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IDENTIFICATION OF FUNGAL INFECTION IN KISSING GOURAMI (*Helostoma temminckii*) CULTURED IN SERUYAN REGENCY, CENTRAL KALIMANTAN, INDONESIA

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ABSTRACT

The kissing gourami (*Helostoma temminckii*) is an economically valuable freshwater fish species with increasing potential for aquaculture development. However, fungal infection remains an important health problem in its culture system. This study aimed to identify the fungal agent infecting cultured kissing gourami and to determine the prevalence and intensity of infection in aquaculture ponds in Seruyan Regency, Central Kalimantan, Indonesia. The study was conducted for 30 days, with fish sampling performed every five days across six observation periods. Fungal examination used mucus samples collected from the body surface, fins, and gills. The fungal agent was identified based on macroscopic and microscopic characteristics. The results showed that the fungal agent identified in infected kissing gourami was *Saprolegnia* sp. The prevalence of infection ranged from 0.7–6.0% , based on 150 fish examined during each sampling period. The lowest prevalence occurred on day 10, while the highest prevalence occurred on day 30. The intensity of infection ranged from 2–7 colonies/fish, indicating very low to low infection intensity. Microscopically, *Saprolegnia* sp. was characterized by long, branched, aseptate hyphae. Clinically, infected fish showed cotton-like white patches, fin erosion, and ulcerative lesions on the body surface. These findings indicate that *Saprolegnia* sp. is an opportunistic fungal pathogen in cultured kissing gourami. Therefore, routine fish health monitoring and proper water quality management are essential to minimize the risk of saprolegniasis in aquaculture ponds.

KEYWORDS: *fungal; kissing gourami; macroscopic; microscopic; Saprolegnia* sp.

ABSTRAK

Ikan biawan (*Helostoma temminckii*) merupakan salah satu ikan air tawar bernilai ekonomis yang memiliki potensi besar untuk dikembangkan dalam kegiatan budidaya. Namun, infeksi jamur masih menjadi salah satu permasalahan kesehatan yang penting pada sistem budidayanya. Penelitian ini bertujuan untuk mengidentifikasi agen jamur yang menginfeksi ikan biawan budidaya serta menentukan prevalensi dan intensitas infeksi pada kolam budidaya di Kabupaten Seruyan, Kalimantan Tengah, Indonesia. Penelitian ini dilakukan selama 30 hari dengan pengambilan sampel ikan setiap 5 hari sekali pada 6 periode pengamatan. Pemeriksaan jamur dilakukan menggunakan sampel lendir yang diambil dari permukaan tubuh, sirip, dan insang ikan. Identifikasi jamur dilakukan berdasarkan karakteristik makroskopis dan mikroskopis. Hasil penelitian menunjukkan bahwa agen jamur yang teridentifikasi pada ikan biawan yang terinfeksi adalah *Saprolegnia* sp. Prevalensi infeksi berkisar antara 0,7–6,0%, berdasarkan 150 ekor ikan yang diperiksa pada setiap periode pengambilan sampel. Prevalensi terendah ditemukan pada hari ke-10, sedangkan prevalensi tertinggi ditemukan pada hari ke-30. Intensitas infeksi berkisar antara 2–7 koloni/ekor yang termasuk dalam kategori intensitas sangat rendah hingga rendah. Secara mikroskopis, *Saprolegnia* sp. ditandai oleh hifa yang panjang, bercabang, dan tidak bersekat. Secara klinis, ikan yang terinfeksi menunjukkan bercak putih menyerupai kapas, erosi sirip, dan luka ulseratif pada permukaan tubuh. Hasil penelitian ini menunjukkan bahwa *Saprolegnia* sp. merupakan patogen jamur oportunistik pada ikan biawan budidaya. Oleh karena itu, pemantauan kesehatan ikan secara rutin dan pengelolaan kualitas air yang baik perlu dilakukan untuk mengurangi risiko terjadinya saprolegniasis pada kolam budidaya.

KATA KUNCI: Ikan biawan; jamur; makroskopis; mikroskopis; *Saprolegnia* sp.

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INTRODUCTIONS

The kissing gourami (*Helostoma temminckii*) is a native freshwater fish species widely distributed across Southeast Asia, including the swamp ecosystems of Kalimantan (Vidthayanon, 2012; Ahmadi, 2021). In Indonesia, this species is locally known as biawan and is recognized for its ecological relevance and economic importance. Its flesh and roe are highly valued by local communities, which contributing to its considerable market demand and relatively high commercial value.

The increasing market demand for kissing gourami has intensified fishing pressure on wild populations. Excessive harvesting, particularly of broodfish and roe-bearing individuals, may reduce the availability of fry and juveniles in natural waters (Hasan *et al.*, 2016). Consequently, aquaculture development has become an important approach to support production while reducing pressure on natural stocks. Previous studies have documented advances in spawning, hatchery techniques, and grow-out practices for kissing gourami (Hasan *et al.*, 2016; Lestari *et al.*, 2024). Nevertheless, the success of aquaculture development is closely linked to effective fish health management.

Disease remains a major constraint in freshwater aquaculture and may cause substantial production losses. Fungal infection is among the common health problems in cultured fish, particularly under stressful conditions associated with poor water quality, high organic matter accumulation, and environmental instability (Abdel-Latif *et al.*, 2020; Rokhmani *et al.*, 2017). Saprolegniasis is one of the most frequently reported fungal diseases in freshwater fish and is commonly associated with *Saprolegnia* spp. This infection primarily affects the external body surface, fins, and gills, and is clinically characterized by cotton-like white to grayish mycelial growths (Barde *et al.*, 2020; Anjum *et al.*, 2023; Ravindra *et al.*, 2022). In severe cases, the infection may lead to tissue damage, impaired growth, and mortality in cultured fish (Lindholm-Lehto & Pylkkö, 2024).

Despite the growing body of research on kissing gourami culture techniques, information regarding fungal infection in this species remains limited, particularly in swamp-based aquaculture systems in Kalimantan. Baseline data on fungal agents, prevalence, and infection intensity in cultured kissing gourami from Seruyan Regency, Central Kalimantan, remain scarce. Therefore, this study aimed to identify the fungal agent infecting cultured kissing gourami and to determine the prevalence and intensity of infection under field culture conditions. The findings are expected to provide essential baseline information for fish health monitoring and disease management in kissing gourami aquaculture.

MATERIALS AND METHODS

Time and Location of Study

This study was conducted over a 30-day period, from July 9 to August 7, 2025. Fish samples were collected from aquaculture ponds at Seruyan Polytechnic, Central Kalimantan, Indonesia. Fungal identification and laboratory analyses were conducted at Aquaculture Laboratory of Seruyan Polytechnic, Central Kalimantan.

Fish Sampling Method

Sampling of kissing gourami (*Helostoma temminckii*) was conducted across 6 sampling periods at five-day intervals. The initial fish population in the culture pond consisted of 150 individuals, and only one pond was used as the experimental unit. During each sampling period, 5 fish were randomly collected from those exhibiting clinical signs of fungal infection, resulting in a total of 30 fish samples examined throughout the study. Fish sampling was carried out using a purposive-random sampling approach. Fish exhibiting clinical symptoms, such as skin lesions, body discoloration, or cotton-like growths on the skin, fins, and gills, were first identified and selected. Subsequently, fish were randomly sampled from this symptomatic group for further laboratory examination. During the study period, water quality parameters were measured in situ, including temperature, pH, and dissolved oxygen (DO), using a multiparameter (Hanna HI 98194).

Fish Examination Method

Fungal isolates were identified using conventional methods consisting of 2 stages: macroscopic and microscopic observation. Macroscopic examination was performed by observing the physical characteristics of fungal colonies associated with visible signs of fungal infection or external abnormalities on the body surface, fins, and gills of fish. Subsequently, a mucus scraping procedure was performed by gently collecting mucus samples from the skin, fins, and gills using a sterile scalpel. The collected mucus samples were placed on a glass slide, mixed with sterile distilled water, and covered with a coverslip for microscopic examination. Observations were performed using a light microscope (Sinher XSZ-107, China) at 160× magnification to identify fungal hyphae and spores (Fradina *et al.*, 2023).

Fungal Prevalence

Fungal identification was conducted based on the identification guidelines described by Fajar *et al.* (2024). The data obtained from observations were

Table 1. Criteria for the prevalence of fungal infections

Prevalence	Category	Description
100 – 99	Always	Very severe infection
98 – 90	Almost always	Severe infection
89 – 70	Common	Moderate infection
69 – 50	Frequent	Very frequent infection
49 – 30	Generally observed	Common infection
29 – 10	Often	Frequent infection
9 – 1	Occasionally	Occasional infection
< 1 – 0.1	Rare	Rare infection
< 0.1 – 0.01	Very rare	Very rare infection
< 0.01	Almost never	No infection observed

subsequently analyzed to determine the prevalence and intensity of fungal infections. Identification of fungal species was initially carried out by comparing the morphological characteristics of the observed fungi with the reference descriptions provided in the identification guide. Fungal prevalence was calculated by dividing the number of infected fish by the total number of fish examined during each sampling period and multiplying the result by 100%, in order to determine the level of infection spread within the observed fish population. Thus, the denominator used in the prevalence calculation refers to the number of fish observed at each sampling event, rather than the total pond population. The prevalence formula used in this study followed the method described by Fajar *et al.* (2024).

$$Prevalence(\%) = \frac{Number\ of\ inf\ ected\ fish}{Total\ number\ of\ fish\ examined\ during\ each\ sampling\ periode} \times 100$$

Fungal Infection Intensity

Infection intensity was determined by calculating the total number of fungal colonies and the number of infected fish. The calculation formula used was according to Fajar *et al.* (2024):

$$Infection\ intensity(\%) = \frac{Total\ number\ of\ fungal\ colonies}{Number\ of\ infected\ fish} \times 10$$

Data Analysis

Observation data were tabulated using Microsoft Excel. Data analysis was conducted descriptively, and the results were presented in the form of tables and graphs. The analysis also included observations of clinical signs in fish through both macroscopic and microscopic identification methods, supported by photographic documentation obtained during the examination process.

RESULTS AND DISCUSSIONS

Fish Mortality

The mortality data recorded over the 30-day culture period showed a fluctuating pattern. Initial mortality was recorded at 0%, then increased to 1.3% on day 1 and reached 3.3% on day 5. Mortality subsequently decreased to 0.7% on day 10, but increased again to 2.7% on day 15 and remained at the same level until day 20. On day 25, mortality decreased to 1.3%, before sharply increasing to 6% on day 30. This fluctuation indicates unstable mortality dynamics, with an increasing trend toward the end of the observation period.

The progression of fungal infection in kissing gourami occurred gradually throughout the observation period. The highest mortality rate was recorded on day 30, reaching 6%, indicating that fungal infection developed progressively and required time to induce severe pathological effects and mortality in the fish population. This condition is likely associated with the opportunistic nature of *Saprolegnia* sp., which commonly infects fish experiencing stress or prior tissue damage. During the early stage of infection, the fungus colonizes the

Table 2. Criteria for Fungal Infection Intensity

Intensity (number of fungi/fish)	Category
1–5	Very low
6–15	Low
16–50	Moderate
51–100	High
>100	Very high

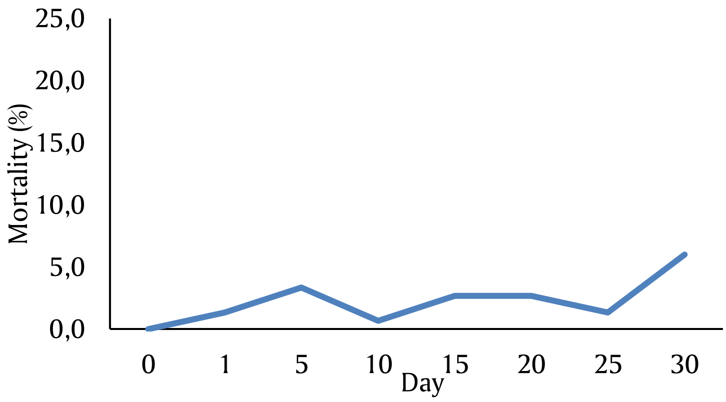

Figure 1. Results of observations on fish mortality

epidermal surface; however, over time the hyphae penetrate deeper tissues, causing skin damage, disruption of osmoregulation, deterioration of physiological condition, and eventually mortality. Lindholm Lehto and Pylkkö (2024) explained that the development of saprolegniasis occurs gradually, beginning with the appearance of mycelial patches and progressing into extensive cotton-like colonies covering the fish body surface.

Furthermore, Juniati *et al.* (2015) reported that Nile tilapia experimentally infected with *Saprolegnia* sp. zoospores exhibited abnormal behavioral responses. The fish showed reduced swimming activity, sluggish movement, and impaired swimming balance. Rahman *et al.* (2025) also reported that the characteristic clinical sign of saprolegniasis is the presence of white cotton-like mycelia on wounded or stressed tissues, accompanied by epithelial damage and physiological deterioration condition in spotted snakehead (*Channa punctata*). During the acute phase of infection, approximately 7–8 days post-infection, fungal colonies developed into cotton-like structures with circular, semicircular, or spiral growth patterns (Das *et al.*, 2012). This condition was subsequently followed by fish mortality within approximately 3–4 days.

The increase in mortality observed during the final phase of the study was also likely influenced by cumulative stress during the rearing period (Figure 1). Although water quality parameters remained within ranges generally suitable for fish survival, environmental fluctuations and the accumulation of organic matter in the culture pond may have induced chronic stress, thereby weakening the fish immune response. Such conditions facilitate the attachment and proliferation of *Saprolegnia* zoospores on damaged tissues or immunocompromised fish. El Gamal *et al.* (2023) stated that *Saprolegnia* infection is closely associated with fish immune status, where environmental stress can increase fish susceptibility to fungal colonization. Susilo and Yusanti (2022) reported that gourami infected with *Saprolegnia* sp. without treatment (control group) experienced mortality rates as high as 82.67%. Similarly, Ravindra *et al.* (2022) observed that mortality in infected fish groups began on day 2 post-infection, with cumulative mortality reaching 100% by day 4.

Prevalence

The prevalence of fungal infection during the 30-day observation period showed a fluctuating pattern, ranging from 0.7% to 6.0%, with all values categorized as "occasional infection". At the beginning of the observation period (day 5), fungal prevalence was recorded at 4.7%, followed by a sharp decline to 0.7% on day 10, representing the lowest prevalence observed during the study. Subsequently, prevalence increased again on days 15 and 20, each reaching 2.7%, before slightly decreasing to 1.3% on day 25. At the end of the observation period (day 30), prevalence increased markedly and reached the highest recorded value of 6.0%.

The prevalence values obtained throughout the study, ranging from 0.7% to 6.0%, were classified as "occasional infection". The relatively low prevalence indicates that fungal transmission within the fish population remained limited and did not develop into a severe outbreak. This low prevalence was likely influenced by relatively stable water quality conditions during the culture period. Dissolved oxygen concentrations ranged from 6.5 to 6.9 mg L^{-1}, water temperature ranged between 28–29°C, and pH values ranged from 6.04 to 6.58. These environmental conditions were still considered suitable for freshwater fish culture and were unlikely to induce severe physiological stress in the fish. However, the slightly acidic pH conditions may still have favored fungal growth. Nuryati *et al.* (2015) reported that fungi can grow optimally in acidic aquatic environments, particularly within a pH range of 3.8–5.6.

Table 3. Prevalence of fungal infection in kissing fish

Sampling time	Total number of sampling	Infected fish	Prevalence	Prevalence category
5	150	7	4.7	Occasional infection
10	150	1	0.7	Occasional infection
15	150	4	2.7	Occasional infection
20	150	4	2.7	Occasional infection
25	150	2	1.3	Occasional infection
30	150	9	6.0	Occasional infection

Infection Intensity

The number of *Saprolegnia* colonies detected in fish during the observation period ranged from 6 to 32 fungal colonies, with the number of infected fish ranging from 1 to 9 individuals. The infection intensity per fish varied between 2 and 7 colonies/fish. Most observations were classified within the very low infection category, whereas the low infection category was observed only on day 10 (6 colonies/fish) and day 25 (7 colonies/fish).

The infection intensity values ranging from 2 to 7 colonies/fish indicate that fungal colonization on the fish body remained at low to very low levels. This finding suggests that most fish were still capable of maintaining physiological resistance against fungal infection development. The relatively low infection intensity was likely associated with stable environmental conditions during the culture period, which may have limited extensive hyphal growth and fungal proliferation. These findings were lower than those reported by Ravindra *et al.* (2022), who observed that *Saprolegnia parasitica* infection in striped catfish resulted in mortality rates reaching 100% within several days after infection. These differences were likely influenced by fish species, fungal virulence, environmental conditions, and the stress level experienced by fish during culture.

The results of this study indicate that *Saprolegnia* sp. acts as an opportunistic pathogen capable of infecting kissing gourami under aquaculture conditions. The presence of physical injuries, stress conditions, and suboptimal environmental quality may have contributed to fungal infection development. Therefore, proper water quality management, routine fish health monitoring, and prevention of physical injuries during culture practices are essential measures to minimize the risk of saprolegniasis in kissing gourami aquaculture systems.

Microscopic Observation

Microscopic examination of *Saprolegnia* sp. revealed long, branched, aseptate hyphae with relatively thin and transparent hyphal walls. Under light microscopy, the hyphae appeared clear with varying diameters. Figure 2 shows the microscopic appearance of *Saprolegnia* sp., indicated by a red circle. Infection of kissing gourami (*Helostoma temminckii*) by *Saprolegnia* sp. was characterized by clinical signs, including white cotton-like patches on the body surface, fin erosion, scale detachment, and open skin lesions. Microscopically, the fungus exhibited long, branched, transparent, and aseptate hyphae, which are consistent with the typical morphological characteristics of *Saprolegnia* sp. These findings are consistent with reports by Anggani *et al.* (2015) and Kusdarwati *et al.* (2016), who described *Saprolegnia* sp. as possessing aseptate hyphal structures and forming cotton-like colonies on infected fish tissues. Based on these morphological characteristics, the fungus was identified as *Saprolegnia* sp.; however, species-level confirmation requires molecular identification techniques.

Clinical Signs

Kissing gourami (*Helostoma temminckii*) infected with *Saprolegnia* sp. exhibited external lesions that were

Table 4. Intensity of saprolegnia infection in kissing gourami

Sampling Time (Day)	Number of Saprolegnia sp.	Number of Infected Fish	Colony/fish	Category
5	15	7	2	Very low
10	6	1	6	Low
15	12	4	3	Very low
20	10	4	3	Very low
25	14	2	7	Low
30	32	9	4	Very low

Figure 2. Observation of *Saprolegnia* sp. under a microscope at 100X magnification

clearly visible, as shown in Figure 3. Ulcerative lesions were observed on the posterior body region, characterized by darkened areas accompanied by scale detachment at the infection site, fin erosion, and open wounds exposing the underlying tissue. Early lesions appeared as grayish-white, cotton-like patches attached to the skin, fins, and occasionally the head region.

As the mycelial growth expanded, the body surface became dull, covered with a thin fungal layer, and lost the shiny appearance typically observed in healthy fish. External tissue damage progressed as the infection developed, and affected areas eventually underwent localized necrosis, causing the skin surface to become damaged and easily slough off. In several cases, fungal colonies covered large portions of the body surface, interfering with fin movement and impairing normal swimming activity (Yasani *et al.*, 2024). These clinical manifestations are consistent with the typical characteristics of saprolegniasis in freshwater fish.

Water Quality

The results of water quality monitoring during the study are presented in Table 5. Dissolved oxygen concentrations remained relatively stable throughout the observation period, ranging from 6.5 to 6.9 mg

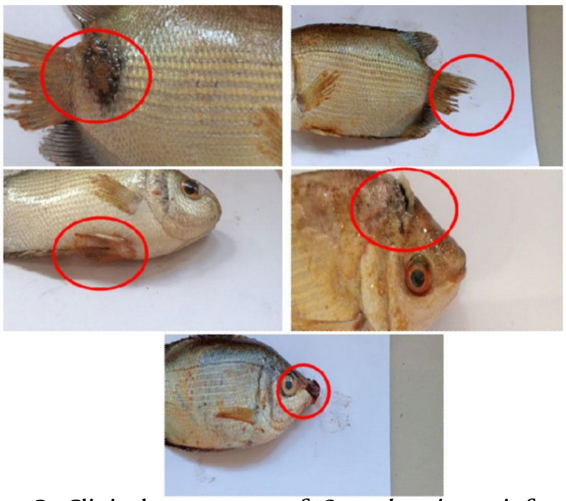

Figure 3. Clinical symptoms of *Saprolegnia* sp. infection

Table 5. Water quality during the observation period

Observation Time (Day)	Dissolved Oxygen (DO) (mg L^{-1})	Temperature (^{0}C)	pH
1	6.7	29	6.04
5	6.9	29	6.12
10	6.6	28	6.17
15	6.7	29	6.43
20	6.5	28	6.58
25	6.5	28	6.58
30	6.7	28	6.58

L^{-1}. Water temperature ranged between 28–29°C, while pH gradually increased from 6.0 to 6.6. Overall, these parameters remained within ranges generally considered suitable for the growth and survival of kissing gourami (*Helostoma temminckii*).

Environmental conditions may also influence the occurrence and development of fungal infections in aquatic organisms. Temperature and pH are among the factors reported to affect the growth and reproductive activity of *Saprolegnia* spp. Liu *et al.* (2017) reported that *Saprolegnia* sp. exhibits optimal growth at temperatures around 25°C. Although the temperature observed in the present study was slightly higher, it remained within the tolerance range reported for aquatic fungi.

Mahboub and Shaheen (2021) further stated that *Saprolegnia* spp. are capable of surviving under a broad range of environmental conditions, including temperatures of 5–40°C and pH levels of 2–11. Variations in pH and temperature may influence fungal development, including hyphal formation and zoospore activity. Therefore, while the water quality recorded during the study remained generally suitable for fish culture, these environmental conditions may also have allowed the persistence of *Saprolegnia* sp. in the culture environment.

CONCLUSIONS

This study demonstrated that kissing gourami (*Helostoma temminckii*) cultured in aquaculture ponds in Seruyan Regency, Central Kalimantan, were associated with *Saprolegnia* sp. infection Macroscopically, the infection was characterized by the appearance of white cotton-like patches, fin erosion, and lesions on the body surface. Microscopically, the fungus exhibited long, branched, and aseptate hyphae, which are morphological characteristics commonly associated with *Saprolegnia* sp. The prevalence of fungal infection ranged from 0.7% to 6.0%, categorized as occasional infection, while the infection intensity ranged from 2 to 7 colonies/fish, classified from very low to low infection intensity. These findings suggest that fungal infections observed in the culture system were associated with *Saprolegnia* sp. However, the identification in this study was limited to macroscopic and microscopic observations; therefore, species-level confirmation would require molecular analysis. Routine fish health monitoring and proper water quality management are essential to minimize the development and spread of fungal infections in aquaculture systems.

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AUTHOR CONTRIBUTIONS

BAT: Conceptualization, methodology, investigation, formal analysis, data curation, writing – original draft. M: Resources, investigation, data curation, visualization, supervision, validation, methodology, writing – review & editing. GAS: resources, investigation, data curation, visualization, writing – review & editing. YY: Supervision, validation, methodology, writing – review & editing. AK: Conceptualization, formal analysis, data curation, writing – original draft. All authors have read and approved the final version of the manuscript.

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