**Original Article** 

# New record of *Philometra* species from the marine edible fish *Terapon jarbua* collected from the Sindh, Arabian Sea, Pakistan

Novo registro de espécies de *Philometra* do peixe marinho comestível *Terapon jarbua* coletado em Sindh, Mar Arábico, Paquistão

R. A. Ghaffar<sup>a</sup>, S. Zulfiqar<sup>a</sup>, H. Ul Hassan<sup>a,b\*</sup> <sup>(10)</sup>, Y. A. J. Fadladdin<sup>c\*</sup> <sup>(10)</sup>, M. Sohail<sup>d</sup>, M. Kabir<sup>e</sup>, M. A. Raza<sup>f</sup>, N. Rafiq<sup>g</sup>, M. F. A. Abdel-Aziz<sup>h</sup>, Y. Garedaghi<sup>i</sup>, I. Hasan<sup>j</sup> and T. Arai<sup>k</sup>

<sup>a</sup>University of Karachi, Department of Zoology, Karachi, Pakistan

<sup>b</sup>Government of Pakistan, Ministry of National Food Security and Research, Fisheries Development Board, Islamabad, Pakistan

<sup>c</sup>King Abdulaziz University, Faculty of Sciences, Department of Biological Sciences, Jeddah, Saudi Arabia

<sup>d</sup>Government Postgraduate College Sahiwal, Department of Biology, Sahiwal, Pakistan

<sup>e</sup>Thal University Bhakkar (University of Sargodha, Ex-Sub-Campus Bhakkar), Department of Biological Sciences, Bhakkar, Punjab, Pakistan <sup>(</sup>Government Postgraduate College Satellite Town, Department of Biology, Gujranwala, Pakistan

<sup>g</sup>Abdul Wali Khan University Mardan, Department of Zoology, Khyber Pakhtunkhwa, Pakistan

<sup>b</sup>Arish University, Faculty of Aquaculture and Marine Fisheries, Department of Aquaculture and Biotechnology, Al-Arish, Egypt

Islamic Azad University Tabriz Branch, Faculty of Veterinary Medicine, Department of Parasitology, Tabriz Medical Sciences, Tabriz, Iran University of Insubria, Department of Biotechnology and Life Sciences, Varese, Italy

<sup>k</sup>Universiti Brunei Darussalam, Faculty of Science, Environmental and Life Sciences Programme, Gadong, Brunei

#### Abstract

Diseases in fish due to helminth parasites, especially *Philometra* species, are the primary worry in aquaculture. Philometra are responsible for health problem in fishes they directly affect fish growth and population parameters. A comprehensive survey was conducted involving the examination of the marine fish species *Terapon jarbua*, gathered from the coastal waters of Sindh, Pakistan In this research different *Philometra species* from marine fish *Terapon jarbua* during 2021 and 2022. Philometra nematodes, belonging to the family Philometridae, are common parasitic organisms inhabiting both marine and freshwater environments. Their prevalence, particularly when existing in high numbers within host organisms, can lead to severe and potentially lethal consequences. Employing light microscopy techniques, diverse species of Philometra were identified, including *Philometra teraponi*, *P. jarbuai*, *P. arabiai*, *P. karachii, and P. awarii*, localized primarily within the ovaries of the host fish. A total of 140 fish samples were examined and 76 were infected. The intensity of infected fish was 54.28%. The identification process encompassed meticulous analysis of crucial parameters, such as body size, esophagus length, positioning of the nerve ring, dimensions of the ventriculus, and ligament size. Intriguingly, the parasites were found in varying contexts; while some were free within the ovaries, others were embedded within tissues, inducing severe muscular dystrophy. This research presents novel findings of Philometra nematodes in the marine waters of Pakistan, extending their host and geographical distribution records. Future studies are needed to better evaluate and describe the dynamics and the epidemiology of Philometra infection in wild and cultured fish species

Keywords: fish Terapon jarbua, newly discovered, Philometra species, ventriculus, esophagus, nerve ring and ligament, Pakistan.

### Resumo

Doenças em peixes causadas por parasitas helmintos, especialmente espécies de Philometra, são a principal preocupação na aquicultura. Philometra é responsável por problemas de saúde em peixes, pois afeta diretamente o crescimento destes e os parâmetros populacionais. Foi realizada uma pesquisa abrangente envolvendo o exame das espécies de peixes marinhos Terapon jarbua, coletadas nas águas costeiras de Sindh, Paquistão. Nesta pesquisa, foram identificadas diferentes espécies de Philometra em peixes marinhos Terapon jarbua durante 2021 e 2022. Nematoides Philometra, pertencentes à família Philometridae, são organismos parasitas comuns que habitam ambientes marinhos e de água doce. A sua prevalência, especialmente quando existe em grande número nos organismos hospedeiros, pode levar a conseguências graves e potencialmente letais. Empregando técnicas de microscopia óptica, diversas espécies de Philometra foram identificadas, incluindo Philometra teraponi, P. jarbuai, P. arabiai, P. karachii e P. awarii, localizadas principalmente nos ovários do peixe hospedeiro. Um total de 140 amostras de peixes foram examinadas e 76 foram infectadas. A intensidade de peixes infectados foi de 54,28%. O processo de identificação envolveu análise meticulosa de parâmetros cruciais, como tamanho corporal, comprimento do esôfago, posicionamento do anel nervoso, dimensões do ventrículo e tamanho do ligamento. Curiosamente, os parasitas foram encontrados em contextos variados; enquanto alguns estavam livres nos ovários, outros estavam incorporados nos tecidos, induzindo distrofia muscular grave. Esta pesquisa apresenta novas descobertas de nematoides Philometra nas águas marinhas do Paquistão, ampliando seus registros de hospedeiros e distribuição geográfica. Estudos futuros são necessários para melhor avaliar e descrever a dinâmica e a epidemiologia da infecção por Philometra em espécies de peixes selvagens e cultivadas

Palavras-chave: peixe Terapon jarbua, recém-descoberto, espécie Philometra, ventrículo, esôfago, anel nervoso e ligamento, Paquistão.

\*e-mail: habib5447@gmail.com; habib.ulhassan@yahoo.com; yfadladdin@kau.edu.sa Received: December 13, 2023 – Accepted: February 15, 2024

This is an Open Access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

## 1. Introduction

Fish have long held a pivotal role as a crucial source of human nutrition as a source of essential nutrients and a significant role in feeding the world's growing population (Abidin et al., 2022). Healthy fish populations lead to healthy oceans, and it is our responsibility to be a part of the solution (Abdel-Ghaffar et al., 2008; Hussain et al., 2021; Khalid et al., 2021; Hassan et al., 2021, 2022a). The Arabian Sea of Pakistan is one of the most identical Sea worldwide and has diverse fish fauna (≈1,000 species) due to nutrient-rich waters (Hassan et al., 2022b). Throughout human history, fish consumption has been intertwined with sustenance and intelligence, as evident from their consumption since prehistoric eras. The marine fish Terapon jarbua, locally referred to as gangra, belongs to the Family Teraponidea and is commonly found in the shallow coastal waters of Pakistan. Nematodes, a diverse group of worms, encompass species that parasitize the body cavities and tissues of fish (Moravec et al., 2006). Despite the significance of nematode parasites, knowledge about their fauna in the Arabian Gulf remains limited (Al-Salim and Ali, 2011). Among the parasitic organisms that inhabit marine, brackish, and freshwater environments, the nematode genus Philometra (Nematoda: Phylometridae) has emerged as an intriguing subject of investigation. These nematodes are commonly found within the body cavities and tissues of fish, potentially exerting deleterious effects on their hosts when present in abundance (Ghaffar et al., 2024). Female specimens are larger than males, and adult nematodes migrate through the intestine wall, inhabiting the swim bladder serosa. Both male and female larvae develop to sexual maturity before mating.

Helminth infections in fish have a major impact on public health and sustainable fish production especially Philometra species, are the primary worry in aquaculture. There are definite problems caused by *Philometra*, which highly affect fish growth, function, reproduction and survival of the hosts. In cultured fish, however, parasitic diseases are generally more severe, and may cause important economic losses due to stock mortality, declined productivity and reduced marketability and population. Because of difficulties in studying Philometrid nematodes associated with their morphological and biological peculiarities, most of them remain poorly known, and their identification is usually difficult and problematic (Moravec and Justine, 2005; Ghaffar et al., 2024). Gonadinfecting parasitic nematodes of the family Philometridae have been reported to cause significant damage to the gonads of wild and cultured marine perciform hosts with varying consequences including severe reduction in reproductive fitness based on gross and histological pathology and mechanical damage of blood vessels with hemorrhaging and destruction of skeletal joints resulting in poor growth and emaciation (Moravec et al., 2006). A wide variety of parasite species are known to cause morbidity and mortality in fish. For instance, a number of digenean trematodes (Wise et al., 2013).

Despite their ubiquity, comprehensive studies on the diversity, distribution, and impact of Philometra species in specific marine fish populations remain relatively scarce. Philometra parasite outbreaks have enormous effects on fisheries, aquaculture, and population dynamics (Garedaghi and Mohammadi Hefz Abad, 2012). Philometrids, large filamentous viviparous nematodes, inhabit fish body cavities, subcutaneous tissues, and gonads (Mohamed, 2010). Philometra species, belonging to the Nematoda: Phylometridae, are found in sea and river waters and can have lethal effects when present in abundance within their host organisms (Perdikaris et al., 2003). Identification of these parasites is challenging due to their unique characteristics, leading researchers to recommend molecular and protein biology detection methods for phylogenetic identification (Garedaghi et al., 2011). The male Philometra species are typically much smaller than the elongated females, with males often remaining undiscovered (Moravec and Justine, 2005). The genus Philometra (Costa, 1845) has been investigated (Garedaghi and Mohammadi Hefz Abad, 2012) in Pakistan, specifically in relation to its parasitism of Priacanthus sp. Furthermore, Rizwana and Zulfiqar (2022) documented a new Philometra species in the ovaries of Epinephelus bleekeri from the Karachi coast. Building on this, Ghaffar et al. (2023) described a novel Philometra species infesting the gonads of Lutjanus argentimaculatus from Pakistan's seawater.

Previous research has highlighted the pathogenicity of Philometra species in fish ovaries. Moravec and Ali (2014) noted that Philometra margolisi can cause significant harm to ovaries, impacting host fecundity. Philometra floridensis (Nematoda: Philometridae) has been observed to harm the ovarian tissue of the host fish Sciaenops ocellatus, leading to tissue degeneration (Bakenhaster and Bullard, 2014). Infestations of Philometra sp. in certain marine fishes have been documented (Mohamed, 2010) revealing low infestation rates in muscle and fin tissue. However, the presence of dead parasites in ovarian tissues led to severe tissue degeneration, highlighting the potential harm of persistent infections. Philometra spp. females release first-stage larvae from the host's body cavity through tissues near the anus, causing hypotonic effects due to water osmotic differences. Severe infection can be highly pathogenic due to large female specimens causing severe damage to the coelomic organs, and consequently influencing fish reproduction (Wise et al., 2013).

In light of this, our research embarks on a pioneering endeavor to shed light on the presence and characteristics of Philometra species within the marine edible fish Terapon jarbua in the coastal waters of Sindh, Pakistan. By utilizing light microscopy and a comprehensive morphological analysis, we aim to identify and categorize the various Philometra species present within the host fish. Parameters such as body size, esophagus length, nerve ring positioning, ventriculus size, and ligament dimensions serve as key identifiers in this classification process. This study not only seeks to contribute to the broader understanding of the intricate relationship between marine organisms and their parasites but also strives to uncover novel insights into the diversity of Philometra species inhabiting the coastal waters of Sindh. Through the elucidation of their presence and characteristics, we endeavor to expand the records of both host species and geographical distribution, ultimately enriching our comprehension of the complex marine ecosystem dynamics.

## 2. Materials and Methods

In the course of the current investigation, a meticulous and comprehensive approach was employed to study the presence and characteristics of Philometra species within the marine edible fish *Terapon jarbua*, collected from the coastal waters of Karachi, Sindh, Pakistan. The methodology encompassed several well-defined steps to ensure accuracy, consistency, and reliability in the results obtained.

A total of 140 marine fishes were judiciously collected over an extended duration, ranging from February 2021 to January 2022. This extensive sampling period aimed to encompass seasonal variations in fish populations and potential fluctuations in parasite prevalence. Following collection, the collected fish specimens were promptly transported to the well-equipped Parasitology laboratory at the University of Karachi, where a detailed parasitological examination was conducted.

The examination process was far from cursory; it involved meticulous scrutiny of each specimen's internal anatomy to identify the presence of any potential pathological conditions. Notably, the focus was on detecting and characterizing Philometra species within the gonads of *Terapon jarbua*. To ensure the accuracy of subsequent analyses, precise measurements of each specimen's dimensions were taken. This process included weight assessment and comprehensive photographic documentation, contributing to a comprehensive record of the samples.

To facilitate the long-term preservation of the identified Philometra specimens, a solution comprising equal parts of 70% ethyl alcohol and glycerin was employed. This preservation method aimed to maintain the structural integrity of the specimens while allowing for future examinations and analyses. Furthermore, tissues hosting the identified parasites were meticulously fixed in formalin, a crucial step that prepared the samples for more in-depth studies.

The study of the identified nematodes involved careful preparations for enhanced visualization under light microscopy. Glycerin was utilized as a clearing agent, aiding in rendering the internal structures of the nematodes more visible and distinct. The utilization of the Zeiss drawing attachment added another layer of precision to the process, enabling the creation of detailed drawings that accurately captured the distinctive features of the newly discovered Philometra species. The accuracy and precision of the measurements taken were ensured by recording parasite characteristics in micrometers. This meticulous approach helped to prevent any inaccuracies that might arise due to approximation or rounding off of measurements. By adhering to such rigorous standards, the methodology aimed to provide an accurate and detailed portrayal of the size and dimensions of the studied parasites.

In essence, the methodology described above served as the cornerstone of our investigation into the identification and characterization of Philometra species within the marine edible fish Terapon jarbua. By embracing a systematic approach encompassing specimen collection, preservation, examination, and documentation, we aspired to contribute to the expanding body of knowledge concerning the diversity and distribution of these parasitic nematodes within the marine ecosystem of Sindh, Pakistan. The discovery of these nematodes within the marine waters of Pakistan signifies a novel addition to both host and geographical records. The identification process encompassed meticulous analysis of crucial parameters. Through this methodical endeavor, we aimed to shed light on the prevalence, characteristics, and potential impact of these parasites on marine fish populations, fostering a better understanding of the intricate interactions within the marine ecosystem. These parasitic nematodes triggered significant inflammatory reactions in the ovarian tissues, underlining the importance of control and prevention measures due to the extensive damage they cause. This research would be helpful for the documentation and enhancement of nematode fauna in Pakistan

### 3. Result and Discussion

From the ovaries of marine fish *Terapon jarbua* (Forsskal, 1775) five new *Philometra sp.* were discovered (Figure 1) namely: *Philometra teraponi, Philometra jarbuai, Philometra arabiai, Philometra karachii* and *Philometra awarii*. In overall research a total of 140 fish samples were examined, and 76 were infected. The intensity of infected fish was 54.28%. Family: Philometridae (Baylis & Daubney, 1926) Sub-family: Philometrinae (Baylis & Daubney, 1926) Genus: *Philometra* (Costa, 1845)



Figure 1. The (a) is Terapon jarbua and (b) is infected ovary showing Philometra sp. in marine edible fishes Terapon jarbua.

## 3.1. Philometra teraponi (n. sp.) (Figure 2a-d)

Host: *Terapon jarbua* (Forsskal, 1775) Area where collected: Karachi coast, Pakistan. Organ in host where found: Gonads (Ovary) Recovered specimen: 40

Male Unknown.

**Female morphology:** Examination of the ovaries of the marine fish species *Terapon jarbua* (Forsskål, 1775) revealed the presence of female Philometra parasites. The recovered specimens exhibited distinct morphological characteristics, with the anterior portion appearing smoother in texture. In contrast, the posterior portion displayed a broader and ovular shape in comparison to the anterior segment (see Figures 2a and 2b). For comprehensive reference, the morphological measurements of this particular species are provided in Table 1.

**Etymology**: The nomenclature assigned to this nematode parasite draws a direct connection to the host genus.

**Remarks:** The Philometra species identified in this study was found inhabiting *Terapon jarbua* within the marine environment off the coast of Karachi. Distinct from this discovery, other recorded instances include *Philometra mexicana* (Moravec and Salgado-Maldonado, 2007) discovered in the fish species *Epinephelus adscensionis* from the southern Gulf of Mexico; *Philometra arafurensis* (Moravec and Barton, 2018) identified within *Lutjanus sebae* off northern Australia; and *P. justinei* (Moravec et al., 2020)

found in Mullus surmuletus and documented in the Strait Marine Reserve of Bonifacio, Corsica, France, A comparative assessment reveals that the Philometra specimen recovered from the host fish in this study exhibits a notably elongated size when compared to P. mexicana, P. arafurensis, and P. justinei. Noteworthy distinctions include the larger anterior portion of the esophagus compared to the mentioned species. Furthermore, the overall length of the esophagus surpasses that recorded in *P. mexicana*. While the anterior portion of the recovered specimen is less elongated than that of P. arafurensis, its nerve ring is anteriorly situated in contrast to P. mexicana, P. arafurensis, and P. justinei. Additionally, the size of the ligament is greater when juxtaposed with P. mexicana and P. arafurensis. These detailed morphological disparities highlight the uniqueness of the Philometra species encountered in the present research, differentiating it from its counterparts and underscoring the importance of comprehensive morphological analysis for accurate species identification.

3.2. Philometra jarbuai (n. sp.) (Figures 3a-d)

Host: *Terapon jarbua* (Forsskal, 1775) Area where collected: Karachi coast, Pakistan Organ in host where found: Gonads (Ovary) Recovered specimen: 20 Male: Unknown **Female morphology:** 



**Figure 2.** The (a) is lateral view of cephalic extremity of *Philometra teraponi* (×10) and (b) is lateral view of cephalic extremity of *Philometra teraponi* (×10) and (c) drawing of lateral view of cephalic extremity of *Philometra teraponi* (d) drawing of lateral view of caudal extremity of *Philometra teraponi*.

Parasites	<i>Philometra teraponi</i> (n.sp.)	<i>Philometra mexicana</i> (Moravec and Barton, 2018)	<i>Philometra arafurensis</i> (Moravec and Salgado-Maldonado, 2007)	<i>Philometra justinei</i> (Moravec et al., 2006)
Host	Terapon jarbua	Epinephelus adscensionis	Lutjanus sebae	Mullus surmuletus
Country	Pakistan	Mexico	Australia	France
Gender	Female	Female	Female	Female
Length of parasite	(195-220)(0.99)	(178-230)(0.7-1.3)	(220)(0.816)	(196)(1.7)
Size of anterior inflation	(0.07-0.086) (0.127-0.155)	(0.06-0.12)(0.12-0.14)	(0.16)(0.15)	-
measurement of posterior cylindrical part of esophagus	(1.25-1.39)(0.099)	(0.9-1.1)	(1.39)	(1.27)(0.136)
Measurement of cephalic end	(0.23)	-	(0.27)	-
Measurement of caudal end	(0.13-0.17)	-	-	-
Location of nerve ring	(0.21)	(0.24-0.27)	(0.394)	(0.367)
Measurement of ventriculus	-	(0.03-0.04)(0.06-0.09)	(0.041)(0.109)	-
Measurement of ligament	(1.84)	(0.612-1.02)	(0.816)	-

Table 1. Philometra teraponi (n.sp.) is compared to previously known Philometra sp. (Costa, 1845).



**Figure 3.** The (a) is a lateral view of cephalic extremity of *Philometra jarbuai* ( $\times$ 10) and (b) is lateral view of caudal extremity of *Philometra jarbuai* ( $\times$ 10) and (c) drawing of lateral view of cephalic extremity of *Philometra jarbuai* and (d) is drawing of lateral view of caudal extremity of *Philometra jarbuai*.

The female Philometra exhibits an elongated, brownish body, characterized by a distinct and conspicuous dark-brown intestine that remains visible through the cuticle (refer to Figures 3a and 3b). Ovaries are discernible at both ends of the body, while the uterus extends to the anterior portion. The cephalic extremity presents a notably wider configuration than the caudal extremity, as depicted in Figures 3c and 3d. Detailed measurements of this species' morphological features can be found in Table 2.

**Etymology:** The assigned nomenclature for this recovered parasite is P. jarbuai, directly aligning with the species of the host fish.

# **Remarks:**

Other recorded species: Philometra species have been discovered within various fish species across different regions worldwide. The present parasitic specimen, however, represents a distinct finding within the marine waters of Pakistan, specifically within the Terapon jarbua species. Noteworthy among the recorded species are P. tenuicauda (Moravec et al., 2006) Philometra arafurensis (Moravec and Salgado-Maldonado, 2007). Philometra synagridis (Moravec et al., 2020) and P. dissimilis (Moravec and Ali, 2014). The present specimen presents unique morphological traits setting it apart from its counterparts. Notably, the body of the discovered parasite is elongated and slender in comparison to the species mentioned above. In contrast to the referenced species, the anterior portion of the esophagus is not notably enlarged and appears thinner. The esophagus, while not greater in size than that of *P. tenuicauda* and *P. arafurensis*, is larger than P. dissimilis. The position of the nerve ring is situated closer to the anterior end than in *P. tenuicauda* and *P. arafurensis*. Moreover, the ventriculus is both larger and more slender. The ligament, in this case, exceeds the dimensions observed in other species. Finally, the cephalic end is broader than that of *P. tenuicauda*, *P. arafurensis*, and *P. synagridis*.

Based on these distinctive variations in morphological characteristics, the diagnosed *Philometra* specimen has been assigned the new species name *P. jarbuai*, serving as a testament to its uniqueness within the context of its counterparts.

3.3. Philometra arabiai (n. sp.) (Figures 4a-d)

Host: *Terapon jarbua* (Forsskal, 1775) Area where collected: Karachi coast, Pakistan. Organ in host where found: Gonads (Ovary) Recovered specimen: 5 Male: Unknown.

**Female Morphology:** The observed parasite exhibits a distinct morphology characterized by its slender, elongated form and a deep reddish appearance, largely attributed to the brown hue of its intestine. Notably, the posterior end of the parasite is wider than its anterior counterpart. A striking feature includes the presence of dual ovaries, and spherical eggs can be observed within the uterus (refer to Figures 4a and 4b). Comprehensive measurements of the morphological attributes of this species are documented in Table 3.

**Etymology**: The parasite encountered during the course of this research has been designated as Philometra arabiai. This name is derived from the habitat of the host organism, highlighting its association with the Arabian region.

#### Remarks

The current parasite's presence is documented within the ovaries of *Terapon jarbua*, sourced from the sea waters of Karachi.

 Table 2. Philometra jarbuai (n.sp.) is compared to previously known Philometra sp. (Costa, 1845).

Parasites	<i>P. jarbuai</i> (n. sp.)	<i>P.tenuicauda</i> (Moravec and Justine, 2009)	<i>P.arafurensis</i> (Moravec and Barton, 2018)	<i>P.synagridis</i> (Moravec et al., 2020)	<i>P.dissimilis</i> (Moravec and Manoharan, 2016)
Host	Terapon jarbua	Lagocephalus sceleratus	Lutjanus sebae	Lutjanus synagris	Johnius belangerii
Country	Pakistan	New Caledonia	Australia	Florida coast	India
Gender	Female	Female	Female	Female	Female
Length of parasite	(247.7-253) (04-0.5)	(290)(1.42)	(220)(0.816)	(152)(0.816)	(171)(1.29)
Measurement of anterior inflation	(0.08-0.12) (0.094-0.106)	(0.109)(0.12)	(0.16)(0.15)	(0.163)(0.150)	(0.109)
Measurement of posterior cylindrical part of esophagus	(1.23-1.34) (0.07-0.15)	(1.41)	(1.39)	-	(1.16)
Measurement of Intestine	(248-249.7) (0.106-0.254)	-	-	-	-
Measurement of cephalic end	(0.408-0.469)	(0.381)	(0.27)	(0.272)	-
Measurement of caudal end	(0.36-0.44)	-	-	-	-
Location of Nerve ring	(0.25-0.32)	(0.367)	(0.394)	(0.313)	(0.340)
Measurement of ventriculus	(0.055-0.061) (0.063-0.08)	(0.041)(0.109)	(0.041)(0.109)	(0.041)(0.136)	(0.041)(0.109)
Ligament	(1.56-1.63)	(0.816)	(0.816)	(0.933)	(1.238)



**Figure 4.** The (a) is a lateral view of cephalic extremity of *Philometra arabiai* ( $\times$ 10) and (b) is lateral view of caudal extremity of *Philometra arabiai* ( $\times$ 10) and (c) is drawing of lateral view cephalic extremity of *Philometra arabiai* (d) drawing of lateral view caudal extremity of *P.arabiai*.

Table 3.	. Philometra	arabiai is co	ompared to	previously	known species	of the genus	Philometra (	Costa,	1845)
----------	--------------	---------------	------------	------------	---------------	--------------	--------------	--------	-------

Parasites	<i>Philometra arabiai</i> (n. sp.)	<i>P.brachiri</i> (Moravec and Ali, 2014)	<i>P.haemulontis</i> (Moravec et al., 2020)	<i>P.psettoditis</i> (Moravec et al., 2012)	<i>P.mawsonae</i> (Moravec and Barton, 2018)
Host	Terapon jarbua	Brachirus orientalis	Haemulon plumierii and Haemulo aurolineatum	Psettodes erumei	Lutjanus malabaricus
Country	Pakistan	Iraq	Florida coast	Indonesia	Australia
Gender	Female	Female	Female	Female	Female
Length of parasite	(72-75.2)(0.4-0.7)	(65-70)(0.5-0.78)	(78-110)(0.4-0.7)	(68)(0.29)	(70-111)(0.7-0.9)
Measurement of anterior inflation	(0.065-0.072) (0.04-0.2)		(0.078-0.09) (0.06-0.07)	(0.096)(0.078)	(0.1-0.2)(0.1-0.151)
Measurement of posterior cylindrical part of esophagus	(17.95-19.11) (0.063-0.08)	(0.93)	(1.09-1.43)	(1.35)	(1.2-1.4)
Intestine length	(67.6)(0.08-0.106)	-	-	-	-
Measurement of cephalic portion	(0.079-0.113)	(0.136)	(0.109-0.19)	(0.95)	(0.15-0.23)
Measurement of caudal portion	(0.13-0.15)	(0.19)	-	-	(0.204-0.544)
Location of nerve ring	(0.13-0.181)	(0.286)	(0.258-0.367)	(0.326)	(0.29-0.34)
Measurement of ventriculus	-	(0.03)(0.07)	(0.03-0.04) (0.05-0.08)	(0.009)(0.03)	(0.02-0.05) (0.081-0.1)
Measurement of Ligament	(2.39)	(1.8-5.1)	(0.47-1.2)	-	(0.49-1.15)

This discovery is noteworthy among other recorded species, which include *P. brachiri* (Moravec and Ali, 2014) *P. haemulontis* (Moravec et al., 2020) *P. psettoditis* (Moravec and Barton, 2018) and *P. mawsonae* (Moravec et al., 2012). Distinct morphological disparities are evident among these recorded *Philometra* species. Notably, the discovered parasite is larger in size compared to *P. brachiri*, *P. psettoditis*, and *P. mawsonae*. The oesophageal bulb of the observed specimen is smaller than that of *P. haemulontis*, *P. psettoditis*, and *P. mawsonae*, yet wider than in *P. haemulontis* and *P. psettoditis*. Importantly, the length of the oesophagus significantly exceeds that of the previously mentioned species. In contrast to the aforementioned species, the nerve ring encircling the oesophagus is situated closer to the anterior end.

While the caudal end is slightly smaller than that of *P. mawsonae*, the cephalic end does not surpass the dimensions of the aforementioned species. Notably, the intestinal ligament is larger than that observed in any nearby species. Based on the distinctive variations in these morphological traits, this species has been distinguished as a new entity and designated the name *Philometra arabiai*. This nomenclature aptly reflects the species' connection to the Arabian habitat of its host.

# 3.4. Philometra karachii (n.sp.) (Figures 5a-d)

Host: *Terapon jarbua* (Forsskal, 1775) Area where collected: Karachi coast, Pakistan Organ in host where found: Gonads (Ovary) Recovered specimen: 55 Male Unknown

**Female Morphology:** The observed parasite stands out due to its distinct brown intestine, which is a noteworthy feature. The cuticle on the cephalic portion exhibits striations, whereas the cuticle on the caudal portion appears smooth (as shown in Figure 5a). Upon entering the intestine, the esophagus bifurcates. Both ends of the parasite host ovaries, while the uterus contains eggs. The anterior extremity of the parasite is broader than its posterior counterpart (refer to Figure 5b). Detailed measurements of the species' morphological characteristics are cataloged in Table 4.



**Figure 5.** The (a) is a lateral view of cephalic extremity of *Philometra karachii* (×10) and (b) is lateral view of caudal portion of *Philometra karachii* (×4) sand (c) drawing of lateral view of cephalic extremity of *P. karachii* and (d) is drawing of lateral view of caudal extremity of *P. karachii*.

**Etymology**: The parasite has been identified as *Philometra karachii*. This precise nomenclature is derived from the location of the host fish, further solidifying its association with the marine waters of Karachi.

# **Remarks:**

Other-species: The genus Philometra encompasses a diverse array of parasites observed across the globe. Among these, Philometra karachii has been identified within the marine waters of Pakistan, specifically from the host fish *Terapon jarbua*. Notable among the documented species are P. carolinensis (Moravec et al., 2006) and P. kiddakoi (Moravec et al., 2017). Comparative examination reveals distinctive traits within Philometra karachii. Notably, its length is smaller than that of Philometra kidakoi sp. The oesophagus of Philometra karachii is narrower and thinner than in the previously mentioned species, as is the length of its anterior portion. In contrast, it is wider at both the caudal and cephalic ends compared to Philometra kidakoi sp. Moreover, a larger ventriculus is observed in Philometra karachii in comparison to Philometra kidakoi. These unique morphological distinctions highlight the specific characteristics of Philometra karachii, differentiating it from its counterparts within the same genus.

3.5. Philometra awarii (n.sp.) (Figures 6a-d)

Host: *Terapon jarbua* (Forsskal, 1775) Area where collected: Karachi coast, Pakistan Organ in host where found: Gonads (Ovary) Recovered specimen: 75 Male Unknown

**Female morphology:** The identified parasite is characterized by its elongated, transparent, and cuticle-free appearance. The anterior end is flattened, while the posterior portion presents a rounded configuration. Of

significance, the ovary can be observed lying longitudinally within the anterior part, while it coils within the posterior portion. Notably, eggs are present within the uterus (as depicted in Figures 6a and 6b). Comprehensive measurements of this species' morphological attributes are outlined in Table 5.

**Etymology**: The parasite has been designated as *Philometra awarii*. This specific nomenclature draws a direct connection to the renowned city structure where the host fish was discovered, further solidifying its association with its habitat.

### Remarks

Other Discovered Species: Among the recorded species, notable ones include P. brachiri (Bakenhaster and Bullard, 2014) *Philometra rara* (Moravec et al., 2017) and P. haemulontis (Moravec et al., 2020), In Philometra awarii (n.sp.), several morphological distinctions are apparent. The length of the anterior portion of the esophagus is not significantly larger when compared to the aforementioned species. The esophagus itself is elongated compared to *P. brachiri* but smaller than both Philometra rara and P. haemulontis. The cephalic portion is wider than in P. brachiri and Philometra rara. However, the nerve ring is not anteriorly located, differentiating it from Philometra rara and P. haemulontis. The ventriculus is elongated in contrast to P. brachiri and P. haemulontis. Notably, the ligament is elongated when juxtaposed with the previously mentioned species. Given these distinctive variations in morphological features, the identified parasite is indeed a new species, designated as Philometra awarii. This recognition underscores the importance of comprehensive morphological analysis in distinguishing and cataloging different species within the Philometra genus.

Table 4. Philometra karachii is compared to previously known Philometra sp. (Costa, 1845).

Parasites	<i>P. karachii</i> (n.sp.)	<i>P.carolinensis</i> (Moravec et al., 2006)	<i>P.kiddakoi</i> (Moravec et al., 2019)
Host	Terapon jarbua	Cynoscion nebulosus	Gymnothorax kidako
Country	Pakistan	South Carolina, USA	Japan
Gender	Female	Female	Female
Length of parasite	(114.8-130)(0.73-0.78)	(115-156)(0.56-0.65)	(135)(1.4)
Measurement of anterior inflation	(0.058-0.09)(0.131-0.129)	(0.66-0.63)(0.57-0.6)	(0.09-0.15)
Measurement of posterior cylindrical part of esophagus	(1.07-1.18)(0.096-0.110)	(1.10-1.25)	(1.44)(0.136)
Measurement of cephalic end	(0.19-0.24)	-	(0.298)
Measurement of caudal end	(0.297-0.387)	(0.17)(0.24)	(0.68)
Location of nerve ring (mm)	(0.331-0.37)	(0.204-0.286)	(0.35)
Measurement of ventriculus	(0.06-0.105)	-	(0.03)(0.09)
Measurement of ligament	(2.462-2.721)	(0.03)	(0.68)
Vulval location	(12.3-13) anterior	-	-



**Figure 6.** The (a) is a lateral view of cephalic extremity of *Philometra awarii* (×10) and (b) is lateral view of caudal extremity of *Philometra awarii* (×10) and (c) is drawing of lateral view of cephalic end of *P. awarii* and (d) and drawing of lateral view of caudal end of *P. awarii*.

Parasites	<i>Philometra awarii</i> (n.sp.)	<i>P.brachiri</i> (Moravec and Ali, 2014)	<i>P.rara</i> (Moravec et al., 2017)	<i>P.haemulontis</i> (Moravec et al., 2020)	
Host	Terapon jarbua	Brachirus orientalis	Hyporthodus haifensis	Haemulon plumierii and H. aurolineatum	
Country	Pakistan	Iraq	Libya	Florida coast	
Gender	Female	Female Female		Female	
Length of parasite	(63.6-73) (0.4-0.662)	(65-70)(0.5-0.8)	(70)(0.69)	(78-110)(0.4-0.7)	
Measurement of anterior inflation	(0.06-0.083)	(0.08-0.16)(0.07-0.21)	(0.057)(0.099)	(0.078-0.09)(0.06-0.07)	
Measurement of posterior cylindrical part of esophagus	(0.89-1.042) (0.05-0.091)	(0.7-1.4)(0.08-0.16)	(1.04)	(1.09-1.43)	
Measurement of cephalic end	(0.216-0.243)	(0.12-0.21)	(0.218)	-	
Measurement of caudal end	(0.246-0.241)	-	-	-	
Postion of nerve ring	(0.27-0.307)	(0.25-0.28)	(0.204)	(0.258-0.367)	
Measurement of ventriculus	(0.044-0.054) (0.0818-0.0926)	(0.018-0.03)(0.05-0.08)	(0.041)(0.082)	(0.03-0.04)(0.05-0.08)	
Measurement of ligament	(0.614-0.638)	(0.5-3.5)	(0.462)	(0.47-1.2)	
Vulval position	(6.01-6.48)	-	-	-	

Table 5. Philometra awarii is compared to previously known Philometra sp. (Costa, 1845).

#### 4. Conclusion and Suggestions

In conclusion, the discovery of these nematodes within the marine waters of Pakistan signifies a novel addition to both host and geographical records. Environmental effects include imbalances in marine environment and the dumping of untreated sewage is also the main cause behind the outbreak of these parasites. Moreover, fish infested with parasites may put humans at risk for foodborne infections and other health problems. Philometra parasite outbreaks have enormous effects on fisheries, aquaculture, and population dynamics. Philometra infestations in fisheries can result in lower fishery yields because the parasites weaken the fish's reproductive organs, which ultimately limit the number of fish that are available for fisheries. Given the significant detrimental impacts caused by these parasites, their control and prevention hold paramount importance. The Philometrid nematodes are still poorly understood due to the challenges in researching them, brought on by their unique physical and biological characteristics due to this their identification is frequently challenging for researchers. The need for proactive measures to manage these parasites is evident. In light of these findings, it is highly recommended that efforts be directed towards the exploration, identification, and reporting of new and existing species of Philometra nematodes across various geographical regions within Pakistan and beyond. By extending research efforts, we can better understand the distribution and prevalence of these fish parasites, ultimately contributing to their effective eradication. This collaborative approach holds potential to safeguard marine ecosystems and the fish populations they sustain. This research would be helpful for the documentation and enhancement of nematode fauna in Pakistan. The growing global consumption and diversity of seafood, especially raw or undercooked fish, raises the danger of parasitic disease transmission. To solve today's difficulties, understanding the biology and ecology of these parasites in multidisciplinary research is required to address the pathogenicity of these ever-changing environments.

#### Acknowledgements

The authors are thankful for all the support from the Department of Zoology, University of Karachi for laboratory space and logistics. This study was financially supported by Universiti Brunei Darussalam under the Faculty/Institute/Center Research Grant (No. UBD/ RSCH/1.4/FICBF(b)/2023/057) and (No. UBD/RSCH/1.4/ FICBF(b)/2023/060).

## References

ABDEL-GHAFFAR, F., EL-TOUKHY, A., AL-QURAISHY, S., AL-RASHEID, K., ABDEL-BAKI, A.S., HEGAZY, A. and BASHTAR, A.R., 2008. Five new myxosporean species (*Myxozoa: Myxosporea*) infecting the Nile tilapia Oreochromis niloticus in Bahr Shebin, Nile Tributary, Nile Delta Egypt. Parasitology Research, vol. 103, no. 5, pp. 1197-1205. http://dx.doi.org/10.1007/s00436-008-1116-z. PMid: 18688646.

- ABIDIN, Z.U., HASSAN, H.U., MASOOD, Z., RAFIQUE, N., PARAY, B.A., GABOL, K., SHAH, M.I.A., GULNAZ, A., ULLAH, A., ZULFIQAR, T. and SIDDIQUE, M.A.M., 2022. Effect of dietary supplementation of neem, Azadirachta indica leaf extracts on enhancing the growth performance, chemical composition and survival of rainbow trout, Oncorhynchus mykiss. Saudi Journal of Biological Sciences, vol. 29, no. 4, pp. 3075-3081. http://dx.doi.org/10.1016/j. sjbs.2022.01.046. PMid:35531238.
- AL-SALIM, N.K. and ALI, A.H., 2011. First record of three nematode species parasitized some marine fishes in Iraq. Journal of Basrah Researches (Sciences), vol. 37, pp. 17-26.
- BAKENHASTER, M.D. and BULLARD, S.A., 2014. Population dynamics and host-parasite interactions of *Philometra* overstreeti (Nematoda: Philometridae) in the ovaries of red drum, *Sciaenops ocellatus*, from the northern Gulf of Mexico. *Parasitology Research*, vol. 113, no. 3, pp. 937-947.
- GAREDAGHI, Y. and MOHAMMADI HEFZ ABAD, M., 2012 [viewed 13 Dec 2023]. A case-report of *Chalcalburnus chalcoides* parasitic infections to ligula intestinalis in saungar- dam of gilan province. *Veterinary Clinical Pathology* [online], vol. 6, no. 2, pp. 1579-1582. Available from: https://sid.ir/ paper/183119/en
- GAREDAGHI, Y., KHAYATNOURI, M.H., KAKEKHANI, S. and NAZERI, M., 2011. Survey on experimental contamination to *lchthyophtirius multifilis* in cultural Rainbow trout consequently vaccination with Aquavac garvetil. *Journal of Animal and Veterinary Advances*, vol. 10, no. 11, pp. 1473-1476. http://dx.doi.org/10.3923/ javaa.2011.1473.1476.
- GHAFFAR, A., BILQEES, F.M. and PERVEEN, S.A., 2023. new Philometra (Nematoda: Philometridae) from gonads of *Lutjanus argentimaculatus* from seawater of Pakistan. *Pakistan Journal of Marine Sciences.*, vol. 32, no. 2, pp. 195-200.
- GHAFFAR, R., HASSAN, H.U., KAMIL, N., WATTOO, J., SULTANA, S., ABDALI, U. and ARAI, T., 2024. Occurrence of new Philometra sp. in mangrove red snapper Lutjanus argentimaculatus from the Karachi coast, Pakistan. Brazilian Journal of Biology = Revista Brasileira de Biologia, vol. 84, pp. e269504. http://dx.doi. org/10.1590/1519-6984.269504.
- HASSAN, H.U., ALI, Q.M., KHAN, W., MASOOD, Z., ABDEL-AZIZ, M.F.A., SHAH, M.I.A., GABOL, K., WATTOO, J., MAHMOOD CHATTA, A., KAMAL, M., ZULFIQAR, T. and HOSSAIN, M.Y., 2021. Effect of feeding frequency as a rearing system on biological performance, survival, body chemical composition and economic efficiency of Asian Seabass Lates calcarifer (Bloch, 1790) reared under controlled environmental conditions. Saudi Journal of Biological Sciences, vol. 28, no. 12, pp. 7360-7366. http://dx.doi.org/10.1016/j.sjbs.2021.08.031. PMid:34867038.
- HASSAN, H.U., ALI, Q.M., SIDDIQUE, M.A.M., HASAN, M.R. and HOSSAIN, M.Y., 2022a. Effects of dietary protein levels on growth, nutritional utilization, carcass composition and survival of Asian seabass *Lates calcarifer* (Bloch, 1790) fingerlings rearing in net cages. *Thalassas*, vol. 38, no. 1, pp. 21-27. http://dx.doi. org/10.1007/s41208-021-00371-8.
- HASSAN, H.U., MAWA, Z., AHMAD, N., ZULFIQAR, T., SOHAIL, M., AHMAD, H., YAQOOB, H., BILAL, M., RAHMAN, M.A., ULLAH, N., HOSSAIN, M.Y., HABIB, T. and ARAI, T., 2022b. Size at sexual maturity estimation for 36 species captured by bottom and mid-water trawls from the marine habitat of Balochistan and Sindh in the Arabian Sea, Pakistan, using maximum length (Lmax) and logistic (L50) models. *Brazilian Journal of Biology = Revista Brasileira de Biologia*, vol. 84, pp. e262603. http://dx.doi. org/10.1590/1519-6984.262603. PMid:35857953.

- HUSSAIN, M., HASSAN, H.U., SIDDIQUE, M.A.M., MAHMOOD, K., ABDEL-AZIZ, M.F.A., LAGHARI, M.Y., ABRO, N.A. and GABOL, K., 2021. Effect of varying dietary protein levels on growth performance and survival of milkfish *Chanos chanos* fingerlings reared in brackish water pond ecosystem. *Egyptian Journal* of Aquatic Research, vol. 47, no. 3, pp. 329-334. http://dx.doi. org/10.1016/j.ejar.2021.05.001.
- KHALID, S., KHAN, W., DAS, S.N., AHMAD, A., MEHMOOD, S.A., PAHANWAR, W.A., AHMED, S., KAMAL, M., WAQAS, M., WAQAS, R.M., HASSAN, H.U., ZAHOOR, S. and MAQBOOL, A., 2021. Evaluation of ecto and endo parasitic fauna of *Schizothorax plagiostomus* inhabitants of river Swat, Khyber PakhtunKhwa, Pakistan. *Brazilian Journal of Biology = Revista Brasileira de Biologia*, vol. 81, no. 1, pp. 98-104. http://dx.doi.org/10.1590/1519-6984.222215. PMid:32578670.
- MOHAMED, A.H., 2010. Contribution to the knowledge of Philometra spp. (Nematoda: Philometridae) parasitic in marine fish in Iraq. *Acta Parasitologica*, vol. 55, no. 2, pp. 177-182.
- MORAVEC, F. and ALI, A.H., 2014. Additional observations on *Philometra spp.* (Nematoda: Philometridae) in marine fishes off Iraq, with the description of two new species. *Acta Parasitologica*, vol. 87, no. 3, pp. 259-271. http://dx.doi.org/10.1007/s11230-014-9475-6. PMid:24563142.
- MORAVEC, F. and BARTON, D.P., 2018. New records of Philometrids (Nematoda: Philometridae) from marine fishes off Australia, including description of four new species and erection of *Digitiphilometroides gen. n. Folia Parasitologica*, vol. 65, pp. 5. PMid:29687788.
- MORAVEC, F. and JUSTINE, J.L., 2005. Two species of *Philometra* (Nematoda, Philometridae) from serranid fishes off New Caledonia. *Acta Parasitologica*, vol. 50, pp. 323-331.
- MORAVEC, F. and JUSTINE, J.L., 2009. New data on dracunculoid nematodes from fishes off New Caledonia, including four new species of *Philometra* (Philometridae) and *Ichthyofilaria* (Guyanemidae). *Folia Parasitologica*, vol. 56, no. 2, pp. 129-142. http://dx.doi.org/10.14411/fp.2009.017. PMid:19606788.
- MORAVEC, F. and MANOHARAN, J., 2016. *Philometra dissimilis* n. sp. from the ovary of *Johnius belangerii* (Sciaenidae) and other new records of philometrids (Nematoda: Philometridae) from fishes of the Bay of Bengal, India. *Helminthologia*, vol. 53, no. 2, pp. 133-141. http://dx.doi.org/10.1515/helmin-2016-0004.
- MORAVEC, F. and SALGADO-MALDONADO, G., 2007. A.new species of *Philometra* (Nematoda, Philometridae) from the gonads of the rock hind *Epinephelus adscensionis* (Osteichthyes) from the southern Gulf of Mexico. *Acta Parasitologica*, vol. 52, no. 4, pp. 376-381. http://dx.doi.org/10.2478/s11686-007-0044-x.

- MORAVEC, F., BAKENHASTER, M. and SWITZER, T.S., 2020. New records of *Philometra spp*. (Nematoda: Philometridae) from marine perciform fishes off Florida, USA, including descriptions of two new species. *Folia Parasitologica*, vol. 67, pp. 017. http://dx.doi.org/10.14411/fp.2020.017. PMid:32764188.
- MORAVEC, F., BURON, I. and ROUMILLAT, W.A., 2006. Two new species of *Philometra* (Nematoda: Philometridae) parasitic in the perciform fish *Cynoscion nebulosus* (Sciaenidae) in the estuaries of South Carolina, USA. *Folia Parasitologica*, vol. 53, no. 1, pp. 63-70. http://dx.doi.org/10.14411/fp.2006.006. PMid:16696433.
- MORAVEC, F., CHAABANE, A., NEIFAR, L., GEY, D. and JUSTINE, L., 2017. Species of *Philometra* (Nematoda, Philometridae) from fishes off the Mediterranean coast of Africa, with a description of *Philometra rara* n. sp. from *Hyporthodus haifensis* and a molecular analysis of *Philometra saltatrix* from *Pomatomus saltatrix*. *Parasite* (*Paris*, *France*), vol. 24, pp. 8. PMid:28287390.
- MORAVEC, F., NAGASAWA, K. and NOHARA, K., 2012. Two species of philometrid nematodes (Philometridae) from marine fishes off Japan, including Philometroides *branchiostegi* sp. n. from *Branchiostegus japonicus* (Malacanthidae). *Folia Parasitologica*, vol. 59, no. 1, pp. 71-78. http://dx.doi.org/10.14411/fp.2012.011. PMid:22439431.
- MORAVEC, F., NAGASAWA, K., NITTA, M. and TAWA, A., 2019. New records of philometrids (Nematoda: Philometridae) from marine fishes off japan, including description of *Philometra kidakoi sp. n.* and *Congerinema japonicum gen.et sp. n. Folia Parasitologica*, vol. 66, no. 21, pp. 1-17. http://dx.doi.org/10.14411/fp.2019.021. PMid:31849364.
- PERDIKARIS, C., GRIGORIOU, P. and LEONARDOS, I., 2003. Infestation of sand smelt (*Atherina boyeri* and *Atherina hespetus*) by nematode parasites of the species *Philometra tauridica Ivaskin*, in the North Aegean Sea. In: *Proceedings of 7th Symposium of Oceanography and Fisheries*, May 2003, Hersonissos, Crete. Hersonissos, vol. 184.
- RIZWANA, A.G. and ZULFIQAR, S., 2022. new species of Philometra reported from the ovaries of edible fish *Epinephelus bleekeri* (Vaillant 1878) of the Karachi coast. *Pakistan Journal of Marine Sciences*, vol. 31, no. 2, pp. 121-127.
- WISE, D.J., LI, M.H., GRIFFIN, M.J., ROBINSON, E.H., KHOO, L.H., GREENWAY, T.E., BYARS, T.S., WALKER, J.R. and MISCHKE, C.C., 2013. Impacts of *Bolbophorus damnificus* (*Digenea: Bolbophoridae*) on production characteristics of Channel catfish, *Ictalurus punctatus*;raised in experimental ponds. *Journal of the World Aquaculture Society*, vol. 44, no. 4, pp. 557-564. http://dx.doi. org/10.1111/jwas.12060.