

The Clot Thickens: Investigating Thrombin Activation of Blood Coagulant Factor XIII

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The final steps of blood coagulation involve fibrinogen, the serine protease thrombin, and the transglutaminase Factor XIII (FXIII). Thrombin cleaves the N-terminal portions of the A α and B β chains of fibrinogen releasing fibrinopeptides A and B (FpA and FpB). This action converts fibrinogen into fibrin which non-covalently self associates into a soft clot. Thrombin also helps to activate FXIII by hydrolyzing the R37-G38 peptide bond within FXIII. The activated FXIII later catalyzes the formation of covalent crosslinks in the fibrin network and for fibrin-enzyme complexes. As a result, a clot that is structurally more stable and degradation resistant is created (1). About 30% of the world-wide population possesses a common polymorphism in which a valine (V34) located in the FXIII activation peptide segment is replaced with a leucine (L34). This conservative change of adding a single methylene group has been correlated with protection against heart attack (2, 3). Biophysical studies are being carried out to better understand how the FXIII activation peptides interact with the thrombin catalytic surface and the roles played by individual substrate and enzyme amino acids.

Kinetic and NMR data indicate that the V34L substitution greatly enhances the ability of thrombin to hydrolyze the FXIII AP (²⁸TVELQG^VVPRGVN^L⁴¹) segment. Benefits are seen both with binding and with catalytic turnover. The key residues that interact with the thrombin active site surface include FXIII AP (³⁴V/L V P R³⁷). An ability to promote through-space interactions between L34 and P36 appears to be a hallmark feature of the V34L polymorphism (4-6). The only other residue thus far to achieve this effect is F34 (7). Thrombin utilizes specific amino acids to create unique binding environments to accept its substrates. The thrombin mutants W215A and W215A/E217A (WE) are helping to probe contributions from the aryl and apolar regions. With these mutants, key features required to orient the FXIII peptides for effective hydrolysis are being hindered. Further knowledge on the roles of individual thrombin and FXIII residues may lead to new strategies to control blood coagulation/anticoagulation processes.

References:

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