RESEARCH STATEMENT

Yohannes Shiferaw
Department of Medicine, UCLA

I. OVERVIEW

My research work is focused on theoretical biophysics. That is, the application of tools from mathematics and physics to solve problems that are inspired by biology. Within this broad field I am particularly interested in problems that have medical relevance, especially in the field of cardiac physiology.

In recent years advances in molecular biology and optical imaging have led to a wealth of experimental data on many important biological processes. In particular, in the field of research cardiology, much is known about the molecular biology of ion channels in heart cells, the movements of intracellular ions, and the dynamics of electrical waves during a heart beat. Thus, it is now possible to address a wealth of new questions regarding important biological processes. Major open problems in the field of cardiology are:

1. What are the essential dynamical features of fibrillation (the state of turbulent electrical activity during a heart attack)?

2. What are the mechanisms, at the cellular and ion channel level, which lead to the initiation of fibrillation?

My goal is to apply tools from mathematics and physics to construct models which can be used to shed light on these processes, and which can also be used to integrate the growing wealth of related experimental data. Furthermore, recent developments in pharmacology and genetic engineering have given experimentalist unprecedented capacity to implement therapeutic strategies. It is my belief that mathematical modeling can play a crucial role to help guide the development of these therapeutic approaches.

II. PAST AND PRESENT WORK

A. Modeling the cardiac cell

A major focus of my recent work has been in the development of mathematical models of cardiac cell dynamics. My approach has been to integrate the known physiology of the cell into a computationally tractable set of ordinary differential equations, and then to apply techniques of nonlinear dynamics to get a deeper understanding of the dynamical behavior. When a cell is stimulated rapidly it is known experimentally that the action potential duration (APD), which is the period of time that the voltage across the cell membrane spends away from its rest state, alternates from beat-to-beat. This dynamical feature, referred to as *alternans*, can be understood mathematically as a period doubling bifurcation, and has been argued to promote ventricular fibrillation (sudden death heart attack). However, the underlying mechanism for this dynamical instability is not known. One possibility, which I have explored in detail, is the role of intracellular *calcium cycling*, which is the process in which calcium is released into the cell from intracellular stores in order to mediate contraction. In order to assess the role of calcium, we developed a computationally tractable model which took into account the wealth of experimental information on calcium imaging in cells, and which also exhibited quantitatively accurate dynamics at high pacing rates. By analyzing the dynamics of this model we were able to uncover the key physiological parameters that controlled the dynamical instability, and gained insight on the underlying physical mechanisms. Also, by developing a nonlinear map that described the beat-to-beat dynamics, we were able to analyze the mathematical nature of the dynamical instability.

Once the nonlinear dynamics of calcium cycling was understood we proceeded to investigate the bi-directional coupling between membrane voltage and intracellular calcium cycling. Here, the challenge was to understand the coupled dynamics of two nonlinear systems; calcium cycling and the dynamics of membrane voltage. The latter being governed by the gating kinetics of ion channels on the cell membrane. Our main finding was that the coupled dynamics could exhibit a rich dynamical behavior which was crucially dependent on the coupling between calcium and voltage. By reducing the model to a two dimensional discrete map we were able to explain several experimentally observed phenomenon. Overall, this work offers a framework that can be used to interpret a variety of experimental results

on the dynamical behavior of cardiac cells. In the future I hope to apply the model to make quantitative predictions that can be directly tested in an experimental setting.

More recently, I have been working on a Markov model of the L-type calcium current, which is based on data acquired in an electrophysiology lab here at UCLA. This work is motivated by recent experiments and simulations which suggest that the gating kinetics of the calcium current plays a critical role in the onset of fibrillation, and thus is a natural target for gene-based therapeutic strategies. We found that the kinetics of the L-type calcium current can be described by a four state Markov model, which cannot be reduced to the standard Hodgkins-Huxley formalism, where gate processes are assumed to be independent. We are now applying this model to evaluate how the detailed kinetics of the L-type calcium current influences tissue scale phenomenon such as spiral waves.

It is my belief that accurate experimentally based mathematical models can guide the development of novel gene-based therapeutic strategies. Recent advances in molecular biology have led to the development of gene transfer technologies which allow unprecedented control of cellular functions. In our lab at UCLA experimentalists have managed to target and slow down the calcium dependent inactivation kinetics of the L-type calcium current. Moreover, they have found that this intervention modifies the membrane voltage dynamics in a way that prevents the onset of fibrillation. However, simply decreasing calcium entry compromises cell contraction which negates the therapeutic advantages. Thus, I am using a mathematical modeling approach to to explore what kinds of interventions are required to prevent fibrillation while maintaining sufficient cell contraction. Ultimately we are hopeful that these studies will lead to novel therapeutic strategies which can be tested in animal hearts.

B. Pattern formation in cells and tissue

Another focus of my research is the study of subcellular pattern formation in cardiac cells. Here, we attempt to describe the spatiotemporal dynamics of subcellular calcium alternans i.e. when the calcium released into the cell can alternate from one beat to the next. This work is motivated in part by recent experiments which show that subcellular calcium alternans can be spatially discordant i.e. different regions of the cell can alternate out of phase. By coarse graining the cell into diffusively coupled units we have managed to

simulate the spatiotemporal dynamics of subcellular alternans. Our most interesting finding is the existence of a pattern forming instability that leads directly to spatially discordant calcium alternans. To analyze the simulation findings we developed a set of amplitude equations that accurately describe the spatial patterns. Using this approach we have shown that the pattern forming instability is analogous to the classic *Turing instability*, where the inhibitor and activator corresponds to voltage and calcium respectively. The physiological implications of our results are potentially very important since the coordination of cell contraction will be disrupted if this pattern formation process develops.

We have extended the above results to study the spatiotemporal dynamics of alternans at the tissue scale (cardiac tissue is modeled as a square lattice of roughly 200×200 cells). In particular, we have investigated the role of calcium cycling in the initiation and dynamics of spatially discordant alternans. An important new finding is that complex tissue scale discordant alternans can be formed when calcium cycling is the underlying mechanism for alternans. This results extend known results in the literature which have focused exclusively on alternans due to membrane voltage instabilities. We have also investigated how the underlying nonlinear dynamics at the cell level controls the spatial scales of alternans patterns at the tissue level. Our main finding is that the spatial scale of the interface separating out of phase regions depends sensitively on the nature of the dynamical instability at the cellular level.

III. FUTURE WORK AND COLLABORATIONS

My future work will be oriented along two distinct but complementary directions. First, I intend to develop data driven mathematical models that can be used to interpret and motivate experimental work in the field of cardiac physiology. It is my view that understanding the dynamics of cellular processes, and how they affect tissue scale dynamics, can help suggest novel therapeutic strategies in the treatment of cardiac arrhythmias. Thus, I intend to construct quantitative models, at both the cellular and tissue scale, that can be used to evaluate the effects of pharmacological or gene-based interventions. In this regard, I plan to strengthen my existing collaborations with experimentalists in the field, so that my modeling work will be directly relevant to ongoing and future experiments.

The second direction in my work will be to study dynamical aspects of biological processes

using tools from mathematics and physics. Here, the goal is to develop a more general qualitative description using analytical tools from nonlinear dynamics, pattern formation, and statistical mechanics. In this regard, my motivation stems from a deep fascination with biological systems, and need to describe them in simple and elegant terms.

Bellow are specific projects that I have in mind:

A. Multiscale modeling of calcium dynamics in cardiac cells

The cycling of calcium in the cardiac cell is fundamental to its function, and has been implicated as a possible cause of various heart rhythm disorders. This process begins when calcium is released from intracellular stores via the stochastic signaling between trigger ion channels on the cell membrane, and calcium gating receptors inside the cell. This signaling process occurs within thousands of sub-micron scale junctions distributed throughout the volume of the cell. These local signaling events then summate in space and time to form the global response that governs the cell behavior. The essential question that I would like to address is the following: *how does the local ion channel dynamics within cells influence tissue scale phenomenon in the whole heart?* This problem involves a vast range of length and time scales, ranging form microseconds and nanometers at the ion channel level, to seconds and centimeters at the tissue scale. It is my belief that mathematical modeling will be essential to understand this hierarchy of interacting scales.

The importance of this problem is illustrated by recent experimental findings that genetic mutations, which modify the behavior of ion channels involved in calcium cycling, can increase the likelihood of a heart attack. These findings show clearly the importance of establishing a link between dynamics at the ion channel level and potentially deadly tissue scale phenomenon. Moreover, this also suggests that genetic interventions which target specific ion channels may be developed to treat heart disorders. My goal is to develop a theoretical and computational framework in which gene therapy approaches can be developed in silico. This research work will involve the following components:

1. *Calcium dependent ion channels*:

   A crucial element of calcium cycling is the stochastic signaling between L-type calcium channels and RyR receptors. The behavior of these channels and their interaction, is

inherently a stochastic process which is not completely understood. My goal is to develop Markov models of the L-type calcium channel and the RyR receptor, that are based on channel structure and known conformational states. I will then apply these models to understand the signaling dynamics between clusters of these channels. My approach is to reduce the complicated dynamics of a many state Markov model to a simple set of differential equations that contains the essential dynamical features. Also, I would like to uncover general principles that dictate the stochastic signaling between physiologically relevant ion channels. The ultimate goal of this work is to develop a computational framework in which the dynamical consequences of a particular genetic modification, say the deletion of a particular channel state, can be evaluated.

2. *Spatiotemporal dynamics of calcium within a cell*: The junctions where calcium release occurs are spatially distributed throughout the cell. Hence, calcium released within a junction can trigger release from neighboring junctions. This spatial coupling endows the calcium system with the properties of a stochastic excitable system. I intend to explore the properties of this system by developing reaction diffusion equations for the spatiotemporal dynamics of calcium within cardiac cells. My approach is to implement the simplified Markov models of ion channels, within a model that involves spatial diffusion. I am particularly fascinated by the connection between stochastic dynamics at the ion channel level, and emergent whole cell dynamics, such as calcium wave propagation. Also, I would like to explore the spatiotemporal pattern formation of calcium release, during spatially discordant alteranns. This work will rely on, and complement, the growing experimental literature on fluorescence imaging of calcium in cardiac cells.

3. *Phenomenological cell models and tissue scale dynamics*: Based on the spatial dynamics described above, I intend to develop a phenomenological model of calcium cycling which accurately describes the macroscopic dynamics of the system. Using this phenomenological model, I would like to explore the nonlinear dynamics of voltage and calcium when a cardiac cell is rapidly stimulated. I am particularly interested in the dynamics during abnormal cell conditions, for instance, during calcium overload conditions, when the cell is close to the onset of spontaneous release. I also intend to apply this phenomenological model to investigate tissue scale consequences of complex

subcellular dynamics. Here, I will focus on pathological conditions, such as the role of calcium cycling in the initiation of triggered activity in the heart, a condition which is known to cause sudden death heart attack.

This work will be done in collaboration with Professor Alain Karma, in the department of physics at Northeastern University, and Zhillin Qu, in the department of cardiology at UCLA.

B. Calcium cycling and ventricular tachycardia

I am interested in studying the spatiotemporal dynamics of calcium in two and three dimensional cardiac tissue. In particular, I will focus on the spatiotemporal organization of calcium during spiral waves (in 2D) and scroll waves (in 3D). As a spiral wave (or scroll wave) rotates over cardiac tissue, where the cells are dynamically unstable to alternans, complex spatial patterns of calcium release should emerge. I would like to understand the organization and dynamics of these patterns. This problem is quite rich from the mathematical point of view as it combines ideas from the theory of excitable systems and nonlinear dynamics. Also, several groups, namely Emilia Entcheva at Stony Brook and Miguel Valderabano at UCLA, have began imaging voltage and calcium during spiral wave activity in two dimensional cardiac cell monolayers. Thus, I am hopeful that theoretical work in this direction can go hand in hand with experiments.

C. The calcium current and ventricular fibrillation

Recently, I have been investigating the role of the L-type calcium current in the initiation and maintenance of fibrillation. This work is motivated by our recent experimental observation (at Cedars Sinai) that fibrillation is preceded by unstable short wavelength excitations which fractionate. My hypothesis is that the high activation threshold for the calcium current allows a premature stimulation to propagate in tissue. This excitation has a wavelength roughly an order of magnitude less than a typical wave, and thus can break when it encounters even moderate tissue heterogeneity. My goal is to develop a mathematical description of this phenomenon, and to investigate its role in the initiation of fibrillation.

D. Spatial pattern formation and optical mapping experiments

I plan to continue my work on the spatiotemporal dynamics of voltage and calcium alternans. In particular the study of the spatiotemporal evolution of discordant alternans in tissue. Recently, my collaborator Dr. Hideki Hayashi at the Cedars Sinai Medical Center has managed to record optical images of discordant alternans in normal and ischemic rabbit hearts. We have found strong evidence that nodal lines, the lines which separate out of phase regions, can have both a dynamic origin, which relies only on the gating kinetics of ion currents, and can also be caused by structural heterogeneities in the heart. I am now using my existing mathematical models of spatial alternans to propose pacing protocols that help distinguish the different mechanisms for discordant alternans.

E. Pattern formation and cell development

I am fascinated by the magnificent spatial organization of cells. For instance, the cell membrane of a cardiac cell contains a periodic array of deep invaginations into the interior of the cell. I would like the understand the dynamical mechanisms that guide the development of these remarkably periodic structures. I am also curious about the mechanisms that underly the formation and maintenance of ion channel clusters on the cell membrane. My view is that these pattern formation processes in the cell are governed by a general set of physical principles that have yet to be uncovered. My goal is to develop novel mathematical models that can shed light on these remarkable biological phenomena.

F. Collaborations

An important aspect of my current and future research is my close ties with experimentalists in the field cardiology. I have collaborated extensively with electrophysiologists at UCLA on single cell measurements of ion channel kinetics, and fluorescence imaging of intracellular calcium. Also, I have collaborated extensively with a group at the Cedars Sinai Medical Center on the analysis of optical mapping of calcium and voltage in rabbit hearts. I feel strongly that my success in this field will depend on a strong relationship between theory and experiment, and I intend to strengthen these collaborations.

IV. CONCLUSION

A major challenge to understand the dynamics of complex biological systems is that all scales are intimately connected. To understand the dynamics of the heart it is necessary to study a hierarchy of interacting scales, from the ion channel level, to cell electrophysiology, and finally to tissue scale dynamics. So that a complete understanding of the system spans a wide range of spatial and temporal scales. It is my view that the application of mathematical techniques and physical principles, in conjunction with a deep knowledge of physiology, will be essential to understand these systems. I find that this research field offers a wealth of fascinating mathematical and physical problems, and also has the potential to yield powerful applications in the field of medical research.

TEACHING STATEMENT

Yohannes Shiferaw

Department of Medicine, UCLA

Much of my teaching experience was during graduate school, at the University of Pittsburgh, where I taught introductory physics to undergraduate students. I was quite successful as a teacher and was awarded the *department of physics award for excellence in teaching* in my last year. Overall, I enjoy teaching and look forward to teaching science and mathematics at the undergraduate and graduate level.

My years teaching undergraduate physics were a constant learning experience and a great challenge. As a result I feel that I have developed the necessary skills to communicate effectively with a broad range of students. In my view, to teach effectively at the undergraduate level it is crucial to understand how students perceive the material. Students have varied backgrounds and abilities and it is the responsibility of the instructor to learn and address the gaps in their knowledge and comprehension. My approach to this problem was to develop a classroom atmosphere where students felt free to ask questions, and to encourage students to communicate on a one-on-one basis outside of the classroom. In this way I felt that I was constantly in touch with the student's perspective, and could then address specific gaps in their comprehension. Also, by carefully grading homework and quizzes, I always tried to gauge the level of understanding of each student in the class. Based on this feedback I would prepare my lectures so that I could go over specific concepts that were not grasped.

I find that many undergraduate students in physics and mathematics work hard in memorizing problem solving methodology rather than trying to understand concepts. Hence, in my lectures I have always stressed the importance of concepts and physical principles. My approach is to present concepts as clearly as possible, and then to teach problem solving as a concrete application of those concepts. To demonstrate how a problem is solved I first outline the essential concepts required, and then proceed to solve the problem in such a way that the essential ideas are not hidden in the mathematical manipulations. Thus by emphasizing ideas I hope to break student's natural tendency to memorize procedure. I also find that a clear description of a physical concept always inspires students to dig deeper into the material. Students are inspired when they sense that they have learned something new,

beyond rote memorization.

As a postdoctoral researcher in the department of Physics at Northeastern University, and at the UCLA medical school, I have had the opportunity to help supervise the research of an undergraduate and two graduate students. This experience has been rewarding and a great learning experience for me. I feel that good supervision requires developing a scientific relationship with a student. This is accomplished by having regular scientific discussions about the research problem at hand. I always challenge the students I work with to explain their findings on a blackboard. In this way we develop an informal conversation on the problem, and I can track their progress while encouraging them to clearly communicate their ideas. On the more practical side I have helped these students by guiding them through the scientific papers in the literature and giving lectures on various relevant topics.

In the future I look forward to teaching at the undergraduate and graduate level. I am especially excited to teach a course in theoretical biology. I feel that this will be an excellent way to teach concepts in physics, mathematics, and scientific computing. My interdisciplinary background in physics and theoretical biology makes me ideally suited for such an undertaking. In particular, I find that cardiac electrophysiology is a wonderful and compelling context in which to teach ordinary differential equations, nonlinear dynamics, and probability theory. To illustrate the kinds of topics that may be taught in an advanced undergraduate or beginning graduate course, I have attached, in the following page, an outline of a course that I would very much like to teach. I am also committed to supervising research projects at both the undergraduate and graduate level. I am confident that students will be drawn to the vast set of interesting research problems in theoretical cardiac physiology, as it involves the opportunity to apply mathematical and physical concepts to help solve problems that have potential medical applications.

Dynamical Systems Modeling of Biological Processes

Instructor: Yohannes Shiferaw

Course description: An examination of the art of making and evaluating mathematical models in biology, and of the dynamical principles inherent in biological systems. The course will develop the idea of differential equations as models for dynamics, and the use of qualitative dynamics and computer simulation as tools for understanding how complex systems behave.

Textbook

Keener, J. and Sneyd, J., Mathematical Physiology. Springer, 1998.

Supplemental reading

Murray, J.D., Mathematical Biology I: An Introduction 3rd Edition, Springer 2002.

Hoopensteadt, F.C. and Peskin, C.S., Modeling and Simulation in Medicine and the Life Sciences. Springer, 2002.

Topics

1. Mathematical foundations of dynamical systems theory: state spaces, vector fields, ordinary differential equations, linear algebra.

2. Computer simulation. introduction to the idea of numerical integration: using simulations to understand qualitative and quantitative properties of dynamical systems. Elementary examples of dynamical systems: mechanics (springs and pendulums), chemical processes. The concept of equilibrium. Oscillatory phenomenon.

3. Cell dynamics. Cellular Electrophysiology. Review of mathematical aspects of electrical circuit theory. Ionic mechanisms in the cell. The Hodgkin-Huxley equations and their simulation. Extensions of Hodgkin-Huxley theory to cardiac and brain cells. Calcium dynamics in cardiac cells.

4. Disease dynamics. differential equations representing the spread of disease in a population. Evaluating therapies and other interventions through modeling.

5. Spatial systems. Spatially-distributed systems modeled by partial differential equations and lattices of ordinary differential equations. Nonlinear wave propagation. Calcium wave propagation. Stability analysis of spatial systems. Pattern formation in development. The Turing instability.

6. Chaos. The idea of simple systems displaying complex dynamics. Elementary mathematical and physical systems displaying chaos. Chaos in physiology: the cases of cardiac arrhythmias and neurologic pathologies.