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Larval *Gnathostoma*: morphological identification and probable source of a case of human eye infection in Assam, India

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Abstract
The present communication deals with morphological identification of a parasite specimen sent from a private eye hospital at Guwahati, Assam after its extraction alive from the aqueous humor in the right eye of a non-tribal rural woman from Assam. The parasite 3 mm X 0.3 mm in size, characterized by the presence of a distinct anterior head bulb armed with four rows of hooklets numbering less than 40 per row and spination along the transverse striations present throughout the body was identified as the third stage larva of a pig gnathostome, *Gnathostoma doloresi*. Interview based investigation to ascertain the source of *G. doloresi* infection reported here for the first time in the 32 year old non-vegetarian house wife having no history of raw fish consumption revealed water used for domestic purpose from a pond adjacent to domestic pig pastured swampy area and the fresh water fishes as the possible sources of infection which might have occurred by accidental ingestion of infected cyclops present in pond water or by direct skin penetration of the larval stage of the parasite during descaling and evisceration of infected fishes prepared for cooking and consumption.

Keywords: *Gnathostoma hispidum*, *Gnathostoma doloresi*, man, eye, Assam, zoonoses

Introduction
Gnathostomiasis is a food borne parasitic zoonosis caused by *Gnathostoma spinigerum*, *Gnathostoma hispidum*, *Gnathostoma doloresi* and *Gnathostoma nipponicum* in Asian countries. The adult parasites living in the stomach of cat, dog, pig and other similar wild animal species require cyclopid copepods (Genera-*Cyclops*, *Eucyclops*, *Mesocyclops*), the aquatic crustaceans as the first intermediate host and the fresh water fishes including eels as the second intermediate hosts for development of early third stage larva (EL3) and advanced third stage larva (AL3) respectively (Liu *et al*., 2020) [1]. Additionally, frogs, snakes and birds serve as paratenic hosts which along with the final host acquire infection through consumption of infected intermediates hosts or paratenic hosts.

Humans are accidental hosts and usually acquire infection through consumption of AL3 contaminated fresh water fish and meat of the paratenic hosts. Drinking of untreated surface ground water contaminated with cyclops carrying EL3 and direct entry of AL3 by skin penetration during handling of contaminated fish or meat have also been suggested as additional risk factors of human infection (Herman and Chiodini, 2009) [2]. The parasitic stage erratically migrates into the sub cutaneous tissues and different visceral organs including eye, brain and spinal cord resulting into manifestation of migratory swelling under the skin and other organ specific complications respectively (Barua *et al.*,2007; Herman and Chiodini 2009; Shubhedar *et al*., 2014; Nawa *et al.*, 2015) [3, 2, 4, 5].

Many human cases of gnathostomiasis caused by *G. spinigerum* and also by *G. hispidum* and *G. doloresi* have been reported from Japan, China, highly endemic Thailand and other Southeast Asian countries where migratory swelling under the skin was a common occurrence (Bravo and Gontijo, 2018) [6]. In India also, sporadic cases of gnathostomiasis in both animals and man have been reported. *G. spinigerum* was reported from cats, dogs and wild felines from different regions including the Northeast (Baruah and Gogoi, 1989; Deka *et al*., 1995; Borthakur and Mukherjee, 2011; Islam *et al*., 2014; Lenka *et al*., 2016) [7, 8, 9, 10, 11]. Additionally, *G. hispidum* and *G. doloresi* were also reported in domestic pigs from the Northeast and Eastern region

(Maplestone, 1930; Yadav and Tandon, 1989; 1994; NEHU 2021) [12. 13. 14. 15]. Twenty-two corresponding human cases of gnathostomiasis recorded so far from India (Pillai *et al.*, 2012; Rawat *et al.*, 2016; Mohanty and Mahapatra, 2020) [16, 17, 18] since 1945 were due to *G. spinigerum* larva involving the eye in 20 cases and the rest with cutaneous involvement. In the Northeast region of India, out of 6 human cases under report, four cases of gnathostomiasis due to *G. spinigerum* have been reported from Assam (Barua *et al.*, 2007; Bhattacharjee *et al.*, 2007; Islam *et al.*, 2011; Das *et al.*, 2014) [3, 19, 20, 21] and one each from the states of Meghalaya (Barua *et al.*, 2007) [3] and Manipur (Mukherjee *et al.*, 2012) [22].

The present communication reports identification of *G. doloresi* from a case of ocular gnathostomiasis in a rural non-tribal woman from Assam and evaluation of the possible risk factors which led to the infection in the patient.

Materials and Methods

A parasite specimen extracted from the aqueous humor in the right eye of a 32 -year -old non-tribal woman from the Kamrup (Rural) district of Assam was forwarded from a private eye hospital at Guwahati, Assam to the Department of Parasitology, College of Veterinary Science, Guwahati for its identification. Preliminary examination of the specimen sent in normal saline solution was done under light microscope to confirm its identity as parasite. The bearer of the specimen cum family member was interviewed to gather some information regarding the patient, travel history, food habits of the family, source of water supply, presence of water body and domestic animals in the surroundings. Morphological details of the specimen cleared in lactophenol were examined under light microscope.

Identification of the specimen

Identification of the received specimen up to species level was done on the basis of observed morphological features in consultation with taxonomic keys and descriptions provided in the literature (Yamaguti, 1961; Akahane *et al.*, 1982; 1994; Nawa *et al.*, 1989; Cho *et al.*, 2007; Jung *et al.*, 2008) [23, 24, 25, 26, 27, 28]. Factual informations thus obtained through interview together with the parasite's identity were analyzed for drawing conclusion on the source of infection.

Results

The parasite specimen cylindrical in shape and milky white in colour measured 3.0 mm in length and 0.3 mm in width (Fig 1). There was presence of a circular head bulb at the anterior end and it was armed with 4 rows of cuticular hooklets (Fig 2). The numbers of hooklets were 32, 36, 34 and 32 from the first row to the fourth row respectively. The hooklets of the first row were smaller than those present in other rows. Each hooklet consisted of irregular four-sided base and a posteriorly directed curved spine at the apex. A pair of lips was seen protruding from the top of the head bulb. The rest part of the body with a constriction behind the head bulb had single pointed cuticular spines along the transverse striations. The spines were densely distributed in the anterior two thirds of the body and gradually decreased both in the size and number towards the tail end (Fig 3). Other morphological features present in the larva could not be studied well since the parasite dispatched in normal saline and received for identification 7 days after recovery from the eye was found to be degenerated. The morphological features on the head bulb and body cuticle were found to be consistent with those of *G. doloresi* (Nawa *et al.*, 1989, Akahane *et al.*, 1998, Imai and Hasegawa, 2001) [26, 29, 30] and the parasite was tentatively identified as the third stage larva of *G. doloresi*.

Fig 1: *Gnathostoma* larva (partially damaged) with host tissue around the head bulb

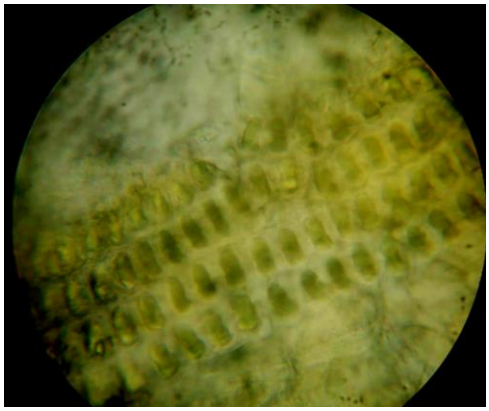

Fig 2: Head bulb (part) of the *Gnathostoma* larva with four rows of hooklets

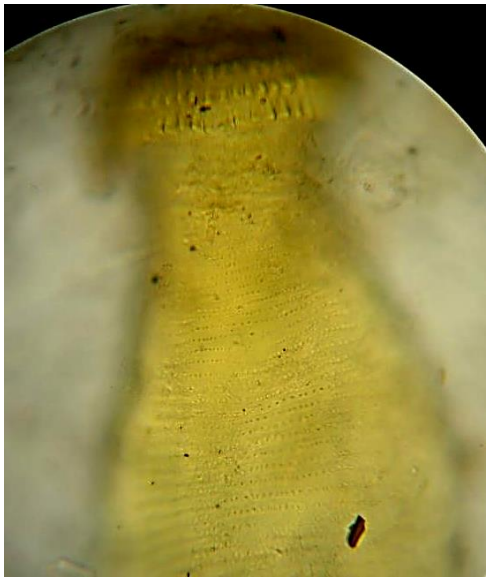

Fig 3: Anterior end of the gnathostome larva showing body spination

Discussion

G. hispidum and *G. doloresi* are the parasites of pigs and their larvae are known to infect humans to cause gnathostomiasis (Akahane *et al.*, 1998; Tuyen *et al.*, 2019) [29, 31]. The larva of *G. doloresi* of the present investigation differed from *G. spinigerum* (Akahane *et al.*, 1994; Baruah *et al.*, 2011) [25, 32] and *G. hispidum* (Akahane *et al.*, 1982; Cho *et al.*, 2007) [24, 27] by the presence of lowest number of hooklets on the head

bulb and these stood at less than 40 in each row as described earlier (Mako and Akahane, 1985; Nawa *et al.*, 1989; Akahane *et al.*, 1998; Imai and Hasegawa, 2001) [33, 26, 29, 30]. Detection of *G. doloresi* larva in Assam for the first time is in accordance with Maplestone (1930) [12] and Yadav and Tandon (1994) [14] who reported occurrence of adults of this species in pigs from the neighbouring states of West Bengal and Meghalaya respectively. Earlier to the report of *G. doloresi* from pig in the year 1930 from West Bengal, Maplestone (1929) [34] also reported a case of human ocular gnathostomiasis in Jalpaiguri district of West Bengal. However, the parasite could be assigned neither to *G. spinigerum* nor to *G. hispidum*. Besides detection of adult *G. spinigerum* in dogs, cats and its larval stage infecting man in Assam, the infection was also recorded in freshwater fishes like *Opiocephalus* and *Channa* in Assam and Bengal respectively (Akahane *et al.*, 1994; Baruah *et al.*, 2011) [25, 32]. These findings strongly suggest that the parasite spp are being naturally circulated among animals and fresh water fishes through cyclops and thereby causing local accidental infection to humans as an autochthonous gnathostomiasis (Vargas *et al.*, 2012; Kim *et al.*, 2013) [35, 36].

Personal interview revealed that the patient was a non-tribal 32 -year- old non-vegetarian rural housewife who had no history of eating raw fish or meat and travel outside the state. The food habit of the family included among others the regular consumption of fresh water fishes by cooking into different menus after proper descaling, evisceration and cleaning. Additionally, side dishes cooked from sun- dried fish were also included less often in their diet. The family had been regularly using pond/tank water for dip bath, washing of clothes, utensils, food materials and for other domestic purpose including livestock. The private pond as reported was adjacent to agricultural field and swampy area where domestic pigs and other animals used to roam freely for pasturing. This situation reflects to the contaminated pond water as the likely source of infection to the patient through accidental ingestion of cyclops carrying the larval stage of the parasite similar to the earlier report of Subhedar *et al*. (2014) [4] who found a vegetarian lady developing creeping eruption in different parts of the body by *Gnathostoma* sp. Moreover, the AL3 might have got entry by skin penetration during cleaning of fishes prior to cooking (Chen and Lin, 1991) [37].

Fish is a major and cheap source of protein for the rural people of Assam and neighbouring states. Owing to high rainfall and seasonal flood, the state has abundant water resource in the form of wetlands and other water bodies (Ramchiary *et al.*, 2014) [38] in which there is ample natural growth of various freshwater fishes. Diversified aquatic flora and fauna present in the water bodies provide a suitable condition to several helminth parasites for completing their life cycle. Besides consumption of fresh fishes by cooking as fry, curry and roasted fish mash, people in rural areas traditionally preserve surplus fishes in the form of sun-dried, kitchen smoke dried and fermented products for future use by cooking as fish pickle, curry or soup (Thapa *et al.*, 2004; Thapa, 2016; Payra *et al.*, 2016) [39, 40, 41].

It has been reported that more than 5000 cases of human gnathostomiasis have been reported worldwide mainly from the endemic countries where 90% of cases were attributed to eating of raw or inadequately cooked fishes (Herman and Chiodini, 2009; Liu *et al.*, 2020) [2, 1]. However, people of the Northeast and also other parts of India may not be accustomed to eating of raw fishes and the traditional culture of cooking might have greatly reduced the infection rate in them compared to maximum cases reported from Japan, China, Thailand and other Southeast Asian countries. Deviation from the custom of cooking or improper cooking may not be totally ruled out since isolated case may occur similar to the report of a case of paragonimiasis in a school going tribal boy from a tea garden of Assam due to frequent eating of raw crabs prepared at his home (Roy *et al.*, 2016) [42].

The patient infected with *G. doloresi* in the present study belonged to a non-tribal family and food habits as disclosed was different from those of tribal people of Assam and other North eastern states who are accustomed to prepare varieties of ethnic dishes from the preserved fish products (Thapa *et al.*, 2004; Thapa, 2016) [39, 40]. In view of high incidence of human and animal gnathostomiasis in the Northeast and Eastern region of the country, a systematic study may reduce the existing gap in understanding the epidemiology of infection which has the potential to emerge in near future as an important zoonosis in the country (Jyothimol and Ravindran, 2015) [43].

Conclusion

The present investigation reports for the first time a case of human gnathostomiasis caused by third stage larva of *G. doloresi* in the right eye of a middle aged non-tribal rural woman from Assam. In absence of habitual consumption of raw fish, it was evidently clear to point out the infected water cyclops or the fresh water fishes as the possible sources of infection to the patient by the larval stage of the parasite that gained entry through mouth or by skin penetration during handling of fishes for descaling and evisceration prior to cooking.

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References

1. Liu G-H, Sun M-M, Elsheikha HM, Sugiyama H, Ando K *et al*. Human gnathostomiasis: a neglected food-borne zoonosis. Parasites & Vectors. 2020;13(1):616.
2. Herman JS, Chiodini PL. Gnathostomiasis, another emerging imported disease. Clin Microbiol Rev. 2009;22(3):484-492.
3. Barua P, Hazarika NK, Barua N, Barua CK, Choudhury B. Gnathostomiasis of the anterior chamber. Indian J Med Microbiol. 2007;25(3):276-278. doi10.4103/0255-0857.34775
4. Subhedar V, Nandedkar S, Jain M, Bajpai T. Gnathostomiasis: A rare case of cutaneous creeping eruptions. Indian J Dermatopathol Diagn Dermatol. 2014;1(1):32-34.
5. Nawa Y, Maleewong W, Intapan PM, Diaz Camacho SP. *Gnathostoma*, Chapter 21, Xiao L, Ryan U, Feng Y (Eds). Food microbiology Series: Biology of Food-Borne Diseases. Boca Raton, London, New York: CRC Press, Taylor & Francis; c2015, p. 405-426.
6. Bravo F, Gontijo B. Gnathostomiasis: An emerging infectious disease relevant to all dermatologists. An Bras Dermatol. 2018;93(2):172-178.
7. Baruah N, Gogoi AR. Incidence of different helminthes in dogs in Guwahati (Assam). J Vet Parasitol.

1989;3(2):155-156.
8. Deka DK, Choudhury S, Chakraborty A. Parasites of domestic animals and birds of Lakhimpur (Assam). J Vet Parasitol. 1995;9(1):21-26.
9. Borthakur SK, Mukherjee SN. Gastrointestinal helminthes of stray cats (*Felis catus*) from Aizawl, Mizoram, India. Southeast Asian J Trop Med Public Hlth. 2011;42(2):255-258.
10. Islam S, Borthakur SK, Ali Ahmed SS. *Gnathostoma* and Gnathostomiasis in Northeast India: IV. Prevalence of *G. spinigerum* in dog. Vet Practitioner. 2014;15(1):20-22.
11. Lenka DR, Johns J, Gopi J, Chandy G, Narayanan PM, Kalarikkal DC, Ravindran R. Occurrence of *Gnathostoma spinigerum* in a leopard cat from Wayanad Wildlife Sanctuary, Kerala. J Parasit Dis. 2016;40(2):555-557.
12. Maplestone PA. Nematode parasites of pigs in Bengal. Rec Ind Mus. 1930;32(part 2):77-105.
13. Yadav AK, Tandon V. Nematode parasite infections of domestic pigs in a sub-tropical and high-rainfall area of India. Vet Parasitol. 1989;31(2):133-139.
14. Yadav AK, Tandon V. Scanning Electron Microscopy of Head and Cuticular Armature of *Gnathostoma doloresi* (Nematoda, Gnathostomatidae). Acta Parasitol. 1994;39:150-152.
15. NEHU. Northeast India Helminth parasite information database. Indian bio-resource Information Network, North Eastern Hill University, Shillong; c2021.
16. Pillai GS, Kumar A, Radhakrishnan N, Maniyelli J, Shafi T, Dinesh KR, *et al*. Intraocular Gnathostomiasis: Report of a Case and Review of Literature. Am J Trop Med Hyg. 2012;86(4):620-623.
17. Rawat P, Gautam M, Jain NC, Jain R. Intraocular gnathostomiasis: A rare case report from Central India. Indian J Ophthalmol. 2016;64(3):235-237.
18. Mohanty A, Mahapatra SK. Intraocular gnathostomiasis: A rare case report of a live Intravitreal worm. Delhi J Ophthalmol. 2020 Jul;31(1). doi: http://dx.doi.org/10.7869/djo.572
19. Bhattacharjee H, Das D, Medhi J. Intravitreal gnathostomiasis and review of literature. Retina. 2007;27(1):67-73.
20. Islam S, Bhattacharjee H, Das D, Medhi J. *Gnathostoma* and gnathostomiasis in North East India: II On the occurrence of an advanced third stage larva of *Gnathostoma spinigerum* Owen, 1836 in human eye with a description of its light microscopic morphology. J Vet Parasitol. 2011;25(1):18-20.
21. Das D, Islam S, Bhattacharjee H, Deka A, Yambem D, *et al.* Parasitic diseases of zoonotic importance in humans of northeast India, with special reference to ocular involvement. Eye and Brain. 2014;6(1):1-8.
22. Mukherjee A, Ahmed NH, Samantaray JC, Mirdha BR. A rare case of cutaneous larva migrans due to Gnathostoma sp. Indian J Med Microbiol. 2012;30(3):356-358.
23. Yamaguti S. Systema Helminthum. New York, London: Interscience Publishers; c1961, p. 3.
24. Akahane H, Iwata K, Miyazaki I. Studies on *Gnathostoma hispidum* Fedchenko, 1872 parasitic in loaches imported from China. JPN J Parasitol. 1982;31(6):507-516.
25. Akahane H, Sano M, Uchikawa R, Masataka K, Mahapatra SD, Dasgupta B, *et al*. Prevalence of larval *Gnathostoma* in Snake-Head fish from Northeast India with reference to their morphological findings. Jpn J Parasitol. 1994;43(2):105-109.
26. Nawa Y, Imai J, Ogata K, Otsuka K. The first record of a confirmed human case of *Gnathostoma doloresi* infection. J Parasitol. 1989;75:166-169.
27. Cho SH, Kim TS, Kong Y, Na BK, Sohn WM. Larval *Gnathostoma hispidum* detected in the red banded odd-tooth snake, *Dinodon rufozonatum rufozonatum* from China. Korean J Parasitol. 2007;45(3):191-198.
28. Jung BK, Lee JJ, Pyo KH, Kim HJ, Jeong HG, *et al.* Detection of *Gnathostoma spinigerum* third stage larvae in snakeheads purchased from a Central part of Myanmar. Korean J Parasitol. 2008;46(4):285-288.
29. Akahane H, Shibue K, Shimizu A, Toshitani S. Human gnathostomiasis caused by *Gnathostoma doloresi* with particular reference to the parasitological investigation of the causative agent. Ann Trop Med Parasitol. 1998;92(6):721-726.
30. Imai J, Hasegawa H. Molting of *Gnathostoma doloresi* (Nematoda: Gnathostomatoidea) in the definite host. J Parasitol. 2001;87(1):14-18.
31. Tuyen NV, Lan NTK, Doanh PN. Morphological and molecular characteristics of adult worms of *Gnathostoma*, Owen 1836 (Nematoda) collected from domestic pigs in Dien Bien Province, Northern Vietnam. Folia Parasitol. 2019;66:010. Doi: 10.14411/fp.2019.010.
32. Baruah N, Islam S, Gogoi AR. Gnathostomiasis in Assam: I. On the prevalence of *Gnathostoma spinigerum* larvae in fishes, experimental infection in first and second intermediate host with a description of an advanced third stage larva recovered from *Opiocephalus punctatus*. J Vet Parasitol. 2011;25(1):11-17.
33. Mako T, Akahane H. On the larval *Gnathostoma doloresi* found in a snake, *Dinodon semicarinatus* from Amami-Oshima Is., Japan. Jpn J Parasitol. 1985;34(6):493-499.
34. Maplestone PA. A case of human infection with a gnathostome in India. India Med Gaz. 1929;64(11):610-614.
35. Vargas TJ de Sousa, Kahler S, Dib C, Cavaliere MB, Jeunon-Sousa MA. Autochthonous gnathostomiasis, Brazil. Emerg Infect Dis. 2012;18(12):2087-2089. doi: 10.3201/eid1812.120367
36. Kim JH, Lim H, Hwang YS, Kim TY, Han EM, *et al.* *Gnathostoma spinigerum* infection in the upper lip of a Korean woman: An Autochthonous case in Korea. Korean J Parasitol. 2013;51(3):343-347.
37. Chen QQ, Lin XM. A survey of epidemiology of *Gnathostoma hispidum* and experimental studies of its larvae in animals. Southeast Asian J Trop Med Public Hlth. 1991 Dec 1;22(4):611-617.
38. Ramchiary H, Kapinder, Goswami UC, Singh HS. Abundance of Diseases in Food Fishes of North East Region of India with reference to economic loss. IOSR J Agric Vet Sci. 2014;7(1):23-33.
39. Thapa N, Pal J, Tamang JP. Microbiol diversity of ngari, hentak and tungtap, fermented fish products of North-East India. World J Microbiol Biotechnol. 2004 Aug;20(6):599-607.
40. Thapa N. Ethnic Fermented and Preserved Fish Products of India and Nepal. J Ethnic Foods. 2016 Mar 1;3(1):69-77. doi: 10.1016/j.jef.2016.02.003
41. Payra P, Maity R, Maity S, Mandal B. Production and marketing of dry fish through the traditional practices in

West Bengal Coast: Problems and Prospects. Int J Fisheries & Aquatic Studies. 2016;4(6):118-123.
42. Roy JS, Das PP, Borah AK, Das JK. Paragonimiasis in a child from Assam, India. J Clin Diagn Res. 2016;10(4):DD06-DD07 doi: 10.7860/JCDR/2016/18160.7616
43. Jyothimol G, Ravindran R. Emerging and Re-emerging Parasitic zoonoses in India. Adv Anim Vet Sci. 2015;3(12):617-628.