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Prevalence of Endoparasites in Two Commercially Important Fish *Sardinella lemuru* and *Glossogobius guiris* from Coast of Cabadbaran and Lake Mainit, Caraga, Philippines

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Abstract

Fish often serves as a staple food and protein source. Most Filipinos depend on fishing activities as a means of living. Fish parasites are diverse and pervasive organisms affecting a variety of hosts, including commercially important fish. Despite being utilized for commercial purposes, limited studies on fish parasitism are done in the Caraga region and are published in the Philippines. This study is one of the few conducted on the prevalence of parasites from the coast of Cabadbaran City and Lake Mainit in Jabonga, Agusan Del Norte, Phils. The two areas were the major fishing ground for different fish varieties sold in the local market. Two fish samples were collected: *Glossogobius guiris* (n=100) and *Sardinella lemuru* (n=100). Standardized protocols for fish parasite investigation were adopted. From the collected fish samples, the gastrointestinal tract was subjected, then examined and isolated. Of the 200 individuals, there were 2 found infected with*G. guiris* and 8 infected with*S. lemuru*. The recorded parasite was identified as Nematodes and was mostly observed in marine fish species (*S. lemuru*) when compared to freshwater fish (*G. guiris*). This implies that the occurrence of these parasites may vary depending on the fish type, maturity, diet composition, and location. A significant difference p=0.0003 was observed in the length-weight and prevalence of fish parasites and infers that size structure is associated with parasitic rate. Thus, the importance of conducting fish parasites could give an understanding of how this ubiquitous organism affects commercially important fish and awareness to the fish-eating community.

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Introduction

In the Philippines, fish livelihood activities greatly contribute to the local and national economies. Fishing has produced a large contribution to the country's development, with the recorded input of an estimated 4.33 billion dollars to the country's financial resources (BFAR, 2016). Due to this record, there is a need for intensive management of fishery activities to maintain the economic stability of the country. Along with this is the careful assessment of the quality of harvest as its Philippine catch value increases over time (Anticamara and Go, 2016). Fish is successfully marketed according to a range of customer key standards (Saeed *et al*., 2022), and such requirements are qualities that are largely influenced by environmental factors (Fritriani*et al*., 2019).

A prevalent problem in aquatic animals is parasitism which contributes to high mortalities among fish stocks (Edeh & Solomon, 2017). This special interaction between organisms involves a parasite harming a host or limiting its abundance. The impact of these organisms stretches out to an ability to wipe out other species due to their ability to cause wide epidemics, especially for disease-causing parasites (Frainer *et al*., 2018). Parasites are classified into ectoparasites, parasites on the external part of the host and endoparasites, parasites within the host's body. Such endoparasites may be located within the flesh and organs of the host organism (Edeh & Solomon, 2017). Flagellates, amoebae, and Haplosporidia are examples of these parasites, to name a few (Lucas *etal*., 2019). The existence of these harmful organisms affects fish quality such that a study made byRamos, 2020 proved that some of these parasite-infested fish products are commercialized.

Despite their ability to cause diseases, fish are also known to be effective bioindicators. In an evaluation performed by Fierro *etal*., 2019, fishes were proven to present information on human agricultural activities to which their setting is subjected, as well as the water pollution in the same setting. In addition, fish examination gives information on the health and environmental disturbances by assessment of the inner organs and tolerance. Fish had also been used in a different study by (Gutiérrez and Agudelo, 2020) in assessing the accumulation of coal and mercury in Colombian waters, which gave significant results. Many factors contribute to the results of these bioassessments, namely, the increase in population (Rabadon & Corpuz, 2021), as well as climate change and the pollution that is brought about by the same factor (Wu *etal*., 2019).

Moreover, the fish-size relationship and the occurrence of parasites in fish products reduce the value and quality to drop, thus changing the marketability of the fish (Abollo *etal*., 2001; Levsen *et al*., 2005; Karl, 2008; Llarena-Reino *etal*., 2013; D'Amico *et al*. 2014; Llarena-Reino *et al*., 2015).

Fish parasite infection and pollution develop in ineffective energy and nutrient consumption and an escalation in energy expenditure of fish through respiration, impacting reproduction and growth (Marcogliese and Pietrock, 2011; Khan, 2012; Shea-Donohue *et al*., 2017; FAO 2020). More importantly, the risk posed by fish-borne parasitic zoonoses to human health (Quiazon, 2015) is estimated to be high, especially since fish is the staple food of fishing communities and eating raw fish is common practice in the region (Soares Magalhães *etal*., 2014; Tenorio and Molina, 2021).

This study utilized *Sardinella lemuru* (Tamban) and *Glossogobiusguiris* (Pidjanga), which is a commercially important fish in the Caraga region and the Philippines as well. Being ranked second in terms of volume in fish production, it is considered to be one of the most abundant fish and an affordable source of protein in the country (Labrador *et al*., 2021). These fish have a good hold on the economy of the Philippines and can affect the state of that part of the country. However, according to a study (Pohle, 2013), consumers have the trend in food without assessing the quality and information of the said product. In this case, consumers are at risk of disease-causing factors brought upon by the products they

buy due to their lack of awareness and lack of assessment from the food industry (Ziarati *etal.*, 2022). In support of this information, the American Society has found that about 260,000 people have reported being sick from consuming fish products. Likewise, Barrett *etal.*, 2017 also recorded 857 outbreaks causing about 4800 illnesses with hospitalizations and deaths in the same country due to polluted fish.

Further, due to their availability and precise identification of species, fish assessments prove to be convenient and useful in scientific investigations. It is also imperative to perform with scientifically proven and applicable methods along with the locally-obtained samples. More importantly, there is a need to use a preventive approach in the maintenance of fish health in our aquatic resources (Assefa & Abunna, 2018). Moreover, Cook, 2017, infers that the foundation of sustainability rests upon three factors which are social, economic, and environmental. There were no studies conducted in the region utilizing the fish samples. Thus, the study aims to provide baseline information to address the gap. Lastly, the focus of the study is to determine the prevalence of endoparasites in *Sardinella lemuru* and *Glossogobius guiris*. While this also touches on the protection and conservation of our water resources as well as the health of the fish-consuming community.

Materials and methods

Fish collection

The fish collection was done with the aid of local fishermen by using gillnet (Pokot). A total of 200 fish samples comprising (n=100 *S. lemuru* and n=100 *G. guiris*) were collected from Coast of Cabadbaran City and Lake Mainit in Jabonga, Agusan Del Norte (Fig. 1).

Fig. 1. Map of the study area, Cabadbaran City and Lake Mainit, Caraga, Philippines.

Samples were sorted and placed in an ice-filled Styrofoam box for transport. Further, the laboratory process was done in the Biology Laboratory of Caraga State University-Main Campus, Ampayon, Butuan City, Agusan Del Norte, Philippines.

Determination of length and weight of fish samples

The fish total weight (TW) was measured using a digital weighing scale, and the total length (TL) was measured using a ruler (Abdel- Ghaffer *et al.*, 2015). The length and weight of the fish samples were taken inside the laboratory.

Isolation and identification of endo-parasites

Individually, the fish sample was dissected through its abdomen by using the scalpel and scissors. The gut of the fish was extracted and placed directly into the Petri dishes with a 0.6% saline solution. Each isolated organ was sliced and cut to observe the presence of the parasite.

Afterward, a dissecting microscope was used for the parasite determination (Kawel *etal.*, 2016). Using the study of Moravec & Van 2015; da Silva *etal.*, 2018; Neves *etal.*, 2020, the identification of helminth parasites.All the observed endo-parasites were preserved in 70% ethyl alcohol (Leela & Rao, 2014).

Data analysis

Data were analyzed using the formula based on the formula of Miah*et al.*, 2013 and Margolis*etal.*, 1982 in analyzing the degree of parasitism in an organism. Graph Pad Prism 8.0.2 was used to determine the significant difference and Pearson Correlation Coefficient.

Where:

Prevalence is equivalent to the number of individuals of a host species infected with a particular parasite species divided by the number of hosts examined.

Formula: $\text{Prevalence rate} = \frac{\text{No. of Host Infected}}{\text{Total No. of Host Examined}} X\ 100$

Mean Intensity is the total number of a particular parasite species in a sample of a host species divided by the number of infected individuals of the host species in the sample.

Formula:

$$\text{Intensity of Infection} = \frac{\text{No. of Parasites Collected in a Sample}}{\text{No. of Infected Host}}$$

Relative Density or Abundance is equal to the total number of individuals of a particular parasite species in a sample of hosts divided by the total number of individuals of the host species (both infected and uninfected) in the sample.

Formula:

$$\text{Density of Infection} = \frac{\text{No. of Parasites Collected in a Sample}}{\text{Total Host Examinded}}$$

Results and discussion

The length and weight of the collected fish samples from the two study areas are presented in Table 1. *S. lemuru* recorded (126.58 ± 0.40) for TL and (17.62 ± 0.18) for TW. In contrast,*G. guiris* recorded (148.68 ± 0.74) for TL and (36.13 ± 0.51) for TW. As observed,*G.guiris* were longer and heavier when compared to *S. lemuru*. Importantly, the relationship between length and weight would develop further explanations about the study. Whereas it also gives a view of how these two components correlate with the prevalence of the parasites present in the two selected fish samples.

Evidently, length and weight play major roles in identifying parasite investigation; hence it is known that size can accumulate and increase the rate of parasites. A significant difference (p=0.0003) was observed in the size (length-weight) of the fish, indicating essential information for parasite accumulation. The length-weight relationship (LWR) is important to offer basic evidence in fisheries biology (Paswan *etal.*, 2012). At the same time, LWR emphasizes the maturity, life span, production, growth, and mortality of the fish stock (AAS, 2000; Ak *etal.*, 2009; King, Lafferti & Morris, 1996; Diaz *et al.*, 2000). Fish size structure is often used to associate contamination as it can tolerate different levels. Thus, fish body weight had a purpose on the prevalence of parasites across infected fishes (Ramos *etal.*, 2020). However, the connection between parasite infection and host body length differed between the host and parasite (Hila Bu and Leong, 1999). Nevertheless, the larger the fish, the larger size of parasites infected and gathered when compared to small fishes. As observed, the total length of the *S.*

lemuru (126.58 ± 0.40) centimeter is quite small in size and the collected parasite was few.

On the other hand, fish are commonly used as biomarkers of infection and are subjected to different studies to assess ecological health.

In contrast, they are susceptible to various environmental degradation and directly impacted by pollution through anthropogenic activities. Fishes are direct transitional hosts of passing for several parasites (Nebel *etal*., 2020). In particular, marine fish species are widely known to be infected by this organism (Ramos, 2020). Parasitic nematodes compose one of the earliest identified groups of helminths in fishes (Molnár *etal.* 2006). Several nematodes are categorically endoparasites infecting sea birds, marine mammals, and fish. About four major anisakids have been proven to infect marine fish species (Dadar *etal*., 2016).

Table 1. Length and weight from two collected fish species on the Coast of Cabadbaran, and Lake Mainit, Caraga, Philippines.

Species name	English name	Local name	Total length (cm)	Total weight (g)
Sardinella lemuru	Bali sardines	Tamban	126.58 ± 0.40*	17.62 ± 0.18*
Glossogobius guiris	Tank goby	Pidjanga	148.68 ± 0.74*	36.13 ± 0.51*

Note - * Significant $p<0.05$.

Further, *G. guiris* is a freshwater fish species and there were no studies conducted yet performing the prevalence of parasitic infection. This present study serves as the first conducted and found very few infected with nematodes. Several works of literature studying the marine fish parasite and when compared to freshwater fish, were scarce. But freshwater and brackish water fish play a foremost role as the basis of human contaminations with food-borne trematodes, and these parasites are getting the most attention as evidence of their diversity and prevalence arises in some Asian countries (WHO, 1995; Chai *etal*., 2005; Chai, 2007).

Notwithstanding the influence of parasites in a fish individual or assemblages, altogether its effect on food safety and socio-economic aspect among coastal communities greatly reliant on the fishery, studies on parasites of commercially important fishes are rare (Gustilo, 2022). Nevertheless, fish is the best and cheap protein source, but numerous diseases involving parasitic infections can cause a threat to the fish-eating community (Yooyen *etal*., 2006). This scenario has not been given importance by the concerned agency of how this ubiquitous organism could possibly affect human health for long exposure. Equally, saltwater fish and freshwater fish are possible sources of parasitic infection affecting humans. Fish become infected with parasites when they feed on intermediate hosts. Though nematodes might be an essential pathogen among fish species, above all, it is typically their role to spread parasites to humans, who turn out to be accidental hosts. (Iqbal *etal*., 2019). Additionally, the physiological condition of fish possibly plays a significant role in the level of infection in different fish and organs (Shalaby *et al*., 1989). Accordingly, numerous diseases involving parasitic contaminations stance a threat to fish cultivation when compared to the natural settings (Ali *etal*., 2016). Moreover, by increasing marine/brackish and freshwater water fisheries along Asia, the economic impact on commercially important aquaculture of fish-borne zoonotic parasites pose a threat to the quality and food safety and thus extending burdensome (Duarte *etal*.,2007).

It is important to recognize how ecological modifications impacted parasites to forecast how they could respond to the changes and how those variations can influence the ecosystem (Iqbal *etal*., 2019). As said, the increased rate of parasites in fish signifies the possible risk to public health.

Inappropriately cooked fish is the foremost source of these contaminations for humans, and this has been reported in many geographical areas (Shamsan and Al-Jobory, 2018). Consequently, there must be public awareness activities concerning the effects on the zoonotic of fish parasites and the threat of eating raw or undercooked fish. Then, it is necessary not to consume raw fish because many Filipinos frequently eat *"kinilaw"* from Tamban, and this can pose a health risk.

Table 2. Prevalence of fish parasites from two collected species on the Coast of Cabadbaran and Lake Mainit, Caraga, Philippines.

Species name	Location	N	Diet composition	Prevalence rate	Intensity of Infection	Density of infection
Glossogobius guiris	Freshwater	100	Piscivores	*2%	1	0.02
Sardinella lemuru	Marine	100	Omnivorous zooplanktivore	*8%	1	0.08

*Significant $p<0.05$.

From the 200 individual fish samples collected, the prevalence rate was observed to be highest in *S. lemuru* (8%) when compared to *G. guiris* (2%). The intensity of infection, both 1% and the density of infection were highest in *S. lemuru* (0.08) and *G. guiris* (0.02), respectively (Table 2). A significant difference of p=0.0003 in the prevalence rate suggests that species type, diet composition, location, and maturity could accumulate different levels of parasite infection. Seeing the prevalence rate of the two fish samples, there were more parasites collected in *S. lemuru;* this might be associated with the location and feeding habits of the fish. The marine area is a vast geographical region where there are large numbers of fish assemblages interacting. This could draw a complex food web where each fish utilized a variety of nutrients. Ecological settings can interplay the health of each organism and that affects physiological ability.

As detected, there were few quantitative values obtained; this might be due to the reason that the number of individual samples was only 100 each, and this influenced the generated data. It was noticed that the larger the samples, the more fish parasites were collected. Also, the size structure (length and weight) influences the presence of these parasites. Further, Pearson Correlation Coefficient revealed errors during the data analysis. It was because the values were too few that affected the data. It is a need to collect large numbers of samples and larger sizes to be able to produce substantial data and to perform the correlation analysis. Therefore, it was impossible to correlate the length-weight relationship to the prevalence of parasites in the present study.

Nonetheless, the collected parasites among the two-fish species represent the occurrence of the latter. Although the prevalence rate is very low,there were still documented. So, this supports the present study of the existence of these parasites. A study showed that various parasites might respond to fluctuations in their location in different ways. In regions with heavy fishing pressure, multi-host life cycles tend to decline, and parasites with composite activities. However, ones with uncomplex life cycles incline to increase (Iqbal *etal*., 2019). This is associated with the feeding habits that fish is reliant on; this is associated with parasite growth. Such that, Anisakid nematodes (Nematoda: Anisakidae) are common parasites of marine fish used as intermediate, paratenic, and definitive hosts (Cruz *etal*., 2007). The evidence on marine fishes' food habits, such as the predator-prey relationship, is valuable to evaluate the function of marine fishes in the environment (Bachok *etal*., 2004).

In addition, the threat associated with fish-borne parasitic zoonoses to human health (Quiazon, 2015; Bao *et al*., 2019) is assessed to be increasing,

specifically as fish is the staple food of fishing communities, and eating raw fish is frequently practiced in the region (Soares Magalhães *et al*., 2014; Tenorio and Molina 2021). Furthermore, as of today, the emergence of zoonoses infection geographically extends from the Philippines, where incidence arises as small epidemics along families located nearby rivers and lakes that directly consume fish (Khalil *etal*., 2014). Along with the importance of public health awareness, the study provides information on the occurrence of these fish-borne parasites. These are roundworms (nematodes), flatworms or flukes (trematodes), and tapeworms (cestodes) (Iqbal *etal*., 2019). In relation, the same study was conducted to examine the parasitic infestation in *S. lemuru* and found the same type of parasites (Ramos *etal*., 2020) While; previous works determined that fish from more contaminated waters were inclined to harbor more parasites than from less polluted area (Ali, 2021).

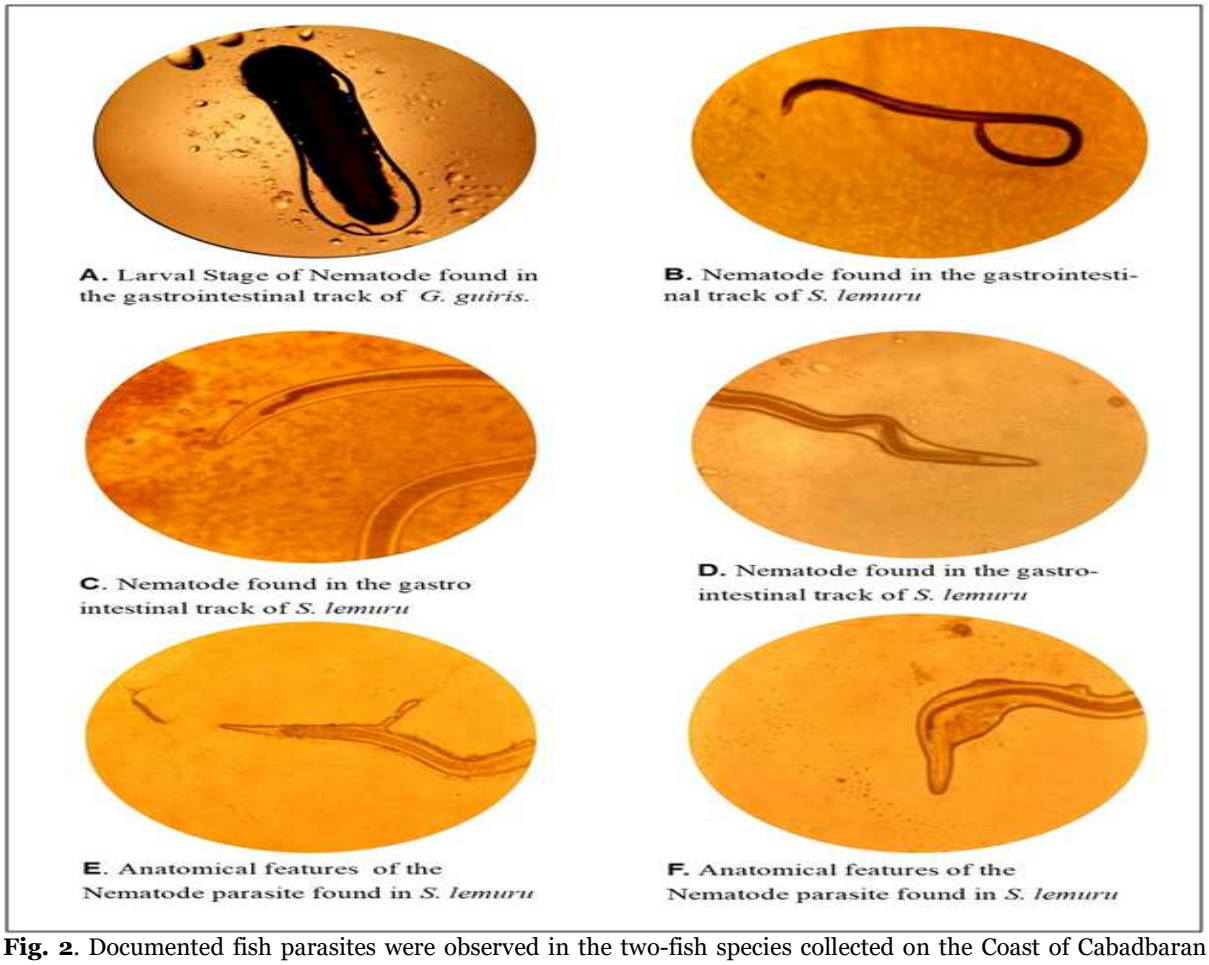

Fig. 2. Documented fish parasites were observed in the two-fish species collected on the Coast of Cabadbaran City and Lake Mainit, Caraga, Philippines.

Accordingly, the foremost reasons stating the diversity of fish parasites and the intensity and incidence of infection can be linked to the feeding habits, life span, mobility throughout its life span, the kinds of environments it comes across, the density of the population, and the size attained, where the large host offers more habitats fit for parasites than small ones (Dhole*etal*., 2010). In particular, nematodes are also playing a vital role in marine ecosystems by perturbing their hosts' population dynamics (Rohde, 1993). Nevertheless, the documented fish parasites from the two collected fish species only prove that they are pervasive organisms that interplay among biological resources. Thus, the present study identified the prevalence of nematodes in the gastrointestinal tract of the fish samples collected from the two study areas. (Fig. 2). Lastly, the relevance of conducting the study would help to

spread awareness about fish parasite contamination and human health risks.

Conclusion

The present study identifies the occurrence of fish parasite-Nematodes between *S.lemuru* and *G. guiris*. Prevalence rates were highest in *S. lemuru* when compared to *G. guiris*. It is possibly due to the following reasons, maturity, feeding habits, and geographical location. However, the recorded intensity of infection and density of infection was too low; this was related to the sample size collected. A significant difference was observed p=0.0003 between the length-weight and prevalence rate, indicating substantial information that each fish species has the ability to harbor parasites depending on its size structure. The larger the fish is, the more it can hoard fish-borne parasites., Thus, the goal of the study is to determine the prevalence of these parasites while associating the awareness of fish-eating communities for their food safety. Many of these parasites could pose a health risk and the threat of eating raw or undercooked fish might develop a health problem.

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