In Vitro Cellular & Developmental Biology - Animal (2023) 59 (Suppl 1):S50–S53
https://doi.org/10.1007/s11626-023-00789-9

Plenary Symposia

PS-1

Women in STEM: Advice I Would Give My Younger Self. ALLISON SONGSTAD. NanoString Technologies, Inc., 530 Fairview Ave N., Seattle, WA 98109. Email: asongstad@nanostring.com

Working in STEM comes with its fair share of obstacles for anyone. However, it can be more challenging for minority groups, such as women and people of color. Sharing experiences and learning from peers and mentors about how they faced their challenges can help us understand how to handle difficulties, like imposter syndrome and feelings of inadequacy. During this session, Allison will talk about her experiences and share advice that helped her overcome challenges throughout her education and early career.

PS-2

Empowering Diversity in STEM: Journey to STEM from a Non-traditional Agricultural Background. J. HOEGER. College of Biological Sciences, University of Iowa, Iowa City, IA 52242. Email: Jasmyn-hoeger@uiowa.edu

Cultivation of diversity in STEM brings forth innovation, alternative perspectives, collaboration, and revolutionary findings. Creating a space for diversity within STEM is imperative, however the challenge lies in marginalized individuals rising above, defying norms, and finding their place within STEM. During the Diversity in Science plenary session I will address my struggles, triumphs, and road to science from a non-traditional agricultural background. Growing up in a small town, on a family farm, and in a large family created barriers for me to defy norms and chase my dreams. Doubt was seeded in my mind early on, I will build upon my experience to provide insight on my strenuous journey that has just begun to provides advice to fellow students and highlight the dangers of academic pressures, unconscious biases, and the importance of mental health. Everyone has a different story and journey to STEM, and it is our journey that makes us unique, cultivates innovation, and changes the world one step at a time. Fundamentally, embracing our heritages and diverse background is key to the cultivation of diversity in science.

PS-3

Your Experience, Your Story, So Tell It! AMBER MAYNARD. Corteva Agriscience. Email: amber.maynard@corteva.com

Is it possible to innovate and drive your work forward with colleagues and employers whose ideas, opinions, and values differ from yours? Join us to learn how story creates space for people to drive meaningful change, one conversation at a time. Audience members will learn about how powerful a tool like authentic storytelling can be to drive innovative and creative workspaces, bridging differences through mindful conversations. Specific takeaways for audience: Learn the importance of getting crystal clear about the "one thing" in order to tell an authentic and compelling story. Understand how critical leadership skills like empathy and emotional intelligence help create safe spaces where colleagues feel empowered to show up authentically and contribute to their fullest potential. Develop awareness of the importance of story for organizations to attract and retain the most qualified talent to drive high levels of innovation and creativity and cultivate workplace environments where everyone can bring the best version of themselves to work each day.

PS-4

Commercialization of a Peripheral Nerve-on-a-Chip: "Trials and Triumphs". M. J. MOORE. Dept. of Biomedical Engineering, Tulane University, New Orleans, LA 70118 and AxoSim, Inc., New Orleans, LA 70112. Email: mooremj@tulane.edu

Currently less than 9% of experimental drugs that enter phase I clinical trials will ever be used in patients, and the number is even lower for drugs targeting the nervous system. Microphysiological systems, also called organs-on-chips, are rapidly being adopted as advanced in vitro models for screening drugs in order to improve this poor track record. Modeling the peripheral nervous system using such devices presents unique challenges owing to its complex structure and organization, and its unique physiological role in information transmission and processing. Past designs of neural microphysiological systems based on microfabricated chip-based platforms and 3D neural organoids have limitations in their abilities to recapitulate appropriate cell-cell interactions, morphology, and functionality as seen in vivo. We have developed a 3D peripheral nerve-on-a-chip that enables physiological evaluation in manners analogous to clinical evaluation that is useful for scientific discovery and drug screening. Deploying this model in a commercial setting has required consideration of reproducibility, quality control, and scalability. The realization of in vitro systems as viable commercial products must undoubtedly take these considerations into account, and addressing these challenges early in product development will facilitate adoption.

PS-5

Better Salads for the Produce Aisle: The Story of Conscious™ Greens Development from Proof-of-Concept to the Marketplace. AARON HUMMEL. Pairwise, Durham, NC. Email: ahummel@pairwise.com

As CRISPR therapies advance through clinical trials, genome editing tools are also enabling significant innovation in crop plants important for consumer health. Pairwise is a health-focused food and agriculture company creating a new consumer-centric category of novel, nutritious foods under our Conscious™ Foods brand. In fact, we are poised to launch our first product, a nutrient dense salad, later this year. Conscious Greens were developed with CRISPR by eliminating myrosinase, the enzyme responsible for mustard oil biosynthesis in fresh Brassica juncea leaves. Salads created with these leaves were served to consumers in three key west coast markets and were enthusiastically received, suggesting acceptance of gene edited foods that confer direct consumer benefits. Conscious Greens will be available in food service and retail this year, making them the first branded food developed with CRISPR in the US market.In this talk I will share the story of Conscious Greens development from proof-of-concept to the marketplace.

PS-6

Disrupting Agriculture While Not Disrupting Global Food Chains. RAYMOND D. SHILLITO. Shillito & Associates, Chapel Hill, NC 27517. Email: rayaacci@gmail.com

Biotechnology, enabled by *in-vitro* biology, has been a huge positive disrupter to the agricultural economy over the last nearly 30 years since the first herbicide and insect resistant crops were introduced. The size of the market is now estimated as above 30 Billion USD although estimates vary widely depending on what is included. It is expected to double by 2030. Taking a product to market requires a cascade of actions. The Ag. Biotechnology sector is highly regulated. While the situation for crop plants is complex, engineered animals are even more difficult to commercialize. It is important to maintain stewardship of every stage of the process, especially with plants. Failure to do so can result in unintended negative disruptive impacts, even to the level of affecting global trade flows. Enabling this technology to reach the market begins with an idea, a proof-of-concept stage, field trials performed in multiple locations/environments and development into viable varieties or breeds. To do so requires regulatory filings in the country where the trials will occur, and a process to comply with or exceed requirements for confinement of the material, and then potentially regulatory filings on a global scale. Principles like the Excellence Through Stewardship (ETS) guidelines can help address product stewardship and quality management throughout the life cycle. The advent of genome editing, which is likely to be regulated in an even more fragmented and discordant way than traditional plant and animal biotechnology will pose additional opportunities and challenges.

PS-7

Microbiomes and Antimicrobial Resistance – a "One Health" Perspective. TIM MCALLISTER and Rahat Zaheer. Lethbridge Research and Development Centre, Agriculture and Agri-Food Canada, Lethbridge, AB T1J 4B1, CANADA. Email: tim.mcallister@canada.ca

Antimicrobial resistance (AMR) is one of the grand health challenges to humanity of the 21st century. In fact, some researchers have projected that AMR infectious diseases will cause more deaths than cancer by 2050. Anthropogenic use of antimicrobials is widespread in human - veterinary medicine, animal - plant agriculture and aquaculture. Prolonged use of antimicrobials (AMU) will invariably result in some, but not all members of the microbiome becoming AMR. However, AMU is not the only factor that impacts AMR in microbiomes. Changes in the type and availability of nutrients, biocides, and heavy metals can exert selective pressures that lead to phylogenetic shifts that alter the AMR profile of microbiomes. Consequently, a true understanding of the ecology of AMR will only be garnered if it is investigated from the perspective of microbiomes within a One

Health context. The current presentation will outline such an approach as it relates to the role that microbiomes in beef cattle and their environment play in One Health AMR.

PS-8

The Second Century of Phytobiome Research. C. T. BULL. Department of Plant Pathology and Environmental Microbiology and The Penn State Microbiome Center, The Pennsylvania State University, 111 Buckhout Laboratory, University Park, PA, 16802. Email: CaroleeBull@psu.edu

Researchers in plant sciences, environmental microbiology, and plant pathology have been studying phytobiomes for over a century. Starting with the microscope, then with culture-dependent approaches, and now with culture-independent approaches, researchers continue to characterize the presence and absence, or diversity of microorganisms associated with plants and plant phenotypes at greater levels of refinement. In 1890, Koch outlined methods for demonstrating disease etiology that went beyond association. Falkow, in 1988, furthered Koch's postulates by adapting them to the demonstration of causal relationship between genes and an observed phenotypes. Similar postulates are now needed to demonstrate causal relationships between phenotypes observed in a phytobiome and the roles of the players. The move from culture-dependent to culture-independent characterization of microbiomes associated with plants relies on high-throughput sequencing and analysis using either an amplicon or metagenomic approach. Culture-independent methods (-omics) are moving us beyond cataloging organisms associated with plants, to understanding the functional relationships among members of the phytobiome in situ. Spatial and temporal multi-omic studies are stimulating new questions as we to tease apart complex phytobiome interactions. Innovative approaches to capture, store, and use microbiome assemblages are allowing us to assess the impact of these assemblages and circle back to principles attributed to Koch. These coupled to advances in diagnostic metagenomics when coupled to whole genome sequences type strains and other reference strains are powerful tools for managing plant health.

PS-9

The Host-Microbiome Interactome: Discovery Inferred from Systems Biology. TOR SAVIDGE. Baylor College of Medicine, Baylor Plaza, Houston, TX. Email: tor.savidge@bcm.edu

Targeted metagenomic sequencing is an emerging strategy to survey disease-specific microbiome biomarkers for clinical diagnosis and prognosis. However, this approach often yields inconsistent or conflicting results due to inadequate study power and experimental bias. We introduce Taxa4Meta, a bioinformatics pipeline explicitly designed to compensate for technical and demographic bias. We designed and validated Taxa4Meta for accurate taxonomic profiling of 16S amplicon data acquired from different sequencing strategies. Taxa4Meta offers tremendous potential in identifying clinical dysbiotic features that can reliably predict human gastrointestinal disease, validated comprehensively via re-analysis of individual patient 16S datasets. Specifically, Taxa4Meta facilitates pan-microbiome profiling of 16S data generated microbiome-based classifiers with excellent utility for stratification of diarrheal patients with Clostridioides difficile infection, irritable bowel syndrome and inflammatory bowel diseases who share common symptoms that are difficult to diagnose and manage. Taxa4Meta represents a novel "best practices" approach to individual microbiome surveys to define dysbiosis at a population-scale level. By integrating this new microbiome profiling strategy with functional systems data we investigate the impact of diet and inflammation on the host-microbiome interactome.

PS-10

Single-cell Analysis of Plant Shoot Meristems Opens a 'Goldmine' for Functional Studies. XIAOSA XU[1], Michael Passalacqua[1], Jesse Gillis[1,2], and David Jackson[1]. [1]Cold Spring Harbor Laboratory, Cold Spring Harbor, NY 11724 and [2]University of Toronto, Toronto, Ontario M5S 3E1, CANADA. Email: xxu@cshl.edu

Shoot meristems determine plant architecture and impact crop productivity. An understanding of these structures requires insights into developmental domains, and the gene networks required to specify them. However, these domains are classified mainly by morphology, or insights from classical genetics, but this knowledge is limited by genetic redundancy and pleiotropy. Here, we constructed a single-cell gene expression atlas of maize inflorescence shoot meristems. We revealed novel developmental markers and validated them by bulk RNA-seq and mRNA in situ. Next, we created a gene co-expression network at single-cell resolution to predict genetic redundancy. We also integrated transcription factors ChIP-Seq with single-cell data to build transcriptional regulatory networks. Finally, we combined Genome-Wide Association Studies with single-cell data to identify yield-associated genes. We further successfully captured rare stem cells in *Arabidopsis* and maize shoot meristems that were largely

missed in previous plant shoot single-cell studies. We identified stem cell markers and validated their expression using a high-resolution spatial analysis approach. We conducted a cross-species single-cell analysis to discover conserved stem cell markers and cell types. Plant stem cells are maintained by a conserved CLAVATA-WUSCHEL (CLV-WUS) pathway. We thus also profiled single cells from the inflorescence shoot apices of mutants of crucial regulators in CLV-WUS pathway. We found hundreds of differential expressed genes (DEGs) in the stem cells by comparing these mutants with wild type. We used multiplex CRISPR/Cas9 to knock out selected DEGs in a family of predicted sugar kinases, and found a striking meristem termination phenotype, validating the predictive power of our single-cell atlas. Together, this comprehensive shoot meristem single-cell atlas will open a 'goldmine' for functional studies at a fundamentally new level, and will be a valuable resource for the plant community. *Funding acknowledgment: NSF*

PS-11

Space - the Final Frontier: The Road to Mapping Transcriptomes in 3D. K. COX[1], E. Schaudy[2], K. Duncan[1], M. Giridhar[2], M. Somoza[2], K. Czymmek[1], and B. Meyers[1]. [1]Donald Danforth Plant Science Center, St. Louis, MO 63132 and [2]Technical University of Munich, Freising, GERMANY 85354. Email: KCox@danforthcenter.org

To understand how genes regulate across tissues and organs, transcriptional data needs to be placed within a spatial, cellular, and organismal context. The rapid evolution of methods to visualize the spatial locations of proteins and transcripts has provided an exciting future to study spatial biology in plants and animals. Additionally, obtaining 3D images of tissues and organs at nanoscale resolution will provide better understanding of how cells are spatially organized and help answer some key biological questions. I will focus my talk on some of the technologies I am developing that could potentially be applied to obtain multi-dimensional transcriptional data in organisms. One area of my research aims to develop a platform that allows for spatially resolved, high-throughput transcriptional profiling at a cellular level in plants. In addition, I will discuss 3D imaging data obtained via X-Ray Microscopy and how potentially combining it with spatial transcriptomics could provide a multi-dimensional view of gene expression in organs. Lastly, I will talk about how I am developing a toolkit to perform expansion microscopy in plant systems to observe subcellular components in 3D.