BACTERIOLOGICAL QUALITY OF IMPORTED FROZEN CHICKEN IN SULAIMANI MARKETS

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Abstract

To study the bacteriological quality of imported frozen chicken meat in Sulaimani markets, a total of 360 samples represented whole chickens (160 samples, 80 for each batch), thighs (120 samples, 60 for each batch) and breasts (80 samples, 40 for each batch) belonged to seven commercial trademarks, distributed on two batches, were tested according to international analytical regulations and guides. The mean values of the total plate count (TPC) in batches 1 and 2 of the whole chicken carcasses ranged 0.540-5.720 $\times10^4$ and 0.250-5.720 $\times10^4$ CFU/ g meat respectively, in thighs they ranged 1.170- 17.520 and 0.730- 12.500 $\times10^4$CFU/ g meat respectively, and in breast they ranged 0.586- 14.380 and 1.174-11.900$\times10^4$CFU/ g meat respectively. The psychrophiles count in whole chicken carcasses ranged 1.950- 13.740$\times10^4$ and 0.900-27.420$\times10^4$ CFU/ g meat, in thighs they ranged 0.554-8.560 and 0.360- 8.000$\times10^4$ CFU/ g meat, and in breast ranged 0.734-8.880 and 1.140- 9.360 $\times10^4$ CFU/ g meat in batches 1 and 2 respectively. Psychrotrophes count in whole carcasses ranged 1.964- 14.280 and 1.286- 21.800$\times10^4$ CFU/ g meat, in thighs they ranged 1.560- 12.640 and 3.42- 7.680 $\times10^4$ CFU/ g meat, and in breast ranged 0.770- 13.140 and 1.178- 8.440 $\times10^4$ CFU/ g meat in batches 1 and 2 respectively. The most probable number (MPN) of both total coliforms and fecal coliforms in the two batches were less than 1 CFU/ g meat in all samples that inspected. Significant differences were recorded among all marks and between both batches for each mark including the three types of meat while all samples were *E. coli* O157: H7, coagulase positive staphylococci, and *Salmonella* free.

KEY WORDS: Bacterial quality, Standard plate count, Psychrophil count, *E. coli* O157:H7, *Staphylococcus, Salmonella.*

*** This research is extracted from a PhD dissertation of the first researcher.**

Introduction

Chicken and poultry products have become popular due to their specific sensory attributes and the increasing tendency of the public to consider white meat as being healthier compared to red meat (Geonaras *et al*., 1995). The presence of pathogenic microorganisms, spoilage microorganisms, or both in poultry is undesirable but unavoidable and lead chicken meat to be deteriorated in quality (Brown, 1982; Russell, 2001). Meat cannot be without microorganisms but their number can be increased by providing the optimum temperature, humidity, and oxygen leading to spoilage while the microbial growth will be static and limited at frozen temperatures (Inoue & Ishikawa, 1997; Nollet, 2007; Biswas *et al*., 2011). In Iraq, Zangana (2006) noticed that the total count of Psychrophilic bacteria in frozen chicken thigh were 4.70 – 4.28 log./ gm meat and the mean value of coliform count in frozen chicken thighs were ranged between 3.86- 4.86 log/ g meat. Alrubaei *et al*. (2007) found that the psychrophilic count of broiler chicken meat stored at 4C° for 0, 3, 6 days were 6.1, 6, 6.5 log / gm meat respectively while Al-Mosawy *et al.* (2008) found that the psychrophilic count in minced breast chicken meat stored at 4C°for 0, 3, 6 day were 6.0, 6.2, 6.5 log/ gm meat respectively. Poultry are highly susceptible to infection with *E. coli* 0_{157}:H_7 (Beery *et al*., 1985; Stavric, *et al*., 1993). Contamination of poultry carcasses with enterotoxigenic *Staphylococcus* isolates of human origin often occurs at processing (Adams & Mead, 1983). In the neighbor Saudi Arabia (KSI), Zeitoun, & Al-Eid (2003) mentioned that two of five marks of imported frozen chicken collected from Al-Hassa revealed no coagulase positive staphylococcal count whereas *Salmonella* was positive in some, while Al - Dughaym & Al-Tabari (2009) mentioned in their study on 10 samples of chicken thighs collected from Al-Ahssa market, that the total mean value of *Staphylococcus aureus* was less than 10^2 CFU/g meat. Foods from animal's origins (especially poultry) are important source of human *Salmonella* infections, especially those which may become contaminated during handling by an ill patient or carrier persons (Gast, 2003). In poultry the cross contamination

is also most likely to occur during evisceration procedures with the use of plucking machines, in which these mechanical devices often rupture the abdominal air sacs and pericardium of the affected birds (Herenda & Franco, 1996 and Lake *et al*., 2002). AL-Dughaym and Altabari (2009) in KSA ranked the samples from carcass cuts (chilled, frozen, fillet and thigh) to minced meat or further processed products as burger, nuggets, frankfurter and meat paste loaf and revealed *Salmonella arizona* was isolated at once from thigh samples. In Kurdistan region of Iraq, the demand for meat increased during the last decades (Iraqi poultry and product annual, 2010), this led poultry meat of different sources to be imported in high quantities. Without regarding the formal inspection may or may not be occur. So, the aim of this study was to assess the bacteriological load of imported frozen chicken meat trademarks available in Sulaimani markets in order to determine their quality for human consumption.

Materials and methods

Sampling: The ultimate inspection included a total number of (360) samples of frozen chicken meat belonged to 7 foreign trademarks were collected in two batches from different parts of Sulaimani city markets. The samples consisted of whole chickens (160 samples- 80 from each batch), thighs (120 samples- 60 from each batch) and breasts (80 samples- 40 each batch). The duration of sampling batch (1) was from the first of February till end of April 2010, while batch (2) was sampled from July till end of September 2010. The whole chicken carcasses were distributed on 40 samples for each of DMIS (Iranian), Gedik (Turkey), Sadia (Brazilian) and Frinal (Brazilian) while thighs were on 40 samples of Sadia (Brazilian), 40 samples of Tyson (American) and 40 samples of AJC (American), and the breasts were from Sadia (Brazilian), 40 samples of Seara (Brazilian). From each trademark (40) samples (whole carcasses, thighs or breasts) were collected and the whole carcass or a number of meat specimens from each sample were obtained aseptically. Both of AJC and Tyson marks were sold in unpacked condition at retail selling. All Samples were transferred inside a Cork chilled box (ice box) to the laboratory of Animal production department, Agriculture faculty, Sulaimani University, as soon as possible to prevent thawing.

-pH measurement

The procedure of **Naveena and Mendiratta (2001)** was used to determine the pH of the specimens

Microbiological tests

The procedure of USDA/ FSIS (1998) was used, aseptically, 50 ± 0.1 g of the sample (Major and minor pectoralis with femur muscles) were used.

Total plate count

The procedure of AOAC 966.23 C (1995) was used.

Psychrophilic bacterial count (APHA, 1992)

The same procedure for counting was used as in Total Plate count; 1ml of the series dilutions, then the plates were incubated at 5-7ºC for 10 days.

Psychrotrophic bacterial count (AOAC, 1995)

The same protocol of pour plate procedure was used, plates were incubated at 20ºC for 4-5 days.

Most Probable Number (MPN) of Coliforms (USDA/FSIS, 1998; Garthright, 1995)

The procedures of 3 set tubes were used. To count the MPN of the fecal source coliforms, a loopful from the positive tubes was inoculated into tubes with lactose broth and incubated at 45.5 ± 0.05°C in covered water bath for 24 ± 2h. The positive Coliform tubes were having acid (change the colour of medium to yellow) and gas. The number of the positive tubes that incubated at 35ºC was considered in the calculation of MPN of total coliforms, while that positive at 45.5ºC was considered to calculate the fecal source (thermotolerant) coliforms. MPN/g of meat was calculated by comparing the positive tubes against standardized MPN tables (USDA/FSIS Laboratory guide book, 2008). The highest lactose positive dilution tubes were confirmed to contain fecal source Coliforms (*Escherichia coli*) (APHA, 1992) by streaking onto Eosin–Methylene Blue agar (EMB) plates, after incubation at 37°C for 24 hours, the plates were examined for typical *E. coli* colonies, which are metallic green sheen. A number (2-3) of isolated colonies were picked out and purified on EMB, then subculture to Nutrient agar slants and incubated at 35ºC for 18 -24 hours to be tested completely. The completed test was achieved by sending the nutrient agar slants to be identified at the species level as *E. coli* through an automated Vitek- 2 system at the Central Health Laboratory, Erbil city, after a

gram stain was done for the slants; those gram negative rods were send.

Identification of *E. coli* O157:H7: Those *E. coli* detected by MPN were completely identified by Vitek 2system which identifies the enteropathogenic strain *E. coli* O157; H7 by development of agglutination with a normal saline suspended colonies.

Identification of Coagulase positive Staphylococci (*Staphylococcus aureus*) (Bennet & Lancette, 1998)

Performed by using Baired-Parker medium (Prondisa, Spain), and also by using Mannitol salt agar, which was prepared according to manufacturer directions (APHA, 1992). Coagulase test was achieved according to Collins & Lyne (1987). The black colonies were surrounded by a clear zone on Baired-parker agar, yellow mannitol fermentative on mannitol salt agar, and clotted the rabbit blood plasma within four hours were presumptively identified as coagulase positive *Staphylococcus aureus,* but for the purpose of complete identification, a number of colonies that showed positive and negative results were purified by streaking on mannitol salt agar and incubated for 24h at 37◦C, and then isolated colonies were sub cultured onto Nutrient agar slants, incubated at the same conditions and were sent to be completely identified by Vitek2 system in Erbil city to be identified at the species level.

Identification of *Salmonella*

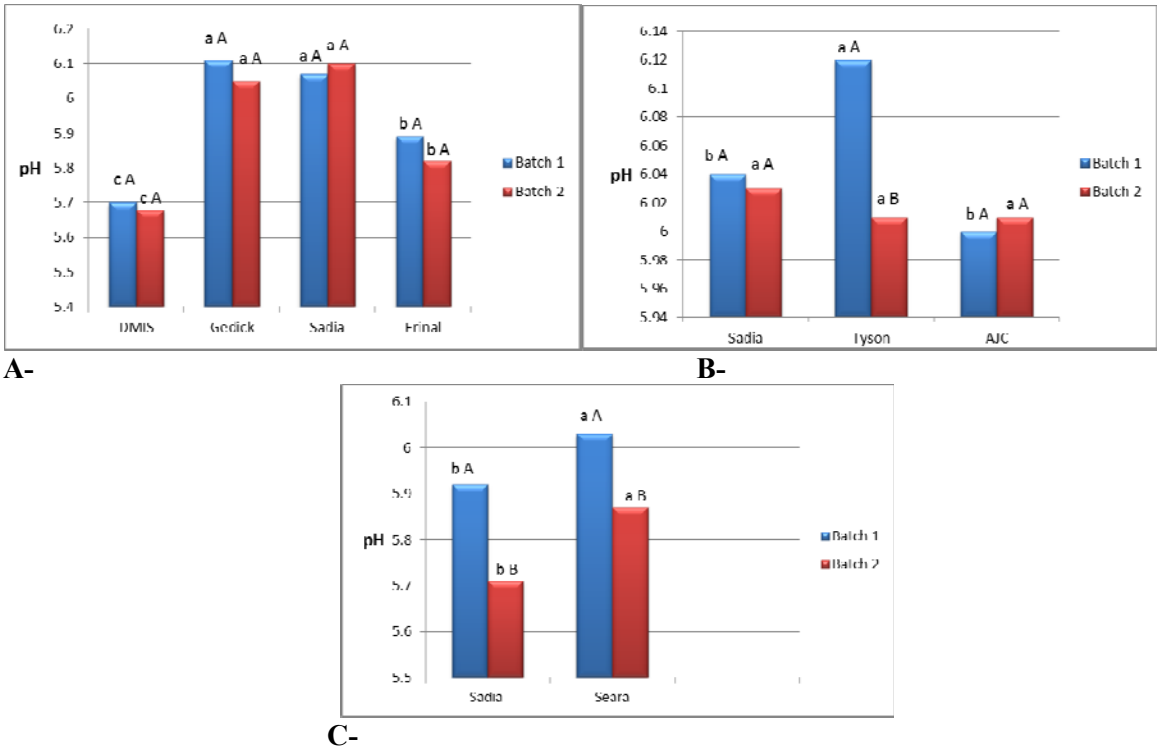
The procedure of Waltman (1999) and Talaska (2004) were used. Three to five suspected colonies were inoculated separately into TSI (Triple Sugar Iron) agar (Merck, Germany) and Urea agar (Biolofe, Italy) and incubated at 35°C for 24 hours, the colonies with characteristics reaction of *Salmonella* were subcultured in duplicates on Nutrient agar slants, incubated at 37°C for 24 hrs and stained by gram staining. The duplicates of the gram negative ones were transferred to the Central Health Laboratory (Erbil) to complete identification by Vitek2 system and to the *Salmonella* National Center in Baghdad city as well. The isolates that gave negative Urea test, and TSI test gave Acid, gas (Yellow, pink, black with gas formation), the biochemical tests of automated Vitek2 system, and the results obtained from Salmonella National Center, all were confirmative for the presence of *Salmonella* spp. in the samples.

Results

pH values: Figure (1) showed the pH mean values of two batches for all inspected marks. In whole carcasses the values ranged 5.70- 6.11 for batch (1) and 5.68- 6.10 for batch (2). Gedik and Sadia did not significantly differ in both batches, while these marks were significantly difference ($p< 0.05$) for both Frinal and DMIS which in turn were different ($p< 0.05$) in between. Also, there were no significant differences between the two batches for each mark. The pH mean values of two batches of chicken thighs ranged 6.00-6.12 for batch (1) and 6.01-6.03 for batch (2). Tyson was significantly different ($p< 0.05$) from the other two marks. All thigh marks showed no significant difference between two batches except for Tyson. The same figure showed the pH mean values of two breast trademarks which ranged 5.92-6.03 and 5.71-5.87 for batch(1 and 2) respectively. Both marks revealed significant differences ($p< 0.05$) in both batches. Also, both batches were significant difference ($p< 0.05$) for each mark.

Bacterial count: The mean values of total bacterial counts of the whole carcasses of two batches for the four trademarks are shown in table (1), the mean values of standard plate count showed significant differences ($p< 0.05$) among all marks for both batches, also, there was a significant difference ($p<0.05$) between the two batches for each mark. All trademarks are within the standard levels of Iraqi quality regulations for frozen poultry meat (ICOSQC- IQS 2270/4, 2006) that specified the SPC of frozen poultry between 10^5- 10^7 CFU/g meat. Mean values of total plate count for the thighs in two batches of four trademarks are shown in table (2). The mean values for all marks were significantly different in both batches ($p< 0.05$). There were significant differences between both batches for each mark ($p< 0.05$), so, all trademarks matched standard levels in Iraqi quality regulations (ICOSQC IQS 2270/4, 2006). The mean values of total bacterial counts of the breasts for two batches of two trademarks are shown in table (3). The mean values for both marks were significantly different ($p< 0.05$) in each batch. There was no significant difference between the two batches for Seara, while for Sadia there was ($p< 0.05$). All trademarks matched the standard levels of Iraqi quality regulations for frozen poultry meat (ICOSQC /IQS 2270/4, 2006).

The psychrophilic bacterial count mean values of the carcasses of two batches for four trademarks are shown in table (1). The mean values showed that there was no significant difference between the two batches of both Gedik and Frinal while there was significant difference ($p< 0.05$) between the two batches for both of DMIS and Sadia. The mean values of psychrophilic bacterial count of the thighs are shown in table (2).The results revealed that all marks, in both batches (1, 2), appeared significantly different ($p< 0.05$). There were significant differences between the two batches for each mark. The mean values of psychrophilic bacterial count of frozen breast are shown in table (3). The results showed the presence of significant difference ($p< 0.05$) between the two marks in both batches. The mean values of psychrotrophic bacterial count of whole carcasses are shown in table (1). There were significant differences ($p< 0.05$) between the two batches for both of DMIS and Sadia while both of Frinal and Gedik were not different significantly. The mean values of psychrotrophic bacteria count of the thighs are shown in table (2). All marks significantly differed ($p< 0.05$) in both batches. Also, there was a significant difference ($p< 0.05$) between two batches for each mark except for AJC mark which showed no difference. The mean values of psychrotrophic in the breasts are shown in table (3). There was a significant difference ($p< 0.05$) between the two marks for both batches. Also, there was significant difference ($p< 0.05$) between the two batches for each mark.



A- **B-**

C-

Figure (1): pH mean values of all chicken trademarks: A-Whole chicken carcasses. B- Chicken thighs. C- Chicken breasts.

-Means having different lower-case at the same row (Trademarks) and upper – case at the same column (Batch) are significantly different.at ($p < 0.05$).

The mean values of most probable number (MPN) of total coliform count of whole carcasses are shown in table (1). There were no significant differences among all marks except for Gedik in both batches, also, there was no significant difference between the two batches for each mark. While the MPN of total fecal coliforms of whole carcasses for two batches of four trademarks are shown in table (2). Batch (1) showed that all marks were not significantly different except for Gedik, batch (2) showed that both DMIS and Frinal were significant difference ($p< 0.05$) with Gedik and Sadia which were not varied in between. *E. coli* were isolated from both batches of Gedik and Sadia carcasses (2). The microbiological analysis results for whole chicken carcasses matched standard levels of Iraqi quality regulations for frozen poultry meat (ICOSQC IQS 2270/4, 2006).

Table (2) showed the MPN of total coliforms mean values of chicken thighs and there were no significant differences ($p< 0.05$) among all marks for both batches. Also, there was no significant difference between the two batches for each mark. The MPN of total fecal coliforms of chicken thighs are shown in table (2). There were no significant differences among all marks in both batches and no significant difference between two batches for each mark.

The mean values of MPN total Coliform count of the breasts are shown in table (3), there was a significant difference ($p< 0.05$) between the two marks in both batches. There was no significant difference between the two batches for each mark. There were no significant differences between two marks for each batch. *E. coli* and their enteropathogenic variant O_{157}:H_7 were not detected in breasts of all trademarks inspected. There were no significant differences with respect to MPN of coliforms among the four inspected marks for whole carcasses except for Gedik (table, 1), which was higher than other marks.

Table (1): Bacterial counts mean values of imported whole frozen chicken carcasses for two batches of four trademarks in (CFU/ g meat).

Traits	Bat-ch	Trade mark			
		DMIS	Gedik	Sadia	Frinal
Total plate count	1	$0.54 \times 10^4 \pm 0.24$ d A	$5.72 \times 10^4 \pm 0.22$ a A	$3.18 \times 10^4 \pm 0.07$ c B	$3.90 \times 10^4 \pm 0.26$ b B
	2	$0.26 \times 10^4 \pm 0.16$ d B	$3.42 \times 10^4 \pm 0.15$ c B	$4.72 \times 10^4 \pm 0.53$ b A	$5.72 \times 10^4 \pm 0.23$ a A
PSP. bacterial count	1	$1.95 \times 10^4 \pm 0.34$ c A	$13.74 \times 10^4 \pm 1.58$ a A	$7.60 \times 10^4 \pm 0.64$ b B	$12.16 \times 10^4 \pm 1.68$ a A
	2	$0.90 \times 10^4 \pm 1.33$ c B	$10.11 \times 10^4 \pm 0.55$ b A	$27.42 \times 10^4 \pm 3.97$ a A	$15.02 \times 10^4 \pm 0.83$ b A
PST. bacterial count	1	$1.96 \times 10^4 \pm 0.73$ b A	$14.28 \times 10^4 \pm 1.11$ a A	$10.54 \times 10^4 \pm 0.94$ a B	$9.50 \times 10^4 \pm 2.67$ a A
	2	$1.28 \times 10^4 \pm 1.16$ c B	$11.62 \times 10^4 \pm 0.66$ b A	$21.80 \times 10^4 \pm 2.67$ a A	$14.22 \times 10^4 \pm 1.56$ b A
MPN of total Coliforms	1	< 0.03 ± 0.0 b A	0.258 ± 0.08 a A	0.030 ± 0.0 b A	< 0.03 ± 0.0 b A
	2	<0.03 ± 0.0 b A	0.132 ± 0.04 a A	0.030 ± 0.0 b A	< 0.03 ± 0.00 b A
MPN of total fecal coliform	1	< 0.03 ± 0.0 b A	0.101 ± 0.09 a A	0.030 ± 0.0 b B	< 0.03 ± 0.0 b A
	2	< 0.03 ± 0.0 c A	0.101 ± 0.09 a A	0.074 ± 0.0 b A	< 0.03 ± 0.0 c A
E.coli	1	ND	ND	ND	ND

(O_{157} :H_7)	2	ND	ND	ND	ND
Coa. pos. *St. aureus*	1	ND	ND	ND	ND
	2	ND	ND	ND	ND
Salmonella Spp.	1	ND	ND	ND	ND
	2	ND	ND	ND	ND

- Means having different lower-case at the same row (Trademarks) and upper – case at the same column (Batch) are significantly different at ($p < 0.05$).
- ND : Not detected. - The symbol (<) was not included in statistical analysis.

Table (2): Bacterial counts mean values of imported frozen chicken thighs for two batches of three trademarks (CFU / g meat)

Traits CFU/g meat	Batch	Trade mark		
		Sadia	Tyson	AJC
Total plate count	1	3.0×10^4 ± 0.53 b B	17.52×10^4 ± 0.29 a A	1.17×10^4 ± 0.36 c A
	2	5.82×10^4 ± 0.22 b A	12.50×10^4 ± 0.70 a B	0.73×10^4 ± 0.35 c B
Psychrophilic bacterial count	1	4.62×10^4 ± 0.51 b B	8.56×10^4 ± 0.31 a A	0.55×10^4 ± 0.46 c A
	2	8.00×10^4 ± 0.39 a A	6.28×10^4 ± 0.30 b B	0.36×10^4 ± 0.58 c B
Psychrotrophic bacterial count	1	1.56×10^4 ± 0.31 b B	12.64×10^4 ± 0.29 a A	6.46×10^4 ± 0.78 c A
	2	3.42×10^4 ± 0.18 b A	7.68×10^4 ± 1.22 a B	4.82×10^4 ± 0.21 c A
MPN of total coliforms	1	0.03 ± 0.0 a A	0.03 ± 0.0 a A	< 0.03 ± 0.0 a A
	2	0.03 ± 0.0 a A	0.03 ± 0.0 a A	< 0.03 ± 0.0 a A
MPN of total fecal coliforms	1	0.03 ± 0.0 a A	< 0.03 ± 0.0 a A	< 0.03 ± 0.0 a A
	2	0.09 ± 0.0 a A	0.03 ± 0.0 a A	< 0.03 ± 0.0 a A
E.coli (O_{157} H_7)	1	ND	ND	ND
	2	ND	ND	ND
Coa. Pos. *St. aureus*	1	ND	ND	ND
	2	ND	ND	ND
Salmonella Spp.	1	ND	ND	ND
	2	ND	ND	ND

Means having different lower-case at the same row (Trademarks) and upper- case at the same column (Batch) are significantly different at ($p < 0.05$).
ND: Not detected. The symbol (<) was not included in statistical analysis.

Table (3): Bacterial counts mean values of imported frozen chicken breast for two batches of two trademarks(CFU / g meat).

Traits	Batch	Trade mark	
		Seara	Sadia
Total plate count	1	$14.38 \times 10^4 \pm 1.20$ a A	$0.586 \times 10^4 \pm 0.56$ b B
	2	$11.90 \times 10^4 \pm 0.99$ a A	$1.174 \times 10^4 \pm 0.53$ b A
Psychrophilic bacterial count	1	$8.88 \times 10^4 \pm 0.67$ a A	$0.734 \times 10^4 \pm 0.74$ b B
	2	$9.36 \times 10^4 \pm 0.59$ a A	$1.140 \times 10^4 \pm 0.62$ b A
Psychrotrophic bacterial count	1	$13.14 \times 10^4 \pm 0.36$ a A	$0.77 \times 10^4 \pm 1.36$ b B
	2	$8.44 \times 10^4 \pm 1.04$ a B	$1.178 \times 10^4 \pm 0.13$ b A
MPN total Coliforms count	1	0.030 ± 0.0 a A	4.58 ± 0.26 a A
	2	0.030 ± 0.0 a A	2.44 ± 0.21 a A
MPN total fecal coliforms	1	< 0.030 ± 0.0 a A	0.03 ± 0.0 a A
	2	< 0.030± 0.0 a A	0.03 ± 0.0 a A
E.coli (O_{157} :H_7)	1	ND	ND
	2	ND	ND
Coa. Pos. *St. aureus*	1	ND	ND
	2	ND	ND
Salmonella Spp.	1	ND	ND
	2	ND	ND

Means having different lower-case at the same row (Trademarks) and upper-case at the same column (Batch) are significantly different at ($p < 0.05$).
ND: Not detected. The symbol (<) was not included in statistical analysis

Discussion

The differences in pH mean values among the whole carcasses (except for both Gedik and Sadia marks), could be due to the stress factors before slaughtering which resulted in drop of glycogen content in meat and less lactic acid formation (Berri, *et al*., 2005), or exhaustion of the bird before slaughtering (Owens & Sam, 2000). The decrease in pH value could be due to liberation of free fatty acids as a result of the lipolytic enzymes, either because of prolonged storage period, or bad storage conditions (Frazier & Westhoff, 1988 and Sams, 2001) which may explain why DMIS mark gave a low pH value. The study results were similar to results of Ali & Al-Zahran (2010) who recorded the pH values in chicken carcasses stored at chilling conditions for 0, 2, 4 and 6 days as 6.00, 6.21, 6.80 and 7.20 respectively. The same explanations above are true to elucidate the differences in pH values in inspected thighs and breasts. The pH values obtained in this study were in agreement with those obtained by Mead (2000) who stated that leg and breast muscles of chicken have pH

values range of (6.1–6.4) and (5.6-5.8) respectively, whilst they were lower than those recorded Al- Hamadany (2009) who recorded the range of pH values in imported frozen chicken thighs sold in Baghdad's markets as 6.60 - 6.90.Unfortunately, there is no related standard for pH in Iraqi quality regulations to be compared with.

Although there were significant differences in total plate count among the inspected carcasses from the collected marks, Gedik had the highest count while DMIS had the lowest. This could be due to many reasons,

the low pH value for DMIS (figure, 1) may had an influence on bacterial growth (Jay *et al.,* 2005), other factors occur through poultry manufacturing processing like slaughtering, eviscerating, scalding, chilling, packaging, freezing, transporting and handling (Bolder, 1998). Also, as being mentioned by ICMSF (2011) that the rate of spoilage is influenced by many factors such as, storage temperature, initial number and type of microorganisms when packaged, type of packaging and chemical composition. The same table revealed significant differences in count between two batches of both Sadia and Frinal marks. The count in batch (2) appeared little higher than batch (1), this may be due to the effect of season; in summer the electric current cut off affects the thawing and refreezing of the products which in turn lead to increase in count as mentioned by Narasimha *et al*. (1998), while DMIS and Gedik appeared not to be affected seasonally because they have been stored in different freezing conditions as they were less demanded by consumers than the other two marks. Indeed, the freezing process inhibits mesophilic bacterial growth (Jay *et al*., 2005). Bailey *et al*. (2000) recorded higher bacterial count than the current study as they reported the mean total plate count in chicken carcasses stored in 4 ºC for 7 days, for 0 till 7 days as 4.6 to 6.6 log CFU/ g meat respectively while their results remained the same when the samples stored by chilling to -18 ºC. Morshedy & Sallam (2009) recorded the total plate count in chicken carcasses stored at 4ºC for 0, 4, 6, days as 5.13, 6.87, 7.4 $\log^{10}$ CFU/g meat respectively. It should be kept in mind that the above two articles used fresh meat but not frozen. Freezing has lethal effect on some microorganisms by formation of ice crystals (Kuntz, 1996) and as a result the count decreased.

The highest psychrophilic count in batch (1) of Frinal and batch (1) of Gedik mark, batch (2) of the Brazilian Sadia, could be due to many reasons, the different manufacturing steps (slaughtering, scalding, defeathering, chilling, handling, packaging) all these factors lead to adding more bacterial count (Bolder, 1998), and the initial bacterial count had an effect on the final count (ICSMF, 2011). Mead (2004) and Jay *et al*. (2005) had mentioned increasing the psychrophilic count within long storage at low temperature. The significant difference ($p< 0.05$) in psychrophilic count between two batches of DMIS and Sadia marks may be due to the seasonal effect on the products, in appropriate storage temperature or fluctuation of storage temperature are the most causes of spoilage (Russell, 2009). The same table showed that the mean value of psychrophilic bacterial count of all inspected marks were higher than total plate count (mesophilic bacteria), this may approve that the increase of count might be due to the contamination of freezers or the storage area with that kind of microorganisms specially *Pseudomonas* spp. or due to the thawing and refreezing processes through processes of marketing handling etc., which lead to rapid spoilage of the products during thawing (Russell, 2001 and Jay *et al*., 2005).

In table (2) which explained the mean value of psychrophilic count in thighs the results showed that AJC mark had the lower psychrophiles count in both batches, this could be due to good manufacturing practices which has a role in decreasing the count (Bolder, 1998 and Mead, 2004) which may be an advantage for those marks with low psychrophilic count. The results obtained here were little lower than those recorded by Al-Hasnawi (2010) who evaluated the quality of thighs meat of imported frozen chicken in Diwaniya city; she claimed that the psychrophilic bacterial count for Tyson thighs as 55.6×10^4 CFU/g meat while for Sadia thighs as 10.6×10^5 CFU/g meat. The current results were also lower than results published by Al-Hamadany (2009) who recorded psychrophilic bacterial count mean value in Sadia chicken thighs collected from Baghdad markets were 10.6×10^5 CFU/g meat, when the average count of other six imported and local marks ranged between 3×10^2 and 2×10^6 CFU/g meat.

The results related with psychrophilic bacterial count mean values of the breasts samples (table, 3) which revealed a rather high count in Seara mark in comparison to Sadia mark for both batches, may explain the role of

several factors have been mentioned before, including the number of microorganisms that contaminate the carcasses through slaughtering, handling, and marketing processes (Bolder, 1998 and Mead, 2004), which differ between the two marks according to the manufacturing processes. The results here agree with those reported by Kenawi *et al*. (2007) who found that the psychrophilic bacterial count in breasts stored at 4ºC in day 1 to 24 increased from 3 log CFU/g meat to 6.8 log CFU/g meat respectively, and results of Gallas *et al*. (2010) who reported that the psychrophilic count in chicken breasts stored under refrigerator condition at 0, 3, 9, 14 days were 3.1, 2.7, 4.4, 6.5 $\log^{10}$ CFU/g meat respectively. Despite the claim of Abu-Ruwaida *et al*. (1996) who studied the microbial shelf life and quality of frozen broiler chickens and reported that prolong frozen storage did not cause substantial changes in the bacterial count of carcasses stored at (-12ºC), as being decreased slightly when stored at (-18ºC), but Modi *et al*. (2005) and Sudheer *et al*. (2011) disapprove that when they reported increasing in psychrophilic count in prolong freezing storage. Even (ICMSF, 2011) confirmed that frozen poultry typically does not undergo microbial spoilage, but they also determined that storage temperature should be controlled to prevent fluctuation which directly have an effect on microbial growth.

The mean values of psychrotrophic bacterial count revealed the lowest by DMIS (table, 2), this could be due to the lowest total bacterial count and psychrophilic. Season may be a strong factor in showing significant differences between the two batches of all marks, due to electrical cut out, freezing and thawing process lead to reactivation of psychrotrophic microorganisms especially in samples collected from grosser shops. Psychrotrophic counts were higher than the mesophilic (table, 1), this mean that most of the mesophiles were adapted to grow in low temperatures (Kraft, 1992; Zeitoun & Al-Eid, 2003; and Jay *et al.*, 2005) as well as a number of psychrophiles that grew in the incubation temperatures of psychrotrophic ones as has been confirmed by several researches (Olson & Nottingham, 1980; Moyer & Morta, 2001 and Jay *et al*., 2005), indeed the storage area and the time of storage may serve as an additional factor (Mead, 2004 and Russell, 2009). The lowest mean value of psychrotrophic bacterial count in the thighs of both batches for the three marks that have been inspected was scored by AJC mark, while Tyson showed the highest in both batches (table, 2). The Psychrotrophes were lower than total aerobic bacterial count, because the samples were bought fresh and the products did not stay for long periods in shops and markets therefor the psychrotrophic bacteria did not have enough time to increase (Jay *et al*., 2005). Tyson mark in batch (1) had a higher count than batch (2) which may be due to the exploration mode of the products in winter (as mentioned earlier) which may led to the growth of mesophilic bacteria at thawing; during refreezing the mesophiles adapt to be psychrotrophs to handle low temperatures (Mead., 2004; Jay *et al*., 2005). The mean value of Psychrotrophic bacterial count of the breasts were higher in batch (1) of Seara (table, 3) which may again be due to the high mesophilic count (total plate count) that has been detected in Seara regarding that most of mesophiles turned to Psychrotrophic and Psychrophils at low temperatures (Lahellec *et al*., 1975; Jay *et al*., 2005). In contrast, samples of the batch (2) of Sadia mark recorded higher count than batch (1) which reflects the effect of summer heat leading to increase in the psychrotrophic count in addition to the high mesophilic count shown in batch (2) of the Sadia. To debate this, some facts should be kept in mind, that Psychrotrophic and Psychrophils have the same minimum temperature ranged -5 to 5 °C but, psycrotrophes have optimum growth temperature at 25 to 30 ºC and Psychrophils have 12-15 ºC, as an optimum range (Olson & Nottingham, 1980; Moyer & Morta, 2001 and Jay *et al*., 2005), and psychrotrophes has been defined as subgroup of mesophiles since their optimum temperatures for growth are in moderately low range and they still grow well at temperatures below the maximum for most mesophiles (Kraft, 1992; Bolder, 1998). Some strains of species that fall into the indicator organisms categories (such as certain coliforms and *Enterobacter* from the family *Enterobacteriaceae*) are psychrotrophic and can multiply on refrigerated raw poultry carcasses and products (ICMSF, 1986).

Some of pathogenic and food poisoning microorganisms are also psychrotrophic in nature like *Salmonella* species*, Staphylococcus, E.coli, Listeria* (Sams, 2001; Mead, 2004 and Jay *et al*., 2005). So, in food quality it's important to count the psychrotrophic bacteria, because any increase may lead to more pathogenic bacterial isolation and identification (USDA /FSIS, 1998).

The sources of coliforms were reported to be due to process of slaughtering, handling and marketing (Mead *et al*., 1993 and Sarlin *et al*., 1998), and the good manufacturing practices (GMP) in addition to that, HACCP may be concerned in manufacturing countries, to control the processing practice (Mead *et al*., 1993; Donald *et al*., 2001), and reducing of count may be due to controlling processes of slaughtering, eviscerating, scalding, chilling, packaging, freezing, transporting and handling (Bolder, 1998). Yet the MPNs for all marks were very low in comparison to results recorded by Khalifa & Abd El-Shaheed (2005) who reported the coliform count in 35 raw chicken samples as $4.1.x10^3$ CFU/g meat and results of Zeitoun and Al-Eid (2003) who studied the safety and quality of frozen poultry at AL-Hassa supermarkets in Saudi Arabiya; 360 samples from seven locally produced companies and from five imported companies were tested, the mean value of coliform count in imported chicken was 1.00 CFU/cm^2.

Table (2) showed count in the frozen thighs, there were no significant differences in the mean value of MPN of coliforms among all the three marks in both batches. This confirms the good hygienic practices by the manufacturers' inspite of that both Tyson and AJC marks were unwrapped at retail outlet. Hygienic habits of the vendors including freezing process may be an additional factor on coliforms count reduction (Bailey *et al*., 2000). Al- Hamadany (2009) reported similar results; she recorded that the frozen chicken thighs of Sadia mark in both retail and whole outlet were coliform free, but the results of AL- Hisnawi (2010) who found that the total coliform count in 15 samples of Sadia mark thighs was 7.3×10^3 CFU/ g meat and in 15 samples of Tyson thigh mark was 6.6×10^3 CFU/ g meat, AL- Hisnawi attributed this difference to bad manufacturing and handling of the products. Although, there were no significant differences in MPN of coliforms between the breasts of the two marks inspected (table, 3). So, the presence of coliform is an indicator of unsanitary conditions through slaughtering processes specially evisceration (Donald et *al*., 2001), or contaminating of the products during handling specially at manufacturing (Mead, 2004) because the products were packaged at retile outlets. There were no significant differences also between the two batches for each mark, which shared the same hygienic and handling conditions. Since there was no specific coliform criteria in the Iraqi quality regulations specified for frozen chicken meat, so the comparison with limits given by H.P.F.B. (2003) which determined coliform limit at $10\text{-}10^3$ CFU/g meat and ICMSF (1986) determined it at 10^2 CFU/g for meat, revealed that all marks for whole chicken and chicken portions were within the standard limits.

As shown in tables (1, 2, 3), *E.coli* O157: H7 was not detected in all inspected samples. This serovar is considered as one of the causatives of food born disease (Feng *et al*., 2001 and Mead, 2004) and so should be included in inspection. The current result is not an exception in this respect if we know that *E.coli* O157 H7 was not applicable in results published by USDA-FSIS (1996) out of 1297 postchill carcass rinse samples from approximately 200 broiler processing plants. *E.coli* is not included in the specific Iraqi standards in both IQS 2270/4 and 3725/4 (Microbiological limits of frozen chicken) (ICOSQC, 2000 and ICOSQC, 2006), whilst all the inspected marks were within the limits introduced by ICMSF (Microbial limits of fresh poultry) (1986) which specified that any food products should be free of this kind of bacteria.

Coagulase positive *Staphylococcus aureus* were not detected in all samples for two batches of all marks (table,1, 2, 3). There are no standard for the incidence of Coagulase positive *Staphylococcus aureus* in Iraqi quality regulations to be compared with (ICOSQC, 2000; ICOSQC, 2006). This was agree with Al-Hamadany (2009) who recorded that the count of *Staphylococcus aureus* in most of imported frozen thighs sold in Baghdad were zero. Contamination of poultry carcasses with enterotoxigenic *S. aureus* of human origin often was mentioned to occur during processing (Adams & Mead, 1983). Indeed coagulase-positive *S. aureus* is typically originated from humans (Donald *et al*., 2001). Even though, both Tyson and AJC marks were unpackaged at retail selling. The effect of freezing on the microorganisms may be the major reason for lowering coagulase positive *staphylococcal* count (Lambert *et al*., 1991 and Jay *et al*., 2005). In a similar study, Al-Hamadany (2009) mentioned that the mean count of *Staphylococcus aureus* in Sadia mark thighs was zero CFU/ g meat in whole sells, and 1×10^2 CFU/g meat in retail sells. While Al -Dughaym & Al-Tabari (2009) in Saudi Arabiya, mentioned in their study on 10 samples of chicken thighs

collected from Al-Ahsa market, that the total mean value of *Staphylococcus aureus* was less than 10^2 CFU/g meats.

Salmonella were also undetected in all samples inspected (Tables, 1, 2, 3), the no detection of *Salmonella* agreed with standard levels of Iraqi quality regulations for frozen poultry meat (ICOSQC IQS 2270/4, 2006).

This means good manufacturing conditions, handling and transporting for the products (Gast, 2003; Sampathkumar *et al*., 2003 and Maed, 2004). The use of prebiotic and probiotic additives to birds feed had been reported to have a good effect on bacterial growth in the birds intestine (Wolfeden *et al*., 2007), but in any case FSIS in 1996, has reported that many produced countries follow the Pathogen Reduction-Hazard Analysis Critical Control Point (HACCP) systems, which set pathogen reduction performance standards for *Salmonella* for slaughter plants and for plants producing raw ground meat products (Bilgili, 2001). Yet freezing had low effect on the *Salmonella* microorganisms (Mead, 2004; ICSMF, 2011), since the last reference reported that Salmonella can live in a frozen product for about one year. AL- Hamdany (2009), in Iraq, reported compatible result when revealed all inspected sample were *Salmonella* free.

Conclusion

Regarding the microbiological quality, all chicken samples (whole chicken carcasses, thighs and breasts) were within Iraqi and international standards and fortunately all samples were free from *Salmonella, E.coli* O15:H7 and coagulase positive *Staphylococcus aureus.* In spite of that there are no standard limits for psychrophilic bacterial count, Psychrotrophic bacterial count, Coliform count, Staphylococcus bacteria, in Iraqi quality standards to be compared with.

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جۆرایە تی بکتیری لە مریشکی هاوردەی بەستوو لە بازاڕەکانی شاری سلیمانی

پوختە

بۆ هەلسەنگاندنی جۆرایەتی بکتیری بۆ گۆشتی مریشکی بەستووی هەوارده کە لە بازارەکانی شاری سلیمانیدا هەیە،لە کۆی ٣٦٠ نموونە مریشکی بەستوو کە حەوت مارکەی بازرگانی لە خۆ گرتبووبە دوو تاوەوە(باج)، لاشە تەواوەکانی مریشکەکان ١٦٠ نموونە بوون، ٨٠ یان لە تاوی یەکەم وە ٨٠ لە تاوی دووەمدابوون. رانەکان ١٢٠ نموونە بوون (٦٠ بۆ هەر تاوێك)، سنگەکانیش ٨٠ نموونە بوون(٤٠ بۆ هەر تاوێك) کە لە بازارەجیاوەزەکانی شاری سلیمانی کۆکرانەوە. ئەژمارکردنی بکتیری گشتی لە باجی یەکەم ودووەمدابۆ لاشەتەواوەکان بە مەودای ٥,٧٢×١٠^٤ و ٠,٢٦- ٥,٧٢×١٠^٤ یەکەی پێکهێنانی نیشینگە / گم گۆشت. لە رانەکانیشدا بە مەودای ١,١٧ -١٧,٥٢ و٠,٧٣- ١٢,٥٠×١٠^٤ یەکەی پێکهێنانی نیشینگە / گم گۆشت تۆمارکرد لە هەردوو باژەکان یەکەم و دووەمدا یەك لەدوای یەك. مەودای ئەژمارکردنی لە سنگدا ١٤,٣٨،١١,٩٠ و ٠,٥٨،١,١٧×١٠^٤ یەکەی پێکهێنانی نیشینگە / گو گۆشت لە باچەکانی یەکەم ودووەمدا یەك دوای یەك.

ژمارەی بەکتییریای هۆگری ساردی لە لاشەکانی مریشکی لەش تەواو بەمودای ١,٩٥-١٣,٧٤ و ٠,٩٠- ٢٧,٤٢×١٠^٤یەکەی پێکهینانی نشینگە /گم گۆشت بوو، لە ڕانەکانیشدا ٠,٥٥١-٨,٥٦ و ٠,٣٦-٨,٠٠×١٠^٤ یەکەی پێکهێنانی نیشینگە / گم گۆشت ، لەسنگدا ٠,٧٣-٨,٨٨ و ١,١٤- ٩,٣٦×١٠^٤ یەکەی پێکهێنانی نیشینگە / گم گۆشت بوو. ژمارەی بەکتیری بەرگری ساردی لە لاشەکانی مریشکی لە ش تە واو بۆ هەردوو باچە کانی یەکەم و دووەم بەمەودای ١,٩٦-١٤,٢٨ و ١,٢٨-٢١,٨٠×١٠^٤ یەکەی پێکهێنانی نیشینگە / گم گۆشت بوو، لە ڕانەکانیشدا٠,٦٤-١٢,٦٤ و ٠,٤٨- ٧,٦٨×١٠^٤ یەکەی پێکهێنانی نیشینگە / گم گۆشت لە باچە کا نی یەکەم و دووەمدا ، لە سنگیشدا٠,٧٧- ١٣,١٤ و ١,١٧- ٨,٤٤×١٠^٤ یەکەی پێکهێنانی نیشینگە / گم گۆشت لە باچە کانی یەکەمدا و دووەمدا یەك لەدوای یەك بوو. زۆرترین ئەگەر ی ژمارەی بکنیریای قوڵۆن لەهەموو نمونەکاندا بە هەردوو باچە کانەوە لە یەك یەکەی پێکهێنانی نشینگە/گم گۆشت کەمتربوو . هەموو نمونەکان لە هەموو مارکەکاندا بە هەردوو باچە کانەوە بکتیریای *E.coli* O157:H7 , وە جۆرە کانی بەکتیریای Coagulase positive *Staphylococcus aureus* وە بکتیریای *Salmonella* یان تیدا نەبووە.

النوعية البكتريولوجية للدجاج المجمد المستورد المباع في اسواق مدينة السليمانية

الخلاصة

لغرض دراسة نوعية لحم الدجاج المجمد من الناحية البكتريولوجية والمباع في اسواق مدينة السليمانية تم فحص ٣٦٠ عينة من الدجاج المجمد حيث تم اخذها من سبعة علامات تجارية اجنبية جمعت بوجبتين (١٦٠ عينة من الذبائح بواقع ٨٠ عينة لكل وجبة) و١٢٠ عينة من الافخاذ (٦٠ لكل وجبة) و٨٠ عينة من الصدور (٤٠) عينة لكل وجبة. وتم اتباع طرق التحليل العالمية في الفحوصات. اظهرت الفحوصات بان كل عينات الذبائح الكاملة كانت ضمن الحدود الميكروبية القياسية، ووجدت فروقات معنوية بين العلامات والوجبات. سجلت معدلات العد الكلي البكتيري مدى ٠,٥٤-٥,٧٢ ×١٠^٤ و ٠,٢٥- ٥,٧٢ ×١٠^٤ وحدة تكوين مستعمرة/غم لحم في الوجبتين الاولى والثانية على التوالي. والعد الكلي البكتيري للافخاذ كان بمدى١,١٧ -١٧,٥٢ و٠,٧٣- ١٢,٥٠ ×١٠^٤وحدة تكوين مستعمرة/غم لحم للوجبة الاولى والثانية على التوالي. سجلت الصدور مدى ١٤,٣٨،١١,٩٠ و ٠,٥٨،١,١٧×١٠^٤ وحدة تكوين مستعمرة/غم لحم لكل من العلامتين التجاريتين **Seara** و **Sadia** على التوالي. اما مدى عدد البكتريا الآلفة للبرودة فقد كان ١,٩٥-١٣,٧٤ و ٠,٩٠- ٢٧,٤٢×١٠^٤وحدة تكوين مستعمرة/غم لحم في الذبائح بينما في الافخاذ فكانت ٠,٥٥-٨,٥٦ و ٠,٣٦-٨,٠٠×١٠^٤وحدة تكوين مستعمرة /غم لحم وفي الصدور كانت ٠,٧٣-٨,٨٨ و ١,١٤- ٩,٣٦ ×١٠^٤وحدة تكوين مستعمرة/غم لحم. ان عدد البكتريا المتحملة للبرودة للذبائح وللوجبتين الاولى والثانية فكانت بمدى ١,٩٦-١٤,٢٨ و ١,٢٨-٢١,٨٠ ×١٠^٤وحدة تكوين مستعمرة/غم لحم . والافخاذ سجلت مدى ٠,٦٤-١٢,٦٤ و ٠,٤٨- ٧,٦٨ ×١٠^٤ وحدة تكوين المستعمرة/غم لحم وللوجبتين الاولى والثانية على التوالي. والصدور سجلت مدى ٠,٧٧- ١٣,١٤ و ١,١٧- ٨,٤٤ ×١٠^٤وحدة تكوين مستعمرة/غم لحم وللوجبتين الاولى والثانية على التوالي. العدد الاكثر احتمالا للبكتريا القولونيه لكل العينات ولكلا الوجبتين اقل من ١وحدة تكوين مستعمرة/غم لحم. وكذلك خلت كل العينات ولكل العلامات وفي كلا الوجبتين من بكتريا *E.coli* O157:H7 و بكتريا Coagulase positive *Staphylococcus aureus* وانواع بكتريا *Salmonella*.