MEASUREMENT OF ANGULAR SCATTERING DISTRIBUTIONS WITH DIGITAL FOURIER HOLOGRAPHY FOR MAPPING SCATTERER SIZE DISTRIBUTIONS OVER LARGE AREAS

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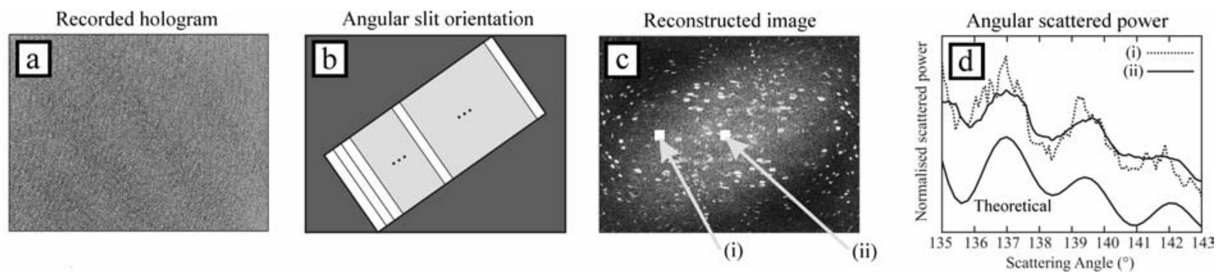
There are many instances in which the microscopic properties of large biological samples (with linear dimensions of up to many millimetres) are desired to be known. Such instances include the counting and assessment of individual cells in large populations, and the *in vivo* assessment of epithelial tumours. If conventional microscopic imaging techniques are used, the structures to be determined are resolved over small fields of view and stage scanning is required to cover the necessary area, a procedure that is slow and laborious. An alternative to conventional image formation is the deduction of the properties of the samples from elastic light scattering – so-called light scattering spectroscopy (LSS) [1,2]. In this class of methods, the frequency spectrum or angular distribution of scattered light is measured and a model (typically Mie theory) is used to deduce local scatterer size distributions and/or relative refractive indices. Such methods have been demonstrated in microscopic format [3], but importantly they have also been demonstrated in macro-imaging [1] formats, in which it has been shown that it is not necessary to resolve the scatterers in order to deduce their properties. In this work, we extend this approach through the use of digital Fourier holography to record full amplitude and phase of scattered waves versus angle for large area samples. The holographic approach provides several key advantages over other methods demonstrated to date. It requires only a single exposure, which is a major advance over particularly LSS imaging methods that require an exposure per spectral or angular component, it conveys a heterodyne sensitivity advantage, its requirement for coherence reduces the contributions from unwanted multiply scattered light, and its coherent registration provides the opportunity to digitally Fourier filter the recorded signal, enabling great flexibility (in the application of a variety of digital amplitude and phase Fourier filters to select, image, and measure desired properties of the sample, for example, scatterer size distributions and relative refractive indices).

To demonstrate the approach, we measured particle size distributions for 1.5×3.0-mm phantoms constructed from polystyrene microspheres in water. Applying the angular slit masks in part (b) of the figure to the recorded hologram in part (a), the scattered power over a range of scattering angles can be recorded for any region of the sample of 10-μm spheres. That corresponding to regions (i) and (ii) in part (c) is compared with theory in part (d).



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