

It is closely comparable to the *M. regulare* reported from southern Shaanxi and also resembles with *Micrhystridium* sp. of Lo described from the Lower Yudoma suite of eastern Siberia<sup>11</sup>. The presence of *Veryhachium* is unique in the assemblage. Small shelly fossils are not reported from this locality so far, but the acritarchs-yielding level in the Korgai syncline has also yielded SSFs in Mussoorie and Garhwal synclines<sup>12-15</sup> and also in Nigalidhar Syncline<sup>16</sup>. This study shows a close similarity in the microfossil yield of the basal Meishucunian levels in the Lesser Himalaya and the Chinese section. Simple *Micrhystridium* population also occur in the lower Yudoma Formation in Siberia<sup>11</sup>. Besides, the acritarchs of basal Tal Formation differ greatly from those of Infrakrol Formation<sup>3</sup>, where large acanthomorphic acritarchs (> 100 µm in diameter) predominate. The presence of smaller acanthomorphic acritarchs in the basal Tal (Cambrian) in comparison to the older Infrakrol Formation (Precambrian) strengthens the concept of decrease in the size of acanthomorphic acritarchs from the Precambrian to the Cambrian<sup>17</sup>.

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## Potential threat to reservoir fishery by fungi in Kumaun Himalaya, India

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Zoosporic fungi are among the most destructive fish pathogens. Eighteen species belonging to *Achlya*, *Aphanomyces*, *Dictyuchus*, *Saprolegnia* and *Pythium* were isolated from Nanak Sagar, which has witnessed mass mortality of fish fauna during 1991-94. Of these, 8 species were also detected from diseased and moribund fish. Species of the parasites and the hosts demonstrated differential pathogenicity and immunity respectively. *Achlya flagellata* and *Saprolegnia parasitica* appeared virulent, while scaly-fish were more vulnerable to fungal infection than the non-scaly fish. The occurrence of fungal species, zoospore density and severity of mycoses was primarily governed by temperature. A water temperature between 22 and 25°C during spring stimulated fungal activity whereas, at above 28°C it retarded the disease process. All the species, except *A. inflata* and *P. vexans* have been reported virulent fish pathogens elsewhere; this is cause of great concern. By virtue of a wide distribution and potentiality to parasitize eggs, fingerlings and adult fish, these fungi along with anthropogenic pressure are most likely to pose threat to reservoir fishery. Thus, research on integrated management of fish mycoses is warranted.

RESERVOIRS, the artificial ecosystems characterized by the existence of both lentic and lotic components, constitute the prime inland fisheries resource in South and South-east Asia. A major increase in inland fish yield in India is expected from reservoirs because of construction of new dams and the improvement in reservoir fisheries management<sup>1</sup>. Beside wide diversity of fish, reservoirs also harbour diverse microorganisms including fungal species which occur as opportunistic pathogens of aquatic organisms, specially of fish<sup>2,3</sup>. Water molds, the zoosporic fungi, are a constant and ubiquitous component of aquatic environments and a continual challenge to fish<sup>4-6</sup>. The ability of these fungi, particularly of those belonging to *Achlya*, *Aphanomyces* and *Saprolegnia* to parasitize eggs, fry, fingerlings and adult fish has been well documented<sup>5-19</sup>. As a general rule, infection starts when the host gets injured either mechanically<sup>9,10</sup> or as a result of infection other than fungal, which often results in epidemic, causing 100% mortality of the infected hosts<sup>10,13,18</sup>. However, the clinical and pathological investigations have revealed that several members of Saprolegniales often act as primary pathogens<sup>5,6,11-19</sup>, thereby play a vital role in minimizing the total output

of fish industry either way. In a preliminary study, prevalence of mycoses was observed at a reservoir in Nainital district<sup>19</sup>. However, in spite of their great economic value, the zoosporic fungi of the reservoirs are least explored.

Kumaun region abounds in lakes and reservoirs. Behgul, Nanak Sagar, Tumaria and Dhaur harbour a high diversity of fish fauna and are well recognized fish production centres of Government of UP, India<sup>20</sup>. During a recent exploration (1991–94) of Kumaun Himalaya for water molds, fungal infection was noticed on several species of fish both in lakes<sup>17</sup> and reservoirs<sup>18,19</sup>. A high prevalence of mycoses on fish species, was, however, observed in Behgul and Nanak Sagar reservoirs<sup>19</sup>. Considering the seriousness of the situation, and importance of reservoirs; occurrence, phenology and pathogenic behaviour of water molds to fish and the influence of physicochemical properties of water on the occurrence and infection of zoosporic fungi were investigated at Nanak Sagar.

A large number of individuals of each major contributing fish species were randomly inspected at the fish catching site of Nanak Sagar (29°55'N and 79°40'E; 200 m amsl) spread over 4662 ha in UP hills. Minute signs of disease were observed using a powerful hand lens. Water samples were also collected at a month interval for two years. Certain important characteristics of the water were also estimated following the standard methods of the American Public Health Association<sup>21</sup>.

Symptomatic fry and adult fish were collected in sterile polythene bags, placed in ice in aseptic glass pots and immediately brought to the laboratory. Portions of infected tissue were washed individually with several changes of sterile tap water, and treated with 0.01%

potassium tellurite solution to avoid bacterial contamination. The infected tissues were then aseptically transferred to sterilized petri plates and baited with sterilized hemp seed (*Cannabis sativa* L.) halves. To isolate zoosporic fungi from water, 20 ml of each sample at different dilutions was poured into a sterilized petri plate and baited with different baits<sup>6</sup>. The plates were incubated at 15°C and fungi that colonized were purified, cultured and identified following various monographs<sup>6,21-22</sup>.

In all, 18 species of zoosporic fungi belonging to 5 genera were isolated (Table 1). *Achlya* and *Saprolegnia* constituted a major proportion, were represented by 8 and 4 species, respectively. Zoospore density of these taxa was recorded in the range of  $5-25 \times 10^3 \text{ L}^{-1}$ , which was higher than the species of *Aphanomyces*, *Dictyuchus* and *Pythium* ( $2.5-5 \times 10^3 \text{ L}^{-1}$ ). These fungi exhibited seasonality of their occurrence, which was closely related with water temperature. Maximum number of species as also high zoospore density of individual species were recorded during February and March (Spring) while a minimum in May and June (Summer). A few species, viz. *Aphanomyces laevis*, *Dictyuchus sterile*, *Saprolegnia ferax*, and sterile isolates of *Achlya* and *Pythium* could resist the summer temperature ( $> 32^\circ\text{C}$ ). Majority of these species have shown adaptability to different habitats in the region<sup>24</sup>.

Of the fungi recovered from water, eight species were also isolated from individuals of 5 fish species exhibiting mycotic symptoms (Table 1). Infection is characterized by depigmentation of body colour, external haemorrhage, descaling, eroded tails and dorsal fins which led to surface swimming and sluggishness and ultimately to death. Colonization of whitish woolly tuft on to infected

Table 1. Occurrence, seasonality and parasitic association of water molds to fish in Nanak Sagar, and elsewhere

Fungal species	Seasonal periodicity	Associated with fish mycoses	
		Nanak Sagar (Reference)	Elsewhere (Reference)
<i>Achlya americana</i>	Spring, rainy		6, 15, 10, 13
<i>A. debaryana</i>	Spring, rainy	18, 19	
<i>A. flagellata</i>	Autumn, spring	19	6, 7, 17
<i>A. inflata</i>	Autumn, winter		
<i>A. klebsiana</i>	Spring–autumn	19	6
<i>A. orion</i>	Rainy, autumn		5, 12, 13
<i>A. prolifera</i>	All season		13
<i>Achlya</i> sp.	All season		6
<i>Aphanomyces laevis</i>	All season	19	13
<i>Aphanomyces</i> sp.	All season		13
<i>Dictyuchus sterile</i>	Spring–rainy		6
<i>Saprolegnia diclina</i>	Autumn–spring	19	6, 17
<i>S. ferax</i>	All season	19	6, 10
<i>S. parasitica</i>	Autumn–spring	19	5, 8, 10, 11, 16, 17
<i>Saprolegnia</i> sp.	Winter–spring		6, 10
<i>Pythium undulatum</i>	Rainy–winter		6, 15
<i>P. vexans</i>	Autumn–spring		
<i>Pythium</i> sp.	All season	19	6

portion is often visible. *Saprolegnia parasitica* was detected from *Mastacembelus armatus*, *Mystus vitatus*, *Tor putitora* and *T. tor*; *S. ferax* from *M. armatus*, *Nandus nandus* and *T. tor*; *A. debaryana* from *M. armatus*, *T. putitora* and *T. tor*; *A. flagellata* was found to be associated with *T. pititora* and *T. tor*; while *A. klebsiana*, *Pythium* sp. infected *N. nandus* and *T. tor*, respectively. These pathogens caused identical signs in different hosts. However, the disease intensity varied with the host and the pathogen.

The infection began during the first week of February and by the middle of the month, substantial proportion of the fish population got infected. By that time, incidence of mycoses on *M. armatus*, *M. vitatus*, *N. nandus*, *T. pititora* and *T. tor* was recorded 36.6, 32.0, 29.8, 28.4, 33.6 per cent, respectively. The infection spread rapidly and peaked by the middle of March, decreased thereafter and was minimum in June when the water temperature was high (32°C).

Water temperature between 22 and 25°C, coupled with a high pH (8.6–8.7) and moderate DO (8.5–9.5 mg/l) favoured zoosporogenesis in fungal forms and their subsequent infection on fish. A maximum disease incidence in March coincided with a water temperature of 23°C, a pH of 8.6 and a DO of 9.5 mg/l. Temperature more than 28°C of May through June, suppressed the growth and pathogenicity of the fungal species, resulting in minimum disease incidence of fish<sup>19</sup>. Retardation of the pathogenic potentiality of the species at higher water temperature may be due to induction of gametes and oospore formation which cannot infect fish and require a certain period of dormancy<sup>18,19</sup>. Since July to September is breeding season for the fish, the reservoir is closed for fishing from July to November. Because of this regulation, no data on infection could be available for this period. However, fish eggs which are highly susceptible to fungal infection<sup>5,6,10,13</sup>, obviously were destroyed during this period. As water molds were abundant during September onwards, their infection on fingerlings and adult fishes is likely. This may be one of the reasons for dwindling fish yield of the reservoir.

Several species of water molds have previously been reported as causal agents of diseases and death in fishes<sup>4-19</sup>. *A. flagellata* which has long been known as fish pathogen<sup>7</sup>, has recently been reported to be associated with *T. tor* and *Puntius ticto* in temperate lakes<sup>17</sup>. Similarly *S. parasitica* parasitized four fish species in Nanak Sagar<sup>19</sup>, has also shown potentiality to parasitize several species of fish<sup>5,8,10,11,16,17</sup>, *A. debaryana* was responsible for epizootic infection on a cat fish<sup>18</sup>. Similarly, *A. klebsiana* on *P. conchionius*, *A. laevis* and *S. diclina* on *P. ticto* and *S. ferax* on *Tor tor*<sup>6</sup> have been reported to be pathogenic.

Moreover, fungal species which were isolated from reservoir but not found associated with fish during the

present investigations, have been reported virulent pathogens of eggs, fry, fingerlings and adults of several fish species elsewhere<sup>5,6,11</sup>. *A. americana* has been reported as parasite of freshwater fish *Lepomis macrochirus*<sup>10</sup> and several tropical and temperate fish<sup>16</sup>. Likewise, *A. orion* has shown its pathogenicity to twelve species of freshwater fish, four belonging to *Puntius*, *Chanda*, *ranga*, *Channa marulion*, *C. punctatus*, *Notopterus notopterus*, *Labeo rohita*, *Colisia lalia* and *Anabas testudineus*<sup>5,12</sup>. *A. laevis*, *D. sterile* and *P. undulatum* have also been reported pathogenic to cold water fish<sup>6,14</sup>. Fish eggs are also attacked by these fungi, infected eggs generally do not hatch and are destroyed, thus causing a colossal loss to the fish industry<sup>5,6,10,13</sup>. A heavily sporulating sterile isolate of *Aphanomyces* has been reported to cause 100% mortality in eggs of *Channa gachua*, while *A. prolifera*, *A. orion* and *Aphanomyces laevis* caused more than 90% egg mortality<sup>13</sup>.

Foregoing discussion reveals that water molds flourishing in Nanak Sagar have wide distribution, and possess great potentiality to parasitize fish<sup>6</sup>. Most of these have a wide host range and any of them can cause substantial loss under congenial conditions. Moreover, a rich mycoflora of parent rivers and within their catchments contribute to the mycoflora of reservoir(s) in terai region. Once established in the reservoir by virtue of parasitism in fish or saprophytic existence, these fungi produce gametes, chlamydo-spores and oospores that persist for many years and are virtually impossible to eliminate, thus, may pose threat to fish under congenial conditions. Further, the more or less similar mycoflora, Ichthyofauna and physio-chemical properties of water of different reservoirs in terai region has made reservoir fish vulnerable to fungal infection. Under intensive system, such as hatcheries and fish farms mycoses has always been a problem and unless adequate hygienic and husbandry methods are employed, grave losses can result. In some habitats these organisms have been responsible for extinction of some fish species. Himalayan Mahseer, *T. pititora*, *T. tor* and some other valuable and rare species of Himalayan fish which are disappearing, are prone to fungal infection<sup>19</sup>, which beside anthropogenic pressure may contribute to their fast depletion. Unistam<sup>25</sup> also reported that water molds were responsible for the extinction of fish species in certain habitats of Australia. Thus, there is an urgent need to undertake prophylactic measures to protect the fingerlings before introducing into the reservoirs. These should be treated with Melachite green, tetracycline, formaline, boric acid and other suggested chemicals in recommended concentrations<sup>26-28</sup>.

In addition, several species of *Olpidiopsis* are known to parasitize on Saprolegniales. *O. achlyae* on *Achlya* sp., *O. fusiformis* on *A. prolifera*, *O. luxurians* on *A. laevis*, *O. saprolegniae* var. *laevis* on *Saprolegnia* sp.

and *O. pythii* on *Pythium* sp. have been reported from Kumaun<sup>24</sup>. Thus, hold potential to suppress population of fish pathogens; and intensive research on biological control of fish mycoses is warranted.

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