DOI: 10.7860/JCDR/2026/80669.23289

Original Article

Microbiology Section

Incidence of *Paragonimus* Metacercariae in Freshwater Crabs in Ammapettai Village: A Pilot Study

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ABSTRACT

Introduction: *Paragonimus westermani* is a significant lung fluke parasite that affects humans by causing a disease called paragonimiasis, transmitted through the consumption of inadequately cooked or pickled freshwater crabs (the second intermediate host) containing metacercariae. This infection is one of the many neglected tropical foodborne parasitic zoonoses found in various parts of the world, particularly in developing countries. It continues to pose a significant public health challenge in regions where such practices of eating undercooked or improperly prepared seafood are common.

Aim: To explore *Paragonimus* metacercariae infestation in freshwater crabs in a rural area.

Materials and Methods: This pilot study involved a survey to find freshwater crabs, which are intermediate hosts for *Paragonimus westermani*, in Ammapettai village over three months from October 2019 to December 2019. Crab consumption is common in this area. A total 80 crabs were collected from ponds, paddy fields, and lakes. Crabs were humanely euthanized by piercing the sternum; legs and claws were then removed. The shell (carapace) was lifted off and discarded. Each crab was carefully dissected, and its body parts- main shell (carapace), claws (chelipeds), and legs (periopods)- were examined separately for parasite infestation. Statistical analysis using the chi-square test was performed to check if there was any significant difference in parasite infestation between different parts of the crab collected in rural area.

Results: Microscopic examination revealed an overall incidence of *Paragonimus* metacercarial infestation in five of 80 (6.25%) crabs. Among other parasites, only monogeneans were detected in two crabs that constitute 2.5% of the total numbers.

Conclusion: The results of the research are helpful for predicting the future risks from the pulmonary paragonimiosis in humans.

Keywords: Foodborne parasites, Intermediate hosts, Lung fluke

INTRODUCTION

Paragonimus westermani is a significant lung fluke that infects humans. It is acquired by consuming inadequately cooked or pickled freshwater crabs (the second intermediate host) containing metacercariae. The infection, known as paragonimiasis or pulmonary paragonimiasis, commonly results in haemoptysis and other respiratory issues. It is one of several neglected tropical foodborne parasitic zoonoses found in various parts of the world, particularly in developing countries [1]. *P. westermani* completes its life cycle in three hosts: one definitive host and two intermediate hosts. The definitive hosts include humans, tigers, and leopards, while the intermediate hosts are freshwater snails and freshwater crabs or crayfish. The adult worms reside in the respiratory tract of the definitive host, where their eggs are typically found in sputum and faeces.

When the raw flesh of an infected crab or crayfish is consumed, the cyst wall is dissolved by gastric juices, releasing metacercariae into the duodenum. The young adult worms then penetrate the intestinal wall, migrate through the diaphragm, and pass through the pleura to reach the lungs, where they attain sexual maturity. Once mature, the worms release eggs into the bronchioles, which are expelled in the sputum, continuing the cycle of infection among the hosts. The movement of adult worms causes mechanical damage, leading to the formation of lesions, including worm cysts and burrows. Additionally, the presence of eggs triggers a foreign body granulomatous reaction, which may eventually soften and create cavities [2].

The clinical manifestations of this infection can be broadly classified into pulmonary and extrapulmonary types, with cases of pulmonary paragonimiasis, particularly those with haemoptysis, frequently being misdiagnosed as pulmonary tuberculosis and extrapulmonary paragonimiasis refers to infection occurring outside lungs, where the parasite migrates to other organs. These forms are clinically significant because they often mimic conditions such as tumours and tuberculosis, leading to misdiagnosis. The major extrapulmonary types include cerebral, cutaneous, pleural, pericardial, abdominal, ocular, and spinal paragonimiasis [3].

Paragonimiasis is most commonly found in various regions of Asia, Africa, South America, including China, Japan, Korea, and the Philippines [4]. In India, literature reviews suggest that paragonimiasis is a highly prevalent yet neglected disease, it is frequently encountered as a trematode infection in wild animals. *Paragonimus westermani* is typically found in the lung parenchyma of infected animals and has been reported during autopsies of tigers in several locations, such as Corbett National Park in Uttar Pradesh (1988), Kanha National Park in Madhya Pradesh, Vandalur Zoological Park in Tamil Nadu (2000), and Assam State Zoo in Guwahati (2001). The first recorded case of *Paragonimus* ova in a human was in Maharashtra in 1919, in a Chinese patient's sputum and faecal samples, suggesting the infection may have been acquired outside India. The first indigenous human case of paragonimiasis in India was reported in Manipur in 1981, followed by additional cases in Maharashtra in 1984 and Manipur in 1986 [5].

The crab eaters are present throughout in India, but still believed that Manipur, Arunachal Pradesh and Nagaland North India identified as endemic foci. The crab eaters were more in Tamil Nadu but actual burden of these infections is still underestimated. There is limited information available from many pockets within the endemic regions of India [5,6].

The earlier studies from the South Part of India, especially in Tamil Nadu witnessed few case studies and from the autopsy of tigers Zoological Park showed existence of these paragonimosis in the study region [7]. No attempt has been made so for to study on the existence and incidence of *P. wertermani* of metacercariae

in fresh water crabs of the region. The aim of the study was to provide information about the metacercariae of *P. westermani* in the fresh water crabs in the study area, which was the first study in Kanchipuram District of Tamil Nadu, India.

The primary objective was to estimate the incidence of *Paragonimus* metacercariae among the second intermediate host, freshwater crabs, in the region, and secondary objective was to examine the distribution of *Paragonimus* metacercariae in various edible body parts of the freshwater crab.

MATERIALS AND METHODS

This was a pilot study which was carried out over a period of three months (October to December 2019) in wet season of Ammapettai Village, Tamil Nadu, India. A total number of 80 crabs were collected from fresh water ponds, rice fields and lakes in the study area. The study protocol was approved by the Institutional Animal Committee (IAEC/SSSMC&RI/2019-09) of Shri Sathya Sai Medical College & Research Institute.

Inclusion criteria: Live fresh water carbs were included in the study.

Exclusion criteria: Dead crabs or crabs showing signs of decomposition at the time of collection were excluded from the study.

Sample size: Sample size was calculated by using the formula: $n=Z^2 \times p(1-p) / d^2$

Z=Z-score (95% confidence), p=expected proportion (use 0.5 for conservative estimate) and d=desired precision (e.g., 0.1 for ±10%) based on the above sample collection formula, 80 fresh water crabs were collected [8].

Survey of Intermediate Host

Fresh water crabs: The infective stage of the parasite, the metacercariae occurs in the muscles of fresh water crabs or cray fishes which serves as the second intermediate host. The infection in the final host is formed by eating inadequately cooked infected crabs. The fresh water crabs were collected from the study area where eating crabs is a common food practice among the inhabitants in this region [Table/Fig-1].

[Table/Fig-1]: Fresh water crab (*Oziotelphusa ravi*).

A total number of 80 crabs were examined to detect infestation with metacercariae of *Paragoniums* over a period of three months during monsoon season.

Parasitological examination: All the collected freshwater crabs were dissected. Crabs were killed by stabbing them through the sternum, later legs and claws were removed and the shell of crab was lifted off in one piece and discarded. The remaining body was divided into two parts: the whole body (Carapace) as one part and claws (Chelipeds) and legs (Periopods) together as second part [Table/Fig-2,3].

[Table/Fig-2]: Whole body (Carapace) of fresh water crab.

[Table/Fig-3]: Claws (Chelipeds) and Legs (Periopods) of fresh water crab.

The two parts of the whole crab of each specimen was minced separately were grained with mortar and pestle. The grained material was filtered through mesh wire sieve and the filterable sediments were washed repeatedly with normal saline in order to get a clear supernatant. The sediment of each part was examined for metacercariae under a dissecting microscope and subsequently with light microscopy by unstained wet mount [9,10].

STATISTICAL ANALYSIS

All the parameters were analysed and tabulated, and statistical analysis was performed using Statistical Package for the Social Sciences (SPSS) version 22.0 and Microsoft Excel 2007. The Chi-square test was also applied wherever applicable to assess the significance of the findings. A p-value <0.05 was considered statistically significant.

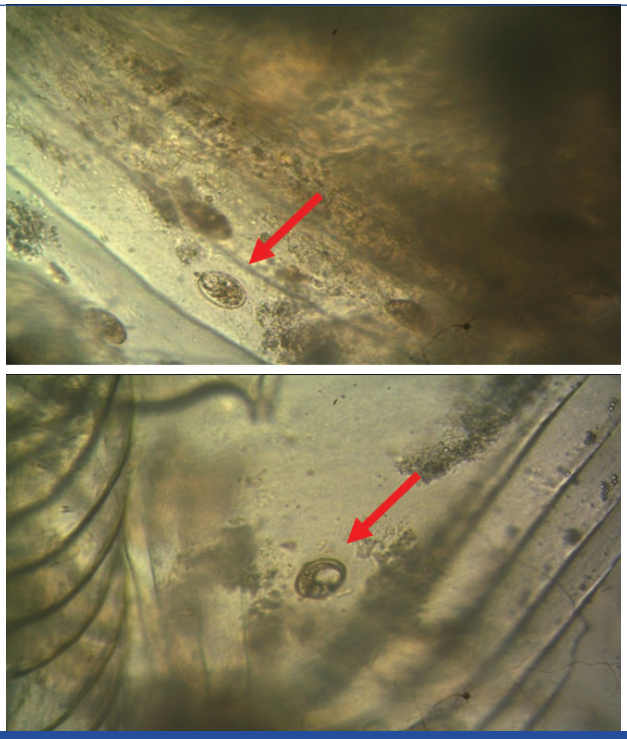
RESULTS

All the species *Oziotelphusa ravi* were collected from locality of Ammapettai Village, Kanchipuram District, Tamil Nadu, India from various sites like ponds, ditches and rice fields in the study area.

The microscopic examination revealed an overall incidence of *Paragonimus* metacercarial infestation in five of 80 (6.25%) crabs. Among other parasites, only monogeneans were detected in two crabs [Table/ Fig-4,5].

Monogeneans are parasitic flatworms belonging to the class Monogenea. They are primarily ectoparasites, commonly found on the gills, skin, or other external surfaces of aquatic hosts, such as fish and crabs. Microscopic characteristics of monogeneans have a flattened body, usually leaf-like or elongate, and exhibit bilateral symmetry. They are generally small, ranging from 0.5 mm to 2 cm in length, though some species may grow larger. The fresh water crab edible parts show the results of detailed examination of five metacercariae in fresh water crabs, giving the distribution of metacercariae in two different parts like whole body showing

three, claws and legs showing two metacercariae in fresh water crab edible parts [Table/Fig-6-8]. There was no significant difference between whole body and claws and legs [Table/Fig-9].



[Table/Fig-4]: The microscopic examination (under low power 10x) fresh water crabs showing *Paragonimus* metacercariae infective form.

Host	Infections	n (%)
Oziotelphusa ravi	*Paragonimus* metacercariae	05 (6.25)
	Other parasite (monogeneans)	02 (2.5)

[Table/Fig-5]: Incidence of Paragonimus metacercarial infection and other parasites in fresh water crab hosts in suspected foci (n=80).

[Table/Fig-6]: The macroscopic examination (under low power10x) fresh water crabs showing other parasite (monogeneans).

[Table/Fig-7]: Mounted slide (4x) monogeneans parasite.

[Table/Fig-8]: Parasite (monogeneans) mounted slide.

Whole body (Carapace)	Claws (Chelipeds) & Legs (Periopods)		χ^2 (p-value)
	Present	Absent	
Present	00	03	0.80 (0.777)
Absent	02	75	

[Table/Fig-9]: Distribution of *Paragonimus* metacercariae in fresh water crab edible parts (n=80).

DISCUSSION

In the present study, the incidence of *Paragonimus* metacercariae in freshwater crabs was found to be 6.25%, a relatively low rate compared to previous studies in India [11]. One such study reported a 59.4% prevalence of intestinal parasites in crabs collected from mountain stream areas. Freshwater crabs in India are often poorly studied due to their secretive and nocturnal behaviour, and even though recent research in biodiversity hotspots like the Western Ghats has revealed new genera and species, many remain undiscovered owing to insufficient habitat surveys [12]. *Oziotelphusa* species, typically found in rice fields, are widely distributed in the low-lying areas of Sri Lanka and India [13,14].

Taxonomy: Superfamily- Gecarcinucoidea Rathbun, 1904, Family: *Gecarcinucidae* Rathbun, 1904, Genus: *Oziotelphusa* Müller, 1887 and species: *Oziotelphusa ravi* sp. (new species). Banana plantation showing holes dug by *Oziotelphusa ravi*, new species; B, burrow of crab next to flooded rice field. The species *Oziotelphusa ravi*, identified in Tamil Nadu, was examined for *Paragonimus* metacercariae infection [15]. The presence of lung flukes was confirmed through dissection of infected crabs, specifically in the gills and thoracic muscles, revealing cystic larvae [16,17].

In the study, metacercariae were isolated from the crabs' muscles using an artificial gastric juice digestion technique. Among the three genera of crabs surveyed from Arunachal Pradesh, Assam, Manipur, and Mizoram, only *Barytelphusa* harboured metacercarial cysts. Crabs from Arunachal Pradesh were found to be positive for the infection, with a prevalence of 26% in *Barytelphusa lugubris* at the Kharshang site [18].

Previous research has identified regions such as Bengal and Assam as endemic areas for human paragonimiasis, with recent reports of the infection in Manipur. Although *Paragonimus* can parasitise a wide range of mammalian hosts, its prevalence and host range in India are not well documented. In recent years, the species *P. hueitungensis* and *P. heterotremus* have been identified as the causative agents of paragonimiasis in Manipur and Arunachal Pradesh, where consumption of crustacean intermediate hosts is common [19,20].

This study also found a low prevalence of other parasitic infections, such as monogeneans (2.5%), in freshwater crabs. Overall, the rate of *Paragonimus* metacercariae infestation was notably low compared to other studies in India.

Limitation(s)

It was conducted in only one small area, which limits the generalisability of the results to other regions. Moreover, the crabs were not selected randomly, which may have bias and affected the accuracy of the outcomes. In addition, the sample size was relatively small, making it difficult to capture the full range of variation in the crab population. Finally, the study was carried out over a short period of three months, which may have overlooked seasonal changes that could influence the results.

CONCLUSION(S)

The findings revealed the presence of other parasitic infections, such as monogeneans, in the region. The study highlights the underdiagnosed issue of endemic haemoptysis, which warrants further research and improved diagnostic practices for human pulmonary paragonimiasis. Additionally, there is a critical need to raise awareness among local populations who commonly consume crabs. Health education should emphasise the importance of consuming properly cooked crabs to mitigate the risk of parasitic infections.

Acknowledgement

Authors sincerely thank ICMR-STS for their support in making this project possible. Authors also extend their gratitude to Stephen Ambu, Associate Dean of Research at the International Medical University, Kuala Lumpur, Malaysia, for his invaluable assistance throughout the research.

REFERENCES

[1] Kalhan S, Sharma P, Sharma S, Kakria N, Dudani S, Gupta A. *Paragonimus westermani* infection in lung: A confounding diagnostic entity. Lung India. 2015;32:265-67.
[2] Furst, TU, Duthaler, B, Sripa, J, Utzinger, JK. Trematode infections: Liver and lung flukes. Infect Dis Clin North Am. 2012;26:399-419.
[3] Singh TS. Occurrence of the lung fluke Paragonimus heterotremusin Manipur, India. Chinese Med Sci J. 2003;18:20-25.
[4] Singh TS, Hiromu S, Devi KR, Singh WA. First case of *Paragonimus westermani* infection in a female patient in India. Indian J Med Microbiol. 2015;33:S156-S159.
[5] KanwarNarain K, Rekha Devi, Mahanta J. Paragonimus and paragonimiasis- A new focus in Arunachal Pradesh, India. Current Science. 2003;84:985-87.
[6] Singh TS, Sugiyama H, Umehara A, Hiese S, Khalo K. Paragonimus heterotremus infection in Nagaland: A new focous of paragonimiasis in India. Indian J Med Microbiol. 2009;27:123-27.
[7] Moudgil AD, Singla LD, Pallavi. Parasitosis in wild felids of India: An overview. J Threatened Taxa. 2015;10:7641-48.
[8] Lwanga SK, Lemeshow S. Sample size determination in health studies: A practical manual. World Health Organization, 1991.
[9] Sachs R, Cumberlidge N. Distribution of metacercariae in freshwater crabs in relation to Paragonimus infection of children in Liberia, West Africa. Annals of Tropical Medicine & Parasitology. 1990;84:277-80.
[10] Kearn G, Whittington I. Sperm transfer in monogenean (platyhelminth) parasites. Acta Parasitologica. 2015;60(4):567-600.
[11] Singh T, Zaman FA, Sugiyama H. Epidemiology and laboratory diagnosis of paragonimiasis. Int J Curr Med App Sci. 2017;16(1):17-27.
[12] Pati SK, Pradhan RN. An overview of the freshwater crabs of the Western Ghats, India. Oceanogr Fish Open Access J. 2020;12(3).
[13] Bahir MM, Yeo DCJ. A revision of the genus Oziotelphusa Müller, 1887 (Crustacea: Decapoda: Brachyura: Parathelphusidae), with description of eight new species. Raffles Bulletin of Zoology. 2005;12(Supplement):77-120.
[14] Pati SK, Sharma RM. Oziotelphusaganjamensis, a new species of freshwater crab (Brachyura: Gecarcinucidae) from south Odisha (Orissa) state, eastern India. Zootaxa. 2012;3528:49-56.
[15] Raj S, Bijukumar A, Peter k. A new species of freshwater crab of the genus Oziotelphusa Müller, 1887 (Crustacea: Decapoda: Brachyura: Gecarcinucidae) from Tamil Nadu, southern India. Zootaxa. 2017;4363(2):225-36.
[16] Kradin RL, Mark EJ. In diagnostic pathology of infectious disease Textbook. 2010.
[17] Muller R. Liver and lung flukes. In: Cox FEG. The Wellcome Trust illustrated history of tropical diseases. The Wellcome Trust, London, United Kingdom; 1996. p. 274-85.
[18] Razaque MA, Mutum SS, Singh TS. Recurrent haemoptysis? Think of Paragonimiasis. Tropical Doctor. 1991;21:153-55.
[19] Singh TS, Mutum S, Razaque MA, Singh YI, Singh EY. Paragonimiasis in Manipur. Indian J Med Res.1993;97:247-52.
[20] Singh TS, Singh YI. TThree types of Paragonimus metacercariae isolated from Potamiscus manipurensis in Manipur. Indian J Med Microbiol. 1997;15:159-62.

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PLAGIARISM CHECKING METHODS: [Jain H et al.]
- Plagiarism X-checker: May 16, 2025
- Manual Googling: Oct 02, 2025
- iThenticate Software: Oct 04, 2025 (2%)

ETYMOLOGY: Author Origin

EMENDATIONS: 7

AUTHOR DECLARATION:
- Financial or Other Competing Interests: None
- Was Ethics Committee Approval obtained for this study? Yes
- Was informed consent obtained from the subjects involved in the study? No
- For any images presented appropriate consent has been obtained from the subjects. NA

Date of Submission: **May 15, 2025**
Date of Peer Review: **Jun 28, 2025**
Date of Acceptance: **Oct 06, 2025**
Date of Publishing: **May 01, 2026**