Appendices: R code

Appendices for "Dryland rock detention structures increase herbaceous vegetation cover and stabilize shrub cover over 10 years but do not directly affect soil fertility" by Ossanna et al. 2024, *Science of the Total Environment*. Included are code and analysis used to make figures and full model output.

Data can be downloaded from the Zenodo archive at https://doi.org/10.5281/zenodo.8310363. The project's GitHub repository can be accessed from https://github.com/lossanna/AVCA_ElkLD.

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Appendices:

- Appendix A: Temporal vegetation trends, Figs 3 & S3
- Appendix B: Coefficient of variation, Figs 4 & S54
- Appendix C: Soil fertility, Figs S5-S7
- Appendix D: Structural equation modeling, models 1-4

Package versions:

- `R version 4.3.1`
- `tidyverse 2.0.0`

  - `dplyr 1.1.2`
  - `stringr 1.5.0`
  - `ggplot 3.4.3`
  - `tidyr 3.2.1`

- `agricolae 1.3-6`
- `plotrix 3.8-2`
- `ggpubr 0.6.0`
- `rstatix 0.7.2`
- `scales 1.2.1`
- `metagenomeSeq 1.42.0, Biobase 2.60.0, BiocGenerics 0.46.0`
- `vegan 2.6-4`
- `dada2 1.28.0, ShortRead 1.58.0, Biostrings 2.68.1` used for DADA2 pipeline (code not included here)
- `lavaan 0.6-16`

Appendix A: Temporal vegetation trends

Setup

```r
library(tidyverse)
library(agricolae)
library(plotrix)
library(ggpubr)
library(rstatix)
```

```r
# Load data ------------------------------------------------------------

notree.all <- read.csv("Herb-and-shrub-cover_2012-2021.csv")
herb.all <- read.csv("Herb-cover_2012-2021.csv")
shrub.all <- read.csv("Shrub-cover_2012-2021.csv")
invasive.all <- read.csv("Invasive-cover_2012-2021.csv")
plant.all <- read.csv("Species-cover_2012-2021.csv")
per.div <- read.csv("Perennial-plant-diversity_2012-2021.csv")
```

```r
# Functions ------------------------------------------------------------

# Convert columns to factor or date as needed
convert.cols <- function(x) {
  x$year.xaxis <- as.Date(x$year.xaxis)

  group.cols <- c("Sample", "Year", "Treatment")

  x[group.cols] <- lapply(x[group.cols], factor)

  return(x)
}
```

```r
# Data wrangling -------------------------------------------------------

notree.all <- convert.cols(notree.all)
herb.all <- convert.cols(herb.all)
shrub.all <- convert.cols(shrub.all)
invasive.all <- convert.cols(invasive.all)
per.div <- convert.cols(per.div)
```

Fig 3a: Grass, forb & shrub cover

```r
# Grass, forb, and shrub cover (notree) --------------------------------

# Find averages by year
notree.avg <- notree.all %>%
  group_by(Treatment, Year, year.xaxis) %>%
  summarise(mean = mean(Cover),
            SD = sd(Cover),
```

```
            SE = std.error(Cover),
            .groups = "keep")

# One-way ANOVA for Control
summary(aov(Cover ~ Year, data = filter(notree.all, Treatment == "Control"))) # p = 4.3e-06
```

```
##             Df Sum Sq Mean Sq F value  Pr(>F)
## Year          5   7815  1563.0    7.12 4.3e-06 ***
## Residuals   177  38854   219.5
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
notree.ctrl <- notree.all |>
  filter(Treatment == "Control")
anova.notree.ctrl <- aov(notree.ctrl$Cover ~ notree.ctrl$Year)
hsd.notree.ctrl <- HSD.test(anova.notree.ctrl, trt = "notree.ctrl$Year")
hsd.notree.ctrl$groups
```

```
##      notree.ctrl$Cover groups
## 2021          42.41935      a
## 2012          31.73194     ab
## 2014          31.58750     ab
## 2018          27.71774      b
## 2013          27.71250      b
## 2015          20.89315      b
```

```
# One-way ANOVA for Treated
summary(aov(Cover ~ Year, data = filter(notree.all, Treatment == "Treated"))) # p = 0.00304
```

```
##             Df Sum Sq Mean Sq F value  Pr(>F)
## Year          5   7804  1560.7   3.739 0.00304 **
## Residuals   178  74291   417.4
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
notree.trt <- notree.all |>
  filter(Treatment == "Treated")
anova.notree.trt <- aov(notree.trt$Cover ~ notree.trt$Year)
hsd.notree.trt <- HSD.test(anova.notree.trt, trt = "notree.trt$Year")
hsd.notree.trt$groups
```

```
##      notree.trt$Cover groups
## 2018         41.69960      a
## 2021         37.47581     ab
## 2014         31.40927    abc
## 2015         30.33669    abc
## 2012         26.72446     bc
## 2013         21.80029      c
```

```r
# Plot with one-way ANOVA letters
notree.ctrl.letters <- hsd.notree.ctrl$groups
notree.ctrl.letters <- notree.ctrl.letters |>
  mutate(Year = rownames(notree.ctrl.letters)) |>
  arrange(Year)
notree.trt.letters <- hsd.notree.trt$groups
notree.trt.letters <- notree.trt.letters |>
  mutate(Year = rownames(notree.trt.letters)) |>
  arrange(Year)

letters.notree2t <- data.frame(x = notree.avg$year.xaxis[1:6],
                               y = c(23, 18, 27, 26, 36.5, 32.5),
                               label = notree.trt.letters$groups)
letters.notree2c <- data.frame(x = notree.avg$year.xaxis[1:6],
                               y = c(35.5, 31.5, 36, 24, 31.5, 47),
                               label = notree.ctrl.letters$groups)

ptext.notree2 <- data.frame(x = as.Date("2019-09-01"),
                            y = 22,
                            label = "ANOVA, p < 0.01")
notree.plot2 <- ggplot(notree.avg, aes(x = year.xaxis, y = mean,
                                       color = Treatment)) +
  geom_line() +
  geom_point() +
  geom_pointrange(aes(ymin = mean - SE, ymax = mean + SE)) +
  xlab(NULL) +
  ylab("Cover (%)") +
  ggtitle("Vegetation cover, 2012-2021") +
  scale_color_manual(values = c("red", "#1F78B4")) +
  theme_bw() +
  theme(legend.position = "bottom") +
  geom_text(data = letters.notree2t,
            mapping = aes(x = x, y = y, label = label),
            color = "#1F78B4",
            size = 3) +
  geom_text(data = letters.notree2c,
            mapping = aes(x = x, y = y, label = label),
            color = "red",
            size = 3,
            fontface = "italic") +
  geom_text(data = ptext.notree2,
            aes(x = x, y = y, label = label),
            color = "gray30",
            size = 2.5) +
  theme(axis.text.x = element_text(color = "black")) +
  theme(plot.margin = margin(t = 0.1, r = 0.1, b = 0.3, l = 0.1, "in")) +
  theme(legend.position = "none")
notree.plot2
```
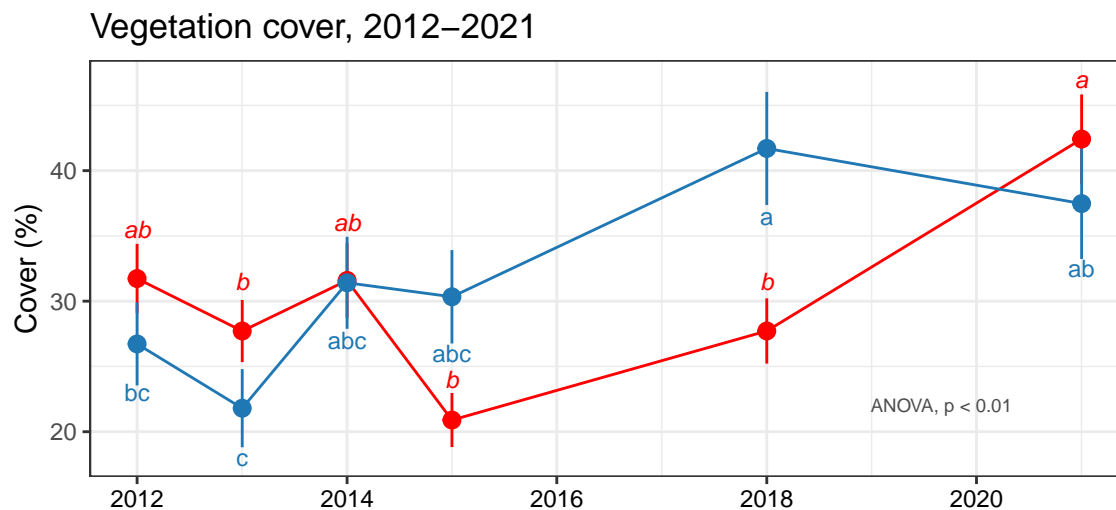
Fig 3b: Herbaceous (grass & forb) cover

```r
# Herbaceous cover ---------------------------------------------------------

# Find averages by year
herb.avg <- herb.all %>%
  group_by(Treatment, Year, year.xaxis) %>%
  summarise(mean = mean(Cover),
            SD = sd(Cover),
            SE = std.error(Cover),
            .groups = "keep")

# One-way ANOVA for Control
summary(aov(Cover ~ Year, data = filter(herb.all, Treatment == "Control"))) # 0.00434
```

```
##               Df Sum Sq Mean Sq F value  Pr(>F)
## Year           5   2821   564.3   3.556 0.00434 **
## Residuals    177  28083   158.7
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```r
herb.ctrl <- herb.all |>
  filter(Treatment == "Control")
anova.herb.ctrl <- aov(herb.ctrl$Cover ~ herb.ctrl$Year)
hsd.herb.ctrl <- HSD.test(anova.herb.ctrl, trt = "herb.ctrl$Year")
hsd.herb.ctrl$groups
```

```
##      herb.ctrl$Cover groups
## 2021        26.78629      a
## 2014        22.28333     ab
## 2012        20.03472     ab
## 2018        19.89718     ab
```

```
## 2013        17.41528        b
## 2015        14.21169        b
```

```
# One-way ANOVA for Treated
summary(aov(Cover ~ Year, data = filter(herb.all, Treatment == "Treated"))) # p = 3.77e-10
```

```
##              Df Sum Sq Mean Sq F value   Pr(>F)
## Year          5   6847  1369.5   12.17 3.77e-10 ***
## Residuals   178  20035   112.6
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
herb.trt <- herb.all |>
  filter(Treatment == "Treated")
anova.herb.trt <- aov(herb.trt$Cover ~ herb.trt$Year)
hsd.herb.trt <- HSD.test(anova.herb.trt, trt = "herb.trt$Year")
hsd.herb.trt$groups
```

```
##        herb.trt$Cover groups
## 2018        24.489919      a
## 2021        22.201613     ab
## 2014        15.139113     bc
## 2015        12.368952     cd
## 2012        11.436828     cd
## 2013         6.929598      d
```

```
# Plot with one-way ANOVA letters
herb.ctrl.letters <- hsd.herb.ctrl$groups
herb.ctrl.letters <- herb.ctrl.letters |>
  mutate(Year = rownames(herb.ctrl.letters)) |>
  arrange(Year)
herb.trt.letters <- hsd.herb.trt$groups
herb.trt.letters <-herb.trt.letters |>
  mutate(Year = rownames(herb.trt.letters)) |>
  arrange(Year)

letters.herb3t <- data.frame(x = herb.avg$year.xaxis[1:6],
                             y = c(9, 5.5, 12.5, 10, 28.5, 18.3),
                             label = herb.trt.letters$groups)
letters.herb3c <- data.frame(x = herb.avg$year.xaxis[1:6],
                             y = c(23.5, 20.5, 26.5, 17, 17, 31),
                             label = herb.ctrl.letters$groups)

ptext.herb3 <- data.frame(x = as.Date("2019-09-01"),
                          y = 8,
                          label = "ANOVA, p < 0.001")
herb.plot3 <- ggplot(herb.avg, aes(x = year.xaxis, y = mean,
                                   color = Treatment)) +
  geom_line() +
  geom_point() +
  geom_pointrange(aes(ymin = mean - SE, ymax = mean + SE)) +
  xlab(NULL) +
```

```
  ylab("Cover (%)") +
  ggtitle("Herbaceous cover") +
  scale_color_manual(values = c("red", "#1F78B4")) +
  theme_bw() +
  theme(legend.position = "bottom") +
  geom_text(data = letters.herb3t,
            mapping = aes(x = x, y = y, label = label),
            color = "#1F78B4",
            size = 3) +
  geom_text(data = letters.herb3c,
            mapping = aes(x = x, y = y, label = label),
            color = "red",
            size = 3,
            fontface = "italic") +
  geom_text(data = ptext.herb3,
            aes(x = x, y = y, label = label),
            color = "gray30",
            size = 2.5) +
  theme(axis.text.x = element_text(color = "black")) +
  theme(plot.margin = margin(t = 0.1, r = 0.1, b = 0.3, l = 0.1, "in")) +
  theme(legend.position = "none")
herb.plot3
```


Herbaceous cover

Fig 3c: Shrub cover
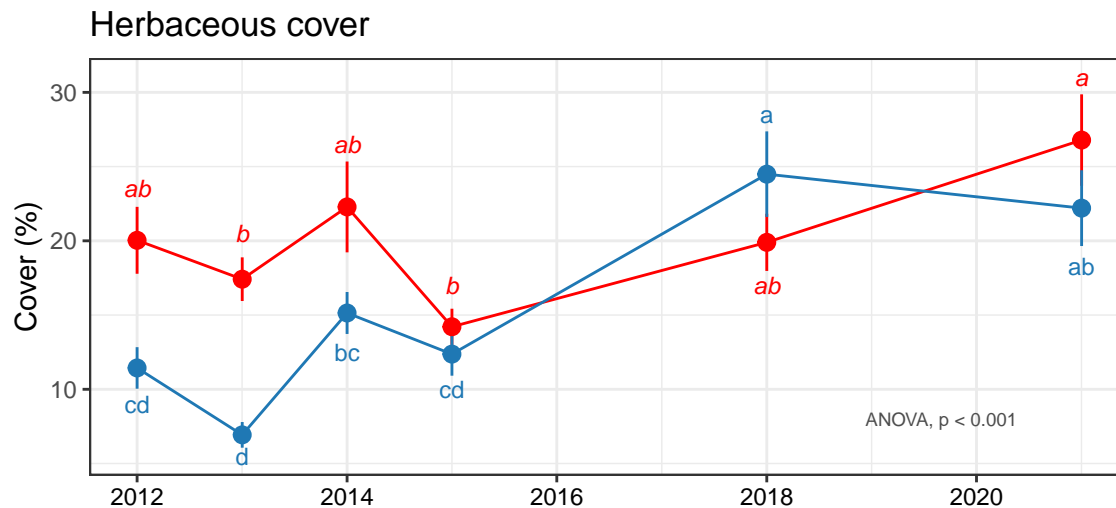
```
# Shrub cover ---------------------------------------------------------------

# Find averages by year
shrub.avg <- shrub.all %>%
  group_by(Treatment, Year, year.xaxis) %>%
  summarise(mean = mean(Cover),
            SD = sd(Cover),
```

```
            SE = std.error(Cover),
            .groups = "keep")

# One-way ANOVA for Control
summary(aov(Cover ~ Year, data = filter(shrub.all, Treatment == "Control"))) # p = 0.0112
```

```
##               Df Sum Sq Mean Sq F value Pr(>F)
## Year           5   1566   313.1   3.062 0.0112 *
## Residuals    177  18100   102.3
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
shrub.ctrl <- shrub.all |>
  filter(Treatment == "Control")
anova.shrub.ctrl <- aov(shrub.ctrl$Cover ~ shrub.ctrl$Year)
hsd.shrub.ctrl <- HSD.test(anova.shrub.ctrl, trt = "shrub.ctrl$Year")
hsd.shrub.ctrl$groups
```

```
##       shrub.ctrl$Cover groups
## 2021         15.633065      a
## 2012         11.697222     ab
## 2013         10.297222     ab
## 2014          9.304167     ab
## 2018          7.820565      b
## 2015          6.681452      b
```

```
# One-way ANOVA for Treated
summary(aov(Cover ~ Year, data = filter(shrub.all, Treatment == "Treated"))) # NS, p = 0.982
```

```
##               Df Sum Sq Mean Sq F value Pr(>F)
## Year           5    232    46.4   0.141  0.982
## Residuals    178  58465   328.5
```

```
# Plot with one-way ANOVA letters
shrub.ctrl.letters <- hsd.shrub.ctrl$groups
shrub.ctrl.letters <- shrub.ctrl.letters |>
  mutate(Year = rownames(shrub.ctrl.letters)) |>
  arrange(Year)

letters.shrub2 <- data.frame(x = shrub.avg$year.xaxis[1:6],
                             y = c(9, 8, 7.4, 4.2, 5.5, 11.8),
                             label = shrub.ctrl.letters$groups)

ptext.shrub2 <- data.frame(x = as.Date("2019-09-01"),
                           y = 6,
                           label = "ANOVA, p = 0.982 \nANOVA, p = 0.011")
shrub.plot2 <- ggplot(shrub.avg, aes(x = year.xaxis, y = mean,
                                     color = Treatment)) +
  geom_line() +
  geom_point() +
  geom_pointrange(aes(ymin = mean - SE, ymax = mean + SE)) +
```

```
  xlab(NULL) +
  ylab("Cover (%)") +
  ggtitle("Shrub cover") +
  scale_color_manual(values = c("red", "#1F78B4")) +
  theme_bw() +
  theme(legend.position = "bottom") +
  geom_text(data = letters.shrub2,
            mapping = aes(x = x, y = y, label = label),
            color = "red",
            size = 3,
            fontface = "italic") +
  geom_text(data = ptext.shrub2,
            aes(x = x, y = y, label = label),
            color = "gray30",
            size = 2.5) +
  theme(axis.text.x = element_text(color = "black")) +
  theme(plot.margin = margin(t = 0.1, r = 0.1, b = 0, l = 0.1, "in")) +
  theme(legend.title = element_blank())
shrub.plot2
```
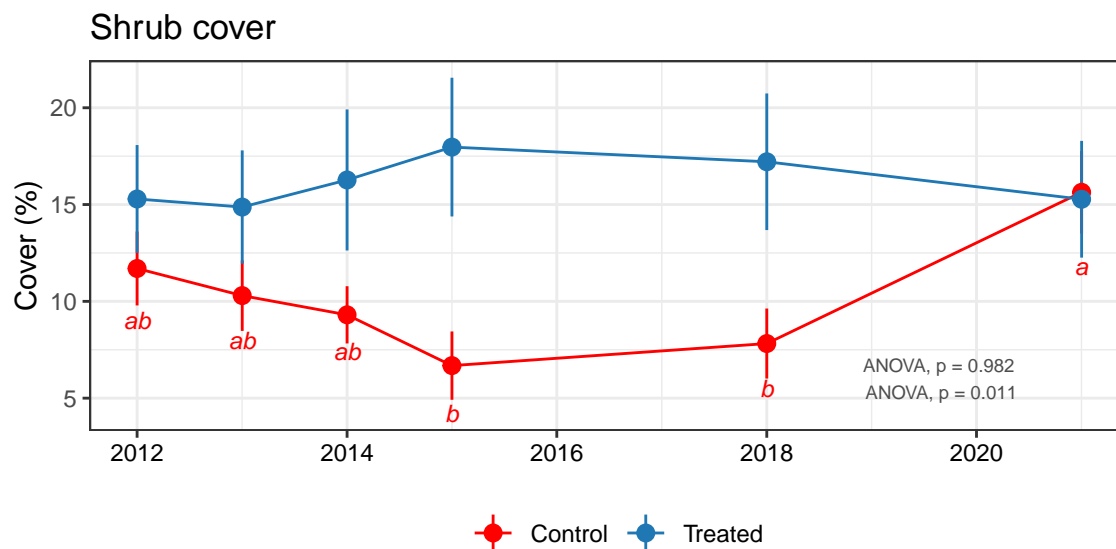

Combine plots for Fig 3

```
# Combine notree, herb & shrub -------------------------------------------

tiff("figures/2023-12_publish-figures/Fig3_temporal_notree-herb-shrub.tiff", units = "in", height = 8.5
ggarrange(notree.plot2, herb.plot3, shrub.plot2,
          ncol = 1, nrow = 3,
          labels = c("(A)", "(B)", "(C)"))

dev.off()
```

Supp Fig 3a: Perennial plant species richness

```r
# Perennial plant richness --------------------------------------------------

# Find averages by year
rich.avg <- per.div %>%
  group_by(Treatment, Year, year.xaxis) %>%
  summarise(mean = mean(rich),
            SD = sd(rich),
            SE = std.error(rich),
            .groups = "keep")

# One-way ANOVA for Control
summary(aov(rich ~ Year, data = filter(per.div, Treatment == "Control"))) # 0.00881
```

```
##              Df Sum Sq Mean Sq F value  Pr(>F)
## Year          5   92.9  18.587   3.189 0.00881 **
## Residuals   177 1031.7   5.829
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```r
rich.ctrl <- per.div |>
  filter(Treatment == "Control")
anova.rich.ctrl <- aov(rich.ctrl$rich ~ rich.ctrl$Year)
hsd.rich.ctrl <- HSD.test(anova.rich.ctrl, trt = "rich.ctrl$Year")
hsd.rich.ctrl$groups
```

```
##      rich.ctrl$rich groups
## 2012       9.866667      a
## 2013       9.133333     ab
## 2018       8.645161     ab
## 2015       8.612903     ab
## 2014       8.200000     ab
## 2021       7.580645      b
```

```r
# One-way ANOVA for Treated
summary(aov(rich ~ Year, data = filter(per.div, Treatment == "Treated"))) # p = 0.0516
```

```
##              Df Sum Sq Mean Sq F value Pr(>F)
## Year          5     76  15.207   2.247 0.0516 .
## Residuals   178   1204   6.766
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```r
# Plot with one-way ANOVA letters
rich.ctrl.letters <- hsd.rich.ctrl$groups
rich.ctrl.letters <- rich.ctrl.letters |>
  mutate(Year = rownames(rich.ctrl.letters)) |>
  arrange(Year)

letters.rich <- data.frame(x = rich.avg$year.xaxis[1:6],
```

```
                                  y = rep(10.4, 6),
                                  label = rich.ctrl.letters$groups,
                                  Treatment = rep("Control", 6))
ptext.rich <- data.frame(x = rep(as.Date("2019-09-01"), 2),
                                  y = c(6.7, 10.3),
                                  label = c("ANOVA, p = 0.009", "ANOVA, p = 0.052"),
                                  Treatment = c("Control", "Treated"))


rich.plot <- ggplot(rich.avg, aes(x = year.xaxis, y = mean,
                                  group = Treatment,
                                  color = Treatment)) +
  geom_line() +
  geom_point() +
  geom_pointrange(aes(ymin = mean - SE, ymax = mean + SE)) +
  facet_wrap(~Treatment) +
  xlab(NULL) +
  ylab("No. of species") +
  ggtitle("Perennial plant species richness, 2012-2021") +
  scale_color_manual(values = c("red", "#1F78B4")) +
  theme_bw() +
  theme(legend.position = "none") +
  geom_text(data = letters.rich,
            mapping = aes(x = x, y = y, label = label),
            color = "black",
            size = 3.5) +
  geom_text(data = ptext.rich,
            aes(x = x, y = y, label = label),
            color = "gray30",
            size = 2.5) +
  theme(axis.text.x = element_text(color = "black")) +
  theme(plot.margin = margin(0.1, 0.1, 0.2, 0.1, "in"))
rich.plot
```
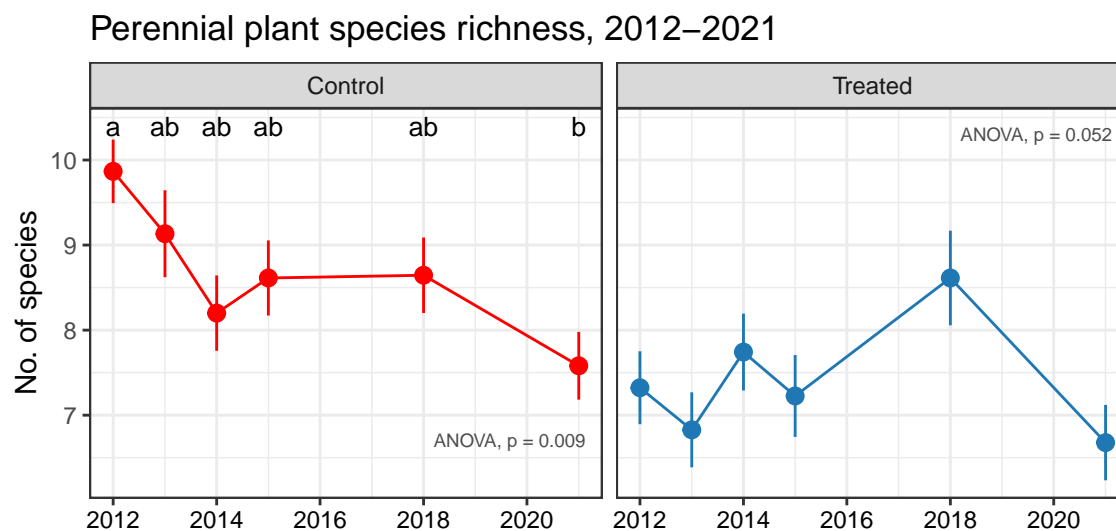

Perennial plant species richness, 2012–2021

Supp Fig 3b: Perennial plant diversity

```r
# Perennial plant diversity (Shannon) -------------------------------------

# Find averages by year
shan.avg <- per.div %>%
  group_by(Treatment, Year, year.xaxis) %>%
  summarise(mean = mean(shan),
            SD = sd(shan),
            SE = std.error(shan),
            .groups = "keep")

# One-way ANOVA for Control
summary(aov(shan ~ Year, data = filter(per.div, Treatment == "Control"))) # p = 0.934
```

```
##              Df Sum Sq Mean Sq F value Pr(>F)
## Year          5  0.185 0.03702    0.26  0.934
## Residuals   177 25.197 0.14235
```

```r
# One-way ANOVA for Treated
summary(aov(shan ~ Year, data = filter(per.div, Treatment == "Treated"))) # p = 0.725
```

```
##              Df Sum Sq Mean Sq F value Pr(>F)
## Year          5   0.59  0.1189   0.568  0.725
## Residuals   178  37.27  0.2094
```

```r
# Plot with one-way ANOVA
ptext.shan <- data.frame(x = rep(as.Date("2019-09-01"), 2),
                         y = c(1.33, 1.33),
                         label = c("ANOVA, p = 0.934", "ANOVA, p = 0.725"),
                         Treatment = c("Control", "Treated"))

shan.plot <- ggplot(shan.avg, aes(x = year.xaxis, y = mean,
                                  group = Treatment,
                                  color = Treatment)) +
  geom_line() +
  geom_point() +
  geom_pointrange(aes(ymin = mean - SE, ymax = mean + SE)) +
  facet_wrap(~Treatment) +
  xlab(NULL) +
  ylab("Shannon diversity index") +
  ggtitle("Perennial plant diversity") +
  scale_color_manual(values = c("red", "#1F78B4")) +
  theme_bw() +
  theme(legend.position = "none") +
  theme(axis.text.x = element_text(color = "black")) +
  geom_text(data = ptext.shan,
            aes(x = x, y = y, label = label),
            color = "gray30",
            size = 2.5) +
  theme(plot.margin = margin(0.1, 0.1, 0.2, 0.1, "in"))
shan.plot
```

Perennial plant diversity
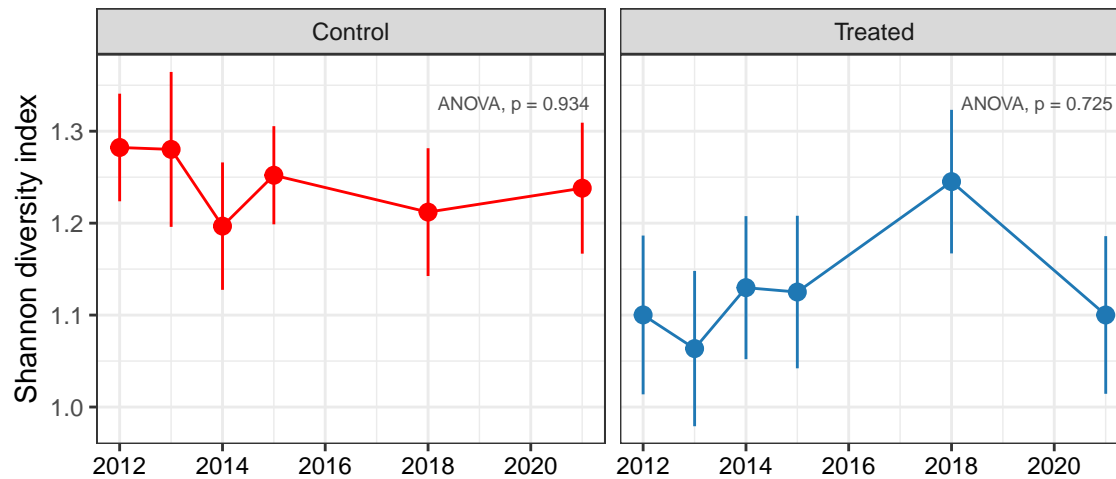
Combine plots for Supp Fig 3

```r
# Combine richness & Shannon ---------------------------------------------

tiff("FigS3_temporal_richness-Shannon.tiff", units = "in", height = 5.5, width = 6, res = 300)
ggarrange( rich.plot, shan.plot,
          ncol = 1, nrow = 2,
          labels = c("(A)", "(B)"))

dev.off()
```

Invasive cover and most common species

```r
# Average cover by year
invasive.all %>%
  select(-year.xaxis) |>
  group_by(Treatment, Year) %>%
  summarise(mean = mean(Cover),
            SE = std.error(Cover),
            .groups = "keep") |>
  mutate_if(is.numeric, round, digits = 2)
```

```
## `mutate_if()` ignored the following grouping variables:
## * Columns `Treatment`, `Year`

## # A tibble: 12 x 4
## # Groups:   Treatment, Year [12]
##    Treatment Year   mean    SE
##    <fct>     <fct> <dbl> <dbl>
## 1 Control    2012   3.34  0.6
```

```
##  2 Control     2013    2.58   0.66
##  3 Control     2014    2.46   0.98
##  4 Control     2015    2.96   0.69
##  5 Control     2018    5.1    1.17
##  6 Control     2021    7.38   1.23
##  7 Treated     2012    3.29   0.84
##  8 Treated     2013    2.18   0.51
##  9 Treated     2014    3.58   0.8
## 10 Treated     2015    5.1    1.08
## 11 Treated     2018    5.17   1.23
## 12 Treated     2021    4.79   0.85
```

```r
# Most common species in Control
plant.all |>
  filter(Native == "Invasive",
         Treatment == "Control") |>
  group_by(Common) |>
  summarise(mean = mean(Cover)) |>
  arrange(desc(mean))
```

```
## # A tibble: 4 x 2
##   Common               mean
##   <chr>               <dbl>
## 1 Lehmann lovegrass   4.49
## 2 Stinkgrass          0.833
## 3 Spreading fantails  0.769
## 4 African lovegrass   0.125
```

```r
# Most common species in Treated
plant.all |>
  filter(Native == "Invasive",
         Treatment == "Treated") |>
  group_by(Common) |>
  summarise(mean = mean(Cover)) |>
  arrange(desc(mean))
```

```
## # A tibble: 8 x 2
##   Common               mean
##   <chr>               <dbl>
## 1 Lehmann lovegrass   4.75
## 2 Buffelgrass         3.41
## 3 Boer lovegrass      0.625
## 4 African lovegrass   0.531
## 5 Spreading fantails  0.367
## 6 Barnyard            0.225
## 7 Rose Natal grass    0.125
## 8 Stinkgrass          0.125
```

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Appendix B: Coefficient of variation

Setup

```r
library(tidyverse)
library(car)
library(scales)
library(ggpubr)
```

```r
# Load data ------------------------------------------------------------

notree.all <- read_csv("Herb-and-shrub-cover_2012-2021.csv")
herb.all <- read_csv("Herb-cover_2012-2021.csv")
shrub.all <- read_csv("Shrub-cover_2012-2021.csv")
per.div <- read_csv("Perennial-plant-diversity_2012-2021.csv")
```

Fig 4a: CV of shrub cover

```r
# Shrub cover ----------------------------------------------------------

# Find CV for each sample over time
shrub.sample <- shrub.all |>
  group_by(Sample, Treatment) |>
  summarise(CV = sd(Cover) / mean(Cover),
            .groups = "keep") # NaNs produced because some have 0 cover and can't divide by 0

# Replace NaNs with 0
shrub.sample[1, 3] <- 0
shrub.sample[5, 3] <- 0
shrub.sample[8, 3] <- 0


# Compare means
wilcox.test(filter(shrub.sample, Treatment == "Treated")$CV,
            filter(shrub.sample, Treatment == "Control")$CV,
            exact = FALSE) # p = 0.01429
```

```
##
##  Wilcoxon rank sum test with continuity correction
##
## data:  filter(shrub.sample, Treatment == "Treated")$CV and filter(shrub.sample, Treatment == "Control
## W = 306, p-value = 0.01429
## alternative hypothesis: true location shift is not equal to 0
```

```r
# Plot
letters.shrub <- data.frame(x = c(1, 2),
                            y = c(2.5, 2.5),
                            label = c("a", "b"))
```

```r
shrub.plot.cv <- shrub.sample |>
  ggplot(aes(x = Treatment, y = CV)) +
  geom_boxplot(alpha = 0.3,
               outlier.shape = NA,
               aes(fill = Treatment)) +
  geom_jitter(size = 1,
              alpha = 0.8,
              aes(color = Treatment)) +
  scale_color_manual(values = c("red", "#1F78B4")) +
  scale_fill_manual(values = c("red", "#1F78B4")) +
  labs(title = "Shrub cover",
       x = NULL,
       y = "Coefficient of variation") +
  theme_bw() +
  theme(legend.position = "none") +
  scale_y_continuous(labels = percent)  +
  theme(axis.text.x = element_text(color = "black")) +
  geom_text(aes(x = 0.95, y = 2.75, label = "Mann-Whitney, \np = 0.014"),
            color = "gray30",
            size = 2.5) +
  geom_text(data = letters.shrub,
            aes(x = x, y = y, label = label),
            color = "black") +
  theme(plot.margin = margin(0.1, 0, 0.1, 0.1, "in")) +
  stat_summary(fun = mean, geom = "errorbar", aes(ymax = after_stat(y), ymin = after_stat(y)),
               width = 0.75, linetype = "dashed") +
  theme(plot.title = element_text(size = 12))
shrub.plot.cv
```
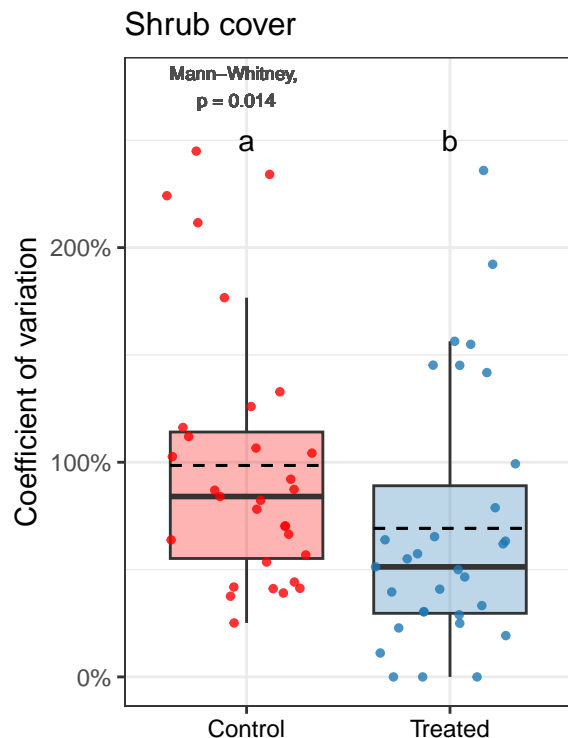
Fig 4b: CV of herbaceous cover

```r
# Herb cover ------------------------------------------------------------

# Find CV for each sample over time
herb.sample <- herb.all |>
  group_by(Sample, Treatment) |>
  summarise(CV = sd(Cover) / mean(Cover),
            .groups = "keep")


# Compare means
wilcox.test(filter(herb.sample, Treatment == "Treated")$CV,
            filter(herb.sample, Treatment == "Control")$CV) # NS, p = 0.148
```

```
##
##  Wilcoxon rank sum exact test
##
## data:  filter(herb.sample, Treatment == "Treated")$CV and filter(herb.sample, Treatment == "Control")
## W = 584, p-value = 0.1479
## alternative hypothesis: true location shift is not equal to 0
```

```r
# Plot
herb.plot.cv <- herb.sample |>
  ggplot(aes(x = Treatment, y = CV)) +
  geom_boxplot(alpha = 0.3,
               outlier.shape = NA,
               aes(fill = Treatment)) +
  geom_jitter(size = 1,
              alpha = 0.8,
              aes(color = Treatment)) +
  scale_color_manual(values = c("red", "#1F78B4")) +
  scale_fill_manual(values = c("red", "#1F78B4")) +
  labs(title = "Herbaceous cover",
       x = NULL,
       y = NULL) +
  theme_bw() +
  theme(legend.position = "none") +
  scale_y_continuous(labels = percent)  +
  theme(axis.text.x = element_text(color = "black")) +
  geom_text(aes(x = 0.95, y = 1.18, label = "Mann-Whitney, \np = 0.148"),
            color = "gray30",
            size = 2.5) +
  theme(plot.margin = margin(0.1, 0.1, 0.1, 0.2, "in")) +
  stat_summary(fun = mean, geom = "errorbar", aes(ymax = after_stat(y), ymin = after_stat(y)),
               width = 0.75, linetype = "dashed") +
  theme(plot.title = element_text(size = 12))
herb.plot.cv
```
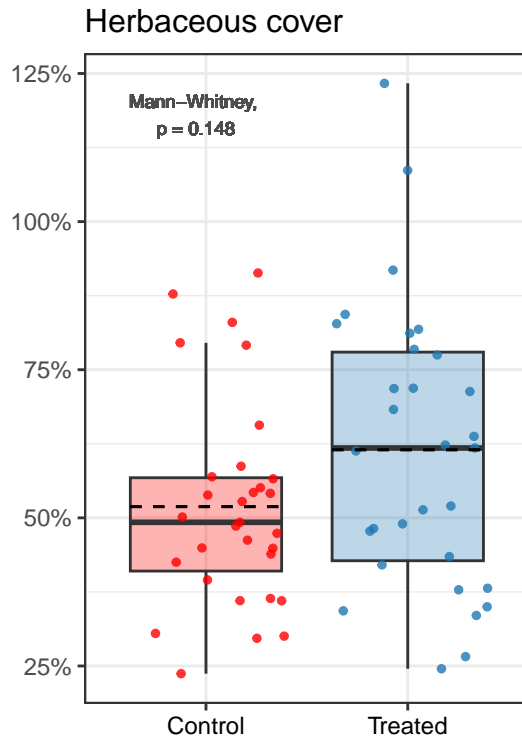
Herbaceous cover

Mann–Whitney,
p = 0.148

Fig 4c: CV of overall veg cover

```
# Notree cover -------------------------------------------------------------

# Find CV for each sample over time
notree.sample <- notree.all |>
  group_by(Sample, Treatment) |>
  summarise(CV = sd(Cover) / mean(Cover),
            .groups = "keep")

# Compare means
t.test(filter(notree.sample, Treatment == "Treated")$CV,
       filter(notree.sample, Treatment == "Control")$CV) # NS, p = 0.882
```

```
##
##  Welch Two Sample t-test
##
## data:  filter(notree.sample, Treatment == "Treated")$CV and filter(notree.sample, Treatment == "Cont
## t = -0.14868, df = 59.846, p-value = 0.8823
## alternative hypothesis: true difference in means is not equal to 0
## 95 percent confidence interval:
##  -0.11574119  0.09972676
## sample estimates:
## mean of x mean of y
## 0.4430859 0.4510931
```

```
# Plot
notree.plot.cv <- notree.sample |>
  ggplot(aes(x = Treatment, y = CV)) +
  geom_boxplot(alpha = 0.3,
               outlier.shape = NA,
               aes(fill = Treatment)) +
  geom_jitter(size = 1,
              alpha = 0.8,
              aes(color = Treatment)) +
  scale_color_manual(values = c("red", "#1F78B4")) +
  scale_fill_manual(values = c("red", "#1F78B4")) +
  labs(title = "Vegetation cover",
       x = NULL,
       y = NULL) +
  theme_bw() +
  theme(legend.position = "none") +
  scale_y_continuous(labels = percent) +
  theme(axis.text.x = element_text(color = "black")) +
  geom_text(aes(x = 0.9, y = 0.95, label = "t-test, p = 0.882"),
            color = "gray30",
            size = 2.5) +
  theme(plot.margin = margin(0.1, 0.1, 0.1, 0.2, "in")) +
  stat_summary(fun = mean, geom = "errorbar", aes(ymax = after_stat(y), ymin = after_stat(y)),
               width = 0.75, linetype = "dashed") +
  theme(plot.title = element_text(size = 12))
notree.plot.cv
```


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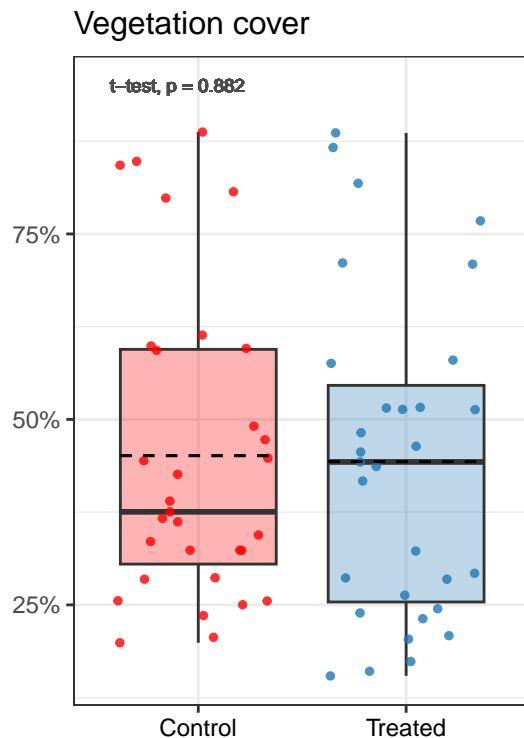
Combine plots for Fig 4

```r
# Combine notree, herb, shrub -------------------------------------------------

tiff("Fig4_CV_shrub-herb-notree.tiff", units = "in", height = 4, width = 7, res = 1000)
ggarrange(shrub.plot.cv, herb.plot.cv, notree.plot.cv,
          ncol = 3, nrow = 1,
          labels = c("(A)", "(B)", "(C)"))

dev.off()
```

Supp Fig 4a: CV of perennial richness

```r
# Richness ---------------------------------------------------------------------

# Find CV for each sample over time
rich.sample <- per.div |>
  group_by(Sample, Treatment) |>
  summarise(CV = sd(rich) / mean(rich),
            .groups = "keep")

# Compare means
wilcox.test(filter(rich.sample, Treatment == "Treated")$CV,
            filter(rich.sample, Treatment == "Control")$CV) # NS, p = 0.138
```

```
## Warning in wilcox.test.default(filter(rich.sample, Treatment == "Treated")$CV,
## : cannot compute exact p-value with ties


##
##  Wilcoxon rank sum test with continuity correction
##
## data:  filter(rich.sample, Treatment == "Treated")$CV and filter(rich.sample, Treatment == "Control")
## W = 586.5, p-value = 0.1375
## alternative hypothesis: true location shift is not equal to 0
```

```r
# Plot
rich.plot.cv <- rich.sample |>
  ggplot(aes(x = Treatment, y = CV)) +
  geom_boxplot(alpha = 0.3,
               outlier.shape = NA,
               aes(fill = Treatment)) +
  geom_jitter(size = 1,
              alpha = 0.8,
              aes(color = Treatment)) +
  scale_color_manual(values = c("red", "#1F78B4")) +
  scale_fill_manual(values = c("red", "#1F78B4")) +
  labs(title = "Perennial plant species \nrichness",
       x = NULL,
       y = "Coefficient of variation") +
  theme_bw() +
```

```r
  theme(legend.position = "none") +
  scale_y_continuous(labels = percent) +
  theme(axis.text.x = element_text(color = "black")) +
  geom_text(aes(x = 0.95, y = 0.65, label = "Mann-Whitney, \np = 0.138"),
            color = "gray30",
            size = 2.5) +
  theme(plot.margin = margin(0.1, 0.1, 0.1, 0.1, "in")) +
  stat_summary(fun = mean, geom = "errorbar", aes(ymax = after_stat(y), ymin = after_stat(y)),
               width = 0.75, linetype = "dashed") +
  theme(plot.title = element_text(size = 12))
rich.plot.cv
```

Perennial plant species richness

Supp Fig 4b: CV of perennial diversity

```r
# Shannon ------------------------------------------------------------------

# Find CV for each sample over time
shan.sample <- per.div |>
  group_by(Sample, Treatment) |>
  summarise(CV = sd(shan) / mean(shan),
            .groups = "keep")

# Compare
wilcox.test(filter(shan.sample, Treatment == "Treated")$CV,
            filter(shan.sample, Treatment == "Control")$CV,
            exact = FALSE) # NS, p = 0.100
```
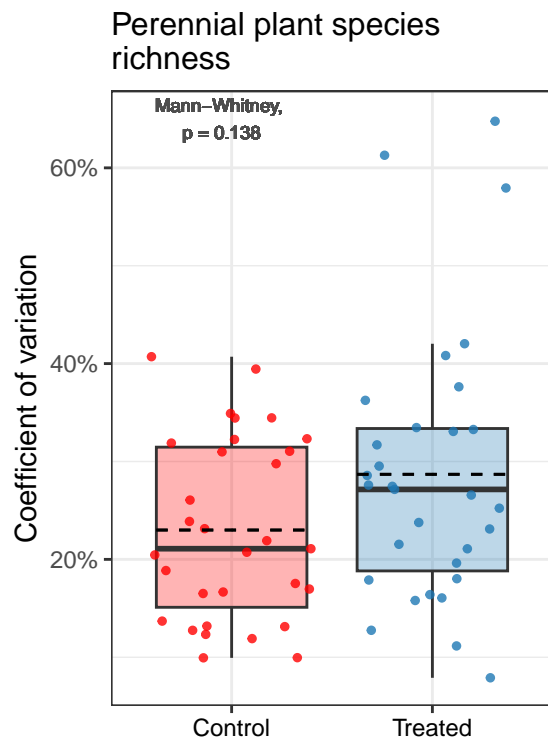
```
## 
##  Wilcoxon rank sum test with continuity correction
## 
## data:  filter(shan.sample, Treatment == "Treated")$CV and filter(shan.sample, Treatment == "Control")
## W = 598, p-value = 0.09952
## alternative hypothesis: true location shift is not equal to 0
```

```r
# Plot
shan.plot.cv <- shan.sample |>
  ggplot(aes(x = Treatment, y = CV)) +
  geom_boxplot(alpha = 0.3,
               outlier.shape = NA,
               aes(fill = Treatment)) +
  geom_jitter(size = 1,
              alpha = 0.8,
              aes(color = Treatment)) +
  scale_color_manual(values = c("red", "#1F78B4")) +
  scale_fill_manual(values = c("red", "#1F78B4")) +
  labs(title = "Perennial plant diversity",
       x = NULL,
       y = NULL) +
  theme_bw() +
  theme(legend.position = "none") +
  scale_y_continuous(labels = percent) +
  theme(axis.text.x = element_text(color = "black")) +
  geom_text(aes(x = 0.95, y = 0.83, label = "Mann-Whitney, \np = 0.100"),
            color = "gray30",
            size = 2.5) +
  theme(plot.margin = margin(0.1, 0.1, 0.1, 0.15, "in")) +
  stat_summary(fun = mean, geom = "errorbar", aes(ymax = after_stat(y), ymin = after_stat(y)),
               width = 0.75, linetype = "dashed") +
  theme(plot.title = element_text(size = 12))
shan.plot.cv
```

Perennial plant diversity

Mann–Whitney,
p = 0.100
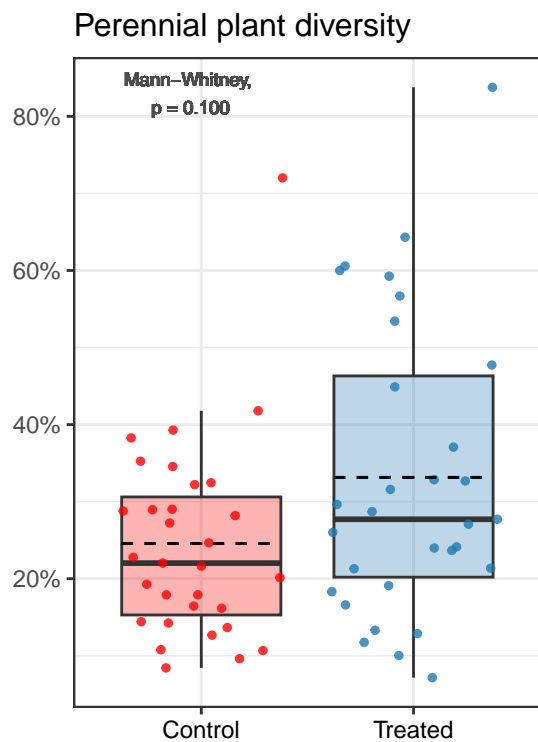
Combine plots for Supp Fig 4

```r
# Combine richness & Shannon ---------------------------------------------

# Supplemental figure
tiff("FigS4_CV_rich-shan.tiff", units = "in", height = 4, width = 5.5, res = 300)
ggarrange(rich.plot.cv, shan.plot.cv,
          ncol = 2, nrow = 1,
          labels = c("(A)", "(B)"))

dev.off()
```

Appendix C: Soil fertility

Setup

```r
library(tidyverse)
library(ggpubr)
library(metagenomeSeq)
library(vegan)
```

```r
# Load data -----------------------------------------------------------

barc.asv <- read.table("bac-arc_clean_asv.txt", sep = "\t",
                        header = T, row.names = 1)
fungi.asv <- read.table("fungi_clean_asv.txt",
                        sep = "\t", header = T, row.names = 1)
meta <- read.csv("sequencing_metadata.csv")

dat.2021 <- read.csv("Veg-soil-elev_2021.csv")
```

```r
# 16S -----------------------------------------------------------------

# Normalization
barc.MR <- newMRexperiment(t(barc.asv))
p <- cumNormStat(barc.MR)
```

```r
## Default value being used.
```

```r
barc.MR <- cumNorm(barc.MR, p = p)
barc.norm <- t(MRcounts(barc.MR, norm = T, log = F))

# Richness and Shannon
meta$barc.richness <- specnumber(barc.norm)
meta$barc.shannon <- diversity(barc.norm, index = "shannon")

# Bray-Curtis distance
barc.dist <- vegdist(barc.norm, method = "bray")
```

```r
# ITS -----------------------------------------------------------------

# Normalization
fungi.MR <- newMRexperiment(t(fungi.asv))
p <- cumNormStat(fungi.MR)
```

```r
## Default value being used.
```

```r
fungi.MR <- cumNorm(fungi.MR, p = p)
fungi.norm <- t(MRcounts(fungi.MR, norm = T, log = F))

# Richness and Shannon
```

```
meta$fungi.richness <- specnumber(fungi.norm)
meta$fungi.shannon <- diversity(fungi.norm, index = "shannon")

# Bray-Curtis distance
fungi.dist <- vegdist(fungi.norm, method = "bray")
```

Fig 5a: Bacteria & archaea NMDS ordination

Note that PERMANOVA model results (`adonis2`) will vary slightly each time and will not exactly match values published in the paper.

```
# NMDS ordination
barc.nmds <- metaMDS(barc.dist, k = 2)
```

```
## Run 0 stress 0.1684425
## Run 1 stress 0.1741199
## Run 2 stress 0.1832087
## Run 3 stress 0.1680461
## ... New best solution
## ... Procrustes: rmse 0.04031523  max resid 0.2924091
## Run 4 stress 0.1703671
## Run 5 stress 0.2005623
## Run 6 stress 0.170816
## Run 7 stress 0.1682434
## ... Procrustes: rmse 0.01454679  max resid 0.09735784
## Run 8 stress 0.1926389
## Run 9 stress 0.1707367
## Run 10 stress 0.1698914
## Run 11 stress 0.1702923
## Run 12 stress 0.1680366
## ... New best solution
## ... Procrustes: rmse 0.002872819  max resid 0.01729991
## Run 13 stress 0.1752061
## Run 14 stress 0.1842293
## Run 15 stress 0.1752072
## Run 16 stress 0.178603
## Run 17 stress 0.1736229
## Run 18 stress 0.1817014
## Run 19 stress 0.1773086
## Run 20 stress 0.167942
## ... New best solution
## ... Procrustes: rmse 0.01258909  max resid 0.07412873
## *** Best solution was not repeated -- monoMDS stopping criteria:
##      1: no. of iterations >= maxit
##     19: stress ratio > sratmax
```

```
barc.nmds$stress
```

```
## [1] 0.167942
```

```
meta$barc.NMDS1 <- barc.nmds$points[ , 1]
meta$barc.NMDS2 <- barc.nmds$points[ , 2]

# PERMANOVA
adonis2(barc.dist ~ meta$Treatment)
```

```
## Permutation test for adonis under reduced model
## Terms added sequentially (first to last)
## Permutation: free
## Number of permutations: 999
##
## adonis2(formula = barc.dist ~ meta$Treatment)
##                  Df SumOfSqs      R2      F Pr(>F)
## meta$Treatment    1   0.2559 0.02881 1.7799  0.029 *
## Residual         60   8.6275 0.97119
## Total            61   8.8835 1.00000
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
# Plot (using dat.2021 for same NMDS values presented in paper):
barc.nmds.plot.21 <- dat.2021 %>%
  ggplot(aes(x = barc.NMDS1, y = barc.NMDS2, color = Treatment, shape = Treatment)) +
  geom_point() +
  scale_color_manual(values = c("red", "#1F78B4")) +
  theme_bw() +
  labs(x = "Axis 1",
       y = "Axis 2",
       title = "Bacteria & archaea",
       color = "Treatment",
       shape = "Treatment") +
  theme(legend.position = "bottom") +
  theme(plot.margin = margin(0.1, 0.2, 0.1, 0.1, "in")) +
  theme(legend.title = element_blank()) +
  geom_text(aes(x = 0.2, y = -0.55, label = "PERMANOVA, p = 0.029"),
            size = 2.5, color = "gray30") +
  geom_text(aes(x = 0.25, y = -0.65, label = "Stress = 0.168"),
            size = 2.5, color = "gray30") +
  theme(plot.title = element_text(size = 12))
barc.nmds.plot.21
```

Bacteria & archaea
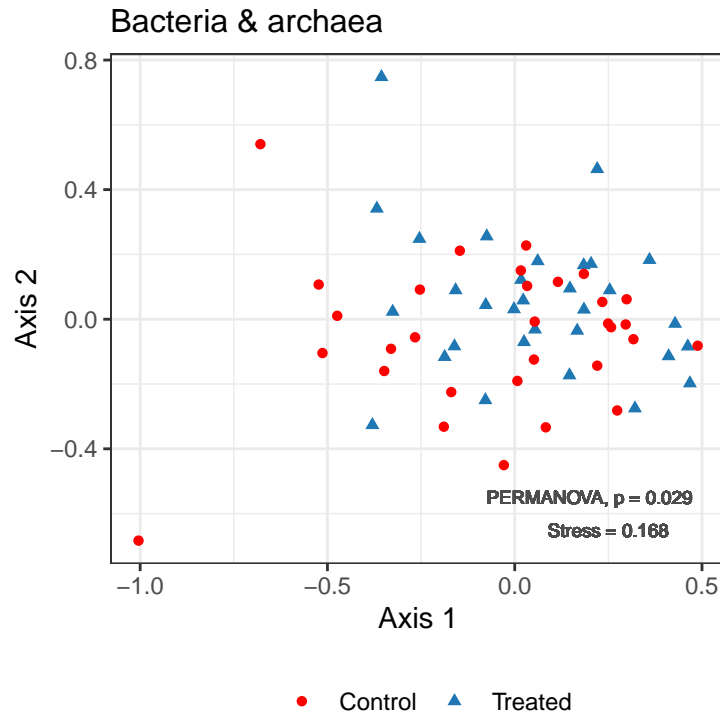
Fig 5b: Fungi NMDS ordination

Note that PERMANOVA model results (`adonis2`) will vary slightly each time and will not exactly match values published in the paper.

```r
# NMDS ordination
fungi.nmds <- metaMDS(fungi.dist, k = 2)
```

```
## Run 0 stress 0.2461785
## Run 1 stress 0.2468725
## Run 2 stress 0.2743682
## Run 3 stress 0.2383982
## ... New best solution
## ... Procrustes: rmse 0.105665  max resid 0.4568876
## Run 4 stress 0.2462807
## Run 5 stress 0.2628147
## Run 6 stress 0.2374924
## ... New best solution
## ... Procrustes: rmse 0.05403049  max resid 0.1819509
## Run 7 stress 0.2469342
## Run 8 stress 0.237575
## ... Procrustes: rmse 0.0540305  max resid 0.2373218
## Run 9 stress 0.2514936
## Run 10 stress 0.2408483
## Run 11 stress 0.2449272
## Run 12 stress 0.2414653
## Run 13 stress 0.2414881
## Run 14 stress 0.2554275
```

```
## Run 15 stress 0.24388
## Run 16 stress 0.2718033
## Run 17 stress 0.2787875
## Run 18 stress 0.2483354
## Run 19 stress 0.2442429
## Run 20 stress 0.2418738
## *** Best solution was not repeated -- monoMDS stopping criteria:
##      3: no. of iterations >= maxit
##     17: stress ratio > sratmax
```

```
fungi.nmds$stress
```

```
## [1] 0.2374924
```

```
meta$fungi.NMDS1 <- fungi.nmds$points[ , 1]
meta$fungi.NMDS2 <- fungi.nmds$points[ , 2]

# PERMANOVA
adonis2(fungi.dist ~ meta$Treatment)
```

```
## Permutation test for adonis under reduced model
## Terms added sequentially (first to last)
## Permutation: free
## Number of permutations: 999
##
## adonis2(formula = fungi.dist ~ meta$Treatment)
##                Df SumOfSqs      R2     F Pr(>F)
## meta$Treatment  1   0.4511 0.02371 1.457  0.012 *
## Residual       60  18.5783 0.97629
## Total          61  19.0294 1.00000
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
# Plot (using dat.2021 for same NMDS values presented in paper):
fungi.nmds.plot.21 <- dat.2021 %>%
  ggplot(aes(x = fungi.NMDS1, y = fungi.NMDS2, color = Treatment, shape = Treatment)) +
  geom_point() +
  scale_color_manual(values = c("red", "#1F78B4")) +
  theme_bw() +
  labs(x = "Axis 1",
       y = "Axis 2",
       title = "Fungi",
       color = "Treatment",
       shape = "Treatment") +
  theme(legend.position = "bottom") +
  theme(legend.title = element_blank()) +
  theme(plot.margin = margin(0.1, 0.2, 0.1, 0.1, "in")) +
  geom_text(aes(x = 0.73, y = -0.7, label = "PERMANOVA, p = 0.011"),
            size = 2.5, color = "gray30") +
  geom_text(aes(x = 0.82, y = -0.8, label = "Stress = 0.236"),
            size = 2.5, color = "gray30") +
  theme(plot.title = element_text(size = 12))
fungi.nmds.plot.21
```
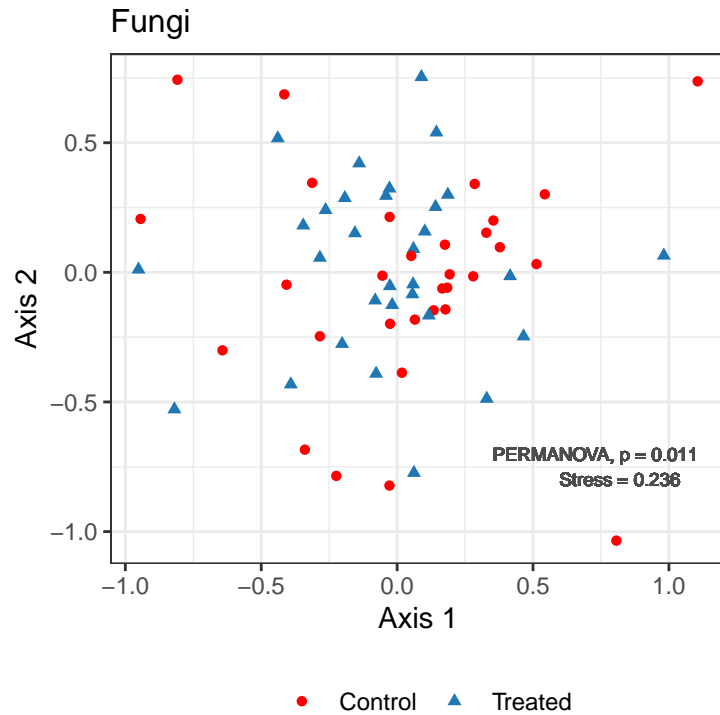
Combine plots for Fig 5

```r
# Combine NMDS ----------------------------------------------------------

tiff("Fig5_Soil-NMDS.tiff", height = 4, width = 7, units = "in", res = 1000)
ggarrange(barc.nmds.plot.21, fungi.nmds.plot.21,
          nrow = 1, ncol = 2,
          labels = c("(A)", "(B)"),
          common.legend = TRUE, legend = "bottom")
dev.off()
```

Supp Fig 5a: Total soil nitrogen

```r
# Total N ---------------------------------------------------------------

# Mann-Whitney
wilcox.test(filter(dat.2021, Treatment == "Control")$TN_ppt,
            filter(dat.2021, Treatment == "Treated")$TN_ppt,
            exact = FALSE) # p-value = 0.1731
```

```
##
##  Wilcoxon rank sum test with continuity correction
##
## data:  filter(dat.2021, Treatment == "Control")$TN_ppt and filter(dat.2021, Treatment == "Treated")$T
```

```
## W = 383.5, p-value = 0.1731
## alternative hypothesis: true location shift is not equal to 0
```

```r
# Plot
tn.plot.21 <- dat.2021 |>
  ggplot(aes(x = Treatment, y = TN_ppt)) +
  geom_boxplot(alpha = 0.3,
               outlier.shape = NA,
               aes(fill = Treatment)) +
  geom_jitter(size = 1,
              alpha = 0.8,
              aes(color = Treatment)) +
  scale_color_manual(values = c("red", "#1F78B4")) +
  scale_fill_manual(values = c("red", "#1F78B4")) +
  labs(title = "Total soil nitrogen",
       x = NULL,
       y = "Total N (mg/g)") +
  theme_bw() +
  theme(legend.position = "none") +
  theme(axis.text.x = element_text(color = "#000000"))  +
  theme(plot.margin = margin(0.1, 0.2, 0.1, 0.1, "in")) +
  geom_text(aes(x = 2, y = 63, label = "Mann-Whitney, \np = 0.173"),
            color = "gray30",
            size = 2.5) +
  stat_summary(fun = mean, geom = "errorbar", aes(ymax = after_stat(y), ymin = after_stat(y)),
               width = 0.75, linetype = "dashed") +
  theme(plot.title = element_text(size = 12))
tn.plot.21
```

Supp Fig 5b: Total soil carbon

```r
# Total C -----------------------------------------------------------------

# Mann-Whitney
wilcox.test(filter(dat.2021, Treatment == "Control")$TC_ppt,
            filter(dat.2021, Treatment == "Treated")$TC_ppt,
            exact = FALSE) # p-value = 0.2397
```
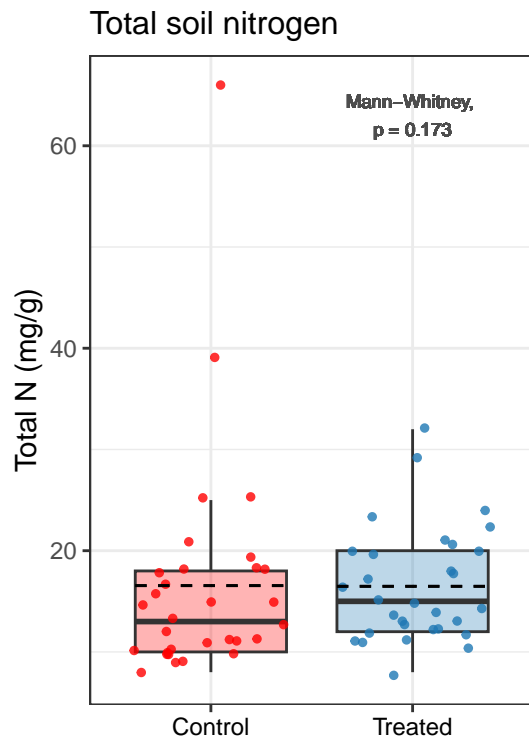
```
##
##  Wilcoxon rank sum test with continuity correction
##
## data:  filter(dat.2021, Treatment == "Control")$TC_ppt and filter(dat.2021, Treatment == "Treated")$`
## W = 396.5, p-value = 0.2397
## alternative hypothesis: true location shift is not equal to 0
```

```r
# Plot
tc.plot.21 <- dat.2021 |>
  ggplot(aes(x = Treatment, y = TC_ppt)) +
  geom_boxplot(alpha = 0.3,
               outlier.shape = NA,
               aes(fill = Treatment)) +
  geom_jitter(size = 1,
              alpha = 0.8,
              aes(color = Treatment)) +
  scale_color_manual(values = c("red", "#1F78B4")) +
  scale_fill_manual(values = c("red", "#1F78B4")) +
  labs(title = "Total soil carbon",
       x = NULL,
       y = "Total C (mg/g)") +
  theme_bw() +
  theme(legend.position = "none") +
  theme(axis.text.x = element_text(color = "#000000"))  +
  theme(plot.margin = margin(0.1, 0.2, 0.1, 0.1, "in")) +
  geom_text(aes(x = 2, y = 670, label = "Mann-Whitney, \np = 0.240"),
            color = "gray30",
            size = 2.5) +
  stat_summary(fun = mean, geom = "errorbar", aes(ymax = after_stat(y), ymin = after_stat(y)),
               width = 0.75, linetype = "dashed") +
  theme(plot.title = element_text(size = 12))
tc.plot.21
```
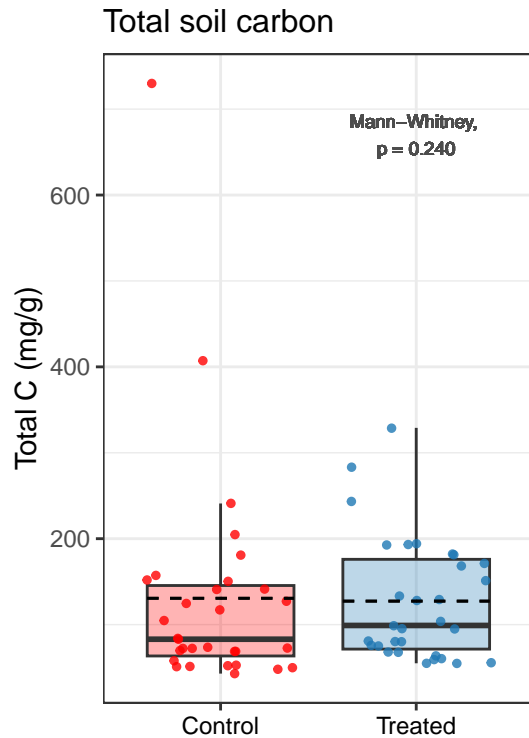
Total soil carbon

Supp Fig 5c: Soil organic matter

```
# Organic matter ------------------------------------------------------------

# Mann-Whitney
wilcox.test(filter(dat.2021, Treatment == "Control")$OM_perc,
            filter(dat.2021, Treatment == "Treated")$OM_perc) # p-value = 0.4332
```

```
##
##  Wilcoxon rank sum exact test
##
## data:  filter(dat.2021, Treatment == "Control")$OM_perc and filter(dat.2021, Treatment == "Treated")$
## W = 424, p-value = 0.4332
## alternative hypothesis: true location shift is not equal to 0
```

```
# Plot
om.plot.21 <- dat.2021 |>
  ggplot(aes(x = Treatment, y = OM_perc)) +
  geom_boxplot(alpha = 0.3,
               outlier.shape = NA,
               aes(fill = Treatment)) +
  geom_jitter(size = 1,
              alpha = 0.8,
              aes(color = Treatment)) +
  scale_color_manual(values = c("red", "#1F78B4")) +
  scale_fill_manual(values = c("red", "#1F78B4")) +
```

```
    labs(title = "Soil organic matter",
         x = NULL,
         y = "Organic matter (%)") +
    theme_bw() +
    theme(legend.position = "none") +
    theme(axis.text.x = element_text(color = "#000000"))  +
    theme(plot.margin = margin(0.1, 0.2, 0.1, 0.1, "in")) +
    geom_text(aes(x = 2, y = 2.14, label = "Mann-Whitney, \np = 0.433"),
              color = "gray30",
              size = 2.5) +
    stat_summary(fun = mean, geom = "errorbar", aes(ymax = after_stat(y), ymin = after_stat(y)),
                 width = 0.75, linetype = "dashed") +
    theme(plot.title = element_text(size = 12))
om.plot.21
```


Soil organic matter
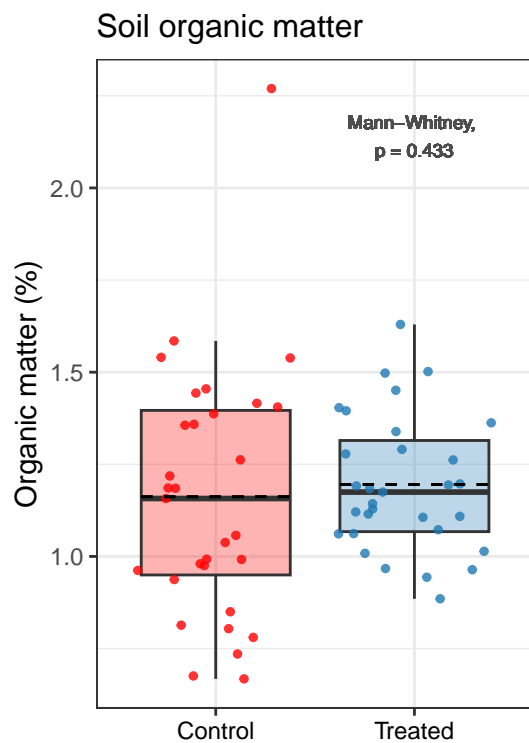
Combine plots for Supp Fig 5

```
# Combine soil chem ---------------------------------------------------------

# TN, TC, OM
tiff("FigS5_Soil-chem.tiff", units = "in", height = 4, width = 7, res = 300)
ggarrange(tn.plot.21, tc.plot.21, om.plot.21,
          ncol = 3, nrow = 1,
          labels = c("(A)", "(B)", "(C)"))


dev.off()
```

Supp Fig 6a: Bacterial & archaeal richness

```
# Barc richness --------------------------------------------------------

# T-test
t.test(filter(dat.2021, Treatment == "Control")$barc.richness,
       filter(dat.2021, Treatment == "Treated")$barc.richness) # NS, p = 0.686
```

```
##
##  Welch Two Sample t-test
##
## data:  filter(dat.2021, Treatment == "Control")$barc.richness and filter(dat.2021, Treatment == "Trea
## t = 0.40607, df = 59.999, p-value = 0.6861
## alternative hypothesis: true difference in means is not equal to 0
## 95 percent confidence interval:
##  -116.6407  176.0601
## sample estimates:
## mean of x mean of y
##  1702.065  1672.355
```

```
# Plot
barc.rich.plot.21 <- dat.2021 |>
  ggplot(aes(Treatment, barc.richness)) +
  geom_jitter(aes(color = Treatment),
              alpha = 0.8,
              size = 1) +
  geom_boxplot(aes(fill = Treatment),
               alpha = 0.3,
               outlier.shape = NA) +
  xlab(NULL) +
  ylab("No. of ASVs") +
  ggtitle("Bacterial & archaeal richness") +
  scale_color_manual(values = c("red", "#1F78B4")) +
  scale_fill_manual(values = c("red", "#1F78B4")) +
  theme_bw() +
  theme(legend.position = "none") +
  theme(axis.text.x = element_text(color = "#000000")) +
  geom_text(aes(x = 2.1, y = 1100, label = "t-test, p = 0.686"),
            color = "gray30",
            size = 2.5) +
  theme(plot.margin = margin(0.1, 0.1, 0.1, 0.1, "in")) +
  stat_summary(fun = mean, geom = "errorbar", aes(ymax = after_stat(y), ymin = after_stat(y)),
               width = 0.75, linetype = "dashed") +
  theme(plot.title = element_text(size = 11.5))
barc.rich.plot.21
```

Bacterial & archaeal richness

t–test, p = 0.686
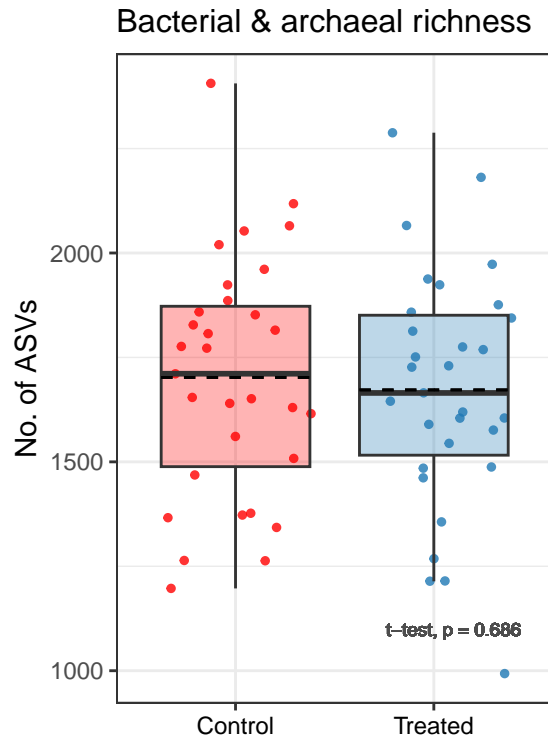
Supp Fig 6b: Fungal richness

```r
# Fungi richness -------------------------------------------------------

# T-test
t.test(filter(dat.2021, Treatment == "Control")$fungi.richness,
       filter(dat.2021, Treatment == "Treated")$fungi.richness) # NS, p = 0.938
```

```
##
##  Welch Two Sample t-test
##
## data:  filter(dat.2021, Treatment == "Control")$fungi.richness and filter(dat.2021, Treatment == "Tre
## t = 0.078, df = 59.653, p-value = 0.9381
## alternative hypothesis: true difference in means is not equal to 0
## 95 percent confidence interval:
##  -61.22215  66.18989
## sample estimates:
## mean of x mean of y
##  406.9677  404.4839
```

```r
# Plot
fungi.rich.plot.21 <- dat.2021 %>%
  ggplot(aes(Treatment, fungi.richness)) +
  geom_jitter(aes(color = Treatment),
              alpha = 0.8,
              size = 1) +
```

```
    geom_boxplot(aes(fill = Treatment),
                 alpha = 0.3,
                 outlier.shape = NA) +
    xlab(NULL) +
    ylab(NULL) +
    ggtitle("Fungal richness") +
    scale_color_manual(values = c("red", "#1F78B4")) +
    scale_fill_manual(values = c("red", "#1F78B4")) +
    theme_bw() +
    theme(legend.position = "none") +
    theme(axis.text.x = element_text(color = "#000000")) +
    geom_text(aes(x = 2.1, y = 70, label = "t-test, p = 0.938"),
              color = "gray30",
              size = 2.5) +
    theme(plot.margin = margin(0.1, 0.1, 0.1, 0.1, "in")) +
    stat_summary(fun = mean, geom = "errorbar", aes(ymax = after_stat(y), ymin = after_stat(y)),
                 width = 0.75, linetype = "dashed") +
    theme(plot.title = element_text(size = 12))
fungi.rich.plot.21
```
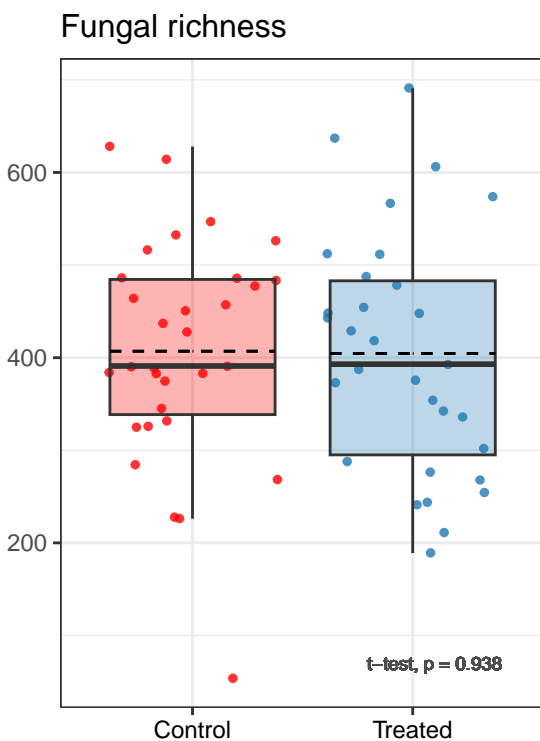

Fungal richness

Combine plots for Supp Fig 6

```
# Combine soil richness ----------------------------------------------

tiff("FigS6_Soil-richness.tiff", units = "in", height = 4, width = 5.5, res = 300)
ggarrange(barc.rich.plot.21, fungi.rich.plot.21,
```

```
        ncol = 2, nrow = 1,
        labels = c("(A)", "(B)"))

dev.off()
```

Supp Fig 7a: Chemoheterotrophic bacteria & archaea

```
# Chemoheterotrophs -----------------------------------------------------

# Mann-Whitney
wilcox.test(filter(dat.2021, Treatment == "Control")$chemoheterotrophy_perc,
            filter(dat.2021, Treatment == "Treated")$chemoheterotrophy_perc) # p = 0.8449
```

```
##
##  Wilcoxon rank sum exact test
##
## data:  filter(dat.2021, Treatment == "Control")$chemoheterotrophy_perc and filter(dat.2021, Treatment
## W = 466, p-value = 0.8449
## alternative hypothesis: true location shift is not equal to 0
```

```
# Plot
chemohet.plot.21 <- dat.2021 %>%
  ggplot(aes(Treatment, chemoheterotrophy_perc)) +
  geom_jitter(aes(color = Treatment),
              alpha = 0.8,
              size = 1) +
  geom_boxplot(aes(fill = Treatment),
               alpha = 0.3,
               outlier.shape = NA) +
  xlab(NULL) +
  ylab("Relative abundance (%)") +
  ggtitle("Chemoheterotrophic \nbacteria & archaea") +
  scale_color_manual(values = c("red", "#1F78B4")) +
  scale_fill_manual(values = c("red", "#1F78B4")) +
  theme_bw() +
  theme(legend.position = "none") +
  theme(axis.text.x = element_text(color = "#000000")) +
  theme(plot.margin = margin(0.1, 0.1, 0.1, 0.1, "in")) +
  geom_text(aes(x = 2, y = 28.7, label = "Mann-Whitney, \np = 0.845"),
            color = "gray30",
            size = 2.5) +
  stat_summary(fun = mean, geom = "errorbar", aes(ymax = after_stat(y), ymin = after_stat(y)),
               width = 0.75, linetype = "dashed") +
  theme(plot.title = element_text(size = 12))
chemohet.plot.21
```

Chemoheterotrophic bacteria & archaea


```
# N-cyclers --------------------------------------------------------------

# Mann-Whitney
wilcox.test(filter(dat.2021, Treatment == "Control")$n.cycler_perc,
            filter(dat.2021, Treatment == "Treated")$n.cycler_perc) # p-value = 0.5854
```
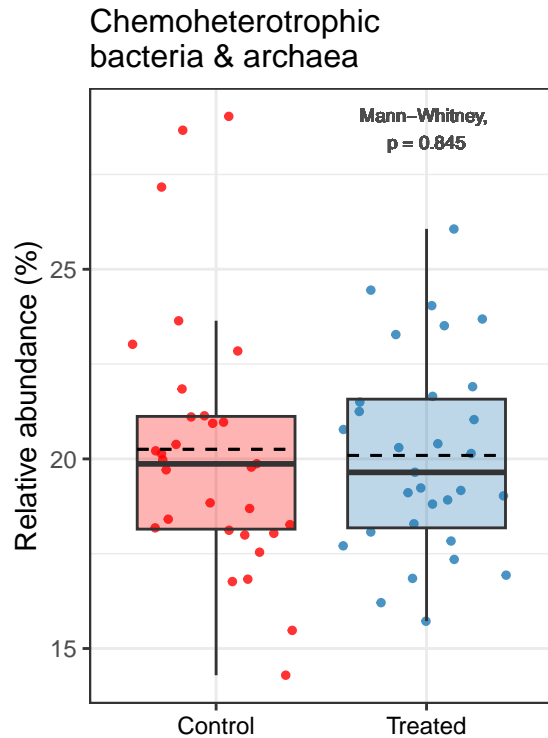
```
##
##  Wilcoxon rank sum exact test
##
## data:  filter(dat.2021, Treatment == "Control")$n.cycler_perc and filter(dat.2021, Treatment == "Trea
## W = 520, p-value = 0.5854
## alternative hypothesis: true location shift is not equal to 0
```

```
# Plot
ncycler.plot.21 <- dat.2021 %>%
  ggplot(aes(Treatment, n.cycler_perc)) +
  geom_jitter(aes(color = Treatment),
              alpha = 0.8,
              size = 1) +
  geom_boxplot(aes(fill = Treatment),
               alpha = 0.3,
               outlier.shape = NA) +
  xlab(NULL) +
  ylab(NULL) +
```

Supp Fig 7b: Nitrogen-cycling bacteria & archaea

```
  ggtitle("Nitrogen-cycling \nbacteria & archaea") +
  scale_color_manual(values = c("red", "#1F78B4")) +
  scale_fill_manual(values = c("red", "#1F78B4")) +
  theme_bw() +
  theme(legend.position = "none") +
  theme(axis.text.x = element_text(color = "#000000")) +
  theme(plot.margin = margin(0.1, 0.1, 0.1, 0.25, "in")) +
  geom_text(aes(x = 2, y = 13.6, label = "Mann-Whitney, \np = 0.585"),
            color = "gray30",
            size = 2.5) +
  stat_summary(fun = mean, geom = "errorbar", aes(ymax = after_stat(y), ymin = after_stat(y)),
               width = 0.75, linetype = "dashed") +
  theme(plot.title = element_text(size = 12))
ncycler.plot.21
```


Nitrogen−cycling
bacteria & archaea
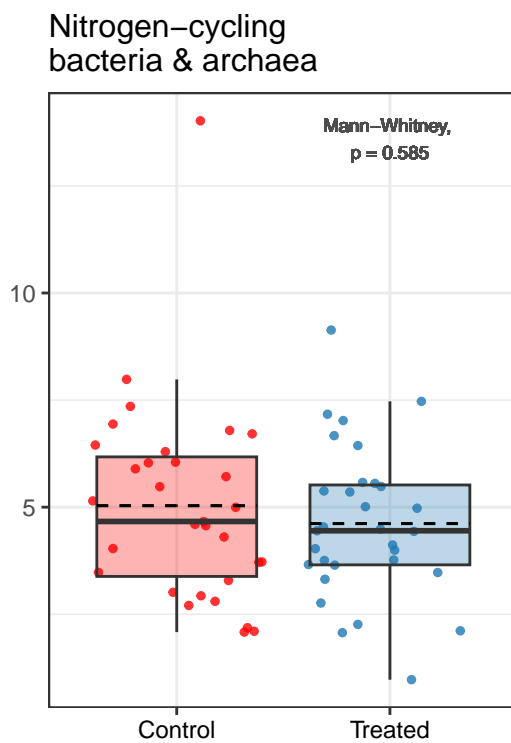
Supp Fig 7c: Saprotrophic fungi

```
# Saprotrophs ----------------------------------------------------------------

# T-test
t.test(filter(dat.2021, Treatment == "Control")$saprotroph,
       filter(dat.2021, Treatment == "Treated")$saprotroph) # NS, p = 0.272
```

```
##
##  Welch Two Sample t-test
##
```

```
## data:  filter(dat.2021, Treatment == "Control")$saprotroph and filter(dat.2021, Treatment == "Treate
## t = -1.1097, df = 59.068, p-value = 0.2716
## alternative hypothesis: true difference in means is not equal to 0
## 95 percent confidence interval:
##  -6.168384  1.767276
## sample estimates:
## mean of x mean of y
##  19.96490  22.16546
```

```
# Plot
sapro.plot.21 <- dat.2021 %>%
  ggplot(aes(Treatment, saprotroph)) +
  geom_jitter(aes(color = Treatment),
              alpha = 0.8,
              size = 1) +
  geom_boxplot(aes(fill = Treatment),
               alpha = 0.3,
               outlier.shape = NA) +
  xlab(NULL) +
  ylab(NULL) +
  ggtitle("Saprotrophic fungi") +
  scale_color_manual(values = c("red", "#1F78B4")) +
  scale_fill_manual(values = c("red", "#1F78B4")) +
  theme_bw() +
  theme(legend.position = "none") +
  theme(axis.text.x = element_text(color = "#000000")) +
  theme(plot.margin = margin(0.1, 0.1, 0.1, 0.25, "in")) +
  geom_text(aes(x = 2, y = 42, label = "t-test, p = 0.272"),
            color = "gray30",
            size = 2.5) +
  stat_summary(fun = mean, geom = "errorbar", aes(ymax = after_stat(y), ymin = after_stat(y)),
               width = 0.75, linetype = "dashed") +
  theme(plot.title = element_text(size = 12))
sapro.plot.21
```

Saprotrophic fungi
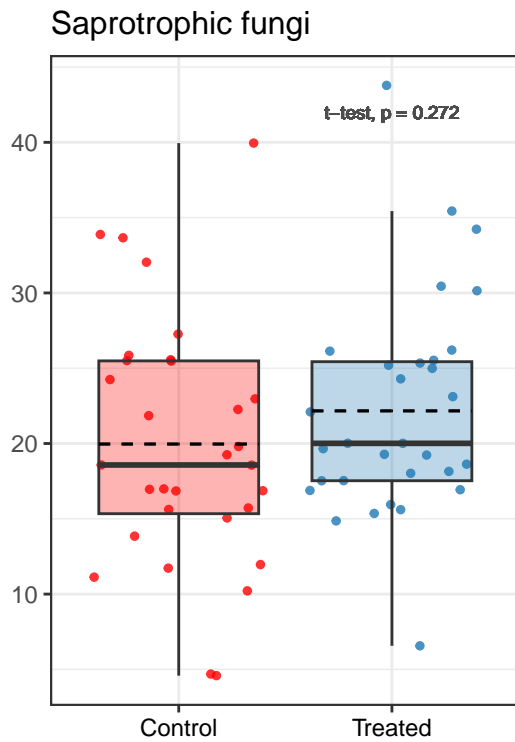
Combine plots for Supp Fig 7

```
# Combine FAPROTAX and FUNGuild ------------------------------------------

tiff("FigS7_Soil-functional.tiff", units = "in", height = 4, width = 7, res = 300)
ggarrange(chemohet.plot.21, ncycler.plot.21, sapro.plot.21,
          ncol = 3, nrow = 1,
          labels = c("(A)", "(B)", "(C)"))

dev.off()
```

Appendix D: Structural equation modeling

Setup

```
library(lavaan)
library(tidyverse)
```

```
# Load data -------------------------------------------------------
```

```
dat.2021 <- read.csv("Veg-soil-elev_2021.csv")
```

```
# Data wrangling --------------------------------------------------
```

```
# Add Control/Treated as binary variable and select only variables needed for SEM
sem.dat.unscaled <- dat.2021 |>
  mutate(rocks = case_when(
    Treatment == "Control" ~ 0,
    Treatment == "Treated" ~ 1)) |>
  select(Sample, rocks, notree, notree.18, herb, herb.18, tree, perveg.richness, perveg.shannon,
         TN_log, TC_log, OM_log, barc.richness, fungi.richness,
         chemoheterotrophy_log, n.cycler_log, saprotroph)

# Center and scale continuous variables
sem.dat <- sem.dat.unscaled |>
  mutate(rocks = as.character(rocks),
         Sample = as.character(Sample)) |>
  mutate_if(is.numeric, scale) |>
  mutate(rocks = as.numeric(rocks))
```

Latent variables

Soil microbiome

```
lvmod.soimic <- '
  # latent variable model
  soil_microbe =~ barc.richness + fungi.richness + chemoheterotrophy_log + n.cycler_log + saprotroph
'
fit.soimic <- sem(lvmod.soimic, data = sem.dat)
summary(fit.soimic, fit.measures = TRUE, standardized = TRUE)
```

```
## lavaan 0.6.16 ended normally after 18 iterations
##
##   Estimator                                         ML
##   Optimization method                           NLMINB
##   Number of model parameters                        10
##
##   Number of observations                            62
##
## Model Test User Model:
```

```
##
##   Test statistic                                  2.723
##   Degrees of freedom                                  5
##   P-value (Chi-square)                            0.743
##
## Model Test Baseline Model:
##
##   Test statistic                                 52.070
##   Degrees of freedom                                 10
##   P-value                                         0.000
##
## User Model versus Baseline Model:
##
##   Comparative Fit Index (CFI)                     1.000
##   Tucker-Lewis Index (TLI)                        1.108
##
## Loglikelihood and Information Criteria:
##
##   Loglikelihood user model (H0)                -412.677
##   Loglikelihood unrestricted model (H1)        -411.315
##
##   Akaike (AIC)                                  845.354
##   Bayesian (BIC)                                866.625
##   Sample-size adjusted Bayesian (SABIC)         835.162
##
## Root Mean Square Error of Approximation:
##
##   RMSEA                                           0.000
##   90 Percent confidence interval - lower          0.000
##   90 Percent confidence interval - upper          0.125
##   P-value H_0: RMSEA <= 0.050                     0.800
##   P-value H_0: RMSEA >= 0.080                     0.134
##
## Standardized Root Mean Square Residual:
##
##   SRMR                                            0.044
##
## Parameter Estimates:
##
##   Standard errors                              Standard
##   Information                                  Expected
##   Information saturated (h1) model           Structured
##
## Latent Variables:
##                  Estimate  Std.Err  z-value  P(>|z|)   Std.lv  Std.all
##   soil_microbe =~
##     barc.richness   1.000                                0.853    0.860
##     fungi.richness  0.342    0.168    2.033    0.042     0.291    0.294
##     chmhtrtrphy_lg  0.158    0.164    0.962    0.336     0.135    0.136
##     n.cycler_log    0.892    0.241    3.699    0.000     0.761    0.767
##     saprotroph      0.461    0.172    2.675    0.007     0.393    0.397
##
## Variances:
##                  Estimate  Std.Err  z-value  P(>|z|)   Std.lv  Std.all
```

```
##     .barc.richness      0.256    0.184    1.392    0.164    0.256    0.260
##     .fungi.richness     0.899    0.165    5.448    0.000    0.899    0.914
##     .chmhtrtrphy_lg     0.966    0.174    5.545    0.000    0.966    0.982
##     .n.cycler_log       0.405    0.160    2.538    0.011    0.405    0.412
##     .saprotroph         0.829    0.156    5.321    0.000    0.829    0.843
##      soil_microbe       0.728    0.247    2.951    0.003    1.000    1.000
```

```r
modindices(fit.soimic, sort = TRUE, minimum.value = 3.5)
```

```
## [1] lhs        op       rhs       mi       epc      sepc.lv  sepc.all sepc.nox
## <0 rows> (or 0-length row.names)
```

```r
# No paths to add.
```

Soil chemistry

- Soil chemistry does not do well as a latent variable because TN, TC, and OM are collinear and must
  be modeled separately.

```r
lvmod.soichem <- '
  # latent variable model
  soil_chem =~ TN_log + OM_log
'
fit.soichem <- sem(lvmod.soichem, data = sem.dat)
```

```
## Warning in lav_model_vcov(lavmodel = lavmodel, lavsamplestats = lavsamplestats, : lavaan WARNING:
##     Could not compute standard errors! The information matrix could
##     not be inverted. This may be a symptom that the model is not
##     identified.
```

```r
summary(fit.soichem)
```

```
## lavaan 0.6.16 ended normally after 13 iterations
##
##   Estimator                                         ML
##   Optimization method                           NLMINB
##   Number of model parameters                         4
##
##   Number of observations                            62
##
## Model Test User Model:
##
##   Test statistic                                    NA
##   Degrees of freedom                                -1
##   P-value (Unknown)                                 NA
##
## Parameter Estimates:
##
##   Standard errors                             Standard
##   Information                                 Expected
##   Information saturated (h1) model          Structured
```

44

```
##
## Latent Variables:
##                   Estimate  Std.Err  z-value  P(>|z|)
##   soil_chem =~
##     TN_log           1.000
##     OM_log           0.874        NA
##
## Variances:
##                   Estimate  Std.Err  z-value  P(>|z|)
##    .TN_log           0.139        NA
##    .OM_log           0.338        NA
##     soil_chem        0.845        NA
```

Model 1

Model for Supp Fig 8.

```r
mod1 <- '
  # latent variables
  soil_microbe =~ barc.richness + fungi.richness + chemoheterotrophy_log + n.cycler_log + saprotroph

  # structure
  notree ~ rocks + notree.18 + tree
  OM_log ~ rocks
  soil_microbe ~ rocks
  notree.18 ~ rocks

  # covariance
  OM_log ~~ soil_microbe
  OM_log ~~ notree
  soil_microbe ~~ notree
'
fit1 <- sem(mod1, data = sem.dat)
summary(fit1, fit.measures = TRUE, standardized = TRUE)
```

```
## lavaan 0.6.16 ended normally after 27 iterations
##
##   Estimator                                         ML
##   Optimization method                           NLMINB
##   Number of model parameters                        22
##
##   Number of observations                            62
##
## Model Test User Model:
##
##   Test statistic                                26.242
##   Degrees of freedom                                30
##   P-value (Chi-square)                           0.663
##
## Model Test Baseline Model:
##
##   Test statistic                               124.316
##   Degrees of freedom                                44
```

```
##    P-value                                        0.000
##
## User Model versus Baseline Model:
##
##    Comparative Fit Index (CFI)                    1.000
##    Tucker-Lewis Index (TLI)                       1.069
##
## Loglikelihood and Information Criteria:
##
##    Loglikelihood user model (H0)               -650.724
##    Loglikelihood unrestricted model (H1)       -637.603
##
##    Akaike (AIC)                                1345.448
##    Bayesian (BIC)                              1392.245
##    Sample-size adjusted Bayesian (SABIC)       1323.026
##
## Root Mean Square Error of Approximation:
##
##    RMSEA                                          0.000
##    90 Percent confidence interval - lower         0.000
##    90 Percent confidence interval - upper         0.080
##    P-value H_0: RMSEA <= 0.050                    0.829
##    P-value H_0: RMSEA >= 0.080                    0.049
##
## Standardized Root Mean Square Residual:
##
##    SRMR                                           0.076
##
## Parameter Estimates:
##
##    Standard errors                             Standard
##    Information                                 Expected
##    Information saturated (h1) model          Structured
##
## Latent Variables:
##                   Estimate  Std.Err  z-value  P(>|z|)   Std.lv  Std.all
##    soil_microbe =~
##      barc.richness   1.000                                0.845    0.852
##      fungi.richness  0.361    0.165    2.194    0.028     0.305    0.308
##      chmhtrtrphy_lg  0.192    0.164    1.167    0.243     0.162    0.163
##      n.cycler_log    0.908    0.194    4.675    0.000     0.768    0.774
##      saprotroph      0.442    0.165    2.680    0.007     0.374    0.377
##
## Regressions:
##                   Estimate  Std.Err  z-value  P(>|z|)   Std.lv  Std.all
##    notree ~
##      rocks          -0.615    0.239   -2.567    0.010    -0.615   -0.307
##      notree.18       0.535    0.107    5.011    0.000     0.535    0.530
##      tree           -0.037    0.106   -0.347    0.728    -0.037   -0.036
##    OM_log ~
##      rocks           0.249    0.250    0.996    0.319     0.249    0.126
##    soil_microbe ~
##      rocks          -0.106    0.237   -0.446    0.656    -0.125   -0.063
##    notree.18 ~
```

```
##     rocks              0.674   0.237   2.844   0.004   0.674   0.340
##
## Covariances:
##                   Estimate  Std.Err  z-value  P(>|z|)   Std.lv  Std.all
##   .soil_microbe ~~
##     .OM_log          0.370   0.129   2.861   0.004   0.439   0.446
##   .notree ~~
##     .OM_log          0.332   0.115   2.877   0.004   0.332   0.393
##   .soil_microbe ~~
##     .notree          0.213   0.107   1.991   0.046   0.252   0.294
##
## Variances:
##                   Estimate  Std.Err  z-value  P(>|z|)   Std.lv  Std.all
##     .barc.richness   0.269   0.138   1.955   0.051   0.269   0.274
##     .fungi.richness  0.891   0.164   5.442   0.000   0.891   0.905
##     .chmhtrtrphy_lg  0.958   0.173   5.535   0.000   0.958   0.973
##     .n.cycler_log    0.395   0.129   3.067   0.002   0.395   0.401
##     .saprotroph      0.844   0.157   5.367   0.000   0.844   0.858
##     .notree          0.740   0.133   5.568   0.000   0.740   0.737
##     .OM_log          0.968   0.174   5.568   0.000   0.968   0.984
##     .notree.18       0.870   0.156   5.568   0.000   0.870   0.885
##     .soil_microbe    0.712   0.213   3.349   0.001   0.996   0.996
```

```r
modindices(fit1, sort = TRUE, minimum.value = 3.5)
```

```
##                        lhs op     rhs    mi     epc sepc.lv sepc.all sepc.nox
## 50           saprotroph ~~  OM_log 4.331 -0.213  -0.213   -0.235   -0.235
## 43 chemoheterotrophy_log ~~  OM_log 3.616  0.203   0.203    0.211    0.211
```

```r
# OM already covaries with soil microbiome latent variable;
#   does not make sense to add paths for saptrotrophs or chemoheterotrophs.
```

Model 2

Model for Supp Fig 9.

```r
mod2 <- '
  # latent variables
  soil_microbe =~ barc.richness + fungi.richness + chemoheterotrophy_log + n.cycler_log + saprotroph

  # structure
  notree ~ rocks + notree.18 + tree
  TN_log ~ rocks
  soil_microbe ~ rocks
  notree.18 ~ rocks

  # covariance
  TN_log ~~ soil_microbe
  TN_log ~~ notree
  soil_microbe ~~ notree
'
fit2 <- sem(mod2, data = sem.dat)
summary(fit2, fit.measures = TRUE, standardized = TRUE)
```

```
## lavaan 0.6.16 ended normally after 32 iterations
##
##   Estimator                                         ML
##   Optimization method                           NLMINB
##   Number of model parameters                        22
##
##   Number of observations                            62
##
## Model Test User Model:
##
##   Test statistic                                31.847
##   Degrees of freedom                                30
##   P-value (Chi-square)                           0.375
##
## Model Test Baseline Model:
##
##   Test statistic                               164.607
##   Degrees of freedom                                44
##   P-value                                        0.000
##
## User Model versus Baseline Model:
##
##   Comparative Fit Index (CFI)                    0.985
##   Tucker-Lewis Index (TLI)                       0.978
##
## Loglikelihood and Information Criteria:
##
##   Loglikelihood user model (H0)               -633.381
##   Loglikelihood unrestricted model (H1)       -617.457
##
##   Akaike (AIC)                                1310.762
##   Bayesian (BIC)                              1357.559
##   Sample-size adjusted Bayesian (SABIC)       1288.340
##
## Root Mean Square Error of Approximation:
##
##   RMSEA                                          0.032
##   90 Percent confidence interval - lower         0.000
##   90 Percent confidence interval - upper         0.103
##   P-value H_0: RMSEA <= 0.050                    0.595
##   P-value H_0: RMSEA >= 0.080                    0.165
##
## Standardized Root Mean Square Residual:
##
##   SRMR                                           0.079
##
## Parameter Estimates:
##
##   Standard errors                             Standard
##   Information                                 Expected
##   Information saturated (h1) model          Structured
##
## Latent Variables:
##                    Estimate  Std.Err  z-value  P(>|z|)   Std.lv  Std.all
```

```
##   soil_microbe =~
##     barc.richness      1.000                                0.808    0.815
##     fungi.richness     0.455    0.167    2.719    0.007      0.367    0.370
##     chmhtrtrphy_lg     0.284    0.169    1.679    0.093      0.229    0.231
##     n.cycler_log       0.948    0.163    5.802    0.000      0.766    0.772
##     saprotroph         0.464    0.167    2.777    0.005      0.375    0.378
##
## Regressions:
##                    Estimate  Std.Err  z-value  P(>|z|)   Std.lv   Std.all
##   notree ~
##     rocks            -0.651    0.237   -2.744    0.006    -0.651   -0.315
##     notree.18         0.593    0.099    6.011    0.000     0.593    0.570
##     tree             -0.033    0.098   -0.336    0.737    -0.033   -0.032
##   TN_log ~
##     rocks             0.177    0.251    0.706    0.480     0.177    0.089
##   soil_microbe ~
##     rocks            -0.102    0.230   -0.444    0.657    -0.126   -0.063
##   notree.18 ~
##     rocks             0.674    0.237    2.844    0.004     0.674    0.340
##
## Covariances:
##                    Estimate  Std.Err  z-value  P(>|z|)   Std.lv   Std.all
##  .soil_microbe ~~
##     .TN_log           0.602    0.145    4.157    0.000     0.747    0.756
##  .notree ~~
##     .TN_log           0.454    0.123    3.696    0.000     0.454    0.532
##  .soil_microbe ~~
##     .notree           0.214    0.104    2.054    0.040     0.266    0.308
##
## Variances:
##                    Estimate  Std.Err  z-value  P(>|z|)   Std.lv   Std.all
##     .barc.richness    0.331    0.100    3.310    0.001     0.331    0.337
##     .fungi.richness   0.849    0.157    5.398    0.000     0.849    0.863
##     .chmhtrtrphy_lg   0.931    0.169    5.508    0.000     0.931    0.947
##     .n.cycler_log     0.397    0.103    3.867    0.000     0.397    0.404
##     .saprotroph       0.843    0.156    5.389    0.000     0.843    0.857
##     .notree           0.746    0.134    5.568    0.000     0.746    0.699
##     .TN_log           0.976    0.175    5.568    0.000     0.976    0.992
##     .notree.18        0.870    0.156    5.568    0.000     0.870    0.885
##     .soil_microbe     0.650    0.184    3.529    0.000     0.996    0.996
```

```
modindices(fit2, sort = TRUE, minimum.value = 3.5)
```

```
##                      lhs op          rhs    mi   epc sepc.lv sepc.all sepc.nox
## 43 chemoheterotrophy_log ~~      TN_log 6.485 0.207   0.207    0.217    0.217
## 29          barc.richness ~~ n.cycler_log 4.776 0.303   0.303    0.835    0.835
## 38         fungi.richness ~~      TN_log 4.552 0.170   0.170    0.187    0.187
## 39         fungi.richness ~~   notree.18 4.096 0.224   0.224    0.260    0.260
```

```
# TN already covaries with soil microbiome latent variable;
#   does not make sense to add path.
# Prior veg (notree.18) is too far removed to have a plausible                    co
```

Model 3

Model for Supp Fig 10.

```r
mod3 <- '
  # latent variables
  soil_microbe =~ barc.richness + fungi.richness + chemoheterotrophy_log +
  n.cycler_log + saprotroph

  # structure
  herb ~ rocks + herb.18 + tree
  OM_log ~ rocks
  soil_microbe ~ rocks
  herb.18 ~ rocks

  # covariance
  OM_log ~~ soil_microbe
  OM_log ~~ herb
  soil_microbe ~~ herb
'
fit3 <- sem(mod3, data = sem.dat)
summary(fit3, fit.measures = TRUE, standardized = TRUE)
```

```
## lavaan 0.6.16 ended normally after 27 iterations
##
##   Estimator                                         ML
##   Optimization method                           NLMINB
##   Number of model parameters                        22
##
##   Number of observations                            62
##
## Model Test User Model:
##
##   Test statistic                                21.703
##   Degrees of freedom                                30
##   P-value (Chi-square)                           0.865
##
## Model Test Baseline Model:
##
##   Test statistic                               113.947
##   Degrees of freedom                                44
##   P-value                                        0.000
##
## User Model versus Baseline Model:
##
##   Comparative Fit Index (CFI)                    1.000
##   Tucker-Lewis Index (TLI)                       1.174
##
## Loglikelihood and Information Criteria:
##
##   Loglikelihood user model (H0)               -653.639
##   Loglikelihood unrestricted model (H1)       -642.787
##
##   Akaike (AIC)                                1351.278
```

```
##    Bayesian (BIC)                                  1398.075
##    Sample-size adjusted Bayesian (SABIC)           1328.856
##
## Root Mean Square Error of Approximation:
##
##    RMSEA                                              0.000
##    90 Percent confidence interval - lower             0.000
##    90 Percent confidence interval - upper             0.052
##    P-value H_0: RMSEA <= 0.050                        0.946
##    P-value H_0: RMSEA >= 0.080                        0.011
##
## Standardized Root Mean Square Residual:
##
##    SRMR                                               0.069
##
## Parameter Estimates:
##
##    Standard errors                                Standard
##    Information                                    Expected
##    Information saturated (h1) model             Structured
##
## Latent Variables:
##                   Estimate  Std.Err  z-value  P(>|z|)   Std.lv  Std.all
##    soil_microbe =~
##      barc.richness    1.000                                      0.850    0.857
##      fungi.richness   0.351    0.163    2.151    0.031    0.298    0.301
##      chmhtrtrphy_lg   0.194    0.163    1.194    0.233    0.165    0.167
##      n.cycler_log     0.899    0.189    4.762    0.000    0.764    0.770
##      saprotroph       0.442    0.163    2.703    0.007    0.375    0.378
##
## Regressions:
##                   Estimate  Std.Err  z-value  P(>|z|)   Std.lv  Std.all
##    herb ~
##      rocks           -0.485    0.234   -2.070    0.038   -0.485   -0.250
##      herb.18          0.391    0.102    3.839    0.000    0.391    0.400
##      tree            -0.102    0.106   -0.965    0.335   -0.102   -0.104
##    OM_log ~
##      rocks            0.249    0.250    0.996    0.319    0.249    0.126
##    soil_microbe ~
##      rocks           -0.106    0.238   -0.443    0.658   -0.124   -0.062
##    herb.18 ~
##      rocks            0.334    0.248    1.347    0.178    0.334    0.169
##
## Covariances:
##                   Estimate  Std.Err  z-value  P(>|z|)   Std.lv  Std.all
##   .soil_microbe ~~
##     .OM_log          0.371    0.130    2.867    0.004    0.438    0.445
##   .herb ~~
##     .OM_log          0.349    0.118    2.954    0.003    0.349    0.405
##   .soil_microbe ~~
##     .herb            0.268    0.112    2.404    0.016    0.316    0.361
##
## Variances:
##                   Estimate  Std.Err  z-value  P(>|z|)   Std.lv  Std.all
```

```
##     .barc.richness      0.261    0.135    1.936    0.053    0.261    0.266
##     .fungi.richness     0.895    0.164    5.451    0.000    0.895    0.910
##     .chmhtrtrphy_lg     0.957    0.173    5.535    0.000    0.957    0.972
##     .n.cycler_log       0.400    0.126    3.172    0.002    0.400    0.407
##     .saprotroph         0.843    0.157    5.369    0.000    0.843    0.857
##     .herb               0.766    0.138    5.568    0.000    0.766    0.813
##     .OM_log             0.968    0.174    5.568    0.000    0.968    0.984
##     .herb.18            0.956    0.172    5.568    0.000    0.956    0.972
##     .soil_microbe       0.720    0.212    3.401    0.001    0.996    0.996
```

```r
modindices(fit3, sort = TRUE, minimum.value = 3.5)
```

```
##            lhs op    rhs    mi   epc sepc.lv sepc.all sepc.nox
## 50 saprotroph ~~ OM_log 4.195 -0.21   -0.21   -0.232   -0.232
```

```r
# OM already covaries with soil microbiome latent variable;
#   does not make sense to add path for saprotrophs.
```

Model 4

Model for Fig 6.

```r
mod4 <- '
  # latent variables
  soil_microbe =~ barc.richness + fungi.richness + chemoheterotrophy_log +
  n.cycler_log + saprotroph

  # structure
  herb ~ rocks + herb.18 + tree
  TN_log ~ rocks
  soil_microbe ~ rocks
  herb.18 ~ rocks

  # covariance
  TN_log ~~ soil_microbe
  TN_log ~~ herb
  soil_microbe ~~ herb
'
fit4 <- sem(mod4, data = sem.dat)
summary(fit4, fit.measures = TRUE, standardized = TRUE)
```

```
## lavaan 0.6.16 ended normally after 30 iterations
##
##   Estimator                                         ML
##   Optimization method                           NLMINB
##   Number of model parameters                        22
##
##   Number of observations                            62
##
## Model Test User Model:
##
##   Test statistic                                25.702
```

```
##    Degrees of freedom                               30
##    P-value (Chi-square)                          0.690
##
## Model Test Baseline Model:
##
##    Test statistic                            148.339
##    Degrees of freedom                             44
##    P-value                                     0.000
##
## User Model versus Baseline Model:
##
##    Comparative Fit Index (CFI)                 1.000
##    Tucker-Lewis Index (TLI)                    1.060
##
## Loglikelihood and Information Criteria:
##
##    Loglikelihood user model (H0)            -638.442
##    Loglikelihood unrestricted model (H1)    -625.591
##
##    Akaike (AIC)                             1320.884
##    Bayesian (BIC)                           1367.681
##    Sample-size adjusted Bayesian (SABIC)    1298.463
##
## Root Mean Square Error of Approximation:
##
##    RMSEA                                       0.000
##    90 Percent confidence interval - lower      0.000
##    90 Percent confidence interval - upper      0.077
##    P-value H_0: RMSEA <= 0.050                 0.847
##    P-value H_0: RMSEA >= 0.080                 0.042
##
## Standardized Root Mean Square Residual:
##
##    SRMR                                        0.066
##
## Parameter Estimates:
##
##    Standard errors                          Standard
##    Information                              Expected
##    Information saturated (h1) model       Structured
##
## Latent Variables:
##                   Estimate  Std.Err  z-value  P(>|z|)   Std.lv  Std.all
##    soil_microbe =~
##      barc.richness    1.000                                    0.812    0.818
##      fungi.richness   0.451    0.167    2.711    0.007    0.366    0.369
##      chmhtrtrphy_lg   0.273    0.168    1.623    0.105    0.222    0.224
##      n.cycler_log     0.940    0.163    5.764    0.000    0.763    0.770
##      saprotroph       0.467    0.166    2.810    0.005    0.379    0.383
##
## Regressions:
##                   Estimate  Std.Err  z-value  P(>|z|)   Std.lv  Std.all
##    herb ~
##      rocks           -0.495    0.233   -2.121    0.034   -0.495   -0.253
```

```
##       herb.18            0.413    0.098    4.236    0.000    0.413    0.418
##       tree              -0.106    0.101   -1.043    0.297   -0.106   -0.107
##    TN_log ~
##       rocks              0.177    0.251    0.706    0.480    0.177    0.089
##    soil_microbe ~
##       rocks             -0.102    0.231   -0.439    0.660   -0.125   -0.063
##    herb.18 ~
##       rocks              0.334    0.248    1.347    0.178    0.334    0.169
##
## Covariances:
##                      Estimate  Std.Err  z-value  P(>|z|)   Std.lv   Std.all
##   .soil_microbe ~~
##    .TN_log             0.604    0.145    4.162    0.000    0.745    0.754
##   .herb ~~
##    .TN_log             0.442    0.123    3.589    0.000    0.442    0.512
##   .soil_microbe ~~
##    .herb               0.256    0.108    2.374    0.018    0.316    0.362
##
## Variances:
##                      Estimate  Std.Err  z-value  P(>|z|)   Std.lv   Std.all
##    .barc.richness      0.325    0.101    3.225    0.001    0.325    0.330
##    .fungi.richness     0.850    0.157    5.397    0.000    0.850    0.863
##    .chmhtrtrphy_lg     0.935    0.170    5.511    0.000    0.935    0.950
##    .n.cycler_log       0.401    0.104    3.873    0.000    0.401    0.408
##    .saprotroph         0.840    0.156    5.383    0.000    0.840    0.854
##    .herb               0.765    0.137    5.568    0.000    0.765    0.798
##    .TN_log             0.976    0.175    5.568    0.000    0.976    0.992
##    .herb.18            0.956    0.172    5.568    0.000    0.956    0.972
##    .soil_microbe       0.656    0.185    3.541    0.000    0.996    0.996
```

```r
modindices(fit4, sort = TRUE, minimum.value = 3.5)
```

```
##                      lhs op          rhs    mi   epc sepc.lv sepc.all sepc.nox
## 38        fungi.richness ~~       TN_log 7.073 0.220   0.220    0.241    0.241
## 29          barc.richness ~~ n.cycler_log 4.802 0.309   0.309    0.855    0.855
## 43 chemoheterotrophy_log ~~       TN_log 4.418 0.177   0.177    0.186    0.186
```

```r
# TN already covaries with soil microbiome latent variable;
#   does not make sense to add path for chemoheterotrophs or fungal richness.
# Effect of soil microbiome is modeled as a single latent variable,
#   so bacterial/archeal richness and N-cyclers do not need to covary.
```