Three-dimensional imaging by optical sectioning in the aberration-corrected scanning transmission electron microscope

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The depth resolution for optical sectioning in the scanning transmission electron microscope is measured using the results of optical sectioning experiments of laterally extended objects. We show that the depth resolution depends on the numerical aperture of the objective lens as expected. We also find, however, that the depth resolution depends on the lateral extent of the object that is being imaged owing to a missing cone of information in the transfer function. We find that deconvolution methods generally have limited usefulness in this case, but that three-dimensional information can still be obtained with the aid of prior information for specific samples such as those consisting of supported nanoparticles. We go on to review how a confocal geometry may improve the depth resolution for extended objects. Finally, we present a review of recent work exploring the effect of dynamical diffraction in zone-axis-aligned crystals on the optical sectioning process.

Keywords: aberration correction; scanning transmission electron microscopy; depth sectioning

1. Introduction

The successful implementation of spherical aberration correctors in the scanning transmission electron microscope (STEM) has allowed an increase in the semiangle of convergence, $\alpha$, of the probe-forming aperture. Not only does this improve the attainable lateral resolution (Nellist et al. 2004), which is inversely proportional to $\alpha$, but also reduces the depth of field, which is inversely proportional to $\alpha^2$. This fact has long been known in the field of light optics (Born & Wolf 1999). Depth of field is the distance parallel to the optic axis over which the sample is in focus. In an aberration-corrected STEM, this distance may only be several nanometres and smaller than the typical thickness of a sample. Clearly the usual interpretation of electron micrographs in terms of the projected structure is no longer possible and a more careful analysis is required. This reduction in depth of field, however, does create the opportunity to retrieve three-dimensional information about the sample. It is possible to optically section a sample in a similar way to confocal scanning optical microscopy (Corle & Kino 1996) and produce a three-dimensional representation of our sample.

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The motivation behind three-dimensional imaging in the electron microscope is to fully understand the three-dimensional structure of materials that determines their chemical, physical and electronic properties. At present, the most common technique for three-dimensional imaging is electron tomography (Midgley & Weyland 2003), and while the volume resolution of this technique is very good (approx. 1 nm³), data acquisition and processing times are long. Another established technique is atom probe tomography (Miller 2000), which can provide atomic layer resolution in the depth direction, but is destructive and cannot be easily applied to samples that are non-conductive.

While STEM optical sectioning is not expected to compete with either of these techniques in terms of resolution, data acquisition is much quicker and recording of the focal series is easily automated. Furthermore, only specific depths of interest need to be probed. Optical sectioning has been demonstrated in previous work for single Hf atoms in SiO₂ (van Benthem et al. 2005) and for small Pt and Au nanoparticles in a TiO₂ powder (Borisevich et al. 2006). The positions of single atoms were located in three dimensions by van Benthem et al. (2005) with an error of less than a nanometre, while Borisevich et al. (2006) showed that the reconstructed shapes of large particles are elongated along the optic axis. In this paper, we present results from experimental STEM optical sectioning of samples containing Pt and Au nanoparticles of varying size. Such nanoparticles present an excellent test sample with which to investigate the resolution limits in STEM optical sectioning and to test the effectiveness of image processing and deconvolution methods applied to the data. We find that the contribution of out-of-focus planes reduces the three-dimensional resolution for laterally extended objects, and we review how a confocal geometry can help restore the resolution for these objects. Finally, we review recent work exploring how dynamical diffraction effects in zone-axis-aligned crystals affect the optical sectioning process.

2. Optical sectioning in scanning transmission electron microscopy

The three-dimensional optical sectioning data can be acquired by recording a series of images of the sample at different defocus values. Incoherent imaging modes, such as annular dark-field (ADF) STEM, have a point-spread function (PSF) that is given by the intensity of the illuminating probe. The depth of field of such an imaging mode is defined by Born and Wolf as \( \lambda/\alpha^2 \) (Born & Wolf 1999). While the full width at half maximum (FWHM) of the PSF in the depth direction is given by (Nellist et al. 2008)

\[
\Delta z_{\text{fwhm}} = 1.7 \frac{\lambda}{\alpha^2},
\]

where \( \lambda \) is the wavelength of the electrons. Our experiments were performed on the Oxford-JEOL 2200MCO with an accelerating voltage of 200 kV (\( \lambda = 0.0025 \text{ nm} \)) and \( \alpha \) of 22 mrad, giving a depth of field of 5.1 nm. Figure 1 shows the \( r-z \) plane containing the optic axis, displaying the intensity of the three-dimensional STEM probe in vacuum. Figure 2 shows a line plot along the \( z \)-axis of the probe intensity showing the 8.8 nm FWHM. All aberrations with the exception of defocus were assumed to be zero.
Three-dimensional imaging

7.0 nm

300 nm

Figure 1. The r–z plane containing the optic axis of the three-dimensional STEM probe (log intensity) with the parameters of the Oxford-JEOL 2200MCO (200 kV and 22 mrad semiangle of convergence). All aberrations with the exception of defocus were assumed to be zero. The lateral width of the probe is approximately 0.1 nm.

Figure 2. Plot of the intensity of the STEM probe along the z-axis of the three-dimensional STEM probe in vacuum. The FWHM in the z-direction is measured to be 8.8 nm. An increase in either accelerating voltage or semiangle of convergence can be used to reduce this width further. \( V_0 = 200 \text{ kV}; \alpha = 22 \text{ mrad.} \)

For an initial test experiment, we recorded a focal series of ADF images of Pt nanoparticles on a powdered carbon support. The particles and the support were dispersed onto an amorphous carbon film. The focal series consisted of 200 images using a focal step size of 4.0 nm. Two images from the series are shown in figure 3, which show that three-dimensional information about the sample has been recorded. If we examine the nanoparticles on the left half of figure 3a, we...
Figure 3. Two images taken from the focal series of Pt particles on a carbon support. We can see that particles come into focus at different values of defocus. For example: examine the particles on the left extreme of (a) and compare the same area in (b) and also with the other particles in the field of view. From this, we can infer that they are at different heights on the support. (a) $\Delta f = 0\text{nm};$ (b) $\Delta f = -112\text{nm}$.

can see that they appear out of focus, while in figure 3b they now appear to be in focus compared with the other particles. From this, we can infer that these particles are at different heights on the support.

To counter the effects of sample drift, each image in the focal series was aligned using a cross correlation with an appropriate reference image. The choice of this reference image is important to ensure proper image registration. It can be chosen by eye as the sharpest image or by some other criterion such as the presence of strong spots in the power spectrum arising from lattice fringes in the image. For the data presented here, the image judged to have most of the particles in or near focus was selected. Nearest neighbour cross correlation was tried but produced unsatisfactory results as the error between each image accumulates and becomes unacceptably large over the entire series.

This reference image is also used to generate a mask for each particle. We are assisted here by the type of sample as it contains nanoparticles that tended not to be clustered together, while the carbon support scatters weakly in comparison. The mean intensity of the reference image is an adequate threshold to separate the particles from the background intensity. An erosion operation (Gonzalez & Woods 2002) is performed to separate any overlapping masks and to remove noise. This is followed by a dilation (Gonzalez & Woods 2002), which ensures that each mask is approximately the right shape and size of each nanoparticle. At this point, we can also estimate the size of each of the particles. Two particles were selected from the Pt dataset (figure 4a) and their integrated intensity measured as a function of defocus (figure 4b).

Immediately striking in figure 4b is the width of the intensity peaks. The intensity profile of the 8.3 nm particle shows an FWHM of 470 nm and the 3.8 nm diameter particle shows an FWHM of 240 nm. These values are much larger than the 8.8 nm FWHM of the probe in the axial direction. We also note that the axial resolution, which we define here as the FWHM of the plot, is smaller for the particle with the smaller diameter.
Three-dimensional imaging

Figure 4. The integrated intensity of particles of varying sizes from the Pt dataset. (a) Pt particles on a carbon support. Two particles of size 8.3 (square) and 3.8 nm (circle) were selected. (b) Integrated intensity of both Pt particles as a function of defocus. The intensity from each particle is significant over a width of hundreds of nanometres. How wide this range is depends on both the size of the particle and also the semiangle of convergence. Solid line, 8.3 nm particle; dotted line, 3.8 nm particle.

(a) Three-dimensional transfer and resolution in incoherent scanning transmission electron microscope imaging

An ADF image is an example of incoherent imaging (e.g. Nellist & Pennycook 1999, 2000) where the recorded intensity from a weak scatterer can be described by a convolution between the PSF, \(|p(r, z)|^2\), and an object function, \(O(r, z)\),

\[
I(r, z) = |p(r, z)|^2 \otimes O(r, z) = P(r, z) \otimes O(r, z),
\]

where the convolution is over both the two-dimensional vector \(r\) representing the position perpendicular to the optic axis, and the coordinate parallel to the optic axis, \(z\). In equation (2.2), we have used an uppercase \(P\) to refer to the probe intensity distribution, the PSF. The form of this distribution is shown in figure 1. Note that the convolution over the \(z\) coordinate is only strictly valid if the form of the probe is unchanged by scattering within the sample. In the case of isolated nanoparticles on a light support, this is a reasonable approximation.

Using the convolution theorem (Bracewell 2000), the Fourier transform of equation (2.2) becomes the multiplication

\[
\tilde{I}(r^*, z^*) = \tilde{P}(r^*, z^*) \tilde{O}(r^*, z^*),
\]

where \(r^*\) and \(z^*\) are the transverse and axial coordinates in reciprocal space, respectively. The function \(\tilde{P}(r^*, z^*)\) is the optical transfer function (OTF) of the electron microscope. For the symmetric probe, such as the case assumed here, the transfer function is completely real. Figure 5 shows the \(r^*-z^*\) plane of the OTF that contains the optic axis.

An important feature of this plot is the large missing cone region where there is no transfer of the object’s spatial frequencies. As will be seen in §2b, this missing cone region has a dramatic effect on the depth resolution of objects that are...
Figure 5. The OTF for a STEM with the parameters of the Oxford-JEOL 2200MCO (200kV and 22 mrad) showing the missing cone region. The size of this region is controlled by the size of the semiangle of convergence. It is this missing cone region that is responsible for the severe elongation of extended objects in the STEM. To emphasize the bounds of the transfer, the contrast in the greyscale has been enhanced.

composed of mainly low lateral spatial frequencies. As indicated in figure 5, the size of this region is controlled by the choice of $\alpha$, the semiangle of convergence. However, even in the presence of a spherical aberration corrector, $\alpha$ is still of the order of tens of milliradians, much smaller than the value for $\alpha$ which can be used in optical microscopy (Corle & Kino 1996). As a consequence of this, many of the spatial frequencies that make up the object are not transferred (Frieden 1967; Streibl 1985; D’Alfonso et al. 2008; Intaraprasong et al. 2008).

(b) Imaging of extended objects

It is often the case that the sample under investigation is composed of objects that are larger than the probe in the transverse direction. We will refer to such objects as extended objects. A point-like object (i.e. an atom) will have Fourier components extending out to high transverse spatial frequencies, whereas extended objects tend to be characterized by mainly low spatial frequencies. As can be seen from figure 5, if only low transverse spatial frequencies of the OTF are explored, then along the optic axis the transfer of the object is severely bandwidth limited, restricting the longitudinal $z$ resolution. It is this loss of resolution that explains the extended intensity profiles seen in figure 4b.

At low transverse spatial frequencies, figure 5 shows that for a maximum transverse spatial frequency, $r_{\text{max}}^*$, present in the object function, the maximum longitudinal spatial frequency transferred by the microscope is given by $\alpha r_{\text{max}}^*$. It therefore follows that for an object with a characteristic transverse length scale, $d$, the longitudinal resolution will be given by

$$\Delta z = \frac{d}{\alpha}. \quad (2.4)$$

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Figure 6. Schematic of the geometry of the STEM probe illuminating a nanoparticle. Experimentally, we see that the intensity of the image of the particle only drops noticeably when the size of the probe becomes comparable with the diameter of the particle (figure 4b). From simple geometry, we can see that this leads to a depth resolution of approximately \( d/\alpha \).

Figure 7. An MI projection through the raw dataset recorded from a sample of Pt particles on a powdered carbon support. The particles are severely elongated parallel to the optic axis, and this makes localizing the particle in three dimensions difficult. The focal series consisted of 200 images, with a focal step size of 4.0 nm.

This result can also be derived by applying the three-dimensional sampling theorem to the case of incoherent imaging (Nugent 1988).

If we consider figure 6, we can show from a simple geometrical viewpoint that the scattered intensity from an extended object only has a noticeable decrease when the probe diameter becomes comparable to the size of the particle. This simple geometric argument also derives the expression in equation (2.4).

We can apply equation (2.4) to the nanoparticles shown in figure 4a. The particle of diameter 8.3 nm should show a longitudinal resolution of 337 nm, and the particle of diameter 3.8 nm, a longitudinal resolution of 172 nm, which is reasonably commensurate with the intensity depth plots in figure 4b.

For our initial test specimen, the Pt nanoparticles, we can see from a maximum intensity (MI) projection, figure 7, that the elongation is quite large. An MI projection displays the most intense voxel parallel to the direction in which the volume is being projected. Although the nanoparticles are distributed at different depths on the support, their locations parallel to the optic axis are obscured by the elongation. There is, therefore, a requirement to reduce this elongation if we wish to accurately determine the position of each nanoparticle.
(c) Deconvolution of experimental images

Given the somewhat complicated three-dimensional form of the PSF and the OTF, it is tempting to explore deconvolution strategies in an attempt to improve the resolution in the longitudinal direction. In light optics, there exists a large body of work concerning the deconvolution of the blurring caused by imaging systems in three dimensions for both confocal and widefield microscopes (McNally et al. 1999; Conchello & Lichtman 2005). At present, confocal imaging is the most common method owing to its optical sectioning capabilities. However, in electron microscopy, we generally use a single objective lens and each focal plane will contain some signal from every other focal plane, and we intend to remove this contribution by a deconvolution method.

In its simplest form, a deconvolution can be performed by dividing equation (2.2) by $\tilde{P}(r^*, z^*)$ and performing an inverse Fourier transform. However, in the presence of noise, the small values of $\tilde{P}(r^*, z^*)$ at high spatial frequencies will amplify the noise in the dataset and produce an unsatisfactory reconstruction. The Richardson–Lucy (RL) algorithm (Richardson 1972; Lucy 1974) has been used successfully by other groups to improve the resolution of two-dimensional images, including, famously, images from the Hubble telescope (Adorf 1995). This algorithm is as follows:

$$O_{n+1}(r, z) = O_n(r, z) \left[ \frac{I(r, z)}{P(r, z) \otimes O(r, z)} \otimes P(r, z) \right] ,$$

where $O_n$ is the $n$th estimated object function. The initial estimate $O_0$ is set equal to the recorded dataset, $I(r, z)$. Each deconvolution was performed on a 2.8 GHz desktop PC with 1 GB of RAM and ran for 100 iterations or until convergence. The parameter

$$\epsilon_n = \frac{\sum |I(r, z) - (P(r, z) \otimes O_n(r, z))|}{\sum I(r, z)}$$

(2.6)

can be used as a measurement of convergence. When the difference between successive values of $\epsilon$ falls below a threshold of $10^{-7}$, the floating-point precision of the computer, the algorithm can be taken to be converged. It is sometimes preferable to stop the algorithm before it converges, especially with a noisy dataset, as the algorithm makes no distinction between noise and signal, and the output may have spurious features present owing to noise. Some trial and error may be necessary to determine an optimum stop criterion for each imaging condition. To reduce calculation time and memory requirements, only a $128 \times 128 \times 100$ pixel subvolume from the whole $512 \times 512 \times 100$ pixel dataset was used. The calculation took approximately 45 min for each dataset.

To test the effectiveness of deconvolution methods, we recorded an ADF STEM focal series from a sample of gold nanoparticles supported on a carbon film, but with the film intentionally inclined at $20^\circ$. In a lateral MI projection of the data (figure 8a) along a direction perpendicular to the tilt direction, the strong elongation of the particles obscured the sample tilt. From figure 8b, we can see that the RL algorithm outputs a volume that has a reduced but still significant amount of blurring along the optic axis. The longer the algorithm runs, the more likely it is that artefacts will appear in each image plane as the higher spatial frequencies that mainly contain information about edges and noise can be
Figure 8. MI projection through Au particles on amorphous carbon that have been tilted 20°. (a) Raw dataset. Clearly the particles are elongated by several hundreds of nanometres along the optic axis. (b) Dataset after 100 iterations using the RL algorithm. While there is some reduction in the blurring, it is still significant.

<table>
<thead>
<tr>
<th>10 nm simulated particle number of iterations</th>
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Figure 9. A simulated spherical particle, 10 nm in diameter. The images were generated by convolving the object with the probe as per equation (2.2). The series consisted of 100 images with a focal step size of 6 nm. (a) Projections through the centre of the raw dataset. (b) Projections through the centre of the dataset after 20 iterations using the RL algorithm (equation (2.5)); note the slight reduction in the elongation, but the presence of an artefact in the xz plane. (c) Projections through the dataset after 45 iterations (the point where the algorithm converged).

Some of the information in the missing cone region can be partially recovered by the RL algorithm owing to the fact that it imposes the condition that the output must contain positive values (Holmes 1988). However, the rather large missing cone region in the OTF combined with the presence of noise in any real dataset makes estimating spatial frequencies in this region problematic and the reconstructions tend to have artefacts present while still retaining the elongation.
of objects parallel to the optic axis. We therefore conclude that the RL algorithm and similar algorithms are only of limited use for the reduction of the elongation of objects in STEM optical sectioning data.

\((d)\) Using prior information

Many deconvolution methods do attempt to reconstruct missing information by making use of prior information. Bayesian methods such as maximum entropy have been applied to electron microscope data (Nellist & Pennycook 1998) and allow a prior probability distribution to be included. In the case of our experiments, we have made several assumptions about the sample. First, we assume that each particle is approximately spherical, consistent with their cross sections in the images. While a real Au or Pt particle is likely to be faceted (Marks 1994), our goal is only to approximate the shape to make our reconstruction possible. Our second assumption is that the particle is located at the depth where its edge is the sharpest. Using these two constraints, we now attempt to localize the particles in our sample.

The sharpness of edges within each image can be measured using the Sobel operator (Gonzalez & Woods 2002). This involves two \(3 \times 3\) matrices (equation (2.7)) that are convolved with each image, \(I_n\), in the focal series. Edge detection enhances the higher spatial frequencies in each image and so can be used to determine whether a particle is in focus or not. From figures 9 and 10, we see that RL deconvolution has a similar effect. Performing edge detection, however, is much quicker. We only deal with one image at a time and remove the need to perform any Fourier transforms which reduces the memory and time requirements of the reconstruction. The data processing takes just a few minutes.

\[
S_x = \begin{bmatrix} 1 & 0 & -1 \\ 2 & 0 & -2 \\ 1 & 0 & -1 \end{bmatrix} \otimes I_n \quad \text{and} \quad S_y = \begin{bmatrix} 1 & 2 & 1 \\ 0 & 0 & 0 \\ -1 & -2 & -1 \end{bmatrix} \otimes I_n. \quad (2.7)
\]
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Figure 11. An MI projection through the dataset processed using prior information from a sample consisting of Au particles on an amorphous carbon film and tilted by 20°. The sharpness of each particle combined with a peak-fitting routine was used to determine its depth. Each particle was assumed to be spherical.

Figure 12. An MI projection through the dataset processed using prior information from a sample consisting of Pt particles on a powdered carbon support. Again, the sharpness of each particle combined with a peak-fitting routine was used to determine its depth. Each particle was assumed to be spherical.

The Sobel operator estimates the image gradient at each pixel point, the magnitude of which is \( S = \sqrt{S_x^2 + S_y^2} \), and so each pixel has a measure of sharpness. Again, it is only the pixels inside the mask used for the summation in §2 that are used. A sum of all the pixels within this mask is performed for each slice in the \( z \)-direction after the dataset has been operated on by the Sobel filter.

Evidently, from figure 10, the use of the Sobel operator can reduce the FWHM of the intensity curve, making the estimate of our particle depth more precise. However, the FWHM is still much greater than the particle size. The next step of our processing method is to locate the height of the particle by peak fitting to the plots created with the Sobel filter in figure 10 to a Gaussian.

Figures 11 and 12 show MI projections through the tilted Au on C and the Pt catalyst on C datasets, respectively, after processing with the Sobel operator and fitting the sharpness curve to a Gaussian. The sample tilt can be observed.
in figure 11, and we can measure the height difference between the top and the bottom of the sample plane as approximately 40nm. The distribution of the particles in the Pt catalyst dataset (figure 12) is more random, as we might expect in a catalyst sample dispersed on a powdered support.

3. Scanning confocal electron microscopy

The background contribution caused by out-of-focus laterally extended objects when optical sectioning was an important motivation in the development of the confocal optical microscope (Minsky 1988). There has been recent work in developing confocal microscopy using electrons (Frigo et al. 2002; Zaluzec 2003; Nellist et al. 2006; Takeguchi et al. 2008), and in our laboratory we have been investigating the prospects for aberration-corrected scanning confocal electron microscopy (SCEM). In this section, we will review the recent theoretical work modelling SCEM imaging.

A schematic diagram of the confocal configuration is given in figure 13. The dashed lines indicate that scattering from points in the sample away from the confocal point will not be accurately focused at a selecting aperture, which by analogy with light optics we will refer to as the pinhole, in the detector plane. The selection by the pinhole increases the depth resolution and reduces the background contribution from out-of-focus layers.

Figure 13. A schematic diagram showing the confocal configuration. Both the pre- and post-specimen lenses are aberration corrected. The confocal plane is the plane where both lenses appear to be focused. The benefit of using the confocal configuration is that electrons scattered outside of the confocal plane are rejected by the pinhole aperture. Solid line, electrons scattered from confocal plane; dashed line, electrons scattered from outside confocal plane.
To make use of the dramatic reduction in depth of field allowed by spherical aberration correction, an instrument fitted with aberration correctors both before and after the sample is required. While still somewhat rare, such double aberration-corrected instruments are currently increasing in number. Careful alignment of both aberration correctors about a mutual axis is required, and a method to do this has been developed (Nellist et al. 2006). The SCEM detector plane is actually the image plane in TEM because what we are essentially doing in the SCEM mode is using an aberration-corrected TEM to image the probe formed by an aberration-corrected STEM. Figure 14 shows the image of the probe that is seen in the absence of a sample, and the pixels inside the circle are summed to replicate the effects of a pinhole aperture. In practice, we typically work with a post-sample magnification of two million times, so that a pinhole size of 200 μm in diameter corresponds to a size of 0.1 nm at the sample. The SCEM configuration is also a very useful diagnostic tool as it can reveal information about instrumental instabilities that are not related to the sample stability. Instabilities and drift of the electron source have been observed in our SCEM datasets, and so the pinhole position must be tracked with the probe position.

In the case of incoherent imaging, it can be shown (Nellist et al. 2008) that the PSF for SCEM is given by the product of two probe intensities. One is the actual illuminating probe intensity, \( P_1(r, z) \), and the other probe intensity, \( P_2(r, z) \), is the intensity that would be formed at the sample if the detector plane pinhole was replaced by an electron source, and the optics between the detector plane and the sample used to focus a probe from this direction. The SCEM imaging process can then be described by the following equation:

\[
I(r, z) = (|P_1(r, z)|^2|P_2(r, -z)|^2) \otimes V'(r, z). \tag{3.1}
\]

Here, \( V'(r, z) \) is an incoherent scattering potential of the sample. The OTF for SCEM imaging is therefore given by the Fourier transform of the product of probe intensities given in equation (3.1). If we assume a microscope fitted with
Figure 15. The OTF of the SCEM. It has been assumed that $P_1(r, z) = P_2(r, z)$ and that the imaging is entirely incoherent as from equation (3.1). This transfer function lacks a missing cone region and therefore we might expect an improvement in the depth resolution over STEM optical sectioning. $V_0 = 200\, \text{kV}; \alpha = 22\, \text{mrad}$.

A symmetric condenser-objective lens and identical aberration correctors, then $P_1(r, z) = P_2(r, z)$ and our SCEM probe function is the square of the STEM probe intensity. The SCEM OTF is therefore given by the autocorrelation function of the STEM OTF (see figure 5 and §2a). This SCEM OTF is shown in figure 15, and comparing with figure 5 we can see that it lacks a missing cone region.

The absence of a missing cone for incoherent SCEM imaging clearly has important implications for three-dimensional optical sectioning of laterally extended objects. Not only should the depth resolution for the nanoparticles discussed in §2a be dramatically improved, but a whole range of potential applications could be imaged, for example, the depth profiling of thin-film layers in devices or other such extended objects.

As there is no equivalent of ADF imaging in SCEM, the incoherent model described above only applies when the scattering is intrinsically incoherent. If elastic scattering is detected, which we refer to as bright-field SCEM (BFSCEM), this theory is not applicable. Indeed a detailed analysis (Cosgriff et al. 2008) shows that in the single-scattering approximation, in which the sample can be treated as a weak-phase object, there will be very little contrast. The BFSCEM imaging mode relies on multiple scattering within the sample to generate contrast, and therefore relatively thick samples must be used. Under these conditions, we expect to see relatively complicated contrast and the interpretation of the data will not be straightforward and will require matching to a simulated image.

If the double aberration-corrected instrument is fitted with an energy filter, then inelastically scattered electrons may be collected, a technique that we refer to as energy-filtered SCEM (EFSCEM). Inelastic scattering is intrinsically partially coherent, and so the incoherent model above is only an approximation. Detailed simulations (D’Alfonso et al. 2008) show that the effects of the partial coherence
are observable in the EFSCMEM data, but that improvements in depth resolution are seen. Experiments investigating both the elastic and inelastic SCEM modes are currently underway in our laboratory.

4. Optical sectioning under dynamical diffraction conditions

In the previous section, we discussed that contrast in BFSCEM requires multiple scattering within the sample. Indeed, in many experiments, it is desirable to orient the sample along a high-symmetry direction where the atomic positions align in discrete columns. For example, one may want to map the depth of individual impurity atoms in the atomic columns of a crystalline sample. We present in this section a review of some recent theoretical work on the propagation of a focused STEM beam in such a sample.

When a STEM probe of atomic dimensions is focused on an atomic column at the entrance surface of an aligned crystal, strong channelling conditions are established (Fertig & Rose 1981). The probe tends to channel along the atomic column with reduced spreading. This channelling is one of the factors that allows the relatively simple interpretation of ADF STEM images (Pennycook & Jesson 1991; Nellist & Pennycook 1999). Channelling can be modelled theoretically using a Bloch wave model. Using the approach described in Nellist & Pennycook (1999) and Cosgriff & Nellist (2007), the wave function in the crystal can be written as a sum of Bloch waves

$$\Psi(r, z, r_0) = \sum_j \int \sum_g \Phi^j_0(k) \Phi^j_g(k) e^{-2\pi i g \cdot r_0} e^{-2\pi i k_z \cdot z} e^{\pi i \Delta f/k^2 \lambda} e^{-2\pi i k \cdot (r - r_0)} dk,$$

where \(r_0\) is the lateral location of the illuminating probe, \(j\) is the Bloch state index, \(g\) represents the reciprocal lattice vectors in the zero layer, \(k\) is the wavevector transverse component of a partial plane wave in the convergent beam forming the probe, \(\Phi^j_g\) is a Fourier component of a Bloch wave, \(k_z^j\) is the forward wave vector of the wave from the dispersion surface and \(\Delta f\) is the defocus. The contribution of a single Bloch state to the wave function can be separated out by examining individual terms in the \(j\) summation. At zero thickness, the contribution of an individual state depends on a phase-linked summation of all the partial plane waves, \(k\), that exist in the convergent beam forming the probe multiplied by the excitation parameter, \(\Phi^j_0\), of that state at that illumination angle. It should be noted that in the presence of absorption this form of the excitation parameter is only an approximation. Figure 16 shows that the channelling states (usually referred to as the 1s states by analogy with the most bound atomic orbital states) for an As column in GaAs have large excitation coefficients even at relatively high semiangles. Prior to aberration correction, typical semiangles of convergence would range up to 10 mrad. Figure 16 shows that up to this angle, the excitation of the 1s states remains strong. However, as pointed out previously by Peng et al. (2004), the illumination angles allowed by aberration-corrected instruments can reach values where the 1s excitation starts to significantly fall off. An interpretation of this is that we are reaching angles where the scattering is becoming more kinematical owing to the large excitation errors involved, and strong dynamical scattering, leading to channelling, is not possible for electrons travelling at these angles.

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A detailed theoretical analysis of depth sectioning for GaAs aligned along the (1 1 0) zone axis (Cosgriff & Nellist 2007) shows that when an aberration-corrected probe is located above an atomic column, while the 1s has the largest excitation, around 60 states have significant excitation, with high-angle, more plane-wave-like states excited by higher angled partial plane waves in the convergent beam.

An optical sectioning experiment can now be simulated by including a defocus term, $\Delta f$, in the phase variation across the partial plane waves converging to form the beam, as is included in equation (4.1). A peak in the intensity of the electron wave function will be seen in the crystal at a depth where the variation in phase owing to the Bloch state eigenvalues maximally compensates for the defocus phase change. In Cosgriff & Nellist (2007), it was shown that at specific depths in the crystal, there is a phase alignment of the contribution of all the states leading to a peak in the wave function intensity. When the probe was located between atomic columns, this maximum was found to occur at the depth selected by the defocus of the probe. However, moving the probe so that a column of atoms was illuminated resulted in a phase alignment and intensity maximum closer to the entrance surface than the position of the focal plane in the sample. This was found not to be related to the 1s state, and is therefore not a channelling effect. The pre-focusing resulted from interference between all states, including the higher angle ones. It was concluded that what was being observed was an atomic lensing effect by the atomic column that pre-focused the intensity maximum. The typical pre-focusing for this case was usually approximately 2 nm.

Superimposed on this pre-focused probe intensity a more quickly varying pendellösung effect from the interference of the 1s state with the remaining Bloch wave ensemble was observed. These channelling oscillations are well known, and are most straightforwardly understood in terms of the interference of the 1s state with the remaining Bloch wave ensemble (Geuens & Van Dyck 2002). The 1s state oscillation is superimposed on the pre-focused broader peak, leading to a relatively complicated depth intensity profile (figure 17). It is therefore possible

![Figure 16. The modulus of the excitation coefficient for the As 1s state as a function of aperture semiangle. The excitation coefficient is circularly symmetric within the aperture. The accelerating voltage used was 300 kV.](http://rsta.royalsocietypublishing.org/)

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Figure 17. Intensity as a function of depth when the probe is located above the As column (solid line) and a schematic pre-focused peak (dashed line) for a probe defocus of $-13.0 \text{ nm}$. An accelerating voltage of 300 kV and a semiangle of 22 mrad were used for this simulation.

Figure 18. Intensity recorded in the SCEM geometry when the confocal condition is broken. The focal points for both the probe and the imaging lens are centred on a Ga column in GaAs $\langle 110 \rangle$ (solid line) and at a point equidistant from all columns (dashed line). Both probes are defocused by the same amount, and the focal difference shown is the sum of both defoci, which is the physical distance between the nominal focal planes for the two probes. At zero focal difference, both probes are centred on the physical centre of the 20 nm thick sample. An accelerating voltage of 200 kV and a semiangle of 30 mrad were used for this simulation.

that in this case the intensity profile in the depth of the crystal can show a double maximum. Such dynamical diffraction effects will naturally affect the SCEM image-forming process. Figure 18 shows the BFSCEM intensity that is recorded as the focal planes for the upper and lower optics are displaced symmetrically from the midplane, with both sets of optics laterally focused on the same Ga column. When both sets of optics are underfocused by 1 nm each (2 nm aggregate), there is an intensity maximum, which presumably results from the atomic column pre-focusing effect. Another maximum occurs when the optics are focused on their
respective closest surface. In this case, it is likely that channelling is playing a major role. The probe incident on the entrance surface channels along the atomic column, emerging as a local intensity maximum on the exit surface, which is then focused on the pinhole by the post-specimen optics. Conversely, when the focal points are translated laterally away from an atomic column, no channelling peak is seen. The maximum seen with both optics overfocused suggests that the crystal produces a slightly divergent effect in this case.

Although the above discussion does elucidate some of the processes involved in optical sectioning influenced by dynamical scattering, it is clear that it is still a complex process and that modelling of the scattering process is required for any kind of data interpretation.

5. Concluding remarks

We have shown experimentally that the missing cone in the OTF of ADF STEM optical sectioning leads to large elongation in the three-dimensional images of laterally extended objects. Although the depth of field of an aberration-corrected STEM may only be a few nanometres, such depth resolutions can only be realized for point-like objects such as single atoms. We have also shown that a commonly used method of deconvolution, the RL algorithm, is unlikely to be effective in reducing this elongation because the method is unable to recover most of the information lost in the missing cone.

Despite this poor resolution, when performing optical sectioning for certain kinds of samples (i.e. nanoparticles), we can generate a three-dimensional reconstruction with some image processing through the use of prior information. The method presented here is very specific to our particular sample, through the assumption of spherical particles with sharp edges. With further work, this method could be extended to particles of any shape, and better sampling when acquiring data would improve the accuracy of future measurements at the cost of increasing the time and memory requirements of the reconstruction. However, this method could not be regarded as a general one. Nonetheless, the method does provide a fast and simple way of three-dimensional mapping of particle distributions in heterogeneous catalyst systems without the requirement of a tomographic series.

In light optics, a confocal geometry is commonly used to reduce the contribution of scattering away from the focal plane, and we have shown how the confocal geometry fills the OTF missing cone in the case of incoherent imaging. Energy-filtered confocal imaging in the TEM is a close approximation to incoherent imaging, but a limitation to the depth resolution in the case of energy-filtered imaging will be the chromatic aberration of the imaging system. The advent of correctors for chromatic aberration will have a significant impact in this regard and will have uses, for example, in elemental mapping of buried layers and thin films.

For confocal TEM based on elastic scattering, multiple scattering is required to generate contrast. Simulations of the optical sectioning process in strong channelling conditions illustrate the complexities introduced in this case, and while a rich dataset may be recorded, its interpretation is likely to require matching to detailed simulations.
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References


