Microbiological evaluation of some frozen and salted fish products in Egyptian markets

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ABSTRACT

Sixty random samples of fish and fish products (frozen fish: Tilapia nilotica and imported basa fillet – salted fish: *Mugil cephalus* and sardine) (15 of each) were collected from different supermarkets in Egypt. All collected samples were microbiologically examined for determination of Aerobic Plate Count (APC), coliforms, *Escherichia coli*, Mould & yeast and *Staphylococcal aureus* counts (log_{10} cfu/g). Isolation and identification of *salmonella*, *liesteria monocytogenes* and *Vibrio parahaemoliticus.* The bacteriological examination revealed that the mean values of APC in the examined fish samples were 4.80± 0.16, 4.63±0.18, 2.35±0.18 and 2.70±0.13 (log_{10} cfu/g) for frozen Tilapia nilotica, Basa salted *Mugil cephalus* and Sardine respectively, 1.48± 0.22, 2.19±0.19, 2.10±0.16 and 2.52±0.11 average coliform counts in examined samples respectively, and *E.coli* count with an average of 2.52±0.14, 2.01±0.17, 1.31±0.11and 1.64±0.14. Mould & yeast average counts were 0.93±0.18, 1.22±0.16, 1.67±0.021 and 1.69±0.18 in examined fish samples, respectively, and the *Staphylococcal* counts were 1.30±0.17, 1.58±0.26, 2.12±0.14, 1.46±0.23 and 2.17±0.17 in examined fish samples, respectively. The incidence of food poisoning organisms (*Salmonella* & *Listeria monocytogenes* and *Vibrio parahaemolyticus* also was investigated and one of both *Listeria monocytogenes* and *Vibrio parahaemolyticus* were isolated in frozen Basa fillet samples with a percentage of 33% and 33% from the examined samples.

Keywords: frozen fish: Tilapia nilotica and Basa ; salted fish: *Mugil cephalus* and Sardine.

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1. INTRODUCTION

Fish and fish products are in the forefront of food safety and quality improvement because they are among the most internationally traded food commodities (Gonçalves & Blaha, 2010). It is an important source of high quality proteins for humans (Tidwell and Allan, 2001). There are increasing in the demand of such good quality protein over the last decade worldwide (Nielson *et al*., 1994). The most serious seafood safety issues resulting in potentially contaminated products are associated with microbial and especially bacterial pathogens. Fish quality is a complex concept involving a whole range of factors which for the consumer include for example: safety, nutritional quality, availability, convenience and integrity, freshness, eating quality and the obvious physical attributes of the species, size and product type (Abbas *et al*., 2008).

Freezing is the most widely used preservation method for fish and it accounted for 49.8% share of total processed fish for human consumption and 20.5% of total fish production in 2008 (FAO, 2010).

The salting process is considered as one of the oldest method of fish preservation and this process is still being used in several places around the world. Salted fish products are popular in many countries and it has proved to be safe for the millenniums even in developed countries (Basti, et al. 2006; Turan, et al. 2007).

Feseikh a traditional Egyptian salted fish has been considered as a popular part of the Egyptian diet especially in certain celebration times as spring day. In addition, Sodium chloride is a flavor enhancer as a consequence of its effect on different biochemical mechanisms by reducing or enhancing the enzymatic activity of some enzymes responsible for the development of different organoleptic parameters (Albarracin et al., 2011). However, the current demand for salted fish is driven more by the flavor of the product than for preservation purposes (Ali, Mariyam, 2012).

In studies of sea food borne pathogens, four major pathogens have emerged as being of significant importance in terms of human health and disease. These include *Listeria monocytogenes*, *Vibrio parahaemolyticus*, *Staphylococcus aureus*, and *Salmonella spp*. (Feldhusen, 2000).

L. monocytogenes has been isolated from fish and seafood products all over the world. The contamination rate of seafood products with *L. monocytogenes* can vary from 0% to more than 50% (Ben Embarek, 1994; Jinneman et al. 1999). Contamination of seafood products with L. monocytogenes depends on many factors such as cleaning and processing procedures, working habits, and the existence of surface persistent L. monocytogenes types in processing facilities. Moreover, raw materials contaminated with Listeria may also be a reason for the contamination of the final product (Rørvik et al. 1997; Miettinen and Wirtanen, 2005).

V. parahaemolyticus is a human pathogen that occurs naturally in the marine environments and is frequently isolated from a variety of seafood including fish, shrimp, crab, lobster, scallop, and oyster (Austin, 2010). This pathogen is a common cause of foodborne illnesses in many Asian countries, including Taiwan, China, and Japan, and is recognized as the leading cause of human gastroenteritis associated with seafood consumption in the United States Jaksic et al. (2002); Su and Liu, (2007).

Therefore, the present study was carried out to investigate the microbiological quality of the frozen fishes (Tilapia nilotica and Basa fillet) and marketed salted fish (*Mugil cephalus*, Sardine) for raising food safety concern

2. Materials and methods

2.1. Collection of samples:

A total of 60 random samples of fish frozen fish: Tilapia nilotica and Basa fillet – salted fish: *Mugil cephalus* and sardine) (15 of each) were collected from different supermarkets in Giza governorate, Egypt. Each sample was kept in a separated sterile plastic bag and preserved in an ice box then transferred to the laboratory under complete aseptic conditions without undue delay for examination. The collected samples were subjected to the microbiological examinations to evaluate their safety and fitness for human consumption.

2.2. Preparation of samples (APHA (2001).

Ten grams from each sample were weighed and stomached with 90ml of 0.1% sterile buffered peptone water using stomacher (Seward stomacher 80 Biomasters, serial No 46464, England to provide a dilution of 10^{-1}. The homogenate was then allowed to stand for 15 minutes at room temperature. From the original suspension, one ml was transferred aseptically with sterile pipette into a test tube containing 9 ml of sterile buffered peptone water 0.1% and mixed well to produce a dilution of 10^{-2} from which further decimal serial dilutions were prepared.

2.3. Determination of Aerobic plate count APHA, 2001.

2.4. Enumeration of Coliform bacteria & Escherichia coli (FDA, 2002).

2.5. Total Mould and Yeast Count (ISO 21527, 2008).

2.6. Isolation and Enumeration of Staphylococcus aureus (FDA, 2001)

2.7. Detection and Enumeration of Listeria onocytogenes (FDA, 2011)

2.8. Isolation and identification of V. parahaemolyticus: According to (ICMSF, 1996).

3. RESULTS

It is evident from the result recorded in table (1) that APC in the examined samples varied from 3.79 to 5.99 with an average value of 4.80±0.16 log cfu/g, 3.40 to 6.15 with an average value of 4.63±0.18 log cfu/g, 0.90 to 2.92 with an average value of 2.35±0.18 log cfu/g and 1.70 to 3.48 with an average of 2.70±0.13 log cfu/g for the examined samples of frozen Tilapia nilotica, Basa, salted Mugil and Sardine, respectively. Table (2) showed that the mean values of Coliform and *E.coli* count (log cfu/g) of examined samples of frozen Tilapia nilotica, Basa, salted Mugil and Sardine were 1.48±0.22 and 2.52±0.14, 2.19±0.19 and 2.01±0.17, 2.10±0.16 and 1.31±0.11 and 2.52±0.11 and 1.64±0.14, respectively. Results achieved in table (3) indicated that the mean values of moulds and yeast count (log cfu/g) of examined samples of frozen Tilapia nilotica, Basa, salted Mugil and Sardine were 0.93±0.18, 1.22±0.16, 1.67±0.21 and 1.69±0.18, respectively. Also, the results recorded in table (4) that the mean values of *Staphylococcal aureus* count (log cuf/g) of examined samples of frozen Tilapia nilotica, Basa, *Mugil cephalus* and Sardine were 1.30±0.17, 1.58±0.26, 2.12±0.14 and 2.17±0.17, respectively. Table (5) showed that incidence of *Salmonella*, *Liesteria monocytogenes* and *Vibrio parahaemoliticus* were not detected in all samples but *Listeria monocytogenes* and *V. parahaemoliticus* were detected in Basa (33, 33%) respectively. Moreover, the results in table (6) showed that 67% and 87% of frozen Tilapia nilotica and Basa fish samples were unaccepted according to E.O.S (2005, 8891). The results achieved in table (7) showed that all samples of salted fish (Mugil and Sardine) were unaccepted according to E.O.S (2005, 1725).

Table (1): Statistical analytical results of Total aerobic plate count log cfu/g in fish samples

No. of positive samples	Frozen fish		Salted fish	
	Tilapia nilotica	Basa fillet	Mugil	Sardin
	15	15	12	13
%	100	100	80	87
Mini.	3.79	3.40	0.90	1.70
Maxi.	5.99	6.15	2.92	3.48
Mean	4.80	4.63	2.35	2.70
SE	0.16	0.18	0.18	0.13

Table (2): Statistical analytical results of *Coliform* and *E. coli* counts log cfu/g in fish samples (N = 15 each)

No of positive samples	Frozen fish				Salted fish			
	Tilapia nilotica		Basa fillet		Mugil		Sardin	
	Coliforms	*E. coli*	Coliforms	*E. coli*	Coliforms	*E. coli*	Coliforms	*E. coli*
	10	5	14	8	12	7	9	6
%	67	33	93	53	80	47	60	40
Min.	0.48	2.01	0.79	1.30	0.79	1.00	2.08	1.00
Max.	2.86	2.78	3.54	2.80	2.79	1.60	2.97	1.95
Mean	1.84	2.52	2.19	2.01	2.10	1.31	2.52	1.64
SE	0.22	0.14	0.19	0.17	0.16	0.11	0.11	0.14

Table (3): Statistical analytical results of Mould and yeast count log cfu/g in fish samples (N = 15)

	Frozen fish		Salted fish	
	Tilapia nilotica	Basa fillet	Mugil	Sardin
No. of positive samples	8	10	12	12
%	53	67	80	80
Mini.	0.15	0.30	0.11	1.00
Maxi.	1.48	2.01	2.78	2.70
Mean	0.93	1.22	1.67	1.69
SE	0.18	0.16	0.21	0.18

Table (4): Statistical analytical results of Staphylococcus aureus count log cfu/g in fish samples

	Frozen fish		Salted fish	
	Tilapia nilotica	Basa fillet	Mugil	Sardin
No. of positive samples	5	5	15	15
%	33	33	100	100
Mini.	0.85	1.00	1.34	1.11
Maxi.	1.75	2.26	2.95	3.23
Mean	1.30	1.58	2.12	2.17
SE	0.17	0.26	0.14	0.17

Table (5): Frequency and percentage occurrence of bacterial isolates of fish samples

Isolates	Frozen fish				Salted fish			
	Tilapia nilotica		Basa fillet		Mugil		Sardin	
	No	%	No	%	No	%	No	%
Salmonella spp.	-	-	-	-	-	-	-	-
Listeria monocytogenes	-	-	1	3.33	-	-	-	-
Vibrio parahaemolyticus	-	-	1	3.33	-	-	-	-

Table (6) Acceptability of the examined samples of frozen fish samples according to EOSQC (2005/288)

	Acceptable limits	Tilapia nilotica Non Accepted N/15	%	Basa fillet Non Accepted No/15	%
APC	≤10^5	*0*	*0*	*1*	*7*
Coliforms	≤ 10^2	*10*	*67*	*13*	*87*
Moulds	Free	*8*	*53*	*10*	*67*
E.coli	Free	*5*	*33*	*8*	*53*
Listeria,monocytogenes	Free	*0*	*0*	*1*	*7*
Salmonella	Free	*0*	*0*	*0*	*0*
Staph aureus	Free	*2*	*33*	*5*	*33*
Vibrio parahaemoleticus	Free	*0*	*0*	*1*	*7*

Table (7) Acceptability of the examined samples of salted fish samples according to EOSQC (2005/3495)

	Acceptable limits	Mugil Non Accepted N/15	%	Sardin Non Accepted N/15	%
APC	*≤10^5*	*0*	*0*	*1*	*7*
Coliforms	≤ 10^2	*13*	*87*	*9*	*60*
Moulds	*Free*	*12*	*80*	*12*	*80*
E.coli	*Free*	*6*	*40*	*6*	*40*
Listeria,monocytogenes	*Free*	*0*	*0*	*1*	*7*
Salmonella	*Free*	*0*	*0*	*0*	*0*
Staph aureus	*Free*	*15*	*100*	*15*	*100*
Vibrio parahaemoleticus	*Free*	*0*	*0*	*0*	*0*

4. DISCUSSION

Fish and fishery products are not only nutritionally important but also important in global trade as foreign exchange earner for a number of countries in the world (Yagoub and Ahmed, 2003). Aerobic plate count on fishes generally do not relate to food safety hazards, but sometimes can be useful to indicate quality, shelf life and post heat processing contamination. The APC of the examined samples with an average value of 4.80±0.16, 4.63±0.18, 2.35±0.18 and 2.70±0.13 log cfu/g for the examined samples of frozen *Tilapia nilotica*, basa (frozen) *Mugil cephalus*, Sardine (salted) respectively. Considering frozen Tilapia nilotica samples higher results $3.08 \times 10^5 \pm 1.31 \times 10^5$ were recorded by Ibrahim et al. (2016). Meanwhile, our results appeared very low when compared to those observed by Edris et al. (2014) and Nayel (2007) who revealed that 60% of the examined samples of salted sardine had frequency range 10^5 to 10^6, also he found that 12% of examined samples of (Fesiekh) were 32% at frequency range 10^5 to 10^6.

There are two main problems associated with frozen storage of fish: hydrolysis and oxidation of lipids and protein denaturation. These problems cause an off taste and dry tough texture. Various factors, such as the freezing temperature, the rate of freezing, vacuum packaging or packaging materials, can affect frozen fish quality (Simeonidou et al. 1997). The aerobic bacterial count could reflect the quality of food sanitation during manufacturing, shipping and storage, and also provides an index of food freshness (Jyh-wei and Yin-hung, 2000).

Comparing to the results recorded in table (2) revealed that, the mean value of coliforms and *Escherichia coli* count in frozen fish (Tilapia nilotica – Basa fillet) and salted fish (*Mugil cephalus* and Sardine) were 1.84± 0.22, 2.52± 0.14 for Tilapia nilotica, 2.19±0.19, 2.01±0.17 for Basa and 2.10± 0.16,1.31±0.11 for Mugil and 2.52± 0.11, 1.64±0.14 for Sardine. Higher findings were observed by Popovic, et al., (2010) of the 60 frozen fish fillets samples analyzed, one fresh winter shellfish sample (0.41%) showed a level of *E. coli* cfu exceeding the given guideline of < 10^2 per g by four-fold, and therefore did not comply with the current legislation. Higher results were observed by Haque et al. (2013) who found the total Coliform counts (TCC) in the sample varied from 9.0×10^5 to1.03×10^5cfu/g of the frozen samples. .

Murad, et al. (2013) isolated *E.coli* from two marks but not from marks white fish fillet and Myanmar of fish fillets. Coliform (including *E.coli*) is a microorganism that can cause food infection inducing sickness in the digestion system, and is unavoidable without proper sterilization method (Jyh-wei and Yin-Hung 2000). Falcao, (2002) have provided evidence for that, the ice used to refrigerating seafood may be contaminated with coliform bacteria which cause human infection, as they discovered the presence of high numbers of coliforms, heterotrophic indicator microorganisms and pathogenic strains in ice used for chilling fish and other seafood; therefore, some of the contamination detected in the current study could be due to the ice used for chilling purposes at processing.

Fish are more liable to contamination with moulds and yeasts from animal and human reservoirs which may contaminate the water in the fishing area. Furthermore, contamination during handling and processing may occur. The contamination was increased in cases of fish caught from polluted areas (Hassan and Abdel Dayem, 2004 and Hassan et al., 2007).The results recorded in table (3) revealed that, the mean value of mould and yeast counts (log10 cfu/g) in the examined frozen fish (Tilapia nilotica -- Basa) and salted fish (*Mugil cephalus* -- Sardine) were

0.93±0.18, 1.22±0.16, 1.67±0.21 and 1.69±0.18.

Higher results than ours were observed by Samaha et al., (2015) who found that 80% of 25 examined imported frozen Basa samples were contaminated with mould, with a mean value of 3.0 $x10^3 \pm 2.1x10^2$ cfu/g. While it also posed that the yeast positive samples of examined 25 samples of imported frozen Basa was 64% and the mean value of total yeast count were 5.22 $x10^4\pm$ $1.80x10^4$ cfu/g, respectively. Fish are subjected to many risks of contamination from various sources either during their presence in their aquatic environment, sewage contamination of harvesting areas or after being harvested by workers, utensils and equipments during transportation, distribution and food preparation (EL-Leboudy2002). Also this result may be due to that very often the fish are displayed in open baskets or on tables beside the gutter or refuse dumps and this encourages fungi attack and subsequent production of toxins. For salted samples a higher findings were observed by Edris et al. (2012).

The incidence of moulds in fish could be attributed to improper sanitation during catching, handling, processing, salting, storage, transportation, distribution and or marketing of fish that could be resulted in undesirable changes of fish and rendering it unfit for marketing and increase the risk of infection with respective disease to consumers as a probable result of aflatoxins production by some fungal strains (Youssef 2011).

Apart from the enteric –organisms, *Staphylococcus aureus* encountered in this study are known enteriotoxic producing microorganism which is poisonous. Results achieved in Table (4) revealed that the mean counts of *Staphylococcus aureus* cfu/g in the examined samples of examined samples of frozen Tilapia nilotica, Basa, salted *Mugil cephalus* and sardine were 1.30±0.17, 1.58±0.26, 2.12±0.14 and 2.17±0.17, respectively.

Considering count of *staph aureus* in the Mugil cephalus and sardine were similar to results recorded by Saad et al. (2015) while a higher findings observed by Edris et al. (2014). Presence of *S.arueus* in food indicates its contamination from the skin, mouth and / or nose of food handlers. Inadequately cleaned equipment may be considered a source of contamination (Thatcher and Clark 1978). Staphylococci can grow best in salty and low water activity-containing foods in which the competing organisms are in reduced numbers (Vishwanath et al. 1998). Basti *et al*. (2006) showed that the *S. aureus* was the most important genus identified from heavy-salted fish and was due to the contamination of fish during capture and subsequent unhygienic handling and processing. We assumed that the isolated *S.aureus* were due to the contamination of fish during capture and subsequent unhygienic handling and processing (Shena et al. 2007). Isolated *S.aureus in* fishery products and fish processing factory workers. Small numbers of this bacterium in fishery products is not a serious problem but food poisoning may occur if the product is handled carelessly during processing, resulting in high multiplication (>1×10^5cfu/g) (Varnam and Evans, 2001; Vishwanath, et al. 1998). In our study also a plate count above 1×10^3 cfu /g *S.aureus,* a maximum acceptable concentration rate of this bacterium for the sardine fish according to ICMSF was obtained. Therefore, consumption of such products may cause a risk by *S. aureus* intoxication to consumers. As the *S. aureus* is an indicator of hygiene and sanitary conditions, the presence of this organism indicates the unhygienic condition during processing, storage et. It is recommended to use sanitary gloves for handling ready-to-eat foods to reduce the problem of *S. aureus* contamination.

Prevalence of some food poisoning organisms (*Salmonella, Listeria monocytogenes* and *Vibrio parahaemolyticus*

The results recorded in table (6) revealed that *Salmonella* failed to be isolated from analyzed samples in this study, which was in agreement with previous studies (Sulieman et al. 2014) in seafood products

Listeria monocytogenes failed to be detected in any examined samples except in frozen Basa fillets it could be isolated with percentage of 3.33% (table 5). In this concern, the literature contains information on *L. monocytogenes* isolation from soil, animals, birds, sewage, silage, stream water, mud, trout and crustaceans. Public health concerns have rapidly expanded from dairy products to processed meats and sea food products. A survey conducted on frozen seafood products in the United States showed some samples of (raw and cooked) shrimp, cooked crab meat, lobster tail, finfish and surimi to be positive for *L. monocytogenes*. In another survey *Listeria* sp. were isolated from 48 of 124 raw seafood samples and 24 of the 48 were *L. monocytogenes*. *L.monocytogenes* is most commonly associated with disease in both animals and humans. Pregnant women, neonates, elderly, or immune-compromised people are particularly susceptible to *Listeria* which manifests as abortion, stillbirth, septicemia, meningitis and meningoencephalitis (WHO, 2004). According to Food Net US, listeriosis was responsible for 30% of foodborne deaths from 1996 to 2005 and had a high case fatality rate of 16.9% (Barton et al. 2011). *V. parahaemolyticus* could be isolated from frozen Basa fillets with percentages 1 sample (3.33%) and failed to be detected in other fish products (Table 5). Higher results obtained by (Baffone, et al. 2000) that isolated *V. parahaemolyticus* from 5% of the examined marine fish samples. The isolation of *V.parahaemolyticus* from these marine fish samples could be attributed mainly to sewage pollution *V. parahaemolyticus* is a halophilic bacterium capable of causing food and waterborne gastroenteritis, wound infections and septicemia in humans. The microorganism is frequently isolated from a variety of raw seafood and shellfish. Consumption of raw or undercooked seafood contaminated with *V.parahaemolyticus* may lead to the development of acute gastroenteritis characterized by diarrhea, headache, vomiting, nausea and abdominal cramps (Caburlotto et al. 2008).

Table (6) showed that 67%, 87% were unaccepted based on their coliform count/g according to E.O.S (2005, 8891) of examined samples of` frozen Tilapia nilotica and Basa fillet respectively. Results achieved in table (7) all examined salted *Mugil cephalus* and salted Sardine samples were unaccepted based on their *staphylococcal aureus* count cfu/g according to E.O.S. (2005,1725)

5. CONCLUSION

Finally, the study concluded that frozen fish samples, which are ready for cooking, have acceptable microbial quality. However, the salted fish samples they may considered of high-risk from emetic toxins liberated from the high incidence of *staphylococcus aureus* found in the examined samples so, special attention should be taken from competent authorities and food business operators. Moreover, consumers are increasingly aware of the dangers of pathogens in salted fish.

6. REFERENCES

Abbas, K.A., Mohamed, A.; Jamilah B. and Ebrahimian, M. 2008. A review on correlations between fish freshness and pH during cold storage. Am. J. Biochem. Biotechnol., 4(4): 416-421.

Albarracin, W., Sanchrz, I.C, Grau, R. and Barat, J.M. 2011. Salt in food processing; usage and reduction; a review. Int. J. of Food Sci. and Technol., 46(7):1329-1336.

Ahmed, A., Dodo, A., Bouba, A.M., Clement, S. and Dzudie, T. 2011. Influence of traditional drying and smoke-drying on the quality of three fish species (*Tilapia nilotica, Silurus glanis and Arius parkii*) from Lagdo Lake, Cameroon. Journal of Animal and Veterinary Advances. 10 (3)301-306.

Ali, Mariyam. 2012. Shelf life determination of the brined Golden mullet during vacuum refrigerated storage using some quality aspect; Acta Scientiarum Polonorum– Technologia Alimmetaria, 11(1):37-43.

American Public Health Association APHA 2001. Compendium of methods for the microbiological examination of Food. 4th ed., Washington ,D.C.

Austin B. 2010. Vibrios as causal agents of zoonoses. Vet. Microbiol. 140:310–317.

Baffone, W., Pianetti, A., Bruscolini, F., Barbieri, E. and Citterio, B. 2000. Occurrence and expression of virulence-related properties of *Vibrio species* isolated from widely consumed seafood products. International Journal of Food Microbiol., 54: 9–18.

Barton Behravesh, C., Jones, T.F., Vugia, D.J., Long, C., Marcus, R., Smith, K.,Thomas, S.; Zansky, S.; Fullerton, K.E., Henao, O.L. and Scallan, E. 2011. Deaths Associated With Bacterial Pathogens Transmitted Commonly Through Food: Foodborne Diseases Active Surveillance Network (FoodNet), 1996– 2005. J. Infect. Dis., 204: 263–267.

Basti, A.A., Misaghi, A. and Salehi, T.Z. 2006. Bacterial pathogens in fresh, smoked and salted Iranian fish. Food Control. 17(3): 183-188.

Ben Embarek, P.K. 1994. Presence, detection and growth of Listeria nomocytogenes in seafoods: a review. Int. J. Food Microbiol. 23:17–34.

Benjakul, S.,Visessanguan, W., Thongkaew, C. and Tanaka, M. 2005. Effect of frozen storage on chemical and gel-forming properties of fish commonly used for surimi production in Thailand. Food Hydrcolloids, 10: 197-207.

Brown G. E. 2004. A Report on the Prevalence of Bacteria specie in Retailed Smoked Fish within Bauchi Metropolis.

Caburlotto, G., Ghidin, V.; Gennari, M.; Tafi, M. and Lleo, M. 2008. Isolation of *Vibrio parahaemolyticus* pandemic strain from a marine water sample obtained in the northern Adriatic. Eurosurveillance, 13: 1-3.

dris, A.A, Amin, A. Reham, Marionette Z. Naseif, Ebtsam M. Abdel Fatah. 2014. Evaluation of Retiled Salted Fish according to Egyptian Standard. Benha Veterinary Medical J. 27 (2):168-176.

El-Leboudy, S. H. 2002. Quality attributed of some local fresh and salted fishes and their improvement. Ph.D. Thesis, Dept. of food Hygiene, Fac. Vet, Med., Cairo University.

Falcao, J. P.; Dias, A. M. G.; Correa, E. F. and Falcao, D. P. 2002. Microbiological quality of ice used to refrigerate foods. Food Microbiol. 19: 269-276.

Feldhusen, F. 2000. The role of seafood in bacterial food-borne diseases. Microbes Infect. 2:1651–1660.

Food and Drug Administration (FDA), 2001. U. S. Department of health and human services. Public Health Service. Food and Drug Administration. College Park, MD 20740.

Food and Drug Administration (FDA), 2002. Enumeration of Escherichia coli and the coliform bacteria: Bacteriological Analytical manual. Chapter 4.

Food and Drug Administration (FDA), 2011. Quantitative Assessment of Relative Risk to Public Health from Foodborne Listeria monocytogenes Among Selected Categories of Ready-to-Eat Foods. Fish and Fishery Products Hazards and Controls Guidance Fourth Edition. Appendix 5: FDA and EPA Safety Levels in Regulations and Guidance.

Foran, J.A., carpenter, D.D., Hamlton, M.C., Knuth, B.A. and Schwager, S. J. 2005. Risk based consumption advice for farmed Atlantic and wild pacific salmon contaminated with dioxin and dioxin-like compound. Environmental Health Perspective. 33: 350-356.

Haque, M. M., Sorrowar, M.G. and Rashid, H. 2013. Effect of frozen storage, radiation and their combined treatments on microorganisms of freshwater Mola fish Amblypharyngodon Mola. Journal of Bangladesh Academy of Sciences. 37 (1) 21-31.

Hassan, A.A. and Abdel- Dayem, R.H. 2004. Prevalence of fungi and mycotoxins in fresh and salted fish. J. Egypt. Vet. Med. Assoc. 64 (1): 1-11 and 59- 68.

Hassan A.A.; Hammad, A.M; El Barawy,A.M.and Manal, A.H. 2007. Incidence of aflatoxigenic fungi in frozen and cannedfishes and trials to inhibit aflatoxin production by use of some minor elements and lupinustermis seeds. Egypt. J. Appl. Sciences. 22 (10B) 351-360.

Huss, H.H. 1995. Quality and quality changes in fresh fish. FAO Fisheries Technical Paper 348 FAO. Rome, Italy.

International Committee on Microbiological Specification for foods (ICMSF) 1996. Micro-organisms in foods 5.characteristics of Microbial Pathogen, Blachie Academic &Professional, London (ISBN0412 47350 X).

International Commission on Microbiological Specification for Foods (ICMSF) 1996. Vibrio parahaemolyticus. Microorganisms in Foods. Characteristics of Microbial Pathogens Blackie Academic & Professional, London. pp. 426–435.

ISO 21527-1. 2008. Microbiology of food and animal feeding stuffs -- horizontal method for the enumeration of yeasts and moulds -- part 1: Colony count technique in products with water activity greater than 0.95.

Jinneman, K.C., Wekell, M.M., and Eklund, M.W. 1999. Incidence and behavior of Listeria monocytogenes in fish and seafood products. In: Listeria, Listeriosis, and Food Safety. Ryser ET and Marth EH (eds.). New York: Marcel Dekker. 601–630.

Jaksic, S., Uhitil, S., Petrak, T., Bazulic, D., and Karolyi, L.G. 2002 Occurrence of Vibrio spp. in sea fish, shrimp and bivalve mollusks harvested from Adriatic Sea. Food Cont. 13:491–493.

Jyh-Wei, S. and Yin-Hung, H. 2000. Investigation for Contamination of Parasite and Aerobic Bacteria in Frozen Tilapia Fillets in Taiwan. J. Food and Drug Analysis. 8(1).

Kaya, Y. and Erkoyuncu, I. 1999. Degˇis¸ik Dumanlama Metotlarının Balık Turlerinin Kaliteleri Uzerine Etkisi. O.M.U. Ziraat Fakültesi Dergisi. 14 (1) 93-105.

Miettinen H and Wirtanen G. 2005. Prevalence and location of Listeria monocytogenes in farmed rainbow trout. Int J Food Microbiol. 104:135–143.

Murad, H. O. M.; Khidhir, Z. Kh. and Arif, E. Dh. 2013. Assessment of the microbial quality of imported frozen fish fillets in Sulaimani markets. Al-Anbar J. Vet. Sci. 6 (1): 24-31.

Nielson, J., Boknaes, N. and Jessen, K. 1994. In quality index method for frozen fish. Proceedings of 24th Annual Meeting of Western European Fish Technologists Association, Nantes, France. 60-62.

Olafsdóttir, G.; Martinsdóttir, E.; Oehlenschläger, J.; Dalgaard, P.; Jensen, B., Undeland, I. and Nilsen, H. 1997. Methods to evaluate fish freshness in research and industry. Trends in Food Science & Technology, 8:258-265.

Popovic, N. T.; Skukan, A. B. ; Dzidara, P.; Coz-Rakovac, R. ; Strunjak-Perovic, I.; Kozacinski, L. ; Jadan, M. and Brlek-Gorski, D. 2010. Microbiological quality of marketed fresh and frozen seafood caught off the Adriatic coast of Croatia. Veterinarni Medicina, 55 (5): 233–241.

Prescott, L.M., Harry, J.P. and Klein, D.A. 1999. Food and Industrial Microbiology. Chapter 43, 4th Edition, New york, Mc Graw Hill publication.

Richards, M.P. 2002. Contributions of blood and blood components to lipid oxidation in fish muscle. PhD Thesis, University of Massachusetts, Amherst, USA.

Rørvik L.M., Skjerve, E., Knudsen B.R., and Yndestad M.1997. Risk factors for contamination of smoked salmon with Listeria monocytogenes Int J Food Microbiol. 37:215–219.

Saad, S.M.; Edris, A.M.; Salem, M. Amani; Hassan, A. Enas and Mostafa, M. Eman. 2015. Incidence of Some Food Poisoning Microorganisms in Salted fish. Benha Vetrenary Medical J. 29. (2):225-229.

Shena, S. S. and Sanjeev, S. 2007. Prevalence of enterotoxigenic Staphylo*coccus aureus* in fishery products and fish processing factory workers, Food Control, 18 (12) 1565- 1568.

Simeonidou, S.; Govaris, A. and Vareltzis, K. 1997. Effect of frozen storage on the quality of whole fish and fillets of horse mackerel (Trachurus trachurus) and Mediterranean hake (Merluccius mediterraneus). ZLebensm UntersForsch, 204: 405-410.

Su YC and Liu C. 2007. Vibrio parahaemolyticus: a concern of seafood safety. Food Microbiol 24:549–558.

Tidwell, J.H. and Allan, G.L. 2001. Fish as food: Aquaculture's contribution. EMBO Reports, 2(11): 958-963.

Turan, H.; Sonmez, G.; Celik, M.Y. and Yalcin, M. 2007. Effect of different Salting process on the storage quality of Mediterranean Muscle role as cheap protein supplement and their significance as (Mytilus galloprovincialis L. 1819). J. Muscle Food, 18: 320-390.

Varela, P. and Fiszman, S.M. 2011. Hydrocolloids in 36:647-655. Fried foods. A review. J. of Food Hydrocolloids, 25:1801-1812.

Varnam,A. H. and Evans, M. G., 2001. Food borne pathegens, wolf publications, London, pp: 157-183.

Vishwanath, W., Lillabati, H. and Bijen, M.1998. Biochemical, nutritional and microbiological quality of fresh and smoked mudeel fish monopterus albus, Food Chemistry. 6 (12): 153-157.

WHO 2004. World Health Organization and Food and Agriculture Organization. Risk assessment of Listeria monocytogenes in ready-to-eat foods, MRA Series No. 4–5. Rome, Italy.

Yagoub, S. O. and Ahmed, T. M. 2003. Pathogenic Microorganisms in fresh water samples collected from Khartoum central market. Sudan J. of Veterinary Science and Animal Husbandry 43(1-2): 32–37.

Youssef, H.H.E.1998. Mycological status of Moloha, smoked herring and frozen mackerel fish in Assuit province. Ph.D. Thesis, Dept. Food Control, Fac. Vet. Med. Assiut University.