

# ERA GTPase of *Staphylococcus aureus*: cloning, expression, purification and preparation for structural research

P-02.1-20

E. Klochkova<sup>I</sup>, S. Validov<sup>I</sup>, A. Bikmullin<sup>I</sup>, D. Islamov<sup>I</sup>, M. Yusupov<sup>I,II</sup>, **K. Usachev<sup>I</sup>**

<sup>I</sup>Kazan Federal University, Kazan, Russia, <sup>II</sup>IGBMC, Strasbourg, France

ERA, is a widely conserved and essential GTPase in bacteria playing a significant role in the regulation of protein synthesis. In bacteria, ERA is required for the maturation of the 30S ribosomal subunit and coordination of cell growth and division cycle. However, the contribution of ERA to protein biosynthesis is not yet fully understood. The study of the effect of ERA on the bacterial protein-synthesizing apparatus will help in finding new targets in the fight against a number of human bacterial pathogens such as *Staphylococcus aureus*. In this study, we have cloned the *era* gene from the genome of *S. aureus* and obtained the construct expressing ERA with six histidine residues on C-terminus. His<sub>6</sub>-ERA was expressed in *E. coli*, purified, and concentrated to 10 mg/ml for further search of crystallization conditions. Crystals of ERA in a complex with GppCp were obtained using the hanging-drop techniques. We also showed that ERA forms a stable complex with a 30S ribosome subunit of *S. aureus*, confirmed using agarose gel electrophoresis and PAGE. Further studies of ERA molecule crystal structure by X-ray diffraction and cryo-EM studies of ERA-30S complexes will prompt active sites responsible for interaction with ribosomal subunit.

This work was supported by the Russian Science Foundation (grant 21-74-20034).