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Microbial Hazard Analysis of Retail Poultry Meat Sold in Unorganized Slaughter Units in Parbhani City of Marathwada Region of Maharashtra State

Patil S. S., Deshmukh V. V.*, Waghmare R. N. and Yeotikar P. V.

Department of Veterinary Public Health & Epidemiology, College of Veterinary & Animal Sciences, Maharashtra Animal & Fishery Sciences University, Parbhani-431 402

*Corresponding Author E-mail: vivekdes@gmail.com

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ABSTRACT

The present study was planned with the objectives to study the microbial load of retail poultry meat sold in unorganized slaughter units in Parbhani city of Marathwada region of Maharashtra State, to detect source of contamination of the retail poultry meat, isolation, enumeration and identification of bacteria of public health significance. A total of 240 swab samples from different sources viz. knife, scalding tank, defeatherer, dressing table, platform, personnel, water and poultry meat, collected from five retail poultry meat shops in six lots, were assessed for microbial quality by evaluating Total Viable Count (TVC), Escherichia coli count, staphylococcal count and by detection of Salmonella. Isolation, identification and enumeration were done by following standard methods. Microbiological processing of 240 samples revealed highest TVC (log CFU/cm^2±SE) of dressing table (6.14±0.04) and platform (6.01±0.05). Mean TVC of other sources in poultry meat shops were found as poultry meat (5.90±0.03), knife (5.85±0.05), defeatherer (5.89±0.06), personnel (5.74±0.06) and water (5.82±0.06). The mean TVC of scalding tank (5.67±0.05) was lowest. Total mean Escherichia coli and staphylococcal counts from all sources was 3.61±0.17 and 3.24±0.14 respectively. The staphylococci were isolated from maximum (34.2%) samples, followed by Escherichia coli (30.8%) and Salmonella (4.6%).The results indicate that the dressing table and platform are the major sources responsible for bacterial contamination of retail poultry meat sold in Parbhani city.

Key words: *Poultry meat, Quality, TVC, Escherichia coli, Staphylococci, Salmonella*

INTRODUCTION

Poultry meat industry is advancing from being an unorganized sector retailing to the large organized sector processing and marketing by way of more modern poultry processing plants, however in the semi-urban areas like Parbhani, the sale is mainly sourced from retail poultry shops only.

Foodborne infections and intoxications have always been important, however given the increased public awareness and media in recent years, assumed more significance as a health hazard.

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The presence of pathogenic and spoilage microorganisms in poultry meat and byproducts remains a significant concern for suppliers, consumers and public health officials worldwide[1]. There have been a number of food-borne illnesses resulting from the ingestion of contaminated foods such as chicken meats as the pathogens that play role in foodborne diseases have a zoonotic origin[2]. These pathogens cause mild to moderate self-limiting gastroenteritis to full-blown, invasive diseases and severe disease complications[3]. Numerous epidemiological reports have implicated foods of animal origin as the major sources associated with illnesses caused by food-borne pathogens[4]. Contaminated raw or undercooked poultry and red meats are particularly important in transmitting these food-borne pathogens[3].

Special attention in poultry meat production is paid to the fact that live animals are hosts to a large number of different microorganisms residing on the skin, feather or in the alimentary tract. During slaughter most of these microorganisms are eliminated, but the subsequent contamination is possible at during feather plucking, evisceration and washing to storage[5].

Total bacterial counts have been used as indices of safety and sanitary conditions under which the food product has been prepared[6]. Foodborne *Escherichia coli* are important pathogens of public health importance. *Escherichia coli* is often suggested as indicator organism because it reliably reflects fecal contamination and indicates a possible contamination of enteric pathogens[7]. *Staphylococcus aureus* is an important organism in relation to food hygiene because many strains produce enterotoxins staphylococcal food poisoning[8].

People from semi-urban area are still using traditional methods of poultry processing. The poor hygienic conditions at retail meat shops due to lack of knowledge of butchers results in the consumers becoming vulnerable to zoonotic food-borne hazards. Therefore the present study was done to study epidemiology of foodborne pathogens in Parbhani city.

MATERIAL AND METHODS

Sample collection and processing: A total of 240 samples were collected from five retail poultry meat shops in Parbhani city. Eight different sources viz. knife, scalding tank, defeatherer, dressing table, platform, personnel, water and poultry meat from each of five poultry meat shops were used for sample collection. A total of six lots from each of eight sources of each shop were collected. For collection of swab samples, sterile cotton swabs (3 cm long and 1 cm in diameter) moistened with 1% peptone water was used. An area of 1 cm^2 was marked with sterile frame of 1 cm x 1 cm for each site. Swabs were rubbed on sites continuously for 30 seconds[9]. Water samples were collected in sterile screw cap test tubes. Poultry meat samples were collected in sterile polyethylene sachets. All these samples were collected for the period from December 2010 to May 2011. Collected samples were brought to the laboratory on ice within one hour and processed immediately.

Processing of samples

Processing of swab samples and bacterial isolation was carried out by the standard methods[10, 11]. A quantity of 10gm of beef sample was minced with the help of sterile scissors and mixed in 90 ml normal saline solution (pH 7.2) in screw cap bottle. Ten fold serial dilutions were made up to 10^{-8} dilution in normal saline solution (pH 7.2). A quantity of 0.1 ml inoculum from 10^{-3} and 10^{-4} dilutions was used by spread plate technique on Plate Count Agar (Hi Media Laboratories, Mumbai) for enumeration of TVC, on Eosin Metheline Blue agar (EMB) (Hi Media Laboratories, Mumbai) and Baird Parker agar (BPA) (Himedia Laboratories, Mumbai) for isolation and enumeration of *Escherichia coli* and staphylococci respectively. Incubation was done at 37^0 C for 24 hours. TVC and enumerations was done by standard formula and expressed as log CFU/cm^2, TVC of water samples was expressed as log CFU/ml and TVC of poultry meat was expressed as log CFU/gm. Typical characteristics colony of *Escherichia coli* on EMB agar as greenish

metallic sheen was counted and isolated. For enumeration and isolation of staphylococci typical black colonies with light yellow zone were considered and enumerated.

For the isolation of *Salmonella* all the samples were pre-enriched in Buffered Peptone Water (Himedia Laboratories, Mumbai) (pH 7.0±0.2) at 37^0 C for 24 hours. Each pre-enriched sample was enriched by using Tetrathionate Broth at 42^0 C for 24 hours. Enrichment of sample was followed by selective plating on Xylose Lysine Deoxycholate (XLD) (HiMedia Laboratories, Mumbai) agar at 37^0 C for 24 hours. Typical *Salmonella* colonies showing black colour with dark centre were considered as positive.

Characterization and identification of isolates

Each isolate of organism obtained on selective medium was further characterized for morphological characterization by using Gram's staining method. Identification of *Escherichia coli, Staphylococcus and Salmonella* was done by biochemical test i.e IMViC, catalase, oxidase, nitrate reduction, H_2S production (for *Salmonella*), coagulase test (for *Staphylococci aureus*), sugar fermentation and motility tests[12].

Statistical analysis

Completely Randomized Design (CRD) was used by using Generalized Linear model with the help of SYSTAT® SOFTWARE VERSION 7.0.

RESULTS

Total Viable Count (TVC) from each source consisting of knife, scalding tank, defeatherer, dressing table, platform, personnel, water and poultry meat was calculated. The results were subjected to generalized linear model by using SYSTAT® software. A highly significant ($p<0.01$) effect of all sources on mean TVC was observed during the study. Comparison of mean TVC between the sources reveals that the dressing table (6.14±0.04) and platform (6.01±0.05) were showing high mean TVC. Both the sources were at par with each other. Retail poultry meat revealed a mean TVC (5.90±0.03) at par with knife (5.85±0.05), defeatherer (5.89±0.06) indicating microbial contamination among these sources. Comparison of mean TVC of personnel (5.74±0.06) with rest of the sources indicated that the water (5.82±0.06) is the only source at par with each other. The scalding tank (5.67±0.05) is the only independent source of bacteria apart from dressing table (6.14 ±0.04). In present study knife, dressing table, platform, personnel and water were found to be main sources of bacterial contamination in retail poultry meat. The TVC of 5.67±0.05 to 6.14±0.04 was observed from all eight sources during study period are shown in details in Table 1. and Fig 1.

Mean *Escherichia coli* counts of different sources were statistically compared to study their effects on *Escherichia coli* counts. Non significant ($p < 0.01$) effect of sources was observed upon mean *Escherichia coli* counts. Mean *Escherichia coli* counts of all the sources are at par ranging from 3.27±0.21 to 3.98±0.44. An attempt was made to obtain *Escherichia coli* isolates from all 240 samples from the 8 sources. A total 74 *Escherichia coli* isolates were obtained from all the sources with overall prevalence of 30.8 % as shown in Table 1. The highest %age of positive samples 46.6 % was seen from dressing table and poultry meat. The lowest %age of positive was observed in personnel (6.6 %). It is clearly evident that dressing table is one of the major sources of *Escherichia coli* contamination followed by water (43.3 %), scalding tank (33.3 %), defeatherer (23.3 %) and platform (16.6 %).

Similar results were observed *as* non significant ($p<0.01$) effect of sources was observed upon mean *Staphylococcal* counts. Mean staphylococcal counts of all the sources are at par ranging from 3.49±0.10 to 3.84±0.14. A total of 82 staphylococcal isolates obtained with overall prevalence of 34.17 % from 240 samples of 8 different sources as shown in Table 1. The highest numbers of Staphylococci were obtained from dressing table (53.33 %) followed by platform (43.33 %), poultry meat (40 %), defeatherer (36.67 %) and knife (33.33 %).

A total of 11 *Salmonella* isolates were obtained from 240 samples with overall prevalence of 4.58 % as shown in Table 1. One isolate of *Salmonella* was obtained from defeatherer having 3.33 % positivity. In a total of 4 samples *Salmonella*e were isolated from dressing table with 13.33 % positivity. Isolation of *Salmonella* from water comprises of 2 isolates with 6.67 % positivity and 4 *Salmonella* isolates were obtained from retail poultry meat with 13.33 % positivity

DISCUSSION

The TVC of 5.67±0.05 to 6.14±0.04 was observed from all eight sources during study period. The TVC of different sources in poultry meat shops observed in excess of 5.0 log CFU/cm^2 has been reported earlier[13,14]. In Present study the *Escherichia coli* count and staphylococcal count were observed within the range of 3.27±0.21 to 3.98±0.44 and 3.49±0.10 to 3.84±0.14 respectively. Earlier worker reported range 2.60 to 4.33 for *Escherichia coli* and 2.47 to 3.48 for *Staphylococcus*.[15]. Investigation of poultry slaughterhouses for microbial analysis to determine bacterial plate count, *Escherichia coli* and *Salmonella* count has been reported[16], and our results are on similar line.

In present study a total 74 *Escherichia coli* isolates were obtained from all the sources with overall prevalence of 30.8 %. Earlier workers also reported prevalence of *Escherichia coli* near about the results shown by the present study[17]. In the same trial a total of 82 positive samples of staphylococci were obtained with overall prevalence of 34.2 %. At last the attempt was done for the isolation of *Salmonella*, a total of 11 isolates of *Salmonella* were obtained with overall prevalence of 4.58 %. Same as that of the present study the CCP in the poultry meat shops was successfully studied by earlier workers[14]. The highest % of positive samples of *Escherichia coli* were obtained from dressing table. The lowest %ages of positive samples were obtained from personnel. Earlier workers found that chopping board (dressing table) as major source of contamination of *Escherichia coli*, Staphylococci and *Salmonella*[18,19]. It is clearly evident that dressing table is one of the major sources of *Escherichia coli*, *Staphylococcus* and *Salmonella* contamination be due to unhygienic condition of the wooden dressing tables in retail poultry meat shops. The cutting block in the retail poultry shops are contaminated from the initial slaughtered birds and this process leads to build up of contaminating bacteria.

All the 74 isolates *Escherichia coli*, 82 isolates of staphylococci and 11 isolates of *Salmonella* shown typical Grams staining reaction, biochemical reactions and sugar fermentation reactions described earlier[12]. Many workers successfully used staining characters, biochemical reactions and sugar fermentation reactions for confirmation of *Escherichia coli*, Staphylococci and *Salmonella* isolated from meat[18,21]. The results in present study are on similar lines.

Table 1: Mean bacterial counts (log CFU/cm^2 ± S.E.) with statistical analysis and prevalence of various organisms in different sources of contamination in retail poultry meat shops

Sr. No.	Sources	Total Viable Count ± SE	*Escherichia coli* ± SE	*Staphylococci* ± SE	*Salmonella*
1	Knife	5.854 ^{bcd} ± 0.05	3.75±0.09	3.60±0.15	--
2	Scalding tank	5.670 ^{e} ± 0.05	3.64±0.12	3.84±0.14	--
3	Defeatherer	5.892 ^{bcd} ± 0.06	3.46±0.06	3.60±0.07	Present (1)
4	Dressing Table	6.138 ^{a} ± 0.04	3.62±0.09	3.62±0.14	Present (4)
5	Platform	6.006 ^{ab} ± 0.05	3.98±0.44	3.49±0.10	--
6	Personnel	5.736 ^{de} ± 0.08	3.27±0.21	3.51±0.20	--
7	Water	5.824 ^{cde} ± 0.06	3.60±0.08	3.67±0.10	Present (2)
8	Poultry meat	5.900 ^{bc} ± 0.03	3.58±0.08	3.51±0.08	Present (4)
F-value		6.927005**	0.721776 ^{NS}	0.713186 ^{NS}	--
Prevalence		--	30.83	34.17	4.58

SE = Standard Error

** = Highly Significant (P<0.01)

$^{a, b, c \text{ and } d}$ = Different superscripts show significant (P<0.05) difference between the sources.

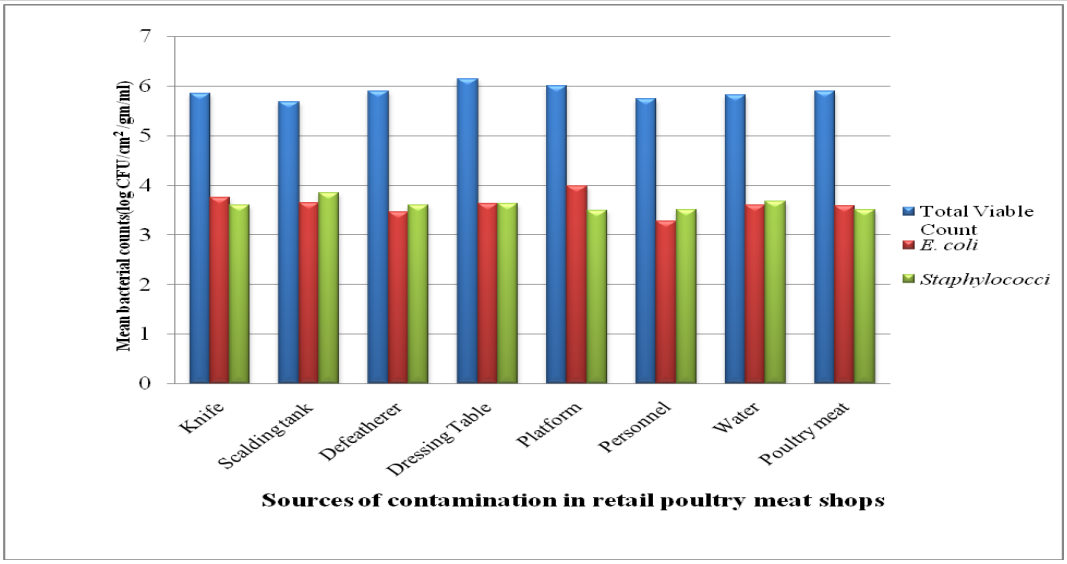

Fig. 1: Comparison of TVC counts at various environmental sources of contamination in the abattoir and traditional meat shops

CONCLUSION

All the sources i.e. knife, scalding tank, defeatherer, dressing table, platform, personnel and water are major sources of bacterial contamination during poultry slaughter process in retail poultry meat shops in Parbhani city. The total viable count of dressing table is (6.14 ±0.04) and the numbers of isolates of bacteria of public health importance obtained from dressing table are found higher than the other sources of contamination of poultry meat. Retail poultry meat shops are important sources of bacteria of public health significance i.e. *E.coli* and *Staphylococci* and *Salmonella* in poultry meat in Parbhani city.

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