We Focus on Distance

Algorithms for Blur-Correction in 3D Electron and Soft X-ray Microscopy

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Outline

Motivation and some background
- Blurring in Transmission Electron Microscopy
- Related Work

Our Results/Contribution
- Analysis of Defocus Gradient Corrected Backprojection
- Fully 3D Frequency Distance Relation
The point spread function (PSF) describes the response of an imaging system to a point source or point object. The degree of spreading (blurring) of the point object is a measure of the quality of an imaging system. In electron microscopy an image obtained resembles a convolution of the ideal projection image with a point spread function.
Blurring: X-ray point spread function

Plots of the normalized soft X-ray point spread function near the geometric focal point of a zone plate objective for defocus 0, 5 and 10 microns. Inserted on the right are the corresponding images acquired by the X-ray microscope at the BESSY II electron storage ring (Berlin, Germany). [Figure courtesy of Roberto Marabini]
A contrast transfer function (CTF) is the Fourier transform of a point spread function. It alters the various frequencies that make up the signal:

- modulating the amplitude at some frequencies
- changing the sign at some frequencies
- completely eliminating some frequencies

As long as the contrast transfer function affects each layer of an object in the same way it does not cause a problem in reconstruction since the blurring and integration commute and one can deblur the projections and then reconstruct.
Blurring: CTF

\[ \int v(x_1, x_2, x_3) \ast h(x_1, x_2) \, dx_3 \]

\[ \int V(\xi_1, \xi_2, x_3) H(\xi_1, \xi_2) \, dx_3 \]

\[ H(\xi_1, \xi_2) \int V(\xi_1, \xi_2, x_3) \, dx_3 \]

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Blurring: Distance-dependent CTF

- CTF changes its shape with the distance from the electron source.
- Each object layer is blurred by a slightly different function.

The effects of distance dependence on CTF blurring become more significant as the desired resolution of reconstructions increases and when larger specimens are imaged.
Example #1
Cross sections of (a) phantom, (b) reconstruction with correction for the distance-dependent contrast transfer function, (c) reconstruction with correction appropriate for the central layer of the specimen, and (d) reconstruction with no correction for the contrast transfer function.
Related Work: DGCBP


**Frequency Distance Relation (FDR)**

The values of the frequencies of each coefficient in the 3D Fourier transform of the projection data obtained from around a single axis rotation are directly related to the distance from the electron source at which the points in the specimen contribute to the coefficient.

This implies that the collected data can be corrected to approximate true projections.
Part A: Analysis of Defocus Gradient Corrected Backprojection (DGCBP) like method

- our contributions
- illustration of results

Part B: Fully 3D extension to Frequency Distance Relation (FDR) method

- our contributions
- illustration of results
PART A:
Defocus Gradient Corrected Backprojection
Part A: Main ideas

In mathematical ideal we can reconstruct an imaged object exactly by applying
• projection
• backprojection
• deblurring

We want to show that similar is true for distance-dependent model:
• distance-dependently blurred projection
• distance-dependent backprojection
• deblurring
should allow us to reconstruct the original object.
Part A: Main ideas

1. show that our projection and backprojection operators are equivalent to the ones that are typically used in the field of image reconstruction (so that we can use established theorems)

2. demonstrate that the **distance-dependent backprojection** applied to the **distance-dependent projections** of an object is approximately equal to the **backprojection** applied to the **projections** of the same object
Part A: EM Operators

**Natterer and Wübbeling**

\[ \mathcal{N} \text{ Ray transform} \quad \mathcal{W} \text{ Backprojection} \]

Let \( f : \mathbb{R}^3 \rightarrow \mathbb{R} \). Then we have

\[
 f = \frac{1}{16\pi^3 \sqrt{2\pi}} \left[ \mathcal{F}^{-1} \hat{r} \right] * \mathcal{WN} f,
\]

where \( \hat{r} \) is defined by \( \hat{r} (\xi_1, \xi_2, \xi_3) = \sqrt{\xi_1^2 + \xi_2^2 + \xi_3^2} \).

**Kazantsev et al**

\[ \mathcal{R} \text{ rotation} \quad \mathcal{C} \text{ compression} \quad \mathcal{P} = \mathcal{CR} \text{ projection} \]

\[ \mathcal{S} \text{ spreading} \quad \mathcal{T} \text{ totaling} \quad \mathcal{B} = \mathcal{TS} \text{ backprojection} \]
Part A: EM Operators

Natterer and Wübbeling

**Ray transform** \( \mathcal{N} \)

\[
\text{Let } f : \mathbb{R}^3 \rightarrow \mathbb{R}. \text{ Then we have } f = \frac{1}{16\pi^3\sqrt{2\pi}} \left[ \mathcal{F}^{-1} \hat{r} \right] \ast \mathcal{WN} f,
\]

where \( \hat{r} \) is defined by \( \hat{r}(\xi_1, \xi_2, \xi_3) = \sqrt{\xi_1^2 + \xi_2^2 + \xi_3^2} \).

Kazantsev et al

**Rotation** \( \mathcal{R} \)

**Compression** \( \mathcal{C} \)

**Projection** \( \mathcal{P} = \mathcal{C}\mathcal{R} \)

**Spreading** \( \mathcal{S} \)

**Totaling** \( \mathcal{T} \)

**Backprojection** \( \mathcal{B} = \mathcal{T}\mathcal{S} \)

\[
\mathcal{B}\mathcal{P} = \mathcal{WN},
\]
Theorem

Let $f : \mathbb{R}^3 \to \mathbb{R}$. Then we have

$$f = DBP f,$$

for which we define a deblurring operator $\mathcal{D}$, for any $v \in \mathbb{R}^3 \to \mathbb{R}$, as

$$\mathcal{D}v = \frac{1}{16\pi^3 \sqrt{2\pi}} \left[ \mathcal{F}^{-1} \hat{r} \right] * v,$$

where $\hat{r}$ is defined by $\hat{r}(\xi_1, \xi_2, \xi_3) = \sqrt{\xi_1^2 + \xi_2^2 + \xi_3^2}$, and
Distance-dependent compressing operator

A projection data set that consists of all distorted 2D projections (micrographs) is defined as a compression of $Rv$ after it has been convolved with a point spread function $h$. The \textit{distance-dependent compressing operator} $C_h$ is defined by

$$C_hw = C[w \ast h]$$

Distance-dependent projection operator

The \textit{distance-dependent projection operator} $P_h$ is defined by

$$P_h = C_hR.$$
Part A: Distance-Dependent EM Operator

Distance-dependent spreading back operator

We define the \textit{distance-dependent spreading back operator} $S_h$ by

$$S_h g = \mathcal{F}_w^{-1} \left[ \frac{\mathcal{F}_G g}{\mathcal{F}_V h} \right].$$

Distance-dependent backprojection operator

The \textit{distance-dependent backprojection operator} is

$$B_h = \mathcal{T} S_h.$$
Irrespective of the choice of the point spread function $h$, we are able to show that for all $v$

$$BPv \approx B_hP_hv,$$

where $\approx$ stands for \textit{approximately equal}.

This, combined with the inversion formula for the ray transform, implies that

$$DB_hP_hv \approx v,$$

and so the required volume can be approximated from the distance-dependently blurred projection data $P_hv$ by applying to it first the distance-dependent backprojection operator and then the deblurring operator.
Part A: Distance-Dependent Inversion

\[ D \quad B_n \quad P_{hv} \quad \approx \quad v \]
Part A: Phantom

We Focus on Distance
PART B:

Frequency Distance Relation
Part B: Operators

We use the same operators for modeling forward projections as in the previous method:

**EM Projection**

\[ \mathcal{R} \text{ rotation} \quad \mathcal{C} \text{ compression} \quad \mathcal{P} = \mathcal{CR} \text{ projection} \]

**EM distance-dependent Projection**

\[ \mathcal{R} \text{ rotation} \quad \mathcal{C}_h \text{ compression} \quad \mathcal{P}_h = \mathcal{C}_h\mathcal{R} \text{ projection} \]
We image a molecule that is an impulse located at \((\hat{x}_1, \hat{x}_2, \hat{x}_3)^T\):

\[
\kappa(x_1, x_2, x_3) = \delta(\hat{x}_1 - x_1) \delta(\hat{x}_2 - x_2) \delta(\hat{x}_3 - x_3).
\]
We image a molecule that is an impulse located at \((\hat{x}_1, \hat{x}_2, \hat{x}_3)^T\):

\[
\kappa(x_1, x_2, x_3) = \delta(\hat{x}_1 - x_1) \delta(\hat{x}_2 - x_2) \delta(\hat{x}_3 - x_3) .
\]

The blur-free projection data \(P_\kappa\) of our molecule \(\kappa\) is

\[
[P_\kappa](\theta, \phi, x_1, x_2) = \delta(\hat{x}_1(\theta, \phi) - x_1) \delta(\hat{x}_2(\theta) - x_2) .
\]
We image a molecule that is an impulse located at \((\hat{x}_1, \hat{x}_2, \hat{x}_3)^T\):

\[\kappa(x_1, x_2, x_3) = \delta(\hat{x}_1 - x_1) \delta(\hat{x}_2 - x_2) \delta(\hat{x}_3 - x_3).\]

The blur-free projection data \(P_\kappa\) of our molecule \(\kappa\) is

\[\left[ P_\kappa \right] (\theta, \phi, x_1, x_2) = \delta(\hat{x}_1(\theta, \phi) - x_1) \delta(\hat{x}_2(\theta) - x_2).\]

The distance-dependently blurred projection data \(P_{h\kappa}\) of \(\kappa\) can is

\[\left[ P_{h\kappa} \right] (\theta, \phi, x_1, x_2) = h(x_1 - \hat{x}_1(\theta, \phi), x_2 - \hat{x}_2(\theta), \hat{x}_3(\theta, \phi)).\]
Part B: Correction I

The 4D Fourier transform of $\mathcal{P}_\kappa$ is

$$[\mathcal{F}_4 \mathcal{P}_\kappa] (\Theta, \Phi, \xi_1, \xi_2) = \frac{1}{(2\pi)^2} \int_0^{2\pi} \int_0^\pi e^{-i(\bar{x}_1(\theta, \phi)\xi_1 + \phi\Phi)} d\phi \ e^{-i(\bar{x}_2(\theta)\xi_2 + \theta\Theta)} d\theta.$$ 

The 4D Fourier transform of $\mathcal{P}_h \kappa$ is

$$[\mathcal{F}_4 \mathcal{P}_h \kappa] (\Theta, \Phi, \xi_1, \xi_2) = \frac{1}{(2\pi)^2} \int_0^{2\pi} \int_0^\pi H (\xi_1, \xi_2, \bar{x}_3 (\theta, \phi)) e^{-i(\bar{x}_1(\theta, \phi)\xi_1 + \phi\Phi)} d\phi \ e^{-i(\bar{x}_2(\theta)\xi_2 + \theta\Theta)} d\theta.$$
**Theorem**

The 4D Fourier transform of the distance-dependently blurred projection data is related to the 4D Fourier transform of blur-free projection data:

\[
[F_4P_{h\kappa}] (\Theta, \Phi, \xi_1, \xi_2) \approx H \left(\frac{\Phi}{\xi_1}\right) [F_4P_{\kappa}] (\Theta, \Phi, \xi_1, \xi_2).
\]
The Fourier coefficients for the corrected projection data of $\nu$ can be obtained by a careful division, so as not to amplify the noise in the data, of the Fourier coefficients of the distance-dependently blurred projection data $P_h \nu$ by the CTF $H$; i.e., as

$$ [\mathcal{F}_4 P_\kappa] (\Theta, \Phi, \xi_1, \xi_2) \approx [\mathcal{F}_4 P_h \nu] (\Theta, \Phi, \xi_1, \xi_2) / H \left( \xi_1, \xi_2, -\frac{\Phi}{\xi_1} \right). $$
[F_4 P_\kappa] (\Theta, \Phi, \xi_1, \xi_2) 
\approx [F_4 P_h \nu] (\Theta, \Phi, \xi_1, \xi_2) / H \left( \xi_1, \xi_2, -\frac{\Phi}{\xi_1} \right).

This correction gives us approximations on the true projection data through the molecule.

The molecule can be reconstructed, based on such corrected projection data, using ANY reconstruction algorithm.
We discussed the distance-dependent nature of the blurring that occurs in microscopic imaging.

We provided mathematical analysis of an algorithm for correction of distance-dependent blurring suggested by Jensen and Kornberg (2000). We developed operators that model data collection process and showed that the distance-dependent projections can be inverted in the reconstruction process by distance-dependent backprojection followed by deblurring.

We provided an extension of frequency distance relation to the case of date collected from arbitrary directions in 3D. This provides a correction of distance-dependently blurred projection data, which then can be used by any known reconstruction algorithm.
Related Work

- D.J. DeRosier, *Correction of high-resolution data for curvature of the Ewald sphere*, Ultramicroscopy (2000)
- J.N. Dubowy, G.T. Herman, *An approach to the correction of distance-dependent defocus in electron microscopic reconstruction*, ICIP 2005
- J.Klukowska, G.T.Herman, I.G.Kazantsev, *Correction of Distance-Dependent Blurring in Projection Data for Fully Three-Dimensional Electron Microscopic Reconstruction*, (submitted to ISBI 2010)
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• I.G. Kazantsev, G.T. Herman, L.Cernetic, *Backprojection-based reconstruction and correction for distance-dependent defocus in cryoelectron microscopy*, ISBI 2008
• I.G. Kazantsev, J.Klukowska, G.T. Herman, L.Cernetic, *Fully three-dimensional defocus-gradient corrected backprojection in cryoelectron microscopy*. (submitted to Ultramicroscopy)
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