Subpixel Localisation of Nanoparticles in Images

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Abstract

Nanoparticle Tracking Analysis (NTA) is an emerging technology for the quantification of particle size, concentration and zeta potential for particles in the size regime of 10 to 1000 nm. The technique allows the visualization of the Brownian motion of particles in liquid suspensions. It is frequently used in commercial and academic applications for the analysis of the physical and chemical properties of dispersions such as solubility, rheology and reactivity which are strongly influenced by the size of the respective particles. Hence, measuring the size of micro- or even nano-sized particles in dispersions plays a central role in chemical and biomedical industries.

With the NTA technique, particles dispersed in liquids are illuminated with an intensive light beam, e.g. from a laser. An image series of the light scattered by the particles is recorded with a sensitive digital camera with a magnification microscope attached to it. From the image series the Brownian motion of the particles is analyzed by first localizing the particle in each video frame, second tracking of the particles from frame to frame, and third computing the Mean Squared Displacement (MSD) along the track of each individual particle. Having the MSD one can estimate the particles diffusion coefficient and apply the Stoke-Einstein relationship to estimate the hydrodynamic size of individual particle. Current NTA systems use background segmentation method to differentiate the particles from background, mostly with fixed threshold approach. Fixed threshold works well for mono-modal dispersion since the brightness of the particles is evenly distributed. Poly-disperse particle solutions on the other hand show a high variation in the particle intensity because the reflected light intensity depends on the particle size and thus it is difficult to find a fixed threshold value.

We propose a new method for NTA which utilizes a multi-scale Laplacian of a Gaussian (LoG) detector on top of the background-subtraction model to localize the particles. Our approach uses an optimized thresholding method for each blob individually to compute a super-resolution position estimate. We show that our method finds more particles in the video with higher precision over the full size-range of tested solutions (20nm-500nm) in comparison to the fixed threshold approach. We further show that the increased efficiency in particle tracking and the higher precision in the localization of the particle center leads to particle size distributions that are narrower (having less variance). Thus, our method is in particular better suited for the analysis of mixtures of poly-disperse particle solutions if the size of the particles in the mixture solution is not too far apart.

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