

# LC-MS/MS SUBMISSION FORM

**Use this form for protein ID's from gel bands when the total target protein amount that can be isolated is <10 pmoles.**

## Wistar Proteomics Facility

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**Submit forms and samples to: Kaye Speicher or Thomas Beer (215) 898-3830/3181**

<b>Date Submitted</b>	<b>P.O. # or Wistar Grant #</b> <small>(credit card payment see below)</small>	<b>P.O. Amount</b>
<b>Submitted by</b>		<b>P.I.</b>
<b>Address</b>		<b>Phone</b>
		<b>Fax</b>
		<b>Email</b>
<b>Credit Card Type (circle one)</b> AMX      MC      Visa		<b>Exp. Date</b>
<b>Credit Card Number</b>		
<b>Name on Credit Card</b>		<b>Daily Limit (if any)</b>

**This method involves the following steps:**

1. **In situ alkylation of cysteines followed by trypsin in-gel microdigestion**
2. **Injection of digest solution onto a nanospray LC - ion trap mass spectrometer**
3. **Search of MS/MS data vs NCBI nr database**
4. **(Optional) More extensive analysis of data for specialized applications or if matches are not found**

**PREPARATION OF SAMPLES IN SDS ACRYLAMIDE GELS**

1. Review and follow "Sequence-Quality SDS-Gel Guidelines" for sample preparation.
2. After electrophoresis immediately stain with either a. or b.
  - a. Coomassie Blue R-250 30-60 minutes. Destain 1-4 hours maximum (background does not need to be completely clear). Rinse in high quality water (such as MilliQ) for 1 hour.
  - b. Invitrogen Colloidal Coomassie Blue G-250 (follow manufacture's guidelines for staining and destaining).
3. Seal in ziploc bag and store at 4°C (add only a few drops of water, gel should not be floating).
4. The entire gel must be sent to us; follow "Gel Shipping Instructions".
5. Include xerox copy or photo of gel with desired band(s) clearly marked.

Sample Name/Code \_\_\_\_\_ Species \_\_\_\_\_

# of Bands \_\_\_\_\_ Pixelation? Yes \_\_\_\_\_ No \_\_\_\_\_ Length of gel run in cm? \_\_\_\_\_

Recombinant? Yes \_\_\_\_\_ No \_\_\_\_\_ If yes, expression system used \_\_\_\_\_

Gel Type (e.g. Laemmli) \_\_\_\_\_ Gel Thickness \_\_\_\_\_ Gel % \_\_\_\_\_

pI (if known) \_\_\_\_\_ List known or suspected post-translational modifications: \_\_\_\_\_

Were reducing conditions used during SDS PAGE (βSH or DTT)? \_\_\_\_\_

Was sample alkylated? \_\_\_\_\_ yes \_\_\_\_\_ no Stain: Coomassie R250 \_\_\_\_\_ Colloidal Blue \_\_\_\_\_ Other \_\_\_\_\_

Is sample radioactive? \_\_\_\_\_ yes \_\_\_\_\_ no Isotope? \_\_\_\_\_ Total cpm in sample? \_\_\_\_\_

Has a copy or photo with band(s) indicated been enclosed? \_\_\_\_\_