

The UPAT's upgraded Confocal Microscopy Facility, is up to our knowledge unique in Greece for its capacities, properties and applications. This set up will allow the integration of the information gained on drug-target structures following their functional characterization (by the SEE-PROT & SEE-STRUCT modules). Insights from structural determination will be validated in live cells, to allow an understanding of how the biomolecule tested affects basic cellular pathways at the molecular levels. In addition, novel bioactive molecules will be evaluated for their efficacy in modulating protein target function and its signaling pathways.

The upgrade of the confocal microscope facility will be complemented by accompanying facilities, already functional in the Medical School of Patras for culturing mammalian cells including tumorigenic cell line and mouse and human stem cells. In addition the Medical School has a fully equipped animal house for animal studies.

The proposed upgrade of the confocal microscope facility will permit the following methodologies, to be provided to users for the "in cell" characterization of protein function and testing mutated proteins and lead compounds.

- Cell-based assays for lead compound validation and assessing effects of mutants. Viability, apoptosis, proliferation, differentiation, and senescence assays in cultured mammalian cells, tumor cell lines and neural stem cells.
- FRET measurements by acceptor photobleaching and sensitized emission for assessing protein-protein and protein-compound interactions and protein modification in live cells. Comparative assessment of wild-type and mutant proteins and the effects of the presence of compounds/peptides.
- Fluorescence Recovery after Photobleaching (FRAP) experiments for the determination of dynamic protein interactions within cells
- Sub-cellular ablations coupled to time-lapse microscopy for the analysis of dynamic cellular responses to damage.

This tool complements the existing UPAT instrumentation for assessing protein function and determining protein-protein interactions and modifications in cells and for validation of lead compounds-peptides by in-cell dynamic measurements of protein-protein and protein-compound interactions. The effects on protein function of targeted mutations guided by structural information (SEE-STRUCT) will be assessed in vivo, while bioactive compounds and interacting peptides characterized structurally either by NMR or by X-ray will be tested in cells. Current UPAT staff has extensive experience with Molecular Cell Biology, functional imaging methodologies and cell based assays and will support the running of the facility.

Confocal in cell imaging details

Written by Administrator

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Access to this infrastructure will be provided to a wide variety of Greek labs, both academic and non-academic, in Patras, in Greece and the South-East Region. The Confocal Microscopy Node has been successfully operating in UPAT since 2007 and is in high-demand by a large number of teams in UPAT and Greece (University of Ioannina and Athens).

The facility will be in close contact with Advanced Light Imaging Facilities at the European Level. UPAT teams have a long-standing collaboration with Centers of Excellence in Advanced Light Imaging in Europe, including the EMBL in Heidelberg, the Max Planck in Dortmund and the NIMR in London.