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Structural and functional analysis of a putative novel nucleotide-metabolizing enzyme TTHA1091

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Nucleotides are component of nucleic acids, essential molecules that carry genetic information. Nucleotides also play important roles in energy metabolism and signal transduction. Therefore, nucleotide metabolic pathways are very important in life processes. Two pathways of nucleotide biosynthesis are known: the *de novo* synthesis from precursor molecules and the salvage pathway from degraded products of nucleic acids. The *de novo* synthesis system has been studied well, but the salvage pathway has been far less studied. Recently, Lott *et al.* determined the crystal structure of a putative novel adenosine kinase PAE2370 from a hyperthermophilic archeon *Pyrobaculum aerophilum* [1]. Homologues of this protein are widely conserved in bacteria and archea, suggesting its functional importance. *Thermus thermophilus* HB8, an extremely thermophilic bacterium, has the structurally homologous protein TTHA1091. Proteins from this eubacterum are extremely stable and suitable for *in vitro* characterization including X-ray crystallograhy. We have already solved X-ray crystal structure of TTHA1091 in the 1.75 Å resolution (PDB ID: 1VGG). In this research, we first analyzed the crystal structure in more detail, and then assessed the adenosine kinase activity of the purified TTHA1091.

TTHA1091 is composed of 161 amino acids and forms a hexameric ring of two trimers (Fig. 1). We identified functionally important regions by mapping the level of evolutionary conservation of each position onto the 3D structure of TTHA1091. Highly conserved amino acid residues are located in the interface between two subunits in the ring and form a pocket-like structure (Fig. 2). These suggested that this pocket-like structure is important for functional role of TTHA1091. In fact, in the determined structure of PAE2307, a histidine residue in this pocket was phosphorylated. Then, we examined whether TTHA1091 had adenosine phosphorylation activity, but have not detected the activity yet. There may be a possibility that other nucleoside or nucleotide are substrate for this protein. Further analyses are now in progress.

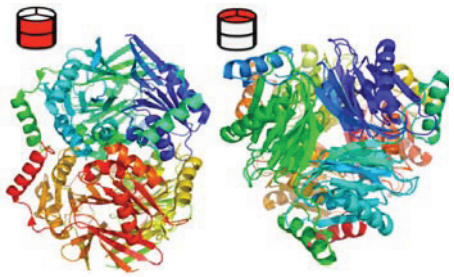

Figure 1. Structure of TTHA1091

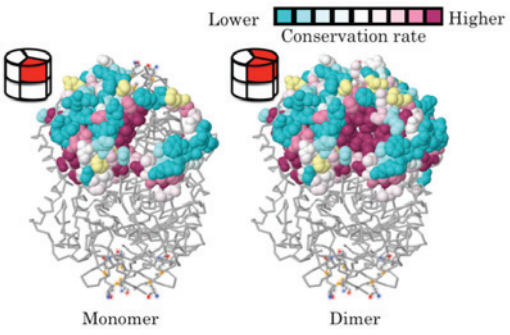

Figure 2. TTHA1091 by mapping the level of evolutionary conservation

Reference

[1] J. S. Lott *et al.* (2006) *J. Biol. Chem.* **281**, 22131-22141