Summary of Progress

The hypothesis for my 2022 Preeclampsia Foundation Vision Grant was that cell-free DNA (cfDNA) would show epigenetic changes indicative of oxidative stress and accerlerated placental aging in preeclampsia pregnancies. Our objective was to use genome-wide DNA methylation data and computational approaches to assess molecular differences in early-onset preeclampsia (early-PE) and late-onset preeclampsia (late-PE) pregnancies compared to controls. Specifically looking at differences in mitochondrial cfDNA in relation to oxidative stress. We are also evaluating epigenetic aging in a cfDNA context, which has never been done before. We are currently assessing these differences through the following specific aims:

Aim 1. Assess DNA methylation patterns of mitochondial cfDNA present in early-PE and late-PE compared to control pregnancies.
Aim 2. Build a cfDNA epigenetic clock to predict gestational age and evaluate whether we observe accelerated aging in early-PE and late-PE.

General progress

The first step to completing the proposed research was to transfer the cell-free methylated DNA immunoprecipitation-sequencing (cfMeDIP-seq) data from University Health Network (UHN) to McMaster University. This look several months as UHN and McMaster legal teams had to negotiate a Data Transfer Agreement (DTA). The DTA was completed and all data was transferred by April 2023. We hired an 8-month co-op student with extensive coding experiences and biology knowledge to complete the proposed work. Our co-op student started May 1st, 2023, and has been working on aim 1 for the past couple months. We also purchased a a computer workstation for Simon to use to complete to proposed work.

Assess DNA methylation patterns of mitochondial cfDNA present in early-PE and late-PE compared to control pregnancies

We first assessed the amount of cell-free mitochondrial DNA present in circulation of healthy control pregnancies and pregnancies subsequently impacted by early-PE, late-PE, and intrauterine growth restriction (IUGR). We found no significant differences in total cell-free mitochondrial DNA (Figure 1).

We also separated pregnancies with male and female fetuses to assess whether male (Figure 2) and female (Figure 3) pregnancies differ.

Again, we found no significant differences in each male and female fetuses when assessing total cell-free mitochondrial DNA in early-PE, late-PE, and IUGR compared to controls. We are currently assessing whether there will be differences in total cell-free mitochondrial DNA in early-PE, late-PE, and IUGR compared to controls when adjusting for body mass index (BMI), gestational age at blood draw, and smoking status.

We are also currently looking for changes in DNA methylation in early-PE, late-PE, and IUGR associated with hypoxia, oxidative stress, and mitochondrial function using a candidate gene approach. These genomic regions will include mitochondrial DNA regions as well as autosomal DNA regions.

Plans for July 2023 - December 2023

For the remainder of 2023, our co-op student will complete aim 2 of the proposed research. We also have plans to follow up aim 1 with wet-lab experiments. This project has also developed into

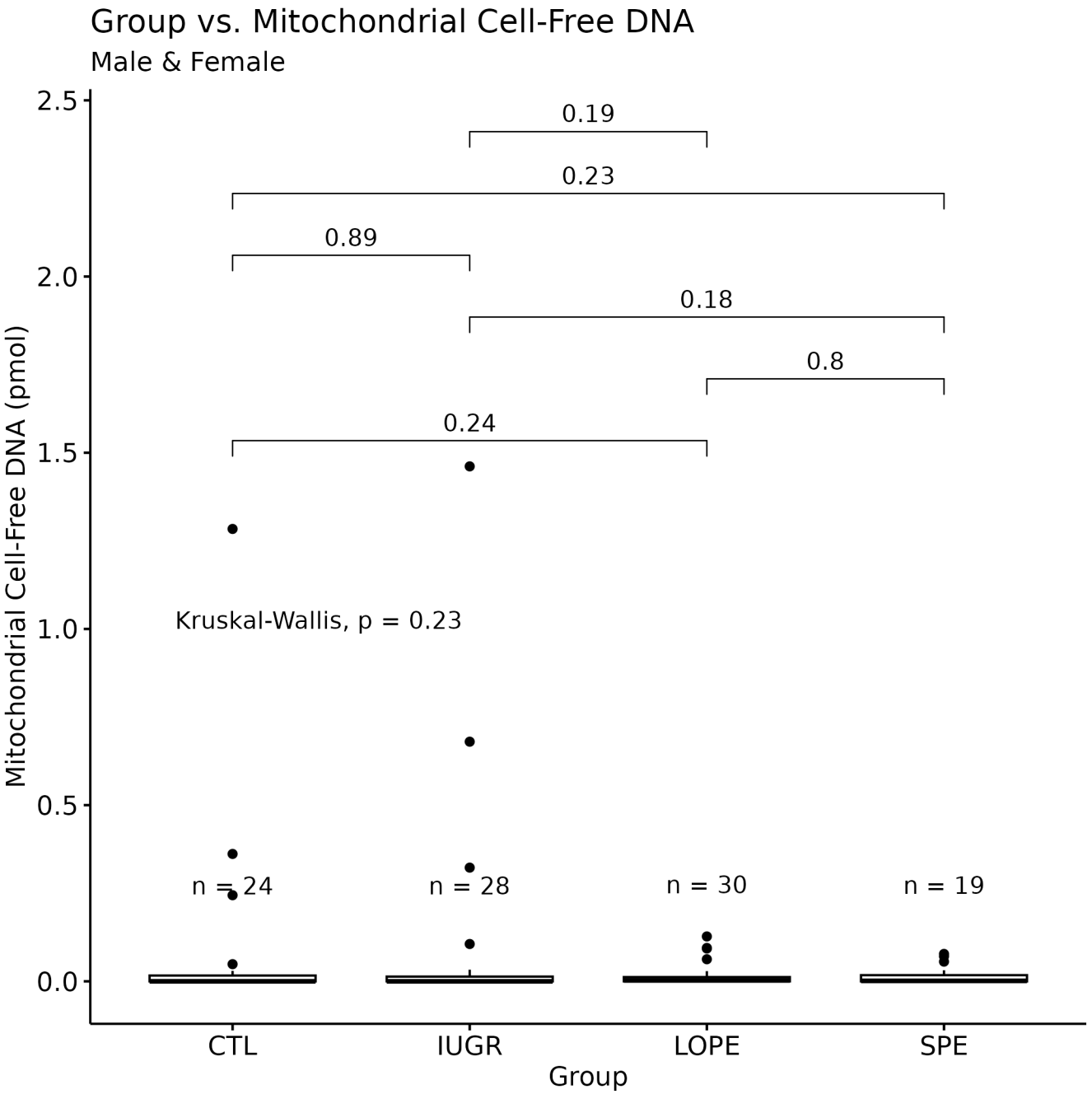

Figure 1: Differences in total cell-free mitochondrial DNA.

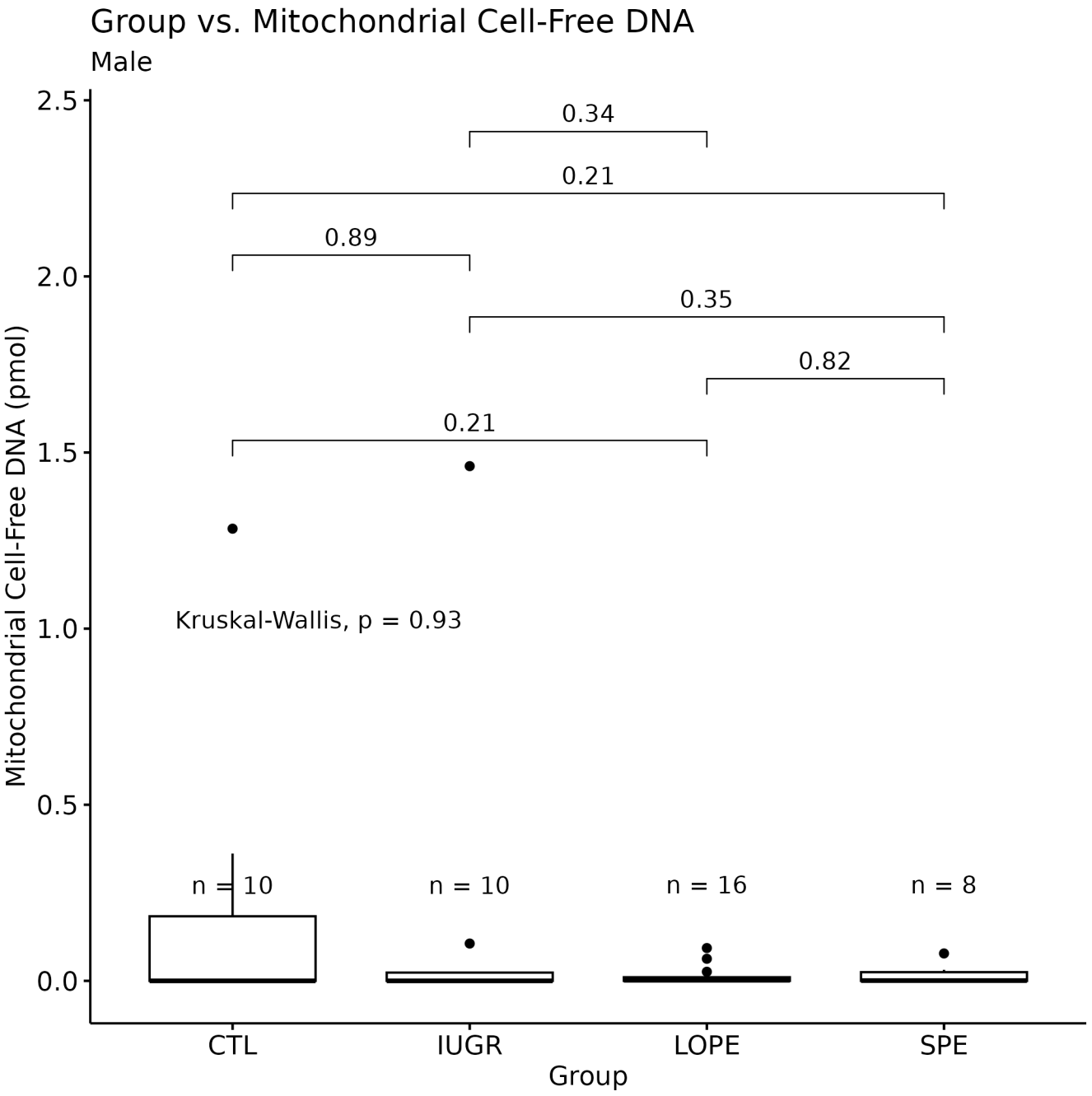

Figure 2: Differences in total cell-free mitochondrial DNA in pregnancies with a male fetus.

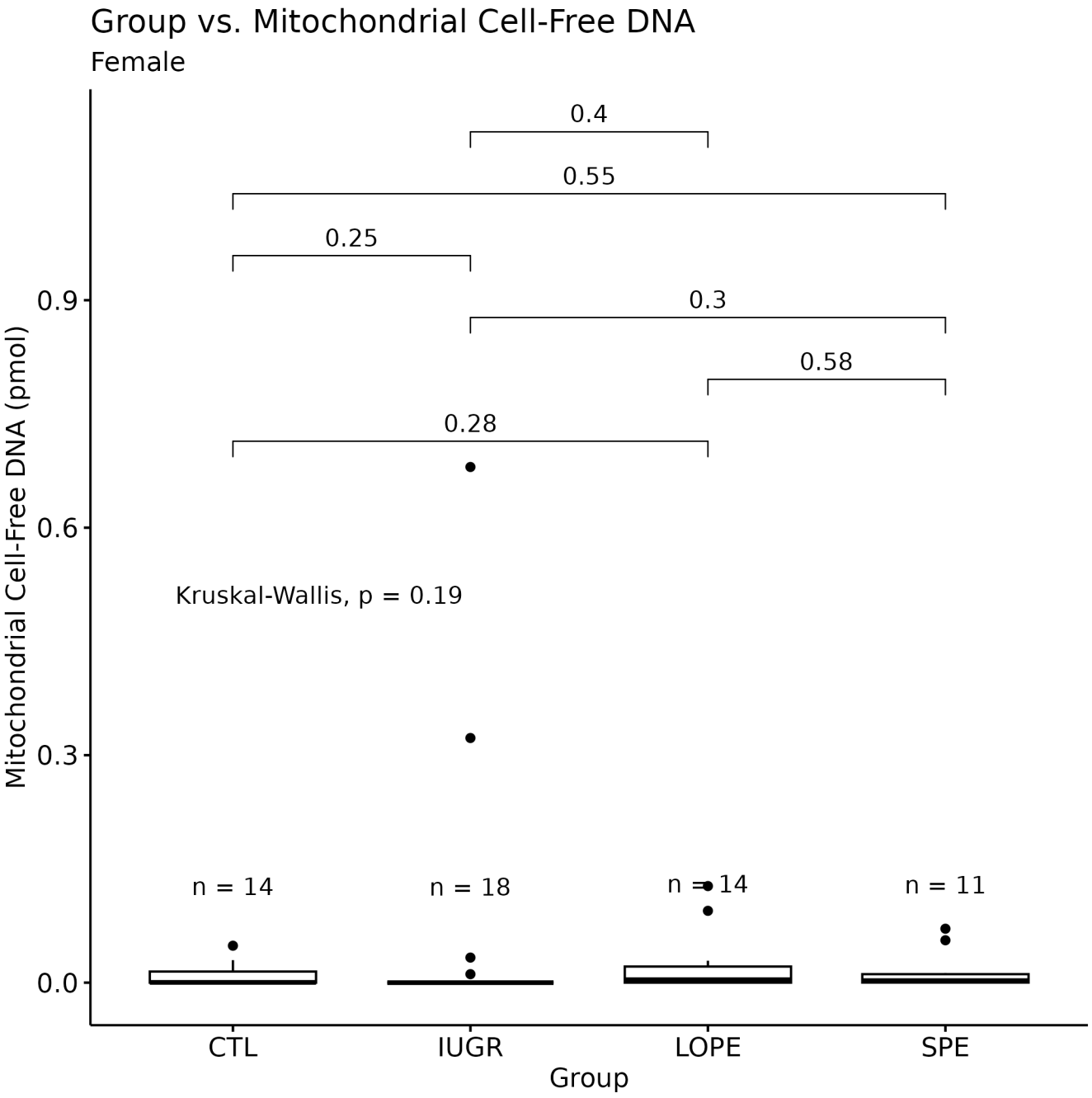

Figure 3: Differences in total cell-free mitochondrial DNA in pregnancies with a female fetus.

a larger research project where we plan to assess molecular changes associated with oxidative stress in early and late placental development using an ”Organ on a Chip” model. This will be in collaboration with Dr. Sandeep Raha and Dr. Boyang Zhang. We are currently preparing a New Frontiers in Research Fund Grant on this new project, where the Preeclampsia Foundation is a named Knowledge User.