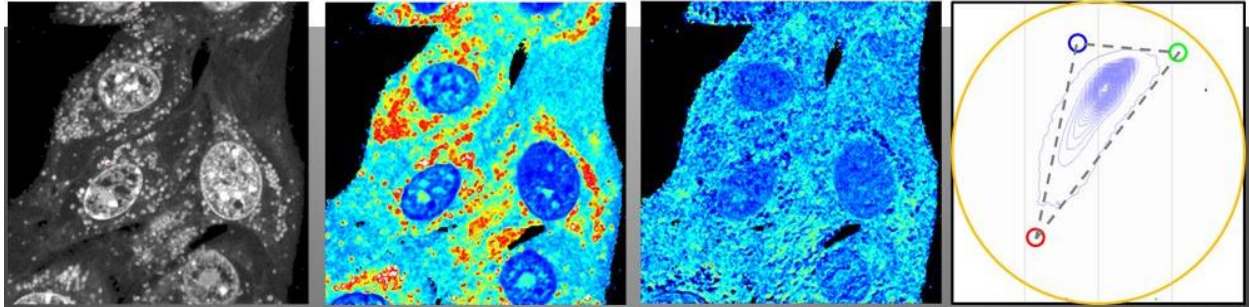


(Preliminary agenda)

2nd Annual Workshop on Advanced Microscopy and Biophotonics

November 23-27 of 2020

Institut Pasteur of Montevideo/Hospital de Clínicas



Organizer: **Dr. Leonel Malacrida**

International speakers:

- Dr. Enrico Gratton (UCI-USA)**
- Dr. David Jameson (U. Hawaii-USA)**
- Dr. Susana Sanchez (U. Concepción-Chile)**
- Dr. Per Niklas Hedde (UCI-USA)**
- Dr. Suman Ranjit (Georgetown U-USA)**
- Dr. Luca Lanzaó (U. Catania)**
- Dr. Luis Bagatolli (I. Ferreyra-Argentina)**
- Dr. Michelle Digman (UCI-USA)**
- Dr. Valeria Levi (UBA-Argentina)**
- Dr. Francesco Cardarelli (U. Pisa-Italy)**
- Dr. German Gunther (U. Chile-Chile)**
- Dr. Laura Estrada (UBA-Argentina)**
- Dr. Belén Torrado (UCI-USA)**
- Dr. Lorenzo Scipioni (UCI-USA)**
- Dr. Alexander Vallmitjana (UCI-USA)**
- Dr. Francesco Palomba (UCI).**

Local speakers:

- Dr. Federico Lecumberry (FIng-UdelaR)**
- Dr. Florencia Irigoín (FMed-UdelaR)**
- Dr. Ariel Fernandez (FIng-UdelaR).**

Course target and goals:

The workshop in Advanced Microscopy and Biophotonics is dedicated to disseminate cutting-edge technology and advanced methods in fluorescence microscopy and biophotonics. The target audience includes postgraduate students, postdocs, and academics from all Americas with a background in the fundamentals of fluorescence microscopy, seeking to learn about recent advances in the combination of spectroscopy with fluorescence microscopy. In the course, we will cover the fundamentals of advanced instrumentation and methods, such as FCS, FLIM, Hyperspectral imaging, Super-resolution, Non-linear microscopy, Deep tissue imaging, light-sheet microscopy, among others. Due to the global COVID outbreak, this year the workshop will be held as a virtual course. The course is organized with lectures of fundamentals (40+5 min), application talks (20+5 min), and an optional advanced section for a virtual practical section (two section 2hs/each) and e-poster. This last section is restricted to 20 students and will be given on the last day. The lectures will be given in English.

Workshop agenda:

Monday November 23

09:45-10:00 – Opening and course introduction. **Leonel Malacrida**

10:00-10:45 – **Lecture 1. David Jameson.** Introduction to fluorescence fundamentals and methods.

10:45-11:30 – **Lecture 2. Leonel Malacrida:** Advanced fluorescence instrumentation and microscopy.

11:30-12:15 – **Lecture 3. Enrico Gratton:** Introduction to fluorescence correlation spectroscopy (FCS) and the photon counting histogram (PCH)

Virtual lunch break and meet with the instructors

(Using zoom rooms I would like to organize small groups with the speakers from the previous sections for more informal and detailed discussion).

13:30-14:15 – **Lecture 4. Per Niklas Hedde:** Raster image correlation spectroscopy (RICS) and *in vivo* applications.

14:15 – 15:00 - **Lecture 5. Per Niklas Hedde:** Number & Brightness (N&B) and its applications for protein oligomerization studies *in vivo*.

15:00-15:45 – **Lecture 6. Enrico Gratton:** The pair-correlation function and its applications for deciphering the occurrence of barriers and obstacles inside the cell.

Virtual coffee break and meet with the instructors

16:15 – 16:40 – **Application talk #1. Susana Sánchez:** Ribosomal proteins detection by the PCH analysis in yeast.

16:40 – 17:05 – **Application talk #2. Laura Estrada:** Deciphering the Dengue virus shuttling in the cell by 2D-pCF.

17:05 – 17:30 – **Application talk #3. Per Niklas Hedde:** Applications of cross-RICS and cross-N&B for protein dynamics within the in vivo cell.

Tuesday November 24

10:00-10:45 – **Lecture 7. David Jameson:** Introduction to lifetime measurements and the phasor plots.

10:45-11:30 – **Lecture 8. Enrico Gratton:** The phasor plots for FLIM and FRET.

11:30-12:15 – **Lecture 9. Leonel Malacrida:** The spectral phasor plot and its applications to hyperspectral imaging.

Virtual lunch break and meet with the instructors

13:30-14:15 – **Lecture 10. Alex Vallmitjana:** Blind four components analysis on FLIM data by the phasor approach.

14:15-15:00 – **Lecture 11. Leonel Malacrida:** Cell metabolism phenotyping and absolute NADH concentration by FLIM-Phasors.

15:00-15:45 – **Lecture 12. Per Niklas Hedde.** Hyperspectral imaging in light-sheet microscope without hyperspectral detection.

Virtual coffee break and meet with the instructors

16:15 – 16:40 – **Application talk #4. Susana Sánchez:** Applications of FLIM-Phasors for biological questions: the pH and Ca⁺⁺ case.

16:40 – 17:05 – **Application talk #5. Suman Ranjit:** FLIM-Phasor for cell metabolism phenotyping.

17:05 – 17:30 - **Application talk #6. Leonel Malacrida:** FLIM and Hyperspectral imaging using phasors to study membrane dynamics by LAURDAN fluorescence.

Wednesday November 25

10:00-10:45 – **Lecture 13. Luca Lanzanó:** Fundamentals of Super-resolution microscopy and its combination with FLIM-Phasors.

10:45-11:30 – **Lecture 14. Lorenzo Scipioni.** The Vespa Microscope for multiplexing microscopy

11:30-12:15 – **Lecture 15. Alexander Vallmitjana:** SR-FLIM opportunities for *in vivo* cellular dynamics measurements

Virtual coffee break and meet with the instructors

13:30-14:15 – **Lecture 16. Enrico Gratton:** The DIVER microscope for deep tissue imaging.

14:15-15:00 – **Lecture 17. Leonel Malacrida:** The single plane illumination microscopy and its applications.

15:00-15:45 – **Lecture 18. Alexander Vallmitjana:** 3D-Single particle tracking and its applications.

Virtual lunch break and meet with the instructors

16:15 – 16:40 – **Application talk #7 Enrico Gratton:** iMSD application for molecular intracellular diffusion studies.

16:40 – 17:05 – **Application talk #8. Lorenzo Scipioni:** Comprehensive correlation analysis for super-resolution dynamic fingerprinting of cellular compartments using the Zeiss Airyscan detector.

17:05 – 17:30 - **Application talk #9. Suman Ranjit:** Applications of the DIVER microscope for biomedical imaging.

Thursday November 26

10:00 – 10:25 – **Application talk #10 Valeria Levi:** Applications of FCS to study the organization of GR in the nucleus.

10:25 – 10:50 – **Application talk #11. Luis Bagatolli:** Uses of solvatochromic probes for water dynamics in yeast.

10:50 – 11:15 - **Application talk #12. Germán Gunther:** Development and application of CAPRYDAA for *in vivo* membrane dynamics.

11:15 – 11:40 – **Application talk #13 Federico Lecumberry:** Applications of imaging processing and AI in advanced fluorescence microscopy.

11:40-12:05 – **Application talk #14. Florencia Irigoín.** The primary cillium membrane organization studied by spectral phasors and LAURDAN.

Virtual lunch break and meet with the instructors

13:30 – 13:55 – **Application talk #15. Per Niklas Hedde:** Fluorescence lifetime detection with particle counting devices and its applications.

13:55 – 14:20 - **Application talk #16. Belen Torrado:** Spectral detection with up/down filters.

14:20-14:45 – **Application talk #17. Ariel Fernandez:** Snapshot polarimetric and phase quantitative microscopy.

14:45-15:10 – **Application talk #18. Michelle Digman:** Evaluation of pre-implantation embryos by FLIM and autofluorescence.

Virtual coffee break and meet with the instructors

15:40 – 16:40 - **Lecture 19: Workshop close and special lecture. David Jameson:** A nano-history of fluorescence.

16:40 – 17:30 – Workshop closing and virtual coffee break with instructors

Friday November 27

10:00 -12:00 e-Poster section 5min/student (20 total presentation)

Virtual lunch break and meet with the instructors

13:30 – 15:30 – Virtual practical section:

Two topics (total 10 students by subgroup)

- **RICS and N&B** (Leonel Malacrida & Nik Hedde)
- **FLIM-Phasor** (Belén Torrado, Alex Vallmitjana and Francesco Palomba)

Virtual coffee break and meet with the instructors

16:00-17:30 – Final discussion