PhD Fellowships in Life Sciences 2023:
Antibiotic resistance and *in vitro* biomarkers of treatment efficacy towards tuberculosis.

This project will be co-supervised by:
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Context / background:
With more than 10 million new infections and 1.5 million deaths (WHO 2021), tuberculosis (TB) represents a major public health concern. The emergence of drug resistant tuberculosis (DR-TB) reinforces the need for rapid and reliable antibiotic susceptibility testing (AST) to choose an effective anti-TB regimen. In 2019, the number of deaths attributable to DR-TB was estimated at 85,000, which represents about 7% of deaths associated with antibiotic-resistant bacteria (1). The treatment of TB, based on the combination of drugs, which is necessary to avoid the emergence of DR-TB and a treatment long enough for effective cure and to avoid relapse, is very challenging. Inappropriate antibiotic regimen might lead to patient relapse and development of antibiotic resistance in *Mycobacterium tuberculosis* (MTB) the etiologic agent of the disease. In addition, such regimens are not without toxic effects for the patient and their duration represents a challenge in terms of compliance.

In a translational and multidisciplinary research we aim to set-up a program for a personalized anti-TB treatment implying, i) validated therapeutic drug monitoring (TDM) program for all current and last generation drugs used against sensitive, multidrug-resistant tuberculosis (MDR-TB) and extensively drug-resistant (XDR) TB empowered with ii) precise AST and rapid biomarkers of minimal inhibitory concentration (MIC) determination and treatment efficacy to make it possible to adapt and even to shorten anti-TB treatments in a safe way to facilitate patient compliance, avoid toxicity while avoiding the development of resistance.

Aim of the project:
The PhD project aims to develop and validate breakthrough fast AST approach providing results on treatment efficacy in less than 24 hours. The first approach that we recently developed and adapted for MTB, called nanomotion-AST, is based on nanomechanical sensors (2,3) and allows to provide a result on the efficacy of a drug in less than 24h; the first approach will explore the correlation between nanomotion-AST, whole genome sequencing and other molecular AST and conventional culture based phenotypic AST. The second approach is to explore the strength of the correlation between i) drug diffusion within MTB cells and MTB drug metabolism rate measured by liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS) and ii) MIC.

Significance and expected results:
This project is part of a global effort aiming at optimizing the clinical use of the currently available anti TB drugs, which should i) improve the cure rates of tuberculosis, ii) permit to adapt drug regimen according to patient for difficult to treat infections, iii) drug toxicity in patients exposed unnecessarily to excessive drug plasma concentrations, iv) shorten regimens for multi-drug-resistant tuberculosis and v) permit to assess the efficacy of new drugs active against *M. tuberculosis*.
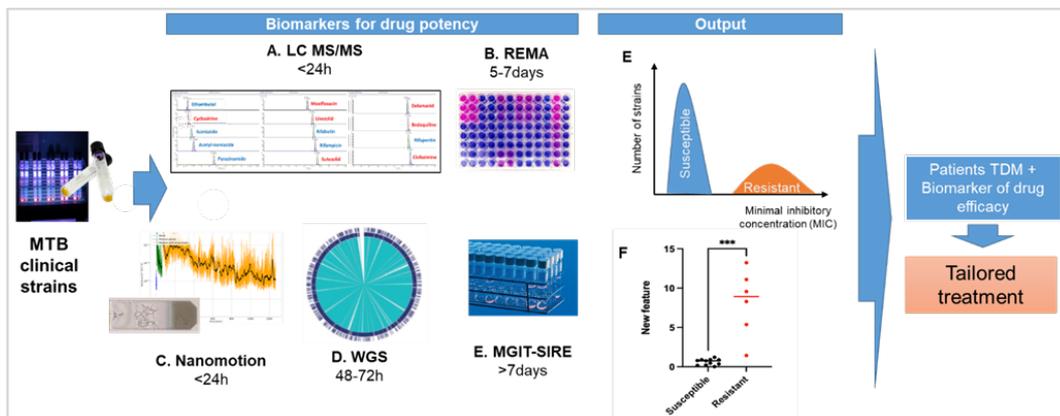
Presentation of the study. Clinical strains of *M. tuberculosis* (MTB) will be exposed to the set of anti-TB drugs presented on the bottom. The activity of the drugs will be assessed using a panel panel of well established antibiotic susceptibility tests and new biomarkers. **A)** LC MS/MS Multiplex liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS) that monitor drug penetrance and degradation **B)** Resazurin microtiter-plate assay (REMA), a culture based microdilution methods providing minimal inhibitory concentrations (MICs) **C)** Nanomotion, a new phenotypic technique, capable of detecting bacterial movements **D)** Whole genome sequencing (WGS) that detects genetic markers of antibiotic resistance and E) MGIT-SIRE Phenotypic detection of resistance.

References:
(1) Murray CJL, Ikuta KS, Sharara F, Swetschinski L, Robles Aguilar G, Gray A, et al. Global burden of bacterial antimicrobial resistance in 2019: A systematic analysis. *The Lancet* 2022; **399**: 629-55.
(2) Stupar P, Opota O, Longo G, Prod'hom G, Dietler G, Greub G, et al. Nanomechanical sensor applied to blood culture pellets: A fast approach to determine the antibiotic susceptibility against agents of bloodstream infections. *Clin Microbiol Infect* 2017; **23**: 400-5.
(3) Longo G, Alonso-Sarduy L, Rio LM, Bizzini A, Trampuz A, Notz J, et al. Rapid detection of bacterial resistance to antibiotics using afm cantilevers as nanomechanical sensors. *Nat Nanotechnol* 2013; **8**: 522-6.