CELL AND MOLECULAR BIOLOGY STUDENT NEWSLETTER

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IN THIS ISSUE

LETTER FROM THE EDITORS

Dear CAMB students, faculty, and alumni,

In this issue of the CAMB Student Newsletter, we highlight recent works by CAMB G&E graduate Philipp Mews and CAMB DSRB graduate Iryna Shakhmantsir. We also sit down with Dr. Cesar de la Fuente, a new faculty member in the Departments of Microbiology, Bioengineering, and Psychiatry, and Derek Sung, a CPM MD/PhD student using Instagram to showcase his research. Finally, we share a BGS-inspired recipe for stuffed peppers.

For additional articles, past publications, and to learn more about the CAMB Student Newsletter team, visit our blog at cambnewsletter.wix.com/blog. Current students interested in contributing to the CAMB Student Newsletter can contact us at camb.studentnews@gmail.com. We hope you enjoy the February 2020 Issue!

Sincerely,
Somdutta Mukherjee and Sylvia Stankov

RESEARCH SPOTLIGHT

Ethanol-derived acetate modifies the brain epigenetic landscape

Aishwarya Pawar

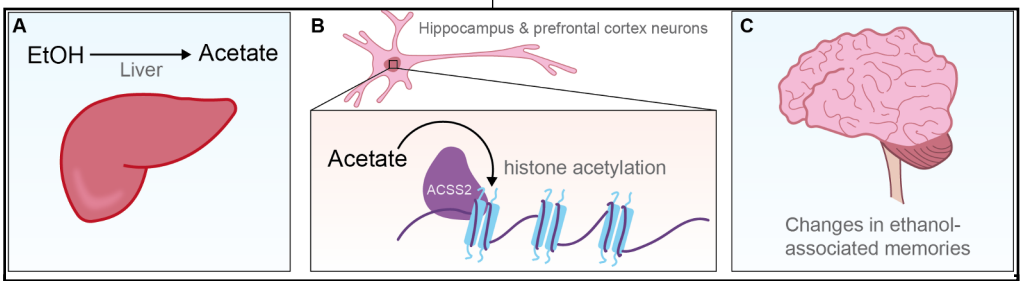
The physiological effects of alcohol on the brain and motor function have been studied extensively for decades. In low doses, alcohol causes euphoria and reduces anxiety, while in higher doses it impairs cognition and balance, and increases response time. Long-term consumption of alcohol is associated with changes in memory and alcohol-related learning, which play important roles in the development of alcohol use disorder. Dr. Philipp Mews, a CAMB graduate from the Genetics and Gene Regulation (now Genetics and Epigenetics) subprogram, explored the link between alcohol metabolism, gene expression, and alcohol-related learning behaviors, and recently published his findings in *Nature*.

Alcohol metabolism begins in the liver when ethanol is broken down into acetate, leading to a rapid increase in blood acetate levels. In a previous study, Mews demonstrated that acetyl- CoA synthase 2 (ACSS2), the metabolic enzyme which converts acetate to acetyl-CoA, is highly expressed in the learning center of the brain - the hippocampus - and is bound to neuronal chromatin to 'fuel' the modification of histone proteins with acetyl groups (Mews et al., *Nature* 2017). The hippocampus is a small, yet complex structure embedded deep in the brain and is associated with memory and learning. Therefore, histone modification changes in this region could affect alcohol-related learning. In their most recent study, Mews and colleagues explored how the rapid rise in blood acetate after ethanol consumption is translated into an immediate increase in acetylation of histones in the hippocampal region of the brain. Using a mouse model, they discovered that this epigenetic modification is facilitated by ACSS2 and affects alcohol-associated learning behaviors.

Dr. Mews, who did his thesis work in Dr. Shelley Berger's lab, conceived the idea for this project in 2013 when he started working on ACSS2. It developed incrementally over the years as a side-project

with help from other co-authors and collaborators. By the time the writing and review process began, Dr. Mews had already taken up his current position as a postdoctoral fellow at the Icahn School of Medicine at Mount Sinai in Dr. Eric Nestler's lab. With regular commutes between Philly and New York on the weekends, thus began a year-long long-distance 'relationship'.

One of the primary techniques utilized by Mews in the study was *in vivo* stable-isotope labeling followed by mass spectrometry. Intraperitoneal injections of labeled ethanol in mice led to the incorporation of the label in the acetyl groups on histones in the hippocampus and prefrontal cortex of the brain. This process was extremely dynamic and detected within just minutes. A knockdown of *Acss2* in the dorsal hippocampus prevented this incorporation, leading Mews and colleagues to conclude that ACSS2 mediated this acetylation of histones in the brain, exploiting the increased levels of blood acetate. A set of *in vivo* and *ex vivo* RNA-seq experiments showed that gene targets for ACSS2 belonged to pathways in neuronal plasticity, as well as ribosomal and mitochondrial functions.



A. The liver metabolizes the ethanol in alcohol to acetate. B. Acetyl CoA synthase 2 (ACSS2) transfers the acetate onto histones in neurons in the hippocampus and prefrontal cortex. C. These histone modifications lead to changes in ethanol-associated memories in mice.

Another exciting experiment done by the group was an ethanol-mediated behavioral study utilizing a method known as conditioned place preference on *Acss2* wild-type and knockdown mice. After prior conditioning to ethanol, wild-type mice showed a higher preference for the spatial compartment with ethanol in comparison to *Acss2*-knockdown mice. These results showed how ACSS2, a metabolic enzyme, opens up the epigenetic landscape for alcohol addiction in the brain.

Lastly, the authors briefly explored how maternal alcohol use affects fetal development during pregnancy. Mass spectrometry on fetal brains at E18.5 revealed that exposure to binge-like drinking in mothers resulted in the deposition of ethanol-derived acetyl groups in the fetal mid- and forebrain. This finding potentiates the inheritance of alcohol-related memory and learning behavior.

Dr. Philipp Mews's work helps bridge the knowledge gap between alcohol metabolism and alcohol-related learning behavior, and it unveils the metabolic enzyme ACSS2 as a potential therapeutic candidate for alcohol abuse and addiction disorders. His current work at Mount Sinai examines how drugs of abuse cause long-lasting changes in chromatin in a key brain region of reward learning. Eventually, he wants his own independent research lab to work on epigenetic dysregulation in psychiatric and substance use disorders.

Scientists in the field of neuro-epigenetics want to understand the role epigenetic regulation plays in brain function in mental health and related disorders. According to Dr. Mews, "the hope is to modulate brain chromatin in modern psychiatry, where psychosocial therapy, in combination with pharmacological treatment, may facilitate epigenetic reprogramming to alleviate symptoms and support disease recovery." His latest work has brought us a step closer to understanding addiction and finding more precise and druggable targets to combat it.

RNA splicing factor mutations that cause retinitis pigmentosa result in circadian dysregulation

Sylvia Stankov

Iryna Shakhmantsir, a recent DSRB alumna from Dr. Amita Sehgal's lab, published her work on retinitis pigmentosa (RP) mutations and circadian rhythms. While studying pre-mRNA processing factors, she noticed that many of the splicing factors that influence circadian timing in fruit flies were also implicated in RP. Iryna's work demonstrates that mice harboring mutations in a splicing factor have longer wheel-running activity and dampened circadian oscillations of critical transcripts. She hopes that her paper can provide a springboard for new studies in both the RP and circadian fields, and wonders whether circadian disruption simply correlates with some forms of RP or is a contributing factor to this debilitating age-progressive disease.

Read about Iryna's full work in the *Journal of Biological Rhythms*.

Iryna Shakhmantsir, PhD

Cesar de la Fuente

James Gesualdi

***I** recently sat down with Cesar de la Fuente, a newly appointed Assistant Professor in the Departments of Microbiology, Bioengineering, and Psychiatry. Cesar leads a diverse group interested in using multiple approaches, including* in-silico *models and synthetic biology, to address global health challenges. Below is a paraphrased transcript of our discussion regarding his time at Penn, goals for the future, and advice for current trainees.*

JG: Could you describe some of the projects in your lab?

CF: Our main goal is to develop new molecular tools and medicines using computers. We are currently focusing on developing new antibiotics to help address the global health challenge of antibiotic resistance, which exerts a massive toll on public health. In the US alone, 35,000 people every year – or one person every 15 minutes – die due to antibiotic resistant infections, and it is likely that this issue will only become more pressing in the future.

For several decades, the scientific community has been unable to effectively develop new antibiotics or discover effective natural products like penicillin. Therefore, our main approach is to use computers to extrapolate from what nature has provided to create new synthetic antibiotic molecules with increased efficacy against drug resistant infections. Ultimately, our ambition is to develop computational tools that are capable of building new molecules for various diseases that are resistant to treatment.

Cesar de la Fuente, PhD

JG: Why do you think there is a lack of development of new antibiotics?

CF: It's a complex and multi-layered problem. Part of the issue is that people thought the problem was solved after the discovery of penicillin and so innovation in these fields had slowed down for many years. Another issue is that antibiotic development is not a profitable business model, and so large pharmaceutical companies have stepped away from this space. Because the lack of profit is a hindrance here, I think the solution will have to involve steps to make profit irrelevant. There are efforts to involve governments and non-profit organizations to address these issues. Ultimately, there needs to be incentives to draw researchers toward these problems. Hopefully, funding from various organizations can be used specifically to help address the growing issue of antibiotic resistance.

JG: How would you describe your mentorship style?

CF: I would say hands-off for the most part. I truly value the time of trainees in my lab and their devotion to our studies. I encourage them to be as creative as possible and try to find projects that they are highly passionate about. I also try to create a lab environment in which folks are generally nice and can get along with each other.

JG: How do you go about working to create a comfortable environment within your lab?

CF: Yeah, that's a very difficult question. I generally try to recruit people that are personable and who enjoy talking about science. I try to recruit individuals that are highly qualified, but not necessarily in the sense of having a spotless academic record. I think it is important to recruit individuals with non-traditional academic backgrounds as well as members of groups that are underrepresented in the scientific community. Doing so can bring different perspectives to the lab which is key to tackling the complex questions we are interested in. This approach has helped us to build a highly diverse group with trainees from all over the world. I also try to create a culture within the lab that is free from hierarchy: I don't consider myself superior to the people that work in my lab in any way. The main difference is that I just happen to have a bit more experience and a different title. I think that helps to encourage people to be comfortable in their workplace. My ultimate goal is to build an exciting environment in which individuals can find what they are truly passionate about.

JG: Do you have any advice for first-year graduate students who are looking to find the lab that's the right fit for them?

CF: It's key to explore different fields early in your training and to not necessarily get stuck on one idea. It's important to get exposed to different ways of thinking in order to truly explore what you are most interested in. There is always a number of possible projects that we have not thought about and perhaps that is where your passion lies. It is also critical to put a lot of thought into your relationships with potential mentors. A good relationship with your advisor is essential, and you should make sure that you can count on them to help you navigate your academic environment. I think it is also important to become accustomed to the fact that learning in science will involve performing a lot of experiments that won't necessarily go how you hoped they would. I think students need to view things like that as part of the process of working toward an evolving hypothesis or a way of tackling a difficult problem, rather than as failures or setbacks. More than failed experiments, I consider these to be important parts of the learning process.

JG: What do you think is the best way for trainees to seek out mentors that they can develop a healthy rapport with?

CF: I would encourage folks to do their best to avoid being shy; set up meetings and interviews with potential mentors that you might be interested in. At this step it is important to believe in yourself and be

confident in your ability to seek out a great mentor. I would also stress the necessity of finding a mentor that will be honest, and one that you can feel comfortable being honest with. A PhD is a major commitment, and it's important to place yourself in an environment in which you are not under unnecessary stress and where you can learn and thrive. A PhD is a special time, as you have the very privileged opportunity of thinking about a problem or set of problems for a long period of time.

JG: What activities do you think new faculty in the life sciences should pursue to help cultivate ideal mentor-mentee relationships?

CF: I think meetings and seminars to encourage discussion and collaboration are critical. Interactions between students and professors, particularly in less formal settings, are important for helping students to become comfortable with their colleagues. It's also important for us as faculty to embrace criticism from trainees: learning is a non-linear process for all of us and it's important to incorporate ideas from diverse perspectives.

JG: What drew you to Penn when you were searching for faculty positions?

CF: I think Penn has a fantastic community, particularly because it places such great value on collaboration and interdisciplinary research. There's also just such a high caliber of faculty here in general, and that leads to the great projects that are always ongoing. The other advantage is that the medical school, engineering school, and undergrad campus are all integrated in a relatively small space. This tends to encourage collaboration and cross-disciplinary studies, which aligns very well with my research interests. So that was one of the key aspects for me. And, of course, Philly is a great city.

JG: Where did you study before your appointment at Penn?

CF: I did my undergraduate studies and master's in biotechnology/bioengineering in Spain, then I did my PhD in microbiology and immunology at the University of British Columbia in Vancouver. Then I went to MIT for a post-doc to expand my knowledge of synthetic biology and computer science. After that, I was hired here. Now my lab works to incorporate concepts from microbiology, synthetic biology, and computer science into our research approach.

JG: Would you say that traveling so far from home for your training added any extra hurdles to your academic journey?

CF: Well, whenever you transition to a new country there are challenges, particularly becoming familiar with a new language, which is especially critical for interviews and that sort of thing. That's certainly a barrier that one needs to overcome, but I think that a good mentor can be especially helpful for dealing with issues like that. That's another example of where I think good mentorship is critical. The way I look at it, every hurdle or challenge is part of the learning process.

JG: What do you think of Philadelphia?

CF: I live in Center City; I think there's a great energy and culture and diversity here. Philadelphia has a lot of heart and passion, which I think are important components of any community.

JG: What are your hobbies outside of the lab?

CF: I play soccer. I was an assistant coach and player at MIT FC when I was there. I am playing here as well but I don't get to play quite as much as I did during my post-doc. I try my best though. I also love to bike, which I think is the best way to get around the city. (*Ed: the author whole-heartedly agrees).* I also love cinema and really enjoy going to movies and concerts.

JG: What's the best movie you've seen this year?

CF: The Irishman. It's worth the time commitment and the story is based in Philly. Joker was also fantastic.

Learn more about Cesar 's work :
https://www.youtube.com/watch?v=xKP9sSMiMqI.
Any interested trainees can contact him at *cfuente@pennmedicine.upenn.edu*.

SPECIAL INTEREST

Derek Sung: @immunofluorescence

Somdutta Mukherjee

Derek Sung, a third year CPM MD/PhD student in Mark Kahn's lab, has found a unique way to showcase his appreciation for how beautiful cells can be - he runs the popular Instagram account, @Immunofluorescence. Derek utilizes immunofluorescence and confocal microscopy to study vascular development during hematopoiesis and in the placenta. He was first introduced to these techniques while working at the Laboratory of Molecular Cardiology at the National Heart, Lung, and Blood Institute at the NIH, and was particularly drawn to its inherent artistry. "Knowing the nuances in staining and imaging makes it as much of an art as a science," he says. Additionally, there are many different ways to analyze a single image and get a lot of data at once, making the techniques both pretty and productive at the same time. Perhaps unsurprisingly, his favorite thing to image is the heart, since that is what he used to study. Derek often immunostains whole mouse embryos, and once he collects what he needs, he looks for anything else that might be interesting. Through these techniques, he has come to appreciate the striking nature of cellular structures such as the cytoskeleton, as well as many different cell types and tissues.

Scrolling through Derek's Instagram page, it is clear that he has not

only mastered the art of immunofluorescence, but confocal microscopy as well. Derek started his account in April of 2017 and, over time, gained a few thousand followers. One day, he received a message from Jerry Fagerberg, manager of the Cell Press Instagram account. That opportunity led to a feature of his work in the Cell Picture Show and an interview for *CrossTalk*, the official blog of Cell Press. Now, Derek has 13K Instagram followers and counting.

Derek Sung, 3rd year MD/PhD student, CPM

A lot goes in to running a successful Instagram account. Over the years, Derek has learned that it's important to post consistently, know your audience, learn when the best times to post are, and write clear and informative captions. This last point is especially important, as the goal of Derek's Instagram page is to not only share his incredible images, but also to help people appreciate science. There are many misconceptions about medicine and science; Derek uses the captions for each post as a way to offer the perspective of a medical student, and help humanize the stories the pictures tell. "People who provide care are also human, and find stresses in their work. As a medical student, a lot of things are first time experiences," Derek notes. From a scientific standpoint, Derek's goal is to communicate the content with less technical jargon, and make science accessible to people of all background levels of understanding.

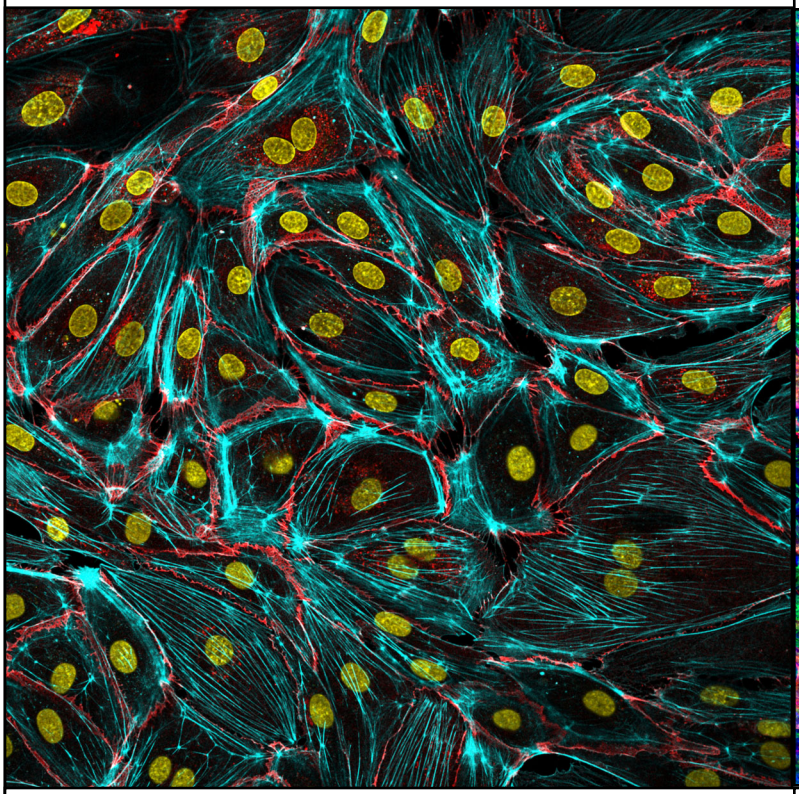
Running a popular Instagram account while being a busy MD/PhD student can be tricky to balance, but Derek doesn't consider managing the account work. He sees it as a fun opportunity to teach people, and to share his research and what he knows. In addition to maintaining @Immunofluorescence, Derek started a podcast called "Beyond the Abstract" with fellow MD/PhD student Ellen White. Joined by various experts in the field, Derek and Ellen discuss basic science papers in a way that anyone can understand. Their podcast highlights the importance of basic science, and aims to show that it is just as important as translational research.

Presenting science in a clear and effective way in order to make it accessible to everyone has become a passion for Derek. He hopes that with his Instagram page and his podcast, he can reach a wider audience. When asked if there is anything he has not stained or imaged yet but would like to in the future, he answered immediately, "The brain. I just haven't come across it in my work. Maybe one day." Wherever he takes his talents next, the results are sure to be illuminating.

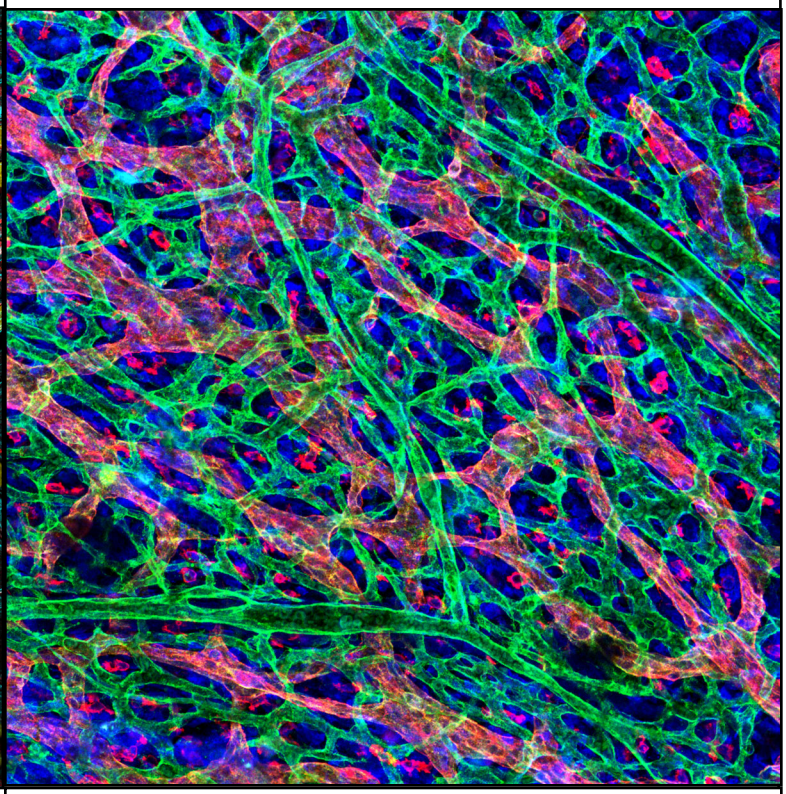
You can follow Derek on Instagram @Immunofluorescence

"Beyond the Abstract" can be found on Spotify, Apple Podcasts, Google Play, or https://beyondtheabstract.captivate.fm/

You can read Derek's interview for *CrossTalk* at http://crosstalk.cell.com/blog/meet-derek-sung



Lymphatic endothelial cells stained for VE-cadherin (red), actin (cyan), and nuclei (yellow). Image courtesy of Derek Sung



Embryonic skin stained for lymphatic (red) and blood (green) vasculature. A cell-cell adhesion protein is stained in blue. Image courtesy of Derek Sung

Recipe

TITLE: B.G.S. (broke grad student) STUFFED PEPPERS

PREP. TIME: Take your time... TOTAL TIME: Don't worry about it SERVES: YOU for 1 week!

INGREDIENTS:

- 1 lb. ground beef
- 1 cup onion, chopped
- 5-6 cloves garlic, chopped
- Salt & pepper
- Chili powder
- 2 cans tomato soup
- ½ lb. cheese
- 1 ½ cup cooked rice
- 8 green peppers

DIRECTIONS:

1. Sauté onion and garlic in skillet, add ground beef and cook until browed. Add your favorite seasonings and tomato soup. Simmer for 10 minutes, covered.
2. Add cheese. Cook on low heat, stirring occasionally until cheese melts. Stir in rice and cool.
3. Cut peppers in half, lengthwise (remove seeds). Boil salted water and cook until barely tender, ~3 minutes. Immediately afterwards, drain water and separate peppers to cool.
4. Stuff peppers with rice mixture. Garnish with cheese.
5. To save leftovers, wrap peppers individually and store in freezer. Reheat at 400 °C for 30 – 40 minutes. Enjoy!

▻ Send us your favorite recipes to be featured in the next CAMB Newsletter!

CAMB Student Newsletter Staff February 2020