Plasma Testing on Salmon Tissue

*uL = Microliters

Day 0 - Preparation Day

1. Prepared E.coli Culture
    a. 10mL Tryptic Soy Broth
    b. 100uL E.coli Stock Solution
    c. 100uL Rifampicin

2. Prepared PBS (Phosphate Buffered Solution)
3. Prepared TSA Plates

Day 1 - Experiment Day

1. Serial Dilution by 5 log reductions
    a. 500uL was transferred from E.coli Culture to a 4500uL tube
    b. Vortexed to mix
    c. Created 5000uL solution that is 1/10 concentrated from E.coli culture
    d. Repeated 4 more times (Total: 5) to create solution with 10^{-5} concentration
2. Salmon Preparation and Inoculation
    a. Prepackaged Salmon Filet was collected and cut into about 1-inch cubed pieces
    b. Each piece was placed on a separate labeled petri dish
    c. Each salmon piece was inoculated with 250uL diluted E.coli culture from step 1
    d. The plates were set to dry for about 1 hour
3. Plasma Treatment
    a. 1min, 3min, and 5 min durations, each with 3 repeats, were used to treat salmon samples under the plasma chamber, Oxyfog.
    b. Details of Plasma Treatment are as follows:
        i. Plates were placed inside the chamber. Seal was then closed
        ii. Recirculation Fan, both DBD Plasma Generators, and 1 Nebulizer were all turned on for the tested duration
        iii. After treatment, all settings were turned off and Heat + Fan setting was turned on for 1 minute with the chamber still sealed. (Intention of this step was to “disable” reactive species present before opening chamber)
        iv. Seal was then opened and plates were taken out of the chamberz
        v. Heat + Fan and Recirculation Fan were then turned on for 1 minute, with chamber open to reset chamber conditions
        vi. Repeated for other tests
4. Liquid Extraction
    a. Samples were then placed in separate centrifuge tubes with 10mL of PBS
    b. Tubes were vortexed until liquid became white.
    c. Liquid was then filtered through coffee filter paper to extract near-pure liquid
5. Plating, Incubation, and Colony Counting

a. 300uL of liquid extract from step 4 was plated on TSA plate. Repeated for all tests
b. Incubated overnight at 37 degrees Celsius
c. After incubation, number of colonies were counted on each plate for analysis