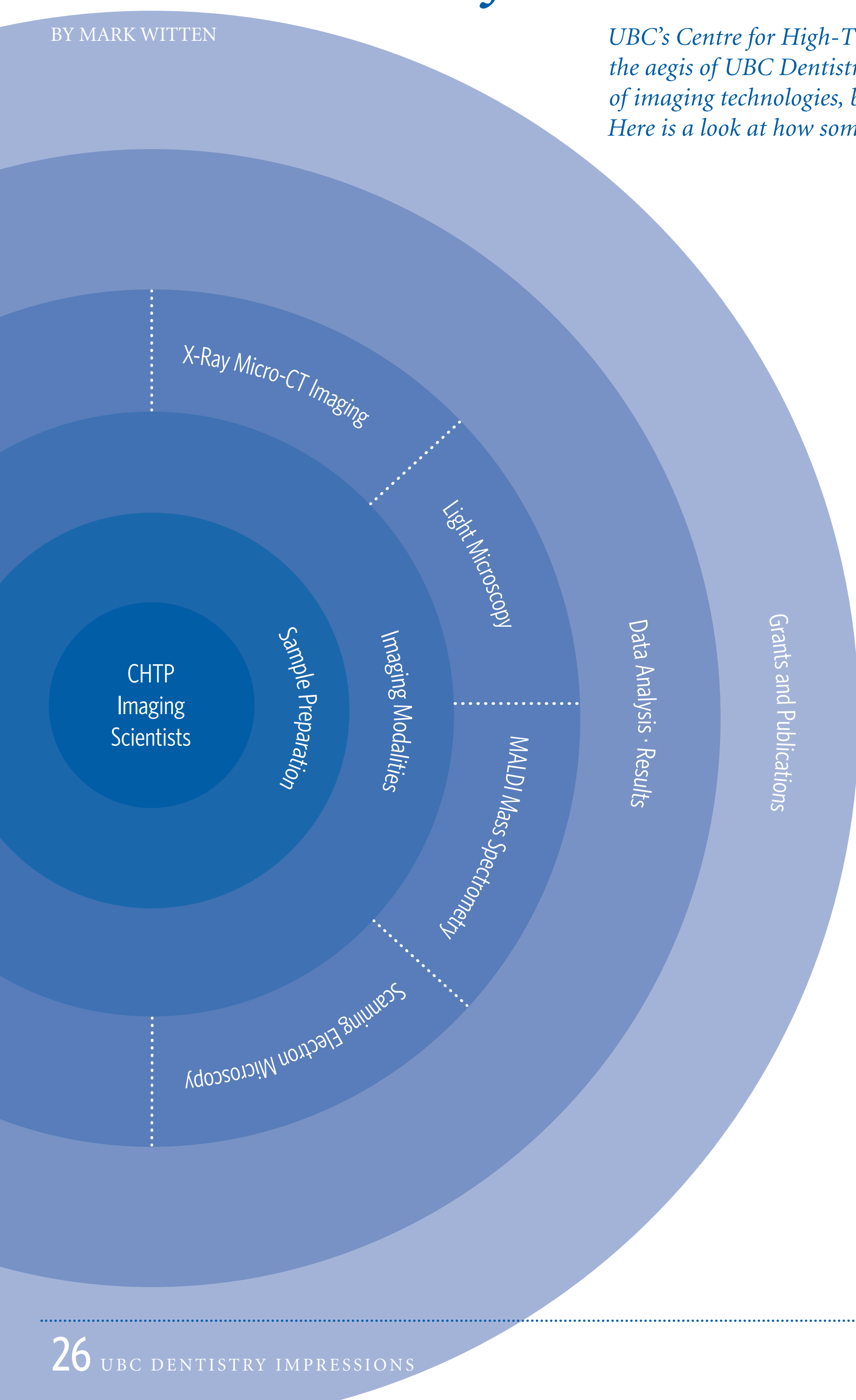
SPRING 18

Rings of Success: Imaging Scientists at the Core of the CHTP

BY MARK WITTEN



UBC's Centre for High-Throughput Phenogenomics (CHTP), under the aegis of UBC Dentistry, is home to not only a comprehensive suite of imaging technologies, but also a team of expert imaging scientists. Here is a look at how some recent studies benefited from their expertise.

MICRO-CT (MICRO-COMPUTED TOMOGRAPHY) IN VIVO SCANNER

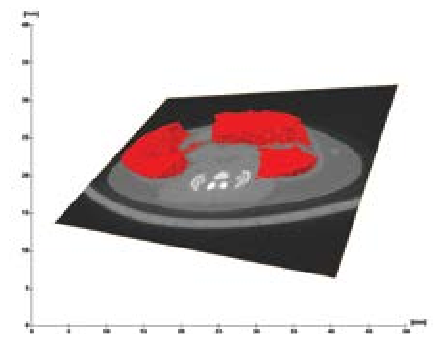
TriFoil eXplore CT 120

Researchers
Dr. James Johnson Lab; Dr. Søs Skovsø, postdoctoral fellow; UBC Department of Cellular and Physiological Sciences, Faculty of Medicine

Summary
The study found that reducing insulin production and secretion in mice made obese by a high-fat diet caused significant weight loss within five weeks. The data obtained through in vivo micro-CT scanning of soft tissues provided the first genetic evidence that obesity can be reversed by acutely and modestly reducing insulin production.

Value add of CHTP
Dr. Skovsø worked closely with a CHTP imaging scientist on both the micro-CT imaging and data analysis, and the project was able to use a pre-approved protocol for micro-CT scanning of soft tissues like fat and muscle. For Dr. Skovsø and other researchers in Dr. Johnson's lab, using micro-CT at CHTP provided greater accuracy and specificity compared to other methods such as micro-DEXA. It allowed the researchers to look at changes in fat distribution in the same mice over time. This enabled them to accurately visualize and measure changes, in both the harmful and less harmful types of abdominal fat, that resulted from lowering insulin levels. It also showed that lowering insulin production to normal levels reverses weight gain in adult mice fed a high-fat diet.

Publication
Page M, Skovsø S, Cen H, Chiu A, Dionne D, Hutchinson D, Lim G, Szabat M, Flibotte S, Sinha S, Nislow C, Rodrigues B, Johnson JD. (2017). Reducing insulin via conditional partial gene ablation in adults reverses diet-induced weight gain. *The FASEB Journal*. doi: 10.1096/fj.201700518R.



Micro-CT image (grey) with fat deposits highlighted red.

MICRO-CT (MICRO-COMPUTED TOMOGRAPHY) SPECIMEN SCANNER

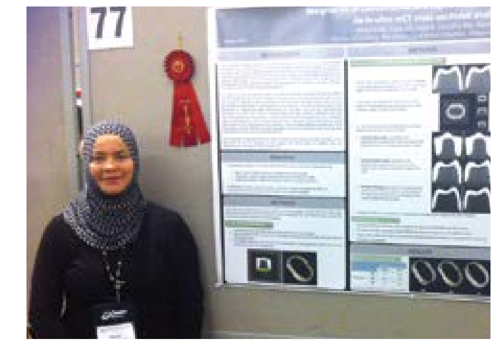
SCANCO Medical µCT100

Researcher
Dr. Nesrine Mostafa, then MSc candidate, now assistant professor in prosthodontics, UBC Department of Oral Health Sciences, Faculty of Dentistry

Summary
Dr. Mostafa's study used micro-CT scanning to systematically assess and compare the marginal fit of crowns using digital technology vs. crowns formed using a conventional lab approach. The study found digitally fabricated crowns showed better fit as compared to conventionally fabricated crowns. (Mostafa continues to access the CHTP for her current research.)

Value add of CHTP
This is one of the first studies to assess the marginal fit of ceramic crowns using a micro-CT scanner and a systematic 2D and 3D analysis. Micro-CT allows a thorough evaluation of crown margins at multiple locations without destroying the samples. It produces 2D and 3D data sets that can be used for quantitative analysis and for visualization by specifying cement thickness. It also allows assessment of the same sample before and after using differing testing variables.

Publication
Mostafa Z, Ruse N, Ford N, Carvalho R, Wyatt C. (2018). Marginal fit of lithium disilicate fabricated crowns using conventional and digital methodology: A three-dimensional analysis. *Journal of Prosthodontics*, 27(2): 145-152. doi: 10.1111/jopr.12656.



Dr. Nesrine Mostafa was awarded second place at the ACP Resident Poster Competition at the 45th American College of Prosthodontics Annual Meeting in Orlando, Florida, October 21 to 24, 2015.

OPT (OPTICAL PROJECTION TOMOGRAPHY) SCANNER

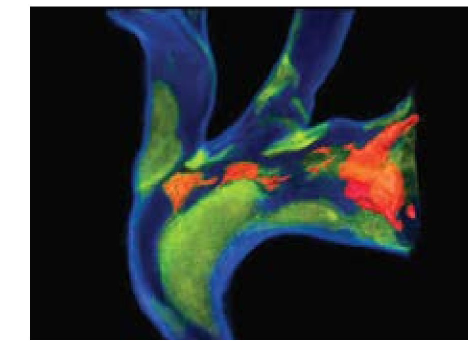
Bioptonics 3001 M

Researchers
Dr. Don Sin Lab, UBC Centre for Heart Lung Innovation; Dr. Jen-erh Jaw, postdoctoral fellow, UBC Experimental Medicine Program, Faculty of Medicine

Summary
A mouse model of plaque rupture was established in which lung exposure to lipopolysaccharide (LPS) leads to lung inflammation and causes acute plaque destabilization. The study also showed that, in this model, circulating neutrophils play an important role in both lung inflammation and plaque rupture. This model could be useful for screening therapeutic targets to prevent acute vascular events related to lung inflammation.

Value Add of CHTP
A CHTP imaging scientist advised Dr. Jaw that OPT would be the best equipment to use for measuring the size and vulnerability of plaques at different stages after exposure to LPS. Technical support for carrying out the experiment was also provided. OPT is an ex vivo micro-imaging tool that doesn't require tissue sectioning and thus preserves the architecture of the blood vessel. By combining the OPT technique with histological findings, the researchers provided convincing evidence that acute lung inflammation induces plaque rupture.

Publication
Jaw J, Tsurata M, Oh Y, Schipilow J, Hirano Y, Ngan D, Suda K, Moritani K, Tam S, Ford N, van Eeden S, Wright J, Man SF, Sin DD. (2016). Lung exposure to lipopolysaccharide destabilisation. *European Respiratory Journal*, 48(1): 205-215. doi: 10.1183/13993003.00972-2015.



Vulnerable atherosclerotic plaque (green) can rupture to form blood clots (red) in the aorta of a mouse. This image was generated by optical projection tomography.

MALDI (MATRIX-ASSISTED LASER DESORPTION/IONIZATION) MASS SPECTROMETER (MS)

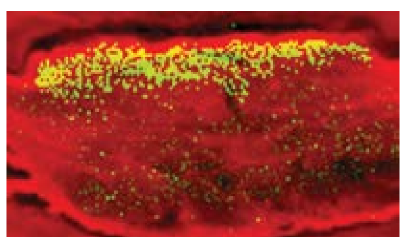
Thermo Scientific LTQ Orbitrap XL

Researchers
Helen Burt Lab; Dr. David Plackett, research associate; UBC Faculty of Pharmaceutical Sciences

Summary
A promising new treatment for superficial bladder cancer has been developed by UBC Pharmaceutical Sciences, the Centre for Drug Research and Development (CDRD) and CDRD Ventures Inc. (CVI). The treatment uses a chemically modified hyperbranched polyglycerol (HPG) as a drug carrier to achieve higher concentrations of the anti-cancer drug docetaxel (DTX) in the bladder wall than when this drug is administered alone in solution. MALDI-MS imaging showed the distribution and concentrations of DTX in surface layers of pig bladder tissue were higher when using an HPG formulation. The findings were comparable with results obtained previously using an established radiolabelled drug method, providing a useful and complementary alternative to the use of radiolabelled or other quantitative methods for measuring drug distribution and concentrations.

Value add of CHTP
MALDI-MS imaging offered a convenient way to visually map and compare the distribution of the new and commercial drug formulations that couldn't be provided by other methods. The imaging scientist's expertise in MALDI-MS also provided a detailed quantitative analysis of the concentrations of the various drug formulations at different tissue depths and locations to complement the qualitative analysis from visual mapping. MALDI-MS imaging is label-free, so it could be used to study drug distribution in the human body in clinical trials.

Publication
Plackett D, Mugabe C, Lin S, Ford N, Liggins R, Burt H. (2016). Hyperbranched polyglycerol-docetaxel treatments for bladder cancer and the characterization of treated bladder tissue using MALDI-MS imaging. Abstract presented at the International Conference on Nanomedicine and Nanobiotechnology in Paris, France, September 28 to 30, 2016.



MALDI-MS image of pig bladder tissue.

FIB-SEM (Focused Ion Beam Scanning Electron Microscope)

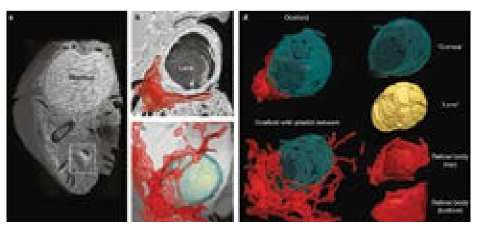
FEI Helios NanoLab 650 DualBeam

Researchers
Dr. Brian Leander Lab; Dr. Gregory Gavelis, postdoctoral fellow; UBC Departments of Botany and Zoology, Faculty of Science

Summary
Single-celled marine plankton, called warnowiid dinoglagellates, evolved a miniature version of a multicellular eye, possibly to help see prey better. The study showed this planktonic predator has an eye-like ocelloid structure, which contains a collection of subcellular organelles that look very much like the lens, cornea, iris and retina of multicellular eyes found in humans and other larger animals.

Value add of CHTP
A CHTP imaging scientist advised Dr. Gavelis that FIB-SEM could provide extremely high resolution images of a unicellular organism in its natural state and that cryofixation of samples would provide the most accurate and detailed reconstruction of a 3D model. The use of FIB-SEM enabled 3D reconstruction of the ocelloid structure and its component parts, including the mitochondrial cornea-like layer, vesicular lens and retinal body. The imaging scientist provided the technical support for the FIB-SEM imaging and 3D reconstruction.

Publication
Gavelis G, Hayakawa S, White III R, Gojobori T, Suttle C, Keeling P, Leander B. (2015). Eye-like ocelloids are built from different endosymbiotically acquired components. *Nature,* 523: 204-207. doi: 10.1038/nature14593.



3D reconstruction and modelling representation of the eye-like ocelloid structure of a marine plankton from FIB-SEM data.

FIB-SEM (Focused Ion Beam Scanning Electron Microscope)

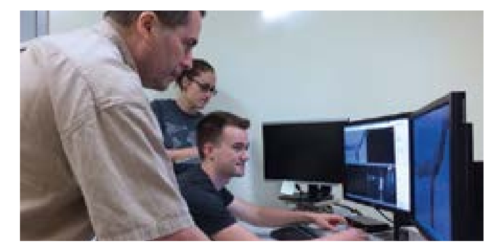
FEI Helios NanoLab 650 DualBeam

Researchers
Dr. Curtis Berlinguette Lab, UBC Department of Chemical and Biological Engineering, Faculty of Applied Science; Kevan Dettelbach, PhD candidate, UBC Department of Chemistry, Faculty of Science

Summary
The Berlinguette lab is focused on solar energy conservation and ways of storing solar energy using thin film materials. The thicknesses and metal loadings of amorphous nickel, iron and iridium oxide films widely used for solar fuel electrocatalysis were determined by cross-sectional SEM and X-ray fluorescence (XRF) spectroscopy measurements. XRF measurements provided a strong linear correlation with the thicknesses determined by cross-sectional SEM. These results highlight that the non-invasive XRF technique is demonstrated to be far superior to the widely used electrochemical surface area (ESA) technique for reporting on thickness and metal loading of thin metal oxide films.

Value add of CHTP
It can be a challenge to measure the thickness of nanometre-scale thin films of low-conductance materials. Cross-sectional SEM accurately determined the thickness of a series of films at the nanometre level on iridium-coated glass samples, which would be impossible to measure with other methods. The CHTP imaging scientist recommended dealing with conductance issues by sandwiching the films between two iridium layers. Using the high resolution SEM, the film thicknesses were imaged and measured, and the backscattered electron imaging capabilities of the machine easily distinguished the thin layer from the iridium layers. This method, which allowed the researchers to get accurate thickness measurements rapidly, can be applied to a wide variety of thin film materials.

Publication
Dettelbach K, Kolbeck M, Huang A, He J, Berlinguette C. (2017). Rapid quantification of film thickness and metal loading for electrocatalytic oxide films. *Chemistry of Materials*, 29(17): 7272-7277. doi: 10.1021/acs.chemmater.7b01914.



Members of the Berlinguette lab using the FEI Helios NanoLab 650 DualBeam.

FIB-SEM (Focused Ion Beam Scanning Electron Microscope)

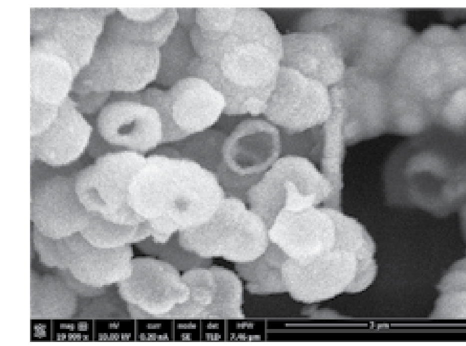
FEI Helios NanoLab 650 DualBeam

Researchers
Dr. David Wilkinson Lab; Baizeng Fang, senior research scientist; UBC Department of Chemical and Biological Engineering, Faculty of Applied Science

Summary
Photocatalytic conversion of CO_2 into a clean energy fuel is a promising avenue for sustainable development. In this study, a one-pot template-free synthesis strategy was used to fabricate CuO-incorporated TiO_2 hollow microspheres in large scale. These were explored for the first time as a catalyst for photodriven conversion with the aid of H_2O, of CO_2 to CH_4 fuels. Because of its unique structural characteristics, the catalyst demonstrates much higher photocatalytic activity toward CO_2 reduction with H_2O into CH_4, compared with a current state-of-the-art photocatalyst. Also, the simple synthesis strategy would enable large-scale production of the hollow microspheres.

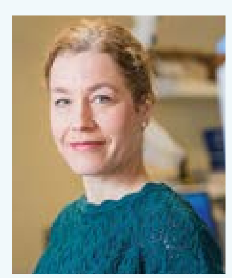
Value add at CHTP
FIB-SEM provided the extremely high resolution scanning needed to visualize and explore the surface morphologies of these TiO_2 hollow microsphere catalyst samples in the nanoscale range. SEM allowed the researchers to look at the newly created catalyst structures with various magnifications and to confirm that TiO_2 hollow microspheres formed and performed as predicted.

Publication
Fang B, Xing Y, Bonakdarpour A, Zhang S, Wilkinson D. (2015). Hierarchical CuO-TiO_2 Hollow Microspheres for Highly Efficient Photodriven Reduction of CO_2 to CH_4. *ACS Sustainable Chemistry & Engineering*, 3(10): 2381-2388. doi: 10.1021/acssuschemeng.5b00724.



FIB-SEM image of the surface morphology of a TiO_2 hollow microsphere catalyst sample.

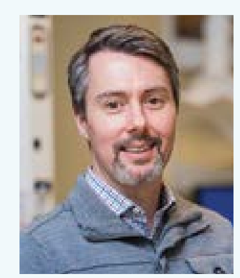
CHTP IMAGING SCIENTISTS



Dr. Nancy Ford, Director, CHTP

Associate Professor, UBC Department of Oral Biological & Medical Sciences, Faculty of Dentistry; PhD, Medical Biophysics, University of Western Ontario

- Is the director of the CHTP, where her work involves optimizing images, minimizing X-ray exposure and navigating massive data sets.
- Research areas and specialties include micro-CT, X-ray imaging, physiological gating, small animal imaging, image quality and cone beam CT.



Dr. Gethin Owen, Technical Director of Electron Microscopy

PhD, Cell Biology, AO Research Institute, Davos, Switzerland

- Manages the SEM facility and has more than 20 years' experience using scanning and transmission electron microscopy as an imaging scientist, researcher and graduate student.
- Provides specialist FIB-SEM imaging services, user training on sample preparation and operating equipment, project design and novel technique development.

Dr. Guobin Sun, X-ray and Optical Imaging Technician

PhD, Cellular and Molecular Biology, UBC

- Provides full-service technical support, training and consulting to users on Zeiss PALM, Olympus LEXT confocal, Nikon Eclipse Ci and Leica SP5 X microscope systems.
- Skilled in micro-CT and OPT imaging, and provides a full range of support, training and consulting services on these instruments to users.

Shujun Lin, MALDI Mass Spectrometry Technician

MSc, Biochemistry, Memorial University

- Set up, operates and maintains the Thermo Scientific LTQ Orbitrap XL mass spectrometer.
- Lin has more than 10 years' experience using MALDI-MS and provides full-service support for users, including sample preparation, data acquisition, imaging data processing and interpretation of results.

Meet the Imaging Scientists at the CHTP

Dr. Nancy Ford, Director
Associate Professor, UBC Department of Oral Biological & Medical Sciences, Faculty of Dentistry; PhD, Medical Biophysics, University of Western Ontario

Dr. Ford is the director of the CHTP, where her work involves optimizing images, minimizing X-ray exposure and navigating massive data sets. Her research areas and specializations include micro-computed tomography (micro-CT), X-ray imaging, physiological gating, small animal imaging, image quality and cone beam CT. Her research focuses on optimizing and accessing imaging protocols for preclinical and clinical computed tomography. Her previous research experience includes working as a research assistant in a digital mammography research group at Sunnybrook Hospital in Toronto, a postdoctoral fellowship in the Imaging Research Labs at the Robarts Research Institute in London, Ontario, and a position, from 2006 to 2011, as assistant professor in the Department of Physics at Ryerson University.

"The centre," says Ford, "has a whole variety of sophisticated imaging instruments available in the same place, including the only MALDI mass spectrometer, the only focused ion beam scanning electron microscope and the only micro-computed tomography specimen and in vivo scanners on the UBC campus. We have a team of imaging scientists with specialized expertise and knowledge. They are dedicated full-time to supporting and training users, and optimizing the techniques to help researchers get the best-quality images and data. The limits of science are pushing forward to visualize smaller and smaller structures. Researchers need to use these more sophisticated and challenging techniques and equipment to solve today's and tomorrow's research problems."

Dr. Gethin Owen, Technical Director of Electron Microscopy
PhD, Cell Biology, AO Research Institute, Davos, Switzerland

Dr. Owen manages the electron microscopy imaging facility and provides specialist imaging services, consulting, user training, project design and novel technique development. He has more than 20 years of experience using scanning and transmission electron microscopy in basic research investigations for both biological and material samples. His previous research experience includes postgraduate work at AO Research Institute, Davos, Switzerland, and postdoctoral fellowships at the New York Structural Biology Center, New York, USA, and the UBC Department of Oral Biological & Medical Sciences, Faculty of Dentistry.

"All of us working at CHTP have an interest in the research users are doing and with our research background can understand the concepts pretty easily. We can advise on the best sample preparation and imaging methods to collect the best-quality data. This is a collaborative kind of work. For example, we often provide technical assistance to prepare and image samples that are difficult, or almost impossible, to image at high resolution because of their constitution. When the researchers agree to use an alternative method that we propose, they not only benefit by getting reliable, high-quality data, but results that would otherwise be impossible to obtain," says Owen.

Guobin Sun, X-ray and Optical Imaging Technician
PhD, Cellular and Molecular Biology, UBC

Dr. Sun has been managing the light microscopy section at CHTP since 2013, and he also manages the micro-CT and optical projection tomography (OPT) imaging equipment and services. Sun provides full-service technical support, training and consultation to users on Zeiss PALM, Olympus LEXT confocal, Nikon Eclipse Ci and Leica SP5 X microscope systems, micro-CT specimen and in vivo scanners, and the OPT scanner. He gained five years of fluorescent imaging experience as a graduate research assistant and research advisor in the cell biology lab of Dr. Catherine Pallen, professor and associate member, UBC Department of Pathology and Laboratory Medicine.

"We're a core facility with many different imaging modalities and diverse users. When researchers come here, they want to get data. We have the equipment and expertise to help them get the best-quality data as quickly as possible, which facilitates and accelerates their research. We can advise users on which imaging method will provide the best measurements of samples and get the highest quality imaging for their purpose," explains Sun. He notes that researchers can use different imaging instruments at the CHTP for the same project, to obtain more data and strengthen their results.

Shujun Lin, MALDI Mass Spectrometry Technician
MSc, Biochemistry, Memorial University

Shujun Lin operates and maintains the Thermo Scientific LTQ Orbitrap XL mass spectrometer, which she set up at the CHTP in 2014. Lin has more than a decade of experience using MALDI mass spectrometry methods to detect peptides, hormones, lipids and drug metabolites in biological specimens, and to determine their relative abundance and distribution in various tissue sections. She has over 20 years of experience using mass spectrometry at the CHTP, the UBC Biomedical Research Centre, Biovail Contract Research (Toronto) and Louisiana State University.

"Users bring us tissue samples and we do the full service," says Lin. "I do everything from setting up protocols, sample preparation, data acquisition, imaging data processing and interpretation of results. Our service is fast, and users appreciate that."

PHOTOS BY PAUL JOSEPH