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## **Image defects and frame alignment**

Wed, 08/01/2018 - 15:34

#1

anaa2

Image defects and frame alignment

Hi,

Has anyone come across this image defects (weird white spirals) when using unblur (see image), and how can one correct for it? Could this cause an issue in frame alignment?

Best wishes,

Noor

Update: I've Just attached two images of a power spectrum post drift correction, both through cisTEM. It appears by removing outlier pixels using remove\_outlier\_pixels command. has improved it a bit. This was done under the default parameters. Is there a way to improve it further?

Another question, I have a pixel size of 0.66 angstroms per pixel, so why is Nyquist to 2.80 angstroms?

Thank you so much,

Noor

File:

- Screen Shot 2018-08-01 at 19.50.14.png
- normal.png
- outlier\_pixel\_removed.png

timgrant

Hi Noor,

Hi Noor,

That looks like ringing artifacts, if you had strong features in the image (like very bright pixels), they could ring after Fourier filtering / possibly phase shifting. These artifacts themselves probably don't cause problems, but the bright pixels that cause them may.

Is this the unblur from cisTEM, or is it the old stand alone unblur?

Thanks,

Tim

Wed, 08/01/2018 - 16:29 (Reply to #2)

anaa2

Hi Tim,

Hi Tim,

This is unblur 1.02 run through RELION GUI. Is there way to correct for these bright pixels?

Best wishes,

Noor

timgrant

Hi,

Hi,

You could try removing outlier pixels from the movie frames prior to aligment, there is a command line program in cisTEM to do this (remove\_outlier\_pixels). The unblur version included with cisTEM is quite a bit faster than the 1.02 version, and likely gives better results. It also automatically removes outlier pixels and so will probably not have this issue.

Cheers,

Tim

Wed, 08/01/2018 - 16:38 (Reply to #4)

anaa2

Hi Tim,

Hi Tim,

Thank you so much. I will do this tomorrow. I will get cisTEM installed. Again, many thanks.

Best wishes,

Noor

Thu, 08/02/2018 - 02:46 (Reply to #5)

Axel

outliers

This is great, I have a couple datasets with this problem, and will test it out.

Axel

Fri, 08/03/2018 - 15:07

timgrant

Hi Noor,

Hi Noor,

If the Nyquist is higher resolution is 2.8A, then cisTEM resizes the power spectra to 2.8A, This is done, because having a higher resolution Nquist is rarely useful in seeing things. This resize is only done for the power spectra, it does not affect the actual data.

If you are worried about fixed pattern noise (like the hot pixels), the parameters I would play with are "minimum shift" -try to increase it to 5 or so, and bfactor - try to reduce it a bit. See if this improves the alignment a little.

Cheers!

Tim

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