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FAQ: 90Plus

Questions and Answers for Driving the Brookhaven **90Plus**

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1. What is the baseline index?

The baseline index (BI) is a measure of how much the autocorrelation function was distorted by a few large particles. Often referred to as "dust", they could be a few aggregates, too few for this technique to characterize properly. A high value (10) corresponds to no dust and a low value (0) corresponds to a noticeable amount of dust. During data analysis, the experimental autocorrelation function must be normalized. In theory, one should use the infinite time baseline B_∞ obtained from the square of the average counts per sampling time. However, in practice, in order to eliminate the effect of a few large particles (dust), a baseline obtained from the measured correlation function at some long delay time is a better choice for normalization. During measurement, this constant background term is measured using a variety of possible methods (see below) to obtain a measured baseline value B_m. For $(B_m/B_\infty-1) < 0.01$, that is a less than 1% difference in baselines, the index is defined by

$$\mathbf{BI = 10\,[\,1 - 100\,|(B_m/B_\infty-1)|\,],} \qquad \mathbf{if\ |(B_m/B_\infty-1)| < 0.01,}$$
$$\mathbf{otherwise,\quad BI = 0.0}$$

If the measured baseline is 1% or more above the infinite time baseline, or the measured baseline is below the infinite time baseline, the baseline index is 0. If the baselines agree exactly (a good thing), the baseline index equals 10. At a 0.1% baseline difference, the baseline index is 9 (which is quite good). If the BI value is 8 or higher, it means that the two measures of the correlation function baseline are in good agreement. Even if the value is lower than 8, if you have reasonable results and repeatable results, then the software has taken care of the less than perfect sample. The low value for baseline index is to alert you about what is going on. If you make a new sample, consider removing the few large dust particles by, for example, filtration or centrifugation. In previous versions of the **90Plus** software the BI was referred to as Sample Quality. The two names refer to exactly the same value, only the name was changed.

2. Why are baselines important?

The first step of data analysis is to normalize the experimental autocorrelation function by dividing each data point by the baseline. These normalized data are then analyzed to obtain particle size. In the most common case, cumulant analysis, the logarithm of the normalized data minus one is fit to a second order power series in delay time. A good baseline index indicates that the data is of high quality, e.g., free of dust. Don't worry if the index is poor; the **90Plus** uses advanced algorithms to recover as much information as possible. If you have reasonable results and repeatable results, then the software has taken care of the less than perfect sample.

3. How are baselines changed in the software?

Click on the menu item: `Setup` → `Normalization`. Select the method desired. Normally the "Autoslope (Slope Analysis)" method is selected. This is described below.

4. **Why are different baseline options available?**

   This is one way that you can recover from an imperfect sample. Varying the baselines changes the way large dust particles are treated in the analysis.

5. **What is *Autoslope* (Slope Analysis) normalization?**

   In this case, the data are examined starting halfway down the autocorrelation function and a series of straight line fits are performed until the slope is less than a small, positive value. Effectively, this is where the decaying autocorrelation function is flat or nearly so. For most samples this works nicely and we recommend using the Autoslope baseline. Sometimes, there are correlation functions that are very noisy or have two distinct decays. In these cases, the Autoslope (Slope Analysis) routine may set the baseline prematurely. In such cases we recommend either "Last Channels" or "Calculated" as baseline choices.

6. **What is *Last Channels* normalization?**

   This places the last eight baseline channels at the last eight delay times. If the baseline index (BI) improves and repeatability is acceptable then the results may be better. However, if there are large variations in Effective Diameter and Polydispersity then there may be some dust and you are better off with Autoslope.

7. **Must a measurement be made again in order to use a different baseline?**

   No. Just recall the data (file/database) and change the normalization method to see the results.

8. **What is the *Dust Filter*?**

   The dust filter is an algorithm that rejects data that are corrupted by scattering from dust. You can turn the "dust filter" on or off during a measurement and you can recall data and turn the dust filter on and off retroactively. However, during and after a measurement, a new filter value cannot be entered. If the dust cutoff value is too high, 100% of the light is accepted for correlation and you have not "clipped" the high or "dust" contributions. Note that for very clean samples, one expects that 100% of the data is retained, provided the dust cutoff is not set too low. If the dust filter value is too low, then the reported effective diameter will be too low.

9. What is the exact algorithm for the *Dust Filter*?

In the data collection the scattered intensity is measured over many short, consecutive time intervals. The data are divided into two sets. The first data set, "unfiltered" includes all of the collected data. The second data set, "filtered" is filled by using the following procedure. The moving mean, I_{mean}, and standard deviation, σ_m, of five sequential intensity values is calculated. On the sixth measurement, the intensity, I_{new}, is compared with the average of the previous five using the dust filter cutoff value "C." If the following test is true, the data is accepted into the second "filtered" data set.

$$| I_{new} - I_{mean} | < C\sigma_m$$

If the test is false, then the data from this measurement is not added to the "filtered" data set. Thus, the "filtered" set is only the result of measurements that meet the inclusion criteria for the dust filter. The autocorrelation function is accumulated and kept for both sets. In this way, the user can turn the dust filter on or off during or after a measurement to observe the effects of data filtering on the results. However, the dust filter cutoff value cannot be changed once a measurement has begun.

10. What value is appropriate for the *Dust Filter*?

Start with a value of 30. The optimal value of the dust filter cutoff is actually related to the expectation of outliers in intensity, which is related to the size of particles being measured. For particle sizes on the order of 10 nm, use a value of about 15. For particle sizes on the order of several 100 nm, use a value of 30. For particle sizes on the order of 1 micron, use a cutoff value of 80. To truly optimize the dust cutoff value, lower the dust cutoff filter until about 98% of the data is retained.

11. How do I know how much data is retained for a given *Dust Filter* setting?

When the dust filter is on, the percent data retained is displayed to the right of the baseline index.

12. What is the *Effective Diameter*?

The effective diameter is the particle diameter determined by a dynamic light scattering measurement. The effective diameter is calculated from the Stokes-Einstein equation (for spheres, see question 27) from the experimentally determined, translational diffusion coefficient. The translational diffusion coefficient is calculated from the accumulated autocorrelation function of the scattered light using the method of cumulants. Thus, the translational diffusion coefficient is intensity-weighted. The Stokes-Einstein equation shows that the hydrodynamic diameter is inversely proportional to the diffusion coefficient. Therefore, it is wrong to say the diameter determined this way is intensity-weighted. The weighting is actually the inverse of the intensity weighting of the reciprocal of the diameter. An average calculated in this way is sometimes called the harmonic mean.

13. What is the *Mean Diameter*?

Consider N runs, each yielding an Effective Diameter. The average of these Effective Diameters (sum divided by N) is called the Mean Diameter.

14. What is the *Combined Diameter*?

The combined diameter is the diameter calculated by treating the data as if it were taken in one long run rather than a series of individual runs. Deselected runs are ignored.

In a size measurement, there are multiple runs. Each run results in an autocorrelation function made up of M correlation coefficients (one for each delay time, τ). Summing the correlation coefficient for a particular delay time over all runs will give the correlation coefficient that would result from a single, long run. Repeating the process for all delay times will then give the correlation function that would result from a single, long run. Finally, this averaged function is analyzed using cumulants to obtain an intensity-weighted average translational diffusion coefficient. The Stokes-Einstein relation (see question 27) is then applied to the averaged translational diffusion coefficient to calculate an average diameter. This is the Combined Diameter.

15. Which one do I use, the *Mean Diameter* or the *Combined Diameter*?

If the Mean Diameter and the Combined Diameter are equal to within one or two standard errors, then there are no statistical outliers and the data are acceptable. In this case, use the Mean Diameter to characterize the central tendency of the size distribution. This is one way to characterize the "size" of a distribution. It is often the safest way to do so.

16. What if there are outliers?

If an Effective Diameter differs from the Mean Diameter by more than twice the standard error, it is an outlier. Delete the outlier by double clicking on the line of data corresponding to the run. The text on that line will turn gray. Deletion can be reversed by double clicking on the same line.

17. What is the upper limit for count rate?

The upper limit for count rate is 300-500 kcps (kilocounts per second) for the avalanche photodiode (APD) detector and 700kcps to 1 Mcps (1 million counts per second) for the photomultiplier tube (PMT) detector.

18. What is the lower limit for count rate?

The lower limit for count rate is 10-100 kcps. The lower limit is really due to a desire to keep measurement durations short. A lower count rate means a measurement will take longer to complete. At low count rates, over a given time period, fewer photons are reaching the detector and there is less data with which to build the correlation function.

19. How do I automatically adjust the incident intensity, and thereby the count rate?

Instruments with a BI-APD (avalanche photodiode) high sensitivity detector and instruments with zeta potential measurement options are equipped with a circular variable neutral density filter. Automatically adjust the count rate by choosing `Setup` → `Incident Power Settings`. Choose the radio button "Optimize Incident Power at start of each measurement." Close the dialog box. When the start button is pressed, the count rate will be optimized so that it is below the upper limit for the count rate. In this process, the neutral density filter wheel will be adjusted such that the count rate is lower than the maximum count rate for the instrument.

20. How do I manually adjust the incident intensity, and thereby the count rate?

Instruments with a BI-APD (avalanche photodiode) high sensitivity detector and instruments with zeta potential measurement options are equipped with a circular variable neutral density filter. Manually adjust the count rate by choosing `Setup` → `Incident Power Settings`. Choose the radio button "Maximize Incident Power at start of first measurement." Close the dialog box. When the start button is pressed, the count rate will be maximized by adjusting the position of a neutral density filter wheel. During this process, click "Bypass" when the desired count rate appears. Or, the software will automatically maximize the count rate. Of course, count rate can also be decreased by lowering the concentration of particles in your sample.

21. How do I export results as plain text (ASCII) to a spreadsheet such as Excel?

Open up a measurement and choose the data you wish to export, such as MSD. Press the "Zoom" button in the upper right of the screen. If you choose Lognormal or MSD data, then press "Lognormal Summary" or "MSD Summary". In the resulting window, press "Copy for Spreadsheet." The data will then be placed on the clipboard as comma delimited data. You can save the data as a text file and import it into Excel, choosing a comma as a delimiter.

22. How do I send data to Brookhaven Instruments when I have a problem?

See question 24, "How do I save the data so I can analyze it at my desk?" Then, e-mail the resulting file to Brookhaven Instruments.

23. How do I send an archive file?

See question 24, "How do I save the data so I can analyze it at my desk?" Then, e-mail the resulting file to Brookhaven Instruments.

24. How do I save the data so I can analyze it at my desk?

Within the software, click on `File` → `Database`. Then, highlight the measurement(s) you wish to send and press the "Archive Selected File(s)" button. A standard save file dialog will appear. Choose a descriptive file name then save the file. This file contains

many details about the measurement that are not available in a screen shot or printed report.

25. Why is 10 mM KNO_3 used as a diluent for the latex reference material?

See question 26, "Why not just use distilled water as a diluent?"

26. Why not just use distilled water as a diluent?

In order to calculate particle size from dynamic light scattering results, the Stokes Einstein equation is used (see question 27). An underlying assumption in the calculation is that the particles are freely diffusing. For dilute electrostatically stabilized dispersions, long range interactions distort free translational diffusion. Adding salt screens the particle-particle interactions, making the assumption of free diffusion and therefore the Stokes Einstein equation to be valid. 10 mM aqueous KNO_3 is often a good choice for a diluent. Most aqueous dispersions, such as the anionic latex used to validate the performance of the 90Plus, are electrostatically stabilized.

27. What is the Stokes-Einstein Equation?

The Stokes-Einstein equation relates the diameter of a spherical particle to the translational diffusion coefficient. It is

$$D = \frac{k_B T}{3\pi\eta d}$$

where

D: Translational diffusion coefficient

k_B: Boltzman's constant

T: Absolute temperature

η: viscosity

d: diameter

This equation is used to transform diffusion coefficients determined by dynamic light scattering into effective diameters presented as measurement results.

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