

UHN Microarray Centre Guidelines for Preparing and Sending Protein Samples For Analysis on the Agilent 2100 Bioanalyzer

Introduction

The Agilent 2100 Bioanalyzer is a microfluidic electrophoresis device that is used to determine the size, concentration and purity of protein samples, including cell lysates, column fractions, antibodies and purified proteins. The Bioanalyzer is able to analyze proteins under reducing (in the presence of DTT) and non-reducing conditions. All assays are run under denaturing conditions.

The UHNMAC currently offers the Protein 230 kit (for proteins between 14-230 kDa in size), the Protein 80 kit (for proteins between 5-80 kDa) and the High Sensitivity Protein 250 kit (for detecting proteins in the low picogram range that are 10-250 kDa in size). All protein assays require 4 μL of sample per well.

This document outlines how to send protein samples for analysis and provides a work order form. Protein samples cannot be processed until the completed checklist and work order form has been received. The Bioanalyzer service is available for \$10 per sample.

Quantity of Protein Required

We request that you send *at least* 10 μL of each protein sample (this will allow for a repeat run, if necessary). For the Protein 80 and 230 kits, each sample concentration should be between 0.1 – 2 $\mu\text{g}/\mu\text{L}$ in a suitable buffer. For the High Sensitivity Protein 250 kit, each sample concentration should be at least 10 $\text{pg}/\mu\text{L}$. For a list of compatible buffers, please see: <http://www.chem.agilent.com/cag/prod/lb/Protein-buffersept-06.pdf>

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Sending Protein Samples

Protein samples should be sent frozen on dry ice. If you are hand delivering the samples, you should ensure that the samples remain frozen during transit and that the samples are handed directly to one of the members of the microarray lab. Please have the lab member that receives your samples sign the "RNA RECEIPT" and retain this receipt for your records.

If you are shipping samples, they should be sent by overnight courier to ensure they arrive frozen. They should be sent such that they will arrive at our laboratory between Tuesday and Thursday. Do not ship a package on Friday, as we do not work weekends. Samples should be sent to the following address:

UHN Microarray Centre
MaRS Centre, TMDT
101 College St. 9-601
Toronto, ON, Canada
M5G 1L7

Please include a completed copy of the checklist and the work order (found at the end of this document) in your shipment. Also include a spreadsheet containing designations and concentrations of each sample with your shipment. You should send an email to geneservice@microarrays.ca informing us that you have sent a shipment. Please provide the tracking number for the courier and your contact number/email address such that we can confirm that your sample has arrived safely.

PLEASE NOTE THAT ANY LEFTOVER SAMPLES WILL BE DISCARDED.

Turnaround Time

Typically, samples are analysed within two business days; however, larger numbers of samples may require more time. We will email your results when they are ready.

Data is provided in a PDF format. This includes a gel-like picture, electropherogram of each sample, and an estimation of sample concentration and purity.

Questions?

If you have any questions about the information contained in this document please contact geneservice@microarrays.ca

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Protein Sample for Bioanalyzer Checklist

Please completely fill out this checklist and submit it with your protein samples.

Orders that do not include this checklist will not be processed. Please check the appropriate boxes and initial the line at the bottom of the form.

NAME: _____ **PHONE NUMBER:** _____

EMAIL: _____

Protein Quantity

1. There is at least **10 μL** of each total protein sample.
2. Each sample is at the appropriate concentration.
(0.1-2 $\mu\text{g}/\mu\text{L}$ for 230/80 kits and >10 $\text{pg}/\mu\text{L}$ for 250 kit)
3. Mass of each sample was estimated by UV (A_{280})
(A list of sample concentrations should accompany samples)

Protein Purification

4. Type of protein sample (cell lysate, column fraction, purified protein): _____
5. Protein purification (if completed) was performed by _____
6. Samples are in (indicate buffer, including concentration and pH): _____

Assay Conditions

- 7 Protein assay required (circle one):
Protein 80 kit (5-80 kDa)
Protein 230 kit (14-230 kDa)
High sensitivity 250 kit (10-250 kDa)
8. Run samples under reducing (in presence of DTT) conditions? Yes/No/Doesn't matter
(circle one)

Initials confirming that all steps have been completed: _____

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Protein Sample Analysis using Agilent Bioanalyzer 2100 Work Order Form

Cost per sample analysis is \$10.00 (Cdn)

We require at least 10 μL of each protein sample at a concentration of 0.1-2.0 $\mu\text{g}/\mu\text{L}$ (for Protein 80 and Protein 230 kits) or $> 10 \text{ pg}/\mu\text{L}$ (for High Sensitivity 250 kit) in a compatible buffer (see: <http://www.chem.agilent.com/cag/prod/lb/Protein-buffersept-06.pdf> for a list of compatible buffers). Samples must be provided in a tube clearly labelled with a unique sample designation on top and the researchers name printed on the side, and must be kept frozen during transport.

Clearly fill out the following

P.I. Name: _____

Billing Address: _____

Preferred method of contact (circle one): phone or e-mail

Phone Number: _____

E-mail: _____

Number of unique samples: _____

Please provide list/spreadsheet of Sample Designations/concentrations

Researcher's Signature

Date

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PROTEIN SAMPLE RECEIPT

Date: _____

Samples delivered by: _____

Samples received by: _____

Number of samples received: _____

Were all samples frozen when received?

YES

NO

Recipients (UHNMAC) Signature

Please note that any leftover samples will be discarded following analysis on the Agilent Bioanalyzer.

University Health Network Microarray Centre

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