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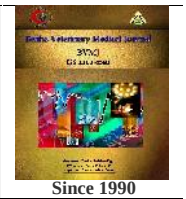

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Original Paper

Bacteriological profile of sheep carcasses in a private Egyptian abattoir

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ABSTRACT

Abomasal disorders can lead to severe and potentially life-threatening conditions for the Sheep are considered one of important protein sources in Egypt; due to its availability, nutritional value, and desirable taste, mutton and its edible offals are of good consumer demands. Hygienic profile of the slaughterhouse facilities and personnel handle fresh carcasses play the major role of its safety and compatibility for human consumption. Therefore, this study aimed to detect the bacteriological profile of ninety random fresh samples of sheep meat, liver, and lungs (30 of each) collected from immediately eviscerated sheep carcasses in private slaughterhouses in Eastern-Owinat, New Valley province, Egypt. Assessment of the bacteriological quality of the examined samples was conducted to investigate their hygienic quality and safety for human consumption. Results showed variable contamination levels in accordance of mean values of aerobic plate counts (APC), Enterobacteriacae counts (EC), coliform counts (CC), and Staphylococcus counts (SC). Accordingly, the liver samples were the highest contaminated samples with APC, EC, and CC with mean values of $3.06x10^5$, $2.1x10^4$, and $1.2x10^4$ CFU/g, respectively; while mutton samples were the most contaminated with SC with incidence and mean value of 8.0x10 (46.66%). Referring to *C. perfringens*, it was detected in 13.3% of the examined liver and lung samples, and 16.66% of examined meat samples; with mean values of 1.7x10, 1.3x10, and 1.5x10 CFU/g, respectively. The obtained results indicated that fresh mutton and edible offals may pose a risk to consumer's health in the absence of food safety knowledge and hygiene application, therefore, strict hygienic measures and authority inspection is strongly recommended

1. INTRODUCTION

Recently, the Egyptian authorities play huge steps to provide red meat of good quality especially with the continuous increase of human population followed by shortage in animal protein sources led the authorities to give much attention to compensate this shortage from other species such as sheep (Hatem et al., 2013).

In Egypt, sheep is an important source of red meat which distributed across the country, especially in the green Nile Valley and Delta at and around rural areas (Galal et al., 2005). It provides consumers with meat and offals of high-quality protein, minerals and vitamins as well as energy supplement (Campbell and Kenney, 1994).

Fresh meat has high decomposition affinity referring to its biological composition and the level of microbial contamination during slaughtering processes results in the spoilage of meat, reduces its shelf-life, and may posse public health hazards (Bacon et al., 2000). In addition, the microbiology of carcass meats is highly dependent on the conditions under which animals are reared, slaughtered and processed (DeWolf et al., 2012).

External contamination of slaughtered carcass is beginning from the moment of bleeding. There are large numbers of potential sources of bacterial meat contamination including contact with the hide, the content of gastrointestinal tracts, water supply, instruments used for dressing, improper worker's hygiene, and even air-borne contamination (Omer et al., 2015).

Contamination of fresh carcass during slaughtering, handling, and storage practices may constitute a public health hazard either due to presence of spoilage and/or pathogenic bacteria responsible for unfavorable changes leading to adverse effects on the consumer through food poisoning and other harmful effects (Abdalla et al., 2009).

Enterobacteriacae comprise different members of bacterial species acting as a hygienic indicator of overall wellness in the abattoir. Moreover, coliforms have been frequently isolated from meat indicating fecal contamination with the gastrointestinal tract content (Omer et al., 2015).

In addition, Staphylococci species as a versatile pathogen are commonly found on the skin and upper respiratory tract of man and animals and can easily contaminate the meat, therefore, presence of staphylococci in meat indicates contamination from the hides during evisceration, and/or contaminated equipment and worker's hands with abrasion and wounds (Murray, 2005).

Slaughtered carcasses may also be contaminated with *C. perfringens* spores through soil, intestinal contents, as well as water used in washing carcasses, cutting boards and by

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unhygienic methods of transportation to the butcher's shops (McEvoy et al., 2004). However, DHH (2015) revealed that vegetative *C. perfringens* can multiply very rapidly in food whereas its heat resistant spores allow its surviving adverse environmental effects. The spores germinate and multiply during cooling and holding of carcasses producing enterotoxins which is responsible for symptoms of *C. perfringens* food poisoning within 8 to 12 h after ingestion, which characterized by a sudden profuse watery diarrhea and moderate to severe cramps and epigastric pain.

Regular investigation of the bacteriological and hygienic quality of fresh meat and edible offals is essential for safety of human consumption; therefore, this study aimed to investigate the bacteriological profile of freshly slaughtered sheep carcass including meat and edible offals (liver and lung) through determination of their level of contamination with some bacteriological indicators.

2. MATERIAL AND METHODS

2.1. Samples:

A total of 90 random samples of fresh mutton, liver, and lung (30 of each) were collected from different sheep carcasses in Eastern-Owainat private abattoir, New Valley province, Egypt. Each sample weighed about 100 g was obtained immediately after removing of fat and connective tissue in the abattoir before chilling or other treatments. The collected samples were kept and transferred to the laboratory without undue delay for the bacteriological examinations.

2.2. Preparation of samples (APHA, 2001):

Twenty five grams of each sample were aseptically added to 225 ml of 0.1% peptone water, the contents were homogenized in stomacher at 450 to 640 strokes\min for 2 minutes; 1 ml of the homogenization was transferred into separate tubes containing 9 ml of sterile peptone water 0.1%, from which ten-fold serial dilutions were prepared.

2.3. Aerobic plate count "APC" (ISO 4833-1, 2013).
2.4. Enterobacteriacae count "EC" (ISO 21528-2, 2017).
2.5. Coliform count "CC" (ISO 4832, 2006).
2.6. Staphylococci count "SC" (ISO 6888-1:1999, A1:2003).
2.7. Detection and enumeration of viable C. perfringens (ISO 7937, 2004).
2.7.1. Identification of C. perfringens (ISO 7937, 2004):
2.7.1.1. Lactose sulfite (LS) broth inoculation
2.7.1.2. Nitrate reduction test
2.7.1.3. Sugar fermentation test

2.8. Statistical analysis:

The obtained results were statistically evaluated by application of Analysis of Variance (ANOVA) test according to Feldman *et al.* (2003).

3. RESULTS

The results of APC as summarized in table (1) declared that the mean values of APC of liver, lung and meat samples were $3.06x10^5 \pm 0.33x10^5$, $3.00x10^5 \pm 0.33x10^5$, and $2.85x10^5 \pm 0.38x10^5$ (CFU/g), respectively. A significant difference has been found between meat and other samples at ($P \leq 0.05$), while no difference was detected between liver and lung samples.

Table 1 Mean values of aerobic plate count (CFU/g) of the examined sheep samples (n=30)

Sample	Min.	Max.	Mean ± S.E.
Liver	1.93 $X10^5$	3.70 $X10^5$	$3.06x10^{5a}$±0.33$x10^5$
Lung	2.10 $X10^5$	3.90 $X10^5$	$3.00x10^{5a}$±0.33$x10^5$
Mutton	1.19 $X10^5$	3.60 $X10^5$	$2.85x10^{5b}$±0.38$x10^5$

Means within the same column of different litters were significantly different at ($P < 0.05$).

Regarding to the importance of Enterobacteriacae and fecal coliform contamination as mentioned in tables (2 & 3), the mean values of Enterobacteriacae and Coliform counts of liver, lung and mutton samples were $2.1x10^4 \pm 0.09x10^4$ and $1.2x10^4 \pm 0.07x10^4$, $1.5x10^4 \pm 0.07x10^4$ and $1.2x10^4 \pm 0.09x10^4$, $1.8x10^4 \pm 0.01x10^4$ and $1.0x10^4 \pm 0.05x10^4$ (CFU\g), with incidence of 33.33 and 26.66, 20.0 and 10.0, 26.66 and 20.00%, respectively. Significant difference was recorded between liver and both of mutton and lung samples at ($P \leq 0.05$).

Table 2 Mean values of Enterobacteriacae count (CFU/g) of the examined sheep samples (n=30)

Sample	+ve Samples		Count (CFU/g)		
	No.	%	Min.	Max.	Mean ± S.E.
Liver	10	33.33	$5.0x10^3$	$3.5x10^4$	$2.1x10^{4b}$±0.09 $x10^4$
Lung	6	20.00	$2.0x10^3$	$3.3x10^4$	$1.5x10^{4a}$±0.07$x10^4$
Mutton	8	26.66	12.0 $x10^3$	$3.5x10^4$	$1.8x10^{4a}$±0.01 $x10^4$

Means within the same column of different litters are significantly different at ($P < 0.05$).

Table 3 Mean values of coliform count (CFU/g) of the examined sheep samples (n=30).

Sample	+ve Samples		Count (CFU/g)		
	No.	%	Min.	Max.	Mean ± S. E.
Liver	8	26.66	$0.2x10^3$	$3.0x10^4$	$1.2x10^{4b}$±0.07$x10^4$
Lung	3	10.00	$0.8x10^3$	$2.9x10^4$	$1.2x10^{4b}$±0.09 $x10^4$
Mutton	6	20.00	$7.0x10^3$	$2.6x10^4$	$1.0x10^{4c}$±0.05$x10^4$

Means within the same column of different litters are significantly different at ($P < 0.05$).

Moreover, Staphylococci was detected in 26.66, 33.33, and 46.66% of the examined liver, lung, and meat samples with mean values of 6.7x10 ± 0.5x10, 7.7x10 ± 0.8x10, and 8.0x10 ± 0.6x10 (CFU/g), respectively. There were significant differences between all of the examined samples at ($P \leq 0.05$) as mentioned in table (4).

Table 4 Mean values of Staphylococci Count (SC) (CFU/g) of examined sheep samples (n=30)

Sample	+ve Samples		Count (CFU/g)		
	No.	%	Min.	Max.	Mean± S. E.
Liver	8	26.66	0.5 X10	3.2 $X10^2$	$6.7X10^{c}$±0.5 X10
Lung	10	33.33	0.2 X10	3.5 $X10^2$	$7.7X10^{b}$±0.8 X10
Mutton	14	46.66	0.2 X10	3.5 $X10^2$	$8.0X10^{a}$±0.6 X10

Means within the same column of different litters are significantly different at ($P < 0.05$).

Also, *C. perferingens* was detected in 13.33% of both liver and lung as well, and 16.66% of the examined meat samples with mean values of 1.7x10 ± 0.01x10, 1.3x10 ± 0.07x10, and 1.5x10 ± 0.09x10 (CFU/g) in liver, lung, and meat samples, respectively. Asignificant difference was reported between all of the examined samples ($P \leq 0.05$) as documented in table (5).

Table 5 Mean values of *C. perfringens* count (CFU/g) of the examined sheep samples (n= 30).

Sample	+ve Samples		Count (CFU/g)		
	No.	%	Min.	Max.	Mean ± S.E.
Liver	4	13.33	1.4x10	6.1x10	$1.7x10^{a}$±.01x10
Lung	4	13.33	0.9x10	4.2 X10	$1.3x10^{c}$±0.07x10
Muscle	5	16.66	1.1x10	5.4 X10	$1.5x10^{b}$±0.09x10

Means within the same column of different litters are significantly different at ($P < 0.05$).

4. DISCUSSION

From the obtained results, it is obvious that liver samples recorded the highest counts of APC and Enterobacteriacae, followed by lung and meat samples which may be explained due to improper practices during evisceration leading to contamination of liver as the first interior organ can be exposed to excreta during evisceration; in addition, lungs have been separated from GIT by mean of diaphragm; moreover, natural rigor mortis and ripening processes result in reduction of bacterial contamination in mutton samples.

The present results of APC of the examined samples as mentioned in Table (1) were somewhat agreed with those reported by Cohen et al. (2006) ($5.1x10^5$ and $3.9x10^5$ CFU/g in meat and liver samples); Bradeeba and Sivakumaar (2013) ($2.2x10^5$ CFU/g in meat), and Ibrahim et al. (2013) ($2.36x10^5$, $2.01x10^5$, and $1.89x10^5$ CFU/g in samples of meat, liver, and lung samples, respectively), while differed with those recorded by Abdalla et al. (2009) (ranged from $1.01x10^3$ to $4.4x10^3$ CFU/g in meat samples), Martineli et al. (2009) ($3.2x10^3$ CFU/g in lung samples), and Elshebrawy (2018) ($3.1x10^7$ CFU/g in meat samples).

The aerobic plate count has been used to assess sanitary quality and safety inspection of meat, which used as an indicator of bacterial population on a sample (APHA, 2001).

Referring to Enterobacteriacae counts as summarized in Table (2) were nearly similar to those recorded by El-Ebidy (2010) ($1.6x10^4$ CFU/g in liver), Aboukahf (2011) ($2.2x10^4$ CFU/g in meat), and Hassanin et al. (2013) ($3.96x10^4$ and $8.53x10^4$ CFU/g in liver and lung samples), while differed with those recorded by Ibrahim et al. (2013) ($1.08x10^5$ CFU/g in meat), Elshebrawy (2018) ($1.3x10^3$ CFU/g in meat).

High determination of Enterobacteriace in fresh meat and edible offals indicated enteric contamination with intestinal contents and declares the fact that the GIT is common habitat of the enteric bacteria and is considered the main source of contamination with these organisms during slaughtering, dressing, evisceration, handling and transportation to butcher shops (Hassanin et al., 2013).

As members of Enterobacteriacae, high coliform counts indicated poor hygienic and environmental quality of slaughterhouse and the carcass consequently. Contamination of fresh meat and edible offals with coliform bacteria begin with improper deskinning from the hide and hair of animal by knives and workers also during evisceration due to puncture of internal organs or from air, worker utensils or clothes , water used for carcass and offal wash (Abdalla et al., 2009). It is widely distributed on hands, bodies and cloths, air and soil, also utensils and equipment as well as in sewage and contaminated water; in addition, human and animals are the primary sources of the pathogenic microorganism that contaminated food and cause foodborne illness (EL-Ansary, 1997).

Results in Table (3) indicated that liver samples were higher coliform contamination than lung and mutton samples which indicated faecal contamination that may come through improper personal hygiene, contaminated hands and water supply may be.

Results of coliform counts are in agree with the results recorded by Aboukahf (2011) ($1.7x10^4$ CFU/g in meat), and Hassanin et al. (2013) (7.44×10^3 and 1.72×10^4 CFU/g in liver and lung samples), while disagreed with those reported by El-Shamy (2011) ($4.2x10^6$ and $4.2x10^6$ in liver and lung samples) and Saif Alnasr (2017) ($7.2x10^2$ CFU/g in meat).

The presence of Staphylococci on fresh carcass indicates low personal and instrumental hygiene especially during handling and storage which may be referred to contamination during dressing and evisceration in the slaughterhouse, as well as contaminated equipment, butcher's hands with abrasions and wounds. Slaughtering of animal beside dressed one in the same area in the slaughter hall, contaminated air from animal crowdedness and workers in the same place and their aerosols contamination with Staphylococci can be expected as it can be carried on human hands, nasal passages or throats (Lasta *et al.* 1992; Acco et al. 2003).

Regarding to Tables (4 & 5), mutton samples showed that highest incidence of Staphylococci and *C. perfringens*, followed by lung and liver, respectively; which may be attributed to that the meat is firstly exposed to contamination with hide, soil, and worker's hands during dehiding, followed by lung which represent the fact of that the respiratory system is the main predilection site of Staphylococci.

Staphylococci counts were in agree with those documented by Rao and Ramesh (1992) (4.2x10 CFU/g in meat), Cohen et al. (2006) ($1x10^2$ and $1.2x10^2$ CFU/g in meat and liver samples), while lower than those recorded by El-Kelany (2010) ($1.2x10^3$ CFU/g in meat), and Ibrahim et al. (2013) ($2.8x10^4$, $2.3x10^4$, and $2.0x10^4$ CFU/g in meat, liver and lung samples, respectively).

Moreover, *C. perfringens* results were agree with the results of Cohen et al. (2006) (1.9x10 and 1.2x10 CFU/g in meat and liver samples, respectively), while disagreed with that reported by Yegoraw et al. (2017) who failed to detect *C. perfringens* in their examined samples.

Clostridium perfringens is one of the most widespread pathogenic bacteria in the environment and is usually found in the GIT of healthy animals in low numbers, where it can contaminate animal carcasses during improper evisceration. Nonetheless, sulfite reducing clostridia, *C. perfringens*, is the most frequently involved anaerobic microorganisms in food spoilage and/or poisoning as it can grow inside the carcasses producing sensory and toxicological changes affecting its safety and quality for consumption (Tholozan et al., 1997). Moreover, its spores persist in soil, sediments, and areas subjected to human or animal fecal pollution (Brynestad and Granum, 2002).

Accordingly, the healthy status of the slaughtered animal, hygienic practices performed during slaughtering and processing; moreover, the temperature, time, storage and distribution conditions were important factors indicating the bacteriological quality of fresh meat sold by butchers (Nortije et al., 1990). Meat is highly susceptible to microbial spoilage, furthermore, it is frequently incriminated in foodborne outbreaks (Olaoye and Nilude, 2010); therefore, regular bacteriological assessment of slaughtered fresh carcasses is essential.

The result variations between different authors may be attributed to the differences in personal hygiene, hygienic practices during slaughtering, dehiding, evisceration, and transportation, different water sources, sources of sampling, and the health status of animals pre-slaughtering.

5. CONCULSION

From the obtained results, it can be concluded that the Egyptian sheep carcasses may be exposed to different

sources of contamination during slaughtering processes which referring it of inferior quality even possess health hazards to consumers. In addition, liver samples revealed the highest APC, EC, and CC contamination levels; while mutton samples were the most contaminated with Staphylococci and *C. perfringens*. It is recommending strict hygienic measures and personal hygiene should be applied in slaughterhouse and during slaughtering process should be applied.

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