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

Dapic, I.; Uwugiaren, N.; Jansen, P.J.; Corthals, G.L.

DOI
10.1021/acs.analchem.7b01937

Publication date
2017

Document Version
Other version

Published in
Analytical Chemistry

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

Irena Dapic[1], Naomi Uwugiaren[1], Petra J. Jansen[1] and Garry L. Corthals[1]

[1]University of Amsterdam, Van 't Hoff Institute for Molecular Sciences, Analytical-Chemistry Group, Science Park 904, 1098 XH Amsterdam, The Netherlands

Corresponding author: Prof. Garry L. Corthals, University of Amsterdam, Van 't Hoff Institute for Molecular Sciences, Analytical-Chemistry Group, Science Park 904, 1098 XH Amsterdam, The Netherlands; E-mail: G.L.Corthals@uva.nl, Phone: +31202525406

Supporting Information

Table of content

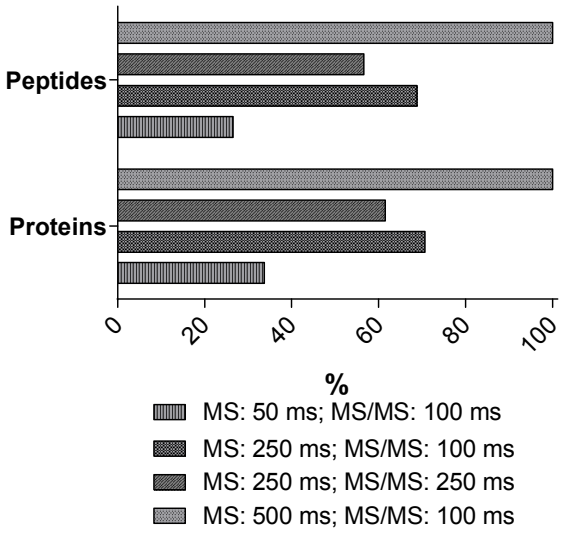


Figure S-1. Influence of the instrumental parameters on number of identified proteins and peptides. Results are displayed for optimization of instrumental parameters for both TOF-MS and TOF-MS/MS accumulation time. Data were recorded in intensity dependent acquisition (IDA) mode with the following parameters: 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.

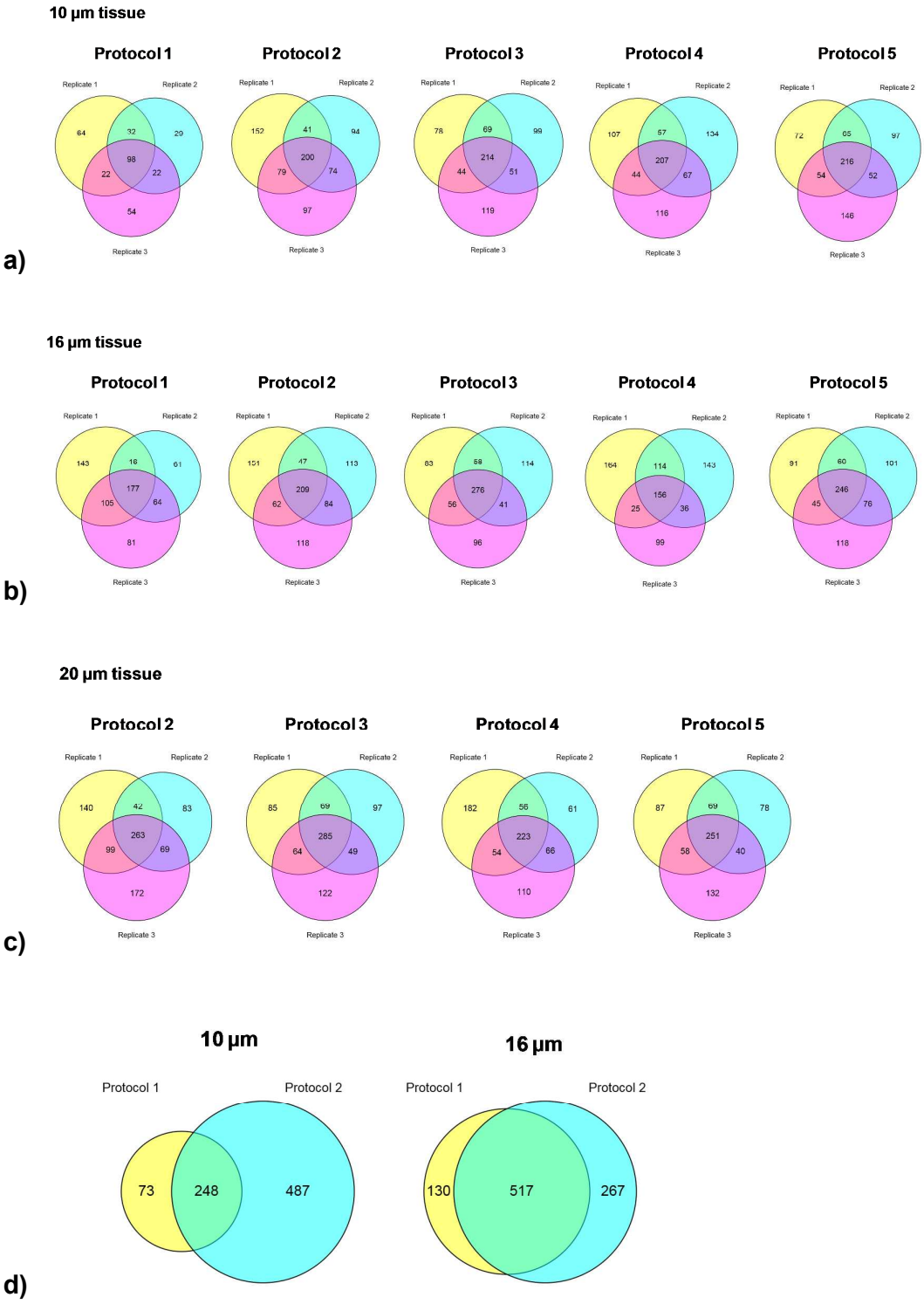
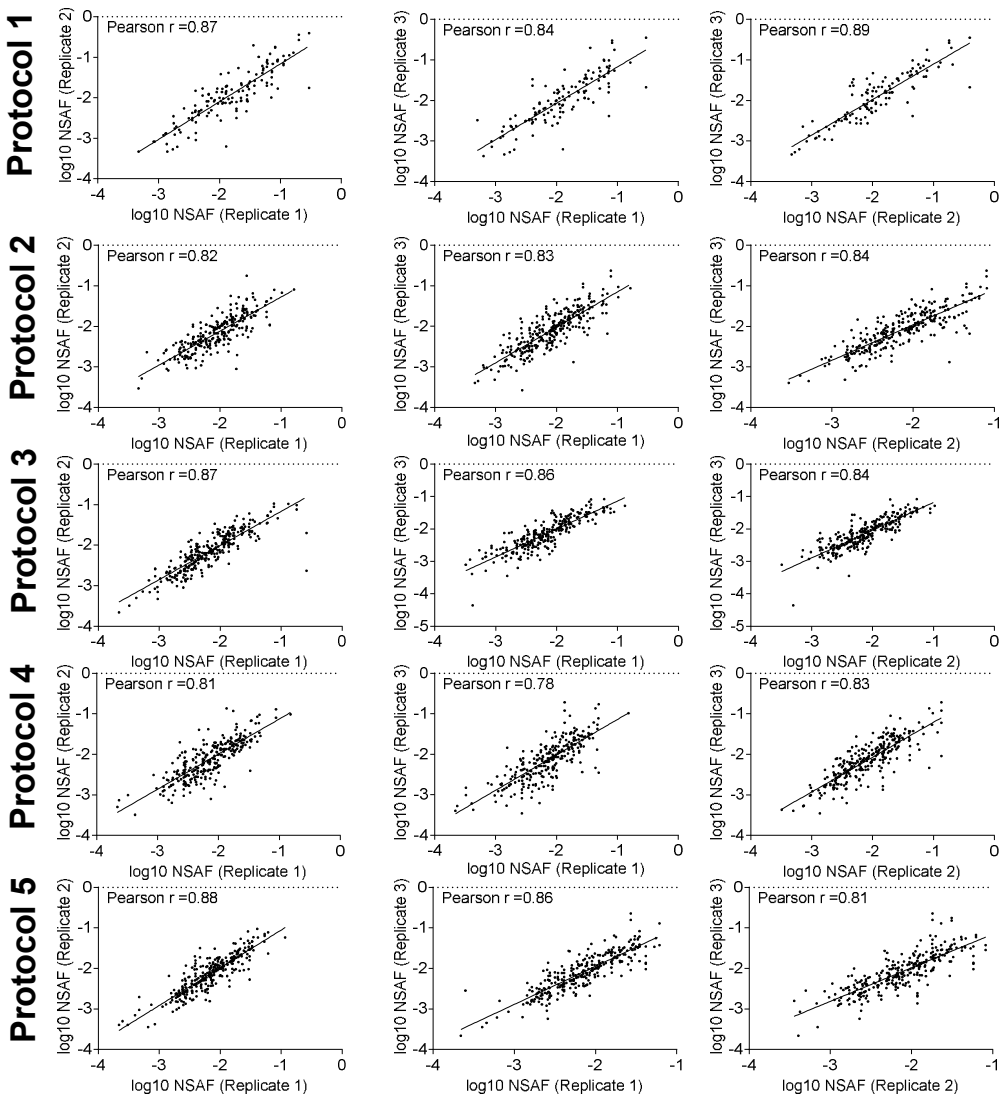
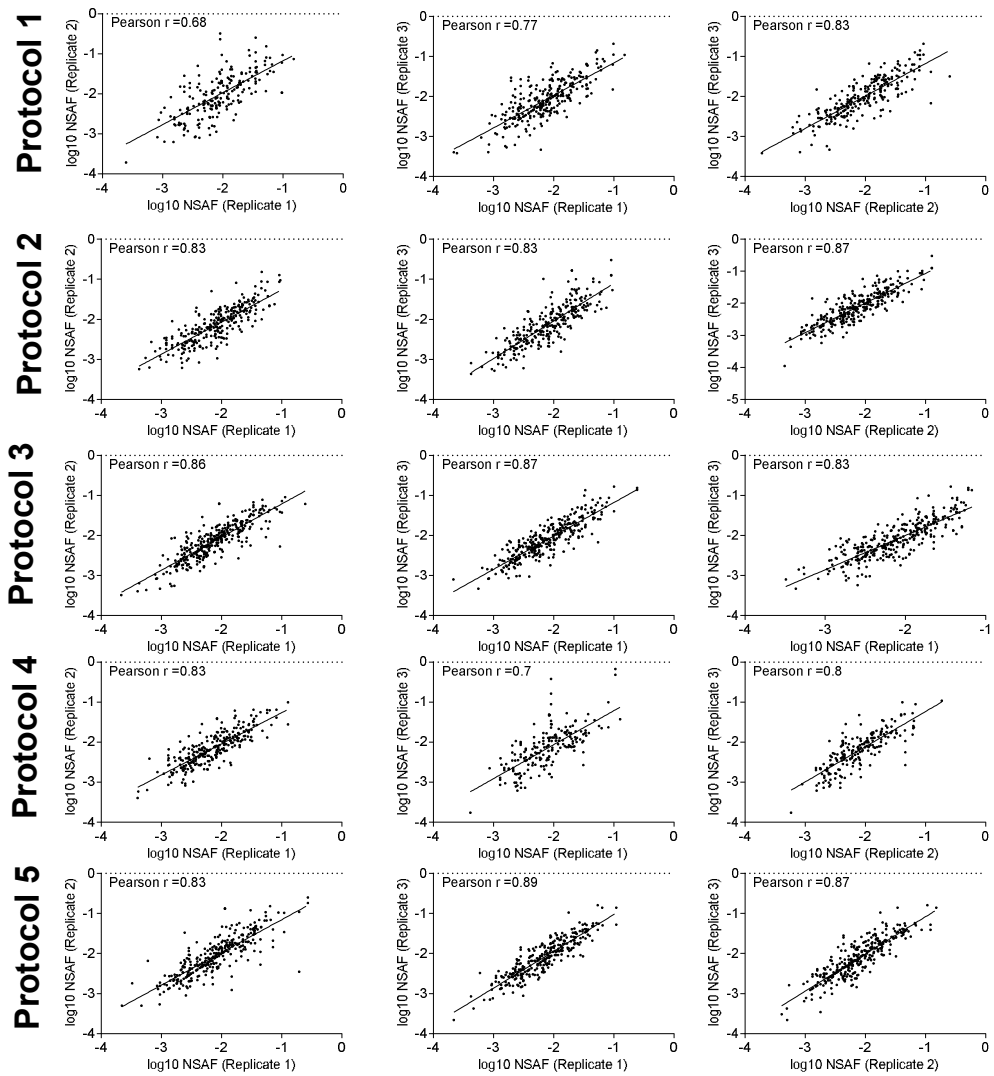


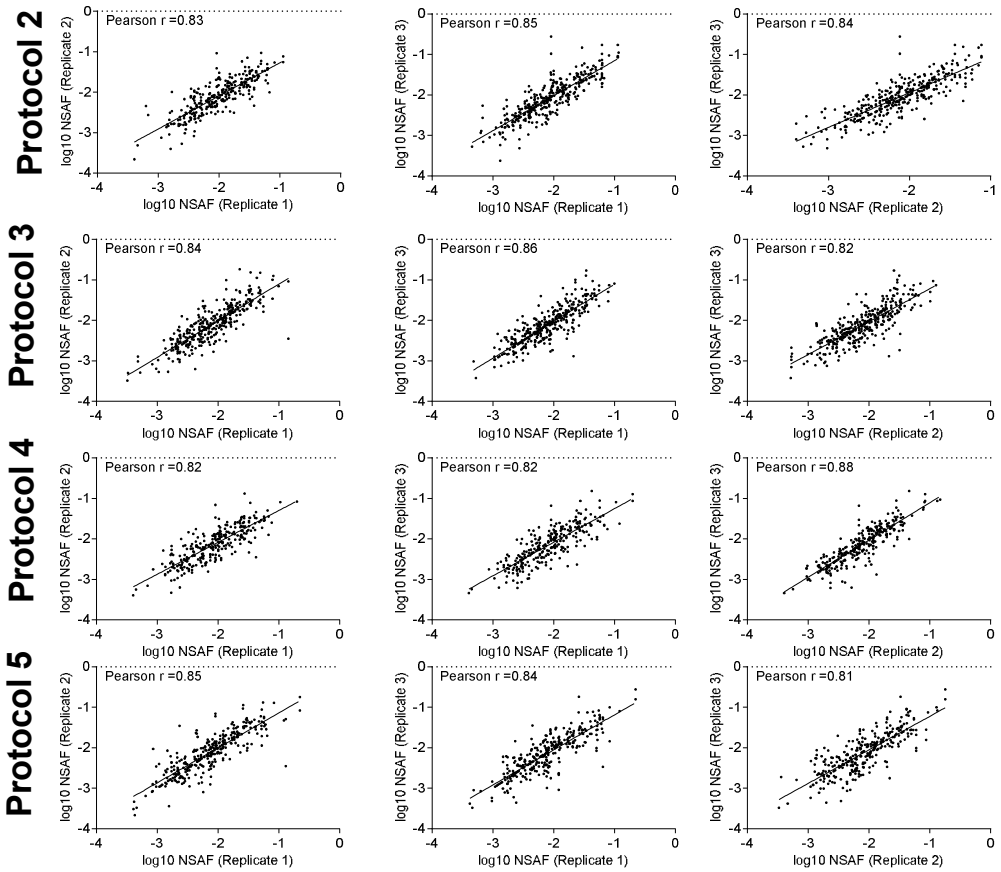
Figure S-2. Venn diagrams showing qualitative reproducibility of the proteins between 3 biological replicates for 10, 16 and 20 µm tissue (a-c); shared proteins for 10 µm and 16 µm thick tissue identified from 3 replicates for Protocol 1 and 2 (d).


Protocol 1
Protocol 2
Protocol 3
Protocol 4
Protocol 5
Pearson r =0.87
Pearson r =0.84
Pearson r =0.89
Pearson r =0.82
Pearson r =0.83
Pearson r =0.84
Pearson r =0.87
Pearson r =0.86
Pearson r =0.84
Pearson r =0.81
Pearson r =0.78
Pearson r =0.83
Pearson r =0.88
Pearson r =0.86
Pearson r =0.81
log10 NSAF (Replicate 1)
log10 NSAF (Replicate 2)
log10 NSAF (Replicate 3)

a)

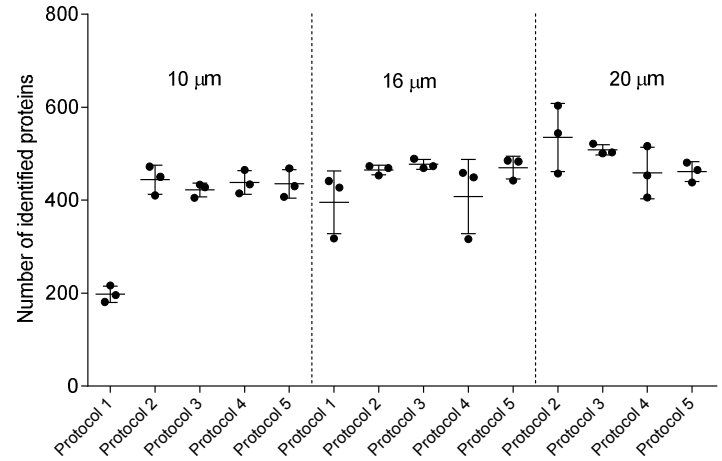
b)


Protocol 1
Protocol 2
Protocol 3
Protocol 4
Protocol 5
Pearson r =0.68
Pearson r =0.77
Pearson r =0.83
Pearson r =0.83
Pearson r =0.83
Pearson r =0.87
Pearson r =0.86
Pearson r =0.87
Pearson r =0.83
Pearson r =0.83
Pearson r =0.7
Pearson r =0.8
Pearson r =0.83
Pearson r =0.89
Pearson r =0.87
log10 NSAF (Replicate 1)
log10 NSAF (Replicate 2)
log10 NSAF (Replicate 3)

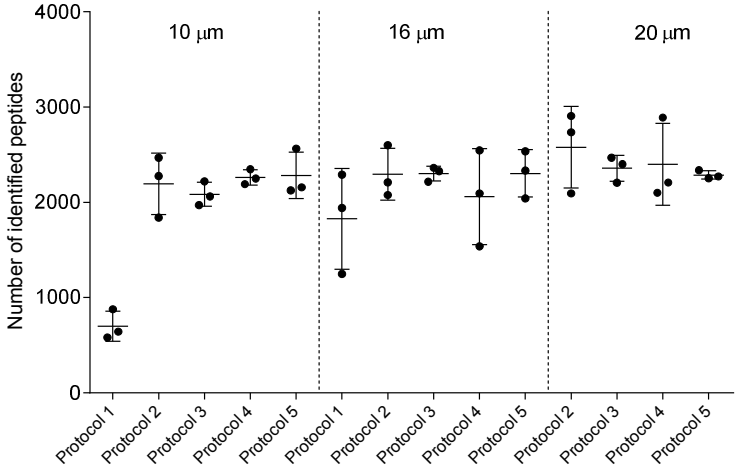


c)

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.

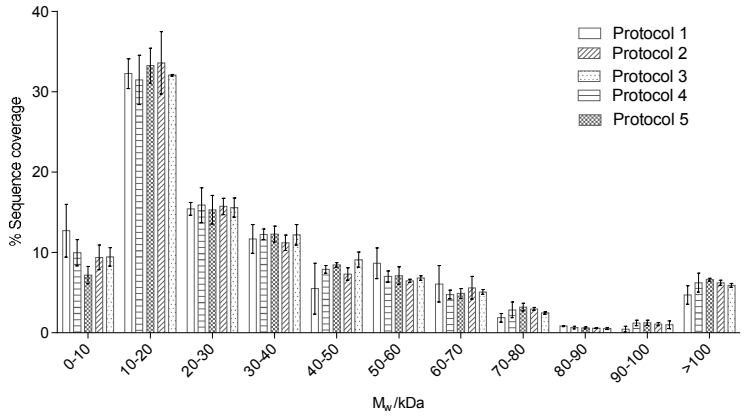


a)

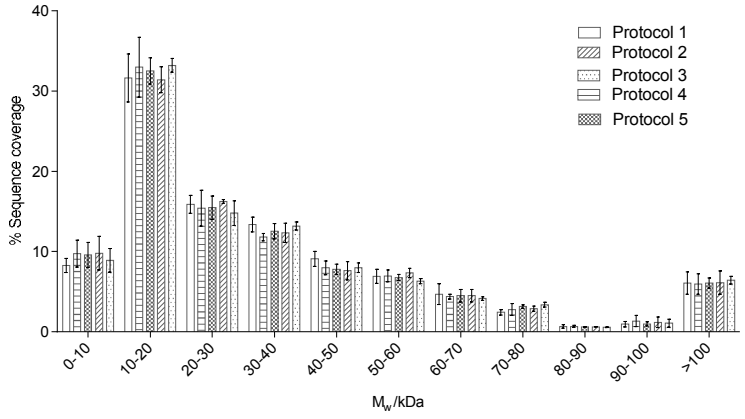


b)

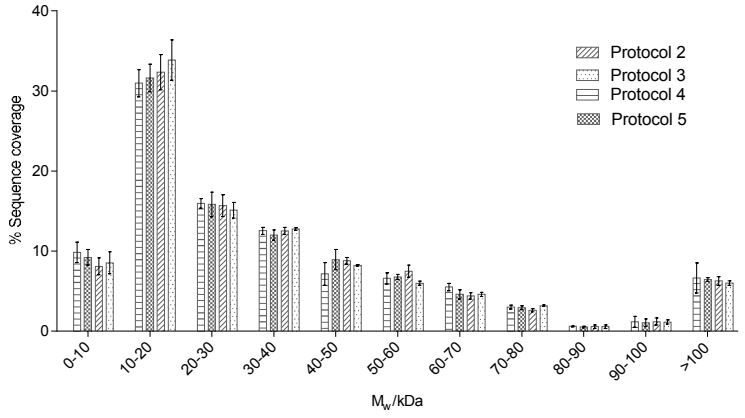
Figure S-4. Number of identified proteins (a) and peptides (b) using different extraction and digestion conditions. Proteins and peptides were identified at 1% FDR in 10, 16 and 20 μm thick tissue after protein extraction and digestion using Protocols 1 to 5. Results are shown as mean±SD.



a)

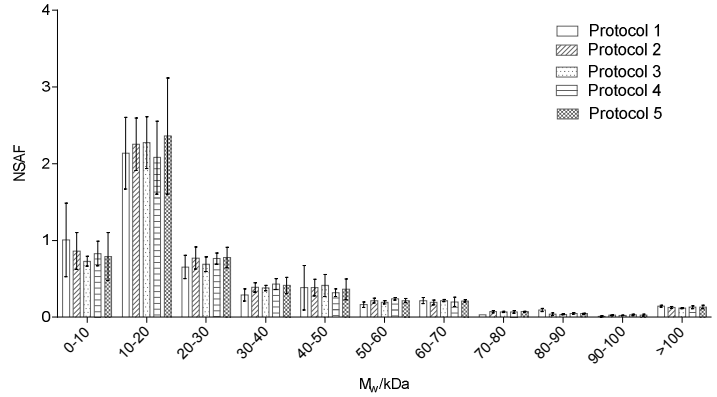


b)

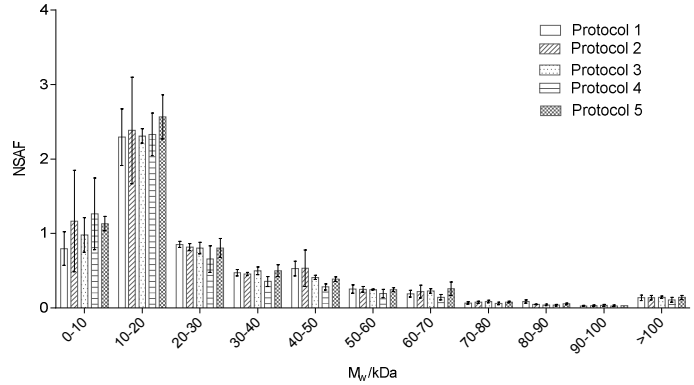


c)

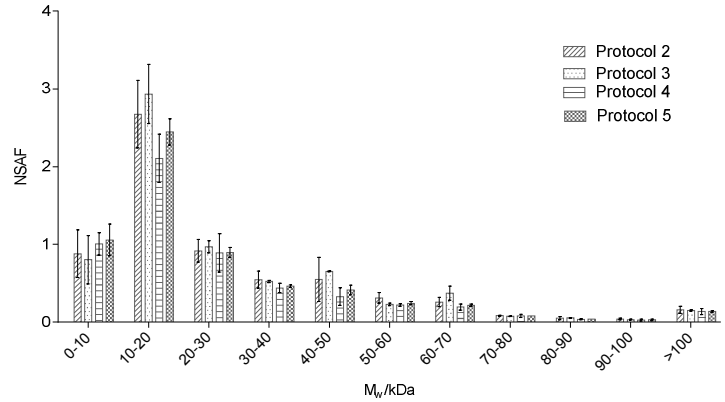
Figure S-5. Results from the study evaluated for the sequence coverage of the proteins extracted from 3 biological replicates distributed according to their M_w: a) 10 µm, b) 16 µm and c) 20 µm thick FF human uterus tissue. Results are shown as mean±SD.



a)



b)



c)

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.