

VIBRIO ANGUILLARUM AS A STRESS-BORNE PATHOGEN IN CULTURED FRESH WATER FISH

A. A SHAHAT * and E. E MEHANA **

*Department of Food Hygiene. Faculty of veterinary Medicine Cairo University

**Department of Pathology and Parasitology. Faculty of Vet. Med., Alexandria University

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SUMMARY

Vibrosis is an economically damaging infectious disease of cultured fresh water fish characterized by red necrotic lesions of the abdominal musculature, exophthalmici, and erythema at the bases of the fins and around the vent. *Vibrio anguillarum* was recovered from 62% of examined clinically affected *Tilapia nilotica* fish. The percentages of isolation from skin lesions, muscle, kidney, spleen and liver tissues were 35%, 22%, 60%, 48% and 43% respectively. The highest recovery rate of *Vibrio anguillarum* was observed in kidney (60%)

The main characteristic histopathological changes as well as the public health importance of the disease were discussed.

INTRODUCTION

Vibrosis is a classical example of a stress-borne disease in summer, that the losses caused by it are highly dependent on the severity of the environmental stresses that precipitated the outbreak (Edwards, 1995).

Highest environmental prevalence is in organically polluted water, high salinity, overcrowding, high temperature and poor hygiene (Kitao et al., 1983 and Noga 1996).

Vibrio anguillarum is the most common fish pathogen that affecting fresh water as well as marine fishes (Hacking and Budd, 1971). Some vibrios produce hemolysin which may cause anemia and proteases which may cause muscle damage, reducing the keeping quality, marketability of fish and so economically losses (Hjeltnes

and Roberts, 1993). Others is a zoonotic pathogen, causing skin ulcers in humans (Love et al., 1981), gastroenteritis or systemic infections, especially in immuno suppressed individuals (Angulo et al., 1994).

Awareness of microbiological health hazard arising from consumption of contaminated fish has been grown in recent years. Therefore, this investigation was planned to study the presence of *Vibrio anguillarum* in *Tilapia nilotica* cultured at El-Qassiem area., Saudi Arabia.

MATERIAL AND METHODS

One hundred moribund fish were collected after visiting a large farm located at El-Qassiem area, Saudi Arabia. Diseased fish placed in a plastic bag submitted to laboratory without undue delay, where they were examined.

For bacteriological examinations, skin lesions, muscle kidney, spleen and liver tissues were examined bacteriologically by using brain heart infusion agar according to Austin and Austin (1993), Inglis et al. (1993) and Quinn et al. (1994). The incubation was carried out aerobically at 25°C for 1-7 days. Isolates were identified using standard procedures for propagation and identification (West et al., 1983).

For histopathological studies a tissue specimen

from skin lesions and its underlying skeletal muscles, spleen, kidney, liver and intestine were fixed in phosphate-buffered formalin then dehydrated in alcohol, cleared in xylol and embedded in paraffin, sectioned at 5-7 μ in thickness were stained with haematoxylin and eosin (Culling, 1974).

RESULTS AND DISCUSSION

Vibrio anguillarum was isolated from 62 out of 100 examined fish samples 62% (Table 1). These findings agree with those reported by Moustafa et al. (1990) and higher than those reported by El-Gaber et al., (1997). These differences could be attributed to the disease problem in fish aquaculture usually arises from interaction of host, pathogen and environment where the last one is the most hazardous critical factor (Snieszko, 1974).

Highest isolation rate could be attributed to environmental stresses particularly high temperature which may reach 50°C in summer season at El-Qassiem area, Saudi Arabia. Organically polluted water, high salinity, poor hygiene and handling resulting in depression of one or several defensive mechanisms (Ellis, 1981). Moreover, in fish culture conditions, crowding and abrasion of surfaces may greatly increase the risk of skin damage with the consequence that a portal of entry is available for invasive vibrios.

Table (1): Prevalence of *Vibrio anguillarum* in the examined samples of clinically affected *Tilapia nilotica* fish.

Samples examined n = 100	No. of Samples positive	Percentage
Skin	35	35
Muscle	22	22
Kidney	60	60
Spleen	48	48
Liver	43	43
Total*	62	62

*Total incidence.

The clinical signs noticed were skin ulcers with excessive mucus production that giving the dark appearance of skin, abdominal distention, exophthalmia, corneal opacity, anorexia and anemia. These clinical observations were nearly similar to those showed by Hjeltnes and Roberts (1993) and Naga (1995). On the other hand, postmortem findings showed dermal haemorrhage, serosanguinous fluid mixed with blood filling the abdominal cavities, visceral petechiation, splenomegaly, liquefactive renal necrosis and necrotic enteritis produces a catarrhal, yellow, mucoid exudate. These septicemic gross pathological changes were similar to those reported by Hjeltnes and Roberts (1993).

Microscopic examination of skin and the underlying skeletal muscles revealed degenerative, ulcerative and necrotic changes in the epidermal cell layers besides dermal ulcerations and hemorrhage. Edema, melanosis, necrotic dermatitis and granuloma like structure in the underlying dermis were also showed (Fig. 1).

Necrotic dermatitis resulted from the effect of endotoxines produced by *Vibrio anguillarum* and the hemorrhage occurred due to hemolysis which led to anaemia, while melanosis and granuloma like structure occurred as immune defense from fish against against vibrio. These results were in the same line reported by Ellis (1981).



Fig. (1): Skin showing dermal edema, melanosis, dermatitis and granuloma like structure.(H & E, 250).

The highest recovery rate of *Vibrio anguillarum* was observed in kidney (60%) Table 1. These results may be attributed to both lymphatic and blood circulation systems pass through the kidney and spleen, which should be viewed as functioning, amongst other activities, as organs of filtration (Ellis et al., 1976).

Hyperactivation of melanomacrophage centers (MMC), hemosiderosis, depletion of the interstitial hematopoietic elements of the spleen were showed (Fig. 2). Hyperactivated MMC occurred as a defense mechanism, hemosiderosis due to erythrocytes destruction under the effect of hemolysin of the vibrios. The renal tubular cells suffered severe degeneration and necrosis besides interstitial hematopoietic elements depletion, cystic dilated renal tubules and hemorrhage were also observed (Fig. 3). The results of splenic and renal lesions were similar to the results of Hjeltnes and Roberts (1993) and Noga (1995).

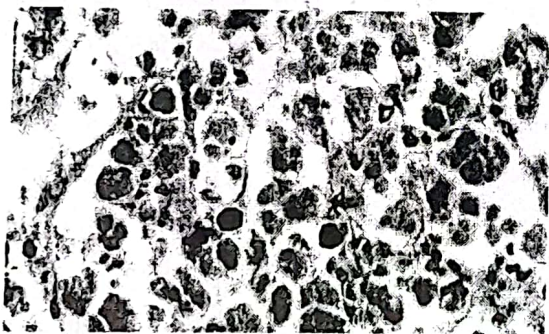


Fig. (2): Spleen showing hyperactivation of melanomacrophages centers, interstitial hematopoietic depletion and haemosiderosis. (H & E, 1000).



Fig. (3): Kidney showing depletion of interstitial hematopoietic elements and necrosis of the renal tubular cells. (H & E, 400).

Microscopically, the hepatocytes showed severe degenerative, necrotic changes and pancreatitis besides lymphocytic infiltration in the portal area and congested hepatic blood vessels. The intestine showed hyperactivated goblet cells, necrotic enteritis with mononuclear cell infiltration and edema of lamina propria with minute eosinophilic granular cells infiltration and lymphocytes (Fig. 4).

The enterotoxines produced by *Vibrio anguillarum* led to the enteric pathological alterations. The same results detected by Abe, (1972).

Conclusions

Vibrio anguillarum is the most serious pathogen of fresh water fishes at present. Infection is in the form of generalized septicemia. Infected fish are anemic, there is a loss of electrolytes and histologically, there is evidence of extensive



Fig. (4): Intestine showed necrotic enteritis. (H & E, 400).

tissue destruction and deep organized granulomatous lesions on various parts of the body may not be apparent until slaughter. These affections reduce the keeping quality and marketability of fish. Avoidance of stresses such as excessive handling, poor hygiene, overcrowding and unbalanced diet. Most promising, as a prophylactic measure is immunization with heat-and formalin killed vaccines. The prophylactic use of antibiotics is not recommended due to the possible selection of resistance strains of bacteria. Controlling the disease by using hazard analysis and critical control points system (HACCP) will minimize its zoonotic hazards.

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