

## CONFOCAL MICROSCOPY CORE UNIT

Diego Megías  
Core Unit Head

Technicians  
Jesús Gómez (PEJ-L)\*, Manuel Pérez (TS)\*\*, Joaquim Soriano (TS)\*\*

\*Plan de Empleo Joven-Licenciado (Youth Employment Plan-Graduate)

\*\*Titulado Superior (Advanced Degree)



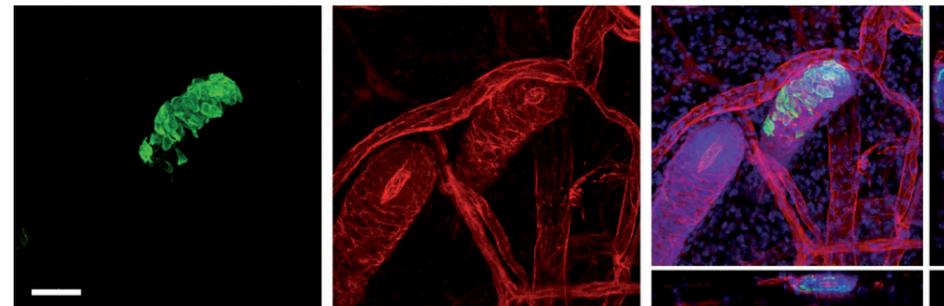
### OVERVIEW

Optical microscopy has traditionally been an indispensable tool in cell biology studies. In fact, one of the main challenges in oncology research is the study of specific markers, expression patterns or individual cells in the tumour environment.

The Confocal Microscopy Unit provides CNIO Research Groups with all the standard methodologies and the latest advances in microscopy. We offer access to state-of-the-art equipment and software packages related to confocal microscopy, including technical and scientific advice and support to the CNIO scientists. The Unit is also actively involved in developing, testing and implementing new microscopy technologies, tools and imaging applications that could be of interest to the Research Groups. Training activities are also an essential component of our mission.

**“The Confocal Microscopy Unit is fully committed to disseminating advanced microscopy methodologies that are useful for cancer research in order to benefit society, always with the aim of increasing our understanding of the cell biology and the disorders of cells that cause cancer.”**

### RESEARCH HIGHLIGHTS



**Figure** Whole-mount of ear hair follicles. A global double-fluorescent reporter mouse (GFP/Tomato) was used for lineage tracing of epidermal cells.

The Confocal Microscopy Unit is equipped with 3 laser scanning confocal systems (Leica SP2 and SP5) that incorporate UV and multiphoton excitation, a white light laser and a Hybrid Detector, as well as 2 wide-field systems (a Deltavision 4D deconvolution station and a Leica DMRI6000 system, equipped with microinjection). All the microscopes are automated and equipped with incubators for live cell imaging.

In addition, the Unit has implemented the use of high-throughput technologies applied to confocal microscopy using 2 different systems:

- An Opera (Perkin Elmer) High Content Screening (HCS) system, which allows running HCS experiments on fixed and live cells in multi-well plates, and enables the monitoring of cell dynamics (translocation, cell division, etc.) through the use of fluorescence.
- A Matrix Screening Application integrated into the SP5 confocal systems, allowing high-throughput feeding of the instrument, not only in multi-well plates, but also in tissue sections.

These advances enable us to increase the level of information obtained from a sample as well as carry out the automated screening of cell behaviour under different treatments.

During 2016, the Confocal Microscopy Unit contributed to the microscopy field in several aspects. It improved the intelligent screening technique with new algorithms for image acquisition, thereby creating new applications in both confocal and conventional fluorescence microscopy. The use of microfluidics with live-cell assays in perfusion chambers has also experienced a significant increase in performance and demand. In addition, the Unit patented a new device for improving hardware autofocus that will be of great relevance in high-resolution automated image acquisition. Moreover, the Confocal Microscopy Unit continues to dedicate a significant effort towards the development and implantation of High-Content Screening technology at the CNIO; for example, in 2016, we provided support for the running of screening assays for compounds that could modify mitotic checkpoints, integrity of nucleoli, DNA Damage, BrdU, cell proliferation, etc.

Last but not least, in the field of intravital microscopy, we already have several ongoing projects that are focused on metastasis and skin alteration studies. ■

#### ► PUBLICATIONS

- Burén S, Gomes AL, Teijeiro A, Fawal MA, Yilmaz M, Tummala KS, Perez M, Rodriguez-Justo M, Campos-Olivas R, Megías D, Djouder N (2016). Regulation of OGT by URI in Response to Glucose Confers c-MYC-Dependent Survival Mechanisms. *Cancer Cell* 30, 290-307.
- Pérez-Guijarro E, Karras P, Cifdaloz M, Martínez-Herranz R, Cañón E, Graña O, Horcajada-Reales C, Alonso-Curbelo D, Calvo TG, Gómez-López G, Bellora N,

Riveiro-Falkenbach E, Ortiz-Romero PL, Rodríguez-Peralto JL, Maestre L, Roncador G, de Agustín Asensio JC, Goding CR, Eyras E, Megías D, Méndez R, Soengas MS (2016). Lineage-specific roles of the cytoplasmic polyadenylation factor CPEB4 in the regulation of melanoma drivers. *Nat Commun* 7, 13418.

- Andradás C, Blasco-Benito S, Castillo-Lliva S, Dillenburg-Pilla P, Díez-Alarcía R, Juanes-García A, García-Taboada E, Hernández-Llorente R, Soriano J, Hamann S, Weners A, Alkatout I, Klapper W, Rock-

en C, Bauer M, Arnold N, Quintanilla M, Megías D, Vicente-Manzanares M, Urigüen L, Gutkind JS, Guzmán M, Pérez-Gómez E, Sánchez C. (2016). Activation of the orphan receptor GPR55 by lysophosphatidylinositol promotes metastasis in triple-negative breast cancer. *Oncotarget* 7, 47565-47575.

- Ramírez-Santiago G, Robles-Valero J, Morlino G, Cruz-Adalia A, Pérez-Martínez M, Zaldivar A, Torres-Torresano M, Chichón FJ, Sorrentino A, Pereiro E, Carrascosa JL, Megías D, Sorzano CO, Sánchez-Madrid

F, Veiga EB (2016). Clathrin regulates lymphocyte migration by driving actin accumulation at the cellular leading edge. *Eur J Immunol* 46, 2376-2387.

#### ► PATENT

- Postigo P.A and Megías Vázquez D. (2016). *Uso de un material para la fabricación de un cubreobjetos, un portamuestras o un recipiente de cultivo celular*. Spanish Patent Application ES201631277.