Biology Department

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managed by Brookhaven Science Associates
for the U.S. Department of Energy

SAMPLE CHARGE

Biology's Scanning Transmission Electron Microscope (STEM)

Per Sample* – non-proprietary……..$391.02**

*See attached for description
**Fee based on FY08 Standard Rates effective October 1, 2007 and published at
http://www.bnl.gov/budget/linkable_files/pdf/FY2007StandardRate.pdf

Dr. Joseph Wall
STEM Principal Investigator

Instructions on How to Initiate Work on Your Sample(s)

Under U.S. Department of Energy regulations, no work can be done on your sample(s) until an advance payment is received for the total number of samples needed.

A user account will be established for the value of the advance payment. As the work is done, appropriate charges for each sample will be applied to your user account. Once funds are depleted, work must stop until the account is increased by another advance payment.

To set up a user account, one of the following is required:

1. A check for the total amount made out to Brookhaven National Laboratory.
2. Wire transfer sent to: J.P. Morgan Chase Manhattan Bank, 270 Park Avenue, New York, NY 10017
   ABA 021 000 021
   Brookhaven National Laboratory Account No. 615-775942
   Swift Code – CHASUS33

The return of any unused balance must be requested in writing.

In all cases, the following information is required:
- Purpose of the project (short scope of work: (e.g., technical services of Biology STEM Facility to do non-propriety samples)
- Full name of principal investigator and authorized users
- The dollar amount
- The expiration date of the order

Mail orders to:
Ms. Georgia L. Irving, Budget Office, Bldg. 460, Brookhaven National Laboratory, P.O. Box 5000, Upton, NY 11973
Phone 631-344-7957; FAX 631-344-2149; irving@bnl.gov

User Rate for Biology's Scanning Transmission Electron Microscope (STEM)

Services Provided

As in the past, specimen solutions can be submitted frozen or on wet ice, either by package delivery or in person. Each solution should be accompanied by 1) a description of the objects of interest (images, gels, references, etc.), 2) approximate concentration, 3) buffer description, 4) estimate of purity and possible contaminants and 5) estimate of heavy atom labeling, if attempted. (Note: users are expected to do their own labeling.)

We will provide hydrophilic thin carbon substrates on holey carbon coated titanium grids, mounted in our holders. We will prepare dilution series and inject sample into a drop of buffer on the film, rinse, add TMV internal control, wash and fast-freeze. This will be done on the day of arrival (for specimens on wet ice), if so arranged, and grids will be stored under liquid nitrogen until freeze dried. Alternatively, grids may be washed with methylamine vanadate and air dried for negative staining.

We will transfer frozen grids to our UHV freeze-dry apparatus, freeze dry overnight and transfer under vacuum to STEM. Grids for negative staining will be processed in parallel with frozen-hydrated grids.

We will collect low-dose dark field STEM images (grid at -160C) suitable for mass measurement or heavy atom cluster mapping. Grids judged too crowded, too sparse or with bad background not suitable for reliable analysis will be imaged in at least 4 widely separated areas, providing a minimum of 12 typical images. Grids judged suitable for data collection will also be imaged in at least 4 widely separated areas, providing a minimum of 36 typical images. Magnification and dose will be chosen for optimum analysis.

We will provide digital STEM images on our FTP site, along with software for viewing & mass analysis. In addition, we will provide preliminary analysis of particle mass distribution, TMV quality & calibration and description of any problems encountered in imaging or analysis. We will provide customized models for mass analysis, if requested. We will provide Materials & Methods sections suitable for publication. We will provide advice on analysis and suggestions for further experiments either by e-mail or telephone.

Guarantee

We will repeat at no charge any grids which fail to give useful data due to: 1) loss of grid in handling, 2) poor coverage of thin carbon or 3) failure of freeze dry as indicated by TMV internal controls.

Description of Service:

Collecting data will be straightforward, since all grids (or samples) are numbered sequentially. Most specimens are arranged in advance by e-mail and submitted by FedEx so we have sender information.

- "Bad samples" are those with film that is so broken that we cannot collect any data.
- "Poor quality" specimens would be those contaminated by salt, detergent or other problems where data may have poor accuracy. This generally results from impurities in the supplied material and would be charged.
- "Difficult" specimens are those where extensive searching is required to find areas of interest, either because the sample is too dilute, too concentrated or falling apart. A "sample" is material submitted in solution (test tube), either frozen or on wet ice, along with a description, concentration in mg/ml and safety information. Any biochemical preparation at our end, other than dilution, would be charged.
- For each "sample," we make at least three "grids" in a dilution series.
- For each "grid", we will provide titanium support and holey film covered with thin carbon mounted in one of our holders. We will apply the user's specimen and freeze dry, as described on our web pages. Freeze dried grids will be transported under vacuum to the STEM and images recorded using the dark field annular detector, suitable for mass measurement. We will record at least 12 images (typically 36-64) and supply these digitally on our ftp site, along with software for viewing and analysis (including documentation & tutorials).

In some cases we may provide automated analysis. In all cases, we will be available by telephone or e-mail to answer questions about the use of the analysis software. Users would be expected to publish results, inform STEM staff and include an appropriate acknowledgement. If requested, we will provide a Materials & Methods section and review manuscripts.