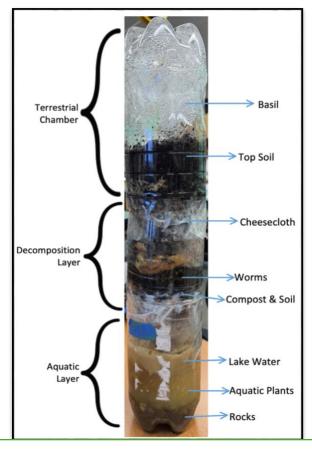
Jai Bindlish James Boyd APES P3 October 29, 2024

# Guided Question: How does organism death affect an ecosystem?



# <u>Claim:</u> Overview of Lab:

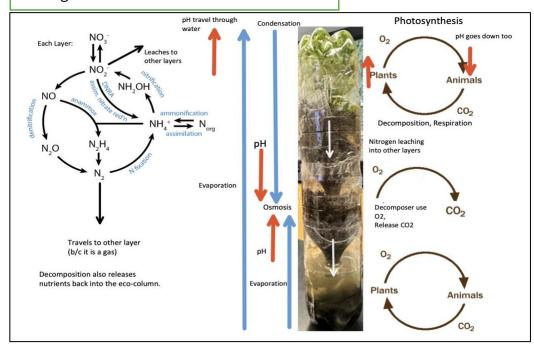
The building of the Eco-column begun on 9/11/24. The structure is made up of three 2-liter bottles to compose different chambers, connected with cheesecloth to maintain water flow. The eco-column contains 3 such layers, Terrestrial, Decomposition, and Aquatic. Terrestrial Layer: contains Topsoil (no added nutrients), basil seeds (for basil plants).

Decomposition Layer: contains household compost (to gradually decompose), soil.

Aquatic Layer: contains lake water (from Lake Sammamish), aquatic rocks, and Lemon Bacopa (aquatic plant).

Measurements were on 9/13, 9/27, 10/11, and 10/24, on 14-day intervals between measurements. The measurements taken for each layer were: Terrestrial – pH, Nitrate, Nitrite, Decomposition- pH, Nitrate, Nitrite, and Aquatic Layer – Dissolved Oxygen, Hardness, Nitrate, Nitrite, Carbonate. Note: Nitrate/Nitrite measured in PPM, Dissolved Oxygen, Hardness, and Carbonate measured in mg/L. There are many ways that energy and molecules flow up the eco column. Capillary Action: the water in the aquatic layer "climbs up" the integrated cheesecloth, so that the autotrophs in the decomposition and terrestrial chambers can get water and grow. The nitrites and nitrates move upward in 3 ways: along with the water, nitrification in the decomposition layer, and plant uptake. Nitrates and nitrites move down by leaching through the soil in the terrestrial and decomposition layers.

## Diagram of Eco-column with labels 9/13



- Diagram of interactions throughout the entire ecocolumn of biotic and abiotic factors and processes including nitrates, nitrites, pH, DO, and processes involving carbon dioxide and oxygen.

These interactions between layers play a massive role in the cycling of nutrients, energy flow, atmospheric composition, and eventually plant and animal life.

There are many ways water especially travels through the system: capillary action with the cheesecloth, water osmosis, and the water cycle evaporation and condensation.

# Terrestrial Layer:

The terrestrial layer in our eco-column was constructed using topsoil, planted with basil seeds and hosed earthworms. This chamber is responsible for nutrient cycling, oxygen production, and potential water filtration for the layers below. **Topsoil** provides rich soil filled with organic matter, minerals, and beneficial microbes. We chose it for its ability to retain moisture, maintain pH close to neutral, and house soil-residing organisms. We planted **Basil seeds** due to their quick germination, ideal for observing plant growth and interactions within a controlled system in a short amount of time. As the basil develops, it absorbs nutrients from the soil, including nitrogen and phosphorus. Through photosynthesis, the basil plants produce oxygen and release it into the system, benefiting organisms in other layers. **Worms** help aerate the soil as they burrow, which improves root oxygen access and promotes microbial activity essential for nutrient cycling. Their digestion process creates nutrient-rich castings, further enriching and maintaining the soil. By day 119, the basil growth will have slowed down, and the topsoil would be completely nutrient deficient.

Time Lapse

9/13

9/27

10/11

10/24

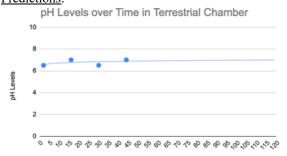
Time Lapse of Terrestrial Layer – Photos taken by Jai Bindlish, JD Kim Predictions:

9/13 – Minimal Growth, Condensation of Water visible on edges of bottle. Soil is very wet.

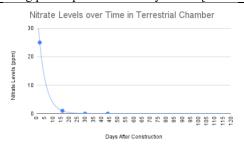
9/27 – Basil Plants have germinated, grown to about 2 inches high. Bottle edges are mostly clear of water.

10/11 – edges of bottle are cloudy, and plants have grown to about 4 inches high. Roots of plants are visible in soil.

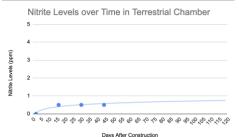
10/24 – plant is fully grown to top of eco-column, roots are very visible and constrained in space. Soil looks less moisturized, edges are clear.



pH levels in the terrestrial chamber of the eco-column found using pH strips and soil analyzation. [data table and graph]



Nitrate levels in terrestrial chamber found using soil analyzation.



Nitrite levels in terrestrial chamber found using soil analyzation.

Date	9/13	9/27	10/11	10/24	1/8*
Nitrate (ppm)	25	1	0	0	0
Nitrite (ppm)	0	0.5	0.5	0.5	0.48-0.53

In the terrestrial layer of the eco-column, pH ranges slightly, but it generally stabilizes near a neutral level at about 7. Such minor changes are normal in ecosystems since organic matter decomposes, plants take up nutrients, and the soil microbes process the compound. pH remaining within this range is optimal for healthy plant growth and microbial activity since nutrient availability can be maintained. The **pHs are predicted** to fall between 6.7 and 7.3, which suggests that the system will have stability in pH. This predicted pH is well within the range of tolerance for the basil plants – between 6.0 and 7.5.

Date	9/13	9/27	10/11	10/24	1/8*
pН	6.5	7	6.5	7	6.7-7.3

In the terrestrial layer data, levels of nitrate and nitrite exhibit stages in the nitrogen cycle: Nitrate: High initial values (25 ppm on 09/13) could be because of ample sources of nitrogen, decomposing organic matter or nutrient input in the soil. Nitrate decreases by 09/27 and goes to zero in the continuing measurements (10/11 and 10/24). This rapid decrease would indicate that either plants (basil) are using the nitrates for growth, or nitrates are leaving the terrestrial layer, leaching down to other layers. **Nitrate Prediction:** 0ppm – This means the basil is absorbing all the Nitrate needed for its growth, in range of tolerance from 0-20. Nitrite: Nitrite usually would appear just for a short period in the nitrogen cycle, since ammonia NH3 is transformed into nitrate by bacteria explaining the initial 9/13 deficiency. Certain imbalance in the microbial population in this soil responsible for the oxidation of ammonia to nitrate causes the increase in levels after 9/13.. The nitrifying bacteria normally convert nitrite to nitrate very fast. The build-up in this case would therefore mean either that there aren't enough nitrite-oxidizing bacteria for nitrite being produced. Worm death could be causing an uptick in Nitrite as decomposers die, they release natural minerals back into the soil. **Nitrite Prediction:** 0.48-0.53ppm – meaning that the nitrite levels are minimal, but enough to support the minimal plants that are present. This is in the range of tolerance of 0-1ppm.

# **Decomposition Layer:**

The decomposition layer in the eco-column is made with topsoil and household compost-like banana peels-and worms. Each plays a different role in the function of the ecosystem. **Topsoil** provides structure and retains moisture and, thus, a stable environment for microbial life to thrive. **Household compost**, like banana peels, contains many nutrients like nitrogen, potassium, and phosphorus that are released into the soil as the compost breaks down. This is all hydrated with lake water from Lake Sammamish. Finally, **worms** are essential decomposers, breaking down organic matter into nutrient-rich castings while aerating the soil through their burrowing activities. Together, they facilitate the decomposition and cycling of nutrients for enrichment of the soil for plant support in the ecocolumn. By 119<sup>th</sup> day, the increased nitrate and nitrite levels will result in leaching and eutrophication inside of the aquatic chamber.

Time Lapse

9/13

9/27

10/11

10/24

Time Lapse of Decomposition Layer - Photos taken by Jai Bindlish, JD Kim

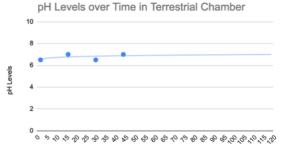
9/13 – chunks of decomposition material still visible, mold is apparent. No water droplets on sides of bottle.

9/27 – chunks are still visible, but some have been buried/broken down.

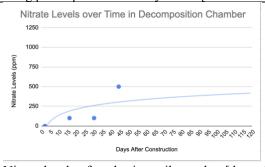
10/11 – layer is much darker, materials are more broken down and soil level seems to have leveled down.

**10/24** – soil is quite dry, most materials are fully broken down, many different solids are visible in soil.

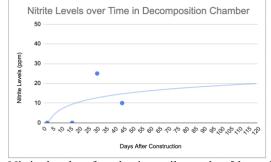
#### Graphs:



pH levels in the decomposition chamber of the eco-column, found using pH strips and soil analyzation. [data table and graph]



Nitrate levels - found using soil samples. [data table and graph]



Nitrite levels – found using soil samples. [data table and graph]

The **pH** levels in this layer were variant, ranging from around 5.5 to 7.5. This is because of the dynamic processes of organic matter getting broken down, where organic acids are released from the organic matter. Different Microbes also affect the pH of the layer, depending on what they consume. **pH prediction**: Although variant, the pH levels on day 119 will be 6.8, according to the line of best fit. This is slightly acidic but makes sense due to the composition of the layer (all different compostable materials). This is in the range of tolerance of 5.5-7.0.

Date	9/13	9/27	10/11	10/24	1/8*
pН	6	7.5	7	5.5	6.6-7.2

Nitrate Levels, contrasted to the terrestrial level, are looking very high and trending up, with the most recent reading at 500ppm. The organic nutrients that are being released into the soil as decomposers (like worms) are breaking them down include Nitrate, increasing the levels. Nitrate Prediction: the line of best fit reflects that the nitrate will be 380-420, outside the range of tolerance for bacteria of 0-20ppm. The nitrate levels going up like this reflects the effects of death on an ecosystem, where when the dead bodies are disposed of by decomposers, they release organic nutrients such as nitrates.

Nitrite Levels are also very high compared to the terrestrial level, with variance in values from 0 to 25ppm. Nitrite Prediction: The line of best fit suggests that the Nitrite Levels will be at about 19-21ppm on day 119, within the range of tolerance of 1-50. This further proves the point of how dead organic material will affect an ecosystem by changing chemical values.

All this excess Nitrate/Nitrite can leach off into other layers, which might be dangerous for the organisms that live there.

Date	9/13	9/27	10/11	10/24	1/8*
Nitrate (ppm)	0	100	100	500	380-420
Nitrite (ppm)	0	0	25	10	19-21

## Aquatic Layer:

This aquatic layer of the eco-column consists of Lemon Bacopa, 1L of Lake Sammamish water, and aquatic rocks. Lemon Bacopa is a robust aquatic plant that utilizes nitrates and other nutrients through active photosynthesis to keep the nutrients down and maintain relatively good water quality. Lake water contributes native microbes to the biodiversity of the system, hence assisting natural processes in water self-purification, since these microbes contribute to the breakdown of organic matter. Pebbles in the aquatic environment provide surfaces upon which beneficial bacteria grow, hence helping in nitrogen cycling by converting ammonia into less toxic forms, such as nitrite and nitrate, usable by plants. These put together create a balanced aquatic environment that supports plants and microbial life within the eco-column for nutrient cycling and oxygen production. In day 119, the aquatic layer will become anaerobic, leveling off at a low value of 5mg/L, due to eutrophication (over concentration of nutrients) causing algal blooms. This will affect the aquatic and the rest of the layers in terms of how much oxygen carried up, effecting organism respiration and plant growth.

Time Lapse

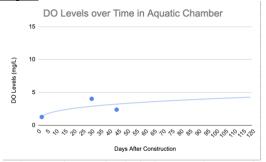
9/13 – tan water, cloudy, rocks and plant are visible. No condensation or water droplets on side are visible.

9/27 – relatively more clear water, plant is visible but looks like floating, condensation is visible.

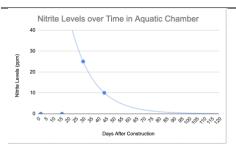
10/11 – water looks darker and dark, nothing much visible

**10/24** – water is black, rocks and plant aren't visible. Condensation is visible at a small level.

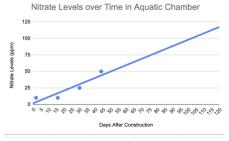
Time Lapse of Aquatic Layer – Photos taken by Jai Bindlish, JD Kim <u>Graphs:</u>



Dissolved Oxygen in Aquatic Chamber Note: Day 16 data excluded due to measuring error.



Nitrite level in aquatic chamber. Found using strips.



Nitrate level in aquatic chamber. Found using strips.

**Dissolved Oxygen** levels have been consistently low in the aquatic chamber. The last recorded value was 2.38mg/L, which is much less than the optimal range of the aquatic plant Lemon Bacopa (6.5-8mg/L). The DO has been consistently under this range, suggesting that the plant might have died due to unhabitable conditions. **Predicted DO** levels based on the line of best fit is 4.37-4.83mg/L, lower than the optimal range of 5-8mg/L. The dissolved oxygen is very low due to a process called Eutrophication – an excess of Nitrogen and Phosphorus among other nutrients, leading to rapid algae growth and eventually ending in oxygen levels being depleted. This also led to the water looking black, it was full of dead organic mass.

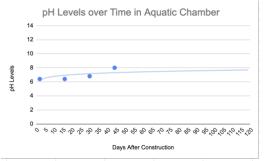
Date	9/13	9/27	10/11	10/24	1/8*
DO (mg/L)	1.27	10.08	4.03	2.38	4.37-4.83

**Nitrite Levels**: the nitrite levels are trending down as time goes on. Day 2 and day 16 both had nitrite levels of 0, then a spike occurred at day 30, then it fell at day 45. The **predicted Nitrite** level for day 119 is 0ppm, which is fine for the plant (as Nitrite is toxic).

Nitrate Levels: the nitrate levels are rapidly increasing as time goes on. The last measurement was at 50ppm. This is mostly due to nitrate leaking from the decomposition layer into the aquatic layer. The **predicted Nitrate** level for day 119 is about 113ppm, way out of the range of 5-20ppm.

The effect of this excess nitrogen in the Aquatic chamber will cause Eutrophication, which is explained in the Dissolved Oxygen graph explanation. Overall, the nitrogen leaks from the decomposition layer, causes access in the Aquatic layer, leading to Eutrophication, which reduces the DO of the water. Organisms that cannot take that reduction of DO will die, and the organic death material is what is turning the water black.

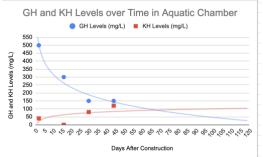
Date	9/13	9/27	10/11	10/24	1/8*
Nitrite (ppm)	0	0	25	10	0
Nitrate (ppm)	10	10	25	50	107-119



**pH Levels:** the pH levels maintain at about 7.5, with varying values due to different microbe activity. The **predicted pH** in the aquatic chamber is 7.9. This is slightly basic, which might affect the organisms that live in this chamber. This is slightly more basic than the aquatic plant Lemon Bacopa prefers (6-7.5).

Date	9/13	9/27	10/11	10/24	1/8*
pН	6.4	6.4	6.8	8	7.5-8.3

pH levels in the aquatic chamber, found using strips



Hardness and Carbonate Levels, found using strips.

**Hardness Levels:** the hardness levels go down as time goes on. The optimal range of hardness for Lemon Bacopa is 36-268.5 mg/L. The first measuring had a value out of range, but the next few were in optimal range. The **predicted Hardness** is about 40ppm, which is in optimal range.

**Carbonate Levels**: carbonate levels go up as time goes on. The **predicted Carbonate** levels for day 119 is 100 mg/L which is in range of tolerance of 71-179ppm. Decomposition of organic matter releases CO2, which will increase the carbonate levels in the ecosystem.

Date	9/13	9/27	10/11	10/24	1/8*
Carbonate (KH)	40	0	80	180	95-105
Hardness (mg/L)	500	300	150	150	38-42

#### Works Cited

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Aloi, P. (2024, April 1). What is topsoil and what are its benefits?. The Spruce. <a href="https://www.thespruce.com/what-is-topsoil-5121325">https://www.thespruce.com/what-is-topsoil-5121325</a> Education, U. C. for S. (n.d.). Center for Science Education. Biogeochemical Cycles | Center for Science Education. <a href="https://scied.ucar.edu/learning-zone/earth-system/biogeochemical-cycles">https://scied.ucar.edu/learning-zone/earth-system/biogeochemical-cycles</a>

Fisheries, N. (n.d.). *Understanding ocean acidification*. <a href="https://www.fisheries.noaa.gov/insight/understanding-ocean-acidification">https://www.fisheries.noaa.gov/insight/understanding-ocean-acidification</a> Schuh, A. M. (n.d.). *Growing basil in Home Gardens*. UMN Extension. <a href="https://extension.umn.edu/vegetables/growing-basil">https://extension.umn.edu/vegetables/growing-basil</a>

### **Evidence:**

# Raw Data:

Raw Data: Terrestrial Layer Nitrite, Nitrate, and pH

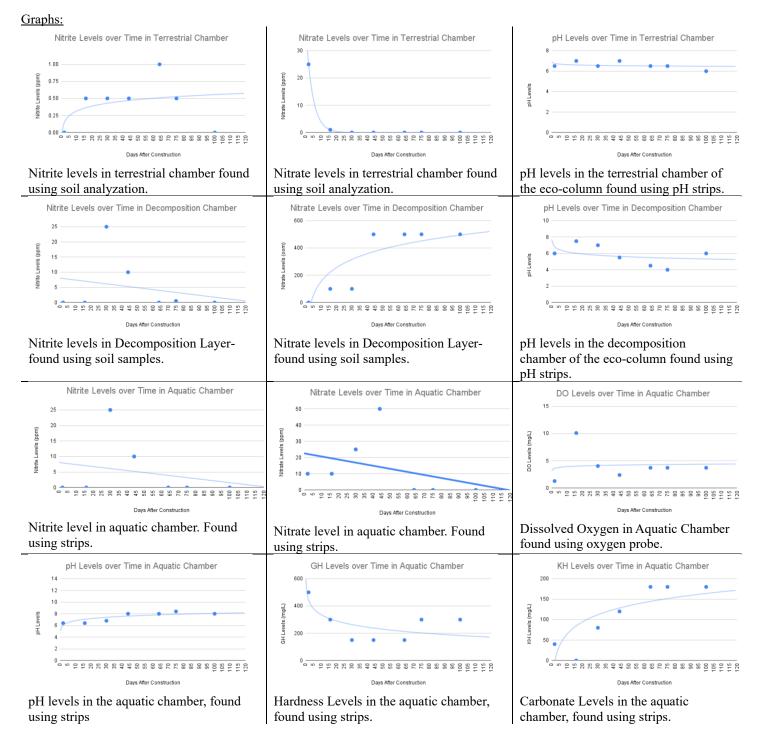
Date	Day #	Nitrite (ppm)	Nitrate (ppm)	pН
9/13	2	0	25	6.5
9/27	16	0.5	1	7
10/11	30	0.5	0	6.5
10/24	44	0.5	0	7
11/14	64	1	0	6.5
11/25	75	0.5	0	6.5
12/20	100	0	0	6
1/08**	119	0	0	6.2-6.6

Raw Data: Decomposition Layer Nitrite, Nitrate, and pH

Date	Day #	Nitrite (ppm)	Nitrate (ppm)	
9/13	2	0 ppm	0 ppm	6
9/27	16	0 ppm	100 ppm	7.5
10/11	30	25 ppm	100 ppm	7
10/24	44	10 ppm	500 ppm	5.5
11/14	64	0 ppm	500 ppm	4.5
11/25	75	0.5 ppm	500 ppm	4
12/20	100	0 ppm	500 ppm	4.5
1/08**	119	0	499-551	4.99-5.51

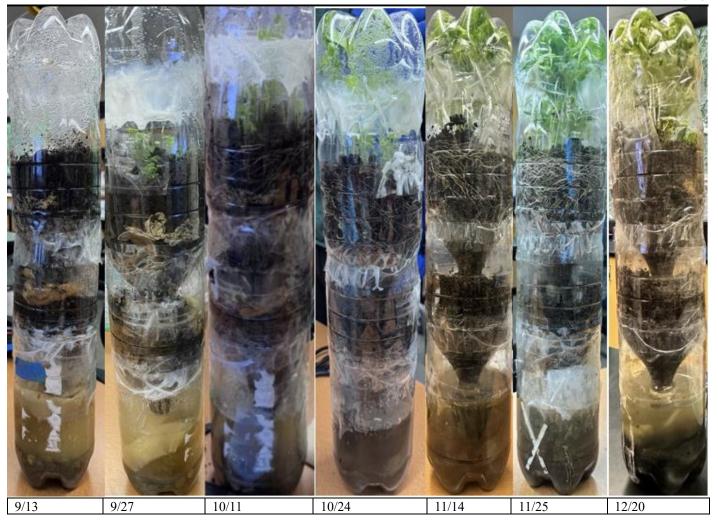
Raw Data: Aquatic Layer Nitrite, Nitrate, DO, pH, GH, and KH

Date	Day	Nitrite (ppm)	Nitrate (ppm)	DO (mg/L)	pН	GH (mg/L)	KH (mg/L)
9/13	2	0	10	1.27 mg/L	6.4	500 mg/L	40 mg/L
9/27	16	0	10	10.08 mg/L	6.4	300 mg/L	0 mg/L
10/11	30	25	25	4.03 mg/L	6.8	150 mg/L	80 mg/L
10/24	44	10	50	2.38 mg/L	8	150 mg/L	120 mg/L
11/14	64	1	0	3.71 mg/L	8	150 mg/L	180 mg/L
11/25	75	0	0	3.92 mg/L	8.4	300 mg/L	180 mg/L
12/20	100	0	0	3.69 mg/L	8	300 mg/L	180 mg/L
1/08**	119	0	0	3.51-3.89mg/L	7.6-8.4	171-189mg/L	171-189mg/L



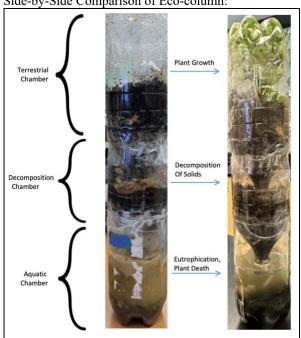
### Eco-column Timeline

- 9/13 No plant growth, lots of water droplets, decomposition material still solid, water quite clear.
- 9/27 Plant has germinated, grown so photosynthesis has started. Decomposition solids have started to break down.
- 10/11- Continued plant growth, decomposition layer decomposition
- 10/24 Roots are exposed, plant is very tall. Water has turned a darker shade.
- 11/14 Basil growth has capped due to space limitations.
- 11/25 Basil has started to wilt; roots have become very congested. Water is a strong black, nothing visible.
- 12/20 Basil has started to die, decomposition layer has finished decomposing, water is very murky and black.



# Reasoning:

Side-by-Side Comparison of Eco-column:



Survived/Died in the Eco-column:

Buivivee	Dica in the Leo-con	4111111.	
Alive		Dead	
-	Basil Plants in Terrestrial Chamber have grown nicely and are behaving regularly.	-	Lemon Bacopa Plant – died due to out of range of tolerance nutrient levels.  Worms in Decomposition Chamber – died due to inconsistent nutrients and food.

On the Left: Side to Side comparison of the eco-column, chambers labeled, significant changes between time periods also noted.

See above in Evidence tab for a full time lapse of the Eco-column from day 1 to 100.

#### Terrestrial Layer:

**pH**: I reject the null hypothesis with  $\pm 5\%$ . Based on the updated prediction (1/8) of pH in the terrestrial chamber, the observed value is 6.2-6.6, which falls outside the range of 6.7-7.3.

**Nitrate**: I fail to reject the null hypothesis with  $\pm 5\%$ . Based on the updated prediction of nitrate in the terrestrial chamber, the observed value is 0, which falls within the expected range of 0.

**Nitrite**: I reject the null hypothesis with  $\pm 5\%$ . Based on the updated prediction of nitrite in the terrestrial chamber, the observed value is 0, which falls outside the expected range of 0.48-0.53.

Note: between the measurements of 11/14 and 11/25, the power in the building housing the eco-column was disrupted (for about 4 days), causing a difference in temperature and light received by the eco-column.

The power shortage (see note above) turned off the lights over the eco-column (until turned back on ~5 days later), which limited the light dependent reactions of photosynthesis in the basil plants (no photons to be received by the plant's chloroplasts'), eventually limiting the amount of CO<sub>2</sub> that gets converted and released as O<sub>2</sub>. This results in higher concentration of CO<sub>2</sub> in the chamber, which directly increases the acidity by dissolving in moisture (in the soil or on sides of eco-column) and reacting with it to form carbonic acid, a weak acid that releases hydrogen ions (H+) when it dissociates, thus making the solution more acidic and lowering the pH value. The data for 12/20 supports this, indicating an unexpected slightly acidic pH of 6 in the terrestrial chamber. The temperature was also disrupted for a period when the building heating wasn't on-briefly affecting optimal environment for enzymes and bacteria, while additionally hampering the travel of water into (and out of) the terrestrial chamber. The colder temperatures slowed evaporation of the water in the aquatic chamber into the terrestrial layer, limiting the water that would then be used in photosynthesis, further impairing the photosynthetic process. Essentially, the environment is more acidic, colder for a period, and is low oxygen. The unexpected acidity disrupts and inhibits natural microbial activity and nitrogen cycling. The unexpected acidity disrupts and inhibits natural microbial activity and nitrogen cycling. This inhibits (by denaturing the enzymes in bacteria) the function of nitrifying bacteria, which facilitate the conversion of ammonium into nitrites and nitrates. The absence of said nitrifying bacteria would result in imbalance in the nitrogen cycle. These bacteria usually prefer more normal to basic conditions (range of tolerance of pH of 7-8), so acidic conditions could stop or slow their activity. The acidic environment also inhibits the productivity of bacteria performing nitrogen fixation, meaning atmospheric nitrogen can't be converted into usable nitrogen (ammonia and nitrate). Furthermore, nitrification significantly slows down in low oxygen environments because the bacteria responsible for nitrification are obligate aerobes [an organism that needs oxygen to cause reactions and grow], meaning they require oxygen to carry out the oxidation reactions necessary for the process; therefore, when oxygen levels are low, the rate of nitrification drastically decreases and can even be completely inhibited. These low oxygen conditions also trigger denitrification, which converts nitrate and nitrite back to natural gas. The disruptions in nitrogen cycling and microbial activity could also trigger feedback loops that increase the issue. For example, reduced nitrogen availability may lead to weaker plant growth, which in turn limits the organic matter returned to the system, reducing substrate availability for decomposers and nitrogen fixers.

Additionally, basil plants are in an active uptake of the nitrogen, especially in nitrate. This is normal activity by the plant, but because other sources of nitrogen may not be available in this soil, it is consuming the small levels of nitrogen (ammonium) available before they can be converted to other forms such as nitrite and nitrate. They are absorbing nutrients at a rate that exceeds the production of nitrates or nitrites by soil microbes. This would thus mean a lesser amount of nitrate in the soil, explaining why the nitrate value was observed to be 0, though the expected range was otherwise. The soil also contains organic material (ungerminated seeds, parts of dead plant) that is undergoing decomposition (in the terrestrial level itself). However, if the decomposition isn't efficient enough, the release of the nutrients (from the dead organism) will slow down. The decrease in decomposition happens based on many different abiotic factors, such as inconsistent temperature, differences in soil compaction, etc. Additionally, the acidic environment denatures bacteria that do decomposition, leading to the buildup of excess waste and limitations on release of nutrients back into the environment. This was a key contributor in slowing down decomposition, decreasing the nutrients in the soil at one time. The combination of all these factors along with the unexpected power outage skewed the pH values and Nitrite values, while keeping the Nitrate values firmly at 0 (but for a different predicted reason). The initial prediction for the Nitrate was 0 due to speculation of the plant absorbing all the nutrients, but the real problem as shown in the later data is a lack of nutrient cycling.

### <u>Decomposition Layer:</u>

**pH**: I reject the null hypothesis with  $\pm 5\%$ . Based on the updated prediction (1/8) of pH in the decomposition chamber, the observed value is 4.99-5.51, which falls outside the expected range of 6.6–7.2.

**Nitrate**: I reject the null hypothesis with  $\pm 5\%$ . Based on the updated prediction of nitrate in the decomposition chamber, the observed value is 499-551, which falls outside the expected range of 380–400.

**Nitrite**: I reject the null hypothesis with  $\pm 5\%$ . Based on the updated prediction of nitrite in the decomposition chamber, the observed value is 0, which falls outside the expected range of 19–21.

The decomposition layer is significantly impacted by the movement of water and dissolved gases within the ecosystem. A large amount of water enters the decomposition layer through evaporation from the aquatic chamber and osmosis from the terrestrial layer above (where water travels from a layer with more water concentration to less concentration). This constant stream of water can carry dissolved carbon dioxide (CO<sub>2</sub>) from other layers into the decomposition chamber. Usually, there is minimal carbon dissolved in the water, but in cold temperatures (power outage), the amount of carbon dissolved can increase. Decomposition (in the eco-column it is

house compost) also releases carbon dioxide (CO<sub>2</sub>) as microorganisms, such as bacteria and fungi, break down organic matter like dead plants, animals, and waste. These microbes respire aerobically, meaning they use oxygen and release carbon dioxide as a byproduct. The carbon in the decomposing material itself is also being released into the atmosphere – a vital part of the carbon cycle (returning of carbon from the dead organisms). This large amount of carbon has the same effect on the decomposition layer as the terrestrial layer: it directly increases the acidity by dissolving in moisture (in the soil or on sides of eco-column) and reacting with it to form carbonic acid, a weak acid that releases hydrogen ions (H+) when it dissociates, thus lowering the pH value, verified by the data collected on 12/20 (a very acidic pH of 5.25).

The unexpected acidity disrupts and inhibits natural microbial activity and nitrogen cycling. This inhibits (by denaturing the enzymes in bacteria) the function of nitrifying bacteria, which facilitate the conversion of ammonium into nitrites and nitrates. The absence of said nitrifying bacteria would result in imbalance in the nitrogen cycle. These bacteria usually prefer more normal to basic conditions (range of tolerance of pH of 7-8), so acidic conditions could stop or slow their activity. The acidic environment also inhibits the productivity of bacteria performing nitrogen fixation, meaning atmospheric nitrogen cannot be converted into usable nitrogen (ammonia and nitrate). This is reflected in the data for 12/20, as there is ample nitrate (499-551ppm) and 0ppm nitrite – there is no way to convert between them, suggesting a bottleneck. Furthermore, another step in the nitrogen cycle ammonification is when ammonia from decomposing organisms, which combines with water and accepts free hydrogen ions in the soil. Without the nitrogen cycle continuing as normal, ammonification wouldn't be able to happen, which would stop one of the biggest balancing factors of the acidity created by carbon dioxide. (the ammonification would reduce the concentration of H+ ions in the soil, increasing pH). The combination of the unexpected levels of CO<sub>2</sub> and acidity led to skewed results for all 3 measurements (pH, Nitrate, Nitrite), leading to the rejection of all 3 predictions.

### Aquatic Layer:

**pH**: I fail to reject the null hypothesis with  $\pm 5\%$ . Based on the updated prediction (1/8) of pH in the aquatic chamber, the observed value is 7.6-8.4, which falls within the expected range of 7.5–8.3.

**Nitrate**: I reject the null hypothesis with  $\pm 5\%$ . Based on the updated prediction of nitrate in the aquatic chamber, the observed value is 0, which falls outside the expected range of 107-119.

**Nitrite**: I fail to reject the null hypothesis with  $\pm 5\%$ . Based on the updated prediction of nitrite in the aquatic chamber, the observed value is 0, which falls within the expected range of 0.

**DO**: I reject the null hypothesis with  $\pm 5\%$ . Based on the updated prediction of dissolved oxygen in the aquatic chamber, the observed value is 3.51-3.89, which falls outside the expected range of 4.37–4.83.

**GH**: I reject the null hypothesis with  $\pm 5\%$ . Based on the updated prediction of GH in the aquatic chamber, the observed value is 171-189, which falls outside the expected range of 38–42.

**KH**: I reject the null hypothesis with  $\pm 5\%$ . Based on the updated prediction of KH in the aquatic chamber, the observed value is 171-189, which falls outside the expected range of 95–105.

Eutrophication occurred in the aquatic environment due to high nitrate levels at the beginning of the eco-column lab (attributed to leaching from above layers), which initiated the growth of algae and started a cascade of ecological changes in the aquatic chamber. Initially, the nitrate concentration exceeded the ecosystem's capacity to process it, promoting rapid algal blooms. These blooms overshadowed the Lemon Bacopa plant, which eventually died due to reduced light availability (water turned very dark due to algae blockage and sediment) and competition for nutrients. The decomposition of the dead plant and algae further disrupted the nitrogen cycle, rapidly depleting both nitrates and nitrites as decomposers consumed these compounds to break down organic matter. The eutrophication process significantly altered water measurements, leading to an increase in pH, with a new predicted value of about 7.6-8.4 that is consistent with the expected outcomes of algal blooms. Algae causes pH to rise as they consume carbon dioxide during photosynthesis, reducing its acidifying effect in the water. However, when they die, they release carbon dioxide into the water, decreasing pH. This volatility is shown in the past few measurements, where we can track algae growth and death. General Hardness (GH), which measures the concentration of divalent cations like calcium (Ca<sup>2+</sup>) and magnesium (Mg<sup>2+</sup>) in water, can indirectly contribute to eutrophication- the high initial levels (GH of 500mg/L) contributed to maintaining conditions ideal for algal life. Elevated calcium and magnesium concentrations provided structural support for algae, enabling them to thrive in the environment. They can also interact with the phosphates in the water, with Calcium ions binding with phosphate (PO<sub>4</sub><sup>3-</sup>) to form calcium phosphate compounds. This increases the phosphate's solubility, increasing the nutrients the algae growing can consume, thereby fueling algal blooms. Furthermore, an increase in carbonate hardness (KH - the amount of carbonate and bicarbonate minerals in water) stabilized the pH and supported continued algal proliferation by providing carbon necessary for photosynthesis. Over time, the ecosystem entered a hypoxic state as dissolved oxygen (DO) levels plummeted (to about 3.51-3.89mg/L in the updated prediction), which is not in the range of tolerance for the Lemon Bacopa plant or most aquatic life. The death and shortly after decomposition of the algae and lemon bacopa created a significant demand for oxygen, which the system could not keep up with. This led to the formation of a dead zone where oxygen levels were too low to support most aquatic life. The combined effects of nitrate depletion, pH elevation, and oxygen reduction are all part of Eutrophication, which made the aquatic chamber severely imbalanced and inhabitable. Eutrophication was factored into the prediction for this aquatic layer, but not to the extent that the data displays. This led to the rejection of multiple measurements, DO, GH, KH, and Nitrite. However, pH and Nitrite were accurately predicted – partially because the pH is balanced (with balancing factors previously explained), and Nitrite always being 0.

The death of organisms is a natural part of an ecosystem's life cycle and plays a massive role in nutrient cycling, flow of energy, and the balance of gases in the atmosphere. While a loss of organism may seem harmful, it decomposes to recycle of essential elements back into the environment and sustain the ecosystem. However, large-scale death of organisms such as eutrophication or habitat disturbance potentially causes ecosystems to collapse and increase greenhouse gas emissions.

Inorganic nutrients are returned to their respective nutrient cycle through decomposers: bacteria, fungi, and detritivores. With the use of enzymes, they degrade a complex organic matter into a simple one such as carbon dioxide CO<sub>2</sub>, methane CH<sub>4</sub>, ammonia, nitrates. These nutrients become available to the producers- plants and algae-through the soil and water, thus serving as the food web's foundation. For example, nitrogen (in any form) from decaying organisms is important for the growth of plants, whereas phosphorus facilitates cellular energy transport (ATP) in producers.

Decomposition releases the essential elements of carbon, nitrogen, and phosphorus to the ecosystem. Carbon is released either as carbon dioxide or methane depending on the oxygen levels. In aerobic decomposition,  $CO_2$  is the major byproduct while anaerobic conditions generate methane-a greenhouse gas. The mass death of organisms can result in higher amounts of these greenhouse gases, thus contributing to global warming and climate change. These factors alter environmental conditions like pH, oxygen levels, and nutrient availability, which effect the nitrogen cycle as well. For example, ammonia released during decomposition can tend to raise the pH of the environment more towards basic. This will influence processes such as nitrification, in which ammonia is oxidized into nitrites and nitrates due to nitrifying bacteria having an optimum pH range. Furthermore, this decomposition in an aquatic environment creates oxygen deficiencies and thus generates anoxic conditions leading to denitrification: the reduction of nitrates into gaseous nitrogen ( $N_2$ ) or even nitrous oxide ( $N_2$ O). All these processes further disturb the natural balance of the nitrogen cycle. They cause disruption in nutrient availability to plants, while building up the proportions of gaseous nitrogenous emissions which are dangerous to environmental health.

For example, in terrestrial ecosystems, large tree or plant death adds organic matter to the soil and releases stored carbon back into the atmosphere. Aquatic organisms can create dead zones upon dying and sinking to the bottom where decomposition depletes dissolved oxygen, which again triggers anaerobic processes leading to methane release and thus worsening greenhouse gas concentrations. This is especially apparent in Eutrophication, where the death of algae blooms cause the oxygen level in the water to become inhabitable for other aquatic life.

When a plant dies, it cannot carry out photosynthesis anymore, thus cannot take up CO<sub>2</sub> and produce oxygen. The local and global carbon balance is disrupted due to the fewer plants removing CO<sub>2</sub> from the atmosphere. This can cause an increase in atmospheric CO<sub>2</sub> over time, reducing photosynthetic organisms and adding to the greenhouse effect. Similarly, in aquatic ecosystems, photosynthetic organisms such as algae have similar impacts when they die on oxygen production and carbon cycling. While it is a natural process, death of organisms and their recycling into production is necessary within ecosystems. Large-scale die-offs through eutrophication, deforestation, and other anthropogenic factors can have destabilizing effects on ecosystems, which, in turn, result in excessive emission of greenhouse gases, nutrient imbalance, and reduced biodiversity. Understanding decomposers and nutrient cycling helps in designing strategies to mitigate these effects, such as reducing nutrient pollution or managing global warming.

In summary, the death of organisms affects ecosystems through nutrient recycling, altered gas dynamics, and energy flow. While it is a critical ecological process, the scale and context of organism death determine whether its effects are beneficial or detrimental.

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