

# Plasminogen: The Clot Buster

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## Abstract

Coagulation and fibrinolysis comprise a complex system involving many proteins designed to form fibrin clots when needed to repair a blood vessel and dissolve these clots when no longer needed. One key fibrolytic protein, plasmin, circulates in the blood as plasminogen, an inactive protein made of several domains. Five domains are kringle domains that help plasmin bind to lysine residues in fibrin, while the catalytic domain exhibits protease activity that fragments the fibrin network. Plasminogen is incorporated within the fibrin network as a clot forms, but the catalytic domain is only able to break down fibrin when plasminogen has been activated and converted to plasmin. One substance known to convert plasminogen into plasmin, tissue plasminogen activator (tPA), is a serine protease that cleaves the peptide bond between Arg561 and Val562 in plasminogen. The endothelial cells of damaged blood vessels slowly release tPA that activates the plasminogen embedded within a fibrin clot. Because increased plasmin levels help dissolve clots, tPA is given clinically to treat conditions caused by blood clots, like heart attack and stroke. To further understand the structural implications of activation, a 3D physical model of plasminogen has been designed and built by the Cedarburg High School SMART (Students Modeling a Research Topic) Team using 3D printing technology.

## Plasminogen has an important role in both normal and abnormal blood clotting and dissolving pathways.

- A fibrin clot, or thrombus, is the final step of the coagulation process that seals off a damaged blood vessel, but unwanted fibrin clots can also form inside undamaged vessels, resulting in thrombosis and obstructed blood flow.
- Plasmin functions in the fibrolytic mechanism to dissolve blood clots, whether formed normally in cases of injury or abnormally in cases of thrombosis.
- Plasmin levels must be carefully regulated; increased levels of plasmin may result in excessive bleeding and decreased levels of plasmin may result in thrombosis.
- A decreased plasminogen level may interfere with the body's ability to dissolve clots and thus also result in thrombosis.
- Because a leading cause of heart attack or stroke is thrombosis, clinical treatments focus on increasing plasmin activation.

## Steps 1-4 illustrate coagulation and fibrinolysis.

1.

### Fibrin forms clots.

- Blood clots develop through a cascade of events that activate fibrin.
- A fibrin network forms that traps cells and debris, preventing blood loss.

2.

### Plasminogen binds clots.

- Plasminogen binds to fibrin when it is not catalytically active.
- The fibrin-binding domains of plasminogen, kringle domains, recognize and bind lysine residues of fibrin.

4.

### Plasmin cleaves fibrin.

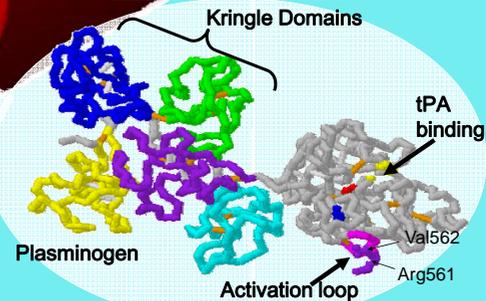
- Plasmin is a serine protease that hydrolyzes the peptide bonds located on the carboxyl side of lysines and arginines in fibrin.
- Cleaving bonds in fibrin leads to the dissolution of the clot.

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## Further Research

- Plasminogen also interacts with cation-independent mannose-6-phosphate receptor (CI-MPR).
- Since CI-MPR targets cellular enzymes to lysosomes and also interacts with several molecules at the cell surface, research is underway to identify residues necessary for binding CI-MPR.

**A zymogram showing plasminogen treated with tPA is catalytically active.** Plasmin (Lane 1) is run as a control. Plasminogen treated with tPA is in Lane 2. The band in each lane indicates protease activity.



kringles 1-3 (1K10.pdb), kringle 4 (2PK4.pdb), kringle5 (5HPG.pdb) and the catalytic domain (1DDJ.pdb)

Each kringle domain is colored a different color. Specific residues are highlighted on the catalytic domain; the catalytic residues are red and blue. Disulfide bonds are orange and hydrogen bonds are white.

3.

### Plasminogen must be activated to dissolve fibrin.

- Tissue plasminogen activator (tPA) is released by endothelial cells in damaged blood vessels.
- tPA is a serine protease and catalyzes the conversion of plasminogen into plasmin by cleaving the bond between arginine 561 and valine 562 and removing the N-terminal portion.
- The two chains are linked together by a disulfide bonds, forming the active plasmin.
- The structure of activated plasmin (Figure B) is more open compared to plasminogen (Figure A).

Figure A: Inactive Plasminogen

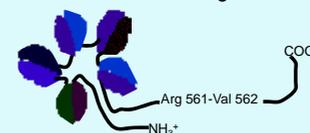


Figure B: Active Plasmin

