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The effectiveness of lempuyang gajah (*Zingiber zerumbet*) rhizome juice in reducing ectoparasite infections in mutiara catfish (*Clarias gariepinus*) fry

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ABSTRACT

Mutiara catfish cultivation is gaining popularity due to its fast growth, feed efficiency, and environmental resilience. However, ectoparasite infections such as *Trichodina sp*., *Gyrodactylus sp*., and *Dactylogyrus sp*. remain a major challenge, causing stress, inhibited growth, and potential mass mortality. The routine use of synthetic antibiotics to combat these parasites raises concerns about resistance and environmental impact, highlighting the need for safe, natural alternatives. This study evaluated the effectiveness of lempuyang gajah (*Zingiber zerumbet*) rhizome juice in reducing ectoparasite infections in Mutiara catfish fry. The experiment involved five treatment concentrations (0, 1, 5, 10, and 15 mL/L), each with four replications. Parameters observed included the number, prevalence, and intensity of ectoparasites. Results showed that lempuyang gajah rizome juice significantly reduced ectoparasite numbers, prevalence, and intensity, with the largest reduction observed at 15 mL/L. The decrease corresponds to increased levels of active compounds such as zerumbone, flavonoids, alkaloids, and saponins, known for their antiparasitic properties. In conclusion, lempuyang gajah rhizome juice is a promising natural and eco-friendly alternative for controlling ectoparasites in Mutiara catfish farming, offering potential solutions to issues of antibiotic resistance and food safety.

Kata kunci:
Intensitas
Parasit
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ABSTRAK

Efektivitas perasan rimpang lempuyang gajah (*Zingiber zerumbet*) dalam menurunkan infeksi ektoparasit pada benih ikan lele mutiara (*Clarias gariepinus*). Budidaya ikan lele mutiara semakin populer karena pertumbuhannya yang cepat, efisiensi pakan, dan ketahanan lingkungan. Namun, infeksi ektoparasit seperti *Trichodina sp*., *Gyrodactylus sp*., dan *Dactylogyrus sp*. masih menjadi tantangan besar, yang menyebabkan stres, pertumbuhan terhambat, dan potensi kematian massal. Penggunaan antibiotik sintetis secara rutin untuk memerangi parasit ini menimbulkan kekhawatiran tentang resistensi dan dampak lingkungan, yang menyoroti perlunya alternatif alami yang aman. Penelitian ini mengevaluasi efektivitas jus rimpang lempuyang gajah (*Zingiber zerumbet*) dalam mengurangi infeksi ektoparasit pada benih ikan lele mutiara. Percobaan melibatkan lima konsentrasi perlakuan (0, 1, 5, 10, dan 15 mL/L), masing-masing dengan empat ulangan. Parameter yang diamati meliputi jumlah, prevalensi, dan intensitas ektoparasit. Hasil penelitian menunjukkan bahwa jus rimpang lempuyang gajah secara signifikan mengurangi jumlah, prevalensi, dan intensitas ektoparasit dengan pengurangan terbesar diamati pada 15 mL/L. Penurunan ini sejalan dengan peningkatan kadar senyawa aktif seperti zerumbon, flavonoid, alkaloid, dan saponin, yang dikenal memiliki sifat antiparasit. Kesimpulannya, sari rimpang lempuyang gajah merupakan alternatif alami dan ramah lingkungan yang menjanjikan untuk mengendalikan ektoparasit pada budidaya lele Mutiara, menawarkan solusi potensial untuk masalah resistensi antibiotik dan keamanan pangan.

Introduction

Mutiara catfish is a distinct population of African catfish, morphologically indistinguishable from other populations such as the Paiton, Sangkuriang, Mesir, and Dumbo catfish (Iswanto *et al.*, 2015). The advantages of the Mutiara catfish include its resistance to disease and adverse environmental conditions, uniform body size, high growth rate, and efficient feed use (Ardyanti *et al*., 2018; Iswanto *et al*., 2016). Due to its high productivity, Mutiara catfish cultivation has become a promising cultivation method and is increasingly sought after by entrepreneurs (Masnita *et al*., 2024).

Despite their numerous advantages, catfish still face common challenges in cultivation, such as the emergence of disease and high mortality rates due to parasitic infections (Siegers *et al*., 2024). These parasites can be endoparasites, which attack the internal organs of the host, or ectoparasites, which attack the external organs of the host. The negative effects of ectoparasites are more obvious physically and can therefore be anticipated promptly (Andayani *et al*., 2024). Some types of ectoparasites frequently found in catfish include *Trichodina* sp. (Haris & Asran, 2015), *Gyrodactylus* sp. (Al Hasyimia *et al*., 2016; Dawo *et al*., 2023; Fachrussyah *et al*., 2024), and *Dactylogyrus* sp. (Al Hasyimia *et al*., 2016; Ekaputri *et al*., 2023; Fransira, 2023; Hastuti *et al*., 2020; Siegers *et al*., 2024). This ectoparasitic infection can weaken the fish's immune system, inhibit growth (Hastuti *et al*., 2020), and has the potential to cause mass mortality (Amelia *et al*., 2023; Fachrussyah *et al*., 2024; Haris & Asran, 2015).

Parasite infestations are generally managed through the administration of synthetic antibiotics (Dawood *et al*., 2021). However, excessive use of synthetic antibiotics risks triggering resistance (Erwiyani *et al*., 2023; Nurhasnawati *et al*., 2016). Furthermore, inappropriate use can also have negative impacts on the environment, fish health, and humans as end consumers in the food chain (Nafiqoh *et al*., 2020; Pelić *et al*., 2024; Shao *et al*., 2021).

The use of natural antibiotics is an alternative to address parasitic infestations in fish (Rosidah, 2022). Various natural ingredients have been studied for their effectiveness as antiparasitic agents, including a solution from the paci-paci plant (*Leucas lavandulaefolia*) at a concentration of 3 g/L, which has been shown to significantly reduce the number of *Trichodina* sp. (Haris & Asran, 2015). Furthermore, a mixture of guava leaves, kipahit, noni, papaya, and turmeric rhizome at a dose of 1.82 g/L also demonstrated effectiveness as an antiparasitic for catfish fry (Nafiqoh *et al*., 2020). Guava leaf infusion at a concentration of 10 ml/L has been reported to be effective in suppressing *Trichodina* sp. attacks on catfish fry (Santrianda & Aji, 2021). Ginger rhizome juice (*Zingiber officinale*) at a concentration of 15 mL/L has been shown to effectively reduce the number of ectoparasites in catfish (Nuriani & Suwartiningsih, 2021). Ethanol extract of neem leaves (*Melia azedarach*) has also been shown to be effective in controlling *Trichodina* sp. parasites in catfish due to its alkaloid, flavonoid, phenol, saponin, steroid, tannin, and terpenoid content (Tran & Tran, 2023).

Plants in the ginger family (*Zingiberaceae*) have great potential as antiparasitics because they contain active compounds such as gingerol (Hossain *et al*., 2019). One type of plant in the *Zingiberaceae* family is lempuyang gajah (*Zingiber zerumbet*). Lempuyang gajah extract contains various bioactive compounds such as alkaloids, flavonoids (especially kaempferol and its derivatives), phenolics (tannins), terpenoids (monoterpenoids, sesquiterpenoids, and steroids), vanillin, zerumbon, zederone, curcumin, and gingerol, which act as antimicrobials, strong antioxidants, anti-inflammatory, anticancer, and therapeutic agents against various infections and pathological disorders (Ramzan & Zeshan, 2023). The results of previous studies showed that lempuyang gajah extract combined with *Boesenbergia pandurate* and *Solanum ferox* was proven to have effective antiparasitic activity in controlling *Argulus* sp. in carp (*Cyprinus carpio*) (Hardi *et al*., 2022). Therefore, research is needed to determine the effectiveness of lempuyang gajah rhizome juice in controlling ectoparasites in Mutiara catfish. This research is crucial given the need for safe and environmentally friendly alternatives to the use of synthetic chemicals that can cause resistance, pollute the environment, and negatively impact human health as end consumers.

Method

This experimental study aimed to test the effectiveness of lempuyang gajah rhizome juice on ectoparasite infestations in Mutiara catfish fry. The experimental design consisted of five treatments of lempuyang gajah rhizome juice concentrations: 0 mL/L, 1 mL/L, 5 mL/L, 10 mL/L, and 15 mL/L, each with four replications, resulting in a total of 20 experimental units. Each experimental unit contained three one-month-old catfish fry. The research stages included making lempuyang gajah rhizome juice, acclimatizing the fry, ectoparasite infection, soaking in lempuyang gajah rhizome juice, and examining for ectoparasites.

The process of making lempuyang gajah rhizome juice began with washing the rhizomes with running water, rinsing with sterile distilled water, cutting, and weighing 500 grams. The rhizome pieces were then blended with 500 mL of sterile distilled water, and the resulting juice was filtered to obtain pure juice. Catfish seeds were acclimatized for three days before treatment.

Ectoparasite infection was carried out according to the method of Nuriani and Suwartiningsih (2021), by placing two catfish showing signs of infection (wounds on the body and excessive mucus production) into each experimental unit for 24 hours. After the infection period, the diseased catfish were removed, and the infected catfish fry were soaked in a solution of lempuyang gajah rhizome juice at the appropriate concentration for five consecutive days.

Observations for the presence of ectoparasites were carried out by scraping the surface of the fish's body with a scalpel and then observing them under a light microscope. Ectoparasite prevalence was calculated using the formula: the number of infected fish divided by the number of fish examined, multiplied by 100%. Meanwhile, intensity was calculated based on the total number of ectoparasites divided by the total number of infected fish (Fachrussyah *et al*., 2024). Data on quantity, prevalence, and intensity were analyzed inferentially using the ANOVA test and then further tested using the Duncan test. Descriptive analysis was also conducted to support the interpretation of the results.

Results and discussion

The results of the study of Mutiara catfish given five treatments of lempuyang gajah rhizome juice concentration showed that out of a total of 60 samples of fish, 47 were infected with ectoparasites, with a total of 156 individuals. The types of ectoparasites identified in this study included *Dactylogyrus* sp., *Gyrodactylus* sp., and *Trichodina* sp. The highest number of ectoparasites was recorded in the treatment without the addition of lempuyang gajah rhizome juice (0 mL/L), with 55 individuals, while the lowest number was found in the 15 mL/L treatment, with 12 individuals (Table 1). The results of the statistical analysis showed that there was a significant difference in the number of ectoparasites between treatments ($p < 0.05$).

The results showed that the highest prevalence of ectoparasite infection occurred in the treatment without the addition of lempuyang gajah rhizome juice (0 mL/L), which was 100%, categorized as always or very severe infection. Conversely, the lowest prevalence was recorded in the 15 mL/L treatment, which was 58%, categorized as very often or very frequent infection (Table 2). The highest average prevalence was obtained in the 0 mL/L treatment at 33%, while the lowest was recorded in the 15 mL/L treatment at 19% (Figure 1). Statistical analysis showed that the prevalence of ectoparasites differed significantly among the treatments tested ($p < 0.05$).

Table 1. Total number of ectoparasites of Mutiara catfish in five treatments

Treatments	Number of Parasites (Individual)			
	Dactylogyrus sp.	Gyrodactylus sp.	Trichodina sp.	Total
0 ml/L	2	5	48	55[c]
1 ml/L	2	2	32	36[bc]
5 ml/L	0	1	24	25[ab]
10 ml/L	0	1	27	28[ab]
15 ml/L	0	0	12	12[a]

Note: The same superscript letter indicates no significant difference (p>0.05), and vice versa.

Table 2. Prevalence of ectoparasites of Mutiara catfish in five treatments

Treatments	Number of fish	Number of Infected Fish	Prevalence (%)				Category
			Dactylogyrus sp.	*Gyrodactylus* sp.	*Trichodina* sp.	Total	
0 ml/L	12	12	8	17	100	100[a]	Always (very severe infection)
1 ml/L	12	10	17	17	75	83[b]	Usually (moderate infection)
5 ml/L	12	9	8	8	67	75[b]	Usually (moderate infection)
10 ml/L	12	9	0	8	75	75[b]	Usually (moderate infection)
15 ml/L	12	7	0	0	33	58[b]	Very often (very frequent infections)

Note: The same superscript letter indicates no significant difference (p>0.05), and vice versa.

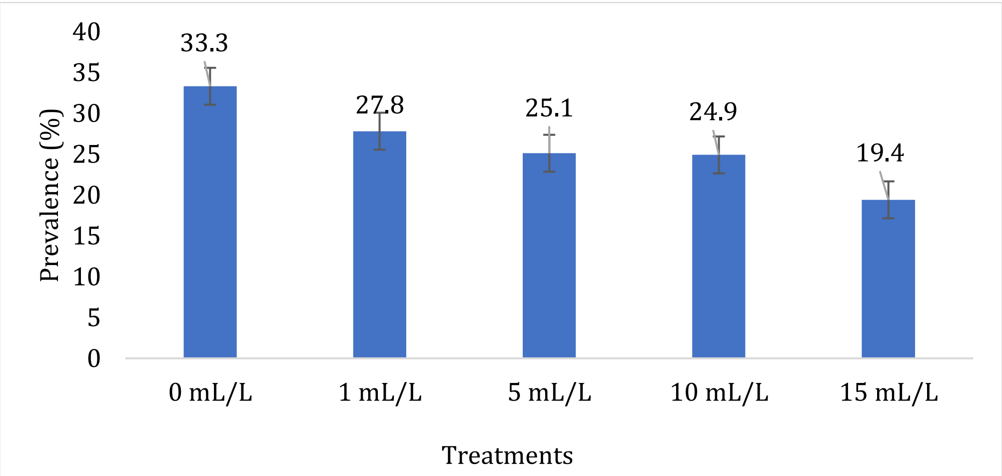

Figure 1. Average prevalence of ectoparasites of Mutiara catfish in five treatments

The highest number and prevalence of ectoparasites in Mutiara catfish were found in the control group or in the treatment without the addition of lempuyang gajah rhizome juice. The administration of this juice showed effectiveness in reducing the number of ectoparasites, where the decrease in the number and prevalence of ectoparasites occurred along with increasing concentration of the applied juice. A similar phenomenon was also reported with the use of ginger juice (*Zingiber officinale*), where increasing the juice concentration resulted in a stronger antiparasitic effect (Nuriani & Suwartiningsih, 2021). This is due to the higher content of active compounds, which can weaken and even kill parasites more effectively (Fu *et al*., 2019).

Lempuyang gajah rhizome, used as a raw material for making juice in this study is known to contain various secondary metabolite compounds that play a role in reducing the number and prevalence of ectoparasites in Mutiara catfish. These active compounds include zerumbone, alkaloids, flavonoids, saponins, tannins, and terpenoids (Affandi & Setyono, 2024). Zerumbon has been shown to kill parasites by triggering reactive oxidative stress (ROS) which causes programmed cell death (apoptosis) (Mukherjee *et al.*, 2016). Alkaloids are toxic to nerves, resulting in paralysis and even death of the parasite (Harahap *et al.*, 2021). Flavonoids work by denaturing cell membrane proteins, thereby damaging membrane permeability, causing leakage of cell contents, and ultimately suppressing growth or killing the parasite (Chandra & Aji, 2023). Saponins cause the release of microvilli so that the parasite cannot attach and dies (Zhou *et al.*, 2021). Tannins damage cell membranes, causing leakage of cell contents, resulting in parasite death (Alavinia *et al.*, 2018). Terpenoids also disrupt membrane permeability and denature proteins, causing leakage of cell contents (Rahim *et al.*, 2016).

Based on the research results, the highest ectoparasite intensity was recorded in the 0 mL/L treatment of 4 individuals per infected fish with a low category. The lowest intensity was found in the 15 mL/L treatment, 1 individual per infected fish, also with a low category (Table 3). The results of statistical tests showed that the ectoparasite intensity differed significantly between treatments ($p < 0.05$).

Table 3. Ectoparasite intensity in Mutiara Catfish in five treatments

Treatments	Number of Infected Fish	Number of Parasites (ind.)	Intensity (individual/infected fish)				Category
			Gyrodactylus sp.	*Dactylogyrus* sp.	*Trichodina* sp.	Total	
0 ml/L	12	55	1	1	6	4^{b}	Low
1 ml/L	10	36	1	1	4	3^{b}	Low
5 ml/L	9	25	1	0	3	2^{ab}	Low
10 ml/L	9	28	1	0	3	3^{b}	Low
15 ml/L	7	12	0	0	2	1^{a}	Low

Note: The same superscript letter indicates no significant difference ($p>0.05$), and vice versa.

Based on data on ectoparasite intensity in Mutiara catfish from five treatments, it was found that increasing the concentration of lempuyang gajah rhizome juice tended to decrease ectoparasite intensity. Of the three parasite species identified, *Trichodina* sp. showed the highest intensity in Mutiara catfish. Ectoparasite inventory results in Sangkuriang catfish fry also showed that *Trichodina* sp. had the highest prevalence rate (Hidayat *et al.*, 2020). Several factors contributing to the high intensity of *Trichodina* sp. ectoparasites include its rapid life cycle and the lack of an intermediate host (Hoar *et al.*, 2020), its ability to reproduce through binary fission, and its spread through direct contact between individuals (Sigit *et al.*, 2019), allowing it to easily reproduce in the rearing environment (Haris & Asran, 2015).

Trichodina sp. is a cosmopolitan protozoan commonly found in high stocking densities and poor water quality. This parasite can cause stress, morphological damage and even death in its host (Munawwaroh & Rahayu, 2017). *Trichodina* sp. populations reach their highest levels in the post-monsoon (post-rainy) season, indicating a significant increase during the transition to the rainy season (Saha & Bandyopadhyay, 2017). *Trichodina* sp. can even survive for two days without attaching to a host (Rokhmani *et al.*, 2020).

Trichodina sp. (Figure 2.a.) is a round, cup-shaped parasite with a diameter of 50–100 μm. It has cilia around its cell, serrated denticles, two nuclei (macronuclei and micronuclei), and an adhesive disc that allows it to attach to the host fish (Yuliani *et al*., 2023). This parasite is known to cause Trichodiniasis in fish (Nugraheny *et al*., 2020). *Trichodina* sp. infection in fish causes itching that encourages rubbing against the bottom or walls of the pond, resulting in wounds, weakness, dull coloration, excessive mucus production, decreased appetite, and a thin, weak body (Sigit *et al*., 2019).

Gyrodactylus sp. (Figure 2.b.) is an ectoparasite with a flat, elongated body, two anterior protrusions, and no eyespots (Fransira, 2023). On the front of its body, it has an attachment device called an opisthaptor, equipped with fine hooks (Shigoley *et al*., 2023), which it uses to attach to its host. This parasite was detected in the skin tissue of Mutiara catfish fry, with an incidence of 10% of a total sample of 60 fry. Infection by this organism is known to cause Gyrodactylosis (Dwilantiani *et al*., 2019). Infected fish generally exhibit behavioral responses such as frequently rubbing their bodies against surrounding objects and frequently rising to the water surface (Sari *et al*., 2020).

Dactylogyrus sp. (Figure 2.c) has a flat, elongated body with four anterior protrusions, two eyespots, and a sucker (Irwandi *et al*., 2017). Three of the 60 Mutiara catfish fry (5%) were detected as infected with *Dactylogyrus* sp. Fish infected with this ectoparasite show clinical signs in the form of changes in body color to pale, excessive mucus production on the skin, fins that appear to be closed, impaired balance when swimming, and increased respiratory rate (Putri *et al*., 2016).

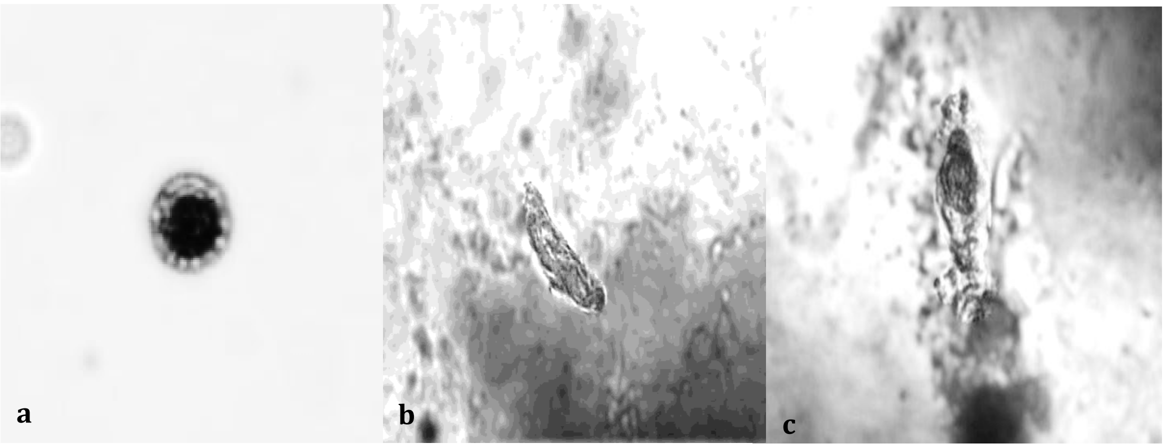

Figure 2. a. *Trichodina* sp., b. *Gyrodactylus* sp., c. *Dactylogyrus* sp.

These findings have important implications for catfish farming, particularly in developing sustainable and health-conscious aquaculture practices. The significant reduction in the number, prevalence, and intensity of ectoparasites following the administration of lempuyang gajah rhizome juice suggests that this natural remedy can serve as an effective, environmentally friendly alternative to synthetic antiparasitic treatments. Incorporating plant-based therapeutics such as lempuyang gajah into routine management strategies can help minimize parasite-related losses, reduce chemical residues in aquaculture systems, and enhance fish welfare. Moreover, the use of locally sourced herbal treatments aligns with cost-effective practices and supports the development of eco-aquaculture, thereby contributing to improved productivity and sustainability in catfish farming operations.

Conclusion

The results of the study showed that administering lempuyang gajah rhizome juice significantly reduced the number, prevalence, and intensity of ectoparasites (*Trichodina* sp., *Gyrodactylus* sp., and *Dactylogyrus* sp.) in mutiara catfish fry, with the highest effectiveness achieved at a concentration of 15 mL/L. This reduction correlated with increased levels of bioactive compounds such as zerumbon, alkaloids, flavonoids, saponins, tannins, and terpenoids, which act through toxic mechanisms, membrane disruption, and induction of oxidative stress in parasites. These findings demonstrate the novelty of using lempuyang gajah rhizome juice as a natural and environmentally friendly alternative for ectoparasite control in Mutiara catfish cultivation, replacing synthetic antibiotics that carry the risk of resistance and contamination. Further research is recommended to evaluate the effectiveness of ginger extract in long-term application, its effects on fish growth and immunity, and the potential for commercial phytopharmaceutical-based formulations for sustainable aquaculture.

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