Changes in muscle temperature and pH in topside muscle (*M. semimembranosus*) in cattle

Potential effects on meat quality

Tova Pettersson

Independent project • 30 credits
Swedish University of Agricultural Sciences, SLU
Department of Animal Biosciences
Agronom djur - Masterprogram
Uppsala 2026

Changes in muscle temperature and pH in topside muscle (*M. Semimembranosus*) in cattle – Potential effects on meat quality

Temperatur och pH-förändringar i innanlåret hos nötkreatur efter slakt – potentiella effekter på köttkvalitet

Tova Pettersson

Supervisor:	**Elin Stenberg, SLU, Applied Animal Science and Welfare**
Examiner:	Anders Karlsson, SLU, Applied Animal Science and Welfare

Credits:	30 credits
Level:	A2E
Course title:	Självständigt arbete i husdjursvetenskap för Agronomer
Course code:	EX1023
Programme/education:	Agronom djur - Masterprogram
Course coordinating dept:	Department of Animal Biosciences
Place of publication:	Uppsala
Year of publication:	2026

Keywords:	PSE, heat-shortening, pre-slaughter stress, blood lactate, lairage time, electrical stimulation

Swedish University of Agricultural Sciences
Faculty of Veterinary Medicine and Animal Science
Department of Animal Biosciences

Abstract

Meat quality is of importance regarding several characteristics like meat colour, visible fat and price, which is important for the consumers when buying meat at the store. Temperature and pH-decline postmortem is of great importance when discussing the quality of the meat. There are several factors along the whole production line that will influence the pH- and temperature decline that occur postmortem, which in turn have an effect on meat quality in the final product. Such factors could be feeding regimes pre-slaughter, age and breed, but also handling of the animals that might cause stress on farm, during transport and at the abattoir. The purpose of this study was therefore to investigate how pH- and temperature decline occurs in beef carcasses at a specific abattoir to investigate if the slaughter process is optimized or can be optimized to ensure a good final meat quality. The present study included 21 bulls, 11 heifers and 13 dairy cows that were slaughtered at two different occasions in the winter season. The carcasses were hanged in the Achilles tendon and half of the carcasses were electrical stimulated. The blood lactate was measured in the exsanguination blood, and the pH- and temperature meters were inserted into the left *semimembranosus* muscle, which registered values every tenth minutes for 24 hours. The results of the study showed that most of the carcasses reached pH 6 at high muscle temperatures, which increases the risk for heat-shortening and PSE. To ensure that pH 6 is reached at desirable muscle temperatures it is important to evaluate the slaughter process at the abattoir, so that animals are in a calm and suitable environment to reduce stressors before slaughter but also that the carcasses enter the coolers fast enough. No differences were seen regarding temperature- and pH-decline during the first 24 hours between sexes, but the muscle temperature at 24 hours postmortem ($p=0.0417$) were significantly higher for cows compared to heifers and bulls. Fat score had a positive correlation ($r=0.64$) to muscle temperature, which means that higher fat scores result in higher muscle temperatures in the carcasses. No differences were seen regarding the electrical stimulation for all parameters tested. However, the lairage time, overnight or not, showed that animals that stayed overnight had higher lactate concentration ($p=0.0070$) in exsanguination blood compared to animals slaughtered the same day due to different animal material in the groups. All bulls slaughtered stayed overnight, the cows were slaughtered the same day as arrival, and the heifers occurred in both groups. Bulls had a higher blood lactate concentration ($p=0.0002$) compared to heifers and cows, which is the explanation for the differences regarding lairage times´ effect on blood lactate concentration. But there were no differences in pH- and temperature decline between lairage times. According to the blood lactate results, animals may have reacted differently to the pre-slaughter situation, and some may have been more stressed/excited than others, it could therefore be of importance to evaluate if there are some steps pre-slaughter that can be optimized to reduce stress/excitement for the animals. Results from this study will be used to discuss optimization of the slaughter process at the specific abattoir.

Keywords: PSE, heat-shortening, pre-slaughter stress, blood lactate, lairage time, electrical stimulation

Table of contents

List of tables

List of figures

Abbreviations

ATP	Adenosine triphosphate
DFD	Dark, firm and dry
ES	Electrical stimulation
MSA	Meat Standards Australia
PSE	Pale, soft and exudative

1. Introduction

Meat quality is of main importance regarding several characteristics of the final product. There are several different factors that the consumers consider when buying meat at the store. The brand and the origin of the meat lead to different expectations on the meat quality (Banović et al. 2009). Acebrón and Dopico (2000) found that the most important factors for consumers when buying beef meat were the colour, visible fat, freshness, and price and presentation of the meat. Acebrón and Dopico (2000) also explained that the most appealing colour according to their study were the brighter colour compared to darker meat, but Steenkamp and Van Trijp (1996) found that the redness of the meat were of importance for the consumers, where darker red colour were more desired, which shows that it varies depending on consumer preferences. Meat colour is affected by the content of myoglobin, but also the muscle composition, physical state and the ultimate pH (Abril et al. 2001). The quality and colour of the meat can be affected by numerous factors from the live animal until the final product (Mullen et al. 2000; Abril et al. 2001). Meat quality can vary between different breeds, ages or feeding strategies, but the handling of the animals on farm, during transport and at the abattoir plays a role regarding stress, which in turn can have a negative impact on the quality. When an animal is stressed the heart rate and blood pressure will increase, but also the levels of blood lactate (Coombes et al. 2014; Boles et al. 2015). Lactate is always produced in the body as a product of the glycolysis (Coombes et al. 2014) but can be a measurement of short-term stress pre-slaughter. Different techniques used during the slaughter process can also affect the meat quality (Mullen et al. 2000). For example, effective postmortem chilling is essential to ensure that carcasses reach the optimal temperature/pH-window (reach pH 6 within 15 to 35 ºC according to MSA). The combined effect of pH and temperature is a well-known factor influencing several meat quality attributes of the final product (Marsh et al. 1981; Meat & Livestock Australia n.d.). Cold-shortening, heat-shortening, dark, firm and dry meat (DFD), and pale, soft and exudative meat (PSE) are examples of quality problems that can occur when the pH and/or temperature decline in the muscle is not ideal.

An abattoir in Sweden has noticed variations in meat quality in *Musculus semimembranosus* in cattle. Meat has appeared more pale in colour and excessive fluid is detected in the vacuum packaged meat. The purpose with this study were therefore to investigate if there are any factors at the slaughter line (from stunning to chilled carcasses) that can be improved in the slaughter system to ensure high quality meat (decrease the incidence of quality problems). In this study, the effect of sex, electrical stimulation and lairage time have been evaluated to measure

effects on blood lactate concentration, slaughter weight, conformation score, fat score, pH-decline and muscle temperature decline.

1.1 Research Questions

The research questions are based on the aim and are the following:

- Does lactate levels, measured from exsanguination blood, affect the decline in pH?
- How does the pH and temperature change in the topside muscle during the first 24 hours after slaughter?
- Are muscle pH and temperature lowered to an acceptable level in the topside muscle, related to stated recommendations?
- Does electrical stimulation affect temp@pH_6 or pH_{24}?
- Are there any differences in temp@pH_6, pH_{24} and $temp_{24}$ in *M. Semimembranosus* from bulls, heifers and dairy cows?
- Does lairage time (overnight or not) influence blood lactate concentration, pH_{24}, $temp_{24}$ and temp@pH_6?
- Which factors in the system could potentially be changed to optimize the slaughter process?

2. Background

2.1 Muscle to meat

The transformation of muscle into meat is a key factor affecting the quality attributes of the final product, like juiciness, tenderness and the meat colour. In the live animal, the muscle maintains energy from adenosine triphosphate (ATP) that is broken down from the glycogen stores, which is needed for the muscle to be able to contract and relax (Paredi et al. 2012; Braden 2013). When the animal has been exsanguinated, most of the blood leaves the body, which also leads to a decrease of oxygen to the muscles (Hui 2012; Paredi et al. 2012; Braden 2013). But there will still be some oxygen bound to the myoglobin that remains in the muscles, and this small amount of oxygen makes it possible for the glycolysis to maintain, but the oxygen will drain rapidly (Paredi et al. 2012; Braden 2013). Braden (2013) explained that when the oxygen is completely depleted, the metabolism of glycogen will convert from aerobic to anaerobic. During the anaerobic glycolysis pyruvate will be fermented to ATP, carbon dioxide and lactic acid. The small amount of ATP that is produced through the anaerobic glycolysis will quickly be used and the store of glycogen will also be depleted. The amount of lactic acid increases, resulting in a drop of pH in the muscles (Paredi et al. 2012; Braden 2013). When ATP is depleted, the muscle will remain stiff, and completion of *rigor mortis* (stiffness after death) has occurred. The time *rigor mortis* is fully completed vary from 1 hour to 24 hours, and there can be differences between species, muscle type, and also depend on the conditions experienced pre- and post-slaughter (Paredi et al. 2012; Matarneh et al. 2023). Completion of *rigor mortis* in beef normally occur at 24 hours postmortem (Bodwell et al. 1965).

2.2 Lactate and its´ impact on meat quality

Exercise in an animal lead to increased heart rate, blood pressure, respiration and levels of blood lactate (Coombes et al. 2014; Boles et al. 2015). These reactions in the body also occur when an animal is stressed. Lactate is the product of the glycolysis, when glycogen is used to produce energy (Coombes et al. 2014). A sufficient amount of glycogen stored in the body is a requirement for an adequate pH-decline post-slaughter to not negatively affect the meat quality. A reduction of glycogen stored in muscle can be due to stress but could also be caused by feeding strategies or temperament. An increased concentration of blood lactate in exsanguination blood can therefore indicate a lower amount of glycogen in muscle at slaughter, which can result in an insufficient pH-decline in muscle post-slaughter, and the meat risks to be classified as dark, firm and dry (DFD)

(Coombes et al. 2014). High concentrations of lactate postmortem can also indicate PSE, as high concentrations might result in a rapid pH-decline in muscle just after slaughter (Aalhus et al. 1998). A study by Immonen et al. (2000) compared low energy diet (10.8 MJ ME) to a high energy diet (12.9 MJ ME) prior to slaughter, which resulted in a higher glycogen storage with the high energy diet, but also a smaller loss of glycogen during transport. The authors explained that the high energy diet served as a protection for stressors, due to adequate amounts of gluconeogenic precursors, which resulted in smaller losses during transport. The higher concentrations of glycogen stored postmortem due to the high energy diet, also resulted in a sufficient pH-decline and a reduced frequency of dark cutting compared to the low energy diet (Immonen et al. 2000).

Reported lactate levels vary considerably between studies. In a Swedish study by Hultgren et al. (2022), measured concentrations ranged from 0.9 to 18.4 mmol/L across different categories of cattle (cows, bulls, heifers and steers), with a mean of 4.74 mmol/L. A study by Sullivan et al. (2024) included steers and heifers raised in feedlots, and reported values ranging from 0.7 mmol/L to 14.80 mmol/L, with a mean of 5.72 mmol/L. The highest concentrations reported in the literature were observed in heifers and steers, where the values ranged from 5.7 to 20.3 mmol/L with a mean of 12 mmol/L (Gruber et al. 2010).

Probst et al. (2013) investigated the effect of gentle touching five weeks before slaughter, meaning that the cattle (bulls, females and castrated males) were stroked lightly with fingers and hands once or twice a week in the head- and neck region, and these were compared to a control group that did not receive this treatment. The treatment sessions of gentle touching occurred for 4 minutes with a break of 45 minutes before the sessions were made once again. A few days after the gentle touching treatments were done, an unfamiliar person who had not been in touch with the animals did an avoidance test to see how they reacted to an unfamiliar human. Probst et al. (2013) claimed that the treatment resulted in a decreased avoidance distance, and the animals were less stressed when they became accustomed to being handled. An indication of less stress for the treated animals could be seen in the lactate concentrations postmortem. Probst et al. (2013) found lactate concentrations between 2.4 and 13.9 mmol/L, with the highest values in the control group, which did not receive the gentle touching treatment. The study included animals of different crossbreds, and some were reared more extensively with less human contact, while other were reared more intensively and had had more contact with humans during their life, but the same patterns of the treatment were seen between the different groups.

Consistently, all studies agree that pre-slaughter stress, like transport, human contact and handling at the abattoir contributes to elevated levels of blood lactate

postmortem (Gruber et al. 2010; Probst et al. 2013; Hultgren et al. 2022; Sullivan et al. 2024). Hultgren et al. (2022) also included loading and unload of animals, transport time, lairage area, contact with both unfamiliar animals and humans, lack of water or feed, but also environmental factors like the weather. Sullivan et al. (2024) included distance travelled to the abattoir, truck waiting time when arriving to the abattoir, space allowance, time in the holding pens before slaughter and the season the animals were slaughtered. Sullivan et al. (2024) observed differences between season and weight, with higher blood lactate levels during summer compared to winter caused by heat stress, and that heavier carcasses tended to have a higher average lactate concentration, but the authors did not conclude any reasons for elevated lactate concentrations for the heavier carcasses.

2.3 Muscle temperature and pH

2.3.1 Recommendations

Muscle temperature at pH 6

The muscle temperature and pH have an important role in the quality of meat, but the parameters also have an important interaction with each other. According to Meat Standards Australia (MSA) the muscle needs to pass pH 6.0 in the temperature window 15 °C and 35 °C. However, if the rate of pH-decline is too slow or too rapid it might result in quality problems, like heat-shortening, cold-shortening or PSE. Locker and Hagyard (1963) found that the lowest risk for shortening occurred in the temperature interval 14-19 °C, which means that heat-shortening still can occur when pH 6 is reached within the interval 15-35 °C, meaning that a lower muscle temperature than 35 °C is to aim for to reduce the severity of heat-shortening. This statement was confirmed by Devine et al. (1999) who found that the shortening was much greater at 35°C compared to at 15°C. Locker and Hagyard (1963), also explained that the risk for cold shortening increased with muscle temperatures below 14 °C, which corresponds to the recommendations by MSA.

Ultimate pH

The recommendation for the ultimate pH, often measured 24 hours postmortem, is 5.3 to 5.7 according to MSA. Meat Standard Sweden (n.d) have set a range from 5.3 to 5.8 (inspired by MSA), which are values from a research project and not values used in the industry. Brazil Beef Quality (n.d) have set an upper limit at pH 5.8 and in a study by Muižniece and Kairiša (2020), the desired pH range is from 5.3 to 5.8, with an upper limit of 5.9. When pH after 24 hours is lower or higher than this, there is a risk for reduced quality, which will be discussed further in section 2.3.2.

Temperature 24h postmortem

There are currently no specific recommendations in national meat standards regarding carcass temperature 24 hours postmortem in Sweden. According to European Union (EU) regulation (EC) No 853/2004 of the European Parliament and of the Council of 29 April 2004 laying down specific hygiene rules for food consumption of animal origin, biproducts should have reached a temperature of 3°C throughout the meat and other meat products should have reached 7 °C before it leaves the abattoir. According to the EU regulation (No 853/2004), carcasses or carcass parts can be allowed to be transported before reaching 7 °C. However, in the US, one of the critical points that are often used is that the surface temperature should be below 4 °C in the first 24 hours, to reduce bacterial growth on the carcasses (Djimsa et al. 2022).

2.3.2 Quality problems related to pH and temperature

The temperature in the muscle at the onset of *rigor mortis* plays an important role for meat quality in the final product (Braden 2013). The ultimate pH varies and is also of importance. The ultimate pH depends on the storage of glycogen, which in turn can be affected by several factors like exercise, feed and stress in different forms (Lawrie 1958). The glycogen stored in muscle postmortem is used to produce lactic acid, which is the cause for decrease of pH. The storage of glycogen is therefore a prerequisite for how much the pH will decrease. Heat shortening, cold shortening, DFD and PSE are common problems when temperature and pH does not hit the ideal window.

Shortening of muscles

Shortening of the muscles might occur when pH 6 is reached outside of the ideal window, which according to MSA is 15-35 °C for beef. According to Meat Standards Australia regarding sheep meat quality, it is explained that if the carcasses are chilled slow the chemical reactions in the carcasses will occur faster which results in a rapid pH decline. That might lead to heat-shortening, which could lead to quality problems in the final product like toughness, increased drip loss and paler colour of the meat. MSA regarding sheep also explained that an incorrect electrical stimulation (ES) can result in carcasses entering *rigor mortis* at a high temperature, above recommended values, which can cause problems with drip loss and colour instability. The lowest risk for shortening is in the interval 14-19 °C, with a maximum of shortening at 0 °C in *sternomandibularis* muscle in beef cattle (Locker & Hagyard 1963). Locker and Hagyard (1963) explained that when the temperature in the muscle is higher than 19 °C when passing *rigor mortis* (pH 6), the level of shortening often increases, which leads to a risk for heat-shortening, which is also called rigor shortening. The authors also explained that if the muscle temperature on the other hand is lower than 14 °C when *rigor*

mortis starts, there will be a risk for cold shortening (Locker & Hagyard 1963). Locker and Hagyard (1963) explained that the shortening in their study was more extensive at 2 °C compared to 37 °C, but they also found that the cold shortening occurred immediate in low muscle temperatures. Devine et al. (1999) and Hertzman et al. (1993) also found that the rigor shortening was more extensive the higher the muscle temperature, which agreed with Locker and Hagyard (1963). The study by Devine et al. (1999) included young bulls (18 months), where the muscle *muscularis longissimus thoracicus et lumborum* (LTL) were removed after slaughter to measure shortening and isometric tension. The ultimate pH was less than 5.6 in all carcasses used in the study (Devine et al. 1999). When entering rigor at 15 °C the muscle shortening was limited, approximately slightly over 10%, but increased with the muscle temperature and were the highest when entering rigor at 35 °C (35 % shortening). The study also showed that the tenderness was the best when rigor starts in 15 °C and decreases with higher muscle temperatures. (Devine et al. 1999). A similar study was made by Hertzman et al. (1993) on young bulls (1-1.5 years of age), where *M. biceps femoris*, *M. semitendinosus* and *M. semimembranosus* were used to compare the effect of rigor shortening at 15 °C versus 37 °C, and how ES (85V, 14Hz and 32 s) had an effect on shortening. In the study the muscles were cut out after exsanguination. The occurrence of *rigor mortis* was mainly affected by the temperature of the muscles compared to muscle type and ES. Hertzman et al. (1993) explained that shortening of the muscles, were much greater at 37 °C than 15 °C, and that ES did not seem to have an effect on rigor shortening at different muscle temperatures. The authors found that the shortening were 22 % higher at 37 °C compared to at 15 °C. Hertzman et al. (1993) also explained that the shortening of the muscles affected the quality of the final product, like tougher meat. The degree of cold shortening was evaluated in the study by Olsson et al. (1994) on young bulls, in *M. semimembranosus* and *M. longissimus dorsi*, and how ES effected the shortening. Authors found that the degree of cold shortening (%) when entering *rigor mortis* were higher the lower the muscle temperature when comparing, 10, 7, 4 and 1 °C. For *semimembranosus* muscle the degree of shortening was approximately 5, 10, 35 and 42 % when entering *rigor mortis* at muscle temperatures of 10, 7, 4 and 1 °C, while *longissimus dorsi* muscle had a shortening of 5, 20, 37, and 50 %, when entering *rigor mortis* at the same muscle temperatures. Olsson et al. (1994) presented the same findings as Hertzman et al. (1993), that the ES did not have an effect on the shortening of the muscles.

DFD

Dark, firm and dry meat (DFD), also called dark cutting is a quality problem that may occur when the ultimate pH in muscles is above the recommended upper values, which differ from 5.7 to 5.9 depending on the different standards

mentioned above (Muižniece & Kairiša 2020; Brazil Beef Quality n.d.; Meat & Livestock Australia n.d.; Meat Standard Sweden nöt n.d.). As Coombes et al. (2014) described, DFD is caused by a poor glycogen storage, leading to lower levels of blood lactate produced which in turn will result in a higher ultimate pH, leading to a higher risk for dark cutting. The causes for a depleted glycogen storage are described in section 2.2.

PSE

Pale, soft and exudative meat (PSE) can occur in beef when pH has dropped quickly, while the muscle temperature is still high (Aalhus et al. 1998). Causes for PSE is stress shortly before slaughter, mainly as a cause of handling at the abattoir, like regrouping, use of electric prods and crowding of animals. (Muižniece & Kairiša 2020).

In a study by Aalhus et al. (1998) a comparison was made between beef carcasses classified as PSE and normal carcasses, regarding muscle temperature, pH and other meat quality parameters. Aalhus et al. (1998) explained that the occurrence of PSE is not as commonly seen in beef as in pork, but the authors referred to two studies by Fischer et al. (1997), that explained that it is most common with PSE in the *psoas major*, *semimembranosus* muscle and the *adductor* muscle in beef, due to its´ slow heat loss, resulting in a high temperature postmortem in these muscles. The study by Aalhus et al. (1998) included ten PSE beef carcasses, and the animals were stunned with a captive bolt gun, and low voltage ES (21 V, 60 Hz, 25 A, for 40 seconds) were carried out on the carcasses during the exsanguination. Seven of the PSE beef carcasses were also electrical stimulated with an additional high voltage ES (470 V, 60 Hz, 1.5 A, for 1 minute) 45 minutes postmortem. Two of these seven carcasses that were exposed to both the low and high voltage ES were placed in a blast chiller (-40 °C) for two hours and then transferred to a cooler of 2 °C, while the rest were placed in the 2 °C cooler for all 24 hours. The control group were also stunned with a captive bolt gun, but no ES were used, and they were cooled in 2 °C for 24 hours. The pH was measured in the *longissimus lumborum* muscle at a depth of 3 centimetres. The pH in the muscle in normal beef carcasses at 45 minutes, 3 hours and 24 hours postmortem were 6.79, 6.27 and 5.69, respectively, and the ultimate pH were 5.55. In the PSE beef carcasses, the muscle pH was 5.98, 5.49 and 5.63 at 45 min, 3 h and 24 h postmortem, with an ultimate pH of 5.54. The muscle temperature in the normal beef carcasses was 39.8 °C, 27.3 °C and 3.9 °C at 45 min, 3 h and 24 h postmortem, respectively, while the PSE beef carcasses had a muscle temperature of 41.1 °C, 26.6 °C and 4.0 °C in the different time stamps. Aalhus et al. (1998) explained that the lower pH and higher muscle temperature at 45 minutes postmortem is typical in PSE carcasses. The differences in the carcasses regarding muscle temperature was seen until 3 hours postmortem, and then the groups did not differ. The authors also

found that the glycogen content (muscle samples taken 5 min, 3 h and 24 h after slaughter) were lower in PSE beef carcasses compared to normal, which is the result of a more rapid glycolysis. The lactate concentration (in muscle) was also higher in the PSE carcasses compared to normal. Aalhus et al. (1998) also found a big difference in drip loss between normal and PSE beef carcasses. Normal carcasses had a drip loss of 13.0 g/kg, while PSE carcasses had 35.8 g/kg, which might lead to poor water holding capacity and binding properties in PSE carcasses. (Aalhus et al. 1998). Authors explained that the characteristics of PSE beef and PSE pork is similar. The same study measured muscle temperature and pH for normal pork carcasses and PSE pork carcasses in the *Longissimus thoracis* and found the same pattern for pork as for beef. At 45 minutes postmortem, the muscle pH was lower and the muscle temperature higher in PSE pork carcasses compared to normal pork carcasses. The lactate concentration and drip loss were also higher in PSE pork carcasses compared to normal. The only thing that differed between the beef and pork carcasses was that PSE beef resulted in greater tenderness compared with normal beef carcasses, while in pork, PSE produced the opposite effect, yielding tougher carcasses than normal pork.

2.3.3 Animal and environmental factors affecting the pH-decline

There are several different factors that might contribute to the differences in pH-decline, like the sex and age of the animal, but also the season the animal is slaughtered, the lairage time at the abattoir and the use of electric prods, which will be further described in the text below. The muscle temperature is more indirectly affected by sex and age, due to changes in weight and conformation over time as well as equipment used. The effect of weight, fat and conformation is further discussed in section 2.3.4.

Sex

Sex is one of the discussed factors that has an effect on the pH-decline, where bulls generally have higher pH_{24} compared to other sexes (Page et al. 2001; Węglarz 2010; Marenčić et al. 2012; Muižniece & Kairiša 2020). Muižniece and Kairiša (2020) compared heifers and bulls, that had a live weight of 499 kg and 530 kg, respectively. Bulls had slightly higher pH_{24} (5.87) compared to heifers (5.66). The study had, as previously described, set a desired pH range between 5.30 and 5.80, with an upper limit of 5.90, which 35 % of the bulls, but only 13 % of heifers exceeded. The majority of the bulls fell within the desired range (65 %), and a small fraction (1 %) had lower values, 5.30 or lower. Among heifers, 86 % were in the desired range, with only 0.4 % with lower values ($pH \leq 5.30$). The authors explained that the differences in pH_{24} between the two sexes are due to heifer's higher stress-tolerance compared to bulls, because bulls are more effected

by hormones and are more often in a state to show dominance which can result in anxiety, which in turn result in higher stress that later on affect muscle pH (Muižniece & Kairiša 2020). The same findings were made in the study by Węglarz (2010) when comparing young bulls (<24 months), older bulls (>24 months), cows and heifers. Live weight differed between summer and winter slaughter seasons, ranging from 520–524 kg in young bulls, 590–636 kg in older bulls, 499–513 kg in cows, and 430–451 kg in heifers. The pH was measured 48 hours postmortem, where young bulls and older bulls had higher pH_{48} compared to the heifers and cows. The pH_{48} also varied depending on slaughter season, which will be discussed below. Young bulls ranged in mean from 5.94 to 6.10 and older bulls from 5.65 to 6.07 between season, while cows had 5.89 and heifers 5.63. Węglarz (2010) concluded that the higher pH_{48} in young bulls and older bulls compared to cows and heifers could be caused by that males are more sensitive to stress factors like heat. The authors also mentioned that there was a higher frequency of proper pH values (pH<5.80) during winter than summer, but that difference were only seen in heifers which during summer had 72.5 % proper values and during winter 86 % proper values. The cows, older bulls and younger bulls had 46.8 %, 68.8 % and 21 % proper pH values, respectively. Marenčić et al (2012) compared bulls (14-16 months) and heifers (13-15 months) regarding pH_{24} in *longissimus dorsi* muscle and found that bulls (5.59) had significantly higher pH_{24} compared to the heifers (5.56), which is consistent with Muižniece and Kairiša (2020) and Węglarz (2010). The differences in pH between the sexes is explained as due to differences in endogenous hormones and temperament (Marenčić et al. 2012). Another study by Page et al. (2001) compared muscle pH in *longissimus* muscle from heifers, steers and bulls. Steers had a hot carcass weight of 350 kg and a pH of 5.51, heifers had a weight of 319 kg and pH of 5.50, while bulls had a weight of 359 kg and a pH of 5.78. The findings by Page et al. (2001), showed that bulls had slightly higher pH than steers and heifers which is consistent with all the studies mentioned previously, but no differences between heifers and steers did occur (Page et al. 2001). However, another study found differences between steers (547 kg) and heifer (487 kg), where heifers (5.52) had slightly higher pH compared to steers (5.46) (Wulf et al. 1997).

The studies above shows that sex influences the meat quality regarding the pH 24 hours postmortem. Heifers generally fall within the desired pH range more consistently than bulls, while steers and cows tend to resemble heifers more closely. Bulls more frequently exceeded the upper limit, which Coombes et al. (2014) explained increases the risk of dark cutting, leading to dark, firm and dry meat.

Age

Age is one of that factors that might contribute to differences in pH-decline and pH_{24}, since differences have been found in studies. Muižniece and Kairiša (2020) compared the ages 12─17 months, 18─23 months and 24 months or older regarding the pH_{24} for bulls and heifers. Age of bulls did not influence the pH_{24}, but a difference could be seen in heifers, where heifers 24 months or older had higher pH_{24} compared to heifers slaughtered at a younger age. The differences between older and younger heifers are that older females tend to be more sensitive to stressors. The pH_{24} for the different ages of heifers were 5.63, 5.66 and 5.69 for 12─17 months, 18─23 months and 24 months or older, respectively. Regarding the pH_{24} for the bulls, the average varied from 5.85 to 5.88. Arik and Karaca (2017) on the other hand found a difference between ages in bulls in *longissimus thoracis* muscle. Bulls older than 30 months (6.05) had a higher pH_{24} compared to bulls younger than 30 months (5.76). In a study by Liza et al. (2024) no differences in pH_{24} in *longissimus dorsi* muscle could be found when comparing animal ages of 1.5 (18 months), 2 (24 months) and 2.5 (30 months) years. The pH_{24} were 5.44, 5.40 and 5.35 for 1.5, 2 and 2.5 years, respectively.

Season

The study previously described by Węglarz (2010) also investigated the effect of seasons on the different sexes. Seasons included were summer which had a mean temperature of 18 °C and winter with a temperature of -7 °C. The sexes compared were younger bulls (<24 months), older bulls (>24 months), cows and heifers. pH_{48} was measured in *M. longissimus thoracis*. According to Węglarz (2010), bulls, both the younger and the older, were affected by the seasons regarding the pH_{48}. For the younger bulls, the pH during the summer (6.10) were higher than during the winter (5.94), and the same pattern was seen in the older bulls where the pH during summer were 6.07 and during winter 5.65. Regarding the heifers and cows no differences were seen. The authors explained that these differences between sex and season is due to that bulls are more sensitive to heat stress and other stress factors compared to heifers and cows (Węglarz 2010). Marenčić et al. (2012) compared pH_{24} during summer, winter, spring and autumn in *longissimus dorsi*. The study included bulls (14–16 months) and heifers (13–15 months). Bulls had during winter a pH of 5.60, spring 5.56, summer 5.60 and autumn 5.58, while heifers had 5.55 during winter, spring and autumn, and 5.61 during summer. Marenčić et al. (2012) found significant differences for bulls with higher values during summer and winter compared to spring, and slightly higher compared to autumn. Regarding the heifers, the pH_{24} during summer were significantly higher compared to the other seasons. The explanation for the differences between seasons is the higher temperatures (above 23 °C) in the summer and lower

temperatures (below 3 °C) during the winter, which is more demanding for the animals to cope with, which results in increase pH-values due to stress.

Lairage time

Regarding the lairage time, a comparison was made by Ferguson et al. (2007) between steers that had stayed overnight (18 hours on lairage), and steers slaughtered the same day (three hours on lairage), and no differences in ultimate pH were found between treatments. The group that stayed three hours on lairage had an ultimate pH of 5.53, and the same value were reported from the group that had stayed a longer time (Ferguson et al. 2007). A similar study by Costa et al. (2019) compared three hours of lairage time versus twelve hours of lairage time on steers and found no differences in pH_{24}. Both groups had a pH_{24} value of 5.57 in *longissimus thoracis et lumborum* muscle. A difference was found by del Campo et al. (2010) on the other hand, regarding pH_{24} between steers that were 2.5 years old, where the two different treatments were three and 15 hours in the lairage area. The steers that stayed three hours had a pH_{24} of 5.82 and steers that stayed 15 hours had 5.56. The authors explained that the longer lairage time might had given the steers the opportunity to restock their glycogen storage, but another explanation could be that they had a chance to recover from the stress (transport) which resulted in less glycogen used before slaughter, leading to a better pH decline. However, del Campo et al (2010) also explained that the differences seen regarding the lairage times effect on the pH, can be due to several things, like the animal material, transport time, how the abattoir is designed, weather conditions and so on. This means that it is tough to conclude if the lairage time were the main difference between the groups or if other factors during the whole experience were of greater importance.

Use of electric prods

Electric prods are used in many abattoirs to simplify the transfer of the animals from unloading of trucks until reaching the stunning area. Warner et al. (2007) made a comparison between animals that were transferred with electric prodders to those that were not, regarding meat quality characteristics. The study included feedlot cattle, females and steers. The animals were divided into a stress group and a control group. The stress group were treated with 6 shocks from the electric prod under 5 to 10 minutes at 15 minutes before slaughter to induce an acute stress response in the animals, while the control group moved in their own pace with as less force as possible. Stunning occurred with a captive bolt gun and blood was collected during exsanguination. Muscle temperature and pH were measured in *longissimus thoracis* muscle every hour from one hour to five hours postmortem. The muscles were removed two days post-slaughter, where the left

sides were used for the consumers eating quality test. The results of the study showed that the stress group (7.12 mmol/L) had higher plasma lactate concentration at the time for exsanguination compared to the control group (4.29 mmol/L). Interestingly, temperature- and pH-decline did not differ between the treatments. Some of the carcasses (7 of 84) reached pH 6 within one hour, where five of them were from the stress group and two from the control group. The authors found that the drip loss were higher in the stress group (2.3 %) compared to the control (1.7 %). The study also included an evaluation from consumers, which resulted in that the meat from stressed animals were characterized as less tender, juicy and with lesser likable flavour. The authors concluded that the consumer eating quality may are easier to predict based on plasma lactate compared to the muscle pH, due to no differences in pH-decline postmortem between the treatments. It is also explained that the mechanism that caused the reduced tenderness is unknown, due to that tenderness normally is affected by pH and muscle temperature (Warner et al. 2007).

2.3.4 Slaughter weight, fatness and conformation – effect on temperature decline

In Sweden, carcass conformation is assessed using the EUROP classification system. The system is standardized for all countries that are a part of EU to ease trade between countries but there are national differences and adaptation to the system. Carcasses are graded with E, U, R, O or P, where E represents excellent muscling and P indicates poor muscling. Each class are divided into plus and minus subdivisions, leading to 15 classes for the conformation. E is the highest grade with high proportion of muscle relative to bone. The fat is classified from a scale from 1 to 5, where 1 indicate very low fat cover and 5 indicates very high fat cover. These classes are also divided with plus and minus subdivisions, leading to 15 classes for fat as well. A moderate fat level (3) is generally preferred (communicated thru each abattoirs own pricing system), cause excessive fat increases trimming of the carcass which gives an extra cost, while a very low fat cover reduces the value of certain cuts (sirloin, ribeye etc.). The final payment to the farmer is based on the classification of fat and conformation but also includes slaughter weight and animal characteristics like sex and age (bulls, younger bulls, steers, cows, heifers, veal calf, fatted calf etc.) (Svenskt Kött, n.d.).

That temperature decline takes longer time for heavier carcasses compared to lighter ones, has been presented in several studies for different muscles such as the *longissimus* muscle, *semimembranosus* muscle, *longissimus lumborum* muscle and *psoas major* muscle (Lochner et al. 1980; Lancaster et al. 2020; Djimsa et al. 2022). Lochner et al. (1980) compared the cooling rate in *longissimus* muscle in 15-month-old steers. The lighter ones had a live weight of 429 kg and a fat

thickness (12^{th} rib) of 0.5 cm, while the heavier group had a weight of 569 kg and a fat thickness of 2.6 cm. Lochner et al (1980) also included two different chilling methods where the two sides of one carcass were cooled with one of the different methods. The right sides of the carcasses were chilled with a rapid chilling method (-2 °C with air moving 90 m/min), and the left side were chilled slow at 9 °C without moving air and transferred to the same cooler as the right sides after 7.5 hours. All carcasses were transferred after 24 hours to a cooler with a temperature of 3 °C. Lochner et al. (1980) found that the heavier carcasses had a slower temperature decline compared to the lighter ones, but the different chilling methods also had an effect, especially in the lighter group. At two hours postmortem the temperature in *longissimus* muscle in the lighter carcasses with the slow cooling had a muscle temperature of 33 °C and with the rapid cooling 28 °C. The heavier carcasses with both chilling methods exceeded 35 °C at two hours postmortem. By 24 hours postmortem the leaner carcasses with both methods had reached a muscle temperature below 5 °C, while the heavier had a muscle temperature slightly over 5 °C. The lighter group that was chilled rapid, had a temperature decline that was twice as fast as the slower method when the temperature in *longissimus* muscle was 30 °C. The heavier carcasses also had a faster temperature decline with the rapid method even though the differences were not as great as for the lighter ones. Lancaster et al. (2020) also made a comparison between sizes of steers, where an average weight of 366 kg (hot carcass weight) were compared to oversized steers (462 kg) regarding the temperature decline in *semimembranosus* muscle, both at a superficial location and a deep location (undefined in the article). The carcasses were chilled in 0 °C. The results showed that the average weight cooled faster than the oversized, but there was no difference in the superficial location temperature decline between the two carcass sizes. The deep location cooled faster for the average weight compared to the oversized, but there was no difference in muscle temperature 48 hours postmortem. The pH also declined fastest in the oversized carcasses in the deep location of the *semimembranosus* muscle, but there was no difference in pH_{48}. Lancaster et al. (2020) concluded that a possible way to overcome the challenges with cooling of deep locations like the *semimembranosus* muscle is by hot boning, but the difficulties is that it might not be possible in the current slaughter chain in abattoirs.

Temperature decline also varies among different muscles. In a study by Djimsa et al. (2022), surface temperature and deep tissue temperature were recorded over time in *semimembranosus* muscle, *longissimus lumborum* and *psoas major* in two different abattoirs (A and B), to compare the effect of different carcass sizes. Two different chilling methods were also compared; one conventional with immediate spray chilling, and one delayed with delayed spray chilling. The heavy carcasses in abattoir A had a hot carcass weight of 445 kg and the lighter had a hot carcass

weight of 360 kg. In abattoir B the heavy carcasses weighted 420 kg and the lighter 355 kg. Djimsa et al. (2022), explained that the lesser mass on the lighter carcasses lead to a faster temperature decline in both abattoirs, but the authors found differences in temperature decline between different muscles where the *semimembranosus* muscle had the slowest temperature decline. The explanation for slower cooling of the *semimembranosus* muscle compared to *longissimus lumborum* and *psoas major*, is that the locations of the *semimembranosus* muscle is in the thickest and the heaviest part of the carcass, which also may have a thicker fat cover and the highest density. Djimsa et al. (2022) also explained that the muscle temperature in the deep tissue varied between lighter and heavier carcasses. The lighter had normally reached below 10 °C in *semimembranosus* muscle by 24 hours postmortem, while heavier carcasses had a muscle temperature above 13 °C at 24 hours postmortem. A pattern that also was seen in the study was that the pH normally dropped more quickly in the heavier carcasses compared to the lighter ones, which corresponds to the study by Lancaster et al. (2020). Djimsa et al. (2022) explained that the rapid pH-decline in heavy carcasses is due to the slow cooling. Jacob and Hopkins (2014) explained in a review that rapid pH decline at high muscle temperatures occurs because elevated temperatures increase the rate of biochemical reactions, leading to a faster drop in pH. Djimsa et al. (2022) and Lancaster et al. (2020) found that heavier carcasses exhibit a faster pH decline, likely due to an increased rate of glycolysis. The higher carcass mass led to elevated internal temperatures, which accelerated glycolysis and consequently resulted in a more rapid drop in pH.

Several studies have shown that carcasses with thicker fat cover cools down slower compared to thinner fat cover (Smith et al. 1976; Aalhus et al. 2001; Djimsa et al. 2022). There also seems to be a positive correlation between carcass weight and carcass conformation, meaning that a higher weight of the carcass also is graded for a higher conformation score (Kause et al. 2015).

The carcass conformation, weight and fat might vary depending on the sex. Bulls are generally heavier than heifers and steers, and heifers have normally a thicker fat cover than bulls and steers (Mueller et al. 2019; Pogorzelska-Przybyłek et al. 2021). In the study by Pogorzelska-Przybyłek et al. (2021), bulls, heifers and steers, all 18 months old at slaughter, were compared regarding slaughter weight, conformation score and fat score. The bulls (508 kg) had significantly higher weight than heifers (460 kg) and steers (464 kg), but there was no significant difference regarding the conformation score. The conformation score was 7.3, 7.2 and 7.4 for bulls, steers and heifers, respectively, and were graded according to EUROP grading system. The heifers (7.7) had significantly higher fat score in comparison to the bulls (4.8) and steers (5.0), using a 15-grading scale. Mueller et al. (2019) made a comparison on 13.5-month-old bulls, heifers and steers. The

bulls had a hot carcass weight of 319 kg and 7.56 cm fat thickness, heifers had a weight of 275 kg and 16.50 cm fat thickness, and the steers had a weight of 274 kg and fat thickness of 11.99 cm. This showed that the heifers had the thickest fat cover compared to bulls and steers (Mueller et al. 2019), which is in agreement with Pogorzelska-Przybyłek et al. (2021). Conroy et al. (2009) made a study to predict fat, bone and meat proportions on bulls, steers and heifers from the carcass conformation and fat content information. The bulls had a pre-slaughter weight of 583 kg and an age of 15 months, the heifers had a weight of 535 kg and 20 months of age, and the steers a weight of 625 kg and an age of 24 months. The bulls had a carcass conformation of 9.8, while heifers and steers had 8.4 and 6.8, respectively, using the EUROP-grading system. The fat score was 7.8, 7.6 and 8.5, respectively for bulls, heifers and steers, using a 15-grade scale. Differences between the sexes were not in focus in this study and had not been made.

2.3.5 Electrical stimulation

Adeyemi and Sazili (2014) explained in their review that electrical stimulation (ES) is used to deplete the glycogen in the carcasses faster, resulting in a more rapid decline of pH. *Rigor mortis* will occur faster and the risk for cold shortening will be decreased with the use of ES. The authors explained that ES often is used to improve the quality of the meat, like tenderness. The effectiveness of ES will depend on the muscle, like the placement of the muscle and its´ properties, but the voltage (V) and duration of the ES is also important (Adeyemi & Sazili 2014). Adeyemi and Sazili (2014) explained that there are three types of ES regarding voltage, which is extra low-voltage ES (<100 V), low-voltage ES (100-110 V) and high-voltage ES (>110V). But the authors also explained that the literature normally only use high and low when explaining the voltage. Low voltage ES is usually used within 20 minutes postmortem, while the high voltage can be used within 60 minutes. Except the different voltage, there will be differences in ampere (A), duration, frequency (Hz) and position of the electrodes for example. The type of animal, costs, the existing slaughter system, are all factors that will influence the ideal method at a specific abattoir (Adeyemi & Sazili 2014).

Effect of different types of voltage

Regarding the voltage, a study by Eikelenboom et al. (1985) compared high voltage ES, low voltage ES and no ES in both *longissimus* muscle and *adductor* muscle of 1.5-year-old bulls. The high voltage ES used electrodes in neck and tail region with pulses of electrical voltage for 1.5 minutes (300 V, 50 Hz) and the low voltage ES were placed through nostrils and was continuous for 1 minute (85 V, 14 Hz). Muscle temperature and pH were measured 1, 2, 4, 6, 8 and 24 hours postmortem. The authors concluded that there was no difference in pH decline

between the high and low voltage treatments, but there were a more rapid decline using ES compared to not using ES. In the *longissimus* muscle one hour postmortem, when using high voltage ES, the pH was 6.02 and muscle temperature 38.1 °C, while the control group had a pH of 7.08 and a muscle temperature of 37.1°C (Eikelenboom et al. 1985). The colour of the meat got a brighter red colour when ES were used, but in general there were no difference between high and low voltage regarding meat characteristics like tenderness. (Eikelenboom et al. 1985). A comparison between high and low voltage were also made by Aalhus et al. (1994) on steers and heifers in two experiments, where a control group and a group treated with both low and high voltage were included. In the first experiment, low voltage ES (21 V, 60 Hz, 0.25 A, 20 seconds) were applied five minutes postmortem in the nose, while the high voltage ES was applied 40 minutes postmortem with a probe in the nuchal ligament (in the neck), after the carcasses had been split in two. Muscle temperature and pH were measured between 12^{th} and 13^{th} rib at a dept of 3 centimetres before and after high voltage stimulation, before cooling, and at three and 24 hours postmortem. Two different chilling methods were used; one conventional chilling (2 °C for 24 hours) and one blast chiller method where the carcasses were blast chilled for 2 hours (-20 °C) before entering the other cooler of 2 °C. The second experiment had the same principle but had a longer duration of the low voltage ES, 40 seconds compared to 20 seconds in the first experiment, and the blast chiller had a temperature of -40 °C instead of -20 °C. The authors found no differences between steers and heifers, and no differences in pH_{24} in both experiments. In the first experiment there were differences in pH between low and high voltage ES at the time before the high voltage ES had been applied, where the low voltage group had a pH of 6.30 and the high voltage group had 6.61. There was also a difference in pH at three hours postmortem where the low voltage group had a pH of 5.72 and the high voltage group had 5.48. In the second experiment there were differences in pH before and after the application of high voltage ES, but also at three hours postmortem. Before the high voltage ES were applied the low voltage group had a pH of 6.10 and the high voltage group 6.77. The authors explained that the more rapid pH-decline in the low voltage group in the second experiment compared to the first, are due to heavier and fatter carcasses in the second experiment, which results in higher temperatures in the muscles which increases the rate of the glycolysis and pH-decline. Another reason for the differences is due to the longer application of ES. In the second experiment after the application of high voltage ES, the high voltage group had a pH of 6.42 and the low voltage group still had 6.10. At three hours postmortem the low voltage group had a pH of 5.61 and the high voltage group 5.88. The muscle temperature decline did not differ between the high and low voltage group in experiment 1, but differences were seen in experiment two where the high voltage group had lower muscle

temperature than the low voltage group before (40.0 °C vs 40.6 °C) and after (40.2 °C vs 40.6 °C) application of high voltage ES. Aalhus et al. (1994) compared the results to Eikelenboom et al. (1985) who found the opposite result, that the voltage did not have an effect on the pH-decline. Aalhus et al. (1994) explained that these differences could be caused by differences in time of application of ES. Interestingly, the authors found PSE conditions in some (9 of 60) of the carcasses in experiment two, with five cases in the group treated with both low and high voltage and two cases in the low voltage group. Low voltage ES were also found to cause PSE conditions in a study by Stiffler et al. (1984) in the *longissimus* muscle on steers, which were explained as due to excessive stimulation.

Effect on temperature when pH 6 occur

Several studies show that the pH declined faster when ES were used (Koh et al. 1987; Agbeniga & Webb 2014; Biraima et al. 2019), but ES did not have an effect on the muscle temperature (Agbeniga & Webb 2014; Biraima et al. 2019). Hopkins et al. (2014) investigated if electrical stimulation had an effect on the muscle temperature when pH 6 occurred in beef steer (322 kg) carcasses with high temperatures in *M. longissimus thoracis et lumborum*. The ES had a duration of 35 seconds (15 Hz) using continuous stimulation with variable voltage. Authors found that carcasses that were electrical stimulated had a mean muscle temperature of 40.9 °C when pH 6 was reached, whereas the non-stimulated carcasses had a mean muscle temperature of 33.3°C. The carcasses that got the electrical stimulation all missed the ideal pH- and temperature window (15-35 °C), according to MSA recommendations, while several of the non-stimulated carcasses also did. Elgasim et al. (1981) made the same findings, that carcasses that were electrically stimulated had a higher muscle temperature when reaching pH 6. The study by Elgasim et al. (1981) included beef cattle (459-510 kg). One side of each carcass were electrical stimulated (600 V, 7 A and 60 Hz) and the other were not electrical stimulated. The study included two different chilling methods, one immediate, meaning that the carcasses were in a cooler of 2 °C for seven days, and the delayed chilling method included twelve hours in 16 °C until entering 2 °C cooler. There was no difference in pH_{24} between treatments. In the immediate chiller, the stimulated carcasses reached pH 6 within three hours and had at that time a muscle temperature of 26.5 °C, while the unstimulated carcasses reached pH 6 after twelve hours and had a muscle temperature of 5 °C. The stimulated carcasses using the delayed method reached pH 6 after two hours and had a muscle temperature of 34.6 °C. The unstimulated carcasses with delayed cooling reached pH 6 after six hours and had a muscle temperature of 24 °C (Elgasim et al. 1981).

2.4 Time frames during the slaughter process

2.4.1 Stunning to chilling

According to Australian standard for the hygienic production and transportation of meat and meat products for human consumption (Chapter 11, section 3), the carcasses need to enter the cooling system within two hours after stunning. European Food safety authority (EFSA) explained that the carcasses need to enter the coolers as soon as possible after slaughter and dressing to minimize growth of bacteria to ensure food safety (Hazards (BIOHAZ) 2014).

In practice the time from stunning until entering the coolers varies. Djimsa et al. (2022) reported a time of 30 to 45 minutes postmortem until entering the cooler in one abattoir and 60 to 75 minutes in another. Hultgren et al. (2022) observed a mean time of just under 60 minutes in the stationary abattoir in their study, while Stevenson et al. (1978) had a dressing time documented of 60 minutes, but no clarification of the time after dressing until reaching the cooler is made.

2.4.2 Time postmortem to pH 6

The time it takes postmortem for the muscle to reach pH 6 can reflect product quality. This measure must be interpreted alongside the temperature at which pH 6 occurs, as both parameters strongly influence the final product quality. In a study by O'Halloran et al. (1997) *M. longissimus dorsi* and *M. semimembranosus* were used to examine the pH decline postmortem in heifers. Authors had divided the carcasses depending on the rate of pH decline, where a fast drop in pH reached pH 6 two hours postmortem, the intermediate at four hours postmortem, and the slow at six hours postmortem. The authors did not find any differences in muscle temperature between the rates of pH-decline, and the muscle temperature when pH 6 occurred is not specified. Nunes et al. (2024) examined the temperature decline postmortem in *longissimus thoracis* muscle on 24-month-old bulls with a cold carcass weight of 296 kg. The study used two different chilling methods, one conventional which cooled the carcasses in 4 °C for 24 hours, and one dynamic chilling method were the carcasses entered coolers with a temperature of 9 °C, which decreased with 1.4 °C each hour the first four hours, and the rest of the time the cooler had a temperature of 3.3 °C. The conventional chilling method reached pH 6 after 2.5 hours postmortem with a muscle temperature of 25.4 °C, while the dynamic chilling method reached pH 6 after 5.8 hours postmortem with a muscle temperature of 14.4 °C (Nunes et al. 2024). In a study by Elgasim et al. (1981) two different chilling methods were compared in combination with use of ES (600 V, 7 A, 60 Hz, 1 min), regarding pH- and temperature decline in *longissimus* muscle in beef cattle (459-510 kg). The immediate chilling refers to cooling the carcasses in 2 °C for seven days, while the delayed meant that the

carcasses entered a cooler with an initial temperature of 16 °C for twelve hours and then entered the cooler of 2 °C for the rest of the time. With use of ES the immediate chilling method reached pH 6 after three hours and had at that time a muscle temperature of 26.5 °C, while the delayed chilling reached pH 6 two hours postmortem with a muscle temperature of 34.6 °C. The non-stimulated carcasses with immediate chilling reached pH 6 at a muscle temperature below 10 °C approximately twelve hours postmortem, while the non-stimulated carcasses with delayed chilling reached pH 6 at six hours postmortem with a muscle temperature around 25 °C (Elgasim et al. 1981). The different studies shows that different chilling regimes and use of ES influence time until pH 6 is reached and therefore differences in temperatures also varies (Elgasim et al. 1981; Nunes et al. 2024).

3. Materials and methods

3.1 Experimental design

3.1.1 Animals and slaughter

The study included 45 beef animals that was slaughtered at two different occasions at an abattoir in Sweden. The animals were randomly selected by the staff at the abattoir, depending on the animals that were supposed to be slaughtered that day in the commercial practice. The requirements for the choice of animals were to have different groups of animals to be able to compare within and between different types of animal categories, to be able to include different type of carcasses when studying the slaughter system. The animals included were of different characteristics like age, sex, breed, conformation and lairage time. The first slaughter occurred December 17, 2025, and included a group of 16 bulls (18 months) that had stayed overnight at the abattoir, and a group of four heifers (41 months) transported to the abattoir the same day as the slaughter. The second slaughter occurred February 11, 2026, and included a group of twelve animals, five bulls (21 months) and seven heifers (25 months) that had stayed overnight, and a group of 13 dairy cows (74 months) transported to the abattoir the same day. The animals were stunned with a pneumatic bolt gun. At the time for exsanguination, blood was collected in cups, 120 ml, to measure blood lactate, with a portable measuring meter (Lactate Plus, Nova Biomedical, Germany). The animals were hanged in the Achilles tendon and half of the carcasses, every second, were electrical stimulated (41 V, 31 A, 300 Hz). Electrical stimulation was applied in the corners of the mouth approximately 20 to 25 minutes postmortem. The time from exsanguination until classification were on average one hour and four minutes (see table 1) and then, after classification, the carcasses entered the blast chiller. The carcasses were in the blast chiller for approximately two hours and temperature in the chiller were 1 °C. After two hours, carcasses entered the second chiller, that had a temperature of 7 °C, where they stayed until 24 hours postmortem.

Table 1. Time frames from exsanguination until pH 6, exsanguination until classification and classification until pH 6.

Parameter	Time		
	Min	Mean	Max
Exsanguination → pH_6	1h 4 min	1h 58 min	5h 59 min
Exsanguination → classification[1]	48 min	1h 4 min	1h 12 min
Classification[1] → pH_6	4 min	54 min	4h 50 min

[1]*Classification is equivalent with the step at the slaughter line where carcasses get their conformation and fatness scores.*

3.2 Temperature and pH measurements

Before and between slaughter occasions, temperature and pH-meters (Mettler Toledo, Seven2Go Pro S8, Schweiz) were calibrated with buffer solution with pH 4 and 7. The probes were also cleaned with pepsin- and ethanol solutions to ensure that there were no remains of protein and fat left on the probes. Probes were placed in tubes with potassium chloride storage solution (KCI 3 mol/l) between measurement occasions. The pH-meter were placed in a plastic bag with a rubber band at the top to minimize contamination of the measuring tools. Cable ties were also placed at one end of each pH-meter, as preparation for easy attachment of the measurement tools to the carcasses. At the abattoir probes were placed in the topside muscle (*M. semimembranosus*) on the internal surface of the carcass within the pelvic cavity, from a ventral angle, see Picture 1. The probes were placed in the left part of the carcass and the temperature- and pH-meter registered values every tenth minutes for 24 hours. All equipment was inserted before each carcass entered the blast chiller, approximately 1 hour postmortem. After 24 hours when the carcass halves were parted into quarters pH-meters were taken out of each muscle. Due to problems with the equipment, 44 out of 45 meters showed correct values for muscle temperature, leading to one of them having a missing value for muscle temperature at pH 6 and muscle temperature 24 hours postmortem.

Picture 1. Placement of the probe in M. Semimembranosus in the carcass

3.3 Analysis of data

The lactate values were achieved immediately with the portable measuring meter (Lactate Plus, Nova Biomedical, Germany) and were written by hand on a paper. Slaughter weight, conformation score and fat score was achieved through slaughter labels from classification. The data from pH- and temperature-meters were transformed to EasyDirect pH program and were manually analysed and values for pH_{24}, muscle temperature at pH 6 and muscle temperature 24 hours postmortem were extracted.

3.4 Statistical analysis

The results were statistically analysed with Statistical Analysis Software (SAS) using Proc Mixed Models, where two different models were used.

For the effect of sex and electrical stimulation the following model was used:

Model: $Y_{ijk} = \mu + S_i + E_j + SE_{ij} + D_k + e_{ijkl}$

For the effect of lairage time at the abattoir, a hierarchical model was used. The model is nested because sex is not comparable between levels, due to unbalanced data. The following model was used:

$Y_{ijk} = \mu + L_i + S_{j\,(i)} + D_k + e_{ijkl}$

where Y_{ijk} is the dependent variable, μ is the grand mean, S_i is the fixed factor of sex of the animal, E_j is the fixed factor of electrical stimulation or not, SE_{ij} is the interaction between sex and electrical stimulation, D_k is the random effect of slaughter date, L_i is the fixed factor of lairage time, $S_{j\,(i)}$ is the fixed factor of sex nested with lairage time, and e_{ijkl} is the residual error. A general Satterthwaite approximation for the denominator degrees of freedom was performed, using the SATTERTH option in SAS. Statistical significance was defined as $p \leq 0.05$, and a tendency to significance $0.05<p \leq 0.10$.

4. Results

Sex had an effect on blood lactate concentration ($p=0.0002$), where the bulls (8.4 mmol/L) had higher blood lactate concentrations in comparison to heifers (5.7 mmol/L) and cows (4.0 mmol/L) (see Table 2). Sex also had an effect on conformation score ($p<0.0001$) and muscle temperature at 24 hours postmortem ($p=0.0417$). Cows (3.41/P+) had lower conformation score compared to heifers (7.33/R-) and bulls (7.98/R). Regarding the muscle temperature 24 hours postmortem the cows (12.7 ºC) had slightly higher muscle temperatures compared to the heifers (11.4 ºC) and bulls (11.1 ºC). There was a tendency to significance for fat score ($p=0.0866$), where cows (6.37/2+) had slightly lower score than heifers (7.93/3). However, sex had no effect on slaughter weight, pH_{24} and muscle temperature at pH 6. The electrical stimulation did not have an effect on any of the parameters tested. Interactive effect was not found for sex and electrical stimulation and are therefore not included in Table 2.

Table 2. Results on blood lactate concentration, slaughter weight, conformation score, fat score, pH_{24}, $temp_{24}$ and temperature at pH_6 for sex (cows, heifers and bulls) and use of electrical stimulation (yes/no).

	Sex			**Electrical stimulation (ES)[1]**				
							Significance	
Parameter	Cows	Heifers	Bulls	Yes	No	SEM[2]	Sex	ES
Blood lactate (mmol/L)[3]	4.0[a]	5.7[a]	8.4[b]	5.8	6.3	0.68	0.0002	0.5579
Slaughter weight (kg)	333.2	312.5	343.6	320.8	338.7	13.61	0.3263	0.2633
Conformation score[4]	3.41[a]	7.33[b]	7.98[b]	6.31	6.17	0.68	<.0001	0.7467
Fat score[5]	6.37	7.93	7.04	6.82	7.41	1.47	0.0866	0.2458
pH_{24}[6]	5.48	5.48	5.47	5.47	5.48	0.07	0.9947	0.8346
$Temp_{24}$ (ºC)[7]	12.7[a]	11.4[b]	11.1[b]	11.5	12.0	1.21	0.0417	0.3010
Temp@pH_6 (ºC)[8]	37.0	37.5	37.1	37.2	37.2	2.86	0.9608	0.9498

[1]Electrical stimulation treatment (41 V, 31 A, 300 Hz), yes or no; [2]SEM = standard error of the mean; [3]Blood lactate measured in exsanguination blood; [4]Conformation score (1-15), where 1 (P-) is the lowest score and 15 (E+) the highest score; [5]Fat score (1-15), where 1 (1-) is the lowest score and 15 (5+) the highest score;[6]pH_{24} = pH measured 24 hours postmortem; [7]$Temp_{24}$ = muscle temperature measured 24 hours postmortem; [8]Temp@pH_6 = the muscle temperature when pH 6 occurs; [a-b]Mean values within rows with different superscripts differ significantly ($p< 0.05$) or show a tendency for significance at $0.05<p\leq 0.10$.

A positive correlation (0.64) was found for fat score and muscle temperature, showing that a higher fat score resulting in a higher muscle temperature in the carcass, see figure 1.

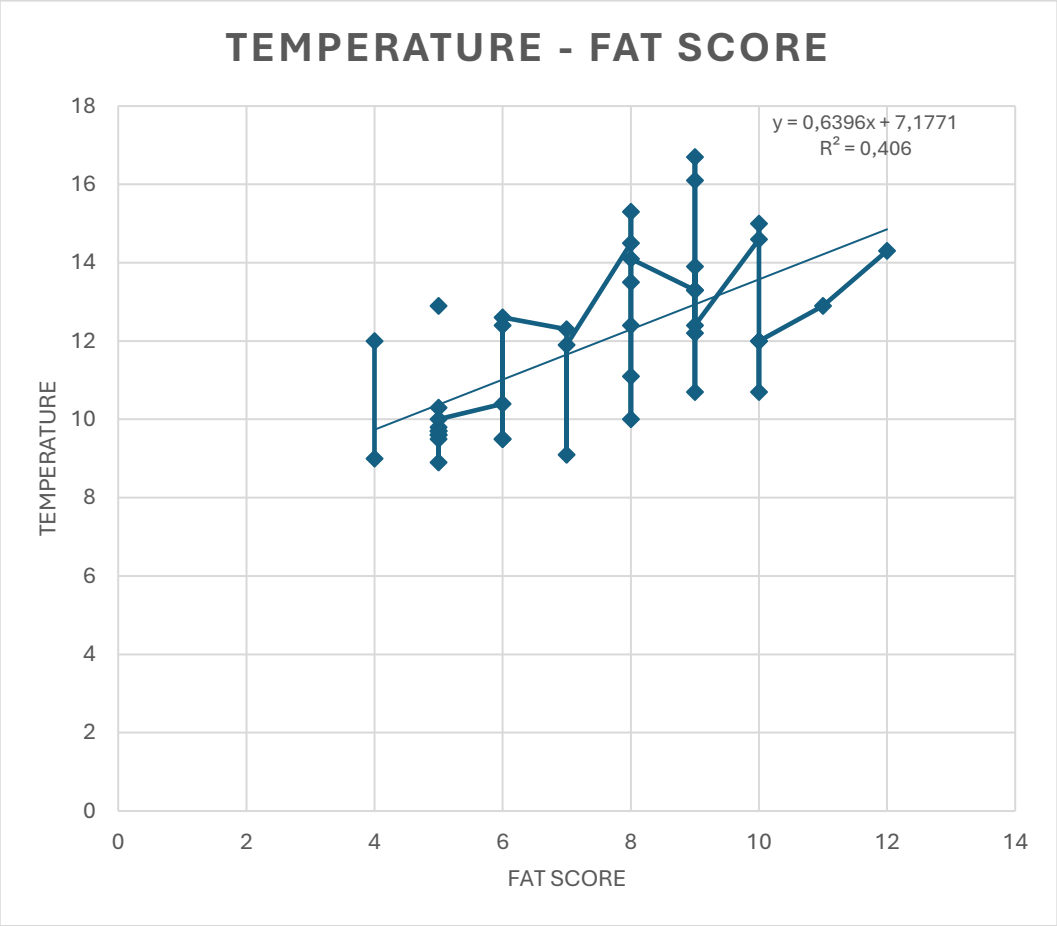

Figure 1. Correlation between muscle temperature and fat score in beef carcasses, based on all carcasses included in the study (bulls, cows and heifers).

The lairage time, if the animals had stayed overnight at the abattoir or not had an effect on blood lactate, where the group that had stayed overnight had higher blood lactate concentrations (see Table 3). There were no differences between lairage time (yes/no) regarding pH_{24}, muscle temperature 24 hours postmortem and muscle temperature at pH 6.

Table 3. Blood lactate concentration, pH_{24}, $temp_{24}$ and temperature at pH_6 for lairage time/overnight (yes/no)

	Lairage time/overnight[1]			
Parameter	Yes	No	SEM[2]	Significance
Blood Lactate (mmol/L)[3]	7.2	4.4	0.69	0.0070
pH_{24}[4]	5.50	5.45	0.10	0.3849
$Temp_{24}$ (ºC)[5]	11.2	12.1	1.30	0.1018
Temp@pH_6 (ºC)[6]	37.0	37.7	3.47	0.6353

[1]Lairage time = if the animals stays overnight or not (yes/no); [2]SEM = standard error of the mean; [3]Blood lactate measured in exsanguination blood; [4]pH_{24} = pH measured 24 hours postmortem; [5]$Temp_{24}$ = muscle temperature measured 24 hours postmortem; [6]Temp@pH_6 = the muscle temperature when pH 6 occurs; Differences are considered significant at $p < 0.05$ and a tendency to significance $0.05 < p \leq 0.10$.

5. Discussion

5.1 Lactate and its´ effect on pH decline

In the present study bulls (8.4 mmol/L) had significantly higher blood lactate concentrations compared to both heifers (5.7 mmol/L) and dairy cows (4.0 mmol/L). In previous studies, lactate concentrations were reported for all animal categories combined, with no sex specific values provided, which prevents comparisons of sex differences between earlier studies and the present study. Coombes et al. (2014) explained that the temperament could have an effect on blood lactate concentrations, which is due to hormones and that the bulls are in a state to show dominance resulting in stress and anxiety (Muižniece & Kairiša 2020). The increased stress due to temperament and hormones might result in elevated concentration of blood lactate which might be one cause for the higher values for bulls compared to heifers and cows in the present study. There were also variations within the sexes in the present study where the bulls ranged from 3.9 to 13.6 mmol/L, heifers from 2.9 to 13.7 mmol/L, and cows from 1.9 to 7.8 mmol/L. The variation in lactate concentration can be caused by several factors, like feeding strategies pre-mortem (Immonen et al. 2000; Coombes et al. 2014). A high energy diet (12.9 MJ ME) resulted in a larger storage of glycogen pre-mortem, but also smaller losses during transport compared to a low energy diet (10.8 MJ ME) (Immonen et al. 2000). The authors explained that it might occurred due to adequate amounts of gluconeogenic precursors with the high energy diet which served as a protection to stress. In the present study, there were no information about the feeding regimes for the different animals included, meaning that feed might be one of the causes for the variations within sex. Hultgren et al. (2022) explained that another stress factor for animals can be contact with unfamiliar animals and humans which happens during transport and at the abattoir. Probst et al. (2013) found that animals that had been used to human contact had shorter avoidance distance to unfamiliar humans and also had lower blood lactate concentrations. In the present study, dairy cows had the lowest blood lactate concentrations and are probably most used to being handled by humans due to milking, compared to beef heifers and bulls. Animals that are more used to humans might experience less stress to unfamiliar humans at the abattoir, which might be the cause for the lower values in dairy cows in the present study. Use of electric prodders was investigated by Warner et al. (2007) and how that affected the blood lactate concentrations post-slaughter. Warner et al. (2007) found that the animals that had been moved with electric prodders (7.1 mmol/L) had higher blood lactate concentration compared to animals that moved without them (4.3 mmol/L), which might also be a factor for differences in the present study, depending on which animals that have been treated with prodders compared to

without. Sullivan et al. (2024) explained that there might be a seasonal effect in lactate concentrations, with higher concentrations during summer due to heat stress, but that effect cannot be included in the present study due to that the slaughter occasions only occurred during the winter season. It is also important to remember that all animals react different to stressors, which also might be a cause for the variation.

Despite these differences in lactate concentrations among and within sexes, no differences were seen regarding the pH-decline during the first 24 hours and therefore does not seem to affect the pH-decline in the present study. But it is important to remember that stress before slaughter like handling of the animals at the abattoir can cause an increased risk for PSE (Muižniece & Kairiša 2020). Stress before slaughter could therefore be a part of several different factors that influenced the fast pH-decline after slaughter, and it would be interesting to investigate if the design of the abattoir where live animals are handled might have had an effect on lactate concentrations.

5.2 pH and temperature decline

The recommended interval for muscle temperature when reaching pH 6 is according to MSA 15 to 35 °C (Meat & Livestock Australia n.d.). In the present study 44 out of 45 animals had recorded values for muscle temperature at pH 6, and the mean muscle temperature were 37.6 °C. Only six of the included animals were, according to MSA, within the recommended interval for pH 6, while the rest of the animals (38) exceeded 35 °C which means that there is a risk for heat shortening or PSE. The lowest value for pH_{24} among all animals included in the study were 5.17 and the highest 5.86, with a mean of 5.48. As previously described, the recommended values for pH_{24} varies among different countries, where the different recommendations range from 5.3 to 5.9 (Muižniece & Kairiša 2020; Brazil Beef Quality n.d.; Meat & Livestock Australia n.d.; Meat Standard Sweden nöt n.d.). There is only one value (5.17) in the present study that is outside of the recommended range (5.3-5.9), but in general pH_{24} is in an acceptable range according to the different standards and recommendations.

5.2.1 Effect of sex

The pH and temperature decline in muscle can be affected by several different factors related to animal material and experiences pre-mortem. In the present study no differences were seen in pH_{24} between bulls, cows and heifers which might had been expected due to previous studies. The sex has in several studies shown to have an effect on the ultimate pH, where bulls have a higher ultimate pH compared to heifers, cows and steers (Page et al. 2001; Węglarz 2010; Marenčić et al. 2012; Muižniece & Kairiša 2020), which is explained by a lower stress

tolerance in bulls due to temperament and hormones. The increased stress seen in bulls might result in that the glycogen storage is used before slaughter, which means that there is a smaller amount that can be used to produce lactic acid postmortem, which result in higher pH and an increased risk for DFD in bulls (Coombes et al. 2014). However, this was not found in the present study.

5.2.2 Effect of age

Animal age has in some studies influenced the ultimate pH, while other have found the opposite (Arik & Karaca 2017; Muižniece & Kairiša 2020; Liza et al. 2024). In the present study the ages have not been included, but it might still be important to consider animal ages when analysing impact of the slaughter system. The average age in the present study were 19 months for bulls, 30 months for heifers and 74 months for cows. Muižniece and Kairiša (2020) did not find any differences in ultimate pH in bulls of different ages, but older heifers (24> months) had higher ultimate pH than younger heifers (12-17, 18-23 months). The older heifers had an ultimate pH of 5.69, which is higher than the pH_{24} for the heifers in the present study (5.48) in the same age group. Arik and Karaca (2017), on the other hand, found that older bulls (30> months) had higher ultimate pH than younger (<30 months). The bulls in the present study had a pH_{24} of 5.47 and can be compared to the younger bulls in the study by Arik and Karaca (2017) which had a much higher ultimate pH (5.76). The differences in pH between the present study and to the previous studies can be due to several factors, like differences in animal material (breed, weight, fat, feeding regimes etc,), but also differences in the slaughter system (stable design, ES etc.).

5.2.3 Effect of season

The slaughter occasions in the present study only occurred during the winter season, which might be a disadvantage of the study. The variation due to season could be of interest to look more into due to findings by Węglarz (2010) and Marenčić et al. (2012), who found that pH varied depending on season. Węglarz (2010) found that both younger (<24 months) and older (> 24 months) bulls had higher pH_{48} during summer than winter, where younger bulls had 6.10 during summer and 5.94 during winter. Older bulls had 6.07 during summer and 5.65 during winter, but heifers and cows did not differ in pH due to season, which was explained by bulls' higher sensitivity to stressors, like heat stress. Marenčić et al. (2012), made the same findings, that the bulls had higher pH during summer (5.60) due to higher environmental temperatures, but also during winter (5.60), due to the colder temperatures, compared to spring (5.56) and autumn (5.58). In the study by Marenčić et al. (2012) a higher pH was also observed in heifers during summer (5.61) compared to other seasons (5.55), also depending on heat stress.

5.2.4 Effect of lairage time

Lairage before slaughter is one discussed pre-slaughter factor that might contribute to fast pH-decline postmortem. In the present study the lairage time (overnight or not) had a significant effect on blood lactate concentration, where the animals that had stayed overnight (7.2 mmol/L) had significantly higher blood lactate concentration compared to animals slaughter the same day (4.4 mmol/L). One explanation for this could be that mainly bulls stayed overnight at the abattoir and bulls had significantly higher blood lactate concentrations compared to heifers and cows, which can be seen in table 2. To evaluate lairage overnight compared to slaughter on the same day a larger number of animals needs to be included in the study do eliminate differences due to differences in animal material between the groups. No differences were seen regarding the ultimate pH (pH_{24}) for different lairage time, which is similar to findings by Ferguson et al. (2007) and Costa et al. (2019). But in the study by del Campo et al (2010) on the other hand, a difference in ultimate pH were found when comparing three hours versus 15 hours lairage time on steers, where the shorter lairage time (5.82) resulted in higher pH compared to the longer (5.56). The difference can be due to that the longer lairage time gave animals the opportunity to restock their glycogen storage or that they had time to recover from the stress. However, del Campo et al. (2010) also explained that the differences might not be caused by the lairage time itself but could be due to other factors like animal material, transport time and design of abattoir, which means that it is difficult to draw conclusions for just the effect of lairage time.

5.2.5 Effect of slaughter weight, fatness and conformation

Temperature decline in carcasses and muscles is affected by slaughter weight, carcass conformation and fatness of the carcass. Animals within the present study had carcass weights ranging from 177 kg to 530 kg, with a mean weight of 332 kg. Comparisons have not been made in the present study for different slaughter weights, but both lighter and heavier carcasses reached pH 6 at muscle temperatures that exceeded MSA: s recommendation (15-35 °C), and no other patterns have been found regarding the muscle temperature and slaughter weight. It is well known that heavier carcasses have a slower temperature decline than leaner carcasses (Lochner et al. 1980; Lancaster et al. 2020; Djimsa et al. 2022). Djimsa et al. (2022) measured the temperature decline in *semimembranosus* muscle, *longissimus lumborum* muscle and in *psoas major* muscle after slaughter and found the same result for all muscles, that the temperature declined faster in lighter (355-360kg) carcasses compared to heavier (420-445kg). However, Djimsa et al. (2022) also found that the temperature decline was slower in the *semimembranosus* muscle compared to the other two muscles, which is explained by the location of the muscle, which is in the heaviest and thickest part of the

carcass, often with a thicker fat cover. Djimsa et al. (2022) and Lancaster et al. (2020), found that pH declined faster in heavier carcasses. Jacob and Hopkins (2014) explained that heavier carcasses often have higher muscle temperatures and that increases the glycolysis, which results in a rapid pH-decline. In the present study, the *semimembranosus* muscle had a mean temperature of 11.9 °C at 24 hours postmortem when the carcasses had a mean weight of 332 kg, for all animals included in the study. This is interesting in comparison to the study by Djimsa et al. (2022) who found that the lighter carcasses (355-360) had reached a muscle temperature of 10 °C in *semimembranosus* muscle by 24 hours postmortem. That showed that even though the carcasses were lighter in the present study, the muscle temperature at 24 hours postmortem were still higher in comparison to the result by Djimsa et al. (2022). The differences in muscle temperature can for example be due to different chilling regimes or fat score, as higher fat score is positively correlated with higher muscle temperature, as seen in the present study (see Figure 1.). The fact the carcass size has an important interaction with the temperature decline, is necessary to consider when evaluating meat quality and cooling condition. The optimal cooling conditions will probably vary depending on the size of the carcass, which is important to consider in the present study as the result showed that several carcasses had high muscle temperatures, and therefore probably not have had optimal cooling conditions.

The weight, conformation score and fat score also vary depending on the sex. Regarding the weight, no significant differences were found in the present study, even though the bulls (343 kg) had the highest average slaughter weight compared to heifers (312 kg) and cows (333 kg). Pogorzelska-Przybyłek et al. (2021) and Mueller et al. (2019) explained that bulls generally have a higher slaughter weight due to a faster growth rate, and a higher weight often results in a higher conformation score (Kause et al. 2015). In the present study the conformation score was significantly lower for cows (3.41) compared to bulls (7.98) and heifers (7.33). The conformation score in the study by Pogorzelska-Przybyłek et al. (2021) were 7.3 for bulls, 7.4 for heifers and 7.2 for steers, while Conroy et al. (2009) had a conformation score of 9.8 for bulls, 8.4 for heifers and 6.8 for steers. These values do not vary so much from the scores seen in the present study for bulls and heifers.

A thicker fat cover often results in slower temperature decline (Smith et al. 1976; Aalhus et al. 2001; Djimsa et al. 2022), which were also found in the present study (see Figure 1). There was a tendency to significance regarding fat score in the present study where cows (6.37) had slightly lower fat score than heifers (7.93). Bulls in the present study had a fat score of 7.04. In the study by Pogorzelska-Przybyłek et al. (2021), heifers (7.7) had significantly higher fat score compared to bulls (4.8) and steers (5.0). The same pattern was made by

Mueller et al. (2019) who compared fat thickness where the heifers (16.5 cm) had the thickest fat cover compared to bulls (7.56 cm) and steers (11.99 cm). In the study by Conroy et al. (2009) the bulls, heifers and steers had a fat score of 7.8, 7.6 and 8.5 respectively. The fat score for heifers in the present study is similar to the findings by Conroy et al. (2009) and Pogorzelska-Przybyłek et al. (2021), but Pogorzelska-Przybyłek et al. (2021) had lower fat score for bulls compared to the present study and to the study by Conroy et al. (2009). These differences are probably due to differences in animal material like breed and age. The cows in the present study also significantly differed from the bulls and heifers regarding muscle temperature at 24 hours postmortem, where the cows had a muscle temperature of 12.7 °C and bulls and heifers had 11.1 °C and 11.4 °C, respectively. This finding is interesting; due to that there were no differences for slaughter weight and that cows had a slightly lower fat score compared to heifers. As previously described by previous research and in the present study (figure 1), a higher fat score is positively correlated with muscle temperature and therefore, it would have been more expected to find higher temperatures in heifers compared to cows due to a tendency for higher fat score. The explanation for this finding is unclear.

5.3 Effect of electrical stimulation

Electrical stimulation is commonly used during beef slaughter, with large variations in voltage, ampere, duration and placement of electrodes for example. In the present study, low voltage ES (41 V, 31 A and 300 Hz) was applied on half of the carcasses included in the study, but no differences were seen regarding pH_{24} which agrees with the findings by Eikelenboom et al. (1985) and Aalhus et al. (1994). The most interesting finding in the study by Aalhus et al. (1994) were the differences in settings of the low voltage ES between two experiments and how that affected the pH-decline. The settings only differed in duration of application, where the first experiment had a duration of 20 seconds while the second experiment had a duration of 40 seconds. The second experiment had a more rapid pH decline compared to the first, which probably were caused by the longer duration, but also in combination with heavier carcasses with higher muscle temperature that increased the glycolysis. But the authors also found that some carcasses (9 of 60) had PSE-like conditions, where two of them had been treated with low voltage ES, and the rest (seven) with a combination of high and low voltage ES. Stiffler et al. (1984) also found carcasses with PSE-like conditions when low voltage ES were used and explained that it probably was due to excessive stimulation. This finding is interesting in comparison to the present study, that the duration of ES might contribute to the PSE-like conditions (pale and watery meat) that have been experienced previously. However, there were no differences regarding ES for any of the parameters included in the study, which

might had been expected, due to that several studies have found that the use of ES leads to a more rapid pH-decline (Elgasim et al. 1981; Eikelenboom et al. 1985; Koh et al. 1987; Adeyemi & Sazili 2014; Agbeniga & Webb 2014; Biraima et al. 2019), this difference was however not observed in the present study between the ES and no ES groups. The rapid pH-decline that occur when ES is used was explained by Adeyemi and Sazili (2014) as due to a more rapid depletion of glycogen. The pH 6 occur faster and the muscle temperature is often higher when reaching pH 6 when ES is used (Elgasim et al. 1981; Eikelenboom et al. 1985; Hopkins et al. 2014). It would have been interesting to evaluate meat colour differences in the present study to see if ES would have resulted in colour differences, as Eikelenboom et al. (1985) found that the use of ES resulted in brighter red colour of the final product.

5.4 Possible quality problem and factors that might optimize the slaughter process

The main problem in the present study is that the pH-decline is rapid while the muscle temperature is still high, which makes it possible that PSE or heat shortening have occurred. The quality attributes of PSE are pale, soft and exudative meat (Van Laack et al. 2000; Woelfel et al. 2002; Barbut et al. 2005; Muižniece & Kairiša 2020) and for heat shortening according to MSA, a paler colour, tougher meat and increase drip loss is common.

In the present study, pH 6 were reached at an average time of 1 hour and 58 minutes, with a mean muscle temperature of approximately 37 °C, for all animals included in the study. In the study by Aalhus et al. (1998) on PSE beef, the pH 6 is reached at a high temperature in the PSE carcasses. At 45 minutes postmortem the PSE carcasses had a pH of 5.98 and a temperature of 41.1 °C, while the normal carcasses had a pH of 6.79 and a temperature of 39.8 °C. Interestingly, Aalhus et al. (1998) explained that the occurrence of PSE in beef is not common, but if it occurs it is often due to slow chilling conditions. Slow chilling conditions occur more often in *psoas major*, *semimembranosus* muscle and in *adductor* muscle, which is interesting in comparison to the present study, as *semimembranosus* muscle is the muscle that has these PSE-like conditions, and the explanation for the problems can therefore partly be caused by characteristics of the specific muscle. As described the pH-decline is very rapid in the present study meaning that the time from exsanguination until the carcasses reaches the cooler is a critical point. If the time is shortened, there is a chance that the carcasses reach pH 6 at lower muscle temperatures than they do today. The time it takes from stunning or exsanguination until reaching coolers varies among abattoirs and the design of the slaughter line. In the present study the average time from exsanguination until classification were 1 hour and 4 minutes, however the

exact time until carcasses enter the cooler is a bit longer in time since carcasses have to wait for some time before entering the cooler after the classification station. In the study by Djimsa et al. (2022) the time postmortem until reaching the cooler were in one abattoir 30 to 45 minutes and in another 60 to 75 minutes. In the study by Hultgren et al. (2022) the carcasses reached the coolers within 60 minutes postmortem, which shows a variation among abattoirs. But another factor that can contribute to a rapid pH-decline in muscle is the use of ES, which may result in reaching pH 6 at high muscle temperatures. Aalhus et al. (1994) compared high voltage ES, low voltage ES, combination of high and low voltage ES, and no ES, and found PSE-like conditions in 9 of 60 carcasses. Two of the carcasses with PSE-like condition were treated with low voltage ES and seven were treated with combination of high and low voltage ES. Stiffler et al. (1984) made the same finding that carcasses exposed to low voltage ES resulted in some carcasses with PSE-like condition, which were explained as due to excessive stimulation. Since low voltage ES is used at the abattoir in the present study, it would be of interest to investigate if other ES settings would decrease occurrence of meat quality problems. Another factor to consider or to evaluate further is the design of the stable where live animals are handled at the abattoir. As mentioned in section 5.1 stress pre-mortem can be a cause for PSE. When arriving to the abattoir the animals are experiencing several things that can be stressful. Hultgren et al. (2022) explained that the unloading of animals can be a cause to stress, but also the contact to both unfamiliar animals and humans. Sullivan et al. (2024) explained that the time spent on the truck before being unloaded contribute to stress, but also the space allowance at the abattoir and the time spent in the lairage area before slaughter. Therefore, it might be valuable to investigate if changes in the live animal areas would contribute to an optimization of the slaughter system by minimising all form of stressors that might occur. Another possible cause for the quality problem is heat-shortening due to that pH 6 is reached at a high muscle temperature, above 35 °C according to MSA. But the previous research describing different occurrence of heat-shortening or shortening in general, do not include any characteristics of the final product as the colour or drip loss in beef. But according to MSA regarding sheep meat, the quality in the final product when experiencing heat-shortening, can also result in paler meat and increased drip loss, as PSE. It could therefore be hypothesised that beef muscles could react in a similar way as the sheep muscle. According to Meat Standard Australia (MSA) pH 6 for beef carcasses should be reached in the interval 15-35°C, but several studies shows that the risk for heat-shortening increases with the temperature even if it still is in this interval. Locker and Hagyard (1963) found that the lowest risk for shortening occurred in the interval 14-19 °C and that the shortening increased with the temperature. Devine et al. (1999) found that the shortening when pH 6 occurred at a muscle temperature of 15 °C were limited and that the heat-

shortening increased with increased muscle temperature and at 35 °C the shortening of the muscle were 35 %. The findings by Locker and Hagyard (1963) and Devine et al. (1999) explained that there still can be heat-shortening even if pH 6 is reached in the interval 15-35 °C, especially in the higher muscle temperatures. Therefore, it is important to understand that the quality problems still can be present even if the average muscle temperature when pH 6 occurs is just below 35 °C. The abattoir in the present study should therefore aim for a temperature that is much lower than 35 °C to ensure to reduce the risk for heat-shortening. It is also important to limit the time from stunning until cooling to reduce the risk for heat-shortening, as it is for PSE, to be able to reach pH 6 in lower muscle temperatures. The main problem for both PSE and heat-shortening is a high muscle temperature and a rapid pH-decline, which means that some of the solutions might reduce both problems if both PSE and heat-shortening is occurring at the abattoir in the present study. In both cases the carcasses need to be chilled more rapid to decrease the risk for both quality problems. For further understanding of the problem, it would be of interest to evaluate the quality in the final product, as colour, drip loss and toughness to get a wider understanding of the quality problems that occurs.

5.5 Evaluation of method and sustainability aspects

To improve the method used in the present study, it would be more representative with a higher number of animals included to avoid unbalanced data. A higher number would make it possible to investigate differences depending on size of the animal, animal age and lairage time. It would also be of interest to analyse more blood parameters to get a wider understanding of the stress situation that the animals might experiences before slaughter and how that might influence the meat quality in the final product. To get a deeper understanding of the severity of the problem it would also be good to evaluate sensory attributes in the final product, which is not made in the present study.

Meat quality is important in several sustainability aspects during the whole process. Reduced quality in the final product, reduces the prices, and are therefore important in an economic perspective for the company. Animals experiencing stress pre-slaughter is one factor negatively affecting the meat quality (Coombes et al. 2014; Muižniece & Kairiša 2020). A good eating quality can often reflect the welfare of the animals and is therefore an important social sustainability and ethical aspect. A good quality and a good welfare also contribute to the reputation of the product and may be the reason that consumers buy from the same brand again. If the final product is of reduced quality, some may be discarded which is negative in an environmental perspective. The part that can be discarded due to

reduced quality also becomes a part of the resources that we do not take advantages of. Discarded meat also means that the animals that have been slaughtered for the human consumption is wasted and in an ethical perspective the death of the animals have not been exploited in a way that take advantages of the life that has been used to provide for people.

Slaughter of all animals awakes a lot of emotions and opinions for the society. The society often concern about the welfare of the animals during production but also regarding research, but animal-based research is according to studies often supported by the consumers if pain and distress is reduced (Fenwick et al. 2009). Fenwick et al. (2009) explains that the three R´s ethics is accepted when using animals in science. The three R´s stands for replacement, reduction and refinement. That means that if possible other alternatives than animals should be used (replacement), decrease the number of animals used in the study (reduce) and create an environment that fulfils the needs of the animals (refinement) (Fenwick et al. 2009). In the present study no animals are being slaughtered just for science, meaning that only animals that had a planned slaughter date have been included. The animals slaughtered in the study have been in the same environment as it always is in this abattoir, meaning that this study does not influence the normal environment at an abattoir, and both these factors is important in an ethical perspective and is probably more acceptable for society.

6. Conclusion

The main meat quality problem is that the rate of pH-decline is very rapid while the muscle temperature is still high, which results in that the muscles enter *rigor mortis* (pH 6) at high temperatures. However, the risk for PSE and heat-shortening increases with the fast pH drop, which might have happened previously in those cases where the final product has been pale and watery. The blood lactate concentrations were higher for bulls compared to heifers and cows but did not seem to influence the pH-decline. The pH_{24} were in the recommended range according to different standards and recommendations, and there were no differences between bulls, heifers and cows regarding pH_{24}, muscle temperature at pH 6 and slaughter weight. But cows had higher muscle temperature at 24 hours postmortem and lower conformation score compared to bulls and heifers. Regarding the fat score, muscle temperature and fat score were positively correlated, meaning that a higher fat score resulted in higher muscle temperatures. The cows also had a tendency to have lower fat scores than heifers. Electrical stimulation on the other hand, did not influence any of the parameters studied. But the effect of lairage time resulted in higher blood lactate concentrations for animals that stayed overnight, which were due to unbalanced groups with mainly bulls overnight, which produced higher blood lactate concentrations than both heifers and cows. Factors that can be improved to optimize the slaughter process and might result in improved meat quality is the time from exsanguination until cooling. A shorter time between exsanguination and the cooler might result in decreased muscle temperatures when pH 6 occurs. The design of the abattoir pre-slaughter should also be evaluated to ensure that the animals' environment is as calm as possible before slaughter, which might contribute to a better quality in the final product.

References

Aalhus, J.L., Jones, S.D.M., Best, D.R., Robertson, W.M., Lutz, S. (1994). The efficacy of high and low voltage electrical stimulation under different chilling regimes. Canadian Journal of Animal Science. 74 (3). 433-442. https://doi.org/10.4141/cjas94-062

Aalhus, J. l., Best, D. r., Murray, A. c. & Jones, S. d. m. (1998). A Comparison of the Quality Characteristics of Pale, Soft and Exudative Beef and Pork. *Journal of Muscle Foods*, 9 (3), 267–280. https://doi.org/10.1111/j.1745-4573.1998.tb00660.x

Aalhus, J.L., Janz, J.A.M., Tong, A.K.W., Jones, S.D.M. & Robertson, W.M. (2001). The influence of chilling rate and fat cover on beef quality. *Canadian Journal of Animal Science*, 81 (3), 321–330. https://doi.org/10.4141/A00-084

Abril, M., Campo, M.M., Önenç, A., Sañudo, C., Albertí, P., Negueruela, A.I. (2001). Beef colour evolution as a function of ultimate pH. *Meat Science*. 58 (1). 69-78. https://doi.org/10.1016/S0309-1740(00)00133-9

Acebrón, L.B. & Dopico, D.C. (2000). The importance of intrinsic and extrinsic cues to expected and experienced quality: an empirical application for beef. Food Quality and Preferences. 11(3). 229-238. https://doi.org/10.1016/S0950-3293(99)00059-2

Adeyemi, K.D. & Sazili, A.Q. (2014). Efficacy of Carcass Electrical Stimulation in Meat Quality Enhancement: A Review. *Asian-Australasian Journal of Animal Sciences*, 27 (3), 447–456. https://doi.org/10.5713/ajas.2013.13463

Agbeniga, B. & Webb, E.C. (2014). Influence of electrical stimulation on carcass and meat quality of Kosher and conventionally slaughtered cattle. *South African Journal of Animal Science*, 44 (5), 58–63. https://doi.org/10.4314/sajas.v44i5.12

Arik, E. & Karaca, S. (2017). The effect of some pre-slaughter factors on meat quality of bulls slaughtered in a commercial abattoir in Turkey. *Indian Journal of Animal Research*, 51 (3). https://doi.org/10.18805/ijar.v0iof.4563

Banović, M., Grunert, K.G., Barreira, M.M. & Fontes, M.A. (2009). Beef quality perception at the point of purchase: A study from Portugal. *Food Quality and Preference*, 20 (4), 335–342. https://doi.org/10.1016/j.foodqual.2009.02.009

Biraima, A.D.A., Mohammed, A.M. & Webb, E.C. (2019). Effects of electrical stimulation and age at slaughter on carcass and meat quality of two Sudanese Baggara beef types. *South African Journal of Animal Science*, 49 (5), 904–913. https://doi.org/10.4314/sajas.v49i5.14

Bodwell, C.E., Pearson, A.M. & Spooner, M.E. (1965). Post-Mortem Changes in Muscle. I. Chemical Changes in Beef. https://ift.onlinelibrary.wiley.com/doi/10.1111/j.1365-2621.1965.tb01838.x [2026-05-05]

Boles, J.A., Kohlbeck, K.S., Meyers, M.C., Perz, K.A., Davis, K.C. & Thomson, J.M. (2015). The use of blood lactate concentration as an indicator of temperament and its impact on growth rate and tenderness of steaks from Simmental×Angus steers. *Meat Science*, 103, 68–74. https://doi.org/10.1016/j.meatsci.2015.01.003

Braden, K.W. (2013). Converting Muscle to Meat: The Physiology of Rigor. I: Kerth, C.R. (editor). The Science of Meat Quality. John Wiley & Sons, Inc. 90-113. http://ebookcentral.proquest.com/lib/slub-ebooks/detail.action?docID=1166768 [2026-01-20]

Brazil Beef Quality (n.d.). *Grading meat*. https://www.brazilbeefquality.com/en/certifying-meat/ [2026-05-09]
Conroy, S.B., Drennan, M.J., McGee, M., Keane, M.G., Kenny, D.A., Berry, D.P. (2010). Predicting beef carcass meat, fat and bone proportions from carcass conformation and fat scores or hindquarter dissection. *Animal*. 4 (2). 234-241. https://doi.org/10.1017/S1751731109991121
Coombes, S.V., Gardner, G.E., Pethick, D.W. & McGilchrist, P. (2014). The impact of beef cattle temperament assessed using flight speed on muscle glycogen, muscle lactate and plasma lactate concentrations at slaughter. *Meat Science*, 98 (4), 815–821. https://doi.org/10.1016/j.meatsci.2014.06.029
Costa, F., Brito, G., Soares de Lima, J., Sant´Anna, A.C., Paranhos da Costa, M.J.R., del Campo, M. (2019). Lairage time effect on meat quality in Hereford steers in rangeland conditions. *Ruminants*. 48. https://doi.org/10.1590/rbz4820180020
del Campo, M., Brito, G., Soares de Lima, J., Hernández, P., Montissi, F. (2010). Finishing diet, temperament and lairage time effects on carcass and meat quality traits in steers. *Meat Science*. 86 (4). 908–914. https://doi.org/10.1016/j.meatsci.2010.07.014
Devine, C.E., Wahlgren, N.M., Tornberg, E. (1999). Effect of rigor temperature on muscle shortening and tenderisation of restrained and unrestrained beef *m. longissimus thoracicus et lumborum. Meat Science*. 51 (1). 61-72. https://doi.org/10.1016/S0309-1740(98)00098-9
Djimsa, B.A., Nair, M.N., Hess, A.M., Belk, K.E. & Woerner, D.R. (2022). The Impact of Carcass Size, Chilling Conditions, and Electrical Stimulation on Beef Postmortem Temperature and pH Decline. *Meat and Muscle Biology*, 6 (1). https://doi.org/10.22175/mmb.13893
Eikelenboom, G., Smulders, F.J.M. & Rudérus, H. (1985). The effect of high and low voltage electrical stimulation on beef quality. *Meat Science*, 15 (4), 247–254. https://doi.org/10.1016/0309-1740(85)90080-4
Elgasim, E.A., Kennick, W.H., McGILL, L.A., Rock, D.F. & Soeldner, A. (1981). Effects of Electrical Stimulation and Delayed Chilling of Beef Carcasses on Carcass and Meat Characteristics. *Journal of Food Science*, 46 (2), 340–343. https://doi.org/10.1111/j.1365-2621.1981.tb04857.x
Fenwick, N., Griffin, G. & Gauthier, C. (2009). The welfare of animals used in science: How the “Three Rs” ethic guides improvements. *The Canadian Veterinary Journal*, 50 (5), 523–530
Ferguson, D.M., Shaw, F.D. & Stark, J.L. (2007). Effect of reduced lairage duration on beef quality. *Australian Journal of Experimental Agriculture*, 47 (7), 770–773. https://doi.org/10.1071/EA05212
Gruber, S.L., Tatum, J.D., Engle, T.E., Chapman, P.L., Belk, K.E. & Smith, G.C. (2010). Relationships of behavioral and physiological symptoms of preslaughter stress to beef longissimus muscle tenderness1. *Journal of Animal Science*, 88 (3), 1148–1159. https://doi.org/10.2527/jas.2009-2183
Hazards (BIOHAZ), E.P. on B. (2014). Scientific Opinion on the public health risks related to the maintenance of the cold chain during storage and transport of meat. Part 1 (meat of domestic ungulates). *EFSA Journal*, 12 (3), 3601. https://doi.org/10.2903/j.efsa.2014.3601
Hertzman, C., Olsson, U., Tornberg, E. (1993). The influence of high temperature, type of muscle and electrical stimulation on the course of rigor, ageing and tenderness of beef muscles. *Meat Science*. 35 (1). 119-141. https://doi.org/10.1016/0309-1740(93)90074-R
Hopkins, D.L., Ponnampalam, E.N., Van De Ven, R.J. & Warner, R.D. (2014). The effect of pH decline rate on the meat and eating quality of beef

carcasses. *Animal Production Science*, 54 (4), 407–413. https://doi.org/10.1071/AN12314
Hui, Y.H. (2012). *Handbook of Meat and Meat Processing*. Taylor & Francis Group. http://ebookcentral.proquest.com/lib/slub-ebooks/detail.action?docID=846037 [2026-04-14]
Hultgren, J., Segerkvist, K.A., Berg, C., Karlsson, A.H., Öhgren, C. & Algers, B. (2022). Preslaughter stress and beef quality in relation to slaughter transport of cattle. *Livestock Science*, 264, 105073. https://doi.org/10.1016/j.livsci.2022.105073
Immonen, K., Ruusunen, M., Hissa, K. & Puolanne, E. (2000). Bovine muscle glycogen concentration in relation to finishing diet, slaughter and ultimate pH. *Meat Science*, 55 (1), 25–31. https://doi.org/10.1016/S0309-1740(99)00121-7
Jacob, R.H. & Hopkins, D.L. (2014). Techniques to reduce the temperature of beef muscle early in the post mortem period – a review. Animal Production Science. 54 (4). 482-493. https://doi.org/10.1071/AN12338
Kause, A., Mikkola, L., Strandén, I. & Sirkko, K. (2015). Genetic parameters for carcass weight, conformation and fat in five beef cattle breeds. *animal*, 9 (1), 35–42. https://doi.org/10.1017/S1751731114001992
Koh, K.C., Bidner, T.D., McMillin, K.W. & Hill, G.M. (1987). Effects of electrical stimulation and temperature on beef quality and tenderness. *Meat Science*, 21 (3), 189–201. https://doi.org/10.1016/0309-1740(87)90089-1
Lancaster, J.M., Buseman, B.J., Weber, T.M., Nasados, J.A., Richard, R.P., Murdoch, G.K., Price, W.J., Colle, M.J. & Bass, P.D. (2020). Impact of beef carcass size on chilling rate, pH decline, display color, and tenderness of top round subprimals. *Translational Animal Science*, 4 (4), txaa199. https://doi.org/10.1093/tas/txaa199
Lawrie, R.A. (1958). Physiological stress in relation to dark-cutting beef. *Journal of Food and Agriculture*. 9 (11). 721-727. https://doi.org/10.1002/jsfa.2740091106
Liza, F.A., Hosssan, M.I., Hashem, M.A. & Rahman, M.M. (2024). Effect of cattle age on the physio-chemical properties of beef. *Meat Research*, 4 (4). https://doi.org/10.55002/mr.4.4.98
Lochner, J.V., Kauffman, R.G. & Marsh, B.B. (1980). Early-postmortem cooling rate and beef tenderness. *Meat Science*, 4 (3), 227–241. https://doi.org/10.1016/0309-1740(80)90051-0
Locker, R.H. & Hagyard, C.J. (1963). A cold shortening effect in beef muscles. *Journal of the Science of Food and Agriculture*. 14 (11). 787-793. https://doi.org/10.1002/jsfa.2740141103
Marenčić, D., Ivanković, A., Pintić, V., Kelava, N. & Jakopović, T. (2012). Effect of the transport duration time and season on some physicochemical properties of beef meat. *Archives Animal Breeding*, 55 (2), 123–131. https://doi.org/10.5194/aab-55-123-2012
Marsh, B.B., Lochner, J.V., Takahashi, G. & Kragness, D.D. (1981). Effects of early post-mortem pH and temperature on beef tenderness. *Meat Science*, 5 (6), 479–483. https://doi.org/10.1016/0309-1740(81)90046-2
Matarneh, S.K., Scheffler, T.L., Gerrard, D.E. (2023). I: Toldrá, F. (editor). *Lawrie´s Meat Science*. Woodhead Publishing. 159-194. https://doi.org/10.1016/B978-0-323-85408-5.00010-8
Meat & Livestock Australia (n.d.). Tips & Tools, Meat Standards Australia beef information kit. *MLA Corporate*. https://www.mla.com.au/marketing-beef-and-lamb/meat-standards-australia/ [2026-04-08]

Meat & Livestock Australia (n.d.). Tips & Tools, Meat Standards Australia – The effect of pH on sheepmeat eating quality. https://www.mla.com.au/globalassets/mla-corporate/blocks/marketing-beef-and-lamb/msa_s5_web.pdf [2026-04-25]

Meat Standard Sweden nöt: MSS nöt (n.d.). *Slaktkroppens pH efter 24 timmar.* https://sluse.instructure.com/courses/8484/pages/slaktkroppens-ph-efter-24-timmar-meat-standard-sweden-not [2026-05-09]

Mueller, L.F., Balieiro, J.C.C., Ferrinho, A.M., Martins, T. da S., da Silva Corte, R.R.P., de Amorim, T.R., de Jesus Mangini Furlan, J., Baldi, F. & Pereira, A.S.C. (2019). Gender status effect on carcass and meat quality traits of feedlot Angus × Nellore cattle. *Animal Science Journal*, 90 (8), 1078–1089. https://doi.org/10.1111/asj.13250

Muižniece, I. & Kairiša, D. (2020). Effect of sex and age on beef cattle meat pH. *Agricultural Science and Practice*, 7 (2), 55–60. https://doi.org/10.15407/agrisp7.02.055

Mullen, A.M., Murray, B., Troy, D. (2000). Predicting the eating quality of meat. The National Food Centre Research Report No. 28. http://hdl.handle.net/11019/113

Nunes, C.L. de C., Vilela, R.S.R., Boas, P.G.V., Silva, J.C., Ramos, J.M.L., Martins, T. da S. & Chizzotti, M.L. (2024). Chilling Rates Impact Carcass and Meat Quality Parameters of Bos indicus Cattle. *Meat and Muscle Biology*, 8 (1). https://doi.org/10.22175/mmb.16908

O´Halloran, G.R., Troy, D.J., Buckley, D.J. (1997). The relationship between early post-mortem pH and the tenderisation of beef muscles. *Meat Science*. 45 (2). 239-251. https://doi.org/10.1016/S0309-1740(96)00074-5

Olsson, U. Hertzman, C., Tornberg, E. (1994). The influence of low temperature, type of muscle and electrical stimulation on the course of rigor mortis, ageing and tenderness of beef muscles. *Meat Science*. 37 (1). 115-131. https://doi.org/10.1016/0309-1740(94)90149-X

Page, J.K., Wulf, D.M. & Schwotzer, T.R. (2001). A survey of beef muscle color and pH. *Journal of Animal Science*, 79 (3), 678–687. https://doi.org/10.2527/2001.793678x

Paredi, G., Raboni, S., Bendixen, E., de Almeida, A.M. & Mozzarelli, A. (2012). "Muscle to meat" molecular events and technological transformations: The proteomics insight. *Journal of Proteomics*, 75 (14), 4275–4289. https://doi.org/10.1016/j.jprot.2012.04.011

Pogorzelska-Przybyłek, P., Nogalski, Z., Sobczuk-Szul, M. & Momot, M. (2021). The effect of gender status on the growth performance, carcass and meat quality traits of young crossbred Holstein-Friesian×Limousin cattle. *Animal Bioscience*, 34 (5), 914–921. https://doi.org/10.5713/ajas.20.0085

Probst, J.K., Hillmann, E., Leiber, F., Kreuzer, M. & Spengler Neff, A. (2013). Influence of gentle touching applied few weeks before slaughter on avoidance distance and slaughter stress in finishing cattle. *Applied Animal Behaviour Science*, 144 (1), 14–21. https://doi.org/10.1016/j.applanim.2012.12.007

Regulation (EC) No 853/2004 of the European Parliament and of the Council of 29 April 2004 laying down specific hygiene rules for food of animal origin. (OJ L 139, 40.4.2004, 55-205). http://data.europa.eu/eli/reg/2004/853/oj

Smith, G.C., Dutson, T.R., Hostetler, R.L. & Carpenter, Z.L. (1976). Fatness, Rate of Chilling and Tenderness of Lamb. *Journal of Food Science*, 41 (4), 748–756. https://doi.org/10.1111/j.1365-2621.1976.tb00717_41_4.x

Standards Australia. (2007). Australian standard for the hygienic production and transportation of meat and meat products for human consumption. AS: 4696:2007. CSIRO Publishing.
Steenkamp, J.E.M. & Van Trijp, H.C.M. (1996). Quality guidance: A consumer-based approach to food quality improvement using partial least squares. *European Review of Agricultural Economics*. 23 (2). 195-215. https://doi.org/10.1093/erae/23.2.195
Stevenson, K.E., Merkel, R.A., Lee, H.C. (1978). Effect of Chilling Rate, Carcass Fatness and Chlorine Spray on Microbiological Quality and Case-Life of Beef. *Journal of Food Science.* 43 (3) 849-852. https://doi.org/10.1111/j.1365-2621.1978.tb02437.x
Stiffler, D.M., Smith, G.C., Savell, J.W., Dutson, T.R., Griffin, C.L., Orcutt, M.W. (1984). Comparison of the Effects of High and Low Voltage Electrical Stimulation on Quality-Indicating Characteristics of Beef Carcasses. *Journal of Food Science*. 49 (3). 863-866. https://doi.org/10.1111/j.1365-2621.1984.tb13229.x
Sullivan, P.A., Davis, M.K., Nair, M.N., Hess, A.M., Mooney, D.F. & Edwards-Callaway, L.N. (2024). Preslaughter factors affecting mobility, blood parameters, bruising, and muscle pH of finished beef cattle in the United States. *Translational Animal Science*, 8, txae035. https://doi.org/10.1093/tas/txae035
SvensktKött. (n.d). *Klassificering av slaktkroppar*. https://svensktkott.se/om-kott/kottkvalitet/klassificering-av-slaktkroppar/. [2026-04-25]
Warner, R.D., Ferguson, D.M., Cottrell, J.J. & Knee, B.W. (2007). Acute stress induced by the preslaughter use of electric prodders causes tougher beef meat. *Australian Journal of Experimental Agriculture*, 47 (7), 782. https://doi.org/10.1071/EA05155
Wulf, D.M., O'Connor, S.F., Tatum, J.D. & Smith, G.C. (1997). Using objective measures of muscle color to predict beef longissimus tenderness1. *Journal of Animal Science*, 75 (3), 684–692. https://doi.org/10.2527/1997.753684x
Węglarz, A. (2010). Meat quality defined based on pH and colour depending on cattle category and slaughter season. *Czech Journal of Animal Science*, 55 (12), 548–556. https://doi.org/10.17221/2520-cjas

Popular science summary

When costumers buy meat at the store, their choices are often made according to the visual appearance of the meat, such as meat colour and the amount of visible fat. Meat characteristics are the result of several biochemical processes, which in turn are affected by the experiences of an animal the time before slaughter, as well as factors after slaughter such as the rate of temperature decline in the carcass and the drop in pH. The storage of glycogen in animals is used in the muscle after slaughter to produce lactic acid, which is the cause for the decline in pH. Several factors that influence the meat characteristics are connected to the animal itself like the sex, age, breed, slaughter weight and carcass conformation for example. However, short-term and long-term stress are also important to consider. Short-term stress can be caused by several different factors from leaving the farm until slaughter, like the transport, loading/unloading of animals, space allowance during transport and at the abattoir, and by unfamiliar humans and other animals. Short-term stress might result in high levels of lactic acid in the blood the time before slaughter as a stress response, which might result in a fast drop in pH while the temperature in muscles is still high. This might result in a quality defect causing pale, soft and exudative meat (PSE) or heat-shortening. Long-term stress before slaughter might reduce the animal's glycogen storage. So, if an animal has been stressed for a longer period of time, the glycogen stores will be used to cope with the stress/excitement, which might result in too low levels of glycogen at slaughter, which in turn results in a low decrease in pH. A low drop in pH, might result in too high pH in the meat, and a meat quality defect causing dark, firm and dry meat (DFD) may occur, which is not wanted by consumers.

An abattoir in Sweden has experienced some quality deviations with the meat in the topside muscle, where the final product is paler and more watery than normal. The purpose with this study was to evaluate the slaughter system and investigate if any particular parts of the system could be optimized to avoid the problem. The study investigated if there were differences between sexes, usage of electrical stimulation and lairage time before slaughter (if animals were at the abattoir overnight or not). Parameters such as blood lactate concentration, carcass composition and the decline in pH and muscle temperature during the first 24 hours after slaughter were measured. The results showed that the carcasses had a rapid pH-decline and a high muscle temperature after slaughter leading to an increased risk for pale, soft and exudative meat (PSE) and heat-shortening. The sex did not seem to influence the pH- and temperature-decline in general, however, cows had a slightly higher muscle temperature 24 hours after slaughter compared to heifers and bulls. There was also a difference seen in blood lactate concentration, where bulls generally had higher values. The effect of electrical

stimulation did not influence any of the parameters tested. But the overnight lairage resulted in higher lactate concentration in blood, which can be explained by an overrepresentation of bulls in this group compared to dairy cows and heifers in the other group. To reduce the occurrence of meat quality deviations, the carcasses need to enter the coolers faster and it would also be good to evaluate the stable to ensure that the environment is as calm as possible for the animals to avoid unnecessary stressors before slaughter.

Publishing and archiving

Approved students' theses at SLU can be published online. As a student you own the copyright to your work and in such cases, you need to approve the publication. In connection with your approval of publication, SLU will process your personal data (name) to make the work searchable on the internet. You can revoke your consent at any time by contacting the library.

Even if you choose not to publish the work or if you revoke your approval, the thesis will be archived digitally according to archive legislation.

You will find links to SLU's publication agreement and SLU's processing of personal data and your rights on this page:

- https://libanswers.slu.se/en/faq/228318

☒ YES, I, Tova Pettersson, have read and agree to the agreement for publication and the personal data processing that takes place in connection with this

☐ NO, I/we do not give my/our permission to publish the full text of this work. However, the work will be uploaded for archiving and the metadata and summary will be visible and searchable.