Protein tyrosine phosphatase 1B (PTP1B) is an important member of protein tyrosine phosphatase (PTPs) family that shares a conserved catalytic cysteine thiol residue. PTP1B acts as a negative regulator of insulin mediated signaling cascade, as inhibition of PTP1B is shown to increase the insulin sensitivity. A key feature of signal transduction pathway involves transient inactivation of PTP1B by an insulin-stimulated burst of hydrogen peroxide. The catalytic cysteine thiol of PTP1B undergoes oxidation in presence of hydrogen peroxide to generate an inactive sulfenic acid, which is reactivated by the action of cellular thiols and reducing enzymes. However, recent evidences suggest an unique chemical transformation to generate sulfenyl amide (3-isothiazolidinone) from sulfenic acid at the active site of PTP1B. The mechanism for the formation of this novel sulfenyl amide is not clear. We employed 1,2-benzisothiazolidinone as chemical model to mimic the important functional groups present at the active site of enzyme and examined the chemical mechanism leading to the formation of sulfenyl amide. Our results supported a direct intramolecular attack of neighboring amide nitrogen onto the sulfenic acid to generate sulfenyl amide.

Generation of the novel sulfenyl amide heterocycle is believed to protect the enzyme against irreversible over-oxidation to sulfinic and sulfonic acids. However, as the claims lacked a strong experimental support, we used our chemical models to estimate the rate constant for the hydrogen peroxide-mediated over-oxidation of sulfenyl amide. Our results suggested that
sulfenyl amide does in fact resist irreversible over-oxidation compared to a sulfinic acid surrogate. Furthermore, we developed a method to generate sulfenyl amide during the hydrogen peroxide-mediated oxidation of PTP1B in aqueous buffer solution. In addition, we determined the rate constant for the formation and over-oxidation of sulfenyl amide at the active site of PTP1B.

We also utilized our model compounds to examine the chemical properties of “over-oxidized” intermediates. Our results suggested that over-oxidation of sulfenyl amide will generate a thiol reversible but hydrolytically labile sulfinyl amide. We extended our knowledge gained from the model studies and investigated the formation of sulfinyl amide during oxidative inactivation of PTP1B. Consistent with our expectations, we were successful in identifying a thiol reversible but hydrolytically labile intermediate at the active site of PTP1B. Furthermore, we determined the rate constant for the hydrolysis of sulfinyl amide formed in PTP1B. Overall, our findings provided first evidence for the formation of this novel thiol-reversible sulfinyl amide heterocycle during the redox regulation of PTP1B. In addition, our results suggest that formation of sulfinyl amide may protect the enzyme against irreversible over-oxidation.

In a different context, we examined possible chemical mechanisms underlying the biological activity of natural product S-deoxy leinamycin. We synthesized a crucial triggering unit 1,2-dithiolan 3-one moiety of the natural product and investigated the generation of reactive oxygen species using DNA cleavage assays. Our results suggested that the reaction of 1,2-dithiolan 3-one heterocycle found in S-deoxy leinamycin with thiols leads to the generation of persulfide intermediate (RSS\(^-\)). The persulfide intermediate then generates biologically active polysulfides, hydrogen sulfide and reactive oxygen species that may ultimately explain the biological activity of S-deoxy leinamycin.