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Trends in the microbial contamination of bovine, ovine and swine carcasses in three small-scale abattoirs in central Italy: A four-year monitoring

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ABSTRACT

The microbial contamination of animal carcasses with respect to the limits established by Regulation (EC) No. 2073/2005 was investigated. Bovine, ovine, and swine carcasses (n = 536 samples) from three small-scale abattoirs were sampled using abrasive sponges and tested for aerobic colony counts (ACC) and *Enterobacteriaceae* in the period 2010–2013.

Mean ACC values reached 1.96 log cfu/cm^2 on bovine carcasses and 2.27 log cfu/cm^2 on both swine and ovine carcasses; *Enterobacteriaceae* counts of 0.01, 0.20 and 0.27 log cfu/cm^2 were found for bovine, swine and ovine carcasses, respectively. Abattoir 1 showed the highest values of ACC; no differences among abattoirs were highlighted for *Enterobacteriaceae*. Compared with swine and ovine carcasses, bovine carcasses showed significantly lower means for both ACC and *Enterobacteriaceae*. The data collected indicated that the management of the three abattoirs met high quality standards, thereby proving that it is feasible to achieve good microbiological quality in abattoirs when adequate Good Hygiene Practices are applied.

1. Introduction

Quality and hygiene are two important parameters that, in food production, must be pursued in parallel. In the specific case of meat processing, the importance of proper hygiene is fundamental in order to prevent the contamination of carcasses by spoilage and/or pathogenic microorganisms, so as to obtain microbiologically optimal products. The compliance with hygiene requirements, aimed at obtaining high quality products, should be implemented along the entire production chain, from slaughtering to processing. The contamination of carcasses may occur during the conversion of living animals into meat for human consumption. In fact, as reported in several studies, meat and meat cuts can be contaminated at various stages during slaughtering by a wide range of microorganisms such as those belonging to the *Enterobacteriaceae* family (*Salmonella*, *Klebsiella*, *Shigella*, *Yersinia* and *Escherichia*) and other pathogens (*Campylobacter*, *Listeria monocytogenes*) or spoilage bacteria (*Acinetobacter*, *Brochothrix*, *Pseudomonas*, *Psychrobacter*) (Bonardi et al., 2013; De Filippis, La Storia, Villani, & Ercolini, 2013; Khen, Lynch, Carroll, McDowell, & Duffy, 2014; Petruzzelli et al., 2010, 2014, 2015; Salmela, Fredriksson-Ahomaa, Hatakka, & Nevas, 2013).

The contamination of carcasses during slaughtering is an inevitable process, since meat, which is initially sterile, can come into direct contact with the skin and digestive tract contents of the slaughtered animals; the microbial cross-contamination of carcasses is greatly influenced by the structure of the slaughtering system, the speed of the slaughtering, the operators' activities and the initial degree of animal cleanliness (Bacon et al., 2000; Blagojevic & Antic, 2014). In this regard, the Regulation (EC) No. 853/2004 (Annex III, Specific Requirements, Section I: Meat of Domestic Ungulates, Chapter IV: Slaughter Hygiene, Paragraph 4) requires food businesses operating slaughterhouses where domestic ungulates are slaughtered, to ensure the cleanliness of animals. In addition, Regulation (EC) No. 854/2004 specifies that it is the responsibility of the Veterinary Official to verify that the animals are slaughtered in clean conditions, appropriate for their use for human consumption.

The European Union food hygiene legislation is intended to protect consumers against potential health risks and to maintain a high level of consumer protection at all stages in the food chain. This must be obtained through the application of appropriate measures that include good hygiene practices (GHP) and risk control throughout the supply chain.

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In order to evaluate the hygiene of the slaughtering process, different microbial indicators (such as total mesophilic aerobes, *Aeromonas*, coliforms, *Enterobacteriaceae*, *Escherichia coli*, and fecal streptococci) have been used (Milios, Drosinos, & Zoiopoulos, 2014); the aerobic colony count (ACC) is commonly used to evaluate the hygiene of the entire meat production process, whereas *Enterobacteriaeceae* and *E. coli* are more frequently used to assess enteric contamination (Ghafir, China, Dierick, De Zutter, & Daube, 2008). Indicator microorganisms can suggest the presence of pathogens since there is a possibility that pathogens might be a positive fraction of indicators (Brown et al., 2000). In addition, indicator microorganisms are easy to detect and are relatively inexpensive to analyze, although there is no proven correlation between these indicators and the occurrence of pathogens. However, it is widely accepted that the amount of pathogens is lower than the number of microbial indicators and the trend in pathogen reduction reflects a decrease in microbial indicators (Brown et al., 2000).

Since the introduction of the so-called "Hygiene Package", including Regulation (EC) No. 852/2004 which abrogated the previous EC Directive 93/43, all food business operators have been obliged to implement permanent procedures based on HACCP principles in order to ensure the safety of food products (Osimani, Aquilanti, Babini, Tavoletti, & Clementi, 2011). The process hygiene criteria for carcasses in the European Union (EU) are laid down in Regulation (EC) No. 2073/2005, and its amendments (Regulation (EC) No. 1441/2007). Process hygiene criteria indicate the acceptable functioning of the production process and set indicative contamination values above which corrective action is required in order to maintain the hygiene of the process in compliance with European Union food law.

The techniques used for the recovery of microbial loadsspread on animal carcasses after slaughtering are pivotal for obtaining sound data to be used for validation and verification purposes (Milios et al., 2014). As regards sampling, destructive and non-destructive methods have been developed and evaluated and it is widely recognized that non-destructive methods, which imply the use of adhesive contact tapes, swabs, sponges and contact agar plates, can be used instead of destructive methods (excision of tissues). On this subject it is worth noting that, although the microbial recovery carried out using non-destructive methods may be lower compared with destructive methods, there is a proportional relationship with microbial loads recovered by excision; hence, the data obtained using non-destructive methods can be as sound as those obtained by destructive methods (Milios et al., 2014). A recent study published by Gallina et al. (2015) highlighted that, although the excision sampling method seemed to be the most efficient in terms of microbial recovery, the use of sponging has proved to be a reliable method for carcass sampling, ensuring food safety for consumers and, at the same time, causing no damage to the carcasses.

The aim of this four-year study (2010–2013) was the evaluation of the hygiene process in 3 small-scale abattoirs through the monitoring of microbial loads (aerobic colony counts and *Enterobacteriaceae*) on carcasses in order to assess the effectiveness of the HACCP system and verify the microbial contamination with respect to the levels of acceptability established by Regulation (EC) No. 2073/2005. In this context, trend analysis may show changes or patterns in the data that are a result of unwanted changes in the slaughtering process enabling the food business operator to take corrective action before the food safety issue gets out of control.

2. Materials and methods

2.1. Sampling

A total of 536 bovine, ovine, and swine carcasses were subjected to microbiological analyses. Samples were collected in 3 small-scale abattoirs (named 1, 2, and 3) located in the Pesaro and Ancona provinces (Marche Region, Italy) and managed by Public Institutions (Table 1).

All three abattoirs had adopted an HACCP system and obtained the CE mark certification; abattoir 1 processes between 20 and 40 *Unité-Gros Bétail* (UGB) per week, while abattoirs 2 and 3 usually process between 40 and 100 UGB per week. Abattoirs 1 and 2 are organized as a single open space, where different lines are located; abattoir 3 consists of separate slaughtering areas where lines for processing different species are neatly divided. In all the abattoirs the slaughtering techniques are the same according to the species to be slaughtered: a captive bolt stunning gun for bovine and an electric stunner for both ovine and swine. Before slaughtering only the swine are washed with cold water and undergo scalding after stunning treatment; no decontamination intervention is carried out on bovine and ovine carcasses, the latter being slaughtered without removing the fleeces.

Samples, as detailed in Table 1, were always collected on the same day of the week by the same Veterinarian over a four-year period (2010–2013) to perform a regular monitoring in the abattoirs in agreement with Regulation (EC) No. 2073/2005 and the ISO 17604 standard

Table 1
Samples of bovine, ovine and swine carcasses (n = 536) collected along seasons in the period 2010–2013 in the three small-scale abattoirs.

Year	Season	Abattoir 1			Abattoir 2			Abattoir 3			Total		
		Bovine #	Ovine #	Swine #	Bovine #	Ovine #	Swine #	Bovine #	Ovine #	Swine #	Bovine #	Ovine #	Swine #
	S	2	–	2	5	–	10	5	–	8	12	–	20
	A	–	5	6	–	–	5	–	5	–	–	10	11
	W	–	–	5	5	5	10	5	–	10	10	5	25
	Sp	–	5	–	5	–	5	–	10	10	5	15	15
2010		2	10	13	15	5	30	10	15	28	27	30	71
	S	3	–	–	–	5	–	5	–	5	8	5	5
	A	–	5	5	5	5	–	5	5	5	10	15	10
	W	3	–	4	–	5	5	–	5	5	3	10	14
	Sp	–	5	5	5	5	4	5	5	9	10	15	18
2011		6	10	14	10	20	9	15	15	24	31	45	47
	S	5	–	5	5	–	5	5	–	5	15	–	15
	A	–	5	10	5	–	5	5	5	10	10	10	25
	W	5	–	5	5	5	5	–	5	10	10	10	20
	Sp	–	5	5	–	5	10	5	5	5	5	15	20
2012		10	10	25	15	10	25	15	15	30	40	35	80
	S	5	5	5	–	5	10	–	5	5	5	15	20
	A	–	5	5	5	5	–	5	–	5	10	10	10
	W	5	–	5	5	–	10	–	5	5	10	5	20
	Sp	–	–	.	5	10	–	5	–	5	10	10	5
2013		10	10	15	15	20	20	10	10	20	35	40	55
Total		28	40	67	55	55	84	50	55	102	133	150	253

S summer, A autumn, W winter, Sp spring.