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Assessing the gut microbiota response to fecal microbiota transplantation in children

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INTRODUCTION

Exposure to certain antibiotics disrupts the composition of a patient's gut microbiome, increasing susceptibility to **recurrent Clostridium difficile infection** (CDI). **Fecal microbiota transplantation** (FMT), a treatment for CDI, consists of transferring fecal material from a healthy donor to a patient's gastrointestinal tract to **restore a healthy gut microbial diversity**. Although there are numerous microbiome studies regarding the efficacy of FMT, there is a **deficit in research regarding** children with recurrent CDI (RCDI). Therefore, to assess the bacterial composition of the gastrointestinal tract pre-FMT and observe compositional changes post-FMT, donor-patient pairs (n=9), with an average patient age of 10 years old, were established.

SAMPLING

- Nine children (age range 2-20 years) with a history of RCDI and without underlying inflammatory bowel disease (IBD) were sampled.
- Six donor stool samples were acquired from Openbiome. Remaining three were collected from patient relatives.
- Patient fecal matter samples were collected via colonoscopy.
- Collection of samples occured prior to FMT and logitudinally after FMT at 2-7 weeks, 8-13 weeks, 14-19 weeks, and 20-24 weeks.

METHODS

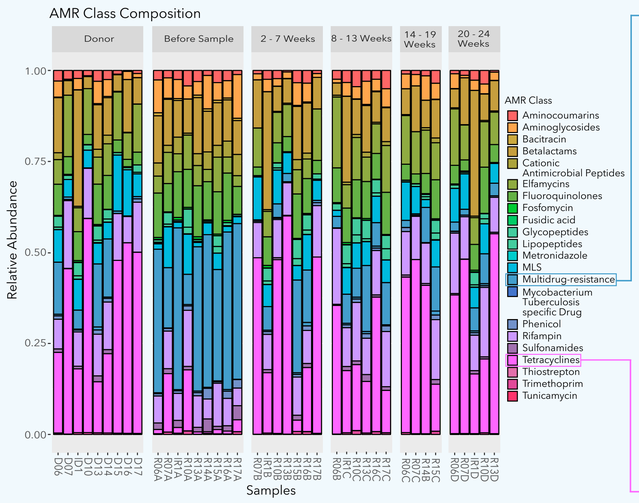
Sample Preparation

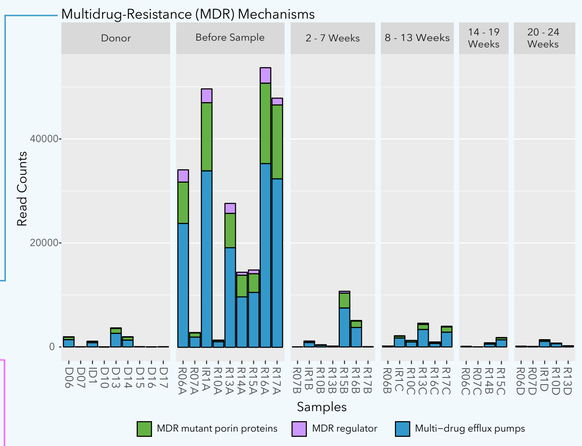
1) Extraction of bacterial DNA using QIAamp DNA Microbiome extraction kit.
2) Paired-end sequencing (single run) on a Illumina HiSeq Platform.

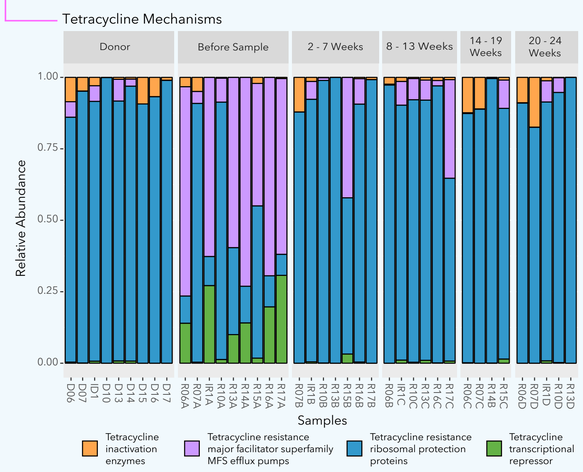
Bioinformatic Methodology

1) QA/QC of raw reads using FastQC/PrinSeq.
2) Pathoscope MAP & ID - Filtered metagenomic reads mapped to the NCBI reference genome databases (Prokaryotes, Fungi, Viruses, Protozoa) for taxonomic classification.
3) Differential abundance analysis and calculated phylogenetic alpha & beta-diversity metrics.
4) Antimicrobial resistance (AMR) determination by using AmrPlusPlus and the MEGARes database.
5) HUMAnN2 - Functional profiling to determine the presence/absence & abundance of metabolic pathways.

ANTIMICROBIAL RESISTANCE



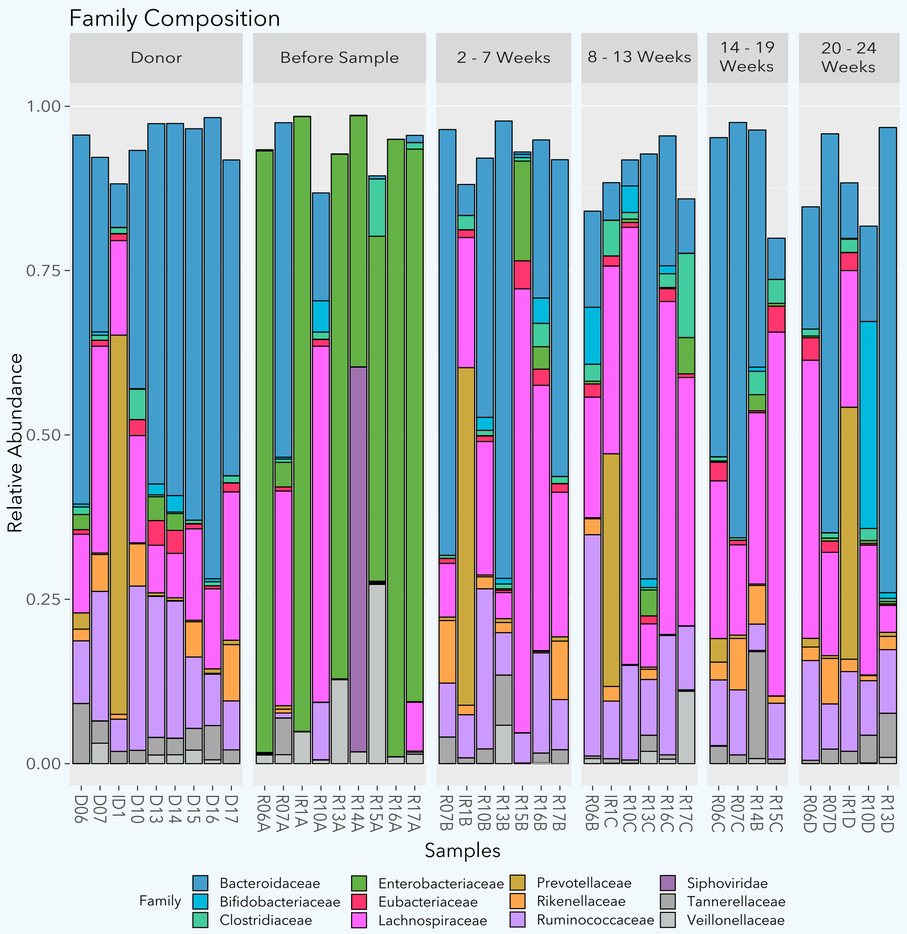




- Identified the relative abundance of antimicrobial chemical class in all samples.
- Top five chemical classes were isolated. Of those, multidrug-resistance (MDR) and tetracycline mechanisms were selected.
- High levels of multi-drug efflux pumps and tetracycline MFS efflux pumps pre-FMT and low levels post-FMT and in donors.
- Miniscule amounts of MDR mutant porin proteins post-FMT.
- Tetracycline transcriptional repressor was found at an abundance avg. of 10.76% pre-FMT compared to 0.42% post-FMT.

MICROBIOME DIVERSITY

Differential abundance analysis on donor, pre-FMT, and post-FMT samples. Microbiome composition is depicted at the taxonomic family-level. Longitudinal post-FMT samples are observed at 2-7, 8-13, 14-19, and 20-24 weeks.

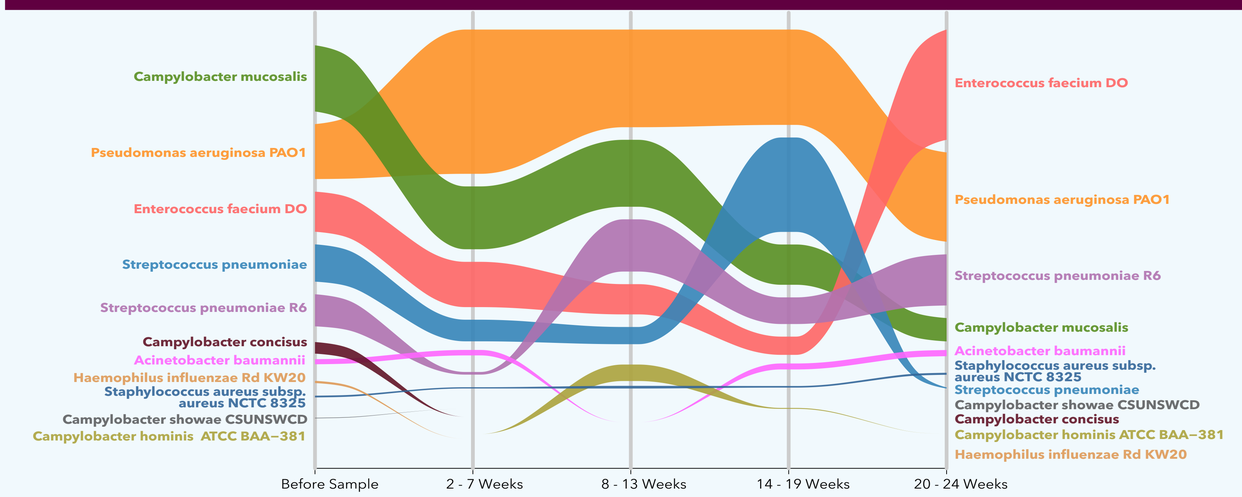


Donor samples:
- High levels of Bacteroidaceae (45.56%).
- Low levels of Enterobacteriaceae (0.949%).

Pre-FMT samples:
- Comprised of Enterobacteriaceae (59.66%).
- R07A & R10A had levels of 31.57% and 52.46% of Lachnospiraceae & 49.18% and 15.01% of Bacteroidaceae.
- R14A had high levels of Siphoviridae (58.51%).

Post-FMT samples:
- Resembled Donor samples composition.
- 67.78% abundance of Bacteroidaceae in R13.
- First sample collection of R15 had 15.09% of Enterobacteriaceae, next sample had 0.41%.
- Slight variations in IR1 for Bacteroidicaceae and Prevotellaceae.

PATHOGENICITY



- World Health Organization's priority pathogen list was utilized to isolate any pathogens found in pre- and post-FMT samples.

ACKNOWLEDGMENTS & REFERENCES

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