IITB-Monash Research Academy

Indian Institute of Technology Bombay | MONASH University

An Indian-Australian research partnership

Project Title:	**Characterizing membrane mimetic biosensors: Towards cell free antibiotic drug screening**	
Project Number	**IMURA0784**	
Monash Main Supervisor (Name, Email Id, Phone)	Professor Mibel Aguilar, mibel.aguilar@monash.edu	*Full name, Email*
Monash Co-supervisor(s) (Name, Email Id, Phone)	Dr. John Lee, john.lee@monash.edu, Senior Post Doc	
Monash Head of Dept/Centre (Name,Email)	Prof. Roger Daly, roger.Daly@monash.edu	*Full name, email*
Monash Department:	Department of Biochemistry & Molecular Biology	
Monash ADRT (Name,Email)		*Full name, email*
IITB Main Supervisor (Name, Email Id, Phone)	Prof. Shobhna Kapoor, shobhnakapoor@chem.iitb.ac.in,	*Full name, Email*
IITB Co-supervisor(s) (Name, Email Id, Phone)	NA	*Full name, Email*
IITB Head of Dept (Name, Email, Phone)	Prof. K.P. Kaliappan, kpk@chem.iitb.ac.in,	*Full name, email*
IITB Department:	Department of Chemistry	

Research Clusters:

Highlight which of the Academy's CLUSTERS this project will address?
*(Please nominate JUST **one.** For more information, see www.iitbmonash.org)*

1	*Material Science/Engineering (including Nano, Metallurgy)*
2	*Energy, Green Chem, Chemistry, Catalysis, Reaction Eng*
3	*Math, CFD, Modelling, Manufacturing*
4	*CSE, IT, Optimisation, Data, Sensors, Systems, Signal Processing, Control*
5	*Earth Sciences and Civil Engineering (Geo, Water, Climate)*
6	*Bio, Stem Cells, Bio Chem, Pharma, Food*
7	*Semi-Conductors, Optics, Photonics, Networks, Telecomm, Power Eng*
8	*HSS, Design, Management*

Research Themes:

Highlight which of the Academy's Theme(s) this project will address?
(Feel free to nominate more than one. For more information, see www.iitbmonash.org)

1	*Advanced computational engineering, simulation and manufacture*
2	*Infrastructure Engineering*
3	*Clean Energy*
4	*Water*
5	Nanotechnology
6	*Biotechnology and Stem Cell Research*
7	*Humanities and social sciences*
8	Design

The research problem

The quest to discover novel antibacterial drugs targeting tuberculosis has seen major paradigm shifts over the last 80 years. To this view, drug-membrane interactions are becoming inevitable towards a complete molecular level understanding of drug's pharmacological properties like transport, toxicity, accumulation and hence efficacy. This is further underscored by the unique membrane components in mycobacterial cell envelope, i.e., the outer membrane (OM) lipids—strikingly distinct from the host eukaryotic membrane lipids—that are presumed to play critical roles in transport and mode of action of anti-tubercular drugs. Thus, despite the enormous therapeutic significance of mycobacterial outer cell membrane as the first line of defence, specific involvement of mycobacterial OM in drug interaction is lacking, largely attributed to the non-availability of appropriate membrane systems capturing the full spectrum of complex mycobacterial lipids. This severely limits our view of gaining significant insights for the unmet need of designing new and more efficient anti-Tb drugs governed by their specific membrane interaction profiles.

Project aims

Thus, to gain insight into the molecular basis underlying the functional organization of the lipid OM membrane, we will carry out a detailed study of the properties of GUVs made from OM lipids or from synthetic lipid mixtures regarding membrane order and domain miscibility, prior to investigating drug-membrane interactions. This project aims to develop molecularly complete membrane scaffolds (i.e., vesicles) highly specific to mycobacterial species and demonstrate them as efficient "proof-of-concept platforms" for investigating biomolecular drug-membrane interactions. Specifically, we will extract OM lipids from mycobacteria and profile characteristic lipids. We will quantify lipid packing in the membrane GUVs by applying two-photon Laurdan fluorescence microscopy, and analyse the topographic features of the monolayers by atomic force microscopy. We will compare first these complex membranes with compositionally simple models, identifying individual species that are critical for the maintenance of high order and lateral demixing. Subsequently, to assess the functional relevance of lipid order and miscibility, we will characterize the perturbing effects exerted by membrane-active agents and known drugs on membranes made from bacteria-derived lipids, correlating with their bactericidal activity.

We foresee to develop a versatile model system in convenient planer geometry compatible with various bio-analytical methods and high-throughput microfluidic devices. In this direction, attempts would be made to generate solid supported bilayers (SLB) derived from mycobacterial OM lipid vesicles coupled with surface sensing techniques. Dual Polarization Interferometry (DPI) would be used to screen key parameters for successful formation of lipid bilayers. Finally, using DPI real-time detection of membrane conformational changes, upon interaction with antibiotic drugs/membrane-active agents would allow for quick screening protocols for testing the efficacy of anti-TB drugs relative their effect on eukaryotic membrane mimics prepared in the similar fashion.

Expected outcomes

Highlight the expected outcomes of the project

1. Generation of novel OM mycobacterial membrane mimics
2. Biophysical characterization of bilayer order and domains organization using fluorescence microscopy, atomic force microscopy and Dual Polarization Interferometry.
3. Development of molecularly complete lipid platforms in appropriate geometries compatible with surface sensitive bio-analytical methods and high-throughput microfluidic devices.
4. Facilitate effective and quick screening of anti-Tb drugs for unspecific membrane toxicity, obviating the complexity of *in vivo* systems.

How will the project address the Goals of the above Themes?

Compared to simple membrane systems made form a few synthetic lipids, biomimetic liposomal model systems comprising of mycobacterial lipids represent an improved novel model of natural mycobacterial cell membrane, in particular the OM for screening for drug-membrane interactions in tuberculosis research. In depth characterization of mycobacterial–specific membranes and detailed understanding of biophysical interaction between them and anti-tubercular drugs would permit leveraging the relationship between affinity and specificity; wherein the gained findings could be applied for rational design of novel compounds which may be weak inhibitors of the target but may concentrate specifically in the mycobacterial membranes driving inhibition. In addition, capturing the whole spectrum of interactions between the drug and lipid membranes rendered by incorporating the natural OM mycobacterial lipid moieties would provide an insightful toolbox for shaping the pharmacological effectiveness of already existing anti-TB drug molecules.

Capabilities and Degrees Required

Masters in Chemistry (Physical or Organic), Physics, or Biophysics. Basic knowledge of Mass spectroscopy (MALTI-TOF, LCMS, ESI-MS), chromatographic separation methods. Basic knowledge and hands-on-experience in microscopy and spectroscopy. Having experience in handling bacterial and mammalian culture would be advantageous. Proficient in written and spoken English.

Potential Collaborators

NA

Select up to **(4)** keywords from the Academy's approved keyword list **(available at http://www.iitbmonash.org/becoming-a-research-supervisor/)** relating to this project to make it easier for the students to apply.

BioScience, Drug-Membrane interactions, Novel Functional Materials, Nanotechnology,