A novel dual-axis reconstruction algorithm for electron tomography

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Abstract. A new algorithm for computing electron microscopy tomograms which combines iterative methods with dual-axis geometry is presented. Initial modelling using test data shows several improvements over both the weighted back-projection (WBP) and Simultaneous Iterative Reconstruction Technique (SIRT) method, and, with increased stability and tomogram fidelity under high-noise conditions.

1. Introduction

In electron tomography, a series of images (projections) are collected over a range of angles. Due to the physical constraints imposed by the microscope, sample, and holder, the range of tilt available for the image series from which the tomogram is constructed is usually limited to ±70°. This leads to a 'missing wedge' of information as seen in Fourier space which results, after reconstruction, in a blurring in the direction of the optic axis.

One method that has been used to try and reduce the problem of this missing wedge is to use dual-axis tomography, in which there are two tilt series, recorded with mutually perpendicular tilt axes, reducing the missing information to a 'missing pyramid'. Penczek et al used dual-axis tomograms, in which two tilt series were taken at 90° and then combined into a single tomogram, to improve resolution [1]. Mastronarde showed how it was possible to selectively combine the two tomograms in Fourier space, and reported that this yielded better results than a simple weighted back projection [2].

This paper reports the development of a process to combine the dual-axis geometry, with the advantages shown by Penczek et al and Mastronarde, with iterative methods such as SIRT [3], to improve the quality of the tomographic reconstruction.

2. The New Algorithm

In the new algorithm, "alternating dual-axis SIRT", the initial reconstruction is formed by combining, in Fourier space, two back-projected reconstructions from each of two tilt series, as suggested by Mastronarde [2]. In this way the initial step is similar to dual-axis weighted back-projection (WBP) methods.

Ideally the angle between the tilt axes of these tilt series would be 90°, but this is not necessary, as shown later. The first iteration and subsequent odd-numbered iterations compare the initial combined reconstruction with the first set of projections (X). As with single-axis SIRT, the reconstruction is...
reprojected, and the difference between reprojections and original projections found and used to further refine the original reconstruction. The first set of projections is used for all odd-numbered iterations. On the second and subsequent even-numbered iterations, the reconstruction is refined using the second set of projections (Y). In this way the method alternates between the two sets. Corrections are made at every iteration to account for the rotation and for any lateral shift between the two series. See Fig. 1

Figure 1. The dual axis iterative algorithm is split into two parts, an initial reconstruction and combination step, and an iterative loop. Depending on whether the iteration number (n) is odd or even, a different image series will be used in each iterative loop.

3. Testing of the Algorithm
In order to test the accuracy of the algorithm and compare it to single-axis SIRT and other methods of combining the two tilt series, test data volumes of 128³ voxels were created and a number of 'tilt series' were created from these volumes. The test object used for the results was a sphere-and-rods arrangement with features along mutually perpendicular axes. The object has a constant density and the image background was set to zero throughout.

For each test case, comparisons were made between several methods. In order to ensure a fair comparison, the total number of iterations was kept constant for iterative methods, and the number of images was halved for each tilt-series when using dual-axis methods, by using 2° spacing instead of 1° spacing. This way the total number of images (total 'dose') was identical for each reconstruction.

The methods studied were single-axis SIRT, single-axis WBP, dual-axis WBP with recombination in Fourier space, dual-axis SIRT with recombination in Fourier-space, and Alternating dual-axis SIRT.

The last two methods have not been reported before. Fourier-combined dual-axis SIRT uses the same Fourier-space method to combine single tomograms as is used in dual-axis weighted back-projection. Alternating dual-axis SIRT is novel in that it uses both series during the iterative process.
to reconstruct a single tomogram, instead of producing two single-axis tomograms which are then combined post-facto. In order to keep the total number of iterations constant, there were only half as many iterations on each series in the Fourier-combined Dual-axis SIRT and Alternating Iterative Dual-axis SIRT compared to the single-axis SIRT method.

4. Comparison of single and dual-axis SIRT methods
The reconstructions were made using tilt series with maximum tilts of ±50°, with an angular spacing of 1° for the single-axis SIRT, and 2° for the dual-axis SIRT combined via Fourier space, and the dual-axis calculated using the newly developed algorithm. Comparisons were made after 4 iterations through to 32 iterations.

It is evident from the results displayed in Fig. 3a that at low iterations, the method of combining two single-axis tomograms is unfavourable, as the increase in reconstruction fidelity is easily offset by the penalties incurred for only having half as many iterations for each series. However, as the total number of iterations increases, the improvement gained with each iteration decreases.

Alternating dual-axis SIRT combines the advantage of filling more of Fourier space with an iterative method that ensures that information from both series is taken into account directly during the reconstruction. Although only half of the data is used at each iteration (a practical limitation imposed by the software), it is the combination of these two factors that results in a far improved fidelity over a wide range of test conditions.

Comparison of single and dual-axis SIRT and WBP methods with different tilt ranges showed that the angular range over which projections are recorded and the number of projections which are acquired within that range has a large effect on the reconstruction fidelity no matter which reconstruction method is used, and that the advantages of switching to dual-axis methods are most obvious at lower tilt ranges, see Fig. 2.

5. Handling of noisy data
Weighted back-projection, single-axis SIRT and dual-axis SIRT reconstructions were performed on test data, using tilt series of ±70°, with 1° sampling for weighted back projection and single-axis SIRT, and 2° sampling for dual-axis SIRT. A noise component was added as a percentage of the true signal.

Any increase in noise relative to the available signal will cause a reduction in the fidelity of the reconstruction. It can be seen from Fig. 3b that using iterative methods improves the reconstruction from a noisy data set, and the dual-axis algorithm seems to perform remarkably well, with very little reduction in fidelity even with large amounts of noise. Clearly the increased 'spread' of data in Fourier
space means that even with the same signal-to-noise (SNR) ratio in the original images, the SNR in the final tomogram will be increased due to the increased sampling of Fourier space overall. The combination of the two data sets may improve the ability of the iterative algorithm to overcome local minima (often caused by noise) in the difference minimisation routine. The introduction of new data at each iteration prevents the program from becoming trapped in these local minima. Added to this is possibly a native smoothing caused by the resampling of data which happens whenever the angle between the series is not exactly 90°.

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**Figure 3.** a) Comparison of single and dual-axis variations of SIRT over a range of iterations. b) Comparison of single and dual-axis tomographic reconstructions, using both WBP and SIRT methods, for different levels of added noise with respect to signal. In each case the tilt range is ±70° and the total number of images in each data set is constant. The comparison used is the Pearson’s Correlation Coefficient (PCC) with respect to the original test data.

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6. **Summary**

It has been shown on test data that alternating dual-axis SIRT has several advantages over conventional iterative methods and weighted back projection, and that this method allows particular scope for improvement of reconstructions in cases where reconstruction fidelity is limited by low signal-to-noise ratio. Using this algorithm, it is possible to improve the quality of reconstructions without increasing the number of projections acquired, and therefore this method seems very suited for low-dose tomography.

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7. **References**

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