

Introduction

The Malvern NanoSight NS300 is an instrument used for the characterization of nanoparticles in solution by measuring particle size and concentration.

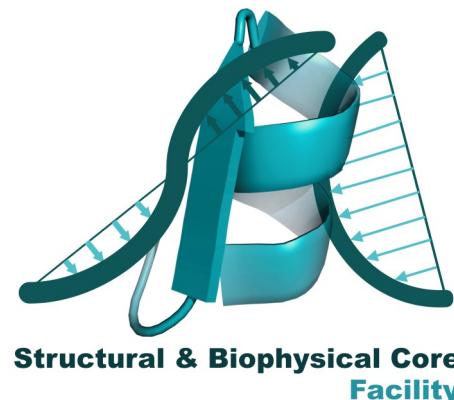
The NS300 uses Nanoparticle Tracking Analysis (NTA) to simultaneously analyze individual nanoparticles by direct observation of diffusion (Brownian motion). Sample particles can be visualized live as the experiment proceeds, providing visual validation of the results.

Applications

- Protein aggregation - sizing and measurement of protein polydispersity
- Drug delivery systems, polymer - based particles, lipid-based carriers, virus-like particles
- Vaccine and drug development - characterizing degradation and aggregation of therapeutic proteins
- Characterizing cellular vesicles - size and concentration of microvesicles, microsomes, exosomes

Features

- Automated analysis of the size distribution and concentration of nanoparticles from a diameter of 10 nm to 2000 nm in a liquid suspension.
- Particle light scattering, fluorescence and Brownian motion is video captured using a 20x magnification microscope and mounted sCMOS camera.
- Sample temperature controlled from 5 °C below ambient to 50 °C .
- Fluorescent labels can be analyzed using 488 nm (blue) and 532 nm (green) excitation lasers with 500 nm and 565 nm long pass emission filters.
- Syringe pump add-on allows for automated multiple analysis at a constant flow rate along with non-destructive sample recovery.



Greg Wasney

Manager, Structural & Biophysical Core Facility

Peter Gilgan Centre for Research & Learning
The Hospital for Sick Children
686 Bay Street, Rm. 21.9708
Toronto, ON. M5G 0A4

Email: greg.wasney@sickkids.ca

Office: 416-813-7209
Office Internal Ext. 307209
Lab: 416-813-7654 ext. 309442

<http://lab.research.sickkids.ca/sbc-facility/>

SickKids®
THE HOSPITAL FOR
SICK CHILDREN

The Hospital for Sick Children's Structural and Biophysical Core Facility



NanoSight - Malvern

NanoSight NS300—Light Scattering

- Measure nanoparticle diameter sizes from 10 nm to 2000 nm in liquid suspension using Brownian motion
- Characterize particle polydispersity, colloidal stability and quality
- sCMOS camera for video capture, particle tracking and concentration determination in solution
- Alternative fluorescence label analysis using blue 488 nm or green 532 nm green lasers.

NanoSight NS300—Nanoparticle Tracking Analysis

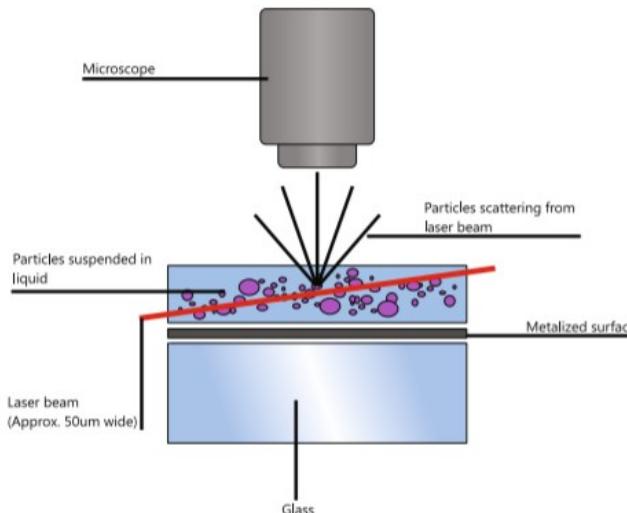
Theoretical background

Nanoparticle Tracking Analysis (NTA) is a method used for measuring particle sizes in liquids by relating the rate of Brownian motion to particle size, as well as the viscosity and temperature of the liquid.

Particles in the liquid are illuminated by a laser and scatter light that is recorded using a microscope mounted camera. The software tracks individual particles frame by frame and calculates particle size according to the following formula derived from the Stokes-Einstein equation:

$$\overline{(x,y)^2} = \frac{2k_B T}{3r_h \pi \eta}$$

Where k_B is the Boltzmann constant; $\overline{(x,y)^2}$ is the mean-squared speed of a particle at temperature T , in a medium of viscosity η , with a hydrodynamic radius of r_h .



Characterizing particle size distribution

Sample: Polystyrene bead standard (100 nm diameter), diluted to appropriate concentration in water.

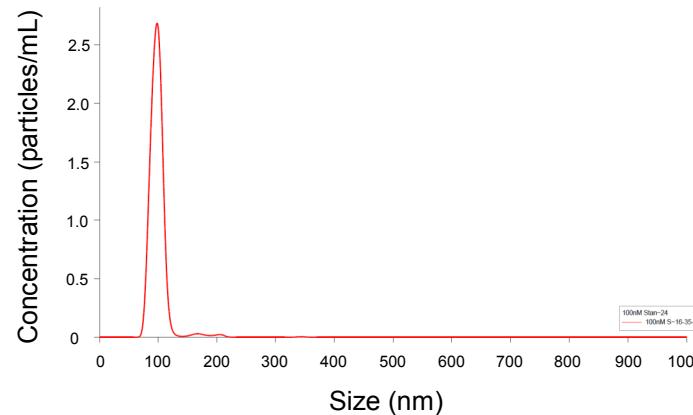
Laser type: Blue 488 nm **Temperature:** Ambient (24.0 °C) **Viscosity:** 0.9 cP (water)

Camera capture settings: sCMOS, 25.0 FPS, 730 second video capture

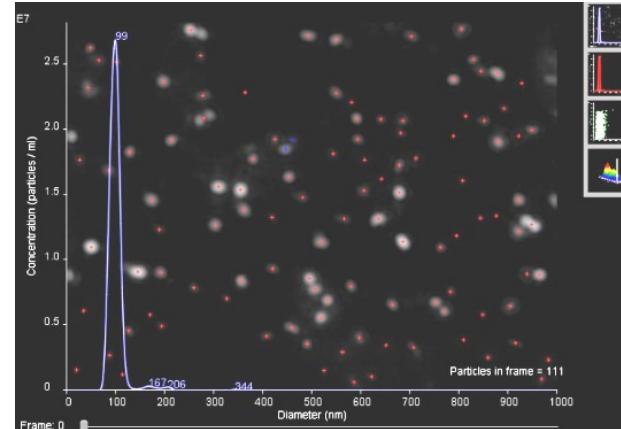
Syringe Pump: 22 μL / min (Relative infusion rate of 100 using a 1 mL syringe)

During the experiment, the sample was pumped by an automated syringe pump through the micro-flow cell where the light scattered from the particles were detected by the mounted digital camera.

Processed data plot of measured particle size and the concentration (number of particles).



During an experiment, the camera detects and records the particles moving through the flow cell. Analysis software tracks the individual particles seen.



Statistics	
Mean	100.3 nm
Mode	94.3 nm
SD	12.8 nm
D10	85.9 nm
D50	96.3 nm
D90	119.2 nm
Concentration	7.22e+008 particles / ml 99.9 particles / frame 87.8 centres / frame

Alternate view: plot of the concentration (number of particles) with changing intensity of the signal and diameter of the particles.

