**Intrinsic pathway**

Damage tissue

Kallikrein*

[XII] [XI XII*]

**Extrinsic pathway**

Trauma

[IX] [IX* & VII VII* & tissue factor VII]

[X & X*]

Prothrombin

Thrombin*

Fibrinogen

Hirudin

Inactive Thrombin-hirudin complex

Fibrin

**Figure 1.3:** Thrombin, a serine protease (*) is dominant in both intrinsic and extrinsic pathways of the blood-clotting cascade. The blood-clotting process is accelerated through a signal amplification mechanism (denoted as pine, curved arrows). Coagulation time is prolonged if thrombin* is inactivated by binding to hirudin. The active and inactive forms of blood-clotting factors are in pink and green respectively.

This amplification mechanism can increase thrombin concentration in blood to 140 nM upon vascular injury (Walz et al., 1985). However, uncontrolled production of thrombin results in thrombosis intravascular coagulation, a phenomenon often associated with major surgery. A clinical resolution of this problem is to find inhibitors, for example, heparin and hirudin with high potency towards thrombin inhibition. Traditionally,
**Figure 2.1:** Smoluchowski's model consists of two interacting spheres, reaction occurs instantaneously once the sphere-A comes in contact with reactive surface of sphere-B.
Figure 2.2: An association reaction occurs only when two heterogeneous reactive patches come together.
Figure 2.3: A diagram illustrates the original model that used by Northrup, Allison and McCammon (1984) to do BD simulation.
Figure 2.4: A schematic representation of diffusion influenced reaction system (Analytical treatment of q-surface in Northrup, Allision and McCammon algorithm). Here \((b, \theta, \phi)\) are the spherical coordinates of the new when the previous position on the \(m\) surface is put on the polar axis. (A reproduced figure from Luty \textit{et al.}, 1992).
Figure 2.5: A practical approach to handle a variable dielectric constant in space is to model the protein system as a dielectric continuum, with two discrete dielectric regions, namely a low dielectric protein region ($\varepsilon=4$) and a high dielectric solvent region ($\varepsilon=78.5$).
Figure 2.6: A water atom with radius (1.4 Å) is used as a probe for rendering the solvent accessible surface of proteins.
Figure 3.1: A schematic representation of the SOD active site and the definition of the reactive spherical shell (in between the two red-dotted lines). The Cu atom is modeled as a (green in figure) 1.87Å sphere.
Figure 3.2: Illustration of collision-free distance $r_{surf}$ ($r_{surf} = \text{radius of superoxide} + \text{distance between protein geometric center and the SOD outermost atom} + \text{radius of the SOD outermost atom}$).
**Figure 3.3:** A two-dimensional representation of partitioning of protein atoms into octants and the collision detection mechanism. The circles in blue and in green are protein atoms and the red circle represents an instance of the moving substrate particle. The circle in blue belongs to the octant 1 and the circle in green belongs to both octants 2 and 3. Therefore, collision detection is needed for overlapping octant-3 only, which is shared by substrate and protein particle. Sequential collision detection is thus quickened by three fold in the three-dimensional situation.
Figure 4.1: The positive and negative electrostatic potential on the SOD surface is shown in blue and red respectively. Positive potential is increased by charge mutation(s) of the labeled residues around the active site of SOD.
Figure 4.2: The surface potential of native barnase. The electrostatic color-code is: blue, positive; red, negative. The residues present in the interface are labeled.

Figure 4.3: The surface potential of native barstar. The interface atoms are labeled.

4.3.2 BD simulation

At infinite ionic strength, the rate constant for the barnase-barstar association obtained from BD simulations is of the same order of
Figure 4.4: Comparison of calculated and experimental results for the free energy of barnase-barstar "encounter" complex at transition state.
Figure 5.1: Graphic representation of electrostatic potential dominance on rate enhancement. The potential in the active site and near region is "dominant" whereas the potential in the far region is "recessive" on rate enhancement.
Figure 5.2: The positively charged binding site of (Fe$^{3+}$)Trx-SOD is surrounded by negatively charged residues (Asp9, Asp10, Asp15, Asp61 and Glu31). This resembles the electrostatic environment of native SOD. Charge neutralization of the negatively charged residues will enhance the local ABF and the overall reaction rate.