**Master’s Thesis Supplementaries**

Department of Ecology, Evolution, and Organismal Biology

College of Science and Math

Kennesaw State University

|  |  |
| --- | --- |
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**I. Standard Operating Procedures**

**I.1.i. Chitosan Biostimulant Granule Phase 1 - Encapsulation SOP**

|  |  |  |  |
| --- | --- | --- | --- |
| **Title:** | CBG Phase 1 – Encapsulation SOP | **Department:** | EEOB |
| **Author:** | Zach Peagler | **Creation Date:** | Oct 24, 2023 |
| **PI:** | Mario Bretfeld | **Revision Date:** | April 17, 2025 |

**Purpose:**

This SOP details how to create CBGs up to the end of the curing process. It covers the preparation of chitosan and TPP solutions, inoculation, and dropwise addition.

**Equipment:**

Stirring hotplate (SC 358 benchtop)

1x 250 mL Erlenmeyer flask (SC 358 benchtop)

3x 1000 mL Erlenmeyer flask (SC 358 benchtop)

0-12x 50 mL Nalgene centrifuge tubes (make sure you use the Nalgene round bottom centrifuge tubes and not the conical centrifuge tubes, which will shatter in the centrifuge)

Autoclave (SC 365 or SL 3080)

Vortex (SC 358 benchtop)

Centrifuge (SC 361 [broken] or SL 3070)

PH meter (SC 358 benchtop)

Scale (SC 358 benchtop)

Dropwise Addition Setup (SC 358 benchtop)

200 – 1000 uL Micropipette (SC 358 benchtop)

Consumables:

5 g Chitosan

10 g Sodium tripolyphosphate

2L DI water

200-1000 uL micropipette tips

Aluminum foil

Autoclave tape

200 – 1000 uL Micropipette tips

**Protocol:**

1. Prepare DI water solution
2. Obtain two 1L Erlenmeyer flasks
3. Fill halfway with DI water
   1. These will be used later as (1) a chitosan solution and (2) a source of sterile DI water
4. Prepare TPP solution
5. Obtain a 1L Erlenmeyer flask
6. Fill to DOUBLE the chitosan solution volume with DI water, again leaving some breathing room on the volume

(we want a 2:1 ratio of TPP:chitosan)

1. e.g. if making 100 mL chitosan solution, make 200 mL TPP solution
2. Use the same concentration TPP as chitosan, 5%
3. 200 mL \* 0.05 = 10 g

(I know we’re going from mL to g here, but we’re assuming a density of 1g/1mL)

1. Mix thoroughly by swirling until all TPP granules have dissolved
2. Measure pH with pH probe and record pH value
3. Autoclave component solutions
4. Once DI water and TPP solutions are mixed, autoclave them
5. Add autoclave tape!
6. Sign the log book!
7. Autoclave time is based on the largest volume of fluid entering the autoclave. Because of this, it can be helpful to prepare the TPP solution in multiple aliquots to reduce the autoclave time, but this is up to your discretion.

(It’s weird to autoclave water and then make it into our solution, but chitosan cannot be autoclaved in solution that contains a strong acid, such as HCl, necessitating this approach. I recommend preparing DI water in bulk so you’re not constantly autoclaving small amounts of DI water.)

1. Prepare chitosan solution (STERILE)
2. Obtain one of the autoclaved DI water solutions and label it appropriately as the chitosan-HCl-microbe solution
3. Add an amount of HCl that is the stoichiometric equivalent to the amine groups on the amount of chitosan you are using
4. For 5% chitosan conc., this is 0.30M
5. Use M1V1=M2V2 to calculate amount of HCl
6. 0.30 M \* 100 mL = 12.08 M \* X mL
7. Weigh out enough chitosan to achieve a 5% concentration for your volume
8. 100 g x 0.05 = 5g
9. Add chitosan while stirring vigorously at 70 C
10. Stir for 30 minutes, or until all the chitosan has dissolved.
11. Solution should be viscous and translucent, mild amber in color
12. Measure pH with pH probe and record value.
13. Chitosan solution inoculation (STERILE)
14. Prior to inoculation, take 3mL samples in cuvette tubes of all chitosan solutions to be inoculated.
15. Obtain a bacterial stock solution from the refrigerator.
16. Do NOT add stock bacterial solution directly to chitosan solution.
17. Vortex stock solution and micropipette desired amount into 50 mL centrifuge tubes.
18. Use the appropriate bacterial calibration curve and the desired cfu/mL of the final product to determine the appropriate volume of bacterial stock to centrifuge.
19. Centrifuge the bacterial stock solution and decant off the supernatant
20. Use the F21 rotor at 10,000 g for 10 minutes.
21. The supernatant might be hazardous, or not, depending on the media. Double check and dispose of it properly.
22. Rinse gently with sterile DI water 3 times
23. Add 2mL sterile DI water to centrifuge tube and vortex
24. Pour into chitosan solution
25. Repeat previous step.
26. After inoculation, vortex and take 3mL samples in cuvette tubes.
27. Use the spectrophotometer in SC 350 to determine the OD600 of the chitosan-microbial solution, making sure to use the respective blanks made in 5.a.
28. Dropwise addition (STERILE)
29. Prior to performing dropwise addition, take a 3mL sample of all TPP solutions in separate cuvette tubes.
30. (This will be used as a blank later.)
31. Hook up the DA setup.
32. Perform dropwise addition.
33. Once no more chitosan solution remains, allow beads to cure in TPP for 30 minutes
34. Post-Cure
35. After 30 minutes, pour beads into Buchner funnel over 1L Erlenmeyer flask.
36. Use the spectrophotometer in SC 350 to determine the OD600 of the post-DA TPP solution, making sure to use the respective blanks made in 6.a.

**I.1.ii. Chitosan Biostimulant Granule Phase 2 - Desiccation SOP**

|  |  |  |  |
| --- | --- | --- | --- |
| **Title:** | CBG Phase 2 – Desiccation SOP | **Department:** | EEOB |
| **Author:** | Zach Peagler | **Creation Date:** | Oct 28, 2023 |
| **PI:** | Mario Bretfeld | **Revision Date:** | April 17, 2025 |

**Purpose:**

Desiccation is necessary for inducing stasis in the bacteria and reducing the weight of the beads for transport. The following protocol is to be used on beads that have already been through Phase 1 and are fully cured.

**Equipment:**

PH meter

Scale

Desiccation chamber

Spectrophotometer

Cuvette tubes

**Consumables:**

Petri dishes

Parafilm

1.5 mL microcentrifuge tubes

**Protocol:**

1. Data Collection
2. Once beads have cured for 30 minutes, transfer them to petri dish.
3. Take a 3mL sample of the post-curing TPP solution and collect the OD600 with the spectrophotometer in SC 350 using the prepared blank from Phase 1.
4. Collect weight by taring scale to empty petri dish, then place petri dish with CBGs onto scale.
5. Collect images by placing petri dish wish CBGs on wooden camera apparatus next to the drying oven.

i. Make sure the overhead light is on and image using the lab camera (Nikon something or other)

1. Collect pH by taking 5 beads, rehydrating them in 1mL DI water in a 1.5mL microcentrifuge tube, and reading the pH with a pH test strip.
2. Desiccation
3. Place beads in vacuum desiccation chamber set to 30C, pulling pressure to 0.5 atm.
4. Leave for 24H.
5. Check desiccation every 24H until fully desiccated, performing tasks 1.c-1.e at every 24H interval.

**I.1.iii. Chitosan Biostimulant Granule Phase 2 - Desiccation SOP**

|  |  |  |  |
| --- | --- | --- | --- |
| **Title:** | CBG Phase 3 - Rehydration SOP | **Department:** | EEOB |
| **Author:** | Zach Peagler | **Creation Date:** | Oct 24, 2023 |
| **PI:** | Mario Bretfeld | **Revision Date:** | April 17, 2025 |

**Purpose**

Collecting data on how CBGs change upon rehydration is crucial to understanding how they degrade. This procedure details data collection tasks to be performed during CBG Phase 3 – Rehydration.

**Reagents**

Sterile DI Water . . . . . . . . . . . . . . . . . . . . . . . . . . . 10 mL

Desiccated CBGs . . . . . . . . . . . . . . . . . . . . . . . . . . .  15 granules

**Equipment**

Sterile 1.5 mL microcentrifuge tubes

Scale

Spectrophotometer

Tweezers

Flame

PH test strips

**Protocol**

1. Once CBGs are fully desiccated, they are ready to be rehydrated.
2. Fill 3x 1.5 mL microcentrifuge tubes with 1 mL of sterile DI water each.
3. Weigh out 5 CBGs.
4. Using a pair of flame-sterilized tweezers, place 5 CBGs in each microcentrifuge tube from the previous step.
5. Take the following measurements at the following time points (5 minutes, 30 minutes, 1 hour, 2 hours, 6 hours, 24 hours, 48 hours, 72 hours)
6. PH
7. Use pH test strips. (Volume is too small to use our pH meter)
8. OD600
9. Use spectrophotometer in SC 350.
10. Prepare a 3mL sterile DI water reference cuvette tube.
11. Pipette 0.300 mL CBG rehydration fluid into 2.7mL of sterile DI water in a cuvette tube.
12. Ref the spectrophotometer on the blank, then read the sample.
13. The OD of the sample is 10x the reading on the machine.
14. Pipette 0.300 mL sterile DI water into CBG microcentrifuge tube to return volume to 1 mL.
15. Following rehydration, remove CBGs from rehydration fluid and weigh.

**I.2. Alginate Microbial Encapsulation SOP**

|  |  |  |  |
| --- | --- | --- | --- |
| **Title:** | Alginate Microbial Encapsulation SOP | **Department:** | EEOB |
| **Author:** | Zach Peagler | **Creation Date:** | Oct 24, 2023 |
| **PI:** | Mario Bretfeld | **Revision Date:** | Sept 30, 2024 |

NOTE: This SOP is meant to be used with the accompanying Alginate Microbial Encapsulation data sheet. Do NOT start the SOP until you have the data sheet ready to go for the trial. - [Link to data sheet.](https://kennesawedu-my.sharepoint.com/:x:/g/personal/zpeagler_students_kennesaw_edu/ESUtnoeackVCsnnu2zOv8pAB-XAzJyRf4QZm52BqEA-jBw?e=UI7fo7&nav=MTVfezAwMDAwMDAwLTAwMDEtMDAwMC0wMDAwLTAwMDAwMDAwMDAwMH0)

**Purpose:**

This SOP details how to create alginate encapsulated microbial granules. It covers the preparation of polymer and cross-linker solutions, inoculation, dropwise addition, curing, desiccating, rehydration, and storage. Following bead creation, beads are subject to microbial viability ([SOP here](https://kennesawedu-my.sharepoint.com/:w:/g/personal/zpeagler_students_kennesaw_edu/EQZ0GKG-KnFPkA5zcVZITxsBBgd5fVnwEmm1vpm9xk-gYA?e=YnX8tA)) and germination assays ([SOP here](https://kennesawedu-my.sharepoint.com/:w:/g/personal/zpeagler_students_kennesaw_edu/EYoqR7fdgZJJqetFa3DmfAwBPnBa_C1d-WwmbzlGyzbJMw?e=Gy5WlC)).

**Equipment:**

|  |  |  |
| --- | --- | --- |
| **Name** | **Location** | **Amount** |
| Stirring hotplate | SC 358 Benchtop B | 1 |
| Vortex | SC 358 Benchtop A | 1 |
| Autoclave | SC 365 or SL 3080 | 1 |
| Centrifuge | SC 361, SL 3070, SL 5070 | 1 |
| PH meter | SC 358 Benchtop B | 1 |
| Scale | SC 358 Benchtop A | 1 |
| Microbial Encapsulation Testbench (MET) | SC 358 Benchtop C | 1 |
| 200-1000 uL Micropipette | SC 358 Benchtop A | 1 |
| 250 mL Erlenmeyer flask | SC 358 Benchtop B | 1 |
| 1000 mL Erlenmeyer flask | SC 358 Benchtop B | 3 |
| Cuvettes | SC 358 Shelf B3 | 2-12 |
| Spectrophotometer | SC 350 | 1 |
| Buchner funnel | SC 358 Shelf B1 | 1 |
| Spatula | SC 358 Benchtop A | 1 |
| Nalgene round bottom centrifuge tubes | SC 358 Benchtop B | 2-12 |
| Bunsen burner | SC 358 Benchtop A | 1 |

**Consumables:**

|  |  |  |
| --- | --- | --- |
| Name | Location | Amount |
| 200-1000 uL micropipette tips | SC 358 Benchtop A | ~10 |
| Aluminum foil | SC 358 Benchtop B | A bit |
| Autoclave tape | Autoclave room | A bit |
| Conical bottom centrifuge tubes | SC 358 Bench B (under the bench) | 2-12 |

**Reagents:**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Name | CAS Number | Location | Amount | Hazards |
| Sodium alginate | 9005-38-3 | SC 358 Benchtop A | 5 g | None |
| CaCl2 | 10043-52-4 | SC 358 Benchtop B | 10 g | Eye irritation |
| DI Water | 7732-18-5 | Autoclave room | 3 L | None |
| Ethanol (70%) | 64-17-5 | SC 358 Benchtop A&B | ~50 mL | Flammable, eye irritation |

**Protocol:**

1. Prepare overnight cultures
2. For the selected microbes, begin culturing them 72 hours (3 days) in advance if harvesting in stationary phase, and 48 hours (2 days) in advance if harvesting in log phase.
3. New cultures must be made for each run of this protocol.
4. Make enough for three batches of beads.
5. Prepare DI water solution
6. Obtain one 1L Erlenmeyer flask
7. Add 500 mL DI water and label accordingly
8. Prepare alginate solution
9. Obtain one 500 mL Erlenmeyer flask
10. Add 250 mL DI water
11. Add 5 g sodium alginate and label accordingly (2% alginate)
12. Add a stir bar and stir on stirring hotplate for 5 minutes
13. Remove stir bar with magnetic stir bar removal tool
14. Prepare CaCl2 solution
15. Obtain one 1L Erlenmeyer flask
16. Add 500 mL DI water
17. Add 10 g CaCl2 and label accordingly (2% CaCl2)
18. Autoclave component solutions
19. Take your DI water, alginate, and CaCl2 solutions and autoclave them
20. Add autoclave tape!
21. Sign the log book!
22. Autoclave time is based on the largest volume of fluid entering the autoclave. Here, our largest volume is 500 mL, which corresponds to the L40 cycle on the autoclave.
23. Once the solutions are out of the autoclave and cooled to under 40C, measure and record the pH of both the alginate and CaCl2 solutions.
24. To a fresh cuvette, add 3mL autoclaved alginate solution.
25. To a second fresh cuvette, add 3mL autoclaved CaCl2 solution
26. Inoculation
27. Retrieve overnight cultures from shaking incubator.
28. Add 3 mL uninoculated growth media to a cuvette.
29. To a second cuvette, add 3 mL overnight culture.
30. Use the spectrophotometer to get the OD600 of the overnight culture.
31. Blank the spectrophotometer with the blank made in 6.b, then read the overnight culture.
32. Use the [microbial dilution calculator](https://kennesawedu-my.sharepoint.com/:x:/g/personal/zpeagler_students_kennesaw_edu/EQtV0Ug1khRNjt0WaPpA944BMEJQs5NaaFw89xSqEvSQfQ?e=P4BXea) to calculate the necessary amount of overnight culture to achieve a bacterial concentration of 1x108 cfu/mL.
33. Do NOT add bacterial solution directly to alginate solution.
34. Vortex overnight culture and micropipette desired amount into 50 mL round bottom centrifuge tubes. The round bottom is important. Conical tubes will shatter.
35. Centrifuge the overnight culture
36. Use the SS34 rotor at 10,000 g for 10 minutes.
37. The supernatant might be hazardous depending on the media. Double check and dispose of it properly.
38. Decant the supernatant.
39. Rinse gently with sterile DI water 3 times
40. Add 2mL sterile DI water to centrifuge tube and vortex.
41. Dump contents of centrifuge tube into autoclaved alginate solution.
42. After inoculation, vortex and take 3mL samples in cuvette tubes.
43. Use the spectrophotometer in SC 350 to determine the OD600 of the inoculated alginate solution, using the blank made in 5.d.
44. Dropwise addition
45. Record pH of both alginate and CaCl2 solutions before dropwise addition.
46. Sterilize and hook up the MET.
47. Perform dropwise addition, shaking at 60 rpm, with the peristaltic pump a quarter turn from the slowest setting.
48. Once alginate is completely dispensed, allow beads to cure for 30 minutes.
49. Post-Cure
50. Obtain and sterilize a Buchner funnel, 1L Erlenmeyer flask, and spatula.
51. Once cure time has elapsed, pour beads into Buchner funnel over 1L Erlenmeyer flask, using the spatula to scrape any recalcitrant beads into the funnel.
52. Transfer beads to a sterile petri dish and weigh.
53. Use the spectrophotometer to determine the OD600 of the post-DA CaCl2 solution, and record its pH.
54. Separate out about half of the beads and store them in sterile conical bottom centrifuge tubes – separating them into two tubes, one for 24C and one for 4C storage.
55. Desiccation
56. Take the remaining half of the beads and put them on uncovered petri dishes and place them into the sterile desiccation chamber (SDC).
57. Plug the SDC in, pull a vacuum down to 0.5 atm, and leave to dry for 24 hours. Check desiccation progress every 24 hours.
    1. It would be awesome if this happened automatically, where the machine takes pictures over time and we could get a sick timelapse of the drying process. For a future version of the SDC)
58. Once beads have been created, add them to the [Microbial Encapsulation Assay Schedule](https://kennesawedu-my.sharepoint.com/:x:/g/personal/zpeagler_students_kennesaw_edu/ETLUVn1pVPlAmNAG2oC31jMBl8itCmgj-KOTf4ubs-E9Ow?e=Qgl0LZ) and perform viability and germination assays on the dates specified therein.

**I.3. Bead Breakdown SOP**

|  |  |  |  |
| --- | --- | --- | --- |
| **Title:** | Bead Breakdown SOP | **Department:** | EEOB |
| **Author:** | Zach Peagler | **Creation Date:** | Oct 2, 2024 |
| **PI:** | Mario Bretfeld | **Revision Date:** | Jan 29, 2025 |

**Note:** This SOP is meant to be used with the accompanying bead breakdown data sheet.  Do not start until you have the data sheet ready. [Link to data sheet](https://kennesawedu-my.sharepoint.com/:x:/g/personal/zpeagler_students_kennesaw_edu/EfPMAHdjHr5BltfFLecHOBsBmHIYbnQB4JZUrcmi8iwghw?e=FwjkFk).

**Purpose:**

The purpose of this SOP is to determine the rate at which microbial beads break down in soil.

**Equipment:**

|  |  |  |
| --- | --- | --- |
| **Name** | **Location** | **Amount** |
| Incubator | SC 358 | 1 |
| Camera + jig | SC 358 | 1 |
| Scale | SC 358 Bench A | 1 |
| Tweezers | SC 358 Bench A | 1 |
| Scissors | SC 358 Bench B | 1 |
| Trowel | SC 358 | 1 |
| Bucket | SC 358 | 1 |
| Graduated Cylinder | SC 358 Benchtop B3 | 1 |

**Consumables:**

|  |  |  |
| --- | --- | --- |
| **Name** | **Location** | **Amount** |
| Petri dishes | SC 358 | 200 |
| Soil | KSU Field Station | 1 kg |
| Microbial beads | SC 358 Benchtop C | 2000 |
| Parafilm | SC 358 Shelf B1 | 200 strips |
| Filter Paper | SC 358 Shelf A2 | 200 rounds |

**Reagents:**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Name** | **CAS Number** | **Location** | **Amount** | **Hazards** |
| Ethanol (70%) | 64-17-5 | SC 358 Benchtop A&B | ~10 mL | Flammable, eye irritation |
| DI Water | 7732-18-5 | SC 358 Benchtop A & B | 1 L | None |

**Protocol:**

1. Collect a trowel and bucket then go out to the KSU field station and collect a kilogram of soil, then return to the lab.
2. Mix the soil thoroughly
3. Collect petri dishes and cut the same number of parafilm strips.
4. Place 10 grams of soil in each petri dish, spreading it thinly and evenly.
5. Add 10 mL  DI water to the petri dish.
6. Collect beads from storage/fresh from the Buchner funnel. Refer to either the [Alginate Encapsulation SOP](https://kennesawedu-my.sharepoint.com/:w:/g/personal/zpeagler_students_kennesaw_edu/Ea8UVwv-edBEk5Scay-AxYMBNPufOSkQq2Qn8qJ8Jd6zsQ?e=U39SoL) or the [CBG Phase 1 SOP](https://kennesawedu-my.sharepoint.com/:w:/g/personal/zpeagler_students_kennesaw_edu/EXKVPrcAvW1JuT6pNXgxrhUByFveKgY6YB994sONuB_y4A?e=jMVamG) for bead creation.
7. Use tweezers to place 10 beads on a paper towel and pat them dry, then transfer them to a weigh boat and collect the weight, then place the 10 beads on a soil filled petri dish.
8. Close the petri dish and seal with parafilm.
9. Label the petri dish with bead id, date, and a unique petri dish id.
10. Repeat for every petri dish.
11. Place plates in the incubator, making sure it’s set to 30C.
12. Once a week for the next 10 weeks, do the following:
13. Collect 10 samples at random from the incubator.
14. Carefully remove the 10 beads from the petri dish and rinse them gently with DI water.
15. Transfer the 10 beads to a paper towel and pat dry.
16. Place the 10 beads on a weigh boat and weigh.
17. Transfer the beads to a clean petri dish and take pictures of them on the camera jig.
18. Once finished with measurements, discard used petri dishes and measured beads.

**II. 3D Prints**

**II.A. Lab Tools**

**II.A.1. Centrifuge Tube (50 mL) Rack**

A blue and white object with holes

AI-generated content may be incorrect.

**II.A.2. Microcentrifuge Tube Rack**

A blue rectangular object with holes

AI-generated content may be incorrect.

**II.A.3. Sterile Desiccation Chamber Electrical Housing**

A white object with holes

AI-generated content may be incorrect.

**II.A.4. Microscope Slide Rack**

A rectangular object with a white surface

AI-generated content may be incorrect.

**II.A.5. Sony E-mount to Leica Microscope Adapter**

**A white object with a white background

AI-generated content may be incorrect.**

**II.B. Greenhouse Tools**

**II.B.1. PhotosynQ MultispeQ V2.0 Tripod Adapter**

A white object with a hole

AI-generated content may be incorrect.

**III. Code**

**III.A. Shared Code**

**III.A.1. Load Packages**

require(lme4)

require(multcomp)

require(lmerTest)

require(MuMIn)

require(rstatix)

require(MASS)

require(pwr)

require(car)

require(tidyverse)

require(showtext)

require(scico)

require(ggpubr)

require(devtools)

install\_github("zachpeagler/ztils", upgrade = "never")

require(ztils)

**III.A.2. Graphical Setup**

a\_palette <- "oslo"

# add fonts

font\_add\_google("Open Sans", family = "open")

font\_add\_google("Montserrat", family = "mont")

# initialize imported fonts

showtext\_auto()

# font sizes

## title

title\_size = 20

## subtitle

subtitle\_size = 16

## caption

caption\_size = 14

## text

text\_size = 12

# custom shapes

four\_shapes = c(15,16,17,23)

**III.B. Tomato Inoculant Location Code**

**III.B.1 Setup**

germdate23 <- "2023-04-14"

treatment\_order23 <- c("Control",

"Soil",

"Foliar",

"Soil+Foliar")

d23\_fg\_file <- "https://raw.githubusercontent.com/zachpeagler/Thesis/refs/heads/main/data/TIP/2023/TIP23\_Fruit\_Greenhouse.csv"

d23\_fl\_file <- "https://raw.githubusercontent.com/zachpeagler/Thesis/refs/heads/main/data/TIP/2023/TIP23\_Fruit\_Lab.csv"

d23\_m\_file <- "https://raw.githubusercontent.com/zachpeagler/Thesis/refs/heads/main/data/TIP/2023/TIP23\_Multispeq.csv"

d23\_li\_file <- "https://raw.githubusercontent.com/zachpeagler/Thesis/refs/heads/main/data/TIP/2023/TIP23\_LI600.csv"

d23\_st\_file <- "https://raw.githubusercontent.com/zachpeagler/Thesis/refs/heads/main/data/TIP/2023/TIP23\_StomatalDensity.csv"

**III.B.2. Clean Data**

d23\_fg <- read.csv(d23\_fg\_file) %>%

mutate(

Row\_num = case\_when(

Row=="A"~1,

Row=="B"~2,

Row=="C"~3,

Row=="D"~4),

Treatment = case\_when(

Row=="A"~"Control",

Row=="B"~"Soil",

Row=="C"~"Foliar",

Row=="D"~"Soil+Foliar",

TRUE ~ NA),

soil = case\_when(

Row=="A"~FALSE,

Row=="B"~TRUE,

Row=="C"~FALSE,

Row=="D"~TRUE

),

foliar = case\_when(

Row=="A"~FALSE,

Row=="B"~FALSE,

Row=="C"~TRUE,

Row=="D"~TRUE

),

BER = case\_when(

BER==0~"FALSE",

BER==1~"TRUE"),

BER = as.logical(BER),

Treatment = factor(Treatment, levels = treatment\_order23),

fruit = 1,

Plant = as.factor(paste0(Row, Pot)),

Pot = as.factor(Pot),

Cluster = as.factor(Cluster),

Date = as.Date(Date, "%m/%d/%Y"),

)

colnames(d23\_fg) <- tolower(colnames(d23\_fg))

d23\_fg <- d23\_fg %>%

rename(mass = weight)

### Summary Fruit Greenhouse

### Sum the fruit, BER, and mass by treatment group, then get ber probability.

d23\_fg\_summary <- d23\_fg %>%

group\_by(treatment, plant) %>%

summarise\_at(vars(fruit, ber, mass),

list(sum=sum)) %>%

mutate(pber = round(ber\_sum/fruit\_sum, 4))

d23\_fg\_summary2 <- d23\_fg %>%

group\_by(treatment) %>%

summarise\_at(vars(fruit, ber, mass),

list(sum=sum)) %>%

mutate(pber = round(ber\_sum/fruit\_sum, 4))

### Means Fruit Greenhouse

### Make summary dataframe with mean and sd of mass by plant.

d23\_fg\_means <- d23\_fg %>%

group\_by(treatment, plant) %>%

summarise\_at(vars(mass), list(mean=mean, sd=sd))

d23\_fg\_means2 <- d23\_fg %>%

group\_by(treatment) %>%

summarise\_at(vars(mass), list(mean=mean, sd=sd))

## Fruit Lab (no BER; marketable fruit only)

d23\_fl <- read.csv(d23\_fl\_file) %>%

rename(pot = plant) %>%

mutate(

row\_num = case\_when(

row=="A"~1,

row=="B"~2,

row=="C"~3,

row=="D"~4),

Treatment = case\_when(row=="A"~"Control",

row=="B"~"Soil",

row=="C"~"Foliar",

row=="D"~"Soil+Foliar",

TRUE ~ NA),

soil = case\_when(

row=="A"~FALSE,

row=="B"~TRUE,

row=="C"~FALSE,

row=="D"~TRUE

),

foliar = case\_when(

row=="A"~FALSE,

row=="B"~FALSE,

row=="C"~TRUE,

row=="D"~TRUE

),

Treatment = factor(Treatment, levels = treatment\_order23),

fruit = 1,

ripeness = abs(1 - round(penetrometer/max(na.omit(penetrometer)), 2)),

logit\_sugar = logit(sugar\_avg/100),

Sugar\_grams = (sugar\_avg/100)\*mass,

row = as.factor(row),

pot = as.factor(pot),

cluster = as.factor(cluster),

plant = as.factor(paste0(row, pot)),

date = as.Date(date, "%m/%d/%Y")

)

colnames(d23\_fl) <- tolower(colnames(d23\_fl))

### Summary Fruit Lab

### Sum the fruit, sugar, and mass by treatment group.

d23\_fl\_summary <- d23\_fl %>%

group\_by(treatment, plant) %>%

summarise\_at(vars(fruit, sugar\_grams, mass),

list(sum=sum))

d23\_fl\_summary2 <- d23\_fl %>%

group\_by(treatment) %>%

summarise\_at(vars(fruit, sugar\_grams, mass),

list(sum=sum))

### Means Fruit Lab

### Make summary dataframe with mean and sd of mass by plant.

d23\_fl\_means <- d23\_fl %>%

group\_by(treatment, plant) %>%

summarise\_at(vars(mass, sugar\_avg, logit\_sugar, sugar\_grams), list(mean=mean, sd=sd))

d23\_fl\_means2 <- d23\_fl %>%

group\_by(treatment) %>%

summarise\_at(vars(mass, sugar\_avg, logit\_sugar, sugar\_grams), list(mean=mean, sd=sd))

## Li-600

d23\_li <- read.csv(d23\_li\_file, stringsAsFactors = T) %>%

mutate(Treatment = case\_when(

Row==1~"Control",

Row==2~"Soil",

Row==3~"Foliar",

Row==4~"Soil+Foliar",

TRUE~NA),

Row = case\_when(

Row==1~"A",

Row==2~"B",

Row==3~"C",

Row==4~"D"

),

Soil = case\_when(

Row=="A"~FALSE,

Row=="B"~TRUE,

Row=="C"~FALSE,

Row=="D"~TRUE

),

Foliar = case\_when(

Row=="A"~FALSE,

Row=="B"~FALSE,

Row=="C"~TRUE,

Row=="D"~TRUE

)) %>%

filter(leak\_pct<10 & gsw > 0 & gsw < 5) %>%

mutate(Date = parse\_date\_time(Date, orders = "mdy"),

Date = as.Date(Date),

Time = parse\_date\_time(Time, orders = "T"),

DaysFromGermination = as.numeric(round(difftime(Date, germdate23, units = c("days")), 0)),

Plant = as.factor(paste0(Row, Pot)),

Treatment = factor(Treatment,

levels = treatment\_order23),

LogitPhiPS2 = logit(PhiPS2, FALSE)

) %>%

group\_by(DaysFromGermination) %>%

mutate(MinutesFromStart = as.numeric(round(difftime(Time, min(Time), units = "mins"), 2))) %>%

ungroup() %>%

mutate(

Time = as.factor(format(Time, "%H:%M:%S"))

)

## multispeq

d23\_m <- read.csv(d23\_m\_file) %>%

rename(Pot = Pot.ID) %>%

mutate(Treatment = case\_when(

Row=="A"~"Control",

Row=="B"~"Soil",

Row=="C"~"Foliar",

Row=="D"~"Soil+Foliar",

TRUE~NA),

Soil = case\_when(

Row=="A"~FALSE,

Row=="B"~TRUE,

Row=="C"~FALSE,

Row=="D"~TRUE

),

Foliar = case\_when(

Row=="A"~FALSE,

Row=="B"~FALSE,

Row=="C"~TRUE,

Row=="D"~TRUE

)) %>%

mutate(Row\_num = case\_when(

Row=="A"~1,

Row=="B"~2,

Row=="C"~3,

Row=="D"~4),

Plant = as.factor(paste0(Row, Pot)),

Pot = as.factor(Pot),

Row = as.factor(Row),

Device.ID = as.factor(Device.ID),

Date = as.Date(time, "%m/%d/%Y"),

DaysFromGermination = as.numeric(round(difftime(Date, germdate23, units = c("days")), 0)),

datetime = parse\_date\_time(time, "%m/%d/%Y %H:%M"),

Time = format(datetime, "%H:%M:%S"),

Time = parse\_date\_time(Time, orders = "T"),

LogitPhiPS2 = logit(Phi2, FALSE),

) %>%

group\_by(DaysFromGermination) %>%

mutate(MinutesFromStart = as.numeric(round(difftime(Time, min(Time), units = "mins"), 2))) %>%

ungroup() %>%

mutate(

Time = as.factor(format(Time, "%H:%M:%S"))

)

## stomatal density

d23\_st <- read.csv(d23\_st\_file) %>%

rename(pot = plant) %>%

mutate(Treatment = case\_when(

row=="A"~"Control",

row=="B"~"Soil",

row=="C"~"Foliar",

row=="D"~"Soil+Foliar",

TRUE~NA),

Treatment = as.factor(Treatment),

Treatment = factor(Treatment, levels = treatment\_order23),

soil = case\_when(

row=="A"~FALSE,

row=="B"~TRUE,

row=="C"~FALSE,

row=="D"~TRUE

),

foliar = case\_when(

row=="A"~FALSE,

row=="B"~FALSE,

row=="C"~TRUE,

row=="D"~TRUE

),

plant = as.factor(paste0(row, pot)),

surface = as.factor(surface)

)

colnames(d23\_st) <- tolower(colnames(d23\_st))

**III.B.3. Exploratory Analysis**

## mass

### PDF, CDF, and KS test

multipdf\_plot(d23\_fg$mass, 100, "all", a\_palette, "mass")

ggsave("23\_mass\_PDF.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_mass\_PDF.png", path = "C:/Github/Thesis/figures/TIP23")

multicdf\_plot(d23\_fg$mass, 100, "all", a\_palette, "mass")

ggsave("23\_mass\_CDF.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_mass\_CDF.png", path = "C:/Github/Thesis/figures/TIP23")

multiks\_cont(d23\_fg$mass, "all")

## check for heteroscedasticity

### in mass as a function of treatment

leveneTest(d23\_fg$mass~d23\_fg$treatment)

### in mass as a function of ber

leveneTest(d23\_fg$mass~d23\_fg$ber)

## box plot of mass by Treatment

ggplot(data = d23\_fg, aes(x= treatment, y = mass,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

## box plot by Treatment colored by BER

ggplot(data = d23\_fg, aes(x= treatment, y = mass,

fill=ber, color=ber)) +

geom\_boxplot(alpha=.5, width=0.6, outliers = TRUE)+

scale\_fill\_manual(values=two\_colors)+

scale\_color\_manual(values=two\_colors)+

guides(fill=guide\_legend(title=str\_wrap("Blossom-End Rot", 10)),

color=guide\_legend(title=str\_wrap("Blossom-End Rot", 10))

)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="right",

legend.title = element\_text(size=subtitle\_size, family="mont",

face = "bold", lineheight=0.5),

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_mass\_by\_ber.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_mass\_by\_ber.png", path = "C:/Github/Thesis/figures/TIP23")

## summarized fruit graphs

### box plot of mean mass of ALL tomatoes by treatment

ggplot(data = d23\_fg\_means, aes(x= treatment, y = mean,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

### box plot of mean mass by treatment

ggplot(data = d23\_fl\_means, aes(x= treatment, y = mass\_mean,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_mass\_mean.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_mass\_mean.png", path = "C:/Github/Thesis/figures/TIP23")

### box plot of mean sugar by treatment

ggplot(data = d23\_fl\_means, aes(x= treatment, y = sugar\_grams\_mean,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_sugar\_mean.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_sugar\_mean.png", path = "C:/Github/Thesis/figures/TIP23")

### box plot of percent BER by treatment

ggplot(data = d23\_fg\_summary, aes(x= treatment, y = pber,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("pBER")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_ber.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_ber.png", path = "C:/Github/Thesis/figures/TIP23")

### box plot of total fruit mass by treatment

ggplot(data = d23\_fg\_summary, aes(x= treatment, y = mass\_sum,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

### box plot of percent BER by treatment

ggplot(data = d23\_fg\_summary, aes(x= treatment, y = fruit\_sum,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Fruit")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_fruit\_count.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_fruit\_count.png", path = "C:/Github/Thesis/figures/TIP23")

## sugar

### PDF, CDF, and KS test

multipdf\_plot(d23\_fl$sugar\_grams, 100, "all", a\_palette, "sugar")

ggsave("23\_sugar\_PDF.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_sugar\_PDF.png", path = "C:/Github/Thesis/figures/TIP23")

multicdf\_plot(d23\_fl$sugar\_grams, 100, "all", a\_palette, "sugar")

ggsave("23\_sugar\_CDF.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_sugar\_CDF.png", path = "C:/Github/Thesis/figures/TIP23")

multiks\_cont(d23\_fl$sugar\_grams, "all")

## check for heteroscedasticity

### in sugar as a function of treatment

leveneTest(d23\_fl$sugar\_grams~d23\_fl$treatment)

###### NOT homoscedastic! Unequal variances!

## box plot of sugar % by Treatment

ggplot(data = d23\_fl, aes(x= treatment, y = sugar\_avg,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Sugar %")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

## box plot of sugar by Treatment

ggplot(data = d23\_fl, aes(x= treatment, y = sugar\_grams,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Sugar (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

## scatter plot of sugar by mass

ggplot(data = d23\_fl, aes(x= mass, y = sugar\_avg,

fill=treatment, color=treatment)) +

geom\_jitter( width=0, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Sugar %")+

xlab("Mass (g)")+

theme\_bw()+

theme(

legend.position="right",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_sugar\_by\_mass.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_sugar\_by\_mass.png", path = "C:/Github/Thesis/figures/TIP23")

## box plot of mean sugar by Treatment

ggplot(data = d23\_fl\_means, aes(x= treatment, y = sugar\_grams\_mean,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Sugar %")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_sugar\_grams\_mean.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_sugar\_grams\_mean.png", path = "C:/Github/Thesis/figures/TIP23")

## box plot of total sugar by Treatment

ggplot(data = d23\_fl\_summary, aes(x= treatment, y = sugar\_grams\_sum,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Sugar (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

# li-600

# PDF, CDF, and KS test

multipdf\_plot(filter(d23\_li, gsw >0)$gsw, 100, "all", a\_palette, "gsw")

ggsave("23\_gsw\_PDF.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_gsw\_PDF.png", path = "C:/Github/Thesis/figures/TIP23")

multicdf\_plot(filter(d23\_li, gsw >0)$gsw, 100, "all", a\_palette, "gsw")

ggsave("23\_gsw\_CDF.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_gsw\_CDF.png", path = "C:/Github/Thesis/figures/TIP23")

multiks\_cont(filter(d23\_li, gsw >0)$gsw, "all")

# check for heteroscedasticity

## in gsw as a function of treatment

leveneTest(d23\_li$gsw~d23\_li$treatment)

## in gsw as a function of time from start

leveneTest(d23\_li$gsw~d23\_li$minutesfromstart)

## box plot by Treatment

ggplot(data = d23\_li, aes(x= treatment, y = gsw, fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.3, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Stomatal Conductance (mol m-2 s-1)")+

# annotate("text", x=1:4, y=3, label = c("a", "b", "b", "b"), size=5, family="open")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=16, family="mont", lineheight=0.8),

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=20, family="open", face="bold")

)

# GSW by date across relative humidity

ggplot(data = d23\_li, aes(x=daysfromgermination, y = gsw,

color = rh\_s)) +

geom\_jitter(width=1, size=2, alpha=1)+

scale\_color\_scico(begin=0.9, end=0, palette=a\_palette)+

# scale\_x\_discrete(guide=guide\_axis(check.overlap=TRUE))+

theme\_bw()+

ylab("Stomatal Conductance (mol m-2 s-1)")+

xlab("Days From Germination")+

labs(

title=NULL,

subtitle = NULL

) +

guides(color=guide\_colorbar(title=str\_wrap("Relative Humidity", 10)))+

theme(

text = element\_text(size=16, family="mont"),

legend.title = element\_text(size=16, family="mont", face="bold"),

legend.text = element\_text(size=14, family="mont"),

legend.position="right",

legend.title.position = "top",

legend.key.height = unit(.4, "cm"),

legend.background = element\_rect(color=two\_colors[2], fill=NA,

linewidth=.5, linetype = 2),

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=20, family="open", face="bold", lineheight = .8)

)

ggsave("23\_gsw\_by\_date.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_gsw\_by\_date.png", path = "C:/Github/Thesis/figures/TIP23")

# GSW by time across relative humidity

ggplot(data = d23\_li, aes(x=time, y = gsw,

color = date,

shape = treatment)) +

geom\_jitter(width=1, size=2, alpha=1)+

scale\_color\_scico(begin=0.9, end=0, palette=a\_palette)+

scale\_shape\_manual(values=four\_shapes)+

scale\_x\_discrete(guide=guide\_axis(check.overlap=TRUE))+

theme\_bw()+

ylab("Stomatal Conductance (mol m-2 s-1)")+

xlab("Time")+

labs(

title=NULL,

subtitle = NULL

) +

guides(color=guide\_legend(title="Date"))+

theme(

text = element\_text(size=16, family="mont"),

legend.title = element\_text(size=16, family="mont", face="bold"),

legend.text = element\_text(size=14, family="mont"),

legend.position="right",

legend.title.position = "top",

legend.key.height = unit(.3, "cm"),

legend.background = element\_rect(color=two\_colors[2], fill=NA,

linewidth=.5, linetype = 2),

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=20, family="open", face="bold", lineheight = .5)

)

ggsave("23\_gsw\_by\_time.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_gsw\_by\_time.png", path = "C:/Github/Thesis/figures/TIP23")

# gsw by relative humidity across treatment groups

ggplot(data = d23\_li, aes(x=rh\_s, y = gsw,

shape=treatment, fill=treatment, color = treatment)) +

geom\_jitter(width=1, size=2, alpha=1)+

scale\_color\_manual(values=four\_colors)+

scale\_shape\_manual(values=four\_shapes)+

scale\_fill\_manual(values=four\_colors)+

#ylim(0,1)+

theme\_bw()+

ylab("Stomatal Conductance (mol m-2 s-1)")+

xlab("Relative Humidity")+

labs(

title=NULL,

subtitle = NULL

) +

theme(

text = element\_text(size=16, family="mont"),

legend.title = element\_text(size=16, family="mont", face="bold"),

legend.text = element\_text(size=14, family="mont"),

legend.position="inside",

legend.title.position = "top",

legend.position.inside=c(0.15,0.75),

legend.key.height = unit(.3, "cm"),

legend.background = element\_rect(color=four\_colors[3], fill=NA,

linewidth=.5, linetype = 2),

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=20, family="open", face="bold", lineheight = .5)

)

# gsw by Minutes from Start

ggplot(data = d23\_li, aes(x=minutesfromstart, y = gsw, color = rh\_s)) +

geom\_point()+

scale\_color\_scico(begin=0.9, end=0, palette=a\_palette)+

theme\_bw()+

ylab("Stomatal Conductance (mol m-2 s-1)")+

xlab("Minutes From Start")+

labs(

title=NULL,

subtitle = NULL

) +

guides(color=guide\_colorbar(title=str\_wrap("Relative Humidity", 8)))+

theme(

text = element\_text(size=16, family="mont"),

legend.position="right",

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=20, family="open", face="bold", lineheight = .8)

)

# multispeq

## phiPS2

### because phiPS2 is a proportion [0:1], we don't have to create PDFs and CDFs or perform KS tests. We can still do those to be thorough, but it's not necessary. For a proportion we can just logit transform it then use a linear model.

### check for heteroscedasticity

###### Pr(>F) of < 0.05 means that the value is homoscedastic

leveneTest(d23\_m$phi2~d23\_m$treatment)

leveneTest(d23\_m$logitps2~d23\_m$treatment)

### box plot of logit PhiPS2 by treatment

ggplot(data = d23\_m, aes(x= treatment, y = logitps2,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("PhiPS2")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

### logit PhiPS2 by date scatterplot

ggplot(data = d23\_m, aes(x= date, y = logitps2,

fill=treatment, color=treatment,

shape = device.id)) +

# facet\_wrap(~plant)+

geom\_jitter( width=2, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("PhiPS2")+

xlab("Date")+

theme\_bw()+

theme(

legend.position="right",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_phi2\_by\_date.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_phi2\_by\_date.png", path = "C:/Github/Thesis/figures/TIP23")

### logit PhiPS2 by time scatterplot

ggplot(data = d23\_m, aes(x= time, y = logitps2,

fill=treatment, color=treatment,

shape = device.id)) +

geom\_jitter( width=0, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

scale\_x\_discrete(guide=guide\_axis(check.overlap=TRUE))+

labs(

title=NULL,

subtitle = NULL

) +

ylab("PhiPS2")+

xlab("Time")+

theme\_bw()+

theme(

legend.position="right",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_phi2\_by\_time.svg", path = "C:/Github/Thesis/figures/TIP23")

ggsave("23\_phi2\_by\_time.png", path = "C:/Github/Thesis/figures/TIP23")

**III.B.4. Models and Post Hoc Tests**

## mass

mod23\_mass <- lm(log(mass\_mean) ~ treatment, data = d23\_fl\_means)

AIC(mod23\_mass)

summary(mod23\_mass)

comps <- glht(mod23\_mass, linfct = mcp(treatment = "Tukey"))

mod23\_mass\_letters <- cld(comps)$mcletters$Letters

print(mod23\_mass\_letters)

## annotated mass boxplot

ggplot(data = d23\_fl\_means, aes(x= treatment, y = mass\_mean,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

annotate("text", x=1:4, y=140, label = mod23\_mass\_letters, size=10, family="open")+

ylab("Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_mass\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width =10)

ggsave("23\_mass\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width =10)

## ber

mod23\_ber <- lm(logit(pber) ~ treatment, data = d23\_fg\_summary)

AIC(mod23\_ber)

summary(mod23\_ber)

comps <- glht(mod23\_ber, linfct = mcp(treatment = "Tukey"))

mod23\_ber\_letters <- cld(comps)$mcletters$Letters

print(mod23\_ber\_letters)

ggplot(data = d23\_fg\_summary, aes(x= treatment, y = pber,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

scale\_y\_continuous(labels = yticks)+

labs(

title=NULL,

subtitle = NULL

) +

annotate("text", x=1:4, y=.85, label = mod23\_ber\_letters, size=10, family="open")+

ylab("Blossom-end Rot (%)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_ber\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width =10)

ggsave("23\_ber\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width =10)

## sugar

mod23\_sug <- lm(logit(sugar\_avg\_mean)~ treatment, data = d23\_fl\_means)

AIC(mod23\_sug)

summary(mod23\_sug)

comps <- glht(mod23\_sug, linfct = mcp(treatment = "Tukey"))

mod23\_sug\_letters <- cld(comps)$mcletters$Letters

print(mod23\_sug\_letters)

## box plot of total sugar by Treatment

ggplot(data = d23\_fl\_means, aes(x= treatment, y = sugar\_avg\_mean,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

annotate("text", x=1:4, y=10, label = mod23\_sug\_letters, size=10, family="open")+

ylab("Sugar (%)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_sug\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width =10)

ggsave("23\_sug\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width =10)

## sug by mass

d23\_fl\_means$sug <- d23\_fl\_means$sugar\_avg\_mean / 100

mod23\_sug2 <- lm(logit(sug) ~ treatment + log(mass\_mean), data = d23\_fl\_means)

AIC(mod23\_sug2)

summary(mod23\_sug2)

comps <- glht(mod23\_sug2, linfct = mcp(treatment = "Tukey"))

mod23\_sug2\_letters <- cld(comps)$mcletters$Letters

print(mod23\_sug2\_letters)

predict\_plot(mod23\_sug2, d23\_fl\_means, sug, mass\_mean, treatment, 100, correction = "logit")+

ylab("Sugar (%)")+

xlab("Mass (g)")+

labs(

title=NULL,

subtitle = NULL

) +

scale\_y\_continuous(labels = yticks)+

guides(fill=guide\_legend(title="Treatment"),

color=guide\_legend(title="Treatment"))+

theme(

legend.position="inside",

legend.position.inside = c(.8,.8),

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

axis.text = element\_text(size=subtitle\_size, family = "mont"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_sugbymass\_pred\_CI.svg", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width = 10)

ggsave("23\_sugbymass\_pred\_CI.png", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width = 10)

## fruit count

mod23\_fc <- glm(fruit\_sum ~ treatment, data = d23\_fg\_summary,

family = poisson(link = "identity"))

AIC(mod23\_fc)

summary(mod23\_fc)

glm\_pseudor2(mod23\_fc)

comps <- glht(mod23\_fc, linfct = mcp(treatment = "Tukey"))

mod23\_fc\_letters <- cld(comps)$mcletters$Letters

print(mod23\_fc\_letters)

ggplot(data = d23\_fg\_summary, aes(x= treatment, y = fruit\_sum,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

annotate("text", x=1:4, y=100, label = mod23\_fc\_letters, size=10, family="open")+

ylab("Total Fruit")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

axis.text = element\_text(size=subtitle\_size, family = "mont"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_fc\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width = 10)

ggsave("23\_fc\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width = 10)

## marketable fruit count

mod23\_fc2 <- glm(fruit\_sum ~ treatment, data = d23\_fl\_summary,

family = poisson(link = "identity"))

AIC(mod23\_fc2)

summary(mod23\_fc2)

glm\_pseudor2(mod23\_fc2)

comps <- glht(mod23\_fc2, linfct = mcp(treatment = "Tukey"))

mod23\_fc2\_letters <- cld(comps)$mcletters$Letters

print(mod23\_fc2\_letters)

ggplot(data = d23\_fl\_summary, aes(x= treatment, y = fruit\_sum,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

annotate("text", x=1:4, y=35, label = mod23\_fc2\_letters, size=10, family="open")+

ylab("Marketable Fruit")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

axis.text = element\_text(size=subtitle\_size, family = "mont"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_mfc\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width = 10)

ggsave("23\_mfc\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width = 10)

## total fruit mass

mod23\_msum <- lm(mass\_sum ~ treatment, data = d23\_fg\_summary)

AIC(mod23\_msum)

summary(mod23\_msum)

comps <- glht(mod23\_msum, linfct = mcp(treatment = "Tukey"))

mod23\_msum\_letters <- cld(comps)$mcletters$Letters

print(mod23\_msum\_letters)

ggplot(data = d23\_fg\_summary, aes(x= treatment, y = mass\_sum,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

annotate("text", x=1:4, y=6000, label = mod23\_msum\_letters, size=10, family="open")+

ylab("Total Fruit Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

axis.text = element\_text(size=subtitle\_size, family = "mont"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_fmsum\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width = 10)

ggsave("23\_fmsum\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width = 10)

## marketable fruit mass

mod23\_mmsum <- lm(mass\_sum ~ treatment, data = d23\_fl\_summary)

AIC(mod23\_mmsum)

summary(mod23\_mmsum)

comps <- glht(mod23\_mmsum, linfct = mcp(treatment = "Tukey"))

mod23\_mmsum\_letters <- cld(comps)$mcletters$Letters

print(mod23\_mmsum\_letters)

ggplot(data = d23\_fl\_summary, aes(x= treatment, y = mass\_sum,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

annotate("text", x=1:4, y=3300, label = mod23\_mmsum\_letters, size=10, family="open")+

ylab("Marketable Fruit Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

axis.text = element\_text(size=subtitle\_size, family = "mont"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_mfmsum\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width = 10)

ggsave("23\_mfmsum\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width = 10)

# fluorescence

## phi2 pca

mod23\_phi2\_3 <- lmer(logit(phips2) ~ treatment + PC1 + PC2 + (1 | device), data = d23\_pca)

AIC(mod23\_phi2\_3)

summary(mod23\_phi2\_3)

r.squaredGLMM(mod23\_phi2\_3)

comps <- glht(mod23\_phi2\_3, linfct = mcp(treatment = "Tukey"))

mod23\_phi2\_3\_letters <- cld(comps)$mcletters$Letters

print(mod23\_phi2\_3\_letters)

mod23\_phi2\_3r <- lm(logit(phips2) ~ treatment + PC1, data = d23\_pca)

predict\_plot(mod23\_phi2\_3r, d23\_pca, phips2, PC1, treatment, 100,

correction = "logit")+

ylim(0.5,0.8)+

ylab("PhiPS2")+

xlab("PC1")+

labs(

title=NULL,

subtitle = NULL

) +

guides(fill=guide\_legend(title="Treatment"),

color=guide\_legend(title="Treatment"))

## phi2

mod23\_phi2 <- lmer(logit(phips2) ~ treatment + daysfromgermination + (1 | device), data = d23\_ps2\_joined)

AIC(mod23\_phi2)

summary(mod23\_phi2)

r.squaredGLMM(mod23\_phi2)

comps <- glht(mod23\_phi2, linfct = mcp(treatment = "Tukey"))

mod23\_phi2\_letters <- cld(comps)$mcletters$Letters

print(mod23\_phi2\_letters)

mod23\_phi2\_2 <- lm(logit(phips2) ~ treatment + daysfromgermination, data = d23\_ps2\_joined)

predict\_plot(mod23\_phi2\_2, d23\_ps2\_joined, phips2, daysfromgermination, treatment, 100,

correction = "logit")+

ylim(0.5,0.8)+

ylab("PhiPS2")+

xlab("Days From Germination")+

labs(

title=NULL,

subtitle = NULL

) +

guides(fill=guide\_legend(title="Treatment"),

color=guide\_legend(title="Treatment"))

ggsave("23\_phi2\_pred\_CI.svg", path = "C:/Github/Thesis/figures/TIP23",

width = 10, height = 6)

ggsave("23\_phi2\_pred\_CI.png", path = "C:/Github/Thesis/figures/TIP23",

width = 10, height = 6)

## gsw

mod23\_gsw <- lm(log(gsw) ~ treatment + rh\_s, data = d23\_li)

AIC(mod23\_gsw)

summary(mod23\_gsw)

comps <- glht(mod23\_gsw, linfct = mcp(treatment = "Tukey"))

mod23\_gsw\_letters <- cld(comps)$mcletters$Letters

print(mod23\_gsw\_letters)

mod23\_gsw2 <- lm(log(gsw) ~ treatment + rh\_s + tref + daysfromgermination + minutesfromstart, data = d23\_li)

AIC(mod23\_gsw2)

summary(mod23\_gsw2)

#predict\_plot(mod23\_gsw2, d23\_li, gsw, rh\_s, treatment, 100,

# correction = "exponential")

predict\_plot(mod23\_gsw, d23\_li, gsw, rh\_s, treatment, 100,

correction = "exponential")+

ylab("Stomatal Conductance (mol m-2 s-1)")+

xlab("Relative Humidity (%)")+

labs(

title=NULL,

subtitle = NULL

) +

ylim(0, 2.5)+

guides(fill=guide\_legend(title="Treatment"),

color=guide\_legend(title="Treatment"))+

scale\_color\_scico\_d(begin = 0.9, end = 0.1, palette = a\_palette)+

scale\_fill\_scico\_d(begin = 0.9, end = 0.1, palette = a\_palette)

ggsave("23\_gsw\_pred\_CI.svg", path = "C:/Github/Thesis/figures/TIP23",

width = 10, height = 6)

ggsave("23\_gsw\_pred\_CI.png", path = "C:/Github/Thesis/figures/TIP23",

width = 10, height = 6)

# stomatal density

d23\_st <- d23\_st %>%

mutate(treatment = as.factor(treatment))

d23\_st\_lower <- d23\_st %>% filter(surface == "Lower")

d23\_st\_sum <- d23\_st\_lower %>%

group\_by(treatment, plant) %>%

summarise\_at(vars(stomatal\_density), list(mean)) %>%

ungroup()

mod23\_st <- lm(log(stomatal\_density) ~ treatment, data = d23\_st\_sum)

AIC(mod23\_st)

summary(mod23\_st)

comps <- glht(mod23\_st, linfct = mcp(treatment = "Tukey"))

mod23\_st\_letters <- cld(comps)$mcletters$Letters

print(mod23\_st\_letters)

ggplot(data = d23\_st\_sum, aes(x= treatment, y = stomatal\_density,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

annotate("text", x=1:4, y=35, label = mod23\_st\_letters, size=10, family="open")+

ylab(str\_wrap("Stomatal Density (stomates/cm^2",20))+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("23\_st\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width =10)

ggsave("23\_st\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP23",

height = 6, width =10)

**III.C. Tomato Inoculant Timing Code**

**III.C.1. Setup**

germdate24 <- "2024-05-01"

treatment\_order24 <- c("Control",

"Transplantation",

"Germination",

"Germ+Trans")

d24\_f\_file <- "https://raw.githubusercontent.com/zachpeagler/Thesis/refs/heads/main/data/TIP/2024/TIP24\_Fruit.csv"

d24\_m\_file <- "https://raw.githubusercontent.com/zachpeagler/Thesis/refs/heads/main/data/TIP/2024/TIP24\_Multispeq.csv"

d24\_li\_file <- "https://raw.githubusercontent.com/zachpeagler/Thesis/refs/heads/main/data/TIP/2024/TIP24\_LI600.csv"

**III.C.2. Data Cleaning**

d24\_f <- read.csv(d24\_f\_file) %>%

filter(mass>0)%>%

rename(pot = plant) %>%

mutate(row = case\_when(

row==1~"A",

row==2~"B",

row==3~"C",

row==4~"D"),

treatment = case\_when(

row=="A"~"Control",

row=="B"~"Transplantation",

row=="C"~"Germination",

row=="D"~"Germ+Trans",

TRUE~NA),

transplantation = case\_when(

row=="A"~FALSE,

row=="B"~TRUE,

row=="C"~FALSE,

row=="D"~TRUE

),

germination = case\_when(

row=="A"~FALSE,

row=="B"~FALSE,

row=="C"~TRUE,

row=="D"~TRUE

),

fruit = 1,

date\_analysis = parse\_date\_time(date\_analysis, orders = "mdy"),

date\_harvest = parse\_date\_time(date\_harvest, orders = "mdy"),

daysfromharvesttoanalysis = as.numeric(round(difftime(date\_analysis, date\_harvest, units = c("days")), 0)),

daysfromgermination = as.numeric(round(difftime(date\_analysis, germdate24, units = c("days")), 0)),

plant = as.factor(paste0(row, pot)),

pot = as.factor(pot),

treatment = factor(treatment, levels = treatment\_order24),

plant = factor(plant, levels = plant\_order24),

BER = as.logical(BER),

fungus = as.logical(fungus),

cracking = as.logical(cracking),

ripeness = abs(1 - round(penetrometer/max(na.omit(penetrometer)), 2)),

sugar\_grams = (sugar\_avg/100)\*mass

)

colnames(d24\_f) <- tolower(colnames(d24\_f))

d24\_f\_export <- d24\_f[,-c(8,9)]

### Make summary dataframe with mean and sd of mass and sugar by plant.

d24\_f\_means <- na.omit(d24\_f) %>%

group\_by(treatment, plant) %>%

summarise\_at(vars(mass, sugar\_avg, sugar\_grams), list(mean=mean, sd=sd))

d24\_f\_means2 <- na.omit(d24\_f) %>%

group\_by(treatment) %>%

summarise\_at(vars(mass, sugar\_avg, sugar\_grams), list(mean=mean, sd=sd))

### Sum the fruit, BER, sugar, and mass by treatment group, then get ber probability.

d24\_f\_summary <- d24\_f %>%

group\_by(treatment, germination, transplantation, plant) %>%

summarise\_at(vars(fruit, ber, fungus, cracking, mass), list(sum=sum)) %>%

mutate(pber = round(ber\_sum/fruit\_sum, 4),

pfungus = round(fungus\_sum/fruit\_sum, 4),

pcracking = round(cracking\_sum/fruit\_sum, 4)

)

### Sum the fruit, BER, sugar, and mass by treatment group, then get ber probability.

d24\_f\_summary\_marketable <- d24\_f %>%

filter(ber == FALSE, fungus == FALSE, cracking == FALSE) %>%

group\_by(treatment, germination, transplantation, plant) %>%

summarise\_at(vars(fruit, mass), list(sum=sum)) %>%

ungroup()

d24\_f\_summary2 <- d24\_f %>%

group\_by(treatment) %>%

summarise\_at(vars(fruit, ber, fungus, cracking, mass), list(sum=sum)) %>%

mutate(pber = round(ber\_sum/fruit\_sum, 4),

pfungus = round(fungus\_sum/fruit\_sum, 4),

pcracking = round(cracking\_sum/fruit\_sum, 4)

)

### sugar summary

d24\_f\_summary\_sugar <- na.omit(d24\_f) %>%

group\_by(treatment, plant, germination, transplantation) %>%

summarise\_at(vars(fruit, mass, sugar\_grams), list(sum=sum))

## Li-600

d24\_li <- read.csv(d24\_li\_file, stringsAsFactors = F) %>%

mutate(row\_let = case\_when(

Row==1~"A",

Row==2~"B",

Row==3~"C",

Row==4~"D"),

Treatment = case\_when(

Row==1~"Control",

Row==2~"Transplantation",

Row==3~"Germination",

Row==4~"Germ+Trans",

TRUE~NA),

transplantation = case\_when(

row\_let=="A"~FALSE,

row\_let=="B"~TRUE,

row\_let=="C"~FALSE,

row\_let=="D"~TRUE

),

germination = case\_when(

row\_let=="A"~FALSE,

row\_let=="B"~FALSE,

row\_let=="C"~TRUE,

row\_let=="D"~TRUE

)) %>%

filter(leak\_pct<10) %>%

rename(Date\_ref = Date) %>%

mutate(Date = parse\_date\_time(Date\_ref, orders = "mdy"),

Time = parse\_date\_time(Time, orders = "T"),

DaysFromGermination = as.numeric(round(difftime(Date, germdate24, units = c("days")), 0)),

plant = as.factor(paste0(row\_let, Pot)),

plant = factor(plant, levels = plant\_order24),

Treatment = factor(Treatment,

levels = treatment\_order24),

logitPS2 = logit(PhiPS2, FALSE)

) %>%

group\_by(DaysFromGermination) %>%

mutate(MinutesFromStart = round(difftime(Time, min(Time), units = "mins"), 2)) %>%

ungroup() %>%

mutate(

Time = format(Time, "%H:%M:%S")

)

d24\_li <- d24\_li[,c(2,3,7,8,10,17,28,32,34,35,36,39,40,114,115,116,117,118,119,120,121,122)]

colnames(d24\_li) <- tolower(colnames(d24\_li))

d24\_li\_export <- d24\_li[,-c(2,3)] %>%

mutate(pot = as.factor(pot))

## multispeq

d24\_m <- read.csv(d24\_m\_file) %>%

mutate(Treatment = case\_when(

Row=="A"~"Control",

Row=="B"~"Transplantation",

Row=="C"~"Germination",

Row=="D"~"Germ+Trans",

TRUE~NA),

transplantation = case\_when(

Row=="A"~FALSE,

Row=="B"~TRUE,

Row=="C"~FALSE,

Row=="D"~TRUE

),

germination = case\_when(

Row=="A"~FALSE,

Row=="B"~FALSE,

Row=="C"~TRUE,

Row=="D"~TRUE

)) %>%

mutate(Row\_num = case\_when(

Row=="A"~1,

Row=="B"~2,

Row=="C"~3,

Row=="D"~4),

Plant = as.factor(paste0(Row, Pot)),

Plant = factor(Plant, levels = plant\_order24),

Pot = as.factor(Pot),

Row = as.factor(Row),

Device.ID = as.factor(Device.ID),

Date = as.Date(time, "%m/%d/%Y"),

time = parse\_date\_time(time, "%m/%d/%Y %H:%M"),

Time = format(time, "%H:%M:%S"),

logitPS2 = logit(Phi2, FALSE),

logitFvPFmP = logit(FvP\_over\_FmP, FALSE)

)

d24\_m <- d24\_m[,c(1,4,5,6,7,8,10,11,12,21,27,30,34,35,41,59,63,64,65,66,67,68,69,70,71)]

colnames(d24\_m) <- tolower(colnames(d24\_m))

**III.C.3. Exploratory Analysis**

## mass

### PDF, CDF, and KS test

multipdf\_plot(d24\_f$mass, 100, "all", a\_palette, "mass")

ggsave("24\_mass\_PDF.svg", path = "C:/Github/Thesis/figures/TIP24")

ggsave("24\_mass\_PDF.png", path = "C:/Github/Thesis/figures/TIP24")

multicdf\_plot(d24\_f$mass, 100, "all", a\_palette, "mass")

ggsave("24\_mass\_CDF.svg", path = "C:/Github/Thesis/figures/TIP24")

ggsave("24\_mass\_CDF.png", path = "C:/Github/Thesis/figures/TIP24")

multiks\_cont(d24\_f$mass, "all")

## check for heteroscedasticity

### in mass as a function of treatment

leveneTest(d24\_f$mass~d24\_f$treatment)

## box plot of mass by Treatment

ggplot(data = d24\_f, aes(x= treatment, y = mass,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

## box plot by Treatment colored by BER

ggplot(data = d24\_f, aes(x= treatment, y = mass,

fill=ber, color=ber)) +

geom\_boxplot(alpha=.5, width=0.6, outliers = TRUE)+

scale\_fill\_manual(values=two\_colors)+

scale\_color\_manual(values=two\_colors)+

guides(fill=guide\_legend(title=str\_wrap("Blossom-End Rot", 10)),

color=guide\_legend(title=str\_wrap("Blossom-End Rot", 10))

)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="right",

legend.title = element\_text(size=subtitle\_size, family="mont",

face = "bold", lineheight=0.5),

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("24\_mass\_by\_ber.svg", path = "C:/Github/Thesis/figures/TIP24")

ggsave("24\_mass\_by\_ber.png", path = "C:/Github/Thesis/figures/TIP24")

## summarized fruit graphs

### box plot of mean mass by treatment

ggplot(data = d24\_f\_means, aes(x= treatment, y = mass\_mean,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("24\_mass\_mean\_box.svg", path = "C:/Github/Thesis/figures/TIP24")

ggsave("24\_mass\_mean\_box.png", path = "C:/Github/Thesis/figures/TIP24")

### box plot of mean sugar by treatment

ggplot(data = d24\_f\_means, aes(x= treatment, y = sugar\_avg\_mean,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Sugar (%)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("24\_sugar\_mean\_box.svg", path = "C:/Github/Thesis/figures/TIP24")

ggsave("24\_sugar\_mean\_box.png", path = "C:/Github/Thesis/figures/TIP24")

### box plot of percent BER by treatment

ggplot(data = d24\_f\_summary, aes(x= treatment, y = pber,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("pBER")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("24\_ber\_box.svg", path = "C:/Github/Thesis/figures/TIP24")

ggsave("24\_ber\_box.png", path = "C:/Github/Thesis/figures/TIP24")

### box plot of total fruit mass by treatment

ggplot(data = d24\_f\_summary, aes(x= treatment, y = mass\_sum,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

### box plot of fruit count by treatment

ggplot(data = d24\_f\_summary, aes(x= treatment, y = fruit\_sum,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Fruit")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("24\_fruit\_count.svg", path = "C:/Github/Thesis/figures/TIP24")

ggsave("24\_fruit\_count.png", path = "C:/Github/Thesis/figures/TIP24")

## sugar

### PDF, CDF, and KS test

multipdf\_plot(filter(d24\_f, sugar\_grams >0)$sugar\_grams, 100, "all", a\_palette, "sugar (g)")

ggsave("24\_sug\_PDF.svg", path = "C:/Github/Thesis/figures/TIP24")

ggsave("24\_sug\_PDF.png", path = "C:/Github/Thesis/figures/TIP24")

multicdf\_plot(filter(d24\_f, sugar\_grams >0)$sugar\_grams, 100, "all", a\_palette, "sugar (g)")

ggsave("24\_sug\_CDF.svg", path = "C:/Github/Thesis/figures/TIP24")

ggsave("24\_sug\_CDF.png", path = "C:/Github/Thesis/figures/TIP24")

multiks\_cont(filter(d24\_f, sugar\_grams >0)$sugar\_grams, "all")

## check for heteroscedasticity

### in sugar as a function of treatment

leveneTest(d24\_f$sugar\_grams~d24\_f$treatment)

###### NOT homoscedastic! Unequal variances!

## box plot of sugar % by Treatment

ggplot(data = d24\_f, aes(x= treatment, y = sugar\_avg,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Sugar %")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

## box plot of sugar by Treatment

ggplot(data = d24\_f, aes(x= treatment, y = sugar\_grams,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Sugar (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

## scatter plot of sugar by mass

ggplot(data = d24\_f, aes(x= mass, y = sugar\_avg,

fill=treatment, color=treatment)) +

geom\_jitter( width=0, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Sugar %")+

xlab("Mass (g)")+

theme\_bw()+

theme(

legend.position="right",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("24\_sug\_by\_mass.svg", path = "C:/Github/Thesis/figures/TIP24")

ggsave("24\_sug\_by\_mass.png", path = "C:/Github/Thesis/figures/TIP24")

## box plot of total sugar by Treatment

ggplot(data = d24\_f\_summary\_sugar, aes(x= treatment, y = sugar\_grams\_sum,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Sugar (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

# li-600

# PDF, CDF, and KS test

multipdf\_plot(filter(d24\_li, gsw >0)$gsw, 100, "all", a\_palette)

ggsave("24\_gsw\_PDF.svg", path = "C:/Github/Thesis/figures/TIP24")

ggsave("24\_gsw\_PDF.png", path = "C:/Github/Thesis/figures/TIP24")

multicdf\_plot(filter(d24\_li, gsw >0)$gsw, 100, "all", a\_palette)

ggsave("24\_gsw\_CDF.svg", path = "C:/Github/Thesis/figures/TIP24")

ggsave("24\_gsw\_CDF.png", path = "C:/Github/Thesis/figures/TIP24")

multiks\_cont(filter(d24\_li, gsw >0)$gsw, "all")

# check for heteroscedasticity

## in gsw as a function of treatment

leveneTest(d24\_li$gsw~d24\_li$treatment)

## in gsw as a function of time from start

leveneTest(d24\_li$gsw~d24\_li$minutesfromstart)

## box plot by Treatment boxplot

ggplot(data = d24\_li, aes(x= treatment, y = gsw, fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.3, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Stomatal Conductance (mol m-2 s-1)")+

# annotate("text", x=1:4, y=3, label = c("a", "b", "b", "b"), size=5, family="open")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=16, family="mont", lineheight=0.8),

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=20, family="open", face="bold")

)

# GSW by date across relative humidity

ggplot(data = d24\_li, aes(x=daysfromgermination, y = gsw,

color = rh\_s)) +

geom\_jitter(width=1, size=2, alpha=1)+

scale\_color\_scico(begin=0.9, end=0, palette=a\_palette)+

# scale\_x\_discrete(guide=guide\_axis(check.overlap=TRUE))+

theme\_bw()+

ylab("Stomatal Conductance (mol m-2 s-1)")+

xlab("Days From Germination")+

labs(

title=NULL,

subtitle = NULL

) +

guides(color=guide\_colorbar(title=str\_wrap("Relative Humidity", 10)))+

theme(

text = element\_text(size=16, family="mont"),

legend.title = element\_text(size=16, family="mont", face="bold"),

legend.text = element\_text(size=14, family="mont"),

legend.position="right",

legend.title.position = "top",

legend.key.height = unit(.4, "cm"),

legend.background = element\_rect(color=two\_colors[2], fill=NA,

linewidth=.5, linetype = 2),

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=20, family="open", face="bold", lineheight = .8)

)

# GSW by time across relative humidity

ggplot(data = d24\_li, aes(x=time, y = gsw,

color = date,

shape = treatment)) +

geom\_jitter(width=1, size=2, alpha=1)+

scale\_color\_scico(begin=0.9, end=0, palette=a\_palette)+

scale\_shape\_manual(values=four\_shapes)+

scale\_x\_discrete(guide=guide\_axis(check.overlap=TRUE))+

theme\_bw()+

ylab("Stomatal Conductance (mol m-2 s-1)")+

xlab("Time")+

labs(

title=NULL,

subtitle = NULL

) +

guides(color=guide\_legend(title="Date"))+

theme(

text = element\_text(size=16, family="mont"),

legend.title = element\_text(size=16, family="mont", face="bold"),

legend.text = element\_text(size=14, family="mont"),

legend.position="right",

legend.title.position = "top",

legend.key.height = unit(.3, "cm"),

legend.background = element\_rect(color=two\_colors[2], fill=NA,

linewidth=.5, linetype = 2),

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=20, family="open", face="bold", lineheight = .5)

)

# gsw by relative humidity across treatment groups

ggplot(data = d24\_li, aes(x=rh\_s, y = gsw,

shape=treatment, fill=treatment, color = treatment)) +

geom\_jitter(width=1, size=2, alpha=1)+

scale\_color\_manual(values=four\_colors)+

scale\_shape\_manual(values=four\_shapes)+

scale\_fill\_manual(values=four\_colors)+

#ylim(0,1)+

theme\_bw()+

ylab("Stomatal Conductance (mol m-2 s-1)")+

xlab("Relative Humidity")+

labs(

title=NULL,

subtitle = NULL

) +

theme(

text = element\_text(size=16, family="mont"),

legend.title = element\_text(size=16, family="mont", face="bold"),

legend.text = element\_text(size=14, family="mont"),

legend.position="inside",

legend.title.position = "top",

legend.position.inside=c(0.15,0.75),

legend.key.height = unit(.3, "cm"),

legend.background = element\_rect(color=four\_colors[3], fill=NA,

linewidth=.5, linetype = 2),

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=20, family="open", face="bold", lineheight = .5)

)

# gsw by Minutes from Start

ggplot(data = d24\_li, aes(x=minutesfromstart, y = gsw, color = rh\_s)) +

geom\_point()+

scale\_color\_scico(begin=0.9, end=0, palette=a\_palette)+

theme\_bw()+

ylab("Stomatal Conductance (mol m-2 s-1)")+

xlab("Minutes From Start")+

labs(

title=NULL,

subtitle = NULL

) +

guides(color=guide\_colorbar(title=str\_wrap("Relative Humidity", 8)))+

theme(

text = element\_text(size=16, family="mont"),

legend.position="right",

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=20, family="open", face="bold", lineheight = .8)

)

# multispeq

## phiPS2

### because phiPS2 is a proportion [0:1], we don't have to create PDFs and CDFs or perform KS tests. We can still do those to be thorough, but it's not necessary. For a proportion we can just logit transform it then use a linear model.

### check for heteroscedasticity

###### Pr(>F) of < 0.05 means that the value is homoscedastic

leveneTest(d24\_m$phi2~d24\_m$treatment)

leveneTest(d24\_m$logitps2~d24\_m$treatment)

### box plot of logit PhiPS2 by treatment

ggplot(data = d24\_m, aes(x= treatment, y = logitps2,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("PhiPS2")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

### logit PhiPS2 by date scatterplot

ggplot(data = d24\_m, aes(x= date, y = logitps2,

fill=treatment, color=treatment,

shape = device.id)) +

# facet\_wrap(~plant)+

geom\_jitter( width=2, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("PhiPS2")+

xlab("Date")+

theme\_bw()+

theme(

legend.position="right",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

### logit PhiPS2 by time scatterplot

ggplot(data = d24\_ps2\_joined, aes(x= time, y = logitps2,

fill=treatment, color=treatment,

shape = Device)) +

geom\_jitter( width=0, height=0)+

ylim(0,1.5)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

scale\_x\_discrete(guide=guide\_axis(check.overlap=TRUE))+

labs(

title=NULL,

subtitle = NULL

) +

ylab("PhiPS2")+

xlab("Time")+

theme\_bw()+

theme(

legend.position="right",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

**III.C.4. Models and Post Hoc Tests**

## ber

mod24\_ber <- lm(logit(pber) ~ treatment, data=d24\_f\_summary)

AIC(mod24\_ber)

summary(mod24\_ber)

comps <- glht(mod24\_ber, linfct = mcp(treatment = "Tukey"))

mod24\_ber\_letters <- cld(comps)$mcletters$Letters

print(mod24\_ber\_letters)

ggplot(data = d24\_f\_summary, aes(x= treatment, y = pber \* 100,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

annotate("text", x=1:4, y=22, label = mod24\_ber\_letters, size=10, family="open")+

ylab("Blossom-end Rot (%)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("24\_ber\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

ggsave("24\_ber\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

## mass

mod24\_mass <- lm(log(mass\_mean) ~ treatment, data=d24\_f\_means)

AIC(mod24\_mass)

summary(mod24\_mass)

confint(mod24\_mass)

comps <- glht(mod24\_mass, linfct = mcp(treatment = "Tukey"))

mod24\_mass\_letters <- cld(comps)$mcletters$Letters

print(mod24\_mass\_letters)

ggplot(data = d24\_f\_means, aes(x= treatment, y = mass\_mean,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

annotate("text", x=1:4, y=200, label = mod24\_mass\_letters, size=10, family="open")+

ylab("Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("24\_mass\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

ggsave("24\_mass\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

## sugar

d24\_f\_means$sug <- d24\_f\_means$sugar\_avg\_mean / 100

mod24\_sug <- lm(logit(sug) ~ treatment, data=d24\_f\_means)

AIC(mod24\_sug)

summary(mod24\_sug)

comps <- glht(mod24\_sug, linfct = mcp(treatment = "Tukey"))

mod24\_sug\_letters <- cld(comps)$mcletters$Letters

print(mod24\_sug\_letters)

ggplot(data = d24\_f\_means, aes(x= treatment, y = sug \* 100,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

annotate("text", x=1:4, y=8.5, label = mod24\_sug\_letters, size=10, family="open")+

ylab("Sugar (%)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("24\_sug\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

ggsave("24\_sug\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

## sugar by mass

mod24\_sugm <- lm(logit(sug) ~ treatment + log(mass\_mean), data=d24\_f\_means)

summary(mod24\_sugm)

comps <- glht(mod24\_sugm, linfct = mcp(treatment = "Tukey"))

mod24\_sugm\_letters <- cld(comps)$mcletters$Letters

print(mod24\_sugm\_letters)

yticks <- function(x) {

x \* 100

}

predict\_plot(mod24\_sugm, d24\_f\_means, sug, mass\_mean, treatment, 100, correction = "logit")+

labs(title = NULL,

subtitle = NULL)+

ylab("Sugar (%)")+

xlab("Mass (g)")+

scale\_y\_continuous(labels = yticks)+

theme\_bw()+

guides(

fill = guide\_legend(title="Treatment"),

color= guide\_legend(title="Treatment"))+

theme(

legend.position="inside",

legend.position.inside = c(.8,.8),

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("24\_sug\_mass\_pred.svg", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

ggsave("24\_sug\_mass\_pred.png", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

## fruit count

mod24\_fc <- glm(fruit\_sum ~ treatment, data=d24\_f\_summary, family = poisson(link="identity"))

summary(mod24\_fc)

confint(mod24\_fc)

glm\_pseudor2(mod24\_fc)

comps <- glht(mod24\_fc, linfct = mcp(treatment = "Tukey"))

mod24\_fc\_letters <- cld(comps)$mcletters$Letters

print(mod24\_fc\_letters)

#pairs(emmeans(mod24\_fc, ~treatment))

ggplot(data = d24\_f\_summary, aes(x= treatment, y = fruit\_sum,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

annotate("text", x=1:4, y=80, label = mod24\_fc\_letters, size=10, family="open")+

ylab("Total Fruit")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("24\_fc\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

ggsave("24\_fc\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

## marketable fruit count

mod24\_mfc <- glm(fruit\_sum ~ treatment, data=d24\_f\_summary\_marketable, family = poisson(link="identity"))

summary(mod24\_mfc)

confint(mod24\_mfc)

glm\_pseudor2(mod24\_mfc)

comps <- glht(mod24\_mfc, linfct = mcp(treatment = "Tukey"))

mod24\_mfc\_letters <- cld(comps)$mcletters$Letters

print(mod24\_mfc\_letters)

#pairs(emmeans(mod24\_fc, ~treatment))

ggplot(data = d24\_f\_summary\_marketable, aes(x= treatment, y = fruit\_sum,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

annotate("text", x=1:4, y=70, label = mod24\_mfc\_letters, size=10, family="open")+

ylab("Marketable Fruit")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("24\_mfc\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

ggsave("24\_mfc\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

## total mass

mod24\_tm <- lm(mass\_sum ~ treatment, data=d24\_f\_summary)

summary(mod24\_tm)

confint(mod24\_tm)

comps <- glht(mod24\_tm, linfct = mcp(treatment = "Tukey"))

mod24\_tm\_letters <- cld(comps)$mcletters$Letters

print(mod24\_tm\_letters)

ggplot(data = d24\_f\_summary, aes(x= treatment, y = mass\_sum,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

annotate("text", x=1:4, y=4000, label = mod24\_tm\_letters, size=10, family="open")+

ylab("Total Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("24\_tm\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

ggsave("24\_tm\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

## total marketable mass

mod24\_tmm <- lm(mass\_sum ~ treatment, data=d24\_f\_summary\_marketable)

summary(mod24\_tmm)

confint(mod24\_tmm)

comps <- glht(mod24\_tmm, linfct = mcp(treatment = "Tukey"))

mod24\_tmm\_letters <- cld(comps)$mcletters$Letters

print(mod24\_tmm\_letters)

ggplot(data = d24\_f\_summary\_marketable, aes(x= treatment, y = mass\_sum,

fill=treatment, color=treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

annotate("text", x=1:4, y=3000, label = mod24\_tmm\_letters, size=10, family="open")+

ylab("Total Marketable Mass (g)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("24\_tmm\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

ggsave("24\_tmm\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP24",

height = 6, width = 10)

# fluorescence

## gsw

mod24\_gsw <- lm(log(gsw) ~ treatment + rh\_s, data = filter(d24\_li, gsw>0))

AIC(mod24\_gsw)

summary(mod24\_gsw)

comps <- glht(mod24\_gsw, linfct = mcp(treatment = "Tukey"))

mod24\_gsw\_letters <- cld(comps)$mcletters$Letters

print(mod24\_gsw\_letters)

predict\_plot(mod24\_gsw, filter(d24\_li, gsw>0), gsw, rh\_s, treatment,

100, correction = "exponential")+

ylab("Stomatal Conductance (mol m-2 s-1)")+

xlab("Relative Humidity (%)")+

ylim(0,2)+

labs(

title=NULL,

subtitle = NULL

) +

guides(

fill = guide\_legend(title="Treatment"),

color= guide\_legend(title="Treatment"))+

scale\_color\_scico\_d(begin = 0.9, end = 0.1, palette = a\_palette)+

scale\_fill\_scico\_d(begin = 0.9, end = 0.1, palette = a\_palette)

ggsave("24\_gsw\_pred\_CI.svg", path = "C:/Github/Thesis/figures/TIP24",

width = 10, height = 6)

ggsave("24\_gsw\_pred\_CI.png", path = "C:/Github/Thesis/figures/TIP24",

width = 10, height = 6)

## phips2

mod24\_ps2 <- lmer(logit(phips2) ~ treatment + daysfromgermination + (1 | Device),

data = d24\_ps2\_joined)

summary(mod24\_ps2)

AIC(mod24\_ps2)

r.squaredGLMM(mod24\_ps2)

comps <- glht(mod24\_ps2, linfct=mcp(treatment="Tukey"))

mod24\_ps2\_letters <- cld(comps)$mcletters$Letters

print(mod24\_ps2\_letters)

mod24\_ps2\_2 <- lm(logit(phips2)~treatment+daysfromgermination, data=d24\_ps2\_joined)

predict\_plot(mod24\_ps2\_2, d24\_ps2\_joined, phips2, daysfromgermination, treatment, 100, correction = "logit")+

ylab("Photosystem II Efficiency (PhiPS2)")+

xlab("Days From Germination")+

ylim(0.55, 0.8)+

labs(

title=NULL,

subtitle = NULL

) +

guides(

fill = guide\_legend(title="Treatment"),

color= guide\_legend(title="Treatment"))+

scale\_color\_scico\_d(begin = 0.9, end = 0.1, palette = a\_palette)+

scale\_fill\_scico\_d(begin = 0.9, end = 0.1, palette = a\_palette)

ggsave("24\_phi2\_pred\_CI.svg", path = "C:/Github/Thesis/figures/TIP24",

width = 10, height = 6)

ggsave("24\_phi2\_pred\_CI.png", path = "C:/Github/Thesis/figures/TIP24",

width = 10, height = 6)

### box plot of logit PhiPS2 by treatment

ggplot(data = d24\_ps2\_joined, aes(x= treatment, y = phips2,

fill=treatment, color=treatment, shape = Device)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Photosystem II Efficiency (PhiPS2)")+

xlab("Treatment")+

annotate("text", x=1:4, y=0.8, label = mod24\_ps2\_letters, size=10, family="open")+

ylim(0.55, 0.8)+

theme\_bw()+

guides(

fill = guide\_legend(title="Treatment"),

color= guide\_legend(title="Treatment"))+

theme(

legend.position="right",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("24\_phi2\_box\_annotated.svg", path = "C:/Github/Thesis/figures/TIP24",

width = 10, height = 8)

ggsave("24\_phi2\_box\_annotated.png", path = "C:/Github/Thesis/figures/TIP24",

width = 10, height = 8)

**III.D. Tomato Inoculant Method Code**

**III.D.1. Setup**

tim\_germ\_date <- "2024-04-01"

tim\_treatment\_order <- c("Control",

"Liquid",

"CG",

"CBG")

tim\_levels <- c("A1", "A2", "A3", "A4", "A5", "A6",

"A7", "A8", "A9", "A10", "A11", "A12", "A13",

"B1", "B2", "B3", "B4", "B5", "B6",

"B7", "B8", "B9", "B10", "B11", "B12", "B13",

"C1", "C2", "C3", "C4", "C5", "C6",

"C7", "C8", "C9", "C10", "C11", "C12", "C13",

"D1", "D2", "D3", "D4", "D5", "D6",

"D7", "D8", "D9", "D10", "D11", "D12", "D13",

"E1", "E2", "E3", "E4", "E5", "E6",

"E7", "E8", "E9", "E10", "E11", "E12",

"F1", "F2", "F3", "F4", "F5", "F6",

"F7", "F8", "F9", "F10", "F11", "F12",

"G1", "G2", "G3", "G4", "G5", "G6",

"G7", "G8", "G9", "G10", "G11", "G12",

"H1", "H2", "H3", "H4", "H5", "H6",

"H7", "H8", "H9", "H10", "H11", "H12"

)

li\_data\_file <- "C:/Github/Thesis/data/TIM/TIM24\_Fluoro.csv"

h\_data\_file <- "C:/Github/Thesis/data/TIM/TIM24\_Height.csv"

DS\_data\_file <- "C:/Github/Thesis/data/TIM/TIM24\_DS.csv"

**III.D.2. Clean Data**

li\_data <- read.csv(li\_data\_file)[,c(2,3,7,8,9,10,17,28,32,34,35,36,39,40)] %>%

filter(leak\_pct<10 & gsw > 0) %>%

mutate(Date = parse\_date\_time(Date, orders = "mdy"),

DaysFromGermination = as.numeric(round(difftime(Date, tim\_germ\_date, units = c("days")), 0)),

Date = as.factor(as.Date(Date)),

Time = parse\_date\_time(Time, orders = "T"),

# Time = as.factor(format(Time, "%H:%M:%S")),

Treatment = factor(Treatment,

levels = tim\_treatment\_order),

Inoculation = case\_when(

Treatment=="Control"~FALSE,

Treatment=="Liquid"~TRUE,

Treatment=="CG"~FALSE,

Treatment=="CBG"~TRUE

),

Chitosan = case\_when(

Treatment=="Control"~FALSE,

Treatment=="Liquid"~FALSE,

Treatment=="CG"~TRUE,

Treatment=="CBG"~TRUE

),

Column = case\_when(

Column==1~"A",

Column==2~"B",

Column==3~"C",

Column==4~"D",

Column==5~"E",

Column==6~"F",

Column==7~"G",

Column==8~"H",

),

Plant = paste0(Column, Row),

LogitPhiPS2 = logit(PhiPS2, FALSE),

Column = as.factor(Column),

Row = as.factor(Row),

Plant = as.factor(Plant),

Plant = factor(Plant, levels = tim\_levels),

P\_atm = P\_atm \* 100

) %>%

rename(Pot=Column) %>%

group\_by(DaysFromGermination) %>%

mutate(MinutesFromStart = as.numeric(round(difftime(Time, min(Time), units = "mins"), 2))) %>%

ungroup() %>%

mutate(

Time = as.factor(format(Time, "%H:%M:%S"))

)

data\_tim\_fluoro <- li\_data[,c(5,16,17,1,2,15,3,4,18,9,10,12,14,11,6,8,19)] %>%

rename(AmbientHumidity = rh\_s,

AmbientTemperature = Tref,

AmbientPressure = P\_atm,

AmbientLight = Qamb,

LeafTemperature = Tleaf

)

DS\_data <- read.csv(DS\_data\_file) %>%

mutate(

Column = case\_when(

Column==1~"A",

Column==2~"B",

Column==3~"C",

Column==4~"D",

Column==5~"E",

Column==6~"F",

Column==7~"G",

Column==8~"H",

),

Inoculation = case\_when(

Treatment=="Control"~FALSE,

Treatment=="Liquid"~TRUE,

Treatment=="CG"~FALSE,

Treatment=="CBG"~TRUE

),

Chitosan = case\_when(

Treatment=="Control"~FALSE,

Treatment=="Liquid"~FALSE,

Treatment=="CG"~TRUE,

Treatment=="CBG"~TRUE

),

Treatment = factor(Treatment, levels = tim\_treatment\_order),

Plant = as.factor(paste0(Column, Row)),

Plant = factor(Plant, levels = tim\_levels),

Row = as.factor(Row),

Column = as.factor(Column)

) %>%

rename(Pot = Column)

data\_tim\_ds <- DS\_data[,c(3,10,11,1,2,12,4,5,6,7,8,9)]

h\_data <- read.csv(h\_data\_file) %>%

mutate(Date = parse\_date\_time(Date, orders = "mdy"),

DaysFromGermination = as.numeric(round(difftime(Date, tim\_germ\_date, units = c("days")), 0)),

Date = as.factor(as.Date(Date)),

Column = case\_when(

Column==1~"A",

Column==2~"B",

Column==3~"C",

Column==4~"D",

Column==5~"E",

Column==6~"F",

Column==7~"G",

Column==8~"H",

),

Inoculation = case\_when(

Treatment=="Control"~FALSE,

Treatment=="Liquid"~TRUE,

Treatment=="CG"~FALSE,

Treatment=="CBG"~TRUE

),

Chitosan = case\_when(

Treatment=="Control"~FALSE,

Treatment=="Liquid"~FALSE,

Treatment=="CG"~TRUE,

Treatment=="CBG"~TRUE

),

Treatment = factor(Treatment, levels = tim\_treatment\_order),

Plant = as.factor(paste0(Column, Row)),

Plant = factor(Plant, levels = tim\_levels),

Row = as.factor(Row),

Column = as.factor(Column)

) %>%

rename(Pot = Column) %>%

filter(Height > 0)

data\_tim\_height <- h\_data[,c(3,7,8,4,6,1,2,9,5)]

**III.D.3. Exploratory Analysis**

#gsw

multipdf\_plot(filter(li\_data, gsw >0)$gsw, 100, "all", a\_palette, "gsw")

multicdf\_plot(filter(li\_data, gsw >0)$gsw, 100, "all", a\_palette, "gsw")

multiks\_cont(filter(li\_data, gsw >0)$gsw, "all")

###### Not significantly different than a Gamma or Normal distribution.

###### Try models with both.

# check for heteroscedasticity

## in gsw as a function of treatment

leveneTest(li\_data$gsw~li\_data$Treatment)

## in gsw as a function of time from start

leveneTest(li\_data$gsw~li\_data$MinutesFromStart)

###### Unequal variances. Uh oh.

## box plot by Treatment boxplot

ggplot(data = li\_data, aes(x= Treatment, y = gsw, fill=Treatment, color=Treatment)) +

geom\_boxplot(alpha=.5, width=0.3, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=str\_wrap("Stomatal Conductance Across Inoculation Treatments in Tomato", 40)

) +

ylab(str\_wrap("Stomatal Conductance (mol m-2 s-1)", 20))+

# annotate("text", x=1:4, y=3, label = c("a", "b", "b", "b"), size=5, family="open")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=16, family="mont", lineheight=0.8),

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=20, family="open", face="bold")

)

# GSW by date across relative humidity

ggplot(data = li\_data, aes(x=Date, y = gsw,

color = rh\_s)) +

geom\_jitter(width=1, size=2, alpha=1)+

scale\_color\_scico(begin=0.9, end=0, palette=a\_palette)+

# scale\_x\_discrete(guide=guide\_axis(check.overlap=TRUE))+

theme\_bw()+

ylab(str\_wrap("Stomatal Conductance (mol m-2 s-1)", 20))+

xlab("Days From Germination")+

labs(

tag="2023",

title=str\_wrap("Stomatal Conductance By Days From Germination Across Relative Humidity in Tomato", 50)

)+

guides(color=guide\_colorbar(title=str\_wrap("Relative Humidity", 10)))+

theme(

text = element\_text(size=16, family="mont"),

legend.title = element\_text(size=16, family="mont", face="bold"),

legend.text = element\_text(size=14, family="mont"),

legend.position="right",

legend.title.position = "top",

legend.key.height = unit(.4, "cm"),

legend.background = element\_rect(color=two\_colors[2], fill=NA,

linewidth=.5, linetype = 2),

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=20, family="open", face="bold", lineheight = .8)

)

# GSW by time across relative humidity

ggplot(data = li\_data, aes(x=Time, y = gsw,

color = rh\_s,

shape = Treatment)) +

geom\_jitter(width=1, size=2, alpha=1)+

scale\_color\_scico(begin=0.9, end=0, palette=a\_palette)+

scale\_shape\_manual(values=four\_shapes)+

scale\_x\_discrete(guide=guide\_axis(check.overlap=TRUE))+

theme\_bw()+

ylab(str\_wrap("Stomatal Conductance (mol m-2 s-1)", 20))+

xlab("Time")+

labs(

title=str\_wrap("Stomatal Conductance By Time Across Date in Tomato", 30)

)+

guides(color=guide\_legend(title=str\_wrap("Relative Humidity", 10)))+

theme(

text = element\_text(size=16, family="mont", lineheight = 1),

legend.title = element\_text(size=16, family="mont", face="bold"),

legend.text = element\_text(size=14, family="mont"),

legend.position="right",

legend.title.position = "top",

legend.key.height = unit(.3, "cm"),

legend.background = element\_rect(color=two\_colors[2], fill=NA,

linewidth=.5, linetype = 2),

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=20, family="open", face="bold", lineheight = .8)

)

# gsw by Minutes from Start

ggplot(data = li\_data, aes(x=MinutesFromStart, y = gsw, shape = Treatment, color = rh\_s)) +

geom\_point(size=3)+

scale\_color\_scico(begin=0.9, end=0, palette=a\_palette)+

theme\_bw()+

ylab(str\_wrap("Stomatal Conductance (mol m-2 s-1)", 20))+

xlab("Minutes From Start")+

labs(

title=str\_wrap("Stomatal Conductance by Minutes From Start Across Relative Humidity and Treatment in Tomato", 40)

)+

guides(color=guide\_colorbar(title=str\_wrap("Relative Humidity", 8)))+

theme(

text = element\_text(size=16, family="mont"),

legend.position="right",

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=20, family="open", face="bold", lineheight = .8)

)

## phiPS2

### because phiPS2 is a proportion [0:1], we don't have to create PDFs and CDFs or perform KS tests. For a proportion we can just logit transform it then use a linear model.

### check for heteroscedasticity

#### Pr(>F) of < 0.05 means that the value is homoscedastic

leveneTest(li\_data$PhiPS2~li\_data$Treatment)

###### Heteroscedastic by treatment.

### box plot of logit PhiPS2 by treatment

ggplot(data = li\_data, aes(x= Treatment, y = PhiPS2,

fill=Treatment, color=Treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=str\_wrap("PhiPS2 by Inoculation Treatment in Tomato", 30)

) +

ylab("PhiPS2")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

### logit PhiPS2 by time scatterplot

ggplot(data = li\_data, aes(x= MinutesFromStart, y = logitPS2, shape = Treatment, color=Qamb)) +

geom\_jitter( width=0, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_scico(begin=0.9, end=0, palette=a\_palette)+

# scale\_x\_discrete(guide=guide\_axis(check.overlap=TRUE))+

labs(

title=str\_wrap("Logit PhiPS2 by Minutes From Start and Inoculation Treatment in Tomato", 40)) +

ylab("PhiPS2")+

xlab("MinutesFromStart")+

theme\_bw()+

theme(

legend.position="right",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

## height

# height

multipdf\_plot(filter(h\_data, Height >0)$Height, 100, "all", a\_palette, "Height")

multipdf\_plot(filter(h\_data, Height >0)$Height, 100, "all", a\_palette, "Height")

multiks\_cont(filter(h\_data, Height >0)$Height, "all")

### box plot of height by treatment

ggplot(data = h\_data, aes(x= Treatment, y = Height,

fill=Treatment, color=Treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title="Height by Inoculation Treatment in Tomato"

) +

# annotate("text", x=1:4, y=35, label = c("a", "ab", "ab", "b"), size=6, family="open")+

ylab("Height (cm)")+

xlab("Treatment")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

# height scatterplot

ggplot(data=h\_data, aes(x=Date, y=Height, color=Treatment))+

geom\_jitter(size=2)+

scale\_color\_manual(values = four\_colors)+

labs(

title="Height by Date and Treatment in Tomato"

)+

ylab("Height (cm)")+

theme\_bw()+

theme(

legend.position="right",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

# RS\_length

multiPDF\_plot(DS\_data$RS\_Length, 100, "all", a\_palette, "R:S Length")

multiCDF\_plot(DS\_data$RS\_Length, 100, "all", a\_palette, "R:S Length")

multiKS\_cont(DS\_data$RS\_Length, "all")

ggplot(data=DS\_data, aes(x=Treatment, y=RS\_Length, color=Treatment))+

geom\_violin()+

geom\_boxplot(width=0.2)+

geom\_jitter()+

labs(

title="Root:Shoot Length by Treatment"

)

ggplot(data=DS\_data, aes(x=Treatment, y=RS\_Mass, color=Treatment))+

geom\_violin()+

geom\_boxplot(width=0.2)+

geom\_jitter()+

labs(

title="Root:Shoot Mass by Treatment"

)

**III.D.4. Models and Post Hoc Tests**

" ## gsw could be normal or gamma

model\_gsw<- lm(log(gsw) ~ Treatment + rh\_s, data = li\_data)

AIC(model\_gsw)

summary(model\_gsw)

model\_gsw2<- lm(log(gsw) ~ Treatment + rh\_s + MinutesFromStart + DaysFromGermination, data = li\_data)

AIC(model\_gsw2)

summary(model\_gsw2)

comps <- glht(model\_gsw2, linfct = mcp(Treatment = "Tukey"))

mod\_gsw\_letters <- cld(comps)$mcletters$Letters

print(mod\_gsw\_letters)

predict\_plot(model\_gsw, li\_data, gsw, rh\_s, Treatment,

correction = "exponential") +

labs(title = NULL, subtitle = NULL) +

ylab(str\_wrap("Stomatal Conductance (mol m-2 s-1)", 20))+

xlab("Relative Humidity (%)")+

theme(

legend.position="right",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("tim\_gsw\_pred.svg", path = "C:/Github/Thesis/figures/TIM",

width = 10, height = 6)

ggsave("tim\_gsw\_pred.png", path = "C:/Github/Thesis/figures/TIM",

width = 10, height = 6)

dfpca <- pca\_data(data\_tim\_fluoro, data\_tim\_fluoro[,c(10:14)])

pca\_plot(data\_tim\_fluoro$Treatment, data\_tim\_fluoro[,c(10:14)])+

xlim(-0.8, 0.8)

dfpca2 <- pca\_data(li\_data, li\_data[,c(9:12, 14, 15, 20)])

pca\_plot(li\_data$Treatment, li\_data[,c(9:12, 14, 15, 20)])+

xlim(-0.8, 0.8)

modgswpc <- lm(log(gsw) ~ Treatment + PC1 + PC2, data = dfpca)

AIC(modgswpc)

summary(modgswpc)

modgswpc2 <- lm(log(gsw) ~ Treatment + PC1 + PC2, data = dfpca2)

AIC(modgswpc2)

summary(modgswpc2)

modps2pc <- lm(logit(PhiPS2) ~ Treatment + PC1 + PC2, data = dfpca)

AIC(modps2pc)

summary(modps2pc)

comps <- glht(modps2pc, linfct = mcp(Treatment = "Tukey"))

mod\_ps2pc\_letters <- cld(comps)$mcletters$Letters

print(mod\_ps2pc\_letters)

modps2pc2 <- lm(logit(PhiPS2) ~ Treatment + PC1, data = dfpca)

predict\_plot(modps2pc2, dfpca, PhiPS2, PC1, Treatment,

correction = "logit") +

labs(title = NULL, subtitle = NULL) +

ylab("PhiPS2")+

xlab("PC1")+

theme(

legend.position="right",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("tim\_ps2\_pred.svg", path = "C:/Github/Thesis/figures/TIM",

width = 10, height = 6)

ggsave("tim\_ps2\_pred.png", path = "C:/Github/Thesis/figures/TIM",

width = 10, height = 6)

model\_fl\_phi2<- lm(LogitPhiPS2 ~ Treatment + DaysFromGermination, data = data\_tim\_fluoro)

AIC(model\_fl\_phi2)

summary(model\_fl\_phi2)

model\_h <- lm(

log(Height) ~ Treatment + DaysFromGermination,

data = h\_data)

summary(model\_h)

confint(model\_h)

predict\_plot(model\_h, h\_data, Height, DaysFromGermination, Treatment, correction = "exponential") +

labs(title = NULL, subtitle = NULL)+

ylab("Height (cm)")+

xlab("Days From Germination")+

theme(

legend.position="right",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold")

)

ggsave("tim\_height\_pred.svg", height = 6, width = 10, path = "C:/Github/Thesis/figures/TIM")

ggsave("tim\_height\_pred.png", height = 6, width = 10, path = "C:/Github/Thesis/figures/TIM")

comps <- glht(model\_h, linfct = mcp(Treatment = "Tukey"))

cld(comps)

model\_h2 <- glm(Height ~ Treatment + DaysFromGermination, data = h\_data,

family = Gamma(link = "log"))

summary(model\_h2)

glm\_pseudor2(model\_h2)

predict\_plot(model\_h2, h\_data, Height, DaysFromGermination, Treatment) +

labs(title = NULL, subtitle = NULL)

model\_RSL <- lm(logit(RS\_Length) ~ Treatment, data = DS\_data)

AIC(model\_RSL)

summary(model\_RSL)

comps <- glht(model\_RSL, linfct = mcp(Treatment = "Tukey"))

mod\_RSL\_letters <- cld(comps)$mcletters$Letters

print(mod\_RSL\_letters)

ggplot(data = DS\_data, aes(x= Treatment, y = RS\_Length,

fill=Treatment, color=Treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Root : Shoot Length")+

xlab("Treatment")+

annotate("text", x=1:4, y=1.5, label = mod\_RSL\_letters, size=10, family="open")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("tim\_RSL\_annotated.svg", path = "C:/Github/Thesis/figures/TIM",

width = 10, height = 6)

ggsave("tim\_RSL\_annotated.png", path = "C:/Github/Thesis/figures/TIM",

width = 10, height = 6)

model\_RSM <- lm(logit(RS\_Mass) ~ Treatment, data = DS\_data)

AIC(model\_RSM)

summary(model\_RSM)

comps <- glht(model\_RSM, linfct = mcp(Treatment = "Tukey"))

mod\_RSM\_letters <- cld(comps)$mcletters$Letters

print(mod\_RSM\_letters)

ggplot(data = DS\_data, aes(x= Treatment, y = RS\_Mass,

fill=Treatment, color=Treatment)) +

geom\_boxplot(alpha=.5, width=0.5, outliers = FALSE)+

geom\_jitter( width=.15, height=0)+

scale\_fill\_manual(values=four\_colors)+

scale\_color\_manual(values=four\_colors)+

labs(

title=NULL,

subtitle = NULL

) +

ylab("Root : Shoot Mass")+

xlab("Treatment")+

annotate("text", x=1:4, y=0.75, label = mod\_RSM\_letters, size=10, family="open")+

theme\_bw()+

theme(

legend.position="none",

text = element\_text(size=subtitle\_size, family="mont", lineheight=0.8),

axis.title = element\_text(size=subtitle\_size, family = "mont", face= "bold"),

title = element\_text(size=title\_size, family="open", face="bold")

)

ggsave("tim\_RSM\_annotated.svg", path = "C:/Github/Thesis/figures/TIM",

width = 10, height = 6)

ggsave("tim\_RSM\_annotated.png", path = "C:/Github/Thesis/figures/TIM",

width = 10, height = 6)

**III.E. Bead Breakdown Code**

**III.E.1. Setup**

bb\_data\_file <- "C:/Github/Thesis/data/BB/BB\_4142025.csv"

**III.E.2. Clean Data**

bb\_data <- read.csv(bb\_data\_file)

bbdat <- na.omit(bb\_data) %>%

mutate(mass\_diff = initial\_weight - end\_weight,

pdiff = mass\_diff/initial\_weight,

ipdiff = pdiff \* -1)

bbnoleaks <- filter(bbdat, X != "leak") %>%

filter(pdiff > 0) %>%

mutate(

logitpdiff = logit(pdiff),

ilpdiff = logitpdiff \* -1)

bbsum <- bbnoleaks %>%

group\_by(polymer, elapsed\_weeks) %>%

summarise\_at(vars(initial\_weight, end\_weight, pdiff), list(mean = mean)) %>%

ungroup()

**III.E.3. Exploratory Analysis**

## EDA

multipdf\_plot(bbnoleaks$end\_weight)

multiks\_cont(bbnoleaks$end\_weight)

ggplot(bbnoleaks, aes(x=elapsed\_weeks, y=ilpdiff, color = polymer))+

geom\_jitter(width=0.3, height = 0, size = 3)+

scale\_color\_scico\_d(begin = 0.8, end = 0.3, palette = "oslo")+

theme\_bw()

ggplot(bbnoleaks, aes(x=elapsed\_weeks, y=end\_weight, color = polymer))+

geom\_jitter(width=0.3, height = 0, size = 3)+

scale\_color\_scico\_d(begin = 0.8, end = 0.3, palette = "oslo")+

theme\_bw()

ggplot(bbnoleaks, aes(x=elapsed\_weeks, y=pdiff, color = medium))+

geom\_jitter(width=0.3, height = 0, size = 3)+

scale\_color\_scico\_d(begin = 0.8, end = 0.3, palette = "oslo")+

theme\_bw()

ggplot(bbnoleaks, aes(x=elapsed\_weeks, y=pdiff, color = inoculant))+

geom\_jitter(width=0.3, height = 0, size = 3)+

scale\_color\_scico\_d(begin = 0.8, end = 0.3, palette = "oslo")+

theme\_bw()

**III.E.4. Models and Post Hoc Tests**

mod1 <- lm(logit(pdiff) ~ elapsed\_weeks + polymer, data = bbnoleaks)

summary(mod1)

AIC(mod1)

predict\_plot(mod1, bbnoleaks, pdiff, elapsed\_weeks, polymer, correction = "logit")+

ylab("Proportion Change in Mass")+

xlab("Elapsed Weeks")+

labs(title = NULL,

subtitle = NULL)+

guides(

fill = guide\_legend(title="Polymer"),

color= guide\_legend(title="Polymer")

)+

theme(

text = element\_text(size=14, family="mont", lineheight=0.8),

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=18, family="open", face="bold")

)

ggsave("BB\_polymer\_pred.svg", path = "C:/Github/Thesis/figures/BB")

ggsave("BB\_polymer\_pred.png", path = "C:/Github/Thesis/figures/BB")

mod11 <- lm(ilpdiff ~ elapsed\_weeks + polymer, data = bbnoleaks)

summary(mod11)

AIC(mod11)

predict\_plot(mod11, bbnoleaks, ilpdiff, elapsed\_weeks, polymer)+

ylab("Mass (g)")+

xlab("Elapsed Weeks")+

labs(title = NULL,

subtitle = NULL)+

guides(

fill = guide\_legend(title="Polymer"),

color= guide\_legend(title="Polymer")

)+

theme(

text = element\_text(size=14, family="mont", lineheight=0.8),

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=18, family="open", face="bold")

)

mod2 <- lm(logit(pdiff) ~ elapsed\_weeks + medium, data = bbnoleaks)

summary(mod2)

AIC(mod2)

predict\_plot(mod2, bbnoleaks, pdiff, elapsed\_weeks, medium, correction = "logit")+

ylab("Change in Mass")+

xlab("Elapsed Weeks")+

labs(title = NULL,

subtitle = NULL)+

guides(

fill = guide\_legend(title = "Medium"),

color = guide\_legend(title = "Medium")

)+

theme(

text = element\_text(size=14, family="mont", lineheight=0.8),

axis.title = element\_text(size=16, family = "mont", face= "bold"),

title = element\_text(size=18, family="open", face="bold")

)

ggsave("BB\_medium\_pred.svg", path = "C:/Github/Thesis/figures/BB")

# inverse logit prediction plot for pdiff

## get group data ready

groups <- sort(unique(bbnoleaks$polymer))

ngroups <- length(groups)

## get predictor range for each group

agg <- stats::aggregate(bbnoleaks$elapsed\_weeks ~ bbnoleaks$polymer,

data = bbnoleaks, range)

dx\_pvar <- data.frame(pvar = numeric(0))

for (i in 1:ngroups) {

tpvar <- data.frame(pvar = seq(agg[[2]][i, 1], agg[[2]][i, 2],

length = 50))

dx\_pvar <- rbind(dx\_pvar, tpvar)

}

dx <- data.frame(group = rep(agg[[1]], each = 50),

pvar = dx\_pvar)

colnames(dx) <- c("polymer", "elapsed\_weeks")

## model

mod <- lm(logit(pdiff) ~ elapsed\_weeks + polymer, data = bbnoleaks)

## make prediction

pred <- stats::predict(mod, newdata = dx,

se.fit = TRUE, type = "response")

dx$mn <- stats::plogis(stats::qnorm(0.5, pred$fit, pred$se.fit))

dx$lo <- stats::plogis(stats::qnorm(0.025, pred$fit, pred$se.fit))

dx$up <- stats::plogis(stats::qnorm(0.975, pred$fit, pred$se.fit))

dx$mn <- dx$mn \* -100

dx$lo <- dx$lo \* -100

dx$up <- dx$up \* -100

## initialize plot

p <- ggplot2::ggplot() +

ggplot2::geom\_point(data = bbnoleaks, ggplot2::aes(x = elapsed\_weeks,

y = pdiff \* -100,

color = polymer))

## loop through treatments

for (g in 1:ngroups) {

flag <- which(dx$polymer == groups[g])

tdx <- dx[flag, ]

p <- p +

ggplot2::geom\_line(data = tdx, ggplot2::aes(x = elapsed\_weeks, y = lo,

color = polymer),

linewidth = 1, show.legend = FALSE) +

ggplot2::geom\_line(data = tdx, ggplot2::aes(x = elapsed\_weeks, y = mn,

color = polymer),

linewidth = 2, show.legend = FALSE) +

ggplot2::geom\_line(data = tdx, ggplot2::aes(x = elapsed\_weeks, y = up,

color = polymer),

linewidth = 1, show.legend = FALSE) +

ggplot2::geom\_ribbon(data = tdx, ggplot2::aes(x = elapsed\_weeks,

ymin = lo, ymax = up,

fill = polymer),

alpha = 0.5)

}

p <- p +

scico::scale\_color\_scico\_d(begin = 0.8, end = 0.3, palette = "oslo") +

scico::scale\_fill\_scico\_d(begin = 0.8, end = 0.3, palette = "oslo")

### make the plot look good (group agnostic)

p <- p +

ggplot2::theme\_bw() +

ylab("Change in Mass (%)") +

xlab("Elapsed Weeks") +

guides(

fill = guide\_legend(title="Polymer"),

color= guide\_legend(title="Polymer")

)+

ggplot2::theme(

text = ggplot2::element\_text(size = 12),

axis.title = ggplot2::element\_text(size = 14, face = "bold"),

title = ggplot2::element\_text(size = 16, face = "bold"),

plot.subtitle = ggplot2::element\_text(size = 14, face = "italic")

)

p

ggsave("BB\_polymer\_pred.svg", path = "C:/Github/Thesis/figures/BB")

ggsave("BB\_polymer\_pred.png", path = "C:/Github/Thesis/figures/BB")