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Microbiological Status of Egyptian Prawn

M.F. Sedik[1], Sawsan A. Roushdy[1], F.A. Khalafalla[2] and Hoda A. El G. Awad[3]

1 Department of Food Hygiene, Faculty of Vet. Med., Cairo University, Giza, Egypt.

2 Department of Food Hygiene, Faculty of Vet. Med., Beni Suef, Egypt.

3 Fish Unit, Animal Health Research Institute, Dokki, Giza, Egypt.

ABSTRACT :

A total of 100 prawn individuals were collected from different markets at Cairo and Giza Governorates. The average counts of aerobes, psychrophiles, enterobacteriaceae, coliforms and staphylococci were $5x10^3$, $3x10^2$, $< 2x10^2$, $<3x10^2$ and $2x10^2$ organisms per gram newly caught prawn sample respectively. Such counts were increased to $7x10^8$, $4x10^5$, 10^8, 20 and $2x10^5$ organisms per gram in samples of unfrozen prawn in shell respectively, reaching its maximum at 10^9, $2x10^7$, $4x10^5$, $2x10^3$ and $8x10^3$ organisms per gram in unfrozen peeled prawn, respectively. In frozen prawn samples (both peeled or in shell) the average counts were lower as compared with unfrozen ones and reached $2x10^7$, $2x10^5$, 10^3, 40 and $2x10^4$ organisms per gram in frozen prawn in shell respectively while $2x10^7$, $2x10^5$, 10^3, 60 and $7x10^3$ organisms per gram in frozen peeled prawn, respectively.

Arizona group, Escherichia coli, Enterobacter group, Proteus group, Providencia group A&B, Shigella group and Staphylococcus aureus were isolated from examined samples with variable percentages. The public health significance of isolated organisms was discussed.

Key words : Microbiological status, Egyptian prawn, Enterobacteriaceae, Psychrophiles, Staphylococci.

INTRODUCTION

Prawn has a highly palatable and digestible quality among consumers all over the world. In the past, problems of handling fresh, iced and frozen prawn and the need for ideal methods of handling and storage of prawn have been emphasized. Several surveys on bacteriological quality of fresh and frozen shrimp have been made (Green, 1949; Surkiewicz et al., 1967; Vanderzant et al. 1973; Zuberi et al., 1985). Moreover, many investigators (Vanderzant et al., 1973; Summer et al., 1982 and Zuberi et al., 1983) explained the importance of processing stages on bacterial load of shrimp. There are extensive literatures on the public health aspects of shellfish bacteriology and the significance of enteric pathogens as cause of food poisoning in man during consumption of fish and fishery products (Shewan, 1962 and Greenwood et al. 1985). Microbial activity is one of the main causes of quality deterioration of prawn (Cobb and Vanderzant, 1970).

The present investigation was carried out to study the bacteriological status of newly caught prawn as well as those collected from markets at Cairo and Giza Governorates, either unfrozen or frozen (peeled or in shell).

MATERIALS AND METHODS

Collection of samples

A total of 110 prawn individuals were collected as follows :

1. Ten prawn samples directly caught from the Red Sea.
2. Twenty five samples directly caught from Lake Karoun.
3. Twenty five raw peeled samples.
4. Twenty five frozen peeled samples.
5. Twenty five frozen samples in shell.

Samples of the 3rd, 4th, and 5th groups were collected from markets at Cairo and Giza. All collected samples were organoleptically acceptable. Such samples were transferred

to the laboratory with minimum of delay to be examined bacteriologically.

Preparation of samples

Samples in shell were beheaded and the shell removed under aseptic conditions. From the muscles of each prawn sample, 10 grams were cut under aseptic conditions into small pieces, then homogenized in 90 ml of 0.1% peptone water using a sterile electrical belender. Ten-fold serial dilutions up to 10^{-6} were prepared from the original dilution (ICMSF, 1974).

Bacteriological examination

1. Aerobic plate count (APC) : The drop plate method recommended by ICMSF (1978) was used. Inoculated plates were incubated for 3 days at 25 °C for enumeration of mesophilic count and for 10 days at 0 °C for enumeration of psychrophilic count.

2. Total Enterobacteriaceae count : The technique recommended by Gork (1976) was applied by using Violet Red Bile Glucose agar (VRBG agar). Inoculated plates were incubated at 37 °C for 24 hrs. Representative colonies were isolated and tested for Gram reaction. The isolates were identified biochemically according to the technique recommended by Finegold and Martin (1982). Salmonellae were typed serologically according to Kauffmann-White schem (Kauffmann, 1974).

3. Most probable number of coliforms (MPN) : Presumptive and confirmed coliforms, fecal coliforms and E. coli were determined according to the 3-tube most probable number procedure recommended by ICMSF (1974).

4. Isolation and Identification of E. coli : A loopful from the positive lauryl sulphate tryptose tube was streaked over Eosin Methylene Blue agar (EMB agar). Inoculated plates were incubated at 37 °C for 24 hours. Typical colonies were tested for IMVIC reactions. The isolates were identified

serologically by using diagnostic sera (Wellcome E. coli agglutinating sera for diagnosis of enteropathogenic types).

5. Staphylococci count : Plates of Baird-Parker's agar were inoculated and incubated at 37 °C for 24 hrs. Suspected colonies were subjected to Gram stain reaction. Isolates were tested by mannitol fermentation (Bailley and Scott, 1974), catalase test (Mac-Faddin 1976) and coagulase test (Gruickshank et al., 1969).

6. Virio parahaemolyticus : The technique adopted was that recommended by Thatcher and Clark (1975), using Thiosulphate Citrate Bile Sucrose agar (TCBS agar). Inoculated plates were incubated at 37 °C for 24 hrs.

RESULTS AND DISCUSSION

From the data obtained (Table 1), it can be concluded that the average aerobic plate counts per gram at 25 °C and 0 °C in newly caught prawn were $5x10^3$ and $3x10^2$ organisms, respectively. Such counts were increased to $7x10^8$ and $4x10^5$ organisms/gram in samples of unfrozen prawn in shell collected from markets, reaching its maximum at 10^9 and $2x10^7$ organisms/gram in unfrozen peeled prawn, respectively. In frozen prawn samples (both peeled or in shell) the average counts were lower as compared with unfrozen ones and reached $2x10^7$ at 25 °C and $2x10^5$ at 0 °C. The average counts of Entero-bacteriaceae and coliforms were $< 2x10^2$ and <3 organisms/gram newly caught prawn, respectively, increasing in unfrozen prawn in shell to 10^8 and 20, and in peeled samples to $4x10^5$ and $4x10^3$ organisms, respectively. In frozen prawn in-shell samples, such average counts were 10^3 and 40 while in frozen peeled samples were 10^5 and 60 per gram, respectively. Staphylococci count of newly caught prawn was less than 10^2 while in unfrozen in-shell prawn and unfrozen peeled prawn they were $2x10^4$ and $8x10^3$ organisms per gram, respectively. Nearly similar results were recorded for frozen in-shell and peeled prawn, each constituting $2x10^4$ and $7x10^3$ organisms per gram, respectively.

Table 1. Summary of data for the bacteriological examination of prawn.

	Type of prawn	Bacterial Count														
		APC* 25°C			APC* 0°C			Enterobacter. count			Coliforms (MPN)			Staphyl. count		
		Min.	max.	mean	min.	max.	mean	min.	max.	mean	min.	max.	mean	min.	max.	mean
1	Newly caught prawn	$2x10^2$	$3x10^4$	$5x10^3$	$2x10^2$	$2x10^3$	$3x10^2$	$<2x10^2$	$<2x10^2$	$<2x10^2$	<3	<3	< 3	$<2x10^2$	$<2x10^2$	$<2x10^2$
2	Unfrozen in shell	$3x10^5$	$9x10^9$	$7x10^8$	10^3	$5x10^6$	$4x10^5$	$4x10^3$	$2x10^9$	10^6	<3	40	20	$4x10^2$	$2x10^5$	$2x10^5$
3	Unfrozen peeled	$3x10^7$	$8x10^9$	10^9	$2x10^4$	$2x10^8$	$2x10^7$	$2x10^3$	$5x10^6$	$4x10^5$	70	10^4	$2x10^3$	$3x10^2$	$8x10^4$	$8x10^3$
4	Frozen in shell	$4x10^2$	$4x10^8$	$2x10^7$	$2x10^2$	$3x10^6$	$2x10^5$	$2x10^2$	$8x10^3$	10^3	<3	$4x10^2$	40	10^2	$2x10^5$	$2x10^4$
5	Frozen peeled	$4x10^4$	10^8	$2x10^7$	$2x10^3$	$9x10^5$	$2x10^5$	$2x10^2$	$3x10^6$	10^3	<3	10^3	60	$2x10^3$	$7x10^4$	$7x10^3$

* APC = Aerobic plate count (organisms per gram).

Generally, shellfish have low bacterial counts when freshly caught, however, the numbers of bacteria increase significantly during handling and distribution until sold in the markets and this substantiates the findings reported in the present investigation and also may explain the problem of handling discussed by many authors (Green, 1949; Surkiewicz et al., 1967 and Vanderzant et al., 1970).

Bacterial counts in all unfrozen peeled prawn samples in this investigation exceeded the limit of the International Commission on Microbiological Specifications for Foods (ICMSF), 1974 (10^6 organisms per gram). This could be attributed to the bad sanitary conditions under which such samples were peeled (hands of workers, containers, even cleaning and temperature of surrounding atmosphere) and this substantiates the findings reported by Surkiewicz et al., (1967) and Vanderzant et al. (1970).

The lower bacterial counts in frozen prawn samples collected from markets as compared with those of unfrozen samples could be attributed to the effect of freezing as it can destroy or lethally injure bacterial cells (Kereluk and Gunderson, 1959 and Vanderzant et al., 1973).

Realizing that mesophiles as well as psychrotrophes can grow at 25 ^{o}C, counts at such degree of incubation were expected. Moreover, owing to the climatic condition of Egypt it is more likely to have higher counts of mesophiles than psychrophiles. Such finding was supported by Shewan (1962).

Table 2 illustrates the incidences of isolated organisms. It can be concluded that Arizona group, Escherichia coli, Enterobacter aerogenes, Providencia group A, Shigella bodyii, Shigella flexneri and Staphylococcus aureus were present in unfrozen in-shell, unfrozen peeled prawn, frozen in-shell and frozen peeled prawn samples at variable percentages. From samples of the unfrozen peeled, frozen in-shell and frozen peeled prawn samples Enterobacter agglomerans and Proteus mirabilis were isolated while Hafnia group could be isolated from unfrozen in-shell prawn samples only. Moreover, Proteus rettigeri and Proteus

Table 2. Frequency distribution of isolated organisms from prawn

	Isolated organisms	Newly caught		Unfrozen in-shell		Unfrozen peeled		Frozen in-shell		Frozen peeled	
		No.	%	No	%	No	%	No	%	No	%
1	Arizona group	-	-	1	4	1	4	2	8	2	8
2	E. coli	-	-	6	24	11	44	2	8	3	12
	a) O_{127} : K_{63} B :			1	4	2	8	1	4	2	8
	b) O_{112} : K_{66} B :	-				-	-	-	-	1	4
3	Enterobacter aerogenes	-	-	4			12	3	12	5	20
4	Enterobacter agglomerans	-	-	-	-	2		1	4	3	12
5	Hafnia group	-	-	1	4	-	-	-	-	-	-
6	Proteus morabilis	-	-	-	-	2	8	1	4	1	4
7	Proteus rettigri	-	-	4	16	3	12	-	-	2	8
8	Proteus vulgaris	-	-	2	8	4	16	-	-	2	8
9	Proteus morgani	-	-	1	4	-	-	-	-	-	-
10	Providancia group A	-	-	3	12	2	8	1	4	4	16
11	Providancia group B	-	-	2	8	-	-	-	-	3	12
12	Salmonella reading	-	-	-	-	2	8	-	-	-	-
13	Shigella boydii	-	-	1	4	1	4	2	8	1	4
14	Shigella flexneri	-	-	3	12	2	8	2	8	2	8
15	Staph. aureus	-	-	2	8	4	16	2	8	3	12

vulgaris were found to exist in unfrozen in-sheep, unfrozen peeled and frozen peeled prawn samples while Proteus morgeni was found in one of the raw prawn in shell samples and Salmonella in two unfrozen peeled samples. It was worth mentioning that in a number of samples, the prawn in its shell contained less organisms than when peeled (i.e. E. coli, Enterobacter agglomerans, Proteus morabilis and others). This was most probably due to the shells acting as a protective cover.

The presence of coliform bacteria in prawn is an undesirable occurrence and the possible presence of enteric pathogens may constitute public health hazard for human consumers (Frazier, 1967). In this respect, Arizona group have been encountered in cases of gastroentritis. Proteus species were found to be implicated in cases of summer diarrhoea in infants, sinusitis as well as urinary tract infections (Fraizer, 1967 and Shewan, 1962).

Escherichia coli serotypes recongnized are pathogenic for both human and warm blooded animals and responsible for colienteritis in children and colibacillosis in adults, also appendicitis, otitits and nephritis (Pyathin and Krivashein, 1980).

Shigellae are not indigenous in foods, however, they cause outbreaks of enterocolitis and have been transmitted through food and water contamination by human excreters (Hobbs, 1974). Salmonella infections play a prominent role in food poisoning (Shewan, 1962; Frazier, 1967 and ICMSF, 1978).

The presence of staphylococcus aureus in prawn indicates its contamination from polluted water in which it was caught or during handling in fishing vessels and in peeling process plants as described by Shewan (1962) and Lawson, (1970).

Failure to isolate Vibrio parahaemolyticus from unfrozen and frozen prawn samples, either in-shell or peeled, could be explained by the sensitivity of the organism to freezing and drying. Such conditions should be relied upon for the destruction of the organisms. This in agreement with

what has been reported by Johnson and Liston (1973), Liston (1974) and Beuchat (1975).

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الحالة الميكروبيولوجية للجمبري المصري

ملخص :

ان الجمبري من القشريات شهية الطعم ويفضله معظم المستهلكين الا انه معرض للفساد السريع لذا يجب تداوله واعداده تحت ظروف صحية سليمة . أجريت الدراسة البكتريولوجية على عدد ١١٠ عينه من الجمبري واتضح نتيجة الفحص أن اعداد الميكروبات الهوائية عند ٢٥ م والمحبة للبرودة والمحبة والقولونية والمكور العنقودي كانت ٥ * $١٠^{٣}$ ، ٣ * $١٠^{٢}$ ، ٢ * $١٠^{٢}$ ، ٣ ، ٢ * $١٠^{٢}$ ، في الجرام على التوالي في الجمبري فور اصطياده وتبين ارتفاع اعداد تلك الميكروبات الى ٧ * $١٠^{٨}$ ، ٤ * $١٠^{٥}$ ، $١٠^{٨}$ ، ٢٠ ، ٢ * $١٠^{٥}$ في الجرام من العينات الطازجة المعروضة بالأسواق على التوالي ووصلت تلك الإعداد الى أقصاها في العينات الطازجة المنزوعة الهيكل الخارجي ($١٠^{٩}$ ، ٢ * $١٠^{٧}$ ، ٤ * $١٠^{٥}$ ، ٢٠ * $١٠^{٣}$ ، ٨ * $١٠^{٣}$ في الجرام على التوالي) . أما في العينات المجمدة فكانت اعداد تلك الميكروبات ٢ * $١٠^{٧}$ ، ٢ * $١٠^{٥}$ ، $١٠^{٣}$ ، ٤٠ ، $١٠^{٣}$ في الجرام بينما في العينات المجمدة المنزوعة الهيكل الخارجي فكانت ٢ * $١٠^{٧}$ ، ٢ * $١٠^{٥}$ ، $١٠^{٣}$ ، ٦٠ ، ٧ * $١٠^{٣}$ في الجرام .

مجموعة الأريزونا والايشريشيا كولاي ومجموعة انيتروبكتير ومجموعة البروتيس ومجموعة البروفيد انشيا أ، ب ومجموعة الشيجلا والميكروب العنقودي السبحي . كما تم مناقشة الأهمية الصحية لتلك الميكروبات المعزولة .