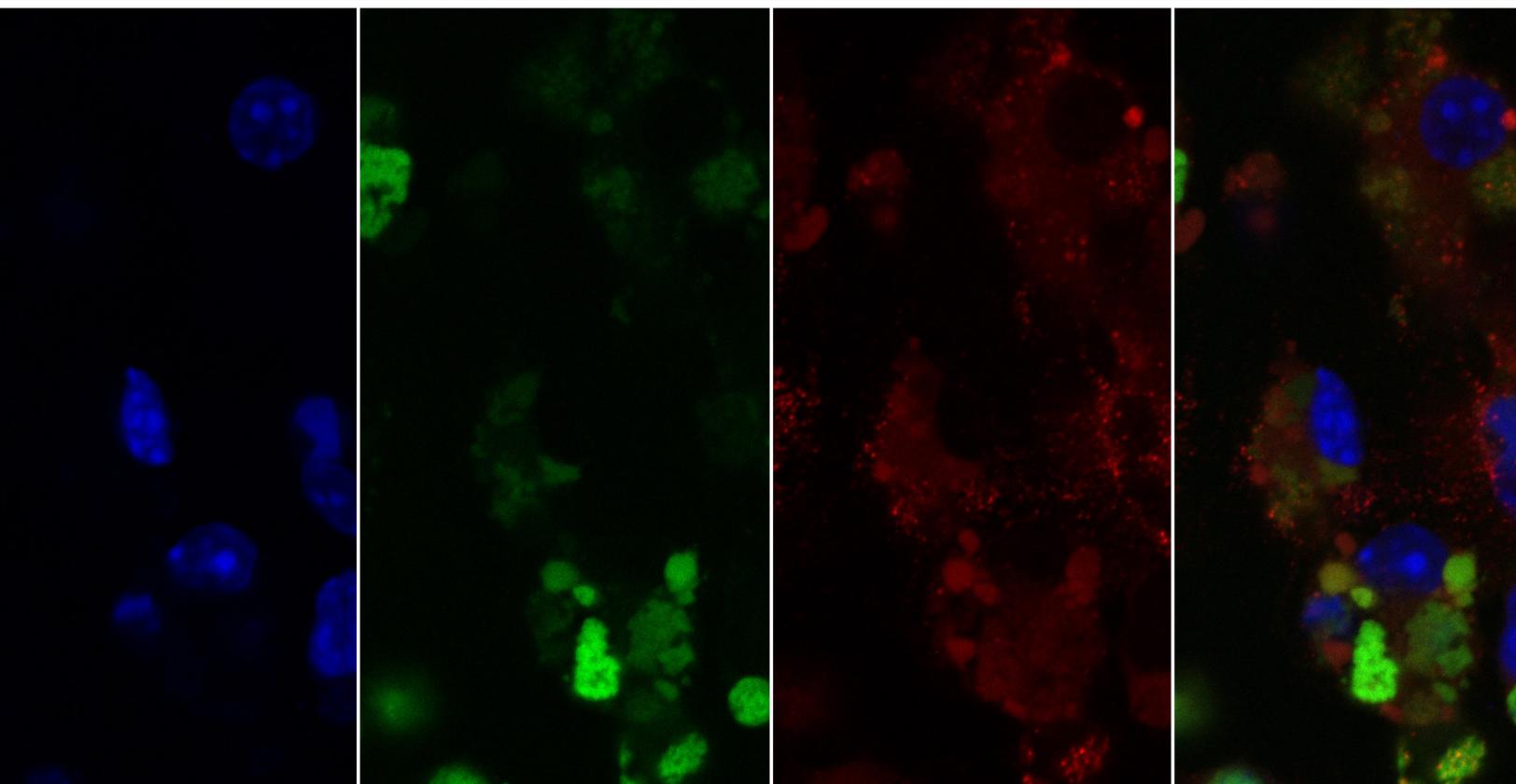


2022

April 20, 2022

Nutrition Symposium

University of Illinois at Urbana-Champaign | Division of Nutritional Sciences



College of Agricultural, Consumer & Environmental Sciences
Nutritional Sciences

NSGSA

Nutritional Sciences
Graduate Student Association



On behalf of the Nutritional Sciences Graduate Student Association (NSGSA), the Division of Nutritional Sciences (DNS), and all participating presenters, we would like to welcome you to the 2022 Nutrition Symposium at the University of Illinois Urbana-Champaign!

The Nutrition Symposium is an important event for sharing ideas across disciplines and with the community. Started in 1994 by NSGSA, the symposium offers graduate students with nutrition-related research on campus an opportunity to present prior to annual national and international scientific meetings and conferences. This symposium offers a first glance at exciting research in areas including metabolic regulation, cancer, gastrointestinal physiology, immunology, physical activity, public health, and bioactive plant compounds. Students will be traveling to present their work at a variety of national and international conferences.

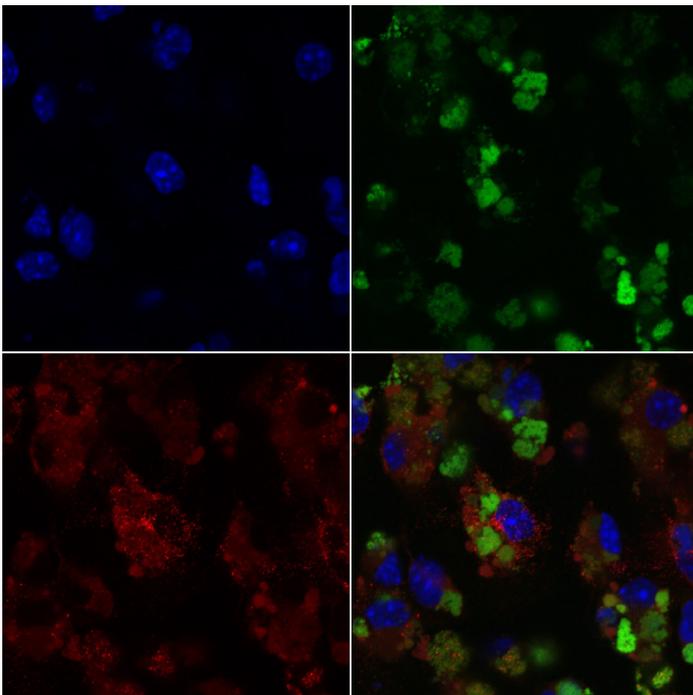
This year, we are honored to have Dr. Elizabeth J. Parks deliver the keynote address, "How Food Becomes You." Additionally, NSGSA is

proud to highlight the work of world-class faculty members through a mini-symposium. This year's presentations highlight the field of metabolic regulation of disease and will feature Drs. Jaume Amengual, Bo Wang, Rex Gaskins, and Kelly Swanson.

We are grateful to the many people involved with this meeting and program. We would first like to thank our keynote speaker, Elizabeth J. Parks. Thank you also to our sponsors – their support is essential to the success and quality of the program. We would also like to recognize the NSGSA Steering Committee and the symposium planning committee, whose members have worked long and hard to organize an excellent program. Most of all, we would like to thank our session chairs, judges, presenters, and attendees for participating in this year's event and making them a success.

The Nutritional Sciences Graduate Student Association Chair and Chair-Elect

nutrsci.illinois.edu



(Cover Image) Composite of single color and merged images of apoptotic cell death bodies (green) engulfed by macrophages (nuclei in blue, F4/80 macrophage marker in red) in a process known as efferocytosis. Macrophages are key regulators of inflammation and promote tissue healing as a result of their capacity to phagocytize apoptotic or dead cells through efferocytosis. For example, this process is essential to promote atherosclerosis resolution and prevent the progression of the disease when lipid-laden macrophages undergo apoptosis and may induce tissue necrosis and more advanced complications. In our study, we explore the effect of retinoic acid, the transcriptionally active form of vitamin A, as a modulator of the macrophage phenotype to promote efferocytosis.

Research image by Ivan Pinos.

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- 8:15 a.m. – 9:00 a.m.**..... **Welcome Breakfast**
Sims Executive Conference Room, ACES Library
Sponsors, presenters, DNS students/faculty/staff invited
- 9:00 a.m. – 9:15 a.m.**..... **Break**
- *9:15 a.m. – 10:15 a.m.** **Oral Session 1: Nutrient Metabolism**
Monsanto Room, ACES Library
9:15 - 9:30: Ziting Chen
9:30 - 9:45: Noah Hutchinson
9:45 - 10:00: Ivan Pinos
10:00 - 10:15: Hania Taha
- 10:15 a.m. – 10:30 a.m.** **Break**
- *10:30 a.m. – 11:30 a.m.** **Oral Session 2: Dietary Patterns & Food Systems**
Monsanto Room, ACES Library
10:30 - 10:45: Elizabeth Gutierrez
10:45 - 11:00: Tori Holthaus
11:00 - 11:15: Ana Mitchell
11:15 - 11:30: Yifan Peng
- 11:30 a.m. – 11:45 a.m.** **Outstanding Faculty Award Presentation**
- 11:45 a.m. – 12:45 p.m.** **Sponsor Network Lunch**
Heritage Room, ACES Library
Sponsors, presenters, DNS students invited; RSVP required
- *12:45 p.m. – 2:45 p.m.** **Faculty Symposium**
Monsanto Room, ACES Library
12:45 - 1:15: Jaume Amengual, PhD
1:15 - 1:45: Bo Wang, PhD
1:45 - 2:15: H. Rex Gaskins, PhD
2:15 - 2:45: Kelly Swanson, PhD
- 2:45 p.m. – 3:00 p.m.** **Break**
- 3:00 p.m. – 3:45 p.m.** **Sponsor Panel**
Bevier Commons, Bevier Hall
Sponsors, presenters, DNS students/faculty/staff invited
- *4:00 p.m. – 5:00 p.m.** **Keynote Address: How Food Becomes You**
Elizabeth J. Parks, PhD, FTOS, University of Missouri
150 Animal Sciences Laboratory
- 5:00 p.m. – 5:15 p.m.** **Break**
- *5:15 p.m. – 6:45 p.m.** **Graduate Student Poster Session**
Heritage Room, ACES Library
Evening Reception; Award Announcements
Sponsors, presenters, DNS Students/faculty/staff invited

* Open to general public

The Nutritional Sciences Graduate Student Association (NSGSA) was founded in the spring of 1973 by students in the program. The mission of the organization is to provide a means of communication among graduate students, faculty, and alumni of the Division of Nutritional Sciences (DNS), which spans multiple colleges and departments.

NSGSA serves as a forum for student opinion and input and provides students the opportunity to expand their experiences as graduate students. Our activities reflect our desire to enrich our experiences as graduate students and promote the importance of the nutritional sciences discipline both within the university and among the surrounding communities of Champaign and Urbana.

NSGSA Board



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Elizabeth Gutierrez	Clara Salame
Megumi Hashida	Leila Shinn

Session Judges

Oral Session 1: Nutrient Metabolism

MC: Steven Kraulis

Judge: Dr. François Reichardt

Judge: Dr. Weinan Zhou

Judge: Dr. Bo Wang

Oral Session 2: Dietary Patterns and Food Systems

MC: Olufemi Fabusoro

Judge: Dr. Hong Chen

Judge: Dr. Elizabeth Parks

Judge: Dr. Brett Loman

Poster Session

Clinical Nutrition and Food Systems 1

Judge: Dr. Miguel Rebollo-Hernanz

Judge: Dr. Marcia Siegel

Clinical Nutrition and Food Systems 2

Judge: Dr. Riley Hughes

Judge: Dr. Elvira de Mejia

Microbiology

Judge: Dr. Brett Loman

Judge: Dr. Corrine Cannavale

Preclinical Metabolism

Judge: Dr. Jacob Allen

Judge: Dr. Diego Hernandez-Saavedra

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Nutritional Sciences

Graduate Student Association

<https://nutrsci.illinois.edu/students/gsa>

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I ILLINOIS

Nutritional Sciences

COLLEGE OF AGRICULTURAL, CONSUMER
& ENVIRONMENTAL SCIENCES

This institution is an equal opportunity provider.

Student Oral Session 1: Nutrient Metabolism

9:15 a.m. - 10:15 a.m.

Monsanto Room, ACES Library

Endometrial glycogen metabolism during early pregnancy in mice

Ziting Chen 17
9:15 a.m. - 9:30 a.m.

Effects of broad-spectrum antibiotic administration and germ-free status on endurance adaptations to regular exercise in mouse muscle tissue

Noah Hutchinson 18
9:30 a.m. - 9:45 a.m.

Retinoic acid exposure promotes phenotypic characteristics of alternative activation in macrophages

Ivan Pinos 19
9:45 a.m. - 10:00 a.m.

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Hania Taha 20
10:00 a.m. - 10:15 a.m.

Student Oral Session 2: Dietary Patterns & Food Systems

10:30 a.m. - 11:30 a.m.

Monsanto Room, ACES Library

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Elizabeth Gutierrez 21
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10:45 a.m. - 11:00 a.m.

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Faculty Mini-Symposium: “Metabolic Regulation of Disease”

12:45 p.m. - 2:45 p.m.

Monsanto Room, ACES Library

Establishment of a mouse model to study HIV-mediated atherogenesis

Dr. Jaume Amengual 13
12:45 p.m. - 1:15 p.m.

Targeting hepatic phospholipid remodeling pathway improves obesity and selective insulin resistance

Dr. Bo Wang 14
1:15 p.m. - 1:45 p.m.

Host-microbiota bile acid co-metabolism in colorectal cancer

Dr. H. Rex Gaskins 15
1:45 p.m. - 2:15 p.m.

Dietary management of companion animal obesity

Dr. Kelly Swanson 16
2:15 p.m. - 2:45 p.m.

Graduate Student Poster Sessions

5:15 p.m. - 6:45 p.m.
Heritage Room, ACES Library

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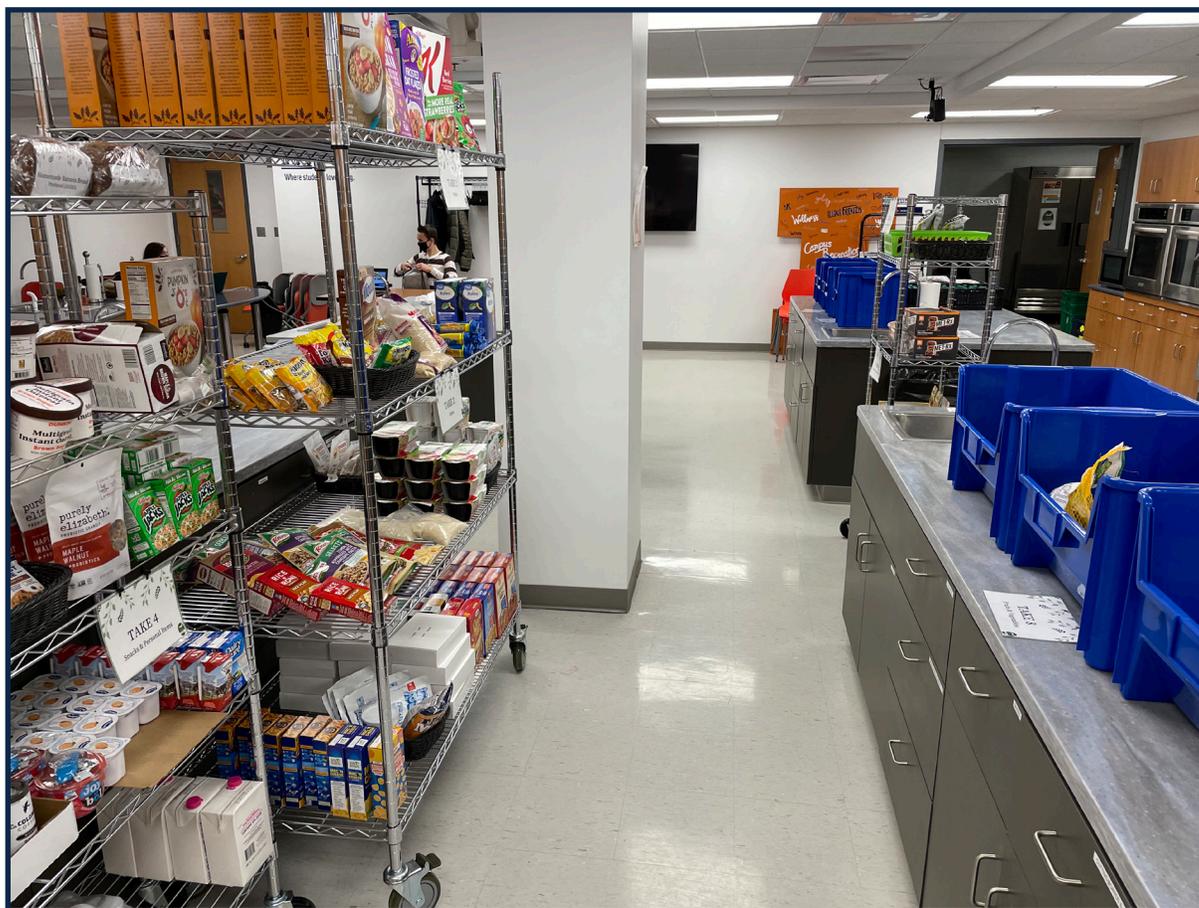
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Ana Mitchell (symposium steering committee member) is conducting an evaluation of the Food Assistance & Well-being Program located at the Activities and Recreation Center to understand more about how the pantry is reaching students, meeting their needs, and delivering key nutrients to students facing food insecurity.

The University of Illinois Division of Nutritional Sciences and the Nutritional Sciences Graduate Student Association would like to acknowledge the generosity of the sponsors and friends of our 2022 Nutrition Symposium.

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 Nlumn

Keynote Speaker

Dr. Elizabeth J. Parks, PhD, FTOS

University of Missouri, *Departments of Nutrition and Exercise Physiology, and Medicine, Division of Gastroenterology and Hepatology*

- *NextGen Institute Investigator*
- *School of Medicine Professor*
- *Clinical Research Center Associate Director*

The Obesity Society President-Elect 2022

“How Food Becomes You”

Dr. Parks received her PhD in Nutritional Biochemistry from the University of California-Davis and then completed a Postdoctoral Fellowship at Berkeley/UCSF in the laboratory of Marc Hellerstein, MD, PhD—an internationally-recognized expert in the study of metabolism. Dr. Parks has held academic positions at the University of Minnesota and the University of Texas Southwestern Medical Center in Dallas, where she collaborated to establish a highly successful research consortium for the study of obesity. In 2013 she moved to the University of Missouri “Mizzou” (MU), where she is a Professor in the Department of Nutrition and Exercise Physiology and in the Department of Medicine’s Division of Gastroenterology and Hepatology. She serves as Associate Director of the Clinical Research Center in the Medical School’s Institute for Clinical and Translational Science, where she conducts studies funded by the federal government and the pharmaceutical industry. She has been recently appointed an Investigator in the new NextGen Institute for Precision Health on the MU campus.

The major research contributions of Dr. Parks’ lab emanate from her development of mass spectrometry techniques to quantitate the delivery and disposal of dietary macronutrients in animal models and in humans. Her seminal studies discovered the contribution of dietary sugars to the development of nonalcoholic fatty liver disease (NAFLD). As a result, two of her highly-cited papers have received placement in the top 1% of all influential publications in the field of Clinical Medicine.



From a national service perspective, Dr. Parks has been a standing member, and chair, of numerous NIH grant review committees. She is a fellow (F) of the American Heart Association (AHA) and The Obesity Society (TOS). Currently, she serves as president-elect of The Obesity Society, the largest professional organization in the U.S. for the study of causes and treatments of obesity. Elizabeth is PI on an NIH project to understand treatments for NAFLD, and also a PI on an NIH-funded grant to direct an annual, isotope short-course at Vanderbilt University. Lastly, her research impact is extended by her serving as a co-investigator and mentor on 9 other NIH grants at Mizzou and around the country.

Dr. Parks’ current research in humans focuses on how both food composition and post-meal metabolism lead to metabolic diseases.

Keynote Address
4:00 p.m. – 5:00 p.m

Faculty Mini-Symposium: Metabolic Regulation of Disease

Abstracts and Biographies

Establishment of a mouse model to study HIV-mediated atherogenesis

Dr. Jaume Amengual

Department of Food Science and Human Nutrition, Division of Nutritional Sciences, University of Illinois Urbana-Champaign, Urbana, IL

ABSTRACT: Even when taking antiretroviral therapies to reduce systemic viral load, patients infected with HIV have more than double the risk of experiencing a cardiovascular event than uninfected subjects. Atherosclerosis is characterized by the accumulation of cholesterol-laden cells in the arterial wall, and it is the leading underlying cause of cardiovascular diseases (CVDs). Observational studies show that HIV patients experience greater degrees of atherosclerosis, presenting inflamed and unstable lesions prone to rupture. The rupture of these lesions culminates in CVD manifestations, such as stroke or myocardial infarction.

We utilized a viral construct specifically designed to study HIV in rodents to evaluate the mechanisms underlying these observations. We infected low density lipoprotein receptor deficient (Ldlr^{-/-}) mice, a standard model for studying atherogenesis. Our data show that Ldlr^{-/-} mice infected with our virus experienced more significant plaque inflammation and necrotic area, a sign of plaque fragility. These changes were accompanied by an increased circulating content of pro-inflammatory monocytes. We failed to observe differences in plasma lipid profile, suggesting that systemic inflammation alone mediates the detrimental effects of HIV on atherosclerosis. Overall, this experimental model will help us establish therapeutic interventions to mitigate CVD incidence in HIV-infected individuals.



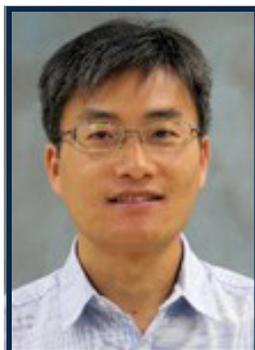
BIOGRAPHY: Dr. Jaume Amengual obtained his PhD in 2009 in Biochemistry at the University of the Balearic Islands. Immediately after, he came to the US to continue his studies on vitamin A at Case Western Reserve University. In 2013, he moved to New York University to train in the Cardiovascular disease department, where he was promoted to research assistant professor after obtaining a career development grant from the AHA. In 2018, he moved to UIUC to continue his research on vitamin A, carotenoids and cardiometabolic diseases.

Targeting hepatic phospholipid remodeling pathway improves obesity and selective insulin resistance

Dr. Bo Wang

Department of Comparative Biosciences, Division of Nutritional Sciences, University of Illinois Urbana-Champaign, Urbana, IL

ABSTRACT: Phospholipids (PLs) are important components of biological membranes and precursors of numerous signaling molecules. PL membranes compartmentalize living cells, form intracellular organelles, and provide platforms for a wide variety of physiological processes. The fatty acyl composition of PLs determines the biophysical characteristics of membranes, including fluidity and the assembly of specific membrane subdomains. Therefore, changes in fatty acyl composition can affect the properties of proteins associated with membranes and influence the biological processes that occur on them. However, due to the difficulty of manipulating fatty acyl composition in animals, the physiological impact of dynamic changes in PL fatty acyl composition has not been explored. We recently have identified a lysophosphatidylcholine acyltransferase 3 (Lpcat3) as a major determinant of PL composition in liver. Previous studies showed that loss of Lpcat3 in the liver results in increased membrane saturation and impaired VLDL secretion and lipogenesis. Our new data demonstrated that hepatic membrane phospholipid composition controlled by Lpcat3 regulates insulin signaling and systemic glucose and lipid metabolism. Hyperinsulinemia induced by high-fat diet (HFD) feeding augments hepatic Lpcat3 expression and membrane unsaturation. Loss of Lpcat3 in the liver improves insulin resistance and blunts lipogenesis in both HFD-fed and genetic ob/ob mouse models. Mechanistically, Lpcat3 deficiency directly facilitates insulin receptor endocytosis and signal transduction, and indirectly enhances Fibroblast growth factor 21 (FGF21) secretion, energy expenditure, and glucose uptake in adipose tissue. These findings provide insights into the pathogenesis of selective insulin resistance that could inform future therapy.



BIOGRAPHY: Dr. Bo Wang received his Ph.D. degree from The Ohio State University and did his postdoctoral training at UCLA. His graduate research focused on elucidating the function of microRNAs in nonalcoholic steatohepatitis (NASH) and liver cancer. His postdoctoral research investigated the function of phospholipid remodeling in lipid metabolism. These studies have been published in journals, such as Cell Metabolism, Cell Stem Cell, JCI, Hepatology, eLife, Oncogene. He has received several honors and awards, such as the NIDDK research scientist career development award, NIH postdoctoral fellowship, American Heart Association postdoctoral fellowship, Distinguished University Fellowship from The Ohio State University.

Host-microbiota bile acid co-metabolism in colorectal cancer

Dr. H. Rex Gaskins

Department of Animal Sciences, Biomedical and Translational Sciences and Pathobiology, Division of Nutritional Sciences, Carl R. Woese Institute for Genomic Biology, Cancer Center at Illinois, Keith W. and Sarah M. Kelley Professor of Immunophysiology, University of Illinois Urbana-Champaign, Urbana, IL

ABSTRACT: Our lab has a decades-long project focused on the roles of genetic background, diet, and bile acid metabolism by the gut microbiota in colorectal cancer (CRC) risk. We have evidence that regardless of disease status, African American subjects, who as a race present with increased risk and mortality from CRC, harbor ten-fold greater abundance of bacteria capable of generating proinflammatory and highly genotoxic hydrogen sulfide than do non-Hispanic white (NHW) subjects. In addition, recent evidence suggests that AA also exhibit a striking depletion of poorly studied isomers of the hydrophobic and toxic secondary bile acids deoxycholic acid (DCA) and lithocholic acid (LCA) that may exert protective effects through the expansion of anti-inflammatory T-regulatory cell (Treg) subset(s) in the gut. Collectively, we hypothesize that CRC health disparities may be ameliorated through the reduction of tumor promoting secondary bile acids DCA and LCA, and enrichment in anti-inflammatory secondary bile acid derivatives.



BIOGRAPHY: Dr. H. Rex Gaskins joined the faculty at the University of Illinois at Urbana-Champaign in 1992 and is the Keith W. and Sarah M. Kelley Professor of Immunophysiology with appointments in the Departments of Animal Sciences, Biomedical and Translational Sciences, and Pathobiology, the Division of Nutritional Sciences, the Carl R. Woese Institute for Genomic Biology and the Cancer Center at Illinois. He obtained the Ph.D. degree in cell biology from The University of Georgia in 1989. From 1989-92, he completed postdoctoral studies in immunology at The Jackson Laboratory in Bar Harbor, Maine. Research in his laboratory focuses on host-intestinal microbe interactions relevant to colorectal cancer with a particular interest in modulation of mucosal inflammation by bile acid and sulfur metabolism. Professor Gaskins has authored 157 peer-reviewed publications and book chapters and has won numerous awards including a Future Leaders Award from the International Life Sciences Institute, a Burroughs Wellcome Fund visiting scientist fellowship (University of Reading), a Sir Frederick McMaster CSIRO Research Fellowship (University of Queensland), being named a University Scholar, the highest honor bestowed by fellow faculty at Illinois, and the Distinguished Scientist Award from the Society for Experimental Biology and Medicine. Professor Gaskins serves as Deputy Director of a NIH-supported Tissue Microenvironment Training Program and Associate Director for Education for the Cancer Center at Illinois.

Dietary management of companion animal obesity

Dr. Kelly Swanson

Department of Animal Sciences, Division of Nutritional Sciences, Kraft-Heinz Endowed Professor in Human Nutrition, University of Illinois Urbana-Champaign, Urbana, IL

ABSTRACT: Obesity is a global epidemic disease not only in humans but also in companion animals, like dogs and cats. Owner misinterpretation of proper body condition, inappropriate feeding, insufficient exercise, and gonadectomy are the primary contributors to pet obesity. Like humans, dogs and cats have many obesity-related comorbidities, including cardiorespiratory disorders, joint diseases, and gastrointestinal disorders that shorten life expectancy, reduce quality of life, and provide an economic burden on pet owners. Given the prevalence and negative impacts of pet obesity, weight management strategies are important for the health and companionship of pets. Various weight management strategies may be used, with increased physical activity and dietary modification serving as the foundation. Increasing meal frequency and limiting treat and snack intake (<10% of daily calories) are usually recommended as well. In regard to nutrient content, most pet diets designed for weight management and/or loss have a low energy density due to substitution of high-calorie ingredients with functional dietary fibers. Such diets also often have increased concentrations of high-quality proteins and micronutrients to preserve lean muscle mass and avoid nutrient deficiency during reduced intake. Increased dietary fiber and protein may also aid in mitigating hunger during weight loss. Several functional ingredients, including L-carnitine, omega-3 fatty acids, and antioxidants are commonly included in such diets. L-carnitine aids in long-chain fatty acid transport and metabolism, especially during weight loss, and has been shown to improve energy expenditure of dogs. Omega-3 fatty acids possess anti-inflammatory and blood lipid-lowering properties. Vitamin C, vitamin E, and other antioxidants are important in limiting oxidative stress. Green tea extract, chromium picolinate, and other functional ingredients may be included to enhance insulin sensitivity. Finally, chondroprotective agents may be used to provide joint support and acid-base balance and dietary mineral concentrations may be manipulated to support urinary health.



BIOGRAPHY: Kelly Swanson is the Kraft Heinz Company Endowed Professor in Human Nutrition and a Professor in the Department of Animal Sciences and Division of Nutritional Sciences at the University of Illinois at Urbana-Champaign. His laboratory studies the effects of nutritional intervention on health outcomes, identifying how diet impacts host physiology and gut microbiota, with primary emphasis on gastrointestinal health and obesity in dogs, cats, humans, and rodent models. To date, Dr. Swanson's laboratory has obtained over \$21 million in research support and published over 215 peer-reviewed journal articles. He has given over 150 invited lectures at scientific conferences around the world and has received 15 research and teaching awards. He has trained approximately 40 graduate students and post-doctoral fellows. Dr. Swanson is also an active instructor, teaching 3-4 nutrition courses to undergraduate and graduate students on campus and online annually. He has been named to the university's 'List of Teachers Ranked as Excellent by Their Students' 27 times. He serves on advisory boards for many companies in the human and pet food industries as well as non-profit organizations, including the Institute for the Advancement of Food and Nutrition Sciences (IAFNS) and International Scientific Association for Probiotics and Prebiotics (ISAPP).

Graduate Student Oral Session Abstracts

Oral Session 1: Nutrient Metabolism

Endometrial glycogen metabolism during early pregnancy in mice

Ziting Chen¹, K. Sandoval¹, and M. Dean¹

¹ Department of Animal Science, University of Illinois at Urbana-Champaign, Urbana, IL.

INTRODUCTION: In humans, at least 30-40% of pregnancies fail, most during the preimplantation period. Prior to implantation too much or too little glucose impairs embryo development. Pregnancy also requires successful decidualization of the stroma, a glucose intensive process. Therefore, optimal levels of endometrial glucose are required to achieve a successful pregnancy. Glycogen, the storage form of glucose, is found in the uterus and may contribute to maintaining glucose concentrations. Our objectives were to 1) determine how glycogen levels change in the luminal epithelium, glandular epithelium, and stroma during early pregnancy; 2) understand the distribution of glycogen metabolizing enzymes (hexokinase I, HK1; glucose-6-phosphatase, G6PC; glycogen synthase, GYS; phospho-glycogen synthase, pGYS; and glycogen phosphorylase, PYG); and 3) determine if the decidua stores glycogen independently of pregnancy.

METHODS: To characterize glycogen during pregnancy, CD-1 mice uteri (n=6) were collected at proestrus, 1.5 days post-coitum (DPC 1.5), DPC 3.5, and DPC 5.5. To artificially induce decidualization, CD-1 mice (n=4) were ovariectomized, primed with steroids, and one uterine horn was stimulated with corn oil. Periodic acid-Schiff staining, with and without diastase pre-treatment, was used to measure glycogen content in endometrial tissues. Immunohistochemistry (IHC) was performed to localize the glycogen metabolizing enzymes.

RESULTS: In the glandular epithelium, glycogen content was highest at proestrus and decreased 71.4% at DPC 1.5 ($P<0.01$) and 62.13% at DPC 3.5 ($P<0.01$). Similarly, in the luminal epithelium glycogen was highest at proestrus, was 46.2% lower at DPC 1.5 ($P=0.061$) and 63.2% lower at DPC 3.5 ($P<0.05$). In contrast, the stroma stored little glycogen during the preimplantation period, and did not change. However, at DPC 5.5, glycogen content increased 7-fold in the decidua surrounding the embryo compared to stroma of proestrus ($P<0.0001$). IHC showed that HK1 was present in the glandular and luminal epithelium. It was undetectable in the stroma. GYS in the luminal and glandular epithelium was highest in the preimplantation period compared to proestrus. It was not detected in stromal cells. At the implantation site, there was a dramatic increase in GYS in the decidua. pGYS was found in glandular and luminal epithelium and was highly expressed in decidua at DPC 5.5. G6PC was highly expressed in the luminal and glandular epithelium from DPC 1.5 to DPC 5.5. PYG increased from proestrus to DPC 3.5 in the glandular and luminal epithelium. The artificially decidualized stroma showed a 5-fold increase in glycogen content ($P<0.05$). The levels of GYS, G6PC, and PYG were increased in the decidualized uterine horn compared to the control.

CONCLUSION: Glycogen in the luminal and glandular epithelium of murine uterus decreased during early pregnancy. This suggests that glycogen in the uterine epithelium supports the preimplantation embryos. In contrast, glycogen in the stroma was constantly low until after implantation. After which, glycogen and GYS expression increased dramatically in the decidua. Thus, decidual glycogen, may be used to support remodeling of the decidua or to provide energy for embryo development.

Effects of broad-spectrum antibiotic administration and germ-free status on endurance adaptations to regular exercise in mouse muscle tissue

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INTRODUCTION: The gut microbiota modulates a variety of physiologic processes. Recent studies from our group have shown that regular endurance exercise induces alterations in the composition of the gut microbiome and metabolome in mice and humans, which possess the capacity to significantly impact host physiology. We hypothesized that the presence of microbes would affect endurance training adaptations in muscular endurance capacity, mitochondrial activity, and gene expression in mice.

METHODS: To discern differences in adaptations from 6 wks. of voluntary wheel running (VWR), we continuously depleted microbes with antibiotics (ABX) and used germ free (GF) mice to compare to control (CON). Male and female C57Bl/6 mice of all groups underwent daily VWR or sedentary (SED) conditions in a 2 x 3 design (VWR/SED, CON/ABX/GF, n=56). After the intervention, treadmill endurance was assessed, and gastrocnemius and soleus tissue were harvested and analyzed for activity of key Krebs cycle enzyme Citrate Synthase (CS) and/or expression of genes indicating metabolic adaptations.

RESULTS: Two-way ANOVA revealed that VWR increased treadmill endurance, ABX had no effect, and that GF status significantly reduced performance. Additionally, VWR increased Citrate Synthase enzyme activity in gastrocnemius muscle tissue in all groups, and ABX and GF status did not reduce VWR's effect, but GF status did reduce it at baseline. VWR also tended to increase expression of genes associated with increased mitochondrial activity (PGC-1 α , CS, Succinate Dehydrogenase) in soleus and gastrocnemius tissue, but ABX had no effect on these observations.

CONCLUSION: From this we conclude that ABX treatment and GF status do not affect VWR induced adaptations in endurance capacity, gastrocnemius mitochondrial activity or expression of metabolic genes in soleus tissue, but that GF status significantly hinders endurance capacity. This indicates that depleting gut microbes does not inhibit muscular endurance training adaptations, but that germ free mice possess hindered endurance exercise capacity.

Erratum for:

Retinoic acid exposure promotes phenotypic characteristics of alternative activation in macrophages

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INTRODUCTION: Macrophages are key players in atherosclerosis, an inflammatory condition that leads to cardiovascular diseases. Classically-activated (M1) macrophages show a pro-inflammatory phenotype, whereas alternatively-activated (M2) are anti-inflammatory. The M1/M2 balance in atherosclerosis determines the fate of the lesions and strategies to increase M2-like macrophages entail promising candidates against atherosclerosis. We have previously shown that vitamin A delays atherosclerosis progression in mice, and others have proposed retinoic acid (RA), the transcriptionally active form of vitamin A, as a modulator of macrophage polarization.

METHODS: Bone marrow-derived macrophages (BMDM) from three wild type C57BL/6J mice were differentiated (M0) and stimulated with LPS/IFN γ (M1) or IL-4 (M2) for 24h, followed by 6h exposure to 1 μ M RA or vehicle (DMSO). RNAseq and gene enrichment analysis was performed on differentially expressed genes to explore regulated pathways by RA. Efferocytosis assays were performed by co-culture of apoptotic-induced Jurkat cells with treated macrophages and subsequent analysis by flow cytometry. Autophagy flux and the lysosomal protein breakdown rate were also analyzed.

RESULTS: RNAseq and differential gene expression analysis showed that RA induced changes in 1,485 genes in M2 macrophages, while 821 and 227 genes were significantly regulated in M0 and M1, respectively (FDR<0.05). Given the lower impact of RA on M1, we focused our subsequent analyses on M0 and M2 macrophages. Arginase 1, a well-known marker of M2 activation, was significantly upregulated by RA in M0 and M2. Western-Blot analyses confirmed the increase of Arg1 protein levels in BMDMs after RA treatment, showing the highest expression in M2 macrophages. Scavenger receptors pathway was significantly regulated in M2 exposed to RA ($p<0.005$), and subsequent efferocytosis assays showed that this group had significantly higher efferocytic capacity compared to M2 with no RA ($p<0.05$) and M0 macrophages ($p<0.005$). Autophagy and Lysosome pathways were also significantly over-represented in M2 macrophages treated with RA, although autophagy flux and protein breakdown rate were not differentially affected by RA in M2 or M0 macrophages

CONCLUSION: Transcriptomic data suggest that M0 and M1 macrophages are less responsive to RA in comparison to M2 macrophages. Our results suggest that RA skews macrophages polarization towards the M2 phenotype when macrophages were already stimulated with IL-4, enhancing alternative activation. Functional assays showed that RA increased efferocytosis in M2 macrophages, a hallmark of alternative activation. Even though RA seems to modulate the transcriptional machinery to induce the expression of autophagy genes, we failed to observe changes in autophagy flux or lysosomal degradation under our experimental conditions. Our data suggest that the responsiveness of M2 macrophages to RA might prompt phenotypic differences that promote an M2-like phenotype.

The feasibility of a carbohydrate-restricted, high-fat diet in head and neck squamous cell carcinoma patients undergoing radiotherapy

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INTRODUCTION: Our previous observational studies indicate that a pretreatment diet high in carbohydrates is associated with increased risk of head and neck squamous cell carcinoma (HNSCC) mortality, while a pretreatment diet high in long-chain fatty acids (FAs), unsaturated FAs, and omega-3 may have beneficial effects on the risk of all-cause mortality, particularly in patients with an advanced stage of HNSCC. The purpose of this study was to conduct a single-blinded randomized controlled trial (RCT) to test the feasibility of a carbohydrate-restricted, high-fat (CRHF) diet in newly diagnosed HNSCC patients who received definitive radiation.

METHODS: 13 Newly diagnosed non-metastatic HNSCC patients were recruited from Augusta Victoria Hospital in East Jerusalem, Palestine and randomized to either Arm A or Arm B. Arm A (N=6) received an intervention of CRHF diet. Arm B (N=7) received a standard diet. Both diets were provided for 2 weeks prior to treatment and during treatment with radiation with enough calories to maintain bodyweight. Feasibility outcomes included recruitment, adherence, attrition, retention, and assessment completion. Safety was assessed by monitoring blood parameters at baseline, start of treatment, and post-treatment.

RESULTS: 53 newly diagnosed HNSCC patients were screened, (N = 20) were eligible. (N = 13) agreed to participate in the study for an enrollment rate of 65%. Reasons for ineligibility included; not meeting eligibility criteria (N = 27); low prognosis (N = 2); and metastatic or recurrent disease (N = 2). Reasons for unenrollment included not being interested to participate (N = 3), could not come for baseline assessment (N= 3), and did not want to be restricted with food (N = 1). Retention rate was 85%, where (N = 2) participants dropped out because they did not like the food provided or not interested to continue. 11 participants completed all study activities. Mean age was 47 years and the average BMI was 26. More than half of the participants were former or current smokers. Most common site was oral cavity and most common stage was stage III. Compliance to meals was 45% (0-72) for Arm A and 67 % (15-98) for Arm B. The mean percentage of weight change for Arm A was lower than Arm B both at the start of treatment and end of treatment. Participants who had higher adherence to the diet had less weight loss during cancer treatment than participants who had lower adherence. Satisfaction rate of the overall quality of the meals was 92% and the overall ranking of the study was 82%. There were no adverse effects reported as a result of the diet.

CONCLUSIONS: The CRHF diet is feasible, acceptable, and safe to implement in HNSCC patients undergoing radiotherapy. This proposed pilot/feasibility RCT is the first step in determining if a CRHF diet is an effective treatment modality in HNSCC patients. A larger sample size is needed to test the efficacy of this diet on HNSCC outcomes.

Oral Session 2: Dietary Patterns & Food Systems

Readily available water is selected most often and suggests potential to reduce child beverage waste during lunch

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INTRODUCTION: The Healthy, Hunger-Free Kids Act requires free, potable water to be available during school meals. Yet, little is known about the impact of beverage selection on waste. Food waste is one aspect that impacts climate change so finding ways to mitigate excess waste is of extreme importance. Overall, food waste results in 1,250 kcals per person per day, 2.6% all U.S. greenhouse gas emissions, 21% of us landfill content, and 21% of us agriculture water usage. The objective of our study was to determine the relationship between beverage selection and the percentage of beverage wasted. Beverage selection choices included water, chocolate milk, white milk, or a combination of a milk choice and water.

METHODS: This is a secondary data analysis of the Time for Lunch study data. A total of 234 observations were collected from school-aged children (n=38) who participated in a 4-week summer camp. Lunches were based on the National School Lunch Program (NSLP) nutrition standards and reimbursement requirements. Mixed effects linear regression was performed using RStudio version 1.3.1093. with random effects, controlling for sex, ethnicity, age, free/reduced-price lunch participation, seated lunchtime, and food consumption.

RESULTS: Participants were majority female (59%), white race (60%), and of non-Hispanic ethnicity (76%). Beverage selection choices included water only (47%), chocolate milk only (17%), white milk only (27%), chocolate milk and water (4.3%), white milk and water (3.0%), or no beverage (1.3%). Children who selected water wasted significantly less of their beverage than children who selected white milk and water ($\beta=39.27$, $p<0.001$) or white milk only ($\beta=31.48$, $p<0.001$). No significant differences were found between those who selected only chocolate milk ($\beta=12.50$, $p=0.08$) or those who selected chocolate milk and water ($\beta=3.10$, $p=0.80$) compared to those who selected water only.

CONCLUSIONS: Water was tolerated at a similar rate as chocolate milk in this study. Research to determine strategies to make water more accessible may help lessen the environmental burden of wasted beverages in the National School Lunch Program and can evaluate how beverage consumption may influence total nutrient intake. Efforts should be made to perform future studies within a diverse population. If water consumption begins to outpace milk in beverage selection, opportunities to further enrich school meals with calcium and vitamin D should be explored.

The MIND dietary pattern is selectively related to cognitive processing speed

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INTRODUCTION: Given that previous research has disproportionately focused on individual nutrients, knowledge of diet patterns and cognitive health is limited. Thus, we investigated the relationship between different diet indices (Mediterranean, Dietary Approaches to Stop Hypertension [DASH], Healthy-Eating-Index-2015 [HEI-2015], and Mediterranean-DASH Intervention for Neurodegenerative Delay [MIND]) and attentional inhibition and neuroelectric function.

METHODS: Adults aged 34.1 ± 6.0 years old ($N = 207$) completed the Dietary History Questionnaire II (DHQII, Past Year and Past Month with Portion Size) to assess adherence to different diet indices. Attentional inhibition was assessed using a modified Eriksen Flanker task while event-related potentials (ERPs) were recorded. The amplitude and latency of the P3 ERP component were used to index attentional resource allocation and information processing speed, respectively. Stepwise linear regression modeling was used to assess the influence of each dietary index on behavioral performance and neuroelectric function following adjustment for significant covariates (e.g., age, sex, intelligence quotient, and body mass index).

RESULTS: Greater adherence to the MIND diet was associated with an earlier incongruent P3 peak latency ($\Delta R^2 = 0.02$, $\beta = -0.14$, $p = 0.04$) but not congruent peak latency ($\Delta R^2 = 0.02$, $\beta = -0.12$, $p = 0.07$). Adherence to the Mediterranean, DASH, and HEI-2015 patterns was not correlated with P3 latency ($p > 0.05$). No associations were observed between the diet indices and attentional inhibition at the behavioral level (i.e., accuracy or reaction time) or P3 amplitude (all p 's > 0.05).

CONCLUSION: Greater adherence to the MIND diet was selectively related to faster information processing speed. This relationship was only evident during incongruent trials, suggesting that the influence of greater MIND diet adherence is particularly beneficial when upregulation of cognitive control is required. Future MIND diet intervention trials are warranted to help inform dietary recommendations for healthy cognitive aging.

Campus food pantry implementation: does nutrient dose promote nutrition security?

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INTRODUCTION: Efforts to alleviate food insecurity have traditionally focused on increasing access to quantity rather than quality of food. Given burgeoning health disparities and lower dietary quality among food insecure individuals, there has been a call to focus on nutrition security—providing consistent access to foods and beverages that promote well-being. The aim of this study was to determine the dose of key nutrients received by students using an on-campus food pantry to determine whether pantry implementation is supporting nutrition security.

METHODS: In August 2020, an on-campus, client-choice, food pantry opened at a large Midwest university; distribution guidelines were based on MyPlate and the pantry was open twice a week, three days apart. Pantry staff tracked student usage, item inventory, and food discarded during the first academic year of implementation. Inventory and waste logs were used to determine items distributed per pantry opening. Items were analyzed for specific nutrients using the Nutrition Data System for Research. Nutrient and distribution data were merged and adjusted for item size and quantity. The mean nutrients distributed was measured and dose-received was calculated as the average nutrients received per person for specific macro- and micronutrients of concern. Values were compared to national recommendations to determine days of adequate intake.

RESULTS: On average, 14 items were selected per student. Nutrients exceeding Dietary Reference Intakes (DRIs) for three days included vitamins A, C, most B vitamins, carbohydrates, and protein, and for men, iron, and women, zinc and magnesium. Nutrients that did not meet the DRIs for three days included vitamin D, and for males, potassium, energy, fiber, and linoleic and alpha linolenic acid. Added sugar made up less than 10% of total calories distributed (9.1%) and Acceptable Macronutrient Distribution Ranges were met for carbohydrates (50.8%) and protein (16.9%) but were exceeded for fat (35.6%).

CONCLUSION: Results suggest distribution standards support students receiving key nutrients, however, more foods fortified with Vitamin D are needed and male students may need access to more food to meet sex specific DRIs. More research on pantry implementation is needed to understand how pantry implementation can best support nutrition security.

The impact of regional produce cooperatives on the distribution of fresh fruits and vegetables in the United States

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INTRODUCTION: The central goal of food banks in the United States is to alleviate food insecurity as federal food assistance programs alone are often insufficient. Increasingly, food banks also work to ensure that food-insecure people have access to sufficient healthy food. Nevertheless, a perceived long-term hurdle to having enough fresh fruits and vegetables (FFV) for food-insecure people is that food banks may not have consistent access to FFV from traditional pathways. Some barriers to the supply of FFV to food banks under the traditional model of procuring food have involved the difficulty of sourcing, perishability, and indivisibility. Due to indivisibility, the traditional model restricted the varieties of FFV that food banks could have, particularly when they had to order each product by truckload size for transportation. Therefore, a new model of FFV distribution to food banks is needed.

One way to achieve this goal is by sourcing through Feeding America's seven regional produce cooperatives (RPCs), launched to serve as regional mixing centers available for food banks in the Feeding America network. The new model using RPCs enables food banks to acquire an increased quantity of FFV at a lower cost than in the past and to form a coordinated model. However, the new distribution model with RPCs has not been rigorously evaluated yet, and therefore, filling the gap on this topic is urgent.

We are interested in whether the overall FFV that a food bank receives can be increased by introducing RPCs. Measuring the overall effect is crucial since it is relevant to how much the FFV food banks can distribute to the local food pantries and agencies and thus to the food bank clients. By answering two questions, we evaluated the impact of RPCs on the overall FFV that food banks receive. First, do RPCs increase the actual amount of FFV at food banks? And second, does the proportion of FFV with respect to the overall food at food banks increase by joining RPCs?

METHODS: To answer the two questions in this study, we used a wide variety of econometric approaches in the difference-in-differences specifications, including the conventional two-way fixed effects (TWFE) and the alternative estimators proposed by Callaway and Sant'Anna under various combinations of weighting strategies with control group settings, which can resolve the potential drawbacks of the TWFE. We applied both quarterly and annual data to both TWFE and the alternative estimators to mutually verify the robustness of each estimator given their pros and cons. We use quarterly, proprietary administrative data for all the 200 food banks in the Feeding America network, which spans from 2012 to 2019, including the pounds of various foods that each food bank received. We linked these data to the timestamp of the food banks' first-time participation in RPCs to determine the treatment timing and, hence, the length of the impact of RPCs on the amount FFV that food banks receive.

RESULTS: The key findings are twofold. First, joining RPCs has a positive impact on the amount of FFV received by food banks. Across models, the average increase was as high as 1.1 million pounds. Second, while the total amount of FFV was increased by RPCs, the proportion of FFV received by food banks remained unchanged. Like with the total amount of FFV, our findings are robust across different specifications.

CONCLUSION: This study identified the effectiveness of running RPCs for the food bank system in terms of supplying more healthy foods in the United States, which can contribute to the design and improvement of policies and programs to increase food banks' access to FFV or other types of healthy and nutritious food. Second, this study is also good practice for the estimators for quasi-experimental design proposed by Callaway and Sant'Anna and shows the various results under different weighting strategies with control group settings.

Graduate Student Poster Session Abstracts

Study Section: Clinical Nutrition & Food Systems 1

An interdisciplinary online weight loss program delivered clinically meaningful weight loss and high participation

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INTRODUCTION: Obesity prevalence has increased in the US, and obesity is associated with comorbidities such as diabetes. Currently no dietary weight management program that can treat obesity reliably and sustainably is available. Individualized Diet Improvement Program (iDip) was developed and tested to achieve sustainable dietary weight loss through informed decision making. Wide variations among participants were observed in weight loss outcome and class attendance, leading to enhancements. We hypothesized that 1) adapting the in-person program to online would increase participation and reduce attrition, and 2) collaborating with behavioral and medical professionals would improve weight loss efficacy by addressing non-dietary barriers to weight loss.

METHODS: Thirty-one patients with BMI ≥ 25 kg/m² and one or more comorbidities were recruited from Carle Clinics and enrolled in a two- year ongoing weight loss study, EMPOWER. EMPOWER is comprised of 19 online sessions (eText) and individual or group coaching sessions (Zoom) by nutrition, lifestyle, or medical professionals. Participants weighed daily using a Wi-Fi -enabled-scale. Body composition and waist circumference were collected at baseline and at six months. A Food frequency questionnaire (FFQ) was obtained at baseline to assess participants' dietary habits. Monthly 24-hr diet records were collected to evaluate diet changes. The focus of dietary changes in EMPOWER was increasing protein and fiber intake while reducing energy intake.

RESULTS: At six-months from baseline, 30 participants remained (96%) and 81% completed at least 15 sessions. The mean weight loss was $-6.6 \pm 5.8\%$ (mean \pm SD) ($p < .001$), whereas mean weight loss in the previous iDip study ($n=22$) was $-5.8 \pm 6.3\%$ ($p < .001$). In EMPOWER, 16 out of 30 participants (53.3%) achieved clinically meaningful weight loss $>5\%$ of initial body weight, whereas in iDip, eight out of 22 participants (36%) achieved $>5\%$ weight loss. No statistical differences were observed in the magnitude of weight loss between the two studies ($p = 0.3$). Mean weight loss of the top, middle, and bottom tertiles ($n=10$ each) of EMPOWER was $-13 \pm 2.9\%$, $-5.8 \pm 1.9\%$, and $-0.6 \pm 2.4\%$, respectively. Protein/kcal and fiber/kcal assessed by FFQ at baseline did not differ among tertiles. Monthly 24-hr records found a significantly higher protein/kcal intake in the top and middle tertiles than the bottom tertile (6.6 vs. 5.3 g/100 kcal, $p < .05$). Although higher intake in fiber/kcal was observed in top and middle tertiles than the bottom tertile, it did not reach statistical significance (1.71 vs. 1.58 g/100 kcal, $p = 0.5$).

CONCLUSIONS: The EMPOWER program resulted in weight loss as effective as the previous study. EMPOWER also achieved low attrition and high participation. The success of weight loss was accompanied by a successful increase in protein/kcal in diets.

Effectiveness of short-term, home-delivered, low sodium meals to sustain changes in dietary behavior in hemodialysis patients in the long term

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INTRODUCTION: Reducing the dietary sodium intake of chronic kidney disease patients receiving hemodialysis (HD) therapy can reduce their volume overload and risk of cardiovascular complications. Previous studies found that dietary education alone is ineffective in reducing the sodium intake of HD patients. A 4-week-study found that short-term low-sodium home-delivered meal provisions could reduce dietary sodium intake, interdialytic weight gain, and blood pressure of HD patients. However, whether changes resulted from short-term feeding of low-sodium meals can be sustained long-term remains unknown. Therefore, the purpose of this study is to determine if short-term feeding of low-sodium meals can “prime” changes in long-term dietary behavior.

METHODS: Eleven HD patients (Age > 18 years old, M = 8, F = 3) who met the inclusion and exclusion criteria have been recruited from a HD clinic in Urbana, IL. Subjects were randomized into two study groups: the control group and the intervention group. Subjects in the control group received standard care for the first 5 months of the study, followed by a 2-month period where they received low-sodium home-delivered meals (2 meals/day at Month 6 and 1 meal/day at Month 7) and dietary education. Subjects in the intervention group received low-sodium home-delivered meals (2 meals/day for Month 1 and 1 meal/day for Month 2) and dietary education for the first 2 months of the study, followed by 3-month continued dietary education. Subjects’ average monthly interdialytic weight gain (IDWG), standardized blood pressure, and sodium intake were measured.

RESULTS: Dietary sodium intake of the intervention group significantly reduced during the first month of home-meal delivery (M0 2309±1356 vs M1 1662±644 p=0.044) and started increasing from Month 2 (M2 1898±756 vs M5 2075±862). The intervention group had a non-significant reduction in IDWG during the first month of home-meal delivery (M0 2.49±1.22 vs M1 2.28±1.11 p=0.134) and increased from Month 2 (M2 2.53±1.41 vs M5 2.59±1.30), while the IDWG of the control group gradually increased (M0 2.59±1.41 vs M1 2.72±1.31 vs M2 2.99±1.27 vs M5 3.29±1.41). No overall interaction effect was found between the IDWG and the sodium intake (p=0.128). The intervention group had a significant reduction in systolic blood pressure (SBP) (M0 154±18 vs M1 150±24 vs M2 145±24 vs M5 143±24 p=0.006), with no change in diastolic blood pressure (DBP) (M0 81±16 vs M1 78±17 vs M2 78±22 vs M5 78±16 p=0.49). An overall interaction effect was found between the SBP and the sodium intake (p=0.025) while no overall interaction effect was found between the DBP and the sodium intake (p=0.143).

CONCLUSIONS: Our study suggests that short-term low-sodium home-meal delivery could reduce dietary sodium intake, IDWG, and blood pressure in HD patients. However, during our study, short-term feeding of low-sodium meals could not “prime” changes in long-term dietary behavior. More studies are needed to investigate the effectiveness of different dietary interventions to sustain changes in the dietary behavior of HD patients in the long term.

Dietary patterns, fiber intake and microbial-derived isobutyrate are associated with executive function in toddlerhood

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INTRODUCTION: Early childhood is characterized by stabilization of the gut microbiome and rapid maturation of executive functions (EF; cognitive flexibility [F]; inhibitory self-control [ISC]; emergent metacognition [EM]), which are vital to the regulation of goal-directed behaviors, academic and social success. While diet in school-aged children and gut microbial-derived volatile fatty acids (VFA) in rodent models are linked to EFs, few studies have explored these in early life among humans. The present study investigated the extent to which dietary patterns, fiber intake and fecal VFA predict EF at 24 mo.

METHODS: Parents and 24-mo-old children (N=291) were recruited from the STRONG Kids 2 cohort study. Parent reported surveys were used to assess EF (Behavioral Rating Inventory of Executive Function for Preschoolers) and diet (Block Food Frequency Questionnaires). To derive dietary patterns, raw frequency responses for diet were used to create 23 food groups, which were imputed into principle component analysis. Analyses were independent of sex, socioeconomic status and energy intake.

RESULTS: Two distinct dietary patterns explaining 30% of the overall variance in diet were evident: higher consumption of fried and sweet foods (DP1) and of vegetables and fruits (DP2). On average, toddlers consumed below the recommendation for fiber intake, 9.10 g/1000 kcals per day. Higher DP1 scores were associated with poorer overall and construct-specific EF (EF: $b=6.146$, $p=0.005$; ISC: $b=4.458$, $p=0.032$; F: $b=4.522$, $p=0.019$; EM: $b=7.322$, $p=0.002$). DP2 was not associated with EF. Toddlers with lower energy-adjusted fiber intake had significantly poorer EF (EF: $b=-1.696$, $p=0.009$; ISC: $b=-1.729$, $p=0.004$; F: $b=-1.149$, $p=0.043$; EM: $b=-1.659$, $p=0.018$). Before adjustment for fiber intake, higher isobutyrate was predictive of poorer EF (EF: $b=0.017$, $p=0.015$; ISC: $b=0.014$, $p=0.036$; EM: $b=0.018$, $p=0.018$). Adjustment for fiber intake nullified the association with ISC and attenuated associations with EF and EM.

CONCLUSIONS: A western-style diet pattern and microbial-derived isobutyrate may have negative implications for EF in toddlers. Additionally, dietary fiber has the potential to influence these relationships.

Production diversity and consumption diversity: panel data evidence from rural India

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INTRODUCTION: The study tries to evaluate the relationship between household agricultural production diversity and consumption diversity since a priori the direction of relationship is ambiguous. If household's production diversity is already high, further increases are unlikely to contribute to increasing dietary diversity: maintaining or increasing production diversity may cause the household to forego benefits from production specialization. In such cases it may be more effective to improve market access for smallholder farmers. A farm household can choose instead to focus on cash crops and use the income towards purchasing a diversified consumption basket.

METHODS: Following Huang et al. (2021) we construct a synthetic control using matching on baseline characteristics and then use the difference-in-difference methodology to calculate causal impacts. We compare households that have significant change in production diversity over time to households with no significant change in production diversity to see if it translated into diversity in diet. For robustness, following Hirvonen & Hoddinott (2017) and Tesfaye & Tirivayi (2019) we use Fixed Effect Instrumental Variable approach to estimate causal impacts. Using fixed effects allows to control unobserved heterogeneity at household level that could affect production diversity & dietary diversity.

RESULTS: Most studies use cross-sectional data, making causal inference difficult due to endogeneity. This study is one of the few studies using longitudinal data based in quasi-experimental setting to obtain causal estimates. Further, we also disaggregate results by different income quantiles where our main variable of interest is women's dietary diversity as measured by total number of food groups woman consumed past 24 hours, including staples, pulses, nuts/seeds, dairy products, meat or fish, eggs, green leafy vegetables, vitamin-A rich fruits, vegetables, and other fruits.

CONCLUSIONS: The study sheds light on whether promoting production diversity is an impactful policy for addressing dietary diversity and consequently tackling undernourishment. This contributes to the need for evidence that tests this association in various contexts and extends beyond Africa (the region accounting for approx. 75% of similar studies). This paper adds to this growing literature by providing empirical evidence in a developing country context using panel data from five states of rural India. Using data on household market participation, we identify how the relationship between household production diversity and dietary diversity differs based on different levels of market access and participation.

Study Section: Clinical Nutrition & Food Systems 2

Strategies and unmet needs to reduce household food waste reported by self-identified food conservers

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INTRODUCTION: In the United States, about 30% of edible food produced is wasted, and 20% is wasted at the consumer level. Yet, an estimated 35% of Americans “put a lot of effort” into wasted food reduction, suggesting an opportunity to learn from these food conservers through positive deviance inquiry. The purpose of the study was to identify food conservation practices, and psychosocial drivers, and unmet waste mitigation needs of self-identified food conservers.

METHODS: Adult, self-identified food conservers were recruited online to complete a one 90-minute virtual focus group and survey consisting of previously validated questions assessing household food waste amounts, behaviors and attitudes. Verbatim focus group transcripts were dual coded and thematically analyzed using a hybrid inductive-deductive approach.

RESULTS: A total of n=27 participants completed the questionnaire and focus groups. The majority of the participants were White (n =13) or Asian (n =11), female (n=18), had a college degree (n=20), had on average 2.6 members in their residence, and made above the U.S. median household income (n=15). Reported strategies to reduce food waste included meal planning, creating and adhering to shopping lists, food inventory management, anti-depth organization of food storage spaces to promote maximum visibility, meal prepping, cooking meals in the home and repurposing leftovers. Many participants were intrinsically motivated to conserve food. Participants reported needing assistance determining optimal produce storage methods and desired opportunities to learn from other food conservers.

CONCLUSIONS: Self-identified food conservers reported a variety of household food waste mitigation strategies, which could be leveraged to positively influence other households. While these findings suggest relatively high food literacy among self-identified food conservers, addressing their unmet needs may improve waste mitigation.

Transitioning a sustainable weight management program to a virtual platform for greater accessibility

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INTRODUCTION: Obesity is a leading public health issue in the United States with 70.2% of adults having either an obese or overweight body mass index (BMI>25). We developed a novel weight management program, the Individualized Diet Improvement Program (iDip) focused on achieving sustainable diet changes. iDip was offered as in-person sessions at the University of Illinois (UIUC) campus. This limitation in time and location was a barrier for workers and residents in distant locations. The objective of the project was to improve accessibility by 1) developing online version of iDip and 2) testing its efficacy.

METHODS: The revised program, EMPOWER, has two components: online-session materials and a mobile app. The eText platform developed by CITL, UIUC was used for session materials. Zoom meeting was used for interactions with clients after a session. A mobile app, MealPlot, was developed using a multi-OS platform in collaboration with Applied Research Institute, College of Engineering, UIUC. 40 participants were recruited from Carle clinics and screened; 32 were eligible and enrolled, and 30 have completed 6-months of the 24-month intervention to date. Primary outcome measures include weight and anthropometrics taken at baseline, 6, 12, and 24 months.

RESULTS: In-class sessions were converted to online. Nineteen lecture sessions were produced with video and text and activities were inserted between modules. All session materials were revised to 6th grade level and non-dietary obstacles were addressed in session material. Activities and an interactive 'make-a-change' assignment apply and test understanding. The answers to activities allow nutrition coaches to customize coaching provided via Zoom call. A Protein Fiber (PF) plot and weight tracking tool was incorporated into MealPlot. MealPlot users can check protein and fiber density of foods, complete 24-hour diet records, and view their weight progress. The plotting of foods helps users with food selection by utilizing a spatial presentation of quantitative values of protein and fiber per calorie for easy comparison of nutrient content. Protein protects muscle mass, and both protein and fiber reduce overeating. The weight chart displays weekly progress charts to monitor energy balance. The EMPOWER trial began in June of 2021 and at the 6-month mark has shown at least as effective as the previous in-person cohort. Mean weight changes of participants at 6 months are -6.7 ± 5.9 kg ($-6.6 \pm 5.8\%$ of initial body weight) in EMPOWER (n = 30) and -5.9 ± 6.3 kg ($-5.7 \pm 6.3\%$ of initial body weight) in iDip (n=22).

CONCLUSIONS: We achieved a successful transition of an in-person weight loss program to the online platform. The EMPOWER program was as effective as iDip in weight loss at 6 months. In the future, the applicability of EMPOWER to underserved populations will be assessed by collaborating with community organizers.

Visceral adiposity, diet quality, and cognitive function in preschool-aged children

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INTRODUCTION: Central adiposity is negatively associated with cognitive function in children and adults. However, the relationship between visceral adipose tissue (VAT) and cognition is understudied in preschool-aged children. In addition, it is not clear whether the influence of VAT is independent of diet quality. The objective of this study was to investigate the relationship between VAT, diet quality, academic skills, and cognitive abilities among preschool-aged children.

METHODS: Children between 4 and 5 years (N=46, 23 females) were recruited from the STRONG Kids 2 cohort study. Dual-energy X-ray absorptiometry (DXA) was used to assess VAT. Woodcock-Johnson Early Cognitive and Academic Development Test was utilized to assess early academic skills (EAS), general intellectual ability (GIA), and expressive language (EL). Covariates accounted for included sex and diet quality. Diet quality was measured using Healthy Eating Index-2015 (HEI) based on 7-day food records.

RESULTS: Bivariate Pearson's correlations were conducted using a one-tailed approach. HEI was positively correlated with both GIA ($r=0.290$, $p=0.009$) and EL ($r=-0.238$, $p=0.025$). VAT was inversely correlated with both GIA ($r=-0.319$, $p=0.009$) and EL ($r=-0.268$, $p=0.022$). However, after the adjustment for covariates, the relationships between VAT with GIA ($r=-0.019$, $p=0.451$) and EL ($r=-0.067$, $p=0.326$) were no longer statistically significant.

CONCLUSIONS: VAT is related to intellectual abilities and expressive language skills in preschool-aged children however following adjustment for diet quality, the relationship was not sustained. These findings and previous literature indicate that influence of dietary factors may potentially mediate the effects of VAT on cognitive abilities in early childhood.

Adherence to eating pattern recommendations and relationship to weight loss at 6 months: findings from the EMPOWER weight management study

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INTRODUCTION: Disrupted eating and sleeping patterns are often present in people with obesity. A consistent eat-sleep cycle may promote health and improve weight loss progress by limiting calorie intake and enhancing weight loss behavior adoption. Recent research has shown the effectiveness of time-restricted eating on weight loss. However, there's controversial evidence on the association between meal frequency and weight loss. In this study, we hypothesized that well-organized teaching materials and coaching could optimize participants' eat-sleep cycle and further increase weight loss.

METHODS: EMPOWER is an ongoing two-year weight management program consisting of 19 online sessions (eText) and a minimum of three online individual coaching sessions. In EMPOWER, the researchers help participants increase control over their health by discovering a sustainable healthy diet and lifestyle through evidence-based education and coaching. The EMPOWER program emphasized the importance of a robust eating-sleeping pattern, including 1) restricting to 10-12 hours eating window, 2) eating balanced breakfast, lunch, and dinner at about the same time every day, 3) maintaining 7-9 hours of sleep, 4) no eating at least 2 hours before going to bed. Online teaching materials (eText), including readings, videos, self-monitoring sheets, and animations, were used to deliver these key points. Individualized coaching was conducted to facilitate the learning process. Eating-sleeping window and meal frequency data were collected with surveys for baseline and 6-month.

RESULTS: At six months from baseline, 30 participants remained in the study with a mean weight loss of -6.7 ± 6.0 kg or $-6.6 \pm 5.8\%$ of initial body weight. Among participants who completed the eating window survey (n=23), compared with baseline, the eating window changed from 12.2 ± 2.0 h to 11.4 ± 1.4 h ($P < 0.05$) on weekdays and from 12.0 ± 2.2 h to 11.0 ± 1.6 h on weekends ($P < 0.05$). 19 out of 23 participants (83%) reported a 12-hour or less eating window at 6-month. Among participants who completed the meal frequency survey (n=25), breakfast frequency was significantly increased on both weekdays ($P < 0.01$) and weekends ($P < 0.01$), while evening snacking frequency was significantly decreased on both weekdays ($P < 0.001$) and weekends ($P < 0.01$). Participants who eliminated their evening snacks (n=9) lost significantly more weight than other participants (n=16) (-11.0 ± 7.8 kg vs -3.7 ± 4.3 kg, $P < 0.05$). There was no significant sleep window change.

CONCLUSIONS: Participants' eating pattern was improved over the 6-month intervention in the EMPOWER program. As intended, the eating window and evening snacking were decreased, whereas eating breakfast was increased. Participants who eliminated evening snacks were more successful in weight loss, suggesting the benefit of this approach.

Study Section: Microbiology

Microbial-derived aromatic amino acid metabolites modify inflammatory signaling and cellular energy status in a human monocyte cell line

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INTRODUCTION: Lactic acid bacteria (LAB) are found in fermented food and have been shown to enhance human health, but the mechanisms are diverse and not fully understood. An understudied property of LAB is the ability to metabolize aromatic amino acids into bioactive metabolites. We hypothesized that the aromatic amino acid xenometabolites of LAB, more specifically indole-3-lactic acid (ILA) and 4-hydroxyphenyllactic acid (4-HPLA), play a role in regulating the host immune function. Monocytes play a vital role in innate immunity and express receptors for both ILA and 4-HPLA. ILA binds to a nuclear receptor, aryl-hydrocarbon receptor (AhR), and both ILA and 4-HPLA bind to a G protein-coupled receptor, hydroxycarboxylic acid receptor 3 (HCAR3). We hypothesized that through binding of these two receptors, ILA and 4-HPLA would alter cellular energy status (cAMP) and attenuate inflammatory signaling of monocytes.

METHODS: Cell Culture. A transformed human monocyte cell line (THP-1) was cultured with either ILA (50 μ M), 4HPLA (50 μ M), or control (PBS), with added immune challenges of either lipopolysaccharide (LPS), Flagellin (FLG) or control (CON). Cells were collected at 6 hr and 24 hr and analyzed for inflammatory cytokine (il6, tnf) anti-inflammatory (il10), antioxidant (sod2) and AhR target genes (cyp1a1) by rtPCR. In a separate experiment, cAMP levels were measured in cell lysates with a competitive ELISA.

RESULTS: ILA and 4HPLA attenuated TNF α gene expression at 24 hr regardless of immune stimuli (main effect $p < 0.05$). HPLA attenuated tnfa expression at 6 hr in response to FLG challenge (treatment x FLG $p < 0.05$). Expression of other inflammatory (il6) or anti-inflammatory (il10) genes were not modified by ILA/4-HPLA ($p > 0.05$). Evidence of AhR binding by ILA was confirmed from an increased expression of Cyp1a1, an AhR target gene. Expression of superoxide dismutase 2 (SOD2), an antioxidant gene associated with the metabolism of superoxide, was also observed. cAMP levels were elevated by ILA and 4HPLA at 24 hr, indicating altered cellular energy status.

CONCLUSIONS: Microbial derived aromatic amino acids metabolites are understudied. Our in vitro work shows that two metabolites, ILA and 4-HPLA, attenuate inflammatory gene expression and alter cellular energy status in a human monocyte cell line. However, the precise mechanisms for how ILA and 4-HPLA interact with monocytes remains to be further investigated. Overall, these results indicate potential anti-inflammatory benefits of amino acid metabolites derived from LAB.

Dietary hesperidin and naringin inclusion affect gastrointestinal microbiota populations in a murine dextran sulfate sodium-induced colitis model

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INTRODUCTION: To evaluate the effect of citrus flavanones, hesperidin and naringin, on gut microbiota populations in a murine dextran sulfate sodium (DSS)-induced colitis model.

METHODS: Male C57BL/6J mice (n=48) fed an AIN93G (control) diet underwent a 14-d acclimation phase while housed in groups of 4; bedding was mixed and redistributed among all cages to normalize gut bacteria. Mice were then housed individually and randomly assigned to 3 treatment groups (n=16/diet): control, control + 50 mg/kg hesperidin (HS), or control + 50 mg/kg naringin (NG) for 14 d. After d14, each group (n=8) was randomly assigned to receive water or water containing 2.5% w/v DSS for 7 d. Mice were then euthanized, with tissues and cecal digesta collected. Fecal samples were collected after acclimation (d0), dietary intervention (d14), and DSS (d21). 16S rRNA Illumina sequencing and QIIME2 were used to assess fecal and cecal microbiota. Intestinal tissues were prepared for histology and histopathology scored by a blinded pathologist. Data were analyzed using Mixed Models procedure of SAS 9.4. The main effects of diet and DSS were tested, with significance set at $p < 0.05$.

RESULTS: DSS shortened cecal length and led to necrosis and immune cell infiltration, but diet was not protective. Based on the Shannon Index, DSS did not affect fecal alpha diversity, but cecal alpha diversity was lower in mice receiving DSS than those receiving water. Prior to DSS, over 10 bacterial taxa were affected by HS or NG consumption over time, including an increased ($p < 0.0001$) relative abundance of fecal *Enterohabdus* in mice fed HS and decreased ($p < 0.05$) relative abundance in fecal *Lachnoclostridium* in mice fed HS or NG. DSS treatment impacted the relative abundance of over 20 bacterial genera in fecal samples, including a large increase ($p < 0.0001$) in *Bacteroides* and large decreases ($p < 0.0001$) in *Lactococcus*, *Clostridium_sensu_stricto_1*, and *Romboutsia*. A few bacterial taxa had a diet*DSS interaction, including *Enterohabdus*, *Lactobacillus*, *Lachnoclostridium*, and *Oscillobacter*. Nearly 30 bacterial genera were impacted by DSS in cecal digesta samples, including large increases ($p < 0.01$) in *Akkermansia*, *Bacteroides*, and uncultured *Oscillospiraceae*, and decreases ($p < 0.01$) in *Muribaculaceae*, *Lactobacillus*, *Lachnospiraceae*, and *Roseburia*. Over 15 bacterial genera in cecal samples had a diet*DSS interaction.

CONCLUSIONS: Hesperidin and naringin did not protect from DSS-induced colitis, but impacted many bacterial taxa.

Gut microbiota profiling reveals a signature of microbiome dysbiosis associated with colitis development in the heterogenous nuclear ribonucleoprotein I (hnRNP I) knock out mice

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INTRODUCTION: hnRNP I is a polypyrimidine tract-binding protein that interacts with heteronuclear RNA (hnRNA). The function of this protein is related to mRNA maturity that could affect gene expression and protein translation. Recent studies suggest that ablation of the hnRNP I gene in intestinal epithelial cells in the knockout (KO) mice have caused spontaneous colitis. Therefore, the objective of this study is to examine gut microbiota quantity and diversity after the hnRNP I gene is knocked out in mice.

METHODS: Fecal samples of wild-type (WT) and KO mice were analyzed at 4-week and 6-week of age. At 6-week of age, bodyweight along with colon weight and length were collected after euthanasia. Fecal samples were saved at -80°C until DNA isolation. Fecal pellets (2 per animal/sampling) were extracted to obtain DNA. Gut microbiota was analyzed by qPCR using genus-specific primers. Results were analyzed using the Mann-Whitney test.

RESULTS: The abundance of gut microbiota was examined in the WT (n=21) and KO (n=26) mice. Genus-specific primers, including *Bifidobacterium* spp., *Enterococcus* spp., *Lactobacillus* spp., and *Clostridia* Cluster XIVa were used in analysis. In the KO mice, *Lactobacillus* spp. and *Enterococcus* spp. were more abundant at 6 weeks of age, comparing to the WT control mice. On the contrary, *Bifidobacterium* spp. showed no difference between KO and WT mice. Interestingly, the *Clostridia* Cluster XIVa was lower in abundance in the KO mice than WT at both 4 weeks and 6 weeks of age.

CONCLUSIONS: Results indicated that the KO mice had lower *Clostridia* Cluster XIVa than WT at both time points. *Clostridia* cluster XIVa belongs to butyrate-producing bacteria. These bacteria relate to mucin-adhered microbiota. This set of data aligns with the previously published results indicating that this cluster of bacteria may be essential to maintain gut homeostasis since they produce butyrate. Butyrate, one of the short-chain fatty acids, is essential for colonocytes as energy, which helps proliferate and maintain the colon from colorectal cancer. The reduction of *Clostridia* cluster XIVa in the hnRNP I knockout mice may be related to the susceptibility of the mice to the development of colitis. Further investigation will focus on the analyses of this bacteria cluster's role in maintaining gut homeostasis and the interactions in gut bacterial receptor-mediated host-microbe communications and colon resident immune systems in the KO mice model.

Fermented foods as a source of microbial-derived immunomodulatory xenometabolites

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INTRODUCTION: Lifestyle factors including diet and exercise influence the gut microbiota and production of downstream immune modulating bioactive ('xeno') metabolites. Indole-3-lactic acid (ILA) and 4-hydroxyphenyllactic acid (4-HPLA) are two microbial-derived aromatic amino acid (ArAA) metabolites that are potential regulators of host immune function and are upregulated in the serum of humans in response to exercise training. While the mechanisms underlying these effects remain unclear, ILA and 4-HPLA are produced through the metabolism of tyrosine and tryptophan by a shared microbial enzyme, phenyllactate dehydrogenase (fLDH) found in select lactic acid bacteria (LAB), many of which can be found in fermented foods. It is plausible to suggest that the presence of ILA and 4-HPLA producing LAB, such as *Lactiplantibacillus plantarum*, may be involved in the anti-inflammatory and immunomodulatory effects of fermented foods. This led us to hypothesize that select fermented foods containing *L. plantarum* also contain microbial-derived ArAA metabolites ILA and 4-HPLA.

METHODS: Fifteen commercially produced fermented foods were homogenized with a stomacher and diluted with PBS. Stomached samples were filtered and sent to the Metabolomics Core at UIUC for liquid chromatography/mass spectrometry (LC/MS) analysis to attain concentrations of microbial-derived ArAA metabolites. Serial dilutions of stomached samples were plated with a spiral platter and cultured in an anaerobic chamber for 48-72 hours. 34 bacterial isolates (at least 2 per food sample) as well as positive control strains of *L. plantarum* were transferred to MRS broth and cultured for an additional 48 hours prior to completing DNA isolation and qPCR to investigate which isolates contain *L. plantarum*.

RESULTS: Results indicate that select fermented foods contain microbial-derived ArAA metabolites including ILA and 4-HPLA. Highest aggregate levels of ILA and 4-HPLA were found in fermented carrots (19.735 μ M and 7.41 μ M, respectively), pickles (9.065 μ M and 6.51 μ M, respectively), and kefir (2.855 μ M and 12.75 μ M, respectively). qPCR analysis revealed *L. plantarum* was evident in many, but not all, fermented foods containing ILA and 4-HPLA. Pickles, Kimchi, and fermented beet products all contained *L. plantarum* isolates while having various levels of ILA and 4-HPLA. Further analysis is needed to determine whether other ILA and 4-HPLA producing LAB are present in fermented foods.

CONCLUSIONS: Fermented foods contain immunomodulatory xenometabolites and represent an ideal opportunity to further examine the significance of microbial-derived ArAA in humans. The amount of ILA and 4-HPLA found in select commercially available fermented food products did not correspond with the detection of *L. plantarum* in bacterial isolates of the foods as predicted, suggesting that additional LAB containing fLDH may be responsible for producing ILA and 4-HPLA in fermented foods. These data help to build a foundation for future studies exploring novel pathways by which select fermented food diets can promote immune modulating xenometabolites.

Study Section: Preclinical Metabolism

Progesterone stimulates glycogen breakdown in bovine uterine epithelial cells via membrane receptors

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INTRODUCTION: Pregnancy loss has been associated with abnormal secretion of glucose into the uterus lumen, but how glucose secretion is regulated to match the changing needs of the embryo is unclear. Glycogen is composed of thousands of glucose molecules, and it is present in the uterus of many species. Our lab has shown that the glycogen content of the bovine uterine epithelium was lower on Day 11 than Day 1 of the cycle. Therefore, our objectives were to 1) make immortalized bovine uterine epithelial (BUTE) and bovine uterine fibroblast (BFIB) cell lines to investigate the hormonal control of glycogen metabolism in the uterine epithelium and 2) to elucidate the role of progesterone on glycogen breakdown in the uterine epithelium.

METHODS: To generate BUTE and BFIB cells, fresh endometrial biopsies were collected from a Holstein dairy cow, digested into a single cell suspension, and plated to allow the formation of individual colonies. Cells were immortalized with large T-antigen, and individual colonies were isolated, expanded, and tested for keratin and vimentin. To determine the effects of progesterone on glycogen concentrations in uterine epithelial cells, BUTE and Ishikawa cells were treated with progestin agonists and antagonists as indicated. Glycogen was isolated with KOH, hydrolyzed to glucose, and then measured. To confirm the expression of membrane progesterone receptors (mPRs), uteri were collected on days 1 and 11 of the estrus cycle, embedded in paraffin blocks, and sectioned for immunohistochemistry.

RESULTS: Western blots and immunofluorescence showed that clone BUTE1A had a high expression of keratin with little expression of vimentin. Clone BFIB2 had a high expression of vimentin with no expression of keratin. Western blots confirmed that BUTE and BFIB cells expressed the enzymes necessary for glycogen metabolism and the nuclear receptors for estrogen and progesterone. Evaluating the effects of progesterone on glycogen levels, we found a high progesterone concentration (10 μ M) decreased glycogen levels in BUTE cells by 99% ($P=0.0002$). RU486 did not inhibit progesterone's effect in BUTE cells, indicating the effect of progesterone was not mediated by nuclear receptors. Therefore, we hypothesized that the effect of progesterone might be mediated by mPRs. RT-PCR confirmed that BUTE cells expressed all five mPRs (α , β , γ , δ , and ϵ). Similar to progesterone, a specific mPR agonist (Org OD02-0, 10 μ M) decreased glycogen levels in BUTE cells by 99% ($P<0.0001$). Confirming these results in a human model, progesterone and Org OD02-0 (10 μ M) decreased glycogen levels to a similar extent in Ishikawa cells ($P=0.0027$) that lack expression of the nuclear progesterone receptor. Indicating a potential for mPRs to regulate glycogen in vivo, immunohistochemistry showed that the bovine uterine epithelium expressed mPRs.

CONCLUSIONS: BUTE cells are a valid model to study the uterine epithelium. High concentrations of progesterone stimulated glycogenolysis and hence may play a role in providing glucose to the embryo. We are currently working to elucidate the downstream signaling pathways activated by mPRs that lead to the breakdown of glycogen.

Tissue lycopene accumulation in transgenic mice lacking one or both carotenoid cleaving enzymes

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INTRODUCTION: Lycopene is one of the most abundant carotenoids found in the body and its antioxidant properties have led researchers to explore its potential as a therapeutic modality for various diseases. Its metabolic pathways are largely undefined, but studies indicate that both β -carotene oxygenase 1 (BCO1) and β -carotene oxygenase 2 (BCO2) are involved in the cleavage of lycopene. This study aims to evaluate the impact of gender and ablation of β -carotene oxygenase 1 (BCO1), β -carotene oxygenase 2 (BCO2) or both on tissue accumulation in lycopene dosed transgenic mice. Previous work from our laboratories suggests that BCO2 appears to be the primary carotenoid cleavage enzyme for lycopene in vivo.

METHODS: Three-week-old C57BL/6 male and female mice (wild type [WT], Bco1^{-/-}, Bco2^{-/-}, Bco1^{-/-} X Bco2^{-/-} double knock out [DKO]) were divided into groups based on genotype (n=8 per group/per gender) and fed a powdered AIN 93G control diet for 2 weeks. Then the mice were gavaged daily for 2 weeks with 1mg of lycopene dissolved in cottonseed oil. At the end of two weeks, the mice were fasted overnight and sacrificed. Liver, serum, and extra-hepatic tissues were harvested. Tissues were preserved in liquid nitrogen and stored at -80 until analysis. Lycopene concentration was measured with high-performance liquid chromatography. Data analyses were performed using one-way ANOVA.

RESULTS: Except for serum, female mice had higher lycopene tissue accumulation, irrespective of genotype. On a concentration basis, liver, duodenum and adrenal lycopene were higher than other tissues. As expected, serum and tissues of DKO mice accumulated the highest lycopene. DKO mice had significantly higher lycopene than Bco1^{-/-} mice in the liver (p<.002), heart (p<.004), adipose (p<.03), and the testes (p<.004). Compared to Bco2^{-/-} mice, DKO mice had greater accumulation in the serum (p<.001), intestine (p<.04), heart (P<.0001), kidneys (p<.0001), adipose (p<.04), and testes (p<.0001). Liver (p<.007) and adrenal (ns) tissues in Bco2^{-/-} mice had higher levels of lycopene than Bco1^{-/-} mice whereas Bco1^{-/-} mice had significantly higher levels in the kidneys (p<.001) and tended to have greater accumulation in other tissues.

CONCLUSIONS: Accumulation of lycopene in tissues depended upon gender, genotype and tissue type. The abolition of both enzymes and mice of a female sex generated a higher accumulation. We are probing whether tissue-specific expression levels of BCO1, BCO2 or carotenoid transport proteins explain differential tissue accumulation across genotypes.

Estradiol may stimulate glycogen synthesis in the bovine uterine epithelium via insulin-like growth factor-1

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INTRODUCTION: Despite interventions and synchronization methods, early embryonic loss in cattle is 30-50%. Embryo survival and subsequent development are dependent on glucose. Post-conception, the embryo primarily uses pyruvate and lactate. At the morula stage, glucose uptake starts to increase dramatically. However, it is unclear how the bovine uterine epithelium meets the increasing glucose needs of the embryo. Research from our laboratory has found that the epithelial cells in the bovine uterus contain higher levels of glycogen, the storage form of glucose, on day 1 than day 11 of the cycle. During estrus, estradiol (E2) stimulates the production of insulin-like growth factor 1 (IGF1) in the uterine stroma of other species, which mediates some of the effects of estradiol. Therefore, our objective was to evaluate the effect of E2 and IGF1 on glycogenesis in immortalized bovine uterine epithelial (BUTE) cells.

METHODS: BUTE and bovine uterine fibroblast (BFIB) cells were maintained in alpha MEM media supplemented with E2, EGF, ITS, L-glutamine, and 10% FBS. Cells were treated as indicated for 48 hours and then collected for analysis. For glycogen, cells were lysed in 30% KOH, glycogen was hydrolyzed to glucose, and glucose was measured with a spectrometer. Metabolomic analysis was conducted by the U of I Metabolomics Core Facility.

RESULTS: Treatment of BUTE cells with E2 (0.1-10 nM) did not stimulate glycogen synthesis. In contrast, treatment of BUTE cells with IGF1 (50 or 100 ng/ml) resulted in a >2-fold increase in glycogen levels ($P < 0.01$). When treated with both E2 (100 nM) and IGF1 (50 ng/ml), only the main effect of IGF1 was significant ($P = 0.0001$). Elucidating the pathway by which IGF1 increases glycogen synthesis, western blots revealed an increase of approximately 7-fold for phospho-AKT after IGF1 treatment ($P < 0.05$) and an increase of >2-fold for phospho-GSK3 β ($P < 0.05$). IGF1 treatment also increased levels of hexokinase and glycogen synthase ($P < 0.05$). We next determined if BFIB cells produced IGF1 in response to E2. Treatment of BFIB cells with estradiol increased IGF1 production, and immunohistochemistry revealed that expression of IGF1 in the stroma was higher on day 1 than on day 11. This agrees with higher levels of glycogen in the epithelium on Day 1. Metabolomics (GS/MS) showed that IGF1 increased 3-phosphoglycerate, lactate, and N-acetyl-glucosamine in BUTE cells, suggesting increased glycolysis and hexosamine biosynthetic pathway activity. IGF1 also resulted in increased levels of glycosylated proteins in BUTE cells ($P < 0.01$).

CONCLUSIONS: This study demonstrated that glycogen synthesis is the result of IGF1 produced in the bovine endometrial stroma due to E2. IGF1 stimulated glycogen synthesis via an AKT/GSK3 β pathway and by increasing expression of glycogenic enzymes. Metabolomic (GS/MS) analysis indicated increased flux through glycolysis and the hexamine biosynthetic pathway. By increasing glycogen levels during estrus, the uterine epithelium may be better able to provide glucose to the elongating blastocyst. Identifying mechanisms of glycogen synthesis and catabolism in the bovine uterus may enable further research toward decreasing early embryonic loss in cattle.

α -Tocopherol restriction exacerbated lipopolysaccharide-induced inflammatory response in α -tocopherol transfer protein-null mice

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INTRODUCTION: Vitamin E (α -tocopherol, α T) can scavenge peroxy radicals as part of its antioxidant function, which is vital for the lipid-rich brain. α T appears to be particularly important for specific cells in the brain, such as cerebellar Purkinje neurons, and for lipid-rich myelin which wraps around axons and modulates signal transmission speed. A range of neurological symptoms, including ataxia and peripheral neuropathy, are common in humans and animals with severe vitamin E deficiency. The α -tocopherol transfer protein-null (Ttpa^{-/-}) mouse model is a valuable tool for studying the molecular and functional consequences of vitamin E deficiency. However, there is a need to establishing a timeline for the development of neurological phenotype in relatively young adult Ttpa^{-/-} mice fed vitamin E deficient diets. Our objective was to assess how dietary α T restriction, followed by lipopolysaccharide (LPS) exposure affected the inflammatory response in Ttpa^{-/-} and wild-type (Ttpa^{+/+}) mice.

METHODS: After weaning (3 weeks of age), male Ttpa^{+/+} and Ttpa^{-/-} littermates (n=36/genotype) were fed an α T deficient diet ad libitum for 4 weeks. At 7 weeks of age, mice were injected with LPS (1 or 10 μ g/mouse) or saline (control) intraperitoneally and sacrificed 4 hours post-injection. Brain and heart IL-6 levels, a marker of inflammatory response, and serum and tissue α T concentrations were measured via ELISA and HPLC-PDA, respectively. Hippocampal Il6, Tnf, and Gpx1 expression, markers of inflammatory and oxidative stress response, were measured via RT-qPCR, and blood immune cell profiles were measured via a hematology analyzer.

RESULTS: α T concentrations in serum and most analyzed tissues were below the limit of detection in Ttpa^{-/-} mice but not Ttpa^{+/+} mice. Circulating white blood cell levels, particularly lymphocytes, were lower in all LPS groups compared to controls (P < 0.01). The 10 μ g LPS groups had elevated IL-6 in the cerebellum and heart compared to controls, confirming an acute inflammatory response (P < 0.01). Hippocampal Il6 and Tnf expression were significantly increased in Ttpa^{-/-} mice that received 10 μ g LPS compared to those that received saline (~20- and ~3-fold higher, respectively) (P < 0.01). Comparing expression patterns by genotype, the 10 μ g LPS-Ttpa^{-/-} mice had ~2-fold lower Gpx1 and ~2-fold higher Il6 expression than the 10 μ g LPS-Ttpa^{+/+} mice. Hippocampal Il6 expression was increased by LPS in a dose-dependent manner (P < 0.05).

CONCLUSIONS: LPS, especially at the higher dose, altered inflammatory markers in the brain, heart, and serum. α T restriction further exacerbated the expression of select hippocampal genes.

Aberrant regenerating-islet derived (reg) gene expression is an early indicator of colonic dysplasia in heterogeneous nuclear ribonucleoprotein (Hnrnp I) knockout mice

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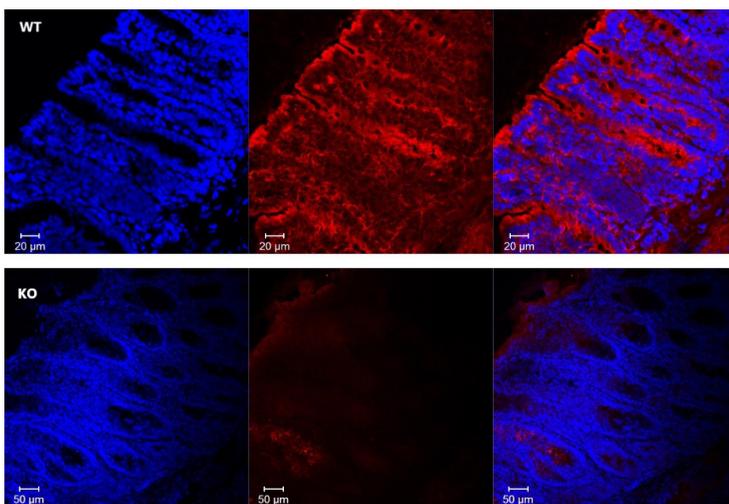
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INTRODUCTION: Reg is an evolutionarily conserved family of proteins with anti-inflammatory and antimicrobial functions, is mainly involved in the defense of intestinal epithelial cells (IEC). Recent studies indicate that the attenuation of colitis depends on the functions of Reg3b. Here, knockout mice with an IEC-specific ablation of the Hnrnp I gene (KO) were examined, which exhibited an increased risk of developing spontaneous colitis. The objective of this study is to examine the gene expression of the Reg genes in the KO mice. We hypothesize that epithelial ablation of Hnrnp I in the colon of the KO mice leads to early aberrant expression of Reg3b and Reg3g.

METHODS: Six weeks old WT and KO mice were sampled for body weight, colon weight and length. Whole colon was opened and jelly-rolled from distal to proximal end for embedding. Total RNA was extracted for quantification of gene expression using qPCR. Colon tissue sections were examined by hematoxylin-eosin staining (H&E) for colon morphology and colonic crypt length measurements. Colon localization of Reg proteins was analyzed using immunofluorescent staining. Results were analyzed using the Mann-Whitney test. Spearman test was used to analyze the correlation. P less than 0.05 is considered significant.

RESULTS: The expression levels of Reg3b, Reg3g, and Il-6 in KO mice were significantly lower than WT mice. Furthermore, expression levels of Reg3b and Reg3g are correlated in both WT and KO groups ($R=0.95$). It is worth mentioning that, in 30% of KO mice, the expression levels of Reg3b and Reg3g are significantly higher than the rest of the KO mice, exhibiting significant dichotomy. Histological analysis showed that the crypt lengths of KO mice were significantly longer than those in the WT mice both in the distal end and proximal colon. More importantly, the KO mice with elevated Reg gene expressions had longer crypt lengths in the distal end. Moreover, KO mice with elongated crypts also exhibited invasive dysplasia in the colon sections.

CONCLUSIONS: Our results indicate that IECs-specific Hnrnp I knockout abolished the expression of the Reg family genes. Further investigations are warranted to determine the potentially detrimental effects from the diminished Reg3 to the overall protection against microbial invasion in the colon. The contradictory upregulation of Reg3b and Reg3g in a subset of the KO mice indicates the extreme responsiveness to the perturbation from the Hnrnp I knockout and may serve as an early indicator of colon dysplasia.



The Gene Expression of the Reg Genes in Heterogeneous Nuclear Ribonucleoprotein (Hnrnp I) Knockout Mice is Significantly Higher than Wildtype Mice

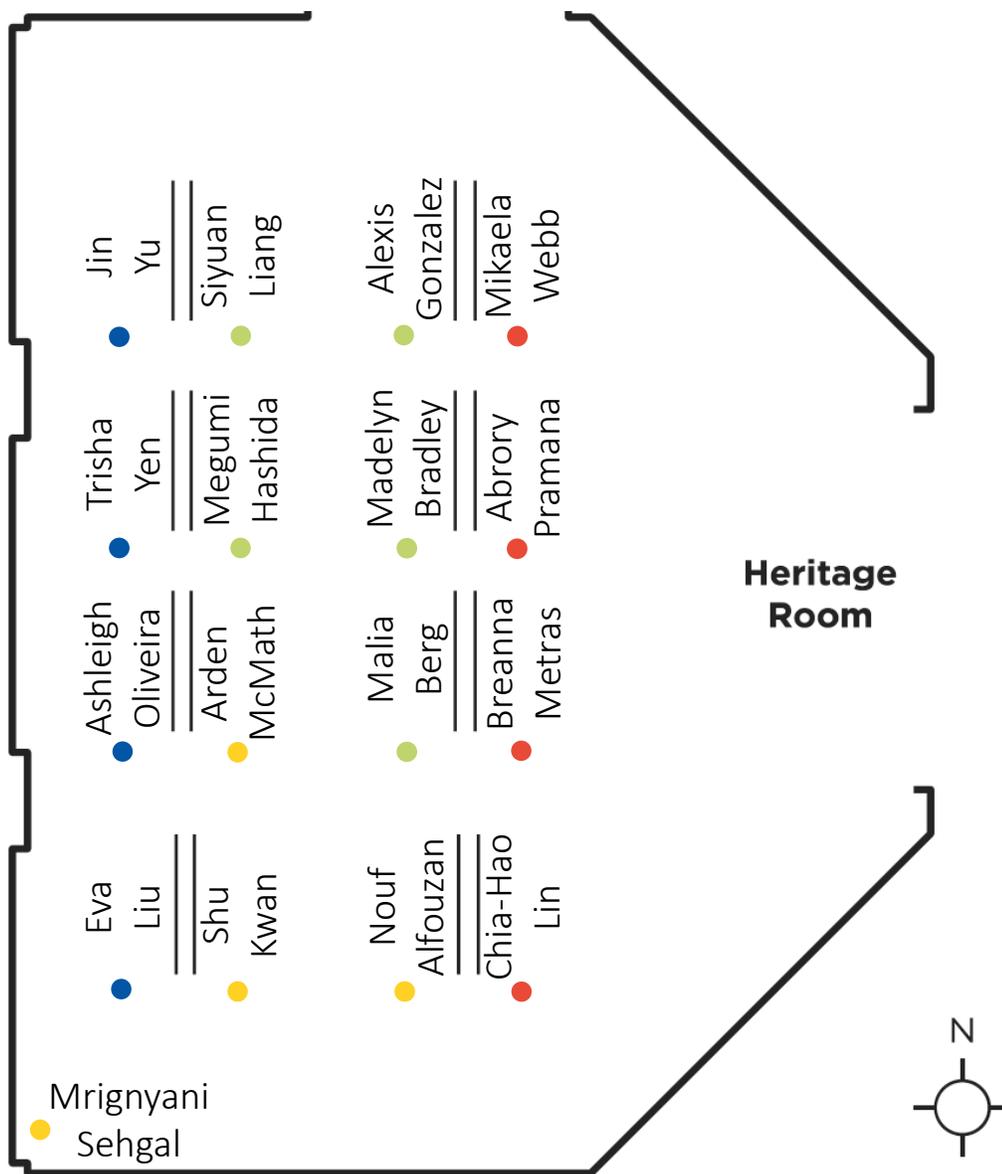
Research Image by Siyuan Liang

Location:

ACES Library, 1st floor

Heritage Room

5:15 – 6:45 pm



- Clinical Nutrition & Food Systems 1
- Clinical Nutrition & Food Systems 2
- Microbiology
- Preclinical Metabolism

