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# Microbial effect of steam vacuum pasteurisation implemented after slaughtering and dressing of sheep and lamb

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ABSTRACT

The main objective of the study was to assess the effect of steam vacuum pasteurisation on carcass contamination with focus on *Escherichia coli*, *Enterobacteriaceae* and total plate count (TPC). Additionally, the effect of an additional tryptone soy agar (TSA) step for resuscitation of *Enterobacteriaceae* after steam vacuum pasteurisation was investigated. Steam vacuum pasteurisation was applied at a temperature of >82 °C for a duration of 10 s on sheep and lamb carcasses (n = 120). Samples were taken immediately: i) after trimming just before the use of steam vacuum and ii) after use of steam vacuum. Nordic Committee on Food Analysis methods were used in microbial analyses. The differences in log reduction were found significant for all of the three microorganisms ($p < 0.05$). For TPC, the general reduction was a 0.65 $\log_{10}$ in the number of colony forming units (CFU) per $cm^2$. For *E. coli*, the median reduction effect on carcasses positive before decontamination was 1.1 $\log_{10}$ CFU/$cm^2$. A large variability of the effect was however found, with 50% of the figures ranging from a 0.24 to 1.62 $\log_{10}$ CFU/$cm^2$ reduction and a 10–90% range of 0–2.1. The number of positive carcasses with *Enterobacteriaceae* after steam vacuum pasteurisation was higher in samples where TSA + violet red bile glucose agar (VRBGA) was used compared to samples where only VRBGA was used ($p < 0.01$). Steam vacuum pasteurisation was found efficient in reducing the total count, read as TPC, as well as the level of *E. coli* and *Enterobacteriaceae*.

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## 1. Introduction

The main principle of steam vacuum pasteurisation is the decontamination of carcass surfaces using hot steam at temperatures ranged 65–100 °C for duration of 5 to 20 s and removal of contaminants from the carcass surfaces by a vacuum system. The use of steam vacuum pasteurisation has been proved to reduce bacterial contamination of raw meat by mean values that ranged from 1 to 4 log CFU/$cm^2$ (Dorsa, Cutter, Siragusa, & Koohmaraie, 1996; Steenberg, Teilmann, & Christensen, 2006; Tarp, 2004). However, good manufacture practice (GMP) and good hygiene practice (GHP) as prerequisites for ensuring minimal microbial contamination and a successful implementation of a hazard analysis critical control point (HACCP) plan are the basis for an optimal hygienic effect of steam vacuum pasteurisation.

Unlike other published decontamination studies, the present work is the only one to focus on steam vacuum pasteurisation of sheep and lamb carcasses taking into consideration resuscitation of the stressed bacteria. This aspect was taken into account by the use of isolation agar suitable for this purpose both before and after steam vacuum pasteurisation. In contrast to most other studies our study was carried out in an abattoir where steam vacuum pasteurisation had already been implemented and used on a daily basis. The only steam vacuum pasteurisation study on lamb (Steenberg et al., 2006) is available as a report where the technique was under trial and had not been implemented in the abattoir as in our study. The formerly published scientific articles on steam vacuum pasteurisation have focused on other species than sheep and a few have examined and revealed the effect on naturally contaminated carcasses in a commercial plant (Corantin et al., 2005; Dorsa et al., 1996; Minihan, Whyte, O'Mahony, & Collins, 2003; Nutsch et al., 1997, 1998).

An outbreak caused by enterohaemorrhagic *Escherichia coli* (EHEC) O103:H25 occurred in Norway between January and March 2006, with 17 confirmed human cases (Norwegian Scientific Committee for Food Safety, 2007). Ten of the 17 cases were children who developed haemorrhagic uremic syndrome (HUS). One child died as a consequence of HUS complications. Norwegian dried fermented mutton sausages were found to be the source of the outbreak. As a result of the *E. coli* O103:H25 outbreak, new principles had to be implemented to support the manual removal of visible contamination (trimming) to remove the invisible contamination of carcasses. The Norwegian cooperative company Nortura, which slaughters about 70% of all

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mammalian food animals in Norway, decided to implement steam vacuum for pasteurisation of carcasses in all ovine slaughter lines in Norway. Food animals represent a reservoir of enteric pathogens. In that context, the slaughtering of sheep involves greater challenges because the animal is relatively small and has a wool fleece, thus increasing the risk of surface contamination at dehiding (Buncic, 2006), which might result in suboptimal hygiene during slaughtering compared with the slaughtering of cattle. Hence, steam vacuum might be more important as a supplement for good hygienic practices in slaughter of sheep and lambs compared to other species.

Pasteurisation of carcasses with hot water or steam has proved to be efficient in reducing bacterial contamination (Sofos, Belk, & Smith, 1999). Other decontamination technologies such as organic acids have also been tested and used for the decontamination of meat (Castillo, Lucia, Goodson, Savell, & Acuff, 1998; Gorman, Sofos, Morgan, Schmidt, & Smith, 1995; Sofos, 1998; Sofos & Smith, 1998). The use of hot water, steam or organic acids has been shown to be an efficient tool for significantly reducing contamination of beef with *E. coli* O157:H7 (Phebus et al., 1997; Sofos et al., 1999).

The aims of our study were:

i) to evaluate the microbiological effects of steam vacuum pasteurisation of ovine carcasses after slaughtering and dressing; and
ii) to investigate the effect of an extra tryptone soy agar (TSA) step for resuscitation of *Enterobacteriaceae* after the use of steam pasteurisation.

## 2. Materials and methods

### 2.1. The slaughter line

The study was conducted in an authorized commercial abattoir in the Northern part of Norway during the seasonal sheep slaughter in the period between September and November. The plant has implemented a HACCP plan and is certified according to the standard required by the British Retail Consortium (BRC). The use of steam vacuum pasteurisation (time/area exposed) might be regarded as a critical control point (CCP) but the plant has not listed this operation as a CCP. However, the abattoir has listed the final visual control as a CCP. At this step, two operators standing at different heights to allow optimal overview go visually through all carcasses after they have passed the veterinary meat inspection and remove any visible contamination using sterile knives. The sheep and lambs were shorn in the lairage a few hours prior to slaughter. Mean weight of sheep and lamb carcasses was 32 and 18 kg, respectively. The line speed was approximately 180 animals per hour. The oesophagus was not rodded as both skinning and evisceration were done with the carcass suspended by its forelimbs. Bagging of rectum by a plastic bag was not performed. However, small balls made of normal drying roll paper (Engros, Narvik, Norway) were forced into the rectum using a hard plastic rod. Thus, faeces were pressed back into the rectum and hence made it easier to eviscerate without causing faecal contamination of the perianal area. Circum anal incision took place while animals were suspending horizontally by all four limbs.

The steam vacuum pasteurisation equipment (SFK System AS, Kolding, Denmark) consisted of two main parts; steam and vacuum units. The steam decontaminates and loosens faecal materials from the carcass surface to facilitate removal by vacuum unit. The steam had a range of temperatures (82–95 °C) and was applied for 10 s on the chest and area around circum anal incision (i.e., spot decontamination) using a hand-held device ended with a nozzle (Steenberg et al., 2006). Dimension of the nozzle was 8 × 3 cm and the diameter of the steam channels was approx. 1.5 mm. The steam was produced from potable water (5 kg per hour) and pressure of the steam entering the hand-held device was 1.5 bar. The steam temperature was measured from the nozzle using a digital thermometer (Testo 105, Testo Limited, Alton, UK).

The removed contaminants were collected in a closed container via a special tube and then emptied by the end of the day. The steam vacuum equipment was fixed on a wall and had two hand-held devices equipped by two hoses each, one for steam and another for vacuum, extended to the terminal in slaughter line where the spot decontamination was performed. Two operators were located in a place between carcass trimming and classification terminals and operated the steam-vacuum equipment. One operator was in charge of steaming the chest area and another of the circle around the circum anal incision.

### 2.2. Collection of samples

The animals sampled were classified as lambs (<1 year of age) or sheep (>1 year of age). A total number of 120 naturally contaminated carcasses (23 sheep and 97 lambs) were sampled in the study. The collection of samples was completed during five different working days within five weeks. The sampling was divided into three intervals during one work-day:

i. In the beginning of the work-day; samples were taken during the first and the second day of sampling (n = 40 carcasses);
ii. in the middle of the work-day; fifty samples were taken during the third and the fifth day of sampling (n = 50 carcasses); and
iii. during the last 3 h of the work-day; thirty samples were taken during the fourth sampling day (n = 30 carcasses).

The idea of dividing the sampling period into three intervals was based on dividing the approximately nine-hour work-day (09:00–18:00) into three equal intervals of 3 h each. Thus, all day intervals were included in the sample collection. The same sheep and lamb carcasses (n = 120 carcasses) were swabbed two times (n = 240 samples):

i. Immediately after trimming and before use of the steam vacuum equipment.
ii. After use of the steam-vacuum equipment.

Two sampling sites were chosen; one on the chest (sampling site 1) and a circle around the circum anal incision (sampling site 2). The 240 samples were equally divided between the two sites and performances of sampling for the two sites were alternated.

The swabbing area was 100 $cm^2$, sampling sites are shown in Fig. 1. For the chest site, right and left sides and vice versa were used respectively before and after steam vacuum pasteurisation at the same carcass. The circle around the anus was divided into two half circles. A half circle was sampled before and the other after the use of steam vacuum. Stainless steel templates (Mathiassen Ventilasjon Blikken, Finnsnes, Norway) with an area of 100 $cm^2$ each were used to accomplish a precise swabbing area of 100 $cm^2$ in the chest and the half circle around circum anal incision sites. In short, an area of 200 $cm^2$ around circum anal incision was obtained by calculating mean diameters of circum anal incision for 10 sheep and lamb carcasses. The two diameters were then used to calculate the area of the circum anal incision for both sheep and lamb carcasses. Given the area, diameter of circum anal incision and the area around circum anal incision, we could calculate the diameter and area of the circle around circum anal incision. Based on these calculations, a template was designed to fit the swabbing area around circum anal incision as depicted in Fig. 1. The templates were made sterile by sinking in sterilisers available in slaughter line using potable water at a temperature of ≥82 °C.

Each sterile spongesicle swab (Biotrace International Plc., Bridgend, Wales, UK) was made wet by adding 10 ml of the 25 ml buffered peptone (Biotrace International Plc.) before sample collection. The remaining 15 ml buffered peptone was added immediately to the wet swab after the sample collection. Swabs were put back immediately after completion of sampling in their sterile bags and sealed. Samples were kept cool in a cooling box at a temperature of approximately 4 °C and shortly freighted to Senja Lab, Finnsnes, Norway, located at a distance

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