

**Current Standard Operating Protocols (SOP), NCBS-CCAMP MS-Facility Metabolomics
Sample Preparation: Extraction of metabolites from Sera/plasma/Tissue**

Purpose: To provide general guidelines for extracting metabolites from Sera/Plasma/Tissue.

Reagents: All solvents and reagents used are of LC-MS quality.

Sample Preparation:

- Prepare the individual stock solutions (STDs) and ISTDs in a suitable solvent.
- Spike the standards and ISTDs (~100 pg) in 200 µl sera (double charcoal stripped human sera).
- Precipitate the proteins from sera by adding 1 mL of acetone (0.1% FA) or methanol and vortex it for ~3mins.
- Centrifuge (14000 rpm, 5 min) and transfer the supernatant to the fresh eppendorf.
- Dry this in speed vacuum and reconstitute it in 50 µl of starting gradient and transfer this into the HPLC vial for the injection.