



## Microbial Profile of Some Ready to Eat Meat Products Retailed for Sale in Al Beida City, Libya

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### ABSTRACT

Ready-to-eat (RTE) meat products have become one of the most important sources of nutrition, especially with changing consumer's dietary and social habits. Unfortunately, it may be loaded with many microorganisms especially accidental post-cooking cross-contamination. The current study was conducted to determine the microbial profile of some ready to eat meat products retailed for sale in Al Beida City, Libya. A total of 75 random samples of ready to eat (RTE) meat products including; Luncheon, Frankfurter and Hot dog (25 samples / each) were collected from different supermarkets and groceries for microbiological evaluation. Statistical analytical results of Aerobic plate count (APC) clarified that the highest mean value was recorded in samples of hot dog ( $4.1 \times 10^5$  cfu/g) followed by luncheon ( $1.4 \times 10^5$  cfu/g) then Frankfurter ( $1.1 \times 10^5$  cfu/g). Concerning Enterobacteriaceae count (EC), the highest mean value was recorded in samples of hot dog ( $6.8 \times 10^4$  cfu/g) followed by Frankfurter ( $6.7 \times 10^4$  cfu/g) then luncheon ( $2.1 \times 10^4$  cfu/g) while the highest mean value of coliforms count was recorded in samples of Frankfurter ( $4.8 \times 10^4$  cfu/g) followed by hot dog ( $4.7 \times 10^4$  cfu/g) then luncheon ( $1.4 \times 10^4$  cfu/g). On the other side, the prevalence of E.coli was 40, 32 and 16% in the examined samples of Luncheon, Frankfurter and Hot dog, respectively and serotyping of Enteropathogenic E.coli isolated samples revealed the presence of O17: H18 (EPEC), O26: H11 (EHEC), O55: H7 (EPEC), O91: H21 (EHEC) and O113: H2 (EPEC) with different rates. Finally, the prevalence of Salmonellae was 16, 12 and 8% in the examined samples of Luncheon, Frankfurter and Hot dog, respectively and serotyping of isolated Salmonellae revealed the presence of *S. Enteritidis*, *S. Virchow* and *S. Heidelberg* with different rates.

**Keywords:** Ready to Eat, Meat, Products, Microbial Profile

### 1. Introduction

Ready-to-eat (RTE) meat products have become one of the most important sources of nutrition, especially with changing consumer's dietary and social habits. Unfortunately, it may be loaded with many microorganisms especially accidental post-cooking cross-contamination so good manufacturing practices and the hygienic conditions of these practices and the hygienic conditions of these products are very important during the procedures of preparation, handling and storage as they are contaminated from different sources this may lead to spoilage of these products and/or act as a public health hazard to consumers.

Fast foods have been defined by FAO as Ready-to-eat foods and beverages prepared and/or sold by vendors especially in streets and other public places for immediate consumption. These foods are well appreciated by consumers, mostly by urban workers because of their taste, low cost, nutrient value and ready availability for immediate consumption. It includes fast foods, junk foods, snacks, beverages, meals, salads, sliced fruits and drinks for a wide variety of people (FAO/WHO, 2009).

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Heat-treated meat products refer to any meat that has been transformed

through heating to enhance flavour or improve preservation. Most processed meats are made from beef, but may also include other meats, such as poultry (Santarelli et al., 2008). Ready to eat meat products are highly demanded due to their biological value, reasonable price, and agreeable taste; also, they represent rapid easily prepared meals and solve the problem of shortage in fresh meat of high price which is not available for many families with limited income (Samapundo et al., 2015). Contaminated meat products may constitute a public health hazard (Datta et al., 2012). The main sources of pathogenic bacteria in food are contaminated raw food, food handlers, dust, water, utensils and insects (Ray, 1996). RTE food has been implicated in cases of food poisoning or gastroenteritis in human beings (Eley, 1996). The presence of Enterobacteriaceae and coliforms in meat products depend upon the meat used for grinding, sanitary conditions, practices during preparation, time and temperature of processing and storage. Also, during cutting and handling, meat surfaces exposed to ambient air provide excellent media for most bacteria. *Escherichia coli* (*E. coli*) is one of the main gastrointestinal inhabitants in most mammalian species, including humans, and birds. Most *E. coli* are commensal, but small proportions are potentially harmful and cause diseases worldwide (Frye and Jackson, 2013). The pathogenic *E. coli* are classified into classes based on the production of different virulence factors and on the clinical manifestations that they cause (Kim et al., 2020). Shiga toxins-producing *E. coli* (STEC) are a group of highly pathogenic strains known as enterohaemorrhagic *E. coli* (EHEC) or verotoxins-producing *E. coli* (VTEC) (Detzner et al., 2020). It is considered as one of the most emerging foodborne zoonotic bacteria causing various clinical signs as watery or bloody diarrhea, and potentially life-threatening syndromes such as hemorrhagic colitis (HC), thrombotic thrombocytopenic purpura (TTP), hemolytic uremic syndrome (HUS) and acute renal failure (Karmali et al., 2010). The pathogenicity of STEC strains is attributed to the production of different virulence factors including two potent phage-encoded cytotoxins as *stx1*, and *stx2*. These toxins are like to those produced by *Shigella dysenteriae* which inhibit protein synthesis in host cell leading to cell death (El Syaed and Mounir, 2020). *Salmonella* is a member of the Enterobacteriaceae, Gram negative, motile, with peritrichous flagella and non-spore forming rods. Also, it is a facultative anaerobic (can grow with or without oxygen) catalase positive and oxidase negative bacteria. However, *Salmonella* is not included in the group of organisms referred to as coliforms (Lawley et al., 2008). More than 2,500 different types of *Salmonella* exist, some of which cause illness in both animals and people. Some types cause illness in animals but not in people. Some serotypes are only present in certain parts of the world (Brands, 2006). Notify, different *Salmonella* spp. were isolated from different types of meat, the commonest non typhoid *Salmonella* were *Salmonella Enteritidis* and *Salmonella Typhimurium* (Ali et al., 2010, Abd El Aziz, 2013, Fardsanie et al., 2016 and El Sisy and Elzanatey, 2019). So the objective of the current study was to assess the microbiological quality of some heat treated meat products including; luncheon, frankfurter and Hot dog retailed for sale in Al Beida City, Libya. In addition, isolation and identification of some potential pathogenic bacteria were attempted.

### 2. Material and methods

#### 2.1. Samples:

A total of 75 random samples (250 g weight of each) of ready to eat meat

products retailed for sale in Al Beida City, Libya were collected from different shops and grocery stores. Twenty five samples each of cooked luncheon, frankfurter and hot dog. The samples were separately put in clean sterile plastic bags, identified and transferred in an insulated ice box to the laboratory under complete aseptic conditions. The collected samples were subjected for microbiological examination.

## 2.2. Microbiological examination:

2.2.1. Preparation of samples was carried out according to APHA (2013).

2.2.2. Determination of Aerobic plate count (APC) according to ISO 4833-1, (2013).

2.2.3. Enterobacteriaceae count according to ISO, (2007).

2.2.4. Determination of coliform count ISO 4832, (2006).

2.2.5. Mold and yeast count according to ISO, (2007).

2.2.6. Screening for Enteropathogenic *Escherichia coli* according to FDA (2002) and serological identification according to Kok et al., (1996).

2.2.7. Detection of *Salmonellae* was performed according to ISO 6579, (2017) and serological identification according to Kauffmann, (1974).

## 3. Results and discussion

Ready to eat meat products are highly demanded due to their biological value, reasonable price, and agreeable taste; also, they represent rapid easily prepared meals and solve the problem of shortage in fresh meat of high price which is not available for many families with limited income (Samapundo et al., 2015). Street vendor foods raise concerns with respect to their potential for serious food poisoning outbreaks due to improper use of additives, the presence of pathogenic bacteria, environmental contaminants, and improper food handling practices based on unrespect of good manufacturing practices and good hygienic practices (Estrada et al., 2004).

Aerobic plate count can provide useful information about the remaining shelf-life of the food in question, and thus highlight potential problems of storage and handling since production and a general indication of the microbiological quality of food not safety. So, high aerobic plate count may indicate unhygienic preparation; inappropriate storage conditions or suggests possible poor temperature control (HPA, 2009).

It is evident from the obtained result in Table (1) that the highest mean value was recorded in samples of hot dog ( $4.1 \times 10^5$  cfu/g) followed by luncheon ( $1.4 \times 10^5$  cfu/g) then Frankfurter ( $1.1 \times 10^5$  cfu/g). The examined samples of hot dog were the most contaminated ones followed by luncheon and Frankfurter. This could be attributed to the fact that Hot dog may receive more handling during preparation as well as addition of spices which may be contaminated with larger number of microorganisms. Such variations may be attributed to difference in quality of meat from the sanitary point of view or ingredients added to meat as vegetables and cheese, the hygienic standard during processing or time and temperature of storage and retailing of product may play a role. Also, the presented data in Table (1) showed that 16, 54 and 98% of the examined samples of luncheon, frankfurter and hot dog, respectively had a total bacterial count more than the permissible limits when compared with EOS, (2005).

These results were in harmony with that of Kasem, (2016) who recorded that the mean value of aerobic plate count was  $8.9 \times 10^3$  cfu/g for luncheon,  $5.2 \times 10^3$  cfu/g for frankfurter and  $7.2 \times 10^3$  cfu/g for hot dog and Salem et al., (2019) who found that the mean value of aerobic plate of examined RTE beef product samples were  $1.48 \times 10^5$  cfu/g in Beef fajitas and  $1.94 \times 10^5$  cfu/g in Hotdog.

Bacterial count of perishable food is used to evaluate its quality and shelf-life. However, high count may be attributed to unsanitary methods of production or exposure to conditions favoring bacterial proliferation as decided by Sharma et al. (1996).

Detection of any or all members of the family Enterobacteriaceae as indicator of food sanitary quality has received the attention of more food scientists. The occurrence of Enterobacteriaceae indicated microbiological and toxigenic bacteria in meat and lead to public health hazard (Mira, 1989). The source of Enterobacteriaceae on meat was shown to be associated with the meat handling work surface. Also, the presence of Enterobacteriaceae in ground beef is an indicator of direct or indirect enteric contamination of meat (Stiles and Lai-King, 1981).

The recorded data in Table (2) showed that the highest mean value of Enterobacteriaceae count was recorded in samples of hot dog ( $6.8 \times 10^4$  cfu/g) followed by Frankfurter ( $6.7 \times 10^4$  cfu/g) then luncheon ( $2.1 \times 10^4$  cfu/g). The presence of Enterobacteriaceae in meat products may be attributed to contamination of meat handler's hands and tools and handling surfaces during all stages of processing especially with fecal contamination.

Nearly similar results were reported by Al-Mutairi, (2011) who reported that Enterobacteriaceae count/g was the lowest among examined sausage samples when compared with other meat products. Similarly, Gouda (1991) reported that Enterobacteriaceae count/g was significantly higher in luncheon meat product samples than in sausage meat product samples.

Coliforms are used as indicator of water pollution or as a general indicator of sanitary condition in the food processing environment (Feng et al., 2002). Also, high Coliforms count indicates poor hygienic quality of meat and is significant as indicator of fecal contamination. Coliforms also have the ability to grow well over wide range of temperature below  $10^\circ\text{C}$  up to

$46^\circ\text{C}$  (Gill et al., 1996).

As shown in Table (3), the highest mean value of coliforms count was recorded in samples of Frankfurter ( $4.8 \times 10^4$  cfu/g) followed by hot dog ( $4.7 \times 10^4$  cfu/g) then luncheon ( $1.4 \times 10^4$  cfu/g).

Accordingly, the examined samples of Frankfurter were the most contaminated ones. This could be attributed to the neglected sanitary measures during their processing, handling and serving of such products.

Similar results were obtained by Salem et al., (2019) who recorded that the mean values of coliform count (cfu/g) were  $2.42 \times 10^4$  and  $1.63 \times 10^4$  in Beef fajitas and Hotdog, respectively.

The presence of coliforms in meat samples suggested mostly fecal contamination and points to potentially severe hazard (Eribo and Jay, 1985). Unfortunately, undercooked meat products have caused much food poisoning incidence associated with coliforms and investigations had established that the bacteria is present in the feces, intestine and hide of healthy cattle from those it could be potentially contaminate meat during the slaughtering process (Duffy et al., 2006).

Variations may be attributed to the processing defect and/or post-processing contamination from workers, utensils and contact surfaces which indicate inadequate hygiene. The presence of high coliform counts in RTE food indicates deplorable poor hygiene and sanitary practices employed in the processing and packaging of this food product.

Mould can grow over an extremely wide range of temperature. Therefore, one can find mold particularly all foods at almost any temperature under which food are held. Besides mold can assists in the putrefactive processes and may produce toxic substances namely mycotoxins which are harmful to man and animal (Frazier and Wasthoff, 1988). Mould count is used as an index of the proper sanitation and high quality products. Mould can assists in putrefactive processes and in other cases; they may impart a moldy odor and taste of food stuffs.

Data presented in Table (4) showed that the highest mean value was recorded in samples of luncheon ( $6.4 \times 10^3$  cfu/g) followed by Frankfurter ( $2.4 \times 10^3$  cfu/g) then hot dog ( $1.9 \times 10^3$  cfu/g).

These results were in harmony with that of Kasem, (2016) who recorded that molds count cfu/g of luncheon samples ranged from  $1 \times 10^2$  to  $1.1 \times 10^3$  with a mean value of  $6.1 \times 10^2 \pm 1.1 \times 10^2$ , frankfurter samples ranged from  $3 \times 10^2$  to  $1.3 \times 10^3$  with a mean value of  $7.2 \times 10^2 \pm 1.3 \times 10^2$  and hot dog samples ranged from  $4 \times 10^2$  to  $1.6 \times 10^3$  with a mean value of  $7.9 \times 10^2 \pm 1.4 \times 10^2$ .

Yeasts normally play a small role in spoilage because they constitute only a small portion of the initial population, because they grow slowly in a comparison with most bacteria and because their growth may be limited by metabolic substances which can be produced by bacteria. Spoilage yeast is those find their way into food being widely distributed into nature resulting in undesirable changes in physical appearance of food (Walker, 1976).

From data presented in Table (5), the highest mean value of yeasts count was recorded in samples of Frankfurter ( $4.9 \times 10^4$  cfu/g) followed by hot dog ( $4.1 \times 10^4$  cfu/g) then luncheon ( $1.5 \times 10^4$  cfu/g). These results were in harmony with that of Kasem, (2016) who found that yeasts count cfu/g of luncheon samples ranged from  $6 \times 10^2$  to  $3.2 \times 10^3$  with a mean value of  $1.1 \times 10^3 \pm 4.1 \times 10^2$  cfu/g, frankfurter ranged from  $9 \times 10^2$  to  $4.2 \times 10^3$  with a mean value of  $1.6 \times 10^3 \pm 3.8 \times 10^2$  cfu/g and hot dog samples ranged from  $8 \times 10^2$  to  $3.6 \times 10^3$  with a mean value  $1.4 \times 10^3 \pm 4.2 \times 10^2$  cfu/g.

*Escherichia coli* is used as an indicator for fecal contamination and poor sanitation during processing; its presence in RTE foods indicates that the food has been prepared under poor hygienic conditions (Khater et al., 2013).

The presented data in Table (6) showed that the incidence of *E.coli* in RTE meat products samples was 40, 32 and 16% in the examined samples of Luncheon, Frankfurter and Hot dog, respectively and serotyping of Enteropathogenic *E.coli* revealed the presence of O17: H18 (EPEC), O26: H11 (EHEC), O55: H7 (EPEC), O91: H21 (EHEC) and O113: H2 (EPEC) with different rates. These results were lower than that of Kasem, (2016) who found that Enteropathogenic *E. coli* was isolated from 36, 48 and 32% of the examined samples of luncheon, frankfurter and hot dog, respectively. The prevalence of *E. coli* in Luncheon were nearly similar to that recorded by Tarabees, et al, (2015) (22.5%). On contrary, these results of Hot dog were higher than that recorded by Oranusi et al., (2011) (11%), Al-Mutairi, (2011) (12%) and Salem et al., (2019) (16%).

Salem et al., (2019) obtained serotypes O127:H6 (ETEC), O119:H6 (EPEC), O55:H7 (EPEC) and O124 (EIEC) in RTE Hotdog.

*Salmonella enterica* serovar typhimurium and *Salmonella enterica* serovar enteritidis are the most frequently isolated serovars from food borne out breaks throughout the world (Herikstad et al., 2002).

The presented data in Table (7) showed that the incidence of *Salmonellae* in RTE meat products samples was 16, 12 and 8% in the examined samples of Luncheon, Frankfurter and Hot dog, respectively and serotyping of isolated *Salmonellae* revealed the presence of *S. Enteritidis*, *S. Virchow* and *S. Heidelberg* with different rates. These results were lower than that of Kasem, (2016) who found that *Salmonella* organisms were recovered from 20, 32 and 40% of the examined samples of luncheon, frankfurter and hot dog, respectively. Moreover, the obtained results disagreed with Amin and Abd El-Rahman, (2015) who could not isolate *Salmonella enterica* from ready to eat meat samples.

Despite the fact that *Salmonella* organisms exit all over the world, it does not mean that Salmonellosis should be accepted as inevitable but every defense must be considered through application of efficient sanitation to control such serious organisms.

**Table (1): Statistical analytical results of Aerobic plate count of RTE meat products**

Meat products (n=25/each)	Min.	Max.	Mean ± S.E.*	Samples exceed permissible limits	
				No	%
Luncheon	1.5×10 <sup>4</sup>	2.3×10 <sup>6</sup>	1.4×10 <sup>5</sup> ± 0.56×10 <sup>4</sup> b	4	16.0
Frankfurter	1.5×10 <sup>4</sup>	4.5×10 <sup>6</sup>	1.1×10 <sup>5</sup> ± 0.22×10 <sup>4</sup> a	14	56.0
Hot dog	5.5×10 <sup>4</sup>	2.5×10 <sup>6</sup>	4.1×10 <sup>5</sup> ± 0.74×10 <sup>4</sup> b	23	92.0

CFS, (2014) stated that APC in heat treated meat products should not more than 10<sup>5</sup> cfu/g

**Table (2): Statistical analytical results of Enterobacteriaceae count of RTE meat products**

Meat products (n=25/each)	Min.	Max.	Mean ± S.E.*	Samples exceed permissible limits	
				No	%
Luncheon	2.4×10 <sup>3</sup>	4.0×10 <sup>5</sup>	2.1×10 <sup>4</sup> ± 0.81×10 <sup>3</sup> b	10	40.0
Frankfurter	1.9×10 <sup>3</sup>	4.5×10 <sup>5</sup>	6.7×10 <sup>4</sup> ± 1.7×10 <sup>3</sup> a	11	44.0
Hot dog	3.0×10 <sup>3</sup>	3.9×10 <sup>5</sup>	6.8×10 <sup>4</sup> ± 1.4×10 <sup>3</sup> a	12	48.0

CFS, (2014) stated that Enterobacteriaceae count in heat treated meat should be less than 10<sup>2</sup> cfu/g.

**Table (3): Statistical analytical results of Coliforms count of RTE meat products**

Meat products (n=25/each)	Min.	Max.	Mean ± S.E.*	Samples exceed permissible limits	
				No	%
Luncheon	1.5×10 <sup>3</sup>	2.6×10 <sup>5</sup>	1.4×10 <sup>4</sup> ± 0.53×10 <sup>3</sup> b	7	28.0
Frankfurter	1.1×10 <sup>3</sup>	3.0×10 <sup>5</sup>	4.8×10 <sup>4</sup> ± 1.2×10 <sup>3</sup> a	4	16.0
Hot dog	1.3×10 <sup>3</sup>	2.9×10 <sup>5</sup>	4.7×10 <sup>4</sup> ± 0.92×10 <sup>3</sup> a	5	20.0

Egyptian Standard (3493/2005) stated that coliforms count in heat treated meat products should be not more than 10<sup>2</sup> cfu/g.

**Table (4): Statistical analytical results of molds count (cfu/g) of RTE meat products**

Meat products (n=25/each)	Min.	Max.	Mean ± S.E.*
Frankfurter	4.0×10 <sup>2</sup>	8.0×10 <sup>4</sup>	2.4×10 <sup>3</sup> ± 0.39×10 <sup>2a</sup>
Hot dog	2.0×10 <sup>2</sup>	9.0×10 <sup>4</sup>	1.9×10 <sup>3</sup> ± 0.39×10 <sup>2a</sup>

Egyptian Standard (3493/2005) stated that heat treated meat products must be free from molds.

**Table (5): Statistical analytical results of yeasts count (cfu/g) of RTE meat products**

Meat products (n=25/each)	Min.	Max.	Mean ± S.E.*
Frankfurter	9.0×10 <sup>3</sup>	1.7×10 <sup>5</sup>	4.9×10 <sup>4</sup> ± 0.8×10 <sup>3a</sup>
Hot dog	9.0×10 <sup>3</sup>	1.9×10 <sup>5</sup>	4.1×10 <sup>4</sup> ± 0.74×10 <sup>3a</sup>

Egyptian Standard (3493/2005) stated that heat treated meat products must be free from yeasts.

**Table (6): Prevalence of Enteropathogenic *E.coli* in RTE meat products**

Serotypes	Meat products	Luncheon (n=25)		Frankfurter (n=25)		Hot dog (n=25)	
		No.	%	No.	%	No.	%
O <sub>17</sub> : H <sub>18</sub> (EPEC)		3	12.0	3	12.0	2	8.0
O <sub>26</sub> : H <sub>11</sub> (EHEC)		2	8.0	1	4.0	0	0.0
O <sub>55</sub> : H <sub>7</sub> (EPEC)		1	4.0	2	8.0	1	4.0
O <sub>91</sub> : H <sub>21</sub> (EHEC)		3	12.0	0	0.0	0	0.0
O <sub>113</sub> : H <sub>2</sub> (EPEC)		1	4.0	2	8.0	1	4.0
<b>Total</b>		<b>10</b>	<b>40.0</b>	<b>8</b>	<b>32.0</b>	<b>4</b>	<b>16.0</b>

**Table (7): Prevalence of *Salmonellae* in RTE meat products**

Serotypes	Meat products	Luncheon (n=25)		Frankfurter (n=25)		Hot dog (n=25)	
		No.	%	No.	%	No.	%
<i>S. Enteritidis</i>		2	8.0	1	4.0	1	4.0
<i>S. Virchow</i>		1	4.0	1	4.0	0	0.0
<i>S. Heidberg</i>		1	4.0	1	4.0	1	4.0
<b>Total</b>		<b>4</b>	<b>16.0</b>	<b>3</b>	<b>12.0</b>	<b>2</b>	<b>8.0</b>

#### 4. Conclusion

The obtained result indicated that the Gram negative coliforms were present predominantly. In view of the microbial implication in handling, slaughtering, dressing, processing and distribution of meat and meat products which may endanger human health. Therefore, to avoid high bacterial load of meat products, the raw meat must be of very low initial bacterial count, application of the HACCP system during processing stages of such products, educational programs must be applied to the workers as learning of such workers about sources of contamination of products and personal hygiene such as, cleaning of their hands after toilet and wearing muzzles on mouth and nose, more over cleaning and sanitation of machines used for processing after each lot to avoid cross contamination.

#### Author contributions

All authors contributed to conception and realization of the work. All the authors have contributed to the paper redaction and given their approval to the final version of the manuscript.

#### Conflict of interests

The authors have not declared any conflict of interests.

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