H. Darling
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DynaStar NanoPro DLS User Guide

The DynaPro NanoStar is a dynamic light scattering (DLS) instrument that is used for the analysis of protein solutions, promiscuous inhibitors, micelles, buffers or other products in solution.

Sample Preparation:

1. Concentration of 1 mg/mL is an ideal starting point
    a. Bring extra buffer to dilute
    b. A lower concentration may work, but it would just have to be tested

2. Samples can be run as-is, centrifuged, or filtered.
    a. Unspun – 5uL
    b. Centrifuged – 10uL
    c. Filtered – 100uL (accounts for dead volume)

3. Run a blank if using surfactant or glycerol

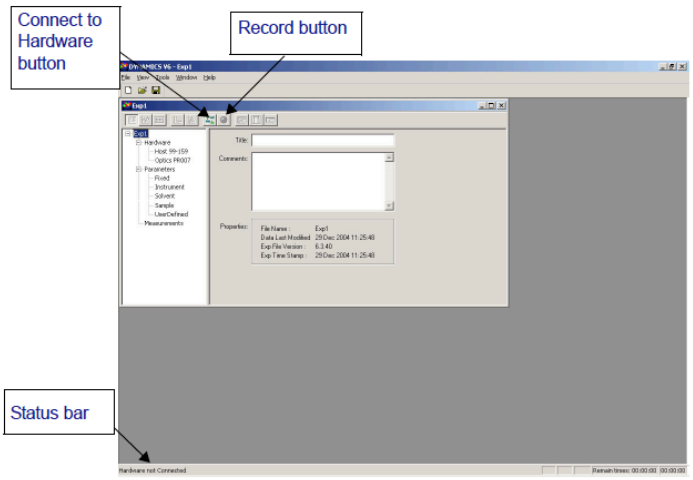
4. Cuvette types
    a. Disposable:
        - Minimum volume =5uL
        - For dynamic light scattering only
        - Tolerates temperatures up to 80°C
    b. Quartz:
        - Minimum volume =1.25uL
        - For static light scattering or dynamic light scattering
        - Used when you have precious protein and/or organic solvents
        - Tolerates temperatures >80°C
        - Replacement cost is $2,500

5. Temperature control
    a. System can be heated to 150°C. If heating a sample add ~100uL of paraffin or silicon oil around the chimney to prevent evaporation
    b. Our system is not currently set up for runs below 20°C. We would need a dry gas source (i.e. nitrogen)

Operation:

1. Power on instrument, laser, and computer
    - Username: wyattlab
    - Password: dynapro
    - Wait 30 minutes before running any experiments
2. Launch “Dynamics” software
    a. File -> open -> preset (configuration/method file)
    b. File -> open -> previous experiment (for comparative reasons i.e. buffer conditions)
    c. File -> open -> new (NOT recommended per technician, saves settings from previous user)

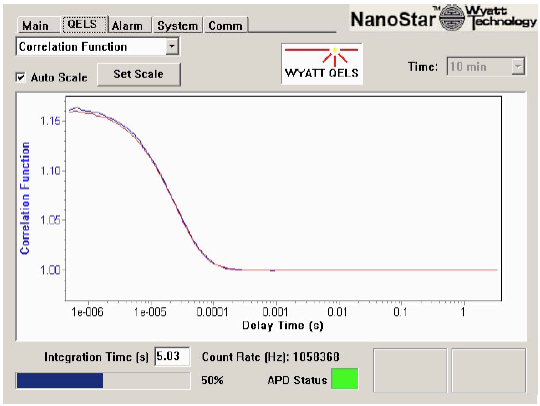
3. Using Preset:
    - Auto attenuation— system focusing is like a pupil and will adjust based on light
    - Automatic 5 minute delay for temperature stability
    - Standard data acquisition is 3 measurements, but can take 5+ if better statistics are needed.
4. Click “connect” icon in toolbar
    - Record button will activate (green circle)
    - Trouble? – Disconnect/connect Ethernet cable then retry or contact Heather Darling (h.darling@vanderbilt.edu room 5128C)



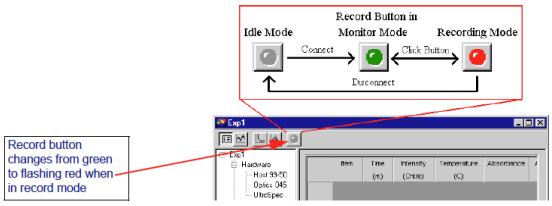
5. Double click on event schedule
    - Under commands and values -> label sample -> enter
    - Save data as -> click edit to open directory
    - Verifies all data is going to this file
6. Load sample in cuvette
    - Insert pipette tip all the way to the bottom of cuvette then slowly pull up while volume is dispensed. This helps prevent air bubble formation
    - For disposable cuvette the sample goes in the center “chimney” indicated below by the dark purple color

7. Place cuvette in cell chamber and close lid
    - Disposable -> make sure “front” faces forward
    - Quartz -> flattened corner positioned on the left front side
8. On the Dynapro instrument highlight the “QUELS” tab
    - Quels = quasi electric light scattering
    - No correlation/flat line indicates no protein in sample (i.e. buffer), air bubble, or cuvette is not pushed all the way in

- Correlation (like below) indicates protein



9. Acquire data -> push green icon on computer
    - Arrow in left column will point to which step the system is on

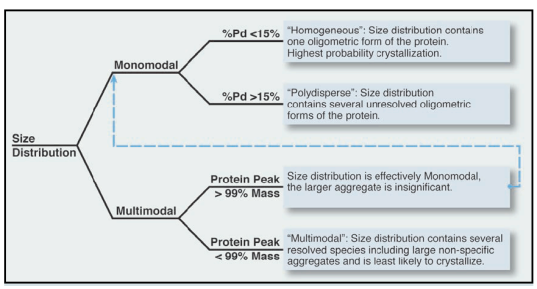


10. Analysis can begin once the green icon comes back -> see next section

Analysis:

Dynamics software can be installed in any computer on campus. Return the disc and license key once this is complete!

1. Tools -> calculations -> optimization
    - Insert molar mass
    - Helps understand expected size
    - Can be used to figure out minimum concentration needed
2. Fit models/data correlation
    - Postulates size distribution as Gaussian (radius, polydispersity)
    - Multiexponential analysis (regularization)
3. If all red data (thrown out data):
    - Right click -> select data filter
    - Lower SOS = better Gaussian -> this can be disabled
    - Tweak baseline to 0.05



Finishing and clean up:

1. Remove cuvette
    - Disposable – discard
    - Quartz – clean using cuvette washer in Megalab
2. Turn off the laser and Dynapro instrument
3. Export data via internet and/or flash drive
4. Log off computer
5. Have a nice day ☺