Analysis Tutorial Overview

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1. **Modified Title**

Visualizing and significance testing on biomass-reduction effects of Cyanobacteria Microcystis (MC) by Amoeba Vanella’s grazing behavior

1. **Modified Research question**

A general grazing effect by freshwater Amoeba had been verified by previous studies (Van Wichelen et al., 2010), is this biomass-reductive effect determined by the Amoeba density?

1. **Modified Objective**

Visualizing the MC growth curve by measuring pigments in 2 days’ interval, the biomass-reductive effect is defined as the gaps between test group lines and control group lines (Weger et al., 2024). Applying statistical analyses to test the significant difference between the treatments of 4 Amoeba density settings (0, 1X, 5X, 10X) (Xinyao et al., 2006).

1. **Overview**

2 previously tested “poor” food source MC strains are included (LE21 22.1 ISO2 MC-, LE21 52.2 ISO3 MC+). The Amoeba Vannella A22.3-1, 1/7/25 was separated to 1X original stock solution, 5X diluted, 10X diluted and standardized to 4 mL. 4 mL of each MC was incubated with each dilution of Amoeba and added up to 40 mL with 32 mL 1X BG-11 media (36 mL for control groups without Amoeba). The workflow could be divided into 6 general steps after the data collection as: step 1) Install/load necessary R packages (ggplot2, readxl, tidyverse, ggthemes, scales, dplyr, writexl, multicompView); 2) Add averages and standard deviations to raw data set mainly by “group\_by() + mean() + sd()”; 3) Visualization of growth curve with new data set mainly by “ggplot() + facet\_grid() + geom\_errorbar() + labs()”; 4) Process One-way ANOVA mainly by “aov\_model <- aov(results ~ treatment, data = df)”; 5) Conduct Tuckey HSD post-hoc test mainly by “tukey <- TukeyHSD(model)”; 6) Adding daily variations to data set mainly by “mutate(Variation = results - lag(results))”.

The growth curves were able to show the distinctive responses of Amoeba density dependent species (22.1 -) and Amoeba density independent inhibitive species (52.2 +) (Urrutia-Cordero, P., et al., 2013; Van Wichelen et al., 2012).). The significance tests identified 1X and 5X treatments to 22.1 – were significantly different from control group (C), while 10X was not. And there is no significance in all the groups in 52.2 +. These results are in accordance with visual inspection on biomass on Day 8 and the growth curve lines. Thus, this workflow and R codes utilized could be regarded as well-designed and reliable, which could be further modified by changing the testing variables to fit in more aims (Van Wichelen et al., 2016)..

1. **References**

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