

MOLECULAR SEROLOGY PROCEDURES MANUAL

Body Fluid Identification by Proteomic Mass Spectrometry – Extraction		
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Body Fluid Identification by Proteomic Mass Spectrometry – Extraction

1 Purpose

- 1.1 Protein extraction from samples to identify specific body fluids on evidence samples using liquid chromatography and mass spectrometry.

2 Protein Extraction Procedure

- 2.1 Procure a frozen tube rack or bucket filled with ice.
- 2.2 Obtain labeled cuttings in 1.5 ml microcentrifuge tubes and place in an ice bucket or frozen tube rack. Compare your sample labels and tube tops to the input sample list in LIMS and confirm that you have the correct samples.
 - Note: Sample size for the extraction should be approximately ¼ of a cotton swab or a 3x3 mm cutting of a stain.
- 2.3 Obtain a 1.5 ml microcentrifuge tubes for your extraction negative. Label it with the associated input LIMS label and tube top label.
 - Note: You will need **one** Extraction Negative for every **nineteen samples in the batch**.
- 2.4 Obtain two 1.5 ml microcentrifuge tubes for each sample for subsequent processing.
- 2.5 Label with the associated output LIMS label and tube top label.
- 2.6 Write a Q on one tube to designate it the Quant Aliquot (1X) tube and an EX on the other to designate it the Extraction tube.
- 2.7 **Label WITNESS**: Have a witness verify that correct tubes are present in the set and the labels on input tubes match labels on output tubes.
- 2.8 Turn refrigerated centrifuge on. Ensure it is set to 4°C.
- 2.9 Retrieve the 1% SDC extraction solution from 4°C refrigerator. Record its identification number in LIMS.
- 2.10 Add 200 µl of 1% SDC to each of the samples and ENEG.
- 2.11 Incubate at room temperature (~25°C) for 30 minutes at 1,000 RPM in a thermomixer. Record instrument and temperature in LIMS.

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- 2.12 Spin tubes in a refrigerated centrifuge (set to 4°C) at 18,000 g for 30 minutes. Record instrument and temperature in LIMS.
- 2.13 Transfer supernatants to corresponding Extraction tubes, discard the sample tube.
- 2.14 Pipette 41 µl from Extraction tubes into Quant Aliquot tubes, for protein quantification.
- 2.15 Store Extraction and Quant Aliquot tubes at -20°C.

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