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***Mycobacterium marinum* Infection in Caviar Fishes and Fisherman's in a Caspian Sea Province in North of Iran**

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Abstract: *Mycobacterium marinum* is the etiologic agent of fish tank granuloma in human. Determination of the fish tank granuloma in Fishermen and *Mycobacterium marinum* infection in Caviar fishes in Ashorade, in south east Caspian Sea were our objectives. Any suspected lesions in 387 subjects of Fishermen and 113 autopsy samples from gills of fish of Caviar were obtained and culture in Lowenstein Jensen media. The mycobacterial species were determined by conventional biochemical tests. No fish tank granuloma was proved in human, but 11 (9.73%) strains of Mycobacteria were isolated from Caviar fishes, that only 2 strains (1.76%) belong to *M. marinum*.

Key words: Caviar fish, *Mycobacterium marinum*, Ashorade, Caspian sea

INTRODUCTION

Mycobacterium marinum is the etiologic agent of fish tuberculosis (Morales and Dunker, 2001) or fish Mycobacteriosis that has a wide distribution in nature. It is an atypical photo chromogen Mycobacterium that classified in Runyon group 1 and found in salt and fresh water. *Mycobacterium marinum* was described by Aronson (1926) from dead fish in a marine aquarium on display at the zoological garden in Philadelphia (Ang *et al.*, 2000).

M. marinum infection, commonly known as fish tank granuloma (Aubry *et al.*, 2002; Cassetty and Sanchez, 2004), occurs following trauma to an extremity that is in contact with an aquarium, salt water, or marine animals and usually presents as a localized granuloma. Fishermen and workers who process saltwater fish, Workers who clean saltwater aquariums, oyster workers, swimmers and aquarium workers are predisposed. A papule or bluish nodule initially appears at the site of trauma. The incubation period of *M. marinum* infection in human is about 2-3 weeks; this nodule subsequently may ulcerate and involve the local joint or tendons. Localized pain and induration are common. Fever, localized lymphadenopathy and systemic infection rarely are observed with the exception of immunosuppressed patients. Tenosynovitis is the most common manifestation of deep invasion,

although septic arthritis and osteomyelitis are well described (Lahey, 2003; Collins, 1985; Leuenberger and Bodmer, 2000).

The localized lesion can increase slowly over several months. These lesions will usually spontaneously heal in 1-3 years (Murdock *et al.*, 1992).

This study was setup to determine the fish tank granuloma in Fishermen and workers who process caviar fish and determination of *Mycobacterium marinum* infection in Caviar fish in Ashorade, in the Golestan province in the south east of Caspian Sea.

MATERIALS AND METHODS

A number of 387 subjects of Fishermen and workers who processed Caviar fish in Ashorade, in Golestan province in south east Caspian Sea, were examined for any types of suspected lesions in the hands, feet and other parts of the body, during the year 2003. Sampling was carried out either by aspiration or biopsy if the granulomatous lesions were observed.

One hundred and thirteen autopsy samples were also obtained from gills of Caviar fish which on examination had grayish lesion with unpleasant odor.

The collected samples were cut to tiny pieces by sterile scissors and were placed to 5 mL of sterile normal saline, then macerated and treated by adding the same

volume of NaOH 4%, shake it 20 min and then neutralized by adding buffer phosphate (pH = 7), It was centrifuged in 3000 rpm for 30 min and cultured on Lowenstein Jensen media incubated at 25 and 37°C.

The growth were assessed every 2-3 days for a period of 1 month and the isolated colonies were evaluated by growth rate, morphology of colony, pigmentation in dark and light and the results of a battery of standard biochemical tests: catalase production test at 25 and 68°C, growth on MacConkey agar plate, NaCl tolerance, niacin production test, tellurite and nitrate reduction test, urease test and Tween 80 hydrolysis (Isenberg, 1998).

RESULTS AND DISCUSSION

From 387 human subjects only one suspected lesion was observed, but *Mycobacterium marinum* was not isolated.

From 113 gills of suspected fish, we isolated 11 (9.73%) strains of Mycobacteria, that only 2 strains belonged to *M. marinum* and the other 9 isolated strains belonged to other environmental Mycobacteria (Table 1).

In this study, only 2 cases of *M. marinum* were isolated from a total of 113 of Caviar fish. There have been some studies in Iran on the presence of *Mycobacterium marinum* and other environmental Mycobacteria in which the prevalence of *M. marinum* demonstrate to be from 7.2% in soil in Mazendran (Rastegar, 1992), 1.6% in breeding pool in Gilan and Mazendran (Ghazesaeed and Mohammad, 1997) and 1.2% from soil in Golestan provinces (Ghaemi *et al.*, 2006).

Gilan, Mazendran and Golestan are 3 provinces in the north of Iran in the south of Caspian Sea.

According to present results in this study and other reports in Golestan province (Ghaemi *et al.*, 2006), it seems *M. marinum* for caviar fish, is not a serious risk factor in this region.

We could not find any fish tank granuloma in Fishermen and workers who process Caviar fish in Ashorade, but some studies in other countries show few cases of this disease such as annual incidence is 0.27 cases per 100,000 in adult patients in USA (Kiel, 2006), 12 cases during 1991-95 in Germany (Leuenerberger and Bodmar, 2000), isolation of 39 cases of *M. marinum* from Spanish men and women during 1991-98, which 35 cases related to the fishes (Casal and Casal, 2001), report of 38 cases in national center of Singapore during 1995-97, out of this 34.2% belong to subjects with fishing industry and 10.5% in those with occupational contact with the fishes (Ang *et al.*, 2000), the detection of 53 cases of the diseases due to contact with fish tanks in France during 1996-98.

Table 1: The frequency of isolated mycobacterium strains from the gills of caviar fish in Ashorade

Mycobacterium sp.	No.	%
<i>M. marinum</i>	2	1.76
<i>M. fortuitum</i>	2	1.76
<i>M. smegmatis</i>	2	1.76
<i>M. goodii</i>	2	1.76
<i>M. scrofulaceum</i>	1	0.88
<i>M. flavescens</i>	1	0.88
<i>M. zolei</i>	1	0.88
Total	11	9.73

All together, in an internet literature review of 99 articles by Jernigan, 652 cases of Skin infection in human by *M. marinum* were reported during 1966-96 (Jernigan and Far, 2000).

These show that the frequency of this disease is rare but it is more common in fisherman and aquarium contact.

Present research was a one year cross sectional study but according to above investigations from various parts of the worlds it seems that we need a long period follow up for case finding.

We are suggesting a study with a longer period and larger sample population of fishermen and other peoples involved to determine the importance of these Mycobacteria in human disease.

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