Chemistry and Molecular Biology Department
and NIH Center for Protease Research
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# "Chemical Approaches to Monitoring Protein Phosphorylation"

**Dr. Mary Kay Pflum**
**Assistant Professor, Department of Chemistry**
**Wayne State University, Detroit, MI**

## Abstract

Protein phosphorylation plays a critical role in a variety of cellular functions. As a result, the monitoring of phosphoproteins in cells represents an important goal for proteomics research. Unfortunately, available methods to identify, quantify, and probe the functional role of phosphopeptides in a cellular context are challenging. To facilitate phosphoprotein analysis, two novel approaches to phosphoprotein and phosphopeptide purification are described. First, the oxidation-reduction condensation was shown to chemically capture phosphopeptides and phosphoproteins with high selectivity. Second, an enzymatic phosphorylation-dependent biotinylation reaction of proteins was developed for phosphoprotein detection and enrichment. To illustrate the utility of the methods, the phosphorylation states of two proteins, the cyclic-AMP response element binding protein and histone deacetylase 1 protein, are discussed. In addition, the chemistry was used to attach various functional probes to phosphoproteins. The combined chemical and enzymatic approaches lay the foundation for development of new chemical tools targeting the phosphoproteome.