# HEALTH HAZARD ASSOCIATED WITH SALTED FISH IN EGYPTIAN MARKET

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#### **Abstract**

Thirty random samples of salted fish (15 each of fesiekh and salted sardine) were collected from different markets in Cairo and Giza Governorates and subjected to physical, chemical and microbiological examination. The physical character of the examined samples did not show any significant abnormality. The chemical examination revealed PH values of 7 and 6.9 for fesiekh and salted sardine respectively, which exceed the permissible limit as recorded by Egyptian Organization for Standardization and Quality Control (EOSQC) (1996). The mean values of moisture % were 49.5 and 50.8 % for fesiekh and salted sardine, respectively, which were within the permissible limit as recommended by EOSQC (1996). Moreover, the maximum values of sodium chloride % were 23 and 25% for fesiekh and salted sardine, respectively. The maximum values of histamine level and Total Volatile Base-Nitrogen (TVB-N) were (35 and 30 mg/100g) and (45 and 40 mg/ 100g) for fesiekh and salted sardine respectively. The values of NaCl and histamine exceeded the permissible limit given by EOSQC (1996). The values of TVB-N exceeded the permissible limits of fresh fish (30 mg/100g) as recommended by EOSQC (2003). Microbiological examination revealed that the mean values of anaerobic count, S. aureus count, Enterobacteriaceae count and coliforms count were  $(3.1 \times 10^4 \text{ and } 1.7 \times 10^3 \text{ cell/g}), (2.2 \times 10^4 \text{ and } 1.5 \times 10^3 \text{ cell/g}), (7.4 \times 10^2 \text{ and } 1.7 \times 10^3 \text{ cell/g})$ 1.6x102 cell/g) and (34.33 and 9.47) for fesiekh and salted sardine, respectively. These values exceeded the permissible limit recorded by EOSQC (1996).

Finally, it was concluded that the process of salting must be done under complete hygienic condition in order to minimize the risk of high bacterial load to become safe for consumer.

### INTRODUCTION

Fish consumption has been increased in the recent years. In the future, fish are considered as one of the important sources of protein for human consumption. Fish like any other food, could be contaminated during handling and processing with many of the well known food poisoning organisms. Contamination of fish with organisms of public health significance remains primarily a problem of handling and processing (WHO, 1999).

It is evident that consumption of salt dried fish may increase the level of salt intake leading to cardiovascular problems (Santosa and Quantick, 1991).

Rodriguez-Jerez *et al.* (1994) studied the count evaluation of total aerobic mesophilic, psychotropic, Enterobacteriaceae, fecal coliform organisms in Spanish-preserved anchovies. They pointed out that the count of such organisms could be used as sanitary index of the product and may produce high concentration of histamine. Sodium chloride concentration was the main factor influencing the decreasing bacterial counts.

Staphylococcus food poisoning is considered as one of the major form of foodborne disease and its toxic symptoms usually appear within 0.5 to 7 or 8 hours after consumption of contaminated food by the Staphylococcal enterotoxins. The common reported symptoms include nausea, vomiting, retching and less frequently diarrhoea, headache, dizziness and weakness reported in minority of cases. There were few deaths recorded, especially in old or very young peoples (Varnam, 1990).

Reilly and Santos (1985) claimed that a high level of histamine indicates poor handling and processing of fish products. They added that delay in salting of fish resulted in higher histamine content. Enterobacteriaceae has important histamine producing activity. Moreover, they are sensitive to elevated sodium chloride concentrations. The isolated Staphylococcus could be considered as halotolerant bacteria. If this kind of microorganisms multiply in salted fish, histamine formation can appear with a risk for consumer's health. The authors concluded that the accumulation of high histamine concentration in salted fish could be due to poor quality of the raw material or to unhygienic handling as the histamine concentration is probably increased due to the presence of the halophilic or halotrolerant microorganisms (Rodriguez-Jerez et al., 1994).

# **MATERIALS AND METHODS**

#### Samples

Thirty random samples of salted fish (15 each of fesiekh and salted sardine) were collected from different markets in Cairo and Giza Governorates. Each sample was wrapped separately in sterile polyethylene bag. The collected samples were transported to the laboratory, where they were subjected to physical, chemical and bacteriological examinations.

# 1. Physical examination (Borgstorm, 1965)

Attachment of scales, muscle texture, colour and odour of the flesh with examination of viscera and abdominal wall were carried out.

# 2. Chemical examination (AOAC, 1990)

2.1 Measurement of pH value

(AOAC, 1990)

2.2 Determination of moisture %

(AOAC, 1990)

The technique was carried out using ten grams of fish flesh, which were placed in a previously weighed porcelain dish, then, dried in hot air oven at 100 °C for four hours till two successive fixed weights were obtained. The moisture content was calculated.

2.3 Determination of sodium chloride % (AOAC, 1990)

It was carried out using silver nitrate (0.1N) precipitation technique.

2.4 Determination of histamine (AOAC, 1990)

The histamine analysis was carried out by thin layer chromatography (TLC). Ten grams of minced fish muscle was weighed and 70 ml methanol was added then, mixed thoroughly, homogenized and filtered. Twenty microliters of the filtrate was spotted with standard of histamine directly on the TLC plate, then solvent system (80 ml acetone: 20 ml ammonia) was used to separate the histamine from sample extract. The plates dried then, sprayed with ninhydrin. The quantitative concentration of histamine was calculated by comparing with the standard.

2.5 Determination of total volatile base nitrogen (TVB-N) (AOAC, 1990)

The Conway's microdiffusion technique was applied. The sample (25 g fish muscle) was extracted by using distilled water acidified by 2M HCl till pH 5.2, then, heat slowly to  $70^{\circ}$  C then, filter. The Conway's dish was covered and incubated at 36 ° C for 2 hours after dispending 2 ml extract (filtrate) with 1 ml

saturated potassium carbonate in outer ring and 2 ml 0.01 N HCl in inner ring. The titration was done (after incubation) with 0.01 N NaOH using methyl red as indicator till faint yellow colour end point. The TVB-N was calculated from the equation.

#### 3. Bacteriological examination (APHA, 1992)

Preparation of fish homogenate

Ten grams from each sample were aseptically placed in a sterile blender with 90 ml of sterile peptone water and homogenized for two minutes, then, serial dilutions were prepared in sterile peptone water 2%, then, subjected to the following examinations:

#### a. Total bacterial count

The total bacterial count per  $cm^2$  /g was done by using the plate count agar in duplicated plates and incubated at 30  $^{\circ}$  C for 48 hours.

#### b. Anaerobic bacterial count

The plate media [reinforced Clostridium media (RCM)] were streaked with 0.1 ml of the first or second dilution, then, incubated anaerobically at 37  $^{\circ}$  C for 48 hours

# c. Staphylococcus aureus count

The *S. aureus* was enumerated using Barid Parker media and incubated at 37 °C for 24-48 hours.

### d. Enterobacteriaceae count

The drop technique was applied using violet red bile glucose agar. Plates were incubated at 37  $^{\circ}$  C for 24-48 hours. All purple colonies were counted.

# e. Coliforms count [most probable number (MPN)]

Using the three tube methods of MacConky broth, tubes showing acid and gas production were considered positive. The MPN was estimated (MPN tables).

# **RESULTS AND DISCUSSION**

# 1. Physical examination

Estimation of the organoleptic quality of the examined samples was illustrated in Table 1. The texture of muscles varied from firm to tender. The variation between the samples of fesiekh and salted sardine referred to the difference in the manufacture. The normal colour of the flesh was red (60% in fesiekh and 66.7% in salted sardine). The abnormal colour of flesh was greyish-yellow. This indicated that

the samples were salted for a long period and the greyish-yellow colour indicated fat oxidation. All the samples had salty odour and taste. Nearly similar results were recorded by Gehan (1996).

#### 2. Chemical examination

The pH values of fesiekh and salted sardine were (7 and 6.9) as recorded in Table 2 which exceeded the permissible limit (6-6.5) as estimated by EOSQC (1996). The obtained results supported those recorded by Ahmed (1976).

The moisture % given in Table 2 revealed that the mean % of fesiekh and salted sardine were 49.5% and 50.8% within the permissible limit (50-55%) given by EOSQC (1996). The moisture of salted sardine was higher than the results obtained by Ahmed (1976), Morshdy (1980) and Gehan (1996). This difference may be due to the seasonal impact on fish composition or due to differences in manner of salting or both.

The values of NaCl% for fesiekh and salted sardine were 23% and 25% as recorded in Table 2. These values exceeded the permissible limit (15-22%) as given by EOSQC (1996).

The maximum values of histamine content of fesiekh and salted sardine were 35 and 30 mg/100g of sample, respectively, as given in Table 2. Such results indicated that histamine content in all samples exceeded the permissible limit (20 mg/100g of sample) given by EOSQC (1996). The high level of histamine in the investigated samples could be attributed to the bacterial decarboxylase activity due to poor quality of raw fish material, mishandling or other causes during their shelf life. Such data agreed with Rodriguez-Jerez *et al.* (1994).

From the results recorded in Table 2, the mean values of total volatile base nitrogen (TVB-N) for fesiekh and salted sardine were 39.5 and 32.2 mg/100g, respectively. The permissible limit of TVB-N was not recommended by EOSQC (1996), but the obtained results exceeded the permissible limits of fresh fish (30 mg/ 100g) as recommended by EOSQC (2003)

# 3. Bacteriological examination

From the results obtained in Table 3, the mean values of aerobic plate count (APC) in fesiekh and salted sardine were  $3.7 \times 10^5$  and  $1.1 \times 10^5$  cell/g. Nearly similar results were obtained by Ahmed (1976) and Rodriguez-Jerez *et al.* (1994).

The mean values of anaerobic bacterial count for fesiekh and salted sardine were  $3.1 \times 10^4$  and  $1.7 \times 10^3$  cell/g, respectively, as shown in Table 3. Nearly similar results were obtained by Elias (1968), and these might be assigned to the same storage-conditions after processing. These limits exceeded the permissible limit ( $10^2$  cell/g) as recommended by EOSQC (1996)

The high *S. aureus* count was due to excessive contamination and handling during the processing. The mean *S. aureus* counts were  $2.2 \times 10^4$  and  $1.5 \times 10^3$  cell/g for fesiekh and salted sardine. Similar findings were recorded by Rashad (1986). *S. aureus* count was significantly correlated to anaerobic count and histamine content in the salted fish samples. These limits exceeded the permissible limit ( $10^2$  cell/g) as recommended by EOSQC (1996)

The mean values of Enterobacteriaceae of fesiekh and salted sardine were  $7.4 \times 10^2$  and  $1.6 \times 10^2$  cell/g, respectively (Table 3). The relatively low count of Enterobacteriaceae may be attributed to the sensitivity of this family to high salt concentration, which agrees with the results reported by Gibsons and Roberts (1986).

The results of Coliforms count (MPN) recorded in Table 3 indicated that the mean coliforms count in fesiekh and salted sardine were 34.33 and 9.47 which exceeded the permissible limit as given in EOSQC (1996). These agree with the results obtained by De Man (1975).

Finally, to obtain high quality of salted fish, the following recommendations should be taken:

- 1. Minimize the contamination of raw material.
- The salting process must be done under complete hygienic conditions (handling, processing).
- Salting of fresh fish must be done without delay to inhibit bacterial growth and to control the histamine level in the final product.

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Table 1. Physical examination of the collected salted fishes (n=15).

Salted sardine %	%	No.		Types of salted fish
40	40	6	F	Texture of muscles
60	60	9	Т	muscles
66.7	60	9	Z	Colour of flesh
33.3	40	6	Ab	of flesh
100	100	15	S	Odour
100	100	15	S	Taste

n= number of the examined samples
N= normal Ab= abnormal

F= firm T= tender S= salty

Types of salted fish	pH value	Je .		Moisture %	е %		NaCI %			Histami	Histamine content	₽	TVB-N		
										(mg/10	0g)		(mg/100g)	0g)	
	Min.	Max.	Mean	Min.	Max.	Mean	Min.	Max.		Min. Ma	Max.	Mean	Min.	X.	Mean
			3S #			± SE						± SE			± SE
Fesiekh	6	7	6.6 ±0.1	45	55	49.5 ±0.8	12	23	19.1 ±0.9	10	35	23.3 ±2.3	25	45	39.5 ±1.3
Salted sardine	5.9	6.9	6.5	47	54	50.8	=	25	18.5	15	30	22.1	22	40	32.2
	0.5	0.9	±0.1	4	7	±0.6	-	22	±1.2	5	5	±1.4	77	4	#1.5

Table 3. Bacterial count of muscles of examined salted fish (n=15).

Table 3. Bacteria	al count of	Table 3. Bacterial count of muscles of examined salted fish $(n=15)$ .	risn (n=15).				ſ
Types of salted fish	fish	Aerobic plate	Anaerobic bacteria	S. aureus	Enterobacteriaceae	Coliforms	
Fesiekh	Min.	2.5×10 <sup>4</sup>	2.0x10²	9.2×10²	2.8x10 <sup>2</sup>	6.0	
	Мах.	9.3×10 <sup>5</sup>	8.0×10 <sup>4</sup>	8.2×10 <sup>4</sup>	1.2x10³	85.0	
	Mean	3.7×10 <sup>5</sup> ± 8.4×10⁴	3.1x10 <sup>4</sup> ± 6.1x10 <sup>3</sup>	2.2×10 <sup>4</sup> ± 6.1×10 <sup>3</sup>	7.4x10² ± 7.4x10	34.33 ± 7.13	
Salted sardine Min.	Min.	1.7x10 <sup>4</sup>	1.8x10²	1.3×10²	1.0x10²	1.0	$\neg$
	Max.	4.7×10 <sup>5</sup>	6.1x10 <sup>3</sup>	6.2x10 <sup>3</sup>	3.3×10²	22.0	
	Mean	1.1×10 <sup>5</sup> ± 3.4×10 <sup>4</sup>	1.7x10³ ± 5.1x10²	1.5×10³ ± 4.9×10²	1.6x10 <sup>2</sup> ± 1.8x10	9.47 ± 1.74	
n= number of the examined samples	the examin	ed samples					

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# المخاطر الصحية المصاحبة للأسماك المملحة في السوق المصرى جهاد فتحي احمد فتح الباب

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أجريت هذه الدراسة على عدد ٣٠ عينة عشوائية من الأسماك المملحة (١٥ عينة فسيخ و١٥ عينة سردين مملح)، تم تجمعيها من عدة سوير ماركت مختلفة في محسافظتى القاهرة و الجيزة وذلك لاستبيان الحالة الصحية لهذه الأسماك وتأثيرها على صحة المستهلك.

وقد خضعت العينات للفحوص الظاهرية، الكيميائية، البكتريولوجية وأظهرت النتائج ما يلى: العينات مقبولة ظاهريا ولم يظهر عليها علامات فساد .

بالفحص الكيميائي وجد ان الحد الاقصى لنمبة الاس الايدروجيني هــو (٧ و ٦,٩) لعينــات الفسيخ و السردين المملح على التوالي وانها أعلى من الحدود المسموح بها طبقا للمواصــفة القياســية المصرية لسنة ١٩٩٦.

كان متوسط نسبة الرطوبة ٩,٥ % و ٩,٠ 0% للفسيخ و السردين المملح على التوالى وانها أعلى من الحدود المسموح بها طبقا للمواصفة القياسية المصرية لسنة ١٩٩٦.

بالإضافة إلى ذلك كان الحد الاقصى لنسبة ملح الطعام ٣٣% و ٢٥% الفسيخ و السيردين الممليح على التوالى وللهستامين والنيتروجين الكلى المتصاعد كانت النسبة (٣٠و ٣٥% مجم /١٠٠ جم) و (٥٠ و ٤٠ مجم /١٠٠ جم) للفسيخ و السردين المملح على التوالى .

وجد ان قيم ملح الطعام والهستامين كانت أعلى من الحدود المسموح بهـــا طبقـــا للمواصــــفة القياسية المصرية لسنة ١٩٩٦. وكانت قيم النيتروجين الكلى المتصاعد أعلى من الحدود المسموح بهــــا للأسماك الطازجة (٣٠ مجم/١٠٠ جم) طبقا للمواصفة القياسية المصرية لسنة ٢٠٠٣.

أوضحت الفحوص البكتريولوجية ان متوسط العد البكتيرى الكلسى للميكروبات اللاهوائية، الميكروب العنقودى الذهبى و الميكروبات المعوية والقولونية هو (۲٫۱×،۱ ، ۱۰×۱۰ ، خلية /جسم) (۲۰×۲۰ ، ۲۱۰×۱۰ خلية /جسم) و (۲۰×۲۰ خلية /جسم) العنيات الفسيخ و السردين المملح على التوالى. وكانت القيم أعلى من الحدود المسموح بها طبقا للمواصفة القياسية المصرية لسنة ١٩٩١. والخلاصة أن عملية التمليح يجب أن تتم تحت ظروف صحية كاملة و ذلك حفاظا على صحة المستهلك.