Crystal Structure of the Cold-Shock Protein CspB from *Bacillus Caldolyticus* and its R3E Mutant

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The cold shock proteins (CspB) from *B. subtilis* and from the thermophilic *B. caldolyticus* differ remarkably in terms of thermostability. Since the sequences of both proteins vary at only 12 positions, it seems worthwhile to study variations of the sequence in conjunction with biophysical and structural investigations in order to gain information about the structural basis of thermostability. The crystal structure of CspB from *B. subtilis* is known [1]. The structure determination of various mutants differing in thermal stability in conjunction with biophysical experiments should lead to a detailed understanding of the energetics of structural transition in proteins.
The diffraction experiments at BW7A resulted in a 1.17 Å dataset for *B. caldolyticus* CspB and a 1.4 Å dataset for the R3E mutant, which are both crystallizing in space group $I4_1$. The structures were solved by molecular replacement with AMORE [2]. Refinement of both structures was carried out with SHELX-97 [3] using anisotropic displacement factors. The final R/R_{free} equals 12.06/18.02 % for the wildtype and 14.08/20.5 % für the R3E mutant structure. Both proteins comprising 66 amino acids each are present as a dimer in the asymmetric unit of the $I4_1$ cell (Fig. 1). Similar to the CspB from *B. subtilis,* each monomer is arranged as a five-stranded β-barrel, which strands β1-β3 are approximately perpendicular to β4-β5. A loop of 14 amino acids residues between β3 and β4 connects the two half-sheets. The interpretation of these structures in terms of the origin of their different thermal stability is in progress.

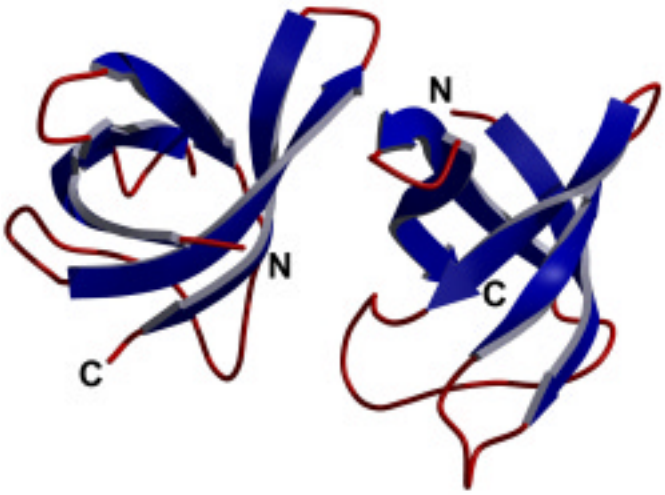

Figure 1: Dimeric structure of CspB from *Bacillus caldolyticus*

References

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