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Assessment of Some Bacterial Zoonotic Microorganisms from Market Fishes in Four Nile Delta Fish Species

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ABSTRACT

Consumption of contaminated fish lead to many of zoonotic bacterial diseases causes severe problems to human health. So, this study aimed to isolate some zoonotic bacteria from fish markets including *E. coli*, *Pseudomonas*, *Salmonella* spp., *Staphylococcus* spp. and Streptococci. This study was carried-out on four hundred samples of fish species classified as; *Clarias gariepinus*, *Cyprinus carpio* (common carp), *Mugil cephalus* and *Oreochromis niloticu* (tilapia) (100 of each). These fishes were obtained from different markets located in Eastern Delta governorates, Egypt. The fishes were subjected to clinical and bacteriological examinations to identify the types of bacterial isolates. The current results indicated that higher incidence of bacterial isolates was observed in *Clarias garipinus*, followed by common carp, *Mugil cephalus* and while low incidence was reported by *O. niloticus*. Although, *Pseudomonas*, Streptococci, *Salmonella* spp. recorded higher loads, while lower was reported by *E. coli* and Staphylococci were observed the public health impotence of zoonotic spp. Isolates were discussed.

Key words: Fresh water fish, Egypt, bacterial infection

INTRODUCTION

Fish is considered one of the most nutritive and highly desirable foodstuffs as fish meat has excellent nutritional value being rich in proteins, vitamins and unsaturated fatty acids. It is also extremely perishable and the safe consumption requires adequate sanitary conditions from the moment of catch, through preparation, sale and consumption (Alghabban, 2014). The most popular freshwater fish species in Egypt are *Oreochromis niloticus, Bagrus bayad* and *Clarias lazera*. The fish flesh, which is the main edible part, the majority of fish infections are usually related to exposed stress.

Most food borne illness caused by *Salmonella* spp., *Staphylococcus* spp. and *Escherichia coli*, usually related to fish consumption. Human infection by fish pathogens is usually through contact with abraded skin, with infected fish while handling or with water or other constituents of an aquatic environment (Acha and Szyfres, 2001; Gauthier, 2014).

In Egypt, *Salmonellae*, *E. coli* and *Staphylococcus* spp. are widely recognized as the principle causes of food poisoning outbreaks occurring due to consumption of contaminated fish and (Hassan and Fatin, 2003).

This study throws a light to detect some bacteria species of zoonotic importance from Nile Delta marketed fish as *Clarias garipinus*, *Cyprinus carpio*, *Mugil cephalus* and *Oreochromis niloticu* collected from Eastern Delta governorates, Egypt, to isolate *E. coli*, *Pseudomonas*, *Salmonella* spp., *Staphylococcus* spp. and Streptococci.

MATERIALS AND METHODS

Four hundred fish samples classified as *Clarias garipinus*, *Cyprinus carpio*, *Mugil cephalus* and *Oreochromis niloticu* (100 of each) were obtained randomly from different markets of Eastern Delta governorates in Egypt.

Clinical examination of fish was performed according to the method described by Schaperclaus (1993). Samples were taken from external body surface at the area between dorsal fin above the lateral line and another were taken from the ventral aspect of the pectoral fin using a sterile bacteriological loop, while sampling from internal organs were taken from liver, spleen, kidney and heart blood.

Surface swabs were aseptically taken from each fish and inoculated into separate sterile tube containing peptone water (1%). In addition, 5 g of fish sample flesh was desiccated in a sterile flask, under complete sterile condition and then 45 mL of sterile peptone water were added and thoroughly mixed using sterile blender for 1-1.5 min at 3000 rpm.

Bacterial isolates were identified by their cultural morphology and biochemical examination (identification) (Cruickshank *et al*., 1975; Collee *et al*., 1996).

The transmission of infection to cultured fish occurs through direct and indirect exposure to fish pathogens, which is facilitated by poor fish health management. The mechanisms by which fish diseases are generally transmitted including a mixture of the following, contaminated water supply, infected eggs or fish stocks and/or contaminated culture facilities, together with environmental conditions associated with the fish culture practice air, ponds, soil, equipment, feed and pollutants (Abd El Shahid *et al*., 2009; Lotfy *et al*., 2011; Aly, 2013).

Statistical analysis: Statistical analysis was carried-out using Chi-square test to detect the differences of the bacterial incidence among examined fish species according to SAS (2004).

RESULTS

The results recorded in Table 1 revealed significance difference ($p<0.01$) between bacterial isolates and fish species (*O. niloticu* and *Clarias garipinus*), while no significance difference was recorded in *Cyprinus carpio* and *Mugil cephalus*). The positive samples incidence was recorded in *Pseudomonas* spp. and followed by *Streptococci* spp. and *Salmonella* spp. while lower one was recorded by *E. coli* and Staphylococci spp. Regarding the results recorded in Table 1 *Pseudomonas* spp. showed high incidence (20%), while lower one recorded by *E. coli* and *Staphylococci* spp. Moreover, the site of examination revealed high incidence (10%) recorded by *Streptococci* spp. from internal organs while *Pseudomonas* spp. showed high incidence (16%) from internal organs. Meanwhile lower results (1%) were recorded by *Streptococci* spp. from internal organs and *E. coli* spp. from external surface.

Table 1: Incidence of bacterial species isolated from surface and internal organs in different examined fish species (n = 100)

	Fish species																			
	Oreochromis niloticus					*Clarias gariepinus*					*Cyprinus carpio*					*Mugil cephalus*				
	No. of positive	Surface		Internal		No. of positive	Surface		Internal		No. of positive	Surface		Internal		No. of positive	Surface		Internal	
Bacterial isolates	sample	No.	%$	No.	%	sample	No.	%	No.	%	sample	No.	%	No.	%	sample	No.	%	No.	%
Escherichia coli	3	1	1	2	2	6	4	4	2	2	5	3	3	2	2	4	2	2	2	2
Pseudomonas	20	4	4	16	16	10	3	3	7	7	22	7	7	15	15	20	8	8	12	12
Salmonella spp.	11	7	7	4	4	20	16	16	4	4	13	9	9	4	4	11	7	7	4	4
Staphylococci	3	2	2	1	1	12	7	7	5	5	6	4	4	2	2	5	4	4	1	1
Streptococci	15	10	10	5	5	20	15	15	5	5	18	12	12	6	6	15	10	10	5	5
Total	52	24	46	28	54	68	45	66	23	33	64	35	55	29	45	55	31	56	24	44

Chi-square: 10.44**, Examined samples for each species: 100, $: Percentage of total samples

Table 2: Prevalence of bacterial isolates of zoonotic importance among different fish species

Fish species	Staphylococci	*Salmonella* spp.	*E. coli*	Streptococci	*Pseudomonas*	Total	Percentage
Oreochromis niloticus	3.00	11.00	3.00	15	20	52.00	52.00
Clarias gariepinus	12.00	20.00	6.00	20	10	68.00	68.00
Common carp	6.00	13.00	5.00	18	22	64.00	64.00
Mugil cephalus	5.00	11.00	4.00	15	20	55.00	55.00
Total	26.00	55.00	18.00	68	72	239.00	59.75
Percentage	6.50	13.75	4.50	17	18	59.75	

Chi-square: 32.55**, n: 100 samples for each fish species

Furthermore, the high incidence of bacterial isolates reveled from *Clarias garipinus* was *Salmonella* spp. and *Streptococci* spp. (20%), while *E. coli* (6%) recorded lower one. Regarding the site of examination, *Salmonella* spp. (16%) recorded the high incidence from the external surface, while the lower one from the same site was recorded by *Pseudomonas* spp. (3%). Meanwhile, the samples taken from internal organs revealed higher incidence (7%) by *Pseudomonas* spp., while lower one was recorded by *E. coli* spp. (2%).

Regarding the results recorded in Table 1 from *Cyprinus carpio,* the high incidence was reveled by *Pseudomonas* spp. (22%) followed by *Streptococci* spp. (18%) and the lower one was recorded by *Staphylococci* spp. (6%). Concerning the site of examination high incidence (12%) was revealed by *Streptococci* spp. and lower incidence (3%) by *E. coli* from external surface, while samples taken from internal organs reveled high incidence (15%) by *Pseudomonas* spp. and lower one was recorded by *E. coli* spp. and *Staphylococci* spp. (2% of each).

The results recorded in Table 1 concerning *Mugil cephalus* reveled high incidence (20%) by *Pseudomonas* spp. and the lower one was recorded by *E. coli* spp. (4%). Meanwhile, the samples taken from external surface showed higher incidence (10%) by *Streptococci* spp. and lower one by *E. coli* spp. (2%), while, higher incidence from samples taken from internal organs (12%) by *Pseudomonas* spp. while lower one was recorded by *Staphylococci* spp. (1%).

The Chi-square statistical results showed high significant results was indicated by *O. niloticu* and *Clarias garipinus* between site of examination and isolated bacteria, while no significant were recorded between fish species (*Cyprinus carpio* and *Mugil cephalus*) samples and site of examination and isolate bacteria.

The results recorded in Table 2 revealed the incidence of different zoontic bacteria that differ significantly among different fish species under the study. The higher incidence of bacterial isolates observed in *Clarias garipinus* (68%), followed by common carp (64%), *Mugil cephalus* (55%) and the lower bacterial incidences observed in *O. niloticus* (52%). The higher incidence of bacterial isolates observed in *Pseudomonas* (18%), Streptococci (17%), *Salmonella* spp. (13.75%) and the least incidence observed in *E. coli* (4.50%) and Staphylococci (6.50%).

DISCUSSION

Concerning the type of examined fish, it is clear that the higher bacterial incidence was observed in *Clarias garipinus*, common carp*, Mugil cephalus* and the lower bacterial incidence was observed in *O. niloticus*. These variations could be attributed to the fish species, environments and methods of catching and extent of handling during catching (Wang *et al*., 1994). In general, the presence of Enterobacteriaceae indicates unsatisfactory hygienic measures during catching and unsanitary condition of the fish environment (Valdivia-Garvayo *et al*., 1997). *Pseudomonas* spp. can cause septicemia in *Oreochromis niloticus* in Egypt that was more prevalent during winter period (El-Sayyad *et al*., 2010; Shayo *et al*., 2012). On the other hand, Newaj-Fyzul *et al*. (2008) illustrated that high prevalence of bacterial pathogens in tilapia especially *Pseudomonas* spp. (60.0%) and *Staphylococcus* spp. ($p<0.05$, χ^2).

The presence of Enterobacteriaceae in fresh water fishes indicates a microbiological proliferation and multiplication of pathogenic and toxigenic micro-organisms constituting public health hazard. Generally, the presence of coliform in fish as *E. coli* serves as index of sanitation under which the fish is handled and associated with the faecal contamination. Such enteric bacteria were previously isolated with different percentages in freshwater from different fish species including Staphylococci, *Salmonella*, *E. coli*, Streptococci and *Pseudomonas* (Hassan, 1995; Austin and Austin, 2012).

The present results agree with those recorded by Heinitz *et al*. (2000), who reported that, *Coliform* do not seem to represent the normal flora of fish but presence indicate contamination when exposed to polluted water.

From results of the current study it is concluded that *O. niloticus*, showed higher incidence of *Pseudomonas* (4% internal, surface 16%), Streptococci (10% internal, surface 5%) *Salmonella* spp. (7% internal, surface 4%) and lower incidence observed by Staphylococci (2% internal, surface 1%) and *E. coli* (1% internal, surface 2%). The obtained results indicated higher incidence of bacterial isolates recorded in *Clarias garipinus* (68%), followed by common carp (64%) and *Mugil cephalus* (55%) and the lower incidence was in *O. niloticus* (52%). Generally, the higher incidence of bacterial isolates was recorded by, *Pseudomonas* (18%), Streptococci (17%), *Salmonella* spp. (13.75%) and the least incidence by *E. coli* (4.50%) and *Staphylococci* spp. (6.50%).

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