CONFOCAL MICROSCOPY CORE UNIT

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Titulado Superior (Advanced Degree)

OVERVIEW

One of the main challenges in oncology research is the study of specific markers, expression patterns, or individual cells in the tumour environment. Optical microscopy has traditionally been an indispensable tool in cell biology studies and has become essential for understanding cancer biology.

The Confocal Microscopy Unit (CMU) provides the CNIO research groups with the latest advances in optical microscopy, offering access to state-of-the-art equipment and image analysis software, including scientific advice and technical support. The Unit is also actively involved in developing and implementing new advanced imaging methods that could have an impact on the work of CNIO research groups. Advanced microscopy training and science disseminating activities are also an essential component of our mission. We organise courses, talks and visits, always with the aim of increasing our understanding of the cellular and molecular disorders that lead to cancer and the study of potential treatments.

"The CMU is committed to providing a deep understanding of the molecular mechanisms involved in tumour progression and treatment responses by applying advanced microscopy methods."

RESEARCH HIGHLIGHTS

The Confocal Microscopy Unit remains dedicated to providing imaging solutions to support research across the CNIO. The team plays a pivotal role in exploring various scientific questions, contributing with valuable insights to a range of projects.

In 2023, the Unit focused on enhancing tissue analysis by implementing immunofluorescence multiplex techniques for simultaneous visualisation and analysis of multiple markers. Together with the Histopathology Unit, the Unit has optimised an automate and reliable multiplex staining protocol for immune panels of 6 cellular markers. Semi-automated acquisition of whole tissue sections was also implemented using the Thunder widefield system to enable spatial analysis and understanding of tissue complexity. These images can be also imported and aligned in the SP8 confocal system for the automated acquisition of highly resolved tissue information. Furthermore, the Unit also developed advanced image analysis algorithms to extract quantitative and spatial information from complex immunofluorescence multiplex images, using deep learning tools for automated cell segmentation, classification, and feature extraction, reducing analysis time, and improving accuracy. Altogether, the Unit's efforts have contributed to improving the CNIO immunofluorescence multiplex tissue analysis platform, facilitating efficient analysis of diverse cellular markers simultaneously in different tissue samples.

To boost the screening capacity at the CNIO, the Unit started to develop Cell Painting high-content methods for cellular morphological profiling. Up to 6 fluorescence dyes are used to label different compartments of the cell to form their unique phenotypic profile by extracting features with automated image analysis. Biological perturbation upon drug treatments can be described by changes in the phenotypic profile providing the possibility to screen for unknown targets.

In June, Ana Cayuela joined the Confocal Microscopy Unit as a bioimage analyst, bringing signinficant experience in implementing image analysis tools, including deep learning for subtracting quantitative information from optical microscopy images. ■

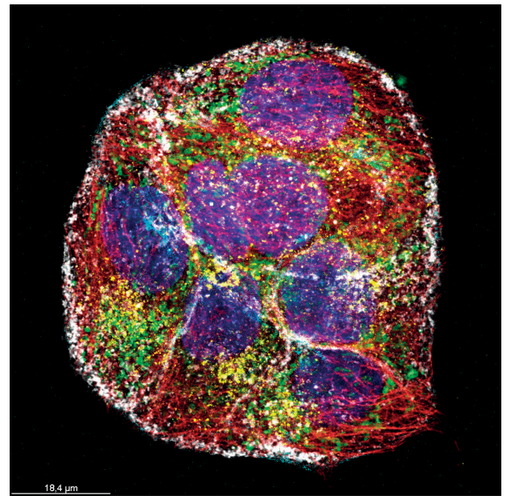

FIGURE 1 Developing immunofluorescence multiplex workflows. Cell Painting image. Nuclei are in blue, microtubules are in red, cell membrane is in white, actin is blue, and lysosomes are in yellow. Image courtesy of Manuel Pérez, Confocal Microscopy Unit.

PUBLICATIONS

- Costa-Machado LF, Garcia-Dominguez E, McIntyre RL, Lopez-Aceituno JL, Ballesteros-Gonzalez Á, Tapia-Gonzalez A, Fabregat-Safont D, Eisenberg T, Gomez J, Plaza A, Sierra-Ramirez A, Perez M, Villanueva-Bermejo D, Fornari T, Loza MI, Herradon G, Hofer SJ, Magnes C, Madeo F, Duerr JS, Pozo OJ, Galindo MI, Del Pino I, Houtkooper RH, Megias D, Viña J, Gomez-Cabrera MC, Fernandez-Marcos PJ (2023). Peripheral modulation of antidepressant targets MAO-B and GABAAR by harmol induces mitohormesis and delays aging in preclinical models. *Nat Commun* 14, 2779.
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