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Bacteriological Quality and Isolation Some Pathogenic Bacteria from Some Meat Products Sold In Some Local Markets of West Libya (Alzawi, Surman, Sabratha and Algelat)

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Abstract This study was aimed to evaluate the bacteriological quality and isolation some pathogenic bacteria from some meat products sold in some local markets of West Libya (Alzawi, surman, sabratha and Algelat) .5 samples of minced meat, beef burger, and sausage were collected from each city subjected to bacteriological analysis. Isolation and identification of some pathogenic and Public Health Hazard bacterial groups (salmonella, E.coli and staphylococcus) were carried out. The obtained results indicated that minced meat has the highest contamination level compared with the other products. The mean values of total bacterial count isolated from minced meat, beef burger and sausage samples were 6 x 10⁸, 3.1x 10⁵ and 5.6x 10⁴CFU/g, respectively. Escherichia coli were detected in 50 % of the examined minced meet samples and 30% of beef burger but not found in sausage samples. Salmonella were isolated from 20 % of minced meat sample and 10% of Beef burger at levels of 6x 10⁴ and 4x 10² CFU/ g, respectively. Data also showed that 20% of minced meet samples and 10% of beef burger samples were contaminated with Staphylococcus aureus at levels of 3x10³, 4x 10² and 2x 10² CFU/g, respectively.

Key words: meat contamination, meat microbial-isolation of bacteria.

الجودة البكتريولوجية وعزل بعض البكتيريا المسببة للأمراض من بعض منتجات اللحوم المباعة في بعض الأسواق المحلية في غرب ليبيا (الزاوية، صرمان، صبراتة والعجيلات)

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الملخص هدفت هذه الدر اسة إلى تقييم الجودة البكتريولوجية وعزل بعض البكتيريا المسببة للأمر اض من بعض منتجات اللحوم التي تباع في بعض الأسواق المحلية في غرب ليبيا (الزاوية، صرمان، صبراتة والعجيلات). تم جمع 5عينات من اللحم المفروم، ولحوم البقر، والسجق من المدن موضوع الدراسة خضعت العينات للتحليل البكتريولوجي. تم إجراء العزل والتعرف على بعض المجموعات البكتيرية المسببة للأمر اض وذات الخطورة على الصحة العامة (السالمونيلا وبكتيريا القالون والمكورات العنقودية) وذلك بمعامل مركز الرقابة على الأغذية فرع زوارة. أشارت النتائج التي تم الحصول عليها إلى أن اللحوم المفرومة كانت أعلى مستوى تلوث مقارنة بالمنتجات الأخرى. كانت القيم المتوسطة لإجمالي عدد البكتيريا المعزولة من اللحم المفروم وعينات برجر اللحم واالسجق x 10⁸ أ 3.1x 10⁵ , و 5.6x 104CFU/g على التوالي. تم الكشف عن الإشريشيا القولونية في 50% من عينات اللحم المفروم التي تم فحصبها و 30% من برغر اللحم البقري ولكن لم يتم العثور عليها في عينات النقانق. تم عزل السالمونيلا من 20٪ من عينة اللحم المفروم و10٪ من برغر اللحم البقري بمستويات 6x 104 و 4x 102 CFU/ 8، على التوالي. أظهرت النتائج أيضاً أن 20٪ من عينات اللحم المفرومة و10٪ من عينات برجر اللحم البقرى ملوثة بالمكورات العنقودية الذهبية بمستويات .2x 10² CFU/g و 3x 10³, 4x 10² على التوالي. الكلمات المفتاحية: تلوث اللحوم –ميكروبات اللحم –عزل البكتيريا.

Introduction

Meat is a very important food to human health due to its composition; Meat is rich in high quality protein, fats, vitamins, minerals and trace elements, so that a huge number of people consume meat and meat products. Generally, meats are very susceptible to quality loss due to microbiological spoilage. The bad and improper processing, handling and storage of meat products lead to spoilage which rises to economic losses and public health hazard.

Meat and meat product such as minced meat appreciated because of its convenience. are

Unfortunately, their shelf- life is limited because the large exposed surface area facilitates spoilage. The rate of deteriorative change depends on meat composition, hygienic practices during cutting, grinding, and preparation, as well as storage conditions. The most important factor in controlling meat spoilage microbial is contamination and their growth, which affect safety and quality (4). Food safety experts, agree that pathogen reduction requires a farm to the table approach. Microbiological testing is designed to address improvements at the plant level, with the

understanding that additional initiatives at other points in the food production chain also are needed. USDA already has begun a number of projects to address these other points, including safe handling instructions for consumers, identification and trace back of animals, and the development of on-farm pathogen prevention models. FSIS established a series of baseline data collection programs to acquire information that provides general microbiological profiles of meat and poultry for selected

microorganisms that are of various degrees of public health concern (19). Baseline studies are to develop pathogen reduction also used performance standards that plans must meet earlier baseline studies (steer/heifer, cow/bull, broiler chicken, market hog, and young turkey) and surveys (raw ground beef, raw ground chicken, and raw ground turkey) included the following microbial analyses of *Escherichia coli; Clostridium* perfringens; Staphylococcus aureus; Listeria monocytogenes; Campylobacter; Escherichia coli 0157:H7; and Salmonella (14). Although the total bacterial count was used in bacteriological examination to reflect the hygienic quality, however, it is evident that coliform group count is considered of much greater value in assessing its quality (7). Salmonella is now established, as one of the most important causes of food – brone illness at worldwide (17). The Staphylococcal genus contains at least 23 species, most important being Staphylococcus aureus. This organism is of major concern to the meat and poultry industries (16).

The purpose of the study was to assess the bacteriological quality for some meat products sold in some cities in western Libya and Isolation and identification of some pathogenic and health hazard bacterial groups was carried out.

Materials and Methods

Samples of meat products:

20 retail samples of minced meat, beef burger, and sausage (5samples of each product collected from four sources of local market of Alzawi, Surman , Sabratha and Algelat(cities in west of Libya).

The analysis were done in microbiology Lab. Food &Drug control center - zwara

Media used:

1-Media used for determination of total bacterial count:

Nutrient agar medium (American Public Health Association(**(1**, 6) was used for the determination of total bacterial count.

2-Media used for isolation of Staphylococcus aureus:

Manitol salt agar media and Vogel Jonson media were used to isolate *Staphylococcus aureus* according to(**6**)

3-Media used for isolation of coliform group bacteria:

Mac Conkey broth, Mac Conkey agar and Eosin methylene blue agar media were used for isolation and identification of coliform bacteria (*E. coli*) according to **(6).**

4-Media used for isolation Salmonella:

The salmonella – shigella – agar medium was used as selective plating

medium as described by (13).

Preparation of samples for bacteriological analysis:

Ten grams of each sample were mixed with 90 ml of sterile saline solution (9 g Na Cl/1L distilled water) under sterile conditions to give 1/10 dilution. Serial dilutions were prepared to be used for counting several types of bacteria.

Determination of total bacterial count:

The total bacterial count was determined using the plate counts technique on a nutrient agar medium coording to procedures of (1) and (6). The plates were incubated at 37°C for 48 hrs.

Isolation of Staphylococcus aureus:

Staphylococcus aureus bacteria was determined according to the method described by (1 and 6) using Vogel Jonson medium plus 1 ml potassium tellurite solution 1 % (w/v) to each 100 ml of sterilized medium which mixed well before pouring in the plates. The plates were incubated at 37° C for 24 hr.

Isolation of coliform bacteria:

Coliform group bacteria were determined using Mac Conkey agar medium according to the procedures described by (1 and 6). The plates were incubated at 37°C for 24 hr.

Isolation of Salmonella:

The presence or absence of *Salmonella* was determined according to the method described by(**13**). *Salmonella* - *Shigella* agar plates were incubated at 35°C for 24 hr. *Salmonella* appeared as black colonies, some of them with metalic sheet.

Results and Discussion

<u>Total count bacteria in alzawia</u>

Data presented in Table (1) illustrated the total count bacteria of minced meat of different sources (A1, A2, A3, A4, A5) were 10x10^{8,} 2x10^{8,} 8x10⁸, 8x10⁸ and 5x10⁸ respectively. The table showed that sample A1 is the highest contamination and sample A2 are the lowest contamination. That is meaning some of sources worked in safe conditions when compared. But generally the values are in agreement with those obtained by (14) found that the aerobic plate counts of two groups ground beef samples were 4.7X10⁶ and 3.5X10⁶ respectively. They concluded that microbiological quality of ground beef depends not only on size but also on length of storage time between grinding and use.) surveyed 124 delicatessen meat products for microbiological quality shortly after purchase and following storage at 22 degree for 24 h. They found that 34.3% of the samples contained aerobic plate count of 107 CFU per g and increased to 62.7% following storage.

(11) determined the microbiological quality for retail packages of frozen ground beef and kofta over a period of six months in Assiut and Cairo. The average total bacterial counts per gram were as follows: 6×10^7 CFU/g for frozen ground beef and 106 for frozen kofta. Also (18) found that the aerobic plate count in 555 samples of minced beef (in Berlin, Germany ranged from 2.7 x 10³ to 9.3 x 10⁶ CF U/g. The result obtained is disagree with Libyan Standard Specification.

Table (1) total bacterial count for minced meat in Alzawia

Sample	No. of samples	Total count		
A1	5	$10x10^{8}$		
A2	5	$2x10^{8}$		
A3	5	$8x10^{8}$		
A4	5	$8x10^{8}$		
A5	5	$5x10^{8}$		
Total and	đ	6.6 x10 ⁸		
average				

A1-A2- A3-A4-A5= Minced meat from different sources in Alzawia

<u>Total count bacteria in surman</u>

Data presented in Table (2) illustrated the total count bacteria of minced meat of different sources (A1, A2, A3, A4, A5) were $8x10^{8}$, $3x10^{8}$, $6x10^{8}$, $8x10^{8}$ and $6x10^{8}$ respectively with average $6.2x10^{8}$. The table showed that sample A1 and A4 is the highest contamination and sample A2 are the lowest contamination. That is meaning some of sources worked in safe conditions when compared. Generally, the values are in agreement with those obtained by **(3)**.the result show the all samples have high contamination and disagree with Libyan Standard Specification.

Table (2) Total bacterial count for_minced meat in Surman

Sample	No. of samples	Total count		
A1	5	8x10 ⁸		
A2	5	3x10 ⁸		
A3	5	6x10 ⁸		
A4	5	$8x10^{8}$		
A5	5	6x10 ⁸		
Total and	l	6.2 x10 ⁸		
average				

A1-A2- A3-A4-A5= Minced meat from different sources in Surman

Total count bacteria in Sabratha

Data presented in Table (3) illustrated the total count bacteria of minced meat of different sources (A1, A2, A3, A4, A5) were 8×10^{8} , 5×10^{8} , 7×10^{8} and 5×10^{8} respectively with average 6×10^{8} . The table showed that sample A1 is the highest contamination and the samples A2, A3 and A5 are the lowest contamination. That is meaning some of sources worked in unsafe conditions because the all samples have high contamination and disagree with Libyan Standard Specification. The result also show that, the values are in agreement with those obtained by (2), (11) and (3).

Table (3) Total bacterial count for_minced meat in Sabratha

Sample	No. of samples	Total count
A1	5	$8x10^{8}$
A2	5	$5x10^{8}$
A3	5	$5x10^{8}$
A4	5	$7x10^{8}$
A5	5	$5x10^{8}$
Total and	1	6 x10 ⁸
average		

A1-A2- A3-A4-A5= Minced meat from different sources in Sabratha

<u>Total count bacteria in Al-agalat</u>

JOPAS Vol.18 No. 4 2019

As shown in table 4 all samples were contaminated by bacteria .the result show the total count bacteria in the samples were $6x10^8$, $4x10^8$, $5x10^8$, $5x10^8$, $5x10^8$ respectively With average 5x10⁸. The obtained results are in agreement with those recorded by (20), They investigated microbial quality of 50 samples of luncheon and minced meat (25 samples of each). They showed that the minced meat has heavier bacterial load than luncheon samples and they traced this result to miss handling, improper hygienic measures during manufacturing and transportation and keeping methods as well as methods of exposure to sale. In addition, (22) examined 80 samples of minced meat, kofta, beef burger and luncheon (20 samples each) from different areas in Cairo and Giza. They found the aerobic plate counts for these previous products were 2.2 x 10⁶, 2.9 x 10³, 2 x 10⁵ and 1.3 x 10⁵, respectively. This result also agreement with (3) he investigated microbial quality of 200 samples of minced meat in Assuit city in Egypt his results show all samples were contaminated by bacteria. These results disagree with Libyan Standard Specification.

 Table (4) Total bacterial count for_minced meat

 in Al-agalat

Sample	No. of samples	Total count		
A1	5	6x10 ⁸		
A2	5	4x10 ⁸		
A3	5	5x10 ⁸		
A4	5	5x10 ⁸		
A5	5	5x10 ⁸		
Total and	l	5 x10 ⁸		
average				

A1-A2- A3-A4-A5= Minced meat from different sources in Al-agalat

<u>Total count bacteria for minced meat of</u> <u>different cities</u>

The analysis show that, the minced meat obtained from **Al-zawia** was the highest contamination (6.6×10^8). on the other hand, the minced meat obtained from **Al-gelat** was the lowest (5×10^8). The result also show the total count bacteria of minced meat for Surman and Sabratha nearly same (6.2×10^8 and 6×10^8). In addition the result show all studied samples were contaminated by bacteria and disagree with Libyan Standard Specification..

Table (5) Total count bacteria for minced meat of different cities

City	Total count
Al-zawia	6.6x10 ⁸
Surman	6.2×10^8
Sabratha	6x10 ⁸
Al-gelat	5x10 ⁸

Data presented in Table (6) showed the total aerobic bacterial count isolated from minced meat, beef burger and sausage samples collected from different sources. The bacteriological analysis indicated that minced meat has the highest contamination level compared with the other products. The total count of aerobic bacteria isolated from minced meat ranged from 5 x 10⁶ to 10 x 10⁸ with an average of 6 x 10⁸ CFU/g. At the same time, the mean values of total bacterial count isolated from beef burger and sausage samples were $3.1x 10^5$ and $5.6x 10^4$ CFU/g respectively.

The obtained results are in agreement with those recorded by **(20)**, They investigated microbial quality of 50 samples of luncheon and minced meat (25 samples of each). They showed that the minced meat has heavier bacterial load than luncheon samples and they traced this result to miss handling, improper hygienic measures during manufacturing and transportation and keeping methods as well as methods of exposure to sale. In addition, **(**22**)** examined 80 samples of minced meat, kofta, beef burger and luncheon (20 samples each) from different areas in Cairo and Giza. They found the aerobic plate counts for these previous products were 2.2 x 10^6 , 2.9×10^3 , 2×10^5 and 1.3 x 10^5 , respectively.

(8) reported that fresh minced meat

tends to have a short shelf life because the quality of the raw ingredients is usually lower (i.e., has higher number of contaminating microorganisms), and is re-contaminated through the grinding /handling process. Mincing and grinding of meat at the retail location can introduce more spoilage microorganisms if proper equipment hygiene and handling measures are not followed. These results are agreement with **(23) and (3)**.

Table 6. Total aerobic bacterial count (CFU/g) of meat products samples

	±						
Meat	No. of	samples Count (CFU/g)					
Products	samples	No.	%	Min.	Max.	Mean	
Minced meat	20	20	100%	5 x 10 ⁸	10 x 10 ⁸	6 x 10 ⁸	
Beef burger	20	20	100%	3 x 104	6 x 10 ⁵	3.1x 10 ⁵	
Sausage	20	20	100%	3 x 10 ³	8x 104	5.6x 104	

Data in Table (7) showed that *E. coli* was detected in 50 % of the examined minced meet samples and 30% of beef burger samples. The average values of the contamination level with *E. coli* were 6×10^4 , 6×10^2 and 4×10^2 in the examined samples of minced meet and beef burger, respectively. On the other hand, *E. coli* couldn't detect in the examined sausage samples. Previous investigators found nearly similar results. (9) and , (12), they detected *E. coli* in 47.37% and 28.3% of the examined minced meat and beef burger samples.

Table 7. *Escherichia coli* count (CFU/g) in meat products samples

Meat Products	No. of		itive Iples	Esch	ia coli t	
Froducts	samples	No.	%	Min.	Max.	Mean
Minced	50	25	50%	Зx	8x	бx
meat	50	25	50%	10^{2}	104	104
Sausage	50	0	0	-	-	-
Beef	50	15	30%	1x	бx	4x
burger	50	13	30%	102	102	102

Fig. (1 and 2): Isolation of *E. coli* from meat products on Mac-Conkey agar and EMB agar media.



Fig. (1) : *E-coli* on Mac-Conkey agar give EMB Pink colonies

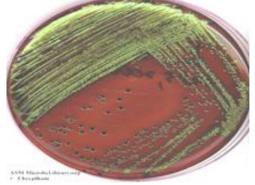


Fig. (2): E-coli on gives green metal shine

Table (8) clears that the incidence of salmonella in miced meat and Beef burger samples were 20 % and 10%, respectively, but not detected in Sausage. Salmonella counts ranged from3x 102 to 8x 10⁴ with mean value of 6x 10⁴ CFU/ g of minced meat, but only ranged from 1x 10² to 6x 10² with a mean value 4x 10²CFU/ g of Beef burger. The obtained results were similar to some extent with that reported by (11), they found that the incidence of Salmonella in beef burger was 6% out of 50 samples and in frozen minced meat was 6% out of 50 tested samples and in fresh minced meat the percentage was 12%. In contrary, they failed to isolate salmonella from any of examined luncheon samples. Salmonella species were detected in 5% of the examined minced meat samples, but not found in any of the examined luncheon or kofta samples. (2), (18), (23),

Table 8. Salmonella counts (CFU/g) in meat products samples

Meat Products	No. of Samples			Salm	ounts	
Flounces	samples	No. %		Min.	Max.	Mean
Minced	20	4	20%	Зx	8x	бx
meat	20	4	20%	10 ²	104	104
Sausage	20	0	0%	_	_	_
Beef	20	2	10%	1x	бх	4x
burger	20	2	10%	10 ²	10 ²	102

Fig. (3 and 4): Isolation of *Salmonella* from meat products on Mac Conkey agar and Bismus sulphate agar media.



Fig.(3): pale yellow colonies of *Salmonella* on Mac. Agar

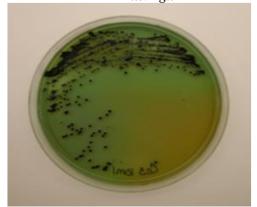


Fig.(4): black colonies of *Salmonella* on Bismus agar

Data presented in Table (4) indicated the incidence of *Staphylococcus spp.* in 10, 5, and 5 samples out of 50 analyzed samples of each of minced meat and beef burger but not in sausage samples. Minced meat showed the highest contamination level (3x 103 CFU/g) Followed by beef burger (4x 102 CFU/g). **(21) and . (15)** who detected *Staphylococcus* in 1.5% and 11.4% of the examined ground meat samples of the examined ground beef samples obtained nearly similar results. On the

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other hand, *Staphylococcus* had not been detected in beef burger or minced meat as reported **(22)**, **(2)**, **(9)**, **(5)**.**(23)** and the same result obtained by **(3)**.

Table 4. *Staphylococcus* count (CFU/g) in meat product samples

Meat Products	No. of samples	Positive samples		Staphylococcus count		
Products	samples	No.	%	Min.	Max.	Mean
Minced	20	4	20%	1x	бх	3x 10 ³
meat		4	20%	10 ²	10 ³	5x 10°
Sausage	20	0	0%	-	-	-
Beef	20	2	10%	2x	бх	4x 10 ²
burger		2	10%	10	10 ²	4X 10 ²

Fig. (5 and 6): Isolation of *Staphylococcus* from minced meat samples on Mannitol salt agar and blood agar media



Fig.(5):yellow colonies of *Staphylococcus* on Mannitol salt agar pale



Fig.(6): B- haemolysis on blood agar with Clear zone around *Staphylococcus* colony

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470