Collaborative Center for Translational Mass Spectrometry

at The Translational Genomics Research Institute
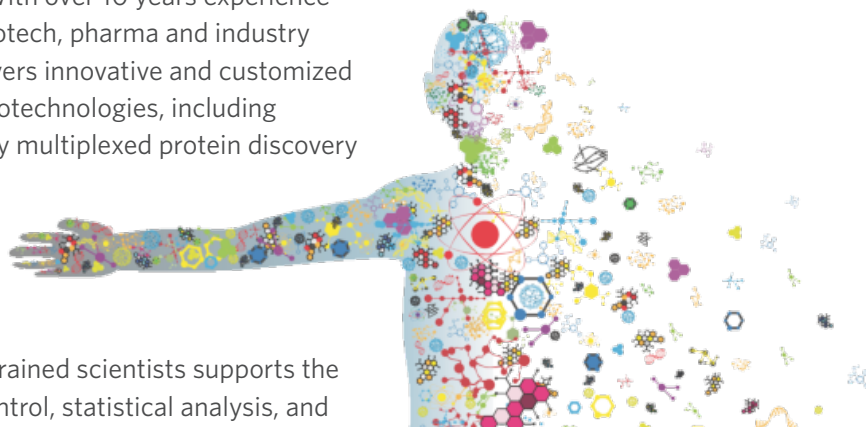
Pirrotte Lab

Collaborative Center for Translational Mass Spectrometry

Accelerating Translational Research and Precision Medicine through Proteomics

The Collaborative Center for Translational Mass Spectrometry (CCTMS) and the laboratory of Dr. Pirrotte is a full service next generation proteomics and small molecule based research facility providing end-to-end solutions tailored for translational and clinical research. With over 10 years experience collaborating with leading academic, biotech, pharma and industry partners, the CCTMS continuously delivers innovative and customized services in mass spectrometry based biotechnologies, including development and optimization for highly multiplexed protein discovery and quantification. Our platforms provide protein separation, characterization, identification and quantification services, featured with high throughput and super-sensitivity. A dedicated team of highly trained scientists supports the optimization of study design, quality control, statistical analysis, and biological interpretation.
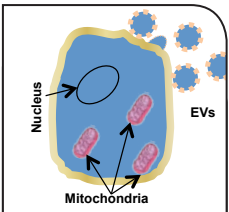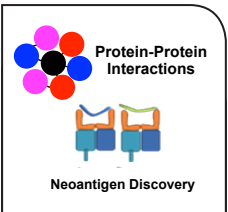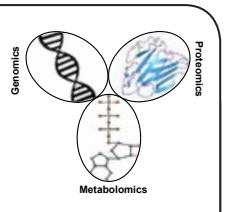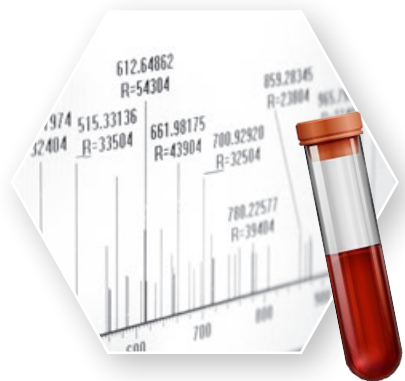
Disease and Systems Biology Applications

The myriad of proteomics and metabolomics applications cover different aspects of human health from identifying biomarkers in cancer to measuring drug delivery, distribution, kinetics and penetrance.

The CCTMS employs highly sensitive mass spectrometry approaches to perform ultra-deep proteomics profiling (i.e., >90% of the expressed proteome) in tissues and tumors. Assays are tailored towards clinical proteomics needs, such as FFPE proteomics or biomarker discovery in biofluids (e.g., plasma, serum, urine, saliva). Approaches for metaproteomic and metabolic profiling of microbial pathogens and microbiomes in longitudinal clinical samples have been established.

In addition, the Center has developed unique approaches that enable comprehensive and functional characterization of protein phosphorylation and kinome discovery to map key signaling pathways in support of the development of novel targeted therapies. Its bioinformatics pipeline provides data integration across proteomics, metabolomics, genomics and transcriptomics to provide a united and actionable overview of tumor biology in precision medicine clinical trials.

Organelle Profiling	Affinity Proteomics	PTM Analysis and Kinome Profiling	Metaproteomics & Metabolomics	Multi-OMICS Integration
Biomarkers in Circulating Biofluids	Cancer Immunotherapy Chromatin Remodeling	Oncogenic Pathways Kinase Inhibitors	Microbiome Studies Metabolic Signatures	ProteoGenomics Variant Analysis

Bioanalytical Services

The CCTMS provides quantification of proteins, metabolites and lipids with unbeatable precision and depth. Innovative services for highly multiplexed protein discovery and quantification in any species and in any biological matrix (*e.g.*, biofluids, cell lines, tissues, tumors and aspirates...), including high abundance protein depletion methods to minimize interference.
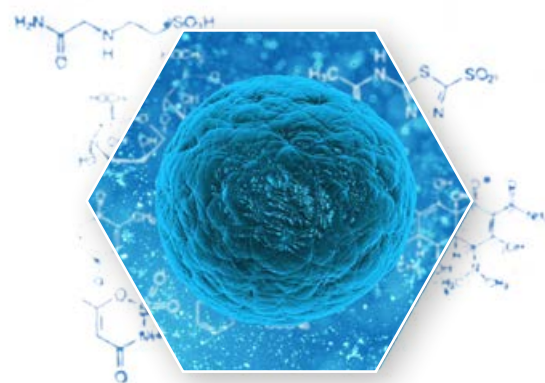
Our Expertise

- Comprehensive identification of proteins, metabolites and lipids in complex biological systems
- Bioinformatics pipelines to integrate proteomics with WGS/WES/RNAseq to observe mutations and genomic variants (*e.g.*, alternate splice junctions, insertions and deletions, fusions...)
- Neoantigen peptide identification and *de novo* peptide sequencing for applications in cancer immunotherapy (*e.g.*, HLA, MHC...)
- Organelle profiling, extracellular vesicles and membrane proteomics
- Oncogenic signaling pathway mapping using quantitative PTM and kinase-substrate analysis.
- On/off-target profiling of kinase inhibitors using unbiased kinome and enzyme profiling
- Characterization of protein-protein interactions to understand assembly of macromolecular complexes
- Metabolic pathway analysis and multiomics data integration
- Microbiome profiling and metaproteomics, identification of novel secondary metabolites in microbial pathogens

Technical Capabilities

- Shotgun proteomics using in-solution, in-gel or MED-FASP strategies
- Robust label-free or labeled proteome quantitation using isobaric/metabolic labeling strategies (*e.g.*, TMT, iTraq, SILAC, SISCAPA, PRM, MRM...)
- Enhanced proteome coverage using multidimensional pre-fractionation strategies, offline and online
- High abundance protein depletion methods for serum, plasma, saliva and urine
- PTM enrichment using targeted (antibody-based) or unbiased approaches (*e.g.* TiO2, IMAC...)
- Untargeted metabolomics and lipidomics, quantitative estimation of primary metabolites of hexose, amino acids, glycerophospholipids, sphingolipids classes and steroids
- Metabolic pathway tracing using stable isotope based fluxomics
- Small molecule analysis (*e.g.* PK/PD, impurity, conjugate analysis, pesticide exposure, assay development and validation...)

Biomarker Discovery & Validation

CCTMS technologies enable unbiased biomarker discovery approaches and targeted biomarker validation, thereby supporting the translation of basic science into applied clinical research. Our technologies enable patient stratification, disease diagnosis and monitoring, and surrogate endpoints. Ability to profile up to 12,000 proteins and 100,000 peptides per sample, up to 20,000 phosphorylation sites and quantify up to 100 target proteins per run in thousands of samples.

State-of-the-Art Facility

The CCTMS houses a 3,400 sq. ft. industrial scale, robotically automated, high-throughput biomarker discovery and validation facility. This facility has state-of-the-art instrument supported by a multidisciplinary team with more than 52 years of post-bachelor experience in the development and integration of targeted, proteomic technologies such as Multiple Reaction Monitoring Mass Spectrometry (MS) and antibody-based enrichment strategies for downstream LC-MS/MS analyses, shot-gun proteomics, PTM analysis (e.g. phosphoproteins), untargeted metabolomics, small molecules, and mass spectrometry based quantification. To address the computational demands required for proteomics data analysis, the CCTMS has access to a 3000+ core HPC system with multi-petabyte filesystems at a state-of-the-art third party data center that TGen partnered with Dell to build. There are also two high-memory SMP systems with 32 cores and 768 GB of RAM, as well as an SMP system with 40 processor cores and 1TB of RAM. These systems are connected to the main TGen network via a 10 Gb Ethernet connection from TGen HQ, allowing the timely transfer of large data sets between locations. The Center also utilizes a custom laboratory information management system (LIMS) for sample management and tracking.

Access from COH through Agilent iLabs solution forthcoming (June 2018).

Major equipment and software programs include:

Equipment

Mass spectrometers:
- Thermo Orbitrap Fusion Lumos ETD
- Thermo Orbitrap Velos Pro with ETD
- Thermo TSQ Altis Triple Quadrupole
- Waters SYNAPT G2 System

Liquid Chromatography:
- Thermo Scientific Vanquish UHPLC
- Dionex U3000 RSLC Nano 1D
- Dionex U3000 RSLC Nano 2D with microflow ternary pump
- Dionex U3000 BioRS Analytical UPLC
- Waters NanoAcquity UPLC
- Waters Acquity UPLC

Additional Equipment:
- Tecan proteomics platform (built-to-purpose)
- Gilson GX274 automated solid phase extraction
- Waters positive pressure system for 96-well plate
- Agilent Offgel Fractionator
- Pressure Bioscience Barocycler

Software

- Proteome Software Scaffold Q+
- Proteome Software Scaffold PTM
- Thermo Scientific Proteome Discoverer 2.2
- Thermo Scientific LipidSearch
- Thermo Scientific Compound Discoverer
- Bioinformatics Solutions PEAKS Studio
- MatrixScience Mascot
- X! TANDEM
- ProteinMetrics Byonic
- Nonlinear Dynamics Progenesis
- MaxQuant
- Perseus
- QIAGEN Ingenuity Pathway Analysis
- Pipelines for Quality Control (developed in-house)
    - proteogenomics
    - phosphoproteomics
    - protein-protein interactions

Director

Patrick Pirrotte, Ph.D.
Assistant Professor of Cancer and Cell Biology
Director, Collaborative Center for Translational Mass Spectrometry

Dr. Pirrotte holds a Ph.D. in Neurobiology and M.Sc. in Biomedical Imaging from the Louis Pasteur University in Strasbourg, France, presently the University of Strasbourg. He pioneered the use of high-resolution dynamic secondary ion mass spectrometry for life sciences. Prior to TGen, Dr. Pirrotte was co-founder and CEO of a surface-based analytics start-up that provided scientific decisional support for heavy industry. He joined TGen in 2014 as Director of CCTMS and is responsible for the scientific and technical development of the Center. His expertise is in biomarker discovery and validation pipelines, with specific interests in high throughput profiling of post-translational modifications. He has published key papers and patents in the field of proteomics and to date, contributed collaboratively to over $13.5M in contracts, grants and subawards to TGen.

Staff

Khyatiben Pathak, Ph.D.
Staff Scientist
Cancer biochemistry, metabolomics, lipidomics, microbial natural products, small molecules and pesticide analysis

Ritin Sharma, Ph.D.
Staff Scientist
Cancer biology, proteomics, kinase profiling, PTM analysis, targeted proteomics

Victoria David-Dirgo, B.S.
Laboratory Manager, Research Associate II
Microbiology, sample preparation for proteomics, PTM analysis, protein-protein interactions

Krystine Garcia-Mansfield, M.S.
Associate Bioinformatician
Proteomics and phosphoproteomic pipeline development, pathway analysis, protein-protein interactions

Apurva Hegde, M.S.
Associate Bioinformatician
Proteogenomics pipeline development, integrated omics data analysis

Blair Morris
Associate Bioinformatician
Multiomics and proteomics pipeline development, data management, user interface development

Marissa McGilvrey, B.S.
Research Associate II
Microbiology, sample preparation for proteomics, metabolomics, microbial natural products, small molecules and pesticide analysis

Kristin Leskoske, Ph.D.
Post-Doctoral Fellow
Cancer biology, signal transduction, PTM analysis