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Seminar Title: “Characterization of an *H. pylori* virulence factor: relevance to glutathione salvage”.

$\gamma$ -Glutamyltranspeptidase initiates extracellular glutathione reclamation by cleaving the  $\gamma$ -glutamyl amide bond of the tripeptide, and is a member of the N-terminal nucleophile hydrolase superfamily. In this emerging class of proteins, the inactive proenzyme undergoes self-processing, leading to activation of the enzyme. *H. pylori*  $\gamma$ -glutamyltranspeptidase (HpGT) is a known virulence factor and was selected as a model system to study the mechanistic details of autoprocessing and amide bond hydrolysis. Biochemical characterizations of the enzyme indicate broad substrate specificity and suggest a functional role following infection. Structural characterizations of HpGT have provided unprecedented insight into the mechanistic details of autoprocessing and substrate recognition.

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I am an Assistant Professor at the University of Nebraska and a member of the Nebraska Redox Biology Center. My research program uses protein crystallography, kinetic studies, and other biophysical techniques to examine structure and function relationships in enzymes vital to cellular redox homeostasis. In particular, we are funded to investigate the mechanistic details of enzymes involved in maintaining reduced thioredoxin and glutathione pools. Recently, we described the structures of oxidized and reduced mouse mitochondrial thioredoxin reductase and are actively pursuing structural studies of the enzyme in complex with its various protein substrates. Comparisons of these structures and further biochemical studies will delineate the biological functions of thioredoxin reductase as well as its mechanism of action. Another active research area involves glutathione homeostasis. The ratio of reduced to oxidized glutathione is primarily maintained by glutathione reductase. However, absolute glutathione levels are controlled in part by glutamate cysteine ligase and  $\gamma$ -glutamyltranspeptidase, which catalyze the committed steps in glutathione biosynthesis and salvage respectively. In higher organisms, the catalytic activity of glutamate cysteine ligase is allosterically activated by a differentially expressed regulatory subunit. We are working to determine representative structures of glutamate cysteine ligase, its regulatory subunit, and the holoenzyme to examine the induced conformational changes in glutamate cysteine ligase that promote increased catalytic efficiency. Since glutathione biosynthesis can be limited by cysteine availability, we are also examining the molecular details of the glutathione salvage enzyme,  $\gamma$ -glutamyltranspeptidase. This ectoenzyme is synthesized as an inactive precursor that must undergo autocatalytic processing to yield active mature enzyme. We are now refining several structures of mature and unprocessed *H. pylori*  $\gamma$ -glutamyltranspeptidase, and these findings will be presented at the conference if I am selected. As outlined, my research program is directly related to the subject area of the conference and I would benefit from participation in this forum. My expertise would allow me to contribute to the stimulating discussions expected at Gordon Research Conferences.