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Structure and Function of Icosahedral Pyruvate Dehydrogenase Complexes from *B. stearothermophilus*.

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Electron cryo-microscopy of “single particles” is useful to determine the 3D architectures of complex cellular assemblies. The pyruvate dehydrogenase (PDH) multienzyme complex, consisting of E1, E2 and E3 enzymes, converts pyruvate to acetyl CoA. We present a model for an 11 MDa icosahedral PDH sub-complex, obtained by docking atomic coordinates of E1 and E2 into a 28 Å structure derived from electron cryo-microscopy. The architecture provides an efficient mechanism for active site coupling by movement of the lipoyl domains in the annular region between the inner E2 core and outer E1 shell of the complex. Analysis of E2 inner cores decorated with increasing amounts of E1 or E3 enzymes indicate a fundamental role for the interdomain linkers in maintaining the annular gap. Our analysis of 42945 images of the E2 core indicates that the top 139 particles leads to a dramatically improved map. Densities corresponding to known alpha-helices can be visualized in a map arising from as few as 9 particles. Application of such methods to other complexes likely will facilitate docking of X-ray atomic coordinates of individual proteins into the density maps obtained by electron cryo-microscopy.