application note

Characterisation of Colloidal Gold Using Dynamic Light Scattering

Introduction

Colloidal gold is a dispersion of suspended nanoparticles of gold which exhibit interesting properties [1]. The colour of the sample is determined by the size and shape of the gold particles present [2]. Figure 1 shows colloidal gold suspensions of various particle sizes. Sizes less than 5nm are yellowish in colour, 5nm to 20nm tend to be reddish in colour whereas particles greater than 100nm are bluish in appearance.

Figure 1: Colloidal gold suspensions of various particle sizes. Photograph courtesy of Dr. Irawati Kandela, University of Wisconsin, BBPIC laboratory

Gold particles in aqueous media attain a negative charge which gives them a strong affinity for various biological macromolecules such as proteins and antibodies [3]. For this reason, colloidal gold is currently used in a variety of biotechnological applications such as DNA-conjugates, imaging probes and diagnostic agents [1,4,5]. In addition, colloidal gold suspensions are being developed for use in advanced electronics and coating applications [6].

Size characterization of colloidal gold is of great importance in order to ensure that the particles are homogenous in diameter and that there are no aggregates present in the dispersion. Size characterization is popularly done using electron microscopy techniques [1,2]. Figure 2 shows a transmission electron micrograph of a colloidal gold sample. Even though the individual gold particles are clearly visible, the majority of them exist as clumps consisting of 2 or more particles.

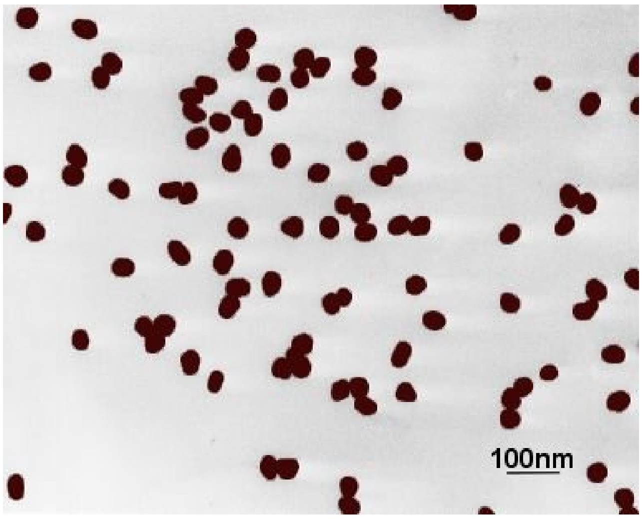

Figure 2: Electron micrograph of colloidal gold sample. Photograph courtesy of Dr. Michael Natan, Department of Chemistry, Penn State University

Even though electron microscopy is an excellent technique for the visualization of particles, it is poor from a statistical point of view as only tens or hundreds of particles are measured. The technique will give a number-based particle size as the numbers of different sized particles are counted.

Dynamic light scattering (DLS) is a non-invasive technique for measuring the size of nanoparticles in a dispersion. The technique measures the time-dependent fluctuations in the intensity of scattered light from a suspension of particles undergoing random, Brownian motion. Analysis of these intensity fluctuations allows for the determination of the diffusion coefficients, which in turn yield the particle size through the Stokes-Einstein equation.

This application note discusses the size characterization of colloidal gold using dynamic light scattering and highlights the differences in the results that might be obtained compared with electron microscopy techniques.

Experimental

All measurements reported in this application note were performed on a Zetasizer Nano S at 25°C. The Nano S contains a 4mW He-Ne laser operating at a wavelength of 633nm and an avalanche photodiode (APD) detector. The scattered light was detected at an angle of 173°.

Results and Discussion

Figure 3 shows the intensity particle size distribution obtained for a colloidal gold sample measured on a Nano S. The plot shows the relative percentage of light scattered by particles (on the Y-axis) in various size classes (on the X-axis). The size distribution obtained is a bimodal with peak means of 13.6 and 339nm respectively. A summary of the peak analysis of the distribution can be found in table 1.

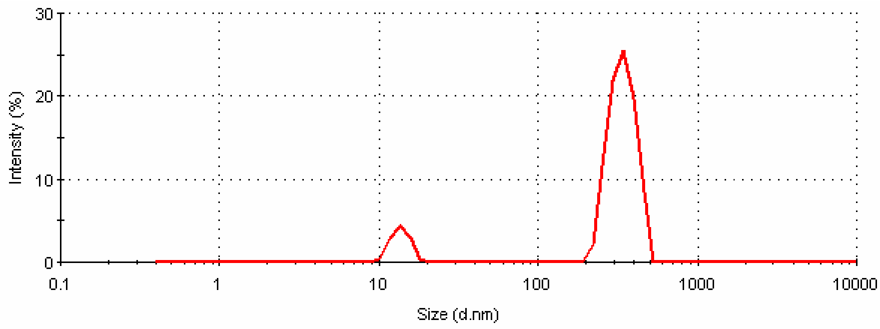

Figure 3: Intensity size distribution obtained for a colloidal gold sample

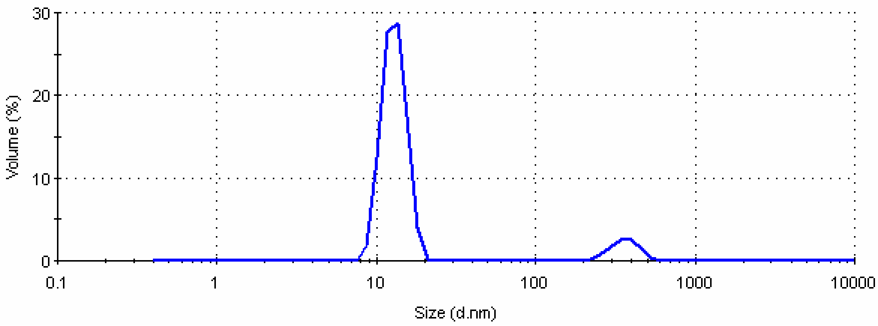

Figure 4: Volume size distribution obtained for a colloidal gold sample

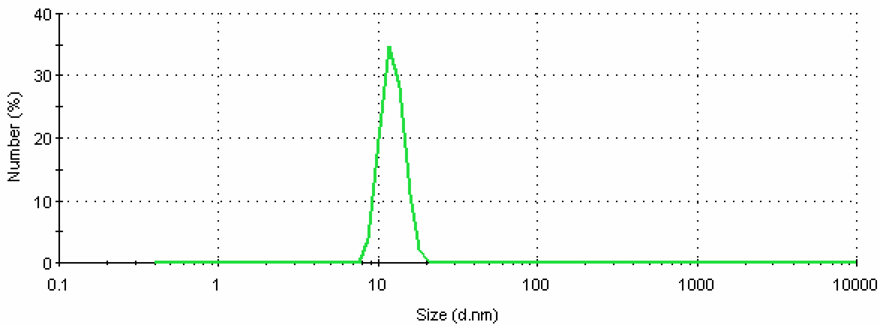

Figure 5: Number size distribution obtained for a colloidal gold sample

The intensity size distribution obtained implies that there are significant aggregates present in this sample. However, conversion into a volume (or mass or weight) based size distribution (figure 4) shows that the aggregates are present in low concentration. The transformation from the intensity data into volume is performed using Mie theory and this conversion requires the particle refractive index (n) and absorption (k) values. The optical properties used for the conversion from intensity to volume in this study were 0.2 (n) and 3.32 (k) respectively [9]. The volume size distribution obtained shows that, on a mass basis, over 90% of the sample consists of small particles around 13nm.

The result can be further converted into a number based size distribution which is shown in figure 5. This distribution is monomodal with a peak mean at 12.4nm. The result suggests that if this sample was characterized using a number based technique, such as electron microscopy, the vast majority of particles visible would be small ones. The presence of large particles would only be seen if sufficient numbers were counted. Even though, on a number basis, this sample contains very few aggregates, they scatter a significant amount of light contributing to the major peak in the intensity size distribution (figure 3). Therefore such a sample would be expected to give quite different results when analyzed by dynamic light scattering and electron microscopy.

On a further note, if the sample illustrated in figure 2 was to be measured by dynamic light scattering, it would not be possible to resolve different sized peaks for the various particle species i.e. single particles, aggregates of 2 particles, aggregates of 3 particles etc. Dynamic light scattering is a low resolution technique being able to resolve materials with a factor of 3 difference in their sizes.

Table 1: Peak analysis for the intensity, volume and number particle size distributions obtained for a colloidal gold sample

	Peak 1		Peak 2	
	Mean (nm)	**%**	**Mean (nm)**	**%**
Intensity	13.6	11	339	89
Volume	13.0	91	363	9
Number	12.4	100	-	-

So a mixture of single particles and aggregates made of 2, 3 or 4 particles would be expected to give a broad single peak with the result obtained being influenced by the larger particles present as they would scatter the majority of the light. The z-average diameter and polydispersity index values are sensitive to the presence of aggregates. The z-average diameter is the mean hydrodynamic diameter and the polydispersity index is an estimate of the width of the distribution. Both of these parameters are calculated according to the International Standard on dynamic light scattering, ISO13321 [10].

Conclusions

The technique of dynamic light scattering is ideally suited to the size determination of colloidal gold. It is very sensitive to the presence of aggregates of particles and the z-average diameter and polydispersity index values could be used as a way of determining sample homogeneity.

For monodisperse and monomodal samples, the results obtained from dynamic light scattering and electron microscopy should be very similar. However, for samples which are polydisperse, the results obtained from dynamic light scattering will be larger than those obtained from electron microscopy studies due to the contribution of scattering from the larger particles

References

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application note

Zetasizer Nano

The Zetasizer Nano system from Malvern Instruments is the first commercial instrument to include the hardware and software for combined static, dynamic, and electrophoretic light scattering measurements. The wide range of sample properties available for measurement with the Nano system include, particle size, molecular weight, and zeta potential.

The Zetasizer Nano system was specifically designed to meet the low concentration and sample volume requirements typically associated with pharmaceutical and biomolecular applications, along with the high concentration requirements for colloidal applications. Satisfying this unique mix of requirements was accomplished using a backscatter optical system and a novel cell chamber design. As a consequence of these features, the Zetasizer Nano specifications for sample size and concentration exceed those for any other commercially available dynamic light scattering instrument, with a size range of 0.6nm to 6um, and a concentration range of 0.1ppm to 40% w/v.

Complementing the patented hardware design is the Malvern software, providing instrument control and data analysis for the Zetasizer Nano System. The software uses self optimizing algorithms to automate the optical set-up required for each individual sample type, and includes a unique "one click" measure, analyze, and report feature designed to minimize the new user learning curve.

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