Fast and Simple Protocols for Mass Spectrometry-Based Proteomics of Small Fresh Frozen Uterine Tissue Sections

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Fast and simple protocols for mass spectrometry-based proteomics of small fresh frozen uterine tissue sections

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Figure S-1. Influence of the instrumental parameters on number of identified proteins and peptides. Results are displayed for optimization of instrumental parameters for both TOF-MS and TOF-MS/MS accumulation time. Data were recorded in intensity dependent acquisition (IDA) mode with the following parameters: TOF-MS ($m/z$: 400-1250; threshold 100 cps; top ions: 30 with charge from +2 to +4; exclusion time: 30 s); TOF-MS/MS ($m/z$: 200-1800). Other parameters regarding instrumental settings were as described in Experimental section.
Figure S-2. Venn diagrams showing qualitative reproducibility of the proteins between 3 biological replicates for 10, 16 and 20 µm tissue (a-c); shared proteins for 10 µm and 16 µm thick tissue identified from 3 replicates for Protocol 1 and 2 (d).
Supporting Information

Protocol 1

Protocol 2

Protocol 3

Protocol 4

Protocol 5

a)
b)
Figure S-3. Quantitative reproducibility of the proteins between 3 biological replicates of FF human uterus tissue. Results are shown as correlation of the NSAF values of the identified proteins for (a) 10, (b) 16 and (c) 20 µm thick FF human uterus tissue.
Figure S-4. Number of identified proteins (a) and peptides (b) using different extraction and digestion conditions. Proteins and peptides were identified at 1% FDR in 10, 16 and 20 µm thick tissue after protein extraction and digestion using Protocols 1 to 5. Results are shown as mean±SD.
Figure S-5. Results from the study evaluated for the sequence coverage of the proteins extracted from 3 biological replicates distributed according to their $M_w$: a) 10 µm, b) 16 µm and c) 20 µm thick FF human uterus tissue. Results are shown as mean±SD.
Figure S-6. NSAF values of the proteins extracted from 3 biological replicates distributed according to their $M_w$: a) 10 µm, b) 16 µm and c) 20 µm thick FF human uterus tissue. Results are shown as mean±SD.