Diagnostic Technologies at the University of Chicago

May 2019
# Available Diagnostic Technologies

## Disease Diagnostics

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Disease Diagnostics
There is currently no standard clinical assay available to measure lipoprotein function, a key link in the triglyceride (TG) clearance pathway.

Dr. John Ancsin has developed a set of blood tests to rapidly screen for deficiencies lipoprotein function in patients with hypertriglyceridemia (HTG) in order to identify a treatment strategy.

Dr. Ancsin has demonstrated that the fluorescence-based lipase assays can assess the activities of lipoprotein lipase (LPL), apolipoprotein, and other plasma factors.

The lipase assays require only one blood sample to provide patient stratification and can also function as a companion diagnostic to support pharmaceutical development of new HTG therapies.

A US nationalized PCT is pending on systems for identifying patient specific deficiencies leading to HTG and informing treatment plans for those patients.

A more compact and efficient version of the lipase assay is under development.

Current methods of diagnosing IPF (X-ray, CT, echocardiogram) cannot predict disease progression or outcome.

Dr. Imre Noth has identified a molecular signature of 52 genes in the peripheral blood of patients with IPF that can reliably predict mortality outcome and assess IPF progression rate.

The technology can serve either as a prognostic or a diagnostic tool by monitoring the expression of the gene signature in peripheral blood mononuclear cells (PBMC).

A four-gene SmartChip qRT-PCR assay has been developed and successfully tested as a prognostic for IPF progression in 74 people.

The University holds both US issued patents and US pending applications on methods of using the biomarkers for assessing idiopathic pulmonary fibrosis.

The assay is ready for use in clinical settings.
Disease Diagnostics

Noth 15-T-034
Genetic Variants Predict Patient Response to N-acetylcysteine (NAC) Therapy for Idiopathic Pulmonary Fibrosis (IPF)

- N-Acetylcysteine (NAC), commonly used in conjunction with other therapeutic agents to break down mucous and lessen the overall decline in lung function, has historically demonstrated little benefit in patients with IPF.
- Dr. Imre Noth has identified loci in two genes, toll interacting protein (TOLLIP) and mucin 5B (MUC5B) whose SNPs indicate whether NAC therapy will improve or worsen the progression of IPF.
- A simple genetic screen of SNPs in TOLLIP and MUC5B allows the identification of IPF patients who will benefit or be harmed from NAC therapy.
- Out of 341 patients screened, the group receiving NAC showed significantly less risk in progression-free survival when they tested positive for both TOLLIP and MUC5B SNPs.
- US and European nationalized PCTs are pending directed to methods for treating idiopathic pulmonary fibrosis.
- Additional studies are underway to further evaluate the therapeutic potential of NAC in screened patient populations.

Cacioppo 15-T-024
Analytical Suite for Improved Time-Varied Data Segmentation

- Electroencephalography is a common method to detect problems in the electrical activity of the brain, however, rapid brain transition states are easily missed with current electroencephalogram (EEG) parameters, resulting in unreliable results and missed diagnosis of brain disorders.
- Drs. Stephanie Cacioppo and John Cacioppo have designed and applied an analytical suite that analyzes complex, high-density brain electrical signals that can distinguish between stable and transition brain states at specific time points and specific brain locations.
- The software program combines a number of various tools that utilize quantitative methods for robust and automatic detection of event-related changes in global brain activity.
- Researchers have measured EEG readings from human subjects and applied the analytical suite to measure changes in brain activity in response to various stimuli.
- The University holds an issued US patent directed to a non-transitory computer-readable medium to determine the stable states in the subject.
- Drs. S and J Cacioppo are looking for commercial partners to further develop and utilize the EEG analysis suite, and expand its application for other large data set analysis programs.
Diagnostic Imaging Technologies
# Diagnostic Imaging Technologies

**Carroll 17-T-074**  
Simultaneous Acquisition of Myocyte Oxygen Usage and Cardiac Strain Data in a Single MRI Scan

- Abnormal myocardial oxygen usage can predict heart attack or heart transplant rejection before the onset of clinical symptoms. However, there are currently no robust and reliable methods for globally measuring myocardial oxygen use and tissue strain despite the capability of this diagnostic tool to offer patients earlier access to lifesaving care.
- **Dr. Timothy Carroll** has developed a method to quantify global myocardial oxygen usage and tissue strain with anatomical imaging.
- Method compatible with existing MRI equipment. Oxygen and strain data are overlaid on the image for easy physician evaluation.
- A proof-of-concept study showed the capability of the method to use a single MRI pulse sequence to acquire two independent datasets and anatomical imaging.
- A PCT application is pending on combined oxygen utilization, strain, and anatomical imaging with MRI.
- The MRI pulse sequencing method is undergoing further validation studies in cardiac applications.

**La Riviere 11-T-132**  
3D Imaging Stain Gives More Complete, Accurate View of Pathology

- Histology images provide pathologists with a limited number of thin 2D slices while X-ray-based 3D imaging does not provide the biological specificity of histology stains.
- **Dr. Patrick La Riviere** has developed X-ray visible stains and computational imaging tools that combine the key benefits of 3D, X-ray-based imaging with histology.
- Method of staining tissues (biological, histological, or pathological samples) with multiple biologically specific heavy metal stains followed by X-ray imaging can generate high-resolution images of 1-2 microns.
- The technique has been validated in zebrafish larvae and juveniles, and can be extrapolated to any tissues traditionally stained with histology stains.
- A US patent is issued on 3D, color histology for multi-stained biological samples.
- The research team is working to scale up the technology to demonstrate it on cm length scales, such as pathology samples.
Diagnostic Imaging Technologies

Popko UCHI 12-T-018
PET Imaging Agent for Diagnosis and Monitoring of Multiple Sclerosis and Traumatic Brain Injury

- Readout from MRI correlates imperfectly with MS pathology; tools are needed to more accurately assess disease status.
- Dr. Brian Popko has exploited the exposure of potassium channels upon neuron demyelination as a mechanism by which to visualize the status and progression of MS.
- Derivatives of 4-aminopyridine, which bind to potassium channels, are labeled with an isotope to visualize disease progression or to monitor remyelination during therapy.
- The imaging agent has been demonstrated to highlight demyelination in rodent spines and brains in autoradiography and microPET experiments.
- US and European patents issued for an imaging agent made of radiolabelled 4-AP or derivatives and uses of the agent for imaging patients with demyelinating disorders. Patent pending in Canada and Australia.
- The investigators are conducting experiments to test the imaging agent in non-human primates

(Left) Luxol fast blue (LFS) staining of control mouse as compared to Shiverer mouse model of demyelination. (Center) 4-AP autoradiography of control mouse as compared to Shiverer mouse model. False color image used to highlight differences. (Right) Quantification of autoradiography signal in the corpus callosum of control and Shiverer (SHI) mice.
Molecular Diagnostic Platform Technologies
Affinity Clamps: A Platform Developing High-Affinity Synthetic Binding Proteins

- Affinity clamp technology could pave the way to understand complex physiological and pathological protein signaling networks and provide unique diagnostic and therapeutic agents.
- Dr. Shohei Koide has created a novel protein engineering platform for developing renewable, high affinity and high specificity antibody-like proteins to diverse and difficult targets in unstructured region of proteins, such as post-translational modifications.
- Current affinity clamps are targeted against phospho-tyrosines and can function as biosensors for the diagnosis of chronic myelogenous leukemia and Noonan syndrome.
- The level of affinity achieved with the clamp technology is three-four orders of magnitude greater than that of FLAG/antibody, c-myc/antibody, and 6xHis-tag/immobilized metal systems.
- Pending US patent application on platform technology and issued US patent on specific clamps.
- The affinity clamps have been optimized to bind a variety of small epitopes. Dr. Koide has also developed a novel protein capture system with a unique peptide fusion tag and its corresponding affinity clamp.

First-in-Class Recombinant Antibodies to Histone Post-Translational Modifications for Chromatin-based Diagnostics

- High-quality, reliable antibodies of high specificity are needed for chromatin-based diagnostics.
- Dr. Shohei Koide has created high-quality recombinant antibodies to histone post-translational modifications using tailored phage-display libraries.
- A series of recombinant antibodies have been generated against tri-methylated residues on histones 3 and 4 that may be useful for the diagnosis of breast cancer, renal cell carcinoma, and other cancers, or as companion diagnostics for histone-modifying drugs.
- Lead antibodies were identified from two libraries and validated against commercially available antibodies; the recombinant antibodies showed greater specificity and reproducibility than their commercial counterparts.
- Issued US patent on compositions and methods of diagnosis and drug screening.
- Histone antibodies to additional post-translational modifications are in development.
### Molecular Diagnostic Platform Technologies

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<th><strong>JUMP-seq: Methods for Detecting Cytosine Modifications</strong></th>
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<td>- 5-methylcytosine (5mc) and 5-hydroxymethylcytosine (5hmc) are important epigenetic markers in mammalian cells. However, current sequencing methods require micrograms of input genomic DNA and cannot obtain sequencing information on a single cell level.</td>
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<td>- Dr. Chuan He has developed new, bisulfite-free method, known as JUMP-seq, for mapping the location of 5hmc within genomic DNA with high resolution.</td>
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<tr>
<td>- The invention method attaches a probe to a modified 5hmc base; the steric hindrance causes a “jump” between DNA polymerase and the genomic DNA. The method identifies the presence of 5hmc by analyzing the jumping patterns to determine the 5hmc location.</td>
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<td>- The inventors have applied this method using only 24ng of genomic DNA, nearly half the input DNA required of the comparable TOP-seq method.</td>
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<td>- A PCT application is pending with claims directed to the JUMP-seq method for detecting 5hmc in a nucleic acid.</td>
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<td>- Interactions between two RNA strands or an RNA strand and protein can have a profound effect on gene expression. Therefore, methods to study these interactions are needed.</td>
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<td>- Dr. Chuan He has developed a kethoxal labeling reagent for use in labeling and crosslinking of nucleic acid allows for the study of RNA intermolecular interactions such as RNA-RNA interactions and RNA-protein interactions. Current chemical probes, such as DMA and SHAPE are limited by a lack of specificity and toxicity.</td>
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<td>- The invention kethoxal compounds can be used to label two ssRNA strands, which can then be crosslinked using dendrimers to study RNA-RNA interactions. Similarly, kethoxal can used to label a ssRNA strand and linked to a diazarine compound to crosslink a proximal protein of interest. Subsequent immunoprecipitation and sequencing enables the study of RNA-protein interactions.</td>
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<tr>
<td>- A PCT application is pending with claims directed to kethoxal compositions and methods of use as a labeling reagent, for studying RNA-RNA interactions, and for studying RNA-protein interactions.</td>
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<td>- The inventors are currently working to improve the CLIP-seq method by investigating other protein crosslinkers.</td>
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Engineered Protein G for Creating Multivalent, Bispecific Recombinant Affinity Reagents

- Protein G is widely used for the purification of antibodies, but current reagents lack specificity to Fabs and subject the antibodies to harsh conditions that may affect product quality.
- **Dr. Anthony Kossiakoff** has engineered Protein G (eProtein G) to bind to Fabs with higher affinity and specificity compared to native Protein G or Protein A.
- eProtein G can be used to purify recombinantly produced Fabs in a pH sensitive fashion or covalently tethered together to create multivalent recombinant affinity reagents to desired targets.
- In in vitro binding assays, use of eProtein G tethering of Fabs against a target antigen enhanced binding compared to the same concentration of Fabs alone.
- US and European nationalized PCTs are pending for compositions and methods of use of Protein G variants.
- Dr. Anthony Kossiakoff is expanding the use of the Protein G platform to diagnostic and therapeutic applications.

(A) Comparison of PH dependent affinity for canonical Protein-G (light grey) as compared to the engineered variants (black). (B) Base stability of canonical protein G (grey) as compared to engineered variants (solid black, dotted line).
How to Partner with the University of Chicago

For more information on partnering with the University of Chicago’s Polsky Center for Entrepreneurship and Innovation including start-up companies and licensable technologies:

polsky.uchicago.edu/tech-commercialization/
polskylicensing@uchicago.edu