

P017 Redox regulation of the thiol isomerase activity of the platelet integrin $\alpha_{IIb}\beta_3$.

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The molecular mechanisms involved in regulating integrin conformational switches are unknown. However, as integrins are known targets for redox modulation, we investigated the effects of altered redox conditions on the functions of the platelet specific integrin $\alpha_{IIb}\beta_3$. We have previously identified an endogenous thiol isomerase activity in this integrin that may regulate integrin activation states. The universal integrin activator, Mn^{2+} , stimulates thiol isomerase activity in purified $\alpha_{IIb}\beta_3$. Kinetic analysis reveals that $\alpha_{IIb}\beta_3$ is an allosteric enzyme that displays positive cooperativity (apparent Hill coefficient of 1.9). Altering the redox conditions by the addition of a nitric oxide donor in combination with glutathione (NO/GSH) causes a significant increase in the saturability of the enzyme activity without affecting the Hill coefficient. In parallel, in intact platelets Mn^{2+} results in activation of the integrin as determined by PAC-1 binding. NO/GSH, added after activation, specifically reverses this activation state. Neither Mn^{2+} or NO/GSH has any effect on platelet secretory responses indicating an integrin-specific effect. Thus, redox agents simultaneously modulate the thiol isomerase activity of $\alpha_{IIb}\beta_3$ and its active conformation in intact platelets suggesting a molecular mechanism for integrin regulation.