3D refinement/CTF refinement issue

Tue, 08/17/2021 - 09:45

#1

charlesb

3D refinement/CTF refinement issue

Hi,
I am curretrly processing a particle stack extracted from relion with cisTEM 1.0.
I first did an auto refine that gave me a final resolution of 2.92A.

I then wanted to make a CTF refinment usinf the cistem "manual refine" option. I used pretty much the same parameters as the auto-refine did, only with the additon of the CTF refinement but I can't get a resolution higher than 3.94A with this method. I tried several parameters and it was always the same.

I may by wrong somewhere in the process, but I can't figure it out.
Do you have any idea how I could resolve this ?

Thanks!
Charles

Fri, 08/20/2021 - 10:55

#2

timgrant

Hi Charles,

Hi Charles,

Hmm - What is the high resolution limit you are using? What is the molecular weight of your particle?

Thanks,

Tim

Mon, 08/23/2021 - 09:56 (Reply to #2)

charlesb

Hi Tim,

Hi Tim,

I've tried several different high resolution limit for my refinement, i would say from 5A to 30A (the basic value) and it was always pretty much the same.
I'm working on human ribosome particle so the molecular weight is 4.3MDa.

Thanks,

Charles

Mon, 08/23/2021 - 12:43

#4

timgrant

Hi Charles,

Hi Charles,

The CTF Refinement needs high res info to work. If you got 2.92 from your autorefine you should be safe to try using 3.5A for the CTF refinement. Try 3.5A and see how that goes.

Also, It's probably a good idea to only refine the CTF when you do that, so just do a local refinement, then unclick the psi,theta,phi,x-shift, y-shit and just do the CTF refinement with a high resolution limit of 3.5A.

Let me know if that works better!

Cheers,

Tim

Fri, 08/27/2021 - 09:54

#5

charlesb

Hi Tim,

Hi Tim,

These parameters were way better, it reached a final resolution of 2.8A. It was definitly a good idea to remove all the psi, theta,... parameters and only do the CTF refinement.

Thank you for your help !

Cheers,

Charles

Source URL:https://cistem.org/3d-refinementctf-refinement-issue