SUMMARY

Post-translational (PTMs) modifications play an important role in regulating protein function. PTMs refer to the addition and removal of a spectrum of biochemical functional groups such as phosphate, acetate, various lipids and carbohydrates. There are over 500 PTMs known so far (1). Site-specific modification of proteins by PTMs including phosphorylation, and ubiquitination, are implicated in signaling pathways that regulate fundamental cellular processes including cell division, DNA replication, apoptosis, and metabolism. Deregulation of proteins involved in these important cellular responses can often lead to disease, and the enzymes catalyzing addition or removal of PTMs have therefore emerged as important therapeutic targets.

Protein phosphorylation is an important PTM known to occur in organisms from archaea, bacteria to eukaryota. Basically, it is an addition of phosphate group to a protein by an enzyme called protein kinase. Based on the specific amino acid residues modified by kinases, they fall into several families. Protein phosphorylations on the serine, threonine, tyrosine, histidine and aspartate residues are the most widely studied.

In a bacterial two-component system, kinases most often modify histidine and aspartate residues. With the advent of genomic sequencing, genes encoding serine-threonine kinases were found extensively amongst bacterial genomes. In the meantime, bacterial tyrosine kinases (BY-kinases) are also being identified. This demonstrates the complexity of bacterial phosphorylation system. Therefore, identification of substrates for these kinases becomes a challenge. Thus, the field of bacterial phosphoproteomics emerged in the past two decades. The mass spectrometry-based phosphoproteomic studies have unambiguously identified phosphorylation events in bacteria on serine, threonine and less commonly on tyrosine residues. Hence, the technological advancement in mass spectrometry based proteomics and immunoaffinity enrichment strategy enabled us to identify large scale low abundant phosphotyrosine proteins in a model organism *Escherichia coli* (*E.coli*). Then, I did the bioinformatics analysis of phosphorylated tyrosine proteome of *E.coli*.

In eukaryotes serine, threonine and tyrosine residues are established to be the frequent targets for kinases. These kinases and the modified proteins are grouped in complex protein networks. It consists of multiple kinases which elicit signals from a cell surface to its nucleus. Signal
transmission inside the cell involves protein changes on three levels which include regulated protein PTMs, protein–protein interactions and changes in the gene/protein expression. Using quantitative mass spectroscopy approach, we can capture the change in phosphorylated state of protein induced upon treatment with and without ligand. Thus, to decipher PTMs- mediated thymic stromal lymphopoietin (TSLP) and interleukin-17 (IL-17) signaling pathways, I annotated reactions from the SILAC-based quantitative phosphoproteomic data and the literature (mainly protein-protein interactions and gene/protein expression events). Both are important immune signaling pathways and are known to be involved in the development of the autoimmune disorders. The reactions specific to TSLP and IL-17 signaling are not available in any pathway database. Therefore, I developed a centralized resource and in silico models of the TSLP and IL-17 signaling pathways.

Because of the amount of data generated from the high-throughput studies, in silico approaches are needed to convert the high-throughput data into the meaningful form. In this proteomics era, analysis and integration of large data sets is challenging task. Thus, both proteomics and bioinformatics fields are emerging simultaneously.