BACTERIAL EVALUATION OF FROZEN CUT – UP DUCK MEAT

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A B S T R A C T

This study was conducted to evaluate microbiological contamination of frozen duck carcasses, and its hazards on public health. 80 samples taken from frozen breast and thigh duck meat (40 of each) from different retail shops were collected for bacteriological examination. The average of APC, Enterobacteriaceae, coliform and *staphylococcus aureus* counts were $9.27x10^4 \pm 2.16 x 10^{4++}$cfu/g, $7.85x10^3 \pm 1.24x10^{3++}$cfu/g, $1.70x10^2 \pm 0.41x10^{2+}$ cfu/g and $2.20x10^3 \pm 0.31x10^{3NS}$ cfu/g in the examined duck breast meat respectively. While for duck thigh meat they were, $3.08 x 10^5 \pm 0.59 x 10^5$ cfu/g, $9.13x10^4 \pm 1.71x10^4$ cfu/g, $3.29x10^2 \pm 0.56x10^2$ cfu/g and $2.96x10^3 \pm 0.47x10^3$ cfu/g respectively. The incidence of isolated *E. coli* was higher in breast than those isolated from thigh (8% and 4%), respectively. Moreover, the incidence of serologically identified *E. coli* as Enteropathogenic *E. coli* (*E coli* O_{55}:H_7 , E coli O_{78} and E coli O_{114}: H_{21}), Enterotoxogenic *E. coli* (*E. coli* O_{125}:H_{18}, *E. coli* O_{127}: H_6) Enteroheamorrhagic *E.* coli (*E. coli* O_{26}: and *E. coli* O_{111}:H_4) and Enteroinvasive *E. coli* (*E. coli* O_{124}). The public health importance of the isolated microorganisms and the suggestive hygienic measures to improve the safety of duck meat were discussed.

Keywords: Duck meat, Enterobacteriaceae, *staph. aureus*, coliform.

(BVMJ-26(2):30-39, 2014)

1. INTRODUCTION

Poultry production has been considered as one of the most important resources of animal production because of their rapid cycle, low price, high level of protein and low fat content consequently, duck meat have been recognized as an important source of protein for human consumption since the old Egyptian ages. In Egypt the peoples nowadays prefer to consume duck meat as it appears more palatable and duck meat contain more fat content in comparison with those of other poultry of similar age or weight (Auckland, 1973 and Brahma et al., 1987). In recent years ducks production has been increased in large scale, as the ducks rearing and management are usually easier in comparison with other poultry species as well as more resistance for diseases (Krogdahl, 1985). In addition, Duck and geese production accounts for about 7.5% of the total world poultry meat production (Pigel, 2004). Poultry carcasses and their parts are frequently contaminated with pathogens, which reach the carcasses from intestinal tract or from fecal material on feed and feathers (Dincer and Baysa, 2004). The level of Enterobacteriaceae as well as aerobic bacterial count in poultry carcasses can be routinely used as indicators of improper hygiene during processing and in correct storage conditions, which can lead to proliferation of pathogens (Robert *et al.*, 1995 and Zweifel *et al.*, 2005). Fecal coliform can be recorded in great numbers on freshly slaughtered carcasses; their

presence in meat generally indicates direct and indirect contamination of fecal origin, improper handling and storage (Charlebois *et al.*, 1991). In addition, *E. coli* was associated with human and animal infections causing suppurative lesions, neonatal septicemia and meningitis (Collins *et al.*, 1991). Moreover, *Staphylococcus aureus* is one of the most food poisoning microorganisms due to production of toxins Therefore, the aim of the present study was to evaluate the bacteriological status of frozen cut-up duck meat (breast and thigh) collected from different retail shops.

2. MATERIAL AND METHODS

2.1. Collection of Samples:

Agrand total of 80 random samples of frozen meat (without skin) of duck cuts classified into samples of breast and thigh (40 of each) were collected from different poultere›s shops at El-Kalyobia Governorate. The collected samples were transferred directly to the laboratory in an ice box under complete aseptic conditions without undue delay and then subjected to following examinations.

2.2. 2.2. Methods:

2.2.1. Preparation of Samples:

The samples were prepared according to the technique recommended by APHA (1992) as follows: twenty five grams of the examined duck meat samples were homogenized in a septic blender jar with 225 ml of 0.1 % sterile buffered peptone water at 2000 RPM for 1-2 minutes to provide a homogenate, from which tenth - fold serial dilutions were prepared. The prepared samples were subjected to the following examination:

2.2.2. Determination of Aerobic Plate Count: According to APHA (1992)

2.2.3. Determination of Enterobacteriaceae count: According to ISO (2004)

2.2.4. Determination of Coliform count: According to APHA (1992)

2.2.5. Isolation and identification of Escherichia coli.

According to (Cruickshank et al., 1975), (Mac Faddin, 2000), (Cheesbrough, 1985) and(Varnam and Evans, 1991)

2.2.6. Isolation and identification of Staphylococcus aureus:

According to ICMSF (1996), (Cruickshank et al.,(1975) and Bailey and Scott, (1978) and APHA,(1992)

3. 3- Results

It is evident from the results recorded in table (1) that APC in the examined samples varied from $2.0x10^3$ to $1.0\ x10^6$ with an average value of $9.27x10^4 \pm 2.16\ x\ 10^{4++}$ cfu/g and $4.0\ x10^3$ to $2.0\ x\ 10^6$ with an average value of $3.08\ x\ 10^5 \pm 0.59\ x\ 10^5$ cfu/g for the examined samples of duck breast and thigh meat respectively. In other words, there is a highly significant difference of APC between the examined duck meat (thigh and breast) *(P< 0.01).* The highest frequency distribution in breast samples was recorded within the range of 10^4- $<10^5$ (62.5%) followed by 10^5- $< 10^6$ (22.5%) and $10^{3-}< 10^4$ (12.5%) and 10^6-$< 10^7$ (2.5%) . while 60% of thigh samples was found within the range of 10^4- $< 10^5$, 32.5% within the range of 10^5-$< 10^6$ and 5.0% within the range of 10^6-$< 10^7$ and 2.5% within the range of 10^3-$< 10^4$ table (2)**.** It is evident from the resuls recorded in table(1) that Enterobacteriaceae in examined samples varied from $2.0x10^2$ to $4.0x10^4$ with an average value of $7.85x10^3 \pm 1.24x10^{3++}$cfu/g for samples of duck breast , and $8.0x10^2$ to$3.0x10^5$ with an average value of $9.13x10^4 \pm 1.71x10^4$ for duck thigh samples, respectively. In other words, there is ahighly significant difference of Enterobacteriaceae between the examined duck meat (thigh and breast) *(P< 0.01).* In table (3) the highest frequency distribution in breast samples was recorded within the range of 10^3-$<10^4$ (75.0%) followed by10^2-$<10^3$ (12.5%) and 10^4-$<10^5$ (12.5%). While (85.0%) of thigh

samples was found in range of 10^3-<10^4, (7.5%) within the range of 10^4-<10^5 and (5.0%) within the range of 10^5-<10^6 (2.5%) within the range of 10^2-<10^3. It is evident from the result recorded in table(1) that coliform count in examined samples varied from 3 to $4.5x10^2$ within an average value of $1.70x10^2 \pm 0.41x10^{2+}$ cfu/g for samples of duck breast, 1.1x10 to $5.0x10^2$ with an average value of $3.29x10^2 \pm 0.56x10^2$ for duck thigh samples, respectively. In other words, there is significant difference of *coliform* count between the examined duck meat (thigh and breast) *(P< 0.05).*

In table (4) the highest frequency distribution in breast samples was recorded within the range of 3-10^2 (92.5%) followed by 10^2-<10^3 (7.5%). While (87.5%) in thigh samples was found in range 3-10^2 , (12.5%) within the range of 10^2-<10^3. It is evident from the results recorded in table (1) that *S. aureus* in examined samples varied from $1.0x10^3$ to $4.0x10^3$ within an average value of $2.20x10^3 \pm 0.31x10^{3NS}$ cfu/g for samples of duck breast ,$8.0x10^2$ to $8.0x10^3$ with an average value of $2.96x10^3 \pm 0.47x10^3$ for duck thigh respectively. There is no significant differences associated with the examined duck meat (thigh and breast) for *staphylococcus aureus* count because the mean value is carrying the same litter in the same column. In table (5) the highest frequency distribution in breast samples was recorded within the range of <10(87.5%) followed by 10^2-<10^3 (12.5%).while (87.5%) in thigh samples was found in the range of <10**,** and **(**10.0%) within the range of 10^2-<10^3, and (2.5%) within the range of 10-<10^2. Results achieved in Table (6) indicated that *E.coli* was isolated from 8% and 4% of examined samples of duck breast and duck thigh, respectively. Moreover, the incidence of serologically identified *E. coli* as Enteropathogenic *E. coli* (*E coli* o_{55}:H_7 , E coli O_{78} and E coli O_{114}: H_{21}), Enterotoxogenic *E. coli* (*E coli* O_{125}:H_{18} *E coli* O_{127}: H_6) Enterheamorrhagic *E.* coli (*E coli* O_{26}: and *E coli* O_{111}:H_4) and Enteroinvasive *E. coli* (*E coli* O_{124})

4. DISSCUSION

It is evident from the result recorded in table (1) that the total APC in examined samples nearly similar to that obtained by Oumokhtar (2000) who menthioned that the mean value of aerobic plate count in chicken meat was $2.9x10^4$ cfu/g. Higher APC in duck meat obtained by Vural et al. (2006) who found that the mean value of APC was 1.48×10^7 in examined 25 chicken breast meat. The higher aerobic plate count in duck meat due to slaughtering and sale of chicken meat in the same place, which provokes the cross contamination of the carcasses. Moreover, the carcasses are kept at ambient temperature, which allow the multiplication of mesophilic micro-organisms. Moreover, the chopping tables, which manufactured from wood were found to be used every day without proper cleanliness. This enhanced the chance of cross contamination for uninfected carcass. As well as the processing of carcass into parts, lead to further spread of contamination by exposing more carcass surface and susceptible fleshy parts to the contaminants if the same cutting tables and knives are used (Satin, 2002). Enterobacteriaceae may be superior to the coliforms as indicators of sanitation (GMPs) because they have collectively greater resistance to the environment than the coliforms and can be colonized in an inadequate sanitation and are sensitive to sanitizers. Thus, the Enterobacteriaceae are useful for monitoring sanitation in food manufacturing plants (Kornacki and Johnson 2001). As well as the Enterobacteriaceae counts are used as a hygiene indicator of foods of animal origin (Arthur et al., 2004 and Crowley et al., 2005)**.** Nearly similar results were obtained by Kozacinski et al.(2006) who found the average number of Enterobacteriacea in chicken breasts with skin was $1.9 \times 10^2 \pm 0.33 \times 10$ cfu/g. Higher total Enterobacteriacae count was obtained by

Table (1): Statistical analytical results of APC, Enterobacteriaceae, Coliform and *S. aureus* counts(cfu/g) in the examined frozen cut-up duck meat samples, (n=40).

	Duck cut-up meat	Min	Max	Mean ± S.E*
APC	Breast	2.0×10^3	1.0×10^6	$9.27\times10^4 \pm 2.16\times10^4$ ++
	Thigh	4.0×10^3	2.0×10^6	$3.08\times10^5 \pm 0.59\times10^5$
Enterobactiacae. Count	Breast	2.0×10^2	4.0×10^4	$7.85\times10^3 \pm 1.24\times10^3$ ++
	Thigh	8.0×10^2	3.0×10^5	$9.13\times10^4 \pm 1.71\times10^4$
Coliform count	Breast	3	4.5×10^2	$1.70\times10^2 \pm 0.41\times10^2$ +
	Thigh	1.1×10	5.0×10^2	$3.29\times10^2 \pm 0.56\times10^2$
S. aureus count	Breast	1.0×10^3	4.0×10^3	$2.20\times10^3 \pm 0.31\times10^3$ NS
	Thigh	8.0×10^2	8.0×10^3	$2.96\times10^3 \pm 0.47\times10^3$

S.E* = Standard error of mean. ++ = High significant differences *(P<0.01).* + = Significant differences *(P<0.05)*. NS = Non-significant differences

Table (2): Frequency distribution of APC /cfu/g in the examined frozen cut –up duck meat samples (n= 40).

Duck cut-up meat	Breast		Thigh	
	No.	%	No.	%
$< 10^3$	-	-	-	-
10^3 - $< 10^4$	5	12.5	1	2.5
10^4 - $< 10^5$	25	62.5	24	60.0
10^5 - $< 10^6$	9	22.5	13	32.5
10^6 - $< 10^7$	1	2.5	2	5.0
Total	40	100	40	100

Table (3): Frequency distribution of Enterobacteriaceae cfu/g in the examined frozen cut-up duck meat samples (n= 40).

Duck cut-up meat	Breast		Thigh	
	No.	%	No.	%
$< 10^2$	-	-	-	-
$10^2 - < 10^3$	5	12.5	1	2.5
$10^3 - < 10^4$	30	75.0	34	85.0
$10^4 - < 10^5$	5	12.5	3	7.5
$10^5 - < 10^6$	-	-	2	5.0
Total	40	100	40	100

Table (4): Frequency distribution of coliform cfu/g in the Examined frozen cut-up duck meat samples (n= 40).

Duck cut-up meat / Interval (CFU/g)	Breast		Thigh	
	No.	%	No.	%
< 3	-	-	-	-
$3 - < 10^2$	37	92.5	35	87.5
$10^2 - < 10^3$	3	7.5	5	12.5
Total	40	100	40	100

Table (5): Frequency total distribution of *Staphylococcus aureus* cfu/g in the examined frozen cut-up duck meat samples (n= 40).

Duck cut-up meat / Interval (CFU/g)	Breast		Thigh	
	No.	%	No.	%
+ve samples	5	12.5	5	12.5
< 10	35	87.5	35	87.5
$10 - < 10^2$	-	-	1	2.5
$10^2 - < 10^3$	5	12.5	4	10.0
Total	40	100	40	100

Table (6): Incidence of serologically identified *E. coli* isolated from frozen cut-up duck meat samples (n= 40).

Duck cut-up meat / *E.coli* Strains	Breast		Thigh		Strain characteristics
	No.	%	No.	%	
O26	1	2.5	2	5	EHEC
O55 : H7	-	-	1	2.5	EPEC
O78	1	2.5	-	-	EPEC
O111 : H4	2	5	1	2.5	EHEC
O114 : H21	1	2.5	2	5	EPEC
O124	-	-	1	2.5	EIEC
O125 : H18	1	2.5	-	-	ETEC
O127 : H6	-	-	2	5	ETEC
Total	6	15	10	25	

EPEC = Enteropathogenic *E.coli.* ETEC = Enterotoxigenic *E.coli.* EIEC = Enteroinvasive *E.coli.* EHEC= Enterohaemorrhagic *E.coli*

Elias (1995) who examined bacteriologically samples from duck carcasses processed at home and poultry's shops and found that the mean value of *Enterobacteriaceae* count per gram was $47x10^6$. The high Enterobacteriaceae counts are an indication of potential microbial contamination during processing, distribution and storage. Their presence in large numbers in food indicates inadequate processing/or recontamination due to cross contamination by raw materials, dirty equipment or unhygienic handling (Ikeme, 1990). As well as presence of Enterobacteriaceae in the food is an indication of improper hygienic measures during the entire sequence of processing (Gill and Landers, 2004). Enterobacteriaceae have an epidemiological importance, as some of their members are pathogenic and may cause serious infections and food poisoning outbreaks to human being. Furthermore, the Enterobacteriaceae count can be taken as indicator of possible enteric contamination in the absence of coliform organisms (Mosupye and Van Holy, 2000). The current results were nearly similar with those obtained by Gad (2004) who examined microbiologically 80 samples of chicken breast and thigh (40 of each). He found that the mean values of total coliform counts were $5.12 \times 10^2 \pm 1.94 \times 10^2$ cfu/ g for breast and $3.44 \times 10^3 \pm 2.84 \times 10^3$ cfu/ g for thigh. Higher coliform count obtained by Chaiba et al. (2007) who found the mean value of coliform count of examined 24 chicken breast meat obtained from poultere s shops was $9.8 \times 10^3 \pm 0.23$ x10 cfu/g. High coliform count indicated poor hygienic quality of meat. The contamination with coliforms may occur during slaughtering, cutting or dressing of carcasses, soiled hands, shopping blocks or knives used for handling and cutting or contaminated water considered as an source of coliforms in meat (Yadav et al., 2006).

The presence of *S .aureus* in a food is usually taken to indicate contamination from the skin ,mouth or nose of workers handling product. Nearly similar results were obtained by Khalifa and Nassar (2001) who examined the bacteriological quality of breast and thigh meat in two game ducks (Pintail and Garganey). They found that the mean count of *S. aureus* in the breast meat of pintail was 3.1 log/g . Higher count obtained by Mohammed- Azza (2003) who mentioned that the *S. aureus* was recorded

in ducks processed in poultere›s shops was 23.3 x 10^3 in muscle. The presence of *S. aureus* in foods commonly indicates contamination that may be directly introduced into the food by workers who have skin lesions containing *S. aureus*, or sneezing or coughing. Presence of *E. coli* in meat indicates a general lack of cleanness during slaughtering, evisceration, dressing, transportation and handling of meat . As well as, *E. coli* may be used as an indicator microorganism because it provides an estimate of faecal contamination and poor sanitation during processing (Eisel *et al.*, 1997). Moreover, the incidence of serologically identified *E. coli* revealed that Enteropathogenic *E. coli* (*E coli* o_{55}:H_7 , E coli O_{78} and E coli O_{114}: H_{21}), Enterotoxogenic *E. coli* (*E coli* O_{125}:H_{18} *E coli* O_{127}: H_6) Enterheamorrhagic *E.* coli (*E coli* O_{26}: and *E coli* O_{111}:H_4) and Entero-invasive *E. coli* (*E coli* O_{124}). Nearly similar results were obtained by Hefnawy and Moustafa (1990**)** and Lee *et al.* (2009), Higher results were obtained by Cenci *et al*. (1992) and Cohen (2007). The presence of *E. coli* in high numbers indicates the presence of organisms originating from faecal pollution. This is due to improper slaughtering techniques, contaminated surfaces and/or handling of the meat by infected food handlers (Nel *et al.*, 2004). Also, the presence of these pathogens can be due to contamination taking place during the meat processing at slaughter house or due to the poor handling of the retailers of meat (Kagambèga *et al.*, 2011).

5. Conclusion

Duck carcasses examined in this study were subjected to various degree of contamination through duck processing specially during plucking and evisceration. Therefore, a concerted effort should be made to maintain sanitary condition in processing, preparation and handling. This can be controlled by applying Hygienic measures during slaughtering, struggling as well as efficient bleeding should be considered. All meat and poultry establishments develop and implement a system of preventive control designed to improve the safety of their products, known as HACCP (Hazard Analysis and Critical Control Points).

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الملخص العربي

لحم البط يعتبر من أشهى أنواع لحوم ا لدواجن وذلك لطعمه المميز واحتوائه على نسبة عالية من الدهون. كما يعتبر لحم البط من اللحوم الحمراء ذات البروتين عالي القيمة والذي يزداد الطلب عليه خاصة في المناسبات. ولكن ذبائح البط قد تتعرض أثناء تجهيزها للتلوث بالعديد من الميكروبات التي قد تصيب المستهلك بالعدوي المباشرة أو عن طريق إفرازالسموم أو قد تؤدي الي فساد المنتج. ولقد استهدفت هذه الدراسة الوقوف على مدي سلامة ذبائح البط المجمد في محيط محافظة القليوبية.
لذا أجريت هذه الدراسة بفحص عدد مائه (80) عينة عشوائية من لحوم البط(الصدور والأوراك) تم جمع هذه العينات من محلات مختلفة من محافظة القلوبية (بمعدل 40 لكل نوع) حيث أجريت الفحوص عليها لتحديد العدد الكلي للميكروبات الهوائية، الميكروبات المعوية والميكروبات القولونية و الميكروب المكور العنقودى الذهبى وكذلك عزل ألأيشريشيا كولاي وتصنيفه سيرولوجيا قد أظهرت النتائج ما يلي: أن متوسط العدد الكلى للميكروبات الهوائية لعينات لحم صدور وأوراك البط على التوالى 9.27×10^4 ± $2.16 \times 10^{4++}$ و3.08×10^5± 0.59×10^5 /جم. بينما كان متوسط العدد الكلى للميكروبات المعوية لعينات لحم صدور وأوراك البط على التوالى 7.85×10^3± $1.24 \times 10^{3++}$ و 9.13×10^4 ± 1.71×10^4 /جم.
على الجانب الآخر، كان متوسط العدد الكلى لميكروبات القولون لعينات لحم صدور وأوراك البط على التوالى 1.70×10^2± $0.41 \times 10^{2+}$ و 3.29×10^2± 0.56×10^2/جم.
بينما كان متوسط العدد الكلى للميكروبات العنقودية الذهبية لعينات لحم صدور وأوراك البط على التوالى2.20×10^3± 0.31×10^3و 2.96×10^3± 0.47×10^3/جم. وعلاوة على ذلك فقد تم عزل ميكروب الأيشريشيا كولاي من من عينات لحم صدور وأوراك البط بنسب 8% و4% على التوالي وبالتصنيف السيرولوجى تبين أن العترات المعزولة هي:
Enteropathogenic *E. coli* (*E coli* O_{55} *:*H_7, *E coli* O78 and *E coli* O_{114} :H_{21}), Enterotoxogenic *E. coli* (*E coli* O_{125}*:*H_{18} *and, E coli* O_{127}*:*H_6), Enterheamorrhagic *E. coli* (*E coli* O_{26} and *E coli* *O111:*H4) and Enteroinvasive *E. coli* (*E coli* O_{124}) . وقد تم دراسة ومناقشة الأهمية الصحية للميكروبات المعزولة ومصادر تلوث لحوم البط التي تم فحصها بالإضافة إلى اقتراح التوصيات اللازمة لضمان سلامة لحوم البط من التلوث من أجل حماية المستهلك.

(مجلة بنها للعلوم الطبية البيطرية: عدد 26(2):30- 39, يونيو 2014)