Facility for Light Scattering
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Operating Instructions for the **NanoBrook Omni**
Brookhaven Instruments
Location: Mason Lab Room B7
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This document is intended as a "quick-start-guide" for users already trained on the NanoBrook Omni. If you require training please contact us.

1. Turn the instrument On, using the switch in the back.

2. Open Software "BIC Particle Solutions"

3. Enter Operator ID = the user's NetID. This brings up the "Main Window."

4. In the "New Measurements" Dropdown Menu at the top of the screen, select "DLS" for DLS/sizing measurements or "PALS" for either zeta potential or IEP titration measurements. Hit the button "New." This brings up a second window titled either "DLS – Sample ID," or "PALS – Sample ID," which will be referred to as the "Measurement Window."

5. Review or change the desired measurement protocol by selecting "SOP" from the row of buttons along the top, which brings up the "SOP Window." Once the SOP details are complete, click OK to return to the Measurement Window.

6. Insert sample into sample chamber, using the electrode as required for PALS measurements. [This step can be done sooner, especially if any sample equilibration is required].

7. Select "Start" in the Measurement Window to begin, and assess both raw and processed data in the Main Window and the Measurement Window.

8. View and transfer data upon completion of the measurement.

Details for items 4, 5, 6, and 7 vary with measurement type (DLS or PALS). The **SOP Window** contains prompts for the user to input the desired measurement protocol and instructions. The **Measurement Window** contains the raw data as it is being collected. The **Main Window** contains both summaries and detailed information on the raw and processed data. Both the Measurement Window and SOP contain features specific to DLS and PALS measurements, and each are outlined below.

DYNAMIC LIGHT SCATTERING

Features of the DLS SOP:
From the bottom of the SOP Window, users have the option to "Load" a previous SOP. For DLS measurements, the option list on the left hand side is as follows:

- Identification
- Instrument Parameters
- Measurement
  - Parameters
  - Automation
    - Time Dependent
    - Temperature Dependent
- Titration
  - Setup
  - pH Titration Measurement
  - Additive Titration Measurement
- Sample Parameters
  - Liquid
  - Particle
- Data Analysis
  - Normalization
  - Size Distribution
  - MW Analysis

Of the items above, the ***options which must always be specified*** are:

1) **Identification**, in which the user sets the **Sample ID** and inputs any notes to accompany the saved data.
2) **Measurement → Parameters**, in which the user sets the **Duration** of each run. A typical value is 30 seconds.
3) **Measurement → Automation → Time Dependent**, in which the users sets the number of **Total Measurements**, as well as the **Time Interval Between Measurements**, which is typically 0 unless a time course is required. The interval is the number of seconds between the end of one measurement and the beginning of the next.
4) **Sample Parameters → Liquid**, in which the dispersing fluid is specified. The default solvent is **Water**, and the dropdown menu contains several common solvents. If the solvent information is missing, the user must select **Unspecified** in the dropdown menu and input the solvent Viscosity and Refractive Index in order to obtain meaningful data.

Other options include:

1) **Instrument Parameters** allows the user to select the standard 90^0 measurement, or switch to forward scattering (15^0) or backscattering (173^0).

Backscattering is recommended to measure small particles, especially when trace amounts of larger particles might be present.

2) **Measurement → Automation → Temperature Dependent** allows the user to specify Starting and Final Temperatures, as well as the Temperature Increment. Note that the Total Measurements requested in Item 3 above will be made for each temperature in the automation series.
3) **Data Analysis → Normalization** defaults to an Automatic calculation, but can be switched to Last Channels if even the largest particles or aggregates are to measured. Last Channels is also the recommended option for samples which exhibit two decays in the Raw Data Correlation Function.
4) **Data Analysis → Size Distribution** defaults to NNLS, but can also be switched to CONTIN. The choice of method here is reflected in the Multimodal Distribution displayed in the Measurement Window. These options can be toggled even after the data has been collected.

Note: Titration instructions are contained in a separate section below.

Upon completion of specifying your SOP, you may either "Save" to overwrite or "Save As," and then click "OK," which closes the SOP window and returns to the Measurement Window. ***It is recommended that you include your name or NetID in the name of your SOP.***

Preparation of DLS sample

Place between 2-4 mL volume of sample into a square plastic cuvette, and insert into the sample chamber. The precise sample volume is unimportant. Plastic covers are available for the plastic cuvettes. Close the top cover of the chamber when the sample is in place. **IMPORTANT NOTE**: For non-aqueous samples, use the square glass cuvettes. ***Glass cuvettes are not disposable***; be careful using them and clean them for the next user.

Begin measurement in the DLS Measurement Window:

Once the sample is in the sample chamber, select "Start" in the upper left hand corner of the Measurement Window. The instrument will equilibrate the sample chamber temperature, adjust the incident laser intensity, and begin measurements.

Data Display in Measurement Window:

Above the plotting window are options to display either Multimodal Distribution, Lognormal Distribution, or Raw Data. The Raw Data option displays both the Correlation Function C(t) and the Count Rate in kcps as a function of time. The Multimodal Distribution shows the size distribution calculated using the fitting method chosen in Data Analysis → Size Distribution. The Lognormal Distribution shows the size distribution as calculated from a Cumulant Analysis fit of C(t).

PHASE ANALYSIS LIGHT SCATTERING

Features of the PALS SOP:
From the bottom of the SOP Window, users have the option to "Load" a previous SOP. For PALS measurements, the list on the left hand side is as follows:

- Identification:
- Instrument Parameters:
- Measurement:
  - Parameters
  - Advanced Settings
  - Automation
    - Time Dependent
    - Temperature Dependent
- Titration
  - Setup
  - pH Titration Measurement
  - Additive Titration Measurement
- Sample Parameters
  - Liquid
  - Particle
- Data Analysis
  - Model

Of the items above, the ***options which must always be specified*** are:

1) **Identification**, in which the user sets the **Sample ID** and inputs any notes to accompany the saved data.
2) **Measurement → Parameters**, in which the user sets either a **Number of Cycles** per measurement (typically 30), or a **Target Residual** value for the raw data fit. The number of cycles accumulates data: the final cycle determines the zeta potential measurement for each particular data point. The number of data points is set in the next item. The Target Residual option is recommended for samples with low mobility.
3) **Measurement → Automation → Time Dependent**, in which the users sets the number of **Total Measurements** (typically 5, but can be increased for more statistics, as is recommended for low-mobility materials), as well as the **Time Interval Between Measurements**, which is typically 0 unless a time course is required. The interval is the number of seconds between the end of one measurement and the beginning of the next.
4) **Sample Parameters → Liquid**, in which the dispersing fluid is specified. The default solvent is **Water**, and the dropdown menu contains several common solvents. If the solvent information is missing, the user must select **Unspecified** in the dropdown menu and input the solvent Viscosity, Refractive Index, and Dielectric Constant in order to obtain meaningful data. pH may also be entered, but will not affect the measurement.

Other options, ***some of which are important for non-polar solvent measurement,*** include:

1) **Instrument Parameters** allows the user to select the cell type. The default **BI-SCP** refers to the Square Plastic Cuvettes used for aqueous samples. **BI-SCGO** must be selected when the Square Optical Glass Cuvettes are used for non-aqueous samples. Similarly, the electrode assembly defaults to the standard electrode **BI-ZEL (1.25 mL)**, but **BI-SREL (1.25 mL)** must be used and selected if the Solvent Resistant electrode is required for non-aqueous samples.
2) **Measurement → Automation → Temperature Dependent** allows the user to specify Starting and Final Temperatures, as well as the Temperature Increment. Note that the Total Measurements requested in Item 3 above will be made for each temperature in the automation series.
3) **Sample Parameters → Particle** does not affect the measurement, but is included in the full Data Summary, and may be helpful for future reference.
4) **Data Analysis → Model** defaults to **Smoluchowski**, for aqueous samples. The **Huckel** option should be selected for non-aqueous samples or samples with very low salt concentrations, and therefore long screening lengths. The third option, the **Henry** model, is identical to the **Huckel** model.

Note: Titration instructions are contained in a separate section below.

Upon completion of specifying your SOP, you may either "Save" to overwrite or "Save As," and then click "OK," which closes the SOP window and returns to the Measurement Window. ***It is recommended that you include your name or NetID in the name of your SOP.***

Preparation of PALS sample

Place 1.7 mL volume of sample into a square plastic cuvette, and insert the electrode into the cuvette. A sample volume of 1.7 mL should allow the electrode enough room in the cuvette with neither spillage nor air bubbles remaining. Place the sample into the sample chamber and plug in the electrode. Close the top cover of the chamber when the sample is in place. **IMPORTANT NOTE**: For non-aqueous samples, use the square glass cuvettes and the **BI-SREL** electrode. ***Glass cuvettes are not disposable***; be careful using them and clean them for the next user.

Begin measurement in the PALS Measurement Window:

Once the sample is in the sample chamber, select "Start" in the upper left hand corner of the Measurement Window. The instrument will equilibrate the sample chamber temperature, adjust the incident laser intensity, and begin measurements. The plot displays the raw data with the fit overlaid. The Residual value displayed on the left shows root-mean-squared residual between the data and the fit.

pH TITRATION OVERVIEW

A dedicated training session is required for pH Titration measurements.

Usage Instructions

1) Turn the titrator unit On.
2) Calibrate pH probe: from top menu of Measurement Window, select **Titrator → Setup and Diagnostics → Recalibrate Probe**. Follow the instructions to calibrate the probe using the buffer solutions provided at pH 4, 7, and 10. If any of the options are greyed out, be sure the Titrator unit is properly turned on; the Measurement Window may need to be closed and re-opened to connect to the titrator.
3) Once the probe is calibrated, gently insert the probe in through the hole in the top of the titrator unit.
4) Confirm the pumps are primed by selecting **Titrator → Setup and Diagnostics → Prime All Pumps**. All four tubes in the titrator unit should drip acid or base into the sample cup. Once they do, hit Cancel.
5) To rinse the cell before starting the measurement, use the upper menu to select **pH Titration → Rinse Cell**. The instrument will empty the cell and prompt the user to remove the sample cup, rinse the impeller, siphons, and pH probe, fill sample cup with rinse liquid, and replace it. The rinse liquid is typically DI water for aqueous samples, or can be the suspending phase for non-aqueous samples. ***It is good practice to rinse the cell both before and after measurements, especially given that the instrument is a part of a shared facility.***
6) Configure SOP as desired for titration measurements (see below).
7) Hit **Start** in the Measurement Window to begin. The software will prompt you to rinse the cell before beginning, and will also prompt you to place your sample in the sample cup.

pH Titration SOP Window

1) **Titration → Setup** indicates the **Contents**, **Type**, **Description**, and **Concentration** of the 4 titration bottles. The fluids provided are: 1) Nitric Acid, 0.1 M, 2) Nitric Acid, 1 mM, 3) Potassium Hydroxide 1 M, and 4) Potassium Hydroxide 1 mM. ***Do not change this information***.

2) **Titration → pH Titration Measurement** contains a check box **Automate pH Titration**. When selected, the menu allows the user to set the **Initial** and **Final pH** as well as the **Number of pH Steps** desired for the measurements. The **Initial Sample Volume** must be entered accurately for efficient pH adjustment. The **pH Adjustment Resolution** is typically set to 0.2; smaller values increase the time required to adjust the pH at each pH point. Note that the measurement protocol specified in the rest of the SOP will apply to ***each*** pH point. **NOTE**: the pH values adjusted by the titrator may not be accurate. It is recommended to check the actual pH values after measurement. These can be found in the right hand side of the Main Window upon selecting the data in left hand side.

DATA TRANSFER AND INSTRUMENT SHUTDOWN

Saving/Exporting Data

You may save data at any time to .xls or .csv format by selecting your data in the left side of the Main Window and using the **Export Data** option from the File menu at the top of the window. Additional data saving options may be found by selecting your data in the left side Main Window and exploring the options available in the upper portion of the right side of the Main Window. You may save your data into a Folder within "**Documents → Research**" labeled with your NetID, and/or transfer your data via USB.

Instrument Shutdown and Cleanup

1) Turn the instrument off using the switch at the back. Turn the titrator off, if necessary.
2) Close the Software window.
3) Log Out of the computer from the Windows Start Menu.
4) **Dispose of all waste in an appropriate fashion**. If you have generated a lot of liquid waste (more than a few mL) or non-aqueous waste, carry it out with you for proper disposal in your lab.
5) **Clean and return** all non-consumables to their proper locations, including the electrodes, glass cuvettes, and the pipettor.
6) After titration measurements, select **pH Titration → Rinse Cell**. Return the pH probe to the buffer solution. Fill a sample cup with DI and place it in the sample holder to slightly immerse the propeller and siphons.