INSTITUT CARNOT Pasteur MI

Department of

Parasites and Insect Vectors

Institut Pasteur

23 & 24 November 2015

Parasitology in the 21st century

Institut Pasteur, Paris

Keynote speakers:

Carolina V. **Barillas-Mury**

Piet **Borst**

Speakers:

Cyrille **Botté**

Markus **Engstler**

Richard **Grencis**

Michael **Grigg**

Mohamed-Ali **Hakimi**

Cynthia **He**

Matthew **Higgins**

David **Horn**

Patricia **Johnson**

Shaden **Kamhawi**

Hugo **Lujan**

Julius **Lukeš**

Geoff **McFadden**

Annette **MacLeod**

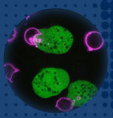
Bill **Petri**

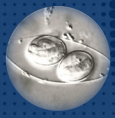
Frédéric **Simard**

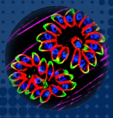
Isabelle **Tardieux**

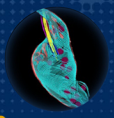
Till **Voss**

Charles **Wondji**









Contents

Introduction

By all rights, parasitology as a field probably should not exist. The organisms we study do not form a coherent phylogenetic group, and are not bound by any common evolutionary history. *Plasmodium* and *Trypanosoma* are as distant from each other as both are from animals. And then there are the insect vectors, generally included as part of the field of parasitology. Parasites may share a common ecological strategy by inhabiting niches within another organism to acquire resources for reproduction, but our parasites are far from the only pathogens with this lifestyle. We may think that they display cunningly, breathtakingly complex and sometimes apparently undefeatable mechanisms for host interaction and resource acquisition, and there we may have a point. Nevertheless, like the apocryphal bumblebee, even though parasitology maybe should not be able to fly, the important observation is that, it does. The essential glue that holds the field together turns out to be the research excellence contributed from parasite and vector systems, often groundbreaking, within the larger fields of cell biology, biochemistry, genomics, epidemiology, ecology and evolution. The scope of parasitology extends from the most modelized, in vitro systems where molecular details can be dissected with precision, to the least modelized natural studies of disease and parasite-vector transmission systems in hospitals and villages. The talks presented in this symposium span the breadth of the field, and we hope will make clear why parasitology has a future in and beyond the 21st Century.

Ken Vernick
Director of the Department

Information

Badges
Name badges must be worn at all times (in the amphitheatre and on the Campus).

Meals and refreshments
Lunch and refreshments will be served in the breakout area. See the scientific programme for specific times.

Mobiles phones
Please ensure that mobile phones are switched off or in silent mode during scientific sessions.

Important

Security measures have been reinforced on the campus and everywhere in Paris. This is likely to cause some delay and it is therefore advised to arrive early on the campus especially on Monday.

Make sure you have your ID with you.

Programme

Monday 23 November

8:30 **Introduction** by Ken Vernick, Director of the Department PIV

Practical information by Philippe Bastin, symposium organiser

Session 1 Fighting resistance in parasites and in insects

Chair: Odile Puijalon

8:45 **Geoff McFadden**
Resistance to the common malaria drug atovaquone cannot be transmitted

9:25 **Frédéric Simard**
Novel Opportunities for Vector Control in a Changing World

10:05 **Charles Wondji**
Signatures of selective sweeps in metabolic resistance to insecticides

10:45 *Coffee break*

Session 2 Parasites in the intestine: a spectrum of behaviours

Chair: Philippe Bastin

11:10 **Bill Petri**
Environmental and genetic factors regulating invasion and killing by *Entamoeba histolytica*

11:50 **Richard Grencis**
Whipworm infection: Modulation of the intestinal microbiome, within and without

12:30 **Julius Lukeš**
Are human intestinal eukaryotes true parasites or rather commensals?

13:15 *Lunch*

Session 3 Doors for escape: Antigenic variation

Chair: Chetan Chitnis

14:40 **Till Voss**
The epigenetics of antigenic variation and sexual commitment in the malaria parasite *Plasmodium falciparum*

15:20 **David Horn**
Antigenic variation in trypanosomes: VSG control by recombination and allelic exclusion

15:50 **Hugo Lujan**
Development of an oral vaccine platform based on protective and adjuvant properties of surface proteins of the intestinal parasite *Giardia lamblia*

16:30 *Coffee break*

KEYNOTE LECTURE

Chair: Artur Scherf

17:00 **Piet Borst**
Base J, its biosynthesis and function

Tuesday 24 November

8:30 **Introduction** by Christian Bréchot, General Director of the Institut Pasteur

SESSION 4 Parasites tuning their hosts

Chair: Rogerio Amino

8:40 **Ali Hakimi**
Beyond the vacuole border: *Toxoplasma* effectors co-opt the host (epi)genetic program

9:20 **Shaden Kamhawi**
Unraveling the virulence of *Leishmania* transmission by vector sand flies

10:00 **Patricia Johnson**
Molecular mimicry: a *Trichomonas vaginalis* homologue of a host cytokine activates human pro-inflammatory and anti-apoptotic pathways

10:40 *Coffee break*

SESSION 5 Unique molecular and cellular features of parasites

Chair: Gerald Spaeth

11:00 **Matthew Higgins**
Structural insights into the interactions at the heart of severe malaria

11:40 **Cyrille Botté**
Membrane biogenesis and role of the apicoplast in Apicomplexa parasites

12:20 **Cynthia He**
Cryo-Electron Tomography of genetically engineered mini *Trypanosoma brucei*

13:00 *Lunch*

SESSION 6 Parasite genetics and evolution

Chair: Luis Quintana-Murci

14:00 **Annette McLeod**
An asexual revolution: population genomics reveals the origin of human infective trypanosomes

14:40 **Michael Grigg**
Genetic Exchange, Surface Antigens and Inflammasome Sensors Activated by Protozoan Parasites

SESSION 7 The how and whys of parasite motility

Chair: Philippe Bastin

15:20 **Isabelle Tardieux**
Toxoplasma high-speed motor(s) and the force(s) operating at the host cell door: a key dynamic step on the way to its indoor niche

15:50 **Markus Engstler**
Always on the move – towards a mechanobiology of trypanosome motility

16:30 *Coffee break*

KEYNOTE CLOSING LECTURE & CLOSING REMARKS

Chair: Ken Vernick

17:00 **Carolina Barillas-Mury**
Plasmodium Evasion of Mosquito Immunity and Malaria Globalization: The Lock and Key Theory

18:00 **Closing remarks** by Ken Vernick, Director of the Department of Parasites and Insect Vectors

Organisers & Invited Speakers

Organisers

Philippe Bastin *Institut Pasteur, France*
Agnès Mer-Appéré *Institut Pasteur, France*
Ken Vernick *Institut Pasteur, France*

Invited speakers

Carolina V. Barillas-Mury *National Institutes of Health, USA*
Piet Borst *Netherlands Cancer Institute, The Netherlands*
Cyrille Botté *Institut Albert Bonniot, France*
Markus Engstler *University of Wuerzburg, Germany*
Richard Grencis *University of Manchester, UK*
Michael Grigg *National Institutes of Health, USA*
Mohamed-Ali Hakimi *Institut Albert Bonniot, France*
Cynthia He *National University of Singapore, Republic of Singapore*
Matt Higgins *University of Oxford, UK*
David Horn *University of Dundee, UK*
Patricia Johnson *University of California, USA*
Shaden Kamhawi *National Institutes of Health, USA*
Hugo Lujan *Center for Research and Development in Immunology and Infectious Diseases, Argentina*
Julius Lukes *Institute of Parasitology, Czech Republic*
Annette MacLeod *University of Glasgow, UK*
Geoff McFadden *University of Melbourne, Australia*
Bill Petri *University of Virginia, USA*
Frédéric Simard *Maladies Infectieuses et Vecteurs : Ecologie, Génétique, Evolution et Contrôle (MIVEGEC), France*
Isabelle Tardieux *Institut Cochin, France*
Till Voss *Swiss Tropical and Public Health Institute, Switzerland*
Charles Wondji *Liverpool School of Tropical Medicine, UK*

Invited Speakers Abstracts

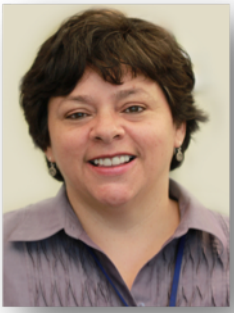
Keynote speakers

Piet Borst
Netherlands Cancer Institute, Amsterdam, Netherlands

Base J, its biosynthesis and function

Although research on trypanosomatids was usually a side-line in my lab, it was always fun and often led to unexpected discoveries: the kDNA maxi-circle (also found by Maurice Steinert); the glycosome; the ability to separate chromosome-sized DNA molecules of protozoa by PFG electrophoresis; the transposition mechanism for antigenic variation (with George Gross); the sequence of telomeric repeats (also found by Liz Blackburn); the growth and shrinkage of chromosome ends; the variable transferrin receptor and its potential role in trypanosome host expansion; and DNA base J.In my talk I shall mainly focus on base J, its biosynthesis and function.

Site web du laboratoire :
http://www.nki.nl/divisions/molecular-oncology/borst-p-group/



Carolina Barillas-Mury
National Institutes of Health, NIAID, Rockville, USA

Plasmodium Evasion of Mosquito Immunity and Malaria Globalization: The Lock and Key Theory

Plasmodium falciparum malaria is transmitted by anopheline mosquitoes. Mosquitoes can mount antiplasmodial responses that effectively limit infection of murine malaria parasites during the early stages of infections, when ookinetes invade the mosquito midgut. However, some Plasmodium falciparum strains can escape these defenses and survive. We recently identified Pfs47, as the surface protein that allows the parasite to evade mosquito immunity by disrupting JNK signaling in midgut cells. P. falciparum malaria originated in Africa and became global as humans migrated to other continents. During this journey, parasites encountered new anopheline mosquito species that were sometimes evolutionarily distant from African vectors. We propose that that the mosquito immune system is a major evolutionary force that continuously selects the parasites circulating in a given region, because only those parasites expressing a Pfs47 haplotype compatible with a given vector species are able to evade antiplasmodial immunity and survive. A new model, the lock and key theory of P. falciparum globalization, is proposed and its implications will be discussed.

Site web du laboratoire :
http://www.niaid.nih.gov/labsandresources/labs/aboutlabs/lmvr/mosquitoimmunityvectorcompetenceunit/Pages/barillasMury.aspx

Speakers

SESSION 1 Fighting resistance in parasites and in insects

Geoff McFadden
University of Melbourne, Melbourne, Australia

Resistance to the common malaria drug atovaquone cannot be transmitted

Drug resistance undermines our ability to control malaria. Mutant parasites resistant to chloroquine, pyrimethamine, and artemisinin emerged, then spread geographically, costing millions of lives and necessitating a constant search for replacement drugs. Resistance control strategies include drug rotation, drug combinations, and pursuing targets refractory to resistance. A less explored strategy is to identify drug targets where resistance mutations are unable to spread from patient to patient, which must occur via mosquitoes. We reasoned that some resistance mutations conferring selective advantage under drug pressure in human stages might suffer a fitness deficit in the mosquito stages thereby reducing transmission, and we focused on mitochondrial electron transport drug targets. Malaria parasites require only modest mitochondrial electron transport in the mammalian blood phase, but electron transport is massively up regulated in the sexual mosquito stages, creating starkly different selection regimes on transport components across the life cycle. We tested the ability of malaria parasites with mutations in the mitochondrial DNA encoded cytochrome B gene (cytB), which are resistant to the widely used electron transport inhibitor atovaquone, to transmit resistance to new hosts via mosquitoes. Five different rodent malaria (Plasmodium berghei) cytB mutants tested were unable to successfully infect mosquitoes and failed to transmit resistance mutations to naïve mice. Outcrossing with atovaquone sensitive lines also failed to facilitate resistance transmission, apparently because the mitochondrion encoded cytB gene is matrilineally inherited. Two tested atovaquone resistant cytB mutants of the human cerebral malaria parasite (P. falciparum) were also unable to successfully infect mosquitoes. Our results suggest that the commonly occurring atovaquone resistance mutations are unable to increase in frequency in the population and cannot spread geographically, which radically improves the long-term prospects for this antimalarial.

Site web du laboratoire : www.geoffmcfadden.com

Frédéric Simard[1], Christophe Paupy[1], Jérémy Boyer[2]
[1] *Maladies Infectieuses et Vecteurs : Ecologie, Génétique, Evolution et Contrôle (MIVEGEC), Montpellier, France*
[2] *UMR CMAEE CIRAD-INRA "Control of exotic and emerging animal diseases", Montpellier, France*

Novel Opportunities for Vector Control in a Changing World

Vector control remains a cornerstone of the fight against malaria and other vector-borne diseases throughout the world. Indoor spraying with residual insecticides and the use of long-lasting insecticide impregnated mosquito nets helped curb mortality and morbidity due to malaria observed since the early 2000s throughout Africa. But the effectiveness of these control methods is now jeopardized by the emergence and rapid spread of insecticide resistance in all major vectors. As they have always done, mosquitoes evolve, they adapt, change their behavior and adjust their physiology to this new environment. The very limited arsenal of molecules used in public health offers few alternatives for sustainable management of these resistances. It is now necessary and urgent to devise new strategies for reasoned and biologically-lucid vector control strategies that will take into account all aspects of vectors ecology to optimize the impact on pathogen's transmission. To do this, it is imperative to consider vectors as part of the ecosystem within which they develop and proliferate, and to take into account all the interactions, abiotic as well as biotic, in which they are engaged and that modulate their life history traits and impact their vectorial capacity. Comparative genetic and genomic analyzes now possible thanks to the many complete genomes of Anopheles published recently, coupled with phenotypic analyzes will identify new targets for more specific and innovative control strategies. Explored in the light of evolutionary biology, these new strategies will help mitigate resistance and sustain disease control through the development of integrated vector management geared towards transmission control.

Site web du laboratoire : http://www.mivegec.ird.fr

Charles Wondji
Liverpool School of Tropical Medicine, Liverpool, UK

Signatures of selective sweeps in metabolic resistance to insecticides

Insecticide resistance in malaria vectors such as *Anopheles funestus* is hindering malaria prevention throughout Africa. Using a combination of microsatellite genotyping and sequencing, we established the population structure of *An. funestus* on an African-wide scale and performed an in-depth analysis of the genetic structure in southern Africa, an area with high resistance to pyrethroids, the only insecticide class recommended for bed nets. This study revealed a strong selective sweep around important cytochrome P450s known to confer resistance to pyrethroids. Moreover, temporal genetic diversity analysis revealed that this selective sweep occurred only after the scaling up of pyrethroid-based vector control interventions notably long lasting insecticide treated bed nets. Furthermore, functional and transgenic analyses revealed that only highly efficient pyrethroid metabolising P450 alleles have been selected replacing poor metabolising alleles. By deciphering the patterns of evolution and spread of insecticide resistance, this study provides vital information to implement suitable resistance management strategies.

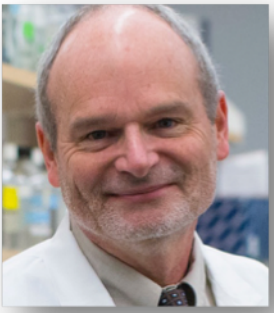
Elucidating the direction and speed of spread of insecticide resistance markers in mosquito populations is crucial in implementing suitable resistance management strategies. Such evolution is strongly associated with patterns of gene flow and the strength of selection. An Africa-wide analysis of patterns of genetic structure in the major malaria vector Anophles funestus revealed that current distribution of insecticide resistance across the continent is strongly influenced by patterns of gene flow. The evolution of insecticide resistance resulted to a major footprint of selective sweep around resistance genes with resistance allele nearing fixation in areas of high resistance. Comparison of polymorphism between pre and post-intervention revealed that implementation of bed nets is the single most important factor explaining the selective sweep. Furthermore, functional and transgenic analyses revealed that only cytochrome P450 alleles able to metabolise insecticides have been selected whereas poor metabolising alleles have been lost.

What is the selective
Is pattern of gene flow in Malaria mosquitoes impacting the distribution of insecticide resistance across Africa? Are there genomic footprints of insecticide-based interventions tools such as bed nets in Malaria vectors? We present an Africa-wide study of patterns of genetic structure in a major malaria vector in Africa, Anopheles funestus, showing that profile of gene flow correlates well with current distribution of insecticide resistance across the continent. Microsatellite and analysis of sequence polymorphism revealed a strong signal of selective sweep around a major insecticide resistance locus driven by two cytochrome P450 genes known metaboliser of pyrethroids. Comparison of polymorphism

between pre and post-bed nets intervention revealed that implementation of bed nets is the single most important factor explaining the selective sweep. Furthermore, functional analyses revealed that the only alleles able to metabolise insecticides have been selected whereas

Site web du laboratoire :
http://www.lstmed.ac.uk/about/people/dr-charles-wondji

SESSION 2 Parasites in the intestine: a spectrum of behaviours



William A. Petri
University of Virginia, Charlottesville, USA

Environmental and genetic factors regulating invasion and killing by *Entamoeba histolytica*

Entamoeba histolytica trophozoites destroy the intestinal epithelium to cause amebic colitis and liver abscess. The steps in pathogenesis include depletion of the host mucosal barrier, adherence to the colonic epithelium, cytotoxicity by amebic trogocytosis and invasion of the colonic epithelium. However most infections do not result in invasion or illness. The genetic and environmental factors that regulate the interaction of parasite and host in the colonic microenvironment will be highlighted.

Site web du laboratoire :
http://www.medicine.virginia.edu/clinical/departments/pathology/Faculty/petri-page

Richard Grencis
University of Manchester, Manchester, UK

Whipworm infection: Modulation of the intestinal microbiome, within and without

Whipworm (Trichuris sp.) is a common nematode infection of man and animals. The parasite lives partially or completely embedded within the epithelial cells of the large intestine. In nature, it survives as a long lived chronic infection with most individuals harbouring low numbers of parasites, and acquired immunity is only partial. Studies in the laboratory model, T. muris, has helped define current paradigms of resitance and susceptibility to gastrointesinal nematode infection. We have previously shown that the host intestinal microflora is important is successful establishment of T. muris infection. Long lived T. muris infection, however, is associated with a marked dysbiosis in the intestinal microbiome, driven by the presence of the parasite. Futhermore, we now have data supporting the acquisition of an intesinal microbiome by the parasite from the host following infection. The intestinal microbiome of the parasite appears to be important for its own fitness with the parasite driven dysbiosis of the host intestinal microbiome reducing subsequent parasite establishment. The data indicates that the three-way interaction between parasite host and microbiome is critical for successful parasitism. The mechanisms uderlying these process will be discussed.

Trichuris muris (SEM of adult worm)

Site web du laboratoire : http://www.manchester.ac.uk/research/richard.grencis/

Julius Lukeš[1,2], Christen Rune Stensvold[3], Kateřina Jirků-Pomajbíková[1], Laura Wegener Parfrey[2,4]

[1] *Institute of Parasitology, Biology Centre, and Faculty of Science, University of South Bohemia, České Budějovice, Czech Republic*

[2] *Canadian Institute for Advanced Research, Toronto, Canada*

[3] *Statens Serum Institut, Copenhagen, Denmark*

[4] *Departments of Botany and Zoology, University of British Columbia, Vancouver, Canada*

Are human intestinal eukaryotes true parasites or rather commensals?

The short abstract could be something like: Human body hosts a multitude of uni- and multicellular eukaryotes, mostly protists and helminths, which are historically thought of as parasites. I will discuss the emerging view that most eukaryotes confined to the human intestinal tract constitute an important segment of intestinal ecosystem, and that their elimination may have a negative effect for our health.

Site web du laboratoire :
http://www.paru.cas.cz/en/section/molecular-parasitology/laboratory-of-molecular-biology-of-protists/

SESSION 3 Doors for escape: Antigenic variation

Till Voss
Swiss Tropical and Public Health Institute, Basel, Switzerland

The epigenetics of antigenic variation and sexual commitment in the malaria parasite *Plasmodium falciparum*

Epigenetic control of heritable gene silencing generates phenotypic diversity in clonally expanding cell populations. *Plasmodium falciparum* blood stage parasites, which cause the most severe form of malaria in humans, employ this strategy to regulate antigenic variation and sexual conversion, two processes essential for parasite survival and transmission. Antigenic variation allows parasites to evade adaptive immune responses in order to establish chronic blood infection. Sexual conversion is the cell fate decision process that occurs in a small subset of proliferating parasites and leads to cell cycle exit and differentiation into gametocytes, which are essential for successful transmission to the mosquito. Intriguingly, both processes are dependent on heterochromatin protein 1 (HP1), a conserved epigenetic regulator of heritable silencing. Our research aims to elucidate in detail how HP1 influences and controls phenotypic variation in *P. falciparum*, and how HP1 itself is regulated in order to facilitate opposite transcriptional states of critical target genes.

Site web du laboratoire :
http://www.swisstph.ch/about-us/staff/detailview.html?tx_x4epersdb_pi1[showUid]=2151&cHash=8c52b6f26e1c2463016b49c9c007dd39

David Horn, Lucy Glover, Sebastian Hutchinson
University of Dundee, Dundee, UK

Antigenic variation in trypanosomes: VSG control by recombination and allelic exclusion

Studies on Variant Surface Glycoproteins (VSGs) and antigenic variation in the African trypanosome, Trypanosoma brucei, have yielded a remarkable range of novel and important insights. However, some long-standing questions persist. We have addressed two of these in particular. First, how is VSG recombination controlled? Second, how do cells restrict expression to a single allele of the VSG gene-family? In relation to the first question, we have characterized DNA repair pathways in some detail and are now developing new tools and assays to facilitate further study. In relation to the second, we have identified VEX1 (VSG exclusion 1), a competence factor that positively regulates VSG expression in cis and negatively regulates VSG expression in trans. I will describe our latest results and current thinking on these projects.

Site web du laboratoire : http://www.lifesci.dundee.ac.uk/groups/david-horn/home

Marianela C. Serradell, Lucia L. Rupil, Roman A. Martino, Alicia Saura, Hugo D. Lujan
Center for Research and Development in Immunology and Infectious Diseases (CONICET-UCC), Cordoba, Argentina

Development of an oral vaccine platform based on protective and adjuvant properties of surface proteins of the intestinal parasite *Giardia lamblia*

Despite the impact of world-wide vaccination, there is still a great need to develop cheap and safe innovative vaccination strategies inducing long-lasting immunity. Since most infectious agents invade the organism via mucosal surfaces, adaptive mucosal immunity plays a central role in protecting the host against infections. Oral vaccines represent a very attractive option because they are not invasive and suitable for mass vaccination programs. However, the main impediment for oral vaccine development has been that orally administered antigens are easily destroyed in the gastrointestinal tract or potentially capable of inducing immune tolerance. The intestinal parasite *Giardia lamblia* expresses variant-specific surface proteins (VSPs) that are extremely resistant to the low pH of the stomach and to intestinal proteases, allowing the parasite to survive in the harsh environmental conditions of the small intestine. Besides, these cysteine-rich molecules are capable to trigger a strong immune response by activating dendritic cells and favoring antigen uptake. We thus hypothesized that the expression onto virus-like particles (VLPs) of *Giardia* VSPs should shield these particles for oral administration. To obtain a proof of principle and, simultaneously, to develop a potential vaccine candidate, we used Influenza Hemagglutinin (HA) as a vaccinal antigen. Our results clearly show that *Giardia* VSP can protect vaccinal antigens inside the gastrointestinal track for oral administration of vaccines, generating strong T and B cell-mediated protective responses. The development of this universal vaccine platform should have a broad application to different parasitic diseases.

SESSION 4 Parasites tuning their hosts

Mohamed-Ali Hakimi
Institut Albert Bonniot, La Tronche, France

Beyond the vacuole border: *Toxoplasma* effectors co-opt the host (epi)genetic program

The obligate intracellular parasite *Toxoplasma gondii* strikes a subtle balance with the host immune system that not only prevents host death but also promotes parasite persistence. Although being enclosed within a parasitophorous vacuole, the parasite actively interfaces with host cell signaling pathways, thereby directing host cell responses. To this end, *T. gondii* delivers effector proteins into the host cell that co-opt host transcription factors and eventually modulate gene expression. Aside from the secretory Rhoptry organelles initially described as the main source of such effectors, Dense Granules are now recognized as critical in delivering products that remain confined at the vacuolar space or traffic beyond the vacuole membrane to the host cell nucleus and contribute to rewire host gene expression.

Site web du laboratoire : http://www.researchgate.net/profile/Mohamed-Ali_Hakimi

Ranadhir Dey[1], Lais da Silva[2], Amritanshu Joshi[1], Shannon Townsend[2], Claudio Meneses[2], Jesus Valenzuela[2], Hira Nakhasi[1], Shaden Kamhawi[2]
[1] *Division of Emerging and Transfusion Transmitted Diseases, Center for Biologics Evaluation and Research, FDA, Bethesda, USA*
[2] *Vector Molecular Biology Section, Laboratory of Malaria and Vector Research, NIAID, NIH, USA*

Unraveling the virulence of *Leishmania* transmission by vector sand flies

Transmission of *Leishmania* by vector bites is complex enhancing virulence of transmitted parasites. This is reflected by an exacerbated disease outcome and the abrogation of protection observed in vaccinated mice challenged by a needle inoculum. This heightened virulence has been attributed to the co-deposition into the bite site of various vector-derived factors with immunomodulatory properties that promote parasite survival and disease establishment. I will present data on novel virulence factors that contribute to the infectious inoculum and demonstrate that vector-transmission of *Leishmania* rapidly drives a unique sustained inflammatory immune response that begins to elucidate its virulence.

Site web du laboratoire :
http://www.niaid.nih.gov/labsandresources/labs/aboutlabs/lmvr/vectormolecularbiologyunit/Pages/valenzuela.aspx#niaid_inlineNav_Anchor

Patricia J. Johnson[1,2], Olivia Twu[1], Yi-Pei Chen[1], Daniele Dessí[3], Anh Vu[2], Frances Mercer[2], Grant C. Stevens[2], Robert T. Clubb[4] & Pier Luigi Fiori[3]

[1] *Molecular Biology Institute*

[2] *Department of Microbiology, Immunology and Molecular Genetics, University of California, Los Angeles, California, USA*

[3] *Dipartimento di Scienze Biomediche, Università degli Studi di Sassari, 07100 Sassari, Italy*

[4] *Department of Chemistry and Biochemistry, University of California, Los Angeles, CA USA*

Molecular mimicry: a *Trichomonas vaginalis* homologue of a host cytokine activates human pro-inflammatory and anti-apoptotic pathways

Trichomonas vaginalis is a common sexually transmitted parasite that colonizes the human urogenital tract where it adheres to epithelial cells and remains extracellular. Infections are often asymptomatic but can be highly inflammatory, depending on the host and the parasite strain. Symptomatic women typically present with vaginitis, whereas infection in men is usually asymptomatic and can lead to untreated, chronic inflammation of the prostate. We have found that *T. vaginalis* secretes a protein (TvMIF) that is 47% similar to human macrophage migration inhibitory factor (HuMIF), a pro-inflammatory cytokine. Anti-TvMIF antibodies were also found in 79% of men seropositive for *T. vaginalis*. As *T. vaginalis* infection is associated with increased risk of aggressive prostate cancer, we asked whether TvMIF might be a molecular mimic of HuMIF thereby inducing cellular pathways linked to inflammation and increasing prostate cell proliferation and invasiveness. We have shown TvMIF to be pro-inflammatory and to bind the human CD74 MIF receptor with high affinity, comparable to that of HuMIF. This in turn triggers activation of ERK, Akt and BAD phosphorylation at a physiologically relevant concentration (80 pM). These data, together with studies showing that TvMIF increases *in vitro* growth and invasiveness of benign and prostate cancer cells, support a possible role for TvMIF in the promotion and progression of prostate cancer in men infected with *T. vaginalis*. A likely intrinsic function for TvMIF in parasite survival will also be discussed.

Site web du laboratoire :
http://people.healthsciences.ucla.edu/institution/personnel?personnel_id=45313

SESSION 5 Unique molecular and cellular features of parasites

Matthew Higgins
University of Oxford, Oxford, UK

Structural insights into the interactions at the heart of severe malaria

The parasites that cause malaria make many and varied interactions with the human host. They invade host cells, including human erythrocytes, and divide within the protected intracellular environment. In the case of Plasmodium falciparum they also send adhesive proteins to infected erythrocyte surfaces, causing them to adhere throughout the body, protecting the parasite within from spleen mediated clearance. I will present our structural studies of the host-parasite interactions involved in both erythrocyte invasion and host tissue adhesion, drawing out their similarities and differences.

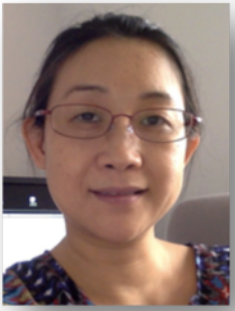
Site web du laboratoire : http://www.bioch.ox.ac.uk/aspsite/index.asp?pageid=808

Cyrille Botté
Institut Albert Bonniot, La Tronche, France

Membrane biogenesis and role of the apicoplast in Apicomplexa parasites

The summary: Apicomplexa parasites are causative agents of major human diseases such as malaria caused by Plasmodium spp., and toxoplasmosis caused by Toxoplasma gondii. These diseases affect and kill millions of people every year. There is no current vaccine and our therapeutical arsenal is reducing each year due to increasing parasite resistance. Throughout their intracellular development, Apicomplexa require large amounts of lipids to generate their daughter cells. The bulk of these membrane lipids are believed to originate from two essential sources: (i) host cell scavenging and (ii) parasite de novo synthesis pathways. Most apicomplexan parasites harbour a non-photosynthetic relict plastid, named the apicoplast, which is essential for parasite survival. We showed that the apicoplast possesses the metabolic machineries to generate, assemble and distribute major lipid precursors to the parasite. Major questions remain to be answered on (i) the nature of lipid products synthesized by the apicoplast, the parasite and those scavenged from the host, (ii) the intracellular destination of such products, (iii) the lipid composition of the parasite compartments, and (iv) the way these lipids are trafficked between these compartments. Our group aims to address these questions and contributing to understand these important mechanisms of membrane biogenesis, to potentially identify new therapeutic targets against Apicomplexa.

Site web du laboratoire : http://www.researchgate.net/profile/Cyrille_Botte



Cynthia Y. He
National University of Singapore, Department of Biological Science, Republic of Singapore

Cryo-Electron Tomography of genetically engineered mini *T. brucei*

Trypanosoma brucei contains a subpellicular array that is composed of >100 stable microtubules crosslinked to each other, forming a bird cage-like structure underneath the plasma membrane. Development of the subpellicular array is tightly linked to biogenesis of the flagellum and the flagellum attachment zone and crucial for cell morphology, during the cell cycle as well as the life cycle development. In this study, we used cryo electron tomography to visualize the 3D organization of the subpellicular microtubule array in genetically engineered mini *T. brucei* cells. 3-dimensional spatial relationship between the flagellum, FAZ and the subpellicular microtubules was analyzed. The results provide an ultrastructural model on how the flagellum drives bihelical cell movement by modifying the arrangement of the subpellicular array.

Site web du laboratoire : http://www.dbs.nus.edu.sg/staff/cynthia.htm

Willie Weir[1], Bernardo Foth[2], Paul Calewell[1], Caroline Clucas[1], Andrew Pountain[1], Pieter Steketee[1], Nicola Veitch[1], Maturin Koffi[3], Thierry de Meeus[4], Jacques Kabore[4], Mohamed Camara[5], Anneli Cooper[1], Andy Tait[1], Vincent Jamoneau[4,6], Bruno Bucheton[5,6], Mathew Berriman[2], Annette MacLeod[1]

[1] *University of Glasgow, Glasgow, UK*
[2] *Wellcome Trust Sanger Institute, Hinxton, UK*
[3] *Université JL. Guédé, Douala, Côte d'Ivoire*
[4] *CIRDES, Burkina Faso*
[5] *Programme National de Lutte contre la Trypnaoso-miase Humaine Africaine, Conakry, Guinea*
[6] *IRD, Montpellier, France*

An asexual revolution: population genomics reveals the origin of human infective trypanosomes

Evolutionary theory predicts that the lack of recombination in strictly asexual diploid organisms results in a reduced ability to purge their genomes of mutations. The predicted consequence of this is that homologous chromosomes will accumulate irreversible mutations and will evolve independently, termed the Messelson effect. In order to examine this phenomenon at a genome wide level, we have examined the genome of an important human pathogen, the main causative agent of human African trypanosomiasis, *Trypanosoma brucei gambiense*, which appears to reproduce clonally. Here we use a population-scale approach to investigate the reproductive strategy of this highly successful parasite to gain an insight into the genomic consequences of clonal evolution. We demonstrate that *T. b. gambiense* is evolving in a strictly asexual manner and is derived from a single human-infective progenitor, which emerged within the last 10,000 years and radiated across West Africa. We demonstrate that homologous chromosomes evolve independently leading to the parasite population consisting of just two distinct haplotypes, confirming at the genome wide level the theoretically predicted Meselson effect. We have identified large regions of loss of heterozygosity within the genome consistent with mitotic recombination and gene conversion events, which we hypothesise to be a short-term compensatory mechanism for these deleterious mutations. This study demonstrates that obligate asexuality is a successful eukaryotic reproductive strategy, which in this case has empowered an ancient pathogen to exploit a new biological niche.

Site web du laboratoire :
http://www.gla.ac.uk/researchinstitutes/bahcm/staff/annettemacleod/

Khan A.[1], Kennard A.[1], Gregg B.[1], Pszenny V.[1], Ramirez JD.[1], Barhoumi M.[1,2], Zhang J.[3], Guizani I.[2], Lukes J.[4], Sacks D.[5], Porcella S.[6], Shen K.[7], Parkinson JP.[3], Tonkin M.[8], Boulanger M.[8], Grigg ME.[1]

[1] *Molecular Parasitology Section, LPD, NIAID, NIH, USA*
[2] *Pasteur Institute at Tunis, Tunisia*
[3] *University of Toronto, Canada*
[4] *University of South Bohemia, Czech Republic*
[5] *Intracellular Parasite Section, LPD, NIAID, NIH, USA*
[6] *RML Genomics Unit, RTS, NIAID, NIH, USA*
[7] *OCICB, NIAID, NIH, USA.* [7] *University of Victoria, Canada*

Genetic Exchange, Surface Antigens and Inflammasome Sensors Activated by Protozoan Parasites

Using population genetic and WGS phylogenomic methods, our work has identified extant genetic exchange among circulating populations of natural isolates of *Giardia, Leishmania,* and *Toxoplasma*. To understand the biological consequences of such genetic admixture, we utilize *Toxoplasma* as our genetic model. Acute virulence during murine *Toxoplasma* infection is highly dependent on expression of polymorphic secreted pathogenesis determinants (SPDs) that are inherited in discrete haploblocks by genetic exchange. SPDs discharged from parasite secretory organelles target host immune signaling pathways and facilitate infection competency. Employing forward, reverse genetic, and genome-wide association (GWAS) techniques, we have identified novel SPDs activating inflammasome pathways, dysregulating immune homeostasis, or altering parasite pathogenesis. eQTL screening of progeny from a collection of *T. gondii* crosses that differentially modulate activation of the host inflammasome has identified three new parasite loci, in addition to GRA15, that upregulate IL-1β. Utilizing GWAS on WGS data from 56 *T. gondii* strains, we have identified four genomic regions (Chromosome VIIa, VIIb, VIII and IX) encoding novel SPDs associated with murine virulence. Finally, utilizing reverse genetics, we show that SRS29C expression negatively regulates murine virulence. The 1.8Å structure of SRS29C possesses a positively charged basic groove mediated principally by three ARG residues (K62, K68, K69) that specifically interact with heparan sulfate, a negatively-charged sulfated glycosaminoglycan (GAG). Mutation of active site residues K62 or K68/69 abrogates GAG binding and restores acute virulence. Our data suggest that the SRS29C-dependent GAG interaction is an important regulator of host immune responses required to maximize parasite transmissibility.

Site web du laboratoire :
http://www3.niaid.nih.gov/labs/aboutlabs/lpd/molecularparasitologyunit/

SESSION 7 The how and whys of parasite motility

Isabelle Tardieux
Institut Cochin, Department of Infection-Immunology-Inflammation, Paris, France

Toxoplasma high speed motor(s) and the force(s) operating at the host cell door: a key dynamic step on the way to its indoor niche

A major interest of my lab is to delve deeper into the molecular machinery/ies that drive(s) *Toxoplasma* tachyzoites into the host cells on which rely their propagation with a primary focus on **force** production during the process. To decipher the origin and features of the force(s) powering *Toxoplasma* entry, we combined kinematic and modeling analysis of cell invasion using parasites that express a fluorescent and functional version of RON2, the core component of the critical tachyzoite junction rapidly assembled at the host cell membrane. This junction serves as the entry door of its indoor niche, a unique dynamic vacuole where to futher develop.

We showed that the majority of invasion events occur with a typical forward rotational progression of the parasite through a **static** junction. However, when tachyzoites encounter resistance in their way to the indoor niche or when the junction is not properly anchored to the host cell cortex, the parasite similarly pulls on the junction that is therefore seen capped backwards together with the host cell membrane to eventually enclose *Toxoplasma* in a functional vacuole.

Next, we analyzed the outcomes of invasion attempts when genetically engineered-tachyzoites were either defective in their ability to (a) decrease host cell cortex tension or (b) to pull on the junction. While both defects differentially alter invasiveness leading to number of abortive events, these as well as the successful sequences are drastically distinct in the molecular machinery/ies engaged by the host cells.

These studies have brought new evidence that (i) tachyzoites need to engage myosin motor with the junction molecular complex to promote force production (ii) this sequence depending on the anchorage of the junction to the host cell cortex and (iii) insertion of the junction triggers substantial host cell membrane dynamics that is spatially and timely restricted during the high speed invasion sequence by motor-competent tachyzoites. Current work aims at uncovering the early signaling pathways and changes at the host cell door during the whole sequence of cell penetration.

Site web du laboratoire :
http://cochin.inserm.fr/Departements/3i/equipe-tardieux-poyart

Markus Engstler
University of Wuerzburg, Wuerzburg, Germany

Always on the move – towards a mechanobiology of trypanosome motility

The life of African trypanosomes is a journey through very distinct environments, encompassing the mammalian bloodstream, various tissues, the cerebrospinal fluid, and the tsetse fly digestive system. This lecture discusses how these parasites may have adapted to the physical conditions prevailing at low Reynolds numbers, in a world without inertia. Two basic aspects will be considered, namely membrane protein diffusion and cell motility. Trypanosomes exploit both processes for survival in the host, and their strategies may shed new light on some fundamental aspects of physics. Towards the end of my lecture, I will highlight the challenges resulting from the marked species-specific differences in trypanosome motility.

Site web du laboratoire :
http://www.zeb.biozentrum.uni-wuerzburg.de/en/people/markus_engstler/