Being open to the unexpected

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ABSTRACT I am grateful to have received the 2019 Women in Cell Biology Mid-Career Award from the American Society for Cell Biology. My lab has been studying aging and longevity regulation since 2005, but along the way we have had some surprises. These unexpected findings have morphed from detours to main directions, changing how I view biology. As I look back I've come to appreciate the importance and joy that can come from being open to these surprise interests and rigorously pursuing them.

Musing on my dubious transition from "early" (presumably promising) to "mid-career" (established, I guess?) I've been struck by how my lab has taken on a life of its own, branching out into directions I hadn't anticipated. When I started out, I had a few ideas that I thought were good, and some adventurous funding agencies took a chance on me, providing the support that these fledgling projects needed. That early stage of the lab was mostly characterized by bravery—on the part of my students, mostly—and hard work (by all of us). My earliest trainees took some half-baked ideas of mine and turned them into real projects addressing reproductive aging (Luo *et al.*, 2010) and cognitive decline (Kauffman *et al.*, 2010), and along the way, developing *Caenorhabditis elegans* as a model for both of these human aging declines. Luckily, my first two students, Shijing Luo and Mandy Kauffman, were unaware of the prevailing views in the field at the time, that "worms could reproduce forever" (they cannot, as they are limited by declining oocyte quality) and that "worms don't have memory" (they do, and it's highly conserved with mammalian memory), and plowed ahead. The lab went on to study both of these areas in quite a bit of detail, identifying pathways that modulate the rates of these declines, characterizing the molecular and cellular details of the decline processes (Stein and Murphy, 2014), and even finding some interventions to slow them down (Templeman *et al.*, 2018; Lakhina *et al.*, 2019). Because those early students and postdocs laid the groundwork, we are now in a position to use worms as a platform to address all kinds of human age-related declines. Most of the projects in my lab use these tools to ask questions that we hadn't even thought were possible when we first started, including identifying new human genes that might affect age-related neurodegeneration (Yao *et al.*, 2018), or finding small molecules that can help stave off neuronal decline (Lakhina *et al.*, 2019). We have also developed new molecular techniques and assays when we needed to answer questions that were hampered by the existing approaches. For example, Rachel Kaletsky tackled the problem of releasing cells from the tough outer cuticle of adult worms (Kaletsky *et al.*, 2016, 2018), enabling a whole suite of new projects. These techniques allowed us to characterize different tissues with age, and identify pathways that regulate the aging of one tissue (especially neurons and oocytes) differently from the rest of the animal.

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But more and more often, we also find things that aren't "supposed to work that way." A student or postdoc shows me their results—results that are rock solid, statistically speaking—that work exactly opposite from the way we thought they would. Or they make an observation that is so weird and unexpected that I feel like they have to drop everything else and just figure out this puzzle. I LOVE this part of science—finding the unexpected, even if it means that I personally have been wrong about an idea or prediction, or that the question doesn't seem to fit with what we initially set out to study. There is nothing more satisfying that knowing that your

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student or postdoc is so technically and experimentally capable that what they are showing you are not bad data, but instead they are pulling aside a curtain to show us how biology really works.

One of the first examples of this happened early in my lab, when we were just doing what we thought was a control—lifespans while blocking matricide—and found that the TGF-beta dauer mutants are actually long-lived, if you just prevent their progeny from hatching inside them. That was surprising to me, since it went against the dogma at the time, that only insulin-IGF-1 signaling mutants could both extend lifespan and affect dauer. My first postdoc, Wendy Shaw, did tons of lifespans on every mutant in the pathway over and over again to be sure we were right, since we knew there would be resistance to that idea. (I still don't know if people know it!) In the end though, this seeming detour (Shaw *et al.*, 2007) nicely complemented the work we've done in my lab, highlighting the interconnections between different signaling pathways that regulate longevity, development, and reproduction (Luo *et al.*, 2010).

Later, my student Cheng Shi was studying a new mutant that extends reproductive span. When he showed me that his wild-type worms shrank by almost 40% (and lived half as long) after they were mated (Shi and Murphy, 2014), he already knew that I'd wonder why no one had reported this before—but he had enough solid data already to convince me, and we decided to drop everything else he was doing to figure this out. Again, what we thought was a detour ended up being an important new line of research that explained to us that what we think of as "longevity" pathways are in fact there to *accelerate aging* when properly coupled to reproduction, and can be hijacked by males to benefit themselves, an ongoing battle of the sexes (Shi *et al.*, 2017, 2019). Our accidental findings and their implications have made me think harder about why longevity pathways exist in the first place. These results made me realize that much of the aging field studies longevity in the absence of consideration of reproduction, when in fact reproduction is the main driver of aging and longevity regulation decisions.

More recently, my student Rebecca Moore found that worms could "remember" behaviors that their mothers, or grandmothers, or even great, great grandmothers, had learned (Moore *et al.*, 2019). On its surface, this seemed like a project on epigenetic inheritance, a topic that my lab had not previously studied. But at its core is the idea that animals can learn information about their environment, interpret that information to make longevity and behavioral decisions, and then pass on this information to their progeny to increase their odds of survival—a way of being "good mothers." In the end, this type of regulation is not so different from the decisions that regulate aging and reproduction, even if it's a brave new world for my lab.

Ultimately, these "weird" things we keep finding are just new aspects of biology that we hadn't anticipated, because biology is way more creative than most of our imaginations. The lab's work is leading us into areas I never thought I'd work on (or swore I never would) because the data are just so compelling. It makes me excited every day to come into the lab and chat with people, finding out what they have discovered.

BOX 1: A few suggestions for getting from here to there.

Since as a mid-career PI, I might be expected to have learned something along the way, I offer a few ideas that I think have helped my lab get to where it is now:

- **Prepare yourself for luck:** Luck is a really big part of what we do. But being ready is important, too: pay attention to students' observations, be trained in different backgrounds, and make sure you are right (quantitation is key).
- **Train students to be their own biggest skeptics, and then listen to them when they find something unexpected**. Most of my lab's best projects come from "weird' observations in carefully measured, quantitative assays, so that even though the results don't match our previous assumptions, they are not wrong. As a PI, the important thing for me to do is figure out which of these things is "real" and decide when to pay attention to it, when to remember it for later, and when to shelve it.
- **Be brave and different**: It's easy—and tempting—to do what everyone else in the field is doing, or to become convinced that only one question is interesting, or to listen to people who tell you that a particular approach can't be done. One of the advantages of being a little more isolated is that my students don't hear as many rumors—like that worms don't have memory, or you can't do biochemistry in worms—and that means they will try to do something that they might have otherwise talked themselves out of trying. You may find yourself in risky territory (sometimes it feels the same as being a middle schooler walking through the haze of high school students smoking outside the door to get to my math classes). But ultimately it will pay off.
- **Have a culture of high standards**: It takes a lot to convince my lab people that something is real. These aren't just my standards, but the standards of the members of the lab. They are quick to call things out, ask for controls, everything—so even if I'm thinking it, I don't have to be bad cop all the time!
- **Respect each other**: My lab has been mostly women and other underrepresented minorities from the start. Normally I don't think about this much, but I notice when I'm in groups predominated by men, they talk over one another and interrupt women—something that does not happen in my lab's group meetings for the most part. My lab people don't pull punches (see a Have a culture of high standards above) but they do listen to one another.
- **Be willing to work hard**: Most projects require putting in hard work, building tools, or carrying out really laborious work. I often thought about my family planting thousands of trees in Kansas when I was a kid while I did thousands of PCRs to build my first microarrays—I was well prepared for hard work. Sometimes you just have to do the boring thing to get work done. I talk about unexpected results, but they often came while we were working hard on something else. The slog is real, and unavoidable.
- **Love the puzzle**: I recently discovered (through Twitter) that I share this love of logic puzzles with other women scientists. I think this is part of why I really like solving these biological puzzles, which helps balance our curiosity-driven science with expectations that we carry out applied research. We are still finding things that "shouldn't be true" but I trust my trainees to do their work well enough that we will figure out the puzzle.

As my lab matures, I have started to think more about how our findings will impact the world. Most of my work is in the aging field, and I do worry about the growing economic imbalances that lead to health care disparities. I don't want our findings to benefit only the rich, but rather I want them to help the majority of people to live better lives. I think this idea will have growing importance as our work extends to the clinic. But along the way, I'll let my lab keep finding the unexpected and solving new biological puzzles, and we'll see where it leads us.

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