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Assessment of bacterial load of poultry meat used at dining hall of Bangladesh Agricultural University campus

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Abstract: The present study was undertaken to investigate the microbiological quality of poultry meat used in different hall of Bangladesh Agricultural University campus. Sanitary condition of dining hall was assessed by using a standard questionnaire. Thirty samples were collected from Isha Khan hall, Fazlul Haque hall and Taposhi Rabeya hall. Microbial load was determined by total viable count (TVC), total coliform count (TCC) and total salmonella count (TSC). Samples were inoculated into various selective media such as plate count (PCA) agar, xylose lysine deoxycholate Agar (XLDA) and MacConkey agar (MCA). It is found that the age of maximum workers (53%) is between 25-35 years. Educational level of dining workers are in same frequency of illiterate and below class eight. Only 6.7% are SSC passed. Most of dining workers have no knowledge on hygienic practices. 43.3% partially know about hygienic practices and 3.3% have no knowledge. Sanitary condition of dining hall is not satisfactory. Only 3.3% disinfectants are used in dining hall. There is no proper washing facilities. About 63.3% washing facilities are in medium level. Only *E. coli* and *Salmonella* spp. were isolated and identified from the samples. The mean value of TVC, TCC and TSC in broiler thigh meats of Isha Khan hall, Fazlul Haque hall and Taposhi Rabeya hall are log 5.65, log 5.77, log 6.06 CFU/g, log 3.26, 3.41, 3.42 CFU/g and log 2.95, log 2.92 log 2.97 CFU/g respectively. The variation of TVC in meats of different dining hall was significant ($p < 0.05$) at 5% level of probability whereas TCC and TSC obtained from meat samples of different hall were not significant ($p < 0.05$). Presence of *Escherichia coli* and *Salmonella* spp. in meat must need particular attention as these organisms are responsible for causing harm to public health. Suggestions have been given to improve the present sanitary condition of dining hall to minimize bacterial load.

Keywords: dining hall; poultry meat; bacterial load; dining workers; hygienic practices

1. Introduction

In recent years, foodborne infections and intoxications have assumed significance as a health hazard. Epidemiological reports suggest that poultry meat is still the primary cause of human food poisoning (Mulder, 1999). Poultry meat is more popular in the consumer market because of advantages such as easy digestibility and acceptance by the majority of people (Yashoda *et al.* 2001). However, the presence of pathogenic and spoilage microorganisms in poultry meat and its by-products remains a significant concern for suppliers,

consumers and public health officials worldwide. Bacterial contamination of these foods depends on the bacterial level of the poultry carcasses used as the raw product, the hygienic practices during manipulation and on the time and temperature of storage (El-Leithy and Rashad, 1989). However, the control and inspection during production, storage and distribution are generally rare. Therefore, it is important to prevent the hazards and to provide a safe and wholesome product for human consumption (Singh *et al.*, 1984).

Undoubtedly the poultry slaughtered and dressed under Bangladesh conditions carry extremely high initial contamination loading from the point of slaughtering process to the point at which the consumers are offered the product. There occurs biomagnification at all levels of handling, poor transport and retailing conditions. Improved hygienic measures will reduce the initial contamination and the proper sanitary applications to the distribution and retailing conditions and the inherent cold chain through to the consumers could in fact meet the challenge to deliver a safe good quality product. To minimize the surface bacterial load in broiler meat it should be maintained strict hygienic measure, maintain proper legislation with government and social awareness. In many countries regulations have been enforced which require frequent cleansing and disinfecting of livestock vehicles. Many producers now expect Veterinarians to be an integral part of their quality assurance programs of foods of animal origin (Hubbert *et al.*, 1996). There are many regulatory agencies responsible for ensuring food safety and quality assurance which are offered to the consumers that will be pure and healthful and of quality claimed such agencies belonging to International forum include the FAO, WHO, UNICEF and CAC respectively. Therefore, the present study was designed with a view to determine the extent of bacterial load of broiler thigh meat obtained from some selected dining hall of Bangladesh Agricultural University, Mymensingh and to assess the sanitary condition of different dining hall.

2. Materials and Methods

2.1. Collection and transportation of samples

All samples were obtained from Isha Khan hall, Fazlul Haque hall and Sultana Taposhi Rabeya hall of BAU campus during the period from July to October 2014. The thigh region muscle were collected aseptically in sterile containers and brought to the laboratory within 30 minutes to determine the total viable bacterial count and occurrences of different microflora gaining access to meat. During transportation the sterile containers were kept cool in iceboxes containing fragments of ice.

2.2. Preparation of sample for bacteriological studies

Each of the raw meat samples was macerated in a mechanical blender using a sterile diluent as per recommendation of International Organization for Standardization (ISO, 1995). 10 (Ten) grams of the thigh meat sample was taken aseptically with a sterile forceps and transferred into sterile containers containing 90 ml of 0.1% peptone water. A homogenized suspension was made in a sterile blender. Thus 1:10 dilution of the samples was obtained. Later on using whirly mixture machine different serial dilutions ranging from 10^{-2} to 10^{-6} were prepared according to the standard method (ISO).

2.3. Enumeration of total viable count (TVC)

For the determination of total bacterial count, 0.1 ml of each ten-fold dilution was transferred and spread on duplicate plate count agar (PCA) using a fresh pipette for each dilution. The diluted samples were spread as quickly as possible on the surface of the plate with a sterile glass spreader. One sterile spreader was used for each plate. The plates were then kept in an incubator at 37°C for 24-48 hours (Figure 2). Following incubation, plates exhibiting 30-300 colonies were counted. The average number of colonies in a particular dilution was multiplied by the dilution factor to obtain the total viable count. The total viable count was calculated according to ISO (1995). The results of the total bacterial count were expressed as the number of organism or colony forming units per gram (CFU/g) of meat sample.

2.4. Enumeration of total coliform count (TCC)

For the determination of total coliform count (TCC), 0.1 ml of each ten-fold dilution was transferred and spread on MacConkey agar (MCA) using a sterile pipette for each dilution. The diluted samples were spread as quickly as possible on the surface of the plate with a sterile glass spreader. One sterile spreader was used for each plate. The plates were then kept in an incubator at 37°C for 24-48 hours presented in Figure 3. The results of the total bacterial count were expressed as the number of organism or colony forming units per gram (CFU/g) of meat sample.

2.5. Enumeration of total salmonella count (TSC)

For the determination of total salmonella count the procedures of sampling, dilution and streaking were similar to those followed in total viable bacterial count (Figure 3). Only in case of salmonella count, Xylose lysine deoxycholate agar (XLDA) was used. The calculation for TSC was similar to that of total viable count.

2.6. Cultural and biochemical examination of samples

The cultural examination of chicken thigh meat samples for bacteriological analysis was done according to the standard method (ICMSF, 1985). The examination followed detail study of colony characteristics including the morphological and biochemical properties.

In order to find out different types of microorganisms in chicken thigh meat samples, different kinds of bacterial colonies were isolated in pure culture from the plate count agar (PCA), Mac Conkey agar (MCA), blood agar (BA) and xylose lysine deoxycholate agar (XLDA) and subsequently identified according to the methods described by Krieg *et al.*, 1994. The isolated organisms with supporting growth characteristics on various media were subjected to different biochemical tests, such as sugar fermentation test for acid or acid and gas, indole production test, catalase test, coagulase test, methyl-red and Voges-Proskauer (VP) test etc. In all cases standard methods as described by Cowan (1985) were followed for conducting these tests.

2.7. Sanitary assessment of dining hall

Sanitary condition of dining hall was assessed by using a standard questionnaire (questionnaire not shown).

2.8. Statistical analysis of experimental data

The data on total viable count, total coliform count and total salmonella count obtained from the bacteriological examination of meat samples of the poultry carcass collected from different dining hall of Bangladesh Agricultural University were analyzed in completely randomised design using computer package subjected to Analysis of Variance using SPSS Software (Version 16, 2007). The differences between means were evaluated by Duncan's Multiple Range Test (Gomez and Gomez, 1984). Correlation between TVC, TCC and TSC were also evaluated.

3. Results and Discussion

Table 1 represents a consumer perception in which the consumers (students) were asked on different hygiene and safety measures that affects the quality and safe meat production. It is shown that most of the consumer like drum stick. It is found that 26.7% like drum stick, 20% breast meat, 20% like thigh, 13.3% like wing and 20% like gilet. It is found that the age of maximum workers (53%) is between 25-35 years. Educational level of dining workers are in same frequency of illiterate and below class eight. Only 6.7% are SSC passed. Most of dining workers have no knowledge on hygienic practices. 43.3% partially know about hygienic practices and 3.3% have no knowledge. Sanitary condition of dining hall is not satisfactory. Only 3.3% disinfectants are used in dining hall. There is no proper washing facilities. 63.3% washing facilities are in medium level.

The variation of TVC in meats of different dining hall was significant ($P < 0.05$) at 5% level of probability as shown in Table 2. The result of total viable count in three different dining hall were differed significantly. The findings are agreement with Rahman and Rahman (1998) and Javadi and Safarmashaei (2011) respectively. The values per gram of meat sample were $\log 5.65$, $\log 5.77$ and $\log 6.06$ respectively. These results signify the fact that the external and exposed surfaces of broiler carcass can become easily contaminated after skinning. Similar observations were also noted by Abu-Ruwaida *et al.* (1994) found the microbial load in broiler meat samples of thigh muscle to be 6.28×10^5 CFU/ gm and 6.6×10^5 CFU/ gm respectively. In another investigation by Anwar *et al.* (2004) reported the TVC per gram of market meat samples of thigh muscle was recorded 5.47×10^6 CFU/ gm and 6.65×10^5 CFU/ gm in thigh muscle respectively. Although, Datta *et al.* (2012) and Chaiba *et al.* (2007) obtained microbial load with a total count of 6.5×10^5 to 6.1×10^6 per gm, but Adu-Gyamfi *et al.* (2009) and Bhandari *et al.* (2013) observed higher count of 6.9×10^6 CFU/ gm, 11.1×10^6 CFU/ gm of meat. At the mean time Al-Mohizea (1994) observed the initial total viable count (\log_{10} CFU/cm²) which ranged from 3.8 to 5.5 with a mean of 4.67. However, Ueno *et al.* (1995) reported the TVC value of marketed broiler meat is 5.06×10^6 / gm of meat it is closely similar to this experiment.

Table 1. Sanitary assessment of dining hall.

Variables	Number of participant	Frequency (%)
Consumer perception		
Drumstick	30	8(26.7)
Breast meat		6 (20)
Thigh		6(20)
Wing		4(13.3)
Giblet		6(20)
Assessment of workers in dining hall		
Age (years)	30	
10-15		0
15-25		5(16.7)
25-35		16(53)
Above 35		9(30)
Education		
Illiterate		10(33.3)
Below class five		8(26.7)
Below class eight		10(33.3)
SSC		2(6.7)
Knowledge on hygienic practice		
Yes	1(3.3)	
No	16(53.3)	
Partially know	13(43.3)	
Sanitation of dining hall		
Disinfectant used	30	
Yes		1(3.3)
No		10(33.4)
Frequently not used		19(63.3)
Washing facilities		
Proper		0
Medium		19(63.3)
Not proper		11(36.7)

Table 2. Determination of mean and standard deviation for microbiological quality of chicken thighs at different hall of BAU campus, Mymensingh.

Retail Market	Total viable count (Mean \pm SD)	Total coliform count (Mean \pm SD)	Total salmonella count (Mean \pm SD)
Isha Khan Hall	5.65 \pm 0.46 ^b	3.26 \pm 0.17 ^a	2.95 \pm 0.28 ^a
Fazlul Haque Hall	5.77 \pm 0.39 ^b	3.41 \pm 0.14 ^a	2.92 \pm 0.31 ^a
Taposhi Rabeya Hall	6.06 \pm 0.34 ^a	3.42 \pm 0.18 ^a	2.97 \pm 0.37 ^a
Least Significant Difference	0.142	0.151	0.298
Level of sig.	**	Not significant	Not significant

In a column figures with same letter do not differ significantly ($p>0.05$) whereas figures with dissimilar letter differ significantly (as per DMRT).

All counts are expressed in logarithms and CFU/g of meat.

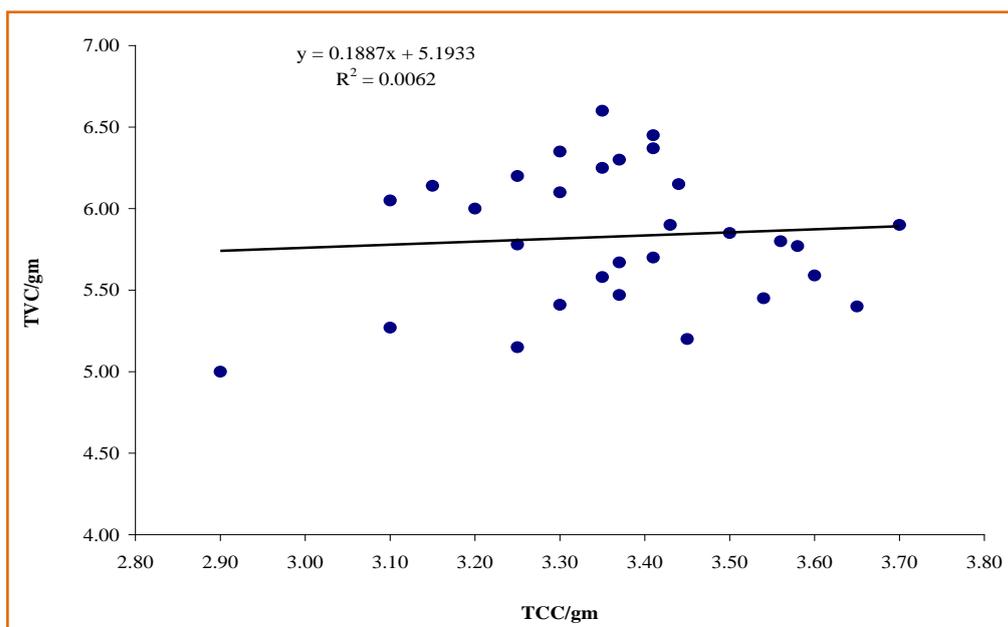


Figure 1. Correlation between total viable count (TVC) and total coliform count (TCC) in CFU/g meat of three dining hall.

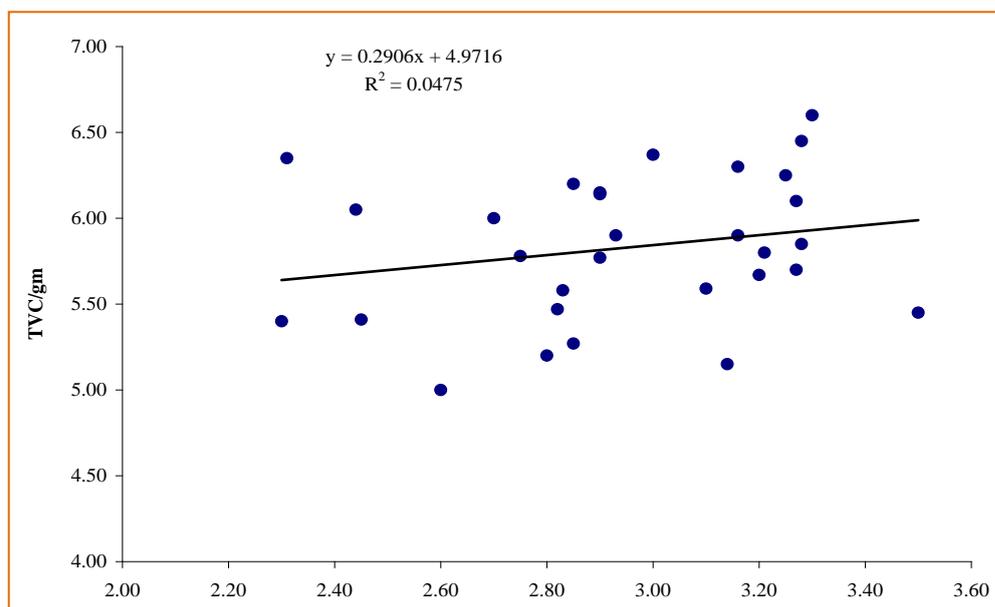


Figure 2. Relationship between total viable count (TVC) and total salmonella count (TSC) in CFU/g meat of three dining hall.

In present study the values of TVC of different broiler carcass exceeded the prescribed maximum microbial limit. ICMSF (1985) recommended that the general viable count of fresh meat tissue at 35°C should be less than 10^6 per gram. Chaiba *et al.* (2007) and Cohen *et al.* (2006) reported the presence of 10^3 to 10^6 aerobic mesophiles per cm^2 on the carcass surfaces after the completion of slaughter, dressing and washing of broiler carcass, although the count varying with site species.

The mean value of TCC per gram in meats of Isha Khan hall, Fazlul Haque hall and Taposhi Rabeya hall were log 3.26, log 3.41 and log 3.42 CFU/gm respectively and these are presented in Table 2. These findings have

proximal relationship with the findings of and Akbar *et al.* (2014), Javadi and Safarmashaei (2011), Sengupta *et al.* (2011) and Anwar *et al.* (2004). On the other hand Bhandari *et al.* (2013) found higher TCC value from fresh broiler meat and was log 6.7 to log 8.4 CFU/gm. Adu-Gyamfi *et al.* (2009), Jacob *et al.* (2008) and Chaiba *et al.* (2007) however examined TCC of internal and external surface of broiler carcass and found log 3.80 and log 4.64 CFU/ gm respectively.

The variation of TCC in meats of different dining hall is not significant ($P>0.05$) as shown in Table 2. Nevertheless no significant variation was demonstrated between the interaction of the three hall. The interpretation of total coliform count in three different dining hall were not differ significantly ($P>0.05$). This findings were agreement with the observations of Datta *et al.* (2012) and Abu-Ruwaida *et al.* (1994) respectively. The isolation of coliform bacteria indicates fecal contamination. Presence of coliforms in the present study might be due to poor quality of water used for washing of meats, vegetables and utensils, inadequate storage of these at ambient temperatures in unhygienic places, maintenance of premises and personal hygiene of dining hall workers. In this study microbiological methods were used to identify bacteria isolated from poultry meat samples. The results of cultural characteristics, Gram's staining, biochemical tests used to identify *E. coli* and *Salmonella* spp. were similar to the findings of Ahmad *et al.* (2013), Svobodova *et al.* (2012), Jacob *et al.* (2008).

The mean value of TSC per gram in meats of Isha Khan hall, Fazlul Haque hall and Taposhi Rabeya hall were log 2.95, log 2.92 and log 2.97 CFU/gm respectively as shown in Table 2. A finding was reported by Adu-Gyamfi *et al.* (2009), Rusul *et al.* (1996) and Baumgartner *et al.* (1992), the authors found TSC values log 3.37, log 3.13, log 3.2 CFU/ gm respectively from the meat of town market. The principal source of Salmonella contaminating broiler carcass includes hands of dining workers, cloths, wiping cloths, tools of workers, knives, skin, eviscerating rack etc. The organisms have been isolated from 50% of the apparently normal healthy individuals. In Fazlul Haque hall the value of TSC in meat was lower than Isha Khan hall but it was highest in Taposhi Rabeya hall (Table 4). The variation of TSC in meats of different hall was not significant ($P>0.05$) as presented in Table 4. The interpretation of Total Salmonella Count in three different hall were not differed significantly ($P>0.05$). No positive correlation and significant variation of TSC was found in three different hall and in differed broiler carcass. This signifies the fact that all these meats are more or less handled in the same manner. The findings are also closely related to the findings of several other researchers (Mead *et al.* 1994, Boonmar *et al.* 1998 and Singh *et al.* 1997 respectively). The variation of TVC in meats of three dining hall was significant ($P<0.05$) at 5% level of probability as shown in Table 2. The result estimated in Figure 1 showed weakly correlated between the total viable count (TVC) and total coliform count (TCC). In this study, total viable counts did not significantly correlated with total coliform count in meats of Fazlul Haque hall and Taposhi Rabeya hall ($P>.05$). The results contradicted with the report of Bhandari *et al.* (2013). The result in Figure 2 reveals that the regression was positively correlated with total viable count (TVC) and total salmonella count (TSC) in meats of different dining hall ($P<0.05$).

4. Conclusions

The present study demonstrated the degree of the microbial contamination during processing of broilers carcasses. The results also indicated that the viable count for microorganisms causing public hazards is appropriate for analysis. Therefore, application of hygienic measurements appears to be important to reduce the contamination of bacteria in dining hall during and after processing of meat.

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Conflict of interest

None to declare.

References

Abu-Ruwaida AS, WN Sawaya, BH Bashti, M Murad and HA Al-Othman, 1994. Microbiological quality of broilers during processing in a modern commercial slaughterhouse in Kuwait. *J. Food Prot.*, 57:887-892.

- Adu-Gyamfi A, J Nketsiah-Tabiri and R Boatun, 2009. Determination of D10 values of single and mixed cultures of bacteria after gamma irradiation. *J. Appl. Sci. Technol.*, 14:13- 18.
- Ahmad MUD, A Sarwar, MI Najeeb, M Nawaz, AA Anjum, MA Ali and NM Anisur, 2013. Assessment of microbial load of raw meat at abattoir and retail. *J. Anim. Plant Sci.*, 23:745-748.
- Akbar A and AK Anal, 2014. Zinc oxide nanoparticles loaded active packaging a challenge study against *Salmonella typhimurium* and *Staphylococcus aureus* in ready-to-eat poultry meat. *Food Control*, 38:88-95.
- Al-Mohizea TS, AS Mashhadi, A Fawwal and A Al-Shalhat, 1994. Microbiological and shelf life assessment of chilled eviscerated whole chicken broilers in Saudi Arabia. *Br. Poult. Sci.*, 35:519-526.
- Anower AKMM, MM Rahman, MA Ehsan, MA Islam, MR Islam, GC Shil and MS Rahman, 2004. Bacteriological profile of dressed broilers and its public health implications. *J. Vet. Med.*, 2:69-73.
- Baumgartner A, P Heimann, H Schmid, M Liniger and A Simmen, 1992. Salmonella contamination of poultry carcasses and human salmonellosis. *Arch. Lebensmittelhyg.*, 43:123-124.
- Bhandari N, DB Nepali and S Paudyal, 2013. Assessment of bacterial load in broiler chicken meat from the retail meat shops in Chitwan, Nepal. *Int. J. Infect. Microbiol.*, 2:99-104.
- Boonmar S, A Bangtrakulnonth, S Pornrunangwong, N Marnrim, K Kaneko and M Ogawa, 1998. Salmonella in broiler chickens in Thailand with special reference to contamination of retail meat with salmonella enteritis. *J. Vet. Med. Sci.*, 60:1233-1236.
- Chaiba A, FF Rhazi, A Chahlooui, BR Soulaymani and M Zerhouni, 2007. Microbiological quality of poultry meat on the Mekne's Market (Morocco). *Internet Journal of Food Safety*, 9:67-71.
- Cohen N and H Karib, 2006. Risque hygie'nique lie' a' la pre'sence des *Escherichia coli* dans les viandes et les produits carne's: Un re'el proble'me de sante' publique? *Technol. Lab.*, 1:4-9.
- Cowan ST, 1985. Cowan and Steel's Manual for Identification of Bacteria (2nd edn.). Cambridge University Press. Cambridge, London.
- Datta S, A Akter, IG Shah, K Fatema, TH Islam, A Bandyopadhyay, ZUM Khan and D Biswas, 2012. Microbiological quality assessment of raw meat and meat products and antibiotic susceptibility of isolated *Staphylococcus aureus*. *Agric. Food Anal. Bacteriol.*, 2:187-194.
- El-Leithy MA and FM Rashad, 1989. Bacteriological studies on ground meat and its products. *Arch. Lebensmittelhyg.*, 40:58-61.
- Gomez KA and AA Gomez, 1984. Statistical procedures for agricultural research. John Wiley and sons, Inc. London, UK (2nd edtn)
- Hubbert WT, HV Hagstad, MH Hinton and KL Hughes, 1996. Food safety and quality assurance: foods of animals origin. 2nd edition, Iowa State University Press, Ames, Iowa.
- ICMSF, 1985. Microorganism in foods; samples for microbiological analysis: principles and specific applications. Recommendation of the International Commission on Microbiological Specification for Foods. Association of Microbiological Societies, Toronto, University of Toronto Press.
- ISO, 1995. Recommendation of the meeting of the subcommittee, International Organization for Standardization, on meat and meat products. ISO/TC-36/Sc-6. The Netherlands. 10-18.
- Jacob RR, C Sethulekshmi, E Nanu and B Sunil, 2008. Evaluation of bacteriological quality of processed chicken. *J. Vet. Anim. Sci.*, 39:23-25.
- Javadi A and S Safarmashaei, 2011. Study of enterobacteriaceae contamination level in premises of poultry slaughterhouse with HACCP system. *J. Anim. Vet. Adv.*, 10:2163-2166.
- Krieg NR, JG Holt, PHA Sneath, JT Staley and ST Williams, 1994. Bergey's Manual of Determinative Bacteriology, Williams & Wilkins, Baltimore, Md, USA, 9th edition.
- Mead GC, WR Hudson and MH Hinton, 1994. Use of a marker organism in poultry processing to identify sites of cross contamination and evaluate possible control measures. *Br. Poult. Sci.*, 35:345-354.
- Mulder RW, 1999. Hygiene during transport, slaughter and processing. In: Poultry Meat Science. Poultry Science Symposium Series (Richardson and Mead eds). CABI Publishing 25:277-285.
- Rahman MM and A Rahman, 1998. Cattle and poultry development activities. In: Kromobikash-O-Karjakrom, DLS, Bangladesh. 1st ed :31-35.
- Rusul G, J Khair, S Radu, CT Cheah and RM Yassin, 1996. Prevalence of Salmonella in broilers at retail outlets, processing plants and farms in Malaysia. *Int. J. Food Microbiol.*, 33:183-194.
- Sengupta R, R Das, S Ganguly and SK Mukhopadhyay, 2011. Survey on microbial quality of chicken meat in Kolkata, India. *International Journal of Research in Pure and Applied Microbiology*, 1:32-33.

- Singh N, MF Slanik and XLiY Wang, 1997. Image analysis as a rapid pathogen detection method for use in poultry industry. *J. Rapid Methods Autom. Microbiol.*, 5:205-214.
- Singh RD, LN Mandal and JN Pandey, 1984. Isolation of aerobic microorganisms from poultry meat at central poultry farm and retail shop. Patna. *Poult. Guide*, 21:71-74.
- Svobodova I, G Borilova, R Hulankova and I Steinhauerova, 2012. Microbiological quality of broiler carcasses during slaughter processing. Acta Vet. Brno, 81:37-42.*
- Ueno H, T Usuku, S Matsuba, K Kawai, T Ohta, T Tusaka and C Morita, 1995. A bacterial survey of raw chicken meat at retail shops in Ebetsu City from 1987 to 1992. *Journal of the Japan Veterinary Medical Association*, 48:281-284.
- Yashoda KP, NM Sachindra, PZ Sakhare and DN Rao, 2001. Microbiological quality of broiler chicken carcasses processed hygienically in a small scale poultry processing unit. *J. Food Quality*, 24:249-259.