

Research Title:	Evaluating the Defense Mechanisms of Selected Plants in Khlong Tamru,				
	Chonburi, Thailand against Biotic and Abiotic Stressors in Soil and Water				
Researchers:	Nattawat Kusoltipcharoen, Methasit Nugate, Pacharapol Sriwisut, Warit				
	Khlaipanpee, Pathawee Chalermpanich, Pranchalee Boonsiri, Sirapat				
	Niyom, and Benyapha Lekmana				
Level:	High School (Grade 10-11)				
School:	Chonradsadornumrung School				
Advisors:	Ms. Rawadee Meesuk and Mr. Marvin Servallos				

ABSTRACT

This current environmental science research aims to evaluate the defense mechanisms of selected plants present in the brackish river of Khlong Tamru, Chonburi, Thailand. Using the standard equipment from Extech, the researchers characterized various water and soil physico-chemical factors such as water temperature, TDS, dissolved oxygen, electrical conductivity, salinity, transparency, water pH, air temperature, soil temperature, soil pH, NPK concentration, and relative humidity. Then, the leaves of three selected plants were gathered for the phytochemical screening. Based on the experimentations, results and gathered data, the researchers concluded that there are significant differences (p<0.05) in water temperature, dissolved oxygen, TDS, electrical conductivity, transparency, salinity, soil temperature (5cm depth), soil pH, and relative humidity except for soil temperature at 10 cm depth (p>0.05). Moreover, Nipa Palm (*Nypa fruticans*), Red Mangrove (*Rhizophora mangle*) and Spurred Mangrove (*Ceriops tagal*) contains various secondary compounds as their defense mechanism against biotic and abiotic stressors in soil and water. For the improvement of the study, further research will be conducted to evaluate antimicrobial activities of the experimental plants against their biotic stressors.

Keywords: Brackish, Biotic stressors, Abiotic stressors, Secondary Compounds

INTRODUCTION

In the past, Khlong Tamru district in Chonburi, Thailand, was merely a modest town with a diverse range of plants growing along its brackish river, which is connected to the Gulf of Thailand. Currently, it is undergoing rapid changes because it is included in the Eastern Economic Corridor (EEC), which aims to boost a region's economic activities. As a result, various infrastructures have been constructed, including factories, bridges, and buildings. Brackish water from Klong Tamru river is beneficial to the various forms of life that are existing in the area. Plants like Nipa Palm, Spurred mangrove, and Red Mangrove are common halophytes that benefits from the river. These plants produce various substances like secondary metabolites that protect them from microbial pathogens and abiotic stresses in their environment (Schafer et al., 2009). These compounds are also responsible for plants biological activities that are also beneficial to human beings. The production of these valuable compounds is linked to the nutrients that plants absorb from the soil and the water. Previous research emphasized that reasonable proportion of nutrition (NPK) factors directly promote the absorption and assimilation of plants, thereby affecting their growth and development (Yildirim et al., 2011). Other factors, like dissolved oxygen (DO) in the water can respond to facilitate the availability of essential nutrients such as NPK, low DO levels can impact the roots' function and stunned growth of the plants (Raven, Evert, & Eichhorn, 2005).

The intensity of anthropogenic activities near the river may have had an immense impact on the survival of plants especially, in terms of their nutrient absorption which has great impact on their growth and development. Given the rapid changes occurring in Khlong Tamru, Chonburi, Thailand that could affect the diversity of life particularly the plants, it is imperative to evaluate the soil and water quality of this place.

The situations above prompted the researchers to conduct environmental research entitled "Evaluating the Defense Mechanisms of Selected Plants in Khlong Tamru, Chonburi, Thailand, against Biotic and Abiotic Stressors in Soil and Water". The current study sought to characterize the physical and chemical features of the river and soil. Most importantly, the defense mechanisms of selected plants in Khlong Tamru, Chonburi, Thailand, against various biotic and abiotic stresses in soil and water were assessed.

Research Questions:

- 1. Is there a significant difference in soil and water parameters measured in the brackish river of Khlong Tamru, Chonburi, Thailand?
- 2. What secondary compounds do selected plants produce as their defense mechanism against biotic and abiotic stressors in soil and water?

Objectives:

- 1. To determine if there are significant differences in the soil and water parameters in Khlong Tamru, Chonburi, Thailand.
- 2. To evaluate the defense Mechanisms of selected plants in Khlong Tamru, Chonburi, Thailand, against biotic and abiotic stressors in soil and water.

Hypotheses:

Alternative: There is a significant difference in the physico-chemical parameters of the brackish river of Khlong Tamru, Chonburi, Thailand and the selected plants possess secondary compounds as their defense mechanism against biotic and abiotic stressors in soil and water.

Null: There is no significant difference in the physico-chemical parameters of the brackish river of Khlong Tamru, Chonburi, Thailand and the selected plants do not possess secondary compounds as their defense mechanism against biotic and abiotic stressors in soil and water.

Materials:

Materials and equipment used in this science project.

Secchi disc	NPK concentration sensor	Beakers
pH, Conductivity, TDS,	Phytochemical screening	Meterstick
Salinity, Temperature Meter	chemicals	
Dissolved Oxygen meter	2 in 1 soil Analyzer	Graduated cylinder
Thermo Hygrometer	Refractometer	Erlenmeyer flask
Stirring rod	Test tube rack	Shovels

Research Methodology

A. Study Site

The study site is in the brackish river of Khlong Tamru, Chonburi, Thailand with a Latitude 13°27'17.9"N, Longitude 100°59'29.1"E.

B. Survey and Preparation of Materials

The researchers went to the



FIGURE 1. The study site on Khlong Tamru, Chonburi, Thailand.

brackish river of Khlong Tamru, Chonburi, Thailand to conduct a field study in the brackish river of Khlong Tamru District to determine the soil and water quality of the place. After the survey and selection of study site, needed laboratory materials and equipment for soil

testing were procured from the science laboratory of Chonradsadornumrung School.

C. Soil Quality Testing

Various soil parameters were considered in assessing the quality of soil in the brackish river of Khlong Tamru, Chonburi such as NPK concentration, soil pH, temperature, moisture, soil texture, soil consistency, soil fertility, also the air temperature and relative humidity of the study site was included.



FIGURE 2. *In situ* measurement of soil quality.

To determine the soil pH, the following steps were carried out: 40 g of dried and sieved soil with 40 mL of distilled water (or other amount in a 1:1 soil to water ratio) was mixed in a beaker using the stirring rod, the mixture was allowed to settle until a supernatant (clearer liquid above the settled soil) formed, the pH of the supernatant was measured using the pH meter. Extech standard thermo-hygrometer was used to determine the air temperature and relative humidity of the study site. Moreover, NPK concentration sensors were used to determine the amount of nitrogen (N), phosphorus (P), and potassium (K) present in the soil.

D. Water Quality Testing

Various water parameters were assessed in the brackish river of Khlong Tamru, Chonburi, Thailand such as water temperature, dissolved oxygen, total dissolved solids (TDS), electrical conductivity, transparency, water pH, salinity, relative humidity, also the air temperature and water color of the study site were included. To determine the water parameters, the following steps were carried out: 500 mL of brackish water was collected in

a beaker for measuring water parameters, the salinity was measured using Optik handheld refractometers. Secchi disk was used to measure the transparency of water. Extech measuring equipment were also used to examine the dissolved oxygen, TDS, electrical conductivity, water temperature and water pH. Visual comparison was used to evaluate the color of the water. In this test 20ml of the sample and 20ml of distilled water were taken in two separate wide mouthed test tubes. The results



FIGURE 3. *In situ* measurement of water quality.

were tabulated (as clear or turbid) by comparing the color of the sample with distilled water. Thermo-hygrometer was used to determine the air temperature and relative humidity of the study site. The hydrosphere protocols from <u>www.globe.gov</u> were used in all the tests needed to evaluate the physico-chemical parameters of the water and soil.

E. Phytochemical Screening

Phytochemical screening procedures from the book of Guevarra et.al., 2005 were applied to evaluate the presence of the secondary compounds responsible for inhibiting the growth of foodborne pathogens like Alkaloids, Saponins, Tannins, Phenolic Compounds, Flavonoids, and Leucoanthocyanins.



FIGURE 4. Testing the secondary compounds of the selected plants.

Results and Discussions

The figures below and on the following page show the data encoded on the Globe website from December 2024 to January 2025. Figures 5 through 12 depict the Globe data entry for air temperature, relative humidity, soil temperature, surface temperature, soil fertility, and soil pH measured in the brackish river of Khlong Tamru, Chonburi, Thailand.

Measurements	Data Counts S	School Info	Site Info	Photos	
O 10 cm Soil Noor O 50 cm Soil Noor		12 [.] - 07 6 [.] -	•	ingland day's average of v * 4 5 12025-2-12	•

Figure 5. Globe Data Entry for soil temperature (5 cm).

School: Chonradsador Site: Khlong Tamru, Chonk	-	ool 🖆			< 🖉 ×
Measurements E	ata Counts	School Info	Site Info	Photos	
Atmosphere Air Temperature Daily Average Temp	▼.	28	Plot only d	isplays day's average of val	• III
Data Date Range: 2024		02-07 21			
		7	0 2 0	N 8 N 9 N	
		ALL COLOR	2024-12-1	5 2025-2-10	Plot X

Figure 7. Globe Data Entry for air temperature.

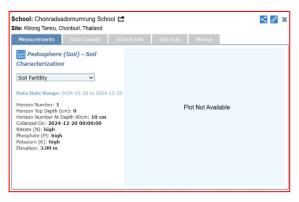


Figure 9. Globe Data Entry for soil fertility.

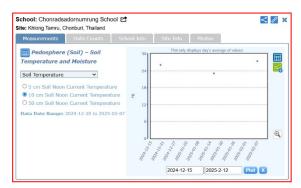


Figure 6. Globe Data Entry for soil temperature (10 cm).

School: Chonradsadornumrung School [Site: Khlong Tamru, Chonburi, Thailand	± 🛛 🖉 🤉
Measurements Data Counts S	chool Info Site Info Photos
Pedosphere (Soll) - Soll Characterization Soll pH v Data Date Range: 2024-12-20 to 2024-12-2 Horizon Number: 1 Horizon Number Control Horizon Number At Depth 900m: 10 cm Collected On: 2024-12-20 00:00:00 Soll pt: S-S pH units PH Method: meter Elevation: 3.00 m	o Plot Not Available

Figure 8. Globe Data Entry for soil pH.

iool I	Info	Site Info	Photos	
	8.5 6.8	•	•	
pH units	3.4			
	1.7	~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~	e e r r r	\$ \$ 8
	024.12.24 C	Contraction of the second	11,100,100,100,100,100,100,100,100,100,	50010 50010 50010 50010 50010 500
		6.8 5.1 Hd 3.4	8.5 6.8 5.1 	8.5 6.8 5.1 2.4

Figure 10. Globe Data Entry for water pH

School: Chonradsadornumrung Site: Khlong Tamru, Chonburi, Thailar		9			K	8 ×
Measurements Data Count	s Sch	ool Info	Site Info	Photos		
Hydrosphere Dissolved Oxygen Ossolved Oxygen via Kit Dissolved Oxygen solitation Dissolved Oxygen solitation Dissolved Oxygen solitation Data Date Range: 2025-01-16 to 2	025-02-07	5 4 3 2 1 0 9 7 7 1 0 9 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7	• * * * * * * * * * * * * *	5 [2025-2-10	· · · · · · · · · · · · · · · · · · ·	

Figure 11. Globe Data Entry for dissolved oxygen

Measurements Data Counts	School Info	Site Info	Photos	
Hydrosphere	30][7		
Water Temperature 🗸	24			
Water Temperature		•		
ata Date Range: 2025-01-16 to 2025-02-	07 18 -			
	6 12			
	5			
				•
	2	2 2 2 3	60. 19. 19.	
	Con Los	1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1	and	13.45 C
	17° 1	2024-12-15		N N Plot X

Figure 12. Globe Data Entry for water temperature

Table 1. Average results of soil characterization measured in the brackish river of KhlongTamru, Chonburi, Thailand.

Parameters	20 Dec. 2024	16 Jan. 2025	7 Feb. 2025
	(4:20 PM)	(9:50 PM)	(3:30 PM)
Soil pH	5.5	5.73	7
Soil Temperature (5 cm)	25	22	27
Soil Temperature (10 cm)	26	23	27.3
Relative Humidity (%)	34.33	44	60.33
Air Temperature (°C)	30.17	26.97	29.57
Soil Color	Brown	Brown	Brown
Soil Texture	Clay	Clay	Clay
Soil Consistency	Firm	Firm	Firm
Nitrogen (N)	High	High	High
Phosphorus (P)	High	High	High
Potassium (K)	High	High	High

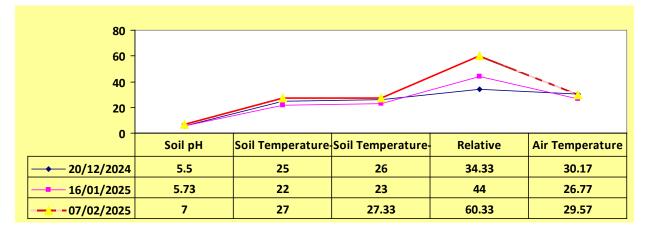


Figure 13. Average results of all soil parameters measured in Khlong Tamru, Chonburi.

Table 1 and Figure 13 shows the average results of all soil parameters measured in the brackish river of Khlong Tamru, Chonburi, Thailand. These results were summarized after 3 series of experiments that started from 12 December 2024 to 7 February 2025. The average soil pH ranges from 5.5 - 7, soil temperature at 5cm depth ranges from $22 - 27^{\circ}$ C, soil temperature at 10cm depth ranges from $23 - 27.33^{\circ}$ C, relative humidity ranges from 34.33 - 60.33%, and air temperature ranges from $26.77 - 30.17^{\circ}$ C. The soil tested from the experimental site also possess the following characteristics: brown color, clay soil structure, clay texture, firm consistency, wet, and has high concentration of Nitrogen (N), Phosphorus (P), and Potassium (K).

One-way ANOVA and Tukey HSD test were used to determine if there was a significant difference in all soil parameters measured quantitatively in the river of Khlong Tamru, Chonburi. It was found out that the p-value corresponding to the F-statistic of one-way ANOVA is lower than 0.05 for air temperature, soil pH, soil temperature measured at 5 cm depth and relative humidity suggesting that the one or more treatments is/are significantly different. For soil temperature (10 cm depth), the p-value corresponding to the F-statistic of one-way ANOVA is higher than 0.05, suggesting that the treatments are not significantly different for that level of significance. It means that there were no significant changes in the soil temperature (10 cm depth) measured for 3 times from the sampling site. **Table 2.** Average results of water physico-chemical parameters measured in the brackish river of Khlong Tamru, Chonburi, Thailand.

Parameters	20 Dec. 2024	16 Jan. 2025	7 Feb. 2025
	(4:00 PM)	(9:50 AM)	(3:30 PM)
Water Temperature (°C)	23.07	20.47	27.43
Dissolved Oxygen (mg/L)	2.05	2.88	1.2
TDS (ppm)	3200	3146	2991
Electrical Conductivity (µS/cm)	6340	6092	6168
Transparency (m)	0.29	0.22	0.17
Water pH	7.56	8.09	7.98
Salinity (%)	30	35	29
Water Color	Turbid	Turbid	Turbid

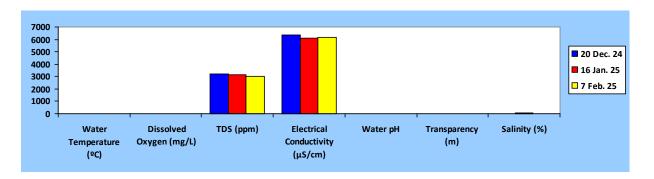


Figure 14. Average results of all water physico-chemical parameters measured in Khlong Tamru, Chonburi.

Table 2 and Figure 14 shows the average results of all parameters measured in the brackish water of Khlong Tamru, Chonburi, Thailand. These results were summarized after 3 series of experiments that started from 20 December 2024 to 7 February 2025. The average water temperature ranges from 20.47-27.43°C, DO ranges from 1.2-2.88 mg/L, TDS ranges from 2991-3200 ppm, Electrical conductivity ranges from 6092-6340 μ S/cm, Transparency from 0.17-0.29 m, water pH from 7.56-8.09, salinity from 29-35%, The water color was consistently turbid throughout the experiment.

Statistical tools namely One-way ANOVA and Tukey HSD test revealed that the pvalue corresponding to the F-statistic is lower than 0.05 for water temperature, DO, TDS, Electrical conductivity, transparency, salinity, and water pH suggesting that one or more treatments is/are significantly different.

Secondary Compounds	Nipa Palm	Red Mangrove	Spurred Mangrove
Alkaloids	+	+	+
Saponins	+	-	+
Tannins	+	+	+
Phenolic Compounds	+	+	+
Flavonoids	+	+	+
Leucoanthocyanins	+	-	+
(+) : possessed by the plan	nt (positive); (-) : is r	not possessed by the	plant (negative)

Table 3. Results of Phytochemical Screening



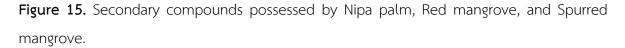


Table 3 and Figure 15 shows the observations done by the researchers during the phytochemical screening of the ethanolic extracts of Nipa Palm (*Nypa fruticans*), Red Mangrove (*Rhizophora mangle*) and Spurred Mangrove (*Ceriops tagal*). After conducting various tests, it was found out that the leaves of Nipa Palm and Spurred mangrove (*C. tagal*) are positive for the presence of Alkaloids, Saponins, Tannins, Flavonoids, Leucoanthocyanins, and Phenolic compounds. The leaves of Red Mangrove are positive for Alkaloids, Tannins, Phenolic Compounds, and Flavonoids.

The researchers used the protocol from the book of Guevara, et.al., 2005; to identify the presence of each secondary metabolites. For Alkaloids, heavy precipitation was used as indication for its presence in plant extract. For saponins, a bubble that is greater than 3 cm above the liquid surface after shaking the test tube. For tannins, the presence of precipitate. For phenolic compounds, change of extract from slightly clear to black after the addition of Ferric chloride. For flavonoids and leucoanthocyanins, color changes (yellow to brown for flavonoids and red to violet for leucoanthocyanins) were used as the basis for their presence after boiling the extract. These secondary metabolites are the defense mechanism of the plants to various biotic and abiotic factors present in water and soil (Briskin, 2000).

Discussion

The results of field measurement, ANOVA, and post-hoc Tukey HSD test showed that there was a significant difference in various water and soil parameters measured in the brackish water of Khlong Tamru, Chonburi, Thailand such as water temperature, water pH, salinity, TDS, electrical conductivity, air temperature, dissolved oxygen, soil temperature measured at 5 cm and relative humidity but there was no significant difference in soil temperature measured at 10 cm depth. These results revealed that plants thriving in the brackish water of the experimental place are experiencing varied physico-chemical factors. Aside from this, there are also biotic stressors that are present in the soil and water. In order for plants to survive in their environment with biotic and abiotic stresses, there is a great need for them to produce essential compounds that will counteract the adverse effects of various factors around them.

Research done by many researchers revealed that plants produce natural products or secondary metabolites with a prominent function in the protection against predators and microbial pathogens (Schafer et.al., 2009). Literally, phytonutrients are produced by the plants to counteract various biotic and abiotic stressors in their environment (Kessler et.al, 2018). The presence of biologically active secondary metabolites in the plants makes them medicinally significant because these substances exert their effects by interacting with human physiology (Kasimala et.al, 2014).

The secondary compounds possessed by the three plants are responsible for their survival and adaptation in their habitat (Cimmino et.al., 2017). It is known that plants produce flavonoids and alkaloids in response to microbial infection. This fact made this substance an efficient antibacterial compound against a variety of pathogenic organisms (Jamine et al., 2007). Moreover, tannins and phenolic compounds have been shown to precipitate microbial protein, which inhibits the growth of microorganisms. This substance prevented the growth of numerous fungi, yeasts, bacteria, and viruses (Prasad et al., 2008).

The soil in the brackish water of Khlong Tamru, Chonburi, Thailand is so rich in nutrients due to high concentration of organic matters and minerals such as nitrogen, phosphorus, and potassium as shown in Table 1. The richness of the soil is noticeable due to the presence of abundant and various plants in the area. The nutrients absorbed by the plants like phosphorus is involved in energy metabolism and photosynthesis during plant growth (Yan et al., 2021). Whereas, potassium plays an important role in carbohydrate and protein metabolism (Hassanein et al., 2021), indicating that a reasonable proportion of nutrition (NPK) factors directly promote the absorption and assimilation of plants, thereby affecting their growth and development (Yildirim et al., 2011). Since plant's biological and physiological needs are satisfied, they were able to produce various compounds both

primary and secondary compounds that are necessary for their growth and development as well as survival in their natural habitat with biotic and abiotic stressors (Briskin, 2000).

Conclusion

Based on the experimentations, results and gathered data, the researchers concluded that there are significant differences (p<0.05) in water temperature, dissolved oxygen, TDS, electrical conductivity, transparency, salinity, soil temperature (5cm depth), soil pH, and relative humidity except for soil temperature at 10 cm depth (p>0.05). Moreover, Nipa Palm (*Nypa fruticans*), Red Mangrove (*Rhizophora mangle*) and Spurred Mangrove (*Ceriops tagal*) contains various secondary compounds as their defense mechanism against biotic and abiotic stressors in soil and water.

Recommendations

For the improvement of the study, further research will be conducted to evaluate antimicrobial activities of the experimental plants against their biotic stressors. Additionally, more plants growing in the brackish water of Khlong Tamru will be studied for their secondary compounds and biological activities.

GLOBE Badges

I am a Collaborator

The effective completion of this environmental research was made possible because of the collective efforts of various individuals. The community living in Khlong Tamru district, allowed the researchers to conduct an investigation about the current condition of the river as well as to research the beneficial effects of the plants thriving in the area. For convenient and effective investigation, the researchers were given a place near the river. During the conduct of the study, the researchers were thoroughly guided and given knowledge by their teachers namely Ms. Rawadee Meesuk and Mr. Marvin Servallos. Furthermore, the soil and water quality testing were carried out properly because of the materials provided by the administrators of the school, including the school van, which was used for seamless transit from school to the study site. Thorough guidance and invaluable ideas from the above names were very significant to completely understand all the scopes of this research. Finally, the researchers of this science project have cooperated to finish the work entirely from the planning stage, experiments, analyzing of data, and packaging of the final research paper.

I Make an Impact

The success of this experiment would greatly benefit the community along Khlong Tamru river, and government officials because the results of soil quality measurement in the natural habitat of halophytes would give them valuable information about the current condition of Samet soil where various halophytes are thriving and other flora are thriving. Moreover, the result of antimicrobial screening is very helpful because the community in Thailand can use the aqueous extracts of halophytes as an alternative method in fighting the adverse effects caused by bacteria. This research will significantly impact including the agriculture sector because the result of allelopathic test will propel them to use organic materials like halophyte extracts in inhibiting the growth of unwanted plants instead of using commercial herbicides that are expensive and harmful to the body. Most importantly, the methods and results gathered in this study have great impact to the community of Chonradsadornumrung School, especially to the students because it serves as an eye opener for them that young learners like the researchers can have a valuable contribution in studying their surroundings like soil quality, also in using the natural resources as alternative agents in inhibiting the growth of pathogenic microbes.

I am a Data Scientist

The researchers have studied systematically the current condition of water and soil sample in the brackish river of Khlong Tamru, Chonburi, Thailand. The results were collected, recorded, and analyzed properly. The water and soil physico-chemical factors were correlated to the secondary compounds produced by the selected plants. In addition, botany and phytochemistry experiments were integrated to this environmental research. All of the data gathered from the field measurement were analyzed using some statistical models like ANOVA (Analysis of Variance) with post-hoc Tukey HSD (Honestly Significant Difference) Test. The results of the analysis were discussed and presented properly. Moreover, the results of the experiment were linked to the research done by other researchers. Lastly, the results presented here will be valuable to the future researchers who will investigate further the quality of the river in Khlong Tamru, Chonburi, Thailand.

I am a STEM Professional

This environmental research is not possible without the invaluable insights from various STEM Professionals. The researchers were able to formulate the research topic and questions by asking their Biology teacher pertaining to possible studies that they can pursue that is relevant to Globe's mission and vision. During data gathering and interpretation, the statistician of the school was consulted about the accurate statistical method that can be used in interpreting the data about soil quality in Samet, Chonburi, Thailand as well as the results of biological testing. From the consultation with the school's statistician, the researchers learned that ANOVA (Analysis of Variance) and post-hoc Tukey HSD (Honestly Significant Difference) Test are the most appropriate tool to analyze the collected data. Moreover, the researchers also asked help from the statistician of the school to calculate and interpret the results of the study.

Acknowledgment

The researchers of the study would like to acknowledge the following for making this environmental science research possible. First, they would like to convey their genuine thanks to the Head of Chonradsadornumrung School English Program, Ms. Rawadee Meesuk for her utmost support, suggestions, and encouragement as well as for providing all the Laboratory equipment and chemicals that they need in their study. Second, heartfelt thanks are also conveyed by the researchers to their Science teacher- Mr. Marvin Servallos for his thorough guidance towards the completion of the study. Third, sincere gratitude is given by the researchers to the administration of the school for providing the transportation needs of the globe researchers. Fourth, the community of Khlong Tamru deserves a huge thank for granting the researchers permission to conduct a study in their place. Finally, the researchers would like to give their special thanks to the committee of Globe IVSS 2025 and Globe Student Research Competition for conducting this prestigious event that enabled young scientists to share their scientific discoveries.

References

<u>Books</u>

Burton, Gwendolyn R. W. et al. (2004). *Microbiology for the Health Sciences. USA,* Lippincott: Williams and Wilkins.

Guevara, Beatrice Q. et al. (2005). *A Guidebook to Plant Screening: Phytochemical and Biological*. Manila, Philippines: UST Publishing House.

<u>Internet</u>

Ballhorn DJ, Kautz S, Heil M, Hegeman AD, 2009. Cyanogenesis of wild lima bean (Phaseolus lunatus L.) is an efficient direct defense in nature. Plant Signaling and Behavior, 4(8): 735-745. Retrieved from https://www.omicsonline.org/open-access/role-of-secondary-metabolites-in-defense-mechanisms-of-plants-0974-8369-3-128.pdf

Briskin, D. (2000). Medicinal plants and phytomedicines. Linking plant biochemistry and physiology to human health. Retrieved from https://academic.oup.com/plphys/article/124/2/507/6098853?login=true

Cimmino et. al. (2017). Amaryllidaceae Alkaloids: Absolute configuration and biological activity. Retrieved from https://onlinelibrary.wilev.com/doi/epdf/10.1002/chir.22719

Rajkaran, A., Adams, J. & van der Colff, D. 2016. Rhizophora mucronata Lam. National Assessment: Red List of South African Plants version 2017.1. Accessed on 2017/12/07. Retrieved from: <u>http://pza.sanbi.org/rhizophora-mucronata-</u>

Rhoades J, Kandiah A and Mashali A 1992 The use of saline waters for crop production (Rome: FAO United Nations). Retrieved from <u>https://iopscience.iop.org/article/10.1088/1755-1315/118/1/012019/pdf</u>

Sawyer C, McCarty P and Parkin G 1994 Chemistry for Environmental Engineering (Singapore: McGraw-Hill, Inc). Retrieved from <u>https://iopscience.iop.org/article/10.1088/1755-1315/118/1/012019/pdf</u>

Saxena et. al. (2013). Phytochemistry of Medicinal Plants. Retrieved from https://www.researchgate.net/publication/284425734_Phytochemistry_of_Medicinal_Plants Shad et. al. (2014). Phytochemical and Biological Activities of Four Wild Medicinal Plants. Retrieved from <u>https://www.researchgate.net/publication/267932850 Biological Activities</u>

Schafer H, Wink M, 2009. Medicinally important secondary metabolites in recombinant microorganisms or plants: progress in alkaloid biosynthesis. Biotechnology Journal, 4(12): 1684- 1703. Retrieved from https://www.omicsonline.org/open-access/role-of-secondary-metabolites-in-defense-mechanisms-of-plants-0974-8369-3-128.pdf

https://www.globe.gov/documents/11865/ae27466a-9a69-4fd6-8244-ae1f7e22d844;

December 10, 2024

https://www.globe.gov/documents/11865/0fd40183-f2ea-480f-82b6-8d62180d9291; December 10, 2024

https://www.globe.gov/documents/11865/3464b426-6d54-4ba2-9cca-8d398fb38ef8; December 10, 2024

https://www.globe.gov/documents/11865/920675f5-56c0-46a3-97b5-74f9953b2ae4; December 10, 2024

Appendix 1

ANOVA (Analysis of Variance) for water temperature that was measured for 3 consecutive times in the brackish river of Khlong Tamru, Chonburi, Thailand.

Treatment \rightarrow	A	В	С
Input Data $ ightarrow$	23.1	20.4	27.4
	23.0	20.5	27.4
	23.1	20.5	27.5

source	sum of	degrees of	mean square	F statistic	p-value
	squares	freedom	MS		
treatment	74.3622	2	37.1811	11,154.3333	1.9439e-11
error	0.0200	6	0.0033		
total	74.3822	8			

Conclusion from ANOVA:

The p-value corresponding to the F-statistic of one-way ANOVA is **lower** than 0.05, suggesting that one or more treatments are significantly different. The Tukey HSD test multiple comparison tests follow. This post-hoc tests would likely identify which of the pairs of treatments are significantly different from each other.

Tukey HSD results for water temperature.

Treatments pair	Tukey HSD Q statistic	Tukey HSD p-value	Tukey HSD inference
A vs B	78.0000	0.0010053	** p<0.01
A vs C	131.0000	0.0010053	** p<0.01
B vs C	209.0000	0.0010053	** p<0.01

Appendix 2

ANOVA (Analysis of Variance) for dissolved oxygen that was measured for 3 consecutive times in the brackish river of Khlong Tamru, Chonburi, Thailand.

Treatment \rightarrow	A	В	С	
Input Data $ ightarrow$	2.02	2.9	1.2	tr
	2.11	2.86	1.2	
	2.03	2.89	1.2	

source	sum of	degrees of	mean square	F statistic	p-value
	squares	freedom	MS		
treatment	4.2507	2	2.1253	2,224.1977	2.4439e-09
error	0.0057	6	0.0010		
total	4.2564	8			

Conclusion from ANOVA:

The p-value corresponding to the F-statistic of one-way ANOVA is **lower** than 0.05, suggesting that one or more treatments are significantly different. The Tukey HSD test

multiple comparison tests follow. This post-hoc tests would likely identify which of the pairs of treatments are significantly different from each other.

Tukey HSD results for dissolved oxygen.

Treatments pair	Tukey HSD Q statistic	Tukey HSD p-value	Tukey HSD inference
A vs B	46.5062	0.0010053	** p<0.01
A vs C	47.8136	0.0010053	** p<0.01
B vs C	94.3198	0.0010053	** p<0.01

Appendix 3

ANOVA (Analysis of Variance) for total dissolved solids (TDS) that was measured for 3 consecutive times in the brackish river of Khlong Tamru, Chonburi, Thailand.

Treatment \rightarrow	A	В	С		source	sum of	degrees of	mean square	F statistic	p-value
Input Data \rightarrow	3212.0	3158.0	2991.0			squares	freedom	MS		
	3194.0	3140.0	2991.0	tr	reatment	70,622.0000	2	35,311.0000	490.4306	2.2474e-07
	3194.0	3140.0	2991.0		error	432.0000	6	72.0000		
					total	71,054.0000	8			

Conclusion from ANOVA:

г

The p-value corresponding to the F-statistic of one-way ANOVA is **lower** than 0.05, suggesting that one or more treatments are significantly different. The Tukey HSD test multiple comparison tests follow. This post-hoc tests would likely identify which of the pairs of treatments are significantly different from each other.

Tukey HSD results for total dissolved solids (TDS).

Treatments	Tukey HSD	Tukey HSD	Tukey HSD
pair	Q statistic	p-value	inference
A vs B	11.0227	0.0010053	** p<0.01
A vs C	42.6619	0.0010053	** p<0.01
B vs C	31.6392	0.0010053	** p<0.01

Appendix 4

ANOVA (Analysis of Variance) for electrical conductivity that was measured for 3 consecutive times in the brackish river of Khlong Tamru, Chonburi, Thailand.

Treatment \rightarrow	A	В	С	source	sum of	degrees of	mean square	F statistic	p-value
Input Data →	6352.0	5951.0	6179.0		squares	freedom	MS		
I	6352.0	6179.0	6179.0	treatment	97,144.2222	2	48,572.1111	9.1377	0.0151
	6316.0	6145.0	6145.0	error	31,893.3333	6	5,315.5556		
				total	129,037.5556	8			

Conclusion from ANOVA:

The p-value corresponding to the F-statistic of one-way ANOVA is **lower** than 0.05, suggesting that one or more treatments are significantly different. The Tukey HSD test multiple comparison tests follow. This post-hoc tests would likely identify which of the pairs of treatments are significantly different from each other.

Tukey HSD results for total electrical conductivity.

Treatments pair	Tukey HSD Q statistic	Tukey HSD p-value	Tukey HSD
pan	Q statistic	p-value	interence
A vs B	5.8996	0.0137979	* p<0.05
A vs C	4.0941	0.0617209	insignificant
B vs C	1.8055	0.4577910	insignificant

Appendix 5

ANOVA (Analysis of Variance) for transparency that was measured for 3 consecutive times in the brackish river of Khlong Tamru, Chonburi, Thailand.

Treatment \rightarrow	A	В	С
Input Data $ ightarrow$	0.31	0.2	0.17
	0.25	0.24	0.17
	0.28	0.22	0.17

source	sum of	degrees of	mean square	F statistic	p-value
	squares	freedom	MS		
treatment	0.0182	2	0.0091	21.0000	0.0020
error	0.0026	6	0.0004		
total	0.0208	8			

Conclusion from ANOVA:

The p-value corresponding to the F-statistic of one-way ANOVA is **lower** than 0.05, suggesting that one or more treatments are significantly different. The Tukey HSD test multiple comparison tests follow. This post-hoc tests would likely identify which of the pairs of treatments are significantly different from each other.

Tukey HSD results for transparency.

Treatments	Tukey HSD	Tukey HSD	Tukey HSD
pair	Q statistic	p-value	inference
A vs B	4.9923	0.0285241	* p<0.05
A vs C	9.1526	0.0015627	** p<0.01
B vs C	4.1603	0.0582234	insignificant

Appendix 6

ANOVA (Analysis of Variance) for water pH that was measured for 3 consecutive times in the brackish river of Khlong Tamru, Chonburi, Thailand.

Treatment \rightarrow	A	В	С
Input Data $ ightarrow$	7.52	7.93	7.9
	7.52	8.07	8.0
	7.63	8.26	8.05

source	sum of	degrees of	mean square	F statistic	p-value
	squares SS	freedom	MS		
treatment	0.4736	2	0.2368	19.0465	0.0025
error	0.0746	6	0.0124		
total	0.5482	8			

Conclusion from ANOVA:

The p-value corresponding to the F-statistic of one-way ANOVA is **lower** than 0.05, suggesting that one or more treatments are significantly different. The Tukey HSD test multiple comparison tests follow. This post-hoc tests would likely identify which of the pairs of treatments are significantly different from each other.

Tukey HSD results for water pH.

treatments	Tukey HSD	Tukey HSD	Tukey HSD
pair	Q statistic	p-value	inference
A vs B	8.2327	0.0027190	** p<0.01
A vs C	6.6276	0.0080047	** p<0.01
B vs C	1.6051	0.5302001	insignificant

Appendix 7

ANOVA (Analysis of Variance) for salinity that was measured for 3 consecutive times in the brackish river of Khlong Tamru, Chonburi, Thailand.

Treatment \rightarrow	А	В	С
Input Data $ ightarrow$	30.0	35.0	29.0
	30.0	35.0	29.0
	30.0	35.0	29.0

source	sum of	degrees of	mean square	F statistic	p-value
	squares	freedom	MS		
treatment	62.0000	2	31.0000	inf	0.0000e+00
error	0.0000	6	0.0000		
total	62.0000	8			

Conclusion from ANOVA:

The p-value corresponding to the F-statistic of one-way ANOVA is **lower** than 0.05, suggesting that one or more treatments are significantly different. The Tukey HSD test multiple comparison tests follow. This post-hoc tests would likely identify which of the pairs of treatments are significantly different from each other.

Tukey HSD results for salinity.

Treatments	Tukey HSD	Tukey HSD	Tukey HSD
pair	Q statistic	p-value	inference
A vs B	inf	0.0010053	** p<0.01
A vs C	inf	0.0010053	** p<0.01
B vs C	inf	0.0010053	** p<0.01

Appendix 8

ANOVA (Analysis of Variance) for relative humidity that was measured for 3 consecutive times in the brackish river of Khlong Tamru, Chonburi, Thailand.

Treatm	ent	А	В	с	source	sum of	degrees of	mean square	F statistic	p-value
\rightarrow						squares	freedom			
Input	Data	34.0	45.0	58.0	treatment	846.8889	2	423.4444	1,905.5000	3.8841e-09
\rightarrow		34.0	44.0	58.0	error	1.3333	6	0.2222		
		35.0	44.0	58.0	total	848.2222	8			

Conclusion from ANOVA:

The p-value corresponding to the F-statistic of one-way ANOVA is **lower** than 0.05, suggesting that one or more treatments are significantly different. The Tukey HSD test

multiple comparison tests follow. This post-hoc tests would likely identify which of the pairs of treatments are significantly different from each other.

Treatments	Tukey HSD	Tukey HSD	Tukey HSD
pair	Q statistic	p-value	inference
A vs B	36.7423	0.0010053	** p<0.01
A vs C	86.9569	0.0010053	** p<0.01
B vs C	50.2145	0.0010053	** p<0.01

Tukey HSD results for relative humidity.

Appendix 9

ANOVA (Analysis of Variance) for air temperature that was measured for 3 consecutive times in the brackish river of Khlong Tamru, Chonburi, Thailand.

Treatment \rightarrow	А	В	С
Input Data $ ightarrow$	29.8	26.3	30.9
	30.4	26.8	30.9
	30.3	27.0	30.9

source	sum of	degrees of	mean square	F statistic	p-value
	squares SS	freedom	MS		
treatment	30.1956	2	15.0978	194.1143	3.5254e-06
error	0.4667	6	0.0778		
total	30.6622	8			

Conclusion from ANOVA:

The p-value corresponding to the F-statistic of one-way ANOVA is **lower** than 0.05, suggesting that one or more treatments are significantly different. The Tukey HSD test multiple comparison tests follow. This post-hoc tests would likely identify which of the pairs of treatments are significantly different from each other.

Tukey HSD results for air temperature.

Treatments	Tukey HSD	Tukey HSD	Tukey HSD
pair	Q statistic	p-value	inference
A vs B	21.5300	0.0010053	** p<0.01
A vs C	4.5544	0.0413047	* p<0.05
B vs C	26.0845	0.0010053	** p<0.01

Appendix 10

ANOVA (Analysis of Variance) for soil pH that was measured for 3 consecutive times in the brackish river of Khlong Tamru, Chonburi, Thailand.

Treatment \rightarrow	А	В	С
Input Data $ ightarrow$	6.0	5.2	7.0
	5.5	6.4	7.0
	5.0	5.6	7.0

source	sum of	degrees of	mean square	F statistic	p-value
	squares	freedom	MS		
treatment	3.9089	2	1.9544	9.4064	0.0141
error	1.2467	6	0.2078		
total	5.1556	8			

Conclusion from ANOVA:

The p-value corresponding to the F-statistic of one-way ANOVA is **lower** than 0.05, suggesting that one or more treatments are significantly different. The Tukey HSD test multiple comparison tests follow. This post-hoc tests would likely identify which of the pairs of treatments are significantly different from each other.

Tukey HSD results for soil pH.

treatments	Tukey HSD	Tukey HSD	Tukey HSD
pair	Q statistic	p-value	inference
A vs B	0.8866	0.7979472	insignificant
A vs C	5.6997	0.0161143	* p<0.05
B vs C	4.8131	0.0331393	* p<0.05

Appendix 11

ANOVA (Analysis of Variance) for soil temperature (5cm) that was measured for 3 consecutive times in the brackish river of Khlong Tamru, Chonburi, Thailand.

Treatment \rightarrow	А	В	С
Input Data $ ightarrow$	25.0	23.0	29.0
	25.0	22.0	26.0
	25.0	21.0	26.0

-					
source	sum of	degrees of	mean square	F statistic	p-value
	squares	freedom	MS		
treatment	38.0000	2	19.0000	14.2500	0.0053
error	8.0000	6	1.3333		
total	46.0000	8			

Conclusion from ANOVA:

The p-value corresponding to the F-statistic of one-way ANOVA is **lower** than 0.05, suggesting that one or more treatments are significantly different. The Tukey HSD test

multiple comparison tests follow. This post-hoc tests would likely identify which of the pairs of treatments are significantly different from each other.

Tukey HSD results for soil temperature (5cm).

Treatments	Tukey HSD	Tukey HSD	Tukey HSD
pair	Q statistic	p-value	inference
A vs B	4.5000	0.0432875	* p<0.05
A vs C	3.0000	0.1650747	insignificant
B vs C	7.5000	0.0043631	** p<0.01

Appendix 12

ANOVA (Analysis of Variance) for soil temperature (10cm) that was measured for 3 consecutive times in the brackish river of Khlong Tamru, Chonburi, Thailand.

Treatment \rightarrow	А	В	С
Input Data $ ightarrow$	26.0	26.0	29.0
	26.0	22.0	26.0
	26.0	21.0	27.0

source	sum of	degrees of	mean square	F statistic	p-value
	squares	freedom	MS		
treatment	29.5556	2	14.7778	4.7500	0.0580
error	18.6667	6	3.1111		
total	48.2222	8			

Conclusion from ANOVA:

The p-value corresponding to the F-statistic of one-way ANOVA is **higher** than 0.05, suggesting that the treatments are not significantly different for that level of significance. The Tukey HSD test multiple comparison tests follow. This post-hoc tests would likely identify which of the pairs of treatments are significantly different from each other.

Tukey HSD results for soil temperature (10cm).

Treatments	Tukey HSD	Tukey HSD	Tukey HSD
pair	Q statistic	p-value	inference
A vs B	2.9459	0.1733142	insignificant
A vs C	1.3093	0.6404332	insignificant
B vs C	4.2552	0.0535661	insignificant