

The Active Site of an ATP-binding Cassette from an ABC Transporter Shows an Induced-Fit Effect with Implications in the Active Transport Reaction

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Introduction: ATP-binding cassette (ABC) transporters are mechanochemically coupled polypeptide complexes responsible for transmembrane solute translocation against cellular concentration gradients. In prokaryotes and archaeobacteria, these complexes are generally composed of four subunits, two of which are membrane-embedded proteins with 6 transmembrane α -helices each that are thought to determine substrate specificity and trajectory. The other two subunits are peripherally associated with the membrane-spanning components and mechanically couple ATP hydrolysis to solute translocation. In higher eukaryotes, these components are fused into one polypeptide containing multiple domains and have been linked to a number of human diseases, the most notable of which are cystic fibrosis and multidrug resistance in advanced tumor cells.

As the individual domains are present as separate genes for many of the prokaryotic transport systems, the roles of the different domains can be studied individually. Therefore, we sought to identify the structural substates of an ATP-binding cassette from an ABC transporter in order to relate the structure of these intermediates to their role in the active transport reaction cycle. We solved the X-ray crystal structure of the ATP-binding cassette from the leucine-isoleucine-valine ABC transporter from *M. jannaschii*, LivF, bound to MgADP and nucleotide free, respectively.

Conclusions: Comparison of these respective structures shows that several conserved active site residues essential for nucleotide hydrolysis are substantially withdrawn from the active site in the apo-form of the enzyme, suggesting a possible induced-fit mechanism by which transport substrate binding may increase the affinity of ATP at the active site of the cassettes. In addition, these data have allowed us to formulate hypotheses about how the energy derived from ATP hydrolysis may be transduced to the transmembrane domains during substrate transport.