Sample Submission Guideline for ProteinSimple NanoPro1000 Assay system

Biophysics Core Facility (BCF), Department of Academic Affairs and Instrument Service, Academia Sinica

I. Sample Submission

1. Apply an account on the BCF reservation system.
2. File sample submission form on-line.
3. After confirming the charges and experiment time through reservation system, lysate samples, primary and secondary antibodies (BCF provides Goat-Anti-Rabbit and Goat-Anti-Mouse secondary antibodies for detection.) may be submitted to BCF.
   Tel: 27855696 x4024; e-mail: szuhuan@gate.sinica.edu.tw
4. Please refer to http://bcf.assic.sinica.edu.tw/NP1000.htm for more.

II. Sample Preparation

5. Before harvest your cells, please contact us for cell lysate preparation procedures, lysis buffers and inhibitors.
6. Spin cell lysate at 12,000g in a 0.22uM Ultrafree-MC filter for 5 minutes to remove possible precipitation.
7. Determine total protein concentration of the cell lysate by BCA protein assay, usually 1~3mg/mL. The minimum concentration required for NanoPRO 1000 depends on the amount of proteins interested in the cell lysate and the affinity of its primary antibody. For pure proteins, try 5uL of 1ug/mL.
8. Please submit your primary antibody information to us so we may prepare your primary antibody solution according to the data sheet. Basically, the primary antibody is used 5-fold concentrated than is used for immune-precipitation experiments. About 8ul diluted antibody is needed for 1 assay. (For example, if the recommended dilution factor for immune-precipitation is 1:100, we will perform a 1:20 dilution for the experiments).
9. Although not always accurate, you may predict molecular weights and isoelectric points of your proteins on the page, http://scansite.mit.edu/calc_mw_pi.html, for picking suitable pH gradient. We recommend running the experiments at widest range of pH 3-10.
10. For common proteins involved in signal transductions, users may look up ProteinSimple antibody database for validation of the primary antibodies.
    http://www.proteinsimple.com/antibody/index.html?message=Please%20login%20to%20view%20and%20submit%20antibodies

III. Getting Experimental Result

11. Raw data can be downloaded through BCF reservation system.
12. BCF offers regular training courses for data analysis. Software, Compass, may be installed upon request. Please note that data analysis is the responsibility of users.
13. BCF may not compensate for your sample loss or data loss under any circumstances (hardware or software failure, operator error, or others). **All experimental results are for research only.**
14. Please acknowledge Biophysics Core Facility, Department of Academic Affairs and Instrument Service, Academia Sinica, if research supported and/or data generated by this laboratory results in publications.