

Immunity Pogil Continued...

- Model 3 and 4 40 minutes
- Allergy Video w/Questions

Pogil Immune In Modela Pathogen

Mihaela Oprea



Pogil Immune In Model Pathogen:

Unveiling Pathogen Interactions with Immune Cells and Model Biomembranes Mengchi Jiao, 2024

The overall goal of my dissertation research is to unveil the pathogen immune cell interactions through the endocytosis pathway by fluorescence microscopy enabled biochemical sensors and biophysical assays. Endocytosis is a critical cellular function that clears foreign materials and protects the host from pathogen attack. However, as can be brought into the intracellular environment of the host cell by endocytosis, pathogens have developed a variety of mechanisms to hijack the endosome maturation so that they can somehow manage to escape, leading to access to replication machinery and successful infection. Therefore, understanding how pathogens escape from endosomes and bypass the innate immune system is critical to preventing infection and developing medical therapeutics. My graduate research provided insights into addressing three fundamental questions regarding pathogen hijacking the endocytosis pathway:

1. How does the anisotropic ligand presentation on pathogens modulate the innate immune response of the host cell during phagosome maturation?
2. How does the interaction between non-enveloped viruses and the lipid membrane result in the virus endosomal escape and host infection?
3. What are the specific roles of capsid protein released peptides and lipids during the dynamic virus membrane interactions?

The anisotropic arrangement of cell wall components is ubiquitous among pathogens, but how this functional asymmetry affects interactions between microbes and host immune cells is not known. In the first part of my thesis, we asked how ligand anisotropy on pathogens modulates phagosome maturation, the process used by host immune cells to degrade internalized microbes. Building on our previous research, we developed two-faced Moon particles as model pathogens that not only display ligands on solely one hemisphere but also simultaneously function as fluorogenic sensors for probing biochemical reactions inside phagosomes during maturation. We show that the anisotropic presentation of ligands on particles delays the start of acidification and proteolysis in phagosomes but does not affect their degradative capacity. Importantly, our work suggests that functional asymmetry provides pathogens with a longer time in the neutral phagosomal environment, making it easier for the pathogens to escape. The second part of my thesis focuses on virus-host membrane interactions. Non-enveloped viruses without a lipid coating enter host cells primarily through the endocytosis pathway. The virus escape requires rearrangement of the viral capsids and conformational change of capsid proteins. However, how the conformational changes enable the endosomal membrane penetration remains unknown. To tackle this question, we have focused on using reovirus as a model to understand how non-enveloped viruses penetrate across the host membrane. Reoviruses virions are digested by extracellular and endosomal proteases to generate entry intermediate called the infectious subviral particles (ISVPs). We sought to investigate the dynamic interactions between ISVPs and lipid membranes by using the giant unilamellar vesicle (GUV) model system. Our work reveals a previously undocumented role of virus particles in inducing local membrane perturbation at the site of dynamic interaction. Specifically, we demonstrate the formation of relatively large pores that approach the size of

particles that are delivered across the membrane. Importantly, the large pore formation requires the presence of ISVPs and the virus-induced membrane rupture depends on cholesterol content in the membrane. We further investigated the three-way interactions of viral proteins, released peptides, and lipid membranes during the membrane entry. We introduced the planar supported lipid bilayer as a model membrane system and applied the single virus tracking technique to dissect the interactions between lipids and viruses. By trajectory analysis, we unveiled the dual roles of the u1N peptides, which not only drove the diffusion of ISVPs on the membrane but also served as receptors to recruit and confine new ISVPs. In addition, we found u1 protein also affected the ISVP-lipid interaction. From our results, we established the burnt bridge mechanism for the initiation of infection, consisting of virus attachment and endosomal escape. Our studies highlighted a well-orchestrated and coherent self-propagating mechanism underlying reovirus membrane interaction, which is critical to understanding the infection of other lethal non-enveloped viruses and the development of anti-viral strategies. In summary, my doctorate research gave insights into the mechanisms by which pathogens hijack the endocytosis pathway to infect the host cell. Learning the infection mechanism will enable us to understand more about complex diseases, facilitating the development of anti-pathogen therapies and promoting public health.

The Mononuclear Phagocyte System in Infectious Disease

Geanncarlo Lugo-Villarino, Céline Cougoule, Etienne Meunier, Yoann Rombouts, Christel Vérollet, Luciana Balboa, 2019-10-04

The Mononuclear Phagocyte System (MPS) of vertebrates is composed of monocytes, macrophages, and dendritic cells. Together, they form part of the first line of immune defense against a variety of pathogens: bacteria, fungi, parasites, and viruses, and thus play an important role in maintaining organism homeostasis. The mode of transmission, type of replication, and mechanism of disease causing differ significantly for each pathogen, eliciting a unique immune response in the host. Within this context, the MPS acts as both the sentinel and tailor of the immune system. As sentinels, MPS cells are found in blood and within tissues throughout the body to patrol against pathogenic insult. The strategy to detect microbial non-self relies on MPS to recognize conserved microbial products known as pathogen-associated molecular patterns (PAMPs). PAMP recognition represents a checkpoint in the response to pathogens and relies on conserved pattern recognition receptors (PRRs). Upon PRR engagement, MPS mounts a cell-autonomous attack that includes the internalization and compartmentalization of intracellular pathogens into toxic compartments that promote destruction. In parallel, MPS cells launch an inflammatory response composed of a cellular arm and soluble factors to control extracellular pathogens. In cases when innate immunity fails to eliminate the invading microbe, MPS serves as a tailor to generate adaptive immunity for pathogen eradication and generation of memory cells, thus ensuring enhanced protection against re-infection. Indeed, MPS cell functions comprise the capture process, migration, and delivery of antigenic information to lymphoid organs, where type 1 immunity is tailored against intracellular microbes and type 2 immunity against extracellular pathogens. However, this potent adaptive immunity is also a double-edged sword that can cause aberrant inflammatory disorders like autoimmunity or chronic inflammation. For this reason, MPS also

tailors tolerance immunity against unwanted inflammation Successful clearance of the microbe results in its destruction and proper collection of debris resolution of inflammation and tissue healing for which MPS is essential Reciprocally as part of the evolutionary process taking place in all organisms microbes evolved strategies to circumvent the actions bestowed by MPS cells Multiple pathogens modulate the differentiation maturation and activation programs of the MPS as an efficient strategy to avoid a dedicated immune response Among the most common evasion strategies are the subversion of phagocytosis inhibition of PRR mediated immunity resistance to intracellular killing by reactive oxygen and nitrogen species restriction of phagosome maturation modulation of cellular metabolism and nutrient acquisition regulation of cell death and autophagy and modulation of pro inflammatory responses and hijacking of tolerance mechanisms among others The tenet of this eBook is that a better understanding of MPS in infection will yield insights for development of therapeutics to enhance antimicrobial processes or dampen detrimental inflammation for the host s benefit We believe that contributions to this topic will serve as a platform for discussion and debate about relevant issues and themes in this field Our aim is to bring expert junior and senior scientists to address recent progress highlight critical knowledge gaps foment scientific exchange and establish conceptual frameworks for future MPS investigation in the context of infectious disease

Methods, Models, and Machine Learning Approaches for Understanding Pathogen-specific Humoral Immunity Tomer Zohar, 2022 The humoral immune response is comprised of vast libraries of polyclonal antibodies capable of recognizing a myriad of targets and directing a spectrum of innate immune functions The complex heterogeneity in antibody profiles across both populations and diseases makes defining mechanisms of protection difficult Understanding these mechanisms and the factors that influence them is essential to defining immunity and helps inform the design of vaccines and therapeutics Thus in this thesis I describe five studies that present the development of experimental and computational methods and machine learning approaches for investigating the mechanisms dynamics and determinants of pathogen specific humoral immunity The first study introduces an assay for probing antigen specific antibody mediated primary monocyte phagocytosis that is capable of capturing subsequent downstream functions The second study describes a machine learning approach for defining the correlates of upper and lower respiratory protection against RSV and methods for evaluating vaccine designs The third study uses machine learning methods to uncover signatures of humoral protection against SARS CoV 2 The fourth study presents a method for longitudinally modelling humoral immunity that was used to investigate the temporal dynamics of antibody features across individuals with varying COVID 19 severity Finally the last study describes a genome wide association screen of pathogen specific polyclonal antibody characteristics and functions that was then validated with transcriptomics data Ultimately the methods described in this thesis present new approaches for investigating underlying phenomena related to pathogen specific humoral immunity

Mathematical Modeling of the Immune System in Homeostasis, Infection and Disease Gennady Bocharov, Burkhard Ludewig, Andreas Meyerhans, Vitaly Volpert, 2020-02-24 The immune system

provides the host organism with defense mechanisms against invading pathogens and tumor development and it plays an active role in tissue and organ regeneration. Deviations from the normal physiological functioning of the immune system can lead to the development of diseases with various pathologies including autoimmune diseases and cancer. Modern research in immunology is characterized by an unprecedented level of detail that has progressed towards viewing the immune system as numerous components that function together as a whole network. Currently we are facing significant difficulties in analyzing the data being generated from high throughput technologies for understanding immune system dynamics and functions, a problem known as the curse of dimensionality. As the mainstream research in mathematical immunology is based on low resolution models, a fundamental question is how complex the mathematical models should be. To respond to this challenging issue, we advocate a hypothesis driven approach to formulate and apply available mathematical modelling technologies for understanding the complexity of the immune system. Moreover, pure empirical analyses of immune system behavior and the system's response to external perturbations can only produce a static description of the individual components of the immune system and the interactions between them. Shifting our view of the immune system from a static schematic perception to a dynamic multi level system is a daunting task. It requires the development of appropriate mathematical methodologies for the holistic and quantitative analysis of multi level molecular and cellular networks. Their coordinated behavior is dynamically controlled via distributed feedback and feedforward mechanisms which altogether orchestrate immune system functions. The molecular regulatory loops inherent to the immune system that mediate cellular behaviors, e.g. exhaustion, suppression, activation and tuning, can be analyzed using mathematical categories such as multi stability, switches, ultra sensitivity, distributed system, graph dynamics or hierarchical control.

GB is supported by the Russian Science Foundation grant 18 11 00171. AM is also supported by grants from the Spanish Ministry of Economy, Industry and Competitiveness and FEDER grant no SAF2016 75505 R, the Mar a de Maeztu Programme for Units of Excellence in R D MDM 2014 0370 and the Russian Science Foundation grant 18 11 00171.

Building a Robust Immune Response Moria Cairns Chambers, 2012. How does your immune system prepare for all of the potential pathogens it might face over the course of a lifetime? Trade offs occur when you invest in responses that are beneficial when fighting one pathogen but are actively detrimental for fighting another. Due to the diversity of pathogens, immunity is potentially rife with this type of antagonism. To appreciate the full scope of potential trade offs, we must think about all of the possible immune responses a host can bring to bear on a pathogen. I propose that an immune response is any response to infection that influences the outcome of that infection. This includes processes that affect either resistance (the ability to clear pathogen) or tolerance (the ability to cope with pathology induced by infection). This broad definition of immunology will bring research of classically non immune physiologies (metabolism, circadian rhythm and mating) into the immune arena. Throughout this thesis, I will explore using *Drosophila melanogaster* as a model, a number of different trade offs in immunity from the antagonism inherent in a resistance response to the benefits and consequences of energy.

expenditure during infection First I demonstrate that there is an inherent trade off due to investment in phagocytosis when flies encounter two different infections *Listeria monocytogenes* and *Streptococcus pneumoniae* *L. monocytogenes* is a facultative intracellular pathogen that harnesses the additional phagocytosis increasing entry into a desirable niche *S. pneumoniae* is an extracellular pathogen that is better cleared by increased phagocytosis I discovered the trade off by comparing and contrasting the phenotypes caused by mutants in two *Drosophila* immunity genes *ets21c* a putative transcription factor and *wntD* a negative regulator of immunity Further exploration of the immune phenotypes of the *ets21c* mutant revealed that these mutants have a range of phenotypes during infection suggesting a complex picture *Ets21c* affects both tolerance and resistance to infection and the class of phenotype observed in *ets21c* mutants cannot be predicted solely by the intracellular versus extracellular nature of the infecting pathogen *Ets21c* mutants also have a strikingly altered basal metabolic state resembling sick wild type flies and have a muted change in transcript levels in response to infection This thesis also deepens our understanding of developmental immune pleiotropy in the *wntD* pathway Pleiotropy itself causes trade offs for while pleiotropy promotes efficiency in the genome it also restricts the ability to evolve *WntD* a negative regulator of the toll pathway impacts both immunity and dorsal ventral development Recently work with the developmental phenotypes led to the discovery of components in the *wntD* signaling pathway I show that these developmental mediators are also involved in immunity and impact survival during *L. monocytogenes* infection *L. monocytogenes* infection causes infection induced anorexia in *Drosophila* and this thesis shows that infection with *L. monocytogenes* affects a number of metabolic pathways at both the transcript and metabolite level This metabolic and transcriptome data generated a number of more specific and mechanistic hypotheses concerning additional potential trade offs First energy stores metabolic intermediates and transcripts for beta oxidation and glycolysis decrease during infection This reduction of available energy can both negatively impact the host when it runs out of energy for essential processes and positively impact the host by restricting the nutrients available to the pathogen By infecting mutants with either initially low energy stores or an inability to access stores we show that access to energy stores is important to the host during infection although the flip side of this trade off remains untested A second potential trade off seen through our metabolomics are changes in the level of an anti oxidant uric acid The flies enzymatically reduce levels of uric acid during *L. monocytogenes* infection A reduction in an anti oxidant should cause the reactive oxygen species to have additional potency This would be helpful in combating the bacterial load but potentially detrimental due to an increase of damage to the host itself However mutants in *uricase* which fail to lower uric acid levels during infection do not have such easily explainable phenotypes potentially due to compensation through other anti oxidants While not conclusive these data suggest that the flies regulate their anti oxidant levels during infection and that this complexly affects immunity To address the dilemma of how to build a robust immune response I contend that one must consider many different variables diversity of pathogens genetic efficiency and the energetic cost Years of evolution have

honed the immune responses with many potential solutions I found that *Drosophila* immune systems are likely constrained by a variety of tradeoffs antagonistic abilities of resistance responses metabolic links with immunity and developmental immune pleiotropy We still need to better understand how these tradeoffs are regulated and their downstream implications Understanding these antagonistic relationships will help us manipulate them to develop more effective treatment as we can tailor medicine to the individual pathogen and the individual person's physiology

How the Immune System Learns from Infections Hongda Jiang, 2022 The immune system is a complex system of cells and molecules that work cooperatively to protect us against pathogenic organisms It can perform complicated tasks such as pattern recognition learning and memory all of which require dynamical coordination among a large number of components across multiple scales Nevertheless the multitude of different components makes it challenging to unveil the mechanistic principles that give rise to these remarkable functions My thesis focuses on how our immune system learns from infections and improves specificity of pathogens recognition on the fly This process is known as affinity maturation where the affinity of B cell receptor improves through Darwinian evolution Although recent progresses in experiments revealed many details what remains is a first principle and quantitative understanding of how different elements come together to achieve the goal Using statistical physics tools and computational modeling I study various aspects of the maturation process including molecular interactions information extraction and evolutionary dynamics To understand how B cells with different affinities are discriminated during affinity maturation we investigate the process of antigen extraction where B cells use cytoskeleton forces to extract antigen molecules from other presenting cell surface We show this process allows a B cell to infer its receptor affinity by measuring the number of extracted antigens Our model highlights the regulatory role of mechanical force Application of a constant force with proper magnitude can enhance discrimination fidelity and usage of a dynamical force that introduces negative feedback can improve discrimination robustness with respect to fluctuations in antigen concentration To illustrate how molecular interactions influence cellular evolution we couple the physical theory of antigen extraction to a minimal model of affinity maturation and simulate ensembles of cell populations under different conditions The multiscale model predicts that the affinity ceiling stems from the physical limit of antigen tether strength and identifies strategies to alleviate the constraint Lastly we present a study on the long term coevolution between evolving pathogen and adaptive immune response Our work reveals that the asymmetric reaction range between immunogenicity the ability of pathogens to induce an immune response and antigenicity the ability of pathogens to interact with antibodies is critical in determining the dynamics of coevolution

Host-pathogen Interaction Using the Whole Blood Models Sravya Sreekantapuram, 2022* In order to investigate the complex interactions between the immune cells and pathogens the aim of this thesis was to establish an ex vivo whole blood model using murine and avian blood and to investigate how the model pathogens *Candida albicans* *Escherichia coli* *Staphylococcus aureus* *Salmonella Enteritidis* and *Salmonella Gallinarum* interact with various host components in this

environment In the first part of our study the established whole blood model was used to understand the immune responses in the peripheral blood of two chicken lines differing in egg laying performance to infection with either *C albicans*, *S aureus* or *E coli* Our results demonstrated chicken line and pathogen dependent differences in pathogen survival immune cells viability and their interactions with the pathogens Comparing different avian leukocyte subsets the bacterial pathogens were found to be most associated with monocytes followed by the granulocytes In contrast *C albicans* more frequently interacted with granulocytes and at a lower rate with monocytes *C albicans* was observed to have stronger impact on immune cell viability in chicken than to the bacterial species These studies were furthermore extended to *Salmonella enterica* sp which are important and widely studied pathogens in chickens The second part of the study provided insights into interactions between murine blood and *C albicans*, *E coli* and *S aureus* Our results indicated relatively low pathogen clearance and pathogen dependent differences regarding rates of association with immune cells Using a filament deficient *C albicans* mutant it was shown that increased filamentation does not explain the reduced killing Likewise the lower absolute number of neutrophils in murine blood could not fully explain higher fungal survival Lack of prior exposure to pathogens and absence of adaptive responses such as antibodies appear to contribute to low pathogen clearance

Evolution of Immune System Against Diverse Antigens Jiming Sheng, 2021 The immune system evolves across the host's lifetime to protect against the wide array of threats in nature While the immune system is capable of evolving and adapting to a single antigen Ag it becomes a challenging task to defend against diverse antigenic targets including mutants of the same pathogen or a wide spectrum of pathogen species First the rapid intra host diversification of highly mutable pathogens such as human immunodeficiency virus HIV or hepatitis C virus HCV creates a coevolutionary arms race with the immune system As a result viruses persist into a chronic infection in most subjects and are only cleared in rare cases In addition The vaccination trials so far to elicit broadly neutralizing antibodies bnAbs against highly mutable viruses have met with failure Second the immune system has to allocate its finite amount of adaptive immune cells against the wide spectrum of pathogens in the environment As memory cells accumulate from each pathogen encounter the host's immune repertoire gradually becomes skewed more adaptive immune cells are dedicated to the frequent pathogens while fewer are reserved for the rare pathogens The skewed repertoire in the elderly has been correlated with immune risk phenotype and a chronic inflammatory response even in the absence of pathogens but whether there is a mechanistic connection remains unknown My dissertation aims to address the following questions regarding the evolution of the immune system against diverse antigenic targets 1 What are the mechanisms and deciding factors behind the distinct coevolutionary outcomes observed in different subjects 2 What makes a viable vaccine design strategy to guide immune system evolution towards bnAbs 3 What are the side effects of a skewed immune repertoire as a result of adapting to different pathogens encountered during host's lifetime Also how can human intervention alleviate these side effects My dissertation shows that 1 different coevolutionary outcomes are decided by the

timing and efficacy of successive narrow and broad antibody Ab responses which in turn are determined by the conservation level and initial diversity of Ag 2 A viable vaccine strategy to elicit bnAbs should balance suppression of strain specific B cells and preserving cross reactive B cells The corresponding optimal selection strength should increase in time as driven by the evolution of B cell cross reactivity 3 Mediated by adaptive innate feedback repeated pathogen encounters during host lifetime and resulting memory inflation may trigger a fragility in which any encounter with a novel pathogen will cause the system to irreversibly switch from health to chronic inflammation CI In addition the onset of CI strongly depends on the history of encountered pathogens the timing of onset can be delayed drastically when the same set of infections is encountered in a specific order

Innate Immune Cell Recruitment and Host Defense in Response to Intracellular Bacterial Infection William Jerome Bunce Vincent,2017 The interactions between invading pathogens and host immune cells that respond to infection is a long and involved relationship In particular a class of pathogens have evolved to not only evade clearance by the cells of the innate immune system but do so by manipulation of the host cytosol This proximity has led to the intricate co evolution of clearance and evasion mechanisms of the host and pathogen respectively How these interactions are carried out in the cellular immune response is the subject of this dissertation In Chapter 2 I build a localized infection model for one such pathogen *Listeria monocytogenes* using the transparent zebrafish larval host Using this model I demonstrate the functional presence of a conserved innate immune sensing pathway the inflammasome that surveils the cytosol for the presence of bacterial flagellin Upon optimal inflammasome activation the host is protected from lethal infection and I show that macrophages are the crucial host cell population in this defense In Chapter 3 I characterize *L. monocytogenes* utilization of the host actin network demonstrating live imaging of this interaction for the first time in vivo Once *L. monocytogenes* is localized within the cytosol it slows down macrophage motility although in an actin independent manner In Chapter 4 I develop a model to study the recruitment and resolution of leukocytes during concurrent wounding and infection This model demonstrates both beneficial and detrimental leukocyte responses and provides evidence that the immune responses to bacterial infection and wounding are separable programs during early stages of immune response Taken together I have shown that macrophages are a crucial part of host defense downstream of inflammasome activation that macrophages activate other cell autonomous responses during their interaction with intracellular pathogens and that macrophage neutrophil interactions can be critical in determining a beneficial or detrimental response to infection These findings highlight the intricate and close evolution of host immune cells and intracellular pathogens as well as the critical nature of inflammatory balance during immune responses

How the Immune System Generates Diversity Mihaela Oprea,1998

Optimal Design Principles in Pathogen Replication and Immune Response Patrick Binder,2022*

Orchestration of an Immune Response to Respiratory Pathogens Andrea Sant,Steven Varga,2019 This eBook is a collection of articles from a Frontiers Research Topic Frontiers Research Topics are very popular trademarks of the Frontiers

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Pathogen sensing in innate immunity ,2010 *Microdomains in the Immune System Control Cell Adhesion and Pathogen Uptake* Alessandra Cambi,2005

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