Exploring Structural Flexibility and Classification Methods for Identifying Intermediate States of Hepatic ABCB Transporters

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Cryo-EM advances in Single Particle Analysis focuses nowadays in the study of the diverse conformations of the macromolecules beyond traditional 3D classification. Continuous flexibility methods, such as cryodrgn or Zernike3D [1, 2], allow to study intermediate states as well as transitions between them, characterizing the full structural spectrum. In this work we have combined flexibility methods, together with static classification consensus strategies, to study the conformational landscape of ABCB4. This receptor is an ATP-binding cassette transporter expressed in hepatocytes that plays a crucial role in the formation of bile by translocating phosphatidylcholine (PC) into canaliculi [3]. While previous studies [4] have provided valuable insights into the structural basis of PC translocation, the precise transition mechanism between discrete conformations remains elusive. Although further characterization is needed to fully understand the mechanism of PC translocation, preliminary studies using these developing flexibility analysis tools [1,5] revealed what might be possible new intermediate states.

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