CNR National Research Council
Institute of Molecular Biology and Pathology

Nikon Microscopy Reference Center for Central-Southern Italy

http://pegaso.ibpm.cnr.it/cge/microscopy/microscopy.html
@Sapienza University - San Lorenzo site

OVERVIEW

In the last decade an impressive development in bioimaging methods has taken place and has transformed microscopy from a descriptive approach into a quantitative, high-content experimental tool. Gaining accurate, high-resolution spatio-temporal information has proven to be key to understand complex biological processes. High-resolution imaging makes it possible to directly correlate specific molecular events with global changes at the single cell level and to visualize stochastic events and cell-to-cell variability that could not be appreciated otherwise. Imaging applications such as time-lapse recording add a new layer of dynamic information that is essential to understand such processes as cell differentiation, intracellular signalling, cell-to-cell communication, migration, invasion, division and others.

The IBPM - Nikon Microscopy Center hosts last-generation equipment with complementary imaging capabilities and applications, including:

- time-lapse recording of live cells over several days,
- detecting intracellular molecular interactions,
- reconstructing cellular and subcellular structures at the 3D level,
- obtaining quantitative information on the molecules or cells that are being imaged.

DATABASE

The image database contains an initial collection of individual entries that support both automatic and manual annotations for image processing and analysis based on Nikon proprietary software
http://www.nis-elements.com/
The database is permanently updated to expand the range and accuracy of image analysis ("machine learning" principle).

WHERE WE ARE

IBPM Laboratories - via degli Apuli 4 - Floor 2
c/o Department of Biology and Biotechnologies
"Charles Darwin", Sapienza University - San Lorenzo compound

CONTACTS

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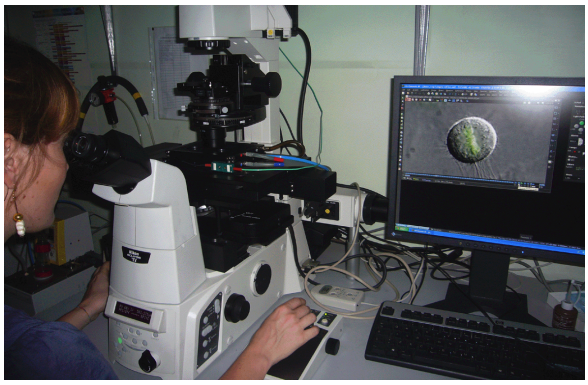
IMAGING APPLICATIONS

The IBPM - Nikon microscopy platform supports versatile applications of fluorescence microscopy for both fixed and live cell samples and provides opportunities for coupling dynamic studies (time-lapse recording) with high resolution qualitative and quantitative image analysis of cells and cellular structures.
Applications and scopes include:
- Studies of dynamic processes in normal and cancer cells, e,g. signalling, intracellular transport, organization of organelles and subcellular structures;
- Cell division and its associated checkpoints;
- Inhibition of tumor cell growth;
- Differentiation of stem and progenitor cells;
- Validation of innovative drugs;
- The cell response to parasites, bacteria and viral infectious agents and their restriction by antibiotics, antibodies or other conditions;
- The induction of different forms of cell death in response to different stimuli, damage and stress conditions;
- The development of biocompatible matrices to support cell growth in studies of tissue regeneration;
- The interaction of nanoparticles with cells in toxicity and therapeutic studies;
- The interaction between different proteins and molecules at the intracellular site(s) in which they take place.

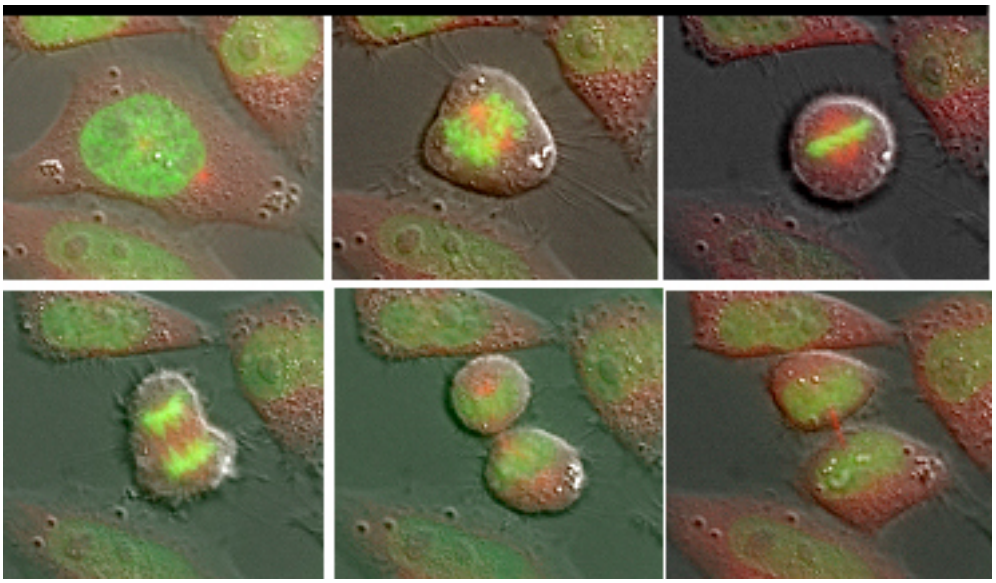
TIME-LAPSE IMAGING OF LIVING CELLS

Nikon Eclipse Ti inverted microscope
www.nikoninstruments.com/en_IT/Products/Microscope-Systems/Inverted-Microscopes/Biological/Eclipse-Ti

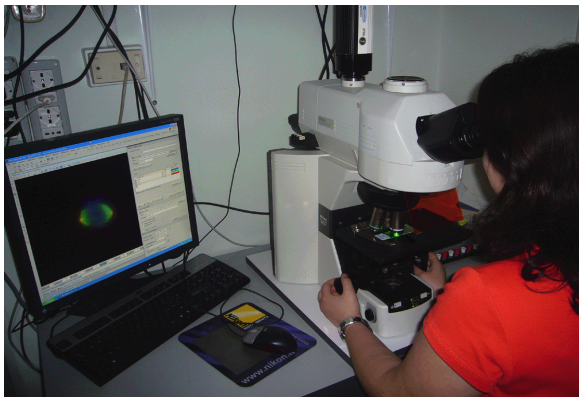


Options:
- automated setting of intervals and duration of recording over days, with automated fluorescence light shutter and joystick;
- multipoint acquisition of fluorescent, DIC and phase contrast images;
- perfect focus device for automatic reset over long recording times;
- OKO-lab stage incubator with controlled T° and CO2 conditions to preserve the sample viability over days.

Video-recording of a human cell during mitotic progression
Chromosomes (H2B-GFP): green; mitotic apparatus (tubulin-mCherry): red



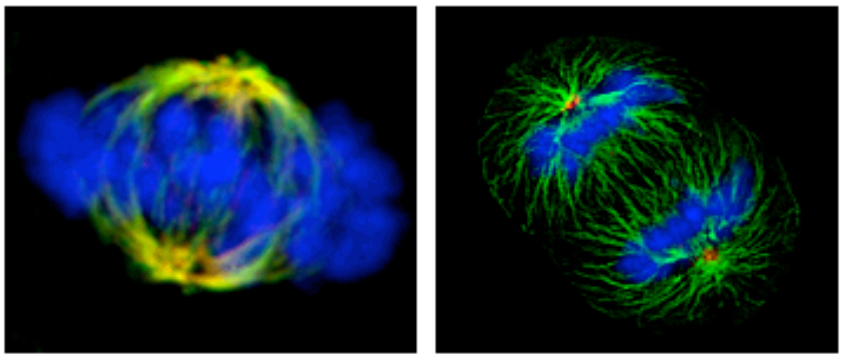
HIGH-RESOLUTION IMAGING OF FIXED CELL SAMPLES



Nikon Eclipse 90i microscope

www.nikoninstruments.com/en_IT/Products/Microscope-Systems/Upright-Microscopes/Biological/Eclipse-90i

Fully integrated high-resolution upright fluorescence microscope with digital imaging head, automated z-axis for 3D image deconvolution and reconstruction, image acquisition in three fluorescence channels and in wide-field, delivering crisp and clean images of fixed cell samples.



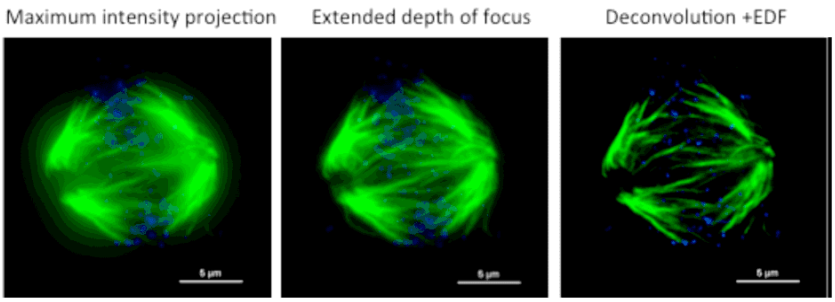
An "Intensilight" illumination source with internal filters allows for modulatable intensity for optimal imaging of fluorescence-stained specimens.

Imaging workstation

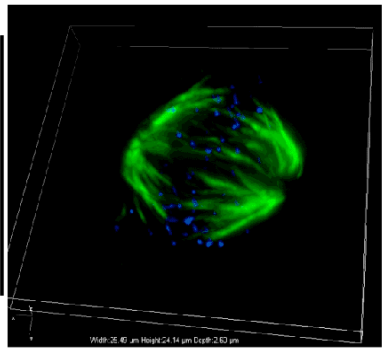
ttp://www.nis-elements.com/

Equipped with Nis-Elements AR 3.2 software (Nikon) for qualitative (deconvolution, 3D-projection, Extended Depth Focus, LUTs, etc) and quantitative image processing.

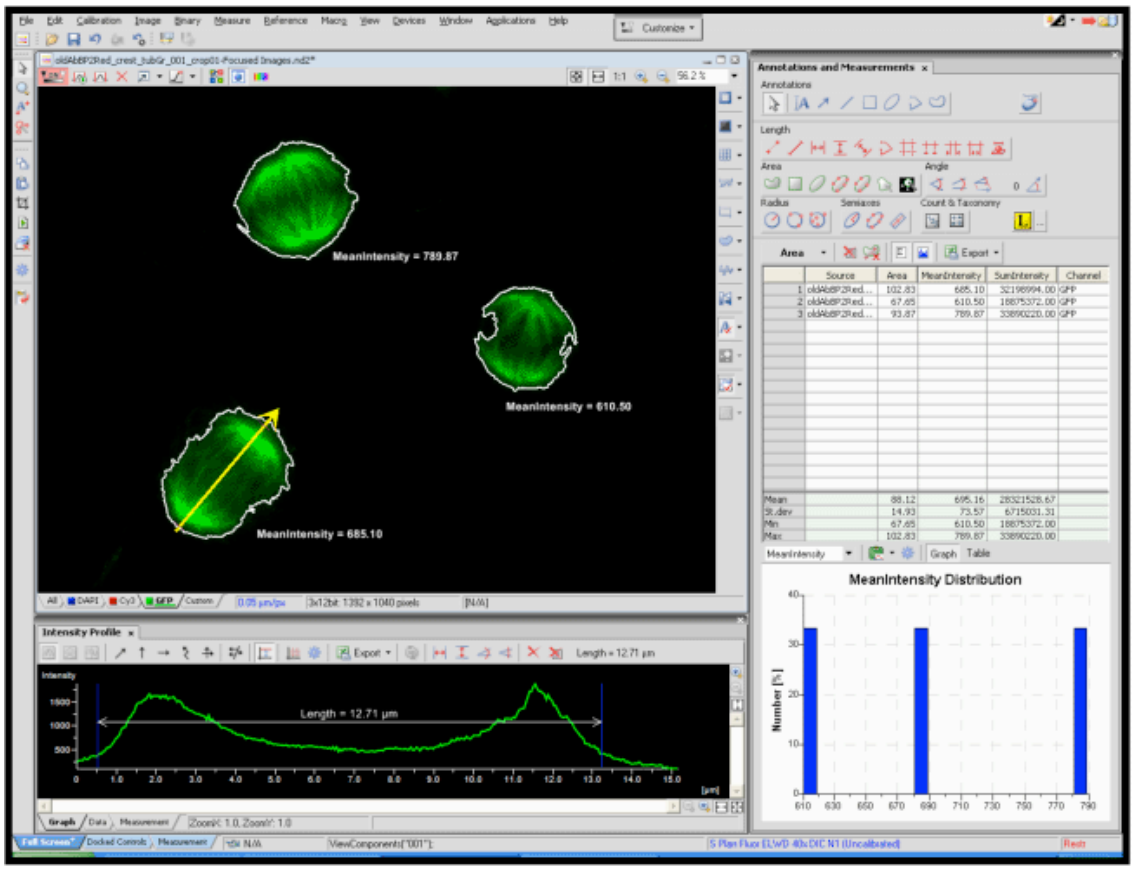
2D processing of z-stacks

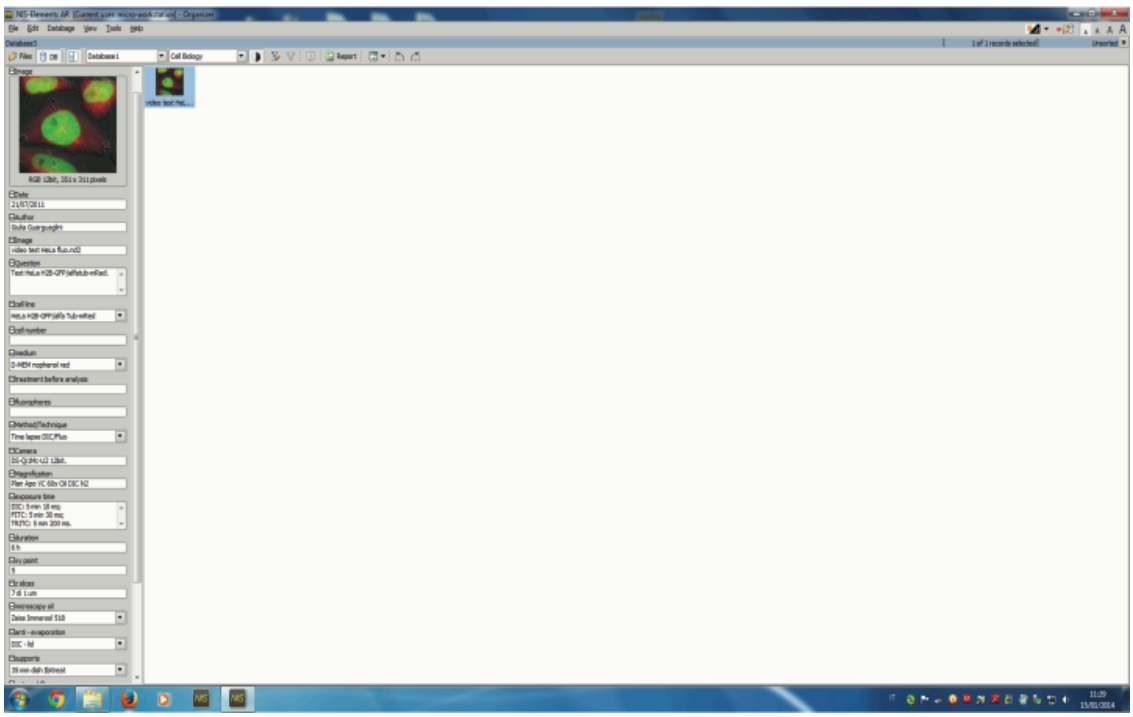


3D projection



Quantitative image analysis



Database for automatic data recording and phenotype collection / analysis with both standard reference and manual annotation modes.

An example of a database entry enabling both automatic and manual annotation-based classifications