WLS_analysis

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Load data

Packages

```
#The vegan package provides tools for descriptive community ecology. It has most basic functions of div
#In general, this package is used for Bray-Curtis and Jaccard analyses.
library(vegan)
## Loading required package: permute
## Loading required package: lattice
## This is vegan 2.4-6
#The phyloseq package seeks to address issues with multiple microbiome analysis packages by providing a
#In general, this package is used for UniFrac analyses.
library(phyloseq)
#Graphing package used in phyloseq. To edit the default setting of a plot, you need to use functions in
library(PMCMR)
## PMCMR is superseded by PMCMRplus and will be no longer maintained. You may wish to install PMCMRplus
#Graphing package used in phyloseq. To edit the default setting of a plot, you need to use functions in
library(ggplot2)
#Figures
library(lattice)
library(gridExtra)
```

Functions

- R-krusk: Function to calculate Kruskal-Wallis for OTUs and variables identified in SIMPER analysis. From A. Steinberger
- Multiplot: for placing 2 or more ggplot2 graphs in one figure. From Cookbook for R
- Pairwise_adonis: Pairwise PERMANOVA (adonis) function to test within categorical variables with more than 2 levels. Modified from P. M. Arbizu

```
source("R_krusk.R")
source("multiplot.R")
source("Pairwise_adonis.R")
```

Data

Load environment from WLS_data_manipulation.Rmd

```
load("WLS_environment_final.rdata")
```

Gradutes-only

Beta-diversity

Tests performed on the graduate samples to avoid possible interactions between spouses and siblings. First, check if the graduate subset is different from the other sample types to ensure the subset is a good representation of the overall data set.

```
#Test if the overall microbiota differs by groups (g, p, s, e). We do not expect significance as these
#Permutational analysis of variance (PERMANOVA)
adonis(OTU ~ Link, data=Met, method="bray", permuations=1000)
##
## Call:
## adonis(formula = OTU ~ Link, data = Met, method = "bray", permuations = 1000)
##
## Permutation: free
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
              Df SumsOfSqs MeanSqs F.Model
                                                 R2 Pr(>F)
## Link
               3
                     0.728 0.24269 0.96004 0.00708 0.557
## Residuals 404
                   102.129 0.25280
                                           0.99292
## Total
             407
                   102.858
                                            1.00000
adonis(OTU ~ Link, data=Met, method="jaccard", permuations=1000)
##
## Call:
## adonis(formula = OTU ~ Link, data = Met, method = "jaccard",
                                                                      permuations = 1000)
## Permutation: free
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
##
              Df SumsOfSqs MeanSqs F.Model
                                                R2 Pr(>F)
                      0.99 0.32987 0.97191 0.00717 0.602
## Link
## Residuals 404
                    137.12 0.33940
                                           0.99283
## Total
             407
                    138.11
                                            1.00000
adonis(wUF.dist ~ Link, data=Met, permuations=1000)
##
## Call:
## adonis(formula = wUF.dist ~ Link, data = Met, permuations = 1000)
## Permutation: free
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
##
              Df SumsOfSqs MeanSqs F.Model
                                                 R2 Pr(>F)
## Link
                    0.1056 0.03521 1.0567 0.00779
## Residuals 404
                   13.4613 0.03332
                                           0.99221
```

```
## Total
             407
                   13.5669
                                            1.00000
adonis(uwUF.dist ~ Link, data=Met, permuations=1000)
##
## Call:
## adonis(formula = uwUF.dist ~ Link, data = Met, permuations = 1000)
##
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
              Df SumsOfSqs MeanSqs F.Model
                                                 R2 Pr(>F)
## Link
               3
                     0.689 0.22952 1.0697 0.00788 0.218
                    86.682 0.21456
## Residuals 404
                                            0.99212
## Total
             407
                    87.370
                                            1.00000
```

There are no differences so we move forward with the graduate subset data set. We will run a number of PERMANOVA (adonis) tests of variables of interest. All tests are run on Bray-Curtis, Jaccard, weighted and unweighted UniFrac distance measures. For variables with missing data, the data is subset to only complete.cases to avoid NA errors in adonis.

Sex and age

```
#Does sex/gender or age correlate to the gut microbiota?
adonis(BC.dist.g ~ gender+age, data = vars.g, permutations=1000)
##
## adonis(formula = BC.dist.g ~ gender + age, data = vars.g, permutations = 1000)
## Permutation: free
## Number of permutations: 1000
##
## Terms added sequentially (first to last)
##
                                                     Pr(>F)
##
              Df SumsOfSqs MeanSqs F.Model
                                                R2
                     0.983 0.98276 3.9154 0.02164 0.000999 ***
## gender
              1
                     0.264 0.26409 1.0521 0.00581 0.348651
## age
              1
## Residuals 176
                   44.176 0.25100
                                           0.97255
## Total
             178
                    45.423
                                           1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(J.dist.g ~ gender+age, data = vars.g, permutations=1000)
##
## Call:
## adonis(formula = J.dist.g ~ gender + age, data = vars.g, permutations = 1000)
## Permutation: free
## Number of permutations: 1000
##
## Terms added sequentially (first to last)
```

```
##
##
              Df SumsOfSqs MeanSqs F.Model
                                                     Pr(>F)
                                                R.2
## gender
                     0.913 0.91259 2.7046 0.01505 0.000999 ***
                     0.353 0.35336 1.0473 0.00583 0.331668
## age
               1
## Residuals 176
                    59.385 0.33742
                                           0.97913
## Total
                    60.651
                                           1.00000
             178
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(wUF.dist.g ~ gender+age, data = vars.g, permutations=1000)
##
## Call:
## adonis(formula = wUF.dist.g ~ gender + age, data = vars.g, permutations = 1000)
## Permutation: free
## Number of permutations: 1000
## Terms added sequentially (first to last)
##
##
              Df SumsOfSqs MeanSqs F.Model
                                                 R2
                                                      Pr(>F)
## gender
               1
                    0.1888 0.188788 4.5515 0.02512 0.000999 ***
                    0.0275 0.027540 0.6640 0.00366 0.861139
               1
## age
                   7.3001 0.041478
## Residuals 176
                                            0.97122
## Total
            178
                   7.5164
                                            1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(uwUF.dist.g ~ gender+age, data = vars.g, permutations=1000)
##
## Call:
## adonis(formula = uwUF.dist.g ~ gender + age, data = vars.g, permutations = 1000)
## Permutation: free
## Number of permutations: 1000
## Terms added sequentially (first to last)
##
##
              Df SumsOfSqs MeanSqs F.Model
                                                R2 Pr(>F)
## gender
                     0.562 0.56205 2.59673 0.01447 0.000999 ***
               1
                     0.175 0.17473 0.80726 0.00450 0.874126
## age
               1
## Residuals 176
                    38.094 0.21644
                                           0.98103
## Total
            178
                    38.831
                                           1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
Since sex is a significant determinant of the microbiota and can impact a number of other variables of interest,
we test if our variables vary by sex.
chisq.test(table(vars.g$gender, as.factor(vars.g$AB)))
## Pearson's Chi-squared test with Yates' continuity correction
##
## data: table(vars.g$gender, as.factor(vars.g$AB))
## X-squared = 0.025893, df = 1, p-value = 0.8722
```

```
chisq.test(table(vars.g$RU57, vars.g$gender))
## Pearson's Chi-squared test with Yates' continuity correction
##
## data: table(vars.g$RU57, vars.g$gender)
## X-squared = 1.115, df = 1, p-value = 0.291
chisq.test(table(vars.g$RU11, vars.g$gender))
## Warning in chisq.test(table(vars.g$RU11, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
## Pearson's Chi-squared test with Yates' continuity correction
##
## data: table(vars.g$RU11, vars.g$gender)
## X-squared = 1.7123, df = 1, p-value = 0.1907
chisq.test(table(vars.g$iq, vars.g$gender))
## Warning in chisq.test(table(vars.g$iq, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
  Pearson's Chi-squared test
##
## data: table(vars.g$iq, vars.g$gender)
## X-squared = 32.254, df = 40, p-value = 0.8032
chisq.test(table(vars.g$yrs.edu, vars.g$gender))
## Warning in chisq.test(table(vars.g$yrs.edu, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
## Pearson's Chi-squared test
## data: table(vars.g$yrs.edu, vars.g$gender)
## X-squared = 9.7847, df = 7, p-value = 0.2011
chisq.test(table(vars.g$cohabYN, vars.g$gender))
## Pearson's Chi-squared test with Yates' continuity correction
##
## data: table(vars.g$cohabYN, vars.g$gender)
## X-squared = 13.675, df = 1, p-value = 0.0002174
chisq.test(table(vars.g$child.sum, vars.g$gender))
## Warning in chisq.test(table(vars.g$child.sum, vars.g$gender)): Chi-squared
## approximation may be incorrect
## Pearson's Chi-squared test
## data: table(vars.g$child.sum, vars.g$gender)
## X-squared = 7.6711, df = 3, p-value = 0.05332
```

```
chisq.test(table(vars.g$groom, vars.g$gender))
## Warning in chisq.test(table(vars.g$groom, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
  Pearson's Chi-squared test
##
##
## data: table(vars.g$groom, vars.g$gender)
## X-squared = 4.5002, df = 7, p-value = 0.7207
chisq.test(table(vars.g$social.sum, vars.g$gender))
## Warning in chisq.test(table(vars.g$social.sum, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
## Pearson's Chi-squared test
##
## data: table(vars.g$social.sum, vars.g$gender)
## X-squared = 14, df = 22, p-value = 0.9015
chisq.test(table(vars.g$dog, vars.g$gender))
##
   Pearson's Chi-squared test with Yates' continuity correction
##
##
## data: table(vars.g$dog, vars.g$gender)
## X-squared = 0.56686, df = 1, p-value = 0.4515
chisq.test(table(vars.g$cat, vars.g$gender))
##
##
  Pearson's Chi-squared test with Yates' continuity correction
##
## data: table(vars.g$cat, vars.g$gender)
## X-squared = 0.98659, df = 1, p-value = 0.3206
chisq.test(table(vars.g$pet.other, vars.g$gender))
## Warning in chisq.test(table(vars.g$pet.other, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
## Pearson's Chi-squared test with Yates' continuity correction
##
## data: table(vars.g$pet.other, vars.g$gender)
## X-squared = 0.0023825, df = 1, p-value = 0.9611
chisq.test(table(vars.g$clean.house, vars.g$gender))
## Warning in chisq.test(table(vars.g$clean.house, vars.g$gender)): Chi-
## squared approximation may be incorrect
## Pearson's Chi-squared test
##
## data: table(vars.g$clean.house, vars.g$gender)
## X-squared = 3.0291, df = 6, p-value = 0.8052
```

```
chisq.test(table(vars.g$bmi, vars.g$gender))
## Warning in chisq.test(table(vars.g$bmi, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
  Pearson's Chi-squared test
##
##
## data: table(vars.g$bmi, vars.g$gender)
## X-squared = 168, df = 160, p-value = 0.3167
chisq.test(table(vars.g$srh11, vars.g$gender))
## Warning in chisq.test(table(vars.g$srh11, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
## Pearson's Chi-squared test
##
## data: table(vars.g$srh11, vars.g$gender)
## X-squared = 2.5354, df = 4, p-value = 0.6383
chisq.test(table(as.factor(vars.g$hbp11), vars.g$gender))
##
  Pearson's Chi-squared test with Yates' continuity correction
##
##
## data: table(as.factor(vars.g$hbp11), vars.g$gender)
## X-squared = 1.4052, df = 1, p-value = 0.2358
chisq.test(table(as.factor(vars.g$hbs11), vars.g$gender))
##
## Pearson's Chi-squared test with Yates' continuity correction
##
## data: table(as.factor(vars.g$hbs11), vars.g$gender)
## X-squared = 0.31741, df = 1, p-value = 0.5732
chisq.test(table(as.factor(vars.g$heart11), vars.g$gender))
##
## Pearson's Chi-squared test with Yates' continuity correction
## data: table(as.factor(vars.g$heart11), vars.g$gender)
## X-squared = 2.3549, df = 1, p-value = 0.1249
chisq.test(table(as.factor(vars.g$arth11), vars.g$gender))
##
## Pearson's Chi-squared test with Yates' continuity correction
## data: table(as.factor(vars.g$arth11), vars.g$gender)
## X-squared = 0.77841, df = 1, p-value = 0.3776
chisq.test(table(as.factor(vars.g$cancer11), vars.g$gender))
##
   Pearson's Chi-squared test with Yates' continuity correction
##
##
```

```
## data: table(as.factor(vars.g$cancer11), vars.g$gender)
## X-squared = 0.50722, df = 1, p-value = 0.4763
chisq.test(table(as.factor(vars.g$stroke11), vars.g$gender))
## Pearson's Chi-squared test with Yates' continuity correction
##
## data: table(as.factor(vars.g$stroke11), vars.g$gender)
## X-squared = 0.27776, df = 1, p-value = 0.5982
chisq.test(table(as.factor(vars.g$IBS), vars.g$gender))
## Pearson's Chi-squared test with Yates' continuity correction
## data: table(as.factor(vars.g$IBS), vars.g$gender)
## X-squared = 4.8242, df = 1, p-value = 0.02806
chisq.test(table(vars.g$walk.ave, vars.g$gender))
## Warning in chisq.test(table(vars.g$walk.ave, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
##
   Pearson's Chi-squared test
##
## data: table(vars.g$walk.ave, vars.g$gender)
## X-squared = 139.61, df = 135, p-value = 0.3753
chisq.test(table(as.factor(vars.g$smokeYN), vars.g$gender))
##
   Pearson's Chi-squared test with Yates' continuity correction
##
##
## data: table(as.factor(vars.g$smokeYN), vars.g$gender)
## X-squared = 0.0023952, df = 1, p-value = 0.961
chisq.test(table(vars.g$meat, vars.g$gender))
## Warning in chisq.test(table(vars.g$meat, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
## Pearson's Chi-squared test
## data: table(vars.g$meat, vars.g$gender)
## X-squared = 8.3147, df = 7, p-value = 0.3057
chisq.test(table(vars.g$poul, vars.g$gender))
## Warning in chisq.test(table(vars.g$poul, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
## Pearson's Chi-squared test
##
## data: table(vars.g$poul, vars.g$gender)
## X-squared = 3.8028, df = 7, p-value = 0.8022
```

```
chisq.test(table(vars.g$pork, vars.g$gender))
## Warning in chisq.test(table(vars.g$pork, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
  Pearson's Chi-squared test
##
##
## data: table(vars.g$pork, vars.g$gender)
## X-squared = 10.397, df = 7, p-value = 0.1672
chisq.test(table(vars.g$sea, vars.g$gender))
## Warning in chisq.test(table(vars.g$sea, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
## Pearson's Chi-squared test
##
## data: table(vars.g$sea, vars.g$gender)
## X-squared = 3.631, df = 4, p-value = 0.4582
chisq.test(table(vars.g$prot.sum, vars.g$gender))
## Warning in chisq.test(table(vars.g$prot.sum, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
##
   Pearson's Chi-squared test
## data: table(vars.g$prot.sum, vars.g$gender)
## X-squared = 18.766, df = 15, p-value = 0.2245
chisq.test(table(vars.g$fruit.sum, vars.g$gender))
## Warning in chisq.test(table(vars.g$fruit.sum, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
##
  Pearson's Chi-squared test
##
## data: table(vars.g$fruit.sum, vars.g$gender)
## X-squared = 22.782, df = 20, p-value = 0.2996
chisq.test(table(vars.g$veg.sum, vars.g$gender))
## Warning in chisq.test(table(vars.g$veg.sum, vars.g$gender)): Chi-squared
## approximation may be incorrect
##
##
   Pearson's Chi-squared test
## data: table(vars.g$veg.sum, vars.g$gender)
## X-squared = 40.631, df = 39, p-value = 0.3984
And correct these P-values for multiple comparison testing.
chi.list= c(0.8722,0.291,0.1907,0.2011,0.8032,0.0002174,0.05332,0.7207,0.9015,0.4515,0.167,0.9611,0.805
p.adjust(chi.list, method="BH")
```

```
## [1] 0.9611000000 0.6545133333 0.6545133333 0.6545133333 0.9244888889

## [6] 0.0033697000 0.4132300000 0.9244888889 0.9611000000 0.7382650000

## [11] 0.6545133333 0.9611000000 0.9244888889 0.6545133333 0.8603173913

## [16] 0.0000016957 0.8429181818 0.6545133333 0.6545133333 0.7013750000

## [21] 0.7382650000 0.8429181818 0.2899533333 0.9611000000 0.6545133333

## [26] 0.9244888889 0.6545133333 0.7382650000 0.6545133333 0.7264941176

## [31] 0.6545133333
```

For the variables that do differ by sex, we will stratify (strata) the model by sex. These include

- cohabYN
- · walk.ave

Antibiotics

Residuals

172

58.200 0.33837

Does antibiotic use correlate to the gut microbiota?

• AB = antibiotic use in the last 6 months, YN

```
vars = data.frame(vars.g$AB)
adonis(BC.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ as.factor(AB), data = vars.g[complete.cases(vars),],
      permuations=1000)
##
## adonis(formula = BC.dist.g[row.names(vars.g[complete.cases(vars),
                                                                          ]), row.names(vars.g[complete
##
## Permutation: free
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
##
                  Df SumsOfSqs MeanSqs F.Model
                                                    R2 Pr(>F)
## as.factor(AB)
                         0.519 0.51945 2.0615 0.01184 0.004 **
                   1
                                               0.98816
## Residuals
                 172
                        43.340 0.25198
## Total
                 173
                        43.860
                                               1.00000
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(J.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ as.factor(AB), data = vars.g[complete.cases(vars),],
      permuations=1000)
##
## adonis(formula = J.dist.g[row.names(vars.g[complete.cases(vars),
                                                                         ]), row.names(vars.g[complete.
##
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
                  Df SumsOfSqs MeanSqs F.Model
                                                    R2 Pr(>F)
## as.factor(AB)
                 1
                         0.533 0.53295
                                       1.575 0.00907 0.004 **
```

0.99093

```
## Total
                173
                       58.733
                                              1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(wUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
      ~ as.factor(AB), data = vars.g[complete.cases(vars),],
      permuations=1000)
##
## Call:
## adonis(formula = wUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                          ]), row.names(vars.g[complete
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
                 Df SumsOfSqs MeanSqs F.Model
                                                    R2 Pr(>F)
                       0.1049 0.104882 2.5225 0.01445 0.003 **
## as.factor(AB)
                  1
## Residuals
                172
                       7.1514 0.041578
                                               0.98555
## Total
                173
                       7.2563
                                               1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(uwUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
      ~ as.factor(AB), data = vars.g[complete.cases(vars),],
      permuations=1000)
##
## Call:
## adonis(formula = uwUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                           ]), row.names(vars.g[comple
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
                 Df SumsOfSqs MeanSqs F.Model
                                                   R2 Pr(>F)
## as.factor(AB)
                  1
                        0.478 0.47768 2.2207 0.01275 0.002 **
                       36.999 0.21511
## Residuals
                172
                                              0.98725
## Total
                173
                       37.476
                                              1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
```

Rural vs. urban

Does rural-urban upbringing or current residence correlate to the gut microbiota?

```
• RU57 = OCF357 = Father's occupation in 1957
```

• RU11 = hf017j1e = 1990 occupation code for graduates

```
##
## Call:
## adonis(formula = BC.dist.g[row.names(vars.g[complete.cases(vars),
                                                                           ]), row.names(vars.g[complete
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
              Df SumsOfSqs MeanSqs F.Model
                                                 R2 Pr(>F)
## RU57
                    0.2119 0.21189 0.8212 0.00696 0.734
## RU11
                    0.3152 0.31516 1.2215 0.01035
                                                    0.170
               1
## Residuals 116
                   29.9307 0.25802
                                           0.98270
                   30.4578
                                            1.00000
## Total
             118
adonis(J.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ RU57+RU11, data = vars.g[complete.cases(vars),],
       permuations=1000)
##
## Call:
## adonis(formula = J.dist.g[row.names(vars.g[complete.cases(vars),
                                                                          ]), row.names(vars.g[complete.
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
              Df SumsOfSqs MeanSqs F.Model
##
                                                R2 Pr(>F)
## RU57
                     0.315 0.31482 0.91801 0.00778 0.674
## RU11
                     0.377 0.37725 1.10005 0.00932 0.233
               1
## Residuals 116
                    39.781 0.34294
                                           0.98290
## Total
                    40.473
                                            1.00000
             118
adonis(wUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ RU57+RU11, data = vars.g[complete.cases(vars),],
       permuations=1000)
##
## adonis(formula = wUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                            ]), row.names(vars.g[complete
## Permutation: free
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
              Df SumsOfSqs MeanSqs F.Model
##
                                                  R2 Pr(>F)
                    0.0281 0.028060 0.65311 0.00554 0.854
## RU57
               1
                    0.0516 0.051578 1.20049 0.01019 0.241
## RU11
               1
## Residuals 116
                    4.9838 0.042964
                                             0.98427
## Total
             118
                    5.0634
                                             1.00000
adonis(uwUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ RU57+RU11, data = vars.g[complete.cases(vars),],
       permuations=1000)
```

```
##
## Call:
                                                                             ]), row.names(vars.g[comple
## adonis(formula = uwUF.dist.g[row.names(vars.g[complete.cases(vars),
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
              Df SumsOfSqs MeanSqs F.Model
                                                 R2 Pr(>F)
## RU57
                    0.1823 0.18225 0.82481 0.00697 0.828
               1
## RU11
                    0.3381 0.33811 1.53017 0.01293 0.032 *
               1
## Residuals 116
                   25.6316 0.22096
                                           0.98010
                                            1.00000
## Total
             118
                   26.1520
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
Education
Does education or IQ correlate to the gut microbiota?
  • yrs.edu = hb103red = years of education
  • iq = IQ (Henmom-Nelson score)
vars = data.frame(vars.g$yrs.edu, vars.g$iq)
adonis(BC.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ yrs.edu+iq, data = vars.g[complete.cases(vars),],
       permuations=1000)
##
## Call:
## adonis(formula = BC.dist.g[row.names(vars.g[complete.cases(vars),
                                                                           ]), row.names(vars.g[complete
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
              Df SumsOfSqs MeanSqs F.Model
                                                R2 Pr(>F)
                     0.285 0.28468
                                     1.117 0.00627 0.280
## yrs.edu
               1
                     0.283 0.28340
                                     1.112 0.00624 0.253
## iq
               1
## Residuals 176
                    44.855 0.25486
                                           0.98749
## Total
                    45.423
                                            1.00000
             178
adonis(J.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ yrs.edu+iq, data = vars.g[complete.cases(vars),],
       permuations=1000)
##
```

adonis(formula = J.dist.g[row.names(vars.g[complete.cases(vars),

Permutation: free

##

Number of permutations: 999

]), row.names(vars.g[complete.

```
## Terms added sequentially (first to last)
##
##
              Df SumsOfSqs MeanSqs F.Model
                     0.354 0.35372
                                    1.0386 0.00583 0.352
## yrs.edu
               1
## iq
                     0.354 0.35398
                                    1.0393 0.00584
## Residuals 176
                    59.943 0.34059
                                            0.98833
## Total
                    60.651
                                            1.00000
             178
adonis(wUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ yrs.edu+iq, data = vars.g[complete.cases(vars),],
       permuations=1000)
##
## Call:
## adonis(formula = wUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                             ]), row.names(vars.g[complete
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
              Df SumsOfSqs MeanSqs F.Model
                                                  R2 Pr(>F)
## yrs.edu
               1
                    0.0395 0.039465 0.93469 0.00525
                                                     0.485
                    0.0459 0.045856 1.08605 0.00610 0.329
               1
## Residuals 176
                    7.4311 0.042222
                                             0.98865
                    7.5164
                                             1.00000
## Total
             178
adonis(uwUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ yrs.edu+iq, data = vars.g[complete.cases(vars),],
       permuations=1000)
##
## adonis(formula = uwUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                              ]), row.names(vars.g[comple
##
## Permutation: free
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
##
              Df SumsOfSqs MeanSqs F.Model
                                                 R2 Pr(>F)
                     0.263 0.26332 1.20764 0.00678 0.122
## yrs.edu
               1
                     0.191 0.19114 0.87661 0.00492
                                                    0.701
## iq
               1
## Residuals 176
                    38.377 0.21805
                                            0.98830
## Total
             178
                    38.831
                                            1.00000
```

Human interaction

Does human interaction correlate to the gut microbiota?

- cohabYN = cohab = Cohabiting with spouse, YN (strata by sex)
- child.sum = HD01701-06 = # of children cohabiting with graduate
- groom = ha103re = Personal grooming score (0-10)
- social.sum = jz023rer + jz024rer = # of social interactions with family and friends in the last 4 wks

```
#Full model
vars = data.frame(vars.g$cohabYN, vars.g$child.sum, vars.g$groom, vars.g$social.sum)
adonis(BC.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ cohabYN + child.sum + groom + social.sum + cohabYN:social.sum,
      data = vars.g[complete.cases(vars),], strata = vars.g[complete.cases(vars),]$gender,
      permuations=1000)
##
## Call:
## adonis(formula = BC.dist.g[row.names(vars.g[complete.cases(vars),
                                                                         ]), row.names(vars.g[complete
## Blocks: strata
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
                      Df SumsOfSqs MeanSqs F.Model
                                                        R2 Pr(>F)
## cohabYN
                            0.3703 0.37031 1.48420 0.01223 0.127
                       1
## child.sum
                            0.2256 0.22562 0.90427 0.00745 0.561
## groom
                            0.2490 0.24897 0.99787 0.00822 0.505
                       1
                            0.2219 0.22194 0.88952 0.00733
## social.sum
                       1
                                                            0.603
## cohabYN:social.sum 1
                           0.5316 0.53156 2.13049 0.01755
                                                            0.004 **
## Residuals
                     115
                           28.6926 0.24950
                                                   0.94723
## Total
                           30.2909
                                                   1.00000
                     120
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(J.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ cohabYN + child.sum + groom + social.sum + cohabYN:social.sum,
       data = vars.g[complete.cases(vars),], strata = vars.g[complete.cases(vars),]$gender,
      permuations=1000)
##
## Call:
## adonis(formula = J.dist.g[row.names(vars.g[complete.cases(vars),
                                                                        ]), row.names(vars.g[complete.
##
## Blocks: strata
## Permutation: free
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
                      Df SumsOfSqs MeanSqs F.Model
                                                        R2 Pr(>F)
                             0.424 0.42442 1.26222 0.01044 0.153
## cohabYN
                       1
## child.sum
                       1
                             0.320 0.32003 0.95178 0.00788 0.522
                             0.339 0.33854 1.00681 0.00833 0.469
## groom
                       1
## social.sum
                             0.315 0.31474 0.93604 0.00775 0.606
                       1
                             0.572 0.57167 1.70014 0.01407
                                                            0.001 ***
## cohabYN:social.sum 1
## Residuals
                     115
                            38.668 0.33625
                                                   0.95154
## Total
                     120
                            40.638
                                                   1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
```

```
adonis(wUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ cohabYN + child.sum + groom + social.sum + cohabYN:social.sum,
      data = vars.g[complete.cases(vars),], strata = vars.g[complete.cases(vars),]$gender,
      permuations=1000)
##
## Call:
## adonis(formula = wUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                          ]), row.names(vars.g[complete
## Blocks: strata
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
                      Df SumsOfSqs MeanSqs F.Model
                                                         R2 Pr(>F)
## cohabYN
                            0.0674 0.067378 1.63138 0.01347 0.156
                       1
                            0.0349 0.034858 0.84401 0.00697 0.580
## child.sum
                       1
                            0.0371 0.037145 0.89937 0.00742 0.640
## groom
                       1
                            0.0354 0.035366 0.85630 0.00707 0.558
## social.sum
                       1
                            0.0787 0.078676 1.90494 0.01573 0.027 *
## cohabYN:social.sum 1
## Residuals
                     115
                            4.7496 0.041301
                                                    0.94935
## Total
                     120
                            5.0030
                                                    1.00000
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(uwUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
      ~ cohabYN + child.sum + groom + social.sum + cohabYN:social.sum,
      data = vars.g[complete.cases(vars),], strata = vars.g[complete.cases(vars),]$gender,
      permuations=1000)
##
## Call:
## adonis(formula = uwUF.dist.g[row.names(vars.g[complete.cases(vars), ]), row.names(vars.g[comple
## Blocks: strata
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
                      Df SumsOfSqs MeanSqs F.Model
                                                        R2 Pr(>F)
## cohabYN
                       1
                          0.2541 0.25415 1.19412 0.00979 0.239
## child.sum
                            0.2745 0.27450 1.28973 0.01057
                       1
                                                            0.086 .
## groom
                       1
                            0.2036 0.20363 0.95676 0.00784
                            0.2264 0.22640 1.06375 0.00872 0.277
## social.sum
                       1
                            0.5295 0.52952 2.48794 0.02039
## cohabYN:social.sum 1
## Residuals
                           24.4760 0.21283
                                                   0.94268
                     115
## Total
                     120
                           25.9642
                                                   1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
#CohabYN alone
vars = data.frame(vars.g$cohabYN)
adonis(BC.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
```

```
data = vars.g[complete.cases(vars),], strata = vars.g[complete.cases(vars),]$gender,
       permuations=1000)
##
## Call:
## adonis(formula = BC.dist.g[row.names(vars.g[complete.cases(vars),
                                                                         ]), row.names(vars.g[complete
## Blocks: strata
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
             Df SumsOfSqs MeanSqs F.Model
                                               R2 Pr(>F)
                    0.549 0.54873 2.1644 0.01208 0.031 *
## cohabYN
              1
## Residuals 177
                   44.874 0.25353
                                          0.98792
## Total
            178
                   45.423
                                           1.00000
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(J.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ cohabYN,
      data = vars.g[complete.cases(vars),], strata = vars.g[complete.cases(vars),]$gender,
      permuations=1000)
##
## Call:
## adonis(formula = J.dist.g[row.names(vars.g[complete.cases(vars),
                                                                        ]), row.names(vars.g[complete.
## Blocks: strata
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
             Df SumsOfSqs MeanSqs F.Model
                                               R2 Pr(>F)
                    0.574 0.57433 1.6921 0.00947 0.024 *
## cohabYN
              1
## Residuals 177
                   60.077 0.33942
                                          0.99053
## Total
            178
                   60.651
                                           1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(wUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ cohabYN,
      data = vars.g[complete.cases(vars),], strata = vars.g[complete.cases(vars),]$gender,
      permuations=1000)
##
## adonis(formula = wUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                          ]), row.names(vars.g[complete
## Blocks: strata
## Permutation: free
## Number of permutations: 999
```

```
##
## Terms added sequentially (first to last)
##
             Df SumsOfSqs MeanSqs F.Model
##
                                                R2 Pr(>F)
              1
                   0.0990 0.098986 2.3621 0.01317 0.043 *
## Residuals 177
                   7.4174 0.041906
                                           0.98683
## Total
                   7.5164
                                           1.00000
            178
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(uwUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ cohabYN,
      data = vars.g[complete.cases(vars),], strata = vars.g[complete.cases(vars),]$gender,
      permuations=1000)
##
## Call:
## adonis(formula = uwUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                          ]), row.names(vars.g[comple
## Blocks: strata
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
             Df SumsOfSqs MeanSqs F.Model
##
                                               R2 Pr(>F)
                    0.386 0.38601 1.7772 0.00994 0.042 *
## cohabYN
              1
## Residuals 177
                   38.445 0.21720
                                          0.99006
## Total
            178
                   38.831
                                          1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
#Socialness alone
##Within cohabiting persons
vars = data.frame(vars.g$social.sum)
adonis(BC.dist.g[row.names(vars.g[complete.cases(vars) & vars.g$cohabYN == "Y",]), row.names(vars.g[com
      ~ social.sum,
      data = vars.g[complete.cases(vars) & vars.g$cohabYN == "Y",], permuations=1000)
##
## Call:
## adonis(formula = BC.dist.g[row.names(vars.g[complete.cases(vars) & vars.g$cohabYN == "Y", ]), r
## Permutation: free
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
              Df SumsOfSqs MeanSqs F.Model
                                                R2 Pr(>F)
                    0.3459 0.34587 1.4415 0.01435
                                                     0.05 *
## social.sum
## Residuals
              99
                   23.7545 0.23994
                                           0.98565
## Total
             100
                   24.1004
                                           1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
```

```
adonis(J.dist.g[row.names(vars.g[complete.cases(vars) & vars.g$cohabYN == "Y",]), row.names(vars.g[comp
              ~ social.sum,
              data = vars.g[complete.cases(vars) & vars.g$cohabYN == "Y",], permuations=1000)
##
## Call:
## adonis(formula = J.dist.g[row.names(vars.g[complete.cases(vars) &
                                                                                                                                                         vars.g$cohabYN == "Y", ]), ro
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
                              Df SumsOfSqs MeanSqs F.Model
                                                                                                     R2 Pr(>F)
                                            0.419 0.41936 1.2769 0.01273 0.056 .
## social.sum
                              1
## Residuals
                              99
                                           32.513 0.32841
                                                                                           0.98727
## Total
                             100
                                           32.932
                                                                                            1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(wUF.dist.g[row.names(vars.g[complete.cases(vars) & vars.g$cohabYN == "Y",]), row.names(vars.g[complete.cases(vars) & vars.g$cohabYN == "Y",]), row.names(vars.g[complete.cases(vars.g] & vars.g[complete.cases(vars.g] & va
              ~ social.sum,
              data = vars.g[complete.cases(vars) & vars.g$cohabYN == "Y",], permuations=1000)
##
## adonis(formula = wUF.dist.g[row.names(vars.g[complete.cases(vars) & vars.g$cohabYN == "Y", ]),
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
                              Df SumsOfSqs MeanSqs F.Model
                                                                                                   R2 Pr(>F)
                                           0.0487 0.048736 1.2118 0.01209 0.239
## social.sum
                               1
                                           3.9816 0.040218
## Residuals
                              99
                                                                                             0.98791
                                           4.0303
                                                                                              1.00000
## Total
                             100
adonis(uwUF.dist.g[row.names(vars.g[complete.cases(vars) & vars.g$cohabYN == "Y",]), row.names(vars.g[c
              ~ social.sum,
              data = vars.g[complete.cases(vars) & vars.g$cohabYN == "Y",], permuations=1000)
##
## Call:
## adonis(formula = uwUF.dist.g[row.names(vars.g[complete.cases(vars) &
                                                                                                                                                            vars.g$cohabYN == "Y", ]),
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
                              Df SumsOfSqs MeanSqs F.Model
                                                                                                     R2 Pr(>F)
                                          0.2859 0.28593 1.3458 0.01341 0.064 .
## social.sum
                               1
## Residuals
                                         21.0331 0.21246
                                                                                           0.98659
                              99
## Total
                                        21.3190
                                                                                            1.00000
                            100
```

```
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
##Within non-cohabiting persons
vars = data.frame(vars.g$social.sum)
adonis(BC.dist.g[row.names(vars.g[complete.cases(vars) & vars.g$cohabYN == "N",]), row.names(vars.g[com
              data = vars.g[complete.cases(vars) & vars.g$cohabYN == "N",], permuations=1000)
##
## adonis(formula = BC.dist.g[row.names(vars.g[complete.cases(vars) &
                                                                                                                                                               vars.g$cohabYN == "N", ]), r
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
                             Df SumsOfSqs MeanSqs F.Model
                                                                                                     R2 Pr(>F)
## social.sum 1
                                         0.4647 0.46470 1.7361 0.04062 0.016 *
                                        10.9744 0.26767
## Residuals 41
                                                                                           0.95938
## Total
                             42
                                     11.4391
                                                                                           1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(J.dist.g[row.names(vars.g[complete.cases(vars) & vars.g$cohabYN == "N",]), row.names(vars.g[comp
              data = vars.g[complete.cases(vars) & vars.g$cohabYN == "N",], permuations=1000)
##
## adonis(formula = J.dist.g[row.names(vars.g[complete.cases(vars) &
                                                                                                                                                            vars.g$cohabYN == "N", ]), ro
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
                             Df SumsOfSqs MeanSqs F.Model
##
                                         0.5139 0.51385 1.4652 0.0345 0.011 *
## social.sum 1
## Residuals 41
                                        14.3784 0.35069
                                                                                           0.9655
## Total
                             42
                                     14.8923
                                                                                           1.0000
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(wUF.dist.g[row.names(vars.g[complete.cases(vars) & vars.g$cohabYN == "N",]), row.names(vars.g[complete.cases(vars) & vars.g$cohabYN == "N",]), row.names(vars.g[complete.cases(vars.g] & vars.g[complete.cases(vars.g] & va
              ~ social.sum,
              data = vars.g[complete.cases(vars) & vars.g$cohabYN == "N",], permuations=1000)
##
## adonis(formula = wUF.dist.g[row.names(vars.g[complete.cases(vars) &
                                                                                                                                                                 vars.g$cohabYN == "N", ]),
## Permutation: free
## Number of permutations: 999
##
```

```
## Terms added sequentially (first to last)
##
##
              Df SumsOfSqs MeanSqs F.Model
                   0.07337 0.073372 1.7404 0.04072 0.046 *
## social.sum 1
## Residuals 41
                   1.72849 0.042158
                                            0.95928
## Total
              42
                   1.80186
                                            1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(uwUF.dist.g[row.names(vars.g[complete.cases(vars) & vars.g$cohabYN == "N",]), row.names(vars.g[c
       ~ social.sum.
       data = vars.g[complete.cases(vars) & vars.g$cohabYN == "N",], permuations=1000)
##
## Call:
## adonis(formula = uwUF.dist.g[row.names(vars.g[complete.cases(vars) &
                                                                              vars.g$cohabYN == "N", ]),
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
              Df SumsOfSqs MeanSqs F.Model
                                                R2 Pr(>F)
                    0.4314 0.43141 1.9549 0.04551 0.006 **
## social.sum 1
## Residuals 41
                    9.0478 0.22068
                                           0.95449
                    9.4792
                                           1.00000
## Total
              42
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
Home environment
Does the home environment correlate to the gut microbiota?
  • dog = dog in house
  • cat = cat in house
  • pet.other = Other pet (bird, reptile, fish) in house
  • clean.house = ha114re = Residence cleanliness score (1-10)
vars = data.frame(vars.g$dog, vars.g$cat, vars.g$pet.other, vars.g$clean.house)
adonis(BC.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ dog+cat+pet.other+clean.house, data = vars.g[complete.cases(vars),],
       permuations=1000)
##
## adonis(formula = BC.dist.g[row.names(vars.g[complete.cases(vars),
                                                                           ]), row.names(vars.g[complete
##
```

0.178 0.17757 0.69904 0.00524 0.924 0.249 0.24906 0.98046 0.00735 0.442

R2 Pr(>F)

Permutation: free

##

dog

cat

Number of permutations: 999

1

Terms added sequentially (first to last)

Df SumsOfSqs MeanSqs F.Model

```
## pet.other
                       0.208 0.20812 0.81930 0.00614 0.711
                       0.221 0.22134 0.87136 0.00653 0.665
## clean.house
                 1
## Residuals
               130
                      33.023 0.25402
                                              0.97473
## Total
                                              1.00000
               134
                      33.879
adonis(J.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ dog+cat+pet.other+clean.house, data = vars.g[complete.cases(vars),],
       permuations=1000)
##
## Call:
## adonis(formula = J.dist.g[row.names(vars.g[complete.cases(vars),
                                                                          ]), row.names(vars.g[complete.
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
                Df SumsOfSqs MeanSqs F.Model
##
                                                   R2 Pr(>F)
                       0.276 0.27604 0.81305 0.00608 0.921
## dog
                 1
                       0.329 0.32871 0.96818 0.00725
## cat
                 1
## pet.other
                 1
                       0.311 0.31137 0.91711 0.00686
                                                      0.628
## clean.house
                 1
                       0.313 0.31302 0.92197 0.00690
## Residuals
                      44.137 0.33951
                                              0.97291
               130
## Total
               134
                      45.366
                                             1.00000
adonis(wUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ dog+cat+pet.other+clean.house, data = vars.g[complete.cases(vars),],
       permuations=1000)
##
## Call:
## adonis(formula = wUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                            ]), row.names(vars.g[complete
## Permutation: free
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
##
                Df SumsOfSqs MeanSqs F.Model
                                                    R2 Pr(>F)
## dog
                      0.0281 0.028096 0.66144 0.00497 0.855
                      0.0475 0.047492 1.11807 0.00840
## cat
                      0.0235 0.023481 0.55280 0.00415
                                                       0.902
## pet.other
                 1
## clean.house
                 1
                      0.0344 0.034381 0.80941 0.00608
                                                       0.647
## Residuals
               130
                      5.5220 0.042477
                                              0.97640
## Total
               134
                      5.6554
                                               1.00000
adonis(uwUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ dog+cat+pet.other+clean.house, data = vars.g[complete.cases(vars),],
       permuations=1000)
##
## adonis(formula = uwUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                             ]), row.names(vars.g[comple
##
## Permutation: free
```

```
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
##
                Df SumsOfSqs MeanSqs F.Model
                                                  R2 Pr(>F)
## dog
                      0.1735 0.17350 0.79469 0.00594 0.916
                      0.1971 0.19712 0.90287 0.00675 0.647
## cat
                      0.2326 0.23263 1.06551 0.00797
## pet.other
                 1
                                                      0.297
## clean.house
                 1
                      0.2093 0.20932 0.95872 0.00717
## Residuals
               130
                     28.3827 0.21833
                                             0.97217
## Total
               134
                     29.1952
                                             1.00000
```

Health

Does health correlate to the gut microbiota?

```
• bmi = body mass index based on height and weight
```

- srh11 = Self-reported health score (1-5)
- walk.ave = (HX472RE+HX473RE)/2 = Average walking speed (sec) (strate by sex)
- hbs11 = High-blood sugar, YN
- hbp11 = High-blood pressure, YN
- heart11 = Heart disease, YN
- arth11 = Arthritis, YN
- cancer11 = Cancer, YN
- stroke11 = Stroke, YN
- IBS = irritable bowel syndrome, YN

```
#overall health
vars = data.frame(vars.g$bmi, vars.g$srh11, vars.g$walk.ave)
adonis(BC.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ bmi+srh11+walk.ave, data = vars.g[complete.cases(vars),],
       strata = vars.g[complete.cases(vars),]$gender,
       permuations=1000)
##
## Call:
## adonis(formula = BC.dist.g[row.names(vars.g[complete.cases(vars),
                                                                          ]), row.names(vars.g[complete
## Blocks: strata
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
              Df SumsOfSqs MeanSqs F.Model
##
                                                R2 Pr(>F)
## bmi
                     0.365 0.36461 1.42693 0.00863 0.052
                     0.276 0.27648 1.08202 0.00654
## srh11
               1
                                                    0.274
## walk.ave
              1
                     0.226 0.22579 0.88365 0.00534
## Residuals 162
                   41.394 0.25552
                                           0.97949
## Total
            165
                    42.261
                                           1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(J.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       bmi+srh11+walk.ave, data = vars.g[complete.cases(vars),],
```

```
strata = vars.g[complete.cases(vars),]$gender,
       permuations=1000)
##
## Call:
## adonis(formula = J.dist.g[row.names(vars.g[complete.cases(vars),
                                                                         ]), row.names(vars.g[complete.
##
## Blocks: strata
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
              Df SumsOfSqs MeanSqs F.Model
                                                R2 Pr(>F)
                     0.444 0.44406 1.30288 0.00788
## bmi
                                                   0.039 *
               1
## srh11
                     0.360 0.35992 1.05602 0.00639
## walk.ave
                     0.308 0.30780 0.90308 0.00546
                                                    0.740
               1
                    55.214 0.34083
                                           0.98026
## Residuals 162
                                           1.00000
## Total
             165
                    56.326
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(wUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ bmi+srh11+walk.ave, data = vars.g[complete.cases(vars),],
       strata = vars.g[complete.cases(vars),]$gender,
      permuations=1000)
##
## Call:
## adonis(formula = wUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                           ]), row.names(vars.g[complete
## Blocks: strata
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
              Df SumsOfSqs MeanSqs F.Model
                                                 R2 Pr(>F)
## bmi
                    0.0472 0.047222 1.11408 0.00676 0.284
## srh11
               1
                    0.0403 0.040271 0.95009 0.00576 0.481
## walk.ave
                    0.0317 0.031706 0.74803 0.00454 0.777
              1
                    6.8666 0.042386
## Residuals 162
                                            0.98294
## Total
             165
                    6.9858
                                            1.00000
adonis(uwUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ bmi+srh11+walk.ave, data = vars.g[complete.cases(vars),],
       strata = vars.g[complete.cases(vars),]$gender,
      permuations=1000)
##
## Call:
## adonis(formula = uwUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                            ]), row.names(vars.g[comple
## Blocks: strata
## Permutation: free
```

```
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
##
             Df SumsOfSqs MeanSqs F.Model
                                                R2 Pr(>F)
                     0.299 0.29876 1.3751 0.00831 0.055 .
## bmi
                     0.218 0.21768 1.0019 0.00605 0.371
## srh11
              1
## walk.ave
                     0.240 0.23992 1.1043 0.00667
              1
                                                    0.267
## Residuals 162
                    35.197 0.21727
                                           0.97896
## Total
            165
                    35.954
                                           1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
#Specific diseases
vars = data.frame(vars.g$hbs11, vars.g$hbp11, vars.g$heart11, vars.g$arth11, vars.g$cancer11, vars.g$st
adonis(BC.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ as.factor(hbs11) + as.factor(hbp11) + as.factor(heart11) + as.factor(arth11) + as.factor(cance)
       data = vars.g[complete.cases(vars),], permuations=1000)
##
## Call:
## adonis(formula = BC.dist.g[row.names(vars.g[complete.cases(vars),
                                                                          ]), row.names(vars.g[complete
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
                        Df SumsOfSqs MeanSqs F.Model
                                                          R2 Pr(>F)
## as.factor(hbs11)
                               0.506 0.50572 2.00886 0.01285 0.005 **
## as.factor(hbp11)
                               0.260 0.25952 1.03090 0.00659
                         1
## as.factor(heart11)
                              0.476 0.47644 1.89255 0.01210 0.006 **
                        1
## as.factor(arth11)
                         1
                              0.172 0.17169 0.68201 0.00436
## as.factor(cancer11)
                         1
                              0.216 0.21558 0.85636 0.00548 0.667
## as.factor(stroke11)
                         1
                              0.174 0.17412 0.69164 0.00442 0.919
## as.factor(IBS)
                         1
                              0.299 0.29913 1.18823 0.00760 0.215
## Residuals
                       148
                              37.258 0.25174
                                                     0.94659
## Total
                              39.360
                       155
                                                     1.00000
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(J.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ as.factor(hbs11) + as.factor(hbp11) + as.factor(heart11) + as.factor(arth11) + as.factor(cance
      data = vars.g[complete.cases(vars),], permuations=1000)
##
## adonis(formula = J.dist.g[row.names(vars.g[complete.cases(vars),
                                                                         ]), row.names(vars.g[complete.
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
                        Df SumsOfSqs MeanSqs F.Model
                                                          R2 Pr(>F)
```

```
## as.factor(hbs11)
                              0.549 0.54890 1.62396 0.01043 0.004 **
                         1
                              0.341 0.34113 1.00928 0.00648 0.399
## as.factor(hbp11)
                         1
## as.factor(heart11)
                              0.495 0.49529 1.46538 0.00941
                                                             0.024 *
## as.factor(arth11)
                              0.276 0.27642 0.81782 0.00525
                                                             0.900
                         1
## as.factor(cancer11)
                         1
                              0.297 0.29733 0.87968 0.00565
## as.factor(stroke11)
                              0.263 0.26348 0.77953 0.00501 0.955
                         1
## as.factor(IBS)
                              0.382 0.38206 1.13037 0.00726 0.175
                         1
## Residuals
                       148
                              50.024 0.33800
                                                     0.95051
## Total
                       155
                              52.628
                                                     1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(wUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ as.factor(hbs11) + as.factor(hbp11) + as.factor(heart11) + as.factor(arth11) + as.factor(cance
       data = vars.g[complete.cases(vars),], permuations=1000)
##
## adonis(formula = wUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                          ]), row.names(vars.g[complete
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
                        Df SumsOfSqs MeanSqs F.Model
                                                           R2 Pr(>F)
## as.factor(hbs11)
                             0.0774 0.077362 1.87129 0.01201 0.025 *
                         1
## as.factor(hbp11)
                             0.0356 0.035631 0.86186 0.00553 0.594
## as.factor(heart11)
                             0.0714 0.071404 1.72718 0.01109 0.060 .
                         1
## as.factor(arth11)
                        1
                             0.0223 0.022300 0.53942 0.00346 0.949
                             0.0388 0.038782 0.93808 0.00602 0.491
## as.factor(cancer11)
                        1
## as.factor(stroke11)
                             0.0295 0.029487 0.71326 0.00458 0.776
                             0.0470 0.047004 1.13697 0.00730 0.287
## as.factor(IBS)
                        1
## Residuals
                       148
                              6.1185 0.041341
                                                      0.95001
## Total
                       155
                              6.4405
                                                      1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(uwUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ as.factor(hbs11) + as.factor(hbp11) + as.factor(heart11) + as.factor(arth11) + as.factor(cance)
       data = vars.g[complete.cases(vars),], permuations=1000)
##
## Call:
## adonis(formula = uwUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                           ]), row.names(vars.g[comple
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
                        Df SumsOfSqs MeanSqs F.Model
                                                         R2 Pr(>F)
## as.factor(hbs11)
                              0.412 0.41164 1.89555 0.01213 0.002 **
## as.factor(hbp11)
                         1
                               0.230 0.23001 1.05914 0.00678 0.294
## as.factor(heart11)
                              0.361 0.36148 1.66454 0.01065 0.009 **
                         1
```

```
## as.factor(arth11)
                               0.207 0.20693 0.95288 0.00610 0.521
                         1
## as.factor(cancer11)
                               0.178 0.17792 0.81927 0.00524 0.834
                         1
## as.factor(stroke11)
                         1
                               0.179 0.17899 0.82421 0.00528 0.830
## as.factor(IBS)
                               0.223 0.22299 1.02685 0.00657 0.358
                         1
## Residuals
                       148
                              32.140 0.21716
                                                     0.94725
## Total
                              33.930
                                                     1.00000
                       155
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
Smoking
Does smoking impact the microbiota?
  • smokeYN = jx013rec = currently a smoker, YN
vars = data.frame(vars.g$smokeYN)
adonis(BC.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ as.factor(smokeYN), data = vars.g[complete.cases(vars),],
      permuations=1000)
##
## Call:
## adonis(formula = BC.dist.g[row.names(vars.g[complete.cases(vars),
                                                                          ]), row.names(vars.g[complete
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
                       Df SumsOfSqs MeanSqs F.Model
                              0.270 0.26958 1.0582 0.00691
## as.factor(smokeYN)
                                                               0.34
                             38.724 0.25476
## Residuals
                      152
                                                    0.99309
## Total
                      153
                             38.994
                                                    1.00000
adonis(J.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ as.factor(smokeYN), data = vars.g[complete.cases(vars),],
      permuations=1000)
##
## adonis(formula = J.dist.g[row.names(vars.g[complete.cases(vars),
                                                                         ]), row.names(vars.g[complete.
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
                       Df SumsOfSqs MeanSqs F.Model
## as.factor(smokeYN)
                              0.362 0.36222 1.0642 0.00695 0.259
                        1
## Residuals
                             51.736 0.34037
                                                    0.99305
## Total
                             52.099
                      153
                                                    1.00000
adonis(wUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
```

~ as.factor(smokeYN), data = vars.g[complete.cases(vars),],

permuations=1000)

```
##
## Call:
## adonis(formula = wUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                             ]), row.names(vars.g[complete
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
                        Df SumsOfSqs MeanSqs F.Model
                                                            R2 Pr(>F)
## as.factor(smokeYN)
                              0.0370 0.036964 0.8926 0.00584 0.574
## Residuals
                              6.2945 0.041411
                                                       0.99416
                      152
                                                       1.00000
## Total
                      153
                              6.3314
adonis(uwUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ as.factor(smokeYN), data = vars.g[complete.cases(vars),],
       permuations=1000)
##
## Call:
## adonis(formula = uwUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                              ]), row.names(vars.g[comple
##
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
                       Df SumsOfSqs MeanSqs F.Model
                              0.215 0.21537 0.99058 0.00647 0.427
## as.factor(smokeYN)
                        1
## Residuals
                      152
                              33.048 0.21742
                                                     0.99353
## Total
                      153
                             33.263
                                                      1.00000
Diet
Does diet impact the microbiota?
  • prot.sum = Times consume protein / wk
  • fruit.sum = # different fruits consume / week
  • veg.sum = # different vegetables consume / week
  • meat = Times consume (red) meat / week
  • poul = Times consume poultry / week
  • pork = Times consume pork / week
  • sea = Times consume seafood / week
#Protein, veg and fruits
vars = data.frame(vars.g$prot.sum, vars.g$fruit.sum, vars.g$veg.sum)
adonis(BC.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ prot.sum + fruit.sum + veg.sum,
       data = vars.g[complete.cases(vars),],
       permuations=1000)
```

]), row.names(vars.g[complete

adonis(formula = BC.dist.g[row.names(vars.g[complete.cases(vars),

Call:

```
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
              Df SumsOfSqs MeanSqs F.Model
##
                                                R2 Pr(>F)
                     0.482 0.48164 1.91298 0.01150 0.007 **
## prot.sum
              1
                     0.215 0.21509 0.85429 0.00514 0.681
## fruit.sum
              1
## veg.sum
              1
                     0.393 0.39348 1.56285 0.00940 0.045 *
## Residuals 162
                    40.787 0.25177
                                           0.97397
## Total
            165
                    41.877
                                           1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(J.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ prot.sum + fruit.sum + veg.sum,
       data = vars.g[complete.cases(vars),],
      permuations=1000)
##
## Call:
## adonis(formula = J.dist.g[row.names(vars.g[complete.cases(vars),
                                                                         ]), row.names(vars.g[complete.
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
              Df SumsOfSqs MeanSqs F.Model
##
                                                R2 Pr(>F)
                     0.522 0.52242 1.54591 0.00932 0.008 **
## prot.sum
               1
## fruit.sum
                     0.308 0.30806 0.91161 0.00550 0.695
               1
## veg.sum
                     0.456 0.45584 1.34889 0.00814 0.034 *
              1
                   54.746 0.33794
## Residuals 162
                                           0.97704
## Total
             165
                   56.032
                                           1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
adonis(wUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ prot.sum + fruit.sum + veg.sum,
       data = vars.g[complete.cases(vars),],
      permuations=1000)
##
## Call:
## adonis(formula = wUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                           ]), row.names(vars.g[complete
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
              Df SumsOfSqs MeanSqs F.Model
                                                 R2 Pr(>F)
##
                   0.1028 0.102773 2.4908 0.01491 0.004 **
## prot.sum
              1
## fruit.sum
                    0.0517 0.051650 1.2518 0.00750 0.201
              1
                   0.0525 0.052510 1.2726 0.00762 0.208
## veg.sum
               1
```

```
## Residuals 162
                   6.6842 0.041261
                                           0.96997
## Total
            165
                   6.8911
                                           1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(uwUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ prot.sum + fruit.sum + veg.sum,
       data = vars.g[complete.cases(vars),],
      permuations=1000)
##
## Call:
## adonis(formula = uwUF.dist.g[row.names(vars.g[complete.cases(vars), ]), row.names(vars.g[comple
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
             Df SumsOfSqs MeanSqs F.Model
                                               R2 Pr(>F)
                    0.290 0.28977 1.33965 0.00808 0.072 .
## prot.sum
              1
## fruit.sum
              1
                    0.197 0.19718 0.91161 0.00550 0.621
## veg.sum
              1
                    0.313 0.31307 1.44737 0.00873 0.054 .
                   35.041 0.21630
                                          0.97768
## Residuals 162
## Total
            165
                   35.841
                                          1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
#Specific proteins
vars = data.frame(vars.g$meat, vars.g$poul, vars.g$pork, vars.g$sea)
adonis(BC.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ meat+poul+pork+sea+meat:poul,
       data = vars.g[complete.cases(vars),],
      permuations=1000)
##
## Call:
## adonis(formula = BC.dist.g[row.names(vars.g[complete.cases(vars),
                                                                         ]), row.names(vars.g[complete
## Permutation: free
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
##
             Df SumsOfSqs MeanSqs F.Model
                                               R2 Pr(>F)
                    0.251 0.25125 1.0030 0.00600 0.425
## meat
              1
## poul
              1
                    0.334 0.33406 1.3336 0.00798 0.093 .
                    0.370 0.36992 1.4767 0.00883 0.050 *
## pork
              1
              1
                    0.341 0.34135 1.3626 0.00815 0.089 .
## sea
                    0.501 0.50053 1.9981 0.01195
                                                   0.007 **
## meat:poul
              1
## Residuals 160
                   40.080 0.25050
                                          0.95709
## Total
            165
                   41.877
                                          1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
```

```
adonis(J.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ meat+poul+pork+sea+meat:poul,
       data = vars.g[complete.cases(vars),],
       permuations=1000)
##
## Call:
## adonis(formula = J.dist.g[row.names(vars.g[complete.cases(vars),
                                                                        ]), row.names(vars.g[complete.
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
             Df SumsOfSqs MeanSqs F.Model
                                               R2 Pr(>F)
## meat
                    0.342 0.34181 1.0140 0.00610 0.403
                    0.386 0.38579 1.1444 0.00689 0.178
## poul
              1
                    0.429 0.42875 1.2719 0.00765
## pork
              1
                                                   0.073 .
                    0.416 0.41641 1.2353 0.00743
                                                   0.073 .
## sea
              1
## meat:poul
              1
                    0.524 0.52377 1.5538 0.00935
                                                   0.005 **
## Residuals 160
                   53.935 0.33710
                                          0.96258
## Total
            165
                   56.032
                                          1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(wUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ meat+poul+pork+sea+meat:poul,
       data = vars.g[complete.cases(vars),],
      permuations=1000)
##
## adonis(formula = wUF.dist.g[row.names(vars.g[complete.cases(vars),
                                                                          ]), row.names(vars.g[complete
##
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
             Df SumsOfSqs MeanSqs F.Model
                                                R2 Pr(>F)
## meat
                   0.0723 0.072346 1.7716 0.01050 0.043 *
              1
                   0.0623 0.062263 1.5247 0.00904 0.090 .
## poul
              1
                   0.0785 0.078518 1.9228 0.01139 0.026 *
## pork
              1
## sea
                   0.0427 0.042669 1.0449 0.00619 0.363
              1
## meat:poul
              1
                   0.1016 0.101576 2.4874 0.01474 0.004 **
                   6.5338 0.040836
## Residuals 160
                                           0.94814
## Total
                   6.8911
                                            1.00000
            165
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
adonis(uwUF.dist.g[row.names(vars.g[complete.cases(vars),]),row.names(vars.g[complete.cases(vars),])]
       ~ meat+poul+pork+sea+meat:poul,
       data = vars.g[complete.cases(vars),],
      permuations=1000)
```

```
##
## Call:
## adonis(formula = uwUF.dist.g[row.names(vars.g[complete.cases(vars), ]), row.names(vars.g[comple
## Permutation: free
## Number of permutations: 999
## Terms added sequentially (first to last)
##
##
             Df SumsOfSqs MeanSqs F.Model
                                             R2 Pr(>F)
## meat
                   0.215 0.21528 1.0004 0.00601 0.387
              1
                   0.259 0.25941 1.2055 0.00724 0.118
## poul
              1
             1
                  0.274 0.27445 1.2754 0.00766 0.100
## pork
## sea
              1
                  0.246 0.24631 1.1446 0.00687 0.156
                   0.416 0.41588 1.9327 0.01160 0.007 **
## meat:poul 1
## Residuals 160
                  34.430 0.21518
                                         0.96062
## Total
            165
                35.841
                                         1.00000
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
```

FDR-correction

Correct P-values for beta-diversity tests across all graduates for multiple comparison testing.

```
BC.list = c(0.000999, 0.346653, 0.002, 0.726, 0.17, 0.277, 0.297, 0.125, 0.551, 0.517, 0.623, 0.003, 0.026, 0.061, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.003, 0.00
 J.list = c(0.000999, 0.323676, 0.002, 0.652, 0.239, 0.361, 0.355, 0.149, 0.533, 0.482, 0.607, 0.006, 0.019, 0.057, 0.008, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.019, 0.0
uwUF.list = c(0.000999, 0.888112, 0.001, 0.863, 0.032, 0.142, 0.738, 0.212, 0.108, 0.541, 0.273, 0.001, 0.046, 0.066
p.adjust(BC.list, method="BH")
## [1] 0.03796200 0.57991304 0.03800000 0.81141176 0.38000000 0.52630000
## [7] 0.53742857 0.29687500 0.77548148 0.75561538 0.78187879 0.03800000
## [13] 0.10977778 0.19316667 0.09025000 0.91500000 0.85988571 0.78187879
## [19] 0.78187879 0.19316667 0.52630000 0.78187879 0.04433333 0.62776000
## [25] 0.04433333 0.91500000 0.78187879 0.91500000 0.40111111 0.57991304
## [31] 0.05971429 0.78187879 0.17100000 0.62776000 0.24971429 0.19584615
## [37] 0.27106667 0.03800000
p.adjust(J.list, method="BH")
## [1] 0.03796200 0.58569943 0.03800000 0.82586667 0.50455556 0.59643478
## [7] 0.59643478 0.37746667 0.75014815 0.70446154 0.79800000 0.05066667
## [13] 0.08022222 0.17246154 0.05066667 0.94075676 0.88377143 0.79800000
## [19] 0.83837500 0.16581818 0.55670000 0.84494118 0.05428571 0.62016000
## [25] 0.08022222 0.94075676 0.84494118 0.94200000 0.38000000 0.55670000
## [31] 0.05066667 0.83837500 0.12160000 0.62016000 0.38000000 0.17246154
## [37] 0.23071429 0.05066667
p.adjust(wUF.list, method="BH")
## [1] 0.0379620 0.8761111 0.0570000 0.8761111 0.5676250 0.7757241 0.6677143
## [8] 0.4092308 0.7757241 0.7954667 0.7757241 0.1456667 0.1786000 0.5742222
## [15] 0.1786000 0.8761111 0.7757241 0.8965946 0.8470323 0.5742222 0.7757241
## [22] 0.8761111 0.1456667 0.7757241 0.1830909 0.9590000 0.7757241 0.8761111
```

```
## [29] 0.6040000 0.7757241 0.0570000 0.4813333 0.4180000 0.1786000 0.2850000
## [36] 0.1520000 0.6677143 0.0570000

p.adjust(uwUF.list, method="BH")

## [1] 0.01266667 0.91211503 0.01266667 0.91211503 0.13933333 0.33305882
## [7] 0.87637500 0.41420000 0.27360000 0.68526667 0.45104348 0.01266667

## [13] 0.17480000 0.19584615 0.03166667 0.93100000 0.91088235 0.41420000
## [19] 0.66172414 0.18309091 0.57152000 0.45104348 0.02850000 0.45104348
## [25] 0.04885714 0.66172414 0.91211503 0.91088235 0.54150000 0.58829630
## [31] 0.19584615 0.81393548 0.13933333 0.58829630 0.33305882 0.24700000

## [37] 0.40111111 0.03040000
```

Alpha-diversity

Tests performed on the graduate samples to avoid possible interactions between spouses and siblings. First, check if the graduate subset is different from the other sample types to ensure the subset is a good representation of the overall data set.

```
summary(aov(shannon ~ Link, data=Met))
##
                Df Sum Sq Mean Sq F value Pr(>F)
                     0.84 0.2804
                                    1.491 0.216
## Link
                   75.96 0.1880
## Residuals
               404
summary(aov((1/simpson) ~ Link, data=Met))
##
                Df Sum Sq Mean Sq F value Pr(>F)
                      486
                          161.99
## Link
                 3
                                    2.114 0.0979 .
                    30964
                            76.64
## Residuals
               404
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
summary(aov(chao ~ Link, data=Met))
##
                   Sum Sq Mean Sq F value Pr(>F)
                   130351
                             43450
                                     1.787 0.149
## Link
               404 9825590
## Residuals
                             24321
summary(aov(ace ~ Link, data=Met))
##
                Df
                     Sum Sq Mean Sq F value Pr(>F)
## Link
                 3
                     207106
                              69035
                                      1.639
                                              0.18
## Residuals
               404 17014109
                              42114
```

There are no differences so we move forward with the graduate subset data set. We will run a number of ANOVA (aov) tests of variables of interest similar to those run on beta-diversity. All tests are run on the Shannon's diversity index and Chao richness estimate. For more information on variables, see the corresponding beta-diversity test.

Sex and age

```
1 0.01 0.0130
                                  0.066 0.798
              176 34.87 0.1981
## Residuals
summary(aov(Met.g$chao ~ gender+age, data = vars.g))
               Df Sum Sq Mean Sq F value Pr(>F)
## gender
               1 115499 115499
                                   5.620 0.0188 *
## age
                1
                     390
                             390
                                   0.019 0.8906
              176 3616849
                           20550
## Residuals
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
Antibiotics
summary(aov(Met.g$shannon ~ as.factor(AB), data = vars.g))
                 Df Sum Sq Mean Sq F value Pr(>F)
                1 1.444 1.4439
                                  7.906 0.0055 **
## as.factor(AB)
               172 31.412 0.1826
## Residuals
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 5 observations deleted due to missingness
summary(aov(Met.g$chao ~ as.factor(AB), data = vars.g))
                 Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(AB) 1 112743 112743
                                    5.601 0.0191 *
## Residuals
                172 3462316
                             20130
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 5 observations deleted due to missingness
Rural vs. urban
summary(aov(Met.g$shannon ~ RU57*RU11, data = vars.g))
               Df Sum Sq Mean Sq F value Pr(>F)
## RU57
                1 0.354 0.3544
                                 1.607 0.2075
                                  2.260 0.1355
               1 0.498 0.4983
## RU11
## RU57:RU11
              1 0.637 0.6370
                                  2.888 0.0919 .
## Residuals
             115 25.363 0.2205
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 60 observations deleted due to missingness
summary(aov(Met.g$chao ~ RU57*RU11, data = vars.g))
               Df Sum Sq Mean Sq F value
##
                                          Pr(>F)
                   49357
                          49357
                                   2.424 0.122275
## RU57
                1
                1 241472 241472 11.857 0.000802 ***
## RU11
                                  4.397 0.038202 *
## RU57:RU11
               1
                    89542
                           89542
## Residuals
              115 2342104
                           20366
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
```

Education

```
summary(aov(Met.g$shannon ~ yrs.edu+iq, data = vars.g))
               Df Sum Sq Mean Sq F value Pr(>F)
## yrs.edu
               1 0.30 0.30411
                                 1.530 0.218
## iq
                1
                  0.01 0.01281
                                  0.064 0.800
## Residuals
              176 34.98 0.19873
summary(aov(Met.g$chao ~ yrs.edu+iq, data = vars.g))
##
               Df Sum Sq Mean Sq F value Pr(>F)
## yrs.edu
               1 124867 124867
                                   6.125 0.0143 *
## iq
                1
                   19584
                         19584
                                   0.961 0.3284
## Residuals 176 3588287
                           20388
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
Human interaction
summary(aov(Met.g$shannon ~ cohabYN + child.sum + groom + social.sum + cohabYN:social.sum, data = vars.
                     Df Sum Sq Mean Sq F value Pr(>F)
                      1 0.322 0.3219 1.751 0.18839
## cohabYN
## child.sum
                      1 0.032 0.0320 0.174 0.67750
## groom
                      1 0.102 0.1018 0.554 0.45840
                      1 0.077 0.0767
## social.sum
                                        0.417 0.51954
## cohabYN:social.sum 1 1.341 1.3408
                                        7.294 0.00797 **
## Residuals
                   115 21.140 0.1838
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 58 observations deleted due to missingness
summary(aov(Met.g$chao ~ cohabYN + child.sum + groom + social.sum + cohabYN:social.sum, data = vars.g))
                      Df Sum Sq Mean Sq F value
                                                Pr(>F)
## cohabYN
                      1
                          35424
                                  35424 2.291 0.132911
## child.sum
                          25037
                                  25037 1.619 0.205808
                      1
## groom
                            884
                                    884
                                        0.057 0.811443
                      1
## social.sum
                           2555
                                   2555
                                        0.165 0.685167
                      1
## cohabYN:social.sum 1 181452 181452 11.733 0.000852 ***
## Residuals
                    115 1778518
                                  15465
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 58 observations deleted due to missingness
summary(aov(Met.g$shannon ~ cohabYN, data = vars.g))
               Df Sum Sq Mean Sq F value Pr(>F)
## cohabYN
               1
                  1.53 1.5300
                                 8.021 0.00516 **
## Residuals
             177 33.76 0.1907
## ---
```

```
## Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
summary(aov(Met.g$chao ~ cohabYN, data = vars.g))
##
               Df Sum Sq Mean Sq F value Pr(>F)
## cohabYN
               1 160740 160740
                                  7.965 0.00531 **
## Residuals
              177 3571997
                            20181
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
summary(aov(Met.g[vars.g$cohabYN == "Y",]$shannon ~ social.sum, data = vars.g[vars.g$cohabYN == "Y",]))
              Df Sum Sq Mean Sq F value Pr(>F)
## social.sum
              1 0.285 0.2853 1.511 0.222
## Residuals
             99 18.695 0.1888
## 26 observations deleted due to missingness
summary(aov(Met.g[vars.g$cohabYN == "Y",]$chao ~ social.sum, data = vars.g[vars.g$cohabYN == "Y",]))
              Df Sum Sq Mean Sq F value Pr(>F)
##
## social.sum
              1 26420
                           26420
                                  1.137 0.289
## Residuals
              99 2301108
                           23244
## 26 observations deleted due to missingness
summary(aov(Met.g[vars.g$cohabYN == "N",]$shannon ~ social.sum, data = vars.g[vars.g$cohabYN == "N",]))
              Df Sum Sq Mean Sq F value Pr(>F)
## social.sum
              1 0.623 0.6227
                                  2.84 0.0995 .
## Residuals
              41 8.989 0.2193
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 9 observations deleted due to missingness
summary(aov(Met.g[vars.g$cohabYN == "N",]$chao ~ social.sum, data = vars.g[vars.g$cohabYN == "N",]))
              Df Sum Sq Mean Sq F value Pr(>F)
              1 64103
                          64103 3.972 0.0529 .
## social.sum
## Residuals
              41 661638
                          16138
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 9 observations deleted due to missingness
Home environment
summary(aov(Met.g$shannon ~ dog + cat + pet.other + clean.house, data = vars.g))
               Df Sum Sq Mean Sq F value Pr(>F)
## dog
               1 0.031 0.03098 0.138 0.710
                1 0.000 0.00003
                                  0.000 0.990
## cat
                                  0.267 0.606
                1 0.060 0.05983
## pet.other
## clean.house 1 0.191 0.19063
                                  0.852 0.358
## Residuals 130 29.086 0.22374
## 44 observations deleted due to missingness
summary(aov(Met.g$chao ~ dog + cat + pet.other + clean.house, data = vars.g))
##
               Df Sum Sq Mean Sq F value Pr(>F)
```

```
31337
                             31337
                                      1.353 0.247
## dog
                 1
                                      0.572 0.451
                             13239
## cat
                     13239
                 1
## pet.other
                      5914
                              5914
                                      0.255 0.614
                      1017
                              1017
                                      0.044 0.834
## clean.house
                 1
## Residuals
               130 3010549
                             23158
## 44 observations deleted due to missingness
```

Health

```
summary(aov(Met.g$shannon ~ bmi+srh11+walk.ave, data = vars.g))
               Df Sum Sq Mean Sq F value Pr(>F)
##
## bmi
                1 0.558 0.5581
                                   2.958 0.0874 .
## srh11
                1
                   0.645 0.6449
                                   3.418 0.0663 .
## walk.ave
                1 0.000 0.0004
                                   0.002 0.9652
## Residuals
              162 30.568 0.1887
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## 13 observations deleted due to missingness
summary(aov(Met.g$chao ~ bmi+srh11+walk.ave, data = vars.g))
##
                   Sum Sq Mean Sq F value Pr(>F)
## bmi
                     94485
                            94485
                                    4.445 0.0365 *
                1
                                    1.356 0.2459
                     28826
                             28826
## srh11
                 1
                                    0.199 0.6561
## walk.ave
                      4231
                             4231
                1
## Residuals
              162 3443638
                            21257
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 13 observations deleted due to missingness
summary(aov(Met.g$shannon ~ as.factor(hbs11)+ as.factor(hbp11)+ as.factor(heart11)+ as.factor(arth11)+
                       Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(hbs11)
                        1 2.195 2.1954 10.916 0.0012 **
## as.factor(hbp11)
                        1 0.360 0.3598
                                           1.789 0.1831
                        1 0.021 0.0211
## as.factor(heart11)
                                          0.105 0.7463
## as.factor(arth11)
                        1 0.027 0.0273
                                          0.136 0.7133
## as.factor(cancer11)
                        1 0.100 0.0998
                                           0.496 0.4823
## as.factor(stroke11)
                        1 0.065 0.0645
                                          0.321 0.5720
## as.factor(IBS)
                        1 0.041 0.0414
                                           0.206 0.6505
## Residuals
                       148 29.765 0.2011
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 23 observations deleted due to missingness
summary(aov(Met.g$chao ~ as.factor(hbs11)+ as.factor(hbp11)+ as.factor(heart11)+ as.factor(arth11)+ as.
##
                           Sum Sq Mean Sq F value Pr(>F)
## as.factor(hbs11)
                        1
                            92522
                                    92522
                                            4.198 0.0422 *
## as.factor(hbp11)
                            17153
                                    17153
                                            0.778 0.3791
                        1
## as.factor(heart11)
                        1
                              214
                                      214
                                            0.010 0.9216
## as.factor(arth11)
                        1
                               35
                                       35
                                            0.002 0.9683
## as.factor(cancer11)
                                    41512
                                            1.884 0.1720
                            41512
                        1
## as.factor(stroke11)
                            15939
                                    15939
                                            0.723 0.3964
```

```
summary(aov(Met.g$shannon ~ as.factor(smokeYN), data = vars.g))
                      Df Sum Sq Mean Sq F value Pr(>F)
                       1 0.035 0.0347 0.177 0.675
## as.factor(smokeYN)
                     152 29.874 0.1965
## Residuals
## 25 observations deleted due to missingness
summary(aov(Met.g$chao ~ as.factor(smokeYN), data = vars.g))
##
                      Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(smokeYN)
                           42360
                                   42360
                                          1.969 0.163
                       1
## Residuals
                     152 3269920
                                   21513
## 25 observations deleted due to missingness
```

Diet

```
summary(aov(Met.g$shannon ~ prot.sum + fruit.sum + veg.sum, data = vars.g))
               Df Sum Sq Mean Sq F value Pr(>F)
## prot.sum
                1 0.066 0.0657
                                  0.351 0.5546
                1 0.169 0.1687
                                  0.900 0.3442
## fruit.sum
                                  3.431 0.0658 .
## veg.sum
                1 0.643 0.6431
             162 30.364 0.1874
## Residuals
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 13 observations deleted due to missingness
summary(aov(Met.g$chao ~ prot.sum + fruit.sum + veg.sum, data = vars.g))
               Df Sum Sq Mean Sq F value Pr(>F)
                                  0.161 0.688
## prot.sum
                1
                     3412
                            3412
## fruit.sum
                    44307
                           44307
                                   2.095 0.150
                1
## veg.sum
                1
                    28453
                           28453
                                   1.345 0.248
              162 3426524
                           21151
## Residuals
## 13 observations deleted due to missingness
summary(aov(Met.g$shannon ~ meat+poul+pork+sea +meat:poul +poul:pork, data = vars.g))
               Df Sum Sq Mean Sq F value
                                          Pr(>F)
## meat
                1 0.196
                          0.196
                                  1.204
                                          0.2743
## poul
                1 0.056
                          0.056
                                  0.342
                                         0.5594
               1 0.090
                         0.090 0.552
## pork
                                         0.4588
               1 0.003
                         0.003 0.019
                                         0.8914
## sea
## meat:poul
               1 4.009
                          4.009 24.627 1.77e-06 ***
## poul:pork
              1 1.004
                          1.004
                                 6.165
                                        0.0141 *
## Residuals
             159 25.884
                         0.163
```

```
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 13 observations deleted due to missingness
summary(aov(Met.g$chao ~ meat+poul+pork+sea +meat:poul, data = vars.g))
##
                   Sum Sq Mean Sq F value Pr(>F)
## meat
                 1
                       624
                               624
                                     0.030 0.8636
## poul
                 1
                       487
                               487
                                     0.023 0.8794
                      1427
                                     0.068 0.7950
## pork
                 1
                              1427
## sea
                 1
                     29595
                             29595
                                     1.404 0.2378
## meat:poul
                 1
                     98236
                             98236
                                     4.661 0.0323 *
## Residuals
               160 3372326
                             21077
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 13 observations deleted due to missingness
```

FDR-correction

```
shannon.list = c(0.153, 0.798, 0.0055, 0.2075, 0.1355, 0.0919, 0.218, 0.8, 0.18839, 0.6775, 0.4584, 0.51954, 0.0079
 \text{chao.list} = c(0.0188, 0.8906, 0.0191, 0.122275, 0.000802, 0.038202, 0.0143, 0.3284, 0.132911, 0.205808, 0.811443, 0.12275, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.000802, 0.00080
p.adjust(shannon.list, method="BH")
          [1] 0.4707692 0.8421053 0.0550000 0.4933333 0.4516667 0.3618182 0.4933333
         [8] 0.8421053 0.4933333 0.8152000 0.7889655 0.7889655 0.0637600 0.0550000
## [15] 0.4933333 0.3618182 0.8152000 0.8492308 0.8152000 0.6761905 0.3618182
## [22] 0.3290000 0.7889655 0.0240000 0.4933333 0.8292222 0.8152000 0.7889655
## [29] 0.7889655 0.8152000 0.8152000 0.7889655 0.6761905 0.3290000 0.5774737
## [36] 0.7889655 0.7889655 0.8914000 0.0000708 0.0940000
p.adjust(chao.list, method="BH")
          [1] 0.1241500 0.9648167 0.1241500 0.3777429 0.0166140 0.1645800 0.1241500
          [8] 0.5336500 0.3777429 0.4396364 0.9648167 0.8944000 0.0166140 0.0690300
## [15] 0.4900435 0.1875545 0.4396364 0.6066667 0.7451786 0.9648167 0.1645800
## [22] 0.4396364 0.3777429 0.1645800 0.5913960 0.9714162 0.9721000 0.3945882
## [29] 0.5946000 0.9721000 0.3945882 0.8944000 0.3900000 0.4396364 0.9648167
## [36] 0.9648167 0.9648167 0.4396364 0.1645800
```

Spouses and siblings

Beta-diversity

Are spouses or siblings more similar? Compare beta-diveristy (Bray-Curtis, Jaccard, weighted and unweighted UniFrac) of spouses, siblings, and unrelated pairs. The unrelated group is defined as pairs that were not siblings, spouses, or in-laws within the dataset. The unrelated group was subsampled to 100 pairs with sex ratios equivalent to the sibling dataset (57% same sex pairs) and P-values were averaged across 1000 iterative subsamples

Since sample sizes and variance are unequal, compare groups by Kruskal-Wallis. Ties exist (values occur more than once in a group) so use Chi-squared method to correct within pairwise comparisons.

```
KW.subsample.fxn = function(rep.number)
{library(PMCMR)
  repeat{
  BC.MF.sample = match.BC.MF[sample(1:length(match.BC.MF), 43, replace=FALSE)]
  BC.MMFF.sample = match.BC.MMFF[sample(1:length(match.BC.MMFF), 57,
                                                                        replace=FALSE)]
  J.MF.sample = match.J.MF[sample(1:length(match.J.MF), 43, replace=FALSE)]
  J.MMFF.sample = match.J.MMFF[sample(1:length(match.J.MMFF), 57, replace=FALSE)]
  wUF.MF.sample = match.wUF.MF[sample(1:length(match.wUF.MF), 43, replace=FALSE)]
  wUF.MMFF.sample = match.wUF.MMFF[sample(1:length(match.wUF.MMFF), 57, replace=FALSE)]
  uwUF.MF.sample = match.uwUF.MF[sample(1:length(match.uwUF.MF), 43,
                                                                       replace=FALSE)]
  uwUF.MMFF.sample = match.uwUF.MMFF[sample(1:length(match.uwUF.MMFF), 57, replace=FALSE)]
  dist.sample = data.frame(BC=c(match.BC.gp.se, match.BC.gs, BC.MF.sample, BC.MMFF.sample),
                   J=c(match.J.gp.se, match.J.gs, J.MF.sample, J.MMFF.sample),
                   wUF=c(match.wUF.gp.se, match.wUF.gs, wUF.MF.sample, wUF.MMFF.sample),
                   uwUF=c(match.uwUF.gp.se, match.uwUF.gs, uwUF.MF.sample, uwUF.MMFF.sample),
                   comp=c(rep(times=(length(match.BC.gp.se)), "1.gpse"),
                               rep(times=(length(match.BC.gs)), "2.gs"),
                               rep(times=100, "3.other")))
#Bray-Curtis
BC.kw = kruskal.test(dist.sample$BC ~ dist.sample$comp)
posthoc.BC.kw.BH = p.adjust(posthoc.kruskal.nemenyi.test(x=dist.sample$BC, g=dist.sample$comp, dist="Ch
#Jaccard
J.kw = kruskal.test(dist.sample$J ~ dist.sample$comp)
posthoc.J.kw.BH = p.adjust(posthoc.kruskal.nemenyi.test(x=dist.sample$J, g=dist.sample$comp, dist="Chis
wUF.kw = kruskal.test(dist.sample$wUF ~ dist.sample$comp)
posthoc.wUF.kw.BH = p.adjust(posthoc.kruskal.nemenyi.test(x=dist.sample$wUF, g=dist.sample$comp, dist="
uwUF.kw = kruskal.test(dist.sample$uwUF ~ dist.sample$comp)
posthoc.uwUF.kw.BH = p.adjust(posthoc.kruskal.nemenyi.test(x=dist.sample$uwUF, g=dist.sample$comp, dist
#Print to table
kw.results = data.frame(
  BC = c(BC.MF.sample, BC.MMFF.sample, BC.kw$p.value, posthoc.BC.kw.BH),
  J = c(J.MF.sample, J.MMFF.sample, J.kw$p.value, posthoc.J.kw.BH),
  wUF = c(wUF.MF.sample, wUF.MMFF.sample, wUF.kw$p.value, posthoc.wUF.kw.BH),
  uwUF = c(uwUF.MF.sample, uwUF.MMFF.sample, uwUF.kw$p.value, posthoc.uwUF.kw.BH),
 label = c(rep("dist",100), "KW.P", "Sp.Sib", "Sp.other", "NA", "Sib.other"))
write.table(kw.results, file="KW.results.csv", append=TRUE, sep=",", col.names=FALSE)
KW.results.final = read.table("KW.results.csv", sep=",")
if(nrow(KW.results.final) >= 105*rep.number){break}
 }}
KW.subsample.fxn(1000)
#Curating results
KW.results.final = read.table("KW.results.csv", sep=",")
colnames(KW.results.final) = c("rep", "BC", "J", "wUF", "uwUF", "label")
#Average P-values
```

```
KW.P.ave = data.frame(
   KW.P = colMeans(KW.results.final[KW.results.final$label == "KW.P" & complete.cases(KW.results.final),
   Sp.Sib = colMeans(KW.results.final[KW.results.final$label == "Sp.Sib" & complete.cases(KW.results.fin
   Sp.other = colMeans(KW.results.final[KW.results.final$label == "Sp.other" & complete.cases(KW.results
   Sib.other = colMeans(KW.results.final[KW.results.final$label == "Sib.other"& complete.cases(KW.result))

write.table(KW.P.ave, file="KW.P.ave.csv", append=TRUE, sep=",", col.names=TRUE)

## Warning in write.table(KW.P.ave, file = "KW.P.ave.csv", append = TRUE, sep

## = ",", : appending column names to file
```

Use linear models to assess if beta-diversity among spouse and/or sibling pairs is explained by variables of interest. Variables found to impact the overall microbiota among graduates (PERMANOVA P < 0.1, Dataset S2), those with considerably more variation in the full dataset (age), and those specific to sibling/spouse relationships (kinship, years cohabitating, closeness) were tested.

Variables specified as diff are the difference between values for individuals within a pair. For example, a spousal pair of person 1 = 70 yrs old and person 2 = 75 yrs old would have an age.diff of 5 yrs. In this way, we are testing if pairs being discordant in one or more variables causes their microbiota to be more or less similar.

Variables in spouses and siblings

Closeness

Self-reported relationship closeness.

- 1 = not at all
- 2 = not very
- 3 = somewhat
- 4 = very

If individuals within a pair did not give the same answer, the lower of the two was used.

```
summary(aov(match.BC.gp.se.gs ~ as.factor(floor(vars.gp.se.gs$closeness))))
                                                {\tt Df \; Sum \; Sq \; Mean \; Sq \; F \; value}
                                                 3 0.0578 0.01927
## as.factor(floor(vars.gp.se.gs$closeness))
## Residuals
                                               173 2.5412 0.01469
##
                                               Pr(>F)
## as.factor(floor(vars.gp.se.gs$closeness)) 0.272
## Residuals
summary(aov(match.J.gp.se.gs ~ as.factor(floor(vars.gp.se.gs$closeness))))
##
                                                Df Sum Sq Mean Sq F value
## as.factor(floor(vars.gp.se.gs$closeness))
                                                 3 0.0352 0.011746
## Residuals
                                               173 1.3652 0.007891
                                               Pr(>F)
## as.factor(floor(vars.gp.se.gs$closeness)) 0.219
## Residuals
summary(aov(match.wUF.gp.se.gs ~ as.factor(floor(vars.gp.se.gs$closeness))))
                                                Df Sum Sq Mean Sq F value
                                                 3 0.0052 0.001729
## as.factor(floor(vars.gp.se.gs$closeness))
                                                                      0.481
## Residuals
                                               173 0.6213 0.003591
```

```
##
                                             Pr(>F)
## as.factor(floor(vars.gp.se.gs$closeness)) 0.696
summary(aov(match.uwUF.gp.se.gs ~ as.factor(floor(vars.gp.se.gs$closeness))))
                                              Df Sum Sq Mean Sq F value
## as.factor(floor(vars.gp.se.gs$closeness))
                                               3 0.0834 0.027787
                                             173 1.0438 0.006034
## Residuals
##
                                              Pr(>F)
## as.factor(floor(vars.gp.se.gs$closeness)) 0.00397 **
## Residuals
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
#Pairwise between closeness levels
pairwise.t.test(match.BC.gp.se.gs, as.factor(floor(vars.gp.se.gs$closeness)), p.adj = "BH")
##
##
  Pairwise comparisons using t tests with pooled SD
##
## data: match.BC.gp.se.gs and as.factor(floor(vars.gp.se.gs$closeness))
##
##
              3
## 2 0.69 -
## 3 0.73 0.69 -
## 4 0.69 0.84 0.47
## P value adjustment method: BH
pairwise.t.test(match.J.gp.se.gs, as.factor(floor(vars.gp.se.gs$closeness)), p.adj = "BH")
##
## Pairwise comparisons using t tests with pooled SD
##
## data: match.J.gp.se.gs and as.factor(floor(vars.gp.se.gs$closeness))
##
              3
##
    1
         2
## 2 0.53 -
## 3 0.74 0.53 -
## 4 0.53 0.95 0.37
## P value adjustment method: BH
pairwise.t.test(match.wUF.gp.se.gs, as.factor(floor(vars.gp.se.gs$closeness)), p.adj = "BH")
##
## Pairwise comparisons using t tests with pooled SD
##
## data: match.wUF.gp.se.gs and as.factor(floor(vars.gp.se.gs$closeness))
##
   1
         2
## 2 0.63 -
## 3 0.66 0.63 -
## 4 0.63 0.63 0.63
##
## P value adjustment method: BH
```

```
pairwise.t.test(match.uwUF.gp.se.gs, as.factor(floor(vars.gp.se.gs$closeness)), p.adj = "BH")
##
## Pairwise comparisons using t tests with pooled SD
##
## data: match.uwUF.gp.se.gs and as.factor(floor(vars.gp.se.gs$closeness))
##
##
     1
## 2 0.283 -
## 3 0.243 0.886 -
## 4 0.059 0.161 0.021
## P value adjustment method: BH
##Pairwise between closeness levels and spouses vs. siblings
vars.gp.se.gs$close.group = paste(as.factor(floor(vars.gp.se.gs$closeness)), vars.gp.se.gs$group, sep="
pairwise.t.test(match.BC.gp.se.gs, vars.gp.se.gs$close.group, p.adj = "BH")
##
## Pairwise comparisons using t tests with pooled SD
##
## data: match.BC.gp.se.gs and vars.gp.se.gs$close.group
##
##
           1.sib 2.sib 3.sib 3.spouse 4.sib
## 2.sib
           0.78 -
## 3.sib
           0.78 0.78 -
## 3.spouse 0.78 0.78 0.78
           0.78 0.78 0.95 0.78
## 4.sib
## 4.spouse 0.78 0.78 0.44 0.78
                                      0.78
## P value adjustment method: BH
pairwise.t.test(match.J.gp.se.gs, vars.gp.se.gs$close.group, p.adj = "BH")
##
## Pairwise comparisons using t tests with pooled SD
##
## data: match.J.gp.se.gs and vars.gp.se.gs$close.group
##
##
            1.sib 2.sib 3.sib 3.spouse 4.sib
## 2.sib
           0.83 -
## 3.sib
            0.83 0.83
## 3.spouse 0.83 0.83 0.83
           0.83 0.83 0.83 0.83
## 4.sib
## 4.spouse 0.83 0.83 0.38 0.83
                                      0.83
##
## P value adjustment method: BH
pairwise.t.test(match.wUF.gp.se.gs, vars.gp.se.gs$close.group, p.adj = "BH")
##
## Pairwise comparisons using t tests with pooled SD
## data: match.wUF.gp.se.gs and vars.gp.se.gs$close.group
##
```

```
## 2.sib
           0.74 -
## 3.sib
           0.82 0.74 -
## 3.spouse 0.82 0.82 -
## 4.sib
           0.94 0.74 0.74 0.74
## 4.spouse 0.74 0.82 0.74 0.82
                                      0.74
## P value adjustment method: BH
pairwise.t.test(match.uwUF.gp.se.gs, vars.gp.se.gs$close.group, p.adj = "BH")
##
## Pairwise comparisons using t tests with pooled SD
##
## data: match.uwUF.gp.se.gs and vars.gp.se.gs$close.group
##
##
           1.sib 2.sib 3.sib 3.spouse 4.sib
           0.370 -
## 2.sib
## 3.sib
           0.370 0.877 -
## 3.spouse 0.252 0.623 0.370 -
           0.328 0.815 0.698 0.713
## 4.sib
## 4.spouse 0.080 0.140 0.005 0.328
                                      0.140
## P value adjustment method: BH
Health
#Heart disease
summary(aov(match.BC.gp.se.gs ~ as.factor(vars.gp.se.gs$heart.diff.cat)))
##
                                           Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(vars.gp.se.gs$heart.diff.cat)
                                            1 0.0241 0.02411
                                                                 1.7 0.194
                                          155 2.1975 0.01418
## Residuals
## 20 observations deleted due to missingness
summary(aov(match.J.gp.se.gs ~ as.factor(vars.gp.se.gs$heart.diff.cat)))
                                           Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(vars.gp.se.gs$heart.diff.cat)
                                            1 0.0152 0.01518 2.006 0.159
                                          155 1.1734 0.00757
## 20 observations deleted due to missingness
summary(aov(match.wUF.gp.se.gs ~ as.factor(vars.gp.se.gs$heart.diff.cat)))
##
                                           Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(vars.gp.se.gs$heart.diff.cat)
                                           1 0.0065 0.006485
                                                                2.061 0.153
                                          155 0.4878 0.003147
## Residuals
## 20 observations deleted due to missingness
summary(aov(match.uwUF.gp.se.gs ~ as.factor(vars.gp.se.gs$heart.diff.cat)))
##
                                           Df Sum Sq Mean Sq F value Pr(>F)
                                            1 0.0037 0.003710
## as.factor(vars.gp.se.gs$heart.diff.cat)
                                                                0.628 0.429
                                          155 0.9154 0.005906
## Residuals
## 20 observations deleted due to missingness
```

1.sib 2.sib 3.sib 3.spouse 4.sib

```
#High blood sugar
summary(aov(match.BC.gp.se.gs ~ as.factor(vars.gp.se.gs$hbs.diff.cat)))
##
                                         Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(vars.gp.se.gs$hbs.diff.cat)
                                         1 0.0859 0.08594 6.081 0.0148 *
                                        150 2.1201 0.01413
## Residuals
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 25 observations deleted due to missingness
summary(aov(match.J.gp.se.gs ~ as.factor(vars.gp.se.gs$hbs.diff.cat)))
##
                                         Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(vars.gp.se.gs$hbs.diff.cat)
                                         1 0.0434 0.04344
                                                            5.731 0.0179 *
                                        150 1.1369 0.00758
## Residuals
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 25 observations deleted due to missingness
summary(aov(match.wUF.gp.se.gs ~ as.factor(vars.gp.se.gs$hbs.diff.cat)))
                                         Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(vars.gp.se.gs$hbs.diff.cat)
                                         1 0.0133 0.01334
                                                           4.195 0.0423 *
## Residuals
                                        150 0.4771 0.00318
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 25 observations deleted due to missingness
summary(aov(match.uwUF.gp.se.gs ~ as.factor(vars.gp.se.gs$hbs.diff.cat)))
##
                                         Df Sum Sq Mean Sq F value Pr(>F)
                                          1 0.0159 0.015867
                                                              2.662 0.105
## as.factor(vars.gp.se.gs$hbs.diff.cat)
## Residuals
                                        150 0.8942 0.005961
## 25 observations deleted due to missingness
Diet
#Distance in protein consumption
summary(lm(match.BC.gp.se.gs ~ match.prot.gp.se.gs))
##
## Call:
## lm(formula = match.BC.gp.se.gs ~ match.prot.gp.se.gs)
##
## Residuals:
       Min
                 1Q
                     Median
## -0.29716 -0.07575 -0.01612 0.08217 0.28846
## Coefficients:
##
                      Estimate Std. Error t value Pr(>|t|)
                      0.643932
                                 0.014968 43.021
                                                    <2e-16 ***
## (Intercept)
## match.prot.gp.se.gs 0.006465
                                 0.005869
                                            1.102
                                                     0.272
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.122 on 173 degrees of freedom
```

```
(2 observations deleted due to missingness)
## Multiple R-squared: 0.006966,
                                   Adjusted R-squared: 0.001226
## F-statistic: 1.214 on 1 and 173 DF, p-value: 0.2721
summary(lm(match.J.gp.se.gs ~ match.prot.gp.se.gs))
##
## Call:
## lm(formula = match.J.gp.se.gs ~ match.prot.gp.se.gs)
## Residuals:
##
        Min
                   1Q
                         Median
                                       30
                                                Max
## -0.259843 -0.051366 -0.005535 0.063490 0.185048
##
## Coefficients:
##
                      Estimate Std. Error t value Pr(>|t|)
## (Intercept)
                      0.777437
                                 0.010991 70.733
## match.prot.gp.se.gs 0.004467
                                 0.004309
                                            1.036
                                                     0.301
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## Residual standard error: 0.08958 on 173 degrees of freedom
     (2 observations deleted due to missingness)
## Multiple R-squared: 0.006171,
                                   Adjusted R-squared: 0.0004263
## F-statistic: 1.074 on 1 and 173 DF, p-value: 0.3014
summary(lm(match.wUF.gp.se.gs ~ match.prot.gp.se.gs))
##
## Call:
## lm(formula = match.wUF.gp.se.gs ~ match.prot.gp.se.gs)
## Residuals:
                         Median
##
        Min
                   1Q
                                       3Q
                                                Max
## -0.136859 -0.036391 -0.007626 0.032091 0.180736
##
## Coefficients:
                       Estimate Std. Error t value Pr(>|t|)
##
## (Intercept)
                      0.229411
                                 0.007156 32.057
                                                    <2e-16 ***
## match.prot.gp.se.gs 0.002438
                                 0.002806
                                            0.869
                                                     0.386
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.05832 on 173 degrees of freedom
     (2 observations deleted due to missingness)
## Multiple R-squared: 0.004344,
                                   Adjusted R-squared: -0.001412
## F-statistic: 0.7547 on 1 and 173 DF, p-value: 0.3862
summary(lm(match.uwUF.gp.se.gs ~ match.prot.gp.se.gs))
##
## Call:
## lm(formula = match.uwUF.gp.se.gs ~ match.prot.gp.se.gs)
## Residuals:
                   1Q
                         Median
                                       3Q
##
        Min
                                                Max
```

```
## -0.175107 -0.049752 -0.003172 0.049159 0.196814
##
## Coefficients:
                       Estimate Std. Error t value Pr(>|t|)
##
## (Intercept)
                       0.594967
                                 0.009619 61.855
## match.prot.gp.se.gs 0.009683
                                 0.003771
                                            2.567
                                                     0.0111 *
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.07839 on 173 degrees of freedom
     (2 observations deleted due to missingness)
## Multiple R-squared: 0.0367, Adjusted R-squared: 0.03113
## F-statistic: 6.592 on 1 and 173 DF, p-value: 0.01109
summary(lm(match.uwUF.gp.se.gs ~ match.prot.gp.se.gs))
##
## Call:
## lm(formula = match.uwUF.gp.se.gs ~ match.prot.gp.se.gs)
##
## Residuals:
##
        Min
                   1Q
                         Median
                                        3Q
                                                 Max
## -0.175107 -0.049752 -0.003172 0.049159 0.196814
##
## Coefficients:
                       Estimate Std. Error t value Pr(>|t|)
##
## (Intercept)
                       0.594967
                                 0.009619 61.855
                                                    <2e-16 ***
## match.prot.gp.se.gs 0.009683
                                 0.003771
                                            2.567
                                                     0.0111 *
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.07839 on 173 degrees of freedom
     (2 observations deleted due to missingness)
## Multiple R-squared: 0.0367, Adjusted R-squared: 0.03113
## F-statistic: 6.592 on 1 and 173 DF, p-value: 0.01109
#Specific protein sources
summary(lm(match.BC.gp.se.gs ~ vars.gp.se.gs$meat.diff*vars.gp.se.gs$poul.diff))
##
## Call:
## lm(formula = match.BC.gp.se.gs ~ vars.gp.se.gs$meat.diff * vars.gp.se.gs$poul.diff)
## Residuals:
##
                   10
                         Median
                                        30
## -0.289279 -0.071626 -0.006431 0.075048 0.285035
##
## Coefficients:
                                                    Estimate Std. Error
##
## (Intercept)
                                                    0.647063 0.014574
                                                              0.010337
## vars.gp.se.gs$meat.diff
                                                   -0.006476
## vars.gp.se.gs$poul.diff
                                                   -0.004548
                                                               0.013067
## vars.gp.se.gs$meat.diff:vars.gp.se.gs$poul.diff 0.015851
                                                               0.007321
##
                                                   t value Pr(>|t|)
## (Intercept)
                                                    44.398
                                                            <2e-16 ***
```

```
## vars.gp.se.gs$meat.diff
                                                   -0.627
                                                            0.5318
## vars.gp.se.gs$poul.diff
                                                   -0.348
                                                            0.7282
## vars.gp.se.gs$meat.diff:vars.gp.se.gs$poul.diff 2.165
                                                            0.0318 *
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.1191 on 169 degrees of freedom
     (4 observations deleted due to missingness)
## Multiple R-squared: 0.05315,
                                   Adjusted R-squared: 0.03634
## F-statistic: 3.162 on 3 and 169 DF, \, p-value: 0.02606
summary(lm(match.J.gp.se.gs ~ vars.gp.se.gs$meat.diff*vars.gp.se.gs$poul.diff))
##
## Call:
## lm(formula = match.J.gp.se.gs ~ vars.gp.se.gs$meat.diff * vars.gp.se.gs$poul.diff)
## Residuals:
                         Median
        Min
                   1Q
                                       3Q
## -0.254621 -0.046544 0.001272 0.059397 0.182297
## Coefficients:
##
                                                   Estimate Std. Error
## (Intercept)
                                                   0.779554 0.010741
## vars.gp.se.gs$meat.diff
                                                  -0.004700 0.007619
## vars.gp.se.gs$poul.diff
                                                  -0.002873
                                                              0.009630
## vars.gp.se.gs$meat.diff:vars.gp.se.gs$poul.diff 0.011010
                                                              0.005396
                                                  t value Pr(>|t|)
                                                   72.576
                                                            <2e-16 ***
## (Intercept)
## vars.gp.se.gs$meat.diff
                                                   -0.617
                                                            0.5381
## vars.gp.se.gs$poul.diff
                                                   -0.298
                                                            0.7658
## vars.gp.se.gs$meat.diff:vars.gp.se.gs$poul.diff
                                                   2.041
                                                            0.0428 *
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.08781 on 169 degrees of freedom
     (4 observations deleted due to missingness)
## Multiple R-squared: 0.04762,
                                   Adjusted R-squared: 0.03071
## F-statistic: 2.817 on 3 and 169 DF, p-value: 0.04075
summary(lm(match.wUF.gp.se.gs ~ vars.gp.se.gs$meat.diff*vars.gp.se.gs$poul.diff))
##
## Call:
## lm(formula = match.wUF.gp.se.gs ~ vars.gp.se.gs$meat.diff * vars.gp.se.gs$poul.diff)
## Residuals:
                         Median
                   1Q
                                       3Q
## -0.129991 -0.033924 -0.005739 0.037445 0.182249
## Coefficients:
##
                                                   Estimate Std. Error
## (Intercept)
                                                   0.231814 0.006991
## vars.gp.se.gs$meat.diff
                                                  -0.002896 0.004958
                                                  -0.006832 0.006268
## vars.gp.se.gs$poul.diff
```

```
## vars.gp.se.gs$meat.diff:vars.gp.se.gs$poul.diff 0.009259 0.003511
##
                                                  t value Pr(>|t|)
## (Intercept)
                                                   33.161 < 2e-16 ***
## vars.gp.se.gs$meat.diff
                                                   -0.584 0.55997
## vars.gp.se.gs$poul.diff
                                                   -1.090 0.27724
## vars.gp.se.gs$meat.diff:vars.gp.se.gs$poul.diff
                                                   2.637 0.00915 **
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.05715 on 169 degrees of freedom
     (4 observations deleted due to missingness)
## Multiple R-squared: 0.06547,
                                   Adjusted R-squared: 0.04888
## F-statistic: 3.946 on 3 and 169 DF, p-value: 0.009409
summary(lm(match.uwUF.gp.se.gs ~ vars.gp.se.gs$meat.diff*vars.gp.se.gs$poul.diff))
##
## Call:
## lm(formula = match.uwUF.gp.se.gs ~ vars.gp.se.gs$meat.diff *
      vars.gp.se.gs$poul.diff)
##
##
## Residuals:
                   10
                         Median
                                       30
## -0.175056 -0.046957 -0.003657 0.051325 0.186175
## Coefficients:
##
                                                  Estimate Std. Error
## (Intercept)
                                                  0.596806 0.009381
## vars.gp.se.gs$meat.diff
                                                  0.001576 0.006653
## vars.gp.se.gs$poul.diff
                                                  0.008810
                                                             0.008410
## vars.gp.se.gs$meat.diff:vars.gp.se.gs$poul.diff 0.006660
                                                             0.004712
##
                                                  t value Pr(>|t|)
## (Intercept)
                                                   63.621
                                                            <2e-16 ***
## vars.gp.se.gs$meat.diff
                                                    0.237
                                                             0.813
                                                             0.296
## vars.gp.se.gs$poul.diff
                                                    1.048
## vars.gp.se.gs$meat.diff:vars.gp.se.gs$poul.diff
                                                    1.413
                                                             0.159
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## Residual standard error: 0.07669 on 169 degrees of freedom
    (4 observations deleted due to missingness)
## Multiple R-squared: 0.08156,
                                   Adjusted R-squared: 0.06526
## F-statistic: 5.002 on 3 and 169 DF, p-value: 0.002386
```

Variables only in spouses

Rural vs. urban

Not tested for siblings as all pairs resided together as children

```
## 29 observations deleted due to missingness
summary(aov(match.J.gp.se ~ vars.gp.se.all$RU.diff.cat))
##
                             Df Sum Sq Mean Sq F value Pr(>F)
## vars.gp.se.all$RU.diff.cat 1 0.0033 0.003346
                                                  0.48 0.491
## Residuals
                             63 0.4387 0.006964
## 29 observations deleted due to missingness
summary(aov(match.wUF.gp.se ~ vars.gp.se.all$RU.diff.cat))
                             Df Sum Sq Mean Sq F value Pr(>F)
## vars.gp.se.all$RU.diff.cat 1 0.00245 0.002450
                                                  1.005
## Residuals
                             63 0.15363 0.002439
## 29 observations deleted due to missingness
summary(aov(match.uwUF.gp.se ~ vars.gp.se.all$RU.diff.cat))
                            Df Sum Sq Mean Sq F value Pr(>F)
## vars.gp.se.all$RU.diff.cat 1 0.0049 0.004942
                                                 0.809 0.372
## Residuals
                             63 0.3847 0.006106
## 29 observations deleted due to missingness
Years cohabiting
summary(lm(match.BC.gp.se ~ vars.gp.se.all$yrs.cohab.marr))
##
## Call:
## lm(formula = match.BC.gp.se ~ vars.gp.se.all$yrs.cohab.marr)
## Residuals:
##
       Min
                 1Q Median
                                  3Q
## -0.27365 -0.07187 -0.01203 0.06711 0.29172
##
## Coefficients:
##
                                 Estimate Std. Error t value Pr(>|t|)
## (Intercept)
                                 ## vars.gp.se.all$yrs.cohab.marr -0.001267
                                           0.001110 -1.141
                                                               0.257
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## Residual standard error: 0.1151 on 92 degrees of freedom
## Multiple R-squared: 0.01396,
                                  Adjusted R-squared: 0.003244
## F-statistic: 1.303 on 1 and 92 DF, p-value: 0.2567
summary(lm(match.J.gp.se ~ vars.gp.se.all$yrs.cohab.marr))
##
## Call:
## lm(formula = match.J.gp.se ~ vars.gp.se.all$yrs.cohab.marr)
## Residuals:
        Min
                         Median
                   1Q
                                      30
                                               Max
## -0.241879 -0.050109 -0.003267 0.054601 0.192958
##
## Coefficients:
```

```
##
                                 Estimate Std. Error t value Pr(>|t|)
                                0.8184200 0.0373019 21.940 <2e-16 ***
## (Intercept)
## vars.gp.se.all$yrs.cohab.marr -0.0010279 0.0008314 -1.236
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.08623 on 92 degrees of freedom
## Multiple R-squared: 0.01635,
                                Adjusted R-squared: 0.005654
## F-statistic: 1.529 on 1 and 92 DF, p-value: 0.2194
summary(lm(match.wUF.gp.se ~ vars.gp.se.all$yrs.cohab.marr))
##
## Call:
## lm(formula = match.wUF.gp.se ~ vars.gp.se.all$yrs.cohab.marr)
## Residuals:
##
                        Median
        Min
                  1Q
                                     3Q
## -0.131040 -0.029903 -0.003345 0.025035 0.185109
## Coefficients:
##
                                 Estimate Std. Error t value Pr(>|t|)
## (Intercept)
                                ## vars.gp.se.all$yrs.cohab.marr -0.0002728  0.0005496  -0.496
                                                              0.621
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## Residual standard error: 0.05701 on 92 degrees of freedom
## Multiple R-squared: 0.00267, Adjusted R-squared: -0.00817
## F-statistic: 0.2463 on 1 and 92 DF, p-value: 0.6209
summary(lm(match.uwUF.gp.se ~ vars.gp.se.all$yrs.cohab.marr))
##
## Call:
## lm(formula = match.uwUF.gp.se ~ vars.gp.se.all$yrs.cohab.marr)
## Residuals:
                1Q Median
       Min
                                  3Q
## -0.15923 -0.05140 -0.01736 0.05751 0.18949
##
## Coefficients:
##
                                 Estimate Std. Error t value Pr(>|t|)
## (Intercept)
                                0.6607260 0.0348422 18.963 <2e-16 ***
## vars.gp.se.all$yrs.cohab.marr -0.0015277 0.0007765 -1.967 0.0522 .
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## Residual standard error: 0.08054 on 92 degrees of freedom
## Multiple R-squared: 0.04037,
                                Adjusted R-squared: 0.02994
## F-statistic: 3.87 on 1 and 92 DF, p-value: 0.05216
```

Closeness

Test if closeness results hold true across just spouses

```
summary(lm(match.BC.gp.se ~ as.factor(floor(vars.gp.se.all$closeness))))
##
## Call:
## lm(formula = match.BC.gp.se ~ as.factor(floor(vars.gp.se.all$closeness)))
## Residuals:
##
        Min
                   1Q
                         Median
                                       3Q
## -0.279084 -0.073207 -0.008726 0.076662 0.289892
## Coefficients:
                                              Estimate Std. Error t value
##
## (Intercept)
                                               0.66454
                                                          0.02643 25.145
## as.factor(floor(vars.gp.se.all$closeness))4 -0.03222
                                                          0.02959 -1.089
                                              Pr(>|t|)
## (Intercept)
                                                 <2e-16 ***
## as.factor(floor(vars.gp.se.all$closeness))4
                                                 0.279
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## Residual standard error: 0.1152 on 92 degrees of freedom
## Multiple R-squared: 0.01272,
                                   Adjusted R-squared: 0.001993
## F-statistic: 1.186 on 1 and 92 DF, p-value: 0.279
summary(lm(match.J.gp.se ~ as.factor(floor(vars.gp.se.all$closeness))))
##
## Call:
## lm(formula = match.J.gp.se ~ as.factor(floor(vars.gp.se.all$closeness)))
## Residuals:
        Min
                   1Q
                         Median
                                       30
## -0.246560 -0.050489 -0.000864 0.061090 0.190911
##
## Coefficients:
                                              Estimate Std. Error t value
##
## (Intercept)
                                               0.79339
                                                          0.01981 40.048
## as.factor(floor(vars.gp.se.all$closeness))4 -0.02477
                                                          0.02218 -1.117
                                              Pr(>|t|)
## (Intercept)
                                                 <2e-16 ***
## as.factor(floor(vars.gp.se.all$closeness))4
                                                 0.267
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## Residual standard error: 0.08636 on 92 degrees of freedom
## Multiple R-squared: 0.01338,
                                  Adjusted R-squared: 0.002653
## F-statistic: 1.247 on 1 and 92 DF, p-value: 0.267
summary(lm(match.wUF.gp.se ~ as.factor(floor(vars.gp.se.all$closeness))))
##
## lm(formula = match.wUF.gp.se ~ as.factor(floor(vars.gp.se.all$closeness)))
##
## Residuals:
```

```
Median
                   1Q
                                       3Q
## -0.132328 -0.031450 -0.007465 0.029408 0.186277
##
## Coefficients:
##
                                               Estimate Std. Error t value
                                                          0.013082 17.862
## (Intercept)
                                               0.233670
## as.factor(floor(vars.gp.se.all$closeness))4 -0.006352
                                                          0.014646 - 0.434
                                              Pr(>|t|)
## (Intercept)
                                                <2e-16 ***
## as.factor(floor(vars.gp.se.all$closeness))4
                                                 0.666
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## Residual standard error: 0.05702 on 92 degrees of freedom
## Multiple R-squared: 0.00204,
                                   Adjusted R-squared:
## F-statistic: 0.1881 on 1 and 92 DF, p-value: 0.6655
summary(lm(match.uwUF.gp.se ~ as.factor(floor(vars.gp.se.all$closeness))))
##
## Call:
## lm(formula = match.uwUF.gp.se ~ as.factor(floor(vars.gp.se.all$closeness)))
## Residuals:
       Min
                 1Q Median
                                   30
## -0.15874 -0.05017 -0.01153 0.05363 0.19457
##
## Coefficients:
                                              Estimate Std. Error t value
##
## (Intercept)
                                                          0.01868 33.023
                                               0.61675
## as.factor(floor(vars.gp.se.all$closeness))4 -0.02832
                                                          0.02091 -1.354
                                              Pr(>|t|)
## (Intercept)
                                                <2e-16 ***
## as.factor(floor(vars.gp.se.all$closeness))4
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## Residual standard error: 0.08141 on 92 degrees of freedom
## Multiple R-squared: 0.01955,
                                   Adjusted R-squared: 0.008888
## F-statistic: 1.834 on 1 and 92 DF, p-value: 0.179
```

Variables only in siblings

Sex

Not tested for spouses because all spousal pairs were male-female

```
## as.factor(vars.gs.all$gender.diff.cat) 1 0.0002 0.000221
                                          81 0.6712 0.008287
## Residuals
summary(aov(match.wUF.gs ~ as.factor(vars.gs.all$gender.diff.cat)))
                                          Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(vars.gs.all$gender.diff.cat) 1 0.0001 0.000059
                                                             0.015 0.902
## Residuals
                                          81 0.3177 0.003923
summary(aov(match.uwUF.gs ~ as.factor(vars.gs.all$gender.diff.cat)))
                                          Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(vars.gs.all$gender.diff.cat) 1 0.0013 0.001326
                                                               0.261 0.611
## Residuals
                                          81 0.4114 0.005079
Genetics
Genetic relatedness defined by a kinship score.
#Subset data to those with kinship scores available
match.BC.gs.relate = subset(match.BC.gs, names(match.BC.gs) %in% row.names(relatedness.gs.match))
match.J.gs.relate = subset(match.J.gs, names(match.J.gs) %in% row.names(relatedness.gs.match))
match.wUF.gs.relate = subset(match.wUF.gs, names(match.wUF.gs) %in% row.names(relatedness.gs.match))
match.uwUF.gs.relate = subset(match.uwUF.gs, names(match.uwUF.gs) %in% row.names(relatedness.gs.match))
summary(aov(match.BC.gs.relate ~ relatedness.gs.match$kinship))
                                Df Sum Sq Mean Sq F value Pr(>F)
## relatedness.gs.match$kinship 1 0.0063 0.006294
                                                      0.43 0.514
                                74 1.0823 0.014626
## Residuals
summary(aov(match.J.gs.relate ~ relatedness.gs.match$kinship))
                                Df Sum Sq Mean Sq F value Pr(>F)
## relatedness.gs.match$kinship 1 0.0050 0.004982
                                                     0.674 0.414
                                74 0.5467 0.007388
## Residuals
summary(aov(match.wUF.gs.relate ~ relatedness.gs.match$kinship))
                                Df Sum Sq Mean Sq F value Pr(>F)
## relatedness.gs.match$kinship 1 0.0006 0.000603
                                                    0.157 0.693
                                74 0.2848 0.003848
## Residuals
summary(aov(match.uwUF.gs.relate ~ relatedness.gs.match$kinship))
                                Df Sum Sq Mean Sq F value Pr(>F)
## relatedness.gs.match$kinship 1 0.0001 0.00005
                                                   0.011 0.917
## Residuals
                                74 0.3412 0.00461
Rural vs. urban
Not tested for spouses as all resided together at time of sampling.
summary(aov(match.BC.gs ~ as.factor(vars.gs.all$RU11.diff)))
##
                                    Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(vars.gs.all$RU11.diff) 1 0.0150 0.01496
                                                         1.25 0.271
                                    35 0.4189 0.01197
## Residuals
```

46 observations deleted due to missingness

```
summary(aov(match.J.gs ~ as.factor(vars.gs.all$RU11.diff)))
                                   Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(vars.gs.all$RU11.diff) 1 0.00822 0.008223
                                                        1.345 0.254
## Residuals
                                   35 0.21401 0.006115
## 46 observations deleted due to missingness
summary(aov(match.wUF.gs ~ as.factor(vars.gs.all$RU11.diff)))
                                   Df Sum Sq Mean Sq F value Pr(>F)
##
## as.factor(vars.gs.all$RU11.diff) 1 0.00082 0.000817
                                                          0.22 0.642
## Residuals
                                   35 0.13019 0.003720
## 46 observations deleted due to missingness
summary(aov(match.uwUF.gs ~ as.factor(vars.gs.all$RU11.diff)))
##
                                   Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(vars.gs.all$RU11.diff) 1 0.00345 0.003446
                                                         0.682 0.415
## Residuals
                                   35 0.17692 0.005055
## 46 observations deleted due to missingness
CohabYN
Cohabiting with a spouse or not
summary(aov(match.BC.gs ~ as.factor(vars.gs.all$cohab.diff.cat)))
                                        Df Sum Sq Mean Sq F value Pr(>F)
                                        1 0.0546 0.05463
## as.factor(vars.gs.all$cohab.diff.cat)
                                                            3.565 0.0626 .
## Residuals
                                        81 1.2410 0.01532
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
summary(aov(match.J.gs ~ as.factor(vars.gs.all$cohab.diff.cat)))
                                        Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(vars.gs.all$cohab.diff.cat) 1 0.0304 0.030437
                                                             3.846 0.0533 .
## Residuals
                                        81 0.6410 0.007914
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
summary(aov(match.wUF.gs ~ as.factor(vars.gs.all$cohab.diff.cat)))
                                        Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(vars.gs.all$cohab.diff.cat) 1 0.01185 0.011846
                                                            3.136 0.0803 .
## Residuals
                                        81 0.30594 0.003777
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
summary(aov(match.uwUF.gs ~ as.factor(vars.gs.all$cohab.diff.cat)))
##
                                        Df Sum Sq Mean Sq F value Pr(>F)
## as.factor(vars.gs.all$cohab.diff.cat) 1 0.0092 0.009176
                                                            1.842 0.179
                                        81 0.4035 0.004982
## Residuals
```

Socialness

```
summary(lm(match.BC.gs ~ vars.gs.all$social.sum.diff))
##
## Call:
## lm(formula = match.BC.gs ~ vars.gs.all$social.sum.diff)
## Residuals:
##
        Min
                   1Q
                         Median
## -0.303079 -0.064872 -0.007305 0.091746 0.263556
##
## Coefficients:
                               Estimate Std. Error t value Pr(>|t|)
##
                                          0.022991 30.008
                               0.689907
                                                             <2e-16 ***
## (Intercept)
## vars.gs.all$social.sum.diff -0.004186 0.002636 -1.588
                                                              0.118
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.1232 on 51 degrees of freedom
    (30 observations deleted due to missingness)
## Multiple R-squared: 0.04713,
                                  Adjusted R-squared: 0.02845
## F-statistic: 2.523 on 1 and 51 DF, p-value: 0.1184
summary(lm(match.J.gs ~ vars.gs.all$social.sum.diff))
##
## Call:
## lm(formula = match.J.gs ~ vars.gs.all$social.sum.diff)
## Residuals:
        Min
                   10
                         Median
                                       30
## -0.250890 -0.041559 0.002072 0.068782 0.172481
##
## Coefficients:
##
                               Estimate Std. Error t value Pr(>|t|)
                               0.808751 0.016916 47.809
## (Intercept)
                                                             <2e-16 ***
## vars.gs.all$social.sum.diff -0.002799 0.001939 -1.443
                                                              0.155
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.09064 on 51 degrees of freedom
    (30 observations deleted due to missingness)
## Multiple R-squared: 0.03924,
                                  Adjusted R-squared: 0.0204
## F-statistic: 2.083 on 1 and 51 DF, p-value: 0.1551
summary(lm(match.wUF.gs ~ vars.gs.all$social.sum.diff))
##
## Call:
## lm(formula = match.wUF.gs ~ vars.gs.all$social.sum.diff)
##
## Residuals:
        Min
                   1Q
                         Median
                                       3Q
## -0.097302 -0.038009 -0.006541 0.031277 0.145307
##
## Coefficients:
```

```
##
                               Estimate Std. Error t value Pr(>|t|)
## (Intercept)
                                         0.009722 24.915
                               0.242229
                                                           <2e-16 ***
## vars.gs.all$social.sum.diff -0.001621 0.001115 -1.454
                                                             0.152
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.05209 on 51 degrees of freedom
     (30 observations deleted due to missingness)
## Multiple R-squared: 0.03982,
                                  Adjusted R-squared: 0.02099
## F-statistic: 2.115 on 1 and 51 DF, p-value: 0.152
summary(lm(match.uwUF.gs ~ vars.gs.all$social.sum.diff))
##
## Call:
## lm(formula = match.uwUF.gs ~ vars.gs.all$social.sum.diff)
## Residuals:
                         Median
        Min
                   1Q
                                       3Q
                                               Max
## -0.186028 -0.043849 -0.000991 0.032925 0.162362
## Coefficients:
##
                                Estimate Std. Error t value Pr(>|t|)
## (Intercept)
                               ## vars.gs.all$social.sum.diff -0.0007563 0.0014941 -0.506
                                                              0.615
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## Residual standard error: 0.06983 on 51 degrees of freedom
    (30 observations deleted due to missingness)
## Multiple R-squared: 0.005, Adjusted R-squared: -0.01451
## F-statistic: 0.2563 on 1 and 51 DF, p-value: 0.6149
Closeness
Test if closeness results hold true across just siblings
summary(aov(match.BC.gs ~ as.factor(floor(vars.gs.all$closeness.ave))))
##
                                             Df Sum Sq Mean Sq F value
## as.factor(floor(vars.gs.all$closeness.ave))
                                             3 0.0152 0.005065
## Residuals
                                             79 1.2805 0.016208
                                             Pr(>F)
## as.factor(floor(vars.gs.all$closeness.ave))    0.816
## Residuals
summary(aov(match.J.gs ~ as.factor(floor(vars.gs.all$closeness.ave))))
                                             Df Sum Sq Mean Sq F value
## as.factor(floor(vars.gs.all$closeness.ave)) 3 0.0109 0.003645
                                                                  0.436
## Residuals
                                             79 0.6605 0.008361
                                             Pr(>F)
## as.factor(floor(vars.gs.all$closeness.ave)) 0.728
## Residuals
```

```
summary(aov(match.wUF.gs ~ as.factor(floor(vars.gs.all$closeness.ave))))
                                                                                                                                                                                             Df Sum Sq Mean Sq F value
## as.factor(floor(vars.gs.all$closeness.ave))
                                                                                                                                                                                               3 0.00952 0.003174
## Residuals
                                                                                                                                                                                              79 0.30826 0.003902
                                                                                                                                                                                              Pr(>F)
## as.factor(floor(vars.gs.all$closeness.ave))
                                                                                                                                                                                                     0.49
## Residuals
summary(aov(match.uwUF.gs ~ as.factor(floor(vars.gs.all$closeness.ave))))
                                                                                                                                                                                             Df Sum Sq Mean Sq F value
## as.factor(floor(vars.gs.all$closeness.ave)) 3 0.0126 0.004212
## Residuals
                                                                                                                                                                                             79 0.4001 0.005064
                                                                                                                                                                                             Pr(>F)
## as.factor(floor(vars.gs.all$closeness.ave))
## Residuals
FDR-correction
BC.sib.spouse.list = c(0.554, 0.257, 0.869, 0.514, 0.271, 0.0626, 0.118, 0.0319, 0.544, 0.272, 0.194, 0.0148, 0.272, 0.194, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 0.0148, 
J.sib.spouse.list = c(0.491, 0.219, 0.871, 0.414, 0.254, 0.0533, 0.155, 0.0305, 0.519, 0.219, 0.159, 0.0179, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.301, 0.3
wUF.sib.spouse.list = c(0.338, 0.657, 0.901, 0.829, 0.832, 0.0717, 0.176, 0.0562, 0.962, 0.754, 0.147, 0.052, 0.672
uwUF.sib.spouse.list = c(0.361, 0.0512, 0.609, 0.911, 0.427, 0.177, 0.61, 0.0266, 0.731, 0.00405, 0.424, 0.106, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.00405, 0.0040
p.adjust(BC.sib.spouse.list, method="BH")
## [1] 0.6331429 0.4352000 0.8690000 0.6331429 0.4352000 0.2504000 0.3776000
## [8] 0.1701333 0.6331429 0.4352000 0.4352000 0.1701333 0.4352000 0.6331429
## [15] 0.7767467 0.1701333
p.adjust(J.sib.spouse.list, method="BH")
## [1] 0.6149714 0.4380000 0.8710000 0.6021818 0.4515556 0.2132000 0.4240000
## [8] 0.2132000 0.6149714 0.4380000 0.4240000 0.2132000 0.4816000 0.6149714
## [15] 0.8168533 0.2132000
p.adjust(wUF.sib.spouse.list, method="BH")
               [1] 0.7608000 0.9508571 0.9610667 0.9508571 0.9508571 0.2868000 0.4693333
## [8] 0.2868000 0.9620000 0.9508571 0.4693333 0.2868000 0.9508571 0.9508571
## [15] 0.7608000 0.2868000
p.adjust(uwUF.sib.spouse.list, method="BH")
                [1] 0.6210909 0.2048000 0.7507692 0.9110000 0.6210909 0.4045714 0.7507692
## [8] 0.1418667 0.8354286 0.0648000 0.6210909 0.3392000 0.0952000 0.8672000
## [15] 0.6040000 0.4045714
Interactions
Do variables that explain beta-diversity differ between spouse and sibling groups?
```

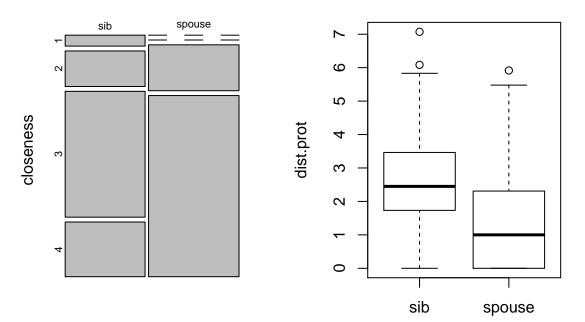
chisq.test(table(vars.gp.se.gs\$group, as.factor(vars.gp.se.gs\$AB.diff.cat)))

```
##
## Pearson's Chi-squared test with Yates' continuity correction
## data: table(vars.gp.se.gs$group, as.factor(vars.gp.se.gs$AB.diff.cat))
## X-squared = 1.7011, df = 1, p-value = 0.1921
#Closeness
chisq.test(table(vars.gp.se.gs$group, as.factor(floor(vars.gp.se.gs$closeness))))
## Warning in chisq.test(table(vars.gp.se.gs$group,
## as.factor(floor(vars.gp.se.gs$closeness)))): Chi-squared approximation may
## be incorrect
##
## Pearson's Chi-squared test
## data: table(vars.gp.se.gs$group, as.factor(floor(vars.gp.se.gs$closeness)))
## X-squared = 59.604, df = 3, p-value = 7.142e-13
#heart.diff
chisq.test(table(vars.gp.se.gs$group, as.factor(vars.gp.se.gs$heart.diff.cat)))
##
## Pearson's Chi-squared test with Yates' continuity correction
## data: table(vars.gp.se.gs$group, as.factor(vars.gp.se.gs$heart.diff.cat))
## X-squared = 0.065908, df = 1, p-value = 0.7974
#hbs.diff
chisq.test(table(vars.gp.se.gs$group, as.factor(vars.gp.se.gs$hbs.diff.cat)))
##
## Pearson's Chi-squared test with Yates' continuity correction
## data: table(vars.gp.se.gs$group, as.factor(vars.gp.se.gs$hbs.diff.cat))
## X-squared = 0, df = 1, p-value = 1
#dist.prot
summary(aov(match.prot.gp.se.gs ~ vars.gp.se.gs$group))
                       Df Sum Sq Mean Sq F value
                                                  Pr(>F)
## vars.gp.se.gs$group
                        1
                            84.5
                                   84.48
                                           42.05 8.99e-10 ***
## Residuals
                      173 347.6
                                    2.01
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 2 observations deleted due to missingness
#meat.diff:poul.diff
summary(aov(vars.gp.se.gs$meat.diff ~ vars.gp.se.gs$group))
                       Df Sum Sq Mean Sq F value
                                                  Pr(>F)
                                           29.38 1.99e-07 ***
## vars.gp.se.gs$group
                       1 40.28
                                   40.28
                      172 235.86
                                    1.37
## Residuals
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 3 observations deleted due to missingness
summary(aov(vars.gp.se.gs$poul.diff ~ vars.gp.se.gs$group))
```

```
##
                        Df Sum Sq Mean Sq F value
## vars.gp.se.gs$group
                         1 16.21 16.214
                                             19.71 1.61e-05 ***
## Residuals
                       172 141.49
                                    0.823
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## 3 observations deleted due to missingness
FDR-correction
p.adjust(c(0.1921,7.14E-13,0.7974,1,8.99E-10,1.99E-07,1.61E-05), method="BH")
## [1] 2.689400e-01 4.998000e-12 9.303000e-01 1.000000e+00 3.146500e-09
## [6] 4.643333e-07 2.817500e-05
It appears that spouses tend to have closer relationships and more similar diets than siblings.
#Plots
par(mfrow = c(1, 2))
plot(table(vars.gp.se.gs\$group, as.factor(floor(vars.gp.se.gs\$closeness))), ylab="closeness")
```

e.gs\$group, as.factor(floor(vars.gp.

boxplot(match.prot.gp.se.gs ~ vars.gp.se.gs\$group, ylab="dist.prot")



OTUs and health

OTUs commonly shared between siblings or spouses (> 50% of pairs) and rare in the overall dataset (< 70% of samples, < 49% of unrelated pairs) were correlated to health outcomes in the whole dataset by Kruskal-Wallis with the Benjamini-Hochberg correction for multiple comparisons.

We will co-op the kruskal.pretty function to run these tests by creating a fake SIMPER output. We will also need to load our unaltered .cons.taxonomy table from mothur as the function cannot take the altered version Tax we have used those far.

#Fake SIMPER output to get OTU list for health analyses

```
fake.simper = data.frame(
  X = c(1:22),
  Comparison = rep("1_2", 22),
 SIMPER = rep(0.001, 22),
  OTU = c("0tu00027", "0tu00035", "0tu00043", "0tu00007", "0tu00019", "0tu00025", "0tu00047", "0tu00004", "0tu0
#Unaltered mothur taxonomy table
Tax.orig = read.table("WLS.final.an.unique_list.0.02.cons.taxonomy", header=TRUE, row.names=1, sep="\t"
We then run Kruskal-Wallis tests for our health variables with NAs removed.
kruskal.pretty(OTU[!is.na(Pheno.sm$hbp11),], Pheno.sm[!is.na(Pheno.sm$hbp11),], fake.simper, c("hbp11")
##
## Attaching package: 'dplyr'
## The following object is masked from 'package:gridExtra':
##
##
       combine
## The following objects are masked from 'package:stats':
##
##
       filter, lag
## The following objects are masked from 'package:base':
##
##
       intersect, setdiff, setequal, union
kruskal.pretty(OTU[!is.na(Pheno.sm$hbs11),], Pheno.sm[!is.na(Pheno.sm$hbs11),], fake.simper, c("hbs11")
kruskal.pretty(OTU[!is.na(Pheno.sm$heart11),], Pheno.sm[!is.na(Pheno.sm$heart11),], fake.simper, c("hea
kruskal.pretty(OTU[!is.na(Pheno.sm$arth11),], Pheno.sm[!is.na(Pheno.sm$arth11),], fake.simper, c("arth1
kruskal.pretty(OTU[!is.na(Pheno.sm$cancer11),], Pheno.sm[!is.na(Pheno.sm$cancer11),], fake.simper, c("c
kruskal.pretty(OTU[!is.na(Pheno.sm$stroke11),], Pheno.sm[!is.na(Pheno.sm$stroke11),], fake.simper, c("s
The IBS variable was not part of the Pheno.sm survey so we curate it from the larger Pheno data set.
```

```
• jx148rer = graduates
```

• px148rer = siblings

```
vars.IBS = data.frame(
   IBS = c(Pheno$jx148rer, Pheno$px148rer),
   ID = c(paste(Pheno$idpriv, "g", sep=""), paste(Pheno$idpriv, "s", sep="")),
   row.names = c(paste(Pheno$idpriv, "g", sep=""), paste(Pheno$idpriv, "s", sep=""))
)

#Subset to data with microbiota samples and IBS data
vars.IBS.sub = as.data.frame(subset(vars.IBS, row.names(vars.IBS) %in% row.names(OTU)))
vars.IBS.all = as.data.frame(vars.IBS.sub[!is.na(vars.IBS.sub$IBS),])
OTU.IBS = subset(OTU, row.names(OTU) %in% row.names(vars.IBS.all))
```

Run Kruskal-Wallis test of new IBS varibale.

```
kruskal.pretty(OTU.IBS, vars.IBS.all, fake.simper, c("IBS"), 'IBS', Tax.orig)
```

Figures

Figure 1

```
set.seed(51234)
#Calculate NMDS
uwUF.g.ordu = ordinate(physeq.tree.g, method="NMDS", distance="unifrac", weighted=FALSE)
## Warning in UniFrac(physeq, ...): Randomly assigning root as -- Otu04244 --
## in the phylogenetic tree in the data you provided.
## Run 0 stress 0.1548755
## Run 1 stress 0.171804
## Run 2 stress 0.1630789
## Run 3 stress 0.173726
## Run 4 stress 0.1855051
## Run 5 stress 0.1834981
## Run 6 stress 0.1866751
## Run 7 stress 0.1641615
## Run 8 stress 0.1728711
## Run 9 stress 0.1937877
## Run 10 stress 0.1705475
## Run 11 stress 0.1643311
## Run 12 stress 0.1579658
## Run 13 stress 0.1838658
## Run 14 stress 0.1867786
## Run 15 stress 0.1578942
## Run 16 stress 0.177159
## Run 17 stress 0.173864
## Run 18 stress 0.1681529
## Run 19 stress 0.1841738
## Run 20 stress 0.1663627
## *** No convergence -- monoMDS stopping criteria:
       20: stress ratio > sratmax
#Create data set of just significant variables
vars.envfit = data.frame(
  AB = ifelse(vars.g$AB == "Y", 2, 1), #Reverse code so that arrow points toward Y responses
  cohabYN = ifelse(vars.g$cohabYN == "N", 1, 2), #Reverse code so that arrow points toward Y responses
  socialness = vars.g$social.sum,
  hbs = ifelse(vars.g$hbs11 == "Y", 2, 1), #Reverse code so that arrow points toward Y responses
  heart = ifelse(vars.g$heart11 == 1, 2, 1), #Reverse code so that arrow points toward Y responses
  meat = vars.g$meat,
  poultry = vars.g$poul,
  prot.sum = vars.g$prot.sum,
  social.cohabY = ifelse(vars.g$cohabYN == "Y", vars.g$social.sum, NA),
  social.cohabN = ifelse(vars.g$cohabYN == "N", vars.g$social.sum, NA),
  row.names = row.names(vars.g))
```

Warning in cor(Hperm, take): the standard deviation is zero

#Fit variables to NMDS

```
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
ef.uwUF.socY = envfit(uwUF.g.ordu ~ social.cohabY, data=vars.envfit, na.rm=TRUE)
ef.uwUF.socN = envfit(uwUF.g.ordu ~ social.cohabN, data=vars.envfit, na.rm=TRUE)
ef.uwUF.hbs = envfit(uwUF.g.ordu ~ hbs, data=vars.envfit, na.rm=TRUE)
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
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ef.uwUF.heart = envfit(uwUF.g.ordu ~ heart, data=vars.envfit, na.rm=TRUE)
ef.uwUF.meat = envfit(uwUF.g.ordu ~ meat, data=vars.envfit, na.rm=TRUE)
ef.uwUF.poul = envfit(uwUF.g.ordu ~ poultry, data=vars.envfit, na.rm=TRUE)
#Figure 1
postscript("FIGURE1.eps", width = 4.2, height = 4.5, horizontal = FALSE, onefile = FALSE, paper = "spec
plot(uwUF.g.ordu, type="n", main="", xlab="", ylab="")
## Warning in ordiplot(x, choices = choices, type = type, display = display, :
## Species scores not available
points(uwUF.g.ordu, pch=21, cex=0.75, display="sites", col="black", bg=c("red", "blue")[as.factor(Pheno.
plot(ef.uwUF.AB, col="red")
## Warning in min(u): no non-missing arguments to min; returning Inf
## Warning in min(x): no non-missing arguments to min; returning Inf
## Warning in max(x): no non-missing arguments to max; returning -Inf
## Warning in min(x): no non-missing arguments to min; returning Inf
## Warning in max(x): no non-missing arguments to max; returning -Inf
## Warning in min(u): no non-missing arguments to min; returning Inf
plot(ef.uwUF.socY, col="black")
plot(ef.uwUF.socN, col="red")
plot(ef.uwUF.hbs, col="red")
## Warning in min(u): no non-missing arguments to min; returning Inf
## Warning in min(x): no non-missing arguments to min; returning Inf
## Warning in max(x): no non-missing arguments to max; returning -Inf
## Warning in min(x): no non-missing arguments to min; returning Inf
## Warning in max(x): no non-missing arguments to max; returning -Inf
## Warning in min(u): no non-missing arguments to min; returning Inf
plot(ef.uwUF.heart, col="red")
plot(ef.uwUF.meat, col="black")
plot(ef.uwUF.poul, col="black")
dev.off()
## pdf
##
```

Figure 2

```
#Figure 2
postscript("FIGURE2new.eps", width = 5, height = 4, horizontal = FALSE, onefile = FALSE, paper = "specilayout(matrix(c(1,2), 1, 2, byrow = TRUE), widths=c(1,1), heights=c(1,1))

#Shannon
boxplot(Met.g$shannon ~ vars.g$cohabYN, ylab="Shannon's diversity", main="", xlab="Cohabitating", col=c"
#Chao
boxplot(Met.g$chao ~ vars.g$cohabYN, ylab="Chao's richness", main="", xlab="Cohabitating", col=c("red", dev.off())

## pdf
## pdf
## pdf
## 2
```

Figure 3

```
#Load results from KW.subsample.fxn
KW.results.final = read.table("KW.results.csv", sep=",")
#Re-organize into data frame
gp.se.gs.boxplot = data.frame(
  BC = c(match.BC.gp.se.gs, KW.results.final[KW.results.final$V6 == "dist" & complete.cases(KW.results.
  J = c(match.J.gp.se.gs, KW.results.final[KW.results.final$V6 == "dist" & complete.cases(KW.results.fi
  wUF = c(match.wUF.gp.se.gs, KW.results.final[KW.results.final$V6 == "dist" & complete.cases(KW.result
  uwUF = c(match.uwUF.gp.se.gs, KW.results.final[KW.results.final$V6 == "dist" & complete.cases(KW.resu
  group = c(rep("1.spouse", length(match.BC.gp.se)), rep("2.sibling", length(match.BC.gs)), rep("3.unre
#Add variable for closeness:pair type
vars.gp.se.gs$closeness.group = ifelse(floor(vars.gp.se.gs$closeness) == 1 & vars.gp.se.gs$group == "sp
                                ifelse(floor(vars.gp.se.gs$closeness) == 2 & vars.gp.se.gs$group == "sp
                                ifelse(floor(vars.gp.se.gs$closeness) == 3 & vars.gp.se.gs$group == "sp
                                ifelse(floor(vars.gp.se.gs$closeness) == 4 & vars.gp.se.gs$group == "sp
                                ifelse(floor(vars.gp.se.gs$closeness) == 1 & vars.gp.se.gs$group == "si"
                                ifelse(floor(vars.gp.se.gs$closeness) == 2 & vars.gp.se.gs$group == "si
                                ifelse(floor(vars.gp.se.gs$closeness) == 3 & vars.gp.se.gs$group == "si
                                ifelse(floor(vars.gp.se.gs$closeness) == 4 & vars.gp.se.gs$group == "si
#Figure 3
postscript("FIGURE3.eps", width = 8, height = 5, horizontal = FALSE, onefile = FALSE, paper = "special"
layout(matrix(c(1,2,3), 1, 3, byrow = TRUE), widths=c(2,2.5,3), heights=c(1,1,1))
boxplot(gp.se.gs.boxplot$uwUF ~ gp.se.gs.boxplot$group, main = "", ylab="Unweighted UniFrac distance",
boxplot(match.uwUF.gp.se.gs~floor(vars.gp.se.gs$closeness), xlab="", ylab="", main="", outpch=NA, ylim
boxplot(match.uwUF.gp.se.gs~vars.gp.se.gs*closeness.group, col=c("blue", "blue", "blue", "red", "blue", "blue",
dev.off()
## pdf
##
```

Figure 4

```
#Calculate relative abundances
abund = OTU/rowSums(OTU)
#Re-code variable to YN letters
Pheno.sm$hbs = ifelse(Pheno.sm$hbs11 == 1, "Y", ifelse(Pheno.sm$hbs11 == 2, "N", Pheno.sm$hbs11))
#Combine OTU abundance with health data
hbs.OTUs1 = data.frame(
  abundance = c(abund$0tu00160*100,abund$0tu00295*100),
  OTU = c(rep("OTU160", 408), rep("OTU295", 408)),
  hbs = rep(Pheno.sm$hbs, 2))
hbs.OTUs2 = data.frame(
  abundance = c(abund\$0tu00004*100, abund\$0tu00019*100, abund\$0tu00074*100),
  OTU = c(rep("OTU004", 408), rep("OTU019", 408), rep("OTU074", 408)),
  hbs = rep(Pheno.sm$hbs, 3))
#Calculate means and standard deviations of OTU abundances within health groups
hbs.OTUs.data1 = aggregate(hbs.OTUs1$abundance, by=list(hbs.OTUs1$OTU, hbs.OTUs1$hbs), FUN = function(x
hbs.OTUs.data1 = do.call(data.frame, hbs.OTUs.data1)
hbs.OTUs.data1$se = hbs.OTUs.data1$x.sd / sqrt(hbs.OTUs.data1$x.n)
colnames(hbs.OTUs.data1) = c("OTU", "hbs", "mean", "sd", "n", "se")
hbs.OTUs.data2 = aggregate(hbs.OTUs2$abundance, by=list(hbs.OTUs2$OTU, hbs.OTUs2$hbs), FUN = function(x
hbs.OTUs.data2 = do.call(data.frame, hbs.OTUs.data2)
hbs.OTUs.data2$se = hbs.OTUs.data2$x.sd / sqrt(hbs.OTUs.data2$x.n)
colnames(hbs.OTUs.data2) = c("OTU", "hbs", "mean", "sd", "n", "se")
#Figure 4
limits1 <- aes(ymax = hbs.OTUs.data1$mean + hbs.OTUs.data1$se, ymin = hbs.OTUs.data1$mean - hbs.OTUs.da
p1 = ggplot(data = hbs.OTUs.data1, aes(x = factor(OTU), y = mean, fill = factor(hbs)))
limits2 <- aes(ymax = hbs.0TUs.data2$mean + hbs.0TUs.data2$se, ymin = hbs.0TUs.data2$mean - hbs.0TUs.da
p2 = ggplot(data = hbs.OTUs.data2, aes(x = factor(OTU), y = mean, fill = factor(hbs)))
plot1 = p1 + geom_bar(stat = "identity", position = position_dodge(0.9)) +
  geom_errorbar(limits1, position = position_dodge(0.9), width = 0.25) +
  labs(x = "", y = "Relative abundance (%)") + scale_fill_manual(name = "HBS", values=c("red", "blue"))
  theme_bw() + theme(panel.border = element_blank(), panel.grid.major = element_blank(), panel.grid.min
plot2 = p2 + geom_bar(stat = "identity", position = position_dodge(0.9)) +
  geom_errorbar(limits2, position = position_dodge(0.9), width = 0.25) +
  labs(x = "", y = "") + scale_fill_manual(name = "HBS", values=c("red", "blue")) +
  theme_bw() + theme(panel.border = element_blank(), panel.grid.major = element_blank(), panel.grid.min
postscript("FIGURE4.eps", width = 5, height = 2.5, horizontal = FALSE, onefile = FALSE, paper = "specia
multiplot(plot1, plot2, cols=2)
dev.off()
## pdf
##
```

Figure S1

```
set.seed(51234)
#Calculate NMDS
BC.g.MDS=metaMDS(OTU.g, distance="bray", k=2, trymax=500)
## Square root transformation
## Wisconsin double standardization
## Run 0 stress 0.1778574
## Run 1 stress 0.1830169
## Run 2 stress 0.2165385
## Run 3 stress 0.1768809
## ... New best solution
## ... Procrustes: rmse 0.03254256 max resid 0.3843774
## Run 4 stress 0.1782439
## Run 5 stress 0.1757846
## ... New best solution
## ... Procrustes: rmse 0.01052766 max resid 0.1034541
## Run 6 stress 0.186635
## Run 7 stress 0.1768495
## Run 8 stress 0.1866905
## Run 9 stress 0.1961226
## Run 10 stress 0.175787
## ... Procrustes: rmse 0.0006641125 max resid 0.005862217
## ... Similar to previous best
## Run 11 stress 0.1870023
## Run 12 stress 0.1790881
## Run 13 stress 0.2053181
## Run 14 stress 0.1951932
## Run 15 stress 0.19719
## Run 16 stress 0.2008779
## Run 17 stress 0.1874823
## Run 18 stress 0.1899205
## Run 19 stress 0.1768658
## Run 20 stress 0.1768485
## *** Solution reached
J.g.MDS=metaMDS(OTU.g, distance="jaccard", k=2, trymax=500)
## Square root transformation
## Wisconsin double standardization
## Run 0 stress 0.1778344
## Run 1 stress 0.1862218
## Run 2 stress 0.1830001
## Run 3 stress 0.1895922
## Run 4 stress 0.1790675
## Run 5 stress 0.1908328
## Run 6 stress 0.1956368
## Run 7 stress 0.1783647
## Run 8 stress 0.2001721
## Run 9 stress 0.1790354
## Run 10 stress 0.1928207
## Run 11 stress 0.1914063
## Run 12 stress 0.1883793
```

```
## Run 13 stress 0.1867394
## Run 14 stress 0.2097437
## Run 15 stress 0.1811278
## Run 16 stress 0.1856737
## Run 17 stress 0.2057145
## Run 18 stress 0.188236
## Run 19 stress 0.1915302
## Run 20 stress 0.1982333
## Run 21 stress 0.2017376
## Run 22 stress 0.1788227
## Run 23 stress 0.1943584
## Run 24 stress 0.1781236
## ... Procrustes: rmse 0.007399325 max resid 0.08542497
## Run 25 stress 0.19052
## Run 26 stress 0.1779312
## ... Procrustes: rmse 0.003673951 max resid 0.04211143
## Run 27 stress 0.1921566
## Run 28 stress 0.1790291
## Run 29 stress 0.1778557
## ... Procrustes: rmse 0.001420418 max resid 0.01682678
## Run 30 stress 0.215146
## Run 31 stress 0.1913911
## Run 32 stress 0.1885353
## Run 33 stress 0.1858878
## Run 34 stress 0.1913819
## Run 35 stress 0.1929349
## Run 36 stress 0.2047987
## Run 37 stress 0.1929133
## Run 38 stress 0.193596
## Run 39 stress 0.1758107
## ... New best solution
## ... Procrustes: rmse 0.03046111 max resid 0.3819559
## Run 40 stress 0.1776132
## Run 41 stress 0.1811713
## Run 42 stress 0.1801865
## Run 43 stress 0.1768579
## Run 44 stress 0.1884731
## Run 45 stress 0.1865361
## Run 46 stress 0.1946415
## Run 47 stress 0.1801935
## Run 48 stress 0.1768662
## Run 49 stress 0.1785746
## Run 50 stress 0.1830558
## Run 51 stress 0.2041382
## Run 52 stress 0.2010643
## Run 53 stress 0.1790965
## Run 54 stress 0.2140271
## Run 55 stress 0.2014982
## Run 56 stress 0.185326
## Run 57 stress 0.2158569
## Run 58 stress 0.1877491
## Run 59 stress 0.19494
## Run 60 stress 0.2141599
## Run 61 stress 0.1891321
```

```
## Run 62 stress 0.1931647
## Run 63 stress 0.1800636
## Run 64 stress 0.1875424
## Run 65 stress 0.1907144
## Run 66 stress 0.1759712
## ... Procrustes: rmse 0.007250359 max resid 0.08512437
## Run 67 stress 0.1758466
## ... Procrustes: rmse 0.003536395 max resid 0.04337283
## Run 68 stress 0.1788659
## Run 69 stress 0.1810522
## Run 70 stress 0.1875395
## Run 71 stress 0.1766162
## Run 72 stress 0.1962701
## Run 73 stress 0.1782816
## Run 74 stress 0.1919053
## Run 75 stress 0.1861325
## Run 76 stress 0.1979306
## Run 77 stress 0.1955984
## Run 78 stress 0.1926229
## Run 79 stress 0.1792036
## Run 80 stress 0.1948191
## Run 81 stress 0.1969628
## Run 82 stress 0.1920004
## Run 83 stress 0.1782967
## Run 84 stress 0.216145
## Run 85 stress 0.1967847
## Run 86 stress 0.1830781
## Run 87 stress 0.1885397
## Run 88 stress 0.1792427
## Run 89 stress 0.1894345
## Run 90 stress 0.1943004
## Run 91 stress 0.1959765
## Run 92 stress 0.1916804
## Run 93 stress 0.1790446
## Run 94 stress 0.1930767
## Run 95 stress 0.2020236
## Run 96 stress 0.1923937
## Run 97 stress 0.1866619
## Run 98 stress 0.1757793
## ... New best solution
## ... Procrustes: rmse 0.001693843 max resid 0.01223205
## Run 99 stress 0.1759575
## ... Procrustes: rmse 0.007292715 max resid 0.08513722
## Run 100 stress 0.1919335
## Run 101 stress 0.1961309
## Run 102 stress 0.1790366
## Run 103 stress 0.1903173
## Run 104 stress 0.1783425
## Run 105 stress 0.2090502
## Run 106 stress 0.4161178
## Run 107 stress 0.1920216
## Run 108 stress 0.178563
## Run 109 stress 0.1977078
## Run 110 stress 0.1800627
```

```
## Run 111 stress 0.1904304
## Run 112 stress 0.2059046
## Run 113 stress 0.1783447
## Run 114 stress 0.1860653
## Run 115 stress 0.1766072
## Run 116 stress 0.1782865
## Run 117 stress 0.1879055
## Run 118 stress 0.2143067
## Run 119 stress 0.1856557
## Run 120 stress 0.2143696
## Run 121 stress 0.1841323
## Run 122 stress 0.1966262
## Run 123 stress 0.2107032
## Run 124 stress 0.2024043
## Run 125 stress 0.1991685
## Run 126 stress 0.1843545
## Run 127 stress 0.1935549
## Run 128 stress 0.1758982
## ... Procrustes: rmse 0.006693901 max resid 0.08532005
## Run 129 stress 0.1938159
## Run 130 stress 0.1860952
## Run 131 stress 0.1989884
## Run 132 stress 0.1885009
## Run 133 stress 0.1862341
## Run 134 stress 0.1785989
## Run 135 stress 0.2049853
## Run 136 stress 0.1785976
## Run 137 stress 0.2047403
## Run 138 stress 0.2088977
## Run 139 stress 0.1888156
## Run 140 stress 0.1946764
## Run 141 stress 0.1765675
## Run 142 stress 0.1758444
## ... Procrustes: rmse 0.003334519 max resid 0.04317937
## Run 143 stress 0.1768781
## Run 144 stress 0.1987545
## Run 145 stress 0.1788026
## Run 146 stress 0.1779653
## Run 147 stress 0.1758474
## ... Procrustes: rmse 0.003402444 max resid 0.04342179
## Run 148 stress 0.1902268
## Run 149 stress 0.2039546
## Run 150 stress 0.1974484
## Run 151 stress 0.1765873
## Run 152 stress 0.1920247
## Run 153 stress 0.1803372
## Run 154 stress 0.1798656
## Run 155 stress 0.1868677
## Run 156 stress 0.1803026
## Run 157 stress 0.1796587
## Run 158 stress 0.1790663
## Run 159 stress 0.192147
## Run 160 stress 0.1906735
## Run 161 stress 0.1915894
```

```
## Run 162 stress 0.1792629
## Run 163 stress 0.1839238
## Run 164 stress 0.1958771
## Run 165 stress 0.201776
## Run 166 stress 0.1782835
## Run 167 stress 0.1785736
## Run 168 stress 0.2113934
## Run 169 stress 0.1971785
## Run 170 stress 0.1967032
## Run 171 stress 0.214885
## Run 172 stress 0.1758913
## ... Procrustes: rmse 0.006672755 max resid 0.08556301
## Run 173 stress 0.20161
## Run 174 stress 0.198531
## Run 175 stress 0.1950551
## Run 176 stress 0.1941418
## Run 177 stress 0.1782492
## Run 178 stress 0.1768658
## Run 179 stress 0.1783474
## Run 180 stress 0.1973157
## Run 181 stress 0.1857687
## Run 182 stress 0.1802025
## Run 183 stress 0.1801703
## Run 184 stress 0.1961555
## Run 185 stress 0.1919891
## Run 186 stress 0.1856448
## Run 187 stress 0.1971833
## Run 188 stress 0.1812108
## Run 189 stress 0.2149783
## Run 190 stress 0.1758009
## ... Procrustes: rmse 0.001199255 max resid 0.01186763
## Run 191 stress 0.2100291
## Run 192 stress 0.1941543
## Run 193 stress 0.1765786
## Run 194 stress 0.1785918
## Run 195 stress 0.1802696
## Run 196 stress 0.2043005
## Run 197 stress 0.1776268
## Run 198 stress 0.1858989
## Run 199 stress 0.2015495
## Run 200 stress 0.1783944
## Run 201 stress 0.4161477
## Run 202 stress 0.1883485
## Run 203 stress 0.1877381
## Run 204 stress 0.1922341
## Run 205 stress 0.1852034
## Run 206 stress 0.1782428
## Run 207 stress 0.1940847
## Run 208 stress 0.178364
## Run 209 stress 0.1872153
## Run 210 stress 0.2048338
## Run 211 stress 0.2143421
## Run 212 stress 0.1813106
## Run 213 stress 0.1765823
```

```
## Run 214 stress 0.1779442
## Run 215 stress 0.1792325
## Run 216 stress 0.1945764
## Run 217 stress 0.1779703
## Run 218 stress 0.4161317
## Run 219 stress 0.1788418
## Run 220 stress 0.1777212
## Run 221 stress 0.2019454
## Run 222 stress 0.200056
## Run 223 stress 0.1785676
## Run 224 stress 0.1934785
## Run 225 stress 0.1929416
## Run 226 stress 0.189122
## Run 227 stress 0.1867236
## Run 228 stress 0.1944087
## Run 229 stress 0.2089062
## Run 230 stress 0.1882561
## Run 231 stress 0.1859053
## Run 232 stress 0.1944056
## Run 233 stress 0.1864893
## Run 234 stress 0.1892151
## Run 235 stress 0.1850967
## Run 236 stress 0.1994451
## Run 237 stress 0.2068457
## Run 238 stress 0.1883604
## Run 239 stress 0.1963499
## Run 240 stress 0.1779844
## Run 241 stress 0.1942086
## Run 242 stress 0.21075
## Run 243 stress 0.1966056
## Run 244 stress 0.1779927
## Run 245 stress 0.1954545
## Run 246 stress 0.1855553
## Run 247 stress 0.4161297
## Run 248 stress 0.1790394
## Run 249 stress 0.2023709
## Run 250 stress 0.1782992
## Run 251 stress 0.1810201
## Run 252 stress 0.1903253
## Run 253 stress 0.1758363
## ... Procrustes: rmse 0.003480458 max resid 0.04327474
## Run 254 stress 0.1777257
## Run 255 stress 0.1905049
## Run 256 stress 0.1768649
## Run 257 stress 0.1880803
## Run 258 stress 0.1954373
## Run 259 stress 0.1792364
## Run 260 stress 0.1844688
## Run 261 stress 0.178364
## Run 262 stress 0.1797154
## Run 263 stress 0.1783397
## Run 264 stress 0.1765699
## Run 265 stress 0.2055737
## Run 266 stress 0.1779793
```

```
## Run 267 stress 0.192255
## Run 268 stress 0.1788459
## Run 269 stress 0.187432
## Run 270 stress 0.1759812
## ... Procrustes: rmse 0.007315288 max resid 0.08490335
## Run 271 stress 0.178841
## Run 272 stress 0.1785707
## Run 273 stress 0.2157835
## Run 274 stress 0.1942939
## Run 275 stress 0.1759012
## ... Procrustes: rmse 0.006838673 max resid 0.08537501
## Run 276 stress 0.206187
## Run 277 stress 0.1933202
## Run 278 stress 0.2020784
## Run 279 stress 0.1783465
## Run 280 stress 0.1928502
## Run 281 stress 0.1788609
## Run 282 stress 0.1782851
## Run 283 stress 0.1875743
## Run 284 stress 0.1887271
## Run 285 stress 0.1885436
## Run 286 stress 0.1811361
## Run 287 stress 0.1906411
## Run 288 stress 0.1765911
## Run 289 stress 0.1782554
## Run 290 stress 0.200926
## Run 291 stress 0.4161383
## Run 292 stress 0.1757835
## ... Procrustes: rmse 0.0006544988 max resid 0.007231332
## ... Similar to previous best
## *** Solution reached
wUF.g.ordu = ordinate(physeq.tree.g, method="NMDS", distance="unifrac", weighted=TRUE)
## Warning in UniFrac(physeq, ...): Randomly assigning root as -- OtuO4170 --
## in the phylogenetic tree in the data you provided.
## Run 0 stress 0.1110309
## Run 1 stress 0.1112774
## ... Procrustes: rmse 0.003638806 max resid 0.02286034
## Run 2 stress 0.1122669
## Run 3 stress 0.1114875
## ... Procrustes: rmse 0.005288243 max resid 0.02933534
## Run 4 stress 0.1113367
## ... Procrustes: rmse 0.004821078 max resid 0.03221992
## Run 5 stress 0.1118956
## Run 6 stress 0.1120194
## Run 7 stress 0.1114885
## ... Procrustes: rmse 0.01008123 max resid 0.03345407
## Run 8 stress 0.4160518
## Run 9 stress 0.1112226
## ... Procrustes: rmse 0.009058642 max resid 0.03666701
## Run 10 stress 0.1111704
## ... Procrustes: rmse 0.003355195 max resid 0.02518748
## Run 11 stress 0.1112287
```

```
## ... Procrustes: rmse 0.01122786 max resid 0.03989851
## Run 12 stress 0.1115027
## ... Procrustes: rmse 0.005896309 max resid 0.02518071
## Run 13 stress 0.4160153
## Run 14 stress 0.1110765
## ... Procrustes: rmse 0.004865465 max resid 0.02148985
## Run 15 stress 0.1113322
## ... Procrustes: rmse 0.006003153 max resid 0.03272031
## Run 16 stress 0.1112682
## ... Procrustes: rmse 0.006793857 max resid 0.03167451
## Run 17 stress 0.4161426
## Run 18 stress 0.1112634
## ... Procrustes: rmse 0.005501874 max resid 0.03138958
## Run 19 stress 0.1111517
## ... Procrustes: rmse 0.011615 max resid 0.03525381
## Run 20 stress 0.1113179
## ... Procrustes: rmse 0.005913694 max resid 0.03059023
## *** No convergence -- monoMDS stopping criteria:
       20: stress ratio > sratmax
#Fit environmental variables
ef.BC.AB = envfit(BC.g.MDS ~ AB, data=vars.envfit, na.rm=TRUE)
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
```

Warning in cor(Hperm, take): the standard deviation is zero ## Warning in cor(Hperm, take): the standard deviation is zero

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## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
## Warning in cor(Hperm, take): the standard deviation is zero
ef.BC.socY = envfit(BC.g.MDS ~ social.cohabY, data=vars.envfit, na.rm=TRUE)
ef.BC.socN = envfit(BC.g.MDS ~ social.cohabN, data=vars.envfit, na.rm=TRUE)
ef.BC.hbs = envfit(BC.g.MDS ~ hbs, data=vars.envfit, na.rm=TRUE)
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
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## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
```

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## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
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## Warning in cor(H, Pw): the standard deviation is zero
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## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
```

```
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
## Warning in cor(H, Pw): the standard deviation is zero
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ef.BC.heart = envfit(BC.g.MDS ~ heart, data=vars.envfit, na.rm=TRUE)
ef.BC.prot = envfit(BC.g.MDS ~ prot.sum, data=vars.envfit, na.rm=TRUE)
ef.BC.meat = envfit(BC.g.MDS ~ meat, data=vars.envfit, na.rm=TRUE)
ef.BC.poul = envfit(BC.g.MDS ~ poultry, data=vars.envfit, na.rm=TRUE)
ef.J.AB = envfit(J.g.MDS ~ AB, data=vars.envfit, na.rm=TRUE)
## Warning in cor(H, Pw): the standard deviation is zero
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ef.J.cohab = envfit(J.g.MDS ~ cohabYN, data=vars.envfit, na.rm=TRUE)
ef.J.socY = envfit(J.g.MDS ~ social.cohabY, data=vars.envfit, na.rm=TRUE)
ef.J.socN = envfit(J.g.MDS ~ social.cohabN, data=vars.envfit, na.rm=TRUE)
ef.J.hbs = envfit(J.g.MDS ~ hbs, data=vars.envfit, na.rm=TRUE)
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ef.J.heart = envfit(J.g.MDS ~ heart, data=vars.envfit, na.rm=TRUE)
ef.J.prot = envfit(J.g.MDS ~ prot.sum, data=vars.envfit, na.rm=TRUE)
ef.J.meat = envfit(J.g.MDS ~ meat, data=vars.envfit, na.rm=TRUE)
ef.J.poul = envfit(J.g.MDS ~ poultry, data=vars.envfit, na.rm=TRUE)
ef.wUF.AB = envfit(wUF.g.ordu ~ AB, data=vars.envfit, na.rm=TRUE)
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ef.wUF.prot = envfit(wUF.g.ordu ~ prot.sum, data=vars.envfit, na.rm=TRUE)
ef.wUF.meat = envfit(wUF.g.ordu ~ meat, data=vars.envfit, na.rm=TRUE)
ef.wUF.poul = envfit(wUF.g.ordu ~ poultry, data=vars.envfit, na.rm=TRUE)
```

```
#Figure S1
postscript("FIGURES1.eps", width = 9, height = 3, horizontal = FALSE, onefile = FALSE, paper = "special
layout(matrix(c(1,2,3), 1, 3, byrow = TRUE), widths=c(1,1,1), heights=c(1,1,1))
plot(BC.g.MDS, type="n", main="Bray-Curtis", xlab="", ylab="")
  points(BC.g.MDS, pch=21, cex=0.75, display="sites", col="black", bg=c("red", "blue")[as.factor(Pheno.si
 plot(ef.BC.AB, col="red")
## Warning in min(u): no non-missing arguments to min; returning Inf
## Warning in min(x): no non-missing arguments to min; returning Inf
## Warning in max(x): no non-missing arguments to max; returning -Inf
## Warning in min(x): no non-missing arguments to min; returning Inf
## Warning in max(x): no non-missing arguments to max; returning -Inf
## Warning in min(u): no non-missing arguments to min; returning Inf
 plot(ef.BC.hbs, col="red")
## Warning in min(u): no non-missing arguments to min; returning Inf
## Warning in min(x): no non-missing arguments to min; returning Inf
## Warning in max(x): no non-missing arguments to max; returning -Inf
## Warning in min(x): no non-missing arguments to min; returning Inf
## Warning in max(x): no non-missing arguments to max; returning -Inf
## Warning in min(u): no non-missing arguments to min; returning Inf
  plot(ef.BC.heart, col="red")
 plot(ef.BC.prot, col="black")
 plot(ef.BC.socY, col="black")
 plot(ef.BC.socN, col="black")
  plot(ef.BC.meat, col="black")
  plot(ef.BC.poul, col="black")
plot(J.g.MDS, type="n", main="Jaccard", xlab="", ylab="")
  points(J.g.MDS, pch=21, cex=0.75, display="sites", col="black", bg=c("red", "blue")[as.factor(Pheno.sm
 plot(ef.J.AB, col="red")
## Warning in min(u): no non-missing arguments to min; returning Inf
## Warning in min(x): no non-missing arguments to min; returning Inf
## Warning in max(x): no non-missing arguments to max; returning -Inf
## Warning in min(x): no non-missing arguments to min; returning Inf
## Warning in max(x): no non-missing arguments to max; returning -Inf
## Warning in min(u): no non-missing arguments to min; returning Inf
  plot(ef.J.cohab, col="black")
 plot(ef.J.socY, col="black")
 plot(ef.J.socN, col="black")
 plot(ef.J.hbs, col="black")
## Warning in min(u): no non-missing arguments to min; returning Inf
## Warning in min(x): no non-missing arguments to min; returning Inf
```

```
## Warning in max(x): no non-missing arguments to max; returning -Inf
## Warning in min(x): no non-missing arguments to min; returning Inf
## Warning in max(x): no non-missing arguments to max; returning -Inf
## Warning in min(u): no non-missing arguments to min; returning Inf
  plot(ef.J.heart, col="black")
 plot(ef.J.prot, col="black")
 plot(ef.J.meat, col="black")
  plot(ef.J.poul, col="black")
plot(wUF.g.ordu, type="n", main="Weighted UniFrac", xlab="", ylab="", xlim=c(-0.03,0.03), ylim=c(-0.03,
## Warning in ordiplot(x, choices = choices, type = type, display = display, :
## Species scores not available
  points(wUF.g.ordu, pch=21, cex=0.75, display="sites", col="black", bg=c("red", "blue")[as.factor(Pheno
 plot(ef.wUF.AB, col="black")
## Warning in min(u): no non-missing arguments to min; returning Inf
## Warning in min(x): no non-missing arguments to min; returning Inf
## Warning in max(x): no non-missing arguments to max; returning -Inf
## Warning in min(x): no non-missing arguments to min; returning Inf
## Warning in max(x): no non-missing arguments to max; returning -Inf
## Warning in min(u): no non-missing arguments to min; returning Inf
 plot(ef.wUF.prot, col="black")
  plot(ef.wUF.meat, col="black")
  plot(ef.wUF.poul, col="black")
dev.off()
## pdf
##
```

Figure S2

```
#Re-code variables to YN letters
vars.g$AB = ifelse(vars.g$AB == 1, "Y", ifelse(vars.g$AB == 2, "N", vars.g$AB))
vars.g$hbs11 = ifelse(vars.g$hbs11 == 1, "Y", ifelse(vars.g$hbs11 == 2, "N", vars.g$hbs11))

#Figure S2ABCD
postscript("FIGURES2a.eps", width = 6, height = 6, horizontal = FALSE, onefile = FALSE, paper = "special layout(matrix(c(1,2,3,4), 2, 2, byrow = TRUE), widths=c(1,1), heights=c(1,1))

#Shannon
boxplot(Met.g$shannon ~ vars.g$AB, ylab="Shannon's diversity", main="", xlab="Antibiotic use last 6mo")
stripchart(Met.g$shannon ~ vars.g$AB, col="black", bg="black", pch=21, vertical=TRUE, method="jitter"
boxplot(Met.g$shannon ~ vars.g$hbs11, main="", xlab="High blood sugar")
stripchart(Met.g$shannon ~ vars.g$hbs11, col="black", bg="black", pch=21, vertical=TRUE, method="jitt#Chao"
boxplot(Met.g$chao ~ vars.g$RU57, ylab="Chao's richness", main="", xlab="1957")
stripchart(Met.g$chao ~ vars.g$RU57, col="black", bg="black", pch=21, vertical=TRUE, method="jitter", boxplot(Met.g$chao ~ vars.g$RU11, main="", xlab="2011")
```

```
stripchart(Met.g$chao ~ vars.g$RU11, col="black", bg="black", pch=21, vertical=TRUE, method="jitter",
dev.off()
## pdf
##
    2
#Figure S2E
postscript("FIGURES2b.eps", width = 3, height = 3, horizontal = FALSE, onefile = FALSE, paper = "specia
layout(matrix(c(1), 1, 1, byrow = TRUE), widths=c(1), heights=c(1))
levelplot(Met.g$shannon ~ vars.g$poul*vars.g$meat, col.regions=function(x)rev(gray.colors(x)), xlab="Po
dev.off()
## pdf
##
#Figure S2F
postscript("FIGURES2c.eps", width = 3, height = 3, horizontal = FALSE, onefile = FALSE, paper = "specia")
layout(matrix(c(1), 1, 1, byrow = TRUE), widths=c(1), heights=c(1))
levelplot(Met.g$shannon ~ vars.g$poul*vars.g$pork, col.regions=function(x)rev(gray.colors(x)), xlab="Po
dev.off()
## pdf
##
```

Figure S3

```
#Calculate linear models
shan.social.cohabY = glm(Met.g[vars.g$cohabYN == "Y",]$shannon ~ vars.g[vars.g$cohabYN == "Y",]$social.
shan.social.cohabN = glm(Met.g[vars.g$cohabYN == "N",]$shannon ~ vars.g[vars.g$cohabYN == "N",]$social.
chao.social.cohabY = glm(Met.g[vars.g$cohabYN == "Y",]$chao ~ vars.g[vars.g$cohabYN == "Y",]$social.sum
chao.social.cohabN = glm(Met.g[vars.g$cohabYN == "N",]$chao ~ vars.g[vars.g$cohabYN == "N",]$social.sum
#Figure S3
postscript("FIGURES3.eps", width = 6, height = 6, horizontal = FALSE, onefile = FALSE, paper = "special
layout(matrix(c(1,2,3,4), 2, 2, byrow = TRUE), widths=c(1,1), heights=c(1,1))
plot(Met.g$shannon ~ vars.g$social.sum, pch=21, col="black", bg=c("red","blue")[vars.g$cohabYN], ylab="
        abline(shan.social.cohabY, col="blue")
        abline(shan.social.cohabN, col="red")
#Chao
plot(Met.g$chao ~ vars.g$social.sum, pch=21, col="black", bg=c("red","blue")[vars.g$cohabYN], ylab="", r
        abline(chao.social.cohabY, col="blue")
        abline(chao.social.cohabN, col="red")
dev.off()
## pdf
##
    2
```

Figure S4

```
#Figure S4
postscript("FIGURES4.eps", width = 9, height = 9, horizontal = FALSE, onefile = FALSE, paper = "special layout(matrix(c(1,2,3,4,5,6,7,8,9), 3, 3, byrow = TRUE), widths=c(1,1,1), heights=c(1,1,1))
```

```
boxplot(gp.se.gs.boxplot$BC ~ gp.se.gs.boxplot$group, main = "", ylab="Bray-Curtis distance", col=c("re
boxplot(match.BC.gp.se.gs~floor(vars.gp.se.gs*closeness), xlab="", ylab="", main="", outpch=NA, ylim=c
  stripchart(match.BC.gp.se.gs~floor(vars.gp.se.gs$closeness), col="black", bg="black", pch=21, vertica
boxplot(match.BC.gp.se.gs~vars.gp.se.gs*closeness.group, col=c("blue", "blue", "blue", "red", "blue", "blue", "
  stripchart(match.BC.gp.se.gs~vars.gp.se.gs$closeness.group, col="black", bg="black", pch=21, vertical-
boxplot(gp.se.gs.boxplot$J ~ gp.se.gs.boxplot$group, main = "", ylab="Jaccard distance", col=c("red","b
boxplot(match.J.gp.se.gs~floor(vars.gp.se.gs\$closeness), xlab="", ylab="", main="", outpch=NA, ylim=c(
  stripchart(match.J.gp.se.gs~floor(vars.gp.se.gs\$closeness), col="black", bg="black", pch=21, vertical"
boxplot(match.J.gp.se.gs~vars.gp.se.gs$closeness.group, col=c("blue", "blue", "blue", "red", "blue", "r
  stripchart(match.J.gp.se.gs~vars.gp.se.gs$closeness.group, col="black", bg="black", pch=21, vertical="
boxplot(gp.se.gs.boxplot$wUF ~ gp.se.gs.boxplot$group, main = "", ylab="Weighted UniFrac distance", col-
boxplot(match.wUF.gp.se.gs~floor(vars.gp.se.gs$closeness), xlab="", ylab="", main="", outpch=NA, ylim=
  stripchart(match.wUF.gp.se.gs~floor(vars.gp.se.gs\$closeness), col="black", bg="black", pch=21, vertic
boxplot(match.wUF.gp.se.gs~vars.gp.se.gs$closeness.group, col=c("blue", "blue", "blue", "red", "blue",
  stripchart(match.wUF.gp.se.gs~vars.gp.se.gs*closeness.group, col="black", bg="black", pch=21, vertica
dev.off()
## pdf
```

Figure S5

##

```
#Figure S5
postscript("FIGURES5.eps", width = 3, height = 3, horizontal = FALSE, onefile = FALSE, paper = "special
plot(match.uwUF.gp.se.gs ~ match.prot.gp.se.gs, bg=c(rep("red",94), rep("blue", 83)), col="black", pch=
    uwUF.spouse = lm(match.uwUF.gp.se.gs[vars.gp.se.gs$group == "spouse"] ~ match.prot.gp.se.gs[vars.gp.se
    uwUF.sib = lm(match.uwUF.gp.se.gs[vars.gp.se.gs$group == "sib"] ~ match.prot.gp.se.gs[vars.gp.se.gs
    uwUF.all = lm(match.uwUF.gp.se.gs ~ match.prot.gp.se.gs)
    abline(uwUF.spouse, col="red")
    abline(uwUF.sib, col="blue")
    abline(uwUF.all, col="black")
dev.off()

## pdf
## pdf
## pdf
```

Figure S6

```
#Figure S6A
postscript("FIGURES6a.eps", width = 3, height = 3, horizontal = FALSE, onefile = FALSE, paper = "special
boxplot(match.prot.gp.se.gs~vars.gp.se.gs$group, ylab="Euclidean distances of protein consumption", main
dev.off()

## pdf
## 2
#Figure S6B
postscript("FIGURES6b.eps", width = 3, height = 3, horizontal = FALSE, onefile = FALSE, paper = "special
plot(table(vars.gp.se.gs$group,floor(vars.gp.se.gs$closeness)), ylab="Closeness", main="")
dev.off()
```

pdf ## 2