Guidelines for Sample Preparation

- It is very important to prevent or minimize contamination of your sample. Problem contaminants include keratins, plasticizers, polymers, detergents, and non-volatile buffers.
- Avoid contamination with keratins.
- Protein storage recommendations – Proteins in coomassie-stained gels are relatively stable at 4C or -20C. Silver-stained protein bands must be analyzed as soon as possible after it is isolated and purified or should be kept frozen (i.e. -20C or -80C).
- Protein solutions containing very low levels of protein (i.e. femtomole or picomole) should be concentrated using Speedvac or ZipTips.
- Coomassie, Colloidal Blue, and similar blue stains are preferred and will generally result in the best peptide recoveries and protein ID results.
- For silver staining, we recommend that you use mass spectrometry-compatible silver stain (such as SilverQuest from Invitrogen). Don't over-stain the gel; stain for the least amount of time possible until you can just see your bands of interest.
- Cut the band out as close as possible; be very selective and cut out only the center of the band; don't include any excess gel material.
- Purity of the protein sample is critical for a successful analysis of the intact molecular weight and identification of a protein. It is critical that the sample does not contain any detergents or polymers (even at trace levels). Avoid using Triton, Tween, SDS, or PEG during any part of the protein preparation.
- Use HPLC/FPLC to purify and collect samples whenever possible.
- Desalt samples prior to sample submission (e.g. HPLC, C18 sep-pack, C18 Ziptip, dialyses). Salts will have a significant impact on ionization, both for ESI or MALDI ionization.
- For more than 10 samples new requisition form needs to be submitted.