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IMPLEMENTATION AND MICROBIOLOGICAL VERIFICATION OF HACCP SYSTEM IN SLAUGHTERHOUSES FOR SMALL RUMINANTS IN BULGARIA

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Abstract

Regulation (EC) 852/2004 covers general hygiene procedures for food at all stages of the production process, from primary production to sale to the EU consumer (so-called "from-farm-to-fork approach"). The requirements of Commission Regulation (EC) 2073:2005 laying down in chapter 2 - "Process hygiene criteria" validated and verified practical application of HACCP system.

In two slaughter facilities were taken swab samples from 48 lamb carcasses during all four seasons from May 2008 to April 2009. The swab samples from 5 different areas on the carcass (outer surface of the thigh; outer surface of the chest bone; the middle of the neck; the middle part of the chest's outer surface; the internal most distal part of the chest cavity) were taken with sterile tampons Peptone water (buffered) Merckotube® (Merck). The determination of total viable counts (TVC) was done in accordance with EN ISO 4833. The number of representatives of *Enterobacteriaceae* family in cfu/mL was established in accordance with ISO 21528-2 and *Salmonella* presence according EN/ISO 6579. Data were statistically processed.

Faecal contamination was pointed as the most important critical control point (CCP). TVC was higher during the summer and autumn, especially in samples from the high-capacity slaughterhouse (> 6 $\log^{10}$ cfu/cm^2). During the colder period of the year there were lower average TVC values (4.09 and 4.32 $\log^{10}$ cfu/cm^2) in the smaller and larger slaughterhouse, respectively. The values of the *Enterobacteriaceae* family microbial counts in the smaller slaughterhouse varied between 1.30 in the spring and 3.18 log10 cfu/cm^2 in the winter. At the high-capacity slaughterhouse, the variation was greater - from 1.27 in the winter to 6.05 in the autumn. *Salmonella* spp. presence was found in samples in one single lamb carcass.

The implementation and functioning of HACCP system is not just a formal act but an active system facilitating the production of quality and safe meat from small ruminants. The current results indicate that the self-control system and HACCP system in the examined facilities does not guarantee completely the fulfilment of the hygiene requirements laying down in Regulation (EC) 852:2004 and Regulation (EC) 2073:2005.

***Key words**: HACCP, Lamb, Hygiene criteria, Salmonella.*

1. Introduction

In response to the increasing risk of food-borne diseases in consumers Regulation (EC) 852/2004 in response of the increasing risks of food-borne diseases in consumables set the integrated approach, based on HACCP principles with the aim to ensure food safety from the place of primary production up to and including placing on the market or export [1]. The requirements set out in Chapter 2 "Process hygiene criteria" of Commission Regulation (EC) 2073:2005 validated and verified the practical application of HACCP system [2]. In Mexico, were surveyed one hundred and sixty enterprises with officially applied HACCP system were surveyed and the main reported benefit in 18% of the studied plants was reduction in microbial counts, while staff training was noted as a significant problem [3]. In 2001, authors from USA pointed that the requirements referred as pathogen reduction (PR) and Hazard Analysis and Critical Control Points (HACCP) included sanitary activities and microbiological testing for developing and following an individual HACCP plan [4]. A systematic review of published data related to the effect of HACCP programs on microbial contamination of carcasses in abattoirs has shown that the results of microbial testing pre- and post-HACCP implementation were reported in only 19 studies. Only Australia reported extensive national data spanning the period from 4 years prior to HACCP implementation to 4 years

post-HACCP, indicating reduction in microbial prevalence and concentration on beef carcasses in abattoirs slaughtering cattle for export [5]. Study in red meat and poultry processing enterprises in Canada for identifying the factors motivating adoption of enhanced food safety controls such as HACCP proved the importance of adopting an "incentive-based regulatory approach" [6]. It is important to note that decontamination interventions during poultry, cattle and pig slaughter should be validated and considered as part of a hazard analysis critical control points (HACCP)-based food safety system which is subject to verification and auditing, and they should never be used as a substitute for good sanitation and proper hygiene practices [7]. In 2012, EFSA organized a technical hearing on meat inspection of small ruminants and made a report, which summarised the information that had been provided by the representatives of the three stakeholder organisations participating in the technical hearing. HACCP implementation and microbiological monitoring as a tool were part of the discussion [8].

The aim of the study was to compare the implemented HACCP plans of two small ruminant slaughter facilities (with low and high production capacity) with the results of microbiological criteria set in Chapter 2 of Commission Regulation (EC) 2073:2005 [2].

2. Materials and Methods

Studies were performed in two types of slaughter facilities in Bulgaria:

A. Small animal slaughter plant with low capacity - up to 20 animals per week with electrical stunning. All slaughter processes were done by two permanent workers with long-term experience and training. The abattoir implemented HACCP system in 2007 and three critical control points (CCP) were developed (skinning, evisceration and cooling of lamb carcasses). Skinning was performed using mechanical equipment for removing the skin from the top down (top-to-bottom). Moving the carcasses on the line was made manually by the working staff.

B. Small animal abattoir with high capacity - from 100 to 500 small ruminants per week with electrical stunning. All slaughter processes were done by separate workers distributed along the chain line. The slaughter facility has applied HACCP system since 2007 when three critical control points were developed (skinning, evisceration and cooling of lamb carcasses). Skinning was mechanical using equipment for removing the skin from the upper part of the carcass down (top-to-bottom). Moving the carcasses on the line was also mechanical.

In both abattoirs dry and wet washing were applied before the carcasses were transffered in the cooling cameras. The HACCP plans in both plants were similar and the fixed CCP were completely the same. All elements of the HACCP plans were practically identical despite the facility size.

The collection of samples from the surface of the processed small animal carcasses (lambs) was performed after the dry and wet washing. Swab samples were taken from 5 different areas of the carcass (outer surface of the thigh - 1; outer surface of the chest bone - 2; the middle of the neck - 3; the middle part of the chest's outer surface - 4; the internal most distal part of the chest cavity - 5) with sterile tampons Peptone water (buffered) Merckotube® (Merck) as presented on fig.1.

Figure 1. Sampling areas

Sampling was performed in compliance with the relevant rules determined by ISO 17604:2003 [9]. Samples were obtained once per season from each facility (in June, September, December 2008, and March 2009), resulting in a total of 120 swab samples from both slaughterhouses. They were immediately transported to the laboratory in a refrigerating container at a temperature of 0 - 4 ^{0}C. The samples were processed immediately after delivery. The total viable count (TVC) of microorganisms, as well as the number of *Enterobacteriaceae* (in cfu/mL) for each swab sample were determined. The swab sample preparation was performed in accordance with the requirements of ISO 6887 [10] in order to prepare the necessary dilutions. The determination of TVC was done in accordance with BSS EN ISO 4833:2004 [11]. The number of representatives of *Enterobacteriaceae* family in cfu/mL was established according to ISO 21528-2:2004 [12]. The results were log transformed and statistically processed by ANOVA using the Microsoft Excel software.

3. Results and Discussion

Interviews were conducted in both abattoirs to characterize the size of the firms. The small animal slaughter plant with low capacity had 7 employees, and one of them was the person responsible for GMP and HAC-

CP implementation and many other duties such as accepting of animals for slaughter, documents, filling forms after cleaning and disinfection. The small animal slaughter plant with high capacity had 25 employees, and one of them was responsible for GMP and HACCP implementation, coordination of hygienic procedures such as cleaning and disinfection and checking the effect of them. The analysis of the HACCP plans of the two enterprises showed high similarity of the technological processes. CCP in both places were the same - CCP 1. Skinning and CCP 2. Evisceration. The downward-pulling machines used for skinning appeared to have a good hygienic effect. Important is to note that both CCP are associated with a risk of fecal contamination of the surface of the lamb carcasses. Dressing/decontamination treatments conducted in both abattoirs included dry toilet and water washing of the lamb carcasses.

Figure 2 presents the total viable counts of the examined samples from lamb carcasses. During the warm seasons (summer and autumn), we registered high values of TVC in both abattoirs, especially in the high capacity plant in the autumn period (up to 7 $\log_{10}$ cfu/cm^2). During the cold period of the year, the lowest values of TVC (4.09 and 4.32 $\log_{10}$ cfu/cm^2) were detected in both the low and high capacity slaughterhouses. During the autumn, winter and spring, the samples taken from the small abattoir showed statistically lower values of TVC ($P < 0.05$) as compared to those obtained from the big size abattoir.

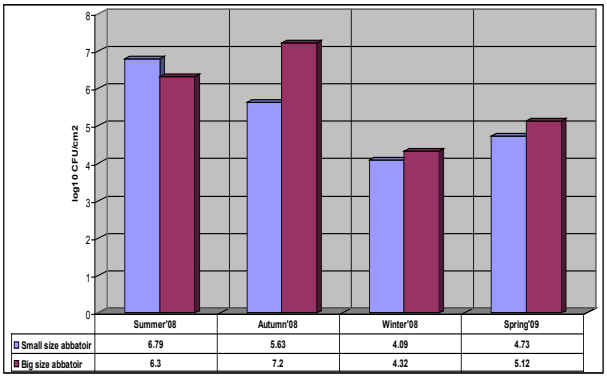

Figure 2. Total viable counts of lamb carcasses, slaughtered in small and big size abattoirs

Figure 2 shows that the *Enterobacteriaceae* counts of the lamb carcasses from the small size abattoir were highest during the autumn, followed by winter and summer and lowest - in the spring. Results from the big size abattoir were obtained only during autumn, winter and spring. The highest data values were observed during the autumn - 6.04, followed by spring - 2.58, whereas the lowest were observed in the winter period - 1.26 $\log_{10}$ cfu/cm^2. The differences between the two slaughterhouses were statistically significant ($P < 0.05$) (Figure 3).

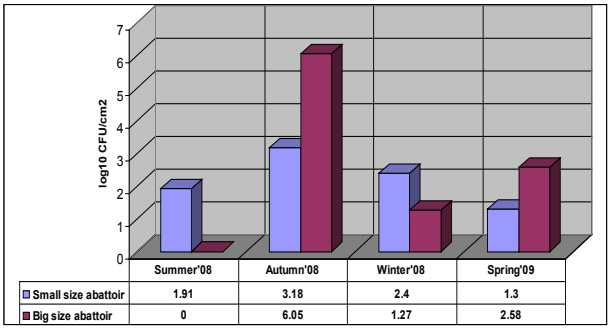

Figure 3. *Enterobacteriaceae* counts of lamb carcasses, slaughtered in small and big size abattoirs

The examinations for detection of *Salmonella* spp. showed that one of the lamb carcasses from the big size abattoir was positive for this food-borne pathogen. The isolates from different parts of the lamb carcass were serotyped as *Salmonella typhimurium*. No other positive results were detected in the examined swab samples.

The studies on the three microbiological criteria (TVC, *Enterobacteriaceae* counts and presence of *Salmonella* spp.) set in Regulation (EC) 2073:2005 [2] which were carried out in two different types of abattoirs for small ruminants were a tool for verifying of the implemented in 2007 HACCP plans. The hygienic requirements for sheep set in Regulation (EC) 2073:2005 [2] for TVC varied from 3.5 log cfu/cm^2 to 5.0 log cfu/cm^2 daily mean log, and for *Enterobacteriaceae* counts - from 1.5 log cfu/cm^2 to 2.5 log cfu/cm^2 daily mean log. These parameters have to be evaluated after dressing before chilling. Additionally, *Salmonella* spp. was tested as a hygienic criterion, also set in the same Regulation. Our results showed that the TVC of the carcasses in both slaughterhouses did not exceed the set limits only during the winter months (4.09 and 4.32 $\log_{10}$ cfu/cm^2). Similar was the case with *Enterobacteriaceae* counts. *Salmonella typhimurium* was found only in one lamb carcass.

Various authors in many developed countries have studied TVC and *Enterobacteriaceae* counts as a criteria for hygienic status of lamb carcasses. Grau [13] in Australia found average value of TVC of lamb carcasses equal to 3.18 log cfu/cm^2; Roberts *et al*. [14] in England proved that the TVC values varied from 2.54 to 3.25 log cfu/cm^2; Kelly *et al*. [15] in Ireland determined average values of 3.77 log cfu/cm^2. In the present moment, all these countries and their abattoirs have implemented the HACCP system and the results of TVC examinations have verified low levels of fecal contamination and good hygienic status. Some authors from south countries as Spain found TVC average value of 4.96 log cfu/cm^2 [16], in India - 5.13 log cfu/cm^2 [17]. Rao and Ramesh [18] in India reported no relation between the level of contamination and the season. More resent studies on hygienic status of lamb carcasses have

shown variations of TVC values from 1.66 to 3.91 log cfu/cm^2, *Enterobacteriaceae* counts from 0.02 to 1.16 log cfu/cm^2, and absence of *Salmonella* spp. in 400 samples from surface of lamb carcasses [19]. Results reported in USA (2007) showed presence of Salmonella spp. in 4.3% of lamb carcasses [20], and other authors noted Salmonella existence in 1.5% of chilled lamb carcasses [21].

4. Conclusions

- In conclusion, we can summarize that TVC of the examined lamb carcasses varied from 4.09 to 6.79 $\log_{10}$ cfu/cm^2 in the small size abattoir and from 4.32 до 7.20 $\log_{10}$ cfu/cm^2 in the big size plant.

- *Enterobacteriacea*e counts varied from 1.30 to 3.18 $\log_{10}$ cfu/cm^2 in the small abattoir and from 1.27 to 6.05 $\log_{10}$ cfu/cm^2 in the big size one. Furthermore, direct relationship between the TVC and the season was observed.

- Using microbiological results as a verification tool we can conclude that the implemented HACCP plans need to be reviewed and the hygienic procedures to be revised. The current results indicate that the self-control system and HACCP system in the examined facilities does not completely guarantee the fulfillment of the hygiene requirements laying down in Regulation (EC) 852:2004 [1] and Regulation (EC) 2073:2005 [2].

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