

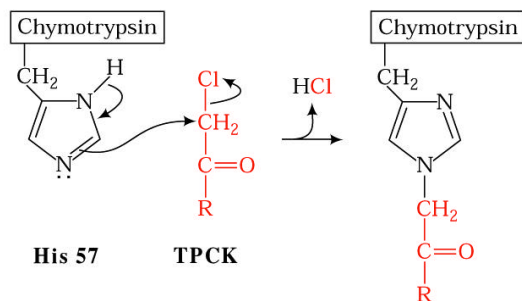
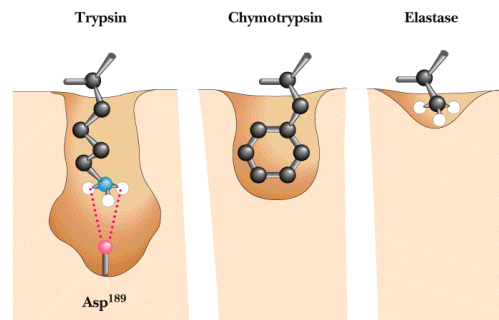
## Enzymatic Catalysis V Proteases - Chymotrypsin

4 classes of proteases

- 1 - zinc protease
  - 2 - serine protease
  - 3 - thiol proteases
  - 4 - aspartyl proteases
- All forms of proteolytic enzymes fit into these 4 classes
  - Aids involves aspartate in active site to complete formation of viral replication proteins
    - protease inhibitors are now used to prevent exposed patients

### Chymotrypsin

- Model of a covalent catalysis mechanism
  - Part of the broad class of serine proteases
  - Serine not generally an active amino acid for acid/base catalysis
  - Catalytic triad of serine, histidine and aspartate responsible for the reactivity of serine in this active site
  - Two phase reaction when an ester is used
    - burst phase - E + S initial reactions
    - steady state phase - EP → E + P (deacylation)
- The first step is the covalent catalysis - where the substrate



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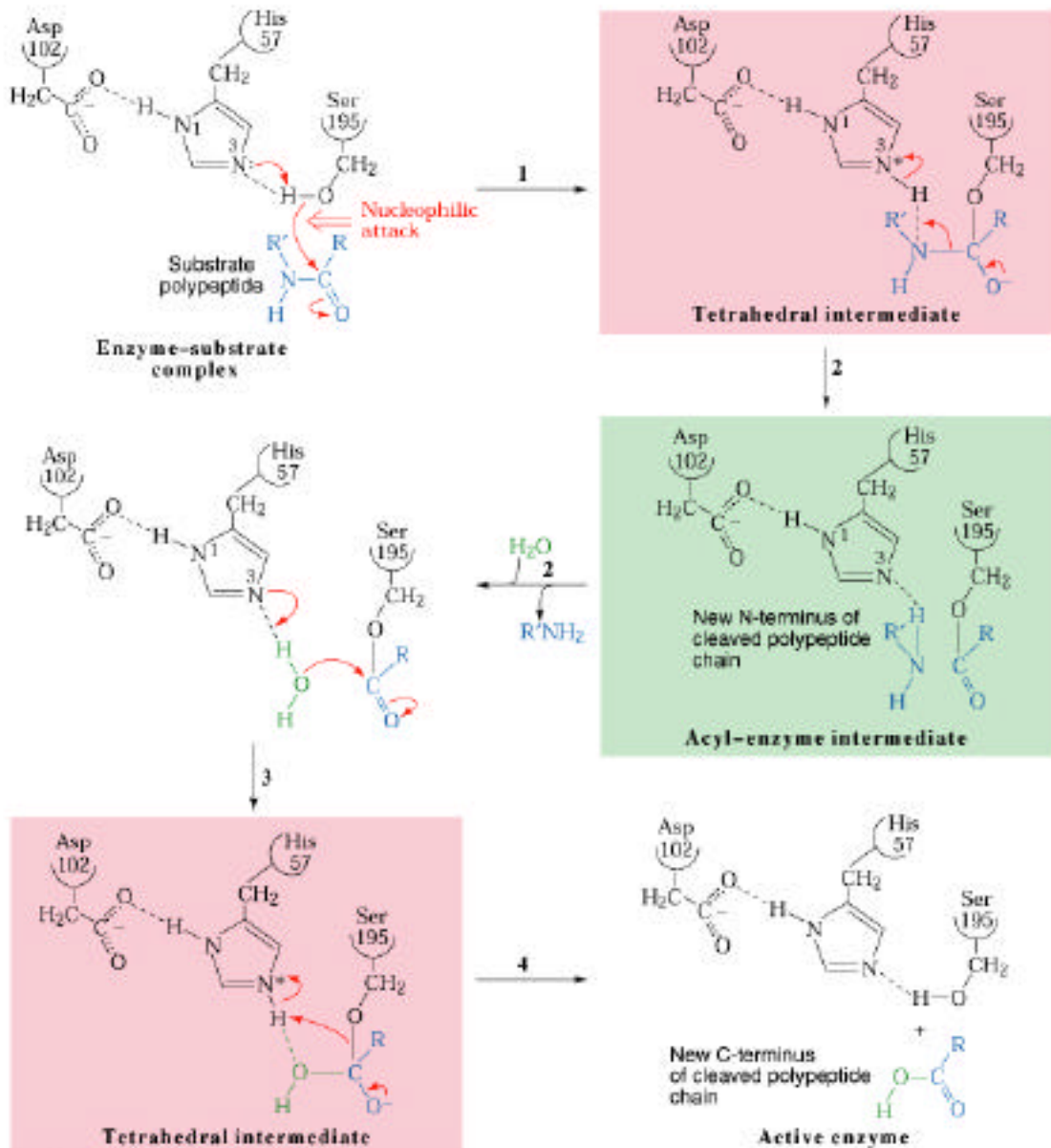
actually is bound to the enzyme itself

- Use of inhibitors show a serine is responsible for first step of reactive Ser
- Another inhibitor TPCK binds to His by affinity labeling
- pH vs. activity also indicate His is important
- Interactions between Asp, His

and Ser make the Ser reactive.

- Ser OH is ionized when H<sup>+</sup> is donated to His.
- Asp "holds" the His in place
- Removing the proton from Ser makes it a better nucleophile

- Attack (of the killer tomatoes) Ser on C=O of peptide and formation of the acyl intermediate
- transition tetrahedral - oxyanion stabilized by NH from peptide chains
- Second stage the reverse takes place where water is activated by the histidine as a base



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