MS07- Nucleic acids and interactions with proteins

Chairs: Prof. Ralf Ficner, Prof. Markus Wahl

MS07-P01

Purification and crystallization of novel archaean protein of the LSM family

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Lsm (Sm-like) proteins are found in representatives of all the three domains of life. They provide biogenesis and functioning of RNA molecules in the cells. Bacterial Lsm proteins called Hfq exhibit RNA-chaperone activity promoting interaction between regulatory sRNA and mRNA during regulation of translation [1], [2]. Eukaryotic Sm proteins are core proteins of the spliceosome while eukarvotic Lsm proteins are involved in the mRNA degradation [3]. Functions of the archaeal Lsm proteins (SmAP) in the cell have been studied pitiable, although there is some data on their participation in the processing of some. Our work concerns with structural and functional studies of an archaeal Lsm protein from Halobacterium salinarum. This protein has remarkable differences of the sequences compared with the homologues and, in fact, represents a minimal Lsm core. My current task is to determine the structure of the protein and its complexes with ribonucleotides and short RNA to define specificity and structural aspects of the Lsm-RNA interaction. We have obtained a genetic construct carrying the gene of the SmAP from H. salinarum (HsaSmAP). The protein was isolated and purified in preparative scale; it has been crystallized and a high-resolution diffraction dataset has been collected at ERSF in Grenoble. The work is supported by RFBR grant #18-04-00222.

References:

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MS07-P02

Comparison of the uridine-binding site of hexameric and heptameric archaeal Lsm proteins

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The Sm and Sm-like proteins are widely distributed among bacteria, archaea and eukarya. They are defined by the ability to adopt the Sm fold, which is comprised of a 5-stranded β -sheet and an N-terminal α -helix. They are participated in many processes connected with RNA-processing or regulation of gene expression. Hetero-heptameric eukaryotic Sm proteins form the core of the uracil-rich small nuclear RNPs that further assemble into spliceosomes and excise introns in eukaryotic pre-mRNAs. Homo-hexameric bacterial Lsm protein Hfq binds polyU RNA sequences in pockets between adjacent monomers at the central pore providing regulation translation of many mRNA. Archaeal Lsm proteins (SmAP, Sm Archaeal Protein) form homo-hexamers and homo-heptamers and appear to bind urudine-rich RNAs, nevertheless, the function of SmAP in the archaeal cells is not clear.

We compare structural organization of the uridine-binding site of SmAP from *Methanococcus jannaschii*, which forms hexamers, and SmAP from *Methanococcus vannielii*, which forms heptamers. The proteins were isolated and purified. Crystals of proteins and their complexes with ribonucleotides were obtained. Using the approach, which has been developed in our group, we analyze single-stranded RNA-binding sites on the surface of the proteins. Comparison of the obtained structures with the known SmAP structures in complex with RNA-fragments reveal significant differences in the RNA-binding site of the proteins.

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Keywords: Lsm protein family, archaea