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Quality Evaluation of Some Fresh and Imported Frozen Seafood

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Abstract: This study was done to evaluate the quality parameters of fresh and imported frozen seafood (fillets and shrimp). A total of 120 seafood samples, fillets and shrimp (fresh and imported frozen) 30 each, collected from fish markets at Giza Governorate were sensory, bacteriological, chemical investigated. Panelists rejected 5.0 (16.6%) and 0.0 (0.0%) of fresh fillets and shrimp samples, while the number raised up to 12.0 (40.0%) and 15.0 (50.0%) of frozen fillets and shrimp samples respectively. Fresh seafood had a significant lower ($p<0.05$) pH values in compared to frozen products. There were observed significant difference ($p<0.05$) in the TBA and TVB-N values between the fresh and frozen seafood samples. All fresh and frozen seafood samples were judge as safe food from microbiological point of view. The total proteolytic, lipolytic, psychrotrophic and pseudomonas/aeromonas counts for all examined seafood samples were lie within the standard permissible limits. *L. monocytogenes* could not be detected from fresh or frozen fillets. Few freqyency of *L. monocytogenes* found in fresh and imported frozen shrimp. As well as *V. parahaemolyticus* and *Y. enterocolitica* were recovered from examined fresh and frozen seafood samples. The public health significance of the isolated organisms was also discussed. The quality of fresh fillets and shrimp were better than that of imported frozen one.

Key words: Fresh fish fillets, frozen, imported, *L. monocytogenes*, quality parameters, shrimp, *V. parahaemolyticus*

INTRODUCTION

Seafood can be defined as any fish or shellfish from the sea used for food. 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 world wide (Nielson *et al*., 1994). The most serious seafood safety issues resulting in potentially contaminated products are associated with microbial and especially bacterial pathogens. Seafood are highly susceptible to both microbiological and chemical deterioration due to its high water content, neutral pH, relatively large quantities of free amino acids, and naturally presence of autolytic enzymes (Jeyasekaran *et al.*, 2006).

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). Changes in flavor, odor, texture and color reflect the level of seafood freshness. Sensory assessment is the use of one or more of the five senses to judge, or form an opinion on, some aspect of quality. The senses in question are sight, smell, taste, touch and hearing (FAO/Codex, 1999).

Chilling and freezing storage are the normally employed methods for fish preservation, however the quality deterioration of stored fish is inevitable with extend shelf-life (Jeon *et al*., 2002). Large quantities of seafood are harvested from cold water; therefore, the microflora is not inhibited as effectively by refrigeration as is the normal microflora of warm blooded animals (Nickelson *et al*., 1980). The predominate microflora of shrimp consisted of Corynebacteria, Pseudomonas, Moraxella and Micrococcus (Vanderzant *et al*., 1970). Seafood are susceptible to all of the common food poisoning organisms as well as to some that are unique to marine products such as *C. botulinum* and *Vibrio* spp. Strains of public health concern common to all food products include *Yersinia enterocolitica*, *Listeria monocytogenes* and *Vibrio parahaemolyticus*.

Listeria monocytogenes is a public health concern in many countries. Several ready to eat foods including smoked fish and pate have been implicated in the outbreaks of human listeriosis (Miettinen *et al*., 1999). Products with a long chilled shelf-life which are consumed without further cooking especially constitute a risk since it has been demonstrated that *L. monocytogenes* could grow to high levels during long term refrigerated storage (Rocourt *et al*., 2003). *Vibrio parahaemolyticus* is recognized as the leading cause of human gastroenteritis associated with seafood consumption in the United States (Mead *et al*., 1999).

Based on the above, seafood safety concerns, challenges and related issues that will continue being of

concern in the 21st century. Therefore, the aim of this study was to evaluate the quality parameters of fresh and frozen seafood (fillets and shrimp) through sensory, bacteriological, chemical testes.

MATERIALS AND METHODS

Collection of samples: A total of 120 seafood samples, fillets and shrimp (fresh and imported frozen) 30 each, were collected from fish markets at Giza Governorate. The samples were wrapped in sterile polyethelene bags, identified then placed in ice-box and rapidly transferred to the laboratory of Food Hygiene Department of Faculty of Veterinary Medicine, Cairo University for further evaluation.

Sensory evaluation:
Sensory evaluation was done according to the technique recommended by FAO/Codex (1999) as follows:

Sensory attributes	Accepted	Rejected
Appearance	Translucent, glossy, natural color	opaque, dull, blood stained, discolored, blackenig on head and body in shrimp
Texture	Firm, elastic	soft, plastic
Odour	Marine, fresh, neutral	sour, stale, spoiled, ammoniacal, putrid
Drip in frozen fillets	Slight, moderate	Abundant

Bacteriological evaluation: Samples homogenate was prepared by homogenization of 10 g of the examined sample with 90 mL of 0.1%. Sterile peptone water with a homogenizer (Universal Laboratory Aid made in poland). From fish/shrimp homogenate, serial decimal dilutions up to 10^6 were performed. The microbiological procedures recommended by the American Public Health Association (APHA, 2002) were applied as follows: the total proteolytic count was performed using skimmed milk agar 10% (*Oxoid*, *CML 31*), total lipolytic count on tributyrine agar (*Oxoid*, *PM4*), total Psychrotrophic count on standard plate count agar (*Oxoid*; *CM 325*) with incubation at 25ºC for 48 h and total pseudomonas/aeromonas count on Glutamate starch phenol red agar medium (*GSP*; *Merck*; *Art 10230*).

Isolation of *Yersinia enterocolitica* on modified Rappaport broth as selective enrichment and incubation at 25ºC for 3 days, then plating on Yersinia Selective Agar Base (*Oxoid*; *CM 653*) to which Yersinia Selective Supplement (Oxoid; SR 109) was added and plates incubated at 32ºC for 18 h.

Isolation of *Listeria monocytogenes* on Buffered listeria enrichment broth (*Oxoid, CM 897*) with listeria selective enrichment supplement (*Oxoid, SR141*) and incubated at 30ºC for 24 h, then selective plating on modified Oxford agar plate (*Oxoid, CM 856*) with listeria selective supplement (*Oxoid, SR 140*), plates were incubated at 35ºC for 48 h. Isolation of *Vibrio parahaemolyticus* with selective enrichment on 0.1% peptone water supplemented with 3% sodium chloride, incubated at 37ºC overnight, then selective plating on Thiosulphate Citrate Bile Succrose Agar (*TCBS*, *BBL*) and incubated at 37ºC for 24 h.

Chemical evaluation: pH was estimated according to the technique described by Duan *et al*. (2010), thiobarbituric acid (TBA) value using the distillation method was applied according to a technique described by the Food and Agriculture Organization (FAO, 1986). TBA values were expressed as mg malondialdehyde/kg. Total Volatile Bases Nitrogen (TVBN) was determined by using the distillation method performed as recommended by the Analytical Methods Committee (AMC, 1979). TVBN were expressed as mg/100 g.

Statistical analysis: The data obtained were analyzed for significance using the General Linear Model (GLM) procedure of the SAS Institute Inc., mean was applied to determine significance between different values (SAS, 2000).

RESULTS AND DISCUSSION

Sensory evaluation of seafood: Sensory evaluation is the most important method today for freshness evaluation of the seafood. Table 1 shows the appearance, texture and odor of examined fresh and frozen fillets and shrimp samples. The number of accepted samples by panelists for fresh fillets and shrimp samples were 25.0 (83.3%) and 30 (100%), respectively. For frozen samples, the acceptable samples number were 18.0 (60%) and 15.0 (50%) respectively. Panelists rejected 5.0 (16.6%) and 0 (0%) of fresh fillets and shrimp samples, while the number raised up to 12.0 (40%) and 15.0 (50%) of frozen fillets and shrimp samples respectively. The organoleptic properties of the examined samples indicated that the products were acceptable according to the panel evaluation, though statistically there was significant difference ($p<0.05$) in the sensory evaluation between the fresh and frozen samples.

Chemical evaluation of seafood: The results of Table 2 and 3 revealed that pH of fresh and frozen fillets samples were 6.5 and 6.6, respectively while for fresh and frozen shrimp were 6.6 and 7.5, respectively. Fresh seafood had a significant lower ($p<0.05$) pH values in compared to frozen products. The pH value of seafood varies from 5.8-7.2 depending on struggling at the time of harvesting but the normal variation is of pH 5.8-6.5. The pH is an important determinant of microbial growth and the high pH seafood has a high spoilage potential and a short shelf-life (Newton and Gell, 1981). Walker and Betts (2000) reported that ultimate pH of meat was significant for its resistance to spoilage because most bacteria grow

Table 1: Sensory evaluation of the fresh and imported frozen fillets and shrimp

		Fillets		Shrimp	
Parameter		Fresh	Frozen	Fresh	Frozen
Accepted, translucent,	Number**	25.0*	18.0	30.0*	15.0
firm and fresh odor	percentage	83.3	60.0	100	50
and offensive	Number***	5.0*	12.0	0.0*	15.0
Rejected, dull, soft	percentage	16.6	40	0.0	50

*: Significantly differ at p<0.05; **: number of accepted samples; ***: number of rejected samples

Table 2: Statistical analysis of chemical examination of fresh and imported frozen fillets

Type of fillets	pH	TBA mg mal/kg	TVB-N mg/100 g
Fresh fillets	6.5±0.30*	0.642±0.120*	19.50±0.70*
Frozen fillets	6.9±0.52	0.959±0.200	23.42±0.95

*: Significantly lower at p<0.05

Table 3: Statistical analysis of chemical examination of fresh and imported frozen shrimp

Type of shrimp	pH	TBA mg mal/kg	TVB-N mg/100g
Fresh shrimp	6.6±1.52*	0.550±0.020*	20.90±2.75*
Frozen shrimp	7.5±1.82	0.959±0.600	24.65±3.50

*: Mean significantly lower at p<0.05

optimally at about pH 7 and not well bellow pH 4 or above pH 9 (Jamilah *et al*., 2008). Microbial load increased with the increase in final pH of the meat (Dharmaveer *et al*., 2007).

The results of TBA and TVB-N values obtained from the chemical analysis of seafood samples are given in Table (2, 3). TBA of fresh and frozen fillets samples were 0.642 and 0.959, respectively, while for fresh and frozen shrimp were 0.55 and 0.959, respectively. TVB-N of fresh and frozen fillets samples were 19.50 and 23.42, respectively, while for fresh and frozen shrimp were 20.90 and 24.65, respectively. The TBA and TVB-N values of the examined samples indicated that all fresh and frozen seafood samples were acceptable according to the The Egyptian Organization for Standardization and Quality Control (EOS, 2005) which recommended that the permissible limit for TBA be not more than 4.5 mg malonaldehyde /Kg and for TVB-N not more than 30 g/100 g. Whatever, there were observed significant difference (p<0.05) in the TBA and TVB-N values between the fresh and frozen seafood samples. Post-harvest the lipids in fish may undergo lipolysis or auto-oxidation (Hardy, 1980), both of which constitute important chemical spoilage processes in fish (Huss, 1995).

Bacteriological evaluation of seafood: The total proteolytic, lipolytic, psychrotrophic and pseudomonas/ aeromonas counts for fresh fillets samples were 2.87±0.14, 2.46±0.08, 4.19±0.32 and 3.65±0.22 Log_{10} cfu/g, respectively. The counts for the same microbial groups for frozen fillets samples were 3.20±0.05, 3.94±0.21, 4.99±0.24 and 3.93±0.33 Log_{10} cfu/g frozen fillets, respectively (Table, 4), as well as these counts in fresh shrimp were 2.10±0.05, 3.00±0.10, 4.10±0.50 and 2.75±0.25 Log_{10} cfu/g, respectively in corresponding to

Table 4: Mean values of bacteriological examination log $_{10}$ CFU/g) of fresh and imported frozen fillets

Microorganism	Fresh fillets	Frozen fillets
Proteolytic count	2.87±0.14*	3.20±0.05
Lipolytic count	2.46±0.08*	3.94±0.21
Psychrotrophic count	4.19±0.32*	4.99±0.24
Pseudomonas/Aeromonas count	3.65±0.22	3.93±0.33

*: Significantly lower at p<0.05

Table 5: Mean values of bacteriological examination log $_{10}$ CFU/g of fresh and imported frozen shrimp

Type of microorganism	Fresh shrimp	Frozen shrimp
Proteolytic count	2.10±0.05*	3.0±0.60
Lipolytic count	3.00±0.10*	3.69±0.52
Psychrotrophic count	4.10±0.50*	5.50±1.04
Pseudomonas/Aeromonas count	2.75±0.25*	3.73±0.90

*: Significantly lower at p<0.05

3.0±0.60, 3.69±0.52, 5.50±1.04 and 3.73±0.90 Log_{10} cfu/g, respectively in frozen shrimp (Table 5) . These counts were significantly lower (p<0.05) in fresh fillets and shrimp than counts of imported frozen one. Flesh of live healthy fish is considered bacteriologically sterile. The largest concentrations of microorganisms are found in the intestine, gills and surface slime. The numbers and types of microorganisms found on freshly caught fish are influenced by the geographical location of the catch and the season and method of harvest (Shewan, 1961). Seafood spoilage required contamination of foods by proteolytic and lipolytic bacterial strains that have the ability to produce extracellular protease and lipases enzymes that can decomposed the protein and fat to low molecular weight substances. Protease enzymes can attack the nitrogen molecules naturally occur in meat causing, severe deteriorative changes in the color and odor of foods even under refrigerated or frozen storage.

In this study, all fresh and frozen seafood samples were judge as safe food from microbiological point of view. The total proteolytic, lipolytic, psychrotrophic and

Table 6: Incidence of isolated organisms from fresh and imported frozen fillets

	Fresh fillets		Imported frozen fillets	
Microorganism	No.**	%	No.**	%
Y. enterocolitica	15.0	50.0	8.0*	26.6
L. monocytogenes	0.0	0.0	0.0	0.0
V. parahaemolyticus	0.0*	0.0	5.0	16.6

*: Significantly lower at p<0.05; **: number of positive samples

Table 7: Incidence of isolated organisms from fresh and imported frozen shrimp

	Fresh shrimp		Frozen shrimp	
Microorganism	No.**	%	No.**	%
Y. enterocolitica	5.0*	16.6	10.0	33.3
L. monocytogenes	3.0*	10.0	5.0	16.6
V. parahaemolyticus	5.0*	16.6	7.0	23.3

*: Significantly differ at p<0.05; **: number of positive samples

pseudomonas/aeromonas counts for all examined seafood samples were lie within the permissible limits recommended by Foster *et al*. (1977) and ICMSF criteria (1974).

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 a dry and tough texture. Various factors, such as the freezing temperature, the rate of freezing, vaccum packaging or packaging materials, can affect frozen fish quality. Frozen fish are often stored in the form of fillets, however, filleting operations can affect frozen fish quality reported that the (Simeonidou *et al.,* 1997).

Pseudomonas, the most common spoilage organism was not isolated from pond water or from shrimp, possibly indicating a difference in the harvesting and handling techniques (Vanderzant *et al.,* 1970). These organisms are capable of causing spoilage because of two important characteristics. First, they are psychrotrophic and thus multiply at refrigeration temperatures. Secondly they attack various substances in the fish tissue to produce compounds associated with off flavor and off odor (Miller *et al.,* 1973).

The data obtained in Table (6, 7) showed that *Y. enterocolitica* was isolated with percentages 50 and 26.6% from fresh and frozen fillets respectively. Also *Y. enterocolitica* could be isolated with percentages 16.6 and 33.3% from fresh and imported frozen shrimp respectively. In this respect, *Y. enterocolitica* has been isolated from raw seafood products and can grow at refrigeration temperatures. Pathogenic strains have been isolated from crabs harvested from cold water (Faghri *et al*., 1984).

L. monocytogenes failed to be detected in any examined samples of fresh and imported frozen fillets (Table 6) and it could be isolated with percentages 10 and 16.6 from fresh and imported frozen shrimp respectively (Table 7). 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 shrimp (raw and cooked), 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*. The highest incidence was found in fresh water catfish in which 15 out of 20 samples were positive for *L. monocytogenes*. Listeriosis has been implied from raw shellfish or raw finfish in an outbreak in New Zealand.

V. parahaemolyticus could be isolated from frozen fish fillets with percentages 16.6 and failed to be detected in fresh fillets. Also it could be isolated with percentages 16.6 and 23.3 from fresh and imported frozen shrimp respectively (Table 6, 7). *V. parahaemolyticus* is a halophilic bacterium capable of causing food and waterborne gastroenteritis, wound infections and septicaemia 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).

CONCLUSION

From the present study, it could be concluded that the sensory evaluation of fresh fish fillets and shrimp were significantly accepted than of imported frozen, as well as the pH ,TBA mg malondialdehyde/kg and TVB-N mg/100 g of fresh seafood were significantly lower at p<0.05 than that of imported frozen. Also, proteolytic, lipolytic and psychrotrophic counts were significantly lower at p<0.05 in fresh seafood than imported frozen. *Y. enterocolitica*,

L. monocytogenes and *V. parahaemolyticus* could be detected in fresh and frozen seafood with different percentages.

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