RESEARCH INSTITUTE FOR MEDICINES

iMed ULisboa

2014 ANNUAL REPORT

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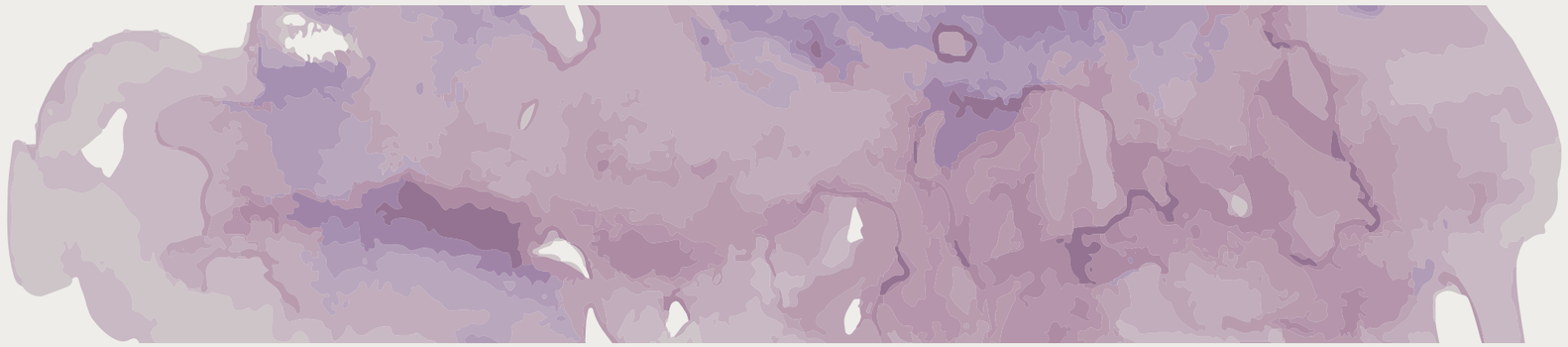
iMed.ULisboa

2014 ANNUAL REPORT



FOREWORD

As part of iMed.ULisboa drive to develop novel treatments and benefit human health through top class research, we have joined forces around the theme *RESEARCH Develops MEDICINES*, ensuring that our research reflects the dynamics of health needs, namely in areas of ageing and ageing-related diseases, such as diabetes, cancer and neurodegeneration, and infection.

iMed.ULisboa capabilities are built around a network of 15 research groups organized in four Program Areas: Drug Discovery, Drug Design, Drug Development, and Drug Usage, spanning the drug discovery and development spectrum, with an emphasis on innovative, multidisciplinary, and collaborative research to develop medicines. In 2014, we hosted 94 integrated researchers holding a PhD and 121 PhD students, in addition to many collaborators and visiting students.

Our research output shows an increasing number of scientific papers published in international peer-reviewed journals, many in high impact journals and with high citation numbers, such as Nature Biotechnology and Nature Reviews in Drug Discovery. Overall, 177 papers were published in 2014. International registered patents have steadily increased to 4 in 2014. Finally, iMed.ULisboa researchers continue to attract competitive external funds, both from contracts with government bodies and industry research contracts.

In addition to the scientific findings and examples of knowledge transfer, other indicators and outputs support our vision and objectives for the future. We are involved in an expanding number of networks and synergies to ensure recognition of iMed.ULisboa value and contributions, aiming at returning the impact of fundamental research to society. We are involved in research infrastructures, locally and at the global level; we have embraced and integrate large consortium initiatives of training programs in pharmaceutical medicines and drug safety, bringing together top universities/institutes and pharmaceutical industry.

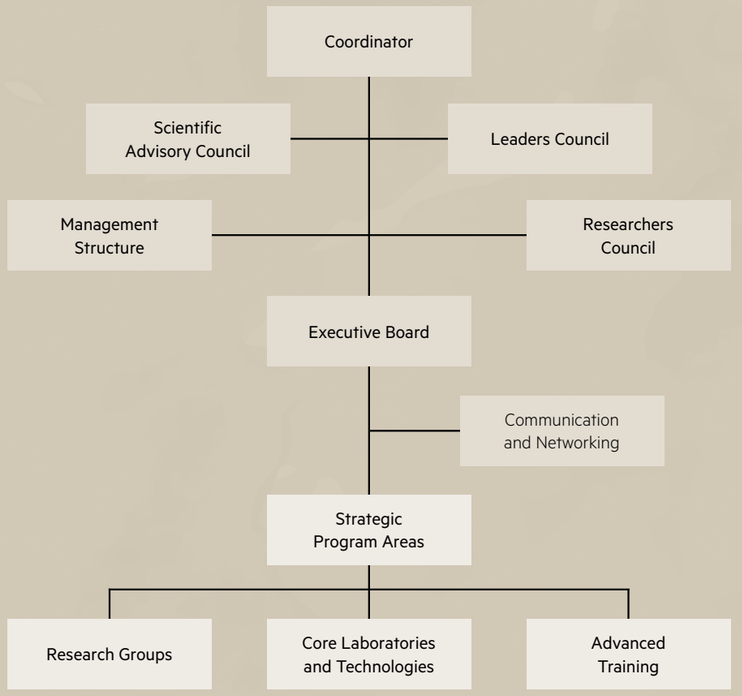
Another fundamental facet of iMed.ULisboa activity is to promote dialogue with the public on science and medicines in general. We have engaged with non-specialist audiences, including public events, school visits, appearances in the media and communicating research to health professionals, patient groups and policy-makers, in collaboration with scientific societies, and the national agencies.

On behalf of iMed.ULisboa, I would like to thank everyone who has collaborated with iMed.ULisboa throughout this year. We strive to build on our unique role in contributing to more effective ways for medicines development to succeed, from understanding fundamental questions, with specific life and health issues in mind, to effectively pursuing unrelenting disease problems in today's society. We look forward to continuing to work together in the upcoming years.

Cecília Rodrigues
Coordinator, iMed.ULisboa

INTRODUCING
iMed.ULisboa

ORGANIZATION AND STRUCTURE



4 PROGRAM AREAS

15 RESEARCH GROUPS

215 RESEARCHERS

SCIENTIFIC ADVISORY BOARD

Independent eminent international scientists at the Scientific Advisory Board advise iMed.ULisboa and ensure our strategic direction is in the best interest of science and society.

Pierluigi Nicotera
German Center for Neurodegenerative Diseases
Bonn, Germany

Ruth Duncan
Cardiff University
United Kingdom

Stephen Caddick
University College London
United Kingdom

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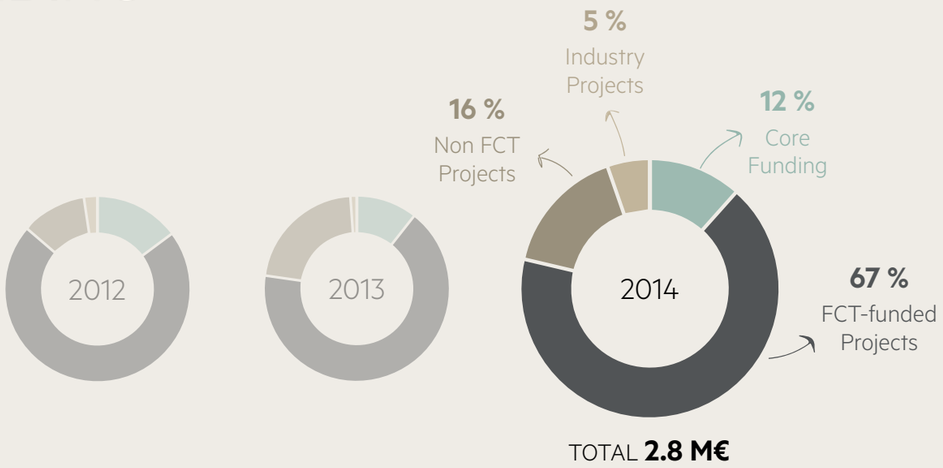
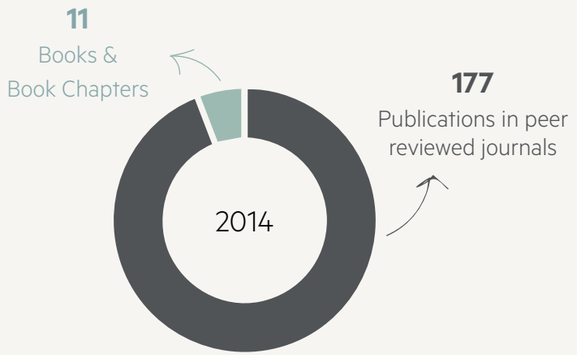
FACTS & NUMBERS

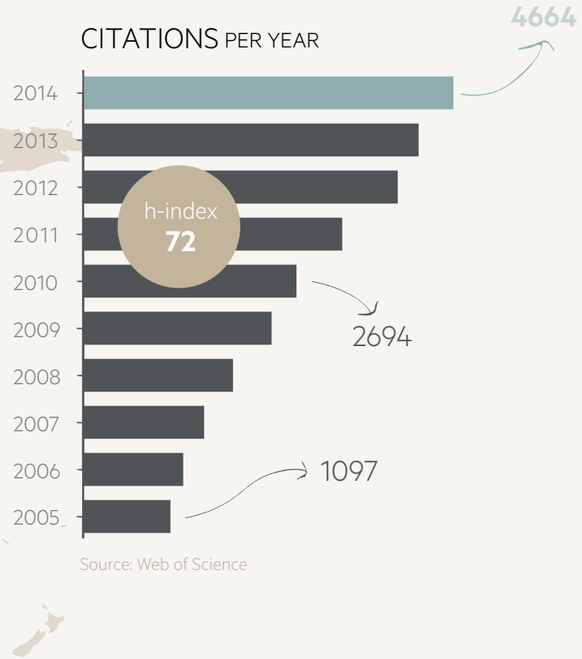
FUNDING



INTERNATIONAL COLLABORATIONS

PUBLICATIONS & CITATIONS





PRIZES & RECOGNITIONS

iMed.ULisboa Scientist, **Beatriz Silva Lima** leads IMI Scientific Committee

iMed.ULisboa scientists, **Cecília M. P. Rodrigues**, **António Almeida** and **José Azevedo Pereira**, funded through the Gilead GÉNESE Program

iMed.ULisboa researcher, **João Vicente**, received the first prize award in the Annual Contest of Innovation at Hovione

Pedro Borrego, **Nuno Taveira** and **Helena Florindo** researchers at iMed.ULisboa were awarded the European prize "Partnering for Cure Research Funding 2014"

Oncognostics, composed by **Joana Amaral**, **Pedro Borralho** and **Rui Castro**, a spin-off team from iMed.ULisboa, wins StartHealth@ULisboa 1st prize

Pedro Borrego, September 2014

Oncognostics, December 2014

PATENTS

Saraiva L, Santos MMM, Pereira NAL, Pereira C, Moreira S, Leão M, Monteiro A, Soares J. "**Tryptophanol-derived oxazoloisoindolinones: small-molecule p53 activators**", WO 2014/207688 (PCT/IB2014/062617, 2014/06/26)

Saraiva L, Santos MMM, Pereira NAL, Pereira C, Moreira S, Leão M, Monteiro A, Soares J. "**Small-molecule p53 activators**" Patent request INPI nº 20131000048238, 2013/06/26

Ribeiro HM, Raposo S. "**Cold process emulsion as vehicle for anti-inflammatory drugs: composition and preparation method**". Portuguese Patent nº105982 M. Propriedade: UL and Laboratórios Edol. Boletim da Propriedade industrial n. 143/2014 from 2014/07/29

Rodrigues CMP, Lucas SD, Finch MD, Low W, Steer C, Munshi CB. "**Deuterated Bile Acids**". Application/ Control Number: 14/386640.

ORGANIZATION OF INTERNATIONAL CONFERENCES

9th World Meeting on Pharmaceutics, Biopharmaceutics and Pharmaceutical Technology
Lisbon, Portugal, March 31 - April 3, 2014
João F. Pinto, Chairman of the Congress and member of the Scientific Board

The International Liver Congress™ 2014
49th Annual Meeting of the European Association for the Study of the Liver
London, United Kingdom, April 9-13, 2014
Cecília M. P. Rodrigues, member of EASL Governing Board

XXIII International Symposium on Medicinal Chemistry
Lisbon, Portugal, September 7-11, 2014
Rui Moreira, Chairman of the Congress and member of the Scientific Board

3 RESEARCH HIGHLIGHTS

DRUG DISCOVERY

THE DRUG DISCOVERY PROGRAM is an initiative to support translational research at the drug target level at iMed.ULisboa. Our drug discovery capabilities are built around a network of research laboratories, institutions and small biotechnology companies. We focus on fully applying our expertise and fostering research excellence on identifying candidate drug-like compounds for selected molecular targets, validating targets/compounds in cell models and pre-clinical studies, and providing a stimulating environment for research training and career development.

The scientific objective of the Drug Discovery Program is to enhance cancer, degenerative and anti-infectious treatments by fostering a highly interactive and vertically integrated drug discovery and development program in which information moves bi-directionally between basic and applied scientists. The Drug Discovery Program comprises a multidisciplinary group of scientists who are committed to making basic laboratory observations on therapeutic targets and potential strategies, exploring their feasibility in pre-clinical models, ultimately translating the most promising approaches and molecules into the clinical setting. Biochemical and molecular biology, microbiology, and neurobiology oriented groups play a pivotal role in iMed.ULisboa research strategy. All participating groups (Cellular Function and Therapeutic Targeting; Host-Pathogen Interactions; Metabolism and Genetics; Molecular Microbiology and Biotechnology; Neuron-Glia Biology in Health and Disease) and associated groups (Medicinal Chemistry; HIV Evolution, Epidemiology and Prevention) are fully integrated laboratories capable of advancing drug discovery projects along the value chain from idea to drug candidate with proof-of-concept, in an highly collaborative environment with the Drug Design and Development Programs.

In addition the Drug Discovery Program at iMed.ULisboa drives a culture of technology platform transfer, through licensing of developed technologies by pharma and biotech industry or collaboration in confidence. Indeed, stimulating creative innovation will be key for biomedical research at academic institutions, to create new opportunities for the industry, and provide faster access to breakthrough drugs by the society.

João Gonçalves

Head of Drug Discovery Program Area

Cecília M. P. Rodrigues

PhD (1996) in Pharmacy (Biochemistry), Universidade de Lisboa

Postdoctoral research at University of Minnesota, USA

Full Professor, Faculdade de Farmácia, Universidade de Lisboa

Selected references

Xavier JM, Morgado AL, Solá S, Rodrigues CMP. Mitochondrial translocation of p53 modulates neuronal fate by preventing differentiation-induced mitochondrial stress. Antioxid Redox Signal. 2014; 21: 1009-1024.

Ferreira DMS, Afonso MB, Rodrigues PM, Pereira DA, Borralho PM, Rodrigues CMP, Castro RE. JNK1-dependent modulation of the p53/miR-34a pathway contributes to apoptosis induced by deoxycholic acid in primary rat hepatocytes. Mol Cell Biol 2014; 34: 1100-1120.

Castro RE, Ferreira DMS, Borralho PM, Machado MV, Cortez-Pinto H, Rodrigues CMP. miR-34a/SIRT1/p53 is suppressed by ursodeoxycholic acid in rat liver and activated by disease severity in human non-alcoholic fatty liver disease. J Hepatol. 2013; 58: 119-125.

Amaral JD, Castro RE, Solá S, Steer CJ, Rodrigues CMP. p53 is a key molecular target of ursodeoxycholic acid in regulating apoptosis. J Biol Chem. 2007; 282: 34250-34259.

Rodrigues CMP, Solá S, Nan Z, Castro RE, Ribeiro PS, Low WC, Steer CJ. Tauroursodeoxycholic acid reduces apoptosis and protects against neurologic injury after acute hemorrhagic stroke in rats. Proc Natl Acad Sci USA. 2003; 100: 6087-6092.

CELLULAR FUNCTION AND THERAPEUTIC TARGETING

PREVIOUS AND CURRENT RESEARCH

Our goal is the identification of novel mechanism-based molecular targets for therapeutic intervention, with a strong focus on understanding and regulating signaling involved in cell death, differentiation and proliferation. We use experimental models of liver, gut and brain disorders involving inflammation, degeneration and regeneration, and test drugs at the preclinical level that may influence disease progression. We have developed bile acids as a group of promising endogenous modulators of cell death/survival, protected by patents and licensed by spin-offs.

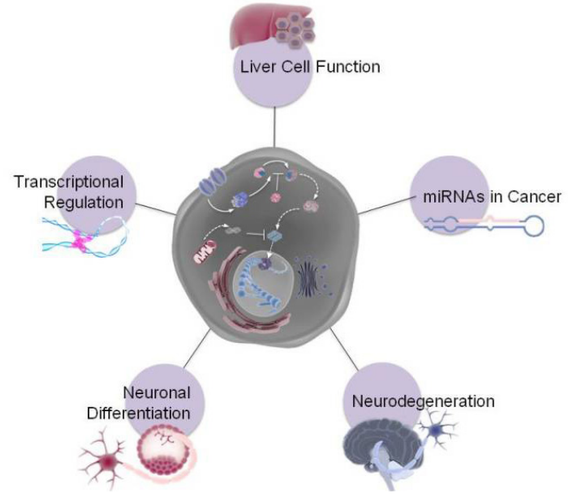
More recently, we have shown that miRNAs regulate liver function and, in particular, that miR-34a expression associates with non-alcoholic fatty liver disease severity. Targeting miR-34a/SIRT1/p53-driven cell death with ursodeoxycholic acid reduces hepatocellular apoptosis and, inversely, toxic deoxycholic acid strongly activates this pro-apoptotic circuit. We have also identified distinct miRNA profiles associated with colon cancer progression. Key miRNA:target gene pathways that influence colon cancer progression and chemotherapy response have also been established, focusing on miR-143 function, as well as innovative methodologies for evaluation of miRNA direct targets in colon cancer.

We have provided evidence for the neuroprotective role of endogenous glutathione S-transferase pi and anti-apoptotic bile acids and showed that mitoprotective bile acids attenuate amyloid pathology, synaptic toxicity, and behaviour deficits. Finally, we have demonstrated that histone deacetylase inhibitors affect neuronal cholesterol metabolism.

Finally, we have also shown that key regulators of neurogenesis interact with apoptosis machinery to direct neural stem cell fate, while reactive oxygen species and key miRNAs stimulate neural differentiation. Further, mitochondrial apoptosis and metabolism also influence neural stem cell fate decision.

FUTURE PROJECTS AND GOALS

Before any potential new therapy can be developed, basic mechanisms of cell function underlying clinically relevant problems must first be identified and thoroughly characterized. We aim to provide an integrative view on the discrete elements of biological systems and how they orchestrate in cells, tissues and organisms. This is key in deciphering how molecular processes change during disease and may be amenable to therapeutic manipulation.



General representation of research activities.

Elsa Anes

PhD (1998) in Pharmacy (Microbiology), University of Lisbon

Posdoctoral research at EMBL, Heidelberg, Germany

Associate Professor, Faculdade de Farmácia, Universidade de Lisboa

HOST-PATHOGEN INTERACTIONS

PREVIOUS AND CURRENT RESEARCH

Our research focuses on the interplay between human cells and several pathogens including *Mycobacterium tuberculosis* (Mtb), Human Immunodeficiency Virus (HIV), Influenza Virus (IV) and *Helicobacter pylori* (HPy). We are interested in characterizing the pathogen determinants involved in these interactions as well as the cell biology and immunobiology of these processes.

During the last decade, we have been committed to understand the pathogenic mechanisms of HIV-2 the interplay between Mtb and phagocytic machinery in infected macrophages. Azevedo-Pereira is expert in the *de novo* infection with HIV in primary human phagocytic cells. Anes is expert in mycobacteria infection and mycobacteria-host interactions.

Mtb success as a pathogen depends on their ability to survive and persist within macrophage and dendritic cells. On the host side of the equation the lab has been developing functional readouts for the environment of the phagosome, and determining how these are modified by immune stimuli and infection. On the bacterial side the group is interested in how the bacterium modifies its intracellular compartment to ensure its survival. This information is being used as the foundation of a high-throughput screen to identify small molecules that kill *M. tuberculosis* inside host cells.

We have two projects aiming to decipher (i) the efficiency, the mechanism and the differences between HIV-1 and HIV-2 cis-infection of Mø and dendritic cells (DC), (ii) how efficiently and rapidly HIV-1 and HIV-2-infected Mø and DC spread the virus to autologous CD4+ T-cells (trans-infection) and (iii) to elucidate the role of cathepsins (Cts) in mycobacterial antigen processing/presentation pathways and/or intracellular killing of mycobacteria either in DC or Mø.

We benefit from recent scientific interaction with Jorge Vitor and Filipa Vale (HPy search for new therapies) and Helena Rebelo de Andrade (IV host interactions and new antiviral targets).

FUTURE PROJECTS AND GOALS

Filipa Vale and Jorge Vitor intend to develop alternative therapies for multi-drug resistant HPy, based on phage therapy including the identification of new lysins and explore their use as a potential "enzybiotic". Rebelo de Andrade basic goals includes to decipher the molecular determinants of pathogenesis and virulence in influenza virus; Influenza antiviral research and exploring antiviral targets. Anes and Azevedo-Pereira objectives are to explore the running projects on the context of HIV infection or during co-infection with Mtb including to define the involvement of Cts in the replication/latency of HIV and during Mtb co-infection; in antigen presentation and, search for signalling networks and microRNAs that controls Cts expression, trafficking host factors or pro-inflammatory events such as inflammasome activation.

Selected references

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Jordao L, Bleck CKE, Mayorga L, Griffiths G, Anes E. On the killing of mycobacteria by macrophages. Cell Microbiol. 2008; 10: 529-548.

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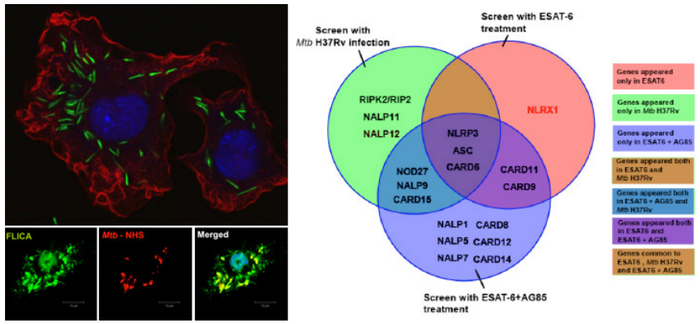

➤ *M. tuberculosis* (green on top, red midlle bottom) infected macrophages activates caspase 1 (FLICA bottom left) to produce the pro-inflammatory cytokine IL1-beta via distinct inflammasomes. Several cytosolic TLR like receptors: NLRs senses Mtb and Mtb PAMPs, ESAT-6 and Ag85.

Ana Paula Leandro

PhD (2001) in Pharmacy (Biochemistry), Universidade de Lisboa

Assistant Professor, Department of Biochemistry and Human Biology

METABOLISM AND GENETICS

PREVIOUS AND CURRENT RESEARCH

The Met&Gen Group aims to study the molecular mechanisms underlying Inborn Errors of Metabolism (IEM) and to identify novel therapeutic targets towards development of new treatments. We have four research areas:

Non-ER-associated Conformational Disorders – Protein misfolding is recognized as a relevant disease mechanism in IEM. We are particular interested in Phenyketonuria, Galactosemia and Classical Homocystinuria. Biochemical/biophysical characterization of the involved misfolded enzymes identified them as targets for conformational/activity rescue (pharmacological chaperones/proteostasis regulators/enzyme activators). Design of enzyme activators were already achieved as well as protein nanoencapsulation systems aiming development of enzyme replacement therapies.

Cellular Methylation Status – S-adenosylhomocysteine and homocysteine are relevant players in cell methylation status with implications in IEM and cardiovascular disease. Using human endothelial cellular models we demonstrated that excess S-adenosylhomocysteine induces hypomethylation of key molecules (DNA, proteins, Sec-tRNA) and is associated with impaired nitric oxide production. Mice models of hyperhomocysteinemia revealed a tissue-specific regulation of homocysteine metabolism and methylation patterns.

Molecular Genetics – Genotyping and gene expression characterization allow inferring disease severity and designing alternative treatments. Splicing mutations are potential targets to antisense therapy and we have demonstrated that a frequent splicing mutation causing Galactosemia is sensitive to LNA. Studying the regulatory mechanisms of PDHA2 testis-specific expression allowed us to envisage somatic activation of this paralog gene as a therapeutic approach for most cases of Pyruvate Dehydrogenase Complex deficiencies caused by PDHA1 mutations.

Mitochondria as Targets for Drugs – Our studies on fatty acid oxidation (mFAO), and energy metabolism contributed to unravel novel mechanisms of drug-associated liver steatosis. New insights were revealed on subcellular transport of fatty acids, the etiology of acylcarnitines as biomarkers of mFAO dysfunction, the carnitine cycle and the CPT1A and CPT2 function. Using mass spectrometry-based targeted-metabolomics novel enzymes of valproate metabolism, intersecting leucine and isoleucine oxidation, and novel mechanisms underlying drug-induced hepatic encephalopathy were defined.

FUTURE PROJECTS AND GOALS

Elucidation of molecular bases of IEM – characterize cellular pathways and players in mutant hPAH proteostasis; elucidate the relationship between S adenosylhomocysteine induced stress and suppression of histone H3K27me3 methylation, and their contribution to proatherogenic endothelial phenotype.

Development of new treatments – evaluate the efficacy of recently developed hPAH-nanoparticles upon cellular/animal models of Phenylketonuria; identify protein stabilizing molecules by HTS of compound libraries; correct splicing-induced defects by antisense oligonucleotides forcing the use of canonical sites thus rescuing wild-type protein production; investigate the modulation of mitochondrial acetyl-CoA dependent pathways and protein acetylation to potentiate or uncover novel pharmacotherapeutic properties of histone deacetylase inhibitors.

Selected references

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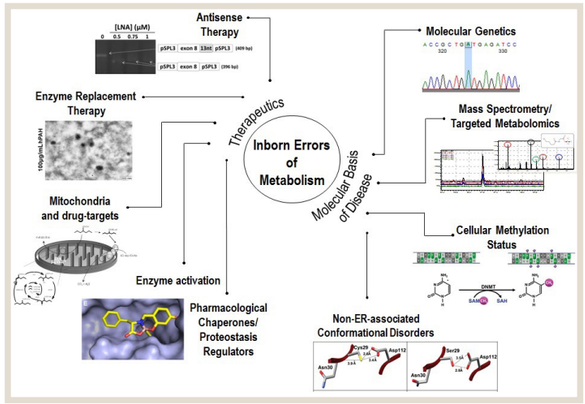


➤ General representation of research activities.

João Gonçalves

PhD (1996) in Pharmacy (Microbiology), Universidade de Lisboa

Postdoctoral research at Harvard Medical School and Scripps Research Institute, USA

Associate Professor, Faculdade de Farmácia, Universidade de Lisboa

Program Area Leader, Drug Discovery at iMed.ULisboa

MOLECULAR MICROBIOLOGY AND BIOTECHNOLOGY

PREVIOUS AND CURRENT RESEARCH

This team is led by Joao Goncalves, and comprises 5 senior research associates. The group has a consolidated track record in several aspects of molecular microbiology and pathogenesis with strong emphasis in biotechnology.

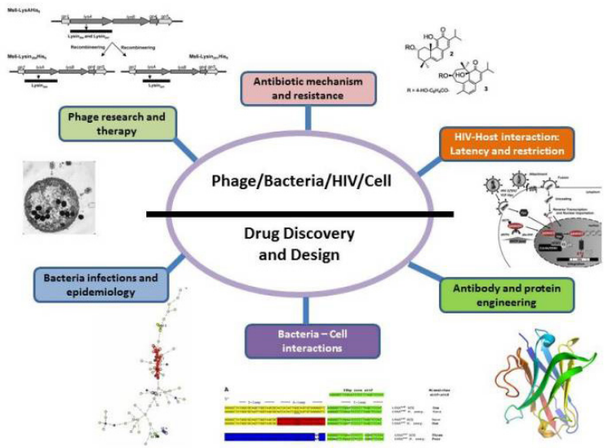
Our main focus is to answer fundamental questions on the molecular mechanisms underlying, promoting and maintaining infectious diseases, how pathogens evolve to other forms of virulence and drug resistance, and the development of molecular strategies to control microbial infection. To achieve the proposed goals, we combine basic research on genetics and molecular biology of mycobacteria and HIV, together with applied protein engineering. We aim to foster the molecular understanding of microbial pathogenesis and the identification of essential steps in the infectious process that can constitute novel targets for therapeutic intervention.

Following this path, we have launched a scientific program directed to the molecular understanding of the interactions between bacteriophages and *Mycobacterium* sp. In addition, we are involved on molecular epidemiology of microbial pathogens (*Mycobacterium tuberculosis*) in hospital and community environments. We also emphasize on factors that are associated with the control of the HIV latency. The use of validated pharmaceutical compound libraries will helps us in this endeavor of modulating viral expression and cellular antiviral defenses. To explore the possibility that viral latency and infection can be both controlled by targeted therapeutics, we developed small antibody scaffolds and full IgG that inhibit HIV-1 infection and helped the specific targeting of lentiviral vectors. Alternative strategies that combine antibody engineering, genetic delivery systems and synthetic biology are being developed to eliminate cells containing viral genomes. The scientific competencies and technologies develop by this group will be a major strength in developing collaboration within iMed.ULisboa, not only in basic molecular microbiology and pathogenesis to identify new therapeutic targets, but also in fostering the design of new biopharmaceuticals.

FUTURE PROJECTS AND GOALS

Our emphasis will be to investigate at molecular level: a) microbial and viral properties associated with infectious diseases and pathogenesis; b) Drug resistance and microbial evolution; c) use the above knowledge to develop innovative biotechnological methods to control infection.

We will boost and sustain our competitive edge in the field of drug discovery and design through greater collaboration and increased multidisciplinary working within iMed.ULisboa. We will seek to identify new biochemical targets for drug development, and will further design those candidate strategies in biodrug design, including new biomolecules and validation of innovative technologies and approaches in areas of unmet need.



General representation of group research activities and goals.

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Dora Maria Tuna Oliveira Brites

PhD (1988) in Pharmacy (Biochemistry), Universidade de Lisboa

Coordinator Investigator, Faculdade de Farmácia, Universidade de Lisboa

NEURON GLIA BIOLOGY IN HEALTH AND DISEASE

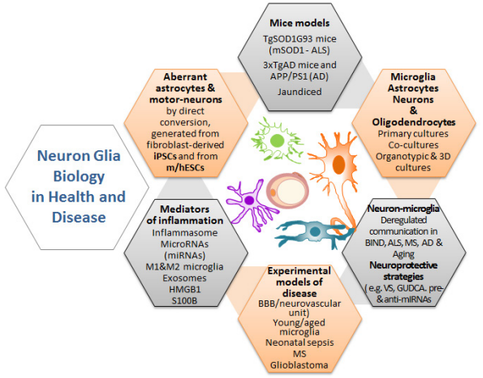
PREVIOUS AND CURRENT RESEARCH

The group of Neuron-Glia Biology in Health and Disease is focused on neuron-glia-vascular interactions during brain development, neurodegenerative diseases and aged-related disorders. To emphasize that from the 15 publications in 2014, 5 of them resulted from interactive collaboration within iMed.ULisboa.

In brain development, we are interested on the sequelae derived from severe neonatal jaundice and sepsis, which we observed to cause neurovascular alterations, astrocytosis and microgliosis with the release of proinflammatory mediators, reduced neuronal density and shrinkage, and myelination deficits. Our studies on the mice model (3xTg-AD) of Alzheimer's disease (AD) pointed out early down-regulation of inflammatory drivers, but increased miRNA-155 expression. These features preceded neuroinflammation and upregulation of high-mobility group box protein 1 (HMGB1). Treatment of young (reactive) microglia with amyloid-β peptide (Aβ) led to decreased phagocytosis, switch to M1/M2 phenotypes, increased HMGB1 expression and inflammasome activation, together with cell senescence. The aged microglia were irresponsive. Interestingly, a vinyl sulfone (produced by the Medicinal Chemistry group) evidenced to reduce the inflammatory profile of Aβ-treated microglial cells. Wild-type and APP/PS1 mice at old age showed hypovascularization and microgliosis in the hippocampus. Astrogliosis was only observed in the AD model. In amyotrophic lateral sclerosis (ALS) the metalloproteinase-9 activation and mitochondria dysfunction in motor-neuron degeneration was counteracted by glycoursodeoxycholic acid. Down- and upregulated proteins were recognized in pre- and symptomatic stages of the SOD1G93A (mSOD1) ALS mice, respectively, and overexpression of miRNA-155 identified as biomarker and therapeutic target. Aberrant astrocytes evidencing the symptomatic stage signature of mSOD1 pups were identified as candidates for targeted therapies. Our studies searching the benefits of a hybrid compound synthesized by the Medicinal Chemistry group revealed its increased benefits over temozolomide. Finally, we demonstrated that S100B was overexpressed in serum/CSF/brain samples of patients with multiple sclerosis (MS) and showed its crucial role in demyelination and associated inflammatory response.

FUTURE PROJECTS AND GOALS

Emphasis will be given to: (i) Impaired neuron-glia communication by neonatal sepsis, AD, ALS, Parkinson's disease and ageing, with a focus at neuroprotection and at recovering astrocytic/microglial function; (ii) Characterization and modulation of miRNA profile in iPSC-generated glia from AD and ALS patients and in cell-derived exosomes; (iii) Neurovascular pathology, therapeutic approaches and strategies to overcome the blood-brain barrier; (iv) New molecular signatures and innovative medicines to brain tumors and the role of neurogenesis; (v) Better understand S100B in MS; and (vi) Novel biomarkers of neurological dysfunction by bilirubin and inflammation, and unique therapeutic solutions aimed at preventing lasting disabilities.



Previous and current research, future goals and experimental models.

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BIOORGANIC CHEMISTRY

MEDICINAL CHEMISTRY

NATURAL PRODUCTS CHEMISTRY

DRUG DESIGN

THE DRUG DESIGN PROGRAM at iMed.ULisboa aims at fully applying chemistry and protein engineering expertise of the participating and associated groups to validate innovative targets and discover novel drug-like compounds and functional proteins with potential for diagnostics and therapeutics for unmet medical needs. This is a chemistry-centered platform oriented to the discovery of biologically active chemical entities that target specific proteins, nucleic acids, or more complex systems such as protein-protein interactions, with the ultimate goal of optimizing their therapeutic properties and value. Working in concert with colleagues from other Program Areas within iMed.ULisboa, we develop solutions for cancer and infectious diseases.

The Drug Design Program includes the Bioorganic Chemistry, Medicinal Chemistry and Natural Products Chemistry core research groups that deliver the chemistry core of the program, and the associated Molecular Microbiology and Biotechnology group that allows new entries into the chemical space beyond small molecules, i.e. peptides and antibody-drug conjugates. These groups make use of a wide-range of technologies and strategies including in silico and biochemical-based screening of focused chemical libraries, fragment- and structure-based design of ligands, bio-guided fractionation of plant extracts, synthetic organic chemistry and development of bioconjugates. State-of-the-art core facilities include a high performance computer cluster with implementation of software for molecular modeling, and an analytical laboratory equipped with mass spectrometry and nuclear magnetic resonance instruments.

Advanced training to forge new generations of young and innovative scientists is at the core of our mission. In close synergism with other program areas, our researchers can find a unique environment for career development and networking within iMed.ULisboa. Importantly, entrepreneurship is a hallmark of this Program Area, with several of the out-of-the-box solutions and methodologies contributing significantly to the patent portfolio of iMed.ULisboa.

Rui Moreira

Head of Drug Design Program Area

Pedro Góis

PhD (2005) in Organic Chemistry, Universidade Nova de Lisboa

Post-Doctoral research at Sussex University; University College of London; IST

Principal Investigator, Faculdade de Farmácia, Universidade de Lisboa

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Active research areas of the Bioorganic Group at iMed.ULisboa.

BIOORGANIC CHEMISTRY

PREVIOUS AND CURRENT RESEARCH

The Bioorganic group aims at applying the principles and methods of organic chemistry to address problems of significant biological relevance, taking inspiration from biology to design and implement new highly efficient chemical processes. The team is nationally diverse and highly multidisciplinary, comprising scientists with organic chemistry, biochemistry, biology and chemical engineer backgrounds, enrolled in multidisciplinary research projects spanning over the interface of organic chemistry and biology.

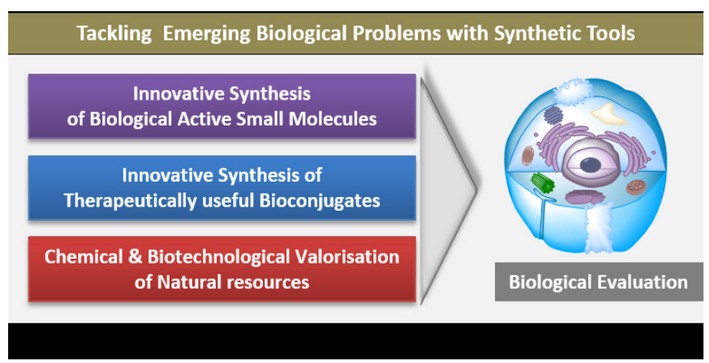
We are currently developing innovative synthetic tools for the preparation of biologically active small molecules based on highly efficient metal (Chem. Eur. J. 2015; ASC 2014, OL 2010; ACIE 2007), organo; (OL 2013; OBC 2011) and bio-catalysis (ChemPlusChem 2015). These efforts recently enabled the preparation of a structurally diverse set of potent antitumor (RSCA 2014) and antiparasitic compounds. The synthesis of several small molecule modulators of human enzymes identified as important therapeutic targets of inflammatory (OBC 2013) and metabolic diseases (RSC Advances 2014), and allowed the toxicity elucidation of key chemical intermediates (Toxicol. Res. 2014).

Empowered by a strong background of methodology design, the group has recently embraced the isolation/synthesis and chemical/biotechnology valorisation of natural resources as a strategy to uncover new biological relevant molecules. We have disclosed a short synthesis of a natural product via microbial transformation of labdanolic acid (Phytochem Lett 2013) and the integrated chemo-enzymatic production of HMF from carbohydrates (ChemSusChem 2013 and 2012).

The use of chemical tools to append chemical probes onto the surface of proteins emerged as a powerful strategy to design new biopharmaceutics and unravel biological mechanisms. Building on our knowledge on the design of water compatible methodologies, we initiated a program to discover new probes and methodologies to selective label biomolecules and construct therapeutically useful bioconjugates. In this area, we have recently disclosed a very innovative methodology that enables the selective functionalization of lysines exposed at the surface of proteins (Chem. Eur. J. 2015; JACS 2012) and we have been exploring folic acid as an efficient targeting molecule for cancer cells (Chem. Com. 2014; OBC 2014).

FUTURE PROJECTS AND GOALS

With the objective of expanding the available tools to prepare biological active small molecules, we will expand our hub of methodologies focusing on the development of bio-inspired and sustainable synthetic methodologies and on the isolation/synthesis and chemical/biotechnology valorization of natural resources. Furthermore, recognizing the importance of establishing generic synthetic methods for the chemical manipulation of proteins, we will actively engage in the discovery of innovative methodologies to selectively modified proteins at different natural residues. These methods will be further applied in the construction of therapeutically useful bioconjugates and in the interrogation of key cellular events.



MEDICINAL CHEMISTRY

Rui Moreira

PhD (1991) in Pharmacy (Pharmaceutical Chemistry), Universidade de Lisboa

Full Professor, Faculdade de Farmácia, Universidade de Lisboa

Program Area Leader, Drug Design at iMed. ULisboa

PREVIOUS AND CURRENT RESEARCH

The research of the Medicinal Chemistry group is driven by a desire to apply chemical and biology concepts to understand the relationship between molecular structure and biological activity, and to identify and optimize small molecules that can be used as starting points for therapeutic intervention.

The design of target-based chemical libraries is one of our major research topics. Heme detoxification into hemozoin by malaria parasites is the primary target of antimalarial drug chloroquine (CQ). We developed novel hemozoin ligands endowed with potent antimalarial activity against CQ-resistant strains (Paulo, 2014). Our libraries also incorporate endoperoxide-based compounds that are selectively activated by parasites to form carbon-centered radicals and release cysteine protease inhibitors, thus providing a dual-acting mechanism of action (Oliveira, 2014). In addition, we pioneered the design of potent agents acting on different stages of the malaria parasite's life-cycle, which led to the discovery of potent dual-stage hybrid compounds that can be used as tools to eradicate the disease (Miranda, 2014).

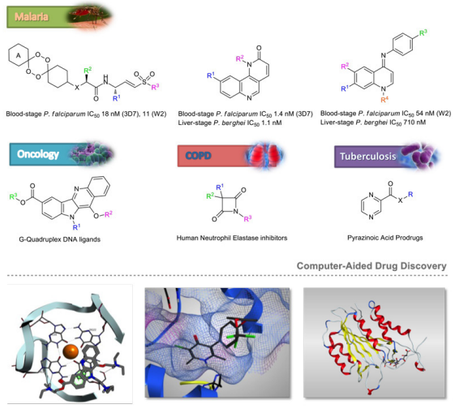
G-quadruplex (G4) DNA structures in telomeres and oncogenic promoter regions are potential targets for cancer therapy. Our group recently discovered new G4 binding chemotypes that selectively decreased Hsp90 and KRAS expression levels in cancer cells (Lavrado, 2013). Restoration of the tumor suppressor p53 activity by inhibition of the p53-MDM2 interaction has also been considered an attractive approach for cancer treatment. Using focused spirooxindole-based libraries we identified novel lead compounds that inhibit the MDM2-mediated p53 regulation hub and present anti-cancer activity (Ribeiro, 2014; Monteiro, 2014).

Human neutrophil elastase (HNE) is a validated target for the treatment of chronic obstructive pulmonary disease and is also a potential target in lung cancer. Using our computational core facility, we developed a virtual screening protocol that enabled the identification of novel chemotypes as HNE inhibitors (Lucas, 2012), which were subsequently optimized for nanomolar inhibitory potency and metabolic stability (Lucas 2013).

FUTURE PROJECTS AND GOALS

The Medicinal Chemistry group will continue to address the current lack of small-molecules capable of selectively interfering with challenging targets involved in infectious diseases and cancer by combining compound library design with advanced computer-aided drug refinement tools to streamline the hit generation process.

Translating lead compounds discovered from phenotypic (cell-based) screenings into drug candidates requires their target(s) to be identified. Suitable strategies, including bioorthogonal click chemistry, will be developed to identify the potential druggable targets of validated lead compounds. Our expertise in covalent inhibitors will also be used to develop activity-based probes to accelerate the discovery novel biomarkers for inflammatory processes.



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Recent achievements of the Medicinal Chemistry group at iMed.ULisboa.

NATURAL PRODUCTS CHEMISTRY

Maria José Umbelino Ferreira

PhD (1990) in Pharmacy (Pharmaceutical Chemistry), Universidade de Lisboa

Associate Professor, Faculdade de Farmácia, Universidade de Lisboa

PREVIOUS AND CURRENT RESEARCH

The Natural Products Chemistry group provides iMed.ULisboa with the expertise in the identification and development of novel hit/lead-drug candidates from natural sources. Having background in organic and medicinal chemistry and advanced spectroscopic techniques, the isolation, structural elucidation and molecular derivatization of novel bioactive chemical scaffolds from plants are the main skills of our group.

We are focused on two main research areas: anti-cancer and anti-infectious agents. Particularly, our main interest is to discover anticancer compounds, with emphasis on targeting multidrug-resistant cancer cells. In silico studies were also performed to achieve this objective. The development of anti-infective molecules from African medicinal plants is also one of our goals. In detail, we are focused on:

1. *Anticancer compounds*

Taking advantage of the great chemical diversity and bioactivity of plant-derived compounds, our group has addressed the multidrug resistance (MDR) issue in cancer by identifying novel anticancer lead scaffolds. We have been focused on the development of reversers of the efflux pump P-glycoprotein (P-gp), the discovery of compounds without cross-resistance in cancer cells exhibiting a drug-resistant phenotype and the development of effective apoptosis inducers.

The development of effective MDR reversers requires a deep comprehension of P-gp efflux mechanism. Using state-of-the-art computational techniques, the murine P-glycoprotein crystallographic structure was used as a molecular target template, enabling further in silico studies on the efflux mechanism and structure-activity relationships. This approach aims to guide molecular derivatization of the lead scaffolds to improve the effectiveness and selectivity of the obtained derivatives.

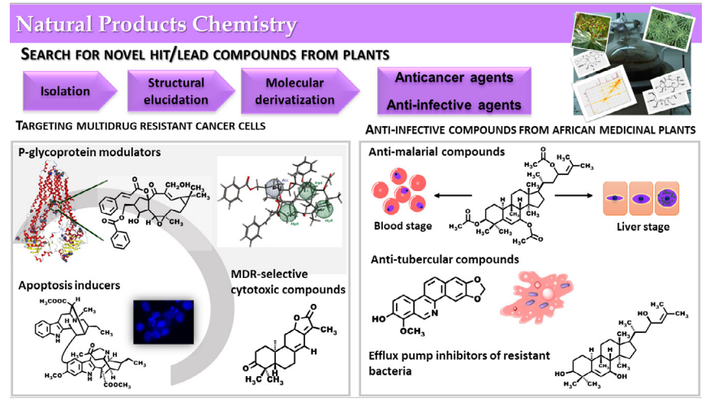
2. *Anti-infective compounds*

The development of hit/lead compounds against infectious diseases such as malaria and tuberculosis, from African medicinal plants used in traditional medicine, is also at the core of our goals. This approach can benefit from the knowledge of plants among natives accumulated over many generations, representing a major strategy for discovering new effective compounds.

FUTURE PROJECTS AND GOALS

For 2015-2020, the major interests of our group will be the development of novel hit/lead-drug candidates by isolation of new bioactive chemical scaffolds from natural sources and molecular derivatization of selected compounds, using hemi-synthetic methodologies. Through this strategy, small libraries of bioactive natural product derivatives, having complex, chiral and highly functionalized structures, will be developed.

We will focus on the search for novel bioactive compounds from plant sources, emphasizing two main areas that intersect with other iMed. ULisboa groups: i) cancer; and ii) infectious diseases.



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Recent achievements of the Natural Products group at iMed.ULisboa.

CHEMICAL BIOLOGY AND TOXICOLOGY

INNOVATIVE PLATFORMS FOR NON-PARENTERAL DELIVERY SYSTEMS

INTRACELLULAR TRAFFICKING MODULATION FOR ADVANCED DRUG DELIVERY

NANOSTRUCTURED SYSTEMS FOR OVERCOMING BIOLOGICAL BARRIERS

DRUG DEVELOPMENT

THE DRUG DEVELOPMENT PROGRAM AREA at iMed.ULisboa represents an interface between Drug Discovery and Design, and potential preclinical development, as well as future clinical development of very innovative approaches arising from those two programs. Meanwhile, the Drug Development Program also introduces main strategic research with scientific and technological innovation. As such, it is supportive of initiatives within the Drug Discovery and Drug Design Program Areas but it also promotes its own initiatives in innovative basic and applied research.

The Drug Development Program is the main and key enabler of supportive research within iMed.ULisboa capable of transforming drug candidates into medicines candidates. For that purpose, we use both innovative technological platforms and innovative validated methods for preclinical development. Our research looks not only to applied areas like cancer, inflammation, genetic disorders and infection, both also through the use of preventive and therapeutic strategies, via relevant research approaches in cell and molecular biology, with major implications in future healthcare practices.

Within this Program main areas of research include: (i) basic cell and molecular mechanisms including the use of biophysical tools; (ii) materials science and (bio)pharmaceutical technology, frequently (but not exclusively) focusing on development of new (bio) molecules and nanomedicines; (iii) pharmacokinetics and biodistribution; (iv) preclinical pharmacology including toxicology, and development of innovative methods and platforms for translational research; and (v) regulatory science.

Beatriz Silva-Lima

Head of Drug Development Program Area

Maria Henriques L. Ribeiro

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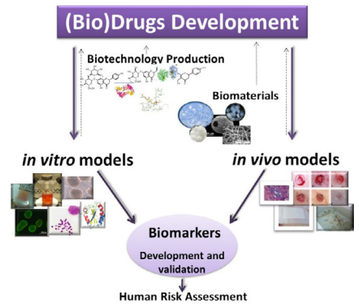

Development of biotechnology-derived drugs, and implementation of alternative methods (e.g. 3D culture models) for toxicological and drug development purposes toward biomarkers and human risk assessment..

CHEMICAL BIOLOGY AND TOXICOLOGY

PREVIOUS AND CURRENT RESEARCH

We aim at developing innovative research for the promotion of healthy living and ageing and contribute to the translational research potential of the institute. For this purpose, a multidisciplinary team with profound knowledge on physico-chemical, toxicology and biological processes uses emerging biotechnology and toxicological approaches for the bioengineering, screening and pre-clinical testing of drug safety and efficacy and investigation of new predictive biomarkers.

Our focus is on drug development, including the design and characterization of value-added biocompounds using biotechnology complementary approaches, the modulation of cancer therapeutics, and the molecular mechanisms of toxicity aiming the development of biomarkers in risk assessment, and the investigation of neuroprotector agents. The specific goals have been to address the challenges associated with: (i) Cell and enzyme-based biotransformation; (ii) Tailoring and biocompatibility studies of biomaterials (iii) Development of 3D culture models for predictive toxicology of drugs; (iv) Modulation of genotoxic stress in human mammary cells; (v) Identification and validation of biomarkers for neurotoxicity and tumoral monitoring; (vi) Risk-benefit analysis of the new biomolecules.

Our research areas focused on:

Biotechnology and Bioengineering Development - We aimed to promote translational research at iMed.ULisboa, developing biotech compounds (e.g. small-molecules, proteins) based on enzyme and cell-culture systems: from bioengineering with process intensification through miniaturization to downstream processing towards biotechnology-derived therapeutics and drug delivery systems; tailoring polymeric (bio)responsive biomaterials networks for high-affinity protein retention; develop and implementation of innovative bioconversions (e.g. (de)glycosylation, lipidation of peptides, production of biosurfactants) towards food and pharmaceutical applications.

Chemical and Cellular Interactions - We were devoted to the study of mechanisms associated with interactions between chemicals and biological systems. In particular, we focused on: a) the development of 3D cell cultures for toxicological and cell therapy studies; b) the development of novel macrocyclic compounds, namely catalytical antioxidants (SOD mimetics) and anti-angiogenic agents (Cu(II) chelators); c) formulation, tailoring and biocompatibility evaluation of acrylic and ceramic based biomaterials and/or drug-loaded medical devices.

Mechanistic Toxicology, Biomarkers and Risk Assessment - We focused on the study of the mode of action (MoA) of neurochemicals and chemical carcinogens in order to identify targets and select endpoints of neurotoxicity and carcinogenicity to be used as predictive biomarkers of disease and/or therapeutics efficacy. Ultimately, the goal was to address protective and antagonistic (pharmacological) complementary strategies in order to prevent or minimize the risk of disease.

FUTURE PROJECTS AND GOALS

Within the framework of iMed.ULisboa the CBT Group perform state of-the-art research for the development of biotechnology-derived drugs and implementation of alternative cell-based methods for predictive toxicology and drug development purposes. Moreover, the identification and development of biomarkers for human risk assessment of xenobiotics (including drugs) and biochemical factors influence in health and disease are also within the aims of our group.

Helena Maria Cabral Marques

PhD (1991) in Pharmacy (Pharmaceutical Technology), Universidade de Lisboa

Associate Professor, Faculdade de Farmácia, Universidade de Lisboa

INNOVATIVE PLATFORMS FOR NON-PARENTERAL DELIVERY SYSTEMS

PREVIOUS AND CURRENT RESEARCH

We are focused on innovative strategies through a multidisciplinary approach integrating materials engineering and physic-chemical characterization of innovative drug delivery systems (technology platforms). Our research is mainly based on the design and development of new carriers for drug transport (polymers, sugars, cellulose derivatives, cyclodextrins), and use of state-of-the-art technologies (spray-drying, freeze-drying, supercritical fluids) to manufacture/produce and characterize these systems and respective materials (e.g. polymorphs state). The main technology platforms are based on the use of micro-/nano-based dosage forms (macromolecular complexation and/or particle engineering), prepared from materials of different nature used for the transport of various bioactive agents for specific diseases. We also thoroughly characterize these platforms.

We have collaborated with several groups within iMed.ULisboa. We are now in the process of strengthening these collaborations and expanding to new ones providing our expertise in the design, development and analysis of drug products with improved performance.

Our main achievements summarized:

1. Understanding the best spray-drying and Supercritical CO2 conditions for drug-cyclodextrins complex formation leading to particles engineered for lung delivery. These technologies allow preparing particles with appropriate characteristics to be used in several new delivery systems.

2. New application of extrusion and co-extrusion technology on the design of new dosage forms (laminar extrusion).

3. Bacterial immobilization (e.g. Lactobacilli) in the new delivery systems

4. Development and characterization of stable polymorphs to carry in the new systems containing olanzapine, gabapentine or paroxetine.

5. Prediction of Drug Distribution in Rat and Humans Using an Artificial Neural Networks Ensemble and a PBPK Model

FUTURE PROJECTS AND GOALS

Our strategy aims to:

(i) Strengthen existing collaborations
(ii) Submit competitive funding applications at national/international level.

These aims will be reached through a multidisciplinary approach involving:

1. Usage of new materials (new or modifications on existing polymers, new cyclodextrins) and stabilization of existing ones (e.g. polymorphism);

2. Design and chemical-physical characterization of new platforms according to the different administration routes specificity;

3. Modulation of the substances' release (in-vitro/in-vivo) from the platforms using a mechanistic perspective;

4. Targeting of the platforms to action sites;

5. Application of statistic tools (QbD) in the design-of-experiments (DoE) in order to construct and optimize these platforms.

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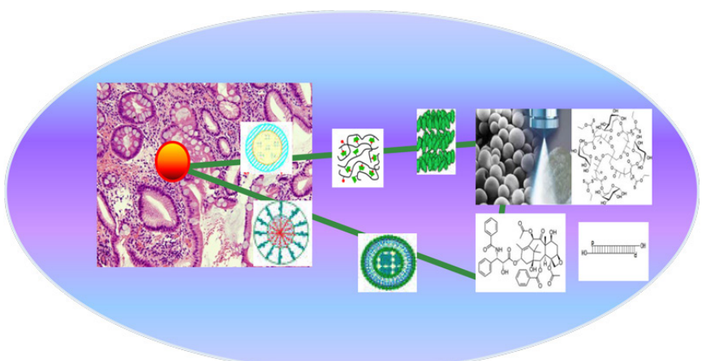

Target organ/tissue reached by innovative platforms for non-parenteral delivery systems.

Helena F. Florindo

PhD (2008) in Pharmaceutical Technology, Universidade de Lisboa, NL

Assistant Professor

INTRACELLULAR TRAFFICKING MODULATION FOR ADVANCED DRUG DELIVERY

PREVIOUS AND CURRENT RESEARCH

Our main scientific strategy covers two main areas towards the development of advanced drug delivery systems (ADDS) adequate for Nanomedicine-based strategies in specific clinically relevant situations (cancer, genetic diseases) and/or related to immunomodulation:

1. Characterization of molecular mechanisms of cellular dynamics both in disease and intracellular trafficking of colloidal carriers-mediated bioactive molecules delivery or effect on cross-talk between immune cells. Specific issues deal with: (i) tumor and/or immune cell targeting (dendritic cells (DCs) and T cells), as well as immunomodulation in disease; (ii) nucleic acid delivery and cytosolic targeting within specific disease mechanisms or immunossupression; (iii) intracellular co-localization of ADDS modulating the mechanisms of disease and disease-response; (iv) interfering with specific cells or intracellular cascades within the mechanisms of disease (e.g. metastisation process and tumor heterogeneity). There is a cross-over in this area with Rodrigues and Fernandez-Llimos (iMed.ULisboa), besides existing international collaborations (e.g. Spain, Israel, Netherlands, Germany, Switzerland).

2. Drug discovery, development and usage.
The group pursues the development of lipid and/or polymeric-based delivery systems for appropriate clinical situations, using "smart delivery strategies" that take advantage from patho-physiological-based challenges to improve the delivery of different agents, including : (i) the use of systems that passively target tumors or liver conditions; (ii) "stealth" long-circulating strategies for inflammation or oncology (primary tumors out of the hepatic space); (iii) targeting modulation using specific ligands to increase the time-residence in pharmacological active sites (tumor microenvironment (TME), metastases and cancer stem cells) and to promote the tumor-associated antigen (TAA) delivery to immune cells and overcome immune evasion through the modulation of immune cell cross-talk and differentiation.

The group is developing new imaging agents for various clinical indications for optical, fluorescent or MRI imaging.

We have well-established collaborations with complementary research groups within iMed. ULisboa (Gonçalves, Góis, Ribeiro, Silva-Lima and Brites research groups).

FUTURE PROJECTS AND GOALS

We aim to rational design multivalent lipid and/or polymeric nanostructure platforms, enabling the delivery of different active molecules (e.g. drugs, antigens, immunomodulators).

Complementary lines of research will be pursued by our team members: host immune modulation; intracellular trafficking and signaling pathways manipulation; biophysics and molecular mechanisms of disease characterization; imaging tools.

ADDS physicochemical characterization will focus the definition of critical quality attributes (CQA) that can be related with relevant critical process parameters (CPP) for the manufacturing processes, establishing appropriate strategies of quality by design (QbD) useful for their lab scale production but also transferable for future pilot and/or industrial batches.

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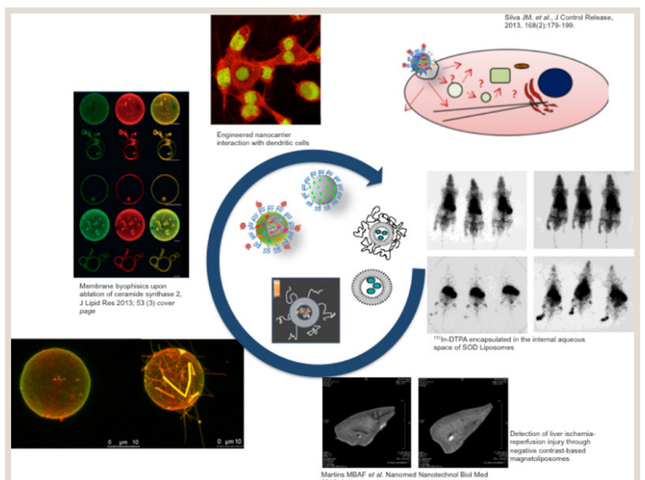
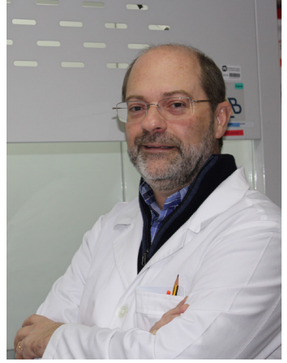


General representation of research group activities.

NANOSTRUCTURED SYSTEMS FOR OVERCOMING BIOLOGICAL BARRIERS



António J. Almeida

PhD (1998) in Pharmacy (Pharmaceutical Technology), Universidade de Lisboa

Full Professor, Faculdade de Farmácia, Universidade de Lisboa

PREVIOUS AND CURRENT RESEARCH

Overcoming biological barriers is the key for the development of many therapeutics and vaccines. Therefore, modulation of transport across biological barriers is one of the major challenges in the drug delivery field.

The group deals with the complex and challenging problems involved in drug transport across biological barriers and delivery at cellular and intra-cellular target sites by means of engineering micro- and nanoformulation of bioactive entities. We aim at developing new drug delivery systems (DDS) adequate for nanomedicine strategies in specific clinically relevant situations (inflammatory, infectious, genetic, autoimmune diseases and cancer) through a multidisciplinary approach involving the existing competencies, as follows:

1. Construction of tailor-made lipid – and polymeric-based nanostructured DDS (nanoDDS) able to improve the performance of bioactive agents directed to specific targets,. Our strategies take advantage from pathophysiological challenges to improve the delivery of drugs, nucleic acids, proteins and antigens including: i) systems that passively target macrophages or phagocytic cells (mostly in infectious diseases but also for liver conditions); ii) stealth long-circulating strategies for cancer therapy as well as dermal and pulmonary delivery for inflammation or genetic diseases and; iii) targeting modulation using specific ligands to increase the time-residence in pharmacological active sites.

2. Development of specific strategies to overcome biological barriers related to the administration route, including comparative exploitation of different routes (parenteral, oral, intranasal, pulmonary, transdermal and topical) to achieve the desired biodistribution profile according to specific physiopathological conditions.

3. In vitro and in vivo characterization of nanoDDS interactions with the biological environment involved in the physiopathology.

4. Vaccine development using nanoDDS, by maximizing immunogenicity without compromising safety and tolerability and elucidation of the immune response.

The research focus allows a wide scientific networking with several groups both outside and within iMed.ULisboa, which is already regarded as a crucial component of our strategy.

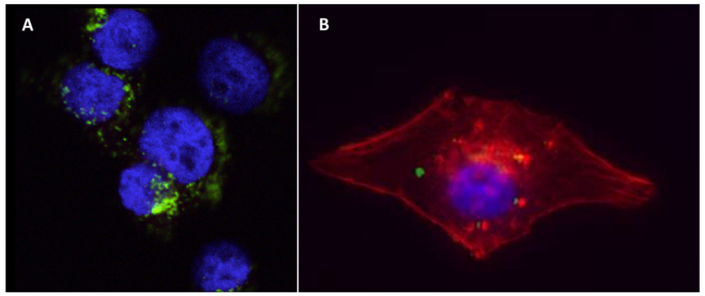
FUTURE PROJECTS AND GOALS

The success of nanoDDS relies on their ability to overcome all the barriers between the site of administration and the intracellular target site, which in turn depends mostly on their surface characteristics. The main future goal is to engineer the nanocarriers' surface to further understand their interactions with the different barriers. Future projects include:

Infectious diseases: NanoDDS will be explored for the treatment of malaria, leishmaniasis, tuberculosis, as well as to improve antibiotic efficacy against biofilm-related infections.

Cancer: Strategies include nanoformulations to specifically target malignant melanoma and lung cancer.

Metabolic diseases: In vivo testing with the stable hPAH-containing nanoDDS.



Fluorescence micrographs of cellular uptake of polymeric nanoparticles (green) by macrophages (A) and osteoblasts (B)..

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HIV EVOLUTION, EPIDEMIOLOGY AND PREVENTION

PHARMACOEPIDEMIOLOGY AND SOCIAL PHARMACY

PHARMACOLOGICAL AND REGULATORY SCIENCES

DRUG USAGE

THE DRUG USAGE PROGRAM AREA at iMed.ULisboa aims at translating medicines research to real-world usage. Focus is on quantitative analysis of population medicines use and outcomes obtained from Health Administrative Databases, where prescription and dispensing profiles are retrieved and clinical data can be included, through a variety of research methods such as epidemiological, statistics and observational research, and evidence-based gathering, or mixed methods and qualitative analysis as well as HIV Evolution. For 2015-20, the Drug Usage Program at iMed.ULisboa will address research topics under determinants of inappropriate use of medicinal products, providing solutions that can be endorsed by Regulatory Agencies and healthcare professionals as a mean for maximizing the benefit-risk ratio of a given therapy and more efficient use of the societal resources spent for health promotion and disease prevention and treatment.

We will translate the potential benefits of medicines into positive health outcomes by understanding and shaping the factors that influence their optimal use. Based on our installed capacities, the Drug Discovery Program will act in the discovery of inter-individual and pathogen genetic variations that account for therapeutic failure and/or adverse drug reactions. We have long been involved in monitoring of drug resistance in HIV-1 and HIV-2 infected individuals.

The Drug Usage Program has already been contributing with input across a number of National Health Authorities to ensure economic harmonization and evaluation of biological medicines and medical devices, as well as standardization in safe medication practice. We host the Pharmacovigilance South Unit, supported by the National Authority of Medicines and Health Products, INFARMED. Drug Usage Program comprises a multidisciplinary group of scientists who are committed to translational research in a collaborative environment with the other Program Areas at iMed.ULisboa, involving the participating groups of Pharmacoepidemiology and Social Pharmacy and HIV Evolution, Epidemiology and Prevention. We highlight here strategic collaborations with the IMI, EMA, and European Network in Pharmacovigilance.

Ana Paula Martins

Head of Drug Usage Program Area

Nuno Taveira

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Associate Professor, Instituto Superior de Ciências da Saúde Egas Moniz (ISCSEM), Caparica

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HIV EVOLUTION, EPIDEMIOLOGY AND PREVENTION

PREVIOUS AND CURRENT RESEARCH

Our objectives are to obtain a better understanding of HIV diversity and evolution in Portuguese speaking countries and to translate this information into the production of better diagnosis tests, better treatment regimens, new antiviral products and new vaccine candidates. Using phylogenetic analysis and sequences obtained from infected patients we have been characterizing the epidemiology, diversity, evolution and drug resistance of HIV in Portugal, Angola, Mozambique and Cape Verde. The results of these studies are helping to explain the origins, dissemination routes and transmission dynamics of HIV in these countries and worldwide. The detailed knowledge on HIV diversity has guided our efforts to produce a new HIV DNA PCR assay which is being used to diagnose HIV-1 infection in children born from infected mothers in Angola. The identification of drug resistance profiles is helping clinicians to offer a better treatment to their patients. Finally, we are testing in the mice model a new HIV vaccine candidate based on envelope antigens obtained from Angolan HIV isolates.

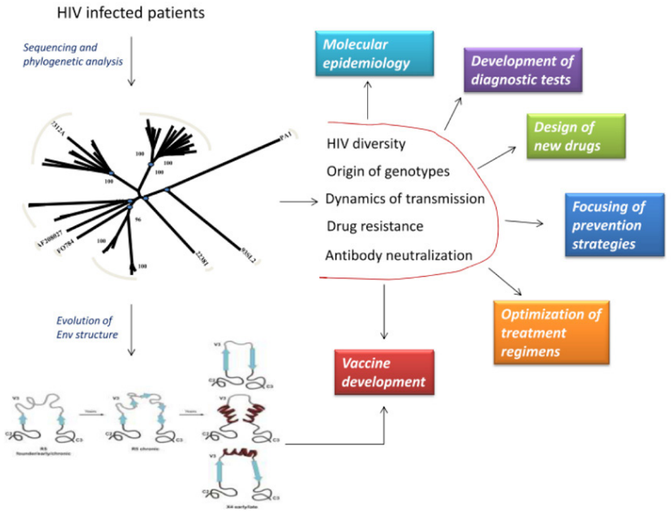
In a second line of research we aim to identify, produce and test at the pre-clinical level new candidate drugs for the treatment and prevention of HIV infection. We design and evaluate the anti-HIV activity of peptides and small molecules synthesized by chemistry departments of several Portuguese Universities. We have already identified three molecules with very potent anti-HIV (nM level). Remarkably two of these also show a potent activity against *Plasmodium berghei*. A collaboration has been set with Miguel Prudêncio at IMM, Portugal, and J. Victor Garcia-Martinez, at the University of North Carolina, Chapel Hill, USA, to test the activity of these compounds *in vivo* (mice model). Our final aim is to produce a candidate microbicide and/or a systemic drug based on these compounds to prevent and/or treat HIV infection and malaria.

FUTURE PROJECTS AND GOALS

HIV cure research – To produce a CRISPR/Cas9-based system to modify the HIV provirus at the RRE and provide the proof-of-concept of its usefulness for curing HIV infection at the cell level.

Vaccine research – Complete the preclinical characterization of our new HIV-1 vaccine.

Research in antiviral agents – Test the microbicide activity of a HEC gel containing the P3 peptide in humanized mice; test the anti-plasmodium and anti-HIV activity of a family of new compounds we have identified recently.



Phenotypic, genetic, evolutionary and structural analysis of HIV isolates from patients residing in Portuguese speaking countries provides basic information enabling the characterization of the HIV epidemic trends in each of the countries, the definition of better diagnostic, treatment and prevention strategies and the development of new antiretroviral drugs and vaccines.

PHARMACOEPIDEMIOLOGY AND SOCIAL PHARMACY

Fernando Fernandez-Llimos

PhD (2003) in Pharmacy, Universidad de Granada, Espanha

Assistant Professor, Pharmacy Practice

PREVIOUS AND CURRENT RESEARCH

During 2014, our group produced 23 items indexed in WoS.

As part of the drug use studies, we completed the assessment of the consumption of antibiotics in Portugal demonstrating a better usage materialized as a reduction in overall consumption, and a reduction of the use of inappropriate antibiotics in primary care (Int J Clin Pharm. 2014), as well as started an investigation on the quality use of OTC medicines in community pharmacies (Res Social Adm Pharm. 2015).

In the Communication in Health area, we continued the work stream following the closed FCT funded project to assess through a specific methodology characteristics of communication between patients and primary healthcare providers, especially with elders (Health Expect. 2013), and also with a focus on medicines package leaflets readability usually limited for patients (Eur J Clin Pharmacol. 2014). In the area of drug information for health care professionals, we could identify serious limitations of the Summary of Product Characteristics in several topics, such us information on dose adjustment for renal impaired patients, or inadequate information for the use in pregnancy and breast feeding that prevent their use in clinical decision making (Br J Clin Pharmacol. 2015).

Aiming to gather the evidence of pharmacists services to improve the outcomes obtained with the use of medicines, we improved a previously designed instrument to characterize pharmacists interventions (J Eval Clin Pract. 2015). But also identified the weaknesses of the most commonly used thesaurus to gather evidence in pharmacy field: the Medical Subject Headings (Am J Health Syst Pharm. 2014 & Res Social Adm Pharm. 2015).

Interaction within iMed with other research groups was accomplished. One example of the cooperative work is the studies in nutrition and toxicology that involved the analysis of food diaries and other exposure and demographic data (J Toxicol Environ Health A. 2014).

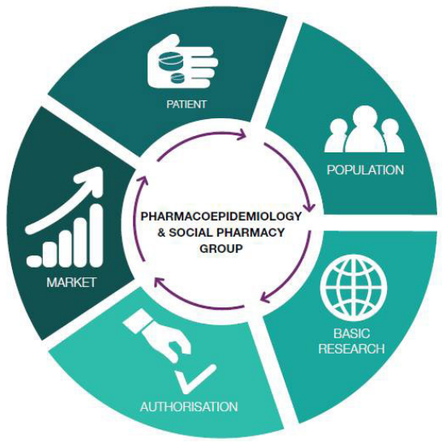
FUTURE PROJECTS AND GOALS

We established the following objectives:

1. To study the positive (efficacy, effectiveness) and negative impact (lack of safety) of medicines use of specific therapeutic classes in specific population groups, with special focus on frail patients, namely elderly, oncologic and transplanted patients.

2. To analyze social, attitudinal, managerial, and cognitive factors associated with therapeutic success of medicines in elderly.

3. To evaluate the elements of the evidence gathering process in order to produce high quality evidence of the effects (desired and undesired) of medicines that may serve as feed-back information for discovery, design and development research units in the production of new therapeutic entities.



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Investigating the use of medicines by individuals and populations in real-world environment.

PHARMACOLOGICAL AND REGULATORY SCIENCES

Beatriz Silva-Lima

PhD (1991) in Pharmacy, Universidade de Lisboa

Full Professor, Faculdade de Farmácia, Universidade de Lisboa

Program Area Leader, Drug Development at iMed.ULisboa

PREVIOUS AND CURRENT RESEARCH

The Group integrates essential translational research competences supportive of First in Human Studies and beyond. The research focus are:

i) Basic and mechanistic pharmacology (*in vitro, in vivo*) covering the pharmacological basis of medicines development / therapeutic use on efficacy and safety, in disease areas with inflammatory components, like ischemia/reperfusion-induced organ injury/dysfunction, reumathoid arthritis or metabolic disorders. Investigational products cover small molecules, natural products, eg herbal medicines and nutraceuticals. Predictive efficacy and safety biomarkers are applied as supportive tools.

ii) Pharmacokinetics (PK) using *in silico, in vitro* and *in vivo* modelling, as well as models of membrane transport and transporters, as factors of drug biodisposition, and characterization of "in life" usage attributes, in special populations. Aquaporins are a strong focus for new medicines development or membrane associated transport mechanisms.

iii) Regulatory science is also a supportive component for drug development research and is a research area *per se*, with a very high level of involvement and experience. We integrate basic science and regulatory requirements on data generation which supports human administration of test molecules, and we contribute to produce and apply international regulatory requirements (European Medicines Agency (EMA) and International Conference of Harmonization) for drug development studies. Several researchers in the Group are highly experienced and internationally recognised Regulatory Scientists contributing to European Commission, EMA, Portuguese Medicines Agency, Academia, on preclinical safety, PK, orphan drugs and medicines assessment and approval. We advise on preclinical "study packages" for clinical trials and marketing of new molecular candidates, including proof of concept in vitro and in animal disease models, PK parameters and safety endpoints.

iv) High training competences, post-graduate, professional, national international, on drug development requirements and strategies is also a strong activity (eg EUDIPHARM, Pharmatrain, CEMDC, SafeSciMet, RAMPS). Master/PhD theses, using data mining eg addressing non-human primates use in medicines research were sponsored.

FUTURE PROJECTS AND GOALS

Research will progress on i) Translational pharmacological science, testing small molecules, natural products, nutraceuticals mode of action/safety and therapeutic potential in inflammatory conditions eg chron, diabetes, cancer. ii) Pharmacokinetics prediction for test molecules through in vitro, modelling, Populational PK. iii) Biological transport research will focus on membrane transport systems function/regulation for understanding transporter defects mechanisms; identifying transport systems as drug targets; screening transporter modulators.iv) Regulatory science platform will integrate research outputs, proposing/identifying specific study design needs, endpoints, tools, markers, towards First in Human studies. Involvement/contribution with national/international training projects Medicines Regulations and Regulators will continue. Collaboration with IMI will progress.

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Translational research towards *in silico, in vitro* and *in vivo* approaches driving new drugs development from experimental setting to human use.

4

FACILITIES & SERVICES

ANIMAL FACILITY

The animal facility has a housing capacity of around 500 rodents (rats and mice) and includes several rooms for animal maintenance and experimental procedures (surgical and others). Metabolic cages (individual cages for monitoring nutrient input and waste output) are available. The facility is also equipped for sterilization of rooms and equipment. Provides support on protocol development and licensing, refinement and advice in experimental procedures, and simple surgical techniques by certified researchers, as well as services of husbandry and routine daily care (feeding, watering, and cage changing) by qualified animal care technicians.

Licensed by the National Authorities "Direção Geral de Alimentação e Veterinária".

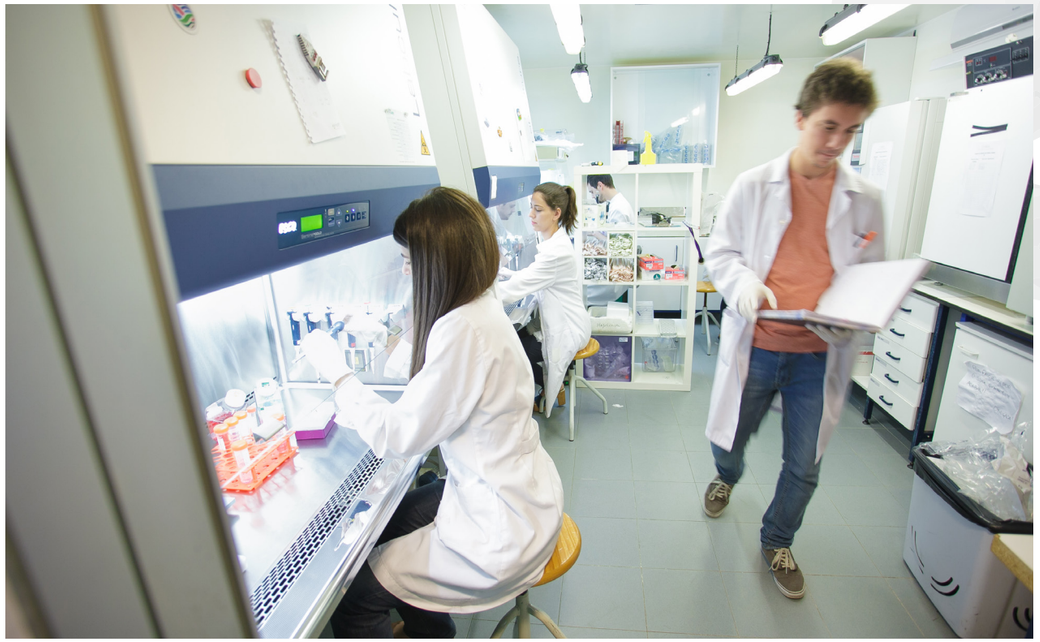
CELL FUNCTION

This facility has dedicated cell culture rooms well equipped to perform all cell culture work. It is tailored for the biological evaluation of cell function, routinely determining the role of transgenes and the cytotoxic and cytoprotective activities of synthetic and natural compounds, in multiple cell models, including immortalized cells (human, monkey, rat, and mouse), primary cultures of liver and brain cells (rat and mouse), and stem cells. This is available as a full service provided by experienced technical personnel.

Additional dedicated equipment include bright field inverted microscope (Zeiss) with image acquisition and system (Leica), high-throughput capabilities with GloMax®-Multi+Microplate Multimode Reader (Promega), accepting 6, 12, 24, 48, 96 and 384-well plates, and equipped with luminometer, fluorescence, and visible/UV absorbance modules and dual injector system for 6 to 96-well plate formats; and xCELLigence RTCA SP (ACEA Biosciences) for real-time label free impedence-based cell analysis in 96 well format.

CELL BIOLOGY

This facility is dedicated to the analysis of gene expression at DNA, RNA and protein level, routinely performing a multitude of biochemical and molecular biology techniques using standard equipment for electrophoresis, western blot, northern bot, as well absorbance plate reader, and gel, membrane and X-ray film imaging (Chemidoc MP and XRS; Bio-Rad). It is also equipped with end-point (Bio-Rad and Thermo) and real time PCR (Applied Biosystems 7300 Real-Time PCR System) thermocyclers, and a Guava easycyte 5HT benchtop flow cytometer for high-throughput cell analysis in 96-well format. Additionally, there is a stopped-flow (HiTech Scientific) and two Patch-clamp setups (Axon Instruments), used for kinetic analyses of ion channel activity in cells. Fluorescence and bright field microscopes (Zeiss) with dedicated cameras (Leica) and acquisition software are available.



COMPUTER ASSISTED DRUG DESIGN

Linux-based high performance computer cluster with 424 CPU cores, 4GB per CPU and 2 TB per node with a specific implementation of state-of-the-art software for molecular modeling, molecular dynamics, virtual screening, and *de novo* design.

RADIOISOTOPE

The radioisotope facility (gamma and beta emission) includes areas for labeling of proteins, other macromolecules and low molecular weight compounds, by chemical modification. The physical proximity to the animal facilities enables in vitro and in vivo studies, namely pharmacokinetics, biodistribution, and metabolite studies. This laboratory has strict rules driven by requirements for improved safety of workers on handling radioactive samples. All new users must perform radiation safety training.

MASS SPECTROMETRY

The mass spectrometry facility is equipped with a Triple Quadruple mass spectrometer (Micromass Quattro Micro API, Waters) with ESI and APCI ion sources. The equipment is suitable for the identification and quantification of small molecules in complex matrices, as biological fluids and extracts of natural products. This facility is also equipped with an Ion-Trap (LCQ-Fleet, Thermo) mass spectrometer dedicated to the characterization of proteins and biological conjugates. The service is available for users on a "do-it-yourself" basis (self-service), for long-term studies. However, an initial training period is required. A technician is also available (full-service)

Equipment is part of the National Mass Spectrometry Network (REDE/1501/REM/2005).

NUCLEAR MAGNETIC RESONANCE

The NMR facility is equipped with a NMR Spectrometer (Bruker Avance 400). It promotes the use of nuclear magnetic resonance spectroscopy in the areas of structure-based drug design, including structure characterization, fragment-based drug design, metabolomics and dynamic host-guest interactions. These activities are part of our strategic interests in thematic areas of drug discovery, drug design and drug development.

BIOSAFETY LEVEL 3 SECURITY

This facility is specifically dedicated to research involving biological pathogens of level 3. It was designed to minimize the risk of personnel and environmental exposure to potential hazardous agents according to European and Portuguese legislation. This facility has an anteroom for material and personnel preparation, and a main procedure room equipped with tree vertical laminar flow chambers (type A2 and type B2), three CO2 incubators (Hera Cell), one normal incubator, two benchtop centrifuges (Eppendorff), a benchtop ultracentrifuge (Beckman), an aerosol-tight microfuge (Eppendorff), a Tecan infinite 200 multimode microplate reader, water baths, freezers, refrigerators, optical microscopes and inverted phase-contrast microscopes (Leica) and a dedicated double door pass-through autoclave (Matachana).

This facility is available to external researchers. All users must have a specific biosafety level 3 training, and must follow strict rules and guidelines while working in the facility.

CENTRO DE FARMACOVIGILÂNCIA - SUL

This facility provides competencies raging from reception, validation, analysis and evaluation of suspected adverse drug reactions, dissemination of pharmacovigilance in the southern region of the country and training in pharmacovigilance field, to scientific activity and research related to drug use safety.

NÚCLEO DE PRESTAÇÃO DE SERVIÇOS

This service provides clinical analyses for diagnosis and monitoring, open to general public, social services, healthcare units, state and private hospitals and non-profit organizations.

The Clinical Biochemistry laboratory offers unique means for the diagnosis of familial hyperbilirubinemias (Gilbert and Crigler Najjar syndromes), as well as for diagnosis and prognosis of various acute and chronic liver diseases, and their therapeutic monitoring. The diagnosis and therapy follow-up of inherited metabolic disorders is performed through the characterization of the metabolic profiling and biomarker identification. This laboratory is member of European Network ERNDIM and one of two laboratories in the country with the technological and scientific knowledge to perform these particular analyses.

The Clinical Microbiology laboratory is a reference in the diagnosis and monitoring of HIV infection/AIDS and other associated infections, such as tuberculosis and fungal infections. The Microbiological Control laboratory provides services for the control of pharmaceutical, biological and cosmetic products, antibiotics and others.

The Molecular Biotechnology laboratory performs immunogenicity assays against biotechnological drugs and quantification in the blood of therapeutic proteins.

ADVANCED **TRAINING**

i3du

Discovery
Design
Development
Usage

PHD PROGRAMME

THE PHD PROGRAMME IN MEDICINES AND PHARMACEUTICAL INNOVATION (i3DU) is a joint initiative of two reference universities in Portugal, the University of Lisbon (ULisboa) and the University of Porto (U.Porto), grouping competences and boosting quality in postgraduate training, in cooperation with the pharmaceutical industry. Both universities will award the PhD degrees (accredited by A3ES) that anchor the i3DU Programme. The consortium involves the Faculties of Pharmacy at ULisboa (FF/ULisboa) and U.Porto (FF/U.Porto), and the Research Centers iMed.ULisboa at ULisboa, and REQUIMTE and IBMC. INEB at U.Porto, in close and effective collaboration with Hovione and Novartis as major industry partners. The i3DU represents a strong commitment of academia and pharma in Medicines and Pharmaceutical Innovation training that meets well-defined standards for high quality international PhD training and is in line with the preconized research-innovation-education triangle in Europe.

Students who thrive in our programmes are expected to be passionate about their subject. Many will be recognized nationally for their scholarship, research, and public engagement, and make original and substantial contributions to their disciplines. Our graduates become idea-leaders who drive our global future by advancing discoveries, broadening knowledge, fostering entrepreneurship, and developing new technologies.

ipSC

iMed.ULisboa Postgraduate Students Commission

2013-2014 iPSC members

The main aims of iMed.ULisboa Postgraduate Students Commission (ipSC) are to contribute towards the complementary formation of postgraduate students of iMed.ULisboa, and to stimulate the interaction between the different scientific fields of the institute, promoting an investigation of excellence, and focused on the surrounding population needs.
The ipSC contributes to complementary training of postgraduate students at iMed.ULisboa, promoting bimonthly workshops, the annual Postgraduate Students Meeting, as well as several social activities.

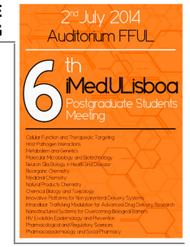
2014 HIGHLIGHTS

Mar — **WORKSHOP**
Intelectual Property: From Academics to Business
by Miguel Santos and Ana Neves

May — **GET TOGETHER PARTY**

Jun — **WORKSHOP**
Quantify what our brain tell us about our results - ImageJ Tutorial
by Adelaide Fernandes

Jul — **VI POST-GRADUATE STUDENTS MEETING**



Oct — **WORKSHOP**
A Light Scent of Bioinformatics
by Jorge Vítor

Nov — **GET TOGETHER PARTY**

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Research
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Medicines