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# Assessment of Species Composition and Seasonal Variation of Phytoplankton in Headwater Streams, Phetchabun Province, Northern Thailand

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#### ABSTRACT

This article aimed to study the species compositions, distribution of phytoplankton, and water quality in the headwater streams of Phetchabun Province. The samplings were collected from 4 stations during the rainy season (September 2022), the winter season (December 2022), and the summer season (March 2023). A total of 35 phytoplankton species were found, with diatoms being the dominant group in both species and quantity across all 3 seasons. In the rainy season, the average phytoplankton density was 226,595 units/m<sup>3</sup>, with the highest density at ST1, Upper Huai Nam Jang. The dominant species found included Phacus quinquemarginatus and Melosira varians. In the winter season, the average phytoplankton density was 5,537,563 units/m<sup>3</sup>, with the highest density at ST1, Upper Huai Nam Jang. The dominant species found included Surirella robusta. In the summer season, the average phytoplankton density was 1,874,367 units/m<sup>3</sup>, with the highest density at ST4, Huai Ban Tok. The dominant species found included Eunotia pectinalis and Surirella robusta. The phytoplankton found at all stations throughout the study included Oscillatoria sp., Synedra ulna and Gyrosigma sp. The values of the diversity index, evenness index, and richness index ranged from 0.99 to 2.14, 0.39 to 0.98, and 0.45 to 0.96, respectively. The concentrations of ammonia-nitrogen, nitrate-nitrogen, and orthophosphate-phosphorus in the water ranged from 0.4 to 0.6mg/ L, 0.02 to 1.70 mg/L, and 0.16 to 0.83 mg/L, respectively. The study results can be used as data for assessing and monitoring water quality, as well as evaluating primary productivity and aquatic animal yield in headwater streams, which serve as indicators of the ecological richness in headwater streams of Phetchabun Province.

### INTRODUCTION

Phetchabun Province is located in the lower northern region of Thailand. The terrain is mostly highlands and complex mountains, making it the source of several important streams and rivers. The headwater areas encompass forest reserves, national









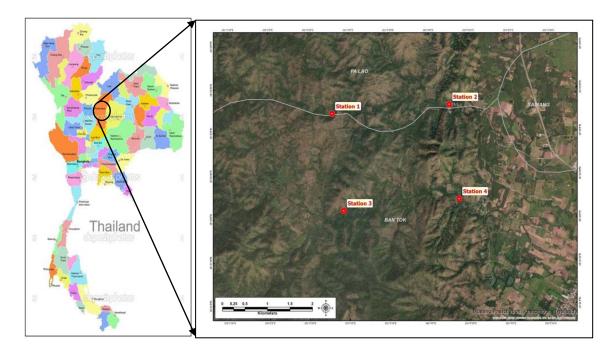
parks, and community agricultural lands. These headwater streams play a crucial ecological role, serving as habitats for native fish species and important aquatic life production sources. They are also a primary water source for community consumption, agriculture, and eco-tourism. However, currently, the headwater streams in Phetchabun Province face degradation due to forest encroachment, deforestation, monoculture agricultural land use, especially the cultivation of economic crops on sloped land (Ngamngon et al., 2022), as well as wildfires and haze during the dry season. This has led to soil erosion and flash floods, where organic substances, chemicals, or various toxins are washed into the rivers, particularly during the rainy season. These factors affect water quality, aquatic animals, and living organisms in the water, leading to changes in the community structure of organisms within the headwater streams, especially phytoplankton, which are the foundation of the food web in freshwater ecosystems.

Phytoplankton are small organisms that float in water bodies. They play a crucial role in aquatic ecosystems as primary producers in the food chain. Since phytoplankton can photosynthesize and produce organic matter, they serve as the main source of energy and food for zooplankton, small aquatic animals, and other aquatic organisms (Capuzzo et al., 2020). The type, quantity, and changes in phytoplankton can indicate the water quality of a water body in various aspects, such as fertility, nutrient deficiency conditions, and can also serve as indicators of pollution or excessive organic matter in the water. Therefore, phytoplankton are important factors that determine the growth and survival rates of aquatic animals. If phytoplankton change, this will inevitably affect the consumers in the next trophic level in the aquatic ecosystem. The study of the composition and seasonal distribution of phytoplankton in the headwater streams of Phetchabun Province aimed to assess water quality parameters, species composition, and seasonal variation of phytoplankton, as well as their relationship with water quality.

### **MATERIALS AND METHODS**

### Study area

Sampling points were established in the headwater stream, which is an aquatic ecosystem in Phetchabun Province, according to the upstream areas with different environmental conditions as follows: Station 1 (ST1), Upper Huai Nam Jang, Mueang District, Phetchabun Province, coordinates 47Q 0718579 E, 1812838 N UTM. Station 2(ST2), Middle Huai Nam Jang, Mueang District, Phetchabun Province, coordinates 47Q 0721181 E, 1813040 N UTM. Station 3 (ST3), Klong Ban Tok, Mueang District, Phetchabun Province, coordinates 47Q 0718841 E, 1810767 N UTM. Finally, Station 4 (ST4), Huai Ban Tok, Mueang District, Phetchabun Province, coordinates 47Q 0721400 E, 1811026 N UTM (Fig. 1).



**Fig. 1.** Location of study site and sampling stations in headwater streams, Phetchabun Province

## Study of water quality in headwater streams, Phetchabun Province

Water quality parameters were measured, and water samples were collected in 4 headwater stream areas of Phetchabun Province, coinciding with the collection of phytoplankton samples, with 3 repetitions per area conducted in September 2022 (rainy season), December 2022 (winter season), and March 2023 (summer season). Water temperature, pH, and dissolved oxygen (DO) were measured at the midpoint of the water source's width and mid-depth at the survey points using a YSI 556 MPS multi-parameter water quality meter. Water samples of 1,000 mL were collected using a vertical-type water sampler at the midpoint of the water source's width and mid-depth at the survey points, stored in plastic bottles, and all samples were placed in a cooling box for laboratory analysis. Nutrient levels in the water were measured for ammonia by the distillation nesslerization method, nitrate by the cadmium reduction method, orthophosphate by the Ascorbic acid method, and water turbidity by the nephelometric method (APHA et al., 2017). For water quality data analysis, the average values of each month's water quality parameters were calculated, and the statistical differences in the average water quality parameters between the sampling months were analyzed using the analysis of variance (ANOVA). Differences between means were assessed using Tukey's honestly significant difference (HSD) test at a 95% confidence level with SPSS for Windows version 23.0.

# Study of the composition and distribution of phytoplankton in headwater streams, Phetchabun Province

Phytoplankton samples were collected from the same 4 areas where water samples were collected, with 3 replicates per area, using plankton nets with a mesh size of 20 microns to filter 20L of water samples at a depth of approximately 30-50cm from the water surface in September 2022 (rainy season), December 2022 (winter season), and March 2023 (summer season). The filtered water samples were stored in plastic bottles and preserved with neutral formaldehyde at a final concentration of 2% before being taken back for analysis to identify species down to the lowest taxonomic rank at the Faculty of Science, Ramkhamhaeng University, Bangkok.

## Classification of phytoplankton

Phytoplankton species were classified by verifying taxonomic accuracy according to Wongrat (2001), Chatmongkolkul and Chantangsee (2005) and Pirapornpisan (2015). Additionally, using the online database AlgaeBase (Guiry & Guiry, 2014). The number of phytoplanktons was counted in a laboratory using the Natural Unit Count method with a high-power compound microscope, sampling 3 replicates per station, and calculating the phytoplankton density. Then, the data were analyzed to calculate the diversity index according to the Shannon-Wiener's diversity index formula (Hurlbert, 1971), the evenness index according to the Pielou's index formula (Washington, 1984), and the species richness index according to the Margalef's index formula (Clarke & Warwick, 1994) using the Primer 5 program.

# Study of the relationship between water quality and phytoplankton density in the headwater streams of Phetchabun Province

The average density of phytoplankton and water quality in each season were analyzed, and the Pearson's correlation coefficient between the average density of all phytoplankton and water quality was calculated.

### RESULTS

# The physical and chemical water quality in the headwater streams of Phetchabun Province

The water quality in the headwater streams of Phetchabun Province is described as follows:

# **Temperature**

Significant differences in temperature were noted, with the highest temperature of  $28.83\pm3.65$ °C observed in the summer season. Other seasons recorded temperature values of  $25.66\pm0.85$ °C and  $23.25\pm1.08$ °C.

### pН

The highest pH of  $8.28\pm0.05$  was found in the rainy season, significantly different from the summer season. The pH for other seasons was  $8.23\pm0.13$  and  $7.80\pm0.20$ .

# The dissolved oxygen (DO)

No significant differences in DO were observed among different seasons. The average DO range was from  $3.97\pm2.14$  mg/L to  $5.48\pm0.49$  mg/L.

# The turbidity

The highest turbidity of  $63.25\pm23.63$  NTU, was found in the rainy season, significantly different from other seasons. The turbidity for other seasons was  $15.6\pm13.69$  NTU, and  $4.15\pm3.3$  NTU. As for nutrient concentrations in the water sources, ammonia-nitrogen, nitrate-nitrogen, and orthophosphate-phosphorus, no significant differences in nutrient concentrations were observed among different seasons. The average nutrient concentrations ranged from  $0.32\pm0.02$  mg/L to  $0.46\pm0.10$  mg/L,  $0.08\pm0.03$  mg/L to  $0.59\pm0.96$  mg/L, and  $0.34\pm0.16$  mg/L to  $0.69\pm0.15$  mg/L, respectively (Table 1).

**Table 1.** Average seasonal water quality in the headwater streams of Phetchabun Province from September 2022 to March 2023

Water quality parameter	September 2022	December 2022	March 2023	<i>P</i> -value
Temperature (°C)	25.66±0.85	23.25±1.08	28.83±3.65	0.020*
pН	$8.28 \pm 0.05$	$8.23 \pm 0.13$	$7.80\pm0.20$	0.003**
Dissolved oxygen (mg/L)	$5.48 \pm 0.49$	$4.18\pm0.80$	$3.97 \pm 2.14$	0.244
Turbinity (NTU)	$63.25\pm23.63$	$4.15\pm3.3$	15.6±13.69	0.002**
Total ammonia (mg/L as nitrogen)	$0.36\pm0.03$	$0.32\pm0.02$	$0.46\pm0.10$	0.064
Nitrate (mg/L as nitrogen)	$0.21 \pm 0.12$	$0.08\pm0.03$	$0.59\pm0.96$	0.414
Orthophosphate (mg/L as phosphorus)	0.59±0.16	0.69±0.15	0.34±0.16	0.051

Mean ± standard deviation

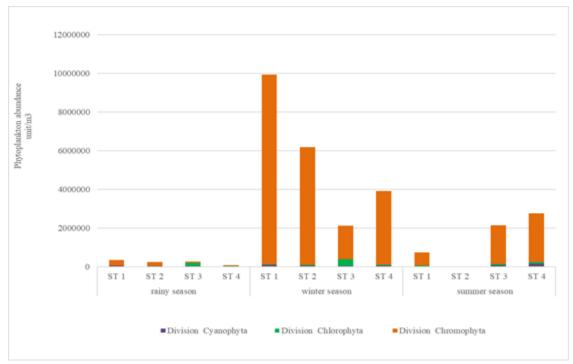
# Composition of species and the distribution of phytoplankton in the headwater streams of Phetchabun Province

From the study of phytoplankton composition in headwater streams, which is a running water ecosystem, a total of 35 species from 32 genera were found. These included 3 species of blue-green algae (Division Cyanophyta), accounting for 8.57%, 12 species of green algae (Division Chlorophyta), accounting for 34.29%, and 20 species of brownish-golden algae (Division Chromophyta), accounting for 57.14% (19 species of diatoms; 95.00% and 1 species of dinoflagellates; 5.00%). The average density of phytoplankton was the highest in the winter season (December 2022), followed by the summer season (March 2023) and the rainy season (September 2022), respectively. The phytoplankton species found at all stations in all 3 seasons included the blue-green algae *Oscillatoria* sp., as well as the diatoms *Synedra ulna* and *Gyrosigma* sp.

When considering the composition and density of phytoplankton by season, it was found that during the rainy season, a total of 15 species from 15 genera of phytoplankton were observed. These included 2 species of blue-green algae, 3 species of green algae, and 10 species of golden-brown algae (9 species of diatoms and 1 species of dinoflagellates). The average density of phytoplankton was 226,595 units/m<sup>3</sup>, with the highest density recorded at ST1, Upper Huai Nam Jang (Fig. 2). The dominant phytoplankton species found were *Phacus quinquemarginatus* and *Melosira varians*, accounting for 34.71% of the total phytoplankton (Table 2). The phytoplankton species found at all stations, which was additionally observed during the rainy season, included *Peridinium* sp. The winter season, a total of 18 species from 17 genera of phytoplankton were observed, including 2 species of blue-green algae, 3 species of green algae, and 13 species of brown algae (12 species of diatoms and 1 species of dinoflagellates). The average density of phytoplankton was 5,537,563 units/m<sup>3</sup>, with the highest density recorded at ST1, Upper Huai Nam Jang (Fig. 2). The dominant phytoplankton species found included Surirella robusta, which accounted for 60.14% of the total phytoplankton (Table 2). The phytoplankton species found at all stations, which was additionally observed during the winter season, included Cymbella tumida, Surirella robusta, and Peridinium sp. The summer season, a total of 22 species from 21 genera of phytoplankton were found, including 2 species of blue-green algae, 7 species of green algae, and 13 species of golden-brown algae (12 species of diatoms and 1 species of dinoflagellates). The average density of phytoplankton was 1,874,367 units/m³, with the highest density recorded at ST4, Huai Ban Tok (Fig. 2). The dominant phytoplankton species found were Eunotia pectinalis and Surirella robusta, which accounted for 40.85% of the total phytoplankton (Table 2). The phytoplankton species found at all stations, which was additionally observed during the summer season, included Gomphonema parvulum, Nitzschia sp., and Surirella robusta.

**Table 2.** Dominant species and the average amount of phytoplankton during the rainy season, winter season, and summer season in the headwater streams of Phetchabun Province from September 2022 to March 2023

Season	Dominant species	Average amount (unit/m³)	Total amount (%)	
Rainy	Phacus quinquemarginatus	85,050	20.80	
	Melosira varians	56,899	13.91	
Winter	Surirella robusta	3,575,813	60.14	
Summer	Eunotia pectinalis	586,575	22.76	
	Surirella robusta	466,233	18.09	



**Fig. 2.** Phytoplankton abundance in the headwater streams of Phetchabun Province from September 2022 to March 2023 (ST1: Upper Huai Nam Jang, ST2: Middle Huai Nam Jang, ST3: Klong Ban Tok and ST4: Huai Ban Tok; During the summer season, ST 2 there is no phytoplankton data because the riverbed is dry, making it impossible to collect samples)

The ecological index of phytoplankton shows that the diversity index ranges from 0.99 to 2.14, with the highest value during the rainy season at the Middle Huai Nam Jang (ST2) and the lowest value during the winter season at the Middle Huai Nam Jang (ST2). The evenness index ranges from 0.39 to 0.98, with the highest value during the rainy season at the Huai Ban Tok (ST4) and the lowest value during the winter

season at the Upper Huai Nam Jang (ST1). The richness index ranges from 0.45 to 0.96, with the highest value at the Upper Huai Nam Jang (ST1) in the summer season and the lowest value during the rainy season at the Huai Ban Tok (ST4) (Table 3).

**Table 3.** Univariate indices of phytoplankton in each area of the headwater streams of Phetchabun Province from September 2022 to March 2023

	Diversity index		Evenness index		Richness index				
Location	Rainy	Winter	Summer	Rainy	Winter	Summer	Rainy	WinterSummer	
	season	season	season	season	season	season	season	season	season
ST1	1.96	1.02	1.92	0.89	0.39	0.73	0.63	0.81	0.96
ST2	2.14	0.99	-	0.89	0.40	-	0.81	0.70	-
ST3	1.33	1.74	1.82	0.69	0.73	0.71	0.48	0.69	0.82
ST4	1.75	1.36	2.01	0.98	0.59	0.81	0.45	0.59	0.74
mean	1.80	1.28	1.92	0.86	0.53	0.75	0.59	0.70	0.84

**Remark**: ST1: Upper Huai Nam Jang, ST2: Middle Huai Nam Jang, ST3: Klong Ban Tok and ST4: Huai Ban Tok

# The relationship between water quality and phytoplankton density in the headwater streams of Phetchabun Province

The relationship between the average density of all phytoplankton and the water quality factors in the headwater streams of Phetchabun Province during the rainy season (September 2022), the winter season (December 2022) and the summer season (March 2023) showed that the average density of all phytoplankton had a highly significant negative correlation (r=-0.624) with water turbidity (P<0.05). For other water quality factors, there was no statistically significant correlation (P>0.05) (Table 4).

### **DISCUSSION**

The water quality in the headwater stream of Phetchabun Province, when considered seasonally, shows that the average temperature is highest during the summer season (P < 0.05). The water temperature changes according to the air temperature, which depends on the season. For pH, and turbidity, the average values are highest during the rainy season (P < 0.05). The DO value also reaches its peak during the rainy season (P > 0.05). Regarding the average concentration of nutrients in the water source, the highest values are observed during the summer season. Except for the orthophosphate-phosphorus concentration, which is highest during the winter season (P > 0.05). This may be because the water temperature in the summer season is suitable for the decomposition

<sup>: -</sup> no data (because during the summer, ST2 in the riverbed dried up, making it impossible to collect phytoplankton samples.)

of organic matter into nutrients (**Khan & Siddique, 1971**). Additionally, the amount of nitrate-nitrogen in the summer season positively correlates with the amount of ammonia-nitrogen. From this study, it was found that the concentration of orthophosphate-phosphorus in the water ranged from 0.16 to 0.83mg/ L, which is higher than the values found in general water sources that range from 0.01 to 0.1mg/ L (**Pitakphon** *et al.*, **2014**). In every season, the highest concentration of orthophosphate-phosphorus was found at ST3, Klong Ban Tok. Generally, the nutrient concentration in upstream streams is lower than in downstream areas. However, since these areas have agricultural activities such as rubber plantations, sweet tamarind fields, and cornfields, when rainwater washes the soil into the water source, it increases orthophosphate-phosphorus levels. The high concentration of orthophosphate-phosphorus in the headwater stream of the study area indicates a deteriorating environmental condition.

The composition of phytoplankton in the headwater stream revealed that goldenbrown algae were the most abundant, with 20 species accounting for 57.14%. Among these, 19 species were diatoms, out of a total of 35 species and 32 genera of phytoplankton. Following these were green algae and blue-green algae, respectively. Diatoms were the main group of phytoplankton in both species and quantity because they can adapt to cling to surfaces and withstand being swept away by currents and turbidity better. This is consistent with the studies of Waiyaka (1998) and Khunpradid (2000), who surveyed phytoplankton in the Mae Sa Stream, Doi Suthep-Pui National Park, Chiang Mai Province, and found that diatoms were the most abundant component. In this study, 35 species of phytoplankton were found, which is relatively low. In headwater streams, which are running water ecosystems, the types and quantities of phytoplankton are less than in standing water ecosystems such as lakes or reservoirs, as well as in large rivers. Additionally, in headwater stream areas, the quantity of phytoplankton is lower than in downstream areas (Nixdorf et al., 2018; Zeng et al., 2023). This is consistent with studies on phytoplankton diversity in the Xijiang River basin in southern China, which found that phytoplankton diversity decreases in headwater stream areas with increasing altitudes (Peng et al., 2024).

The distribution of phytoplankton in headwater streams is influenced by various factors such as topography, altitude, water flow characteristics, sunlight exposure, and the nature of riparian vegetation, which affect water temperature and nutrient levels (**Biggs** *et al.*, 2011). In this study, it was found that phytoplankton distribution varies by season and sampling site. During the rainy season, both the types and quantities of phytoplankton were at their lowest. The factors directly affecting distribution during the rainy season are physical factors, including water current speed, turbidity, and sunlight. In headwater streams, the fast-flowing water can cause phytoplankton quantities to fluctuate significantly depending on rainfall, which affects water current speed. Phytoplankton are quickly swept away, leaving little time for growth. Additionally, during the rainy season,

the water is highly turbid, reducing sunlight penetration. Sunlight is a crucial factor for phytoplankton photosynthesis; insufficient light decreases their growth rate, leading to a reduction in overall phytoplankton biomass, even with adequate nutrients (Basu & Pick, 1996). Furthermore, the phytoplankton found during the rainy season are more likely to come from other sources rather than being directly grown in the stream (Devercelli et al., **2018).** This study found a strong negative correlation (r=-0.624, P<0.05) between turbidity and the average density of all phytoplankton. During the winter seasons and summer seasons, the water flow in the upstream is slow or stagnant. Some areas of the river exhibit characteristics of pools, leading to the sedimentation of nutrients in the water. Additionally, the clear water allows phytoplankton sufficient time to grow, reproduce, and undergo photosynthesis effectively. This results in an increase in the diversity of phytoplankton species and an overall rise in phytoplankton biomass, particularly those requiring intense light and nutrients. These phytoplankton are often the types that can remain suspended in the water column (planktonic) (Nixdorf et al., 2018). Studies have shown that during the winter, the concentration of orthophosphatephosphorus in the water is at its highest, which correlates with the average density of the most abundant phytoplankton species. Orthophosphate is crucial for energy production (ATP) and essential cellular processes. When there is sufficient orthophosphate along with nitrate, it significantly accelerates photosynthesis and phytoplankton proliferation (Herawati et al., 2025). However, if the orthophosphate concentration in the water is too high, it can lead to eutrophication and algal blooms of certain phytoplankton species under suitable conditions. For the average density of phytoplankton, when considering the sampling stations, it was found that ST3, Klong Ban Tok, had the lowest average phytoplankton density. Although the orthophosphate-phosphorus content in the water source in this area was the highest, it can be explained by the general characteristics of the two banks of the river in the area, which are covered with large trees along the entire river. The shade from the trees covering the stream reduced the amount of light reaching the water surface, resulting in less sunlight reaching the water surface. Since light is a crucial limiting factor for the photosynthesis of phytoplankton, the overall density of phytoplankton decreased accordingly. Additionally, the shade also affected the water temperature, reducing heat accumulation in the water source. This cooler air also reduced the growth of some types of phytoplankton (**Devercelli** et al., 2018).

The dominant phytoplankton found in the headwater streams of Phetchabun Province are mostly diatoms. During the rainy season, the dominant phytoplankton include *Phacus quinquemarginatus* and *Melosira varians*. While in the winter season, the dominant phytoplankton include *Surirella robusta*. Whereas. in the summer season, the dominant phytoplankton include *Eunotia pectinalis* and *Surirella robusta*. The phytoplankton found at all stations throughout the study include *Oscillatoria* sp, *Synedra ulna* and *Gyrosigma* sp. This is consistent with the study by **Teunissen et al. (2014)**, who found that several genera of diatoms, such as *Achnanthidium*, *Gomphonema*, *Cymbella*,

and *Navicula*, are frequently found and often dominant in headwater streams because they can adhere well to surfaces or withstand high water flow. This study during the rainy season found *Phacus quinquemarginatus* to be the dominant species at ST3, Klong Ban Tok, indicating significant changes in water quality, particularly the increase of organic matter and nutrients such as nitrogen and phosphorus in the water source. Water quality data showed that orthophosphate-phosphorus levels in the stream were high throughout the study period. Various species of the genus *Phacus* often thrive in water bodies with high organic matter content or in semi-polluted to moderately polluted conditions (**Luo** *et al.*, 2017), which may be due to the runoff of organic substances from agricultural areas into the water source. Therefore, the dominance of this phytoplankton species indicates the initial stages of eutrophication in the stream.

The ecological index of phytoplankton shows that the diversity index, evenness index, and species richness index have average values ranging from 1.28 to 1.92, 0.53 to 0.86, and 0.59 to 0.84, respectively. These values are similar to the study by **Zeng** *et al.* (2023), which examined the upstream of the Pearl River in China and found that the average diversity index and evenness index of phytoplankton ranged from 1.32 to 1.51 and 0.83 to 0.87, respectively. The diversity index of living organisms ranged from 1 to 3, indicating that the water source is still suitable for the survival of living organisms (**Tudorance** *et al.*, 1975). This study suggests that the headwater stream in Phetchabun Province has conditions suitable for the survival of phytoplankton and aquatic organisms at a moderate level.

### **CONCLUSION**

The diversity of phytoplankton in the headwater streams of Phetchabun Province was found to include 35 species across 32 genera, with diatoms being the main group both in terms of species and quantity. Phytoplankton has the highest average density during the winter season (P< 0.05). The average density of all phytoplankton had a highly significant negative association with water turbidity. Increased water turbidity is a significant ecological indicator that signals environmental problems and the quality of freshwater sources. In terms of nutrients in the water source, the orthophosphate-phosphorus levels were found to be higher than in typical natural water sources that range from 0.01 to 0.1 mg/L, which may be the main cause of eutrophication. This could lead to the degradation of the water source, so water quality and aquatic organism monitoring should be conducted every season throughout the year. This will provide a comprehensive understanding of water body dynamics, allow for the assessment of ecosystem health, and enable the timely identification of potential problems. For guidelines for on reducing runoff from highland agricultural areas into water bodies focus on soil and water management to reduce topsoil and chemical erosion. This includes

planting cover crops and contour planting, creating buffer zones between agricultural areas and water bodies, promoting reduced chemical use in agriculture, and proper fertilizer management.

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