Ultimate Recovery of Low-Frequencies in Thin-film ZPC-TEM by Inverse Projector

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We have developed a method for Zernike phase-contrast (ZPC) transmission electron microscopy (TEM) to recover very low frequency components. The ZPC-TEM using a Zernike phase plate (ZPP) made with thin amorphous carbon-film realizes very high contrast images [1], because it can visualize low frequency components, except for very low frequency components. The lack of the very low frequency components is caused by the phase plate design with a finite-sized central hole. While phase-plates with smaller hole-size is required to recover lower frequency components, the carbon-film with a smaller hole is exposed by stronger electron beam around the central beam-spot, and its lifetime becomes shorter due to the problem of charging. To solve the problem, we develop a method for recovering the very low frequency components by computer calculation, even for larger holes

As a mathematical formulation, the lack of low-frequency components is described by a projector in the frequency space. Inverse of a projector is ill-posed problem. To fix such ill-posed issues, traditionally, additional information has been supplied as supports in the real space. In the context, iterative projection algorithm (IPA) has been developed by using two conditions (constraints), one that defined in the frequency space and the other that given as supports as an additional information in the real space. We analytically determined the inverse projector with the help of the supports in the real space, as an infinite series of linear operators. As a practical approach to fix the huge dimension of the inverse projector, a computer-implementation of the inverse projector automatically gives a kind of the IPA. We also find a computational method for correcting images distorted by misalignment of the phase plate.

The proposed method allows us to use a ZPP with a larger hole, which makes the ZPC-TEM friendly in the beam alignment and the ZPP lifetime. Another virtue of the method is automatic settlement of the issue of image fringing, which has been a major trouble in ZPP inevitable by using finite-sized central holes [2] (refer to Figure 1).

The applicability of the method with improved ZPPs is to be reported with biological samples such as virus (refer to Figure 2).

References:

- [1] R. Danev and K. Nagayama, Ultramicroscopy, 88 (2001) 243.
- [2] R. Danev, S. Kanamaru, M. Marko, K. Nagayama, J. Struct. Biol., 171 (2010) 174.



Figure 1. A simulation of ZPC-TEM image for two phantoms (rings with internal structures), and a reconstruction by the inverse projector method. There arises image-fringing effect in the ZPC image due to the deficit of low frequency components. The method reconstructs the internal structures correctly.



Figure 2. The original ZPC-TEM image of a virus, which includes no very low frequency components due to the finite-sized hole of the ZPP, and the resultant image whose very low frequency components are recovered by computer calculation with the inverse projector method. The curves below the images indicate brightness-plots on the x-axis of the images. The inverse projector is performed with the segmentation (support) as information of the specimen. The segmentation is semi-automatically generated with a mathematical morphology algorithm.