MOLECULAR BIOSCREENING

Molecular BioScreening is the newest infrastructure from iMed.ULisboa that offers an innovative and integrated approach of cell-based medium- to high-throughput assays for screening small molecules (natural or synthetic) and biologics, ultimately leading to discovery of new therapeutics. It combines the power of relevant cell models, phenotypic screens and live cell functional assays to both model disorders and search for drugs to treat disease.

The initial steps in drug discovery include the selection of a target, development of an assay and screening against a compound library. This infrastructure provides a platform to identify molecules that modify the function of genes, cells or biochemical pathways to internal and external project partners, in both academia and industry.

After the initial screening and hit identification, the primary hits are optimized using multiple criteria including structure activity relationships, selectivity, physicochemical properties that result in a lead compound series. Selected compounds may undergo specific molecular testing, to evaluate biological targets and mechanisms of action, and later on pre-clinical pharmacokinetics and pharmacodynamics, toxicity and efficiency testing. The collaborative environment at iMed.ULisboa facilitate access to faculty with special expertise in a variety of related disciplines, including bioinformatics and advanced data analysis, as well as medicinal chemistry, biomolecular engineering, and chemical probe synthesis. This strategy will ideally lead also to the discovery of biomarkers and molecular targets.

Goals

- Support basic and translational biomedical research by designing, optimizing, and running screening assays that deliver answers to questions of scientific interest;
- Discover new medicines in a faster and more efficient fashion, promoting innovation in drug discovery, design, and development;

- Promote institutional cooperation strategies between academia and industry.
- Integrate regulatory science in early compound screening and development stages to diminish attrition success.

The Molecular BioScreening infrastructure provides cell-based assays including phenotypic assays where the underlying targets are unknown, using human and non-human cell lines, primary cells, stem cells and induced pluripotent stem cell-derived cells to recapitulate human biology.

Why choose a phenotypic screening approach?

In recent years, cell-based assays have emerged as a more physiological alternative for hit discovery. The phenotypic screening approach measures cellular properties in response to test compounds. Cell-based high-throughput screening often assumes no a priori knowledge of direct molecular targets and aims to identify modulators of a pathway in the more physiological environment of a cell, offering the advantage of using intact cells in their native environment. It enables lead discovery for many diseases in which a drug target has not been identified and/or validated, and allows to interrogate entire pathways of interest providing the opportunity for multiple potential intervention points.

Ongoing projects

The infrastructure operates mostly as a collaborative resource. In addition, as member of the Academic Drug Discovery Consortium (http://www.addconsortium.org/), iMed.ULisboa has established partnerships such as that with a renowned multinational pharma company to evaluate the therapeutic potential of a 250.000 compound library. There are also several ongoing projects with start-up companies to identify human and animal-derived recombinant antibodies in viral infection, cancer and inflammation.

Available primary screens

Primary screens are designed and optimized to deliver solutions that help achieve specific experimental goals. These include, among others:

Cytotoxicity / Cell Viability

Cellular viability is one of the most common phenotypic assays performed. Using cell lines and the MTS method, this assay evaluates compound cytotoxicity at several standardized concentrations allowing to characterize cytotoxicity profiles by establishing dose-response curves and calculating IC_{50} or EC_{50} values. Antibody-derived mechanisms of cytotoxicity are also available, such as ADCC and CDC, to discover or optimize antibody-based therapies.

This assay is available in 96-well format and it can be applied to both pure compounds and crude extracts.

Regulated cell death

Regulated cell death is characterized by biochemical and morphological features and is described as contributor to the pathogenesis of several diseases. Therefore, identifying modulators of forms of regulated cell death can prompt the development of lead compounds for therapeutic strategies.

This assay is optimized in 384-well format to evaluate compound libraries.

Inquiries regarding other specific assays are welcome from academia and biotechnology or pharmaceutical industries seeking solutions in bioscreening.

Instrumentation and resources

This infrastructure brings together all the scientific potential and know-how at iMed.ULisboa to provide the most complete molecular screening service and in-depth capability to perform biological evaluation of cell function in several contexts and cellular models. It is quipped with instrumentation, automation, and software for running medium- to high-throughput screens using a variety of assay technologies. The facility is highly flexible, making it possible to run assays of any size, using 96- and 384-well plates, and includes:

- Cell culture rooms with biosafety level 2 containment laminar flow hoods, CO_2 incubators, inverted microscopes coupled to imaging system and support equipment;
- Bright-field, fluorescence and confocal microscopes with dedicated cameras, imaging and acquisition systems;
- Automatic liquid handling platform (Multidrop Combi Reagent Dispenser; Thermo Scientific) for 6 to 1536-well plates;
- Multi-label plate readers (GloMax®Multi-Microplate Multimode Reader; Promega) for 6 to 384-well plates, accommodating luminescence, fluorescence and visible/UV absorbance modules, and dual injector system for 6 to 96-well plate formats;
- Real-time label free impedance-based cell analysis (xCELLigence RTCA SP; ACEA Biosciences) for in 96-well format;
- End-point thermocyclers (Bio-Rad and Thermo Scientific);
- Real-time PCR systems in 96-well format (7300 Real-Time PCR – Applied Biosystems) and in 384-well format (QuantStudio™7 – Applied Biosystems);
- Flow cytometry cell analysis (Guava Easycyte 5HT flow cytometer; Merck Millipore) in 96-well format;
- Gel electrophoresis and transfer systems (Trans-Blot Turbo System; Bio-Rad), absorbance plate readers (Bio-Rad), gel/membrane and X-ray film imaging (Chemidoc MP and Chemidoc XRS; Bio-Rad);
- Biosafety level 3 facilities;
- Animal facilities.

The Molecular BioScreening infrastructure is available to external researchers.

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