PJN
ISSN 1680-5194

PAKISTAN JOURNAL OF NUTRITION

ANSInet
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan
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Pakistan Journal of Nutrition 10 (3): 224-228, 2011
ISSN 1680-5194

Enumeration of Listeria and Enteric Bacteria of Public Health Significance on Meat Tables Before and After Sales of Meat in Ibadan Municipal Abattoir, Nigeria

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Abstract: A bacteriological assay was done on meat tables in Ibadan municipal abattoir, Nigeria. Swab samples from 1 cm^2 of the surface were obtained from meat tables before and after the sales of meat. The swab samples were analyzed for *Listeria* sp Count (LSC), *Listeria monocytogenes* Count (LMC), Coliform Count (CC), *Enterobacteriaceae* Count (EC) and *Salmonella* sp Count (SSC). The mean values for LSC, EC and SSC were 8.20±0.06 $log_{10}CFU/cm^2$, 8.81±0.05 $log_{10}CFU/cm^2$ and 7.32±0.20 $log_{10}CFU/cm^2$ respectively for before sales while after meat sales was 10.47±0.05 $log_{10}CFU/cm^2$ (LSC), 11.47±0.03 $log_{10}CFU/cm^2$ (EC) and 11.37±0.04 $log_{10}CFU/cm^2$ (SSC). The mean values for before and after meat sales of Coliform Count (CC) were 8.35±0.07 $log_{10}CFU/cm^2$ and 10.86±0.05 $log_{10}CFU/cm^2$ respectively while *Listeria monocytogenes* Count (LMC) for before and after meat sales were 7.59±0.06 $log_{10}CFU/cm^2$ and 9.78±0.07 $log_{10}CFU/cm^2$ respectively. The mean values ($log_{10}CFU/cm^2$) showed that EC> LSC>SSC for before meat sales while after sales showed that EC>SSC>LSC. A significat difference ($p<0.05$) was observed in all the counts before and after meat sales. A positive correlation ($R^2 = 0.396$) ensued for LMC and LSC before sales. Results reflect poor hygienic conditions of the meat tables and non adherence to HACCP programs.

Key words: Meat tables, hygienic conditions, food borne diseases

INTRODUCTION

Wood has been used for many centuries in the preparation and display of meat products. Recently, the use of wood in the meat industry worldwide has been diminishing, especially in the areas of meat processing, packaging and transportation aids. This is mainly because of modifications brought to the regulation in developed world. In Nigeria, wood is still been used for the production of meat carcasses and construction of retail meat tables. Wood is a porous and absorbent material where organic matter along with bacteria can become entrapped; cross-contamination is a main concern (Lauzon, 1998).

Meat is an ideal medium for the development and reproduction of micro-organisms particularly bacteria and rapid growth can be expected unless control is effected. Microbial contamination can cause spoilage of meat, reduces shelf life of meat and causes public health hazards (Rao, 1992). The microbial contamination of carcasses occurs mainly during processing and manipulation such as skinning, evisceration, storage and distribution at slaughterhouses and retail establishments (Gill, 1998; Abdalla *et al*., 2009). In developing countries, some traditional methods of handling, processing and marketing of meat undermine quality whereas poor sanitation leads to considerable loss of product as well as to the risk of food-borne disease (Garcia de siles *et al*., 1997).

Bacteria which are responsible for the most food borne disease contaminate meat directly and indirectly from animal excreta at slaughter process; also they may be transferred from the surfaces, utensils and other equipment (Arnold International Students, 1993; Yen, 2003). The external contamination of meat constitutes a constant problem in most developing countries abattoirs where they are potential sources of infection (Lawrie, 1979). The microbial surface contamination of carcasses has been repeatedly reported to have a significant effect on the meat shelf life. Moreover, Contaminants may also include pathogens which can penetrate into the meat (Elmossalami and Wassef, 1971).

Slaughtering is a suitable progress for the contamination of the carcass by partially pathogenic bacteria (Forsythe and Hayes, 1998) so that all surfaces in contact with meat should be taken under control or kept clean to minimize the risk of bacteria contamination (Butterorth-Heineinann, 2000). Unsanitary methods spread such diseases as Salmonellosis, Cholera, *E. coli* food poisoning and Listerosis that cause contamination of the meat, a serious public health concern (Neil Trent *et al*., 2002). Fecal matter is a major source of contamination and could reached carcasses through direct deposition as well as by indirect contact through contaminated and unclean carcasses equipment, surfaces, workers, installations and air (Borch and Arnder, 2002). Aerobic Plate Count (APC) and

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Enterobacteriaceae Count (EBC) are generally used as hygienic indicators in the food chain (Warriner *et al*., 2002; Nel *et al*., 2004; Zweifel *et al*., 2005). Aerobic Plate Count (APC) depicts general microbial contamination. The *Enterobacteriaceae* Count (EBC) is a marker of possible fecal contamination. Feaces are the main source of pathogens such as *E. coli* 0157:H7, *Salmonella* or *Campylobacter* (WHO, 1995). Anonymous (2001) stated that *Enterobacteriaceae* count becomes a public health issue when the count is above acceptable value of 2.0logCFU set by decision of 2001/471/EC of the European commission. A standard of less than 1.3 log10 CFU and 1.3 log10 CFU were used for aerobic plate count and coliform count respectively by decision of 2001/471/EC of the European commission.

Ibadan Municipal Abattoir is a major Abattoir located in Ibadan North local government area of Ibadan, Oyo State, Nigeria. Ibadan is the largest city in West Africa and the second largest In Africa with land size covering an area of 240 km^2 and with human population of over 2 million by 2005 census. The city is located on geographic grid longitude 35°. SE, latitude 7°22 (Filani, 1994). Animals slaughtered in Bodija abattoir alone accounts for 65.23% of the total animal in Oyo State (Abiola, 1995).

This study enumerated *Listeria* sp., *Listeria monocytogenes*, *coliform*, *Enterobacteriaceae* and *Salmonella* sp. of bacteria loads of public health importance on meat tables before and after the sales of meat and to determine the bacteriological quality of the meat tables.

MATERIALS AND METHODS

Study design: The study was to assess the bacteria loads of public health importance viz *Listeria* sp., *Listeria monocytogenes*, *Enterobacteriaceae* and *Salmonella* sp. before and after the sales of meat on 100 meat tables at different locations in Ibadan municipal, abattoir, Nigeria. The tables were selected by stratified random sampling methods. A total of 100 samples were collected from the surfaces of meat tables. Fifty samples were collected before meat sales (7.00 am) and remaining fifty samples collected after meat sales (4.00 pm). During collection, sterile swab sticks were soaked with sterile distilled water and placed on a measured 1 cm^2 as a point of collection on a meat table. The samples were kept in packs on transit to the laboratory to help maintain the microbial loads as described in the compendium of methods for the microbial examination of foods (American Public Health Association, 1992).

Bacteriological assessment: 1 ml of the homogenized swab samples were suspended in 9 ml of sterile peptone water and vortexed. A dilution factor of 10^{-6} (before meat sales) and 10^{-8} (after meat sales) was used. 0.1 ml of the appropriate dilution was inoculated onto surfaces of three selective agar plates namely Deoxycholate citrate Hyne agar for *Salmonella* (Biotec lab. Ltd., UK.), *Listeria* selective agar (with antibiotic supplement) for *Listeria monocytogenes* (Fluka 7014, Germany.) and MacConkey for *Enterobacteriaceae* (Biotec lab. Ltd., UK.). All the samples were done in replicates and incubated at 37°C for 18-24 h. The enumeration of bacteria isolates was carried out according to Barrow and Feltham (1993). The plates were assayed for *Listeria* sp. Count (LSC), *Listeria monocytogenes* Count (LMC), Coliform Count (CC), *Enterobacteriaceae* Count (EC) and *Salmonella* sp. Count (SSC) were done according to method specified in FDA (Food and Drug Administration, 1995). Colony Forming Units (CFU) per ml of sample were calculated using the dilution factor of each and converted to $log_{10}CFU/cm^2$ values. The counts were reported as means ± Standard Error of Mean (SEM).

Statistical analysis: The data were analyzed using SPSS 15 software 2006 (Statistical package for the social science SPSS Inc. and Chicago, IL, USA.) All bacteria counts were converted to $log_{10}CFU/cm^2$ for analysis and ANOVA was performed. Statistical significance was set at a P value of <0.05 and $p<0.01$. Charts was plotted using Microsoft Excel 2009.

RESULTS AND DISCUSSION

The mean values for *Listeria* sp Count (LSC), *Enterobacteriaceae* Count (EC) and *Salmonella* sp. Count (SSC) were 8.22$log_{10}CFU/cm^2$, 8.81$log_{10}CFU/cm^2$ and 7.32$log_{10}CFU/cm^2$ respectively for before sales with significant differences ($p<0.05$) between them while after meat sales were 10.47$log_{10}CFU/cm^2$ (LSC), 11.47$log_{10}CFU/cm^2$ (EC) and 11.37$log_{10}CFU/cm^2$ (SSC). (Table 1). The Coliform Count (CC) mean values were 8.35$log_{10}CFU/cm^2$ and 10.86$log_{10}CFU/cm^2$ before and after meat sales respectively while *Listeria monocytogenes* Count (LMC) was 7.59$log_{10}CFU/cm^2$ before meat sales and 9.78$log_{10}CFU/cm^2$ after meat sales. The descending trend of mean values ($log_{10}CFU/cm^2$) before meat sales was EC> LSC>SSC whilst after sales was EC>SSC>LSC (Fig. 1). A significant difference ($p<0.01$) in all the counts was observed between before sales and after sales (Table 2 and Fig. 1). There was a positive correlation between *Listeria* sp Count (LSC) and *Listeria monocytogenes* Count (LMC) (Table 3 and Fig. 3). Also, between *Enterobacteriaceae* Count (EC) and Coliform Count (CC) (Table 3). A correlation significant exists (R^2) in before sales between *Listeria monocytogenes* Count (LMC) and *Listeria* sp Count (LSC) (Table 3).

Table 1: Mean values of *Enterobacteriaceae*, *Listeria* and *Salmonella* counts in Log_{10}cfu/cm^2 before and after sales

Variables (n)	*Enterobacteriaceae* count	*Listeria* sp. count	*Salmonella* sp. count
Before sales (50)	8.81±0.05^a	8.20±0.06^b	7.32±0.20^c
After sales (50)	11.47±0.03^a	10.47±0.05^a	11.37±0.04^a
	Coliform count	LM count	
Before sales (50)	8.35±0.07^b	7.59±0.06^b	na
After sales (50)	10.86±0.05^a	9.78±0.07^a	na

Means with the same superscripts are not significantly different at 0.05 levels values represent; Mean LogCFU/cm^2 ± standard error of mean; na-not applicable

Table 2: Mean values comparing counts in Log_{10}cfu/cm^2 before and after meat sales within each group of organisms

Specie	Variable (logcfu/cm^2)	n-50	Mean (log_{10}CFU/cm^2)	t
Enterobacteria	Coliform count	Before sales	8.35±0.07	-27.22**
		After sales	10.85±0.06	
	Enterobacteriaceae	Before sales	8.81±0.05	-40.66**
		After sales	11.47±0.04	
Listeria sp.	LM Count	Before sales	7.62±0.06	-28.48**
		After sales	9.78±0.07	
	Listeria sp.	Before sales	8.20±0.06	-29.59**
		After sales	10.47±0.06	
Salmonella sp.	*Salmonella* sp.	Before sales	7.32±0.20	-20.69**
		After sales	11.40±0.04	

**Mean t-value significant at 0.01 level of significance

Table 3: Correlation between same group of organisms before and after meat sales

Specie	Variable (logcfu/cm^2)		Mean (log_{10}CFU/cm^2)	Correlation (R^2 values)
Enterobacteriace	Coliform count	Before sales	8.35±0.07	0.056
	Enterobacteriaceae	Before sales	8.81±0.05	
	Coliform count	After sales	10.85±0.06	0.068
	Enterobacteriaceae	After sales	11.47±0.04	
Listeria sp.	LM Count	Before sales	7.62±0.06	0.396*
	Listeria sp.	Before sales	8.20±0.06	
	LM Count	After sales	9.78±0.07	0.127
	Listeria sp.	After Sales	10.47±0.06	

*Correlation significant at 0.05 level of significance

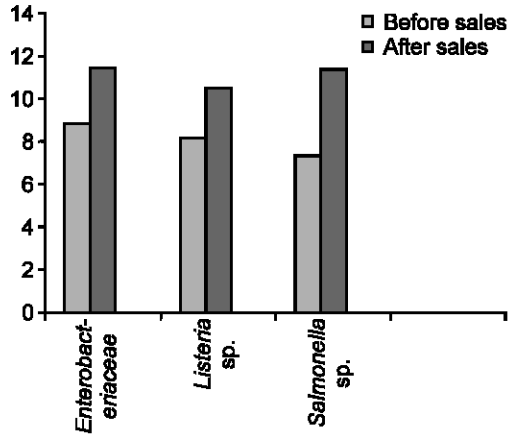

Fig. 1: *Enterobacteriaceae*, *Listeria* and *Salmonella* sp. counts before and after sales

Fig. 2: Comparison between *Listeria* sp. and *Listeria monocytogenes* counts

Meat carcasses are traditionally processed and marketed on wooden surfaces (meat tables) in Ibadan municipal abattoir, Nigeria thus allowing for environmental contamination. Microbial contamination of meat results in spoilage, reduced shelf life and cause public health hazards. It is generally accepted that microbial loads on surfaces and equipment vary in different food plants depending on the microbial quality of the food (Evans *et al*., 2004).

Fig. 3: Comparison between enterobacteriaceae and coliform count

The mean values ($log_{10}CFU/cm^2$) showed that EC predominates and coliform, CC followed by LSC and its virulent strain: LMC and the least being SSC were present on meat table before the meat sales while EC predominates and coliform followed by SSC and LSC and its virulent strain: LMC. The $log_{10}CFU/cm^2$ values obtained for EC and CC were higher than the other counts, this in agreement with the work of Warriner *et al.* (2002); Nel *et al.* (2004); Zweifel *et al.* (2005) in which Aerobic plate count and EC were reported as hygienic indicators in food chain. Aerobic plate count depicts general microbial contamination and EC is a marker of possible fecal contamination. The extremely high count found this study could be as result of high level of unhygienic practices by the abattoir workers and the wood surface used.

The mean count for *Enterobacteriaceae* (8.81-11.47$log_{10}CFU/cm^2$) and coliform (8.35-10.86$log_{10}CFU/cm^2$) were higher than standards used in food processing industry as set by decision of 2001/471/EC of the European commission (Anonymous, 2001). A standard of less than 1.3logCFU/cm^2 was set for aerobic plate count, less than 2.0logCFU/cm^2 for EC and less than 1.0logCFU/cm^2 for CC. The *Enterobacteriaceae* and coliform counts in this study were also higher than reports made in earlier works done on meat contact surfaces by Samaha and Draz (1993) in which he reported that EC and CC were 5.0logCFU/cm^2 and 3.68logCFU/cm^2 of cattle carcasses surfaces inside the slaughter halls in Alexandria city, Egypt. Omer Cetin *et al.* (2006) reported 3.0logCFU/cm^2 for total aerobic count, 2.08logCFU/cm^2 for CC and 1.23logCFU/cm^2 for EC. The high EC and CC values obtained from the meat tables before and after meat sales must be decreased by applying HACCP procedures because *E. coli* and its subspecies and coliform group bacteria can cause serious problems on public health. Thus, it is an obligation to eliminate or minimize the presence of bacteria (Lowe *et al.*, 2001). Kusumaningrum *et al.* (2003) reported that training and supervision to ensure proper hand washing and appropriate cleaning and sanitation procedures will reduce cross-contamination.

Salmonella was all samples after sales following 48 h of incubation. This could be attributed to increase contamination from human factors. In addition *Salmonella* sp. need a longer incubation time (48 h) for growth when compared to *E. coli*.

The results in this study indicates that the bacteriological quality of the meat tables were found to be higher when compared to European commission standards and other similar works done by Samaha and Draz (1993) and Omer Cetin *et al.* (2006) for *Enterobacteriaceae* count and coliform count. The counts obtained for after sales showed increased environmental contamination and bacteria multiplication. Results also reflect poor condition of our meat table where carcasses are placed and unhygienic practices involved in the abattoir. The levels of microbial contamination of meat tables in Ibadan Municipal Abattoirs reflect the hygienic status of meat production in the developing world and meat tables made of wood. Wood as a food contact surface has diminished because is a porous and absorbent material where organic matter along with bacteria can become entrapped; cross-contamination is a main concern (Lauzon, 1998). Glass is sometimes used for food contact surfaces because of its smooth and corrosion-resistant surface (Dunsmore *et al.*, 1981). Stainless steel resists impact damage better than glass but is vulnerable to corrosion, while rubber surfaces are prone to deterioration and may develop surface cracks where bacteria can accumulate (LeClercq-Perlat and Lalande, 1994). Equally important in meat contact surface is the clean ability of the surface once bacteria forming have attached. This is a major concern in meat processing surfaces. Furthermore, high values showed that the effective cleaning, sanitation programs and safe handling procedures were not applied as required on the meat tables. The production of good quality and safe meat products will be possible by HACCP procedures. One of the most important steps in developing a HACCP system is the establishment of critical control points. In terms of fresh meat processing, safety hazards cannot be eliminated but they can be prevented or reduced. (Pearce *et al.*, 2004). Other control measures should include an extensive education programs for proper hygiene, improvement of managements and introduction of different surfaces other than wood for the display of meat.

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