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Reducing the missing wedge: High-resolution dual axis tomography of inorganic materials

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Abstract

Electron tomography is a powerful technique that can probe the three-dimensional (3-D) structure of materials. Recently, this technique has been successfully applied to inorganic materials using Z-contrast imaging in a scanning transmission electron microscope to image nanomaterials in 3-D with a resolution of 1 nm in all three spatial dimensions. However, an artifact intrinsic to this technique limits the amount of information obtainable from any object, namely the missing wedge. One way to circumvent this problem is to acquire data from two perpendicular tilt axes, a technique called "dual axis tomography." This paper presents the first dual axis data at high resolution for inorganic materials, and by studying a CdTe tetrapod sample, demonstrates the increase in information obtained using this technique.

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Keywords: Dual axis electron tomography; Z-contrast imaging; Three-dimensional data

1. Introduction

Over recent years, nanotechnology has become a key component in the field of materials science. Technological advances are yielding ever smaller, more complex and anisotropic nanostructures that need to be characterized in all three dimensions. Unlike previous transmission electron microscopy (TEM) experiments in which the analysis of bulk single or poly-crystal thin films was often undertaken assuming uniformity parallel to the beam, many nanomaterials that are now studied have a finite size and shape in all three dimensions, and are not necessarily uniform in any direction. This new demand on materials characterization has led to the development of electron tomography for a full three-dimensional (3-D) analysis of nanomaterials.

Tomography has been used in many branches of science for nearly half a century. Although X-ray tomography was first used in medicine in what is now known as the CAT scan (computer-assisted tomography) [1], it has more recently been applied to materials science and engineering to reconstruct many microscopic 3-D structures such as metallic foams [2], and in the biological sciences to reconstruct cell structures [3]. Unfortunately, due to the wavelength of X-rays and other mechanically limiting factors, conventional X-ray tomography cannot yield a resolution better than about 1 μm. On the other end of the resolution spectrum, the atom probe field ion microscope (APFIM) has been developed over many years to produce 3-D reconstructions with single atom resolution [4]. The limitations with this technique, however, are that the sample must be conducting, withstand high field stresses, and can only produce reconstructions of relatively small volumes. The optimum technique for a nanometer-scale analysis of a wide variety of materials is electron tomography.

Electron tomography has been used very successfully in the biological sciences to study many cell structures, viruses, etc. [5,6] using primarily bright-field (BF) TEM. Attempts have been made to apply this same technique to

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inorganic materials, but have often led to the reconstruction artifacts arising from the violation of the projection requirement, which states that the signal used for tomographic reconstructions must be a monotonic function of a physical property [7]. In general, the intensity of BF and dark-field (DF) images of crystalline materials, whose contrast depends almost entirely upon the diffraction condition of the crystal, does not have a monotonic relationship with the thickness of the sample [8]. One way to circumvent this problem is to use an incoherent signal for image formation, such as that used in Z-contrast imaging in the scanning transmission electron microscope (STEM). Z-contrast images are formed by collecting the high-angle scattered electrons (40–100 mrad at 200 kV) on an annular dark field detector. Detecting the scattered intensity at these high angles and integrating over a large angular range effectively averages coherent effects between neighboring atomic columns in the specimen [9–11]. Therefore, diffraction contrast in the sample is minimized, and the projection requirement is fulfilled. This imaging technique yields reliable and quantifiable 3-D reconstructions of inorganic (crystalline) materials, and as such, is the technique used for all the results shown in this paper. STEM tomography has been applied to semiconducting cadmium telluride nanostructures called tetrapods. A perfect tetrapod has four legs in tetrahedral symmetry, and as such, there is a good chance that one of the legs will be in an orientation that is perpendicular to the tilt axis. The high symmetry of the tetrapod allows the effects of the missing wedge to be clearly illustrated and to demonstrate how these effects are reduced by implementing dual axis tomography.

2. Image acquisition

All the experimental data presented here were acquired on an FEI Tecnai F20 microscope at an accelerating voltage of 200 kV, equipped with a Super-Twin objective lens and a Fischione high angle annular dark field (HAADF) detector. Electron tomography must be performed using a specialized high-tilt tomography holder, with the width of the holder determined by the size of the pole piece gap. For single axis tomography, the Fischione Advanced Tomography Holder was used, which is only 4 mm wide and 1.7 mm thick (Fig. 1). This holder is limited by the microscope goniometer to a tilt of $\pm 80^\circ$. In practice, this very high tilt is rarely achieved due to shadowing from the specimen, either from grid bars if present, or simply the change in thickness of the specimen if ion milled or focused ion beam thinned. The inability to obtain information from the specimen beyond some maximum tilt leads to a "missing wedge" of data, resulting in reconstruction artifacts, the most apparent being an elongation of the reconstruction perpendicular to the tilt axis. Fig. 2 shows a 2-D slice of Fourier space in which the uneven sampling is evident and the missing wedge of information is determined by the maximum tilt angle, α. By recording a second

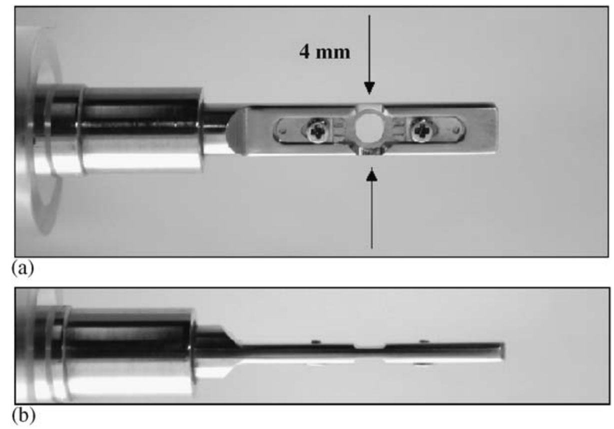

Fig. 1. Single tilt Fischione Advanced Tomography Holder (a) viewed from the top and (b) viewed from the side. Its small dimensions of 4 mm × 1.7 mm make it ideal for tomography, with the tilt range only limited by the specimen or microscope goniometer.

tilt series of the same volume of the sample around a tilt axis perpendicular to the first, considerably more of Fourier space is sampled and the "missing wedge" is reduced to a "missing pyramid." This form of dual axis tomography will minimize many reconstruction artifacts and allow us to gain more information about our samples.

Here, we present the first high-resolution dual axis data of inorganic materials using a new Dual-Axis Advanced Tomography Holder by Fischione. As can be seen from Figs. 3 (a) and (b), the tip of this holder is slightly thicker than the single axis holder (5 mm × 3 mm) to allow internal sample rotation capabilities, limiting the tilt range to $\pm 70^\circ$. At the back of the holder, two knobs can be seen. The knob labeled (A) in Fig. 3(c) allows for free 360° rotation of the sample to find the best orientation to start the tilt series. Once this is done, that position can be clamped (using knob labelled (B)), the first tilt series can be acquired, and then the knob labeled (C) can be used to rotate the sample by 90° with an accuracy of less than 1°. Then the second, perpendicular, tilt series can be taken from the same volume of the sample.

To achieve ~1 nm resolution in all three dimensions in the reconstruction, a tilt series of ~140 images must be acquired for a volume of $\sim 200\,nm^3$ or less. This can be achieved either by recording a tilt series at equal angular increments (e.g. every 1° over $\pm 70^\circ$), or by using the Saxton scheme, in which the total number of images is specified, and the higher tilt angles are sampled more than the lower tilt angles, known to improve the quality of the final reconstruction [12]. Both methods will take, on average, ~3 h once the microscope is aligned and the volume of interest is found.

3. Tomographic reconstruction

Once the tilt series has been acquired, the reconstruction procedure can be started. A very important first step is the alignment of the images within the data set. This can

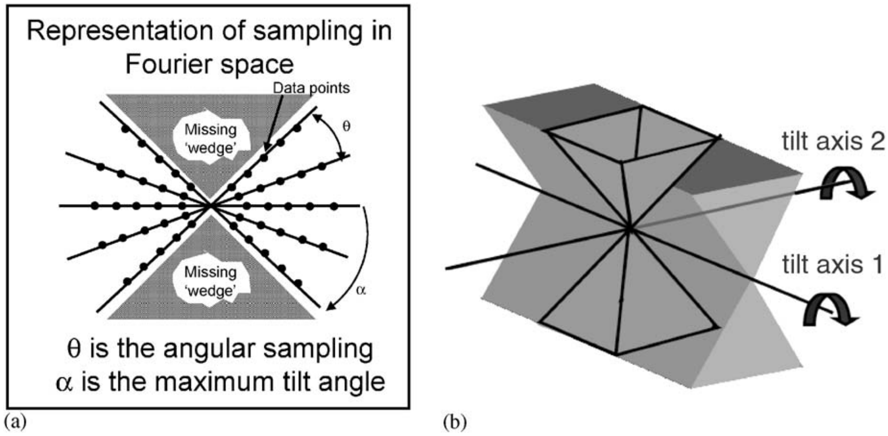

Fig. 2. (a) A schematic showing the sampling of data in Fourier space. The limited range of tilt leads to a missing wedge, resulting in reconstruction artifacts, which is reduced by dual axis tomography to a missing pyramid (green) as shown in (b).

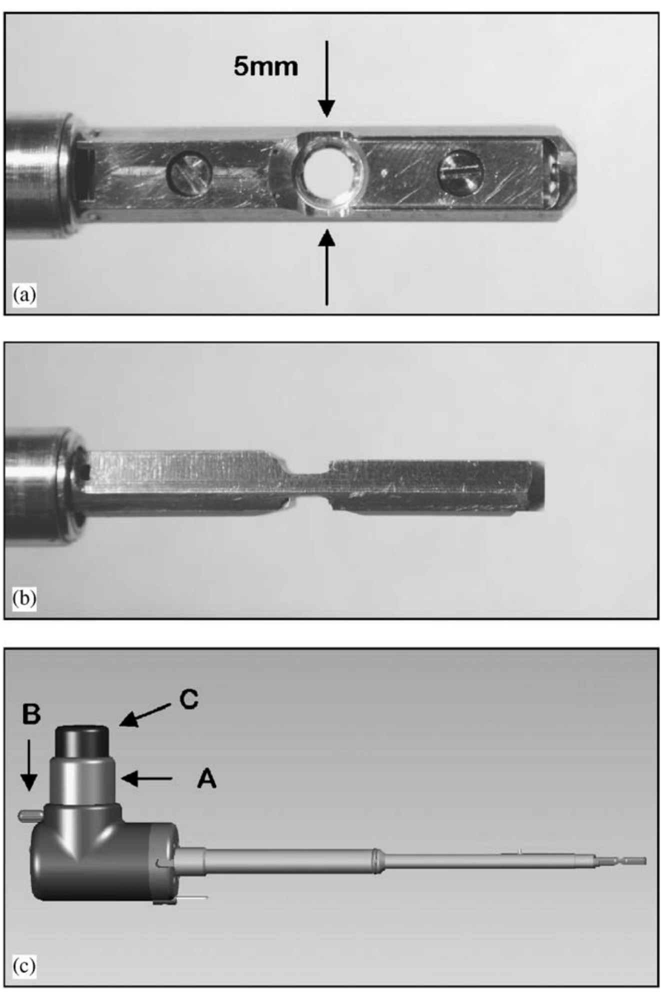

Fig. 3. Fischione Dual-Axis Advanced Tomography Holder (a) viewed from the top, (b) viewed from the side, (c) shows a schematic of the whole holder, and illustrates the rotation capabilities. Knob A allows for free 360° rotation of the sample, knob B is used to clamp down the desired starting position, and knob C is used for 90° rotations from that starting position.

usually be achieved with a cross correlation algorithm available in most software packages. However, in some cases, the geometry of the sample is such that a satisfactory cross correlation cannot be achieved, and the alignment must be performed manually image by image, a very laborious, but necessary, task. Once the images are correlated, the whole data set also has to be aligned with respect to the tilt axis of the goniometer, which rarely coincides with the optic axis of the microscope, the effects of which are discussed in Ref. [8]. Finally, the data is ready to be reconstructed.

The reconstruction is performed using a backprojection method in which an image, or projection, is "smeared" back into an object space at the angle of the original projection. Using a sufficient number of projections, from different angles, the superposition of all the backprojected images will, in an ideal case, return the original object [13,14]. Unfortunately, reconstructions by backprojection are always blurred with an enhancement of low frequencies and a loss of fine spatial detail. This is an effect of the uneven sampling of spatial frequencies in the series of original images, or projections. Assuming an even sampling of Fourier space in each projection, this will result in a proportionately greater sampling density near the center of Fourier space compared with the periphery (Fig. 4), which leads to an under-sampling of the high spatial frequencies of the object and a "blurred" reconstruction. This can be partially corrected in Fourier space using a weighting filter (a radially linear function in Fourier space, zero at the center and a maximum at the edge). This weighting filter has the effect of rebalancing the frequency distribution in Fourier space and minimizing the blurring in real space, an approach known as weighted backprojection.

Reconstructions using the backprojection method will always be 'imperfect' because of the limited sampling and poor signal-to-noise ratio (SNR). However, by noting that each projection is a 'perfect' reference, the quality of the

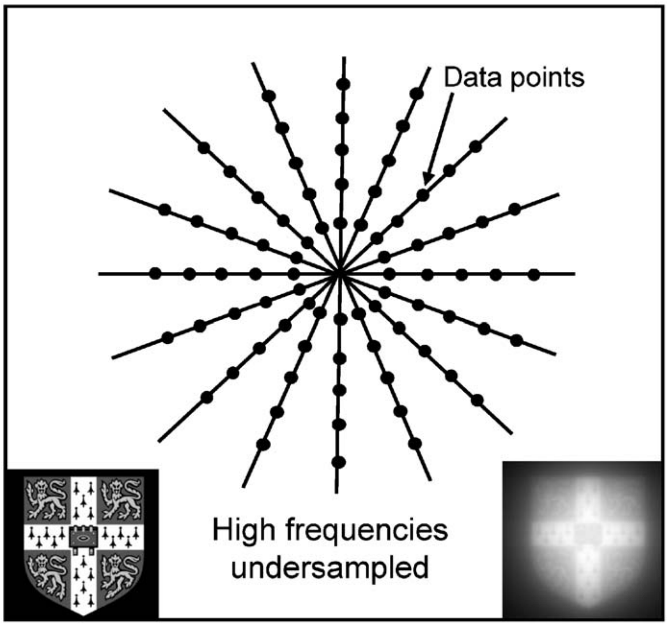

Fig. 4. A representation of the sampling of data points in Fourier space, showing that the higher frequencies are sampled much less than the lower frequencies. This leads to a blurring in a simple backprojection reconstruction, as the higher frequencies are undersampled (inset on bottom right), as compared to the original image (bottom left).

reconstruction can be improved. If the (imperfect) reconstruction is re-projected back along the original projection angles the re-projections, in general, will not be identical to the original projections (images). The difference between them will be characteristic of the deficiency of the reconstruction from the limited dataset. This difference can be back-projected into reconstruction space, generating a 'difference' reconstruction, which can then be used to modify the original reconstruction in order to correct the imperfections in the backprojection. This constrains the reconstruction to agree with the original projections. As the 'difference' is also being back-projected, a single operation will not fully correct the reconstruction and the comparison operation must be repeated iteratively until a 'best' solution is reached [15]. This method is called the Simultaneous Iterative Reconstruction Technique (SIRT).

Although these methods are known to succeed using data from a single tilt axis, their implementation becomes more difficult when applied to dual axis data. One way to reconstruct the two sets of data is to reconstruct each data set individually, either using a weighted back projection, or SIRT. The two data sets can be summed in Fourier space [17], with the resulting reconstruction benefiting from information in both tilt series. However, a more rigorous and more accurate algorithm has been developed by us called alternating dual-axis SIRT (ADA-SIRT) [16].

In this new algorithm, the initial reconstruction is formed by combining, in Fourier space, the two back-projected reconstructions from the two tilt series, as before [17]. As with single-axis SIRT, the reconstruction is reprojected, and the difference between reprojections and original images are used to further refine the reconstruction. The first iteration and subsequent odd-numbered iterations compare the initial combined reconstruction with the first set of projections (first tilt series), and on the second and subsequent even-numbered iterations, the reconstruction is refined using the second set of projections (second tilt series). In this way, the method alternates between the two sets of data. Corrections are made to account for the rotation and for any lateral shift between the two series at every iteration. These must be determined beforehand, but it has been shown previously that it is possible to align tomograms reconstructed from mutually perpendicular tilt series, even without the use of fiducial markers [18]. This routine is based on the Riemann sum procedure in the IDL programming language, and IDL is used for the dual axis reconstructions.

4. Results and discussion

Two perpendicular tilt series were taken of a CdTe tetrapod sample. Each tilt series contained 69 images, the first series ranging from -70° to $+70^\circ$, and the second series from -65° to $+70^\circ$ (acquiring two perpendicular series over exactly the same tilt range from the same volume is very difficult due to the shadowing effects discussed previously). Before the acquisition of the data sets, a suitable region of the sample was chosen, and it was rotated at low magnification to ensure that the same area of the grid remained in the field of view. Fig. 5 illustrates this rotation at low magnification, with arrows indicating the center of the grid in both images. Fig. 5(b) is rotated 90° clockwise with respect to Fig. 5(a), and the accuracy of the rotation is less than 1°. Fig. 6 shows two Z-contrast images at 0° projection, one from each tilt series. Again, the second tilt series (Fig. 6(b)) has been rotated 90° clockwise with respect to the first (Fig. 6(a)), and the arrows indicate locations of the same two tetrapods in the field of view.

The reconstructions of this sample highlight the effects of the missing wedge. Fig. 7(a) shows a reconstruction of the first tilt series using single axis SIRT. With respect to the original images from Fig. 6, it can be seen that certain legs of the tetrapods are either missing or weak, as indicated by the arrows. It is important to note that only legs in certain orientations are missing (perpendicular to the arrows) and legs that are even a few degrees away from this orientation are present. This effect is because the tilt axis is parallel to the direction of the arrows; legs that are perpendicular to the axis will be strongly affected by the missing wedge, and will not appear in the reconstruction. Fig. 7(b) is a reconstruction of the second tilt series, also performed using single axis SIRT. The reconstruction has been rotated to match the orientation of the first one for comparison. As this tilt series is at 90° to the first, one would expect that legs in the perpendicular direction would be missing now, and this is clearly the case as can be seen in Fig. 7(b). Again, the tilt axis is parallel to the direction of

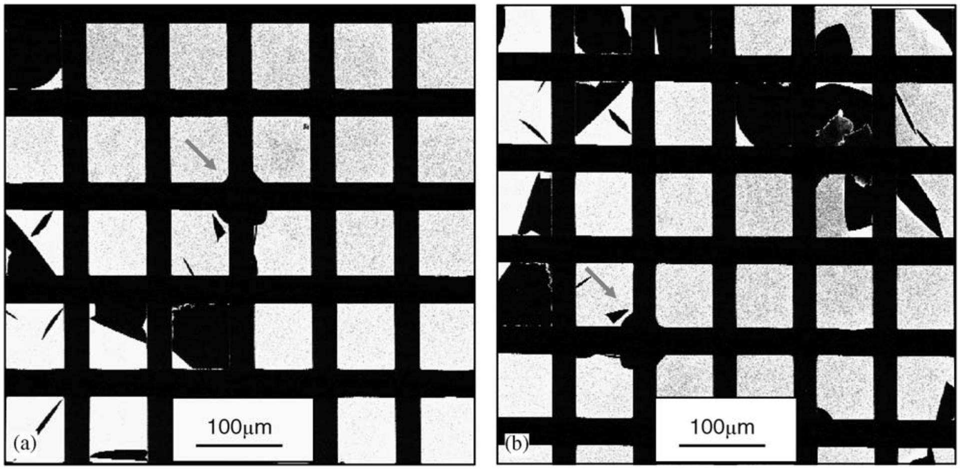

Fig. 5. Low-magnification STEM images that show the grid bars of the sample. The arrows indicate the center of the grid, and it can be seen that image (b) had been rotated 90° clockwise with respect to image (a) with an accuracy of less than 1°.

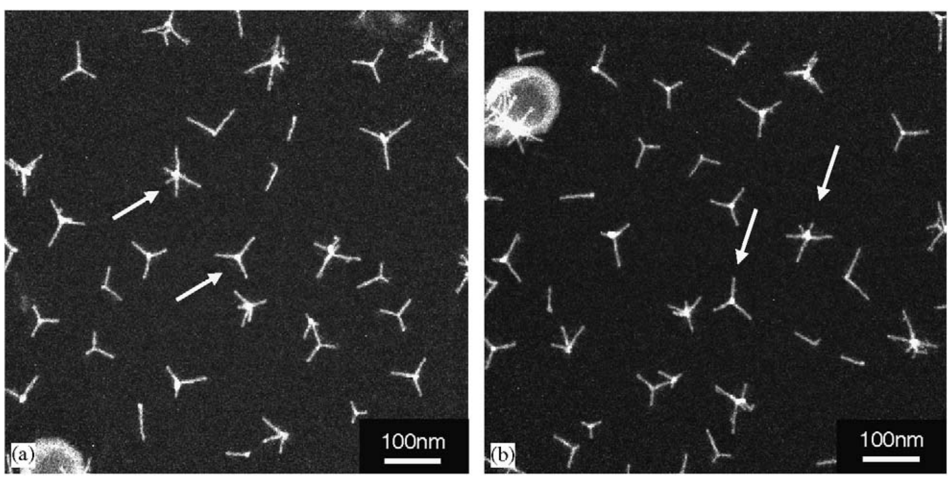

Fig. 6. Z-contrast images of CdTe tetrapods in two perpendicular orientations at 0° tilt. The arrows indicate the location of the same tetrapods in both images, and it can be seen that image (b) had been rotated 90° clockwise with respect to image (a).

the arrows. Although anticipated from theory, it is still quite striking to see this effect in practice with legs missing in one tilt series reconstructed perfectly in the second, perpendicular, series, and vice versa.

Combining the two data sets, we should be able to reconstruct all legs in all directions. As described previously, this can be done by combining the reconstructions in Fourier space or by doing a more rigorous reconstruction using alternating dual axis SIRT. Fig. 7(c) shows a reconstruction using the first of these methods, and we see that this reproduces all legs in one reconstruction, reducing the effect of the missing wedge. At this reduced pixel resolution and scale, the data does not seem suffer from any artifacts. However, performing the reconstructions at full pixel resolution on a single tetrapod illustrate the differences in the two reconstruction techniques. Fig. 8(a) shows a reconstruction of one tetrapod having summed the two data sets in Fourier space, and shows that all of the legs are present, but that the reconstruction has suffered from noise. Fig. 8(b) shows the same tetrapod reconstructed using ADA-SIRT and visualized in the same exact way. Again all legs are present using this method, but there is clearly a reduction in artifactual noise in Fig. 8(b), indicating the constraints imposed in the ADA-SIRT algorithm have a clear beneficial effect on the quality and resolution of the reconstruction.

While dual axis tomography is undoubtedly time-consuming, this work illustrates, compared to single axis tomography, that there is significant improvement in the amount of 3-D information that can obtained from nanoscale materials. The development of accurate 3-D characterization at the nanoscale enables many important structural features to be studied, including the size, shape and faceting of small particles such as quantum dots and catalysts and accurately observing fine detail in materials such as semiconductor devices.

Fig. 7. 3-D reconstructions of a dual axis tilt series. (a) is a reconstruction of the first tilt series and shows that some of the legs of the tetrapods are missing or weak (compared to the original images in Fig. 6) due to the missing wedge, as indicated by the arrows. (b) is a reconstruction of the perpendicular tilt series showing that while the missing legs in (a) are present in this data set, there are a different set of missing legs, again indicated by the arrows. (c) is a dual axis reconstruction of the two data sets (Fourier sum) and illustrates that no legs are missing because the missing information has been greatly reduced. The tilt axes in (a) and (b) are parallel to the direction of the arrows.

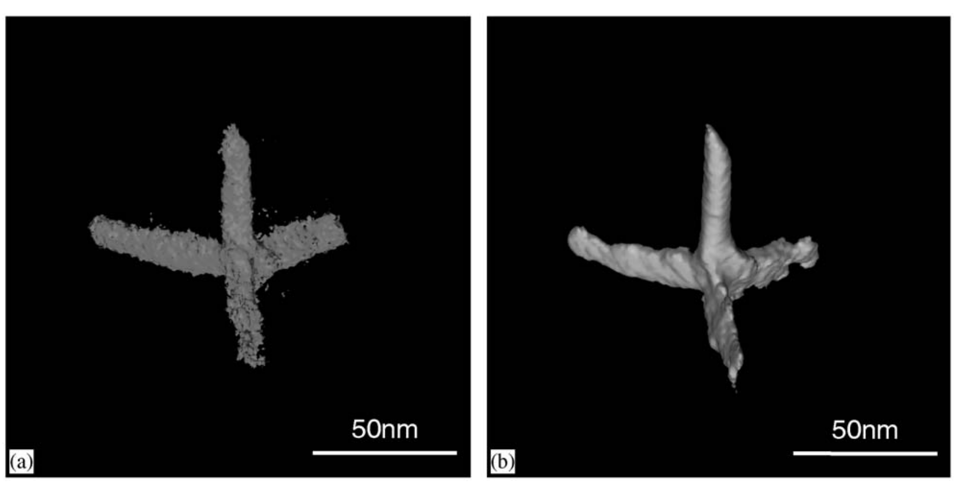

Fig. 8. Dual axis reconstructions of the same tetrapod, both visualized by surface render. (a) shows the reconstruction of the data summed in Fourier space and (b) shows the reconstruction using ADA-SIRT. There is a marked reduction of noise in (b), illustrating that ADA-SIRT yields an improved reconstruction.

5. Conclusions

High-resolution electron tomography has proven to be a powerful technique that has great implications for all of the nanosciences, and is now becoming widely used. With single axis tomography, a resolution of 1 nm^3 is attainable, making it possible to elucidate the structure–property relationships of ever smaller nanostructures. However, the amount of 3-D information that can be obtained is often sub-optimal due to a limited tilt range, and leads to reconstruction artifacts brought about by the "missing wedge" of data. Using dual axis tomography, this missing wedge can be reduced to a "missing pyramid" by acquiring two tilt series about mutually perpendicular axes.

Here we have presented the first high-resolution dual axis tomographic data of inorganic materials using a dual axis holder of novel design from Fischione, showing direct experimental evidence of the deleterious effect of the missing wedge by means of a CdTe tetrapod sample. This is reduced when the two data sets are combined, and it is shown that a new algorithm, alternating dual axis SIRT, yields considerably improved tomograms compared to a standard Fourier sum. While the improvement in resolution has not yet been quantified, this work clearly illustrates that the amount of 3-D information obtained from the material is drastically increased, and the fidelity of the reconstruction greatly improved.

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References

[1] G.N. Hounsfield, A Method and Apparatus for Examination of a Body by Radiation such as X or Gamma Radiation, The Patent Office, London, England, 1972.
[2] J. Banhart, Progr. Mater. Sci. 46 (2001) 559.
[3] C. Larabell, M.A. Le Gros, Mol. Biol. Cell 15 (2004) 957.
[4] M.K. Miller, Atom-probe Tomography: Analysis at the Atomic Level, Kluwer Academic/Plenum Press, New York, 2000.
[5] J. Frank, T. Wagenknecht, B. McEwen, M. Marko, C. Hsieh, C. Mannella, J. Struct. Biol. 138 (1–2) (2002) 85.
[6] W. Baumeister, R. Grimm, J. Walz, Trends Cell Biol. 9 (1999) 81.
[7] P.W. Hawkes, The electron microscope as a structure projector, in: J. Frank (Ed.), Electron Tomography: Three-dimensional Imaging with the Transmission Electron Micrsocope, Plenum Press, New York/London, 1992.
[8] P.A. Midgley, M. Weyland, Ultramicroscopy 96 (2003) 413.
[9] S.J. Pennycook, L.A. Boatner, Nature 336 (1988) 565.
[10] E.M. James, N.D. Browning, Ultramicroscopy 78 (1999) 125.
[11] P.D. Nellist, S.J. Pennycook, Ultramicroscopy 78 (1999) 111.
[12] W.O. Saxton, W. Baumeister, M. Hahn, Ultramicroscopy 13 (1984) 57.
[13] R.A. Crowther, D.J. de Rosier, A. Klug, Proc. Roy. Soc. Lond. A. 317 (1970) 319.
[14] M. Radermacher, Weighted backprojection methods, in: J. Frank (Ed.), Electron Tomography: Three-dimensional Imaging with the Transmission Electron Microscope, Plenum Press, New York/London, 1992.
[15] P. Gilbert, J. Theor. Biol. 36 (1972) 105.
[16] J. R. Tong, I. Arslan, P. A. Midgley, J. Struct. Biol. 153 (2006) 55.
[17] D.N. Mastronarde, J. Struct. Biol. 120 (1997) 343.
[18] J. R. Tong, M. Weyland, R. E. Dunin-Borkowski, P. A. Midgley, Proceedings of the XIII European Congr. Microscopy, vol. I, 2004, p. 227.