

Standard Operating Procedure

SOP194

Title: Use and maintenance of Leica RM2125 RTS rotary microtome.

Location: H27

1. PURPOSE

To describe the use and maintenance of the Leica RM2125 RTS rotary microtome that is located in the CL2 CBE laboratory H27.

2. SCOPE

This SOP applies to CBE personnel using the Leica RM2125 RTS rotary microtome for the sectioning of fixed soft tissues that have been embedded in wax or other hard polymers. The microtome is assigned to H27 which is used for histological processing and this system should not be transferred to another lab. Only authorised and trained personnel may use the microtome.

3. RESPONSIBILITIES

3.1. Authorised laboratory staff shall:

- (i) Work in accordance with the instructions for operation and maintenance detailed in this SOP, Risk Assessment and the operator manual.
- (ii) Carry out the routine inspection, cleaning, and maintenance of the equipment, as required.
- (iii) Record any adverse events and alarms that indicate non-conformance or malfunction on the Maintenance Record and notify the Laboratory Manager/Responsible Person.

3.2. The Responsible Person/Laboratory Manager shall:

- (i) Ensure that authorised laboratory personnel are given suitable information, instruction, training and supervision in the correct use and maintenance of the equipment.
- (ii) Investigate any reported problems, adverse event, alarms, or non-conformities associated with equipment usage.
- (iii) Organise the maintenance, repair, or servicing of the equipment by trained and authorised contract / service personnel.

4. EQUIPMENT AND MATERIALS

1. Leica RM2125 RTS rotary microtome and accessories
2. Blades
3. Brush
4. Appropriate PPE (labcoat, gloves, overshoes and eye protection, where appropriate)

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5. PROCEDURE

5.1. Description of the system

The Leica RM2125 RTS is a manually operated rotary microtome for creating thin sections of specimens of varying hardness for use in routine and research laboratories in the fields of biology, medicine, and industry. It is designed for sectioning soft paraffin specimens as well as harder specimens, as long as they are suitable for being cut manually.

Note: Operating temperature range is 10°–35°C, and relative humidity range is 20-80% non-condensing humidity. Section thickness range is 0.5- 60µm.

5.2. Overview of components

Leica RM2125 RTS (from the right)



Leica RM2125 RTS (from left)



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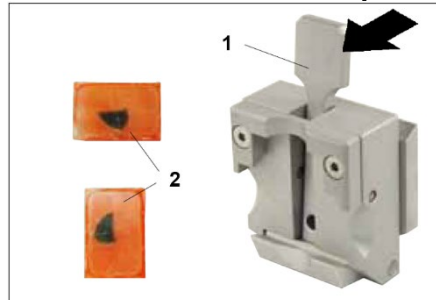
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5.3. Operation

5.3.1. Clamping specimen on universal cassette clamp

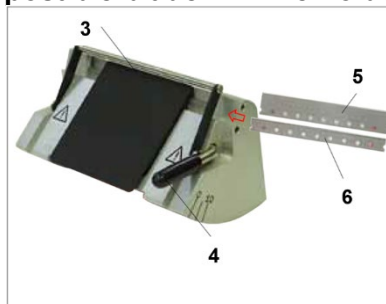


1. Always clamp the specimen block BEFORE clamping the knife or the blade.
2. Lock the handwheel and cover the knife edge with the knife guard prior to any manipulation of knife or specimen, prior to changing the specimen block and during all work breaks.
3. Activate the handwheel lock by allowing the handwheel handle to lock in place.
4. To insert a specimen into the specimen clamp, push lever (1) forwards.
5. Mount the cassette (2) horizontally or vertically as required.
6. To clamp the cassette, release the lever.

Note: Before clamping the cassette into the universal cassette clamp, remove excess wax on the outside of the cassette to ensure that the cassette clamps in securely.

Wax deposits on the outside of the cassette can make the universal cassette clamp dirty. The dirt prevents the cassette from clamping in securely and can lead to sections being too thick or thin, chattering within the section and, in the worst-case scenario, damage to the specimen. Prior to sectioning, the user has to verify that the specimen is clamped securely and, if necessary, remove wax deposits from the universal cassette clamp.

5.3.2. Clamping the knife / disposable blade in knife holder E



1. Carefully insert knife or disposable blade into the knife holder and clamp.
2. Fold knife guard (3) downward.
3. To insert the blade, rotate clamping lever (4) forward.
4. Carefully push in the blade (5 or 6) from the side.
5. To clamp the blade, rotate clamping lever (4) back upwards.

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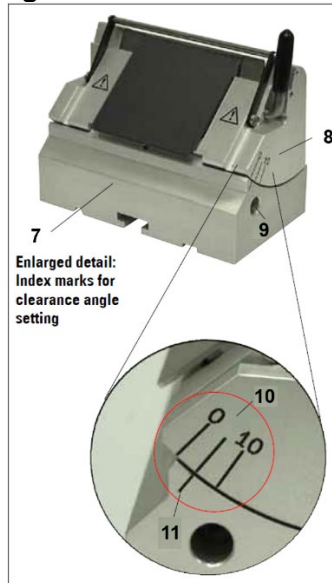
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5.3.3. Adjusting the clearance angle



1. The index marks (0°, 5° and 10°) for adjustment of the clearance angle (10) are located on the right side of the knife holder (8).
2. There is also an index mark (11) on the right side of the knife holder base (7) which serves as a reference point when adjusting the clearance angle.
3. When using the knife holder base (non-orientable), loosen the Allen screw to release the clamp by turning Allen key No. 4 (9) counterclockwise.
4. When using the knife holder base (with lateral movement), rotate the lever on the right side of the knife holder base counterclockwise.
5. Move the knife holder until the index mark of the desired clearance angle coincides with the reference line on the knife holder base.
Example: Enlarged detail showing a clearance angle setting of 5°.
Note: The recommended clearance angle setting for knife holder E is approx. 1° - 3°.
6. Firmly hold the knife holder in this position and rotate the lever (9) or Allen screw (depending on the knife holder base used) clockwise to clamp it.

5.3.4. Specimen retraction (specimen orientation)



- (i) To prevent the knife or blade from touching the overlapped specimen while returning to the upper end position, the specimen is retracted 40 µm when retraction is activated.

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Note: The user can switch the specimen retraction ON or OFF.

- (ii) To do so, use an Allen key with handle, No. 4 (included in the delivery package) and turn the screw depicted in the detail picture (Fig. 13a) so that the red dot indicates "OFF" = retraction is disabled. Red dot on "ON" means = retraction is switched on.

- (iii) Important if retraction is enabled:

The specimen must NOT be oriented or approached in the retraction phase (if the black dot on the handwheel is visible – see enlarged detail Fig. 13 – you are in the sectioning phase)! The previously retracted specimen will advance by the retraction value PLUS the selected section thickness before the next section.

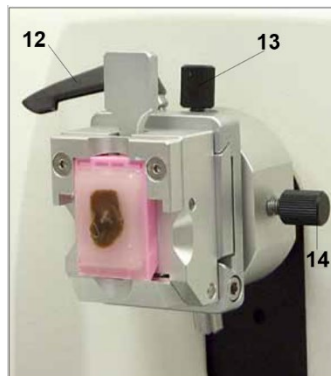
There is a danger of the specimen and knife being damaged by a section that is too thick.

The same applies to the "rocking mode", during which the specimen is trimmed by rocking motion (no complete rotation of the handwheel).

Carry out "rocking mode" ONLY in the sectioning phase – NEVER in the retraction phase!

5.3.5. Orienting the specimen

The specimen orientation allows for simple position correction of the specimen surface when the specimen is clamped into place.



1. Run the specimen to the rear end position by turning the coarse feed wheel.
2. Loosen the clamping lever in front on the microtome base plate and slide the knife holder base with knife holder until it is almost in front of the specimen.
Note: Specimen blocks must NOT be oriented during the retraction phase!
If a block is oriented during retraction, the block will advance by the retraction value PLUS the selected section thickness before the next section.
This may cause damage to both specimen and knife!
3. Move the specimen holder fixture to the upper end position by turning the handwheel and engage the handwheel lock.
4. To release the clamp, turn the eccentric lever (12) counterclockwise.
5. Turn setscrew (14) to orient the specimen in north-south direction. Turn setscrew (13) to orient the specimen in east-west direction.
6. To lock the current orientation, turn the eccentric lever (12) clockwise.

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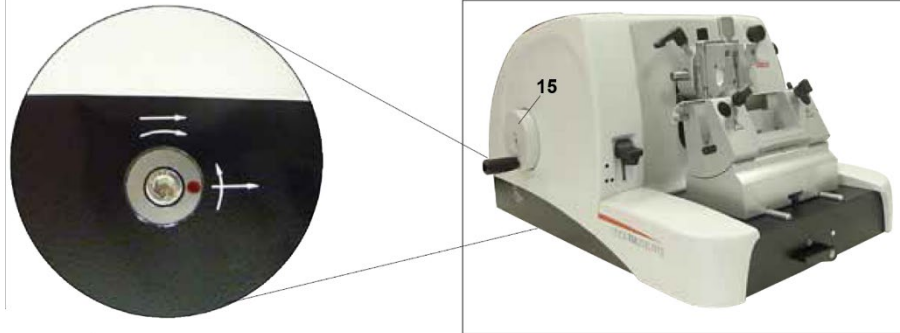
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5.3.6. Cutting (trimming) the specimen



5.3.6.1. Coarse feed wheel (15)

The coarse feed serves for a fast horizontal forwards movement of the specimen – towards the knife – and backwards – away from the knife.

The instrument can be used optionally with either clockwise rotation or counterclockwise rotation for the coarse feed wheel (15).

To do so, use an Allen key with handle, No. 4 (included in the delivery package) to turn the screw as shown in the detail picture.

1. Red dot at 3 o'clock: turning the coarse feed wheel counterclockwise (see curved arrow) means the specimen advances.

Turning the coarse feed wheel clockwise means the specimen retracts (away from the knife).

2. Red dot at 12 o'clock: turning the coarse feed wheel clockwise (see curved arrow) means the specimen advances.

Turning the coarse feed wheel counterclockwise means the specimen retracts (away from the knife).

Note: When the rear- or front-end position is reached, the coarse feed wheel will be difficult to rotate (if you continue turning it at this point anyway, the torque limitation will be exceeded – this is not a malfunction!).

In the front-end position, no more feed motion takes place.

5.3.6.2. Trimming the specimen with the coarse feed



1. Release the handwheel lock. To do so, pull the handle of the handwheel out to the right and use the lever (19) to release the brake.

- Brake engaged
- Brake released

2. Bring the specimen closer to the knife by rotating the coarse feed wheel and trim it by simultaneously rotating the handwheel (18) until the desired specimen plane is reached.

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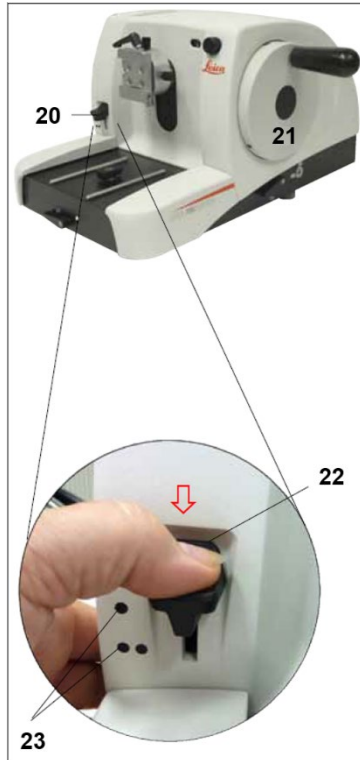
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5.3.6.3. *Trimming the specimen by setting a large section thickness*

1. Set a correspondingly large section thickness (e.g. 50 μm) using the section thickness setting knob (17) at the front of the microtome on the right.
The current setting is displayed in the section thickness window (16).
2. Trim the specimen by rotating the handwheel (15) until the desired specimen plane is reached.

5.3.6.4. *Trimming with the mechanical trimming function*



The Leica RM2125 RTS is equipped with a mechanical trimming function that is activated via the trimming lever (20).

The trimming lever has 3 click stops:

0 μm , 10 μm , and 50 μm .

The points (23) mark the two trimming stages:

● = 10 μm

●● = 50 μm

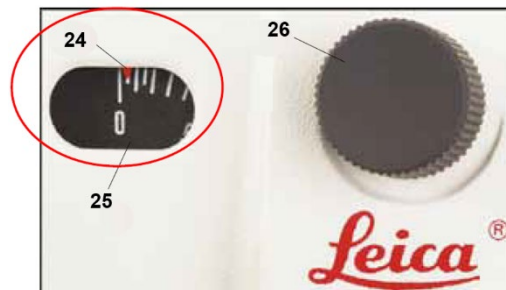
1. To activate the trimming function, press the lever downwards into the desired position and keep it pressed down.
2. With each rotation of the handwheel, a feed motion of 10 μm or 50 μm takes place.
3. After you let go of the lever, it springs back to its original position (zero position). The trimming function is thereby deactivated.

Note: The section thickness that has been set is not added to the selected trimming value.

If the section thickness that has been set is greater than the selected trimming value, the section thickness is fed.

4. Bring the specimen closer to the knife by rotating the coarse feed wheel.
5. Select the desired trimming stage.
6. Trim the specimen by rotating the handwheel (21) until the desired specimen plane is reached.
7. Release the trimming lever (22).

5.3.7. Sectioning



Note: Always rotate the handwheel at a uniform speed. The rotation speed of the handwheel must be adapted to suit the hardness of the specimen.

For harder specimens, use a slower speed.

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CAUTION: The handwheel continues turning if it has been rotated very quickly and then released – this can cause crushing or other injuries!

8. Set the desired section thickness using the section thickness setting knob (26) at the front of the microtome on the right, or check the set value in the viewing window (25). The red indicator (24) indicates the selected section thickness (on the scale).
9. Use a different area of the cutting edge for trimming than for sectioning.
10. To do so, move the knife holder laterally on the knife holder base accordingly (see Chap. 5.9.7) or, when using the knife holder base without lateral movement, move the knife or disposable blade in the knife holder.
11. For sectioning, turn the handwheel (15) evenly in a clockwise direction.
12. Pick up the sections and mount them on microscope slides.

5.3.8. Changing specimens

Note: Lock the handwheel and cover the knife edge with the knife guard prior to any manipulation of knife or specimen, as well as prior to changing specimens and during all work breaks!

1. Move the specimen to the upper end position by turning the handwheel and engage the handwheel lock.
2. Cover the sectioning edge with the knife guard.
3. Remove the specimen from the specimen clamp and mount a new specimen.
4. Run the object clamp with the coarse feed back far enough until the new specimen can start being cut.

5.4. Cleaning and Maintenance

5.4.1. Cleaning

Note: Always remove the knife / blade before detaching the knife holder from the instrument. Always put the knives back into the knife case when not in use! Never place a knife anywhere with the cutting edge facing upwards and never try to catch a falling knife! When using cleaning agents, observe the manufacturer's safety instructions and the laboratory regulations valid in the country of use. Do not use any of the following for cleaning the outside surfaces of the instrument: alcohol, detergents containing alcohol (window cleaner!), abrasive cleaning powders, solvents containing acetone or xylene. Xylene or acetone will damage the finished surfaces! Ensure that liquids do not enter the interior of the instrument during cleaning! Move the specimen to the upper end position by turning the handwheel and engage the handwheel lock.

Before each cleaning, carry out the following preparatory steps:

- Move the specimen clamp to the upper end position and activate the handwheel lock.
- Remove the blade from the knife holder and insert it in the receptacle at the bottom of the dispenser, or remove the knife from the knife holder and put it back in the knife case.
- Remove knife holder base and knife holder for cleaning.
- Remove the specimen from the specimen clamp.
- Remove section waste with a dry brush.
- Remove specimen clamp and clean separately.

Instrument and outside surfaces:

- If necessary, the varnished outside surfaces can be cleaned with a mild commercial household cleaner or soap water and then be dried with a moist cloth.

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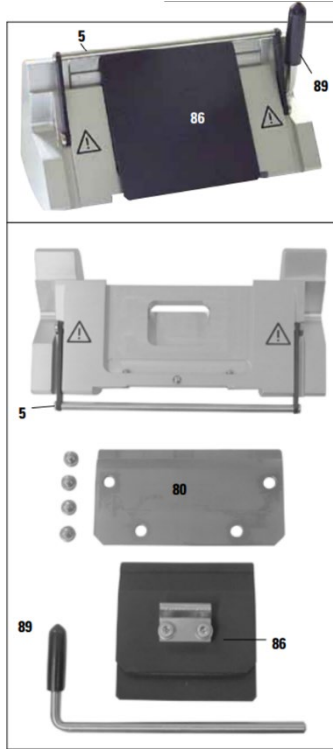
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Knife holder E



Take the knife holder apart for cleaning. The front pressure plate can be removed for cleaning. To do so, proceed as follows:

1. Fold knife guard (5) downward.
2. Rotate the blade clamping lever (89) downward.
3. Carefully remove the blade and dispose of it properly.
4. Pull out the clamping lever (89) sideways.
5. Remove pressure plate (86).
6. Clean all parts of the knife holder.

Note: If several knife holders are cleaned at the same time, the parts must NOT be mixed up! Failure to adhere to this may result in sectioning problems!

For cleaning and removal of paraffin, do not use xylene or cleaning fluids containing alcohol (e.g. glass cleaner).

7. Lay the removed parts on an absorbent cloth into the drying chamber (up to a max. of 65 °C) and let the paraffin contamination run off. **Note:** There is a burn hazard when removing the parts from the drying chamber (65 °C). Wearing safety gloves is recommended!

8. After cleaning the moving parts, apply a thin coat of drive part oil to them.

9. Reassembly takes place in reverse order.

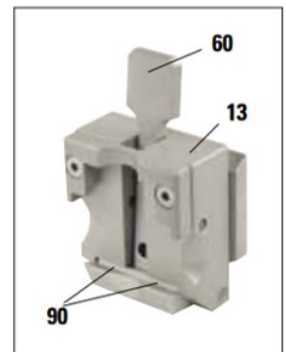
10. When performing the installation, make sure that the upper edge of the pressure plate (86) is parallel and level with the upper edge of the rear pressure plate (80). If necessary, adjust the pressure plates.

Universal cassette clamp

1. Detach cassette clamp (13) for a thorough cleaning, removing all paraffin residues.
2. For cleaning, do not use xylene. Use xylene substitutes or paraffin removers such as "Para Gard."
3. The cassette clamp (13) can also be placed in a drying chamber heated to a maximum of 65 °C, until the liquid wax escapes.

Note: There is a burn hazard when removing the parts from the drying chamber (65 °C). Wearing safety gloves is recommended!

4. Remove paraffin residues with a dry cloth.
5. After such a cleaning procedure in an oven, always be sure to lubricate the axle and spring of the clamping lever (60).



5.4.2. Maintenance

Note: Only authorized and qualified service personnel may access the internal components of the instrument for service and repair!

The instrument is basically maintenance-free. To ensure trouble-free operation of the instrument over a long period of time, the following is recommended:

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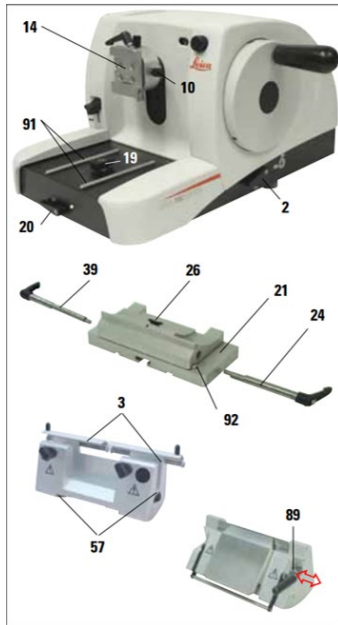
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- Have the instrument checked at least once a year by a qualified service technician authorized by Leica. At the end of the warranty period, enter into a service contract. For more information, please contact your local Leica technical service center.



- Clean the instrument every day.
-
- Once a month, lubricate the following parts with the included oil No. 405 (1-2 drops are enough):
 - The moveable parts of the specimen holder fixture (10) and the dovetail receptacle (Fig. 14).
 - T-piece (19) on the microtome base plate.
 - Clamping levers (2) and (20) on the microtome.
 - Guide rails (91) for the knife holder base on the microtome base plate.
 - The clamping lever (39) and (24) on the right and left on the knife holder base.
 - The guide (92) of the lateral movement on the knife holder base (21).
 - The T-piece (26) on the knife holder base (21).
 - The sliding surfaces of the knife guard (3) and knurled nuts (57) on knife holder N.
 - The clamping lever (89) on knife holder E.
 - Shaft (90) of the clamping lever of the cassette clamp (see Fig. 36).

Special Safety Note: Equipment contains sharp edges. Ensure you are wearing PPE, have been fully trained and use caution when using.

5.5. Equipment Malfunction

- (i) If any part of the equipment fails or malfunctions, seek advice from the Laboratory Manager or Responsible Person. Unauthorized repairs may damage the microtome or alter its functionality, which may void the warranty. With permission of the Responsible Person, consult the operator's instruction manual to access fault finding and troubleshooting procedures.
- (ii) If any problem cannot be rectified according to the troubleshooting procedures a "do not use" notice must be put on the microtome and the Laboratory Manager informed. The manufacturer must be contacted for advice and external maintenance or servicing might need to be arranged.
- (iii) All problems and corrective actions must be recorded in the equipment maintenance log.
- (iv) If the equipment fails to work or malfunctions and cannot be rectified according to troubleshooting procedures detailed in the Operator and Users Manuals the Laboratory Manager must be informed and a "Do Not Use" notice should be posted on the equipment. Contact the manufacturer for advice and coordinate with the Lab Manager for external maintenance and servicing.

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6. DOCUMENTATION

- 6.1 Equipment Maintenance Log
- 6.2 Microtome Training Agreement

This form can be found on the Loughborough University LEARN module for the Centre of Biological Engineering under QS-Form templates.

6.1. SOP Version History

Version Reviewed	Date Revised/ Reviewed	Revision Summary	New Version Number
		[Insert specific changes from previous SOP] < e.g. changes in accountabilities, process steps, deviation actions, or records >.	

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