

Formation of Fibrin Clot (Clotting Cascade)



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This is just an excerpt of a full-length report for this pathway. To access the complete report, please download it at the <u>Reactome Textbook</u>.

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Introduction

Reactome is open-source, open access, manually curated and peer-reviewed pathway database. Pathway annotations are authored by expert biologists, in collaboration with Reactome editorial staff and cross-referenced to many bioinformatics databases. A system of evidence tracking ensures that all assertions are backed up by the primary literature. Reactome is used by clinicians, geneticists, genomics researchers, and molecular biologists to interpret the results of high-throughput experimental studies, by bioinformaticians seeking to develop novel algorithms for mining knowledge from genomic studies, and by systems biologists building predictive models of normal and disease variant pathways.

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This document contains 4 pathways (see Table of Contents)

Formation of Fibrin Clot (Clotting Cascade) 7

Stable identifier: R-HSA-140877

Compartments: extracellular region



The formation of a fibrin clot at the site of an injury to the wall of a normal blood vessel is an essential part of the process to stop blood loss after vascular injury. The reactions that lead to fibrin clot formation are commonly described as a cascade, in which the product of each step is an enzyme or cofactor needed for following reactions to proceed efficiently. The entire clotting cascade can be divided into three portions, the extrinsic pathway, the intrinsic pathway, and the common pathway. The extrinsic pathway begins with the release of tissue factor at the site of vascular injury and leads to the activation of factor X. The intrinsic pathway provides an alternative mechanism for activation of factor X to the formation of a multimeric, cross-linked fibrin clot. Each of these pathways includes not only a cascade of events that generate the catalytic activities needed for clot formation, but also numerous positive and negative regulatory events.

Literature references

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Kisiel, W., Fujikawa, K., Davie, EW. (1991). The coagulation cascade: initiation, maintenance, and regulation. *Biochemistry*, 30, 10363-70. ↗

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Extrinsic Pathway of Fibrin Clot Formation 7

Location: Formation of Fibrin Clot (Clotting Cascade)

Stable identifier: R-HSA-140834

Compartments: extracellular region



Factor VII, the protease that initiates the normal blood clotting cascade, circulates in the blood in both its proenzyme (factor VII) and its activated (factor VIIa) forms. No clotting occurs, however, because neither form of the protein has any catalytic activity when free in solution. Blood clotting is normally initiated when tissue factor (TF), an intrinsic plasma membrane protein, is exposed to the blood by injury to the wall of a blood vessel. TF is then able to bind factor VIIa from plasma, and possibly also factor VII, to form complexes capable of catalyzing the conversion of factor X, from plasma, into its activated form, factor Xa. Factor Xa catalyzes the conversion of additional factor VII molecules to their activated form, increasing the amount of tissue factor: factor VIIa complex available at the site of injury, accelerating the generation of factor Xa, and allowing the activation of factor IXa as well. This process is self-limiting because as levels of factor Xa increase, tissue factor: factor VIIa complexes become trapped in the form of catalytically inactive heterotetramers with factor Xa and the protein TFPI (tissue pathway factor inhibitor). At this point the intinsic pathway, as an independent source of activated factor X, is thought to become critical for the continuation of clot formation (Broze 1995; Mann et al. 2003).

The nature of the initial tissue factor: factor VII complexes formed is controversial. One model, building on the observation that the complex of factor VII and TF has low but measurable proteolytic activity on factor X, suggests that this complex begins the activation of factor X, and that as factor VIIa accumulates, tissue factor: factor VIIa complexes also form, accelerating the process (Nemerson 1988). A second model, building on the observation that normal plasma contains low levels of activated factor VII constitutively, suggests that complexes with factor VIIa form immediately at the onset of clotting (Rapaport and Rao 1995). The two models are not mutually exclusive, and in any event, the central roles of tissue factor and factor VIIa in generating an initial supply of factors IXa and Xa, and the self-limiting nature of the process due to the action of TFPI, are all well-established.

Literature references

Broze, GJ Jr. (1995). Tissue factor pathway inhibitor and the revised theory of coagulation. *Annu Rev Med*, 46, 103-12.

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Intrinsic Pathway of Fibrin Clot Formation 7

Location: Formation of Fibrin Clot (Clotting Cascade)

Stable identifier: R-HSA-140837

Compartments: extracellular region



The intrinsic pathway of blood clotting connects interactions among kininogen (high molecular weight kininogen, HK), prekallikrein (PK), and factor XII to the activation of clotting factor X by a series of reactions that is independent of the extrinsic pathway and that is not subject to inhibition by TFPI. It is thus essential for the prolongation of the clotting cascade: while the reactions of the extrinsic pathway appear to be sufficient to initiate clot formation, those of the intrinsic pathway are required to maintain it (Broze 1995; Davie et al. 1991; Monroe et al. 2002). The intrinsic pathway can be divided into three parts: 1) reactions involving interactions of kininogen, prekallikrein, and factor XII, leading to the activation of factor XII, 2) reactions involving factor XI, factor IX, factor VIII, and von Willebrand factor (vWF) leading to the activation of factors VIII and IX, and 3) reactions that inactivate factor XIIa and kallikrein.

Kininogen, prekallikrein, and factor XII were first identified as proteins needed for the rapid formation of clots when whole blood is exposed to negatively charged surfaces in vitro. Early studies in vitro identified several possible sets of interactions, in which small quantities of one or more of these proteins 'autoactivate' and then catalyze the formation of larger quantities of activated factors. Subsequent work, however, suggests that these factors form complexes on endothelial cell surfaces mediated by C1q binding protein (C1q bp), that the first activation event is the cleavage of prekallikrein by prolylcarboxypeptidase, and that the resulting kallikrein catalyzes the activation of factor XII (Schmaier 2004).

The second group of events, occurs in vivo on the surfaces of activated platelets (although most biochemical characterization of the reactions was originally done with purified proteins in solution). Factor XI binds to the platelet glycoprotein (GP) Ib:IX:V complex, where it can be activated by cleavage either by thrombin (generated by reactions of the common pathway) or by activated factor XII (generated in the first part of the intrinsic pathway). Activated factor XI in turn catalyzes the activation of factor IX. Simultaneously, factor VIII, complexed with vWF, is cleaved by thrombin, activating it and causing its release from vWF. Activated factors VIII and IX form a complex on the platelet surface that very efficiently converts factor X to activated factor X. (Activated factors X and V then form a complex that efficiently activates thrombin.)

While these two groups of events can be viewed as forming a single functional pathway (e.g., Davie et al. 1991), human clinical genetic data cast doubt on this view. Individuals deficient in kininogen, prekallikrein, or factor XII proteins exhibit normal blood clot formation in vivo. In contrast, deficiencies of factor XI can be associated with failure of blood clotting under some conditions, and deficiencies of vWF, factor VIII, or factor IX cause severe abnormalities - von Willebrand disease, hemophilia A, and hemophilia B, respectively. These data suggest that while the second group of events is essential for normal clot formation in vivo, the first group has a different function (e.g., Schmaier 2004).

Finally, reactions neutralize proteins activated in the first part of the intrinsic pathway. Kallikrein forms stable complexes with either C1 inhibitor (C11nh) or with alpha2-macroglobulin, and factor XIIa forms stable complexes with C11nh. The relevance of these neutralization events to the regulation of blood clotting is unclear, however. The physiological abnormalities observed in individuals who lack C11nh appear to be due entirely to abnormalities of complement activation; blood clotting appears to proceed normally. This observation is consistent with the hypothesis, above, that factor XIIa plays a limited role in normal blood clotting under physiological conditions.

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Common Pathway of Fibrin Clot Formation 7

Location: Formation of Fibrin Clot (Clotting Cascade)

Stable identifier: R-HSA-140875

Compartments: extracellular region



The common pathway consists of the cascade of activation events leading from the formation of activated factor X to the formation of active thrombin, the cleavage of fibrinogen by thrombin, and the formation of cleaved fibrin into a stable multimeric, cross-linked complex. Thrombin also efficiently catalyzes the activation of several factors required earlier in the clotting cascade, thus acting in effect as a positive regulator of clotting. At the same time, thrombin activates protein C, which in turn catalyzes the inactivation of several of these upstream factors, thereby limiting the clotting process. Thrombin can be trapped in stable, inactive complexes with: antithrombin-III (SERPINC1), a circulating blood protein; heparin cofactor II (SERPIND1) which inhibits thrombin in a dermatan sulfate–dependent manner in the arterial vasculature; protein C inhibitor (SERPINA5) that inhibits thrombin in complex with thrombomodulin; and Protease nexin-1 (SERPINE2) that inhibits thrombin at the vessel wall and platelet surface. The quantitative interplay among these positive and negative modulators is critical to the normal regulation of clotting, facilitating the rapid formation of a protective clot at the site of injury, while limiting and physically confining the process.

These events are outlined in the drawing: black arrows connect the substrates (inputs) and products (outputs) of individual reactions, and blue lines connect output activated enzymes to the other reactions that they catalyze.

Literature references

Kisiel, W., Fujikawa, K., Davie, EW. (1991). The coagulation cascade: initiation, maintenance, and regulation. *Biochemistry*, 30, 10363-70. ↗

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