Adverse Effects of Glucocorticoids are Exacerbated in the Presence of Obesity

# Obesity is a rapidly growing epidemic in the US and is associated with an array of metabolic co-morbidities, such as insulin resistance and liver steatosis. Chronically elevated circulating glucocorticoids, which can occur due to Cushing’s Syndrome or from taking commonly prescribed steroid hormones chronically, lead to increased fat mass and similar co-morbidities. With this in mind, we hypothesized that having elevated glucocorticoids in the presence of obesity would further exacerbate these metabolic complications.

# Dexamethasone-Induced Insulin Resistance is Worsened in the Presence of HFD-Induced Obesity

* Show ITTs from NCD and HFD dex mice
* Fasting glucose---find serum samples for this and insulin—HOMA IR
* Human data stratified by BMI and HOMA-IR
* CLAMPS, tissues from CLAMPS—better to look at the signaling it these
  + Insulin signaling (pTyr-IR/IRS and pAKT) differences-mRNA and protein in mouse adipose tissue, \*muscle and liver
  + If muscle ins sig is reduced then in vitro muscle info (direct effect)—lots of citations needed (invitro and in vivo)
  + Biological effect in muscle?
    - Dex or something else from HFD
  + Not

# HFD-Induced Liver Steatosis is Worsened in Dexamethasone Treated mice

* Liver TGs
* Lipid staining and H&E staining of livers
* Check AST/ALT levels in serum
* Human data here too
* Check liver TG synthesis enzymes (same logic as Erin’s paper, maybe its steatotic secondary to FFA flux)---via qPCR or other analysis?

# Dexamethasone Causes Decreased Fat Mass and IN HFD-Fed Mice

* Body composition (fat mass and percent)
* Food consumption
* RER???-in progress and CLAMS from 70d if needed
* Fat pad weights
* Fat pad cell size/staining
  + Lean mass here instead maybe, come back to this with atrophy

# Dexamethasone Treatment Results in Increased Lipolysis (70 day dex mice)

* Cell culture
  + Continuous dex treatment leads to decreased lipid accumulation and greater glycerol release-Shown, but need to get up to N=3-check if got for this and below
  + Increased ATGL/HSL (*Lipe*) mRNA in vitro with Dexamethasone- Shown, but need to get up to N=3
    - qPCR of bar and pde3b
  + Protein
  + Identification of GRE at *Pnpla2* (sequence + EMSA)
  + CHiP of *Pnpla2* with GR
* Mouse data
  + Increased basal lipolysis, and isoproterenol and fasted (chow)
    - Glycerol
    - TG (maybe not include)
    - FFA---make sure none of this is already in paper we published!

qPCR of Pnpla2 (at least) from these lysates and western blotting

# Dexamethasone-Induced Lipolysis is increased in HFD-Fed Mice

* FFA/Glycerol from HFD/NCD Water/Dex from Fasted
* Mouse iWAT qPCR data - increased lipolytic transcripts
* Perform westerns on lipolysis proteins
* More GR occupancy on *Pnpla2*  with HFD

# Dex/HFD Promotes Muscle Breakdown (may be its own paper)

* Immediately after going on Dex increased RER/VO2- have shown, but still need to do in HFD/older chow cohort
* Lean Mass
* Energy expenditure/RER at baseline and at end of 70 day cohort (more VO2 after adjusting for lean mass)- have shown, but still need to do in HFD/older chow cohort?---we have performed clams at initial dex treatement in the HFD and NCD cohorts (not analyzed)
* Lean Mass
* Grip
* Muscle weights
* Measure muscle FOXO and atrogene mRNA/protein-have shown atrogenes upregulated in short term dex muscles
* Muscle sections—higher atrophy

Elevated Glucocorticoids/

Dexamethasone

**Working Hypothesis**

HFD-induced Obesity

Atrogenes?

Insulin Sensitivity

Dex alone (directly?)

Lipolytic mRNA transcripts (directly?)

Liver Steatosis?

Lean Mass

Strength

Fat Mass

Lipolysis