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THE ROLE OF OTOLITHS AND SCALES AS BIOLOGICAL INDICATORS: EVALUATING LIFE QUALITY OF FISH IN FRESHWATER ENVIRONMENTS

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ABSTRACT

Fish quality of life is influenced by where they live. Changes in aquatic environment quality due to human activities have become a global problem. These changes, or runoff into the water, affect fish scales, including lepidont scales. In addition to scales, otoliths can also predict water quality. *Helostoma temminckii* lives in Sepuncung Lake. To understand the microstructural characteristics of scales and otoliths from freshwater and marine environments, identify the correlation between aquatic environment and fish quality of life, and develop a model to predict fish quality of life as a bioindicator of aquatic environment reflected in otolith and scale (lepidont) condition. We captured fish using bamboo traps and fish nets. Scales were taken between the dorsal fin and lateral line; the anterior end was cut, cleaned, stained using keratin dye, and observed using a microscope with 1000x magnification. Otoliths were collected and shaved with an otolith shaver and observed under a microscope. Results show that *H. temminckii* has a dense dark ring around the focus; farther from the focus is a light ring, and there were light lepidont abnormalities in every part of the scales. This means the fish experiences stress only during the early stages of life. As they grew, they found an environment with less stress or disturbance. Scales (lepidont) can be used as a parameter to assess fish life quality and aquatic environmental conditions without sacrificing the fish.

Keywords: Abnormalities, Anthropogenic, Aquatic environment, Bioindicator, Runoff.

1. INTRODUCTION

Changes in aquatic environmental quality from human activities, such as heavy metal pollution, agricultural runoff, and climate change, have become a global issue affecting aquatic ecosystems worldwide. Heavy metal pollution, such as lead (Pb), has been reported to affect fish health and biodiversity in Southeast Asian waters, including Indonesia.

Their environment influences fish quality of life. If the water is disturbed, it can threaten fish survival. Consequently, such disturbances will affect the quality of life of the fish. Water is crucial to fish survival and growth because it is their habitat. Polluted water hinders fish growth.

Mahboob et al.[1] states that introducing pollutants into a water body affects the growth patterns of organisms and has physiological effects on them. This can be assessed by examining fish physical condition. Two body parts of fish that can indicate water quality are scales and otoliths. Scales are believed to indicate water quality.

Fish scales are small, rigid plates that grow on the fish skin to protect their bodies. Masood et al.[2] states that fish scales are small, rigid plates that protect against certain diseases and also help fish move. Scales are the part of the fish that comes into direct contact with water; therefore, if pollutants are present in a body of water, scales are likely to bear the brunt of declining water

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quality or pollution. Viana et al.[3] states that scales are the part of the fish directly exposed to toxins and various contaminants. Thus, scales can accumulate contaminants and serve as a chemical fingerprint of the environmental pollution events the fish experience.

Changes or contaminants in the water affect fish scales, including lepidonts. Lepidonts are tooth-like structures that function as hooks, aiding movement and anchoring the scales to the fish's body. Khanna et al.[4] states that lepidonts are damaged when scales are directly exposed to pollutants, which can harm the fish. Therefore, fish scales are believed to serve as a basis for determining water quality. In addition to scales, otoliths are also believed to indicate the quality of a water body.

Otoliths are biomineral structures in the inner ear that function as balance organs and detect direction and sound in all teleost fish[5]. Otoliths are organs that correlate with fish growth. If a body of water is disturbed, it can affect fish growth, and these changes can be observed in the otoliths. Suyatna et al.[6] states that fish otolith size follows fish size and is positively correlated. Otoliths are parts of fish that do not come into direct contact with the water. Although they do not come into direct contact with water, fish otoliths are thought to reflect water quality. This is because poor water conditions affect fish growth.

Sepuncung Lake is an oxbow lake formed when the meandering Kampar River was cut off, creating a separate body of water[7]. Oil palm plantations surround the lake, and this may affect the waters of Lake Sepuncung. During the rainy season, the lake receives runoff from oil palm plantation activities; this runoff is believed to contain many pollutants such as pesticides, herbicides, and others that can enter the water.

Sepujung Lake is a freshwater ecosystem with high biodiversity potential. One of the species found in the lake is the tambakan (*Helostoma temminckii*). This fish is very popular in Riau. The tambakan is a freshwater species native to tropical regions, specifically Southeast Asia[8]. The lake's current conditions are believed to be affected by runoff from oil palm plantation activities, which may impact the fish's survival.

Fish scales can be used to measure annual growth patterns, and fish otoliths record the environmental history experienced by the fish throughout its lifetime. This study is highly relevant to sustainable fisheries management and the conservation of aquatic ecosystems. The results are expected to inform the development of more effective aquatic management policies, particularly in addressing the challenges of environmental pollution. Although several studies have investigated otoliths and scales as indicators of environmental quality, research integrating both indicators remains limited. Therefore, this study can fill this gap by providing insights into the relationship between the microstructure of otoliths and scales and environmental parameters in freshwater and marine environments.

2. RESEARCH METHOD

Time and Place

This study was conducted from March to June 2025 at Sepuncung Lake, Kampar Regency, and Dumai seawater, Dumai City.

Method

Fishermen captured fish using bamboo traps and fish nets, with a minimum of 30 fish/location. Scales were taken between the dorsal fin and lateral line; the anterior end was cut, scales were cleaned and put into alcohol, then were stained using a keratin dye for 15-30 minutes, and then the scales were washed in running water and observed using an Olympus CX 21 microscope with 1000x magnification.

Otoliths were processed following the Windarti and Simarmata[9] method. The water quality parameters observed were temperature, pH, dissolved oxygen, and heavy metal (Pb). Primary data were obtained through direct observation of the samples and qualitative and quantitative

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analysis. Data were analyzed using descriptive and inferential statistics, including the Pearson Correlation test in SPSS (Statistical Product and Service Solutions).

3. RESULT AND DISCUSSION

General Conditions

An oxbow lake forms in a meandering river when the main channel is cut off[7]. This lake forms through erosion and sedimentation over time. During the rainy season, water volume increases, and the oxbow reconnects with the main river, allowing water from the river to flow into the oxbow. During the dry season, the lake becomes disconnected from the main river, causing the lake's water volume to decrease. This phenomenon can lead to differences in the lake's ecosystem between the rainy and dry seasons.

Lake Sepuncung, located in Rantau Baru Village, is one of the oxbow lakes in Pelalawan Regency. Rantau Baru Village is in Pangkalan Kerinci District, the capital of Pelalawan Regency. Rantau Baru Village has three oxbow lakes: Lake Kayangan, Lake Teluk Benderas, and Lake Sepuncung. This lake covers an area of 6.58 ha, of which 3.52 ha is open water, and aquatic plants cover 3.08 ha.

Figure 1. General Conditions of Lake Sepuncung

Correlation between Length and Weight of Fish

The relationship between fish length and weight serves as a criterion for determining the quality of a water body. A healthy water body will support good fish growth, and vice versa. Fish living under environmental stress divert energy from growth to maintenance to survive. This hinders fish growth and disrupts the growth process. This aligns with Mahé et al.[10] statement that length, weight, and the quality of the aquatic environment in which fish reside are significantly related. Fish living in good water quality will exhibit healthy growth in both length and weight. The total length of the tambakan caught in Sepuncung Lake ranged from 158 to 240 mm, with a body weight ranging from 31.33 to 353.63 g. The relation between length and weight of *H. teminckii* is shown in Figure 2.

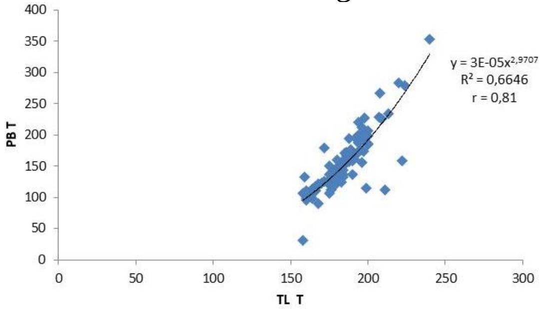


Figure 2. Relation between length and weight of *H. teminckii*

The regression equation for farmed fish in Lake Sepuncung is $y = 3E\text{-}05x^{2.97}$. The coefficient of determination (R^2) is 0.66. With a value of (b) = 2.97, the growth pattern of tambakan in Lake Sepuncung is isometric, where an increase in body length is proportional to an increase in body weight. Meanwhile, r = 0.81 indicates a strong relationship between body length and body weight. These findings align with research by Ahmadi[11], whose analysis of the relationship between body length and weight in *H. temminckii* in the Batang River yielded b = 2.90. This implies that this fish's growth pattern is negatively allometric but approaching isometric, meaning body length increases faster than body weight. These results are similar because the fish share nearly identical habitats rivers and oxbow lakes and because many factors, including species, feed, and aquatic environment, influence body shape.

The regression equation results with the b value illustrate the growth pattern (b = 2.97 for farmed fish in Lake Sepuncung).

These results likely reflect the study site's adequate habitat characteristics and food availability, yet heavy metal levels remain relatively high. This lead (Pb) pollution pressure can disrupt fish growth and food availability in aquatic environments. This can directly indicate environmental and physiological stress in fish, further strengthening their role as indicators. The growth patterns of organisms, particularly fish, depend on the environmental conditions in which they reside and the availability of food.

Otolith

Otoliths are fish organs that detect vibrations and function as balance organs. According to Lowensten[12], otoliths function as balance organs in fish. These organs are located in a cavity (sacculus) within the inner ear. According to Windarti & Simarmata[9], otoliths form through calcium carbonate deposits that accompany fish growth; as fish grow, the otoliths in the sacculus increase in size. The calcium carbonate deposition rate in otoliths varies and is generally related to fish growth. If the fish grows rapidly, calcium carbonate deposition around the otolith is less dense; if it grows slowly, calcium carbonate accumulation on the otolith is denser. Differences in calcium carbonate density create the light and dark ring patterns on the otolith.

Environmental conditions strongly influence dark and light growth ring patterns; in this study, Pb content had a particular effect. Extremely high Pb levels can disrupt calcium carbonate deposition, making it irregular and resulting in darker growth rings. Heavy metals can also affect otolith biomineralization, causing a denser and more irregular structure. Dark growth rings form when the growth rate slows, so otolith growth slows and calcium carbonate accumulation becomes denser[9].

Based on this study, the otoliths of *H. teminckii* range in length from 4.22-10.00 mm and in weight from 0.04-0.16 g. The sagittal otoliths of this fish are oval or elliptical, with a sharper front end and a rounded rear end. The edges of these otoliths are not completely symmetrical, with the back side being slightly more curved than the front. This oval or elliptical shape is a common characteristic of fish in the order Anabantiformes as well as some groups of freshwater fish that have compressed bodies and move relatively slowly (Figure 3). The growth rings in the otoliths of *H. teminckii* fish were shown in Figure 4.

Figure 3. *H. teminckii's* otolith

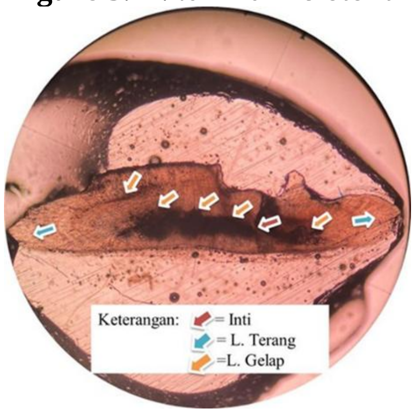


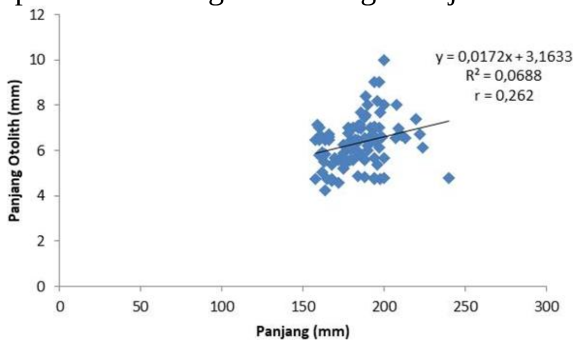
Figure 4. The growth rings in the otoliths of *H. teminckii*

This study only measured the thickness of the dark rings. This is because the number of dark rings in this otolith could not be counted, as the otolith has very dense dark growth rings. According to Windarti & Simarmata[9], dark growth rings form when fish have a slow growth rate, so otolith growth is slow, and calcium carbonate accumulation is dense. Light growth rings form when fish have a relatively fast growth rate, so the otolith grows rapidly, and calcium carbonate accumulates less densely.

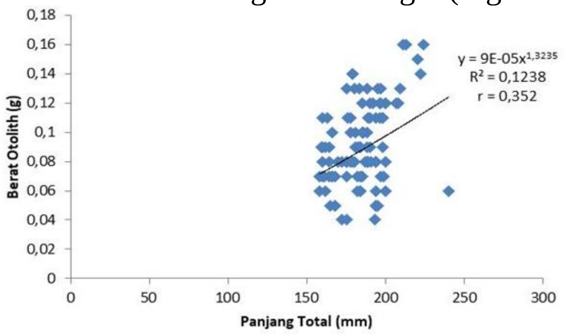
In this fish, nearly all of the otolith has the same pattern of dark rings. They are almost all located near the center and even cover the focus. The formation of dark rings near the Nucleus indicates that, during the early stages of their lives, these fish experienced stress and disturbances. As they grew, these fish moved into less stressful environments, so the farther they were from the nucleus, the lighter they became. The dark ring patterns indicate that heavy-metal exposure was highest during the juvenile phase, which then subsided. According to Sinaga et al.[13], the first dark ring near the center indicates that, during the early stages of fish growth, the fish experienced stress and disturbances in its life.

The Total Length and length & Weight of the otolith relation

Otolith measurements of *H. teminckii* include otolith length and weight (Figure 5).



a



b

Figure 5. The graph of the relation between total length and the length (a)-weight(b) of *H. temminckii*'s Otolith

Helostoma temminckii living in Sepunjung Lake follows y = 0,0172x + 3,1633, with a coefficient of determination (R^2) = 0,0688 and a correlation coefficient (r) = 0,262. For this fish, the correlation coefficient indicates a positive but weak relationship between total length and otolith length. It shows that otolith length increases as total length increases, but otolith length increases more slowly than total length. Campana[14] state that otolith growth is conditional, so size is not always proportional or linear with fish body length.

The *H. teminckii* that live in the Sepunjung Lake have y = 9E-05x1.3235 with a determination coefficient (R^2) = 0,1238, the correlation coefficient (r) = 0,352, and b = 1,3235. Similarly, the relationship between total length and otolith length is shown by the (r) values. This fish has a 12% correlation. The growth pattern of total length with weight was negative allometric (B<3). This means that the body length of this fish grows faster than the otolith weight. This is because otolith weight is influenced by the formation of dark rings, which cause calcium carbonate deposits on the otolith, making the rings increasingly dense. The longer a fish faces adversity in its life, the thicker its otolith becomes. According to Jawad & Mahe[15], otoliths are directly influenced by the environmental conditions where the fish live. If fish live in poor conditions, this affects the length and weight of their otoliths.

Fish scales are small, rigid plates that grow on the skin of animals, particularly fish, and serve to protect the body against certain diseases as well as support the fish's movement. Each scale consists of several key components. The focus is the central point, which is the first part of the scale. Circuli are surface features that typically form a continuous line but are often interrupted by grooves. Radii are grooves that extend outward toward the edge of the scale, and at the posterior edge, there is an additional structure called a cteni[16].

One of the most important parts of a scale is the lepidont. The lepidont is a tooth-like structure that functions as a hook, aiding the fish's movement and anchoring the scales to the fish's body. Khanna et al.[4] states that the lepidont is located in the inner cavity of the scale's circuli. This tooth-like structure is arranged at regular intervals and functions as a hook, aiding the fish's movement and anchoring the scales to the fish's body. The image showing the lepidont structure of the scales of this fish can be seen in Figure 6.

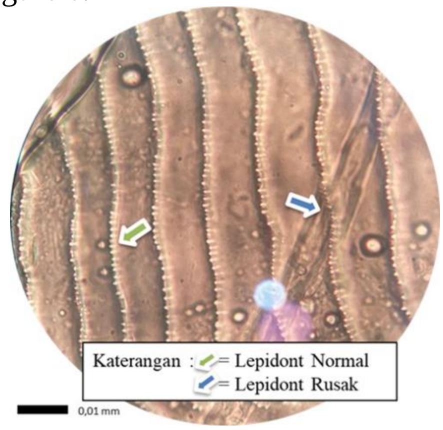


Figure 6. Lepidont of *H. teminckii*

Based on scale observations, this fish has ctenoid scales. Ctenoid scales are serrated along their outside edges and are typically found on fish with spikey fins. These scales have a stenii structure at the posterior end and a comb-like structure at the anterior end. Physically, ctenoid scales are oval or round in shape, with sizes varying depending on the fish species. Ctenoid scales are usually smaller than cycloid scales but larger than ganoid scales. Ctenoid scales are located on the fish's skin, covering the entire body and situated beneath the mucus.

Physiologically, Pb can damage fish scales by accumulating in hard tissues, impairing mineralization, and disrupting the supporting scale tissue. According to Dai et al.[17], Pb disrupts hard tissues by weakening their structure and interfering with mineralization in bones and scales.

Scales are among the organs that interact directly with water. Therefore, if the water contains very high levels of heavy metals (Pb), this can cause changes in the annuli, and heavy metal accumulation can damage the scales. High levels of heavy metals (Pb) can cause damage to the circuli and lepidons. Damage increases as heavy-metal concentration rises. This can deform the calcium/chitin in the scales, leading to brittleness or cracking of the lepidons. Sun et al.[18] reported severe deformities and damage in scales collected from waters with high heavy metal levels. Heavy metals accumulate and affect scales more than other organs, as scales are directly exposed to environmental conditions.

The scales of *H. teminckii* living in Lake Sepunjung waters are still in good condition, although they show some minor abnormalities (Figure 6). This is because the aquatic environment of Lake Sepunjung is still in good condition, but lepidonts show minor abnormalities due to heavy metals that can affect scale condition. These heavy metals are estimated to come from runoff from palm oil plantations located around the lake.

Heavy metal pollution causes these abnormalities. This damages the scale structure. Heavy metal pollution causes these abnormalities. This damages the scale structure, particularly in lepidonts. This indicates that fish scales can indicate pollution, particularly heavy metal contamination, in the water. This aligns with Azab et al.[16] statement that abnormalities in fish scales, particularly the lepidont, can indicate that the waters where the fish live contain heavy metals and may endanger human life.

The Relationship between Otolith and Scale Parameters with Water Quality

Otoliths are organs within a fish's body formed by calcium carbonate deposits that accumulate as the fish grows. Although otoliths do not come into direct contact with water, they are significantly influenced by the environment in which the fish lives. Poor aquatic conditions affect fish growth, which in turn impacts calcium carbonate deposition in the otoliths. If fish growth is disrupted, calcium carbonate accumulates in the otoliths, resulting in dark rings; the opposite is also true. These deposition variations create dark and light rings. This aligns with Campana & Thorrold[19] statement that the surrounding environment influences otolith growth. Additionally, otoliths can serve as a "fingerprint" to distinguish a fish's origin whether from a healthy or polluted environment. Thus, the relationship between otoliths and water quality is very strong.

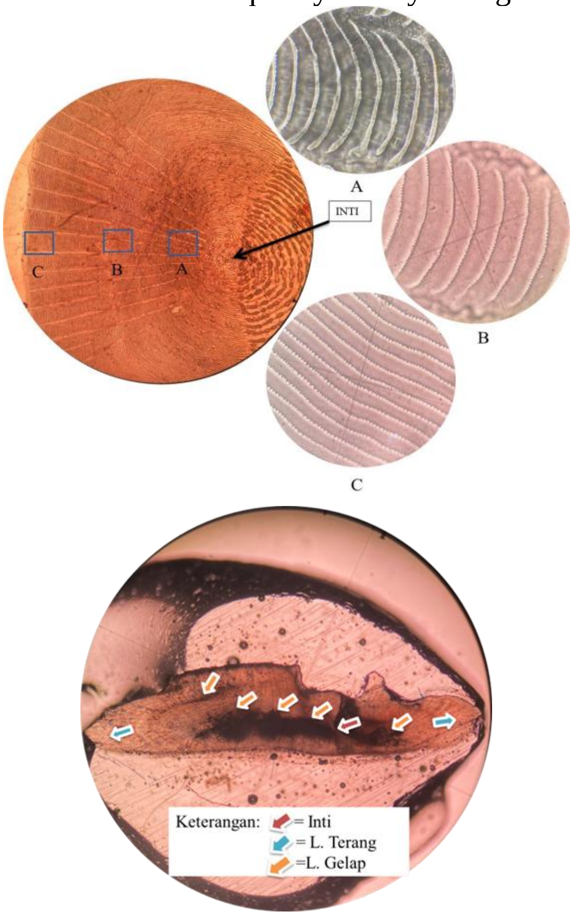


Figure 7. The relation between scales and otoliths of *H. temminckii*

Scales are the outermost part of a fish's body. Because they are in direct contact with water, the aquatic environment affects the condition of the fish's scales. If the aquatic environment is polluted, it will negatively affect the fish, especially their scales. Scales exposed to pollutants may develop abnormalities. One part of the scale that is often affected is the lepidont. The lepidont is the tooth-shaped part of a fish's scale. If water quality is poor or the water is polluted, the lepidonts will exhibit abnormalities. Pollutants may weaken bonding strength, causing lepidonts to break or become loose. This aligns with Khanna et al.[4] statement that fish exposed to pollution will experience changes in their scales, particularly in the lepidonts. Pollution-induced abnormalities in lepidonts damage the scale structure, loosening scales through damage or breakage of the lepidonts (Figure 7).

This study shows a relationship between otoliths and scales. This relationship can be used to describe the environmental conditions in freshwater. Otoliths start growing from the core and continue to the next stage, forming a circular pattern. This is consistent with scale growth, which also starts from a focus and continues until the scale is fully formed. Lippitsch[20] states that scales begin to grow from the focus and continue throughout the fish's lifetime; the surface pattern, typically linear and following the scale's contour, usually consists of continuous lines but is often interrupted by radius-shaped grooves.

According to this study, otoliths and scales have similar shapes that can indicate the water conditions where the fish live. In freshwater, most otoliths of *H. teminckii* have a thick dark ring around the focus, and the farther from the core, the lighter the ring becomes. This is consistent with the growth of this fish's scales, where the scales exhibit lepidont damage around the focus, but the lepidont condition improves farther from the focus. This indicates the fish experienced stress early in life, which decreased as it grew. In the Stepwise correlation test (Pearson Correlation), the correlation coefficient (r) was 0.66. This indicates a significant relationship between dark rings and scale damage.

This research found a relationship between otoliths, scales, and environmental quality, particularly regarding heavy metals (Pb). This relationship may also indicate heavy-metal (Pb) status in both freshwater and marine environments. In farmed fish from Lake Sepunjung, there is a thick dark ring surrounding the nucleus, and the further away from the nucleus, the lighter the ring becomes. This is consistent with the scales of these fish, where the scales show lepidothelial damage only around the core, but the lepidothelial layer improves as one moves farther from the core. In the Pearson correlation test examining the relationship between the percentage of dark rings in the otoliths and Pb concentration, the p-value was 0.001 ($p < 0.05$). This indicates a strong relationship between otolith damage and Pb concentration.

Regarding the relationship between lepidont damage on the scales and Pb concentration, the P-value was 0.61 ($P > 0.05$). This indicates a weak relationship between lepidont damage and Pb concentration. This is because fish are exposed to Pb only at the beginning of their life cycle, and this exposure decreases as they grow.

In *H. teminckii* fish in Sepunjung Lake, otoliths and scales correlate with each other and with environmental conditions, especially heavy metals (Pb). This indicates that otoliths and scales can reflect the condition of the aquatic environment where the fish live. Both organs can serve as bioindicators of aquatic-environment conditions, particularly lepidonts in scales. Thus, scales can serve as bioindicators of aquatic-environment condition without sacrificing the individual fish being studied.

Scales are the part of a fish that is directly affected by the water. If heavy metal (Pb) accumulates in the water at a dangerous level, it will cause direct damage to the scales. Scale damage can be observed immediately, making it possible to assess aquatic environmental conditions without sacrificing the fish. Thus, fish scales can be used as bioindicators of aquatic environmental conditions without sacrificing the fish. Heavy metal concentrations influence accumulation and effects on the scales. Therefore, scales can serve as bioindicators of heavy metal pollution, a major contributor to environmental contamination[21,23].

Water Quality

Water is the habitat for aquatic life, so water quality must be maintained. Kasry et al.[24] states that, biologically and ecologically, the presence of aquatic organisms in water bodies, particularly fish, significantly impacts water quality. Water quality includes physical, chemical, and biological aspects. The water's self-purification capacity significantly influences its ability to tolerate a certain amount of pollutants. This aligns with Kordi & Tancung[25], who state that poor-quality water, or water contaminated by pollutants that disrupt the aquatic environment, can cause disease or abnormalities in aquatic organisms, especially fish.

Table 1. Lake Sepunjung water quality

Water Quality	Parameter	Water quality results (Monthly)			Water quality standard*
		1	2	3	
pH		5	5	5	6-9
Temperature	^{O}C	29	30	29	28-30
DO	mg/L	4,9	5,1	5	4
Pb	ppm	0,038	0,14	0,12	0,03

pH is an index of hydrogen ion concentration that affects aquatic organisms. In this study, the pH value remained relatively constant across all sampling points and did not change in Lake Sepunjung, remaining at 5. This indicates that the water conditions in Lake Sepunjung are acidic but still normal, as Lake Sepunjung is a lake located on peatland. This is in accordance with Arifin et al.[26] statement that freshwater fish have an acid critical point at pH 5.0 and a base critical point at pH 9.0. The pH conditions in these waters remain tolerable for fish and other aquatic organisms; therefore, the pH levels found in this study do not significantly affect the organisms living there. The pH in these waters still supports the survival of aquatic organisms and is not a major disturbing factor. Highly alkaline or highly acidic water conditions can endanger organism survival and disrupt metabolic and respiratory processes[27]. Water temperature is one of the most important factors for aquatic organisms. Temperature significantly affects aquatic life and growth. Water temperature is influenced by season, latitude, time of day, air circulation, cloud cover, and water flow and depth.

In this study, water temperatures in Lake Sepunjung ranged from 29 to 30 °C. The water temperatures recorded were relatively consistent from one month to the next. This indicates that the lake's water temperatures are within normal ranges and do not disrupt the lives of the organisms in the lake.

Water temperature in this area remains within normal ranges and meets quality standards, so it does not affect the organisms living in the water. This also indicates that temperature conditions in this area continue to support aquatic life and do not pose a major threat. Effendi[28] states that significant changes in temperature affect the metabolic rate of aquatic organisms.

Dissolved oxygen is necessary for aquatic organisms to respire. Oxygen is also required to oxidize organic and inorganic substances. The primary sources of oxygen in a body of water are diffusion from the atmosphere and photosynthesis by organisms living in that body of water[29].

Dissolved oxygen levels in this study were still relatively normal. In Lake Sepunjung, the levels ranged from 4.26 to 4.76. These values indicate that dissolved oxygen conditions in the lake remain normal, meaning oxygen levels are still sufficient to support the survival of aquatic life. According to Government Regulation No. 22 of 2021 (Class III), the dissolved oxygen quality standard is 4 mg/L. Although slightly above the standard, the oxygen levels are still suitable for aquatic life and do not threaten its sustainability.

Heavy metals are chemicals that can cause significant problems for aquatic environments. Household activities, agriculture, fisheries, ports, and industrial operations contribute to increased levels of heavy metals in water bodies. Lead (Pb) is one heavy metal particularly hazardous to aquatic environments. Anthropogenic activities around coastal areas can generate waste that enters the ocean through runoff or river flow.

The concentration of heavy metals (Pb) in the waters of Sepunjung Lake ranges from 0.038 to 0.14 mg/L. This indicates that Pb accumulation in these waters is poor because it exceeds the water quality standard set by Government Regulation No. 22 of 2021, which is 0.03 mg/L. Oil palm plantations surround Sepunjung Lake, which influences water quality. The lake receives runoff from oil palm plantation activities, which contains pollutants such as pesticides, herbicides, and other substances that can enter the water. This is reflected in very mild damage to the scales and growth ring patterns on the otoliths, indicating recovery (light rings are at the edges) and fish growth approaching isometric.

4. CONCLUSION

Tambakan (*H.temminckii*) from Lake Sepunjung showed mild otolith abnormalities and a more normal otolith pattern (dark rings only around the focus). Among the observed environmental parameters (temperature, pH, dissolved oxygen, and Pb), only the concentration of the heavy metal (Pb) showed a significant effect (correlation) on fish health, as reflected in changes in the microstructure of otoliths and scales (lepidonts). Fish scales (lepidonts) can be recommended as a bioindicator parameter for predicting water quality. Abnormalities in lepidonts correlate with pollution levels (particularly Pb) and allow evaluation of fish and environmental quality without sacrificing the organism.

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