Lipid transfer mechanism: Crystal structures of human VAPA protein and its complex with CERT FFAT motif

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Ceramide is one of the lipids in mammalian cells, and plays important roles in cell growth, differentiation and apoptosis. It is also a precursor for sphingomyelin, a major component of lipid membrane. Ceramide is synthesized in the the endoplasmic reticulum (ER) and transported to the Golgi apparatus for conversion to sphingomyelin. The ceramide transport occurs in a nonvascular manner and is mediated by CERT, a cytosolic ceramide transfer protein [1].

CERT is a 68 kDa protein, and the C-terminal START domain recognizes and transfers ceramide specifically [2, 3]. CERT has an FFAT motif (two phenylalanies (FF) in an acidic tract), in the middle region of the protein. FFAT motif is known for targeting motif of the ER, and its consensus sequence is EFFDAxE. VAMP associated protein A (VAPA) is an integral membrane protein on the ER membranes. VAPA has a cytosolic major sperm protein (MSP) domain at its N-terminus. It has been shown that the MSP domain of VAPA interacts with the FFAT motif of CERT [4]. These results suggest that the interaction between the VAPA MSP domain and the ER-targeting FFAT motif of CERT is important for the ceramide transfer from the ER to the Golgi.

To clarify the molecular mechanisms of the ceramide transfer by VAPA and CERT, we have determined the crystal structures of the VAPA MSP domain, and its complex with the FFAT motif of CERT, at 1.7 and 2.3 Å resolutions, respectively (PF-AR NE3A, and PF-AR NW12A). The crystal structure of the VAPA MSP domain has an immunoglobulin β fold, and the VAPA MSP domain binds to the FFAT motif in the central hydrophobic pocket of the protein. Based on these structures, the mechanisms of ceramide transport and ER targeting will be discussed.

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[4] Kawano, *et al.*, (2006) *J. Biol. Chem.* **281**, 30279-30288.