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Cutting FFPE Sections for Imaging Mass Cytometry

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1 Works for me

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ABSTRACT

FFPE block are cut into thin sections.

MATERIALS

NAME	CATALOG #	VENDOR
FFPE block		
Rotary Microtome	HM355S	Thermofisher
FFPE Block Cooling Unit	TES99.430	
Stretching table		
Deionized Water		
Microtome Blade		
Brushes		
Glass Slides Superfrost 25x75x1.0mm	10.0344.01	Thermo Fisher Scientific

SAFETY WARNINGS

The microtome blade is very sharp, care must be taken to avoid injury.

Cooling the FFPE Block

1 1.1 Cool down the FFPE block

- a. Switch on the cooling device and set it to 4°C
- b. Put the FFPE block into the cooling device.

Switching on the microtome

7 1.2 Prepare the microtome

- a. Connect the cooling unit in the back with the cable and switch it on at the front.
 - b. Switch on the microtome using the button at the back of the machine.
 - c. Install the water bath with the filter.
- d. Add deionised water to the water bath and lower the compartment until the grid is slightly covered with water.
 - e. Place a fresh blade in the blade holder.
- f. Check the water flow. If water is not flowing rinse the tubing with a syringe filled with water.
 - g. Set the water flow to 3 or 4.
 - h. Fix the precooled FFPE block to the specimen clamp holder.
- i. Adjust the angle between blade and FFPE block by unlocking the clamping lever and then adjusting the orienting screws accordingly.
- j. Set the trimming (Trim) and cutting (Feed) thickness by pressing the top knob once or twice, respectively.
- k. Unlock the wheel with the mechanical hand wheel brake on the right-hand side underneath the wheel.

1.3 Switch on the stretching table

a. Switch it on and set it to 37°C.

Cutting

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3 1.4 Trimming

- a. Always use a new sharp blade.
- b. Trim carefully and make sure that your cutting angle is fine, if needed readjust.
- c. As soon as you think you are cutting tissue, take up a cut and have a look under the microscope.
- d. Exchange the water bath before starting cutting sections that you intend to collect.

1.5 Cutting

- a. Cut in a uniform and gentle rotation.
- b. Let the cut slide down into the water bath.
- c. Wait until the cut is flattened before taking it up on a glass slide.
- d. Wipe the glass slide dry on the bottom side before placing it on top of the tretcher
- e. Exchange water on a regular basis if sample produces a lot of paraffin debris and between samples.

Switching off

4 1.6 Switching off the microtome

- a. Block the wheel to avoid any injuries as soon as you are done cutting
- b. Dispose of the blade
- c. Stop the water flow by turning the wheel down to 0.
- d. Discard the water, clean the water bath, filter and grid, place them to dry.
- e. Clean the water slide.
- f. Clean the microtome from paraffin debris.
- g. Switch off the microtome at the back and disconnect the cooling block.
- h. Switch off the FFPE Block cooling device.

Section drying and storage

5 1.7 Drying sections

a. Dry the sections overnight or for **1-2 hours** at 37°C. Do not extend the drying for too long.

1.8 Storage of FFPE sections

a. Store all FFPE fresh cut and dried sections at -80°C or stain them within a week.