

Original Article

HOST RANGE STUDIES OF *APHANOMYCES LAEVIS* IN SOME FRESH WATER TROPICAL FISHES, ORIGINALLY ISOLATED FROM *GARRA GOTYLA* (G.), A NEW HOST RECORD.

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ABSTRACT

**Objective:** To find out the host range of fungi isolated from fish *Garra gotyla* (Gray)

**Methods:** During the present study 28 specimens of fresh water tropical fish *Garra gotyla* (Gray.) were collected from Larpur reservoir, Bhopal for fungal isolation. On the basis of taxonomic characters, the fungi isolated was identified and experimental infection studies were conducted on twelve different species of fishes, viz. *Carrasius auratus*, *Chanda ranga*, *Cirrhinus mrigala*, *Ctenopharengedon idella*, *Labeo rohita*, *L.bata*, *Mastacembalus armatus*, *Mystus cavasius*, *Nandus nandus*, *Puntius sarana* *P.ticto* and *Garra gotyla* itself by inoculating the isolated fungi

**Results:** From *Garra gotyla*, *Aphanomyces laevis*, (de Bary).have been reported first time, it's a new host record for the given isolated species of fungi and it was found pathogenic to all the fishes causing infection and mortality within six days of experiment. Maximum infection and early mortality was observed in *Puntius spp.* (*P.sarana* and *P.ticto*). In most of the experimental fishes infection was developed in form of mycelial growth and in some fishes de scaling and red spots were observed. Only in *Mystus seenghala* a lesion was developed with hyphal growth.Mortality of fishes was 100%.

**Conclusion:** *Gara gotyla* is a new host record for *Alaevis*. Experimental studies showed, given fungi is having wider host range with high parasitic ability

**Keyword:** *Garra gotyla* *Aphanomyces laevis*, Parasitic fungi.

INTRODUCTION

Several zoosporic fungi cause infection in fish. *Aphanomyces laevis* is one of the parasitic fungi responsible for causing infection in fresh water fishes. *A.laevis* was first reported as parasite in *Xiphophorous maculatus* by [1]. Later on some other workers reported the isolation of this fungi from infected fish and prawn, [2], [3], [4], [5],[6],[7], [8], and [9]. Reports on pathogenicity and host range of *A.laevis* are very rare, [10] reported pathogenicity on eggs of *Channa striatus* and [11] reported its pathogenicity on *Nile tilapia* Present study describes the isolation of *A.laevis* from fresh water tropical fish *Garra gotyla* (Gray.) which is a new host record of this species of fungi and also describes the experimental exposure of the isolated strain of fungi on some fresh water fishes to find its host range by which the virulence of given species of fungi can be found out.

MATERIALS AND METHODS

For the present study a total number of 28 specimens of *Garra gotyla* (Gray) were collected from Larpur reservoir in sterilized polythene bags and brought to the laboratory. They were kept in aquaria for further observations. Most of the collected fishes showed mycelial growth over the body and in few specimens lesions were found. Cultures were prepared by taking small inocula from fish. The methods of [12] and [13] were used for preparation of cultures and to purify them. Sabourauds Dextrose Agar (SDA) was used to prepare the cultures and Sorghum seeds (Jowar) were used as bait to prepare wet cultures for identification of species. To avoid bacterial contamination all the cultures were prepared in aseptic condition and by using streptomycin sulphate (100mg/ml) in media. Cultures were grown on 18-22 °C. Fully grown cultures were identified with the help of monographs of [14] and [15]. Fishes was identified by [16]. To determine the host range of isolated species of fungi zoospores concentration were prepared by using haemocytometer. Healthy fishes of twelve different species viz. *Carrasius auratus*, *Chanda ranga*, *Cirrhinus mrigala*, *Ctenopharengedon idella*, *Labeo rohita*, *L.bata*, *Mastacembalus*

*armatus*, *Mystus cavasius*, *Nandus nandus*, *Puntius sarana*, *P.ticto* and *Garra gotyla* were taken and kept in aquaria of 10 L. capacity species wise separately. They were kept under observation for three days with continuous aeration and fed with artificial feed. For experimental purpose fishes were injected subcutaneously with 0.1 ml concentration of zoospores. Concentrations of zoospores were used according to the weight of fish used. Fishes were observed for six days from the day of inoculation and morbid fish were immediately removed from experimental troughs and aquariums. Experimental fishes were observed for visible symptoms like de coloration of skin, de scaling, mycelial growth and ulcerations.

RESULTS

In the present study *A.laevis*, (de Bary) was isolated from naturally infected fresh water fish *Garra gotyla*, which is a new host record for the given species of fungi. (Fig-1.). *A.laevis* was identified on the basis of its taxonomic characters like Hyphae, slender, branched 4-8µm thick, zoosporangia filamentous, 350-800 µm long and 4-8.5 µm in diameter. Zoospores arranged in a single row, rod shaped; encysted zoospores 5.5-8.5 µm, oogonia few, terminal 15-25 µm in diameter. Antheridia numerous, declinous rarely androgynous. Oospores spherical single, 12-20 µm in diameter.

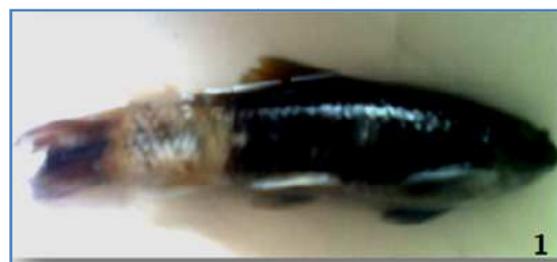


Fig. 1: *Garra gotyla* found infected with *Aphanomyces laevis*.

Artificial infection studies of isolated strain of fungi on eleven different species of fresh water tropical fishes showed it is a parasitic fungi lethal to all the fishes tested. All the inoculated specimens lead to infection and mortality within six days of

experiment. Maximum visible infection and mortality was observed on the fourth day. Among the fishes tested most virulent affect of this fungus was observed on *Puntius spp.* (*P.sarana* and *P.ticto*) in which all the ten inoculated fishes died within four days. (Table-1).

**Table 1: Pathogenicity of *Aphanomyces laevis* on ten different species of fresh water fishes, showing the host range**

S. No.	Fungi inoculated	Fish used	Mean L(cm)	Mean Wt(gm)	No. of fish used	Conc. Of zoospores (ml)	Mycoses evident within six days.					% of survival
							2	3	4	5	6	
1.	<i>Aph.laevis</i>	A	9.51±2.00	12.38±1.12	06	125	2	1	3	1	3	0
2.	<i>Aph.laevis</i>	B	9.46±2.24	10.14±2.01	10	120	2	2	5	1	-	0
3.	<i>Aph.laevis</i>	C	12.21±1.26	10.12±2.14	06	125	2	1	1	2	-	0
4.	<i>Aph.laevis</i>	D	10.26±2.24	11.24±1.12	06	125	1	2	1	1	1	0
5.	<i>Aph.laevis</i>	E	10.21±1.46	9.24±3.12	06	125	-	-	3	1	2	0
6.	<i>Aph.laevis</i>	F	10.36±2.12	10.14±1.21	06	120	-	2	2	-	2	0
7.	<i>Aph.laevis</i>	G	11.34±2.12	10.21±2.16	06	125	-	2	1	2	1	0
8.	<i>Aph.laevis</i>	H	10.13±1.16	9.26±1.24	06	120	2	-	2	1	1	0
9.	<i>Aph.laevis</i>	I	10.24±1.12	9.65±2.12	06	120	1	2	1	2	-	0
10.	<i>Aph.laevis</i>	J	9.84±2.34	10.16±1.4	10	120	5	3	2	-	-	0
11.	<i>Aph.laevis</i>	K	8.62 ±2.12	8.42± 3.62	10	125	4	2	4	-	-	0
12.	<i>Aph.laevis</i>	L	11.21±2.26	10.46±3.12	06	125	1	1	3	1	-	0

\*Aph.-*Aphanomyces*(A- *Carrasius auratus*, B- *Chanda ranga*, C- *Cirrhinus mrigala*, D-*Ctenopharengoedon idella*, E- *Labeo rohita*, F- *L.bata*, G- *Mastacembalus armatus*, H- *Mystus seenghala*, I- *Nandus nandus*, J- *Puntius ticto*, K- *P.sarana* & L-*Garra gotyla*.)

Percentage of survival was zero. All the experimental fishes showed external visible symptoms similar to naturally infected fishes like white cottony outgrowths, descaling, decoloration of skin and ulcerations. (Fig-2A-2k)



**Description of infected fishes artificially infected with *Aphanomyces laevis*.** Fig2A- 2K De scaling in *C. auratus*, loss of epidermis in *C.ranga*, caudal region infection in *C.mrigala*, cottony out growth on anterior region of *C.idella* & *L.rohita*, de scaling & decoloration in *L.bata*, mycelia growth on posterior region of *M armatus*, a large ulcer with hyphal growth in *M.seenghala*, *N.nandus* with hyphae covering the body, patch. *P sarana* with cottony out growths & infection on anterior portion and *P.ticto* with cottony patch

## DISCUSSION

During the present investigation fungi, *Aphanomyces laevis* (de Bary) have been reported from *Garra gotyla* as a new host record. Some workers reported it from fresh water fishes, *Labeo rohita* and *Cirrhinus mrigala* [6], *Channa striatus* [17]. [18] Isolated this species of fungi from *Mastacembalus armatus*, *Mystus viatus*, *Nandus nandus*, *Tor putitora* and *Tor tor*. [19] reported this fungi from *Aptocheilus panchax* and [20] reported the isolation of *A.laevis* from *Mystus seenghala*. All these reports of isolation of *A.laevis* were from naturally infected fishes in which the fungi may be primary or secondary pathogen.

Reports on artificial infection studies with *A.laevis* in fishes rare and not well documented. [4] tested this species of fungi on eggs of *Channa striatus* with 100% mortality. [11] tested it on *Nile Tilapia*. Present findings are in agreement with these reports giving wider host range of *A.laevis* in eleven different species of fresh water fishes with 100% mortality showing its virulent parasitic ability.

## CONCLUSION

It was found from the above study that *Garra gotyla*, a fresh water tropical fish is a new host record for *A.laevis* and pathogenicity tests on twelve different species of fishes showed the isolated strain of fungi is having wider host range and virulence in nature causing infection in fishes with 100% mortality.

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## REFERENCES

1. Vishniac HS & Nigrelli RF. The ability of Saprolegniaceae to parasitize platy fish. *zoologica*, 1957; 42; 131-134.
2. Scott WW & O'Bier AH, 1962, Aquatic fungi associated with diseased fish and fish eggs. *Prog. Fish Cult.* 1962; 24: 3-15.
3. Scott WW. Fungi associated with fish diseases. *Dev. Ind. Microbiol*; 5:109123.
4. Shrivastava GC & Shrivastava RC. Two fungal parasites of eggs of *Channa striatus*
5. *Bl.Curr.Sci*, 1976; 44:817-818.

6. Shrivastava GC & Shrivastava RC. Ability of saprolegneous fungi to Parasitize *Colisa fasciatus* Bl.Geobios, 1977a; 4 :31-32.
7. Shrivastava RC. Fungal parasites of certain fresh water fish of India, Aquaculture, 1980; 21:387-392
8. Prabhujji SK, Shrivastava GC & Shina SK. Some saprolegneous fungi in *Macro brachium lamarrei* H.M.Edw. and their eggs. Curr.Sci, 1984; 53(20):1104-1106.
9. Khulbe RD. Studies on aquatic phycomycetes of Nanital in relation to taxonomy, physiology, ecology and pathology, 1985 ; D.Sc thesis Kumaun University, Nanital pp335.
10. Khulbe RD. A manual of aquatic fungi (Chytridiomycetes & Oomycetes). Daya Publishing Housing House, 2001; Delhi: 255.
11. Shrivastava RC. *Aphanomyces*. a new threat to fish population, Mycosen, 1979a; 22:25-30
12. Hashem, E.H. Ali and M.B Salahy-A. Pathogenicity and oxidative stress in *Nile tilapia* caused by *Aphanomyces laevis* and *Phoma herbarum* isolated from farmed fish. Dis. Aquat. Organ Mar. 2011; 16:94 (1) 17-28.
13. Fraser GC, Calinan RB, Calder LM. *Aphanomyces species* associated with red spot disease an ulcerative disease of estuarine fish from eastern Australia. J.of fish diseases, 1992; 15; 173-181.
14. Khulbe RD. The Parasitic water molds (Taxonomy, Ecology, Pathology and Control). Publ by Almora Book Depot, Almora, India, 1993; pp 1-143.
15. Scott WW. A monograph of genus *Aphanomyces*. Verginia Agric..Exp.Sta, Tech.Bull. 1961; 151:95pp.
16. Willoughby LG. Fungi and Fish Diseases. Pisces Press, Sterling, UK. 1994; p. 57.
17. Jhingran VG. Fish and fisheries of India. Revised and enlarged second edition. 1999; Hindustan publishing corporation, Delhi (india), 666pp.
18. Chinabut S, Roberts RJ, Willoughby GR & Pearson MD. Histopathology of snake head *Channa striatus*, (Bloch). Experimentally infected with the *Aphanomyces* fungus associated with Epizootic ulcerative syndrome (EUS) at definite temperature. J. of fish diseases 1995; 18:41-47.
19. Khulbe RD, Joshi C & Bisht GS. Fungal diseases of fish in Nanak Sagar, Nanital, India. Mycopathologia, 1995; 130 (2): 71-74.
20. Mondal SK & De AB. A fatal Oomycotic disease of fresh water fish *Aplochelius panchax* in India caused by *Aphanomyces laevis*. Mycopathologia, 2001; 154: 21-24.
21. Chauhan R. Study on certain fungal diseases in culturable and non-culturable Species of fishes of Upper lake, Bhopal. J. Chem. Bio. Phy. Sci. 2012; B, Vol- 2, No.1810-1815.