Comparative modelling can be envisaged as two steps. The first is to solve the inverse folding problem: to define all those sequences that can adopt a particular fold. Operationally this is more usefully posed as defining whether a new sequence belongs to any of the known folds. It involves projecting constraints from a three-dimensional fold onto a one-dimensional sequence. For this step we have calculated amino acid substitution tables in terms of local structural environmental parameters, which can be used to generate sequence templates for secondary structures, structural motifs and tertiary folds. The second step is to use the sequence, together with the knowledge that the protein belongs to a family of known fold, to construct a model. This form of protein modelling and prediction involves placing constraints from a known fold on a related protein sequence. The two steps require similar knowledge of the structures of protein families, and this knowledge can be expressed as rules that relate both local and global three-dimensional patterns in the sequence of amino acids in a polypeptide chain. The method is comparative but exploits a broader knowledge-base of non-homologous protein structures.


Various mechanisms of the protein stability have so far been studied, and several strategies to enhance the stability are now proposed. We have recently made lots of mutant proteins of ribonuclease H from Escherichia coli (E.coli RNase H, 17.6kDa), and studied the conformational stability and their crystal structures. As a result, several of the mutant proteins obtained the remarkable thermal stability, due to very local amino acid replacements. For example, Lys95–Gly is considered to stabilize the local left-handed α-helical conformation by a Gly residue (S. Kimura et al., J. Biol. Chem., 1992, 267, 22014-22017). His82–Pro may stabilize the short turn structure (K. Ishikawa et al., Protein Eng., 1993, 6, 85-91). Val74–Leu, fills the cavity in the hydrophobic core (K. Ishikawa et al., Biotechnology, 1993, in press). All three mechanisms are localized, and the characteristic features of individual amino acids contribute to the increase of the thermal stability. Analyses of the crystal structures of the wild-type and mutant proteins of E.coli RNase H less than 1.8 Å resolution reveal that the global conformational change of all those mutant proteins deviate very little from that of the wild-type protein. It means that these local structural changes can be permitted and even suitable for the original global conformation. The additivity of the mutations was confirmed (S. Kimura et al., J. Biol. Chem., 1992, 257, 21535-21542), and the structural analyses of the associated protein from Thermus thermophilus show that these local mechanisms are used in the thermophilic protein (K. Ishikawa et al., J. Mol. Biol., 1993, 230, 529-542).

MS-05.01.05 RULE-BASED APPROACHES TO COMPARATIVE MODELING by Y.Z. Zhu, M.J. Johnson, H. Wako, R. Sewaldin, N. Srinivasan, K. Guruprasad, L. Sun, B. Roberts, S. Ruffino, Y. Edwards, T. Muckle*. Imperial Cancer Research Fund Unit of Structural Molecular Biology, Department of Crystallography, Birkbeck College, University of London, Malet Street, London WCIE 7HE, UK.