



Association of Biomolecular Resource Facilities

Business Office:

2019 Galisteo Street, Bldg. I, Santa Fe, NM 87505

Tel: 505-983-8102 ♦ Fax: 505-989-1073 ♦ Email: abrf@abrf.org

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Re: ABRF-PRG08 Proteomics Research Group (PRG) Qualitative Proteomics Study

Dear Colleagues,

Thank you for agreeing to participate in the "PRG2008 Qualitative Proteomics Study" conducted by the ABRF Proteomics Research Group (PRG). Accompanying this letter are two sample vials. If for some reason the samples did not arrive in good condition, please notify the PRG by sending an e-mail to abrfPRG2008@yahoo.com. Results returned by **December 1** will be included in the PRG presentation at the 2008 ABRF Meeting (Feb. 9 – 12, 2008, Salt Lake City, UT) and will be posted electronically.

PRG2008. The main goals of the 2008 PRG study are for participants to identify the proteins present in two similar samples and to determine any qualitative differences among the proteins in the samples. As a result of this study, it is anticipated that it will not only be possible to compare different strategies used for qualitative characterization of proteins, but also, information will be gained about methods used to maximize protein sequence coverage. The study was designed such that it could be addressed by a wide variety of proteomic methodologies. We intentionally selected samples that could be quickly and successfully analyzed using well-established techniques that can be performed by participants at a variety of skill levels. At the same time, we anticipate that the study would have some aspects that are challenging enough to benefit from the use of new or emerging technologies that are being investigated in more experienced laboratories. With participation from a wide range of scientists in laboratories who use varied analytical approaches and instrumentation, it will be possible to compare the capabilities of diverse methods for qualitative protein characterization. Results from participants will be submitted using an on-line survey, as in previous studies. In addition, we are now asking participants to provide a short summary of the approach(es) they used and the key results they obtained. The PRG expects to publish the tabulated results with links to participants' (anonymous) individual summaries so that information about effective protocols and novel strategies can be readily shared with the proteomics community.

Sample Information. The study samples are supplied in two vials (labeled "A" and "B") and consist of separate preparations of related human proteins that were expressed in *Escherichia coli* and affinity purified. The cloning vectors that were used produced an N-terminal His₆-tagged fusion protein that included a thrombin cleavage site in the His-tag. In both preparations, the sequence GSHM was retained with the protein after thrombin cleavage to remove the His-tag. Vials A and B contain 5 µg and 10 µg of protein, respectively. The samples were prepared from aqueous solutions that also contained small amounts of salts. To the best of our knowledge, there are no appreciable quantities of interfering substances that contain primary amines and/or free thiols. The samples have been successfully dissolved in 25 – 50 mM ammonium bicarbonate with or without 20% acetonitrile; as well as 0.1% formic acid; we anticipate that other solvents can be used.

Returning Results. When you submit your results, please provide: (1) the identification of major protein(s) in each sample; (2) any differences in protein composition found between samples A and B; and (3) the sequence coverage obtained for the major protein(s) in each sample, with a goal of attaining maximum coverage.

As noted above, submission of results will be via an on-line survey. The survey will be accessible at http://www.surveymonkey.com/s.aspx?sm=pErV5pSei4nsucjiY255bA_3d_3d. This survey address will also be posted on the PRG website. When submitting your results, you will be asked to supply a 5-digit identification number so that all entries remain anonymous. It is requested that you use UniProt accession numbers when reporting results. In addition to the on-line survey, please prepare a brief written summary of the experimental methods used and the corresponding results obtained from your analysis. If you used more than one independent experimental strategy to obtain your results, please enter each one separately in the on-line survey. It will be necessary to use a separate 5-digit identification number for each entry. This will allow us to associate the results obtained with each analytical approach. The written summary of your methods and results can be combined into one document. Please indicate any qualitative differences between samples A and B that were observed and provide supporting information. Use your 5-digit identification number as the file name for your written summary and save the file in pdf format; e-mail the file to anonymousPRG2008@yahoo.com. Please remember to keep your written report anonymous, as it will be posted along with the study results.

Please note that if your browser is set to accept cookies, you will be able to revisit the survey form and make changes in your entries until the submission deadline, as long as you are using the same computer. If you need to make changes in your entry but are using a different computer, it will be necessary for you to fill out the complete survey again using the same 5-digit identification code you used previously. Before compilation of the results, we will retain only the most recent submission and remove prior, duplicate entries. Remember that your results will be listed only by their 5-digit identification number. So, please select a unique number that you will recognize. We would like to emphasize that information about all analyses is vital to this study, whether successful or not. All results will be compiled in a completely anonymous manner (even to members of the PRG), so there is absolutely no need to feel shy about submitting negative results.

Any updates about the study will be posted on the ABRF PRG website: www.abrf.org/PRG

If you have any questions about filling out the online data analysis form, please e-mail the PRG at prg2008@googlegroups.com.

We thank you for your support of the ABRF and we look forward to your participation in this study.

The ABRF Proteomics Research Group

Nathan A. Yates (Chair) - Merck & Co. Inc.

Allis S. Chien - Stanford University

David B. Friedman - Vanderbilt University

Jeroen Krijgsveld - Utrecht University

William S. Lane - Harvard University

Kathryn S. Lilley - University of Cambridge

Michael MacCoss - University of Washington

Nicholas E. Sherman - University of Virginia

Susan T. Weintraub - Univ. of Texas Health Science Center at San Antonio

H. Ewa Witkowska - UCSF

Chris W. Turck (EB Liaison) - Max Planck Institute of Psychiatry

Vendors and Commercial Service Labs

PLEASE NOTE:

If you are a vendor or commercial service lab and wish to participate in this study, please read the following:

ABRF Research Group Studies are conducted for the benefit of our members and the field at large to help them evaluate their own technical level in comparison to their colleagues, to provide education in techniques and strategies to which they normally might not be exposed, and to give an overview of the current capabilities of the “average” lab in carrying out a challenging analysis.

The ABRF welcomes the participation of vendors and for-profit labs, provided that they abide by the ABRF guidelines for the use and distribution of data derived from these studies, as follows:

- An ABRF Research Study is not a competition and under no circumstances should it be referred to as such. Words and phrasing that imply a competition—such as ‘winner’, ‘best of’, etc., are strictly forbidden.
- Representations and publications should not be deceptive and should fairly emphasize any differences between any data comparisons. For example, instrument reliability cannot be fairly concluded by comparing 5-year old instruments in the field used in the study with a vendor's new instrument.
- Any comparisons to or use of ABRF data should prominently indicate: the number of samples the vendor received, the number of runs performed by the vendor, and whether the actual characteristics of the sample were known by the vendor at the time the vendor's analysis was performed.
- Uses of or comparisons to ABRF data should specifically emphasize that many factors will affect analytical results and that the data obtained in the company's R & D lab may exceed feasible expectations for an "average" resource or research facility under routine conditions.
- Publications and presentations should contain a disclaimer stating that ABRF prepared and provided the sample to all members and vendors, but did not participate in the vendor's study and does not endorse any specific manufacturer, instrument or strategy.
- Vendors are strongly encouraged to distribute potential publications to the ABRF Executive Board and Research Group Chairperson for comments regarding compliance with these guidelines.

***Recipient:* We recommend that this document be distributed to the appropriate marketing and senior personnel in the company to ensure compliance. A copy of this document (Vendor ABRF Study Participation Guidelines.pdf) can be found at www.abrf.org under the Forms and Documents menu.**