j. Egypt. net. med. Assac 77, no 2. 313 - 336 (2017)

#### **RISK EVALUATION OF SOME FISH AQUACULTURE**

By

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#### ABSTRACT

Fishes are generally regarded as safe, nutritious and beneficial but, aquaculture products have sometimes been associated with certain food safety hazards and the majority of which are usually related to exposed stress. In Egypt, tilapias aquaculture yield represents the most popular cultured species are mainly Oreochromis niloticus. The current study was conducted on tilapias farms at Damietta governorate. Therefore, a total of 50 tilapias fish samples (25 sample from a risky southern area and 25 samples from a non-risky northern area) were random collected from farms in the two local areas for bacterial and chemical risks evaluation. Where, the obtained results showed that incidence of coliforms contamination were 100% and 84% in risky and non-risky area respectively. While, by using conventional culture method a different pathogenic bacterial species were isolated and identified in the flesh muscles with isolation rates 44%, 40% and 60% in risky area. Whereas, the rates were 16%, 8% and 24% in non-risky area for pathogenic E. coli, L. monocytogens and Staph. aureus respectively. Salmonella spp. failed to be isolated from the two areas. Isolates of pathogenic E. coli were subjected to serological identification as O86a, O146, O114, O142 and untypable serovars. The study was conducted to analyze some organochlorine pesticides residues by Agilent gas chromatograph GC. The results revealed rates of 100% heptachlor, 40% dieldrin, 48% DDD, 64% DDT, 16% gamma BHC and 20% delta BHC. Contamination of fish in risky area was exceeding the permissible limits, comparing to the non-risky area where the same residues are detected by lower ratios and within the permissible limits, except for DDD and DDT that was higher than the permissible limit. Organophosphorus pesticides were not detected in all examined samples in the two areas. Cadmium residues were detected in all examined fish flesh samples within the permissible limits, while lead and mercury heavy metal residues were not detected in both risky and non-risky study areas. The public health significance of isolates and chemical pollutants was described.

#### <u>Keywords:</u>

Aquaculture - fish - bacteria - *E. coli* - *Salmonella* spp. - *Staph. aureus* - *L. monocytogenes* - insecticides - heavy metals - risks - GLC - AAS.

#### INTRODUCTION

Fish is considered one of the safest nutritive and highly desirable foodstuffs as fish meat has excellent nutritional value being rich in proteins, vitamins, minerals and unsaturated fatty acids that provide clear health benefits (Emikpe et al., 2011 and Alghabban, 2014). On a global scale, most of the world's population about 56% derives at least 17-20% of its animal protein intake from fish (FAO, 2002 and 2014). Almost one third of fish used for human consumption are produced in aquaculture (ICMSF, 2005; Hastein et al., 2006 and FAO, **2012**). About, 87.1% of the total aquaculture production was from developing countries (FAO, **1997** and **2002**). On a national scale, in a survey of Egyptian aquaculture (Shehadeh and Feidi, 1996) found that the most popular cultured species are tilapias, mainly Oreochromis niloticus, about 35% of total aquaculture production. Tilapias are categorized as the second most widely farmed fish in the world (Adebayo-Tayo et al., 2012). Tilapias are an important aquaculture production for food supply because of their faster growth rate, tolerant to harsh environment and ease culture technique. Tilapias are more suitable for culture in the shallow and small ponds (Suresh and Lin, 1992 and Romana-Eguia and Eguia, 1999). Moreover, aquaculture products have sometimes been associated with certain food safety risks that will differ from region to region, from habitat to habitat and will vary according to the method of production, management practices and environmental conditions where the majority of fish hazards are usually related to exposed stress (WHO, 2007). According to Twarowska et al., (1997), water used for aquaculture is restricted to irrigation drainage water of variable salinity and water drawn from coastal and inland lakes. The largest problem regarding aquaculture in wastewater effluent is the accumulation of bacterial pathogens, pesticides and heavy metals in the fish with the possible public health hazards. The hazards associated with human pathogenic bacteria in the fishes produced in aquaculture can be categorized into two groups: The first contains bacteria (indigenous) naturally originate in the aquatic environment, such as Clostridium botulinum, Vibrio spp., Aeromonas hydrophila and Listeria monocytogenes. While, the second group consists of bacteria (non-indigenous) that contaminate the aquatic environment from human or animal reservoirs or otherwise, introduced to the aquatic environment, such as Staphylococcus aureus, Salmonella spp., Shigella spp. and faecal

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coliforms such as Escherichia coli (FAO, 1994; Jay, 2000; Jayasinghe and Rajakaruna, 2005 and Lyhs, 2009). In studies of seafood borne pathogens, some of the common major pathogens have emerged as being of significant importance in terms of human health and diseases; these include Listeria monocytogenes, Vibrio parahaemolyticus, Staphylococcus aureus, Salmonella spp., Yersinia enterocolitica, Campylobacter jejuni and faecal coliforms such as *Escherichia coli*, are widely recognized as the principle causes of food poisoning outbreaks occurring due to consumption of contaminated fish (Venugopal et al., 1999; Feldhusen 2000; Acha and Szyfres, 2001; Hassan and Fatin, 2003; Novotny et al., 2004 and Gauthier, 2014). Pesticides compounds include organophosphorus, organochlorine, carbamate and pyrethroid derivatives used to destroy different pests. The hazardous chemicals in fish may be traced to the direct uptake of compounds present in the water and in fish diet that may bioaccumulate (Arnot and Gobas, 2004). Therefore, fish absorb these compounds directly by water or by ingesting contaminated food. In particular, organochlorine insecticides are highly stable under different environmental conditions with long persistent nature leading to chronic adverse effects on humans (Monirith et al., 2000). Generally, chemicals used at farm level can be accumulated in fish and could cause chronic health risks to fish consumers. They have the potential to gradually accumulate in the human body causing certain organ or system malfunction, namely, cancer, nerve problems and immunological problems (Garrett et al., 1997; Reilly and Kaeferstein, 1999 and Hans, et al., 2000). Toxic heavy metals have received considerable attention due to their risks, considered among the most serious contaminants of aquatic ecosystems due to their accumulation in biota (Dural et al., 2006) and biomagnification in the food chain (Mansour and Sidky, 2002; Erdogrul and Ates, 2006; Erdogrul and Erbilir, 2007). The main sources of heavy metal pollution are the sewage disposal, agriculture drainage water containing pesticides and fertilizers as well as the industrial effluents (Unger and Newman, 2002; Santos et al., 2005; Singh et al., 2007). Moreover, among the heavy metals that can be toxic under certain conditions are lead, mercury and cadmium have a cumulative effect causing serious health risks to humans, especially to young children as carcinogenic and mutagenic effects as well as endocrine disruption (Twarowska et al., 1997; Kalay and Canli, 2000 and Brar, 2009). Unpleasant, fish and fishery products cause up to 30% of the food borne illnesses in developed countries (Nilsson and Gram, 2002 and Valdimarsson et al., 2003). One of the essential things in food hygiene is the examination of food, especially for the presence of hazards. This is very

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much needed for the protection and maintenance of community health against the possible risks (Sujatha *et al.*, 2011). Finally, risk evaluation of aquaculture fish is largely reflected the condition and safety of aquatic environments in which they were cultured (Erondu and Anyanwu, 2005 and Ekpo *et al.*, 2010). The current study was therefore, planned for the risk evaluation of some aquaculture fish to some bacterial pathogens, pesticides and heavy metals in two local different areas: risky area (southern area/Shatta village) and non-risky area (northern area) at Damietta governorate and compared to the obtained results between the two areas also, and to declare potential public health significance.

### **MATERIAL AND METHODS**

### I.Study area:

The study was conducted on two areas, the first one is Shatta village a fishermen village located in the northern east direction to Damietta governorate; it is characterized by many aquaculture fish farms. The main growing water supply is an extension of the water of Lake Manzala, which lies between some governorates in eastern delta, Egypt. It is considers a risky area and more vulnerable exposure to constant pollution from different sources, notably the most dangerous one is the untreated domestic human sewage wastes; in addition to the agricultural (pesticide residues and organic fertilizers) and industrial pollution. Moreover, the presence of garbage recycling factory at neighboring of this area whereas smoke spreads in the environments as a result of garbage burning. Conversely, the second non-risky as a comparison area is located in the northern to the first risky area, where all environmental factors are nearly normal.

### II.<u>Sampling:</u>

A total of 50 samples of aquaculture fresh tilapias spp., were obtained from aquaculture farms, located in two different local areas (25 each of risky area (southern area/Shatta village) and non-risky area (northern area)) were randomly collected from each farm at Damietta governorate and packed immediately in well labeled, clean, polyethylene bags and kept in a cooler and then taken immediately to the Damietta sea port-food inspection laboratory for bacteriological analysis. Analysis of heavy metal residues were conducted by central Lab of residue analysis of pesticides and heavy metals in food (ARC). Analysis of pesticide residues and serological confirmation of isolates were performed in (AHRI).

**Bacteriological Analysis:** 

1.Coliforms count (MPN/g) (FDA, 2002).

2. Isolation of Escherichia coli (FDA, 2002).

3. Isolation of *Salmonella* species (ISO 6579: 2002).

4. Isolation of *Listeria monocytogenes* (FDA, 2011).

5. Isolation of *Staphylococcus aurues* (FDA, 2001).

IV. Analysis of pesticide residues (Mills et al., 1972 and Mosaad et al., 2008).

V. Analysis of heavy metal residues (ICP-OES, 2003).

## Statistical analysis:

Statistical analysis was performed using SPSS statistical program for windows (Version 16), (SPSS Inc., Chicago, IL, and USA). Data were expressed as minimum, maximum and mean  $\pm$  SD, while P values by the ANOVA-test.

## RESULTS

 Table (1): Prevalence of coliforms count (MPN/g) in examined tilapia aquaculture fish flesh (n=50).

Sampling area	Number of examined	Positive samples		Coliforms count (MPN/g)			
	samples	No.	%	Min.	Max.	Mean±SD	
Risky area (southern area/Shatta village)	25 (Marketing sizes)	25	100	15	240	54.400±57.01023	
Non-risky area (northern area)	25 (Marketing sizes)	21	84	3	75	22.392±21.64628	

 Table (2): Prevalence of identified bacterial isolates of examined aquaculture tilapia fish flesh (n=50).

Bacterial isolates	(southern a	y area area/Shatta age)	Non-risky area (northern area) Positive samples		
	Positive	samples			
Γ	No.	%	No.	%	
Pathogenic <i>E. coli</i>	11	44	4	16	
Salmonella spp.	-	-	-	-	
Listeria monocytogens	10	40	2	8	
Staphylococcus aureus	15	60	6	24	

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 Table (3): Serotyping of pathogenic *Escherichia coli* isolated from the examined aquaculture tilapia fish flesh samples (n=50).

Bacterial isolates Samples	No. of	Positive		serogroup EC)	Noof	0/ -6
	samples of pathogenic <i>Escherichia coli</i>	Poly- valent sera	Mono- valent sera	No. of serovars	% of serovars	
		50 15	1	<b>O8</b> 6a	3	20
			2	O146	3	20
<i>E. coli</i> 50	50		3	0114	3	20
			5	0142	3	20
			Unty	pable	3	20

**Table (4):** Prevalence of organochlorine pesticides (OCs) residues in examined aquaculturetilapia fish flesh of the risky area (southern area/Shatta village) (n=25).

Organochlorine pesticides (OCs)	Permissible limits		Positive samples		<b>Concentrations (ppm)</b>			
	µg/kg	ppm	No.	%	Min.	Max.	Mean±SD	
Heptachlor	<b>200</b> <sup>1</sup>	0.2	25	100	0.12	0.80	0.189±0.035	
Dieldrin	<b>300</b> <sup>1</sup>	0.3	10	40	0.12	0.90	0.550±0.262	
DDD	100 <sup>2</sup>	0.1	12	48	0.50	0.88	0.709±0.121	
DDT	100 <sup>2</sup>	0.1	16	64	0.10	0.19	0.160±0.027	
Gamma BHC	200 <sup>2</sup>	0.2	4	16	0.47	0.88	0.620±0.195	
Delta BHC	<b>200</b> <sup>2</sup>	0.2	5	20	0.22	0.30	0.274±0.032	

<sup>1</sup>(CAC, 2009) - <sup>2</sup>(EU, 2011)

**Table (5):** Prevalence of organochlorine pesticides (OCs) residues in examined aquaculturetilapia fish flesh of the non-risky area (northern area) (n=25).

Organochlorine pesticides (OCs)	Permissible limits		Positive samples		Concentrations (ppm)			
pesticides (OCs)	µg/kg	ppm	No.	%	Min.	Max.	Mean±SD	
Heptachlor	<b>200</b> <sup>1</sup>	0.2	18	72	0.15	0.25	0.20±0.260	
Dieldrin	<b>300</b> <sup>1</sup>	0.3	7	28	0.24	0.35	0.29±0.038	
DDD	100 <sup>2</sup>	0.1	6	24	0.66	0.94	0.80±0.103	
DDT	100 <sup>2</sup>	0.1	10	40	0.66	0.93	0.81±0.098	
Gamma BHC	200 <sup>2</sup>	0.2	3	12	0.17	0.47	0.22±0.084	
Delta BHC	200 <sup>2</sup>	0.2	-	-	-	-	-	

<sup>1</sup>(CAC, 2009) - <sup>2</sup>(EU, 2011).

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Table (6): Prevalence of heavy metal residues in examined aquaculture tilapia fish flesh of the risky area (southern area/Shatta village) and non-risky area (northern area) (n=25 each).

Heavy metal	Permissible limits <sup>3</sup>	Area of examination	samples		Concentrations (mg/kg - ppm)			
residues	(mg/kg - ppm)	examination	No.	%	Min.	Max.	Mean±SD	
Lead (Pb)	0.3	risky area non-risky	-	-	-	-	-	
Mercury (Hg)	0.3	risky area non-risky	-	-	-	-	_	
*Cadmium	2.0	risky area	25	100	0.0161	0.0262	0.0210± 0.005**	
(Cd)		non-risky	25	100	0.0288	0.0720	0.0518± 0.0217**	

#### <sup>3</sup>(CAC, 2012)

\*Marine bivalve molluscus and cephalopods

\*\* Similar letter means non-significance difference at (P>0.05)

#### DISCUSSION

In available literatures there is no data similar with the obtained current findings of the comparison between aquacultures under risk and other under normal conditions at the same area. But, nearly all researches deal with retail fish and fish products or aquaculture fish-processing environments as sediment of fish farming ponds and growing water supplies. As well as, investigation of the pathogens in gills, skin, mouth and viscera. Recorded data in (Table 1) indicated that out of 50 aquaculture tilapia fishes that evenly of marketing sizes using conventional culture coliforms count (MPN/g) microbiological method. 25 samples (100%) were positive for coliform with a count ranged from 15 to 240 with a mean value was 54.400 MPN/g of risky area (southern area/Shatta village). While, 21 samples (84%) were positive for coliform with a count ranged from 3 to 75 with a mean value was 22.392 MPN/g of non-risky area (northern area) respectively. As far as, a very few available literatures dealing with the coliforms count and pathogenic bacteria detected in fish flesh reared in aquaculture. Nearly similar finding was declared by (Olugbojo and Ayoola, 2015). While,

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higher results were investigated by (Gabr and El Alfy, 2009 and Mandal et al., 2009), who study the abundance of coliforms bacteria in tilapia sampled from different sources where, significantly higher density of coliform was detected in the muscles of tilapia sampled from fish ponds. Initially, it has been well known that both fresh and brackish water fishes can harbor human pathogenic bacteria, particularly the coliform group (Ramos and Lyon, 2002). The presence of coliforms in fish flesh demonstrates the level of biological pollution of their environment because coliforms are not the normal flora of bacteria in fish, where there has been coliform contamination from warm blooded animals; their presence is used as bioindication that other pathogenic organisms in the environment (Chao et al., 2003). Unfortunately, the presence of different bacteria species including human pathogenic bacteria in fish can be linked to direct contact with a contaminated water environment and ingestion of bacteria from sediments or contaminated feed. Thus, bacteria detected in fish reflect the condition and safety of aquatic environments (Hastein et al., 2006 and WHO, 2007). Aquaculture products can harbor pathogenic bacteria which are part of the natural microflora of the environment. As shown in (Table 2), out of 50 aquaculture tilapia fishes using conventional culture microbiological methods. 25 samples of the risky area (southern area/Shatta village) revealed that different species of bacteria were isolated and identified in the flesh with isolation rate was 44% pathogenic Escherichia coli, 40% Listeria monocytogens and 60% Staphylococcus aureus. While, 25 samples of non-risky area (northern area) the isolation rate was 16% pathogenic Escherichia coli, 8% Listeria monocytogens and 24% Staphylococcus aureus. While, Salmonella spp. failed to isolate in all inspected samples. Referring to the serotyping results in (Table 3), revealed that 15, pathogenic *E. coli* isolates from aquaculture tilapias flesh positive samples were serologically identified as serogroup enteropathogenic (EPEC), polyvalent 1 monovalent O86a serovars 3 (20%), polyvalent 2 monovalent O146 serovars 3 (20%), polyvalent 3 monovalent O114 serovars 3 (20%), polyvalent 3 monovalent O142 serovars 3 (20%) and untypable serovars 3 (20%). While, emerging pathogen E. coli O157:H7 were absent in all the analyzed samples. For pathogenic *Escherichia coli*, lower findings were reported by (Elsherief et al., 2014), that detect (8%) of E. coli in tilapias farm fish which serotyped as O111:H4 (EHEC) and O125:H21 (ETEC). On contrary, Hassan et al., (2012) failed to isolate E. coli from muscle of O. niloticus. On the other hand, the detection of pathogen E. coli O157:H7 in agreement finding were declared by (Surendraraj et. al., 2009; Barbosa et. al., 2014) in failure of E. coli O157:H7 detection. On contrary, positive result of

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E. coli O157:H7 was recorded by (Soliman et. al., 2010). From the public health point of view, risks of food borne pathogens have increased during the last 20 years (USGAO, 1996). Thus, along with nutritional benefits from the consumption of fish, the potential risk to human health occurs. Escherichia coli are often used as an indicator for fecal contamination. Some strains of E. coli are capable of causing food borne disease, ranging from mild enteritis to serious illness and even death (WHO, 1999). It is one of the most significant food borne pathogens that have gained increased attention in recent years to be responsible for a wide range of illnesses in humans, including hemorrhagic colitis and hemolytic uremic syndrome (Nakanishi et al., 2009; Jasson et al., 2010 and Mathusa et al., 2010). Although, most E. coli strains are non-pathogenic, some of them are highly pathogenic (Kuhnert, 2000). Enteropathogenic *E. coli* (EPEC) had been recognized by the (WHO, 1999), is the main cause of diarrhea among children in developing countries (Dean et al., 2005). It is generally transmitted by contaminated food and water then colonizes the small intestine, where it becomes attached to epithelial cells and produces typical lesions named "attaching and effacing" (A/E) (Kaper et al., 2004). Also, for Salmonella spp. higher findings were mentioned by (Elsherief et al., 2014 and Olugbojo and Ayoola, 2015), that identified S. typhimurium and S. enteritidis strains. In essence, the relationship between fish and Salmonella has been described by several scientists; some believe that fish are possible carriers of Salmonella which are harbored in their intestines for relatively short periods of time and some believe that fish get actively infected by Salmonella. The organism was never recovered from the flesh of the fish but, was isolated from viscera and epithelium (Pullela, **1997**). Aquaculture products can become contaminated with *Salmonella* through the use of water, unsanitary ice, containers and poor hygienic handling practices (FAO, 2010). Whereas, for Listeria monocytogens lower prevalence of L. monocytogenes (3.3%) was seen in fresh O. niloticus purchased from fish farm in Beheira governorate by (Edris et al., 2014). L. monocytogenes, an invasive food borne pathogen capable of causing a serious disease called listeriosis (Barton et al., 2011). Listeriosis is a life-threatening disease, despite its low incidence, the mortality rate may reach 20-40% in those who are susceptible, including fetuses, newborns, immunocompromised individuals also, are more vulnerable (as those with AIDS, diabetes mellitus, cancer, organ transplant recipients, chronic diseases or who are on certain immune-suppressing arthritis drugs or cancer chemotherapy), elderly and pregnant women (Farber and Peterkin, 1991; Schuchat et al., 1991 and Lianou and Sofos, 2007).

Furthermore, for *Staphylococcus aureus* nearly similar result was confirmed by (Olugbojo and Ayoola, 2015). S. aureus is one of the most important etiological agent predominantly associated with sporadic staphylococcal food poisoning around the world (Balaban and Rasooly, 2000; Le-Loir et al., 2003 and EFSA and ECDC, 2010), and in many countries, it is the main bacterial agent causing food borne diseases. S. aureus is a versatile human pathogen capable of causing staphylococcal food poisoning, toxic shock syndrome, pneumonia, postoperative wound infection and nosocomial bacteremia. Patients who are highly susceptible to infection, including those with a low neutrophil count, should be supplied with a low microbial diet including transplant recipients, people with cancer, human immunodeficiency, virus/acquired immunodeficiency syndrome, diabetes, older and pregnant women (Lund, 2014). Concerning to the results in (Table 4), could represent the mean concentrations of some organochlorine pesticides (OCs) residues in examined aquaculture tilapias flesh samples of the risky area (southern area/Shatta village), with 0.189±0.035 ppm for heptachlor by 100%,  $0.550\pm0.262$  ppm for dieldrin by 40%,  $0.709\pm0.121$  ppm for DDD by 48%, 0.160±0.027 ppm for DDT by 64%, 0.160±0.027 ppm for gamma BHC by 16% and  $0.274\pm0.032$  ppm for delta BHC by 20%. In comparison, with the maximum residue limits (MRL) of these pesticides, all these values were exceeded the permissible limits have been established by (CAC, 2009) and (EU, 2011). Likewise (Table 5), could characterize the mean concentrations of some organochlorine pesticides (OCs) residues in examined aquaculture tilapias flesh samples of the non-risky area (northern area), with 0.20±0.260 ppm for heptachlor by 72%, 0.29±0.038 ppm for dieldrin by 28%, 0.80±0.103 ppm for DDD by 24%, 0.81±0.098 ppm for DDT by 40%, 0.22±0.084 ppm for gamma BHC by 12% and delta BHC (benzene hexachloride or hexachlorobenzene /HCB) not detected in all fish flesh samples of the non-risky area. In comparison, with the maximum residue limits (MRL) of these pesticides, these values were not exceeded but below the high end of the maximum allowable limits have been established by (CAC, 2009) and (EU, 2011) except for DDD (dichlorodiphenyl-dichloroethane) and DDT (dichloro-diphenyl-trichloroethane) that markedly higher than the permissible limits have been recognized by (EU, 2011), which may be due to a previous or an illegal use. Thought, previous studies have detected organochlorine pesticides (OCPs) in aquaculture tilapias flesh with variable findings. Some investigators have obtained nearly similar around the current detected findings, (Easton et al., 2002; Bagumire et al., 2008; van Leeuwen et al., 2009; Botaro et al., 2011; Yahia and Elsharkawy, 2013 and

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Masci et al., 2014). Whereas, organophosphorus pesticides (OPs), could not be detected in all examined tilapias aquaculture flesh samples in the two areas of the current study. Understandably, OPs are degraded rapidly by hydrolysis on exposure to sunlight, air and soil although; slight amounts can be detected in some food and drinking water. Their ability of getting degraded easily has made them an attractive alternative to the persistent organochloride pesticides. Most, OPs are degraded by microorganisms in the environment as a source of phosphorus and/or carbon. Thus, the OP pesticides can be hydrolyzed and detoxified by carboxylesterase and phosphodiesterase enzymes (Ghosh et al., 2010). Pesticides cause untoward effects on man in two ways. Firstly, they have direct effects on the health of persons who use them and secondly, their remnants accumulate in foodstuffs which also produce side effects on consumer (Yazgan and Tanik, 2010). The side effects include short term illness like abdominal cramps, vertigo, headaches, diplopia, nausea, ocular disturbances and dermatopathies. While, long term adverse effects include increased possibility of respiratory failures, depression, nervous defects, prostate cancer, leukemia and infertility. These effects are considered one of the major health risks all over the world (Ghasemi and Karam, 2009 and Yazgan and Tanik, 2010). Pesticides may increase the likelihood of Parkinson disease (Hoek and Dawson, 2007 and Bradman et al., 2011). Appropriate poisoning in many countries, especially in developing countries is considered the second causes of mortalities after infectious diseases (Farshad, 2000). OCPs are frequently detected in human lipid tissues and breast milk (Hernández et al., 2002). From the results obtained in (Tables 6), it is obvious that neither lead (Pb) nor mercury (Hg) present in the fish flesh of both risky area and non-risky. On the other hand, the cadmium was recorded in this study in both fish flesh of risky area and non-risky. The minimum concentration of cadmium (Cd) in fish muscles of the risky area was 0.0161 mg/kg (ppm) and the maximum value was 0.0262 mg/kg (ppm) with a mean value  $0.0210\pm0.005 \text{ mg/kg}$  (ppm). While, the mean value of cadmium (Cd) residue concentration in fish muscles of non-risky area was 0.0518±0.0217 mg/kg (ppm) with a minimum value 0.0288 mg/kg (ppm) and maximum value 0.0720 mg/kg (ppm). The cadmium mean concentration was below the level recommended by (CAC, 2012), 2.0 mg/kg (ppm) this level was recommended for cadmium residues in marine bivalve molluscus and cephalopods, the fish was not included the codex. The permissible limit recommended by (CAC, 2012) for both lead (Pb) and mercury (Hg) as methyl was 0.3 mg/kg (ppm) for each heavy metal. Nearly similar results were recorded for cadmium in tilapias and

other fish species; the range of cadmium (Cd) concentration was 0.0466-0.0686 ppm. On contrary, the same study reported lead (Pb) residue range 0.06-0.44 ppm and total mercury (Hg) range 0.075 - 0.151 ppm for tilapias and other species in Egyptian market (Marzouk et al., 2016). Whereas, the concentration of heavy metals in the fish muscles of fishes collected from the different regions in this study were within the permissible levels are from the heavy metals point of view safe for human consumption and public health, this results agreed with that obtained by (Marcussen et al., 2007; Bagumire et al., 2008; Taweel et al., 2011; Botaro et al., 2012 and Olusola et al., 2012). While, higher finding was declared by (Saeed and Shaker, 2008), who mentioned that the edible part of Nile tilapia (O. niloticus) samples; showed higher levels of Cd in (Lake Edku and Manzala) and Pb in (Lake Manzala). The statistical analysis proved that there was non-significant difference at (P>0.05) between the concentration of cadmium residues level in the fish muscles of both risky and non-risky area in Shatta village aquaculture. Heavy metals in aquatic environment are transferred through the food chain into humans. It is well known that the edible portion fish muscles are not an active tissue to accumulate heavy metals but, it was detected in muscles of some fish in contaminated regions exceeded the permissible levels (Uysal et al., 2008). Heavy metals under certain conditions have a cumulative effect causing deleterious health risks to humans through food chain, high level of mercury (Hg) can damage kidneys, CNS that can cause memory loss, slurred speech, hear loss, coordination lack, loss of sensation in fingers and toes, reproductive problems, coma and possible deaths. In addition, developing fetus brain is more sensitively affected (Vannoort and Thompson, 2006). Long term or high dose exposure to cadmium (Cd) can lead to kidney failure and bone softening and prostate cancer (Gray et al., **2005 and Vannoort and Thompson, 2006)**. Whereas, lead (Pb) can built up in the body and affects the central nervous system and kidneys. Children and babies are particularly at risk from damage to their CNS, which can cause learning difficulties and behavioral changes (WHO, 2000). Finally, the current study revealed that the contamination of aquaculture fish with some hazards may give an indication about bad sanitary conditions under which fish were exposed resulting in public health risks

#### .CONCLUSION

Results of the current study were showed the comparative difference between the two study areas where, the risky southern area fish aquacultures were under major hazards have the

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potential of affecting human beings to the presence of bacterial, opportunistic pathogens, chemical pollutants indicate contamination from different sources and may cause food borne diseases to susceptible individuals. In addition, a general trend from the results declares an increase of some organochlorine pesticides in this area above the MRLs indicating a recent abuse in this investigated area. Almost, these pesticides and their derivatives have been banned in most parts of the world. However, the results of the study have shown that they are still being used in our countries. Also, heavy metal residues detected in similarity for cadmium pollution; while, lead and mercury residues were not detected in both risky and nonrisky study areas. On the contrary, the non-risky northern area was acceptable and in save side for hazards presence; that findings of this area were to some extent similar with the risky area but with low incidence or may be non-detectable and within the permissible limits. Unfortunately, exception for DDD and DDT was found in higher level above the permissible limits which may be due to an illegal uses. Considerations must be submitted to aquaculture stakeholders should work in union to provide guideline and policies that would promote an environmentally friendly and supportable industry. In conclusion, further investigations of farmed fish especially for the presence of hazards are necessary for protection and maintenance of the community health from possible risks by keeping food borne hazards to a minimum.

### RECOMMENDATION

Hazards in aquatic environment are difficult but not impossible to be controlled by many available preventive measures. It is now generally recognized that it is impossible to fully guarantee the safety of food but, national and international monitoring programs exist to ensure that, the levels present are acceptable (Hastein *et al.*, 2006). Although, it is possible to reduce the probability of accidents happening by implementing a well-designed, HACCP program based principles at all stages of the food business intended to improve food safety and for protection of the consumer's health (NACMCF, 1992; FAO, 1994 and Reilly and Kaeferstein, 1997). All measures must be taken for preventing or decreasing biological and chemical risks in animal originated food. Thus, application of risk analysis to the aquaculture sector, which produces nearly half the fish that is consumed worldwide (FAO, 2014), has become very important. Guidelines for performing risk analysis have been brought out by the Codex Alimentarius Commission or Codex Committee on Fish and Fishery Products (Karunasagar, 2008). Guidelines should be provided by relevant stakeholders on how to

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achieve a basic level of environmental protection within the vicinity of aqua farms. It is recommended and suggested that, the whole food chain needs to be restructured and stakeholder should work together in controlling the possible hazards. Farmed fish should be under periodic health inspection to minimize the human health risks. The water supply that is suspected of having any biological or chemical contamination should not be used for aquaculture or should be treated well before it is used. Food contamination monitoring is an essential component of assuring the safety of food supplies and managing health risks at the national level. Population should also be educated on the proper measures to be introduced in sewage and wastes disposal and not to the aquatic environments, as this would pollute the water and cause health risks. Also, veterinarians should be pushed to enforce the legislation on all food businesses and understand the importance of doing so. One of the essential things in food hygiene is the examination of food, especially for the presence of hazards. This is very much needed for the protection and maintenance of community health.

#### ACKNOWLEDGEMENT

The authors would like to express their sincere thanks and gratitude to all of Dr. Randa G. Abo-Samra; Dr. Rania L. Salem; Dr. Samah A. Aboushehatta; Dr. Mohammed R. El-Bediwi and Dr. Nahla H. Khalil, the researches members of Food Hygiene Department-Damietta Sea' Port Food Inspection Lab., AHRI, ARC, Egypt for their appreciated kind help and dedicated assistance in carrying out to complete the practical bacteriological part of this research.

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#### الملخص

الأسماك مصدر ممتاز للبروتين عالى الجودة, ومع ذلك فهي عرضة للملوثات البيئية المختلفة, وتلاحظ في العقود الأخيرة انتشار مزارع الأحياء المائية لإنتاج الأسماك, وفي مصر تمثل مزارع إنتاج أسماك البلطي حوالي 35٪ من إجمالي إنتاج المزارع السمكية للأنواع الأخرى, وقد أجريت الدراسة على مزارع أسماك البلطي في محافظة دمياط, حيث تم فحص عدد 50 عينة (25 عينة من المنطقة المعرضة للمخاطر وهي الجنوبية/قرية شطا) و (25 عينة من المنطقة الغير معرضة للمخاطر وهي الشمالية) جمعت بطريقة عشوائية من المزارع بالمنطقتين, وذلك لتقييم المخاطر البكتيرية والكيميائية المختلفة. حيث أظهرت النتائج التي تم الحصول عليها أن نسبة تلوث عضلات الأسماك بميكروبات المجموعة القولونية كانت 100٪ و84٪ في المنطقتين المعرضة للمخاطر وغير المعرضة للمخاطر على التوالي. وذلك باستخدام طريقة الزرع البكتيريولوجي التقليدية المرجعية, كما تم عزل أنواع مختلفة من البكتيريا المسببة للأمراض من عضلات الأسماك وذلك بمعدلات عزل 44٪ و40٪ و60٪ في المنطقة المحفوفة بالمخاطر. في حين كانت 16٪ و8٪ و24٪ في المنطقة الغير محفوفة بالمخاطر وهى على التوالى الإي كولاي المُمرضة والليستيريا مونوسيتوجينس والمكور العنقودي الذهبي الاستافيلوكوكس أوريس، في حين لم يتم الكشف عن وجود بكتيريا السالمونيلا في جميع العينات التي تم فحصها من المنطقتين, وبالكشف السيريولوجي لمعزولات بكتيريا الإي كولاي أكد تواجد عترات متنوعة لبكتيريا الإي كولاي O86a، 0146، 0114، O142 وسلالات untypable, وبتحليل بعض بقايا المبيدات الكلورينية العضوية (OCPs) تواجدت بمعدلات تلوث 100٪ heptachlor و40٪ dieldrin و48٪ DDD و64٪ DDT و16 و26٪ gamma BHC و20٪ delta BHC في المنطقة المعرضة للمخاطر بأعلى من الحدود المسموح بها، وبالمقارنة بالمنطقة الغير معرضة للمخاطر تواجدت نفس البقايا وبنسب أقل وفي الحدود المسموح بها باستثناء DDD وDDT كانتا أعلى من الحد المسموح به والتي قد تكون نتيجة لاستخدامات غير مشروعة, في حين لم يتم الكشف عن المبيدات الفسفورية العضوية (OPs) في جميع العينات التي تم فحصها من المنطقتين, كما تم الكشف عن وجود بقايا الكادميوم في عضلات جميع عينات السمك في الحد المسموح به، في حين لم يتم الكشف عن بقايا المعادن الثقيلة من الرصاص والزئبق في كل من منطقتي الدر اسة المعرضة للمخاطر والغير معرضة للمخاطر, وقد تم مناقشة النتائج والأهمية الصحية مع وضع التوصيات اللازمة. الكلمات الدالة:

الأسماك - مزارع الأسماك - البكتيريا - الإيكولاي - السالمونيلا - الميكروب العنقودي الذهبي - الليستيريا مونوسيتوجين -المبيدات الحشرية الكلورينية والفوسفورية - المعادن الثقيلة.

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