Rational Donor Fecal Microbiota Transplantation in HIV (REFRESH Study)

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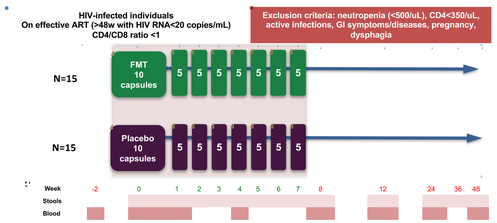
ABSTRACT #284

INTRODUCTION

- Chronic inflammation persists in people with HIV (PWH) persists despite ART and drives an excess risk of adverse events and mortality.
- Because abnormalities in the microbiota have been linked with persistent inflammation and immune recovery in PWH, manipulation of the gut bacterial communities has been pursued as a strategy to improve clinical outcomes. However, the gut microbiota has remained so far an elusive target for interventions.
- It is unknown whether oral fecal microbiota transplants (FMT) can affect the gut microbiota and systemic immunity of HIV-infected individuals.

METHODS

Study Design and Outcomes



clinicaltrials.gov: NCT0300894

- **Outcomes:**
  - Primary: 48-week safety
  - Secondary : 48-week efficacy (changes in CD4+/CD8+ T cells)
  - Exploratory:
    - Fecal microbiota changes and donor's microbiota engraftment
      (Illumina platform, Q/C, trimming using trimmomatic, OTU picking using Kraken
      Taxonomic assignment using SILVA database, rarefaction and subsampling
    - Plasma biomarkers of inflammation and bacterial translocation: commercial ELISA kits run in triplicate, in batch

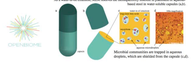
Study procedures, data (collected in REDCap) and safety were monitored by a *Fundación SEIMC-GESIDA*.

P values computed using piecewise linear mixed models to compare trajectories (time by group interaction).

FMT Preparation



- Three stool donors were selected from a universal donor stool bank based on fecal abundance of:
  - *Fecalibacterium prausnitzii*
  - *Bacteroides spp.*
  - Butyrate
  - Prevotella spp.
- Openbiome's capsules: 0.7 g of stools each, microbial emulsion matrix technology.
- Each participant was assigned to one single donor. The capsule intake was supervised by the research nurse.



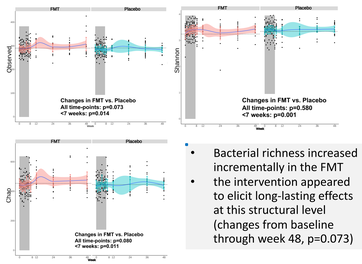
RESULTS

Description of the Study Population

	FMT n=14	Placebo n=15
Age, mean (SD), years	48 (10)	46 (12)
Male, n (%)	12 (86)	13 (87)
Body mass index, median (P25-P75), kg/m2	24 (20-26)	25 (23-27)
Years since HIV diagnosis, median (P25-P75)	19 (7-26)	5 (3-610)
Risk factor, n (%)		
MSM	9 (64)	11 (73)
HTX	0 (0)	3 (20)
IDU	5 (36)	1 (7)
AIDS diagnosis, n (%)	4 (29)	3 (20)
HIV RNA <20 copies/mL	15 (100%)	15 (100%)
Nadir CD4, median (P25-P75), cells/uL	110 (56-187)	259 (160-300)
CD4+ T cell counts, median (P25-P75), cells/uL	703 (457-852)	545 (454-684)
CD4/CD8 ratio, median (P25-P75), cells/uL	0.58 (0.45-0.78)	0.73 (0.39-0.96)
Use of antibiotic in past 6 months, n (%)	3 (21)	2 (13)
On triple ART, n (%)	15 (100)	15 (100)

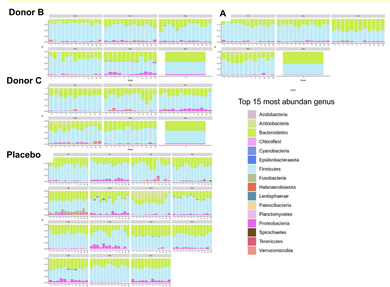
Between January and May 2017, 47 participants were screened to participate in the study; 17 were not eligible and the remaining 30 were randomized into treatment or placebo groups. A total of 29 subjects completed the 48-week follow-up

Changes in alpha diversity



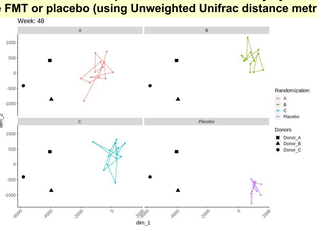
- Bacterial richness increased incrementally in the FMT
- the intervention appeared to elicit long-lasting effects at this structural level (changes from baseline through week 48, p=0.073)

Relative proportion of taxa at the genus level



Beta Diversity

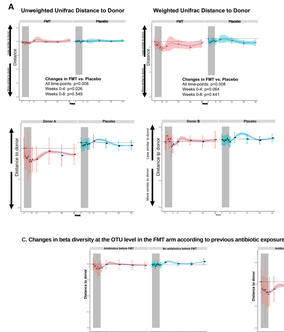
Principal Component Analysis (PCA) representing donor microbiota profiles and HIV-infected recipient microbiota community dynamics after the FMT or placebo (using Unweighted Unifrac distance metric)



After FMT, recipient microbiota profiles remain distinct from the donors, but more pronounced changes are observed in the FMT group.

Beta Diversity

Engraftment of donor's microbiota on study participants. Unifrac distances from recipients to donor)



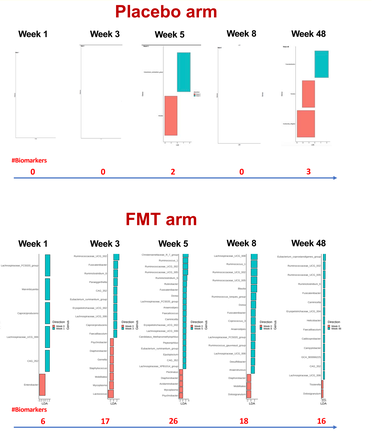
- For unweighted Unifrac, the peak effect occurred at week 3 and maintained a *plateau* until week 8.
- For weighted Unifrac, the effect was incremental after each FMT, and there was no evidence of a plateau.
- The intervention affected the dynamics of the microbiota at the beta diversity level, yet the effect at this level tended to disappear once the FMT intervention was discontinued.

The effect on weighted Unifrac distances was again more pronounced for donor A

C: Changes in beta diversity at the OTU level in the FMT arm according to previous antibiotic exposure

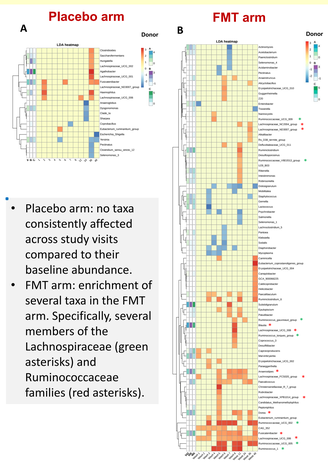
The engraftment was more apparent in the four subjects with recent antibiotic exposure

LEfSe of LDA distances from baseline to different timepoints in each group

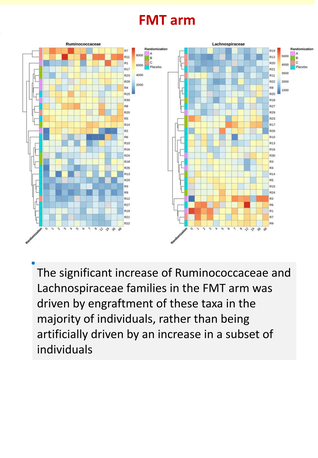


The number of taxa driving differences in microbiota composition across key time-points (weeks 1, 3, 5, 8, and 48) drastically differed in the placebo arm (range, 0–3) compared to the FMT arm, in which six bacterial biomarkers were identified at week 1, 16 at week 3, 26 at week 5, and 16 taxa were still found to drive differences at week 48

Heatmap of LDA distances at the genus level from recipients to donors at each time point

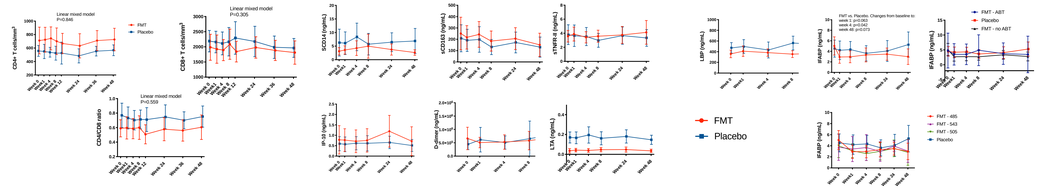


- Placebo arm: no taxa consistently affected across study visits compared to their baseline abundance.
- FMT arm: enrichment of several taxa in the FMT arm. Specifically, several members of the Lachnospiraceae (green asterisks) and Ruminococcaceae families (red asterisks).



The significant increase of Ruminococcaceae and Lachnospiraceae families in the FMT arm was driven by engraftment of these taxa in the majority of individuals, rather than being artificially driven by an increase in a subset of individuals

Changes in CD4+ T cells, CD8+ T cells and plasma biomarkers – only significant decreases of IFABP concentrations in the FMT arm



CONCLUSIONS

- Repeated oral capsular FMT from rationally selected donors was safe in PWH, attenuated HIV-associated dysbiosis, and ameliorated a marker of intestinal damage that independently predicts mortality.
- Gut microbiota manipulation using a non-invasive and safe strategy of FMT delivery is feasible and deserves further investigation.

Safety: No serious adverse events attributable to the intervention. All participants maintained virologic suppression throughout the study. In the FMT arm, five individuals reported mild abdominal distension, flatulence, and diarrhea starting some hours after capsules intake until 48–72 hours after. By the end of the study, two patients reported improvement in their bowel movements from the baseline. These symptoms were not appreciated in the placebo arm.

ACKNOWLEDGMENTS

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