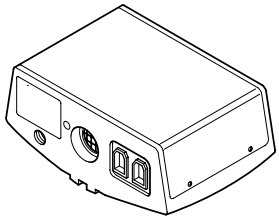
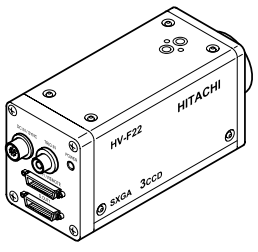
2.6 MIRAX MIDI System Overview for Single User Solution

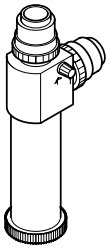


2.7 System Overview for Network Solution

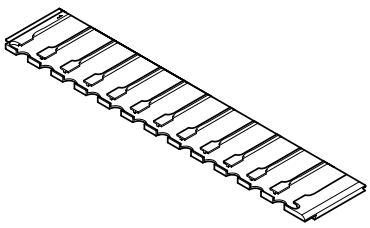


2.8 Objective Lenses, Cameras and Adapter

View/Drawing	Name and technical specifications
	In standard delivery version, the MIRAX MIDI is configured with a 20x / 0.8 Plan-Apochromat objective.
	40x/0.95 Corr Plan-Apochromat objective (optionally available)
	AVT Stingray F-146C FireWire (MIRAX Fast Scan option)
	AxioCam MRm Rev. 3 (MIRAX Monochrome Scan option)
	Hitachi HV F22CL (MIRAX 3Chip Scan option)

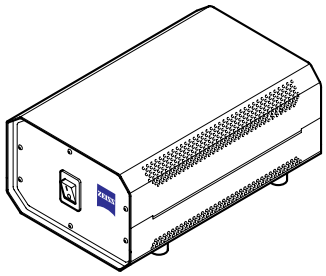
	Double camera adapter (option) for use of two cameras
Without illustration	Preview camera with white backlight LED illumination and reflected light illumination
Without illustration	The MIRAX MIDI comes with a C-mount adapter 1x in standard delivery version.
Without illustration	C-mount adapter 0.63x (optional)
Without illustration	Optovar 1.25x ACR P&C (optional, for transmitted light only and only for MIRAX MIDI BF/FL)
Without illustration	Optovar 1.6x ACR P&C (optional, for transmitted light only and only for MIRAX MIDI BF/FL)

2.9 Accessory Components

Graphical view	Name and technical specifications	Ordering code
	Insert for 12 slides	000000-1507-124
	Barcode stickers (1000 pieces)	415000-0101-000

☞ The MIRAX MIDI may not be operated with other than original slide inserts.

2.10 Power Supply Unit and Light Source

Graphical view	Description and technical specifications	Ordering code
	Power supply unit (24 V)	-
	HAL 12 V, 5 W for standard configuration, transmitted-light mode	000000-0467-436

2.11 Accessories and Spare Parts for MIRAX MIDI BF/FL

Graphical view	Name and technical specifications	Ordering code
Without illustration	Calibration slide for fluorescence mode	415006-9010-000
Without illustration	Light guide HXP 120 with liquid fill, 2 m	000000-0482-760
Without illustration	Trigger cable for HXP 120 illuminator (only required if AxioCam MRm is used)	000000-0485-260
Without illustration	AxioCam FireWire HS/MRc5/HR/MR-Rev. 3 Trigger Cable Set T/R (D)	426506-0002-000
Without illustration	HXP 120 lamp module, replacement	000000-0482-730
Without illustration	Reflector module FL EC P&C (without filter sets)	424931-0000-000

2.12 Software Option

Graphical view	Name and technical specifications	Ordering code
Without illustration	MIRAX scanning module, extended depth of sharpness	415010-0113-000

2.13 Technical Data

Dimensions (width x depth x height)

MIRAX MIDI base unit ..... approx. 1000 mm x 600 mm x 600 mm
Control computer ..... approx. 206 mm x 560 mm x 440 mm
Monitor (19") ..... approx. 420 mm x 240 mm x 490 mm
Power supply unit (24 V) ..... approx. 170 mm x 300 mm x 110 mm

Weight

MIRAX MIDI BF (brightfield) ..... approx. 23 kg
MIRAX MIDI BF/FL (brightfield and fluorescence) ..... approx. 26 kg
Power supply unit (24 V) ..... approx. 2 kg

Ambient conditions

Transportation/shipment (packed)

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

Storage

Permissible ambient temperature ..... +10 °C to +40 °C
Permissible relative air humidity (non-condensing) ..... max. 75 % at 35 °C

Operation

Permissible ambient temperature ..... +10 °C to +40 °C
Permissible relative air humidity ..... max. 75 % at 35 °C
Altitude of place of installation ..... max. 2000 m
Air pressure ..... 500 hPa to 1060 hPa
Degree of pollution 2

Operating data

Category of operating environment ..... closed room facilities
Protection class I
Degree of protection ..... IP 20
Electrical safety ..... under DIN EN 61010-1 (IEC 61010-1) and taking CSA and UL regulations into account
Over-voltage category II
Radio interference suppression ..... under EN 55011 class B
Noise immunity ..... under DIN EN 61326
Base unit input voltage ..... 24 V DC ±1 V
Power consumption of the complete device (PC, monitor and power supply unit MIRAX) ..... 270 VA

Power supply unit (24 V)

Category of operating environment ..... closed room facilities
Protection class I
Degree of protection ..... IP 20
Input voltage ..... 100 to 240 V AC
Input frequency ..... 50/60 Hz
Power consumption ..... max. 50 VA
Secondary voltage ..... 24 V DC
Secondary current ..... max. 3 A
Fusing ..... 2 x T 2,5 A/H, 250 V, 5 x 20 mm

Light sources

Halogen lamp (transmitted light operation): ..... HAL 12 V, 5 W
Average lifetime performance of halogen lamp (000000-0467-436) ..... 4000 h
Lamp module for HXP 120 (for MIRAX MIDI BF/FL) ..... 120 W
Mercury short arc reflector lamp:
Average lifetime performance of lamp module (000000-1313-162) ..... 2000 h

The present manual includes the description of the fluorescence compact light source HXP 120. Besides the HXP 120, the X-Cite 120 may be used with the MIRAX MIDI!

Software

MIRAX MIDI operation control ..... included in MIRAX MIDI delivery scope
MIRAX Viewer ..... freeware
MIRAX scanning module, extended depth of sharpness (optional) 415010-0113-000

MIRAX MIDI standard configuration

Objective lens ..... Plan-Apochromat 20x/0.8
Inserts ..... 2 inserts for 12 slides each
Camera ..... AVT Stingray F-146C, AxioCam MRm or Hitachi HV-F22 CL
Camera adapter ..... C-mount adapter 1x

Identification of slides:

Slides are identified by preview camera (TIS DFK21F04): Barcode area is saved as a picture and barcode label is interpreted as a character string, with capability for interpretation of the following types of 1D and 2D barcodes:

- Code39 (ASCII encoding)
- Code93 (ASCII encoding)
- Code128 (UCC/EAN128) (ASCII encoding)
- Code Interleaved 2of5
- EAN8 (Numeric encoding)
- EAN13 (Numeric encoding)
- UPC-A (Numeric encoding)

- DataMatrix (Numeric encoding, Alpha encoding, AlphaNumericPunc encoding, AlphaNumeric encoding, ASCII encoding, IS08 encoding)
- Maxicode (Encoding mode 2, 3, 4, 5, 6)
- MicroPDF417 (Standard encoding type)
- PDF417 (Standard encoding type)
- QR (QR code Model 1, 2 encoding)

Image digitization

AVT Stingray F-146C FireWire
1/2" type progressive SCAN IT CCD
Pixel size ..... 4.65 µm x 4.65 µm
Pixel resolution with 20x objective and C-mount adapter 1x (interpolated) ..... 0.23 µm

Hitachi HV-F22 CL
3x 1/2" type CCD
Pixel size ..... 4,65 µm x 4,65 µm
Pixel resolution with 20x objective and C-mount adapter 1x ..... 0.23 µm

AxioCam MRm Rev. 3
2/3" type progressive SCAN IT CCD
Pixel size ..... 6.45 µm x 6.45 µm
Pixel resolution with 20x objective and C-mount adapter 1x ..... 0.32 µm
Cooling ..... Peltier element

Control computer and required system resources (minimum)

CPU ..... 2 x Intel 2.6 GHz
RAM ..... 3 GB
Hard disk ..... 130 GB
Ports ..... 2 x FireWire
1 x USB 2.0
GB Ethernet
Input units ..... keyboard, mouse

3 START UP

3.1 Initial Start Up

The base unit is shipped in a commercial polyethylene container with outer carton packaging as is customary in the trade. The base unit is firmly screwed to a wooden board.

This base unit set includes two slide inserts, slides, barcode stickers, scan camera, replacement lamps, tooling, and 1x C-mount adapter, as well as cables (2x FireWire, 1x USB). The control computer and monitor come in separate outer carton packages!

If the MIRAX 3Chip Scan option is used, two CameraLink cables and one power supply cable are delivered instead of a FireWire cable.

The MIRAX MIDI classifies as a product requiring installation by Manufacturer personnel! For a proper operation of the device, installation and initial start up must be performed by Zeiss Service technicians.

You should dispose of the original packing in accordance with local regulations or keep it for later storage or reshipment of the product to the Manufacturer.

When choosing the place of installation you should take into consideration that the slide insert will be moved beyond the basic measures of the base unit during operation (50 cm on both sides).

3.2 MIRAX MIDI Transportation

- The user should move the MIRAX MIDI only over short distances.
- The device can be transported safely only by two persons.
- Care must be taken to ensure that the scan camera is not moved during the transport.

- Turn power to the MIRAX MIDI and its peripherals off and detach all cables to external instruments.
- Remove the slide insert.
- Lift the device from below holding it by its base plate (not by the protective covers).
- Once the device is at its designated installation place, establish the cable connections again.

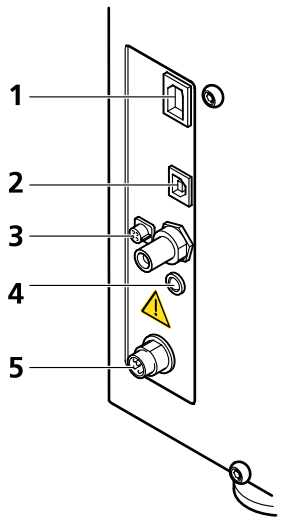


Fig. 3-1 Terminals at base unit

3.3 Ports and Operation Control Elements

3.3.1 Base Unit

The connector back panel provides the terminal point for the power supply cable (+24 V DC) (3-1/**5**) to link the device with the 24 V power supply unit, a ready-for-operation LED indicator (3-1/**4**), and a port for the trigger cable (3-1/**3**) for scan camera operation (for Sony camera only).

The MIRAX MIDI further provides a terminal for its computer connection via USB control cable (3-1/**2**) and a FireWire terminal for connecting the preview camera (3-1/**1**) with the control computer.

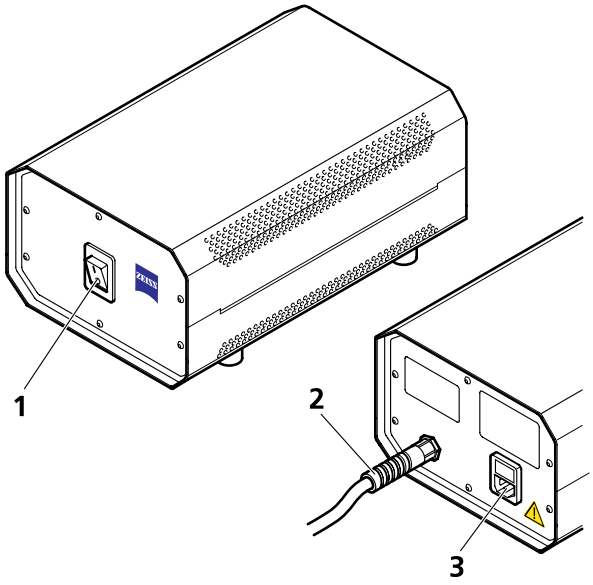


Fig. 3-2 Terminals at 24 V power supply unit

3.3.2 Power Supply Unit (24 V)

The power supply unit is designed to supply the base unit with direct voltage of 24 V.

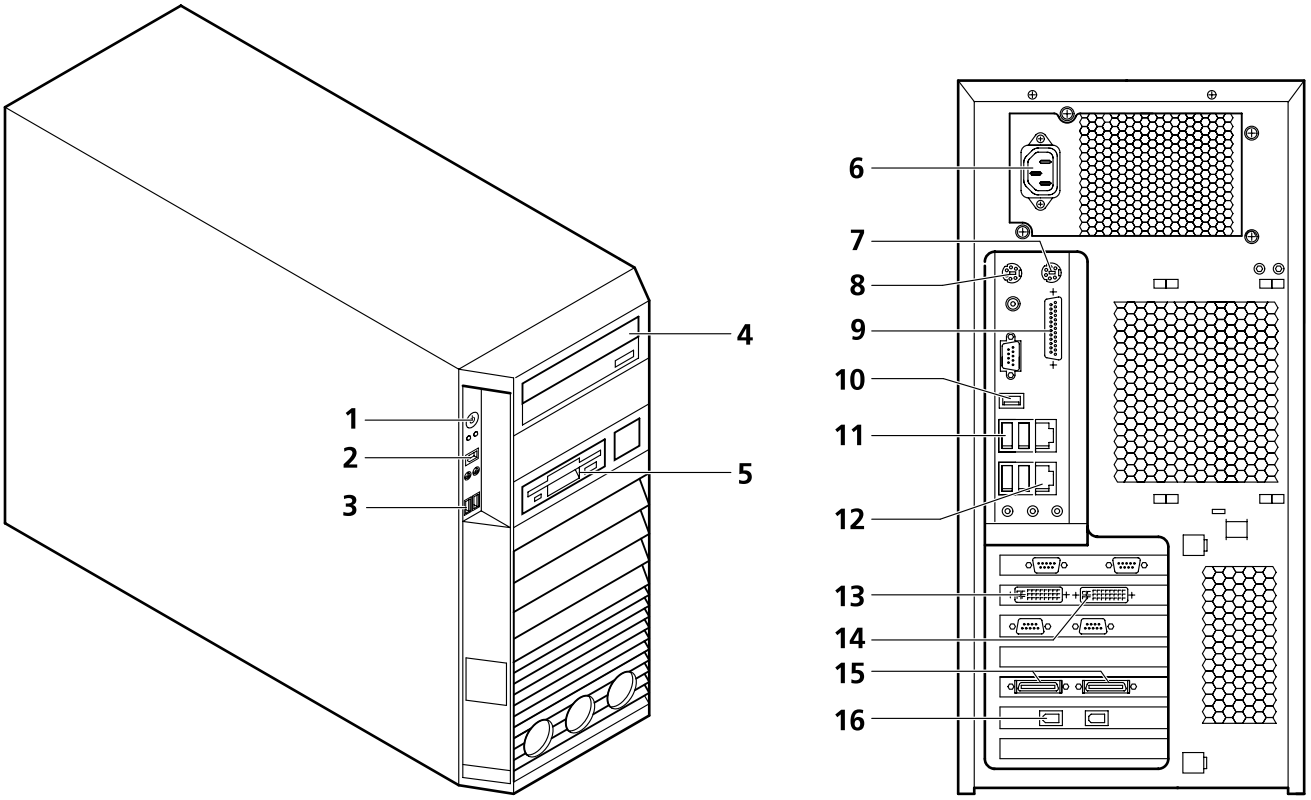
Located at its front is the power switch (3-2/**1**).

Its back panel contains the line power inlet (3-2/**3**) with integrated fuse compartment and the non-detachable power cable (3-2/**2**) for the base unit.

3.3.3 Monitor

For the monitor terminals and the locations of its control elements please refer to the special user manual which is included in the delivery.

3.3.4 Control Computer



1 ON/OFF switch
2 FireWire port
3 2x USB port
4 DVD drive (RW)
5 3.5" disk drive
6 Power input connector
7 Mouse connector
8 Keyboard connector
9 Printer connector (connector for the Matrox dongle; barcode detection software)
10 FireWire port – to be used for the connection of the preview camera (consider labels, if any)
11 4x USB ports – use one of these ports to establish the connection to the MIRAX DESK basic unit and to connect dongles (included in delivery)
12 Network connector
13 DVI port - to be used for connection of monitor
14 DVI port
15 Cameralink ports – to be used for connection to the scan camera (Hitachi HV-F22CL). Consider labels. This plug-in card is provided only if this scan camera type is used.
16 FireWire port – to be used for connection to scan camera (consider labels, if any)
The computer is provided with a separate board for the AVT Stingray F-146C camera. This board is equipped with a FireWire B port.

Fig. 3-3 Terminal points and control elements of control computer

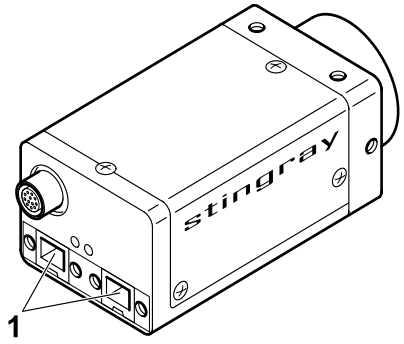


Fig. 3-4 Terminal at AVT camera

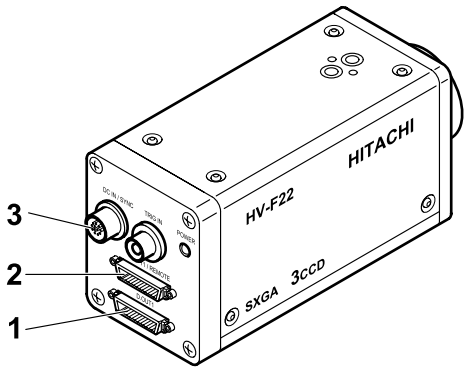


Fig. 3-5 Terminals at Hitachi camera

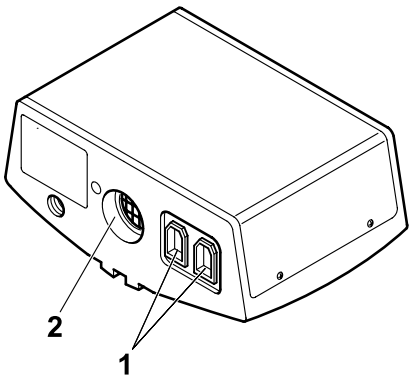


Fig. 3-6 Terminals at AxioCam MRm

3.3.5 Scan Camera

As scan camera can be used:

a) AVT Stingray F-146C FireWire (brightfield and fluorescence applications)

The terminals for the video output port (3-4/**1**) FireWire (IEEE 1394b) are located at the back of the camera.

b) Hitachi HV F22CL (brightfield and fluorescence applications)

The terminals for the video output port (3-5/**1** and **2**) and the terminal for the power supply (3-5/**3**) are located at the back of the camera.

c) AxioCam MRm Rev. 3.0 (for fluorescence applications only)

The terminal for the video output port (3-6/**1**) FireWire (IEEE 1394/iLink) is located at the back of the camera. If an HXP 120 fluorescence device is used, the connection to the lamp (to control the lamp-internal shutters) will be established via the trigger terminal (3-6/**2**).

3.3.6 Cabling of Product Components

For required cable connections, the supplied cables must be plugged as shown in Fig. 3-1 to Fig. 3-6, respectively.

All cable connections must be checked for firm seating.

A defective cable must not be connected. It must be replaced with a cable found to be in proper operating condition. Contact the Zeiss Service for support in such cases.

3.4 Connecting the HXP 120 Illuminator (MIRAX MIDI BF/FL)

For operating the UV radiation source HXP 120 observe in any case the health and safety at work provisions. The national legal regulations have to be complied with as well.

- Follow also the operating instructions for the HXP 120 illuminator provided by the Manufacturer.
- Do not expose yourself to the UV radiation of the HXP 120 illuminator.
- Never look directly into the light guide, when the HXP 120 illuminator is switched on. **In case of non-observance your eyes may be damaged.**
- Never remove the light guide from the MIRAX MIDI BF/FL device when the HXP 120 illuminator is switched on.
- Never connect the light guide to the MIRAX MIDI BF/FL device when the HXP 120 illuminator is switched on.
- Never remove the housing from the MIRAX MIDI BF/FL device when the HXP 120 illuminator is connected and switched on.

The fiber-coupled HXP 120 illuminator outputs the light that is required for excitation in fluorescence mode.

Line power is supplied at the back of the illuminator (3-7/**2**).

Connection between the HXP 120 illuminator and the MIRAX MIDI BF/FL is maintained via a light guide.

One end of the light guide (3-7/**3**) must be plugged into the terminal (3-7/**4**) at the back of the illuminator, the other end (3-8/**3**) into the illumination adapter (3-8/**2**) at the back of the MIRAX MIDI BF/FL. This end must be additionally fixed with a screw (3-8/**4**).

Use clamping screw (3-8/**1**) at the coupling of the MIRAX MIDI BF/FL to fix the collimator (3-8/**2**) of the illumination adapter.

The **Shutter remote** socket (3-7/**1**) of the HXP 120 can be used to connect a trigger cable for Shutter remote control.

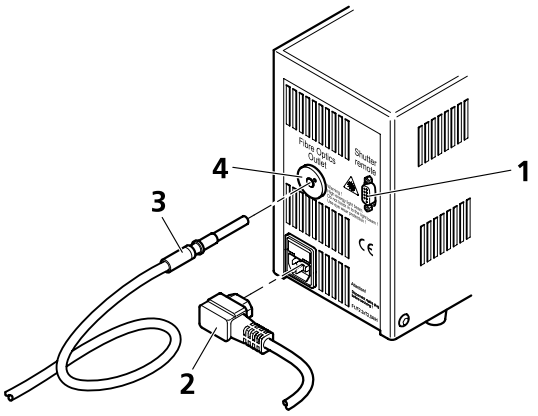


Fig. 3-7 Terminals of HXP 120 illuminator

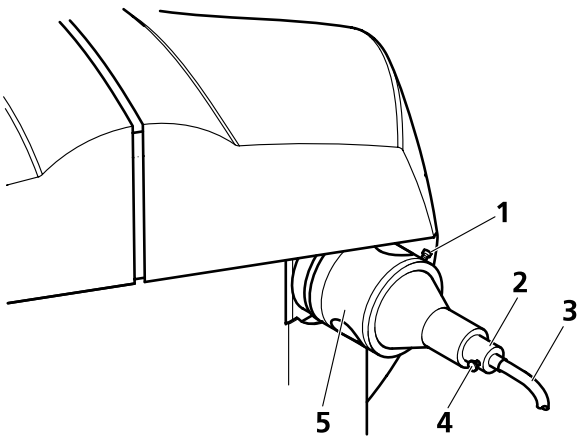


Fig. 3-8 Illumination adapter at the back

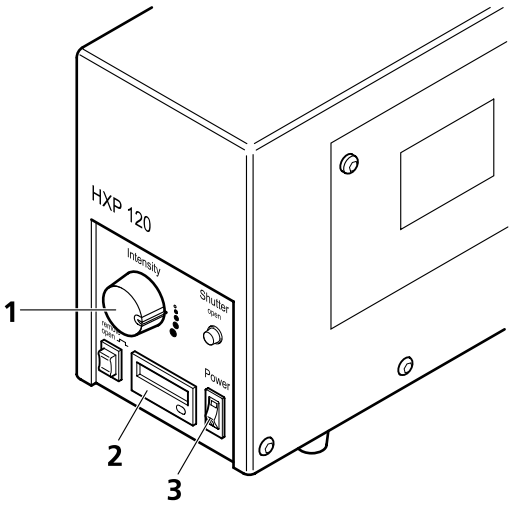


Fig. 3-9 Switching the HXP 120 illuminator on and off

3.5 Switching the Device On

To transfer all components to power-on state, the 24 V power supply unit, the control computer and the monitor must be switched on individually.

- Begin by switching the 24 V power supply unit on. Use the power switch (3-2/**1**) for this purpose.
- Press the on/off switch of the computer (3-3/**5**) to switch the control computer on.
  The operating system will start.
- Switch the monitor on.
- For working with MIRAX MIDI BF/FL, switch the HXP 120 illuminator on with power switch (3-9/**3**) and use setting wheel (3-9/**1**) to adjust for maximum illumination intensity. Reduce the illumination intensity only if the required exposure times would be too short, entailing a camera output with insufficient stability. Check the elapsed-hour meter (3-9/**2**) to make sure that the remaining lifetime of the lamp allows successful digitizing (change lamp, if necessary).
- Wait twenty minutes until the lamp has reached its nominal brightness!
- Start the MIRAX control software (see 0).

The MIRAX MIDI is ready for operation.

3.6 Switching the Device Off

- Exit the MIRAX MIDI control software.
- Shut down computer as required by Windows routine rules.
- Switch 24 V power supply unit off, using the power switch (3-2/**1**).
- Switch monitor off.
- Switch HXP 120 illuminator (3-9/**2**) off, using the power switch (for MIRAX MIDI BF/FL only). Take into account a cooling time of about 20 minutes before switching the HXP 120 on again.

4 OPERATION

4.1 Preparing Slides and Inserts

☞ You may use any type of slides and cover slips that meet the following specifications:

Slides		**Cover slips**	
Length:	75.0 to 76.2mm	Length:	max. 50mm
Width:	25.0 to 26.0mm	Width:	max. 24mm (recommended: 22 mm)
Thickness:	0.95 to 1.05mm	Thickness:	No. 1 and 1.5 (0.13 to 0.16mm and 0.16 to 0.19mm, respectively)

- 45° beveled corners
- Grounded edges
- Recommended: Colored area for placement of barcode stickers

Or

Slides		**Cover slips**	
Length:	75.0 to 76.2mm	Length:	max. 50mm
Width:	24.7 to 26.0mm	Width:	max. 24mm (recommended: 22 mm)
Thickness:	0.95 to 1.05mm	Thickness:	No. 1 and 1.5 (0.13 to 0.16mm and 0.16 to 0.19mm, respectively)

– Cut edges
– Recommended: Colored area for placement of barcode stickers

The following indications apply to all slides:

The distance between the slide edges and those of the mounted cover slip should be 1 mm to 2 mm. The edges of slide and cover slip must be parallel with each other.

Over 0.4 mm thick stickers (or sticker combinations) must not be used.

Please check that no part of a cover slip protrudes from any of the slide's edges when mounting the cover slip onto the slide.

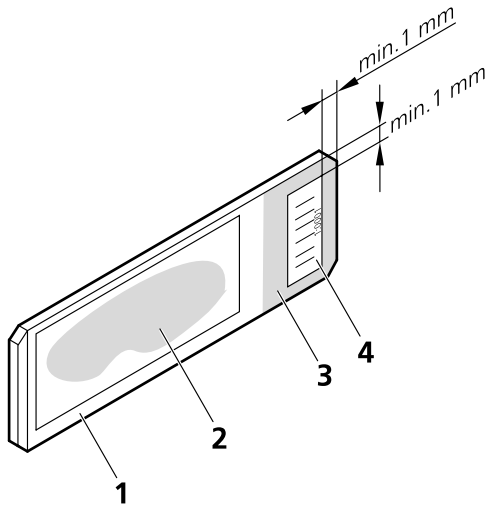


Fig. 4-1 Slide with barcode sticker

4.1.1 Sticking Barcode Label on Slide

A slide's barcode area (4-1/**3**) is recorded by the preview camera and displayed by software. Make sure that a marginal stripe of 1 to 2 mm remains on all sides between the sticker label and the barcode area outer limits.

- Strip off a barcode label from the barcode roll and stick it wrinkle-free on the colored area (4-1/**3**) as shown in Fig. 4-1.
- Note that the barcode label (4-1/**4**) must be stuck on the specimen side (with cover slip) (4-1/**2**) of the slide (4-1/**1**).
- There should be a circumferential gap of 1 to 2 mm between barcode sticker's outer edges and the edge of the barcode area. This is necessary for safe identification of barcode.
- Check that the barcode sticker does not protrude from the slide edges, nor must it be stuck on a part of the cover slip surface.
- As long as the overall thickness does not exceed 0.4 mm, several labels may be stuck on top of one another.

4.1.2 Putting Slides in an Insert

Only original Carl Zeiss inserts are allowed for use with the MIRAX MIDI!

The inserts are designed to hold a maximum of 12 slides.

Push slide (4-2/**1**) into the insert (4-2/**3**) with its barcode end (4-2/**2**) first and the cover slip at the top.

For transportation of a filled insert (4-2/**4**), for example, for introducing it into, or removing it from, the device, the insert should be slightly tilted to its closed side, to prevent the slides from falling out.

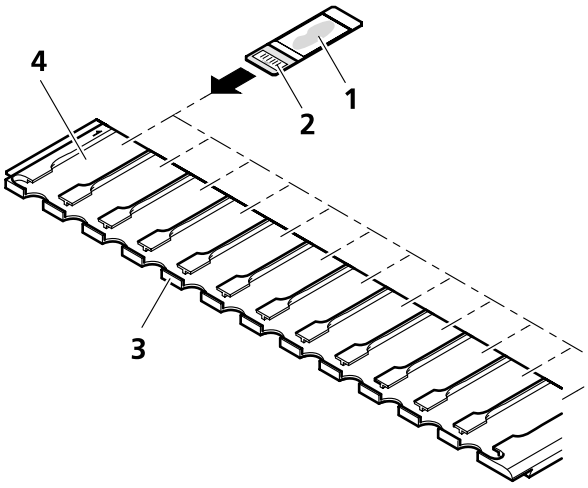


Fig. 4-2 Inserting slides

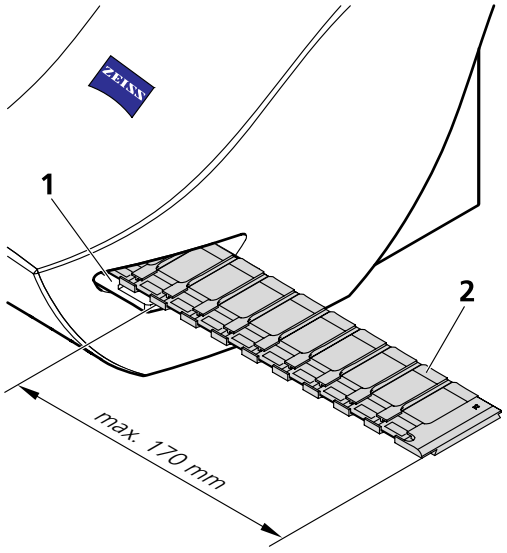
Whether slides will be safely seated and be able to perform guided motion in an insert depends on the cleanness of that insert. For this reason, one should make sure that the inserts are free from dust (compressed air may be used for cleaning) and free from glass breakage and other mounting media residues.

4.1.3 Inserting and Removing Inserts

The MIRAX MIDI has two lateral openings that allow the insert to be moved.

Crush hazard
Do not reach into the movement area of the insert, while a digitization process is going on. Even though the insert is introduced on the right, it may be moved partly out of the device on the left resp. right (80 mm resp. 170 mm, Fig. 4-3).

When placing other objects near the device, consider the movement of the slide insert beyond the basic measures of the base unit during operation. Make sure that no objects protrude into the movement area of the slide insert.



- Introduce the filled insert (4-3/**2**) into the right (viewed from the user's position) opening (4-3/**1**) for digitization. The insert must be introduced completely until a resistance is felt.
- Once the slides have been digitized, the insert is moved to the output position and can be removed there.

Fig. 4-3 Introducing / removing an insert

4.1.4 Special Features in Applying the Double Adapter for Use of two Cameras (Option)

The double adapter for use of two cameras has a manual changeover switch (4-4/**1**) for the light path to the cameras.

☞ Please make sure that the light path is really guided to the camera, which shall be used subsequently.

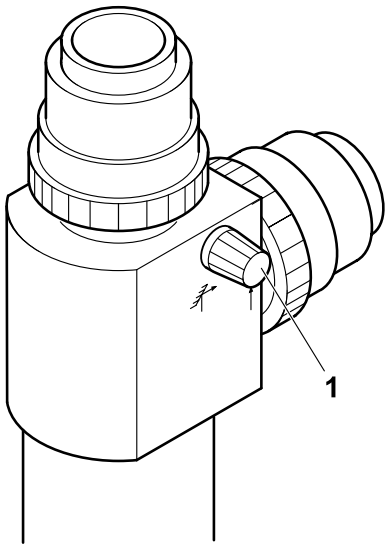


Fig. 4-4 Changeover switch of the double adapter

5 SOFTWARE DESCRIPTION

5.1 Software Version

The software described below refers to the MIRAX MIDI control software version 1.12. Due to technical progress, the screenshots contained in this section may differ slightly from those of the used version.

Fig. 5-1 **MIRAX software icon**

5.2 Starting/ Terminating the Scanning Program

- Double-click the program icon (Fig. 5-1) on the desktop.
  A MIRAX control software session starts.
- Alternatively, trigger program with:
  Start ▶ Programs ▶ Carl Zeiss ▶ Mirax Scan

☞ If your device is equipped with a double adapter for use of two cameras (option), you will find two program icons on the desktop. In the standard case, the monochrome camera and a color camera are used. Then, the desktop is provided with the following two program icons:

MIRAX MIDI – Fluorescence - to start the monochrome camera supporting software,
and
MIRAX MIDI – Brightfield - to start the color camera supporting software. The color camera can also be used for the fluorescence mode.

The double adapter is provided with a manual changeover switch for the light path to the cameras. Please make sure that the light path is really guided to the camera, which shall be used subsequently (see section 4.1.4).

As part of the start-up routine, all motors, the preview camera and the scan camera will be initialized.

On successful initialization, the program will output a selection window (Fig. 5-2).

The scanner is able to work in two modes for brightfield applications (Brightfield Scan Mode):

- ***Automatic***
- ***Manual***

If using the MIRAX MIDI BF/FL, these modes will equally be available for fluorescence jobs (Fluorescent Scan Mode) (see section 5.5).

☞ If your device has been equipped with the MIRAX Monochrome Scan Option, the Brightfield Scan Mode is not available for MIRAX MIDI BF/FL.

☞ It is recommended to use specimens with identical staining for a given insert filling in ***Automatic*** mode.

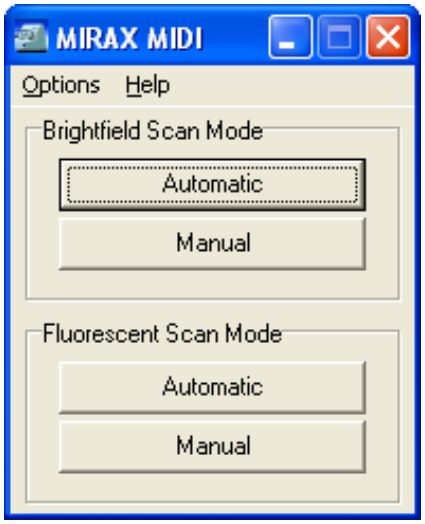


Fig. 5-2 Selection window

Selections for digitization mode should be made following this general rule:

Automatic should be used for digitizing a greater number of specimens.

Manual should be selected:

- if only individual slides need to be digitized,
- where contrast ratio is critical due to specimen staining and where ideal parameters need to be determined for subsequent use in ***Automatic*** mode,
- if profiles shall be defined for ***Automatic*** mode.

A running control program session can be terminated by clicking on the cross in the title bar of the selection window.

5.3 Use of Profiles

The MIRAX control software offers the option of using predefined profiles (brightfield and fluorescence). The profiles are defined in the Manual mode. If profiles are used in the Automatic mode, no further detail settings are required, except for the indication of the memory location of the virtual slides, because all information is provided by the profile. Without profiles, the settings defined in the individual input and dialog fields will be used. In Automatic mode, each slide can be linked with one profile. This can be done by manually entering or selecting the profiles, or profile - slide assignments can be imported with a file.

Profiles are storing the following information:

Scan Mode

- Brightfield or fluorescence

Scan Options

- Fine focus frequency
- Coarse focus frequency
- Compression procedure: JPEG, PNG, or BMP; in case of JPEG: image quality in %
- Use of a fixed compensation image: YES/NO (brightfield)
- Use of Stitching: YES/NO
- Use of database: YES/NO + settings
- Use of Bayer mask: YES/NO

Determination of the area to be scanned

- Use of threshold value: YES/NO + threshold value
- Use of a fixed scan area: YES/NO + path to the image of the scan area
- Use of the distance to smaller areas: YES/NO + size of the areas
- Filling of holes: YES/NO
- Use of marker: YES/NO + threshold value of the marker
- Use of limitation (Use Selection box): YES/NO + size

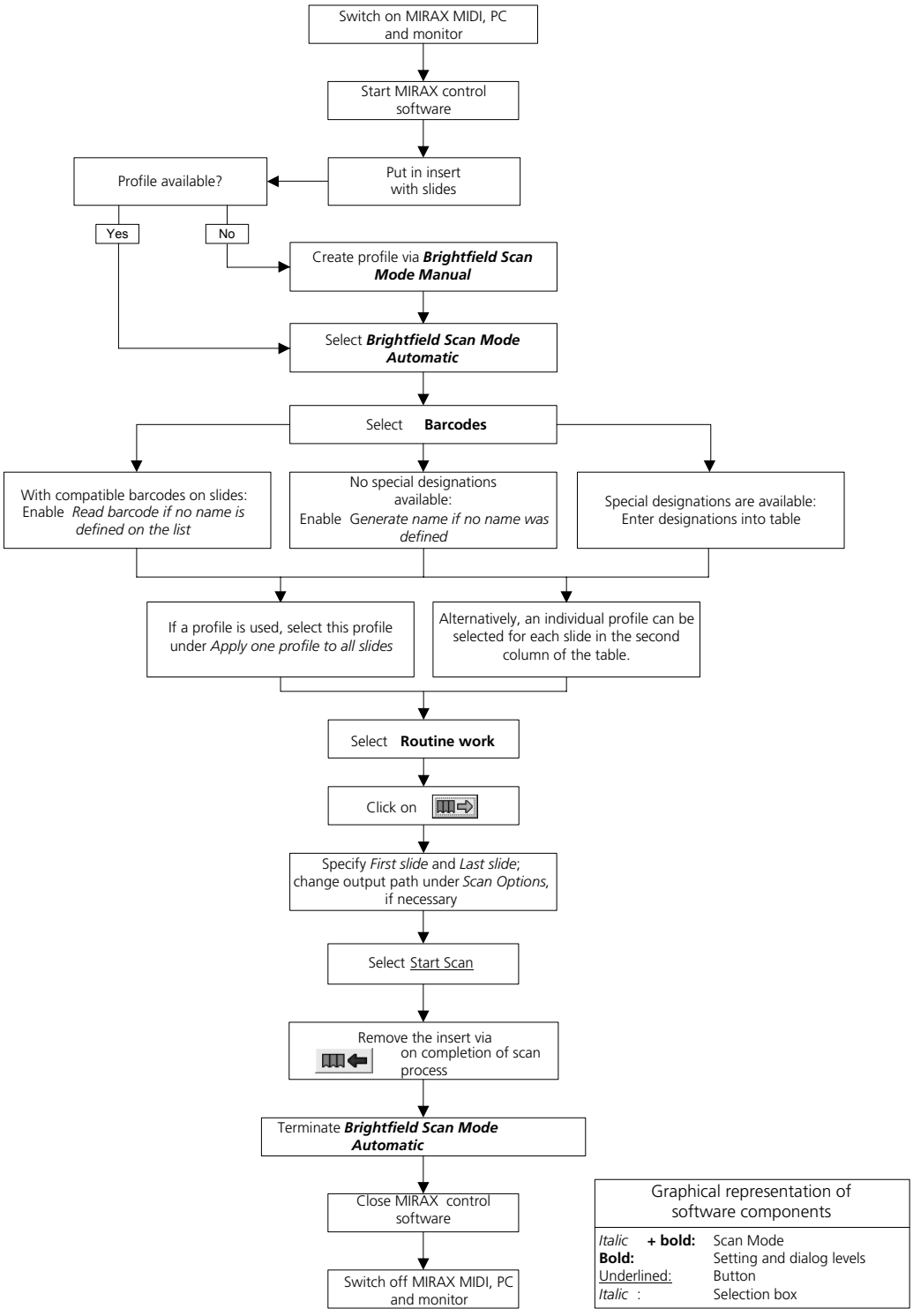
Other settings

- Use of Optovar: Position in filter wheel and name
- Limitation of focus range: YES/NO + lower limit / upper limit
- Use of extended depth of sharpness (for fluorescence, this is a channel-specific entry): YES/NO + step size and number of levels
- Use of Gain (fluorescence)
- Use of Exposure weight (fluorescence)
- Exposure time (fluorescence)
- Manual focus plane (fluorescence)
- Use of a fixed compensation image: YES/NO (fluorescence, channel-specific)

5.4 Brightfield Applications

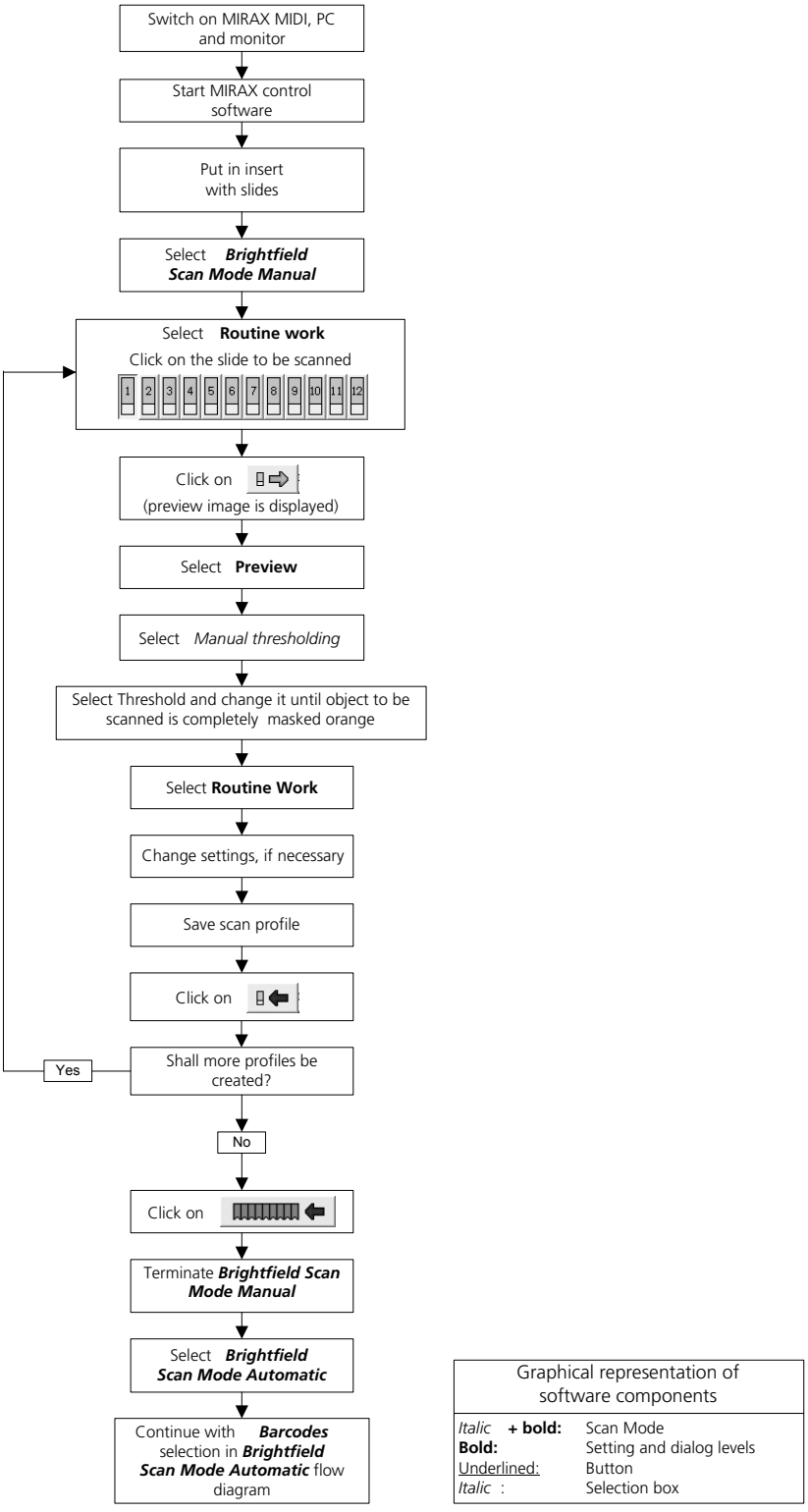
5.4.1 Digitizing Slides in *Automatic* Mode (Standard Procedure 1)

This work procedure should be applied when a greater number of slides needs to be digitized.



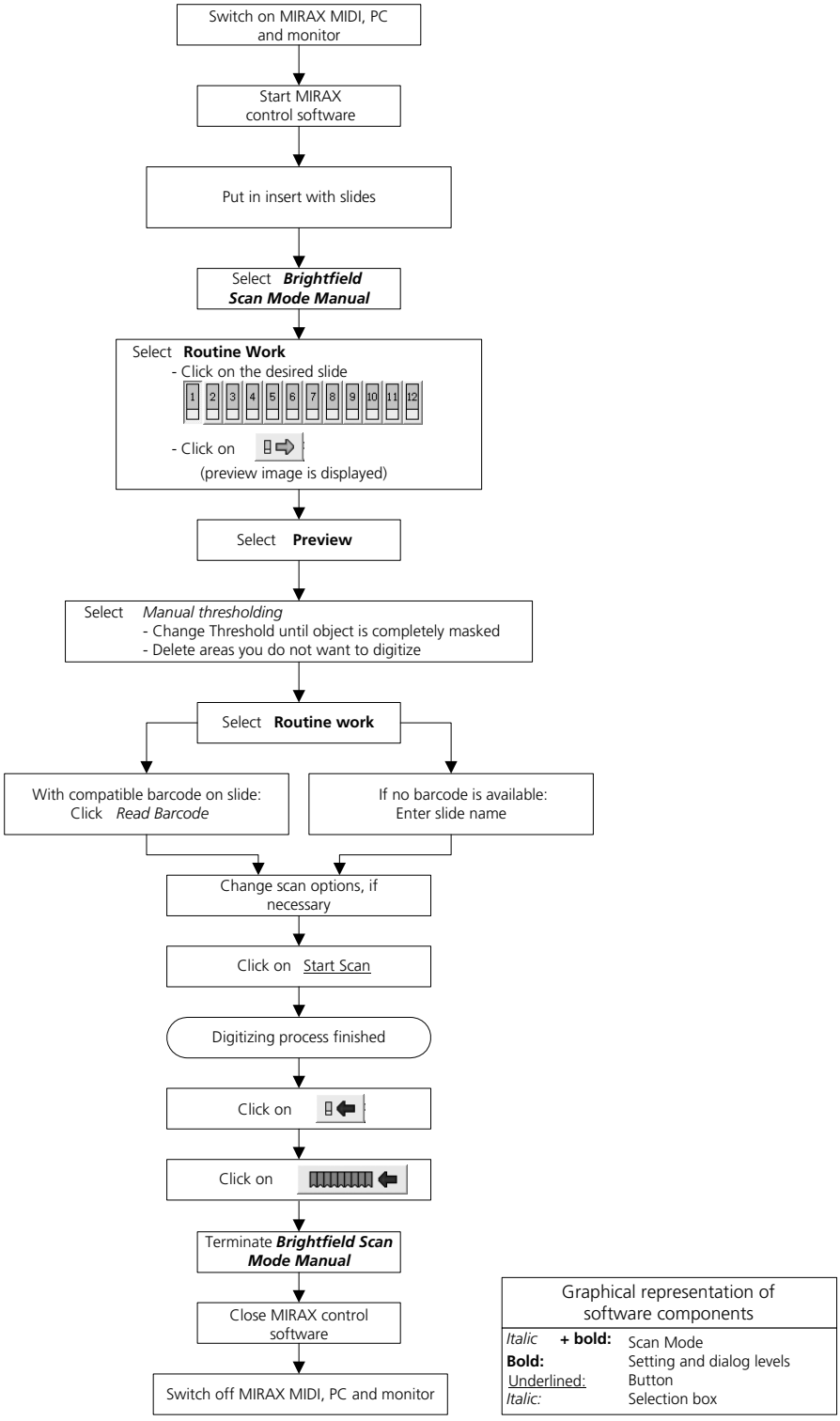
5.4.2 Creating a Profile (Standard Procedure 2)

This procedure is appropriate when a profile shall be defined for its subsequent use in Automatic mode.



5.4.3 Digitizing Slides in *Manual* Mode (Standard Procedure 3)

This work procedure is intended for individual slides.



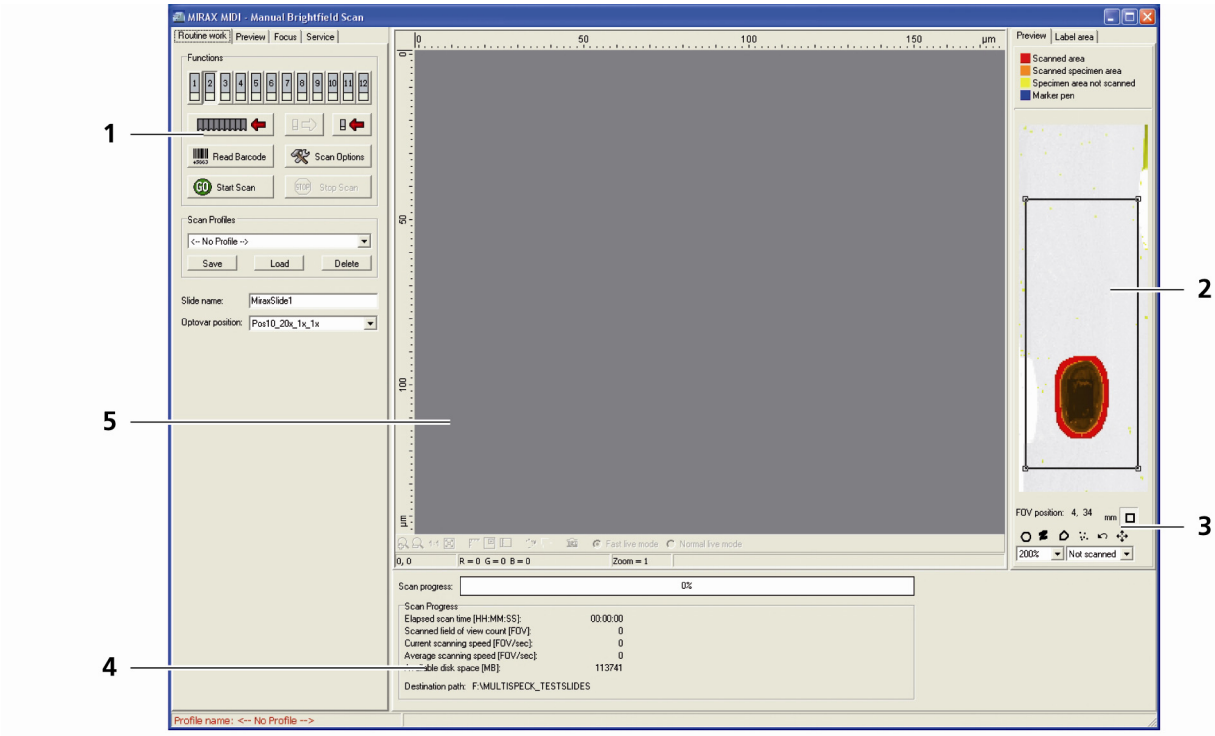
5.4.4 *Brightfield Scan Mode Manual* Mode

Manual mode is intended for digitization of single slides. It can also be selected for slides which are distinguished by e. g. low-contrast staining so the area for digitization should be determined manually. The most important function, however, is the definition of profiles which shall be used in the Automatic mode.

So, available tab functions can be used to determine and define parameter settings as appropriate to achieve optimal digitization results. Once selected, these settings can be applied in ***Automatic*** mode.

Following a click on Manual in the mode selection window below *Brightfield*, the display and dialog window of the ***Manual*** mode will open with pertaining tabs (Fig. 5-3). These are:

- **Routine work:** Fundamental scanner settings, loading and unloading of inserts and slides, administration of profiles, and triggering of digitization process
- **Preview:** Settings for specimen recognition
- **Focus:** Settings for focusing sequences
- **Service:** Auxiliary operation and maintenance functions



1 Dialog and function block with scanner functions and scanner settings
2 Preview image recorded with preview camera (areas to be digitized are shown in red/orange color)
3 Toolbar for editing of preview image and for manual positioning of slide in relation to optical axis
4 Sub-area informing about the progress of a running digitization
5 Live image and camera image display field

Fig. 5-3 ***Manual*** **dialog and display window**

Clicking on the cross in the window's title bar will terminate ***Manual*** mode. The mode selection window turns active again.

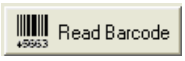
A slide which is contained in the device at that moment will automatically return into its insert as soon as a manual mode session is ended.

5.4.4.1 Routine work Tab

Routine work is the tab that provides basic digitization functions.

Data entry options or function trigger options are as follows:

- With a click on a slide included in the symbol 

, the selected slide will be moved to the transfer position.
- With a click on the symbol  , the selected slide will be transported from the insert to the specimen holder; a preview image is automatically generated and displayed.
- A compatible barcode on the slide will be captured automatically, once the 

button has been actuated. It will also appear in the *Slide name* display box.
- With a click on the symbol  , the slide will be moved back into the insert.
- With a click on the symbol  , the slide insert will be moved to a position where it can be removed by the user.

Slide name:
This input field is available for manual entry of a desired slide name, under which the image data relating to this slide are to be saved.

- The *Scan Profiles* field is used to administrate the profiles:
  A click on Save (Fig. 5-5) will save the current settings. The system will ask for a new profile name.
  All existing profiles can be selected via the selection menu and enabled by clicking on the Load button. The current profile will be overlaid at the bottom on the left Profile name: HE_stain_std. The current profile will be deleted by clicking on the Delete button.

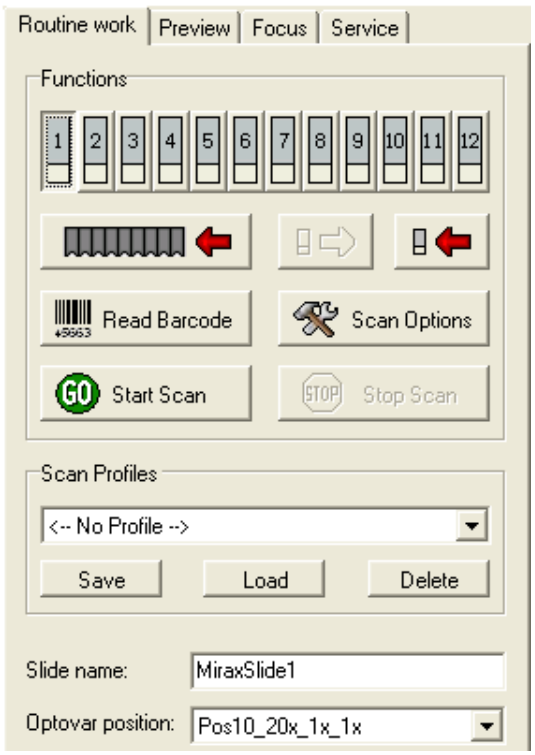


Fig. 5-4 Routine work tab

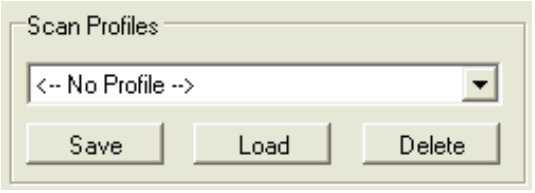


Fig. 5-5 *Scan Profiles* field

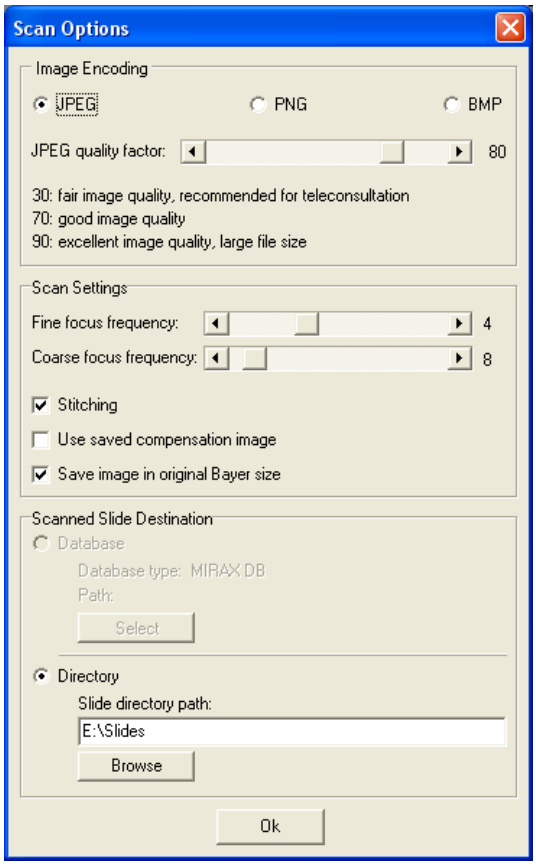
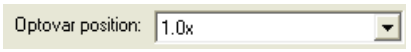
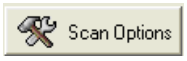


Fig. 5-6 Scan Options

- The selection field 

is used to select an Optovar for digitizing purposes. This option can only be used if one or several Optovars have been installed.

- Following a click on the 

button, the **Scan Options** dialog window will open. It provides parameter options for digitization process settings.

Image Encoding and *Scan Settings* allow parameter selections which will determine the resulting quality of a virtual slide. Fig. 5-6 shows preset values.

The table below provides a more detailed explanation of *Image Encoding*, *Brightfield Compensation* and *Scan Settings*:

Parameter	Explanation
Image encoding	Sets image format for individual image fields.
JPEG	Saves image fields as Joint Photographic Experts Group files (jpeg) with a certain degree of loss. The resulting quality can be adjusted with the *JPEG quality factor* slider button. Great values yield images of high image quality. Such files will, however, be quite big. Small values result in low-quality images and small files. The default setting of **80 %** provides the best possible compromise of image quality and file size, with smaller value settings (e. g. 70%) still yielding images of a high quality level.
PNG	Saves the various images as Portable Network Graphics files (png). This method guarantees loss-free saving of image information. PNG was raised to the standard procedure by W3C in 2003 (ISO standard ISO/IEC 15948:2003), also refer to BMP.
BMP	Provides loss-free saving of the various images as Windows bitmap files (bmp). Since the compression algorithm proves less effective with BMP than with PNG files, the resulting files are greater with the BMP option than with the PNG option.
Fine focus frequency	Allows you to define after how many in a series of snapshots fine focusing (focusing through a small z-range with few steps) is to be performed. This setting will influence the speed of the digitization process, because a fine focus routine will be carried out on completion of a defined number of image fields and focusing is a time-consuming sequence. The default setting of **4** is applicable to the majority of specimens. For a specimen with strong non-planarity, you should pick a smaller value for this parameter. The same applies to cytological specimens. A setting of "one" will define that focusing is to be performed for each image field. Although this results in 100% focused image sequences, it also takes essentially more time to digitize.
Coarse focus frequency	Allows you to define after how many in a series of snapshots coarse focusing (focusing through a great z-range accomplished in several steps) is to be performed. This setting will influence the speed of digitization, because a coarse focusing sequence will be carried out after a specified number of image fields and focusing itself is a time-consuming procedure. Coarse focusing takes more time than fine focusing. The default setting of **8** is OK for most specimens. Basically, the statements for *Fine focus frequency* apply to this parameter as well. Typically, this setting should be twice as great as the setting for *Fine focus frequency*.
Stitching	By turning the *Stitching* function on, the user defines that the various tiles of a digitized slide will seamlessly stitch together in a subsequent MIRAX Viewer session with no perceivable positive or negative overlapping. If this function is active, digitization will automatically work with a greater overlap, which leads to increased digitization time (about 27% longer than with this function in disabled state). The resulting files will also be bigger by as much. When this function is deactivated, the shortest digitization time and the smallest file size are attained, but overlapping or underlapping may occur in the resulting image.

Parameter	Explanation
Use saved compensation image	If this option is disabled, several reference images of empty slide areas will be recorded automatically for each slide at the beginning of the digitization process for white balancing and shading. On the other hand, where slides almost fully covered by a specimen are used (e. g. smears), the automatic search for empty slide areas is bound to fail in some cases. You should note that action to search for empty image fields is restricted to *Use selection box* (see section 5.4.4.2). By turning this option on, you may use a previously recorded reference image (see section 5.4.4.4) as compensation image for mathematical setting off against the digitized fields.
Save image in original Bayer size	If this option is on, the various image fields will be saved with interpolated pixel resolution (0.23 µm per pixel with standard configuration). If it is disabled, you will work with reduced pixel resolution (0.32 µm per pixel for standard configuration), and the resulting files will be smaller (by about 40%). This option is not available for the MIRAX Monochrome Scan option and the MIRAX 3Chip Scan option, because these cameras do not have a Bayer mask.

The following table provides explanations to the various selections which are available for the *Scanned slide destination* field:

Parameter	Explanation
Database	This is to enter the required information if a database interfacing is used.
Directory	If this function is enabled, the virtual slides will be saved into a directory to be specified. A Browse button is available for conveniently choosing a directory via the special selection window. If a non-existing path is specified, this path will be created by the software on acknowledgement. This indication will not be saved in the profiles.

All parameter inputs will be saved and will be available for reuse during the next software session.

To initiate digitization of a loaded slide, you need to click on the Start Scan button.

If the selected slide is a representative of a greater lot of slides, it makes sense to save the settings via a profile (or profiles) and to continue operation in the ***Automatic*** mode.

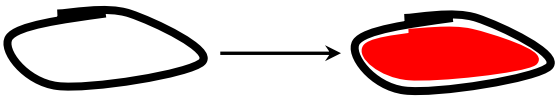
5.4.4.2 Preview Tab

Two options for the determination of the threshold value are offered under *Use threshold* in the **Preview** register card. A given threshold setting defines which areas will be masked (shown red and orange in the slide preview). Only these masked areas will be included in a digitization process at high resolution:

Scan threshold specimen
A desired threshold value is selected directly by changing the threshold setting with the help of the arrow buttons in the *Specimen threshold level* selection box.

Small threshold values result in a more sensitive scanning of the areas to be digitized (but also dirt portions or other impurities will be preferentially recorded and included in subsequent digitization). Greater threshold values are known to be insensitive to impurities, but it may occur that some of the desired specimen areas fail to be masked and, hence, will not be digitized.

Scan area inside the marker
If a specimen is identified with the above option as one that is not to be digitized (e.g., extremely weak contrast), the specimen may be circled with a black marker pen. The threshold value defining the marker can be set in the *Marker threshold level* selection window. *The Specimen threshold level* to be selected defines the threshold value for the specimen within the marker. If the whole area located within the marker is to be digitized after all, irrespective of the threshold value of the specimen, the *Scan everything within marked* control box must be activated.



If option *Fill holes in scan area* is active, any non-masked area within a given masked area will also be masked. This function may prove helpful where, e. g., a specimen exhibits a contrast that decreases from outside to inside. When you work with default settings, it may happen that an inner area of little contrast (e. g. fatty tissue) is interpreted as not selected for digitization. In such cases, this function will mark also these areas as due for digitization (red).

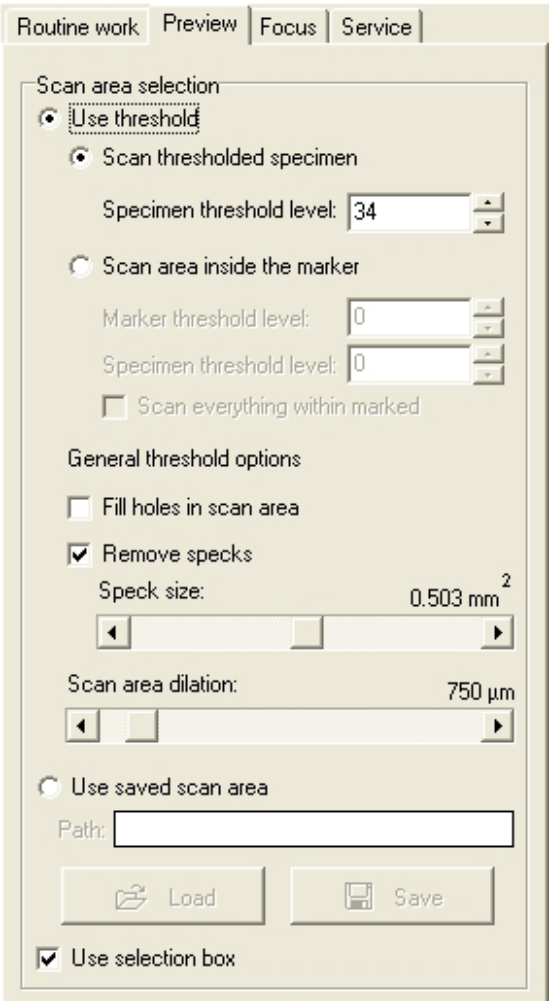


Fig. 5-7 Preview tab

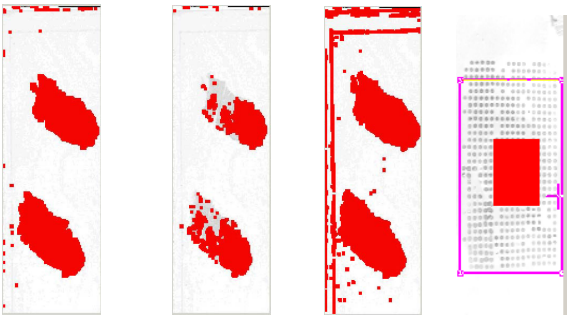
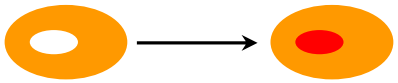
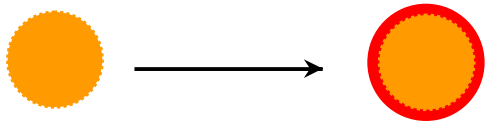


Fig. 5-8 Threshold values and masking



Remove specks
If the threshold value is used for the detection of the specimen, high-contrast dirt and other artifacts will be recognized as well. Those artifacts are small in length. The *Remove specks* function may be used to exclude them from digitization. When this function is activated, all the areas which would be recognized as a specimen based on the threshold value, but are smaller than the value set under *Speck size*, will not be digitized. A value of 0.5 mm² has proved best suited for standard specimens.

By activating the *Scan map dilation* function, the red or orange masked area can be extended. This edge extension is specified in µm. The value cannot be changed continuously, because the edge is extended discretely by one image field of the camera each time. The default value is **250 µm** (in the standard configuration); i.e. the area detected by the preview camera (shown in orange and depending on the threshold value) is extended by one image field (shown in red color).



Use saved scan area
Various toolbar resources are provided (Fig. 5-15) to the user for selection of areas to be included in or excluded from digitization.

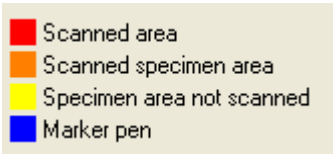
You may save a manually defined mask via Save and call this mask again via Load if required at some later point in time.

The option *Use saved preview* appears suitable where slides have to be handled, which contain small single objects or areas of interest that cannot be resolved by the preview camera with adequate precision, for example, a given number of cytological samples.

The edges of a coverslip will be detected by the preview camera and included as areas of interest to be digitized, depending on the particular threshold value setting (see Fig. 5-8; preview image subtitled "low"). Since the digitization of such edges is a time-consuming process that occupies extra memory capacity of the hard disk, you may use the *Use selection box* function to exclude these areas from digitization. If this function is active, a frame will be superimposed on the slide (5-9/**1**). Since, normally, the position of a coverslip can be reproduced, this function guarantees that only those masked areas will be included in digitization, which are located within a user-definable area (rectangle), while excluding not desired areas from digitization, although these may have been masked.

This rectangle can be enlarged or diminished by dragging with the mouse. To achieve this, position the mouse pointer on one of the edge lines, click with the mouse and change the rectangle's size. All areas of interest previously masked red or orange which are now located outside the altered frame line are thus eliminated, and solely the masked areas within the new boundary frame will be digitized.

Various colors are used in the slide preview to show the user the latest state:

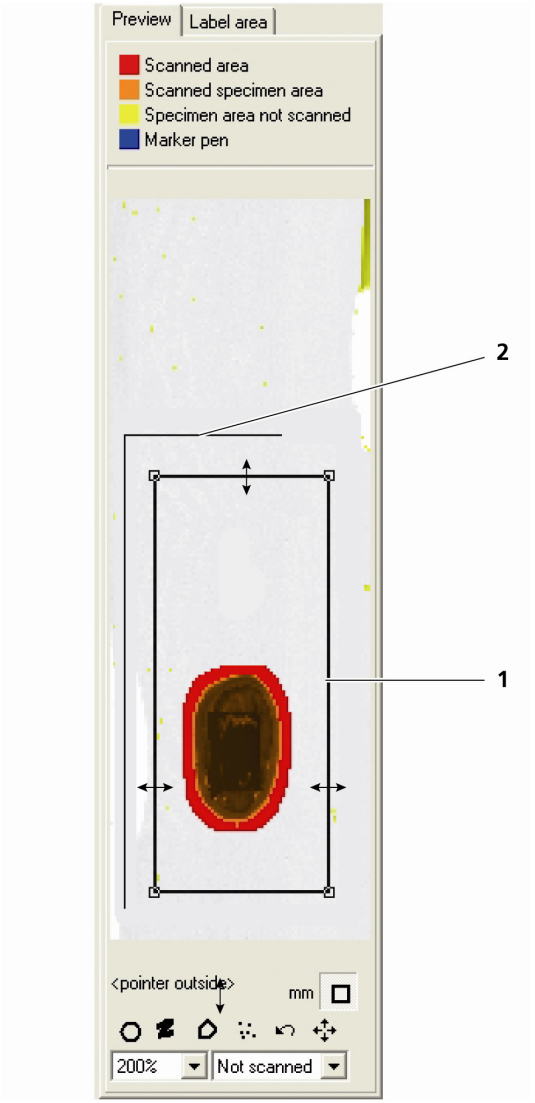


Red (*Scanned area*): These areas will be digitized later at high resolution. They are defined by the user and not directly derived from the threshold value (e.g. the filling of holes or user-specified areas).

Orange (*Scanned specimen area*): These areas will be digitized later at high resolution. They are recognized by the selected threshold value (see above).

Yellow (*Specimen area not scanned*): These areas have been detected by the preview camera, but they will not be digitized, for example due to their position outside the selected area (frame; selection box).

Blue (*Marker pen*): The system shows that a marker pen has been detected here (is only indicated if the option *Scan marker specified area* has been enabled). The inside areas appear in red color and will be digitized.



1 Frame (all areas outside the frame will be excluded from digitization; e. g. the coverslip edges)
2 Edges of coverslip

Fig. 5-9 Use selection box option

5.4.4.3 Focus Tab

On selection of the **Focus** tab (Fig. 5-10), you may make online adjustments as required in manual mode. You can also work with the focus tool in offline mode.

The *Live* sub-area enables a live image of the scan camera, once the Live button is turned active. This live image is displayed in the live image and camera image display field (see section 5.4.5). This option has no effect on a running digitization process and no live image will be displayed. If this is required, you need to select option *Show images during scanning* in the **Service** tab.

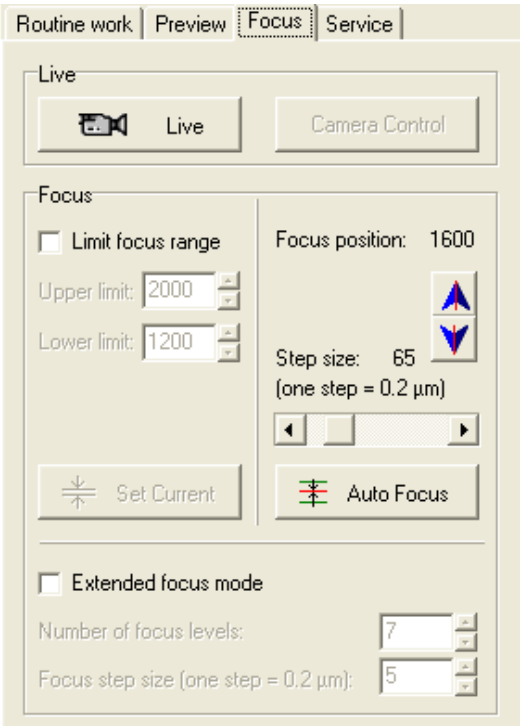


Fig. 5-10 Focus register tab

Focus is the sub-area for setting restrictions to the focus range. If *Limit focus range* is active, you may define a lower focus range limit via *Lower limit* and an upper focus range limit via *Upper limit*, using the respective arrow keys.

This function may prove necessary if, as a result of a soiled coverslip surface, focusing is targeted on a faulty spot on the coverslip surface rather than the actual specimen you want to digitize.
With the help of the Set current button you can set the *Lower limit* value to the current focus value (1600 in the given example) –32 and the *Upper limit* value to the current focus value +32.

Clicking of the Auto Focus button will initiate a focusing sequence in the image marker position. A restriction to the focus range possibly set (*Limit focus range*) will be ignored.

☞ For Auto Focus, the live image display should be turned off, because this function will reduce the processing speed of any other function that runs concurrently.

The *Step size* selection box allows you to define a manual focus step size with the help of the two arrow buttons.

The blue vertical arrow buttons are intended for manual focusing. This will have no influence on the way slides are digitized in automatic mode.

With the mechanical (cross) stage function (5-15/**9**) in active state, a slide can be viewed like under a microscope. As a necessary precondition for this, the Live button must also be active.

Furthermore, the **Focus** tab may include a possibility to work with an *Extended focus mode* function. This extended focus functionality is provided as an option.

If this function is active, several images will be captured in different focus planes for each image field. The regions of strongest contrast which belong to given images of a z-stack are combined into an image that will contain the maximum contrast portions of all images recorded for this image field. This method guarantees a maximum in depth of sharpness. While exerting no influence in terms of the file size, it does have an impact on digitization speed, because a set of images has to be shot and mathematically combined into a single image.

Once the increased depth of sharpness function was turned on via the *Extended focus mode* control box (Fig. 5-10), you may determine a desired number of focus planes via the *Number of focus levels* selection box. The currently valid focus value (defined by z-value as determined by auto-focusing) is always interpreted as the starting point and point of reference. A value of "5" means that the resulting image will be calculated, using the current focus value, two over-focused images and two under-focused images as mathematical input. For this reason, you are advised to pick an uneven number for *Number of focus levels*.

The mutual spacing between the individual images in z-direction can be specified via *Focus Step size*. It designates the number of motor steps (one motor step roughly equals 0.2 µm).

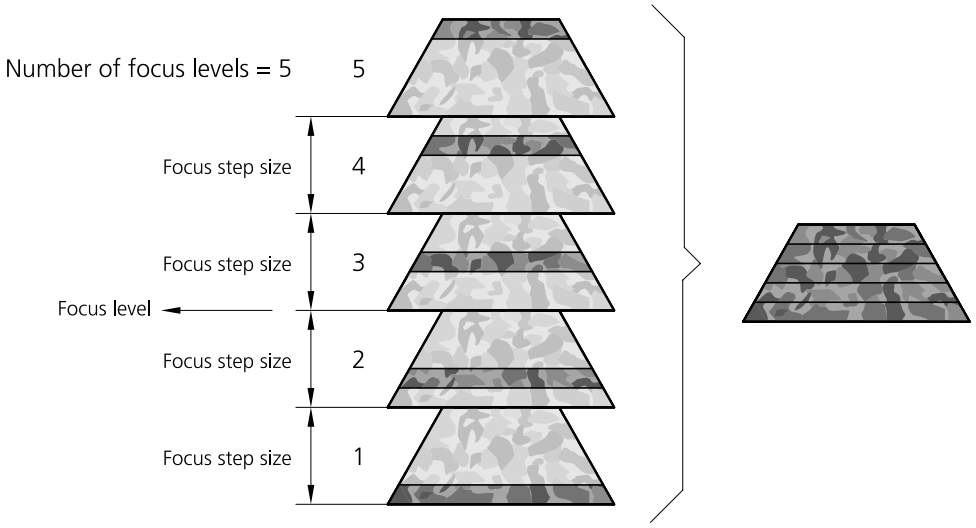


Fig. 5-11 Graphical view of *Extended focus* process (bright areas are unsharp zones, the darker areas are sharp zones)

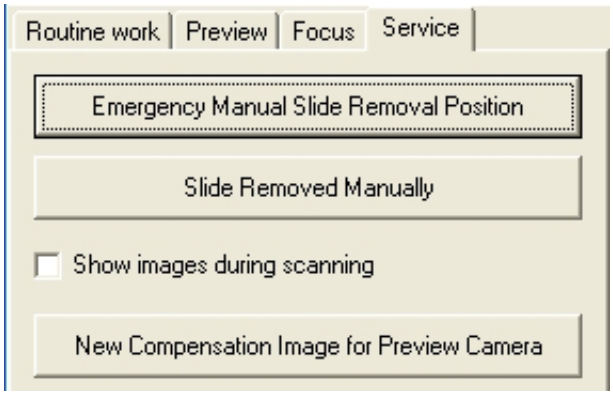


Fig. 5-12 Service tab

5.4.4.4 Service Tab

The **Service** tab contains specific control and maintenance functions that can be turned active.

If the Emergency Manual Slide Removal Position button was actuated, the MIRAX MIDI will move the specimen holder into the position for removal, i.e. the slide can be removed by the operator after the cover has been opened (see section 6.2.5).

On completion of this procedure, the system should be able to detect the absence of a slide by itself. If there are still problems in terms of the system suspecting a slide in the holder, you may click on the Slide Removed Manually button, thus notifying the software that the slide has been removed.

If the *Show images during scanning* control box is active, the currently scanned image field will appear in the live image or camera image display field (5-13/**3**).

☞ *Show images during scanning* should be activated only in exceptional cases or for control purposes, because this will slow down the digitization process.

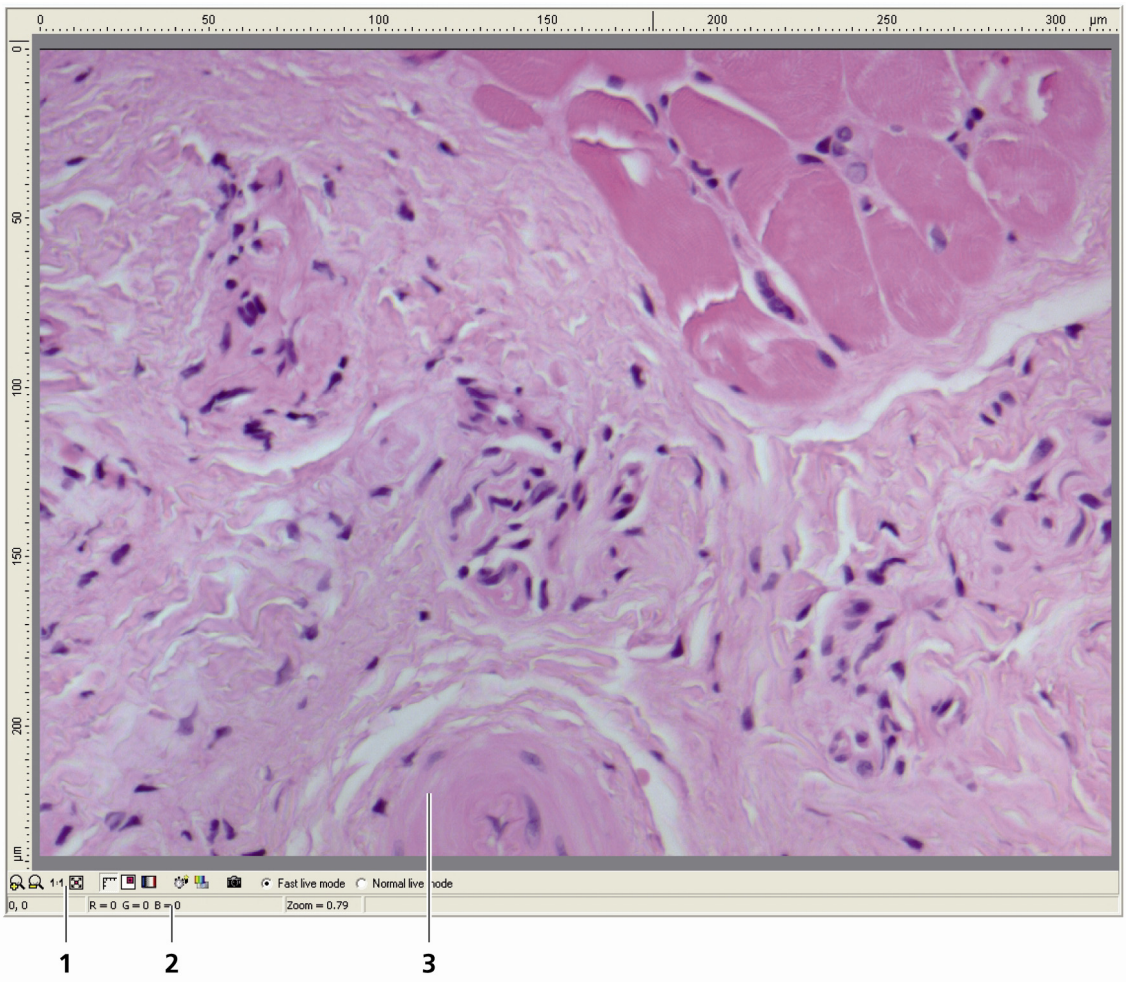
To make this function operational for digitization, you must activate the *Use saved compensation image* item in the scan settings via the *Scan Options* button of the **Routine work** tab.

If the preview image of a slide fails to deliver a satisfactory result, you should generate a new compensation image for the preview camera. This procedure is initiated by clicking on the New Compensation Image for Preview Camera button. For this procedure to be successfully completed, the user should take care that no slide is on the specimen holder. If the system still finds the device to contain a slide, the slide will be transported back into its insert.

5.4.5 Display Field for Live or Camera Images

The display field for live or camera images shows images of the scan camera (Fig. 5-13).

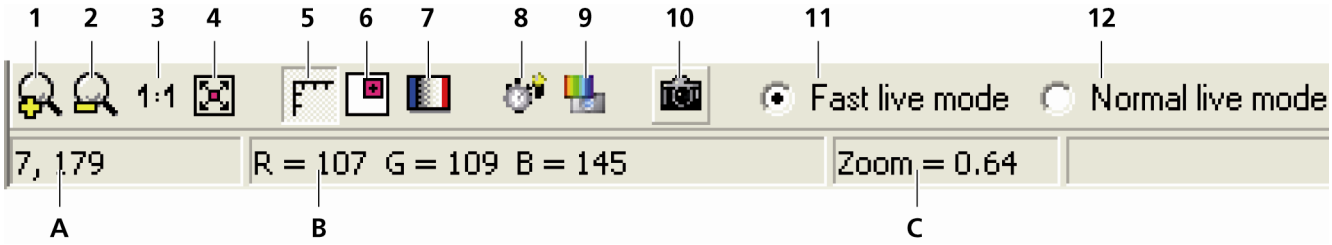
The live image will only be displayed if there is no digitization process running at the moment. To enable display during digitization, the *Show images during scanning* option of the **Service** tab must be activated.



1 Toolbar
2 Status bar
3 Live or camera image

Fig. 5-13 Display field for live or camera images

Different actions for the displayed live image can be triggered with the toolbar, like zooming or determination of exposure time, etc. (Fig. 5-14). However, these possibilities are not available for camera images during digitization.



Toolbar

1 Enlarges the live image.
2 Scales the live image down.
3 The live image is displayed in a way that one pixel on the monitor corresponds to one pixel on the camera chip.
4 Changes the size of the live image in a way that it completely fills the display window area.
5 Displays or suppresses the display of the horizontal and vertical image bar with scaling.
6 Superimposes or hides the magnifying window.
7 Activates red staining of all overexposed pixels.
8 Re-calculates the exposure time for the scan camera.
9 Triggers another white-fader routine (for color cameras only)
10 Activates a dialog window with which the current live image can be saved as a bitmap or JPEG image.
11 With this mode activated, the live image will always be shown with a frequency of at least 10 images per second. If an exposure time of more than 100 ms is set, the pixel values will be multiplied accordingly (if an exposure time of 200 ms is set, the pixel values will be multiplied by 2 and the system will reset the exposure time to 100 ms), i.e. the resulting images are characterized by a higher noise level.
12 If this mode is active, the images will be taken with the user-specified exposure time.

Status bar

A Current position of the image mark related to the live image
B Red (R), green (G) and blue (B) value of current pixel at the mouse pointer
C Zoom factor of the current live image (1:1 means that one pixel on the camera corresponds to one pixel on the monitor, i.e. a zoom of 1.00)

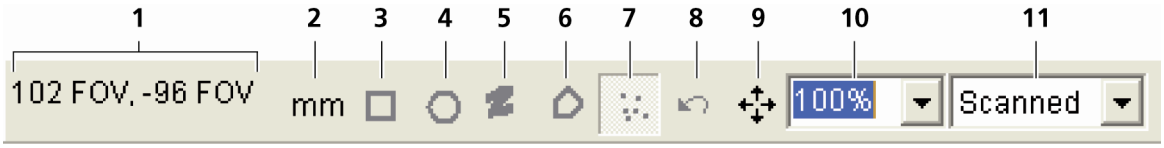
Fig. 5-14 Toolbar and status bar display field for live or camera images

5.4.6 Toolbar for Preview Image

Regardless of which tab is currently selected, the toolbar will always be displayed. The most important toolbar functions allow you to select the areas you want to be digitized or not digitized, when the area to be digitized has to be selected manually (e. g. in the case of certain cytological specimens) or if unnecessary areas (e. g. dirt) are to be deleted.

These functions are described in Fig. 5-15 under **3** to **8** and **11**. These functions are helpful if the **Preview** or **Focus** tab was selected.

The mechanical stage function (5-15/**9**) activates the shifting of the slide relative to the optical axis. The shifting is controlled by clicking with the left mouse-key into the preview image. It is recommended to enlarge the preview image via scaling (5-15/**10**) for a more simple navigation. The current position is displayed via a cross. A click on one of these symbols (5-15/**3**, **4**, **5**, **6** or **7**) disables this mode again.



1 Coordinates of cursor position (status display)
2 Button for specifying the cursor position (mm or numbering)
3 Button for selecting a filled rectangle
4 Button for selecting a filled circle
5 Button for selecting a free-hand line
6 Button for selecting a filled polygon
7 Button for selecting dots
8 If changes were made in the preview image, a maximum of 40 changes can be undone step-by-step by clicking this button in one-click mode.
9 Tool to place the image mark at any desired position on the slide by a click; when the tool is active the slide can be shifted continuously in two coordinates to the display system in the same way as for the mechanical stage of a microscope. The shift towards z-direction (focusing) must be done via the settings in the **Focus** tab. This function can be disabled again by clicking on one of the symbols (**3** to **7**).
10 Scaling for the preview image (selection in preset steps using the selection box)
11 Processing mode for selected areas (in combination with the symbols **3** to **7**):
Not scanned: Selected areas will be deleted from the preview
Scanned: Selected areas will be added

Fig. 5-15 Toolbar for area selection in preview window

5.4.7 Preview Image

The preview area displays two preview images each depending on the selected tab:

Preview tab	**Label area** tab

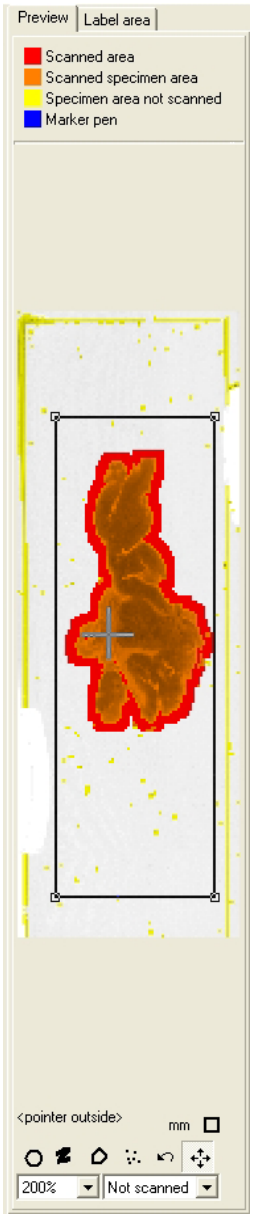
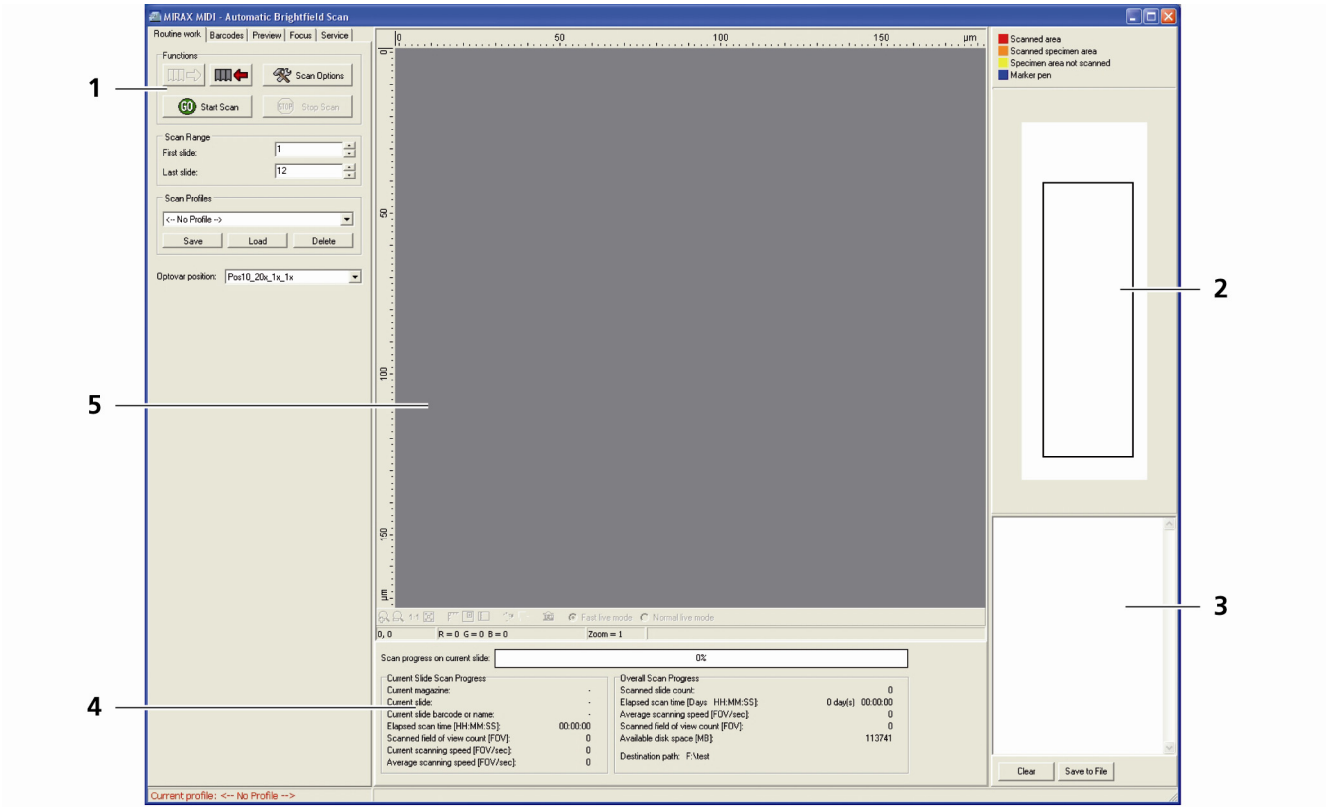



Fig. 5-16 Specimen preview image	**Fig. 5-17** Barcode area preview image
This figure displays the specimen image, with a mask to show the area being digitized and with a black frame if *Use selection box* is active.	This figure displays the barcode area image. With the help of checkbox *Rotate image by 180 degree* the image can be rotated by 180 degrees in cases when there is lettering upside down. This image will be saved together with the digitization data and will be available in the MIRAX Viewer.

5.4.8 *Brightfield Scan Mode Automatic*

After a click on Automatic, the display and dialog window for ***Automatic*** operation will open with these tabs (Fig. 5-18):

- **Routine work:** Provides fundamental scanner settings and initiates scan process.
- **Barcodes:** Setting and input options for identification of slides and assignment of profiles
- **Preview:** Settings for optimized preview images
- **Focus:** Settings for focusing (e.g. limitation of focus range)
- **Service:** Auxiliary functions to support operation control and maintenance



1 Dialog and function block for scanner functions and scanner settings
2 Preview image as recorded by preview camera (areas subject to digitization are shown in red/orange color)
3 Logsheet window with digitization process data
4 Information field showing latest state of digitization process of currently processed slide and overall progress state
5 Display field for camera images

Fig. 5-18 ***Automatic Brightfield Scan*** **dialog and display window**

A click on the cross in the title bar of the window will terminate ***Automatic*** mode operation. The mode selection window will turn active again. A slide which may be contained on the specimen holder at this moment will automatically be pushed back into the insert.

As the MIRAX control software offers the option to use predefined profiles, you should make use of it.

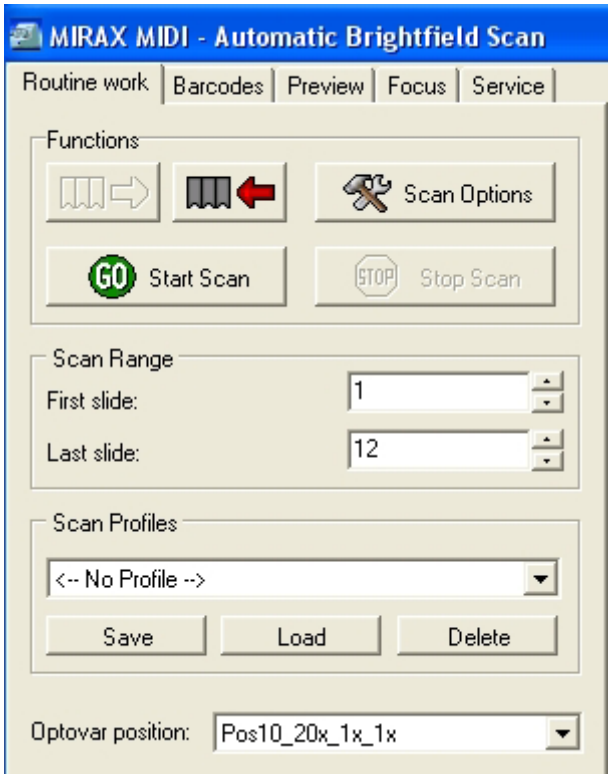


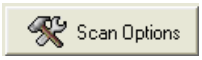
Fig. 5-19 Routine work tab

5.4.8.1 Routine work Tab

Routine work designates the tab with fundamental digitization functions.

The following data can be entered and functions triggered here:

- First, the insert must be loaded by clicking on the  button. During the loading process the system is checking the positions for slides.
- Specification of first slide of the batch to be digitized *(First slide*).
- Specification of last slide of the batch to be digitized (*Last slide*).
- With a click on the  button the insert will be moved to a position where it can be taken out by the customer. Slides possibly situated in the specimen holder are moved beforehand into the insert.

By clicking on the 

button, you can open the **Scan Options** dialog window (see section 5.4.4.1). It provides setting options for the digitization process. These setting values will be valid for all slides of a batch that are to be digitized, unless profiles have been defined.

Use of Optovars

If the device is equipped with Optovars, you may select the desired Optovar in the selection box beside *Optovar position*.

☞ These settings are only applicable if no profile has been defined. If profiles are used, the settings stored in the profiles will be used.

! Before initiating a digitization process, you should check for adequate memory space on the destination drive! Available capacity can simply be read in the last line (**Available disk space**) of the information field of *Current Slide Scan Progress*.

For the automatic mode, the use of profiles defined in the manual mode is recommended (section 5.3). You may select a predefined profile via the selection menu dealt with in this section. This profile will be loaded after clicking on Load. The profile name will appear in the window at the bottom on the left (e.g.: Profile name: HE_stain_std). If changes are made to settings which may be saved in the profiles (e.g. via Scan Options), the profile name will no longer appear at the bottom on the left, but the message ← *No Profile* → will be overlaid, showing that no profile exists for these settings. To accept these changed settings, you may save a new profile by clicking on the Save button.

Once all desired parameter settings and file-related entries have been made, a digitization process can be triggered by clicking on the 

button. In particular, the settings in the **Barcodes** tab have to be checked before starting (see section 5.4.8.2).

The first selected slide is pushed onto the specimen holder and positioned so that the preview camera will generate a preview image, which is a necessary precondition for selection of the areas to undergo digitization. Besides, the preview camera creates an image of the barcode area that may serve as a working input for barcode interpretation.

A running digitization process may be interrupted at any point by pressing the Stop Scan button.

Stop Scan will be disabled unless a digitization process was triggered.

If a running digitization process is interrupted by pressing the Stop Scan button, the slide which is being digitized at this moment will be placed back into its insert.

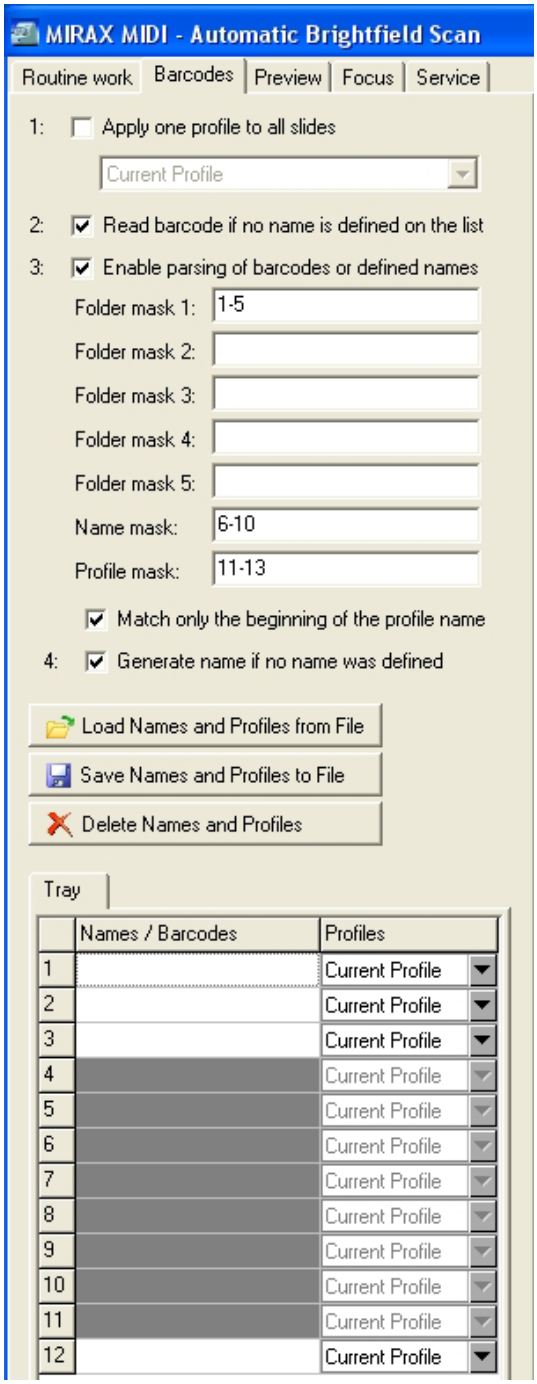


Fig. 5-20 Barcodes tab

5.4.8.2 Barcodes Tab

The **Barcodes** tab is intended for setting identification features to the various slides and for assigning profiles.

Four modes can be set in this tab:

1: If the option *Apply one profile to all slides* is active, one profile is used for all slides of a batch. The assignments in the lower part of the tab are disabled. *Current Profile* is the profile that has been loaded in the **Routine work** tab under *Scan Profiles*. You may, however, select any profile via the selection menu.

2: If the option *Read barcode if no name is defined on the list* is active, the barcode will be read and used as name for the virtual slide. This kind of name forming will take place only in the case that no name has been entered in the table in the lower half of the tab.

3: A barcode can be used to define the output directory, the name and the profile to be used. To do this, option 3 is enabled and the positions of the characters in the string (= barcode) are defined in the mask fields. It is possible to define areas using the character –. Furthermore, characters located at different places of the string may be combined by means of a comma.

Example:

Character string: MIRAX_MIDI_123

Mask: 1-5 → MIRAX

Mask: 7-10 → MIDI

Mask: 1-5,12-14 → MIRAX123

Example:

Barcode: Path_DoeJohn_003_FAST_HE

Path is the path for saving

DoeJohn is the name of the virtual slide

003_FAST_HE is the profile name

The barcode control for the example above would have to look like this:

Folder mask 1:	1-4
Folder mask 2:	<empty>
Folder mask 3:	<empty>
Folder mask 4:	<empty>
Folder mask 5:	<empty>
Name mask:	6-12
Profile mask:	14-24

Up to five subdirectories can be coded in a barcode (*Folder mask 1* … *Folder mask 5*). In administrating and creating profiles, the user will prefer descriptive names to recognize them easier (e.g.: FF04_CF08_HE_23). Coding of such profiles in the barcode requires a great part of the available space in the barcode. For this case, the option *Match only the beginning of the profile name* may be enabled. Then, the system will use only the first characters to identify a profile. The name may begin, for example, with an unequivocal number used as identification. The profile could be named, for example, 005_FF04_CF08_HE_23. You would then enter 1-3 into the profile mask and enable the option *Match only the beginning of the profile name*. Now, only the number 005 would be used to identify the profile. Make sure that only one profile name beginning with the characters 005 exists.

4: If no name has been defined in the table below and the option *Generate name if no name was defined* is active, a name will be generated automatically:

1M<slide position>

Example: 1M10 (10th slide position)

☞ The algorithm for automated number generation will assign identical names to slides in the same insert slot each time a given insert is refilled.

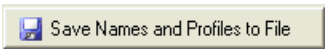
For this reason, data regarding digitized slides must be separately saved before loading new slides into the device. Otherwise, the contents of directories with identical names will be overwritten with the data of newly digitized slides without an acknowledgement request.

The above numbering from 1 to 4 represents the operating sequence (if the options were enabled).

Example: Items 2 and 4 are active. – First, the system will check whether a name has been entered into the table. If this is not the case, the barcode will be read (No. 2). If there is no barcode, a name will be given automatically (No. 4).

In the table below, the user may enter names into the column *Names / Barcodes* to identify the virtual slides. If several profiles shall be used, one profile for each slide can be defined via the selection menu (column: *Profiles*). If an insert was loaded, only those positions will be active for which a slide has been detected.

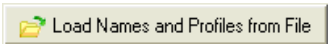
Current Profile represents the profile loaded in the **Routine work** tab under *Scan Profiles*.

The user may save the entered names and profile assignments by clicking the 

button. Before saving, the user may define a name. The information will be saved in the format *comma separated values (csv)*:

1;<position number>,<slide name>,<profile name>

This information can be imported into the EXCEL program, where the user may change the data and save them again in the csv format.

By clicking on the 

button, the user may import these changed data or other assignments again.

The 

button will be pressed to delete all entries contained in the profile assignment table.

☞ If a directory of the same name already exists in an automatic batch digitization operation, a name will be generated that will contain the original name, followed by the date and time of digitization.

5.4.8.3 Preview Tab

The **Preview** tab provides various options for defining the areas for masking (shown red/orange in software viewing screens). Only the masked areas will then be digitized with high resolution. The options are to be used in the same way as described for the manual mode (section 5.4.4.2).

☞ The use of profiles is recommended, as the automatic mode does not provide a preview image to check the settings.

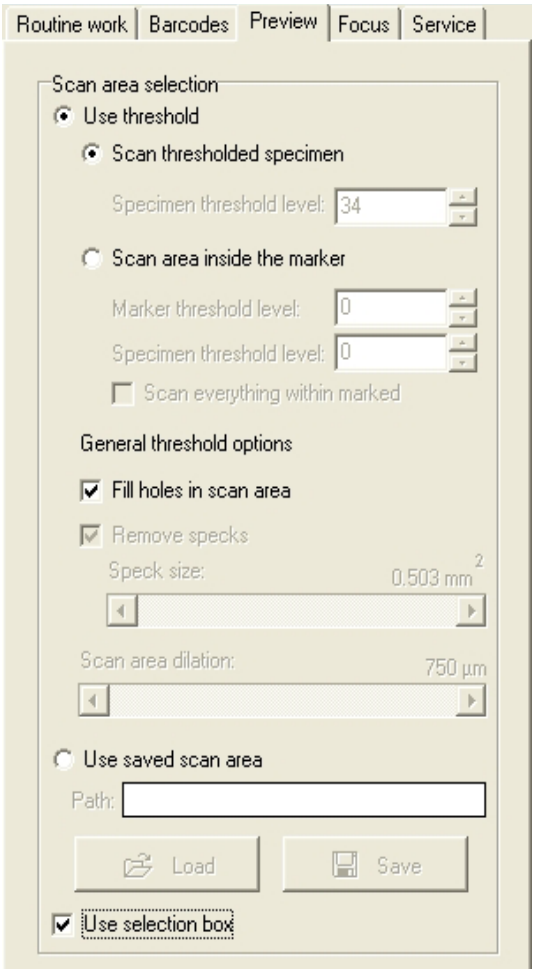


Fig. 5-21 Preview tab

5.4.8.4 Focus Tab

On selection of the **Focus** tab (Fig. 5-22), you may make online adjustments as required during a running digitization process.

Focus is the sub-area for setting restrictions to the focus range. If *Limit focus range* is active, you may define a lower focus range limit via *Lower limit* and an upper focus range limit via *Upper limit*, using the respective arrow keys.

This function may prove necessary if, as a result of a soiled coverslip surface, focusing is targeted on a faulty spot on the coverslip surface rather than the actual specimen you want to digitize.
Furthermore, the **Focus** tab may include a possibility to work with an *Extended focus mode* function. This extended focus functionality is provided as an option.

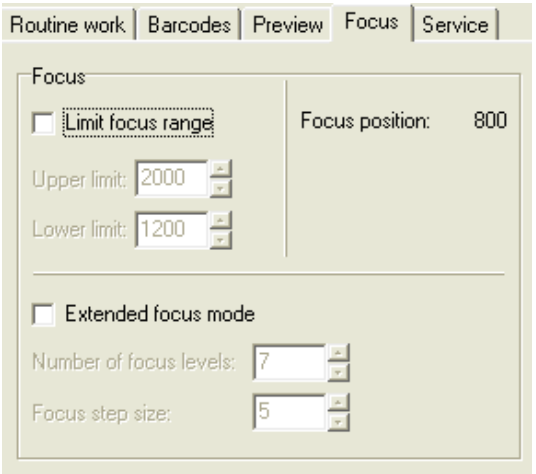


Fig. 5-22 Focus tab

If the function is active, several images will be captured in different focus planes for each image field. The regions of strongest contrast which belong to a given image are combined into an image that will

contain the maximum contrast portions of all images recorded for this image field. This method guarantees a maximum in depth of focus. While exerting no influence in terms of the file size, it does have an impact on digitization speed, because a set of images has to be shot and mathematically combined into a single image.

Once the increased depth of focus function was turned on via the *Extended focus mode* control box, you may determine a desired number of focus planes via the *Number of focus levels* selection box. The currently valid focus value (defined by z-value as determined by auto-focusing) is always interpreted as the starting point and point of reference. A value of "5" means that the resulting image will be calculated, using the current focus value, two over-focused images and two under-focused images as mathematical input. For this reason, you are advised to pick an uneven number for *Number of focus levels*.

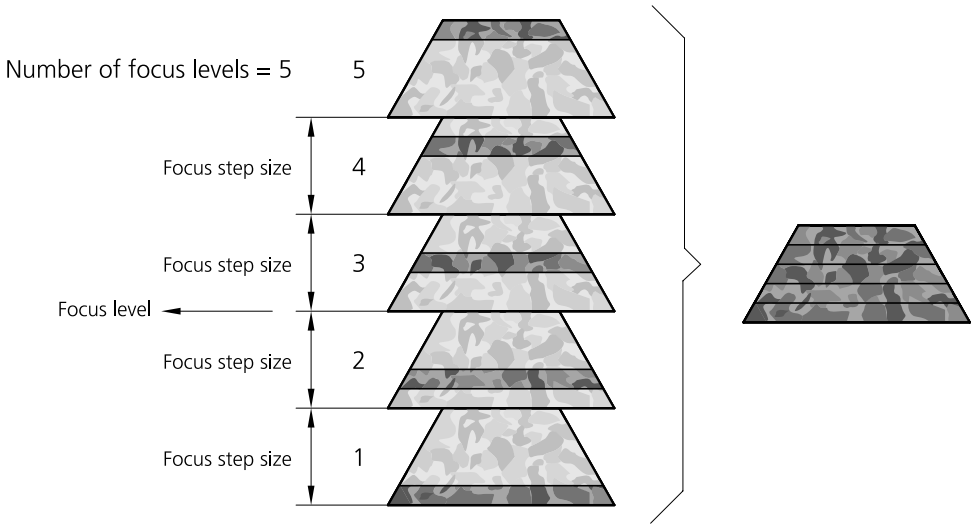


Fig. 5-23 Graphical view of *Extended focus* process (bright areas are unsharp zones, the darker areas are sharp zones)

The mutual spacing between the individual images in z-direction can be specified via *Focus step size*.

5.4.8.5 Service Tab

The **Service** tab allows you to enable certain control and maintenance functions.

If a jammed slide causes the digitization process to be interrupted, you may click the Emergency Manual Slide Removal Position button in order to transfer that slide to a position where it can be accessed for error removal. Action described in the fault finding and elimination procedure (refer to table on page 92 et seq.) should not be initiated before the slide has reached this position.

By pressing the Slide Removed Manually button, you can notify the software that a slide was removed manually.

If the *Show images during scanning* check box is turned on, the image field which is currently being digitized will appear in the display field 5-18/**5**.

☞ *Show images during scanning* should only be turned on in exceptional cases when you work in ***Automatic*** mode, because the digitization process will become slower as a consequence.

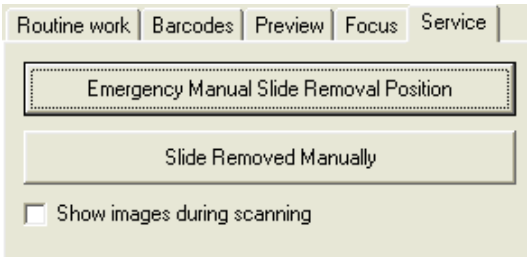


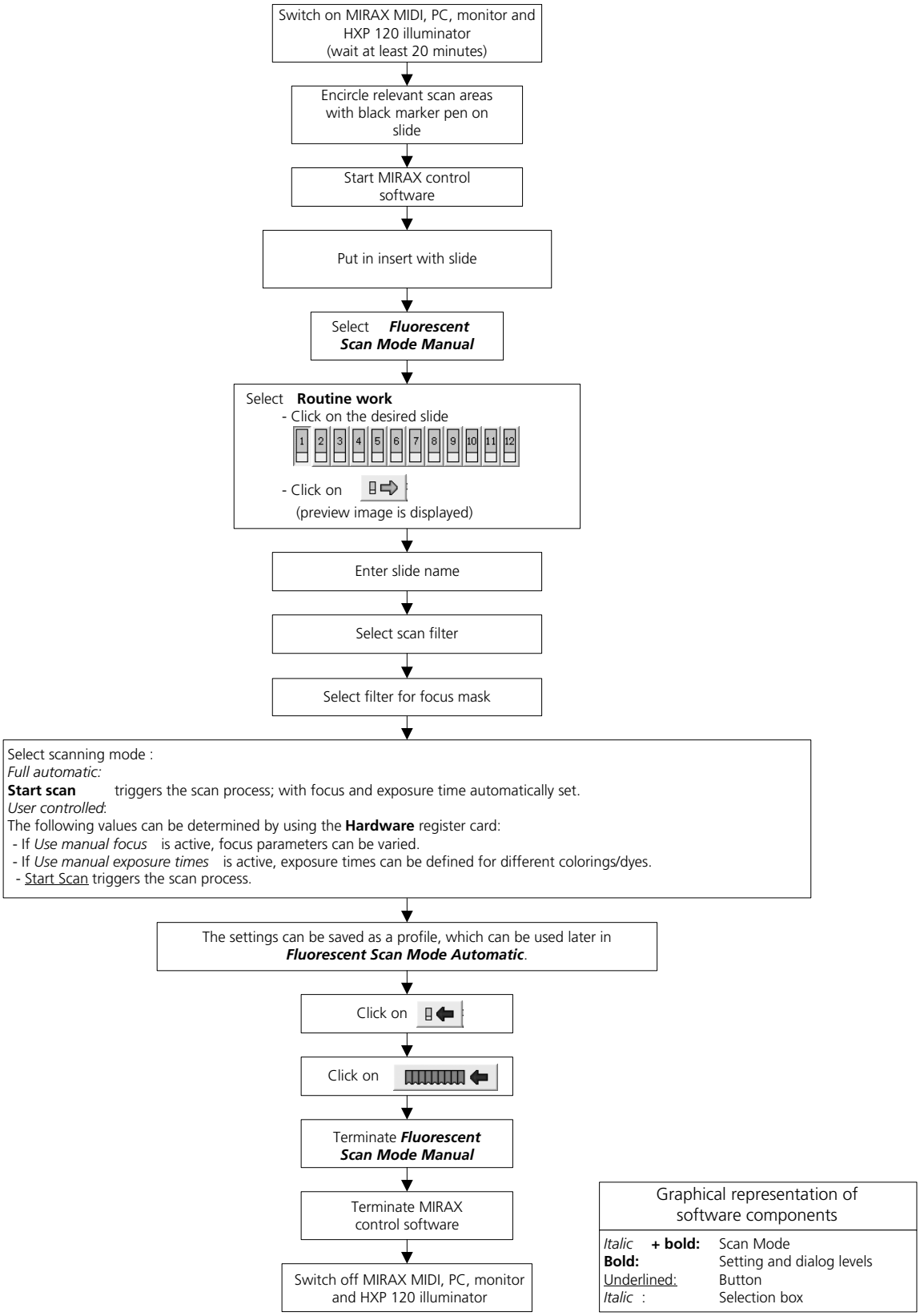
Fig. 5-24 Service tab

5.5 Fluorescence Applications (MIRAX MIDI BF/FL)

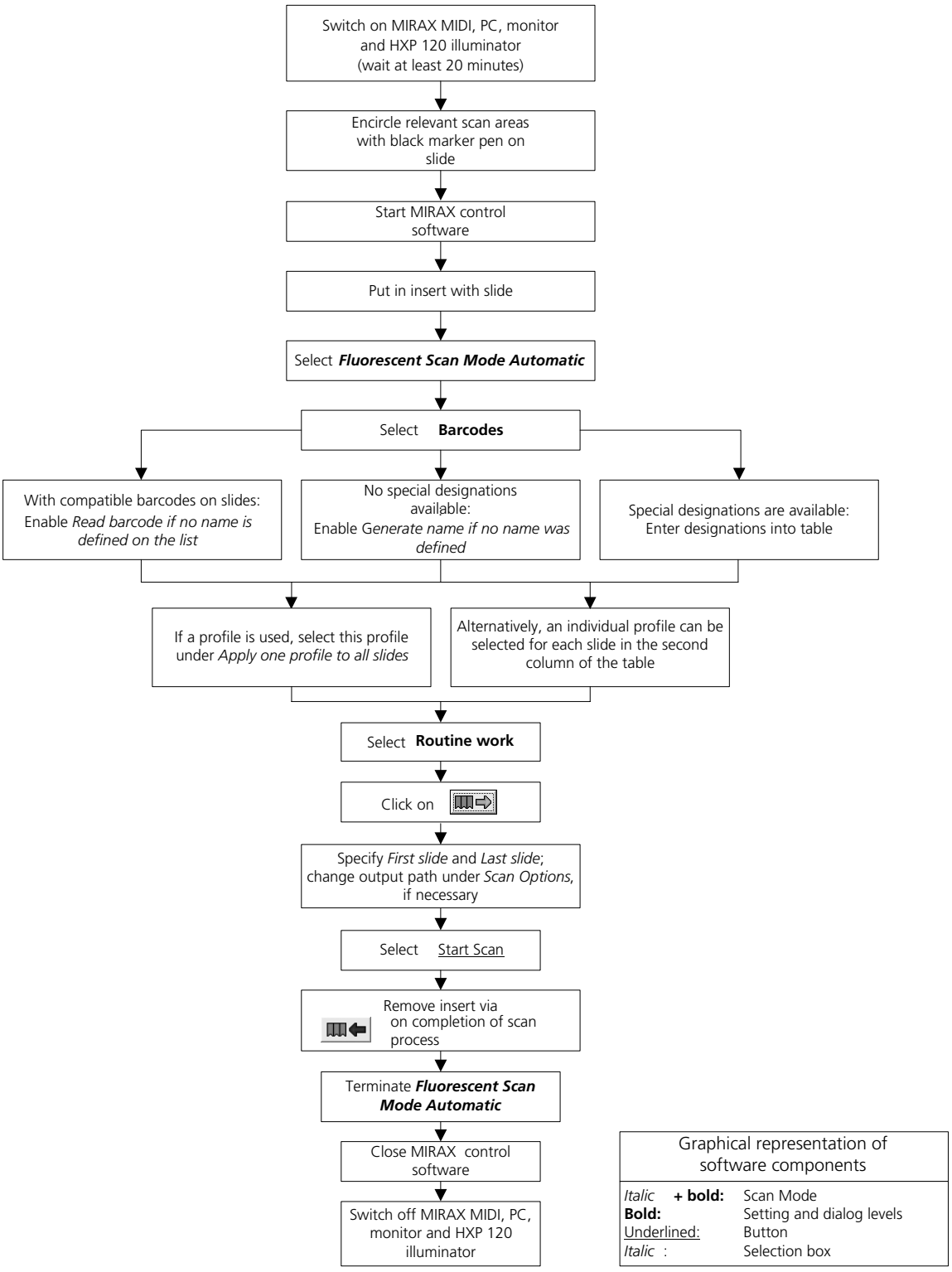
The operating screen for fluorescence application closely follows that of brightfield application.

Therefore the following section is intended to provide a survey of the Fluorescence workflow and to deal with the features that are specific to fluorescence (see sections 5.5.1 and 5.5.2).

5.5.1 Digitizing Slides in *Fluorescent Scan Mode Manual*



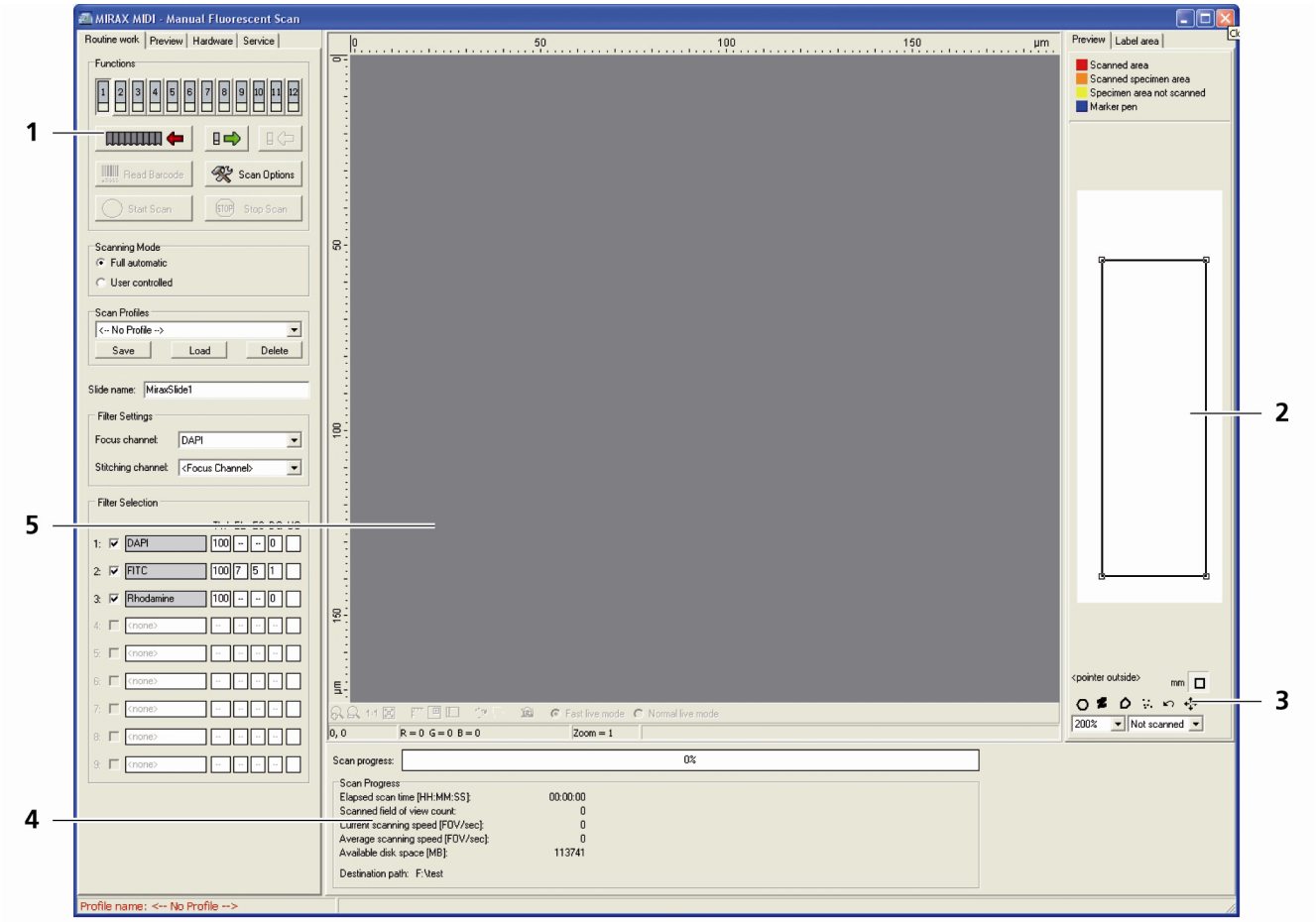
5.5.2 Digitizing Slides in *Fluorescent Scan Mode Automatic*



5.5.3 *Fluorescent Scan Mode Manual*

After a click on Manual in the *Fluorescent Scan Mode* selection area in the mode selection window, the ***Manual*** mode display and dialog window with the tabs (Fig. 5-25) will appear:

- **Routine work:** Basic scanner settings, loading and unloading of inserts and slides, administration of profiles, and triggering of the digitizing process
- **Preview:** Settings for preview images
- **Hardware:** Settings for focus (e.g.: limitation of focus range) and exposure time
- **Service:** Auxiliary functions for operation and maintenance



1 Dialog and function block with scanner functions and scanner settings
2 Preview image recorded with preview camera (areas to be digitized are shown in red/orange color)
3 Toolbar for editing of preview image and for manual positioning of slide in relation to optical axis
4 Sub-area informing about the progress of a running digitization
5 Live image and camera image display field

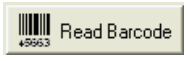
Fig. 5-25 ***Manua Fluorescent Scan*** **dialog and display window**

5.5.3.1 Routine work Tab

The **Routine work** tab is the input and dialog level for basic scanner functions.

The following data can be entered and functions initialized:

Functions

- With a click on a slide included in the symbol  , the selected slide will be moved to the transfer position.
- With a click on the symbol  , the selected slide will be transported from the insert to the specimen holder; a preview image is automatically generated and displayed.
- A compatible barcode on the slide will be captured automatically, once the  button has been actuated. It will also appear in the *Slide name* display box.
- You can start the scanning process with a click on the Start Scan button. A click on Stop Scan will terminate the scanning process.
- With a click on the symbol  , the slide will be moved back into the insert.
- With a click on the symbol  , the slide insert will be moved to a position where it can be taken out by the user.

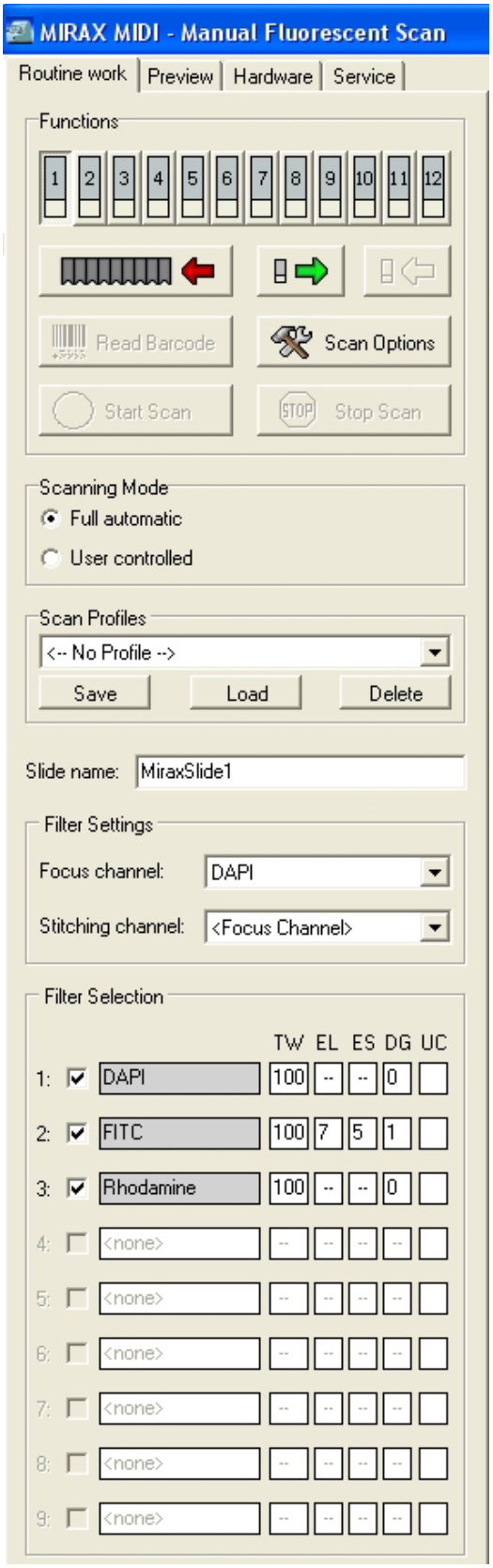


Fig. 5-26 Routine work tab

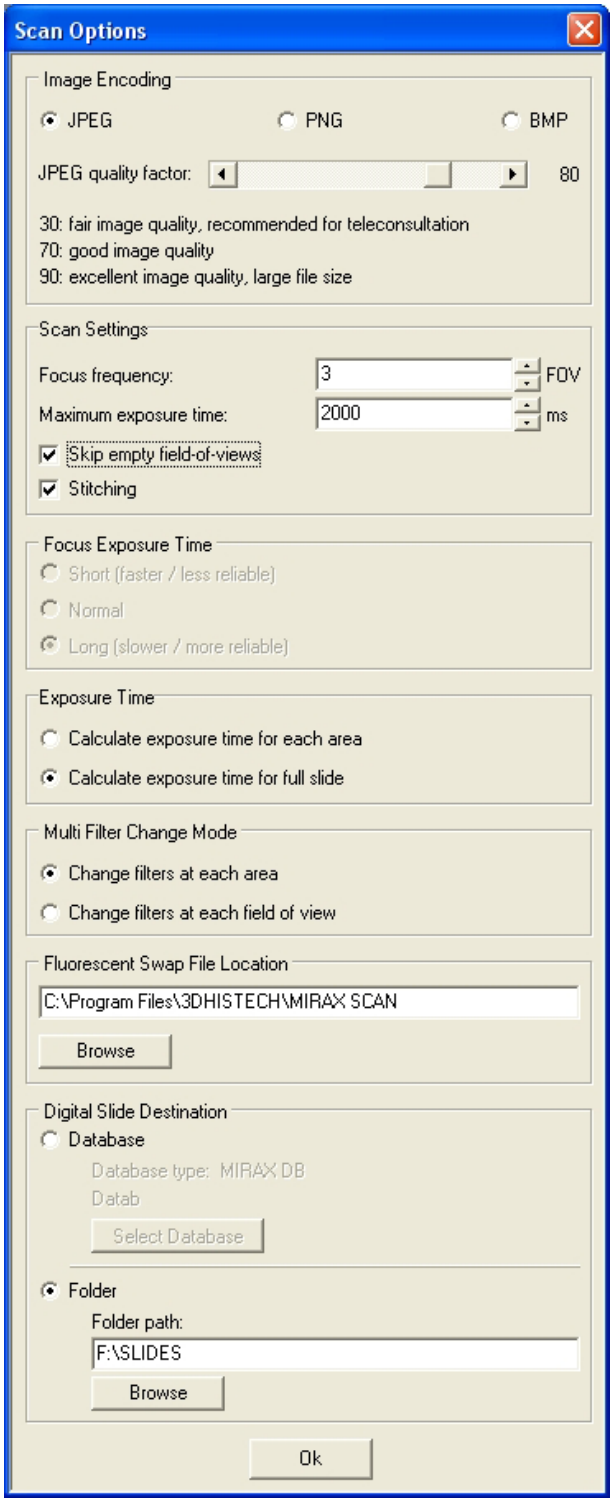
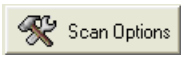


Fig. 5-27 Scan Options

A click on the 

button will open the **Scan Options** dialog window (Fig. 5-27).

The *Scan Options* selection window allows you to set parameters determining the image quality and other features.

The individual data items are explained in the following table:

Parameter	Explanations
Image encoding	Sets image format for individual image fields.
JPEG	Saves image fields as Joint Photographic Experts Group files (jpeg) with a certain degree of loss. The resulting quality can be adjusted with the *JPEG quality factor* slider button. Great values yield images of high image quality. Such files will, however, be quite big. Small values result in low-quality images and small files. The default setting of **80 %** provides the best possible compromise of image quality and file size, with smaller value settings (e. g. 60%) still yielding images of a high quality level.
PNG	Saves the various images as Portable Network Graphics files (png). This method guarantees loss-free saving of image information. PNG was raised to the standard procedure by W3C in 2003 (ISO standard ISO/IEC 15948:2003), also refer to BMP.
BMP	Provides loss-free saving of the various images as Windows bitmap files (bmp). Since the compression algorithm proves less effective with BMP than with PNG files, the resulting files are greater with the BMP option than with the PNG option.
Focus frequency	This value determines the number of image fields beyond which new focusing takes place. This value should be selected in dependency of the unevenness and size of the specimen.
Maximum exposure time	In this field, the maximum exposure value used by the software is determined. If the value is too high, digitization time will increase markedly.
Skip empty field-of-views	If this option is active and the first channel (focus channel) recognizes an image field without fluorescence signal, no snapshots will be taken for the remaining channels. The activation of this option is recommendable if many empty image fields are to be expected (e.g. TMA).
Stitching	By turning the *Stitching* function on, the user defines that the various tiles of a digitized slide will seamlessly stitch together in a subsequent MIRAX Viewer session with no perceivable positive or negative overlapping. If this function is active, digitization will automatically work with a greater overlap, which leads to increased digitization time (about 27% longer than with this function in disabled state). The resulting files will also be bigger by as much. When this function is deactivated, the shortest digitization time and the smallest file size are attained, but overlapping or underlapping may occur in the resulting image.

Parameter	Explanations
Focus Exposure Time	This field is used to define the length of time intended for determining the focus. Three settings are possible: Short: The algorithm applied allows speedy focusing. Depending on the specimen, the focus quality may be reduced thereby. This setting is particularly advisable for sensitive specimens with a tendency to bleach out. Normal: In this case the standard algorithm is used for focusing. It represents the optimum between focus quality and focus duration and is the best choice for the majority of specimens. Long: This algorithm produces the best focus quality, but requires a clearly longer time than Normal setting. This option should be selected only in the case of focusing problems with *Normal* setting and in the case of intense, strong fluorescent staining.
Exposure Time	This option is only taken into account if no exposure times were manually predefined! There are two possibilities - you may either have the exposure time determined for each camera image (*Calculate exposure time for each area*) or one value will be used for the complete slide (*Calculate exposure time for full slide*). It is recommended to use the second possibility.
Multi Filter Change Mode	This is to define, in the case of a multi-channel digitization, if filters are to be changed for each field of view (*Change filters at each field of view*) or if filters are to be changed after digitization of the slide (*Change filters at each area*). In order to optimize speed, the filter change should not take place until the slide has been digitized with one channel. If a very precise colocalization is required, filters should be changed for each field of view.
Fluorescent Swap File Location	In the course of the digitization process, temporary files up to a size of 60 Gbytes - depending on the settings - are created. Because of their size, these files should be stored on a partition that provides enough space, and this space should not be available for other processes as well (e.g. as memory location for digital slides).
Digital Slide Destination	
Database	This is to enter the required information if a database interfacing is used.
Folder	If this function is enabled, the virtual slides will be digitized into a directory to be specified. A Browse button is available for conveniently choosing a directory via the special selection window. If a non-existing path is specified, this path will be created by the software on acknowledgement.

Scanning Mode

Provides the user with a choice between ***Full automatic*** and ***User controlled*** mode. In ***Full automatic*** mode, focus and exposure time are automatically determined by the program.

The ***User controlled*** mode provides the user with the possibility to define focus and exposure times manually. The selection of the scanning mode influences the number of options in the **Hardware** tab.

Scan Profiles

The *Scan Profiles* field is used to administrate the profiles:

A click on Save will save the current settings (Fig. 5-28). The system will ask for a new profile name.

All existing profiles can be selected via the selection menu and enabled by clicking on the Load button. The current profile will be overlaid at the bottom on the left Current profile: Fluorescence_3_color.

The current profile will be deleted by clicking on the Delete button.

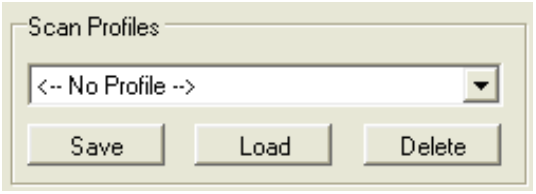


Fig. 5-28 Scan Profiles field

Filter Settings

The *Focus channel* function in this input field allows you to define the channel used for creating the focus map. It should be a channel with an exposure time as short as possible, but also with a sufficiently covered area. For cell nuclei stained with DAPI, e. g., select the blue channel.

In addition to that, you can define the channel to be used as master channel for the stitching of the images (*Stitching channel*).

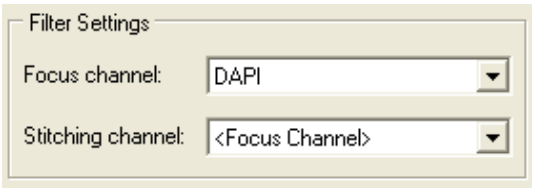


Fig. 5-29 Filter Settings field

☞ In the *Filter Settings* selection boxes you can select only channels that have been activated beforehand in the *Filter Selection* field.

Slide name:

This input field is available for manual entry of a desired slide name, under which the image data relating to this slide are to be saved.

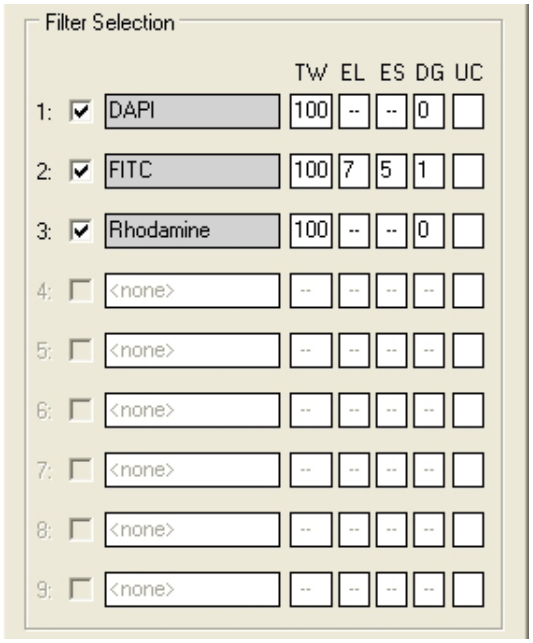


Fig. 5-30 Filter Selection field

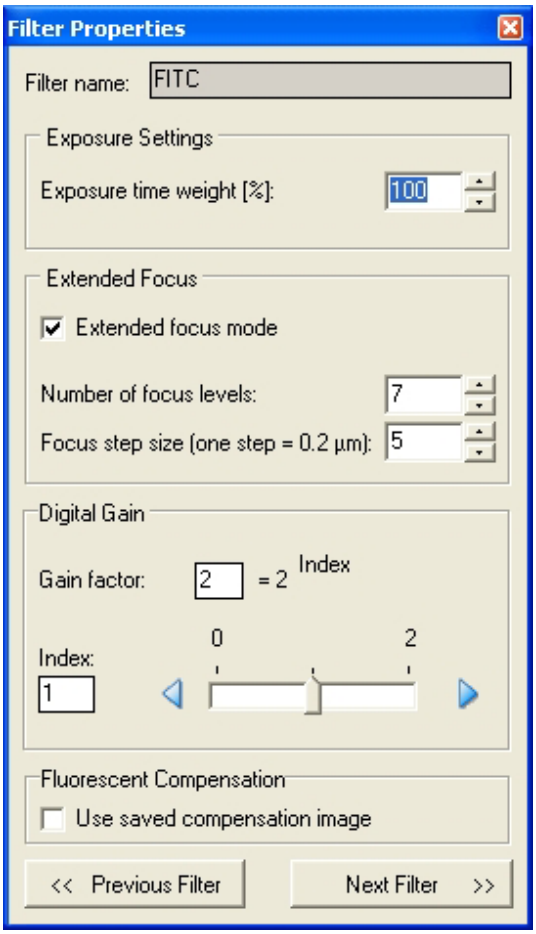


Fig. 5-31 Filter Properties

Filter Selection

In this input field you can select the filters from the list of all defined filters. Activate the filters which shall be used for the subsequent digitization process by marking the check boxes in front of the respective filters. In addition to that, further filter-specific properties can be assigned here. You can enter them directly or call them by means of a window (see Fig. 5-31) Click for this purpose onto the field behind the filter name.

The following properties can be defined:

Exposure time weight (**TW**): If the system determines the exposure time automatically, the brightest structure will be used as reference. This can result in an underexposed appearance of certain structures. This option allows you to define an appropriate percentage for increasing or reducing the automatically determined exposure time (a value of 140 % would increase the exposure time by 40 %).

Extended Focus: The Extended Focus function can be used to extend the depth of sharpness, provided this option has been installed. For a more detailed description see section 5.4.4.3. The number of focus planes is indicated under **EL** and the distance between the individual focus planes appears under **ES**.

Use saved compensation image (**UC**): If this option is activated, each image recorded is mathematically set off against a compensation image. A compensation image must be created for that (see 6.3.2.2.). Activation of this option is recommended to avoid shading artifacts in image fields.

Digital Gain (**DG**): Here, the index for digital gain can be entered if a camera capable of using digital gain (such as AxioCam MRm) is employed. If other cameras are used, no entry will be displayed. The dialog for digital gain is shown in Fig. 5-31.

If several filters have been activated, you can move to the previous or next filter properties by clicking the << Previous Filter and Next Filter >> buttons.

☞ Only the filters selected in this section can also be selected in the following tabs.

5.5.3.2 Preview Tab

The **Preview** register card offers the same options as in the brightfield mode (see 5.4.4.2).

For digitizing, use of the *Scan area inside the marker* option is recommended. The contrast is usually too weak for the specimens to be defined through the threshold value.

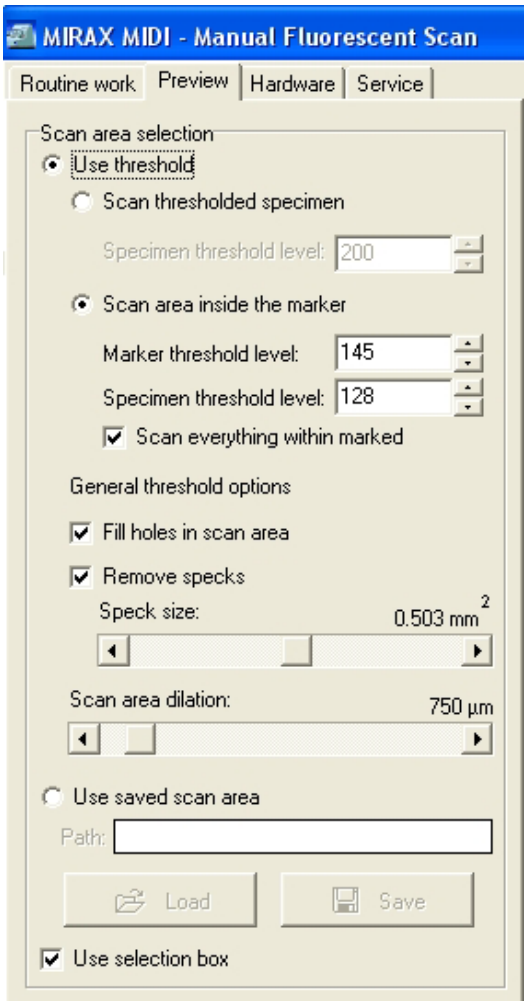


Fig. 5-32 Input field of the Preview tab

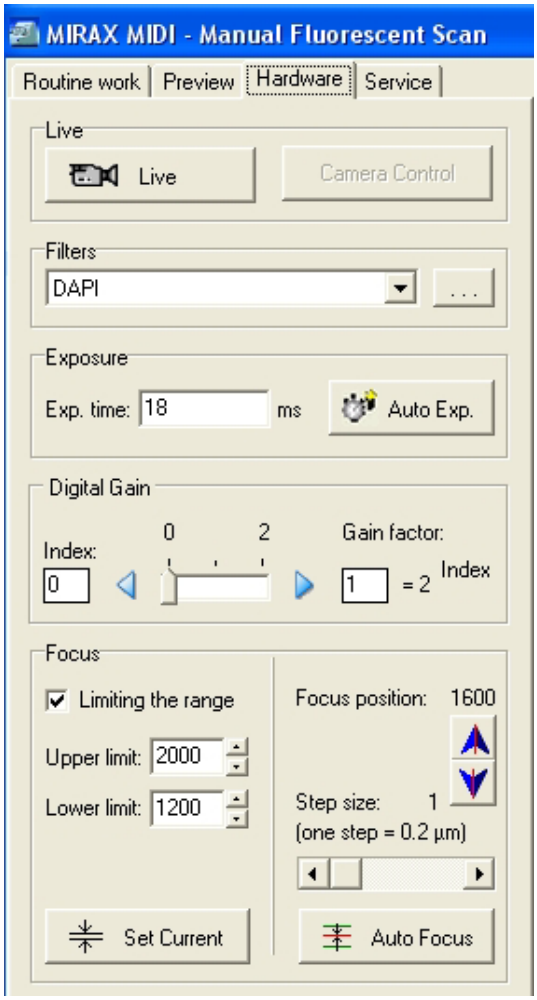


Fig. 5-33 Input area of the Hardware tab if the Full automatic mode is used

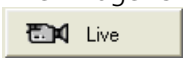
5.5.3.3 Hardware Tab

The layout and the number of possible settings depend on the *Scanning Mode* selection in the **Routine work** tab.

(1) Full automatic

If in the **Routine work** tab the *Full automatic* option has been selected under *Scanning Mode*, the exposure time and the focus will be determined automatically. For this reason, this tab is used to define basic focus properties or to explore the virtual slide manually.

First, a suitable filter has to be selected from the list of defined filters in the *Filters* selection box.

The live image is activated and displayed after clicking the 

button. Only after having clicked this button, the selected filter will be rotated into position. Otherwise, an empty space will be rotated into position and no light will fall on the specimen.

Even if the live image has been activated, the empty space will be moved into position again after 10 s, the last snapshot, however, will still be displayed.

If the camera supports the application of a digital gain function (e.g. AxioCam MRm), this gain can be determined by changing the index value in the *Digital Gain* panel:
(gain value = 2^{Index}).

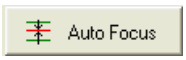
The resulting value is shown under *Gain factor*.

The *Exposure* input field allows you to enter a suitable exposure time or to have a suitable exposure time determined automatically by clicking on the 

button.

Focus is the sub-area for setting restrictions to the focus range. If *Limiting the range* is active, you may define a lower focus range limit via *Lower limit* and an upper focus range limit via *Upper limit*, using the respective arrow keys.

This function may prove necessary if, as a result of a soiled coverslip surface, focusing is targeted on a faulty spot on the coverslip surface rather than the actual specimen you want to digitize.
With the help of the Set current button you can set the *Lower limit* value to the current focus value minus the value **32** and the *Upper limit* value to the current focus value plus the value **32**.

Clicking on the 

button will initiate a focusing sequence in the image marker position.

The *Step size* selection box allows you to define a manual focus step size with the help of the two arrow buttons.

The blue vertical arrow buttons are intended for manual focusing. This will have no influence on the way slides are digitized in automated mode.

With the mechanical (cross) stage function (5-15/**9**) in active state, a slide can be viewed like under a microscope. As a necessary precondition for this, the 

button must also be active.

Furthermore, the **Focus** tab may include a possibility to work with an *Extended focus mode* function. This extended focus functionality is provided as an option. For a detailed description of this function see section 5.4.4.3.

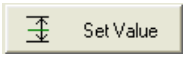
(2) User controlled

If in the **Routine work** tab the *User controlled* option has been selected under *Scanning Mode*, the user can preset the exposure time. This way, digitizing becomes reproducible (in the other case, the exposure time(s) is/are calculated separately for each slide). Furthermore, the time required for digitization is reduced (no calculation of the exposure time). The user may also preset one or several coarse focus values. This option is activated by enabling the *Use manual focus* control box. In spite of that, the system will always carry out a fine focusing routine.

☞ Activating this function implies the determination of a value, since the default value might be wrong and, in this case, focusing would not be performed correctly.

There are two ways to enter the coarse focus value. Before that, however, the live image must be turned on and a suitable exposure time must be selected.

Coarse focus:

This option is for setting a coarse focus value manually. First, the displayed image has to be set sharply. It should be used the same channel as will be used later for the focus card. The current z-value will be accepted by clicking on the 

button. This option is recommended for very flat specimens.

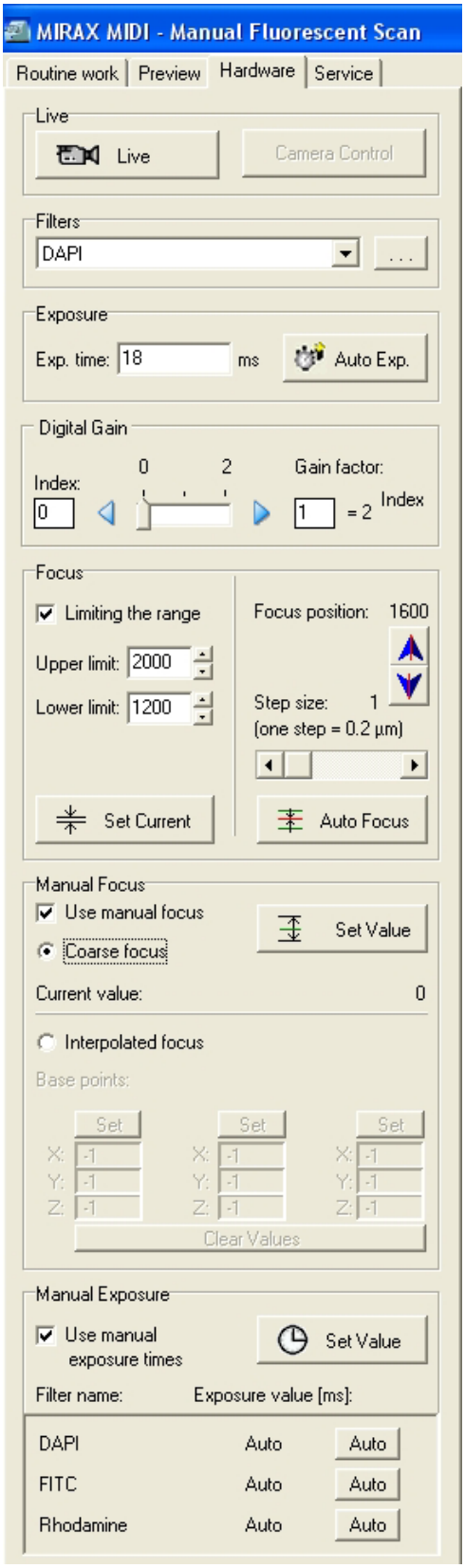
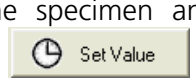


Fig. 5-34 Input area of the Hardware tab, if the User controlled mode is used

Interpolated focus:

If this option is selected, the coarse focus level will be calculated from three different focus points of the object. To do so, a manual focusing sequence of the object must be carried out at three points. Once the correct focus value has been determined, it can be saved with a click on the Store button. To delete the saved values, a click on the Clear All button is required. The software will then calculate a coarse focus level from the three values determined this way as a basis for the fine focusing routine. The user can only define the start parameters here, which, however, will increase the focus speed.

The enabled *User controlled* option allows the user, apart from the focus value setting, also the determination of the exposure times. For this purpose, the *Use manual exposure times* control box has to be activated.

The values are typically set on *Auto*, i.e., the exposure times are determined automatically. Presetting a value is only possible after having selected the corresponding filter and activated the live image. An exposure value will be determined in the field under *Exposure*. It is recommended to go to various positions in order to determine the intensity distribution throughout the specimen and define a representative value. This value will be accepted for presetting by clicking the 

button. This procedure must be repeated for all other filters.

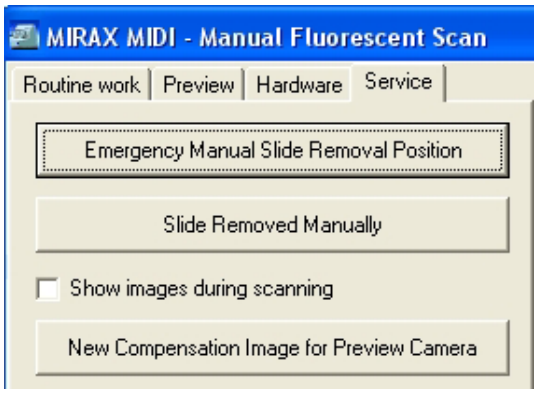


Fig. 5-35 Service tab

5.5.3.4 Service Tab

The **Service** tab allows you to enable certain control and maintenance functions.

If a jammed slide causes the digitization process to be interrupted, you may click the Emergency Manual Slide Removal Position button in order to transfer that slide to a position where it can be accessed for error removal. Action described in the fault finding and elimination procedure (see pages 91 et. seq.) should not be initiated before the slide has reached this position.

By pressing the Slide Removed Manually button, you can notify the software that a slide was removed manually.

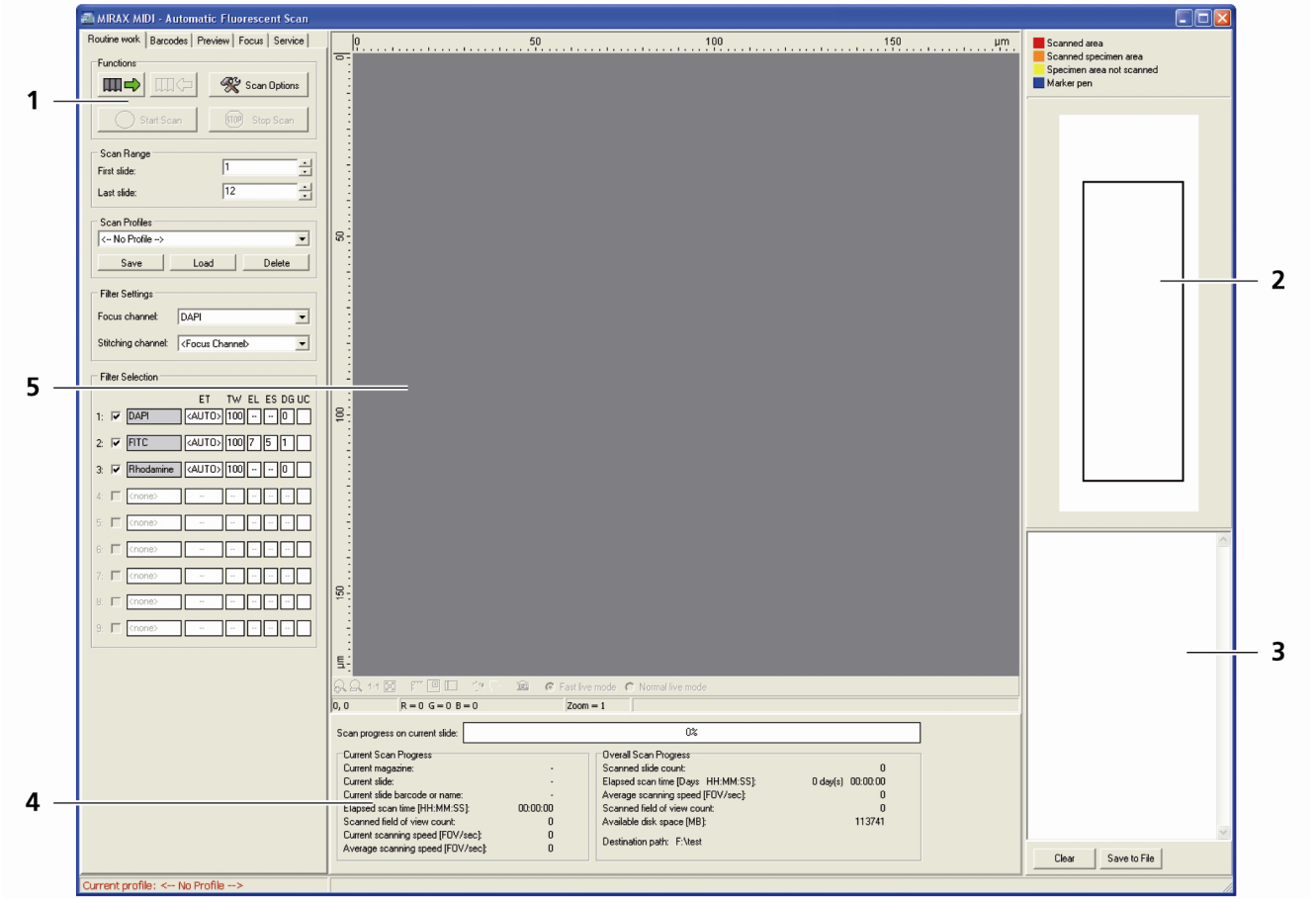
If the *Show images during scanning* check box is turned on, the image field which is currently being digitized will appear in the display field 5-18/**2**.

☞ *Show images during scanning* should be activated only in exceptional cases, as it slows down the digitization process.

5.5.4 *Fluorescent Scan Mode Automatic*

After a click on Automatic, the display and dialog window for ***Automatic*** operation will open with these tabs (Fig. 5-36):

- **Routine work:** Provides fundamental scanner settings and initiates scan process
- **Barcodes:** Setting and input options for identification of slides and assignment of profiles
- **Preview:** Settings for optimized preview images
- **Focus:** Settings for focusing (e.g. limitation of focus range)
- **Service:** Auxiliary functions to support operation control and maintenance



1 Dialog and function block for scanner functions and scanner settings
2 Preview image as recorded by preview camera (areas subject to digitization are shown in red color)
3 Logsheet window with digitization process data
4 Information field showing latest state of digitization process of currently processed slide and overall progress state
5 Display field for camera images

Fig. 5-36 *Automatic Fluorescent Scan* dialog and display window

A click on the cross in the title bar of the window will terminate ***Automatic*** mode operation. The mode selection window will turn active again. A slide which may be contained in the specimen holder at this moment will automatically be pushed back into the insert.

As the MIRAX control software offers the option to use predefined profiles, you should make use of it.

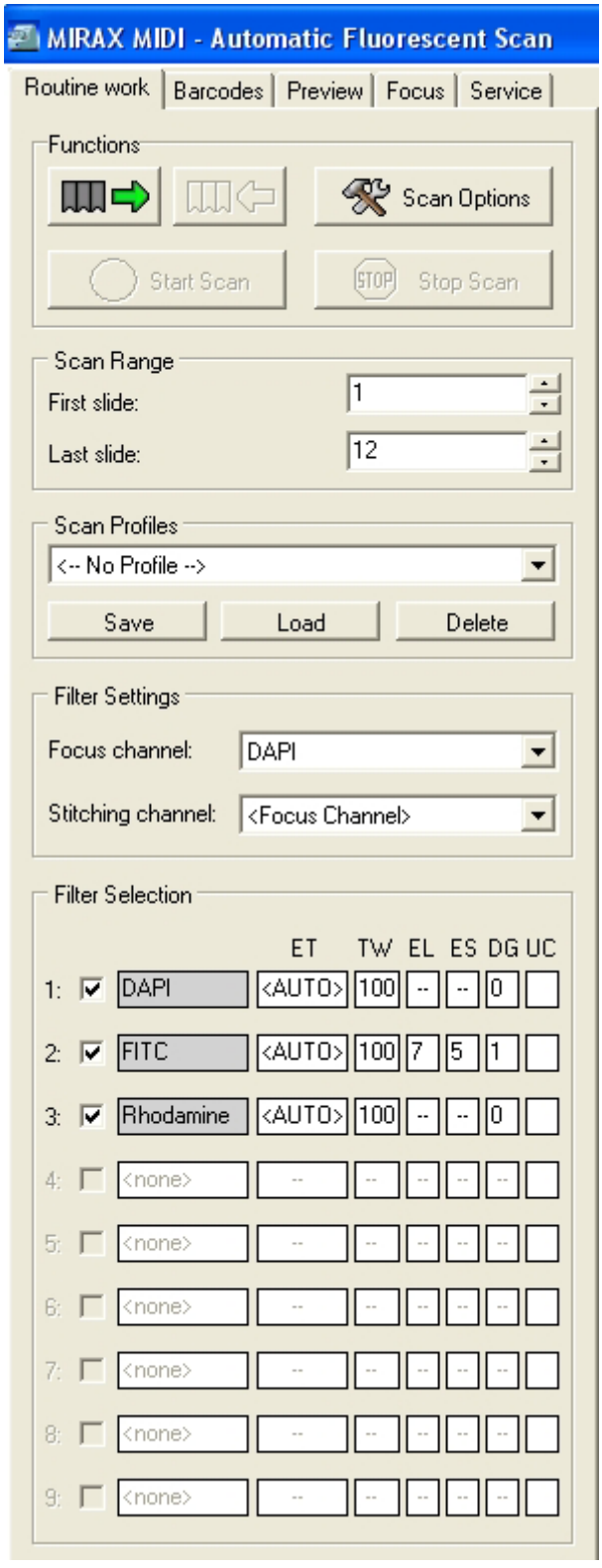


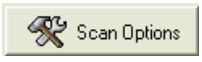
Fig. 5-37 Routine Work input and dialog level

5.5.4.1 Routine work Tab

Routine work designates the tab with fundamental digitization functions.

The following data can be entered and functions triggered here:

- First, the insert must be loaded by clicking on the  button. During the loading process the system is checking the positions for slides.
- Specification of first slide of the batch to be digitized *(First slide*).
- Specification of last slide of the batch to be digitized (*Last slide*).
- With a click on the  button the insert will be moved to a position where it can be taken out by the customer. Slides possibly situated in the specimen holder are moved beforehand into the insert.

By clicking on the 

button, you can open the **Scan Options** dialog window (see section 5.5.3.1). It provides setting options for the digitization process. These setting values will be valid for all slides of a batch, which are to be digitized, unless profiles have been defined.

Before initiating a digitization process, you should check for adequate memory space on the destination drive! Available capacity can simply be read in the last line (**Available disk space**) of the information field of *Current Slide Scan Progress*.

For the automatic mode, the use of profiles defined in the manual mode is recommended (section *Scan Profiles*). You may select a predefined profile via the selection menu dealt with in this section. This profile will be loaded after clicking on Load. The profile name will appear in the window at the bottom on the left (e.g.: Current profile: Fluorescence_3_color).

Filter Settings

The *Focus channel* function in this input field allows you to define the channel used for creating the focus map. It should be a channel with an exposure time as short as possible, but also with a sufficiently covered area. For cell nuclei stained with DAPI, e. g., select the blue channel.

In addition to that, you can define the channel to be used as master channel for the stitching of the images.

☞ In the *Filter Settings* selection boxes you can select only channels that have been activated beforehand in the *Filter Selection* field.

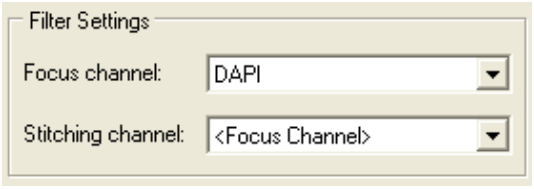


Fig. 5-38 Filter Settings field

Filter Selection

In this input field you can select the filters from the list of all defined filters. Activate the filters which shall be used for the subsequent digitization process by marking the check boxes in front of the respective filters. In addition to that, further filter-specific properties can be assigned here. You can enter them directly or call them by means of a window (see Fig. 5-39). Click for this purpose onto the field behind the filter name.

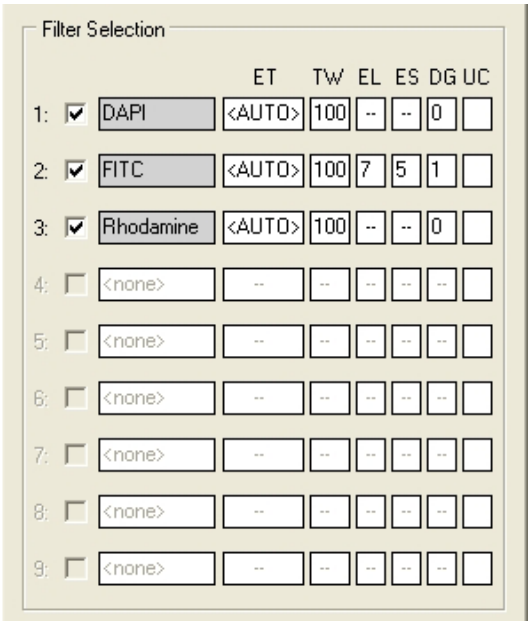


Fig. 5-39 Filter Selection field

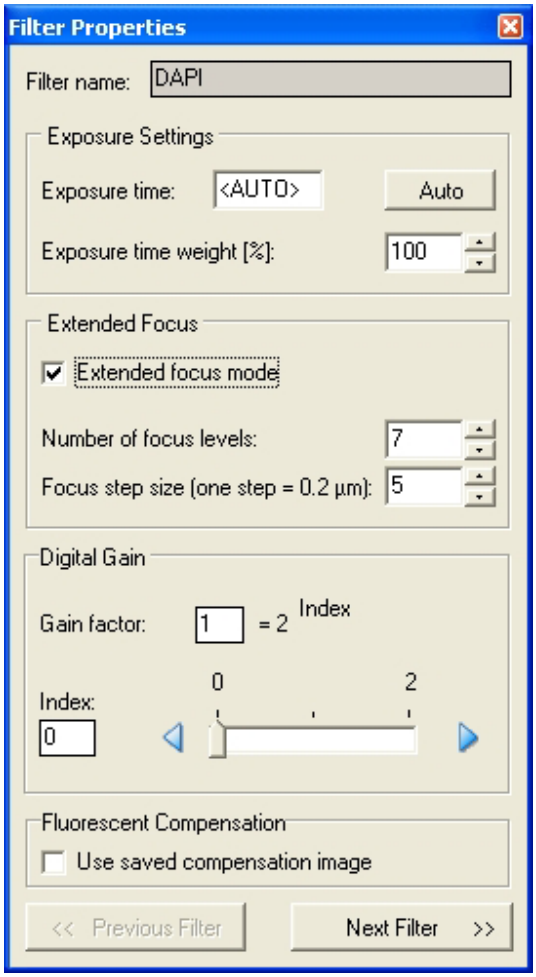


Fig. 5-40 Filter Properties

The following properties can be defined:

Exposure time (**ET**)*:* This field is used to enter the desired exposure time, which is indicated in ms. If <Auto> has been entered, the exposure time will be determined automatically. <Auto> will be entered by clicking the **Auto** button.

Exposure time weight (**TW**): If the system determines the exposure time automatically, the brightest structure will be used as reference. This can result in an underexposed appearance of certain structures. This option allows you to define an appropriate percentage for increasing or reducing the automatically determined exposure time (a value of 140 % would increase the exposure time by 40 %).

Extended Focus: The Extended Focus function can be used to extend the depth of sharpness, provided this option has been installed. For a more detailed description see section 5.5.4.3. The number of focus planes is indicated under **EL** and the distance between the individual focus planes appears under **ES**.

Digital Gain (**DG**): If the camera used has a digital gain function, you can enter here the index for the digital gain.

Use saved compensation image (**UC**): If this option is activated, each image recorded is mathematically set off against a compensation image. A compensation image must be created for that (see 6.3.2.2.). Activation of this option is recommended to avoid shading artifacts in image fields.

If several filters have been activated, you can move to the previous or next filter properties by clicking the << Previous Filter and Next Filter >> buttons.

☞ Only the filters selected in this section can also be selected in the following tabs.

If changes are made to settings that may be saved in the profiles (e.g. via Scan Options), the profile name will no longer appear at the bottom on the right, but the message ← *No Profile* → will be overlaid, showing that no profile exists for these settings. To accept these changed settings, you may save a new profile by clicking on the Save button.

☞ These settings are applicable only if no profile has been defined for the digitization process. If profiles are used, the settings stored in the profiles will be applied.

Once all desired parameter settings and file-related entries have been made, a digitization process can be triggered by clicking on the 

button. In particular, the settings in the **Barcodes** tab (section 5.5.4.2) have to be checked before starting. For starting, continue as explained in section 5.4.8.1.

5.5.4.2 Barcodes Tab

The **Barcodes** tab is intended for setting identification features to the various slides and for assigning profiles.

Four modes can be set in this tab:

1: If the option *Apply one profile to all slides* is active, one profile is used for all slides of a batch. The assignments in the lower part of the tab are disabled. *Current Profile* is the profile that has been loaded in the **Routine work** tab under *Scan Profiles*. You may, however, select any profile via the selection menu.

2: If the option *Read barcode if no name is defined on the list* is active, the barcode will be read and used as name for the virtual slide. This kind of name forming will take place only in the case that no name has been entered in the table in the lower half of the tab.

3: A barcode can be used to define the output directory, the name and the profile to be used. To do this, option 3 is enabled and the positions of the characters in the string (= barcode) are defined in the mask fields. It is possible to define areas using the character –. Furthermore, characters located at different places of the string may be combined by means of a comma.

Example:

Character string: MIRAX_MIDI_123

Mask: 1-5 → MIRAX

Mask: 7-10 → MIDI

Mask: 1-5,12-14 → MIRAX123

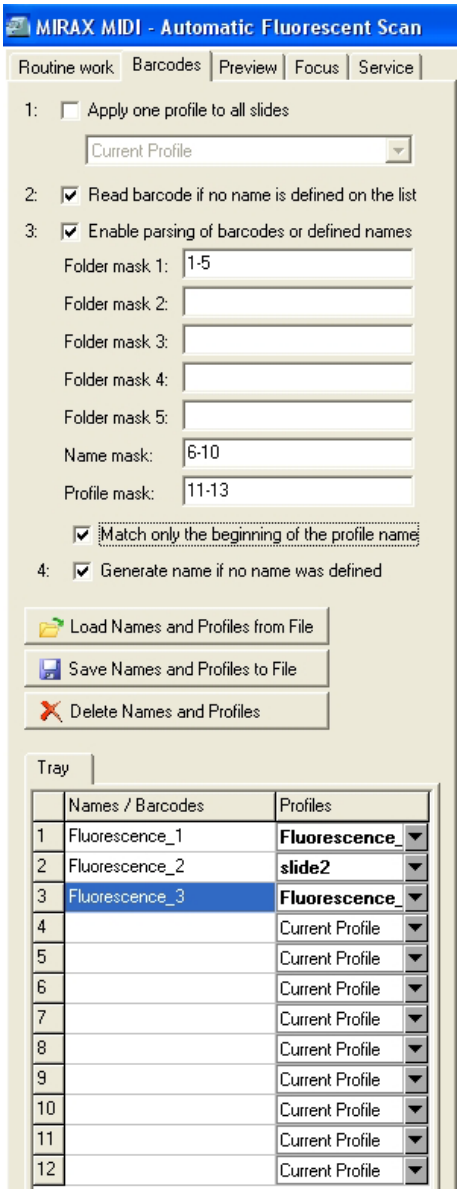


Fig. 5-41 Barcodes input and dialog level

Example:

Barcode: Path_DoeJohn_003_FAST_HE

Path is the path for saving

DoeJohn is the name of the virtual slide

003_FAST_FL is the profile name

The barcode control for the example above would have to look like this:

Folder mask 1:	1-4
Folder mask 2:	<empty>
Folder mask 3:	<empty>
Folder mask 4:	<empty>
Folder mask 5:	<empty>
Name mask:	6-12
Profile mask:	14-24

Up to five subdirectories can be coded in a barcode (*Folder mask 1* … *Folder mask 5*). In administrating and creating profiles, the user will prefer descriptive names to recognize them easier (e.g.: FF04_CF08_HE_23). Coding of such profiles in the barcode requires a great part of the available space in the barcode. For this case, the option *Match only the beginning of the profile name* may be enabled. Then, the system will use only the first characters to identify a profile. The name may begin, for example, with an unequivocal number used as identification. The profile could be named, for example, 004_F04_3C_26. You would then enter 1-3 into the profile mask and enable the option *Match only the beginning of the profile name*. Now, only the number 004 would be used to identify the profile. Make sure that only one profile name beginning with the characters 004 exists.

4: If no name has been defined in the table below and the option *Generate name if no name was defined* is active, a name will be generated automatically:

1M<slide position>

Example: 1M10 (10th slide position)

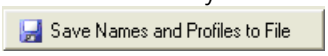
☞ The algorithm for automated number generation will assign identical names to slides in the same insert slot each time a given insert is refilled.

The above numbering from 1 to 4 represents the operating sequence (if the options were enabled).

Example: Items 2 and 4 are active. – First, the system will check whether a name has been entered into the table. If this is not the case, the barcode will be read (No. 2). If there is no barcode, a name will be given automatically (No. 4).

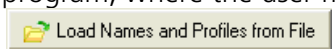
In the table below, the user may enter names into the column *Names / Barcodes* to identify the virtual slides. The insert can be selected by using tabs. If several profiles shall be used, one profile for each slide can be defined via the selection menu (column: *Profiles*). If an insert was loaded, only those positions will be active for which a slide has been detected.

Current Profile represents the profile loaded in the **Routine work** tab under *Scan Profiles*.

The user may save the entered names and profile assignments by clicking on the 

button. Before saving, the user may define a name. The information will be saved in the format *comma separated values (csv)*:

1;<position number>;<slide name>;<profile name>

This information can be imported into the EXCEL program, where the user may change the data and save them again in the csv format. By clicking on the 

button, the user may import these changed data or other assignments again.

The 

button will be pressed to delete all entries contained in the profile assignment table.

☞ If a directory of the same name already exists in an automatic batch digitization operation, a name will be generated that will contain the original name, followed by the date and time of digitization.

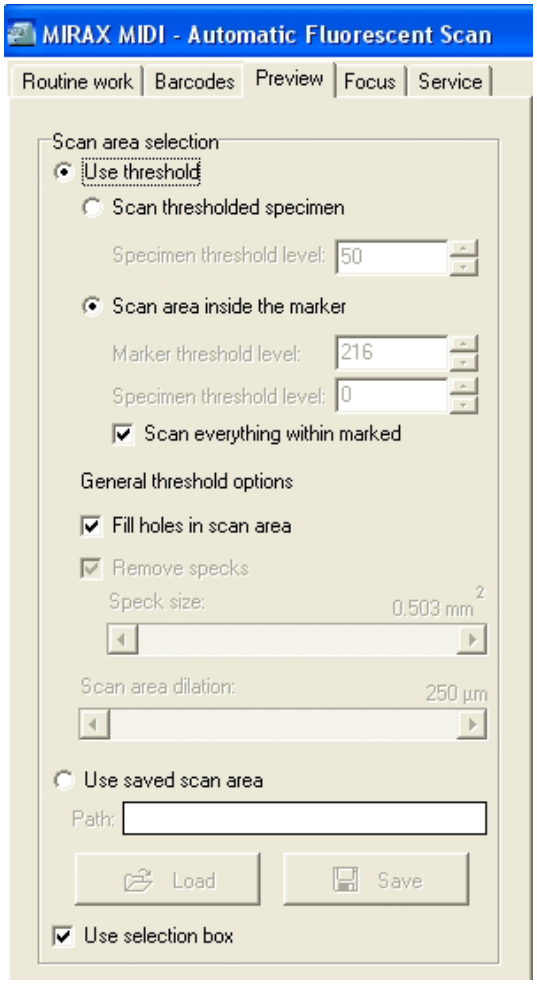


Fig. 5-42 Input field of the Preview tab

5.5.4.3 Preview Tab

The **Preview** register card offers the same options as in the brightfield mode (see 5.4.4.2).

For digitizing, use of the *Scan area inside the marker* option is recommended. The contrast is usually too weak for the specimens to be defined through the threshold value.

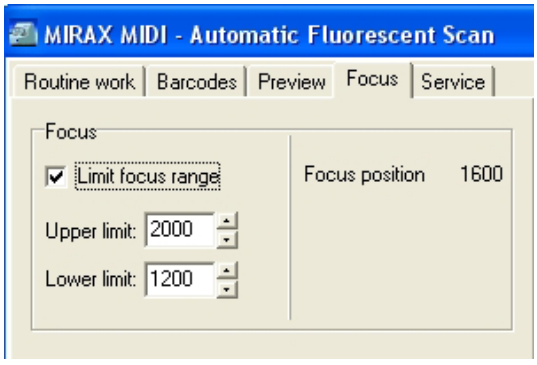


Fig. 5-43 Input field of the Focus tab

5.5.4.4 Focus Tab

On selection of the **Focus** tab (Fig. 5-43), you may make online adjustments as required during a running digitization process.

Focus is the sub-area for setting restrictions to the focus range. If *Limit focus range* is active, you may define a lower focus range limit via *Lower limit* and an upper focus range limit via *Upper limit*, using the respective arrow keys.

This function may prove necessary if, as a result of a soiled coverslip surface, focusing is targeted on a faulty spot on the coverslip surface rather than the actual specimen you want to digitize.

5.5.4.5 Service Tab

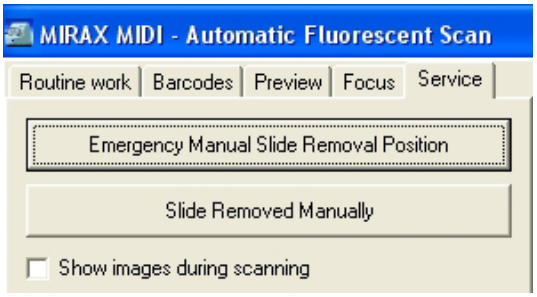


Fig. 5-44 Input field of the Preview tab

The **Service** tab allows you to enable certain control and maintenance functions.

If a jammed slide causes the digitization process to be interrupted, you may click the Emergency Manual Slide Removal Position button in order to transfer that slide to a position where it can be accessed for error removal. Action described in the fault finding and elimination procedure (see page 92 et seq.) should not be initiated before the slide has reached this position.

By pressing the Slide Removed Manually button, you can notify the software that a slide was removed manually.

If the *Show images during scanning* check box is turned on, the image field which is currently being digitized will appear in the display field 5-18/**2**.

☞ *Show images during scanning* should only be turned on in exceptional cases when you work in ***Automatic*** mode, because the digitization process will become slower as a consequence.

6 PREVENTIVE MAINTENANCE, TROUBLE SHOOTING, ADJUSTMENTS AND SERVICE

Switch off the device and disconnect power supply before beginning any maintenance work. Whenever working on the device take care that no liquid gets into it.

6.1 Preventive Maintenance (Base Unit)

Preventive maintenance for the MIRAX MIDI is limited to the following work items:

- Turn power off after each MIRAX MIDI operating session and put on protective cover (to prevent settling of dust or penetration of moisture).
- Do not install the MIRAX MIDI in a damp room. Max. allowed humidity: $\leq$ 75 %.
- Cover exposed tubes with dust protection caps.
- Breathe on water-soluble dirt (coffee, cola, etc.) and wipe it off with a lint-free cotton patch or a piece of cloth slightly moistened with water, to which a soft detergent may be admixed.
- Use a cotton swab or a lint-free piece of cotton cloth together with special L optics cleaning solution to wipe off stubborn oily or greasy dirt.
  This cleaning mixture consists of 90 volume% of gasoline and 10 volume% of isopropanol (IPA). These components are also known as:
  Gasoline: surgical spirit, petroleum ether
  Isopropanol: 2-Propanol,
  Dimethylcarbinol,
  2-Hydroxypropane

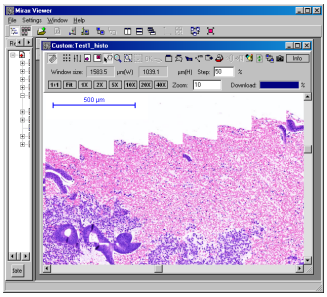
For operation in moist and warm climatic zones, you should make sure that the MIRAX MIDI is installed in bright, dry and properly ventilated rooms with $\leq$ 75 % air humidity.

Precision mechanical and optical instruments are threatened by fungal contamination of their surfaces if operating in any of the following environments:

- Relative air humidity > 75% prevailing for more than three days with temperatures between +15° C and +35° C.
- Dark rooms without air flow.
- Sedimentation of dust and fingerprints on optical surfaces.

6.2 Trouble Shooting

Problem	Cause	Action for error removal
The MIRAX MIDI does not work.	Electric power supply broken	24 V power supply unit is not on. • Turn power supply unit on.
		Control computer is not on. • Turn control computer on.
		• Check cable connections.
Control program has been interrupted.	Slide jammed	• Click Emergency Manual Slide Removal Position button to move slide into initial position. • Terminate control program session. • Turn control computer and 24 V power supply off and disconnect power plug. • Push protective covers apart (see 0). • Remove slide manually (see 6.2.5). • Push protective covers together again. • Reconnect 24 V power supply unit and control computer to line power supply. • Turn 24 V power supply unit on. • Turn control computer on. • Launch control program.
	Insert jammed	• Terminate control program session. • Turn control computer and 24 V power supply off. Disconnect power plug. • Push protective covers apart (see page 88). • Remove insert manually. • If a jammed insert cannot be removed, you should contact service personnel. • Push protective covers together again. • Reconnect 24 V power supply unit and control computer to line power supply. • Turn 24 V power supply unit on. • Turn control computer on. • Launch control program.
	Name of currently selected slide for digitization already exists and is open in MIRAX Viewer	• Close virtual slide in MIRAX Viewer.

Error message during scan camera calibration	Halogen lamp defective	• Replace halogen lamp (see 6.2.3).
Image screen dark		
Virtual slide improperly focused almost across its entire surface.	Focus range enabled	Disable *Limiting focus range*.
Poor image quality	Imaging errors (e.g. chromatic aberration)	• Contact service.
Virtual slides in MIRAX Viewer exhibit poor stitching: 	Scan camera not properly adjusted (e. g. due to loose fixture)	• Proceed as explained in section 6.3.4 • Contact service.
Slide is often positioned with fault.	Mech. guide error in transporting system	• Contact service.
Preview image is too bright (flare/glare effects) or too dark.	Invalid compensation image for preview camera	• Record new compensation image for preview camera (see section 5.4.4.4)
Digitization process is extremely slow	*Extended focusing* option is on	• Turn *Extended focusing* option off if not required.
Blurred image in ***Manual*** mode	Focus range is on	• Turn *Limiting focus range* off (see Fig. 5-10).
	False focus range setting	• Change focus range setting.
All single images exhibit a shading effect (brightfield)	Algorithm failed to find empty image fields	• Record reference image with empty slide (see section 5.4.8.1, *Use saved compensation image*)
All single images exhibit a shading effect (fluorescence)	No compensation image is activated.	• Determine and activate the compensation image for the fluorescence channel used (see 6.3.2.2.).
Error message that no empty image can be found	Algorithm cannot find appropriate empty image fields	• Record reference image with empty slide (see section 6.3.2.1) and activate it.

Fluorescence Applications (optional)		
The shutter of the HXP120 cannot be triggered, in spite of trigger connection via AxioCam MRm.	HXP 120: Shutter is permanently open.	• Activate shutter remote control with the button at the front of the HXP120.
No suitable exposure time can be found. Signal is too weak.	HXP 120: Shutter is closed. Intensity is too low.	• Deactivate the remote control of the shutter via the button on the front of the HXP120. • Increase intensity with the rotary button at the front of the HXP 120.
	HXP 120 is switched off.	• Switch light source on.

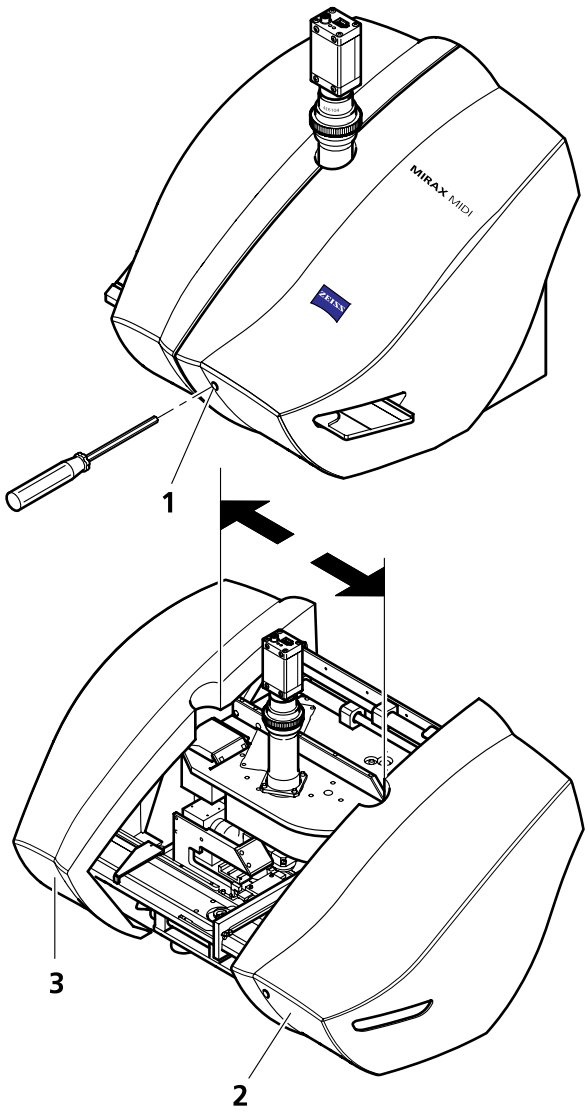


Fig. 6-1 Pushing covers apart or removing them

6.2.1 Push Protective Covers apart or remove them

For replacement of the halogen lamp (or to perform other maintenance work) and to interrupt a running digitization procedure because of slide jamming or insert jamming, the protective covers of the MIRAX MIDI need to be pushed apart in order to eliminate the problem. To improve access to special points you may remove the covers completely.

Remember to terminate the scan program and turn power to the control computer and the 24 V power supply unit off before pushing the covers apart (exception: filter change) or removing them.

The protective cover is composed of two halves anchored to each other.

At the front of the cover (6-1/**2**) there is a depression (6-1/**1**):

- Push with a blunt, long object (e.g. 3 mm Allan key) against this depression to release the anchorage. A "clack" can then be heard.
- Now the two halves can be pushed apart.

If necessary, one or both halves can be removed completely. For this purpose, push them to the right or to the left until they can be detached.

- On completion of the maintenance or repair work, the cover halves must be pushed together again and locked. A "clack" can then be heard clearly.

Make sure that no cables are squeezed when you are pushing the cover halves together. This refers in particular to the cable(s) of the scan camera.

6.2.2 Replacing Fuses

Make absolutely sure that the power plug is disconnected before you replace fuses.

The line power inlet and the fuse compartment are located at the back of the 24 V power supply unit. The fuse compartment contains two fuses of type T 2.5 A / H / 250 V.

- Disconnect power plug.
- Pull fuse holder (6-2/**3**) out to the front. Use a small screwdriver if necessary for convenience.
- Remove fuses (6-2/**2**) from fuse holder and replace them with new fuses.
- Push fuse holder (6-2/**3**) into the fuse compartment (6-2/**1**) to mechanical stop position.
- Insert power plug.

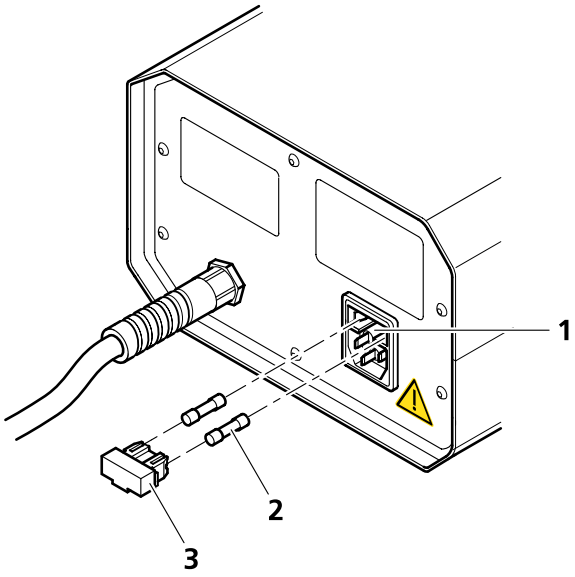


Fig. 6-2 Replacing fuses

6.2.3 Replacing Halogen Lamp

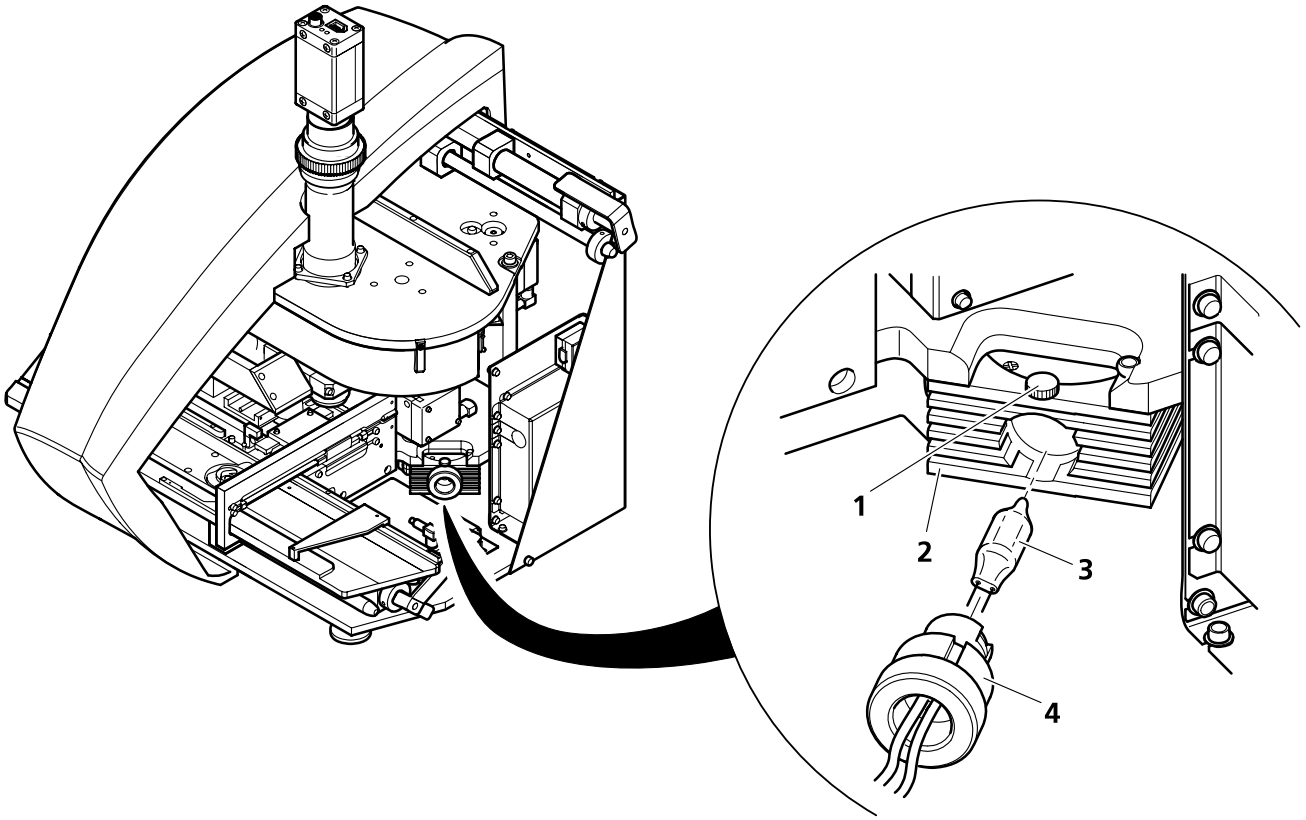


Fig. 6-3 Replacing halogen lamp

- Close MIRAX control software.
- Turn power supply off and disconnect power plug.
- Pull cover apart (see section 0).
- Hold lamp mount (6-3/**4**) with your hand.
- Loosen knurled screw (6-3/**1**) on the upper side of the heat sink (6-3/**2**), until the lamp mount can easily be pulled out sideways.

Caution! Hot surface!
Allow the lamp surface to cool down sufficiently.
Wait at least about fifteen minutes.

- Pull defective lamp (6-3/**3**) out from the top and replace it with a new lamp (000000-0467-436).

Avoid touching the new lamp bulb with bare fingers during replacement action!

- Carefully push the lamp mount into the heat sink to mechanical stop position. Make sure that the groove at the lamp mount is properly oriented (the groove is oriented toward the knurled screw). Lock the lamp mount with the knurled screw, once the mount is correctly positioned.
- Push the cover halves together again (see section 0).
- Connect power plug and turn power supply on.
- Start MIRAX control software.

6.2.4 Replacing Lamp Module (MIRAX MIDI BF/FL)

This section recites some extracts from the HXP 120 operating manual. For full information, notably instructions regarding safety, you are strongly advised to consult that manual. No alignment is required for the lamp module. Accordingly, the module is at once operational after a lamp replacement.

Caution! Hot surface!
Allow the lamp surface to cool down sufficiently.
Wait at least twenty minutes.

- Disconnect power plug (or rather separate all poles from the mains).
- Unfix screw in lateral cover plate of the lamp housing and remove plate from lamp housing.
- Disconnect plug of the lamp.
- Loosen pressure bolt with the lever.
- Remove lamp module.
- Carefully retrieve the new lamp module (000000-0482-730) from its transporting container. Hold the lamp module only by its ceramic part.
- Insert the new lamp module. Make sure that the groove in the contact surface of the lamp captures the corresponding pin of the lamp mount.

Incorrect handling may considerably reduce the lamp module's useful life. Hold it by the ceramic part in all cases! Never touch the lamp's glass surface or the inside surface of the reflector. Skin oil is damaging to these parts.

- Re-establish plug-in connection.
- Tighten pressure bolt again with the lever.
- Screw the lateral cover-plate down to the housing.
- Connect power plug.
- Set hours meter to zero.

A new compensation image for fluorescence should be generated following each change or modification of filter (see 6.3.2.2).

The fiber-optic cable of the HXP 120 has to be exchanged at regular intervals as well.

6.2.5 Removing a Slide in Case of Fault

If a slide got jammed or has suffered breakage during a digitization process, digitization must be aborted. Having done this, you should try, by clicking the Emergency Manual Slide Removal Position button, to move the slide to a position in which it can easily be removed (section 5.4.4.4).

Crush hazard!
Remember to cancel the running digitization process, terminate the software session and turn power off before you remove a jammed slide.

- Push protective covers apart as described in section 0.
- Position the slide (Fig. 6-4/**2**).
- Remove the slide carefully. The clamp (Fig. 6-4/**1**) can be pushed slightly to the left to facilitate slide removal.

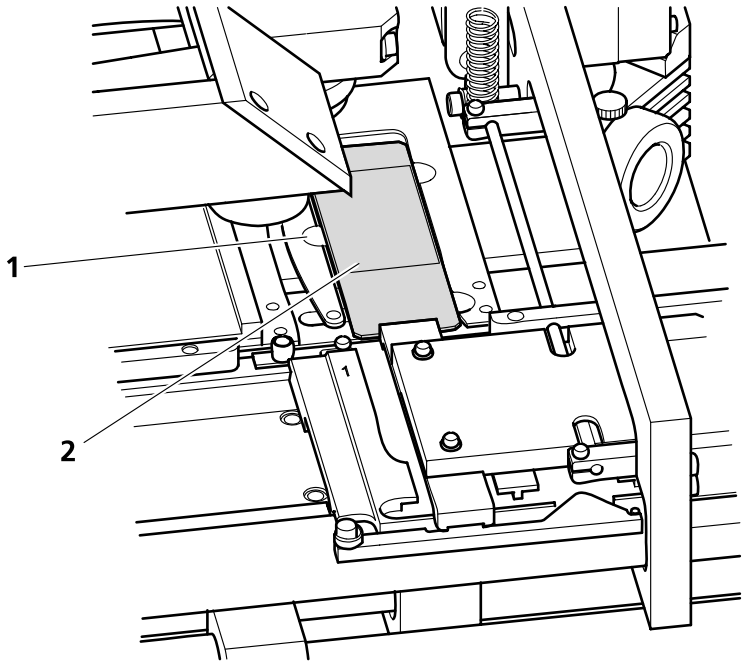


Fig. 6-4 Removing a slide

- Push the protective covers together again (see section 0).
- Restart the software.

Please note that any manipulation performed on a mechanical component by the user must be followed by a complete device initialization routine (software restart).

6.2.6 Installing and Removing the Filter Module (MIRAX MIDI BF/FL)

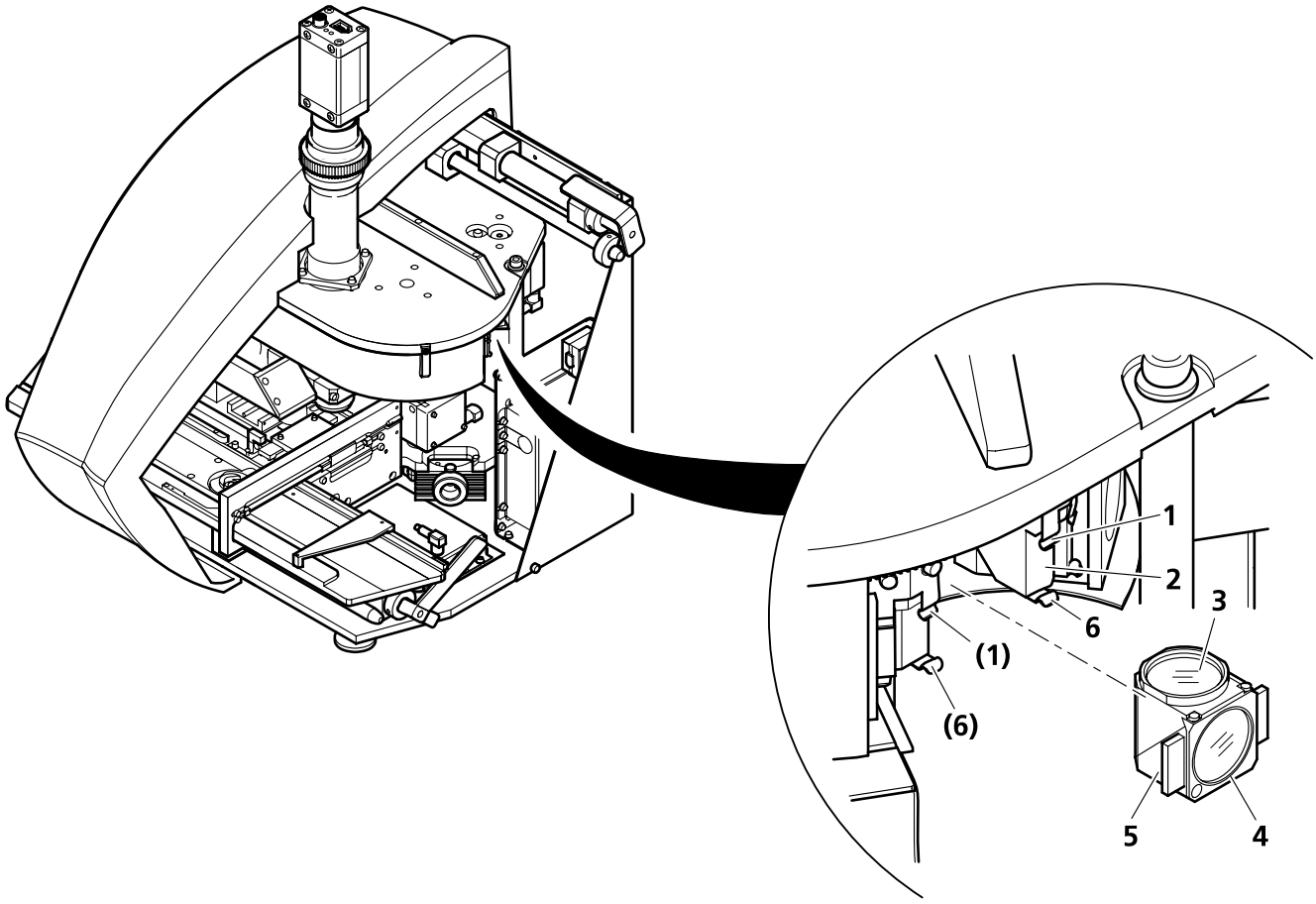


Fig. 6-5 Installing and removing the filter module

- Turn HXP 120 illuminator off.

Crush hazard!
Cancel the running digitization process before you proceed with filter module installation or removal.

- Pull protective cover halves apart or remove the right part completely, as described in section 0.
- Select the desired filter in the corresponding software window (see section 6.3.1). The filter will be positioned in such a way that the user can access it without having to turn the filter wheel manually. Enter the respective parameters also into the input fields.
- For installation, hold the filter module (6-5/**5**) by its handling pads (one on the right, one on the left side of the module) and introduce it in forward tilted position from above into the upper spring-loaded clamps (6-5/**1**) at the filter wheel. Having done this, press the filter module down, until it safely engages with the bottom spring-loaded clamps (6-5/**6**) of the filter wheel.

☞ Mounting position of the filters: excitation filter (6-5/**4**); emission filter (6-5/**3**).

- For removal, slightly tilt the filter module out of its upper spring-loaded fixtures at first, then pull it out of the bottom spring-loaded components and take it off.
- Push the right part of the protective cover in again and push both cover parts together (see section 6.2.1).
- Turn the HXP120 illuminator on again (consider the cooling time possibly required).

☞ A new compensation image for fluorescence should be generated following each change or modification of filter (see 6.3.2.2.).

6.3 Settings

6.3.1 Defining Fluorescence Filters

The definition of the fluorescence filters is done before opening the actual working area of the fluorescence software by clicking on **Options** ▶ **Microscope Settings** (Fig. 6-12). The selection window shown in Fig. 6-6 will appear. Select the **Base Settings** tab.

This register card is used to define the basic parameters of the device.

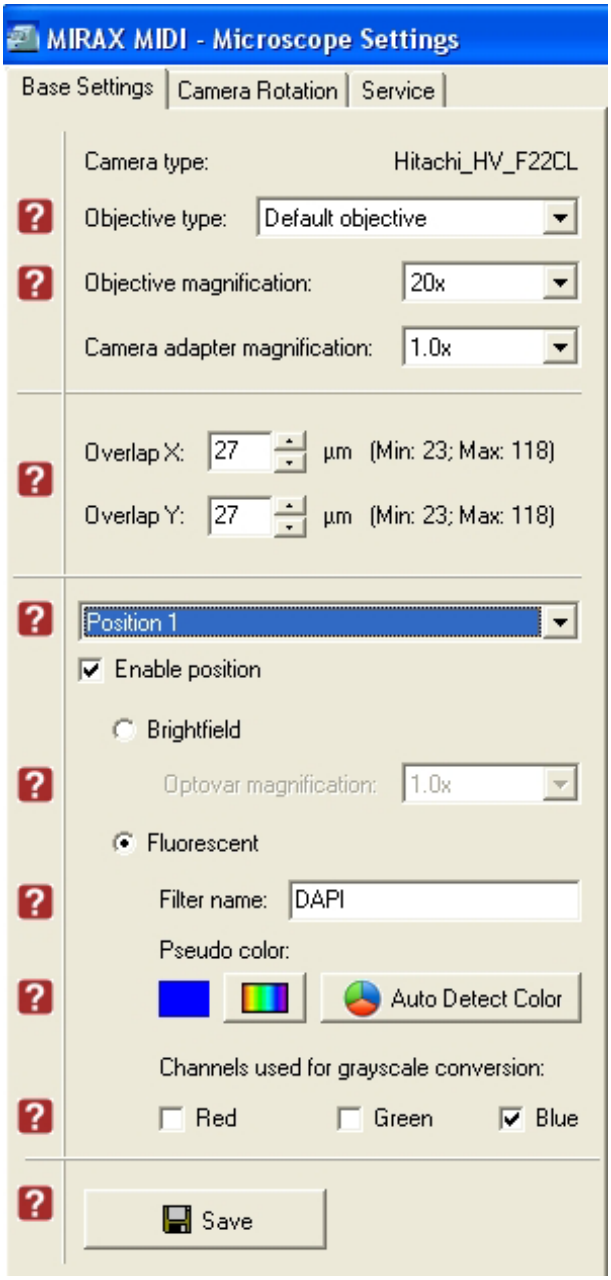
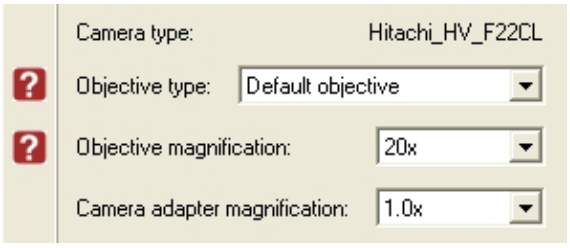
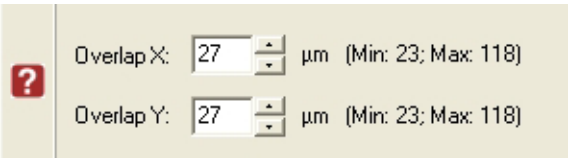
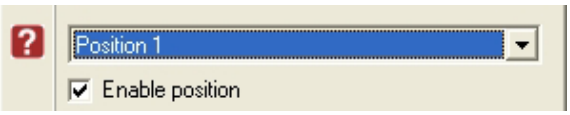
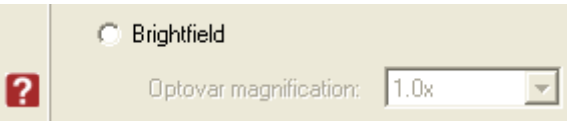
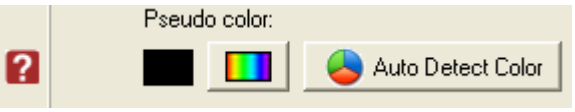
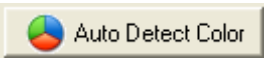


Fig. 6-6 Base Settings tab

The following entries are possible:

	This area contains the changeable optical components. The entries are to be changed only after a change of the hardware configuration. *Camera type:* This is a Service entry and can be changed by Service only.
	This area serves to define the overlap in digitizing the individual part images. These entries should not be changed!
	Position of the filter in the filter wheel – the filter will be positioned in such a way that the user can change / insert the filter - once the covers have been removed - without having to turn the filter wheel manually (see section 6.2.6). The *Enable position* selection box indicates whether this filter wheel position is used or not.
	If the above selected position for brightfield applications is used, *Brightfield* must be activated here. If Brightfield has been activated, an Optovar magnification can be selected in the selection menu located below that (1.0x means that no Optovar is used, e.g. for position 10).
	If the above selected position for fluorescence is used, *Fluorescent* must be activated here. The name of the channel is entered in the *Filter name* box.
	False color, in which the channel will be displayed later in the MIRAX Viewer (selectable by a mouse-click on ).  Automatic determination of the false color. This function can only be used if a specimen has been inserted. The procedure may take several minutes.
	Selection of the channel or channels (RGB) used by the camera (Bayer mask) to create a grayscale channel (for yellow emission light, for example, the green (*Green*) and red (*Red*) channel should be enabled). This function is available for color cameras only.

For inserting the filters into the filter wheel see section 6.2.6.

6.3.2 Recording Permanent Compensation Images for Brightfield and Fluorescence Applications

6.3.2.1 Brightfield Applications

At the beginning of the digitization process, several reference images of empty slide areas are usually recorded automatically for white balancing and shading. If slides which are almost fully covered by a specimen (such as smears) are employed, the automatic search for empty areas on the slide cannot always be successful. You will know that this is the case when, for instance, white balancing and shading in the resulting virtual slides do not exhibit the quality you are used to (e.g., a darker area along the edges for each image field).

For this purpose, a compensation image must be recorded first. This is done in the Image Compensation window. To call this window up, **Image Compensation** is to be selected under **Options** in the main window (see Fig. 6-7).

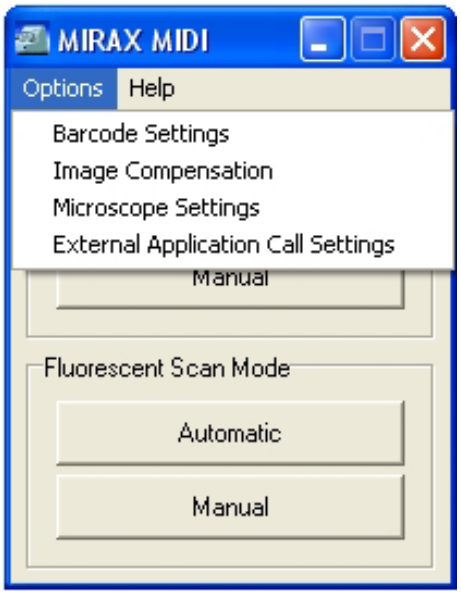


Fig. 6-7 Calling up the Compensation Dialog

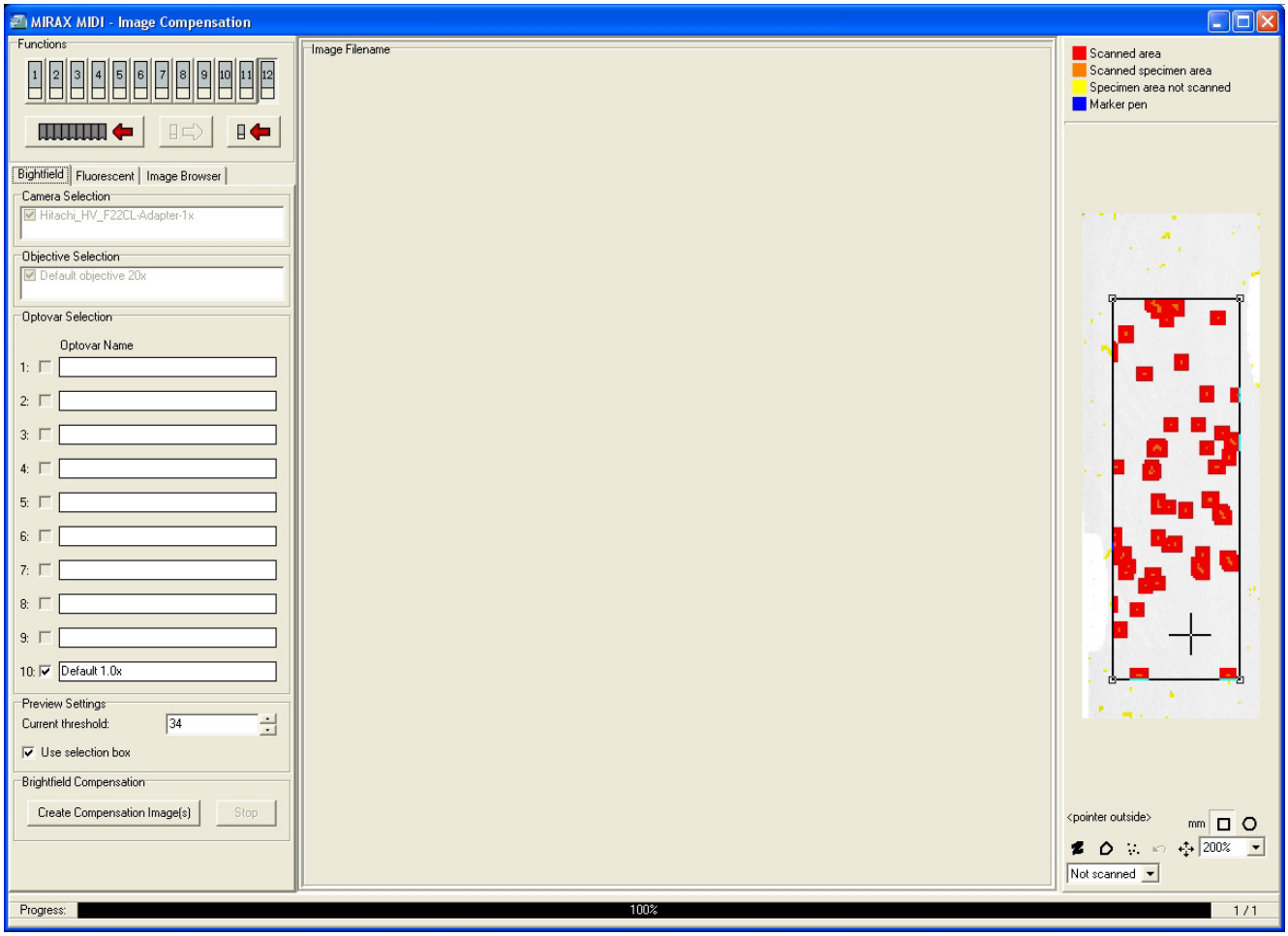


Fig. 6-8 Recording a compensation image (brightfield)

- In order for image compensation to be employed, a cover slip must be placed on a slide without specimens, with the slides matching exactly the combination to be used later on.
- Place this slide into an insert.
- Using the relevant buttons, move the insert into the instrument and the prepared slide onto the stage.
- Activate the **Brightfield** tab.
  All the defined brightfield channels possible are shown now. All the channels for which a compensation is to be performed can be selected via the check box in front of each channel.
- A threshold value needs to be defined under *Preview Settings*. It should be in the neighborhood of 20, so that any dirt that may exist will not be included in the calibration process.
- To start calibration, click on the Create Compensation Image(s) button.
  Progress can be followed under *Progress* in the lower part of the window. The calibration process may be interrupted at any time by clicking on Stop.

Completion of the calibration procedure is indicated in a window.

In order for this function to be activated for digitization, the *Use saved compensation image* entry under *Scan Options* in the **Routine Work** register card must be activated (see 5.4.4.1).

The compensation image is stored separately for each brightfield channel. The compensation image can be viewed for verification. To do so, click on the **Image Browser** tab (Fig. 6-9).

All compensation images are displayed, complete with their optical configuration. With a click on a channel, the compensation image will be shown in the middle of the window. Alternatively, you can leaf through the compensation images via the Previous Image and Next Image buttons. It must be made sure that no artifacts will appear in the image. By a click on the Export Image… button, the image can be exported in various formats for documentation purposes.

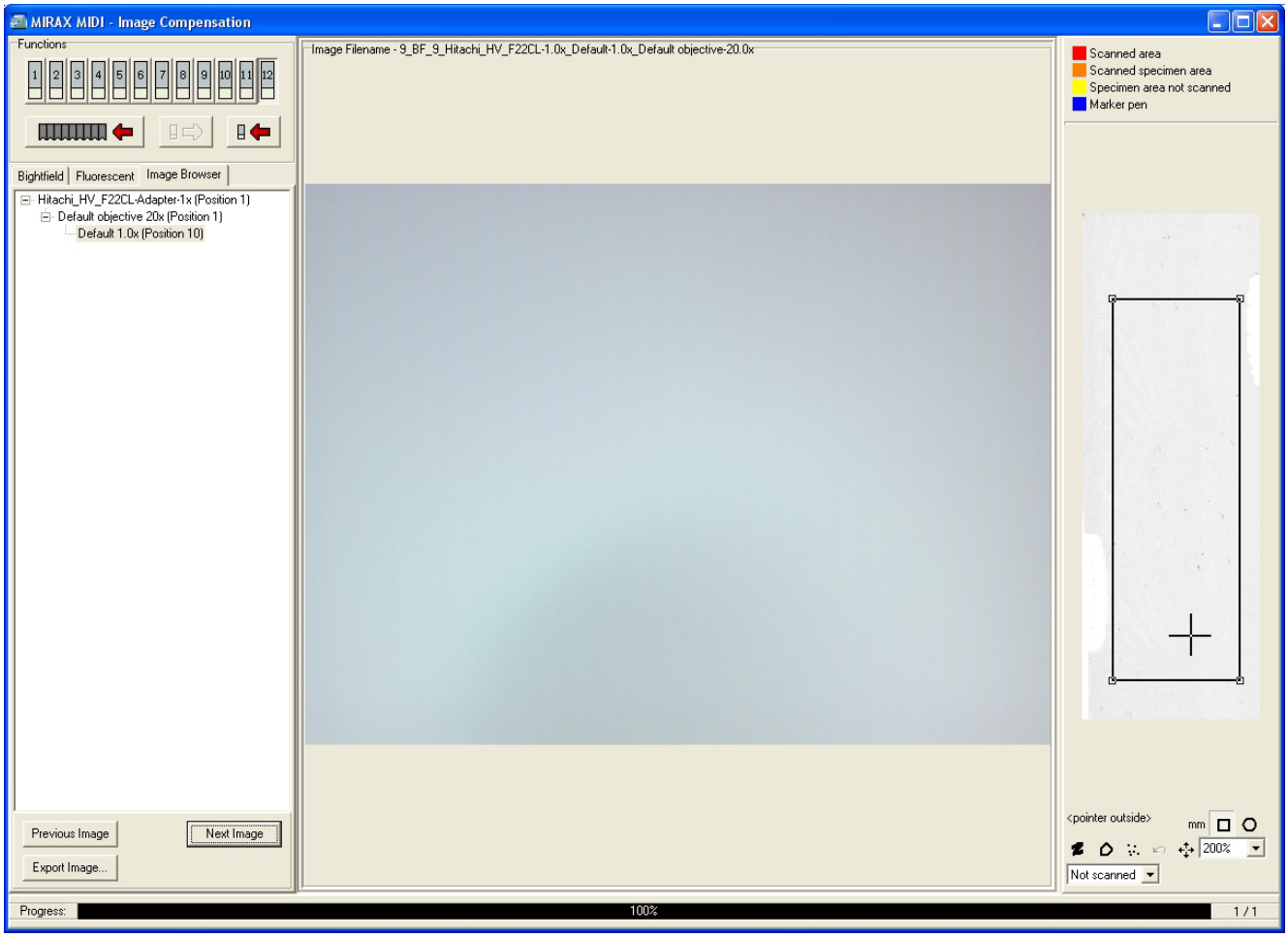


Fig. 6-9 Image Browser

6.3.2.2 Fluorescence Applications (MIRAX MIDI BF/FL)

A compensation image cannot be generated automatically in fluorescence applications. A separate compensation image must be recorded for each fluorescence channel beforehand. The compensation image should be recorded right after the insertion of a reflector module.

A calibration slide is required for the generation of a fluorescence compensation image. Use of the Zeiss calibration slide (415006-9010-000) is recommended.

The calibration procedure corresponds to the one used for brightfield applications (see 6.3.2.1).

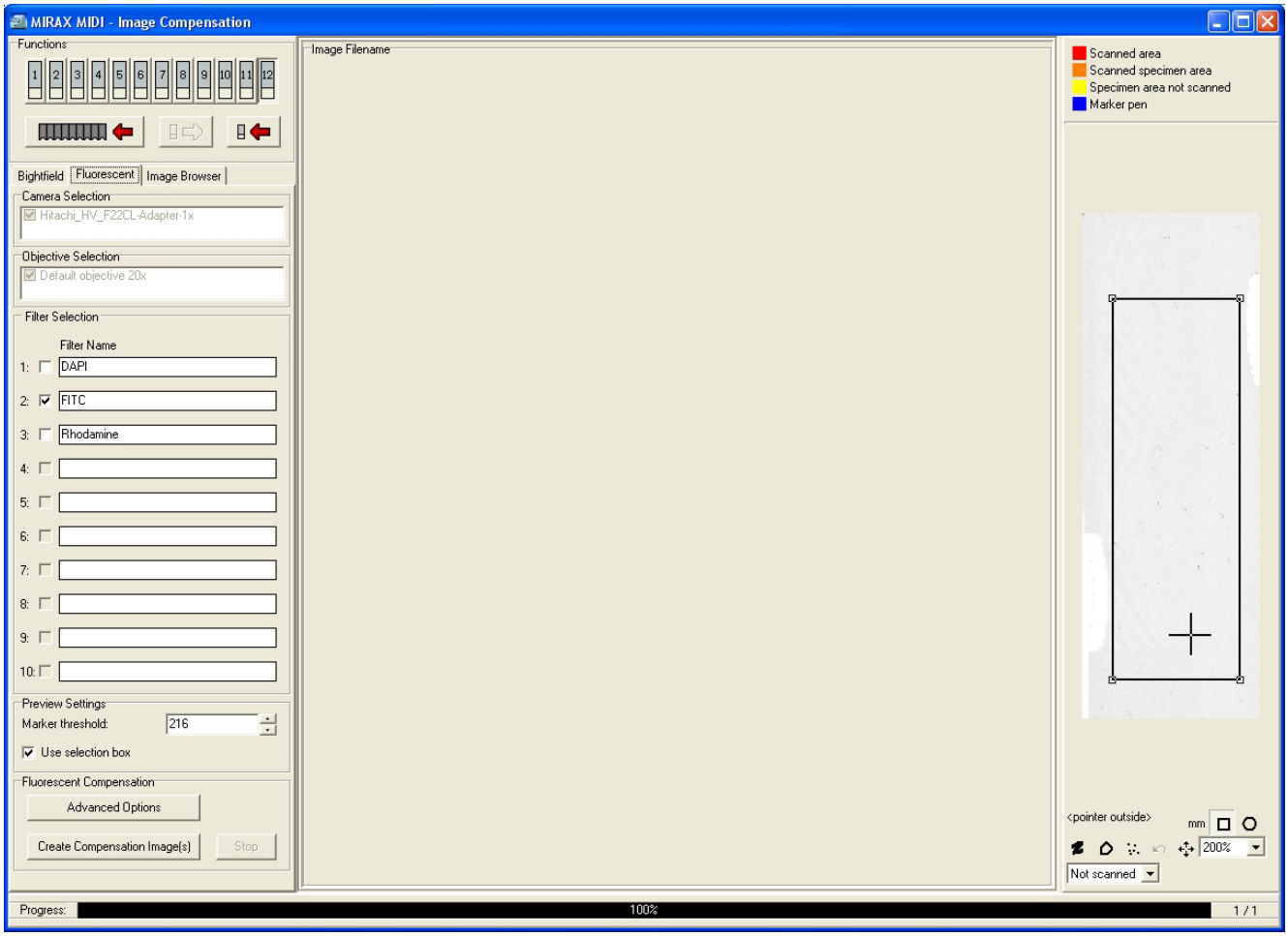


Fig. 6-10 Recording a compensation image (fluorescence)

- To call up the *Image Compensation* window, select **Image Compensation** under **Options** in the main window (see Fig. 6-7).
- Insert the calibration slide for fluorescence in an insert.
- Using the relevant button, move the insert into the instrument and the prepared slide onto the stage.
- Activate the **Fluorescent** tab.
  All the defined fluorescence channels possible are shown now.
- Using the check box in front of each channel, select the channels for which compensation is to be performed.

- A threshold value must be defined under *Preview Settings*. The threshold value should be in the neighborhood of 20, so that any dirt that may exist will not be included in the calibration process.
- The maximum exposure time can be determined under Advanced Options.

☞ If an error message is displayed during the calibration procedure, the maximum exposure time should be increased. The exposure time should not exceed 2,000 ms. Should an increase in exposure time prove insufficient, the calibration slide is no longer usable and must be replaced by a new one.

- To start calibration, click on the Create Compensation Image(s) button.
  Progress is shown under *Progress* in the lower part of the window. The calibration process can be interrupted at any time by clicking on Stop.

Completion of the calibration procedure is indicated in a window.

In order for the compensation image to be activated for digitization, the **UC** option in the **Routine Work** register card needs to be activated for the relevant channels (see page 65).

The compensation image is stored separately for each fluorescence channel. The compensation image can be viewed for verification. To do so, click on the **Image Browser** tab (Fig. 6-9).

All compensation images are displayed, complete with their optical configuration. With a click on a channel, the compensation image will be shown in the middle of the window. Alternatively, you can leaf through the compensation images via the Previous Image and Next Image buttons. It must be made sure that no artifacts will appear in the image. By a click on the Export Image… button, the image can be exported in various formats for documentation purposes.

6.3.3 Determining the Barcode Type

The MIRAX MIDI is able to read barcodes by default. This is not the case in devices equipped with the preview camera TIS DFK21F04 - for such devices the correct barcode type, which is to be used later, must be defined.

Select for this purpose **Options** from the main menu and then the **Barcode Settings** function.

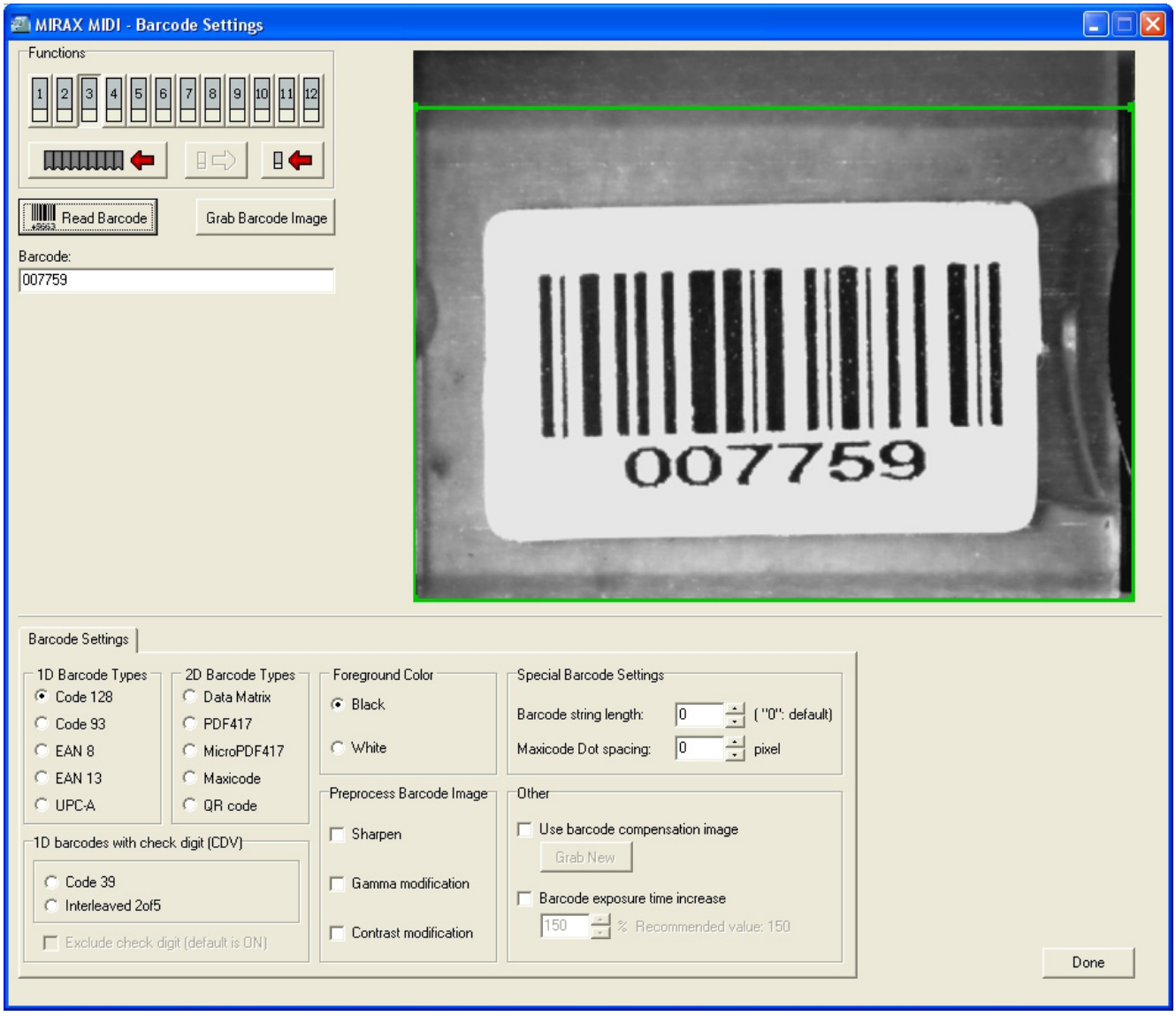
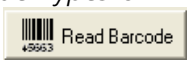


Fig. 6-11 Settings for barcode recognition

A window will appear (Fig. 6-11) where the following steps have to be carried out:

- With a click on a slide included in the symbol
  

  , the selected slide will be moved to the transfer position.
- With a click on the symbol  , the selected slide will be transported from the insert to the specimen holder.

- Now, the barcode type used has to be enabled under *1D Barcode Types* or *2D Barcode Types*. Once this has been done, the barcode can be read by clicking on the 

button – the barcode area will be shown in the window and the read barcode indicated under *Barcode*. If the selected barcode type does not correspond to the barcode stuck on the slide (i.e. the barcode has not been recognized), another barcode type has to be selected.

The two barcodes Code39 and Interleaved 2of5 include a checksum code. By default, the check digit is not indicated. If the barcode shall be indicated with the check digit, the function *Exclude check digit* must be deactivated.

- If the barcode cannot be read although the correct type has been selected, the image can be optimized via the settings under *Preprocess Barcode Image*, until the barcode can be read. Another possibility for optimization is offered at *Other*: Improved barcode identification can be reached with the help of a compensation image. Before this function can be used, a compensation image must be captured. This will be initiated by clicking on the *Grab New* button. Beforehand, a slide with a not printed barcode has to be inserted.

If the barcode cannot be read in spite of having proceeded as described, please contact the service to have the problem explored (e.g. barcode area out of focus).

- A green frame will be presented. It marks the area to be used for the interpretation of the barcode. The size and position of the frame can be changed with the aid of the mouse. This limitation is recommended if two barcodes are located in the barcode area and the barcode to be used for identifying the slide shall be specified.

If you have limited the area, make sure that the barcode is located precisely within this area.

The settings under *Special Barcode Settings* should not be changed.

- With a click on the symbol  , the slide will be moved back into the insert.
- With a click on the symbol  , the slide insert will be moved to a position where it can be removed by the user.
- The settings are saved by clicking on the Done button.

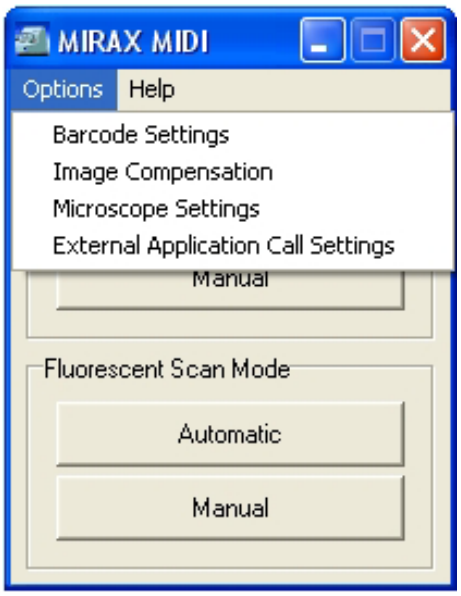


Fig. 6-12 **Calling Microscope Settings**

6.3.4 Determining the scan camera rotation angle

If the stitching quality is inadequate (e.g. double contours at the edge of the image fields), an improvement can be reached by redetermining the angle of the scan camera. Please contact your local Zeiss service if the image quality does not improve by using this function.

If this calibration function is not correctly used, it may happen that the outcome of the digitization process is inadequate. That is why this function has to be used with great care.

The function is called via the main window under **Options**, clicking on **Microscope Settings**. Select the register card *Camera Rotation*. The window opened now allows you to carry out this calibration step by step. For this purpose, a slide containing a specimen is used. Ideally, the specimen should be stained with hemalum/eosin.

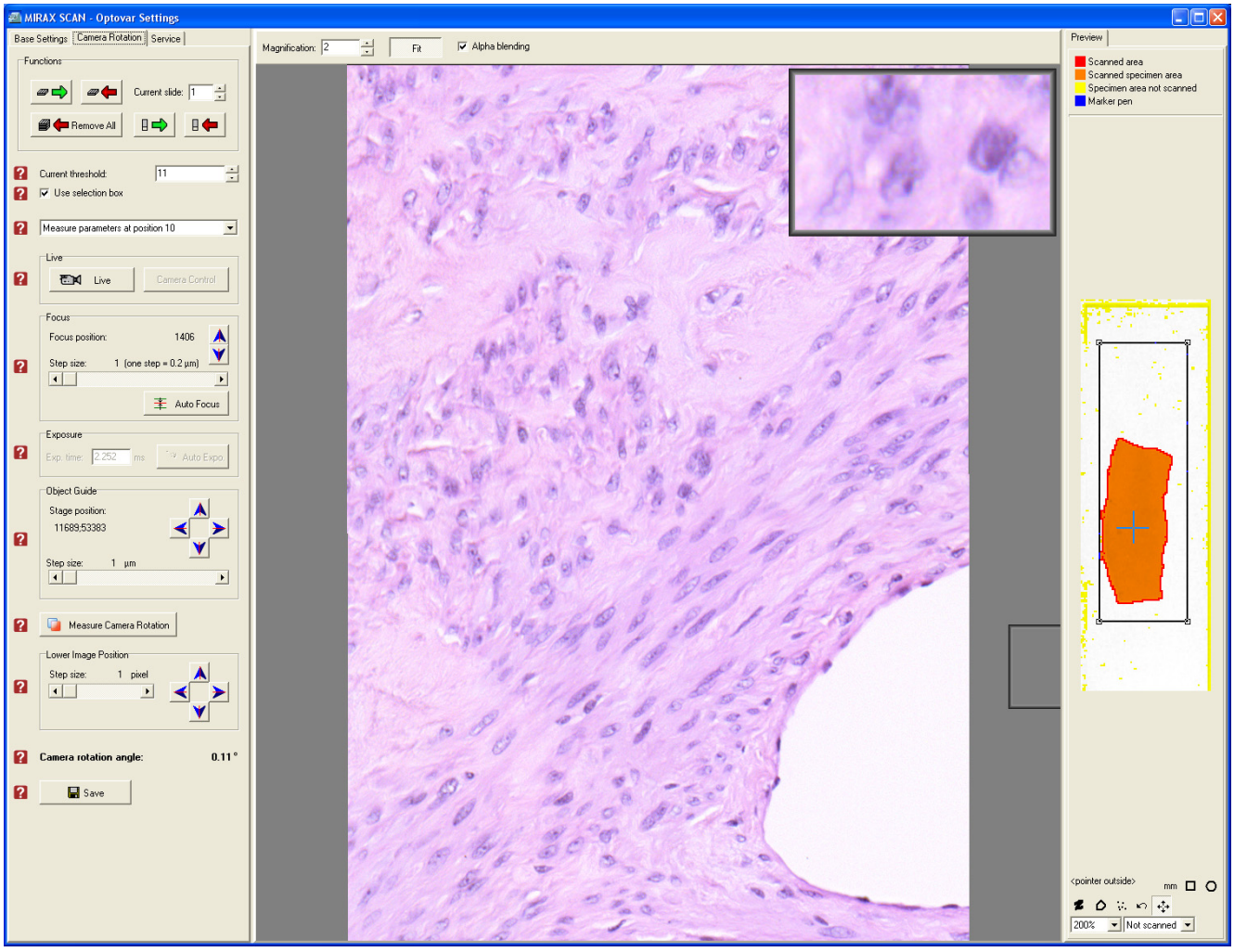


Fig. 6-13 Overview for determining the camera rotation angle

First, the selected slide must be moved to the transfer position. For this purpose click on a slide included in the symbol 

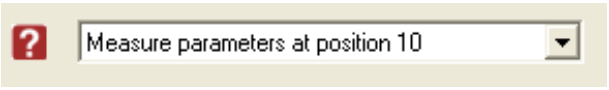
.

With a click on the symbol  , the selected slide will be transported from the insert to the specimen holder.

The following steps must be performed one after another. You may call the corresponding help function by clicking on  .

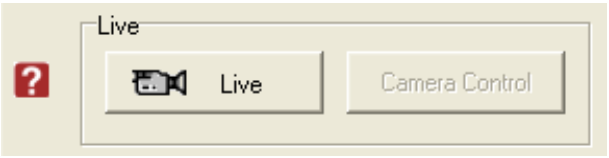
Step 1:

Select at *Current threshold* the threshold for identifying the specimen. A suitable threshold allows complete masking of the specimen. If the *Use selection box* function is activated, the identification area can be limited in order to exclude, for example, the edge of the coverslip.



Step 2:

This field allows you to select the position to be used for subsequent measurements. Position 10 is the standard channel for brightfield.

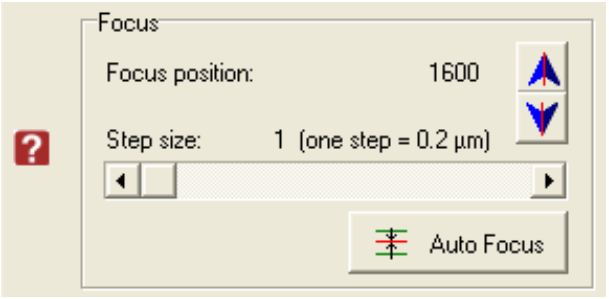


Step 3:

Activate the camera image via the Live button.

Step 4:

Activate the mechanical stage function by clicking on the  button (on the right side below). Click now on the specimen area on the right side (preview image) to select a spot that is as homogeneous as possible.

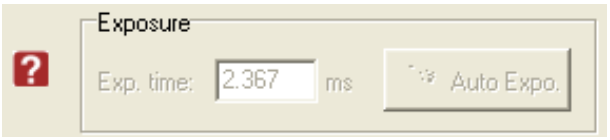


Step 5:

Initiate automatic focusing via the Auto Focus button (you may also use the blue arrows for manual focusing).

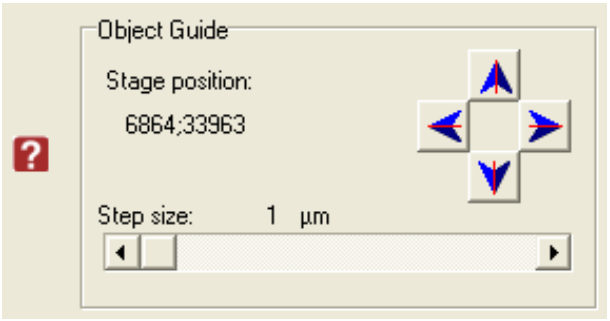
Step 6:

In the case of brightfield measurements (i. e. for color cameras), nothing can be entered in this panel, because the system determines the exposure time automatically. In the case of measurements for fluorescence channels, an exposure time can be entered here or it can be determined automatically via the Auto Expo. button.



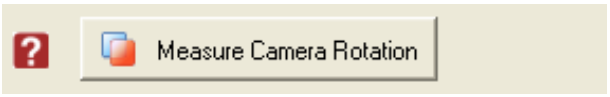
Step 7:

If positioning cannot be reached with sufficient accuracy by using the preview image, the specimen can be shifted by means of the blue arrow buttons. *Step size* is used to adjust the size of the individual steps.



Step 8:

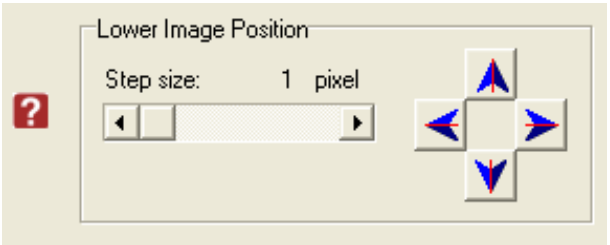
With a click on the Measure Camera Rotation button the system will take a snapshot of the current image and another snapshot of an adjacent image field. Both snapshots will be displayed in the main window. The system will determine automatically the camera rotation angle (see step 10).



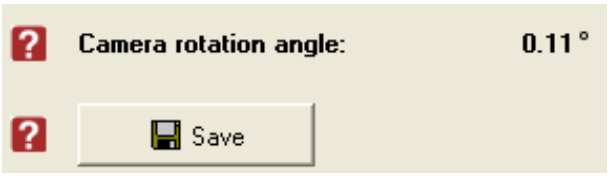
Step 9:

If the two snapshots are not aligned correctly (for example, if the deviation of the camera rotation angle is too large), the lower image can be shifted by using the blue arrow buttons.

For exact alignment or rather for checking the alignment, a window will be superimposed above on the right with an enlarged display of the transition of both snapshots. The magnification factor can be changed at *Magnification*.



The Fit button should be activated for displaying both snapshots simultaneously. By default, a blending filter is used to improve the transition of both image fields. This filter can be deactivated via *Alpha blending* to facilitate better manual adjustment.



Step 10:

Here, the camera rotation angel is indicated. Values > 0.5° or < -0.5° appear in red color and cannot be saved. In this case the camera must be readjusted. Contact the Service for this purpose.

Finally, click on the Save button to write the changed values into the EEPROM of the device.

In the end, push the slide back into the insert and remove the insert.

6.3.5 Calling an external Program

The MIRAX control software can call an external program and transfer corresponding parameters. The preconditions to initiate an external program call are as follows:

- Digitizing of a slide is finished (Scan Finished Call).
- An error has occurred (Error Occurred Call).

The settings are defined via the **External Application Call Settings** window (it is opened from the main window under **Options** with **External Application Call Settings**).

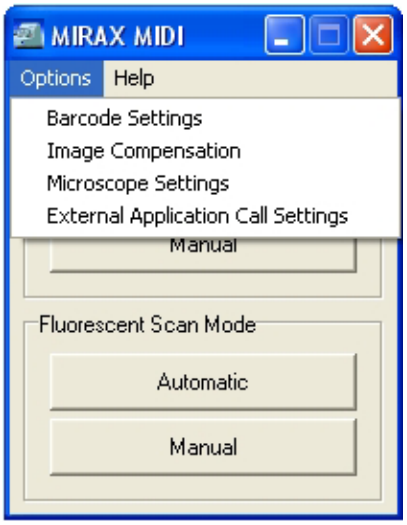


Fig. 6-14 Opening of External Application Call Settings

Up to four settings are possible for either situation (see Fig. 6-15):

Application:
This field is to define the program (including the path) to be called (e.g. a converter for another image format).

Start in:
This field is to select the path for executing the program. Normally, this is the path where the program to be executed is located.

Parameters:
This entry allows parameters to be transferred to the external program, provided that this action is supported by the external program. This can be any character string. The MIRAX control software provides two defined parameters: <Slidename> and <Errortext>. If <Slidename> is displayed, the name of the virtual slide will be transferred; if <Errortext> is indicated, the error message will be transferred in the case of error; this is recommendable if, for example in the case of error, a user shall be informed.

Run:
Through a selection menu one may specify the presentation window in which the external program will be executed.

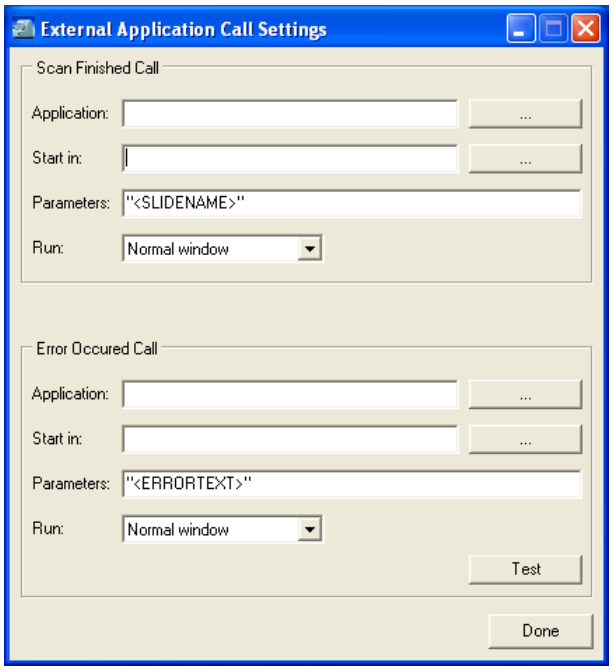


Fig. 6-15 Calling an external program

6.4 Requesting Service

Unless expressly described in this manual as a user work item, work of any kind requiring intervention in mechanical, optical or electronic parts of the device, including intervention into its internal and its electrical system, may not be performed by anyone other than Carl Zeiss Customer personnel or specifically **authorized** expert personnel.

To ensure that your MIRAX MIDI will remain optimally set and continues in a faultless operating condition over a longer period of time, we recommend that you enter into a Service and Maintenance Agreement with Carl Zeiss.

For supplementary purchase orders or to request service, please contact your local Carl Zeiss representative.

For further information, you can also visit us on the Internet at:

mikro@zeiss.de

http://www.zeiss.de