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FIRST PERSON

First person – Ana Romarowski

First Person is a series of interviews with the first authors of a selection of papers published in Journal of Cell Science, helping early-career researchers promote themselves alongside their papers. Ana Romarowski is the first author on 'Super-resolution imaging of live sperm reveals dynamic changes of the actin cytoskeleton during acrosomal exocytosis', published in Journal of Cell Science. While completing this research, Ana was a postdoctoral research associate in the lab of Mariano Buffone at Instituto de Biología y Medicina Experimental, Buenos Aires, Argentina. She is now a postdoctoral research associate in Pablo E. Visconti's lab, University of Massachusetts, Amherst, USA. Her research interests are in reproductive biology.

How would you explain the main findings of your paper in lay terms?

Sperm have a particular vesicle in their heads called the acrosome. In all other cell types the process in which a vesicle is released is called exocytosis. In sperm, the release of the acrosome is called acrosomal exocytosis and is an absolute requirement for fertilization in mammals. Acrosomal exocytosis shares many features with all other exocytotic processes, one of them being regulation by the actin cytoskeleton. Actin is a protein that can act as a unit or as a polymer. Actin polymerization can serve as a scaffold for other proteins and also as a barrier to the occurrence of certain processes (i.e. the exocytosis of a vesicle), whereas actin depolymerization can facilitate those processes. Using super-resolution microscopy in live sperm, we observed that sperm possess different polymerized actin structures within the sperm head. Interestingly, we found that only some of them are depolymerized before acrosomal exocytosis takes place. This is the first time that we have been able to see actin cytoskeleton dynamics and the process of acrosomal exocytosis simultaneously in live sperm.

Were there any specific challenges associated with this project? If so, how did you overcome them?

One of the challenges associated with this project was to be able to study actin cytoskeleton dynamics in live cells. All the studies that have been done before have used phalloidins, which are toxic and not capable of crossing the cell plasma membrane. But as acrosomal exocytosis is also a dynamic process, we were interested in studying it in real time using live cells. For that purpose, and for the first time in sperm, we used the novel membrane-permeable fluorescent probe SiR-actin, which binds to filamentous actin *in vivo*. Another challenge was how to study the actin cyotskeleton changes and acrosomal exocytosis simultaneously. For that we loaded the sperm with SiR-actin, and also with FM4-64, which allowed us to observe the process of acrosomal exocytosis. Setting up the optimal conditions for using both probes together with live cells was a challenge that we successfully overcame.

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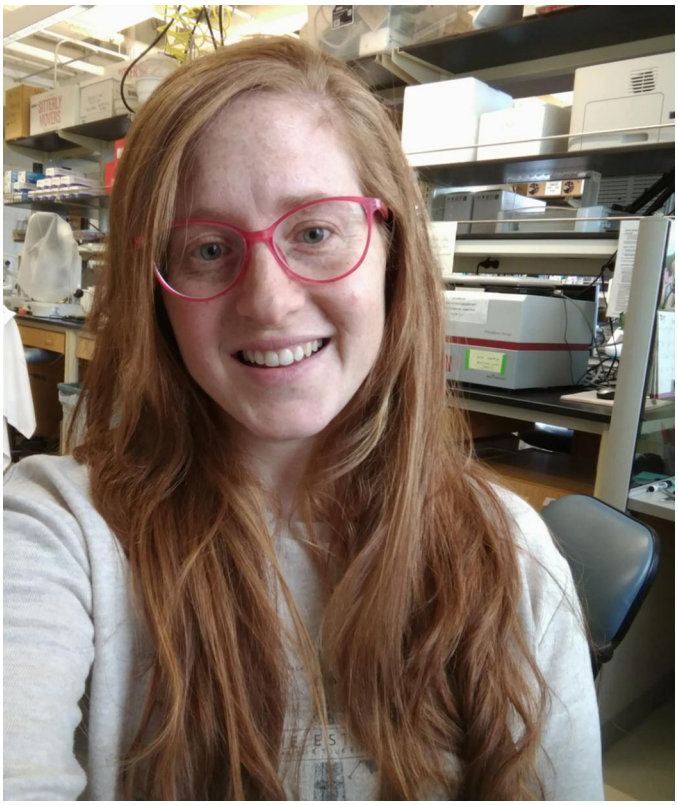

Ana Romarowski

When doing the research, did you have a particular result or 'eureka' moment that has stuck with you?

"Both mentors helped me to think critically and to find the best way to achieve the goal of the project."

The use of SiR-actin revealed to us the presence of novel filamentous actin structures within the sperm head, and I would say novel because, unlike commonly used phalloidin staining (which homogenously stains the sperm head), SiR-actin shows filamentous actin in specific regions of the sperm head. As these filamentous actin structures were novel, we coined a name for each of them in relation to their localization within the sperm head. When we were recording and saw one of those actin structures being depolymerized and then acrosomal exocyotsis taking place, the involvement of the actin cytoskeleton in this exocytotic process became evident, which was indeed an exciting moment for us.

Why did you choose Journal of Cell Science for your paper?

We chose Journal of Cell Science for our paper because it publishes research articles of high-level science and the scope of the journal is broad. We are very honored to have the opportunity to publish in this journal of excellence.

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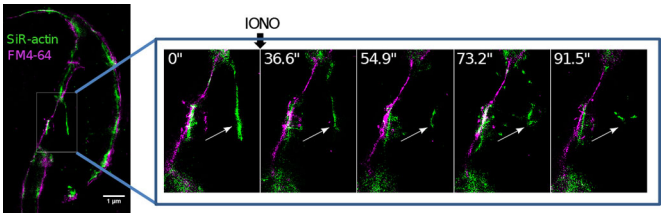

Representative super-resolution image of a sperm, which at the beginning of the recording possesses the septum F-actin structure (left). In higher magnification, the time course sequence of that structure after addition of ionomycin is shown. SiR-actin (green) and FM4-64 (magenta).

Have you had any significant mentors who have helped you beyond supervision in the lab?

My PhD mentor Dr Mariano Buffone had the idea of pursuing this project and he helped me through all the difficulties I encountered during this process. I was fortunate to have had a supervisor like Mariano and I am thankful to him for trusting me with this amazing project. We discussed all my results together and he helped me find the best way to explain them when writing this manuscript. I enjoyed our meetings because he transmits to me that desire to move forward and work as much as I can. I had fun working in his laboratory, and also felt comfortable to express my opinions. Also, Dr Alberto Darszon in Mexico allowed me to do the project at his lab using the amazing microscopy facility there. Both mentors helped me to think critically and to find the best way to achieve the goal of the project.

“Go and do.”

What motivated you to pursue a career in science, and what have been the most interesting moments on the path that led you to where you are now?

I have always been interested in understanding how things happen. I am a very curious person and a scientific career allows me to ask questions and answer them: that is one of the things I enjoy most in my life! I started studying philosophy because I had the naive idea that if I studied the mother of all sciences I was going to be able to understand everything. However, after my first two years I realized that I needed something more concrete, so a friend of mine recommended that I move to biology. I still remember today how much I enjoyed the first subject of my biology degree: a molecular biology classes given by the excellent Dr Alberto Kornblihtt. This inspired my desire to dedicate myself to biology research.

Who are your role models in science? Why?

When doing research you usually spend more time in your lab than in your own house, so I picked as my role model in science someone who I had the opportunity to work with. A researcher that I admire is Dr Mariano Buffone, my PhD supervisor. I deeply admire some of his qualities as a researcher: his creativity, his ability to catch the audience by passionately transmitting his ideas, his ability to read many papers per unit of time and above all his perseverance when things do not work out.

Also, I always try to keep in mind something my grandfather used to say: “Go and do”, which implies having a committed attitude in life that pushes you to action and to not remain in pure speculation, which leads to nothing.

What’s next for you?

Currently I have moved from Argentina to the USA and I am a postdoctoral research associate in Dr Pablo Visconti’s laboratory at the University of Massachusetts, Amherst. My postdoctoral training will complement the skillset I acquired during my PhD and expand my knowledge beyond the male gamete. To build upon the investment the government of Argentina made in my education, I will continue to contribute to the field of reproductive biology as I want to become an independent researcher and educator in my home country.

Tell us something interesting about yourself that wouldn’t be on your CV

Most of my time I spend doing research but I would never be happy enough without spending time with the people I love. My future husband, my family and my friends are the ones who allow me to be happy in life.

Reference

Romarowski, A., Felix, A. L., Gimenez, P. T., Gervasi, M. G., Xu, X., Luque, G. M., Gimenez, G. C., Cardenas, C. S., Gomez, H. V. R., Krapf, D. et al. (2018). Super-resolution imaging of live sperm reveals dynamic changes of the actin cytoskeleton during acrosomal exocytosis. *J. Cell Sci.* **131**, jcs218958.