Nanosuspension characterization: Application of NTA to research from drug delivery to exosomes research

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Abstract
NanoParticle Tracking and Analysis (NTA), has been commercially developed over the past six years and now, with over 500 systems installed, is being considered a key characterisation technique in many fields of colloid characterisation particularly in the application of nanomedecines research and development. Here we discuss the technique, its application to nanobiomedecine and how recent developments are further enabling this field.

In this method a laser beam passes through a suspension at a low angle. The particles scatter light which is collected onto a CCD camera by a microscope-type configuration (Fig 1a). Particles, between 10-2000nm, are tracked individually (Fig 1b) and their diffusion coefficient and therefore size calculated directly from their speed (Fig 1c).

This technique gives significant advantage over traditional light scatter techniques (e.g. DLS) as the individual tracking of particles results in a better ability to cope with polydispersed suspension and results in higher resolution [1]. It also generates directly a measure of particle concentration which is critical in understanding many of the applications to which it is applied.

More recently, other parameters have been developed for further characterisation, such as scatter intensity (see figure 2) and zeta potential, all on a particle by particle basis. The technique can be integrated with fluorescence filters to allow fluorescently labelled/loaded particles to be selectively analysed. This allows for more complete characterisation of different nanoparticles within a single sample.

This technique has variously been applied to liposomes and other drug delivery particles, exosomes (with an aim towards a diagnostic measurement), viral vaccines and VLPs and in the field of protein aggregation and nanoparticle toxicology.

In the field of toxicology, the ability to measure nanoparticles and to assess the level of aggregation of nanosuspensions dispersed in various media is critical. NTA is appropriate for rapidly measuring samples as the optimal concentration for detection is very low (10^8 particles/mL) and such samples are commonly polydisperse and has been used extensively to this end [2]. The output, a number-based size distribution and absolute concentration measurements, directly provides the data in the appropriate format.

The technique is also used to directly generate a measure of viral concentration. In this regard, the ability of NTA to determine virus count through direct visualization, is of significant value [3]. It has advantage over both plaque assay (measurement times are of just a few minutes and aggregation can be assessed) and qPCR (as all viral particles will be measured whether or not they contain DNA). The application with fluorescence filters to allow fluorescently labelled/loaded particles to be selectively analysed can additionally be of particular import where the suspension is not purified.

In the field of pharmaceuticals, a crucial question under inspection is that of protein aggregation and its measurement thereof. In this field NanoSight has been identified as a technique suitable for characterizing this and has crucial benefits over previously available techniques[4].

Here we will present the technique and demonstrate, with examples, how it is a powerful, high resolution multi-parameter method with the capabilities to characterize and monitor particles in both water and biological environments for drug development and toxicological studies. Data will be shown demonstrating the accuracy and consistency of the technique both in ideal and non-ideal sample types.

References
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[3] Anderson, B., et al. Bacteriophage, Volume 1, Issue 2 (2011), 86-93.
[4] Carpenter, J. F., Randolph, T. W., Jiskoot, W., Crommelin, D. J., Middaugh, C. R. and Winter, G. Journal of Pharmaceutical Sciences, (2010), 99: 2200–2208.

Figures

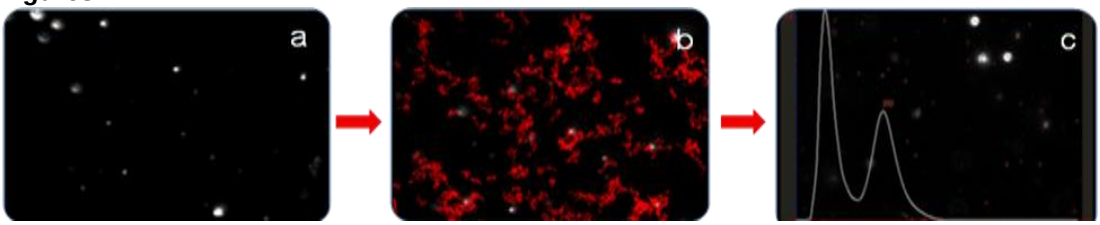

Figure 1: a) Scattered light from particles collected on CCD, b) partices tracked and c) size distribution calculated.

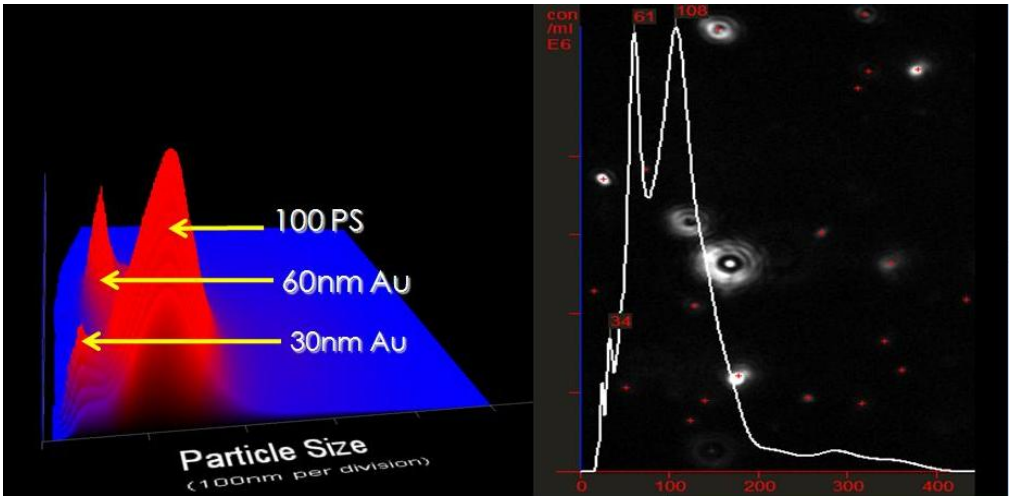

Figure 2: Trimodal measurement of a mixture of 30 and 60nm gold and 100nm latex polystyrene particles.