

Preliminary Results

Our preliminary results are largely from the flight muscles insects that utilize asynchronous flight muscle. These include the Hemipteran *Lethocerus indicus*, and the Dipteran *Drosophila melanogaster*. We plan to investigate other vertebrate striated muscle thick filaments.

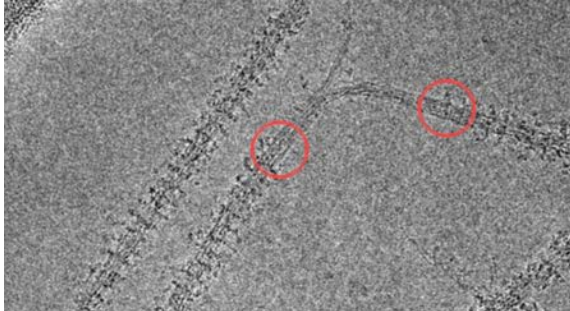


Figure 1. Electron micrograph of a frozen-hydrated *Lethocerus* thick filament recorded in integration mode on the DE-64. The red circles outline the first crown (crown-0). At the end of the bare zone. Heads are ordered into shelves of density called “crowns”. Although all the parts of the thick filament showing ordered myosin heads are useful for a helical reconstruction, only those for which crown-0 can be identified are useful for the asymmetric reconstruction. If the asymmetric reconstruction shows sufficient variability along the filament, potentially classification can place filament segments where their placement is not defined by the image itself.

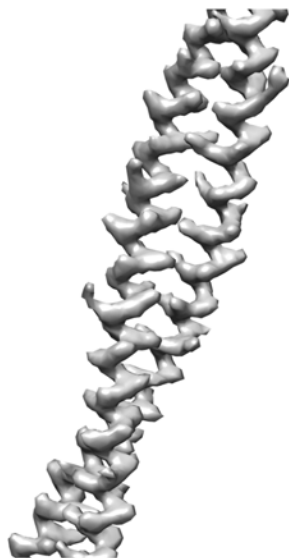


Figure 3. A portion from the 2.8Å reconstruction of flight muscle thick filaments from *Drosophila melanogaster* showing well-resolved side chains. This region from the myosin α -helical coiled coil tail but not the same region shown in Figure 2.

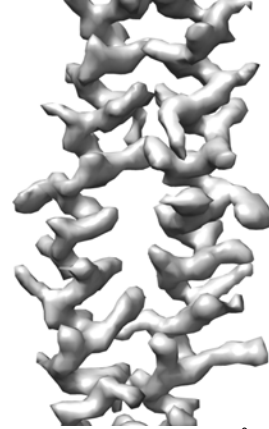


Figure 2. A portion of the 2.7 Å reconstruction (FSC calculation) from *Lethocerus indicus* showing well resolved side chains. This region from the myosin α -helical coiled coil tail.

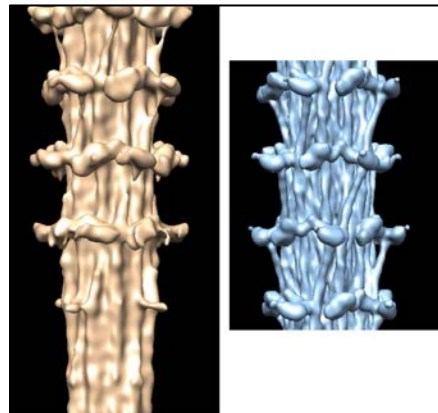


Figure 4. Comparison of *Lethocerus* thick filaments reconstructions. (left) The crown-0 reconstruction which imposed only rotational symmetry on 542 motifs (2,168 asymmetric units), reached a resolution of 16Å by FSC criteria. Images were recorded in integration mode on a DE-64 detector. (right) Previously published 6Å helical reconstruction from ~24,000 segments, low pass filtered to 16Å. Images recorded on a DE-20 direct electron detector operated in integration mode. The DE-20 detector is inferior to both the DE-64 and K3. Both reconstructions show the poorly ordered myosin heads projecting from the backbone about equally, but also resolve the S2 linkage from the heads to the backbone, though not as well as in the crown-0 reconstruction with symmetry imposed.

Bibliography

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