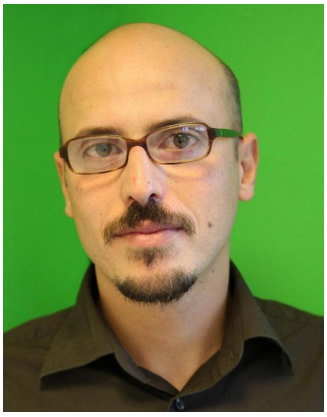


First Name / Last name: Charles Baroud
Contact : *charles.baroud@pasteur.fr*

Unit: Physical microfluidics and Bioengineering
IP Department or IP: G&G
Secondary affiliation: None

Main domain 1: Bacteria

Main domain 2: technological and methodological developments.

Attractive synopsis:
In my group we are using microfluidic tools and imaging to study the emergence of antibiotic resistance at the scale of individual cells.

Research projects in relation with AMR:

We have developed a range of microfluidic tools that allow us to observe the growth of bacterial colonies, starting from one cell or a few cells. The techniques also allow the modulation of the environment, e.g. to add or remove antibiotics, while observing the response of the cells. With these methods, we are currently addressing the emergence of resistance to antibiotics, in collaboration with other groups at Pasteur, for example by correlating the SOS response of individual cells with their ability to grow under antibiotic stress. Quantitative measurements are then confronted with mathematical models in order to understand the distribution of behaviours in the population.

3 Publications

- Amselem, G., Guermonprez, C., Drogue, B., Michelin, S., & Baroud, C. N. (2016). Universal microfluidic platform for bioassays in anchored droplets Lab on a Chip. *Lab on a Chip*, *16*, 4200–4211. https://doi.org/10.1039/C6LC00968A

- Amselem, G., Sart, S., & Baroud, C. N. (2018). Universal anchored-droplet device for cellular bioassays. *Methods in Cell Biology*, *148*, 177–199. https://doi.org/10.1016/bs.mcb.2018.05.004

- Barizien, A., Jammalamadaka, M. S. S., Amselem, G., & Baroud, C. N. (2019). Growing from a few cells : combined effects of initial stochasticity and cell-to-cell variability. *J. Roy. Soc. Interface. Soc. Interface*, *16*, 20180935. https://doi.org/10.1098/rsif.2018.0935

First Name / Last name: Frédéric Barras
Contact : fbarras @pasteur.fr.....

Unit: SAMe
IP Department or IP: Dpt of Microbiology
Secondary affiliation: Dpt of Genome and Genetic

Main domains 1: Bacteria,

Main domain 2: mechanism of resistance and dissemination

Attractive synopsis:

Understanding molecular, metabolic and physiological mechanisms of antibiotic activity and of phenotypic resistance.

Research projects in relation with AMR:

- Role of Fe-S cluster proteins in resistance/sensitivity to antibacterial compounds.
- Influence of metals on resistance/sensitivity to antibacterial compounds.
- Influence of iron bioavailability on resistance to quinolones.
- Mechanism of resistance to aminoglycoside under anaerobic respiratory conditions.
- Optimizing Fe-S protein production to enhance antibiotic production (ERA BioTech with G. Bokinsky, NL)
- Search for antibacterial compound targeting Fe-S associated process (project).

3 Publications

- Ezraty, B., Vergnes, A., Banzhaf, M., Duverger, Y., Huguenot, A., Brochado, A.R., Su, S.-Y., Espinosa, L., Loiseau, L., Py, B., Typas, A. and Barras, F. (2013) Fe-S cluster biosynthesis controls uptake of aminoglycosides in a ROS-less death pathway. ***Science*** 340:1583-1587.

- Hérisse M., Duverger Y., Martin-Verstraete I., Barras F., Ezraty B. (2017) Silver potentiates aminoglycoside toxicity by enhancing their uptake. ***Mol. Microbiol.*** doi: 10.1111/mmi.13687

- Chareyre S., Barras F. and P. Mandin (2019) A small RNA controls bacterial sensitivity to gentamicin during iron starvation. ***PLoS Genet***: in press

First Name / Last name: Gregory Batt
Contact: *gregory.batt@inria.fr*

Unit: InBio - Experimental and computational methods for modeling cellular processes
IP Department or IP: Computational Biology Department
Secondary affiliation: Genomes and Genetics Department

Main domain 1: Bacteria, Yeast

Main domain 2: Technological and methodological developments.

Attractive synopsis:

We are developing models to characterize individual and collective antibiotic resistance to β-lactams, together with algorithms and experimental platforms for their automated and efficient calibrations.

Research projects in relation with AMR:

In β-lactam treatments, the resistance of a bacterial population depends on the antibiotic dose but also on the cell density. Indeed, when dying, bacteria release an enzyme, a β-lactamase, in their environment that degrades the antibiotic and hence allows not-dead-yet cells to escape the treatment and repopulate their niche.

We propose an efficient approach for the phenotypic characterization of clinical isolates subjected to β-lactam treatments. Our models capture the subtle interplay between cell growth, antibiotic-mediated killing and antibiotic degradation by β-lactamases. Moreover, we develop an experimental platform combining software and hardware for their efficient and automated calibration.

3 Publications

- HR Meredith, V Andreani, HR Ma, AJ Lopatkin, AJ Lee, DJ Anderson, G Batt, and L. You (2018). Applying ecological resistance and resilience to dissect bacterial antibiotic responses, Science Advances, 4(12):eaau1873.

First Name / Last name: Christophe BELOIN
Contact : *christophe.beloin@pasteur.fr*

Unit: Génétique des Biofilms
IP Department or IP: Microbiology
Secondary affiliation:

Main domains 1: **Bacteria**

Main domain 2: mechanism of resistance and dissemination, alternative strategies.

Attractive synopsis:

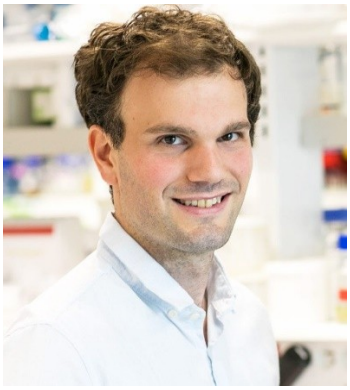
We aim at understanding the molecular mechanisms beyond the extreme tolerance of biofilms towards antibiotics and the link between this tolerance and evolution of resistance in order to identify novel strategies to fight biofilm-associated infections and to reduce emergence of antibiotic resistance.

Research projects in relation with AMR:

Biofilms are communities of microorganisms that interact with surfaces, where they display unique properties compared to free-floating, individual microorganisms. In clinical contexts, biofilms are responsible for many infections that are difficult to eradicate due to biofilm extreme tolerance towards antibiotics. This phenotypic tolerance is mainly associated to high level of persister bacteria that survive antibiotic treatments. We aim at characterizing molecular mechanisms of persistence within *E. coli* biofilms and developing approaches to eradicate persisters. We have shown that combination of stress response and starvation within biofilms plays a key role in formation of persister bacteria. Moreover, we identified several molecules potentiating antibiotic efficacy against persisters and enabling eradication of pathogenic biofilms in a clinically relevant *in vivo* model of catheter-associated infection. Recently we showed using experimental evolution that emergence of resistance is facilitated within biofilms.

3 Publications

- Lebeaux D, Ghigo JM, Beloin C*. Biofilm-related infections: bridging the gap between clinical management and fundamental aspects of recalcitrance toward antibiotics. Microbiology and molecular biology reviews : MMBR. 2014;78(3):510-43.
- Lebeaux D, Chauhan A, Letoffe S, Fischer F, de Reuse H, Beloin C*, et al. pH-mediated potentiation of aminoglycosides kills bacterial persisters and eradicates *in vivo* biofilms. The Journal of infectious diseases. 2014;210(9):1357-66.
- Usui M*, Yokoo H, Tamura Y, Nakajima C, Suzuki Y, Ghigo JM, Beloin C*. Zinc acetate potentiates the action of tosufloxacin *Escherichia coli* biofilm persisters. Antimicrobial agents and chemotherapy. 2019 Apr 1. pii: AAC.00069-19.



First Name / Last name: **David Bikard**
Contact : *david.bikard@pasteur.fr*

Unit: **Synthetic Biology**
IP Department or IP: **Microbiology**
Secondary affiliation: **Genomes and Genetics**

Main domain 1: Bacteria

Main domain 2: alternative strategies, technological and methodological developments.

Attractive synopsis:

The Synthetic Biology group is developing CRISPR tools to study and fight antibiotic resistant bacteria.

Research projects in relation with AMR):

The synthetic biology group has two main interests related to antimicrobials. (1) In collaboration with Eligo Bioscience we are investigating the feasibility of using CRISPR as a sequence specific antimicrobial. This strategy relies on the delivery of CRISPR systems to bacterial populations in vivo using bacteriophage derived vectors. The CRISPR system can be programmed to target antibiotic resistance genes to either killed or resensitize target bacteria. (2) Our group also focuses on the development of high-throughput screens based on the ability of the dead-Cas9 (dCas9) protein to block gene expression. Such screens can be useful for the identification of genetic interactions as well as interactions between genes and drugs. This type of screens can lead to the identification of novel antibiotic targets as well as novel synergistic target combinations.

3 Publications

- Bikard, D., Euler, C.W., Jiang, W., Nussenzweig, P.M., Goldberg, G.W., Duportet, X., Fischetti, V.A., and Marraffini, L.A. (2014). Exploiting CRISPR-Cas nucleases to produce sequence-specific antimicrobials. Nature Biotechnology *32*, 1146–1150.
- Bikard, D., and Barrangou, R. (2017). Using CRISPR-Cas systems as antimicrobials. Current Opinion in Microbiology *37*, 155–160.
- Rousset, F., Cui, L., Siouve, E., Becavin, C., Depardieu, F., and Bikard, D. (2018). Genome-wide CRISPR-dCas9 screens in E. coli identify essential genes and phage host factors. PLOS Genetics *14*, e1007749.

First Name / Last name: Ala-Eddine Deghmane
Contact : *ala-eddine.deghmane @ pasteur.fr*

Unit: Invasive Bacterial Infections
IP Department or IP: Global Health
Secondary affiliation: Microbiology

Main domains 1: Bacteria

Main domain 2:: surveillance and epidemiology, mechanism of resistance and dissemination,
new molecules, alternative strategies,

Attractive synopsis:

Emergence and mechanisms of resistance to beta lactams in invasive bacterial infections

Research projects in relation with AMR:

The polysaccharide-encapsulated bacteria *Neisseria* meningitidis, *Haemophilus influenzae* (Hi) and *Streptococcus pneumoniae* are leading causes of serious bacterial infections. Beta lactams are the first line antibiotics in the treatment. These bacteria share the same ecological niche with possible DNA exchanges. They also present similarity of the mechanisms of resistance to beta lactams through modifications of penicillin binding proteins. We use genomic approaches, structure-function analysis and in vivo validation using relevant animal models. A major force of our work is the use of large collections of clinical isolates from diseased subjects as well as establishing databases for molecular characterisation of resistance to beta lactams. Our work also brings together complementary expertise through collaborations in the fields of molecular epidemiology, pathogenesis and structure-function analysis.

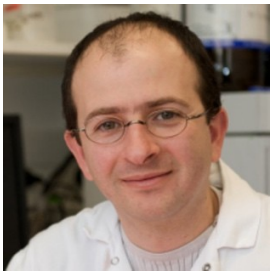
3 Publications

- Hong E, Deghmane AE, Taha MK. Acquisition of beta-lactamase by Neisseria meningitidis through possible horizontal gene transfer. Antimicrob Agents Chemother. 2018 Jun 25;62(9):e00831-18.

X

- Belkacem N, Hong E, Antunes A, Terrade A, Deghmane AE, Taha MK. Use of Animal Models To Support Revising Meningococcal Breakpoints of beta-Lactams. Antimicrob Agents Chemother. 2016 Jul;60(7):4023-7.

- Zarantonelli ML, Skoczynska A, Antignac A, El Ghachi M, Deghmane AE, Szatanik M, et al. Penicillin resistance compromises Nod1-dependent proinflammatory activity and virulence fitness of *Neisseria meningitidis*. Cell Host Microbe. 2013;13(6):735-45.



First Name / Last name: Ivo GOMPERTS BONECA
Contact : *bonecai @ pasteur.fr*

Unit: Biology and genetics of the bacterial cell wall
IP Department or IP: Microbiology
Secondary affiliation: Immunology

Main domains 1: Bacteria

Main domain 2: (possibly more than one response): mechanism of resistance and dissemination, new molecules, alternative strategies.

Attractive synopsis:

Searching for new therapeutic strategies targeting the assembly of the cell wall of bacteria

Research projects in relation with AMR:

Our laboratory is specialized in the bacterial cell wall and is focused in developing new antibiotics targeting the assembly of the cell wall by new unexplored mechanisms. We have chosen to tackle two distinct processes in the assembly of the bacterial cell envelop. One is aimed at the lipoprotein maturation pathway and in particular the last step catalysed by the essential Lnt enzyme. The second is aimed at the peptidoglycan assembly complexes, in particular, the one required for cell elongation. In this latter approach, we developed a FRET assay to screen for molecules that interfere with the PBP2-MreC complex at the core of the elongasome protein complex assembly.

3 Publications

- **...** El Ghachi M, P-J Matteï, C Ecobichon, A Martins, S Hoos, C Schmitt, F Colland, C Ebel, M-C Prévost, F Gabel, P England, A Dessen, **IG Boneca**. 2011. Characterization of the elongasome core PBP2:MreC complex of *Helicobacter pylori.* ***Mol Microbiol***. 82(1):68-86.
- **...** Contreras-Martel C, A Martins, C Ecobichon, D Maragno Trindade, P-J Matteï, S Hicham, P Hardouin, M El Ghachi, **IG Boneca***, A Dessen*. **2017**. Molecular architecture of PBP2:MreC core bacterial cell wall synthesis complex. ***Nat Comm***. 8(1):776. (* co-corresponding author).
- **...** Gélis-Jeanvoine S., S Lory, J Oberto and **N Buddelmeijer. 2015**. Residues located on membrane-embedded flexible loops are essential for the second step of the apolipoprotein N-acyltransferase reaction. ***Mol. Microbiol***. 95: 692-705.

First Name / Last name: Brice SPERANDIO
Contact : *brice.sperandio@pasteur.fr*

Unit: PMM
IP Department or IP: BCI
Secondary affiliation: INSERM U1202

Main domains 1: Bacteria

Main domain 2: New molecules, alternative strategies

Attractive synopsis:

Deciphering the regulation of human innate antimicrobial mechanisms to develop innovative immuno-stimulatory molecules.

Research projects in relation with AMR:

Recurrent enteric infections in children of low income countries, chronic inflammatory bowel diseases, nosocomial diseases, myelosuppressive and immunosuppressive therapies are, among others, clinical situations that require strong boosting of mucosal innate defenses to respectively avoid luminal bacterial overgrowth causing malnutrition, chronic inflammation, opportunistic infection, and deadly translocation. My challenge is to translate fundamental findings in the genetic and epigenetic regulation of antimicrobial mechanisms of the epithelium into drugs and interventions boosting their expression. In contract with the high-throughput screening platform of Institut Pasteur Korea, we aim to identify molecules from the pharmacopoeia inducing strong expression of epithelial antimicrobial molecules, without triggering significant inflammation. Such candidate molecules would represent a breakthrough in the field of anti-infectious strategies in an era of limitation of antibiotic discovery and use, in face of the spread of resistance.

3 Publications:

- Sechet E., Telford E., Bonamy C., Sansonetti P.J. & Sperandio B. (2018) Natural molecules induce and synergize to boost expression of the human antimicrobial peptide beta-defensin-3. *PNAS* E9869-E9878
- Bonamy C., Sechet E., Amiot A., Alam A., Mourez M., Fraisse L., Sansonetti P.J. & Sperandio B. (2018) Expression of the human antimicrobial peptide beta-defensin-1 is repressed by the EGFR-ERK-MYC axis in colonic epithelial cells. *Nature Scientific Reports* 18043
- Fischer N., Sechet E., Friedman R., Amiot A., Sobhani I., Nigro G., Sansonetti P.J. & Sperandio B. (2016) Histone deacetylase inhibition enhances antimicrobial peptide but not inflammatory cytokine expression upon bacterial challenge. *PNAS* E2993-E3001

First Name / Last name: Sylvain Brisse
Contact : sylvain.brisse@pasteur.fr

Unit: BEBP (Biodiversity and Epidemiology of Bacterial Pathogens)
IP Department or IP: Global Health
Secondary affiliation: G& G

Main domains 1: Bacteria

Main domain 2: surveillance and epidemiology, technological and methodological developments.

Attractive synopsis:

We use genomics and bioinformatics to understand the global dissemination of multidrug resistance strains and we provide to the community, unified nomenclatures ("genomic taxonomies of strains") that allow global communication on strain subtypes and their tracking across time, geography and activity sectors ("One Health").

Research projects in relation with AMR:

Several ongoing projects on epidemiology and ecology of Klebsiella:

- One Health Klebsiella (MedVetKlebs; funding: One Health EJP), 10 EU partners
- klebNET: Global and One health Klebsiella (35 partners, international)
- SpARK (coord. Ed Feil; JPIAMR funding)
- CRISPR-ATTACK (coord Beisel Chase, JPIAMR funded): CRISPR applications against Klebsiella
- klebGAP (coord. Arnfinn Sundsfjord, Funding by a Norwegian Foundation): epidemiology and pathogenesis of Klebsiella

We also study antimicrobial resistance in *Corynebacterium diphtheriae*

3 Publications

- Perrin A, 27 authors Brisse S. 2017. Evolutionary dynamics and genomic features of the Elizabethkingia anophelis 2015 to 2016 Wisconsin outbreak strain. Nat Commun 8:15483.
- Bialek-Davenet S, Criscuolo A, Ailloud F, Passet V, Jones L, Delannoy-Vieillard AS, Garin B, Le Hello S, Arlet G, Nicolas-Chanoine MH, Decre D, Brisse S. 2014. Genomic definition of hypervirulent and multidrug-resistant Klebsiella pneumoniae clonal groups. Emerging infectious diseases 20:1812–20.
- Moura A, 28 authors Brisse S. 2016. Whole genome-based population biology and epidemiological surveillance of Listeria monocytogenes. Nature Microbiology 2:16185.

Carmen Buchrieser
Contact: *cbuch @ pasteur.fr*

Unit: Biology of Intracellular Bacteria
IP Department or IP: Department Microbiology
Secondary affiliation: Department Genomes and genetics

Main domain 1: Bacteria

Main domain 2: alternative strategies, technological and methodological developments.

Attractive synopsis:
We are investigating how immunological responses, metabolic signalling, metabolic fluxes, and subcellular architecture of host cells are modified during infection by intracellular pathogens using *Legionella pneumophila* as a model. This knowledge will be used to identify immunometabolic drugs to tackle infection and to target host pathways instead of the pathogen.

Research projects in relation with AMR:
Influence of the lung microbiome on transmission of antibiotic resistance and analyses of the development of antibiotic resistance during long-term treatment of pneumonia: We are characterizing pulmonary samples from patients with pneumonia due to *Legionella* or *Pseudomonas* using cutting edge technologies (metagenomics, resistome analyses, 3C-metagenomics and pathogen evolution monitored by whole genome sequencing). Combined analyses will lead to knowledge with impact on treatment strategies and may result in the identification of novel microbiome markers suggesting increased sensitivity to develop pneumonia, severity of infection and antibiotic resistance.
Immunometabolism and *Legionella pneumophila* infection a new way to tackle infection: We combine high-content microscopy, metabolic profiling and metabolomics to monitor multiple cellular immunometabolic phenotypes in infected cells. We aim to reveal new, general mechanisms and strategies of how pathogens counteract the host defence and how the host responds to infection. An integrative view of infection-induced changes on subcellular architecture, metabolism and immunity is of utmost importance in order to understand how intracellular bacteria cause infection and disease and, in the era of a scaring rise of antibiotic resistance worldwide, may allow the usage of immunometabolic drugs and/or the generation of novel anti-infectious drugs attacking the disease mechanisms instead of the microbes.

Publications
- Escoll P, Song OR, Viana F, Steiner B, Lagache T, Olivo-Marin JC, Impens F, Brodin P, Hilbi H, Buchrieser C. (**2017**) *Legionella pneumophila* Modulates Mitochondrial Dynamics to Trigger Metabolic Repurposing of Infected Macrophages. ***Cell Host Microbe.*** 22(3):302-316.e7.
- Escoll P, Buchrieser C. (**2018**) Metabolic reprogramming of host cells upon bacterial infection: Why shift to a Warburg-like metabolism? ***FEBS J.*** 285(12):2146-2160.
- Rolando M, Escoll P, Nora T, Bauvy C, Bedia C, Daniels C, Abraham G, Stogios PJ, Skarina T, Christophe C, Dervins-Ravault D, Cazalet C, Hilbi H, Rupasinghe T, Tull D, McConville M, Ong SY, Hartland E, Codogno P, Levade T, Nader T, Savchenko A and C. Buchrieser. (2016) *Legionella pneumophila* S1P-lyase targets host sphingolipid metabolism to restrain Autophagy. ***Proc Natl Acad Sci U S A.*** 113(7):1901-6

First Name / Last name: Nienke BUDDELMEIJER
Contact : *nienke.buddelmeijer@pasteur.fr*

Unit: Biology and Genetics of the Bacterial Cell Wall (BGPB)
IP Department or IP: Microbiology
Secondary affiliation: Immunology

Main domain 1: Bacteria

Main domain 2: new molecules, alternative strategies, technological and methodological developments.

Attractive synopsis:

Lipoprotein modification in bacteria: a novel target for antibiotics

Research projects in relation with AMR:

We study the molecular mechanism of bacterial lipoprotein modification, in particular of the essential integral membrane enzymes prolipoprotein:phosphatidylglycerol diacylglyceryltransferase (Lgt) and apolipoprotein Nacyltransferase (Lnt), and screen for small inhibitory molecules using high-throughput screening (HTS) with specific fluorescence *in vitro* assays and *in silico* design. We use medicinal chemistry approaches to optimize potential hits, and biophysical data and crystal structures of enzyme-inhibitors complexes for the development of novel antibacterial agents.

3 Publications

- Wiktor M., Weichert D., Howe N., Huang C., Olieric V., Boland C., Bailey J., Vogeley L., Stansfeld P. J., Buddelmeijer N., Wang M., Caffrey M. (**2017**) Structural insights into the mechanism of the membrane integral N-acyltransferase step in bacterial lipoprotein synthesis. Nature communications 8, 15952. PMID: 28675161. DOI:10.1038/ncomms15952.
- Gélis-Jeanvoine S., Lory S., Oberto J. and Buddelmeijer N. (**2015**) Residues located on membrane-embedded flexible loops are essential for the second step of the apolipoprotein N-acyltransferase reaction. Mol. Microbiol. 95: 692-705. PMID: 25471278. DOI: 10.1111/mmi.12897.
- Pailler J., Aucher W., Pires M. and Buddelmeijer N. (**2012**) Prolipoprotein diacylglycerol ::diacylglyceryltransferase (Lgt) has seven transmembrane segments and its essential residues are located in the membrane. J. Bacteriol. 194: 2142-2151. PMID: 22287519. DOI: 10.1128/JB.06641-11

First Name / Last name: Dominique CLERMONT
Contact : *dominique.clermont@pasteur.fr*

Unit: Collection of the Institut Pasteur
IP Department or IP: Microbiology
Secondary affiliation: /

Main domains 1: Bacteria

Main domain 2: Mechanism of resistance and dissemination

Attractive synopsis:

Screening for metabolites potentially usable as new antimicrobials and Study of the history and evolution of resistance in ESKAP pathogens (*Enterococcus faecium*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter baumanii*, *Pseudomonas aeruginosa*) are expanding the bacterial landscape of innovative solutions to combating AMR today.

Research projects in relation with AMR:

The Collection of the Institut Pasteur (CIP) maintains a broad diversity of prokaryotic species from which new antimicrobial compounds can be discovered. The CIP facilitates access to biological resources in France and abroad following safety standards for health and the environment in compliance with regulations and laws, and ensuring maximum traceability. In addition, it is involved in different research projects focused on bacterial genomics and proteomics, as well as on preservation and distribution of control strains. This is done within Europe and non-European countries, in order to facilitate cross-disciplinary experimental works and to innovate with new better-performing methodologies involving partnerships between private and public laboratories. Bioinformatics pipelines are being developed in collaboration with C3BI to implement classical resistance gene prediction workflows among the main pathogens. The added value of CIP is the provision of bacterial strains isolated from clinical samples before the discovery of most antibiotics used nowadays. Detection of what is not already known as mechanism of resistance is the ultimate goal of our genomic research.

3 Publications

- Evolutionary dynamics and genomic features of the Elizabethkingia anophelis 2015 to 2016 Wisconsin outbreak strain. Perrin et al., 2017. Nature Communications.
- Global phylogeography and evolutionary history of Shigella dysenteriae type 1. Njamkepo et al., 2016. Nature Microbiology.
- Streptococcus agalactiae clones infecting humans were selected and fixed through the extensive use of tetracycline. Da Cunha et al., 2014. Nature Communications.

First Name / Last name: Hilde DE REUSE
Contact : hdereuse@pasteur.fr

Unit: *Helicobacter* pathogenesis unit
IP Department: Department of Microbiology
Secondary affiliation: Department of Genomes and Genetics

Main domain 1: Bacteria

Main domain 2: mechanism of resistance and dissemination, biomarkers and diagnostic, new molecules, alternative strategies.

Attractive synopsis:
Understanding the mode of action of existing medications and investigating new targets and molecules to fight infection by *Helicobacter pylori*, a priority pathogen for AMR that is responsible for 800,000 deaths every year worldwide.

Research projects in relation with AMR:
Helicobacter pylori is a bacterial pathogen of major importance that colonizes the stomach of half of the human population worldwide and is associated with the development of gastric cancer that kills about 800,000 individuals annually. *H. pylori* is till now the only bacterium recognized as a class 1 carcinogen by WHO and is part of the recent list of Ab-resistant "priority pathogens" that pose the greatest threat to human health. We develop two projects related to AntiMicrobial Resistance aimed at: (i) understanding the mode of action of bismuth in the new anti-*H. pylori* medication that associates antibiotics with this metal and, (ii) defining novel *H. pylori* therapeutic targets, validating molecules that are efficient against *H. pylori* growth both *in vitro* and in an animal model and defining the corresponding mechanisms.

3 Publications

- Salillas S, et al. Efficacy of flavodoxin inhibitors against *Helicobacter pylori* drug-resistant clinical strains and in *Hp*-infected mice. *Journal of Medicinal Chemistry* (*in revision*) *Patent:* ***PCT/ES2014/070011*** "Design, synthesis and efficacy testing of nitroethylene- and 7-nitrobenzoxadiazol-based flavodoxin inhibitors against *Helicobacter pylori* drug-resistant clinical strains and in *H. pylori*-infected mice"
- El Mortaji L, Aubert S, Galtier E, Schmitt C, Anger K, Redko Y, Quentin Y, De Reuse H*. (2018) The sole DEAD box RNA helicase of the gastric pathogen *Helicobacter pylori* is essential for colonization. *mBio* 9:e02071.
- Fischer F+, Robbe-Saule M+, Turlin E, Mancuso F, Michel V, Richaud P,. Veyrier F J, De Reuse H* and Vinella D (2016) Characterization in *H. pylori* of a nickel transporter essential for colonization that was acquired during evolution by gastric *Helicobacter* species. *PLoS Pathog* 12(12):e1006018

First Name / Last name: Laurent DEBARBIEUX
Contact : *laurent.debarbieux@pasteur.fr.*

Unit: Molecular Biology of Gene in Extremophiles
IP Department or IP: Microbiology
Secondary affiliation:

Main domain 1: Bacteria

Main domain 2: (possibly more than one response):, biomarkers and diagnostic, alternative strategies, technological and methodological developments.

Attractive synopsis:

Using animal models, we are elucidating the mechanisms supporting the efficacy of bacteriophages for treating infections caused by AMR bacteria.

Research projects in relation with AMR:

Our team has developed three complementary projects encompassing the study of:
i) molecular mechanisms of bacteriophages infecting antibiotics resistant strains of *Pseudomonas aeruginosa* and *Escherichia coli*, which are uncovering original bacterial targets for developing putative novel antibacterials;
ii) the efficacy of bacteriophage treatments for pulmonary infections, which includes deciphering the role of the immune system during phage therapy;
iii) the dynamic interactions between bacteriophages and bacteria in the intestinal microbiota with ecological and evolutionary impacts.
Our expertise in animal models in the field of bacteriophage research is internationally recognized.

3 Publications

- Comparative transcriptomics analyses reveal the conservation of an ancestral infectious strategy in two bacteriophage genera. Blasdel BG, Chevallereau A, Monot M, Lavigne R, Debarbieux L. (2017) ISME J. 11(9):1988-1996.
- Synergy between the Host Immune System and Bacteriophage Is Essential for Successful Phage Therapy against an Acute Respiratory Pathogen. Roach DR, Leung CY, Henry M, Morello E, Singh D, Di Santo JP, Weitz JS, Debarbieux L (2017) Cell Host Microbe, Jul 12;22(1):38-47
- The gut microbiota facilitates drifts in the genetic diversity and infectivity of bacterial viruses. De Sordi L, Khanna V, Debarbieux L. Cell Host Microbe. 2017 Dec 13;22(6):801-808.

First Name / Last name: Caroline Demangel
Contact : caroline.demangel@pasteur.fr

Unit: Immunobiology of Infection
IP Department or IP: Immunology
Secondary affiliation: Cell Biology and Infection

Main domain 1: Mycobacteria

Main domain 2: Host-pathogen interactions, Innate immunity, Mycobacterial virulence lipids, Host-derived antimicrobials

Attractive synopsis:

We aim to determine whether host-derived lipids may function as natural antibiotics, and can be considered as effectors of the innate immune system.

Research projects in relation with AMR:

Mycobacterium tuberculosis (Mtb), the causative agent of human Tuberculosis (TB), caused 1.6 million deaths in 2017 and it is estimated that 23% of the world's population has a latent TB infection. It is therefore urgent to develop new TB treatments able to control the emergence of drug-resistance and reduce the toxicity that is associated with current antibiotic therapies. Host-directed therapies (HDTs) represent promising approaches to fight TB since the manipulation of host immunity has the potential to improve bacterial clearance and limit the development of immunopathology. Moreover, because they target host proteins HDT should avoid the development of bacterial resistance. Our current project in relation with AMR aims to determine if lipid synthetic enzymes have the potential to modulate protective immunity in TB patients without inducing bacterial resistance.

3 Publications

- **Not yet**

Guillaume / Dumenil
Contact : *guillaume.dumenil@inserm.fr*

Unit: Pathogenesis of vascular infections
IP Department or IP: BCI
Secondary affiliation:

Main domains 1: Bacteria

Main domain 2: new molecules, alternative strategies

Attractive synopsis:

An anti-virulence strategy based on the inhibition of type IV pili is efficient on various bacteria including antibiotic resistant Neisseria gonorrhoeae

Research projects in relation with AMR:

The main objective of the project is to develop innovative drugs to treat gonorrhoeae with little or no emergence of resistance. In terms of mechanism, the drug would prevent *N. gonorrhoeae* attachment to the urogenital epithelium by blocking the surface expression of the bacterial type IV pili. At the molecular level these drugs inhibit an ATPase responsible for the assembly of type IV pili. Given the widespread expression of type IV pili among human pathogens such drugs would have broad applications. This new class of therapeutics based on the inhibition of virulence rather than bacterial survival bears the advantage of decreasing the emergence of resistance.

3 Publications

Aubey F, Corre J-P, Kong Y, Xu X, Obino D, Goussard S, Lapeyrere C, Souphron J, Couturier C, Renard S, Duménil G (2019) Inhibitors of the Neisseria meningitidis PilF ATPase provoke type IV pilus disassembly. Proceedings of the National Academy of Sciences: 201817757.

First Name / Last name: Bruno Dupuy
Contact: bdupuy@pasteur.fr

Unit: Pathogenesis of Bacterial Anaerobes
IP Department or IP: Microbiology
Secondary affiliation:

Main domain 1: Bacteria

Main domain 2: Mechanism of resistance and dissemination, New molecules, Alternative strategies

Attractive synopsis:

Development of new therapeutic potentials against *Clostridium difficile*

Research projects in relation with AMR:

The 1st project aims to synthesize in collaboration with the group of Pierre VERHAEGHE (CNRS Laboratoire de Chimie de Coordination, Université de Toulouse, CNRS) amino-acid polymers, called **peptidomimetics**, as simplified analogs of natural antimicrobial peptides (AMPs), in a view to study their antibiotic potential against the bacterium ***Clostridium difficile***, the leading cause of intestinal nosocomial post-antibiotic infections from occidental countries.
The 2nd project aims to synthesize in collaboration with the groups of Axel Hartke and Thierry Lequeux (U2RM and UMR6507, University of Caen) new inhibitors of DltA and DltC. Compounds combinations are increasingly seen as attractive options to fight the inexorable spread of antibiotic resistance and the use of Dlt inhibitors in combination with antibiotics will be tested for 5 major Gram-positive pathogens including *C. difficile*.

3 (max) Publications

- Not yet, ..
- **...**
- **...**

First Name / Last name: Olivier DUSSURGET
Contact : olivier.dussurget@pasteur.fr

Unit: *Yersinia* research unit
IP Department or IP: Department of Microbiology
Secondary affiliation: Department of Immunology

Main domains 1: bacteria

Main domain 2: new molecules

Attractive synopsis:

Design and synthesis of NAD kinase inhibitors as novel antibacterial agents

Research projects in relation with AMR):

Multidrug resistance is a major public health problem requiring urgent development of new antibiotics and thus identification of new bacterial targets. Our consortium selected the nicotinamide adenine dinucleotide kinase (NADK) as a valuable target for drug development. Structural and functional characterization of NADK led us to decipher its original enzymatic mechanism and to synthesize the first inhibitor active against pathogenic bacteria *in vitro*. Using a structure-based approach, we designed a new lead compound showing antibacterial activity in murine models of infection using clinically relevant pathogenic bacteria, including antibiotic-resistant isolates. Our data provide proof of concept that NADK is a druggable target. We are currently developing compounds with enhanced antibacterial activity and improved pharmacokinetic/pharmacodynamic properties.

3 Publications

- Paoletti J, Assairi L, Gelin M, Huteau V, Nahori MA, Dussurget O, Labesse G, Pochet S. 8-Thioalkyl-adenosine derivatives inhibit *Listeria monocytogenes* NAD kinase through a novel binding mode. *Eur. J. Med. Chem.,* 2016, 124, 1041-1056.
- Gelin M, Poncet-Montange G, Assairi L, Morellato L, Huteau V, Dugue L, Dussurget O, Pochet S, Labesse G. Screening and in situ synthesis using crystals of a NAD kinase lead to a potent antistaphylococcal compound. *Structure,* 2012, 20, 1107-1117.
- Pochet S, Labesse G, Gelin M, Assairi L, Dussurget O, Poncet-Montange G. Novel antibacterial compounds, Patent EP20100290679, WO2012090136A1.

First Name / Last name: Jost Enninga
Contact : *jost.enninga@pasteur.fr*

Unit: **Dynamics of host-pathogen interactions**
IP Department or IP: **Cell Biology and Infection**
Secondary affiliation: **-**

Main domain 1: Bacteria

Main domain 2: mechanism of resistance and dissemination, biomarkers and diagnostic, alternative strategies, technological and methodological developments.

Attractive synopsis:

Using an interdisciplinary research approach, we investigate how bacterial pathogens subvert host cells to reach intracellular niches to escape from antimicrobials and the host immune response.

Research projects in relation with AMR:

Many bacterial pathogens are internalized within host cells through active or passive mechanism. Upon uptake, they subvert host pathways to form intracellular niches that allow them to persist and to escape antibiotics as well as the host immune response. These pathogen-specific niches are highly important in the formation of antimicrobial resistance. Our team investigates the molecular and cellular pathways involved in the intracellular niche formation. For this, we develop innovative experimental strategies and novel biosensors that can be used for the identification of new antimicrobials against different pathogens, such as Shigella, Salmonella, Listeria and Mycobacterium tuberculosis.

3 Publications

- Voznica J, Gardella C, Belotserkovsky I, Dufour A, **Enninga J**, Stévenin V., *Identifying parameters of host cell vulnerability during Salmonella infection by quantitative image analysis and modeling.* ***Infect Immun***. 2017, pii: IAI.00644-17.
- Mellouk N., Weiner A., Aulner N., Schmitt C., Elbaum M., Shorte S., Danckaert A. and **Enninga J.** *Shigella subverts the host recycling compartment to rupture its endocytic vacuole.* **Cell Host Microbe,** 2014, Oct. 8(16):517-530
- Simeone R., Bobard A., Lippmann J., Bitter W., Majlessi L., Brosch R. and **Enninga J.**, *Phagosomal translocation of Mycobacterium tuberculosis results in toxicity and host cell death*, **PLoS Pathogens**, 2012 Feb;8(2):e1002507.

First Name / Last name: Jean-Marc GHIGO
Contact : *jmghigo@pasteur.fr*

Unit: Génétique des Biofilms
IP Department or IP: Microbiology
Secondary affiliation:

Main domains 1: **Bacteria**

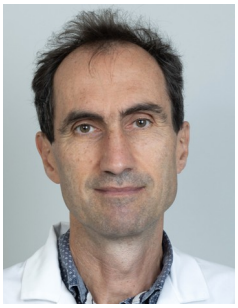
Main domain 2: Biofilm, tolerance to antibiotic, bacterial genetics.

Attractive synopsis:
Identification of functions promoting the rise and fall of antibiotic-tolerant bacterial biofilms

Research projects in relation with AMR (non confidential):
One of the hallmarks of surface-attached bacterial communities called biofilm is their high tolerance to antimicrobials and the difficulty to eradicate pathogenic bacterial biofilms plays a key role in recurrence of biofilm infections. Although biofilm tolerance is multifactorial, it mostly originates from subpopulations of non-growing persistent bacteria (persisters) that are able to resume growth and repopulate biofilm after ineffective antibiotic treatments. We showed that the SOS stress response induced in heterogeneous and nutrient-deprived biofilm microenvironments is involved in this phenotype and we are now exploring how tolerance emerging from bacterial biofilm communities could lead to the emergence of antibiotic resistance.

3 Publications

• Usui M*, Yokoo H, Tamura Y, Nakajima C, Suzuki Y, Ghigo JM, and Beloin C*. **(2019**). Zinc acetate potentiates the action of tosufloxacin on *Escherichia coli* biofilm persisters. ***Antimicrobial Agents and Chemotherapy***. Apr 1. pii: AAC.00069-19. doi: 10.1128/AAC.00069-19. (*co-corresponding authors)
• Szczesny, M., C. Beloin and J.-M. Ghigo **(2018)**. Increased osmolarity in biofilm triggers RcsB-dependent lipid A palmitoylation in *Escherichia coli*. ***mBio***Aug 21;9(4). pii: e01415-18. doi: 10.1128/mBio.01415-18.
• Létoffé, S., S. Chalabev, J. Dugay, F. Stressmann, B. Audrain, J.C. Portais, F. Letisse, and J.M. Ghigo. (2017). Biofilm microenvironment induces a widespread adaptive amino-acid fermentation pathway conferring strong fitness advantage in Escherichia coli. PLoS Genetics. May 19;13(5) doi: 10.1371.



First Name / Last name: Philippe Glaser
Contact : pglaser@pasteur.fr

Unit: Ecology and Evolution of Antibiotic Resistance
IP Department or IP: Microbiology
Secondary affiliation: Genomes and Genetics

Main domain 1: Bacteria

Main domain 2: surveillance and epidemiology, mechanism of resistance and dissemination

Attractive synopsis: We are addressing a major threat in antibiotic resistance: the global dissemination of multidrug resistant (MDR) lineages and of antibiotic resistant genes. We are focusing on carbapenemase producing enterobacteriaceae which are considered by WHO as “critical” priority pathogens.

Research projects in relation with AMR:
In tight collaboration with the team headed by Thierry Naas at the Bicêtre hospital and the National Reference Centre for Carbapenemase producing (CP) Enterobacteriaceae we are characterizing the genetic and functional bases for the dissemination of specific lineages of CP-*Escherichia coli* and CP-*Klebsiella pneumoniae*. By associating genomics, phylogenetic analyses, isolates phenotyping and metadata from the clinic we aim to identify candidate loci and mutations contributing to the dissemination of at risk clones. We are characterizing their evolutionary trajectories aiming to determine trough modelling the external factors contributing to their selection and dissemination. Hypotheses on the candidate factors are further challenged at the bench by reconstructing or complementing mutant strains, deriving experimental evolution studies, transcriptional analyses and by in depth characterizing clinical isolates from single outbreaks and from single patients.

3 Publications

- Patiño-Navarrete R, Rosinski-Chupin I, Cabanel N, Gauthier L, Takissian J, Madec J-Y, Hamze M, Bonnin RA, *Naas T, & *Glaser P (2018) Stepwise evolution and convergent recombination underlie the global dissemination of carbapenemase-producing Escherichia coli bioRxiv.
- Jousset AB, Bonnin RA, Rosinski-Chupin I, Girlich D, Cuzon G, Cabanel N, Frech H, Farfour E, Dortet L, *Glaser P, & *Naas T (2018) 4.5 years within-patient evolution of a colistin resistant KPC-producing Klebsiella pneumoniae ST258. Clin. Infect. Dis.
- *Dortet L, *Glaser P, Kassis-Chikhani N, Girlich D, Ichai P, Boudon M, Samuel D, Creton E, Imanci D, Bonnin R, Fortineau N, & Naas T (2018) Long-lasting successful dissemination of resistance to oxazolidinones in MDR Staphylococcus epidermidis clinical isolates in a tertiary care hospital in France. J. Antimicrob. Chemother. 73(1):41-51.

First Name / Last name:
Contact : *simonetta.gribaldo@pasteur.fr*

Unit: Evolutionary Biology of the Microbial Cell
IP Department or IP: Microbiology
Secondary affiliation: Genomes and Genetics

Main domain 1: Bacteria, Archaea

Main domain 2: (possibly more than one response): mechanism of resistance and dissemination, alternative strategies, technological and methodological developments.

Attractive synopsis:
By studying fundamental cellular processes in bacteria and archaea, our research can discover novel targets for antimicrobial agents

Research projects in relation with AMR):

Most of our knowledge on fundamental cellular processes in prokaryotes come from a limited number of models and/or pathogens. Our expertise is large-scale comparative genomics and phylogenomics across the whole diversity of bacteria and archaea (cultured and uncultured). We have established robust reference phylogenies which are essential to frame their diversity and carry out evolutionary analyses. We are now developing two new experimental models, one bacterial (*Veillonella parvula*) and one archaeal (*Methanobrevibacter smithii*), both representing abundant but largely understudied components of the human microbiota. Our objective is to discover new mechanisms for the biogenesis and functioning of bacterial and archaeal cell envelopes, including cell wall synthesis, cell division, appendices, and motility, which may identify new targets for antimicrobials.

3 Publications

- Antunes LS, Poppleton D, Klingl A, Criscuolo C, Dupuy B, Brochier-Armanet C, Beloin C, and Gribaldo S (2016) Phylogenomic analysis supports the ancestral presence of LPS outer membranes in the Firmicutes. *Elife* Aug 31;5. pii: e14589.
- Poppleton DI, Duchateau M, Hourdel V, Matondo M, Flechsler J, Klingl A, Beloin C, Gribaldo S (2017) Outer membrane proteome of Veillonella parvula : a diderm Firmicute of the human microbiome. Front Microbiol 8 : 1215.
- Borrel G, Adam PS, McKay LG, Chen L, Sierra-García IN, Sieber CMK, Andersen GL, Li WJ, Hallam SJ, Muyzer G, Maia de Oliveira V, Inskeep WP, Banfield JF, and Gribaldo S (2019) Wide diversity of methane and short-chain alkane metabolisms in uncultured archaea. *Nat Microbiol.* 4(4):603-613.

First Name / Last name: Didier Guillemot
Contact : *didier.guillemot@pasteur.fr*

Unit: Pharmacoepidemiology and Infectious Diseases
IP Department or IP: Global Health
Secondary affiliation: none

Main domain 1: Bacteria

Main domain 2: mechanism of resistance and dissemination, new molecules, technological and methodological developments.

Attractive synopsis:

We aim at decipheing dynamic causal links between human contacts, bacterial intrinsic transmission capacities for resistant gene vectorization, factors affecting human and environmental microbiota disruption, and the burden of antibiotic resistance

Research projects in relation with AMR:

Beyond the traditional investigation and analysis techniques (biostatistics and biomathematics) used in infectious disease epidemiology we develop specific methodological approaches initiated within our research group in recent years as a result of:
1. Dynamic contact monitoring of antibiotic resistant bacteria (ARB) dissemination;
2. Temporospatial analysis and microspatial simulation of ARB dissemination
3. Dynamic Network Analysis and Agent Based Simulation

3 (max) Publications

- Cheysson F, Vibet M-A, Guillemot D, Watier L. Estimation of exposure-attributable fractions from time series: a simulation study. Stat Med. 25 June 2018. https://doi.org/10.1002/sim.7818
- Duval A, Obadia T, Martinet L, Boëlle PY, Fleury E, Guillemot D, Opatowski L, Temime L; i-Bird study group. Measuring dynamic social contacts in a rehabilitation hospital: effect of wards, patient and staff characteristics. Sci Rep. 2018. https://doi.org/10.1038/s41598-018-20008-w
- Domenech De Cellès M, Pons-Salort M, Varon E, Vibet M–A, Caroline Ligier, Letort V, Opatowski L, and Guillemot D. Interaction of Vaccination and Reduction of Antibiotic Use Drives Unexpected Increase of Pneumococcal Meningitis. 2015. Sci Rep 2015. https://doi.org/10.1038/srep11293

First Name / Last name: Mélanie Hamon
Contact : *melanie.hamon@ pasteur.fr*

Unit: G5 Chromatin and Infection
IP Department or IP: Cell Biology and Infection
Secondary affiliation:

Main domains 1: Bacteria

Main domain 2: new molecules, alternative strategies, technological and methodological developments.

Attractive synopsis:

Our work focuses on bacteria-host interactions, with the aim to target host mechanisms essential for bacterial growth and find alternatives to current direct-acting anti-bacterial drugs.

Research projects in relation with AMR (non confidential):

In our research projects, we aim to characterize original mechanisms of chromatin-bacteria cross talk, focusing on two pathogens, *Streptococcus pneumoniae* and *Listeria monocytogenes*. Indeed, recent studies, including my own, have shown that pathogenic bacteria induce chromatin modifications in order to reprogram host transcription during infection, and this process is essential for efficient bacterial replication. Targeting such host mechanisms are therefore detrimental for infection and could provide alternative treatment against pathogen invasion.

In a collaborative project we are also developing new tools based on modified DNA aptamers coupled to photodynamic therapy for the specific detection and eradication of individual bacterial strains.

3 Publications

- Hamon M.A., Batsche E, Regnault B, Tham TN, Seveau S, Muchardt C, Cossart P. Histone modifications induced by a family of bacterial toxins. *Proc Natl Acad Sci*. 2007. 104: 13467-72.
- Eskandarian, H. A., Impens, F., Nahori M.A., Soubigou G., Coppée J. Y., Cossart P., Hamon, M. A. A role for SIRT2-dependent histone H3K18 deacetylation in bacterial infection. *Science*. 341, 1238858 (2013)
- Pereira, J.M., Chevalier, C., Chaze, T., Gianetto, Q., Impens, F., Matondo, M., Cossart, P., and Hamon, M.A. (2018). Infection reveals a modification of SIRT2 critical for chromatin association. *Cell Reports* **23,** 1124–1137 (2018).**...**

First Name / Last name: Molly Ingersoll
Contact : *molly.ingersoll@pasteur.fr*

Unit: Group of Mucosal Inflammation and Immunology (ICD Unit)
IP Department or IP: Immunology
Secondary affiliation:

Main domain 1: Bacteria

Main domain 2: mechanism of resistance in the host, biomarkers and diagnostic, alternative non-antibiotic strategies, technological and methodological developments.

Attractive synopsis:
We are developing non-antibiotic based immunomodulatory therapeutic approaches to combat multidrug resistant urinary tract infections

Research projects in relation with AMR (non confidential):
Treatment for urinary tract infection (UTI) relies upon antibiotics, which only treat acute UTI, do not prevent recurrence, and are not efficacious against rapidly disseminating multidrug resistant uropathogenic *E. coli* (UPEC). We are identifying novel treatment strategies targeting host pathways to treat UTI without antibiotics. Our data suggest that the innate response shapes the strength and nature of the long-term adaptive response to UTI. We test how manipulating the innate host response impacts development of UPEC-specific long-lasting immunity, focusing on three non-antibiotic based therapies approved for use in humans in other contexts.

3 (max) Publications

- Zychlinsky Scharff A, Rousseau M, Lacerda Mariano L, Canton T, Consiglio CR, Albert ML, Fontes M, Duffy D, **Ingersoll MA**. Sex differences in IL-17 contribute to chronicity in male versus female urinary tract infection. JCI Insight, 2019 May 30;5. pii: 122998. doi: 10.1172/jci.insight.122998
- Rousseau M, Goh HM, Holec S, Albert ML, Williams RB, **Ingersoll MA*** & Kline KA. Bladder catheterization increases susceptibility to infection that can be prevented by prophylactic antibiotic treatment. JCI Insight. 2016 Sep 22;1(15):e88178. PubMed PMID: 27699248 *co-last author
- Mora-Bau G, Platt AM, van Rooijen N, Randolph GJ, Albert ML & **Ingersoll MA**. Macrophages Subvert Adaptive Immunity to Urinary Tract Infection. PLoS Pathog. 2015 Jul 16;11(7):e1005044. doi: 10.1371/journal.ppat.1005044. eCollection 2015 Jul. PubMed PMID: 26182347.

First Name / Last name: Daniel LADANT
Contact : *daniel.ladant@pasteur.fr*

Unit: Biochimie des Interactions Macromoléculaires
IP Department or IP: Département de Biologie Structurale et Chimie - CNRS UMR 3528
Secondary affiliation: Département de Microbiologie

Main domains 1: Bacteria

Main domain 2: new molecules, technological and methodological developments.

Attractive synopsis:

Our goal is to exploit an innovative and robust target-based *in vivo* screening pipeline based on a robust bacterial two hybrid screening technology (BACTH) to identify small-molecules able to block the assembly of bacterial cell division machinery.

Research projects in relation with AMR:

The bacterial cell division machinery (divisome) is attracting considerable attention as a key target for new antibiotics, as its components are very specific to the prokaryotic kingdom [1]. Our objective is to exploit a robust and cost-effective experimental screening pipeline to identify novel compounds targeting key protein-protein interactions (PPI) implicated in the divisome assembly. This technique, based on a bacterial two hybrid assay (BACTH; developed in the laboratory), can detect PPI with simple phenotypic assays in *Escherichia coli* [2,3]. Collections of compounds are screened to identify high-quality hits able to block *in vivo*, in the native environment (e.g. cytosol, membrane or periplasm) of bacterial cells, critical PPI among key divisomal components. The BACTH screening pipeline will also be used to identify inhibitors of PPI implicated in other key processes in bacterial physiology. *(In collaboration with Hélène Munier-Lehmann, plateforme Criblage Biologique et Chimiothèques, Unité de Chimie et Biocatalyse, & Fabrice Agou, Plate-Forme de criblage chémogénomique et biologique, Institut Pasteur.)*

3 Publications

1. Den Blaauwen, T., Andreu, J.M. and Monasterio, O.(2014) Bacterial cell division proteins as antibiotic targets. Bioorganic Chem. 55, 27–38.
2. Karimova G., Dautin N., and Ladant D. (2005) Interaction network among *Escherichia coli* membrane proteins involved in cell division as revealed by bacterial two-hybrid analysis. *J. Bact*. 187:2233-2243
3. Ouellette, S. P., Karimova, G., Davi, M., and Ladant, D. (2017). Analysis of membrane protein interactions with a bacterial adenylate cyclase–based two-hybrid (BACTH) technique. *Current Protocols in Molecular Biology*, *118*,20.12.1–20.12.24.doi: 10.1002/cpmb.36

First Name / Last name:
Contact: *marc.lecuit@pasteur.fr*

Unit: **Biology of Infection Unit**
IP Department or IP: **Cell Biology and Infection**
Secondary affiliations: **Global Health, Virology**

Main domains 1: Bacteria

Main domain 2: surveillance and epidemiology, mechanism of resistance and dissemination, biomarkers and diagnostic, alternative strategies

Attractive synopsis:

We follow an interdisciplinary approach to understand Listeria biology, its pathogenic potential and saprophytic life, and its dynamics within and outside the host.

Research projects in relation with AMR:

Listeria monocytogenes (Lm) is one of the deadliest foodborne pathogens. Although acquired antimicrobial resistance is rare in Lm, listeriosis is a difficult-to-treat infection, with a case fatality rate of up to 30% under appropriate therapy. We study the pathophysiology of listeriosis to precisely determine (i) the routes taken by Lm from the gut lumen to its final target organs, the brain and the fetal-placental unit, (ii) the niches where it resides, and (iii) Lm within-host dynamics. Beside improving our basic understanding of the biology of infections, this may help improve the clinical efficacy of antimicrobial strategies. We also aim to identify the niches in which Listeria thrive outside the host, and the transmission routes from animals to food and human, to try to understand why Lm, in contrast to most other foodborne pathogens, exhibits such a low degree of antimicrobial resistance, anticipating that this may lead to new way to prevent resistance acquisition in other bacterial species of medical interest.

3 Publications

- Epistatic control of intrinsic resistance by virulence genes in Listeria. Scortti M, Han L, Alvarez S, Leclercq A, Moura A, Lecuit M, Vazquez-Boland J. **PLOS Genet**. **2018**;14(9):e1007525
- Clinical features and prognostic factors of listeriosis: the MONALISA national prospective cohort study. Charlier C, …, Lecuit M; MONALISA study group. **Lancet Infect Dis. 2017**;17(5):510-519
- Whole genome-based population biology and epidemiological surveillance of Listeria monocytogenes. Moura A, ..., Lecuit M, Brisse S. **Nat Microbiol. 2016**;10;2:16185

First Name / Last name: Emmanuel LEMICHEZ
Contact : *emmanuel.lemichez@pasteur.fr*

Unit: Bacterial Toxins Unit
IP Department or IP: Microbiology
Secondary affiliation: Cell Biology of Infection

Main domains 1: Bacterial Toxins, Virus

Main domain 2: new molecules, alternative strategies

Attractive synopsis:
We pursue novel promising approaches aiming at fighting toxi-infections through development of chemical compounds acting directly on host components to confer to cells resistance properties to the arsenal of pathogens and stimulate innate immune responses. This shall offer the possibility to act specifically on pathogenesis without disturbing the microbiota of the host and minimizing development of resistance.

Research projects in relation with AMR:

To tackle the question of pathogen resistance, we follow two original research directions. One promising strategy **deals with the targeting of host regulators of the vesicular trafficking using chemical compounds** to inhibit the action of a large spectrum of bacterial protein AB toxins. These toxins are among the most potent poisons found in Nature. Acting selectively on host factors to disarm pathogens is without risk of selecting resistant microbes and shall offer protection without impacting the microbial flora. Specifically, we are characterizing the mode of action of a family of compounds we have isolated with collaborators from the CEA. These small compounds have the capacity to block the action of various AB-like toxins notably those produced by gastrointestinal tract pathogens, such as diarrheagenic strains of *E. coli* and *Clostridium difficile*. The second research axis to solve the question of antibiotic resistance rise deals with **stimulating the host defenses**. Our project is aiming at **providing therapeutic and prophylactic molecules to stimulate simultaneously innate and acquired immune defenses,** by mimicking the immunostimulatory effect of Rho GTPase activation by the CNF1 toxin. Such therapy or/and prophylaxis could be used as alternative or complements to antibiotics, antivirals, anti-parasitic or anti-fungal therapies.

3 Publications

- Mahtal N, Brewee C, Pichard S, Visvikis O, Cintrat JC, Barbier J, **Lemichez** E*, **Gillet** D*
  Screening of a Drug Library Identifies Inhibitors of Cell Intoxication by CNF1.
  ChemMedChem. 2018 Apr 6;13(7):754-761. doi: 10.1002/cmdc.201700631.

- Wu Y et al. ABMA, a small molecule that inhibits intracellular toxins and pathogens by interfering with late endosomal compartments.
  Sci Rep. 2017 Nov 14;7(1):15567. doi: 10.1038/s41598-017-15466-7.

- Michel G, Ferrua B, Munro P, Boyer L, Mathal N, Gillet D, Marty P, **Lemichez** E.
  Immunoadjuvant properties of the RhoA activating factor CNF1 in prophyloactic and curative vaccination against *Leishmania infantum*
  PLoS One. 2016 Jun 3;11(6):e0156363. doi: 10.1371/journal.pone.0156363.

First Name / Last name: Giulia MANINA
Contact: *giulia.manina@pasteur.fr*

Unit: G5 Microbial Individuality and Infection
IP Department or IP: Cell Biology and Infection

Main domain 1: Bacteria

Main domain 2: mechanism of resistance and persistence, new molecules, alternative strategies, technological and methodological developments.

Attractive synopsis:

Drug enhancers that target phenotypic variation to accelerate treatment and prevent resistance.

Research projects in relation with AMR:

Tuberculosis ranks among the top ten causes of mortality and is the most frequent and deadly drug-resistant respiratory infection. About 10% of infected people experience symptoms during their lifetime, a risk that rises in certain vulnerable groups, and about a quarter of the world's population is an asymptomatic reservoir of infection. Our group focuses primarily on the ability of *Mycobacterium tuberculosis* to persist within the host and despite long-term therapy. We found that, prior to drug exposure, phenotypically distinct subpopulations exist that display different drug susceptibility. In light of this, we hypothesized that phenotypic variation favors persistence and consequently treatment failure. We screen for molecules that reduce phenotypic variation and homogenize bacterial susceptibility to drugs. Lastly, we aim to enhance the treatment of drug-recalcitrant tuberculosis.

3 Publications

- Phenotypic Heterogeneity in *Mycobacterium tuberculosis*. *Microbiol Spectr*. 2016 doi: 10.1128/microbiolspec.TBTB2-0021-2016.
- Preexisting variation in DNA integrity predicts the fate of single mycobacterial cells under stress (Manuscript in revision in the EMBO Journal).
- Multiplexable microfluidic culture chamber for imaging monolayer growth of single cells (Patent Application submitted to the European Patent Office).

:

First Name / Last name: Didier MAZEL
Contact : *mazel@pasteur.fr.*

Unit: Plasticité du Génome Bactérien
IP Department or IP: Genomes and Genetics
Secondary affiliation: Microbiologie

Main domains 1: Bacteria

Main domain 2: (mechanism of resistance and dissemination, new molecules, alternative strategies.

Attractive synopsis:

We fight resistance from both side: understanding the genetics behind its development and proposing alternate antimicrobial strategies

Research projects in relation with AMR:

Integrons are responsible for the capture and spread of most AB resistance genes in Gram negative pathogens. We are studying the recombination processes which have led to their evolutionary success. In parallel we study the selective effect driven by low AB concentrations such as those released in nature after waste water treatment. We are trying to understand how these low stresses are involved in resistance evolution. In addition, we study the transporters of a class of AB, in order to develop a strategy to improve their uptake. Finally, we are developing novel antimicrobial strategy, highly specific for a given pathogen, able to kill it in the context of the microbiota, and which will prevent the dysbiosis that broad host range ABs are usually causing.

3 Publications

- Lopez-Igual R, Bernal-Bayard J, Rodriguez-Paton A, Ghigo J-M and **Mazel D**. Engineering synthetic toxin-intein weapons as specific antimicrobials. *Nature Biotechnology* (2019) DOI : 10.1038/s41587-019-0105-3
- Escudero JA, Loot C, Parissi V, Nivina A, Bouchier C and **Mazel D**. Unmasking the ancestral activity of integron integrases reveals a smooth evolutionary transition during functional innovation. *Nature Communications* (2016) **7:**10937. doi: 10.1038/ncomms10937.
- Loot C, Nivina A, Cury J, Escudero JA, Ducos-Galand M, Bikard D, Rocha EPC and **Mazel D.** Differences in integron cassette excision dynamics shape a trade-off between evolvability and genetic capacitance. *mBio* (2017) **8**(2) pii: e02296-16. doi: 10.1128/mBio.02296-16

First Name / Last name: Javier PIZARRO-CERDA
Contact: *pizarroj@pasteur.fr*

Unit: Yersinia Research Unit, National Reference Center 'Plague & Other Yersinioses'
IP Department or IP: Microbiology
Secondary affiliation: Immunology, Cell Biology & Infection

Main domains 1: Bacteria

Main domain 2: (possibly more than one response): surveillance and epidemiology, new molecules, alternative strategies, technological and methodological developments.

Attractive synopsis:

We investigate both novel compounds against essential bacterial genes as well as novel bactericidal molecules against Gram-positive pathogens.

Research projects in relation with AMR:

In our Unit, we investigate different bacterial pathogens for their capacity to produce novel bactericidal molecules, or as models for the development of novel therapeutic drugs. In this context, we investigate the mechanisms of action of Listeriolysin S, a bacteriocin produced by Listeria monocytogenes *which displays a bactericidal activity against a restricted group of Gram-positive bacteria including* Staphylococcus aureus*. We are also developing novel compounds against the NAD kinase, which is an essential gene widely distributed among bacteria. Within the framework of the activities of the National Reference Centre 'Plague & Other Yersinioses', we also perform surveillance of antibiotic resistances present in different* Yersinia *species including* Y. enterocolitica, Y. pseudotuberculosis *as well as* Y. pestis.

3 Publications

- Gelin M, Poncet-Montange G, Assairi L, Morellato L, Huteau V, Dugué L, Dussurget O, Pochet S, Labesse G. **2012**. Screening and in situ synthesis using crystals of a NAD kinase lead to a potent anti-staphylococcal compound. *Structure* 20: 1107-17.

- Quereda JJ, Dussurget O, Nahori MA, Ghozlane A, Volant S, Dillies MA, Regnault B, Kennedy S, Mondot S, Villoing B, Cossart P, Pizarro-Cerda J. **2016**. Bacteriocin from epidemic Listeria strains alters the host intestinal microbiota to favor infection. *Proc Natl Acad Sci USA* 113: 5706-11.

- Quereda JJ, Nahori MA, Meza-Torres J, Sachse M, Titos-Jiménez P, Gomez-Laguna J, Dussurget O, Cossart P, Pizarro-Cerdá J. **2017**. Listeriolysin S Is a streptolysin S-like virulence factor that targets exclusively prokaryotic cells *in vivo*. *mBio* 8: e00259-17.

First Name / Last name: Eduardo ROCHA
Contact : *erocha@pasteur.fr*

Unit: Microbial Evolutionary Genomics
IP Department or IP: Genomes & Genetics
Secondary affiliation: Microbiology & Bioinformatics

Main domains 1: Bacteria

Main domain 2: mechanism of resistance and dissemination, biomarkers and diagnostic.

Attractive synopsis:

Our lab uses bioinformatics, genomics, metagenomics, and experimental approaches to understand how mobile genetic elements drive bacterial adaptation by way of horizontal gene transfer, including the acquisition of antibiotic resistance genes.

Research projects in relation with AMR:

Our lab has expertise in microbiology, bioinformatics, large scale genomics, modelling, experimental evolution, and molecular genetics. Our experimental work uses a diverse panel of *Klebsiella pneumoniae* strains, one of the most important nosocomial pathogens, to understand how its adaptation results from the interplay between the bacterial capsule and horizontal transfer driven by mobile genetic elements. Other key projects involve the use of metagenomics to study the perturbations caused in the human microbiome by antibiotic therapy in a clinical assay, the study of population genomics of nosocomial pathogens, the experimental evolution of associations between bacteria and eukaryotes, and the identification of the molecular mechanisms of horizontal transfer in bacteria.

3 Publications

- Rendueles O, Sousa JAM, Bernheim A, Touchon M, Rocha EPC (2018) Genetic exchanges are more frequent in bacteria encoding capsules. ***PLoS Genetics*** 14: e1007862.
- Bradley P, Bakker HCD, Rocha EPC, McVean G, Iqbal Z (2019) Real-time search of all bacterial and viral genomic data. ***Nature Biotechnology*** 37:152-9
- Cury J, Oliveira P, de la Cruz F, Rocha EPC (2018) Host range expansion and genetic plasticity drive the trade-off between integrative and extrachromosomal mobile genetic elements. ***Mol Biol Evol*** 35:2230–2239.

First Name / Last name: Lhousseine Touqui
Contact : *lhousseine.touqui@pasteur.fr*

Unit: Mucoviscidose et Bronchopathies Chroniques
IP Department or IP: Infection et Epidémiologie

Secondary affiliation:

Main domain 1: Bacteria.

Main domain 2: Alternative strategies

Attractive synopsis:
The use of antimicrobial peptides (AMPs) combined with nanoparticles (NPs) to fight multi-resistant bacteria.

Research projects in relation with AMR:
AMPs are and considered as the front-line of host defense molecules against invading pathogens (Salzman et al., 2010). Among AMPs, the human cathelicidin LL-37 (Fahy et a., 2005) and group IIA secreted phospholipases A2 (sPLA2s-IIA) (Nevalainen et al., 2008) are known to exert potent bactericidal effects on *Pseudomonas aeruginosa* and *Staphylococcus aureus*, respectively. These bacteria are known to exhibit high antibiotic resistance and the use of AMPs may represent an “alternative strategy” to control infections by these bacteria. However, the presence of biological lung barriers, such as mucus and biofilm, represent physical barriers that may prevent the access of AMPs to bacteria colonizing the respiratory system in patients with cystic fibrosis. Polymeric nanoparticles (NPs) are highly promising drug delivery vehicles that can protect AMPs against degradation by proteases and help AMPs to overcome mucus and biofilm in CF airways. We propose the use of combinations of silver NPs (AgNP) to AMPs for the optimization of the AMPs delivery and efficacy in killing multi-resistant bacteria in CF airways and other diseases characterized by mucus over-production.

3 Publications

- Geitani R, Ayoub Moubareck C, Touqui L, Karam Sarkis D. Cationic antimicrobial peptides: alternatives and/or adjuvants to antibiotics active against methicillin-resistant Staphylococcus aureus and multidrug-resistant Pseudomonas aeruginosa. BMC Microbiol. 2019 Mar 8;19(1):54. doi: 10.1186/s12866-019-1416-8.
- Pernet E, Guillemot L, Burgel PR, Martin C, Lambeau G, Sermet-Gaudelus I, Sands D, Leduc D, Morand PC, Jeammet L, Chignard M, Wu Y, **Touqui** L. Pseudomonas aeruginosa eradicates Staphylococcus aureus by manipulating the host immunity. Nat Commun. 2014 Oct 7;5:5105. doi: 10.1038/ncomms6105.
- Wu Y, Raymond B, Goossens PL, Njamkepo E, Guiso N, Paya M, **Touqui** L. 10.1038/ncomms6105.cType-IIA secreted phospholipase A2 is an endogenous antibiotic-like protein of the host. Biochimie. 2010 Jun;92(6):583-7. doi: 10.1016/j.biochi.2010.01.024. Epub 2010 Feb 6. Review.

First Name / Last name: Sven Van-Teeffelen
Contact : *sven.van-teeffelen@pasteur.fr*

Unit: G5 Microbial Morphogenesis and Growth
IP Department or IP: Microbiology
Secondary affiliation:

Main domain 1: Bacteria

Main domain 2: mechanism of resistance, technological and methodological developments.

Attractive synopsis:

Our lab works on the ability of cells to protect their mechanical integrity against cell-wall damage caused by antibiotics and other stresses, largely using single-cell time-lapse microscopy.

Research projects in relation with AMR (non confidential):

Our lab is interested in how cells maintain their cell envelopes intact and how they coordinate envelope expansion with growth. The cell envelope is a major target of antibiotics, and we are interested in the question whether cells actively respond to cell-envelope damage through antibiotics and other stresses. To that end, we investigate individual enzymes that might repair cell-wall damage as it occurs, using tools from single-cell and single-molecule microscopy, genetics, molecular biology, and physical modeling. Furthermore, we are interested in the response of cells to envelope perturbations, for example the response of macromolecule synthesis, which can provide an important contribution to antibiotic tolerance.

3 (max) Publications
None of our work on this subject is published at this point.

First Name / Last name: François Xavier Weill
Contact : *francois-xavier.weill @pasteur.fr*

Unit: Enteric Bacterial Pathogens
IP Department or IP: Global Health
Secondary affiliation: Microbiology

Main domain 1: Bacteria

Main domain 2: surveillance and epidemiology, mechanism of resistance and dissemination,

Attractive synopsis:

Thanks to a unique of historical bacterial isolates and collaboration with clinical laboratories, food and veterinary agencies at national or international level, we can carry out large-scale genomic studies on global collections, to determine population structures, phylogeographic patterns, and genetic evolution of antibiotic resistant enteric bacterial populations.

Research projects in relation with AMR:

Our research studies are strongly linked to our public health and reference activities (National and WHO Reference Centers for *Salmonella* spp, *Shigella* spp, enteric pathovars of *E. coli* and *Vibrio cholerae*). We work on three partly overlapping themes: (i) identification and dynamics of enteric bacterial populations resistant to antibiotics (ii) development of new bacterial typing and diagnostic tools, (iii) population structures and the evolution of genetically monomorphic pathogenic agents. We are interested, in particular in new emerging enteric bacterial pathogen populations (antibiotic-resistant and/or epidemic) and at clarifying the key steps leading to the emergence of these populations.

3 Publications

- FX Weill *et al*. Genomic insights into the 2016-2017 cholera epidemic in Yemen. ***Nature***, 2019;565: 230–233

- A Tran-Dien, S Le Hello, C Bouchier, FX Weill. Early transmissible ampicillin resistance in zoonotic *Salmonella enterica* serotype Typhimurium in the late 1950s: a retrospective whole-genome sequencing study. ***Lancet Infectious Diseases*****,** 2018;18:207-214.

- E Njamkepo, N Fawal, A Tran-Dien, …69 authors.., FX Weill. Global phylogeography and evolutionary history of *Shigella dysenteriae* type 1. ***Nature Microbiology*****,** 2016;1:16027.