UNIVERSITY OF MANITOBA

CANADA FOUNDATION FOR INNOVATION 15-6
Innovation Fund

Notice of Intent

1. Completed NOIs must be submitted by the Associate Dean (Research)/Research Liaison Officer of the "Lead" Unit to the Office of Research Services to: Birtukan.Gebretsadik@umanitoba.ca by May 15, 2018.

Proposed name of project: **Manitoba Nanoimaging Disease (MIND)**	**Estimated Total Project Costs:** approx. $6 Mill.
Designated Project Leader/Faculty/Dept: T. Klonisch, FHS, Human Anatomy & Cell Science (HACS) **List Principal Users/Faculty/Dept:** **1.** K. Coombs, FHS, Medical Microbiology **2.** M. Essig, FHS, Radiology **3.** P. Nickerson, FHS, Internal Medicine **4.** A. Halayko, FHS, Physiology **5.** L. Kirshenbaum, FHS, Physiology **6.**	
'Lead' Unit ADR/RLO: **Name: Rady Faculty of Health Sciences**	

Briefly describe (max 2 pages, 12 pt. font size, 2 cm margins):

- The proposed research and how it is world-class, innovative and demonstrates clear benefits to Canada.
- The infrastructure and how it will enhance the University's existing research capacity.
- The excellence of the team, including expertise and existing collaborations necessary to conduct the proposed research.
- Plans to secure matching funds and the potential funding sources for the operation and maintenance of the infrastructure.

MIND: Manitoba Nanoimaging Disease

The new **ManItoba Nanoimaging Disease (MIND**) node realizes a mutual research vision proposed by members of the Faculty of Health Sciences to establish a novel, unique, and world class platform in Canada that integrates advanced clinical imaging of animals and patients with 3D ultrastructural imaging of biological structures in health and disease. The proposed cutting edge microscopy equipment extends and empowers existing expertise in 3D super-resolution and bright field fluorescence imaging technologies at FHS, University of Manitoba. This CFI proposes a novel integrated strategy by combining clinical and ultrastructural imaging for an unprecedented insight into disease processes. We will use advanced clinical imaging of patients using established dynamic MRI and PET technologies to guide tissue sampling at disease sites. These biopsies are then processed and analysed employing cutting-edge three-dimensional (3D)-nanoimaging technologies. Emerging evidence emphasizes the importance of the tissue micro-environment as a key regulator of pathological processes and cellular responses to treatment. Currently, UofM is lacking the ultrastructural imaging equipment required to visualize the intricate cellular networks of normal and diseased tissue micro-environments. We recognize that biological studies on surgically removed bulk tumor tissue is of limited value. Dynamic MRI imaging and PET scans identify marked structural and functional heterogeneity within tumors and other diseases. Using different techniques, this heterogeneity can now even be traced to the single cell level. This reflects diverse cellular populations being inter-connected in complex and intricate cellular communication networks which shape their cell responses depending on the tissue micro-environmental context. The impact disease processes have on the dynamic cellular and structural composition and changes in biological functions of affected micro-environments is only starting to emerge. Our proposal focuses on the urgent need to integrate patient-centered and radiology-guided tissue acquisition with 3D volumetric ultrastructural imaging to target structurally and functionally distinct micro-environments and connectomes in patient tissues.

Patient tissues obtained by advanced clinical image-guided tissue biopsies are processed for structural image analysis using a world class high resolution field emission gun (FEG) 200keV **TITAN TALOS** transmission electron microscopes (TEM) with integrated cryogenic sampler technology. This cryo-TEM instrument is tasked with applications that extend the research capabilities and expertise far beyond existing boundaries to allow 3D image acquisition of subcellular structures in their native state. TALOS is an invaluable complementary tool to functional studies by providing ultrahigh resolution images of subcellular organelles alloing us to nano-visualize cell stress/death, cell-cell connections in tissue/disease specific micro-environments, monitor viral life cycle in human cells, and study (auto)immune tissue responses in transplants, tumor or neurodegenerative diseases, to give just some examples.

Dual beam focused ion beam (FIB) SEM capacity with cryotissue processing and automated milling technology provides 3D SEM (scanning EM) imaging of complex healthy and diseased patient tissue regions of defined metabolic states, including heart, kidney, lung, brain or virally infected cells and tissues. The dual beam SEM is part of a new **correlative microcopy workstation** which includes a specialized **confocal fluorescence microcopy unit** enabling the seamless integration of SEM images with localized fluorescent signals of labeled antigens and advanced morphometry in these tissues. These new capabilities of **MIND** complement existing super-resolution SIM and conventional 3D fluorescence microscopy at the imaging platform of the Dept. of Human Anatomy and Cell Science (HACS).

MIND will propel the University of Manitoba to becoming the first place in Canada that directly links patient advanced clinical imaging-guided biopsy collection with 3D volumetric nano-resolution capture of ultrastructural changes. **MIND** addresses key research themes within the UofM. Using a set of carefully selected high-end microscopes capable of correlative imaging of biopsy materials mapped to

structurally and functionally distinct tissue regions provides unique **and integrated** high-resolution "zoom lens" to visualize dynamic ultrastructural changes in normal and diseased tissue micro-environments. This innovative ultrastructure platform has the potential to empower translational research, spark new clinical research directions, and reveal unique structural insights into treatment failure, development of drug resistance, immunological tissue rejection or tumor cell niches. **MIND** also elevates the analysis of genetically modified animal models. **MIND** also sparks new exciting alliances by combining correlative immunofluorescence protein detection with proteomic analysis on tissue sections. Glioma brain tumors exemplify how **MIND** excels translational research. As a systemic brain disease, glioma tumor cells co-exist with normal brain cells for several years (low grade) and infiltrate cortical and subcortical brain networks. Glioma patients show lower global brain network efficiency believed to contribute to cognitive impairment, resulting in difficulties in processing speed, executive functioning, verbal learning, and language. These symptoms are positively correlated with increasing tumor volume and grade and likely reflect altered neuronal connectivity as brain cells are forced to adapt to the presence of tumor cells. Structural changes in brain connectomics and the extent/ dynamics of underlying cellular and molecular adaptations in glioma patients are largely unknown. Combining advanced MRI imaging-guided tumor biopsy with **MIND** will 3D visualize unique ultrastructural changes in networks of patient brains with high and low tumor burden and vascularity. Combined use of TALOS, FIB-SEM and laser-scanning microscopy for multicolor immune-tracing of specific cellular/subcellular markers permits exceptional subcellular imaging down to low-nanometer resolution of intricate cellular network interactions to reveal new structural and molecular relationships between the brain connectomes and tumor cells. The Klonisch team complements these studies with the use of mouse xenograft models of patient-derived brain tumor initiating stem-like cells which have well-defined molecular parameters to interrogate the effect of specific tumor promoters and tumor plasticity on brain networks.

By closing the existing imaging gap between advanced clinical imaging and high-resolution structural imaging, **MIND** will spur discoveries into novel disease concepts and inform/monitor new therapeutic strategies. **MIND** will be embedded within the established Histomorphology and Ultrastructural Imaging service platform at HACS and excel EM services to the entire community of health researchers at UofM and other R&D communities in Manitoba, in Canada and abroad.

The team has recognized expertize in:
(i) Electron microscopy and 3D fluorescence imaging (Coombs, Klonisch)
(ii) Dynamic MRI and PET functional imaging and clinical decision making (Essig),
(iii) CRC-I in Airway Cell and Molecular Biology (Halayko),
(iv) CRC-I in Molecular Cardiology, Director Institute of Cardiovascular Sciences (Kirshenbaum)
(v) Clinical researchers, immunology/biomarkers in renal transplantation medicine (Nickerson)
(vi) Coombs, Klonisch, Kirshenbaum, Halayko have mutual publications on various cell death topics. Klonisch has current grant support together with Drs. Coombs and Essig.

Lead: Dr. T. Klonisch, (Anatomy/ Surgery/ Medical Microbiology)
Principal Users: Drs. **K. Coombs** (Medical Microbiology, Manitoba Centre for Proteomics and Systems Biology), **M. Essig** (Radiology), **A. Halayko** (Physiology/ CHRIM), **L. Kirshenbaum** (Pharmacology & Therapeutics/ Inst. Cardiovascular Sciences ICS); **P. Nickerson** (Internal Medicine/Immunology)