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Imaging of weak phase objects by a Zernike phase plate

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ABSTRACT

Analysis of the imaging of some simple distributions of object phase by a phase plate of Zernike type shows that sharp transitions in the object phase are well transmitted. The low-frequency components of the complete object function are attenuated by the plate. The behaviour can be characterised by a cut-on parameter defined as the product of the cut-on frequency of the plate and a characteristic dimension of the object. When this parameter exceeds a value of the order of unity, a sharp boundary in the object is imaged by a Zernike plate as a dark lining inside the boundary with a white outline or halo outside the boundary, in agreement with reported observations. The maximum diameter of objects that can be imaged accurately is inversely proportional to the diameter of the hole for beam transmission in the phase plate.

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1. Introduction

Phase plates are currently of interest in transmission electron microscopy (TEM) for improving the imaging of weak phase objects. When inserted at a suitable position in the column, a phase plate can change the phase of a specific range of spatial frequencies and can provide maximum image contrast for these frequencies on passing through focus, thereby eliminating their contrast reversal and simplifying the interpretation of images. Use of a phase plate is also expected to reduce the electron dose needed for imaging and thus reduce specimen damage.

One frequently used type of plate is rotationally invariant and is described as having Zernike geometry. A simple form of Zernike plate consists of a thin sheet of material of known mean inner potential and provided with a central hole of radius r_1 somewhat greater than that of the unscattered electron beam. The thickness of the plate is chosen to change the phase of the scattered electrons by the desired amount relative to the unscattered beam, as described by Danev and Nagayama [1]. This type of plate has an outer boundary at the maximum aperture that is convenient but has no further structure. Other possible ways have been demonstrated for producing a change of phase that is independent of rotation around the axis. One method uses electrodes arranged so that direct and scattered electrons see different potential distributions as they pass through the structure. The electrical lengths for the two paths differ, providing a phase difference which can be varied in operation, as described for example by Schultheiss et al. [2]. Another method being investigated uses a thin ring carrying magnetic flux which produces a phase difference by the Aharonov–Bohm effect [3]. Such a ring can either advance or retard the phase of scattered electrons, depending on the direction of circulation of flux which can be reversed by turning over the ring in its holder. In principle, a Zernike plate provides the desired phase difference at radii down to some minimum value r_2. For the simple sheet, r_2 coincides with r_1 but for other types of plate the two radii differ. Electrons passing the plate at radii between r_1 and r_2 may be intercepted, and those at radii less than r_1 will not be changed in phase.

Phase or amplitude contrast transfer functions show how the transmission varies with frequency, but do not reveal the result of imaging an object that superimposes many spatial frequencies. By considering the imaging of strong phase objects, Beleggia has shown [4] that the optimum phase change for the plate is a function of object phase shift. Further detailed simulations [5,6] have modelled strong phase objects and the transfer function of the objective lens. The purpose of the present paper, in contrast, is to obtain analytic results for the imaging of weak phase objects by the phase plate alone, omitting any absorption or defects of lenses. Although the objects to be used are much simpler than real biological specimens, the analytical descriptions allow the change of image with object size to be demonstrated clearly. The results not only confirm that objects that are sufficiently small can be imaged accurately, but also show that the 'white halo' and other artefacts that appear with larger objects are due to the high-pass filtering action of the phase plate, independent of any lens effects. The cut-on frequency of a Zernike plate and a characteristic dimension of the object can be combined to form a dimensionless 'cut-on parameter' whose value characterises the behaviour produced by this combination.

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The process of imaging by successive Fourier transforms, as described in section 8.6.3 of Born and Wolf [7] and chapter 5 of Goodman [8], is convenient for analysing the effect of a phase plate. This process defines the imaging behaviour of a coherently-illuminated perfect lens, ignoring quadratic functions of spatial coordinates and magnification. According to section 5.3.2 of [8], this neglect of quadratic factors is acceptable provided the object is sufficiently small in comparison with the lens aperture, as is likely to be satisfied in electron microscopy.

The results reported here are expressed in terms of various special functions, as defined by Abramowitz and Stegun [9] and the NIST Digital Library of Mathematical Functions [10]. This analysis omits effects both of charging and of loss in the object or phase plate, and applies only to weak phase objects.

2. General phase variation

At the object, a scattering centre acts as a source of a spherical wave. An element of this wave initially diverging at an angle θ to the axis arrives at the back focal plane (BFP) at a radius $r_s(\theta)$. We adopt the common approximation that

$$r_s \approx \theta f$$

where f is the focal length of the objective lens. To simplify later analysis, the incident and scattered wave vectors $\mathbf{k_0}$ and $\mathbf{k}(\theta)$ are defined here to have magnitude $2\pi/\lambda$, where λ is the electron wavelength, and their difference, $\mathbf{q}(\theta)=\mathbf{k}(\theta)-\mathbf{k_0}$, is a spatial frequency in angular measure. From the geometry of scattering for $\theta \ll 1$, $q \approx k\theta = 2\pi\theta/\lambda$ so $q \approx 2\pi r_s/\lambda f$. Since a Zernike plate produces a phase change only for $r > r_2$, it does so only for q values greater than $2\pi\, r_2/\lambda f$. This threshold value of q is is denoted here by q_0:

$$q_0 = 2\pi r_2/\lambda f$$

and is 2π times the corresponding quantity defined in [1,5,6] as the 'cut-on' frequency. It will be shown below that wave components with $q < q_0$ contribute little to the contrast, and so the plate acts as a high-pass filter.

When transforms of this sort are calculated without the phase plate, the integrals have ranges from 0 to ∞ or $-\infty$ to ∞, and suitable evaluations can be found without difficulty. However, to model a phase plate that provides a step change of phase at the cut-on frequency, it is necessary to evaluate integrals with the cut-on frequency as one of the limits. Very few suitable integrals are then available, and this restricts the types of object for which analytic solutions can be obtained. A semi-analytic solution is presented here for an object that produces uniform phase change over a cylindrical radius b. It is possible to find analytic solutions somewhat more easily for equivalent systems in 1D Cartesian coordinates, and two such solutions are presented here for comparison. However, the Cartesian analysis implies that not only the object but also the phase plate is of strip form and hence these solutions do not represent the behaviour of strip objects with a rotationally invariant Zernike plate. The spatial frequency spectra of all these objects contain components down to zero frequency and so are useful for modelling extended objects.

The mean potential in an object is more negative than the vacuum potential, so an electron travelling through matter moves with slightly greater average momentum than one of the same energy in the surrounding space. Hence, within the object, the electron wavelength is smaller, k is greater and the phase change kz is increased, relative to the same distance of transit z outside the object (section 4.2 of [11]). The phase changes induced both by a typical phase object relative to the direct beam, and by a Zernike plate on the scattered wave components relative to the direct beam, are thus both positive.

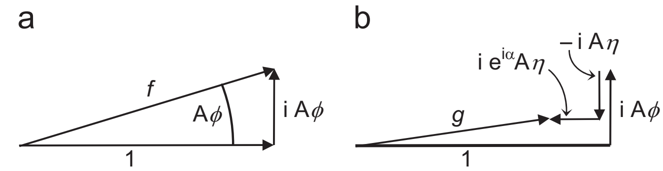

Fig. 1. (a) Wave incident on the object (shown as unit amplitude), wave f at exit from the object and scattered wave $iA\phi$. (b) Wave f as in Fig. 1(a), wave g after passing through the phase plate and wave components $(A\eta)$ introduced by the phase plate (drawn here for $\alpha = \pi/2$).

Consider a wave described by

$$\psi = \exp\, i[kz - \omega t + A\phi]$$

where A is the magnitude of phase shift and ϕ is a real function of x or of r. Different functions ϕ will be specified for different objects, with a maximum magnitude of 1. We ignore any attenuation and make the weak-phase object approximation by assuming that $A \ll 1$. The exponential can then be expanded to first order in A as

$$\psi \approx (1 + iA\phi)\ \exp\ i(kz - \omega t)$$

From here on we consider only far-field transforms, and omit the factor $\exp\ i(kz - \omega t)$. The remaining complex amplitude of the total wave

$$f = 1 + i\,A\phi \tag{1}$$

is a function of spatial coordinates. At any given point in the object exit plane it can be represented on a phasor diagram as in Fig. 1(a).

The effect of the phase plate will be found by defining a phase function ϕ for the object, finding the (spatial) frequency spectrum of the whole wave, applying the phase change α from the plate as a function of frequency and then transforming again to find the modified wave g in coordinate space. The analyses below express the results for g in the form

$$g = 1 + iA\phi - i\,(1 - \exp\ \ i\alpha)A\eta \tag{2}$$

where η is a function of a transverse coordinate and of q_0, the cut-on frequency of the plate. If η is equal to ϕ, then (2) gives the same result for g as if the vector $iA\phi$ were rotated by α. Just as the phase change of the original wave by the object is said to add the vector of magnitude $A\phi$, so the phase plate adds further components $(A\eta)$ both opposing that vector and in the direction $(\alpha + \pi/2)$. The resulting intensity is, from (2),

$$|g|^2 = 1 - 2A\eta\ \sin\ a + O(A^2)$$

Thus if the phase plate advances the scattered component $(iA\phi)$ in Fig. 1(a) and so rotates it counter-clockwise, the total wave g then becomes smaller in amplitude than the initial wave (Fig. 1(b)), the intensity is reduced and the image becomes darker. A general rotation by α is analysed here, with phase advance (as provided by a thin Zernike plate) shown by a positive value of α.

The relative distribution of intensity over the image is proportional to $\sin\, \alpha$ which is constant over the image, and to η which varies over the image. When the object is sufficiently large (to be quantified later), $|\eta|$ is small over most of the image and the variation of intensity remains proportional to A^2. When the object is sufficiently small, the magnitude $|\eta|$ increases to the same order as ϕ and the intensity contains a component proportional to A. It will be shown that the distribution of η in the image plane may differ substantially from that of ϕ.

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