CELL AND MOLECULAR BIOLOGY STUDENT NEWSLETTER

Volume 4, Issue 2
June 2019

IN THIS ISSUE

LETTER FROM THE EDITORS

Dear CAMB students, faculty, and alumni,
Summer is here! In this issue of the CAMB Student Newsletter, we highlight the Pie a P.I. event held in March. Thank you to all who donated to the Philadelphia Education Fund's Donald McKinney Center for STEM Education. We raised money and had a good time de-stressing by throwing whipped cream in P.I.s' faces. Thanks again to the P.I.s for being such good sports!
In this issue we also discuss the pros and cons of companies that offer genetic testing services. We spoke with Priya Chatterji (Genetics & Epigenetics) about her research investigating the role of RNA-binding proteins in the progression of colorectal cancer. Additionally, we catch up with Developmental, Stem Cell, and Regenerative Biology alumnus Michael Convente about his position as a medical writer for Scientific Pathways. Finally, test your knowledge of science in pop culture with a fun crossword that rounds out our Summer issue!
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 June 2019 Issue!
Sincerely,
Lexy Stanley and Somdutta Mukherjee
Editors-in-Chief

RESEARCH SPOTLIGHT

The unique LIN28B-IMP1 regulatory axis: A double edged sword

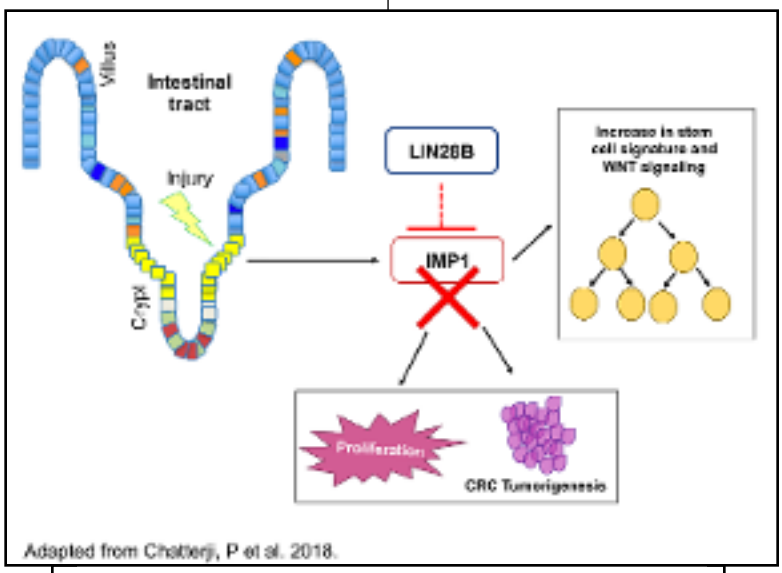
Julianne Davis

Although new cases of colorectal cancer (CRC) are on the decline, CRC is amongst the top four most commonly diagnosed cancers in the United States according to the latest NCI SEER report. Underlying risk factors such as smoking, obesity, and family genetic history can lead to malignant transformation of intestinal epithelial cells. At the molecular level, mutations in the genes APC, LKB1, MLH2, and MYH have been associated with colorectal tumor initiation or progression. However, regulation at the transcriptional-translational interface by RNA-binding proteins (RBPs) has not been well characterized within CRC. To clarify the role of the RBPs LIN28B and IMP1 in intestinal cell differentiation, regeneration, and malignant transformation, CAMB alumna Dr. Priya Chatterji and colleagues from the laboratory of Anil Rustgi recently defined a novel signaling axis between LIN28B, IMP1, and the downstream WNT signaling pathway.

LIN28B is a master transcriptional regulator that suppresses let-7 miRNA. Subsequently, this suppression increases the transcription of of let-7 miRNA targets including IMP1. Previous members from the Rustgi laboratory established that IMP1 acts as an oncogene

Priya Chatterji, G&E

in IMP1-overexpression CRC xenograft models. Priya encountered an early obstacle when determining the LIN28B regulated translatome and deciphering downstream targets of LIN28B unique to intestinal epithelium repair and colorectal tumor initiation. Priya stated that “since both LIN28B and IMP1 are RNA binding proteins that bind thousands of targets, it was important to figure out how to focus my study. My collaboration with Dr. Premal Shah at Rutgers to do the ribosome profiling really guided my study.” Priya performed ribosomal profiling in SW480 CRC cells and observed that IMP1 was the most notably enriched post translational regulatory element when LIN28B was overexpressed. Subsequent CRISPR/Cas9 deletion of IMP1 in two CRC cell lines followed by RNA-seq and gene set enrichment analysis (GSEA) led to the discovery that IMP1 was a significant translational regulator of pathways associated with colorectal cancer, in particular the WNT signaling pathway.



The LIN28B-IMP1 signaling axis in the intestinal epithelium. LIN28B overexpression with IMP1 loss leads to tumor progression in colorectal tumor cells and rapid expansion of intestinal stem cells. (Adapted from Chatterji, P et al. 2018)

Priya first examined clinical datasets to search for correlations between LIN28B and IMP1 in CRC patients. She found that LIN28B and IMP1 transcript levels positively correlated when interrogating colorectal cancer patient RNA- seq dataset from The Cancer Genome Atlas database. Additionally, previous studies implicate IMP1 as an oncogene and driver of tumor progression in CRC, independent of LIN28B expression. Using intestine-specific LIN28B overexpression genetic murine models or LIN28B overexpression xenograft murine models, Priya manipulated IMP1 expression to characterize IMP1 regulation on cell differentiation and CRC tumor growth properties. She found that regardless of IMP1 protein expression level, tumor volume decreased in mice injected with LIN28B overexpressing colorectal cancer cells. On the contrary, mice with intestinal-specific LIN28B overexpression acquired multiple tumors and increased counts of proliferating cells in normal adjacent intestinal tissue with IMP1 loss. Notably, there was no tumor formation in genetic models of IMP1 loss alone. However, the combination of LIN28B overexpression with IMP1 loss led to undifferentiated tumors. These data imply that IMP1 acts as an inhibitor in colorectal tumor initiation when LIN28B is overexpressed within the intestine.

Building upon the inhibitory role of IMP1 in CRC tumor initiation, Priya hypothesized that IMP1 may regulate cell stemness and epithelial regeneration within normal intestinal epithelia. Intestinal epithelial-specific deletion of IMP1 led to the regeneration of crypt cells in mice following whole body irradiation - a stressor used to ablate most intestinal cell types. To further examine the stem cell compartment effected by IMP1 loss, Priya interrogated the mRNA expression of stem cell-related genes including WNT target genes and found an increase in mRNA expression of these genes. Furthermore, beta-catenin, a protein important for nuclear transcription of WNT target genes, was more abundant in the nucleus of LIN28B-overexpressing cells, and enriched for genes that encompass the WNT signaling pathway. Additionally, GSEA performed on RNA-seq of IMP1 wildtype or IMP1 deleted intestinal stem cell tissue disclosed a WNT target gene enrichment signature in the absence of IMP1. These observations led to the hypothesis that IMP1 impacts downstream canonical WNT targets in the intestine.

To complement her hypothesis, Priya utilized an IMP1 overexpression model to see if IMP1 overexpression had an opposite effect on tumor growth or intestinal regeneration in the presence of LIN28B overexpression. Priya recounts that, “it was necessary to test my hypothesis out in multiple systems both in vitro, in vivo and ex vivo.” IMP1 overexpression resulted in no tumor formation irrespective of LIN28B overexpression status, suggesting IMP1 expression by itself was not enough to generate tumors. When comparing LIN28B overexpression/IMP1 deletion to a LIN28B/IMP1 double overexpression model, the double overexpression model showed a decrease in WNT target mRNA expression. This finding suggested that IMP1 was a switch for WNT signaling and IMP1 expression may be a result of another upstream protein involved in tumor initiation.

Taken together, Priya’s findings illustrate a more intricate signaling network between LIN28B, IMP1, and WNT downstream targets. The presence, absence, or overexpression of LIN28B defines the effects of IMP1 expression on tumor burden, differentiation potential, and epithelial cell repair within the colorectal tumor environment or normal intestinal ecosystem. IMP1 loss promotes expansion of the intestinal stem cell compartment and augments LIN28B-mediated colorectal tumor progression. Ongoing efforts from Priya and other members of the Rustgi lab to further investigate IMP1 expression in colonic epithelial repair resulted in the additional European Molecular Biology Organization Reports publication, “Posttranscriptional regulation of colonic epithelial repair by RNA binding protein IMP1/IGF2BP1" published in late March. Using conventional therapeutic approaches to target signaling pathways in CRC is difficult due to toxicity in normal intestinal epithelium. This study suggests that inducing IMP1 may be a beneficial therapeutic strategy for a subset of CRC cases in which LIN28B is overexpressed.

Chatterji, P, Hamilton, K E *et* al., *Genes & Development*, 2018

Pie a P.I. Day

Felicia Peng

March holds a beloved holiday for nerds all around the world: Pi Day. The classic way to celebrate this holiday is through feasting on pie, but here in CAMB we put a twist on this tradition. For the third year in a row, the CAMB Student Newsletter team hosted its annual "Pie a P.I." event on Thursday, March 14. As the name suggests, this event involves gathering round and watching CAMB faculty members get pies thrown in their faces. In the days leading up to Pi Day, those wanting to pie a P.I. purchased $1 raffle tickets to enter for the chance to be one of the lucky pie throwers. The brave targets this year were CAMB faculty members Mary Mullins (DSRB), Jean Bennett (GTV), Joe Baur (CPM), Ben Prosser (CPM), Chris Lengner (DSRB), and Dan Kessler (chair-CAMB).

While "Pie a P.I." is a great way for students and post docs to have fun at the expense of faculty members, at the heart of the event is a good cause. This year the money raised from the raffle is benefitting the Philadelphia Education Fund's Donald McKinney Center for STEM Education , which was formerly known as STEM Initiatives. The revamping of STEM Initiatives is in honor of Donald McKinney, who was a chemistry teacher and founder of the Philadelphia Education Fund's Math and Science Coalition. In keeping with his legacy of improving Philadelphia's public education system, the McKinney Center seeks to provide K-12 students with strong educational programs in the math and sciences. To achieve this goal, educators are provided access to STEM professional development opportunities and resources, which should ultimately strengthen their STEM curriculum. Furthermore, the McKinney Center's Philadelphia STEM Ecosystem establishes collaborations between schools, nonprofit organizations, businesses, and government agencies that aim to increase accessibility to quality STEM education. By increasing the standard of education and the number of STEM opportunities students are exposed to, the McKinney Center envisions that "all Philadelphia students will graduate from high school with strong [STEM] knowledge and skills [that will enable] them to succeed in a 21st century workforce and society."

DUTCH APPLE PIE

INGREDIENTS

5 large Granny Smith apples
½ cup white sugar
2 tbsp all-purpose flour
½ tsp ground cinnamon
2 tbsp lemon juice
½ cup white sugar
½ cup all-purpose flour
½ cup butter
1 pie crust of choice (9 inches)

DIRECTIONS

Preheat oven to 425 degrees F. Peel, core, and slice apples. Combine ½ cup sugar, 2 tbsp flour, and cinnamon. Pour mixture over apples in pie crust, and sprinkle lemon juice on top. Cut ½ cup sugar, ½ cup flour, and ½ cup butter together, and top pie with mixture. Enclose pie with two 15 inch pieces of parchment paper, folding edges up 3 times. Bake pie for 1 hour. Remove pie from oven, split parchment paper open, and cool pie on a wire rack.

Faculty from left to right (top photo): Dan Kessler, Chris Lengner, Joe Baur, Jean Bennett, Ben Prosser, Mary Mullins.

First-time participants Baur and Prosser were eager to get involved with "Pie a P.I." When asked why he volunteered to have pie thrown in his face, Baur reflected that "it seemed like a nice opportunity to help raise money for a good cause." Prosser shared similar sentiments, and also noted that his participation in the event boosted lab morale. He remarked, "[We] P.I.s can sometimes be tough on our trainees. I imagine that it was a bit of healthy catharsis for my own trainees to see me take a pie in the face. I could sense this in their anticipatory smiles and devious grins throughout the day prior to my pie-ing." The only complaints Baur and Prosser had were that the pies were not as real or delicious as they had anticipated, though it is unclear if their requests for blueberry and pecan pie, respectively, will be met in future years.

Whether you support "Pie a P.I." for its contribution to STEM education or its humbling of P.I.s, the CAMB Student Newsletter team would like to extend a thanks to all the donors, faculty, and volunteers that participated in the event. As a result of your support, we were able to continue to make this event a great success and raise money for the Donald McKinney Center for STEM Education.

More information on the McKinney Center for STEM Education at www.philaedfund.org/programs/advancing-education/mckinney-center

The Future of Genealogy Services

James Gesualdi

Over the last decade, DNA sequencing technologies have become more accessible, and a number of companies offering to unlock the secrets of our genome have become increasingly popular. These companies market themselves as tools for learning about ancestry and potentially connecting with distant relatives by analyzing their customers' genomes.

Example of a DNA Relatives Map generated by 23&Me. This map shows the individual the locations of all the 23&Me members whose submitted DNA is a close match to theirs.

DNA extracted from saliva samples sent in by patrons is sequenced, and loci that are highly variable among different ethnic groups are compared to corresponding loci in existing libraries. The largest of these companies, Ancestry.com and 23&Me, have user libraries containing the genomes of over 15 million individuals (1). The software then determines the population to which the customer's input is most likely to correspond to and compiles a genealogy report (2). For example, if 35% of the analyzed loci from a customer's genome correspond to the standing library of Sub-Saharan African sequences, the report will tell the customer that they are 35% Sub-Saharan African. Of course, a person's genome - and more so a library of several million people's genomes - contains much more interesting information than an elaborate family tree.

Genome analysis can also detect sequence variants that are associated with genetic disorders, and provide other health related information. 23&Me offers this service in one of its packages that includes analysis of one's predisposition to diseases in addition to ethnic background. Using information about their customers' health also allows for discovery of new disease associated variants; data from 23&Me's library has been mined in over 100 publications since the company's inception in 2006 (3). Furthermore, 23&Me recently announced a partnership with pharmaceutical giant GlaxoSmithKline (GSK). It is unclear what this collaboration may yield, but it seems that GSK will use disease associated variations discovered by 23&Me to develop new therapies. GSK may also be interested in contacting 23&Me customers for later clinical trials in an attempt to expedite the normally slow developmental process for therapeutics (3).

Genealogy services have also made headlines recently for helping to close homicide investigations, the most notable of which is the case of The Golden State Killer. To confirm that suspect Joseph James DeAngelo was indeed the Golden State Killer, police uploaded DNA gathered from multiple crime scenes to the open source library of GEDmatch, another genealogy company. They found that crime scene DNA had partial matches to the DNA of DeAngelo's relatives who had used GEDmatch's services in the past. These partial matches eventually led police to DeAngelo, who was apprehended in 2018 after confirmation that DNA he left behind at a restaurant matched the crime scene isolates (4, 5).

The success of this methodology has led to a partnership between the FBI and the genealogy company FamilyTreeDNA (5). This collaboration will grant the FBI access to nearly 2 million genetic profiles, effectively doubling the amount of genomic data they previously had access to through open source libraries (5). Allegedly, the FBI will not have totally unrestricted access to FamilyTreeDNA's archives, but will be able to upload crime-scene samples to their database and search matches. This has engendered apprehension among the company's customers, as there is now a possibility that some of their relatives may be caught up in a criminal investigation. In fact, FamilyTreeDNA has been stricken from a list of genealogy companies adhering to a set of voluntary privacy guidelines maintained by the Future of Privacy Forum, whose president called the deal "deeply flawed" and "out of line with industry best practices and with consumer expectations" (5). Obviously, the idea of sharing such intimate information stirs up controversy, and 23&Me's collaboration with GSK is no exception.

As stated on their website, 23&Me reserves the right to use their customers' genomic information to sell them products or services. Given their nascent partnership with GSK, one can imagine that genealogy reports may soon come with information about predisposition to certain conditions and advertisements for the latest GSK products available to treat those conditions. 23&Me's privacy policy assures customers that it will never share personal genetic information without an individual's explicit consent, but some people worry that this protection is only promised in theory and will not be implemented in practice. In an interview with Live Science, Tiffany Li, a fellow at Yale Law School's Information Society Project, said that this agreement is buried in 23&Me's Terms of Service, which "no one really reads". She went on to suggest that as a result, most customers will inadvertently agree to the sharing of their genomic data, and end up "paying to help the company make money with [their] data" (3). As 23&Me's database continues to grow, their customers will have to trust them to remain true to their promise to withhold genomic data from third parties in the face of the profitability of this information.

23andMe, a popular service for genetic testing and genealogy.

The scientific community has a unique understanding of the potential applications of the databases that these companies have amassed. The majority of scientists would agree that the generating a robust genomic database containing information from a representative sampling of the population is invaluable for biomedical research efforts. However, the marketing of this technology as a consumer product is a different story, as there are currently no robust regulatory measure to protect consumers from having their information shared with various third parties. As scientists, it is our responsibility to ensure that our friends outside of the lab have an appreciation for the power of the information they are handing over to these corporations. As such, we must help ensure that policy keeps pace with technology, and advocate for legislation protecting consumers from the appropriation of their genomic material and guaranteeing fair compensation should they consent to the use of their personal data for research purposes. These companies could potentially be instrumental to the general progress of biomedical research, but we will have to do our part to advocate for their fair and ethical behavior with the genomic data of their customers.

See full list of references on the online version of this article

WHERE ARE THEY NOW?

Michael Convente

Sam Sander Effron

For Dr. Michael Convente, a 2016 alum from DSRB, a professional move out of academia didn't mean giving up on the scientific enterprise. After finishing his doctoral training in the Shore Lab, Michael began working as a medical writer for Scientific Pathways, a medical communications agency within the Nucleus Global network. His office is based in nearby Hamilton, NJ, so Michael is even able to stay local in Philadelphia, and makes the daily commute by train. Scientific Pathways may be relatively new on the scene, but it already has offices in the U.S. and the U.K., producing deliverable content and consulting on medical communications strategy for both academic and private clientele.

Despite no longer being at the bench, Michael still uses the skills he gained from his Ph.D. training on a daily basis. Scientific Pathways is a small company with fewer than a dozen employees, so his responsibilities change as often as projects are assigned. While larger organizations may have distinct teams for business, design, and writing, Michael's roles expand to fulfill the needs of the client.

Most of his day-to-day duties fall under publication support and medical affairs consulting. His manuscript work is focused on clinical trials, for which he both writes and edits content. On the business side, he develops deliverables for medically trained personnel or administrative staff within a medical organization. For example, a project for healthcare professionals might be a written overview of pharmaceuticals in use for a given disease. Conversely, an assignment for regulatory staff may be a slideshow presentation that briefs a legal team on relevant information for a compound that their company has in development.

As a medical writer, Michael lives in an intellectual space that is similar to that of his academic years. However, his day-to-day work feels quite different. For starters, his assignments are broken up as smaller projects based on clients' specific requests. Because he is based in the commercial world, deadlines come and go much faster than the crawl of research, which methodically chugs along, one hypothesis at a time. That being said, Michael does miss the independence of creatively designing his own experiments. And in turn, the elation of acquiring that new piece of data on the frontier of scientific knowledge is unmatched.

These differences aside, there are many parallels that call back to his role as a graduate student. Writing an F31 grant application is not all that different from crafting a persuasive pitch for a prospective client. Similarly, the time management skills required to run several experiments simultaneously are also applicable to the prioritization needed to juggle multiple projects with staggered deadlines.

Michael Convente, DSRB

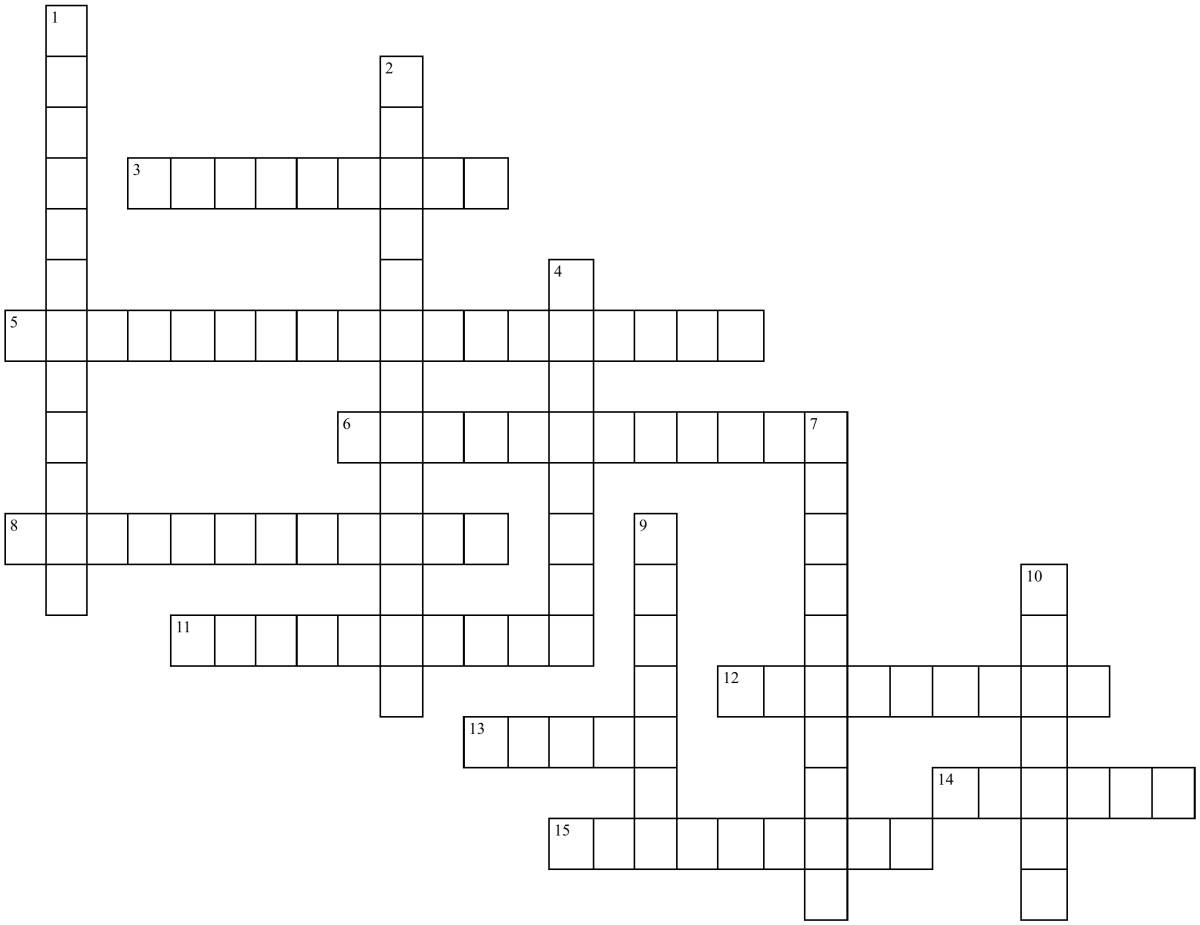
Michael cites his extracurricular involvements at Penn as integral to his successful transition to this new line of work. He spent time with the Penn Biotech Group, a student group out of Wharton that offers life science consulting services to biotech and healthcare companies. Interfacing with external organizations, he gained real-world experience with project management in a professional setting, and practiced new ways to apply his technical training to situations beyond academia.

He advises current students to take advantage of the many non-research opportunities available at Penn, including groups such as the Penn Science Policy and Diplomacy Group, which operates in the fields of science communication, policy, and diplomacy. Many student groups are relatively low-commitment, and provide a welcoming environment to practice useful skill-sets while exploring other modes of science engagement. Participating in these external activities provides an avenue to learn 'soft skills', like interdisciplinary collaboration and written communication, that are applicable to careers that extend far beyond the bench (no offense, Western blotting!).

Just like how labs here at Penn each have vastly different cultures, there is tremendous variety between professional workplace environments. Michael suggests that job-seekers do their homework on careers and companies of interest. Whether researching online or chatting at a networking happy hour, there are many ways to learn about all types of organizations ranging from massive pharmaceutical groups to one-room STEM outreach non-profits. It's also important to do some soul-searching, and consider your own skills, passions, and desires that you want to incorporate into a future career to help guide your decision.

Michael enjoys his work as a medical writer, which allows him to remain well-integrated within the scientific community, albeit from a new angle. While he misses the personal attachment that came with working tirelessly on his own projects, he welcomes the satisfactory feelings of progress that now come much more regularly. Michael looks back fondly on his time at Penn, and is grateful for all the experiences along the way that helped get him to where he is now.

SCIENCE IN POP CULTURE



Across

3. Author of Pale Blue Dot, narrator of Cosmos
5. Vulcan greeting
6. Planet Earth narrator
8. Mary Shelley Novel
11. A botanist in space
12. "Nothing spreads like fear" movie
13. Director of the Hayden Planetarium
14. Elon Musk, CEO
15. Blue pill or red pill?

Down

1. Life uh… finds a way
2. Cooper, Sheldon et al.
4. Flux Capacitor inventor
7. a.k.a Walter White
9. The science guy
10. Primate advocate