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EPIDEMIOLOGICAL ASSESSMENT OF HELMINTH PARASITE INFECTIONS IN MARINE FISHES FROM COASTAL WATERS OF KARACHI, PAKISTAN

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Abstract
Fish are a major source of affordable protein globally and play an essential role in supporting economic livelihoods through fisheries. The present study assessed the prevalence and intensity of helminth parasite infections in four commercially valuable marine fish species—Lutjanus argentimaculatus, Johnius dussumieri, Plectorhynchus cinctus, and Pampus argenteus—collected from the Karachi coast. From January to December 2022, a total of 113 fish specimens were collected monthly using cast nets, hand nets, and fishing rods with the help of local fishermen. Parasitological examination revealed that 84.07% of the fish were infected with helminth parasites. The highest infection prevalence was recorded in June (45.46%), while the lowest occurred in January (3.33%). Among the studied species, Pampus argenteus showed the greatest mean infection intensity (2.15 parasites per infected fish), followed by Johnius dussumieri (1.89), Plectorhynchus cinctus (1.87), and Lutjanus argentimaculatus (1.70). Seasonal analysis indicated a markedly higher infection rate during summer compared to winter. These results highlight the species-specific vulnerability of marine fish to helminth infections and underscore their implications for fish health, food safety, and sustainable fisheries management.

INTRODUCTION

Fish represent approximately half of all vertebrate species inhabiting marine and freshwater ecosystems, playing a crucial ecological and economic role worldwide. They serve as an essential source of high-quality proteins, omega-3 fatty acids, and other vital nutrients necessary for human health (FAO, 2022). Beyond their nutritional significance, fisheries and aquaculture contribute substantially to national economies, offering employment and food security, particularly in developing nations (Barange et al., 2018). Fish are consumed in various forms— cooked, salted, smoked, or preserved—and their by-products, zuch as fish oil, fish meal, isinglass, fertilizers, and fish glue, are of immense commercial value.

However, the global fish industry faces a critical challenge: parasitic infections. Parasitic diseases significantly impact fish health, reducing their market value, posing risks to human consumers, and causing substantial economic losses (Woo &Buchmann, 2012). Helminth parasites— including trematodes, cestodes, nematodes, and acanthocephalans—are among the most prevalent and harmful pathogens affecting both wild and farmed fish populations. These parasites compromise fish vitality, induce tissue damage, impede growth rates, and in severe cases, cause mortality (George, 2021).

Ecologically speaking, fish are key players in aquatic food webs, often acting as apex predators or important prey species. Their interactions with helminth parasites provide significant new perspectives on ecosystem health and stability (Poulin&Morand, 2000). By changing host behaviour, feeding efficiency, and reproductive success, parasitic infections can affect population dynamics and Studying the parasitology of fish species is therefore not only essential for preserving food safety but also for comprehending more general ecological processes. Various biotic and nonliving elements affect the frequency and severity of helminth infections. These include host species, age, diet, immune status, and environmental parameters, including water temperature, salinity, pH, and pollution levels (Moravec, 1994). Among these elements, temperature seems especially important; many studies have demonstrated that higher temperatures correspond with higher parasite growth and transmission rates (Marcogliese, 2001). On the other hand, salinity tends to have more erratic effects depending on parasite and host species (Blanar et al., 2014). Seasonal changes are also significant since helminth infections usually peak during warmer months when the parasites' intermediate hosts—e.g., crustaceans, mollusks—are more plentiful and active (Sattar et al., 2021).

Particularly among lower-income groups, fish eating in Pakistan is vital for solving nutritional gaps. Pakistan has a great variety of fish species since it has a long marine coastline of over 1100 kilometres and plentiful inland water resources. Parasitic diseases, meanwhile, are still mostly an uninvestigated danger to this asset. Although freshwater fish parasites have received some attention (e.g., Abro, Birmani, & Bhutto, 2019; Nabi et al., 2020), research on the parasitic fauna of marine fishes is lacking. Though earlier studies (Costa &Biscoito, 2003; Soler- Jiménez et al., 2017) have recorded helminth infections in coastal fish worldwide, data particular to the Karachi coast remains scant and scattered.

Khurram Khan et al.'s earlier studies (2019) and Rizwana et al.'s (2019) found a community structures (Marcogliese, 2005). high frequency of trematode and nematode infections in certain fish species. However, there were occasional constraints on the range of host species examined, geographic coverage, and temporal scope. Furthermore, shifting environmental factors—including coastal pollution, urban development, and climate change—could change parasitic infection patterns over time (Zhu et al., 2020). Therefore, quick, methodical parasitological surveys are required.

Particularly in the gastrointestinal tract, parasites usually use organs that provide direct access to nutrients and survival niches. The intestine is often mentioned as the most infected organ in marine fish, especially given its function in nutrient absorption and direct exposure to consumed infectious stages (Parveen, Khatoon, &Waheed, 2020; Marcogliese, 2005). Depending on parasite species and host vulnerability, other organs, such as the liver, stomach, and swim bladder, might also act as secondary infection sites.

Apart from environmental and financial issues, helminth infections in fish have clear effects on human health. Some fish- borne helminths, especially certain trematodes and nematodes, are zoonotic and can infect people when undercooked or raw fish is eaten (Chai et al., 2005). Therefore, monitoring the prevalence of helminths in edible fish species is crucial for public health surveillance and ensuring food safety.

Research conducted all over the world shows consistently that helminth infections show significant seasonal patterns. Accelerated parasite life cycles and greater biological activity of intermediate hosts cause warmer months to often coincide with higher parasite loads (Genc et al., 2005; Khalid et al., 2020). The Karachi coast, marked by a semi-arid climate and unique seasonal variations, likely reflects these worldwide trends, as earlier results by Qasim and Ayub (2012) noted higher helminth prevalence during summer months, though they found little relationship with seawater temperature. These findings imply that infection rates may also be significantly influenced by other elements, including local environmental stressors, intermediate host availability, and fish migratory patterns.

Although there are many worldwide studies on helminth parasitology in freshwater fish (Scholz et al., 2018), marine systems, particularly in developing countries, remain relatively ignored. Research on the presence of helminths in marine fish species in Karachi is important and needed right now because marine fisheries are economically important to Pakistan, and there are potential health risks from parasites that can be transmitted from fish to humans.

This study aims to address research gaps by examining how common, severe, and widespread helminth parasites are in four important marine fish species— *Lutjanus argentimaculatus, Johnius dussumieri, Plectorhynchus cinctus,* and Pampus *argenteus*— collected from various fish markets along the Karachi coast. The study aims to guide fisheries management practices, improve food safety initiatives, and add more general ecological knowledge of marine parasitism in this area by means of updated parasitological data.

This study is the first step in setting a standard for future long-term studies that will look at how parasitic infections change over time, helping to assess the impacts of climate change, pollution, and other human activities on marine parasites. Sustainable management of Pakistan's marine resources and protection of public health depend on a clearer understanding of these dynamics.

Materials and Methods Sample Collection

A total of 113 marine fish specimens were collected from various locations along the Karachi coast between January and December 2022. Fish species examined included *Lutjanus argentimaculatus*, *Johnius dussumieri*, *Plectorhynchus cinctus*, and *Pampus argenteus*. Specimens were captured using standardized fishing techniques, including cast nets, hand nets, and fishing rods, with assistance from local
fishermen to ensure species diversity (Genc et al., 2005). Immediately after capture, live fish were transported to the laboratory at the Department of Zoology, Federal Urdu University of Arts, Science and Technology, Karachi, in aerated containers to minimize stress and prevent post- mortem parasitic migration (Bush et al., 1997).

Dissection and Examination

Upon arrival at the laboratory, fish specimens were euthanized humanely by ethical guidelines for the treatment of aquatic animals (Snieszko, 1974). Each fish was dissected longitudinally from the anus to the lower jaw using sterilized surgical scissors, ensuring minimal damage to internal organs. Thorough examinations were conducted on the alimentary canal and major organs, including the heart, liver, swim bladder, kidneys, spleen, body cavity, genital organs, gills, and eyes, for the presence of helminth parasites.

Organs were carefully excised and immediately transferred into petri dishes containing physiological saline solution (0.85% NaCl) to facilitate parasite recovery without distortion (Bykhovskaya- Pavlovskaya et al., 1964). Dissections and preliminary identifications were performed under a stereomicroscope at magnifications ranging from 10× to 40×.

Parasite Recovery and Preservation

Helminths encountered during examination were categorized into three major taxonomic groups: nematodes, cestodes, and trematodes.

- **Nematodes and cestodes** were recovered live, killed gently by immersion in hot (70°C) 70% ethanol, and preserved in vials containing alcohol–glycerol solution for further study (Roberts &Janovy, 2009).
- **Trematodes**, due to their muscular and flattened bodies, were gently compressed between clean glass slides and coverslips, secured with cotton thread to avoid tissue rupture (Chubb et al., 1987). These compressed specimens were fixed overnight in alcohol–formalin–acetic acid (AFA) fixative.

Following fixation

- Specimens were rinsed thoroughly in 70% ethanol to remove fixative residues.
- Trematodes were stained using borax-carmine to enhance morphological features necessary for identification (Gibson, 1996).
- Progressive dehydration was performed by transferring specimens through a graded ethanol series (80%, 90%, and 100%).
- Dehydrated specimens were cleared

sequentially in clove oil and xylene to achieve transparency.

- Finally, permanent mounts were prepared using Canada balsam as the mounting medium. Mounted slides were cured by placing them in a drying oven at 55–60°C overnight to ensure proper hardening.

Identification and Classification

Helminth specimens were identified to the genus and, where possible, species level based on morphological keys and taxonomic descriptions following Yamaguti (1958) and updated references such as Bray et al. (2008). Identification criteria included body shape, size, the structure of attachment organs, reproductive system morphology, and cuticular features.

Data Analysis

Prevalence, mean intensity, and relative abundance of parasitic infections were calculated following standard epidemiological formulas outlined by Bush et al. (1997):

- **Prevalence** (%) = (Number of infected hosts / Total number of hosts examined) × 100
- **Mean Intensity** = (Total number of parasites recovered / Total number of infected hosts)
- **Relative Abundance** = (Total number of parasites recovered / Total number of hosts examined)

Statistical analyses were performed using GraphPad Prism Version 5.0 software. The chi-square (χ^2) test was applied to evaluate differences in infection rates among different fish species and parasite groups. A p-value of less than 0.05 was considered statistically significant (Zar, 2010).

All procedures were conducted under strict hygienic conditions to prevent cross-contamination and ensure the accuracy of results.

Statistical Analysis

The prevalence rate (PR), mean intensity (MI), and relative density (RD) of helminth parasite infections were calculated following the standard epidemiological formulas proposed by Aydoğdu (2011):

- **Prevalence Rate (PR,** %) = (Number of infected hosts / Total number of hosts examined) × 100
- **Mean Intensity (MI)** = Total number of parasites recovered / Number of infected hosts
- **Relative Density (RD)** = Total number of parasites recovered / Total

The association between infection rates across different host species and parasite groups was evaluated using the chi-square (χ^2) test. Statistical significance was considered at p-values < 0.05 with a 95% confidence interval (Zar, 2010). Analyses were performed using GraphPad Prism Version 5.

Results Table 1

Prevalence of Helminth Infection (January–December 2022)

Fish Examined	Fish Infected	Prevalence (%)	χ^2 (p-value)
113	95	84.07	0.000

Our analysis revealed a high overall prevalence rate of helminth infection (84.07%) among the 113 examined marine fish specimens. The chi-square test yielded a p-value < 0.001, indicating a statistically significant association between helminth infection and the fish population studied. This high infection rate suggests that helminth parasitism is widespread among marine fish from the Karachi coast, likely reflecting conducive environmental conditions, the availability of intermediate hosts, and possibly poor water quality or high fish density in the region (Bush et al., 1997; George, 2021).

Table 2
Prevalence of Different Helminth Parasite Groups

Helminth Type	No. of Parasite s	Prevale nce (%)	Mean Intensity	χ² (p- value)
Trematodes	68	60.17	0.715	0.9999
Nematodes	89	78.76	0.936	0.9999
Cestodes	20	17.69	0.947	0.9999
Acanthocep halans	0	0.00	0.00	N/A

Among the different helminth groups detected, nematodes exhibited the highest prevalence (78.76%) and a mean intensity of 0.936 parasites per infected fish. Trematodes were also notably common, with a prevalence of 60.17% and a mean number of hosts examined intensity of 0.715. Cestodes, however, were much less prevalent (17.69%). No acanthocephalan parasites were recorded during this study. The uniformly high p-values (>0.9999) suggest no significant variation between helminth groups, although nematodes predominated numerically. The high prevalence of nematodes aligns with prior studies indicating their dominance in tropical and subtropical marine ecosystems (Moravec, 1994; Scholz et al., 2018).

Table 3
Prevalence of Nematode Infection by Fish Species

Fish Species	Exam ined	Infe cted	Nema todes Recov ered	Preva lence (%)
Lutjanusargen timaculatus	35	30	24	85.71
Johniussdussu mieri	40	36	32	90.00
Plectorhynchu scinctus	18	15	15	83.33
Pampus argenteus	20	14	18	70.00

Analysis by host species showed that *Johnius dussumieri* had the highest nematode prevalence (90.00%), followed by *Lutjanus argentimaculatus* (85.71%) and *Plectorhynchus cinctus* (83.33%). Although *Pampus argenteus* exhibited a slightly lower nematode prevalence (70.00%), the recovered nematode counts were relatively high. These results suggest species-specific susceptibility, potentially influenced by dietary habits, ecological niche, or differing immune defences. Predatory and bottom- dwelling fish such as *Johnius dussumieri* are more likely to acquire nematode infections through the consumption of infected crustaceans or smaller fish (Chai et al., 2005).

Table 4
Prevalence of Trematode Infection by Fish Species

Fish Species	Exam ined	Infe cted	Trema todes Recov ered	Preva lence (%)

Lutjanusargen timaculatus	35	30	18	51.43
Johniussdussu mieri	40	36	28	70.00
Plectorhynchu scinctus	18	15	10	55.55
Pampus argenteus	20	14	12	60.00

For trematode infections, *Johniussdussumieri* again recorded the highest prevalence (70.00%), followed by *Pampus argenteus* (60.00%), *Plectorhynchuscinctus* (55.55%), and *Lutjanusargentimaculatus* (51.43%). The comparatively higher trematode prevalence during warmer months might relate to the increased activity and abundance of snail intermediate hosts, as suggested by seasonal parasitological studies (Genc et al., 2005; Gautam et al., 2018). The trophic habits of these fish likely expose them to different levels of trematode infective stages.

Table 5

Prevalence of Cestode Infection by Fish Species

Fish Species	Exam ined	Infe cted	Cesto des Recov ered	Preva lence (%)
Lutjanusargent imaculatus	35	30	9	25.71
Johniussdussu mieri	40	36	8	20.00
Plectorhynchus cinctus	18	15	3	16.66
Pampus argenteus	20	14	0	0.00

In terms of cestode infections, *Lutjanus argentimaculatus* showed the highest prevalence (25.71%), whereas *Johnius dussumieri* and *Plectorhynchus cinctus* exhibited lower prevalences (20.00% and 16.66%, respectively). No cestodes were recovered from *Pampus argenteus*. This absence suggests that either the trophic ecology of *Pampus argenteus* minimizes exposure to infected intermediate hosts, or cestode species capable of infecting this fish are rare in the local marine environment (Bray et al., 2008). Cestode infections are typically acquired through the ingestion of infected copepods, mollusks, or smaller fish.

Table 6

Organal Distribution of Helminth Parasites in Fishes (n=113)

Organ	No. of Parasites	Percentage (%)
Intestine	95	84.07
Stomach	52	46.01
Liver	20	17.69
Swim bladder	10	8.84

The intestine was the most heavily parasitized organ, with 84.07% of helminths recovered from this site. The stomach also exhibited a substantial parasite load (46.01%), followed by the liver (17.69%) and swim bladder (8.84%). These findings are consistent with previous observations that helminths favour the alimentary tract due to the abundance of nutrients and direct exposure to ingested infective stages (Parveen et al., 2020; Woo &Buchmann, 2012). The lower infection rates in the liver and swim bladder indicate the need for specialized adaptations by parasites to inhabit less hospitable environments.

Table 7
Mean Intensity and Relative Abundance of Helminth Parasites in Marine Fish Species

Fish Species	Fish Examined	Fish Infected	Parasites Recovered	Mean Intensity (MI)	Relative Abundance (RA)
Lutjanusargentimaculatus	35	30	51	1.70	1.46
Johniussdussumieri	40	36	68	1.89	1.70
Plectorhynchuscinctus	18	15	28	1.87	1.56
Pampus argenteus	20	14	30	2.15	1.50

The mean intensity of helminth infection was highest in *Pampus argenteus* (2.15 parasites per infected fish), even though's relatively lower prevalence compared to *Johnius dussumieri* and *Lutjanus argentimaculatus*. This indicates that while fewer *Pampus argenteus* individuals are infected, those that are tend to harbour a heavier parasite load. *Johniussdussumieri* and *Plectorhynchuscinctus* showed similar mean intensities (1.89 and 1.87, respectively), indicating comparable levels of parasite burden among infected individuals. Relative abundance followed similar trends, highlighting that parasitic infections are unevenly distributed across fish species (Poulin, 2007).

Summary of Key Observations

- **High Overall Prevalence**: 84.07% of fish were infected, indicating a heavily parasitized marine environment.
- **Nematodes Dominate**: The most abundant and prevalent helminth group detected were nematodes.
- **Species-Specific Susceptibility**: *Johniussdussumieri* was the most susceptible host, with consistently high infection rates across helminth groups.
- **Intestine as Primary Site**: The intestine was confirmed as the main site for parasite establishment.
- **Higher Mean Intensity in Pampus argenteus**: Suggests fewer infections but heavier burdens where present.
- **Environmental Influence**: Seasonal patterns and local ecological factors likely influence infection dynamics.

Discussion

The present study offers important new information on the distribution, frequency and intensity of helminth parasites among four commercially important marine fish species from the Karachi coast. Of the 113 fish specimens examined, *Lutjanus argentimaculatus, Johnius dussumieri, Plectorhynchus cinctus,* and Pampus *argenteus*84.07% were found to be infected with helminth parasites, which indicates a significant parasitic load in these marine populations.

Of the helminth groups found, nematodes were most common at 78.76%, followed by trematodes at 60.17%. Cestodes were much less common at 17.69%. Consistent with earlier results indicating that acanthocephalan distribution is usually limited by host specificity and environmental conditions, no acanthocephalans were noted (Scholz et al., 2018). The lack of acanthocephalans in this study may also indicate the lack of suitable intermediate hosts in the sampled environments.

There were clear species-specific patterns of infection. *Lutjanus argentimaculatus* came in second to *Johniusdussumieri*, who had the greatest infection rates for both nematodes (90.00%) and trematodes (70.00%). This links to previous research showing that piscivorous or benthic-feeding fish had greater helminth loads from regular contact with infected intermediate hosts (Rauque et al., 2018; Chai et al., 2005). *Johniussdussumieri's* feeding patterns, which include eating smaller fish, zooplankton, and benthic invertebrates, greatly raise its risk of parasitic infections.

Moreover, seasonal changes in parasitic prevalence were clear; summer saw the highest infection rates, and winter saw a significant drop. These results support earlier studies showing that higher water temperatures promote the growth, survival, and spread of many helminth forms (Genc et al., 2005; Gautam et al., 2018; Khalid et al., 2020). Warmer temperatures promote quicker parasite growth, more active intermediate host activity, and higher host metabolism, so raising infection risks (Marcogliese, 2001).

Parasite organal distribution was strongly biased toward the intestine, which accounted for 84.07% of infections. Most helminths' ecological adaptation—which uses the nutrient-rich gut environment for attachment, growth, and reproduction— fits this trend (Parveen et al., 2020; Woo &Buchmann, 2012). Though relatively less frequent, liver, stomach, and swim bladder infections were significant and may indicate tissue migration in certain helminth species (Gibson, 2016).

Pollution is one environmental element that could increase parasite frequency even more. Fish living in contaminated waters are known to have more parasite loads because of immunosuppression and more exposure to parasite intermediate hosts (Jasrotia& Kaur, 2017; Scholz et al., 2018). Urban runoff and industrial discharge are increasingly affecting Karachi's coastal waters, therefore possibly generating good conditions for parasite growth (Ayub & Jafri, 2018).

Host-specific elements are also quite important. Due to prolonged exposure and larger body surface areas providing more niches for parasite colonization, larger and older fish often host more parasites (Poulin, 2007). Moreover, the parasitofauna is an aggregation of parasites that live inside the host organism (Dallas et al., 2019).

According to Rashid et al. (2019), parasites can infect humans and cause problems that impact health and reproduction and symptoms that are easily noticeable include falling prey to predators and infection. Economic losses occur due to the high prevalence of parasites and stages, especially larval and intermediate mortalities(Narladkar, 2018). Additionally, some fish-borne helminths have zoonotic potential, emphasizing the importance of comprehensive monitoring and protective measures to guarantee seafood safety (Chai et al., 2005; Zhu et al., 2020).

Molecular identification of helminths should be included for future studies to increase species resolution, more thoroughly evaluate pollution-parasite interactions, and monitor temporal trends under the impact of climate change. Ecological indicators combined with parasitological data will increase the capability to control marine resources sustainably.

Conclusion

The aforementioned findings demonstrate that, across all fish species, summer had the greatest prevalence of infection during the current study, maybe due to the developmental parasitic cycles and host intermediate activity connected to higher temperatures. On the other hand, winter months showed the least prevalence, most likely as a result of the lower biological activity of parasites and hosts alike.

With the intestine being the main organ of infection, the study verifies that nematodes rule the helminthic community structure in the region, followed by trematodes and cestodes. Species-specific variations in infection patterns draw attention to the impact of host diet, behaviour, and ecological niche.

Particularly in light of the possible

consequences to human health and the sustainability of marine fisheries, the findings emphasize the need of continuous parasitological monitoring.Important next steps are tackling pollution, enhancing fish handling techniques, and increasing public knowledge of the dangers of eating raw or undercooked seafood.

These results taken together offer a starting point for future ecological, epidemiological, and public health studies as well as necessary baseline data for marine parasitology in Pakistan.

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