



Samples preparation guidelines for 10X Genomics Visium, Fresh-Frozen Non-Cytassist

These guidelines relate to the Visium non-Cytassist protocol for the part of sectioning, fixation, staining and imaging if they are NOT performed at the EPFL Histology core facility.

Interactions with GECF on experiment day

- **IN SHORT:** After the histology part, bring to the GECF the tissues positioned on the Visium slide and fixed/stained/imaged as described in 10XG protocols. Give us sample names, positions on the slide (with a clear scheme) and desired permeabilization time (prior optimization necessary). Later send us the images.

Slides preparation

Most of the guidelines are in our main guidelines document. But here are some additional considerations gathered from 10XG documents:

Key Considerations

Slide Handling (before sectioning)


- Equilibrate Visium slides to cryostat temperature before cryosectioning. Just put the slide in cryostat.
- Store unused slides in original packaging and keep sealed. DO NOT remove desiccant. If necessary, store original packaging in a secondary container such as a resealable bag.

Freezing and Embedding

- Snap freeze samples in a bath of isopentane and liquid nitrogen.
- Store frozen samples at -80°C in a sealed container for long-term storage prior to embedding.

Cryosectioning

- Equilibrate OCT tissue block to the cryostat chamber temperature for 30 min.
- Place tissue sections on the Capture Area* within the fiducial frame on the slide.



*Visium Gateway Gene Expression Slide contains 2 Capture Areas

Slide Handling (after sectioning)

- Maintain slides containing sections in a low moisture environment.
- Keep slides cold and transport slides on dry ice.
- DO NOT leave slides at room temperature.

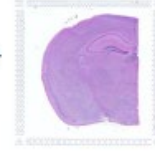
Sample Storage

- Store slides individually in a sealed container at -80°C for up to 4 weeks to avoid multiple freeze thaw cycles. If using an unsealed slide mailer, store in a secondary sealed container, such as a resealable bag.

Sample Preparation

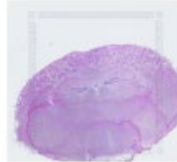
Tissue Placement

- Tissue samples should be placed on the capture area within the fiducial border

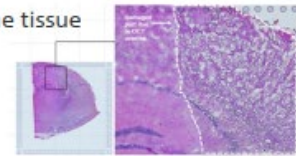


- Avoid:

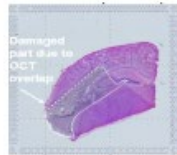
- Covering the fiducial border



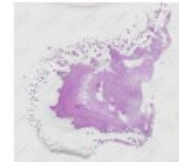
- OCT overlapping the tissue



- Folding of the tissue



- Condensation formation



Versions log

- vA.02: removed elements now in our main guidelines document