Obeying Anfinsen: a Serpin that folds to the most Stable State

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Members of the serpin superfamily of protease inhibitors represent an exception to Anfinsen's conjecture and fold to a native metastable conformation, rather than the theoretically most stable "relaxed" conformation. Inhibitory serpins utilise metastability to inhibit target proteases. Unfortunately, as a consequence of metastability, serpins are conformationally labile, and vulnerable to mutations that promote the formation of inactive loop sheet polymers. Polymerisation of human serpins is the critical factor in the development of a number of degenerative diseases (serpinopathies).

Eukaryote serpins are sensitive to mild heating, however, to our surprise, we have identified serpins in thermophilic prokaryotes. A structural study on the serpin thermopin reveals that this molecule is able to adopt the native and cleaved state and inhibits a metastable α-lytic-like protease. In contrast, the high-resolution crystal structures of another prokaryote serpin, tengpin, reveals that the serpin domain of this molecule folds spontaneously and rapidly to most stable (i.e. relaxed) conformation. This is an exciting result, since tengpin represents the first serpin identified to date that obeys Anfinsen's conjecture. Furthermore, the X-ray crystal structures of tengpin reveals the structural basis for a novel mechanism for loop-C-sheet serpin-polymerisation. Analysis of the structural data provides striking insight into the mechanism of serpin metastability and the structural basis for serpin polymerisation.

[1] a) Irving J.A., et al., *Structure 2003*; b) Fulton K.F., et al., *J Biol Chem,* 2005.

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