

Tissue Sectioning for CODEX imaging in the Phenocycler Fusion 2.0

Guidelines:

Tissue slides should ideally be prepared freshly, however storage at 4°C for up to 6 months is possible.

Optimization of tissue assembly and placement is recommended to reduce the number of total slides to be imaged – reducing costs and imaging time.

It is critical not to exceed a thickness of 10 µm as this may disrupt the autofocus capabilities of the microscope.

For best results, the tissue should be completely adhered to the slide with minimal tears or folds. To ensure that tissue sections are not damaged, it is critical that the tissue slides are stored properly and not stacked on top of one another.

Recommended Materials:

We recommend sectioning on SuperFrost Plus™ microscopy slides (e.g. Fisherbrand™ Cat.No. 12-550-15 or EpreDia™ J1800AMNZ).

For tissues prone to detachment or in special cases alternative slides such as SuperFrost Ultra Plus™ or coated slides can be considered.

Sectioning:

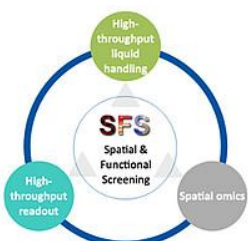
The ideal tissue thickness depends on the cellularity and scientific evaluations. For dense tissues such as Lymphoma, we recommend 2µm thickness.

Standard sectioning procedures can be used as established locally. Afterwards, the slides should be dried overnight at room temperature on a clean surface (with the tissue facing up) or on the angled slide holder. Afterwards, slides should be shipped at 4°C with a desiccant.

Tissue arrangement:

The imageable area is 35 x 18mm large and positioned on the microscopy slide as detailed in Figure 1. Tissue outside of this area (under the adhesive portion of the flow cell) can affect bonding of the flowcell and cause experiment failures. Ensure the tissue is placed on the positively charged side of the slide to ensure proper adhesion.

Tissues can be arranged in a number of ways to ensure maximal utilization of the imageable area. A single large tissue section can be imaged (Fig. 2A), a number of smaller tissues can be sectioned on the slide after each other – ensuring they don't overlap – (Fig 2B) or a tissue microarray can be assembled and sectioned onto the slide (Fig 2C).



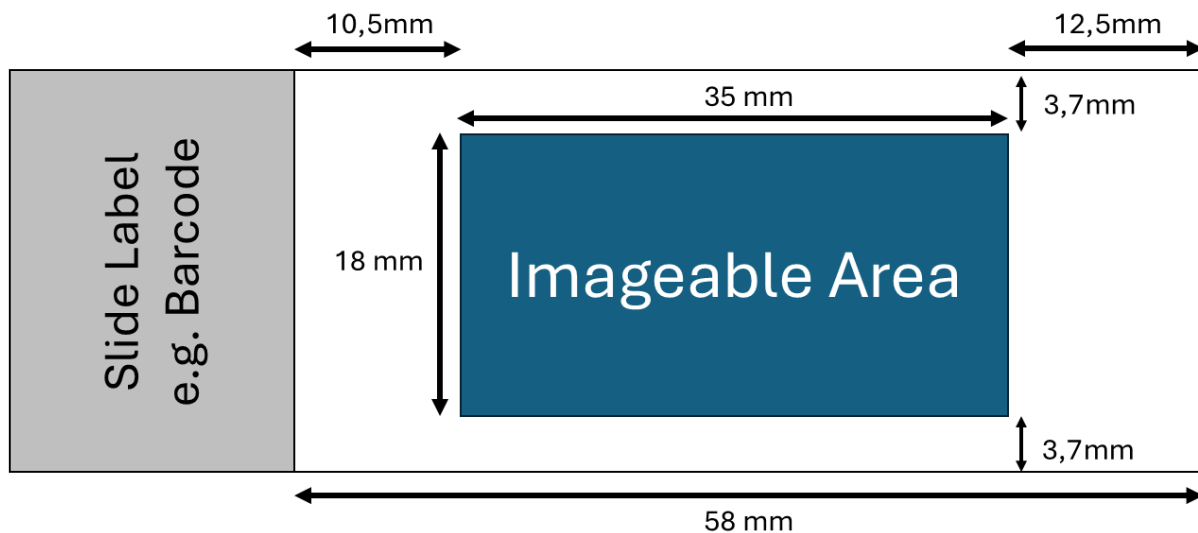


Figure 1 Tissue Placement Guide: Make sure that the tissue section is placed in the centre of the slide. Tissue under the adhesive portion of the flow cell can affect bonding of the flowcell and cause experiment failures. Ensure the tissue is placed on the positively charged side of the slide to ensure proper adhesion.

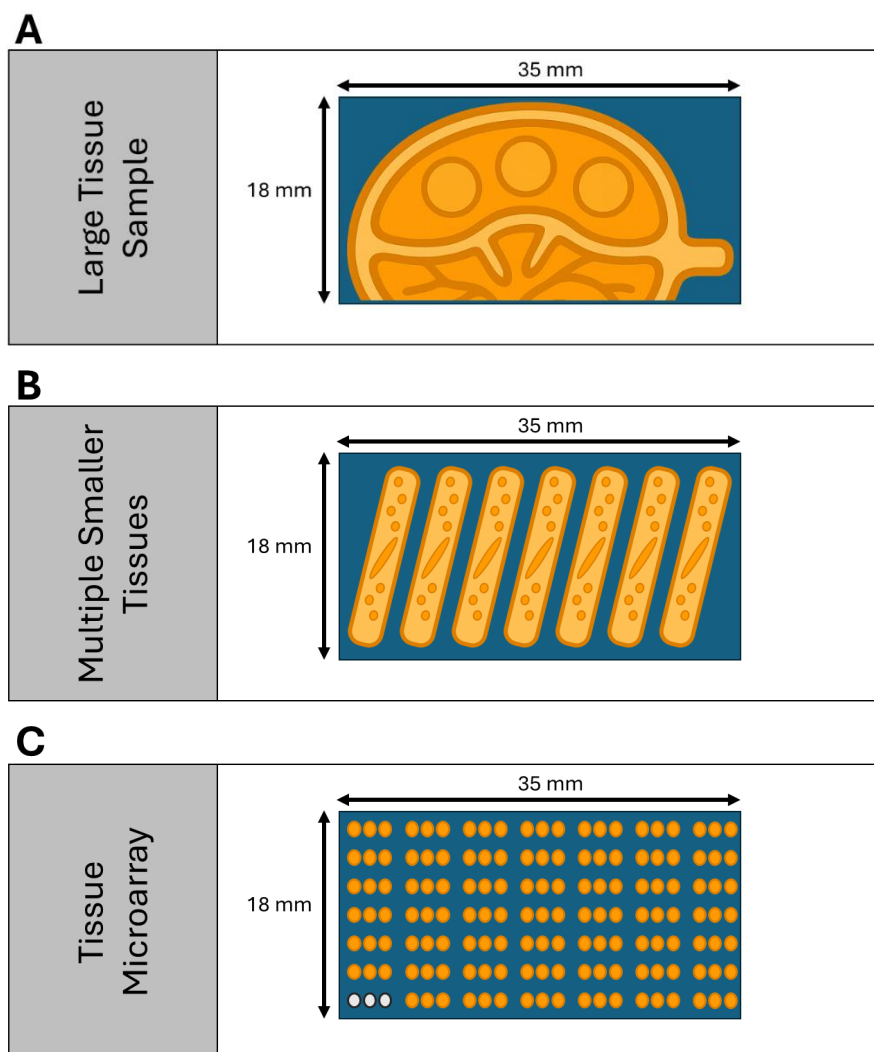


Figure 2 Tissue Arrangement: The entire imaging area can contain a single tissue specimen (A) or be filled with multiple smaller tissue specimens, which is particularly relevant if the original tissues are not suitable for the generation of a tissue microarray (B). If the original tissues are suitable for assembly in a tissue microarray, any format of core size, replicates and orientation can be placed within the 18x25mm area (C).

