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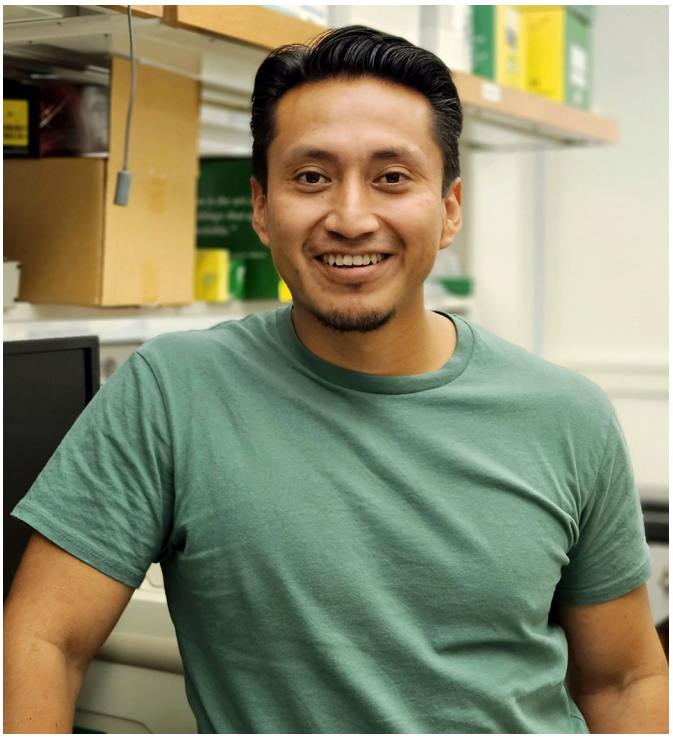
FIRST PERSON

# First person – David Anaguano

First Person is a series of interviews with the first authors of a selection of papers published in Journal of Cell Science, helping researchers promote themselves alongside their papers. David Anaguano is first author on 'Time-resolved proximity biotinylation implicates a porin protein in export of transmembrane malaria parasite effectors', published in JCS. David is a PhD student in the lab of Vasant Muralidharan at Center for Tropical Emerging Diseases, University of Georgia, Athens, GA, USA, investigating the mechanisms protozoan parasites utilize to maintain a successful infection within their hosts.



David Anaguano

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

Malaria remains a significant global health issue, with its most lethal and widely distributed causative agent being the apicomplexan parasite known as *Plasmodium falciparum*. Within the human host, *P. falciparum* parasites are capable of infecting and developing within erythrocytes. However, due to the unfriendly nature of these host cells for parasite growth, *P. falciparum* has evolved the capacity to extensively remodel the host cell by exporting their own proteins during the initial hours post invasion. The mechanisms underlying the transport of these exported proteins across multiple membranes are still not well understood. My work focused on investigating how membrane-associated proteins traverse the parasite plasma membrane, a process that occurs within a narrow time frame. I successfully developed an assay to identify interacting partners during the transport of a membrane bait protein across the parasite plasma membrane. I found several proteins that could potentially serve as exporting partners, including some that were previously known to play a role in protein export. To validate these findings, I applied several types of microscopy to confirm the localization of both proteins within the periphery of the parasite during early stages of invasion. Our results strongly support the applicability of our assay to identify protein interactors within time-sensitive biological scenarios.

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

Given that my primary objective was to capture parasites when the bait protein was at a specific localization during its transport, the most technical challenge I faced was synchronizing the parasites within a small window of time and maintaining this synchronization across the different sample collections and replicates. To address this challenge, I carried out a meticulous standardization of the initial assay and carefully planned out sample collection times. Another key challenge was that conventional immunofluorescence microscopy could not offer the resolution I needed to discern the precise localization of the protein candidates. To overcome this limitation, I had the valuable opportunity to learn ultrastructure expansion microscopy from Dr Sabrina Absalon at Indiana University School of Medicine, a pioneer in this technique for malaria parasites. Implementing this innovative technique significantly improved the robustness of my results.

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

After dedicating so much work to collecting the samples and creating the different cell lines, I experienced an overwhelming sensation of relief the moment I first observed one of my candidate proteins localizing to the parasite periphery. Coincidentally, this finding occurred right before I left for my Christmas vacation, which made my visit to my family back home an awesome time.

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

I chose the Journal of Cell Science because of its reputation in the field of cell biology, where I wanted my work to be highlighted.

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

My mentors have played a crucial role in shaping both my professional and personal growth throughout my entire career as a researcher. I consider myself fortunate to have had mentors who prioritize mental health and personal time, recognizing their importance in ensuring success during graduate school. One quote from my undergraduate mentor has had a profound impact on me: "You can only be successful if you help others to be successful". I aspire to carry this forward and support others when I have the opportunity to mentor.

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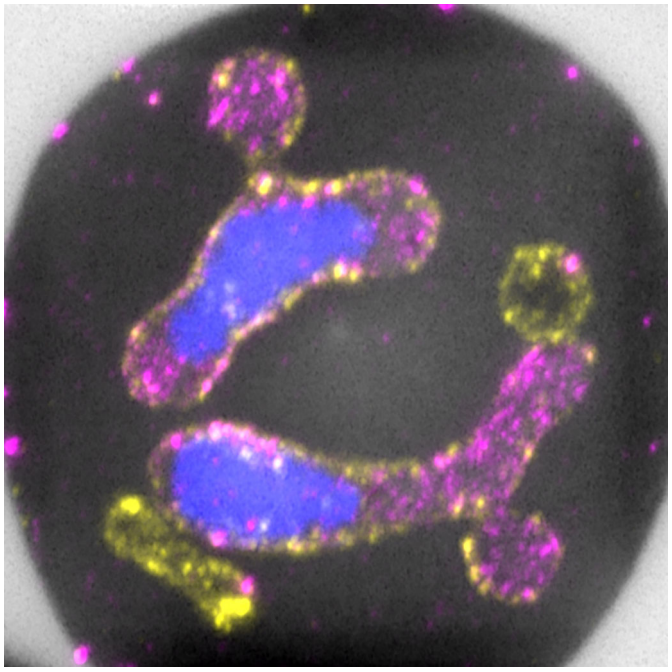

**GAPM1 localizes to the host-parasite interface.** GAPM1$^{mNG\text{-}apt}$ parasites showing the localization of GAPM1 in early-ring stages (4 hpi) using U-ExM. Sample was stained with NHS-Ester (greyscale), anti-HA (magenta), anti-RAP1 (yellow) and Sytox (blue). Selected *Z*-stack images were projected as a combined single image.

### 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 been interested in biology since I was a child and learnt about DNA at school. My mom often reminds me how we used to talk about DNA and that I would mention I wanted to study it one day. However, it was not until my final year as an undergraduate student that I decided to pursue a career focused on the biology of parasites. During that time, I was working on my undergraduate project, which involved collecting vectors of Leishmaniasis in an endemic region of my home country, Ecuador. This experience allowed me to witness first-hand the significant impact of this neglected parasitic disease on the lives of people in rural areas where access to public health services is limited or non-existent. After this experience, I was motivated to apply for a Fulbright scholarship, with the goal of pursuing graduate studies in the US and starting my career as a researcher.

### What's next for you?

I am still fascinated by protozoan parasites and their unusual ways of surviving within human hosts, so I will be moving from the US to Europe to start a postdoc working on a different aspect of the biology of malaria parasites.

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

When I am not at my bench trying to make experiments work, I am either playing soccer, working out, or sharing a beer with friends. I consider personal time and activities outside the lab work to be essential to help students navigate through the difficulties that graduate school can present.

#### Reference

**Anaguano, D., Dedkhad, W., Brooks, C. F., Cobb, D. W. and Muralidharan, V.** (2023). Time-resolved proximity biotinylation implicates a porin protein in export of transmembrane malaria parasite effectors. *J. Cell Sci.* **136**, jcs260506. doi:10.1242/jcs.260506