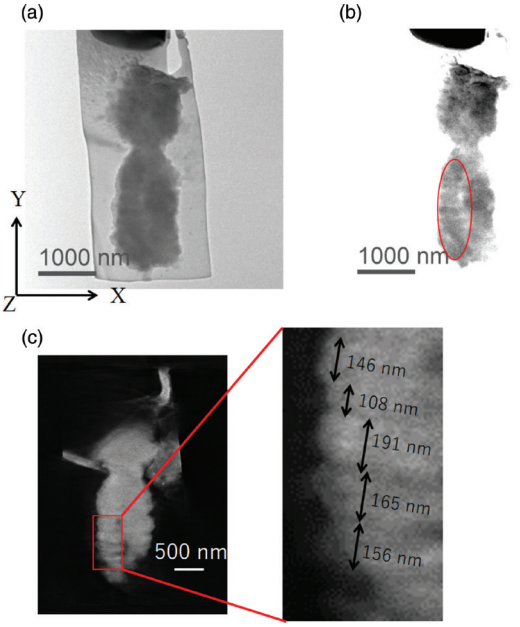
Highlights from Microscopy AND Microanalysis

Biological Applications

Higher-order Structure of Human Chromosomes Observed by Electron Diffraction and Electron Tomography by M Hayashida, R Phengchat, M Malac, K Harada, T Akashi, N Ohmido, and K Fukui, *Microsc Microanal* | doi:10.1017/S1431927620024666

It is well known that two DNA molecules are wrapped around histone octamers and are folded together to form a single chromosome. However, the nucleosome fiber folding within a chromosome remains an enigma, and the higher-order structure of chromosomes is not understood. We employed electron diffraction (ED) with a very high camera length to non-invasively analyze the internal structure of chromosomes. By switching between imaging and diffraction modes, the relationship between the direction of chromosomes and periodic features was observed. Results revealed the presence of structures with 100 to 200 nm periodic features perpendicular to the chromosome axis in unlabeled isolated human chromosomes. We also visualized the 100 to 200 nm periodic features perpendicular to the chromosome axis in an isolated chromosome whose DNA molecules were specifically labelled with OsO_4 using electron tomography (Figure).

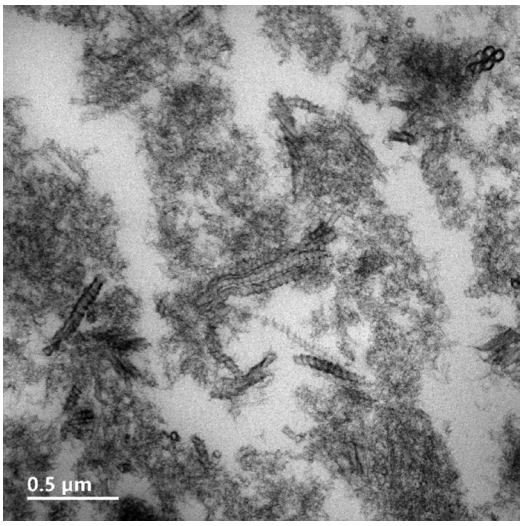


(a) A TEM image of a chromosome labelled with OsO_4. (b) Contrast enhanced image of (a). (c) Z-slice images from the tomogram of the chromosome.

Materials Applications

Microscopy of Polyurea Grease by MA Thorseth, JD Harris, J Gu, J Cuthbert, L Huffman, K Capaldo, and Z Jia, *Microsc Microanal* | doi: 10.1017/S1431927620024794

Understanding the phase structure of lubricating grease thickener in the base oil phase can provide critical insight into improving the grease lubricating performance. However, observation of the intact thickener microstructure has historically proven to be difficult. Previously reported observations of grease structures were performed by scanning electron microscopy (SEM) after the removal of the oil phase, which may alter the microstructure, or by freeze-fracture SEM, which results in poor contrast between phases. Direct transmission electron microscopy (TEM) imaging of thin slices of grease prepared by cryo-microtomy has rarely been reported, likely due to the difficult sample preparation. This article compares various sample preparation techniques of polyurea greases, the most prevalent non-soap grease variety, for microscopic observation. A technique is demonstrated to successfully collect and stain thin sections of grease for examination by TEM, enabling observation of unique microstructures of the grease thickener. Most of the thickener is present in clusters of tangled fibers (Figure), but other conformations similar to those observed in block copolymers were observed by TEM tomography.



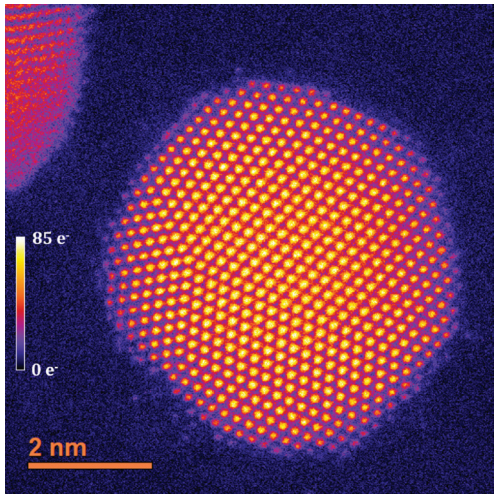
Bright-field TEM image of the microstructure of polyurea grease after staining with RuO_4 vapors.

Microscopy AND Microanalysis

Techniques Development

Development of a Practicable Digital Pulse Read-Out for Dark-Field STEM by T Mullarkey, C Downing, and L Jones, *Microsc Microanal* | doi:10.1017/S1431927620024721

With beam damage becoming the resolution-limiting factor for many technologically relevant samples, there is an ever-growing interest in low-dose scanning transmission electron microscopy (STEM). Lowering both the beam-current and pixel dwell-time is an appealing route to follow as these are available to all operators. Unfortunately, common annular dark-field (ADF) detectors begin to produce unacceptable image artefacts under these conditions (streaks). Fortunately, these conditions also reveal single electron impacts in the detector's output, which we now exploit with our new technique. Using a USB-powered streaming oscilloscope, we pass the ADF detector output to a laptop where it is analyzed using MATLAB. Crucially, this analysis converts the analog output of the detector to a digital signal where each '1' represents a single electron impact. By registering *only* electron impacts as signal, and each with equal intensity, we remove these erroneous signals, eliminate background noise, and circumvent any inhomogeneity in the ADF detector. This new approach to STEM imaging produces clean, high signal-to-noise ratio low-dose digital images where every scattered electron counts (Figure).



This first-ever digital image of gold nanoparticles on carbon is the result of rigidly aligning and summing 20 low-dose digital HAADF image frames. Each was captured with a beam current of ~5 pA and a dwell-time of 2 µs. The colorbar has units of integer electron impacts per pixel.