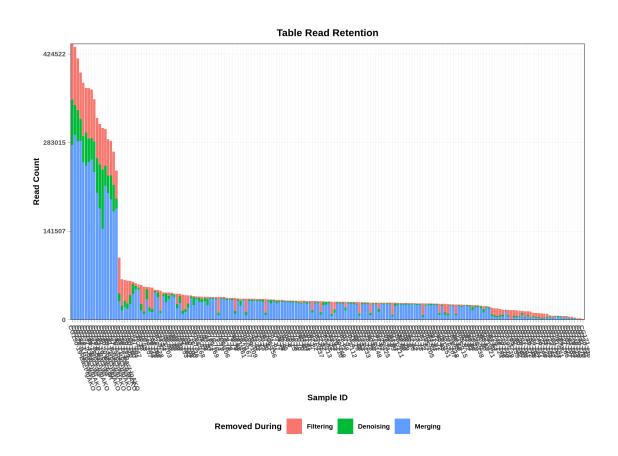
## **MMEDS** Analysis Summary

Clemente Lab
Icahn School of Medicine at Mount Sinai

(Dated: May 16, 2022)

#### I. Table Statistics Summary

#### a. Dada2 Statistics



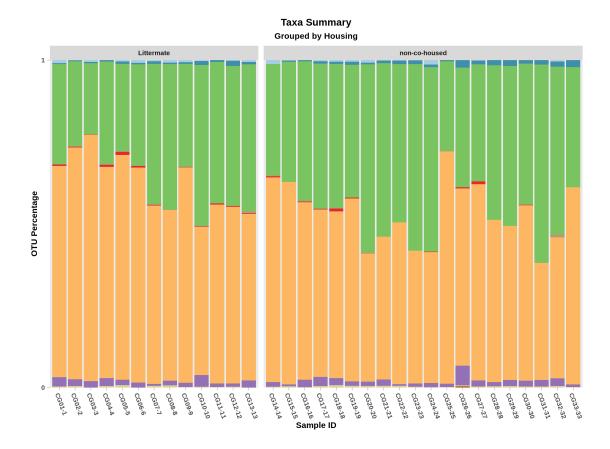
The above plot represents number of input reads (total bar length) and retained (magenta) after quality control filtering, including denoising and chimera checking.

#### II. Taxonomy Summary

#### a. Interpreting Taxonomy Results

Taxonomy plots represent the abundance of different taxa using stacked plots on a per-sample or per-group (averaged) basis. Data is normalized so that abundances per sample or per group add up to 100%. When using group-based taxonomy plots, it should be noted that only average abundances are shown per group and taxa: this can induce visual biases when a small number of samples in a group have significantly higher abundance of a given taxa compared to the rest of samples in the group, and give the (incorrect) impression that the group as a whole has high high abundance of the taxa.

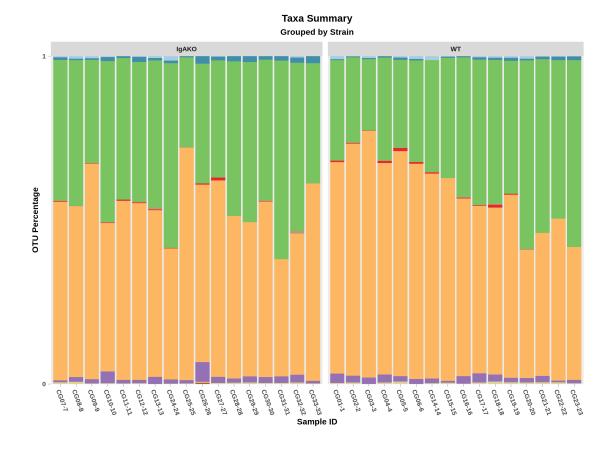
#### b. Phylum Level



Legend grouped by Housing
Color Abundance OTU

45.07% k\_\_Bacteria;p\_\_Bacteroidetes
51.17% k\_\_Bacteria;p\_\_Firmicutes

The above plot represents the percentage of each sample belonging to particular taxon summarized at the Phylum level.



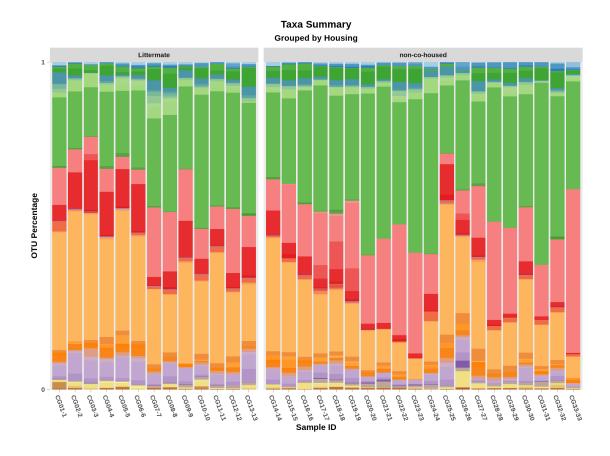
# Legend grouped by Strain Color Abundance OTU 39.07% k\_\_Bacteria;p\_\_Bacteroidetes

 $k\_Bacteria; p\_Firmicutes$ 

57.40%

The above plot represents the percentage of each sample belonging to particular taxon summarized at the Phylum level.

#### c . Genus Level



Legend grouped by Housing
Color Abundance OTU

37.24%

k\_Bacteria;p\_Bacteroidetes;c\_Bacteroidia;o\_Bacteroidales;f\_S24-7;g\_

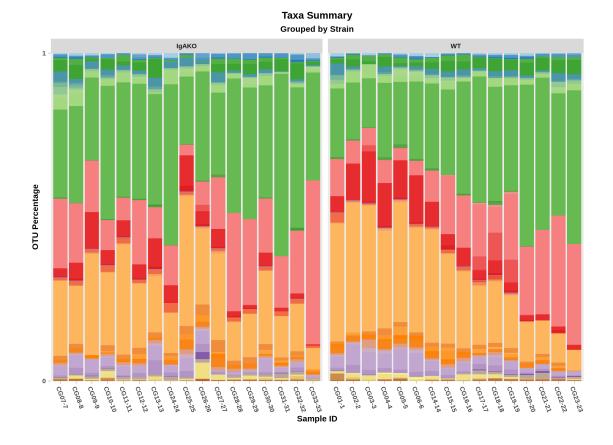
22.88%

k\_Bacteria;p\_Firmicutes;c\_Bacilli;o\_Lactobacillales;f\_Lactobacillaceae;g\_Lactobacillus

19.75%

k\_Bacteria;p\_Firmicutes;c\_Clostridia;o\_Clostridiales;f\_Lachnospiraceae;\_\_

The above plot represents the percentage of each sample belonging to particular taxon summarized at the Genus level.



```
Legend grouped by Strain
Color Abundance OTU

30.59%
k__Bacteria;p__Bacteroidetes;c__Bacteroidia;o__Bacteroidales;f__S24-7;g__
21.31%
k__Bacteria;p__Firmicutes;c__Bacilli;o__Lactobacillales;f__Lactobacillaceae;g__Lactobacillus
13.05% k__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridiales;__;__
26.59%
k__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridiales;f__Lachnospiraceae;__
```

The above plot represents the percentage of each sample belonging to particular taxon summarized at the Genus level.

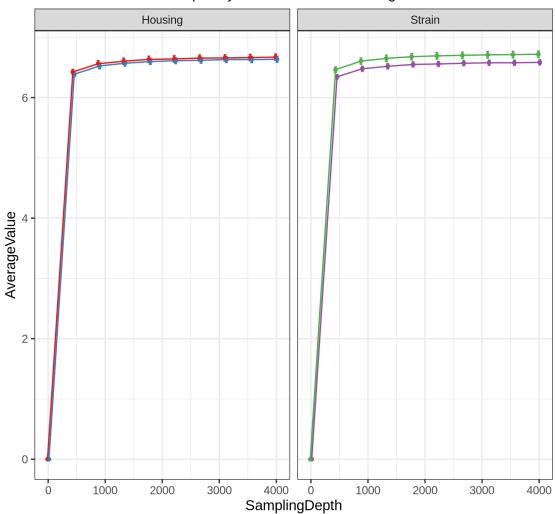
#### III. Alpha Diversity

#### a. Interpreting Alpha Diversity Results

Alpha diversity estimates the amount of microbial diversity present in a sample or group of samples. There are several measures that can be used for alpha diversity, including observed features, Shannon's diversity or Faith's phylogenetic diversity. Because diversity estimates depend on the total number of sequences assigned to each sample, rarefaction curves are constructed to show the relation between alpha diversity (on the vertical axis) and sequencing depth (on the horizontal axis). Curves that gradually plateau as sequencing depth increases suggest that additional sequencing effort would not substantially yield additional results in terms of currently not observed diversity; curves that continue to increase suggest additional sequencing effort might be required to saturate the estimate.

#### b. Shannon Diversity

Alpha Diversity
Grouped by Discrete Metadata Categories





Littermate

non-co-housed

Legend for Strain Color Metadata



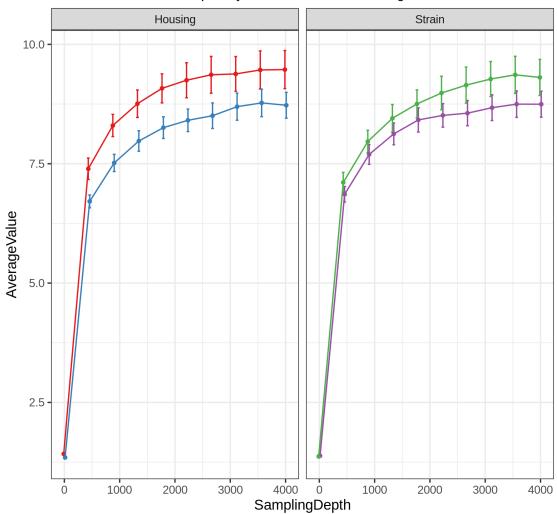
IgAKO



The above plot represents the average value of alpha diversity at each sampling depth. The error bars show the standard error within each group. Groups are determined by the metadata value in each category specified in the plot.

## **c** . Faith's Phylogenetic Diversity

Alpha Diversity
Grouped by Discrete Metadata Categories

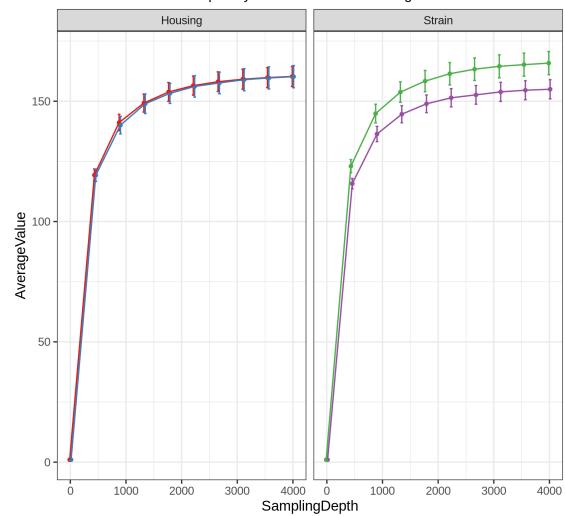




The above plot represents the average value of alpha diversity at each sampling depth. The error bars show the standard error within each group. Groups are determined by the metadata value in each category specified in the plot.

## ${\bf d}$ . Observed ASV

Alpha Diversity
Grouped by Discrete Metadata Categories



Legend for Housing
Color Metadata

Littermate
non-co-housed

Legend for Strain
Color Metadata

IgAKO

The above plot represents the average value of alpha diversity at each sampling depth. The error bars show the standard error within each group. Groups are determined by the metadata value in each category specified in the plot.

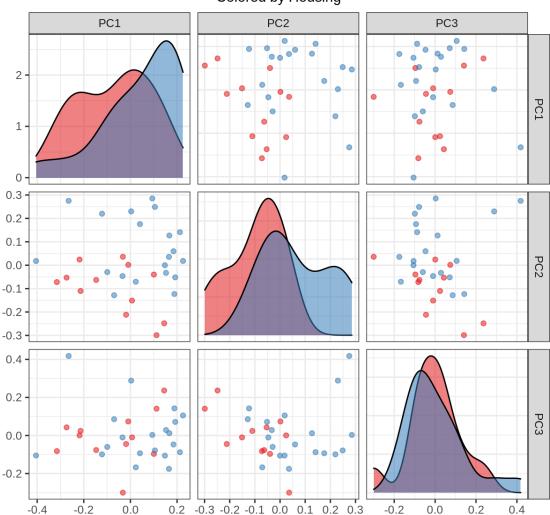
#### IV. Beta Diversity

#### a. Interpreting Beta Diversity Results

Beta diversity estimates how similar or dissimilar samples are based on their microbiome composition. Different to alpha diversity, which is estimated per sample, beta diversity is a distance that is calculated between pairs of samples. Samples that are similar to each other in their microbiome composition will have a low distance between them based on beta diversity, while those that are very different in their composition will have a large distance. Principal Coordinate Analysis (PCoA) is an ordination technique that visually represents the samples based on their beta diversity distances to facilitate the identification of clusters or gradients of samples. By default, the first three principal coordinates are shown in PCoA plots.

#### b. Bray-Curtis, grouped by Housing



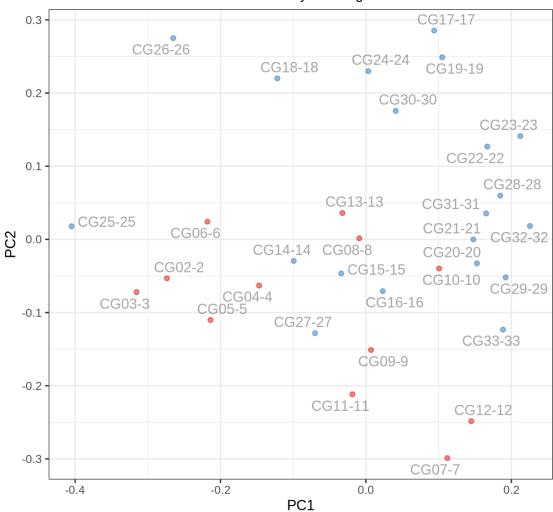




Littermate non-co-housed

The above plot represents the first three compenents created when performing Principle Component Analysis on the Beta diversity of the samples.

PC1 vs. PC2
Colored by Housing

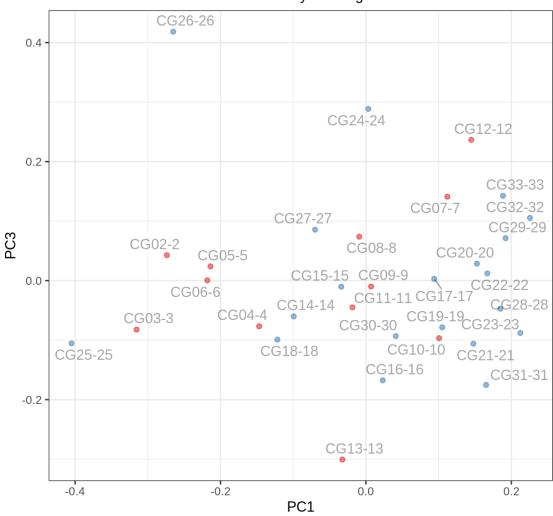


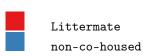
Legend for Housing Color Metadata



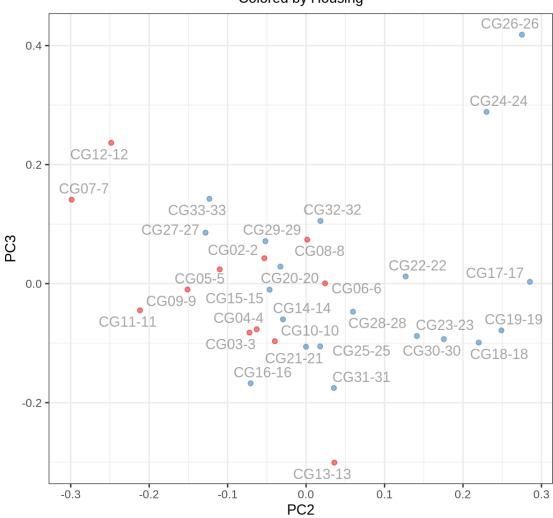
Littermate non-co-housed

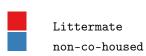
PC1 vs. PC3
Colored by Housing





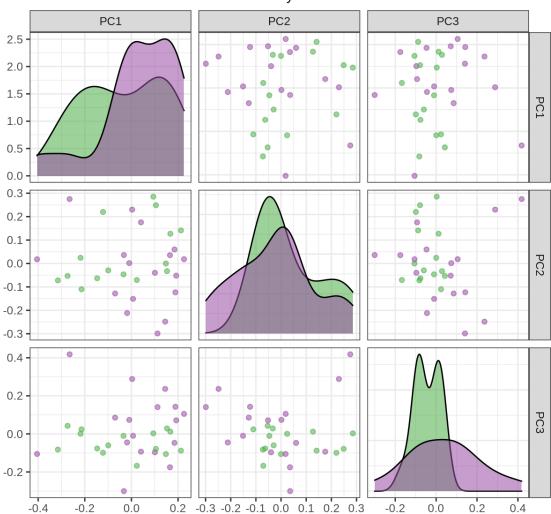
PC2 vs. PC3 Colored by Housing





## c . Bray-Curtis, grouped by Strain

# PCA plot Colored by Strain

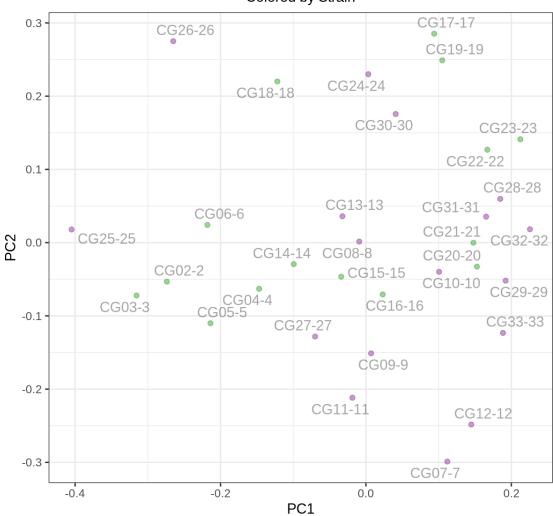


Legend for Strain Color Metadata



The above plot represents the first three compenents created when performing Principle Component Analysis on the Beta diversity of the samples.

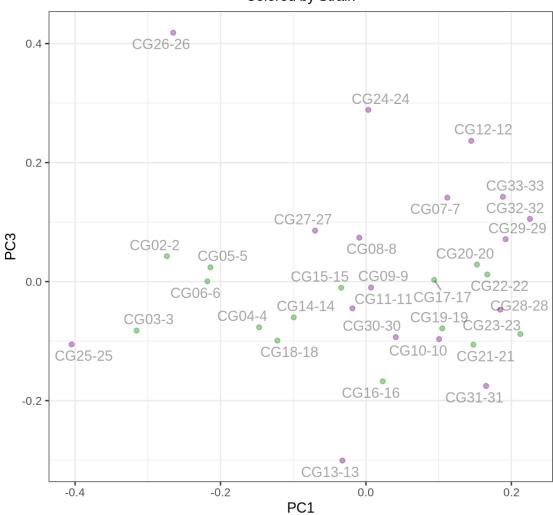
PC1 vs. PC2 Colored by Strain





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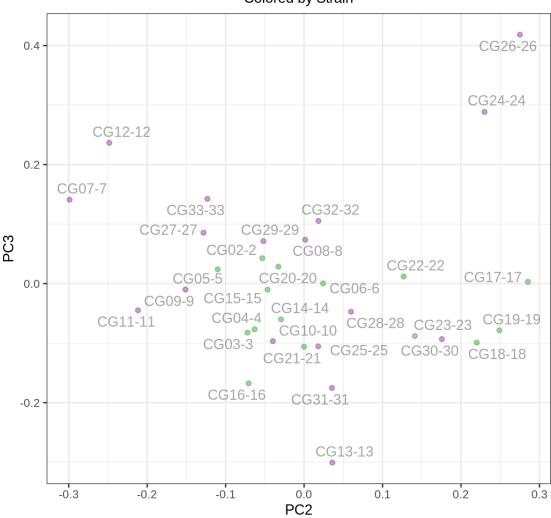
PC1 vs. PC3 Colored by Strain





IgAKO

PC2 vs. PC3 Colored by Strain

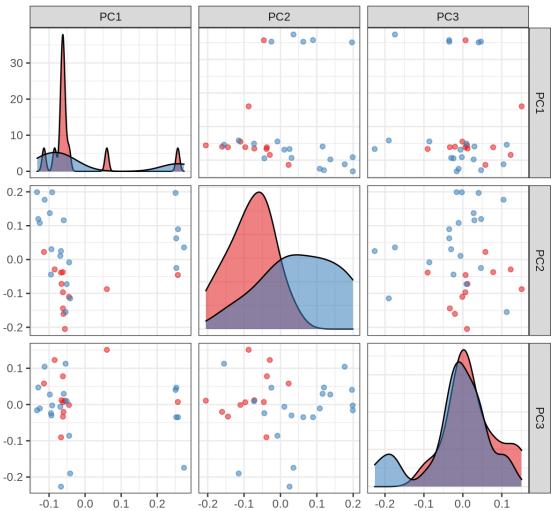




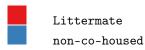
IgAKO

## d. Unweighted UniFrac, grouped by Housing



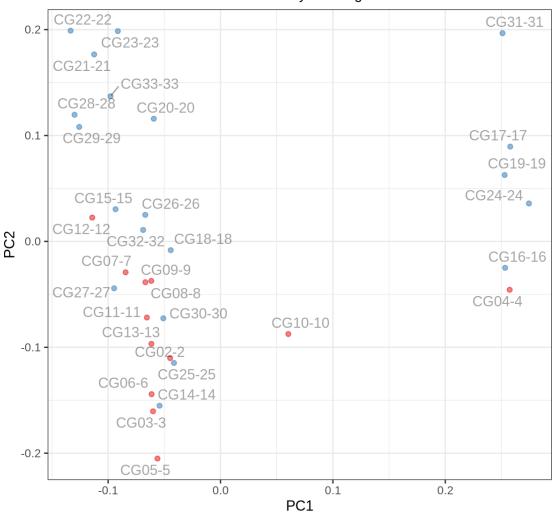


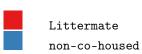
Legend for Housing Color Metadata



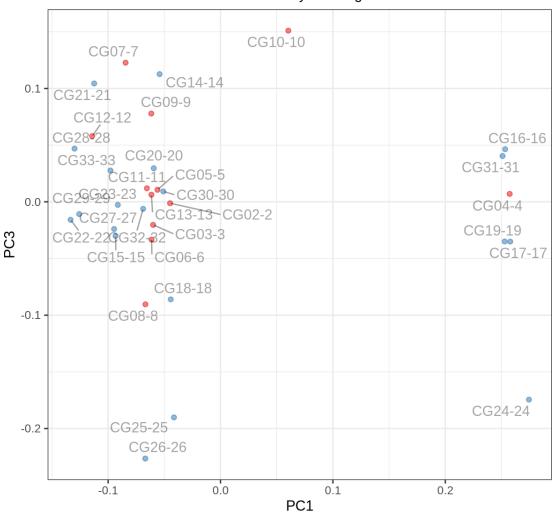
The above plot represents the first three compenents created when performing Principle Component Analysis on the Beta diversity of the samples.

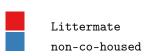
PC1 vs. PC2 Colored by Housing



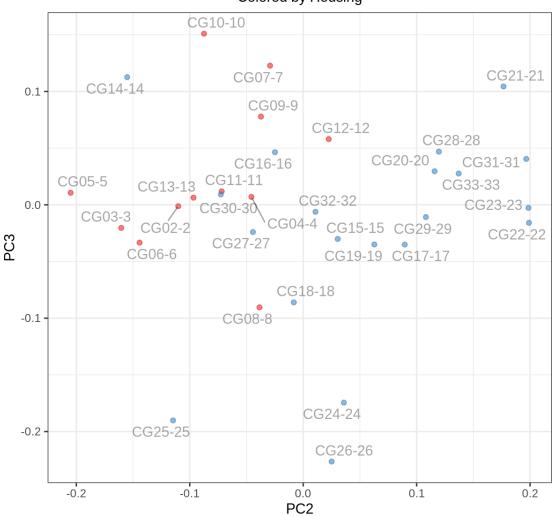


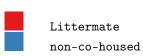
PC1 vs. PC3
Colored by Housing



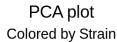


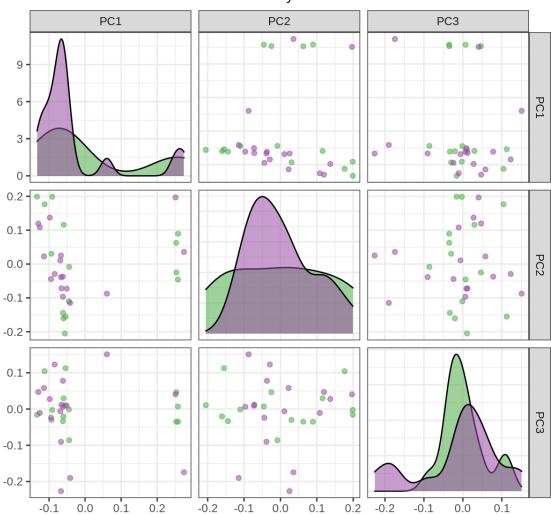
PC2 vs. PC3
Colored by Housing





## e. Unweighted UniFrac, grouped by Strain



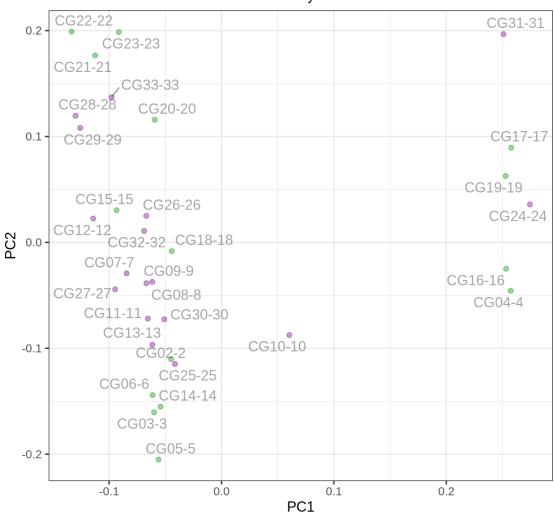


Legend for Strain Color Metadata



The above plot represents the first three compenents created when performing Principle Component Analysis on the Beta diversity of the samples.

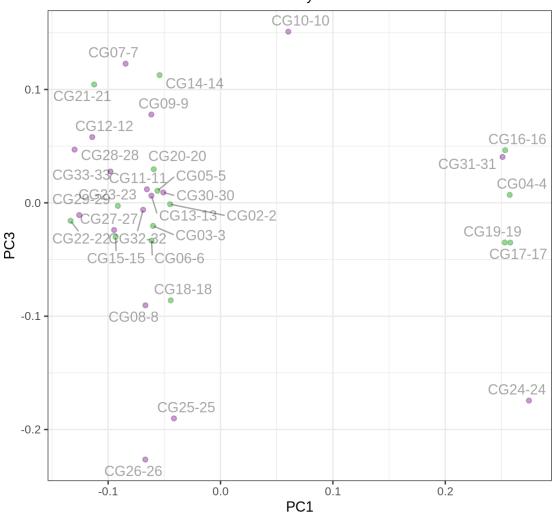
PC1 vs. PC2 Colored by Strain





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PC1 vs. PC3 Colored by Strain

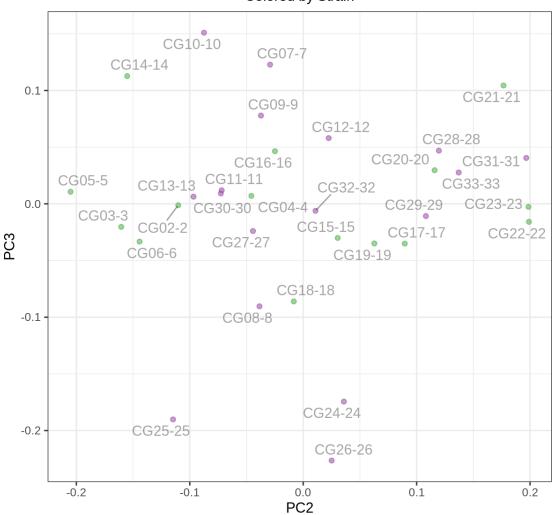




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 $\mathtt{WT}$ 

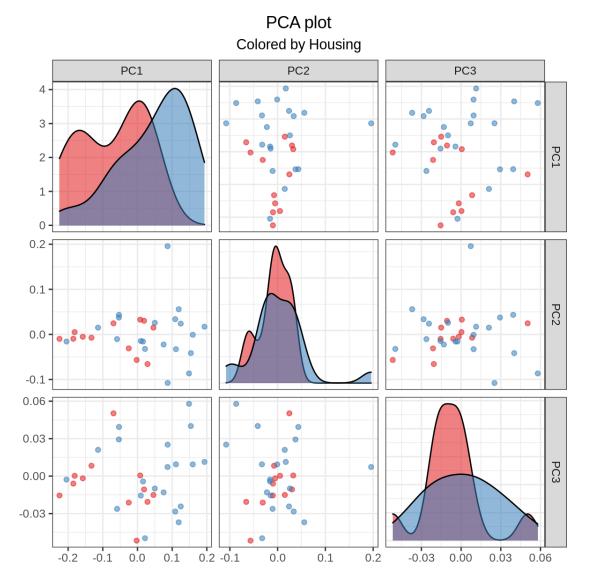
PC2 vs. PC3 Colored by Strain

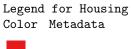


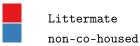


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## f. Weighted UniFrac, grouped by Housing

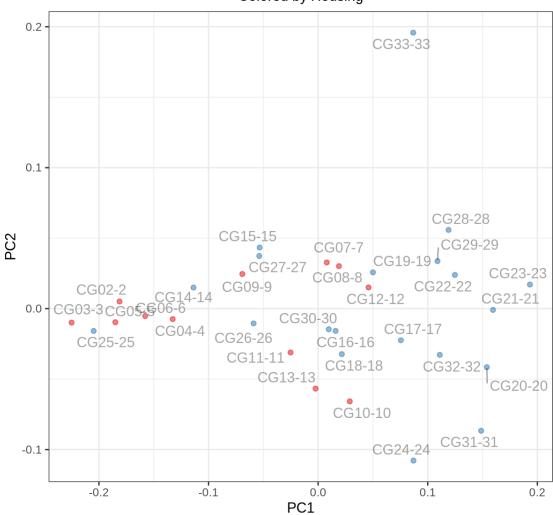


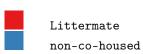




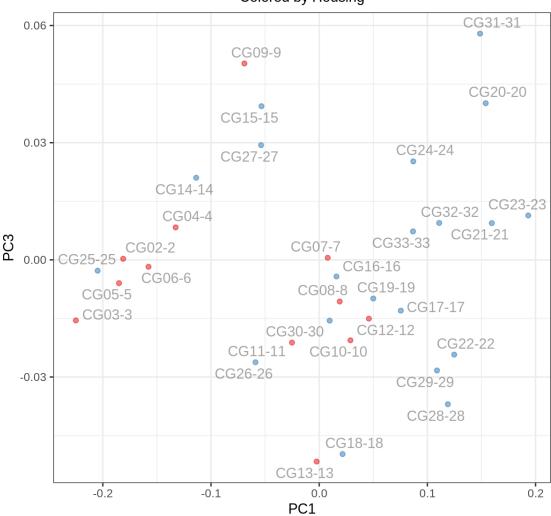
The above plot represents the first three compenents created when performing Principle Component Analysis on the Beta diversity of the samples.

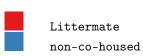
PC1 vs. PC2 Colored by Housing



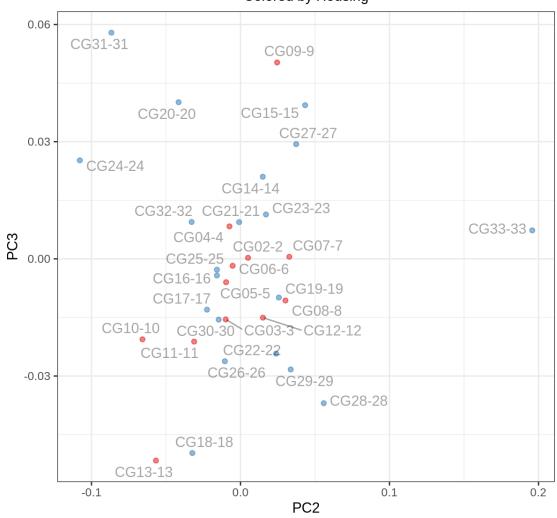


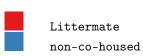
PC1 vs. PC3
Colored by Housing





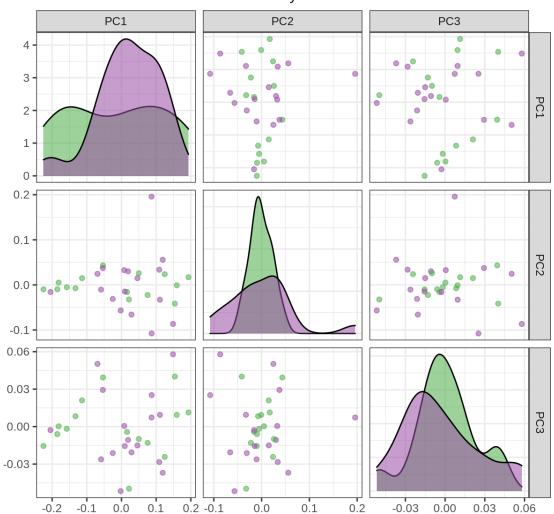
PC2 vs. PC3 Colored by Housing





## ${f g}$ . Weighted UniFrac, grouped by Strain



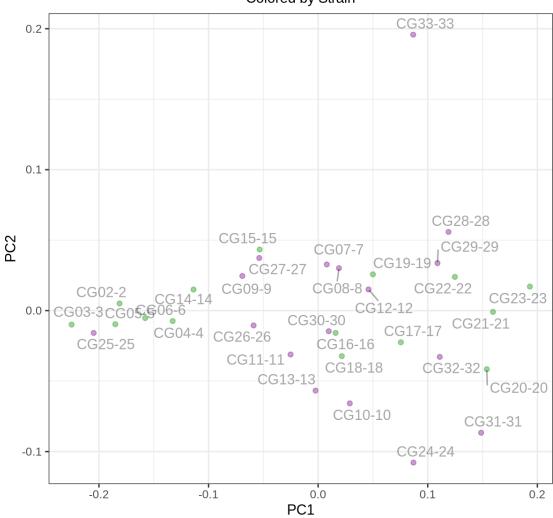


Legend for Strain Color Metadata



The above plot represents the first three compenents created when performing Principle Component Analysis on the Beta diversity of the samples.

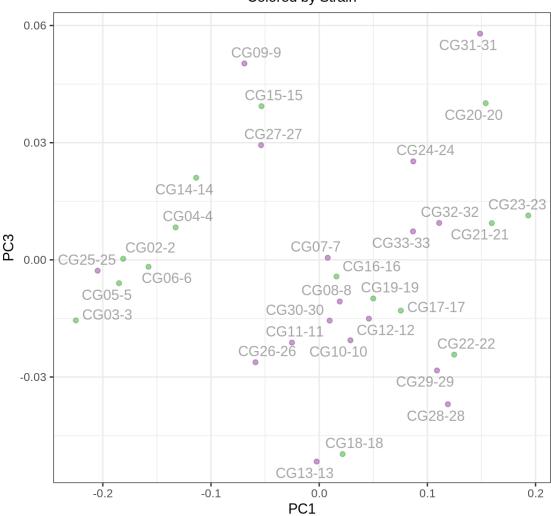
PC1 vs. PC2 Colored by Strain





IgAKO

PC1 vs. PC3 Colored by Strain

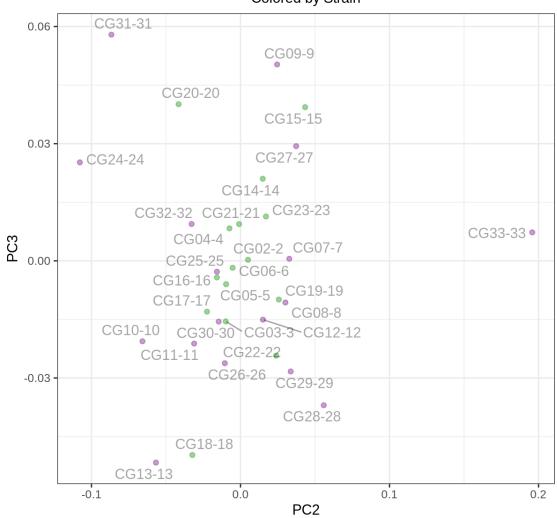




IgAKO

 $\mathtt{WT}$ 

PC2 vs. PC3 Colored by Strain





IgAKO