Supporting Material

Specific and Selective Inhibitors of Proprotein Convertases Engineered by Transferring Serpin B8 Reactive-Site and Exosite Determinants of Reactivity to the Serpin α1PDX

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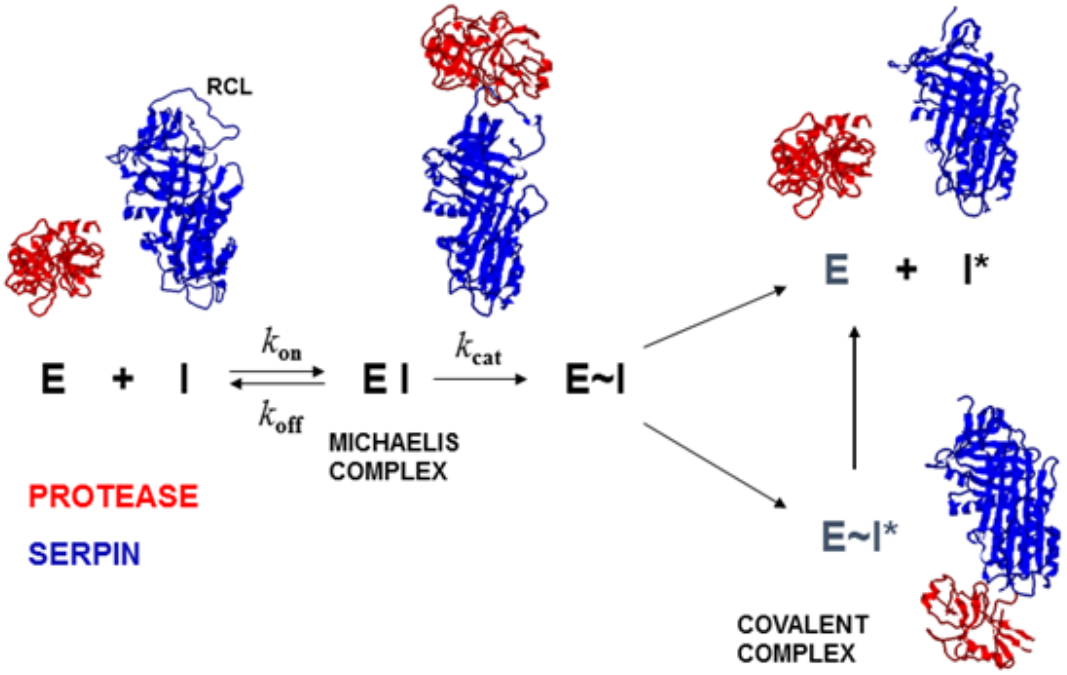

Figure S1. Mechanism of protease inhibition by serpins. The catalytic site of the protease (E) binds to the serpin (I) reactive center loop (RCL) to form an equilibrium binding Michaelis complex (EI). The Michaelis complex then undergoes the chemical reaction step (k_{cat}), in which a covalent-acyl bond (E~I) is formed. The covalent complex can be rapidly resolved either by the hydrolysis of the bond, which leads to the formation of cleaved serpin (I*) and the regeneration of the free protease (E), or by a conformational transition step that traps the protease in a stable inhibitory covalent complex with the inhibitor (E~I*). The inhibitory complex slowly dissociates into cleaved serpin and free enzyme. A 1:1 stoichiometry of inhibition is indicative of an efficient mechanism of inhibition.

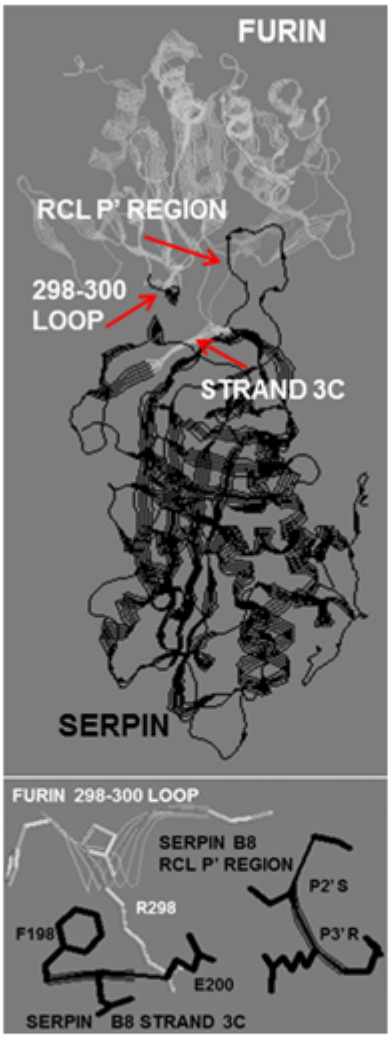

Figure S2. Model of the Michaelis complex structure formed by furin and serpin B8. The relative positions of the serpin RCL and strand 3 of β-sheet C, and the protease 298-300 loop are shown. The serpin exosite residues are located on strand 3C. The modulation of the interaction between the serpin RCL P' region and the PC 298-300 loop (Arg298 forms part of the S2' pocket) by the serpin strand 3C exosite residues (F198 and E200 in serpin B8; Y222 and E224 in α1PDX) was described in reference 10. Here, the RCL primed regions of serpin B8 and α1PDX are reported to determine different selective exosite interactions between furin and PC4-PC7.