

ARTISAN®
TECHNOLOGY GROUP

Your **definitive** source for quality pre-owned equipment.

Artisan Technology Group
(217) 352-9330 | sales@artisantg.com | artisantg.com

Full-service, independent repair center
with experienced engineers and technicians on staff.

We buy your excess, underutilized, and idle equipment
along with credit for buybacks and trade-ins.

Custom engineering
so your equipment works exactly as you specify.

- Critical and expedited services
- In stock / Ready-to-ship
- Leasing / Rentals / Demos
- ITAR-certified secure asset solutions

Expert team | Trust guarantee | 100% satisfaction

All trademarks, brand names, and brands appearing herein are the property of their respective owners.

Visit our website - *Click HERE*

Protein Solutions Dynamic Light Scattering Machine Protocol

Version Number: 2
Date: 05/19/04, 9/11/06
Authors: J. Jancarik/ R. Pufan
Edited by: R. Kim
Reviewed by: D. Das, A. DeGiovanni

Materials/Reagents/Equipment	Vendor
Equipment	
DynaPro99 (1 Rm 308) & Dyna Pro (2 Rm 115) Serial number: 99-24 (1) & 2004-020 (2) Model: MSHTC/12 (1) & MSXTC (2)	Wyatt Technology Corp., 6300 Hollister Ave., Santa Barbara, CA 93117 Tel. 805-7=681-9009 http://www.wyatt.com Cost: $31,329.44 Previously known as Protein Solutions/DynaPro instruments.
Cuvettes 12 µl – Cat #: 003030 2 µl	

IMPORTANT: DO NOT CHANGE ANY of the settings on the instruments. If you are uncertain as to how to proceed or if there is anything wrong contact Jarmila Jancarik or Debanu Das. Mark in the log book next to the machine any problems: date, name, describe problem.

Procedure

1. Turn the power on the DynaPro 99 and the Temperature Controller (both switches on the front) – warm up is 15 min.

2. Wash thoroughly the 12 µl cuvette with 0.2 µ filtered distilled water. DO NOT use anything else. Cuvettes are stored in chromic/sulfuric or nitric acid, so rinse thoroughly. Use 0.2 µ filtered distilled water also as your standard by pipetting 20 µl into the cuvette. The cuvette has windows on three sides so **do not** put your fingers over the windows (if necessary use Kim wipes to clean the outside). Tap gently on counter if you see any bubbles. Put the lid on the cuvette.

3. Insert the cuvette into the holder with the frosted side to the left.

4. On the desktop double click on the Dynamics V6 icon. **First time users**: click on the "DLSFolder" icon on the desktop and create a folder for yourself; this is where you will be saving all of your data. For each protein open a new experiment file. When opening a new

Artisan Scientific - Quality Instrumentation ... Guaranteed | (888) 88-SOURCE | www.artisan-scientific.com

experiment the machine will connect automatically – a green circle will appear on the toolbar. When opening a preexisting experiment, you must click on the double arrow button to connect. The circle on the next button will become green. You are now ready to start reading.

5. First you need to test the water to make sure that your cuvette is clean. Click on the icon to the right of the green circle and look at the readings that appear under intensity. For the instrument in room 308, the counts should be around 8000; for the one in room 115, the counts should be around 13000. The fluctuations should not be more than 1000 up or down, otherwise you need to rinse the cuvette more; if still dirty, then add chromic/sulfuric or nitric acid solution and let it soak either overnight or at least 5 min. Once the cuvette is clean close this screen, NOT the program.

6. Remove water. Make sure to spin your sample for a few minutes on a tabletop centrifuge before reading. Pipette 20 µl (12 µl is the minimum volume) of your protein at 1-2 mg/ml and insert it into the cuvette. Insert cuvette into DynaPro99.

7. Click on the green button on the Experiment window. The acquisition time is set to 10 seconds for a total of 20 acquisitions. If your sample is aggregated or precipitating the instrument will give a warning with three options: resume, stop, and help. If you don't do anything the experiment will continue, after the third warning the software will disconnect to protect the laser. If you hit resume, the experiment will also continue. DO NOT hit resume more than five times, as the laser will be destroyed. If you don't want to be disconnected from the instrument take your sample out as soon as possible. If you hit stop the machine will also disconnect.

8. If you still want to get an idea about your sample you have two options. For instrument # 1, dilute your sample; for instrument # 2 if you click on the button next to the round circle – the same one you use to check the water – on the left side of the screen under laser power, you can decrease the power as low as 10%. Make sure you mark on your experiment the % you read at if it is not 100. When you are done, make sure to bring the intensity back to 100%. DO NOT change anything else.

9. The 4th button on the main window gives the correlation graph, and the 5th button on the window gives the regularization, which is the histogram.

10. The ideal results for the protein should be: radius – between 2 and 5 nm and the polydispersity less than 20%. If the results are higher than that, try increasing or decreasing the salt concentration, add DTT or TCEP if cysteines are present, glycerol or propanediol or any reagent the purifier may recommend. Save your readings in your folder on the computer. If you need to print your results go to **File**, select **Print**. Check on Correlation View, Regularization View and Results Summary.

Artisan Scientific - Quality Instrumentation ... Guaranteed | (888) 88-SOURCE | www.artisan-scientific.com

11. After you are done, close the Dynamics V6 program and switch off the DynaPro99 and the Temperature Controller. Wash cuvette with 0.2 µ filtered distilled water. Add acid and cap the cuvette for storage.

12. Software remarks: at times you will notice a 0% polydispersity. This is due to a mathematical calculation. In such cases assume that the polydispersity is less than 15%. In dilute samples you will get a buffer peak. This will be the first peak on your regularization view. You can eliminate it by unmarking it at the bottom of the page.

NOTE: Dynamic light scattering should be performed pre and post SEC. The fractions should be pulled based on the DLS results. DLS should also be performed to check on the condition of stored proteins if they need to be used for further set ups.

Artisan Scientific - Quality Instrumentation ... Guaranteed | (888) 88-SOURCE | www.artisan-scientific.com

Artisan Technology Group is an independent supplier of quality pre-owned equipment

Gold-standard solutions

Extend the life of your critical industrial, commercial, and military systems with our superior service and support.

We buy equipment

Planning to upgrade your current equipment? Have surplus equipment taking up shelf space? We'll give it a new home.

Learn more!

Visit us at **artisantg.com** for more info on price quotes, drivers, technical specifications, manuals, and documentation.

Artisan Scientific Corporation dba Artisan Technology Group is not an affiliate, representative, or authorized distributor for any manufacturer listed herein.

We're here to make your life easier. How can we help you today?

(217) 352-9330 | sales@artisantg.com | artisantg.com

ARTISAN® TECHNOLOGY GROUP