

FUNDACIÓN DE LA COMUNIDAD VALENCIANA
CENTRO DE INVESTIGACIÓN PRÍNCIPE FELIPE

MORE RESEARCH BETTER HEALTH

ANNUAL REPORT
2018



PRINCIPE FELIPE
CENTRO DE INVESTIGACION



The mission of the **Centro de Investigación Príncipe Felipe** is to enhance knowledge of the molecular basis of human diseases, with the aim of translating benefits to patients. Our core values are excellence, collaboration, innovation, transparency, and equality.

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BOARD OF TRUSTEES

The Board of Trustees celebrated three meetings in 2018: 25th of June, the 22nd of October, and the 14th of December.

President

Hble. Sra. Dña. Ana Barceló Chico

Universal and Public Healthcare Counsellor of the Government of Valencia

Vice-President

Excmo. Sr. D. Santiago Grisolia

Valencia Council of Culture Chairman

Members

Narcís Vázquez Romero

María Amparo García Layunta

Ana M^a Ávila Peñalver

Carmelina Pla Silvestre

Vicente Boluda Fos

Manuel Llombart Bosch

Antonio Pellicer Martínez

New members since December 14th

María A. Blasco Marhuenda

Carmen Ayuso García

Isabel Fariñas Gómez

Óscar Marín Parra

Secretary (non-voting member)

Deborah Burks

In accordance with the Spanish Transparency Legislation (Spanish Royal Decree 451/2012 of March 5) and the By-Laws of the CIPF Foundation, the Board of Trustees are not remunerated.



FOREWORD



Deborah J. Burks Director

2018 has been a great year for the CIPF; our research groups have advanced with cutting-edge scientific projects and together, we have all achieved historic institutional goals. Early in the year, the new management team and principal investigators developed a five-year strategic plan (2018 to 2023) which was enthusiastically approved by our Board. This strategic scientific plan was implemented mid-2018 with the overall aim of positioning the CIPF on the map of international centres of research excellence. Our mission is to perform cutting-edge biomedical research with the goal of translating discoveries to the clinic so that our efforts benefit those who need it most: patients in the public healthcare system.

One of our major accomplishments was to abolish conditions that have mandated closure dates and subjected employees to frozen salaries since the budget cuts of 2011. Following a negotiation process, the labour union and director of CIPF signed an agreement to end this unfavourable situation and to implement a plan for updating salaries and revising job categories. Many thanks to the labour union for your cooperation and dedication to the employees of this research institute. Improving work conditions is a necessary step forward in making the CIPF a more competitive research centre.

In 2018, we named a new Scientific Advisory Board (SAB) composed of prestigious and internationally recognized scientists. The member of the new SAB will play an important role in guiding and evaluating the implementation of our new strategic plan and are key to developing international collaborations. We are grateful to these outstanding scientists for agreeing to advise and promote the CIPF. The SAB includes: Sharon Gerecht, Director of the Johns Hopkins Institute for NanoBiotechnology (INBT); the endocrinologist and geneticist, Jens C. Brüning, (Director of the Max Planck Institute for Metabolism Research); Michelle Bradbury, Director of Imaging Research, Sloan-Memorial Kettering Cancer Centre; Leslie B. Vosshall, Director of the Kavli Neural Systems Institute at The Rockefeller University; Josep Tabernero, Director of the Vall d'Hebron Institute of Oncology (VHIO); Antonio Zorzano, Head of Molecular Medicine Programme, IRB Barcelona; Adriana Maggi, Director of the Centre of Excellence on Neurodegenerative Diseases, University of Milan; Sabine Werner, Chair of the Institute of Molecular Health Sciences, ETH Zurich.

We are also indebted to the new members of our Board of Trustees who joined us in 2018: President, Ana Barceló Chico, Minister of Public Healthcare of Generalitat Valenciana, and the elected new members Drs. Maria A. Blasco (CNIO), Isabel Fariñas (UV), Óscar Marín (UCL), and Carmen Ayuso (Foundation Jimenez Diaz), all of whom are recognized for the excellence of their scientific research as well as their strong commitment to translating biomedical research to the benefit of all patients.

67% of our researchers are female and thus, we are committed to supporting women in science and scientific management. For the first time in its history, the CIPF has a Gender Equality Plan 2018-2022 and a committee to oversee our objectives such as equal opportunity and flexible work time. The CIPF recognises that good science includes consideration of gender and sex differences in experimental design and interpretation of results. Thus, to promote good scientific practice one of our strategic goals is the creation of a Gender in Biomedical Research program. At the end of 2018, we signed an agreement with the University of Valencia to create a mixed research unit headed by Dr. Amparo Oliver, full professor of Psychology. Dr. Oliver is an expert in statistics and study design and brings years of experience in research related to loneliness and ageing.

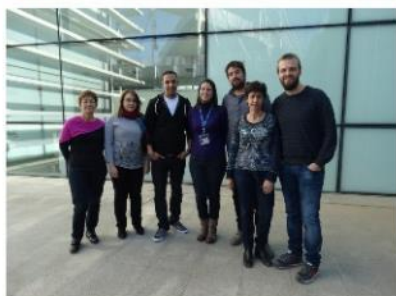
2018 has been an important year for the recruitment of new research groups. Dr. Enric Esplugues spent more than a decade at Yale University and has joined us as a Senior Investigator who leads our Immunobiology Group. The laboratory of Dr. Esplugues will focus on the cellular and molecular mechanisms of the immune, research that has a tremendous translational potential for drug discovery. Dr. Pietro Fazzari is a Ramon y Cajal fellow who has joined the center as a junior group leader. His group, Cortical Circuits in Health and Disease, pursues research on circuit development and neuronal regeneration. They are developing new approaches to restore cortical function upon stroke or brain injuries.

Finally, a special mention is reserved for two principal investigators who have contributed to the excellence of the CIPF during their long, very productive careers. Dr Erwin Knecht and Dr M^a Eugenia Armengod retired in July 2018 after more than 30 years as principal investigators and leaders in their respective fields. Dr Armengod made important contributions to our understanding of the molecular mechanisms which regulate mitochondrial RNA. Moreover, Dr. Armengod was director of the Instituto de Investigaciones Citológicas, predecessor of the CIPF, from 1995 to 2000 and implemented many policies that modernized the research lines and management of the institute. Dr. Knecht is considered one of the founders of the field of autophagy and has pioneered this new area of science which has led to important discoveries about the molecular basis of ageing and pathological processes related with many human diseases from cancer to rare diseases. He was an investigator of the national network CIBERER. Both dedicated years of service to the Ministry of Science as evaluators and panel members. Both are exemplary mentors who throughout the years have trained dozens of doctoral students and postdocs.

Maria Eugenia and Erwin: We dedicate this annual report from 2018 to both of you. Thank you for your scientific contributions and for sharing your knowledge, wisdom, and experience with us.

The core values at the CIPF as defined by our new strategic plan include excellence, collaboration, innovation, transparency, and equality. I cannot think of two people who better represent these values than Maria Eugenia and Erwin. You will be missed for your intelligence, for your rigor, for your sense of humor, for your mentoring skills, and your passion for science.

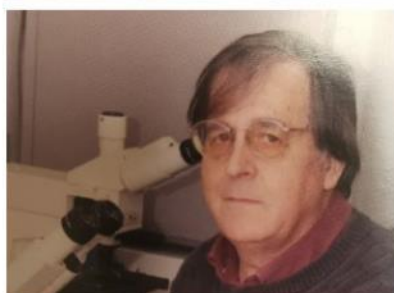
Retirement Tribute



40 YEARS OF SCIENCE Dr. M^a Eugenia Armengod

First Woman Director Instituto de Investigaciones Citológicas,
Group leader of the CIPF RNA Modification & Mitochondrial Diseases Lab

Retirement Tribute



40 YEARS OF SCIENCE Dr. Erwin Knecht

Group leader of the CIPF Intracellular Protein Degradation
Principal Investigator, CIBERER.

A photograph of a laboratory interior, heavily tinted with a blue color overlay. The scene shows a workbench with a sink, a rack of pipette tips, and various laboratory equipment and supplies. Shelves in the background are filled with boxes and containers. The word "RESULTS" is prominently displayed in white, bold, sans-serif capital letters in the lower right quadrant, underlined.

RESULTS

2018

FUNDED RESEARCH PROJECTS

| ENTITY | 2018 ACTIVE PROJECTS | |
|--------------------------------|-------------------------|----------------|
| International funding | 6 | 5,90% |
| European Commission | 4 | |
| Private funding | 2 | |
| National Public funding | 48 | 47,05% |
| MINECO | 21 | |
| Ministry of Education | 9 | |
| Ministry of Health | 2 | |
| ISCIII Health Institute | 16 | |
| National Private funding | 17 | 17,65% |
| Regional funding | 20 | 19,60% |
| Regional Ministry of Education | 17 | |
| Regional Ministry of Health | 1 | |
| Regional Ministry of Industry | 2 | |
| Industrial Contracts | 10 | 9,80% |
| Total | 101 | 100,00% |

2018

PUBLICATIONS



Papers were published in indexed journals



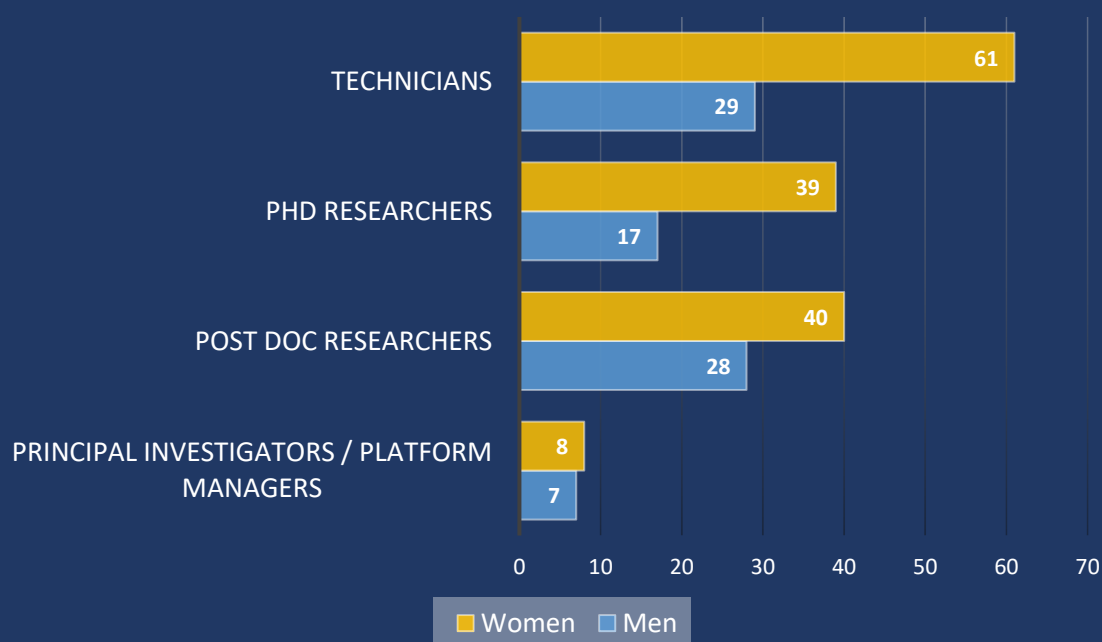
Published in Q1

| | Average Impact Factor | Indexed Publications | Publications in 1st quartile (Q1) Journals | Percentage of publications in Q1 Journals |
|------|--------------------------|-------------------------|--|---|
| 2017 | 5,31 | 93 | 61 | 66% |
| 2018 | 5,67 | 82 | 65 | 79% |

2018 HUMAN RESOURCES

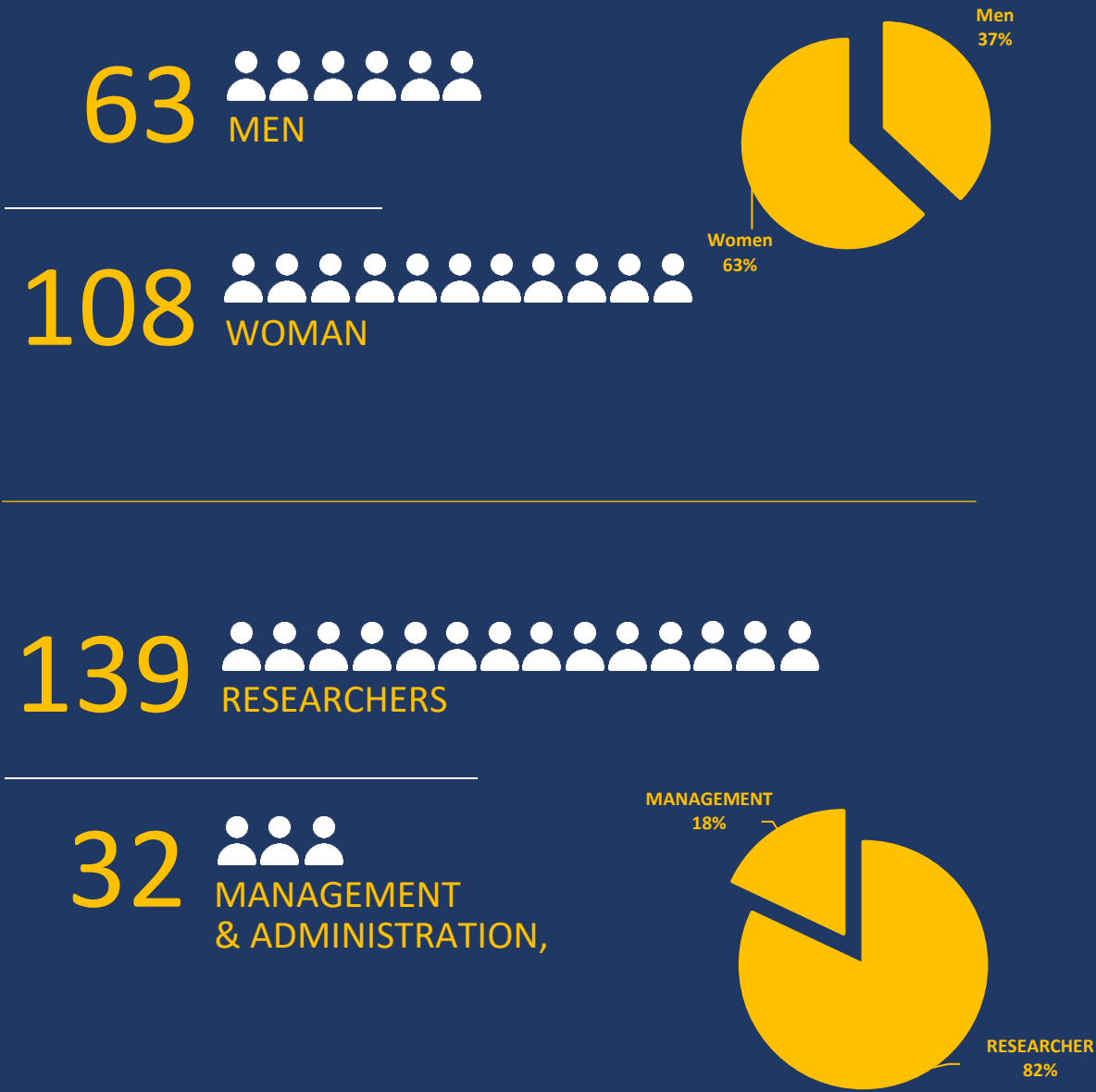
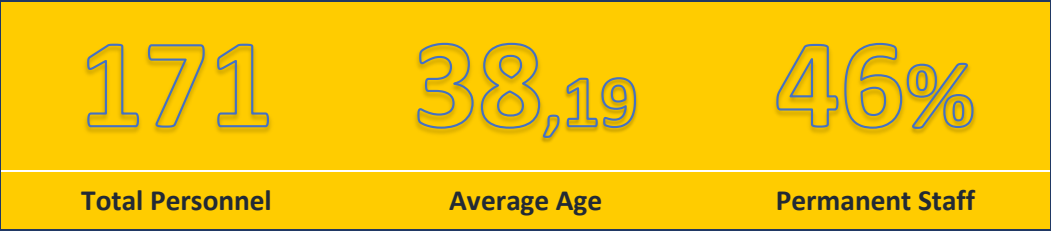
| PERSONNEL | NUMBER | % | MEN | WOMEN |
|--------------------------|--------|----|-----|-------|
| Principal Investigators | 15 | 4 | 7 | 8 |
| Postdoctoral Researchers | 68 | 18 | 28 | 40 |
| Predoctoral Researchers | 56 | 15 | 17 | 39 |
| Technicians | 90 | 23 | 29 | 61 |

Research Staff



| 2018 CIPF Personnel | Number |
|---------------------|------------|
| Collaborators | 117 |
| Students | 98 |
| Staff | 171 |
| Total | 386 |

2018 STAFF

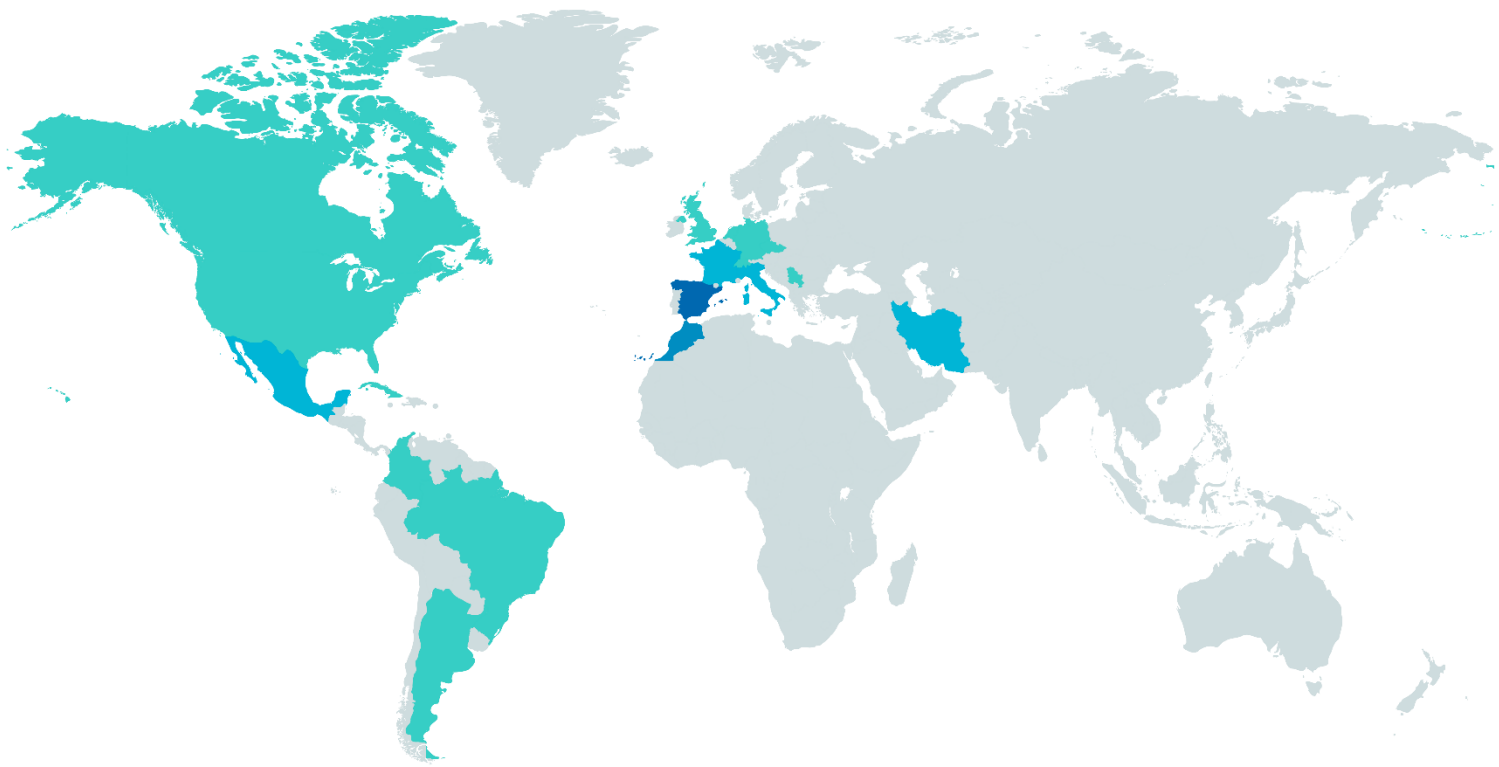


NATIONALITIES CIPF STAFF

| STAFF | Men | Women | Total |
|--------------------|-----------|-----------|-----------|
| Researchers | 25 | 54 | 79 |
| Spanish | 16 | 48 | 64 |
| German | 1 | 0 | 1 |
| Croatian | 1 | 0 | 1 |
| U.S. | 0 | 1 | 1 |
| French | 0 | 1 | 1 |
| Italian | 4 | 2 | 6 |
| Moroccan | 1 | 0 | 1 |
| Ukrainian | 1 | 1 | 2 |
| Argentina | 0 | 1 | 1 |
| British | 1 | 0 | 1 |
| Technical | 21 | 38 | 59 |
| Spanish | 19 | 36 | 55 |
| Armenian | 0 | 1 | 1 |
| French | 1 | 0 | 1 |
| Italian | 1 | 1 | 2 |
| Management | 18 | 15 | 33 |

NATIONALITIES CIPF COLLABORATIONS

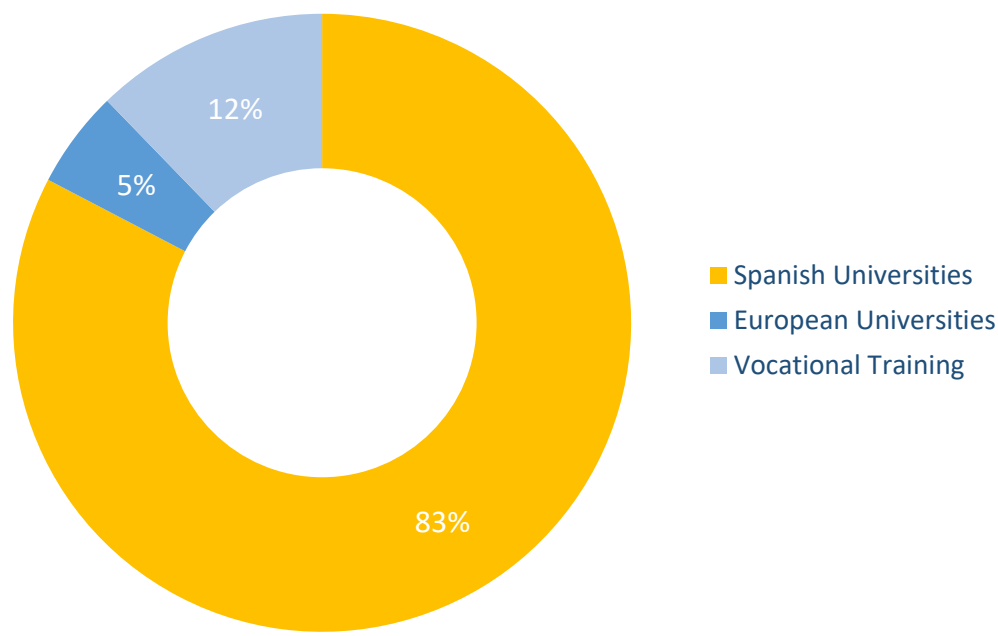
| COLLABORATIONS | Men | Women | Total |
|--------------------|-----------|-----------|-----------|
| Researchers | 31 | 54 | 85 |
| Spanish | 23 | 43 | 66 |
| U.S. | 1 | 0 | 1 |
| Italian | 0 | 1 | 1 |
| Moroccan | 2 | 2 | 4 |
| Argentina | 0 | 1 | 1 |
| Brazilian | 0 | 1 | 1 |
| Canadian | 0 | 1 | 1 |
| Colombian | 1 | 0 | 1 |
| Cuban | 1 | 0 | 1 |
| Dutch | 0 | 1 | 1 |
| British | 1 | 0 | 1 |
| Iranian | 0 | 2 | 2 |
| Mexican | 2 | 0 | 2 |
| Serbian | 0 | 1 | 1 |
| Swiss | 0 | 1 | 1 |
| Technical | 9 | 23 | 32 |
| Spanish | 6 | 20 | 26 |
| Romanian | 1 | 0 | 1 |
| French | 1 | 1 | 2 |
| Italian | 0 | 1 | 1 |
| Czech | 0 | 1 | 1 |
| U.S. | 1 | 0 | 1 |



2018 STUDENTS

PhD, Master's and Undergraduate

| 2018 STUDENTS | Men | Women | Total |
|-----------------------|-----|-------|-------|
| | 41 | 57 | 98 |
| Spanish Universities | 34 | 47 | 81 |
| European Universities | 3 | 2 | 5 |
| Vocational Training | 4 | 8 | 12 |





SCIENCE



Genetics and Genomics of Neuromuscular and Neurodegenerative Diseases

Group members

Group Leader
Carmen Espinós Armero

Collaborator Researchers
Vincenzo Lupo

Post-Doctoral Scientist
Natalia Sotelo

PhD Students
Paula Sancho Salmerón
Ana Sánchez-Monteagudo
Candela Machuca Arellano

Technicians
Lola Martínez-Rubio
Amparo Andrés-Bordería

Collaborators
Marina Guillot
Claudia Vázquez
Andrea Català

Overview

In the Laboratory of Genetics and Genomics of Neuromuscular and Neurodegenerative Diseases (<http://espinos.cipf.es>), our main mission is to understand the molecular basis of neurological rare diseases; in particular, hereditary peripheral neuropathies, neurodegeneration with brain iron accumulation and related movement disorders with cerebellar atrophy, Wilson's disease, and hereditary ataxias. We work the genetic analysis of these diseases and in the development and validation of new strategies and new tools, which applied to clinical and health care, provide us with a more accurate and cost-effective diagnosis. Our aim is also to investigate the effect that genes and new mutations have on the central and peripheral nervous system through the use of different assays performed in animal and cellular models. Our research not only contributes to the improvement of people's quality of life, but also helps our knowledge about the mechanisms underlying the disease and, consequently, the discovery of new possible therapeutic targets. It is well known that age, diet and quality of life in general, together with other environmental factors and the same genetic background specific to each individual, contribute to the variability observed in some diseases with the same primary molecular cause. This later information together with the molecular characterization of these diseases, some of them clinically and genetically very heterogeneous, provide us with a better correlation between the genetic cause and the specific phenotype of each patient, a solid basis for personalized medicine.

Research Results

Characterization of the molecular bases of the neurodegeneration with brain iron accumulation disorders (NBIA; ENACH acronym in Spanish). NBIA is a group of inherited neurologic disorders in which iron accumulates in the brain, mainly in the basal ganglia. NBIA are rare diseases with a prevalence of 1-3/1,000,000. Currently we have recruited 134 probands with NBIA or NBIA-mimic. Genetic test of the NBIA genes has allowed us to identify the causative mutation in 54 index cases. The subsequent analysis of 38 patients by a customized targeted-next generation sequencing of 498 genes involved in NBIA and other movement disorders (MoveDisord-498 panel) has led to the molecular diagnosis in 19 patients. Additionally, two families have been investigated by whole exome sequencing and we have discovered a new gene involved in human pathology and the second case linked to mutations in PLEKHG2.

Genetic epidemiology, identification of genetic modifiers and characterization of the microRNA profile associated with the Wilson disease. The Wilson disease (WD) is a rare disorder with an estimated prevalence of 1/30,000. This research line includes three goals: (i) to characterize genetically the WD patients from the Valencian Land and to gain insight into the genetic epidemiology; (ii) to characterize genetic variants that could modify the phenotype of WD patients; and (iii) to identify the microRNA profile that could be useful for prognosis and new therapies for WD. (i) We have performed the genetic study of ATP7B, including introns, promotor and, 3' and 5'-UTRs regions, in 26 probands and achieved the definitive diagnosis in 22 probands. The remaining 4 cases have been investigated by whole exome sequencing. We have detected clinical mutations in CCDC115, gene involved in congenital disorder of glycosylation type IIO (CDG2O), in one family (Fig. 1).

The remaining cases are still under study. (ii) We have investigated three pairs of sibs who present with disparate clinical pictures by whole exome sequencing. After filtering 60 genes related to copper metabolism, different changes are of interest, such as the LMNA c.1255C>T, previously associated with laminopathies. Functional studies using patient's fibroblasts are undergoing. (iii) Twenty WD patients have been analyzed by miRNAseq in a comparative study using twenty controls, paired by age and sex. The informatics analyses of miRNAs that are under- or over-expressed have revealed that 18 miRNAs present a differential expression. Among them, miR-485 and miR-122, which are related to iron metabolism and liver disease. These findings are currently under-validation using qPCR.

Molecular bases which underlie the Charcot-Marie-Tooth (CMT) disease and hereditary motor neuropathies (HMN). This group of hereditary neuropathies shows a wide genetic heterogeneity. Moreover, the clinical and genetic overlap existing between CMT and HMN make the genetic diagnosis even more difficult. This overlap suggests that genetically both diseases should be considered into the same group. To date, more than 80 related genes have been described, and all the incoming genetic findings help the reclassification of phenotypes in patients with difficult diagnosis. The objective of our laboratory is to actively participate in the generation of this new information: discover new genes associated with the disease, new mutations and redefine the correlation between the genotype and the phenotypic variability of these peripheral neuropathies. In our laboratory, using exome sequencing we have identified new genes associated with neuromuscular disorders, and through the implementation of an in-home diagnostic (currently named Neuro119) gene panel we have achieved a promising diagnostic performance for sporadic and familiar cases with difficult diagnosis.

To characterize the MORC2 gene involved in a new form of Charcot-Marie-Tooth disease (CMT). In 2016, we described the MORC2, as a gene involved in a new axonal CMT form, CMT2Z. At present, the number of families published at international level is 33, seven of them identified in our clinical series, so that this gene is proposed as a relatively common gene in axonal forms of CMT. The aim of this study is to investigate the disease mechanism associated with MORC2. We have analyzed the most frequent mutation, p.R252, and the most severe clinical mutation, p.S87L, using a rat neuronal cell model and patients' fibroblasts, by means of a transcriptomics approach. Additionally, we have also investigated the expression of Morc2 in the neuronal model and observed that is mainly detected in the nuclei of the cells, and also in axons, which agrees with the axonal phenotype described in CMT2Z patients (Fig. 2).

Figures

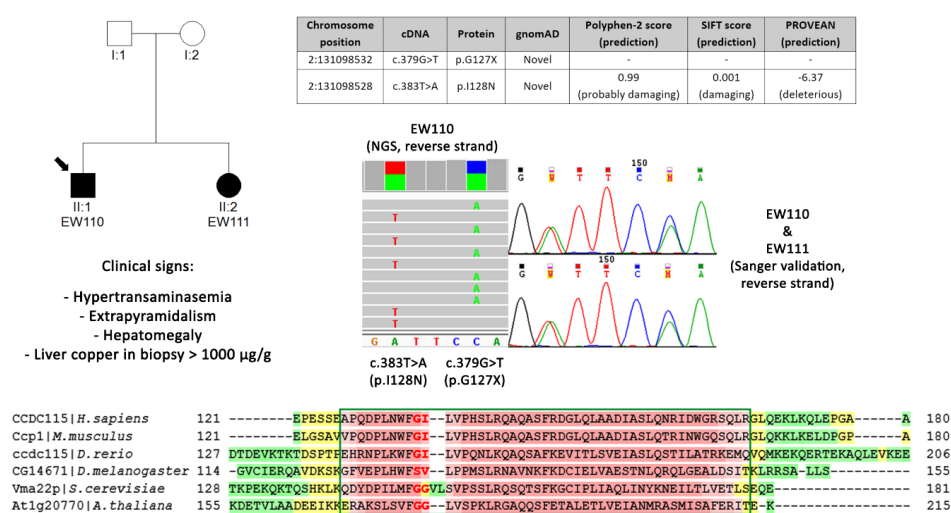


Figure 1. Wilson-mimics. In a family with clinical diagnosis of Wilson’s disease (WD) and negative results for mutational screening of ATP7B, two mutations (c.383T>A/c.379G>T) by whole exome sequencing were identified in CCDC115, gene involved in congenital disorder of glycosylation type IIO (CDG2O). Both disorders present abnormal accumulation of copper in liver. In our clinical series, 85% of patients with WD are carriers of ATP7B mutations. This figure highlights that in patients with clinical suggestive of WD and no ATP7B mutations, disorders with abnormal accumulation of copper must be taken into account.

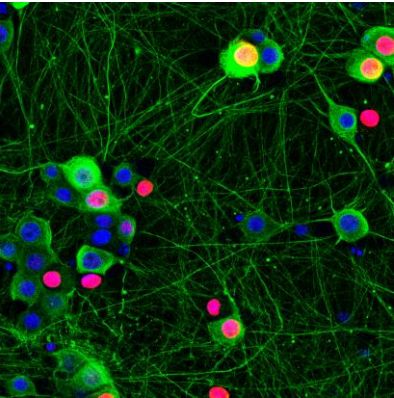


Figure 2. Wild-type Morc2 expression in purified rat sensory neurons. Analysis of MORC2 subcellular localization in neuronal cultures infected with viruses carrying depicted constructs. Virally mediated gene expression was induced for 4 days with doxycycline. The expression of endogenous Morc2 was predominantly detected in the nuclei of the cells, and also in axons, which agrees with the axonal phenotype described in patients with MORC2 mutations. (MORC2 is in red; SMI32 for neurofilaments in green; DAPI for nuclei in blue. Scale bar=20 µm.)

Publications

Tello C, Darling A, Lupo V, Pérez-Dueñas B, Espinós C*. **On the complexity of clinical and molecular bases of neurodegeneration with brain iron accumulation.** Clin Genet 2018; 93: 731-40. (Invited Review)

Calpena E, López del Amo V, Chakraborty M, Llamusi B, Artero R, Espinós C, Galindo MI. **The Drosophila junctophilin gene is functionally equivalent to its four mammalian counterparts and is a modifier of a Huntington poly-Q expansion and the Notch pathway.** Dis Model Mech 2018; 11 (1). pii: dmm.029082. doi: 10.1242/dmm.029082.

Abbott JA, Meyer-Schuman R, Lupo V, Feely S, Mademan I, Oprea SN, Griffin LB, Alberti MA, Casasnovas C, Aharoni S, Basel-Vanagaite L, Züchner S, De Jonghe P, Baets J, Shy M, Espinós C, Demeler B, Antonellis A, Francklyn C. **Substrate interaction defects in histidyl-tRNA synthetase linked to dominant axonal peripheral neuropathy.** Hum Mut 2018; 39: 415-32.

Frasquet M, Lupo V, Chumillas MJ, Vázquez-Costa JF, Espinós C, Sevilla T. **Phenotypic features of two patients diagnosed with PHARC syndrome and carriers of a new homozygous mutation in the ABHD12 gene.** J Neurol Sci 2018; 387: 134-138.

Machuca-Arellano M, Vilches A, Clemente E, Pascual-Pascual SI, Bolinches-Amorós A, Artero-Castro A, Espinós C, León-Rodríguez M, Jendelova P, Erceg S. **Generation of a human iPSC line from a patient with autosomal recessive spastic ataxia de Charlevoix-Saguenay (ARSACS) caused by a mutation in SACSIN gene.** Stem Cell Res 2018; 31: 249-252.

Lupo V*, Frasquet M, Sánchez-Monteagudo A, Pelayo-Negro A, García-Sobrinho T, Sedano MJ, Pardo J, Misiego M, García-García J, Sobrido MJ, Martínez-Rubio D, Chumillas MJ, Vilchez JJ, Vázquez-Costa JF, Espinós C*, Sevilla T*. **Characterizing the phenotype and mode of inheritance of patients with inherited peripheral neuropathies carrying MME mutations.** J Med Genet 2018; 55:814-823.

Machuca-Arellano C, Vilches A, Clemente E, Pascual-Pascual SI, Bolinches-Amorós A, Artero-Castro A, Espinós C, León-Rodríguez M, Jendelova P, Erceg S. **Generation of human induced pluripotent stem cell (iPSC) line from an unaffected female carrier of mutation in SACSIN gene.** Stem Cell Res 2018; 33: 166-170.

Darling A, Aguilera-Albesa S, Tello C, Serrano M, Tomás M, Camino-León R, Fernández-Ramos J, Jiménez-Escrig A, Poó P, O’Callaghan M, Ortez C, Nascimiento A, Candau Fernández Mesaque R, Madruga M, Arrabal L, Roldán S, Gómez-Martín H, Garrido C, Temudo T, Jou-Munoz C, Muchart J, Huisman TAGM, Poretti A, Lupo V, Espinós C, Pérez-Dueñas B. **PLA2G6-associated neurodegeneration: New insights into brain abnormalities and disease progression.** Parkinsonism Relat Disord 2018; doi: 10.1016/j.parkreldis.2018.10.013. pii: S1353-8020(18)30447-4.

Sancho P, Bartesaghi L, Miossec O, García-García F, Ramírez-Jiménez L, Siddell A, Åkesson E, Hedlund E, Laššuthová P, Pascual-Pascual SI, Sevilla T, Kennerson M, Lupo V, Chrest R*, Espinós C*. **Characterization of molecular mechanisms underlying the axonal Charcot-Marie-Tooth neuropathy caused by MORC2 mutations.** Hum Mol Genet 2019; doi: 10.1093/hmg/ddz006.



ENFERMEDADES RARAS Unidad Mixta IIS La Fe - CIPF



**GENERALITAT
VALENCIANA**
Conselleria de Sanitat
Universal i Salut Pública

**TOTS
A UNA
VEU**



Rare Diseases CIPF-IISLAFE Joint Research Unit

Group members

CIPF

Genetics and Genomics of neuromuscular and neurodegenerative diseases

Group Leader

Carmen Espinós Armero

Researchers

M. Dolores Martínez-Rubio, Ana Sánchez-Monteagudo, Paula Sancho, Amparo Andrés-Bordería, Candela Machuca, Isabel Hinarejos, Isabel Bustos, Claudia Vázquez-Calvo.

Retina Degeneration

Group Leader

Dunja Lukovic

Researchers

Verónica del Buey

Bioinformatics and Biostatistics Unit (UBB)

Group Leader

Francisco García-García

Researchers

Marta R Hidalgo García, Sandra Alandes Esteve.

Stem Cell Therapies in Neurodegenerative Diseases

Group Leader

Slaven Erceg

Researchers

Ana Artero, Ovsanna Kepenekyan, M^a Amparo Pérez, Francisco Javier Rodríguez, Regina Rodrigo, Lorena Olivares, Manuel Pérez.

TFGs, TFM's and Doctoral Thesis

Maria Isabel Hinarejos Martínez TFG (2017). Co-directed by Vincenzo Lupo and Rafael Pascual Vázquez Manrique.

Sandra Alandes Doctoral Thesis. Co-Directors Francisco García García and José M. Millán. (Starts on Dec 2018).

Candela Machuca Doctoral Thesis. Co-Directors Slaven Erceg and Carmen Espinós. (Starts on July 2017).

IISLAFE

Group Leader

José M. Millán

Researchers

Elena Aller, Teresa Jaijo, Gema García, Rafael Vazquez, Carla Fuster, Ana Rodríguez, Ana Pilar Gómez, Belén García, M^a Dolores Sequeda, José Bono.

Group Leader

Marina Berenguer

Researchers

Eva Silgo, Ângela Carvalho

Group Leader

Juan J. Vilchez

Researchers

Teresa Sevilla, Luis Bataller, Nuria Muelas, Elvira Millet, Victoria Cortés, Inmaculada Azorín, Maria Pilar Martí, Juan Francisco Vázquez, Marina Frasset, Herminia Argente, Javier Poyatos, Roger Vilchez, María Isabel Nieto, Paula Lizandra, Yaiza Dubón

Figures

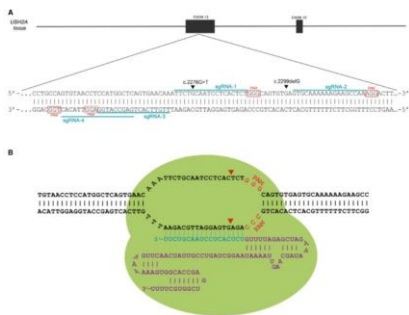


Figure 1. Diagram Showing the CRISPR Construct Designs for the USH2A Locus Editing.

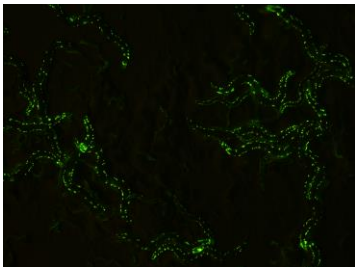


Figure 3. Polyglutamine diseases worm model that express 40 CAG repeats in frame with a fluorescent protein (YFP) in muscle cells. Long Repeats produce a polyQ aggregation pattern progressive and age-dependent manner.

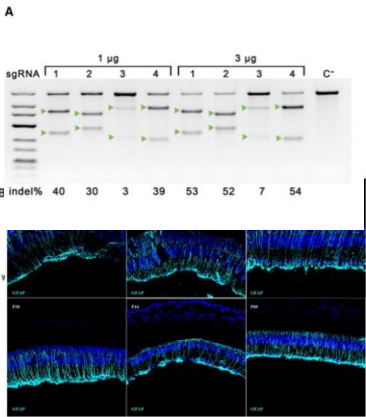


Figure 2. CRISPR Assay on HEK293 Cells. (A) Products of the T7E1 assay resolved on a 2% agarose gel. (B) Chromatograms showing the result of Sanger sequencing of PCR products obtained after amplifying genomic DNA, and subcloned into a plasmid for E. coli transformation.

Figure 4. Progression of reactive gliosis in rd10 retinas. Representative photomicrographs of retinal sections showing GFAP (Müller cell activation) content in DAPI-counterstained sections at different postnatal days of retinas of rd10 mice. At least, six retinas were used per group and age.

Congress Communications

Alandes Esteve S, García-García F, García-García G, Millán JM. JBI-2018. **Application of genomics for a precision medicine in hereditary dystrophies of the retina: Usher syndrome.** XIV Symposium on Bioinformatics). Granada, 14-16 November 2018.

Sancho P, Bartesaghi L, Miossec O, García-García F, Ramírez-Jiménez L, Lupo V, Chrast R, Espinós C (2018). **Characterization of neuronal molecular mechanisms underlying CMT2Z neuropathy.** 15th International Congress on Neuromuscular Diseases (ICNMD). Vienna (Austria).

Committees, Working Groups and Alliances

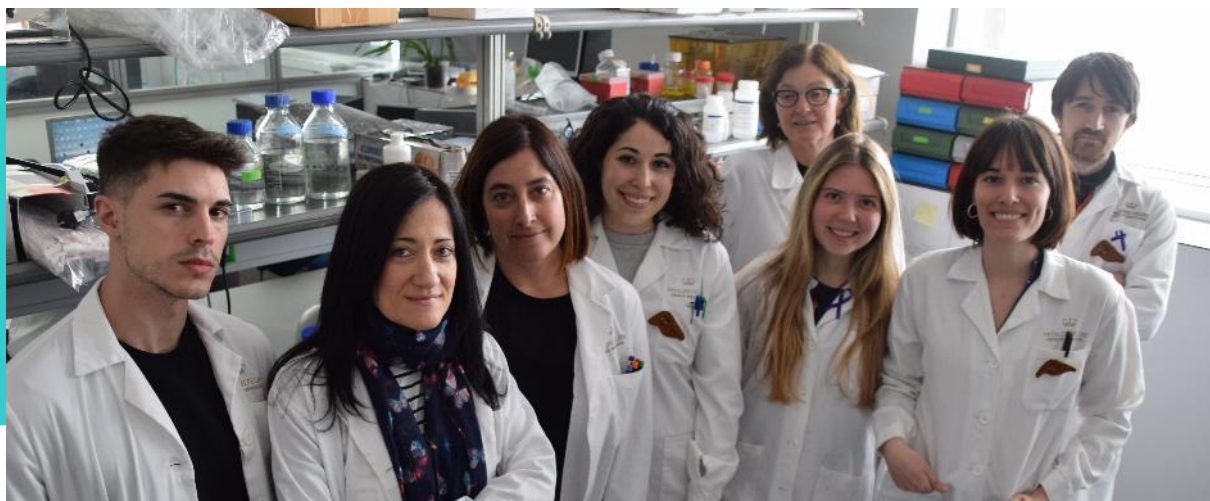
Carmen Espinós and José M. Millán are members of the Scientific Committee of the *Alianza para la investigación traslacional en Enfermedades raras de la Comunidad Valenciana*.

Publications

Machuca-Arellano M, Vilches A, Clemente E, Pascual-Pascual SI, Bolinches-Amorós A, Artero-Castro A, Espinós C, León-Rodríguez M, Jendelova P, Erceg S. **Generation of a human iPSC line from a patient with autosomal recessive spastic ataxia de Charlevoix-Saguenay (ARSACS) caused by a mutation in SACSIN gene.** Stem Cell Res 2018; 31: 249-252.

Machuca-Arellano C, Vilches A, Clemente E, Pascual-Pascual SI, Bolinches-Amorós A, Artero-Castro A, Espinós C, León-Rodríguez M, Jendelova P, Erceg S. **Generation of human induced pluripotent stem cell (iPSC) line from an unaffected female carrier of mutation in SACSIN gene.** Stem Cell Res 2018; 33: 166-170.

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Molecular Endocrinology

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Overview

The mission of our laboratory is to define the roles of insulin and IGF-I receptor signal transduction in the regulation of various physiological systems, with the overall goal of understanding how defects in signal transduction may provoke human diseases such as obesity, diabetes, liver disease, and neurodegeneration. Our research focuses on insulin receptor substrate (IRS) proteins which are the major intracellular targets of the activated insulin receptor. Loss of *Irs2* in both mice and humans is associated with a reduced mass of pancreatic beta cells and peripheral insulin resistance, hallmarks of Type 2 diabetes. Interestingly, the *Irs2*-knockout model has also provided evidence that this molecule plays a key role in brain development and function; *Irs2* deficiency causes a developmental reduction in brain size. Moreover, we have observed that in adult brains the lack of *IRS2* leads to an accumulation of phosphorylated tau, a classic feature of the neurofibrillary tangles associated with Alzheimer's disease, consistent with the hypothesis that defects in insulin action may contribute to neurodegenerative disorders. Our goal is to utilize information from basic analysis of insulin signaling to understand metabolic diseases. The development of new therapeutic strategies for preventing and treating obesity and diabetes is a public health priority.

Research Results

Role of *IRS2* in Liver Regeneration

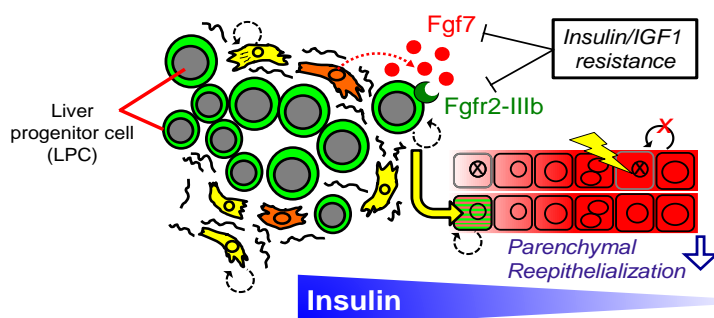
Insulin resistance is a systemic state of reduced insulin sensitivity and the hallmark of metabolic disease, which affects around 1 in 3 Spanish adults over the age of 35 years. We use the liver as a model organ in which to study the negative impact of insulin resistance on tissue repair. The liver is a highly regenerative organ at the forefront of insulin's metabolic actions. Insulin resistance is associated with progression of Nonalcoholic Fatty Liver Disease (NAFLD), which affects around 25.8% of the Spanish population and can lead to liver scarring/cirrhosis and liver cancer. We model how insulin resistance influences the adaptive response to injury by genetic deletion or silencing of insulin receptor substrate 2 (*IRS2*) – an

adaptor molecule responsible for the transmission of insulin and IGF-1 receptor signalling. We are particularly interested in how bipotent liver progenitor cells (LPCs), the facultative hepatic stem cells that emerge in response to chronic damage, are influenced by IRS2 deletion, and, how local cellular communication with the fibrotic microenvironment is affected.

To better understand how stem cells respond to metabolic growth factors, we have developed a range of new molecular tools that enable us to visualize and study key aspects of insulin/IGF1 signalling at the cellular level. We are using these tools, in combination with the CIPF's world-class cytometry services and high-content image analysis platforms to discover new aspects of how insulin/IGF1 signalling and insulin resistance can affect the stem cell's ability to survive, proliferate and differentiate in response to signals from their fibrotic microenvironment. We are also studying how "stem-like" cells in the context of HCC respond to insulin/IGF1 and how the spatial patterning of insulin/IGF1 sensitivity may underlie the heterogeneity of this incurable neoplasm by influencing cell fate, drug resistance, tumour growth and cell survival.

Generation of IRS2 Reporter Mouse Model

Deletion of *Irs2* in mice causes diabetes owing to a reduced beta cell mass and peripheral insulin resistance. IRS2 signals are required for beta cell compensation under conditions of metabolic stress such as high fat feeding. However, the precise mechanisms by which IRS2 signaling regulates the genetic program of beta cell development and adaptive proliferation remain unknown. To investigate the role of IRS-2 signaling in pancreatic progenitor cells, we have developed a multi-disciplinary approach that centers around a novel reporter mouse model. We are using this model to define the spatial and temporal expression of IRS2 during stages of embryonic pancreas development and during the normal ageing of adult mice using this new reporter model where GFP and luciferase are driven by the *Irs2* promoter. This tool will also enable us to isolate specific populations of pancreatic progenitors which express *Irs2* in order to analyse genetic networks that regulate insulin/IGF1-mediated beta cell proliferation and survival in adults. Completion of these aims will not only improve our understanding of how IRS2 signaling regulates beta cell development and survival but will provide new insights into the etiology of diabetes and may identify new markers for the early diagnosis and treatment of beta cell failure. IRS2 represents a rationale target for protecting existing beta cells in the adult pancreas or for engineering robust human insulin-producing cells from pluripotent stem cells.



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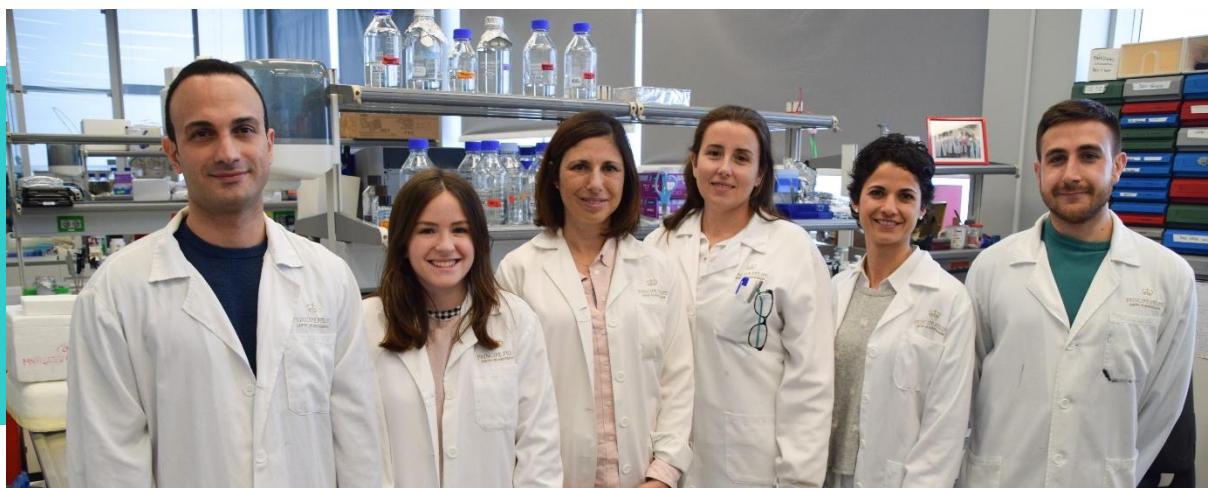
Changes in CDKN2A/2B expression associate with T-cell phenotype modulation in atherosclerosis and type 2 diabetes mellitus. Vinue A, Martínez-Hervás S, Herrero-Cervera A, Sánchez-García V, Andrés-Blasco I, Piqueras L, Sanz MJ, Real JT, Ascaso JF, Burks DJ, González-Navarro H. *Transl Res.* 2019 Jan;203:31-48. doi: 10.1016/j.trsl.2018.08.003. Epub 2018 Aug 15.

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Oncogenic Signalling

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Overview

Cancer is, by its incidence and mortality, a public health problem of enormous magnitude. Elucidation of the molecular mechanisms underlying tumour development remains an enormous challenge for basic research, as well as representing an essential step in the development of new drugs. Also, a major challenge of modern medicine is the development of personalized cancer treatment therapies. Research in the laboratory is focused on the molecular mechanisms that control cancer progression and metastasis. We study the molecular interaction between cell signalling and protein homeostasis on cell proliferation and differentiation, and how dysregulations of these signalling pathways and protein abundance are causes for pathophysiological processes and regulate cancer cell progression and metastasis. The experimental approach developed in our laboratory is based on the integration of cellular/molecular biology, mouse models and patient samples analysis. Specifically, the laboratory exploits the potential for genetic and pharmacological manipulation of 2D and 3D (organoids) cellular cancer models, and in patient-derived xenograft (PDX), and exploits publicly available cancer patient databases to complement information with the primary results of patients obtained in collaboration with local hospitals.

Research Results

1. Molecular mechanisms and signaling pathways involved in the epithelial-mesenchymal transition and cancer stem cells.

The epithelial-mesenchymal (EMT) transition is an evolutionarily conserved fundamental development process in which epithelial cells undergo the conversion to motile mesenchymal cells. EMT is essential for embryonic development and is also involved in tissue repair, organ fibrosis and cancer progression. During EMT carcinoma cells lose their epithelial characteristics and acquire invasive properties. The expression of biological markers for cell epithelial integrity, such as E-cadherin, is repressed, thus stabilizing the loss of epithelial junctions during EMT, with subsequent activation of β -catenin signaling. On the other hand, the

activation of transcription factors of the SNAIL, ZEB or AP-1 families potentiate the repression of epithelial markers and activate the transcription of genes associated with the mesenchymal phenotype such as N-cadherin, vitronectin, fibronectin, vimentin, etc. Recently, the translation factor eIF5A2 has been included in the network of positive regulatory elements that activate EMT, with defined oncogenic functions.

We have focused in the regulation and function of transcription and translation factors that induce epithelial cells to enter into the mesenchymal/stem-cell in lung cancer:

The AP-1 transcription factor consists of a family of homo- and heterodimers of bZip proteins including, among others, the Jun family members (c-Jun, JunB and JunD). We have performed chromatin immunoprecipitation experiments followed by massively parallel DNA sequencing (ChIP-seq) to identify new JunB target genes. The results have revealed new direct transcriptional targets of JunB involved in cell proliferation and differentiation processes. The regulation of these genes mediated by JunB represents a novel mechanism by which JunB can contribute to cancer progression.

eIF5A is a highly evolutionary conserved specific translation factor, and it is the only known protein that undergoes a post-translational modification that generates the hypusine residue, which is necessary for its activity. Overexpression of eIF5A2 isoform is frequently observed in cancer suggesting that aberrant expression of eIF5A2 may be responsible for the malignant behavior of cancer cells. We have shown that genetic inactivation of eIF5A2 in lung cancer cells results in a decrease of vimentin and an increase of E-cadherin expression, indicating that cells adopt a more epithelial phenotype. In addition, inactivation of eIF5A2 factor impairs actin cytoskeleton organization and migration of lung cancer cells. Taken these results into account, together with the highly selective nature of the hypusine modification and its susceptibility to pharmacological inhibition, makes eIF5A2 a very attractive therapeutic cancer target.

2. Development of lung cancer models to study tumor heterogeneity and plasticity.

Despite rapid advances in drug development and surgical procedures, lung cancer remains the leading cause of cancer-related death worldwide. The overall 5-year survival rate is approximately 15%. Surgery is still considered the best option for the treatment of non-small cell lung cancer (NSCLC), however, lung cancer is usually diagnosed at an advanced stage, and less than 25% of NSCLC patients are considered candidates for surgical therapy. Chemotherapy is another important therapeutic strategy for the treatment of cancer, but it cannot eliminate all tumor cells due to drug resistance. In recent years, genetic mutations have been identified in lung tumors and therefore drug-targeted therapy has been developed directed against some of them. However, even providing individualized therapies, acquisition of resistance to treatment is one of the main causes of the high mortality of lung cancer. Cancer stem cells (CSCs) represent a subpopulation of cells distinguishable from the majority of the tumor for its unique ability to drive tumorigenesis and metastasis. They play a crucial role in relapse due to their inherent resistance to current therapies. In our laboratory we investigate the biology of CSCs and analyze whether these specific cells in each tumor dictate the response to treatment. In particular, our research has focused on: i) isolation and characterization of CSCs from lung human tumor biopsies; ii) identification of activated signaling pathways in these cells; (iii) identification of new biomarkers for CSCs; iv) in vivo studies of the biology of CSCs in mouse xenograft models; and v) the search for new therapies targeted against CSCs.

To investigate lung tumor biology, in collaboration with Hospital Universitari i Politècnic La Fe and Hospital Universitario de La Ribera, we are developing lung cancer models which can adequately represent tumor heterogeneity and predict in vivo drug sensitivity. Specifically, we are developing Patient-derived Xenograft Models (PDX) and lung cancer organoids. These cancer models retain largely the histological and genetic characteristics of the original tumor. The fact of amplifying the patient's tumor tissue in several mice and analyzing it at different times, relatively quickly, allows a better understanding of the molecular changes that drive metastasis and resistance to therapies. Therefore, this model can be used for testing the efficacy of compounds with antitumor activity, to guide in the design of a personalized treatment, or for the identification of new biomarkers.

Figures

Figure 1 Proposed role of eIF5A isoforms in cancer. eIF5A isoforms are involved in multiple cellular functions including translation initiation, mRNA decay, cell cycle progression, cell survival, retroviral infection and translation elongation. eIF5A undergoes post-translational modifications that regulate its subcellular localization. eIF5A protein is hypusinated and converted into a mature and active form in the nucleus, and then exported to the cytoplasm. Acetylation of eIF5A might accumulate in the nucleus. The mature eIF5A protein, eIF5A(Hyp), may influence translation of a subset of mRNAs encoding onco-proteins, tumor suppressors, apoptosis and autophagy regulators, leading to the development of tumour cell invasion and metastasis.

Functional crosstalk of post-translational modifications on eIF5A

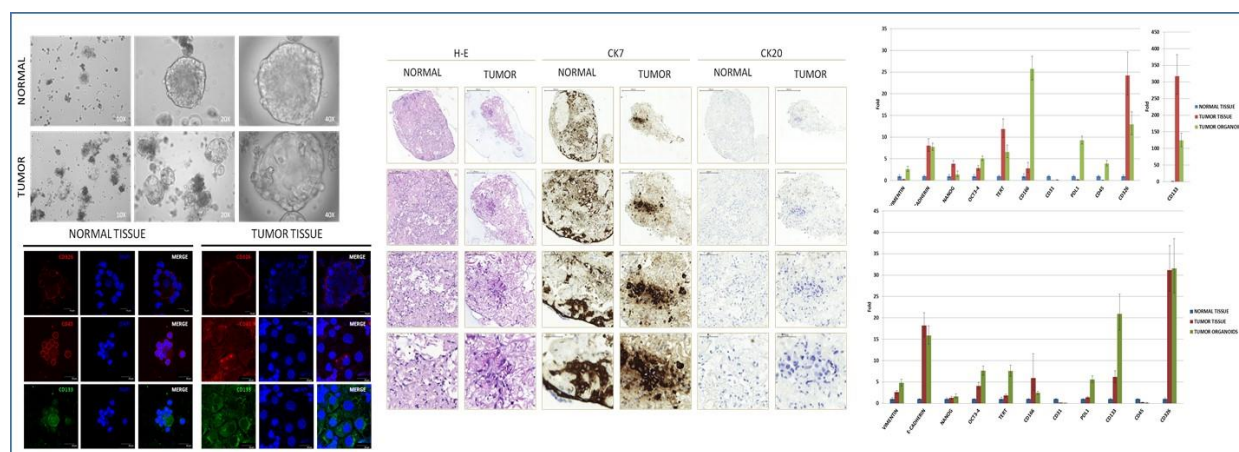
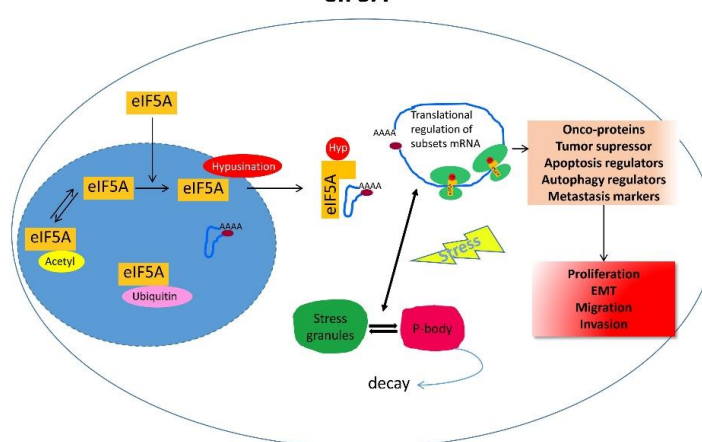


Figure 2 Patient-derived lung tumour organoids. (top panel) Optical microscopy images of patient-derived lung tumour organoids and their adjacent healthy lung tissue acquired with progressive magnification (10x, 20x, 40x). (lower panel) Fluorescence immunocytochemistry of CD326, CD45 and CD133 antibodies in organoids derived from lung adenocarcinoma. CD326+ shows the epithelial origin of the tumor. CD45+ cells are tumor-infiltrating lymphocytes (TIL). CD133 is a cancer stem cell (CSC) marker. (central panel) Comparative histological and immunohistochemical images of lung adenocarcinoma derived organoids and normal lung tissue derived organoids. (right panel) RNA expression of indifferentiation, inflammation, and cancer stem cells markers.

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Unit 1. Pathophysiology of Rare Diseases. Dr. Ibo Galindo.

Dr. Ibo Galindo's group (IDM-UPV) works on the generation of disease models in *Drosophila melanogaster*. In the past they have characterized a model of Charcot-Marie-Tooth (CMT) neuropathy caused by mutations in the GDAP1 gene and have described the alteration of energy metabolism as one of the possible mechanisms of disease. The group of Espinós (CIPF) is focused on the genetics of rare neuronal diseases, and characterized the gene JUNCTOPHIIN-1 (JPH1) as a genetic modifier of GDAP1. A collaboration between both groups has shown that the *Drosophila* junctophilin gene is equivalent to the 4 human junctophilins (JPH1-4) and presents neural, muscular and cardiac phenotypes. In addition, they have shown that this gene is a modifier of the polyglutamine expansions that cause Huntington's disease and spinocerebellar ataxia type 3. At present, both groups continue to collaborate to demonstrate that the expression levels of JPH1 are important for its effect as a modifier; and that in addition to GDAP1 they can modify the phenotype of mutations in another gene that causes CMT, MITOFUSIN-2.

The group is also developing models for Dravet syndrome, a very low prevalence infantile epilepsy caused by dominant mutations in the SCN1A gene, which codes for a voltage-gated sodium channel. It is a serious pathology that produces frequent epileptic seizures, a cognitive and behavioral deterioration, and can even cause the death of the infant. The urgency in finding new treatments is complicated by the fact that this is also a very heterogeneous disease: no two patients are equal in terms of the manifestation of the disease nor in terms of the response to commonly used anticonvulsant medications. Our goal is to reproduce the mutations of patients in *Drosophila*, and use this organism to develop new treatments. To do this, we eliminate the *Drosophila* gene that codes for this sodium channel, and instead we introduce the human gene in which we have introduced the clinical mutations by directed mutagenesis. So far we have generated the mutation of the *Drosophila* gene; this mutant is lethal in homozygosis and has a convulsive phenotype in heterozygosis, as in the case of clinical mutations. At this time we are generating the mutant cDNAs for insertion in the corresponding locus in the fly. To carry out this project, we have the support and sponsorship of the patient association ApoyoDravet.

Projects and contracts

De genes a tratamientos en enfermedades raras neurodegenerativas y neuromusculares. (PROMETEO/2018/135). Programa Prometeo para grupos de investigación de excelencia, Conselleria d'Educació, Investigació, Cultura i Esport de la Generalitat Valenciana. Federico Pallardó (Coordinator, UVEG), Carmen Espinós (CIPF),

Máximo Ibo Galindo (UPV), José María Millán (IIS-La Fe), Teresa Sevilla (IIS-La FE), Pascual Sanz (IBV-CSIC)

Generación de modelos en *Drosophila melanogaster* mediante knock-in de mutaciones de pacientes. Asociación ApoyoDravet. Máximo Ibo Galindo (UPV).

Utilización de *Drosophila melanogaster* como sistema de cribado. Fase II. Contract with Biomar Microbial

Technologies, S.A. Máximo Ibo Galindo (UPV), Mar Orzaez (CIPF)

Unit 2. Pathophysiology of Rare Diseases. Dr. Ramón Martínez.

The aim of our unit is to address the disease mechanisms with the ultimate goal of developing new treatments from a multidisciplinary point of view and apply the latest advances in nanomedicine to solve health problems. Our lines of research include the following:

Lectin-gated and glycan functionalized mesoporous silica nanocontainers for targeting cancer cells over expressing Lewis X antigen

The Joint Unit (UPV-CIPF) has been developing the design of gating systems that actively target carbohydrate from membrane receptors of cancer cells. The Lewis X (Lex) antigen-targeted delivery system is based on mesoporous silica nanoparticles (MSNs) loaded with ATTO 430LS dye, functionalized with a Lex derivative and capped with a fucose-specific carbohydrate-binding protein (Aleuria aurantia lectin (AAL)). This design takes advantage of the affinity of AAL for Lex overexpressed receptors in certain cancer cells.

Smart gated magnetic silica mesoporous particles for targeted colon drug delivery.

Inflammatory bowel diseases (IBD) are autoimmune, inflammatory, chronic malignances affecting the gastrointestinal tract tissue, mainly the colon, being ulcerative colitis and Crohn's disease the most important. Therefore, the Joint unit (UPV-CIPF) have carried out a research in order to design an oral colon drug delivery device. As a result of the research a nano and microdevice were developed and its efficacy in the treatment of Inflammatory bowel diseases was evaluated in vivo in a TNBS colitis induced rat model. The nano and microdevices consisted on magnetic mesoporous silica particles loaded with safranin O or with hydrocortisone and functionalized in the external surface with bulky azo derivatives. The developed systems can be used to specifically deliver drugs or other agents in the last part of intestine while decreasing the systemic absorption; both effects contribute to increase efficacy for the treatment of inflammatory bowel disease and to reduce adverse effects of drugs.

Design of oligonucleotide-capped mesoporous silica nanoparticles for the detection of miRNA-145 by duplex and triplex formation.

The development of new strategies to detect microRNAs (miRNAs) has become an important challenge in the biomedical field. The Joint Unit (UPV-CIPF) have been working in the design of oligonucleotide-gated silica nanoparticles for the detection of miRNA-145. The design consists of mesoporous silica nanoparticles loaded with a fluorescent reporter and capped with specific DNA oligonucleotide which acts as a molecular gate blocking the pores. The opening of the gated system and fluorescent reporter delivery is selectively controlled by DNA-miRNA recognition.

Mesoporous Bioactive Glasses Equipped with Stimuli-Responsive Molecular Gates for Controlled Delivery of Levofloxacin against Bacteria.

Throughout 2018, the Joint Unit (UPV-CIPF) focused part of its research in the development of mesoporous bioactive glasses equipped with stimuli-responsive gates for controlled delivery of drugs as an option to fight against bone infection, which is one of the most common problems regarding bone regeneration therapies. The mesoporous bioglass was functionalized with polyamines and capped with adenosine triphosphate (ATP) as the molecular gate for the controlled release of the antibiotic levofloxacin. The solid was characterized and tested successfully against bacteria.

Gold Nanostars Coated with Mesoporous Silica Are Effective and Nontoxic Photothermal Agents Capable of Gate Keeping and Laser Induced Drug Release.

The collaborative research activities carried out into the Joint Unit have allowed the design and development of a novel drug photorelease system based on gold nanostars (AuNSts) coated with a Mesoporous Silica shell and capped with paraffin as a suitable thermosensitive molecular gate. Dox-loaded nanoparticles showed no cytotoxicity toward HeLa cells, until they were irradiated with 808 nm laser, provoking paraffin melting and drug release. This system can be of general use for the fabrication of new drug photodelivery systems containing AuNSts.

Targeted delivery of inflammation inhibitors by Mesoporous Nanocarriers in acute lung injury.

Acute lung injury (ALI) is a severe pulmonary disorder with high percentages of mortality and morbidity in intensive care units. In the context of the Joint Unit (UPV-CIPF) the specific delivery of anti-

inflammatory drugs to the lungs is being analysed in cellular and in vivo models, both using drugs approved for clinical use and new drugs developed at the CIPF.

Screening of a combinatorial hexapeptide library for the identification of new senolytic agents

Senescence is a state of permanent cell cycle arrest. Despite its physiological role in tissue remodelling, organism development and tumour growth control, accumulation of senescent cells causes tissue degeneration linked to aging and contributes to the development of numerous age-related diseases such as arteriosclerosis, osteoarthritis, cancer or neurodegenerative diseases. In contrast, drug-induced senescence has recently been approved for the treatment of specific types of tumours. However, among the side effects of these drugs, the accumulation of senescent cells in the organism produces negative consequences for patients, including aging related diseases and the appearance of new metastases. The specific elimination of these cells by a mechanism known as senolysis, contributes positively to health improvement. That is why the development of molecules capable of selectively eliminating these senescent cells has become a very active field of research.

In the context of the Joint Unit (UPV-CIPF) the screening of a hexapeptide combinatorial library has led to the generation of two new senolytic peptides and to the identification of a new molecular target. Patentability studies are ongoing.

Evaluation of the antitumoral activity of a senolytic nanoparticle in a triple negative breast cancer model

High mortality and lack of appropriate treatment for triple-negative breast cancer, together with the expectative opened by the latest clinical trials of senescence-inducing activity in this type of patients, have aroused the interest in the study of combination therapies of senescence induction as antitumor treatment plus selective elimination of those senescent cells to avoid negative side-effects.

In the context of the Joint Unit (UPV-CIPF) we have set up a senescence induction model in immunocompetent triple negative breast cancer mice and evaluated the activity of a senolytic mesoporous nanoparticle and different nanoprobe for in vivo detection of senescence.

Figures

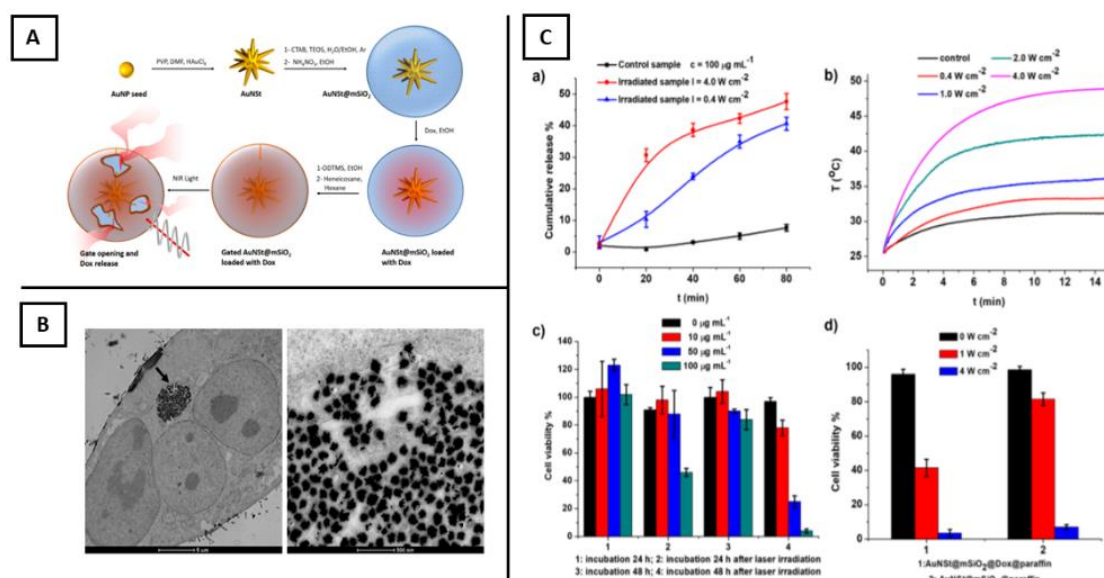
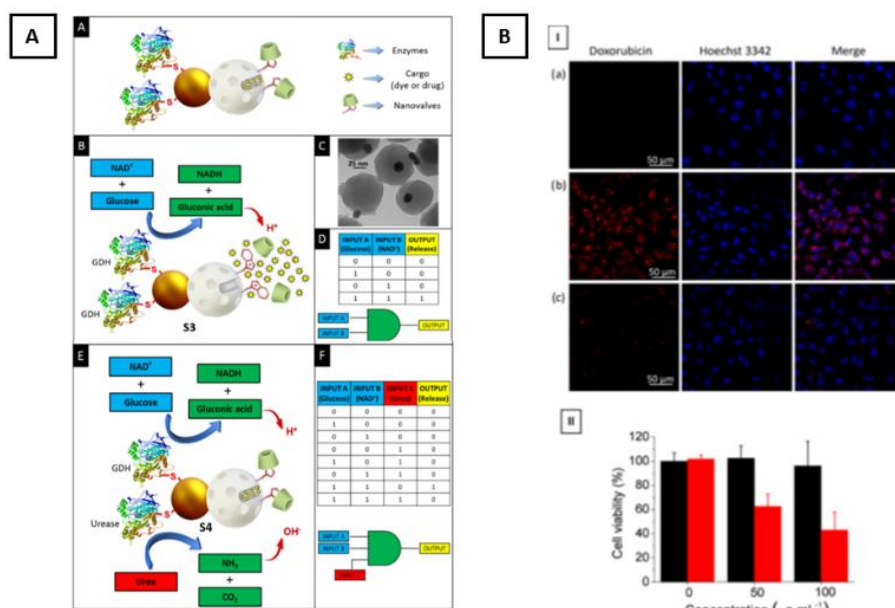


Figure 1. A. Representation of the drug photorelease system AuNSt@mSiO₂@Dox@paraffin based on AuNSts coated with a mesoporous silica shell and paraffin as a thermosensitive molecular gate. The delivery of the entrapped cargo (Dox) is triggered by NIR laser irradiation. B. TEM pictures of HeLa cells after incubation with AuNSt@mSiO₂@Dox@paraffin nanoparticles. C. (a) Cumulative release of Dox from AuNSt@mSiO₂@Dox@paraffin aqueous suspensions and (b) NIR-light-induced heating of bulk nanoparticle suspensions at different laser power densities. (c) Viability of HeLa cells in the presence of AuNSt@mSiO₂@Dox@paraffin nanoparticles. (d) Cell viability of HeLa cells in the presence of AuNSt@mSiO₂@Dox@paraffin and AuNSt@mSiO₂@paraffin nanoparticles upon laser irradiation (808 nm) at different laser power densities during 10 min.



microscopy images of HeLa cells showing controlled doxorubicin release from nanodevice S3DOX. (a) HeLa cells with no treatment; (b) HeLa cells treated with S3DOX in a medium containing glucose; (c) HeLa cells treated with S3DOX in the absence of glucose. From left to right: doxorubicin fluorescence, DNA marker (Hoechst 3342) fluorescence, and combined (merge). Bottom (II): cell viability assays of HeLa cells treated with different S3DOX concentrations (0, 50, and 100 $\mu\text{g mL}^{-1}$) in the absence (black bars) or presence (red bars) of glucose (25 mM).

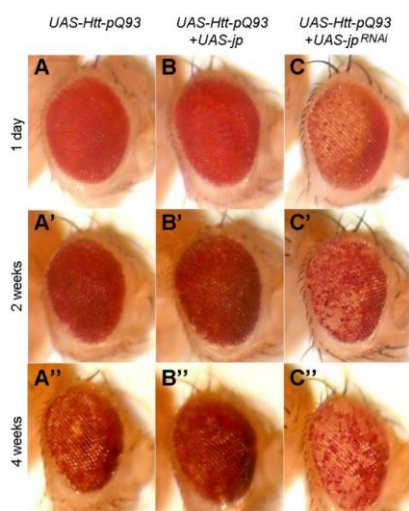


Figure 3. Jp levels modify the phenotype of a pathological Htt poly-Q expansion. (A–A”) Progressive degeneration and depigmentation over 4 weeks upon expression of Htt-pQ93 in the eye driven by GMR-Gal4. (B–B”) Co-expression of Jp ameliorates the depigmentation. (C–C”) Co-expression of Jp RNAi induces earlier and faster progressing depigmentation.

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Physiopathology of Rare Diseases CIPF-INCLIVA Joint Research Unit

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INCLIVA

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Overview

The main objectives that we are currently addressing in our CIPF-INCLIVA research unit include both the genetic and epigenetic regulation and also the physiopathological mechanisms of rare diseases. The main goal of our unit is to improve the diagnostic and therapeutic tools in order to ameliorate the quality of life of those patients suffering these low prevalence pathologies.

In 2018 our main achievement was to obtain a Prometeo research grant (GVPROMETEO2018-135) entitled: “From genes to therapy in neuromuscular diseases” principal investigator was Federico Pallardó (INCLIVA-UV) with members of the research team, Carmen Espinós and Máximo Ibo Galindo (CIPF). Other members of the research Project are Teresa Sevilla and JM Millán from IIS La Fe and Pascual Sanz (CSIC-IBV). In the framework of this Project several talks and meeting communications have been organized. Also, the CIPF-INCLIVA mixt unit has organized meetings on rare diseases and postgraduate courses. There is web page in process of construction (<https://gt2n.es/>).



Oncology Research CIPF-UCV Joint Research Unit

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CIPF

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Overview

During 2018 this Unit has maintained its activity in three fundamental scenarios:

1. Histological, immunohistochemical and molecular techniques, available to the Researchers of the CIPF and the UCV. The interpretation and discussion of the results has produced a very close collaboration, with a series of researchers, and has favored multidisciplinary and team work.
2. We have served as a Reference Center for clinicopathological consultations and incorporate new techniques in specific fields such as: Hematopathology (FHIS, rearrangements); Nephropathology (Ultrastructure); Neuropathology (Ultrastructure).
3. We have collaborated in the area of teaching Biomedicine, giving practical teaching in the laboratory to students of different degrees of the UCV, especially medical students. Additionally, we have collaborated in the technical assistance and supervision of doctoral works.

Research results

The fundamental diseases in which we have planned a molecular study is Rosai-Dorman disease and Hodgkin lymphoma. This last disease is a model to study the body's reaction to neoplasia. This model may be a paradigm in the immunobiology of the treatment of neoplasms and may be mutations in the K-ras pathway, the basis of the onset and progression of this disease. On the other hand, the Rosai-Dorman disease can be a good model to study the importance it can have in the hematological neoplasms, the alterations of the autophagy and the emperipolesis.

Publications

A CASE REPORT OF LIPOMA-LIKE HIBERNOMA IN AXILLA. A RARELY BENIGN TUMOR OF BROWN ADIPOSE TISSUE. Ricardo Rubini Costa¹, Antonio Torregrosa Gallud², José Miguel Rayón³, Jerónimo Forteza Vila⁴ Case Studies in Surgery 4, (1-5) 2018 DOI: 10.5430/css.v4n1p1

AN IMMUNOHISTOCHEMICAL SCORE TO PREDICT THE OUTCOME FOR ORAL SQUAMOUS CELL CARCINOMA. Monteiro LS¹, Diniz-Freitas M², Warnakulasuriya S^{3,4}, Garcia-Caballero T⁵, Forteza J⁶, Fraga M⁷ DOI: 10.1111/jop.12682 J ORAL PATHOL MED 47, (175-381) 2018.

COMBINED PLASMA RICH IN GROWTH FACTORS AND ADIPOSED-DERIVED MESENCHYMAL STEM CELLS REMOTES THE CUTANEOUS WOUND HEALING IN RABBITS. Deborah Chicharro¹, Jose M. Carrillo¹, Mónica Rubio¹, Ramón Cugat², Belén Cuervo¹, Silvia Guil³, Jerónimo Forteza⁴, Victoria Moreno⁵, Jose M. Vilar^{6*} and Joaquín Sopena¹ Chicharro et al. BMC Veterinary Research (2018) 14:288 doi: 10.1186/s12917-018-1577-y

HISTOLOGICAL FEATURES OF CEREBELLAR NEUROPATHOLOGY IN PATIENTS WITH ALCOHOLIC AND NONALCOHOLIC STEATOHEPATITIS. Tiziano Balzano, PhD, Jerónimo Forteza, MD, PhD, Irene Borreda, BS, Pilar Molina, MD, PhD, Juan Giner, MD, PhD, Paola Leone, MS, Amparo Urios, PhD, Carmina Montoliu, PhD, and Vicente Felipo, PhD. J Neuropathol Exp Neurol (1-9) 2018 DOI: 10.1093/jnen/nly061

IMBALANCE OF IMMUNOLOGICAL SYNAPSE-KINAPSE STATES REFLECTS TUMOR ESCAPE TO IMMUNITY IN GLIOBLASTOMA. Laura R. Díaz,^(1,2) Elena Saavedra-López,^(1,2) Leire Romarate,^(1,2) Izaskun Mitxitorena,^(1,2) Paola V. Casanova,^(1,2) George P. Cribaro,^(1,2) José M. Gallego,⁽³⁾ Ana Pérez-Vallés,⁽⁴⁾ Jerónimo Forteza-Vila,⁽⁵⁾ Clara Alfaro-Cervello,⁽⁶⁾ José M. García-Verdugo,⁽⁶⁾ Carlos Barcia Sr.,⁽³⁾ and Carlos Barcia Jr.^(1,2) JCI INSIGHT Published: September 20, 2018 Reference information: JCI Insight. 2018; 3(18): e120757. DOI: 10.1172/jci.insight.120757

INSULIN RESISTANCE DISRUPTS EPITHELIAL REPAIR AND NICHE-PROGENITOR FGF SIGNALING DURING CHRONIC LIVER INJURY Fátima Manzano-Núñez^{1,2}, María José Arámbul-Anthony^{1,2}, Amparo Galán Albiñana², Aranzazu Leal Tassias², Carlos Acosta Umanzor^{1,2}, Irene Borreda Gascó³, Antonio Herrera², Jerónimo Forteza Vila^{2,3}, Deborah J. Burks^{1,2}, Luke A. Noon^{1,2*} PLOS Biology | January 29, 2019 (1-32). DOI: 10.1371/journal.pbio.2006972

SELECTIVE TARGETING OF COLLAGEN IV IN THE CANCER CELL MICROENVIRONMENT REDUCES TUMOR BURDEN. Fernando Revert^{1,2}, Francisco Revert-Ros^{1,2}, Raúl Blasco^{1,3}, Aida Artigot¹, Ernesto López-Pascual^{1,2}, Roberto Gozalbo-Rovira^{1,8}, Ignacio Ventura^{1,2,9}, Elain GutiérrezCarbonell^{1,10}, Nuria Roda⁴, Daniel Ruiz-Sanchis¹, Jerónimo Forteza⁵, Javier Alcácer⁶, Alejandra Pérez-Sastre^{1,2,11}, Ana Díaz⁷, Enrique Pérez-Payá⁴, Juan F. Sanz-Cervera³ and Juan Saus^{1,2,4} www.impactjournals.com/oncotarget/.

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THE CEREBELLUM OF PATIENTS WITH STEATOHEPATITIS SHOWS LUMPHOCYTE INFILTRATION, MICROGLIAL ACTIVATION AND LOSS OF PURKINJE AND GRANULAR NEURONS Tiziano Balzano¹, Jerónimo Forteza², Pilar Molina^{3,4}, Juan Giner³, Ana Monzó³, Jennifer Sancho-Jiménez³, Amparo Urios¹, Carmina Montoliu^{4,5} & Vicente Felipo¹ SCIENTIFIC REPORTS | (2018) 8:3004 | Page 1-12. DOI: 10.1038/s41598-018-21399-6.

TUMOR MICROENVIRONMENT-TARGETED POLY-L-GLUTAMIC ACID-BASED COMBINATION CONJUGATE FOR ENHANCED TRIPLE NEGATIVE BREAST CANCER TREATMENT Juan J. Arroyo-Crespo, Ana Armiñán,*, David Charbonnier, Leandro Balzano-Nogueira, Francisco Huertas-López, Cristina Martí, Sonia Tarazona, Jerónimo Forteza, Ana Conesa, María J. Vicenta,* Biomaterials 186 (2018-21) DOI: 10.1016/j.biomaterials.2018.09.023

RECELLULARIZED ACELLULAR MATRIX VS NON-RECELLULARIZED ACELLULAR MATRIX IN A RAT MODEL FOR IN VIVO NEO-BLADDER REGENERATION Esteve, María José; Mellado-Lopez, Mara; Forteza, Jerónimo; Moreno-Manzano, Victoria; David Vera-Donoso, César JOURNAL OF UROLOGY Volume: 199 Issue: 4 Pages: E1100-E1100 Supplement: S Meeting Abstract: MP81-09 Published: APR 2018



Neurobiology

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Ana Agustí

Overview

The lab performs basic and translational research on cognitive, motor and sleep alterations in different pathological situations, including: minimal and clinical hepatic encephalopathy (HE), hyperammonemia and developmental exposure to food and environmental contaminants. The aims are in animal models: 1. Unveil the molecular mechanisms leading to neurological impairment; 2. Identify new therapeutic targets for its treatment; 3. Design and assess new therapeutic procedures to reverse neurological impairment. In patients: 1. Study the mechanisms, diagnosis and treatment of neurological impairment; 2. Bring to the clinic the therapeutic procedures developed in animal models; 3. Identify early diagnostic procedures for neurological impairment; 4. Bring to the clinic the diagnostic procedures identified.

Our recent results indicate that the process leading to cognitive and motor impairment in HE (Fig. 1) involves: induction by liver failure of hyperammonemia and peripheral inflammation which, acting synergistically induce neuroinflammation, which alters GABAergic and glutamatergic neurotransmission and cGMP levels, leading to cognitive and motor impairment. There are mechanisms by which neuroinflammation, intra- and extra-cellular cGMP and GABAergic and glutamatergic neurotransmission modulate each other. This leads to the neurological alterations, but also offers the possibility of restoring cognitive and motor function by acting on any of the components of the system: cGMP, neurotransmission or neuroinflammation.

The research of the group focuses now in identifying the mechanisms by which: 1. hyperammonemia and peripheral inflammation induce neuroinflammation; 2. neuroinflammation impairs neurotransmission and cognitive and motor function; 3. neuroinflammation, intra- and extra-cellular cGMP and GABAergic and glutamatergic neurotransmission modulate each other and cognitive and motor function. Unveiling these mechanisms (which also occur in other pathologies) is allowing us to identify new targets and assess new therapeutic treatments that restore cognitive and motor function in HE (and could restore them also in other pathologies).

Research Results

The cerebellum of patients with steatohepatitis shows lymphocyte infiltration, microglial activation and loss of Purkinje and granular neurons

Peripheral inflammation contributes to minimal hepatic encephalopathy in chronic liver diseases, including patients with steatohepatitis. Studies in animal models indicate that this would be mediated by neuroinflammation, especially in cerebellum. Infiltration of lymphocytes in brain may induce neuroinflammation. Neuroinflammation in cerebellum of patients with chronic liver diseases has not been studied in detail. Our aim was to analyze by immunohistochemistry in cerebellum of patients with different grades of liver disease: a) neuronal density in Purkinje and granular layers; b) microglial activation; c) astrocytes activation; d) peripheral lymphocytes infiltration; e) subtypes of lymphocytes infiltrated.

Post-mortem cerebellum and liver tissues were collected from patients with different grades of disease, from mild steatohepatitis to cirrhosis and hepatic encephalopathy. Steatohepatitis patients were classified as SH1, SH2 and SH3.

Patients with SH1 show Th17 and Tfh lymphocytes infiltration in the meninges, microglia activation and Bergman glia damage in the molecular layer and a small loss of Purkinje and granular neurons. White matter remains unaffected. At worse stages (SH2, SH3, cirrhosis) activation of microglia and astrocytes is extended also to white matter and there is a further loss of Purkinje neurons.

The results reported support that neuroinflammation in cerebellum occurs at early stages of liver disease, even before reaching cirrhosis, and is associated with infiltration of peripheral Th17 and Tfh lymphocytes.

This study has been published in Balzano et al, (2018). Scientific Reports; 14:8(1):3004

Increasing extracellular cGMP in cerebellum in vivo reduces neuroinflammation, GABAergic tone and motor in-coordination in hyperammonemic rats.

The above study shows that patients with chronic liver disease show neuroinflammation in cerebellum that must contribute to the impairment in motor coordination in these patients. To understand the underlying mechanisms and how to reverse it we performed a study in a rat model: rats with chronic moderate hyperammonemia.

The aims of this work were to assess whether chronic intracerebral administration of cGMP to hyperammonemic rats: 1) restores motor coordination; 2) reduces neuroinflammation in cerebellum; 3) reduces extracellular GABA levels and GABAergic tone in cerebellum; and also 4) to provide some advance in the understanding on the molecular mechanisms involved.

The results show that rats with chronic hyperammonemia show neuroinflammation in cerebellum, including microglia and astrocytes activation and increased levels of IL-1b and TNFα and increased membrane expression of the TNFα receptor. This is associated with increased glutaminase expression and extracellular glutamate, increased amount of the GABA transporter GAT-3 in activated astrocytes, increased extracellular GABA in cerebellum and motor in-coordination. Chronic intracerebral administration of extracellular cGMP to rats with chronic hyperammonemia reduces neuroinflammation, including microglia and astrocytes activation and membrane expression of the TNFα receptor. The data support that extracellular cGMP restores motor coordination in hyperammonemic rats by reducing microglia activation and neuroinflammation, leading to normalization of extracellular glutamate and GABA levels in cerebellum and of motor coordination. This study has been published in: Cabrera-Pastor et al. (2018) Brain Behav Immun.; 69:386-398. (Figure 1)

Hyperammonemia alters membrane expression of GluA1 and GluA2 subunits of AMPA receptors in hippocampus by enhancing activation of the IL-1 receptor.

Rats with chronic hyperammonemia reproduce many of the cognitive alterations of patients with MHE. We used this animal model to analyze the mechanisms responsible for altered functional connectivity in hippocampus. As functional connectivity is mediated by neurotransmission we analyzed the mechanisms by which hyperammonemia alters glutamatergic neurotransmission in hippocampus.

The aims of this work were: 1) assess if increased IL-1b levels and activation of its receptor are responsible for the changes in GluA1 and/or GluA2 membrane expression in hyperammonemia; 2)

identify the mechanisms by which activation of IL-1 receptor leads to altered membrane expression of GluA1 and GluA2.

We identify the pathways by which hyperammonemia alters membrane expression of GluA1 and GluA2. Hyperammonemia increases IL-1 β , enhancing activation of IL-1 receptor. This leads to activation of Src, reflected in increased phosphorylation of Tyr416. These steps are common to the pathways leading to altered membrane expression of the GluA1 and GluA2 subunits. The changes in membrane expression of both GluA1 and GluA2 are reversed by blocking the IL-1 receptor with IL-1Ra or by inhibiting Src with PP2.

After Src activation the pathways for GluA2 and GluA1 diverge. The enhanced activity of Src in hyperammonemic rats increases phosphorylation of GluN2B at Tyr1472 and membrane expression of GluN2B, leading to activation of MAP kinase p38. Activated p38 binds to and reduces phosphorylation at Thr560 and activity of PKC ζ , resulting in reduced phosphorylation at Ser880 and enhanced membrane expression of GluA2.

This work identifies two pathways by which neuroinflammation alters glutamatergic neurotransmission in hippocampus. The steps of the pathways identified could be targets to normalize neurotransmission in hyperammonemia and other pathologies associated with increased IL-1 β by acting, for example, on p38 or PKC δ . This study has been published in Taoro-Gonzalez et al (2018) J Neuroinflammation.; 15(1):36 (Figure 2)

Figures

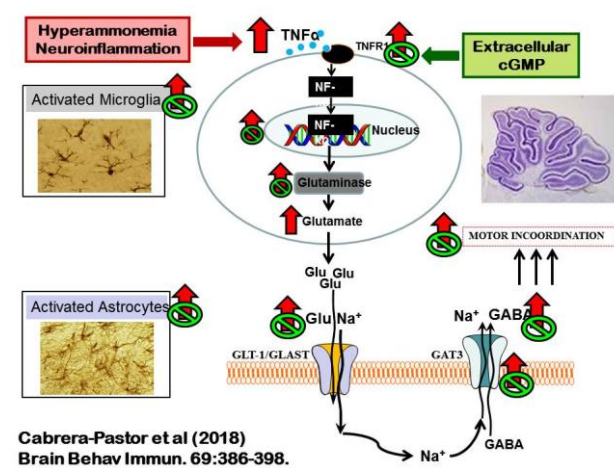


Figure 1. Increasing extracellular cGMP in cerebellum in vivo reduces neuroinflammation, GABAergic tone and motor incoordination in hyperammonemic rats.

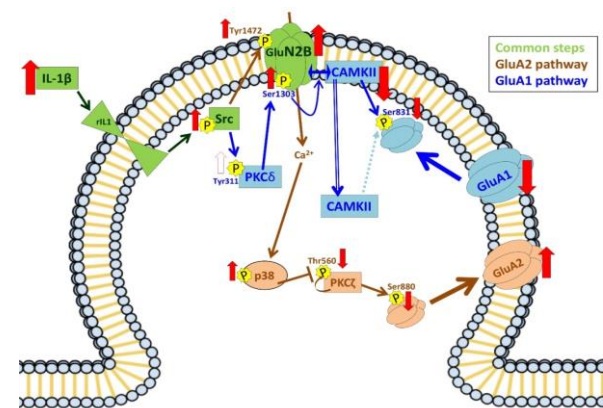


Figure 2. Hyperammonemia alters membrane expression of GluA1 and GluA2 subunits of AMPA receptors in hippocampus by enhancing activation of the IL-1 receptor

Publications

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Doctoral Thesis

Raquel García García. Efectos de la hiperammonemia y la encefalopatía hepática sobre la conectividad neuronal en cerebro de pacientes cirróticos y modelos animales. Implicaciones terapéuticas. Doctorado en Neurociencias. Facultad de Ciencias Biológicas, Universidad de Valencia, 25th October 2018 Sobresaliente cum Laude, Directed by: Carmina Montoliu, Cristina Forn, Vicente Felipe

Lucas Taoro Gonzalez. Mecanismos moleculares del deterioro cognitivo en encefalopatía hepática mínima. Papel de la neuroinflamación en la alteración de la neurotransmisión. Doctorado en Neurociencias. Facultad de Ciencias Biológicas, Universidad de Valencia, 5th November 2018,

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International Committees

Member of the Editorial Board of: *International Journal of Molecular Medicine*,

World Journal of Gastroenterology, *Open Gastroenterology Journal*, *Journal of Hepatology*, *Review Editor de Frontiers in Neuroenergetics*, *World Journal of Experimental Medicine*, *Neuroimmunology and Neuroinflammation*, *New Horizons in Translational Medicine*, *The Open Journal of Medicine*, *International Journal of Neurology Research*, *Journal of Neuroscience and Cognitive Studies*

Member of the Scientific Advisory Board of ACS Chemical Neuroscience, American Chemical Society, since March 2018.

Member of the Action Group A3 on "Prevention and early diagnosis of frailty and functional decline, both physical and cognitive, in older people" European Innovation Partnership on Active and Healthy Ageing (EIP on AHA) European Commission.

Full Member World Academy of Science. 2018. <http://www.worldacademyofsciences.com/members.html>

Management activities

Member of the Academic Commission of the Doctoral Program on Neurosciences of the Valencia University

Training activities

Coordinator of the Section "Technological tools for the diagnosis and treatment of human pathologies" Master on Biomedical Research, Medicine Faculty and Valencia University.

Professor in the following Masters:

"Master on Neurological Diseases" Biotechnology. Universidad Politécnica de Valencia. Vicente Felipe

"Master on Basic and Applied Neurosciences". Universidad de Valencia" Vicente Felipe

"Master on Biomedical Research" Facultad de Medicina, Universidad de Valencia. Marta Llansola, Andrea Cabrera, Tiziano Balzano.



Molecular and Cellular Pathology of Alcohol

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**Olga Valverde
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Overview

We are interested in the molecular and cellular actions of ethanol in the adult and developing brain. Prenatal alcohol exposure is a leading preventable cause of birth defects, mental retardation and neurodevelopmental disorders (FASD) including the fetal alcohol syndrome (FAS). The new pattern of binge-alcohol drinking during adolescence not only induces the neurotoxicity associated with behavioural and cognitive deficits, but also increases the vulnerability to alcohol dependence. Since, alcohol abuse can also induce brain damage and neurodegeneration; we are also interested to investigate the molecular actions of ethanol in the adolescent and adult brain. We use neural cells in primary culture and animal models, which mimic the alterations observed in alcohol-related pathologies.

Research Results

During the last years we have demonstrated the role of the neuro-immune system and TLR4 response in the actions of ethanol on the brain. Indeed, the innate immune response in the central nervous system (CNS) participates in both synaptic plasticity, but over-activation can cause neural damage. Our results have shown that alcohol stimulates brain immune cells, microglia and astrocytes, by activating innate immune receptors TLRs (Toll-like receptors) and (NOD)-like receptors (inflammasome NLRs) triggering signaling pathways, which culminate in the production of pro-inflammatory cytokines and chemokines, causing neuroinflammation and neural damage. Therefore, our aims during this year have been: i) to study the molecular mechanisms of the actions of ethanol on the immune receptors (TLRs, NLRs) in cortical glial primary culture; ii) to assess the impact of the innate immune response and TLRs in the brain damage and behavioral dysfunction induced by alcohol abuse; iii) to investigate new serum biological markers, such as cytokines or chemokines, exosomes, associated with neuroinflammation that could prevent brain damage; iv) to study potential gender differences in alcohol-induced neuroinflammation; v) to evaluate new treatments and/or drugs that could block neuroinflammation and neurodegeneration induced by

ethanol abuse in adult and in adolescents; vi) To assess the potential role of neuroinflammation and TLRs response in the brain developmental dysfunction observed in FASD.

Alcohol-induced neuroinflammation and the TLR4 response are associated with important changes in miRNAs in cortical region: Deep sequencing analysis.

Alcohol abuse can induce brain injury and neurodegeneration. We have demonstrated that ethanol by activating the innate immune receptors, Toll-like (TLRs) and NOD-like, receptors (inflammasome NLRs), causes neuro-inflammation and brain damage. Recent evidence indicates the role of the small non-coding RNA, mRNAs, as important regulators of gene expression, participating in the neuro-immune system response as well as in neurodegenerative and neurological disorders. Therefore, we explore the modulatory role of the miRNAs induced by chronic alcohol intake through the TLR4 response in the brain. We used cortices of WT and TLR4-KO with and without chronic alcohol treatment and the next-generation sequencing (NGS) technique to identify the miRNA profiles that could be differentially expressed.

The bioinformatics pipelines identified several miRNAs and a regulatory pathways network related to the TLR4-response and immune system alterations.

Using SGS we identified miRNAs that were differentially expressed in the chronic alcohol-treated versus untreated WT or TLR4-KO mice. We observed a differential expression of miR-183 Cluster (C) (miR-96/-182/-183), miR-200a and miR-200b, which were down-regulated, while miR-125b was up-regulated in alcohol-treated WT versus (vs.) untreated mice. These miRNAs modulate targets genes related to the voltage-gated sodium channel, neuron hyper-excitability (Nav1.3, Trpv1, Smad3 and PP1-y), as well as genes associated with innate immune TLR4 signaling response (Il1r1, Mapk14, Sirt1, Lrp6 and Bdnf). Our results show the relationship between alcohol intake and miRNAs expression and open up new therapeutically targets to prevent deleterious effects of alcohol on the brain (see Fig.1)

Binge-like ethanol treatment in adolescence impairs autophagy and hinders synaptic maturation: Role of TLR4

Adolescence is a developmental period of brain maturation in which remodeling and changes in synaptic plasticity and neural connectivity take place in some brain regions.

Alcohol is a neurotoxic compound and we have shown that binge-like ethanol treatment in adolescent mice induces synaptic and myelin alterations in the prefrontal cortex and causes long-term cognitive and memory dysfunctions. These events were associated with ethanol-induced activation of the TLR4 response, triggering neuroinflammation, neural damage and behavioral alterations. However, the potential participation of autophagy in long-term neurochemical and cognitive dysfunctions induced by binge ethanol drinking in adolescence is uncertain. Therefore, we evaluated whether binge ethanol drinking alters autophagy pathways by contributing to adolescent synaptic dysfunctions, and if the immune receptor TLR4 response participates in these events.

Using wild-type (WT) and TLR4-deficient (TLR4-KO) we show that binge-like ethanol exposure in adolescence impairs autophagy machinery by increasing autophagy inhibitor mTOR by lowering LC3-II levels and accumulating p62. Inhibition of mTOR, by rapamycin, restores the levels of excitatory scaffolding synaptic proteins (PSD-95 or SHANK3), p62, and partly reestablishes the LC3-II levels in the prefrontal cortices of ethanol-treated WT mice. Elimination of the TLR4 receptors using TLR4-KO mice prevents autophagy dysfunctions and reduces the number or size of the synaptic connections induced by ethanol. These results suggest the role of autophagy dysfunctions in the structural synaptic plasticity alterations induced by binge alcohol in adolescence, and support the participation of the TLR4 response in these events (see Fig.2).

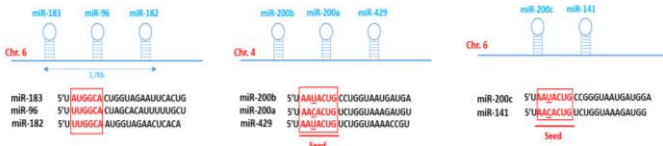
Astrocyte-derived exosomes as inflammatory mediators induced by ethanol: Role of TLR4

Ethanol activates glial cells through Toll-like receptor 4 (TLR4) responses triggering neuroinflammation. Recent evidences indicate the participation of exosomes, tiny cytoplasmic vesicles (30-100 nm), in the intercellular signaling and in the regulation and amplification of neuroinflammation. Here we evaluate the involvement of the exosomes secreted by astroglial cells in ethanol-induced inflammatory response, and the potential role of TLR4 in this process. We observed that the total number of secreted nanovesicles was higher in ethanol-treated WT astrocyte than in untreated cells. No changes were observed in the amount of isolated exosomes between untreated and

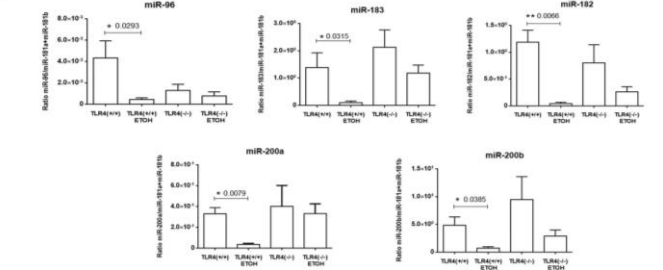
ethanol-treated TLR4-KO cells. These results suggest that astrocyte-derived exosomes could act as cellular transmitters, amplifying the neuroinflammatory response induced by ethanol through TLR4 activation.

Figures

A



B



C

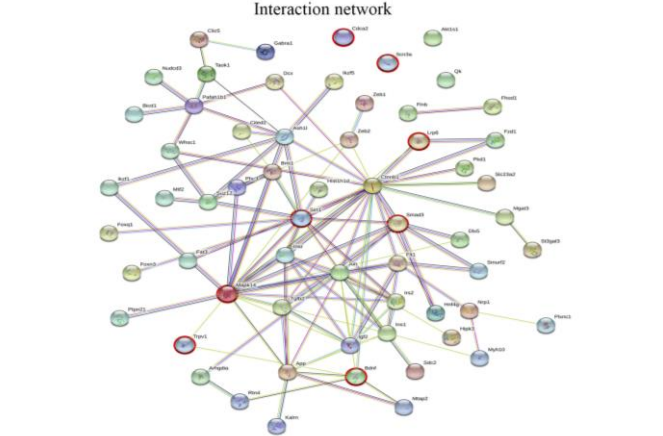
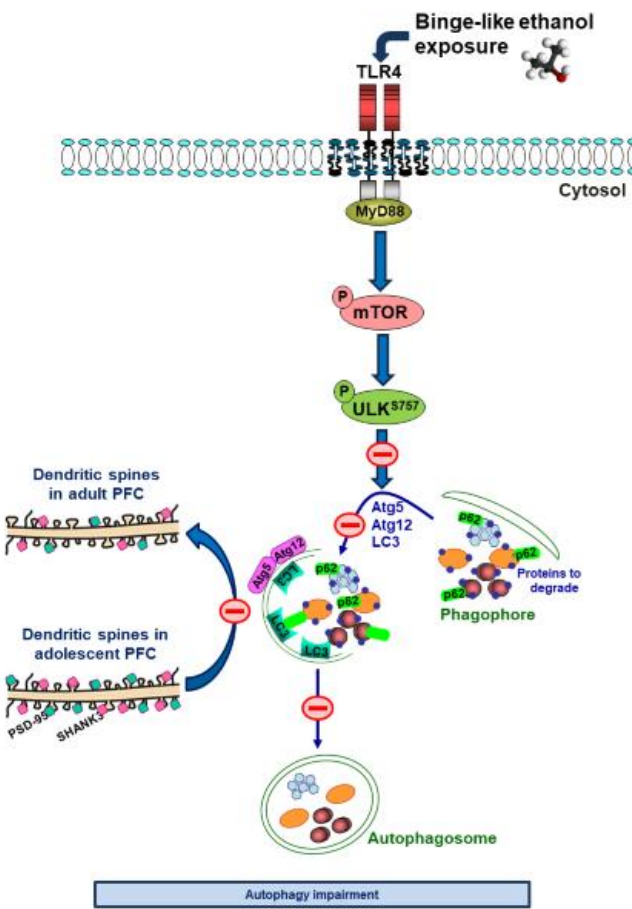


Figure 1 Genomic organization, structure and expression of miR-183 and the miR-200 gene clusters involved in alcohol abuse and the TLR4 immune response. (A) Homology sequence with a conserved seed of the miR components of the mmu-miR-183 cluster (miRs-183, -96, -182) and the miR-200a/b. (B) The RT-qPCR shows a differential expression at the levels of miR-183, miR-182, miR-96 (miR-183C), miR-200a and miR-200b in the cortices of the ethanol-treated vs. untreated WT and TLR4-KO mice. n= 9-11 independent experiments. *p<0.05, **p<0.01 (Student's t-test). (C) The interactome PPI (protein-protein interaction) network of miR-183C and the miR-200 family in alcohol-induced neuroinflammation in the WT and TLR4-KO mice cortex.

Figure 2: Schematic model of the effects of ethanol in the autophagy pathway through the TLR4 response. Binge-like ethanol exposure in adolescence, by activating the TLR4 immune response, impairs the autophagic pathway by increasing the phosphorylation form of mTOR, and ULK. This model suggests that by lowering the levels of LC3-II, ethanol causes p62 accumulation, impairing phagophore and autophagosome formation. Deficient autophagy could induce defects in synaptic scaffold proteins PSD-95 and SHANK3 in dendritic spines (synaptic refinement), which is a hallmark of adolescent synaptic maturation. MyD88: myeloid differentiation primary response gene 88.



Publications

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Montesinos J, Pascual M, Millán-Esteban D, Guerri C Binge-like ethanol treatment in adolescence impairs autophagy and hinders synaptic maturation: Role of TLR4 *Neurosci Lett*. 24; 682:85-91, 2018.

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Congresses Presentations

Congress of the International society for Biomedical research on Alcoholism, Kyoto, September 9-13, 2018, Participation as Speaker in two symposia

Symposium: NOVEL MECHANISMS OF ETHANOL-INDUCED DAMAGE TO THE DEVELOPING BRAIN : Chairs: C.Guerri and F. Valenzuela

Oral Presentation: C. Guerri1, J.Montesinos1, D.Millan-Esteban1, M. Pascual: Role of the TLR4 immune response in alcohol-induced brain dysfunctions in a model of Fetal Alcohol Spectrum Disorders. -Symposium

Oral presentation: C. Guerri1, J.Montesinos1, D.Millan-Esteban1, M.Pascual. Binge-Like Ethanol Treatment in Adolescence Impairs Autophagy and Affects Synaptic Maturation: Role of the Neuroimmune Activation

Congreso Nacional de Sociodrogalcohol, Toledo, 8-10 March 2018

Participation in two symposia: Oral presentation

Guerri, C.1; Pascual, M.1,2; Montesinos, Exposición prenatal al alcohol y neuroinflamación.

Pascual-Mora, M and Guerri, C Consumo de alcohol en atracción durante la adolescencia: Perfil inflamatorio y diferencias de género

University courses

Biochemistry and Biomedical Science, University of Valencia, September 2018

Pathological consequences of alcohol metabolism (C Guerri).

Master in Biotechnology, Polytechnic University, Valencia, Alcohol, Immune system and Neurodegeneration, June 2018

Course of Neurobiology of Addiction –July 2-6, 2018, University of Oporto, Oporto

Dissemination activities

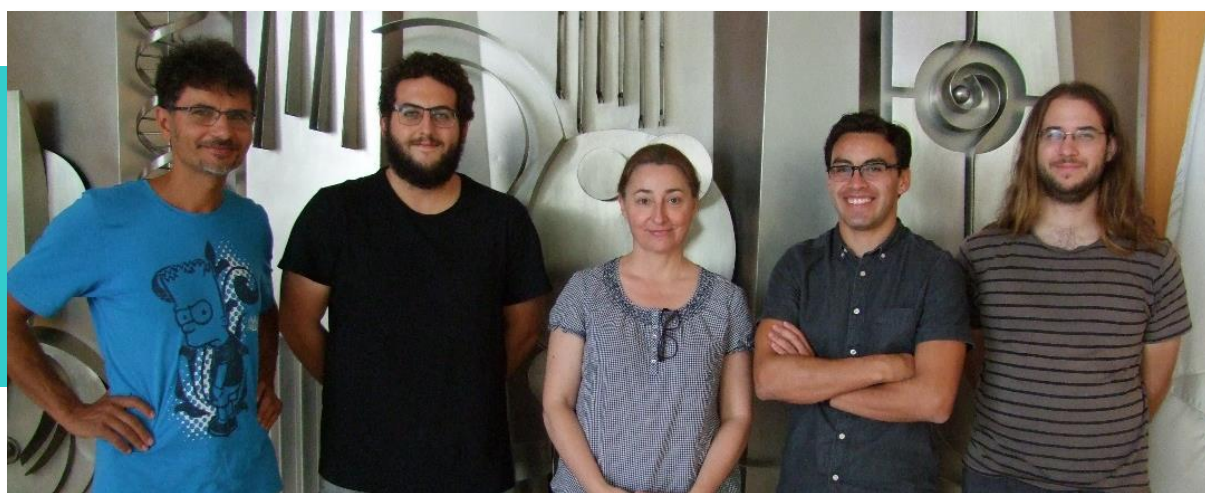
Lecture, C.Guerri “III Jornades sobre el Trastorn de l’Espectre Alcohòlic Fetal” June 2-3, Barcelona 2018

Participation in 1-Jornadas in Neuroscience, DONES en NEUROCIÈNCIA, 2-02- 2018, UV, Valencia

Participation within the congress “VI Internacional Congreso de Estudiantes de Farmacia” UV, “Galénica Moderna: Encapsulando ideas para administrar salud. Debate in the “research career”

Participation within the “Sexto Ateneo Biomédico”, Cátedra FISABIO –UV.

Microbiome: new advances in biomedical research.



Biomedical Imaging CIPF-FISABIO Joint Research Unit

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FISABIO

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Joaquim Àngel Montiel

Overview

The Research Unit CIPF/FISABIO is focused on the study of Brain Connectivity. The brain is a complex network of interconnected regions both structurally and functionally. The functional communications between brain regions play a key role in cognitive processes. Mental activities involve activation of neural networks in the brain areas involved creating various circuits in order to perform complex cognitive functions. The analysis of brain connectivity allows understanding the organization of the human brain, how distinct neural networks perform various tasks or how activation patterns are altered in pathological situations.

The Joint Research Unit develops methods to detect and quantify these connectivity types and their relation with brain function and combines different mathematical techniques to analyze images. It then interprets and integrates biological and medical knowledge to obtain models that explain and predict brain function in normal and pathological conditions. Our objectives are: to advance in the understanding of the mechanisms involved in the neurological impairment in different pathological situations, to better understand its causes, to develop early diagnostic procedures of cognitive impairment and to evaluate the usefulness of therapeutic procedures to reverse or prevent it.

Research Results

National projects in execution during 2018:

Challenges Collaboration 2016 (Ministry of Economy and Competitiveness) RTC-2016-5334-1 01/07/2016 - 06/30/2018 "CELÉXITA: Connectome-based knowledge Extraction for diagnosis and Therapy evaluation". 2018 III CALL FOR FISABIO AID: HELPS TO CARRY OUT THE DOCTORAL THESIS 2018 OF THE FOUNDATION FOR THE PROMOTION OF HEALTH AND BIOMEDICAL RESEARCH OF THE VALENCIAN COMMUNITY (FISABIO). UGP-18-159 27/09 / 2018-

26/09/2019 "MACHINE LEARNING IN MAGNETIC RESONANCE DESCRIBING THE PHYSIOPATHOLOGY OF LUMBAR PAIN" John Jairo.

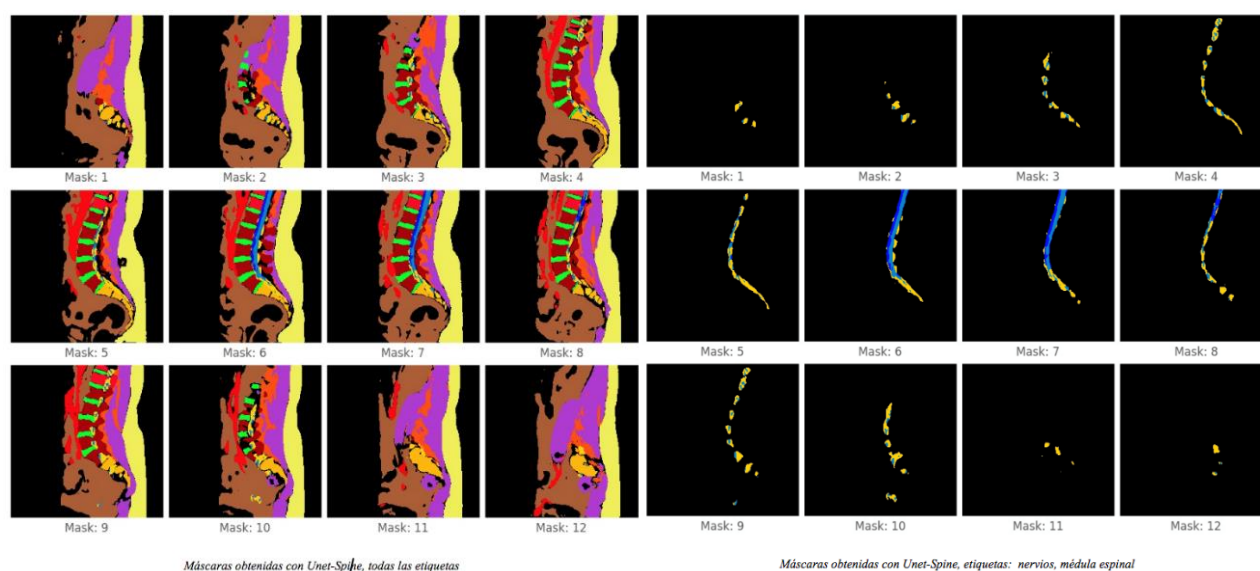
Predoctoral Assistance of the Ministry of Education:

2018 SUBSIDIES FOR THE RECRUITMENT OF RESEARCH PERSONNEL OF PREDOCTORAL CHARACTER. CONSELLERIA D'EDUCACIÓ, INVESTIGACIÓ, CULTURA I ESPORT. ACIF / 2018/285 "MACHINE LEARNING IN MAGNETIC RESONANCE DESCRIBING THE PHYSIOPATHOLOGY OF PAIN LUMBAR" 01/09 / 2018- 31/08/2021

International projects in execution during 2018.

Awarded in 2018; Execution 01/01/2019 - 12/31/2021; Deep-Learning and HPC to Boost Biomedical Applications for Health - DeepHealth (Grant agreement ID: 825111). European Commission - DG RTD. H2020-ICT-2018-2019. Partner María de la Iglesia Vayà. CEIB-CSUSP, CIBERSAM, FISABIO-CIPF Execution 01/01/2016 - 12/31/2018, Euro-BioImaging Preparatory Phase II - Project - EuBI PPII (Grant agreement ID: 688945). European Commission - DG RTD. H2020-EU.1.4.1.1. INFRADEV-1-2015-2. Partner María de la Iglesia Vayà. CEIB-CSUSP, CIBERSAM, FISABIO-CIPF

Figures



Publications

Rojas GM, Alvarez C, Montoya CE, Church-Vayá M, JE Cisterns, Gálvez M. Study of Resting-State Functional Connectivity Networks Using EEG Electrodes Position As Seed. Front Neurosci. 2018 Apr 24; 12: 235. doi: 10.3389 / fnins.2018.00235. eCollection 2018. PubMed PMID: 29740268<https://www.ncbi.nlm.nih.gov/pubmed/29740268>.



Bioinformatics and Biostatistics

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Overview

The increasing volume of biological data and the need to address biological and clinical problems with computational procedures, prompted the creation of our Bioinformatics and Computational Biology Program.

In this context, the main objective of UBB is research into new methods and tools of computational analysis in Biomedicine and Biotechnology, as well as the design, implementation and application of these methodologies in biomedical studies that generate relevant knowledge that allows us to improve diagnosis and prognosis in many diseases.

UBB main goals include the following: Development of new methodologies of computational analysis in biomedical and biotechnological research.

To provide statistical and bioinformatic advice for the planning of research projects for CIPF groups, improving their scientific quality and increasing their competitiveness in funding calls.

Teaching data analysis methods and bioinformatics tools to students and researchers.

To establish collaborations with other knowledge fields within and outside the CIPF, in which the computational analysis is the basis for the biomedical research.

Create synergies with the strategic and operational objectives established by the Conselleria de Sanitat Universal i Salut Pública.

Administration and scientific coordination of the CIPF computer cluster.

Research Results

In 2018, the computational methods and tools generated by the UBB have been applied in several biomedical studies, allowing a better characterization of the diseases and the obtaining of relevant knowledge that provides a more precise diagnosis and a personalized treatment of the patients, in the following areas:

Precision Medicine in Genomics

The study of survival levels in non-small cell lung cancer and the characterization of its tumoral subtypes, has generated information that improves the application of oncological treatment lines (Provencio M et al., Collaboration UBB-CIPF, Hospital Puerta del Hierro).

Genetics Service of the La Fe Sanitary Research Institute and UBB-CIPF are developing a genomic application of precision medicine in the Usher syndrome, through the co-supervision of Sandra Alandes' thesis and whose first results have been presented at the Bioinformatics Symposium in Granada, 2018. Collaborative project between University General Hospital of Valencia, IMEGEN Company and CIPF, funded by the Valencian Innovation Agency: "Biomarkers for precision oncology in lung, colorectal and melanoma cancer".

Functional characterization of omics studies

The use of functional enrichment methods in studies on the effect of alcohol (microRNA-Seq) and the assessment of the modulating role of cardiomyocytes (proteomics) has improved the understanding of functional aspects in these scenarios, providing two publications with two CIPF groups: Ureña-Peralta JR, et al. (Molecular and Cellular Pathology of Alcohol) and Ontoria-Oviedo I, et al. (Cardiovascular Repair). Signaling pathway activity analysis methods have produced relevant results in studies of cancer and viral diseases, generating 5 publications of interest: Cubuk C, et al., Fourati S, et al., Hidalgo MR, et al., Amadoz A, et al., Ferreira PG, et al.

Gender differences in health from big data approaches

Clinical and epidemiological indicators show a large number of diseases and health scenarios where gender differences are detected but the underlying causes of these changes are still unknown. The detection of these biological, clinical and psychosocial causes would allow us to be more precise in the diagnosis and in the personalized selection of treatments of patients. In this project, we present several big data approaches whose goal is to improve the knowledge that explain gender differences in 1) non small cell lung cancer, 2) non alcoholic fatty liver disease, 3) effect of alcohol on development, 4) schizophrenia and 5) loneliness.

Clinical, omics and, biomedical image are integrated in supervised models to predict groups of interest and improve the selection of health interventions. From these big data approaches we provide bottom-up prototypes to generalize at a population level in a next step. This collaborative project is being carried out among multidisciplinary groups: Hepatology and Liver Transplantation Unit, III La Fe; Hospital Puerta del Hierro, Madrid; Department of Methodology at Behavioral Sciences, University of Valencia; CIPF-FISABIO Joint Research Unit of Biomedical Imaging and several CIPF units: Molecular and Cellular Pathology of Alcohol, Oncogenic Signalling, Molecular Neuroendocrinology and UBB.

We have an AECC grant collaboration for a student ("Gender differences in lung cancer") and six master projects are linked with this project. First results have been presented at the Bioinformatics Symposium in Granada, 2018. Finally, also in this year we have prepared the protocols for the use and coordination of the computational infrastructure of the CIPF, and currently researchers from 8 research institutions are sharing this computational resource.

Our course on Bioinformatics and Biostatistics: WODA (Web-based Omics Data Analysis), "How to determine the sample size in my experiments?" And the teaching participation in 6 masters of Bioinformatics and Computational Biology.

Bioinformatic and biostatistical support in 46 consultations of CIPF groups.

Publications

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Genomics and Gene Expression

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Pedro Salguero García

Overview

The Genomics of Gene Expression group is interested in understanding the functional aspects of gene expression at the genome-wide level and across different organisms and its relationship with diseases and traits.

During 2018, the group has worked in different projects related to the development of statistical methods and tools for multi-omic data integration. We have developed new strategies for the combination of RNA-seq data (transcription) and ATAC-seq (transcriptional regulation) in the context of myeloid cell lineage differentiation and in order to infer transcriptional networks. We have continued the development of Paintomics 3, a software that allows to integrate any type of multiomic data on the basis of KEGG pathways.

Another important area of research is the development of methods and tools for the study of the expression of alternative isoforms. We have evaluated different algorithms for the differential isoform expression analysis and published the first method to study differential splicing in time series experiments. We have investigated the role of alternative splicing (AS) in T1D to discover that this regulatory mechanism is prevalent in disease-related genes, and developed two new software tools to identify isoforms from third generation sequencing data, and to study the functional implications of alternative splicing. Finally, we have established a new research path that makes use of two cutting edge methods, single-cell RNAseq and CRISPR, to develop novel methods for the analysis of alternative isoform expression and functional role.

Research Results

The Genomics of Gene Expression lab is interested in understanding functional aspects of gene expression at the genome-wide level and across different organisms and its relationship with diseases and traits.

The group has developed statistical methods and software tools that analyze the dynamic aspects of transcriptomes, integrate these with other

types of molecular data and annotate them functionally, with a special focus on Next Generation Sequencing (NGS) data.

We are creators of bioinformatic software tools such as Blast2GO, Paintomics, maSigPro, NOISeq, Qualimap, SQANTI, with thousands of users worldwide. We have coordinated two major EU projects: the STATegra project on multiomics data integration (11 partners) and the Marie Curie DEANN action to create a NGS network with American countries (16 partners). In addition, we are leaders of WP4 in ChroMe (12 partners). We have (co-)organised numerous bioinformatics and NGS conferences -including the largest conference in the field of Computational Biology ISMB-, and given specialised courses in bioinformatics in more than 10 countries and 5 continents with more than 550 attendees.

Through a PROMETEO grant from the Generalitat Valenciana and in collaboration with the Universidad Politécnica de Valencia and the Instituto Valenciano de Biomedicina, our batch correction algorithm has been improved and the manuscript submitted awaiting publication. Furthermore, in collaboration with the Quantitative Modelling of Cell Metabolism group from Technical University of Denmark, we have developed a novel multiomic integration strategy to combine transcriptomics data with metabolomics data via COBRA methods.

We participate in the Marie Curie ITN ChroMe that seeks to understand the relationship between chromatin and metabolic disorders. Within this network, we are committed to the development of new bioinformatics tools to assist scientists in the study of data sets combining metabolomics, gene expression and epigenomics information. We have continued the development of Paintomics 3, which visualizes any type of multiomic data within the frame of KEGG pathways, and published the tool in Nucleic Acids Research Journal in its annual web server edition. Moreover, during 2018, Paintomics 3 has been improved by adding support for additional pathway databases as well as new features to provide more insightful results.

In addition, within the framework of the ChroMe project, we have created PEAR, a new tool to infer pathways from scientific literature, which incorporates Deep Learning methods to identify biological entities in scientific texts. This work will soon be published in Bioinformatics.

Another important area of research of the group is the development of methods and tools for the functional analysis of the expression of alternative isoforms, which is the consequence of alternative splicing (AS), an important regulatory process to expand the complexity of transcriptomes and proteomes in superior eukaryotes. We have improved tappAS (<http://tappas.org>), a Java desktop application for alternative functional splicing analysis released in 2017. tappAS represents functional annotation with isoform resolution and implements novel statistical methods for identifying AS-regulated pathways, studying differences in polyadenylation sites, evaluating differential inclusion in functional motif transcripts as a consequence of AS, and interpreting the functional consequences of differential expression of isoforms.

Within this research area, we have started two projects that aim to extend our work in the field of alternative splicing. The first of them uses single-cell RNAseq data to study isoform co-expression. We are currently developing a computational method to infer co-regulatory relationships between modules of transcripts, gaining insight into splicing as an additional layer that contributes to cell identity and cell type-specific biological processes. In 2018, we have published the first review study on single-cell isoform studies in Genome Biology, which will serve as a critical guide on experimental and computational methods for the field. The second research line uses CRISPR to perform isoform-specific knock-outs, aiming to validate the functional role of candidate alternative splicing events. We have initiated the validation of our experimental approach in a mouse embryonic stem cell line on a previously selected candidate, Ctdnn1.

Publications

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Metabolomics

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Overview

The Joint Research Unit has been traditionally associated with the “Clinical Metabolomics” strategy initiated in 2005, under the coordination of Pineda-Lucena (CIPF), that facilitated the actualization of infrastructures, both Nuclear Magnetic Resonance (NMR) and Mass Spectrometers (MS) available at different research institution located in Valencia, including those located at the CIPF. These efforts, focused on the strengthening of infrastructures required for promoting the development of new diagnostic and therapeutic procedures, were possible thanks to the support of the Conselleria de Sanitat Universal i Salut Pública (FEDER funds) of the Valencian Government. Later on, the Conselleria de Sanitat Universal i Salut Pública adopted the firm decision, based on the results obtained so far, to further promote this research, and this strategy evolved towards another one termed “Innovative and Precision Drugs” that is currently coordinated by Pineda-Lucena (IIS La Fe), being the CIPF one of the institutions involved in the development of this initiative. The main objective of this strategy is to provide a wider framework for all the research associated with the development of new diagnostic (metabolomics) and therapeutic (drugs) procedures. It should be noticed that the development of this new strategy, that is closely linked to the current collaboration between the CIPF and the IIS La Fe, has facilitated that both institutions, as well as INCLIVA, benefited from significant economic investments in infrastructure during the last years.

Therefore, with regards to the scientific advances achieved since the initial agreement between both institutions, it seems pretty clear that the establishment of this Joint Research Unit has promoted not only the interactions between the CIPF and the IIS La Fe in clinical metabolomics, but has also facilitated the extension of this fruitful collaboration to the understanding of the molecular mechanisms involved in the onset of different diseases with a high socio-economic cost, the development of new clinical procedures (diagnostic, prognostic and predictive), and the characterization of the mode-of-action of drugs using metabolomics approaches.

Moreover, it is important to underline that the interactions between both institutions have translated in a number of activities including: (i) the publication of a significant number of scientific manuscripts associated with the clinical application of metabolomics in different fields, (ii) the coordinated involvement in strategic projects promoted by the Conselleria de Sanitat Universal i Salut Pública (Clinical Metabolomics, Innovative and Precision Drugs), and (iii) the application to regional, national and European projects.

Research Results

The establishment of the Joint Research Unit in Clinical Metabolomics between the CIPF and the IIS La Fe has facilitated the coordinated involvement in a number of competitive and strategic projects that have benefited both institutions. At the same time, the Joint Research Unit has significantly contributed to the activity of different Technology Services from the CIPF.

Publications

Since its formal inception (2016), the members of the Joint Research Unit in Clinical Metabolomics have published a significant number of scientific manuscripts (18) including the affiliation of both research institutions. All of these publications have focused on subjects directly associated with the scientific focus of the Joint Research Unit. Only in 2018, five articles were published in research areas closely related to the strategy in metabolomics (NMR, MS) and, therefore, aligned with the objectives of the Joint Research Unit: (1) Palomino-Schätzlein; M.; Becker; J.; Schulze-Sünninghausen; D.; Pineda-Lucena; A.; Herance; J.R.; Luy; B. "Rapid two-dimensional ALSOFAS-HSQC experiment for metabolomics and fluxomics studies: Application to a ¹³C-enriched cancer cell model treated with gold

nanoparticles". *Analytical and Bioanalytical Chemistry*, 410: 2793 – 2804, 2018.

(2) Prieto; J.; Seo; A.Y.; León; M.; Santacatterina; F.; Torresano; L.; Palomino-Schätzlein; M.; Giménez; K.; Vallet-Sánchez; A.; Ponsoda; L.; Pineda-Lucena; A.; Cuezva; J.M.; Lippincott-Schwartz; J.; Torres; J. "Myc induces a hybrid energetics program early in cell reprogramming". *Stem Cell Reports* 6: 1479 – 1492, 2018.

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(4) Palomino-Schätzlein; M.; Simó; R.; Hernández; C.; Ciudad; A.; Mateos-Gregorio; P.; Hernández-Mijares; A.; Pineda-Lucena; A.; Herance; J.R. "Metabolic fingerprint of insulin resistance in human polymorphonuclear leucocytes". *PLoS One* 13: e0199351 - e0199351, 2018.

(5) Pastor; M.; Fernández-Calle; R.; Di Geronimo; B.; Vicente-Rodríguez; M.; Zapico; J.M.; Garmage; E.; Coderch; C.; Pérez-García; C.; Lasek; A.W.; Puchades-Carrasco; Pineda-Lucena; A.; de Pascual-Teresa; B.; Herradón; G.; Ramos; A. "Development of inhibitors of receptor protein tyrosine phosphatase β/ζ (PTPRZ1) as candidates for CNS disorders". *European Journal of Medicinal Chemistry* 14: 318 – 329, 2018.



Polymer Therapeutics

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Overview

Polymer therapeutics have attained clinical proof-of-concept with multiple marketed products and two polymer therapeutics reaching the US top-ten best selling drugs. However, there still exist opportunities to expand and develop this platform in areas such as i) delivery of single/combination anti-cancer agents against novel molecular targets, ii) development of innovative polymeric materials with defined architectures, and (iii) treatment of diseases other than cancer, including neurodegeneration. The Polymer Therapeutics Laboratory at the CIPF focuses on these exciting research areas. Our research activity includes the design of advanced polymer conjugates - novel therapeutic and/or diagnostic tools with applications in metastatic cancer, neurodegeneration, and tissue regeneration, among others. The development of polypeptide-based biodegradable carriers, the use of combination therapies, and the design of nanoconjugates for novel molecular targets represent approaches we follow to achieve effective treatment options. Additionally, the implementation of quantitative tools to assess the fate of polymer therapeutics allows the investigation of how spatial conformation affects trafficking, pharmacokinetics, and whole body biodistribution, so allowing the exploration of a range of clinical applications.

Research Results

Polymer-based Combination Therapeutics for Triple Negative Breast Cancer - MyNano

In 2018, we continued our work on the European Research Council Consolidator funded project “MyNano”, resulting in multiple publications related to both combination conjugate design and the tools employed to assess treatment response.

During the development of pH-responsive biodegradable polyglutamic acid (PGA)-based combination conjugates with the aim of optimizing anticancer effects, we found that conjugates bearing Doxorubicin (Dox) and aminoglutethimide (AGM) via pH-sensitive linkers led to the specific release of Dox within the tumor micro-environment. Our second related study focused on the linkers employed between the PGA backbone and the conjugated drugs, revealing that the inclusion of a small flexible linker can

modify the spatial conformation of the conjugate to promote synergistic drug release and improve biological activity.

Finally, we characterized preclinical models of TNBC and associated metastasis employed to assess conjugate treatment response. Through collaborations with the Forteza (CIPF-IVP) and Pineda-Lucena (IIS La Fe) laboratories, we highlighted crucial differences between the immunodeficient MDA-MB231Luc human and the immunocompetent 4T1 mouse models that could aid the development of therapies and diagnostic tools.

In parallel within this project, we are also developing novel star-polypeptides with the ability to self-assemble into nanostructure of different sizes and shapes, from spheres to nanofilaments. We performed in-depth physico-chemical studies to investigate the stabilization effect of the core and the length of polypeptide chains on the stability of the nanostructures. Furthermore, we have developed different crosslinking strategies to secure such stability under physiological conditions that permit their use as targeted nanocarriers.

Developing Novel Polymer-based conjugates as Immunotherapeutic Agents

Using the star-shaped crosslinked PGA carriers developed within “MyNano”, we recently received a Proof-of-Concept European Research Council grant (“POLYMMUNE”) with the Florindo laboratory (Univ. Lisbon, Portugal) to extend their application to immunotherapy for melanoma. To this end, we are currently developing nanoconjugates using metastatic melanoma antigens (single and combination therapy) in the hope of generating a cheap and effective means to activate a broad immune response against melanoma without any unwanted side-effects. We believe that our approach will result in the off-the-shelf production of an effective cancer vaccine that will quickly and safely target different melanoma types. This approach, when fully developed, can be easily extended to other types of advanced tumors.

Diagnosing Early Osteoarthritis with Polyglutamate-conjugated Fluorescent Probe

In collaboration with the Dive (Université Paris-Saclay) and Nagase (University of Oxford) laboratories, we developed a PGA-conjugated matrix metalloproteinase 13 (MMP-13)-cleavable fluorescent resonance energy transfer-based (FRET) probe to aid in the imaging of the early molecular changes in osteoarthritic joints in the hope of fostering the development of novel and effective disease-modifying drugs. Application of this novel PGA-conjugate probe allowed the detection of early biochemical events that occur in a surgically induced murine model of osteoarthritis before major histological changes. Furthermore, we proved the utility of the probe to monitor the in vivo efficacy of an orally bioavailable MMP-13 inhibitor that effectively blocks cartilage degradation during the development of osteoarthritis.

Crossing Challenging Biological Barriers: The Blood-Brain Barrier and the Skin

In collaboration with the Viña and García-Verdugo laboratories (Univ. Valencia), we have developed a “Trojan Horse” strategy, based on novel LRP-1-targeted nanosystems that ameliorated cognitive decline in an in vivo model of Alzheimer's disease upon conjugation of neuroprotective and anti-inflammatory drugs. A collaboration with the Battaglia laboratory (UCL, UK) will facilitate the optimization of targeting residue surface density allowing a higher level of BBB crossing and the mechanisms involved using in vitro 3D BBB models. To achieve intranasal delivery carriers, we have developed star-shaped PGAs with reversible disulfide chemistries that disassemble under reductive media to enhance chemical adsorption on the mucosa and to allow greater diffusion/penetration rates in the brain. Additional surface modifications with targeting motifs have improved mucosal barrier crossing. Using this system, we have studied mucodiffusion and ex vivo permeation assays in a sheep mucosal model and neuroprotection in a primary microglia model via treatment with a combination conjugate comprising an anti-inflammatory drug and anti-oxidation drug, and will soon assess pediatric glioblastoma therapy via conjugate treatment in patient-tumor derived tumorspheres via a collaboration with the Carcaboso-Montero laboratory (IRSJdD, Barcelona).

A RETOS grant supports another of our biological barrier-associated research project (“PolyPepSkin”) involving Polypeptide Therapeutic Solutions S.L. The primary goal of this project is to design novel drug delivery systems with different physico-chemical characteristics to enhance the penetrability of small molecules into different skin layers. We have already demonstrated that the conjugation of anti-

inflammatory drugs to polypeptide-based carriers increased the penetrability and therefore the activity of the drug in inflammatory skin diseases, such as psoriasis.

Developing Polymer-based Therapies for the Treatment of Castration-Resistant Prostate Cancer

Patients with castration-resistant prostate cancer (CRPC) currently face low survival levels, thereby necessitating the development of novel biomarkers to stratify patients and novel targeted therapies with greater efficiency. We are involved in a PROMETEO project (“BioChip”) together with the López-Guerrero (FIVO) and Pineda-Lucena (IIS La Fe) laboratories studying both the noted aspects in a CRPC patient subtype positive for a specific fusion gene (T2E). With the help of an ASEICA grant, we are now focusing on the polymer-conjugation of small drug inhibitors of poly ADP ribose polymerase (PARPi) as they represent a potentially exciting treatment option. We believe that the creation of PARPi conjugates will lead to improved patient responses and, therefore, improved survival rates in CRPC patients.

Figures

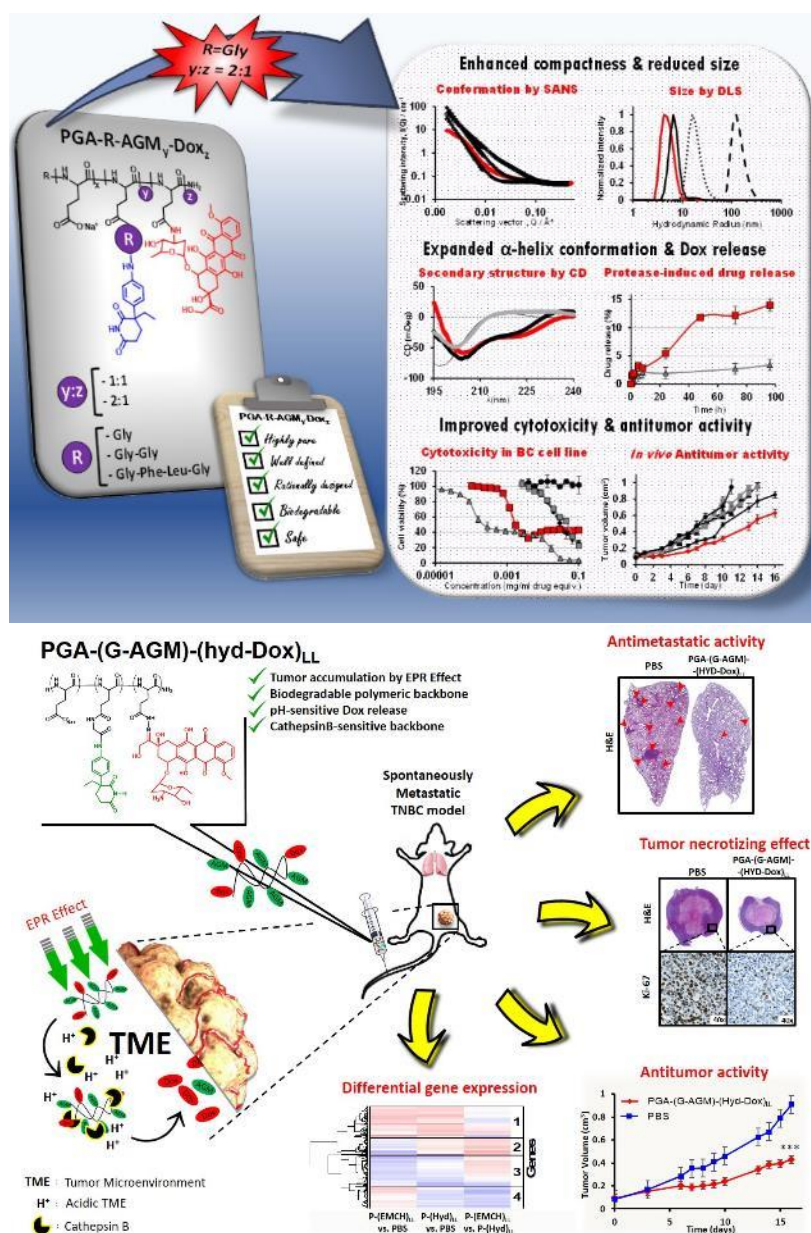


Figure 1. Polymer-based Combination Drug Therapy for Triple Negative Breast Cancer (TNBC) (MyNano). TNBC models and Rationally designed combination conjugates.

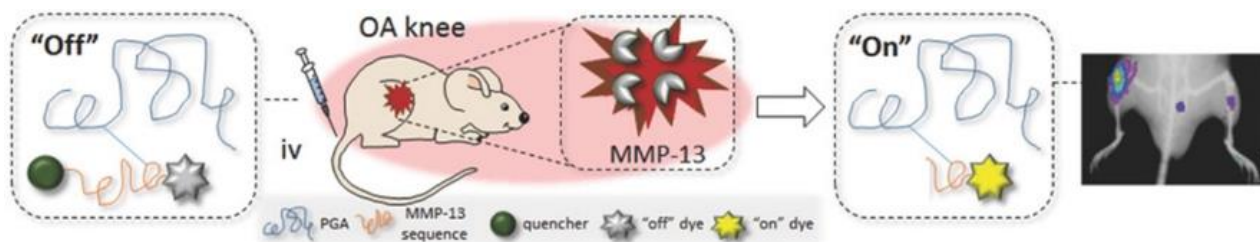


Figure 2. Diagnosing Early Osteoarthritis with Polyglutamate-conjugated Fluorescent Probes.

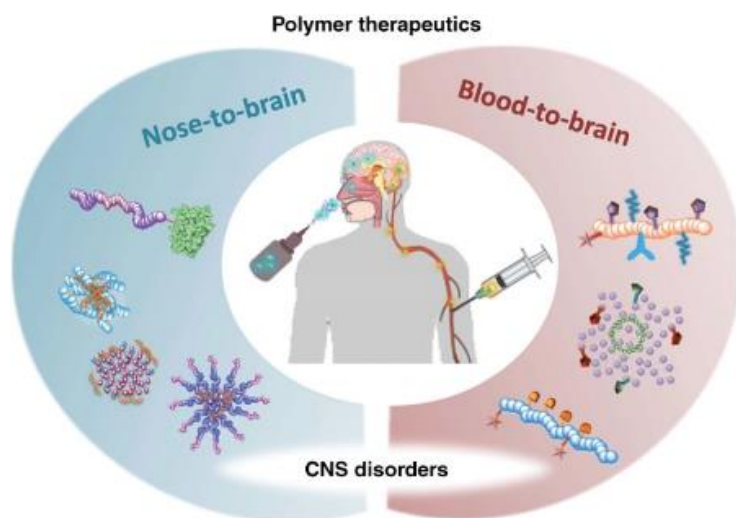


Figure 3. Crossing Barriers with Polymers to Treat Diseases and Disorders of the Brain.

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Protein and Peptide Chemistry

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Overview

Protein-protein-interactions (PPIs) govern almost all important processes in living organisms. The primary focus of the Protein and Peptides Research group is the identification and development of new modulators of PPIs from the apoptosis and inflammation pathways to re-establish the equilibrium in pathological situations such as cancer, neurodegenerative diseases or ischemia-re-perfusion associated damages.

Research Results

Our research line is focused on the study of transmembrane interactions from Bcl-2 family proteins. Bcl-2 family proteins control the permeabilization of the mitochondrial outer membrane, an irreversible step in the path of a cell towards death. Alterations of Bcl-2 proteins have been found in several tumors such as lymphoma, small-cell lung carcinoma, breast cancer, hepatocellular carcinoma or prostate cancer. Understanding the molecular mechanism of the Bcl-2 interactions that take place in the cytosol has been crucial for the development of the first anti-tumor drug targeting one of the proteins of this family, Bcl-2. (Venetoclax was approved in 2016 to chronic lymphocytic leukemia with 17p deletion). Our work has focused on understanding, at a molecular level, the interaction equilibria that these proteins establish in the membranes. We have evidenced the existence of homo- and hetero-interactions between the transmembrane segments of the Bcl-2 proteins and demonstrated that these interactions are crucial for the modulation of apoptosis. Our next objectives are; i) to identify drugs targeting the transmembrane hetero-interactions of Bcl-2 proteins and induce cell death in tumors (Bcl-TMInhs) and ii) to establish a “platform of Bcl-2 functional analysis” to understand how transmembrane somatic mutations found in cancer patients affect the canonical functions of these proteins.

In the inflammation field our research is focused in the development of new inflammasome modulators and their evaluation in disease models. The immune system protects the body from injury or infection triggering inflammation. However, deregulated inflammation is at the origin of several diseases such as autoimmune disorders, neurodegeneration or cancer. The inflammasome, a crucial macromolecular complex from the

innate immune system, is responsible of the activation of the potent pro-inflammatory cytokine IL- β and the inter-connection between innate and adaptative immune responses. We seek to develop new inflammasome modulators as therapies to fight against human diseases.

Nanoparticles for specific targeting and drug delivery in disease models. Our group is part of the “Join unit on disease mechanisms and nanomedicine”, a scientific platform for the collaboration between investigators from the Polytechnic University of Valencia and the CIPF. This joint unit aims to approach disease treatments from a multidisciplinary point of view and to apply last advances in nanomedicine to solve health problems. In particular, our group is involved, in collaboration with the group of Prof. Martínez-Mañez, in the development of nanoparticles for the specific targeting of inflammatory diseases, senescence and cancer.

Figures

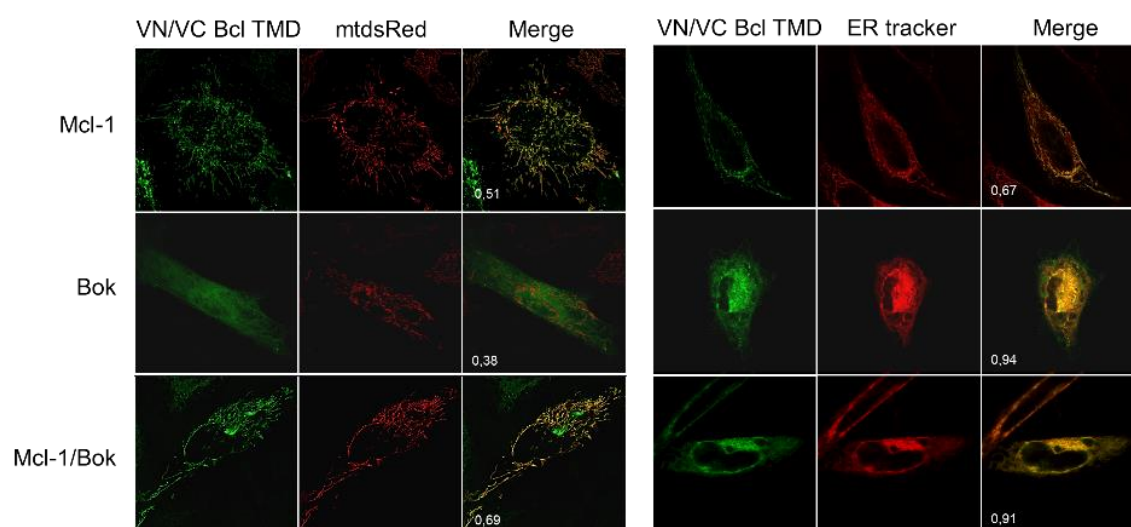


Figure 1. Mcl-1 TMD hetero-oligomerizes with Bok TMD and displace it from the ER to the mitochondria. a) Oligomerization analysis of Mcl-1 TMD and TMDs from the pro-apoptotic proteins Bax, Bak and Bok, measured by BiFC system in HCT 116 cells. ****P < 0.01** b) The pro-apoptotic effect of VN Bok TMD is partially inhibited by the presence of VN Mcl-1 TMD, measured by caspase-3 activity, in HCT 116 cells co-transfected with VN Bcl constructs. ***P < 0.5.** c) Intracellular localization of VN/VC chimeras was assessed by confocal images of Hela cells transfected with TMD constructs (green). Cells stably express a mitochondrial matrix marker (red).

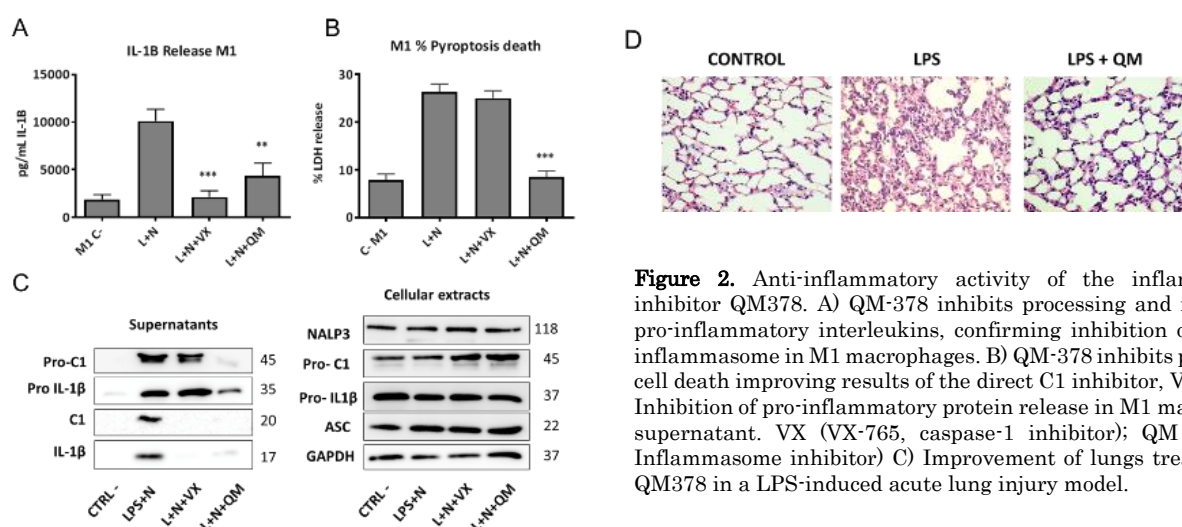


Figure 2. Anti-inflammatory activity of the inflammasome inhibitor QM378. A) QM-378 inhibits processing and release of pro-inflammatory interleukins, confirming inhibition of NLRP3 inflammasome in M1 macrophages. B) QM-378 inhibits pyroptotic cell death improving results of the direct C1 inhibitor, VX-765. C) Inhibition of pro-inflammatory protein release in M1 macrophage supernatant. VX (VX-765, caspase-1 inhibitor); QM (QM378; Inflammasome inhibitor) C) Improvement of lungs treated with QM378 in a LPS-induced acute lung injury model.

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Research in Biomedicine: Bridges and Obstacles. Orzaez M. X Congreso de Estudiantes de Farmacia Valencia

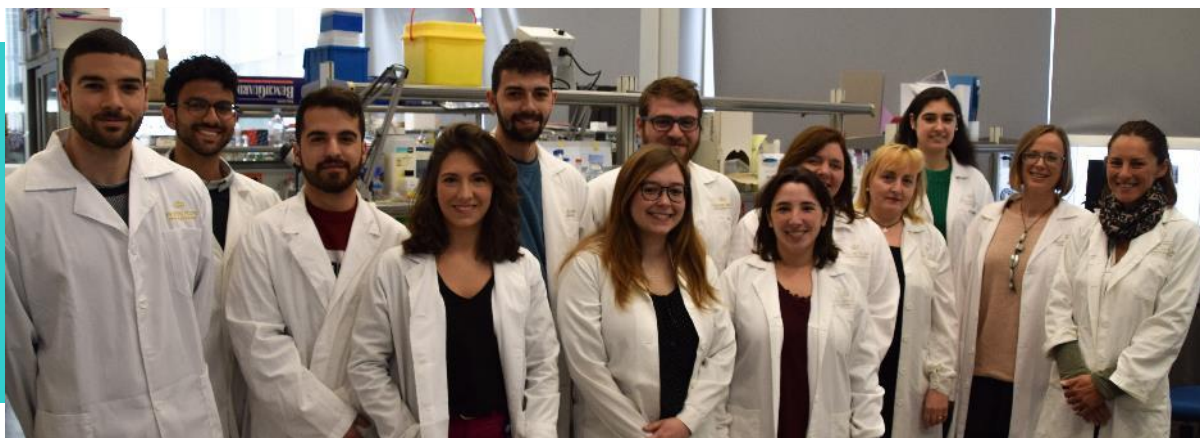
Endothelial senescence enhances tumor cell migration in vitro. Alejandra Estepa-Fernández, Ramón Martínez-Máñez, Mar Orzáez. International Workshop on Sensors and Molecular Recognition. Valencia 2018

Nanodevices for the efficient co-delivery of CRISPR-Cas9 editing machinery and an entrapped drug to solve inflammatory disorders. Alba García-Fernández, Gema Vivo Llorca, Mónica Sancho, Félix Sancenón, José Ramón Murguía, Ramón Martínez-Máñez and Mar Orzáez. XII International Workshop on Sensors and Molecular Recognition. Valencia 2018.

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Mesoporous silica nanoparticles for the treatment of navitoclax resistant triple negative breast cancer. Gema Vivo Llorca, Alba García-Fernández, Danya Bandeira-Lima, Félix Sancenón, Ramón Martínez-Máñez and Mar Orzáez. XII International Workshop on Sensors and Molecular Recognition. Valencia 2018.



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Overview

In the Neuronal and Tissue Regeneration laboratory (NTRL) we work in the Regenerative Medicine field, looking for efficient cellular therapies adapted to the demand of the pathological process and its clinical translation from the experimental models. The functional rescue of the neuronal activity lost after a spinal cord injury (SCI) constitutes the primary research line of the group. The SCI is a very complex and multiphasic process that leads us to prospectively direct the design of alternative therapies which involve the combination of stem cells transplantation, pharmacological treatments including nanomedicines, tissue engineering and biomaterials, electrical-magnetic stimulation, neuronal interfaces and/or neuro-rehabilitation, with the final objective of favoring neuroplasticity with functional results.

The NTRL develops therapies at the preclinical level with the ultimate goal of efficient translation to clinical practice. Proof of this is the creation of FactorStem (www.factorstem.es) a biotechnological company whose objective is the development and application of cellular therapies in veterinary medicine. The NTRL works closely with different reference hospitals for translational research (Hospital Politècnic I Universitari La Fe, Valencia, Hospital Casa de la Salud, Valencia, Hospital Vall d'Hebron, Barcelona). For additional information visit <http://cipf.es/en/web/portada/regeneracion-neuronal>.

Research Results

Biohybrids for Spinal Cord Injury Repair: Spinal cord injuries (SCI) result in the loss of sensory and motor function with massive cell death and axon degeneration. We have previously shown that transplantation of spinal cord-derived ependymal progenitor cells (epSPC) significantly improves functional recovery after acute and chronic SCI in experimental models, via neuronal differentiation and trophic glial cell support. We recently proposed and generated an improved procedure based on transplantation of epSPC in a tubular conduit of hyaluronic acid (HA), containing poly (lactic acid) (PLLA) fibers creating a “biohybrid” scaffold in tight collaboration with an expert group on Biomaterials led by Dr Monleón in the Polytechnic University of Valencia. First, in vitro experimentation showed that the PLLA microfibers included in the conduit induce a preferential neuronal fate of the epSPC rather than glial differentiation, favoring elongation of cellular

processes. The safety and efficacy of the biohybrid implantation was evaluated in a complete SCI rat model. The conduits allowed efficient epSPC transfer into the spinal cord, improving the preservation of the neuronal tissue by increasing the presence of neuronal fibers at the injury site and by reducing cavities and cyst formation. The biohybrid-implanted animals presented diminished astrocytic reactivity surrounding the scar area, an increased number of preserved neuronal fibers with a horizontal directional pattern, and enhanced co-expression of the growth cone marker GAP43 (see overall Image). The biohybrids offer an improved method for cell transplantation with potential capabilities for neuronal tissue regeneration, opening a promising avenue for cell therapies and SCI treatment. (Martínez C et al, *J Tissue Eng Regen Med.* 2019 Mar;13(3):509-521. doi: 10.1002/term.2816).

Figures

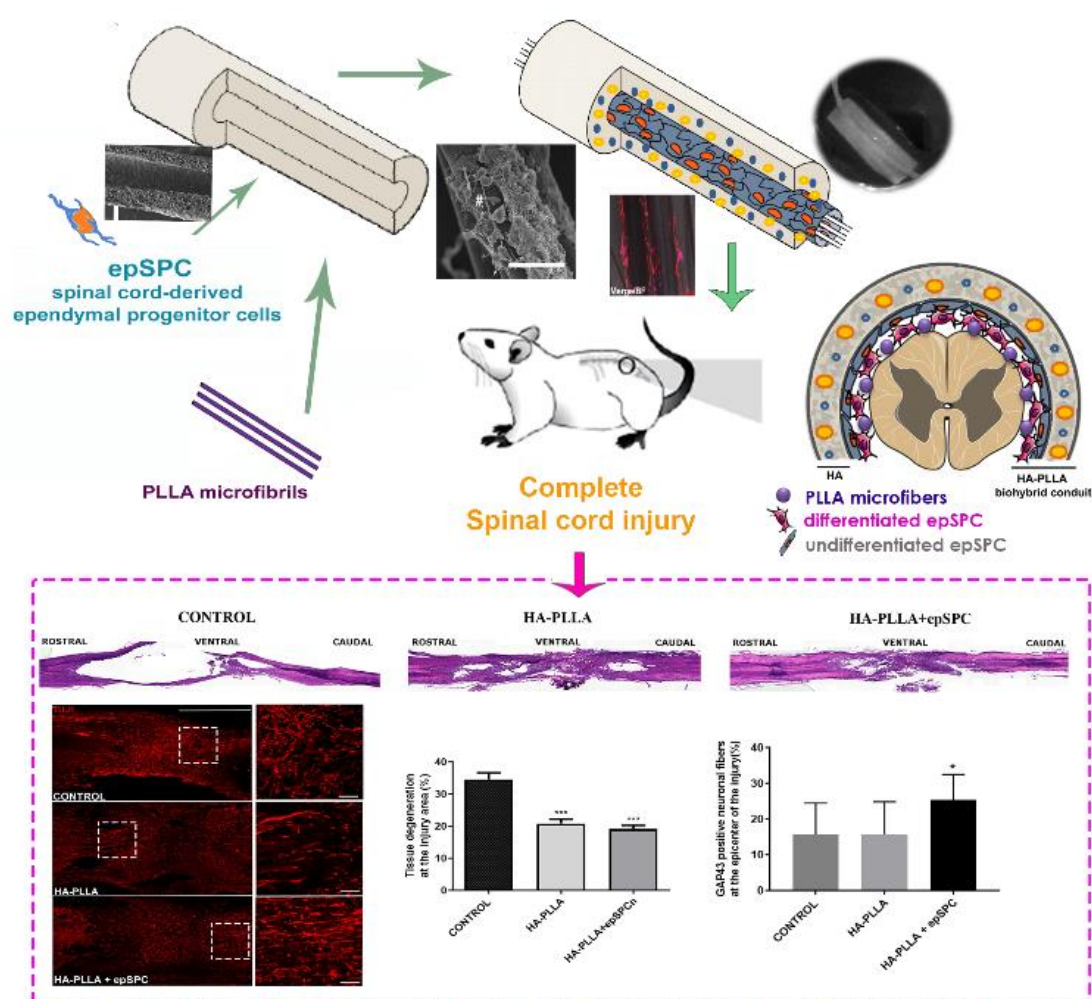


Figure 1. Overall image representing the Biohybrids for Spinal Cord Injury Repair: upper panel: The HA conduits with highly porous structure filled with PLLA micro fibrils in the central channel are laid out parallel to the conduit's axis and covered by epSPC seeded into the lumen. Lower panel: The scaffold, longitudinally open is placed covering both stumps immediately after sectioning the spinal cord. Three weeks after complete section, the hematoxylin-eosin anatomical staining of the spinal cord longitudinal sections showed a decreased cavity area only in the HA-PLA + epSPC implanted group, the group of animals that were implanted with the whole scaffold (HA-PLA + epSPC) showed a significant increase in the number of preserved neuronal fibers, TUJ-1 positive fibers (red) at the injury site and enhanced percentage of neuronal fibers expressing GAP43, a growth cone marker (green), indicating an activated process for axonal re-growth.

Publications

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FM19G11 and Ependymal Progenitor/Stem Cell Combinatory Treatment Enhances Neuronal Preservation and Oligodendrogenesis after Severe Spinal Cord Injury. Alastrue-Agudo A, Rodríguez-Jiménez FJ, Mocholi EL, De Giorgio F, Erceg S, Moreno-Manzano V. *Int J Mol Sci.* 2018 Jan 9;19(1). pii: E200. doi: 10.3390/ijms19010200

Combined plasma rich in growth factors and adipose derived mesenchymal stem cells promotes the cutaneous wound healing in rabbits. Chicharro D1, Carrillo JM1, Rubio M1, Cugat R2, Cuervo B1, Guil S3, Forteza J4, Moreno V5, Vilar JM6, Sopena J1. *BMC Vet Res.* 2018 Sep 21;14(1):288. doi: 10.1186/s12917-018-1577-y.

Therapeutic Strategies to Spinal Cord Injury (book title). Chapter title: FM19G11 and Ependymal Progenitor/Stem Cell Combinatory Treatment ISBN 978-3-03897-406-2 (Pbk); ISBN 978-3-03897-407-9 (PDF)

Culturing adult stem cells for cell-based therapeutics (Book chapter), ISBN 978-1-78984-867-0. IntechOpenScience. Elisa Oltra and Victoria Moreno Manzano

Electrical stimulation increases schwann cells proliferation inside hyaluronic acid conduits, that is published in 2018 EMF-Med 1st World Conference on Biomedical Applications of Electromagnetic Fields (EMF-Med). Cristina Martínez Ramo, María José Morillo Bagues, Fernando Gisbert Roca, Laura Rodríguez Doblado, Tomás García Sánchez, Luis Mir, Ester Giraldo Reboloso, Victoria Moreno Manzano, Manuel Monleón Pradas 10.23919/EMF-MED.2018.8526063

PSMA-Targeted Mesoporous Silica Nanoparticles for Selective Intracellular Delivery of Docetaxel in Prostate Cancer Cells. Eva Rivero-Buceta, Carla Vidaurre-Agut, César David Vera Donoso, Jose María Benlloch, Victoria Moreno Manzano and Pablo Botella. 10.1021/acsomega.8b02909

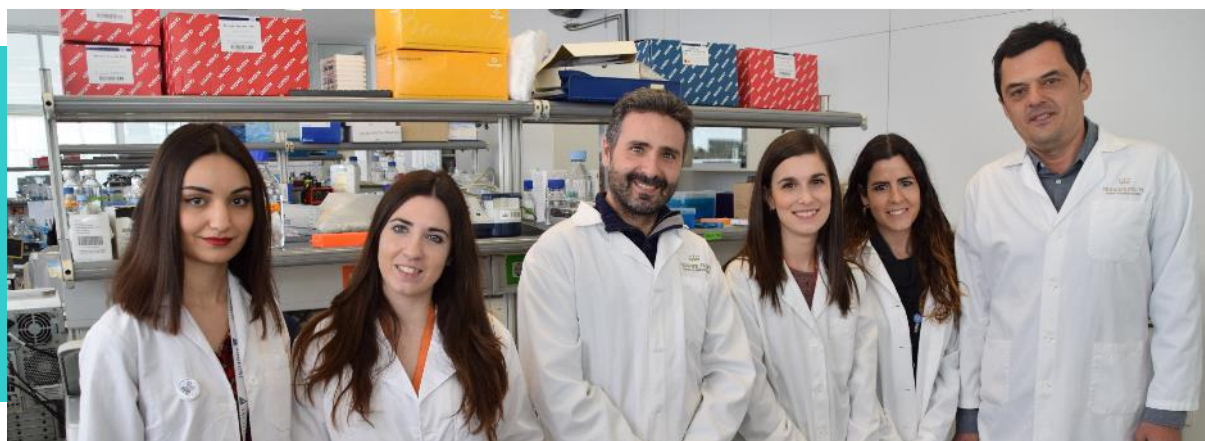
Collaborators

National: M^{re} Jesús Vicent (CIPF), Ana Conesa (CIPF), Slaven Erceg (CIPF), Francisco Javier de Lucio (Univ

Alcalá, Madrid), Manuel Monleón Prades (UPV, Valencia), Mireia García Roselló (CEU, Valencia), Angel Maria Hernández (CEU, Valencia), Maria Teresa Miras Portugal (Univ. Complutense, Madrid), Fivos Panetsos (Univ. Complutense, Madrid), Jose Manuel García Verdugo (UV, Valencia), Pablo Botella (ITQ-UPV, Valencia), Angel Serrano (UVC, Valencia), Michael Edel (Hospital Clinic Barcelona), Xavier Navarro Acebes (UAB, Barcelona).

International: Martin Balastik (IMG, República Checa), Renato Mantegazza y la Dra Stefania Marcuzzo (Carlo Besta, Milan), Lluís Mir (LEA, EBAM, Paris), Claudia Consales (ENEA, Rome, Italy).

Clinical: Cesar David Vera Donoso (Hospital la Fe de Valencia), Miguel Angel González-Viejo (VHIR-Barcelona), Ferran Pellice (VHIR-Barcelona), Ramón Cugat (Fundación García Cugat, Traumatología, Barcelona (Spain), Ignacio Muñoz (Hospital Casa de la Salud, Valencia).



Stem Cell Therapies in Neurodegenerative Diseases

Group members

Group Leader

Slaven Erceg

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Bankia Dual Fellowship

Maria Bañuls

Overview

The focus of our group is development of new cell therapy approaches using pluripotent stem cells such as human embryonic stem (hES) cells and induced human pluripotent stem (ihPS) cells as well as adult stem cells in the treatment neurodegenerative diseases. Our aim is to develop clinically acceptable protocols for neural differentiation in animal-free conditions as therapeutic tool for treatment spinal cord injury, different types of ataxias and retinal dystrophies and to test them in animal models. We also apply combinatorial approaches using small molecules to increase the success of cell-based therapy. In order to understand disease mechanisms we are using patient's ihPS cells to create human cellular models such as 2D cellular systems and 3D organoids.

Research Results

Cell therapy and role of GSK inhibition and astrocytes in axonal regeneration after spinal cord injury

Spinal cord injury (SCI) leads to permanent impairment of sensory and motor function bellow the injury mostly because the most axons fail to regenerate. Growth inhibitory molecules associated with myelin debris also attenuate axon regeneration after SCI. During many years of SCI research, many lines of investigation were focused on searching the strategies to promote the axonal growth of central nervous system (CNS). Astrocytes play a complex role in repair after spinal cord injury (SCI) contributing to glial scar formation. Although astrocytes are involved in these detrimental events following injury, they are also capable of providing neuroprotection and supporting axonal growth. Understanding the origin and differentiation patterns of astrocyte lineage cells after SCI is important for optimizing cell selection and pre-differentiation paradigms for transplantation strategies.

There are several aims of this study:

- 1) to determine the astrocyte population after SCI in different time points after SCI

- 2) to evaluate in vitro different subpopulations of astrocyte progenitors generated from multipotent and pluripotent stem cells in order to find the optimal cell source for axonal growth after SCI
- 3) to use the optimal cell source in combination with biomaterials in animal model of SCI to investigate axonal growth and locomotor recovery.
- 4) Inhibit GSK pathway to trigger astrocyte migration and neurogenesis

3D organoids to model ARSACS diseases

Autosomal recessive spastic ataxia of Charlevoix–Saguenay (ARSACS) is a rare neurodegenerative disorder characterized by ataxia, spastic paraparesis, polyneuropathy, thickening of the retina and evidence of superior cerebellar vermis atrophy representing the possible relationship between cerebellar dysfunction and ARSACS disease. We generated the iPSC lines from 1 ARSACS patients and one healthy sibling using non-integrative Sendai virus methodology as it was established during the first year. These iPSC lines were characterized related to pluripotency markers, teratoma formation, in vitro differentiation capacity and virus silencing. At the same time we apply the protocol to generate cerebellar cells from generated iPSC lines from ARSACS patients and control to test whether this in vitro model could be a faithful human in vitro model of ARSACS disease. In our project, we pretend to perform functional comparative analysis of cerebellar neurons (mainly granule neurons and Purkinje cells) to ones obtained from healthy individual in order to investigate the disease mechanisms and search therapy. Importantly these cells can be studied in detail in the context of the human genetic background, a crucial feature of the cellular model. Equally important is that it provides us with a unique opportunity for the testing of novel therapeutic agents in the presence of the human background, which has the potential to significantly reduce the cost of preclinical studies in animal models

Cell therapy for RPE related retinal dystrophies

Hereditary retinal dystrophies (HRD) represent a significant cause of blindness, affecting mostly retinal pigment epithelium (RPE) and photoreceptors (PRs), and currently suffer from a lack of effective treatments. Highly specialized RPE and PR cells interact mutually in the functional retina, with primary HRDs affecting one cell type leading to a secondary HRD in the other. Phagocytosis is one of the primary functions of the RPE and studies have discovered that mutations in the phagocytosis-associated gene MERTK lead to primary RPE dystrophy. Treatment strategies for this rare disease include the replacement of diseased RPE with healthy autologous RPE to prevent PR degeneration. The generation and directed differentiation of patient-derived human iPSCs may provide a means to generate autologous therapeutically-relevant adult cells including RPE and PR. However, the continued presence of the MERTK gene mutation in patient-derived iPSCs represents a significant drawback. Recently, we reported the generation of an iPSC model of MERTK-associated RP that recapitulates disease phenotype and the subsequent creation of gene-corrected RP-iPSCs. The main aims of this study are to differentiate gene-corrected RP-iPSCs into RPE and investigate whether these cells will recover both wild-type MERTK protein expression and the lost phagocytosis in vitro and in vivo. These findings provide proof-of-principle for the utility of gene-corrected iPSCs as an unlimited cell source for personalized cell therapy of rare vision disorders.

Publications

Machuca C, Vilches A, Clemente E, Pascual-Pascual SI, Bolinches-Amorós A, Artero Castro A, Espinos C, Leon Rodriguez MA, Jendelova P, Erceg S. (2018) Generation of a human induced pluripotent stem cell (iPSC) line from an unaffected female carrier of mutation in SACSIN gene. *Stem Cell Res.* 2018.

Lukovic D, Artero Castro A, León M, Del Buey Furió V, Cortón M, Ayuso C, Erceg S. Generation of a human iPSC line from a patient with Leber congenital amaurosis caused by mutation in AIPL1. *Stem Cell Res.* 2018 Dec;33:151-155.

Jendelova P, Sykova E, Erceg S. (2018) Neural Stem Cells Derived from Human-Induced Pluripotent Stem Cells and Their Use in Models of CNS Injury. *Results Probl Cell Differ.* 2018;66:89-102.

Machuca Arellano C, Vilches A, Clemente E, Pascual-Pascual SI, Bolinches-Amorós A, Ana Artero Castro A, Espinos C, Leon Rodriguez MA, Jendelova P, Erceg S. (2018) Generation of a human iPSC line from a patient with Autosomal recessive spastic ataxia of Charlevoix-Saguenay (ARSACS) caused by mutation in SACSIN gene. *Stem Cell Research* (in press). IF:2 (Q2)

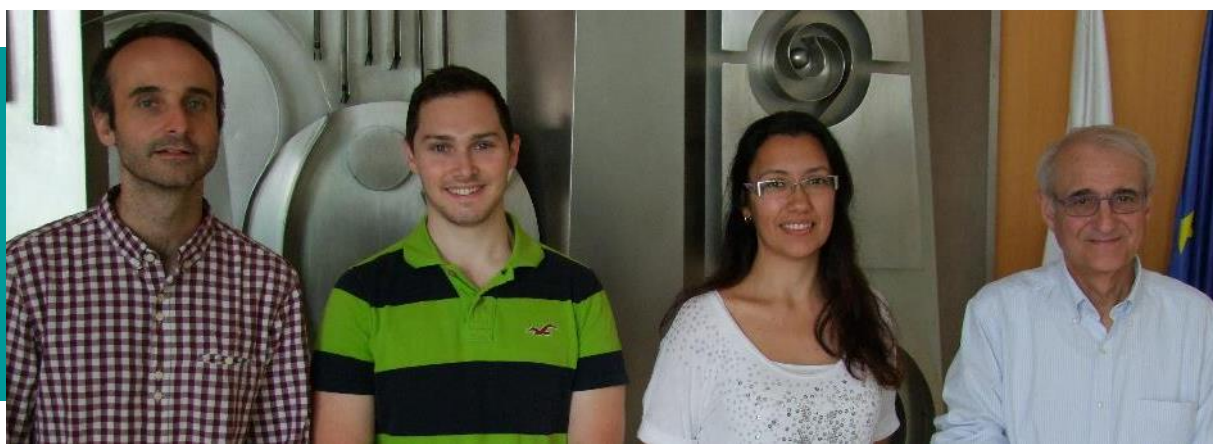
Artero Castro A, Leon MA, del Buey Furió V, Erceg S, Lukovic D (2018). Generation of a human iPSC line by mRNA reprogramming. *Stem Cell Research* 28:157-160. IF:2(Q1).

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Alastrue-Agudo A, Rodriguez-Jimenez FJ, López Mocholi E, De Giorgio F, Erceg S, Moreno-Manzano V. FM19G11 and Ependymal Progenitor/Stem Cell Combinatory Treatment Enhances Neuronal Preservation and Oligodendrogenesis after Severe Spinal Cord Injury (2018). *International Journal of Molecular science.* 9:19(1). IF:3,2 (Q1)

Artero Castro A, Lukovic D, Jendelova P, Erceg S, (2018) Human induced pluripotent stem cell models of retinitis pigmentosa. *Stem Cells* 36(4):474-481. IF:5,9(Q1)



Organic Molecules

CIPF-UV Joint Research Unit

Group members

UV

Group Leader

Santos Fustero

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Daniel Mark

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Cristina Morales

Miriam Rodenes

Alberto Llobat

Arancha Maté

Overview

Fluorinated organic compounds are playing an ever-increasingly important role in pharmaceutical research. Recent surveys suggest that approximately 25% of all newly approved small molecule drugs contain fluorine. This increasing prevalence has been driven by a deeper understanding by the medicinal chemistry community in applying fluorination to drug candidates to address many of the commonly encountered challenges in drug design.

Introducing fluorine into a pharmacologically active compound is a very effective means for modulating its physicochemical and biological properties. Because of fluorine's small atomic radius, the carbon-fluorine bond is only slightly longer than a carbon-hydrogen bond and, as a result, fluorine substituents have very low steric profiles. However, the very high electronegativity of fluorine relative to carbon imparts a strong dipole moment to the carbon-fluorine bond. Thus, fluorine substituents often engage in hydrogen-bonding and dipole-dipole interactions with adjacent functional groups on the bioactive molecule itself or with the biological target of interest.

The myriad of effects that fluorination provides offers the medicinal chemist many opportunities to utilize fluorine to solve a long list of problems typically encountered in drug discovery. Fluorination has been effectively used to improve the potency of drug candidates as well as their permeability. Fluorine has also found widespread use in improving the pharmacokinetics of drug targets through blocking sites of oxidative metabolism.

Research Results

We have developed a convenient and userfriendly metal-free protocol for the regioselective anti-Markovnikov hydrofluorination of olefins that uses readily available, inexpensive reagents (m-CPBA and HF·Py) and mild reaction conditions (CH_2Cl_2 , 0 °C, under air). The reaction has shown good functional group tolerance and is suitable for the late-stage fluorination of

complex organic molecules bearing a double bond. An additional benefit of our one-pot protocol is the stereospecific nature of the two processes, namely syn-epoxidation and anti-epoxide opening, as well as from extremely practical and economical reaction conditions. The development of an asymmetric version of this methodology is currently under study in our laboratories.

We have developed a simple, metal-free and highly regioselective procedure for the synthesis of (Z)- β -fluorovinyl sulfones by hydrofluorination reaction of alkynyl sulfones using TBAF, one of the cheapest and most commonly available fluoride sources. Furthermore, we have investigated the reactivity of this new class of compounds which are only recently introduced in the chemical literature. In this sense, we have described the first studies into the hydrogenation of fluorovinyl sulfones, obtaining a range of hydrogenated products in good yields and selectivities; a challenging reaction given the competing hydrodehalogenation that has hindered the use of similar substrates in hydrogenation processes. Both β -fluorovinyl sulfones and their hydrogenation products β -fluoroalkyl sulfones may find applications in medicinal and agrochemical sciences. Further studies towards an enantioselective version are currently being carried out in our laboratory.

A new methodology for the synthesis of enantiomerically enriched bicyclic δ -sultams is described, involving an initial organocatalytic intramolecular aza-Michael reaction of vinyl sulfonamides bearing a conjugated ketone at a remote position. The resulting Michael adducts were then subjected to an intramolecular conjugate addition over the vinyl sulfone moiety, thus rendering the final bicyclic sultams containing two stereocenters. The key point of this strategy relies on the use of vinyl sulfonamides as both, nitrogen nucleophiles and Michael acceptors. The use of phosphazene-derived bases avoided the racemization of the intermediate derivatives, rendering 6-membered ring bicyclic δ -sultams in enantiomerically enriched manner with a small erosion of enantiopurity. Anyway, after recrystallization, final sultams were obtained in almost enantiomerically pure form. Nevertheless, the enantioselective synthesis of either 5-membered ring products or benzofused derivatives was found to be out of the scope of our strategy.

Publications

Metal-Free and User-Friendly Regioselective Hydroxyfluorination of Olefins. *Org. Lett.* 2018, 20, 2338–2341

A metal-free and regioselective approach to (Z)- β -fluorovinyl sulfones and their chemoselective hydrogenation to β -fluoroalkyl sulfones. *J. Fluorine Chem.* 2018, 206, 108–116.

Recent advances in the synthesis of functionalized monofluorinated compounds. *Chem. Commun.*, 2018, 54, 9706–9725

Asymmetric Vinylogous Mannich-Type Addition of α,α -Dicyanoalkenes to α -Fluoroalkyl Sulfinyl Imines. *Adv. Synth. & Catalysis* 2018, 360, 366 – 373

Dual Role of Vinyl Sulfonamides as N-Nucleophiles and Michael Acceptors in the Enantioselective Synthesis of Bicyclic δ -Sultams. *Adv. Synth. & Catalysis* 2018, 360, 2885–2893

Recent progress in the application of fluorinated chiral sulfinimine reagents. *J. Fluorine Chem.* 2018, 216, 57–70.

Biorelevant fluorine-containing N-heterocycles. *Fluorine in Life Sciences: Pharmaceuticals, Medicinal Diagnostics, and Agrochemicals* 2018, Elsevier, Ed G. Haufe & F Leroux Academic Press (AP) Chapter 16 pp 575–599. Book Chapter



Cardiovascular Repair

CIPF-IISLAFE Joint Research Unit

Group members

IISLAFE

Lab

Group Leader

Pilar Sepúlveda

Investigators

Akaitz Dorronsoro

Imelda Ontoria

Post-Doctoral Scientists

María Ciria Calduch

PhD Scientists

Hernán González-King

Sandra Tejedor

Rafael Sánchez

Marta Gómez

Ignacio Reinal

Overview

Cardiovascular diseases are a major health problem in developed countries. Among them, heart failure has a limited treatment and cell therapy has become an alternative tool to conventional therapies. The Joint Unit for Cardiovascular Repair is a collaborative program between IISLAFE and the Prince Felipe Research Center. The group aims to develop basic research with a quality translational component, which will contribute to the development of new treatments for ischemic diseases. The study of the cellular mechanisms triggered by adult stem cells and the factors that influence the cell death of cardiomyocytes in apoptotic and inflammatory environments will allow the identification of molecular targets for the development of new drugs. The scientific activity of the group focuses on the use of adult stem cells in preclinical models of cardiac repair, the study of exosomes as mediators of cellular communication, the use of iPSC as a tool for the testing of cardiotoxicity in the heart, as well as the evaluation of new materials for cardiac repair.

Research Results

Mesenchymal stem cell (MSC)-derived exosomes are under intensive study for their applications in MSC-based therapeutics. Several studies point that paracrine factors secreted by MSC have a major therapeutic potential than MSC itself. Our work showed that the increased angiogenic potential exhibited by MSCs overexpressing hypoxia inducible factor-1 α (HIF-1 α) is due, to a great extent, to the overexpression of the Notch ligand Jagged1 loaded into exosomes. This finding describes a new signaling mechanism independent of cell-to-cell contact mediated through Jagged1-loaded in exosomes and sheds new light on how exosomes could mediate the beneficial effects of MSCs. Because of this, we are interested in MSC genetic modification to improve MSC-derived exosomes therapeutic potential in cardiovascular diseases.

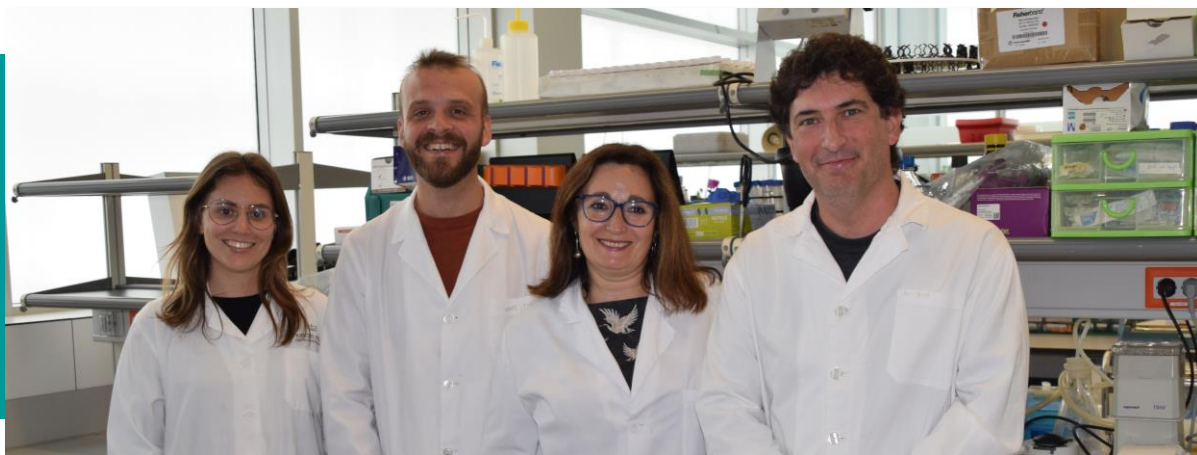
Induced-pluripotent stem cells (iPSC) are the most realistic system for disease modelling. In our group, differentiation of iPSC derived from different

patients to cardiomyocytes and endothelial cells allowed us to understand the molecular mechanisms of different cardiovascular diseases as Transposition of Great Arteries (TGA) or cardiotoxicity derived from anthracycline-based chemotherapy.

Cardiotoxicity derived from anthracycline-based chemotherapy is one of the most important side effects after treatment. In our laboratory, we have analyzed miRNA patterns from serum samples of breast cancer patients and we have established a miRNA signature which is able to distinguish between patients with high or low risk to develop cardiotoxicity after anthracycline-based chemotherapy. Consequently, clinicians will be able to treat specifically each patient according to their miRNA profile.

Publications

Ontoria-Oviedo I, Dorronsoro A, Sánchez R, Ciria M, Gómez-Ferrer M, Buigues M, Grueso E, Tejedor S, García-García F, González-King H, García NA, Peiró-Molina E, Sepúlveda P. Extracellular vesicles secreted by hypoxic AC-10 cardiomyocytes modulates fibroblast cell motility. *Front Cardiovasc Med*. 2018 Oct 25; 5:152



New group

Molecular and Cellular Immunology

Group members

Group Leader

Enric Esplugues

Investigators

Salvador Meseguer

Beatriz Perez-Benavente

Hanaa Zbakh

Lab Manager

Mari-Paz Rubio

Overview

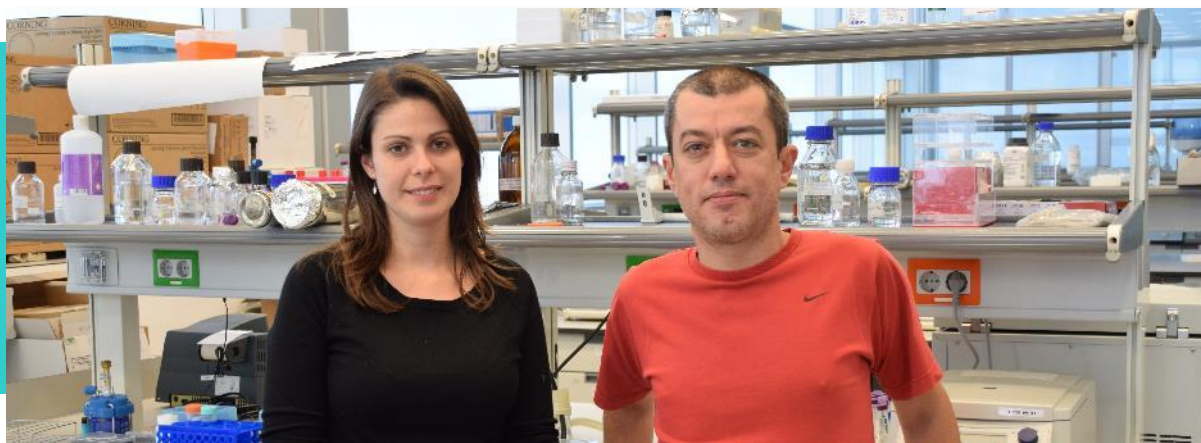
The major focus of research in our laboratory is the regulation of the immune responses in the context of infection, autoimmune disease and cancer. In all these processes inflammation plays a fundamental physiological role. We are interested in how different molecular and cellular regulatory mechanisms control inflammatory processes in pathological conditions. Of interest are to understand the links between inflammation and cancer and inflammation and metabolism with a particular emphasis on how metabolism governs this process.

Currently, our work is focused on the role of metabolism in regulating T helper cell development and function. To address fundamental questions in immune cell metabolism and how this impacts on protective immunity to infection, autoimmunity and cancer we use a wide variety of in vivo and in vitro approaches combining advanced genetic modeling of mice and immunologic techniques.

Publications

Effector T_H17 Cells Give Rise to Long-Lived TRM Cells that Are Essential for an Immediate Response against Bacterial Infection. Amezcua Vesely MC, Pallis P, Bielecki P, Low JS, Zhao J, Harman CCD, Kroehling L, Jackson R, Bailis W, Licona-Limón P, Xu H, Iijima N, Pillai PS, Kaplan DH, Weaver CT, Kluger Y, Kowalczyk MS, Iwasaki A, Pereira JP, Esplugues E, Gagliani N, Flavell RA. *Cell*. 2019 Aug 22;178(5):1176-1188.e15. doi: 10.1016/j.cell.2019.07.032.

Microbiota-driven interleukin-17-producing cells and eosinophils synergize to accelerate multiple myeloma progression. Calcinotto A, Brevi A, Chesi M, Ferrarese R, Garcia Perez L, Grioni M, Kumar S, Garbitt VM, Sharik ME, Henderson KJ, Tonon G, Tomura M, Miwa Y, Esplugues E, Flavell RA, Huber S, Canducci F, Rajkumar VS, Bergsagel PL, **Bellone M**. *Nat Commun*. 2018 Dec 3;9(1):4832. doi: 10.1038/s41467-018-07305-8.



New group

Cortical Circuits in Health and Disease

Group members

Group Leader

Pietro Fazzari

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Carmen Navarro

Research Assistant

Héctor Carceller Cerda

PhD Student

Ángela Rodríguez

Overview

Bridging the gaps between basic and translational neurobiology.

The cortex is the most complex and developed region of the brain. Higher cognitive functions, memories, emotions, they all depend on the proper formation and function of cortical circuits. Alterations of cortical wiring at neuritic or synaptic level underlie different brain pathologies: i) neurodevelopmental disorders, such as Schizophrenia; ii) neurodegenerative disorders, such as Alzheimer's Disease; iii) acute brain injuries provoked, for instance, by stroke or cerebral trauma.

Our aim is to understand the structure and function of cortical circuits in physiological and pathological conditions. Specifically, we investigate the role of disease-linked genes in the development and connectivity of excitatory/inhibitory cortical neurons using state-of-the-art approaches such as genetic, molecular neurobiology, morpho-functional analysis and electrophysiology. In last years, we determined role of the schizophrenia risk-gene *NRG1* in excitatory/inhibitory neurons^{2,3,4}. Currently, we are investigating the molecular signalling underlying *NRG1* function to identify the effectors of this gene and new therapeutic targets to treat Schizophrenia. Another long term goal of the lab is to apply our knowledge on circuit development to study neuronal regeneration in the adult: we develop new approaches in vitro and in vivo to discover new treatments.

Research Results

Neuroprotective role of *Nrg1* in cortical ischemia. *Nrg1* is a schizophrenia risk gene that controls the formation of excitatory and inhibitory synapses in cortical circuits. While different *Nrg1* isoforms are expressed during development, adult neurons primarily express CRD-*Nrg1* that is characterized by a highly conserved intracellular domain (*Nrg1*-ICD). We and others showed that the *Nrg1* intracellular signaling promotes dendrite elongation and excitatory connections during neuronal development. However, the role of *Nrg1* intracellular signaling in adult neurons and in pathological conditions remains largely unaddressed. Here, we investigated the role of *Nrg1* intracellular signaling in neuroprotection in aging and stroke.

Our bioinformatic analysis showed that Nrg1 intracellular domain is extremely conserved during evolution and that NRG1 expression significantly decreases with age in the human frontal cortex. Hence, we first tested whether Nrg1 signaling may affect pathological hallmarks of aging such as DNA damage and cellular stress in an in vitro model of neuronal aging. Our data did not suggest a role for Nrg1 in this context.

Previous studies showed that the soluble EGF domain of Nrg1 could alleviate brain ischemia, a pathological process that involves the generation of free radicals, reactive oxygen species and excitotoxicity. Hence, we tested the hypothesis that Nrg1 intracellular signaling could be neuroprotective in stroke. We found that Nrg1 expression significantly increased neuronal survival upon oxygen-glucose deprivation, an established in vitro model for brain ischemia. Notably, the specific activation of Nrg1 intracellular signaling by expression of the Nrg1 intracellular domain was sufficient to protect neurons from ischemia. Time-lapse experiments further confirmed that Nrg1 intracellular signaling increases the survival of ischemic neurons at different time points (Figure 1).

Finally, we investigated the relevance of Nrg1 intracellular signaling in stroke in vivo. Using viral vectors, we expressed the Nrg1 intracellular domain in cortical neurons that were subsequently challenged by a focal hemorrhagic stroke. We found that Nrg1 intracellular signaling could improve neuronal survival in the infarcted area.

Altogether, our data showed that Nrg1 intracellular signaling is neuroprotective upon ischemic lesion both in vitro and in vivo. The neurotoxic effect of stroke is complex and involves different mechanisms such as the generation of reactive oxygen species, excitotoxicity and inflammation. Therefore, further studies will be required to determine the molecular bases of the neuroprotective effect of Nrg1 intracellular signaling. In conclusion, our work indicates that the stimulation of Nrg1 intracellular signaling may be a promising target for the treatment of cortical ischemia.

Figures

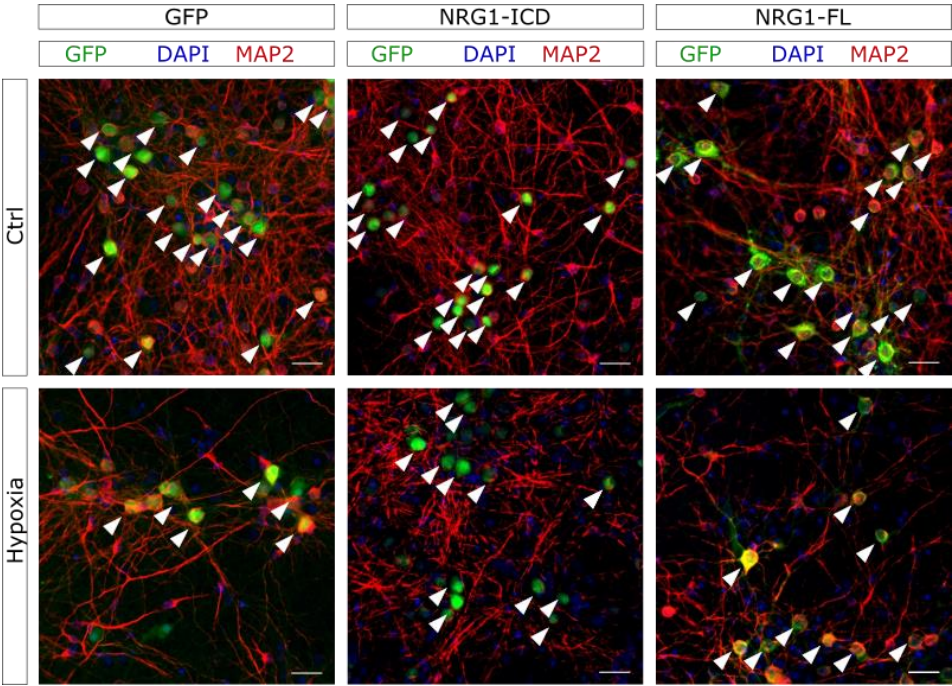


Figure 1. Nrg1 signaling is neuroprotective in hypoxia. Representative pictures of cortical neurons expressing GFP or Nrg1 in control and hypoxic conditions. Neuronal survival is increased by Nrg1

A photograph of a woman and a man in a laboratory setting. The woman, on the left, has curly hair and is looking towards the right. The man, on the right, is looking through a microscope. The background shows a wall with various posters and diagrams. The entire image is overlaid with a dark blue filter.

SOCIETY

As a publically-funded research center, we open our doors to society through events, talks, and guided tours. We take our science to the public and we bring the public into our institute to see our research. Public engagement is a priority for European research institutions, funding bodies, and evaluators. We are committed to involving the public, patients, and policy makers in our science.

In 2018 we organised for the first time events to celebrate the European Researchers' Night and the International Day of Women and Girls in Science. Both of them were highly successful in engaging the public and promoting science. In February, some of our female scientists joined the 11F initiative (11 February <https://11defebrero.org/>) and visited schools to share their knowledge and experience. One principal objective of these actions is to stimulate scientific curiosity and to encourage young girls to pursue scientific careers by providing a dialogue between women and girls in science.

We also recognise and highlight the active participation of our researchers at festivals and events such as the Pint of Science and VLC Data Beers, for example. This is great example of taking our science out to the public and generating public forums to discuss research topics.

Finally, our press office and research staff were actively involved in providing guided visits to schools and patient groups throughout the year.

The mission of CIPF's Communications and Events Department is to bring the Centre's research activity closer to those who can benefit the most from it, especially patients. We work to improve the public understanding of biomedical research. In order to fulfil this objective, we use all the information channels available, both traditional and digital.

In 2018, our news items received extensive media coverage with many spotlights in the press, either online or in print, and also via series of appearances on radio and television programs. Throughout the year, we set up interviews and work with local, regional, national and international media to provide accurate information about our scientific discoveries, events and management news.

2018 EVENTS

FBR LECTURES Series Conferences

| | | |
|---|-----------------------|------------|
| Fuel and Oil of the Cancer Engine | A. Carracedo | 19/01/2018 |
| Coordinating Gene Expression: links between transcription and RNA fate | J. Mellor | 09/02/2018 |
| Genome-wide study of transcription complexity and ribosome dynamics | V. Pelechano | 23/03/2018 |
| Remote signaling and stem cell-niche biology in the adult brain | I. Fariñas | 13/04/2019 |
| Targeting the extracellular matrix to restore neurological function and memory | J. Fawcett | 25/05/2019 |
| Targeting necroptosis and ferroptosis in experimental disease models, therapeutic perspectives | P. Vandenabeele | 15/06/2018 |
| Vaccines for Antibiotic Resistant Bacteria: "Our Experience with Multidrug Resistant Acinetobacter baumannii" | Michael J. Mc Connell | 16/11/2018 |
| Mitochondrial dynamics and its role in the pathophysiology of metabolic disorders | A. Rada Iglesias | 23/11/2018 |
| Mitochondrial dynamics and its role in the pathophysiology of metabolic disorders | A. Zorzano | 30/11/2018 |
| BrainCure: Therapy for Neurodegeneration with Brain Iron Accumulation | J. A. Sánchez-Alcázar | 14/12/2018 |



A. Carracedo



J. Mellor



V. Pelechano



I. Fariñas



J. Fawcett



P. Vandenabeelellor



M. McConnell



A. Rada Iglesias



A. Zorzanochano



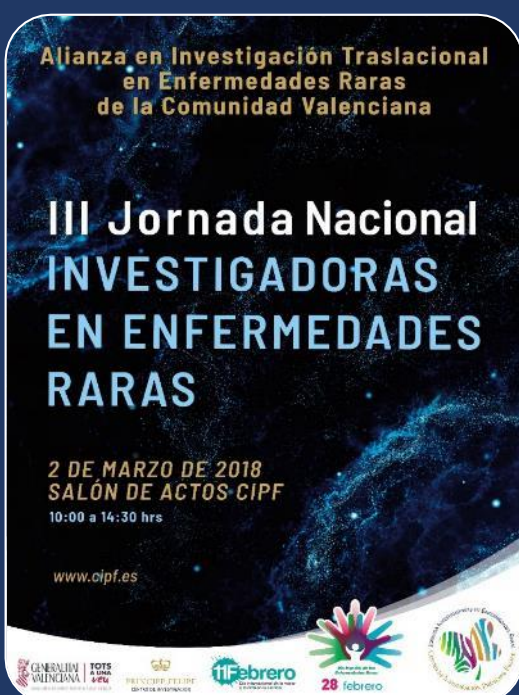
J.A. Sánchez Alcázarariñas

CIPF Seminars

| | | |
|---|-----------------|------------|
| From synapses to assemblies: neural circuit function and dysfunction | Isabel del Pino | 05/02/2018 |
| Single-cell RNAseq for the study of isoforms: how is that possible? | A. Arzalluz | 09/02/2018 |
| Liver 'organ on a chip | A. Wells | 13/03/2018 |
| Towards the Design of Personalised Polymer-based Combination Conjugates for Advanced Stage Breast Cancer Patients | M.J. Vicent | 16/03/2018 |
| Resolving tissue heterogeneity by Single-Cell Genomics | H. Heyn | 12/04/2018 |
| Cell-specific functions of EGFR in inflammation and cancer | Maria Sibilia | 18/05/2018 |
| Macrophages in tumor angiogenesis, lymph angiogenesis and immunity | M. Mazzone | 21/05/2018 |
| Targeting Nuclear Hormone Receptors in Inflammatory Disease | E. Esplugues | 01/06/2018 |
| Mitochondria: More is different | Fco. J. Iborra | 08/06/2018 |
| Looking at evolutionary systems through an evolutionary genomics prism | T. Gabaldón | 25/06/2018 |
| Active WNT vampirization by glioblastoma network leads to tumor growth and neurodegeneration | S. Casas | 19/10/2018 |
| Measuring mitochondrial respiration and other bioenergetic parameters using the O2kFluoRespirometer from Oroboros Instruments | S. Messeguer | 10/12/2018 |
| La neuroepigenética en la enfermedad de Huntington | Luis M. Valor | 21/12/2018 |

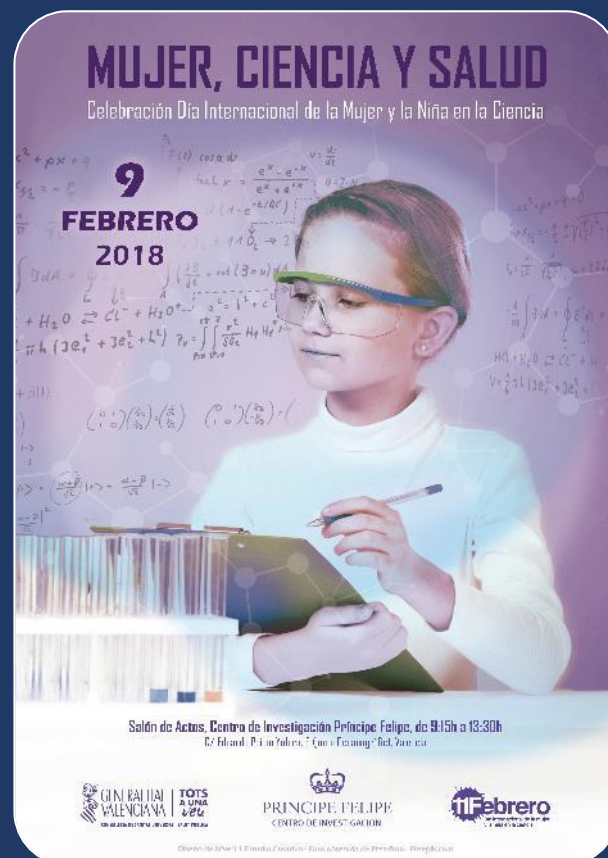
Scientific Conferences

| | |
|--|------------|
| III Jornada Nacional de Investigadoras en Enfermedades Raras | 02/03/2018 |
| 12th International Symposium on Polymer Therapeutics | 28/05/2018 |
| KICK-OFF: CLUSTER CIPF Plataforma para la Investigación en la Comunidad Valenciana | 21/06/2018 |
| Bioinform@tics Valencia 12/07/2018 | 12/07/2018 |
| I Valencian Vesicles Workshop | 21/09/2018 |
| Jornada BIOVAL | 27/11/2018 |



General Conferences

| | |
|---|------------|
| Jornada PHLL | 15/01/2018 |
| Women, Science and Health Conference 9/2/2018 | 09/02/2018 |
| I Jornada de Comunicación con las Asociaciones de Pacientes, Conselleria de Sanitat Universal | 17/05/2018 |
| Jornada EDEM | 26/09/2018 |
| European Researchers Night | 28/09/2018 |



Visits

| | |
|--|---|
| Ayuntamiento de Valencia | IVASP, Institut València de Seguretat Pública |
| Unidad de Documentación Clínica y Admisión (UDCA), Hospital Universitario Doctor Peset | SSZFK Slovenia |
| IES Consuelo Aranda, Alberic | Centro de Formación Profesional Santa Ana |
| Amparo Matíes, Club de Encuentro Manuel Broseta | IES Riu Túria, Quart de Poblet |
| Vicente Boluda, Asociación Valenciana de Empresarios, AVE Chairman | IES La Senda, Quart de Poblet |
| Caxton College | Apoyo Dravet |
| IES Pare Arques, Cocentaina | Universida Católica de Valencia |
| Colegio Jesús-María | IES Jorge Juan Port, Sagunt |
| IES Leopoldo Querol, Vinaròs | IES Tirant lo Blanc, Gandía |
| CEU San Pablo | Palacio de Congresos Valencia |
| CEIP El Molí, Torrent | |





| | |
|---|-----|
| Press Releases | 21 |
| Media appearances (press and online) | 138 |
| Radio and Television Appearances and Interviews | 8 |

SCImago ranking in which the CIPF is in the 9th position of Spanish research centers



New link between metabolic syndrome and liver cancer

BIOMEDICINA Y SALUD: Patología

Nuevo vínculo entre síndrome metabólico y cáncer hepático

Un estudio publicado en *Nature Communications* relaciona las alteraciones del metabolismo y los mecanismos moleculares del cáncer hepático. Los investigadores esperan que esta ruta abra nuevos modos de intervención para reducir la progresión de la enfermedad y el desarrollo de cáncer en pacientes con hígado graso no alcohólico.

Más información sobre: [cáncer hígado](#) [NAFLD](#) [autofagia](#) [síndrome metabólico](#)

SINC | [Ir a la página de SINC](#) | 28 noviembre 2018 11:30

International Biomedical Research Leaders will present their results at the CIPF in Valencia

europapress / c. valenciana

Líderes en investigación biomédica internacional presentarán sus resultados en el CIPF de València

Publicado 15/11/2018 11:13:36 [ver](#)

VALENCIA. 15 Nov. (EUROPA PRESS) -

El Centro de Investigación Principe Felipe (CIPF) acogerá este viernes la quinta edición del Ciclo de Conferencias The Future of Biomedical Research Lecture Series, una iniciativa con la que se fomenta y promueve un entorno de diálogo entre la comunidad científica, creando un espacio para abordar la actualidad de investigaciones biomédicas de vanguardia.

CIPF collaborates with Mount Sinai in the discovery of a link between metabolic syndrome and liver cancer

europapress / c. valenciana

El CIPF colabora con el Mount Sinai en el descubrimiento de un vínculo entre el síndrome metabólico y el cáncer hepático

SANIDAD ends the ERE and appoints a new scientific committee to promote the CIPF

Sanidad finaliza el ERE y nombra un nuevo comité científico para impulsar el Centro de Investigación Príncipe Felipe

EUROPA PRESS 15/06/2018

La Conselleria de Sanidad terminará con el Expediente de Regulación de Empleo que se aplicó al Centro de Investigación Príncipe Felipe (CIPF) en el año 2011. lo que permitirá eliminar la reducción salarial que aún se aplicaba a 15 trabajadores, y se ha nombrado asimismo a un nuevo comité científico para los próximos cinco años con el objetivo de hacer "más competitivo" y relanzar el centro.



Ana Barcelo announces the creation of the first scientific strategic plan of the CIPF

elperiodic.com

Sanidad y Salud

Ana Barceló anuncia la creación del primer Plan Estratégico Científico del Centro de Investigaciones Príncipe Felipe

25/06/2018 | elperiodic.com

La consellera de Sanidad Universal y Salud Pública, Ana Barceló, se ha incorporado este lunes como presidenta al Patronato del Centro de Investigación Príncipe Felipe (CIPF). A la sesión ordinaria también se han incorporado como nuevas vocales M^a Amparo García Layunta, directora general de Asistencia Sanitaria, y Carmelina Pla Silvestre, directora general de Eficiencia y Régimen Económico.



European Reserchers Night in CIPF

Celebramos la Noche Europea de L@s Investigador@s en Valencia



Volver

El 28 de septiembre se celebra la Noche Europea de L@s Investigador@s. Por eso, bajo el lema "Ven a descubrir con nosotros", ese mismo viernes se celebrarán en el Centro de Investigación Príncipe Felipe de Valencia actividades gratuitas para todos los públicos, prometiendo ser una tarde tan científica como divertida para dar a conocer qué se hace desde el centro. Desde FEDER participamos gracias al CIBERER y estuvimos representados por Juan Carlos González, Miembro de nuestra Junta Directiva y Representante Territorial, y por todo nuestro equipo de voluntariado: Santi, Paloma, Encarni y Carlos. sin ellos, no habría sido posible.

En este marco, se celebrarán talleres y experimentos, se explicará qué es la investigación en biomedicina, tendrá lugar una exposición del CIBERER y la de "Menos Raras", se explicará qué es un biobanco, se abrirá la posibilidad de preguntarle a científicos e incluso se podrá disfrutar de un monólogo científico de la mano de Carlos Manuel Cuesta.

La jornada cuenta con el apoyo de la Conselleria de Sanitat Universal i Salut Pública de la Generalitat Valenciana, el Fondo Europeo de Desarrollo Regional, el Centro de Investigación Príncipe Felipe, el CIBERER y el CIBERDEM.



28 de septiembre: La noche de los investigadores

Última actualización: 02/10/2018.

Me gusta 0 | Compartir | Twitter | G+ | Compartir | Share | Guardar

ciberdem isciiii

Buscar

Investigadores del CIBERDEM participan en la Nit Europea de la Investigación del CIPF



CIBERDEM Reserchers participate European Reserchers Night in the CIPF



EVAP/BPW 2018 Integrity Prize to Deborah Burks Interview at Levante TV



La Qüestió: 'Bates Blancues'. À punt



TVE informs about 'DESCUBRE SUMMER COURSE'

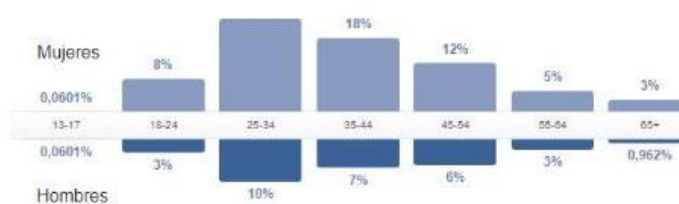
| | | |
|--------------------|-------|-----------------------|
| Twitter Followers | 4.000 | 58% women and 42% men |
| Facebook Followers | 1.450 | 70% women and 30% men |
| LinkedIn Followers | 2.910 | |
| YouTube Views | 4.000 | 60% women and 40% men |

Características de los seguidores ©

Datos para: Sector ▼

Sectores principales

| Sectores principales | Seguidores | % de seguidores |
|--|------------|-----------------|
| Investigación | 837 | 25,43% |
| Biotechnología | 486 | 14,76% |
| Enseñanza superior | 358 | 10,87% |
| Atención sanitaria y hospitalaria | 190 | 5,77% |
| Industria farmacéutica | 188 | 5,71% |
| Sanidad, bienestar y ejercicio | 152 | 4,62% |
| Servicios y tecnologías de la infor... | 91 | 2,76% |
| Servicios médicos | 58 | 1,76% |
| Productos químicos | 51 | 1,55% |
| Alimentación y bebidas | 39 | 1,18% |



| | | | | | | | |
|--|-----|--------|-----|-------|------|-----|--------|
| CIPF Laboratorio de Química de Péptidos y Proteínas Dra. M. Orzáez | 429 | 5,8 % | 196 | 4,9 % | 3,23 | 665 | 11,6 % |
| Clúster CIPF Plataforma para la Investigación en la Comunidad Val... | 208 | 18,3 % | 296 | 7,4 % | 1,48 | 537 | 9,4 % |
| Presentación Dra. Victoria Moreno Manzano | 511 | 5,1 % | 254 | 6,3 % | 1,57 | 497 | 8,7 % |
| CIPF Laboratorio de Señalización Oncogénica Dra. Rosa Farràs | 307 | 5,2 % | 97 | 2,4 % | 3,35 | 348 | 6,1 % |
| Programa DESCUBRE 2015 - Estudiantes | 415 | 8,4 % | 113 | 2,8 % | 2,26 | 276 | 4,8 % |
| Presentación Dra. Mª Jesús Vicent | 152 | 5,3 % | 106 | 2,6 % | 2,24 | 256 | 4,5 % |
| Presentación Dr. Slaven Erceg | 339 | 4,7 % | 168 | 4,2 % | 1,17 | 218 | 3,8 % |
| Presentación Dr. Vicente Felipo | 188 | 5,3 % | 76 | 1,9 % | 2,15 | 172 | 3,0 % |
| CIPF Laboratorio de Regeneración Tisular y Neuronal | 256 | 6,3 % | 74 | 1,8 % | 1,59 | 147 | 2,6 % |
| Presentación Dra. María Eugenia Armengod | 188 | 1,6 % | 60 | 1,5 % | 2,15 | 136 | 2,4 % |
| CIPF Servicio de Genómica y Genética Traslacional (SGGT) | 367 | 3,8 % | 56 | 1,4 % | 2,24 | 135 | 2,4 % |
| Noche Europea de los Investigadores 28 de septiembre 2018 | 286 | 8,4 % | 233 | 5,8 % | 0,32 | 125 | 2,2 % |



The background image shows a surgical suite. In the center is a large, adjustable operating table. Above it are two large, articulated C-arm X-ray machines. To the left, there's a mobile cart with various monitors and medical equipment. The entire scene is bathed in a warm, orange-yellow light, and a semi-transparent orange filter is applied over the entire image.

Technology

Core Facilities and Shared Scientific



Group leader **Ernesto de la Cueva**

Overview

Cutting edge biomedical translational research relies on a constantly evolving set of technologies designed to help advance scientific knowledge and to accelerate the transfer of experimental findings to medical practice. At CIPF these essential resources are managed through its Core Facilities, an organization designed to offer researchers access to the latest technology and best technical expertise.

The CIPF Core Facilities strive to provide fast, reliable, flexible and cost-effective services, offering in-house scientific support with a high degree of customization to match individual research needs.

The main mission of CIPF Core Facilities is to help scientists answer complex biological questions and facilitate the execution of advanced scientific projects. In order to achieve their goals, the Cores maintain state-of-the-art scientific instrumentation, provide specialized technological support and advice users in techniques and experimental design. Our services are offered both to CIPF affiliated researchers and to the broader scientific community. The Core Facilities organization foster a culture of effective knowledge transfer facilitating that technological advances revert to the society, the public health system and the industry.

A coordinated management of existing resources and capacities, an outstanding collaborative and interdisciplinary culture, and the concentration of diverse technology and singular infrastructures, allows us to offer a comprehensive and high quality technology portfolio. As a consequence of all the above, the Cores can be kept up to date providing added-value solutions and scientific applications.

The Core Facilities operate under a best practice approach and are ISO9001:2015 certified.

Overall, 2018 has been a year of growth both in activity and portfolio of resources, with a significant increase not only in the number of services provided, but also in the diversity and specialization of supported activities.

The organization of training, education and communication events, as well as the participation of Core Facilities personnel in scientific activities, have been areas of relevance where a lot of effort have been put with a significant increase in sponsored activities. All in all, the behaviour of the Core Facilities during 2018 consolidates the positive trend observed during the last years few years.

Animal Models and Comparative Medicine



Overview

Progress of biomedical sciences and the development of more efficient and safer diagnostics and therapeutic strategies require in-depth knowledge of diseases and the underlying mechanisms that cause them. For many diseases, this can only be achieved replicating those mechanisms or pathological conditions in a model organism. Therefore, the use of animal models is essential to understand how a biological process works in the context and with the complexity that only a living organism can provide.

Current regulations can only authorize the use of animals for experimental purposes if the model cannot be replaced in a valid *in vitro* alternative. Likewise, it imposes the refinement of the procedures and the reduction of the number of animals used, as two fundamental parts of the experimental design. This ethical approach to the use of animals is what has come to be known as "concept of the three R's" (replacement, reduction and refinement). The main goal of the Comparative Medicine Unit is to provide the standards and the environment that guarantee that animal use is performed in compliance with regulations and best scientific practice.

During the last year, the Unit has been involved in multiple projects: the evaluation of the oncogenic capacity of tumor stem cells isolated from non-small cell lung cancer patients, the generation of different PDX orthotopic models of cancer, the development of a heterologous model for the assessment of treatments for endometriosis lesions, a biohybrid assay for the promotion of axonal growth and regeneration of the central and peripheral nervous system, the development of new strategies in the treatment of spinal cord injuries (modulation of reactive astroglia after the transplantation of neural progenitors), among others.

The Unit operates an exceptional 700 m² experimental surgery facility where more than 30 training programs of minimally invasive surgical techniques have been delivered, especially in the field of laparoscopic genitourinary urology, gastrointestinal laparoscopy, thoracoscopy and arthroscopy.

Advanced Light Microscopy



Overview

Advanced microscopy provides users with a wide range of techniques and key tools in the field of biomedical research, but also in other scientific areas such as nanoscience and nanotechnology, new materials, QC and QA, etc. Amongst all available technologies, confocal microscopy is probably one of the most relevant to biomedical research. Not only because of its great resolution power, but also because of a greater sample penetration capacity. Confocal microscopy allows the capture of images in different focal planes that can be processed to generate three-dimensional reconstruction of structures.

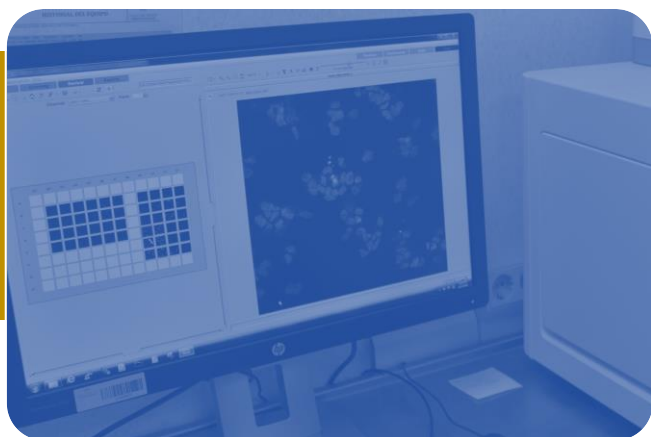
The CIPF Advanced Light Microscopy Unit offers a wide range of applications in microscopy techniques and data analysis, including conventional optical microscopy, fluorescence, in vivo imaging, confocal techniques, super-resolution, multiphoton, etc.

In 2018 the Unit has worked in different research projects of CIPF laboratories and has collaborated with other research institutions such as the Valencian Institute of Pathology (IVP), the Institute of Materials Technology of the Polytechnic University of Valencia, the PROCREA foundation, the IVI foundation, the Institute of Molecular and Cellular Plant Biology (IBMCP), etc.

During 2018, the Unit has collaborated with the Health Research Institute (IIS) of the Hospital La Fe of Valencia in a research project for the evaluation of mitochondrial activity and respiration rates of human oocytes as a marker of metabolic activity and oocyte viability.

During 2018 two new techniques have been fully implemented and made available to users: super-resolution microscopy and improved HTS.

Flow Cytometry and Cytomics



Overview

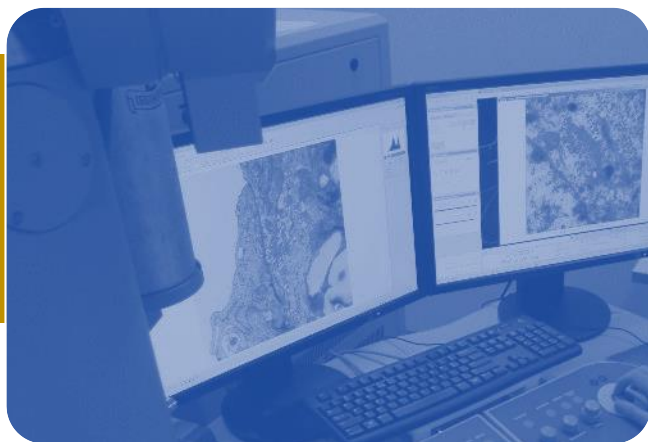
The CIPF Flow Cytometry and Cytomics Unit provides technological solutions for high-speed polychromatic analysis and cell separation, that along with its vast experience in cytomic analysis applied to biomedicine, biotechnology and translational medicine, allows us to offer users high-quality, expert experimental support. In addition, the Unit also has experience in the application of cytomics methods to the field of microbiology and environmental sciences.

In 2018, the Unit has participated in many different research projects related to sample immunophenotyping, antigen expression, cell cycle analysis, cytotoxicity, functional analysis of cell lines, primary culture and ex vivo samples, specific applications for clinical and biotechnology use of microorganisms, functional characterization and immunophenotyping of stem cells, analysis of real-time parameters (in flux), multiplex analysis of soluble proteins, cell separation for obtaining purified populations based on immunophenotype and/or specific features.

The Unit has extensive experience in functional and high content studies (HCA) using image analysis in adherent cells and tissue sections. One of the most relevant scientific projects during 2018 has been a collaboration for the use of molecular characterization of lung cancer propagating cells as a predictive tool and for the design of personalized therapies.

In addition, the service has organized many different training activities, such as the 1st edition of the workshop "Flow cytometry: Technical bases" (accredited by the Commission for Continuing Training of Health Professions), in close partnership with UVEG and INCLIVA Foundation, the 5th edition of the "International Summer School on Cytometry" of the European Society of Clinical Cell Analysis (ESCCA).

Transmission Electron Microscopy



Overview

Electron microscopy uses an accelerated electron beam which, upon impacting the sample, generates different signals that provide information about its atomic structure. In Transmission Electron Microscopy (TEM), transmitted electrons are detected to generate conventional, dark-field, high-resolution transmission images. Due to its high resolution power, TEM allows the development of useful applications in the fields of biomedical research, biotechnology and diagnostics.

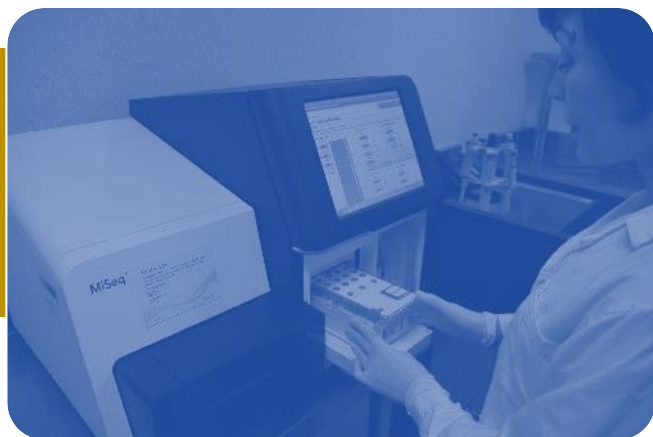
The Electronic Microscopy Unit at CIPF collaborates with internal and external research groups offering an invaluable tool to address some scientific questions. The Unit also provides technological support to pathology services for the diagnosis of some renal, muscular, dermal and peripheral nerve diseases, as well as for certain tumors.

In 2018, the Unit has participated in various research projects, such as a new collaboration with the stem cell department of the Andalusian Center for Molecular Biology and Regenerative Medicine (CABIMER) for the study of skeletal muscle of mice mutant (deficient) for Pax8 gene. Other project required the study of mitochondrial morphology, number of mitochondria, ultrastructure of the fibers (bands, triads, etc.) and the general appearance of adipocytes.

Additionally, the Unit has carried out during 2018 several new initiatives: set-up of new cryo-electron microscopy (cryo-EM) applications allowing the study of particles, cells and organelles in a quasi-native state, a new method for the analysis of individual particles (nanoparticles, macromolecular complexes, etc.) either isolated or purified, the optimization of a sample preparation protocol (vitrification), software control tools, or the development of protocols for the characterization of exosomes (negative staining).

In line with its history as a training and user capacitation service, the Unit has organized during 2018 multiple training sessions in TEM technology and applications for both internal and external staff. In addition, the service has hosted visits of secondary school students.

Genomics and Translational Genetics



Overview

The Genomics and Translational Genetics Service offers advanced genetic analysis and genomics applications. To achieve its main goal, the service integrates CIPF scientific advances in the field of human genetics, genomics and bioinformatics, with the knowledge generated through different collaboration projects and the experience in genetic diagnosis. This goal is to provide genetic information and tools to health professionals interested in the diagnosis and prognosis of hereditary pathologies.

In particular, at CIPF we focus on the study of hereditary peripheral neuropathies, hereditary ataxia/spastic paraparesis, neurodegenerative diseases related to iron accumulation in the brain, among other motion disorders. Using our growing scientific know-how and in collaboration with clinicians we have designed custom gene panels for the diagnosis of some of these neurological disorders.

In the area of genomics, we focus on supporting users in their microarray, qPCR and NGS experiments. Some of the most commonly used applications are: aCGH, differential expression, miRNAs identification and analysis, inter-individual genetic variation, epigenetic profiling, among others.

During 2018, the unit has reinforced and expanded its service portfolio. New diagnostic panels for peripheral hereditary neuropathies, in particular for distal spinal atrophy (AED), spinal muscular atrophy (SMA), Charcot-Marie-Tooth (CMT) and amio-trophic lateral sclerosis (ALS) have been designed and validated.

Another addition to the portfolio is the use of gene arrays applied to oncology (covering approximately 2,410 relevant regions in carcinogenic processes), autism (with more than 1,500 genes involved in developmental disorders and syndromes, including 150 genomic regions associated with autism) or other applications related to diagnostic genetics in general.

Nuclear Magnetic Resonance



Overview

The Nuclear Magnetic Resonance (NMR) Unit provides advanced applications for the characterization of the chemical structure of both small molecules and macromolecules and tools used to elucidate the molecular mechanisms underlying their biological activity.

The NMR Unit also designs and carries out applications and experimental designs for the development of drugs, being this one of its main expertise areas. Another interesting and increasingly demanded application is the characterization of the metabolomic profile in a varied group of biological samples (serum, urine, cerebrospinal fluid, cells, etc.). The Unit has been involved in many different projects in this area in close partnership with hospitals and other research institutions.

The Unit offers its solid experience in the characterization of different chemical compounds and macromolecules, in structural analysis of molecules and in the thorough interpretation of NMR spectra. This robust and wide background translates into numerous scientific collaborations. Throughout 2018 the NMR Unit has participated in a diverse range of projects and activities.

NMR technology has been applied at CIPF as an approach to the elucidation of the metabolomic profiles associated with different biological samples, each one of them representing a particular biochemical, pharmacological or pathological process.

It is worth mentioning as well the dedication of the NMR Unit to the organization of training and educational activities (such as the advanced course of MRI or metabolomics and the training of undergraduate and master students).

Proteomics



Overview

The Proteomics Unit manages a number technologies and equipment with the main goal of providing the CIPF scientific community, collaborators and other partners with the instrumental support and the scientific advice needed to carry out their proteomics projects.

The fundamental activity of the Proteomics Unit focuses on the support provided to researchers, both with technical aspects of sample analysis and providing experimental design or methodological advice on more general aspects related to proteomics and protein chemistry. One of the most relevant projects in which the Unit has participated during 2018 has been the study of the interactome of the transmembrane domains of Bcl-2 proteins and its potential role as an antitumor target.

Screening Platform



Overview

During the last few years, the activity of the Pharmacological Screening Unit has specialized in the development of specific trials for drug discovery and development, in areas ranging from new therapeutic strategies to fight cancer to regenerative medicine (stem cells) or infectious diseases.

One of the most significant projects in which the service has been involved is MyNano, a project conceived to study the role of exosomes in cancer and their suitability in the search for new anticancer drugs. In this particular field, we have worked on the identification of targets with potential synergy when administered with specific antitumor agents in melanoma models. Other projects include massive screening for the identification of activators of caspase 9 or the development of chemotherapy strategies based on combinations of endocrine active compounds for the treatment of hormone-dependent cancers.

The CIPF screening platform has been involved in the identification of targets associated with the maintenance of pluripotency and/or activation in different types of stem cells, including hematopoietic umbilical cord cells.

Other projects have used massive screening approaches based on polarized fluorescence techniques (for HIV) or screening for candidate identification in lung cancer.

In addition, the screening service is working on the development of massive screening assays for the identification of exosome modulators in human cell lines or for the identification of new FXN modifying genes in drosophila cell lines.

The unit participates in multiple projects and initiatives of organizations such as EU-Openscreen (European network of screening platforms) or REDEFAR (Spanish network for drug discovery), GEIVEX (Spanish Group for Research in Extracellular Vesicles), or those organized by SDDN (Spanish Drug Discovery Network).



Projects

Horizon 2020 - EU (*EU Framework Programme for Research and Innovation*)

| GRANT TYPE | TITLE | PRINCIPAL INVESTIGATOR |
|-----------------------|--|-----------------------------|
| Collaborative Project | MyNano: Towards the design of Personalised Polymer-based Combination Nanomedicines for Advanced Stage Breast Cancer Patients | M ^a Jesús Vicent |
| Collaborative Project | Exploiting Protein Complexes that induce Cell-death | Mar Orzáez |
| Collaborative Project | Ensuring long-term sustainability of excellence in chemical biology within Europe and beyond | M ^a Jesús Vicent |
| ERC Proof of Concept | Off the self polipeptide based immunotherapy for Advanced Melanoma Treatment | M ^a Jesús Vicent |

ISCIII - ES (*Instituto de Salud Carlos III*)

| GRANT TYPE | TITLE | PRINCIPAL INVESTIGATOR |
|-------------------------|---|------------------------|
| Research Platform | Caracterización molecular de células propagadoras de cáncer de pulmón como herramienta predictiva y para el diseño de terapias personalizadas | Rosa Farràs |
| Research Platform | Plataforma de Proteómica, genotipado y líneas celulares | Slaven Erceg |
| Research Platform | Plataforma de Bionformática | Ana Conesa |
| FIS Project | Avanzar en el diagnóstico, la prognosis y la terapia de enfermedades neurodegenerativas raras | Carmen Espinós |
| FIS Project | Estudio preclínico de terapia celular con progenitores neurales derivados de hESC e iPSC combinado con modulación de astrogliosis en tratamiento de lesiones medulares. | Slaven Erceg |
| FIS Project | Estudios clínicos, bases genéticas y biomarcadores pronósticos en enfermedades raras neurodegenerativas | Carmen Espinós |
| FIS Project | Desarrollo de un sistema integrado para la medicina personalizada orientado al diagnóstico y descubrimiento de genes de enfermedad usando secuenciación de nueva generación | Joaquín Dopazo |
| FIS Project | Estudio preclínico de potencias regenerativo de astrocitos derivados de células madre en tratamiento de lesión medular en ratón | Slaven Erceg |
| FIS Project | 3D retinas derivadas de células iPS como herramienta para encontrar terapias eficaces para enfermedades hereditarias de la retina | Dunja Lukovic |
| RETICS | Red de Trastornos Adictivos | Consuelo Guerri |
| CIBER | CIBER de Diabetes y Enfermedades Metabólicas Asociadas (Ciberdem) | Deborah J. Burks |
| Miguel Servet Programme | Contratación de doctores de acreditada trayectoria investigadora en centros del ámbito del SNS | Carmen Espinós |
| Miguel Servet Programme | Contratación de doctores de acreditada trayectoria investigadora en centros del ámbito del SNS | Slaven Erceg |
| Miguel Servet Programme | Contratación de doctores de acreditada trayectoria investigadora en centros del ámbito del SNS | Dunja Lukovic |

MSCBS - ES (*Ministerio de Sanidad, Consumo y Bienestar Social*)

| GRANT TYPE | TITLE | PRINCIPAL INVESTIGATOR |
|--------------------|---|------------------------|
| National Drug Plan | Neuroinflamación y alteraciones en la plasticidad cerebral en adolescentes con abuso de alcohol: Diferencias de género, biomarcadores y terapias. | Consuelo Guerri |
| National Drug Plan | Biomarcadores de neuroinflamación asociados al consumo de alcohol en adolescentes y nuevas terapias para paliar la neuroinflamación | Consuelo Guerri |

MINECO - ES (*Ministerio de Economía y Empresa*)

| GRANT TYPE | TITLE | PRINCIPAL INVESTIGATOR |
|---------------------------------|--|---------------------------------|
| Research Projects | Biohíbridos para la promoción del crecimiento axonal y la regeneración en el sistema nervioso central y periférico | Victoria Moreno |
| Research Projects | MicroRNAs como biomarcadores en la neuroinflamación asociada al abuso de alcohol: implicaciones diagnósticas y terapéuticas | Consuelo Guerri |
| Research Projects | Polímeros Terapéuticos diseñados para cruzar la Barrera Hematoencefálica para el tratamiento de desordenes neurodegenerativos-Explorando la Ruta Intranasal. | M ^a Jesús Vicent |
| Research Projects | Bases moleculares de las alteraciones neurológicas (cognitivas y motoras) en hiperamonemia y encefalopatía hepática. Implicaciones terapéuticas | Vicente Felipo |
| Research Projects | Modificación de tRNAs bacterianos y mitocondriales: funciones biológicas e implicaciones patológicas | M ^a Eugenia Armengod |
| Research Projects | Buscando una terapia para la epilepsia mioclónica progresiva de Lafora | Erwin Knecht |
| Research Projects | Bases moleculares de las alteraciones neurológicas (cognitivas y motoras) en hiperamonemia y encefalopatía hepática. Implicaciones terapéuticas | Vicente Felipo |
| Research Projects | Descifrando y modulando el interactoma transmembrana de las proteínas Bcl-2 como diana antitumoral | Mar Orzáez |
| Research Projects | Regulación de la adaptación y proliferación de células Beta por el Sustrato del Receptor de Insulina 2 | Deborah Burks |
| Research Projects | Señalización de NRG1 en circuitos corticales: información sobre las bases moleculares de la esquizofrenia | Pietro Fazzari |
| Retos Colaboracion | Desarrollo de una plataforma de terapia génica para enfermedades genéticas renales | M ^a Jesús Vicent |
| Retos Colaboracion | Desarrollo de terapias tópicas basadas en sistemas de transporte polipeptídicos | M ^a Jesús Vicent |
| Ramón y Cajal Programme | Incorporación de investigadores nacionales y extranjeros con una trayectoria destacada en centros de I+D | Pietro Fazzari |
| Formación Personal Investigador | Sonia Vicent - Polímeros Desarrollo de nanoconjugados para el tratamiento de cáncer de próstata resistente a castración | M ^a Jesús Vicent |
| Formación Personal Investigador | Angeles Arzalluz Luque - Nuevos métodos para los retos emergentes en el análisis de datos de secuenciación masiva | Ana Conesa |

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|---------------------------------|--|---------------------|
| Formación Personal Investigador | Raquel Garcia Garcia - Bases moleculares de las alteraciones neurológicas (cognitivas y motoras) en hiperamonemia y encefalopatía hepática. Implicaciones terapéuticas | Vicente Felipo |
| Personal Técnico de Apoyo | Nerea Marín Izquierdo – Ayudas a la Contratación de Personal Técnico de Apoyo | Ernesto de la Cueva |
| Juan de la Cierva Formación | Michele Malaguarnera – Ayudas a la contratación laboral de jóvenes doctores para completar su formación investigadora posdoctoral | Vicente Felipo |

MECD - ES (Ministerio de Educación, Cultura y Deporte)

| GRANT TYPE | TITLE | PRINCIPAL INVESTIGATOR |
|---------------------------|--|------------------------|
| Ayudas FPU | Jose Miguel Pardo - Contratos predoctorales para la realización de tesis doctorales. | Rosa Farràs |
| Ayudas FPU | Lorena de la Fuente - Contratos predoctorales para la realización de tesis doctorales. | Ana Conesa |
| Ayudas FPU | Lucas Taoro - Contratos predoctorales para la realización de tesis doctorales. | Vicente Felipo |
| Ayudas FPU | Paula Sancho - Complejidad Genética y fisiopatología de neuropatías hereditarias sensitivo y/o motoras | Carmen Espinós |
| Ayudas FPU | Paula Izquierdo - Contratos predoctorales para la realización de tesis doctorales. | Vicente Felipo |
| José Castillejo Programme | Leo Puchades – Ayudas para estancias que favorezcan e incentiven la movilidad de jóvenes doctores | Ernesto de la Cueva |
| Ayudas FPU / Estancias | Jose Miguel Pardo - Holanda | Rosa Farràs |

CEICE - GVA (Conselleria de Educación, Investigación, Cultura y Deportes)

| GRANT TYPE | TITLE | PRINCIPAL INVESTIGATOR |
|---|--|------------------------|
| PROMETEO Programme | Mecanismos moleculares y cerebrales de las alteraciones cognitivas y motoras en hiperamonemia y encefalopatía hepática. Implicaciones terapéuticas y diagnósticas. | Vicente Felipo |
| PROMETEO Programme | The next systems biology: desarrollo de métodos estadísticos para la biología de sistemas multiómica | Ana Conesa |
| Grupos Emergente - GV | Estudio del papel de la GTPasa RAB11 en cáncer de próstata para su uso como biomarcador | Ana Armiñan |
| Grupos Emergente - GV | Diseño de sistemas de transporte de fármacos intranasales como plataforma nanotecnológica para el tratamiento | Inmaculada Conejos |
| Grupos Emergente - GV | Organoides tumorales de cáncer de pulmón como modelo personalizado para estudiar la biología del tumor y la respuesta a fármaco | Carolina Gandia |
| Captación proyectos internacionales - APE | Diabetes and different co-morbidities Subvenciones para la captación de proyectos europeos u otros programas de carácter internacional | Vicente Felipo |
| Postdoctoral - APOSTD | Tiziano Balzano | Vicente Felipo |

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| Postdoctoral - APOSTD | Beatriz Pérez - Deconstrucción de las redes transcripcionales de JunB en la proliferación celular | Rosa Farràs |
| GRISOLIA Programme | Paola Leone - Papel de la inflamacion periferica y neuroinflamacion en el deterioro cognitivo y motor en encefalopatía hepática | Vicente Felipo |
| Predoctoral - ACIF | Irene Dolz Pérez - Desarrollo de Polímeros Terapéuticos para el tratamiento de la Piel | M ^a Jesús Vicent |
| Predoctoral - ACIF | Estefania Lucendo - El interactoma de los dominios transmembrana de las proteínas BCL-2 como diana antitumoral | Mar Orzáez |
| Predoctoral - ACIF | Fátima Manzano - El papel del substrato receptor de insulina 2 (IRS") en cáncer hepatocelular y su implicación en la heterogenidad intra tumoral. | Luke Noon |
| Predoctoral - ACIF | Jose Pardo Palacios | Ana Conesa |
| Predoctoral - ACIF | Maria Sancho Alonso | Vicente Felipo |
| Predoctoral - ACIF | M ^a José Arámbul | Luke Noon |
| Estancias Predoctoral - BEFPI | Irene Dolz Pérez - BERLIN | M ^a Jesús Vicent |

CSUIP - GVA (Conselleria de Sanidad Universal y Salud Pública)

| GRANT TYPE | TITLE |
|----------------------------|---|
| Internacionalización - AFI | Acciones dirigidas a impulsar y gestionar la participación en los programas de investigación internacionales en materia de biomedicina, sanidad y salud pública |

AVI - GVA (Agencia Valenciana de la Innovación)

| GRANT TYPE | TITLE | PRINCIPAL INVESTIGATOR |
|---------------------------------|---|------------------------|
| Proyectos colaboración - INNEST | Biomarcadores para oncología de precisión en cáncer de Pulmón, Colorrectal y Melanoma | Francisco García |

FOUNDATIONS & OTHER PRIVATE ENTITIES

| GRANT TYPE | TITLE | PRINCIPAL INVESTIGATOR |
|----------------------|--|-----------------------------|
| AECC - Predoctoral | Design of novel targeted Polymer Therapeutics as combination therapy for the treatment of Brain Metastasis- Overcoming the Blood Brain Barrier | M ^a Jesús Vicent |
| AECC - Predoctoral | Brain Drug Delivery using polymer therapeutics as intranasal platform towards paediatric glioma treatment | M ^a Jesús Vicent |
| AECC - Predoctoral | Nuevas estrategias terapéuticas contra el cáncer de pulmon basadas en el control de la síntesis de proteínas mediada por pliaminas | Rosa Farràs |
| AECC – Prácticas Lab | Alejandro Vegas Campos | Ernesto de la Cueva |

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| AECC – Prácticas Lab | Mario García | Jerónimo Forteza |
| AECC – Prácticas Lab | Microentorno | Francisco García |
| AFM | CORRET: Cell therapy with genetically corrected retinal pigment epithelium in hereditary retinal dystrophies | Slaven Erceg |
| AFM | An integrative approach to develop cellular models and characterize disease mechanisms implicated in CMT2Z, a newly described axonal form of neuropathy | Carmen Espinós |
| ASEICA - Premio | Identificación de nuevos biomarcadores y desarrollo conjugados poliméricos de combinación en cáncer de próstata metastásico | M ^a Jesús Vicent |
| La Marató de TV3 | Transplant of combined cell therapy from clinical grade iPSC-derived cells with neuroprotective small chemicals in a SCI rat model for central regeneration of spinal pathways | Victoria Moreno |
| La Marató de TV3 | Dissecting protein trafficking in retinal neurodegeneration by super-resolution imaging on animal models and human iPSCs | Slaven Erceg |
| La Marató de TV3 | Neurodegeneration with Brain Iron Accumulation: Clinical Assessment and Genetic Characterization by means of a Spanish Multi-Centre Research Network | Carmen Espinós |
| La Marató de TV3 | Combinatory treatment of Neural precursor cells and a new nanoconjugate of Fasudil for the clinical application in Acute Spinal Cord Injury | M ^a Jesús Vicent |
| INDACEA | Medicina de precisión en síndrome de Dravet | Ibo Galindo |
| PROYECTO DRAVET | Generación de modelos en Drosophila melanogaster mediante knock-in de mutaciones de pacientes | Ibo Galindo |
| FPAW-Wilson | Avanzar en el diagnóstico, la prognosis de la enfermedad de Wilson | Carmen Espinós |
| INDACEA | Validación e implementación de un herramienta diagnóstica | Carmen Espinós |
| Fundación Ramón Areces | Caracterización de MORC2, nexo de unión de neuropatías hereditarias sensitivo-motoras | Carmen Espinós |

RESEARCH CONTRACTS

| ENTITY | TITLE | PRINCIPAL INVESTIGATOR |
|-------------|--|------------------------|
| LAB. ESTEVE | Biohíbridos para la promoción del crecimiento axonal y la regeneración en el sistema nervioso central y periférico | Victoria Moreno |
| BIONOS | Study of anti-oxidant in retina of diabetic mouse model | Deborah Burks |
| BIONOS | Estudio del efecto sobre la autofagia de un anti-oxidante en fibroblastos de pacientes con la enfermedad de Lafora | Erwin Knecht |
| BIOMAR | Asesoramiento y Apoyo Tecnológico | Ibo Galindo |
| VIVANTA | Regeneración de tejido periodontal con células madre, factores de crecimiento y biomateriales | Victoria Moreno |

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|----------------------------------|--|-----------------|
| SPIRAL THERAPEUTICS | Characterization of Apoptosome Inhibitors | Mar Orzáez |
| GRUNRNTHAL PHARMA | Traumatic spinal cord injury pain | Victoria Moreno |
| FLORIDA UNIVERSITY | Realización de análisis integrador de datos transcriptómicos y metabolómicos | Ana Conesa |
| NATIONAL INSTITUTE OF BIOLOGY | Realización de análisis integrador de datos transcriptómicos y metabolómicos | Ana Conesa |

ANNUAL REPORT 2018

FUNDACIÓN DE LA COMUNIDAD VALENCIANA CENTRO DE INVESTIGACIÓN PRÍNCIPE FELIPE



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