AFM and cryoEM studies of the giant Mimivirus

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Mimivirus has a 7,400 Å diameter virion and a 1.2 Mbp dsDNA genome containing many genes that are not normally in other viruses [1-3]. The genome size, the physical size of the virus and the ability to perform many of the functions of a simple cell make it difficult to define the boundary between a virus and a cell [3]. The Mimivirus major capsid protein was found to be homologous to the major capsid proteins of Paramecium bursaria Chlorella virus 1 and other large dsDNA viruses [4-5]. Therefore, the structure of Mimivirus capsomers had been assumed to be a trimeric double “jelly roll” folds with a diameter of ~75 Å and thickness of ~70Å as in the aforementioned large dsDNA icosahedral viruses. Initial cryo-electron microscopic (cryoEM) studies [2, 6] have shown that Mimiviruses have multiple layers of proteins and lipid membranes that surround its nucleocapsid. The viral surface is covered by a dense layer of 1200 Å-long fibers creating difficulties for reconstructing a three dimensional map of the virus [2]. This problem has been partially overcome by digestion of the fibers with enzymes. Here we report the use of atomic force microscopy (AFM) and cryoEM to investigate the structure of Mimiviruses. We show that the capsomer separation is about 140 Å, twice as big as observed in other large dsDNA icosahedral viruses indicating a different structural arrangement of the double jelly-roll motifs within each capsomer. We also confirm that Mimivirus has a unique pentameric vertex identified by a star-fish shaped feature facilitating the delivery of the genome to the host [6-7]. This “star-fish” shaped feature distorts the previously-assumed icosahedral symmetry of Mimivirus [2]. CryoEM reconstruction based on only five-fold showed the nucleocapsid has a defined enveloped structure with one depressed face towards the starfish shaped feature associated vertex.

References

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**FIG. 1.** AFM images of de-fibered Mimiviruses. (a) AFM image of de-fibered virus showing one face of the virion. (b) A starfish-shaped feature on a de-fibered virus. The scale bars represent 1000 Å.

**FIG. 2.** CryoEM reconstruction of Mimivirus applying only five-fold symmetry averaging. Surface shaded rendering (a,c) and central slices (b,d) of cryoEM reconstruction of untreated Mimivirus. (a,b) look down the starfish-shaped feature associated vertex and (c,d) looking from one side. The coloring is based on radial distance from the center of the virus. The scale bars represent 1,000 Å.