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# **©** Cutting FFPE Sections for Imaging Mass Cytometry

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We use this protocol and it's working

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#### Abstract

FFPE block are cut into thin sections.



### **Materials**

### **MATERIALS**

- **⋈** FFPE block
- **☒** Rotary Microtome **Thermofisher Catalog** #HM355S
- Stretching table
- **X** Deionized Water
- Microtome Blade
- **X** Brushes
- **⊠** Glass Slides Superfrost 25×75×1.0mm **Thermo Fisher Scientific Catalog #**10.0344.01

# 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

### 2 **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

### 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 stretcher.
- 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.