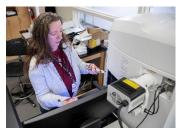




# **NBCC Advanced Imaging Guide**

The Network Biology Collaborative Centre (<a href="nbcc.lunenfeld.ca">nbcc.lunenfeld.ca</a>) at the LTRI offers a wide range of microscropes from day-to-day workhorses to super-resolution instruments. This guide provides information on the imaging applications we offer and the instrumentation that is available for analysis.

#### I. Who we are





The NBCC Advanced Imaging node is overseen by LTRI's Senior Scientist and NBCC co-Director Dr. Laurence Pelletier who is an expert in microscopy and super-resolution applications. The facility is co-managed by Louise Brown who has over 20 years' experience within LTRI's microscopy core and by Dr. Johnny Tkach who oversees the super-resolution microscopes. Director of Operations Dr. Karen Colwill provides administrative and logistical support.

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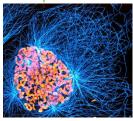
Website: <a href="mailto:nbcc.lunenfeld.ca/facilities/advanced-imaging">nbcc.lunenfeld.ca/facilities/advanced-imaging</a> Research Resource Identifier (RRID): SCR 025389

## II. How we help

The NBCC provides full support for your imaging needs. We work with you on experimental design, guide you to the appropriate instrument, provide training and assisted use on our scopes, and help with image and data analysis.

# III. Applications

# A. Super-Resolution Imaging



Super-Resolution imaging goes beyond the diffraction limit of light to provide more detailed analysis of sub-cellular structures. We offer three different super-resolution options. The SoRa and DeepSIM systems provide up to 100 nm resolution with standard fluorophores in fixed or live samples. The STEDYCON system can achieve 30 nm resolution for the ultimate detail.

SoRa example

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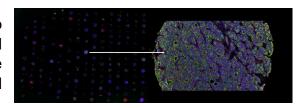


## B. Dynamic Live-Cell Imaging

Our widefield DeltaVision system is ideal for standard imaging or live-cell imaging using fluorescently tagged molecules or live cell compatible dyes. Temperature, humidity and CO<sub>2</sub> control are available for long-term live cell imaging to reveal molecule dynamics.

### C. Whole-Slide Scanning

Our Zeiss Axio Z1 slide scanner will allow up to 100 slides to be imaged with either a Brightfield or Fluorescent camera, allowing the whole slide to be digitized at either 20x or 40x, giving a full picture of the entire specimen.



#### D. Widefield and Confocal Imaging

We also offer a wide range of widefield and confocal microscopes to meet your imaging needs. All our instruments are detailed under Core Instrumentation.

## E. Image Analysis

We offer the use of Visiopharm, NIS Elements, Volocity, CellProfiler, and ImageJ software. We provide training, advice and support for all your image analysis needs.

## IV. Core Instrumentation

#### A. Super-Resolution Instruments

**Nikon CSU-SoRA:** The Nikon CSU-SoRa is a spinning disk microscope that uses optical reassignment to achieve super-resolution. It offers super resolution (~100 nm) of live or fixed samples with good fluorescence intensity. Its field of view is on the scale of a single mammalian cell, and it can accommodate standard secondary fluorophores (e.g., AlexaFluor), live dyes and fluorescent proteins. The objectives range from 10x air to 100x oil and excitation wavelengths extend from 405 nm (DAPI) to 647 nm (Cy5).

**Nikon CREST v3 DeepSIM:** The Nikon CREST v3 DeepSIM is a spinning disk microscope using structured illumination. It offers super resolution (~ 100 nm) of live or fixed samples with a field of view that can accommodate multiple mammalian cells. Since it is less phototoxic than the SoRa, it is ideal for longer term super resolution live imaging. This scope can accommodate standard secondary fluorophores (e.g., AlexaFluor), live dyes and fluorescent proteins. The objectives range from 10x air to 100x oil and excitation wavelengths extend from 405 nm (DAPI) to 730 nm (Cy7).

**Abberior STEDYCON:** The Abberior STEDYCON is a super resolution microscope using stimulated emission depletion. It offers super resolution ( $\sim$ 30 nm) of fixed samples with a field of view up to 90 x 80  $\mu$ m. This system works best with Abberior STAR dyes. Outfitted

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with a 100x 1.49 NA oil objective and 405, 488, 561 and 640 nm lasers. STED depletion laser of 775 nm permits super-resolution using red and far-red fluorophores.

#### B. Confocal Instruments

**Nikon A1:** The Nikon A1 is a point scanning confocal with 4x to 63x objectives, an adjustable pinhole, and 4 laser (405, 488, 561, 647 nm) illumination.

**Leica Quorom:** The Leica Quorom is a spinning disc confocal with 5x air to 100x oil objectives, a fixed pinhole, and illumination by 100 W halogen, EXFO X-cite 120 or 4 lasers (405, 488, 561, 647 nm)

**Andor BC43:** The Andor BC43 is a spinning disc and widefield confocal with 2x air to 63x oil objectives, a fixed pinhole, and 4 laser (405, 488, 561, 647 nm) illumination.

#### C. Widefield Instruments

**Deltavision Elite:** the Deltavision Elite is a widefield instrument that achieves 200 nm resolution with deconvolution. The instrument has a fully enclosed stage that controls temperature, CO<sub>2</sub>, and humidity for long-term imaging. It has an inverted microscope, 4x air to 100x oil objectives, solid state illumination and 390 nm (DAPI) to 632 (Cy5) filters.

**Olympus BX61:** We have two BX61 widefield instruments. One that is capable of histology only and one that can do both histology and fluorescence.

**Olympus IX81 Inverted:** The IX81 is a widefield instrument with structured illumination and deconvolution.

**DMIL LED:** The DMIL LED is a basic histology instrument with widefield fluorescence.

#### D. Slide Scanner

**Zeiss AxioScan:** The Zeiss AxioScan enables both brightfield and fluorescent scanning. The lenses range from 5x to 40X with 5 different filters (DAPI, FITC, Cy3, Cy5 and Aqua).

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