

Fungal attack on *Tilapia mossambicus* in culture pond, leading to mass mortality of fishes.

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Abstract:-

This study was conducted on mass mortality of *Tilapia mossambicus* in culture pond of University campus, Bhopal. In December, 2012 fungal infection was observed on body of fishes in form of cottony mycelium. Anterior region of body was most affected area and fishes suffered from severe infection followed by death. Fishes were examined regularly for the period of one month. Isolation of fungi revealed the presence of six species of fungi viz. *Achlya Americana*, *Achlya proliferoids*, *Aphanomyces laevis*, *Pythiopsis* species, *Saprolegnia diclina* and *Saprolegnia parasitica*. Total 196 isolates were cultured by using three different agar media viz. Corn Meal Agar and Potato Dextrose Agar. Maximum percentage of isolates were contributed by *Saprolegnia parasitica* (52%) and minimum were of *Achlya Americana* (5%). Temperature, pH and DO of water were measured.

Key words :- *Tilapia mossambicus*, mass mortality, *Saprolegnia parasitica* and Fungal attack.

Introduction:-

Like other fresh water fishes *Tilapia* is also subjected to fungal infection. Stress is one of the causes of mycotic infection leading to mass mortalities. Pillai, 1983 reported *Fusarium* as the cause of mass mortality of fish. Sharouny and Badarn, 1995 reported seventeen species of Oomycete fungi in causing fungal infection in *Tilapia*. Okene and Olufemi, 1997 isolated eighteen fungal species in *Tilapia* cultured ponds in Nigeria. Some other workers have also reported fungal infection in *Tilapia* were Osman et al., 2008; Refai et al., 2010; Ali et al., 2011; Hussian et al., 2013 and Eli et al., 2011. In India very less work have been reported on mycotic infection in *Tilapia* and the role of environmental factors in mass mortality. Present study have been designed to find out the fungal species associated with infection leading to mass mortality of fish.

Material and Methods

For present investigation, *Tilapia mossambica* were found infected with fungal infection in university pond during December, 2012. Mass mortality was observed. A total number of 196 fishes showing external symptoms were collected from pond and examined in laboratory to note external symptoms and to prepare cultures for identification of species of fungi. Cultures were prepared by taking small inocula from fish. The agar plates were incubated at $16 \pm 2^\circ\text{C}$ for the growth of cultures. Growth of colonies were observed in 24 hours. For full growth of colony, plates were kept for 4-6 days in incubator. To avoid bacterial contamination all the glass wares, instruments and media were sterilized, along with all aseptic conditions, Streptomycine sulphate 100mg/ml were used in media. Inoculation was done in Laminar flow in sterilized conditions.

Preparation of pure cultures. Pure cultures were prepared by picking up small tuft of mycelium and grown on agar media (Potato Dextrose Agar). For the development of zoospores and reproductive structures cultures were prepared on baits. For that pretiplates were filled with 20-30 ml sterilized tap water, small piece of media with fungal growth were kept in these plates and baited with different baits at temperature $16 \pm 2^\circ\text{C}$. Glycine seeds (Soyabean seeds) and Sorgham seeds (Jowar) were used as baits.

Identification of the isolates

All pure cultures were examined for colonial growth, morphological features and microscopical characteristics. For identification, slides were prepared from each colony by taking small tuft of mycelium and stained with Lacto-phenol cotton blue. The slides were observed under microscope. Identification of fungi was carried out on the basis of keys of Coker, 1923; Johnson, 1956, Khulbe, 1993 and 2001.

Water temperature, pH and Dissolved oxygen have been measured by using mercury thermometer, Digital pH meter and D.O meter (Elite-Hach)

Results and Discussion:-

A total number of 684 fishes were died in culture pond in University campus . Among them 196 infected fishes were examined in living condition. Infected fishes showed cottony mycelium over the body, descaling was observed in most of the fishes .Some eroded scales with with starting stage of lesions was observed .Anterior region was found most affected area . In some fishes mouth was found completely covered with hyphae and mycelium was also observed covering the eyes..Fishes were observed regularly for one month. Fig .1, 2&3



Fig -1. Showing fungal hyphae covering whole body surface of Tilapia.



Fig-2. Showing moribund fishes infected with fungus collected from culture pond.



Fig-3. Showing cottony mycelium covering eye and fungal growth inside mouth.

All the fishes showed visible infection in the period of 10-12 days and within 12 days mass mortality was observed. Mass mortality in Tilapia due to mycotic infection have been reported by Pillai, 1983.

Isolation and culture of fungi revealed the presence of six different species of zoosporic fungi viz. *Achlya Americana*, *Achlya proliferoids*, *Aphanomyces laevis*, *Pythiopsis species*, *Saprolegnia diclina* and *Saprolegnia parasitica*. Most of the fungi appeared as mixed infections. Maximum percentage of isolates were contributed by *S. parasitica* (52%). It was found most virulent and pathogenic fungi. Among the isolated species, (13%) of isolates were of *S.diclina* followed by *A.laevis* (12%), *Pythiopsis* (10%), *A.proliferoids* (8%) and *A Americana* (5%). These findings have been supported by the reports of Sharony and Budana, 1995 who isolated the given species of fungi from *T.mossabicus*. Above findings are also supported by works of Ashran et al., 2007; Abu and Atta, 2008; Refai et al., 2010 and Hussian et al., 2013. **Fig.4.**

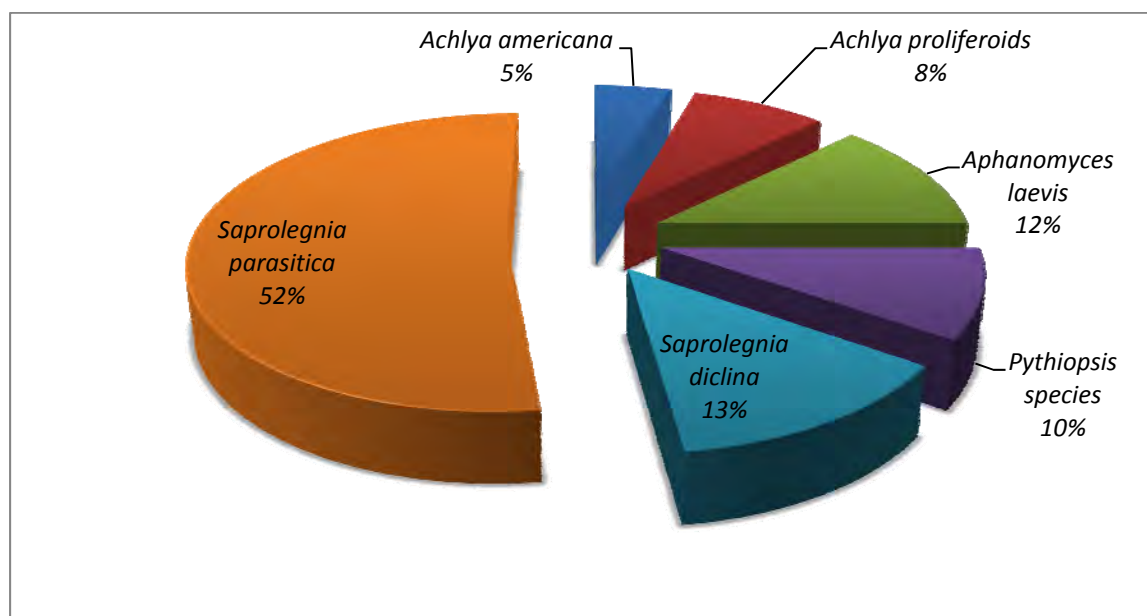


Fig-4. Species wise % of isolated species of fungi from infected Tilapia

Water temperature recorded during the study was 16 ± 2 °C ; p H values recorded were 7.9 ± 1.6 and Dissolved oxygen during the study was found to be 6.8 ± 3.0 mg/l. Low temperature in December and alkaline p H along with high DO favours the growth of fungi and concentration of spores in the water have been increased. Change in temperature may be one of the stress factor to mak fish more susceptible to fungal infection. These findings are similar to the reports of Khulbe, 2001 and Chauhan, 2014a.

Conclusion:-In the present study it was observed that fungal infection lasts for 10-12 days and in all the fishes and sudden decrease of temperature may be the stress factor along with low temperature and alkaline range of pH increases the concentration of zoospores in water and parasitic activity of fungi. These may be the causes of mass mortality of fish. Study also showed *S. parasitica* is a fast growing fungi with high parasitic activity which leads to fish mortality.

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References :-

- [1] T.W,Johnson The genus *Achlya*: Morphology and Taxonomy. Univ. of Michigan Press, 1956, Ann. Arbor. 180 pp.
- [2] R.D. Khulbe . The parasitic water molds. Publ. Almora. Book Depot, 1993.Almora, pp. 144.
- [3] R.D. Khulbe Fungal attack in fresh water fishes and evaluation of control measures .Aquaculture feed and health , Biotech consortium India Ltd. New Delhi, 2000 pp. 140-152.
- [4] R.D. Khulbe A manual of aquatic *fungi* (Chytridiomycetes & Oomycetes). Daya Publishing Housing House, 2001.Delhi: 255.
- [5] W.C.Coker. The Sarolegniaceae with notes on other water molds.Univ. of North Carolina, 1923,Carolina Press Chapel Hill,North Carolina,USA.201.
- [6] C.T Pillai and Y.M,Freitas .Fungal infection causing mass mortality of fresh water fish *Tilapia mossambica* , CMFR institute, 1983,Cochin,Kerala.
- [7] M M A., Hussian W H ,Hussian and M A. Mohhamad ,Pathogenicity of *Achlya proliferoids* and *Saprolegnia diclina*(*Saprolegniaceae*) associated with saprolegniasis out breaks in cultured Nile *Tilapia*(*Oreochromis niloticus*).World j. of fish and marine sci.2013; 5(2),188-193.
- [8] M.K. Refai, A. Laila, M .K, Mohamed and S.M.S, Shima., The assessment of Mycotic settlement of freshwater fishes in Egypt. J. Amer. Sci.,2010; 6(11), 595- 602.
- [9] Ali, E.H. Hashem,M and AL- Salahy,M.B. . Pathogenicity and oxidative stress in Nile *Tilapia* caused by *Aphanomyces laevis* and *Phoma herbarum* isolated from farmed fish.Dis.Aquat.Organ. ,2011, 16: 94(1):17-28.
- [10] Eli,A , Briyai, O.F and Abowel,J.F.N. A review of some fungi infection in African fish *Saprolegniasis*, Dermal mycoses; *Branchiomyces* infections , Systemic mycoses and *Dermocystidium*. Asian journal of medical sciences. 2011 .3 (5):198-205.

- [11] H.M ,Osman, , W.E ,Solman, , A.E, Noor, E.I ,Deen, and A.L Mohamed, Induction of Saprolegniasis in Oreochromis niloticus with special reference to its biological control. *Glo bal veterinaria*2, 2008. (1):32-37.
- [12] El Ashram, A.M.M., A.M. Abd El Rhman and S.F. Sakr, A contribution to saprolegniosis in 21. Smith, S.N., R. Chohan, S.G. Howitt and cultured Nile tilapia (*Oreochromis niloticus*) with special reference to its control. *Egyptian Journal of Aquatic Biology and Fisheries*, 2007, 2: 943-955. 98: 389-395
- [13] Abou El and M.E, Atta.. Saprolegniosis in freshwater cultured tilapia (*Oreochromis niloticus*) and trials for control by using Bafry D50/500. *Proceeding of 8th International Symposium on Tilapia in Aquaculture*, 2008, Cairo, Egypt, pp;1403-1418.
- [14] R, Chauhan. Diversity of aquatic fungi of six water bodies of Bhopal in relation to its abiotic parameters. *Int. J of Green and Herbal Chemistry*, 2014, Vol.3, No.2, pp.425-433.
- [15] A.N, Okaeme and B.E, Olufemi Fungi associated with Tilapia culture ponds in Nigeria. *J. Aquat. Trop.* 1997.12:267-274.
- [16] H.M Sharouny and R.A, Badran. Experimental transmission and pathogenicity of some species of zoosporic fungi to Tilapia fish. *Mycopathologia* ,1995,132(2): 95-103.