

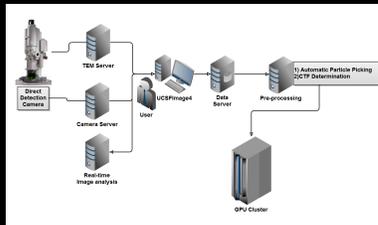
# Combining electron counting and beam-induced motion correction to achieve near atomic resolution single particle cryoEM

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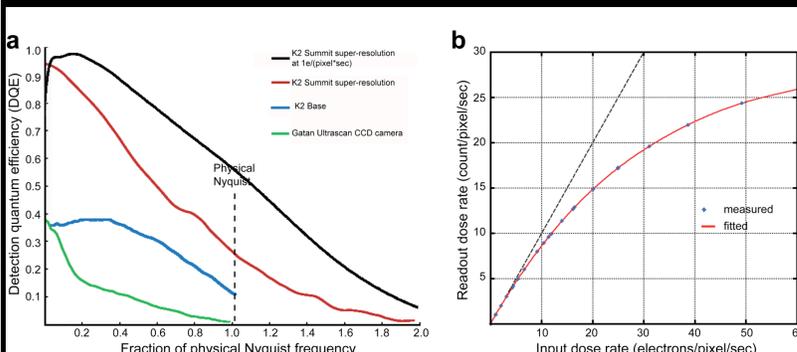
## K2 camera with the Tecnai Polara



- \* K2 is fully embedded into our data acquisition procedures, both with UCSFTomo and UCSFImage4.
- \* Motion correction is implemented both on-the-fly with data acquisition and during image processing.
- \* Identified optimal dose rate for data collection.

Xueming Li, Shawn Zheng,  
Paul Mooney, Chris Booth

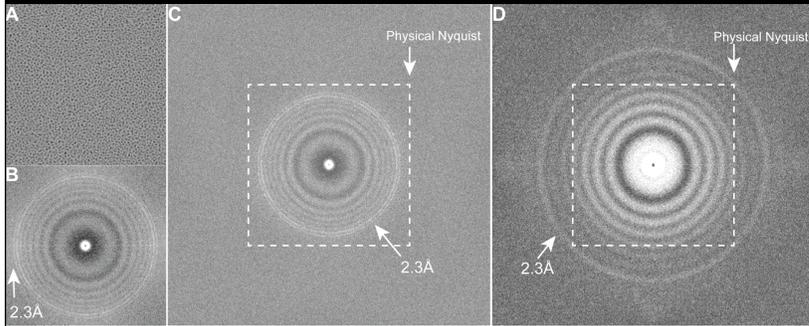
## DQE and dose rate



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- \* Motion correction is implemented both on-the-fly with data acquisition and during image processing.
- \* Identified optimal dose rate for data collection.
- \* DQE at 1 e<sup>-</sup>/(pixel\*sec) dose rate is close to an ideal camera.

Xueming Li, Paul Mooney

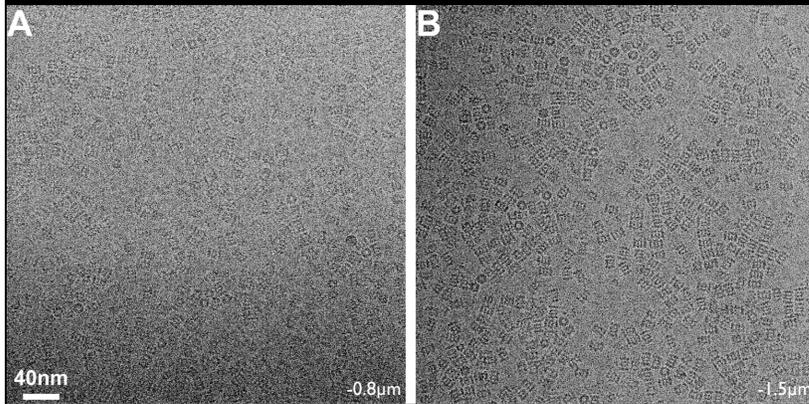
## K2 is suitable for high-resolution low-dose imaging



- \* thin Pt/Ir film recorded at a magnification of 39kX,  $\sim 1\text{\AA}/\text{pixel}$ ;
- \*  $\sim 17\text{e}/\text{\AA}^2$  on specimen and  $\sim 17\text{e}/\text{pixel}$  on camera;
- \* D: super resolution image, 23kX,  $\sim 1.8\text{\AA}/\text{pixel}$ , dose rate  $\sim 31\text{ e}/\text{pixel}\cdot\text{sec}$ ,  $\sim 28\text{e}/\text{\AA}^2$  on specimen and  $\sim 93\text{ e}/\text{pixel}$  on camera;

Chris Booth

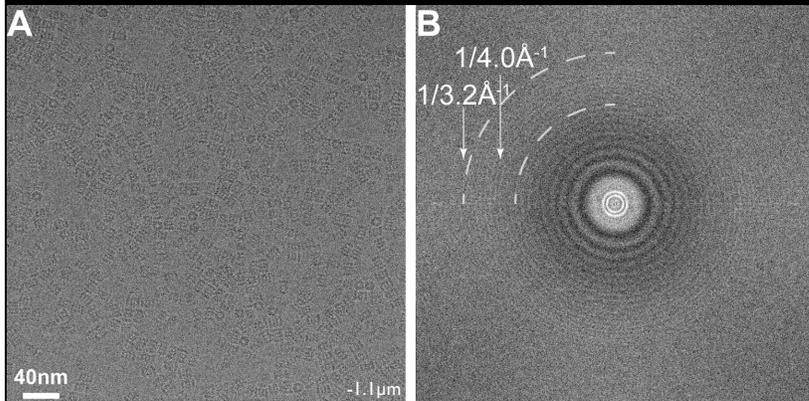
## K2 image of frozen hydrated protein samples, T20S



- \* 300kV, 31kX mag,  $\sim 10\text{e}/\text{pixel}/\text{sec}$ ;  $\sim 1.2\text{\AA}/\text{pixel}$ ,  $25\text{e}/\text{\AA}^2$ , 3.5sec exposure;

Xueming Li, Kiyoshi Egami

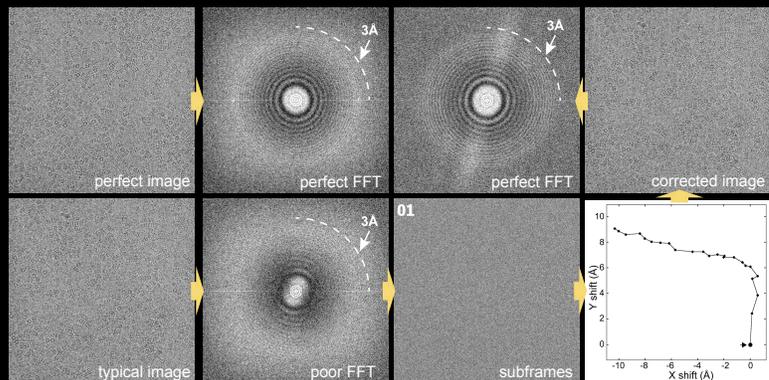
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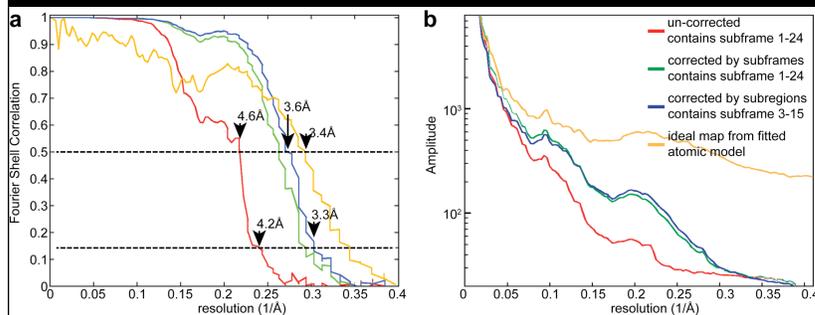
## Charge induced image blurring can now be corrected



\* Instead of recording one image at a time, we fractionate a single exposure into 24 subframes. By re-aligning all subframes, we can correct motion induced image blur.  
 \* We can restore most of images to near perfect quality suitable for atomic resolution single particle cryoEM, ground breaking leap in high-resolution image acquisition.

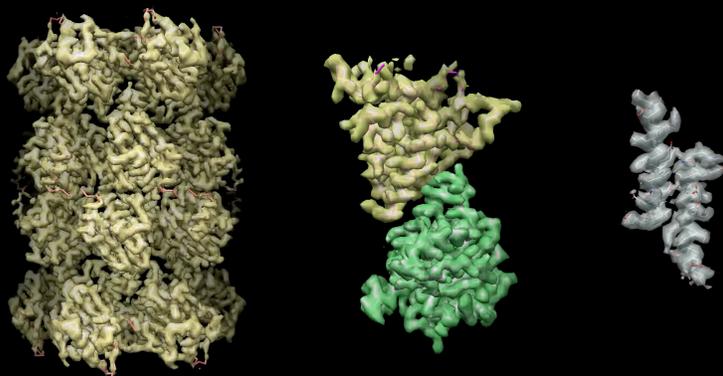
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## Beam induced motion



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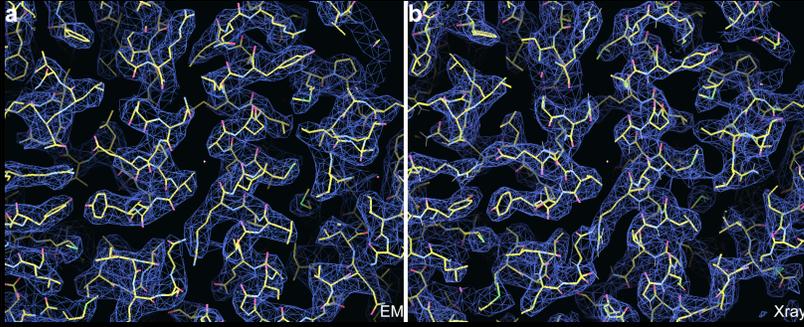
## We achieved resolution comparable with X-ray crystallography



\* We determined a 3D reconstruction of archaeal 20S proteasome to the resolution of  $\sim 3.3$  Å, comparable to the resolution of X-ray crystal structure, 3.4Å.

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## Acknowledgements

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