Bettersize
BETTER PARTICLE SIZE SOLUTIONS

APPLICATION NOTE

Characterizing the size and size distribution of monosaccharide molecules

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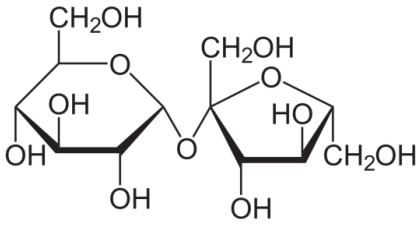
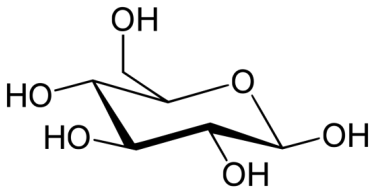
Introduction



Structural formula of sucrose (molecular weight 342.3Da)



Structural formula of glucose (molecular weight 180.16 Da)

The lower limit of effective particle size detection by dynamic light scattering (DLS) technique is influenced by various factors, including the instrument's optical path design, laser power, detector type, and correlator's computational ability. This limit has been a matter of concern for users.

Previous studies often verified the lower detection limit of DLS using small molecule samples, such as sucrose (molecular weight: 342.3 Da) and vitamin B1 (molecular weight: 337.29 Da). These small molecules, falling within the range of 300-400 Da, pose significant challenges for DLS equipment due to their extremely low molecular weights, weak scattering signals, and rapid decay of correlation functions.

In this application note, a challenge was undertaken using the BeNano nanoparticle size analyzer from Bettersize to measure glucose, a sample with even lower molecular weight. Glucose has a smaller molecular size compared to sucrose and vitamin B1, with a molecular weight of only 180.16 Da, and its scattering ability is lower compared to sucrose samples, making the measurement even more demanding.

Instrumentation

The BeNano 180 nanoparticle size analyzer from Bettersize Instruments was utilized for the experiment. It incorporates a laser source with a wavelength of 671 nm and a power of 50 mW. Scattered light signals were acquired using an APD detector positioned at an angle of 173 degrees. To optimize the signal-to-noise ratio, single-mode optical fibers were employed for signal transmission. The BeNano analyzer is equipped with a high-speed correlator operating at the nanosecond level, enabling fast correlation calculations for the rapid decay of correlation functions associated with small particles.

Viscosity correction

Since the viscosity of aqueous solutions of sucrose and glucose varies with concentration, viscosity information was corrected prior to measurement. To accomplish this, polystyrene spheres with a nominal diameter of 100 nm were added to the solutions. By measuring the diffusion coefficient (D) of the polystyrene spheres, viscosity information was obtained using the Stokes-Einstein equation, which relates viscosity (η) to diffusion coefficient (D) and particle diameter (d_H).

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

This approach is commonly employed in DLS to determine the viscosity of unknown solutions, particularly in cases where sample volume is limited and cannot meet the requirements of rheometers or viscometers. Given that sucrose and glucose molecules are small and exhibit extremely weak scattering, their contribution to the signal relative to the 100 nm polystyrene spheres is negligible. The measured viscosities are presented in Tables 1 and 2.

Table 1. Glucose samples

No.	Solution concentration	Solution viscosity
1	20%	1.53 mPa.s/cp
2	10%	1.15 mPa.s/cp

Table 2. Sucrose samples

No.	Solution concentration	Solution viscosity
1	20%	1.77 mPa.s/cp
2	10%	1.25 mPa.s/cp

Experimental

Solutions of glucose and sucrose at concentrations of 10% and 20% were prepared. The BeNano's built-in temperature control system maintained a test temperature of 25°C ± 0.1°C. Given that the sample particles are extremely small and exhibit weak scattering, the presence of impurities like dust can significantly impact the detection process. To address this, a 220 nm PTFE filter was used to filter the sample during the experiment. The filtered sample was then directly injected into the cuvette.

Each sample was measured at least three times to assess the reproducibility of the results and to obtain the standard deviation of the results.

Results and Discussion

Based on the raw scattered light signals, the correlation functions and size distribution results are obtained.

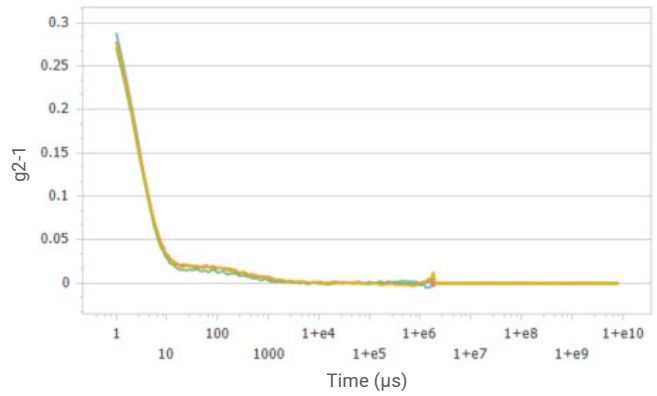


Figure 1. Correlation functions of glucose solution at a concentration of 20%

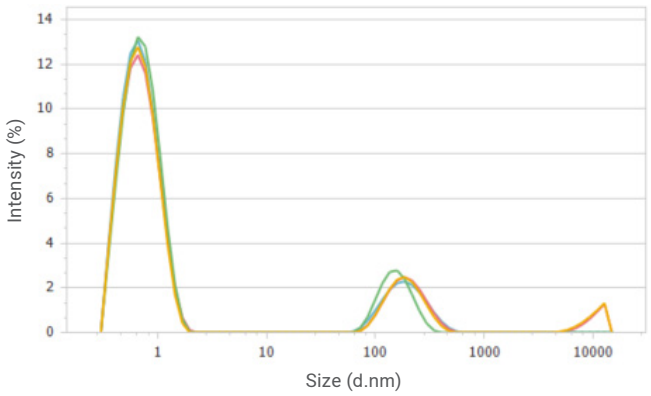


Figure 2. Intensity distributions of glucose solution at a concentration of 20%

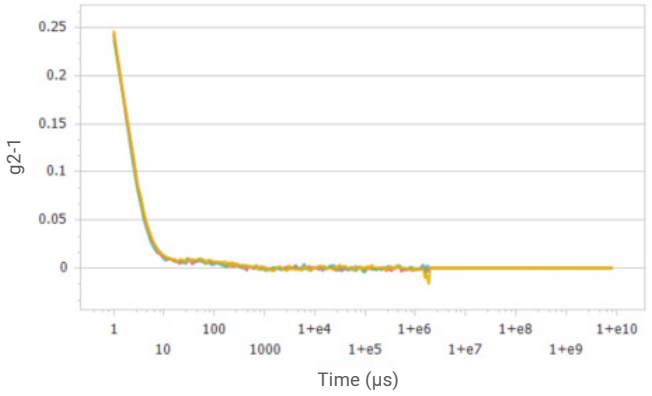


Figure 3. Correlation functions of glucose solution at a concentration of 10%

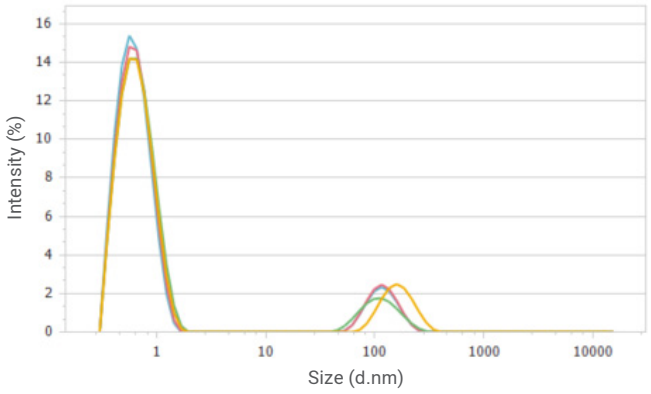


Figure 4. Intensity distributions of glucose solution at a concentration of 10%

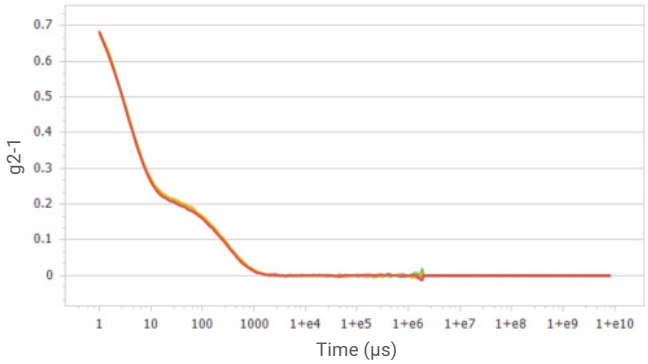


Figure 5. Correlation functions of sucrose solution at a concentration of 20%

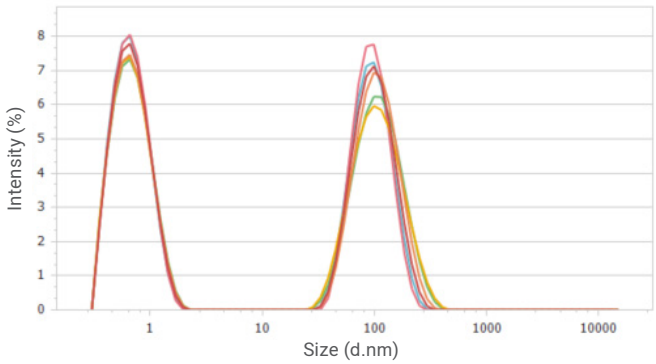


Figure 6. Intensity distributions of sucrose solution at a concentration of 20%

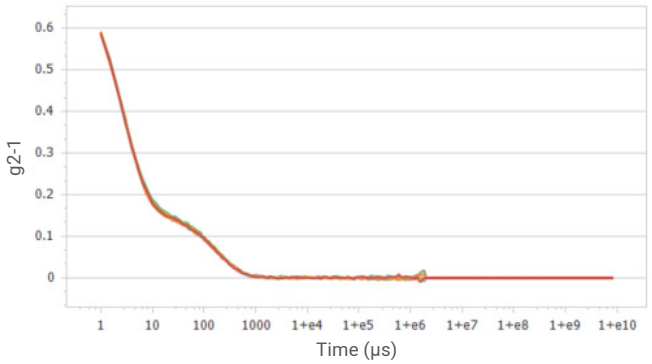


Figure 7. Correlation functions of sucrose solution at a concentration of 10%

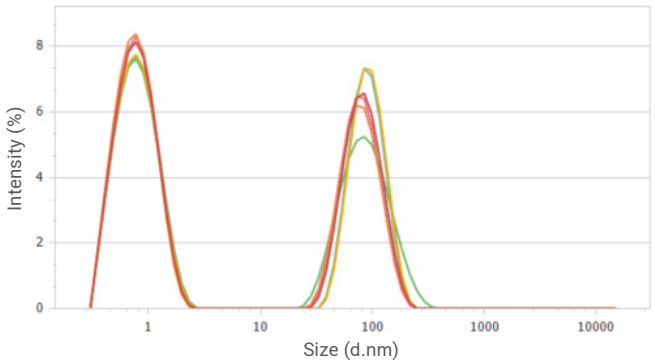


Figure 8. Intensity distributions of sucrose solution at a concentration of 10%

The correlation functions show a rapid decay for small particles, indicating their faster Brownian motion compared to larger particles. These correlation functions exhibit a favorable signal-to-noise ratio and demonstrate high repeatability. The correlator's capability to perform very-short-time correlation calculations ensures a sufficient number of signal points on the correlation function, enabling accurate determination of small particles.

Table 3. Particle size results for sucrose and glucose

Sample Name	Average hydrodynamic size (d.nm)
20% sucrose	27.29±1.01
10% sucrose	11.86±0.93
20% glucose	0.98±0.01
10% glucose	0.81±0.06

Figures 1, 3, 5, and 7 display the correlation functions for multiple tests conducted at different concentrations. The results demonstrate excellent reproducibility of correlation functions for sucrose and glucose solutions, exhibiting two distinct decays. This indicates the presence of at least two or more fractions with significantly different particle sizes in these solutions. Previous studies reveal the existence of large molecular aggregates in sucrose solutions, which aligns with the particle size distribution curves depicted in Figure 6 and Figure 8.

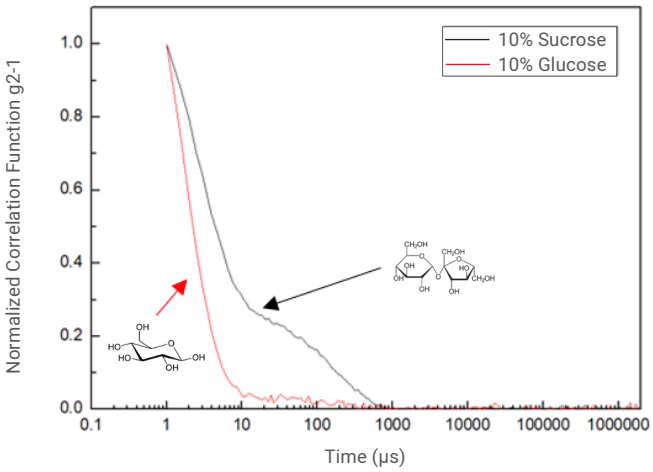


Figure 9. Normalized correlation functions for sucrose and glucose at a concentration of 10%

Figure 10 illustrates the normalized correlation functions of the 10% sucrose and 10% glucose samples. It is evident that the first decay of the correlation function for the glucose sample exhibits a faster decay rate. This behavior can be attributed to the smaller glucose molecule, which possesses a higher diffusion rate.

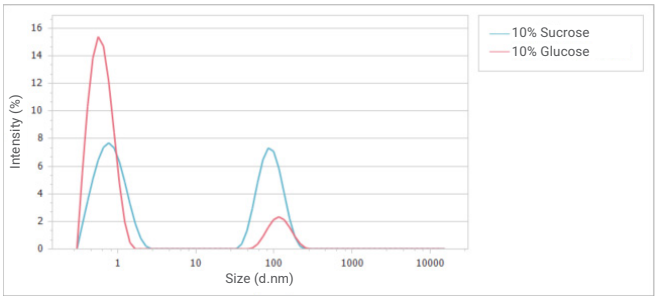


Figure 10. Overlay of intensity distributions of glucose and sucrose solution at a concentration of 10%

Comparing the size distribution of 10% sucrose and 10% glucose, as shown in Figure 10, reveals a peak around 1 nm for both samples. This peak corresponds to the contribution of monosaccharides. Additionally, an agglomerate peak around 100 nm is observed, representing the contribution of polysaccharides.

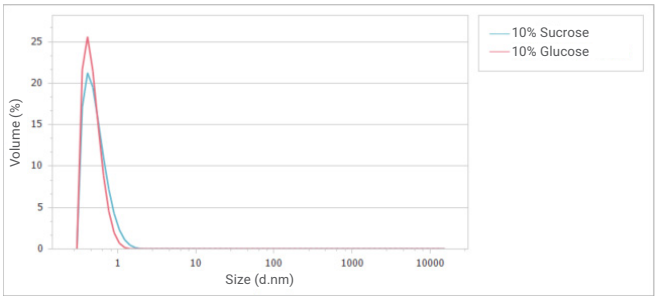


Figure 11. Overlay of volume distributions of glucose and sucrose solution at a concentration of 10%

Figure 11 presents the volume distribution curve, indicating that the majority of the two samples in solution consist of very small monosaccharides, with minimal levels of polysaccharides.

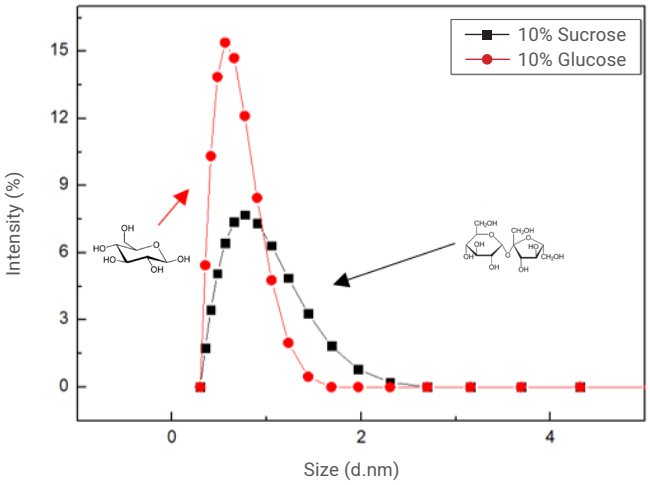


Figure 12. Magnified correlation functions for sucrose and glucose at a concentration of 10%

To gain more insights into the differences in monosaccharide sizes, the monosaccharide peaks around 1 nm are magnified, and the coordinates are changed to linear coordinates in Figure 12. It is evident that the monosaccharide peaks in the glucose solution have a narrower size range compared to those in the sucrose solution. This discrepancy is attributed to the relatively smaller size of glucose molecules.

Conclusions

This application note demonstrates the characterization of the size and size distribution of monosaccharide molecules, specifically glucose. The BeNano, equipped with a high-speed correlator, was used to measure glucose, which has a molecular weight of 180 Da. Viscosity correction was performed using polystyrene spheres, and measurements were conducted at different concentrations. The results showed distinct correlation functions and size distributions for sucrose and glucose samples, highlighting the presence of monosaccharides and polysaccharides. The BeNano system demonstrated reliable detection capabilities for small particles like glucose.

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