



# Effect of 60NM Polystyrene Latex Nanoparticles Concentration Towards Calculation of Measurement Uncertainty and Z-Score in Particle Size Analysis Using Dynamic Light Scattering Principle

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## Abstract

The dynamic light scattering technique is used to determine the particle size distribution macro and nanomolecules in colloids. The determination of size is crucial for some of the application such as nanogold production for in-vitro diagnostic rapid test kit production. The measurement of the size is very crucial as different size of particle will provide different applications propose. Therefore, during the measurement the important point of consideration is the measurement of uncertainty which contributes to the ultimate measurement of the size. There few aspect that is being considered in the measurement of uncertainty, which are reference standard, resolution, diff value of equipment and concentration of sample. However, in this study we measure the size distribution of polystyrene latex sphere (PLS) of 60nm at different concentration of 1 drop to 20 drop in 1ml of 10mM of NaCl. The experiment was carried out to prove that the concentration does not affect the measurement particle size when measured using hydrodynamic size distribution calculation. The result indicates the measurement of particle size of 1 drop to 20 drop of PLS was in the range of 64.53 to 65.62, which is in the range of hydrodynamic size of the standard PLS. Therefore, the concentration does not affect the measurement of the particle size and the effect of concentration can be omitted from the measurement of uncertainty calculation.

**Keywords:** Polystyrene, Latex, Nanoparticles, Z-Score, Light Scattering.

## 1. Introduction

Dynamic light scattering (DLS) is a technique commonly employed to measure particle distribution size in colloids. A monochromatic beam of light will be scattered in all direction when come in contact with molecules in solution. The intensity fluctuation will be caused by Brownian motion of the present molecules in the solution. By using the diffusion coefficient the hydrodynamic size of the molecule can be obtained [1]. Many factors can influence this measurement technique; hence certain parameters must be strictly adhered to or controlled to prevent measurement errors. In this endeavour, National Institute of Standards and Technology (NIST) have provided several guidelines in terms of procedures, data analysis and reporting of DLS measurements to increase accuracy and robustness of the data obtained. In the current work, the effect of particle concentration in DLS measurements of 60 nm polystyrene latex spheres were determined by calculating measurement of uncertainty (MU) and z-score values. The measurement of uncertainty is very crucial to determine the actual measurement of the sample taking in consideration of all the possible errors that can contribute to hydrodynamic diameter of the sample [2]. Besides, measurement of uncertainty calculation is a must if a measurement is carried out according to ISO 17025:2017 standard [3]. The analysis of this investigation is expected to provide clarity for other researchers in the field about the effects of sample concentration in DLS measurements and the requirement of inclusion/exclusion of sample concentration for measurement of uncertainty calculation.

## 2. Materials and Methods

Nominal 60 nm (catalog number LTX3060A and LTX3060B) polystyrene latex spheres (PLS) were purchased from Thermo Scientific. To prepare the samples for analyse, 1 drop of 60nm concentrated PLS was diluted in 1ml of filtered aqueous 10 mM NaCl and light scattering was observed at a scattering angle of 173° at temperature 25°C. The analysis was repeated by added 5, 10, 15, 20 drops of 60nm concentrated PLS, respectively. The data was collected and analyzed with particle size analyzer (Zetasizer Nano S, Malvern Instruments). Measurements were repeated five times in order to determine the z-score and measurement of uncertainty for 60nm polystyrene latex.

## 3. Results and Discussions

The experimental design for determine the concentration effect for DLS using the 60 nm polystyrene latex, and for determining the uncertainty associated with repeatability is presented. The method of uncertainty evaluation as outlined in the Guide to Expression of Uncertainty in Measurement (GUM) published by the International Organization for Standardization (ISO) were used [2-4]. The repeatability uncertainty, which is a so-called Type A uncertainty, is needed for measurement uncertainty calculation by combining with the Type B uncertainty. Table 2 showed the uncertainties of

Type B contributing to the uncertainty in the 60 nm polystyrene latex.

**Table 1:** Summary of Type A statistical analysis for particle size uncertainty

Samples	Size (nm)	Mean	Standard deviation	Standard Uncertainty, $\mu$ s	Degree of freedom
60nm (1 drop)	65.33	65.35	0.331993976	0.14847222	4
	65.34				
	65.73				
	64.83				
	65.51				
60nm (5 drop)	65.30	64.98	0.480385262	0.21483482	4
	65.35				
	64.83				
	64.20				
	65.21				
60nm (10 drop)	65.93	65.62	0.382727057	0.171160743	4
	65.63				
	65.51				
	65.03				
	65.98				
60nm (15 drop)	64.70	64.75	0.331013595	0.14803378	4
	65.23				
	64.42				
	64.92				
	64.49				
60nm (20 drop)	65.17	64.53	0.387388694	0.173245491	4
	64.49				
	64.34				
	64.14				
	64.50				

**Table 2:** Summary of Type B uncertainties contributing to the uncertainty in the 60 nm polystyrene latex

Item	Uncertainty Component, $\mu$	Value (a)	Units	Formula	Distribution	Divisor	Degrees Freedom (n-1)	Standard Uncertainty, $\mu$ s
1	Reference standard	4.00000000	nm	$u_b = a/k$	Rectangular	2.00	$\infty$	2
2	Resolution (1/2x(0.01nm))	0.00500000	nm	$u_c = a/\sqrt{3}$	Rectangular	$\sqrt{3}$	$\infty$	0.002887
3	Drift value of equipment	3.00000000	nm	$u_d = a/\sqrt{3}$	Rectangular	$\sqrt{3}$	$\infty$	1.732051

The relative combined uncertainty ( $\mu_c$ ) of the particles size measurement is expressed as

$$\text{combined uncertainty, } \mu_c = \sqrt{\mu_{\text{repeat}}^2 + \mu_{\text{reference}}^2}$$

The calculation expanded uncertainty (U) of the particles size measurement is followed as

$$U = k \mu_c \quad (95\% \text{ Confidence level; Coverage factor, } k = 2)$$

The results of the 60nm polystyrene latex nanoparticle are shown in Table 3

**Table 3:** Result for 60nm polystyrene latex

Samples	Result
60nm (1 drop)	65 $\pm$ 5
60nm (5 drop)	65 $\pm$ 5
60nm (10 drop)	65 $\pm$ 5
60nm (15 drop)	65 $\pm$ 5
60nm (20 drop)	65 $\pm$ 5

The z-score, also known as standard score, is a measurement used in statistics [5,10]. It is the measurement of the number of standard deviations a specific number is above or below a mean. The formula to calculate z-score is

$$z = (x - \mu) / \sigma$$

where, z is the z-score, x is the value to be standardized,  $\mu$  is the mean of the given set of data, and  $\sigma$  is the standard deviation of the given set of data. Satisfactorily values of z-scores are from -2 to 2 (acceptable results). The values of z-scores between -2 and -3 and between 2 and 3 are doubtful (suspicious results), while the value of z-scores lower than -3 and greater than 3 are unsatisfactory (unacceptable results) [6-9]. Table 4 show the result for z-score. The results showed satisfactory based on the z-score.

**Table 4:** Results for z-score

Samples	Size (nm)	z-score
60nm (1 drop)	65.33	0.55
	65.34	0.57
	65.73	1.30
	64.83	-0.40
	65.51	0.89
60nm (5 drop)	65.30	0.49
	65.35	0.58
	64.83	-0.40
	64.20	-1.58
	65.21	0.32
60nm (10 drop)	65.93	1.68
	65.63	1.11
	65.51	0.89
	65.03	-0.02
	65.98	1.77
60nm (15 drop)	64.70	-0.64
	65.23	0.36
	64.42	-1.17
	64.92	-0.23
	64.49	-1.04
60nm (20 drop)	65.17	0.25
	64.49	-1.04
	64.34	-1.32
	64.14	-1.70
	64.50	-1.02

## 4. Conclusions

The hydrodynamic diameter measurement of 60 nm of polystyrene latex spheres has no changes while the concentration of the PLS is being increased from 1 drop up to 20 drops in 1ml of 10 mM NaCl. Therefore, it can be concluded the concentration of PLS

does not has to be taken in consideration for the calculation of measurement of uncertainty.

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