New Methodologies for Preparing and Imaging Cryo-EM Samples.

Gan Li¹, Michael S Westphall², Mr Austin Z. Salome², Kenneth W Lee², Joshua J Coon³, Timothy Grant¹

¹Morgridge Institute for Research, UW-Madison, ²UW-Madison, ³UW-Madison, Morgridge Institute for Research
gli243@wisc.edu

We have investigated using native mass spectrometry to land isolated protein complexes onto EM grids, subsequently solving their structure by negative stain EM. I will discuss these results and the potential for MS-EM to provide ideal samples for single-particle cryo-EM. I will also discuss a new imaging method we are developing, “Defocus Sweep Imaging”, which, via computational microscope control, allows the microscope defocus to be changed as movies are recorded. This allows high resolution close-to-focus information to be recorded at the beginning of an exposure when radiation damage is low, and lower resolution far-from-focus data to be recorded at the end of the exposure when the sample is highly damaged.

Figure 1