HiTs@UNIPD

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The Imaging Facility of the Department of Biology (DeBio) hosts the High Content Imaging System (HCI) Operetta (Perkin Elmer).
This highly flexible instrument allows to perform both low and high throughput studies based on fluorescent assays.

The Hits Facility @Unipd includes:

- ✓ The operetta High Content Imaging System equipped with a JANUS Liquid Handler Workstations, a Plate handler, Cell incubator, Operetta;
- ✓ Two Harmony software licences;
- ✓ Precision XS pipetting system (Biotek) for sample preparation;
- ✓ Qiacube for automated Miniprep preparation;
- ✓ chemical or genomic libraries;
- ✓ assistance in HT image acquisition.

Our High content Imaging Facility is involved in screenings and in small-scale research projects that could benefit from the use of the available equipment and expertise in automation, assay development and data acquisition.

The versatility of Operetta makes it an ideal instrument for analysis of different experimental models, from cell monolayers to complex biological systems (3D-acquisition), as shown below.

Low Throughput approaches:

Migration, Cell Cycle, Differentiation, Colony Formation

Nuclear size, shape, granularity

Organelles morphology

Cell death

Cell shape, neurite outgrowth

Quantification of cytosolic markers

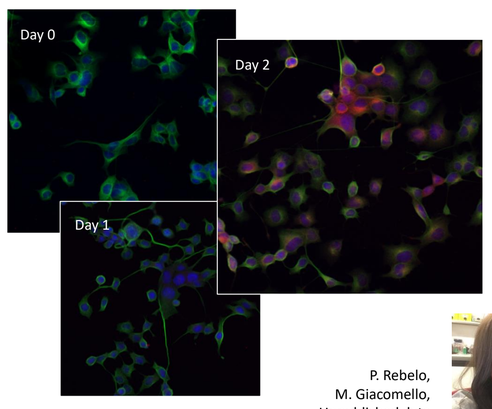
Plasma membrane translocation

Mitochondria membrane potential in muscle fibers (tibialis)

Isolation of fibers from muscle
20nM TMRM for 20 min

C. Bean, F.Chemello, M. Giacomello, Unpublished data

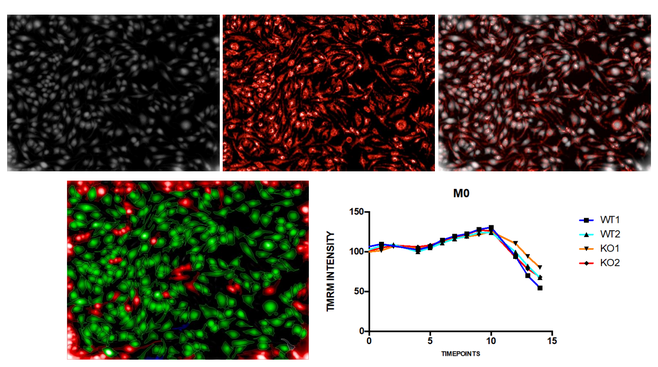
NSC34 differentiation to motor neurons-like cells.



Tubulin III, green; MAP2, red; Hoechst, blue.

P. Rebelo, M. Giacomello, Unpublished data

Pharmacological Profiling



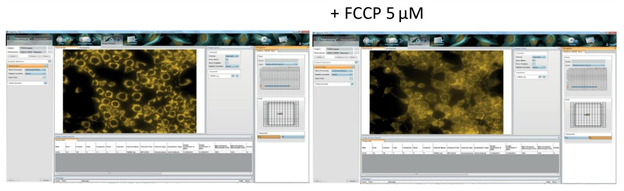
Bone-marrow derived murine macrophages. Loaded with TMRM.

T. Scolaro, F. Munari; Prof. A. Viola, Unpublished data

Mitochondria membrane potential

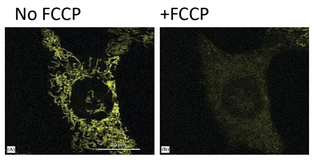
Operetta

+ FCCP 5 µM



Confocal Microscope

No FCCP +FCCP



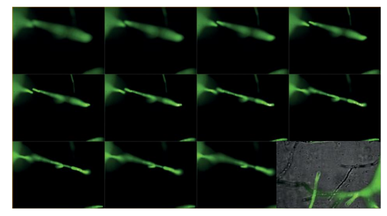
Fibroblasts from patients: seeded 24 hours before analysis, 3000 cells/well, 20nM TMRM for 30 min

R. Quitana Cabrera, M. Giacomello, E. Schon et al

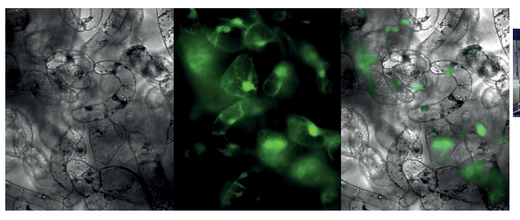
Plant Biology

Physcomitrella + YC3.6

A. Alboresi, Prof T. Morosinotto, Unpublished data



Grapes + Hyper



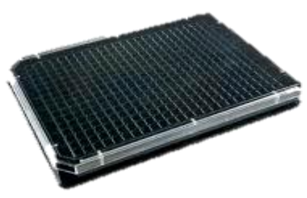
E. Barizza, M. Zottini, Unpublished data

High Throughput approaches: an example

A high content imaging screen for mitochondria-Endoplasmic Reticulum contact sites based on a FRET probe

Day 1

seed cells on 384 well plates (automated pipetting system, Precision XS, Biotek)

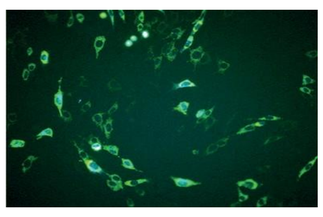


Day 2

shRNA transduction (Mission Library, SIGMA, 10000 genes)

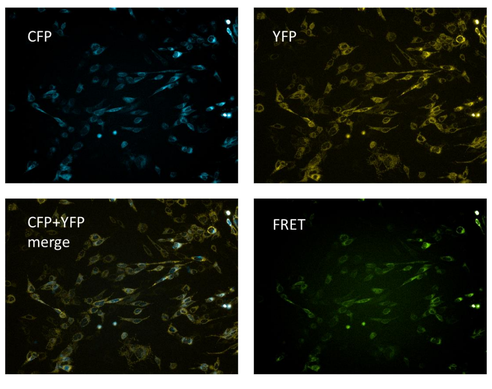
Day 3

Transfection of the FRET Probe

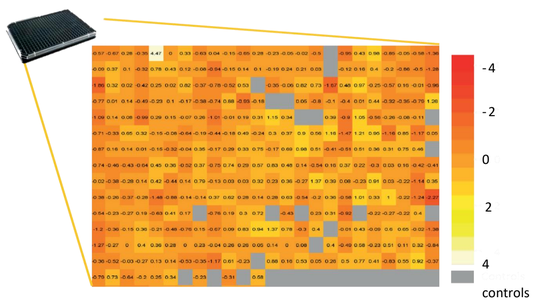


Day 4

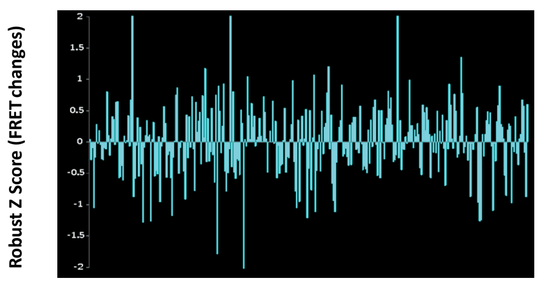
Image Acquisition



Robust Z Score (FRET changes)



12,000 mouse genes



about 200 putative MERCs modulators
10 structural

The MERCS-HCS Team:

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Further information available at:
https://www.biologia.unipd.it/servizi/servizi-alla-ricerca/servizi-dipartimentali/imaging-facility/high-throughput-screening-facility/
Booking through the following website:
http://service.biologia.unipd.it/gestione_dipartimento/login.

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