Sex-Specific Effects of Dietary Genistein on Survival, Growth, and Gene Expression in ΔF508 Cystic Fibrosis Mice

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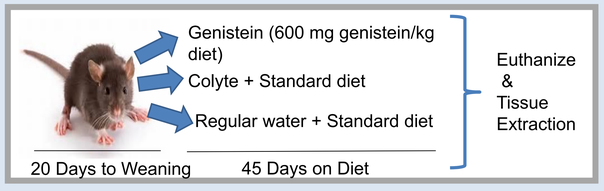
INTRODUCTION

- The ΔF508 mutation is the most common cause of cystic fibrosis (CF), leading to severe intestinal disease in mice and resembling meconium ileus in human patients. This mouse mutation requires laxatives for survival (1).
- ΔF508 CF mice were fed one of three diets (regular, regular + laxative Colyte, or genistein-enriched) for 45 days post-weaning to assess survival and physiological outcomes.
- Key Findings on survival and weight show Genistein improved survival, especially in females (87% vs. 71% with Colyte), and increased body weight in males by 16% compared to Colyte-fed mice (2).
- Genistein is a known activator of the CFTR chloride channel, including the ΔF508 mutation (3).
- Genistein exerts sex-specific effects on survival, metabolism, and gene regulation in CF mice, emphasizing the need to investigate its molecular mechanisms and potential therapeutic applications.

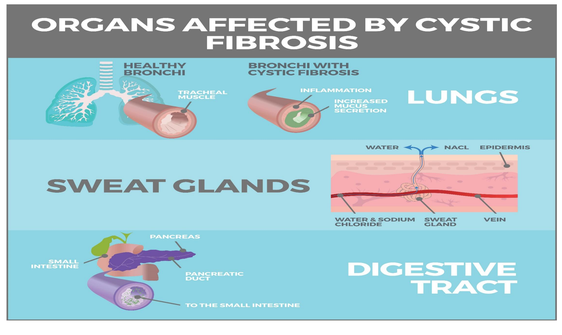
METHODS

- Number of ΔF508-CF mice for the study = 78 M & 72 F



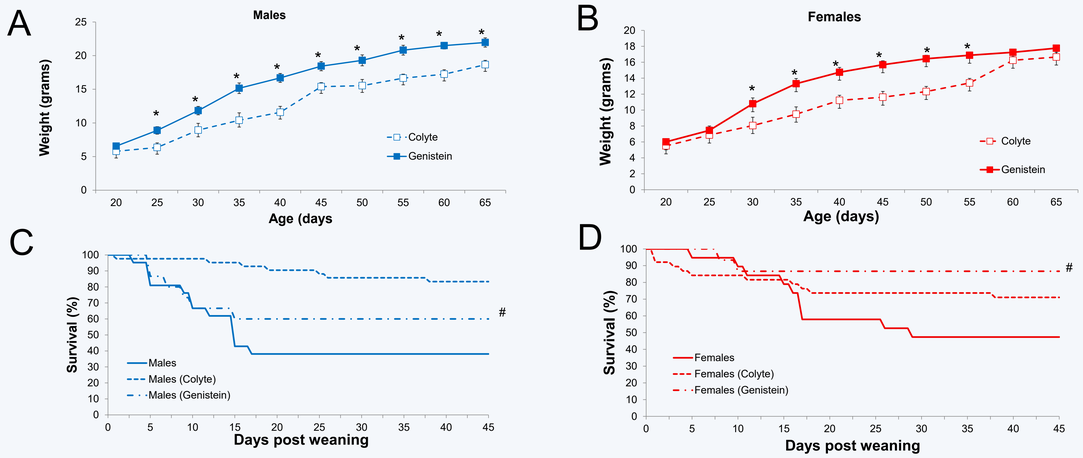
Molecular Analysis:

- RNA extraction: Performed from flash-frozen liver and small intestine using Qiagen RNeasy microkits (Qiagen, Valencia, CA).
- cDNA synthesis: Generated from 1 µg RNA using iScript cDNA synthesis kits (Bio-Rad, Hercules, CA).
- qPCR analysis: Conducted using 0.5 µg cDNA, run in triplicate to quantify target gene expression.



RESULTS

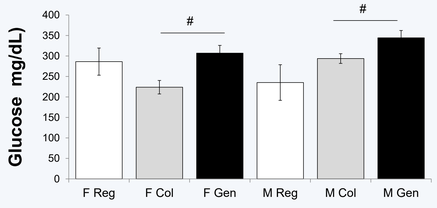
Figure 1: Effects of Genistein on Weight and Survival



Effect of genistein diet on weight of the DF-508 CF mouse. **A. Male Weight:** Genistein diet significantly increased weight versus colyte throughout the entire study (colyte n=8 and genistein n=14**).** **B. Female Weight:** Genistein-fed group gained weight earlier than those fed colyte, but end weight was the same for both groups (colyte n=9 and genistein n=15).). **C. Male Survival:** Males fed genistein diet survived less well, 60%, than those fed colyte, 83.3% (colyte n=42 and genistein n=15**).** **D. Female Survival:** **G**enistein diet improved survival rates to 86.7%, compared to those fed colyte, 71.1% (colyte n=38 and genistein n=15). Data are mean±SEM. # denotes significance, *P*<0.05.

Absence of CFTR in the intestine does not correlate with the observed growth reduction in CF (4)

Figure 2: Effects of Genistein on serum glucose



Effect of genistein serum glucose in DF-508 CF mouse. Genistein diet increased serum glucose levels in both males and females compared to colyte. n = 6-10/group. Data are mean±SEM. # denotes significance, *P*<0.05.

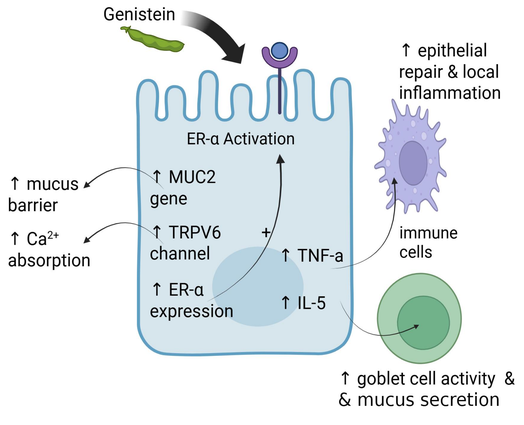
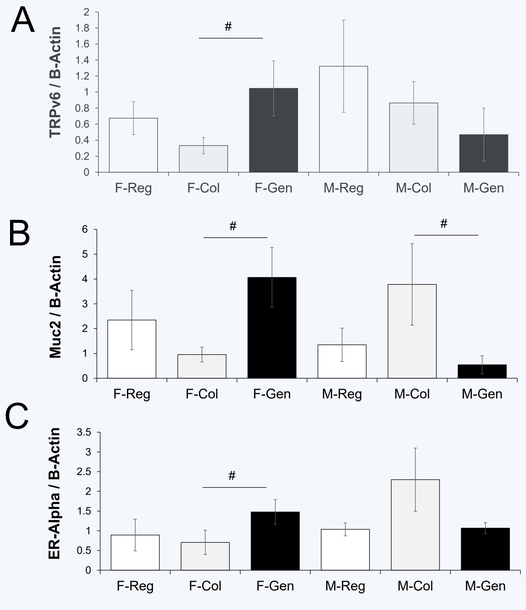


Figure 3: Effect of Genistein on Gene Expression of TRP6v6. Muc2 & ER-Alpha



Effect of genistein intestinal genes. **A. TRPpv6:** Genistein diet increased TRPv6 versus colyte in females**. B. Muc2:** Genistein increased Muc2 expression in females but down-regulated it in males. **C. ER-Alpha: G**enistein increased ER-alpha expression in females. n=3-6/group.Data are mean±SEM. # denotes significance, *P*<0.05.

Figure 4: Effect of Genistein on intestinal inflammatory markers

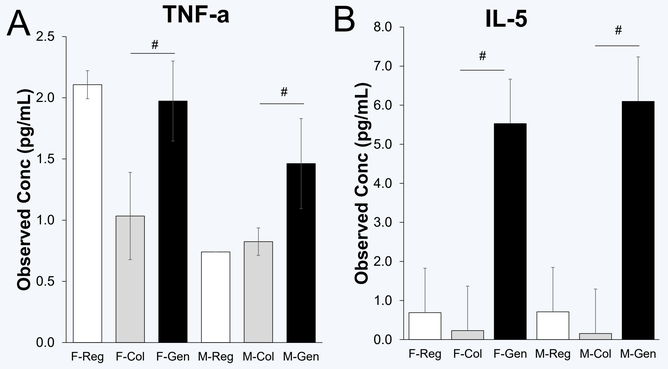


Figure 4. A. Intestinal TNF-alpha. This pro-inflammatory cytokine was increased with genistein versus colyte (both males and females). **B. Intestinal IL-5.** This cytokine was increased with **g**enistein (both males and females). n=3-5/group. Data are mean±SEM. # significance, *P*<0.05.

CONCLUSIONS

- Genistein (Gen) treatment improved survival rates in female mice compared to standard Colyte care.
- In males, Gen administration led to an overall increase in body weight.
- Female mice on Gen diet showed a faster early weight gain, but their final body weight was similar to males.
- Serum glucose levels were elevated in both male and female Gen-treated mice.
- Gene expression analysis showed:
  - Upregulation of TRPv6, MUC2, and ERA in females.
  - Downregulation of MUC2 in males.
- Serum TNF-alpha and IL-5 were increased with genistein diet.

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