### H.pylori Rhesus Microbiome Analysis

Noah Siegel

### Data used in analysis

#### Library

```
suppressPackageStartupMessages({
  library(sjPlot)
  library(readxl)
  library(phyloseq)
  library(microbiome)
  library(DESeq2)
  library(qiime2R)
  library(tidyverse)
  library(tidyMicro)
  library(kableExtra)
  library(magrittr)
  library(ggpubr)
  library(microeco)
  library(tidytree)
  library(RColorBrewer)
  library(questionr)
  library(rmarkdown)
  library(stringr)
})
```

ASV counts the taxa table are linked below:

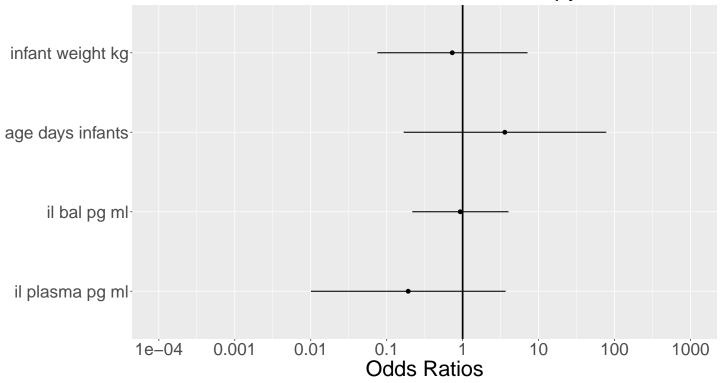
**ASV** counts

Taxa table

Table 1: H.pylori Metadata

| SampleID              | Treatment         | site | Site_status                              | sex    | log10_cfu.gm | IL8_Lavage | IL8_Plasma |
|-----------------------|-------------------|------|--|--------|--------------|------------|------------|
| LMiller_00458.BAL     | H.pylori_(-)      | BAL  | H.pylori_(-)_BAL                         | Female | 0.00         | NA         | 463.376    |
| LMiller_00458.Swab    | H.pylori_(-)      | Swab | H.pylori_(-)_Swab                        | Female | 0.00         | NA         | 463.376    |
| LMiller_00459.BAL     | H.pylori_(-)      | BAL  | H.pylori_(-)_BAL                         | Female | 0.00         | NA         | 258.416    |
| LMiller_00459.Swab    | H.pylori_(-)      | Swab | H.pylori_(-)_Swab                        | Female | 0.00         | NA         | 258.416    |
| $LMiller\_00461.BAL$  | $H.pylori_(+)$    | BAL  | $H.pylori_(+)_BAL$                       | Female | 7.66         | NA         | 704.550    |
| $LMiller\_00461.Swab$ | $H.pylori_(+)$    | Swab | $H.pylori_(+)_Swab$                      | Female | 7.66         | NA         | 704.550    |
| $LMiller\_00462.BAL$  | H.pylori_(-)      | BAL  | H.pylori_(-)_BAL                         | Female | 0.00         | 19.104     | 1075.626   |
| $LMiller\_00462.Swab$ | H.pylori_(-)      | Swab | H.pylori_(-)_Swab                        | Female | 0.00         | 19.104     | 1075.626   |
| $LMiller\_00463.BAL$  | H.pylori_(-)      | BAL  | H.pylori_(-)_BAL                         | Female | 0.00         | 11.590     | 423.590    |
| $LMiller\_00466.BAL$  | $H.pylori_(+)$    | BAL  | $H.pylori_(+)_BAL$                       | Female | 3.81         | 11.201     | 642.148    |
| $LMiller\_00466.Swab$ | H.pylori_(+)      | Swab | H.pylori_(+)_Swab                        | Female | 3.81         | 11.201     | 642.148    |
| $LMiller\_00467.BAL$  | $H.pylori_(+)$    | BAL  | $H.pylori_(+)_BAL$                       | Female | 6.09         | 15.511     | 425.782    |
| $LMiller\_00467.Swab$ | $H.pylori_(+)$    | Swab | $H.pylori_(+)_Swab$                      | Female | 6.09         | 15.511     | 425.782    |
| $LMiller\_00468.BAL$  | H.pylori_(-)      | BAL  | H.pylori_(-)_BAL                         | Female | 0.00         | 15.511     | 660.911    |
| $LMiller\_00468.Swab$ | $H.pylori_{-}(-)$ | Swab | H.pylori_(-)_Swab                        | Female | 0.00         | 15.511     | 660.911    |
| $LMiller\_00469.BAL$  | $H.pylori_(+)$    | BAL  | $H.pylori_(+)_BAL$                       | Female | 6.38         | NA         | 1279.789   |
| $LMiller\_00469.Swab$ | $H.pylori_(+)$    | Swab | $H.pylori_(+)_Swab$                      | Female | 6.38         | NA         | 1279.789   |
| $LMiller\_00473.BAL$  | H.pylori_(-)      | BAL  | H.pylori_(-)_BAL                         | Female | 0.00         | 20.100     | 608.508    |
| $LMiller\_00473.Swab$ | H.pylori_(-)      | Swab | H.pylori_(-)_Swab                        | Female | 0.00         | 20.100     | 608.508    |
| $LMiller\_00474.BAL$  | $H.pylori_(-)$    | BAL  | H.pylori_(-)_BAL                         | Female | 0.00         | 44.699     | 443.989    |
| $LMiller\_00476.BAL$  | H.pylori_(+)      | BAL  | H.pylori_(+)_BAL                         | Female | 5.48         | NA         | 476.431    |
| LMiller 00476.Swab    | $H.pylori_(+)$    | Swab | H.pylori_(+)_Swab                        | Female | 5.48         | NA         | 476.431    |
| $LMiller\_00477.BAL$  | $H.pylori_(+)$    | BAL  | H.pylori_(+)_BAL                         | Female | 5.81         | NA         | 715.056    |
| LMiller_00477.Swab    | $H.pylori_(+)$    | Swab | H.pylori_(+)_Swab                        | Female | 5.81         | NA         | 715.056    |
| $LMiller\_00481.BAL$  | H.pylori_(-)      | BAL  | H.pylori_(-)_BAL                         | Female | 0.00         | 17.622     | NA         |
| $LMiller\_00481.Swab$ | H.pylori_(-)      | Swab | ${\rm H.pylori}\_(\text{-})\_{\rm Swab}$ | Female | 0.00         | 17.622     | NA         |

# Odds ratios of variables based on H. pylori status



|                      | OR    | 2.5~% | 97.5~%   | p     |
|----------------------|-------|-------|----------|-------|
| (Intercept)          | 0.018 | 0.000 | 110.146  | 0.410 |
| $infant\_weight\_kg$ | 0.243 | 0.000 | 7254.525 | 0.786 |
| $age\_days\_infants$ | 1.042 | 0.955 | 1.177    | 0.413 |
| $il\_bal\_pg.ml$     | 0.993 | 0.852 | 1.158    | 0.921 |
| $il\_plasma\_pg.ml$  | 0.996 | 0.985 | 1.001    | 0.271 |

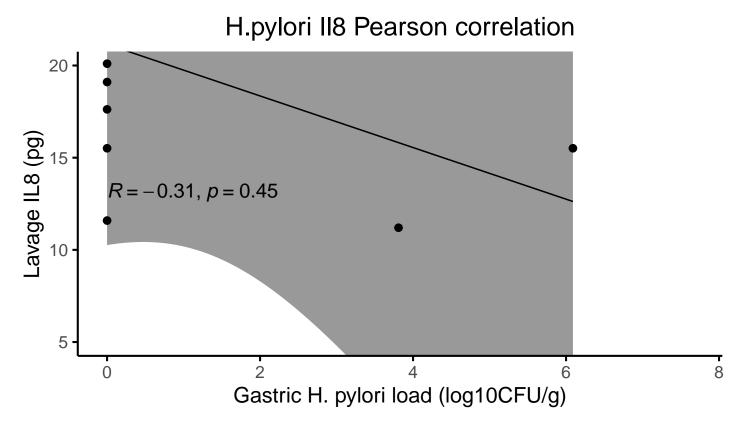


Figure 1: This plot only includes animals that had material sequenced and not all 25 animals from the study

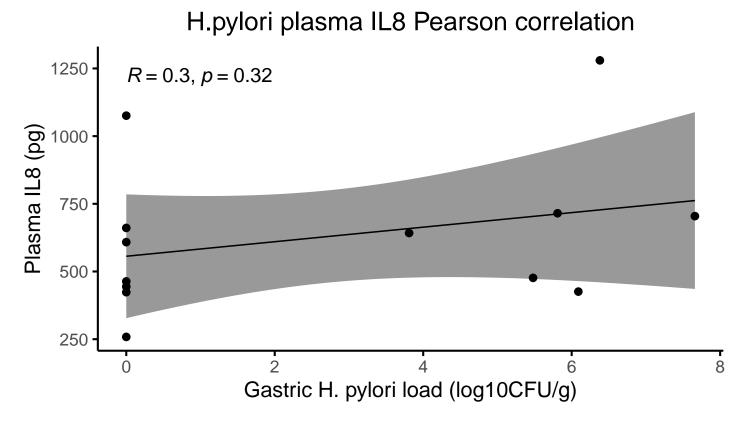
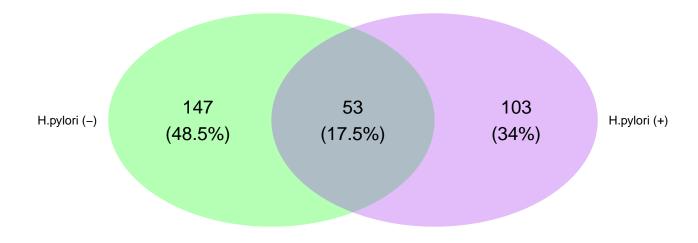
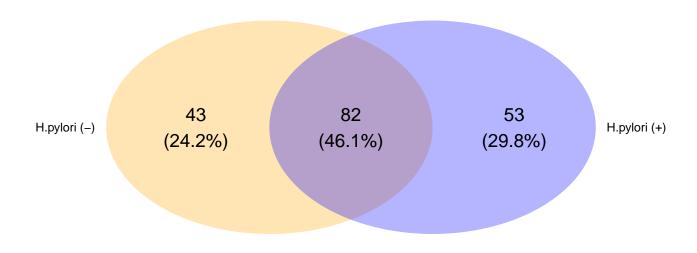


Figure 2: This plot only includes animals that had material sequenced and not all 25 animals from the study

# Overlapping taxa between H. pylori (+) and (-) in lavage

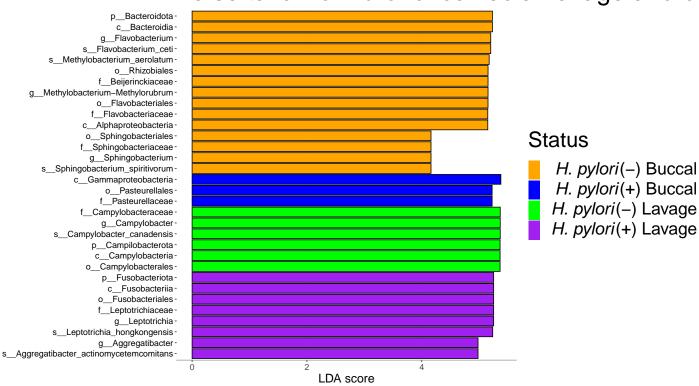


### Overlapping taxa between H. pylori (+) and (-) in buccal cavity



Lefse analysis and differential abundance for both sites

## Lefse taxa from bronchoalveolar lavage and or



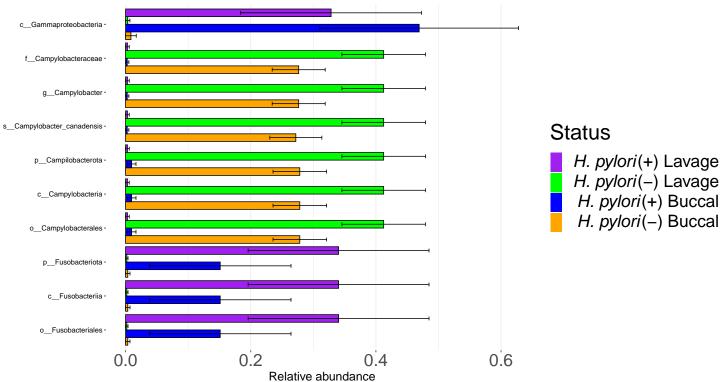
g\_\_Campylobacter-

-5.0

-25

s\_\_Campylobacter\_canadensis

### Lefse relative abundance from bronchoalveolar lavage



Lefse and differential abundance for bronchoalveolar lavage

#### Lefse Taxa from Bronchoalveolar Lavage p\_\_Proteobacteria c\_\_Alphaproteobacteria o\_\_Rhizobiales f\_\_Beijerinckiaceae g\_\_Methylobacterium-Methylorubrum s\_\_Methylobacterium\_aerolatum - Flavobacteriales f Flavobacteriaceae g\_\_Flavobacterium s\_\_Flavobacterium\_ceti-**Status** c\_\_Bacteroidia p\_\_Bacteroidota o\_\_Bacteroidales -H. pylori(-) Lavage o\_\_Sphingobacteriales -H. pylori(+) Lavage f\_Sphingobacteriaceae g\_\_Sphingobacterium s\_\_Sphingobacterium\_spiritivorumc\_\_Clostridia p\_\_Firmicutes p\_Campilobacterota c\_\_Campylobacteria o\_\_Campylobacterales f\_Campylobacteraceae -

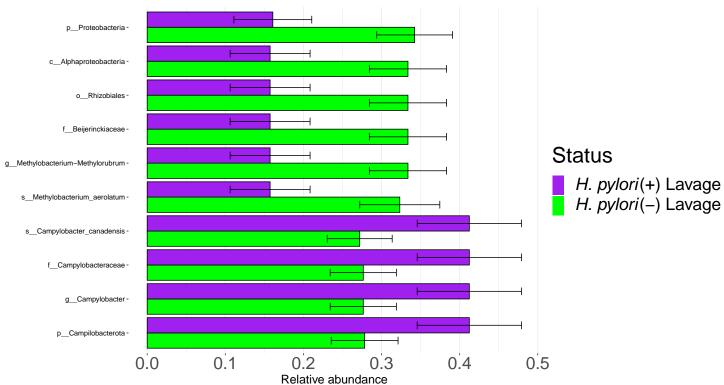
0.0

LDA score

2.5

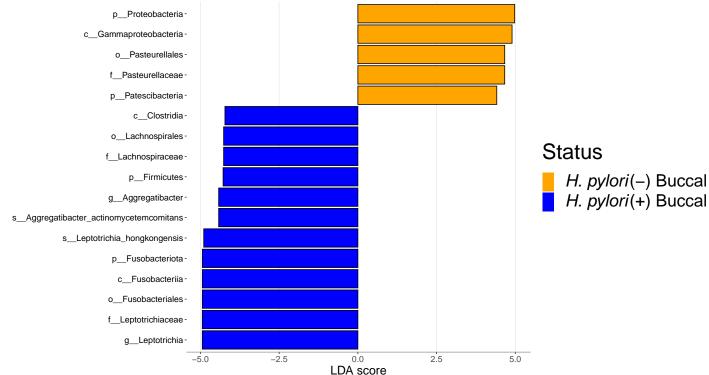
5.0



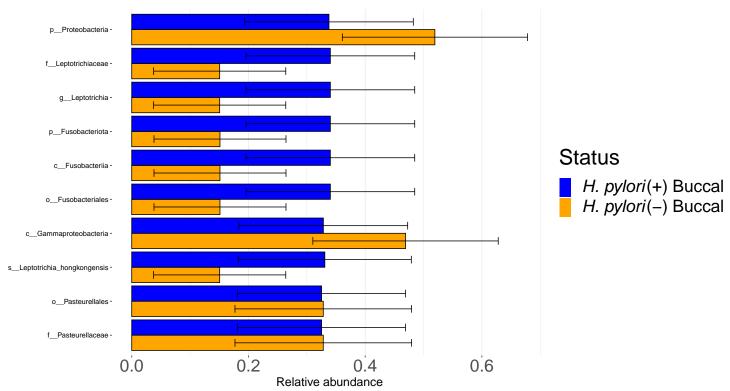


Lefse and differential abundance for oral swabs

# Lefse taxa from oral swabs



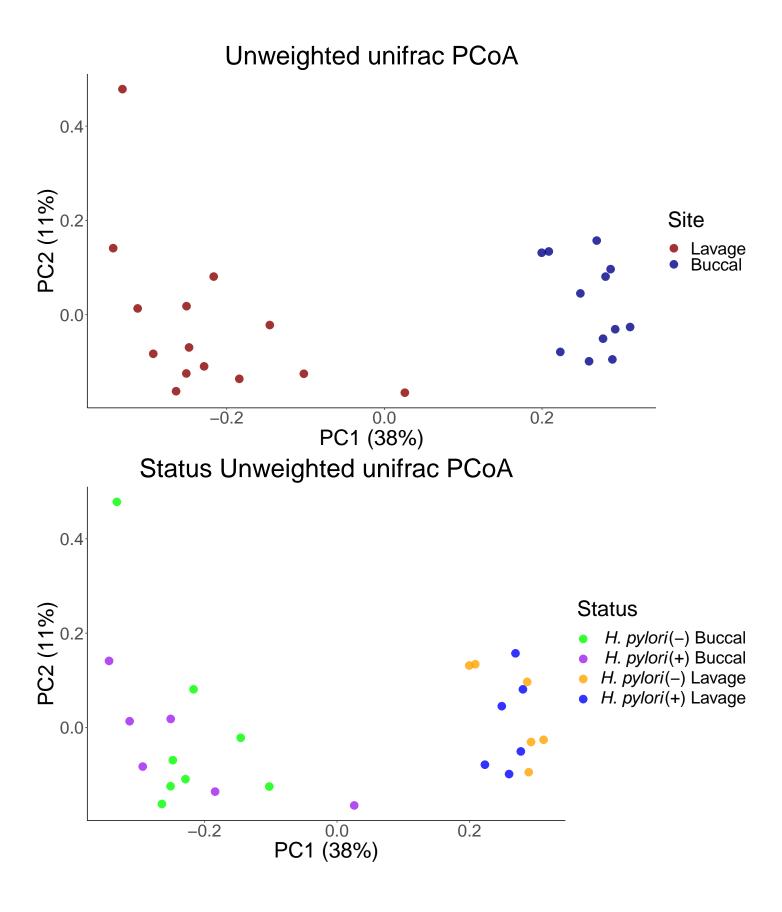
### Lefse relative abundance from oral swabs

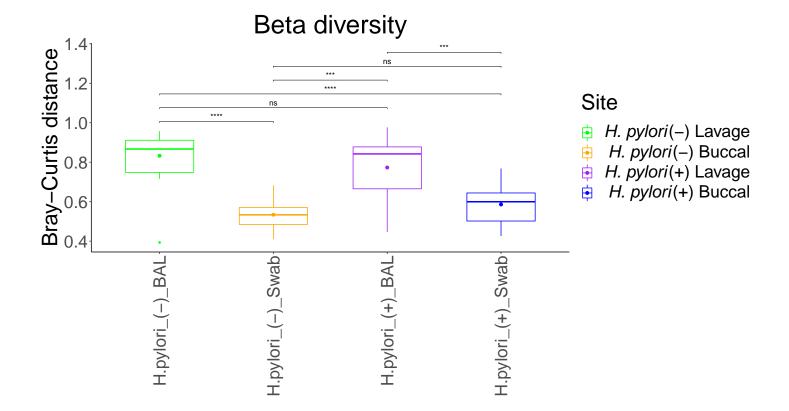


#### Beta diversity

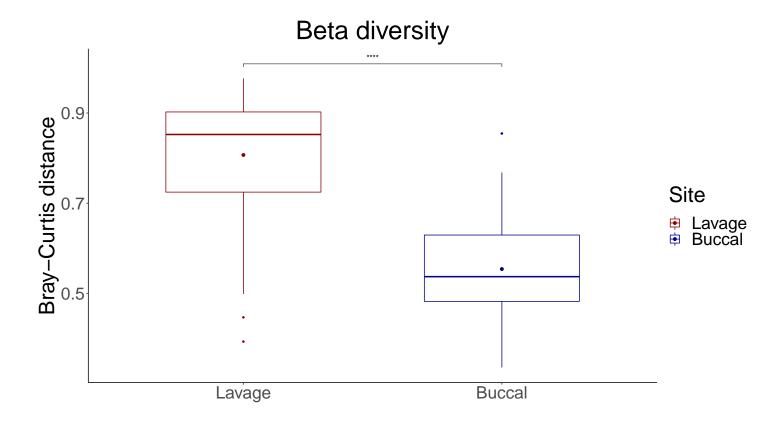
#### Unweighted unifrac PCoA plots

```
PC1
                   PC2
                              PC3
                                         PC4
                                                     PC5
                                                                PC6
                                                                           PC7
## 1 0.38311 0.1122317 0.08255574 0.05624649 0.04518453 0.04178459 0.03697168
            PC8
                      PC9
                               PC10
                                          PC11
                                                     PC12
                                                                PC13
## 1 0.03441675 0.0305229 0.0292508 0.02395872 0.0227312 0.01881898 0.01538496
          PC15
                     PC16
                                PC17
                                            PC18
                                                         PC19
                                                                     PC20
##
## 1 0.0140825 0.01226748 0.01155323 0.008776171 0.007488221 0.004824027
                        PC22
                                    PC23 PC24 PC25 PC26
##
            PC21
## 1 0.003528799 0.002630896 0.001679594
                                             0
```



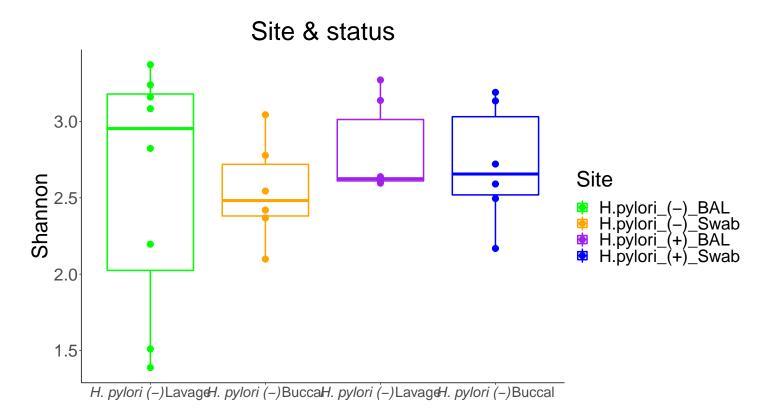


#### **Bray-Curtis**



### Alpha Diversity

Shannon index by site and H. pylori status



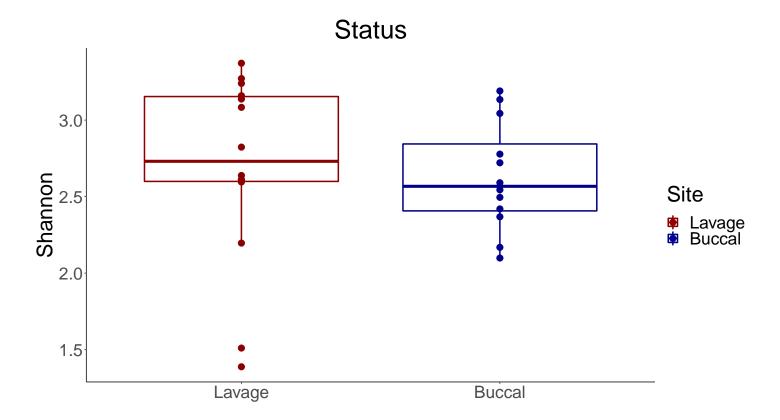
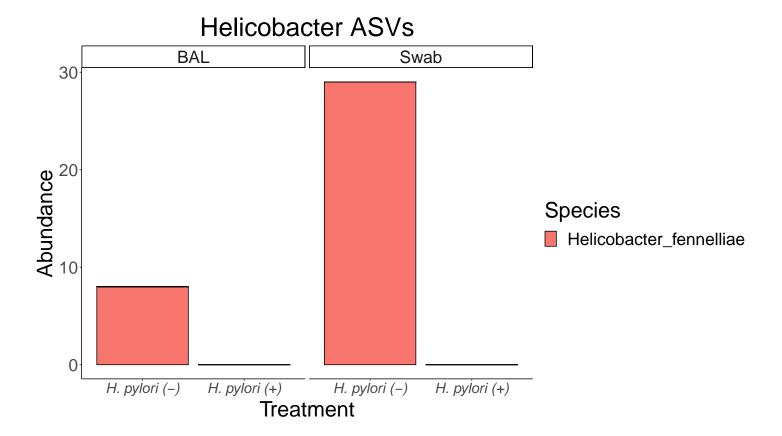


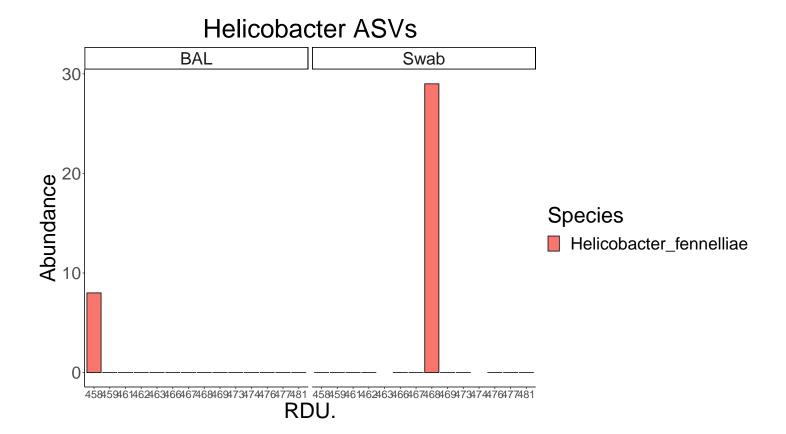
Table 2: All alpha diversity measurees

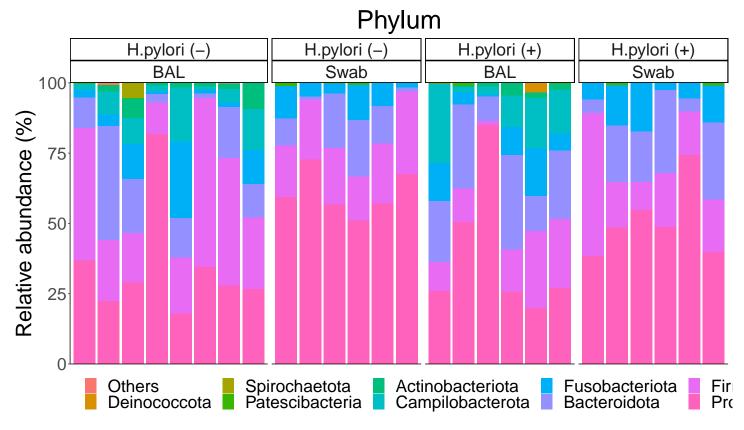
| Groups  | Measure  | Test method          | p.value                                   | Significance         |
|---|--|----------------------|---|----------------------|
| H.pylori_(-)_BAL vs H.pylori_(-)_Swab H.pylori_(-)_BAL vs H.pylori_(+)_BAL H.pylori_(-)_BAL vs H.pylori_(+)_Swab H.pylori_(-)_Swab vs H.pylori_(+)_BAL H.pylori_(-)_Swab vs H.pylori_(+)_Swab   | Observed<br>Observed<br>Observed<br>Observed                       | KW<br>KW<br>KW<br>KW | 0.220<br>0.651<br>0.081<br>0.262<br>0.520 |                      |
| H.pylori_(+)_BAL vs H.pylori_(+)_Swab<br>H.pylori_(-)_BAL vs H.pylori_(-)_Swab vs H.pylori_(+)_BAL vs H.pylori_(+)_Swab<br>H.pylori_(-)_BAL vs H.pylori_(-)_Swab<br>H.pylori_(-)_BAL vs H.pylori_(+)_BAL<br>H.pylori_(-)_BAL vs H.pylori_(+)_Swab   | Observed<br>Observed<br>Chao1<br>Chao1<br>Chao1                    | KW<br>KW<br>KW<br>KW | 0.109<br>0.202<br>0.220<br>0.651<br>0.081 |                      |
| H.pylori_(-)_Swab vs H.pylori_(+)_BAL<br>H.pylori_(-)_Swab vs H.pylori_(+)_Swab<br>H.pylori_(+)_BAL vs H.pylori_(+)_Swab<br>H.pylori_(-)_BAL vs H.pylori_(-)_Swab vs H.pylori_(+)_BAL vs H.pylori_(+)_Swab<br>H.pylori_(-)_BAL vs H.pylori_(-)_Swab | Chao1<br>Chao1<br>Chao1<br>Chao1<br>ACE                            | KW<br>KW<br>KW<br>KW | 0.262<br>0.520<br>0.109<br>0.202<br>0.220 |                      |
| H.pylori_(-)_BAL vs H.pylori_(+)_BAL H.pylori_(-)_BAL vs H.pylori_(+)_Swab H.pylori_(-)_Swab vs H.pylori_(+)_BAL H.pylori_(-)_Swab vs H.pylori_(+)_Swab H.pylori_(+)_BAL vs H.pylori_(+)_Swab   | ACE<br>ACE<br>ACE<br>ACE   | KW<br>KW<br>KW<br>KW | 0.651<br>0.081<br>0.262<br>0.520<br>0.109 |                      |
| H.pylori_(-)_BAL vs H.pylori_(-)_Swab vs H.pylori_(+)_BAL vs H.pylori_(+)_Swab H.pylori_(-)_BAL vs H.pylori_(-)_Swab H.pylori_(-)_BAL vs H.pylori_(+)_BAL H.pylori_(-)_BAL vs H.pylori_(+)_Swab H.pylori_(-)_Swab vs H.pylori_(+)_BAL               | ACE<br>Shannon<br>Shannon<br>Shannon<br>Shannon                    | KW<br>KW<br>KW<br>KW | 0.202<br>0.439<br>1.000<br>0.796<br>0.109 |                      |
| H.pylori_(-)_Swab vs H.pylori_(+)_Swab<br>H.pylori_(+)_BAL vs H.pylori_(+)_Swab<br>H.pylori_(-)_BAL vs H.pylori_(-)_Swab vs H.pylori_(+)_BAL vs H.pylori_(+)_Swab<br>H.pylori_(-)_BAL vs H.pylori_(-)_Swab<br>H.pylori_(-)_BAL vs H.pylori_(+)_BAL  | Shannon<br>Shannon<br>Shannon<br>Simpson<br>Simpson                | KW<br>KW<br>KW<br>KW | 0.337<br>0.423<br>0.540<br>0.519<br>0.699 |                      |
| H.pylori_(-)_BAL vs H.pylori_(+)_Swab<br>H.pylori_(-)_Swab vs H.pylori_(+)_BAL<br>H.pylori_(-)_Swab vs H.pylori_(+)_Swab<br>H.pylori_(+)_BAL vs H.pylori_(+)_Swab<br>H.pylori_(-)_BAL vs H.pylori_(-)_Swab vs H.pylori_(+)_BAL vs H.pylori_(+)_Swab | Simpson<br>Simpson<br>Simpson<br>Simpson                           | KW<br>KW<br>KW<br>KW | 0.699<br>0.109<br>0.423<br>0.262<br>0.493 |                      |
| H.pylori_(-)_BAL vs H.pylori_(-)_Swab H.pylori_(-)_BAL vs H.pylori_(+)_BAL H.pylori_(-)_BAL vs H.pylori_(+)_Swab H.pylori_(-)_Swab vs H.pylori_(+)_BAL H.pylori_(-)_Swab vs H.pylori_(+)_Swab   | InvSimpson<br>InvSimpson<br>InvSimpson<br>InvSimpson<br>InvSimpson | KW<br>KW<br>KW<br>KW | 0.519<br>0.699<br>0.699<br>0.109<br>0.423 |                      |
| H.pylori_(+)_BAL vs H.pylori_(+)_Swab<br>H.pylori_(-)_BAL vs H.pylori_(-)_Swab vs H.pylori_(+)_BAL vs H.pylori_(+)_Swab<br>H.pylori_(-)_BAL vs H.pylori_(-)_Swab<br>H.pylori_(-)_BAL vs H.pylori_(+)_BAL<br>H.pylori_(-)_BAL vs H.pylori_(+)_Swab   | InvSimpson<br>InvSimpson<br>Fisher<br>Fisher<br>Fisher             | KW<br>KW<br>KW<br>KW | 0.262<br>0.493<br>0.606<br>0.699<br>0.796 |                      |
| H.pylori_(-)_Swab vs H.pylori_(+)_BAL<br>H.pylori_(-)_Swab vs H.pylori_(+)_Swab<br>H.pylori_(+)_BAL vs H.pylori_(+)_Swab<br>H.pylori_(-)_BAL vs H.pylori_(-)_Swab vs H.pylori_(+)_BAL vs H.pylori_(+)_Swab<br>H.pylori_(-)_BAL vs H.pylori_(-)_Swab | Fisher<br>Fisher<br>Fisher<br>Coverage                             | KW<br>KW<br>KW<br>KW | 0.631<br>0.631<br>1.000<br>0.927<br>NaN   | NA                   |
| H.pylori_(-)_BAL vs H.pylori_(+)_BAL H.pylori_(-)_BAL vs H.pylori_(+)_Swab H.pylori_(-)_Swab vs H.pylori_(+)_BAL H.pylori_(-)_Swab vs H.pylori_(+)_Swab H.pylori_(+)_BAL vs H.pylori_(+)_Swab   | Coverage<br>Coverage<br>Coverage<br>Coverage                       | KW<br>KW<br>KW<br>KW | NaN<br>NaN<br>NaN<br>NaN<br>NaN           | NA<br>NA<br>NA<br>NA |
| H.pylori_(-)_BAL vs H.pylori_(-)_Swab vs H.pylori_(+)_BAL vs H.pylori_(+)_Swab H.pylori_(-)_BAL vs H.pylori_(-)_Swab H.pylori_(-)_BAL vs H.pylori_(+)_BAL H.pylori_(-)_BAL vs H.pylori_(+)_Swab H.pylori_(-)_Swab vs H.pylori_(+)_BAL               | Coverage<br>PD<br>PD<br>PD<br>PD                                   | KW<br>KW<br>KW<br>KW | NaN<br>0.366<br>0.699<br>0.439<br>0.631   | NA                   |
| H.pylori_(-)_Swab vs H.pylori_(+)_Swab<br>H.pylori_(+)_BAL vs H.pylori_(+)_Swab<br>H.pylori_(-)_BAL vs H.pylori_(-)_Swab vs H.pylori_(+)_BAL vs H.pylori_(+)_Swab   | PD<br>PD<br>PD   | KW<br>KW<br>KW       | 0.749 $0.631$ $0.777$                     |                      |

### Helicobacter positive respiratory samples

Helicobacter positive samples separated by site







 ${\bf Figure~3:~Microbiota~Composition~at~Phylum~level}.$ 

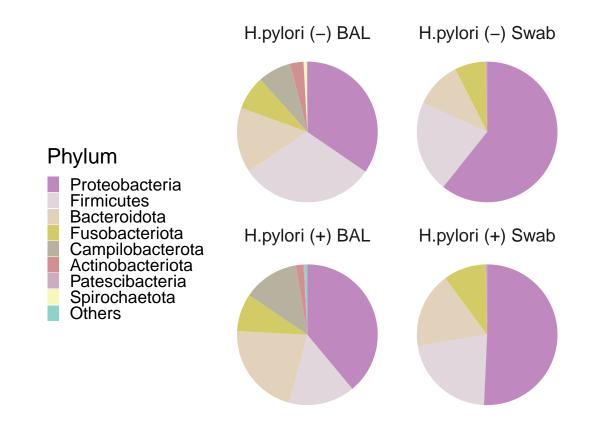


Figure 4: Microbiota Composition at Phylum level.

 Table 3: Phylum Average Relative Abundance

| Taxonomy  | Sample  | Average Percent Abundance             |
|---|---|---------------------------------------|
| Acidobacteriota<br>Actinobacteriota<br>Actinobacteriota<br>Actinobacteriota<br>Actinobacteriota | H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (+) BAL<br>H.pylori (+) Swab<br>H.pylori (-) Swab  | 0.1%<br>3.1%<br>1.8%<br>0.1%<br>0%    |
| Bacteroidota Bacteroidota Bacteroidota Bacteroidota Campilobacterota                            | H.pylori (+) BAL<br>H.pylori (+) Swab<br>H.pylori (-) BAL<br>H.pylori (-) Swab<br>H.pylori (+) BAL  | 21.7% $17.4%$ $14.9%$ $10.8%$ $12.9%$ |
| Campilobacterota<br>Campilobacterota<br>Deinococcota<br>Desulfobacterota<br>Firmicutes          | H.pylori (-) BAL<br>H.pylori (-) Swab<br>H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (-) BAL   | 7.7% $0%$ $0.6%$ $0.1%$ $31.1%$       |
| Firmicutes Firmicutes Firmicutes Fusobacteriota Fusobacteriota                                  | H.pylori (+) Swab<br>H.pylori (-) Swab<br>H.pylori (+) BAL<br>H.pylori (+) Swab<br>H.pylori (+) BAL | 21.7% $21%$ $15.2%$ $9.8%$ $8.6%$     |
| Fusobacteriota<br>Fusobacteriota<br>Latescibacterota<br>Patescibacteria<br>Patescibacteria      | H.pylori (-) BAL<br>H.pylori (-) Swab<br>H.pylori (-) BAL<br>H.pylori (-) Swab<br>H.pylori (+) Swab | 7.8% $7%$ $0%$ $0.4%$ $0.3%$          |
| Patescibacteria<br>Patescibacteria<br>Proteobacteria<br>Proteobacteria<br>Proteobacteria        | H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (-) Swab<br>H.pylori (+) Swab<br>H.pylori (+) BAL  | 0.3% $0%$ $60.7%$ $50.7%$ $38.9%$     |
| Proteobacteria<br>Spirochaetota   | H.pylori (-) BAL<br>H.pylori (-) BAL  | $34.5\% \ 0.7\%$                      |

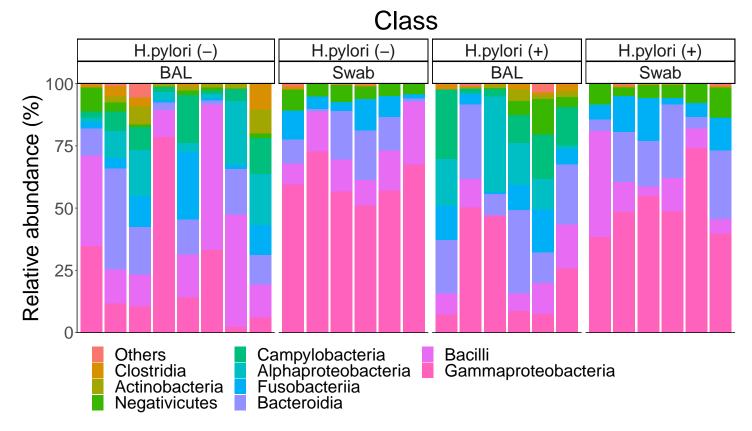


Figure 5: Microbiota Composition at Class level.

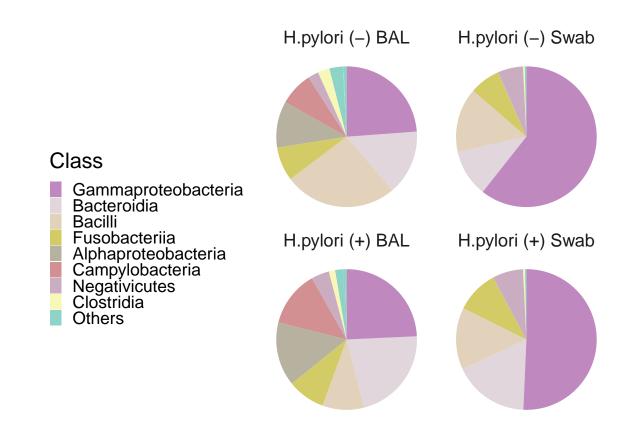


Figure 6: Microbiota Composition at Class level.

 Table 4: Class Average Relative Abundance

| Taxonomy  | Sample  | Average Percent Abundance             |
|---|---|---------------------------------------|
| Actinobacteria<br>Actinobacteria<br>Alphaproteobacteria<br>Alphaproteobacteria<br>Bacilli     | H.pylori (-) BAL<br>H.pylori (+) BAL<br>H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (-) BAL    | 3.1%<br>1.7%<br>14.7%<br>10.7%<br>26% |
| Bacilli Bacilli Bacilli Bacteroidia Bacteroidia   | H.pylori (-) Swab<br>H.pylori (+) Swab<br>H.pylori (+) BAL<br>H.pylori (+) BAL<br>H.pylori (+) Swab | 14.9% $14.2%$ $9.6%$ $21.7%$ $17.4%$  |
| Bacteroidia Bacteroidia Campylobacteria Campylobacteria Clostridia                            | H.pylori (-) BAL<br>H.pylori (-) Swab<br>H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (-) BAL   | 14.9% $10.8%$ $12.9%$ $7.7%$ $2.7%$   |
| Clostridia<br>Clostridia<br>Clostridia<br>Deinococci<br>Fusobacteriia                         | H.pylori (+) BAL<br>H.pylori (+) Swab<br>H.pylori (-) Swab<br>H.pylori (+) BAL<br>H.pylori (+) Swab | 1.5% $0.4%$ $0.4%$ $0.6%$ $9.8%$      |
| Fusobacteriia<br>Fusobacteriia<br>Fusobacteriia<br>Gammaproteobacteria<br>Gammaproteobacteria | H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (-) Swab<br>H.pylori (-) Swab<br>H.pylori (+) Swab | 8.6% $7.8%$ $7%$ $60.7%$ $50.7%$      |
| Gammaproteobacteria<br>Gammaproteobacteria<br>Negativicutes<br>Negativicutes<br>Negativicutes | H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (+) Swab<br>H.pylori (-) Swab<br>H.pylori (+) BAL  | 24.3% $23.8%$ $7.1%$ $5.8%$ $4.1%$    |
| Negativicutes<br>Spirochaetia   | H.pylori (-) BAL<br>H.pylori (-) BAL  | $2.4\% \\ 0.7\%$                      |

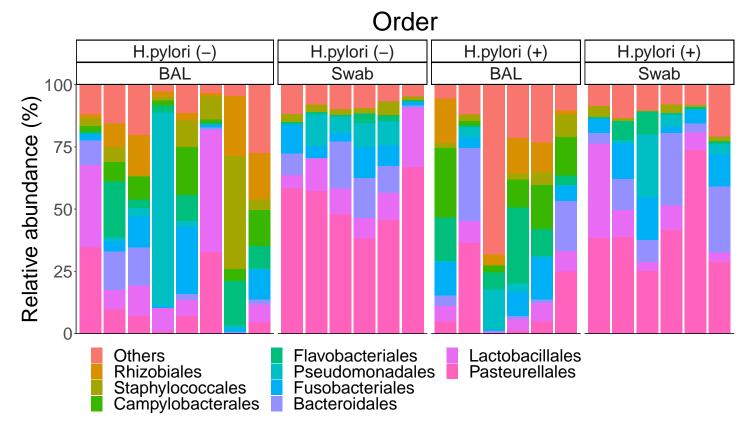


Figure 7: Microbiota Composition at Order level.

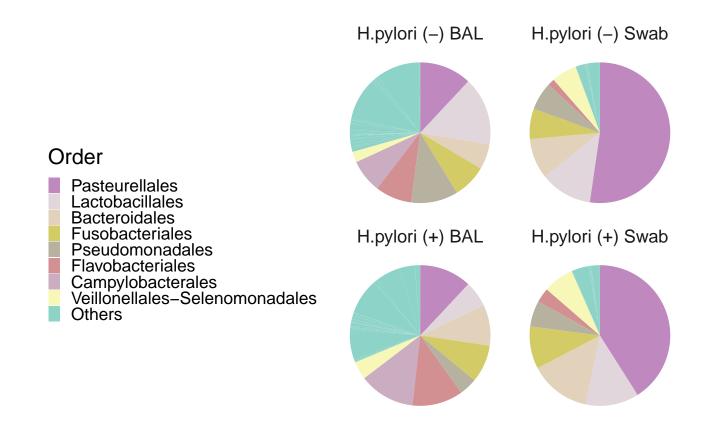


Figure 8: Microbiota Composition at Order level.

 Table 5: Order Average Relative Abundance

| Taxonomy   | Sample  | Average Percent Abundance              |
|--|---|--|
| Bacteroidales Bacteroidales Bacteroidales Bacteroidales Burkholderiales                  | H.pylori (+) Swab<br>H.pylori (+) BAL<br>H.pylori (-) Swab<br>H.pylori (-) BAL<br>H.pylori (+) BAL  | 14.1%<br>9.4%<br>9.3%<br>5.8%<br>6.8%  |
| Campylobacterales Campylobacterales Flavobacteriales Flavobacteriales Fusobacteriales    | H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (+) Swab   | 12.9%<br>7.7%<br>11.7%<br>8.4%<br>9.8% |
| Fusobacteriales Fusobacteriales Fusobacteriales Lactobacillales Lactobacillales          | H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (-) Swab<br>H.pylori (-) BAL<br>H.pylori (+) Swab  | 8.6% $7.8%$ $15.7%$ $12.2%$            |
| Lactobacillales Lactobacillales Pasteurellales Pasteurellales Pasteurellales             | H.pylori (-) Swab<br>H.pylori (+) BAL<br>H.pylori (-) Swab<br>H.pylori (+) Swab<br>H.pylori (+) BAL | 12% $6%$ $52.3%$ $41%$ $12%$           |
| Pasteurellales Pseudomonadales Pseudomonadales Pseudomonadales Pseudomonadales           | H.pylori (-) BAL<br>H.pylori (-) BAL<br>H.pylori (-) Swab<br>H.pylori (+) Swab<br>H.pylori (+) BAL  | 12% $10.8%$ $6.4%$ $5.9%$ $4.1%$       |
| Rhizobiales Rhizobiales Sphingomonadales Staphylococcales Veillonellales-Selenomonadales | H.pylori (-) BAL<br>H.pylori (+) BAL<br>H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (+) Swab   | 9.6%<br>8.3%<br>5.8%<br>10.1%<br>7.1%  |
| Veillonellales-Selenomonadales<br>Veillonellales-Selenomonadales                         | H.pylori (-) Swab<br>H.pylori (+) BAL   | 5.8%<br>4.1%                           |

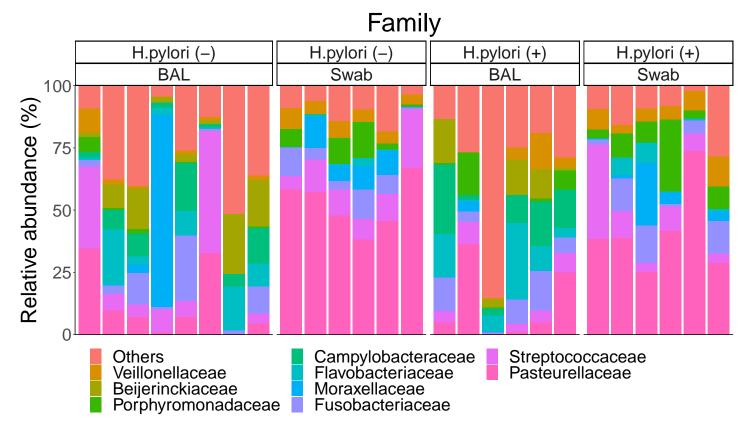


Figure 9: Microbiota Composition at Family level.

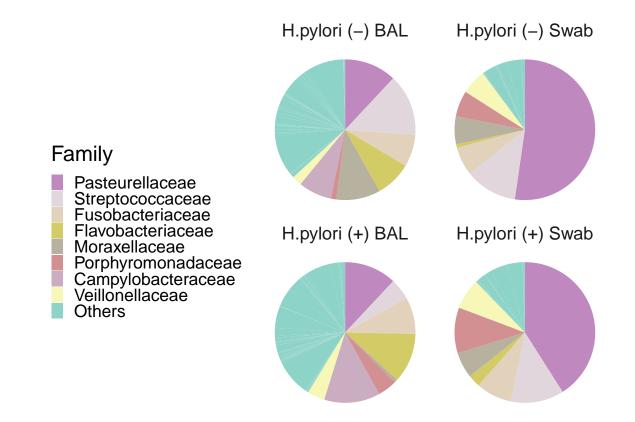


Figure 10: Microbiota Composition at Family level.

Table 6: Family Average Relative Abundance

| Taxonomy  | Sample  | Average Percent Abundance              |
|---|---|--|
| Beijerinckiaceae<br>Beijerinckiaceae<br>Campylobacteraceae<br>Campylobacteraceae<br>Flavobacteriaceae | H.pylori (-) BAL<br>H.pylori (+) BAL<br>H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (+) BAL    | 9.5%<br>8.1%<br>12.9%<br>7.7%<br>11.5% |
| Flavobacteriaceae Fusobacteriaceae Fusobacteriaceae Fusobacteriaceae Fusobacteriaceae                 | H.pylori (-) BAL<br>H.pylori (+) BAL<br>H.pylori (+) Swab<br>H.pylori (-) BAL<br>H.pylori (-) Swab  | 8.3% $8.5%$ $8.2%$ $7.4%$ $6.6%$       |
| Moraxellaceae<br>Moraxellaceae<br>Moraxellaceae<br>Oxalobacteraceae<br>Pasteurellaceae                | H.pylori (-) BAL<br>H.pylori (-) Swab<br>H.pylori (+) Swab<br>H.pylori (+) BAL<br>H.pylori (-) Swab | 10.1% $6.4%$ $5.9%$ $5%$ $52.3%$       |
| Pasteurellaceae Pasteurellaceae Pasteurellaceae Porphyromonadaceae Porphyromonadaceae                 | H.pylori (+) Swab<br>H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (+) Swab<br>H.pylori (-) Swab | 41% $12%$ $12%$ $10.5%$ $5.9%$         |
| Porphyromonadaceae<br>Prevotellaceae<br>Prevotellaceae<br>Sphingomonadaceae<br>Staphylococcaceae      | H.pylori (+) BAL<br>H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (+) BAL<br>H.pylori (-) BAL    | 4.4% $4.3%$ $4.1%$ $5.8%$ $8.4%$       |
| Streptococcaceae<br>Streptococcaceae<br>Streptococcaceae<br>Streptococcaceae<br>Veillonellaceae       | H.pylori (-) BAL<br>H.pylori (+) Swab<br>H.pylori (-) Swab<br>H.pylori (+) BAL<br>H.pylori (+) Swab | 14.2% $12.2%$ $12%$ $4.9%$ $7%$        |
| Veillonellaceae<br>Veillonellaceae  | H.pylori (-) Swab<br>H.pylori (+) BAL   | 5.8% $4%$                              |

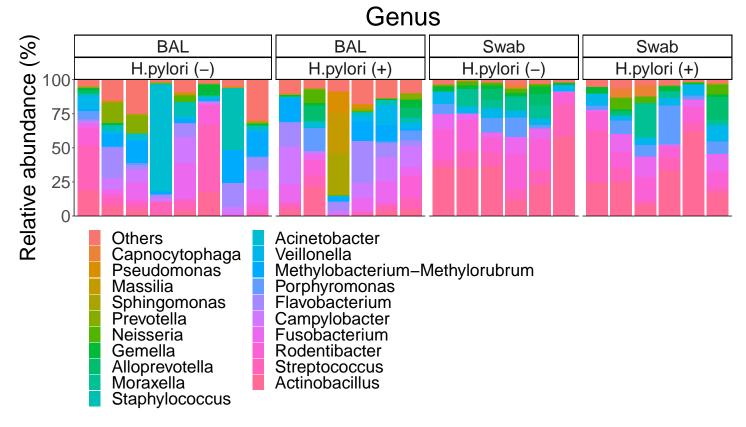


Figure 11: Microbiota Composition at Genus level.

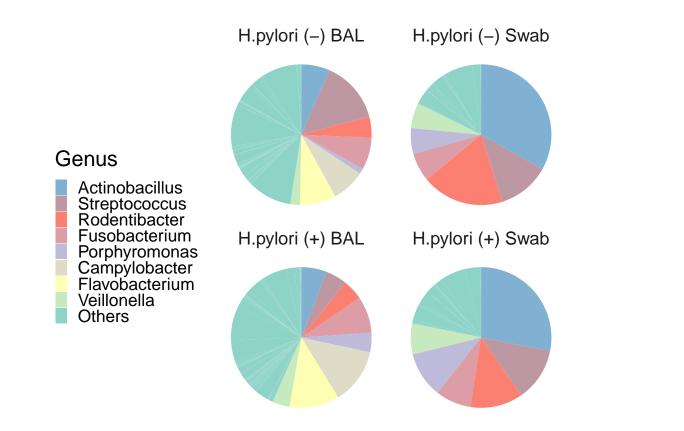


Figure 12: Microbiota Composition at Genus level.

 Table 7: Genus Average Relative Abundance

| Taxonomy   | Sample   | Average Percent Abundance               |
|--|--|---|
| Acinetobacter Actinobacillus Actinobacillus Actinobacillus Actinobacillus                  | H.pylori (-) BAL<br>H.pylori (-) Swab<br>H.pylori (+) Swab<br>H.pylori (-) BAL<br>H.pylori (+) BAL   | 10.1%<br>33.1%<br>28.1%<br>6.6%<br>5.9% |
| Campylobacter Campylobacter Flavobacterium Flavobacterium Fusobacterium                    | H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (+) BAL<br>H.pylori (-) BAL<br>H.pylori (+) BAL     | 12.9%<br>7.7%<br>11.5%<br>8.3%<br>8.5%  |
| Fusobacterium Fusobacterium Fusobacterium Massilia Methylobacterium-Methylorubrum          | H.pylori (+) Swab<br>H.pylori (-) BAL<br>H.pylori (-) Swab<br>H.pylori (+) BAL<br>H.pylori (-) BAL   | 8.2%<br>7.4%<br>6.6%<br>4.9%<br>9.5%    |
| Methylobacterium-Methylorubrum<br>Moraxella<br>Moraxella<br>Porphyromonas<br>Porphyromonas | H.pylori (+) BAL<br>H.pylori (-) Swab<br>H.pylori (+) Swab<br>H.pylori (+) Swab<br>H.pylori (-) Swab | 8.1%<br>6.4%<br>5.9%<br>10.5%<br>5.9%   |
| Rodentibacter Rodentibacter Rodentibacter Rodentibacter Sphingomonas                       | H.pylori (-) Swab<br>H.pylori (+) Swab<br>H.pylori (-) BAL<br>H.pylori (+) BAL<br>H.pylori (+) BAL   | 18.9%<br>12.1%<br>4.9%<br>4.7%<br>5.4%  |
| Staphylococcus<br>Streptococcus<br>Streptococcus<br>Streptococcus<br>Streptococcus         | H.pylori (-) BAL<br>H.pylori (-) BAL<br>H.pylori (+) Swab<br>H.pylori (-) Swab<br>H.pylori (+) BAL   | 8.4%<br>14.2%<br>12.2%<br>12%<br>4.9%   |
| Veillonella<br>Veillonella   | H.pylori (+) Swab<br>H.pylori (-) Swab   | 7%<br>5.8%                              |

### Lavage Genus Abundance

Grouped by H. pylori status

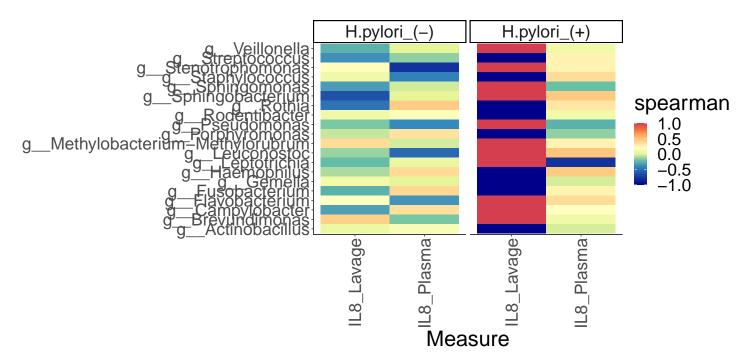


Figure 13: IL8 and lavage genus abundance Spearman correlations separated by H. pylori status

## Lavage Genus Abundance

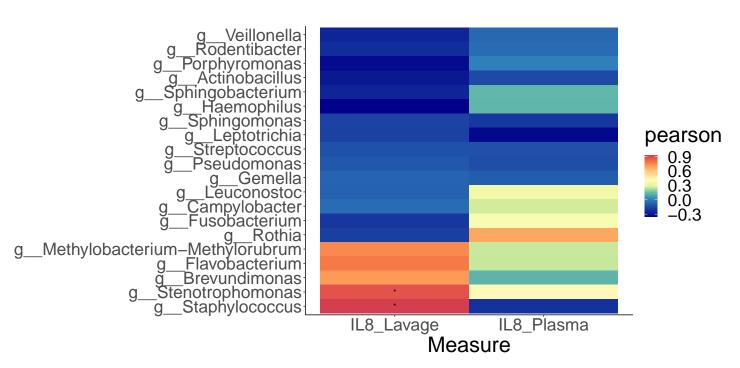


Figure 14: IL8 and lavage genus abundance Spearman correlations independent of H. pylori status

# Lavage Alpha Diversity

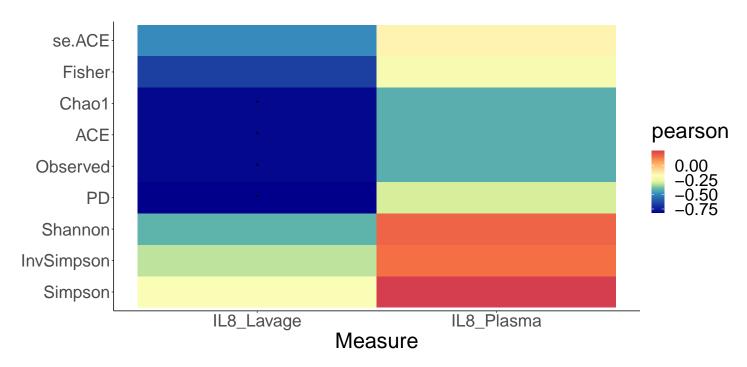


Figure 15: IL8 and lavage alpha diversity Spearman correlations inpendent of H. pylori status.

## **Buccal Cavity Genus Abundance**

Grouped by H. pylori status

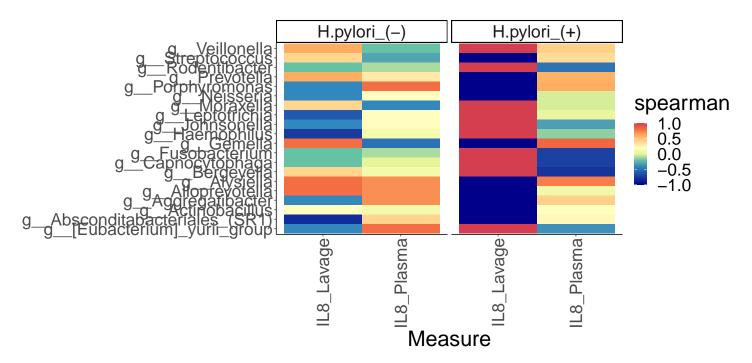


Figure 16: IL8 and Buccal genus abundance Spearman correlations separated by H. pylori status

## **Buccal Cavity Genus Abundance**

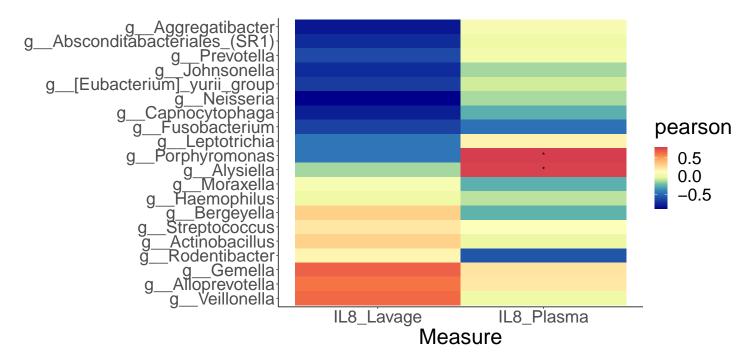


Figure 17: IL8 and Buccal genus abundance Spearman correlations independent of H. pylori status

# **Buccal Cavity Alpha Diversity**

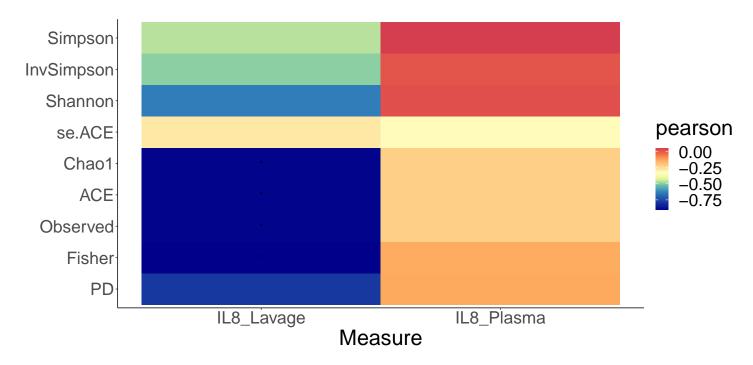


Figure 18: IL8 and Buccal alpha diversity Spearman correlations independent of H. pylori status

#### Record session information

```
## R version 4.1.3 (2022-03-10)
## Platform: x86_64-conda-linux-gnu (64-bit)
## Running under: Ubuntu 18.04.6 LTS
##
## Matrix products: default
## BLAS/LAPACK: /srv/conda/envs/notebook/lib/libopenblasp-r0.3.20.so
##
## locale:
##
   [1] LC_CTYPE=en_US.UTF-8
                                    LC_NUMERIC=C
##
   [3] LC_TIME=en_US.UTF-8
                                   LC_COLLATE=en_US.UTF-8
   [5] LC_MONETARY=en_US.UTF-8
                                    LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=en_US.UTF-8
                                   LC_NAME=C
   [9] LC ADDRESS=C
                                   LC TELEPHONE=C
##
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] stats4
                 stats
                           graphics grDevices utils
                                                          datasets methods
## [8] base
##
## other attached packages:
## [1] rmarkdown_2.14
                                     questionr_0.7.7
## [3] RColorBrewer_1.1-3
                                     tidytree_0.3.9
## [5] microeco_0.3.2
                                     ggpubr_0.4.0
##
   [7] magrittr_2.0.3
                                     kableExtra_1.3.4
## [9] tidyMicro_1.48
                                     forcats_0.5.1
## [11] stringr_1.4.0
                                     dplyr_1.0.9
## [13] purrr_0.3.4
                                     readr_2.1.2
## [15] tidyr_1.2.0
                                     tibble_3.1.7
                                     qiime2R_0.99.6
## [17] tidyverse_1.3.1
## [19] DESeq2 1.34.0
                                     SummarizedExperiment 1.24.0
## [21] Biobase_2.54.0
                                     MatrixGenerics_1.6.0
## [23] matrixStats 0.62.0
                                     GenomicRanges 1.46.1
## [25] GenomeInfoDb_1.30.0
                                     IRanges_2.28.0
## [27] S4Vectors 0.32.3
                                     BiocGenerics 0.40.0
## [29] microbiome_1.16.0
                                     ggplot2_3.3.6
## [31] phyloseq_1.38.0
                                     readxl 1.4.0
## [33] sjPlot_2.8.10
##
## loaded via a namespace (and not attached):
##
     [1] estimability_1.3
                                bit64_4.0.5
                                                        knitr_1.39
##
     [4] DelayedArray_0.20.0
                                 data.table_1.14.2
                                                        rpart_4.1.16
##
    [7] KEGGREST_1.34.0
                                RCurl_1.98-1.7
                                                        generics_0.1.2
##
   [10] timeSeries_3062.100
                                RSQLite_2.2.8
                                                        VGAM_1.1-7
##
   [13] bit_4.0.4
                                tzdb_0.3.0
                                                        webshot_0.5.3
##
   [16] xml2_1.3.3
                                 lubridate_1.8.0
                                                        httpuv_1.6.5
                                                        fBasics_3042.89.1
##
   [19] assertthat_0.2.1
                                xfun_0.31
##
    [22] hms 1.1.1
                                 evaluate 0.15
                                                        promises 1.2.0.1
##
   [25] fansi_1.0.3
                                dbplyr_2.2.0
                                                        igraph_1.3.1
##
   [28] DBI 1.1.2
                                geneplotter_1.72.0
                                                        htmlwidgets 1.5.4
   [31] ellipsis_0.3.2
                                backports_1.4.1
                                                        insight_0.17.1
##
##
   [34] permute 0.9-7
                                picante_1.8.2
                                                        annotate_1.72.0
##
   [37] deldir_1.0-6
                                vctrs_0.4.1
                                                        sjlabelled_1.2.0
##
   [40] abind_1.4-5
                                 cachem_1.0.6
                                                        withr_2.5.0
    [43] checkmate_2.1.0
                                 emmeans_1.7.4-1
##
                                                        vegan_2.6-2
##
    [46] svglite_2.1.0
                                 cluster_2.1.3
                                                        ape_5.6-2
##
   [49] lazyeval_0.2.2
                                 crayon_1.5.1
                                                        genefilter_1.76.0
##
   [52] pkgconfig_2.0.3
                                 {\tt zCompositions\_1.4.0-1 \ labeling\_0.4.2}
##
    [55] nlme_3.1-157
                                nnet_7.3-17
                                                        rlang_1.0.2
```

| ## |       | spatial_7.3-15     | lifecycle_1.0.1      | miniUI_0.1.1.1                    |
|----|-------|--------------------|----------------------|-----------------------------------|
| ## | [61]  | modelr_0.1.8       | randomForest_4.7-1.1 | cellranger_1.1.0                  |
| ## | [64]  | datawizard_0.4.1   | Matrix_1.4-1         | carData_3.0-5                     |
| ## | [67]  | Rhdf5lib_1.16.0    | boot_1.3-28          | reprex_2.0.1                      |
| ## | [70]  | base64enc_0.1-3    | png_0.1-7            | viridisLite_0.4.0                 |
| ## | [73]  | stabledist_0.7-1   | parameters_0.18.1    | bitops_1.0-7                      |
| ## | [76]  | rhdf5filters_1.6.0 | Biostrings_2.62.0    | blob_1.2.3                        |
| ## | [79]  | GUniFrac_1.6       | jpeg_0.1-9           | rstatix_0.7.0                     |
| ## | [82]  | ggeffects_1.1.2    | ggsignif_0.6.3       | scales_1.2.0                      |
| ## | [85]  | memoise_2.0.1      | plyr_1.8.7           | zlibbioc_1.40.0                   |
| ## | [88]  | compiler_4.1.3     | tinytex_0.39         | clue_0.3-60                       |
| ## |       | lme4_1.1-29        | cli_3.3.0            | ade4_1.7-19                       |
| ## |       | XVector_0.34.0     | htmlTable_2.4.1      | Formula_1.2-4                     |
| ## | [97]  | MASS_7.3-57        | mgcv_1.8-40          | tidyselect_1.1.2                  |
| ## | [100] | stringi_1.7.6      | highr_0.9            | yaml_2.3.5                        |
| ## | [103] | locfit_1.5-9.5     | latticeExtra_0.6-30  | ggrepel_0.9.1                     |
| ## |       | grid_4.1.3         | tools_4.1.3          | parallel_4.1.3                    |
|    |       | rstudioapi_0.13    | foreach_1.5.2        | foreign_0.8-82                    |
| ## | [112] | statip_0.2.3       | gridExtra_2.3        | $scatterplot3d_0.3-42$            |
| ## | [115] | farver_2.1.0       | Rtsne_0.16           | stable_1.1.6                      |
| ## | [118] | digest_0.6.29      | $shiny_1.7.1$        | Rcpp_1.0.8.3                      |
| ## | [121] | car_3.0-13         | broom_0.8.0          | performance_0.9.0                 |
|    |       | later_1.2.0        | httr_1.4.3           | AnnotationDbi_1.56.1              |
|    |       | effectsize_0.7.0   | sjstats_0.18.1       | colorspace_2.0-3                  |
|    |       | rvest_1.0.2        | XML_3.99-0.10        | fs_1.5.2                          |
|    |       | modeest_2.4.0      | truncnorm_1.0-8      | splines_4.1.3                     |
|    |       | yulab.utils_0.0.4  | rmutil_1.1.9         | $statmod_1.4.36$                  |
|    |       | multtest_2.50.0    | systemfonts_1.0.4    | xtable_1.8-4                      |
|    |       | jsonlite_1.8.0     | nloptr_2.0.3         | timeDate_3043.102                 |
|    |       | R6_2.5.1           | ${\tt Hmisc\_4.7-1}$ | NADA_1.6-1.1                      |
|    |       | pillar_1.7.0       | htmltools_0.5.2      | mime_0.12                         |
|    |       | glue_1.6.2         | $fastmap_1.1.0$      | $minqa_1.2.4$                     |
|    |       | DT_0.23            | BiocParallel_1.28.3  | codetools_0.2-18                  |
| ## | [157] | mvtnorm_1.1-3      | utf8_1.2.2           | lattice_0.20-45                   |
|    |       | interp_1.1-3       | survival_3.3-1       | $biomformat_1.22.0$               |
|    |       | munsell_0.5.0      | rhdf5_2.38.0         | <pre>GenomeInfoDbData_1.2.7</pre> |
|    |       | iterators_1.0.14   | labelled_2.9.1       | sjmisc_2.8.9                      |
|    |       | haven_2.5.0        | $reshape2_1.4.4$     | gtable_0.3.0                      |
| ## | [172] | bayestestR_0.12.1  |                      |                                   |
|    |       |                    |                      |                                   |