ASESSMENT OF THE HYGIENIC CHARACTERISTICS OF THE BEEF SLAUGHTER AND DRESSING PROCESS AND PREVALENCE OF *E. Coli* O157:H7 IN A SMALL SLAUGHTERHOUSE IN ZULIA STATE, VENEZUELA

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Background

In beef carcass dressing procedures the major hazard is the contamination with fecal organisms. It is widely recognized that assessment of the hygienic risk in a carcass dressing process should involve enumeration of an organism indicative of fecal contamination, such as *E. coli*, at various points of the process. Research has demonstrated that cross contamination of carcasses during dressing operations carry some level of bacteria of potential public health importance (Ingram and Roberts, 1976; Roberts, 1980; Chabelois et al.,1991; Doyle, 1991; Nortjé *et al.*, 1989; Nortjé *et al.*, 1990; Biss and Hathaway, 1995). Therefore, the goal of modern slaughter and dressing system is to reduce such contamination to the lowest practicable level. In Venezuela exist more than 280 small municipal abattoirs under surveillance by the Health Ministry. However no study has been undertaken to monitor bacterial contamination of beef carcasses during dressing operations nor the prevalence of emerging bacteria such as *E. coli* O157: H7 in these establishments.

Objectives

To assess the hygienic characteristics of the beef slaughtering and dressing process in a small slaughterhouse by determining *E. Coli*, aerobic bacteria and coliform counts, and prevalence of *E. Coli* O157: H7 at four points in the process and three sites of the carcasses.

Methods

Microbiological contamination of 36 beef carcasses in a small municipal slaughterhouse of the Zulia State, Venezuela, was evaluated, at four points of the standard dressing process: after dehiding, after evisceration, after carcass splitting and after washing. Samplings were carried out every 15 days, three times per week, during three consecutive months. Collection of microorganisms on each carcass was performed every other day at three different anatomical regions: anal, rump and cranial brisket, by means of a sterile cotton swab of an undelimited area of approximately 100 cm^2 (Gill *et al.*, 1996). Each swab was homogenized by stirring into a flask containing 100 ml of 0,1 % (wt/vol) pH 7.0 peptone water. A 1-ml portion of each homogenate was used to prepare 10-fold dilutions to 10^{-3} in 0.1% (wt/vol) peptone water. Portions of 0.1 ml of the homogenate and each dilution were spread on triplicate plates of plate count agar (Difco) and incubated for 48 h at 30 °C, to determine the levels of aerobic bacteria (TPC) (APHA, 1992). The most probable number (MPN) method was used to determine the level of coliforms (APHA, 1992; AOAC, 1995). *Escherichia coli spp* and of *E. coli* O157: H7 were determined. For the detection of E. *coli* O157: H7, the strains were taken from a commercial E.C broth and spreaded on plates containing MacConckey agar, which were incubated at 37 °C for 24 h. Lactose positive colonies were transferred onto a commercial nutritive agar and then they were made to react with a commercial antiserum (O157:H7) for the detection of O157: H7 serologic group. Positive colonies produced an agglutination reaction (Martínez, 1997). A completely randomized design with three replications was used in a 3^2 x 4 x 6 factorial arrangement. All bacterial counts (CFU/cm^2) were transformed to log values. The log-transformed data were subjected to the analysis of variance (ANOVA) using de general linear model (GLM) procedure of Statistical Analysis System (SAS) computer program, version 6.12 for Windows. Tukey's Studentized Range was used for mean comparison of the principal effects. When the interaction between two or more factors was found significant, the Least Squares method (LSMEAN) method was used for mean separation.

Results and discussion

Regarding to sampling day, higher TPC were obtained on Wednesday ($p < 0.001$) surpassing 1000 CFU/cm^2. This represented an increment of 57 and 37 % as compared to Monday and Friday, respectively. For total (TCC) and fecal coliforms counts (FCC), the highest levels were recorded on Wednesday and Friday, showing an increment of 65% and 40%, respectively, as compared to Monday. These results are different from to those obtained by Charlebois *et al.*(1991) and Dickson and Anderson, (1992) who did not detect significant differences among sampling days in FCC. Even though the slaughtering room and the equipment were washed at the end of the process it was observed that the cleaning operations were thoroughly performed only on Fridays. It can be presumed that the increase in count level observed on Wednesdays and Fridays was a result of the accumulation of bacterial population due to bad manufacturing practices.

The variance analysis for sampling week revealed that the highest levels ($p<0,001$) TPC, were observed during the first week of sampling, at mean >3000 CFU/ cm^2. Non-significant differences ($p>0.05$) were observed for subsequent weeks. Coliform counts (CC) exhibited the same trend. The differences in these results were due to the hand washing, the use of glove and clean clothe of personnel during the dressing process, starting after the first week up to the end of sampling of this investigation.. According to Nortjé *et al.*(1990), there could be an improvement in the degree of contamination if the hands and tool of operators were thoroughly cleaned.

Constant levels of TPC and CC were observed in all operational steps considered, except after washing, where an increase of 57%, 53 % and 62 % of TPC, TCC and FCC, was observed, respectively. These results were different from the observations of other researchers (Cacciarelli et al., 1983; Dickson and Anderson, 1992), in this case, carcass washing was quite ineffective for removal of surface bacteria. It was believed that the water used for washing operations had a poor microbiological quality.

Degree of contamination at the three carcass sites varied. Higher levels of TPC and CC were observed on the anal region ($p<0.001$). Population of aerobic bacteria and coliforms had the same trend on this site: 2X and >7X as compared to cranial brisket and rump, respectively.

Overall evaluation of the data revealed highly significant differences for the sampling week x sampling day interaction ($p<0.001$). There was a general trend for higher bacterial counts every sampling day during the first week, with the highest count occurring on Fridays. Despite of these results, it can be said that a considerable variability in count levels was observed.

Analysis of variance detected differences for sampling week x operational step interaction on TCC and FCC, showing high variability through the whole sampling phase, indicating bad manufacturing practices.

A considerable variability in the level of microbiological contaminant was observed when sampling week x anatomical region interaction became significant However all bacterial counts were higher during the first week at the anatomical sites with the highest level ($p<0.001$) on the anal region. This week-to-week trend of bacterial counts has also been reported by Charlebois. *et. al*, (1991) stressing the requirement of adequate controls at the slaughterhouses to minimize carcass contamination during dressing operation.

For operational step x anatomical region interaction the results showed a significant increment of bacterial counts after carcass washing in all carcass sites, however, the highest counts were found on the anal region.

E. coli prevalence.

Presence of *E coli* was observed, however, counts were significantly inferior to those found for TPC an CC. A frecuency analysis performed on the overall data, showed that only 26.7 % out of 1296 samples, gave a positive test for *E coli* . The serological analysis did not detect the presence of *E. coli* O157:H7.

Conclusions

Results from the present study reveled that hygienic condition of the beef carcass is of a multi- factorial complexity. The microbial status of the final product depends on the conditions surrounding operational steps, and management of personnel involved in the dressing process.

The high variability of microbial counts after the washing step, regardless of sampling conditions, indicated an inefficient sanitation program or an inadequate technological program in operation.

Pertinent Literature

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