Use this page to provide an abstract (up to 350 words) outlining the research described in your poster. **This OUTLINE MUST BE APPROVED AND SIGNED BY THE DEPARTMENT HEAD OR MAJOR PROFESSOR.** (Note: *email notification by your advisor to Ms. Debbie Koch can be substituted for the signature)*

The goal of my current proposed study is to investigate how dietary polyphenols may be used to prevent the adverse of multigenerational consequences of maternal obesity(OB). Using a OB murine model induced by high-fat high-sugar (HFHS) diet, I am testing the hypothesis that grape polyphenols can alter the gut microbiota to improve the maternal metabolic profile and prevent adverse maternal and fetal outcomes.

In the US and developing countries 60% of reproductive-age women are overweight or OB, which are main risk factors for gestational diabetes (GD). Unmanaged OB or GD predisposes women to developing type-2 diabetes (T2D) after the pregnancy. Due to increased placental transport of nutrients in the intrauterine environment, offspring may develop hyperinsulinemia and perinatal/neonatal complications such as macrosomia, dystocia, hypoglycemia, respiratory distress syndrome, and jaundice. Due to epigenetic changes promoted by an adverse intrauterine environment, offspring are at risk for developing OB and T2D prior to puberty. Epidemiological evidence shows a positive correlation between consumption of polyphenol-rich foods (e.g. fruits, vegetables, teas, spices) and protection from metabolic disease. Polyphenols may have therapeutic value for OB and GD, but efficacy and safety of polyphenols during pregnancy remains to be investigated.

Plant-derived bioactive compounds, such as grape polyphenols (GP), have been shown to have anti-hyperglycemic, antioxidant, and anti-inflammatory effects. In previous work, male mice fed a high-fat diet (HFD) supplemented with GP showed attenuated body weight gain, adiposity, intestinal and systemic inflammation, and improved glucose tolerance. These metabolic improvements were accompanied with a bloom in Akkermansia muciniphila, a gut microbe associated with metabolic resilience in mice and humans (REF). I will investigate whether GP may be able to attenuate the symptoms of OB in HFHS-diet-fed dams, regulate the placental transport of nutrients, and improve fetal outcomes.

The first experiment was initiated in summer 2018. To induce the OB phenotype, female mice were fed HFHS or HFHS diet formulated with GP (HFHS-GP) and given water with 20% sucrose, as previously described (REF). Controls groups will consist of female mice fed low-fat diet (LFD) and water. Body weights were monitored, oral glucose tolerance tests (OGTT) were performed, and fecal samples were collected before and during diet intervention. When mice fed HFHS or HFHS-GP showed 25% weight gain they were mated with males. On GD 17.5 dams were euthanized. Placenta and yolk sac were collected for gene/protein expression analysis of glucose, lipid acid and amino acid transporters. Fetal weights were measured. Maternal intestinal tissues and liver were harvested for gene expression analyses related to lipid and glucose metabolism and inflammatory cytokines. Cecal short chain fatty acids and serum bile acids will be analyzed by our established GC-MS and LC-MS methods.

Based on data, mice fed with HFHS-GP gained less body weights than ones fed with HFHS. Akkermansia muciniphia bloomed in the group of HFHS-GP, which is in accordance with expectance. I have been working to measure the level of protein expression and transcription of nutrient transporters, using western blot and Taqman assay, respectively. Also, other metabolic makers will be detected, and serum bile acids will be analyzed to explain the mechanism of GP on maternal obesity.

The above, proposed plan of research is approved and accepted.

DATE SIGNATURE OF DEPARTMENT HEAD OR MAJOR PROFESSOR - PRINT NAME

Effects of grape polyphenols in a murine model of maternal obesity

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Background

In the US and developing countries 60% of reproductive-age women are overweight or obese (OB), which are main risk factors for gestational diabetes (GD), defined as glucose intolerance developed during pregnancy . Maternal overweight/obesity (OB) and GD are independently associated with high risk of adverse maternal and fetal outcomes, with combined OB-GD having worse outcomes. OB or GD-related pregnancies result in increased placental transport of nutrients, which can lead to fetal hyperinsulinemia and perinatal/neonatal complications (e.g. macrosomia, dystocia, hypoglycemia, respiratory distress syndrome, jaundice). Due to epigenetic changes promoted by an adverse intrauterine environment, offspring are at risk for developing OB and type-2 diabetes prior to puberty. Except for lowering carbohydrate intake, there are no specific dietary guidelines for pregnant women with OB or GD.

Epidemiological evidence shows a positive correlation between consumption of polyphenol-rich foods (e.g. fruits, vegetables, teas, spices) and protection from metabolic disease. The efficacy and safety of polyphenols during pregnancy requires investigation. Male C57BL/6J mice fed HFD supplemented with grape polyphenols (GP) had decreased inflammation, improved oral glucose tolerance, and gut microbiota changes consistent with metabolic health. The goal of the present studies was to investigate effects of GP supplementation in pregnant dams fed high-fat, high-sugar (HFHS).

Methods

Female wild type C57BL/6J mice were fed a HFHS (46% kcal fat, 30% kcal sucrose) diet or HFHS diet supplemented with GP (HFHS-GP). All mice received 20% sucrose-containing water. A control group of females were fed LFD (15% kcal fat, 1% kcal sucrose). Body weight was monitored twice a week. HFHS-fed females were mated with males when they showed a 25% increase in body weight. As a matched control, a LFD-fed female was also mated at this time. Oral glucose tolerance tests were performed on Day 21 after diet intervention and on and gestational day 15.5 (GD 15.5). Fecal samples were collected at baseline, D14, and GD 17.5 for gut microbiota analysis. On GD 17.5, dams were sacrificed. Serum and tissues including placenta, liver, intestinal segments, livers and brains were collected. Liver weights, fetal weights, and relative abundance of *Akkermansia muciniphila* was measured.

Experiment design

Results

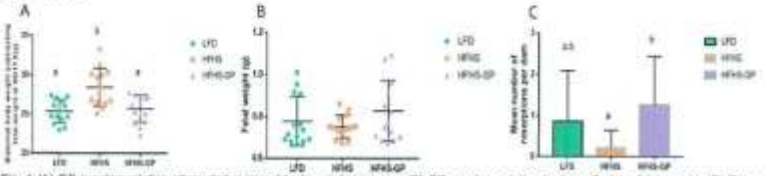

Fig. 1 (A) GP supplementation attenuated maternal body weight increase. (B) GP supplementation had no effect on fetal weight. (C) Fewer resorptions were observed in HFHS group. Significant differences were detected by one-way anova followed by Tukey's test. Same letter means no significant differences and different letters indicate significant differences, p<0.05.

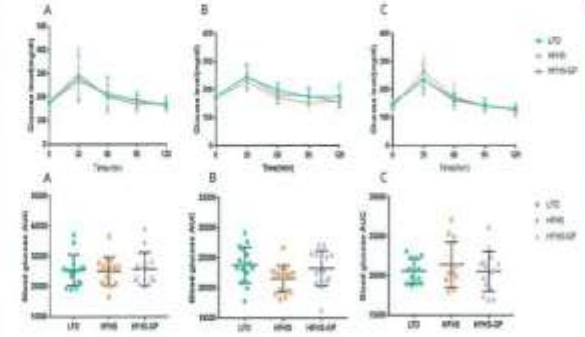

Fig.2 Oral Glucose Tolerance Test at: (A) baseline (B) 3 weeks on indicated diets and (C) GD 15.5. Differences in glucose metabolism were not observed.

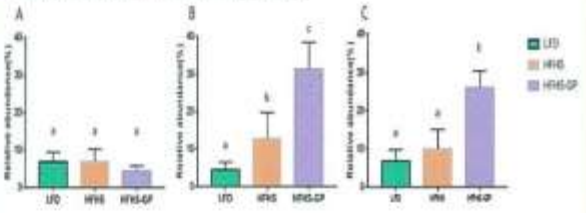

Fig. 3 GP induced an increase in the relative abundance of *Akkermansia muciniphila*. qPCR was performed on genomic DNA extracted from fecal samples collected at (A) baseline, (B) D14 on indicated diets and (C) GD17.5 and using universal primers and primers specific to *A. muciniphila*. Significant differences were detected by one-way anova followed by Tukey's test. Same letter means no significant differences and different letters indicate significant differences, p<0.05.

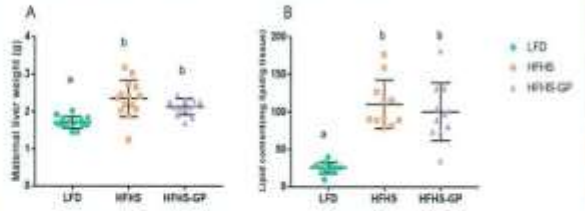

Fig. 4 (A) Maternal liver weight. (B) Lipid content in maternal livers was determined by Folch's method. Significant differences were detected by one-way anova followed by Tukey's test. Same letter means no significant differences and different letters indicate significant differences, p<0.05.

Conclusions

GP supplementation of HFHS diet alleviated maternal weight gain and increased the relative abundance of *Akkermansia muciniphila* which is associated with improvement of metabolic profile.

Mean fetal weight and number of fetuses per dam were similar in the LFD, HFHS and HFHS-GP groups. Fewer resorptions occurred in HFHS group.

Future work

Anatomy of placenta and factors involved in placental nutrient transport

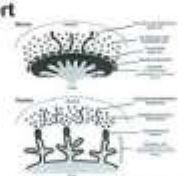

Fig. 5 Comparative anatomy of the human and mouse placenta. Adapted from "Genes, development and evolution of the placenta," by Cross, J. C., et al, 2003, *Placenta*, 24(2), p.123-130. Copyright 2002 by Elsevier Science Ltd. Adapted with permission.

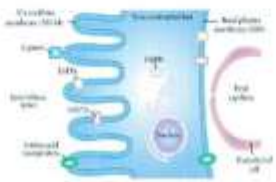

Fig. 6 Transporters mediating the transfer of amino acids, glucose (GLUTs), and fatty acids (FATPs) are expressed in both plasma membranes of the syncytiotrophoblast. Extracellular lipases release fatty acids from maternal lipoproteins and intracellular binding proteins (FABPs) guide the fatty acids within the cytosol of the syncytiotrophoblast. Adapted from "Regulation of nutrient transport across the placenta," by Lager, S., & Powell, T. L., 2012, *Journal of pregnancy, 2012, 179827*. Copyright 2012 by S. Lager and T. L. Powell. Adapted with permission.

1. The effect of GP on gene/protein expression of nutrient transporters in placenta and small intestine of dams will be investigated.

Perform qRT-PCR to detect changes in:
a). nutrient transporters involved in in glucose, amino acid and lipid transport in placenta(GLUT3, LAT1, LAT2, FATP1), liver(SGLT1 and GLUT2) and jejunum(LAT1, LAT2 and GLUT2).
b). Inflammatory markers in colon and liver tissue

2. Serum concentration of triglycerides, cholesterol, insulin and inflammatory markers will be measured.

References

Lager, S., & Powell, T. L. (2012). Regulation of nutrient transport across the placenta. *Journal of pregnancy*, 2012, 179827.

Dubé, E. *et al.* (2012). Modulation of fatty acid transport and metabolism by maternal obesity in the human full-term placenta. *Biology of reproduction*, 87(1), 14-1.

Dalhaes, F. *et al.* (2018). Altered maternal and placental lipid metabolism and fetal fat development in obesity: Current knowledge and advances in non-invasive assessment. *Placenta*, 69, 118-124.

Roopchand, D. E., *et al.* (2015). Dietary polyphenols promote growth of the gut bacterium *Akkermansia muciniphila* and attenuate high fat diet-induced metabolic syndrome. *Diabetes*, 64, 2847-2858.