SURVEY OF *AEROMONAS HYDROPHILA* IN THREE MARINE FISH SPECIES FROM NORTH WEST ARABIAN GULF, IRAQ

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(Received 21 Otober 2011 , Accepted 16 November 2011)

Keywords; Arabian Gulf , *platycephelus indicus*, hemolytic activity .

ABSTRACT

This study preformed on 74 samples of marine water fishes (24 of *Acanthopagrus lalus*, 27 of *platycephelus indicus* and 23 of *Cynoglossus arel*). Which collected from north west Arabian Gulf of Basrah. All samples were examined for the presence of *Aeromonas hydrophila* in muscles. 24 isolates of *Aeromonas hydrophila* were obtained, (33.3%) from *Acanthopagrus lalus*, (44.4%) from *platycephelus indicus*, and (17.39%) from *Cynoglossus arel*. Then all isolates were examined for their ability to hemolytic activity as a virulence factor, the higher percentage of hemolytic activity isolates was found in *Acanthopagrus lalus*.

INTRODUCTION

Fish is a very perishable, high-protein food that typically contains a high level of free amino acids and volatile nitrogen bases which are essential for human consumption, in addition to high polyunsaturated fatty acids (González-Fandos *et al*., 2005).

The bacterium *Aeromonas* is considered as one of the newly emerging water and food born pathogens (Merino *et al*.,1998; Gugnani, 1999). In fish *Aeromonas* typically causes haremorrahgic septicemia and has been implicated in different outbreaks associated with heavy losses (Son *et al*., 1997). Isolation of these organisms have been reported from a variety of food including fishes (Aditthepchaikram *et al*., 2008).

Aeromonas species are short, gram negative, facultatively anaerobic, non spore forming, motile bacilli with a single flagellum and can ferment glucose with or without producing of gas (Andrade *et al*., 2006). This bacteria is widely distributed in a aquatic environment (Fiorentini *et al*., 1998). This bacteria infect humans and cause septicemia, gastroenteritis, acute diarrhea urinary tract infection and ear infection (Koneman *et al*., 1994; Topic *et al*., 2000).

Aeromonas group produce number of potential virulence factors, including, enterotoxins, haemolysins, cytotoxins and proteases (Burke *et al*., 1982; Ljungh and Wadström, 1983) mentioned that the hemolytic activity is strongly associated with enterotoxin production in members of *Aeromonas* genus. Rogulska *et al*., (1994) reported that the hemolytic activity of *Aeromonas* species act as marker of pathogenicity.

MATERIALS AND METHODS

Isolation and diagnosis:

Seventy four samples of marine fishes (24, *Acanthopagrus lalus*; 27, *Platycephalus indicus*; 23, *Cyanolossus arel*) were collected by net from marine water of north west Arabain Gulf 29 40 787 N 48 43 750 E (Awama 1), then the specimens were transferred to the laboratory under septic condition for bacteriological examination.

Enrichment method used to analyze the samples according to Okrend *et al.,* (1987). 12.5 g of muscle tissue from intestine from each sample was added to 112.5 ml of trypticase soybroth containing 5 mg ampicillin /ml and blended for 2 minutes, Then diluted up to 10^3 in butterfieds phosphate dilutent, then the count was carried out by aforementioned dilution as recommended by Palumbo *et al.,* (1989) using Macconkey manitol ampicillin agar. The number of colonies which showed red color in countable plates was enumerated as *Aeromonas* organism.

Hemolytic activity test

Determination of hemolytic activity of the isolated strains was carried out using 5% sheep blood agar as recommended by Rogulska *et al.,* (1994).

Biochemical tests

Biochemical tests such as gram stain, motility, Indole, Voges proskeur, Methyl red, Urase, H_2S, Nitrate reduction, Catalase, Oxidase, Glucose, Maltose, Sucrose, Hemolysis, Gelatin liquification, Ornithine decarboxylase and NaCl tolerance were used for diagnose *Aeromonas hydrophila*, table (1) was shows that.

RESULTS AND DISCUSSION

This study was preformed to detect the incidence of *hydrophila* in marine fishes, Biochemical tests were used for diagnosis of *hydrophila* as presented in Table (1). The results in Table (2) was shows that 8(33.3%), 12(44.4%), 4 (17.39%) isolates were obtained from *Acanthopagrus lalus*, *Platycephalus indicus* and *Cynoglossus arel* respectively. Figure 1 shows the colonies of *Aeromonas hydrophila*, Figure 2 shows infection in *Cynaglossus areal*.

Table 1: Morphological and biochemical characteristics of *hydrophila* isolated from marine fishes collected from NW Arabian Gulf.

Sr.	Characteristic	*hydrophila* isolated
1.	Gram stains	-
2.	Shape	Rod
3.	Motility	M
4.	Indole test	+
5.	Voges proskeur test	-
6.	Methyl red test	+
7.	Urase test	-
8.	H2S gas	-
9.	Nitrate reduction test	+
10.	Catalase test	+
11.	Oxidase test	+
12.	Glucose	+
13.	Maltose	+
14.	Sucrose	-
15.	Hemolysis	ß
16.	Gelatin liquification	+
17.	Ornithine decarboxylase	+
18.	NaCl 0%	+
19.	NaCl 6%	+

Table 2: Incidence of *hydrophila* in three marine Fish species

Marine fishes	No.of specimens	No.of isolates	Percentage %
Acanthopagrus lalus	24	8	33.3
Platycephalus indicus	27	12	44.4
Cynoglossus arel	23	4	17.39

The present results disagree, with those reported by Okrend *et al*., (1987); Palumbo *et al*., (1989) and Freitas *et al.,* (1992) since the authors pointed out that hemolysin was detected in 100% of *hydrophila* strains recovered from some varieties of food.

Abyta *et al*., (1994) identified *hydrophila* as the primary enterophogenic species, In addition, beta hemolytic strains of *Aeromonas* are assigned to *hydrophila* (Deodhor *et al*., 1991), Varnam and Evanus, (1991) reported that a number of phenotypic characters have been proposed as a markers of enteropathogenicity of *Aeromonas* species and they added that the most important of these markers was hemolysin production.

The information given by the achieved results revealed that *Aeromonas* organism existed in the examined fishes and therefore the foods may play a significant role in the epidemiology of gastroenteritis for human, so the strict hygienic measures, good food handling practice at home, properly clean, sanitary equipments and contact surfaces should be recommended to avoid contamination with *Aeromonas* organism. *Aeromonas hydrophyla* consider as etiologic agent for tail/fin diseases and hemorrhagic septicemia of fresh water fishes.

Fig (1): colonies of *Aeromonas hydrophila*.

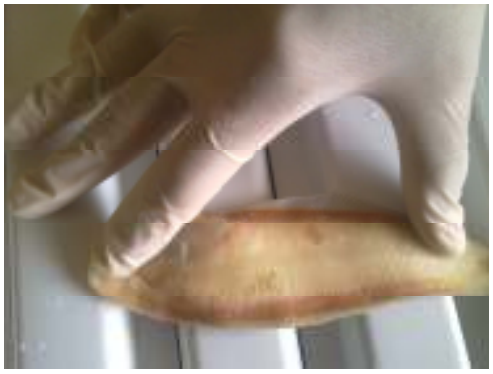

Fig (2): *Cynoglossus arel* infected with *Aeromonas hydrophila*.

مسح لجرثومة *Aeromonas hydrophila* في ثلاثة أنواع من الاسماك البحرية من شمال غرب الخليج العربي - العراق

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الخلاصة

أجريت هذه الدراسة على 74 نموذج من ثلاثة انواع من اسماك المياه البحرية (*Acanthopagrus lalus , Platycephalus indicus , Cyanoglossus arel*) والتي جمعت من شمال غرب الخليج العربي في البصرة. وتم التحري عن تواجد جرثومة *Aeromonas hydrophila* في عضلات الانواع الثلاثة من الاسماك. واظهرت النتائج عن وجود 24 عزلة من الجرثومة منها 8(33.3%) في *Acanthopagrus lalus* و 12(44.4%) في *Platycephalus indicus* و 4(17.39%) في *Cyanoglossus arel*. كما تم اختيار العزلات إلى قابليتها على تحليل الدم كعامل ضراوة، وبينت الدراسة إن العزلة التي حققت أعلى نسبة مئوية للضراوة تم الحصول عليها من *Acanthopagrus lalus*.

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