CPAC sample processing recommendations

Sample types and requirements (setup/installation phase)

- Cells
 - Frozen cell pellets (dry ice), washed 1x with PBS, collected by scraping
 - Sample processing using the Preomics Kit (1-100 ug material),
 - Minimal input requirement: 300 000 cells
 - one shot samples: 10 000 cells
 - pellet of 1x 6-well

Native Tissue

- Transfer frozen (dry ice)
 - Sample processing: tissue homogenization (mixer, douncer), sonication in vial tweeter
 - Sample processing using the Preomics Kit (1-100 ug material),
 - input requirement: 2 mm biopsy punch (lengths ca. 0.5-1 cm); biopsy slice (10-20 μm thickness of a tissue block, 1cmx1-2 cm)
 - for statistical data evaluation three punches/slices (biological replicates) are required

Formalin-fixed Tissue (FFPE)

- o Transfer RT
 - Sample processing: de-waxing using a series of Xylene/Heptane and Ethanol; tissue homogenization (mixer, douncer), sonication in vial tweeter
 - Sample processing using the Preomics Kit (1-100 ug material), protocol for FFPE tissue
 - input requirement: 1 mm biopsy punch (lengths ca. 0.5-1 cm) or biopsy slice (10-20 μm thickness of a tissue block, 1cm x 1.5 cm);
 ~weight input material 0.5-1 mg

 for statistical data evaluation three punches/slices (biological replicates) are required

Quality Control Standards

- Bovine Fetuin/A1AG as spike-in reference for sample processing at the protein level
- iRT spike tides as reference for MS performance and Spectronaut evaluation
- QuiC software as tool to monitor MS performance(Biognosys, Spectronaut pipeline)