ASCB AWARD ESSAY

A Tale of Mother and Daughter

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Submitted September 29, 2009; Revised October 1, 2009; Accepted October 7, 2009

Loving science and nature and being a scientist can be very different, yet the two are so intertwined in a scientist's life that you will certainly experience both aspects. This essay presents my perspective on how, as one who loves science and nature, I came to fall in love with centrosome behavior in stem cells and how I came to run a lab as a scientist. When I started, there was a big gap between my love for science and my experience as a scientist. I filled this gap by learning a "laid-back confidence."

Before the beauty of cell biology (or whatever you love), who you are (i.e., your age, gender, or race) is immaterial. Yet history shows that the ease with which you can pursue science is influenced by who you are. This has certainly been my experience. The key is to find a way to fill in the gap between who you are and what you are (i.e., a scientist), a goal in which we must all support each other. It is my hope that this essay will convey something helpful to those who are at early stages of their career and might be encountering obstacles because of who they are.

EARLY DAYS

My childhood was spent in a small town in Japan. Until leaving for college in Kyoto, I barely remember being conscious of my gender. My parents hardly treated me as a girl (or a boy), with their only expectation being that I explain things logically. (Just asking them to buy something was almost like writing a NIH grant, at least relative to the ability of a 10-year-old.) This certainly influenced me in that I did not think my gender would matter in my future pursuits—which turned out to be biology.

The reason I decided to pursue biology was a combination of my fascination with all living creatures and my inclination toward the logical. After reading a series of novels and essays by Albert Camus during high school, I decided that he was the most logical person of those who have addressed the meaning of life. His ultimate question was whether there is a meaning to life or not. This must be the most urgent and nagging question, since the negative answer would mean logical termination of your life! Although biology cannot directly answer this philosophical question, it gives us good ideas as to why human beings have evolved to have a brain capable of asking such a question.

Certainly, the way I was nurtured by my parents prepared me to become a biologist. However, during my undergraduate

and graduate school years, I was shocked by people's expectation that I, as a female, must fill a woman's role. These years were the toughest of my life, as I was unwilling to adopt such a role. It was around this time that I almost decided not to pursue a career as a scientist. In retrospect, I believe I was undergoing a sort of adjustment as a result of being split between what I truly loved and other people's expectations. My husband, who is one of the few people who does not really see me as a woman (don't ask me how we ended up marrying!), suggested that we find postdoctoral positions in the United States, a move that profoundly changed my life. Years later, he told me that I was the reason for the move, as I clearly seemed depressed and was declining. This move turned out, without a doubt, to be one of the best decisions in my life, although it was not really a decision I made by myself.

FASCINATED BY CELL BIOLOGY

My fascination with asymmetric stem cell division, which produces a new stem cell and a differentiating cell, started during my postdoctoral years with Minx Fuller at Stanford University. Before this time, both as a graduate student in Mitsuhiro Yanagida's laboratory and a postdoctoral fellow in Shunichi Takeda's laboratory, I studied mechanisms that allow cells to create their exact copies by carefully dividing in two. Being used to cells' elaborate way of making exact copies, I thought the idea of asymmetric division was unusual, though I knew that it was essential to the development of multicellular organisms.

Often we do not know what we really love until we have found it. This was the case when I first observed *Drosophila* male germ line stem cells (GSCs) expressing the marker GFP- α -tubulin in Minx Fuller's laboratory. The GSCs were nicely aligned in a rosette around the hub cells, illustrating the beautiful nature of cellular arrangements in the context of tissue architecture. Hub cells function as a major component of the stem cell "niche," which specifies stem cell identity by sending a signaling ligand to GSCs. I really fell in love with this system upon seeing the GSC mitotic spindle for the first time. Seeing the GSC mitotic spindle nicely oriented toward the hub cells was almost like being in outer space and realizing that the Earth was not everything after all. The shape of mitotic spindle in male GSCs was almost boringly exactly what I was accus-

DOI: 10.1091/mbc.E09-08-0658

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Yukiko M. Yamashita is the recipient of the 2009 American Society for Cell Biology Women in Cell Biology (WICB) Junior Award.

tomed to seeing. However, this spindle was nicely oriented perpendicularly toward the hub, a feature that one could never appreciate if viewing cells in isolation. I remember having the distinct feeling that this is where cell biology must meet developmental biology.

The orientation of the mitotic spindle with respect to the hub was not the only surprise. My assumption, coming from cell biology textbooks, was that two centrosomes that have duplicated at the G1/S transition should separate right around the G2/M transition (in prophase; i.e., that the two centrosomes would stay together for a long time until the G2/M transition). However, male GSCs almost always had separated centrosomes, presumably because centrosomes separate from each other much earlier in the cell cycle. More surprisingly, one centrosome was always closely associated with the hub cells, and the other appeared to be migrating toward the opposite side (Yamashita *et al.*, 2003).

FOLLOWING THE MOTHER AND DAUGHTER

After this observation that stereotypical centrosome positioning prepares the oriented mitotic spindle, Minx and I started getting asked repeatedly at meetings whether mother-daughter centrosome differences might underlie this stereotypical centrosome behavior. By then, it had long been known that the mother centrosome differs from the daughter because of the age difference in centrioles (Piel *et al.*, 2000, 2001). I began looking for a way to distinguish mother and daughter centrosomes. However, because *Drosophila* did not have any known mother centriole-specific components such as ϵ -tubulin (Chang *et al.*, 2003), simple antibody staining would not work. I created constructs with γ -tubulin (e.g., GFP- γ -tubulin and DsRedTimer- γ -tubulin) with the vague hope that, if I could come up with perfect fixation/extraction conditions, I might be able to see the centriolar population without seeing the PCM population, an approach that failed.

One day, I saw a newly published article describing a new *Drosophila* centriolar component that is incorporated into the centriole only during duplication and stays there forever (or at least for a very long time; Martinez-Campos *et al.*, 2004). Almost at that moment, I ordered oligos to construct a transgene for transient expression of a tagged form of this centriolar marker (PACT). I tried multiple tags and expression systems such as the DsRed-Timer tag, which changes its color over time, hoping I might be able to label the mother centrosome green and the daughter red. I also tried the *gal4/gal80* temperature-sensitive system for temporal control of expression.

In the end, the unexciting method of heat-shock-induced GFP-PACT worked. This approach allowed me to clearly see that the GFP-marked daughter centrosome is always away from the hub (Figure 1; Yamashita *et al.*, 2007). Without a doubt, making this observation was one of the best moments in my life. I really do not know why the mother-daughter centrosome issue fascinates me so much. Probably it's because this clearly illustrates the simplicity and beauty of nature.

SCIENCE AS A CAREER FOR WOMEN

Having been enjoying science for a long while, I have rarely been reminded of gender (except when it was pointed out that perhaps centrosomes in *male* GSCs should be called "father and son centrosomes" rather than mother and daughter centrosomes). This is due, in part, to "laid-back confidence," one of the best things I learned from Minx Fuller. That is, there is no need to fight, struggle, or sacrifice anything. Just love science.

Hub, centrosome Daughter centrosome Germ cells

Figure 1. An example of a GFP-marked daughter centrosome migrating away from the hub. The daughter centrosome is marked by transient expression of GFP-PACT (green). Centrosomes are marked by anti- γ -tubulin antibody (red). Hub cells are marked by anti-Fasciclin III antibody (red). Germ cells are marked by anti-Vasa antibody (blue).

And you can do whatever you set out to accomplish. It was such a comfort for me to (gradually) acquire this confidence. Without this, I would not have been able to decide to pursue an independent career in academia.

The environment for female scientists is much better than that for women in other career paths, because of the flexibility and relatively fair evaluation system in science. However, this should not be used as an excuse to avoid further improvement in female scientists' working environments. We still hear depressing reports of female scientists disappearing along the career path, the tenure ratio for women not being equal to that for men, or comments about women's incapability. I believe that laid-back confidence can be key in improving the situation for women in science. The progress might be slow and inefficient, a pace that we scientists have difficulty accepting ("can't we get the result in a week?"). Nevertheless, we must continue to progress steadily and build evidence that women can contribute to science.

ACKNOWLEDGMENTS

I thank Minx Fuller, Sean Morrison, Tony Mahowald, Allan Spradling, James Nelson, Takashi Toda, and Kentaro Nabeshima, and Hauro and Tomoko Yamashita for inspiration, encouragement, guidance, and support. I would not be where I am now without these people.

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