## **Contrast Enhancement of Nano-materials Using Phase Plate STEM**

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Phase contrast of biological molecules composed of light elements can be enhanced by using a phase plate (PP) because a phase contrast transfer function (PCTF) can be modified from a sine type to a cosine type. Since many biological molecules were observed with high contrast by phase plate transmission electron microscopy (P-TEM), P-TEM is as an effective method. However, charge up of the PP is one of the most serious issues in P-TEM. The PP has to be set on a back focal plane (BFP) of an objective lens (OL) of an electron microscope. A diffraction pattern is formed on the BPF and an electron beam at the center of the diffraction pattern which corresponds to unscattered wave gives high density electron current. Therefore, an electron irradiation of the unscattered wave onto the PP should cause a local charge up of the phase plate.

Another issue in P-TEM is in a structural limitation of an instrument. Since distance between a sample plane and BFP of the OL is very small, some of the instrument which has very high spatial resolution can't be used for P-STEM. The BFP of the high spatial resolution electron microscope, is not in between the gap of the pole piece and the PP can't be set on the BFP. In this case an additional optical system such as a transfer lens is necessary [1]. Thus, the P-TEM study can't be done using existing high resolution electron microscopes. The Instrument which realizes high resolution P-TEM optics have to be newly introduced.

Phase plate scanning transmission electron microscopy (P-STEM) would resolve these issues [2]. In STEM mode the PP have to be set on a front focal plane (FFP) of the OL. The PP can be set on condenser aperture (CA) plane and the CA plane can be made to be conjugate with the FFP of the OL by adjusting illumination optical system. Thus, P-STEM could easily applied to any existing instruments. Since intensity of the illuminating electron on the CA plane is homogeneous, the local charging does not occur on the PP and the charge up issue of the PP is also resolved. In fact the PCTF did not show an apparent change during P-STEM observations as shown in Figure 1. This is a big advantage in P-STEM against P-TEM.

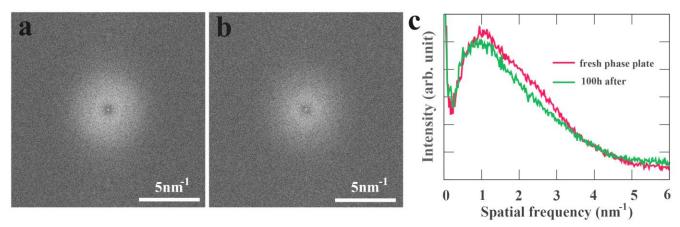
An optical condition of PSTEM was realized and a sine type PCTF can be modified into a cosine type PCTF when a Zernike type PP with a  $\pi/2$  thickness was used. In the present paper applications of P-STEM to several kinds of nano-particles are demonstrated. Figure 2 is an example of the P-STEM image of quantum dots (Q-dots). A STEM image taken with conventional STEM (C-STEM) optics was also shown as a reference. Fig. 2 (a) is a P-STEM image and Fig. 2(b) is a C-STEM image of quantum dots. Dark dot images are the Q-dots and are aggregated. Image contrast of the Q-dots in (a) is higher than that in (b). Moreover, a background pattern which corresponds to a carbon film pattern is also clearer in (a). The power spectra obtained from these images are shown at the right bottom of the each image. The power spectrum in (a) has higher intensity around the center than that in (b) showing modification of the PCTF. The sizes of the Q-dots are around 1nm~10nm and these sizes correspond to the spatial

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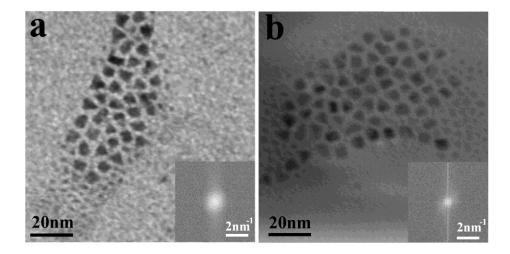
frequency at which intensity of the power spectrum in (a) is higher than that in (b). Thus, the higher image contrast of the P-STEM image is due to the modification of the PCTF. [3]

## References:

- [1] F. Hosokawa, R. Danev and K. Nagayama, J. Electron Microscopy 54 (2005) 317-324.
- [2] H. Minoda et al, Microscopy **64** (2015) 181-187.
- [3] This development was supported by SENTAN, AMED.



**Figure 1.** Power spectra at the initial stage of the PP (a) and after 100h (b). Figure (c) compare the PCTFs of (a) and (b). An amorphous carbon film was used as a specimen.



**Figure 2.** Comparison of a P-STEM image (a) and a conventional STEM image of quantum dots.