BrainVectors Newsletter n° 1

Marie Curie Industry-Academia Partnerships and Pathways - Contract n° 286071

The *BrainVectors* project

Welcome of the coordinator

Dear All,

I have the pleasure in this first *BrainVectors* newsletter to welcome all the PIs of the consortium and the two recruited researchers, **Marie Humbert-Claude**, **Ludivine Breger** and **Felix Junyent-Herena**. I wish to Marie, Ludivine and Felix to achieve the best in their research and that their tenure in Lausanne and Lund, respectively, will be a great opportunity for their career. I wish to say also welcome to all the 26 researchers staff members of the partners' labs, who will visit the partners' laboratories along the 48 months and implement thus the core *BrainVectors* research program.

I have the privilege to be the coordinator of this exciting network of cutting-edge academic and industrial laboratories that are specialist of gene transfer into brain by using vectors derived adeno-associated viruses, canine adenovirus and lentiviruses. Our common approach is to drive gene transfer by switching on/off the expression of foreign genes delivered into cells and tissues, analyse the effects of such expression in 2D/3D cells systems and mice and assess the immune responses at cellular and molecular levels. Our goal is to establish the pharmaceutical settings of our vectors to use them in novel gene therapy-based protocols for Parkinson's disease and other neuropathies.

Behind this, we shall strive to open the borders of the academia environment to the industry and vice versa, by transferring from one to the other sector different approaches, working methods and career prospects for researchers. Only with this *open-innovation* vision, it will be possible to bridge the two environments and, thus, to overcome the several questions that remain still open in the translational neurobiology research today.

In this Newsletter, I also invite you to communicate between each other, share information and materials, open your lab to the visiting researchers to exchange experiences and create with them unique opportunities to achieve new knowledge and innovative applications. You have started already this intra-network interaction with the first secondment of **Stefania Piersanti** (UniRM), who has spent two months in IBET in Oeiras, and with **Gloria Gonzales** (FIMA), who will collect cell lines from all partners to study her vectors in them. I am delighted in seeing such a dynamism and cooperation already and I am proud to be the coordinator of a network where the colleagues show initiative and enthusiasm.

I invite you also to implement training the initiatives of **Mauro Mezzina** (EASCO), who will develop the *BrainVectors* educational and dissemination program in organizing meetings and delivering individual coaching to researchers. Mauro's experience will help us to work collectively and to spread outside the consortium the new knowledge acquired.

Finally, I invite you to come to Madrid this October 24th and 25th, 2013 for our first forthcoming *BrainVectors* consortium meeting and I look forward to meeting you for important interactions and discussions in Madrid in October.

With warm regards,

Liliane

Partners profiles

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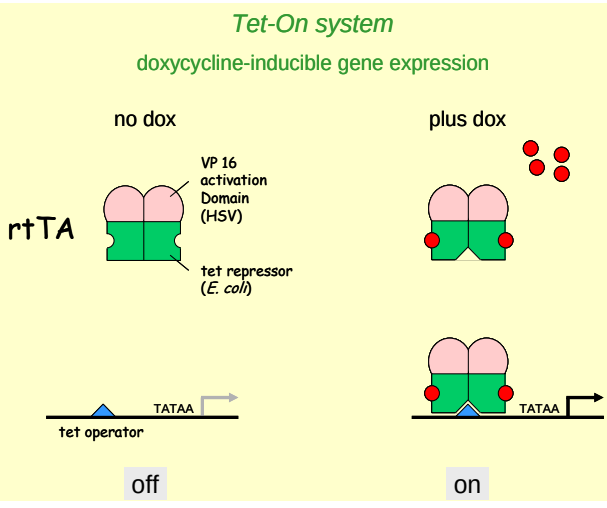
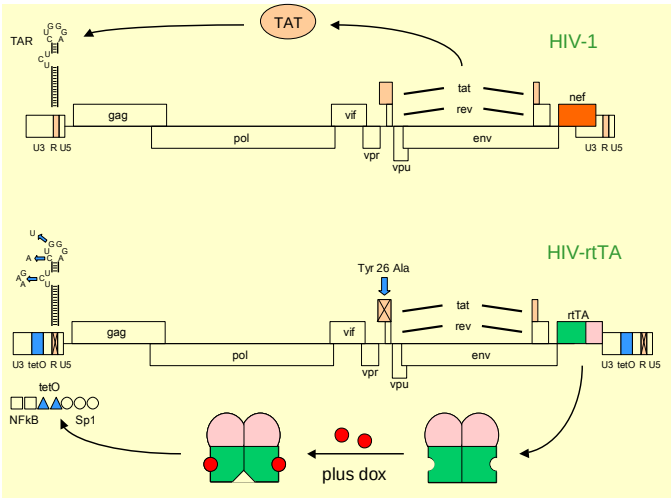
RESEARCH: Technology for the regulation of gene expression in mammalian cells and tissues is essential for both functional genomic research and applications in medicine and biotechnology. The most widely used regulatory circuit is the so-called Tet-On system that is based on the E. coli tetracycline repressor protein (TetR) and doxycycline (Dox) as effector molecule. We previously constructed a genetically modified HIV-1 virus variant of which the gene expression and replication is controlled by the Tet-On system. This HIV-rtTA variant can be turned on and off at will by the addition or removal of Dox, respectively. It was developed as a live-attenuated vaccine candidate and is currently being tested in the SIV-macaque AIDS model. We use this unique HIV-rtTA virus as a tool to develop new Tet-On systems. For example, we developed systems that are more active and more Dox-sensitive, which makes them very suitable for applications in brain and other tissues where reduced Dox levels can be reached.





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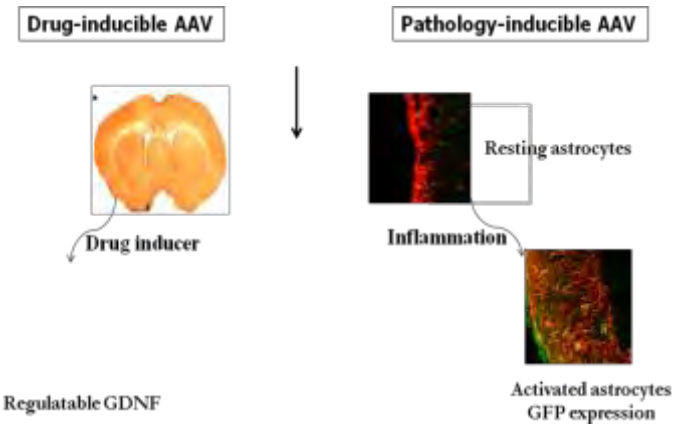
RESEARCH: The group develops viral vectors based on AAV for regulatable and specific gene expression in neuroprotective and anti-inflammatory therapeutic strategies for Parkinson's disease. The vectors are evaluated *in vivo* in phenotypic rodents models for Parkinson's disease.

Another goal of the group is to develop pathology-inducible vectors to monitor disease progression and therapeutic interventions *in vivo*. Recombinant AAV vectors of various serotypes (1,2,5,8,9) are produced by co-transfection of HEK-293T cells, purified by iodixanol gradients and titrated to determine viral genomes and capsids using described methods (Lock et al., Hum.Gene Ther. 21:1273-85, 2010).



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A.Chtarto, X.Yang, O.Bockstael, C.Melas, D.Blum, J.-M.Jaspar, M.Levivier , J.Brotchi, T.Velu and L.Tenenbaum .(2007). "Controlled delivery of glial cell line-derived neurotrophic factor by a tetracycline-inducible AAV vector". Exp. Neurol. 204(1):387-399.

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RESEARCH: The clarification of virus-cell interactions may be the most exciting aspect of modern virology, revealing similarities among virus families, the diversity of tropism, trafficking, and pathogen propagation. Our core theme is the study of Adenoviridae. Human adenoviruses normally cause subclinical symptoms, but in sporadic cases they can be lethal in infants and immunocompromised patients. Our approaches transverse disciplines as diverse as virology, cellular and molecular biology, immunology, physical biochemistry, neurology, biochemistry, and gene transfer.

The Kremer lab 2013

http://www.igmm.cnrs.fr/spip.php?rubrique35

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The team:




Cecilia Lundberg, PI Ludivine Breger Luis Quintino Erika Elgstrand Wettergren

and Christina Isaksson, technical engineer (not shown)

http://www.med.lu.se/expmed/cns_gene_therapy

RESEARCH: Development of gene therapy vectors to treat neurodegenerative disease and damage in the CNS. The goal of our research is to develop vectors for use in gene therapy to treat diseases in the brain. Furthermore, we also aim to explore ways to modify CNS function by gene transfer.

In our lab we are constantly advancing the vector production and constructions to achieve as effective and specific lentivectors as possible.

One of our objectives is to develop cell-specific recombinant viral vectors to treat the hallmarks features of Parkinson's disease (PD) such as dopaminergic cell death and L-DOPA induced dyskinesia. To achieve this goal we will use and further refine the cell-specific lentiviral vectors developed by our group during the last couple of years. Thus, we are constructing lentiviral vectors specific for PD by analyzing expression patterns in the PD striatum. We are combining our cell-specific technology with the possibility to down regulate proteins using RNA interference. In these projects we are not only developing tools to study the patophysiology of PD but also investigating possible new treatment paradigms for PD and proof-of-concepts that can be transferred to the study and treatment of other brain disorders.

Another objective for our research is to develop lentiviral vectors that can be regulated. This can be achieved either by giving inducing drugs or by utilizing the natural regulation of promoters in our vectors. These vectors will be improved for further use in the PD models.

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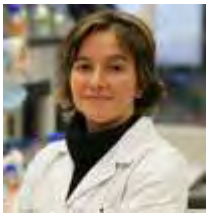
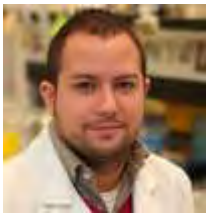




Jose Luis Lanciego Gloria Glez-Aseguinolaza Alberto Rico Daniel Moreno

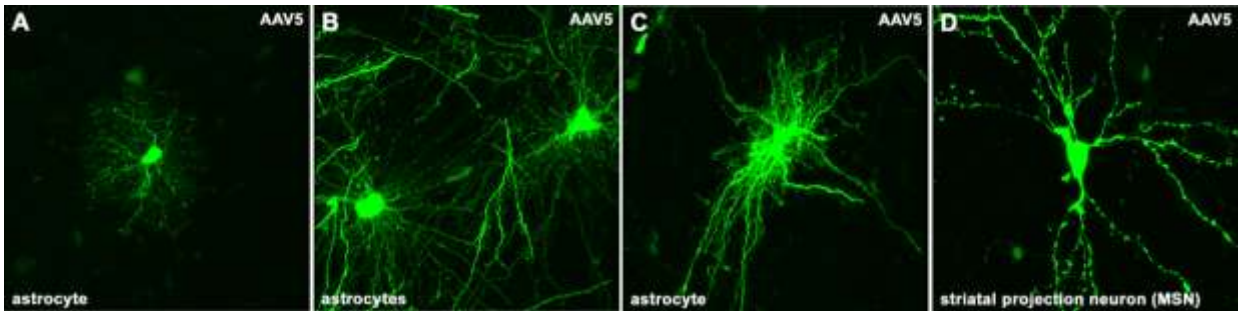
and Jose Diego Pignataro (not shown).

http://www.cima.es/labs/ver/14/neuroanatomia-ganglios-basales/

http://www.cima.es/labs/ver/49/terapia-genica-hepatitis-virales-lab-302/

RESEARCH: Neurodegenerative diseases cause a significant burden on the elderly population in Europe. Parkinson's disease (PD) affects 1.2 million people in Europe and with the increasing life expectancy this number will rise, putting more pressure on health care. Treatment of PD is only symptomatic, and therefore, there is an urgent need for more efficient therapies. With this goal in mind our group is working in four research area:

- The study of basal ganglia circuits underlying the pathophysiology of Parkinson's disease using animal models of this neurodegenerative disease (rodents and primates).
- Screening of G-protein-coupled receptor (GPCR) heteromers in basal ganglia nuclei. Synthesis and validation of newly designed drugs targeting GPCR heteromers in the monkey model of PD.
- Development of central nervous system specific tetracycline inducible AAV vectors.
- Controlled release of neurotrophic factors (GDNF and relatives) using either AAV-inducible viral vectors.



Legend to figure: Following the delivery of a GFP-carrying AAV5 into the post-commissural putamen of the monkey, both astrocytes (A-C) and striatal MSNs (D) are infected and highly express the reporter.

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RESEARCH: GenIbet is a biopharmaceutical CMO (Contract Manufacturing Organization) offering highly specialized microbial, cell culture and viral propagation process development and cGMP manufacturing services to research groups, biotech and pharma companies.GenIbet's core activity is the manufacture and supply of materials for use in early stage drug development, pre-clinical studies and cGMP manufacturing for Phase I and II clinical trials.

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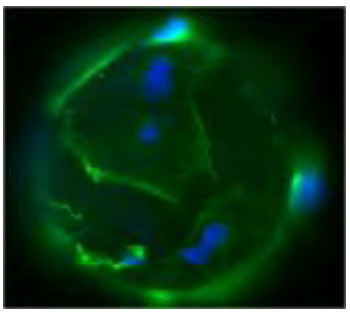
RESEARCH: Our research falls into 2 main areas : complex bio-molecules for diagnostics and therapeutics, such as recombinant proteins including MAbs, virus like particle (VLP) vaccines and vectors for gene therapy (retro-, lenti-, adeno- and, more recently, AAV) ; development of methodologies for pre-clinical research and cell therapy/regenerative medicine, namely in 3-D cell models and stem cells. In both areas, our goal is the development of highly productive bioprocesses and shorter development timelines by studying the mechanisms underlying cell growth, cell metabolism and product formation to rationally develop and optimize production processes.

The team:



mesenchymal stem cells on microcarriers

http://www.itqb.unl.pt/labs/animal-cell-technology

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Biopharmaceutical and Novel Therapies Services we provide to Academia and Industry:

Animal Cell Technology

• Cell Line Development

• Bioprocess Development for Recombinant Proteins, mAbs, Vaccines and Viral Vectors for Gene Therapy

• Upstream and Downstream Design & Development

• In vitro 3D Models for Pre-Clinical Research

• Stem Cells for Cell Therapy & Drug Discovery

Biopharmaceuticals: Discovery, Production and Characterization

• Pilot Plant Unit (Bioprocess Development and Production up to gram quantities) - Access to GMP production

• cGMP Analytical Services: Cell Based Assays, Mass Spectrometry; Glycosylation and Characterization

• 3D protein structure determination by X-ray crystallography and/or NMR

• Molecular modeling applied to protein structure and enzyme kinetics

• NMR spectroscopy for drug screening and hit optimization

• New targets and more efficient combinatorial therapies

Drug Delivery

• Nano/Micro Encapsulation & Impregnation

• Release kinetics

• Formulation

Biochemical Engineering Tools

• Novel Tools for Extraction and Process Monitoring

Modeling of Downstream Processing: in Particular Porous Systems

• Systems Biology for Cell Culture and Process Development

• Synthetic Biology Applied to Bioseparation Processes

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RESEARCH: The research activity of A Bosch's group is focused on the development of gene therapy strategies for diseases affecting the nervous system, both central (lysosomal storage diseases) and peripheral (pain, genetic and acquired neuropathies) and on the elucidation of the molecular mechanisms implicated in the development of these pathologies combining the use of animal models, tissue cell culture and gene therapy viral vectors.

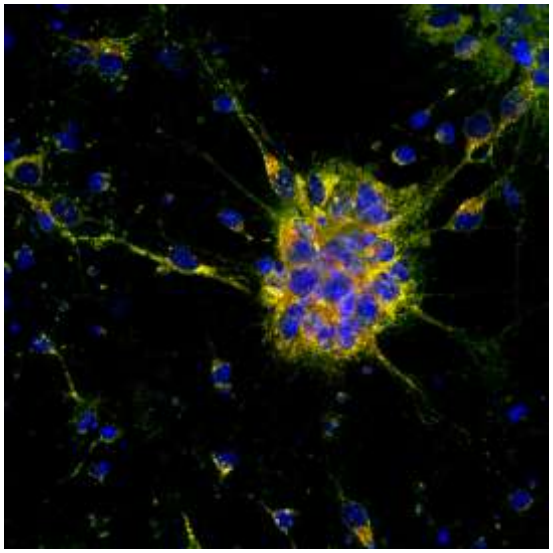

Figure 1: Primary cultures of cortical mouse neurons are used to characterize the molecular pathology of neurodegenerative diseases like Mucoplysaccharidosis type VII.

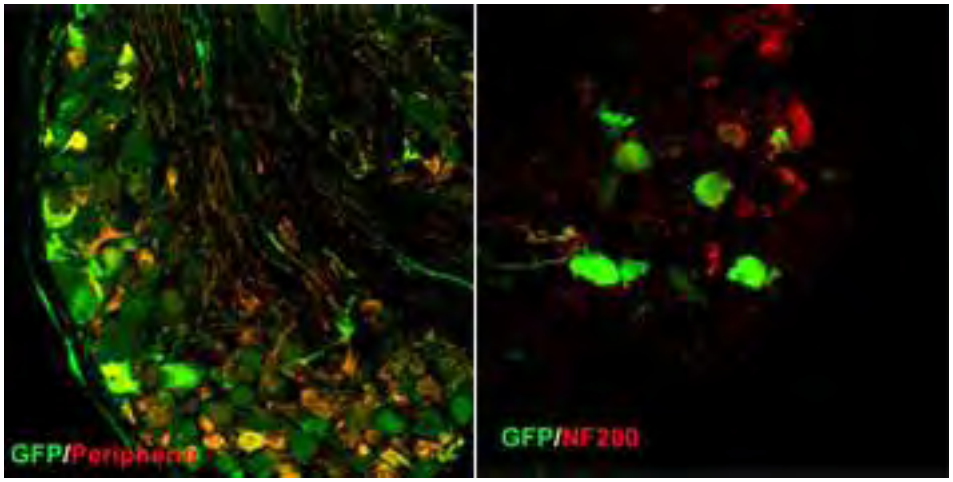

Figure 2: AAV transduction in DRG organotypic cultures target only sensory neurons. Different subpopulations of sensory neurons can be targeted with different serotypes of AAV vectors.

The team:

http://bioquimica.uab.es/paginas_cat/recerca_grups.php?idarea=6&nomarea=TERAPIA%20G%C8NICA%20I%20MODELS%20D%27ANIMALS%20TRANSG%C8NICS

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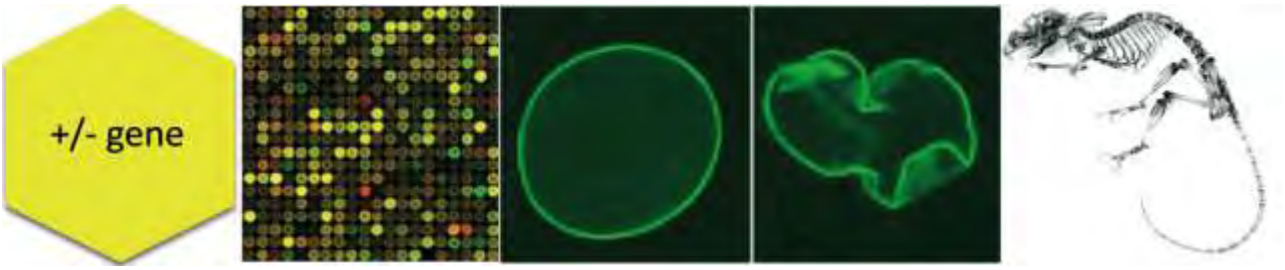
RESEARCH: We develop two research lines, one centered on **a)** viral gene therapy vectors and one **b)** studying **"**telomeraging" biology**.**

a) We analyze by single gene, single pathway and high throughput analysis the modulation events induced by different viruses, including AAV, lentivirus, adenovirus. We also use viral vectors to model human diseases which help designing therapeutic strategies and contribute to the understanding of disease mechanisms.

b) We study mammalian genes implicated in genome/telomere metabolism. We are specifically interested in ageing mechanisms at the molecular, cellular and organismal levels.



The team:

www.saggiolab.co

1. Piersanti S, Astrologo L, Licursi V, CostaR, Roncaglia E, Gennetier A, Ibanes S, Chillon M, Negri R, Tagliafico, Kremer EJ, Saggio I. (2013) Differentiated neuroprogenitor cells incubated with human or canine adenovirus, or lentiviral vectors have distinct transcriptome profiles. Plos One 26 (8) e69808.

2. Piersanti S, Tagliafico E, Saggio I. (2013) DNA microarray to analyze Adenovirus-host interactions. Methods in Molecular Biology Humana Press. In press.

3. Cherubini G, Naim V, Caruso P, Burla R, Bogliolo M, et al. (2011) The FANC pathway is activated by adenovirus infection and promotes viral replication-dependent recombination. Nucleic Acids Research 39: 5459-5473.

4. Caruso P, Burla R, Piersanti S, Cherubini G, Remoli C, et al. (2009) Prion expression is activated by Adenovirus 5 infection and affects the adenoviral cycle in human cells. Virology 385: 343-350.

5. Sacchetti B, Funari A, Michienzi S, Di Cesare S, Piersanti S, et al. (2007) Self-renewing osteoprogenitors in bone marrow sinusoids can organize a hematopoietic microenvironment. Cell 131: 324-336.

FIRALIS

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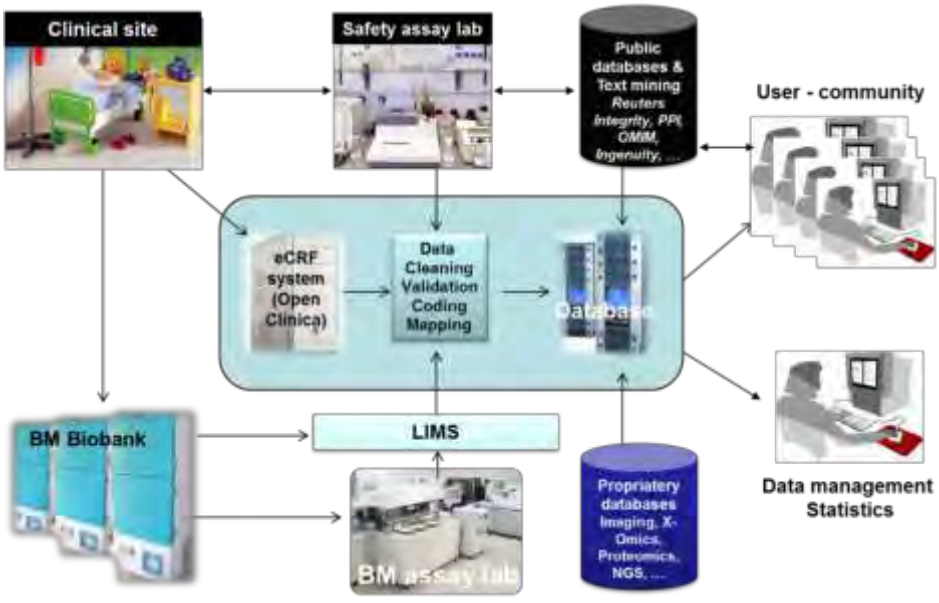
FIRALIS is a life sciences company developing innovative biomarkers (BM) for clinical use, with a specific focus on the inflammatory, auto-immune and cardiovascular fields. FIRALIS' mission is to create value via BM science, through discovery and translational development of BMs & BM-based diagnostic tools toward the bedside.

FIRALIS offers a wide spectrum of biomarker services from discovery to the clinical qualification; also research-use-only kits are developed and serve as a starting point to validate clinical utility as *in vitro* diagnostic devices. The worldwide leadership of FIRALIS as a respected expert in the biomarker field is proven by several major EU funds granted since its creation in 2008 such as the consortia SAFE-T, MitoCare, Imagint, Fibrotarget,…

Firalis clinical biomarker qualification platform

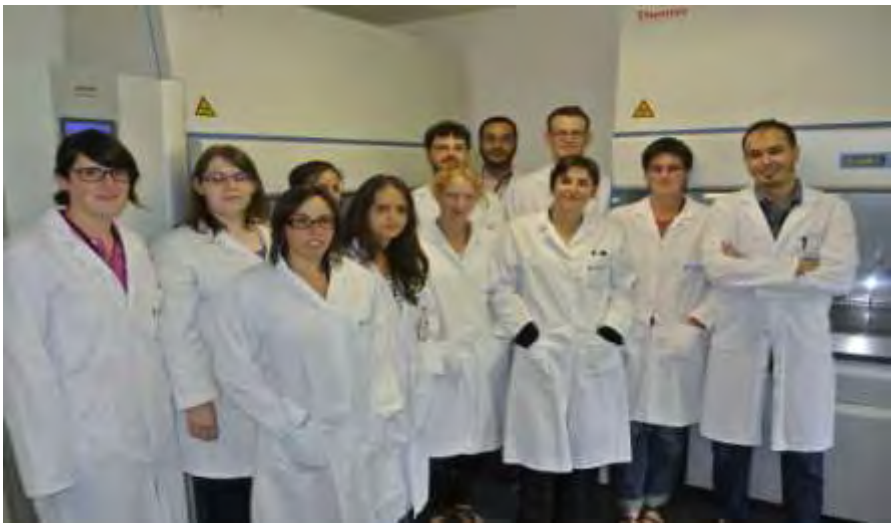
The qualification of biomarkers requires close cross-talks between numerous fields of expertise. To achieve this, Firalis has created a network of clinical centers of excellences around its technical platform. The laboratory comprises an ISO 17025 accredited bioanalytical platform, a biobank of extensively annotated samples, and a statistic and data management group. The various groups are organized around a state-of-the-art infrastructure including a laboratory information management system (LIMS) and a biomarker oriented clinical database. This organization promotes a strong synergy across disciplines to accelerate the translation of candidate biomarkers toward qualification in key contexts of use.



Firalis team operates in a high quality environment for each of its activities:

- ISO 17025: for testing and calibration laboratories
- ISO 9001: Quality Management Systems
- ISO13485: Assay development activities
- NFS96-900F: Quality of biological resource centers (BRCs)

The team:



Relevant publications :

- Altar et al., Rationale and design of the 'MITOCARE' Study: a phase II, multicenter, randomized, double-blind, placebo-controlled study to assess the safety and efficacy of TRO40303 for the reduction of reperfusion injury in patients undergoing percutaneous coronary intervention for acute myocardial infarction.Cardiology 2012;123:201–207
- Matheis et al., A generic operational strategy to qualify translational safety biomarkers. Drug Discov Today. 2011 Jul;16(13-14):600-8
- Vidal I, Blanchard N, Alexandre E, Gandillet A, Chenard-Neu MP, Staedtler F, Schumacher M, Bachellier P, Jaeck D, Firat H, Heyd B, Richert L. Improved xenogenic hepatocyte implantation into nude mouse liver parenchyma with acute liver failure when followed by repeated anti-Fas antibody (Jo2) treatment. Cell Transplant. 2008;17(5):507-24.
- Hartmann et al., Gene expression profiling of skin and draining lymph nodes of rats affected with cutaneous contact hypersensitivity. Inflamm Res. 2006; 55(8), 322-34Ginhoux F, Turbant S, Gross DA, Poupiot J, Marais T, Lone Y, Lemonnier FA, Firat H, Perez N, Danos O, Davoust J. HLA-A*0201-restricted cytolytic responses to the rtTA transactivator dominant and cryptic épitopes compromise transgene expression induced by the tetracycline on system. Mol Ther. 2004 Aug 10(2):279-89.

Profiles of the recruited researchers

BREGER Ludivine

Lund University, Lund, Sweden

Nationality: French

Age: 29

Date of recrutement: 01 May 2013

ludivine.breger@med.lu.se

Flash CV

Post-doctorate researcher (since 2013):
Wallenberg Neuroscience Centre, Department of Experimental Medical Science, Lund, Sweden.

Ph.D. (2010 - 2013): School of Pharmacy and Pharmaceutical Sciences, Cardiff, UK.

Thesis title: *Parameters impacting the outcome of cell replacement therapy for Parkinson's disease: a preclinical study*.

M.Sc. (2009): specializing in Tissue, Cell & Gene Biotherapies, University of Evry , France.

B.Sc. (2007): specializing in Biology, University of Evry, France.

My past research

My academic career has been driven by a strong interest in neurodegenerative diseases and translational research. Following a Bachelor's degree in Biology and Physiology, I completed a Master's degree specializing in Cell and Gene Biotherapies. Being fascinated by the potential of cell therapy, I chose to perform the main part of my practical training in the Institute for Stem cell Therapy and Exploration of Monogenic diseases (I-STEM). In parallel, I began to engage in scientific communication for the public. I was awarded a Ph.D. scholarship from Cardiff School of Pharmacy and Pharmaceutical Sciences to work on cell therapy for Parkinson's disease. My project focused on reducing the risk of adverse effects following cell transplantation. This work generated important data that I presented at international conferences and led to the publication of a review and two scientific articles in well-

respected, peer-reviewed journals (with a third one in preparation). Along with my passion for research, teaching is also a serious interest of mine, for which I received the Higher Education Academy recognition as an associate fellow. My Ph.D. studies also gave me the opportunity to further develop my communication skills, regularly engaging with patients' groups to inform and educate them about latest technologies and scientific breakthroughs.

My research in BrainVectors

Having developed a strong interested in Parkinson's disease during my Ph.D., I whished to carry on working in this field but sought to further develop my technical skill by studying molecular biology. I therefore, joined Prof Cecilia Lundberg's team a year ago to work on the BrainVectors project. My project is to generate a bidirectional Tet-On regulated glia derived neurotrophic factor (GDNF) lentiviral vectors, as well as appropriate controls (namely luciferase and green fluorescent protein) and test their expression *in vitro*. The bidirectional Tet-On regulated GDNF cassette was designed based on the construct chosen our collaborators in Lausanne, Switzerland. Expression and inducibility of the vectors will be tested in cell culture before being sent to collaborators for *in vivo* assessment. The second main goal is to produce lentiviral vectors for cell-specific expression using a 2-vectors inducible system. To that purpose, co-transfection of 2 vectors will be used: 1) rtTAV16 construct under control of a cell specific promoter, 2) GFP transgene controlled by the tetracycline responsive element.

My career objectives

Research and teaching constitute my main professional interests. Consequently, my long-term goal is to obtain a lectureship in one of the European leading research centre for neuroscience and become a world-expert researcher in neurodegenerative diseases. Although I am at an early stage of my career, my ambitious and pro-active nature has allowed me to build the strong foundation of an international scientific career, along with valuable teaching experience. After completion of the BrainVectors project, I will apply for an international Marie Sklodowska Curie outgoing fellowship. If successful, this would be a major step in the pathway to independence an it would allow me to expend my professional network outside of Europe.

HUMBERT-CLAUDE Marie
CHUV, Lausanne

Nationality: French

Age: 35

Date of recruitment: 01 July 2013

marie.humbert-claude@chuv.ch

Flash CV

- Experience

July 2013 – Sep 2015: **Post-Doctoral position in Gene Therapy (2 years)**
Laboratory of Gene therapy for Parkinson's disease, CHUV, Lausanne, Switzerland.
July 2011 – Sept 2012: **Post-Doctoral position in Neurobiology (1 year and 3 month)**
Laboratory of Neurobiology,CNRS UMR 7637, ESPCI ParisTech Paris, France.
Nov 2009 – Dec. 2010: **Hospital Pharmacist, (1 year),** *Vevey, Switzerland.*
2003-2004 – 2005-2008: **Pharmacist Internist, (4 years,** in parallel of the PhD) *Paris, France.*

- Experimental skills

- Pharmacology: Bindings and activity measurements on G-protein-coupled-receptor (GPCR).
- Cell biology: Cell culture, primary neuronal cultures, transfections, videomicroscopy.
- Vectorology: AAV production, validation, injection (P2-safety).
- Animal tests: Stereotaxic experiments: Rat models of Parkinson's disease and Behavioural tests.

- **Imaging skills:** Magnetic Resonance Imaging (MRI) on rat brain: Anatomic and DTI.

- Education

❑ ***Life Science Education***
2005 – 2010 **PhD in Neuropharmacology** (*INSERM U894, Paris, France)*.
2004 – 2005 **Master 2 of Pre-Clinical and Clinical Pharmacology** (*Paris, France).*
2003 **Master 1 of Science in Biological and Medical Sciences** *(Toulouse, France).*
❑ ***Pharmaceutical Education***
2009 **Hospital Pharmacist Degree***, Hospitals in Paris, France.*
2003 - 2008 **Pharmacist Internist,** *Hospitals in Paris, France.*

1997 – 2003 **Pharmaceutical studies,** *Paul Sabatier University (U.P.S.) Toulouse, France.*

My past research

I did my PhD in the Psychiatry and Neurosciences Center, INSERM, Paris, in the Laboratory of Neurobiology and Molecular Pharmacology directed by Dr Jean-Michel Arrang. I investigated new target in order to developed new drug candidate in neuropsychiatry. Notably, I showed that the activation of the dopaminergic D_2R and histaminergic H_3R receptors in the brain is additive (Biochem. Pharmacol., 2007). The second focal point of my studies leads me to clozapine, an atypical antipsychotic, and its effects on the histaminergic system. I used radioligands binding on *ex vivo* brain tissues and second messenger assay on cell culture. This work demonstrated that clozapine interacts with the four histamine receptors at clinically relevant concentrations. It may explain the atypical antipsychotic profile of clozapine, as well as its side effects (Psychopharmacology, 2012). Finally, I investigated the role of the histamine H_3 receptor as a potential target in the treatment of Parkinson's disease. I explored the effects of ciproxifan,

a H_3 ligand, on a model of Parkinson's disease: I did stereotaxic injections of 6-OHDA into rat brain and evaluated akinesia on behavioural tests. We observed a partial improvement of the initiation of movement in two different tests (stepping test and limb-use asymmetry test) (article submitted in Neuroscience).

In parallel of my PhD, my hospital pharmacist position allowed me to work in hospitals and in a close relationship with clinicians, which provides me a strong knowledge about diseases, drugs and patient's care.

Then, I did my post-doctoral position, in the Laboratory of Neurobiology of ESPCI, CNRS, Paris, in the Zsolt Lenkei`s team. I worked on the relationship between the cannabinoïd receptor type-1 (CB_1R) and the neuronal plasticity. In collaboration with physicists, I studied the impact of CB_1R activation on neuronal connectivity using Magnetic Resonance Imaging (MRI) on rat brain (Poster for 26th Congress of ECNP). I also contributed to the assessment of the effect of CB_1R agonists on the development of the axons, by perfoming videomicroscopy on primary neuronal cultures from rat hippocampus (Article submitted to ELife).

My research in BrainVectors

Today, I am working for BrainProject, a European project involving 11 European partners, aimed at assessing the efficacy and safety a new treatment for Parkinson's disease. This work is performed in the team "gene transfer for Parkinson's disease", Centre Hospitalier Universitaire Vaudois, Lausanne. During the first year, I dedicated my work to the production of AAV-derived viral vectors that deliver hGDNF in response to low doses of doxycyline and with an undetectable background in absence of doxycyline. This work included the genetic construct, production, purification, dosage and functional validation of the vector. In a close collaboration with Genibet (Portugal), we are making viral vectors that present high criterion of purity. Moreover, in order to avoid any risk of potential immune response, a second transactivator "V16im" has been provided by the Univers:ity of Amsterdam.

I participate to the production and validation of these new generation of vector s: AAV1/2-V16im-hGDNF, compared to a control vector, the AAV1/2-V16im-GFP. The next step is to assess the immune profile of these vectors in humanized mice .

JUNYENT Felix
IGMM-CNRS, Montpellier
Nationality: Spanish
Date of recruitment: November 2013
Age: 33
felix.junyent@igmm.cnrs.fr

Flash CV

2004- Graduate in Biology, University of Barcelona, Spain

2008- PhD in Biomedicine, University of Barcelona, Spain

2008-2013- Postdoctoral position in CIBERNED, Barcelona, Spain

2009-2013- Associate lecturer, Department of Biochemistry, Faculty of Medicine, Universitat Rovira i Virgili

42 peer-reviewed papers in international journals in neuroscience field and 9 book chapters

My past research

During my PhD I was involved in the study of taurine in the brain. Specifically, I studied the neuroprotective role of taurine in an experimental model of epilepsy based on kainic acid injections. After my PhD I did a postdoc during five years where I was studying different pathways involved in neuronal apoptosis. Mainly, my research project during that period was to determine the specific role of the three different c-jun N-terminal kinases (JNK) in neurodegeneration, using the different knock-outs for these isoforms.

My research in BrainVectors

Since I started in the host institution I've been involved in the production of CAV-2 virus expressing eGFP, luciferase or GDNF under the control of Doxycycline.

The pAC1-V16-rTA-eGFP, pAC1-V16-rTA-luciferase, pAC1-V16-rTA-GDNF construtcs have been cloned in pTCAV12a vector and after we generated the pCAV with the constructs by homologous recombination.

After, the virus CAV-rTA-eGFP, CAV-rTA-luciferase and CAV-rTA-GDNF have been produced in DKZeo cells.

Now, I'm involved in testing the virus that we have produced in the lab in vitro and in vivo. We will test the virus in 293T cells without and with doxycycline at different concentrations. We will also infect primary neuronal cultures.

Moreover, the virus produced will be injected in the mice brains at different regions and different concentrations of doxycycline will be test to determine the inducibility of gene expression

My career objectives

While having a strong background in neuroscience and in neurodegenerative diseases, the research and technical skills that I'm acquiring in BrainVectors project will allow me to further diversify and extended my knowledge, mainly in gene therapy tools that can be useful for the treatment of neurodegenerative diseases. That will allow me to broaden my views and give me the opportunity to consider the applicability of the techniques that I'm using to develop new therapeutic strategies for neurodegenerative diseases (gene therapy).

Together will help me to reach a level of maturity on research that will help me to follow my career and have more success to become an independent researcher.