

# Blocked and Loaded: Does Sunscreen Reduce Phytoplankton Growth in Marine Ecosystems?

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## ABSTRACT

Along British Columbia's approximately 46,500 km of coast, phytoplankton serve as a foundational part of ecosystems. In existing literature, organic UV filters (OUVFs) such as homosalate have been shown to have adverse effects on phytoplankton biomass in tropical climates. However, there is a limited understanding of these impacts in temperate environments. In this study, we used chlorophyll extraction and spectrophotometry to quantify the changes in Chlorophyll *a* (Chla), Chlorophyll *b* (Chlb), and Chlorophyll *c<sub>1</sub>* + *c<sub>2</sub>* (Chlc) in relation to varying quantities of exposure to sunscreen that contained high mass percentages of Homosalate (15%), Octocrylene (10%), Octisalate (5%), and Avobenzone (3%). The objective of this research was to quantitatively determine if these chemicals would have an impact on phytoplankton biomass. The hypothesis was that sunscreen would decrease chlorophyll levels, as phytoplankton responded negatively to OUVFs. We hypothesized that increased exposure to sunscreen would decrease the relative chlorophyll in samples, as phytoplankton levels would stagnate in agreement with existing literature focusing on tropical environments. We determined there was a significant impact on relative Chlc amounts within trials ( $p = 0.015$  in ANOVA test), and that results showed a negative linear correlation across all chlorophylls. This information has the potential to necessitate oceanography, marine biology, and ecological research on British Columbia's (BC) coast, especially as more chemicals make their way into the province's natural waterways which could impact the biodiversity within ecosystems.

**Keywords:** Sunscreen, Phytoplankton, Toxicology, Chlorophyll.

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## INTRODUCTION

British Columbia's South Coastal region covers approximately 46,552 km<sup>2</sup><sup>[1]</sup>, which includes Tower Beach and Wreck Beach, both popular tourist destinations for swimming and enjoying Vancouver's scenic views. Between 2018 and 2023, the number of visitors increased by 20%, with over 870,000 visitors in 2023<sup>2</sup>. As this beach becomes more popular, there will be an increase in the chemicals entering the water via sunscreen and other topical products, including organic UV filters (OUVFs) such as oxybenzone and homosalate. Within Vancouver's waters, there are ecosystems with high levels of variation and large populations of phytoplankton, which play a vital role in the marine food web as primary producers<sup>3</sup> and produce up to 20% of the oxygen present in Earth's biosphere<sup>4</sup>.

There is significant research concluding that OUVFs have negative effects on the health of coral reef sites<sup>5,6</sup> and freshwater populations<sup>7,8</sup>; however, to our knowledge there is a gap in research focusing on these effects in colder marine regions. This uncertainty in the impact of OUVFs in colder climates may lead to environmental harm or damage to the marine ecosystem including the BC coast. Analysis of OUVFs may realize a need to evaluate environmental protections in these regions, and conduct more analyses on these products.

Our study aims to answer the question, "What effect does sunscreen have on the biomass of Pacific Northwestern phytoplankton?", during which we will use a Trichromatic method of spectrophotometry and associated equations to quantify chlorophyll (Chl) in samples as an indicator of biomass and phytoplankton health. We hypothesize that an increase in the presence of sunscreen will result in a decrease in calculated chlorophyll, indicating a reduction in phytoplankton biomass due to the toxicological effects of homosalate, octocrylene, octisalate, and avobenzone as discussed in existing research. We further expect this study to help assess the ecological effects and risks of common OUVFs in commercial sunscreens.

## METHODS

### SAMPLE COLLECTION

This study was conducted using water samples taken from Tower Beach, located on the northern side of the Point Grey Peninsula (Fig. 1). During the period of the study from February 17th to March 8th 2025, the average surface water temperature was  $6.46 \pm 0.94$  degrees Celsius (this was the average recorded at Soames Creek in Gibsons<sup>9</sup>). Seawater was collected from a rocky outcrop using a small container, then poured into a two-litre jug for transport.



**Figure 1.** Map and image of Tower Beach, British Columbia, with sample collection site indicated. This location was selected for its ease of recognition and easy access to deeper water for sample collection.

### SAMPLE CULTIVATION AND EXPOSURE

Following collection, samples were strained through a 200  $\mu\text{m}$  sieve to remove zooplankton and other debris. Samples were stored in a one-litre Erlenmeyer flask and a one-litre glass bottle; this discrepancy was due to the glassware available during the study. Samples were fed once daily with aquarium-grade "Plant Pack Enhancer: NPK" Phosphorus, Potassium, and Nitrogen mixtures from Seachem Laboratories<sup>10</sup> in standardized amounts (Table 1). Between 6 am and 10 pm, samples were exposed to full spectrum LED lights, with 6500 K white light, 450 nm blue light and 660 nm red light.

**Table 1.** Daily nutrient quantities during each stage of Phytoplankton cultivation.

	1 L Erlenmeyer Flask	0.9 L Bottle	200 mL Erlenmeyer Flask
<b>Nitrogen</b>	16.0 $\mu\text{L}/\text{day}$	15.0 $\mu\text{L}/\text{day}$	3.5 $\mu\text{L}/\text{day}$
<b>Phosphorus</b>	32.0 $\mu\text{L}/\text{day}$	29.0 $\mu\text{L}/\text{day}$	6.5 $\mu\text{L}/\text{day}$
<b>Potassium</b>	40.0 $\mu\text{L}/\text{day}$	36.0 $\mu\text{L}/\text{day}$	8.0 $\mu\text{L}/\text{day}$

Calculations were based off of resources from manufacturer, available here: <https://www.seachem.com/downloads/charts/Plant-Dose-Chart.pdf>.

After 7 days of growth, samples were subdivided into five Erlenmeyer flasks with 200 mL of phytoplankton sample each (Fig. 2). Flasks were then exposed to varying quantities of a standard solution containing 10 g/L of "Neutrogena Hydro Boost Water Gel Lotion Sunscreen with SPF 50"<sup>11</sup>, starting at 4 mL and increasing by 2 mL increments up to 10 mL per beaker. This product was selected due to its high mass percentages of Homosalate (15%), Octocrylene (10%), Octisalate (5%), and Avobenzone (3%) which are the highest amounts permitted by Health Canada<sup>12</sup>. Samples continued to populate for an additional 5 days before

extraction.



**Figure 2.** Image of experimental setup used for sample cultivation and exposure. The sample was subdivided into these Erlenmeyer flasks after 7 days of growth, at the same time they were exposed to sunscreen treatments.

#### PIGMENT EXTRACTION

Our extraction procedure was based on the work of Walsham et al.<sup>13</sup>; we adapted their procedure to avoid drying and storing samples. To determine the chlorophyll amount within each sample, beakers were drained through vacuum filtration, before being soaked on filter paper in 99% acetone for 60 seconds and muddled with a mortar and pestle. The solution was then adjusted to a final volume of 10 mL with acetone. The resulting liquid was then analyzed in 3 mL amounts with a spectrophotometer (Tables 4-7), with pre-determined wavelengths (630 nm, 647 nm, 664 nm, and 750 nm) absorption recorded, we then used published equations from Jeffrey and Humphrey<sup>14</sup> to provide the quantity of Chlorophyll *a*, *b*, *c*<sub>1</sub> + *c*<sub>2</sub> within our samples (Eqs. 1, 2, 3). Within these equations, *V*<sub>e</sub> is the extracted volume (0.003 L in our trials), *V*<sub>f</sub> is the total filtered volume (0.2 L in our trials), and *L* is the Cuvette light-path in centimeters (1 cm in our trials).

$$\text{Chlorophyll } a (\mu\text{g ml}^{-1}) = [11.85(E_{664} - E_{750}) - 1.54(E_{647} - E_{750}) - 0.08(E_{630} - E_{750})] \frac{V_e}{LV_f} \quad (1)$$

$$\text{Chlorophyll } b (\mu\text{g ml}^{-1}) = [-5.43(E_{664} - E_{750}) + 21.03(E_{647} - E_{750}) - 2.66(E_{630} - E_{750})] \frac{V_e}{LV_f} \quad (2)$$

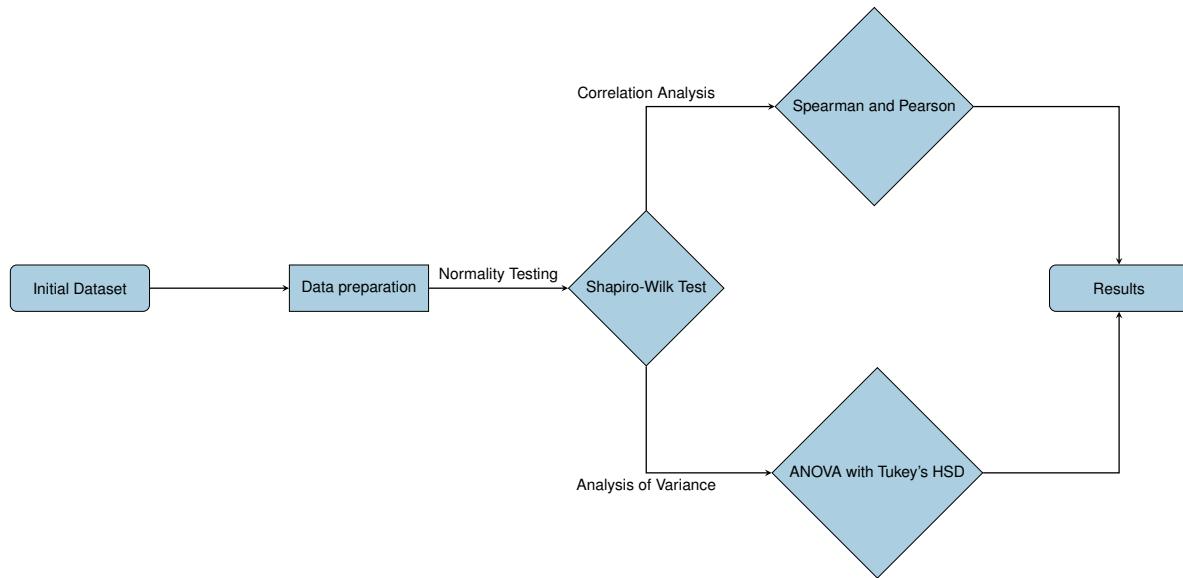
$$\text{Chlorophyll } c_1 + c_2 (\mu\text{g ml}^{-1}) = [-1.67(E_{664} - E_{750}) - 7.60(E_{647} - E_{750}) + 24.52(E_{630} - E_{750})] \frac{V_e}{LV_f} \quad (3)$$

## STATISTICAL ANALYSIS

To analyze the results of our study, we first calculated the relative chlorophyll content (R-Chl) in each treatment (Eq. 4), this value was used for all statistical analysis as it normalized our dataset (Fig. 3).

$$\text{Relative Chlorophyll} = \frac{\text{Chlorophyll in Treatment}}{\text{Chlorophyll in Control}}. \quad (4)$$

Using R-Chl values for each treatment, we established normal data distribution using the Shapiro-Wilk test, with a null hypothesis that our treatments have normal distribution. Both chlorophyll *a* and *b* resulted in  $p > 0.05$ , with Chl*c* being an outlier with a p-score of  $p = 0.028$  (Table 8). These results mean we fail to reject the null hypothesis for Chl*a* and Chl*b*, thus our data for Chl*a* and *b* has a normal distribution. Only Chl*c* does not reject the null hypothesis, and consequently is not normally distributed. After establishing these characteristics of our data, we used the SciPy statistics library to perform ANOVA tests with Tukey's Post-hoc and Spearman and Pearson Correlation tests on our data, all the resulting tables are presented in Appendix B.



**Figure 3.** Schematic representation outlining our methods of statistical analysis on the collected dataset.

## RESULTS

The results of our study show that exposure to sunscreen and OUVFs does have an impact on chlorophyll levels within trials. We used both ANOVA and Tukey's Post-hoc tests to quantify the correlation between chlorophyll levels and exposure to OUVFs (Table 9), only Chl*c* had significant changes, as seen by its scores

of  $F = 5.298$  and  $p = 0.048$  (Table 9), though Chl $a$  and Chl $c$  revealed no significant differences across different sunscreen volumes, which was confirmed by the Tukey post-hoc test with a score of  $p = 0.0109$  (Table 12). The ANOVA results conclude that Chl $a$  and Chl $b$  levels do not vary significantly with changes in sunscreen volume, but Chl $c$  are significantly affected by sunscreen volume, with a notable decrease observed at 10.0 mL of exposure.

In addition to the ANOVAs (Table 9), we ran our data through Pearson and Spearman correlation tests to measure the strength and direction of the relationship between sunscreen exposure and chlorophyll levels. The tests found Chl $c$  has a significant moderate negative correlation (Pearson:  $r = -0.639$ ,  $p = 0.002$ , Spearman:  $r = -0.572$ ,  $p = 0.008$ ), affirming the ANOVA results. Chl $b$  had somewhat vague values, indicating a marginally non-significant negative linear relations ( $r = -0.388$ ,  $p = 0.091$ ) (Table 13). Chl $a$  showed a weak negative correlation, though it was not statistically significant as both tests were well above our alpha value (Pearson:  $p = 0.222$ ; Spearman:  $p = 0.144$ ). These results can be clearly seen when looking at the greatest percent change using either the mean (Table 2) or individual samples (Table 3).

**Table 2.** Greatest percent changes in each trial, using mean of treatment.

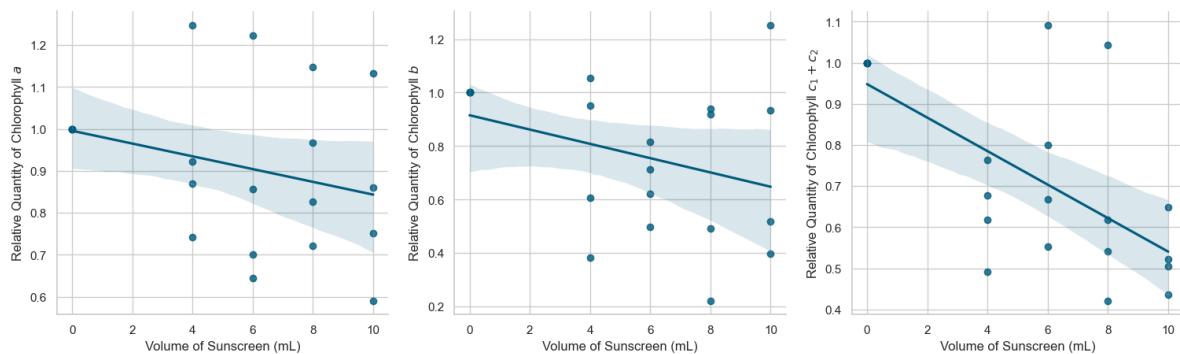
Chlorophyll Type	Greatest Percent Change	Treatment (Sunscreen Volume)
<i>a</i>	-16.59%	10 mL
<i>b</i>	-35.82%	8 mL
<i>c</i>	-47.26%	10 mL

To determine the greatest percentage change in each chlorophyll type (Chl $a$ , Chl $b$ , Chl $c$ ) relative to the control (0 mL of sunscreen), we calculated the mean values for each treatment and compared them to the control mean.

**Table 3.** Greatest percent changes in each trial, using individual treatments.

Chlorophyll Type	Greatest Percent Change	Treatment (Sunscreen Volume)
<i>a</i>	-40.95%	10 mL
<i>b</i>	-77.84%	8 mL
<i>c</i>	-57.90%	8 mL

To determine the greatest percentage change in each chlorophyll type (Chl $a$ , Chl $b$ , Chl $c$ ) relative to the control (0 mL of sunscreen), we used individual data points.



**Figure 4.** Scatter plots of Relative Chlorophyll *a*, *b*, *c*<sub>1</sub> + *c*<sub>2</sub> levels. Graphs have been overlaid with fits using Seaborn<sup>15</sup> regplot functions. These plots visualize the negative correlations found in Chlorophyll *c* and potentially Chlorophyll *b* in relation to sunscreen exposure.

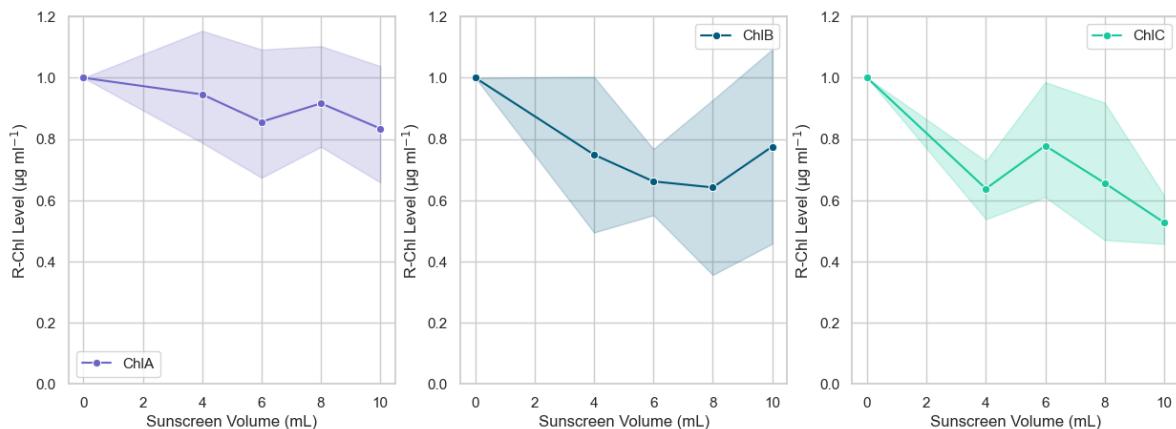
The overview of our results is that: Chl*a* shows no significant correlations with sunscreen exposure, Chl*c* exhibits a significant negative correlation, and Chl*b* shows a marginally non-significant negative linear correlation, but this relationship is not strong enough to be conclusive. The strongest effect of sunscreen was observed in Chlorophyll *c*, with the strongest negative correlation and changes in mean R-Chl amounts.

## DISCUSSION

Across Chl*b* and Chl*c*, we noted a decrease in the R-Chl present in treatment samples (Fig. 5). These trends present in our data agree not only with our expectations but also with the findings of Prakash and Anbumani<sup>5</sup>, as well as Li et al.<sup>8</sup> The behaviour of R-Chl values in our data agrees with our hypothesis, as we did see a negative correlation between the amount of exposure to sunscreen and the resulting chlorophyll levels.

The research done is not without limitation; due to time and resource constraints, the research was performed with a small sample size. Future research testing a greater number of treatments, and limiting the variation between collection times is suggested. Our concentration of 10 g/L was intended to overshoot the realistic presence of sunscreen in marine ecosystems, which leaves room for studies focused on smaller quantities. It is believed that larger data sets could minimize the ambiguity of the statistical analysis, and allow for more nuanced research questions such as "Are these trends reflected in varying ecological locations?" or "Do seasonal variations in climate affect the impacts of these processes?". Further refinement of the cultivation and extraction techniques is highly encouraged, as all of the techniques used are prone to human error, which can be combatted by automated equipment.

Additionally, the cultivation samples can be further regulated to minimize error. Due to the availability of glassware, there was inconsistency in the containers phytoplankton were grown in. While temperatures were



**Figure 5.** Scatter plot of mean Relative Chlorophyll *a*, *b*,  $c_1 + c_2$  levels. These plots visualize the negative trends when generalizing the results of our experiment through averaging individual R-Chl data points.

maintained at room-temperature, there may have still been minor fluctuations not accounted for, as well as higher temperature that may have been higher than typical environment. We also did not study the effects of currents, UV degradation, and chemical breakdown of the sunscreen in our study which may have introduced uncertainty and error. Errors may exist due to accidental contaminants in the water from foot traffic, although this is likely minimal.

Our findings conceivably hold significance across several fields, both in research and industry. This study urges further research into the ecological impacts of OUVFs on biodiversity loss and nutrients production within marine environments, which could have long-reaching impacts on marine and coastal ecosystems and species. On a global scale, the findings are potentially important for fisheries and aquaculture as pillars of economies. There are still unknowns about the impacts these chemicals could have on fisheries and conservation efforts, which are both important areas of research in British Columbia. In summary, this study is significant to the long-term well-being of British Columbia's marine ecosystems, and concludes that organic Ultraviolet filters have negative affects on the phytoplankton exposed when using chlorophyll as a benchmark for phytoplankton biomass.

## CONCLUSIONS

This study was undertaken to investigate the impact of sunscreen on cold-water phytoplankton. Negative correlations were found between the exposure of phytoplankton to sunscreen containing organic UV filters and the relative amounts of Chlorophyll *c*. The research was inconclusive for a correlation with Chlorophyll *a* and *b*, which suggests further research should be undertaken. We are confident that we answered our research question and supported our hypothesis, though the findings should be interpreted with caution

given the small initial dataset and limitations of the study. With mounting uncertainties of the impacts of OUVFs in British Columbia's fragile marine ecosystems, the question still remains how human actions can prevent further damage.

## ACKNOWLEDGMENTS

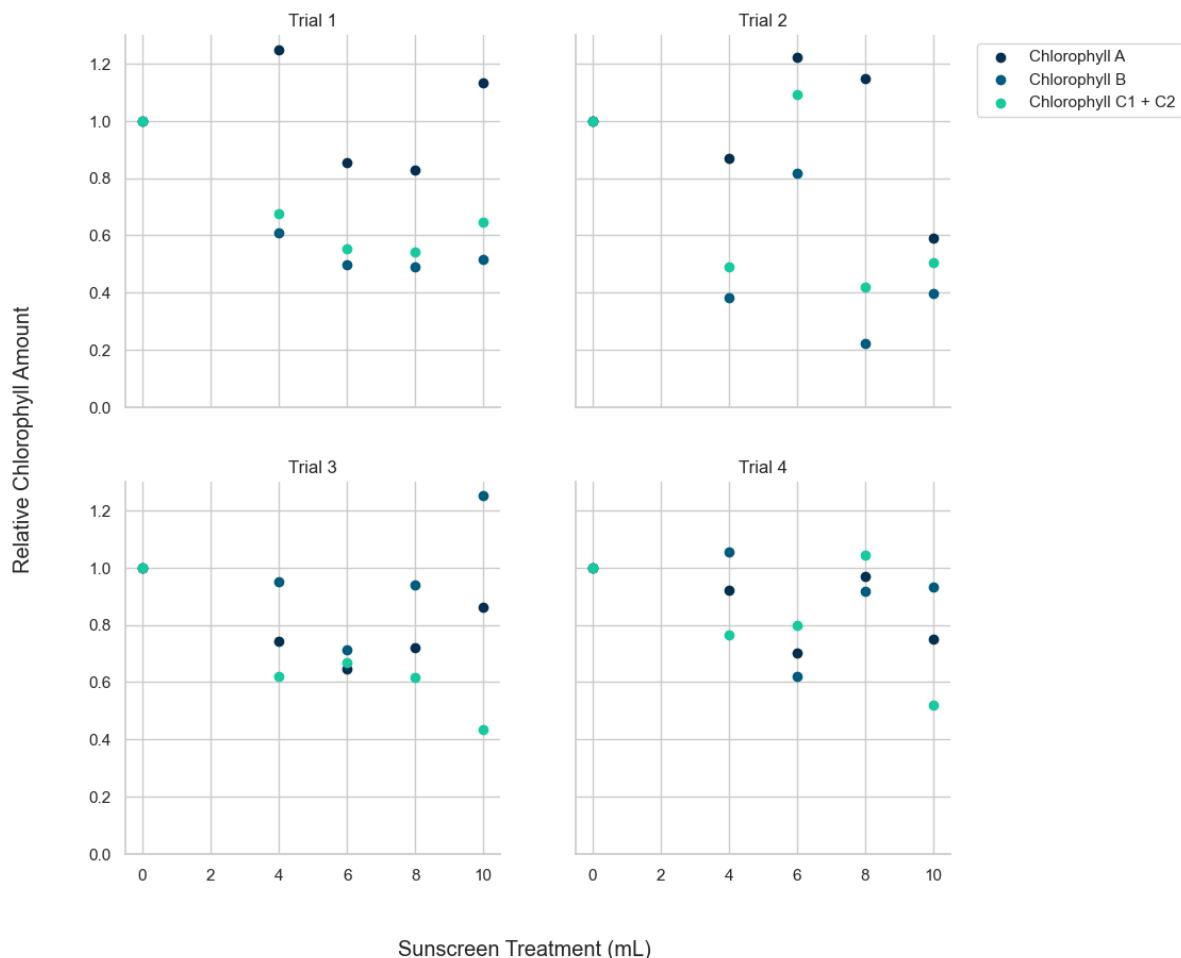
We would like to express gratitude to our mentor Solveig van Wersch for her mentorship and guidance throughout this project. We'd also like to extend our appreciation to Niloo Nasiri Faskhodi for overseeing sample growth when needed. Additionally, this paper would not be possible without the support of Dr. Guillaume Bussiere, and Johanna Marshall, who offered us assistance and expertise over the course of our research.

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## APPENDIX A: VISUAL REPRESENTATIONS



**Figure 6.** Scatter plots of Relative Chlorophyll *a, b, c<sub>1</sub> + c<sub>2</sub>* levels for each trial. These plots visualize the individual behaviours of each chlorophyll within each trial. There are notable negative linear trends within the data points.

## APPENDIX B: STATISTICAL RESULTS

**Table 4.** Absorbance data for Trial 1

Volume of Sunscreen (mL)	630.000nm	664.000nm	647.000nm	750.000nm	635.000nm
0	0.13410	0.14105	0.13358	0.10508	0.13380
4	0.07314	0.09415	0.07435	0.05094	0.07218
6	0.04456	0.05711	0.04488	0.02725	0.04372
8	0.05175	0.06372	0.05205	0.03483	0.05125
10	0.06757	0.08616	0.06774	0.04697	0.06596

These results were collected from the Spectrophotometry software, using the specified wavelengths.

**Table 5.** Absorbance data for Trial 2

Volume of Sunscreen (mL)	630.000nm	664.000nm	647.000nm	750.000nm	635.000nm
0	0.28910	0.38008	0.29881	0.21907	0.28592
4	0.09396	0.18444	0.10315	0.05366	0.09110
6	0.28690	0.39918	0.29372	0.21392	0.28284
8	-0.11471	0.01395	-0.10160	-0.15742	-0.12831
10	0.09796	0.15297	0.10201	0.06348	0.09532

These results were collected from the Spectrophotometry software, using the specified wavelengths.

**Table 6.** Absorbance data for Trial 3

Volume of Sunscreen (mL)	630.000nm	664.000nm	647.000nm	750.000nm	635.000nm
0	1.40387	1.41137	1.54416	1.22706	1.39943
4	0.83629	0.8398	0.9859	0.69574	0.82934
6	0.76775	0.76779	0.86913	0.64648	0.76254
8	0.82774	0.82933	0.97453	0.68886	0.81922
10	0.92375	0.93929	1.14111	0.76748	0.91641

These results were collected from the Spectrophotometry software, using the specified wavelengths.

**Table 7.** Absorbance data for Trial 4

Volume of Sunscreen (mL)	630.000nm	664.000nm	647.000nm	750.000nm	635.000nm
0	0.78477	0.786	0.90073	0.64773	0.77825
4	0.63657	0.64033	0.76999	0.50921	0.62911
6	0.52208	0.52131	0.58704	0.42649	0.51739
8	0.69562	0.69508	0.79705	0.56255	0.68828
10	0.45602	0.46113	0.58006	0.35236	0.44918

These results were collected from the Spectrophotometry software, using the specified wavelengths.

**Table 8.** Shapiro-Wilk Normality test results for collected dataset.

Chlorophyll Type	Statistic (W)	p-value	Conclusion
A	0.967	0.698	Normally distributed
B	0.935	0.190	Normally distributed
C1 + C2	0.891	0.028	Not normally distributed

Results were calculated using the SciPy Python package.

**Table 9.** ANOVA test results for collected dataset.

Chlorophyll Type	F-statistic	p-value	Conclusion
A	0.175	0.942	No significant differences
B	5.298	0.048	Significant differences
C1 + C2	2.520	0.169	No significant differences

Results were calculated using the SciPy Python package.

**Table 10.** Tukey's Post-hoc test results for Chlorophyll *a*.

Group1	Group2	Mean Difference	p-adj	Lower	Upper	Reject
0.0	4.0	-0.0543	0.9949	-0.4911	0.3824	False
0.0	6.0	-0.1437	0.8443	-0.5804	0.2931	False
0.0	8.0	-0.0835	0.9744	-0.5203	0.3532	False
0.0	10.0	-0.1659	0.7661	-0.6026	0.2709	False
4.0	6.0	-0.0894	0.9674	-0.5261	0.3474	False
4.0	8.0	-0.0292	0.9995	-0.466	0.4075	False
4.0	10.0	-0.1116	0.9299	-0.5483	0.3252	False
6.0	8.0	0.0601	0.9925	-0.3766	0.4969	False
6.0	10.0	-0.0222	0.9998	-0.459	0.4145	False
8.0	10.0	-0.0823	0.9757	-0.5191	0.3544	False

Results were calculated using the SciPy Python package.

**Table 11.** Tukey's Post-hoc test results for Chlorophyll *b*.

Group1	Group2	Mean Difference	p-adj	Lower	Upper	Reject
0.0	4.0	-0.2512	0.711	-0.8608	0.3584	False
0.0	6.0	-0.3385	0.4547	-0.9481	0.2711	False
0.0	8.0	-0.3582	0.4015	-0.9678	0.2515	False
0.0	10.0	-0.225	0.7836	-0.8346	0.3846	False
4.0	6.0	-0.0873	0.9912	-0.6969	0.5223	False
4.0	8.0	-0.107	0.9813	-0.7166	0.5027	False
4.0	10.0	0.0262	0.9999	-0.5834	0.6358	False
6.0	8.0	-0.0197	1.0	-0.6293	0.59	False
6.0	10.0	0.1135	0.9768	-0.4961	0.7231	False
8.0	10.0	0.1332	0.959	-0.4765	0.7428	False

Results were calculated using the SciPy Python package.

**Table 12.** Tukey's Post-hoc test results for Chlorophyll *c*.

<b>Group1</b>	<b>Group2</b>	<b>Mean Difference</b>	<b>p-adj</b>	<b>Lower</b>	<b>Upper</b>	<b>Reject</b>
0.0	4.0	-0.3622	0.0616	-0.7381	0.0137	False
0.0	6.0	-0.222	0.3967	-0.5979	0.1539	False
0.0	8.0	-0.3438	0.0811	-0.7197	0.0321	False
0.0	10.0	-0.4726	0.0109	-0.8485	-0.0967	True
4.0	6.0	0.1402	0.7773	-0.2357	0.5161	False
4.0	8.0	0.0184	0.9999	-0.3575	0.3943	False
4.0	10.0	-0.1104	0.8898	-0.4863	0.2655	False
6.0	8.0	-0.1218	0.851	-0.4977	0.2541	False
6.0	10.0	-0.2506	0.2869	-0.6265	0.1253	False
8.0	10.0	-0.1288	0.8245	-0.5047	0.2471	False

Results were calculated using the SciPy Python package.

**Table 13.** Pearson and Spearman correlation analysis test results for collected dataset.

<b>Chlorophyll Type</b>	<b>Pearson Correlation</b>	<b>Pearson p-value</b>	<b>Spearman Correlation</b>	<b>Spearman p-value</b>
Chl $a$	-0.286	0.222	-0.339	0.144
Chl $b$	-0.337	0.147	-0.388	0.091
Chl $c$	-0.639	0.002	-0.572	0.008

Results were calculated using the SciPy Python package.