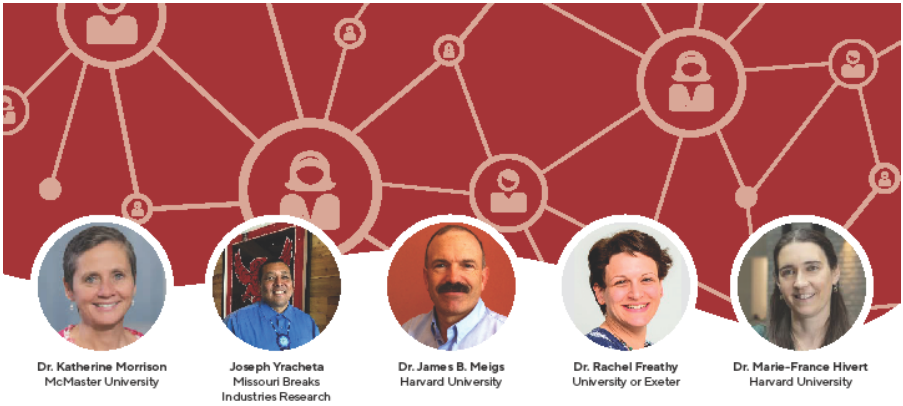

Dr. Katherine Morrison
McMaster University

Joseph Yracheta
Missouri Breaks Industries Research

Dr. James B. Meigs
Harvard University

Dr. Rachel Freathy
University or Exeter

Dr. Marie-France Hivert
Harvard University

9TH ANNUAL DIABETES RESEARCH SYMPOSIUM

Collaboration trumps competition: Lessons from scientific discoveries in type 2 diabetes

Event Details

Streamed via Zoom

18
November 2020
9:00AM - 12:00PM

19
November 2020
8:00AM - 2:00 PM

Agenda at a Glance

- Addressing obesity to prevent Type 2 diabetes: Making the Connection
  Dr. Katherine Morrison
- Civil Engagement and Big Data Research: How Tribal Repositories Can Potentially Broker Vital Partnerships
  Joseph Yracheta
- Translation of Type 2 Diabetes Genetics to Diverse Populations
  Dr. James B. Meigs
- iCARE Participant Advisory Group
- What can genetic studies of birth weight tell us about its relationship with later Type 2 diabetes
  Dr. Rachel Freathy
- Gestational diabetes and epigenetics
  Dr. Marie-France Hivert

Join via Zoom: https://us02web.zoom.us/j/86772614975?pwd=QnoxejNIOCtXaXZ2ZlhSblhIKysrQT09

DREAM

The Children's Hospital Research Institute of Manitoba

Rady Faculty of Health Sciences | University of Manitoba

DREAM Diabetes Research Envisioned and Accomplished in Manitoba 2020 SYMPOSIUM

Collaboration trumps competition: Lessons from scientific discoveries in type 2 diabetes

9th Annual Diabetes Research Symposium

Wednesday November 18th, 2020

Poster Session *Judges: Dr. Morrison, Mr. Yracheta, Dr. Meigs, Dr. Freathy, Dr. Hivert*	
9:00 – 11:00	**Individual poster presentations** – ZOOM
11:15-12:00	**Panel discussion**
*Announce Top 3 presentations	

Thursday November 19th, 2020

Pediatric Grand Rounds *Chair: Dr. Liz Sellers*	
8:00 – 9:00	7th Annual Dr. Heather Dean Lecture for Excellence in Pediatric Diabetes Research **Dr. Katherine Morrison** - Addressing obesity to prevent Type 2 diabetes: Making the Connection
Chairs: Dr. McGavock, Marylin Carino, Dr. Vern Dolinsky	
9:00 – 9:10	Opening Prayer by Knowledge Keeper Sherry Copenace
9:10 – 9:15	Opening remarks by Drs. Dolinsky and McGavock
9:15 – 9:40	**Joseph Yratcheta** – Civil Engagement and Big Data Research: How Tribal Repositories Can Potentially Broker Vital Partnerships
9:45 – 10:10	**Dr. James B. Meigs** – Translation of Type 2 Diabetes Genetics to Diverse Populations
10:10- 10:30	***BREAK***
10:30 – 11:00	**iCARE Participant Advisory Group**
11:05 – 11:30	**Dr. Rachel Freathy** – What can genetic studies of birth weight tell us about its relationship with later Type 2 diabetes
11:35-12:00	**Dr. Marie-France Hivert** – Gestational diabetes and epigenetics
12:00-12:20	***BREAK***
Top 3 poster presentations	
12:20-12:25	Presenter #1
12:25-12:30	Presenter #2
12:30-12:35	Presenter #3
12:35-12:45	Questions
12:45-1:05	***BREAK***
1:05 – 1:50	**Panel discussion**

WELCOME!

GREETINGS FROM THE CHRIM SCIENTIFIC DIRECTOR

Welcome to the 9th Annual DREAM Diabetes Research Symposium!

The DREAM symposium has become a platform to highlight recent discoveries by DREAM researchers and their trainees that lead to improvement of the health of children living with, or at risk for type 2 diabetes. In addition, the symposium provides an opportunity to hear from some of the world's best and brightest stars in diabetes and health research. This year's 9th annual symposium is focused on Nature or Nurture – Preventing & Treating Type 2 Diabetes in Youth. The symposium will begin with the 7th Annual Heather Dean Lecture in Excellence in Diabetes by Dr. Katherine Morrison from McMaster University. This will be followed by presentations from renowned keynote speakers, local patient partners in learning, and trainees. On behalf of CHRIM I genuinely invite you to enjoy the day and acquaint yourself in these and other exciting new areas of diabetes research.

Terry Klassen
CEO and Scientific Director, CHRIM

GREETINGS FROM THE DREAM CO-DIRECTORS

Welcome to the 9th Annual Diabetes Research Envisioned and Accomplished in Manitoba (DREAM) Symposium. This is our first Virtual Experience and we look forward to sharing this new format with colleagues across Canada. This year we are excited to introduce the topic of large scale collaboration and the importance of consortia in tackling challenges related to type 2 diabetes in youth. From big data within Indigenous communities, to the environmental and genetic underpinnings of the developmental origins of diabetes, large scale collaboration is the future of our work. We are pleased host an outstanding group of international and national speakers that span the continuum of research from basic discoveries to clinical investigation and patient engagement in pediatric diabetes. We are pleased to partner with the Department of Pediatrics on the 7th annual Dr. Heather Dean Lecture in Excellence in Diabetes Research, which will be given by Dr. Katherine Morrison, a clinician scientist and pediatric endocrinologist at the McMaster University.

We are also highlighting a new virtual format with the poster presentations this year, so be sure to attend the poster presentations Wednesday November 18th, as the top three will be selected to present their posters during the symposium. We hope that you will enjoy this symposium and that you will come away with a new appreciation of the forms of community- and patient-oriented research that are currently underway in the realm of diabetes in youth.

Vern Dolinsky & Jon McGavock
Co-Directors, DREAM

DR. HEATHER DEAN LECTURE

EXCELLENCE IN DIABETES

The 9th Annual DREAM Symposium marks the 7th Annual Dr. Heather Dean Lecture in Excellence in Diabetes. This lecture is named in honour of one of the University of Manitoba's most recognized and trailblazing clinician scientists. Dr. Dean has been a pillar in several communities in Manitoba for nearly 40 years, including but not limited to the medical community, the pediatrics and child health community, the farming community, the sporting community and most famously, the knitting community. Dr. Dean has inspired countless trainees, patients, and families during her tenure in the province. Without Dr. Dean's commitment to diabetes in children and vision for team-based care, the DREAM team would not exist. The Annual Dr. Heather Dean Lecture symbolizes the excellence in clinical care, research and interdisciplinary collaboration in the area of pediatric endocrinology that Dr. Dean has embodied and cultivated in the province of Manitoba. We hope that the lecture will also serve as an annual source of inspiration for young hearts and minds in the same way that Dr. Dean has inspired us over the past 30 years.

DR. KATHERINE MORRISON

Addressing obesity to prevent Type 2 diabetes: Making the Connection

Dr. Katherine Morrison is a pediatric endocrinologist, Professor and Associate Chair – Research in the Department of Pediatrics and Co-Director of the Centre for Metabolism, Obesity and Diabetes Research at McMaster University. She is a clinician researcher and is the Medical Director of the Pediatric Weight Management and Pediatric Lipid Clinics at McMaster Children's Hospital. Her research is centered around the etiology, consequences and treatment of obesity and lipid disorders in children. Dr. Morrison's work is supported by the Heart and Stroke Foundation of Canada, the Canadian Institutes of Health Research, CFI, HAHSO and the Ontario Ministry of Health. Perhaps most importantly – her first faculty position was in Winnipeg working with and learning from Heather Dean!

Objectives:

- Connecting in the clinic: Be aware of recent updates on the approach to the assessment and management of the child or adolescent with obesity
- Connecting with the science: Be familiar with the science related to emerging pathways associated with obesity in childhood and its consequences
- Connecting with each other: Consider ways we can connect to enhance knowledge translation, research and clinical care related to childhood obesity

KEYNOTE SPEAKERS

JOESEPH YRATCHETA

Civil Engagement and Big Data Research: How Tribal Repositories Can Potentially Broker Vital Partnerships

Joseph Yracheta is an Indigenous American and a working researcher at the Lakota community-based company called Missouri Breaks Industries Research, Inc. (MBIRI). Currently enrolled in the DrPH program at Johns-Hopkins, he studies the intersection of Environmental Health and Genomics.

Mr. Yracheta's main passion is the proper use of Genomics & Omics in the biomedical, anthropological, historical, cultural, legal and land rights jurisdictions of Native communities. Mr. Yracheta feels that in the burgeoning field of Precision Health and Genomics, all Amerindigenous people (including Latin American Indigenous & Mestizo groups) must engage to ensure return of economic, educational and health benefit. He feels that the most important outcome of his work would be to encourage & support the sustainability of Indigenous culture and sovereignty utilizing STEM & Big Data technologies to grow and advance the power of Tribal Nations as well as undo the 528 year history of Settler-Colonial injustice to America's First & descendant people.

DR. JAMES B. MEIGS

Translation of Type 2 Diabetes Genetics to Diverse Populations

Dr. Meigs is Professor of Medicine at Harvard Medical School and Massachusetts General Hospital. His group studies causes and prevention of type 2 diabetes and cardiovascular disease, approached using biochemical and genetic epidemiology of insulin resistance, type 2 diabetes and cardiovascular disease, and health services translational research to improve type 2 diabetes and CVD prevention and clinical care. He is active research mentor and also is a practicing primary care internal medicine physician at MGH.

iCARE PARTICIPANT ADVISORY GROUP

The iCARE (Improving Renal Complications in Adolescents with Type 2 Diabetes through Research) is a study that will help to explain why youth with type 2 diabetes (T2D) develop early kidney disease. The iCARE Participant Advisory Group works with youth and caregivers of youth living with type 2 diabetes in Manitoba to help make treatment and outcomes better for patients.

Jackie McKee, iCARE co-chair & advisor:

"I live in Waywayseecappo First Nation. I'm Ojibwe with Irish decent and I am a mother of 5 children. I was diagnosed with Type 2 diabetes 5 years ago and many of my family members live with this disease - but I do not let this disease define me. My oldest son was diagnosed with Type 2 diabetes 2 months before his 13 birthday. This was not a complete shock but devastating news for my son and my family. Throughout his teenage years he was seen at the Children Adolescent clinic at the Health Science Centre. As the co-chair of the iCARE Advisory Group I am very passionate about this research. I have been given a rewarding opportunity to contribute in a meaningful way to assist in the direction of type 2 diabetes research."

Jennifer Lopez, iCARE youth co-chair & advisor:

"I was born and raised in Winnipeg to parents who originated from the Philippines. I was diagnosed the summer right before I entered grade 10. I joined the PAG group because I want to help inform and educate individuals about type 2 diabetes especially in youth. I am currently in the Child and Youth Care at Red River College and I think through both being part of the PAG and as an upcoming child and youth care practitioner, I have a voice that I can use to advocate for young people living with type 2 diabetes."

Sydnee Monias, iCARE youth advisor:

"I live in Winnipeg and I'm Oji-cree. As far as I know, type 2 diabetes started with my grandmother and grandfather. Then my mom and 2 of my aunties were diagnosed, and then me. I love being a part of iCARE, I like the support it gives me and the help to understand more."

DR. RACHEL FREATHY

Early Growth Genetics Consortium – maternal and offspring genetic determinants of child obesity and early life growth

Rachel Freathy is an Associate Professor and Wellcome Trust Senior Research Fellow at the University of Exeter, United Kingdom. Her research aims to use genetics to understand the factors affecting the growth of a fetus in utero and the associations between birth weight and later life diseases such as Type 2 diabetes.

Rachel is leading genome-wide association studies and Mendelian randomization studies of birth weight and placental weight in the Early Growth Genetics (EGG) Consortium, an international collaboration of more than 150 investigators from more than 40 studies. These studies have recently identified 190 genetic loci associated with birth weight, highlighted genetic links with later life disease, and shown causal associations between maternal obesity-associated factors and birth weight.

DR. MARIE-FRANCE HIVERT

Gestational diabetes and epigenetics

Dr Hivert is an Associate Professor in the Department of Population Medicine at Harvard Medical School. She is a clinical investigator with primary focus on the etiology and primordial prevention of obesity and related co-morbidities, particularly type 2 diabetes and gestational diabetes. Her interests also include fetal metabolic programming mechanisms and the integration of genetics, epigenetics, and environmental factors contributing to obesity and related disorders. She is the Principal Investigator (PI) of Genetics of Glucose regulation In Gestation and Growth (Gen3G), and co-PI of Project Viva, two independent prospective pre-birth cohorts that investigate the health determinants of mothers and children. She is currently involved in many international consortia investigating the genetics determinants of glycemic regulation during and outside of pregnancy, as well as epigenetics mechanisms linked to prenatal exposures and fetal programming. At Harvard Medical School, she is the director of the Curricular Theme “Nutrition and Lifestyle Medicine” and the co-director or the Advance Integrated Science Course “Metabolism, Nutrition, and Lifestyle Medicine”.

ABSTRACT #1

INVESTIGATING THE MOLECULAR AND METABOLIC REGULATORS OF RHYTHMIC INSULIN SECRETION OVER 24 HOURS

Nivedita Seshadri and Christine A. Doucette

Chrono-disruption is an important risk factor shown to augment type 2 diabetes (T2D). Our lab has previously demonstrated that healthy β cells secrete insulin in a rhythmic, diurnal manner regulating glucose homeostasis; however, the molecular and metabolic factors that regulate rhythmic insulin secretion remain elusive. We hypothesize that the pancreatic β-cell clock controls temporal changes in metabolic pathways that regulate glucose-sensing signals and GSIS capacity.

Bmal1, a core circadian clock machinery component, was knocked down in MIN6 β-like cells using siRNA. Both control and Bmal1KD-MIN6 cells were then synchronized and used to interrogate changes in metabolic gene expression (Cpt-1, Slc2a2, Ldha, and Ucp2), glucose-stimulated ATP production and fuel oxidation (glucose and fatty acids; using the Seahorse XF-24) at 4 and 16 hours post-synchronization (ZT4 and ZT16, respectively).

We have established that Ucp2 expression is rhythmic over 24 hours and its rhythmic expression is essential for controlling daily rhythmic GSIS via glucose-stimulated ATP production and glucose-stimulated ROS production. Knock-down of Bmal1 chronically elevated Ucp2 ultimately impairing GSIS rhythms via reduction in glucose-stimulated ATP production. Notably, rhythmic Ldha expression (a disallowed gene) was also lost and similar results were observed for Slc2a2 and Cpt-1 expression. Examination of control MIN6 cells reveal that at ZT4 when GSIS capacity is highest, MIN6 cells show higher rates of glucose oxidation compared to ZT16 when GSIS is suppressed. Concomitantly, at ZT4, MIN6 cells have lower rates of fatty acid oxidation than at ZT16. Furthermore, when both Cpt-1a and Ucp2 were inhibited using Etomoxir and Genipin respectively, cells at ZT16 had significantly lower basal and maximal oxidation in the presence of fats when compared to control cells at ZT16.

Overall, our results demonstrate that diurnal rhythms of GSIS are driven by the circadian clock via temporal control of primary metabolic pathways. This study will help gain a mechanistic insight into β-cell function over 24hours and how these pathways are altered in T2D.

ABSTRACT #2

LARGE EXTRACELLULAR VESICLES (EVs) ARE POSITIVELY ASSOCIATED WITH THE EXERCISE RESPONDER PHENOTYPE IN ADOLESCENTS WITH OBESITY

Taiana M. Pierdoná, Alexandria Martin, Patience Obi, Samira Seif, Benjamin Bydak, Ashley Eadie, Keith Brunt, Jonathan M. McGavock, Martin Sénéchal, Ayesha Saleem

Introduction: Heterogeneity in response to exercise training exists, though the underlying mechanisms remain unclear. This variance in response to exercise training might be mediated through differential release of EVs. The purpose of this study was to evaluate if changes in EVs after acute aerobic exercise (AE) were associated with responder's phenotype following 6-weeks of resistance exercise training (6W-RT).

Methods: This is a single-blind secondary analysis of the EXIT trial (#02204670), which included 11 inactive adolescents (15.7±0.5 years) with obesity (BMI ≥ 95th percentile). Adolescents underwent a bout of AE (60% heart rate reserve, 45 min) at baseline and then a 6W-RT. Primary outcome was the categorization of responders (RE) or non-responders (NRE) based on changes in insulin sensitivity. The primary exposure variables were EV size, stability, and yield. EVs were isolated using size exclusion chromatography. These analyses were performed using blood samples collected at baseline (AT0), during (AT15,30,45) and after AE (AT120).

Results: Overall, there was a general increase in EV production in both groups with AE. Average EV size was larger in RE (~146.9 nm) vs. NRE (~124.1nm; $p<0.05$). Average EV size at AT0 was associated with absolute change in Matsuda index following 6-weeks of resistance training ($r=0.44$, $p=0.08$). EV size distribution revealed RE preferentially expressed EVs between 150 – 250 nm in size, whereas NRE expressed EVs between 50 – 150 nm ($p<0.05$). At baseline, RE-EVs contained ~25% lower TSG101 protein ($p<0.05$), ~85% higher MMP2 content, while CD63 levels remained unchanged. We do not know the mechanistic link between EV size/cargo, and the individual response to exercise.

Main Findings: Our data suggest that adolescents with obesity that respond to exercise training produce larger EVs, with lower exosome- and higher microvesicle-specific protein expression. RE-EVs also had higher EV protein yield during AE. EV size was positively associated with the responder phenotype.

ABSTRACT #3

A COMPARISON OF CLINICAL AND SOCIAL CHARACTERISTICS OF CANADIAN YOUTH LIVING WITH TYPE 2 AND TYPE 1 DIABETES

Marylin Carino, Yesmino Elia, Elizabeth Sellers, Jacqueline Curtis, Jonathan McGavock, James Scholey, Jill Hamilton, Cheril Clarson, Teresa Pinto, Stasia Hadjiyannakis, Luc Mertens, Constantine Samaan, Josephine Ho, Munier Nour, Constadina Panagiotopoulos, Mary Jetha, Melissa Gabbs, Farid H. Mahmud*, Brandy Wicklow*,Allison Dart*

Background: The objective was to describe the clinical and social characteristics of 2 Canadian cohorts of adolescents with diabetes.

Methods: Participants from the Improving Renal Complications in Adolescents with type 2 diabetes through Research (iCARE) study (n=322) and the Early Determinants of Cardio-Renal Disease in Youth with Type 1 Diabetes (n=199) study were compared.

Results: Adolescents were 14-15 years of age. The T2DM cohort had a shorter duration of diabetes. Both groups had HbA1c's above target. T2DM cohort predominantly Indigenous; T1DM 58.3% European/Caucasian, with a high proportion (41.7%) of visible minority groups (Afro-Caribbean, Asian/Pacific Islander, Hispanic). Prevalence of obesity, hypertension, left ventricular hypertrophy, albuminuria, and hyperfiltration are higher in T2DM cohort. The T1DM cohort was more socially and economically advantaged in all four dimensions of health inequalities.

Main Findings: There are significant differences in clinical and social characteristics of adolescents with T2DM and T1DM in Canada. Both have inadequate glycemic control with evidence of onset and progression of diabetes-related complications.

ABSTRACT #4

TUNING MUSCLE ENERGY HOMEOSTASIS: THE ROLE OF NIX IN MITOPHAGY AND ADAPTATION

Jared Field, Matthew Martens, Joseph Gordon

Introduction: Exercise is instrumental in reversing derangements to muscle metabolism, such as insulin-resistance during type 2 diabetes. Mitochondrial quality control is important for efficient use of metabolic fuel, but the mechanism remains unclear. We observed that deletion of the gene Nix, a regulator of mitochondrial clearance (mitophagy) and calcium signals, in muscle caused the accumulation of dysfunctional mitochondria in mice. The objective is to determine the role of Nix in regulating mitophagy and muscle adaption as part of muscle metabolism.

Methods: To determine the effect of Nix, the muscle-specific deletion of Nix in mice was achieved using Cre-lox recombination and human skeletal actin-Cre. To assess the mechanism of Nix, a cell culture model of C2C12 myotubes were stimulated with electrical pulses to induce contraction (1hr) then fluorescent microscopy or biochemical assays were performed.

Results: Deletion of Nix in the muscle of mice caused the appearance of ragged red fibers only in males (N=3, $p<0.001$). This phenotype is a diagnostic marker of myopathy caused by accumulation of dysfunctional mitochondria. Myotubes that underwent contractile activity increased expression of Nix (~1.9-fold, $p<0.05$) and increased mitophagy markers (P62, ~5-fold, $p<0.05$; LC3A-II, ~4-fold, $p<0.05$). The increase in mitophagy was cross-validated with a fluorescent mitophagy indicator (~1.4-fold, $p<0.05$); however, when Nix protein levels were reduced by sh-RNAs, the effect was blocked ($p<0.05$) implicating Nix as a mitophagy regulator. Additionally, nuclear calcium accumulation increased in response to contractile activity (~2-fold, $p<0.05$) and this response was blocked by partial loss of Nix ($p<0.05$). Nix regulates calcium-dependent transcription factors, activating NFATc3 (~3-fold, p,0.05) and deactivating HDAC5 (~0.5-fold, p-value) which target adaptive gene expression (myoglobin, ~4-fold, $p<0.05$).

Conclusion: Together these data show that Nix regulates both mitophagy and adaptive gene expression in muscle, both are important components of metabolism and are known to be disrupted during the onset of diabetes.

ABSTRACT #5

MULTI-OMICS PROFILING OF RAT OFFSPRING EXPOSED TO GESTATIONAL DIABETES REVEALS CARDIOMETABOLIC DISEASE DEVELOPMENT WITH AGE

Stephanie M. Kereliuk, Prasoon Agarwal, Gabriel M. Brawerman, Laura K. Cole, Bo Xiang, Mario A. Fonseca, Grant M. Hatch, Jonathan McGavock, Vernon W. Dolinsky

Introduction: Through unknown mechanisms, fetal exposure to gestational diabetes mellitus (GDM) increases the risk for cardiovascular disease development later in life. We hypothesize that fetal exposure to GDM induces alterations in cardiomyocyte metabolism and induces left ventricular (LV) dysfunction with age.

Methods GDM was induced in female rats with a high fat (45% kcal) and sucrose diet prior to mating, throughout pregnancy and lactation. Lean control females received a low fat (10% kcal) diet. Fetal rat ventricular cardiomyocytes (FRVC) were isolated from e20.5 offspring for U-13C glucose flux analysis and calcium handling. The cardiac transcriptome and metabolome were measured in 3-month old offspring. LV morphology and function were assessed in the offspring from e18 to 12-months of age by transthoracic ultrasound.

Results: Offspring exposed to GDM exhibited increased LV posterior wall thickness across their life course (fetal to 12-months of age; $p<0.05$) and impaired LV filling beginning at 6-months of age ($p<0.05$). Consistent with the development of diastolic dysfunction in vivo, alterations in calcium flux and re-uptake were observed in FRVC isolated from GDM offspring compared to Lean controls ($p<0.05$). When FRVC were treated with isoproterenol, U-13C glucose metabolic flux through glycolysis and the citric acid cycle was reduced in GDM offspring. In 3-month old offspring metabolomics revealed an altered acylcarnitine profile. These metabolic changes corresponded to altered gene expression patterns associated with glucose metabolism and fatty acid transport pathways (e.g. Irs2, Slc2a4, Pfkfb2, Pdk4 and Cpt1a) identified by RNAseq.

Conclusions: Multi-omic profiling revealed GDM-induced alterations in the cardiac gene expression profile leading to modified metabolite levels in the offspring. These alterations corresponded with mitochondrial dysfunction, impaired cardiomyocyte metabolic flux and contractility, in concert with LV hypertrophy and diastolic dysfunction in the rat offspring. Our findings identify several mechanisms that link early-life GDM exposure to the development of cardiovascular disease later in life.

ABSTRACT #6

DOES EXPOSURE TO DIABETES IN UTERO CAUSE GREATER SUSCEPTIBILITY TO HYPERTENSION IN OFFSPRING? A TRIANGULATION APPROACH

Nicole Brunton, Meghan Azad, Allison Dart, Jonathan McGavock

Background: Exposure to diabetes in utero is associated with adverse offspring cardiovascular health outcomes. The literature supporting the above statement has traditionally relied on correlative evidence and mechanistic animal models. Recent advances in analytical approaches provide new opportunities to use observational data to infer a causal link between exposure to diabetes in utero and offspring cardiovascular health in humans.

Objective: We will triangulate results from three complimentary study designs to test the hypothesis that exposure to diabetes in utero is a causal determinant of elevated blood pressure in adolescent offspring.

Methods: Study#1 will be a sibling analysis to minimize confounding from genetics and shared environments. We will create a sibling-paired birth cohort using administrative data from the Manitoba Centre for Health Policy to compare hypertension risk before 18 years of age between siblings that were differentially exposed to diabetes in utero. Study#2 will be a Mendelian Randomization study applied to a birth cohort created within the EArly Genetics and Lifecourse Epidemiology Consortium that will use maternal and paternal (negative control) genetic risk scores for diabetes as statistical instruments to test for unbiased associations with offspring blood pressure in adolescence. Study#3 will be a Recall – by – Genotype study for detailed phenotyping of offspring to account for measurement error and temporal variations in blood pressure. Offspring from the Manitoba site of the CHILD study will be recruited at 13 years of age based on their mother's genetic risk score for diabetes. We will compare 24-hour ambulatory blood pressure measurements and vascular morphology between offspring within the highest and lowest quintiles for their mother's genetic risk score for gestational diabetes.

Importance: The results from this project could further inform early life public health strategies to support women during their pregnancies as a population-based strategy to prevent heart disease later in life.

ABSTRACT #7

SKELETAL MUSCLE-DERIVED EXTRACELLULAR VESICLES (EVs) - UPTAKE KINETICS AND EFFECT OF CHRONIC EXERCISE

Patience O. Obi, Samira Seif, Carlynn Davidson, Ben Bydak, Joseph W. Gordon, and Ayesha Saleem

Introduction: All living cells release extracellular vesicles (EVs) that package biological cargo, and constitute one of the primary methods of cellular communication. EVs can be taken up by cells and can subsequently affect recipient cell function. We and others have shown that exercise evokes an increase in systemic EV release, and these EVs can potentially mediate pro-metabolic effects of chronic exercise (CE) such as an increase in mitochondrial biogenesis. We do not know the effect of CE specifically on skeletal muscle-derived EVs, nor the kinetics of EV uptake by skeletal muscle.

Methods: We precipitated EVs from conditioned media from C2C12 myoblasts (MBs) using total exosome isolation kit, and from myotubes (MTs) using differential ultracentrifugation, a gold standard of EV isolation. MB-EVs were labelled with PKH67 and specificity of labelling established. Next, we evaluated the effect of EV dose, treatment time (24-72 h) and freezing on EV uptake by MBs through fluorescent imaging. MTs were electrically paced (3h/d x 4d @ 14V) using C-PACE EM stimulator (IonOptix) to mimic CE in vitro, EVs isolated and characterized biophysically by size and zeta potential (stability) using dynamic light scattering.

Results: EV uptake was measured specifically via PKH67 labelling, and increased concomitantly with treatment time and EV concentration. One freeze-thaw cycle at -20ºC decreased EV uptake, though the effect of dose and time remained. CE produced smaller EVs (50-200 nm) vs. control (100-250 nm) that were equally stable.

Conclusions: Our data suggest that EV uptake by skeletal muscle significantly increases with time and concentration, can be measured specifically by PKH67 labelling, and is decreased after just one freeze-thaw cycle. CE reduced average size, but did not affect stability of skeletal muscle-derived EVs. The effect of skeletal muscle-derived EVs after CE on modulating recipient cell function (e.g. metabolism) remains to be elucidated.

ABSTRACT #8

THE EFFECT OF THE HNF-1α G319S VARIANT ON LIVER FUNCTION AND YOUTH ONSET T2D

Manuel Sebastian, Taylor S. Morriseau, and Christine A. Doucette

Background: Youth-onset Type 2 diabetes (T2D) in Anishininiiwuk (Oji-Cree) linguistic group of central Canada are among the highest in the world, and a genetic variant in the hepatic nuclear factor-1a (HNF-1a) gene, known as HNF-1a G319S, strongly associates with T2D in this population. HNF-1a is known to play an important role in the liver, and the liver plays a central role in controlling glucose homeostasis, but it is unclear how the G319S variant influences liver function. We hypothesize that the G319S variant promotes greater hepatic glucose production, and triglyceride storage, which historically may have conferred an advantage during states of fasting, but in modern times, promotes hyperglycaemia and T2D.

Methods: CRISPR/Cas9 gene editing was used to knock-in the HNF-1a G319S G>A single nucleotide substitution into C57BL6 mice. G/G (wild type), G/S (heterozygous) and S/S (homozygous) mice were fed chow diet, sacrificed at 6 months of age, and liver tissues were collected for gene expression, triglyceride content, and glycogen content measurements. Additionally, pyruvate tolerance tests were performed to assess endogenous hepatic glucose production capacity.

Results Lipogenic genes such as PPARG were elevated, and key lipolytic genes such as PPARa is down regulated in S/S mice. However, no differences in steatosis, liver triglyceride or glycogen content were seen across the three genotypes. Additionally, female S/S displayed elevated endogenous glucose production during a pyruvate tolerance test.

Conclusion: The HNF-1a G319S variant appears to trigger a shift in liver metabolism towards increased lipid synthesis, but not storage, and increased endogenous glucose production, at least in females on a chow diet.

ABSTRACT #9

INCREASED HEPATIC GLUCONEOGENESIS AND ALTERED HEPATIC LIPID METABOLISM CONTRIBUTE TO THE DEVELOPMENT OF GESTATIONAL DIABETES MELLITUS IN PREGNANT ADIPONECTIN KNOCKOUT MICE

Brittany L. Gruber, Laura K. Cole, Bo Xiang, Grant M. Hatch and Vernon W. Dolinsky

Introduction: Gestational diabetes mellitus (GDM) is a common pregnancy-related condition with implications for both maternal and neonatal health. Genetics and lifestyle both contribute to development of GDM, but evidence suggests that low levels of circulating adiponectin increases the risk for GDM. Adiponectin is a fat derived hormone that improves insulin sensitivity. We hypothesize that adiponectin deficiency causes fatty liver during pregnancy, contributing to the development of GDM.

Methods: We compared the glucose and insulin tolerance of pregnant (3rd trimester) adiponectin knockout (KO) (strain B6;129-Adipoqtm1Chan/J) and wild-type (WT) mice, and assessed parameters of hepatic metabolism, mitochondrial function and fatty acid metabolism. Impact of adiponectin supplementation was measured by administering adenovirus mediated full length adiponectin at the end of the second trimester and comparing to control containing GFP.

Results: In the third trimester, fasting pregnant adiponectin KO mice are hyperglycemic on a low-fat diet (9.2mmol/L vs. 7.7mmol/L in controls, $p<0.05$) and glucose intolerant relative to WT controls. Adiponectin KO mice have elevated gluconeogenesis (determined by pyruvate tolerance test), which is accompanied by increases in hepatic gluconeogenic genes such as Pck2 (>2 fold) which is not responsive to insulin. Pregnant adiponectin KO mice also develop hepatic steatosis, and a 3-fold elevation in hepatic triglycerides ($p<0.05$) relative to wild-type. Gestational weight gain and food consumption were similar in KO and wild-type mice. Adenoviral-mediated adiponectin supplementation to pregnant adiponectin KO mice improved glucose tolerance, prevented fasting hyperglycemia, suppressed hepatic gluconeogenesis and attenuated fatty liver development.

Conclusion: Adiponectin deficiency is associated impaired insulin sensitivity and hepatic steatosis during pregnancy. Consequently, adiponectin deficiency contributes to insulin resistance and hyperglycemia characteristic of GDM. Adiponectin supplementation rescues the effects of adiponectin deficiency on insulin sensitivity and hepatic lipid metabolism.

ABSTRACT #10

UNRAVELLING INTERACTIONS BETWEEN SEX, DIET, AND GENETICS USING EXPERIMENTAL MODELS OF EARLY-ONSET TYPE 2 DIABETES

Taylor S. Morriseau, Kristin L. Hunt, Cuilan Nian, Vernon W. Dolinsky, Francis Lynn, Christine A. Doucette

Background: Manitoban Indigenous youth, particularly female adolescents, experience the highest rates of early-onset type 2 diabetes in Canada. 40% of these youth harbor the HNF-1αG319S gene variant that appears to drive pancreatic β-cell dysfunction compounded by modern dietary stress. We hypothesize that the HNF-1αG319S variant alters β-cell metabolism and insulin secretion in a sex- and diet-dependent manner.

Objective: To define how the HNF-1αG319S variant interacts with metabolic fuels to modulate pancreatic β-cell function and whole-body metabolism.

Methodology: CRISPR/Cas9 was used to knock-in the G>A.955 substitution into MIN6 β-cells ("G319S-MIN6") and C57/BL6 mice (G/S and S/S) compared to control (G/G). In vitro, glucose-stimulated insulin secretion (GSIS) and Seahorse oxygen consumption assays were performed. In vivo, body weight, insulin sensitivity, and ex vivo GSIS were assessed in both sexes.

Results: Glucose-stimulated insulin secretion (GSIS) in female G/S islets was reduced 3.5-fold relative to G/G mice, but this impairment was not observed in G319S-MIN6 or in male islets. Instead, basal insulin secretion (BIS) was suppressed (>2.8-fold) in male-derived models, potentially driven by increases in fatty-acid β-oxidation in the fasted state (1.5-fold elevation). At the whole-body level, suppressed BIS in male G/S mice translated into reduced fasting plasma insulin (3.4-fold) and elevated ketones (2.6-fold), with no changes in body weight or insulin sensitivity.

Conclusions: In female G/S mice, impaired GSIS may contribute to accelerated diabetes onset under modern dietary stress. This will be assessed by placing G319S-expressing mice on a "Western" high-carbohydrate diet. In male-derived models, the G319S variant shifts β-cell metabolism to mimic a fasted state, resulting in elevated fatty-acid oxidation and suppressed BIS, potentially triggering ketogenesis and reducing systemic glucose utilization. Whether a high-fat, low-carbohydrate diet (one that resembles the content of traditional food sources) better aligns with metabolic dependencies in male and female G319S-expressing mice will be further evaluated.

ABSTRACT #11

THE NEXT GENERATION BIRTH COHORT: SCREENING FOR DYSGLYCEMIA AND ALBUMINURIA IN HIGH RISK CHILDREN

Yash R. Rawal, Elizabeth A.C. Sellers, Allison Dart, Christy Pylypjuk, Brandy A. Wicklow

Introduction: Youth onset type 2 diabetes (T2D) is increasing worldwide. The prevalence of dysglycemia and microalbuminuria prior to diagnosis remains unclear. This project examined the presence of dysglycemia and albuminuria prior to diagnosis in youth with T2D and compared rates of albuminuria in youth onset T2D compared to normoglycemic youth.

Methods: A sub-analysis of data from a prospective cohort of children at risk of T2D (The Next Generation birth cohort) was performed. All children had a biennial orange glucose tolerance test (OGTT) starting at age 7 and annual albumin-creatinine ratio at 12 months. We looked at a 3-year period prior to diagnosis for youth developing T2D and normoglycemic youth matched for age. Descriptive analysis included proportions, means and medians.

Results: Of the 118 participants (11.2 ± 2.7 years, 100% First Nations, 51% female, 39% T2D), 22% of children with T2D had microalbuminuria at diagnosis compared to the 7% of normoglycemic children. Only one participant had microalbuminuria prior to diagnosis. Participants who had albuminuria at diagnosis had a fasting blood glucose (BG) of 14 mmol/L, 2-hour post 75g OGTT BG of 18.85 mmol/L and A1C of 9.23%. None of the children who developed T2D had impaired glucose tolerance or impaired fasting glycemia detected on biennial OGTT in the 3 years prior to diagnosis.

Conclusions: Although the children appear to be at greater risk of diabetes related complications, unlike adults, children are diagnosed without a significant period of preceding dysglycemia. In addition, almost a quarter of the children develop albuminuria co-incident with diabetes onset.

ABSTRACT #12

GESTATIONAL DIABETES EXPOSURE ALTERED CARDIAC MITOCHONDRIAL PROTEIN ACETYLATION IN THE MOUSE OFFSPRING

Mateusz M Tomczyk, Bo Xiang, John A. Wilkins, Vernon W. Dolinsky

Background: Gestational diabetes mellitus (GDM) affects 5-10% of pregnancies and puts offspring at risk for cardiovascular complications later in life. Previously we showed that GDM induced cardiac hypertrophy and mitochondrial dysfunction in the offspring. Reduced cardiac energy production resulting in an inability to pump blood as well as increased oxidative stress contributes to the development of diabetic cardiomyopathy. SIRT3 is the main mitochondrial lysine deacetylase that controls the acetylation of mitochondrial proteins and their enzymatic activity. SIRT3 protein levels are reduced in the hearts of GDM offspring. We hypothesize that decreased SIRT3 in the hearts of GDM offspring alters the acetylation of mitochondrial proteins and contributes to mitochondrial dysfunction.

Methods: Female mice were fed a high-fat high-sucrose (HFS) diet to induce GDM for 6 weeks prior to pregnancy. Lean control dams received a low fat (LF) diet. Four weeks after birth, male offspring were weaned and randomly assigned to HFS or LF postnatal diets. Mice were sacrificed and cardiac mitochondria were isolated. An anti-acetylated lysine antibody was used to enrich for tryptic digested peptides containing Acetyl-K that were analyzed by mass spectrometry (QTRAP LC-MS/MS, n=5).

Results: MS data of mitochondrial cardiac peptides revealed maternal and postnatal diet-induced alterations in the acetylation of peptides corresponding to proteins involved in cardiac energy production and oxidative stress resistance. Specifically, offspring of GDM dams exhibited hyperacetylation of Idh2, Aco2, Pdha1 which was attenuated in offspring from Lean dams, suggesting a maternal effect. Postnatal HFS diet in the Lean and GDM offspring was associated with hyperacetylation of Etfdh, Acadvl, Hadha, which was prevented by a LF postnatal diet.

Conclusion: GDM exposure and postnatal HFS diet induces differential acetylation of peptides involved in important mitochondrial processes when compared to LF control diet. Differential acetylation of these proteins could contribute to mitochondrial dysfunction present in diabetic cardiomyopathy.