

A V-type c subunit in an archaeal ATP synthase: Probing the Na⁺ binding site in the rotor

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A₁A₀ ATP synthases are the energy producers in the third domain of life, the archaea. These enzymes are unique and share properties with both, the bacterial F₁F₀ ATP synthases and the eukaryal V₁V₀ ATPases. Most of the archaea have an F-type like proteolipid (c subunit) in their rotor rings, consisting of one hairpin with one ion binding site. In contrast to that some other archaea, e. g. *Pyrococcus furiosus*, have an unusual proteolipid (1). The c subunit from this hyperthermophilic archaeon was isolated by chloroform/methanol extraction, purified and the molecular mass was determined with MALDI-TOF-MS. The proteolipid of *P. furiosus* is a 16 kDa protein, arisen by gene duplication, with two hairpins and two putative ion binding sites. Labeling of the ion binding sites with the ATP synthase inhibitor N,N - Dicyclohexylcarbodiimide (DCCD) clearly showed, that only one ion binding site is functional and that the labeling process is pH-dependent. The effect of Na⁺ on the labeling is object of the current research. Based on these results and the recent finding, that the A₁A₀ ATP synthase of *P. furiosus* has a rotor with 10 c subunits (2), a homology model of the c ring is present, giving first insights into the structure of an archaeal rotor ring with its ion binding site.

(1) Müller V (2004) An Exceptional Variability in the Motor of Archaeal A₁A₀ ATPases: From Multimeric to Monomeric Rotors Comprising 6-13 Ion Binding Sites. J Bioenerg Biomembr 36: 115-125

(2) Vonck J, Pisa KY, Morgner N, Brutschy B and Müller V (2009) Three-dimensional structure of A₁A₀ ATP synthase from the hyperthermophilic archaeon *Pyrococcus furiosus* by electron microscopy. J Biol Chem 284: 10110-10119