. Structural and Functional Study of the Major Cold Shock Protein of E. Coli, CspA, as a Model of RNA Chaperone

M.R. Quintero Bernabeu , R. Konrat, K. Kloiber

.Max F. Perutz Laboratories, Vienna, Austria

CspA is the major cold shock protein from Escherichia coli, which is transiently overproduced upon cold-stress conditions. It induces adaptation of the cell to temperatures as low as 10-15 degrees and displays RNA chaperone activity. It has been reported to participate in a negative feedback mechanism regulating its own gene expression by interacting (directly or indirectly) with its 5' leader region. The latter contains an 11-base sequence (cold box) that is conserved among cold shock genes. Albeit there is evidence strongly corroborating the ss-nucleic acid binding capacity of CspA (on a molecular level), the detailed events which lead to its chaperone activity have yet to be elucidated.

.Here we use NMR spectroscopy to investigate the interaction of CspA (on an atomic level) with a model 9 nt synthetic RNA (5'-UUU-AUU-AUT-3'). We found that this short RNA binds strongly to CspA and induces little exchange broadening in the ligated state compared to longer RNAs. The RNA binding site of CspA to S6 has been located using an automated binding site mapping procedure developed in our lab. This approach, which uses triple resonance data of the apo-state together with 3D structural information will be introduced here. Furthermore we will show that the binding interface with respect to this short nucleic acid is coincident with the aromatic face of the protein previously reported to be involved in contact with a 24 nt ss-DNA that contains the coldbox motif. With this we corroborate the assumption that our structural and dynamics studies of the interaction between CspA and S6 are of biological relevance.

▪

This work has been carried out with financial aid of FWF (Austrian Science Foundation, SFB-17 project)