October 7, 2008

Dear colleagues,

Please find enclosed the ABRF-2009 ESRG samples that you requested from the ABRF Edman Sequence Research Group. This is the 21st study in an annual series designed to aid laboratories in evaluating their abilities to obtain and interpret amino acid sequence data. The purpose of this year's study is to investigate both traditional and alternative methods for obtaining the N-terminal sequence of a protein. Participants may use any available Edman, mass spectrometric or biochemical method or technology they choose, or a combination of techniques. The goal is to obtain as much amino acid sequence from the N-terminus of the protein as possible by whichever technique is chosen. In addition to obtaining the N-terminal sequence of the proteins, we are also asking participants to try to identify the proteins by conducting a database search.

For this year's study, two vials (Test Sample 1 and 2) each containing a unique protein are provided. Each vial contains approximately 1 nanomole of protein for analysis. The ESRG committee has determined that both proteins are soluble in a variety of buffers. Potential buffers that may be used include:

a. 0.1% trifluoroacetic acid
b. 70% formic acid
c. 1% acetic acid
d. Guanidine-HCl

It is recommended that both test samples be allowed to solubilize for > 1 hour before beginning an analysis.

The object of this year's study is to ascertain the state of the art for determining an N-terminal sequence using a mass spectrometry technique as compared to traditional Edman degradation chemistry. In order to do this comparison, the ESRG is asking that you attempt to get as much N-terminal sequence as possible from each test sample by whichever technique you've chosen. Those performing Edman sequencing should expect to be able to sequence 30+ cycles for each protein. The challenge is to find out if there are any mass spectrometry techniques that can supply equivalent N-terminal sequencing data. If you have the time, enough protein has been provided in each tube to be able to perform multiple analyses.

The ESRG is asking participants to fill out an Excel data file with the results from your analysis. The Excel file can be found on the ABRF web site. Go to the Research Groups tab and select the Edman Sequencing Research Group bullet point. Under "Studies" select the "ESRG 2009 Data Sheet". If you cannot open the file or prefer a paper copy, contact Steve Smith (jssmith@utmb.edu) and he will fax you a paper copy. For those performing Edman degradation chemistry, we ask that you include yield data with the Excel file. All other techniques need only submit sequence data. In addition, we also ask that you submit two other items. First is a request for the protocol (including instrumentation) you used. A place for a short protocol can be found on the Excel form. If however, more space is required, the protocol can be submitted as a separate word or text file. Second we request copies

of the mass spectra for each analysis. These should be submitted as powerpoint, excel or pdf files. If you have collected N-terminal sequence data by more than one technique, please submit these results as separate data sets.

The files with the results should be e-mailed to Dr. Robert English (rdenglis@utmb.edu) as attachments by December 1st, 2008. If you are returning paper copies and/or pc formatted cd's, please mail to:

Dr. Robert English
Biomolecular Resource Facility
Mass Spec Core Lab, Room 2.234 BSB
Mail Rt. 0635
University of Texas Medical Branch
Galveston, Texas 77555-0635 USA

In order to ensure anonymity, Robert English will remove all identifying marks prior to forwarding the data to the sequence committee for analysis. An e-mail will be sent to your facility with a three-digit code to allow you to evaluate your results as compared to others. The sequencing results will be presented at ABRF 2009, February 7 - 10, 2009 in Memphis, Tennessee, and will also help guide future potential studies and tutorial sessions.

If your sample arrived damaged or if you have questions about the study, please contact Steve Smith at the above e-mail address. Equipment failures and "no data obtained" analyses are as important to us as data from "successful" runs. Please send us your results whatever happens. Thank you for your participation in this study!

The deadline for receiving data for inclusion in the study is December 1, 2008.

The Edman Sequencing Research Group:

Peter Hunziker (chair)
University of Zurich

Steve Smith
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Monsanto Co.

Nancy Denslow (EB liaison)
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