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FIRST PERSON

First person – Daniela Sparvoli

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. Daniela Sparvoli is first author on 'Diversification of CORVET tethers facilitates transport complexity in *Tetrahymena thermophila*', published in JCS. Daniela conducted the research described in this article while a postdoc in Aaron P. Turkewitz's lab at The University of Chicago, IL. She is now a postdoc in the lab of Maryse Lebrun at Université de Montpellier, France, where she is interested in understanding the molecular mechanisms underlying membrane trafficking in the ciliate *Tetrahymena thermophila,* a unicellular eukaryote.

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

Each cell is divided into multiple membrane-bound compartments, which are interconnected by a constant exchange of material. Regulating what gets exchanged, and when this occurs, is critical to the proper functioning of the cell. Exchange occurs via carriers called vesicles, which have to be precisely targeted between specific donor and acceptor compartments. This targeting involves protein complexes on the vesicle surface. Two related complexes, called CORVET and HOPS, function in animals and fungi. In our study, we show that in a unique evolutionary lineage, which includes the single-celled protist *Tetrahymena thermophila*, the HOPS complex is not present, but instead the cells express six distinct CORVET complexes. We find that each CORVET complex is associated with a distinct pathway, and in some cases we could infer which element of the CORVET complex is responsible for this specificity. Our findings suggest that the expansion in the number of CORVET complexes in this lineage, over evolutionary time, was important in generating the elaborate network of compartments that exist in protists like *Tetrahymena*.

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

I think that the main challenge of this project was to identify the subunit composition of the six CORVETs by immunoprecipitation and mass spectrometry. The major difficulty arose from the fact that all the subunits in the complexes are expressed at very low levels in *Tetrahymena.* Thus, in order to isolate intact complexes in amounts sufficient for protein analysis, we had to look for alternative approaches to scale up the amount of cells but not the quantity of the resuspension buffer. We overcame this issue by using cryomilling, which consists of quickly freezing cell pellets and then reducing them into small particles. In this way, the cell powder can be easily dissolved in a reasonable amount of buffer. The technique was successfully used in other systems, and it allowed us to considerably increase the amount of starting material, while keeping the experimental volumes manageable.

Daniela Sparvoli's contact details: Université de Montpellier-UMR5235, Place E. Bataillon Bât 24, 34095 Montpellier, France.
E-mail: daniela.sparvoli@umontpellier.fr

Daniela Sparvoli

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

I am not sure I had a true 'eureka' moment, but my favorite memory of this research project was when we received the results of the mass spectrometry analysis for the six Vps8 pulldowns. It was exciting to finally know the subunit composition of the *Tetrahymena* CORVET complexes, and see how it appeared largely consistent with evolutionary relationships inferred from previous phylogenetic studies.

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

The Journal of Cell Science is a rigorous and well established peer-reviewed scientific journal, which publishes outstanding and original discoveries in many areas of cell biology. We believe that our research findings contribute to advancing the understanding of cell biology, and thus fit very well within the scope of the journal. The journal is aimed at a broad audience with diversified interests in cell biological processes, so it is well-suited to promote our research and our model system *Tetrahymena thermophila*. Our current understanding of membrane trafficking relies mainly on studies conducted on animals and fungi, members of the opisthokonta group, which represents just one of the five major eukaryotic lineages. We believe that cell biological studies from non-Opisthokont clades, such as the SAR supergroup to which ciliates belong, will expand our understanding of the structure and function of endomembrane networks, providing a more complete and accurate understanding of the evolution of modern cells. JCS is certainly a scientific journal that values this plurality in science.

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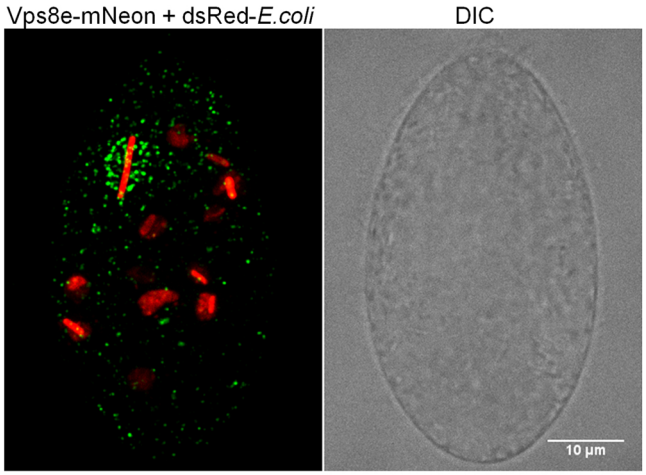

Vps8e-containing CORVET complex localizes to food vacuoles in *Tetrahymena thermophila*. Fluorescence image of a single *Tetrahymena thermophila* cell expressing the mNeon-tagged Vps8e subunit of the CORVET complex Vps8e-containing CORVET complex (8E-CC) (left panel), with a paired differential interference contrast (DIC) image (right panel).To visualize food vacuoles, *Tetrahymena* cells were fed with dsRed-expressing bacteria prior to imaging. The Vps8e–mNeon subunit, and consequently the 8E-CC complex, appear in numerous small puncta throughout the cell cytoplasm (green dots), with a small fraction concentrated around a food vacuole (labeled by red bacteria) near the cell anterior.

Have you had any significant mentors who have helped you beyond supervision in the lab? How was their guidance special?

During my PhD and postdoc, I met several inspiring scientists who fueled my passion and interest in science, but I can firmly assert that my number one mentor has been my postdoc supervisor, Prof. Aaron Turkewitz. Joining Aaron's lab, at the University of Chicago, has been a wise choice for my career. He is not only a brilliant scientist, but also an excellent teacher, who had a very important influence on my growth as a scientist. He taught me how to rigorously develop a research project by continuously stimulating my thinking with challenging questions, and by discussing the best experiments to find the answers to our biological problems. He joined my enthusiasm for a good results, and encouraged me in the tough moments. Most importantly, he trusted me and valued my judgment. He contributed to building up my self-confidence as a researcher, which I think is an important skill in order to become a proficient scientist. He is my role model in science.

What's next for you?

I will continue to be a researcher since working at the bench is what I like the most. Being able to manipulate nature in order to understand it is a privilege, and despite the tough moments, which are common to all jobs, it is absolutely rewarding. I recently joined Maryse Lebrun's lab (Laboratory of Pathogen Host Interactions), at the University of Montpellier, where I will apply my expertise and experience with free-living ciliates to advance the understanding of secretion in the related phylum Apicomplexa. By taking advantage of forward genetics, a tool well developed in *Tetrahymena*, I will be searching for genes important for regulated exocytosis in this ciliate, and looking for homologs in apicomplexan parasites, which, in turn, could be relevant for secretion during host cell invasion, a complex process still not fully understood.

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

When I was a kid I wanted to be an astronaut, I guess like almost everyone. I was fascinated by the mysteries of the universe, as evidence I still have plenty of magazines at home, for non-expert readers, related to this topic. Growing up I started reading more specific books on the subject: about planets, stars, galaxies, black holes… aliens! However, it was during high school that I dropped the idea to travel to the moon and beyond, and I fell in love with biology, deciding to pursue a career in science. I did not change my mind about the beauty of the universe, but I simply shifted my interests from the macroscopic to the microscopic world.

In addition to my science skills, an interesting thing about me is that, apparently, I am an extraordinary tiramisu and pizza maker, which I guess is not surprising since cooking is essentially an applied science!

Reference

Sparvoli, D., Zoltner, M., Cheng, C.-Y., Field, M. C. and Turkewitz, A. P. (2020). Diversification of CORVET tethers facilitates transport complexity in *Tetrahymena thermophila*. *J. Cell Sci.* **133**, jcs238659. doi:10.1242/jcs.238659