International Baccalaureate

ENVIRONMENTAL SYSTEMS
AND
SOCIETIES

Extended Essay

Effect Of Number of Cigarette Butts In Water Medium On Photosynthesis Rate Of *Elodea canadensis*

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1. Introduction

Last year when I visited a seaside in Izmir, I have noticed that the beaches were filled with cigarette butts. The locals didn't seem to get affected by much however, from the way the sea was foamy and blurry, it was clear that the sea quality was getting affected. Later on, I have learned from the municipality informing poster that cigarette butt pollution was a big problem for that coastal region. This made me think that although the pollution was not at a level to visibly harm human health, it was influencing the sea which can be associated with marine life problems that can surface in the near future. So, I felt the need to investigate the effect of cigarette butts with respect to its amount on the aquatic life.

Among all types of litter, the most common waste is cigarette butts. Cigarette butts are carried as runoff from streets to drains, to rivers, and ultimately to the ocean and beaches, cigarette filters are the single most collected item in international beach cleanups each year. (Novotny et al., 2009) This most seen litter is pervasive, long-lasting, and a toxic form of marine debris. (*What Is the Most Commonly Found Ocean Litter?*, n.d.) Which brings the question of what might be the most littered item's effect on the ecosystem it is dumped to? A 2006 laboratory study found that cigarette butts were found to be acutely toxic to a freshwater organism and a marine bacterium (microtox) and that the main cause of toxicity was attributed to nicotine and ethylphenol in the leachates from cigarette butts. (Novotny et al., 2009) This shows that the contents of cigarettes are capable of disturbing local organisms and bacteria, having the potential of becoming a great stress on the local marine ecosystem. Moreover, according to a study on burned and unburned cigarettes' effect on water chemistry and microbes living in beach sediments, the beach sediment showed change by means of trans metals, pH and native microbes. Results of the study illustrated that cigarettes not only litter a clean beach, but they also introduce metals into an environment and change the local population of microbes, which are key to a healthy ecosystem. Both studies indicate that cigarette butts have a significant effect

on the balance of aquatic systems and opens new doors for researches on how cigarette butts might be affecting ecosystems and how we can fix these situations.

An interesting idea was that, if the cigarette filter had an impact on water chemistry and photosynthetic native microbes, would it have an effect on photosynthetic plants as well? In terms of biological processes, photosynthetic plants are significant for mediums where they are present because most importantly, they supply oxygen which in its absence, the collapse of the ecosystem can be observed due to the uptake of the oxygen needed for the breakdown of organic material, and maintenance of aquatic lives without stresses. Moreover, in terms of energy flow, photosynthesis has great importance in an ecosystem as the product of photosynthesis form Gross Primary Productivity (GPP) which is the total gain in energy or biomass per unit area per unit time, being a determining factor on the rate of growth of individual organisms, thus how fast an ecosystem can recover. When respiration is taken out from GPP, Net Primary Productivity (NPP) is found which is the actual gain of energy or biomass of photosynthetic plants. The more plants photosynthesize the faster they will be able to grow and have the ecosystem mature, increasing resistance to disturbances. Resilience of an ecosystem which is affected greatly by photosynthesis rate is significant especially in 21th century as humans pollute environment more than ever. As cigarette butts are the leading polluters, aquatic biomes and ecosystems that cigarette butts are mostly thrown to have no choice but go through some changes over time.

Elodea canadensis which is a photosynthetic underwater plant is important in lake ecosystems because apart from photosynthesizing, it provides habitat for aquatic invertebrates, fish, and amphibians, and it is a major food source for ducks, geese, beaver and muskrat (Sonneborn, 2013). So, the photosynthesis of this species which provides itself oxygen necessarily for growth and also for the ecosystem, is significant by means of both resonance of the ecosystem and present food chain. If Elodea doesn't photosynthesize enough, the growth of the plant is

going to slow down and will not be as big and filling as it used to be which has the chance of becoming a stress on the food chain by causing elodea eaters to consume more Elodea in numbers or consume more of other available primary producers. On another point, the balance of the ecosystem that highly depends on photosynthetic organisms can be disrupted due to depletion of oxygen caused by decrease in photosynthesis of Elodea and same amount of uptake of oxygen by organisms which can put local aquatic organisms like fish or other plants in stress lowering the overall productivity of the lake. Considering these possible effects, the aim of this investigation is to see the effect of cigarettes on the photosynthesis rate of Elodea which can give an idea about the future of a lake ecosystem after being targeted as cigarette butt dump. So, I came up with the research question: "What is the effect of the number of cigarette butts in water medium, (0, 2, 4, 6, 8) on photosynthesis rate of *Elodea canadensis*, measured with volume of oxygen produced in cm^3 over 1 hour at 25ºC at 100kpa?"

In terrestrial plants photosynthesis happens via stomata (tiny openings on epidermis on leaves which play role in gaseous exchange and transpiration) into the air. (Admin, 2022) In contrast, submerged water plants like *Elodea canadensis* lack stomata and release oxygen from the stem or more specifically through the aerenchyma. (Submerged Plants, n.d.) In pondweeds there is a specialized tissue in the stem called aerenchyma which is composed of large spaces that are filled with gas as the plant photosynthesizes. Gases can diffuse through the aerenchyma so that the plant can respire and photosynthesize. So, in order to investigate the factors that affect photosynthesis, such as cigarette butts, pondweeds are a good option due to the presence of aerenchyma which can allow the observer to visually see the produced gas in the form of bubbles.

If the aquatic plant is taken from an outdoor pond in winter, as it is darker and colder, the plant won't be photosynthesizing at a particularly fast rate which can make the result of this investigation harder to observe. Therefore, prior to the experiment, plant can be allowed to sit

in an aerated water tank with a source of very bright light (e.g. 30watt LED work lamp) for two weeks so that photosynthetic system of the plant can be active again.

2. Hypothesis

As the number of cigarette butts increase in water medium, the photosynthesis rate of *Elodea canadensis* will decrease.

Cigarette butts can become a great stress on living organisms affecting the quality of water. Water indirectly influences the rate of photosynthesis resulting in leaves wilting which lowers the metabolic activity that leaves perform. It is suggested that the more cigarette butts will be present, the more photosynthesis rate is going to drop due to an increase in trans-metals and toxic materials released from cigarette butts which in return is going to deteriorate the functions of the photosynthetic water plant, Elodea.

3. Methodology

3.1 Identification Of Variables

Variable Type	Explanation	Method Of Management	
Independent Variable	Number of Cigarette Butts Put In Medium of Elodea	Varying amounts of cigarette butts have been put in same amount of water in sealed jars and have been kept for 9 days in room temperature as other researches suggested that it took 9 days for cigarettes to release their contents fully into water.	
Dependent Variable	Rate Of Photosynthesis of Elodea Calculated from Volume of Oxygen Produced (cm^3/min)	In order to calculate the rate of photosynthesis of Elodea pieces that are put in solutions with different concentrations of cigarette butts, a closed system that shows the volume of produced oxygen has been set up.	
Variable Type	Explanation	Reason To Control	Method Of Control

Controlled	Length of Elodea Pieces	Length of plant pieces are proportional to the amount of oxygen produced as the more leaves there are the more photosynthesis which takes place on leaves take place, affecting the rate of photosynthesis, dependent variable.	The lengths of pieces have been kept the same as 6 cm.
	Length of leaves of Elodea	The length of leaves affect the photosynthesis taking place on leaves so they have to be approximately the same length to not influence the rate of photosynthesis, a dependent variable.	Outstandingly long leaves have been shortened and outstandingly short leaves have been taken out.
	Temperature of solutions	Temperature of solutions can change the dissolved oxygen and thus the volume of produced oxygen that is going to be measured.	Temperatures have been checked with thermometers and solutions have been kept in the same medium.
	Sodium bicarbonate concentrations in solutions	Sodium bicarbonate concentrations can affect the visible oxygen bubble produced which can temper with the results	3 grams of bicarbonate has been added to all solutions
	Exposure time of cigarette butts	Cigarette butts have to stay in water medium for at least 9 days for trans metals to infuse into water, and exposure time of cigarette butts can affect the amount of trans metals, which can temper with the reuslt	For all solutions, water has been exposed to cigarette butts for 9 days
	Brand of cigarette butt	Brand of cigarette can change the type and ratio of chemicals in the cigarette and this difference can have an effect on the dependent variable.	All cigarette butts have been chosen from the same brand.
	Length of cigarette	the length of a cigarette can indicate how much that cigarette has been smoked and this factor can influence	Cigarette butts that are approximately the same length have been chosen.

		the amount and type of chemicals that are going to be released when they will be put in water for the experiment. This can temper with results as the categorization by number of cigarettes would be wrong.	No calculation has been carried out for this process, outstandingly longer or shorter cigarette butts haven't been used for the experiment.
	Ruler	Rulers may have different scale divisions which would make the overall measurements inconsistent and inaccurate	Same ruler has been used for all produced oxygen measurements from test tubes.

Table 1: Table indicating variables and their method of management

3.2 Materials

Materials	**Quantity (For 1 Trial Of 5 Experimental Groups)**
Sodium bicarbonate	15 grams
200 ml beaker (∓0.05)	5
Funnel	5
Test tube	5
Elodea	1
Jars	5
Ruler (∓0.05)	1

Marker	1
Thermometer (∓0.05)	1
Scissors	1
Tweezer	1

Table 2: Table indicating needed materials for the experiment

3.3 Procedure

Figure 1: Picture of experimental design of all variable groups

Figure 2: Picture of experimental design of 0 cigarette control group

1- Keeping one jar as a control group, put in order, 2, 4, 6, 8 smoked cigarette butts in separate jars, and fill them up to 300 milliliters, and seal them,
2- Keep them for 9 days at room temperature as it takes 9 days for the metals and other contents in cigarettes to fully infuse into water,
3- After 9 days, take out cigarette butts from the jars with a tweezer and dispose them to a cigarette disposal bin.

4- Followingly, add 3 grams of sodium bicarbonate to each solution and mix them with rod as to supply the solutions with CO_2 which makes it easier for photosynthesized oxygen bubbles to be more visible,
5- Cut 6 cm pieces from elodea with scissors while also making sure leaves are approximately of the same length by cutting particularly long leaves and taking out outstandingly short leaves. Additionally, cut the stem of each Elodea piece to make it easier for bubbles to get out of the body of the plant,
6- Place 2 pieces of 6 cm Elodea at the bottom of the beaker,
7- Trap the elodea pieces with an upside-down funnel, keeping every leaf in the funnel,
8- Fill a test tube with the solution that is going to be used for that experimental group and while holding the top of the tube turn it upside down and put on top of the funnel without letting the solution in the test tube leak,
9- Fill the rest of the beaker with the leftover solution in the jar,
10- Mark **the initial volume** of the solution in the test tube, start the timer and after 1 hour mark **the final volume,** and determine how many cm^3 of oxygen has been produced in 1 hour,
11- Repeat steps 5, 6, 7, 8, 9 for all experimental groups of 2, 4, 6, 8 numbers of cigarette butts,
12- With obtained data, make a table, analyze data, and draw a graph.

3.4 Justification

There are several methods of measuring photosynthesis rate: measuring the uptake of CO_2, production of O_2, production of carbohydrates, and increase in dry mass. However, in order to measure the uptake of CO_2, either algae or equipment such as Infra-Red Gas Analyzer (IRGA) is needed which are hard to find. CO_2 production can also be monitored by putting the plant in

a plastic bag, but in this investigation the plant needs to be photosynthesizing in water so that the effect of cigarette in water bodies can be observed, which doesn't fit this method. As for the measurement of dry mass it is relatively time consuming. On the other hand, the method of measurement of oxygen production is the most practical and least time consuming for measuring the photosynthesis rate of a small water plant of elodea that is being affected by cigarette butts in water.

The production of oxygen can be measured via two ways in which either oxygen bubbles are counted or audus apparatus is used. Cigarette butts are indirectly a limiting factor which prolongs the time needed for them to have an effect on the photosynthesis rate of Elodea. Unlike cigarette butts which can take effect over longer time, counting bubbles are usually used for measuring instantly affecting factors like light. So, in terms of experimental design, funnels and test tubes are utilized in order to observe the effect of cigarette butts over a time duration of 1 hour which wouldn't be as practical if the method of counting the produced oxygen bubbles were chosen. So, this experimental setup that is similar to audus apparatus allowing the accumulation of produced oxygen is the most suitable for the independent variable for this investigation.

The appropriate setup involves placement of an aquatic plant at the bottom of a beaker full of sodium hydrogen carbonate solution with a funnel turned upside down and a test tube on top trapping the plant pieces. As the plant photosynthesizes, the gases produced are released from the end of the stem as bubbles. In the setup, funnel and test tube are used to collect the gas that has been produced. This setup allows the observation of photosynthesis rate over a longer period of time as the bubbles are collected without needing to be counted manually.

In this investigation the effect of independent variable of number of cigarette butts in water medium on the dependent variable of photosynthesis rate of Elodea is researched. Analysis of Variance-Single Factor is useful when comparing multiple independent groups with one categorical

independent variable, thus it will be used to compare and analyze the data set composed of 5 variable groups. In order to represent the statistical significance in between data groups, bar graph will be drawn because it is expected for large differences among groups to be seen which is more suitable for bar graph rather than a line graph that display smaller changes in trend.

3.5 Risk Assessment And Safety

Hazard	Risk	Precaution
Scissors	Cut skin	Don't run with scissors. If skin is cut, stop the bleeding, clean the cut, and put plaster on it.
Glassware	Cut skin	Avoid using glassware irresponsibly. If glassware is broken, section off and clean up the area. If skin is cut, stop the bleeding, clean the cut, and put plaster on it.
Cigarette butts solutions	Irritate skin Irritate eye	Wear protective gloves, coat and eye protection. Avoid spilling solutions. In contact with skin, flush skin with water and soap. In contact with eyes, flush eyes with plenty of water for at least 15 minutes.

Sodium bicarbonate	Irritate skin Irritate eye	Wear protective gloves, coat and eye protection. In contact with skin, flush skin with plenty of water and soap for at least 15 minutes while removing contaminated clothing. In contact with eyes, flush eyes with plenty of water for at least 15 minutes. Get medical aid.

Table 3: Table indicating hazards, risks, and precautions

Environmental Considerations: Dispose of cigarette butts to cigarette disposal bins in order not to pollute the environment. Dispose of cigarette butt solutions to a disposal plant so that they wouldn't mix into water bodies and soil, threatening aquatic life and degrading soil. Use minimum amount of chemicals so that they wouldn't be wasted.

Ethical Considerations: As Elodea is a living organism, use only needed amount of Elodea.

4. Data Analysis

4.1 Raw Data

Number of Cigarette Butts In 300 ml Water Medium	Trials	Reading of The Volume Of Produced Oxygen In The Test Tubes (cm^3)(∓ 0.05)
0	1	8.70
	2	8.50
	3	8.30
	4	8.80

	5	8.70
2	1	7.40
	2	7.30
	3	7.60
	4	7.40
	5	7.00
4	1	5.50
	2	5.50
	3	5.20
	4	5.30
	5	5.50
6	1	4.30
	2	4.10
	3	4.50
	4	4.20
	5	4.30
8	1	3.70
	2	3.50
	3	3.80
	4	3.70
	5	3.60

Table 4: Table indicating raw data obtained from experiment including trial numbers, number of cigarette butts variable, and reading of test tubes

Outlier Determination:

Outlier is an abnormally distanced value from other values which indicate random value. After determining them, outlier values shouldn't be taken into calculation in order to only work with reliable values. To find outliers; lower quartile and upper quartile should be found followed by the interquartile range. Afterwards, the range between upper fence and lower fence would give the range of non-outliers.

First Quartile and Third Quartile: boundary for the first 25% of the data points which is calculated by finding the median of the data below median, and boundary for the last 25% of data points which is calculated by finding the median of the data above median.

Interquartile Range: range of data between first and third quartile of data set.

Upper boundary and Lower boundary: Top and bottom limit of range of data set for which data to be accurate

For 0 cigarette butts in water medium:

$Q_1 = (n+1) \times \frac{1}{4} = \frac{8.50+8.30}{2} = 8.50$

$Q_3 = (n+1) \times \frac{3}{4} = \frac{8.80+8.70}{2} = 8.75$

IQR: $Q_3 - Q_1 = 8.75 - 8.50 = 0.25$

Upper fence = Q_3 + (1.5 x IQR) = 8.75 + (1.5 x 0.25) = 9.13

Lower fence = Q_1 - (1.5 x IQR) = 8.50 - (1.5 x 0.25) = 8.13

Any value that is not in between 9.13cm³-8.13cm³ for 0 cigarette butt in water medium is an outlier. Same calculations have been carried out for other numbers of cigarette butt variables and no outlier has been found. Therefore, all values presented in raw data is going to be used for processed data.

4.2 Processed Data

Photosynthesis rate was calculated for each trial of number of cigarette butts variable, using the following formula:

$$Photosynthesis\ rate = \frac{volume\ of\ oxygen\ produced\ in\ cm^3}{Total\ time\ in\ hour}$$

Sample calculation for Trial 1 of 0 cigarette butts in water medium:

$$Photosynthesis\ rate = \frac{8.70}{1.00} = 18.70\ cm^3/hour$$

This procedure was repeated for each value. The photosynthesis rate of Elodea per each trial is established in Table 5.

4.3 Sample Mean, Standard Deviation, Standard Error, 95% Confidence Interval Calculation

In order to assess the spread of data of photosynthesis rate, standard deviation, standard error, and confidence interval were calculated with the following formulas, where; X_i stands for individual value in sample, μ refers to mean of sample, and N stands for count of values in sample.

Sample calculation for 0 cigarette butts in water medium:

Sample mean = $\mu = \sum_{i=1}^{n} x_i = \frac{8.70+8.50+8.30+8.80+8.70}{5} = 8.60$

Sample mean uncertainty = $\frac{max-min}{2\sqrt{n}} = \frac{8.80-8.30}{2\sqrt{5}} = 0.60$

$$\frac{0.60}{8.60} \times 100 = \pm 7\%$$

Standard deviation = $\sigma = \sqrt{\frac{\sum(x_i-\mu)^2}{N}}$

$$\sqrt{\frac{(8.70-8.60)^2+(8.50-8.60)^2+(8.30-8.60)^2+(8.80-8.60)^2+(8.70-8.60)^2}{5}} = 0.179$$

Standard error = $\sigma_{\bar{x}} = \frac{\sigma}{\sqrt{n}} = \frac{0.179}{\sqrt{5}} = 8 \times 10^{-2}$

95% Confidence Interval = $1.96 \times \sigma_{\bar{x}} = 1.96 \times 8 \times 10^{-2} = 0.157$

The calculations were repeated for all number of cigarette butts in water medium variables and presented in Table 5.

Number of Cigarette Butts In 300 ml Water Medium	Trials	Photosynthesis Rate of Elodea (cm^3/hour)	Mean of photosynthesis rate of 5 Trials	Percentage Sample Mean Uncertainty (%)	Standard Deviation	Standard Error of The Mean (SEM)	95% Confidence Interval (Margin of Error)
0	1	8.70	8.60	6.98	0.179	8×10^{-2}	0.157
	2	8.50					
	3	8.30					
	4	8.80					
	5	8.70					
2	1	7.40	7.34	1.83	0.196	9×10^{-2}	0.172
	2	7.30					
	3	7.60					
	4	7.40					
	5	7.00					
4	1	5.50	5.40	1.24	0.126	6×10^{-2}	0.111
	2	5.50					
	3	5.20					
	4	5.30					
	5	5.50					
6	1	4.30	4.28	2.09	0.133	6×10^{-2}	0.116
	2	4.10					
	3	4.50					
	4	4.20					
	5	4.30					
8	1	3.70	3.66	1.83	0.102	5×10^{-2}	0.089
	2	3.50					
	3	3.80					
	4	3.70					
	5	3.60					

Table 5: Table indicating processed data including photosynthesis rate per each trial of numbers of cigarette butt variable, mean of photosynthesis rates, percentage mean uncertainty, standard deviations

Standard error of the mean which indicates how much the sample would vary if the experiment were to be repeated with new samples is important to estimate the accuracy and consistency of the sample showing how well the sample represents a population. In Table 5, standard errors of means of photosynthesis rates are close to zero with values ranging from 0.05 to 0.09 indicating that the estimations of mean photosynthesis rates are close to their true values' means. Another statistical fact that shows calculations are accurate is sample mean uncertainties in between 1.24% – 6.98% range that tells the degree of error associated with measurement is low. The standard deviations of 0.179, 0.196, 0.126, 0.133, 0.102, which are all smaller than 1 and are relatively close to zero tells that the data set is not spread but the data are precise by being gathered.

The 95% confidence intervals are in the range band of 0.089 and 0.172 which is slightly narrow showing that the uncertainty is low enough for the individual values of photosynthesis rates to be precise. Therefore, the calculations of 95% Confidence Interval and Standard Deviation show that both the sample data and individual data in the sample are precise, while percentage sample mean uncertainty and standard error of the mean illustrate that data are accurate.

4.4 Further Analysis

There are 5 variable groups of different varying cigarette butt number that water medium is exposed to as follows: 0, 2, 4, 6, 8 cigarette butts. In order to compare 5 variable groups in terms effect of different number of cigarette butts on photosynthesis rate, the relationship between independent and dependent variable can be figured via Analysis of Variance (Anova).

Null Hypothesis (H_0): There is not a significant mean difference between rate of photosynthesis of elodea kept in 300 ml water medium that are exposed to different number of cigarette butts for 9 days.

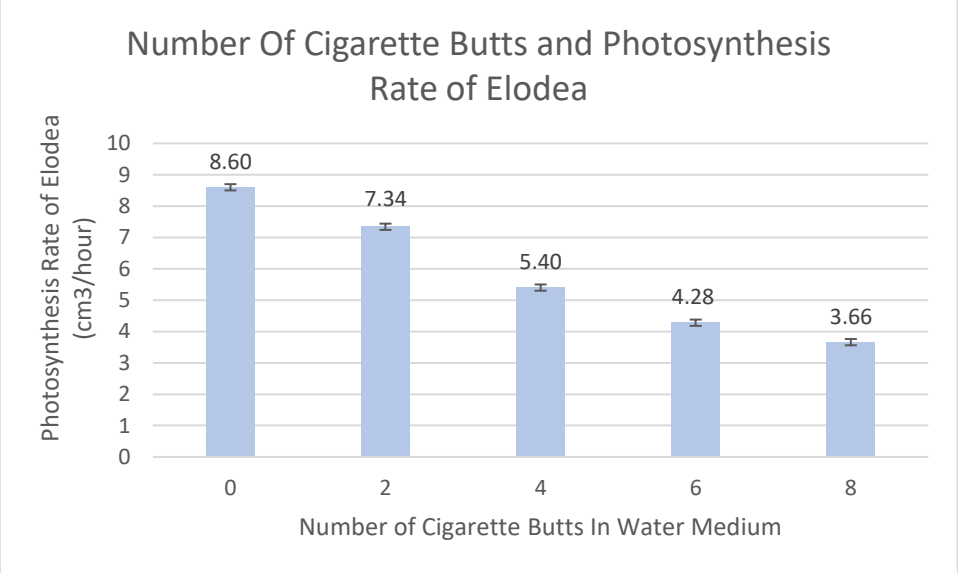
Alternative Hypothesis (HA): There is a significant mean difference between rate of photosynthesis of elodea kept in 300 ml water medium that are exposed to different number of cigarette butts for 9 days.

ANOVA

Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	86,2296	4	21,5574	753,7552	0	2,866081
Within Groups	0,572	20	0,0286			
Total	86,8016	24				

Table 6: Anova Analysis Evaluation

The Anova Analysis which helps compare and analyze means of two or more groups showed results of F-value 753.7552 and P-value 0. Large F-value indicates that a great difference among the group means is present in other words; variations between 0, 2, 4, 6, 8 numbers of cigarette butt independent variable groups are significant. So, with each common difference of 2 cigarette butts, the rate of photosynthesis changes. The P-value being lower than 0.05 supports that there is a statistically significant difference while providing evidence to reject the null hypothesis. Therefore, H_0 is rejected and H_1 is accepted.



Graph 1: Graph showing photosynthesis rate per number of cigarette butts variable with error bars denoting standard deviation

The graph can help visualize the data and relation between dependent and independent variable better while also including the coverage of standard deviation which can tell how reliable data might be. The bar graph, demonstrates dependent variable of photosynthesis rate group means from

most to least as: 8.60, 7.34, 5.40, 4.28, 3.66 in each bar which the values are worked out from y-axes while on x-axes number of cigarette butts independent variable is displayed. The overall trend shows a gradual decline with a decreasing gradient. The peak photosynthesis rate value belongs to control group of 0 cigarette butt number while the lowest point is 8 cigarette butt number group's in which a constant decline with a slight downward curve could be figured which describes an inversely proportional relationship between dependent and independent variable as cigarette butt number increases. From this graph the photosynthesis rate for more number of cigarette numbers cannot be projected as at some point the cigarette butts will become a limiting factor that is going to stabilize the photosynthesis rate which can not be determined with the current amount of data.

5. Discussion And Evaluation

In this experiment, the causations of random errors can be due to measurement and experimental design. Although random errors were tried to be minimized, it is not 100% possible for random errors to not occur at all. So that there wouldn't be an error with the measurement of volume in the test tube, same ruler has been used for all trials and experimental groups, however there is still a chance of error due to human eye capabilities. In order to control the rate of photosynthesis per piece before they were exposed to cigarette butts, Elodea was cut into pieces of same length and leaves were cut when necessary to ensure all pieces had same length of leaves. However, ruler was not used to measure the size of each individual leaf as they were too small. This, could have caused small variations between each Elodea piece's photosynthesis rate which would be random error.

Limitations	Effect On Result And Why	Suggested Improvements
Light Intensity	The experiment took place in a place where each Elodea could see sunlight however, individual light sources weren't placed in front of each experimental design. Therefore, there is a high chance that sunlight reaching each Elodea varied slightly which might have affected the rate of photosynthesis by little.	30 watt LED work lamp can be arranged in front of each experimental group per trial at same distance, allowing all Elodeas to receive light of same intensity from same distance and angle.
The investigation focused on effect on Elodea	The results of the experiment is only applicable to Elodea or similar aquatic plants	The experiment can be conducted with different aquatic plants to see whether the change in rate of photosynthesis would follow approximately the same trend.
There were limited number of experimental groups	The effect of 5 varying number of cigarette butts were observed with 5 trials per experimental group. This limits the scale of application of this investigation as a projection can not be made based on the current amount of data.	A more inclusive experiment can be executed with more trials and with a wider range of number of cigarette butt variable.
Certain type of cigarette butts were chosen	As same brand of cigarettes had to be used for all groups, trans metals and toxic materials that were relased were only limited to the variety and concentration in that brand.	A larger scale experimantation including more brands of cigarettes with filtered and unfiltered differentitation can be carried out.

Table 7: Table indicating limitations, effect on the results and reason, and suggested improvements

6. Conclusion

The world is against many ongoings that contribute to environmental problems, and climate change; such as pollution that can be in the form of soil, air, and water pollution. Cigarette butts are leading polluters which have the potential to disrupt especially aquatic ecosystems by tempering with the quality of local niche, health and density of living organisms, thus present food web. Photosynthetic activities of aquatic organisms are a good indirect way of measuring the effect of a pollution. In lead of my interest in the effect of cigarette butts on photosynthetic aquatic plants, the investigation was made to find the change in photosynthesis rate of *Elodea* plant with varying number of cigarette butts. As discussed in the hypothesis, it was expected for the rate of photosynthesis to decrease as the number of cigarette butts were increased based on the assumption that trans-metals and toxic elements in cigarette butts would deteriorate the photosynthetic systems in the leaves of the *Elodea canadensis,* lowering the photosynthesis rate. Derived from this, an experimental design that allowed the observation of rate of photosynthesis via the measurement of volume of accumulated produced oxygen was set as it was most practical for implementing this measurement for a living aquatic plant. The results of the experiment revealed values in line with hypothesis.

In Table 5, mean of 5 trials of photosynthesis rate per each independent variable group was as follows: 8.60, 7.34, 5.40, 4.28, 3.66 which visibly showed that numerically values decreased. The data was accurate and precise because standard errors in range of 0.05-0.09 showed that data were close to their true values and standard deviations in between 0.102-0.196 indicated that sample data were gathered together being precise.

Sample mean uncertainty and 95% confidence interval also supported that individual data accuracy was high and precise. So, it can be said that the calculated values of photosynthesis rate of Elodea were reliable.

However, it had to be statistically supported by evidence that the change in between groups were significant owing to the single factor of number of cigarette butt independent variable. Therefore, Anova Analysis was carried out which exhibited F-value of 753.7552 and P-value of 0 that

supported that there was a significant mean difference. Graph 1 that illustrated the visualization of data set including the general pattern that displayed a decreasing trend. As number of cigarette butts increased, the photosynthesis rate decreased establishing an inverse relationship. In conclusion, from this experiment with research question: "What is the effect of the number of cigarette butts in water medium, (0, 2, 4, 6, 8) on photosynthesis rate of *Elodea canadensis*, measured with volume of oxygen produced in cm^3 over 1 hour at 25ºC at 100kpa?" it can be deduced that increase in number of cigarette butts in water medium decreases the photosynthesis rate of *Elodea canadensis* in cm^3 per hour establishing an inverse relationship. So, it can be deduced that cigarette butts thrown to water can reduce photosynthesis rate of Elodea in an aquatic ecosystem, thus lower the NPP as respiration will take up the products produced by photosynthesis. Which in return they can lower the growth of Elodea, cause O_2 to become a limiting factor, reduce niche quality and lower resilience of the ecosystem that can even lead to its collapse, if no measurement is taken on the long run. Derived from this conclusion, it wouldn't be wrong to suggest the application of minimizing cigarette butts thrown to environment, especially in coastal areas, in order to lessen the exposure time of cigarette butts as well as their amount. To start from, human activity can be altered as it is the most fundamental level of change. Via utilization of media and community groups to raise awareness and spread the act of reducing throwing cigarette butts, masses can be guided to not pollute environment with cigarette butts. Followingly, governments can be persuaded or pressured to make legislations and regulations regarding this problem, as well as cigarette butts can be cleaned from contaminated cites.

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