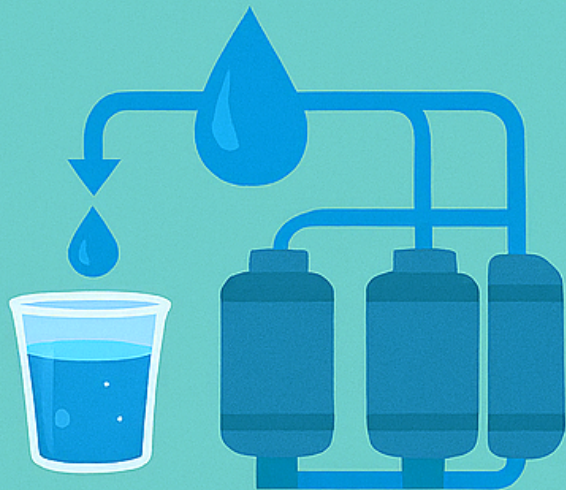
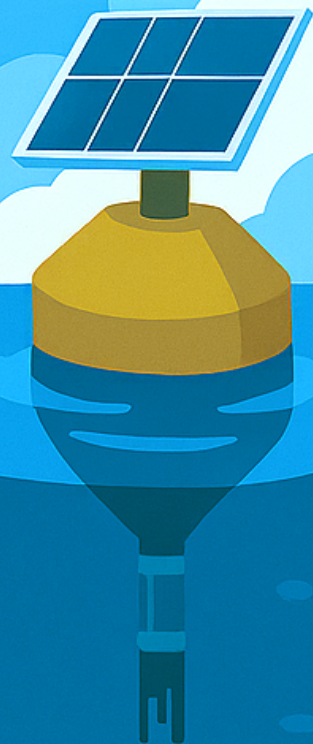


# Clean Water, Smart Science & Health

Category 2



# A Mangrove-Inspired Real-Time Water Quality Monitoring System

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## **Abstract**

Mangrove forests are a defining feature of Indonesia's coastal identity, supporting biodiversity, coastal protection, and community livelihoods. Indonesia contains the largest mangrove area in the world, accounting for roughly 20–23% of global mangrove forests (Spalding et al., 2010). Despite their ecological importance, monitoring environmental conditions in mangrove ecosystems is still often conducted through manual observation and periodic sampling, which may delay the detection of sudden disturbances affecting water quality.

Delayed detection of water quality changes can slow environmental response and allow disturbances to escalate. This study investigates whether a real-time sensor-based monitoring system can improve early detection of water condition changes compared to conventional observation methods.

The research introduces an autonomous monitoring prototype that measures key water quality indicators including pH, Total Dissolved Solids (TDS), and turbidity using integrated sensors connected to a microcontroller. To enable long-term deployment in coastal environments, the device operates using solar energy with a rechargeable battery, is housed in a 3D-printed waterproof casing, and transmits measurements remotely through a radio communication module.

By continuously collecting environmental data, the prototype is expected to provide faster detection of water quality fluctuations and demonstrate how sensor-based monitoring can support more responsive management of vulnerable mangrove ecosystems.

## **Keywords**

Mangrove forest, Water quality monitoring, Autonomous monitoring prototype

## **1 Introduction**

Mangrove forests are among the most important coastal ecosystems in the world due to their role in maintaining biodiversity, protecting shorelines from erosion, storing carbon, and supporting fisheries and coastal livelihoods. Indonesia possesses the largest mangrove forest area globally, representing approximately 20–23% of the world's mangrove ecosystems. These ecosystems provide ecological and economic benefits for coastal communities, particularly in tropical regions where mangroves act as natural barriers against storms, tidal waves, and coastal degradation.

Despite their importance, mangrove ecosystems are increasingly threatened by pollution, coastal development, sedimentation, and climate change. One major concern in mangrove conservation is the decline of water quality caused by waste discharge, excessive sediment accumulation, and environmental contamination. Changes in water quality can negatively affect mangrove biodiversity, aquatic organisms, and the overall stability of coastal ecosystems.

Currently, environmental monitoring in mangrove areas is often conducted through manual observation and periodic water sampling. These traditional monitoring methods are time-consuming and may delay the detection of sudden environmental disturbances. Because measurements are not collected continuously, rapid changes in water conditions may remain undetected until the damage becomes more severe.

Advancements in digital sensor technology and microcontroller systems provide opportunities to improve environmental monitoring through real-time data collection. Sensors capable of measuring pH, Total Dissolved Solids (TDS), and turbidity can continuously monitor changes in water conditions and provide immediate

information regarding environmental quality. When combined with autonomous power systems and wireless communication, these technologies can support long-term environmental monitoring in remote coastal ecosystems.

To address this issue, this research proposes the development of an autonomous water quality monitoring prototype designed specifically for mangrove ecosystems. The system integrates pH, TDS, and turbidity sensors connected to a microcontroller for continuous monitoring. The prototype is powered by solar energy with a rechargeable battery, protected using a waterproof 3D-printed casing, and equipped with a radio communication module for remote data transmission.

By providing continuous and real-time environmental monitoring, this research aims to support faster detection of water quality changes and contribute to more responsive management strategies for vulnerable mangrove ecosystems.

### ***1.2 Problem Statement***

Mangrove ecosystems are highly sensitive to changes in water quality caused by pollution, sedimentation, and environmental disturbances. However, current monitoring practices in many coastal areas still rely on manual sampling and periodic observations that cannot provide continuous environmental data.

As a result, sudden changes in water quality may not be detected immediately, which can delay environmental response efforts and increase ecological damage. In addition, many existing monitoring systems are expensive, require constant human supervision, or are not designed for long-term deployment in remote coastal environments.

Therefore, there is a need for an autonomous, low-cost, and real-time monitoring system capable of continuously evaluating critical water quality conditions associated with mangrove ecosystem health.

### ***1.3 Research Questions***

1. How reliable and stable is the developed Arduino Uno-based monitoring system in continuously measuring pH, TDS, and turbidity within mangrove water environments?
2. How effective is LoRa communication technology in supporting continuous real-time environmental monitoring within remote mangrove ecosystems?

### ***1.4 Research Objectives***

1. To develop and evaluate an Arduino Uno-based autonomous monitoring system capable of continuously measuring pH, TDS, and turbidity within mangrove ecosystems.
2. To evaluate the stability, reliability, and real-time transmission performance of the integrated LoRa communication system for remote mangrove environmental monitoring applications.

### ***1.5 Research Boundaries***

The study focuses only on three water quality parameters: pH, Total Dissolved Solids (TDS), and turbidity.

- The prototype is designed specifically for monitoring water conditions in mangrove ecosystems.
- The system uses a microcontroller-based monitoring platform.
- Power supply is limited to solar energy and rechargeable battery storage.
- Data transmission is limited to radio communication modules.
- The research focuses on prototype development and monitoring performance evaluation only.

## **1.6 Research Benefits**

### **Theoretical Benefits:**

1. Contributing to research on the application of sensor technology in environmental monitoring systems.
2. Expanding scientific understanding of autonomous monitoring systems for coastal ecosystem management.
3. Supporting studies related to real-time environmental monitoring in mangrove ecosystems.

### **Practical Benefits:**

1. Providing a low-cost prototype for continuous mangrove water quality monitoring.
2. Supporting faster environmental response through real-time monitoring data.
3. Assisting environmental conservation efforts in vulnerable coastal ecosystems.
4. Serving as an educational model for integrating environmental science with digital technology.

## **2 Literature Review**

### **2.1. Digital Sensors**

#### **2.1.1 Digital Sensors in Environmental Monitoring Systems**

Previous studies have demonstrated that digital sensors are widely utilized in environmental monitoring systems due to their ability to provide continuous and real-time measurements with relatively low signal distortion (Sensorex, n.d.). According to O'Donnell (2022), digital sensor systems are commonly applied in wastewater treatment and water quality monitoring because they support faster data acquisition and improved transmission efficiency compared to conventional monitoring approaches.

Recent advancements in microcontroller integration and wireless communication technologies have further expanded the use of digital sensors in autonomous environmental monitoring systems, particularly within remote ecosystems where

continuous manual observation is difficult to maintain.

#### **2.1.2 Ideal Water Quality Conditions for Mangrove Ecosystems**

Mangrove ecosystems require relatively stable water quality conditions to maintain ecosystem health, biodiversity, and nutrient cycling processes. According to Alongi (2008), water quality parameters such as pH, salinity, and suspended sediment concentration strongly influence mangrove productivity and ecosystem stability.

pH plays an important role in regulating biological and chemical reactions within mangrove waters. Most mangrove ecosystems develop optimally under slightly acidic to slightly alkaline conditions, generally ranging between pH 6.5 and 8.5.

Total Dissolved Solids (TDS) reflect dissolved ion concentration and salinity conditions within water systems. Excessive dissolved solid concentrations may disrupt aquatic organism survival and environmental balance. Similarly, high turbidity levels may reduce sunlight penetration and negatively affect photosynthetic activity within mangrove ecosystems (Khan et al., 2020).

Therefore, monitoring pH, TDS, and turbidity is important for evaluating mangrove ecosystem conditions and detecting early environmental disturbances.

### **2.2 Sensor Principles, Accuracy and Limitations.**

#### **2.2.1 pH Sensor**

A pH sensor measures a solution's acidity or alkalinity on a 0 to 14 scale—where below 7 is acidic, 7 is neutral, and above 7 is alkaline—which dictates its chemical behavior across various processes (KACISE®, n.d.). Measurement accuracy relies heavily on managing controllable factors, such as performing frequent calibrations

with fresh buffer solutions and minimizing temperature differences between the buffer and the sample. With a well-hydrated sensor and properly grounded, high-impedance equipment, realistic accuracies range from +/- 0.03 to +/- 0.05 pH units, while strictly controlled lab conditions can achieve accuracies of +/- 0.02 pH units (Hamilton Company, n.d.).

### **2.2.2 TDS Sensor**

Total Dissolved Solids (TDS) are the dissolved substances present in water which influence its taste, safety and usability, hence monitoring TDS is essential for maintaining clean water standards, especially since industrialization and population growth has increasingly threatened water quality (Adjovu, Stephen, James, & Ahmad, 2023). TDS sensors can accurately distinguish between clean and dirty water, achieving an average percentage error accuracy of only 0.008894%, with 7 out of 12 water samples meeting clean-water criteria based on PPM values. Its limitations, however, still exist. As sensor performance may vary under different environmental conditions, and improvements in sensor reliability, calibration and integration with other monitoring technologies are needed. (Setiawan, Elmi, Zukhruf Z., & Juniani, 2024)

### **2.2.3 Turbidity Sensor**

Turbidity measurements monitor particle mixtures in various reactions and require calibration with a formazine standard per ISO 7027:1999. Although simple and inexpensive, these probes have significant accuracy limitations. Their reliability is hindered by environmental sensitivities (such as temperature, stray light, and mechanical disturbances), early signal saturation at higher concentrations, and an inability to separate absorption from scattering effects. Additionally, the formazine standard itself has stability issues. Consequently, this environmental sensitivity and limited measurement range make turbidity probes less reliable than advanced methods like PDW spectroscopy (Mützenberg, Hass, Khanh, & Reich, 2016).

## **2.3 Hypothesis**

An autonomous real-time sensor-based monitoring system measuring pH, TDS, and turbidity is expected to provide continuous environmental monitoring and support earlier detection of water quality fluctuations within mangrove ecosystems.

## **2.4 Research Gap**

Previous studies have demonstrated the potential of digital sensors and IoT-based systems for environmental water quality monitoring. Several monitoring systems have successfully integrated pH, TDS, and turbidity sensors to evaluate aquatic environmental conditions in real time. In addition, wireless communication technologies have increasingly been utilized to improve remote environmental monitoring capabilities.

However, many existing monitoring systems remain limited by high implementation costs, dependence on stable internet infrastructure, limited autonomous operation, or lack of suitability for long-term deployment within remote coastal ecosystems such as mangrove environments. In addition, limited studies have evaluated the stability and reliability of low-cost sensor systems under saline and high-sediment mangrove environmental conditions.

Therefore, this research proposes the development of an autonomous Arduino Uno-based monitoring system capable of continuously measuring pH, TDS, and turbidity while transmitting environmental data remotely through LoRa communication technology. The proposed system aims to support low-cost, real-time, and long-range environmental monitoring specifically designed for mangrove ecosystem applications.

### **3 Methodology, System Development, and Results**

#### **3.1 Research Design**

The objective of this research is to design and develop an autonomous monitoring device that is capable of continuously measuring pH, Total Dissolved Solids (TDS), and turbidity. Furthermore, the system aims to facilitate real time environmental management by automatically transmitting the collected data wirelessly to a mobile interface to allow users to monitor water conditions remotely and respond promptly to environmental fluctuations.

#### **3.2 Research Methodology**

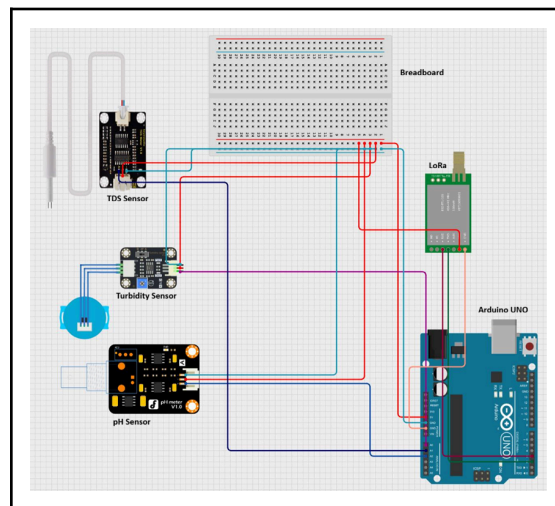
The research methodology was structured around the development, deployment, and evaluation of the autonomous water quality monitoring prototype.

Water samples were collected from mangrove coastal areas located in Tanjung Pasir, Tangerang Regency, Banten, Indonesia. Sampling was conducted within shallow mangrove water channels influenced by tidal and sedimentary environmental conditions.

##### **3.2.1 System Design**

The prototype was built around an Arduino Uno microcontroller, which functioned as the central processing unit responsible for processing continuous environmental monitoring data. Three sensors were integrated into the system, consisting of a pH sensor, a Total Dissolved Solids (TDS) sensor, and a turbidity sensor. A LoRa communication module was also integrated to enable long-range wireless data transmission for real-time environmental monitoring applications.

The system was powered using a battery supply and enclosed within a waterproof protective casing to minimize environmental exposure and prevent water intrusion into the electrical components during field deployment.



### ***3.2.3 Sensor Calibration and Baseline Testing***

Before environmental measurements were conducted, all sensors were calibrated and tested to improve measurement accuracy and reliability. Standard buffer solutions with known pH values were used to calibrate the pH sensor and establish baseline measurement accuracy. This calibration process ensured that the sensor produced readings consistent with standard reference values.

The TDS sensor was calibrated through comparative measurements using a commercial laboratory-grade TDS meter as the reference instrument. Calibration adjustments were performed to minimize measurement deviation and improve ppm measurement consistency.

The turbidity sensor was validated through comparative testing under different water clarity conditions, including clear and high-sediment water samples. This procedure was conducted to verify the sensor's ability to distinguish variations in turbidity levels during environmental monitoring applications. During continuous monitoring trials, sensor measurements were recorded every 10 seconds over a 60-second observation period to evaluate sensor stability, repeatability, and short-term measurement consistency. A 60-second monitoring duration was selected to evaluate short-term sensor stability and transmission consistency during continuous environmental monitoring.

### ***3.2.4 TDS Sample Dilution Procedure***

Due to the original mangrove water samples exceeding the maximum measurement range of the TDS sensor, a serial dilution procedure was performed prior to measurement.

Initially, the mangrove water samples were diluted using distilled water at a 1:10 ratio to reduce dissolved solid concentration. However, since the resulting measurements still approached the upper detection limit of the sensor, a second 1:10 dilution was conducted using the previously diluted sample.

This resulted in an overall dilution factor of 1:100 relative to the original mangrove water sample concentration.

The serial dilution procedure was applied consistently across all samples to maintain measurement uniformity and minimize experimental variability during sensor analysis.

### ***3.2.5 Warning and Detection System***

The monitoring system was programmed to compare sensor measurements against predefined ideal environmental ranges for mangrove ecosystems. When sensor readings exceeded the recommended threshold ranges, warning indicators were automatically generated to notify users regarding potentially unfavorable environmental conditions.

The warning system operated by continuously evaluating pH, TDS, and turbidity measurements against predefined environmental thresholds stored within the Arduino Uno microcontroller. For pH measurements, values below 6.5 or above 8.5 were classified as outside the ideal mangrove water quality range. Similarly, abnormal increases in TDS concentration and excessively high turbidity levels were treated as indicators of potential environmental disturbance.

Once abnormal conditions were detected, the monitoring system transmitted warning notifications together with real-time sensor data through the LoRa communication module. This feature enables early detection of water quality changes that may negatively affect mangrove ecosystem stability and supports continuous remote environmental monitoring applications.

**Table 3.2.5. Warning Threshold**

Parameter	Ideal Range	Warning Conditions
pH	6.5-8.5	<6.5 > or 8.5
TDS	Stable moderate levels	Sudden abnormal increase
Turbidity	Low-moderate	Excessively high turbidity

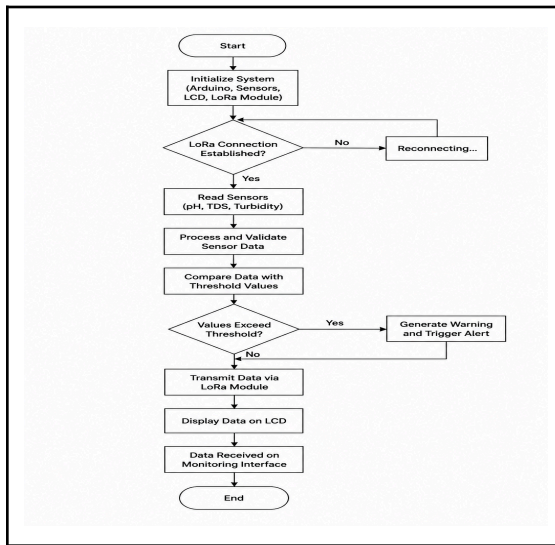


Figure 3.2.5. Operational workflow of the autonomous mangrove water quality monitoring system.

Figure 3.2.5 illustrates the operational workflow of the monitoring system, beginning from environmental data acquisition through sensor measurements, followed by data processing, threshold comparison, warning detection, LoRa transmission, and remote user monitoring.

### 3.3 Results

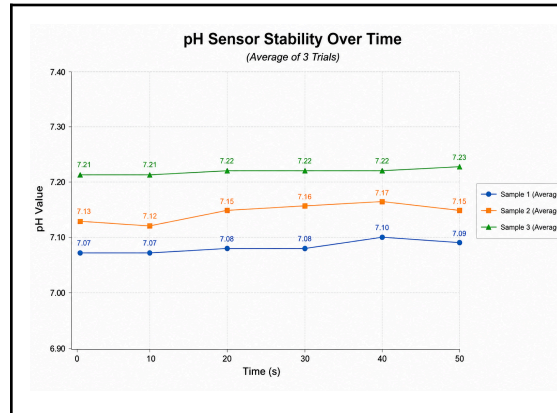


Figure 3.3.1. pH Sensor Stability Over Time

Figure 3.3.1 presents the stability profile of average pH measurements obtained from three mangrove water samples during a 60-second observation period. The graph was included to evaluate the temporal consistency and repeatability of the Arduino Uno pH sensor during continuous environmental monitoring.

Sample 3 consistently produced the highest pH values, indicating slightly more alkaline conditions compared to the other samples, while Sample 1 showed the lowest measurements. Only minor fluctuations were observed throughout the monitoring period, suggesting that the pH sensor maintained relatively stable and consistent measurements over time.

**Table 3.3.1. pH Sensor Analysis**

Sample	Reference pH av.	Arduino pH	%Error
Sample 1	6.62	7.11	7.40%
Sample 2	6.62	7.18	8.46%
Sample 3	6.74	7.24	7.42%

#### Reliability Statistics

Standard Deviation (SD): 0.065

Coefficient of Variation (CV): 0.91%

The pH sensor produced percentage error values ranging from 7.40% to 8.46%, indicating acceptable agreement with the reference instrument. The low standard deviation value (0.065) and coefficient of variation value (0.91%) indicate minimal measurement fluctuation and strong sensor consistency during repeated monitoring. These results suggest that the pH sensor was capable of producing stable real-time environmental measurements.

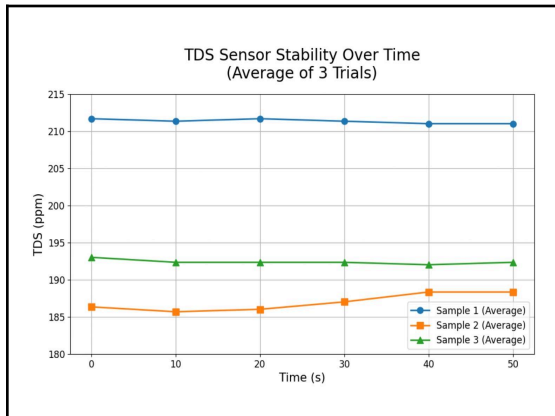


Figure 3.3.2. TDS Sensor Stability Over Time

Figure 3.3.2 illustrates the stability profile of average TDS measurements obtained from three mangrove water samples throughout the monitoring period. Sample 1 consistently produced the highest TDS values, indicating a greater concentration of dissolved solids compared to the other samples.

Minor fluctuations were observed during repeated measurements. The larger variability observed in the TDS sensor may be associated with conductivity sensitivity limitations commonly observed in low-cost TDS sensors, particularly under saline or particle-rich environmental conditions. Variations in dissolved ion concentration and suspended particles within the mangrove water samples may also have contributed to the observed measurement fluctuations. Despite these variations, the overall measurement trends remained relatively stable throughout the monitoring period.

Table 3.3.2. TDS Sensor Analysis

Sample	Reference TDS ppm	Arduino TDS ppm	%Error
Sample 1	246.00	211.40	14.07%
Sample 2	201.00	188.40	6.27%
Sample 3	218.67	192.30	12.06%

### Reliability Statistics

Standard Deviation (SD): 12.309

Coefficient of Variation (CV): 6.24%

The TDS sensor produced percentage error values ranging from 6.27% to 14.07%, indicating moderate agreement with the reference instrument. The higher standard deviation value (12.309) and coefficient of variation value (6.24%) indicate greater variability compared to the pH sensor. This variability may have been influenced by dissolved ion concentration changes and conductivity sensitivity limitations commonly observed in low-cost TDS sensors under saline environmental conditions. Nevertheless, the overall measurement trends remained relatively stable throughout repeated monitoring.

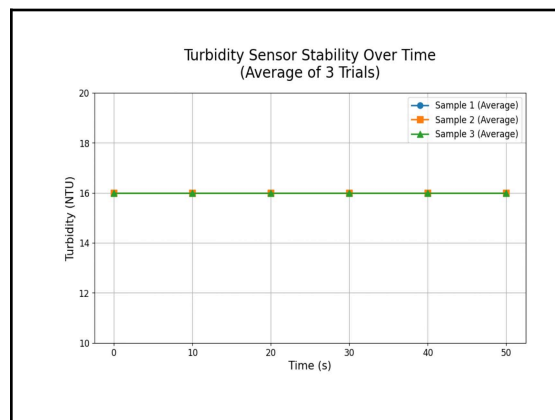


Figure 3.3.3. Turbidity Sensor Stability Over Time

Figure 3.3.3 presents the stability profile of turbidity measurements recorded from three mangrove water samples during continuous monitoring. All samples consistently produced identical turbidity values of 16 NTU throughout the observation period.

The absence of significant fluctuation indicates very high sensor stability and repeatability during repeated measurements. The identical measurements may also suggest that the tested samples possessed relatively similar suspended particle concentrations during the monitoring period.

**Table 3.3.3. Turbidity Sensor Analysis**

Sample	Arduino Turbidity NTU
Sample 1	16
Sample 2	16
Sample 3	16

The turbidity sensor consistently produced identical readings of 16 NTU across all tested samples. The absence of fluctuation indicates very high measurement stability, repeatability, and reliability during continuous monitoring. The consistent measurements may also indicate relatively similar suspended particle concentrations among the tested mangrove water samples.

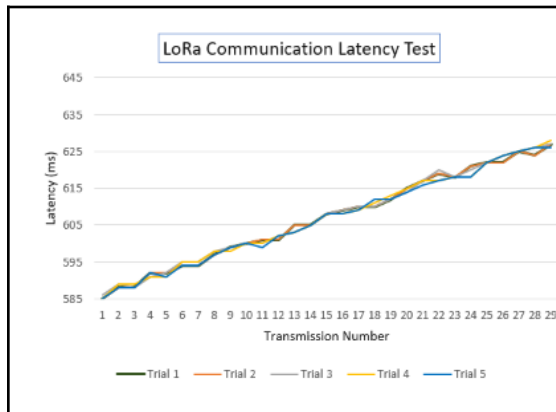


Figure 3.3.4. Stability Profile of LoRa Communication Latency During Repeated Wireless Transmission Trials

Figure 3.3.4 illustrates the stability profile of LoRa communication latency during repeated wireless transmission trials. The highly overlapping trends observed across all five trials indicate strong

consistency and repeatability of the wireless communication system.

Only minimal latency fluctuations were observed throughout repeated transmissions, demonstrating relatively stable wireless communication performance. These results suggest that the LoRa module was capable of maintaining reliable real-time environmental data transmission during continuous monitoring operations.

**Table 3.3.4. Statistical Results of LoRa Communication Latency**

Trial	Mean Latency (ms)	SD	CV
Trial 1	606.90	12.57	2.07%
Trial 2	606.90	12.57	2.07%
Trial 3	607.07	12.59	2.07%
Trial 4	606.90	12.59	2.08%
Trial 5	606.62	12.58	2.07%

The LoRa communication latency analysis demonstrated highly stable wireless transmission performance across all repeated trials. The low standard deviation and coefficient of variation values indicate minimal fluctuation between latency measurements, while the highly overlapping trends further support the consistency and repeatability of the communication system. These findings suggest that the LoRa module was capable of maintaining reliable real-time environmental data transmission during continuous monitoring operations.

**Overall Sensor Performance Analysis**

Based on the graphical and statistical analysis, the developed Arduino Uno-based monitoring system demonstrated satisfactory stability and repeatability during continuous environmental measurements. The relatively small fluctuations observed in the pH and turbidity sensors indicate strong measurement consistency, while the TDS sensor exhibited slightly greater variability under saline environmental conditions.

Nevertheless, the overall performance of the integrated monitoring system remained relatively stable throughout repeated testing. The demonstrated stability of the sensor system and the reliability of the LoRa communication module suggest that the developed autonomous prototype possesses strong potential for supporting continuous real-time monitoring within vulnerable mangrove ecosystems. By enabling faster detection of environmental fluctuations, the system may contribute to more responsive conservation and environmental management strategies in coastal environments.

### **3.4 Limitations**

Several limitations were identified during this study. The TDS sensor possessed limited measurement capability under highly concentrated saline conditions, requiring serial dilution prior to testing. In addition, environmental factors such as dissolved particle movement, temperature variation, and sensor sensitivity may have influenced measurement stability during monitoring.

The study was also limited by relatively short observation duration and a restricted number of sampling locations, which may not fully represent long-term environmental variability within mangrove ecosystems. Future research should involve extended monitoring duration, additional sampling locations, and higher-precision environmental sensors to further improve system accuracy and reliability.

### **4 Conclusion**

This research successfully developed an autonomous Arduino Uno-based monitoring system capable of continuously measuring pH, Total Dissolved Solids (TDS), and turbidity within mangrove water environments. The developed prototype demonstrated relatively stable sensor performance and reliable LoRa communication during repeated environmental monitoring trials,

indicating its capability to support continuous real-time monitoring within mangrove ecosystems.

Overall, the proposed system demonstrates strong potential as a low-cost environmental monitoring solution for supporting earlier detection of water quality fluctuations and improving conservation efforts within vulnerable coastal ecosystems. Future research may involve longer monitoring duration, additional environmental parameters, and broader field deployment to further improve monitoring accuracy and system reliability.

### **Acknowledgements**

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# The Biofiltration Efficacy of Tanjung Pasir Mangroves in Mitigating Nitrate Loading and Stabilizing pH from the Cisadane River

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## Abstract

In 2025, over 70% of Indonesia's rivers are polluted by anthropogenic waste, which eventually flows into coastal waters and accumulates in marine ecosystems, increasing the risk of eutrophication, algal blooms, and hypoxia. Data from the Cisadane River bank in Tanjung Burung showed high nitrate (41.7 ppm) and toxic nitrite levels (1 ppm), indicating incomplete denitrification and poor water quality.

These conditions highlight the importance of natural coastal filters, especially mangrove forests. Indonesia has around 3.4 million hectares of mangroves, about 23% of the world's mangroves. Mangroves improve water quality by trapping pollutants and supporting processes such as denitrification and nutrient uptake. Our results showed significantly better water conditions in mangrove areas, including lower nitrate levels (10 ppm), and pH remained stable at 8.4 due to seawater carbonate (40 ppm), which helps bind heavy metals and phosphate.

However, mangrove forests are declining by around 0.57% annually because of forest degradation and climate change. This study demonstrates how river pollution directly affects coastal ecosystems and shows that mangrove rehabilitation can reduce pollutants and help prevent eutrophication. Parameters tested included temperature, salinity, pH, TDS, BOD, COD, nitrite, carbonate, nitrate, and phosphate.

## Keywords

*Mangrove rehabilitation, environmental sustainability, water quality*

## 1. Introduction

Indonesia is a maritime country with a coastline stretching over 108,000 km and a water area of approximately 6.4 million km<sup>2</sup> [1][2]. However, these marine areas are highly vulnerable to pollutants from anthropogenic activities such as the disposal of household, industrial, and agricultural wastes into the river, as over 70% of Indonesia's rivers are polluted with anthropogenic wastes such as nitrate [3]. These pollutants will eventually flow into the sea. This condition causes damage to the marine ecosystem by disrupting the

wildlife and reducing water quality by increasing the risk of eutrophication, algal blooms, and hypoxia [4].

Indonesia's geographical conditions make it rich in marine biodiversity and coastal ecosystems such as mangrove forests, coral reefs, and seagrass beds. Indonesia has the largest area of mangrove forests in the world, covering about 3.5 million hectares or about 23% of the world's mangroves [5]. Nonetheless, the area of mangrove forests is decreasing at an alarming rate, by about 19,501 hectares annually, due to forest degradation and climate change [6]. This decline in mangrove areas not only affects the decrease in biodiversity but also the social welfare of coastal communities, especially fishermen, including those that do not have access to basic necessities, as they are highly dependent on the availability of marine resources and natural sources of clean water [7][8].

Damage to marine ecosystems due to pollution, the reduction and damage in mangrove forests, poses a challenge for nations to achieve the Sustainable Development Goals (SDGs), specifically SDG 6.3 and 14. SDG 6.3 focuses on water quality by reducing pollution, minimizing the release of hazardous materials, and improving wastewater treatment. SDG 14 aims to conserve and protect coastal ecosystems and biodiversity, as well as increase the economic benefits through fisheries and aquaculture management [9][10]. Mangrove forest conservation needs to be done immediately. In line with Pope Francis' call in the *Laudato Si* encyclical letter, article 48, which emphasizes the importance of human awareness and responsibility to act in efforts to preserve the environment sustainably [11].

Mangrove roots provide a breeding ground for an array of microorganisms. The dense roots themselves act as a buffer for pollution, causing the speed of rushing water to decrease as it passes through the roots, causing the pollutants to settle in the soil rather than in the sea. Mangrove roots also act as bioremediators for nutrients such as phosphate, nitrates, and ammonia. The uptake of excess nutrients greatly decreases the chances of eutrophication occurring [12][13].

## 2. Purpose of the Investigation

This research aims to evaluate the ability of mangrove ecosystems as a naturally occurring biofilter, which is achieved by measuring, analyzing, and comparing the differences in water quality parameters of the Cisadane River and water quality after passing through the mangrove population. By doing this, the role of mangroves as biofiltering agents can be determined quantitatively.

## 3. Methodology

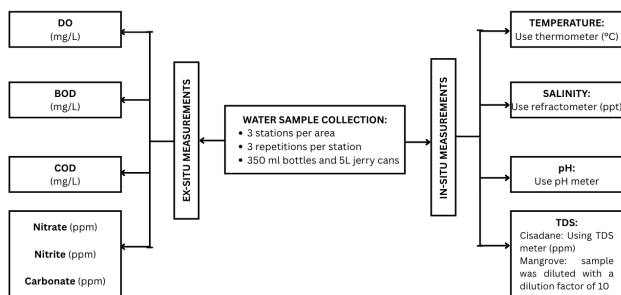


Figure 1: Flow diagram of water sampling and measurement parameters

### 3.1 Preparation of Water Sample

Samples of water were collected from three sampling stations for each area (Tanjung Pasir Mangrove and Cisadane River), with three repetitions at each station. Sampling was carried out using 350 ml bottles and 5L jerry cans.



Figure 2: Mangrove water sample collection

### 3.2 Determining Temperature

A thermometer was prepared and used for in-situ measurements.

### 3.3 Determining Salinity

A refractometer was prepared and used for in-situ measurements. The refractometer is first calibrated by putting a drop of aquadest onto the prism and closing the cover, making sure that the boundary line reads 0 ppt (parts per thousand). After that, wipe it dry, then apply a drop of the water sample onto the prism. Point the refractometer

toward a light source and read the salinity value where the boundary line intersects the scale. Finally, rinse the prism with aquadest and dry it after each use to prevent cross-contamination.

### 3.4 Determining pH Levels

A pH meter was prepared, and the tip of the tester was inserted into the liquid to be tested. The readings on the display were recorded once they stabilized. The pH meter should be washed and dried after each use to ensure accuracy.

### 3.5 Determining Total Dissolved Solids (TDS)

A TDS meter was prepared, and the tip of the tester was inserted into the liquid to be tested. The readings on the display were recorded once they stabilized. The TDS of water is expressed in parts per million (ppm). The TDS meter should be washed and dried after each use to ensure accuracy.

### 3.6 Determining Dissolved Oxygen (DO) and Biochemical Oxygen Demand (BOD)

#### 3.6.1 Sample Preparation

The 350 ml bottle was submerged and allowed to be filled completely before the cap was securely tightened underwater to prevent any air bubbles from being trapped. Immediately after collection at each station, 2 ml of  $\text{MnSO}_4$  (0.101M) solution and 2 ml of Alkaline-Iodide-Azide solution were added to the sample. This addition aims to fix the dissolved oxygen content, ensuring that the oxygen concentration during transport from the mangrove area to the laboratory remains unchanged. Besides that, water from each station was also collected in separate 5L jerry cans for 5 days for final DO ( $\text{DO}_5$ ) measurements.

#### 3.6.2 Thiosulphate ( $\text{S}_2\text{O}_3$ ) Titration



Figure 3: Water sample BOD preparation

50 mL of the sample was measured using a volumetric pipette and relocated into a 300 mL Erlenmeyer. 5 drops of sodium thiosulfate ( $\text{Na}_2\text{S}_2\text{O}_3$ ) solution (0.025 M) were titrated into the sample while swirling the Erlenmeyer,

creating a straw-colored solution. This was done to eliminate the excess iodine. 1 mL of 1% amyllum or starch solution was added to the Erlenmeyer, causing the solution to turn a deep blue color. Essentially, the blue color becomes the indicator of the presence of iodine, which, in the Winkler Method, is released in the same proportion as the dissolved oxygen. Sodium thiosulfate is titrated into the Erlenmeyer until the blue color disappears, and the solution turns colorless and clear. This process was repeated 3 times for each location of the sample collection. The total volume of sodium thiosulfate used during the process is used to determine the concentration of dissolved oxygen present in the sample using the formula:

$$DO \text{ (ppm)} = \frac{V \times N \times 8000}{\text{aliquot volume (mL)}} \quad (1)$$

After 5 days,  $DO_5$  is recorded, and the BOD can be calculated using the formula:

$$BOD_5 = DO_0 - DO_5 \quad (2)$$

### 3.7 Determining Chemical Oxygen Demand (COD)

100 mL of the sample was relocated to a 300 mL Erlenmeyer. 10 mL of  $H_2SO_4$  (8M) and 10 mL of  $KMnO_4$  (0.01M) were added and swirled, and the solution was heated until its color changed from fuchsia to a brownish red. After 30 minutes, 10 mL of  $Na_2C_2O_4$  (0.00625M) was added to the solution.  $KMnO_4$  was then titrated into the solution until it turned a light pink color. This process was repeated 3 times for each location of the sample collection. The total volume of  $KMnO_4$  used during the process is used to determine the chemical oxygen required to oxidize the organic matter present using the formula:

$$COD \text{ (ppm)} = \frac{(V_1 - V_0) \times C \times 8000}{V_s} \quad (3)$$



Figure 4: Water sample COD preparation and titration

### 3.8 Nitrate, Nitrite, and Carbonate

Test strips were prepared and dipped in each bottle for a few seconds, with results recorded after the paper had dried. The nitrate, nitrite, and carbonate levels were determined using an indicator scale provided on the packet. This process was conducted as soon as the samples were obtained.

## 4. Results and Discussion

### 4.1 Water Parameters Results

Table 1. Water parameters of each sample

Parameters	Water Samples	
	Tanjung Pasir Mangrove	Cisadane River
Total Dissolved Solids (TDS)	8333.33	165.5
Temperature (°C)	31.3	28.3
Salinity (ppt)	2.5	0
Nitrite (ppm)	1	1
Nitrate (ppm)	10	41.667
pH	8.4	6.273
Carbonate (ppm)	40	13.333
BOD (ppm)	3.343	2.333
Initial Dissolved Oxygen (ppm)	4.454	3.223
COD (ppm)	166.064	120.884

### 4.2 Analysis

#### 4.2.1 Tanjung Pasir Mangroves

Based on the data, mangrove waters have a higher BOD of 3.343 ppm, which is caused by the constant cycle of shedding of natural organic carbon, such as leaves, roots, and branches, which increase the concentration of organic matter, indicating that a significant amount of oxygen is consumed by aerobic microorganisms. High BOD in mangroves signifies that there is more organic matter available for degradation by microbes. It is important to note that the decomposition performed by anaerobic microorganisms is not recorded in the BOD value, meaning that the total organic matter processed by the ecosystem is higher than what the BOD alone can reflect.

The high level of organic input prompts dense microbial communities, which contribute to nutrient cycling and organic material decomposition. Mangroves have a high TDS value of 8333.3 ppm alongside relatively clear water, suggesting low TSS and that most of the organic matter is dissolved rather than suspended. A significant contributor to TDS is salinity, measured at 2.5 ppt, classifying the water as low-salinity brackish water (oligohaline). The sodium and chloride ions from the water significantly increase the TDS. However, high TDS can reflect organic matter, microbial degradation byproducts, and dissolved pollutants.

In the nutrient cycle, nitrogen is permanently lost through denitrification, which is when anaerobic bacteria use nitrate as an electron acceptor for respiration, while consuming organic carbon as their source of energy. Nitrification produces nitrate as a byproduct of ammonium oxidation that accumulates in the water column, which subsequently becomes available for denitrifying bacteria. Though it does not affect COD directly, nitrification exerts additional oxygen demand during the oxidation of ammonium to nitrate, contributing to the depletion of DO in the water column. Since the COD in mangroves is at a higher level while the nitrate level is low, this suggests that the microbes are actively consuming nitrate in denitrification as an electron acceptor, effectively removing nitrogen from the environment as  $N_2$  gas while COD remains high due to the presence of organic matter. High COD indicates the presence of a large amount of oxidizable organic and inorganic matter in the water.

Theoretically, the pH of the water would be lower and more acidic due to this microbial activity, as the microbes in mangrove waters are very active in decomposing organic matter. However, based on the data, mangrove waters are more basic and higher in pH. The water in mangrove ecosystems such as Tanjung Pasir has high salinity, meaning that it is brackish and is partly made up of seawater. Seawater has a natural carbonate-bicarbonate buffering system where excess  $H^+$  ions absorbed from  $CO_2$  are neutralized using carbonate ions ( $CO_3^{2-}$ ) to form bicarbonate ( $HCO_3^-$ ), preventing acidification. Based on the data, mangrove waters have high levels of carbonate (40 ppm), which then acts as the buffer that neutralizes acids sourced from microbial decomposition and  $CO_2$  dissolved within the system, maintaining alkaline conditions, creating ideal conditions for marine biota to improve immunity and support shell growth on shellfish by providing carbonate ions.

The level of dissolved oxygen (DO) in mangroves, 4.454 ppm, indicates that there is photosynthetic activity performed by algae and plants, as well as the slight movement of water during high tide and low tide, which is crucial for the biodiversity of the area. High BOD and microbial activity are suppressing DO. The DO concentration in mangroves indicates that the ecosystem in Tanjung Pasir is able to perform tidal re-aeration to replenish oxygen, maintaining ideal conditions for biodiversity. The nitrate levels in mangroves are low, showing a 75% significant decrease compared to nitrate levels in Cisadane. This indicates that mangroves can function as a biological filter, removing nitrogen from the ecosystem. The temperature in mangroves is also around  $3^\circ C$  higher than temperatures in Cisadane, which influences microbial decomposition rates, which subsequently affects DO levels.



Figure 5: Mangrove sampling site

#### 4.2.2 Cisadane River

Cisadane is a river that runs through heavily urbanized, industrialized, and agricultural areas, where toxic runoff, domestic waste, and industrial discharge are discarded into the water. Because of this, the water in the Cisadane River has a nitrate concentration of 41.667 ppm, which critically exceeds the standards of river water quality classification 2 of 10 ppm (Class 2). This level of nitrate indicates the presence of nitrification and a slower rate of denitrification, which is suppressed due to the continuous water flow that incorporates atmospheric oxygen into the water and maintains aerobic conditions in the water, as reflected in the measured DO, which is not in line with the anoxic conditions required for denitrification to occur effectively, though the primary nitrogen load originates from anthropogenic waste. High levels of nitrate are the main risk factor for algal blooms, which rapidly deplete dissolved oxygen and create hypoxic conditions.

The DO of Cisadane is classified as Class 3 under Indonesia's Government Regulation No. 22 of 2021, with a minimum requirement of 3 ppm, with a measurement of 3.223 ppm. This means that the water is considered acceptable for fisheries, animal husbandry, and agriculture, but is too degraded for direct human consumption. The low DO measurement reflects the high oxygen consumption used by aerobic microorganisms for decomposing pollutants as well as nitrification processes, leaving less dissolved oxygen available in the water. Nitrification releases  $H^+$  ions and consumes oxygen, reducing the DO and pH of the water.

The BOD of Cisadane is recorded at 2.3 ppm, which meets the minimum requirements of Class 2 (requiring a maximum of 3 ppm) under Indonesia's Government Regulation No. 22 of 2021, showing that the level of biodegradable organic pollution is within acceptable levels. However, BOD only captures oxygen demand from biologically degradable material, while non-biodegradable pollutants are recorded in the COD. The COD is recorded at 120.884 ppm, which is considerably high because of the high level of pollutants available in the water.

The TDS of the Cisadane River is relatively low despite high pollution, which might sound contradictory, but is explained by the nature of the pollutants. Inorganic wastes

and heavy metals contribute to TDS, while plastics and solid waste may not dissolve or decompose directly. Though it contributes to TSS, it may release dissolved chemicals or microplastics, which will contribute to TDS over time. Agricultural waste, such as pesticides or fertilizers that contain nitrates and phosphates, can raise TDS, but part of it is organic. However, this is offset by the high water volume and flow of water, which also constantly dilutes dissolved solids, keeping the TDS reading low despite heavy contamination. The visibly murky water is due to high TSS or total suspended solids, which comes from floating waste and sediment, rather than dissolved solids.

The pH of Cisadane is quite low, which is due to low carbonate levels of 13.3 ppm and low alkalinity, which, in turn, has weak carbonate-bicarbonate buffering capacity. Cisadane also does not have saltwater influence, this being one of the reasons why carbonate ions cannot be replenished, and the ones that do enter the water are consumed by the high acid load. The high amounts of organic pollutants drive heterotrophic aerobic microbial respiration, producing large quantities of CO<sub>2</sub>, which dissolves into H<sub>2</sub>O to form carbonic acid, lowering pH.



Figure 6: Cisadane water sample collection

#### 4.2.3 Comparison

Mangrove water has a higher BOD (3.34 mg/L) than Cisadane River (2.33 mg/L) due to the accumulation of littered mangrove leaves and branches that increase natural organic matter. Despite the higher BOD, mangrove areas showed lower nitrate levels (10 mg/L) compared to the Cisadane River (41.7 mg/L), reflecting a reduction by around 75%. This indicates that mangrove ecosystems have the ability to retain nutrients, facilitating the removal of nitrate through denitrification by microbes. In contrast, the high nitrate levels in Cisadane River may increase the risks of eutrophication, algal blooms, and hypoxic conditions.

Mangrove water is more alkaline and higher in pH (pH 8.4) due to the carbonate-bicarbonate buffering system which is supported by the high concentration of carbonate (40 mg/L) in the water. Conversely, the river, lacking this buffer system and containing excess CO<sub>2</sub> from decomposing waste, shows acidic conditions (pH 6.27). Dissolved oxygen (DO) concentration is higher in mangrove areas (4.45 mg/L) than in Cisadane River (3.22 mg/L) due to photosynthetic activity and tidal re-aeration cycles. Both

locations have 1 mg/L nitrite, which exceeds the Class 2 water quality standard according to Indonesia's Government Regulation No. 22 of 2021 with a maximum concentration of 0.06mg/L, indicating water quality concerns. However, microbes in mangrove areas may facilitate denitrification which converts nitrite into nitrogen gas through denitrification, unlike the water system in Cisadane River.

## 5. Conclusion

In conclusion, the data obtained from Tanjung Pasir suggests that mangroves can function as a naturally and locally occurring biofilter within coastal environments. Mangroves act as a nutrient trap, lowering nitrate levels before river water enters the sea, which may help reduce the risk of eutrophication in coastal waters. Moreover, mangroves can function as a temporary sink for pollutants and organic matter. The high COD also indicates that oxidizable pollutants or organic matter is present in the water, which may be decomposed by microbes or undergo sedimentation. In addition, mangroves maintain pH and stable alkaline conditions through carbonate-bicarbonate buffering systems. Mangroves maintain dissolved oxygen concentrations at around 4.45 ppm, indicating that photosynthetic activity and re-aeration processes are still happening alongside microbial activity. Despite the water being only about one metre deep, this DO level is enough for fish and other aquatic organisms to thrive.

Further research should investigate the biodiversity within the mangrove ecosystem, such as microbial communities, fish, and shellfish, to better understand the correlation of biodiversity and water purification processes.

Coastal management should consider prioritizing mangrove restoration as a low-cost and local solution for water quality improvement and conserving coastal habitats.

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# Water is Life, Sustainability is Future: Green Photocatalyst from Rubberwood Biochar–TiO<sub>2</sub> for Wastewater Purification

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## Abstract

Water pollution from textile dyes poses serious environmental and public health risks due to the chemical stability and bioresistance of synthetic colorants. This study developed a green photocatalytic composite by immobilizing titanium dioxide (TiO<sub>2</sub>) onto rubberwood biochar to degrade methylene blue (MB) under neon light irradiation. The composite was evaluated under varying conditions including catalyst type (activated biochar vs. normal coal), catalyst dosage (0.1–0.2 g), UV irradiation, and contact time (15–210 min) at an initial MB concentration of 20 ppm. Dye removal was quantified by UV-Vis spectrophotometry. The biochar–TiO<sub>2</sub> composite under UV irradiation achieved approximately 94–96% MB removal within 15 minutes at a dosage of 0.2 g, outperforming normal coal under both irradiated and non-irradiated conditions. The enhanced performance is attributed to the synergistic coupling of adsorption by the biochar surface and photocatalytic oxidation by TiO<sub>2</sub>. This material offers a cost-effective, sustainable solution for textile wastewater treatment while valorizing rubberwood agricultural waste, supporting circular economy goals in Thailand.

**Keywords:** *rubberwood biochar, titanium dioxide, methylene blue, photocatalysis, wastewater treatment*

## 1 Introduction

The rapid expansion of textile and dyeing industries has resulted in widespread contamination of water bodies with synthetic dyes. Methylene blue (MB), a cationic thiazine dye, is among the most prevalent textile pollutants due to its structural stability, resistance to biodegradation, and toxicity to aquatic ecosystems [1, 2]. Conventional treatment methods such as coagulation, biological treatment, and membrane filtration are often insufficient to achieve complete mineralisation of such recalcitrant compounds [2].

Advanced oxidation processes, particularly heterogeneous photocatalysis using titanium dioxide (TiO<sub>2</sub>), have attracted considerable research interest. TiO<sub>2</sub> generates reactive oxygen species—hydroxyl radicals (•OH) and superoxide ions (O<sub>2</sub>•<sup>-</sup>)—upon light activation, capable of degrading a broad spectrum of organic pollutants [3]. However, nano-TiO<sub>2</sub> in suspension suffers from post-treatment separation challenges and particle agglomeration, reducing catalytic efficiency [1].

Immobilising TiO<sub>2</sub> onto solid supports such as activated carbon has been shown to circumvent these limitations. Thanurat et al. [1] demonstrated that granular activated carbon (GAC) impregnated with TiO<sub>2</sub> via the sol-gel method achieved up to 96.7% MB removal through combined adsorption and photocatalytic oxidation, significantly surpassing plain GAC (67.5%) which operated by adsorption

alone. The superior performance was attributed to the anatase crystal structure of TiO<sub>2</sub>, confirmed by X-ray diffraction (XRD), and the increased porosity of the composite material observed by scanning electron microscopy (SEM).

Similarly, Phohtitontimongkol et al. [2] prepared TiO<sub>2</sub>-coated activated carbon from tamarind wood using phosphoric acid chemical activation and reported 97.76% MB removal efficiency after 9 hours at a catalyst dosage of 4.0 g per 100 mL. Their study identified 500 °C as the optimal calcination temperature for anatase crystal formation and ten coating cycles as the minimum for complete surface coverage. Martins et al. [3] further showed that sol-gel-derived TiO<sub>2</sub>/activated carbon composites maintain high photocatalytic activity for the degradation of persistent antibiotics, underscoring the versatility of this approach. Sin et al. [4] confirmed through response surface methodology that TiO<sub>2</sub>/GAC systems can be optimised systematically for dosage, pH, and irradiation time.

Despite these advances, most studies rely on commercially produced activated carbon from coconut shell or wood char, which requires energy-intensive chemical activation. Rubberwood (*Hevea brasiliensis*), a widely available agricultural residue in Thailand following latex extraction, represents an underutilised lignocellulosic biomass that can be converted into biochar with desirable surface properties. This study therefore investigates rubberwood-derived biochar as a novel, regionally sustainable support for TiO<sub>2</sub>, evaluating its performance in MB degradation under neon light irradiation and comparing its effectiveness with non-activated normal coal under identical conditions.

## 3 Content

### 3.1 The Purpose of the Investigation

This study aimed to evaluate the photocatalytic MB removal performance of a rubberwood biochar–TiO<sub>2</sub> composite under neon light irradiation and to compare it with normal coal (non-activated) under both UV and non-UV conditions across a range of catalyst dosages and contact times.

**Hypothesis:** The rubberwood biochar–TiO<sub>2</sub> composite under UV irradiation will achieve significantly higher MB removal efficiency than normal coal owing to the synergistic combination of surface adsorption by the high-porosity biochar and photocatalytic oxidation by anatase TiO<sub>2</sub>.

### 3.2 Method of the Investigation

#### Materials

Methylene blue (MB, analytical grade) was used as the model dye pollutant at an initial concentration of 20 ppm. Two carbon materials were studied: (1) rubberwood-derived activated biochar (hereafter “activated carbon”), prepared by thermal carbonisation followed by TiO<sub>2</sub> loading via sol-gel impregnation; and (2) commercially available normal coal without activation (hereafter “normal coal”). TiO<sub>2</sub> was loaded onto both carbon supports following the sol-gel method analogous to that reported by Thanurat et al. [1]. All absorbance measurements were performed with a UV-Vis spectrophotometer.

## Variables

Independent variables: carbon type (activated biochar vs. normal coal), UV irradiation (present or absent), catalyst mass (0.10 g and 0.20 g for activated carbon; 0.10, 0.25, 0.50, and 1.00 g for normal coal), and contact time (15.0, 30.0, 45, 60, 120, and 150 min for activated carbon; 30.0, 60.0, 120.0, 180.0, and 210.0 min for normal coal). Dependent variable: percentage MB removal (% removal), derived from absorbance readings. Controlled variables: initial MB concentration (20.0 ppm), solution volume, stirring speed, and spectrophotometric settings.

## Procedure

A calibration curve was first established using standard MB solutions (0.0, 2.5, 5.0, 10.0, 15.0, and 20.0 ppm) to confirm linearity between concentration and absorbance. For each experimental condition, MB solution (20.0 ppm) was placed in sealed glass bottles with a specified mass of catalyst. Triplicate samples were prepared for each condition to enable statistical analysis. For UV experiments, neon light was applied throughout the contact period. At each designated time point, absorbance was measured and percentage removal was calculated as:

$$\% \text{ Removal} = [(A_0 - A) / A_0] \times 100$$

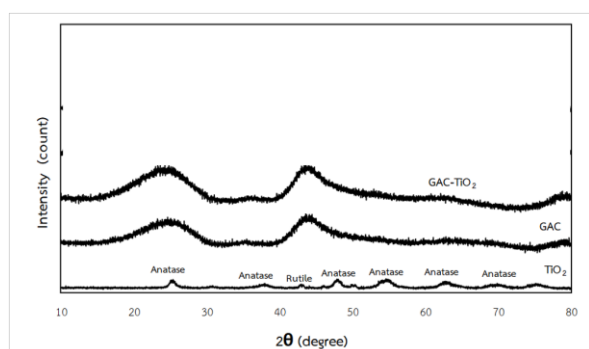
where  $A_0$  is the initial absorbance and  $A$  is the absorbance at time  $t$ .

## Comparison with known methods

Relative to the approach of Thanurat et al. [1], who employed coconut-shell GAC and UV-C lamps, this study uses rubberwood biochar and neon irradiation, representing a more regionally accessible and energy-efficient setup. Unlike Phothisontimongkol et al. [2], who activated tamarind wood carbon with phosphoric acid ( $H_3PO_4$ ), the present work employs rubberwood biochar without acid activation, reducing chemical waste. The study also examines short contact times (15.0–45.0 min), extending beyond the equilibrium-focused designs of prior work to assess practical rapid-treatment scenarios.

## 3.3 Results of the Experiment

Treatment efficiency of GAC-  $TiO_2$  was examined in term of methylene blue removal. From the results, GAC- $TiO_2$  of immobilized practices showed similar treatment efficiency as 95% yields by adsorption and photocatalytic oxidation processes. Crystal structure and element composition of GAC- $TiO_2$  were analyzed by X-ray diffraction (XRD) in Fig 1.



**Fig 1** Crystal structure and element composition of GAC- $TiO_2$  were analyzed by X-ray diffraction (XRD)

A standard calibration curve for MB confirmed a strong positive linear relationship between concentration and absorbance across the range 0.0–20.0 ppm (Table 1), validating spectrophotometric quantification.

**Table 1: MB calibration curve data**

Concentration (ppm)	Absorbance
0.0	0.000
2.5	0.314
5.0	0.673
10.0	1.181
15.0	1.578
20.0	1.781

The calibration curve of standards is  $R^2 = 0.998$ . Normal coal without UV showed moderate and variable MB removal (Table 2). At 1.0 g mass and 30.0 min, removal was 74.33%, improving to 83.08% at 180.0 min. Decreasing the coal mass to 0.10 g increased the maximum removal to 90.29% at 180.0 min, suggesting that surface saturation at high dosages limits performance when  $TiO_2$  loading is absent.

**Table 2: % MB removal – Normal coal, No UV (20.0 ppm, selected conditions)**

Mass (g)	Time (min)	Avg. Absorbance	% Removal±S.D.
1.00	30.0	0.452	74.33±0.08
1.00	180.0	0.298	83.08±0.02
0.50	60.0	0.288	83.65±0.04
0.25	60.0	0.230	86.94±0.05
0.10	180.0	0.171	90.29±0.02

Activated biochar without UV performed substantially better, achieving 93.36%–93.53% at 0.2 g across 30–60 min (Table 3). This indicates strong intrinsic adsorption capacity, consistent with the expanded pore structure of the activated biochar surface.

**Table 3: % MB removal – Activated biochar, No UV (20 ppm, selected conditions)**

Mass (g)	Time (min)	Avg. Absorbance	% Removal±S.D.
0.10	45.0	0.155	91.20±0.01
0.10	120.0	0.146	91.71±0.01
0.20	30.0	0.117	93.36±0.02
0.20	45.0	0.119	93.24±0.02
0.20	60.0	0.114	93.53±0.00

Under UV irradiation, activated biochar- $TiO_2$  delivered the highest overall performance (Table 4). At 0.20 g, removal reached 96.06% within 15 min and remained above 91.83% throughout the 150-min experiment. At 0.10 g, removal ranged from 88.73% to 93.97%. Normal coal under UV irradiation produced comparatively lower removal rates of

83–88% (0.1 g) and 72–83% (0.2 g), confirming that UV activation alone, without the structural benefits of the activated biochar support, provides limited enhancement.

**Table 4: % MB removal – Activated biochar, UV irradiation (20 ppm, all conditions)**

Mass (g)	Time (min)	Avg. Absorbance	% Removal±S.D.
0.10	15.0	0.107	93.97±0.04
0.10	30.0	0.142	92.00±0.04
0.10	45.0	0.144	91.89±0.04
0.10	60.0	0.119	93.30±0.01
0.10	120.0	0.172	90.31±0.03
0.10	150.0	0.200	88.73±0.06
0.20	15.0	0.079	96.06±0.02
0.20	30.0	0.079	95.55±0.00
0.20	45.0	0.103	94.20±0.01
0.20	60.0	0.145	91.83±0.02
0.20	120.0	0.123	93.07±0.03
0.20	150.0	0.114	93.58±0.01

Across all conditions, the ranking of MB removal was: Activated Carbon + UV > Activated Carbon (No UV) > Normal Coal (No UV) > Normal Coal + UV. The divergence between activated biochar and normal coal is attributable to the significantly higher surface area and porosity of the biochar, enabling greater TiO<sub>2</sub> dispersion and dye adsorption, consistent with observations reported by Thanurat et al. [1] and Phothitontimongkol et al. [2].

## 4 Conclusion

This study demonstrated that rubberwood biochar–TiO<sub>2</sub> composite is an effective photocatalytic material for MB removal from synthetic wastewater. Under optimal conditions (0.20 g catalyst, UV irradiation, 15.0 min contact time), the composite achieved approximately 96.0% removal, supporting the stated hypothesis that synergistic adsorption–photocatalysis significantly outperforms either process individually. Performance was consistent across the 15.0–150.0 min observation window, indicating rapid dye uptake.

These results are in agreement with prior work on GAC–TiO<sub>2</sub> composites [1, 3, 4] and TiO<sub>2</sub>-coated wood-derived activated carbons [2], while introducing rubberwood biochar as a promising, locally abundant alternative precursor that does not require acid activation.

Future investigations should address: (1) systematic optimization of calcination temperature and TiO<sub>2</sub> loading cycles specific to rubberwood biochar; (2) characterization of the composite by SEM and EDS to confirm anatase crystal structure and titanium distribution; (3) performance under visible-light irradiation to enable solar-driven treatment; (4) reusability and catalyst stability over multiple cycles; and (5) evaluation using real textile effluent containing mixed dye matrices and competing ions.

## Acknowledgements

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# Smart Water Monitoring: Automated Detection of Microplastics in Water Using Polarization Imaging and Machine Learning

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## Abstract

Microplastic contamination in freshwater environments is an escalating concern, as microplastics accumulate in living organisms and enter the food chain, posing risks to both ecosystems and human health. While established techniques such as Fourier Transform Infrared Spectroscopy (FTIR) and Raman Spectroscopy offer high accuracy, they are limited by cost, complexity, and the requirement for expert supervision, making them unsuitable for field or community-level use. To address this, the project aims to develop an automated system for microplastic detection and classification in water using polarization-based optical imaging combined with deep learning. Samples are imaged under polarized light at angles of 0°, 45°, 90°, and 135° to highlight differences in the optical properties of microplastics, before being fed into a YOLOv11 model for detection and classification of polyethylene terephthalate (PET) and polypropylene (PP) microplastics. Experimental results showed that the system achieved an overall classification accuracy of 77.2% in distinguishing PET and PP microplastics. However, the model still shows limitations in separating background objects from microplastics, which affects overall precision. These findings suggest that polarization-based imaging combined with YOLOv11 has potential as a low-cost, user-friendly automated system for water quality monitoring at the field or community level.

## Keywords

Microplastics, Polarization Imaging, Deep Learning, Freshwater Sources

## 1 Introduction

Microplastics are plastic particles smaller than 5 mm, including nurdles, microbeads, plastic fragments, and synthetic fibers. They originate from direct manufacturing at small sizes (primary microplastics) and from the breakdown of larger plastic items (secondary microplastics) [6,12]. Once accumulated in the environment, they can enter the food chain and have been detected in human blood, placenta, and lungs, posing risks such as gastrointestinal disorders, chronic inflammation, hormonal disruption, and potential effects on fetal development [9]. The United Nations Environment Programme further notes that microplastics can act as carriers for toxic substances such as heavy metals and endocrine disruptors, increasing the risk of chronic diseases such as heart diseases and cancer [13].

Microplastic contamination in freshwater sources is particularly severe. The World Health Organization (WHO) recognizes that while the health impacts remain incompletely

understood, reducing contamination and developing monitoring approaches is urgently needed [14].

Current detection tools remain limited in cost and accessibility. Standard techniques such as Fourier Transform infrared spectroscopy (FTIR) and Raman spectroscopy can identify microplastics accurately but require expensive specialized equipment and expert personnel [3,1]. This highlights the need for easy-to-use, low-cost instruments that can rapidly assess microplastic concentrations in water.

Light polarization is a phenomenon in which the oscillation direction of light waves is confined to a single plane rather than spreading in all directions [2,7,11]. Microplastics possess birefringent properties that rotate the plane of polarized light [8], enabling their detection. When two polarizing films are placed perpendicular to each other, light cannot pass through; however, when a microplastic particle is placed between them, it rotates the polarization direction and allows some light to pass through, revealing the particle's presence.

Plastics such as PET (Polyethylene Terephthalate) and PP (Polypropylene) have molecular structures that affect the direction of polarized light. Their small size, irregular surface, and non-uniform shape cause significant light scattering, affecting polarization state [8,10]. Research published in Marine Pollution Bulletin confirmed that microplastics have distinct optical characteristics compared to inorganic particles (e.g., sand), and using Mueller matrix polarimetry, microplastic particles create unique "optical fingerprints" that can be distinguished by changes in polarized light depending on refractive index and particle shape [4, 5].

Building on this principle, this project develops a low-cost prototype microplastic detection device [15] using adjustable polarizing sheets, a light source, and a camera within an opaque enclosure. The system operates together with an application for processing and displaying microplastic measurement data, making initial screening accessible at community and school levels.

## 1.1 Objectives

This study aims to:

1. To investigate the factors affecting polarized light transmission and the birefringent characteristics of microplastics in rotating the polarization plane.
2. To develop and evaluate a deep learning model for classifying microplastic types from polarization-based optical images.

3. To build a prototype device for semi-automated, low-cost detection and classification of microplastics in water.
4. To develop an application for processing and displaying microplastic measurement data, including particle count and risk-level classification.

## 2 Methodology

### 2.1 Factors Affecting Polarized Light Transmission and Microplastic Birefringence

An experimental setup was designed using a mobile phone screen as the light source, with two polarizing films placed at an angle to each other. The experiments were divided into two configurations:

**Experiment 1:** To investigate the relationship between the rotation angle of the plastic sheet and the transmitted light. The polarization axis of the polarizing films was fixed, and the plastic sheet placed between the two films was rotated. Differences in the resulting images were observed, as shown in Fig.1.

**Experiment 2:** To investigate the relationship between the rotation angle of the polarizing film axis and the transmitted light. The polarizing film attached to the mobile phone screen and the plastic sheet were kept fixed, while the upper polarizing film was rotated. Differences in the resulting images were observed, as shown in Fig.2.



Figure 1: Photographs of observations from Experiment 1 at the same observed height, showing differences in transmitted polarized light at varying plastic sheet rotation angles.

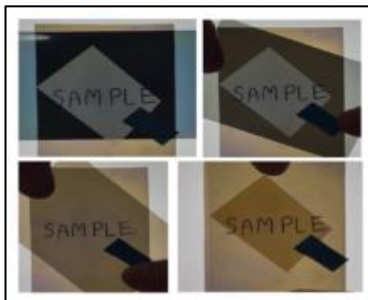


Figure 2: Photographs of observations from Experiment 2 at the same observed height, showing differences in transmitted polarized light at varying polarizing film rotation angles.

## 2.2 Data Collection Device Design for Deep Learning Model Training

### 1. Device Design for Data Collection

The data collection device consisted of fixed polarizing film, a tablet screen as the light source, rotatable polarizing film (at  $0^\circ$ ,  $45^\circ$ ,  $90^\circ$ , and  $135^\circ$ ), opaque cylindrical tube (to eliminate ambient light interference), black opaque fabric (used to cover and seal the enclosure from ambient light) and a smartphone camera for image capture. The experimental setup is illustrated in Fig.3.

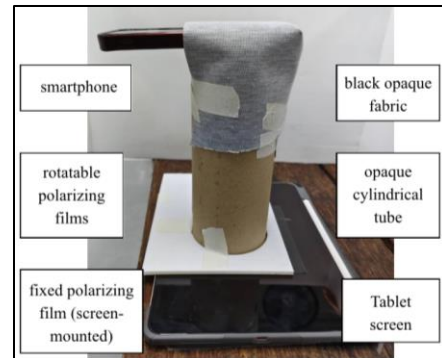


Figure 3: Device setup for training image data collection

### 2. Data Collection for Deep Learning Model Training

The data collection process was carried out in seven sequential steps as follows:

1. Prepare PET and PP plastic sheets by cutting both types into different shapes and sizes, as shown in Fig. 4 and 5. The plastic sheets were placed at varying angles, as the orientation angle is one of the factors affecting the rotation of the polarization plane.
2. Place the plastic sheets between the two polarizing films one at a time, rotating each sheet to different angles to observe changes in the captured images. Replace each sheet with one of a different size and shape one at a time.
3. Observe and capture images of the plastics between the two polarizing films at angles of  $0^\circ$ ,  $45^\circ$ ,  $90^\circ$ , and  $135^\circ$  relative to each other, for both PET and PP types, collecting 350 images per angle per type.
4. Analyze and screen the quality of captured images, selecting only clear images free from external interference such as light sources other than the experimental setup.
5. Store the screened images as a dataset and divide them into a training set and a test set.
6. Train the Deep Learning model using the training set, selecting ResNet-50 as the model and training on Google Colab.
7. Evaluate the classification performance of the model.

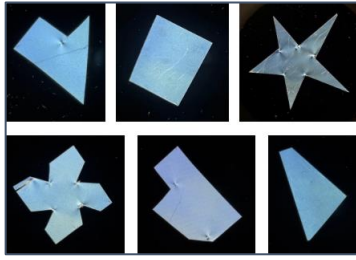


Figure 4: Photographs of PET plastic sheets of varying shapes and sizes captured for data collection

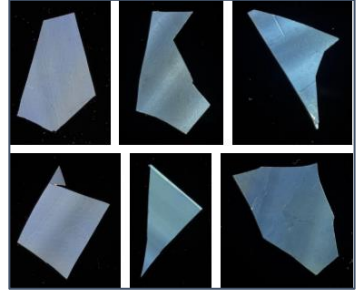


Figure 5: Photographs of PP plastic sheets of varying shapes and sizes captured for data collection

## 2.3 Experimental Setup Design for Microplastic Detection Training Data Collection

The experimental design setup process was carried out in five sequential steps as follows:

1. PET and PP plastic materials were mechanically reduced to microplastic-scale particles through grinding with a plastic grinder.
2. Design a container for holding water sample with microplastic particles using clear acrylic, which have no birefringent properties that would rotate the polarization plane. The container was specified to be 2 mm thick with a size of 6×6 cm<sup>2</sup>, with a laser-engraved groove of 1 mm depth for holding the water sample in a 4×4 cm<sup>2</sup> area. A clear acrylic cover sheet of 2 mm thickness and 5×6 cm<sup>2</sup> was placed on top to press the water sample flat against the base acrylic sheet.
3. Attach a polarizing film to the bottom of the base acrylic sheet with the polarization axis oriented horizontally as the polarizer. A second polarizing film was then attached to the top of the acrylic cover as the analyzer, creating four angular configurations: 0°, 45°, 90°, and 135°.
4. Set up the experiment by placing the prepared microplastics water sample into the container and covering it with the acrylic cover fitted with the analyzer film. The sample was then examined using a Primo Star optical microscope with the light level set to 2, using the lowest magnification — a 4x objective lens and 10x eyepiece lens. Focus was adjusted using the coarse focus knob to achieve maximum clarity. Microplastic particles were then located by scanning the sample without changing

any other parameters, and images were captured using a smartphone camera, as shown in Fig. 6.

5. Photographs of PET and PP microplastic samples were captured under controlled lighting conditions from multiple viewpoints and orientations to increase dataset diversity. The resulting images were stored and organized into a dataset for training and testing the Deep Learning model.



Figure 6: Experimental setup for Deep Learning model training data collection

## 2.4 Data Preparation and Deep Learning Model Development

### 2.4.1 Image Dataset Preparation

1. A total of 638 microplastic images obtained from screened microscope imaging in Section 2.3 were converted to .jpg format and stored in folders to serve as the image dataset for model processing and training.
2. An object detection project was created on the Roboflow platform with the task type set to Object Detection and target classes defined as PET and PP. The microplastic image dataset was then uploaded to the platform, and class labels were assigned to each image, as shown in Fig. 7, before incorporating the data into the model training dataset.
3. The dataset was divided into a training set (87%), test set (4%), and validation set (8%). Additional images were generated using Data Augmentation techniques to increase dataset diversity for model training, with the following augmentation parameters:
  - Flip: Horizontal
  - Rotate 90°: Clockwise, Counter-Clockwise
  - Rotation: ±15°
  - Hue: ±20°
  - Saturation: ±25%
  - Brightness: ±20%
  - Exposure: ±10%
  - Blur: Up to 2px
  - Noise: Up to 1.05% of pixels

- The image dataset was downloaded for use in model training and performance evaluation on a computer.

#### 2.4.2 Development Environment Setup for Model Training and Testing

- A project folder was created for model development and execution using Python in Visual Studio Code. The required libraries were then installed, including PyTorch and Ultralytics.
- Code was developed for model training and object detection inference using the YOLOv11 model, utilizing the previously prepared dataset for training.
- The trained model was tested by inputting images into the program. The model then processed each image and produced output images with bounding boxes and plastic type labels indicating the detected microplastics.

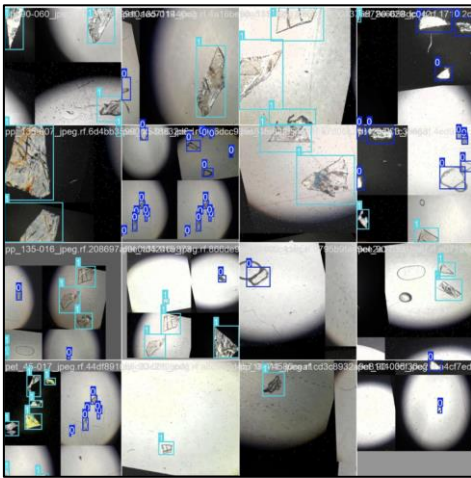


Figure 7: Example of class label assignment to PET and PP microplastics images

### 2.5 Prototype Device Design for Capturing Microplastic Images from Water Samples

**Water Sample Container:** A container for holding microplastic particles in water was designed using clear acrylic sheets. The water sample chamber measures  $2.6 \times 2.6$  cm<sup>2</sup> with a depth of 0.2 cm, capable of holding approximately 1.4 mL of water sample, as shown in Fig. 8.

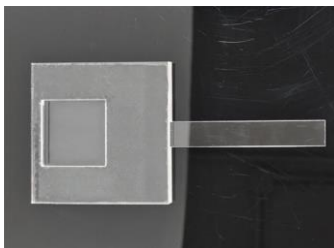


Figure 8: Water sample container

**Prototype Device:** The prototype device was installed inside an opaque box to prevent interference from external light, as shown in Fig. 9. The main components are as follows:

- Smartphone Camera — used to capture images of the sample for light intensity and particle characteristic analysis.
- AA Battery Holder — holds 5 AA batteries.
- AA 1.5V Batteries (×5) — used as the power supply for the system (total voltage: 7.5V).
- Arduino Uno Microcontroller Board — controls and commands the servo motor to rotate to specified angles according to the program and manages the operational sequence during image capture.
- Sample Container — holds the microplastic-mixed water sample, made from clear acrylic, as shown in Fig. 8.
- Frosted Acrylic Diffuser Sheet — distributes light from the LED evenly and reduces hot spots before entering the polarizing assembly.
- Polarizer Sheet — adjusts the direction of light to produce linearly polarized light.
- Analyzer Sheet — connected to the motor to rotate and adjust to specified angles: 0°, 45°, 90°, and 135°.
- MG90S Servo Motor — controls the rotation of the Analyzer sheet to stop precisely at specified angles: 0°, 45°, 90°, and 135°, via signals from the microcontroller.
- Push Button Switch — controls the initiation of the polarizing sheet rotation system.
- STP Switch — controls the on/off operation of the Arduino board by connecting/disconnecting the battery to the system.
- 12W White LED Panel Light (Light Source) — powered on and warmed up for approximately 1–2 minutes to stabilize light intensity.

#### Initial Device Configuration

- Measurement angles: 0°, 45°, 90°, 135°
- Settling time: 3 seconds per angle
- Number of measurements: 4 images captured per sample
- The Arduino board was configured to control servo motor rotation and tested for computer connectivity.

#### Prototype Device Operating Procedure

- The prototype device is an opaque box to prevent interference from external light.
- The LED Panel Light was powered on and warmed up for approximately 1–2 minutes to stabilize light intensity.
- Light from the source passes through the diffuser sheet and polarizer sheet to produce linearly polarized light.
- The polarized light travels through the sample container holding the microplastic-mixed water sample.

5. Light passing through the sample enters the analyzer sheet, which is mounted on a holder and connected to the servo motor.
6. The analyzer sheet was initialized at 90° and the servo motor reference position was set via the Arduino board.
7. The system was powered on using the STP switch.
8. The data recording process was initiated by pressing the push button, commanding the servo motor to rotate the analyzer sheet to the specified angles: 0°, 45°, 90°, and 135° sequentially.
9. At each angle, the position was held for 3 seconds to capture images of the water sample using the smartphone camera.
10. After completing rotation and image capture at all four angles, the system commands the servo motor to return the analyzer sheet to the initial position at 90° to reset to the starting point.
11. Images captured at polarization angles of 0°, 45°, 90°, and 135° are fed into the application for data preparation before being processed by the YOLOv11 model to analyze and classify microplastic types from the polarized image data of the water sample

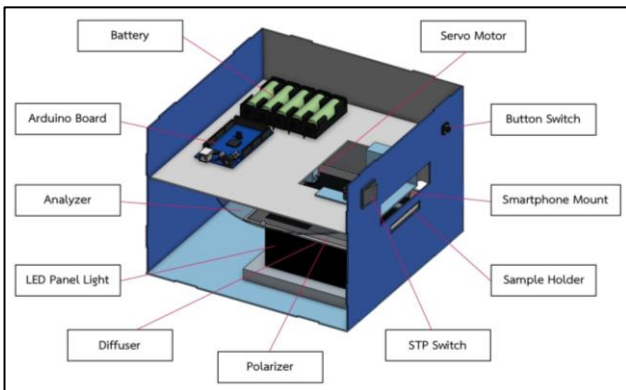


Figure 9: Prototype device for microplastic detection in water

## 2.6 Application Design and Development for Microplastic Data Processing

The developed application serves as a tool for managing microplastic measurement data and supports the operation of the PET and PP microplastic detection and classification system, enabling more systematic and user-friendly use for general users.

### 2.6.1 Application Workflow and AI Model Interfacing

- The application was designed to allow users to input microplastic sample images captured from the prototype device for analysis.
- The images are submitted to the AI processing system developed in Section 2.4, with classification results displayed clearly to the user.

The workflow begins with the user uploading images captured from the prototype device into the application. The data is then sent to the processing system, where the model

analyzes and classifies the microplastic types before returning the results to be displayed through the application.

### 2.6.2 Application Development Using Flutter

The application was developed using the Flutter framework to support cross-platform compatibility and to connect with a data storage system for recording sample data and measurement results. The application was designed to align with the operational workflow of the microplastic detection and classification system, enabling efficient integration with the Deep Learning model processing pipeline.

### 2.6.3 UI/UX Implementation

The user interface was developed for practical use, with the following details:

- **Home Page:** Displays the application logo.
- **Data Page:**
  - Basic information fields for recording sample details including location name, date, and water sample type.
  - Image upload function for sample images captured from the prototype device.
  - Measurement results displaying microplastic type classification and quantity detection.
- **History Page:** Displays previous measurement records.

The development of this application enables the microplastic detection and classification system to be practically deployed, reducing data management complexity, and improving the convenience of displaying measurement results, as shown in Fig. 10.

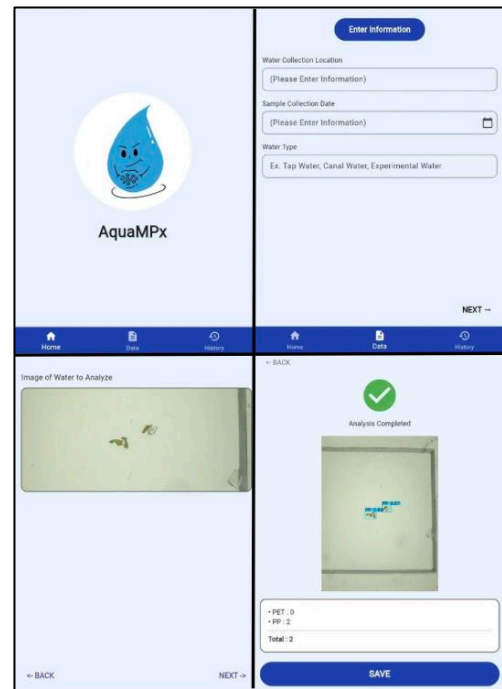


Figure 10: Example of the application user interface (UI) and user interaction

### 3 Results and Discussion

#### 3.1 Polarized Light Transmission and Microplastic Polarization Results

A study of the factors affecting changes in the polarization plane of light passing through plastic sheets was conducted to serve as the foundation for designing the imaging system and data preparation process. From preliminary studies and experiments, the key factors affecting polarized light behavior were found to be:

- The angle between the polarization axes of the polarizer and analyzer sheets.
- The orientation angle of the plastic sheet relative to the polarization axis of light.

This investigation revealed that measuring a single light intensity value may not sufficiently reflect the unique characteristics of each plastic particle type, particularly when multiple particles are present within the same image.

#### 3.2 Large Plastic Classification Results Using Deep Learning

This experiment investigated the effect of the angle between polarizing sheets on light intensity changes in large plastic samples. PET and PP plastics were photographed under four different polarizing angles: 0°, 45°, 90°, and 135°.

The results revealed that varying the angle between the polarizing sheets significantly affected image brightness and contrast. Both plastic types allowed some light to pass through even when the polarizing sheets were perpendicular to each other, demonstrating their ability to rotate the polarized light direction. When comparing plastic types, PET plastic exhibited more pronounced variation in light intensity, while PP plastic showed relatively uniform brightness, as shown in Fig. 11 and Fig. 12.

Plastic image data captured at various polarizing angles was used to develop a machine learning model for classifying large plastic types. The dataset was divided into two sets: a training set and a testing set.

The results showed that the developed model was able to correctly classify plastic types in the test dataset. Model performance evaluation yielded an accuracy of **0.9706**, demonstrating high classification accuracy from photographic images, as shown in Fig. 13.

These results indicate that plastic images captured at various polarizing angles can be effectively used for plastic type classification through Deep Learning model development, and can serve as a foundation for further development and expansion toward microplastic classification in subsequent steps.

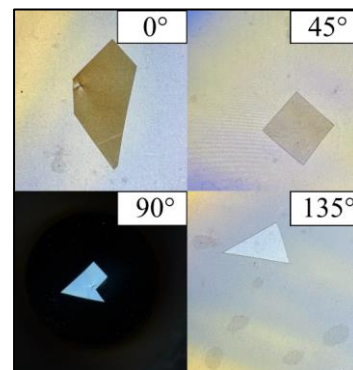


Figure 11: Example photographs of PET plastic at polarization angles of 0°, 45°, 90°, and 135°

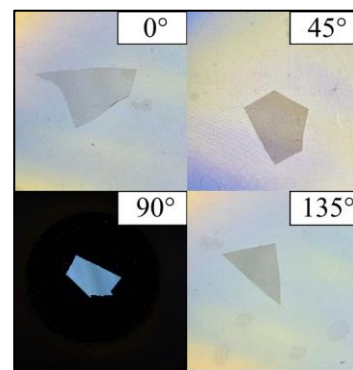


Figure 12: Example photographs of PP plastic at polarization angles of 0°, 45°, 90°, and 135°

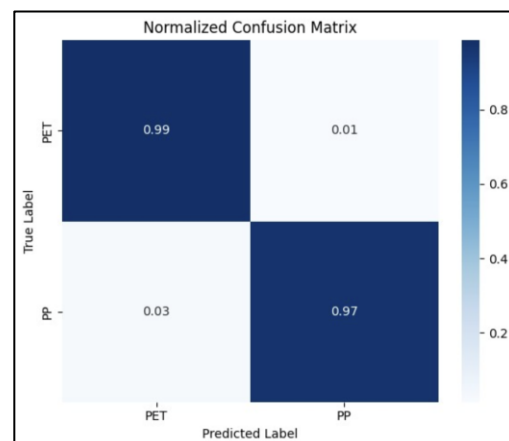


Figure 13: Confusion matrix showing the performance of the developed ResNet-50 model for PET and PP large plastic detection

#### 3.3 Results of Microplastic Image Data Collection

Microplastic samples were photographed using a microscope at four polarizing angles: 0°, 45°, 90°, and 135°. The results showed that light transmitted through PET and PP plastics exhibited distinct characteristics at different angles, as shown in Fig. 14 and Fig. 15.

When comparing the two microplastic types, PET microplastics showed more pronounced differences in background contrast and light intensity variation compared to PP microplastics. PP microplastics exhibited relatively uniform brightness with minimal variation. These

characteristics reflect the differences in optical properties and molecular structure between the two microplastic types.

From the experimental results, it can be concluded that optical testing using the polarization method can effectively detect microplastics, and the differences in light intensity obtained can serve as foundational data for microplastic type classification in subsequent steps.

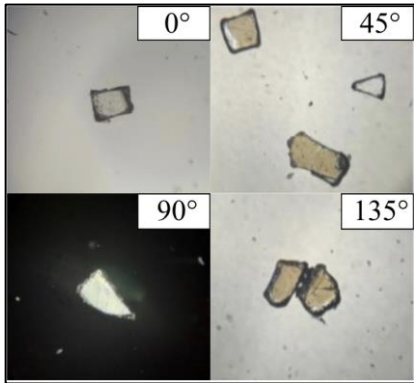


Figure 14: Example photographs of PET microplastic at polarization angles of 0°, 45°, 90°, and 135°

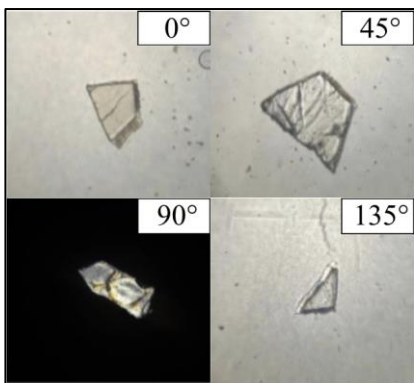


Figure 15: Example photographs of PP microplastic at polarization angles of 0°, 45°, 90°, and 135°

### 3.4 Model Training and Evaluation Results

#### 3.4.1 Microplastic Detection Results

The results demonstrated that the YOLOv11 model could consistently detect PET and PP microplastic particles across all four polarization angles — 0°, 45°, 90°, and 135° — even though the same sample can appear visually different at each angle due to changes in light intensity and contrast, as shown in Fig. 16. This suggests the model has learned to recognize the underlying characteristics of each plastic type regardless of how the polarization angle affects the appearance of the image.

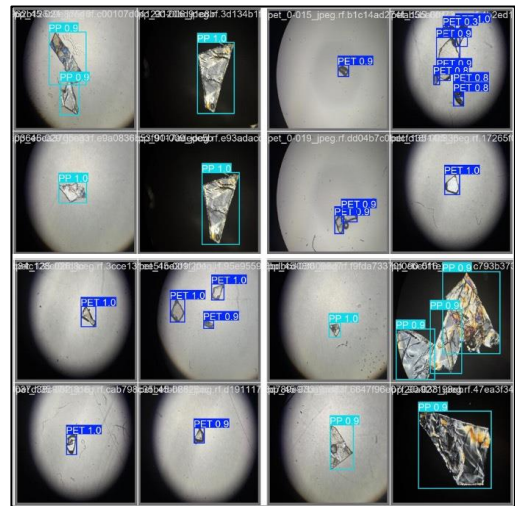


Fig. 16: Example of PET and PP microplastic detection and classification by the YOLOv11 model

#### 3.4.2 Model Training Results

The YOLOv11 model was trained for microplastic detection of PET and PP types over 100 epochs to enable the model to effectively learn microplastic characteristics from microscope images. The results showed that the training loss — including box loss (object localization), classification loss (type classification), and distribution focal loss (DFL loss) — consistently decreased as the number of training epochs increased, as shown in Fig. 17. This indicates that the model progressively improved its ability to identify the distinguishing characteristics of microplastics.

Regarding the validation set, the loss values showed slight fluctuations during certain training periods, as shown in Fig. 18. This is a normal characteristic when learning from diverse image data. However, the overall loss did not increase abnormally, indicating that the model did not suffer from overfitting the training data.

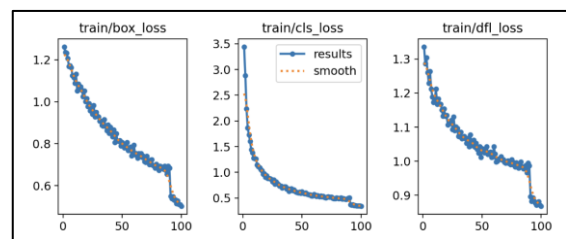


Figure 17: Graphs illustrating the trends of box loss, classification loss, and DFL loss of the training dataset across 100 training epochs

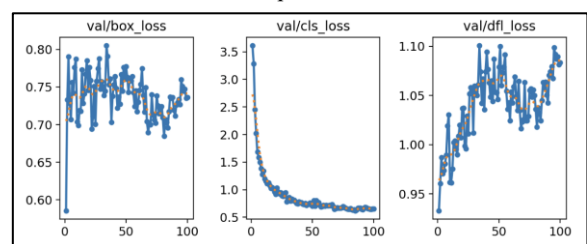


Figure 18: Graphs illustrating the trends of box loss, classification loss, and DFL loss of the validation dataset across 100 training epochs

#### 3.4.3 Model Performance Evaluation

Model performance evaluation was conducted to analyze the

capability of the YOLOv11 model in detecting and classifying PET and PP microplastics from microscope images, using statistical metrics along with results obtained from testing the model on a previously unseen test dataset.

**Model Accuracy:**

From the graphs showing Precision, Recall, and mAP values across training epochs, as shown in Fig. 19, these metrics increased rapidly during the initial training period and showed a stable trend as the number of epochs increased. At epoch 100, the model achieved approximate values of Precision  $\sim 0.90$ , Recall  $\sim 0.85$ ,  $mAP@50 \sim 0.90$ , and  $mAP@50-95 \sim 0.72$ , reflecting the model's ability to learn microplastic characteristics with stability and effectiveness.

Furthermore, the  $mAP@50$  and  $mAP@50-95$  values were at satisfactory levels, demonstrating that the model was capable of accurately localizing and classifying microplastic types under both general conditions and conditions requiring higher levels of object localization precision.

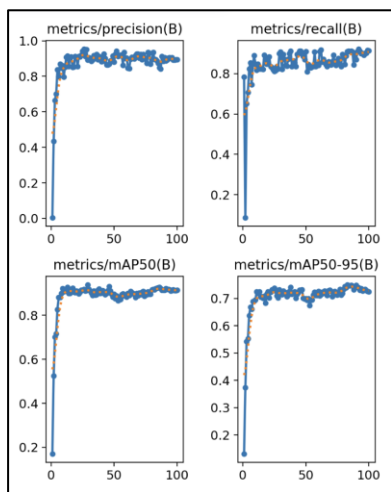


Figure 19: Graphs illustrating the trends of Precision, Recall,  $mAP@50$ , and  $mAP@50-95$  across 100 training epochs

**Precision-Recall Curve:**

The Precision-Recall Curve, as shown in Fig. 20, demonstrates that the model achieved high performance in microplastic classification. PET microplastics showed an Average Precision of **0.966**, while PP microplastics achieved an AP of **0.879**, reflecting the greater classification difficulty of PP. Considering all classes overall, the model achieved an  $mAP@0.5$  of **0.923**, demonstrating the system's ability to detect microplastics with high accuracy and reliability.

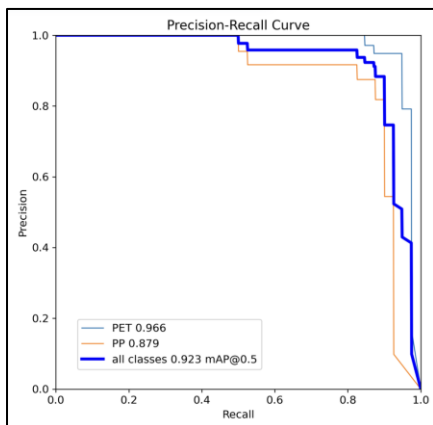


Fig. 20: Precision-Recall Curve for PET and PP microplastic classification

**Confusion Matrix:**

The Confusion Matrix, as shown in Fig. 21, presents the classification results of microplastics into PET, PP, and background classes. The model demonstrated high efficiency in classifying PET microplastics, achieving a Precision of **0.90**, Recall of **0.95**, and F-measure of **0.92**, indicating accurate and comprehensive PET detection. For PP classification, the model achieved a Precision of **0.79**, Recall of **0.85**, and F-measure of **0.82**, which was slightly lower due to some misclassification into the background class. Nevertheless, the overall model Accuracy of **0.78** and Specificity were at a satisfactory level, reflecting that the developed Deep Learning model can appropriately distinguish microplastics from the background and has the potential for application in an automated microplastic detection system in water.

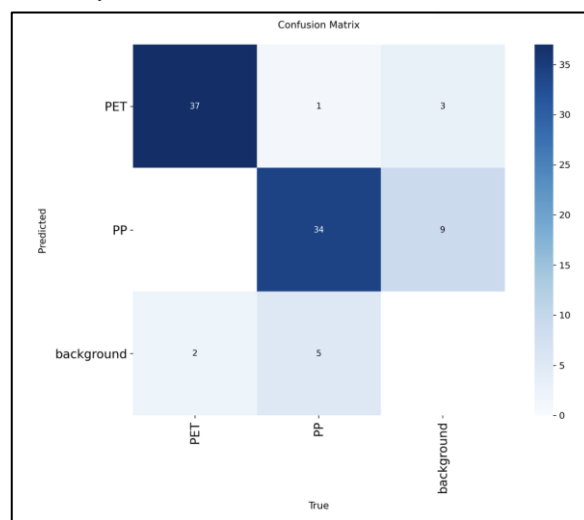


Figure 21: Confusion Matrix showing PET, PP, and background classification results

Overall performance evaluation results demonstrate that the YOLOv11 model is capable of effectively detecting and classifying PET and PP microplastic types, and can be applied to the analysis of microplastic images captured under polarized light.

**3.5 Prototype Device Testing Results**

Testing of the developed prototype device revealed that it operated correctly according to the designed principles, utilizing a structure and components that are readily accessible, resulting in reduced development costs.

These experimental results demonstrate that the developed prototype device can effectively address microplastic detection requirements under the concept of simple design, practical usability, and low cost.

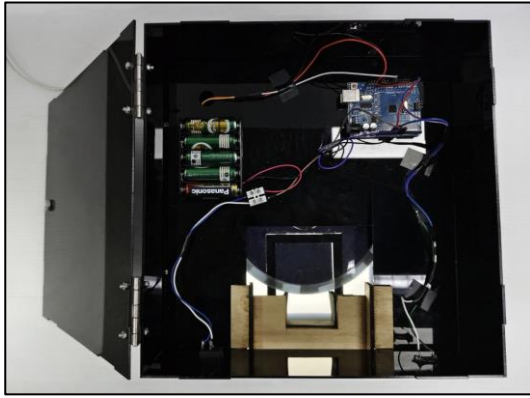


Figure 22: Prototype device for microplastic detection in water (actual device)

### 3.6 Application Development Results for Data Display

Testing of the application revealed that it fully supports integration with the microplastic detection and classification system. The application can receive input data from users, transmitting it to the processing system, and displaying detection results along with measured quantities and classification of PET and PP microplastic types, as shown in Fig. 23.

These results demonstrate that the developed application is practically functional and plays a significant role in enabling the microplastic detection and classification system to operate efficiently, particularly in terms of improving user convenience and accessibility.



Figure 23: Microplastic detection and classification results displayed by the processing system

## 4 Conclusion

This project set out to build a simple, affordable system for detecting microplastics in water, and the results suggest it is achievable. By combining polarization-based optical imaging with Deep Learning and the AquaMPx mobile application, the system was able to detect and classify PET and PP microplastics in a way that is practical for community and school-level use.

The YOLOv11 model performed well overall, reaching an mAP@0.5 of 0.923. PET classification was particularly strong at an Average Precision of 0.966, while PP reached 0.879, a slightly lower score that likely reflects PP's more uniform behavior under polarized light, making it inherently harder to distinguish.

It is worth noting that all testing was carried out using artificially prepared samples in controlled conditions. While this was a necessary starting point, real freshwater environments are far more complex, and future testing in natural water sources will be essential to validate the system's reliability.

The AquaMPx application worked as intended, allowing users to upload images, view classification results, and track measurement history, making the system accessible to non-specialist users without requiring technical expertise.

Going forward, the team plans to test with real water samples, expand detection to more plastic types, and refine PP classification accuracy, with the broader goal of making microplastic monitoring more accessible and widely applicable for real-world field applications.

## 5 Acknowledgements

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# Assessment of olive stone-derived biochar as a sustainable solution for reducing pharmaceutical pollution in wastewater

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## Abstract

*This research paper examines pharmaceutical pollution and its existence in both drinking- and wastewater. The rapid growth of the pharmaceutical industry, together with the sometimes excessive use of medications, has increased the concentration of active compounds and their metabolites in the environment. Many of these can persist for long periods of time, posing risks to both aquatic organisms and human health. In this context, the present paper proposes a hypothetical improvement to current filtration systems, specifically those used in Barcelona, with the aim of increasing their sustainability and efficiency. The proposal involves the development of a biochar-based filter material produced from olive pits, an abundant organic waste in the region, and analyses the potential benefits of this material for water filtration as well as its environmental sustainability. It also outlines the procedure that would be followed to recreate a sample of the biochar and evaluate its effectiveness through chromatography. Additionally, a model of the filtration system designed to support this material and optimise the filtration process is presented.*

## Keywords

Pharmaceutical pollution, wastewater filtration, biochar, sustainability

## Hypothesis

Olive stone-derived biochar could be a more effective and sustainable alternative for wastewater filtration and the elimination of pharmaceutical compounds found in it.

## Introduction

Freshwater is one of the most essential and limited natural resources on the planet. Agriculture, urbanisation, tourism and public water supply place pressure on freshwater sources increasing the demand for clean and accessible water around the world. [1] In parallel, urban areas generate large amounts of wastewater as a result of domestic use, industrial

activities and urban runoff. This wastewater must be treated before being released back into the environment in order to prevent harm to ecosystems and human health. Consequently, the quality of natural water bodies is directly linked to how efficiently wastewater is treated. Contaminants that are not fully removed can accumulate in rivers, lakes and coastal water, making sustainable water management a priority for modern societies. [2]

Among the many pollutants present in wastewater are pharmaceutical compounds. The first reports on their presence in the environment were published in the 1970s, and since then, this topic has been increasingly common in scientific literature. [3] Such substances originate mainly from pharmaceutical manufacturing facilities, the livestock industry, hospitals and healthcare centres, and everyday human consumption of medicines. [4] Once released, they are transported through urban wastewater infrastructure and, in many cases, eventually diffuse into natural water bodies. [5]

The presence of pharmaceutical residues in aquatic ecosystems raises numerous environmental and public health concerns. These compounds are designed to interact with biological systems, potentially altering physiological processes in aquatic species and disrupting ecosystems. [6] To address this issue, wastewater treatment plants rely on conventional physical and chemical processes. These methods are effective for traditional pollutants, however they are often limited in their ability to eliminate pharmaceutical compounds completely. More advanced treatment technologies exist, but they are frequently expensive and suppose high energy consumption. [7] Therefore, a more sustainable effective solution must be sought out.

## 1. Pharmaceutical pollution

Medicines are chemical products used in diagnosis, treatment or cure, as well as in the alteration or prevention of diseases, health conditions or the functionality of the body.

The global pharmaceutical market is a subsegment of the healthcare sector. More than 12,000 pharmaceutical manufacturing plants are registered worldwide, with annual production exceeding 60 billion prescription and over-the-counter drug doses. Europe operated more than 2,000 plants, supplying over 15 billion doses focused on sterile injectables, oncology drugs, and active pharmaceutical ingredients (APIs). [8] Spain, in 2024, had 181 significant pharmaceutical manufacturing plants. Catalonia is the country's leading region, with 79 production plants, accounting for 44% of the total. [9, 10]

As the pharmaceutical industry has expanded globally, risks of environmental impact have also increased. Pharmaceutical contamination is the presence of drug residues and their metabolites in the environment. These emerging contaminants harm the ecosystem by affecting flora and fauna and potentially human health. Their generation is closely linked to the growth of the pharmaceutical industry over the years, which has also increased the associated environmental impact. This contamination occurs mainly because drugs contain APIs in their formulation, which are responsible for fighting infections, curing diseases, modifying metabolism when it is altered, or relieving the pain of those who take them. [11] They are designed to maintain their chemical structure and function over a long period of time, approximately 24 months or more from their sale and use, in order to carry out their therapeutic function in organisms. The long duration of the drug means that, even after it has left the body, the compound remains active and, therefore, may pose a risk of contaminating the environment or altering the metabolism of an organism that might accidentally ingest it. [12]

### 1.1. Active pharmaceutical ingredients

APIs are the components responsible for the therapeutic effect of a drug, designed to diagnose, treat, cure, or prevent diseases in the body. They act by modifying biological processes, such as fighting infections (antibiotics), relieving pain (analgesics), regulating the nervous system, or modulating the immune response. [9]

For example, in aspirin (brand name), the active ingredient is acetylsalicylic acid. Acetylsalicylic acid (ASA), is a drug of the salicylate family, often used as an analgesic. At low doses and with long-term treatment, ASA irreversibly blocks the formation of thromboxane A2 (a lipid) in platelets, producing an inhibitory effect on platelet aggregation. [23]

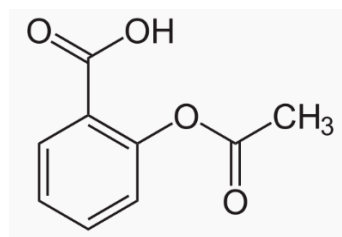


Figure 2: Structure of ASA [23.1]

### 1.2. Common drugs in wastewater

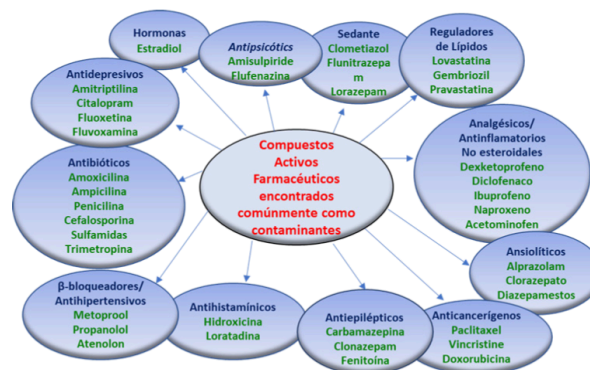


Figure 1: Common APIs as pollutants [12]

The most abundant compounds in wastewater are:

- Analgesics and anti-inflammatories, such as acetylsalicylic acid, ibuprofen, naproxen and diclofenac;
- Hormones, such as synthetic estrogens and others present in contraceptive hormone therapies;
- Antibiotics, which directly contribute to the development of bacterial resistance;
- Antidepressants and neuroleptics, such as fluoxetine and others that alter wildlife behaviour;
- Antiparasitic and pesticide chemicals, present in hygiene products and veterinary use;
- Metabolites and degradation products, which result from the transformation of drugs after being metabolised and are often more persistent;
- Other chemicals, such as triclosan, nonylphenols and parabens, present in many medications and consumer products. [12]

These compounds present dangers to the ecosystem because of their chemical properties: they are difficult to degrade and toxic to aquatic organisms, potentially

causing malformations and affecting their growth and reproduction. [13, 14] The main pathways of contamination are related to the consumption and disposal of drugs and metabolites in urine and faeces. It is also related to the improper or insufficient disposal of unused or expired medications. [15]

### 1.3. Pathways into natural bodies

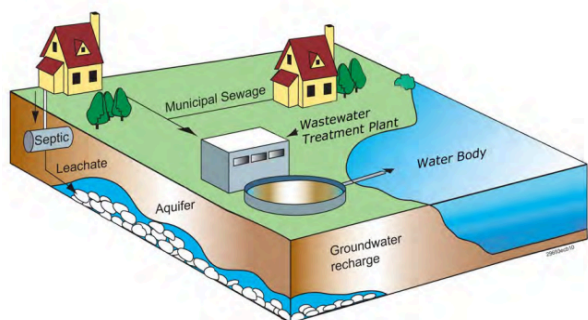


Figure 3: Pathway of pharmaceuticals post-use [x]

The main entry routes of pharmaceuticals into the aquatic environment are wastewater and wastewater treatment plants, but they can also enter via landfills or the disposal of leftover medications. Wastewater includes urban, hospital, industrial, and agricultural or livestock sources. In addition, drug residues can be deposited in soil through direct excretion (animals) or due to the reuse of organic waste. Medicines that are normally ingested can reach the environment when they are carried away by shower or toilet water, and in the case of agriculture and livestock farming (even more so in fish farms), medicines enter directly as plant growth factors or pesticides. [12]

As for the metabolic pathway of drugs within the human body, they are ingested, distributed through the bloodstream, metabolised, absorbed, and excreted. Once the drug has performed its intended function, the APIs, additional excipients, and their metabolites produced during metabolism are released from the human body, in this case through sweat, urine, saliva, and feces. These contaminating components mainly originate from human and animal consumption and from expired medications. [16]

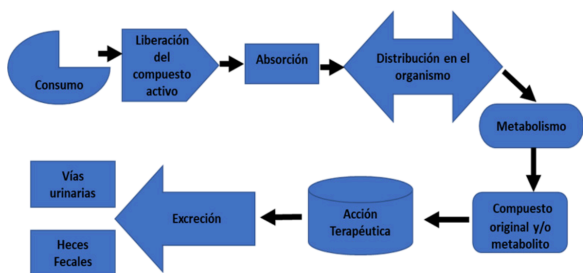


Figure 4: Route of APIs within the human body [12]

### 1.4. API persistence in the environment

Pharmaceutical compounds are considered environmental contaminants due to their incomplete removal in wastewater treatment plants. These compounds exhibit certain properties: pharmacological action (the ability to interact with the body to carry out their therapeutic function), solubility in water or oils, polarity (the most suitable solvent), and stability (maintaining their chemical integrity during and after use). [12, 15]

Many of these substances are persistent due to their properties, meaning they resist natural degradation. Even when compounds are not chemically very stable, their everyday use and their constant introduction into waters as contaminants leads to high concentrations in aquatic systems, pseudo-persistence, maintaining detectable levels in lakes, rivers, or groundwater. It should be emphasised that the degradation process and its longevity in the environment depend heavily on environmental conditions: temperature, salinity, redox state, microbial activity, and solar exposure. Therefore, the transformation of a chemical in the environment is determined by a combination of specific chemical properties and environmental conditions. [13]

These factors can cause bacterial activity to decrease or increase. Moreover, some drugs undergo structural transformations upon ingestion, converting into metabolites (metabolic products) while remaining biologically active and persistent. In other words, these drugs do not always disappear but rather transform into other compounds that remain biologically active and chemically more stable, making their persistence and duration in the environment even longer. One example would be diclofenac, which can partially degrade in water and form different compounds to the original (4'-hydroxydiclofenac or 5'-hydroxydiclofenac). [13]

The drugs most commonly detected in wastewater treatment plants include antibiotics (such as sulfonamides, ciprofloxacin, and tetracycline), as well as anti-inflammatory drugs and analgesics such as ibuprofen, diclofenac, and acetaminophen. Although concentrations vary by location and treatment plant, these compounds are regularly found in wastewater and aquatic ecosystems. [12]

### 1.5 Biological impacts

Emerging contaminants include chemicals such as APIs, surfactants, illicit drugs (substances that are chemically different but mimic the pharmacological

effects of a particular substance), and hydrocarbons. This pollution has generated concerns due to its presence in the environment, such as soil, sediments, surface waters, and groundwater. Studies have confirmed that these components have been found in water streams within a biologically active range. For instance, high water solubility facilitates the rapid transport of these pollutants through aquatic systems, allowing them to reach drinking water sources more easily and disperse contamination over long distances. [17]

To study the environmental risk of drugs, all types of variables must be considered: consumption volumes, their physicochemical properties such as water solubility, metabolism within the organism, behavior in the environment (e.g., degradation and mobility), and their ecotoxicity. Many of the properties that are important for the therapeutic effectiveness of medicines can be problematic from an environmental perspective, such as high stability or high water solubility.

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Substances with so-called PBT properties are considered highly critical for the environment as they:

- Degrade slowly and remain in the environment for a long time (Persistent);
- Accumulate within organisms (Bioaccumulative);
- Represent risks to humans and/or environmental organisms, and can be carcinogenic or alter the hormonal system (Toxic). [18]

Due to these properties, PBT substances should not enter the environment. They represent a risk regardless of their concentration, and their long-term effects on humans and the environment are difficult to predict. This also applies to pharmaceutical substances with PBT properties, although very few active ingredients are currently known to fall into this category. [18]

In addition to PBT criteria, it is essential to monitor parameters such as Chemical Oxygen Demand (COD) and Biological Oxygen Demand (BOD). The presence of pharmaceutical residues can significantly alter these values, indicating a degradation of water quality and lower oxygen availability for aquatic life. [19]

Hormonal drugs, upon reaching an organism that does not require them, can interfere with internal biological system processes, such as reproduction, development, and growth. [19]

Another type of drug with a significant impact on our environment is antibiotics. If their elimination is not handled correctly, it can lead to bacterial resistance. While this phenomenon is already a growing concern, its impact could escalate exponentially, as antibiotic residues in wastewater act as a permanent training ground for the development of resistant "superbugs"; these could lead to disrupting natural biodegradation processes within ecosystems. [15, 19]

## 1.6 Effects on aquatic organisms

The presence of APIs in seas and rivers causes them to be absorbed or consumed by aquatic organisms, which are not the target species for which these substances were designed. These compounds bioaccumulate in non-target organisms, causing various types of physiological damage. Recent studies have shown that even at sub-lethal concentrations, pharmaceuticals like Diclofenac can cause severe tissue damage in the liver and kidneys of salmonids, while beta-blockers such as Propranolol significantly reduce the heart rate and metabolic activity of various crustacean species. [18]

Among the main negative impacts, the decrease in reproduction among affected organisms is particularly concerning. In fish species such as trout, Japanese medaka, and goldfish, it has been demonstrated that these compounds cause teratogenic effects, specifically malformations and morphological or functional alterations during development. Some drugs act as endocrine disruptors, altering the hormonal systems of fish. It has been proven that these compounds cause the feminisation of fish, thereby severely affecting their reproduction by reducing the male population and, consequently, the fertilisation rates of females. [19]

Additionally, APIs inhibit chlorophyll biosynthesis in microalgae, causing severe damage and even death, as these organisms lose the ability to convert inorganic matter into the nutrients necessary for survival. In other words, at slightly higher concentrations, APIs can have lethal effects. [20]

### 1.6.1. Reproduction impacts

Active ingredients gravely alter reproduction by acting as endocrine disruptors. These compounds interfere with hormones, affecting oocyte maturation, reducing sperm quality, altering sexual cycles, and inducing feminisation in fish, amphibians, and mammals, which ultimately decreases birth and survival rates. [21, 22]

Key mechanisms include:

- Endocrine disruption: many chemical active ingredients mimic or block natural hormones (such as estrogen and progesterone), deregulating ovarian cycles and testosterone production.
- Impact on female reproduction: they can alter oogenesis in the ovary, interfere with the preparation of the endometrium for implantation, and affect zygote development.
- Impact on male reproduction: they affect the formation of sperm (spermatogenesis) and reduce testosterone production, impairing overall sperm quality. [21, 22]

### 1.6.2. Impact on photosynthetic organisms

When certain compounds (especially herbicides and some antibiotics) reach plants or algae, they can block chlorophyll biosynthesis, causing what is known as chlorosis (yellowing and loss of photosynthetic capacity). This disruption of the primary producers has a "bottom-up" effect on the entire food web, as the lack of algae reduces the food source for zooplankton and higher trophic levels. [20, 23]

They primarily affect organisms in three different ways:

- Inhibition of the PPO enzyme: many herbicides inhibit the enzyme protoporphyrinogen oxidase (PPO), which is essential for the synthesis of the heme group and chlorophyll. When blocked, precursors accumulate that, in contact with light, generate singlet oxygen (free radicals) that destroy cell membranes.
- Electron transport blockade: active ingredients such as triazines bind to chloroplast proteins. While this does not inhibit chlorophyll production directly at first, it renders it useless and eventually leads to degradation via photooxidation. The plant receives light but

cannot channel the energy, literally "burning" its own chlorophyll.

- Magnesium chelation: the chlorophyll molecule contains a magnesium atom ( $Mg^{2+}$ ) at its center. Some active ingredients with chelating properties can sequester magnesium from the environment or replace it with heavy metals, creating non-functional chlorophyll molecules that cannot absorb light. [20, 23]

Active substance	Use*	Most sensitive species	Toxicity	Effect concentration	Reference
<b>Analgesics</b>					
Acetylsalicylic acid	VMP, HMP	Fish	moderate	NOEC = 283 µg/L	JanusInfo.se / Assessment Report Aggrin
Diclofenac	HMP	Fish, mussels	very high	NOEC = 0.44 µg/L	EU-WFD, Circa (2022)
Ibuprofen	HMP	Fish	very high	NOEC = 6.88 µg/L	EU-WFD, Circa (2022)
Ketoprofen	HMP, VMP	Green algae	high	NOEC = 17.8 µg/L	JanusInfo.se / Assessment Report Orudis
Naproxen	HMP	Water flea	high	NOEC = 33 µg/L	JanusInfo.se / Assessment Report Naproxen Entero
Paracetamol	HMP, VMP	Water flea	moderate	NOEC = 100 µg/L	JanusInfo.se / Assessment Report Paracetol
<b>Antibiotics</b>					
Amoxicillin (Amoxicillin Penicillin Acid)	VMP, HMP	Cyanobacteria	very high	NOEC = 0.8 µg/L	Andreucci et al., 2004
Azithromycin	HMP	Cyanobacteria	very high	NOEC = 0.019 µg/L	EU-WFD, Circa (2022), Oekotoxzentrum 2015
Clarithromycin	HMP	Cyanobacteria	very high	E.C10 = 0.13 µg/L	EU-WFD, Circa (2022), Baumann et al. 2015
Doxycycline	VMP, HMP	Cyanobacteria	high	NOEC = 22.4 µg/L	Assessment Report Allidox
Erythromycin	VMP, HMP	Cyanobacteria	very high	E.C10 = 0.5 µg/L	EU-WFD, Circa (2022)
Oxetracycline	VMP, HMP	Cyanobacteria	high	EC50 = 261 µg/L	Assessment Report OXTRA

Figure 5: Table of effects of APIs on different species of fish [22]

### 1.7. Human health and antibiotic resistance

Anti-inflammatory drugs, antibiotics, and painkillers are among the most widely used pharmaceuticals globally. Sewage treatment plants typically remove only 20–30% of diclofenac due to its low biodegradability and limited adsorption on activated sludge. Continuous release of these drugs into the environment results in chronic exposure for organisms, as they remain active and toxic even at low concentrations. Studies show that such exposure may lead to health issues like Alzheimer's, obesity, thyroid disorders, and cancer. [15]

Therefore, effective treatment of pharmaceuticals at their source is essential before discharge into aquatic environments. Addressing these challenges requires a multifaceted approach involving improved drug manufacturing practices, proper disposal methods, enhanced wastewater treatment, and increased awareness among healthcare professionals and the public. [14]

Another major problem observed is that antibiotics present in low concentrations across various natural water sources are causing many microorganisms, especially pathogenic bacteria, to develop resistance to these compounds. This phenomenon occurs because sub-lethal doses of antibiotics do not kill the bacteria but instead provide a selective pressure that favors the survival of individuals with resistance genes (ARGs). [24]

This could lead to a global health crisis if the majority of bacteria develop resistance to antibiotics. At that point, there would be no way to control the infections caused by these organisms in both humans and livestock. Furthermore, it is important to consider that many antibiotics, such as amoxicillin, are not fully metabolised by the body; about 60% is excreted in its active form. Even when degraded, it transforms into stable metabolites like penicilloic acid, which, despite losing its antibacterial capacity, can still provoke hypersensitivity and allergic reactions in humans, further complicating the clinical landscape of antibiotic pollution. [15]

### 1.8. Amoxicillin

Among the pharmaceuticals present in the environment, antibiotics such as amoxicillin are especially relevant because of their relationship with bacterial resistance. In Barcelona's case, one antibiotic group clearly predominates over the others: penicillins, the group to which amoxicillin belongs, represents approximately one third of all antibiotics used in the hospital setting. In 2022, according to the PRAN indicator, the following national data was obtained, demonstrating the predominance of penicillins compared with other antibiotics. [24]

PRAN INDICATOR (Spain, community sector)	DHD
Total systemic-use antibiotics (group J01)	17.48
Penicillins (J01C)	12.24
Other beta-lactams (J01D)	2.50
Macrolides, lincosamides and streptogramins (J01F)	3.19
Quinolones (J01M)	1.98
Tetracyclines (J01A)	1.58
Sulfonamides and trimethoprim (J01E)	0.56
Other antibacterials (J01X)	0.61

Figure 6: DHD of different drugs in Barcelona, self-made from information from [24]

DHD refers to the defined daily doses of a medication per 1,000 inhabitants per day.

On the other hand, within the penicillin group, the consumption of amoxicillin and amoxicillin/clavulanic acid in primary care from the ICS of Barcelona in 2025 was measured in packages, DDD and DHD:

Active ingredient	Packages	DDD	DHD
Amoxicillin	103,253	1,084,435.77	2.217
Amoxicillin-clavulanic acid	64,277	804,052.16	1.644

Figure 7: DDD and DHD of amoxicillin in Barcelona, self-made from information from the Catalan Health Institute (ICS)

DDD indicates the assumed average dose per day for a drug used for its main indication in adults.

Of this total amount consumed, according to official data from the medicine's technical data sheet, between 50% and 85% of the administered dose of amoxicillin is excreted through urine within a 24-hour period. In addition, an important proportion of this amount is excreted without being metabolised, meaning in its active form. In other words, the original molecule is broken down, but it may still retain partial biological activity or contribute to resistance, for example through compounds such as penicilloic acid. [36]

Amoxicillin works by stopping the growth of bacteria. Antibiotics like amoxicillin are not effective in treating colds, the flu, and other viral infections. It has the following molecular formula: C<sub>16</sub>H<sub>19</sub>N<sub>3</sub>O<sub>5</sub>S.

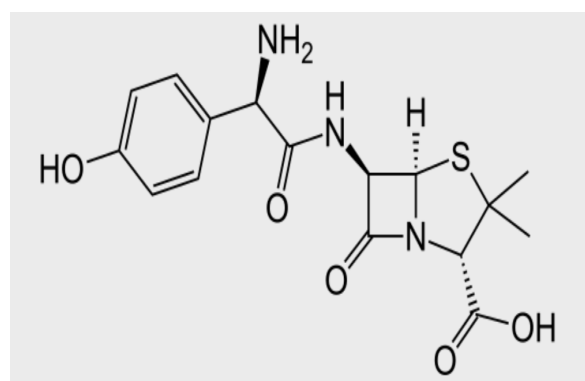


Figure 8: Amoxicillin structure [36.1]

Amoxicillin is available as a tablet, chewable tablet, or suspension (liquid) to be taken orally. It is usually taken every 12 hours (twice a day) or every 8 hours (three times a day). Amoxicillin can be taken with food to prevent stomach disruptions. The length of the treatment depends on the type of infection. [36]

According to the Spanish Agency for Medicines and Health Products [37], Amoxicillin is indicated for the treatment of the following infections in adults and children:

- Acute bacterial sinusitis;
- Acute otitis media;
- Acute streptococcal tonsillitis and pharyngitis;
- Acute exacerbation of chronic bronchitis;
- Community-acquired pneumonia;
- Acute cystitis;

- Asymptomatic bacteriuria in pregnancy;
- Acute pyelonephritis;
- Typhoid and paratyphoid fever;
- Dental abscesses with disseminated cellulitis;
- Prosthetic joint infection;
- Helicobacter pylori eradication;
- Lyme disease.

Amoxicillin is partially excreted in the urine in the inactive penicilloic acid form in amounts equivalent to 10 - 25% of the initial dose. That means that penicilloic acid is the metabolite of amoxicillin. [36]

### 1.8.1. Penicilloic acid

Penicilloic acid is any of several acids which are obtained from the penicillins by the hydrolytic opening of the lactam ring (by the action of a beta-lactamase). Hypersensitivity is the most important adverse effect of the penicillins. The major antigenic determinant of penicillin hypersensitivity is its metabolite, penicilloic acid, which reacts with proteins and serves as a hapten to cause an immune reaction. [38]

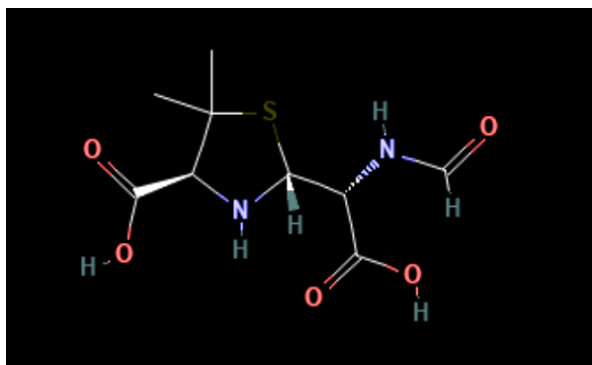


Figure 9: 2D penicilloic acid structure [38]

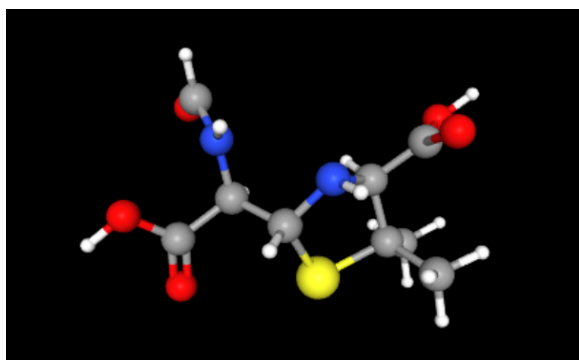


Figure 10: 3D penicilloic acid structure [38]

This acid has a molecular weight of 262.29 g/mol, a 141 Å<sup>2</sup> topological polar surface area and 17 heavy atoms. The largest specimens of this molecule can reach 1nm and the smallest of these cannot be calculated with precision but are about 0.025nm. [38]

This acid has several subtypes, one of which is benzylpenicilloic acid (BPNLA). This component can be harmful for human health and different organisms for the following reasons: [39]

- Cellular toxicity (cytotoxicity): BPNLA stops the growth of several types of cells (such as SK-N-SH or MRC-5).
- Alteration of the cell cycle: it blocks cells in the G1 phase, preventing them from passing to the S phase (DNA synthesis) and dividing correctly.
- Induction of apoptosis: toxic levels of BPNLA cause programmed cell death (apoptosis), mainly due to morphological changes in the cell.
- Presence in the food chain: the real danger lies in its formation and stability. It is generated when meat or milk with penicillin residues are subjected to high temperatures, making it resistant to cooking. On top of this, while penicillin is regulated, its degradation products, such as BPNLA, are often not monitored, despite being present in products such as yoghurt or cooked meat.
- Acute toxicity: in animal models (mice), an LD50 (standard measure of the acute toxicity of a substance) of 8.48 g/kg has been determined intraperitoneally. Although this is a relatively high value, the study warns that continued consumption of small doses through animal-derived foods could have long-term public health significance. [39]

Despite these risks, it must be clarified that the toxic concentrations tested in laboratories are higher than those typically found in the meat of animals treated with therapeutic doses of penicillin. [39]

## 2. Current water filtration in Barcelona

The water network in Barcelona is currently quite complex because it combines various resources and is designed both to guarantee supply and to manage droughts like the one the region recently experienced. Water is primarily supplied by the Ter river (the most important source, water is captured inland and brought

to the metropolitan area through the Ter-Llobregat pipeline system), the Llobregat river (which also contributes a significant share), and several aquifers (especially those of the Llobregat delta, which are used mainly in times of need). However, in recent years two alternative sources have been introduced, which are used on a smaller scale: seawater desalination and the reuse of treated water (reintroducing it into the river's flow before it is captured again by the water treatment plants). [25]

The urban water cycle in Barcelona is a continuous process: first, water is drawn from natural sources, then it is taken to plants where it is treated to make it potable for consumption, next it is transported and stored in reservoirs before being distributed to homes and industries, where it is used for daily activities; Once used, wastewater travels through the sewer system to treatment plants, where it is cleaned using physical, chemical, and biological processes, and can finally be reclaimed for reuse (for example, for irrigation or industrial uses) or returned to the natural environment, thus closing an increasingly circular and efficient cycle to address water scarcity. [26]

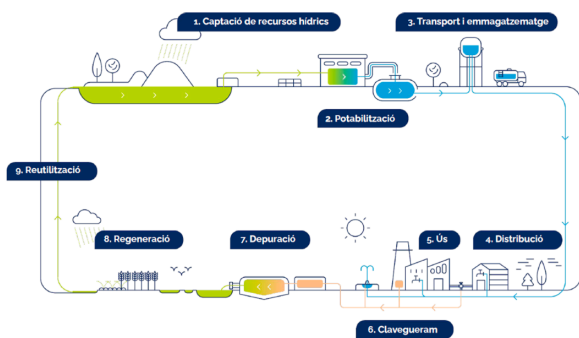


Figure 11: Cycle of water used in Catalonia [25]

The wastewater treatment plants (WWTP) that focus on treating the water of Barcelona are:

- Besòs WWTP: treats wastewater from the Northern metropolitan area;
- Baix Llobregat WWTP (El Prat): one of the largest, key for the South and for reuse;
- Montcada i Reixac WWTP: serves inland areas. [27]

In contrast, the WWTPs that purify the water before the consumption of the metropolitan area of Barcelona are:

- Ter WWTP (Cardedeu): treats water from the Ter river;

- Llobregat WWTP (Abrera): treats water from the Llobregat river;
- Sant Joan Despí WWTP: one of the most important, managed by Aigües de Barcelona, with advanced technologies (including partial desalination). [27]

The water purification process that is carried out in water treatment plants uses a series of steps. First of all, the water passes through a series of physical barriers in which all unwanted elements such as leaves or twigs are trapped. After this, pre-oxidation is done, reagents are added to the water to oxidise the organic matter. The first reagent added is ozone, which oxidises the organic matter in the water coming from the reservoir. The second reagent added to the water is PAC (Polyaluminium Chloride) which acts as a flocculating agent for the matter dissolved in the water; that is, it groups it in the form of flocs to facilitate its elimination in subsequent stages (sludge separation). In order to distribute the chemical reagents throughout the water column, the addition process takes place in tanks called “mixing chambers”, where the water is stirred by fast agitators. After the addition of reagents, the water is led to the flocculators, where there is slow agitation to “fatten” the flocs that have formed in this first stage. Then decantation is done, where the flakes settle to the bottom of the decanters by gravity, separating from the clear water. Following this, the water flows through sand and gravel filters. Sand and gravel filters are used to remove small particles that still remain in the water after decantation, such as very fine clays and silts (suspended soil), remains of organic matter (decomposed leaves, microscopic algae) or microorganisms. Finally, there are two options to inhibit the taste of water; activated carbon and nanofiltration. [27, 28]

Activated carbon filtration relies on adsorption, where molecules adhere to the material’s extensive surface area, providing moderate to good effectiveness for certain substances such as caffeine and ibuprofen; however, many pharmaceutical compounds are either too small or do not bind efficiently, allowing them to pass through, especially since pores around 50 nm are common in this system and permit smaller particles to escape. In terms of sustainability, this method has notable drawbacks because activated carbon has a relatively short lifespan and typically needs to be replaced every couple of years, requiring energy-intensive regeneration in high-temperature ovens that also generate significant gas emissions. A typical example is standard pitcher filters, which are mainly designed to enhance taste and remove chlorine

rather than fully eliminate pharmaceutical contaminants. [29, 30, 31, 32, 33]

Ultra/nanofiltration is a filtration approach that relies on a physical barrier with highly controlled and extremely small pore sizes of around 20 nm, significantly smaller and more precise than those found in activated carbon filtration, which allows for a more selective separation of contaminants; typically used in industrial settings, nanofiltration is effective at removing multivalent ions and organic compounds, while ultrafiltration targets larger molecules, benefiting from this tighter pore structure compared to activated carbon systems where larger and less uniform pores allow smaller particles to pass through. [28]

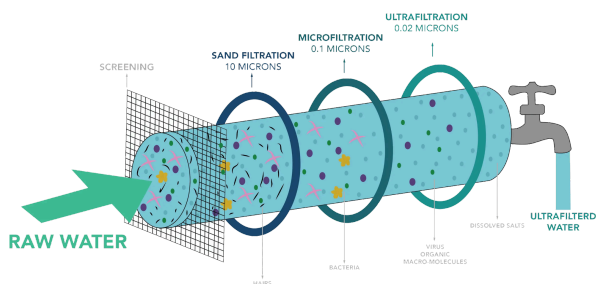


Figure 12: Diagram of the ultrafiltration process [28]

When water has remains of minerals or salts (or is directly salt water), the reverse osmosis process is implemented. Currently, since the rivers that supply water to Barcelona have high quantities of minerals and carry a good proportion of salts, this process is always used. It is a filtration method based on physical separation, where water is forced through a semipermeable membrane with extremely small pore sizes of around 0.1 nm, far smaller and more restrictive than those in activated carbon filtration, preventing a wide range of contaminants from passing through; as a result, it achieves high effectiveness (around 90–99%) in removing substances such as drugs, hormones, salts, and metals that cannot fit through these pores, although this performance comes with important considerations, including higher costs, the production of wastewater due to the waste-to-water ratio, and greater maintenance requirements compared to activated carbon systems. [34, 35]

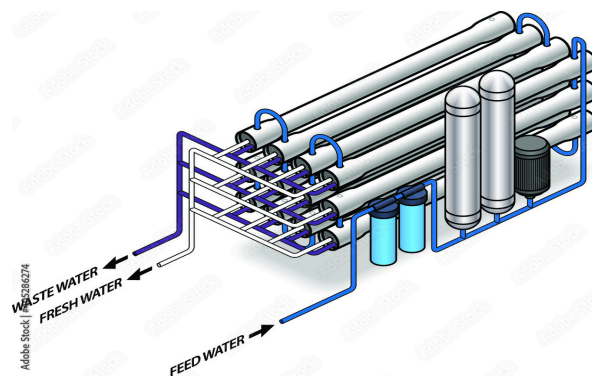


Figure 13: Diagram of reverse osmosis system [34]

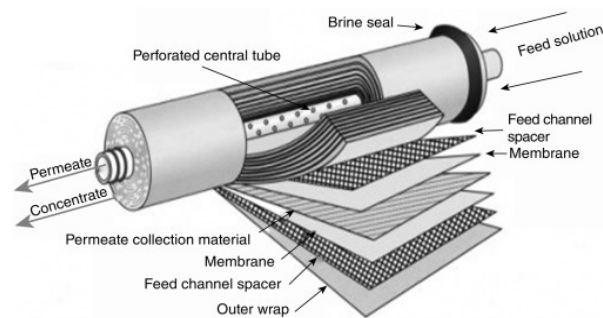


Figure 14: Diagram of reverse osmosis system [34]

### 3. Limitations of WWTPs

The main problem that WWTPs suppose is their sustainability, with many components having a very poor service life and needing to be replaced very frequently. Apart from that, in non-saline regions (unlike Barcelona), the maximum treatment applied to the water is nanofiltration, with a filtration capacity for particles larger than 20 nm. This is not sufficient to remove all APIs and other remaining contaminants. The solution currently being applied in Barcelona, reverse osmosis, is also problematic from a sustainability standpoint, as, upon requiring replacements, it pollutes on a large scale. [7, 8, 15]

Activated carbon-based filtration systems need to be replaced every 6-12 months. Approximately 2 to 6 kg of CO<sub>2</sub> are emitted for every kilogram of activated carbon produced. Additionally, to create this compound, very high temperatures (600-1000°C) are required, which results in enormous energy consumption. Furthermore, certain acids, such as phosphoric acid, are used in the activation process, which produces contaminated liquid waste. [31, 32]

The reverse osmosis system does have a longer lifespan, between 5 and 8 years. However, when producing the necessary components, it pollutes on a much larger scale. For every square meter of fabric produced, between 5 and 10 kg of CO<sub>2</sub> are emitted,

assuming each filtration cylinder requires 280 m<sup>2</sup>, and, as there are at least 50 of these in a single treatment plant, this figure scales up to 70,000 kg (minimum) of CO<sub>2</sub> each time these materials are renewed.

#### 4. Biochar for water filtration

Biochar, carbon generated from biomass, is produced by heating organic waste to high temperatures (300-700°C) in a low-oxygen environment, a process known as pyrolysis. Through it, the material is broken down into smaller molecules of gas and solids. [40] A portion of the gases yielded are condensable, and can be converted into bio-oil as a renewable fuel alternative, while the others are non-condensable, also known as syngas, and can be used as sustainable and versatile energy carriers. [41] As for the solid components that become biochar, they are obtained mainly from the lignin portion of the biomass. Lignin is an organic polymer that acts as a binder in plants, and is the second most abundant renewable carbon source, accounting for approximately 30% of the Earth's non-fossil carbon. [42]

Pyrolysis can be classified into several types based on heating rates:

- Torrefaction (up to 300°C): occurs at relatively low temperatures, below those needed for the significant breakdown of biomass components like cellulose, hemicellulose, and lignin.
- Slow pyrolysis (300-500°C): takes place over a long period of time, running from hours to days, allowing biomass to decompose gradually. The primary product formed during both torrefaction and slow pyrolysis is biochar.
- Fast pyrolysis (600-1000°C): involves rapid heating, resulting mainly in the production of bio-oil, along with smaller amounts of biochar and syngas.
- Flash pyrolysis (around 1000°C): occurs extremely quickly, in less than one second, and primarily produces vapours and aerosols. [43]

Char filters, whether made from activated carbon or biochar) operate through two processes. First, pollutants diffuse into the material's pores (absorption), after which these molecules bind to the surface of the char (adsorption). For this to happen, the number of reactive sites for the compounds to adhere to must be

substantial, underscoring the importance of having a large surface area and high porosity. [44]

However, building upon these shared adsorption mechanisms, the performance of biochar in water filtration differs from that of activated carbon in several aspects. The latter is viewed as the standard filtration material due to its high surface area (500-1500 m<sup>2</sup>/g) [32] and uniform pore structure, which allow the efficient adsorption of a wide spectrum of contaminants, including organic compounds, chlorine, and heavy metals. This high degree of porosity is achieved through additional activation processes that involve chemical treatments or high-temperature steam activation. [45]

In contrast, biochar exhibits a lower surface area and less uniform pore distribution, which results in a comparatively lower adsorption efficiency. Its performance is dependent on certain production conditions such as feedstock type and pyrolysis temperature. For instance, higher temperatures promote the development of a more porous structure and greater surface area, while lower and intermediate temperatures favour yield larger quantities and preserve a greater number of surface functional groups, which also play a significant role in adsorption. As a result, biochar production involves a balance between factors, rather than having a single optimal temperature. [45]

Despite its lower standard performance, biochar presents advantages in terms of cost and sustainability. Unlike activated carbon which requires very energy-intensive and costly processing, biochar is produced from organic waste materials with a relatively low energy input. This, apart from reducing production costs, contributes to the valorisation of waste thus making biochar a more environmentally sustainable option for water filtration. Additionally, according to Richardson et al. (2025), modified or engineered biochar can achieve adsorption capacities comparable to activated carbon for certain pollutants, which could, in the future, narrow the performance gap between these two materials. [45]

### 5. Practical framework

#### 5.1. Proposed solution

Based on the aforementioned problems that have been identified in current filtration systems in terms of the removal of pharmaceutical compounds, a more sustainable and cost-effective purification method is proposed as a substitution for activated carbon. The main objective is to explore biochar as an adsorbent

material capable of filtering said contamination, ensuring that the organic waste needed for its production is of proximity. One of the viable sources for this biomass in Spain are olive stones, deriving from a product that represents a high percentage of the agricultural goods generated in the country. This idea is based on the valorisation of an abundant agricultural residue, repurposing a by-product of the olive oil industry into a functional material for environmental remediation.

However, pharmaceutical contamination encompasses an extensive number of drugs, and in consequence, active components. It is for this reason that for the sake of this project the example of amoxicillin, the most prescribed antibiotic in the area, will be used.

### 5.1.1 Properties for adsorption

Olive stones are an important byproduct generated during olive oil extraction. As a lignocellulosic material, they are mainly composed of cellulose, hemicellulose, and lignin, along with smaller amounts of proteins, fats, phenolic compounds, free sugars, and polyols. This composition provides a carbon-rich structure, which makes olive stones a suitable precursor for biochar production. [35]

The lignocellulosic structure gives olive stones notable rigidity and stability, while their naturally irregular morphology contributes to the development of a porous structure after processing. In addition, their favorable elemental composition, characterised by high carbon content and low nitrogen and sulfur levels, enhances their suitability for thermal conversion processes such as pyrolysis. [46]

As a result, olive stones can be transformed into biochar, a carbon-based material with properties that make it suitable for environmental applications, particularly in water treatment. According to several studies that have analysed the properties of olive stones biochar, it is a carbon-rich material characterised by its well-developed internal structure. During the pyrolysis process at 500 degrees, the olive stone undergoes a significant increase in carbon content (rising from 46.9% in the raw stone to 84.1% in the biochar), thus creating a stable carbon matrix ready for filtration purposes. [47, 48]

The key properties that make olive stone biochar a good natural adsorbent filter are its high porosity and large specific surface area. To understand how this biochar works, activation after carbon formation is essential. While the original precursor has a negligible surface area, thermal treatment and subsequent

activation with CO<sub>2</sub> generate a sophisticated porous network. For example, biochar activated for 3 hours reaches a high specific surface area of 1349.3 m<sup>2</sup>/g and a total pore volume of 0.655 cm<sup>3</sup>/g, where micropores are predominant. This extensive surface provides a large number of active sites available for contaminant capture. [47]

In the specific case of antibiotic molecules, such as amoxicillin and its derivatives, biochar acts as an effective molecular trap. The material can capture these pollutants through several scientific mechanisms described in the article. Its porous structure allows for “pore filling”, where molecules become physically trapped within micro- and mesopores.

### 5.1.2. Availability and industrial context

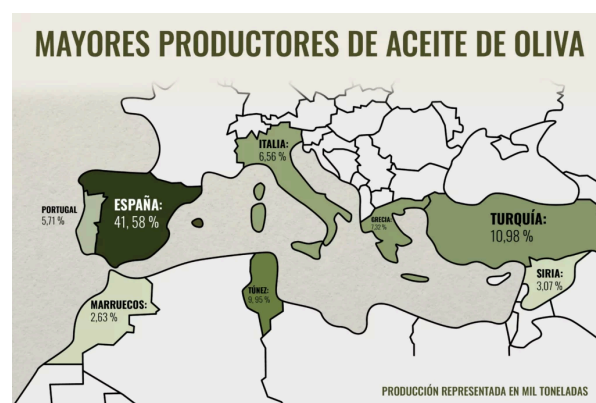


Figure 15: Countries according to production of olive oil [53]

Spain is among the countries in the world that contribute the most to the olive oil industry. According to the Ministry of Agriculture, Fisheries and Food. [49] In the 2024/2025 season Spain produced around 1.29 million tonnes of olive oil, which involves processing several million tonnes of olives each year. This volume is part of a global olive-growing area of approximately 11.7 million hectares, of which 86.5% is dedicated to olive oil production. [50, 51]. Within this context, Spain is the leading producer with approximately 2.7 million hectares, representing 24% of the global total, followed by Tunisia with 1.8 million hectares, Italy with 1.1 million hectares, and Greece, Morocco, and Turkey as other major producing countries. [52, 53]

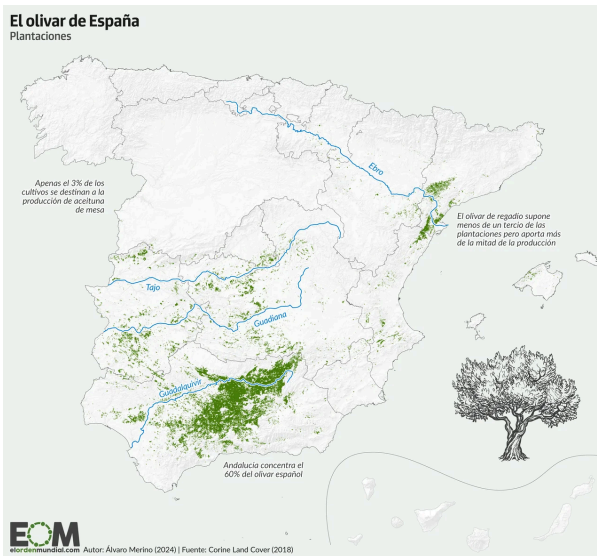


Figure 16: Regions in Spain with highest olive production [54]

Regarding national production within the industry, Andalusia accounts for nearly 60% of Spain's olive-growing area, followed by Castilla-La Mancha (16%), Extremadura (11%), Catalonia with 4%, and the Valencian Community with 3%. [54]

From an industrial perspective, the oil extraction process generates a large amount of by-products. According to data from the Official State Gazette (BOE, 2022), approximately 80% of the weight of processed olives corresponds to by-products, while only 20% is converted into oil. Among these by-products is the olive stone (pit), which accounts for roughly 15% to 20% of the total weight of the fruit. [55]

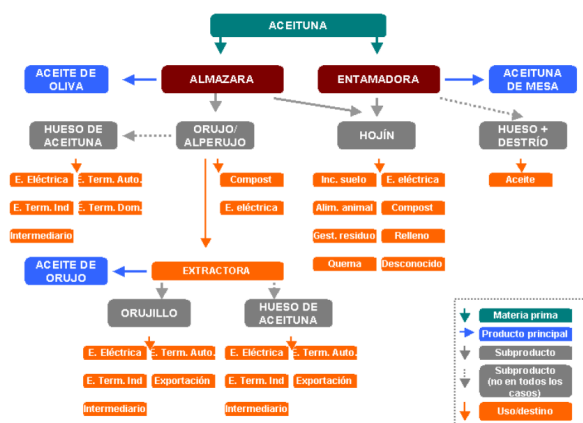


Figure 17: Utilisation model of the olive agro-industry [55]

This high volume makes olive pits a very abundant residue, especially in Mediterranean countries such as Spain, where production is intensive and highly geographically concentrated. However, this by-product

is not mainly discarded, but instead reused, primarily as biomass for energy production. [56]

### 5.1.3. Management of olive waste

As mentioned previously, the olive industry generates a large amount of agro-industrial waste as a result of the extraction and processing of olives. The main by-products include olive pomace, olive mill wastewater, pruning residues, and olive stones.

From an environmental perspective, these residues can have a significant impact if they are not properly managed. In particular, olive pomace, which consists of a mixture of water, organic matter, and solid residues, has a high moisture content and contains potentially toxic phenolic compounds. If it is discharged without control, it can lead to soil and water pollution. For this reason, public administrations have already developed regulatory frameworks to manage this waste, through regional legislation that establishes strict conditions regarding their use, dosage, and control in order to prevent negative environmental impacts. [57]

Currently, there are already reuse strategies in place. For example, the Regional Government of Andalusia uses these residues mainly for energy purposes (biomass, biogas) or agricultural applications (compost and organic soil amendments). However, the approximately 400,000 tonnes of olive stones generated annually require further innovation. [58]

In this context, the present proposal suggests the valorisation of olive stones through their transformation into biochar. The ash content is an important chemical property when assessing the suitability of olive stone by-products for applications such as soil amendments, structural materials, and bioenergy. Their high carbon (50.1–53.7%) and oxygen (38.8–41.9%) content enhances their calorific value, while low nitrogen and sulfur content reduces emissions, making them an environmentally friendly biomass option. Compared to other biomass materials, olive stone ash content is lower than that of wood species such as *Scytalidium* (7.5%) and wheat straw (6.1%), which increases its potential as an efficient and sustainable energy source. [46]

## 5.2. Visit to a wastewater treatment plant

The visit to the ETAP de Sant Joan Despí has helped the team understand in a practical and realistic way how the water purification process works in the Barcelona metropolitan area. Although the different stages of water treatment were already known

theoretically, observing the facilities in operation allowed for a better understanding of the industrial scale of the process, as well as the technical and energetic complexity involved in guaranteeing drinking water for millions of people.

During the visit, it was possible to directly observe how water from the Llobregat River arrives at the plant with a high concentration of salts, organic matter, and sediments, which explains why this water treatment plant requires much more advanced processes than other plants located in non-saline regions. One of the most relevant pieces of information extracted from the visit was that partial desalination through reverse osmosis is not simply a complementary system, but an essential and permanent part of the treatment of Llobregat water.

Another interesting aspect that was learned during the visit was the amount of waste generated throughout the treatment process, observing what happens to the sludge produced during decantation and to the filtration membranes once they reach the end of their useful life. This information helped establish a clearer connection between water treatment and the environmental sustainability problems later discussed in the project.

Finally, the visit helped the team better understand the strategic importance of the Sant Joan Despí treatment plant within Barcelona's water system, especially during periods of drought. It was observed how the plant is prepared to adapt to different water qualities depending on the availability of water resources, combining river water, reclaimed water, and advanced treatment processes to guarantee water supply.

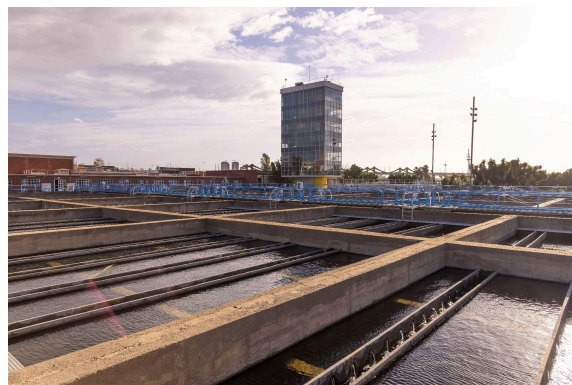


Figure 18: What is seen is an Archimedes screw, designed, when rotating, to lift water, fluids or solid materials (such as grain) from a lower area to a higher one. It consists of a

helical screw located inside a cylinder, arranged on an inclined plane. In this case, it serves to carry water from the physical separation area to the activated carbon pools. Image taken by authors.



Figure 19: Water extraction point of the Llobregat river during a period of drought. Image taken by authors.



Figures 19 & 20: Different processes can be seen, such as oxidation and filtration from organic matter to reduce hardness and ensure water quality before advanced treatment. Image taken by authors.



Figure 21: One of the first processes to which water is subjected, the filtration of large bodies through physical barriers.

### 5.3. Methodology

It is important to emphasise that the following methodology is entirely hypothetical. Although the process has been carefully designed and is scientifically grounded, it was not carried out experimentally due to the lack of access to the necessary resources. In particular, the research would require a non-school laboratory and specialised equipment, such as chromatographic systems, which were not available. Therefore, the steps described below represent what would have been done under appropriate conditions.

Biochar would have been produced from olive stone through a controlled pyrolysis process. The raw material would be cleaned, dried, and subjected to thermal treatment in an inert atmosphere to obtain a carbon-rich material with developed porosity. Additional activation processes would have been applied to improve its adsorption capacity.

To evaluate the effectiveness of the biochar, an aqueous solution containing amoxicillin (and its metabolite) would have been prepared. This solution would be brought into contact with the biochar under controlled conditions, allowing the adsorption process to take place. Samples would be taken at different times to study the removal efficiency of the contaminant. The concentration of amoxicillin before and after treatment would have been determined using chromatographic techniques, specifically HPLC. This method would allow the identification and quantification of the compound, making it possible to assess the reduction in concentration after filtration.

As the experimental procedure could not be performed, an AI-based video simulation has been developed to visually represent the process. This simulation illustrates the main steps of biochar production,

filtration, and contaminant removal, providing a clearer understanding of the proposed methodology.

Finally, a small-scale prototype of the filtration system has been designed using 3D designing technology. This model represents how the biochar could be implemented in a real filtration device, demonstrating the practical application of the proposed system.

#### 5.3.1. Biochar creation process

Olive pits would be used as the raw material for biochar production. Initially, they would be repeatedly washed with tap water in order to remove impurities and organic residues. Subsequently, they would be subjected to a drying process in an oven at 105 °C for 24 hours to eliminate residual moisture.

Once dried, the olive pits would be subjected to a pyrolysis process, consisting of thermal decomposition in the absence of oxygen. This process would be carried out in a tubular furnace under an inert nitrogen atmosphere. The temperature would gradually increase until reaching 500 °C, with a heating rate of 10 °C/min, and be then maintained for 30 minutes. [47]

After the thermal treatment, the system would be allowed to cool naturally to room temperature. The solid material obtained corresponds to the biochar, characterised by a carbon-rich structure with an initial development of porosity.

In order to improve the adsorptive properties of the obtained biochar, it would be subjected to three different activation processes (each of these activation methods as a different experimental group): a physical activation with carbon dioxide (CO<sub>2</sub>) and two chemical activations using phosphoric acid (H<sub>3</sub>PO<sub>4</sub>) and potassium hydroxide (KOH). To reliably compare the results, all samples would be treated under similar thermal conditions, varying only the activating agent used.

In the case of physical activation with CO<sub>2</sub>, the previously obtained biochar would be placed in a tubular furnace under an inert nitrogen atmosphere in order to prevent combustion reactions. The temperature would rise to approximately 750 °C. After reaching this temperature, the nitrogen flow would partially be replaced by carbon dioxide, and these conditions would be maintained for a period between 30 and 180 minutes. [47]

On the other hand, chemical activation would be carried out by impregnating the biochar, ensuring homogeneous contact between the activating agent and

the surface of the material, using the two activating agents separately, phosphoric acid and potassium hydroxide. For the impregnation of these two activators, an aqueous solution at 50% would be prepared in the case of phosphoric acid, and 56 g of KOH in 1 L of distilled water in the case of potassium hydroxide. Subsequently, the impregnated sample would be introduced into the tubular furnace and subjected to thermal treatment under controlled conditions, increasing the temperature to approximately 750 °C. [47]

Finally, the system, including the three activations and the control group, would be cooled again under an inert atmosphere. This process would significantly increase the specific surface area and porosity of the material, improving its adsorption capacity.

### 5.3.2. Efficacy evaluation

To evaluate the capacity of biochar as a filtering material, an amoxicillin solution in distilled water with a known concentration would be prepared. Specifically, a stock solution of 1 g/L.

The adsorption tests would be carried out in batch mode, in which the adsorbent and the contaminated solution are brought into direct contact in a closed container, without continuous flow. For this purpose, 500 mL of amoxicillin solution would be mixed with a determined amount of activated biochar (approximately 0.5 g), establishing an adsorbent dose of 1 g/L.

The adsorption tests would be carried out in batch mode, in which the adsorbent and the contaminated solution are brought into direct contact in a closed container, without continuous flow. For this purpose, 500 mL of amoxicillin solution would be filtered through a self-made mechanism (depicted and described in point 5.3.5.), maintaining controlled conditions of temperature (20 °C) and pH close to neutrality (~6.8). The contact time would vary between 5 and 420 minutes in order to study the adsorption kinetics.

### 5.3.3. Chromatography

To determine the concentration of amoxicillin present in the samples after the adsorption process, liquid chromatography would be used, a widely applied method for the separation, identification, and quantification of compounds in solution.

In this study, high-performance liquid chromatography (HPLC) would be used, in which the mobile phase

consists of a mixture of solvents that transport the sample through a chromatographic column containing the stationary phase. As the components pass through the column, they are separated according to their interactions with the stationary phase.

The identification of amoxicillin would be carried out by spectrophotometric detection at a specific wavelength. According to the European Pharmacopoeia, amoxicillin shows a characteristic absorbance in the ultraviolet region, and it is commonly detected at 254 nm. In the obtained chromatogram, amoxicillin is identified by its retention time, which corresponds to the time the compound takes to pass through the column from injection to detection.

To ensure correct identification, the retention time and the obtained signal would be compared with those of a standard amoxicillin solution. The match between both parameters confirms the presence of the compound in the sample. Likewise, a sample corresponding to the control group would be analysed, consisting of an amoxicillin solution without biochar subjected to the same experimental conditions. The comparison of its chromatogram with that of the treated samples made it possible to verify that any variation in the observed concentration was due to the adsorption process and not to degradation of the compound or other external factors.

This procedure would make it possible to accurately evaluate the effectiveness of the adsorbent material by comparing the initial and final concentration of the contaminant under different experimental conditions.

This information was learned due to a visit that was made to the laboratories of the Faculty of Pharmacy at the University of Barcelona, where it was possible to observe the necessary machinery.



Figures 21, 22 & 23: Equipment used to carry out chromatographies. Images taken by authors.

The results obtained by an analysis like this are collected in specific computer programs (which vary according to the machinery) and a report, like the one below, is obtained, in which information is obtained about the height, area and width of the peak awaited.

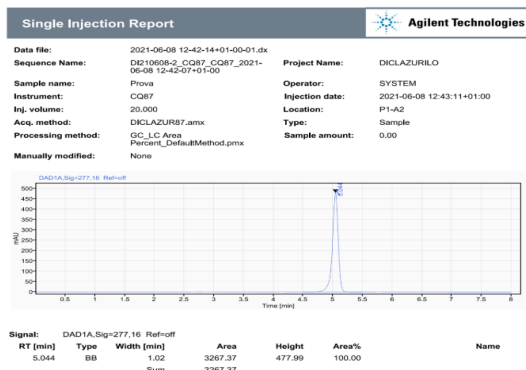


Figure 24: Example of a chromatography, resource given to the team by Barcelona's Faculty of Pharmaceutics

### 5.3.4. AI video simulation



As previously explained in the methodology section, the experimental procedure proposed in this project could not be physically carried out due to the lack of access to the necessary laboratory conditions and specialised equipment. In particular, the production of biochar through controlled pyrolysis and the subsequent chromatographic analysis would require facilities and instruments that were not available in a school laboratory environment. For this reason, the creation and testing of the olive stone-derived biochar remain hypothetical and are based entirely on bibliographic research and scientifically supported procedures.

Nevertheless, the methodology presented throughout the project describes the steps that would have been followed under appropriate laboratory conditions. These steps include the preparation and drying of the olive stones, the pyrolysis process under an inert atmosphere, the activation treatments designed to improve porosity and adsorption capacity, and finally the filtration and chromatographic analysis of

amoxicillin solutions in order to evaluate the efficiency of the proposed material.

To provide a clearer understanding of the theoretical procedure, an AI-generated video simulation was developed. The video visually recreates the different stages of the proposed experiment, including the production of biochar, the filtration process, and the hypothetical removal of pharmaceutical contaminants from water. It must be noted that, in the video, the adsorption process through biochar is not depicted to be carried out with the prototype described in the next point, as limitations were encountered while generating the video. Furthermore, although the simulation does not represent real experimental results, it serves as an educational and illustrative tool that allows the proposed methodology to be understood more easily and realistically.

### 5.3.5. Filter prototype

A prototype mechanism has been designed for the filtration of the amoxicillin solutions. It should be emphasised that this model could be adapted to different WWTP in accordance to specific conditions, such as the pipe radius, the direction in which the water is injected, or the length of the system.

This 3D recreation was created using the free CAD 3D project creation tool. In order to do this, the team members had to develop the necessary skills to use the servers.

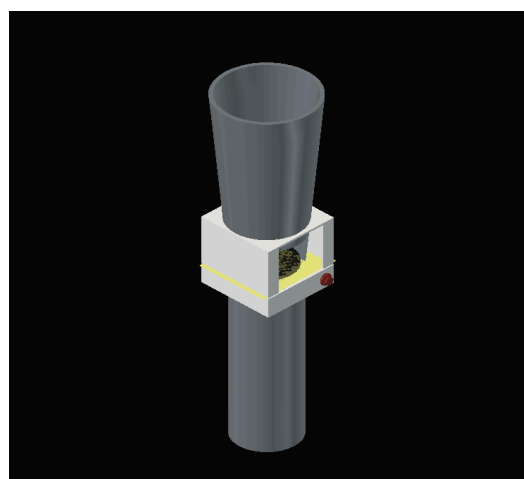


Figure 25: Angular view of the model

The water would enter through the top of the pipe, move through the filtration process in the filter inside the control box, and exit through the lower pipe.

The structure's body consists of 3 main parts: the pipe body through which the water would flow, the control box, and the filter. The upper pipe has a conical shape

to accelerate the water as it enters the outlet, thereby slightly optimising the energy used to transport it at the desired speeds. Due to the need for a surface to support the biochar, a filter bed was created that allows water to pass through. However, to be able to remove and replace this layer when necessary, a cut was made in the pipe, as seen below.

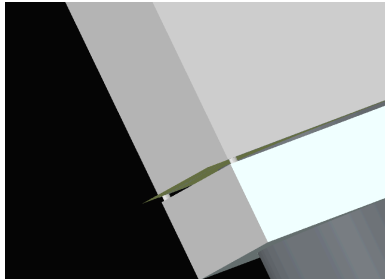


Figure 26: Image showing the aforementioned cut

The presence of this cut makes the control box, which is responsible for holding the two parts of the pipe together, even more important.

In addition, this duct has a hole that connects directly to the interior of the control box. This hole is covered with a hermetic door which is not specified in the three-dimensional visualisation to be able to see the interior.

The control box is the central part of the model, from where the entire filtration process can be controlled. It has three main functions.

First, it connects the upper part of the pipe with the interior through joints in the corners (which do not obstruct the passage of the filter).

Secondly, the filter does not hold itself up on its own, so a support system has been created for it based on cylindrical rods. This structure is a series of rods of different lengths that cross the entire diameter of the pipe. Being circular, they favor the inertia of the filter and do not obstruct its movement, while supporting it. Depending on the characteristics of the WWTP and the amount of biochar, a greater or lesser number of rods will be needed. For the hermetic sealing of the model at the joints, a variant of silicone called polydimethylsiloxane will be used [59], which will prevent water from spilling once the filter is inserted.

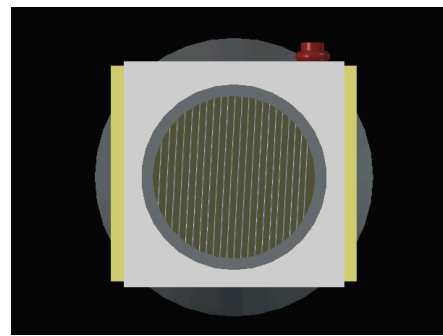


Figure 27: Rods that support the filter

The last function of the control box would be to regulate the amount of water flow, since it is advisable that the water to be filtered be in contact with the biochar for as long as possible. This is achieved due to an external valve (red in the three-dimensional representation) which reduces the radius of the pipe 10 centimeters below the filter (and the biochar).

The filter would be inserted through one of the openings in the pipe, sliding over the internal rods until it reaches the other part, where its movement would be fixed. This layer, responsible for maintaining the generated solid, while allowing water to pass through, would be a microporous PTFE membrane. This has a narrow pore size distribution. The membranes that fit into this group have a distribution of pore diameters of 0.001mm - 10mm. [60, 61, 62]

## 5.4. Sustainability of the proposed filter

Sustainability is an increasingly important factor in the development of water treatment technologies due to the growing pressure on global water resources. Therefore, sustainable water treatment aims to reduce environmental footprint by minimising energy consumption and reducing waste generation, as well as guaranteeing access to clean and safe water. [63]

For this reason, the proposed olive stone-derived biochar filter has been designed considering sustainability criteria. By transforming olive industry waste into an adsorbent material, the proposal intends to reduce dependence on environmentally compromising filtration methods.

### 5.4.1. Technical aspects

As explained throughout the project, olive stone biochar presents physicochemical properties that support its potential use as a filtration material for wastewater treatment. From a technical perspective, the proposed filter could theoretically be adapted to existing water treatment plants. The design developed during the project is intended to allow flexibility in its

implementation and scalability depending on the treatment requirements. Furthermore, the continuous production of olive industry residues in Spain suggests that the raw material required for large scale biochar production would remain constantly available.

However, important technical limitations must also be considered. Although activated biochar can improve adsorption performance, its efficiency generally remains lower than that of conventional activated carbon. Additionally, the effectiveness of biochar depends strongly on production conditions, something that can affect the consistency of the final material. Therefore, although the proposal is technically viable according to the research conducted, testing would be necessary to evaluate its real performance.

#### **5.4.2. Environmental impact**

The main sustainability advantage of the proposed filter is its use of olive stones, because, by transforming this waste product into an adsorbent material, it contributes to waste valorisation. In addition, it could theoretically reduce dependence on more environmentally demanding filtration systems based on industrial activated carbon or membrane technologies. The low sulfur and nitrogen content of olive stones also supports lower pollutant emissions during thermal treatment processes compared to other raw materials. Furthermore, the pyrolysis process also produces secondary products such as bio-oils and syngas that can be reused as energy sources.

Nevertheless, the environmental impact of the proposed system is not entirely negligible. The production of biochar through pyrolysis still requires energy consumption, and activation processes may involve intensive thermal or chemical treatments.

Regarding recyclability, biochar materials could be reused after adsorption processes through heating in order to remove the pharmaceuticals and later applied as a soil amendment. [64] However, repeated regeneration cycles may decrease adsorption efficiency, and the accumulation of contaminants could complicate disposal. Since these aspects were not experimentally tested during this project, the recyclability and long term performance of the proposed filter remain theoretical and require further investigation.

#### **5.4.3. Economic viability**

The proposed filtration system presents several economic advantages due to the abundance of olive industry residues in Spain. As these by-products are

widely available, the raw material required for biochar production would not depend on expensive extraction or manufacturing processes. Additionally, its local production would reduce transportation and import costs associated with conventional filtration materials.

There are still important economic challenges, however. Pyrolysis and activation process require specialised equipment and controlled production conditions, which may increase operational costs. Furthermore, large-scale integration into wastewater treatment facilities would require industrial infrastructure and the adaptation of existing systems. Consequently, although olive stone biochar could represent a more economically sustainable alternative in the long term, its economic viability would depend on further technological development.

### **6. Conclusions**

Pharmaceutical pollution has become an increasingly important environmental issue due to the constant release of active pharmaceutical compounds and their metabolites into aquatic ecosystems. As explained throughout this paper, conventional wastewater treatment systems are often unable to completely eliminate these substances, and their continuous presence contributes to toxicity in aquatic organisms, imbalances, endocrine disruption, and the development of antibiotic-resistant bacteria. In addition to the limitations in filtration efficiency, many traditional water filtration systems also present sustainability problems. This makes the search for more effective and sustainable water filtration systems necessary.

As a result of the bibliographic research carried out, olive stone-derived biochar was identified as a viable alternative to conventional filtration materials. The proposal is based on the valorisation of a locally abundant agricultural residue generated by the olive oil industry, transforming it into a potentially useful absorbent material through pyrolysis and activation processes. Focus has been placed on the adsorption of amoxicillin and its metabolite as a representative example of pharmaceutical contamination in Barcelona, as it is the most widely consumed antibiotic.

The investigation demonstrated that olive stones possess suitable properties for biochar production due to their lignocellulosic composition and high carbon content. Furthermore, the scientific literature analysed indicates that, after pyrolysis and activation, olive stone biochar can develop a porous structure with a relatively large surface area, characteristics associated with adsorption capacity. Different adsorption

mechanisms described in the literature, including pore filling and surface interactions, suggest that biochar is capable of retaining pharmaceutical contaminants under certain conditions.

However, the initial hypothesis of this project has ultimately been partially refuted. Although the research supports the technical feasibility of using olive stone biochar as a filtration material, the bibliographic analysis also demonstrates that its adsorption and filtration performance does not surpass that of current advanced systems such as activated carbon filtration or reverse osmosis. These technologies continue to provide higher contaminant removal efficiencies and more reliable filtration capacities, particularly for small pharmaceutical compounds.

Therefore, although olive stone biochar cannot currently be considered a superior replacement for existing filtration technologies, it may still represent a complementary and more sustainable alternative for certain water treatment applications. This project emphasises the importance of continuing research into environmentally responsible filtration materials capable of reducing both pharmaceutical contamination and the ecological impact associated with conventional purification systems.

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## Hydro Credit Project

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### Abstract

The degradation of water quality threatens ecosystems and communities, leading to unsafe water sources. Current solutions are often reactive and unsustainable for the community. This paper presents the Water Quality Trading (WQT) system in Thailand as a potentially more effective approach for managing pollutants such as nitrogen and phosphorus. Based on the carbon credit model, the proposed framework allows polluters to purchase credits and polluters who reduce pollution to earn tradable credits. The study focuses on the Bang Pa-in reach of the Chao Phraya River, Ayutthaya Province. A pilot policy framework was designed around the U.S. EPA WQT Toolkit, enabling industrial plants to buy nutrient reduction credits from upstream farms for proven pollution control practices. WASP8 was used to model total nitrogen and phosphorus concentrations under two scenarios: one following current discharge limits, one applying the WQT framework. Even with just two point sources included, the policy scenario cut total phosphorus loading by 58.1% and total nitrogen loading by 58.3%. A companion web platform, BPI-NCE, was built to handle credit issuance, trading, and compliance reporting across three user groups. Together, the results suggest that WQT is a workable and scalable approach to watershed nutrient management in Thailand.

### Keywords

Water Quality Trading, Thailand, Nitrogen, Phosphorus, Credit

## 1 Introduction

Water pollution remains a critical challenge in Thailand, particularly in watersheds subject to compounding pressures from industrial activities, rapid urban expansion, and intensive agricultural practices. These pressures directly threaten aquatic ecosystems, public health, and economic sectors dependent on water resources, including fisheries, tourism, and domestic water supply. Prolonged deterioration of water quality further contributes to hypoxia, eutrophication, and the loss of aquatic biodiversity, underscoring the urgency of effective and lasting management solutions [2].

Given the complexity of these challenges, water quality management cannot rely on any single approach. It requires the integration of scientific knowledge to understand and simulate pollutant behaviour with legal and policy frameworks that establish standards and regulatory obligations, and economic mechanisms that create incentives for efficient pollution reduction. Traditional command-and-control approaches, however, often lack the flexibility needed to address pollution at the watershed scale in a cost-effective manner. As a result, market-based mechanisms such as Water Quality Trading (WQT) [19] have gained increasing attention as complementary tools for

environmental management, offering the potential to achieve equivalent or greater environmental outcomes at lower overall cost.

In response to these opportunities and constraints, this study is conducted under the "WATER IS LIFE" project, which aims to develop a WQT framework tailored to the Thai regulatory and institutional context. The primary objective is to design a trading system and supporting policies that allow pollution sources to exchange water quality credits within defined environmental criteria. The framework draws on established international WQT principles while adapting them to Thailand's legal, economic, and data limitations. Key challenges include the absence of clearly defined discharge rights, regulatory frameworks that do not yet formally support credit trading, and concerns regarding the reliability of baseline data. To address these constraints, the project proposes a digital platform web-based to facilitate credit trading with enhanced transparency, reduced transaction costs, and improved monitoring, verification, and reporting capacity.

To demonstrate how such a system would function under real-world conditions, this study applies the Water Quality Analysis Simulation Program (WASP) model [21] to Bang Pa In district as a representative case study. The simulation allows for visualization of pollutant transport, changes in water quality parameters, and projected outcomes under varying trading scenarios all within a data-limited environment reflecting Thailand's current conditions. By integrating hydrological modelling with policy design, this research evaluates both the technical feasibility and the institutional prerequisites for implementing a WQT system in Thailand at the watershed level.

### 1.1 The purpose of the investigation

1. To design a Water Quality Trading (WQT) framework tailored to Thailand's legal, regulatory, and institutional context.
2. To establish a credit-based trading system that allows pollution sources to exchange water quality credits within defined environmental criteria, as a more flexible and cost-effective alternative approaches.
3. To apply the Water Quality Analysis Simulation Program (WASP) to a real watershed, simulating pollutant transport and projected water quality outcomes under varying trading scenarios.
4. To propose a web-based digital platform that facilitates credit trading with greater transparency, lower transaction costs, and improved monitoring, verification, and reporting capacity.

## 2 Context and Methodology

### 2.1 Drafting Thai Water Quality Trading Policy

#### 2.1.1 Understanding the Core Concept

Water Quality Trading is a market-based environmental policy mechanism that allows one regulated discharger (the "buyer") to meet its water quality-based effluent limitation by purchasing verified pollution reduction credits [20] from another party (the "seller") that has reduced its own discharges below the level required of it. The fundamental premise is economic: different sources of pollution within the same watershed face vastly different costs to achieve the same marginal reduction in a given pollutant. By allowing those who can reduce pollution cheaply to sell credits to those for whom reduction is expensive, WQT achieves the aggregate environmental outcome at a lower total cost while maintaining or improving overall water quality in the receiving waterbody.

These pollution sources generally fall into two categories. A point source is pollution that originates from a single, identifiable location such as a factory or sewage treatment plant discharging effluent through a permitted outflow pipe. Because the origin is discrete and traceable, point sources are subject to direct regulatory oversight. A nonpoint source, by contrast, has no single identifiable origin; pollution instead accumulates from many scattered inputs across a landscape, such as rainwater washing fertilizer, sediment, and other contaminants from agricultural land into a waterway. Because nonpoint sources cannot be tied to one responsible party or location, they are far more difficult to regulate through traditional permitting. WQT is particularly valuable in this context precisely because it creates a mechanism for nonpoint sources which are often able to reduce pollution at a much lower cost to generate credits that regulated point sources can purchase to meet their obligations.

A credit represents a verified unit of pollutant load reduction, typically expressed in pounds per day or kilograms per day. Credits are only generated when a seller reduces its discharge below its established baseline which is the pollution control requirement that would apply in the absence of trading. A buyer may only use purchased credits to satisfy a water quality-based effluent limitation (a limit derived from what the waterbody needs to meet water quality standards), not a technology-based limit (a minimum treatment standard applied regardless of receiving water quality). This distinction is critical: WQT does not allow sources to buy their way out of basic pollution control obligations. It allows them to satisfy requirements above that baseline more efficiently.

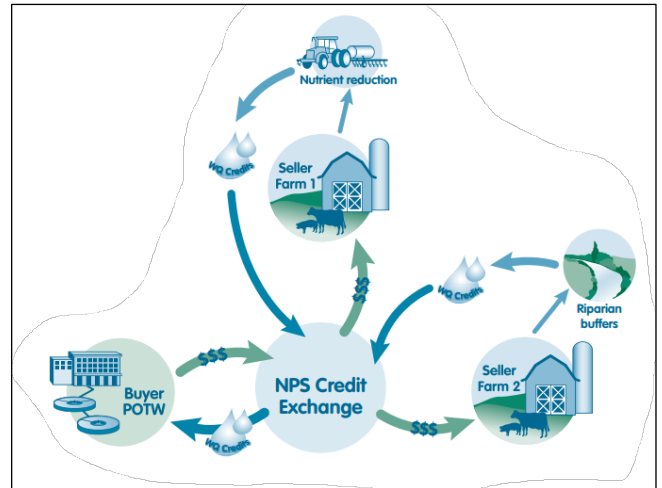


Figure 1: Credit Trading System

#### 2.1.2 Analysing Watershed assessment and feasibility

The process begins with a rigorous characterisation of the target watershed. The analyst must identify the dominant pollutants driving water quality impairment, typically nitrogen, phosphorus, and BOD for Thai river basins, and map every significant source across point and nonpoint categories. Critically, this process must quantify the marginal cost of pollution reduction for each source type: what does it cost an industrial facility to remove pre-established removal efficiencies one kilogram of total phosphorus through treatment [1], compared with what it costs a rice farmer to achieve the same reduction through conservation tillage or a riparian buffer? If this cost differential is not large, the economic rationale for trading collapses. Data from the PCD's (Pollution Control Department) monitoring network, the Department of Water Resources, and the Royal Irrigation Department should be synthesised here [10,13,14].

#### 2.1.3 Legal authority and regulatory foundation

Before any trade can be authorised, the legal basis must be unambiguous. For Thailand, the most practical pathway is a Ministerial Regulation under the Water Resources Act 2018 that formally creates the category of "pollution reduction credits," defines eligible pollutants, and specifies that WQT is a supplement for existing effluent standards. In parallel, PCD and DIW (Department of Industrial Works) must jointly develop and issue discharge monitoring permits for all point source participants in the pilot basin [6]. These permits are the scaffolding on which trading provisions will be hung; without them, there is no verifiable baseline from which credits can be measured.

### 2.1.4 Baseline setting and credit generation rules

This process defines the floor below which reductions become credits. For regulated point sources, the baseline is their most stringent applicable effluent standard that credits are generated only for reductions below that level, so treatment technology obligations remain non-negotiable. For agricultural nonpoint sources, baselines must be derived through a combination of standard pollutant loading models, field surveys, and local expert knowledge from agricultural extension officers. The regulatory authority must then publish an approved BMP (Best management practices) list with pre-established removal efficiencies [20] and uncertainty ratios for each practice, giving farmers clear and predictable rules and giving permit writers a defensible basis for credit approval. A credit should be defined as one unit of load reduction (kilogram per day) at the buyer's discharge location, with adjustments applied for distance.

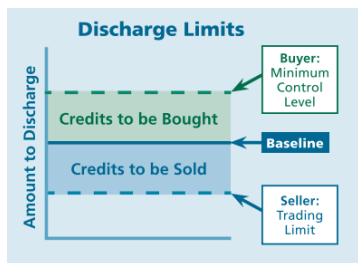


Figure 2: Discharge Limits

### 2.1.5 Trade ratio derivation

Trade ratios are the mathematical safeguard that ensures environmental integrity across every transaction [20]. The ONWR (Office of the National Water Resources), working through the relevant river basin subcommittee, should commission a watershed hydrological model to calculate delivery ratios: the fraction of a pollutant load reduced at an upstream location that actually reaches the downstream waterbody of concern. For the Chao Phraya, delivery ratios of 3:1 or greater are likely for upstream agricultural trades given the river's length and attenuation dynamics [20]. Equivalency ratios must also be calculated where different chemical forms of the same pollutant are being traded. Finally, uncertainty ratios are applied to nonpoint source credits specifically, to buffer against measurement and modelling imprecision. All ratios should be basin-specific and subject to periodic revision as monitoring data accumulate.

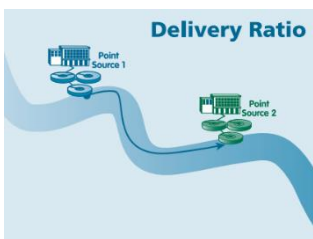


Figure 3: Delivery Ratio

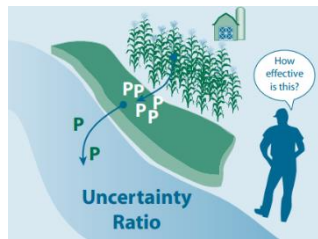


Figure 4: Uncertainty Ratio

### 2.1.6 Credit exchange design and permit integration

Once baselines and ratios are established, the trading mechanism itself must be built. A third-party credit exchange, operated by a basin subcommittee office, an accredited NGO (Non-Governmental Organisation), or a designated government body should aggregate credits from multiple agricultural sellers, verify BMP performance, set standard credit prices, and sell credits to industrial buyers. This pooled structure removes the burden of individual negotiation from both farmers and industry. The permitting authority must then formally incorporate trading provisions into point source discharge permits, either by referencing the trade agreement or embedding its key terms as permit conditions. A publicly accessible digital registry tracking credits generated, transferred, and retired is essential for transparency and public trust.

### 2.1.7 Monitoring, verification, and adaptive management

A WQT program without credible verification will fail both environmentally and politically. Point source buyers must submit verified end-of-pipe discharge monitoring data at prescribed frequencies. Nonpoint source BMPs must be subject to random field audits by the permitting authority or the credit exchange. Ambient water quality monitoring stations should be placed at strategic points within the trading zone to detect any localised water quality violations that aggregate credit accounting might miss. An annual public evaluation report, covering both environmental outcomes (measured changes in loading at key monitoring stations) and economic outcomes (cost per unit of reduction achieved through trading versus technology compliance) provides the feedback loop for adaptive management, enabling the regulatory authority to revise trade ratios, update the BMP list, and adjust program rules as evidence accumulates.

## 2.2 Simulating Water Quality Analysis

### 2.2.1 Model and Program Selection

The Water Quality Analysis Simulation Program version 8 (WASP8) [21], developed by the U.S. Environmental Protection Agency, was selected to simulate nutrient transport and fate in the study reach. The model was configured through five sequential setup steps within the WASP8 interface.

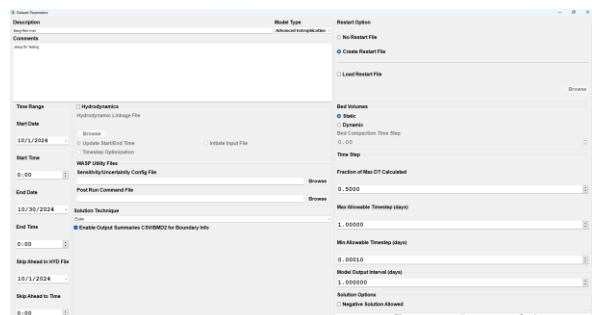
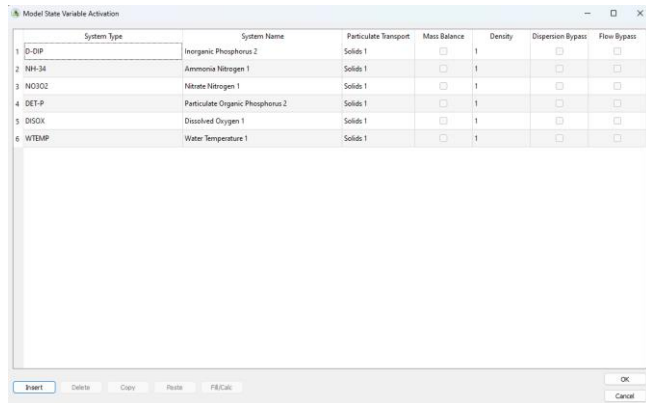


Figure 5: Initialize project

## 2.2.2 Dataset initialization

A new project dataset was created to define the simulation period and the initial concentration of each water quality constituent across all segments. Initial values for dissolved oxygen (DO), biochemical oxygen demand (BOD), ammonia-nitrogen (NH<sub>3</sub>-N), nitrate-nitrogen (NO<sub>3</sub>-N), dissolved inorganic phosphorus (DIP-P), and detrital phosphorus (DET-P) were assigned based on field measurements from REO-6 monitoring station CH18 [12].



System Type	System Name	Particulate Transport	Mass Balance	Density	Dispersion Bypass	Flow Bypass
DIP	Inorganic Phosphorus 2	Solids 1	<input type="checkbox"/>	1	<input type="checkbox"/>	<input type="checkbox"/>
NH3-N	Ammonia Nitrogen 1	Solids 1	<input type="checkbox"/>	1	<input type="checkbox"/>	<input type="checkbox"/>
NO3-N	Nitrate Nitrogen 1	Solids 1	<input type="checkbox"/>	1	<input type="checkbox"/>	<input type="checkbox"/>
DET-P	Particulate Organic Phosphorus 2	Solids 1	<input type="checkbox"/>	1	<input type="checkbox"/>	<input type="checkbox"/>
DO	Dissolved Oxygen 1	Solids 1	<input type="checkbox"/>	1	<input type="checkbox"/>	<input type="checkbox"/>
WTEMP	Water Temperature 1	Solids 1	<input type="checkbox"/>	1	<input type="checkbox"/>	<input type="checkbox"/>

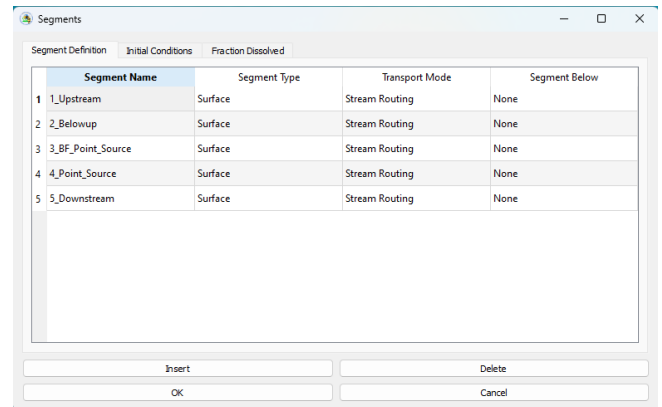
Figure 6 : Dataset Characterization

## 2.2.3 Function System

The EUTRO module was selected as the functional system to enable simulation of nitrogen and phosphorus cycling, algal dynamics, and dissolved oxygen interactions. Constituent state variables activated for this simulation were NH<sub>3</sub>-N, NO<sub>3</sub>-N, DIP-P, DET-P, DO, and BOD, consistent with the pollutants targeted by the WQT framework.

## 2.2.4 Segment Definition

The study reach is located within Bang Pa In district, Ayutthaya Province, along the Chao Phraya River. It was discretised into five longitudinal segments totalling approximately 18.5 km. Segment geometry including length, surface width, and cross-sectional depth was derived from Google Earth imagery and Royal Irrigation Department (RID) bathymetric data (2019) [13]. A uniform depth of 11.21 m was applied across all segments based on the bathymetric survey average.

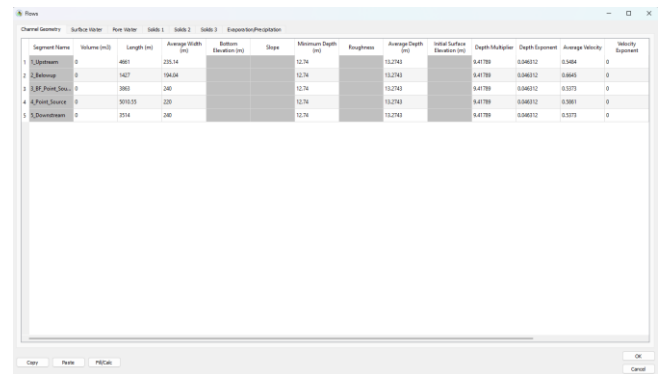


Segment Name	Segment Type	Transport Mode	Segment Below
1_1_Upstream	Surface	Stream Routing	None
2_2_Belowup	Surface	Stream Routing	None
3_3_BF_Point_Source	Surface	Stream Routing	None
4_4_Point_Source	Surface	Stream Routing	None
5_5_Downstream	Surface	Stream Routing	None

Figure 7: Watershed Delineation

## 2.2.5 Flow Module

The flow module was used to define both advective transport and point source inputs. Upstream boundary flow was specified using daily discharge records from RID gauging station C.29A for October 2024 [7,14], representing wet season conditions (1,000–2,000 m<sup>3</sup>/s). Point source flows were entered separately for PS-1 (TTW WWTP, 14,000 m<sup>3</sup>/d discharging to Segment 4) [17] and PS-2 (Rojana Industrial Park WWTP, 48,000 m<sup>3</sup>/d discharging to Segment 3) [7,15], with constituent concentrations assigned according to each scenario.

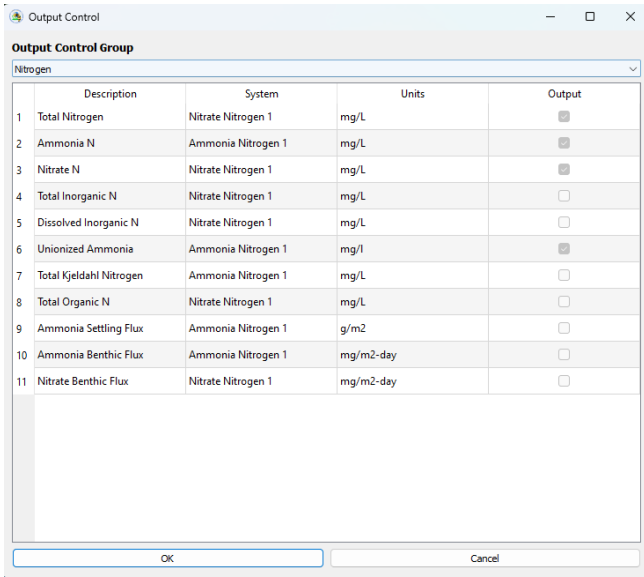


Segment Name	Volume (m <sup>3</sup> )	Length (m)	Average Width (m)	Bottom Elevation (m)	Slope	Minimum Depth (m)	Frictionless	Average Depth (m)	Total Surface Elevation (m)	Depth Multiplier	Depth Exponent	Average Velocity	Stability Exponent
1_1_Upstream	0	4801	233.14			12.76		11.2763		0.47789	0.548512	0.3484	0
2_2_Belowup	0	1427	194.04			12.76		11.2763		0.47789	0.548512	0.3484	0
3_3_BF_Point_Source	0	980	240			12.76		11.2763		0.47789	0.548512	0.3375	0
4_4_Point_Source	0	593.53	240			12.76		11.2763		0.47789	0.548512	0.3383	0
5_5_Downstream	0	3714	240			12.76		11.2763		0.47789	0.548512	0.3375	0

Figure 8: Hydrological Flow Modeling

## 2.2.6 Output Control

The output control panel was configured to record time-series concentrations of all activated constituents at each segment boundary. Segment 5 (downstream compliance point, corresponding to REO-6 station CH20) was designated as the primary reporting location for scenario comparison.



Description	System	Units	Output
1 Total Nitrogen	Nitrate Nitrogen 1	mg/L	<input checked="" type="checkbox"/>
2 Ammonia N	Ammonia Nitrogen 1	mg/L	<input checked="" type="checkbox"/>
3 Nitrate N	Nitrate Nitrogen 1	mg/L	<input checked="" type="checkbox"/>
4 Total Inorganic N	Nitrate Nitrogen 1	mg/L	<input type="checkbox"/>
5 Dissolved Inorganic N	Nitrate Nitrogen 1	mg/L	<input type="checkbox"/>
6 Unionized Ammonia	Ammonia Nitrogen 1	mg/l	<input checked="" type="checkbox"/>
7 Total Kjeldahl Nitrogen	Ammonia Nitrogen 1	mg/L	<input type="checkbox"/>
8 Total Organic N	Nitrate Nitrogen 1	mg/L	<input type="checkbox"/>
9 Ammonia Settling Flux	Ammonia Nitrogen 1	g/m2	<input type="checkbox"/>
10 Ammonia Benthic Flux	Ammonia Nitrogen 1	mg/m2-day	<input type="checkbox"/>
11 Nitrate Benthic Flux	Nitrate Nitrogen 1	mg/m2-day	<input type="checkbox"/>

Figure 9: Model Output Specification

## 2.3 Designing Digital Platform

The BPI-NCE (Bang Pa-in Nutrient Credit Exchange) platform is a web-based system designed to operationalise the WQT framework within the Bang Pa-in sub-watershed, Ayutthaya Province. It mediates nutrient credit transactions between industrial point-source (PS) buyers and agricultural non-point-source (NPS) sellers under the regulatory oversight of Thailand’s Pollution Control Department (PCD), tracking both Total Nitrogen (TN, reconciled annually) and Total Phosphorus (TP, reconciled monthly).

### 2.3.1 System Architecture

The platform adopts a role-separated, three-application architecture deployed as a TypeScript monorepo. Three independent Next.js 14 web applications [22] serve distinct classes of users, enforcing the principle of least privilege at the network boundary so that no portal can access another’s routes or data.

Table 1: Three-portal architecture

Portal	Primary Users & Functions
Authority Portal	PCD officers approve BMP applications, issue credit certificates, execute compliance reconciliation
Buyer Portal	Industrial point-source facilities browse credit market, submit purchase requests, file discharge monitoring reports
Seller Portal	Agricultural non-point-source farms submit BMP applications, track inspection status, view credit earnings

All three applications and their shared packages are managed in a single pnpm workspace orchestrated by Turborepo. Shared packages consolidate the tRPC API router [16], PostgreSQL schema and migrations (Drizzle ORM) [3], a React component library, and pure business-logic functions

(credit calculation, compliance reconciliation) so that logic is written once and consumed consistently across every portal.

### 2.3.2 Technology Stack

Technology selections were governed by three criteria: end-to-end type safety, regulatory auditability, and deployment simplicity appropriate for a pilot-scale government-adjacent system.

Table 2: Core technology stack

Layer	Technology
Frontend	Next.js 14 (App Router) , React 18, TypeScript 5
API	tRPC v11 end-to-end type-safe procedures
Database	PostgreSQL via Drizzle ORM ACID-compliant, versioned migrations
Authentication	NextAuth.js v4 [8] with signed JWT sessions, Passwords are hashed using bcrypt, following OWASP recommendations [9]
Deployment	Railway (container PaaS) three independent services, CI/CD via GitHub

The combination of tRPC and Drizzle ORM ensures that a schema change propagates as a TypeScript compile error to every API caller, preventing the class of silent contract-drift failures common in REST-based systems. All three portals are deployed independently on Railway [11] via a GitHub-triggered CI/CD pipeline, allowing zero-downtime updates to individual portals without affecting the others.

### 2.3.3 Trading and Compliance Workflow

The end-to-end trading workflow proceeds through eight role-gated stages:

- NPS seller submits a BMP application specifying practice type, parcel area (rai), and GPS coordinates.
- A field inspector conducts an on-site audit and records a PASS, FAIL, or CONDITIONAL result with dual-signature validation.
- A permit officer approves the application, initiating a 12-month monitoring lock period.
- Upon lock expiry, the permit officer issues a credit certificate with net credits calculated from the formula below.
- A PS buyer browses available certificates, submits a purchase request specifying quantity, agreed price per kg, and target compliance period.
- The permit officer approves the purchase; the certificate transitions to SOLD and the transaction to SETTLED.
- The buyer files a Discharge Monitoring Report declaring actual discharge and credits applied.

- The system automatically computes compliance status (COMPLIANT, DEFICIENT, or MCL\_VIOLATION).

### 2.3.4 Credit Calculation

Credit certificates are issued following the 12-month BMP monitoring period. Net tradable credits are derived from gross pollution reduction adjusted by a set of watershed-specific ratios, consistent with U.S. EPA Water Quality Trading methodology [20]:

$$\text{Net Credits (kg)} = [\text{Gross Reduction} \div (\text{DR} \times \text{LR} \times \text{ER} \times \text{UR})] \div \text{RR}$$

Table 3: Credit calculation parameters

Parameter	Role in calculation
Gross Reduction	Measured pollutant reduction attributed to the BMP practice
DR — Delivery Ratio	Accounts for transport losses between farm parcel and receiving waterbody
LR — Location Ratio	Adjusts for spatial proximity of seller to buyer's discharge point
UR — Uncertainty Ratio	Discounts gross reduction for monitoring and modelling imprecision
RR — Retirement Ratio	Permanently retires a share of credits to ensure net environmental benefit (pilot: 2 - 50% sellable)

All ratio values are drawn from the active trading\_ratio\_versions record at the time of certificate issuance and stored immutably on the certificate row, ensuring retrospective ratio updates do not alter previously issued credits.

Compliance is evaluated per reconciliation period as: Net Obligation = Actual Discharge – WQBEL – Credits Applied. A non-positive result yields COMPLIANT status; a positive result within the minimum control level (MCL) yields DEFICIENT (eligible for additional credit purchase); and any discharge exceeding the MCL yields MCL\_VIOLATION regardless of credits held, preserving the regulatory floor that credits cannot substitute for on-site treatment obligations.

### 3 Results and Discussion

This study compares two scenarios. **Scenario A** represents current discharge conditions with no trading policy in effect. **Scenario B** implements the proposed Bang Pa-in WQT Pilot Policy Framework, under which each point source must meet a three-tier discharge obligation combining on-site treatment and credit purchases. The following subsections describe the structure of Scenario B in detail.

#### 3.1 Policy Adoption under Scenario B

Scenario B operationalises the Bang Pa-in WQT Pilot Policy Framework through a three-tier discharge obligation imposed on each point-source (PS) facility within the Bang Pa-in Industrial Estate. The tiers are cumulative: a facility must satisfy the lower tier before the upper tier becomes relevant, and credit purchases are permitted only in the uppermost tier.

##### 3.1.1 Tier 1 — Technology Floor (Minimum Control Level)

Each facility is required to first achieve its Minimum Control Level (MCL) through on-site treatment, independently of any credit market activity. Under Section F of the Bang Pa-in WQT Policy, all industrial dischargers within the estate must attain at least 60% Total Nitrogen (TN) removal and 70% Total Phosphorus (TP) removal through secondary biological treatment with basic nutrient removal. This obligation is grounded in the Ministry of Industry Notification B.E. 2537 and IEAT Notification 029/2567 [4,5]. Credit purchases cannot substitute for the MCL under any circumstances.

Table 4: Minimum control level requirements for all PS buyers under Scenario B (Bang Pa-in WQT Policy, Section F).

Requirement	Value	Legal Basis
Minimum TN removal (on-site)	≥60%	Section F, Bang Pa-in WQT Policy; Ministry of Industry Notification B.E. 2537
Minimum TP removal (on-site)	≥70%	Section F, Bang Pa-in WQT Policy; IEAT Notification 029/2567
Credit substitution for MCL	<b>Not permitted</b>	Section 2.1 and Section F, Bang Pa-in WQT Policy

##### 3.1.2 Tier 2 — Water Quality-Based Effluent Limit (WQBEL)

Where the receiving waterbody fails to meet Thailand’s Class 3 water quality standards, a more stringent water quality-based effluent limit (WQBEL) is imposed above the technology floor. The WQBEL for this pilot is derived from the U.S. EPA eutrophication guideline of  $TN \leq 0.30$  mg/L, a threshold already exceeded by approximately two-fold at REO-6 monitoring station CH18 ( $NH_3-N + NO_3-N = 0.58$  mg/L, February 2026). The effective TP outfall target is set at 1.0 mg/L for both PS-1 and PS-2, consistent with interim ambient water quality criteria applied in the pilot watershed model. It is precisely because meeting these tighter limits through conventional treatment can be very expensive that trading becomes economically attractive and environmentally justified [Section 2.2, Bang Pa-in WQT Policy].

##### 3.1.3 Tier 3 — Credit Purchase to Bridge the Gap

The gap between the MCL and the WQBEL constitutes the only portion of the discharge obligation that may be offset through credit purchases (Section 7, Bang Pa-in WQT Policy). Each facility independently reduces its effluent by approximately 20% beyond the MCL through on-site upgrades, then purchases verified credits from upstream non-point source (NPS) sellers — specifically, farmers implementing approved Best Management Practices (BMPs) under Section G.2 of the policy — to bridge the remaining gap to the WQBEL. The resulting effluent reduction structure for each point source is summarised in Table 2.

Table 5: Point source effluent reduction structure under Scenario B, showing on-site treatment and credit purchase components for PS-1 and PS-2.

Parameter	PS-1 TTW WWTP	PS-2 Rojana WWTP
Flow (m <sup>3</sup> /d)	14,000	48,000
Scenario A — TP outfall (mg/L) [TBEL level]	2.0	2.5
On-site reduction (~20% beyond MCL)	1.6 mg/L	2.0 mg/L
WQBEL target (mg/L) [met via credit purchase]	1.0	1.0
Scenario B — effective TP outfall (mg/L)	1.0	1.0
TP load Scenario A (kg/d)	28	120
TP load Scenario B (kg/d)	14	48

##### 3.1.4 Trading Ratios Applied

Because a kilogram of pollutant reduced upstream does not exert the same effect at the downstream compliance point as a kilogram reduced at the discharge pipe, all credits are subject to a set of watershed-specific trading ratios specified in Section E of the Bang Pa-in WQT Policy Framework.

These ratios are applied at the time of credit certificate issuance and stored immutably, so that subsequent ratio revisions do not alter previously issued certificates. The ratios applied under Scenario B are presented in Table 3.

Table 6: Trading ratios applied under Scenario B (Bang Pa-in WQT Policy, Section E).

Ratio	Value	Applied to	Basis (Section E)
Delivery Ratio (DR)	1.0	PS-1 TTW	Direct discharge adjacent to Seg. 4; no attenuation
Delivery Ratio (DR)	0.70	PS-2 Rojana	~5–8 km via canal; preliminary estimate
Uncertainty Ratio (UR)	1.5	All PS–NPS trades	Phase 1; no established field monitoring; reduces to 1.25 after 2 years of NPS data
Retirement Ratio (RR)	2:1	All trades	Pre-TMDL Phase 1; 1 kg retired per 2 kg generated; ensures net watershed reduction
Equivalency Ratio (ER)	1.3–1.8	PS–NPS (TN)	Agricultural organic-N vs. ammonium-N; 55–75% bioavailability in Thai tropical conditions
Location Ratio (LR)	1.5–4.0	NPS sub-catchments	Varies by canal path length and residence time in irrigation network

### 3.1.5 Compliance Determination

A facility’s compliance status is evaluated per reconciliation period using the net obligation formula derived from Section 7 of the Bang Pa-in WQT Policy:

$$\text{Net Obligation} = \text{Actual Discharge} - \text{WQBEL} - \text{Credits Applied}$$

Three compliance states are defined, consistent with Sections 7 and F of the policy. A non-positive net obligation yields **COMPLIANT** status. A positive net obligation where actual discharge remains within the MCL yields **DEFICIENT** status, indicating that additional credit purchases are required to achieve compliance. Any discharge exceeding the MCL yields **MCL\_VIOLATION** status regardless of credits held, and is subject to enforcement action; credits cannot remedy an MCL breach under any circumstances.

## 3.2 Simulation Results

### 3.2.1 Model Configuration and Boundary Conditions

The WASP8 EUTRO module was applied to an 18.5 km reach of the Chao Phraya River within Bang Pa-in district, Ayutthaya Province, discretised into five longitudinal segments. Simulations were conducted under wet-season conditions representative of October 2024, during which upstream discharge at RID gauging station C.29A ranged from 1,000 to 2,000 m<sup>3</sup>/s. Upstream boundary concentrations were held constant across both scenarios, based on field measurements recorded at REO-6 [12] monitoring station CH18 in February 2026: dissolved oxygen 5.2 mg/L, BOD 2.1 mg/L, NH<sub>3</sub>-N 0.14 mg/L, NO<sub>3</sub>-N 0.44 mg/L, and TP 0.05 mg/L. The combined TN proxy (NH<sub>3</sub>-N + NO<sub>3</sub>-N) of 0.58 mg/L exceeded the U.S. EPA recommended guideline of 0.30 mg/L [18,23], indicating pre-existing nutrient enrichment in the upstream reach independent of the two modelled point sources.

The two point sources were PS-1, representing TTW Water Treatment Plant discharging 14,000 m<sup>3</sup>/d into Segment 4, and PS-2, representing Rojana Industrial Park WWTP discharging 48,000 m<sup>3</sup>/d into Segment 3. The total effluent volume of 62,000 m<sup>3</sup>/d (0.72 m<sup>3</sup>/s) was about 0.07 % of the total river flow in wet-season conditions. The model output was assessed at Segment 5, which was the same as REO-6 monitoring station CH20, the designated compliance point.

### 3.2.2 Scenario Definitions

Two scenarios were compared. Scenario A (Baseline) represented prevailing discharge conditions with no trading policy in effect, each point source operating at its technology-based effluent limit (TBEL) without additional on-site treatment or credit purchases. Scenario B (WQT policy) was based on the implementation of the proposed framework, where each facility reduced its discharge by approximately 20% over the TBEL through on-site treatment, and the remaining shortfall relative to the water quality-based effluent limit (WQBEL: TP ≤ 1.0 mg/L) was met through the purchase of verified agricultural credits. Trading ratios used in Scenario B included a delivery ratio (DR) of 0.70 for PS-2, an uncertainty ratio (UR) of 1.5, and a retirement ratio (RR) of 2:1. Compliance status was determined using the expression: Net Obligation = Actual Discharge – WQBEL – Credits Applied ≤ 0.

### 3.2.3 Simulated Concentration Output

Simulated nutrient concentrations at the compliance point under both scenarios are presented in Table 1. Scenario A produced TN and TP concentrations of 0.5842 mg/L and 0.0510 mg/L, respectively. Scenario B yielded 0.5816 mg/L (TN) and 0.0504 mg/L (TP), representing absolute reductions of 0.0026 mg/L (–0.4% TN) and 0.0006 mg/L (–1.2% TP).

Table 7: WASP8-simulated nutrient concentrations at the compliance point (Segment 5, REO-6 station CH20) under Scenario A (Baseline) and Scenario B (WQT Policy).

Parameter	Scenario (Baseline)	Scenario B (WQT Policy)
TN (mg/L)	0.5842	0.5816
TP (mg/L)	0.0510	0.0504
Δ TN	—	-0.0026 mg/L (-0.4%)
Δ TP	—	-0.0006 mg/L (-1.2%)

The modest magnitude of the between-scenario concentration difference is attributable to the extreme dilution ratio characteristic of wet-season flow. The combined point-source discharge of 0.72 m<sup>3</sup>/s constitutes approximately 0.07% of total river discharge, resulting in rapid attenuation of the effluent-associated nutrient signal within the first downstream segments. This outcome reflects a physical constraint of the study season rather than any deficiency of the trading mechanism. Under dry-season conditions, when river flow is reduced to 50–150 m<sup>3</sup>/s, point-source contributions would represent a substantially larger fraction of total flow, and the concentration effect of trading-induced reductions would be correspondingly more pronounced.

### 3.2.4 Pollutant Loading Reduction

Loading-based analysis demonstrated substantial reductions in the mass of pollutant discharged, independent of dilution effects. Daily TP and aggregate TN loads for each point source under both scenarios are presented in Table 2.

Table 8: Daily pollutant loading at each point source and aggregate totals under Scenario A (Baseline) and Scenario B (WQT Policy). TP loads are presented per facility; TN totals are aggregate.

Source	Load A (kg/d)	Load B (kg/d)	Reduction
PS-1 (TTW WWTP)	28	14	-50.0%
PS-2 (Rojana WWTP)	120	48	-60.0%
<b>Total TP</b>	<b>148</b>	<b>62</b>	<b>-58.1%</b>
<b>Total TN</b>	<b>618</b>	<b>258</b>	<b>-58.3%</b>

Under Scenario B, PS-1 reduced its TP discharge from 28 to 14 kg/d (-50.0%), while PS-2 reduced from 120 to 48 kg/d (-60.0%). In aggregate, total daily TP loading fell from 148 to 62 kg/d (-58.1%), and total TN loading from 618 to 258 kg/d (-58.3%). These reductions represent the mass of nutrients prevented from entering the Chao Phraya River on each operating day.

It is emphasised that only two point sources were included in the present simulation under wet-season conditions, producing a conservative lower-bound estimate of programme-wide impact. Extension of the model to include additional permitted industrial and municipal dischargers within the sub-watershed, combined with dry-season simulations, would be expected to yield both larger aggregate loading reductions and more detectable ambient concentration effects at the compliance point. Notwithstanding these constraints, the 58% loading reduction achieved with only two facilities illustrates the scalability of the WQT mechanism and its potential to deliver meaningful nutrient abatement across the broader watershed as programme participation expands.

### 3.3 Platform Results

The BPI-NCE (Bang Pa-in Nutrient Credit Exchange) platform is designed as a web-based system to operationalise the proposed WQT framework within the Bang Pa-in sub-watershed. The platform adopts a role-separated, three-portal architecture, in which each portal serves a distinct class of stakeholder under the regulatory oversight of the Pollution Control Department (PCD), Regional Office 13. Two pollutants are tracked concurrently: Total Nitrogen (TN), subject to annual reconciliation, and Total Phosphorus (TP), subject to monthly reconciliation. The three portals and their respective roles are summarised in Table 3.

Table 9: Summary of BPI-NCE portal roles and primary functions.

Portal	User Role	Primary Functions
Authority	PCD Officers	Approve BMPs, issue credit certificates, approve purchases, run compliance reconciliation
Buyer	PS Facilities	Browse credit market, submit purchase requests, file discharge monitoring reports, view compliance status
Seller	NPS Farms	Register BMP applications, track inspection status, view issued credit certificates and earnings

#### 3.2.1 Authority Portal

The Authority Portal is designed for exclusive access by PCD officers and serves as the regulatory control centre for the trading programme. The Programme Overview dashboard is intended to provide real-time system-wide metrics, including the number of registered industrial buyers and farm sellers, the volume of net credits currently listed on the market, and any purchase requests awaiting officer review. A time-series chart of monthly average total nitrogen concentration at an ambient monitoring station is embedded in the dashboard, enabling officers to track ambient water quality trends alongside programme activity. A chronological audit log records all system events with actor identity and timestamp, supporting full regulatory traceability.

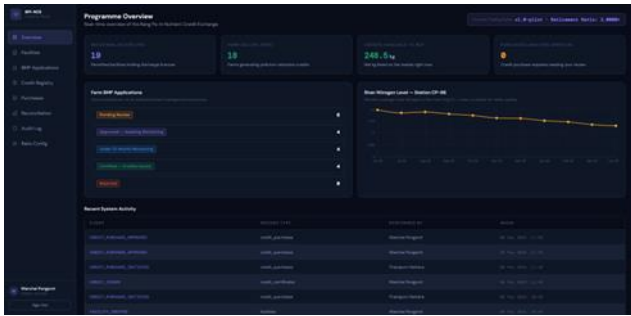


Figure 10: Authority Portal — Programme Overview dashboard showing system-wide metrics, BMP pipeline status, river nitrogen trend, and recent audit activity.

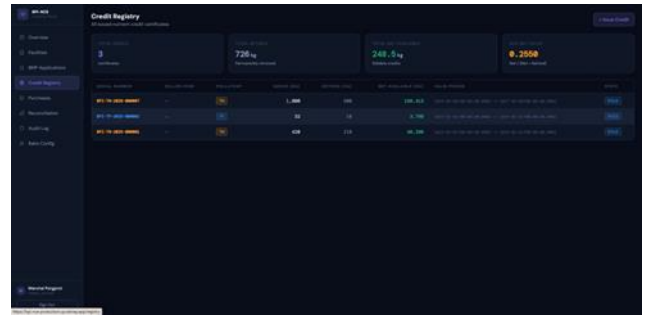


Figure 12: Authority Portal — Credit Registry showing issued certificates, gross and net volumes, retirement amounts, and overall programme totals.

The BMP Applications module is designed to enable officers to review all submitted applications through a filterable list displaying seller identity, BMP type, parcel area, estimated TN and TP reductions, submission date, monitoring lock status, and current state. Available states include PENDING, APPROVED, MONITORING, CERTIFIED, and REJECTED. Officers are able to approve pending applications, initiating a 12-month monitoring lock period, or to certify applications for which the monitoring period has been completed and field inspection passed.

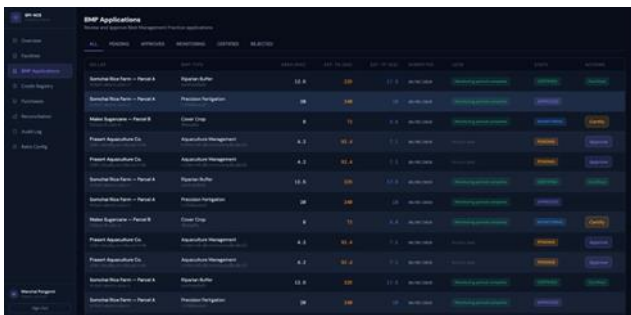


Figure 11: Authority Portal — BMP Applications list with filterable states, showing example applications from Somchai Rice Farm, Malee Sugarcane, and Prasert Aquaculture.

The Credit Registry module is designed to display all issued credit certificates with their gross reduction, retired volume, net available credits, valid period, and current state. Credit issuance is performed through an “Issue Credit” modal in which the officer selects a certified BMP application, specifies the pollutant type and gross reduction volume, and enters the applicable trading ratios (DR, LR, ER, UR). The retirement ratio is automatically populated from the active trading ratio version. A live calculation panel displays the resulting net credit volume in real time before the certificate is finalised.

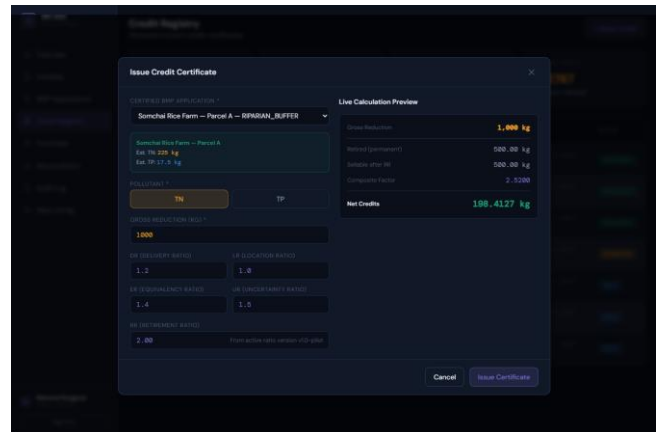


Figure 13: Authority Portal — Issue Credit Certificate modal with live net credit calculation based on user-entered trading ratios.

The Purchase Approvals module is designed to list all pending buyer purchase requests for officer review, displaying the buyer facility, certificate reference, pollutant type, quantity, agreed price per kilogram, total cost, and current status. Officers may approve or reject each request, with approved transactions triggering automatic settlement and state transitions in both the certificate and purchase records.

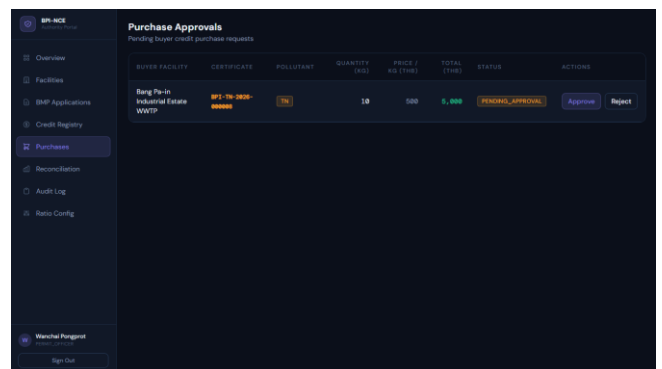


Figure 14: Authority Portal — Purchase Approvals page listing pending buyer credit purchase requests awaiting PCD review.

The Compliance Check Tool (Reconciliation module) is designed to allow officers to enter a facility’s measured discharge data and credits applied in order to compute and formally record its compliance status. The tool evaluates the net obligation as: Actual Discharge – WQBEL – Credits Applied. A result of zero or below yields a COMPLIANT determination; a positive result within the MCL yields DEFICIENT; and any discharge exceeding the MCL yields MCL\_VIOLATION regardless of credits applied. Past compliance records for each facility are displayed alongside the input form to support longitudinal monitoring.

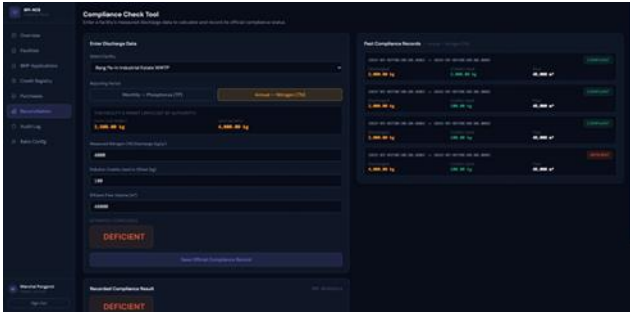


Figure 15: Authority Portal — Compliance Check Tool showing discharge data entry, live compliance estimation, and historical compliance records for a selected facility.

### 3.2.2 Buyer Portal

The Buyer Portal is designed to provide industrial point-source facilities with the tools necessary to monitor their compliance position, procure nutrient credits, and file discharge monitoring reports. The Compliance dashboard is intended to present the facility’s current TN (annual) and TP (monthly) reconciliation status, together with a Compliance Simulator. The simulator allows buyers to interactively adjust their projected discharge volume and credit purchase quantity to preview the resulting compliance determination prior to committing to any transaction, thereby reducing the information asymmetry inherent in credit-based compliance strategies.

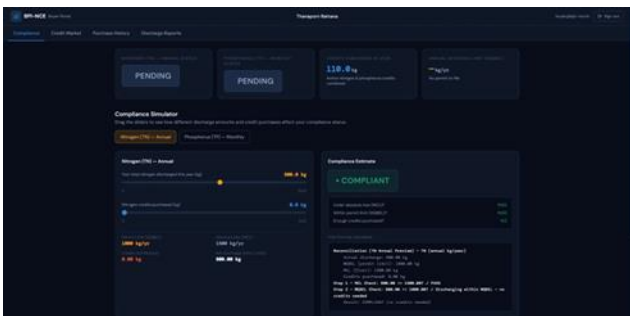


Figure 16: Buyer Portal — Compliance dashboard with current reconciliation status and interactive Compliance Simulator showing live net obligation calculation.

The Credit Market page is designed to list all PCD-verified certificates currently available for purchase, with filter controls for pollutant type (TN or TP). Each listing displays the certificate identifier, source zone, available quantity (kg), validity date, and status. This page enables buyers to identify credits suitable for their compliance period and pollutant requirements before initiating a purchase request.

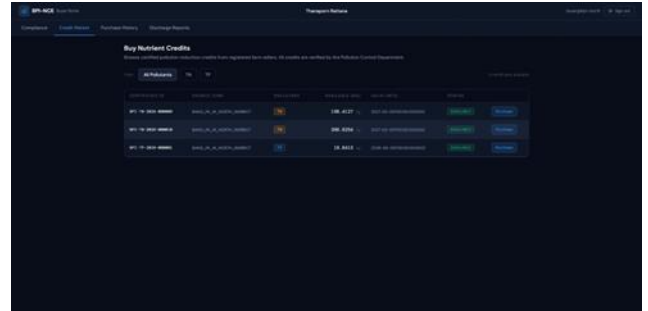


Figure 17: Buyer Portal — Credit Market listing available nutrient credit certificates verified by the Pollution Control Department.

Purchase requests are submitted through a modal dialogue in which the buyer specifies the quantity to purchase (kg), the agreed price per kilogram (within the permitted range), and the target compliance period. Total cost is calculated automatically. Submitted request will be routed to the Authority Portal for PCD approval before settlement. Credits are reserved immediately upon submission, but not transferred until approval is granted.

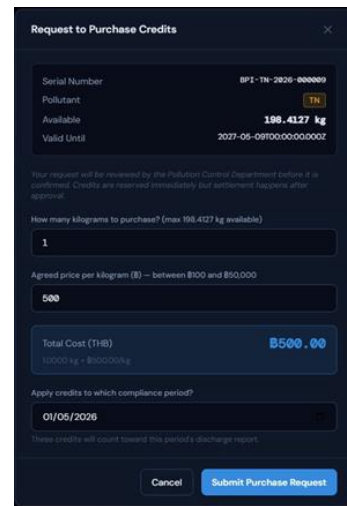


Figure 18: Buyer Portal — Request to Purchase Credits modal showing quantity, price, total cost, and compliance period selection.

The Purchase History page is designed to present all credit transactions made by the facility, grouped by compliance period, showing certificate reference, pollutant type, quantity, price per kilogram, total paid, and settlement status. Settled transactions include a downloadable invoice, while pending transactions display their current approval status. This view is intended to support compliance record-keeping and internal audit by facilities.

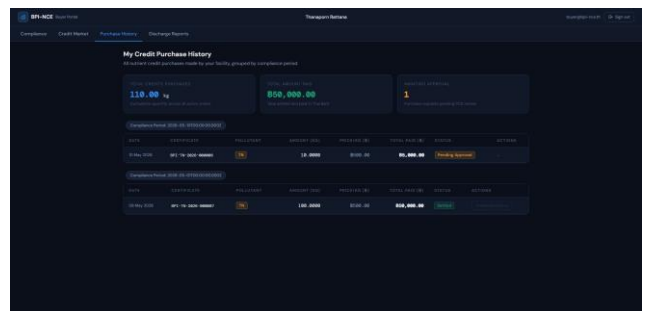


Figure 19: Buyer Portal — Purchase History showing settled and pending credit transactions grouped by compliance period.

Discharge Monitoring Reports are filed through the Discharge Reports page, which provides separate forms for the monthly TP report and the annual TN report. Each form accepts the reconciliation period end date, actual pollutant discharge (kg), effluent flow volume (m<sup>3</sup>), and credits applied (kg). A live reconciliation preview is displayed as data are entered, allowing buyers to verify their net obligation before formal submission. Submitted reports are stored and made available for review through the Authority Portal’s reconciliation workflow.

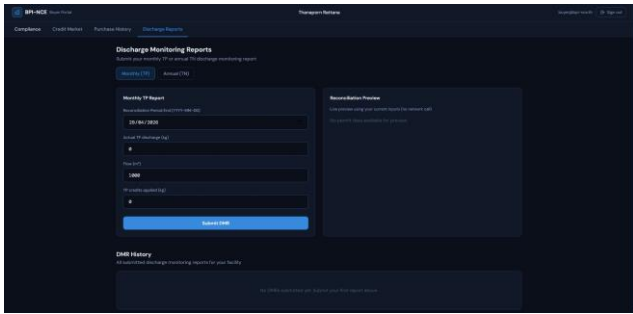


Figure 20: Buyer Portal — Discharge Monitoring Reports page with monthly TP report form and live reconciliation preview.

### 3.2.3 Seller Portal

The Seller Portal is designed to enable agricultural non-point-source participants to register BMP projects, monitor their progress through the inspection and certification pipeline, and track their issued credit certificates and earnings. The Dashboard is intended to provide an overview of the seller’s current credit balance, active BMP projects with monitoring progress indicators, and earnings awaiting settlement, offering a single-screen summary of the seller’s position within the programme.

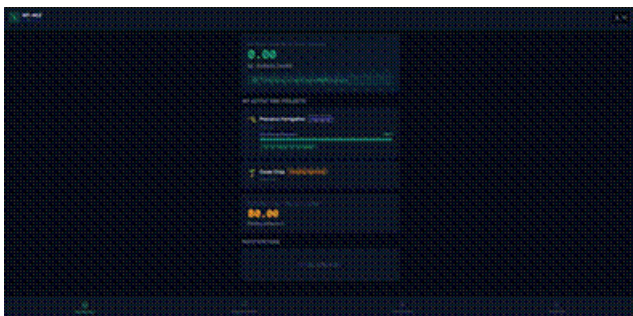


Figure 21: Seller Portal — Dashboard showing available credit balance, active BMP projects with monitoring progress, and earnings awaiting settlement.

BMP applications are submitted through the Register BMP page, on which the seller selects the practice type (Riparian Buffer, Precision Fertigation, Cover Crop, or Aquaculture Management) and enters the parcel area in rai. Upon submission, a confirmation screen displays the estimated TN and TP reductions associated with the registered parcel and informs the seller that a PCD field inspection will be scheduled within 30 business days. The page also displays the full list of the seller’s existing applications with their current states (PENDING APPROVAL, APPROVED, MONITORING, or CERTIFIED), enabling sellers to track the lifecycle of each registered practice.

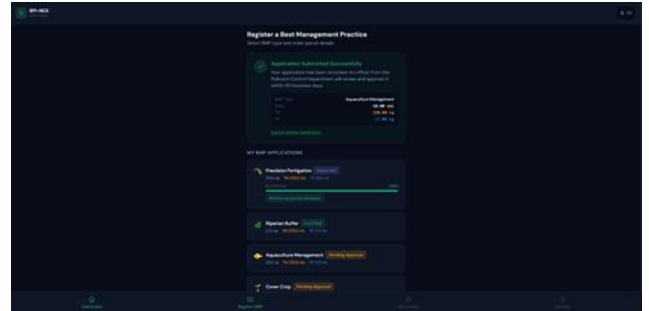


Figure 22: Seller Portal — Register BMP page showing successful submission confirmation and the seller’s existing application list with current states.

The My Credits page is designed to display all issued credit certificates held by the seller, including the gross reduction, the portion permanently retired under the active retirement ratio, and the net volume available for sale. Each certificate entry provides an expandable disclosure explaining why a portion of the gross reduction is retired rather than sold, ensuring transparency for sellers regarding how the retirement ratio affects their sellable volume. The Earnings page tracks income from completed credit sales, displaying total earned and amounts pending settlement, and is restricted to income from credit sales only as seller accounts are not permitted to purchase credits.

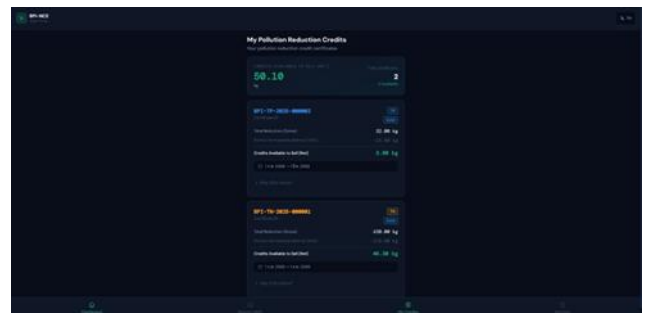


Figure 23: Seller Portal — My Pollution Reduction Credits page showing issued certificates with gross reduction, retirement deduction, and net sellable volume.

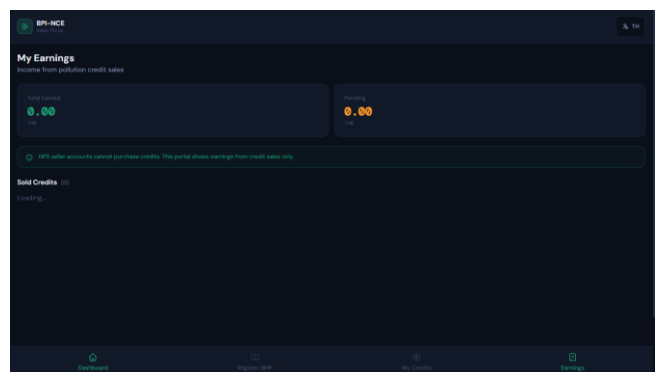


Figure 24: Seller Portal — Earnings page tracking income from credit sales with total earned and pending settlement amounts.

## 4 Conclusion

This project is about creating a Water Quality Trading model for Thailand. It is focused on making policies creating tools and building digital infrastructure for Water Quality Trading. This includes making a policy framework and a prototype app for Water Quality Trading to monitor, calculate and manage water pollution credits. The goal of Water Quality Trading is to build a system that connects people and organizations across a watershed.

The project also made a sandbox simulation using the Water Quality Analysis Simulation Program. This simulation shows how a Water Quality Trading system could work in watershed conditions. The sandbox is not for use now but it helps demonstrate how Water Quality Trading works. It shows what happens when pollutants are released how water quality changes and how Water Quality Trading can reduce pollution and improve management.

The study found that combining water quality science with policies market-based tools and digital technology can be a way to manage water pollution than traditional methods. This is especially important in watersheds with pollution sources like industrial zones, urban areas and farms. A Water Quality Trading system can give people a reason to reduce pollution, which helps them meet environmental goals at a lower cost.

However there are challenges that need to be addressed before a Water Quality Trading system can be used in Thailand. These challenges include gaps in water quality monitoring data uncertainties in how water pollution credits are calculated, legal and regulatory issues, governance structures and involvement from stakeholders. Because of this the sandbox and framework created in this project should be seen as a starting point for research and testing in the Thai context.

Overall this project is not about simulating water quality it is about creating a new Water Quality Trading model for managing water pollution. This Water Quality Trading model combines science, policy, technology and economic incentives into one system. It lays groundwork for sustainable watershed management and future environmental advancements in Thailand. The Water Quality Trading model is a way to manage water pollution and it has the potential to make a big difference, in Thailand.

## Acknowledgements

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# Microbial Contamination of Reusable Water Bottles Among Students of Different Age Groups

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## Abstract

Reusable water bottles have become increasingly common in schools due to growing environmental awareness and the availability of water refill stations. They support daily hydration and reduce plastic waste, making them an environmentally friendly alternative to single-use bottles. However, their hygiene is often overlooked. If not cleaned regularly, reusable bottles can accumulate bacteria and other microorganisms that enter through the mouth, hands or air and multiply in warm, moist conditions. Our research study examines how students' cleaning habits influence the hygiene of their reusable water bottles. The research question asks whether bottles that are cleaned regularly contain fewer microorganisms than those cleaned rarely or not at all. The hypothesis is that more frequently cleaned bottles will show lower microbial contamination. To test this, microbiological analysis will be conducted on bottles used by students. Samples will be collected from the inner surfaces using sterile swabs and cultured on agar plates to measure microbial growth. The contamination levels of regularly cleaned bottles will be compared with those cleaned less often. The results are expected to demonstrate the importance of proper bottle hygiene and encourage students to clean their reusable bottles more consistently to reduce potential health risks.

## Keywords

Reusable water bottles, microbial contamination, hygiene practices, cleaning frequency, student behavior,

## Introduction

### 1. Microbial contamination and types of microorganisms

Microbial contamination refers to the presence and growth of microorganisms in reusable water bottles. It is described as widespread across all student age groups, from preschool to university [13]. Reusable water bottles can contain microorganisms because they provide suitable conditions for microbial growth. Water bottles "provide optimal growth conditions for microorganisms" and can become contaminated through contact with hands, surfaces, and the user's mouth [12]. Common microorganisms found in reusable water bottles include coliform bacteria,

heterotrophic bacteria, *E. coli*, *Pseudomonas*, *Staphylococcus*, *Salmonella*, *Shigella*, and *Streptococcus* [13], [14]. Biological contaminants found in water bottles include "bacteria, virus, protozoa, yeast, fungus, and their toxins," which can cause waterborne diseases such as gastroenteritis [9]. Studies have shown that reusable bottles contain coliform bacteria, fecal coliforms, and heterotrophic bacteria, indicating contamination levels that exceed drinking water guidelines in many cases [1]. Some studies also identified antimicrobial-resistant bacteria such as *Klebsiella grimontii* and oral microbiota transferred during drinking [13]. These microorganisms can originate from the mouth, hands, water sources, or the surrounding environment [14].

### 2. Factors influencing contamination levels hygiene practices

Cleaning behavior varies widely among students. Some clean their bottles daily, while others refill the same bottle for months without washing [1]. University students were reported to clean bottles infrequently and incorrectly [13]. Poor hygiene practices are a major factor influencing contamination. Reusable bottles "pose hazards, such as disease-causing organisms, associated with poor water bottle hygiene practices" [5]. For example, some students "continually refilled the same water bottle for months without washing it" [1]. There is also no clear guideline for cleaning frequency, and improper cleaning increases contamination risk [5]. However, some studies found no statistically significant relationship between cleaning frequency and bacterial levels [6].

### 3. Bottle material and design

Bottle material and design influence contamination levels. Stainless steel may have antimicrobial properties [6], while plastic bottles and complex designs are harder to clean [15]. PET bottles were found to have higher microbial loads (68.8 cfu/ml) compared to stainless steel (35.4 cfu/ml) [9]. Bottle design also affects cleanliness, as "difficulties associated with various bottle designs and materials" can

lead to contamination [10]. Direct mouth-contact bottle designs allow transfer of oral bacteria into the bottle [17], and biofilm formation on materials allows bacteria to persist even after cleaning [20].

#### 4. Cleaning frequency, usage patterns, and source factors

Contamination is influenced by how often bottles are cleaned. Studies show that “the longer water bottles are left unclean, the higher the microbial count” [6]. However, some results showed no statistically significant difference between cleaning frequencies, suggesting contamination may persist regardless of cleaning method [6]. Usage behaviors such as cleaning frequency, cleaning methods, storage conditions, and sharing practices all affect contamination [1]. Other usage behaviors affecting contamination include exposure to air and surfaces, storage at room temperature, and contact with contaminated water sources [6]. Contamination can also result from source water quality and environmental conditions. In younger children, contamination often occurs at home due to caregiver handling and household hygiene [15].

#### 5. Differences across student age groups

There is no clear scientific evidence that microbial contamination differs systematically by student age group [14], and there is limited direct comparison between age groups, but differences in behavior suggest variation in contamination. Indirect comparisons suggest that:

- Preschool and nursery populations often show very high contamination levels, with up to 100% exceeding safety standards [14], [15].
- Elementary school students show moderate contamination levels, with studies indicating 13.3% exceeded total coliform limits and 64.4% exceeded heterotrophic bacteria guidelines [1].
- University students show variable but still significant contamination (e.g., 22%–78% exceeding thresholds) [13], [16].

The only direct comparison found higher bacterial counts in adult bottles (~75,000 CFU/mL) than in children (~34,000 CFU/mL) [8]. Overall, differences across age groups are influenced by confounding factors such as geography, bottle material, hygiene responsibility, and environmental conditions rather than age alone [14].

#### 6. Review of scientific studies

The theoretical understanding is based on 12 studies

conducted between 2002 and 2025 across multiple countries and educational levels [14]. Several studies demonstrate widespread contamination in reusable water bottles. Key findings include:

- Contamination rates ranging from 22% to 100% exceeding safety standards [13], [14].
- A study in Ecuador found 73% of reusable bottle water samples contaminated with coliform bacteria, mainly due to inadequate cleaning [4].
- Extremely high bacterial counts ( $8.03 \times 10^6$  CFU/mL) in some preschool populations [14].
- Rapid microbial growth inside bottles, with bacteria increasing up to 200-fold within 24 hours, reaching 1–2 million CFU/mL within one day [8], [18].
- Research comparing bottle types showed no significant difference between hydration systems, indicating contamination occurs regardless of container type [2].

Most studies examined only one age group, and only one study directly compared different age groups [8]. These findings show that contamination is common across different environments and user groups.

#### 7. Hygiene behavior and student practices

Student behavior strongly influences contamination levels. Research shows:

- Hygiene knowledge does not always translate into proper cleaning practices [5].
- Female students were more likely to clean bottles frequently than male students [5].
- Many students continue unsafe practices despite awareness [7].

In school environments, bottles are exposed to multiple contamination sources, including sharing and contact with surfaces [12].

#### 8. Health implications

Although no study confirmed direct illness caused by bottle contamination, microbial contamination poses health risks. Reusable bottles can contain pathogens that lead to disease, and contamination may result in “water-borne disease such as gastroenteritis” [9]. Pathogens such as *Salmonella* and *Shigella* have been detected in bottles [14], and antimicrobial-resistant bacteria have also been identified [20]. Poor hygiene practices contribute significantly to

disease burden, with inadequate water and sanitation responsible for a large percentage of global illness [4]. There is also evidence linking poor bottle hygiene to diarrhea-related diseases in young children [15]. Younger children may be more vulnerable due to developing immune systems [15], and reusable bottles may also act as carriers of bacteria, increasing the risk of disease transmission [10].

## 9. Settings

This study was designed to investigate the relationship between students' age group and microbial contamination of reusable water bottles. The research was conducted in a school environment using reusable water bottles owned and used by students. The study focused on bottles used during normal school activities, as reusable bottles are frequently refilled, handled, stored at room temperature, and exposed to contamination from the mouth, hands, air, and surrounding surfaces.

The microbiological investigation was carried out using DipSlide TVC with Redspot / Coliforms dipslides. This test type was selected because dipslides are suitable for assessing microbial contamination on both liquids and surfaces, and can be used by dipping into liquid samples, pressing onto surfaces, or applying a swabbed sample onto the agar surface [22], [23]. The TVC side was used to estimate total viable bacterial contamination, while the Redspot / Coliform side was used to assess possible coliform contamination [22].

### 9.1. Sampling

Reusable water bottles were sampled from students from different age groups who used them during the school day. Students were grouped according to age so that microbial contamination could be compared between age categories. The main comparison groups were:

1. younger students, aged 7-10 years old;
2. middle age group students, aged 12-16 years old;
3. older students, aged 17-19 years old].

Sampling was carried out aseptically to reduce the risk of contamination from the researcher, the surrounding environment, or sampling equipment. Each bottle was labelled with a sample code rather than the student's name to maintain anonymity. Information recorded for each sample included the bottle code.

Sample water was collected from each reusable water bottle, as each refill may contaminate water in may also receive microorganisms transferred from the user's mouth or hands. 100 ml of water was taken from each bottle. Dipslide guidance allows samples to be applied by dipping in the water for 10 seconds to have a viable count [24].

## 9.2. Microbial investigation

Microbial contamination was investigated using DipSlide TVC with Redspot / Coliforms dipslides and Plate Count Agar (PCA) plates. Each dipslide was removed carefully from its sterile container by holding only the cap or handle. The agar surfaces were not touched directly, as touching the agar could introduce contamination and affect the reliability of the results [24].

A water sample from each bottle was collected using the sterile container supplied with the dipslide. Both agar surfaces of the dipslide were then exposed to the water sample according to the manufacturer's instructions. The TVC side was inoculated to estimate the overall level of viable bacterial growth. The Redspot / Coliform side was inoculated to investigate possible coliform contamination. After inoculation, each dipslide was returned immediately to its original sterile container and closed securely.

In addition to the dipslide method, Plate Count Agar (PCA), also known as Standard Methods Agar, was used for enumeration of aerobic mesophilic microorganisms. The medium was prepared according to the manufacturer's instructions by suspending the required amount of dehydrated PCA powder in distilled water, heating it until completely dissolved, and sterilizing it by autoclaving at 121 °C for 15 minutes. The medium was then cooled to approximately 45–50 °C before being poured into sterile Petri dishes [28], [29]. Serial dilutions of each water sample were prepared aseptically, and an appropriate dilution was inoculated into sterile Petri dishes. Molten PCA was added, mixed gently by rotating the plates, allowed to solidify, inverted, and incubated.

The inoculated dipslides were incubated upright at 33.4 °C for 46 hours. If manufacturer-specific instructions are followed, dipslides are commonly incubated at approximately 30–35 °C for 24–48 hours to allow bacterial colonies to develop [25], [26]. The same incubation conditions were used for all samples so that microbial growth could be compared fairly between age groups.

For PCA plates, incubation was carried out under aerobic conditions. For aerobic plate count according to the FDA BAM method, plates are incubated at 35 °C for 48 ± 2 hours [30]. For the ISO horizontal pour-plate method, colony counts are determined after aerobic incubation at 30 °C [31]. Standard microbiological methods also describe PCA or Standard Methods Agar as a suitable medium for enumeration of aerobic microorganisms in food, dairy, and environmental samples [32]. In this investigation, the same incubation conditions were applied to all PCA plates to allow comparison between samples.

After incubation, the dipslides and PCA plates were examined for colony growth. The TVC side of the dipslide was used to estimate total viable count, while the Redspot / Coliform side was used to assess the presence or absence of coliform bacteria. Colony density was compared with the manufacturer's reference chart where available, as dipslide results are commonly interpreted semi-quantitatively by

comparing visible colonies with a reference chart to estimate contamination level in CFU/ml or CFU/cm<sup>2</sup> [26], [27]. PCA plates within the countable range were selected, colonies were counted, and the results were expressed as colony-forming units per milliliter (CFU/mL).

Used dipslides, PCA plates, and other contaminated materials were handled as biological waste. After observation, they were disposed of according to laboratory safety procedures. Used dipslides and other microbiological materials should be autoclaved, incinerated, or disinfected using an appropriate bleach solution before disposal, depending on local laboratory rules [25].

## 10. Results

### 10.1. Overview of microbial results

A total of 25 reusable water bottle samples were collected from students aged 7 to 19 years. Each sample was examined using two microbiological methods: **DipSlide TVC** with Redspot / Coliforms and Plate Count Agar (PCA) Petri dish counting.

The DipSlide TVC method was used as a semi-quantitative estimate of total viable bacteria, while the Redspot / Coliform side was used to estimate coliform or possible *E. coli* contamination. The PCA Petri dish method was used to obtain a more direct aerobic plate count of bacteria in CFU/ml.

For comparison by age, the samples were grouped as follows:

Table 1. Sample distribution by age group

Age group	Age range	Number of samples
Younger students	7–10 years	9
Middle age group students	12–16 years	10
Older students	17–19 years	6

The results showed that microbial contamination was present in many reusable water bottles, but the level of contamination varied between samples and between age groups.

### 10.2. Total dipslide TVC results

The DipSlide TVC results showed bacterial growth in 17 out of 25 samples, indicating that viable bacteria were detected in most tested water samples. The remaining 8 samples showed no count on the DipSlide TVC side. Detected results ranged from 10<sup>2</sup> to 10<sup>7</sup> CFU/ml, showing clear differences in contamination level between bottles. The highest DipSlide TVC result was found in sample 19, from an 18-year-old student, with a value of 10<sup>7</sup> CFU/ml.

Other high DipSlide TVC results were found in samples 1, 18, and 21, each with 10<sup>6</sup> CFU/ml. These results show that several samples had notably elevated bacterial contamination levels, while others had little or no detectable growth using the DipSlide TVC method. This supports DipSlide TVC as a useful screening tool overall.



Image 1: Growth of bacteria on DipSlide TVC and Redspot / Coliform agar after incubation

Table 2. DipSlide TVC result count

DipSlide TVC result	Number of samples
No count	8
10 <sup>2</sup> CFU/ml	3
10 <sup>3</sup> CFU/ml	2
10 <sup>4</sup> CFU/ml	6
10 <sup>5</sup> CFU/ml	2
10 <sup>6</sup> CFU/ml	3
10 <sup>7</sup> CFU/ml	1

### 10.3. Redspot / Coliform and E. coli results

The Redspot / Coliform side showed detectable coliform or E. coli-type contamination in 12 out of 25 samples. In 13 samples, no coliform or E. coli count was detected.

Table 3. E. coli / coliform result count

E. coli / coliform result	Number of samples
No count	13
10 <sup>1</sup> CFU/ml	1
10 <sup>2</sup> CFU/ml	8
10 <sup>4</sup> CFU/ml	1
10 <sup>6</sup> CFU/ml	2

The highest E. coli / coliform counts were found in sample 19, from an 18-year-old student, and sample 21, from a 19-year-old student. Both samples had an E. coli / coliform count of 10<sup>6</sup> CFU/ml. Another notable result was found in sample 1, from a 7-year-old student, which had an E. coli / coliform count of 10<sup>4</sup> CFU/ml.

Most positive E. coli / coliform samples were low-level positives, mainly 10<sup>2</sup> CFU/ml. However, samples 19 and 21 were much higher and therefore represented the greatest concern for possible hygiene-related contamination.

### 10.4. PCA petri dish results

The PCA Petri dish results showed that many samples contained a high aerobic bacterial load. Unlike the DipSlide method, the PCA method gave more detailed count ranges, such as 18,000–24,000 CFU/ml or 11,000–14,000 CFU/ml. Several samples were recorded as 60,000+ CFU/ml, meaning that the exact count was above the upper recorded range. Therefore, the true total PCA count is likely higher than the calculated value.

Table 4. PCA Petri dish result category count

PCA result category	Number of samples
Below 10,000 CFU/ml	8
10,000–30,000 CFU/ml	8
30,000–60,000 CFU/ml	1
60,000+ CFU/ml	8



Figure 2: PCA agar plate Sample 22 and 23

Using the lower value of 60,000 CFU/ml for samples recorded as **60,000+**, the minimum total PCA count was: Because eight samples were recorded as 60,000+, the real PCA total was probably higher than this value.

Table 5. Samples with PCA count of 60,000+

Sample	Age	DipSlide TVC	E. coli / coliform	PCA Petri dish
5	10	10 <sup>5</sup>	10 <sup>2</sup>	60,000+
6	10	10 <sup>4</sup>	10 <sup>2</sup>	60,000+
19	18	10 <sup>7</sup>	10 <sup>6</sup>	60,000+
20	18	no count	no count	60,000+
21	19	10 <sup>6</sup>	10 <sup>6</sup>	60,000+
23	15	10 <sup>5</sup>	10 <sup>2</sup>	60,000+
24	15	no count	no count	60,000+
25	8	no count	no count	60,000+

### 10.5. Comparison between dipslide and PCA petri dish counts

The DipSlide TVC and PCA Petri dish results did not

always match exactly. This is expected because the two methods measure bacterial contamination in different ways. The DipSlide TVC method gives an estimated contamination level based on visible growth and comparison with a reference chart. It is therefore semi-quantitative. The PCA Petri **dish** method gives a more direct colony count and is usually more sensitive for total aerobic bacteria.

Some samples showed high PCA counts even when the DipSlide TVC result was recorded as “no count.” For example:

These differences may be explained by several factors. PCA plates are designed for the enumeration of aerobic mesophilic microorganisms and may support the growth of a wider range of bacteria. DipSlides, however, are interpreted by visual comparison and may not detect all bacteria equally. Uneven distribution of bacteria in the water sample, differences in inoculation, dilution, incubation, and agar type may also affect the results.

Therefore, PCA results are useful for showing the overall bacterial load, while the DipSlide Redspot / Coliform side is more useful for indicating possible coliform contamination.

## 10.6. Age group comparison

The average microbial contamination level was calculated for each age group. For the calculation, “no count” was treated as zero. For PCA ranges, the middle value was used. For 60,000+, the value 60,000 CFU/ml was used, so PCA averages are minimum estimates.

Microbial contamination differed between age groups. The older students aged 17–19 years had the highest DipSlide TVC results, ranging from no count to  $10^7$  CFU/ml, and the highest E. coli / coliform results, ranging from no count to  $10^6$  CFU/ml. This indicates that the older age group had the highest overall contamination, especially for total viable bacteria and coliform-type bacteria.

The younger students aged 7–10 years also showed microbial contamination. Their DipSlide TVC results reached up to  $10^6$  CFU/ml, and their E. coli / coliform results reached up to  $10^4$  CFU/ml. This shows that contamination was present in younger students’ bottles, although the highest values were lower than those found in the older group.

The middle age group aged 12–16 years generally showed lower DipSlide TVC and E. coli / coliform results compared with the younger and older groups. However, PCA Petri dish counts still reached 60,000+ CFU/ml, showing that general aerobic bacterial contamination was present even when DipSlide or coliform results were lower. Overall, the highest contamination was found in the older age group, but high PCA counts occurred in all age groups. This suggests that age may influence microbial contamination, but other factors such as bottle cleaning

habits, refilling frequency, storage conditions, and contact with hands or mouth may also have affected the results.

## 10.7. Comparison by individual age

When the results were compared by individual age, the highest contamination was found at ages 18 and 19. Age 18 included the highest DipSlide TVC result, reaching  $10^7$  CFU/ml, and also had an E. coli / coliform result reaching  $10^6$  CFU/ml. Age 19 also showed high contamination, with both DipSlide TVC and E. coli / coliform results reaching  $10^6$  CFU/ml.

Lower contamination was observed at ages 12, 13, and 16, where most DipSlide TVC and E. coli / coliform results were low or recorded as no count. This suggests that bottles from these ages generally had lower levels of total viable bacteria and coliform-type bacteria.

However, PCA Petri dish results showed that high general aerobic bacterial contamination was still possible even when DipSlide TVC or E. coli / coliform results were low. This means that some bottles contained bacteria that were detected more clearly by the PCA method than by the DipSlide method.

Overall, the individual age comparison showed that contamination was not evenly distributed across all ages. The strongest contamination was seen in the older students, especially at ages 18–19, but some younger and middle-age samples also showed high PCA counts.

## 10.8. Main findings

The results show that reusable water bottles used by students contained different levels of microbial contamination. The most important findings were:

1. DipSlide TVC growth was detected in 17 out of 25 samples.
2. E. coli / coliform growth was detected in 12 out of 25 samples.
3. PCA Petri dish counts were high in several samples, with 8 samples recorded as 60,000+ CFU/ml.
4. Older students aged 17–19 years had the highest average DipSlide TVC and E. coli / coliform counts.
5. The middle age group generally showed lower DipSlide and E. coli / coliform counts, although some samples still had high PCA counts.
6. PCA and DipSlide results did not always match, because PCA measures total aerobic bacteria more directly, while DipSlide TVC gives an estimated count and the Redspot / Coliform side focuses on coliform-type bacteria.
7. High PCA counts with no E. coli / coliform count suggest that many bacteria present in the bottles were probably non-coliform environmental or general aerobic bacteria.

## 10.9. Discussion

The results support the idea that reusable water bottles can contain microbial contamination after normal school use. The highest contamination was found in the older student group, especially among students aged **18 and 19 years**. This may suggest that bottle hygiene behavior, cleaning frequency, refilling habits, or length of use may influence microbial growth.

However, the results also show that contamination was not only related to age. Some younger and middle-age samples also showed high PCA counts. Therefore, while age group may have some relationship with contamination level, other factors such as cleaning routine, bottle material, bottle design, storage at room temperature, and contact with hands or mouth are also likely to be important.

Overall, combining the DipSlide and PCA Petri dish results gives a broader understanding of the microbial condition of the water bottles. DipSlide results helped identify estimated total viable count and possible coliform contamination, while PCA Petri dishes showed the wider aerobic bacterial load in the samples. Together, the methods showed that several reusable water bottles had considerable microbial contamination and that regular cleaning is important to reduce bacterial growth.

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# The mechanism of dehydration in the mouth, airways and lungs caused by vaping.

Why the vape is killing the water in your airways.

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## Abstract

In this profile research project, we investigated how substances found in vapes, particularly propylene glycol (PG), interact with water and biological environments. The aim is to better understand the potential role these compounds may play in the body's water balance, especially in tissues such as the mouth, throat and lungs. We analyze the physicochemical properties of PG, with a specific focus on its hygroscopic behavior and the possible implications for moisture regulation in biological models. To deepen our perspective, we consulted a pulmonary pathologist and attended a national conference on smoking and vaping in healthcare, integrating current medical and societal insights into our approach. Additionally, we conducted a model experiment in which PG was exposed to various moist materials to study the dynamics of water uptake and release. This approach provides a broader understanding of how components of vape liquid may behave in a humid biological context.

## Keywords

Vape, Propylene-Glycol, Dehydration, Mucous membranes.

## 1 Introduction

### 1.1 Reason for investigation

#### 1.1.1 Why are we participating in Water is Life (W.I.L.)?

Water is Life first came into the picture through our then enthusiastic biology teacher and now mentor: John van Heeswijk. After his stories about the conference in 2024 in Canada (Oak Bay High) and the news that W.I.L. will be held in Vught in 2026 at Maurick College, we were interested; We went to several teachers to ask what they have done with Water Is Life and if they wanted to supervise new profile research groups. That's how we ended up with Mr. Markus. We sat down with him and he explained to us in detail how the profile paper is put together. He has shown us examples. After this we both felt like starting this new adventure. We started researching which water-related topic we wanted to talk about.

#### 1.1.2 Why do we care about this topic?

At the time we started this profile paper, vaping was just new and all the rage. It is often said that vaping is healthier than smoking and it is seen as a good alternative. We don't vape ourselves, because it has such negative consequences. We are curious why it is so bad. This was a reason for us to include it in our profile paper.

We are also both interested in the workings of the human body. Pim wants to study physiotherapy and Jippe wants to study medicine. We thought that many of the profile papers of water is life would be about the water in our living environment and a bit more on a macro level. That is why we wanted to think a little more out of the box and do research into water at the cellular level, because water is really vital there.

#### 1.1.3 How did we arrive at this research question?

We had decided to participate in W.I.L. after which we started brainstorming. We thought of all kinds of different things. During the brainstorming session, we quickly turned to vaping, because we felt that not much was known about this and because the subject interested us. We thought about how to combine vaping with water, but we quickly came to the question: what is the effect of vaping on water in the human body?

## 1.2 Research purpose

### 1.2.1 Purpose of our research.

Our research question is: what is the influence of vaping on the water in the cells of the respiratory tract.

With this research, we also want to show the lesser-exposed side of why vaping is so bad. A lot is already known about the consequences of the high dose of nicotine and other harmful substances in the vapes. But less is known about or the direct effects of vaping on the respiratory mucosa.

## 2 Research

### 2.1 vaping basics

#### 2.1.1 How does a vape work?

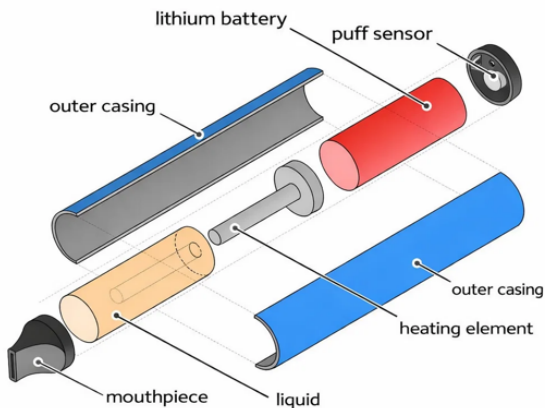


Figure 1: Ingredients of vape

How a vape works:

When you suck on a vape, the heating element activates via the puff sensor. Liquid in the vape evaporates and the vapor ends up in your lungs, which can cause irritation or damage. [1]

The liquid contains several substances, including nicotine. This is a form of addictive nicotine that is 'soft' in the throat and is quickly absorbed by the body. That's how you become addicted unnoticed.

#### 2.1.2 Difference between smoking and vaping.

To start with the more theoretical part of our research, we first need to look at the difference between the traditional cigarette and the more new vape. Both cause dehydration, but in their own way. A cigarette is roughly made up of tobacco and paper. When tobacco is burned, the addictive substance nicotine is released. [2] Other toxins are also released such as lead, cadmium, mercury and tar. [3] The nicotine in a cigarette can cause narrowed blood vessels, [4] so that less saliva is produced in the mucous membranes and this can cause local dehydration. When cigarettes are burned, up to 7000 substances are released. Vapes or e-cigarettes contain a chemical liquid that evaporates when burned. The chemicals contained in the vapes immediately extract moisture from the contact areas such as the mucous membranes in the mouth, throat and lungs. This process is called hygroscopy. [5]

## 2.2 Congress and specialists

### 2.2.1 Congress: (No) Smoking in Healthcare

On 9-10-2025 we went to a conference together about smoking and vaping. This conference was organized by the

organization SMOKE-FREE CARE. We were invited by Dr. Danielle Cohen and went along as her guests. It took place in Amersfoort, where we traveled by train. At the congress we followed several rounds of presentations, a round on Smoking and vaping and a round on Young people and vaping. [6] We also met other students who came from Leiden.

With dr. Cohen, we talked about the consequences of vaping and what she is trying to do to reduce vaping among young people. Because vaping is relatively new, doctors and researchers can still investigate few consequences. This is because there are no "long-term vapers" yet.

Dr. Cohen herself founded [Vapen #jouwkeuze](#) together with a colleague. They provide teaching packages for schools to make young people aware of the fact that they are victims of a dark and cunning industry, the tobacco industry. [7] She also gives presentations herself and she did that at our school as well. From there we came into contact with her.



Figure 2: conference vaping in healthcare.

#### 2.2.2 Questions for Dr. Cohen.

As mentioned earlier, we spoke with pathologist D. Cohen, who specializes in the lungs. In addition, she has made it her task to inform young people who are addicted to smoking and vaping about the harmful effects. Medical students also took part in the conversation.

We let Dr. Cohen read some of our literary research and asked follow-up questions about it. Dr. Cohen said that dehydration only occurs on the surface of the lungs and does not dry out the entire lungs. Macrobiotics, the 'good' bacteria, are affected in your mouth and throat. The mucous membranes are also permanently affected when vaping. She admits that she did not know the mechanism of the sodium channels that are extra active.

We asked the question in which phase we as a society were in the research into vapes. Are we really at the beginning or do we already know a lot about addictive e-cigarettes? Dr. Cohen said that although we are still in the beginning,

we already know enough to know that the vape is extremely harmful. As a doctor, do you continue to spend the money on deeper research, or do you spend the money on prevention and action? Dr. Cohen chose the second option. She says that if you can not solve the problem now, you need to make sure to learn as much as possible about the subject. So you will not be faced by big surprises in a few years. That is why further research is now being done.

When we asked the question whether the dehydration caused by vaping causes permanent damage, Dr. Cohen gave a hopeful answer. The nicotine in vapes and cigarettes pinch the blood vessels, so that less fluid is distributed over your body. As soon as you stop smoking and vaping, they open again in a brief time. This reduces the risk of cardiovascular disease quickly. She also says that the activity of the sodium channels is increased when vaping due to the propylene glycol, so that does not mean that more sodium channels are added and that cell structure is affected. It is therefore likely to recover in the short term. She also indicates that it is difficult for pathologists to see if a cell area has dried out.

Children now regularly come to the hospital with complaints that are due to vaping, according to Dr. Cohen. She does not yet see young people vaping at her work because she often looks at cells from people with visible diseases caused by longer periods of smoking and vaping, such as cancer and COPD. She does indicate that she speaks a lot with pediatricians who do have patients who have complaints from vaping. Asthma is a major problem, but so are mental effects such as concentration problems and sleeping problems. It bothers her that there are no concrete figures on these problems in the Netherlands because this is not yet reported enough. As a result, only the worst symptoms and consequences stand out, and the milder symptoms of vaping quickly fade into the background.

Finally, she gives us the tip not to lose sight of the big picture. It's not just the nicotine or not just the dehydration from the vape, it's the combination that makes the vape so incredibly harmful.

### 2.2.3 Questions for dr. Slaats

We spoke with pediatric pulmonologist M. Slaats. She provided us with some insight into the significance of our research.

We asked her whether she observed a difference between complaints caused by smoking and those caused by vaping. She explained that she does not see children with smoking-related complaints in her department. This is mainly because smoking at a young age has become less popular and more difficult than vaping. According to her, vaping causes dehydration in the airways, partly due to nicotine, which results in a thicker layer of mucus in the respiratory system. She also stated that diseases such as Chronic Obstructive Pulmonary Disease and pulmonary fibrosis can be caused by vaping, although these conditions generally develop later in life. She concluded this topic by

emphasizing that it remains uncertain when these future patients will begin to experience symptoms. She expects that children who start vaping at an early age may already suffer from chronic coughing with thick mucus in early adulthood, which could eventually lead to permanent lung damage. She further stressed that many of the harmful effects of vaping are caused by the combination of various toxic substances in e-cigarettes and the dehydration they cause.

Dr. Slaats stated that the vaping problem is still not being taken seriously enough. She believes that insufficient measures are currently being taken in the Netherlands to reduce and prevent vaping. She also mentioned that schools often deal with the vaping issue too casually. In fact, she described it as "absurd" that so many people dismiss the problem and fail to take responsibility.

We concluded the interview by asking how she views the future. She responded that she hopes the government will soon take stronger action to ban vapes. She also hopes that schools will provide students with greater awareness and understanding of the risks associated with vaping. According to her, the best solution would be to support all addicted young people through addiction treatment programs, ultimately leading to a completely vape- and smoke-free generation.

She ended by remarking that, as doctors, they are always willing to help patients, but rather no complications that could have been avoided.

## 2.3 Personal experiences

### 2.3.1 Help, I suffer from dehydration.

In the next piece, we will discuss experiences of vaping young people on online forums such as reddit. So these are user experiences and we have not been able to verify them.

*'Vaping dehydrates the hell out of me.'* This user indicates that he is very addicted to the e-cigarette and always suffers from a dry mouth and thirst for a while after vaping, even though the user indicates that he drinks a lot of water. A response to this person is given by another user who indicates that he is also bothered by this. This person has completely stopped drinking alcohol and caffeine, but vapes. Even now, this person indicates that they are *'often dehydrated'*. This person even indicates that they once had to go to the hospital because they became unwell after feeling extremely dehydrated. Of course, we cannot determine with certainty that this is due to vaping.

A subsequent user indicates that they do not suffer from dehydration at all when vaping. He/she says that it can feel uncomfortable but cannot lead to extreme dehydration. This user also indicates that it is nonsense to drink an extra 650 to 1000 milliliters of water to prevent dehydration. *'This is utterly bullshit'* the user indicates. According to him/her, drinking 650 to 1000 milliliters is a guideline that is spread among young people, but it is just a fable. The

editor's advice: 'just drink what feels good and a little more when you're thirsty.' [8]

So you can see that one person clearly feels dehydrated, while the other does not have that at all. Is the dehydrated feeling just a coincidence or is it due to vaping?

## 2.4 Pubmed research dehydration

### 2.4.1 Vaping-induced proteolysis causes dehydration of the airway surface.

The following study is one of the effects of PG in the medical literature. Pubmed is a free to visit research website where many medical studies are shared. We have incorporated this research into several parts. Difficult words are printed in italics and will be explained later.

In this study, scientists at the University of North Carolina are investigating whether vaping affects the *epithale sodium channel*. They do this by measuring the *proteage levels* in the. The researchers cultured bronchial cell cultures and exposed them to *bronchoalveolar lavage fluid*. The height of the liquid on the airway surface was also measured. This study was tested on the cultures of non-smokers/vapers (control group), smokers and vapers.

There was increased ENaC activity *in the group of smokers and the group of vapers*. This was not the case with the control group. This means that much more moisture goes into the cell and the surface dries out. The height of the liquid on the surface was also considerably less. The conclusion is therefore that there is clearly dehydration on the surface of the mucous membrane, which in turn can cause further diseases and disorders. [9]

- *Epithale sodium channel (EnaC)*: a gate that regulates the humidity of your mucous membrane cells in your airways.
- *Proteases*: protein-degrading enzyme.
- *Protégé mirror*: the amount you have of the protein-degrading enzyme.
- *bronchoalveolar lavage fluid*: that is fluid from the lungs that has been sucked out by means of a viewing examination.

## 2.5 Propylene-glycol

### 2.5.1 Why is there Propylene-Glycol (PG) in a vape?

PG is added because it is an excellent flavor enhancer. It also promotes the 'hit', the feeling in the throat while vaping. [10]

The review article *A review of constituents identified in e-cigarette liquids and aerosols* (February 2021) shows that in all analyzed studies; propylene glycol was found in the e-cigarettes examined. [11]

### 2.5.2 What is the function of propylene glycol? (PG)

The e-liquids in a vape consist largely of PG. This is a substance that provides smoke-like vapor in the e-cigarette. The chemical formula of this substance is  $C_3H_8O_2$ . The substance belongs to the group of 'dioles', which are alcohols with two OH groups. PG has a freezing point of -50 degrees Celsius (222 degrees

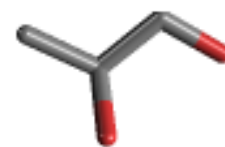


Figure 3: Structural formula PG

Kelvin) which makes the substance useful for antifreeze. It is a color and odorless substance that has consistency of cough syrup. [12] This substance is used in the food industry, among other things, because the handy thing about the substance is that it easily absorbs moisture into the food. For example, it is found in flavorings, candy and soft drinks. [13]

PG is actually a very useful substance. It is biodegradable and is used in many foods because it easily draws out moisture. But that's exactly why it's so harmful in vapes. PG carries the nicotine and flavorings in vapes. The PG is absorbed into the lungs, broken down into lactic acid and pyruvate and eventually excreted through urine. [14] What researchers have recently discovered is that when PG is heated, it can break down into formaldehyde and acetaldehyde, which can cause cancer in the long term.

Why does PG cause dehydration?

As mentioned earlier, the substance easily absorbs liquid. This is called hygroscopic. When you inhale PG with a vape, it comes into direct contact with the outer layer of your airways. The PG attracts fluid in your mucous membranes of your mouth, throat and lungs. The fluid is therefore absorbed by the PG, as it were. The result is that your mucous membranes dry out. The same thing happens deeper in your airways; PG removes fluid in the mucus layer that protects your lungs. The mucus layer thus becomes thinner and less effective. As a result, you can get a dry cough and experience a poor feeling. [15] You can compare it to putting salt on a piece of tomato. The salt pulls the moisture out of the tomato; PG does this in your airways.

### 2.5.3 Why does PG extract moisture?

To find out, we need to delve more into the chemistry of the substance PG. As mentioned earlier, PG is a diol, an

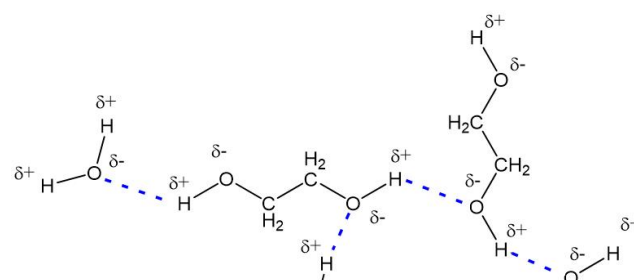
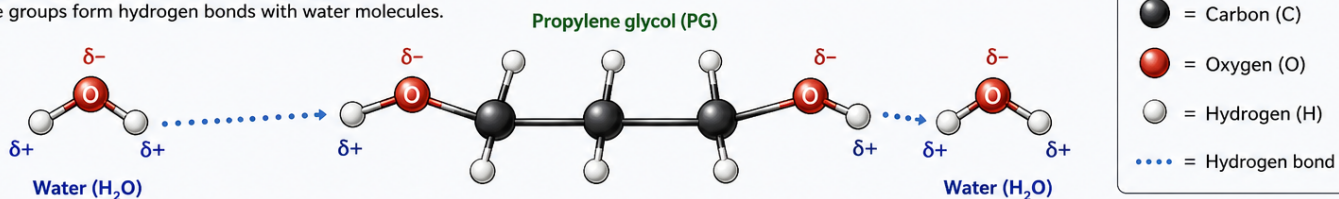


Figure 4: Hydrogen bonds

## 1. HYDROGEN BONDING: PG ATTRACTS WATER

Propylene glycol (PG) has two  $-OH$  groups. These groups form hydrogen bonds with water molecules.



The  $-OH$  groups of PG are polar and form hydrogen bonds (dotted lines) with water molecules. This allows PG to attract and hold water.

## 2. DIFFUSION: WATER MOVES FROM HIGH CONCENTRATION TO PG

Because PG binds water, it creates a lower water concentration. Water moves by diffusion from the moist tissues (high concentration) to the PG (low concentration) until equilibrium is reached.

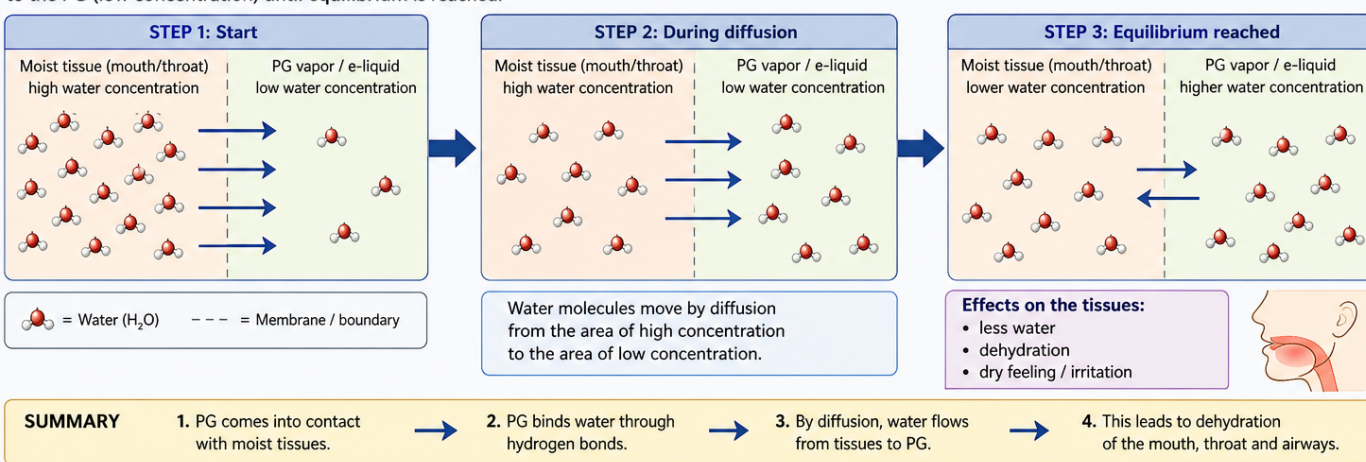


Figure 5: Hydrogen bonds and Diffusion

alcohol with two OH groups. These OH groups make molecule polar and able to form hydrogen bonds. [16] Water molecules attract each other via hydrogen bonds, these bonds are not very strong, but they are important. PG also forms such bonds with water, so that the water adheres to the PG, so the water is retained by the PG. Diffusion also plays a major role in dehydration with PG. There is a relatively large amount of water in the mucous membrane and relatively little in PG. Through diffusion, the water goes from a high concentration to a low one to create an equilibrium. [17]

## 2.6 Consequences on the mucus membranes

### 2.6.1 Consequences of dehydrated mucous membranes.

What are the consequences if your mucous membranes dry out? That is a nice question since this question encompasses the whole idea of water (dehydration) and life.

To answer that question, you first need to know what the function of your mucous membranes is. Your mucous membranes have a protective function. They stop bacteria, viruses and dirt. For example, substances stick to the mucus. Your mucous membranes also contain immune

cells and antibodies that can fight intruders. Feeding into your airways small cilia together with mucus dirt towards the throat, so you can swallow it or spit it out, so it also has a transport function. Finally, your mucous membrane keeps your tissues moist. Mucous membranes produce mucus so that surfaces do not dry out. This helps with breathing, swallowing and digesting food.

Small cracks or irritations can occur in your oral mucosa when it dries out temporarily. As well as a dry mouth/throat and difficulty swallowing and talking. This is because there is less saliva. Prolonged dry mouth can cause more serious problems. Such as gum infections, fungal infections and chronic pain when eating or speaking.

In the airways, the consequences are already a lot greater. The mild symptoms, coughing a lot and a greater risk of infection occur with short-term dehydration of the airways. If we look at the longer term, you will see that the cilia work less well and the dirt is removed less effectively. If your airways are dehydrated for a longer period, you will even see that dirt and thick mucus start to accumulate. This can lead to chronic inflammation, increased risk of bronchitis, permanent damage to cilia and the breakdown

of mucous membrane tissue. These are already serious problems that make your life a lot more difficult.

Your lungs are highly dependent on moist mucous membranes for gas exchange and defense. Temporary dehydration leads to a feeling of tightness and less effective oxygen uptake. Your mucus also thickens because your mucous membranes can no longer regulate the moisture supply properly, so dust particles, bacteria and viruses linger longer. After suffering from dried out mucous membranes in your lungs for a long time, extremely harmful and permanent problems arise. These include chronic bronchitis, pneumonia, damage to alveoli, scarring, reduced lung capacity and a high risk of COPD or emphysema. Long-term inflammation can cause lungs to become stiffer due to fibrosis (scarring), which significantly reduces the uptake of oxygen. This is also irreversible.

For example, we see that dehydration of the mucous membranes is not something to be neglected. Permanent damage to the lungs and permanent irritations in the throat are important consequences to consider.

### 3 Practical research

#### 3.1 Research question and hypothesis.

In this experiment, it is investigated whether PG can absorb moisture from the environment because of its hygroscopic properties. This property is thought to contribute to the drying effect that e-cigarette users experience in the mouth and throat.

The hypothesis is that propylene glycol absorbs water vapour from the air, and that this uptake increases as the relative humidity in the immediate vicinity is higher.

#### 3.2 Purpose of the experiment.

By means of this research we want to show that PG extracts water from a moist environment, such as occurs in the respiratory tract.

#### 3.3 Experimental design.

To investigate the hygroscopic effect of PG, multiple experimental setups were used in which PG was exposed to different humidity levels.

Each setup consisted of a sealed container containing a small dish filled with 6 milliliters of PG. Moist materials were placed around this dish, such as wet paper and pork. The container was covered to prevent direct contact with the outside air, so that only water vapor absorption could take place through the air.

The complete arrangement is shown in the figure below.



Figure 6: experimental setup



Figure 7: experimental setup

#### 3.4 Setup with PG and moist materials.

For this experiment, we created three different setups:

1. Control setup: an empty container with a dish containing 6 milliliters PG. Covered with plastic wrap and a lid.
2. Container with damp kitchen paper on both sides of the dish with 6 milliliters PG. Covered with plastic wrap and a lid.
3. Container with pork tenderloin cut into small pieces around the dish with 6 milliliters PG. Covered with plastic wrap and a lid.

Pork was used as a model for biological tissue due to its relatively high-water content and structural similarity to human tissue. [18]

The control group was used to see if the PG does not extract moisture from the air present.

In the experiment, we made sure that there is no direct contact between the moist substance and the PG. So that the moisture is absorbed by the PG through the air. A process that is similar to the vapor that comes out of a vape.

### 3.5 Measurement procedure.

For each setup, the initial mass of the dish was determined with PG using a scale that weighed the grams to 2 decimal places. After a predetermined exposure time, the mass was measured again.

The mass increase was interpreted as the amount of absorbed water vapor.

The experiment was conducted several times to test the reproducibility of the results.

We pay close attention to safety and use lab coats, glasses, face masks and gloves.

### 3.6 Results

In the first experiment, with an exposure time of 2 hours, only a limited mass increase was observed. However, due to leakage in the control group, these results could not be reliably used for comparison.

#### 3.6.1 Mass Increase in Propylene Glycol After 24 Hours of Exposure (Experiment #2)

In the second experiment, the exposure time was extended to about 24 hours. A clear increase in the mass of the propylene glycol was observed in all setups.

The results of this experiment are shown in Figure 8. In this group, setup 1 is the control group, setup 2 with wet paper and 3 with pork.

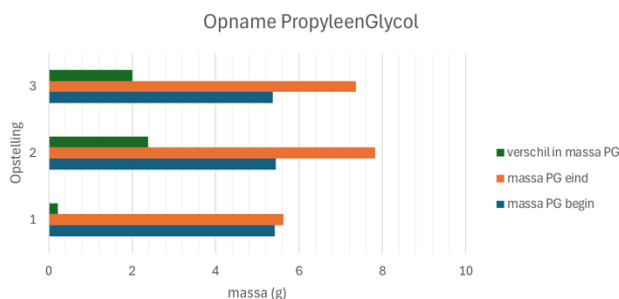


Figure 8: results experiment 2

As can be seen in Figure 8, the mass increase in the setups with moist materials is significantly larger than in the control group. The increase in wet paper and pork is in the order of magnitude of a factor of 10 compared to the control group.

#### 3.6.2 Mass Increase in Propylene Glycol After 24 Hours of Exposure (Experiment #3)

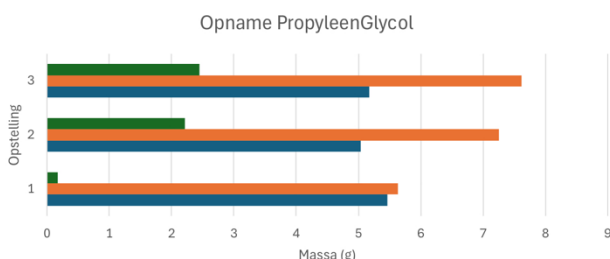


Figure 9: results experiment 3

A repeat of the experiment (experiment #4) yielded similar results, as shown in Figure 9. As before, setup 1 is the control group, setup 2 with wet paper and setup 3 with pork.

### 3.7 Discussion

This measurement also shows that the presence of moist materials leads to a greatly increased mass increase in PG. Although there are slight differences between the materials, the overall trend remains consistent.

During the study, some experimental limitations occurred. In the first practical, the control group turned out to be leaky, so this measurement was not usable for analysis. In a fourth version of the experiment, two setups were lost due to a fall, so no data could be collected.

In addition, the quantities and surfaces of the moist materials used are not standardized. This may have led to variations in the evaporation rate and thus in the measured mass increase.

Small differences in exposure time (just below or above 24 hours) may also have affected the results.

Nevertheless, the results have been shown to be reproducible in two independent experiments (experiment #2 and #3). This strengthens the reliability of the conclusion that PG absorbs more moisture in an environment with higher humidity.

A limitation of the model used is that it is a simplified representation of the human situation. In the body, in addition to hygroscopic, physiological processes such as blood flow, mucus production and active fluid regulation also play a role.



Figure 10: Setup 2 with wet paper.

### 3.8 Research conclusion

From this experiment, it can be concluded that PG is able to absorb water vapor from the environment. This hygroscopic effect depends on the ambient humidity: in a humid environment, the mass increase is significantly greater.

The results thus support the hypothesis that PG can extract moisture from the immediate environment.

Combined with existing literature, this suggests that PG may contribute to dehydrating effects on the mucous membranes in the respiratory tract when using e-cigarettes. However, further studies are needed to fully confirm this effect in a biological context.

### 4 Overall conclusion

In this profile paper, we have investigated in a well-researched way how PG, one of the main components of the e-liquids in vapes, extracts moisture from the mucous membranes of the respiratory tract. We approached our research question, what is the influence of vaping on the water in the cells of the respiratory tract, from several angles: theoretically, experimentally and through experts.

The literature is well explained how PG as a hygroscopic substance forms hydrogen bonds with water molecules and extracts moisture from the mucous membranes in the respiratory tract via diffusion. This is complemented by a study from North-Carolina showing that vaping leads to increased ENaC activity in the respiratory tract, which dries out the mucosal surface.

The experiment is a simplified but reliable study, in our opinion. By exposing PG to moist materials, such as wet paper and pork, in sealed containers, we showed that PG absorbs significantly more moisture in a humid environment than in normal air, even by a factor of ten times greater. We repeated the experiments, which increases the reproducibility a little bit. Limitations must be taken into account, such as the non-standardised quantities of material and the simplified representation of physiological reality.

The conversation with pulmonary pathologist Dr. Cohen adds a lot of value. She confirms that the dehydration takes place on the mucous membrane surface, that the sodium channels are likely to recover when you stop vaping, but the combination of all the harmful substances in the vape is what makes the vape so dangerous.

All in all, we are satisfied with this profile paper and the results that have come out. We hope to eventually be able to convey a message about the serious physical consequences of vaping. In answer to our research question: PG in vapes actively dries out the mucous membranes of the respiratory tract. As a result, irritation of the airways that can lead to pneumonia, COPD, bronchitis

and pulmonary fibrosis. Which leads to a lower quality of life.

### Acknowledgements

We would like to begin by thanking our mentor and initiator, John van Heeswijk. Thanks to his enthusiasm, we were able to achieve a great deal. We noticed how much energy and effort he puts into the entire Water is Life week, and it means a lot to us to see how proud he is of it.

Secondly, we would like to thank our supervisor, Patrick Markus. From the very beginning, he believed in what we were doing and guided us in the right direction through regular check-ins. During every Dalton hour we spent with him, he always made time for us without hesitation, which we appreciated. We would also like to thank Mr. Markus for his clear explanations about the conference itself. We were always welcome to ask questions and consistently received the encouragement we needed. We hope he can look back on our collaboration as a successful one, just as we do.

Furthermore, we are highly appreciative of everyone who helped us get into contact with specialists. This helped us a lot.

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At the beginning of our project, we received a strong start through our conversation with Dr. Cohen. With her help, we gained a broad perspective on vaping in general. She also helped us formulate an appropriate and well-structured research question. We wish her the best of luck with her work in raising awareness among young people about the consequences of vaping.

The help of the educational support staff was very welcome. Helping us gather the items we needed for the experiment and standing by if needed.

Lastly, we would like to thank Dr. Slaats for helping us through the final stages of our project. With her extensive expertise as a pulmonologist, she helped us clarify our complex research and gave it significant meaning.

Working on this profile paper was sometimes challenging, but we learned a great deal and genuinely enjoyed the entire process.



Figure 11: Pim (left) & Jippe (right)

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# Awareness of Estonian Companies Using Marine Raw Materials Regarding Environmental Impact Assessment and Data Availability

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## Abstract

The European Commission is increasingly focusing on the sustainability of production, creating a need to assess the environmental impact of companies and their products. However, current environmental assessment methods often lack detailed parameters for products made from marine raw materials such as seaweed. Although these methods are being developed further, it remains unclear whether companies will be able to meet future requirements.

As part of the AlgaeProBanos project, this study explored the readiness of Estonian companies using marine raw materials to conduct environmental assessments. A survey was carried out among nine companies. The questionnaire examined the origin of their raw materials, their awareness and use of environmental assessment methods, and their ability to provide the data required for such assessments. Respondents were also asked which types of assessment questions they would be able to answer and what support they would need to obtain the necessary data in the future.

The results showed that only a few companies hold environmental certifications and many highlighted the need for improved communication and cooperation between businesses, government institutions and scientists.

## Keywords

environmental awareness, environmental footprint, life cycle assessment, environmental product declaration, data availability

## Introduction

The European Commission is working to ensure sustainable production, which includes assessing the environmental impact of companies. Various assessment methods are in use, each with different criteria that are either recommended or that companies are required to follow before marketing their products. For companies that use marine-based products, the criteria in current assessment methods are insufficient, and efforts are being made to improve this.

The research is being conducted as part of the European Commission's project "Monitoring and Evaluation Sustainability Assessment Framework of Micro- and Macroalgal Value Chains", AlgaeProBanos (APB). AlgaeProBanos brings together 26 experts and industry partners from the Baltic and North Sea regions to accelerate product development and market access for sustainable algae products. One of the project's objectives is to develop a science-based life cycle assessment (LCA) method for products made from raw materials sourced from aquatic environments, grounded in quantitative data and metrics, as well as an Environmental Product Declaration (EPD) for these products. [1]

Unlike the traditional view, according to which the three pillars of sustainability – Environmental, Social, and Governance (ESG) [2] – are separate entities, a sustainable vision should view the economy as part of a larger ecological system. In the environmental sphere, ecological and biological parameters are measured, such as biodiversity, the use of natural resources, and positive and negative impacts on the natural environment. [1]

The aim of my work is to find answers to the following questions with the help of the businesses participating in the pilot project:

- a) What environmental labels and environmental indicators are companies aware of?
- b) What environmental impact assessment indicators do companies already use?
- c) What data necessary for obtaining indicators is available to companies?
- d) What positive effects do the companies' activities have on the environment?
- e) What are the companies' recommendations for the assessment method?

During my research I surveyed nine Estonian companies that use sea mud, seaweed, reeds, or shells in their products.

In this study, I will examine the companies' views on environmental assessment and the collection of data required for it. I used Google Forms to conduct the survey.

## 1. Overview of the Marine Environment and Economy

### Estonia's Blue Economy

The original meaning of the blue economy was an economy that restores the environment [3]. Blue economy encompasses all economic sectors related to the oceans and seas. A major challenge today is to increase the sustainability and resilience of the blue economy. The Organisation for Economic Co-operation and Development (OECD) calls on countries to make decisions and take action towards this goal at both local and international levels. [4]

The goal of the Estonian Maritime Cluster (EMK) is to develop and grow Estonia's blue economy sector based on sustainable plans. The EMK's key development priorities are sustainability, digitalization, and international competitiveness. The EMK network consists of the largest Estonian organizations, companies, and professionals involved in the blue economy, whose goal is to increase the share of the maritime economy in Estonia's GDP (gross domestic product). [5]

### Marine Resources and Their Use

When thinking about marine resources, most people think of fish. In addition to fish, the sea provides a great many minerals, such as manganese nodules (which usually also contain cobalt, copper, nickel, and iron), which cover a large part of the ocean floor; sand and gravel [6]. These minerals are essential for industry and construction. A significant portion of marine resources also consists of crude oil and natural gas, which are used in the transportation sector and for generating electricity. Many countries that possess these fossil resources, such as Norway, are working toward more sustainable production where crude oil and natural gas accounted for 40% of exports in 2015 [7].

In Estonia the use of sea mud is widespread. Sea mud is used as therapeutic mud in various spas and resorts [8], as well as in cosmetic products [9]. Clay, including sea clay, is used as a building material and can be used to create art and ceramics. Another widespread use of clay is in cosmetics. Many toothpastes contain clay, and clay masks are used in skincare. Clay is also used in soaps, powders, exfoliating creams, and wraps. Different types of clay are used for different purposes. [10]

Around the world, particularly in Asian countries, the cultivation of macroalgae is a significant industry. Conditions in the Baltic Sea vary depending on the season, and the low salinity of the Baltic Sea water limits the

potential for cultivating these species in Estonia. However, cultivating macroalgae is possible and would help combat eutrophication (the excessive accumulation of nutrients in seawater) and expand habitats for fish and invertebrates. There is high demand for algae, as they are used in food, cosmetics, and fertilizers, as well as for producing biofuels. [11]

Edible mussels are farmed in the western part of the Baltic Sea and the North Sea, and they can also be farmed in Estonian waters. Edible mussels are a good source of protein and other nutrients and farming them has a lower environmental impact on the sea than the fishing industry. In the lower-salinity areas of the Baltic Sea, mussels do not grow as large, but they can still be used for the production of fertilizers and various animal feeds. [12]

Reeds are versatile plants that grow quickly. Not all of their potential uses have been discovered yet, but they can be used as building and fuel materials and are also frequently used in handicrafts. Animals no longer use overgrown shores, so utilizing reeds would also be environmentally friendly, as cutting them restores animal habitats and removes excess nutrients from the Baltic Sea. At the same time, growing areas must not be overharvested, and the amount of reed cut must be monitored, as the reed must not be allowed to die out. [13]

### Threats and Problems in the Baltic Sea

The Baltic Sea and its resources are threatened by a number of different factors. The most significant factor is eutrophication, or the excessive accumulation of nutrients in the water. The causes of eutrophication are the slow water exchange in the Baltic Sea and human activities (agriculture, forestry, industry, wastewater). As a result of eutrophication, the water becomes murkier, oxygen depletion occurs, and various algae, including blue-green algae, begin to proliferate. [14]. As they decompose, blue-green algae release toxins known as cyanotoxins. These can be liver or nerve toxins and may irritate the skin. Cyanobacteria are potentially toxic, meaning that the same species of cyanobacteria may sometimes be toxic and sometimes not. Toxins can remain in the water for 1 to 4 weeks after the cyanobacteria have completely decomposed. [15]

In addition to eutrophication, there is also a great deal of marine debris in the Baltic Sea, most of which is plastic. Plastic does not decompose but breaks down into microplastics. Marine debris is harmful to all marine organisms because they can become entangled in it and develop digestive problems. To date, not all of the effects are yet known [14]. Due to the geographically low water temperature and slow water exchange, the nutrient load in the Baltic Sea (substances and materials entering the environment as a result of human activity) persists for a long time [14], which is why it is good that efforts to reduce pollution in the Baltic Sea began as early as the 20th century [16].

As a result of climate change, seawater is warming, which facilitates the spread of invasive species [14]. In addition, overfishing is a major problem, depleting fish stocks faster than they can recover. The disappearance of even a single fish species has a major impact on the ecosystem. The situation for fish species in the Baltic Sea has not improved significantly, which is why the Estonian Nature Fund has developed a “fish guide”, a set of guidelines for making sustainable fish choices, encouraging people to fish and eat fish more consciously [17].

## 2. Different Assessment Methods

### Principles for Assessing the Environmental Footprint of Products

The European Union has developed a method for assessing environmental footprints that covers both the environmental footprints of products and organizations. The environmental footprint measures a product’s impact on the environment. The pilot phase of the method began in 2013 [18] and lasted until 2018. During this time, 19 Product Environmental Footprint Category Rules (PEFCRs) and two Organizational Environmental Footprint Sector Rules (OEFSRs) were developed [18]. Several methods were updated with regard to technical aspects [18], and the results were presented in 2019 [18]. In 2021, the European Commission issued new recommendations to European Union member states and repealed the original 2013 recommendations [18]. EU member states are required to simplify access to the data necessary for the environmental footprint method, improve measurement capabilities, and encourage the inclusion of such data in databases [18].

An environmental footprint takes into account the environmental impact of products and may consider individual parameters, such as water and electricity, throughout the entire product lifecycle [19]. The assessment method developed by the European Union helps establish regulations and rules that organizations are able to follow [20]. Environmental footprint assessment methods include methods for measuring the environmental footprint of products and organizations and provide harmonized rules for assessing the environmental footprint of products and organizations [21].

### Life Cycle Assessment

Life Cycle Assessment (LCA) is a standardized method [22] that evaluates the environmental impact of a product, service, or process throughout its entire life cycle, taking into account all resources and influencing factors. A product’s life cycle begins with the acquisition of raw materials, includes production, transportation, use, and recycling, and ends with the product’s final disposal in a landfill. In other words, it covers the product’s entire lifespan, from cradle to grave. Life cycle analysis is used to assess both the negative and positive aspects of a product’s environmental impact. This assessment method can be used

to compare different products and technologies. A major drawback is the large volume and high cost of the necessary data. [23]

Today, the theory of planetary boundaries, which was developed in Stockholm in 2009, is used to describe changes in economic activity. The planetary boundaries framework addresses nine areas within the Earth system that are the most affected by human activity: climate change, novel entities, stratospheric ozone depletion, atmospheric aerosol loading, ocean acidification, modification of biogeochemical flows, freshwater change, land system change, and biosphere integrity. Exceeding these limits could trigger irreversible changes in the Earth’s biosphere. As of 2025, seven out of nine aspects have exceeded safe limits. [24]. The three areas of planetary boundaries most frequently discussed are: climate change, loss of biodiversity, and environmental pollution. All three elements are inextricably linked, which is why they must be addressed simultaneously [25].

Companies assess the life cycle of their products to examine the sustainability and environmental impact of product creation. The sustainability of consumption is examined through life cycle analysis and assessment as well [26]. The life cycle analysis method is an important tool for assessing the impact of economic activities on planetary boundaries [27].

The standard for life cycle analysis methods still needs to be updated so that production and research can be more efficient and rely on more solid foundations [28]. In addition to refining the life cycle, it is also necessary to establish reliable methods for collecting the necessary data at all stages of the product life cycle. If data is unavailable, synthetic data must be used, which is derived retrospectively from the product’s raw materials and mimics known real-world data. Synthetic data does not provide the most accurate results, but it is a better option than omitting data from the analysis and disregarding its impact. [29]

An Environmental Product Declaration (EPD) is an extension of life cycle assessment that includes sector-specific, agreed-upon measurable parameters and environmental impacts, which are presented to the consumer in a product passport accompanying the product. An EPD may also include information on how the company contributes to improving the state of the environment. [30]

### Currently Piloted Assessment Method

The demand for healthy and natural products is constantly growing. This demand can be met using traditional and new, previously untapped raw materials (innovative raw materials) sourced from water bodies. The cultivation of algae and mussels has essentially no negative environmental impacts; moreover, doing so can help restore and improve natural environments. [1]

The research is being conducted as part of the European

Commission's "Monitoring and Evaluation Sustainability Assessment Framework of Micro- and Macroalgal Value Chains" (AlgaeProBanos, APB) project. APB brings together 26 experts and industry partners from the Baltic and North Sea regions to accelerate product development and market access for sustainable algae solutions. APB supports six business pilots and startups to bring eight algae products to market. The second objective of the project is to develop a science-based LCA method and an EPD for products made from raw materials sourced from the aquatic environment, based on quantitative data and metrics. [31]. A key component of the toolkit is the framework for assessing the sustainability of algae-based products, the LCA, which includes data on energy and material flows and a wide range of environmental, social, economic, and corporate governance indicators.

Unlike the traditional perspective, which views the three pillars of sustainability (social, environmental, and economic) as separate entities, a sustainable vision should regard the economy as part of a larger ecological system [1]. In the environmental sphere, ecological and biological parameters are measured, such as biodiversity, the use of natural resources, and positive and negative impacts on the natural environment.

### 3. Research Methodology

#### Criteria for Selecting the Research Sample

My supervisor and I wrote to Estonian companies whose products use seaweed, mussels, fish, mud, clay, or reeds sourced from the sea.

I used a list compiled during the AlgaeProBanos project of Estonian companies that use marine resources – excluding raw fish – in their production. There are nine such companies in Estonia. The goal of the pilot survey was to receive responses from at least half of the companies. We sent the questionnaire to five companies, and all of them agreed to respond.

To find the rest of the companies, importers, and service providers, we used Google's search engine, but some of the companies were recommended by my personal contacts.

I limited my focus to companies that use raw materials sourced from the sea, as my work centers on the sea. Companies that use freshwater raw materials were therefore excluded from the study sample.

This is a pilot study being used to design the survey and evaluation to be conducted as part of the project "Monitoring and Evaluation Sustainability Assessment Framework of Micro- and Macroalgal Value Chains".

#### Conducting the Survey

During the course of the study, I examined how businesses

view environmental impact assessments and the collection of data required for them. For this project, I used Google Forms to survey various companies.

The questions were primarily multiple-choice and designed to provide answers to the following: a) From which countries or water bodies do the raw materials used in the companies' products/services originate? b) Which eco-labels and environmental indicators are the companies aware of? c) What environmental impact assessment metrics are companies already using? d) What data necessary for obtaining these metrics can companies access? e) What positive environmental impacts result from the company's operations? f) What are the companies' recommendations for the assessment method?

We collected background information on all companies that was necessary for the study — an overview of their economic activities and raw material use, the origin of raw materials, and product categories. Nine of the questions were multiple-choice. The last two questions (10. If you needed to assess the environmental impact of your products, what information would you need? Where could you obtain this information? Who could provide this information? 11. Other suggestions, comments, or questions that arose during the survey) were open-ended.

We sent a letter of introduction and a request to conduct the study to the companies' public email addresses, asking them to complete the questionnaire. The companies that participated in the study are kept anonymous in my work, as, in accordance with the APB project agreements, the data will not be disclosed until the project is completed in 2027. The companies are numbered in the order in which they responded. In total, we sent the questionnaire to 14 companies and received responses from nine. The estimated time required to complete the survey was 20–40 minutes.

### 4. Results

#### Company Overview

We received responses from nine Estonian companies that we selected based on the criteria mentioned earlier. I personally sent the survey to five companies, none of which responded.

Of the responding companies, four were manufacturers (companies 2, 4, 5, 8), three were importers (companies 3, 6, 7), one was a service provider (company 1), and one was both a service provider and a manufacturer (company 9). The number of products in the companies' product portfolios varied significantly. Company 7 has one product, while Company 2 has 32 products. Most have fewer than ten products. Seaweed is used by seven companies, while mud is used by three companies. Only Company 3 uses shells, and only Company 8 uses reeds. Fish and clay are both used by two companies, and fish is not the sole raw material for either company; one also uses seaweed, and the other uses seaweed and shells. (see Table 1)

**Table 1.** Overview of companies based on economic activity and raw material use (“+” indicates which option the company selected)

Options	Company nr								
	1	2	3	4	5	6	7	8	9
Manufacturer		+		+	+			+	+
Importer			+			+	+		
Service provider	+								+
Nr of products, pcs.	4	32	10	3	14	20	1	3	7
Seaweed	+		+	+	+	+	+		+
Mud	+	+		+					
Shells			+						
Reed								+	
Clay	+			+					
Fish			+						+

The main categories into which the companies’ products fall are cosmetic and or food-related products. Three companies deal with cosmetics, two with human food, one with pet food, one with dietary supplements, and one with food contact products. One company sells therapeutic mud. (see Table 2)

**Table 2.** Product categories

Product category	Company nr								
	1	2	3	4	5	6	7	8	9
Cosmetics				+	+	+			
Therapeutic mud		+							
Food						+			+
Pet food			+						
Dietary supplements							+		
Food contact products (straws)								+	

Five companies source their raw materials from the Baltic Sea, three of which also source from Estonian waters. Some companies source their raw materials from Norway or Europe more broadly, but one company sources its raw materials from the United States.

## Companies' Knowledge of Various Options for Assessing Environmental Impact

Most companies were aware of at least two methods or labels used to assess environmental impact. Only Company 2 was unaware of any. All other companies knew what an eco-label and a product’s environmental footprint were. Seven companies were also familiar with the organic label. Five companies are familiar with EPDs and five companies are familiar with ESG. The least well-known is LCA, which only four companies are aware of. (see Table 3)

**Table 3.** Overview of businesses’ responses to the question: “Various types of analysis are used to assess the environmental impact of products and services. Please check all options you are aware of.”

Product evaluation options	Company nr								
	1	2	3	4	5	6	7	8	9
Eco-label	+		+	+	+	+	+	+	+
Organic label (food products)	+		+	+		+	+	+	+
Environmental footprint	+		+	+	+	+	+	+	+
Life Cycle Analysis (LCA)	+					+		+	+
Environmental Product Declaration (EPD)	+					+	+	+	+
ESG	+		+			+	+	+	
Not aware of any		+							

**Table 4.** Overview of companies’ responses to the question: “Please check all options that your company already applies to a specific product or the entire production process (e.g., you already have a label or certificate, or you have conducted an assessment).”

Product evaluation options	Company nr								
	1	2	3	4	5	6	7	8	9
Eco-label					+	+			
Organic label (food products)						+			
Environmental footprint								+	
Life Cycle Assessment (LCA)								+	
Environmental Product Declaration (EPD)									
ESG								+	
None of the above	+		+	+					+
Other certificates		+					+		

Four of the responding companies do not use any assessment methods or hold any certifications. Two companies use an eco-label, and one of them also uses an

organic label. Company 8 uses the most assessment methods, employing LCA and ESG in addition to assessing the product's environmental footprint. (see Table 4)

## Companies' Assessments of the Availability of Necessary Data

I asked the companies to list the environmental impacts of their products of which they were already aware. Companies 3 and 5 currently had no information regarding the environmental impact of their products. Six companies were able to specify the size of the marine area required for their product and indicate what proportion of their raw materials is locally sourced. Five companies knew which habitats are affected by the production of their products. Four companies knew how their production affects eutrophication and how much waste is generated during production. Companies 6 and 9 had information on twelve out of seventeen different environmental impacts. (see Table 5)

Next, I examined what kind of information companies could find on their own, from databases, or with the help of the government or researchers regarding the environmental impact of their products. I also considered how this information could support better decisions in product development and sustainability planning.

The number of possible answers from companies 1, 7, and 8 increased significantly with the option of external assistance. Previously, they had answers to fewer than ten questions, and now they can answer nearly all of them. Company 9 could answer all questions with such assistance. The capabilities of the other companies remained the same. (see Table 6)

## The Positive Environmental Impacts of Business Operations and Recommendations Regarding the Accessibility of Information

Four companies preserve biodiversity through their operations, and three companies enhance biodiversity. Two of these companies chose both preserving the environment and enhancing biodiversity. Two companies sequester CO<sub>2</sub> as a result of their activities, and two companies restore habitats. The activities of two companies have no positive impact on the environment. Company 5 was unable to answer. (see Table 7)

According to Companies 5 and 8, manufacturers and raw material suppliers should be the primary sources from which they obtain data to assess their environmental impact. Cooperation partners were also mentioned. Company 5 specifically mentioned Tallinn University; the other respondents did not provide further details. Company 1 proposed the idea of creating an automated application where researchers and companies could exchange information, and the information would be available to companies free of charge.

**Table 5.** Companies' responses to the question: "When assessing the environmental impact of your products, you, as a business owner, would need to know the following information. Please indicate the answers where you already know the impact of your product on the natural environment. Select all that apply."

Questions asked	Company nr								
	1	2	3	4	5	6	7	8	9
How much marine area is required for growing/harvesting raw materials (ha, m <sup>2</sup> )?	+	+				+	+	+	+
What habitats does the production of your product affect?	+					+	+	+	+
What key species does the production of your product affect?						+		+	+
How does production contribute to eutrophication?	+					+		+	+
How much nitrogen do you use in production?	+					+			
How much phosphorus do you use in production?	+					+			
Does production result in chemical pollution (amount of chemicals used, e.g., herbicides, insecticides, antibiotics, algicides)?	+					+			+
Does cultivation or production result in SO <sub>2</sub> (sulfur dioxide) emissions?						+			+
Can production increase the number of invasive species?									+
How much greenhouse gas is emitted?							+		
Are there emissions of any other greenhouse gases besides CO <sub>2</sub> ?						+			
What percentage of the energy used comes from renewable energy sources?	+								+
Do your activities help preserve biodiversity in the marine environment?									+
Do your activities in the marine environment increase biodiversity?							+	+	+
What percentage of raw materials is locally sourced (from Estonia or the Baltic Sea)?	+	+		+		+	+		+
How much waste is generated during production (kg, t)?	+					+		+	+
What is the volume of wastewater generated during production?						+			
No possible answer			+		+				

**Table 6.** Companies' responses to the question: "When assessing the environmental impact of products, you, as a business owner, would need to know the following information. Please indicate which of the following questions you would be able to answer (on your own, using various databases, with government assistance, with the help of researchers, etc.). Select all that apply."

Questions asked	Company nr								
	1	2	3	4	5	6	7	8	9
How much marine area is required for growing/harvesting raw materials (ha, m <sup>2</sup> )?	+	+				+	+	+	+
What habitats does the production of your product affect?	+					+	+	+	+
What key species does the production of your product affect?	+					+	+	+	+
How does production contribute to eutrophication?	+					+		+	+
How much nitrogen do you use in production?	+					+			+
How much phosphorus do you use in production?	+					+			+
Does production result in chemical pollution (amount of chemicals used, e.g., herbicides, insecticides, antibiotics, algicides)?	+					+	+	+	+
Does cultivation or production result in SO <sub>2</sub> (sulfur dioxide) emissions?						+	+	+	+
Can production increase the number of invasive species?	+						+		+
How much greenhouse gas is emitted?	+						+	+	+
Are there emissions of any other greenhouse gases besides CO <sub>2</sub> ?	+					+	+	+	+
What percentage of the energy used comes from renewable energy sources?	+						+	+	+
Do your activities help preserve biodiversity in the marine environment?	+						+	+	+
Do your activities in the marine environment increase biodiversity?	+						+	+	+

What percentage of raw materials is locally sourced (from Estonia or the Baltic Sea)?	+	+			+		+	+	+
How much waste is generated during production (kg, t)?	+						+	+	+
What is the volume of wastewater generated during production?	+						+		+
No possible answer				+		+			

**Table 7.** The positive environmental impacts of businesses

Positive impacts	Company nr								
	1	2	3	4	5	6	7	8	9
Remove nutrients from the aquatic environment	+							+	+
Absorb CO <sub>2</sub>						+			+
Restore habitats						+		+	
Their activities help preserve the biological diversity of the marine environment						+	+	+	+
Contribute to the biological diversity of the marine environment through their activities				+		+			+
Do not occur		+	+						

## 5. Generalization and Conclusions

### Analysis

Companies currently use algae that have grown naturally or in cultivation. One of the biggest environmental problems in the Baltic Sea is eutrophication, which refers to the excessive accumulation of nutrients. Research has shown that large-scale algae and mussel farming is an effective way to combat eutrophication. Algae cultivation could also benefit people and businesses in the Baltic Sea countries, as evidenced by the fact that seven of the responding companies use algae in their products. The responding companies source their raw materials from the Baltic Sea and other European regions, but raw materials are also imported from the United States.

As shown in Table 3, companies are most familiar with eco-labels and the product environmental footprint method, but few are familiar with life cycle assessment (LCA). It is

recommended that companies become familiar with the LCA method, as it will soon be important for products sold on the European Union market. Environmental footprints are discussed more widely around the world than LCA, which may explain the four companies' awareness of LCA. EPDs are set to be adopted more widely in the EU market, and five of the responding companies are already familiar with this assessment method. At present, none of the responding companies have implemented EPDs, but they still have time before EPDs are more widely adopted in the EU.

A comparison of Tables 3 and 4 reveals a clear discrepancy between the assessment methods companies are aware of and the methods they actually use. Awareness is widespread, but few companies have conducted an environmental impact assessment. Only a few of the responding companies actually hold environmental labels, such as Companies 5 and 6, which have an eco-label, and Company 8, which has conducted an environmental footprint assessment, LCA, and ESG. Companies 1, 3, 4, and 9 do not hold any environmental labels. The fact that companies have not yet pursued environmental certifications indicates that the European Union still needs to work on the sustainability and implementation of the green and blue economies.

As things stand, two companies would be able to obtain the data needed to conduct a detailed and comprehensive environmental impact assessment. Only three companies would face difficulties in conducting an environmental impact assessment due to a lack of data and its inaccessibility. The remaining four could conduct a preliminary analysis, but they lack the means to answer some of the questions. This situation is promising, as I initially thought that only a very small number of companies would be able to answer the questions. It would be even better if all companies could find answers to the questions necessary for assessing environmental impacts, but that is not the objective of this study.

Six of the nine responding companies have a positive impact on the environment through their activities, which was to be expected, as seaweed cultivation has positive environmental effects and the majority of companies use seaweed in their products. Companies 6 and 9 have the most positive impact on the environment due to their activities, as both have four of the positive environmental impacts we listed.

Company 1 proposed the idea of creating an app where companies, researchers, and raw material suppliers can communicate with one another and share information. It was noted that the app should be free of charge. The idea proposed by Company 1 could be something worth pursuing in the future. Such an app would help all participants easily connect with one another. The app could

include a feature allowing companies to submit their data to the government or the EU and, through this, obtain labels and necessary certifications, thereby creating a public and transparent database that would facilitate the assessment of companies' environmental impacts.

## **Self-Analysis**

It's possible that the estimated time required for the survey deterred some companies, as 40 minutes is quite a long time – though in hindsight, that estimate was a bit of an exaggeration. One company even expressed dissatisfaction that the survey took so long, even though the introduction to the questionnaire stated that it would take some time.

Companies were also more willing to respond when the email came from the Estonian Marine Institute. The emails I sent received no reply, which suggests that a 11th-grade student's research project isn't exactly a priority. My advisor and I wrote emails that were almost identical, so presumably the wording of the emails did not play a major role in how open the companies were to participating in the study. It seems that the study is perceived as less important.

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