

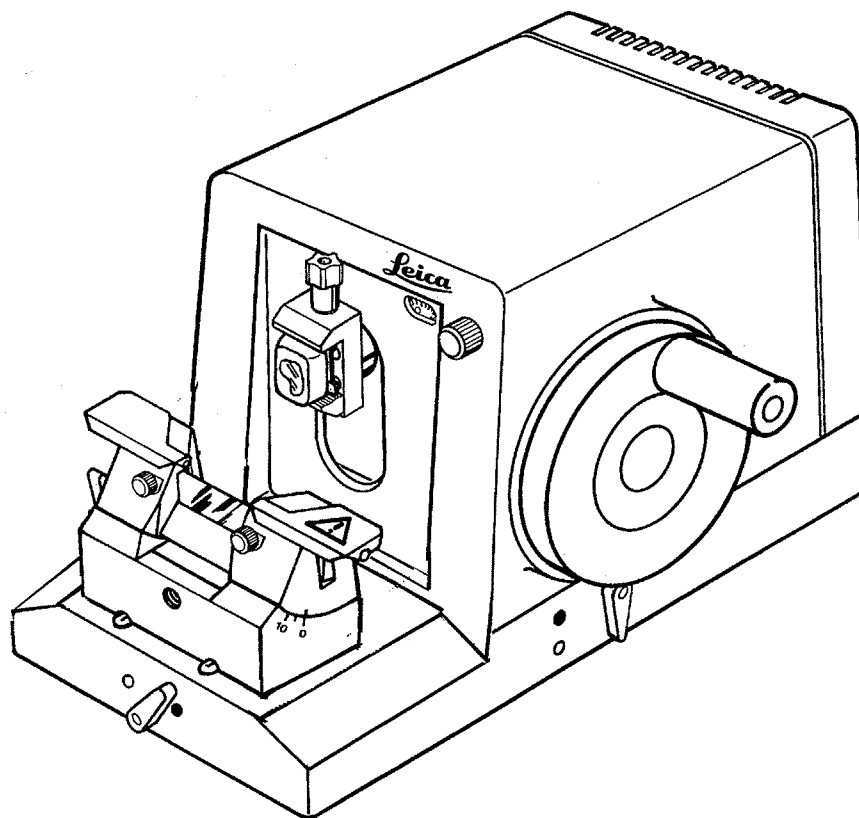
**Gebrauchsanweisung  
Operating Instructions  
Notice d'emploi  
Manual de instrucciones**

**RM2025**

*Leica Microsystems Inc.*

*1-800-248-0665*

*1-847-317-7212*



## **Operating instructions**

# ***RM2025***

## **Rotary microtome**

**Always keep this manual together with the instrument!**






**Before working with the instrument read this instruction manual carefully!**

**Subject to change!**

**V 2.0 E - 07/93**

***Leica***

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## 1. Technical specifications

Sectioning thickness range: 1 - 60  $\mu\text{m}$

Section thickness selection:

1  $\mu\text{m}$  increments from 0 - 10  $\mu\text{m}$

2  $\mu\text{m}$  increments from 10 - 20  $\mu\text{m}$

5  $\mu\text{m}$  increments from 20 - 60  $\mu\text{m}$

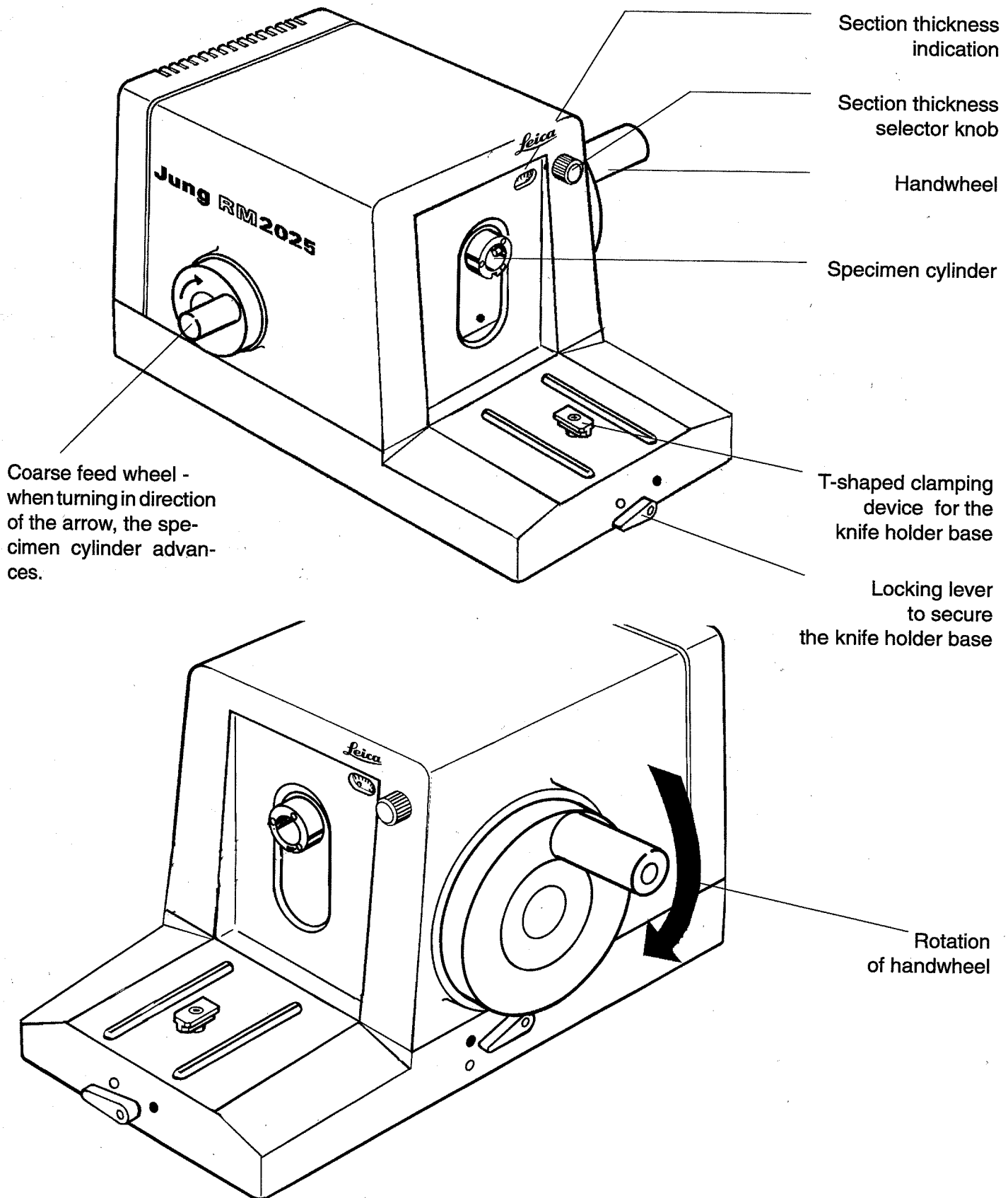
Horizontal stroke 25 mm  
(via cross roller bearings)

Vertical stroke 59 mm  
(via micrometer spindle)

Maximum specimen size H x B = 55 x 40 mm  
or: Standard cassettes

Dimensions and weight: Length: 47 cm  
Width: 40 cm  
Height: 27 cm  
Weight: 29 kg

## 2. General view of the microtome



### 3. Installing the instrument

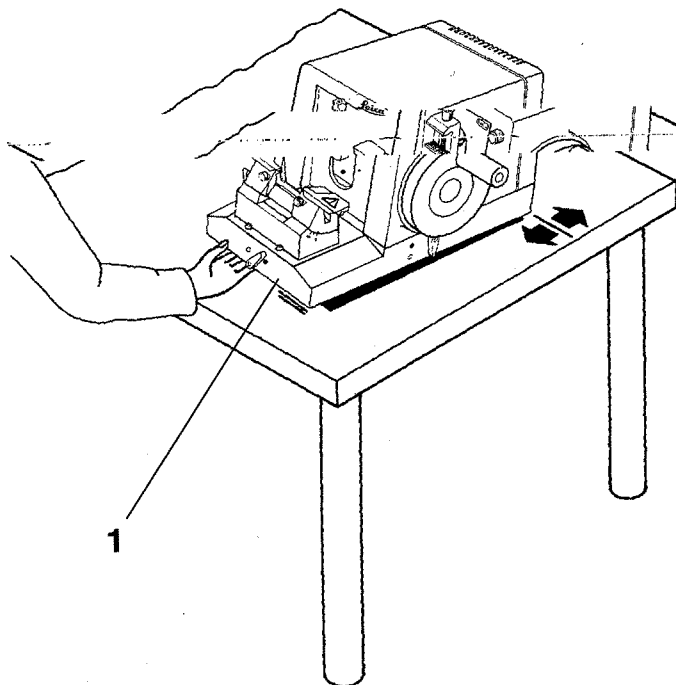


**Do not use the handwheel for carrying the microtome!**

- Install the microtome on a stable working bench provided with an easy -to-clean-surface.
- To move the instrument, slightly lift it at the front as shown in the drawing (1).

#### **Errors which should be avoided when installing the microtome:**

- Do not expose the instrument to direct sunlight or other sources of heat, as the paraffin will soften, thus giving unsatisfactory sectioning results.
- Instruments which cause vibrations should not be placed near the microtome.
- Vibrations will worsen the section quality considerably - the block will show an undulating surface.
- When installing the microtome make sure that the instrument is easily accessible and thus easy to operate.



### 4. Cleaning and maintenance

#### **Maintenance**

All moving parts of the microtome which are accessible should be lubricated once a month with oil type 405; these parts are e.g. the locking levers and their shafts, clamping screws and sliding surfaces of the knife holder base.

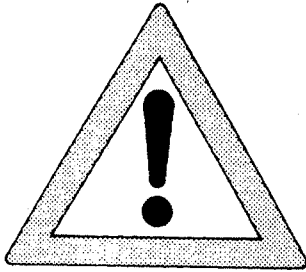
#### **Cleaning**

- Remove section waste (using e.g. a dry brush).
- For cleaning the outer surfaces, use mild detergents.
- For disinfection alcoholic disinfectants should be used; optionally you can also use alcohol or cleaning fluid.



**Caution: Long term exposure of the painted surfaces to acetone will cause damages of the paint finish!**

## 5. Safety instructions



- Please read the operating instructions carefully **before** you start to work with the instrument.

- Warning triangles mark the dangerous parts of the knife.

- The locking position of the knife holder levers are marked as follows:

- = locked
- = unlocked

### Attention:

- When attaching a specimen to the holder, lock the handwheel and cover the knife edge with guards.

- If possible, always mount the specimen **prior** to the knife!

- **Always** make sure that exposed parts of the cutting edge are covered with the knife guards.

- When finishing work, either protect the cutting edge entirely with the knife guards or remove the knife from the holder.

### RISK OF INJURY!

- Knives which are not in use should **always** be stored in the knife case!

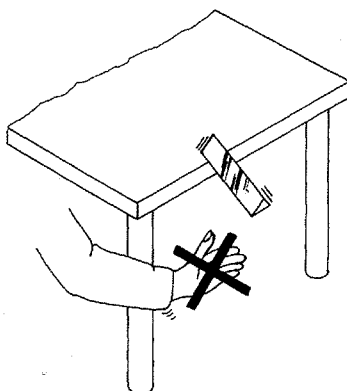
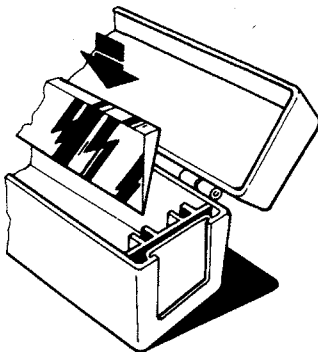
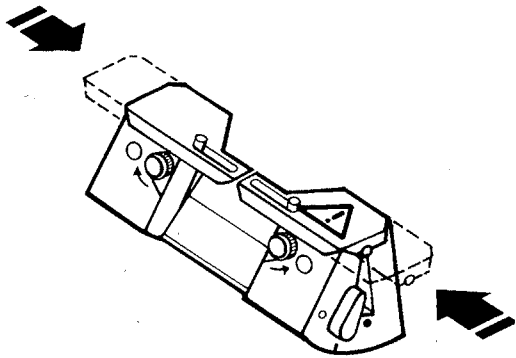
- **Never** place a knife on its back, thus exposing the cutting edge!

Before exchanging knife holders always remove the knife!

### RISK OF INJURY! !

- **Never** try to catch a falling knife!

### RISK OF INJURY!



## 6. Quick release specimen holding system

### Inserting and exchanging specimen holding systems (with non-orienting adapter)

The microtome is available in two basic versions:

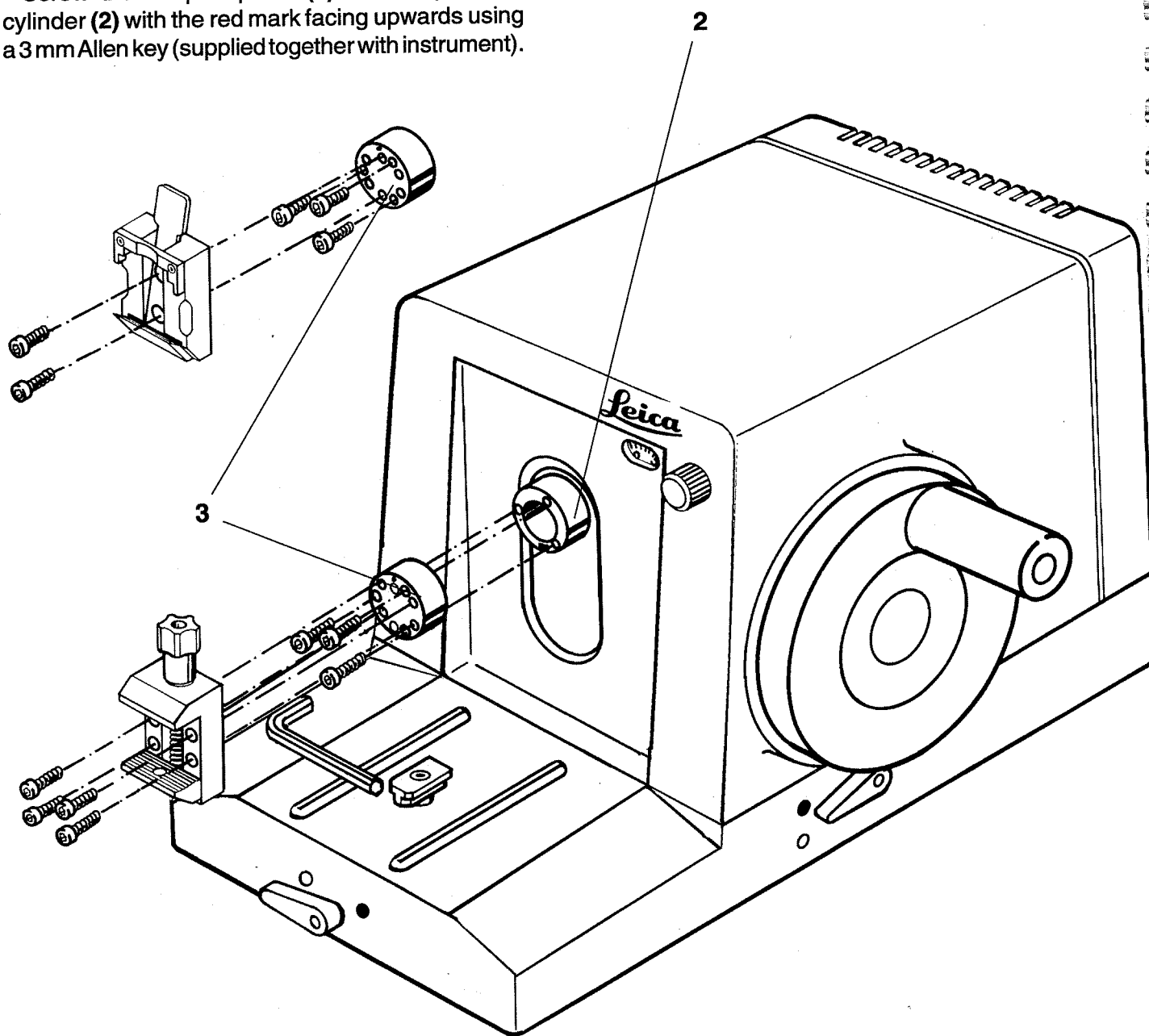
1. With non-orienting adapter
2. With x/y-orienting adapter.

#### 6.1 Non-orienting adapter piece

- Screw the adapter piece (3) to the specimen cylinder (2) with the red mark facing upwards using a 3 mm Allen key (supplied together with instrument).

- Screw the specimen clamp to the adapter piece as shown in the drawing below.

- To exchange the specimen clamps, simply loosen the screws, remove the clamp and mount another one.





## 6. Quick release specimen holding system

### 6.2 X/Y-Orienting adapter piece

The orienting adapter piece allows for the optimisation of the position of the cutting surface of the clamped block.

- Screw the adapter to the specimen cylinder (2) as shown in the drawing below, with the red mark (8) facing to the right.
- Unscrew the spring bushing (8) until the red ring becomes visible.
- Pull out the eccentric bolt (5).
- Now the specimen clamps can be mounted onto the adapter piece.

### Inserting and exchanging specimen holding systems (with x/y-orienting adapter)

To **mount** the specimen clamps proceed as follows:

- A groove (10) is provided in the clamping shaft (9).
- The groove (10) must be in line with the red mark (6) when mounting the specimen clamp.
- Mount the specimen clamp.
- Insert the eccentric lever (5) with the pull-out-grip (7) facing upwards.
- Screw in the spring bushing (8) until the red mark is not visible any longer.

(When sectioning very hard materials, screw in the spring bushing (8) completely).

- Turn down the pull-out grip (7) thus tightening the eccentric lever (5).

The specimen clamp is **oriented** via the two orienting screws (4a) and (4b).

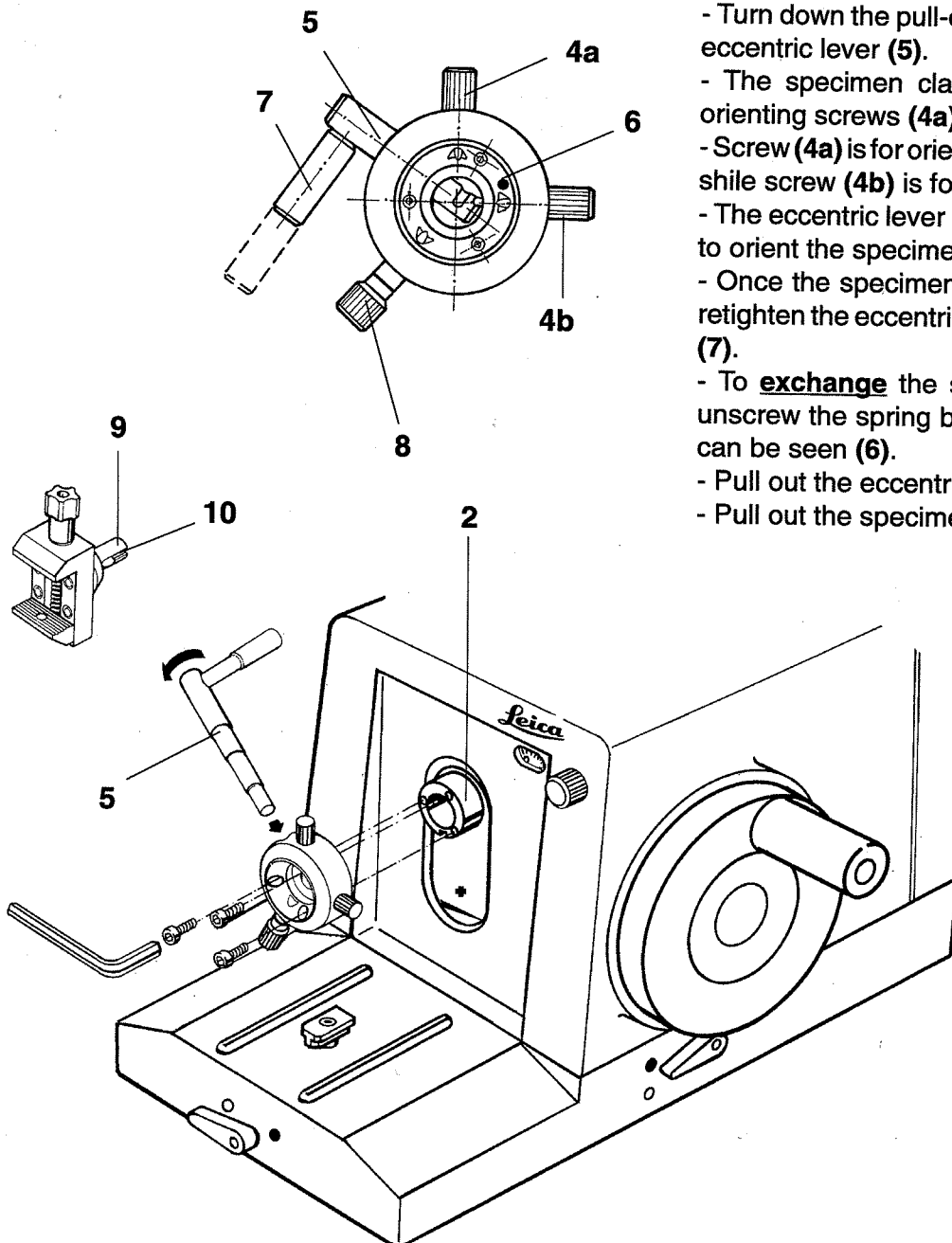
- Screw (4a) is for orienting in North/South direction, while screw (4b) is for East/West direction.

The eccentric lever (5) must be unlocked in order to orient the specimen.

- Once the specimen is oriented, do not forget to retighten the eccentric bolt (5) with the pull-out-grip (7).

To **exchange** the specimen clamping systems unscrew the spring bushing (8) until the red mark can be seen (6).

- Pull out the eccentric lever (5).
- Pull out the specimen clamp.



## 7. Orienting specimen clamps and inserts



### Please note:

The specimen block must protrude from the clamp to avoid striking the metal parts of the clamp.

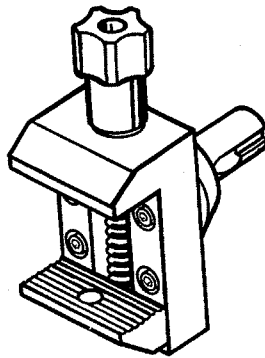


Attention: when exchanging specimens, always protect the knife edge and lock the instrument.

**RISK OF INJURY!**

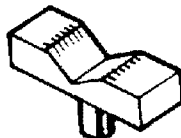
### 7.1 Standard specimen clamp

The standard specimen clamp is designed for rectangular blocks, as are e.g. paraffin blocks or embedding rings.



#### 7.1.1 Vee insert

The vee insert is mounted in the lower jaw of the standard specimen clamp (orienting and non-orienting version) and serves for clamping round specimens.



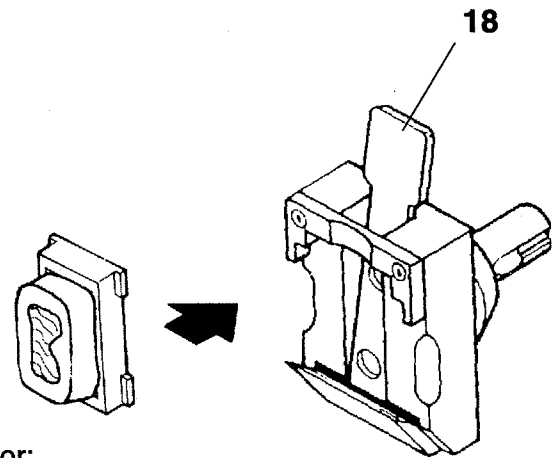
### 7.2 Universal quick release cassette clamp

The universal quick release cassette clamp is designed for Leica cassettes as well as for all other standard cassette types.

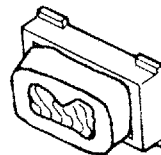
- Cassettes may be clamped vertically or horizontally (as required).
- To mount the cassette:
  - Pull the spring loaded lever (18) to the front.
  - Mount the cassette vertically or horizontally.
  - Let the lever (18) flip back.
  - The built-in spring system secures the cassette.

### Please note:

Excess paraffin should be removed regularly from the clamps with cleaning fluid. From time to time apply a light coat of oil to the springs. This is particularly important in case that you work with ice to cool down the block surface, as the melt water will eventually get into the springs and will cause them to rust and thus break.

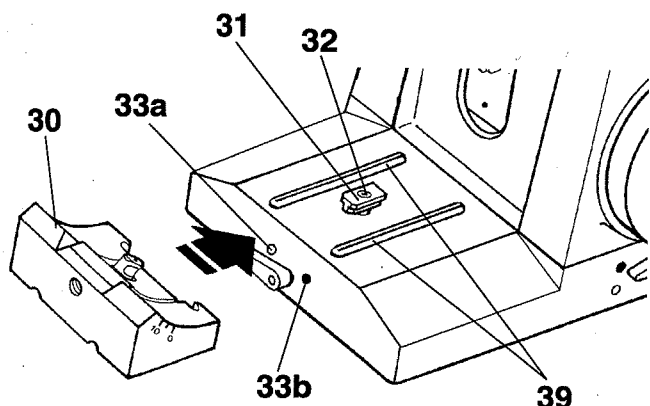


or:



(Other types of specimen clamps are available on request).

## 8. Knife holder base



### Mounting the knife holder base

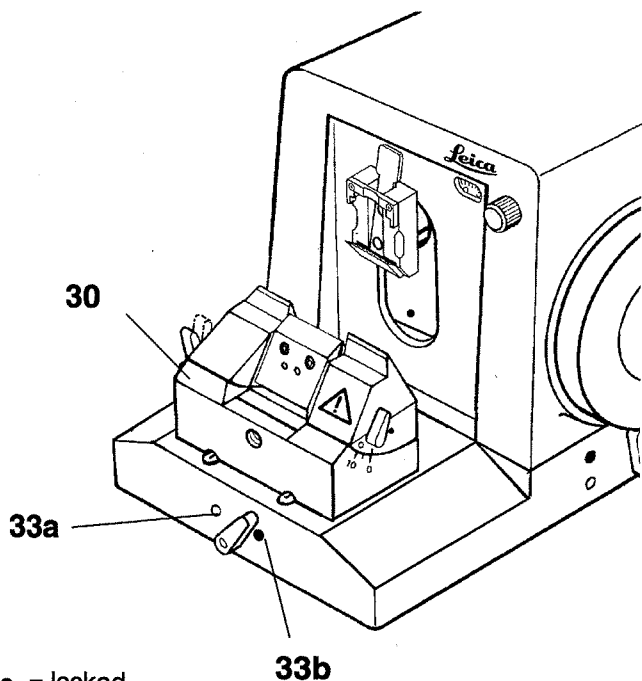
The knife holder base (30) is designed to accommodate the different knife holders. It is mounted on the microtome base plate - optionally without knife holder or with the knife holder already mounted.

- Release the locking lever (33) turning it to the left (position (33a)).

- Place the knife holder base (30) on the slideways (39) on the microtome base plate and turn the locking lever (33) to the right (33b) to secure it.

### Readjustment of the clamping system

Should the locking lever (33) not clamp tightly on the right side (33b) in the ideal position of 45°, the T-piece on the microtome base plate can be readjusted turning the screw (32) with a 4 mm Allen key until the locking lever (33) clamps tightly in the ideal position.



- = locked
- = unlocked

## 9. Knife holders

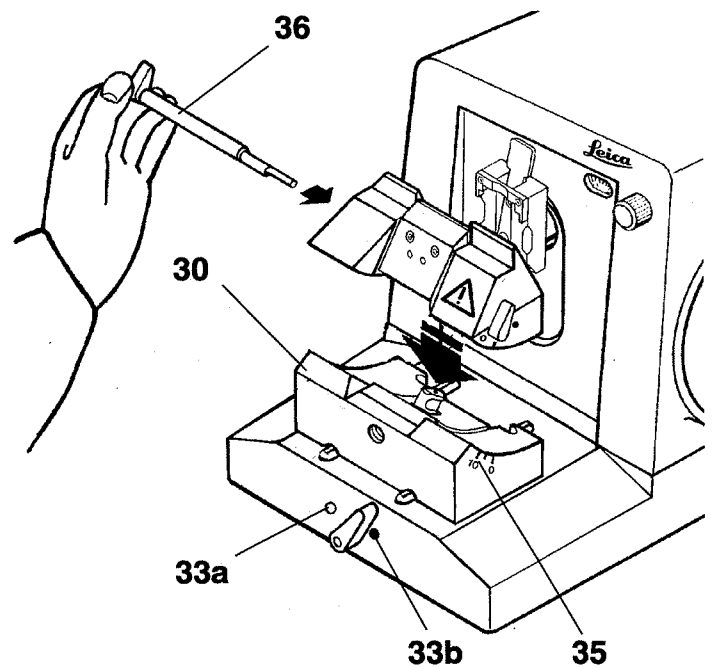
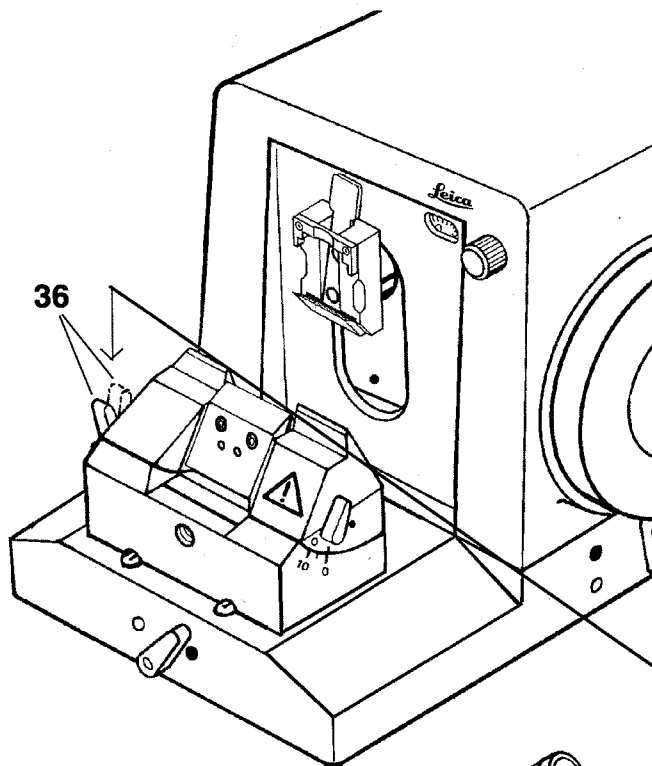


**CAUTION: PLEASE REFER TO SAFETY INSTRUCTIONS (CHAPTER 5)!**

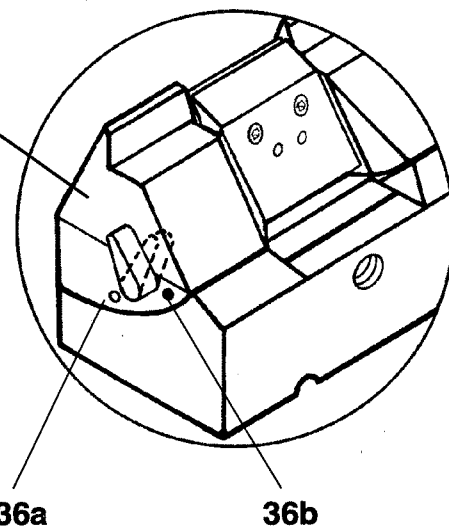
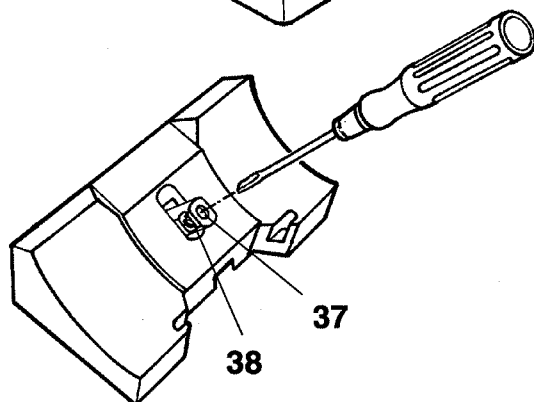
### 9.1 Mounting the knife holder

- Mount the knife holder on the knife holder base (39).
- Insert the eccentric lever from the left and secure in position (36b).
- The ideal clamping position is about 10° (36b).
- If the lever does not clamp at 10°, the tightening pin (37) can be readjusted via screw (38).
- Before adjusting the clearance angle the eccentric lever must be released to position (36b).
- The scale (35) on the right side of the knife holder base helps adjusting the appropriate clearance angle.

**Caution: Do not insert knives or disposable blades while the knife holder is not yet securely mounted on the microtome!**  
**Before exchanging knife holders, for reasons of safety always remove knives or disposable blades!**



- = locked
- = unlocked



## 9. Knife holders

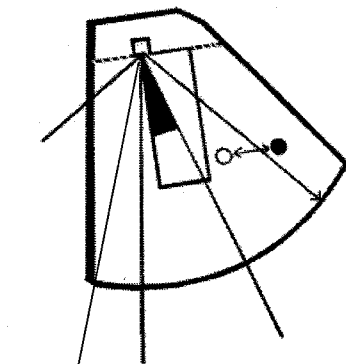
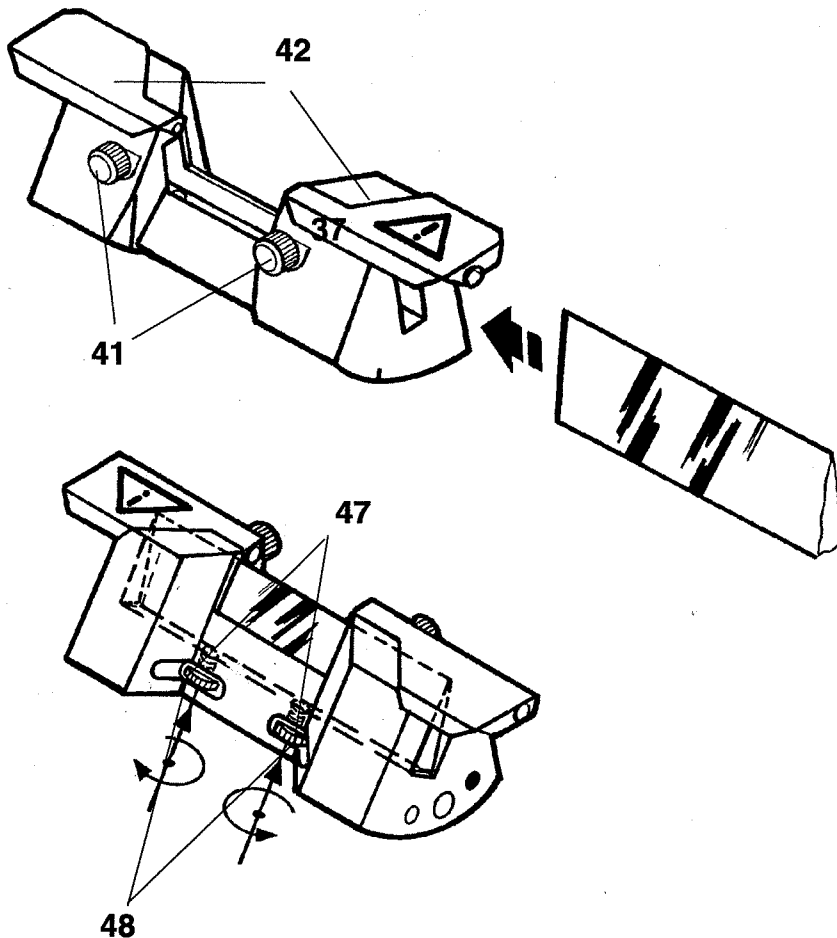


**CAUTION: PLEASE REFER TO SAFETY INSTRUCTIONS (CHAPTER 5)**

### 9.2 Knife holder N for standard knives

- The knife holder N ist designed for conventional steel and tungsten carbide knives up to 16 cm long.
  - **Attention:** Before inserting the knife, screw down completely the knife height levelling bolts (47) with the wheels (48) and also release completely the clamping screws (41) to avoid that the knife edge will be damaged.
  - Insert the knife laterally as shown in the drawing below.
- Raise the levelling bolts (47) by means of the wheels (48) until the knife edge reaches the appropriate height for sectioning (see drawing (40)).
- Secure the knife retightening the screws (41).

- The knife edge is covered with the adjustable knife guard (42).
- Always use the knife guard (42) to cover completely all parts of the knife edge which need not to be exposed for sectioning.



40 Ideal position of the knife in the knife holder: the knife edge is exactly at the height of the lower end of the groove in the knife holder.

## 9. Knife holders



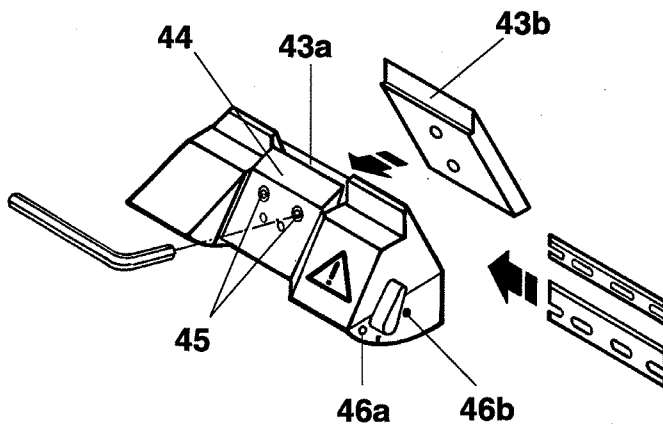
**CAUTION: PLEASE REFER TO SAFETY INSTRUCTION (CHAPTER 5)!**

### 9.3 Knife holder E for disposable blades Type I

The knife holder E is available in two different versions:

#### Type I (with exchangeable locating plate):

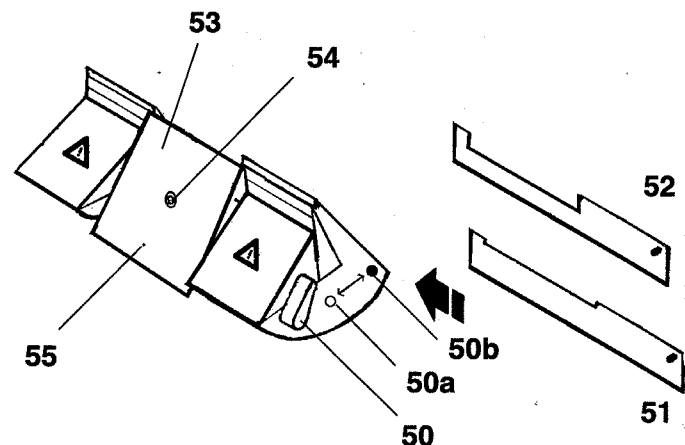
- Before inserting the blade, turn the locking lever (46) to the unlocked position (46a).
- Insert the blade laterally in the groove between the pressure plate (44) and the locating plate (43a or b).
- Secure the blade moving the locking lever (46) to the locked position (46b).
- The edges of pressure plate (44) and locating plate (43) have to be exactly parallel.
- If the blade clamping is not uniform on both sides (this problem may occur when changing to a new brand of blades), the screws (45) of the pressure plate (44) should be readjusted using a 3 mm Allen key.
- Care should be taken not to overtighten the screws.
- The relocation of the lever (46) to the clamping position (46b) should not be too tight.
- If you wish to change from high profile to low profile blades or vice versa, the locating plate (43a; b), which is fixed with two screws, has to be exchanged. The locating plates can be ordered separately. For details, please contact your local sales organisation.



### Knife holder E for disposable blades Type II

When working with the knife holder E, Type II proceed as follows:

- Turn the locking lever (50) on the right of the knife holder to the front position (51a) (= unlocked).
- Insert the blade rail for either low profile (51) or high profile (52) blades approx. halfway from the right side of the knife holder.
- Put in the matching blade (high or low profile) and insert the blade rail completely.
- Turn the locking lever (50) to the locking position (50b).
- With both knife holder E models all common disposable blade brands can be used.
- If you change from one brand to another, it may be necessary to readjust slightly the clamping force of the clamping plate (53).
- To do this, carefully adjust the screw (54) in the middle of the clamping plate with a 3 mm Allen key. Do not turn the adjusting screw (54) by more than 45° at once.
- The screw is adjusted correctly when the blade rail cannot be moved with the locking lever being in locked position (50b). When the locking lever (50) is released (51a), the blade has to be easily movable.
- Do not adjust by yourself the small screw (55) located at the bottom of the clamping plate.



## 9. Knife holders



**CAUTION: PLEASE REFER TO SAFETY INSTRUCTION (CHAPTER 5)!**

### 9.4 Quick release knife holder Z for standard knives

The knife holder Z is provided with a large clamping plate which ensures very stable clamping. Due to the central clamping principle the knife can be placed nonsymmetrically in the knife holder with the greatest clamping stability always provided in the cutting area.

- Screw down completely the knife height levelling bolts (68) by means of the knurled wheels (69) until reaching the desired cutting height.

- When working with reconditioned knives, especially when they have been resharpened many times, use the distance piece (67) which is placed in the knife holder as shown in the drawing in order to achieve an appropriate height of the cutting edge.

- If the clamping plate (63) is not exactly parallel to the cutting edge, it has to be adjusted turning the adjusting screws (64), while the eccentric lever is in position (66a).

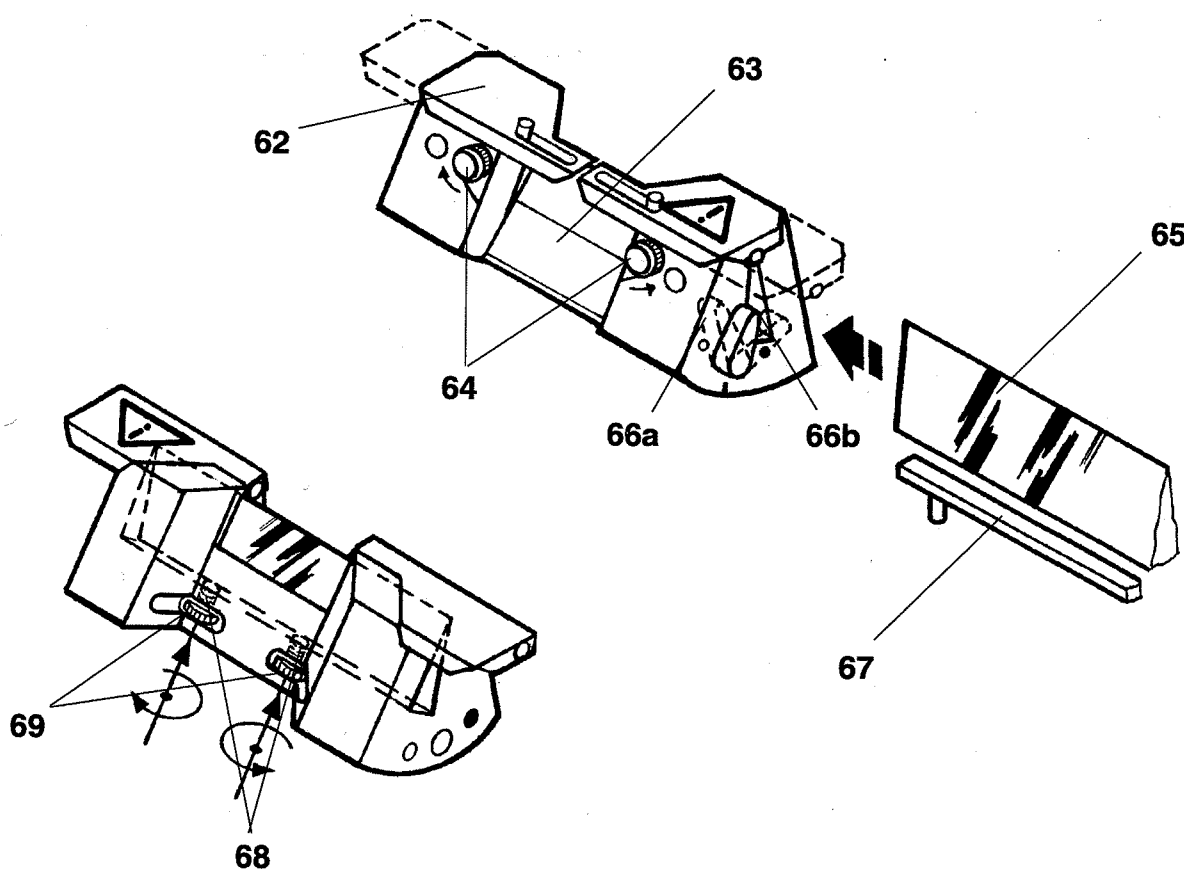
- The clamping plate being adjusted correctly, its upper edge and the knife edge have to be exactly parallel.

- To secure the knife move the clamping lever to position (66b).

The clamping lever must clamp tightly in position (66b).

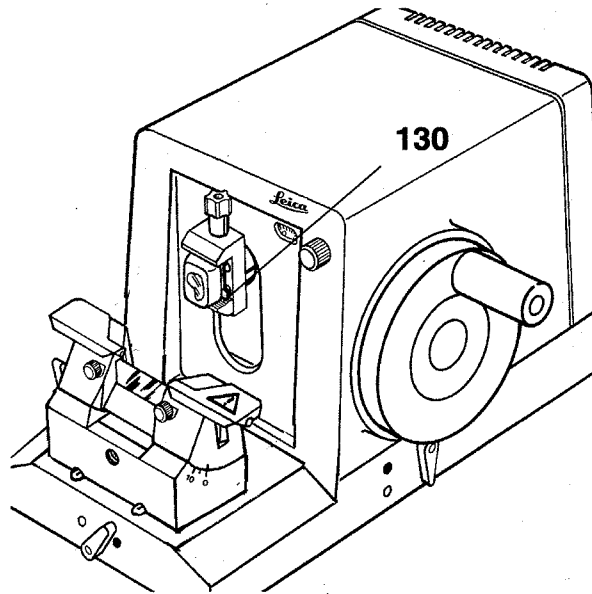
- To adjust the position of the knife or to remove the knife from the holder release the clamping lever (position (66a)).

- Make sure that all parts of the cutting edge which are not required for sectioning are covered with the knife guards (62).



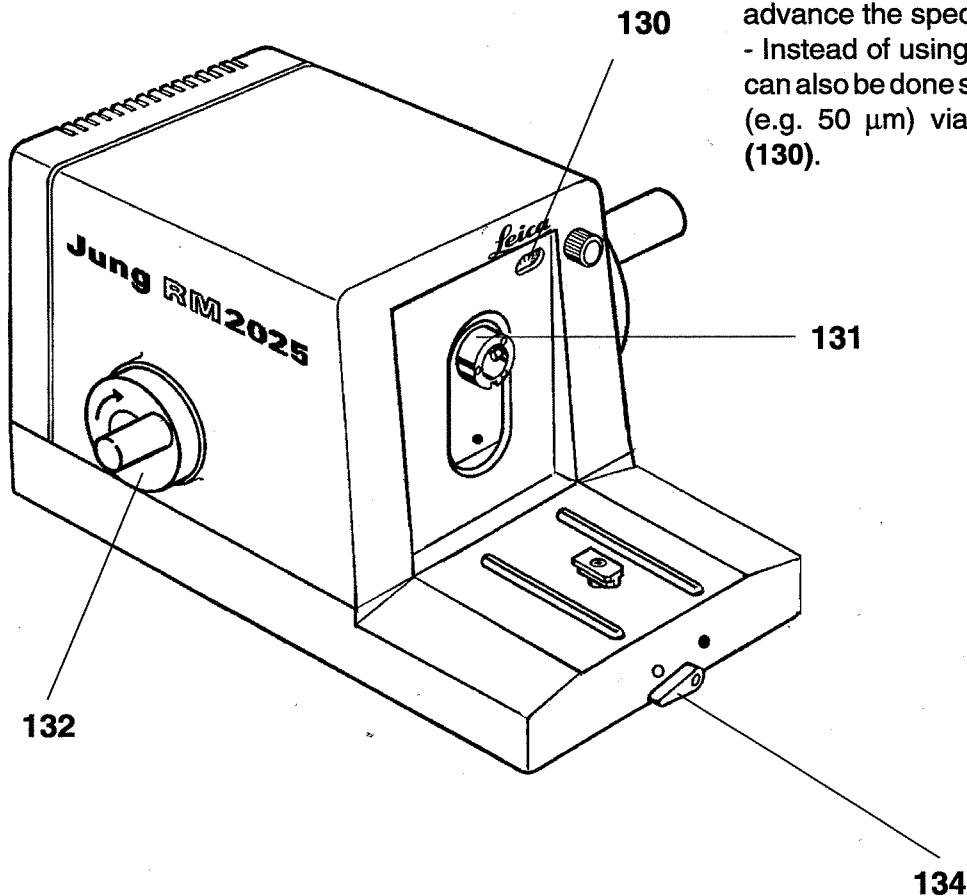


## 10. Trimming



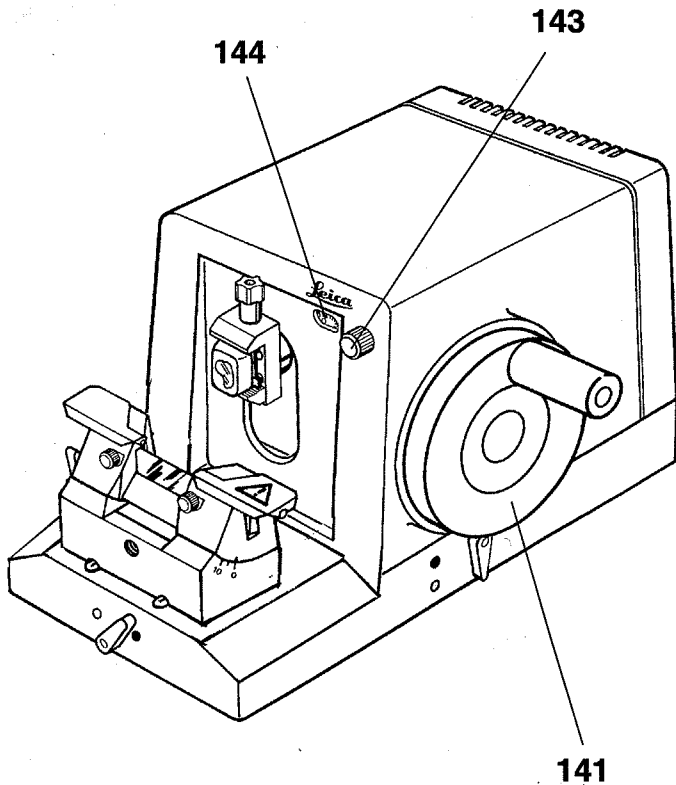
**CAUTION:** Care must be taken to assure that the specimen is carefully positioned in the specimen clamp. Damage to the knife can be caused, if the knife touches metal parts of the specimen clamp.

- Prior to the specimen trimming with the coarse feed (132), the horizontal specimen cylinder (131) and thus the specimen should be positioned at the rear limit.
- For trimming bring the knife holder as close as possible to the specimen.
- To position the knife holder close to the specimen, release the clamping lever (134), move the knife holder together with the knife holder base towards the specimen and retighten the lever (134).
- the coarse feed wheel (132) is positioned on the left side of the microtome. The arrow on the coarse feed wheel indicates the rotational direction to advance the specimen towards the knife.
- Instead of using the coarse feed wheel trimming can also be done selecting a large section thickness (e.g. 50  $\mu\text{m}$ ) via the section thickness selector (130).





## 11. Sectioning



A high quality microtome and a **knife in excellent condition** are the basic requisites for good sectioning results.

Other important factors to keep in mind are:

- the hardness of the specimen
- the cutting speed
- the knife angle,
- correct knife clamping,
- correct specimen clamping.

First select the appropriate clearance angle.

- The sections should show as little compression as possible - no tears or breaks.
- The smaller the clearance angle, the less compressed sections.
- The harder the specimen the larger the clearance angle should be.

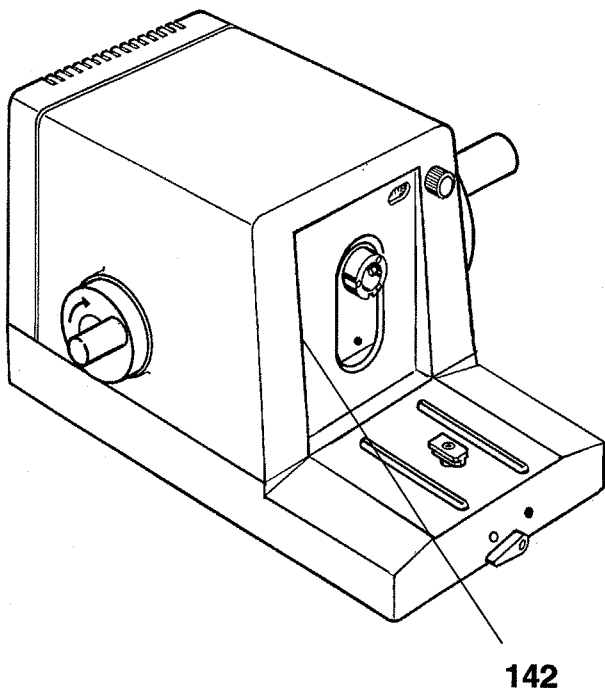
- Always start with a clearance angle of  $0^\circ$  selected on the scale on the right side of the knife holder base.

- If the sectioning results are not satisfactory, the clearance angle has to be increased in approx.  $2^\circ$  increments.

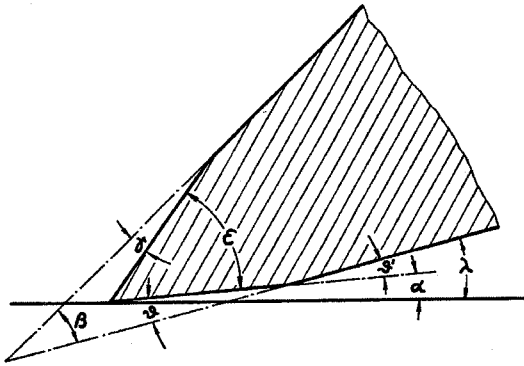
When using disposable blades, please note that all disposable blades use different clearance angles. There is no common rule to suit the great number of different specimen types. The appropriate clearance angle should be selected from one specimen to another.

- The section thickness is selected with knob (143).
- The selected micron value is indicated in the display (144).

- Cutting is done via the large handwheel (141) either turning it by  $360^\circ$  or swinging the wheel by  $\pm 90^\circ$  in its upper position.



## 12. Mikrotome knives



The schematics illustrate the geometry of cutting edge.

$\alpha$  = clearance angle (to be adjusted on the knife holder)

$\beta$  = wedge angle

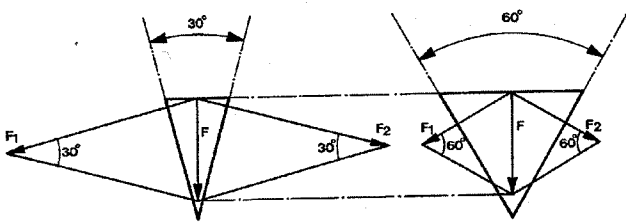
$\gamma$  = upper exterior facet angle

$\delta'$  = lower exterior facet angle

$\delta' = \gamma$  = alternative angle to  $\delta$

$\epsilon$  = facet angle

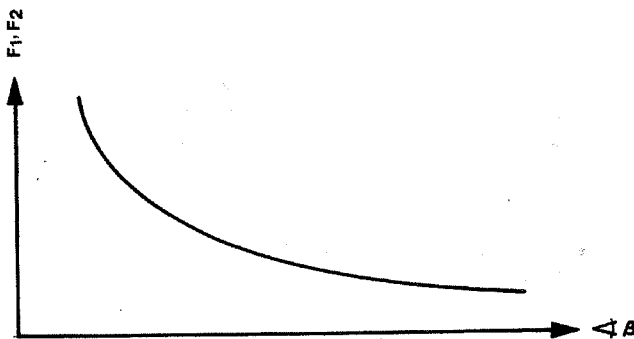
$\lambda$  = inclination angle ( $\alpha + \delta'$ )



The side forces  $F_1$  and  $F_2$  are dependent on the facet angle  $\epsilon$ :

At a small facet angle (e.g.  $30^\circ$ ), great side forces are produced, which act on the specimen.

At a large facet angle (e.g.  $60^\circ$ ), the side forces are lower. The stability of the cutting edge is increased, however.  $F$  remains constant.



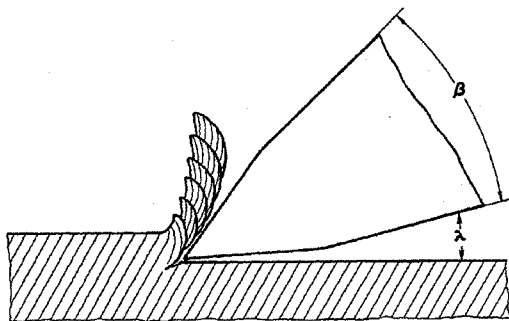
Interrelationship between facet angle and side forces:

The formula for calculating the side force is as follows:

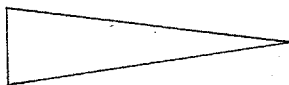
$$F_1 = F_2 = \frac{F}{2 \sin \frac{\epsilon}{2}}$$

$F$  = Cutting force

$F_1 = F_2$  = Side forces that separate the material.



If the inclination angle  $\lambda$  or facet angle  $\epsilon$  is very large, the sections will tear.



c-profile knives  $\epsilon = 27^\circ$  (for paraffin)



d-profile knives  $\epsilon = 45^\circ$  (for plastics)

## 13. Trouble shooting

### Cause

### Remedy

#### 13.1 No ribboning

13.1.1 Paraffin is too "hard".

Re-embed the specimen in a paraffin with a lower melting point.

Or:

Immerse the entire block in a paraffin with a lower melting point and trim the block so that a thin "sticky" layer remains on the upper and lower edge of the block.

13.1.2 Clearance angle is too big.

Decrease the knife inclination towards the block.

13.1.3 Selected section thickness is too big..

Correct section thickness feed.

13.1.4 Cutting edge of the knife is blunt..

Exchange, reposition or sharpen the knife.

13.1.5 None of the above applies

Unroll the section and hold it lightly against the knife using a brush.

## 13. Trouble shooting

Cause	Remedy
<b>13.2 Crooked ribbons</b>	
13.2.1 The individual sections are wedge-shaped.	Trim the block so the upper and lower edge of it are parallel to each other and of the same width.
13.2.2 Edges of the block are parallel to each other, however not to the knife edge.	Clamp the block securely and orient via the specimen clamp orientation so the upper and lower edge are parallel to the knife edge.
13.2.3 Irregularities in the knife edge.	Move the knife laterally until a part of the knife edge without irregularities is used for sectioning.
13.2.4 The block consists of paraffins of different consistencies.	Remove inhomogeneous paraffina and re-embed.
13.2.5 The sides of the block have different temperaturea, caused by light sources, heaters, or draughts.	Avoid places with these adverse temperature conditions.  The microtome should be set up in a place where the temperature is uniform. Wait until the block and microtome have assumed the temperature.
<b>13.3 Sections vary in thickness or are skipped (i.e. only every second section is cut). Thick and thin sections.</b>	
13.3.1 Knife inclination angle is too small, and thus the clearance angle is too small as well. A cutting stroke, which did not produce a section, is followed by a return stroke, in the course of which the tissue is compressed.	The optimum inclination angle can be found by systematically trying different angles.
13.3..2 lack of stability in clamping devices.	Check all screwed connections and clampings and readjust, if necessary.

## 13. Trouble shooting

Cause	Remedy
<b>13.4 Sections are extremely compressed, wrinkled and jammed together.</b>	
13.4.1 Cutting edge is blunt.	Resharpener or exchange the knife, or maybe reposition sideways.
13.4.2 Room temperature is too high.	Cool the trimmed block and knife in very cold or ice water immediately before sectioning.  Or:  Re-embed the tissue in "harder" paraffin.
13.4.3 Knife inclination angle is too small so the facet bevel rubs over the block.	Increase angle (see 13.3.1).
13.4.4 Knife edge is contaminated with paraffin	Wipe both sides of the knife with a cotton moistened with cleaning fluid. When cleaning the knife, always wipe from the bottom towards the knife edge; never wipe from the knife edge downwards, as thus the cutting edge would be damaged and there would also be an enormous risk of injury!
13.4.5 The paraffin has been used before several times. Sections break etc.	Re-embed the specimen in new paraffin.

## 13. Trouble shooting

### Cause

### Remedy

#### 13.5 Sections crumble and specimen may tear out.

13.5.1 Material was incompletely dehydrated or not properly cleared.

Dehydrate and clear.

13.5.2 When soft and mushy, material is incompletely infiltrated with paraffin.

Reinfiltrate and re-embed (can rarely be remedied if dehydration was incomplete).

13.5.3 Alcohol not completely removed by clearing fluid before infiltrating with paraffin.

Remove alcohol.

13.5.4 Specimen stayed too long in paraffin bath, or paraffin bath was too hot.

Can hardly be remedied as mostly the tissue is irretrievably damaged.

13.5.6 Specimen (tissue) is too hard for the paraffin used.

Re-embed in a "harder" paraffin or plastic (HistoResin).

#### 13.6 Split ribbon, or lengthwise striation in the ribbon.

13.6.1 Nicks in the knife.

Use another part of the cutting edge, or sharpen the knife.

13.6.2 Knife edge is dirty.

Clean (see 13.4.4).

13.6.3 Specimen may be too large for paraffin method.

Try celloidin embedding.

13.6.4 When working with disposable blades: Teflon coat is coming off.

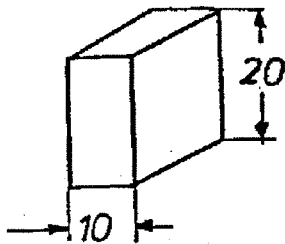
Clean the blade or do one section of considerable thickness (20 - 30  $\mu\text{m}$ ).

## 13. Trouble shooting

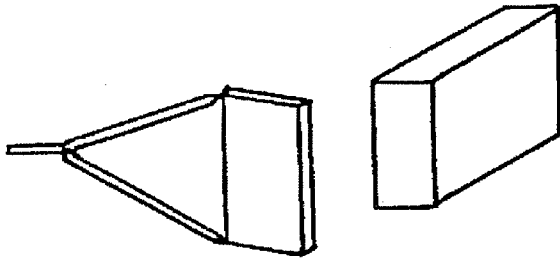
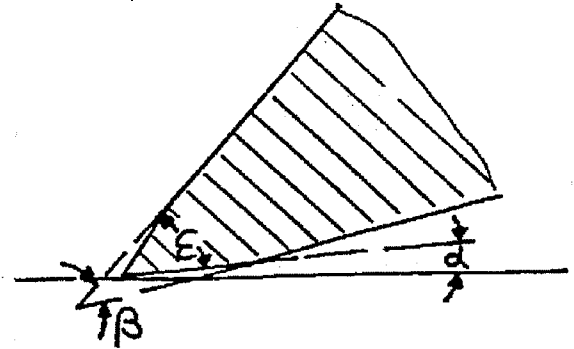
Cause	Remedy
13.6.5. Scratches are caused by hard particles, especially contaminations in the paraffin.	Filter or decant the melted paraffin.
Crystals from remainders of killin fluid. Calcareous or siliceous particles in the block.	Wash more thoroughly! Decalcify; desilicify.
<b>13.7. Knife rings on cutting stroke. Sections are scratched and show chatter marks.</b>	
13.7.1 Knife inclination angle is inappropriate.	See 13.3.1.
<b>13.8. Section stick to the knife.</b>	
13.8.1 Knife edge is dirty.	Clean the knife.
<b>13.9. Undulation in the surface of the sections.</b>	Readjust angles.
<b>13.10. Sections fly away. They stick to the microtome or other objects close to the microtome.</b>	Air humidity must be increased.

## 13.1 How to set the right clearance angle

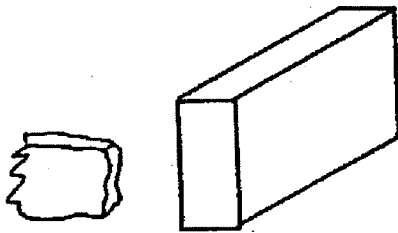
How to set the appropriate clearance angle:



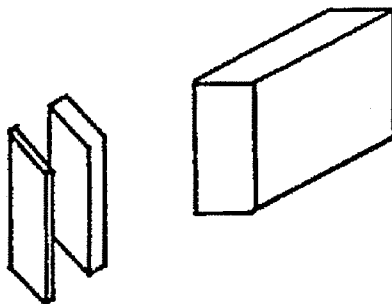
$\alpha$  = Clearance angle  
 $\beta$  = Wedge angle  
 $\epsilon$  = Facet angle



Section done at an optimal clearance angle



Compressed section



Thick-thin sections

1. Secure a paraffin block of 10 mm width and 20 mm height (any length) in the specimen clamp. Use compasses to compare the length of the section with the height of the block. **The clearance angle is optimal when section and block are of the same height.** However, in reality there will always be a very slight compression which is normal.
2. **Too much compression**
  - The set clearance angle is too big,
  - or: the paraffin is too warm,
  - or: the melting point of the paraffin is too low, compared to the room temperature.
3. **Thick-thin section**
  - the set clearance angle is too small
  - or: the clearance angle is even zero causing the knife edge to squeeze the block.
4. **Sections curl - no ribbing**
  - the melting point of the paraffin is too high compared to ambient temperature; i.e. the paraffin is too cold.
  - or. the set section thickness is too big.



## 14. Ordering information

Adapters for orienting specimen holders .....	0402 09157
Adapters for non-orienting specimen holders .....	0399 09262

### Specimen clamps

Orienting standard specimen clamp .....	0402 09173
Non-orienting standard specimen clamp .....	0402 09378
Vee insert for round specimens .....	0402 09299
Orienting universal quick release cassette clamp .....	0402 09166
Non-orienting universal quick release cassette clamp .....	0394 09174

<b>Knife holder base</b> for knife holder N,E Types I and II and Z .....	0427 21114
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### Knife holders

Standard knife holder N .....	0402 09236
Disposable blade holder E fType I, for high profile blades .....	0402 09376
Disposable blade holder E Type I, for low profile blades .....	0402 09386
Locating plate for high profile blades .....	0402 09471
Locating plate for low profile blades .....	0402 09472
Disposable blade holder E Type II .....	0427 21163
Blade rail for high profile blades (Type II) .....	0427 21173
Blade rail for low profile blades (Type II) .....	0427 21174
Quick release knife holder Z .....	0402 09374

### Sectioning accessories for paraffin sections

Standard knife, 16 cm, profile c .....	0216 07100
Standard knife, 16 cm, profile d .....	0216 07132
Variable plastic case for 16 cm knives .....	0213 11140
Dispenser with 50 disposable blades, low profile .....	0358 13583
Dispenser with 50 disposable blades, high profile .....	0358 12881

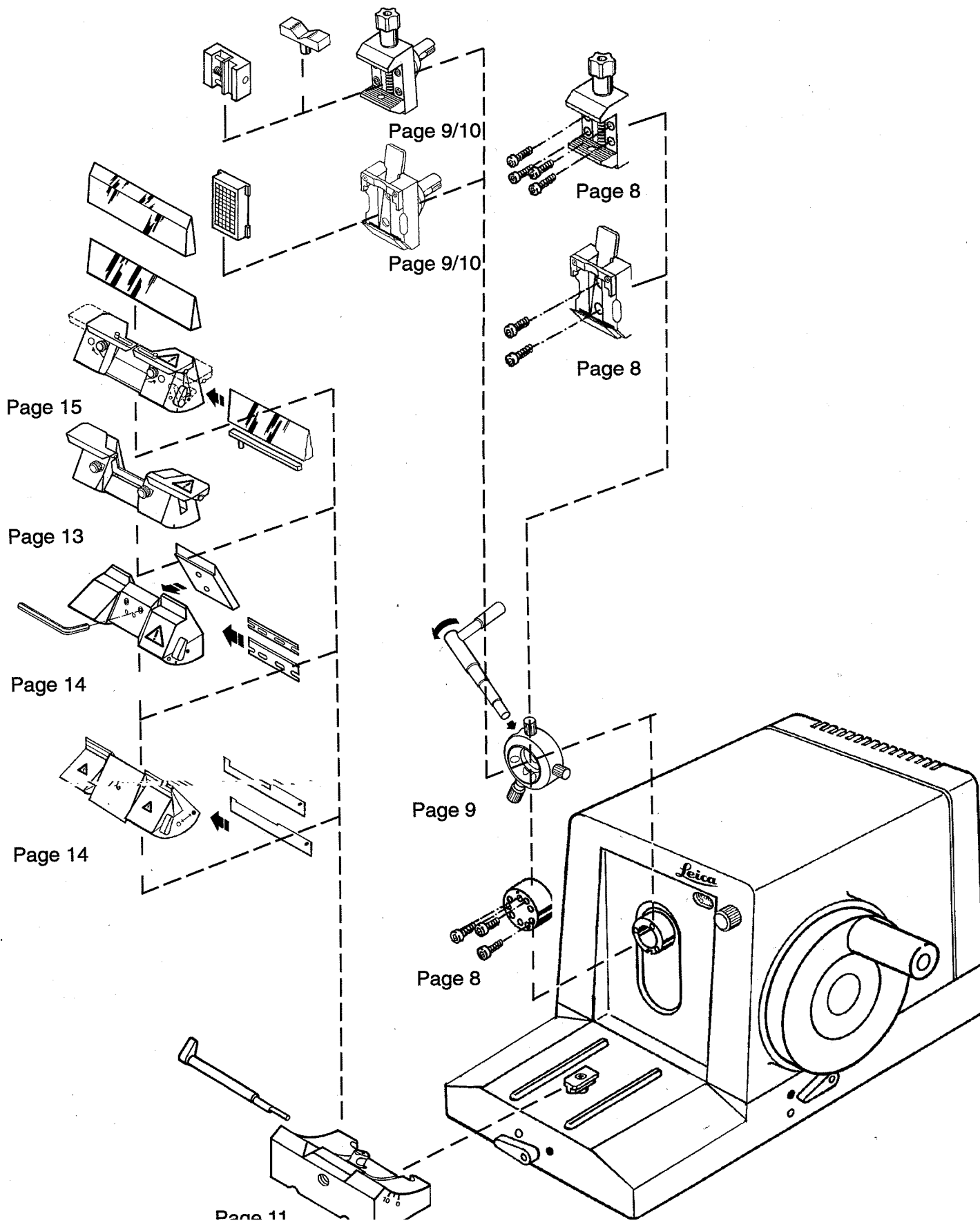
### Paraffins

Histowax 2,5 kg .....	0374 08585
Histowax, large bag, 25 kg .....	0374 14374

### Embedding cassettes

250 cassettes, white .....	0394 12312
dto., grey .....	0394 08972
dto., yellow .....	0394 08974
dto., red .....	9394 08976
dto., green .....	0394 08978
dto., blue .....	0394 08980
250 lids, white .....	0394 12315
dto., grey .....	0394 08982
dto., yellow .....	0394 08984
dto., red .....	0394 08986
dto., green .....	0394 08988
dto., blue .....	0394 08990

# 15. RM 2025 - Outline Drawing



## 16. Notes