

Pseudomonas Epiphytic Growth and Virulence Analysis

2024 Summer

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Load Libraries

```
pacman::p_load(ggplot2, readxl, RColorBrewer, paletteer, viridis, wesanderson, ggbeeswarm,
  dplyr, tidyverse, devtools, emmeans, cowplot, knitr, survival, here, tibble,
  survminer, lubridate, formatR, gridExtra, ggsurvfit, gtsummary, tidycmprsk, install = FALSE)
```

Load Data

```

# Define the desired order of the treatments
strains_order <- c("194", "200", "204", "205", "215", "216", "220", "221", "227",
  "228", "B728a", "Cit7", "pisi", "Control")

strains_df <- read_excel("data/strains_R.xlsx")

epi_growth_data <- read_excel("data/pseud_epi_growth_2024summer_R.xlsx")

# Convert the strain variable to a factor with the specified order
epi_growth_data$treatment <- factor(epi_growth_data$treatment, levels = strains_order)

# Join the strains dataframe to epi growth
epi_growth_data <- left_join(epi_growth_data, strains_df, by = "treatment")

# Load virulence data
aphid_virulence_data <- read_csv("data/virulence_new_all.csv")

## Rows: 4973 Columns: 6
## -- Column specification -----
## Delimiter: ","
## chr (3): date, treatment, replicate
## dbl (3): individual, censored, time
##
## i Use `spec()` to retrieve the full column specification for this data.
## i Specify the column types or set `show_col_types = FALSE` to quiet this message.

# Convert the treatment variable to a factor with the specified order
aphid_virulence_data$treatment <- factor(aphid_virulence_data$treatment, levels = strains_order)

# Join the strains dataframe to virulence
aphid_virulence_data <- left_join(aphid_virulence_data, strains_df, by = "treatment")

```

Load colors

```

# Extract the default ggplot colors
default_colors <- (scales::hue_pal())(length(strains_order) - 1)

# Create a named vector for strain colors, ensuring 'Control' is black
strain_colors <- setNames(c(default_colors, "black"), strains_order)

# Load species colors
species_colors <- c(`P. fluorescens` = "dodgerblue2", `P. syringae` = "springgreen3",
  `P. putida` = "sienna4", Control = "black")

species_colors_manual <- c(`194` = "sienna4", `200` = "dodgerblue2", `204` = "dodgerblue2",
  `205` = "dodgerblue2", `215` = "springgreen3", `216` = "dodgerblue2", `220` = "sienna4",
  `221` = "dodgerblue2", `227` = "dodgerblue2", `228` = "dodgerblue2", B728a = "springgreen3",
  Cit7 = "springgreen3", pisi = "springgreen3", Control = "black")

```

Goals

- Create Kaplan-Meier curve for Pseud. virulence data
- Use stats (Wilcox?) to determine statistical significance of each strain
- Compare virulence data with epiphytic growth ability

Virulence Analysis

Note: *In order to help me with this analysis, I am using the following sites - Survival Analysis in R and Hazard Ratio: Interpretation & Definition.*

Calculate survival probabilities for each strain and create dataframe

Survival model

```
# Fit the survival model
km_fit <- survfit(Surv(time, censored) ~ treatment, data = aphid_virulence_data)
```

Extract survival probability

```
# Fit the survival model
km_fit <- survfit(Surv(time, censored) ~ treatment, data = aphid_virulence_data)

# Extract survival probabilities at specific time points
time_points <- c(72)
km_summary <- summary(km_fit, times = time_points)

# Calculate standard error (assuming fit includes the necessary information)
std_error <- summary(km_fit, times = time_points)$std.err

# Initialize empty lists to store the results
times_list <- list()
treatment_list <- list()
surv_prob_list <- list()
std_error_list <- list()

# Loop over each treatment group and extract survival probabilities at
# specified time points
for (i in 1:length(km_fit$strata)) {
  treatment_name <- names(km_fit$strata)[i]
  for (t in time_points) {
    idx <- which(km_summary$time == t & km_summary$strata == treatment_name)
    if (length(idx) > 0) {
      times_list <- c(times_list, t)
      treatment_list <- c(treatment_list, treatment_name)
      surv_prob_list <- c(surv_prob_list, km_summary$surv[idx])
      std_error_list <- c(std_error_list, km_summary$std.err[idx])
    } else {
```

```

        times_list <- c(times_list, t)
        treatment_list <- c(treatment_list, treatment_name)
        surv_prob_list <- c(surv_prob_list, NA)
        std_error_list <- c(std_error_list, NA)
    }
}

# Create the data frame
surv_probs <- data.frame(time = unlist(times_list), treatment = unlist(treatment_list),
    surv_prob = unlist(surv_prob_list), std_error_data = unlist(std_error_list))

# Replace 'treatment=' with an empty string
surv_probs$treatment <- gsub("treatment=", "", surv_probs$treatment)

# Convert the treatment variable to a factor with the specified order
surv_probs$treatment <- factor(surv_probs$treatment, levels = strains_order)

# Join the survival dataframe to strains
surv_probs <- left_join(surv_probs, strains_df, by = "treatment")

# Print the data frame
print(surv_probs)

```

```

##      time treatment  surv_prob std_error_data      strain      -80
## 1    72      194 0.12244898   0.019117897 field sample #8    194
## 2    72      200 0.02230483   0.009003784 field sample #14   200
## 3    72      204 0.05281690   0.013272241 field sample #17   204
## 4    72      205 0.07118644   0.014971038 field sample #18-1  205
## 5    72      215 0.22222222   0.024497697 field sample #26-1  215
## 6    72      216 0.15625000   0.021395412 field sample #26-2  216
## 7    72      220 0.17229730   0.021949800 field sample #29-2  220
## 8    72      221 0.09363296   0.017828335 field sample #29-3  221
## 9    72      227 0.16949153   0.021844143 field sample #35    227
## 10   72      228 0.10447761   0.018684542 field sample #36    228
## 11   72      B728a 0.10996564   0.018339411      B728a ΔFlgK      B728a
## 12   72      Cit7 0.17073171   0.022210773      Cit7          14
## 13   72      Control 0.81678082   0.011319228      Control      Control
## 14   72      pisi 0.74151436   0.022370650      1704B          6
##      species_complex      species_match      collector
## 1    P. fluorescens      P. paralactis      Kathryn L Herr
## 2    P. fluorescens      P. fluorescens      Kathryn L Herr
## 3    P. fluorescens      P. rhodesiae      Kathryn L Herr
## 4    P. fluorescens      P. orientalis      Kathryn L Herr
## 5      P. syringae      P. syringae      Kathryn L Herr
## 6      P. putida      P. putida      Kathryn L Herr
## 7    P. fluorescens      P. pergaminensis      Kathryn L Herr
## 8    P. fluorescens      P. fluorescens      Kathryn L Herr
## 9    P. fluorescens      P. pergaminensis      Kathryn L Herr
## 10   P. syringae      P. lijiangensis      Kathryn L Herr
## 11   P. syringae P. syringae pv. Syringae Alan Collmer/J. Lindemann?
## 12   P. syringae      P. syringae      <NA>
## 13      Control      Control      Control

```

```
## 14      P. syringae          P. syringae          David Baltrus
##              source      ANI              genome_match      Reseq/Genbank
## 1      Pisum sativum      98.44 Pseudomonas paralactis DSM 29164 GCA_001439735.1
## 2      Pisum sativum      <NA>              <NA>              <NA>
## 3      Pisum sativum      98.92 Pseudomonas rhodesiae CIP104664 GCF_024169765.1
## 4      Pisum sativum      95.78 Pseudomonas orientalis DSM 17489 GCF_001439815.1
## 5      Pisum sativum      95.57 Pseudomonas syringae DSM 10604 GCA_000597765.1
## 6      Pisum sativum      95.42 Pseudomonas putida S13.1.2 GCF_000498395.2
## 7      Pisum sativum      98.64 Pseudomonas pergaminensis 1008 GCF_024112395.1
## 8      Pisum sativum      <NA>              <NA>              <NA>
## 9      Pisum sativum      98.22 Pseudomonas pergaminensis 1008 GCF_024112395.1
## 10     Pisum sativum      96.6 Pseudomonas lijiangensis LJ2 GCF_018968705.1
## 11 Phaseolus vulgaris      <NA>              <NA>              CP000075
## 12     Pisum sativum      <NA>              <NA>              AEAJ 00000000
## 13              Control Control              Control              Control
## 14 Citrus leaf surface      <NA>              <NA>              AEAI 00000000
```

```
##
```

```
## 1
```

```
## 2
```

```
## 3
```

```
## 4
```

```
## 5
```

```
## 6
```

```
## 7
```

```
## 8
```

```
## 9
```

```
## 10
```

```
## 11
```

```
177. https://doi.org/10.1146/annurev.py.28.090190.001103
```

```
## 12
```

```
177. https://doi.org/10.1146/annurev.py.28.090190.001103
```

```
## 13
```

```
## 14 Baltrus, D. A., Nishimura, M. T., Romanchuk, A., Chang, J. H., Mukhtar, M. S., Cherkis, K., Roach
```

Post-hoc test for survival

```
# Perform pairwise log-rank tests between treatment groups and the control
# group
pairwise_tests_treatment <- pairwise_survdif(Surv(time, censored) ~ treatment, data = aphid_virulence_

# Print the pairwise tests
print(pairwise_tests_treatment)
```

```
##
```

```
## Pairwise comparisons using Log-Rank test
```

```
##
```

```
## data: aphid_virulence_data and treatment
```

```
##
```

```
##      194      200      204      205      215      216      220      221      227
```

```
## 200      2.3e-12 -      -      -      -      -      -      -      -
```

```
## 204      1.6e-08 0.26489 -      -      -      -      -      -      -
```

```
## 205      3.7e-06 0.04009 0.38289 -      -      -      -      -      -
```

```
## 215      4.6e-05 < 2e-16 < 2e-16 7.8e-16 -      -      -      -      -
## 216      0.20589 3.0e-06 0.00054 0.00915 1.3e-05 -      -      -      -
## 220      1.2e-05 0.34987 0.95899 0.61067 7.5e-10 0.04787 -      -      -
## 221      0.06597 1.7e-06 0.00063 0.01560 3.0e-08 0.57084 0.05899 -      -
## 227      0.00048 0.05275 0.34910 0.75895 1.7e-08 0.14997 0.65606 0.23547 -
## 228      0.02457 2.3e-05 0.00265 0.03730 5.6e-09 0.52684 0.02668 0.75895 0.15756
## B728a    0.30241 3.8e-15 2.0e-10 1.5e-07 0.00193 0.15635 0.00089 0.00913 0.00558
## Cit7     0.97833 6.8e-10 1.0e-06 8.4e-05 0.00044 0.38289 0.00217 0.11439 0.01459
## Control < 2e-16 < 2e-16 < 2e-16 < 2e-16 < 2e-16 < 2e-16 < 2e-16 < 2e-16 < 2e-16
## pisi     < 2e-16 < 2e-16 < 2e-16 < 2e-16 < 2e-16 < 2e-16 < 2e-16 < 2e-16 < 2e-16
##          228      B728a    Cit7      Control
## 200      -      -      -      -
## 204      -      -      -      -
## 205      -      -      -      -
## 215      -      -      -      -
## 216      -      -      -      -
## 220      -      -      -      -
## 221      -      -      -      -
## 227      -      -      -      -
## 228      -      -      -      -
## B728a    0.00265 -      -      -
## Cit7     0.05646 0.46912 -      -
## Control < 2e-16 < 2e-16 < 2e-16 -
## pisi     < 2e-16 < 2e-16 < 2e-16 0.00042
##
## P value adjustment method: BH
```

```
# Perform pairwise log-rank tests between species_complex groups and the
# control group
pairwise_tests_species <- pairwise_survdif(Surv(time, censored) ~ species_complex,
      data = aphid_virulence_data)

# Print the pairwise tests
print(pairwise_tests_species)
```

```
##
## Pairwise comparisons using Log-Rank test
##
## data: aphid_virulence_data and species_complex
##
##          Control P. fluorescens P. putida
## P. fluorescens < 2e-16 -      -
## P. putida      < 2e-16 0.0084 -
## P. syringae    < 2e-16 < 2e-16 2.6e-10
##
## P value adjustment method: BH
```

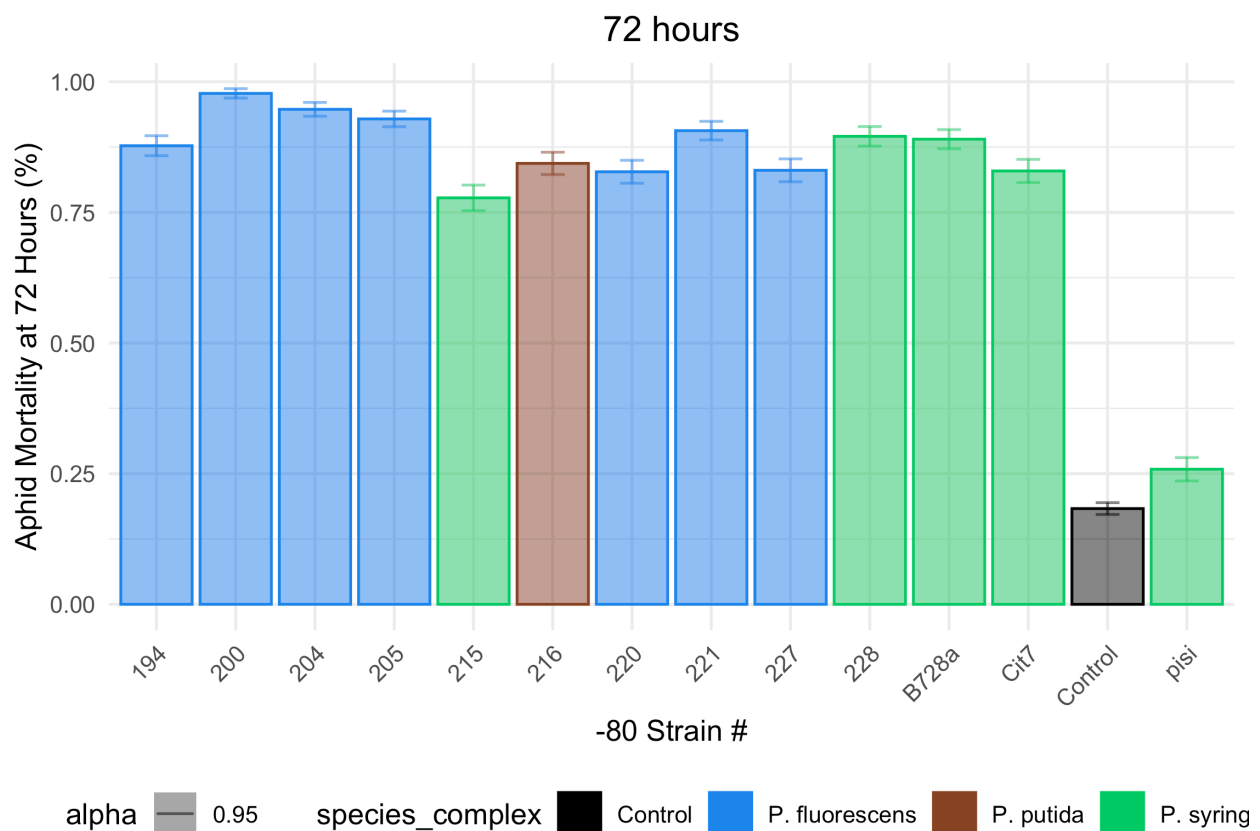
Plot Survival Dataframe at different times

```
surv_plot <- ggplot(data = surv_probs, aes(x = treatment, y = (1-surv_prob), color = species_complex, f
      geom_col() +
```

```

geom_errorbar(aes(ymin = (1-surv_prob) - std_error_data, ymax = (1-surv_prob) + std_error_data),
              width = 0.3, # Width of error bars
              position = position_dodge(width = 0.9)) + # Dodge bars slightly
labs(title = "72 hours",
     x = "-80 Strain #",
     y = "Aphid Mortality at 72 Hours (%)") +
theme_minimal() +
scale_fill_manual(values = species_colors) +
scale_color_manual(values = species_colors) +
theme(plot.title = element_text(hjust = 0.5),
      axis.text.x = element_text(angle = 45, hjust = 1), legend.position = "bottom")
surv_plot

```



Make Kaplan-Meier Plot

Cohort Survival Curve

```

# Plot the Kaplan-Meier survival curves by treatment
surv_curv_treatment <- survfit2(Surv(time, censored) ~ treatment, data = aphid_virulence_data) %>%
  ggsvfit(type = "survival") + labs(x = "Hours", y = "Overall survival probability") +
  theme_minimal() + scale_fill_manual(values = species_colors_manual) + scale_color_manual(values = s
  theme(plot.title = element_text(hjust = 0.5), axis.text.x = element_text(angle = 45,

```

```

    hjust = 1), legend.position = "right")

# Plot the Kaplan-Meier survival curves by species complex
surv_curv_species <- survfit2(Surv(time, censored) ~ species_complex, data = aphid_virulence_data) %>%
  ggsurvfit(type = "survival") + labs(x = "Hours", y = "Overall survival probability") +
  theme_minimal() + scale_fill_manual(values = species_colors) + scale_color_manual(values = species_
  theme(plot.title = element_text(hjust = 0.5), axis.text.x = element_text(angle = 45,
    hjust = 1), legend.position = "right")

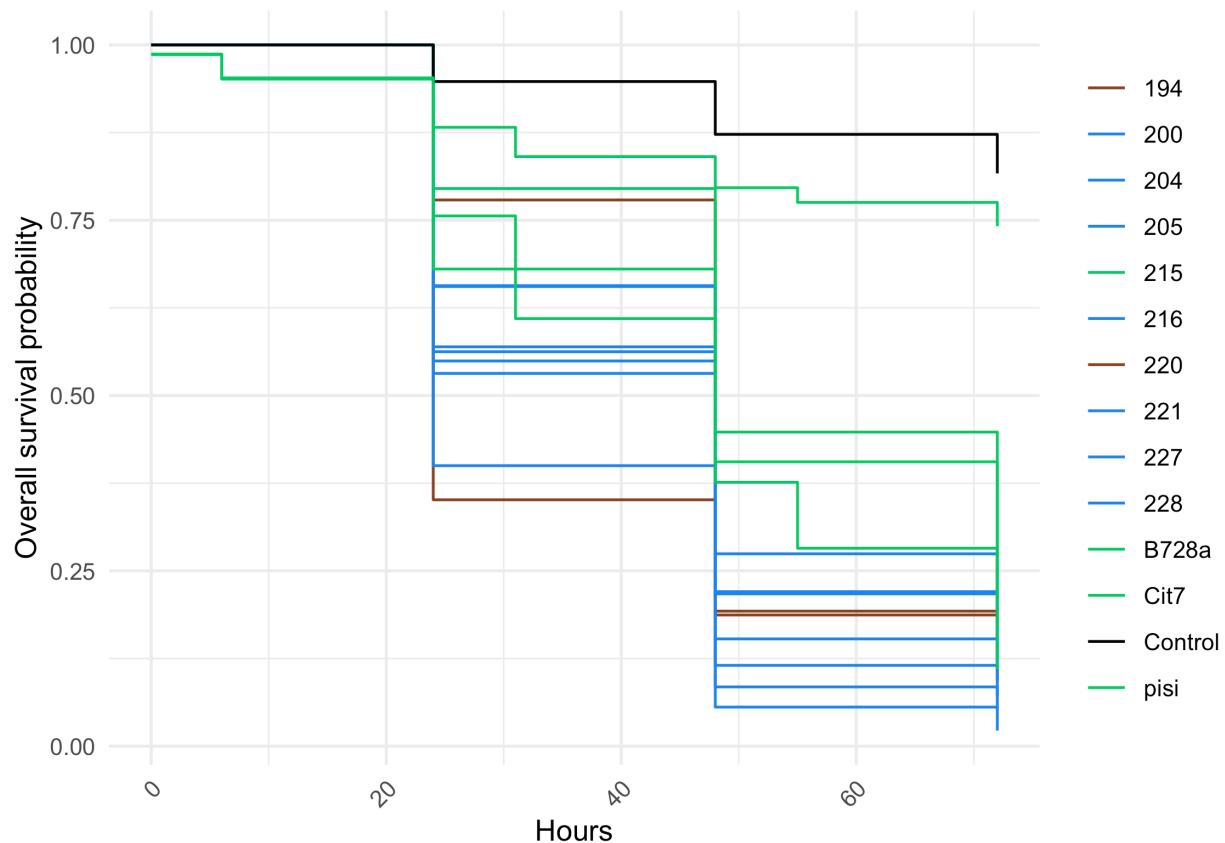
# show plots
surv_curv_treatment

```

```

## Warning: No shared levels found between `names(values)` of the manual scale and the
## data's fill values.

```

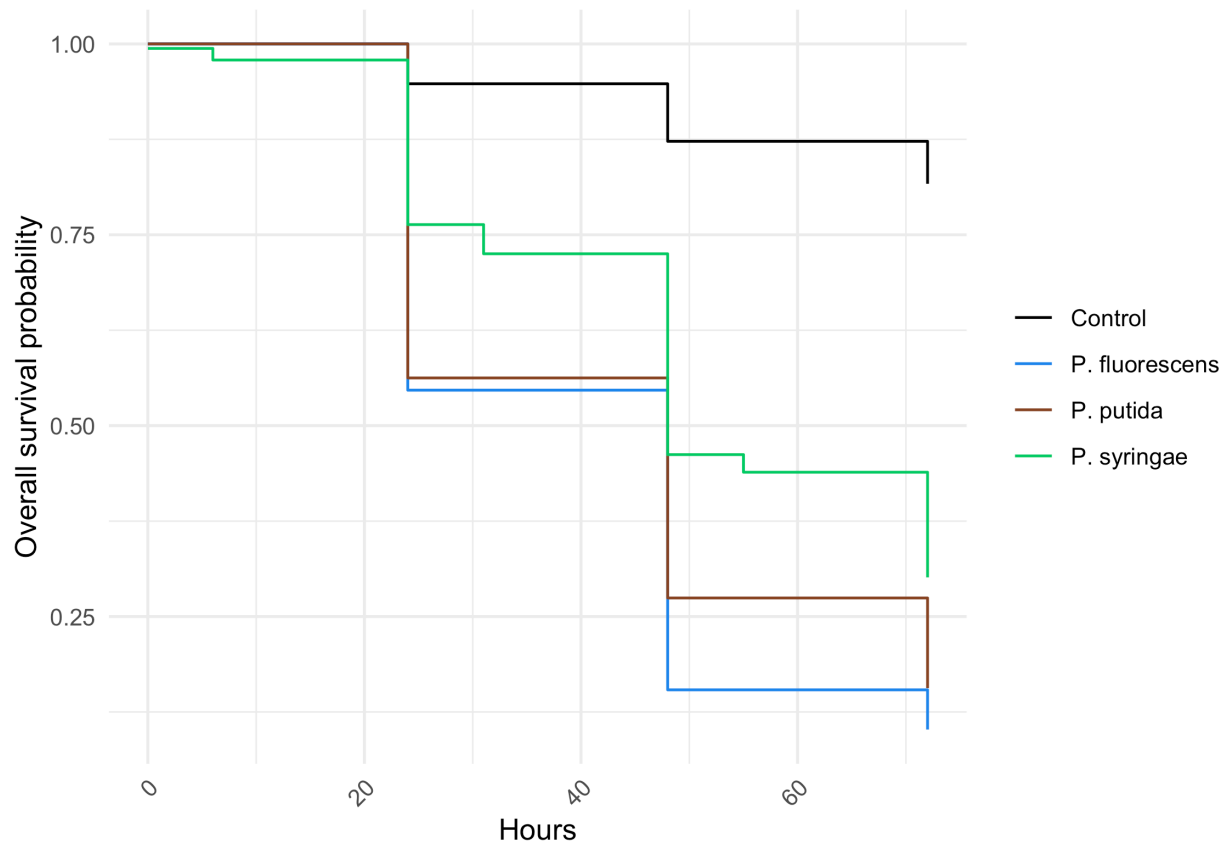


```
surv_curv_species
```

```

## Warning: No shared levels found between `names(values)` of the manual scale and the
## data's fill values.

```

Epiphytic Growth Analysis

Calculate mean/variance epiphytic growth ability

```
# Replace NA with a lower value or remove them for visualization Remove rows
# with NA in CFU_per_10_leafdiscs
epi_growth_clean <- epi_growth_data %>%
  filter(!is.na(CFU_per_10_leafdiscs))

# Convert CFU_per_10_leafdiscs to numeric, handling scientific notation
epi_growth_clean$CFU_per_10_leafdiscs <- as.numeric(gsub("<", "", epi_growth_clean$CFU_per_10_leafdiscs))

## Warning: NAs introduced by coercion

# Subset data to remove unfinished strains and extract the letter part from the
# 'plant_rep' column
epi_growth_clean <- epi_growth_clean %>%
  mutate(plant_rep_letter = substr(plant_rep, 1, 1))

# Calculate the mean and SD CFU per strain for each plant rep letter and retain
# the specified columns
epi_growth_mean_per_rep <- epi_growth_clean %>%
```

```

group_by(treatment, plant_rep_letter) %>%
summarise(mean_CFU_per_10_leafdiscs = mean(CFU_per_10_leafdiscs, na.rm = TRUE),
  sd_CFU = sd(CFU_per_10_leafdiscs, na.rm = TRUE), sample = first(sample),
  person = first(person), species_complex = first(species_complex), block = first(block),
  treatment = first(treatment), plant_rep = first(plant_rep), .groups = "drop")

# Calculate the mean and SD CFU per strain for each plant rep letter and retain
# the specified columns
epi_growth_mean_per_strain <- epi_growth_clean %>%
  group_by(treatment) %>%
  summarise(mean_CFU_per_10_leafdiscs = mean(CFU_per_10_leafdiscs, na.rm = TRUE),
    sd_CFU = sd(CFU_per_10_leafdiscs, na.rm = TRUE), sample = first(sample),
    person = first(person), species_complex = first(species_complex), block = first(block),
    treatment = first(treatment), plant_rep = first(plant_rep), .groups = "drop")

epi_growth_mean_per_strain

```

```

## # A tibble: 13 x 8
##   treatment mean_CFU_per_10_leafdi~1 sd_CFU sample person species_complex block
##   <chr>          <dbl>    <dbl> <chr>  <chr>  <chr>      <chr>
## 1 194          156472. 1.43e5 194-A1 Havi   P. fluorescens 2-1
## 2 200          261214. 1.81e5 200-A1 Havi   P. fluorescens 2-6
## 3 204          186888. 1.89e5 204-A1 Havi   P. fluorescens 2-3
## 4 205          207750. 2.40e5 205-A1 Sara   P. fluorescens 3
## 5 215           1000 0      215-A1 Sara   P. syringae    3
## 6 216           6788. 7.61e3 216-A1 Sara   P. putida      3
## 7 220          172543. 1.89e5 220-A1 Sara   P. fluorescens 5
## 8 221          259556. 1.93e5 221-A1 Havi   P. fluorescens 2-7
## 9 227           82872. 1.53e5 227-A1 Sara   P. fluorescens 5
## 10 228          22875 2.37e4 228-A1 Sara   P. syringae    5
## 11 B728a        71144. 1.36e5 B728a~ Sara   P. syringae    4
## 12 Cit7         61906. 1.17e5 Cit7~~ Havi   P. syringae    2-3
## 13 pisi         11607. 3.51e4 pisi~~ Havi   P. syringae    2-4
## # i abbreviated name: 1: mean_CFU_per_10_leafdiscs
## # i 1 more variable: plant_rep <chr>

```

Plot Epiphytic Growth

```

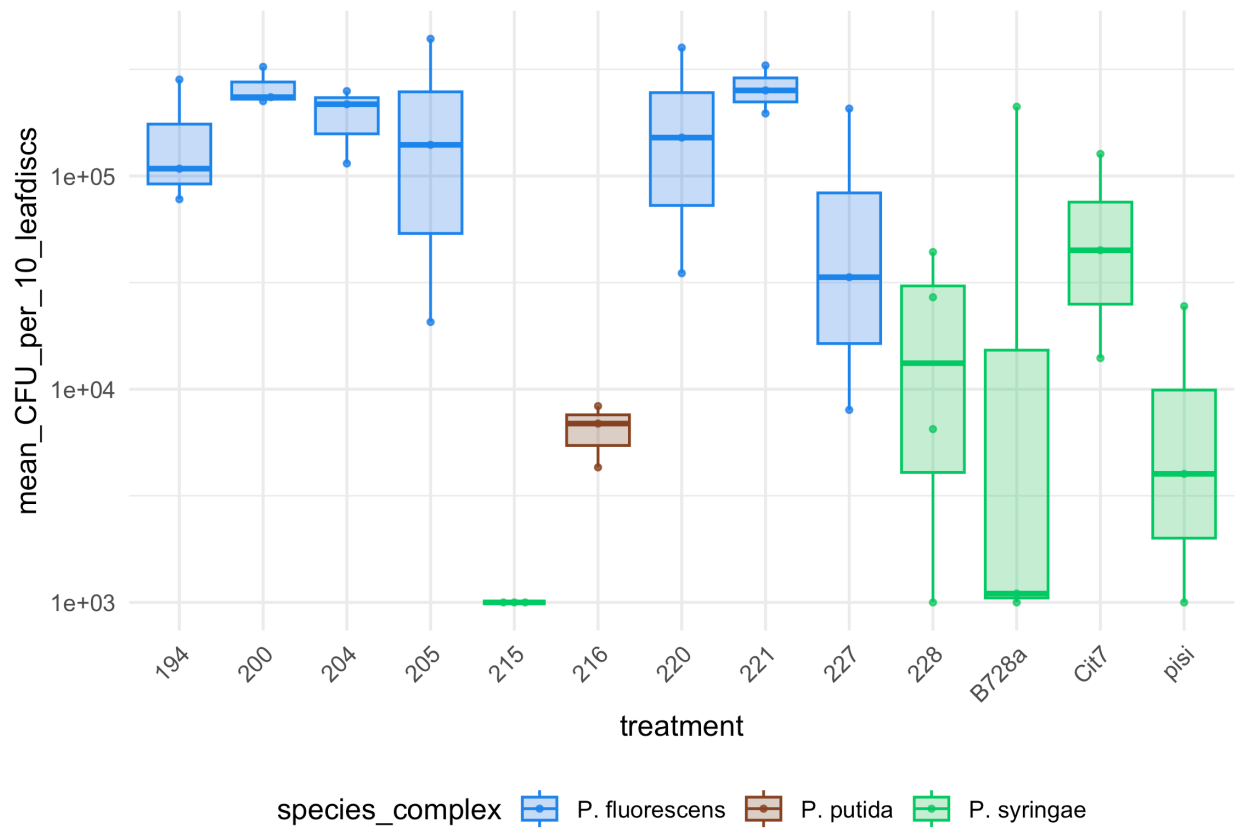
# plot by treatment
epi_growth_treatment <- ggplot(data = epi_growth_mean_per_rep, aes(x = treatment,
  y = mean_CFU_per_10_leafdiscs, color = species_complex, fill = species_complex)) +
  geom_boxplot(outlier.shape = NA, alpha = 0.3) + geom_beeswarm(stroke = 0.5, size = 0.8,
  alpha = 0.8) + theme_minimal() + scale_fill_manual(values = species_colors) +
  scale_color_manual(values = species_colors) + scale_y_log10() + theme(plot.title = element_text(hjust = 1),
  axis.text.x = element_text(angle = 45, hjust = 1), legend.position = "bottom")

# plot by species complex
epi_growth_species <- ggplot(data = epi_growth_mean_per_rep, aes(x = species_complex,
  y = mean_CFU_per_10_leafdiscs, color = species_complex, fill = species_complex)) +
  geom_boxplot(outlier.shape = NA, alpha = 0.3) + geom_beeswarm(stroke = 0.5, size = 0.8,
  alpha = 0.8) + theme_minimal() + scale_fill_manual(values = species_colors) +

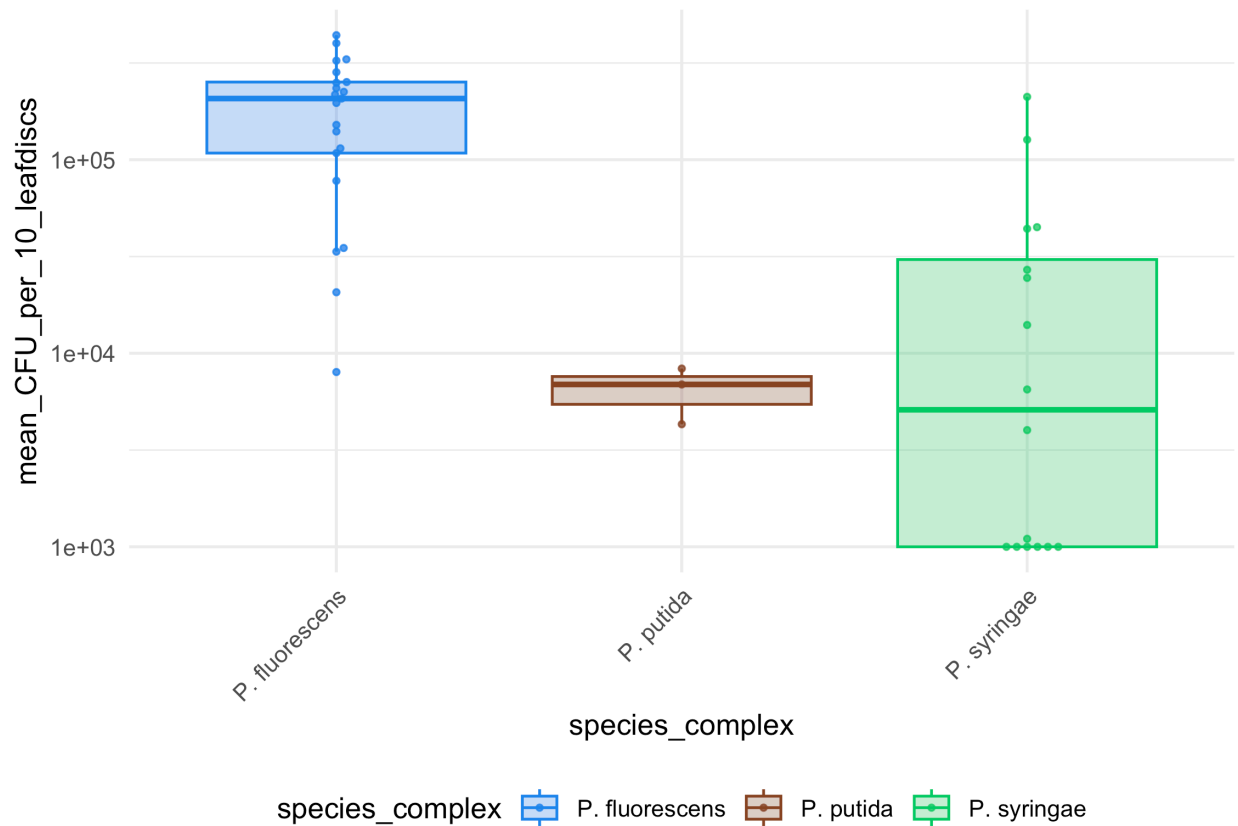
```

```
scale_color_manual(values = species_colors) + scale_y_log10() + theme(plot.title = element_text(hjust = 1))
axis.text.x = element_text(angle = 45, hjust = 1), legend.position = "bottom")
```

```
# show plots
epi_growth_treatment
```



```
epi_growth_species
```



Epiphytic growth stats

```
# stats per treatment
epi_growth_mod_treatment = lm(mean_CFU_per_10_leafdiscs ~ treatment, data = epi_growth_mean_per_rep)

emmeans(epi_growth_mod_treatment, pairwise ~ treatment)
```

```
## $emmeans
## treatment emmean      SE df lower.CL upper.CL
## 194        156472 58153 27    37151  275793
## 200        261214 58153 27   141893  380535
## 204        193911 58153 27    74590  313232
## 205        200222 58153 27    80901  319543
## 215           1000 58153 27  -118321  120321
## 216           6511 58153 27  -112810  125832
## 220        195467 58153 27    76146  314788
## 221        259556 58153 27   140234  378877
## 227          82872 58153 27   -36449  202193
## 228         19625 50362 27   -83710  122960
## B728a        71144 58153 27   -48177  190466
## Cit7         61906 58153 27   -57416  181227
## pisi          9839 58153 27  -109482  129160
##
```

```

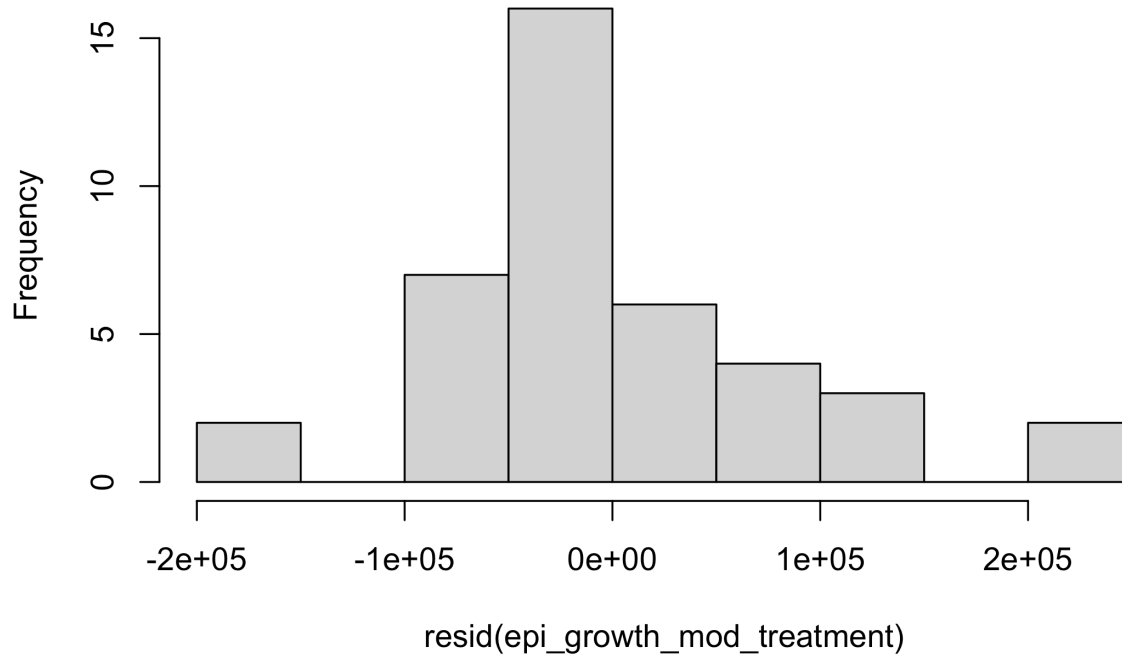
## Confidence level used: 0.95
##
## $contrasts
## contrast      estimate      SE df t.ratio p.value
## 194 - 200      -104742 82241 27  -1.274  0.9829
## 194 - 204       -37439 82241 27  -0.455  1.0000
## 194 - 205       -43750 82241 27  -0.532  1.0000
## 194 - 215       155472 82241 27   1.890  0.7869
## 194 - 216       149961 82241 27   1.823  0.8218
## 194 - 220       -38994 82241 27  -0.474  1.0000
## 194 - 221      -103083 82241 27  -1.253  0.9849
## 194 - 227        73600 82241 27   0.895  0.9992
## 194 - 228       136847 76930 27   1.779  0.8434
## 194 - B728a      85328 82241 27   1.038  0.9970
## 194 - Cit7       94567 82241 27   1.150  0.9926
## 194 - pisi      146633 82241 27   1.783  0.8415
## 200 - 204        67303 82241 27   0.818  0.9997
## 200 - 205        60992 82241 27   0.742  0.9999
## 200 - 215       260214 82241 27   3.164  0.1318
## 200 - 216       254703 82241 27   3.097  0.1503
## 200 - 220        65747 82241 27   0.799  0.9998
## 200 - 221        1658 82241 27   0.020  1.0000
## 200 - 227       178342 82241 27   2.169  0.6194
## 200 - 228       241589 76930 27   3.140  0.1381
## 200 - B728a     190069 82241 27   2.311  0.5282
## 200 - Cit7      199308 82241 27   2.423  0.4582
## 200 - pisi     251375 82241 27   3.057  0.1624
## 204 - 205       -6311 82241 27  -0.077  1.0000
## 204 - 215       192911 82241 27   2.346  0.5064
## 204 - 216       187400 82241 27   2.279  0.5489
## 204 - 220       -1556 82241 27  -0.019  1.0000
## 204 - 221      -65644 82241 27  -0.798  0.9998
## 204 - 227      111039 82241 27   1.350  0.9733
## 204 - 228       174286 76930 27   2.266  0.5573
## 204 - B728a     122767 82241 27   1.493  0.9456
## 204 - Cit7      132006 82241 27   1.605  0.9132
## 204 - pisi     184072 82241 27   2.238  0.5748
## 205 - 215       199222 82241 27   2.422  0.4589
## 205 - 216       193711 82241 27   2.355  0.5003
## 205 - 220        4756 82241 27   0.058  1.0000
## 205 - 221      -59333 82241 27  -0.721  0.9999
## 205 - 227      117350 82241 27   1.427  0.9601
## 205 - 228       180597 76930 27   2.348  0.5052
## 205 - B728a     129078 82241 27   1.569  0.9246
## 205 - Cit7      138317 82241 27   1.682  0.8853
## 205 - pisi     190383 82241 27   2.315  0.5258
## 215 - 216       -5511 82241 27  -0.067  1.0000
## 215 - 220      -194467 82241 27  -2.365  0.4945
## 215 - 221      -258556 82241 27  -3.144  0.1372
## 215 - 227       -81872 82241 27  -0.996  0.9979
## 215 - 228       -18625 76930 27  -0.242  1.0000
## 215 - B728a     -70144 82241 27  -0.853  0.9995
## 215 - Cit7      -60906 82241 27  -0.741  0.9999
## 215 - pisi      -8839 82241 27  -0.107  1.0000

```

```
## 216 - 220      -188956 82241 27  -2.298  0.5368
## 216 - 221      -253044 82241 27  -3.077  0.1563
## 216 - 227      -76361 82241 27  -0.928  0.9989
## 216 - 228      -13114 76930 27  -0.170  1.0000
## 216 - B728a    -64633 82241 27  -0.786  0.9998
## 216 - Cit7     -55394 82241 27  -0.674  1.0000
## 216 - pisi     -3328 82241 27   -0.040  1.0000
## 220 - 221      -64089 82241 27  -0.779  0.9998
## 220 - 227      112594 82241 27   1.369  0.9704
## 220 - 228      175842 76930 27   2.286  0.5443
## 220 - B728a    124322 82241 27   1.512  0.9409
## 220 - Cit7     133561 82241 27   1.624  0.9068
## 220 - pisi     185628 82241 27   2.257  0.5626
## 221 - 227      176683 82241 27   2.148  0.6323
## 221 - 228      239931 76930 27   3.119  0.1441
## 221 - B728a    188411 82241 27   2.291  0.5410
## 221 - Cit7     197650 82241 27   2.403  0.4705
## 221 - pisi     249717 82241 27   3.036  0.1688
## 227 - 228       63247 76930 27   0.822  0.9997
## 227 - B728a    11728 82241 27   0.143  1.0000
## 227 - Cit7     20967 82241 27   0.255  1.0000
## 227 - pisi     73033 82241 27   0.888  0.9993
## 228 - B728a    -51519 76930 27  -0.670  1.0000
## 228 - Cit7     -42281 76930 27  -0.550  1.0000
## 228 - pisi       9786 76930 27   0.127  1.0000
## B728a - Cit7    9239 82241 27   0.112  1.0000
## B728a - pisi    61306 82241 27   0.745  0.9999
## Cit7 - pisi     52067 82241 27   0.633  1.0000
##
## P value adjustment: tukey method for comparing a family of 13 estimates
```

```
hist(resid(epi_growth_mod_treatment))
```

Histogram of resid(epi_growth_mod_treatment)

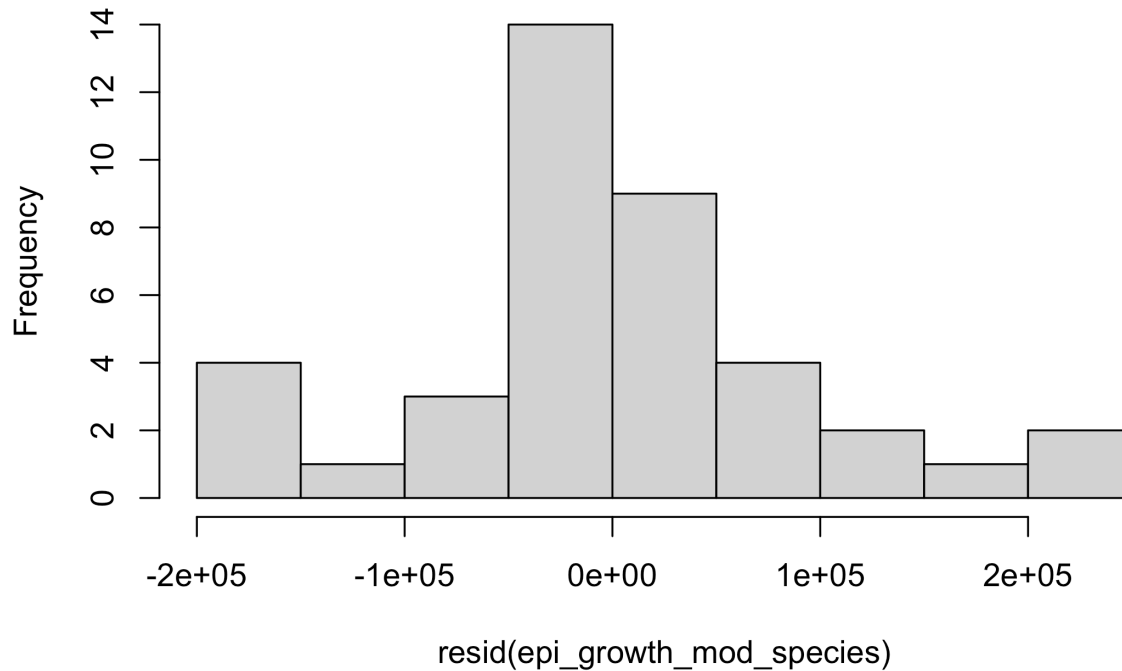


```
# stats per species
epi_growth_mod_species = lm(mean_CFU_per_10_leafdiscs ~ species_complex, data = epi_growth_mean_per_rep)
emmeans(epi_growth_mod_species, pairwise ~ species_complex)
```

```
## $emmeans
## species_complex emmean SE df lower.CL upper.CL
## P. fluorescens 192816 21344 37 149568 236064
## P. putida 6511 56472 37 -107912 120934
## P. syringae 31885 24453 37 -17661 81432
##
## Confidence level used: 0.95
##
## $contrasts
## contrast estimate SE df t.ratio p.value
## P. fluorescens - P. putida 186305 60371 37 3.086 0.0105
## P. fluorescens - P. syringae 160931 32458 37 4.958 <.0001
## P. putida - P. syringae -25374 61539 37 -0.412 0.9108
##
## P value adjustment: tukey method for comparing a family of 3 estimates
```

```
hist(resid(epi_growth_mod_species))
```

Histogram of resid(epi_growth_mod_species)



Combine epiphytic and virulence data

```
# Combine strain_stats and surv_probs using left_join
epi_virulence_data <- left_join(epi_growth_mean_per_strain, surv_probs, by = "treatment")

# Print the combined data
print(epi_virulence_data)
```

```
## # A tibble: 13 x 21
##   treatment mean_CFU_per_10_leaf~1 sd_CFU sample person species_complex.x block
##   <chr>          <dbl>    <dbl> <chr>  <chr>    <chr>          <chr>
## 1 194          156472. 1.43e5 194-A1 Havi    P. fluorescens 2-1
## 2 200          261214. 1.81e5 200-A1 Havi    P. fluorescens 2-6
## 3 204          186888. 1.89e5 204-A1 Havi    P. fluorescens 2-3
## 4 205          207750. 2.40e5 205-A1 Sara    P. fluorescens 3
## 5 215           1000 0        215-A1 Sara    P. syringae    3
## 6 216           6788. 7.61e3 216-A1 Sara    P. putida      3
## 7 220          172543. 1.89e5 220-A1 Sara    P. fluorescens 5
## 8 221          259556. 1.93e5 221-A1 Havi    P. fluorescens 2-7
## 9 227           82872. 1.53e5 227-A1 Sara    P. fluorescens 5
## 10 228          22875. 2.37e4 228-A1 Sara    P. syringae    5
## 11 B728a       71144. 1.36e5 B728a~ Sara    P. syringae    4
## 12 Cit7        61906. 1.17e5 Cit7~~ Havi    P. syringae    2-3
```



```
## 13 pisi 11607. 3.51e4 pisi-- Havi P. syringae 2-4
## # i abbreviated name: 1: mean_CFU_per_10_leafdiscs
## # i 14 more variables: plant_rep <chr>, time <dbl>, surv_prob <dbl>,
## # std_error_data <dbl>, strain <chr>, `~80` <chr>, species_complex.y <chr>,
## # species_match <chr>, collector <chr>, source <chr>, ANI <chr>,
## # genome_match <chr>, `Reseq/Genbank` <chr>, Paper <chr>

# Calculate correlation coefficient between survival probability and epiphytic
# growth ability
correlation <- -1 * cor(eps_virulence_data$surv_prob, eps_virulence_data$mean_CFU_per_10_leafdiscs,
  method = "pearson")
```

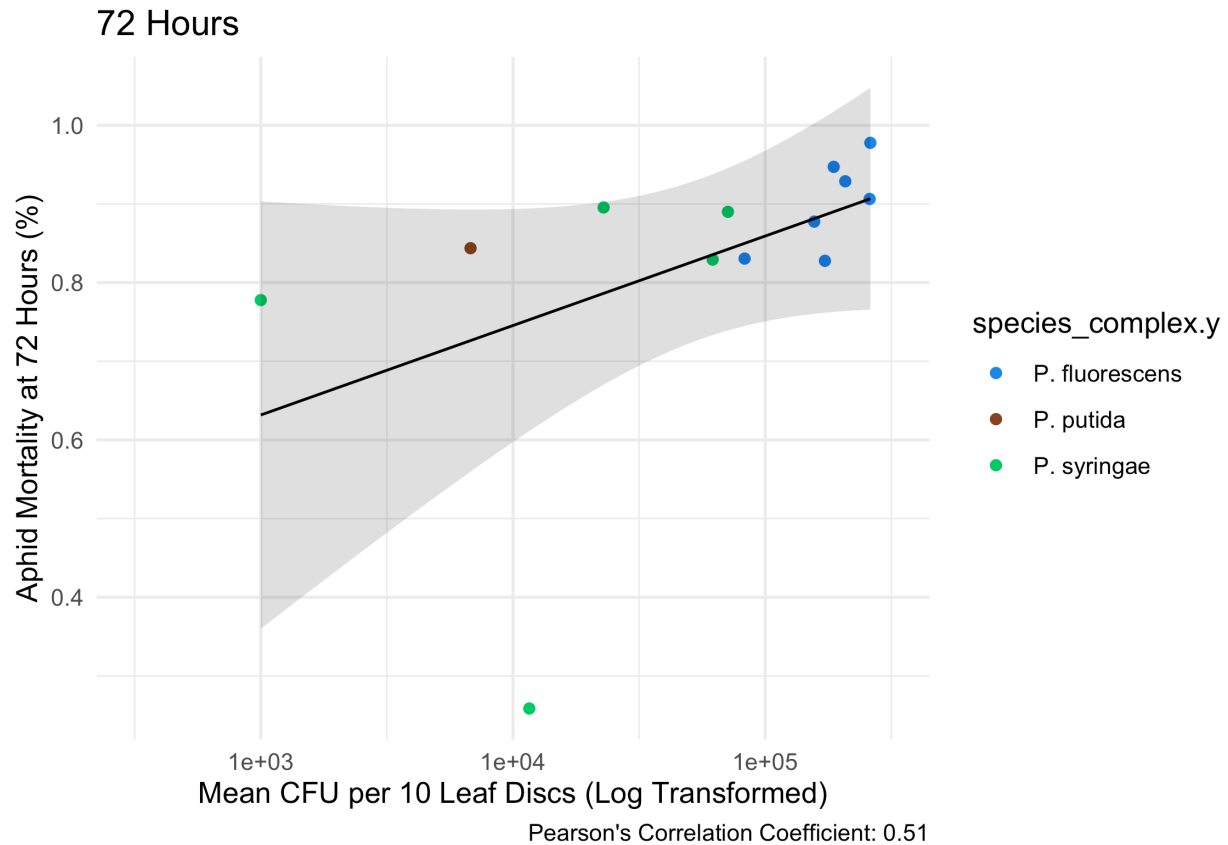
Plot eps virulence plot

```
# Create scatter plot for 72 hours
eps_virulence_plot_strains <- ggplot(eps_virulence_data, aes(x = mean_CFU_per_10_leafdiscs, y = 1-surv_
  geom_point(aes(color = species_complex.y)) +
  geom_smooth(method = "lm", se = TRUE, color = "black", fill = "black", alpha = 0.15, linewidth = 0.5)
  labs(title = "72 Hours",
    x = "Mean CFU per 10 Leaf Discs (Log Transformed)",
    y = "Aphid Mortality at 72 Hours (%)",
    caption = paste("Pearson's Correlation Coefficient:", round(correlation, 2))) +
  scale_x_continuous(trans = "log10", limits = c(10^2.5, 10^5.5)) + # Apply log transformation to x-axis
  scale_fill_manual(values = species_colors) +
  scale_color_manual(values = species_colors) +
  theme_minimal()

eps_virulence_plot_strains

## `geom_smooth()` using formula = 'y ~ x'

## Warning: No shared levels found between `names(values)` of the manual scale and the
## data's fill values.
```



Session Information

```
devtools::session_info()
```

```
## - Session info -----
## setting value
## version R version 4.4.0 (2024-04-24)
## os      macOS Ventura 13.4
## system  x86_64, darwin20
## ui      X11
## language (EN)
## collate en_US.UTF-8
## ctype   en_US.UTF-8
## tz      America/New_York
## date    2024-07-01
## pandoc  3.1.11 @ /Applications/RStudio.app/Contents/Resources/app/quarto/bin/tools/x86_64/ (via rm
##
## - Packages -----
## ! package      * version date (UTC) lib source
## P abind         1.4-5   2016-07-21 [?] RSPM
```

##	P backports	1.4.1	2021-12-13	[?]	CRAN	(R 4.4.0)
##	P beeswarm	0.4.0	2021-06-01	[?]	CRAN	(R 4.4.0)
##	P bit	4.0.5	2022-11-15	[?]	CRAN	(R 4.4.0)
##	P bit64	4.0.5	2020-08-30	[?]	CRAN	(R 4.4.0)
##	P broom	1.0.6	2024-05-17	[?]	CRAN	(R 4.4.0)
##	P broom.helpers	1.15.0	2024-04-05	[?]	CRAN	(R 4.4.0)
##	P cachem	1.0.8	2023-05-01	[?]	CRAN	(R 4.4.0)
##	P car	3.1-2	2023-03-30	[?]	RSPM	
##	P carData	3.0-5	2022-01-06	[?]	RSPM	
##	P cellranger	1.1.0	2016-07-27	[?]	CRAN	(R 4.4.0)
##	P cli	3.6.2	2023-12-11	[?]	CRAN	(R 4.4.0)
##	P colorspace	2.1-0	2023-01-23	[?]	CRAN	(R 4.4.0)
##	P cowplot	* 1.1.3	2024-01-22	[?]	CRAN	(R 4.4.0)
##	P crayon	1.5.2	2022-09-29	[?]	CRAN	(R 4.4.0)
##	P data.table	1.15.4	2024-03-30	[?]	CRAN	(R 4.4.0)
##	P devtools	* 2.4.5	2022-10-11	[?]	RSPM	
##	P digest	0.6.35	2024-03-11	[?]	CRAN	(R 4.4.0)
##	P dplyr	* 1.1.4	2023-11-17	[?]	CRAN	(R 4.4.0)
##	P ellipsis	0.3.2	2021-04-29	[?]	RSPM	
##	P emmeans	* 1.10.2	2024-05-20	[?]	RSPM	
##	P estimability	1.5.1	2024-05-12	[?]	RSPM	
##	P evaluate	0.23	2023-11-01	[?]	CRAN	(R 4.4.0)
##	P fansi	1.0.6	2023-12-08	[?]	CRAN	(R 4.4.0)
##	P farver	2.1.2	2024-05-13	[?]	CRAN	(R 4.4.0)
##	P fastmap	1.1.1	2023-02-24	[?]	CRAN	(R 4.4.0)
##	P forcats	* 1.0.0	2023-01-29	[?]	CRAN	(R 4.4.0)
##	P formatR	* 1.14	2023-01-17	[?]	RSPM	
##	P fs	1.6.4	2024-04-25	[?]	CRAN	(R 4.4.0)
##	P generics	0.1.3	2022-07-05	[?]	CRAN	(R 4.4.0)
##	P ggbeeswarm	* 0.7.2	2023-04-29	[?]	CRAN	(R 4.4.0)
##	P ggplot2	* 3.5.1	2024-04-23	[?]	CRAN	(R 4.4.0)
##	P ggpubr	* 0.6.0	2023-02-10	[?]	RSPM	
##	P ggsignif	0.6.4	2022-10-13	[?]	RSPM	
##	P ggsvrfit	* 1.1.0	2024-05-08	[?]	CRAN	(R 4.4.0)
##	P glue	1.7.0	2024-01-09	[?]	CRAN	(R 4.4.0)
##	P gridExtra	* 2.3	2017-09-09	[?]	RSPM	
##	P gt	0.10.1	2024-01-17	[?]	CRAN	(R 4.4.0)
##	P gtable	0.3.5	2024-04-22	[?]	CRAN	(R 4.4.0)
##	P gtsummary	* 1.7.2	2023-07-15	[?]	CRAN	(R 4.4.0)
##	P here	* 1.0.1	2020-12-13	[?]	CRAN	(R 4.4.0)
##	P hms	1.1.3	2023-03-21	[?]	CRAN	(R 4.4.0)
##	P htmltools	0.5.8.1	2024-04-04	[?]	CRAN	(R 4.4.0)
##	P htmlwidgets	1.6.4	2023-12-06	[?]	CRAN	(R 4.4.0)
##	P httpuv	1.6.15	2024-03-26	[?]	RSPM	
##	P km.ci	0.5-6	2022-04-06	[?]	RSPM	
##	P KMsurv	0.1-5	2012-12-03	[?]	RSPM	
##	P knitr	* 1.46	2024-04-06	[?]	CRAN	(R 4.4.0)
##	P labeling	0.4.3	2023-08-29	[?]	CRAN	(R 4.4.0)
##	P later	1.3.2	2023-12-06	[?]	RSPM	
##	P lattice	0.22-6	2024-03-20	[?]	CRAN	(R 4.4.0)
##	P lifecycle	1.0.4	2023-11-07	[?]	CRAN	(R 4.4.0)
##	P lubridate	* 1.9.3	2023-09-27	[?]	CRAN	(R 4.4.0)
##	P magrittr	2.0.3	2022-03-30	[?]	CRAN	(R 4.4.0)
##	P Matrix	1.7-0	2024-03-22	[?]	CRAN	(R 4.4.0)

##	P memoise	2.0.1	2021-11-26	[?]	CRAN	(R 4.4.0)
##	P mgcv	1.9-1	2023-12-21	[?]	CRAN	(R 4.4.0)
##	P mime	0.12	2021-09-28	[?]	CRAN	(R 4.4.0)
##	P miniUI	0.1.1.1	2018-05-18	[?]	RSPM	
##	P munsell	0.5.1	2024-04-01	[?]	CRAN	(R 4.4.0)
##	P mvtnorm	1.2-5	2024-05-21	[?]	RSPM	
##	P nlme	3.1-164	2023-11-27	[?]	CRAN	(R 4.4.0)
##	P pacman	0.5.1	2019-03-11	[?]	CRAN	(R 4.4.0)
##	P paletteer	* 1.6.0	2024-01-21	[?]	RSPM	
##	P pillar	1.9.0	2023-03-22	[?]	CRAN	(R 4.4.0)
##	P pkgbuild	1.4.4	2024-03-17	[?]	RSPM	
##	P pkgconfig	2.0.3	2019-09-22	[?]	CRAN	(R 4.4.0)
##	P pkgload	1.3.4	2024-01-16	[?]	RSPM	
##	P profvis	0.3.8	2023-05-02	[?]	RSPM	
##	P promises	1.3.0	2024-04-05	[?]	RSPM	
##	P purrr	* 1.0.2	2023-08-10	[?]	CRAN	(R 4.4.0)
##	P R6	2.5.1	2021-08-19	[?]	CRAN	(R 4.4.0)
##	P RColorBrewer	* 1.1-3	2022-04-03	[?]	CRAN	(R 4.4.0)
##	P Rcpp	1.0.12	2024-01-09	[?]	CRAN	(R 4.4.0)
##	P readr	* 2.1.5	2024-01-10	[?]	CRAN	(R 4.4.0)
##	P readxl	* 1.4.3	2023-07-06	[?]	CRAN	(R 4.4.0)
##	P rematch2	2.1.2	2020-05-01	[?]	CRAN	(R 4.4.0)
##	P remotes	2.5.0	2024-03-17	[?]	CRAN	(R 4.4.0)
##	P renv	1.0.7	2024-04-11	[1]	CRAN	(R 4.4.0)
##	P rlang	1.1.3	2024-01-10	[?]	CRAN	(R 4.4.0)
##	P rmarkdown	2.26	2024-03-05	[?]	CRAN	(R 4.4.0)
##	P rprojroot	2.0.4	2023-11-05	[?]	CRAN	(R 4.4.0)
##	P rstatix	0.7.2	2023-02-01	[?]	RSPM	
##	P rstudioapi	0.16.0	2024-03-24	[?]	CRAN	(R 4.4.0)
##	P scales	1.3.0	2023-11-28	[?]	CRAN	(R 4.4.0)
##	P sessioninfo	1.2.2	2021-12-06	[?]	RSPM	
##	P shiny	1.8.1.1	2024-04-02	[?]	RSPM	
##	P stringi	1.8.3	2023-12-11	[?]	CRAN	(R 4.4.0)
##	P stringr	* 1.5.1	2023-11-14	[?]	CRAN	(R 4.4.0)
##	P survival	* 3.6-4	2024-04-24	[?]	CRAN	(R 4.4.0)
##	P survminer	* 0.4.9	2021-03-09	[?]	RSPM	
##	P survMisc	0.5.6	2022-04-07	[?]	RSPM	
##	P tibble	* 3.2.1	2023-03-20	[?]	CRAN	(R 4.4.0)
##	P tidycmprsk	* 1.0.0	2023-10-30	[?]	CRAN	(R 4.4.0)
##	P tidyr	* 1.3.1	2024-01-24	[?]	CRAN	(R 4.4.0)
##	P tidyselect	1.2.1	2024-03-11	[?]	CRAN	(R 4.4.0)
##	P tidyverse	* 2.0.0	2023-02-22	[?]	CRAN	(R 4.4.0)
##	P timechange	0.3.0	2024-01-18	[?]	CRAN	(R 4.4.0)
##	P tzdb	0.4.0	2023-05-12	[?]	CRAN	(R 4.4.0)
##	P urlchecker	1.0.1	2021-11-30	[?]	RSPM	
##	P usethis	* 2.2.3	2024-02-19	[?]	RSPM	
##	P utf8	1.2.4	2023-10-22	[?]	CRAN	(R 4.4.0)
##	P vctrs	0.6.5	2023-12-01	[?]	CRAN	(R 4.4.0)
##	P vipor	0.4.7	2023-12-18	[?]	CRAN	(R 4.4.0)
##	P viridis	* 0.6.5	2024-01-29	[?]	RSPM	
##	P viridisLite	* 0.4.2	2023-05-02	[?]	CRAN	(R 4.4.0)
##	P vroom	1.6.5	2023-12-05	[?]	CRAN	(R 4.4.0)
##	P wesanderson	* 0.3.7	2023-10-31	[?]	RSPM	
##	P withr	3.0.0	2024-01-16	[?]	CRAN	(R 4.4.0)

```

## P xfun          0.43    2024-03-25 [?] CRAN (R 4.4.0)
## P xml2          1.3.6    2023-12-04 [?] CRAN (R 4.4.0)
## P xtable        1.8-4    2019-04-21 [?] RSPM
## P yaml          2.3.8    2023-12-11 [?] CRAN (R 4.4.0)
## P zoo           1.8-12   2023-04-13 [?] RSPM
##
## [1] /Users/zahavahrojer/Desktop/Cornell/Hendry Lab/pseud-epi-growth/renv/library/macos/R-4.4/x86_64
## [2] /Users/zahavahrojer/Library/Caches/org.R-project.R/R/renv/sandbox/macos/R-4.4/x86_64-apple-darw
##
## P -- Loaded and on-disk path mismatch.
##
## -----

```