Your Name: Jess Lovescryo
Your PI's Name: Rea
Your collaborator Name(s): Chelsea Doescience
Collaborator PI's Name(s): Parker
Project Name (3-5 words): Apoferritin for benchmarking

Describe specimen, biological relevance, and brief background:

Specimen: Equine spleen apoferritin has a molecular weight of about 444,000 and is composed of 24 subunits (MW 18,500). These are arranged in 432 symmetry to form a nearly spherical hollow shell with outside and inside diameters approximately 130 Å and 75 Å respectively. The large cavity inside the molecule can store up to 4,500 Fe atoms.
Background: Until recent years, apoferritin had remained a difficult structure to determine by cryo-EM, even with the advent of direct electron detectors, because the contrast in individual particle images was insufficient to resolve the characteristic structural motif of a four α-helix bundle. Since then, multiple groups have reached resolutions better than 3 angstroms, thus establishing apoferritin as an appropriate benchmarking protein for TEMs. The goal of this project is to assure that our current instrumentation can achieve comparable resolutions to other TEM setups.
Biological significance: The ferritins are a class of iron-storage proteins in both prokaryotes and eukaryotes, keeping iron in a soluble and non-toxic form. Ferritin that is not combined with iron is called apoferritin. Apoferritin catalyzes the oxidation of Fe(II) which it retains inside the molecule as the ferric hydrolysate.

Specimen molecular weight: 440kD
Specimen dimensions: 13 nm sphere

Data collection on either the Talos Arctica or Titan Krios requires upload of cryo images (with scale bars) or 2D classes with a description of your previous TEM usage and processing scheme:
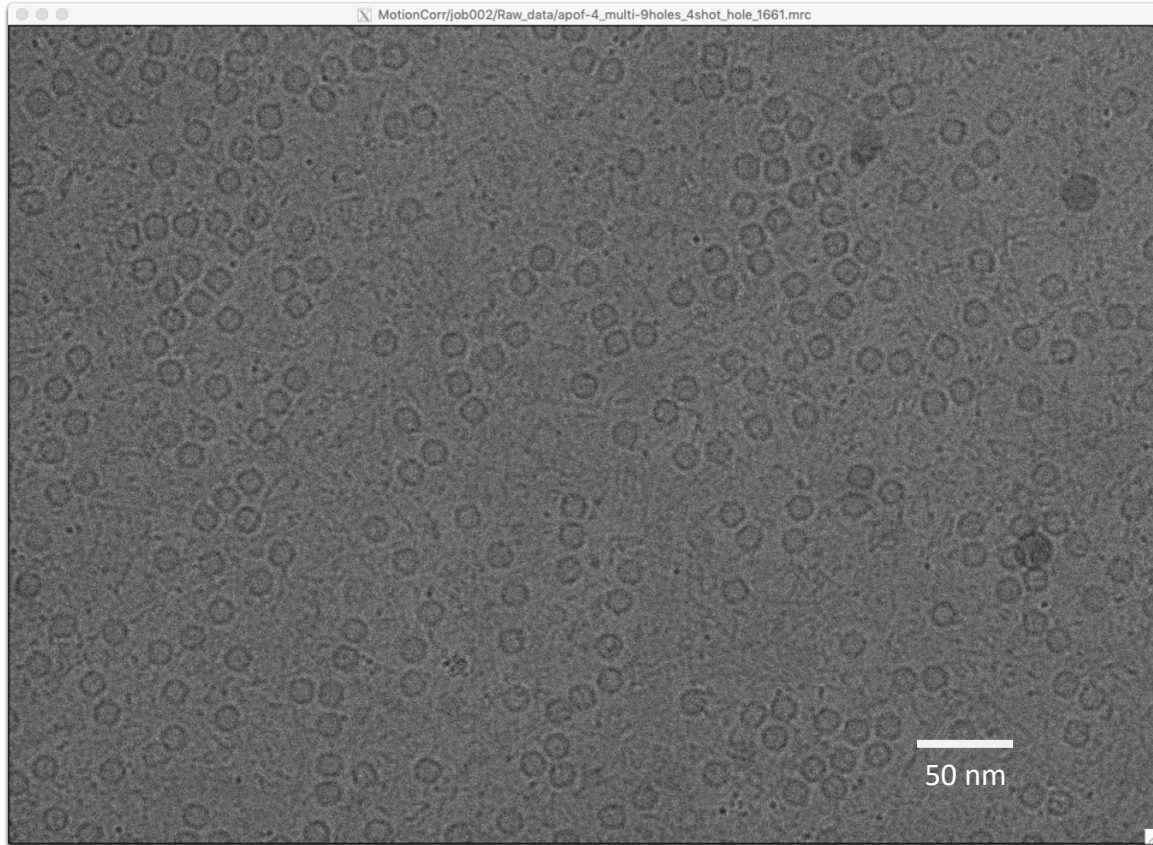
Cryo image of apoferritin collected on a 300kV Titan Krios utilizing multi-shot/multi-hole data acquisition. Sample concentration was a bit lower than hoped for, but I was able to get decent 2D classes as shown here. I used Relion 3.0 to process the data with the exception of particle-picking, which was performed in crYOLO.
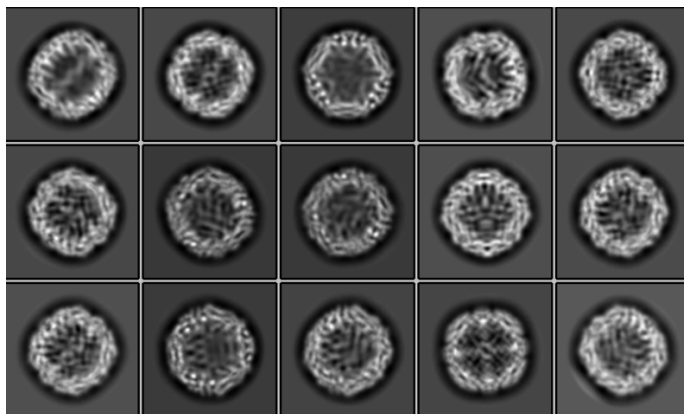
Requested microscope (Talos Arctica or Titan Krios): Titan Krios
Phase plate (yes or no): no
Requested number of sessions (up to 4): 2