CHAPTER 5

Electron Holography of Nanostructured Materials

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5.1 Introduction

The technique of electron holography, which was originally described by Gabor,\textsuperscript{1} is based on the formation of an interference pattern or “hologram” in the Transmission Electron Microscope (TEM). In contrast to most conventional TEM techniques, which only allow the spatial distribution of image intensity to be recorded, electron holography also allows the phase shift of the high-energy electron wave that has passed through a specimen to be measured directly. The phase shift can, in turn, be used to provide information about local variations in magnetic induction and electrostatic potential in and around the specimen. This chapter begins with an outline of the theoretical background and experimental procedures that are required to obtain and analyse phase information from electron holograms. Medium-resolution applications of the off-axis, or “side-band”, TEM mode of electron holography to the characterisation of nanostructured materials are then described, followed by a description of high-resolution electron holography and alternative forms of electron holography.
5.1.1 Basis of Off-Axis Electron Holography

The TEM mode of off-axis electron holography involves the examination of an electron-transparent specimen using defocused illumination from a highly coherent Field-Emission Gun (FEG) electron source. The region of interest is positioned so that it covers approximately half the field of view. The application of a voltage to an electron biprism results in the overlap of a “reference” electron wave that has passed through vacuum with the electron wave that has passed through the specimen, as shown schematically in Figure 5.1(a). If the electron source is sufficiently coherent, then an interference fringe pattern (an electron hologram) is formed in the overlap region, in addition to an image of the specimen. The amplitude and the phase shift of the specimen wave are recorded in the intensity and the position, respectively, of the fringes. For studies of magnetic materials, a Lorentz lens (a high-strength minilens) allows the microscope to be operated at high magnification with the objective lens switched off and the sample in magnetic-field-free conditions.

Figure 5.2(a) shows a representative electron hologram of a thin crystal of the mineral hematite. Changes in the positions of the holographic interference fringes are visible in the inset to Figure 5.2(a).

An expression for the intensity distribution in an off-axis electron hologram can be obtained by considering an addition of a tilted plane reference wave to a complex specimen wave, in the form

\[ I_{\text{hol}}(r) = |\psi_f(r) + \exp[2\pi i \mathbf{q}_r \cdot \mathbf{r}]|^2 \]  \hspace{2cm} (5.1)

where \( \mathbf{r} \) is a two-dimensional vector in the plane of the sample, the tilt of the reference wave is specified by the two-dimensional reciprocal space vector \( \mathbf{q} = \mathbf{q}_r \), and the electron wavefunction in the image plane is given by the expression

\[ \psi_i(r) = A_i(r) \exp[i\phi_i(r)] \]  \hspace{2cm} (5.2)

where \( A \) and \( \phi \) refer to amplitude and phase, respectively. By combining these two expressions, eqn (5.1) can be rewritten in the form

\[ I_{\text{hol}}(r) = 1 + A_i^2(r) + 2A_i(r)\cos[2\pi \mathbf{q}_r \cdot \mathbf{r} + \phi_i(r)] \]  \hspace{2cm} (5.3)

From eqn (5.3), it can be seen that there are three separate contributions to the intensity distribution in a hologram: the reference image intensity, the specimen image intensity, and a set of cosinusoidal fringes, whose local phase shifts and amplitudes are equivalent to the phase and amplitude, respectively, of the electron wavefunction in the image plane.

In order to extract phase and amplitude information, an off-axis electron hologram is first Fourier transformed. From eqn (5.3), it can be shown that the
Figure 5.1 (a) Schematic illustration of the experimental setup used for generating off-axis electron holograms. The specimen occupies approximately half the field of view. Essential components are the field-emission gun electron source, which provides coherent illumination, and the electron biprism, which causes overlap of the object and (vacuum) reference waves. The biprism is usually a wire, below 1 μm in diameter, located in place of one of the conventional selected-area apertures. The sample and reference waves can be considered as originating from two virtual sources, S1 and S2. The Lorentz lens allows imaging of magnetic materials in close-to-field-free conditions. (b) Schematic diagram illustrating the use of specimen tilt to provide an inplane component of the vertical magnetic field, H, of the conventional electron microscope objective lens for in situ magnetization-reversal experiments.
Figure 5.2 Sequence of image processing steps used to convert an off-axis electron hologram into a phase image. (a) Representative off-axis electron hologram of the region of interest on a specimen (the edge of a thin foil of hematite containing nanoscale inclusions of maghemite). The inset shows a magnified image of the holographic interference fringes in the outlined region. Coarser fringes, resulting from Fresnel diffraction at the edges of the biprism wire, are visible at the upper right and lower left corners of the original hologram. (b) Reference hologram obtained from a region of vacuum immediately after recording the hologram of the specimen. (c) Modulus squared of the Fourier transform of the electron hologram shown in (a), comprising a central peak, two sidebands, and a diagonal streak arising from the presence of the Fresnel fringes in the hologram. (d) One of the sidebands is extracted from the Fourier transform of the hologram, and a circular mask with smooth edges is used to reduce its intensity radially to zero. If required, the streak from the Fresnel fringes can be removed in a similar manner, by assigning a value of zero to pixels inside the region shown by the dashed line. (e) Analysis of the inverse Fourier transforms of sidebands from the specimen hologram and the reference hologram yields the complex image wave of the specimen, from which the phase is initially calculated modulo 2\pi. (f) Phase unwrapping algorithms are used to remove the 2\pi phase discontinuities from (e) to yield a final unwrapped phase image.
Fourier transform of a hologram can be written in the form

\[
\text{FT}[h_{\text{ref}}(r)] = \delta(q) + \text{FT}[\hat{A}^2(r)] \\
+ \delta(q + q_c) \otimes \text{FT}[A_l(r) \exp[i\phi_l(r)]] \\
+ \delta(q - q_c) \otimes \text{FT}[A_l(r) \exp[-i\phi_l(r)]]
\]  

(5.4)

Equation (5.4) describes a peak at the reciprocal-space origin corresponding to the Fourier transform of the reference image, a second peak centred at the origin corresponding to the Fourier transform of a conventional bright-field TEM image of the sample, a peak centred at \(q = -q_c\) corresponding to the Fourier transform of the desired image wavefunction, and a peak centered at \(q = +q_c\) corresponding to the Fourier transform of the complex conjugate of the wavefunction. Figure 5.2(c) shows the Fourier transform of the hologram shown in Figure 5.2(a). In order to recover the complex electron wavefunction, one of the two “sidebands” in the Fourier transform is selected, as shown in Figure 5.2(d), and then inverse Fourier transformed.

As phase information is stored in the lateral displacement of the holographic interference fringes, long-range phase modulations arising from inhomogeneities in the charge and the thickness of the biprism wire, as well as from lens distortions and charging effects (e.g., at apertures) can introduce artifacts into the reconstructed wavefunction. In order to take these effects into account, a reference hologram is usually obtained from vacuum alone by removing the specimen from the field of view without changing the optical parameters of the microscope (Figure 5.2(b)). Correction is then possible by performing a complex division of the recovered sample and vacuum wavefunctions in real space, and then calculating the phase (the arctangent of the ratio of the imaginary and real parts) of the resulting complex wavefunction, to obtain the distortion-free phase of the image wave\(^2\) (Figure 5.2(e)). The acquisition of a reference hologram has the additional advantage that the use of the same locations for the sidebands determined from the sample and vacuum holograms removes any tilt of the recorded wave that might be introduced by an inability to locate the exact (subpixel) position of the sideband frequency in Fourier space. Figure 5.2(e) illustrates the fact that a phase image that is calculated digitally is initially evaluated modulo \(2\pi\), meaning that \(2\pi\) phase discontinuities that are unrelated to specimen features appear at positions where the phase shift exceeds multiples of this amount. If required, the phase image may then be “unwrapped” by using suitable algorithms,\(^3\) as shown in Figure 5.2(f).

The recorded phase shift can be used to measure the electrostatic potential and also the inplane component of the magnetic induction in the specimen. Neglecting dynamical diffraction (i.e. assuming that the specimen is thin and weakly diffracting), the phase shift can be expressed in the form

\[
\phi(x) = C_F \int V(x, z) \, dz - \left(\frac{e}{\hbar}\right) \iint B_\perp(x, z) \, dx \, dz
\]  

(5.5)
where

$$
C_E = \left( \frac{2\pi}{\lambda} \right) \left( \frac{E + E_0}{E(E + 2E_0)} \right)
$$

(5.6)

\( z \) is the electron-beam direction, \( x \) is a direction in the plane of the specimen, \( B_\perp \) is the component of the magnetic induction perpendicular to both \( x \) and \( z \), \( V \) is the electrostatic potential, \( \lambda \) is the (relativistic) electron wavelength, and \( E \) and \( E_0 \) are, respectively, the kinetic and rest mass energies of the incident electron.\(^4\) \( C_E \) takes values of \( 7.29 \times 10^6 \), \( 6.53 \times 10^6 \) and \( 5.39 \times 10^6 \) rad \( V^{-1} \ m^{-1} \) at accelerating voltages of 200 kV, 300 kV and 1 MV, respectively. If neither \( V \) nor \( B_\perp \) varies along the electron-beam direction within a sample of thickness \( t \), then eqn (5.5) can be simplified as

$$
\phi(x) = C_E V(x) t(x) - \left( \frac{e}{\hbar} \right) \int B_\perp(x) r(x) \, dx
$$

(5.7)

Hence, by making use of eqns (5.5)–(5.7), information about \( V \) and \( B_\perp \) can be recovered from a measured phase image, as described subsequently.

### 5.1.2 Experimental Considerations

Several practical issues must be addressed in order to record and analyse an electron hologram successfully. A key experimental requirement is the availability of a reference wave that can be overlapped onto the region of interest on the specimen. This restriction, which usually implies that the hologram must be recorded from a region close to the specimen edge, can be relaxed if a thin, clean, weakly diffracting region of electron-transparent support film, rather than vacuum, can be overlapped onto the region of interest.

Electron holograms have traditionally been recorded on photographic film. However, digital acquisition using charge coupled device (CCD) cameras is now used widely due to their linear response, high dynamic range and high detection quantum efficiency, as well as the immediate accessibility of the recorded information.\(^3\) Whether a hologram is recorded on film or digitally, the field of view is typically limited to approximately 5 \( \mu \)m, either by the dimensions of the recording medium or by the sampling and the contrast of the holographic fringes.

The high electron beam coherence required for electron holography necessitates the use of a FEG electron source, a small spot size, a small condenser aperture size and a low gun extraction voltage. The coherence may be maximised by adjusting the condenser-lens stigmators in the microscope to provide elliptical illumination that is wide in a direction perpendicular to the biprism when the condenser lens is overfocused.\(^5\) The contrast of the holographic fringes is also determined by the mechanical stability of the biprism and the point spread function of the recording medium. Should the fringe contrast decrease too much, reliable reconstruction of the image wavefunction will not be possible. A phase detection limit for electron holography\(^7\) can be determined from the effect on a
recorded hologram of Poisson-distributed shot noise, the detection quantum efficiency and the point spread function of the CCD camera, and the fringe contrast. In practice, averaging of the phase is often implemented, particularly if the features of interest vary slowly across the image or in one direction.

A final artifact results from the presence of Fresnel diffraction from the biprism wire, which is visible in Figures 5.2(a) and (b), and causes the diagonal streak at the lower left of Figure 5.2(d), as well as subsequent phase and amplitude modulations of both the specimen wave and the reference wave.\textsuperscript{8} These effects can be removed to an extent by using a reference hologram (Figure 5.2(b)), and by Fourier-filtering the sideband before reconstruction of the image wave (Figure 5.2(d)). More advanced approaches for removing Fresnel fringes from electron holograms based on image analysis\textsuperscript{9} and double-biprism electron holography\textsuperscript{10} have recently been introduced. Great care should also be taken to assess the effect on the reference wave of long-range electromagnetic fields that may extend outside the sample and affect both the object wave and the reference wave.\textsuperscript{11}

Further details about the theoretical background and practical aspects of electron holography can be obtained from recent books\textsuperscript{12-14} and review papers.\textsuperscript{15-19}

### 5.2 The Mean Inner Potential Contribution to the Phase Shift

If a phase image is associated solely with variations in mean inner potential and specimen thickness, then electron holography can be used to characterise local variations in specimen morphology and composition. For a specimen that has uniform structure and composition in the electron-beam direction, in the absence of magnetic and long-range electrostatic fields (such as those at depletion regions in semiconductors), eqn (5.7) takes the simpler form

\[
\phi(x) = C_E V_0(x) \tau(x)
\]  
(5.8)

where the mean inner potential, \(V_0\), is the volume average of the electrostatic potential in the specimen. Theoretical values for \(V_0\) can be obtained by assuming that the specimen can be described as a collection of free atoms and using the expression

\[
V_0 = \left( \frac{\hbar^2}{2\pi m e^2} \right) \sum_e f_e(0)
\]  
(5.9)

where \(f_e(0)\) is the electron scattering factor at zero scattering angle (with dimensions of length), \(\Omega\) is the unit cell volume, and the sum is performed over all atoms in the unit cell.\textsuperscript{9} Although calculated values for \(f_e(0)\) have been tabulated,\textsuperscript{20,21} the use of eqn (5.9) typically leads to overestimates for \(V_0\) by approximately 10% because the redistribution of electrons due to bonding, which results in a contraction of the electron density around each atom is neglected.\textsuperscript{22,23}
Experimental measurements of $V_0$ have been obtained using electron holography, in combination with independent measurements of specimen thickness profiles. Values for $V_0$ of 13.0 ± 0.1, 14.5 ± 0.2, 17.2 ± 0.1 and 14.3 ± 0.2 V have been obtained from cleaved wedges and cubes of MgO, GaAs, PbS and Ge, respectively.\textsuperscript{24-26} By analysing GaAs, InAs, GaP and InP wedges, values of 14.2 ± 0.2, 14.5 ± 0.2, 14.4 ± 0.2, 14.5 ± 0.1 V, respectively, were obtained,\textsuperscript{27} while wedge-shaped Si samples with stacked Si oxide layers on their surfaces were used to measure the mean inner potentials of the oxide layers.\textsuperscript{28} Si nanospheres coated in layers of amorphous SiO$_2$ have been used\textsuperscript{29} to determine values of 12.1 ± 1.3 V, 11.9 ± 0.9 V and 10.1 ± 0.6 V for crystalline Si, amorphous Si and amorphous SiO$_2$, respectively. Similar measurements obtained from spherical latex particles embedded in vitrified ice were used to provide values of 8.5 ± 0.7 and 3.5 ± 1.2 V for the two materials, respectively.\textsuperscript{30} While cylindrical GaN nanowires suspended over holes in a carbon support film were used\textsuperscript{31} to measure a value for $V_0$ of GaN of 16.7 ± 0.3 V. Care is always required when measuring $V_0$ using electron holography, as dynamical contributions to the phase shift can complicate the determination of $V_0$ from crystalline samples. Equation (5.8) is no longer valid when the sample is tilted to a strongly diffraacting orientation, as the phase shift can then vary nonlinearly with sample thickness, and is also very sensitive to small changes in sample orientation. Such effects can be simulated by using either multislice or Bloch-wave algorithms. Additional experimental factors that may affect measurements of $V_0$ include the chemical and physical state and the crystallographic orientation of the specimen surface\textsuperscript{32} and specimen charging.\textsuperscript{33-35}

If $V_0$ is known, then measurements of phase shift can be used to determine the local specimen thickness, $t$. Examples of the measurement of specimen shapes from phase images include the characterisation of faceted ZrO$_2$ crystals,\textsuperscript{36} carbon nanotubes\textsuperscript{37} and bacterial flagellae.\textsuperscript{38} Such measurements can, in principle, be extended to three dimensions by combining electron holography with electron tomography, as demonstrated by the analysis of tilt series of electron holograms of latex particles.\textsuperscript{39}

The specimen thickness can also be inferred from a holographic amplitude image in units of $\lambda_{im}$, the mean free path for inelastic scattering, by making use of the expression

$$\frac{t(x)}{\lambda_{im}} = -2 \ln \left( \frac{A_s(x)}{A_r(x)} \right)$$  \hspace{1cm} (5.10)

where $A_s(x)$ and $A_r(x)$ are the amplitudes of the sample and reference holograms, respectively.\textsuperscript{39} If desired, the thickness dependence of both the phase and the amplitude image can then be removed by combining eqns (5.8) and (5.10) as

$$\frac{\phi(x)}{-2C_F \ln \left( \frac{A_s(x)}{A_r(x)} \right)} = V_0(x) \lambda_{im}(x)$$ \hspace{1cm} (5.11)
to generate an image, in which the contrast is the product of the local values of
the mean inner potential and the inelastic mean free path. These parameters
depend only on the local composition of the sample. Equation (5.11) can
therefore be useful for interpreting images obtained from samples with varying
composition and thickness.  

5.3 Measurement of Magnetic Fields

When examining magnetic materials using electron holography, the conven-
tional TEM objective lens is usually switched off, as its strong magnetic field
would be likely to saturate the magnetisation in the sample in the electron-beam
direction. Instead, a high-strength minilens (a Lorentz lens) located below the
objective lens can be used to provide high magnification (~ 50–75 k×) with the
sample either in a magnetic-field-free environment or in a chosen (precalibrat-
ed) vertical magnetic field provided by using an intermediate setting of the
TEM objective lens.

5.3.1 Early Experiments

Early examples of the examination of magnetic materials using electron
holography involved the reconstruction of electron holograms using a laser
bench, and included the characterisation of horseshoe magnets, 42 magnetic
recording media 43 and flux vortices in superconductors. 44, 46 The most elegant
of these experiments involved the confirmation of the Aharonov–Bohm effect, 47
which states that when an electron wave from a point source passes on either
side of an infinitely long solenoid the relative phase shift that occurs between
the two parts of the wave results from the presence of a vector potential. The
Aharonov–Bohm effect provides the only observable confirmation of the
physical reality of gauge theory. Electron holography experiments were carried
out on 20 nm thick permalloy toroidal magnets that were covered with 300 nm
thick layers of superconducting Nb, which prevented electrons from penetrat-
ing the magnetic material and confined the magnetic flux by exploiting the
Meissner effect. The observations showed that the phase difference between the
centre of the toroid and the region outside it was quantised to a value of 0 or π
when the temperature was below the Nb superconducting critical temperature
(5 K), i.e. when a supercurrent was induced to circulate in the magnet. 48, 49
Although the observed quantisation of magnetic flux, and the measured phase
difference with the magnetic field entirely screened by the superconductor,
appeared to provide confirmation of the Aharonov–Bohm effect, alternative
explanations for these observations are still proposed. 50

5.3.2 Experiments Involving Digital Acquisition and Analysis

Recent applications of electron holography to the characterisation of magnetic
fields in nanostructured materials have almost invariably made use of digital
recording. The off-axis mode of electron holography, combined with digital image processing, is ideally suited to the characterisation of such materials because unwanted contributions to the contrast from local variations in composition and specimen thickness can be removed from a phase image more easily than from images recorded using other phase-contrast techniques. For example, the Fresnel and Foucault modes of Lorentz microscopy and differential phase contrast (DPC) imaging provide signals that are approximately proportional to the first or the second differential of the phase shift. These techniques enhance contributions to the contrast from rapid variations in specimen thickness and composition, as compared to the weak and slowly varying magnetic signal of primary interest.

Of particular interest for magnetic materials is the digital determination of the gradient of a phase image. If \( V \) and \( B_\perp \) do not vary in the electron-beam direction in the specimen, then the phase gradient can be written in the form

\[
\frac{d\phi(x)}{dx} = C_x \frac{d}{dx} \left\{ V(x) t(x) \right\} - \left( \frac{e t}{h} \right) B_\perp(x) t(x)
\]  

(5.12)

According to eqn (5.12), for a specimen of uniform thickness and composition, the phase gradient is directly proportional to the inplane component of the magnetic induction in the specimen, \( i.e. \)

\[
\frac{d\phi(x)}{dx} = -\left( \frac{e t}{h} \right) B_\perp(x)
\]  

(5.13)

A graphical representation of the strength and direction of the local magnetic induction can therefore be obtained simply by adding contours to a recorded phase image. A phase difference of \( 2\pi \) between such contours then corresponds to an enclosed magnetic flux of \( 4 \times 10^{-15} \) Wb. Unfortunately, even if the first term in eqn (5.12) is negligible, great care is required when interpreting contoured phase images acquired from general specimens, in which the region of interest may have variable thickness and finite lateral extent, because the gradient of the magnetic contribution to the phase shift then contains contributions from the internal demagnetising field and stray magnetic fields outside the sample.

The primary advantage of the digital analysis of electron holograms is that the magnetic and mean inner potential contributions to the measured phase shift can be separated, particularly at the edges of magnetic nanoparticles, where rapid variations in specimen thickness can dominate both the phase and the phase gradient. Several approaches can be used to achieve this separation. The sample may be inverted to change the sign of the magnetic contribution to the phase and a second hologram recorded. The sum and the difference of the two phase images can then be used to provide twice the magnetic contribution, and twice the mean inner potential contribution, respectively. Alternatively, two holograms may be acquired from the same region of the specimen at two different microscope accelerating voltages. In this case, the magnetic signal is independent of accelerating voltage, and subtraction of the two phase images can be used to
provide the mean inner potential contribution. A more practical method often
involves performing a magnetisation reversal experiment \textit{in situ} in the electron
microscope, and subsequently selecting pairs of holograms that differ only in the
(opposite) direction of the magnetisation in the specimen. Just as when turning
the specimen over, the magnetic and mean inner potential contributions to the
phase shift are calculated by taking half the difference, and half the sum, of the
resulting phase images. The mean inner potential contribution can then be
subtracted from all other phase images acquired from the same specimen region. In
\textit{in situ} magnetisation reversal, which is required both for this purpose and
for performing magnetisation reversal experiments in the TEM, can be achieved
by exciting the conventional microscope objective lens slightly and tilting the
specimen to apply known inplane magnetic fields, as shown schematically in
Figure 5.1(b). In practice, if the two remanent magnetic states are not exactly
equal and opposite to each other, then it may be necessary to repeat the switching
process several times so that nonsystematic differences between switched pairs of
phase images average out. Such differences, which can lead to artifacts in the final
magnetic-induction map, are often identified by inspection. By varying the
applied field, it is possible to record a series of images that correspond to any
desired point on a remanent hysteresis loop or magnetisation reversal cycle.

\subsection{Isolated Magnetic Nanoparticles}

Analytical expressions for the phase shift of a magnetic particle, based on eqn
(5.5), can be derived for a uniformly magnetised sphere of radius \(a\), magnetic
induction \(B_{\perp}\) (along \(y\)) and the mean inner potential \(V_0\) in the form

\[
\phi(x, y) |_{x^2 + y^2 \leq a^2} = 2C_\mu V_0 \sqrt{a^2 - (x^2 + y^2)} + \left( \frac{\mu_0}{\hbar} B_{\perp} a \right) \left( \frac{x}{x^2 + y^2} \right) \left\{ 1 - \left( 1 - \left( \frac{x^2 + y^2}{a^2} \right) \right)^{\frac{3}{2}} \right\} \tag{5.14}
\]

\[
\phi(x, y) |_{x^2 + y^2 > a^2} = \left( \frac{\mu_0}{\hbar} B_{\perp} a \right) \left( \frac{x}{x^2 + y^2} \right) \tag{5.15}
\]

Graphical representations of eqns (5.14) and (5.15) are shown in Figure 5.3
for a uniformly magnetised 100 nm diameter spherical particle of iron, on the
assumption that \(V_0 = 22\) V and \(B_{\perp} = 2.2\) T. The total phase shift (Figure 5.3(c))
is the sum of mean inner potential (Figure 5.3(a)) and magnetic (Figure 5.3(b))
contributions. Line profiles, generated from Figures 5.3(a)–(c) along a line
passing through the centre of the particle in a direction perpendicular to its
magnetisation direction, are shown in Figures 5.3(d)–(f). As mentioned above,
the inplane component of the magnetic induction integrated in the electron-
beam direction can be visualised by adding contours to the magnetic contribution
to the phase shift \(\phi_{\text{MAG}}\), as shown in Figure 5.3(h) in the form of the
Figure 5.3  Simulations of the phase shift associated with a 100 nm diameter spherical particle of iron. The particle is assumed to be magnetised uniformly in the vertical direction, with $V_{th} = 22$ V and $B_z = 2.2$ T. The mean inner potential contribution to the phase shift is shown in (a), the magnetic contribution in (b), and the sum of the two contributions in (c). A small amount of noise has been added to the simulations. (d)-(f) show line profiles, taken from images (a)-(c) horizontally through the centre of the particle (*i.e.* in a direction normal to the magnetisation direction). The analytical forms of these graphs are given by eqns (5.14) and (5.15). In (d) $\text{min} = -0.3$, $\text{max} = 14.5$; in (e) $\text{min} = -9.2$, $\text{max} = 9.1$; in (f) $\text{min} = -8.7$, $\text{max} = 18.7$. (g)-(i) Magnetic phase contours ($4 \times$ amplification; 1.57 rad spacing), corresponding to the cosine of four times each of the phase images shown in (a)-(c). (j) Colour map derived from the gradient of the magnetic contribution to the phase shift shown in (b). The hue and intensity of the colour are used to label the direction and magnitude, respectively, of the in-plane component of the magnetic induction integrated in the electron-beam direction, using the colour wheel shown in (k) (red = right, yellow = down, green = left, blue = up). The colours can be combined with the contour map, as shown in (l).
cosine of four times the phase image. The horizontal and vertical derivatives of the magnetic contribution to the phase shift \((d\phi_{\text{MAG}}/dx)\) and \((d\phi_{\text{MAG}}/dy)\) can be used to generate either an arrow map or a colour map (Figure 5.3(j)), in which the direction and magnitude of the projected inplane magnetic induction are represented by the hue and intensity of the colour, respectively, according to the colour wheel shown in Figure 5.3(k). The colours can also be combined with the cosine image if desired (Figure 5.3(l)). Equations (5.14) and (5.15) have been used to interpret phase images recorded from Co particles suspended over a hole in a carbon support film. In this study, the magnetic induction and mean inner potential of each particle were determined by fitting experimental line traces to simulations similar to that shown in Figure 5.3(f).

Figure 5.4 shows experimental results obtained using electron holography from an isolated 50 nm diameter single crystal of magnetite (Fe₃O₄) from a bacterial cell. Figure 5.4(a) shows a high-resolution TEM image of the crystal. The three-dimensional morphology and orientation of the crystal (Figure 5.4(b)) were determined by applying electron tomography to a series of High-Angle Annular Dark-Field (HAADF) images of the particle taken over a high range of specimen tilt angles. The tomographic reconstruction reveals that the particle is elongated slightly in the \([111]\) direction in the plane of the specimen, as indicated by the white arrow in Figure 5.4(a). Electron holograms of the magnetite crystal were acquired in magnetic-field-free conditions, both at room temperature (Figure 5.4(c)) and at 90 K (Figure 5.4(d)). The magnetic contribution to the phase shift was isolated by performing a series of in situ magnetisation reversal experiments, as described previously. The direction of the inplane component of the applied field is indicated by the black double arrow. Both images show uniformly magnetised single-domain states, including the characteristic return flux of an isolated magnetic dipole (Figure 5.3(h)). In both cases, the remanent magnetisation direction appears to make a large angle to the applied field direction. At room temperature, the phase contours in the crystal make an angle of \(\sim 30^\circ\) to its \([111]\) elongation direction (Figure 5.4(c)). The contours appear to be parallel to the \([111]\) elongation direction at 90 K (below the Verwey transition for magnetite; Figure 5.4(d)). Figure 5.4(e) shows a line profile generated from the magnetic phase image that was used to create Figure 5.4(c), taken along a line passing through the centre of the crystal in a direction perpendicular to the phase contours. A least-squares fit of the experimental phase profile to eqn (5.14) and (5.15) yielded a value for \(B_\perp\) of 0.6 ± 0.12 T. This value is equal to the room-temperature saturation induction of magnetite, suggesting that the magnetisation direction of the particle lies in the plane of the specimen, close to the [131] crystallographic direction. This direction corresponds to the longest diagonal dimension of the particle, which is consistent with shape anisotropy dominating the magnetic state of the crystal at room temperature. The 90 K phase profile yielded a value for \(B_\perp\) of 0.46 ± 0.09 T. This value is lower than the saturation induction of magnetite at 90 K, suggesting that, at remanence, the magnetisation direction in the crystal is tilted out of the plane by \(\sim 40^\circ\) to the horizontal. This direction is close to either [210] or [012]. Below the Verwey transition, the
Figure 5.4  (a) High resolution image of a faceted 50 nm diameter magnetite crystal in a magnetotactic bacterium. (b) Visualisation of a high-angle annular dark-field tomographic reconstruction of the same particle. (c) and (d) Magnetic induction maps showing remanent states recorded from the particle at room temperature and at 90 K, respectively. The images were recorded after tilting the sample by ±30° and applying a vertical 2 T magnetic field using the TEM objective lens. The inplane component of the applied field was directed along the black double arrow. The mean inner potential contribution to the phase shift has been removed from each image (see text for details). (e) Line profile of the magnetic contribution to the phase shift recorded from the magnetite particle at 90 K (filled circles), taken through the centre of the particle in a direction normal to the contours shown in (d). The solid line shows a least-squares fit of eqns (5.14) and (5.15) to the data. Such fits were used to provide values for the inplane component of $B$ of 0.60 ± 0.12 T at room temperature and 0.46 ± 0.09 T at 90 K.
magnetocrystalline anisotropy of magnetite is known to increase considerably in magnitude, and to switch from $<111>_{\text{cubic}}$ to $[001]_{\text{monoclinic}}$. The $[001]_{\text{monoclinic}}$ easy axis can lie along any one of the original $<100>_{\text{cubic}}$ directions. Both the [100] and the [001] direction of the original cubic crystal lie close to the observed remanence direction, suggesting that magnetocrystalline anisotropy has a more significant effect on the remanence direction at 90 K than at room temperature. The fact that the remanence direction in Figure 5.4(d) is perpendicular to the applied field direction suggests that this choice may be influenced by the morphology of the crystal.

When acquiring electron holography results similar to those shown in Figure 5.4, it is important to recognise that a sample with uniaxial anisotropy constant, $K$ and saturation magnetisation, $M_s$ that is tilted by an angle of 30° is expected to reverse magnetically when the vertical field reaches 0.52 $B_K$, where $B_K = 2K/M_s$ is the coercivity for fields applied along the anisotropy axis, as shown in the form of a schematic diagram in Figure 5.5. It is also important to relate experimentally acquired magnetic-induction maps, such as those shown in Figures 5.4(c) and (d), to theoretical predictions of the effect of the size and shape of a particle on its magnetic state. In Figure 5.6, the upper solid line shows the theoretical boundary between single domain and two domain states for magnetite nanoparticles. The dashed line shows the boundary between single-domain and single-vortex states, as predicted by micromagnetic simulations.

Figure 5.5 Schematic illustration of magnetic switching of a nanoparticle in the TEM, achieved by tilting the sample by 30° and applying a magnetic field using the conventional microscope objective lens. A uniaxial particle with anisotropy constant, $K$ and saturation magnetisation, $M_s$, initially oriented horizontally and magnetised to the right, is tilted by an angle of 30° to the horizontal. A chosen current is passed through the objective lens of the TEM, exposing the sample to a downward magnetic field of up to 2 T. The direction of magnetisation in the particle is predicted to switch when the vertical field reaches 0.52 $B_K$, where $B_K = 2K/M_s$. The objective lens is then switched off and the sample tilted back to the horizontal.
Figure 5.6 Calculated equilibrium threshold sizes for superparamagnetic, single-domain, vortex and two-domain magnetic states in magnetite particles, shown as a function of particle length and axial ratio. The upper solid line shows the boundary between single-domain and two-domain states. The lower solid lines show the sizes for the onset of superparamagnetic behaviour, with relaxation times of $4 \times 10^5$ years and 100 s. The dashed line shows the boundary between single-domain and vortex states for uniaxial ellipsoidal particles, calculated using finite element micromagnetic methods. Open circles show the sizes and aspect ratios of closely spaced magnetite blocks in an exsolved titanomagnetite inclusion imaged using electron holography by Harrison et al. Reproduced from Ref. 57.

For isolated equidimensional particles, the equilibrium single-domain–single-vortex transition is predicted to occur at a particle size of 70 nm, while the transition to a superparamagnetic state is expected to occur below a particle size of 25–30 nm. The observation of a stable single-domain state in the approximately equidimensional 50 nm diameter crystal shown in Figure 5.4 is in agreement approximately with the expected behaviour.

5.3.2.2 Nanoparticle Rings

An illustration of the characterisation of magnetostatic interactions between particles that each contain single magnetic domains is provided by the
examination of rings of 20 nm diameter crystalline Co particles. Such rings are appealing candidates for high-density information-storage applications because they are expected to form chiral domain states that exhibit flux closure. The magnetisation directions of the present nanoparticle rings cannot be reversed by applying an inplane external field. As a result, phase images were obtained both before and after turning the specimen over. The resulting pairs of phase images were aligned in position and angle, and their sum and difference calculated as described previously. Figure 5.7(a) shows a low-magnification bright-field image of the Co rings. Many self-assembled structures are visible, including five- and six-particle rings, chains and closely packed aggregates. Figures 5.7(b)–(d) show magnetic flux closure states in four different rings, measured using electron holography at room temperature in magnetic-field-free conditions. The magnetic flux lines, which are formed from the cosine of 128 times the magnetic contribution to the measured phase shift, reveal the inplane induction within each ring ensemble. Subsequent electron holography experiments have been used to show that the chirality of each flux closure state can be switched in situ in the TEM by using an out-of-plane magnetic field.

5.3.2.3 Nanoparticle Chains

There are few experimental measurements of the critical sizes at which magnetic particles that are arranged in chains are large enough to support magnetic vortices rather than single domains. Previous electron holography studies of magnetic nanoparticle chains did not provide direct images of such vortex states. Figures 5.8(a) and (b) show the remanent magnetic states of two chains of Fe$_{56}$Ni$_{44}$ particles, whose average diameter of 50 nm is expected to be close to the critical size for vortex formation. For a 75 nm Fe$_{56}$Ni$_{44}$ particle sandwiched between two smaller particles (Figure 5.8(a)), closely spaced contours run along the chain in a channel of width 22 ± 4 nm. A comparison of this result with micromagnetic simulations indicates that the particle contains a vortex, whose axis is parallel to the chain axis, as shown schematically in Figure 5.8(c). In Figure 5.8(b), a vortex can be seen end-on in a 71 nm particle at the end of a chain. The positions of the particle's neighbours determine the handedness of the vortex, with the flux channel from the rest of the chain sweeping around the core to form concentric circles (Figure 5.8(d)). The vortex core, which is perpendicular to the chain axis, is only 9 ± 2 nm in diameter. The larger value of 22 nm observed in Figure 5.8(a) results from magnetostatic interactions along the chain. Figure 5.8(c) shows similar particles with an alloy concentration of Fe$_{0.10}$Ni$_{0.90}$, which contain wider flux channels of diameter ∼ 70 nm, and single-domain states when the particles are below ∼ 100 nm in size. The complexity of such vortex states highlights the importance of controlling the shapes, sizes and positions of closely spaced magnetic nanocrystals for storage and recording applications.

Magnetic-induction maps similar to those shown in Figure 5.8 have been obtained from chains of magnetite (Fe$_3$O$_4$) and greigite (Fe$_3$S$_4$) crystals in
Figure 5.7 (a) Low-magnification bright-field image of self-assembled Co nanoparticle rings and chains that were deposited directly onto an amorphous carbon support film. Each Co particle has a diameter of between 20 and 30 nm. (b)-(e) Magnetic-induction maps showing remanent states in four different nanoparticle rings, in the form of magnetic phase contours (128× amplification; 0.049 rad spacing) formed from the magnetic contribution to the measured phase shift. The mean inner potential contribution to the phase shift has been subtracted from each image (see text for details). The outlines of the nanoparticles are marked in white, while the direction of the measured magnetic induction is indicated both using arrows and according to the colour wheel shown in (f) (red = right, yellow = down, green = left, blue = up). Reproduced from Ref. 63.
magnetotactic bacteria. The magnetic properties of the bacterial iron oxide and sulfide nanocrystals, which are typically 30 to 120 nm in size and arranged in linear chain configurations, depend on crystal structure, morphology, orientation and spacing. For highly aligned chains of crystals, magnetostatic interactions move the boundary between single domain and vortex states to larger particle sizes, and promote the stability of single-domain states. This effect enables bacteria to grow single-domain crystals to larger sizes than would otherwise be possible, optimising the magnetic moment of the chain. Similarly, crystals that are small enough to be superparamagnetic at room temperature, if they were isolated, are constrained to be single domain by magnetostatic interactions with neighbouring crystals in the chain. Shape anisotropy appears to be the most important factor controlling the magnetic microstructure of these bacterial crystals, followed by interparticle interactions, with magnetocrystalline anisotropy being least important. The strict biological control that results in the presence of uniform morphologies, sizes and orientations of magnetite crystals appears to be much weaker in greigite-producing bacteria.
5.3.2.4 Two-dimensional Nanoparticle Arrays

The magnetic behaviour of the rings and chains of nanomagnets described above contrasts with that of a two-dimensional array of closely spaced crystals. Figure 5.9(a) shows a chemical map, obtained using three-window background-subtracted elemental mapping, of a crystalline naturally occurring magnetite-ulvöspinel (Fe₃O₄·Fe₂TiO₄) inclusion in clinopyroxene,⁷¹,⁷² in which the distribution of Fe is shown in blue and Ti in red. The sample exsolved during slow cooling yielding an intergrowth of ferrimagnetic magnetite-rich (Fe₃O₄) blocks separated by paramagnetic ulvöspinel-rich (Fe₂TiO₄) lamellae.⁷³ Magnetic-induction maps obtained from this region, which are shown in Figures 5.9(b) and (c), were recorded by tilting the specimen in zero field and then turning the objective lens on fully to saturate the sample, in order to provide a known starting point from which further fields could be applied. The objective lens was then turned off, the specimen tilted in zero field in the opposite direction and the objective lens excited partially to apply a known inplane field to the specimen in the opposite direction. Finally, the objective lens was switched off and the sample tilted back to 0° in zero field to record each hologram. Figures 5.9(b) and (c) show that the magnetic-domain structure in this sample is both complicated and unexpected. The remanent state of such a system might be expected to involve adjacent blocks being magnetised in an

![Image](image_url)

**Figure 5.9** (a) Chemical map of a finely exsolved naturally occurring titanomagnetite inclusion within pyroxene, acquired using a Gatan imaging filter. The blue regions show the positions of magnetite (Fe₃O₄) blocks, which are separated from each other by paramagnetic ulvöspinel (Fe₂TiO₄) lamellae. (b) and (c) show magnetic-phase contours, measured using electron holography from the same region after magnetising the sample using two different values of the magnetic field provided by the conventional microscope objective lens. The inplane component of the applied field was always vertical on the page. The black contour lines show the direction and magnitude of the projected inplane magnetic induction, which can be correlated with the positions of the magnetite blocks (outlined in white). The direction of the measured induction is also indicated using colours and arrows. Each image was acquired with the specimen in magnetic-field-free conditions. The outlines of the magnetite-rich regions are marked in white, while the direction of the measured magnetic induction is indicated using arrows and colours. Reproduced from Ref. 72.
alternating manner along their elongation directions. However, strong interactions between adjacent magnetite blocks (which are separated by \( \sim 15 \) nm) constrain their remanent magnetisation direction to lie almost perpendicular to their elongation direction and to the applied-field direction. The chains of blocks form superstates that are magnetised parallel to the chain axes but perpendicular to the easy axes of the individual blocks, even though the inplane component of the applied field was parallel to their elongation directions. In an earlier electron holography study, arrays of magnetite blocks in a similar sample that were expected to display single vortex states at remanence were found to be primarily single-domain states.\(^{74}\) The majority of the blocks examined in this study, whose dimensions are plotted in Figure 5.6, would be expected to display single vortex states at remanence if they were isolated and at equilibrium. In an array of strongly interacting particles, the demagnetising energy, which normally destabilises the single-domain state with respect to the vortex state in an isolated particle, is reduced greatly.

It is interesting to note that, in the titanomagnetite system, mean inner potential contributions to the measured phase shift can be removed using a different procedure to that used for the rings and chains of particles described above. Although both thickness and composition vary in the region of interest, the different compositions of magnetite and ulvöspinel are compensated by their densities in such a way that their mean inner potentials are equal. As a result, only a thickness correction may be required. The local specimen thickness across the region of interest can be determined in units of inelastic mean free path by using energy–filtered imaging. This thickness measurement can then be used to determine the mean inner potential contribution to the phase shift, which was in turn used to establish the magnetic contribution to the phase shift.

5.3.2.5 Lithographically Patterned Magnetic Nanostructures

Specimen preparation can present a challenge for electron holography of nanomagnet arrays. In order to examine square arrays of Ni pillars that were fabricated on Si substrates using interferometric lithography,\(^{75}\) specimens were prepared for TEM examination using focused ion beam (FIB) milling so that a single row of pillars could be imaged in cross section.\(^{76}\) In order to protect the pillars from the damaging effects of implantation and physical damage during FIB milling, the sample was first coated with a 200–500 nm layer of amorphous carbon. A 1 \( \mu \)m thick Pt strip was then deposited onto the carbon layer. A single row of pillars was cut out of the array using a 30 kV Ga\(^+\) ion beam by the lift-out method, and placed on a carbon-coated Cu TEM grid. The presence of the layer of amorphous carbon under the protective Pt strip meant that during the final stage of the milling process the Pt fell away from the sample without the need for milling from the top surface. Figure 5.10 shows representative examples of magnetic-induction maps recorded from a single row of 57 nm diameter 115 nm high Ni pillars that were originally fabricated on Si in a square array of side 100 nm. At remanence, the pillars are each in close-to-single-domain magnetic
Electron Holography of Nanostructured Materials

Figure 5.10 Magnetic-induction maps acquired from a single row of Ni pillars prepared using focused ion beam milling from a square array of pillars grown directly onto a Si substrate. Each pillar is 57 nm in diameter, 115 nm in height and separated from its neighbours by 100 nm. The images were acquired in magnetic-field-free conditions, after saturating the pillars downwards by using the conventional TEM objective lens and then applying reverse inplane fields (upwards) of (a) 0 Oe, (b) 450 Oe and (c) 920 Oe. The phase contours have a spacing of 0.049 rad. The mean inner potential contribution to the phase shift has been removed from each image (see text for details). Reproduced from Ref. 76.

states. Pillars that are magnetised parallel to their neighbours appear to show a stronger return flux and a weaker stray magnetic field than pillars that are magnetised antiparallel to their neighbours. Care is clearly required when preparing specimens using FIB milling, as the magnetic moments of the pillars shown in Figure 5.10 are smaller than expected for pure Ni, possibly because of oxidation or damage sustained during specimen preparation. In a similar study, square arrays of Co disks were prepared in plan-view geometry using FIB milling.77

Similar results to those shown in Figure 5.10 have been obtained from a wide range of larger lithographically patterned structures that were prepared directly onto electron-transparent substrates, many of which show multidomain behaviour and complicated switching mechanisms.78-81 Few phase contours are visible outside such elements when they support magnetic flux closure states. Electron holography has also been used to provide information about magnetic
interactions between closely separated ferromagnetic layers within individual spin valve elements. The presence of two different phase contour spacings at different applied fields in such elements is associated with the reversal of the magnetisation directions of the ferromagnetic layers in each element at different fields. The switching field of each layer may also be influenced strongly by magnetostatic interactions between the two layers.

5.3.2.6 Nanowires

An important question relates to the minimum size of a nanostructure in which magnetic fields can be characterised successfully using electron holography. Although 4 nm diameter single-crystalline Co nanowires have been characterised successfully, this measurement is challenging because the mean inner potential contribution to the phase shift at the centre of a 4 nm wire relative to that in vacuum is 0.57 rad (assuming a value for $V_0$ of 22 V), whereas the step in the magnetic contribution to the phase shift across the wire is only 0.032 rad (assuming a value for $B$ of 1.6 T). Although the magnetic contribution to the measured phase shift for an isolated wire was measured successfully in this study, the closely spaced contours along the wire were not straight, resulting from smoothing a signal that is noisy and weak, as also demonstrated in a more recent study.

5.3.2.7 Quantitative Measurements, Resolution and Micromagnetic Simulations

A particular strength of electron holography is its ability to provide quantitative information about magnetic properties. For example, the magnetic moment of a nanoparticle can be obtained from the expression

\[ m_x = \frac{\hbar}{e} \int_{y=-\infty}^{y=+\infty} \int_{x=-\infty}^{x=+\infty} \frac{\partial}{\partial y} \phi_{\text{MAG}}(x,y) \, dx \, dy \]  

(5.16)

where, $\phi_{\text{MAG}}$ is the magnetic contribution to the phase shift and $y$ is a direction perpendicular to $x$ in the plane of the specimen. According to eqn (5.16), the magnetic moment in a given direction can be obtained by measuring the area under the first differential of $\phi_{\text{MAG}}$ evaluated in the perpendicular direction. The contribution of stray magnetic fields to the moment is included in this calculation if the integration is carried out over a large enough distance from the particle.

The spatial resolution that can be achieved in a phase image is determined primarily by the spacing of the holographic interference fringes. However, the contrast of these fringes decreases as their spacing is reduced, and the recording process is also dominated by Poisson-distributed shot noise. These parameters are affected by the illumination diameter, exposure time and biprism voltage. The final phase resolution and spatial resolution are therefore
Electron Holography of Nanostructured Materials

inherently linked, as a small phase shift can be measured with high precision and poor spatial resolution, or with poor precision and high spatial resolution. In each of the examples described above, the recorded phase images were smoothed slightly to remove noise, and the spatial resolution of the magnetic information is typically between 10 and 20 nm. This procedure is necessarily subjective, and care is required to ensure that artifacts are not introduced. Higher spatial and phase resolution may also be achieved by recording several holograms of each area of interest and subsequently averaging the resulting phase images.

It is therefore necessary to compare electron holographic measurements with micromagnetic simulations due to the sensitivity of the magnetic domain structure in nanoscale materials and devices to their detailed magnetic history and to their microstructure and chemistry. Differences between starting magnetic states that are too small to be distinguished visually, as well as inter-element coupling and the presence of out-of-plane magnetic fields, are important for the formation of subsequent domain states, and in particular for determining the sense (the handedness) with which magnetic vortices unroll. The sensitivity of the domain structure to such effects, as well as to the three-dimensional physical and chemical microstructure of the specimen, emphasises the need to correlate high-quality experimental holographic measurements with micromagnetic simulations.

5.3.2.8 Cross-sectional Layered Specimens

One of the most challenging problems for electron holography of magnetic materials is the quantitative measurement of the magnetic properties of nanoscale magnetic layers when examined in cross section. In such samples, the weak magnetic signal must be separated from rapid and unknown variations in both composition and thickness. By rearranging eqns (5.8) and (5.12), it can be shown that specimen thickness effects may be eliminated by calculating the difference between the phase gradients of images, between which the magnetisation has been reversed, divided by the average of their phases and multiplied by a constant and by the value of the mean inner potential of each magnetic layer separately. Formally, this procedure can be written as

$$\frac{C_{\text{g}}\delta V_0(x,y)}{e} \left\{ \frac{\Delta[\phi(x,y)]}{\phi(x,y)} \right\} = \frac{\Delta B_\perp(x,y)}{(1 - (e/C_{\text{g}}\delta V_0(x,y)))\left\{ \frac{\int B_\parallel(x,y)I(x,y)dx}{I(x,y)} \right\}}$$

(5.17)

According to eqn (5.17), by combining appropriate phase profiles and their gradients (evaluated in a direction perpendicular to the layers), both the magnitude and the sign of $\Delta B_\perp(x,y) = 2B_\perp(x,y)$ are obtained exactly if the magnetisation reverses exactly everywhere in the sample (the denominator on the right-hand side of the equation is then unity). Furthermore, nonzero values are returned only in regions where the magnetisation has changed. Equation
(5.17) has been applied successfully to the characterisation of a cross-sectional magnetic tunnel junction sample.\textsuperscript{99} Similar approaches have also been used to remove specimen thickness and mean inner potential contributions from phase shifts measured from holograms of La$_{x}$Ca$_{1-x}$MnO$_{3}$ acquired at different temperatures.\textsuperscript{90,91}

5.4 Measurement of Electrostatic Fields

In this section, the application of electron holography to the characterisation of electrostatic fields is reviewed. Examples are taken from the characterisation of fringing fields outside biased nanowires, dopant contrast at depletion layers in semiconductors and the characterisation of interfaces at which both charge redistribution and changes in chemistry are possible.

5.4.1 Electrically Biased Nanowires

Early experiments on tungsten microtips demonstrated that electron holography could be used to measure electrostatic fringing fields in biased samples.\textsuperscript{92} Further studies were carried out on pairs of parallel 1 \( \mu \)m diameter Pt wires held at different potentials\textsuperscript{93} and on single conducting wires.\textsuperscript{94} More recent experiments have involved the use of electron holography to map electrostatic potentials around the ends of electrically biased carbon nanotubes. In one such study, a three-axis manipulation electrode was used to position a multi-walled carbon nanotube approximately 6 \( \mu \)m from a gold electrode.\textsuperscript{95} Contoured phase images recorded before a bias was applied between the nanotube and the electrode showed a featureless phase gradient profile around the nanotube, whereas closely spaced contours were observed around the nanotube tip when a voltage of up to 120 V was applied. The threshold voltage for field emission was observed to be approximately 70 V. Comparisons with simulations provided a value of 1.22 V nm$^{-1}$ for the electric field at the nanotube tip when the bias voltage was 120 V. This field was stable over time, even when the emission current varied. Results obtained from a similar experiment are shown in Figure 5.11. Figure 5.11(a) shows a design drawing of a side-entry TEM specimen holder that allows specimens to have up to three electrical contacts applied to them \textit{in situ} in the TEM, while allowing tilt angles in excess of 70° to be used for electron tomography. The end of the holder, which contains a removable cartridge that allows two independent electrical contacts to be made to the specimen, is shown in the inset to Figure 5.11(a). A third electrical contact can be moved towards the specimen using micrometers and piezoelectric drives. The removable cartridge is compatible with a FIB workstation, and allows the same specimen to be examined under an applied bias in a scanning electron microscope. Figure 5.11(b) shows a defocused bright-field image of bundles of single-walled carbon nanotubes that were placed in the cartridge shown in Figure 5.11(a). The application of a voltage resulted in attraction of the nanotubes towards a gold electrode and deflection of the incident electrons.
Figure 5.11  (a) Design drawing of an ultrahigh-tilt three-contact cartridge-based electrical biasing nanopositioning TEM specimen holder, with coarse and fine three-axis motion of the moveable contact provided by micrometers and piezoelectric crystals, respectively. The inset shows the location of the cartridge and the position and motion of the third contact. (b) Defocused bright-field image of a specimen containing bundles of single-walled carbon nanotubes, placed in the cartridge shown in (a), with a voltage applied between the tubes and a gold needle that was brought to within 1–2 μm of them in the nanopositioning specimen holder. (c) Contoured phase images obtained from an off-axis electron hologram of a carbon nanotube bundle that has a voltage applied to it in situ in the TEM. Reproduced from Ref. 96.

by the electric field at the end of each nanotube bundle. Figure 5.11(c) shows a contoured holographic phase image of the end of a nanotube bundle. The electric field is strongest where the contours are most closely spaced at the end of the bundle.96

Under certain circumstances, it may be useful to apply a voltage to a sample to separate a parameter of interest from other, unwanted contributions to the contrast.96 An example of how this separation may be achieved is shown in Figure 5.12. Figure 5.12(a) shows a holographic amplitude image of two needles, one of Fe and the other of W, which have been brought to a separation of 560 nm using the nanopositioning specimen holder shown in Figure 5.11(a). The acquisition of phase images with different voltages applied between the two
Figure 5.12 (a) Electron holographic amplitude image of the ends of Fe and W needles, which have been positioned with a separation of 560 nm using the nanopositioning specimen holder. (b) Phase contours measured using electron holography showing the inplane component of the magnetic induction integrated in the electron-beam direction. The contour spacing is $2\pi$ rad. (c) Phase contours arising from the projected electrostatic potential alone, integrated in the electron-beam direction, with the W needle at +5 V with respect to the Fe needle. The contour spacing is $6\pi$ rad. (d) As for (c) but with the W needle at −5 V. The slight asymmetry about the horizontal axis in the contours shown in (c) and (d) almost certainly arises because of the long-range electrostatic field, which perturbs the wave that is overlapped onto the sample to form the hologram. Reproduced from Ref. 96.

needles allows the contribution to the phase shift from the electric field between the needles to be separated from magnetic and mean inner potential contributions to the contrast, as well as from any diffraction contrast that may be present (Figure 5.12).

5.4.2 Dopant Potentials in Semiconductors

Off-axis electron holography promises to fulfill the requirements of the semiconductor industry by providing a technique that can be used to provide quantitative information about dopant potentials in semiconductors with nanometer spatial resolution. In a specimen of uniform thickness, the measured phase shift is expected to provide a quantitative measure of the variation in potential associated with the presence of dopant atoms. Attempts to tackle this problem have been made since the 1960s using many forms of electron interferometry, both experimentally$^{97,98}$ and theoretically.$^{99,100}$ However, TEM specimen preparation is now known to have a profound influence on phase shifts measured from doped semiconductors. In addition to surface depletion resulting from the presence of the specimen surfaces, the electrostatic potential in the specimen may be affected by oxidation, physical damage and the
implantation of Ar and Ga during preparation of the sample for electron microscopy, as well as by irradiation by high-energy electrons during TEM examination. The effects of specimen preparation, and in particular the electrical state of near-surface regions, may account for many of the anomalous results in early experiments. However, recent studies indicate that it may be possible to resolve these problems.

The first unequivocal demonstration of two-dimensional mapping of the electrostatic potential in an unbiased doped semiconductor using electron holography was achieved for metal-oxide-semiconductor (MOS) Si transistors.\textsuperscript{101} The source and drain regions were visible in phase images with a spatial resolution of 10 nm and an energy resolution of close to 0.10 eV. Differential thinning was discounted as a cause of the observed phase shifts and an optimal specimen thickness of 200–400 nm was identified for such experiments. The transistors were prepared for TEM examination using conventional mechanical polishing and Ar-ion milling. A 25 nm thick electrically altered layer was identified on each surface of the specimen, which resulted in measured phase shifts across each p-n junction that were lower than predicted. More recently, electron holography studies of transistors have been compared with process simulations.\textsuperscript{102} In this study, specimens were prepared primarily using tripod wedge polishing, followed by limited low-angle Ar-ion milling at 3.5 kV. Surprisingly, no electrically altered surface layer had to be taken into account to quantify the results.

The electrostatic potential across an abrupt p-n junction is shown schematically in Figures 5.13(a) and (b). In Figure 5.13(a), the effect of the unknown electrical state of the specimen surface on the potential is included phenomenologically by assuming the presence of electrically altered layers of uniform thickness on the specimen surfaces. The true junction potential is then assumed to lie within specimen thickness $t_{ds}$, which is smaller than the total specimen thickness, $t$. The graph in Figure 5.13(b) is drawn on the assumption that the “transition regions” on each side of the depletion region are negligibly small—an assumption that is unlikely to be true in practice, but that can be assessed experimentally by calculating the charge density profile from a measured phase profile. It should be noted that care is required both with specimen preparation and with interpretation of measured phase shifts because a small change in specimen thickness (of 5–10%) can result in similar contrast to that across a p-n junction.

Recently, electrostatic potential profiles have been measured from reverse-biased Si p-n junctions that were prepared for TEM examination using FIB milling.\textsuperscript{103–106} FIB milling is the technique of choice for preparing TEM specimens from site-specific regions of integrated circuits. Specimens for in situ electrical biasing were prepared by using a 30 kV FEI 200 FIB workstation to machine parallel-sided electron-transparent membranes at the corners of 1 × 1 mm 90° cleaved squares of wafer. This geometry allowed electrical contacts to be made to the front and back surfaces of each specimen using a modified single-tilt holder.\textsuperscript{103} Care was taken to expose the region of interest to the focused beam of Ga ions only at a glancing angle to its surface.
Figure 5.13 (a) Schematic diagram showing the cross-sectional geometry of a TEM specimen of uniform thickness that contains a symmetrical semiconductor $p$-$n$ junction. $t_{ad}$ is the "electrically active" specimen thickness and $W$ is the width of the depletion region over which the potential changes. The layers at the top and bottom surfaces of the specimen represent electrically passivated or depleted layers, whose physical and electrical nature is affected by TEM specimen preparation. (b) Schematic diagram of the electrostatic potential profile across the $p$-$n$ junction. $V_{bi}$ is the built-in voltage. The sign convention for the potential is consistent with the mean inner potential of the specimen being positive relative to vacuum. (c) Representative phase image reconstructed from an off-axis electron hologram of a FIB-milled Si $p$-$n$ junction sample. The sample edge is at the lower right of the image. No attempt has been made to remove the phase "wraps" lying along this edge. (d) and (e) Line profiles of the measured phase shift across the Si $p$-$n$ junction, shown as a function of (d) specimen thickness for three different unbiased FIB-milled "trench" specimens and (e) reverse-bias voltage for a single specimen whose crystalline thickness was measured to be 390 nm. The phase images were averaged over a distance of approximately 100 nm on the specimen to form the profiles, in which the zero of phase has been chosen to lie on the left (the $p$-) side of the junction. (f) Schematic diagram showing, in cross section, the physical and electrical structure of a FIB-milled TEM specimen inferred from results such as those shown in (d) and (e). Reproduced from Ref. 104.
Figure 5.13 (Continued)

Figure 5.13(c) shows a representative phase image recorded from an unbiased Si p-n junction sample, whose thickness was measured to be 550 nm using convergent beam electron diffraction. The p-type and n-type regions are delineated as areas of darker and lighter contrast, respectively. The additional “grey” band at the specimen edge is likely to be associated with the presence of an electrically altered layer, which is visible in cross section but is thought to extend around the entire specimen surface. Line profiles across the junction were obtained from phase images acquired with different reverse bias voltages applied to a specimen of 390 nm crystalline thickness (Figure 5.13(d)), as well as from several unbiased specimens (Figures 5.13(d) and (e)). Each profile is qualitatively consistent with the expected potential profile for a p-n junction in a specimen of uniform thickness.\(^{167}\) In Figure 5.13(e) the height of the potential step across the junction, \(\Delta \phi\), increases linearly with reverse bias voltage \(V_{\text{applied}}\) according to the equation

\[
\Delta \phi = C_E (V_{\text{bi}} + V_{\text{applied}}) t_{\text{el}}
\]  

(5.18)

where \(C_E\) is defined in eqn (5.6) and the p-n junction is contained in an electrically active layer of thickness, \(t_{\text{el}}\), in a specimen of total thickness, \(t\). Analysis indicates that 25 ± 5 nm of the crystalline thickness on each surface...
of the TEM specimen is electrically altered. Depletion widths across the junction measured from the line profiles are higher than expected, suggesting that the electrically active dopant concentration in the specimen is lower than it’s nominal value. External electrostatic fringing fields were never observed outside any FIB-milled specimens, indicating that their surfaces are equipotentials in both unbiased and biased specimens. Analysis of the results shown in Figures 5.13(c)-(e) suggests that a layered structure may be present in the TEM membrane, with amorphous outer surface layers surrounding inner, crystalline electrically altered surface layers, themselves surrounding crystalline electrically active material, as shown schematically in Figure 5.13(f).

Figure 5.14(a) shows a holographic phase image of a GaAs p-n junction that was prepared for TEM examination using FIB milling. In contrast to results obtained from FIB-milled Si specimens of similar thickness, such as that shown

![Figure 5.14](image)

**Figure 5.14** (a) and (b) Wrapped electron holographic phase images of a focused ion beam milled GaAs p-n junction of crystalline thickness 240 nm, recorded at room temperature (a) before and (b) after in situ annealing the specimen at 500°C for 30 min. (c) Phase profiles across a focused ion beam milled GaAs p-n junction in a specimen of crystalline thickness 300 nm, measured both before (open circles) and after (closed circles) annealing at 500°C. Reproduced from Ref. 108.
in Figure 5.13(c), the step in phase across the GaAs junction is indistinct and
the phase image is noisy. Figures 5.14(b) and (c) show that in situ annealing of
this specimen in the TEM can be used to increase the phase shift across the
junction, while at the same time decreasing noise in the recorded phase image.
These results suggest that annealing can be used to remove defects resulting
from Ga⁺ implantation and to reactivate dopant atoms in the thin specimen. A
similar, although smaller, improvement is seen for FIB-milled Si specimens.108
Although Figure 5.14(a) shows that in situ annealing improves recorded phase
images and decreases the thickness of the electrically inactive surface layers, the
measured built-in voltage across the junction, \( V_{bi} \), is still incorrect. Recent work
has shown that \( V_{bi} \) may be improved for FIB-prepared GaAs specimens by
taking care in setting the incident electron beam current during examination
(and therefore to the rate at which charge is dissipated from the area of interest)
and by providing high-quality electrical contacts to the region of interest. All of
these factors, in combination with simulations of electrostatic potentials in TEM
specimens,109,110 must be understood in order to develop electron holo-
graphy into a technique that can be used to characterise semiconductor dopant
potentials reliably.111–114 Details about the use of electron holography to map
transistor structures reliably have recently been presented.115,116

Charging effects during electron irradiation of a semiconductor can be seen
directly in two dimensions in Figure 5.15, which shows contoured phase images
acquired from a specimen that contains a linear array of transistors located
several \( \mu \)m below the surface of a wafer and separated from its surface by
metallisation and oxide layers. Conventional “trench” FIB milling was used to
prepare the specimen, with a nominal thickness of 400 nm. Figure 5.15(a)
shows eight-times-amplified phase contours obtained from the edge of the
specimen, which contains an array of tungsten contacts separated by regions of
silicon oxide. Instead of the expected phase distribution, which should be
proportional to the mean inner potential multiplied by the specimen thickness,
eliptical contours are visible in each silicon oxide region, and an electrostatic
fringing field is present outside the specimen (at the top of Figure 5.15(a)). Both
the elliptical contours and the fringing field are associated with the build-up of
positive charge in the oxide layers. Figure 5.15(b) shows a similar phase image
obtained after coating the specimen on one side with approximately 20 nm of
carbon. The effects of charging are now absent, there is no fringing field outside
the specimen edge and the phase contours follow the change in specimen
thickness. If the charge is assumed to be distributed through the thickness of
the specimen, then the electric field in the oxide is approximately 2 \( \times 10^7 \) V/m.
This value is just below the breakdown electric field107 for thermal SiO₂ of
10⁸ V m⁻¹. If FIB milling from the substrate side of the wafer is used to prepare
the specimen, then charging no longer occurs, presumably as a result of Si
redepsoition onto the specimen surface.117

Although questions still remain about phase contrast observed at simple
\( p-n \) junctions, electron holographic data have recently been obtained from more
complicated semiconductor device structures, in which changes in
composition as well as doping concentration are present, such as a strained
Figure 5.15 (a) Eight-times-amplified phase contours (0.785 rad spacing) obtained from a cross-sectional semiconductor device specimen of nominal thickness 400 nm prepared using conventional "trench" focused ion beam milling. The amorphous layers on either side of the W contact are formed from Si oxides of different densities. Charging of the oxide layers results in the presence of electrostatic fringing fields in the vacuum region outside the specimen edge, as well as elliptical phase contours within the oxide layers between adjacent W contacts. (b) shows an equivalent phase image obtained after coating the specimen on one side with approximately 20 nm of carbon to remove the effects of charging. The phase contours now follow the expected mean inner potential contribution to the phase shift in the oxide layers, and there is no electrostatic fringing field outside the specimen edge. Reproduced from Ref. 35.

\[ n\text{–Al}_{0.1}\text{Ga}_{0.9}\text{N/ In}_{0.1}\text{Ga}_{0.9}\text{N/ p–Al}_{0.1}\text{Ga}_{0.9}\text{N} \] heterojunction diode, in which strong piezoelectric and polarisation fields are used to induce high two-dimensional electron gas concentrations and internal electrostatic potentials across InGaN quantum wells with thicknesses ranging from 2 to 10 nm.

5.4.3 Space-Charge Layers at Grain Boundaries

Electron holography has also been used to characterise space-charge layers at doped and undoped grain boundaries in electroceramics. At such boundaries, several contributions to the electron holographic phase shift can complicate interpretation. The space-charge distribution that is predicted to form at such a grain boundary is often described as a double (back-to-back) Schottky barrier. For Mn-doped and undoped grain boundaries in SrTiO$_3$, a decrease
in the measured phase shift at the boundary relative to that in the specimen was observed using electron holography.\textsuperscript{121} The changes in phase measured at the doped boundaries were larger in magnitude and spatial extent than at similar undoped boundaries. Possible contributions to the contrast from changes in density, composition, specimen thickness, dynamical diffraction and electrostatic fringing fields were considered and the remaining contributions to the measured phase shifts at the doped boundaries were attributed to space charge. The results were finally interpreted in terms of a narrow (1–2 nm) region of negative grain boundary charge and a wider (3–5 nm) distribution of positive space charge. Defocus contrast has been used\textsuperscript{122} to assess possible space-charge contributions to electrostatic potential profiles across grain boundaries in doped and undoped SrTiO$_3$. The contrast observed in these experiments was not consistent with a dominant contribution to the signal from space charge. In similar electron holography studies, variations in electrostatic potential at undoped and Ca-doped grain-boundary dislocations in YBa$_2$Cu$_3$O$_{7-\delta}$ were studied\textsuperscript{123} and space charge layers with widths of approximately 150 nm were measured at grain boundaries in ZnO.\textsuperscript{124} In an earlier study, defocus contrast recorded from delta-doped layers in Si and GaAs was also attributed to the presence of space charge.\textsuperscript{125} Related experiments have been performed\textsuperscript{126,127} to measure polarization distributions across domain boundaries in ferroelectric materials such as BaTiO$_3$ and PbTiO$_3$. There are many opportunities for further work in this area.

5.5 High-Resolution Electron Holography

Aberrations of the objective lens, which result in modifications to the amplitude and phase shift of the electron wave, rarely need to be taken into account when characterising magnetic and electrostatic fields at medium spatial resolution, as described in Sections 5.3 and 5.4. However, these aberrations must be considered when interpreting electron holograms that have been acquired at atomic resolution, in which lattice fringes are visible.

The back focal plane of the objective lens contains the Fraunhofer diffraction pattern, i.e. the Fourier transform, of the specimen wave $\psi_s(r) = A_s(r) \exp[i\phi_s(r)]$, denoted $\psi(q) = \text{FT} [\psi_s(r)]$. Transfer from the back focal plane to the image plane is then represented by an inverse Fourier transform. For a perfect thin lens, neglecting magnification and rotation of the image, the complex image wave would be equivalent to the object wave $\psi(r)$. Modifications to the electron wave that result from objective-lens aberrations can be represented by multiplication of the electron wavefunction in the back focal plane by a transfer function of the form

$$T(q) = B(q) \exp[iZ(q)] \quad (5.19)$$

In eqn (5.19), $B(q)$ is an aperture function that takes a value of unity for $q$ within the objective aperture and zero beyond the edge of the aperture. The
effects of two primary objective lens aberrations, defocus and spherical aberration, are included in the phase factor in the form

$$\chi(q) = \pi \Delta z \lambda q^2 + \frac{\pi}{2} C_s \lambda^2 q^4$$  \hspace{1cm} (5.20)

where $\Delta z$ is the defocus of the lens and $C_s$ is the spherical aberration coefficient. The complexity of eqn (5.20) increases rapidly as further aberrations are included. The complex wave in the image plane can then be written in the form

$$\psi_i(r) = \text{FT}^{-1}\left[\text{FT}[\psi_s(r)] \times T(q)\right]$$  \hspace{1cm} (5.21)

$$= \psi_s(r) \otimes t(r)$$  \hspace{1cm} (5.22)

where $t(r)$ is the inverse Fourier transform of $T(q)$, and the convolution $\otimes$ of the specimen wave $\psi_s(r)$ with $t(r)$ represents the smearing of information that results from lens imperfections. Since both $\psi_s(r)$ and $t(r)$ are in general complex, the intensity of a conventional bright-field image, which can be expressed in the form

$$I(r) = |\psi_s(r) \otimes t(r)|^2$$  \hspace{1cm} (5.23)

is not related simply to the structure of the specimen.

The effects of lens aberrations can be removed by multiplying the complex image wave by a suitable phase plate corresponding to $T^*(q)$ to provide the amplitude and the phase shift of the specimen wave $\psi_s(r)$ rather than the image wave $\psi_i(r)$. In this way, the interpretable resolution of the image can be improved beyond the point resolution of the electron microscope. The optimal defocus that maximises the resolution of the reconstructed specimen wave after correction of aberrations\textsuperscript{128-131} is given by the expression

$$\Delta z_{\text{opt}} = -\frac{3}{4} C_s (\lambda q_{\text{max}})^2$$  \hspace{1cm} (5.24)

where $q_{\text{max}}$ is the maximum desired spatial frequency.

Figure 5.16 illustrates the application of aberration correction to a high-resolution electron hologram of crystalline Si imaged along the $[110]$ zone axis, at which characteristic "dumbbell" contrast, of spacing 0.136 nm, is expected.\textsuperscript{132} The original hologram was acquired using an interference fringe spacing of 0.05 nm on a CM30 FEGTEM, which has a point resolution of 0.198 nm and an information limit of 0.1 nm at 300 kV. Figures 5.16(a) and (b) show, respectively, the reconstructed amplitude and phase shift of the hologram after aberration correction using a phase plate. The phase image reveals the expected white "dumbbell" contrast, at a spatial resolution that is considerably better than the point resolution of the microscope, after lens aberrations, including residual astigmatism and off-axis coma, have been measured and removed. The projected atom column positions are visible as dark contrast in the amplitude image. High-resolution electron holography is clearly an exciting area of research, with many recent developments and applications of the technique to a wide range of materials problems.\textsuperscript{133,134}
Figure 5.16 High-resolution (a) amplitude and (b) phase images of the aberration-corrected specimen wave reconstructed from an electron hologram of [110] Si, obtained at 300 kV on a CM30 FEGTEM. The spacing of the holographic fringes was 0.05 nm. The sideband contains [111], [220], [113] and [004] reflections, corresponding to lateral information of 0.136 nm. The characteristic Si dumbbell structure is visible only after aberration correction. Reproduced from Ref. 132.

5.6 Alternative Forms of Electron Holography

Many different forms of electron holography can be implemented both in the TEM and in the Scanning TEM (STEM). There are also several ways in which the off-axis mode of TEM electron holography can be implemented. A full discussion of these various schemes, which include interferometry in the diffraction plane of the microscope, and reflection electron holography, is beyond the scope of this chapter. However, in this section, a few of the more important developments in this area are reviewed.

The need for a vacuum reference wave is a major drawback of the standard off-axis mode of TEM holography since this requirement restricts the region that can be examined to near the specimen edge. In many applications, the feature of interest is not so conveniently located. The implementation of a Differential Phase Contrast (DPC) mode of electron holography in the TEM enables this restriction to be overcome. DPC imaging is well established as a technique in the STEM, involving the use of various combinations of detectors to obtain magnetic contrast. DPC contrast can also be obtained using far-out-of-focus STEM electron holography (see below). An equivalent TEM configuration can be achieved by using an electron biprism located in the condenser aperture plane of the microscope. The hologram is acquired under out-of-focus conditions, and is in effect the superposition of a pair of Fresnel images. For characterisation of both components of the inplane induction without removing the sample from the microscope, a rotating biprism or a rotating sample holder is required.

An alternative scheme that is conceptually similar to the differential mode of electron holography in the TEM, but which does not require the use of an
electron biprism or a field-emission electron gun, is termed amplitude-division electron holography. Division of the amplitude of the electron wave can be achieved by using a crystal film located before the specimen. The lattice fringes of the crystal film are then used as carrier fringes. The specimen can be inserted into the normal object plane by placing a single-crystal thin film and the sample of interest on top of each other. The single crystal film is tilted to a strong Bragg condition and used as an electron-beam splitter. The hologram plane contains two defocused images of the specimen that are shifted laterally with respect to one another. One of these images is carried by the direct beam and the other by the Bragg-reflected beam. When the distance between the two images is greater than the size of the object, the images separate perfectly and interfere with adjacent plane waves to form an off-axis electron hologram. The defocus of the object can be corrected at the reconstruction stage by using a phase plate, although high coherence of the incident illumination is then required. The coherence used when forming the image therefore determines the spatial resolution of the final reconstructed image.

An approach that can be used to increase the sensitivity of electron holography is termed phase-shifting electron holography. This approach is based on the acquisition of several off-axis holograms while the phase offset (the initial phase) of the image is changed. Electron holograms are recorded at successive values of the incident-beam tilt, such that the phase is shifted by at least $2\pi$ over the image series. The advantages of the phase-shifting approach are greatly improved phase sensitivity and spatial resolution. Care is required if the object is out of focus, as tilting the beam will also induce an image shift between successive images. Very small phase shifts have been observed from individual unstained ferritin molecules using this approach.

A real-time approach for acquiring and processing holograms has been demonstrated by using a liquid-crystal panel to reconstruct holograms. Holograms were recorded at video rate and transferred to a liquid-crystal spatial light modulator located at the output of a Mach–Zender interferometer. The liquid crystal panel was illuminated using a He-Ne laser, and interference micrographs were observed at video rate on the monitor beside the microscope as the specimen was examined. In an alternative configuration, a liquid-crystal panel was used as a computer-controlled phase plate to correct for aberrations.

Whereas an off-axis electron hologram is formed by the interference of an object and a reference wave that propagate in different directions in the electron microscope, the simplest way of recording an electron hologram without using an electron biprism involves using the transmitted wave as the reference wave to form an inline hologram. Gabor’s original paper described the reconstruction of an image by illuminating an inline hologram with a parallel beam of light and using a spherical aberration correcting plate and an astigmatism corrector. The reconstructed image is, however, disturbed by the presence of a “ghost” or “conjugate” twin image. If the hologram is recorded and subsequently illuminated by a plane wave, then the reconstructed image and a defocused conjugate image of the object are superimposed on each other. The most effective method of separating the twin images is to use Fraunhofer
inline holography. Here, inline holograms are recorded in the Fraunhofer
diffraction plane of the object. Under this condition, the conjugate image is so
blurred that its effect on the reconstructed image is negligible.\textsuperscript{145}

The STEM holographic mode used for DPC imaging, which has similarities
with the TEM differential mode of electron holography described above, is a
point projection technique in which a stationary beam in a STEM is split by a
biprism preceding the sample so that two mutually coherent electron point
sources are formed just above the specimen. In this operating mode, the
objective lens is excited weakly so that the hologram is formed in the diffraction
plane rather than the image plane.\textsuperscript{146} By defocusing the objective lens greatly, a
shadow image of the object is formed, which has the appearance of a TEM
hologram, although it is distorted by spherical aberration and defocus. The
image magnification and the separation of the sources relative to the specimen
are flexible in this configuration, and can be adjusted by changing the biprism
voltage and/or the objective or post-specimen lens settings. The far-out-of-focus
mode of STEM holography has been applied to the characterisation of a range
of magnetic materials.\textsuperscript{147}

A rapid approach that can be used to visualise equiphase contours involves
superimposing a hologram of the specimen onto a vacuum hologram acquired
under identical conditions, with the specimen removed from the field of view.\textsuperscript{148}
Interference between the holographic fringes in the two images then provides
widely spaced, low-contrast bands that reveal phase contours directly and by
defocusing the combined image, the unwanted finely spaced holographic
fringes can be removed.

5.7 Discussion, Prospects for the Future and
Conclusions

In this chapter, the technique of off-axis electron holography has been
described, and its application to a variety of materials has been reviewed.
A selection of results has been presented from the characterisation of magnetic
fields in arrangements of closely spaced nanocrystals and patterned elements, to
electrostatic fields in field emitters and semiconductors. \textit{In situ} experiments,
which allow magnetisation reversal processes to be followed and electrostatic
fields in working semiconductor devices to be characterised, have been
described, and the advantages of using digital approaches to analyse electron
holograms have been highlighted. High-resolution electron holography and
alternative modes of holography have also been described. Although the results
that have been presented are specific to the dimensions and morphologies of the
examples chosen, they illustrate the ways in which holography can be adapted
to tackle a range of materials problems.

Future developments in electron holography are likely to include the develop-
ment and application of new forms of electron holography and instrumenta-
tion, the introduction of new approaches for enhancing weak signals and the
formulation of a better understanding of the effect of sample preparation on phase images recorded from semiconductors and ferroelectrics.

A particularly exciting prospect involves the combination of electron holography with electron tomography to image electrostatic and magnetic fields inside nanostructured materials in three dimensions rather than simply in projection. This approach has been used to image magnetic fringing fields outside materials in three dimensions, by acquiring two ultrahigh-tilt series of electron holograms about orthogonal axes. If each phase image is differentiated in a direction perpendicular to the tilt axis, then standard tomographic reconstruction algorithms can be used to calculate the three-dimensional distribution of the component of $B$ that lies parallel to the tilt axis, based on the relations

$$\frac{d}{dy} \phi(x,y) = -\left(\frac{e}{\hbar}\right) \int_{z=-\infty}^{z=+\infty} B_z(x,y,z) \, dz \quad (5.25)$$

and

$$\frac{d}{dx} \phi(x,y) = +\left(\frac{e}{\hbar}\right) \int_{z=-\infty}^{z=+\infty} B_x(x,y,z) \, dz \quad (5.26)$$

After determining $B_x$ and $B_y$ in three dimensions in this way, $B_z$ can be inferred by making use of the criterion that $\nabla \cdot B = 0$. The application of this approach to the characterisation of magnetic fields inside nanostructured materials is complicated by the fact that the (often dominant) mean inner potential contribution to the measured phase shift must be removed at each sample tilt angle. This requirement can be achieved if each tilt series is recorded both before and after reversing the direction of magnetisation in the specimen (e.g., using the microscope objective lens). Subsequently, half of the difference between pairs of reversed images acquired at each tilt angle can be used to provide the magnetic contribution to the phase shift. Four tilt series of holograms are therefore required. The fact that eqns (5.25) and (5.26) are expected to hold can be illustrated analytically for a uniformly magnetised sphere. The region of interest must clearly lie close to a large enough hole to allow electron holograms to be acquired at high sample tilt angles about two axes, without either the region of interest or the hole being shadowed by other parts of the specimen. The difficulty of finding such a region means that the distribution of crystals that is imaged should be isolated and small, so that the magnetic signal from the region of interest decreases to close to zero at the edges of the field of view.

In conclusion, the unique capability of electron holography to provide quantitative information about magnetic and electrostatic fields in materials at a resolution approaching the nanometer scale, coupled with the increasing availability of field-emission-gun transmission electron microscopes and quantitative digital recording, ensure that the technique has a very promising future.


Acknowledgements


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