# Incidence of ulcer type of disease in wild fishes of Bangladesh

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#### Abstract

Disease occurred in wild fish species investigated in different water bodies like canals, ditches, beel, haor, flood plain etc. in 17 districts of Dhaka division. Haemorrhagic lesions were observed on the body surface of affected fishes. Incidence of the disease in the investigated water bodies ranged from 0 to 100%. In total 19 fish species were found to be affected and prevalence of infection ranged from 0.0 to 100.0%. *Channa punctatus* and *Puntius ticto* were severely affected in all locations. Percentage of infection in these fishes ranged from 0.0 to 100.0. The highest infection was observed in Netrokona, Kishoreganj, and Mymensingh districts. Bacterial genera isolated from the lesions of these affected fishes were *Aeromonas, Pseudomonas, Flavobacterium, Micrococcus*, and *Staphylococcus*. Among these isolates *Aeromonas* was the dominant. Abundance of *Aeromonas* in the lesions among the investigated bacteria ranged from 75 to 90%. Five identified *Aeromonas hydrophila* were examined for their pathogenicity and were able to infect the experimental fish, silver barb (*Puntius gonionotus*). The pathogen *Aeromonas hydrophila* was thus considered to have an association with the outbreak of ulcer type of disease in the investigated fish species.

Key words: EUS, Aeromonas hydrophila

## Introduction

Bangladesh is uniquely endowed with the diverse of very rich and extensive inland and marine fishery resources which mainly include rivers, flood plains, estuaries coastal belt and vast sea waters. Disease has become a major problem in fish production both in culture system and wild condition in Bangladesh (Rahman and Chowdhury 1996). In Bangladesh the fish has been suffering from ulcer type of diseases of different expressions including epizootic ulcerative syndrome (EUS), bacterial haemorrhagic septicaemia, tail and fin rot, bacterial gill rot, dropsy, columnaris disease, fungal disease and parasitic disease (Chowdhury 1997). In an investigation of the fish farms by spot B. Majumder et al.

observation many farmed fish species were found to suffer from diseases caused by bacterial pathogens (Chowdhury 1993).

Considering the importance and immediate need for the country research works, the present study was planned to achieve the prevalence of the gross fish disease occurred in wild fish species in Dhaka division, to determine the percentage composition of bacteria in the lesions of the affected fishes and to perform pathogenicity test of some recovered aeromonad isolates suspected as pathogen.

## Materials and methods

#### Investigation of the occurrence of fish disease

Investigation of the occurrence of disease in fishes were carried out on the basis of observation and sampling of fish from different waterbodies in 17 districts of Dhaka division of Bangladesh starting from November'98 and continued up to March'99. Fish were sampled randomly from three wild waterbodies (canal, beel, haor, ditches and flood plain) of one randomly selected thana of each district. Gross diagnosis of ulcer type of disease in the investigated fish was based on eye observation on the spot having haemorrhagic lesions on the fish body. A total of 100 fish samples were grossly examined from each waterbody. Total fish species and the infected fishes were counted and percentage of infection was calculated.

## Collection of fish

Mymensingh, Netrokona, Kishoreganj, Tangail and Jamalpur among 17 districts were selected for collection of severely affected fish for isolation of bacteria. Bacterial isolates were collected from the lesions of body surface of the farmed fish viz., Channa punctatus, C. striatus, Mastacembelus armatus, M. pancalus and Puntius ticto. The samples were brought to the laboratory immediately after collection for bacteriological study.

## **Bacteriological Investigation**

Tryptone Soya Agar (TSA) was used for culture and swabs were aseptically taken on the culture plate with a sterile inoculating loop and spreaded. The plates were incubated at 25°C for 36-48 hours. Twenty colonies were randomly separated from this initial culture and individual culture was maintained on TSA slants at 4°C for characterization. Morphological and biochemical characters were studied following the methods described in the Cowan and Steel's Manual for the identification of Medical Bacteria edited by Barrow and Felthem (1993) and confirmed with the help of Bergey's Manual for Systematic Bacteriology (Volumes 1 and 2) edited by Krieg and Holt (1984). Percentage composition of the identified bacteria isolated from the lesions was based on the 20 colonies initially separated.

## Artificial infection in fish

For pathogenicity test five identified A. hydrophila isolates were tested on silver barb (Puntius gonionotus) of about 15 g in weight. The experimental fish were acclimatized for 3 days in the laboratory condition and checked their health before use in the experiment. Stock suspension of individual bacteria was prepared in sterile tap water taking 18-24 h culture. In a 40l capacity aquarium, 15l of bacterial suspension was prepared in the tap water with the stock suspension in such way that the bacterial dose become  $3-5\times10^8$ CFU/ml. The exposure dose of bacteria was followed after Chowdhury (1997). Ten fish were exposed to bacterial suspension in the aquarium under aerated condition at room temperature ranged 27-28°C. After 24 h of exposure 80% of bacterial suspension was exchanged with tap water and from the following day 60% of water was exchanged at every 24 h. Two replications were set up for the same isolate and thus in total 20 fish were used for each bacterial isolate. Experimental period was 2 weeks. For each set of experiment control fish were maintained in the same way without exposure to bacteria. No food was applied during the experimental period. Infection was recorded by observation of lesions, mortality and continued by re-isolation of bacteria from the experimental fish.

## **Results and discussion**

Incidence of ulcer disease in the investigated natural waterbodies were recorded from 0.0 to 100.0% in different districts (Fig. 1). The result was correlated with the reports of Chowdhury (1998) where he observed 40 to 50% of the non-farmed waterbodies were affected by ulcer disease at Mymensingh in Bangladesh.



Fig. 1. Prevalence of EUS disease in natural waterbodies in different districts of Dhaka.

A: Dhaka B: Narayanganj C: Munshiganj D: Narshingdi E: Manikganj F: Gazipur G: Mymensing H: Kishoreganj I: Netrokona J: Jamalpur K: Sherpur L: Tangail M: Faridpur N: Madaripur O: Shariatpur P: Rajbari Q: Gopalganj

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Nineteen different types of wild fishes viz., Amblypharyngodon mola, Anabas testudineus, Channa orientalis, C. punctatus, C. striatus, Clarias batrachus, Colisa fasciatus, Glossogobius giuris, Heteropneustes fossilis, Macrognathus aculeatus, Mastacembelus armatus, M. pancalus, Mystus aor, M. vittatus, M. tengara, Nandus nandus, Notopterus notopterus, Puntius ticto and Wallago attu were found to be affected by the ulcer disease. Percentage of infection in wild fish species by ulcer disease ranged from 0.0 to 100.0%. The disease was most prevalent in Netrokona, Kishoreganj and Mymensingh districts and Channa punctatus and Puntius ticto were found to be severely affected in case of wild fish species (Table 1). Barua et al. (1990) observed 22 different species affected by ulcer disease and percentage of fish infection ranged from 0.0 to 100.0 and the disease was most prevalent in Bogra, Netrokona and Chandpur.

Bacterial genera recovered from the lesions of affected wild fishes were identified as *Aeromonas, Pseudomonas, Flavobacterium, Micrococcus* and *Staphylococcus*. The results were correlated with the findings of Chowdhury and Baqui (1997) where they isolated the bacterial flora from fish organs were *Aeromonas*, Coryneformes, *Flavobacterium*, Enterobacteria, *Maraxella, Acinetobacter* and *Bacillus. Aeromonas* was detected as the dominant bacteria in the lesion of affected farmed fish species and varied from 75 to 90% of the total bacterial count which are shown in Fig. 2. *Pseudomonas* was the second dominant bacteria found in the lesion of affected fish species. According to Chowdhury et al. (1995), *Aeromonas* sp. as the dominant bacteria with high percentage in the kidney of EUS affected fishes in Bangladesh. Rahman et al. (1998) also reported that *Aeromonas* sp. in the kidney of different affected fishes were found to be 80-90% of the total bacterial content.



Fig. 2. Comparative percentage composition of bacterial genera in the investigated farmed fish species.

In the pathogenicity test, all the *A. hydrophila* isolates causing an ulcer type disease and mortality. In this study all of the selected isolates were able to cause lesions and mortality in the experimental fish. Appearance of lesion and mortality of fishes varied

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from one isolate to another. The isolate AA2 showed highest infection (100%) with 90% mortality (Fig. 3).

**Fig. 3.** Patterns of the appearance of lesions and mortality caused by different *A. hydrophila* isolates in experimental infection.

Chowdhury (1998) found 75-80% mortality in carp and catfish with exposure dose  $3-5x10^8$  CFU/ml of *Aeromonas hydrophila* isolates experimentally and the present study support the previous work.

The present study provides information about the occurrence of ulcer disease in farmed fishes of Dhaka division. It also provides association of the bacterial pathogen A. *hydrophila* with the disease. Knowledge of this study will be helpful to fish pathologist, fish culturists and researchers to detect ulcer disease and to take necessary measures against the bacterial pathogen.

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(Manuscript received 9 March 2000)