Cut the Crap: Mechanical Bowel Preparation Does Not Improve Engraftment of Fecal Microbiota Transplant

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Introduction

- The gut microbiome plays a crucial role in modulating local and systemic inflammation.[1]
- By influencing the balance and composition of the gut microbial community, fecal microbiota transplantation (FMT) has been established as an effective treatment for recurrent and refractory Clostridioides difficile colitis and has shown therapeutic potential in other gastrointestinal, metabolic, allergic, rheumatic, and neuroinflammatory pathologies that involve intestinal dysbiosis.[2]
- If the therapeutic effect of FMT relies on the replacement of pro-inflammatory microorganisms with those that are anti-inflammatory, the ability to engraft the donor microbiome within the recipient bowel is paramount to the success of the treatment and the development of animal models.

Hypothesis

We hypothesized that mechanical bowel preparation before FMT would improve colonization of donor microbiota and result in a recipient mouse microbiome that more closely resembles that of the human donor.

Methods

Mice either underwent four oral-gastric gavages administered at 20-minute intervals with 200 µL of polyethylene glycol (PEG) at 425 g/L or phosphate-buffered saline (sham). Three mice from each group were euthanized and their colons were harvested to verify PEG efficacy. Each cohort then received FMT with 200 µL of stool resuspended in PBS (100 g/L) from human patients with an active inflammatory bowel disease (IBD) flare, healthy human donors (HD), or PBS control.

Stool was collected at baseline prior to bowel pretreatment. Mice underwent three rounds of FMT: four hours after bowel pretreatment, on day 2, and on day 4. Stool was collected again on day 7.

Figure 1. Experimental Groups

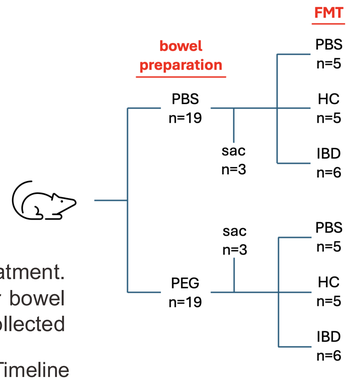
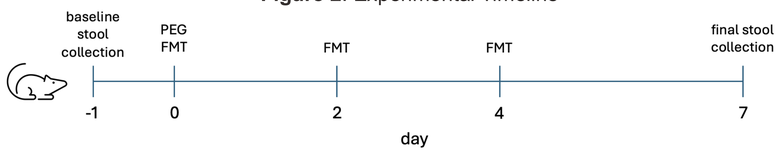


Figure 2. Experimental Timeline



Genomic DNA was PCR amplified with primers sIDTP5_515F and sIDTP7_806R targeting the V4 variable region of microbial small unit ribosomal RNA genes. PCR products underwent 16s rRNA sequencing on an Illumina MiSeq i100+.

Results

- The gut microbial structure of recipient mice at the genus level much more closely resembled the recipients' baseline microbiota than that of the transplanted human stool 7 days post-FMT.
- The genus Bacteroides, absent from the native murine microbiome, was engrafted into all mice that received FMT from a human donor.

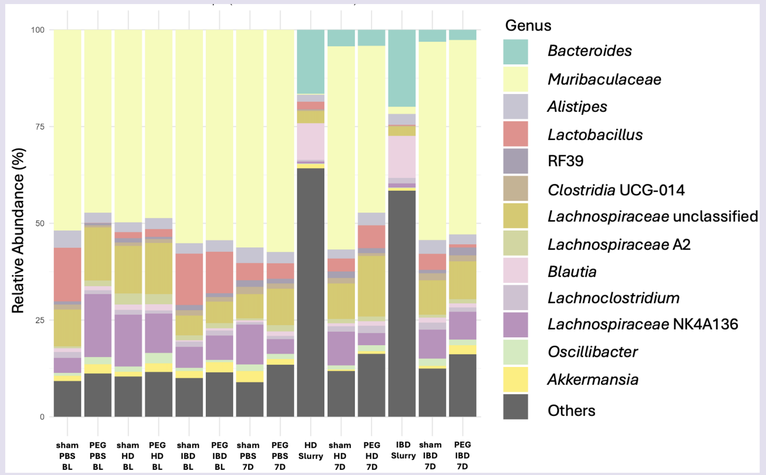


Figure 3. Relative baseline and post-engraftment abundances of gut microbial taxa at the genus level for each experimental group and the donor slurries.

- Post-engraftment microbiota of mice did not differ significantly between those that were pretreated with PEG or PBS, regardless of whether they received FMT with IBD or HD stool

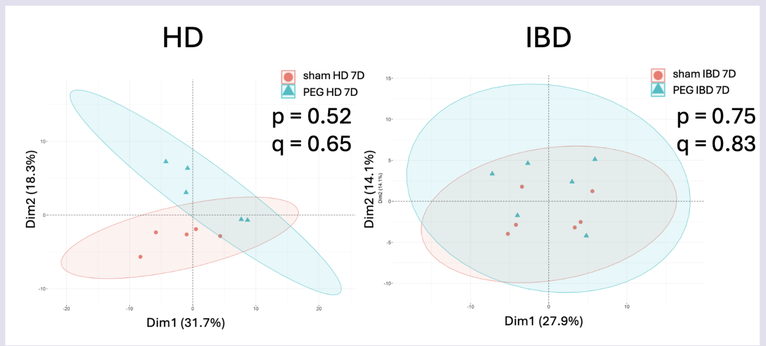


Figure 4. Principal Component Analysis (PCA) plots comparing post-engraftment microbiota in mice that received PEG or PBS pretreatment.

Results

- Mice that received FMT with IBD stool demonstrated a more significantly reshaped microbiome 7 days post-FMT compared to baseline than mice that received FMT with HD stool.

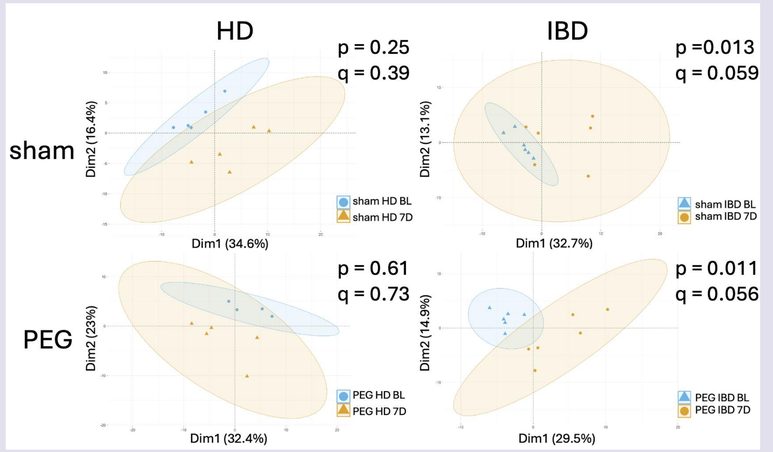


Figure 5. Principal Component Analysis (PCA) plots comparing baseline and post-engraftment microbiota at the genus taxonomic level.

Conclusions

- **Mechanical bowel preparation prior to FMT did not improve engraftment of human microbiota onto mice.**
- **The therapeutic effect of FMT demonstrated in other studies may be due to subtle but significant alterations in specific anti-inflammatory taxa rather than broad changes to the microbiome.**
- **Inflammatory proteins or structural components of stool related to dysbiosis may play a role in mucosal colonization.**

References

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2. Karimi, M. et al. Safety and efficacy of fecal microbiota transplantation (FMT) as a modern adjuvant therapy in various diseases and disorders: a comprehensive literature review. Front Immunol 15, 1439176 (2024). https://doi.org/10.3389/fimmu.2024.1439176

Acknowledgements

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