



# Longitudinal Analysis of Microbiome Profiles During Antibiotics Treatment and Recovery in a Canine Model

Thomas M. Kuntz<sup>1,2</sup>, Tobyn Branck<sup>4</sup>, Artemis S. Louyakis<sup>4</sup>, Jacob T. Nearing<sup>1,3</sup>, Matthew I. Jackson<sup>4</sup>, Christoph Brockel<sup>4</sup>, Dayakar V. Badri<sup>4</sup>, Kelsey N. Thompson<sup>1,3</sup>, Curtis Huttenhower<sup>1,3</sup>

<sup>1</sup>Department of Biostatistics, Harvard T.H. Chan School of Public Health, Boston, MA, United States, <sup>2</sup>Harvard Chan Microbiome in Public Health Center, Harvard T. H. Chan School of Public Health, Boston, MA, United States, <sup>3</sup>Infectious Disease and Microbiome Program, Broad Institute of MIT and Harvard, Cambridge, MA, United States, <sup>4</sup>Science and Technology Center, Hill's Pet Nutrition, Inc., Topeka, KS, United States



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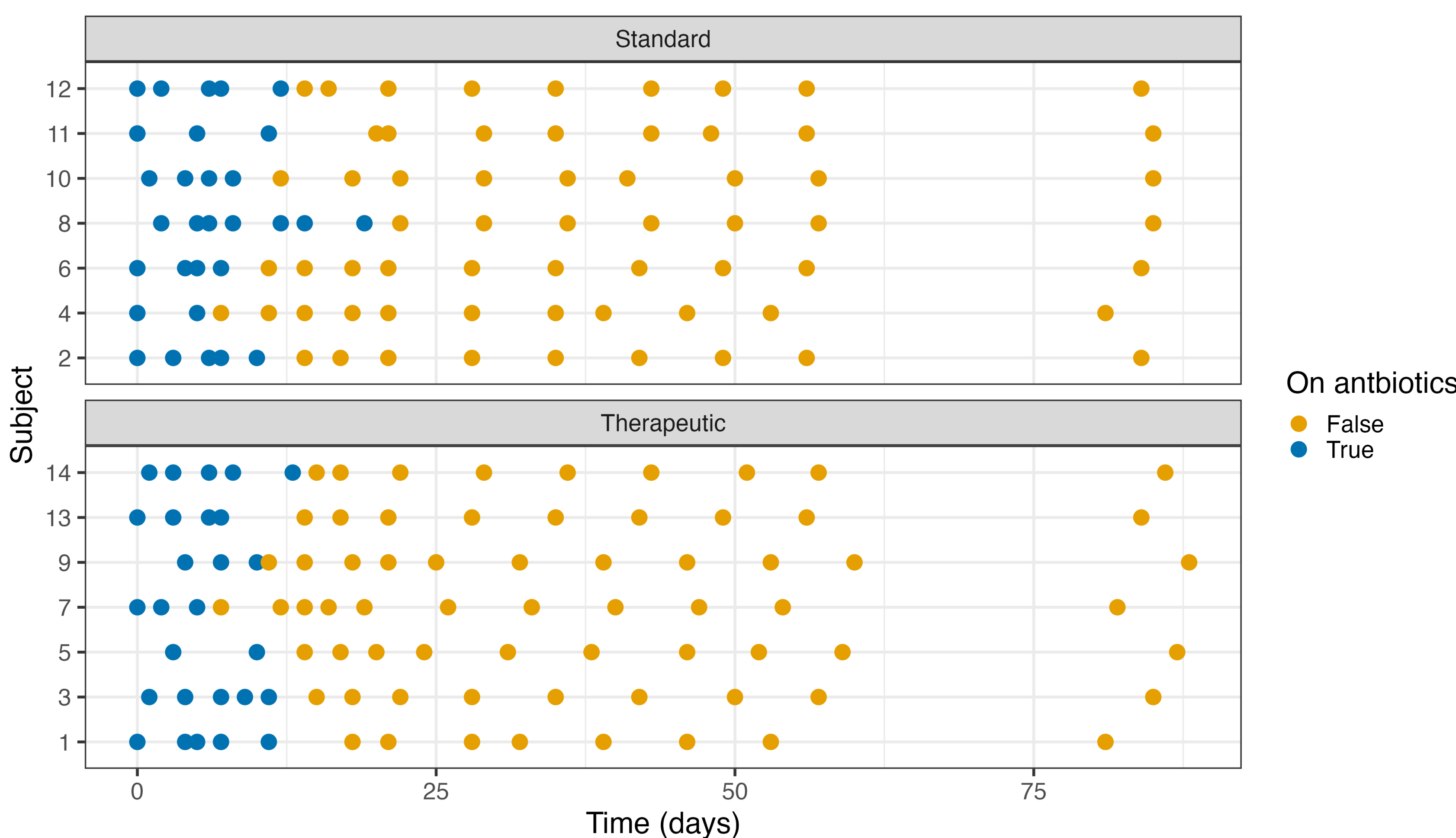
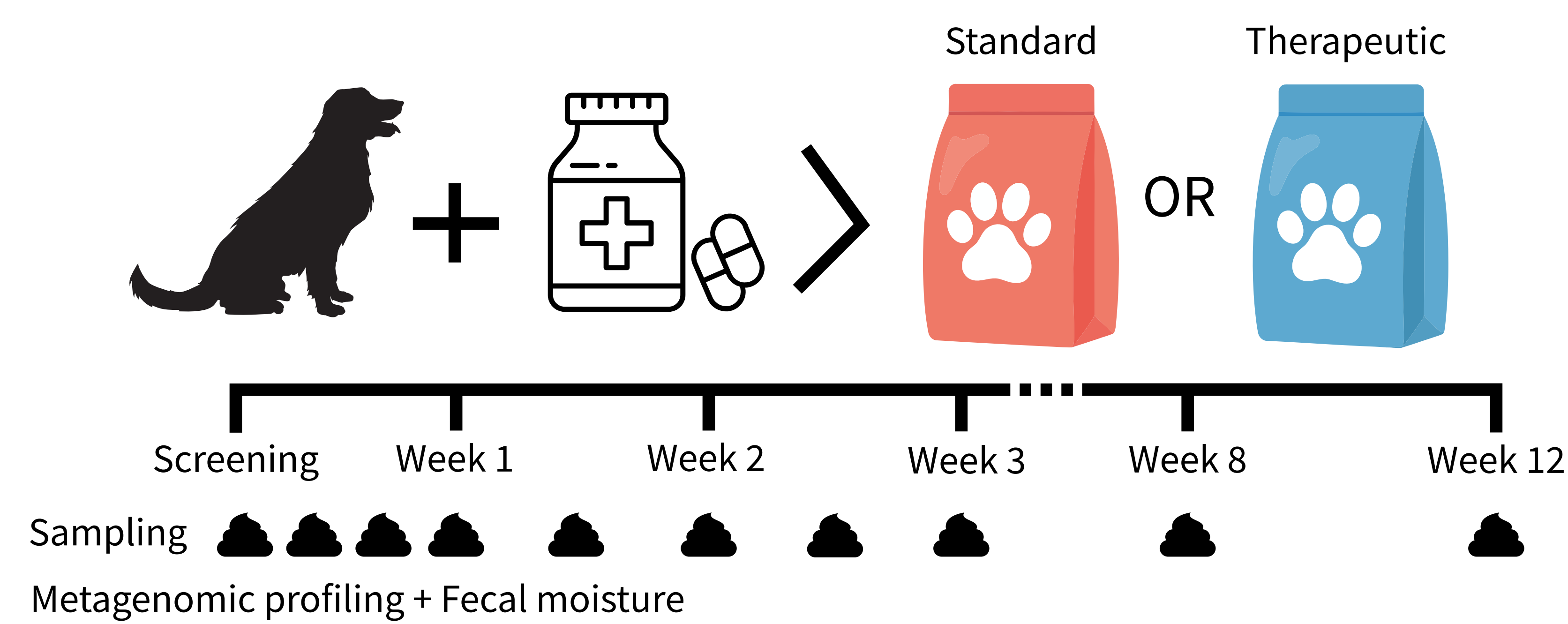


## Antibiotics treatment disrupts the microbiome

Antibiotics (ABX) are among the most prescribed medications, yet treatment often results in strong perturbations of the gut microbiome via off-target anti-microbial activity. When the host microbial community is depleted, there is an increased risk of opportunistic infection or dominance by native antibiotic-resistant microbes, e.g. *Enterococcus faecalis*. In healthy subjects, return to a stable microbial community is typically achieved in the weeks following treatment; however, the trajectory and completeness of this recovery is variable. Therefore, promoting a faster return of the microbiome to a "healthy" state is key in reducing microbially-linked antibiotic side effects.

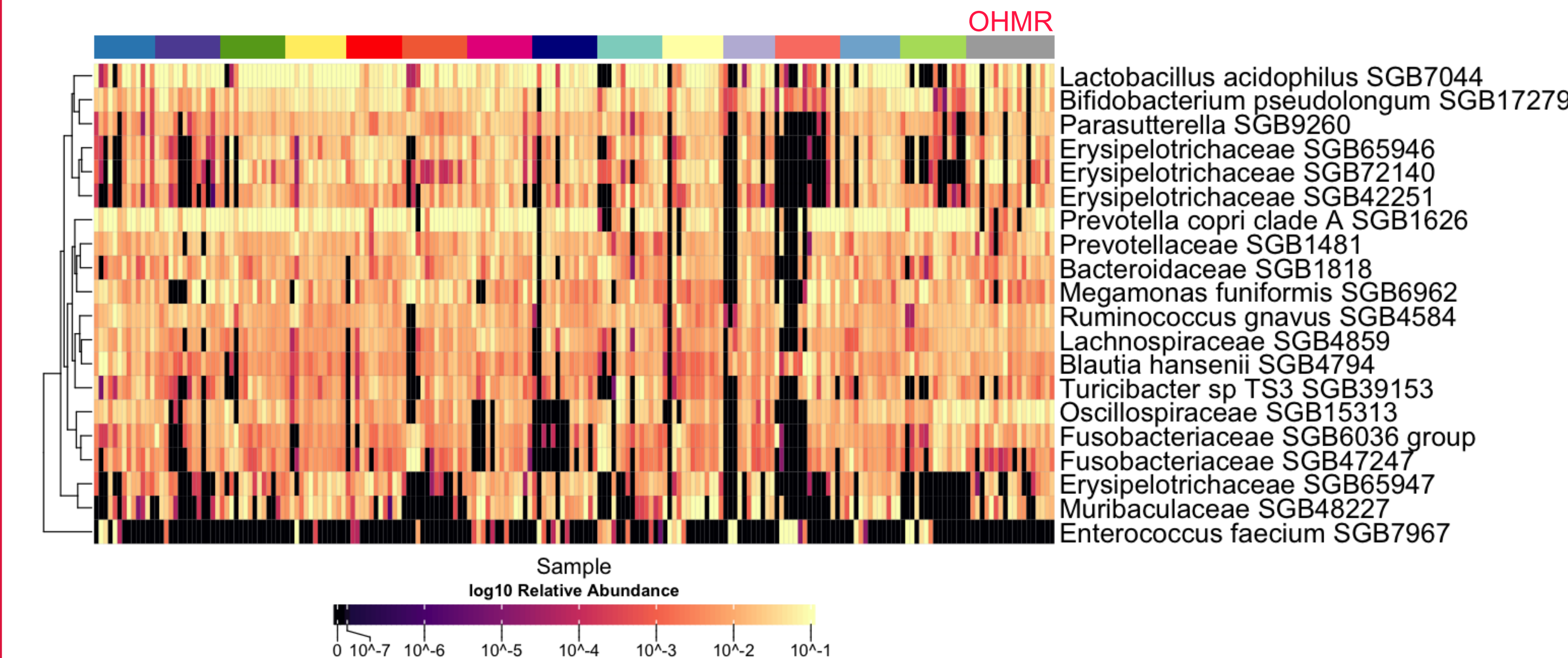
## Longitudinal canine gut microbiome samples

In our preliminary analyses, 14 dogs (of 40 recruited) with clinical indications requiring antibiotic treatment were fed either a standard maintenance (n = 7) or a veterinary therapeutic food (n = 7) enriched in fiber. Fecal samples were periodically collected during and after ABX treatments and associated sequencing data and metadata were collected.



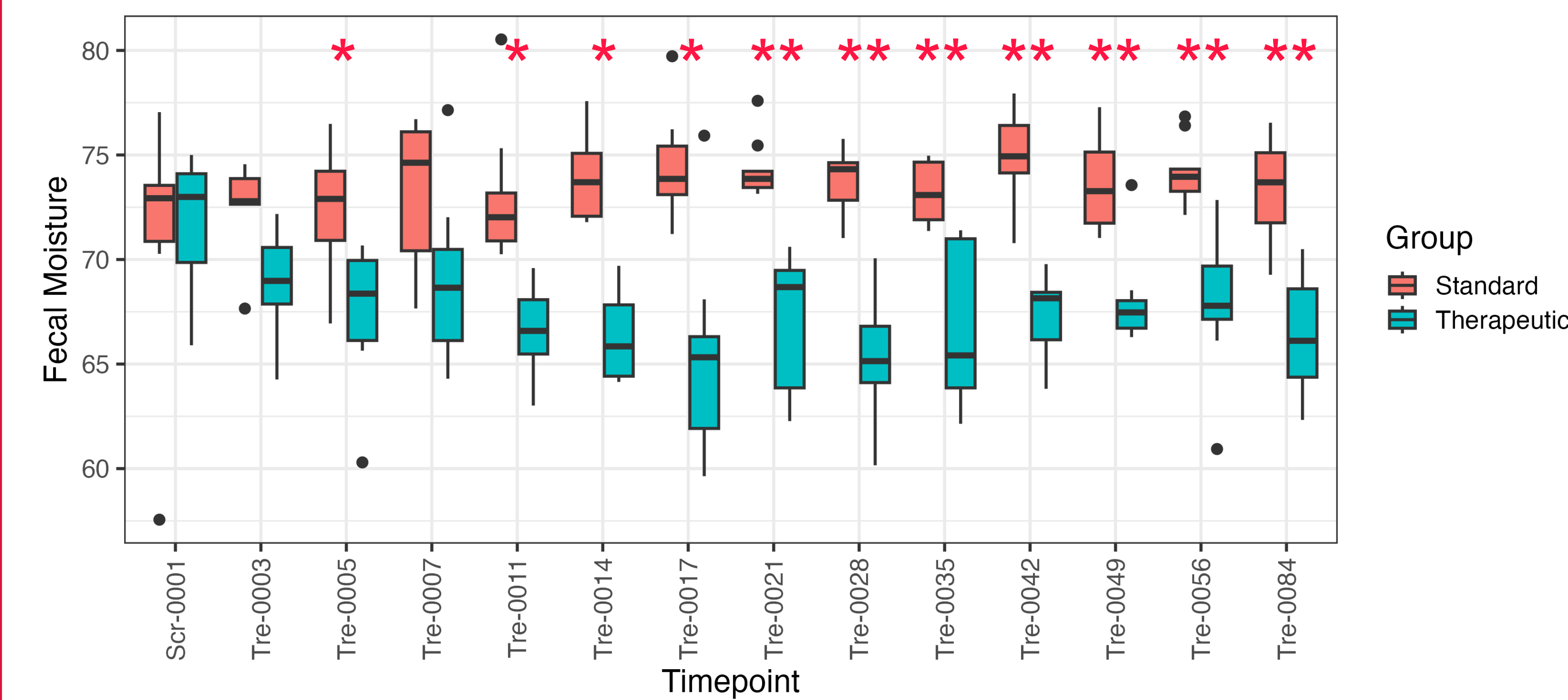
Sampling timepoints for the pilot cohort of dogs colored by antibiotics status. Timepoints are denser near the start of treatment to capture potentially rapid microbiome changes.

## Taxonomic profiles are similar to healthy comparators with some clear perturbations



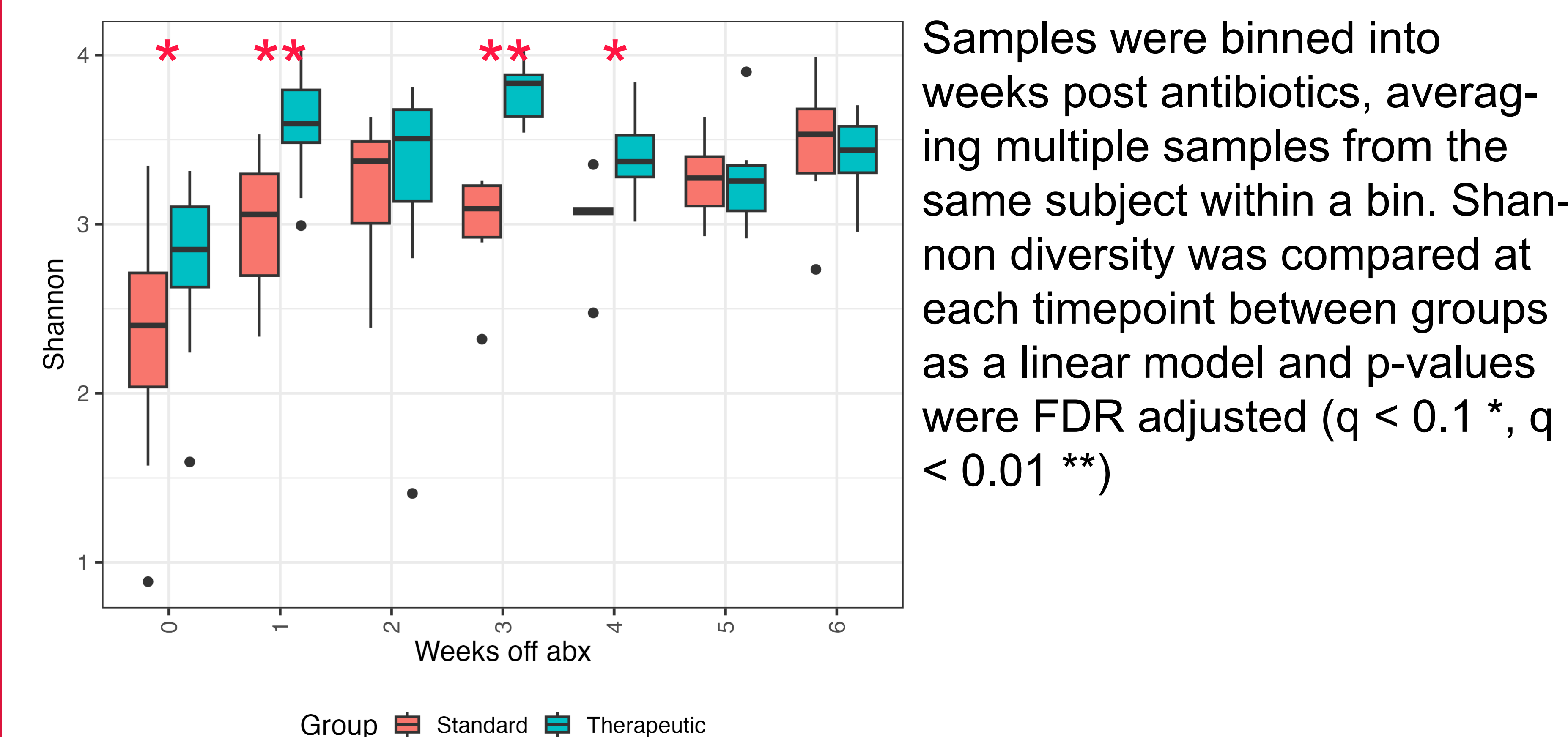
Relative abundance of top 20 species-level genomic bins (SGBs) produced by MetaPhlan 4, ordered by time and grouped by dog (colored bars). Right-most group (gray bar) is 19 healthy samples from One Health Microbiome Resource (OHMR) selected to serve as a baseline (from the same location, no recent medication or chronic conditions).

## Fecal moisture differs across foods over time



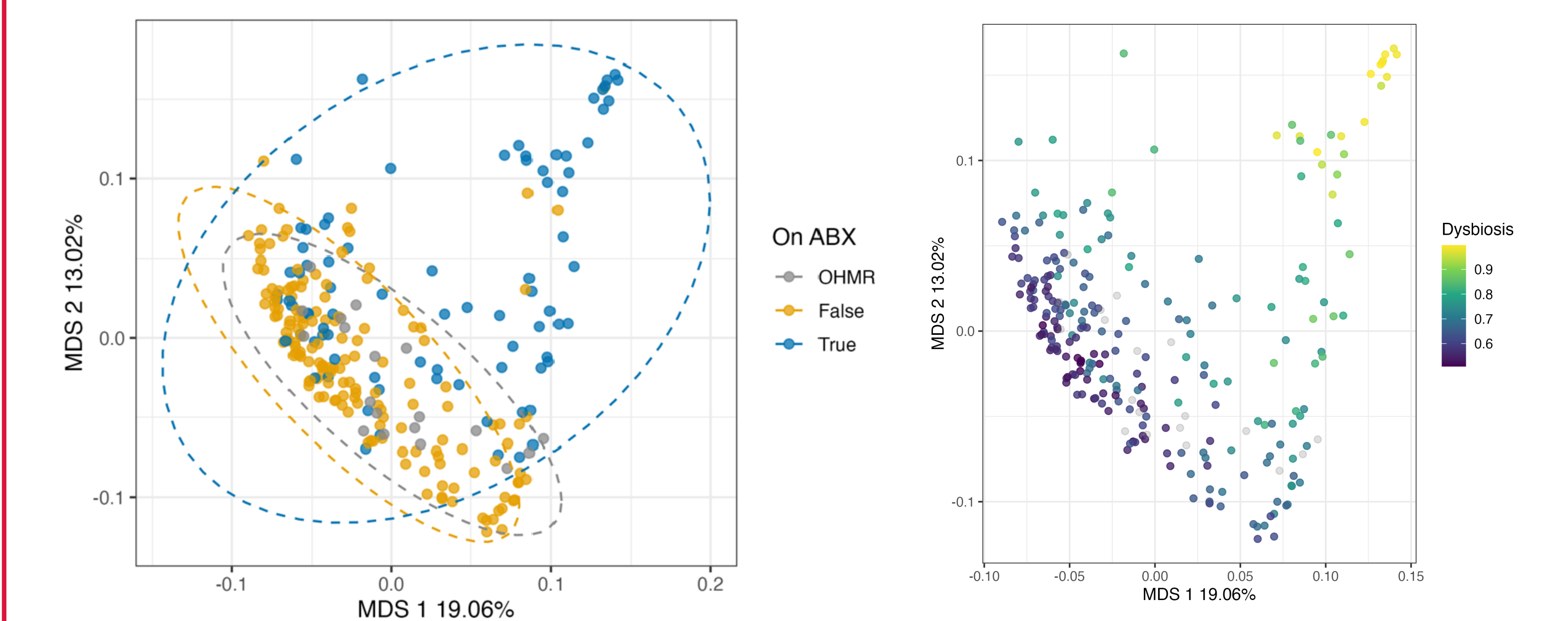
Fecal moisture was compared at each timepoint between groups as a linear model and p-values were FDR adjusted ( $q < 0.1$  \*,  $q < 0.01$  \*\*)

## Diversity differs across foods post-antibiotics



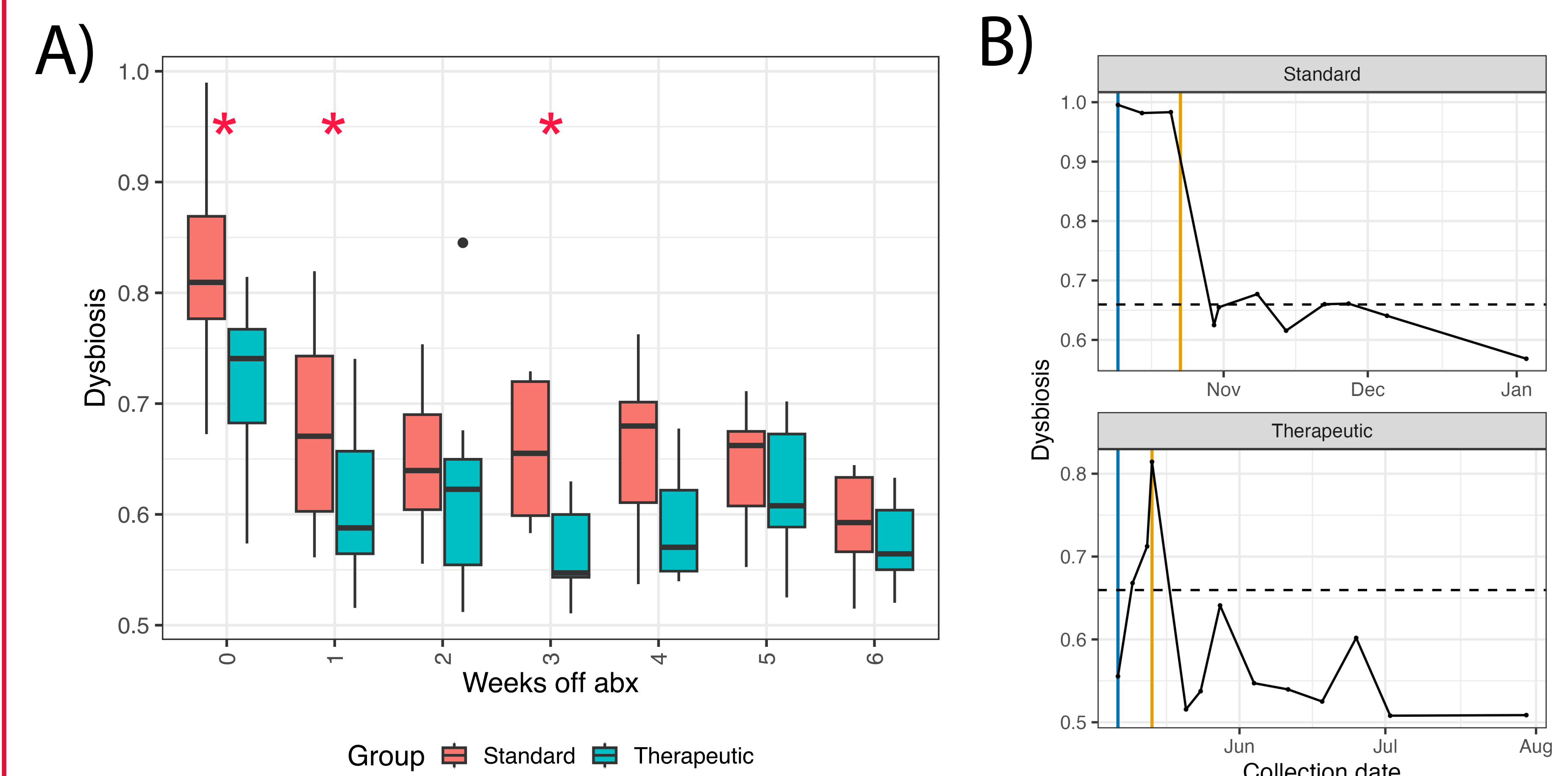
Samples were binned into weeks post antibiotics, averaging multiple samples from the same subject within a bin. Shannon diversity was compared at each timepoint between groups as a linear model and p-values were FDR adjusted ( $q < 0.1$  \*,  $q < 0.01$  \*\*)

## Bray-Curtis dissimilarity shows antibiotics related differences from a healthy baseline



PCoA of experimental samples on-ABX (blue), post-ABX (yellow), and OHMR baseline samples. Some but not all ABX samples are well separated from baseline. Distance from "healthy" is quantified by "dysbiosis", defined here as the median dissimilarity from baseline samples (Lloyd-Price 2019).

## Dysbiosis is significantly lower in therapeutic group shortly after end of treatment



A) Dysbiosis was compared at each timepoint between groups as a linear model and p-values were FDR adjusted ( $q < 0.1$  \*)  
B) Two subject-level dysbiosis trajectories are shown. Start and end of treatment are indicated by blue and orange lines, respectively. Dotted horizontal line indicates a cutoff of 90th percentile of scores within the baseline samples.

## Acknowledgments

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[tkuntz@hsph.harvard.edu](mailto:tkuntz@hsph.harvard.edu)  
<http://huttenhower.sph.harvard.edu>

