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# Process hygiene of pig carcasses in one large-scale slaughterhouse in the west of Serbia, during 48 months

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**Abstract.** This study was conducted to determine microbial contamination of pig carcasses during four years in one slaughterhouse. The numbers of total viable counts and *Enterobacteriaceae* and the presence/absence of *Salmonella* spp. are the process hygiene criteria for pig carcasses. We collected 240 samples from April of 2015 to April of 2019, with swabs being continually taken from the carcasses of pigs every month for 48 months in slaughterhouse in the west of Serbia. Over 48 consecutive months of testing, *Salmonella* spp. presence was detected on 1.67% of the pig carcasses, while the determined mean numbers of *Enterobacteriaceae* were  $0.18 \pm 0.37$  log CFU/cm<sup>2</sup>, and the mean total viable count of aerobic bacteria was  $1.88 \pm 0.85$  log CFU/cm<sup>2</sup>. The process hygiene criteria results for the tested pig carcasses showed that for total viable count of aerobic bacteria, 95.35% of carcasses fell into the satisfactory process hygiene group, while 4.17% belonged to the acceptable group. *Enterobacteriaceae* numbers showed 97.90% of the tested pig carcasses belonged to the satisfactory process hygiene group, and 2.10% of carcasses belonged to the acceptable group.

## 1. Introduction

Meat consumption is increasing worldwide due to rapid population growth, urbanization, changing consumer preferences and income growth. Global meat consumption increased by 58% during the past 20 years and in 2018, reached 360 million tonnes [1]. That has resulted in increased concerns and challenges, above all in the field of meat safety and hygiene. To date, the best approach to food safety is a preventive approach, by managing food production from primary production to the consumer. The main responsibility for food safety lies with the Food Business Operators (FBO), who define and implement appropriate measures for good hygienic and manufacturing practice, as well as other procedures based on Hazard Analysis and Critical Control Point (HACCP) principles, in order to achieve the food safety objectives defined in the food regulations. The presence of some microbial indicators is a consequence of direct or indirect contamination of the food with fecal material [2]. The numbers of total viable counts (TVC) and *Enterobacteriaceae* (*EC*) and the presence/absence of *Salmonella* spp. are the process hygiene criteria for pig carcasses.

During the last few decades, infections with *Salmonella* spp. have been recognized as a major hazard to humans in most developed countries, primarily through contaminated food of animal origin. The genus *Salmonella* covers more than 2400 different serotypes, and although all serotypes must be considered as potential human pathogens, only a limited number of serotypes is attributed as a cause of infection in humans and animals. Although *Salmonella* can survive for long periods in the



environment [3], it is assumed carrier animals are the major source of infection for both animals and humans.

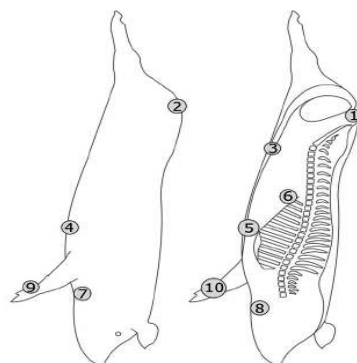
Pig carcasses contaminated with *Salmonella* cannot be recognized during the current veterinary inspection after slaughter. Good manufacturing principles are important to prevent cross-contamination of carcasses during the slaughter process [4]. Cross-contamination in the slaughterhouse is also a big problem from the aspect of meat safety [5], which is confirmed by the increased prevalence of *S. enterica* from farm to slaughterhouse. Furthermore, the contamination/infection of pigs with *Salmonella* spp. can occur at any point from the farm to the slaughterhouse, although it should be emphasized that the slaughterhouse has an important role in this process. The surfaces of the lairage and stunning box are almost always contaminated with *Salmonella*, and these surfaces can be sources of cross contamination, ultimately increasing *Salmonella* prevalences on carcasses on the slaughter line [6]. Operations at the point of slaughter can also have an effect on pig carcass contamination with *Salmonella* [7].

*EC* are very widespread in the environment, and they are also an integral part of the gastrointestinal microbiota of humans and animals. One of the most important places for contamination of pig skin with enterobacteria is the stunning box, which each pig touches. There is also a high risk of meat contamination with gastrointestinal tract contents during pig evisceration. This evisceration is the processing step that most contributes to bacterial contamination on carcass surfaces, because afterward, there is no primary treatment that could reduce the number of bacteria. The technology of pig skin removal after slaughter also carries a high risk of contaminating carcasses/meat with enterobacteria [8]. Moreover, any inadequate procedures during technological operations on the slaughter line can lead to contamination of pig carcasses [9].

The aim of this study was to follow the process hygiene of pig carcasses in one large-scale slaughterhouse during a period of four years. Monitoring hygiene in the slaughterhouse was conducted through process hygiene examinations of pig carcasses and validation of the HACCP system according to the self-control plan of this FBO.

## 2. Materials and Methods

Every slaughterhouse should have a self-control plan specifying time and frequency of sampling, which is regulated according to the: slaughter practice for each animal, design of risk-based process control assurance or harmonized monitoring programs, production volume and the epidemiological status of the area from which the animals originate. The numbers of microorganisms on carcasses were determined according to standard methods [10]. In this study, we used the non-invasive swab sampling method. The swab method is the preferred method for carcass sampling according to HACCP requirements for European Union slaughterhouses [11]. The carcass sites from which samples are taken must be described in the self-control plans, edited by the FBOs. However, since the purpose of this study was to examine those carcass sites where the probability of contamination was the greatest, the recommended standard sampling sites on pig carcasses were used in this study, as shown in Figure 1 [10].



**Figure 1.** Suitable sites for taking samples from pig carcasses [10]. Sites are: 1) Pelvic channel internal, 2) Pelvic channel external, 3) Abdominal, 4) Xiphoid external, 5) U Xiphoid internal, 6) Pillar of diaphragm, 7) Submaxillary external, 8) Submaxillary internal, 9) Forefoot external aspect, 10) Forefoot internal aspect

### 2.1. Process hygiene criteria for pigs

The microbiological criteria for production process hygiene control of pig carcasses were: TVC [12], EC count [13] and the presence/absence of *Salmonella* spp. [14]. Regulations in the EU (No. 2073/2005) [15] and in Serbia [16-18] prescribe limits for process hygiene test results for pig carcasses (Table 1).

**Table 1.** Process hygiene criteria for pigs – non-destructive sampling method [15-18]

Microorganism	Sampling plan		Limits		Analytical reference method	Stage where the criterion is applied	Action in the case of unsatisfactory results
	N	c	m	M			
Total Viable Counts			3.3 log CFU/cm <sup>2</sup>	4.3 log CFU/cm <sup>2</sup>	ISO 4833		Improvements in slaughter hygiene and review of process controls
EC			1.3 log CFU/cm <sup>2</sup>	2.3 log CFU/cm <sup>2</sup>	ISO 21528-2	Carcasses after dressing	
<i>Salmonella</i>	50	3 (5)*	Absence in the area tested per carcass		EN/ISO 6579	but before chilling	Improvements in slaughter hygiene and review of process controls, origin of animals and of the biosecurity measures in the farms of origin

\*[16]; n=number of units comprising the sample; c = number of sample units giving values between m and M.

### 2.2. Samples from slaughterhouse

A total of 240 swabs from randomly selected pig carcasses were collected in one slaughterhouse in Kolubara district, West Serbia. This study lasted for a period of four years, from April 2015 to April 2019. Five samples were collected once a month. Swabs were taken on the slaughter line after the final washout before chilling, in order to monitor compliance with the process hygiene criteria. Process

hygiene was followed and compared with Serbian regulation [16], and followed up by comparison with new Serbian regulation [17] from its date of validation, August 2018.

### 2.3. Statistical analysis

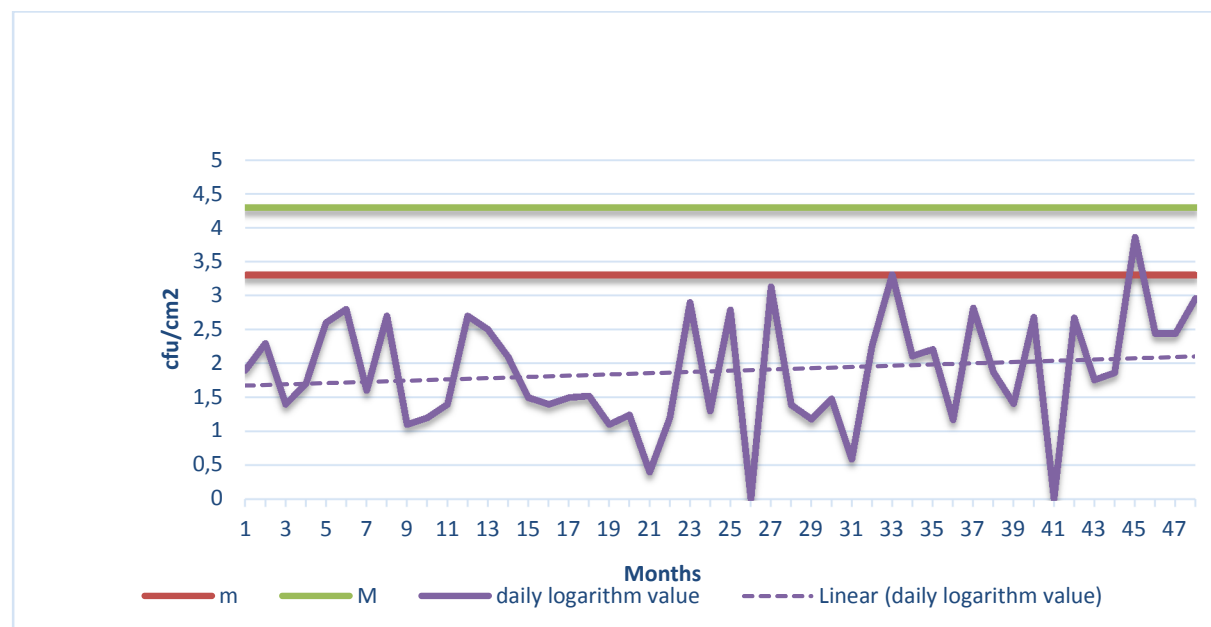
Statistical analysis of the results was conducted with Microsoft Office, Excel program 2016 and GraphPad Prism version 7.00 software. Firstly, the average logarithm value of TVC and *EC* counts for each carcass was calculated (based on previously transformed log values of these bacterial counts for each of four corresponding sites on each carcass), and then the average daily logarithm was calculated. The results were expressed as the mean  $\pm$  standard deviation. The average daily logarithm of *Salmonella* spp. was not calculated, taking into account the regulatory requirement defining only the absence or presence of *Salmonella* spp.

## 3. Results and Discussion

Levels of TVC on the pig carcasses ranged from undetected to 3.86 log CFU/cm<sup>2</sup>, while *EC* levels ranged from undetected to 1.86 log CFU/cm<sup>2</sup>. *Salmonella* spp. were detected on 1.67% of carcasses, while the mean number of *EC* on the carcasses was 0.18 $\pm$ 0.37 log CFU/cm<sup>2</sup>, and the mean TVC of aerobic bacteria was 1.88 $\pm$ 0.85 log CFU/cm<sup>2</sup>.

### 3.1. TVC numbers and trend

The results of process hygiene testing on pig carcasses in this slaughterhouse showed that for TVC, 95.38% of carcasses fell into the satisfactory process hygiene group (equal to or less than 3.3 log CFU/cm<sup>2</sup>), while 4.17% belonged to the acceptable group (3.3-4.3 log CFU/cm<sup>2</sup>) (Figure 2). Results reported previously [19] were similar to the results in our study, as 97% of the carcasses in that study fell into the satisfactory group and 3% fell into the acceptable group. The linear trend for the mean daily TVC on the pig carcasses, which increased over the 48 months, is shown in Figure 2.

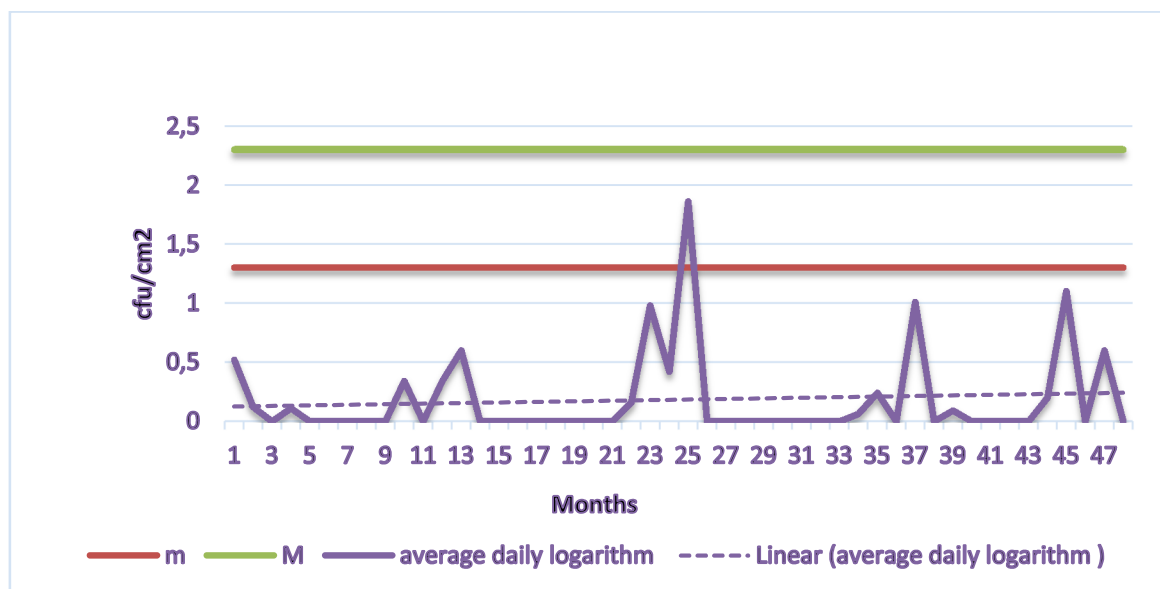


**Figure 2.** Trend analysis of total viable count for pig carcasses 2015-2019

### 3.2. Enterobacteriaceae numbers and trend

*EC* numbers on pig carcasses in the slaughterhouse were such that 97.9% of tested pig carcasses belonged to the satisfactory process hygiene group (equal to or less than 1.3 log CFU/cm<sup>2</sup>) and 2.1% of carcasses belonged to the acceptable group (1.3-2.3 log CFU/cm<sup>2</sup>) (Figure 3). Similar results were

found by Milojević *et al.* [19], who reported that 99% of tested pig carcasses belonged to the satisfactory group and 1% belonged to the acceptable group. Figure 3 shows the slightly increasing linear trend for *EC* over our 48 month study.



**Figure 3.** Trend analysis of *Enterobacteriaceae* for pig carcasses 2015-2019

### 3.3. *Salmonella* spp. presence/absence

The presence of *Salmonella* spp. was detected on 4 of the 240 pig carcasses examined. The regulatory limit for detection of *Salmonella* spp. is 3 times in 50 samples. These current results differ from the results of Mrdovic *et al.* [20], who carried out research in another district in Serbia, but detected the presence of *Salmonella* spp. only twice during a period of six years (2011-2016). Because *Salmonella* spp was detected at the slaughterhouse, the origin of animals and biosecurity measures on the farms of origin had to be checked, process controls reviewed, and slaughter hygiene improved.

## 4. Conclusion

We conclude that more than 95% of tested pig carcasses at slaughter in this FBO's premises had satisfactory process hygiene indicators, *EC* (97.9%) and TVC (95.38%).

The FBO was required to perform corrective actions because of the presence of *Salmonella* spp. on the pig carcasses at slaughter. The FBO had to improve slaughter hygiene and review measures for process control, check the origin of the animals and examine biosecurity measures in the farms of origin according to their self-control plan.

The process hygiene indicators and microbial quality of meat for consumption are closely related to public health, and so the FBO must have proper control over the production process. Linear trends of the process hygiene data for both TVC and *EC* showed increasing numbers of both bacteria indicators of process hygiene on the pig carcasses over time.

FBOs should more respectful of their requirements to fulfill good manufacturing practice (GMP) and good hygiene practice (GHP) measures and improve production hygiene in the slaughterhouse. The pre-requisite GHP and GMP programs must work effectively before HACCP is applied. HACCP is the best system currently available for maximizing the safety of meat and meat products, as well as food in general, and requires the FBO to proactively recognize, control and/or eliminate relevant hazards that could compromise product safety.

### Acknowledgment

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