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Structural conformation of *Bacillus stearothermophilus* F1 protease and effect of modification on its thermostability

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A thermophilic *Bacillus stearothermophilus* F1 that produced an extremely thermostable serine protease was isolated, cloned, sequenced and expressed in *Escherichia coli*. Its amino acid sequence was used to predict the three dimensional structure, to provide better insights into relationship between protein structure and biological function as well as open opportunities for protein engineering. The amino acid sequence of F1 protease was modeled onto the crystal structure of thermolysin that had 61% sequence identity. In order to investigate further the determinant of F1 protease stability, one mutant (W200R) of F1 protease was designed. From computational work, the mutated F1 protease showed additional three new ion pairs at that point. Site directed mutagenesis was then carried out and the mutated F1 protease expressed. The half-life of the W200R mutant protease at 85°C was 10 minutes longer than the wild type enzyme. The increased in thermostability obtained shows the importance of ion pairs in determining protein stability.