

# What's in Your Hutch?

## An evaluation of the microbial community present in dairy calf hutches and its relationship with the calf microbiome

Department of Animal Science

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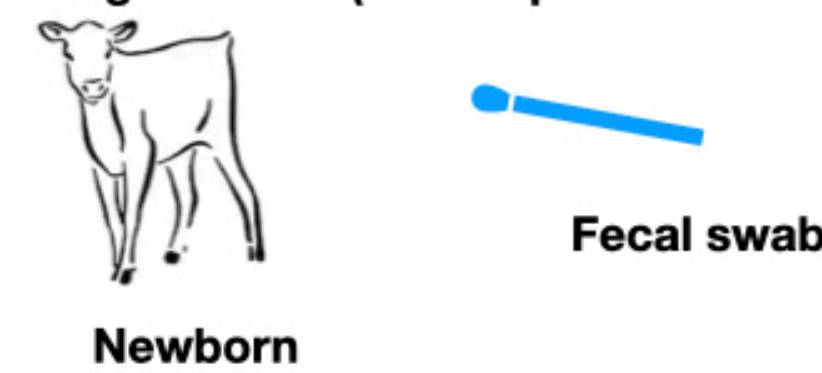
### Introduction

- Calf immune systems are very fragile for the first few months of life. Infections from bacteria such as *E. Coli* can cause severe diarrhea and even death.
- Colonization of the microbiome by microbes that are essential to the rumen environment begins after weaning.
- Individual calf hutches were developed to isolate the calf and protect them from disease spread from other calves.
- The objective of this study was to determine the microbiomes of the hutch and the calf, and how the two influence each other over time.

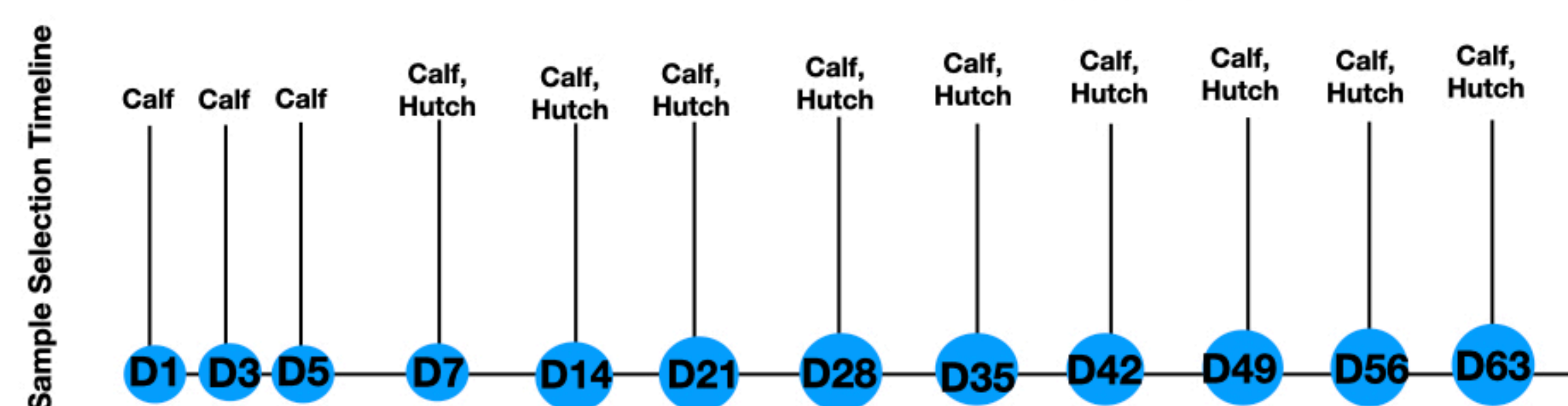
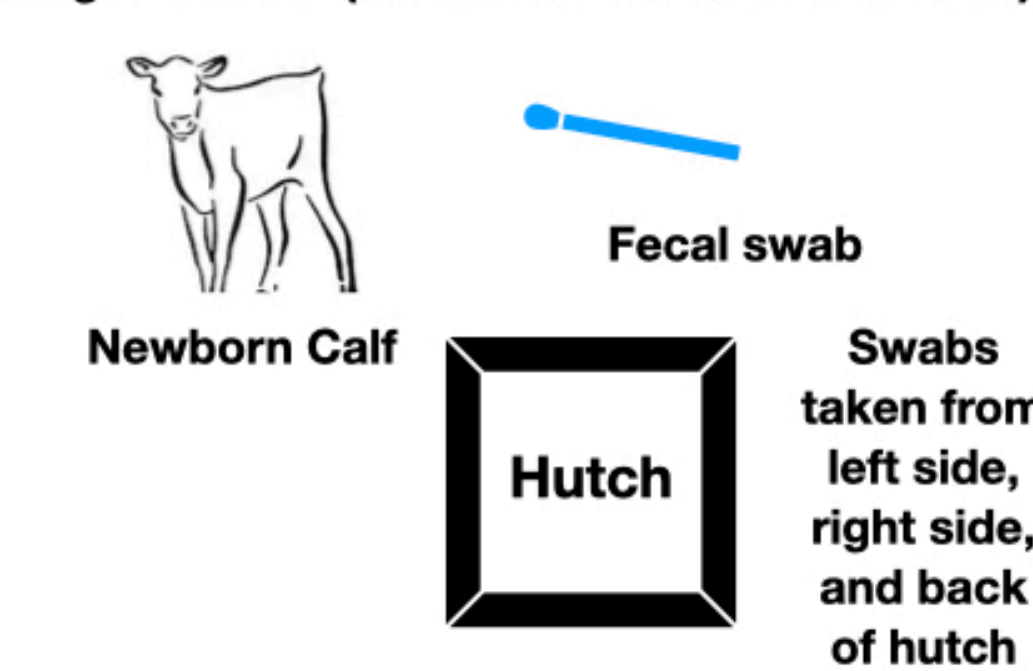
### Methods

#### Sample Collection

Stage 1: Birth (First exposure to microbes)



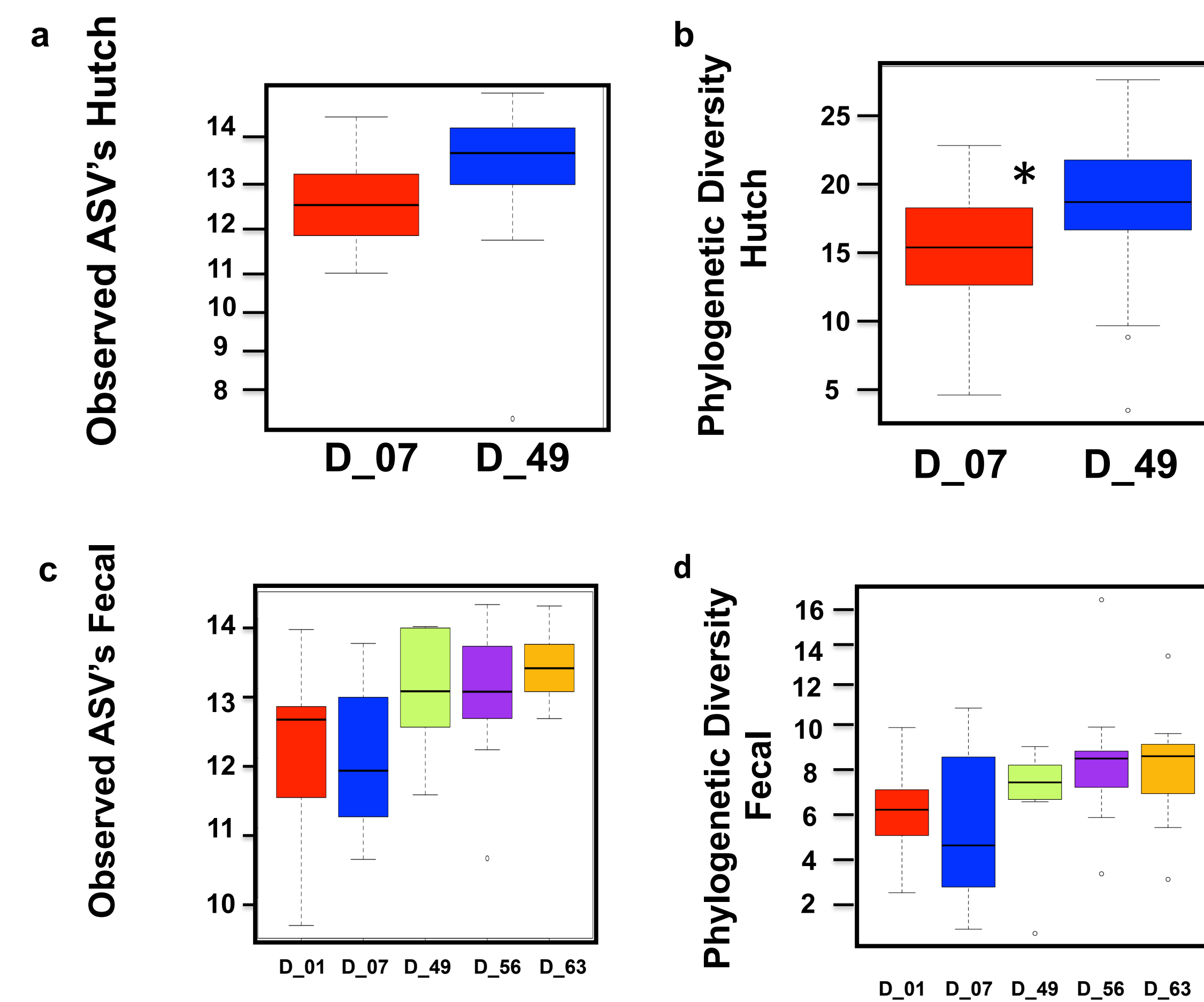
Stage 2: Hutch (Establishment of microbiome)



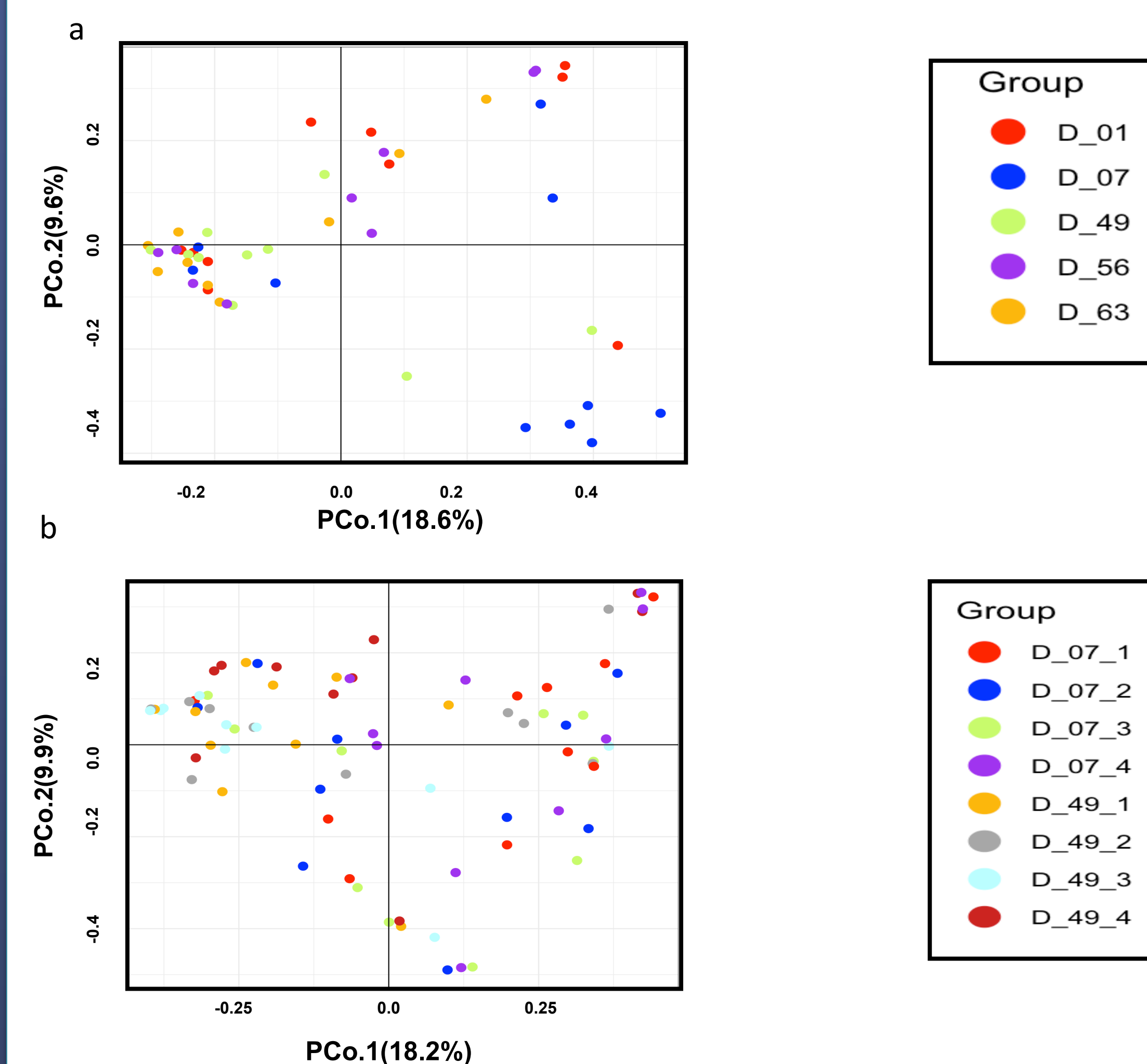
#### Data Analysis

- DNA was extracted from swabs using QIAgen Powersoil DNA extraction kit and sent to the University of Minnesota Genomic Center for sequencing and amplification of the 16s rRNA V4 region
- Sequences were filtered and trimmed using a bioinformatics pipeline within Qiime2 and DADA2, run through MSI.
- High quality sequences were sorted into Amplicon Sequence Variants (ASVs) with assigned taxonomy using the GreenGenes database.
- Different packages in the R statistical software was used to run multivariate and univariate statistical analyses, as well as distance based-ordination and similarities analyses of bacterial communities in hutches and calves.

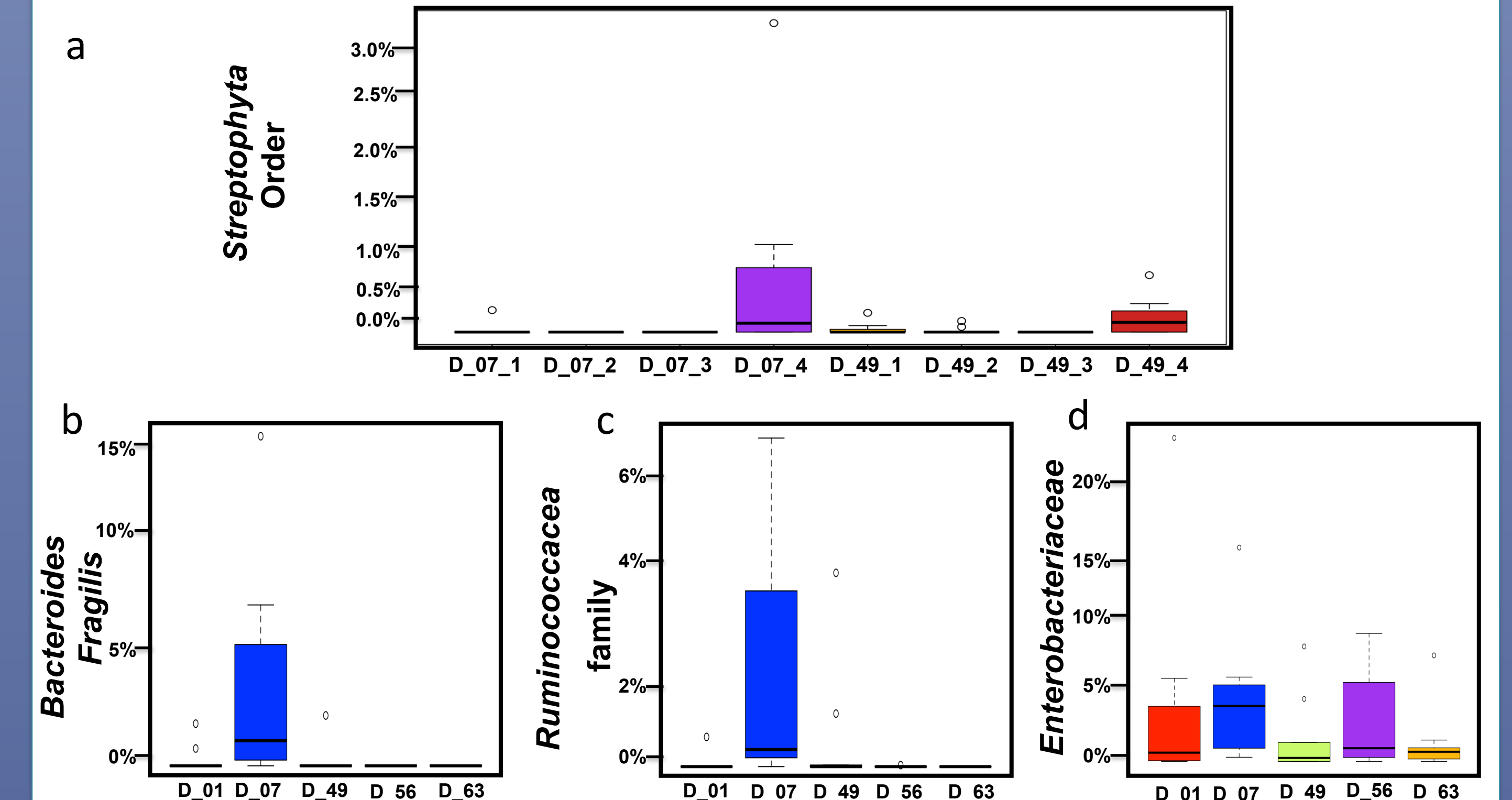
### \* Results



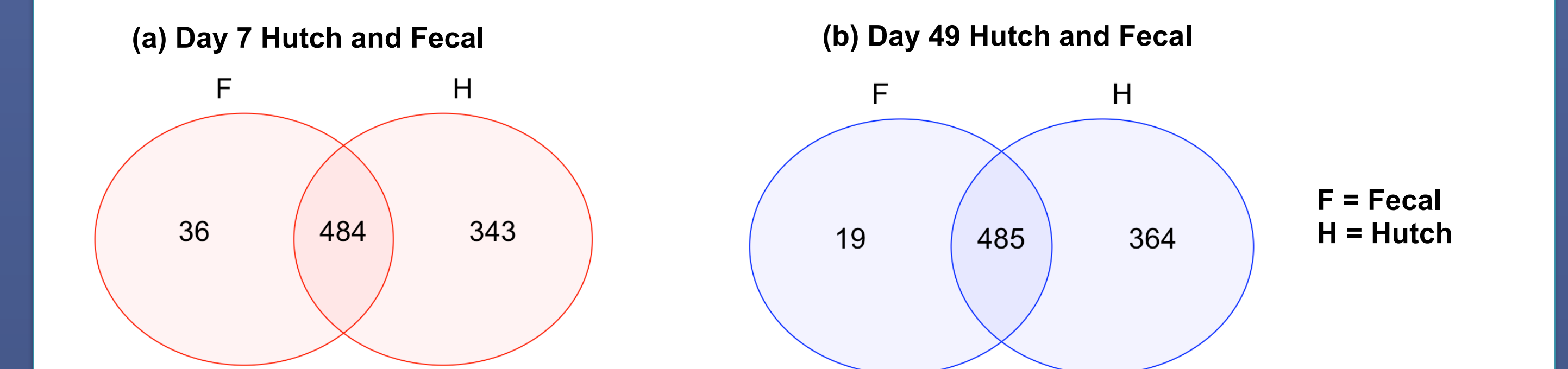
**Figure 2. The microbiome diversity of the hutch and calf change over time.** These distinctions were seen for alpha diversity in the hutch (a,b) and calf samples (c,d); Alpha diversity was statistically significant in the hutch at both time points (Observed ASV's,  $p=0.003069$ ; Phylogenetic Diversity,  $p=0.02207$ ). The change over time calf microbiome diversity was found to be statistically significant (Observed ASV's,  $p=0.01$ ), but the changes in phylogenetic diversity were not found to be statistically significant ( $p=0.1958$ ).



**Figure 3. Beta Diversity.** (a) Calf samples varied between individuals and were statistically significant (PERMANOVA,  $R^2=0.11659$ ,  $p=0.016$ ). (b) Hutch samples also varied between surfaces and showed statistical significance, (PERMANOVA,  $R^2=0.12582$ ,  $p=0.004$ ).



**Figure 4. Indval, and Enterobacteriaceae distribution.** Calf samples had two distinct indval species (b,c) that had statistically significant changes over time (*B. Fragilis*  $p=6.2e-5$ , *Ruminococcaceae*  $p=3.6e-5$ ). Hutch samples only had one (a), but the change in *Streptophyta* not significant ( $p=0.2$ ). Enterobacteriaceae distribution in calves changed over time but was not statistically significant (d) ( $p=0.07$ ).



**Figure 5. Venn diagrams showing number of ASVs shared between fecal and hutch samples.**

### Conclusions

- The microbiome of the calf hutch increases in both the number of bacterial species present and the populations of those species from day 7 to day 49.
- The calf microbiome is highly variant between individuals and has a significant increase in number of bacteria present over time, especially around day 7 in *B. Fragilis* and the family *Ruminococcaceae*. However, the number of *Enterobacteriaceae* did not change significantly over time.
- Both the calf and hutch share lots of ASV's, but the number of these shared ASV's did not change from day 7 to day 49.

### Acknowledgements

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