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The Rocking Phase Plate – another step towards improved stability

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Phase plates are promising tools for enhancing contrast, especially in cryo-transmission electron microscopy (cryo-TEM). As an alternative to defocusing, contrast of weak phase objects -like most cryo samples- can be improved in close to focus images by inserting a phase plate into the back focal plane of the TEM (1). The phase plate adds an additional phase shift of ideally π/2 to the scattered electrons and thus enhances image contrast. Their advantages for electron tomography and single particle acquisition have been demonstrated recently (2). Nevertheless, phase plates are still no tool for routine application. This is mostly due to difficult handling and limited durability of actual phase plate designs. For more than 15 years, carbon based Zernike phase plates have been the most used type (3). Recently, a new type of hole free phase plates -Volta phase plate- has been made available. The hole free design has reduced ringing artifacts due to the missing hole edge. This is on cost of a variation of the phase shift over time.

Here, we present our experimental results from a rocking phase plate. We use a classical Zernike type phase plate which is based on a thin iridium film instead of carbon. This improves the long term stability of the phase plate. To overcome the ringing artifact problem, we move the phase plate on a circular path during acquisition (see figure 1). This rocking mode virtually smooths the edge of the central hole. Additionally, the diameter of the central hole can be larger compared to classical Zernike phase plates which facilitates the positioning of the phase plate inside the TEM. The rocking mode is possible due to a special, piezo based positioning system, which can position the phase plate with nanometer precision (4).

We were able to show that the rocking mode settings can be used without interference of the phase plate hole edge during image recording. Furthermore, we could not find any resolution loss caused by the moving phase plate. Figure 2 shows a comparison between the phase plate in a) steady and b) rocking mode. No cutoff can be detected in the power spectra of the rocking phase plate in comparison to the power spectrum calculated from a micrograph recorded with a not moving phase plate.

In conclusion, the rocking mode could be an alternative method to acquire phase plate data. Currently, we are recording high-resolution single particle datasets to demonstrate up to which resolution the rocking phase plate can be used.

References:

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Fig. 1: Illustration of the phase plate rocking mode.

Fig. 2: Power spectra of TEM-micrographs in a) steady mode and b) rocking mode.

Fig. 1

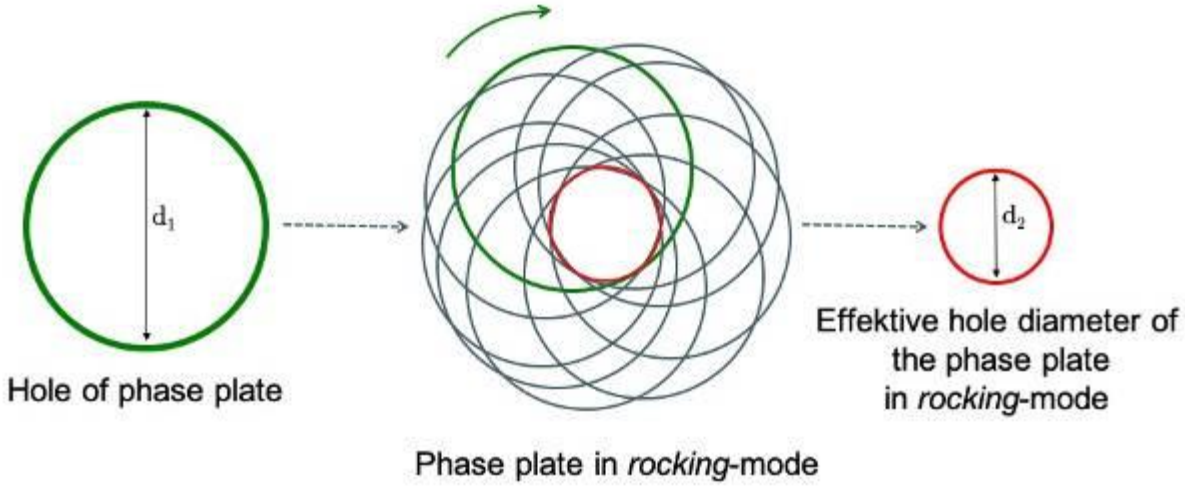
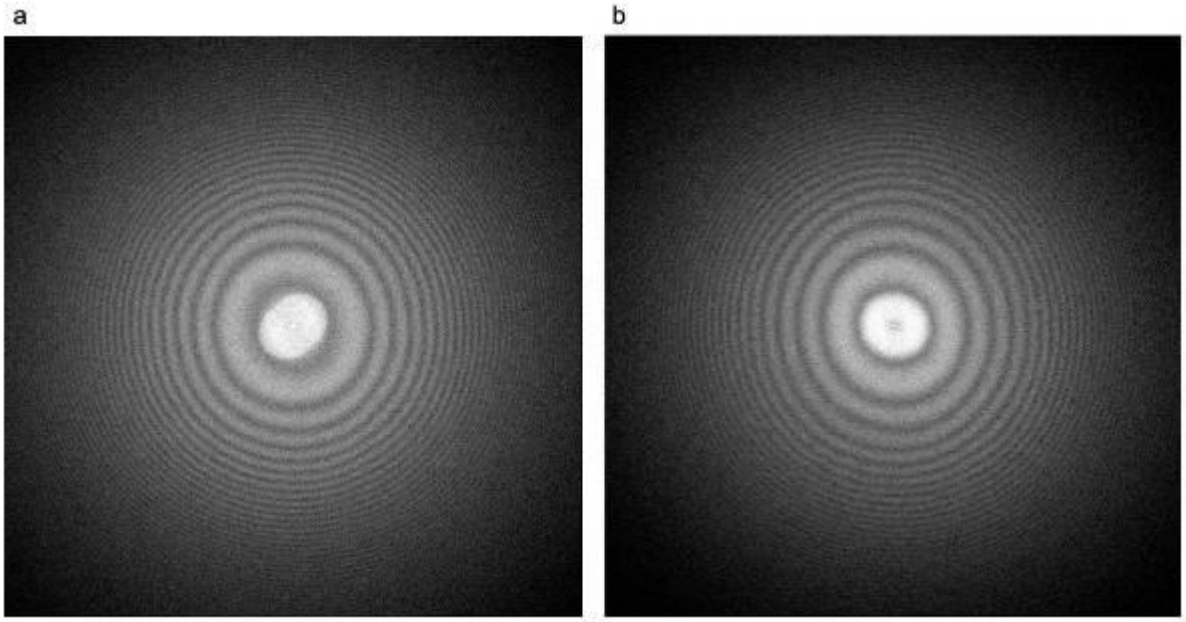

d1
d2
Hole of phase plate
Phase plate in *rocking*-mode
Effektive hole diameter of
the phase plate
in *rocking*-mode

Fig. 2


a
b