# Effects of Glucose and Insulin Levels on Intervertebral Disc Metabolic Responses in an *in vitro* Rat Tail Model

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*The authors declare no conflicts of interest related to this study.*

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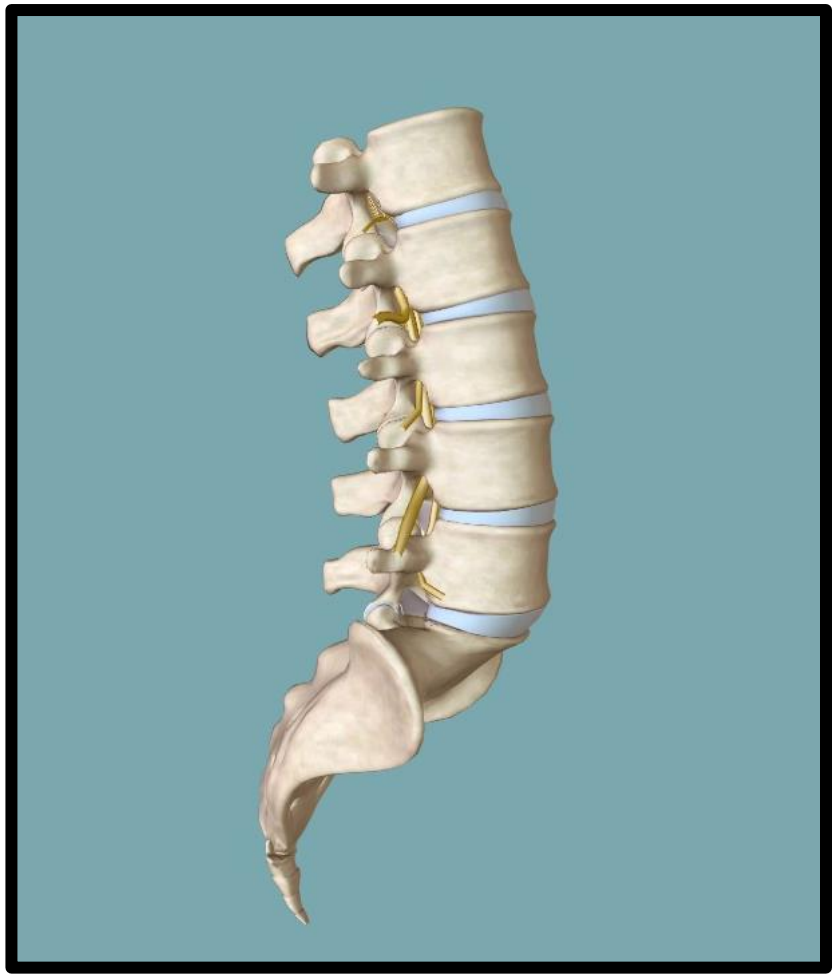
# INTRODUCTION AND PURPOSE

- Intervertebral disc (IVD) degeneration is associated with debilitating neck and low back pain, and there is evidence that there is a relationship between diabetes on IVD degeneration

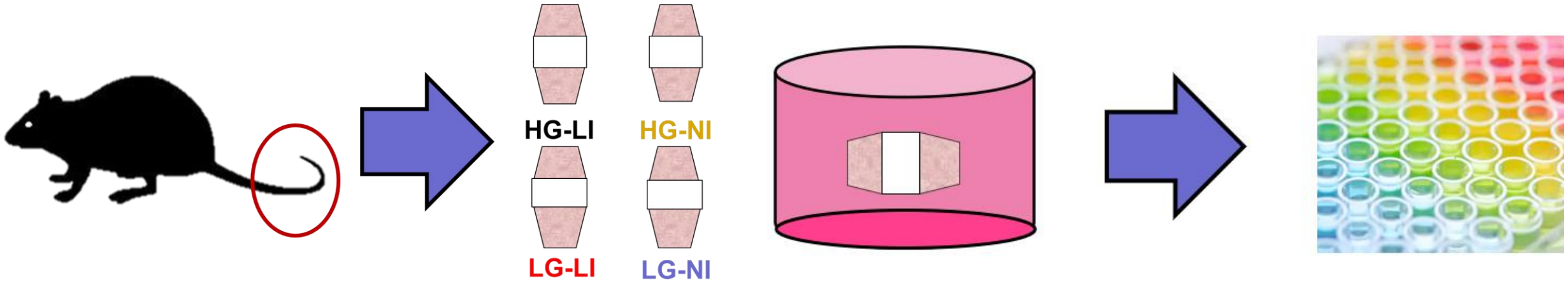
- The metabolic effects of diabetes, and varying levels of glucose and insulin, on the IVD have not been established

- This study was designed to characterize the metabolic responses of IVDs in varying concentrations of glucose and insulin using a rat tail whole organ IVD culture model



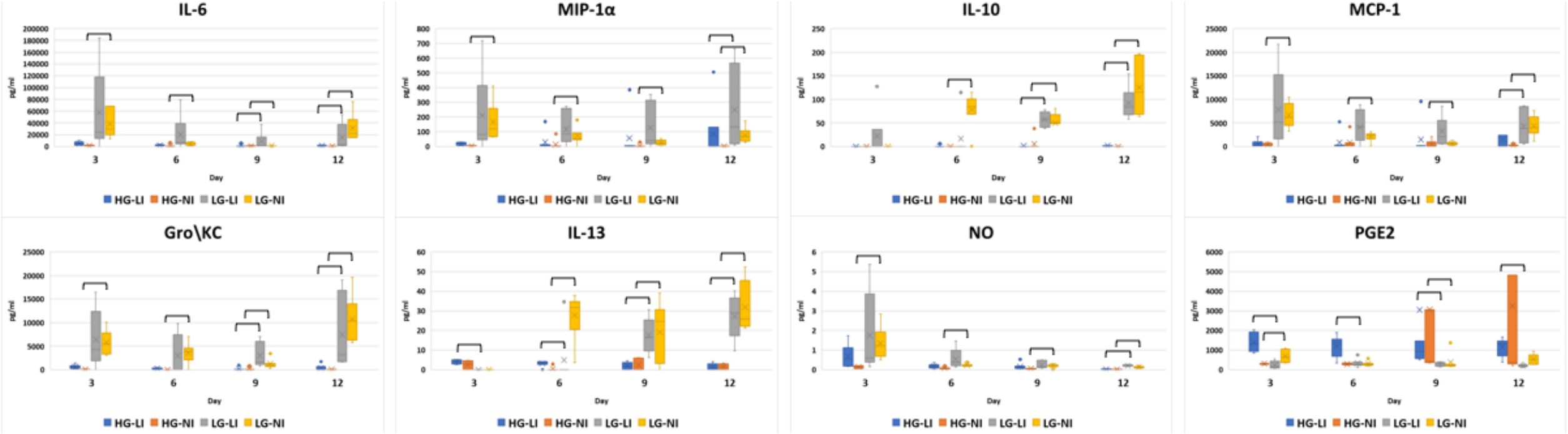
# METHODS

1. With IACUC approval (ACUC#9435), tails were collected from skeletally mature Sprague Dawley rats (n=6) euthanatized for reasons unrelated to this study.
2. IVD whole organ explants (n=24) were created and were randomly assigned to one of four culture groups based on media insulin (1 µg/ml low insulin [LI] or 10 µg/ml normal insulin [NI]) or glucose (4500 µg/ml high glucose [HG], or 1000 µg/ml low glucose [LG]) concentration.
3. Explants were cultured for 12 days with media changed and collected on day 3, 6, 9, and 12 for biomarker analysis
4. Media were analyzed for concentrations of NO, PGE2, MIP-1α, IL-6, IL-10, MCP-1, GRO-KC, and IL-13.
5. A Kruskal Wallace with post-hoc analysis and Bonferroni correction were used to determine significant differences between groups at each time point with significance set at $p<0.05$.



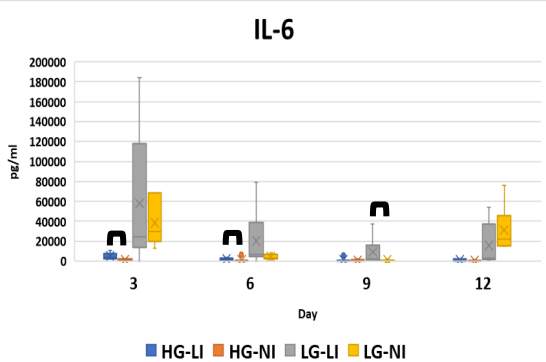
# RESULTS – Effects of Glucose Levels

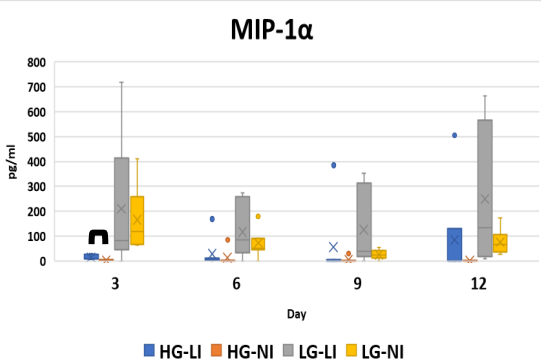
- The HG-LI group produced significantly higher levels of IL-13 (days 3-6), PGE2 (days 3-12), and IL-6 (days 9-12), and significantly lower levels of IL-13 (days 9-12), MIP-1α (day 12), IL-10 (days 9-12), GRO/KC (days 9-12), and NO (Day 12), compared to the LG-LI during culture.
- The HG-NI group produced significantly higher levels of PGE2 (day 9), and significantly lower levels of IL-13 (days 6-12), PGE2 (day 3), MIP-1a (day 3-12), IL-6 (days 3-12), IL-10 (days 6-12), MCP-1 (day 3-12), GRO/KC (days 3-12) and NO (days 3-12) compared to the LG-NI group during culture.

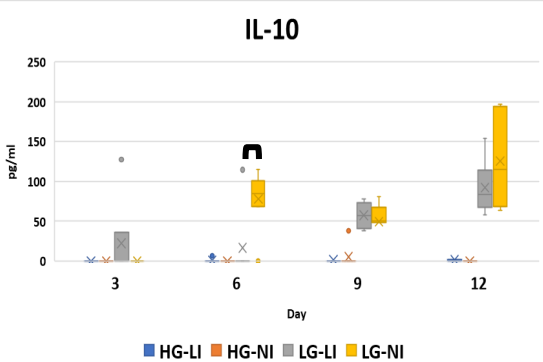


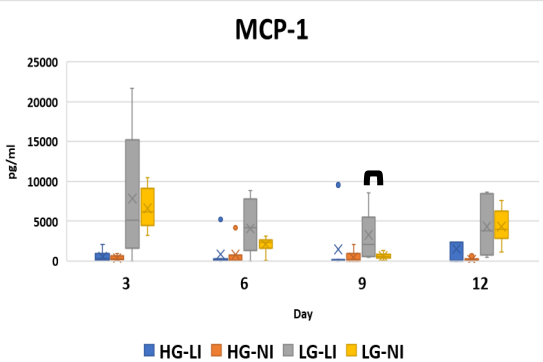
# RESULTS – Effects of Insulin Levels

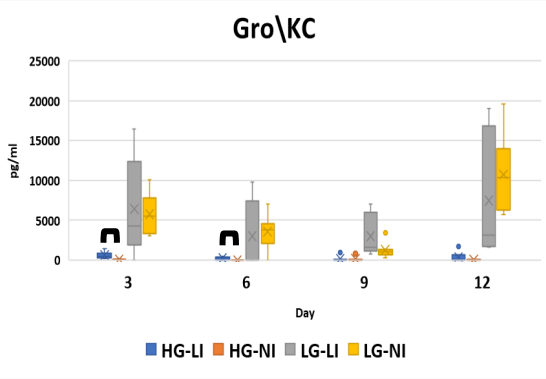
- The HG-LI group produced significantly higher levels of IL-13 (Days 3-6), PGE2 (days 3-6), MIP-1a (day 3), IL-6 (days 3-6), GRO/KC (days 3-6), and significantly lower levels of NO (days 3-6), compared to the HG-NI group during culture.
- The LG-LI group produced significantly higher levels of IL-6 (day 9), MCP-1 (day 9), and NO (day 12), and significantly lower levels of IL-13 (day 6), PGE2 (day 12), and IL-10 (day 6), compared to the LG-NI group during culture.

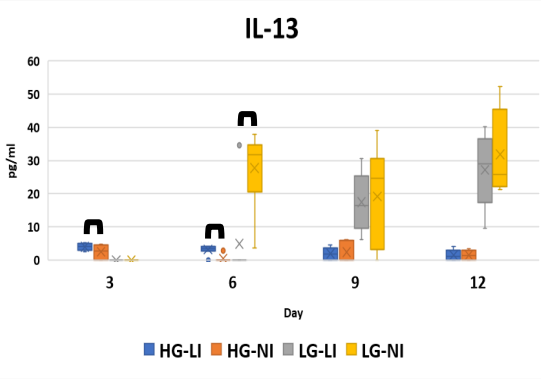


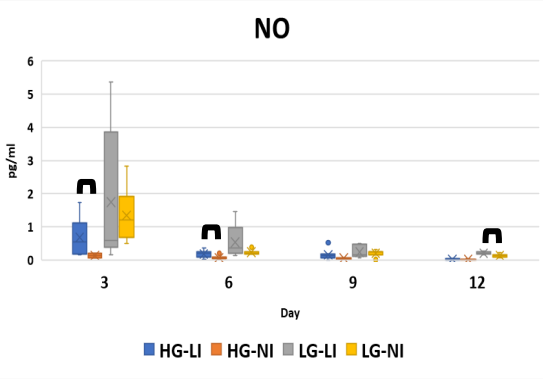



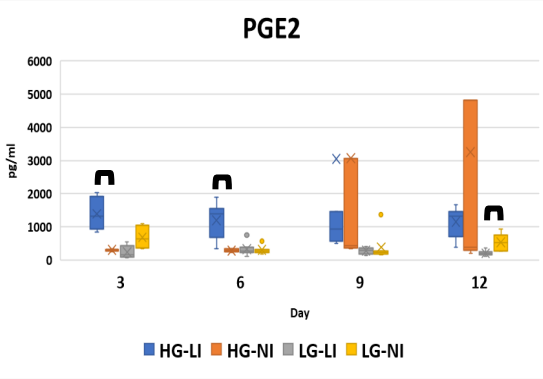



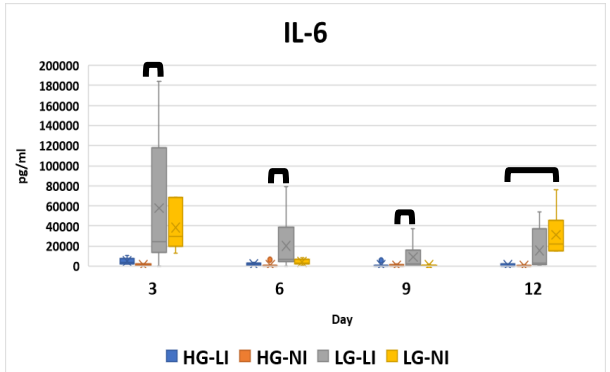



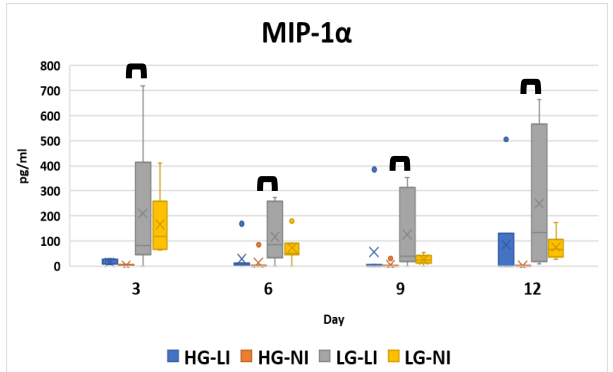



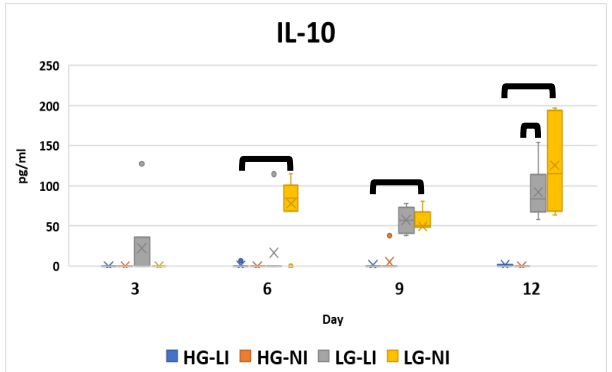



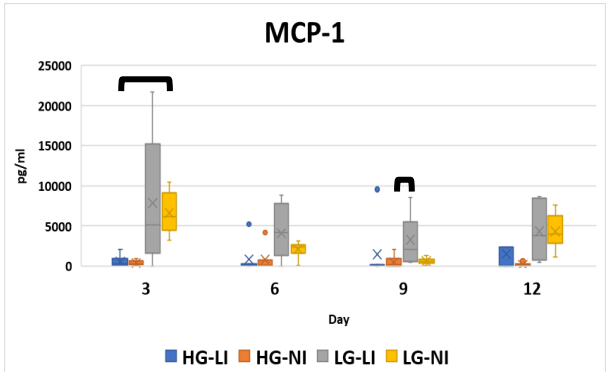
# RESULTS – Effects of Glucose and Insulin Levels

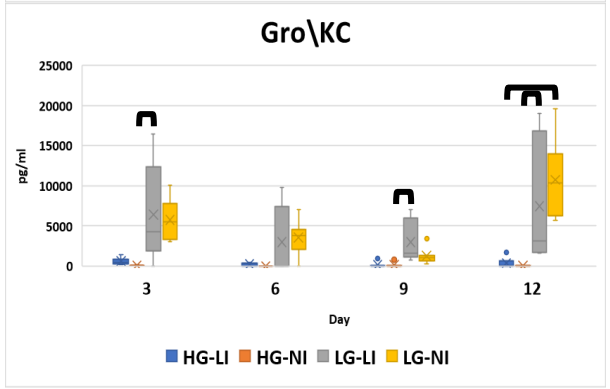
- The LG-LI group had significantly higher production of MIP-1a (days 3-12), IL-13 (days 9 and 12), IL-6 (days 3-9), IL-10 (day 12), GROKC (days 3, 9, 12), MCP-1 (day 9), and NO (days 6 and 12) compared to the HG-NI group.
- The LG-NI group had significantly higher production of MCP-1 (day 3), IL-10 (days 6-12), IL-13 (days 9 and 12), IL-6 (day 12), and GRO\KC (day 12), and significantly lower production of IL-13 (day 3) and PGE2 (days 6 and 9), compared to the HG-LI group.

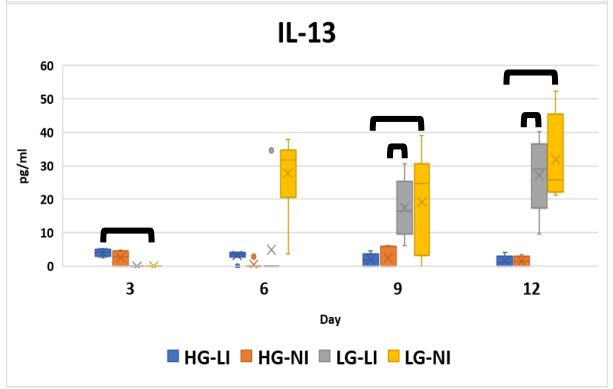


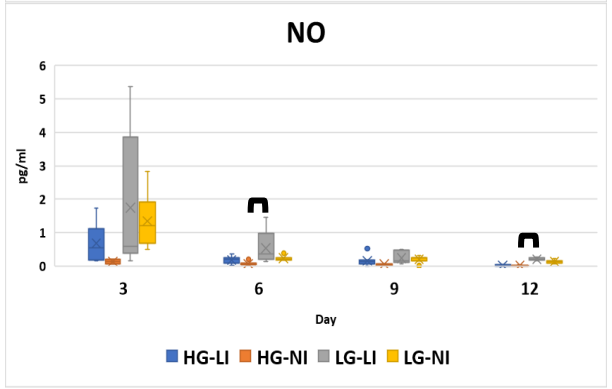



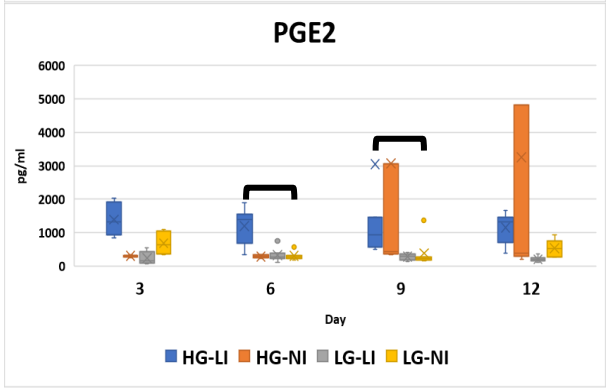



# Conclusions

- **The data from this study indicate that inflammatory metabolic responses of IVDs are sensitive to changes in glucose and insulin levels**
- **A low-glucose environment appears to be more pro-inflammatory than a high-glucose environment, and low insulin conditions appears to exacerbate the effects of glucose concentration**
- **These results suggest that insulin and glucose levels may play direct roles in exacerbating the inflammation-mediated effect on IVDs in diabetic patients**

# Questions?

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