Experimentalist Meets Theoretician: A Tale of Two Scientific Cultures

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This is the story of how a small team of experimentalists and theoreticians collaborated to develop a theoretical model for vesicle formation during endocytosis. In telling our story, we hope to distil some general conclusions about the purpose and value of theoretical models and how best to navigate collaborations between experimentalists and theoreticians. We encountered challenges in building and publishing our model, but through our experiences we gained insight into how such collaborations can be profitably conducted. We also developed opinions about how theoretical models should be evaluated by peer reviewers and editors. During the evolution of our theoretical model, we educated each other, organized our thoughts and our data, developed a conceptual framework for understanding the mechanochemistry of endocytosis, and generated testable hypotheses that stimulated new experiments.

Getting Started

"Just as houses are made of stones, so is science made of facts; but a pile of stones is not a house and a collection of facts is not necessarily science."

—Jules Henri Poincaré

In sharing this story about a collaboration that has produced two theoretical modeling papers (Liu et al., 2006, 2009), we hope to communicate ideas that we have evolved about why it is good for experimentalists and theoreticians to collaborate, how such collaborations can be most productive with the least pain, what a theoretical model should and should not attempt, and why all of the effort is worthwhile.

Detailed quantitative microscopy and genetic studies had enabled the Drubin laboratory to acquire an enormous "pile of facts" describing the order and timing of events along the endocytic pathway in budding yeast (Kaksonen et al., 2005). But how could these facts be organized into a quantitative model for endocytosis? Key questions included the following: What are the underlying mechanisms that insure the proper sequence and timing of events? How do these events bend the plasma membrane? How does a yeast cell pinch off a vesicle without the GTPase dynamin, which is essential for endocytic vesicle scission in mammalian cells? Why is the process so robust against disruptive events? Although experiments had described the molecular events in exquisite detail, they were insufficient to answer these questions. A theoretical model was needed that was based on physics and expressed in mathematical equations. This model should make clear the principles governing the collective behaviors of the molecules and the mechanics of membrane remodeling.

Mutual Education and Iterative Pruning

"It’s better to know nothin’ than to know what ain’t so."

—Joshua Billings

For this collaboration, we recruited a biophysics postdoc, Jian Liu and, sequentially, two experimentalist postdocs, Marko Kaksonen and Yidi Sun. We started with informal, unstructured meetings, often in one of Berkeley’s cafés. The experimentalists described their data and interpretations, and the theoreticians speculated on physical explanations. The experimentalists stressed the remarkable precision of the timing and sequencing of events during endocytosis, and how the events were affected by perturbations. The theoreticians focused on mechanochemical aspects of the process, such as the stiffness and chemical composition of the plasma membrane, the forces required to deform the membrane, and how actin assembly and myosin might shape the membrane into an endocytic tubule. At first, it seemed as though the experimentalists and theoreticians were from different worlds, speaking to one another in alien tongues.

Vesicle formation is fundamentally a mechanical process. Consequently, the theoreticians found the simple cartoon sequence that the experimentalists had envisioned to be incomplete because it did not take into account the mechanical aspects of the process. The theoreticians asked many questions that the experimentalists had not thought about, such as where the energy for deforming the membrane and driving the scission reaction came from, how forces from actin polymerization could be coupled to the membrane, how membrane curvature might affect biochemical reaction rates, and how lipid phase boundaries might contribute to vesicle scission.

These were not the kinds of issues that the experimentalists were accustomed to thinking about. Thus, one immediate benefit from these meetings was that the experimentalists began to organize their data and their thinking to respond to these unfamiliar queries. These conversations with the theoreticians benefited the experimentalists, who profited from understanding the physical and chemical as-
pects of the endocytic process, and they were able to formulate new ideas for designing future experiments as a result of this understanding.

The theoreticians eagerly offered ideas about underlying principles that might govern the endocytic process. The experimentalists, however, often had to stop the theorists in their tracks, and point out more facts that the experimentalists had not told them about. Frequently, these additional facts would throw a wrench into the theorists’ models. The experimentalists would offer alternative ideas, but the theoreticians would often reciprocate by throwing a wrench into the works by pointing out that they violated some law of physics. And so it went for many sessions. One group would propose ideas, and the other would shoot most of them down, based on facts or physics. It was, however, a mutual education process between scientists with very different backgrounds and perspectives.

There was a positive trend to these meetings; participants contributed new ideas, and a pruning process ensued wherein the experimentalists and theoreticians would correct each other. Eventually, we reached a point where the physics allowed a set of possibilities, which the experiments had not yet refuted. This process eventually nucleated a model, which became successively more focused and fine grained. Although an abundance of data constrained the model, there were still plenty of assumptions, because not everything about the system was known. The final model will probably survive only until the next set of experiments is performed, at which point some of the assumptions will have to be modified, or abandoned. Wise theorists do not cling to their models when they conflict with the facts.

**Models Evolve by Cycles of Revision**

“Everything should be made as simple as possible—but no simpler!”

—Albert Einstein

A brief digression is instructive. We consider here the evolution of Oster’s Brownian Ratchet model, which stimulated experiments that forced it to undergo several refinements. In the first iteration of this model, a mechanism was proposed by which actin filaments could assemble against and move an object. The object itself moved by Brownian motion, and the polymerizing filament rectified the diffusion (Simon et al., 1992; Peschel et al., 1993). Experiments, however, showed that the original model could not be correct because *Listeria* and *Escherichia coli* moved at the same speed even though *Listeria* are smaller than *E. coli* (Theriot, 2000). The Brownian ratchet model was therefore modified to allow for the fluctuations in the filament and membrane in addition to the cell itself (Mogilner and Oster, 1999). This solved the problem that the velocity was independent of size. A subsequent study then showed that during assembly filaments are attached to the cell surface, so how could polymerization push (Carlier et al., 2003)? To fit these data, another revision of the model was necessary that took into account the attached filament subpopulation (Mogilner and Oster, 2003).

Several important lessons derive from this process in which a model is proposed and then iteratively modified when additional information becomes available. The first version of the model accommodated only data known at the time it was published. When additional facts emerged, it was necessary to modify the model to accommodate the new facts. Importantly, publication of the original model advanced the field by stimulating experiments, and the new data led to refinements of the model, and a better understanding of the mechanism. Rarely is the first version of a model the final word on the subject—for that matter, rarely is the first report on experimental studies the final word.

**Challenges and Benefits**

“A model should not fit all the facts, since not all of the facts are right.”

—Francis Crick

After the brainstorming sessions, generating the detailed mathematical model for endocytosis involved a great deal of calculation, and many more back-and-forth discussions. A major focus was evaluating the reliability of the data and estimating the physical parameter values from the published literature. This part of the process was not easy, and choices could be controversial because, in some cases, the parameters had not been measured or were not known accurately, and in other cases there were conflicting opinions about the parameter values. To address these uncertainties, sets of parameter plots, or “phase diagrams,” were generated to examine how the system would be affected by differences in the values of the key parameters. The phase diagrams made experimentally testable predictions for how the system should respond when key parameters were varied, and which variables were the most sensitive to perturbations.

Collaborations can go awry even when those involved have similar backgrounds. When experimentalists and theoreticians get together, differences in their intellectual cultures can impede working together harmoniously. Every discipline has its own terminology and jargon. The experimentalists know a wealth of detailed facts and the theoreticians know a great deal of physics. For such a collaboration to work, it is crucial that each group educates the other—but sometimes it is not fun to be corrected. Theoreticians make assertions about what can and cannot work based on the laws of physics, but experimentalists want to see empirical evidence. The culture of the experimentalist dictates that something is only true if you can measure it. When asked for the evidence, the theorist may reply that there is no need for evidence, because physics dictates that it must be so.

One sine qua non for a productive collaboration is the willingness of the theorists to master all of the experimental data. This is no small commitment, for the body of experimental data can be considerable, and it is generally unclear in the beginning what is relevant to possible models and what is not. But the theorist cannot delegate to the experimentalist the entire task of seeing which data support and which refute a particular proposed mechanism, for that involves a more detailed knowledge of the physics than the experimentalists can be expected to possess. For the experimentalists’ part, it is crucial that they thoughtfully organize their data to facilitate communication with the theorists, that they patiently communicate the experimental data to the theorists, that they understand that performing calculations can take a long time, and that they are willing to part with preconceived notions about mechanisms.

Our collaboration was indeed difficult at times, but in the end, the effort and periods of frustration produced satisfying and worthwhile results. We educated each other and refined our model to fit the facts and the physics. Out of our many discussions emerged a simple idea: membrane curvature can orchestrate the sequence of biochemical reactions that shape the membrane and pinch off the endocytic vesicle (Liu et al., 2009).
Publishing a Theory Paper for a General Audience

“Education is learning what you didn’t even know you didn’t know.”
—Daniel J. Boorstin

When it came time to publish our model, we had to contend with several issues. What should a modeling paper do or not do? How much mathematical detail should be reported? Who is the audience? During the peer review process, what are the criteria for evaluating a modeling paper?

We wanted our work to have the greatest impact possible, which meant that it should be accessible to the largest audience possible. Making a theory paper accessible can be challenging because many biologists are turned off by equations. So, it is the responsibility of the theorists to clearly exposit the physics, and in this the experimentalists are essential to ensure that the paper is comprehensible and interesting for nontheoreticians.

When writing for a general audience, it is a good rule to include very few equations in the main body of the text. In addition, it is crucial to communicate the main concepts of the model in pictures. The majority of the math should be relegated to appendices, the methods, or online supplemental data.

Although developing our model took a year, publishing it took even longer. We wanted to publish our work in a biological journal; yet, most biological journals do not have editors qualified to handle theory manuscripts or clear criteria for evaluating a theory paper. At two different journals, the editors chose to send the manuscript to four reviewers, two experimentalists and two theorists. Pleasing such a diverse set of reviewers was remarkably challenging. Several reviewers wanted us to “prove” that our model was true by performing additional experiments. But this was a modeling paper, not an experimental paper. So, what should a theory paper accomplish and what should be the criteria for evaluating such an article?

What a Theory Paper Should Accomplish

“Success consists of going from failure to failure without loss of enthusiasm.”
—Winston Churchill

A theoretical model should organize the experimental facts, clearly state the assumptions on which it is based, make testable predictions, and present a (hopefully) new conceptual framework for thinking about a biological phenomenon. A model is not meant to be the final word on a subject but a beginning that invites empirical tests of its validity.

In physics, theory goes hand in hand with experiment—but not yet in biology. Developing models and doing experiments are complementary activities but doing either is a full time job. So collaborations between modelers and experimentalists are the only way to bring together these two perspectives. The laws of physics tell us what cannot happen, and what could happen. But only experiments tell what does happen.

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