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Human α_1-Antitrypsin Variant with Enhanced Conformational Stability at the Cost of Activity

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Native strain of inhibitory SERPINS (Serine protease inhibitors) is thought to be used in the facile conformational switch to play biological regulation.
Many heat stable variants of α_1-antitrypsin, a prototype of inhibitory serpins, increased their stability by reducing the native strain.
We combined sixteen thermostable mutations of α_1-antitrypsin(M16) of which mutation sites were distributed over the whole molecule. M16 was more stable than a noninhibitory serpin, ovalbumin, and still maintained native conformation in 8M urea.
Interestingly, M16 lost most, if not all, of its inhibitory activity. Most M16 molecules partitioned into substrate pathway in a elastase binding assay. M16 did not make a binary complex with a 14-mer peptide mimicking the sequence of reactive site loop (RSL) during 24 hr incubation at 50℃, suggesting the rate of RSL insertion is extreamely retarded.
It seems that M16 can not overcome the energy barrier of the transition from native state to enzyme-inhibitor complex.