Mathematics of the Cell: Integrating Genes, Biochemistry and Mechanics
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1 Overview of the Field

1.1 Introduction

Biology is being touted by many as THE science of the 21st century, much as physics was at the forefront of science in the 20th century. Over the last two decades, mathematical modeling, analysis and computation have played an important in this transition. Mathematical biology is barely one hundred years old, and for a long time its focus was the deep mathematical analysis of equations inspired by biology, much like the landmark 1952 paper of Alan Turing that proposed a biologically abstract model for pattern formation during tissue development. Coincidentally, in the same year, Hodgkin and Huxley developed a biologically concrete model to understand the electrical activity in nerve cells. In the last two decades, the number of mathematically deep and biologically realistic studies has undergone explosive growth. The impact of mathematics on biology, physiology and medicine is increasing, while biologically-relevant mathematics is becoming more diverse and sophisticated.

Scientific and technological changes are accelerating, and it is important to foresee paradigmatic shifts in science and prepare in advance. Many scientists have started to realize that individual biological processes can no longer be studied in isolation but need to be considered as integrated events. Thus, the next shift is the rise of *Integrative Biology*, loosely defined as multidisciplinary research across all levels of biological organization, from molecules to the biosphere. We focus on the cell level of biological organization because cells are the smallest autonomous units of life and occupy the midpoint between the molecular and macroscopic scales. In order to understand how living systems are built and function, we need to understand the mathematical principles that underlie cellular organization and function. It is in the cell where we will first understand the basic processes of life at the molecular level in a physiological context. The cell provides the natural coordinate system in space and time onto which we have to map and integrate genomic, biochemical and mechanical information about the molecular networks that make up living systems.

Mathematical cell biology will be an integrative hub of much of modern quantitative biological research. Mathematics will provide the structure, abstraction and language to integrate across scales. Thus, future Mathematical cell biology will require bringing together scientists and mathematicians with different areas of expertise able to use a diversity of techniques and work at the interfaces of disciplines. It is essential to

make mathematicians a part of the process of biological discovery and to make biologists understand and appreciate the power of mathematics and to be able to actively participate in model development.

1.2 Mathematical Cell Biology

The exact tools that have been used previously in physical and chemical sciences may simply not be directly applicable to cell biology. This has nothing to do with vitalism, but simply with the fact that cell biology occurs on multiple and radically different scales, both in terms of time, space, and complexity. Understanding how cellular-scale behaviors arise from molecular actions is fundamentally difficult due to the large number of many different kinds of molecules all interacting in complex networks. Another difficulty is that cell biological systems consist of thousands of molecules and so are not microscopic, but they are not macroscopic either; often, fluctuations of chemical or physical quantities in the cell are comparable in magnitude to the average values of these quantities. The main difference between modeling in biology and physics stems from the inherent redundancy and heterogeneity of evolved molecular machines that have to be elucidated. This makes cell modeling very difficult but also unavoidable as new technologies produce staggering amounts of data about the spatiotemporal behavior of molecular assemblies. With increasing frequency, these data are quantitative – correlation functions, statistical regressions, and other similarly sophisticated forms – that cannot be reduced to simple qualitative statements, so mere qualitative cartoon drawing in the discussion section of a paper is not sufficient. Rather, to integrate and make sense of these data, quantitative modeling is needed as hypotheses generating machine and a natural endpoint for the experimental efforts.

2 Recent Developments and Open Problems

There are a few specific areas of cell biology where mathematical modeling is especially useful.

2.0.1 Cytoskeletal dynamics

All cells with a nucleus have dynamic polymers collectively referred to as the cytoskeleton. Two out of three primary filament types are actin filaments and microtubules. The cytoskeleton is involved in a wide range of cellular functions including cell division and cell migration. Further, the cytoskeleton provides a structural framework within the cell, allowing it to both exert and respond to extracellular stimuli. Microtubules are the most rigid structures in the cell, typically emanating from the microtubule organizing center (centrosome) next to the nucleus. Actin filaments can assume a variety of different conformations including bundles, where the filaments are arranged in a parallel or antiparallel fashion, or branched networks. These branched networks typically found at the edge of migrating cells, form a broad thin structure called a lamellipod. The cytoskeleton is involved in determining cell shape and movement. It should not be surprising that actin filaments and microtubules do not act alone. Rather, their function is regulated by a multitude of associated proteins. Some of these associated proteins help to polymerize or depolymerize the filaments, crosslink filaments into networks or bundles. Motor proteins exert forces between filaments, organelles, cell membrane and extracellular matrix. Biochemistry and cell biology have given us a wealth of data about the components and parts of this system, but many details are still missing. More than that, how the system is organized to work coherently in space and time is still an elusive question. Given the inherent complexity of this system, mathematical and physical models are playing an increasingly important role in elucidating the underlying behavior of the cytoskeleton and its role in many cellular functions. Open problems include, but are not limited to: how are cytoskeletal elements self-organize into contractile or expanding structures, what are the mechanisms behind history-dependent mechanochamical behavior of the cytoskeleton, what are the differences in prokaryotic and eukaryotic cytoskeletal dynamics,

2.0.2 Cell polarity

When the cells move directionally, or even when they are stationary but functioning in a group of other cells or preparing for division, they polarize their morphology and distributions of key molecular players inside the cells become asymmetric. This symmetry break poses a challenging problem that is a right up the alley of mathematicians: how is a combination of spatial-temporal positive and negative feedbacks coupled

to achieve the polarization? How does this process work in the presence of huge fluctuations? Ever since the fundamental Turings discovery, reaction-diffusion models of interacting chemicals were investigated and applied to explain various polarization events. However, recently, mechanical pathways were also discovered to be able to break symmetry, even in the absence of the underlying biochemistry. However, more often than not, mechanical and biochemical pathways are coupled to achieve faithful and rapid polarization. The details, and more importantly, the design principles of these couplings are not clear.

2.0.3 Cell signaling

Cell signaling is part of a complex system of communication that governs basic cellular activities and coordinates cell actions. The ability of cells to perceive and correctly respond to their microenvironment is the basis of development, tissue repair, and immunity as well as normal tissue homeostasis. Errors in cellular information processing are responsible for diseases such as cancer, autoimmunity, and diabetes. By understanding cell signaling, diseases may be treated effectively. Cell is constantly bombarded by tens of environmental stimuli and stresses. Meanwhile, there is a very finite number of 'hubs in signaling relays inside the cell. How does the cell convert the multitude of outer signals into another multitude of behaviors using a very finite signal processing toolbox? This is a great problem for a mathematician.

Traditional work in biology has focused on studying individual parts of cell signaling pathways. Mathematical biology research helps us to understand the underlying structure of cell signaling networks and how changes in these networks may affect the transmission and flow of information. Such networks are complex systems in their organization and may exhibit a number of emergent properties including bistability and ultrasensitivity. Analysis of cell signaling networks requires a combination of experimental and theoretical approaches including the development and analysis of simulations and modeling.

2.0.4 Role of aqueous medium in cell processes

Cells are filled, besides cytoskeleton, with fluid cytoplasm. The cytoplasm is a very strong electrolyte. Recently, it became clear that cells can move and generate forces by converting cytoskeletal contractions into squeezing the fluid and generating hydrostatic pressure. This pressure delaminates cell membrane from underlying actin producing bubbles. The same pressure can move nucleus as a piston inside the cell. Besides hydrostatic pressure, the osmotic pressure is a big part of the picture because ion concentrations inside and outside the cell vary a lot. Osmotic and hydrostatic pressures create flows that transport molecular cargo and help cell movements. Understanding this biofluid dynamics is impossible without solving complex problems of fluid mechanics coupled with statistical physics of polyelectrolytes.

2.0.5 Cell motility

Cells crawl by, first, pushing out the front, then it assembles tight adhesions to the surface at the leading edge and weakens such adhesions at the rear, and finally the cell develops contractions that pull up the weakly adherent rear toward the strongly adherent front, completing the motility cycle. This process is an important part of wound healing, morphogenesis and cancer, among many other biological and medical phenomena but it is the elegance of the seemingly simple, yet underlined by layers of complexity, motile cycle that inspired thousands research papers in the last four decades. The devil, of course, is in the details, and it is these increasingly meticulous and numerical molecular details of the motile machinery that require mathematical modeling. First, we have to understand the actin treadmill how dynamic network of filaments translocates in space by assembly/disassembly. Then, we have to figure out how myosin and actin self-organize to contract the cell body. Finally, we have to elucidate mathematically how tens of adhesion proteins first assemble at the leading edge gluing it to the substrate and then disassemble at the cell rear letting it go.

2.0.6 From individual cell to tissues

Developmental biology is the study of the process by which multiple cells form tissues and by which organs grow and develop. Modern developmental biology studies the genetic control of cell growth, differentiation and morphogenesis. In recent years, mathematical modelling of developmental processes has earned new respect. Not only have mathematical models been used to validate hypotheses made from experimental data,

but designing and testing these models has led to testable experimental predictions. There are now impressive cases in which mathematical models have provided fresh insight into biological systems, by suggesting, for example, how connections between local interactions among system components relate to their wider biological effects.

2.1 Objectives of the workshop

One of the most effective ways to achieve goals of collaboration and integration of theoretical and experimental approaches is to organize a small meeting at which mathematicians and biologists discuss in depth recent advances, new paradigms and trends, and plan future collaborations. The format of BIRS 5-day workshop was ideal for such a meeting. We started to pursue the goals described above 7 years ago, with the first meeting focused on Mathematical Biology of the Cell, and devoted to defining this discipline. The meeting was a big success, and was followed by the second in 2011. In the second meeting we concentrated on the narrower area of mathematics of cytoskeleton and cell motility and division, which has attracted a large number of modeling efforts. One of the best signs of its success is that a great number of collaborations were started at that meeting.

We want to emphasize that this area is not settling down into a comfortable routine. In fact, the emerging challenge we are facing is that we have to go past successful mathematical modeling of certain aspects (i.e. biochemical or mechanical) of cell behavior, and start integrating genetic, biochemical and mechanical models into multi-scale mathematical platforms that will allow for prediction of cell behavior in physiological circumstances. This poses a set of mathematical, computational and biological problems that we hoped to discuss, define and start planning to solve in 2014. And so, we continued in 2014 with having unprecedented level of interaction between experimentalists and modelers and transitioning into the systems level of modeling cell biological phenomena.

3 Presentation Highlights

A number of talks were devoted to discussions of actin dynamics and relevant self-organization phenomena. David Sept explained that highly unusual characteristics of parasites actin result from isodesmic polymerization rather than the nucleation-elongation kinetics of conventional eukaryotic actins. These findings expand the repertoire of how actin functions in cell motility and offer clues about the evolution of self-assembling, stabilized protein polymers. Thomas Pollard showed that in yeast cell division punctate protein structures arise in separate locations in the cortex and join each other around the equator of the cell by a diffuse and capture mechanism to form ”nodes”, the precursors of the cytokinetic contractile ring During mitosis nodes grow actin filaments and a search, capture, pull and release mechanism organizes the nodes into the contractile ring. Alexander Bershadsky discussed the processes of actin cytoskeleton self-organization driven by actin assembly and cross-linking and myosin II contractility. He showed how computational modeling demonstrates the evolution of the radial pattern of stress fibers into the chiral pattern and how self-organization of the actin cytoskeleton provides built-in mechanism of establishing left-right asymmetry. This mechanism may play a key role in a variety of morphogenetic processes. W. Bement brought to our attention a fascinating discovery that anaphase onset in frog and echinoderm embryos is associated with cortical excitability, manifested as waves of Rho activity and F-actin that traverse the underside of the plasma membrane. Remarkably, the excitability entails F-actin mediated Rho inhibition. He proposed that chaotic phase is explained by the development of cortical excitability which is normally restricted to a discrete portion of the cell cycle Cdk1 and that excitability provides the cell with the means to balance the conflicting needs of speed, precision and flexibility during cell fission.

Other talks addressed how actin and myosin generate force. Anders Carlsson investigates mechanisms by which polymerized actin can exert forces with the required orientation and magnitude. He showed three possible mechanisms leading to endocytic invagination: a) lateral segregation of nucleation-promoting factors into an inner core and an outer ring creates curvature-generating forces via differences in polymerization rates, b) spontaneous curvature of a coat-protein layer bends the membrane, and c) motor activity creates a contractile ring that buckles the coat protein layer. He proved that mechanism a) by itself can produce invagination, but mechanisms b) and c) cannot. However, either mechanisms b) or c) can reduce the requirements

for mechanism a). Ben Fogelson asked how an individual stress fiber behaves and, in particular, how much force it generates. By using data from cells grown on micropatterns, he was able to construct a simple 1-D model of actomyosin force production to explain a puzzling peak in force production at intermediate stress fiber lengths. Cecile Sykes talked about reconstitution of the actin cortex of cells inside liposomes, and using it as a simplified system to study endocytosis. She showed how these cortices contract in the presence of myosin motors, and how such experiments shed light of the mechanisms of cell shape changes.

A group of talks were devoted to modeling various aspects of cell movement. K. Keren posed the problem of how actin network translocates forward and illustrated that the complex reaction-diffusion problem for the polymeric and monomeric actin produces solutions that compare well with experimental measurements, and thus diffusion is able to move the monomers effectively forward. In a related talk, Garegin Papoian reported development of detailed physico-chemical, stochastic models of lamellipodia and filopodia, which are projected by eukaryotic cells during cell migration, and contain dynamically remodeling actin meshes and bundles. His simulations showed that some processes, such as binding and unbinding of capping proteins, may be dominated by rare events, where stochastic treatment of filament growth dynamics is obligatory. The talk shed light on how actin transport due to diffusion and facilitated transport such as advective flow and active transport, tunes the growth dynamics of the branched actin network. Wolfgang Loosert described how simple physical measurements of shape dynamics and motion reveal an underlying wave-like process of the cellular scaffolding that drives persistent migration and how wave-like dynamics of the scaffolding contributes to the ability of cells to recognize and follow surface nano-topography, and allows cells to couple to each other when moving in groups. A couple of talks addressed a very important problem of cells moving in 1D and 3D. David J. Odde explored the behavior of the motor-clutch model, and assessed which model parameters control the stiffness at which sensing is optimal. Using a Master Equation approach, he developed an analytical description of the model, and obtained a dimensionless number that defines the optimal substrate stiffness. He speculated that the motor-clutch model may be useful for in silico identification of combination drug targets for brain cancers, and is generally applicable to animal cell adhesion and migration in 1D, 2D, and 3D environments. Damir Khismatullin reported development of a fully three-dimensional computational model of amoeboid chemotaxis by incorporating the intracellular force field due to actin polymerization into our algorithm for passive cell deformation and adhesion, known as VECAM. The resulting model (VECAM-Active) takes into account passive mechanical properties of the cell, extracellular diffusion of chemoattractant molecules, intracellular release and diffusion of signaling molecules, intracellular active force generation, cell adhesion and physiological shear flow conditions. Using VECAM-Active, Damir has investigated the amoeboid movement of leukocytes and cancer cells in a rectangular microchannel. His simulation data indicated that the model captured a number of deformation patterns of motile cells: from a finger-like projection, which is a feature of cells migrating through the endothelial layer or into a chemoattractant-filled micropipette, to a lamellipodium-like projection that is observed for many cells actively migrating on a flat substrate.

Another group of talks focused on cell signaling. Sasha Jilkine introduced adipogenesis, the differentiation process of adipocyte (fat cell) formation from precursor cells contributing to increase of fat tissue in obesity. Previous work has found that some of these proteins increase and then decrease significantly during differentiation. She showed that three coupled network motifs found in adipocytes can explain these observations. Andre Levchenko asked how cells exposed to a diverse range of molecular cues are informed about the appropriate direction of migration. These cues can change in space and in time, and co-exist in a consistent or contradictory fashion. How live cells interpret these cues and compute the appropriate program of polarization and directional migration is not well understood. He discussed recent findings and modeling suggesting how integration of such cues can take place through a relatively simple set of molecular circuits. Orion Weiner focussed on recent advances in optogenetics, which enable us to interrogate signaling cascades in a manner that has been difficult or impossible with previous tools. Alba Diz-Muoz started by saying that far from being a passive participant, the plasma membrane is now known to physically, as well as biochemically, influence cell processes ranging from vesicle trafficking to actin assembly. In particular, changes in plasma membrane tension regulate cell shape and movement. She recently found that membrane tension (the force a cell has to overcome to protrude the plasma membrane) is necessary and sufficient to determine leading edge size and number. Research in recent years has shed light on the role of forces in cytoskeletal organization, but how changes in membrane tension are translated into changes in cell signaling is unknown. She reported how to use a combination of atomic force microscopy and fluorescence imaging of intracellular signals to uncover how neutrophils sense tension.

Many talks tackled the problem of cell polarity. Stan Maree discussed a fascinating similarity between animal and plant cells with respect to the organization of cytoskeletal elements in the regions of active protrusive growth and cell wall extension. He showed a multiscale model of a motile keratocyte, describing how the molecular players cause cell polarity and deformation. He then contrast this to the cell shape changes that occur in the pavement cells in the leaves of plants that form jigsaw-like patterns. He posed, by mathematically and computationally exploring the system, that similar modular principles play a role in animal and plant cells. Adriana Dawes turned her attention to highly conserved molecular players in polarization, including Par proteins, Rho proteins and actomyosin. Using a combination of modeling and experiments, she demonstrated the likely interactions between these key players responsible for initiation and maintenance of polarization in early embryos of the nematode worm C. elegans. Vernica A. Grieneisen addressed how computational and mathematical approaches combined with molecular studies and in vivo microscopy can help us understand polarity on three different levels: on the scale of the tissue, the cellular and subcellular tissue level.

A few talks discussed microtubules and molecular machines that use them. Dan Fletcher described recent work investigating the effect of volumetric confinement on the assembly of mitotic spindles in Xenopus egg extract, in particular, a limiting component model that addresses the effect of volumetric confinement on cytoskeletal assembly. This talk contributed to the growing view that cytoskeletal structures in cells are defined not only by their molecular components but also by the boundary conditions imposed on them. Melissa Gardner discussed length control of the metaphase mitotic spindle that is thought to be achieved through a mechanism in which spindle pole separation forces from plus-end directed motors are balanced by forces from minus-end directed motors that pull spindle poles together. Paradoxically, she describe that in contrast to this notion, metaphase mitotic spindles with inactive Kinesin motors often have shorter spindle lengths, along with poorly aligned spindle microtubules. A mechanistic explanation for this paradox is unknown. She showed that results of computational modeling, in vitro reconstitution, live-cell fluorescence microscopy, and electron microscopy suggest that the yeast Kinesin motor can efficiently align spindle microtubules along the spindle axis. Holly Goodson started by saying that the concept of critical concentration is a central idea in the understanding of biological polymers such as actin and microtubules. Classically, the critical concentration is accepted to be a single discrete value with several equivalent definitions. However, the mathematical underpinnings of this understanding are based on analysis of equilibrium polymers, which is problematic because these cytoskeletal filaments are instead steady-state polymers. This incongruity raises questions about whether present understanding of critical concentration as it applies cytoskeletal filaments is complete or even approximately correct. She has used computational models of systems of dynamic microtubules to investigate this issue.

Two fascinating talks were about the role of water in cells. Yoichiro Mori formulated a prototypical mathematical model that couples membrane mechanics with chemical diffusion and osmotic water flow. He then presented a numerical scheme for this problem, and demonstrated some computational examples. Sean Sun mathematically analyzed cellular pressure and volume control by considering both cytoskeletal dynamics and active regulation of cellular osmotic content. He showed that water permeation across the cell membrane is a major contribution to the slow phase of cellular mechanical response. He further demonstrated that water permeation alone can drive cell motility in confined environments. The last finding is significant for cancer cell motility in some situations.

4 Scientific Progress Made

There are a number of new understandings that emerged from the meeting. First, the concept of a motile cell as a free boundary object emerged, in which local boundary movements are governed by distributed spatial-temporal mechanochemical process inside the cell. Second, a few signaling pathways coupled to mechanical forces found their first mathematical representation. Third, a number of people realized how to make 3D, rather than 2D, modeling of cells practical and found a few open problems that need mathematical investigation. Forth, actomyosin cortical flow creating friction with the extracellular environment was realized to be a universal mechanism of coupling cell deformations to the substrate. Fifth, a number of design principles of multi-scale self-organization of cytoskeletal systems was classified. In general, we started to see general design principles of cell dynamics from mathematical point of view.

5 Outcome of the Meeting

The main result of the meeting is new collaborations that resulted from it. K. Keren started to collaborate with A. Mogilner on modeling collective migration of fish keratocytes they already had a working model of individual cells as free boundary objects, and now they started to use microscopy, biochemical perturbations and partial differential equations to understand how cells keep collective polarity and coherent movements. G. Papoian is planning to apply his 3D modeling methods to simulating D. Fletchers in vitro 3D actin assay with the goal to predict the hysteresis effects. A. Carlsson and W. Loosert are combining their experimental and theoretical efforts to elucidate actin waving behavior and its role in cell locomotory behavior. A. Levchenko and L. Edelstein-Keshet conceived a new idea on how complex stochastic Rac/Rho pathway leads to cell polarization. S. Sun is helping E. Paluch to simulate blebs in complex environments. J. Allard and A. Upadhyaya collaborate on developing very detailed models of cell-cell interactions. A few groups of people started to write grant proposals after finding ways to enhance each other research. Generally speaking, there is a growing sense of mathematical cell biology as a vibrant subdiscipline with an above average degree of cohesion of theory and experiment that is the most important outcome of this meeting.

6 What Lays Ahead

Cell biology is transitioning into a quantitative science characterized by increasing integration of modeling into experiment. In this transition, we have to proceed with numerous, often arbitrary, assumptions about the nature of processes and parameter values governing cell systems. One great future challenge is to improve quantitative experimental methods with an eye toward synchronizing modeling and experiments. Then, frequent back-and-forth between theory and experiment using models of varying scope and level of realism will allow us to overcome the arbitrariness and uncertainty. Another significant challenge is to make switching from one type of model to another a more standard, less ad hoc procedure, to ease modeling use and integration between theory and experiment. Models along this course will be impermanent and should be judged by how useful they are and what we can learn from them, not by how close we are to the elusive whole-cell model.