People & Ideas

Marilyn Farquhar: From the beginning

Farquhar studies cellular secretion, endocytosis, and podocytes in the kidney.

Few people have a perspective on cell biology that can compare to Marilyn Farquhar's. She was there at the very beginning and, over the course of her career, has both witnessed and helped guide the evolution of the field. She has served as faculty at the University of California, San Francisco (UCSF) and at Yale. And, as the first head of the Department of Cellular and Molecular Medicine at the University of California, San Diego (UCSD), she nurtured the development of the department's research and educational programs.

Through her research, Farquhar has explored the inner secrets of cellular function. Her work has revealed novel aspects of cell junctions (1, 2), endocytosis (3, 4), secretion (5), podocyte biology and pathology (2, 6), and G protein functions (3, 7). And she has even more in store for us, as we heard when we spoke with her at her office at UCSD.

A STROKE OF LUCK

You went to college in the late 1940s. Wasn't that unusual for women at that time?

It was less usual than it is now. Today, the number of women surpasses men in colleges and medical schools. But in my case, it was assumed by my parents—especially my mother—that I would go to college, from the day I was born. My mother had started out at Mills College, but when her father had a bad year in farming she had to come home, so she was determined that my sister and I would get a college education. It was my good fortune that I was born in California, which has the University of California—the top public university in the nation. Thus, I was provided with a first-class education for very little money.

I was originally interested in medicine. My role model was Frances Zumwalt, a friend of my mother's and a pediatrician who practiced out of her home. So, I went to Berkeley as a premed student, graduated, and went to medical school at UCSF for a year and a half before deciding to enter a PhD program.

How did you first come to use the electron microscope?

As a graduate student I started working in the laboratory of a pathology professor who had been one of my medical school instructors. It happened that, a year after I started, my professor purchased the first electron microscope at UCSF. He was interested in glomerular disease, so I was involved in that work and later collaborated with others at the University of Minnesota to examine renal biopsies. We were lucky to be the first to see glomerular pathology at the electron microscope level. Among other things, out of that work came the first description of the now diagnostic foot process loss, or "foot process effacement," that occurs in the nephrotic syndrome. But I had always been interested in endocrinology, so for my PhD thesis project I studied secretion in pituitary cells using the techniques of an experimental endocrinologist.

I was particularly fascinated by the Golgi because with a light microscope it appeared to be only empty vacuoles but with the electron microscope the Golgi was clearly recognizable and waxed and waned during secretion. I was studying secretory granule formation in the Golgi, granule discharge by exocytosis, and other phenomena that are now quite well known. But we were seeing them in pituitary cells for the first time. It was an exciting time because whatever you looked at in the electron microscope was new.

"I was particularly fascinated by the Golgi."

AT THE START OF IT ALL

You did your postdoc at Rockefeller…

I wrote to Keith Porter and to George Palade saying that I was looking for a position in an electron microscopy lab. Keith didn't have any room in his lab, but George wrote back saying he had a project about the kidney glomerulus. So I ended up in George's lab. At Rockefeller I got what I had never had before: formal training in the new field of cell biology.

PHOTO COURTESY OF MARILYN FARQUHAR

Marilyn Farquhar

That's where you did your first work on cell–cell junctions…

Yes. We were using electron-dense tracers to establish the permeability properties of normal and nephrotic glomeruli and the role of the podocyte in protein uptake. It was during that work that I discovered the junctions. We named tight junctions and adherens junctions, and using tracers we established that the tight junction serves as a seal that prevents passage of proteins along the intracellular spaces. That paper is unusual in today's terms because it is so detailed and has so many figures. One couldn't write a paper in that style nowadays and have it accepted. However, it turned out to be my most-cited paper.

Over the years I have continued to study junctions in the podocyte, which has modified junctions called slit diaphragms. Recently we carried out a mass spec proteomics analysis in collaboration

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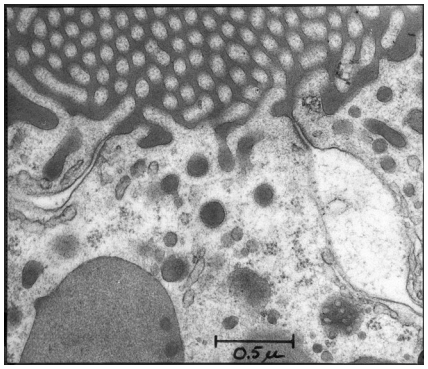

IMAGE COURTESY OF MARILYN FARQUHAR

An electron micrograph from Farquhar's 1963 paper in *J. Cell Biol.* showing the presence of tight junctions between kidney tubule cells.

with John Bergeron and discovered some new proteins that we think may be components of the slit diaphragms.

After Rockefeller, you went back to UCSF and started your own lab…

At UCSF I continued to work on junctions but also returned to my old loves, using tracers and cytochemistry to probe the secretory process in pituitary cells and leukocytes. That work yielded the first description of crinophagy, the process by which secretory granules are taken up and disposed of in multivesicular bodies and lysosomes, as well as the first insights into the biogenesis of leukocyte granules.

Later, I took a sabbatical back at Rockefeller. I had intended to stay only a year but ended up staying longer because George and I were married. I had a small lab within the department, and George and I collaborated where there was a mutual interest. For example, while he was working on Golgi fractionation, I was doing enzyme cytochemistry on the fractions. But we maintained our separate labs and separate programs throughout our entire careers.

A LONG TRIP

You were at Yale for a number of years before moving to UCSD. What has been your research focus?

Well, I never gave up studying the kidney glomerulus, secretion, Golgi, or junctions. I've carried these interests throughout my entire career.

At Yale we became interested in the fate of secretory granule membranes that merge with the cell membrane during exocytosis. We obtained evidence that these membranes are recycled and reach the Golgi in secretory cells. We also identified several glomerular components (megalin, podocalyxin, heparan sulfate proteoglycans) important for glomerular functions.

We discovered megalin as the target of an autoimmune kidney disease known as passive Heymann nephritis, which is an animal model of a glomerular autoimmune disease called membranous glomerulonephritis. We decided that to understand the molecular mechanisms involved in this disease we needed to identify the target antigen. It turned out that megalin is located in clathrin-coated pits at the base and the sides of the foot processes in the glomerulus. That led us to try to understand what this protein does.

When we found that it is in the LDL receptor superfamily, we understood it is an endocytic receptor. It turned out to be the main receptor in the proximal tubule responsible for recovery of many proteins and vitamins that are filtered through the glomerulus. We studied its trafficking and found that megalin recycles from pericentriolar recycling endosomes instead of from early endosomes and that ARH, a specific adaptor for members of the LDL receptor family, follows megalin throughout its intracellular journey. Most recently we found that ARH actually directs megalin to the endocytic recycling compartment, and we showed that the reason megalin goes to the recycling compartment is so that it can be proteolytically processed to produce fragments that can influence megalin transcription.

> "Currently, our focus is on one particular molecule called GIV."

It was a long trip from the identification of megalin as a target antigen of an autoimmune disease to understanding the functional significance of its trafficking route. Now we have other more pressing projects.

Such as?

At UCSD I started studying G proteins because I wanted to understand how signaling occurs from intracellular membranes. The classical paradigm assumes that heterotrimeric G proteins are found only at the plasma membrane. So, when G proteins were found inside the cell and, in particular, when Gαi3 was found in the Golgi, I had to find out what it was doing on intracellular membranes. Over the last 15 years we have discovered a number of regulators (several novel GAPs and GEFs) of heterotrimeric G proteins that are found on intracellular membranes, and it has become clear that G proteins regulate many cell processes including Golgi functions, endocytosis, autophagy, and cell migration.

Currently, our focus is on one particular molecule called GIV, which stands for "G protein–interacting protein associated with vesicles." This protein serves as a GEF for Gαi proteins. It regulates cell migration in response to growth factors and determines the fate of growth factor receptors. It even promotes survival of podocytes early after glomerular injury. We're now trying to understand how it connects G protein signaling to so many different cell processes.

1. Farquhar, M.G., and G.E. Palade. 1963. *J. Cell Biol.* 17:375–412.
2. Fukasawa, H., et al. 2009. *J. Am. Soc. Nephrol.* 20:1491–1503.
3. Beas, A.O., et al. 2012. *Mol. Biol. Cell.* 23:4623–4634.
4. Shah, M., et al. 2013. *J. Cell Biol.* 202:113–127.
5. Smith, R.E., and M.G. Farquhar. 1966. *J. Cell Biol.* 31:319–347.
6. Farquhar, M.G., et al. 1961. *J. Exp. Med.* 113:47–66.
7. Ghosh, P., et al. 2008. *J. Cell Biol.* 182:381–393.

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Farquhar and her husband, George Palade, hiking in Colorado.