## **Supplementary Information**

## Nanoscale Characterization of the Biomolecular Corona by Cryo-Electron

## Microscopy, Cryo-Electron Tomography, and Image Simulation

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**Supplementary Figure 1.** (a) Protein corona (hard and soft) formation on the surface on nanoparticles (no wash) (b) hard protein corona formation on the surface of nanoparticle (with washing step).



**Supplementary Figure 2.** Cryo-EM images of the PS-COOH NPs (a) original plasma diluted in Sorensen's phosphate buffer to 50%. There is little evidence for presence of large cluster and aggregate; (b) NPs with corona in 2% solution showing the distribution of biomolecules and their association with the surface of the NPs. Similar to the plasma, there is little evidence for large clusters.





































Supplementary Figure 3. TEM images showing the 3D tomographic volume slices of 10% corona coated NPs. The size of each image panel is  $1\mu m$ 

































































































































Supplementary Figure 4. TEM images showing the 3D tomographic volume slices of 50% corona coated NPs. The size of each image panel is  $1\mu m$ .