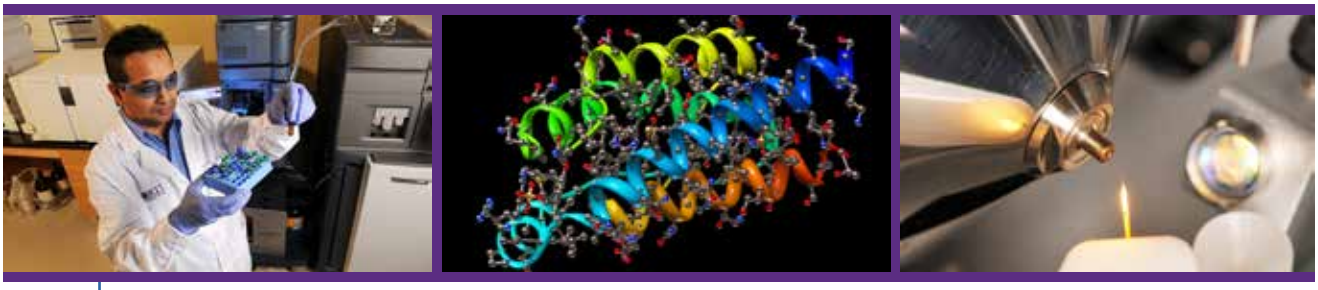
Protein Science and Proteomics



RTI International provides comprehensive protein analysis and proteomics profiling using state-of-the-art instrumentation. Our research activities include protein identification, peptide mapping, characterization of sites of protein modification, and use of label-free and stable-isotope-labeled differential proteomics approaches for unsupervised protein target discovery and validation. We use subcellular fractionation, enrichment, and targeted multiple reaction monitoring (MRM) approaches to characterize subproteome, post-translational modifications, and protein binding partners.

Protein sciences and proteomics measure the expression, post-translational state, and biological function of proteins, allowing us to derive detailed information about the molecular biology of the living cell and the organism.

Approach and Capabilities

RTI applies proteomics-based approaches in multiple scientific disciplines, including toxicology, drug discovery, and molecular medicine. For example, we identified and validated a novel role of proteasome in the activation of extracellular regulatory kinase.

In addition to broad-spectrum proteomics, we use targeted proteomics to investigate subcellular proteomes and generate the highest quality data relevant to the biological question. By employing different subcellular fractionation and enrichment techniques, we can characterize subproteome or post-translational modifications. For example, we use biotin-acyl chemistry to selectively enrich and profile palmitoylated membrane proteins.

We have also performed large-scale studies using 2-D gel electrophoresis, shotgun proteomics, target protein quantification and identification, and post-translational modification characterization on a variety of starting material, including bacterial and mammalian cell cultures, serum, plasma, saliva, and both fresh and formalin-fixed paraffin-embedded tissues.

Our protein science and proteomics laboratory supports several sample preparation methods, such as high-abundant-serum-protein depletion; membrane isolation; lipid-raft isolation and mitochondrial isolation; and enrichment of phosphopeptides, glycol-proteins, and palmitoylated proteins. We support labeled differential proteomics analysis such as SILAC (stable isotope labeling of cell culture) and iTRAQ technologies and label-free differential proteomics analysis for both gel-based and gel-free samples using high-resolution OrbiTrap as the mass analyzer. Additional research activities include characterization of a purified protein or peptides, protein-protein interaction mapping, and epitope mapping.

Instrumentation and Informatics

Our instrumentation capabilities include high-resolution mass spectrometry platforms, high-throughput discovery proteomics platforms, and quantitative targeted proteomics platforms. The bioinformatics workflow of our protein sciences and proteomics pipeline is supported by MASCOT, SEQUEST, and X!-Tandem for database searches and by Scaffold and ProteoIQ for data processing and compilation. The GeneGo pathway mapping tool is available to compile differential proteomics data and determine the target protein or pathway. Our protein sciences and proteomics program emphasizes the importance of sample preparation, analytical data collection, and proper bioinformatics distillation of analytical data to minimize false discovery rates.

Selected Publications and Presentations

Bencharit, S., S. Baxter, J. Carlson, W.C. Byrd, M.V. Mayo, M.B. Border, H. Kohltfarber, E. Urrutia, I. Saldarriaga, E.L. Howard-Williams, S. Offenbacher, M.C. Wu, and J.B. Buse. (2013). Salivary proteins associated with hyperglycemia in diabetes: A proteomic analysis. *Molecular BioSystems 9*(11), 2785–2797.

Carlson, J., S.A. Baxter, D. Dreau, and I.V. Nesmelova. (2013). The heterodimerization of platelet-derived chemokines. *Biochimica et Biophysica Acta—Proteins and Proteomics 1834*(1), 158–168.

Border, M.B., S. Schwartz, J. Carlson, C.F. Dibble, H. Kohltfarber, S. Offenbacher, J.B. Buse, and S. Bencharit. (2012). Exploring salivary proteomes in edentulous patients with type 2 diabetes. *Molecular BioSystems 8*(4), 1304–1310.

Merrick, A.B., S. Dhungana, J.G. Williams, J.J. Aloor, S. Peddada, K.B. Tomer, and M.B. Fessler. (2011). Proteomic profiling of S-acylated macrophage proteins identifies a role for palmitoylation in mitochondrial targeting of phospholipid scramblase 3. *Molecular and Cellular Proteomics 10*, 1–13.

Dhungana, S., B.A. Merrick, K.B. Tomer, and M.B. Fessler. (2009). Quantitative proteomic analysis of macrophage rafts reveals compartmentalized activation of the proteasome and of proteasome-mediated ERK activation in response to lipopolysaccharide. *Molecular and Cellular Proteomics 8*, 201–213.

Dhungana, S., J.G. Williams, M.B. Fessler, and K.B. Tomer. (2009). Epitope mapping by proteolysis of antigen—antibody complexes. Invited book chapter. Pp. 87–110 in *Methods in Molecular Biology, Epitope Mapping Protocols 524*. Edited by U. Reineke and M. Schutkowski. Clifton, NJ: Humana Press.

Dhungana, S., M.B. Fessler, and K.B. Tomer. (2009). Epitope mapping by differential chemical modification of antigens. Invited book chapter, Pp. 119–134 in *Methods in Molecular Biology, Epitope Mapping Protocols 524*. Edited by U. Reineke and M. Schutkowski. Clifton, NJ: Humana Press.

More Information

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