

P019 The initial substrate binding site of γ -secretase is located on presenilin near the active site

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Alzheimer's γ -secretase cleaves the transmembrane domain of the APP and this enzyme is a founding member of aspartyl intramembrane proteases. This protease is a complex of four integral membrane proteins, with presenilin as the catalytic component. The substrate transmembrane domain appears to adopt a helical conformation upon initial interaction with the protease. Additional evidence supports the presence of an initial docking site for substrate on γ -secretase that is distinct from the active site. We showed that peptides designed to adopt a helical conformation inhibit γ -secretase. Importantly, these inhibitors do not compete with an active-site directed photoaffinity reagent, which suggests that helical peptides do not bind to the active site of γ -secretase. Here we report that photoprobes based on helical peptide inhibitors bind specifically to the PS subunit interface, at a site distinct but close to the active site. The location of the helical peptide binding site suggests that substrate passes between the two PS subunits to access the active site. An aggressive Alzheimer-causing mutation in PS1 strongly reduced photolabeling by a transition-state analogue but not by helical peptides, providing biochemical evidence that the pathological effect of this PS mutation is due to alteration of the active site topography.