SHORT COMMUNICATION



Microbiological investigation on the effect of rinsing of intentionally soiled roe deer carcasses

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Abstract

Reduction of the microbial load of soiled carcasses is essential in the production of game meat. Rinsing with water is a common practice in handling game carcasses to remove any visible contamination. In this study, microbiological investigations were performed on carcasses of roe deer (*Capreolus capreolus*), inoculated with a mixture of gastrointestinal content and then either rinsed (n=3) or unrinsed (n=3). Water rinsing may have short-term effects on bacterial contamination related to soiling. However, introducing water into the body cavity may promote bacterial growth during cold storage.

Keywords Game meat · Microbial contamination · Gastrointestinal content · Water washing · Soiled carcasses

1 Introduction

In contrast to livestock meat production, several uncontrollable factors influence the microbiological quality of game meat products at the primary production stage. Disregarding the level of influence of natural conditions, factors such as damage to the abdominal area or an inadequate evisceration technique affect the microbial load (ML) of the muscle surfaces (Branciari et al. 2020; Mirceta et al. 2017). In the field, the presence of soiling is one of the most noticeable indications of the unsuccessful shot or inadequate evisceration practice (Avagnina et al. 2012; Paulsen and Winkelmayer 2004). Nevertheless, carcasses that are visually clean may also contain relevant MLs (Korkmaz et al. 2022a; Paulsen and Schopf 2016; Paulsen et al. 2022). Rinsing with water is

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one of the most recommended corrective measures to reduce the visible soiling of carcasses as well as the resulting ML (Deutz 2014). As previously discussed for roe deer shot by certified hunters in Germany, rinsing with drinking water at ambient temperature was not always effective, and the corresponding reduction of the ML was not always reproducible (Korkmaz et al. 2022a). The latter is partly due to the fact that the initial ML of surfaces of roe deer's abdominal cavity (belly flap or fillet) was not always visibly associated with soiling. Thus, this incongruence impeded a clear statement on the effectiveness of rinsing on the ML reduction of soiled roe deer carcasses (Korkmaz et al. 2022a). In this study, we aimed to assess the effect of water rinsing on the ML of carcasses directly at harvesting in the field, with a limited set of samples. By experimentally soiling with gastrointestinal content (GIC), we intended to reproduce similar MLs for the rinsed and the unrinsed carcasses. And thus, eliminating this confounder and achieving comparability of ML values prior to treatment at a single hunting day with its particular weather conditions.

2 Results and discussion

Using deliberate contamination of the roe deer body cavities with a freshly prepared mixture of GIC, we intended to standardize the initial microbial conditions of body cavities to better elucidate the impact of rinsing on the ML of game carcasses. Experiments were conducted directly in the hunting ground using freshly shot carcasses under field conditions. Table 1 shows an overview of the basic experimental conditions of each trial. In the context of the practical focus of the experiments, some ambient as well as individual factors could not be controlled, and consequently differed between trials (Table 1). Since ambient temperature, have been described to affect the microbiological quality of game carcasses during the harvesting (Korkmaz et al. 2022b; Paulsen and Winkelmayer 2004; Branciari et al. 2020), the comparing of rinsed and unrinsed carcasses were evaluated separately within each trial. Further influencing factors such as body weight, elapsed time between shot and evisceration of the animals, and the time between the evisceration and the further processing (Korkmaz et al. 2022b; Paulsen and Winkelmayer 2004; Paulsen et al. 2022) are additional challenges for the interpretation of the outcome of the experiments. Despite the low number of carcasses and the different conditions between the hunts for the microbial contamination and development, the subsequent inclusion of further animals for experimental soiling was not undertaken for ethical reasons. The number of animals was also chosen based on results of a previous study, where a higher sample number did not allow further insights on the microbiological effects of rinsing soiled body cavities (Korkmaz et al. 2022a). Nevertheless, we stress that the practical context of this study as well as the use of 2 different muscle surfaces bring valuable results. We obtained baseline data in the field for a basic measure of hygiene that is important for hunters and stakeholders alike. It could be a critical point in the primary production chain of game meat.

In accordance with previous reports (Korkmaz et al. 2022a; Paulsen and Schopf 2016), the ML of visually clean body cavities considerably differed prior to soiling both between and within the trials. The bacteriological load on the surface samples for every trial are presented in Fig. 1. According to an investigation of Paulsen et al. (2022) in 352 hunted roe deer, bacterial counts of clean body cavities can differ considerably between animals even without perforation of structures of the gastrointestinal tract. As expected, sampling before soiling (BS) vs. sampling after soiling (AS) resulted in a general increase of the ML in belly flaps and fillets of soiled carcasses (Fig. 1). The increase in bacterial counts occurred independently from the initial ML and with single exceptions for the Enterobacteriaceae and E. coli. However, the level of increased ML differed between trials, which may be related to the different bacterial composition of GIC mixtures. In trial 1 and 2, the bacterial load in the mixture ranged from 4.3 to 7.1 \log_{10} CFU/g for the total aerobic colony count (TAC), from 3.7 to 4.4 log₁₀ CFU/g for Pseudomonas spp., from 2.5 to 4.2 log₁₀ CFU/g for Lactobacillus spp., and from 2.5 to 4.7 \log_{10} CFU/g for *Enterobacteriaceae*. *E. coli* was either below the limit of detection or reached counts of 4.7 \log_{10} CFU/g. Due to technical issues, data of trial 3 was not considered. Although, the proportions of the GIC for preparation of the mixture was comparable between trials, divergence on bacterial content of the mixtures may have occurred due to differences in the microbial content in the segments of the gastrointestinal tract, and may explain the apparent incongruences BS to AS in unrinsed carcasses, especially for both fecal indicators (Fig. 1). The microbial communities may differ between the sections of the gastrointestinal tract as well as between the studied roe deer individuals (Li et al. 2014), which may be influenced by the diet composition in different habitats (König et al. 2020; Liu et al. 2019).

Rinsing soiled belly flap surfaces consistently reduced the TAC as well as the Pseudomonas spp. count to a level similar to or lower than the initial ML, as determined by sampling 20 min after rinsing (Fig. 1). However, the effects of rinsing were incongruent between trials for Lactobacillus spp., Enterobacteriaceae, and E. coli on the same surfaces. In contrast to belly flaps and with one exception for E. coli (trial 1), the ML on rinsed fillet surfaces remained above the initial bacterial counts, with a maximal difference of 1.40 log₁₀ CFU/cm² observed for Lactobacillus spp. in trial 1 (Fig. 1). Differences on the effect of rinsing are possibly due to the more irregular surface of fillets compared to belly flaps after field evisceration. The irregular surface may have promoted bacterial attachment and consequently reduced the short-term effects of rinsing (Delaquis and Mccurdy 1990; Dickson 1988).

Regarding the MLs after cold storage, bacterial development during 3 days at +4 °C did not only differ between rinsed and unrinsed body cavities, but also between meat cuts of single animals. While counts for TAC (trial 1 - 3), Pseudomonas spp. (trial 2 and 3), Lactobacillus spp. (trial 1 and 3) and Enterobacteriaceae (trial 1 and 3) considerably increased on the rinsed belly flap surfaces, slight reductions of the TAC (trial 1 and 2), the counts of Lactobacillus spp. (trial 1 and 2) as well as the counts of E. coli (trial 1 and 2) were observed for rinsed fillets after the storage (Fig. 1). Interestingly, the ML decreased in 2 of 3 unrinsed body cavities, disregarding the meat cuts after the 3-day cold storage, except for the counts of Pseudomonas spp. on fillets. Thus, these results support the hypothesis that residual water may promote bacterial growth on meat surfaces (Sofos 2014).

Trial no	Carcass no	Sex	Body	Time	Water	Temperatur	e (°C)				ЬH			
			weight (kg ^a)	('mm')	volume (ml ^c)	Ambient	Pelvis ^d		Back ^e		Pelvis ^d		Back ^e	
			1				At hunting day	After cold storage	At hunting day	After cold storage	At hunting day	After cold storage	At hunting day	After cold storage
1	1^{f}	Male	11	145	1680	11	33	7	28	6	6.7	6.4	5.8	6.2
	2	Female	18	180	I		27	4	28	4	5.5	5.8	5.5	6.0
2	ю	Female	9	75	1840	0	20	3	19	2	5.7	5.7	5.5	5.6
	4	Male	13	150	Ι		14	4	10	2	5.5	5.7	5.5	5.9
3	5	Male	14	240	7200	9	34	5	29	5	5.6	5.8	5.7	5.7
	9	Female	12	45	I		19	5	19	5	5.8	6.1	5.7	6.0
^a After evi	sceration of the	carcass												

Table 1 Description of natural experimental conditions for inoculated and rinsed (n=3) or unrinsed (n=3) roe deer carcasses at each trial

^fEviscerated without opening the pelvis, all other carcasses were eviscerated with opening pelvis

^eTemperature or pH measured in the muscle between the 13th and 14th spinous process of the thoracic spine

^dTemperature or pH measured in the muscle close to the pelvis

^bElapsed between killing and evisceration

^cUsed for rinsing the carcass



Fig. 1 Bacterial counts determined on rinsed and unrinsed meat surface of roe deer body cavities that were intentionally soiled with GIC mixture in 3 different experimental trials (1 - 3). Total aerobic colony count (A), *Pseudomonas* spp. (B), *Lactobacillus* spp. (C), *Enterobacteriaceae* (D), *E. coli* (E). Sampling was performed before soiling

(BS), after soiling (AS) or after soiling and rinsing (ASR), and after cold storage for 3 days at +4 °C (AC). The values are presented for individual carcasses; values below the limit of detection are given as 0

3 Conclusions

In conclusion, the microbiological investigation after this experimental approach showed that rinsing of soiled roe deer body cavities may acutely reduce the bacterial load directly caused by fresh soiling under field conditions. However, rinsing with water may further facilitate the growth of remaining bacteria during cold storage. Further experimental studies are required to better understand the effects of rinsing on the shelf life of game meat under different storage temperatures. Based on this and previous observations (Korkmaz et al. 2022a) as well as considering ethical issues, soiling complete body cavities should be avoided in future studies, since the practice compromises the hygienic quality of the whole carcass and the obtained information is limited. Instead, similar to previous studies that examined the effect of washing meat from slaughtered animals (Castillo et al. 1998), future studies should rather use meat cuts of game carcasses including muscles with different surface characteristics and perform them under controlled laboratory conditions. Because there, potential influencing factors such as bacterial contamination load, rinsing regime or temperature can be modulated. This may also permit i.e. the examination of hot water rinsing, which was reported to reduce bacterial counts on livestock carcasses (Bosilevac et al. 2006). Overall, regardless of whether a carcass is visually clean or whether rinsing successfully removed visual soiling, all game products should be cooked to a core temperature of 70 °C for at least 2 min prior consumption.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s00003-023-01417-0.

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Declarations

Conflict of interest The authors declare that they have no conflict of interest.

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