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There is no easy answer

Factors affecting meat quality in lambs

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There is no easy answer – Factors affecting meat quality in lambs

Abstract

This thesis examined the effects of production system, slaughter method, carcass handling procedures and storage method on the quality of meat from intact male lambs. Quality aspects of the meat assessed were carcass quality, meat quality indicators (measured on carcasses), technological meat quality (measured on meat samples) and sensory attributes. Four different production systems were studied: indoor feeding with silage and concentrate; cultivated pasture with or without concentrate feeding; and grazing on semi-natural pasture. Different slaughter methods (small-scale and large-scale), including different stunning methods, chilling regimes and use or not of electrical carcass stimulation, were tested. Storage methods studied included differences between fresh or frozen meat, to evaluate the effect of freezing.

Differences in feeding intensity in the four production systems affected lamb live weight gain. Carcass weight, dressing percentage, conformation score and fatness score were also affected by production system. On comparing slaughter systems, a difference was found for fatness score, but not for carcass weight or conformation score. Production system and slaughter method both had no effect on pH measurements 24 hours after slaughter. Production system had no effect on meat colour, but slaughter method affected lightness of the meat. Freezing did also cause colour differences. Production system or slaughter method did not result in differences in thawing or cooking loss. Frozen storage caused an increase in total fluid losses (thawing and cooking losses) and some differences in meat colour (redness, lightness). Tenderness (measured as Warner-Bratzler shear force, WBSF) did not differ between the production systems or between the different slaughter methods, but freezing increased WBSF compared with fresh meat. Production system affected some sensory attributes of the meat (resistance to cutting, hay odour, leafy flavour), as did slaughter method (colour appearance, fatty flavour). Storage method caused differences in fatty odour, frying flavour, sour flavour, fatty flavour, liver flavour and juicy, texture and a tendency for a difference for mushy texture.

Keywords: lamb, meat quality, production systems, carcass quality, pH, muscle temperature, meat colour, thawing and cooking losses, Warner-Bratzler shear force, sensory assessment

Det finns inget enkelt svar – Faktorer som påverkar köttkvaliteten hos lamm

Abstract

Denna avhandling har undersökt hur produktionssystem, slaktmetoder, slaktkroppshantering och lagringsmetoder påverkade kvaliteten hos kött från intakta bagglamm. Kvalitetsaspekter hos köttet har köttet benämnts som slaktkroppens kvalitet, köttkvalitetsindikatorer (mätt på slaktkroppen), teknologisk köttkvalitet (mätt från köttprov) och sensoriska tester. De olika produktionssystemen som studerades var; inomhusutfodring med ensilage och kraftfoder, åkermarksbete med eller utan kraftfoder och naturbete. Olika slaktmetoder (småskalig och storskalig) inkluderande olika bedövningsmetoder, nedkylningsmetoder och användning eller inte av elstimulering av slaktkroppar testades. De studerade lagringsmetoderna innefattade skillnader mellan färskt eller fryst kött, för att utvärdera effekten av frysning.

Utfodringsintensitet påvisade en effekt på daglig tillväxt. Slaktkroppsvikt, slaktutbyte, konformationsklass och fettklass påverkades alla av produktionssystem. Vid jämförelse av slaktsystem påvisades en skillnad för fettklass men inte för slaktvikt eller konformationsklass. Varken produktionssystem eller slaktmetod påvisade effekt av pH-mätningar 24 timmar efter slakt. Produktionssystemet påvisade ingen effekt på köttets färg men slaktmetod påverkade däremot köttets ljushet. Varken produktionssystem eller slaktmetod påverkade köttets vätskeförluster i form av upptiningssvinn eller koksvinn. Fryslagring gav en ökad vätskeförlust (upptining och kokning) samt viss skillnad i köttets färg (rödhet och ljushet). Mörheten(mätt som Warner-Bratzler skärmotstånd, WBSF) skilde sig varken mellan de olika produktionssystemen eller mellan slaktmetoderna, frysning å andra sidan ökade WBSF jämfört med färskt kött. Utfodring påverkade de sensoriska egenskaperna: skärmotstånd, hö lukt och lövsmak. Slaktmetod påverkade de sensoriska egenskaperna färg och fet smak. Förvaringsmetod orsakade skillnader i fet lukt, steksmak, sur smak, fet smak, leversmak, saftig textur och en tendens till skillnad för mosig textur.

Nyckelord: lamm, köttkvalitet, produktionssystem, slaktkroppskvalitet, pH, muskeltemperatur, köttfärg, tinings- och koksvinn, Warner-Bratzler skärmotstånd, sensorisk bedömning

Dedication

Till mamma

“Let us not take ourselves too seriously. None of us has a monopoly on wisdom.”
Queen Elizabeth II

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List of publications

This thesis is based on the work contained in the following papers, referred to by Roman numerals in the text:

I. Stenberg, E., Karlsson, A., Öhgren, C. & Arvidsson Segerkvist, K. (2020). Carcass characteristics and meat quality attributes in lambs reared indoors, on cultivated pasture, or on semi-natural pasture. *Agricultural and Food Science*, vol. 29, pp. 432–441.

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IV. Stenberg, E.; Arvidsson-Segerkvist, K.; Karlsson, A.H.; Ólafsdóttir, A.; Hilmarsson, Ó.Þ.; Gudjónsdóttir, M. & Thorkelsson, G. (2022). A Comparison of Fresh and Frozen Lamb Meat—Differences in Technological Meat Quality and Sensory Attributes. *Animals*, vol. 12, 2830.

The contribution of Elin Stenberg to the papers included in this thesis was as follows:

I. Planning the research jointly with the co-authors, collecting samples and conducting experimental measurements, processing of data and responsible for compiling the manuscript.

II. Planning the research jointly with the co-authors, collecting samples, processing of data and responsible for compiling the manuscript.

III. Planning the research jointly with the co-authors, collecting samples and conducting experimental analysis, processing of data and responsible for compiling the manuscript.

IV. Planning the research jointly with the co-authors, collecting samples and conducting experimental analysis, processing of data and responsible for compiling the manuscript.

Abbreviations

a*	Redness
b*	Yellowness
BCFA	Branched chain fatty acids
DFD	Dark, firm and dry
DM	Dry matter
e.g.	*Exempli gratia*/for example
HA	Hectare
i.e.	*id est*/in other words
L*	Lightness
LWG	Live weight gain
LTL	*Musculus longissimus thoracis et lumborum*
N	Newton
NMR	Nuclear magnetic resonance
PSE	Pale, soft and exudative
WBSF	Warner-Bratzler shear force

1. Introduction

Sheep are fantastic creatures! It is not difficult to understand why our ancestors domesticated these amazing animals and why they are still an important form of livestock in the modern agricultural world. Sheep provide many excellent products that have been helpful for the survival of mankind over thousands of years and are still very important today (Mazinani & Rude, 2020). These products include wool, meat, milk, sheepskins, leather, open landscapes, biodiversity and, last but not least, lambs.

Today, domestic production accounts for only approximately 30% of the lamb meat consumed in Sweden, meaning that 70% of all lamb meat consumed is imported (Lannhard Öberg, 2022). Thus the sector is small, but from another perspective this imbalance provides an opportunity to increase domestic production without a need to increase actual meat consumption, which would be positive for the Swedish trade balance. However, any expansion in domestic production of lamb meat will be dependent on achieving economically sustainable farm enterprises, which will depend in turn on many different societal factors such as politics and consumer demand.

In order to make future sheep production more successful, it will be necessary to increase consumer demand for Swedish lamb meat and to produce more uniform meat. To this end, it is of critical importance to standardise and improve meat quality. The eating quality of Swedish lamb meat currently varies in both eating quality attributes and in the size of valuable cuts, mainly due to use of different production systems, different breeds, and varying age at slaughter and slaughter weight (Carlsson & Arvidsson Segerkvist, 2018). It is well known that meat quality is a complex subject that is influenced by a multitude of factors from field to fork (Sañudo et al., 1998). These factors include e.g. the feeding strategies used in different

production systems, sheep breed and sex, pre-slaughter conditions, slaughter methods, handling of carcasses after slaughter and storage of meat before consumption (Sañudo et al., 1998). All these factors also interact to determine the outcome in terms of meat quality, as indicated in a mind map in Figure 1. Determination of meat quality is thus very complex and can be difficult to assess.

Figure 1. Illustration of factors affecting meat quality included in the conducted studies.

1.1 Assessment of meat quality

Lamb meat is often described as tender, but the degree of tenderness may have changed over recent decades. For example, comparisons of instrumental tenderness scores, measured as Warner-Bratzler shear force (WBSF), indicate a decrease in tenderness in Icelandic lamb meat in 2018 (Thorkelsson et al., 2018) compared with 2003 (Sañudo et al., 2003). This decrease in tenderness may be related to different factors, such as breeding for lean carcasses with increased muscle mass (Eiríksson & Sigurdsson, 2017) and post-slaughter procedures such as different strategies for chilling and ageing (Warner et al., 2010). An effect on meat quality can therefore derive from different factors, making it important to include the whole production system from rearing on the farm to end-product on the plate when seeking to identify possible influencing factors.

The final step in the value chain of meat is the consumer, with meat as the end-product, and eating quality is of high importance from a consumer perspective. In particular, tenderness, juiciness and flavour are often listed as the most important quality attributes for meat in general (McIlveen & Buchanan, 2001). For lamb meat in particular, flavour is described as the most important attribute, followed by tenderness and juiciness (Pethick et al.,

2006a; Young et al., 1997). Therefore, sensory attributes must be assessed when discussing meat quality in order to obtain a complete picture of eating quality.

This thesis investigated the effects of different factors in the production system for lamb meat on meat quality. Based on the factors mentioned above, the investigations were designed to cover the effect of feeding (pasture, concentrate) and rearing of lambs on the farm, of different slaughter processes and of different storage methods on different quality aspects of the final product (live weight gain, carcass quality, technological meat quality and sensory attributes).

1.1.1 Muscle becomes meat

After an animal has been slaughtered, the conversion of muscle to meat begins. This process involves several biochemical changes over time. According to the open public food database at the Swedish Food Agency, uncooked lamb meat without fat cover consists of 76.5% water, 20% protein, 2.5% fat and 1% ash (vitamins and minerals) (Swedish Food Agency, 2023). When assessing meat quality, it is crucial to understand the basic processes that occur when muscle becomes meat after the animal has been slaughtered. One important factor is feeding regime (amount, quality, composition), as it influences e.g. live weight gain (LWG) of the animal. Adequate feeding enables muscle glycogen storage, which in turn governs the desired pH decline in muscles after slaughter. Glycogen serves as an important energy supply for energy production in post-mortem muscle when oxygen is no longer present. In this process, glycogen is converted into energy (as ATP), with lactic acid as a by-product, which lowers muscle pH due to lactic acid accumulation within the muscle. In the living animal, the lactic acid is transported away from the muscle by the blood system. This process is thus glycogen-dependent and it is therefore crucial to have adequate glycogen levels in muscle prior to slaughter to achieve an optimal pH decrease (Bendall, 1973). Conversion of glycogen into lactic acid stops when there is no more glycogen available or when enzymes responsible for this process are deactivated due to low pH. Adequate glycogen storage in muscle can be achieved by appropriate feeding and it is therefore important that slaughter animals are in positive energy balance (growth phase). The current recommendation in Australia is that lambs should grow by at least 100 g/day during the last two weeks prior to slaughter (MLA, 2019a). Further, it is

important that glycogen stores in the animal are not depleted before slaughter due to e.g. stress reactions or fasting. This can be avoided by offering adequate feeding to promote weight gain and prevent weight loss prior to slaughter (MLA, 2019a).

1.1.2 Effects of pH and temperature

As mentioned, muscle pH is strongly dependent on transformation of glycogen into lactic acid during anaerobic glycolysis, where lactic acid accumulation in muscle leads to a pH drop from neutral (pH 7) to a final pH of approximately 5.3 to 5.8. The pH decrease can be monitored over time, in relation to the muscle temperature or by itself as a final value. Final pH at 24 h post-slaughter, or pH_{24} as it is often called, should not fall below the recommended value of 5.7 according to Meat Standards Australia (MLA, 2019b), while 5.4-5.8 is the desirable range according to Alliance Group Limited (New Zealand) (Alliance Group Limited, 2010). Measured pH is often used as an indicator of tenderness in meat (Toohey *et al.*, 2006; Thompson *et al.*, 2005; Geesink *et al.*, 2000). For example, muscle pH in relation to muscle temperature can be used as a tool to predict the tenderness of meat (Devine *et al.*, 1993). To promote more tender meat and avoid quality problems, pH and temperature should also decrease at a specific recommended rate. Measurements are specifically made to identify the temperature at which *rigor mortis* occurs (pH 6.0), in order to detect problems within production systems, slaughter systems or a specific carcass. Problems to avoid in this particular context are DFD (dark, firm and dry) meat, PSE (pale, soft and exudative) meat, heat toughening and cold shortening, all of which have a negative effect on several meat quality parameters. For example, Locker and Hagyard (1963) established that if beef muscle is in pre-rigor condition (pH about 6.0-6.4) when muscle temperature decreases below 10-15°C, cold shortening can occur, resulting in toughening of the meat (Locker & Hagyard, 1963). Some muscles are more susceptible to cold shortening than others. It has been shown in pigs that the intensity of cold shortening increases with increasing proportion of red muscle fibres, with the exception of the *Musculus longissimus dorsi* (Bendall, 1975). The temperature window within which pH 6.0 should be reached depends on slaughter method (electrical stimulation or not) and suspension method (Achilles tendon or pelvic suspension). Ageing time is also affected by the different recommendations. An Achilles tendon-suspended carcass that has

not received electrical stimulaation should enter *rigor mortis* at a temperature between 8 and 18 °C and should be aged for a minimum of ten days at 1 °C, according to Australian recommendations (MLA, 2019b). An Achilles tendon-suspended carcass that has been electrically stimulated should reach pH 6.0 between 18 and 35 °C and ageing time is a minimum of five days at 1 °C. For a tender-stretched (pelvic-suspended) carcass without electrical stimulation, a temperature between 8 and 35°C at pH 6.0 is recommended and it should be aged for a minimum of five days at 1°C (MLA, 2019b).

1.2 Meat quality parameters

1.2.1 Carcass quality

Today, lamb carcass quality is described as carcass weight, carcass conformation, carcass fatness and dressing percentage. All these descriptors of the carcass are used by the abattoir to calculate a value for each individual carcass. The carcass value is not linear in terms of increasing muscles and fat. Rather, it is more of a "best paid" structure where a price reduction or increase is based on specific scoring. For example, a fat score outside a specific threshold incurs a price reduction. Carcass conformation and fatness are usually assessed manually, including in Sweden, by a certified grader at the abattoir. In European countries, the EUROP scale is used for scoring of conformation and fatness. The number of classes in the Swedish version of the system is 15 for both conformation and fat, where a score of 1 refers to poor conformation/very low fat and a score of 15 refers to excellent conformation/very high fat. The number of classes may differ between countries that use the EUROP classification system.

1.2.2 Meat quality indicators

As described in previous sections, measurements of pH and muscle temperature can be used as indicators of meat quality, for example expected tenderness. In particular, such measurements are used to detect and predict potential meat quality problems such as DFD, PSE, cold shortening and heat toughening. Repeated measurements of pH alone can be carried out in the same or different muscles post-slaughter, to monitor developments over time. The final pH value is usually measured at 24 hours after slaughter. The temperature data obtained can be analysed together with pH at the same time-

points in order to chart the parallel decline in temperature and pH, which can be used to detect and predict potential risks of meat quality problems.

1.2.3 Technological meat quality attributes

Technological meat quality assessed within the work of this thesis was assessed as fluid loss, instrument-measured meat colour and Warner-Bratzler shear force (WBSF).

Fluid loss

The loss of fluid from a meat sample can be expressed in different ways. In this thesis, it was determined as thawing loss, cooking loss and total fluid loss. These parameters refer respectively to the loss of fluid from the meat sample during thawing (thawing loss), cooking (cooking loss) and combined losses from both thawing and cooking. Another way of analysing the water content in muscle is by using low field nuclear magnetic resonance (LF-NMR), which detects water distribution within muscle samples and how strongly those specific water populations are bound to proteins within each sample by their relaxation times.

Instrumental meat colour

Meat colour can be measured according to the CIELAB system (International Commission on Illumination, 1976). The colour attributes analysed in this thesis were lightness (L*), redness (a*) and yellowness (b*). These specific colour attributes are important from the consumer's point of view when assessing the meat package in a purchase situation.

Warner-Bratzler shear force (WBSF)

In the WBSF method, the tenderness of cooked meat is measured using an instrument that determines the force needed, often expressed in Newtons (N), to cut through a specific meat sample of a particular size. The cut is made orthogonal to the fibre direction of each muscle sample. The underlying concept for this type of instrumental measurement is to mimic the chewing situation in a person's mouth. The force needed to cut through the meat sample with the instrument can thus be equated to the force needed for the molar teeth to bite through the meat, and the results can be used to discuss tenderness of the meat sample. However, there may be different perceptions on what constitutes tender meat. Recommendations regarding this concept are available and are presented later in the thesis.

1.2.4 Sensory attributes

To assess how meat samples might be perceived by a consumer in a more detailed way, a sensory panel can be used to evaluate and score meat for different attributes. These attributes are often related to odour, flavour, appearance and texture. The sensory attributes can also be compared with the results of technological meat quality analysis, to compare differences that the panel could identify and those that might not have been detected. The sensory panels used in the thesis were both trained panels. A trained panel is used to describe individual samples without ranging them in any order of acceptance. Another common type of panel, the called consumer panel, can be used for preference testing of different samples.

1.3 Factors affecting meat quality

The effect of different factors on lamb meat quality were evaluated in different studies in this thesis, as described in Papers I-IV. As mentioned, there are many factors affecting meat quality attributes and in various ways. Some of these different factors are described below and related to the work conducted within this thesis.

1.3.1 Sex

Meat from intact male lambs may have a higher incidence of undesirable flavour and aroma attributes. This may be connected to age at slaughter, with increasing age increasing the risk of undesirable flavour and aroma compounds (Gkarane *et al.*, 2017). This in turn is related to increased levels of specific branched chain fatty acids (BCFA), which contribute to increased intensity of *e.g.* lamb flavour, lamb aftertaste and fatty aftertaste in meat (Gkarane *et al.*, 2020). In order to confirm whether intact male lambs and castrates differ in terms of sensory attributes, further research needs to be conducted on different breeds and feeding regimes. In previous studies, shear force values obtained for meat from castrates and intact male lambs of different ages did not reveal any differences before the lambs reached more than 300 days of age, which may imply that castration affects meat quality in lambs (Young *et al.*, 2006), but this needs further evaluation. In particular, more research on the effect of intact male lambs is needed in order to fill the knowledge gap in this particular field.

1.3.2 Breeding

Meat quality attributes can be both heritable and affected by *e.g.* diet. Shear force values and meat colour stability are examples of attributes that can be influenced by genetic selection, while *e.g.* meat fatty acid composition is to a high degree influenced by diet composition (Jacob & Pethick, 2014). Breeding for increased muscle mass and less fat in the carcass may negatively affect the eating quality of lamb meat according to Eiríksson and Sigurdsson (2017). An incentive for this type of breeding strategy (selection) is created by the EUROP classification system used at abattoirs, which rewards muscular carcasses with a limited amount of fat. However, Pannier *et al.* (2014) found that selection for high lean meat yield can negatively affect the eating quality of lamb meat as assessed by consumers, which can cause future problems for the lamb meat industry if not monitored carefully. Pethick *et al.* (2006b) suggest that if this breeding strategy continues, future improvements in meat quality will have to be derived from carcasses with reduced fatness and more muscle.

1.3.3 Rearing model

Feeding strategy and diet composition have been identified as factors affecting meat quality in different regards. Different feeding strategies, including e.g. pasture-fed lambs or grain-fed lambs, can cause meat to appear different in flavour, but can also have an effect on glycogen storage in muscles, directly affecting the quality of meat (Watkins et al., 2013). The relationship between LWG and pH has been examined in previous studies, with varying results (Majdoub-Mathlouthi et al., 2013; Hopkins et al., 2005). Majdoub-Mathlouthi et al. (2013) did not find any differences in final pH as an effect of different feeding strategies that promoted differences in LWG, whereas Hopkins et al. (2005) found that meat pH was affected by feeding strategy, with higher pH in meat from animals fed a diet promoting lower LWG than in meat from animals fed a diet promoting higher LWG. The feeding strategy for lower LWG, which resulted in increased pH, also increased the shear force values and therefore reduced the tenderness of the meat (Hopkins et al., 2005). The opposite has also been observed, e.g. Pethick et al. (2005) found that lambs gaining weight had higher pH_{24} values compared with lambs not gaining weight. The concept of LWG and final pH are thus affected by other factors in the system. Diet composition, with particular differences in grain-based diets vs. pasture-based diets, may affect

sensory aspects of the meat and in particular the flavour of the meat (Resconi et al., 2009; Arsenos et al., 2002; Priolo et al., 2002; Fisher et al., 2000). Live weight gain may also influence sensory attributes of the meat, with meat from low LWG lambs resulting in higher final pH and differences in the sensory attributes aroma, flavour and texture compared with lambs with high LWG (Campbell et al., 2012).

Evaluation of sensory attributes

Based on findings in previous studies regarding the importance of flavour attributes in relation to overall satisfaction of meat quality, sensory evaluation is of the utmost importance when assessing meat quality (Pethick et al., 2006a). The flavour attributes relate to the species-characteristic flavour and odour that can occur in lamb meat (Young et al., 1994). It is therefore important to understand and acknowledge the factors affecting such attributes, in order to minimise the risk of unwanted specific sensory attributes occurring in the meat. Sensory attributes are affected by the animal's diet, but may also be partly affected by breeding (Jacob & Pethick, 2014). In addition, Pethick et al. (2005) showed that having lambs still growing before slaughter prevented decreases in consumer scores for meat juiciness and flavour-liking observed for meat from with lambs losing weight before slaughter. Differences in meat pH may also result in sensory differences, where high final pH scores lower in sensory evaluations for overall flavour and odour, sheep meat flavour and odour, and foreign odour than meat of low pH (Young & Braggins, 1996). However, it is important to note that the sensory effect of low pH in that study was seen at 5.66 and 'high' pH was 6.81.

Consumers may differ in the sensory attributes they prefer and which they regard as non-preferable, sometimes described as off-flavours or odours. Such differences may relate e.g. to diets based on milk, concentrate or pasture. A high proportion of concentrate in the diet (compared with pasture) can decrease species-specific sensory attributes (Young & Braggins, 1996). Moreover, maize-finished lambs have been reported to receive lower sensory scores for the attributes strong, grassy and lamby compared with forage-finished lambs (Bailey et al., 1994). However, diets based on forage or pasture can also differ in composition and should not be regarded as equivalent, as their effects on sensory attributes can differ (Bailey et al., 1994). Hence, sensory attributes of meat are directly linked to the composition of the diet fed to animals, due to transfer of feed components

(Watkins et al., 2013). The more species-specific flavours and odours originate from volatile fatty acids (stored as fat) (Young & Braggins, 1996; Hornstein & Crowe, 1963). Some consumers prefer meat from lambs fed a specific type of diet and some may prefer or accept meat with different sensory profiles (Sañudo et al., 2007). The sensory profile of meat is linked to liberation of flavour compounds. The cooking process results in the formation and degradation of compounds in the meat, and the release of aroma volatiles. The reactions involved include Maillard reactions between amino acids and reducing sugars and degradation of lipids (Mottram, 1998).

1.3.4 Slaughter system

When an animal is slaughtered, in Sweden and in many other parts of the world, it must be stunned before exsanguination. Stunning can be performed in different ways. The "head-to-leg" electrical stunning method has been shown to affect meat quality in terms of enhancement of the post-mortem pH decline, which could be positive if cold shortening is a risk during the chilling process (Petrović et al., 1993). Stunning method may therefore also contribute to meat quality and should not be overlooked when analysing the slaughter system. To avoid cold shortening, electrical stimulation can be applied in the production chain at the abattoir (Pouliot et al., 2012). This procedure can be used on carcasses up to 30 minutes after slaughter to avoid cold shortening and promote tenderness (Hopkins & Ferrier, 2000). Electrical stimulation can also be used to hasten the pH decline in muscles post-mortem and avoid cold shortening when fast chilling of the carcasses is part of the system at the abattoir (Campañone et al., 2006). Electrical stimulation can therefore be used to optimise the eating quality of the meat under slaughter conditions, by controlling the pH decline in relation to muscle temperature (Abhijith et al., 2020; Leygonie et al., 2012). Decreasing tenderness in lamb meat could be due to changes in the slaughter system, which may be connected to increased risk of cold shortening when fast chilling is applied after electrical stimulation (Warner et al., 2010). The slaughter system therefore needs to be optimised in terms of the combination of practices applied at the specific abattoir to avoid such problems, both with and without application of electrical stimulation at the slaughter line. Electrical stimulation is not used in all sheep and lamb slaughter worldwide, for instance it is not used at Swedish abattoirs.

There are different types of electrical stimulation system for lamb carcasses, which are usually divided into low-voltage and high-voltage stimulation. A third category, medium-voltage stimulation, has also been developed in order to get an increased effect compared with low voltage in a system that is not as unsafe in terms of working environment as high-voltage stimulation (Shaw et al., 2005). There are many different definitions of low-, medium- or high-voltage stimulation and the definitions may differ between studies. Low-voltage stimulation is often defined as use of maximum 45 V peak (Shaw et al., 2005; Morton & Newbold, 1982) or below 100 V (Cetin et al., 2012); medium-voltage stimulation as use of maximum 450 V peak (Shaw et al., 2005); and high-voltage stimulation as use of 100 V peak (Cetin et al., 2012), 850 V peak (Morton & Newbold, 1982) or 1100 V peak (Shaw et al., 2005). Current (Ampere) and frequency (Hertz) may also differ depending on e.g. animal species and equipment used. Electrical stimulation can be used at different stages after the animal has been slaughtered, for example it can be applied to wool-on carcasses or after dressing (Shaw et al., 2005). Low-voltage stimulation has been used on lamb carcasses with successful results in avoiding cold shortening when fast chilling has been applied (Polidori et al., 1999). This finding contradicts claims that low-voltage stimulation is not enough to enhance rapid pH decline and that high-voltage stimulation should therefore be used (Pommier et al., 1989). However, Morton and Newbold (1982) concluded that low-voltage stimulation has to be applied soon after slaughter in lamb carcasses, since a functioning nervous system has to be present for low-voltage stimulation to give an effect. The nerve system in lamb carcasses deteriorates within 30 minutes post-slaughter (Chrystall et al., 1980), and therefore higher voltage than low-voltage stimulation may be necessary to obtain the optimal effect of electrical stimulation if not performed soon after stunning (Morton & Newbold, 1982; Chrystall et al., 1980). Low-voltage stimulation has generally been shown to have no effects on cooking loss compared with unstimulated carcasses (Pouliot et al., 2012; Hopkins & Ferrier, 2000; Lee et al., 2000). An attribute that has been found to be affected in previous studies is meat colour (Pouliot et al., 2012; Toohey et al., 2006; Hopkins et al., 2005; Hopkins & Ferrier, 2000). Other attributes reported to be both affected and unaffected by electrical stimulation are WBSF (Pouliot et al., 2012; Toohey et al., 2008; Hopkins & Ferrier, 2000; Lee et al., 2000; Polidori et al., 1999; Pommier et al., 1989; Solomon & Lynch, 1988; Solomon et al., 1986;

Chrystall et al., 1984; Savell et al., 1977) and different sensory attributes (Pouliot et al., 2012; Pommier et al., 1989; Solomon & Lynch, 1988; Solomon et al., 1986; Savell et al., 1977). The use of electrical stimulation as a tool to obtain positive effects on lamb meat quality attributes and the level of voltage and other specific settings that should be used need further study, in order to optimise the effect of different slaughter systems and animal materials on meat quality.

1.3.5 Storage method

Lamb meat is often viewed as a seasonal product in the Northern hemisphere, which is explained by the traditional lambing period occurring in spring due to the normal seasonal reproductive cycle in sheep (Chemineau et al., 2008). Therefore, it is more common to have a short slaughter season in the autumn, which is followed by sales of fresh meat in local markets, while the surplus can be stored as frozen meat. Use of frozen storage has increased the opportunity to store meat for longer periods, not just for shipping but also for stabilising the market and offering consumers lamb meat all year around and not just for a limited season (Pietrasik & Janz, 2009; Wheeler et al., 1990). Freezing surplus meat reduces the risk of lowering the market price in the autumn and enables storage of the meat until market prices are higher and conditions are more profitable by selling meat in all seasons (Campañone et al., 2006). Since meat is sold both fresh and frozen, it is important to understand how freezing may affect meat quality and to optimise the storage conditions without decreasing the eating quality of the product. Freezing is used to preserve meat quality as close to the quality of fresh meat as possible under longer-term preservation (Smith et al., 1968). A review by Leygonie et al. (2012) of previous literature concluded that freezing can cause increased fluid losses from meat, but there are still inconsistencies about the effect of freezing on other parameters, such as tenderness and meat colour. Method of freezing, for example if the freezing procedure is fast or slow, also affects meat quality attributes. A slow freezing process often results in larger ice crystals, both intracellular and intercellular (between and within the muscle cells). Fast freezing, on the other hand, promotes small intracellular ice crystals which do not disrupt the muscle cells as much as larger crystals. However, the increased disruption of muscle tissue with slower freezing may result in greater fluid loss from the thawed and cooked meat (Petrović et al., 1993; Luyet, 1964). On the other hand, it may result in

a decrease in shear force (Duckett et al., 1998). Longer duration of frozen storage also increases fluid loss compared with shorter duration (de Paula Paseto Fernandes et al., 2013). Therefore freezing and thawing in itself can cause disruptions in muscle structure that affect quality attributes. Frozen storage can be combined with storage under chilled conditions, which may also affect meat quality attributes, since the ageing time increases. Chilled storage could potentially be applied before or after freezing and may have an effect on e.g. tenderness (Coombs et al., 2017). The combined effect of chilled storage and freezing of meat needs to be studied further, to obtain more scientific evidence that can be applied to promote increased or stable meat quality attributes (Coombs et al., 2017)

2. Aims and objectives

The overall aim of this thesis was to investigate impacts of different factors in the production system for lamb meat and in subsequent handling of carcasses and meat on meat quality attributes. Specific objectives were to:

- Determine how the four most commonly used lamb production systems in Sweden affect live weight gain (LWG), carcass quality and meat quality. The hypotheses tested were that: 1) higher feeding intensity improves growth rate, and carcass and meat quality; and 2) concentrate allowance increases LWG, and thus carcass and meat quality (Paper I)

- Identify how different feeding strategies affect sensory attributes of lamb meat (Paper II)

- Investigate whether small-scale slaughter systems affect quality attributes in lamb meat compared with large-scale slaughter systems; and assess whether breeding for carcasses with a higher proportion of muscle and less fat can affect lamb tenderness scores over time (Paper III)

- Evaluate the combined effect of chilling and freezing storage, and determine whether frozen lamb meat displays differences in meat quality attributes compared with fresh meat. The hypothesis tested was that freezing leads to higher fluid loss, which influences meat quality (Paper IV)

3. Materials and methods

This chapter provides an overview of the materials and methods used in the different studies reported in Papers I-IV. For a more detailed description, see the respective papers.

3.1 Empirical studies

Papers I-IV are based on data obtained in two different studies, conducted in Sweden and Iceland. The Swedish study was carried out at SLU Götala Beef and Lamb Research Centre and at a private farm, both located near the city of Skara in southern Sweden (Papers I and II). The Icelandic study was performed at two different abattoirs in Iceland (Papers III and IV). The overall aim of the Swedish study was to investigate the effects of feeding and rearing system on carcass and meat quality (Papers I and II), while the overall aim of the Icelandic study was investigate the effect of slaughter method and storage regime on meat quality (Papers III and IV). Both studies involved intact male lambs used in domestic production in the two countries. Based on the results obtained, it was possible to investigate the effects on meat quality of different factors throughout the whole production chain from rearing of the living animal on the farm to cooked meat on the plate.

3.2 Animals, experimental design and housing

Paper I

In Paper I, 80 crossbred intact ram lambs (Dorset x Fine Wool) were included in the study. These lambs were either 50:50 (n=36) or 75:25 (n=44) Dorset:Fine Wool. Groups of 20 animals each were assigned to one of four production models for weaned male lambs. Group 1 lambs were kept on

indoor feeding; group 2 and 3 lambs were kept on cultivated pasture, with and without supplementary concentrate, respectively; and group 4 lambs were kept on semi-natural pasture. Group 1 lambs were housed indoors throughout and were fed a total mixed ration consisting of silage *ad libitum* and a constant amount of 0.8 kg concentrate per lamb and day. Group 2 and 3 lambs grazed two different enclosed grass-clover leys of in total 1.0 ha. In addition to pasture, Group 2 lambs were given 0.3 kg of concentrate per lamb and day. Group 4 grazed a semi-natural pasture. All lambs had free access to water, salt and minerals. The chemical composition of the silage and pastures in terms of metabolisable energy (MJ kg^{-1} dry matter, DM) was 11.4 (group 1), 11.5 (group 2), 10.4 (group 3) and 11.6 (group 4), while the digestible protein content was (g kg^{-1} DM) 126.3 (group 1), 140.0 (group 2), 126.5 (group 3) and 179.8 (group 4).

Paper II

In Paper II, 32 crossbred intact ram lambs from the four production models compared in Paper I were included. The feeding and rearing process in Paper II were identical to those in Paper I.

Paper III

In Paper III, 10 pairs of twin intact male lambs from the same producer were included in the study. The breed used was the native Icelandic sheep breed and the lambs were on average 160 days old at slaughter. All lambs were kept on the same type of pasture throughout the grazing season before slaughter. The lambs were slaughtered at two different abattoirs (one large-scale, one small-scale), with one of each pair of twins slaughtered at each abattoir. The animals at both abattoirs were kept in lairage for the same amount of time (10-12 hours) before slaughter. After slaughter, the large-scale abattoir applied electrical carcass stimulation (10 A, 80 V for 60 s) and the carcasses were then chilled at 2-4°C before sampling at 30 hours after slaughter. The small-scale abattoir used captive bolt stunning and chilled the carcasses in two stages, first at 10-15°C for six hours followed by 3-4°C until sampling 30 hours after slaughter.

Paper IV

Paper IV compared fresh and frozen meat samples from the lambs in Paper III. The right *Musculus longissimus thoracis et lumborum* (LTL) from the

carcasses was used to investigate the effect of freezing on quality attributes of the meat. The frozen samples were subjected to two different treatments that followed the ordinary procedures at each abattoir and the different slaughter methods used in Paper III. Samples from the large-scale abattoir were frozen on the day after slaughter and thereafter kept in frozen storage at -24°C for three months. The procedure at the small-scale abattoir was to age the samples for four days at 2°C before frozen storage at -24°C for three months.

3.3 Data collection and analysis

Paper I

Silage and pasture samples fed to the lambs in Paper I were sampled daily during the whole study period and later pooled to one feed sample per feed treatment before being sent for analysis. All animals were weighed once a week, to determine the average daily weight gain throughout the rearing period. At slaughter, slaughter weight, carcass conformation and fatness were assessed. Over the 24 h following slaughter, pH and temperature were recorded every 10 min, using pH and temperature loggers inserted in the LTL muscle. Meat samples from 32 animals (the first eight animals slaughtered in each of the four treatment groups) were collected after six days of ageing (Achilles tendon-suspended, 4°C). Meat samples, all from the LTL muscle on the right side of the carcass, were immediately frozen and stored at -20°C until analysis for colour, thawing loss, cooking loss and instrumental tenderness (WBSF).

Paper II

In Paper II, samples of 32 LTL from the left side of the carcass of lambs in the Swedish study were thawed and cooked using the sous vide method to an internal temperature of 65.5 ± 1.2°C. The samples were then chilled overnight at 4°C. On the next day, the samples were sliced into 5-mm slices and held at 70°C for 10 minutes before being used in sensory analysis. Sensory analysis was carried out on five testing occasions, by a trained panel consisting of six panellists.

Paper III
In Paper III, fat and conformation scores and carcass weight data were collected at both abattoirs on the day of slaughter. pH and temperature loggers were used to record data 24 hours after slaughter. Fresh meat samples (left LTL), aged for 6-7 days, were then analysed for meat colour before being cooked sous vide for one hour at 68°C, followed by further analysis of cooking loss, WBSF and sensory attributes. Sensory analysis was performed by a trained panel (6-10 panellists) on five test occasions.

Paper IV
The frozen lamb meat assessed in Paper IV was thawed overnight at 4°C before being analysed for fluid loss, colour, WBSF, LF-NMR and sensory attributes. Colour and LF-NMR determination was carried out on uncooked meat, whereas WBSF, sensory analysis and combined fluid losses were determined on cooked meat. The cooking process and further analysis was carried out in the same way as in Paper III.

3.4 Statistical analyses

The data obtained were analysed in general using Proc Mixed in Statistical Analysis Software (SAS 9.4) (Papers I-IV). A general Satterwaite approximation for the denominator degrees of freedom was performed, using the SATTERTH option in SAS (Papers I-IV). Differences were considered significant at $p \leq 0.05$ and a tendency for significance was assumed at $0.05 < p \leq 0.10$. In Paper I, two statistical models were used to analyse differences, where production system, with four sub-classes, was used as a fixed effect in the models and, for technological meat quality, day of analysis was used as random effect. In Paper II, sensory data were analysed with production system and assessor as random factors. In Paper III, the data were analysed with slaughter system as fixed effect and twin lamb pair as random effect. In Paper IV, analysis of technological meat quality and sensory data was performed with twin lamb pair as random effect, slaughter system as fixed effect, sample treatment (fresh/frozen) as fixed effect and also an interactive effect between slaughter system and sample treatment. The relaxation data were maximum-normalised prior to further analysis. The normalised data were also analysed by principal component analysis (PCA) using the Unscrambler X® software, as described by Pedersen et al. (2002), to look

for similarities between variables and differences between treatments. Data were centred and weighted with the inverse of the standard deviation (SD) of each variable before analysis.

4. Summary of results

4.1 Paper I

Pasture height as an average over the study period was lower for group 4 lambs reared on semi-natural pasture (5.6 cm) than for group 2 lambs (9.5 cm) and group 3 lambs (9.2 cm) reared on cultivated pasture. Live weight gain over the study period was significantly affected by production system ($p<0.001$) and followed the same order as feeding intensity, with group 1 lambs having the highest LWG (377 g/day) followed by lambs from group 2 (287 g/day), group 3 (244 g/day) and group 4 (211 g/day). Age at slaughter was therefore also affected by LWG ($p<0.001$) and ranged from 149 days for group 1 lambs to 167, 177 and 194 days for group 2, 3 and 4 lambs, respectively. There were significant differences in carcass weight between the groups ($p<0.001$), with group 4 lambs having lower carcass weight than lambs in the other groups. Dressing percentage ($p<0.001$), carcass conformation ($p=0.011$) and fat score ($p=0.039$) were also significantly affected by production system, where group 4 had the lowest scores and lowest dressing percentage. There were no differences in pH after 24 hours, pH after 6 days, thawing loss, cooking loss, meat colour or tenderness measured as Warner-Bratzler shear force between lambs from the different production systems.

4.2 Paper II

Significant effects of production system on sensory attributes were observed for three meat attributes: resistance to cutting ($p=0.0323$), hay odour ($p=0.051$) and leafy flavour ($p=0.0777$). Resistance to cutting was scored

higher for meat from group 2 lambs (43) compared with meat from lambs in the other groups (group 1 score 37, group 3 score 34, group 4 score 39). A tendency for significant differences was found for both hay odour and leafy flavour. Hay odour was scored higher for meat from group 4 lambs (33) compared with meat from lambs in the other groups (group 1 score 30, group 2 score 29, group 3 score 29). Leafy flavour followed the same pattern, with scores for group 4 lamb meat (35) being higher than those for meat from the other groups of lambs (31, 32 and 22 for group 1, 2 and 3, respectively).

4.3 Paper III

Fatness scores differed between the two slaughter systems ($p=0.0002$), where the small-scale abattoir received higher scores (3.5) for fatness than the large-scale abattoir (2.3). Significant differences were also found for lightness (L*) ($p=0.0073$), where meat from the large-scale abattoir had higher values (38.3) than meat from the small-scale abattoir (36.5), and for the sensory attribute colour appearance ($p=0.0089$), where meat from the small-scale abattoir was darker (higher score, 33) than meat from the large-scale abattoir (score 30). Fatty flavour was also scored differently for the two slaughter systems ($p=0.0370$), with meat samples from the small-scale abattoir receiving a higher score (18) compared with meat from the large-scale abattoir (15).

4.4 Paper IV

A tendency for a significant difference ($p=0.0654$) between fresh and frozen meat was found for lightness (L*), where fresh meat was lighter (37.5) than frozen meat (36.7). A difference in redness (a*) was also found ($p=0.0080$), with fresh meat having higher redness scores (19.5) compared with frozen meat (18.8). The opposite was found for yellowness (b*), where the fresh meat had lower yellowness score (4.5) ($p=0.0006$) compared with frozen meat (6.0). Total fluid losses (thawing loss + cooking loss) were greater ($p<0.0001$) for frozen meat (27%) compared with fresh meat (15%). This difference in fluid loss was reflected in measured relaxation time for different water populations within the meat samples. Transverse relaxation time differed for two of the water populations tested (T21 and T22; see Paper IV), where the relaxation time was higher for both T21-fresh (48.7 ms) and T22-

fresh (224.1 ms) compared with T21-frozen (41.0 ms) and T21-frozen (105.5 ms). An increase in the water population A22, corresponding to an increased proportion of water within the extra-myofibrillar space, was found for the frozen meat samples compared with the fresh samples. There was also a tendency for a significant difference in shear force (WBSF) (p=0.0837), with higher values (indicating greater toughness) found in the frozen meat (51 N) compared with the fresh meat (46 N).

Interactive effects between slaughter systems and meat treatment (fresh/frozen) were found for both redness (a*) (p=0.0011) and WBSF (p=0.0269). For samples from the small-scale abattoir, redness was higher in frozen meat (19.0) compared with fresh meat (18.8), while for samples from the large-scale abattoir a decrease in redness was seen in frozen meat (18.5) compared with fresh meat (20.2). For WBSF, a decrease was found in frozen meat (47.6 N) compared with fresh meat (49.0 N) from the small-scale abattoir, but an increase was found in frozen meat (53.7 N) compared with fresh meat (43.4 N) from the large-scale abattoir.

For sensory attributes, differences were found for fatty odour (p=0.0229), frying flavour (p=0.0340), sour flavour (p=0.0204), fatty flavour (p=0.0003), liver flavour (p=0.0222) and juicy texture (p=0.0001). A tendency for a significant difference was found for mushy texture (p=0.0714). Fatty odour was scored higher in frozen meat (score 32) compared with fresh meat (score 27), as was frying flavour (fresh score 24, frozen score 29), sour flavour (fresh score 25, frozen score 31), fatty flavour (fresh score 16, frozen score 22) and liver flavour (fresh score 41, frozen score 45). Juicy texture was scored higher for fresh meat (score 49) compared with frozen meat (score 35) and there was a tendency for mushy texture to be scored higher in fresh (score 16) compared with frozen (score 14) meat.

5. General discussion

The vast majority of previous research involving male lambs has focused on wethers (castrated male lambs), which reflects the situation in lamb production worldwide. Hence, the studies described in Papers I-IV in this thesis, which were conducted on intact male lambs, make a novel contribution to the existing knowledge base on lamb production. They also more accurately reflect the situation in the Nordic lamb production, where male lambs are commonly not castrated. For such countries, research and recommendations on lamb production should include the category of intact males, in order to optimise meat quality attributes based on differences between intact and castrated male lambs.

5.1 pH_{24} value as an indicator of meat quality

The applicability of using carcass pH measured 24 hours after slaughter (pH_{24}) as an indicator of potential quality problems or negative effect on tenderness was verified in this thesis. However, the results obtained in Papers I-IV questioned the validity of discarding meat with measured values above the recommended pH_{24} threshold of 5.7 (MLA, 2019b) or 5.4-5.8 (Alliance Group Limited, 2010) because of a higher risk of meat quality problems. For example, the pH_{24} values in meat from the Swedish lambs studied in Papers I and II (5.77 and 5.83, respectively) were both higher than the threshold set by Meat and Livestock Australia (MLA), but this did not result in any differences between meat samples from the different production systems in terms of elevated WBSF values, i.e. greater meat toughness. The pH_{24} values in Paper I did not differ significantly between the rearing treatments, due to large variation between lambs within groups 1-4, but the numerical difference was compared against recommended thresholds.

An interesting finding on comparing all results (Papers I-IV) was that WBSF values for meat samples from the Icelandic study were higher (49 N and 43 N for the small-scale and large-scale slaughter systems, respectively, and 46 N and 51 N for fresh and frozen meat, respectively) than those for meat samples from the Swedish study (34 N, 46 N, 32 N and 35 N for meat from lambs in group 1, 2, 3, and 4, respectively). This was despite the fact that pH_{24} was lower for the Icelandic fresh and frozen meat (5.65 and 5.63, respectively) (Papers III and IV) compared with meat from lambs in group 1, 2, 3, and 4 in the Swedish study (5.83, 5.66, 5.77 and 5.59, respectively) (Papers I and II). Therefore it is likely other factors within each system also affected WBSF and that pH_{24} may not be solely responsible for tenderness development in muscle.

5.2 Critical thresholds for pH_{24} and WBSF as indicators of meat quality

Based on previous statements, it is clear that pH should be used as an indicator of meat quality, but not as the only determinant of quality. The pH decline in muscle is very important and should be acknowledged as such, but the findings in this thesis showing that final pH or pH_{24} may differ depending on differences in the rearing, slaughter and/or meat storage system used for lamb meat should be taken into consideration.

With regard to meat tenderness and considering the WBSF results presented earlier for lamb meat for all treatments analysed in this thesis, previous studies have tried to establish thresholds for when meat is considered as tender or not. However, tenderness is a parameter that is influenced by other meat attributes, *e.g*. juiciness and fat content, and by consumer opinion, and it is therefore rather difficult to formulate recommendations that take into account all possible conditions. A WBSF limit of 49 N to classify meat as acceptably tender has been suggested (Shorthose *et al.*, 1986), but Huffman *et al.* (1996) concluded that 40.2 N is the upper limit when evaluating consumer satisfaction of beef. A more recent recommendation for lamb meat in Australia sets a limit of approximately 29.4 N or less (MLA, 2019a). This supports findings by Miller *et al.* (2001), who suggested a threshold of below 29.4 N for 100% consumer acceptability of beef. Miller *et al.* (2001) proposed an interval of 29.4 N to 45.1 N to describe the transition from slightly tender meat to slightly tough meat (with

93% consumer acceptability), and a threshold for tough meat of above 45.1 N. In light of the differences between all these previously reported thresholds for meat tenderness, it can be concluded that more research is needed to obtain consistent results on what should be classified as tender lamb meat, if such classification is important.

5.3 Changes over time in critical thresholds for WBSF

Another interesting reflection based on the large variation in recommended values is that the concept of 'tender meat' may have changed over time, since the more recently suggested recommendations or thresholds for tender meat set lower WBSF values than older recommendations. This may be due to different circumstances in different studies, but one explanation could be that tenderness today is a topic of conversation and an important attribute when considering the concept of total or overall meat quality. Another explanation could be that the increase in living standards has resulted in higher expectations on quality for all kinds of products, including meat. For example, a study by Mioche *et al.* (2002) found that meat samples which were chewed and assessed as being ready to swallow had a shear force value of 23.2 N. Comparing this 23.2 N for swallowing with the recommendation by MLA (2019a) and Miller *et al.* (2001) of below 29.4 N for tender meat implies that this type of consumer would theoretically consume meat that needed little to no chewing. When meat is chewed in the mouth aroma compounds are liberated, which increases the ability of the consumer to experience different flavours and textures. Therefore, a consumer prioritising tenderness and ease of mastication may not appreciate the flavour attributes of meat. To extend this concept even further, Warner *et al.* (2021) suggested that a single cut-off value for meat tenderness is no longer appropriate, due to different reasons relating to variations in samples from different muscles and different conditions, consumer variations, consumer preferences, differences in WBSF measurements and also the fact that consumers are not all solely interested in tenderness when consuming meat. To conclude, tenderness is influenced by multiple attributes and it may not be optimal to establish recommendations on WBSF, but until there is a better system to use it may still be the best way forward.

5.4 Measurement of pH

As regards measurements of pH, it could also be debated whether 24 hours after slaughter is the optimal time for measurement of final pH to predict tenderness of meat. The results in Paper I clearly showed that pH in muscle had decreased further six days after slaughter, resulting in meat from lambs in all groups (1-4) having pH values below the current recommendations. It would therefore be highly interesting to further evaluate the possibility of accepting higher pH_{24} values than that recommended today, if it could be shown that the further decrease in pH after 24 hours results in the same "final" tenderness of the meat to suit consumer and market demands. The practical dilemma of when to measure pH could thus be questioned, since carcasses or meat cuts are present at the abattoir for a limited amount of time and measurements after six days may be impossible in practice. Another complicated issue relating to pH is the actual measurement procedure and interpretation of results. The actual measured value can vary widely depending on the equipment used, probe sensitivity and the skill and experience of the individual performing the measurements in interpreting the values obtained. Since the measurement in itself is sensitive, the interpretation of results could be an area for improvement. However, until a better solution is established, this type of measurement is still the best to use for indicating meat quality problems related to pH decline in muscle.

5.5 Effect of freezing on meat quality

Paper IV examined the effect of freezing on meat quality. It is important to note that meat samples in Papers I and II were frozen and thawed prior to analysis, so effects of freezing on meat quality detected in Paper IV may also apply to the results presented in those papers. For example, when comparing the total fluid losses from meat samples, it can be assumed that freezing had an effect on fluid losses from meat from the Swedish lambs analysed in Papers I and II. The combined losses of fluid during thawing and cooking for meat from lambs in the four different rearing groups were: group 1 28.6%, group 2 28.9%, group 3 26.9% and group 4 29.2%. Fluid losses from fresh meat from Icelandic lambs in Paper III were 14% (small-scale abattoir) and 16% (large-scale abattoir), while fluid losses from fresh and frozen meat in Paper IV were 15% and 27%, respectively. Based on the comparison of fresh and frozen lamb meat in Papers III and IV, it can be deduced that freezing

also had an impact on fluid losses from meat samples in Paper I. The reason for freezing the samples in Papers I and II was that the study design was adapted to suit the circumstances prevailing at the time, meat analyses were performed elsewhere and the animals were not slaughtered all at once. Since day of analysis might have affected the parameters analysed, meat samples were stored frozen to allow samples from lambs in the different rearing groups to be analysed at the same time, and not in the order of when animals were slaughtered. This approach allowed samples from lambs in the different rearing groups to be equally divided over each day of analysis, to even out the risk of differences relating to that. Overall, however, the effect of freezing on meat quality attributes in Papers I and II may not have been of high importance, since the aim was to evaluate differences between the treatment groups and since all meat samples were treated in the same way. The effect of freezing should thus have been equal and differences detected should be an effect of treatment.

Freezing could be a positive solution for providing consumers in the Nordic region with lamb meat all year around. The potential negative effects of freezing seen in this thesis, with increased fluid losses, decreased scores for juiciness and increased intensity for many sensory attributes, could be regarded as a problem. However, it could also be possible to market those particular products to consumers who appreciate that specific type of sensory sensation and meat texture obtained and who use an appropriate cooking method for such meat. Freezing could also be used to minimise food wastage or to stabilise the market price and availability of meat over a longer period. To understand potential opportunities and threats raised by this meat preservation method if adapted commercially to a higher degree, consumer acceptance of frozen lamb meat should be evaluated in future studies.

5.6 Differences in consumer preferences and sensory panel assessments

Studies comparing the results of sensory testing of meat in different countries have demonstrated that consumer preferences for meat from animals fed different diets may vary between individual countries, or that clusters of countries may have similar preferences for a certain type of sensory profile (Font i Furnols *et al.*, 2009). The sensory testing performed on meat from Swedish lambs (Paper II) and Icelandic lambs (Papers III and IV) in this

thesis was carried out by two different trained panels. This complicated direct comparison of results, since the scoring of attributes by the panellists differed slightly between the Icelandic and Swedish study, as it depended on how the different panels described different attributes. It would be easier to compare the results of different studies if sensory attributes were described according to a standard formula. With the present system, it can be difficult to know whether similar attributes, such as grassy flavour, hay flavour or leafy flavour, relate to the same flavour attribute or not. This concept may affect the training stage performed by panellists, since they distinguish these sensory attributes during training on the test samples and naming of attributes is carried out based on consensus of all assessors in the panel. It is also important to acknowledge that this type of sensory testing does not give results about preferred samples based on likability or preference. Rather, it gives an evaluation of meat samples based on the attributes detected by the panellists during the training sessions and then scored in the test sessions for intensity or other pre-defined scales. This approach is also important when considering differences in consumer acceptability of different sensory profiles or meat quality attributes, as different consumers appreciate different attributes and also rate the same attribute differently. For example, Glitsch (2000) assessed differences in consumer perceptions of beef meat based on results from a consumer survey in six European countries and found that differences occurred in the ranking of different eating quality attributes. For example, tenderness was ranked first in some countries, but in others flavour was the most important attribute of beef (Glitsch, 2000). Hence, consumer perception differs and it is therefore important to understand how different sensory attributes and other attributes are affected by different factors in the production system, in order to provide the right type of quality for a specific consumer.

This thesis showed that differences in the sensory profile of meat may also arise depending on different feeding strategies, slaughter methods and freezing of meat. When this topic is raised with consumers and producers, they are usually convinced that feeding must have a great effect on the sensory outcome of meat. However, from the results obtained in Paper II, it is clear that some sensory attributes were affected by feeding and rearing, but the majority of attributes were not. The major effect of feeding seen was on more pasture-influenced attributes (odour and flavour), and not on more species-specific attributes of lamb meat. These results are interesting since

they refer to intact male lambs reared in four systems with different feeding intensities and therefore slaughtered at different ages. Previous literature suggests that intact male lambs of a specific slaughter age may have elevated levels of species-specific attributes compared with castrates slaughtered at the same age (210 days) (Sutherland & Ames, 1996). That was not the case in Paper II, where only pasture-specific odour and flavour attributes differed between the treatment groups. The effect of slaughter age of intact male lambs on sensory attributes, especially species-specific attributes, needs to be investigated further. It would also be interesting to analyse the composition of both cultivated pastures and semi-natural pasture in terms of species diversity and relate any differences to sensory attributes. Based on visual assessments in the field, species composition differed between the pastures used in Papers I and II. In particular, the semi-natural pasture contained different herbs and plant species that were not present in the cultivated grass-clover pastures.

One characteristic of meat is thus that consumers may differ in terms of the sensory attributes that they consider most important and also in terms of non-favourable attributes (Sañudo *et al.*, 2007). The sensory evaluation performed in Paper II did not detect many differences in sensory attributes, which could be interpreted as a positive finding considering that the sensory attributes of lamb meat may differ depending on farm conditions, such as geographical location and the types of pastures that are available. Previous studies have found that variations in carcass pH may cause changes to the sensory profile (flavours and odours) of cooked lamb meat (Young & Braggins, 1996), but this was not observed in the studies reported in this thesis. Hence, it can be suggested that changes in the sensory attributes hay odour, resistance to cutting and leafy flavour are not explained by changes in pH (non-significant results), but rather are connected to the composition of the diet. Lamb production, even within a country, may differ and the potential to produce lamb meat in different production systems without risking major differences in the sensory profile of the meat is an important finding in this thesis. However, the results are based on only one study under specific circumstances and further trials are needed to enable stronger conclusions to be drawn.

5.7 Consumer perceptions and prejudices

An important consumer perspective that should never be overlooked is the meat quality attributes that consumers consider most important. It has been suggested in previous literature that tenderness is the most important factor from a consumer perspective when eating beef (McIlveen & Buchanan, 2001). This is not the case with lamb meat, where flavour is reported to be the most important factor from a consumer perspective (Pethick *et al.*, 2006a). It is therefore very important to acknowledge the sensory profile of meat tested and not only focus on the technological parameters, in order to obtain a full picture of meat quality. The suggestion that flavour is the most important attribute may be connected to unjustified expectations of off-flavours in lamb meat, and it might not be the most important attribute if no off-flavours are expected (Risvik, 1994). It may also be connected to fear of experiencing more species-specific flavours that are not necessarily described as off-flavours, but which consumers are hesitant about. These attributes include *e.g.* lamb meat flavour or lamb meat odour, which may be connected to a prejudice that lamb tastes like 'woolly sweater' among consumers with no experience of eating lamb meat. This hesitancy or fear of specific flavour expressions may cause consumers to avoid lamb meat. Tenderness is still an important attribute, but the anxiety about experiencing off-flavours when eating lamb meat may make flavour the most important attribute.

5.8 Combination of technological analysis and sensory evaluation of meat

Technological and sensory analyses were both included in Papers I-IV in this thesis, in order to get a broader picture of how meat quality was affected by the different treatments compared. Technological meat quality and sensory attributes were both informative and important when analysing meat quality. The ideal situation when analysing the technological attributes of meat with the help of instruments is that the result is linked in some way to the eating experience (sensory profile). The results from the technological quality evaluations in Paper IV were reflected in the results of the sensory evaluation in terms of *e.g.* fluid loss and the sensory attribute juicy texture. The increased fluid loss as an effect of freezing was associated with lower scores for the texture attribute juicy in sensory evaluation of the frozen and thawed

meat. Other results from the technological evaluations were not reflected in the results from the sensory evaluation, *e.g*. in Papers I and II WBSF values and the sensory attribute tenderness did not follow the same pattern. This discrepancy has been reported previously by *e.g*. Oltra *et al.* (2015) and Sañudo *et al.* (1996) when comparing results for WBSF and sensory tenderness. It can therefore be concluded that both technological analysis and sensory evaluation should be applied to meat samples in order to obtain as much information as possible about specific meat quality characteristics.

Since consumer preferences also differ depending on *e.g*. culture and origin (Font-i-Furnols & Guerrero, 2014), a single worldwide threshold for WBSF value may not be optimal, as discussed previously. Consumer preferences may also vary within a country and result in different consumer segments with different preferences (Gracia & de-Magistris, 2013). While the technological measurements established for WBSF, meat colour and fluid loss can be used to set recommendations on acceptable meat quality, using a trained sensory panel to test meat for specific attributes will produce results that complement the technological measurements and give broader understanding of the overall quality of specific meat samples. Complementing WBSF values with scores for sensory tenderness and resistance to cutting permits evaluation of the instrumental method and helps understand how sensory testing can be refined to optimise the accuracy of the evaluation. Comparison of the results from the sensory evaluation and technological testing of meat quality in Papers I-IV in this thesis revealed that even significant differences in some parameters may not be important from a consumer point of view. For example, comparison of differences in colour measurements (L*, a* and b*) with the sensory attribute colour appearance showed that measured colour differences were not detected by the trained consumer panel. This indicates that the small numerical difference between samples was not large enough to cause a difference detectable with the human eye. Luo *et al.* (2001) tested this by placing ellipses on the so-called chromatic diagram used to represent the CIELAB colour space and found that the possibility of the human eye detecting a difference in colour depended on colour saturation and specific colour, for example the red nuance of the different samples. With this in mind, it is therefore difficult to propose an acceptable range for L*, a* and b*, since the combined result of all three also affects the outcome. The results presented in this thesis thus refer to detectable differences when L*, a* and b* were

compared to colour attributes evaluated by the sensory panel. Future studies involving both technological meat colour (L*, a* and b*) and sensory evaluation could seek to identify the acceptable range for meat colour based on consumer preference. In a study by Mancini *et al.* (2022) in which a trained panel performed a Farnswort-Munsell 100-Hue test, the results showed that at a set illumination expressed as A, the panellists who passed the test were able to discriminate a change of 0.95 in a* and 0.9 in b*.

In Papers III and IV, the attribute colour appearance was evaluated by the panel, rather than separate evaluations of redness and yellowness. Thus the interpretation of increased redness for some samples analysed in Papers III and IV could also reflect darker appearance of the meat. Together with the decreased L* value found in Paper III, it is therefore suggested that the numerical increase of 1.4 in a* from the small-scale abattoir (a* = 18.8) to the large-scale abattoir (a* = 20.2) contributed to the difference in colour appearance reported by the sensory panellists, even though differences in a* between treatments were non-significant. The differences in redness (0.7) in Paper IV were therefore too small to cause a difference in assessed meat colour. This is also supported by the results in Paper I, where the maximum variation in a* between groups 1 and 4 was 0.5 and there were no differences in the sensory attribute pinkness. However, the difference in yellowness (1.5) in Paper IV was large enough to be detected by visual assessment according to (Mancini *et al.*, 2022). This indicates that differences in meat colour may be very complex to detect, and that a combination of differences in lightness, redness and yellowness may cause a visual detectable difference, rather than differences in only one of the three colour attributes. It could also be debated whether redness has more influence over colour differences than yellowness, based on the results in Paper IV. In that study, no difference was found in sensory colour measurements even though the difference in yellowness should have been detectable by visual assessment and could therefore have caused a difference between samples. This difference was thus not reflected in the results. An important aspect to consider is that differences in meat colour may not be regarded as unfavourable from a consumer point of view, depending on personal preference and magnitude of the difference.

5.9 Quality variations in Swedish lamb meat

Variations in the quality of Swedish lamb meat could originate from the many different factors mentioned previously, providing many different opportunities for change. First, variability does not have to be a bad thing in meat quality. The problem in the case of Swedish lamb meat is that consumers or chefs at restaurants may prefer imported meat (larger quantities with similar quality) to domestically produced lamb meat (smaller quantities with variation in quality) (Carlsson & Arvidsson Segerkvist, 2018). It is important to bear in mind that domestic production not only provides meat from lambs that are reared, slaughtered and processed under Swedish regulations, but also provides other important benefits, such as job opportunities in rural areas, open landscapes and increasing biodiversity in pastures. One way to decrease potential negative differences in meat quality attributes such as tenderness is through use of electrical stimulation (Polidori *et al.*, 1999), since an effective method for decreasing differences in quality would result in a larger quantity of meat of the desired quality being available on the market. The results for two different slaughter systems compared in Paper III showed that all meat quality parameters for the electrically stimulated carcasses displayed less variation around the mean compared with the non-stimulated carcasses. A slaughter system with electrical stimulation of carcasses and fast chilling may therefore be more effective in decreasing the variation within the meat quality attributes tested (lightness, redness, yellowness, fluid losses and WBSF). Optimisation of such slaughter systems and meat quality attributes is a relevant issue which could be explored in future studies. The potential to use electrical stimulation as a tool to reduce the variability in the quality of domestic lamb meat should also be further evaluated.

6. Main conclusions

Many factors affect the quality attributes of lamb meat, and therefore each production system needs to be optimised according to its own specific conditions. Measured indicators of meat quality can be of assistance when trying to exclude or include meat of normal or problematic quality. The main conclusions drawn from the work in this thesis were as follows:

- Time of measurement and pH thresholds to be used in different systems for rearing slaughter lambs should be evaluated and optimised before implementation in practice.

- In order to promote adequate glycogen storage in muscles, thereby ensuring an adequate pH decline and high eating quality, lambs should be in positive energy balance (growth phase) at the time of slaughter.

- Live weight gain is influenced by increased feeding intensity, but increased LWG did not influence technological meat quality attributes.

- Intact male lambs can be reared in both extensive and intensive systems (indoor feeding, cultivated pastures and semi-natural pasture in this thesis) without risking differences in technological meat quality attributes. However, carcass quality can be affected by low LWG (semi-natural pasture), resulting in lower scores for conformation and fatness and lower carcass weight and dressing percentage.

- Differences in diet composition did not dramatically affect the sensory profile of lamb meat, however, the few differences found were due to diet and not species specific attributes.

- Technological tenderness, measured as WBSF values, confirmed that decreased tenderness scores are an effect of breeding for carcasses with more muscle and less fat.

- Slaughter method had only minor effects on sensory attributes such as colour appearance and fatty flavour. Differences in colour appearance detected by sensory panels were reflected in the results for measured colour of lightness (L*).

- Freezing increased fluid losses, and thus increased the scores given for many of the sensory attributes tested, meaning that the intensity of specific attributes often increased as an effect of frozen storage.

- Differences in e.g. instrumental meat colour (L*, a* and b*) may not be of practical importance if they are not detectable (visible to the human eye) in sensory evaluations by trained panellists. Thus comparison of technological attributes and sensory evaluation scores is important when scientifically analysing meat samples.

7. Future perspectives

This thesis identified some of the main factors affecting the quality attributes of lamb meat. To build upon the findings, future studies should:

- Evaluate whether Swedish pelt breeds (*e.g*. Gotland pelt) differ from common crossbreeds in terms of technological meat quality and sensory attributes.

- Further investigate sensory attributes of intact ram lambs of different slaughter ages and fed different diets.

- Investigate the potential advantages for meat quality attributes of using electrical stimulation of lamb carcasses in Swedish slaughter systems.

- Further investigate how to optimise slaughter systems and preservation methods to maintain meat quality throughout the whole process until cooking.

- Further investigate freezing of meat, to determine how to optimise quality attributes during preservation, thawing and cooking.

- Develop specific pH recommendations for Swedish lamb production, since this thesis showed that it may not be appropriate to adopt current established recommendations.

- Further investigate when to measure final pH, to get an appropriate indication of meat quality problems.

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Popular science summary

Annual consumption of lamb meat in Sweden is currently only 1.5 kg per person and domestic production of lamb meat covers only around 30% of current consumption, while the remaining 70% of all lamb meat consumed in Sweden is imported. There is thus great potential for expansion of the domestic lamb production sector, e.g. the 70% that is imported today could be produced locally in Sweden without increasing domestic consumption. In order for domestic production to increase, it is essential that production is profitable for farmers who would like to invest in rearing lambs and that the domestic market for lamb meat is stable.

An obstacle to expansion of the domestic sector is that previous studies have found that Swedish restaurants and consumers prefer to buy imported meat instead of locally produced Swedish lamb meat, citing varying quality of Swedish lamb meat as one reason. In order to improve lamb meat quality and reduce perceived variations in quality, this thesis sought to identify factors that influence important quality parameters of lamb meat produced in Nordic conditions. Different factors were analysed in two different studies which included comparisons of rearing systems (intensive to extensive), slaughtering of the animals (slaughter process) and the storage method used for meat (cold storage or freezing). Many different factors such as feeding with concentrate or pasture, stunning method, electrical stimulation of the carcass post-slaughter, cooling process and freezing/cold storage regime were investigated.

The major lamb-producing countries castrate virtually all ram lambs, which means that the majority of research results published to date refer to castrated ram lambs, while the Nordic countries do not castrate ram lambs. Therefore all analyses in this thesis were carried out on intact male lambs.

Meat quality can be measured in many different ways, a range of which were assessed in this thesis. For example, growth rate of the animal and post-slaughter changes in pH and temperature in the muscle over time are commonly used indicators of meat quality problems. The growth rate of an animal provides information about the energy status of the muscles in the carcass, e.g. if the animal's weight is increasing prior to slaughter there are probably energy reserves in the muscle in the form of glycogen. After slaughter, this glycogen is converted to lactic acid and, as lactic acid accumulates in the muscle, the pH drops. If there is not enough glycogen in the muscle before slaughter, not enough lactic acid is formed to provide a sufficient pH reduction, which means that the meat risks developing quality problems. For conversion of muscle into high-quality meat, this decline in pH in must reach an optimal threshold and also needs to occur at an optimal pace (assessed by measurement of muscle temperature) in order to promote tenderness in the meat. By weighing animals to detect potential negative energy balance in the muscle before slaughter and by measuring and monitoring the pH and temperature drop in the muscle after slaughter, possible meat quality problems can be predicted and avoided. Additional quality analyses of technological meat quality parameters, carried out with help of technical devices, are usually performed and include e.g. Warner-Bratzler shear force (WBSF), meat colour and total fluid losses during thawing and cooking the meat. Technological measurement of meat colour usually involves analysing different aspects of colour, e.g. lightness (L*), redness (a*) and yellowness (b*).

Sensory assessments, in which a trained expert panel evaluates meat samples based e.g. on odour, visual appearance, texture and flavour, were also carried out on individual lamb meat samples in this thesis. The attributes were graded by the panellists on a scale from 1 to 100, with higher scores usually associated with increased intensity of flavour, appearance or texture. The results revealed that feeding intensity (intensive/extensive) affected carcass characteristics, i.e. slaughter weight, dressing percentage, conformation and fatness scores, all of which are directly linked to the profitability of production. Rearing the lambs on semi-natural pasture resulted in slower growth, lower slaughter weight and lower carcass scores compared with more intensive rearing on cultivated pasture or indoors with concentrate feed. However, rearing did not affect post-slaughter pH in the muscle after 24 hours or after six days. The technological meat quality

characteristics analysed (thawing loss, cooking loss, meat colour and shear force) were also not affected by production system. There were only a few sensory differences between the production systems, e.g. resistance to cutting was highest for meat from lambs reared on cultivated pasture (grass-clover) and given supplementary concentrate. There was a tendency for a difference in hay odour and leafy flavour of the meat, where meat from the group of lambs reared on semi-natural pasture scored higher on both attributes than meat from lambs in the other groups.

A comparison between two slaughter methods, where one included electrical stimulation and rapid chilling of carcasses (abattoir A) and the other slower chilling and no electrical stimulation (abattoir B), showed that meat lightness (L*) and the sensory attribute colour were affected, with both indicating that meat from abattoir B was darker than meat from abattoir A. Freezing affected meat colour, fluid loss and WBSF values. Frozen meat tended to be darker than fresh meat and less red, but more yellow, than fresh meat. Fluid losses were much greater for frozen meat (27%) compared with fresh meat (15%). There was also a tendency for higher WBSF in frozen meat compared with fresh. Slaughter method affected the sensory attribute fatty flavour, with meat from abattoir B scoring higher than meat from abattoir A.

Overall, frozen storage was the treatment that affected sensory attributes most strongly, giving higher scores in the frozen meat for fatty odour, frying flavour, sour flavour, fatty flavour and liver flavour. The characteristics that gave higher scores in the fresh meat were juicy texture and a tendency for mushy texture.

In summary, there are many different factors that influence the different quality characteristics of lamb meat. Indicators of quality problems, such as pH and temperature in the muscle post-slaughter, are a good help in detecting quality problems, but specific thresholds need to be identified for these indicators in Nordic lamb production. To achieve the pH reduction that ensures high quality development in the meat, lambs sent to slaughter should be in a growth phase so that they have sufficient glycogen stores in their muscles. Technological and sensory analyses of lamb meat from different rearing models in this thesis indicated that intact male lambs can be reared under the conditions described without risking negative quality problems with the meat. Carcass quality was affected by rearing model, which indicates that it is important to optimise production system based on site-

specific farm conditions in order to get lambs within the best paid range in terms of fat and conformation scores of the carcass, but also to ensure that the carcass weight is sufficient to optimise the price per carcass. Breeding to produce carcasses with a greater proportion of muscle and less proportion of fat is perhaps not the best option to ensure good eating quality and eating experience, since over time there has been a reduction in tenderness as an effect of this breeding method. Freezing meat is a good method to increase the storage possibilities, but this thesis showed that it increases fluid losses during thawing and cooking and enhances the intensity of many measured sensory attributes, resulting in a real quality difference between fresh and frozen meat. However, statistically significant differences in meat parameters may not always be of high practical value, e.g. in some cases in this thesis significant differences in instrument-measured meat colour could not be detected by the sensory panel. It is therefore important to combine technological and sensory results in order to obtain a full picture of lamb meat quality.

Populärvetenskaplig sammanfattning

Den årliga konsumtionen av lammkött i Sverige är för närvarande 1,5 kg per person, och den inhemska produktionen av lammkött utgör idag endast omkring 30 % av konsumtionen. Det betyder att vi idag importerar 70 % av allt lammkött som konsumeras i Sverige. Dessa siffror kan ses både som ett smärre nederlag eller som en möjlighet till potentiell utveckling. De 70 % som vi idag importerar skulle kunna produceras lokalt i Sverige utan att öka den inhemska konsumtionen. För att den inhemska produktionen skall kunna öka krävs dock att produktionen är lönsam för de lantbruksföretag som vill satsa på lammproduktion. För att potentiellt öka den inhemska produktionen behövs dessutom en marknad som efterfrågar det som produceras.

Tidigare arbete har bland annat identifierat varierande kvalitet av lammkött som en bidragande orsak till att Svenska restauranger och konsumenter väljer bort det svenska lammköttet och istället väljer att köpa importerat lammkött. För att förbättra kvaliteten och förhoppningsvis minska dess variation har denna avhandling fokuserat på att identifiera faktorer som påverkar viktiga kvalitetsparametrar hos kött från intakta bagglamm som producerats under nordiska förhållanden. Olika faktorer har analyserats genom utförande av två olika studier som innefattat jämförelser av olika uppfödningssystem (intensiv till extensiv), slakt av djuren (slaktmetod) samt jämförelse av lagringsmetod för kött (kyllagring eller frysning). På detta sätt har många olika faktorer så som utfodring, bedövningsmetod, elstimulering av slaktkroppen, nedkylningsprocess samt kyllagring/frysning av kött undersökts.

De stora lammproducerande länderna kastrerar i stort sett alla bagglamm vilket medför att majoriteten av hittills publicerade forskningsresultat baseras på just kastrerade bagglamm. Då vi i de nordiska länderna har som

tradition att inte kastrera har därför alla studier i denna avhandling utförts på intakta bagglamm.

Köttkvalitet kan mätas på många olika sätt, denna avhandling inkluderar en variation av dessa möjliga metoder. Indikatorer för kvalitetsproblem är exempelvis djurets dagliga tillväxt samt pH- och temperatur sänkningen i muskeln efter slakt. Tillväxten hos ett djur kan ge oss en bild av musklernas energistatus, om djuret ökar i vikt innan slakt, finns troligtvis energireserver i muskeln i form av glykogen. Detta glykogen omvandlas efter slakt till mjölksyra som är sur (pH), och när mer och mer mjölksyra ansamlas i muskeln sjunker pH. Om det däremot inte finns tillräckligt med glykogen i muskeln innan slakt bildas inte tillräckligt med mjölksyra för att ge en tillräcklig pH sänkning vilket leder till att köttet riskerar att utveckla kvalitetsproblem. När muskeln omvandlas till kött behöver denna pH sänkning, förutom att nå optimal nivå, också ske i lagom takt (därav mätning av muskelns temperatur) för att gynna bland annat köttets mörhet. Genom att väga djuren för att se potentiell negativ energibalans i muskeln innan slakt samt att mäta och följa både pH och temperatursänkningen i muskeln efter slakt, kan eventuella köttkvalitetsproblem förutsägas och undvikas. Ytterligare kvalitetsanalyser som görs med hjälp av tekniska apparater brukar kallas för teknologiska köttkvalitetsparametrar och innefattar bland annat skärmotstånd, färg och vätskeförluster. Vätskeförluster kan analyseras och beskrivas på många olika sätt, ett av dem är att mäta den totala vätskeförlusten vid upptining och kokning av kött. Teknologisk mätning av köttets färg görs vanligtvis genom att analysera färgen i olika delar, exempelvis; ljusheten (L*), rödheten (a*) och gulheten (b*).

Sensoriska analyser, där en tränad expertpanel utvärderar köttprover genom att bland annat dofta, titta, skära och smaka, har också genomförts i de båda studierna. Attributen har graderats av den sensoriska panelen på en skala från 1 till100, där ökad poäng oftast är förknippat med ökad smakintensitet, färg eller mörhet. Sensoriska analyser i dessa studier har genomförts med hjälp av tränad sensorisk panel som utvärderat provernas egenskaper individuellt utan rangordning sinsemellan. Resultaten ifrån dessa studier visar att utfodringsintensiteten (intensiv/extensiv) påverkade slaktkroppsegenskaperna, dvs. slaktvikt, slaktutbyte, form- och fettklass, vilka alla är direkt kopplade till produktionens lönsamhet. Uppfödning på naturbete resulterade i lägre tillväxt, slaktvikt och klassning (konformation och fett) jämfört med mer intensiv uppfödning på åkermarsbete eller på stall.

Däremot påverkade uppfödningen inte pH i muskeln efter 24 timmar eller efter sex dagar. De teknologiska köttkvalitetsegenskaper som analyserades; upptiningssvinn, koksvinn, köttfärg och skärmotstånd, påverkades inte heller av uppfödningsmodell. De sensoriska skillnaderna mellan uppfödningsmodellerna var endast ett fåtal, skärmotstånd var högre för gruppen som gått på åkermarksbete och fått kraftfoder jämfört med de andra grupperna. En tendens till skillnad fanns för hödoft och lövig smak, där gruppen på naturbete fick högre poäng för båda dessa attribut jämfört mot de andra grupperna.

Vid en jämförelse mellan två slaktmetoder, där den ena inkluderade elstimulering och snabb nedkylning (slakteri A) och den andra långsammare nedkylning och ingen elstimulering (slakteri B) påverkades köttets ljushet (L*) samt sensoriska attribut för färg, där de båda visade att kött från slakteri B var mörkare än från slakteri A. Frysning påverkade köttets färg, uppmätta vätskeförlust samt skärmotstånd. Det frusna köttet hade en tendens till att vara mörkare än det färska köttet samt att det frysta var mindre rött men mer gult än det färska köttet. Vätskeförlusterna var mycket större i det frysta köttet (27 %) jämfört mot det färska (15 %). Det fanns även en tendens till högre skärmotstånd i det frysta köttet jämfört mot det färska. Slaktmetod påverkade som beskrivet ovan köttets färg men även attributet fet smak, där kött från slakteri B fick högre poäng än det från slakteri A.

Fryslagring var det som påverkat de sensoriska egenskaperna allra mest. Egenskaper som hade högre poäng för det frusna köttet var; fet doft, steksmak, sur smak, fet smak och smak av lever. De egenskaper som däremot gav högre poäng för det färska köttet var saftig textur samt en tendens för mer smulig textur.

Sammanfattningsvis så finns det många olika faktorer som påverkar köttets olika kvalitetsegenskaper. Indikatorer på kvalitetsproblem, så som pH- och temperatursänkning i muskeln är ett bra hjälpmedel för detektering av kvalitetsproblem. Vilka gränser som bör användas för dessa mätningar bör utredas ytterligare för att passa den nordiska lammproduktionen. För att åstadkomma den pH-sänkning som gynnar köttets kvalitetsutveckling positivt bör lamm som går till slakt vara i tillväxtfas så att deras glykogenförråd är tillräckliga vid slakt. Resultaten från de teknologiska och sensoriska analyserna av kött ifrån de olika uppfödningsmodellerna inkluderade i denna avhandling indikerar att intakta bagglamm kan födas upp under dessa förhållanden utan att riskera negativa kvalitetsproblem på köttet.

Slaktkroppskvaliteten påverkades av uppfödningsmodell, vilket tyder på att det är viktigt att optimera uppfödningsmodellen utifrån gårdens förutsättningar för att dels hamna inom det bäst betalda intervallet vad gäller fett och konformation av slaktkroppen men även se till att slaktkroppsvikten är tillräcklig för att optimera priset per slaktkropp. Avel för att få fram slaktkroppar med större andel muskel och mindre andel fett är kanske inte det bästa alternativet för att säkerställa en god ätkvalitet och ätupplevelse. Resultat över tid visar på minskad mörhet som en effekt av denna avelsmetod. Att frysa kött är en bra metod för att öka lagringsmöjligheterna för kött. Frysning gav dock ökade vätskeförluster vid upptining och kokning samt ökad intensitet för många av de uppmätta sensoriska attributen, vilket påvisar en reell kvalitetsskillnad mellan färskt och fruset kött. Att skillnader kan ses statistiskt är kanske inte alltid av lika högt praktiskt värde som rent teoretiskt. Så var fallet med de uppmätta skillnaderna i köttets färg där vissa signifikanta skillnader i uppmätt färg med instrument inte kunde detekteras av den sensoriska panelen. Det är därför viktigt att kombinera de teknologiska och sensoriska resultaten för att se vilka likheter och skillnader som finns.

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I

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Carcass characteristics and meat quality attributes in lambs reared indoors, on cultivated pasture, or on semi-natural pasture

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This study evaluated the effects of different lamb production systems on live weight gain (LWG), carcass quality and meat quality. Four production systems for weaned intact male lambs were examined: indoor feeding with grass silage and concentrate (group 1), grazing on cultivated pasture with (group 2) or without (group 3) concentrate, and grazing on semi-natural pasture (group 4). Live weight, carcass weight, dressing percentage, carcass conformation, fatness and pH decline were recorded at slaughter, and *M. longissimus thoracis et lumborum* was analysed for colour, thawing and cooking loss, pH after 24 hours and 6 days, and Warner-Bratzler shear force. LWG was strongly affected by production system, being highest for group 1 and lowest for group 4 (p<0.001). Group 4 had the lowest conformation (p=0.002) and fat scores (p<0.001). Hence, production system affected age at slaughter, live weight gain, weight at slaughter, carcass conformation and fatness scores, but caused no differences in meat quality attributes in intact male lambs.

Key words: production system, intact male lamb, live weight gain, pH, colour, Warner-Bratzler shear force

Introduction

Domestic lamb and sheep meat production was in 2019 5.090 tonne, which corresponds to 30.7% of Sweden's total consumption in 2019 (Lannhard Öberg 2020). This suggests that there is potential for expansion of Swedish lamb meat production. Rising demand for high-quality Swedish lamb means that data are needed on the optimal way to rear lamb under Swedish conditions, so as to produce meat with high and consistent eating quality. It is well known that the eating quality and size of valuable cuts of Swedish lamb currently vary more than those of imported lamb and sheep (Carlsson and Arvidsson Segerkvist 2018). Several factors may explain this variation, including some related to primary production such as production system, choice of breed and/or cross, age at slaughter and carcass weight. The Swedish lamb meat production is characterized by few large and many small farms, with a large verity of breeds and production systems (Carlsson and Arvidsson Segerkvist 2018, Lannhard Öberg 2020). The average herd size was 33 ewes and/or rams in 2019 (Jordbruksverket 2019). It has also been shown that feedstuffs and feeding regime can also affect lamb meat quality (Watkins et al. 2013). In particular, pasture and concentrate may have different effects on meat flavour (Fisher et al. 2000, Arsenos et al. 2002, Priolo et al. 2002, Resconi et al. 2009). Feeding strategy can also influence glycogen storage in muscles, which in turn affects post-mortem muscle metabolism and thereby meat quality. Glycogen in muscles is converted into lactic acid under anaerobic conditions after slaughter, which reduces the pH of the muscle tissue. Since glycogen serves as 'fuel' in this process, it is essential to ensure that glycogen storage in the muscles prior to slaughter is sufficient to enable an adequate decline in pH (Bendall 1973). The official recommendation in Australia is for crossbred lambs to gain 100–150 g day^{-1} in the last two weeks pre-slaughter, to ensure that the animals are in positive energy balance (growth phase) and have adequate glycogen depots in muscle (MSA 2015a). The effects of different feeding strategies on lamb meat quality can be evaluated by measuring the carcass pH, which is a useful indicator of various meat quality parameters. Specifically, the pH at 24 hours after slaughter (pH_{24}) is commonly used as an indicator of tenderness in meat (Geesink et al. 2000, Thompson et al. 2005, Toohey et al. 2006). Carcass pH (and thus meat quality) is sensitive to many factors pre-slaughter, during slaughter and post-slaughter, and can therefore vary between production systems (Sañudo et al. 1998). It is thus very important to understand how different production systems affect meat quality, in order to help producers deliver the consistent meat quality demanded by consumers.

The aim of this study was to determine how the four most commonly used lamb production systems in Sweden affect live weight gain (LWG), carcass quality and meat quality. The hypotheses tested were that: 1) higher feeding intensity improves growth rate and carcass and meat quality; and 2) concentrate allowance increases LWG, and thus carcass and meat quality.

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Material and methods

Animals and experimental design

The experiment was performed between 29 June and 26 October 2016, at SLU Götala Beef and Lamb Research, Swedish University of Agricultural Sciences (SLU), Skara, Sweden (58°42'N, 13°21'E) and at a private farm outside Skara, Sweden (58°20'N, 13°26'E). In total, 80 crossbred weaned intact ram lambs (Dorset x Fine Wool) were included in the study, 36 of which were 50:50 crosses and 44 were 75:25 Dorset:Fine Wool crosses. The experiment was approved by the Ethics Committee on Animal Experiments, Gothenburg, Sweden (Registration No. 53-2016). Immediately prior to the study, the lambs were weighed and divided into four groups of 20 individuals each, equally balanced by breed crosses. All lambs were weaned just prior to the study. Average live weight (26.4±2·7, 26.8±2·8, 26.4±3.1 and 26.0±2.7 kg for group 1, 2, 3 and 4, respectively) and age (85.25±6.0, 85.2±5.9, 84.45±5.3 and 84.4±6.0 days for group 1, 2, 3 and 4, respectively) were similar for the four groups at the start of the experiment. Each group was assigned a unique feeding treatment and followed that treatment throughout the experiment, with all groups starting treatment on 29 June. The treatments were: 1) indoor rearing; 2) cultivated pasture with a concentrate supplement daily; 3) only cultivated pasture; and 4) semi-natural pasture (Table 1). Group 4 was reared on the private farm, while the other three groups were reared at SLU Götala Beef and Lamb Research farm.

Table 1. Feeding strategies used for groups 1–4

Group	Treatment
Group 1, indoor	Silage *ad libitum* + 0.8 kg concentrate per lamb daily
Group 2, pasture	Cultivated pasture + 0.3 kg concentrate per lamb daily*
Group 3, pasture	Cultivated pasture with no concentrate
Group 4, pasture	Semi-natural pasture with no concentrate

*Group 2 received 0.4 kg of concentrate per lamb daily between 28 September and 4 October, due to poor pasture availability.

Experimental diets

Feed values and chemical composition of the experimental feeds are presented in Table 2. Group 1 was fed a diet consisting of silage *ad libitum* and 0.8 kg of a standard commercial concentrate (Fårfor Lamm 500, Lantmännen, Västerås, Sweden) per lamb per day to promote rapid growth. The seed mix for the silage consisted of 76% timothy (*Phleum pratense* L.), 18% red clover (*Trifolium pratense* L.) and 6% white clover (*T. repens* L.). The silage was harvested on 27–31 May, and was fertilised in late April with around 30 tonne ha^{-1} of cattle manure, providing 1.5 kg N $tonne^{-1}$. A commercial additive (mixture of formic acid and propionic acid) was added to the herbage before ensiling. The animals in group 1 had free access to water, salt and minerals, and were housed indoors in an enclosed pen made from metal gates and with wheat straw bedding.

Groups 2 and 3 were kept on cultivated pasture in two different enclosures of 1.0 ha, both were divided into three grazing paddocks of 0.3 ha each. Both groups had access to one of the three paddocks at a time and they were moved once a week. Groups 2 and 3 differed in feeding intensity (Table 1), but both groups had daily access to a salt and mineral block and were given free access to water in a tub in each paddock. The seed mix for the cultivated pastures consisted of 50% timothy, 20% meadow fescue (*Festuca pratensis* Huds.), 15% perennial ryegrass (*Lolium perenne* L.), 10% red clover and 5% white clover. The cultivated pasture for groups 2 and 3 was fertilised on 3 April with 250 kg of Axan (Yara AB, Malmö, Sweden) (total nitrogen 27.0%, nitrate nitrogen 13.5%, magnesium 0.4%, sulphur 3.7% and calcium 6.0%) and then on 5 April with approximately 30 tonne ha^{-1} of cattle manure (1.5 kg N $tonne^{-1}$). The forage was harvested on 10 June. The regrowth was not fertilised after harvesting.

Group 4 was kept on unimproved semi-natural pasture (Table 2) with daily access to salt and minerals. The semi-natural pasture contained trees and shrubs and was on hilly land. This group was moved to new pastures on three occasions (31 August, 8 September, 11 October) due to poor quality and growth of the pasture late in the season. Lambs on semi-natural pasture had free access to water in a pond, stream or tub.

Due to illness, two lambs were removed from the experiment, one from group 1 and one from group 2. Data on these animals were excluded from further analyses, so the results are based on data for 78 lambs in total.

Table 2. Chemical composition and feeding values of the experimental feeds

	Group 1	Group 2	Group 3	Group 4	Groups 1+2
	Silage	Cultivated pasture	Cultivated pasture	Semi-natural pasture	Concentrate
n	3	4	4	4	1
Dry matter, g kg^{-1}	297 (31)[1]	233 (33)	228 (22)	223 (39)	878
Crude protein, g kg^{-1}	168 (16.2)	183 (29.3)	168 (16.8)	198 (28.6)	185
Digestible protein, g kg^{-1}	126 (15.0)	140 (27.6)	127 (15.8)	180 (69.6)	-
Metabolisable energy, MJ kg^{-1} DM	11 (0.3)	12 (0.4)	10 (0.5)	12 (0.3)	13
NDF, g kg^{-1}	507 (16.6)	421 (40.5)	453 (10.7)	396 (31.4)	278
Ash, g kg^{-1}	70 (3.5)	90 (1.7)	83 (8.5)	86 (2.5)	81
Crude fat, g kg^{-1}	-	-	-	-	48[2]
Crude fibre	-	-	-	-	91[2]
Starch, g kg^{-1}	-	-	-	-	238[2]
AAT, g kg^{-1}	-	-	-	-	130[2]
PBV[5], g kg^{-1}	-	-	-	-	20[2]

Group 1 = indoor feeding with silage; Group 2 = cultivated pasture with supplemented concentrate; Group 3 = cultivated pasture only; Group 4 = semi natural pasture; [1] = Standard deviation in brackets; NDF=neutral detergent fibre; AAT=amino acids absorbed in the duodenum; PBV=protein balance in the rumen; [2] = Values from feed producer

Special treatments during the experiment

All animals were dewormed (ivermectin, 0.8 mg ml^{-1}) at the start of the experiment. All animals other than those in group 1 were dewormed again after four weeks. Animals in group 4 were dewormed a third time, six weeks after the second treatment.

Feed sampling and analyses

Throughout the experiment, silage samples were collected daily and pasture samples were taken once a week and stored at –20 °C until analysis. Pasture samples were cut with a handheld machine, along a W-shaped route in each pasture according to Frame (1993). Before analysis, samples were pooled to obtain representative samples for consecutive 4-week periods. For mineral analysis, samples were pooled to obtain a representative sample for the whole experimental period. The height of the cultivated pastures was measured each week in conjunction with weighing the lambs, immediately before the lambs were released into a new paddock. Sward height was measured according to Frame (1993), following the W-shaped route, with a rising plate meter (0.3 × 0.3 m, weight 430 g). The same sampling procedure was used for the semi-natural pasture, although the lambs in this group were not moved to a new pasture each week.

Crude protein was analysed according to Dumas (1831) and digestible protein levels were calculated using the digestibility coefficient of Spörndly (2003). Ash content was analysed by combustion at 525 °C. Dry matter (DM) measurement was performed by drying samples at 60 °C for 16 hours and then at 130 °C overnight. Neutral detergent fibre (NDF) was analysed as described by Chai and Udén (1998), using 100% neutral detergent solution, amylase and sulphite. Metabolisable energy (ME) was determined by incubation in rumen fluid and buffer for 96 hours (Lindgren 1979) and then calculating the ME concentration based on *in vitro* disappearance of rumen organic matter, as described by Lindgren (1983).

Weighing and body condition scoring of the lambs

All lambs were weighed once a week on a portable scale (Iconix 21, Iconix New Zealand Ltd, New Zealand). Body condition scoring (BCS) was performed according to Swedish standards, using five condition classes ranging from 1 (very lean) to 5 (very fat) (Eggertsen 2007). The target for the lambs for slaughter was BCS 3 and live weight 47–50 kg. The lambs were divided into 10 slaughter groups, with 6–8 animals in each group.

Slaughter

For practical reasons, all animals were gathered and kept indoors on the farm on the night before slaughter. All had free access to water and silage until transport to a commercial abattoir, located about 10 minutes' drive from

the farm. All animals were transported in a horse trailer driven by staff from the university (SLU) to the abattoir at approximately 08:00 h. All procedures at the abattoir, such as lairage before slaughter, were varied as little as possible. The lambs were rendered unconscious by captive bolt stunning and then exsanguinated within 6 ± 2 seconds. Carcass weight (hot carcass weight × 0.98) and carcass grade were recorded. Conformation and carcass fatness were assessed manually by a certified classifier using the EUROP-scale, which has 15 classes ranging from 1 (poor conformation/very low fat) to 15 (very excellent conformation/very high fat). Dressing percentage was calculated as carcass weight/live weight × 100. Muscle pH was recorded, in the *M. longissimus* muscle, 24 hours after slaughter of each animal (Seven2Go pro, Metler Toledo, Schwerzenbach, Switzerland). Between test occasions, the pH probe was cleaned with pepsin solution to remove residual protein and with ethanol to remove residual fat, in accordance with the manufacturer's instructions. The probe was then re-calibrated with pH 4.0 and 7.0 buffer solutions.

Meat quality analyses

All carcasses were hung by the Achilles tendon for six days at 4 °C and then the right *longissimus* muscle was collected from the first eight slaughtered animals in each group (in total 32 lambs). Immediately after sampling, these meat samples were vacuum-packed and kept frozen at –20 °C until analysis.

For each meat sample, the colour of the thawed meat, weight loss after thawing, weight loss after cooking and Warner-Bratzler shear force (WBSF) of the cooked meat were determined. The preparations required to measure all these physical variables meant that it was only possible to process four samples per day. Therefore, we chose to analyse one sample from each sample group on each day of analysis. An additional seven analysis sessions were performed for the remaining samples. All analyses were completed within two weeks. Before each analysis session, four meat samples (one each from groups 1–4) were removed from the freezer, unpacked, weighed (start weight), repacked in a new vacuum bag and thawed for 15 hours at 4 °C. The samples were then tempered in a 20 °C water bath for one hour, unpacked and reweighed to get the weight loss after thawing. After removal of the fat cover, colour measurements were performed on the surface of the longissimus muscle, at eight locations, using DigiEye (VeriVide, Enderby, UK) and mean of the eight measurements was calculated. For WBSF measurements, repacked samples were placed in a water bath pre-heated to 75 °C for one hour or until the core temperature reached 70 °C. The samples were then allowed to cool in ice water for one hour, after which the meat was unpacked, weighed to get cooking loss and then placed in bags to reach room temperature. Cylindrical samples with diameter 15 mm (7–10 replicates/meat sample) were punched out in the longitudinal direction of the fibres for measurement of WBSF using an Instron 5542 instrument (Instron Ltd., High Wycombe, UK). A total of 7–10 measurements were performed and the mean value was calculated. Each sample was placed in a wedge-shaped recess under the cutting blade. The blade was 1 mm thick and moved downwards through a rectangular hole at a speed of 50 mm min^{-1}. The maximum force measured was used as a measure of the cutting resistance of the sample.

Statistical analysis

Statistical analyses were performed using the Mixed procedure in SAS (SAS 9.4, SAS Inst. Inc., Cary, NC, USA). Two statistical models of the following forms were created, with production system (with four sub-classes) included as a fixed effect.

Live weight gain, age at slaughter and carcass characteristics were analysed using the model:

$$Y_{ij} = \mu + P_i + e_{ij}$$

Thawing and cooking loss, colour and WBSF were analysed using a model which included day of analysis as a random effect:

$$Y_{ij} = \mu + P_i + d_{ij} + e_{ijk}$$

where Y_{ij} is the dependent variable, μ is the grand mean, P_i is the fixed effect of the production system, d_{ij} is the random effect day of analysis, and e_{ij} and e_{ijk} are the residual error ($\sim N(0, \sigma^2)$). A general Satterthwaite approximation for the denominator degrees of freedom was performed, using the SATTERTH option in SAS.

Differences were considered significant at $p<0.05$ and indicative of tendencies at $0.05 \leq p < 0.10$.

Results

Feeding intensity and live weight gain

The amount of pasture available to group 4 (5.6 cm on average over the experimental period) was less than that for the other pasture groups (9.5 cm for group 2, 9.2 cm for group 3). Production system had an effect (p<0.001) on LWG, which followed the intensity of the feeding treatments, i.e. group 1 lambs had the highest LWG, followed by group 2, group 3 and group 4, respectively (Table 3). As expected, LWG affected the age at slaughter, which differed between all four groups (p<0.001). Group 4 lambs had the highest age at slaughter. Live weight at slaughter also differed between the groups (p=0.011). Group 1 had the highest live weight at slaughter, while group 2 had higher live weight at slaughter than group 3 (Table 3). There were between-group differences in LWG during the last 14 days pre-slaughter. In group 4, growth rate per day in the last 14 days prior to slaughter exceeded the overall growth rate during the experiment.

Table 3. Live weight (LW), age and live weight gain (LWG) of lambs reared using four different production systems

Parameters	Group 1	Group 2	Group 3	Group 4	SEM	Significance
n	19	19	20	20		
LW at start, kg	26.4	26.8	26.4	26.0	0.65	NS
LW at slaughter, kg	50.6^{a}	50.3^{ab}	48.3^{c}	48.9^{bc}	0.54	0.011
Days in experiment	65^{d}	82^{c}	91^{b}	109^{a}	2.7	<0.001
Age at slaughter	149^{a}	167^{b}	177^{c}	194^{d}	2.9	<0.001
LWG, g day^{-1}	377^{a}	287^{b}	244^{c}	211^{d}	7.9	<0.001
LWG 14d^{1}, g day^{-1}	322^{a}	287^{ab}	244^{b}	319^{a}	0.0	0.043

Group 1 is reared by indoor feeding with silage and concentrate; group 2 on cultivated pasture with 0.3 kg concentrate per lamb daily; group 3 on cultivated pasture and group 4 on semi-natural pasture. SEM = standard error of the mean; NS = non-significant (p>0.05); $^{a-d}$ = Mean values within rows with different superscripts differ significantly (p<0.05); ^{1} = Average LWG during the last 14 days pre-slaughter.

Carcass quality

Production system had a significant effect on carcass weight (p<0.001), with group 4 having lower carcass weight than the other three groups. Group 4 had lower conformation (p=0.011), fat score (p=0.039) and dressing percentage (p<0.001) than all other groups (Table 4).

Table 4. Carcass quality and meat quality of lambs reared using four different production systems

Parameter	Group 1	Group 2	Group 3	Group 4	SEM	Significance
Carcass quality						
n	19	19	20	20		
Carcass weight	21.6^{a}	21.3^{a}	20.9^{a}	18.8^{b}	0.47	0.003
Conformation^{1}	9.2^{a}	8.7^{a}	8.7^{a}	7.9^{b}	0.24	0.002
Fatness^{2}	7.4^{a}	7.7^{a}	7.4^{a}	6.5^{b}	0.17	< 0.001
Dressing, %	42^{a}	42^{a}	41^{a}	37^{b}	0.4	< 0.001
pH_{24}	5.83	5.66	5.77	5.59	0.098	NS
Meat quality						
n	8	8	8	8		
Age at slaughter, d	146^{a}	163^{b}	172^{c}	193^{d}	3.5	< 0.001
pH_{6d}	5.45	5.40	5.45	5.41	0.032	NS
Thawing loss, %	4.3	4.8	4.1	5.0	0.40	NS
Cooking loss, %	24.3	24.1	22.8	24.2	0.87	NS
Colour L*	37.1	37.2	36.5	35.7	0.69	NS
Colour a*	16.9	16.5	16.6	16.4	0.28	NS
Colour b*	7.5	7.0	6.8	7.1	0.31	NS
WBSF, N(cm^{2})$^{-1}$	33.8	45.9	31.9	34.9	4.88	NS

Group 1 is reared by indoor feeding with silage and concentrate; group 2 on cultivated pasture with 0.3 kg concentrate per lamb daily; group 3 on cultivated pasture and group 4 on semi-natural pasture. SEM = standard error of the mean; $^{a-b}$ = Mean values within rows with different superscripts differ significantly (p<0.05).^{2} = According the EUROP system, where 1=P-, 2=P, 3=P+, 4=O-, 5=O, 6=O+; 7=R-, 8=R, 9=R+, 10=U-, 11=U, 12=U+, 13=E-, 14=E and 15=E+; ^{3} = According the EUROP system, where 1=1-, 2=1, 3=1+, 4=2-, 5=2, 6=2+; 7=3-, 8=3, 9=3+, 10=4-, 11=4, 12=4+, 13=5-, 14=5 and 15=5+; NS = non-significant (p>0.05); WBSF = Warner-Bratzler shear force

Meat quality indicators and technological meat quality attributes

There were no differences between groups with respect to the technological meat quality attributes, i.e. pH after 24 hours, pH_{6d} (six days after slaughter), thawing loss, cooking loss, colour (L*, a* and b*) and WBSF (Table 4).

Discussion

In this study, higher feeding intensity resulted in higher LWG, with group 1 lambs having the highest LWG, but there was no effect on meat quality attributes. Live weight gain and/or live weight at slaughter differed from those in comparable studies (e.g. Young et al. 1994, Pethick et al. 2005, Campbell et al. 2012).

The LWG values were mostly higher than in previous studies, where some of the treatments described even gave negative LWG (Young et al. 1994, Pethick et al. 2005, Campbell et al. 2012). However, the nutrient content of the feeds used in those studies is only briefly described, so it is difficult to compare the results with those in the present study. The energy content of the cultivated pasture for group 3 was lower than that for group 2, even though both groups grazed the same field (albeit different parts). This difference in energy content was most likely due to differences in the establishment and relative abundance of various plant species, i.e. the sward composition in the pasture, influencing the nutritional content in different areas of the pasture. Although the nutrient content in the semi-natural pasture was good (Table 2), it should be noted that the amount of pasture available for group 4, based on sward height, was less than that for the other pasture groups (groups 2 and 3). This observation is important for understanding the comparatively low growth rate of group 4, which was most likely adversely affected by pasture availability, as the nutritional content of the pasture was high and should have resulted in higher growth. However, the LWG in group 4 lambs (211 g day^{-1}) was similar to that in a study by Lind et al. (2009), who observed LWG of 230 g day^{-1} on semi-natural pastureland in northern Norway. Thus, it can be concluded that rearing lambs on semi-natural pasture in the temperate climate zone is feasible. The LWG_{14d} for group 4 was higher than the average LWG for the whole rearing period. The reason for this remains unclear. To increase the weight at slaughter for animals reared on semi-natural pastures (group 4), the lambs could: i) be given supplementary feed or ii) lambing time could be brought forward in the spring, in order to enable earlier release to pasture and ensure that the lambs have time to grow and reach slaughter maturity before the nutritional quality and availability of semi-natural pastures decreases in the autumn.

As expected, the high-intensity feeding systems (groups 1 and 2) and the intermediate production system (group 3) all yielded carcasses with higher conformation and fatness scores than the extensive feeding system (group 4). However, although group 4 lambs had the lowest scores for both conformation and fatness, the carcasses would still qualify for the highest payment per kg carcass weight according to the current price list at two Swedish abattoirs (HKScan 2020, KLS 2020). The dressing percentage was lower for group 4 (37%) compared with the other groups (41–42%). This could be explained by the higher age at slaughter for group 4 lambs, which is associated with greater loss of weight in terms of head, bones and intestines (Muir et al. 2008). This loss of weight probably did not derive from greater rumen fill at slaughter, since the amount of pasture available to lambs reared under semi-natural conditions was limited. As expected, feeding intensity affected both growth per day and number of days to slaughter. Although group 3 lambs were reared at a lower intensity than those in groups 1 and 2, they were ready for slaughter at around the same time as group 1 and 2 lambs, despite being raised solely on cultivated pasture. Overall, there were no appreciable differences between groups 1, 2 and 3 with respect to carcass conformation and fatness scores.

In meat from groups 1 and 3, the pH_{24} value exceeded 5.7 which is considered as the upper limit for a good eating quality (MSA 2015b). This outcome was unexpected, since high growth rates are associated with high glycogen storage in muscles, which normally causes the expected post-slaughter pH decline. However, our results are consistent with those of Pethick et al. (2005), who found that meat from animals fed a high-energy diet had higher pH_{24} than meat from animals fed low-energy diets. That study also found that the high-energy group unexpectedly lost a greater proportion of glycogen between farm and slaughter than lambs raised on pasture. Pethick et al. (2005) suggested that these animals may have been more predisposed to lose glycogen in response to stress, a trait which could be of metabolic or behavioural origin. In the present study, even though groups 1 and 3 meat had high pH_{24} values, there was no expected effect of high pH on the tenderness of the meat (WBSF values). Other studies have found that lower pH_{24} values are associated with more tender meat, e.g. Devine et al. (1993) found that meat is most tender when its final pH is 5.5–5.7. Moreover, two of the pH_{24} values observed in the present study were above the upper limit of 5.7 recommended by Meat Standards Australia (MSA 2015b). Based on this standard,

there was a risk of meat from groups 1 and 3 having lower eating quality than that from the other groups. Commercial research in New Zealand has established that the desirable pH_{24} range for lamb is 5.4–5.8, with values of 5.8–6.0 being associated with intermediate quality (Alliance Group Ltd 2010). Based on these criteria, groups 2, 3 and 4 had more numerically preferable pH_{24} values than group 1.

In the literature, the pH at 24 hours post mortem is often defined as ”final” pH (e.g. Koohmaraie et al. 1991, Koohmaraie et al. 1995, Watanabe et al. 1996, McGeehin et al. 2001, Díaz et al. 2002, Priolo et al. 2002, Sañudo et al. 2003, Velasco et al. 2004, Pethick et al. 2005, Teixeira et al. 2005, Majdoub-Mathlouthi et al. 2013, Majdoub-Mathlouthi et al. 2015). However, in the present study a continuous decline in pH was seen between pH_{24} and pH_{6d} (Table 4). In contrast, Koohmaraie et al. (1995) found that the pH of lamb carcasses 24 hours post mortem (pH_{24} = 5.6) was identical to that six days later, and that carcass pH values then rose slightly (to 5.7) between seven and 21 days post mortem. It is thus not clear whether pH generally drops significantly after 24 hours or not. However, the results presented both in this study and in Koohmaraie et al. (1995), suggest that describing the pH at 24 hours post mortem as “final” or “ultimate” may be inaccurate.

Technological meat quality parameters determined in this study can only be compared with those in previous work to a limited extent. Comparisons between studies are hampered by differences in e.g. feeding intensity, LWG, breed, sex, intact or castrated ram lambs, carcass weight and slaughter method, or the fact that some of these factors are barely described. These differences derive from the many different production systems used for lamb, which have different prerequisites such as climate and tradition. Thus, previous studies have found no differences in WBSF (Berge et al. 2003, Rodrígues et al. 2008, Karaca et al. 2016), significant differences in WBSF (Sañudo et al. 2003), no differences in meat colour (Díaz et al. 2002, Pethick et al. 2005), significant differences in meat colour (Priolo et al. 2002) and no differences in water loss (Díaz et al. 2002, Rodrígues et al. 2008, Karaca et al. 2016) when comparing different lamb types and production systems.

Berge et al. (2003) did not find any differences in WBSF (2.17–3.69 kg, equal to 21.3–36.2 N) between indoor-reared entire male lambs fed concentrate compared with entire male lambs reared on different types of pasture, with or without concentrate. The results in Berge et al. (2003) are not fully comparable with ours, due to differences in animal age at slaughter (3.5–7 months), carcass weight (10.4–19.7 kg) and feeding systems, but were similar to those in our study. Karaca et al. (2016) studied the effect of two feeding systems where lambs were fed a finishing diet of either alfalfa hay (1750 g day^{-1}) or alfalfa hay (1250 g day^{-1}) + 500 g of barley per lamb and day, with both diets balanced to give equal energy intake in the two groups. Live weight gain for both groups (90 and 32 g day^{-1}) was low compared with that in the present study, but these values were significantly different. The WBSF values did not differ between groups in that study (45.7 and 43.9 N), as also found in the present study. However, carcass weights reported by Karaca et al. (2016) (17.5 and 15.7 kg) were lower than those in the present study. Significant differences in average daily gain between groups were observed in both that and the present study, indicating that differences in growth rate do not automatically result in differences in WBSF. Further, Karaca et al. (2016) found no differences in either meat colour (L*, a* or b*) or cooking loss, corresponding to the results of the present study. A study by Rodrígues et al. (2008) found differences in LWG when comparing different feeding intensities (straw + pelleted commercial concentrate compared with whole barley grain + protein supplement), with a LWG per day of 272 g for the straw group and 371 g for the barley group. On the other hand, Rodrígues et al. (2008) did not find any differences in meat colour, WBSF or water-holding capacity. However, carcass weight was lower (by 12 kg) than in the present study, hampering direct comparison of the results.

Sañudo et al. (2003) compared different production systems from six European countries, based on either grass or concentrate or a combination of both. The WBSF results in that study revealed significant differences between systems related to different feeding strategies, age at slaughter, carcass weight and breeds. However, the results are not unequivocal, indicating a combined effect of the factors mentioned above on WBSF. Nevertheless, the results indicate that WBSF values are affected by age at slaughter when comparing entire male lambs with similar carcass weights. The youngest animals had the lowest WBSF values in the study by Sañudo et al. (2003), but no such effect was seen in the present study. The lack of differences in technological meat quality attributes in the present study is a positive finding, since it indicates that all four production systems compared can be used in practice without altering the technological properties of the lamb meat. Pethick et al. (2005) found significant differences in LWG when comparing different feeding intensities (pasture, moderate-energy pellet, high-energy pellet, straw). They found no differences in L* and b*, as in the present study, but observed differences in a*, with meat from lambs in the pasture and moderate-energy treatment having a darker red colour than the high-energy pellet group, in contrast to the present study. Pethick et al. (2005) attributed the differences in a* to elevated ultimate pH levels in the high-energy pellet (pH 5.66) and straw (pH 5.67) feeding treatments compared with pasture

(pH 5.57) and moderate-energy pellet (pH 5.59). This could also explain the lack of differences in colour in the present study, since pH_{24} did not differ significantly between groups and may therefore not have influenced any of the colours (L*, a* or b*). As in the present study, Díaz et al. (2002) did not observe any differences in colour (L*, a* or b*) or water-holding capacity when comparing concentrate and pasture for fattening lambs. However, Priolo et al. (2002) recorded differences in L*, with a grass-fed group producing darker meat than the stall-fed group, and saw a tendency for differences in b*, with the grass-fed group having a lower yellowness index than a stall-fed group. Priolo et al. (2002) attributed the difference in L* to numerically higher ultimate pH for the grass-fed group (pH 5.62) compared with the stall-fed group (pH 5.57). However, this difference was non-significant, and it is thus questionable whether it can explain the difference in L* between grass-fed and stall-fed lambs.

Conclusions

There were differences in LWG between the four production systems studied here, but the results indicate that intact ram lambs can be reared under intensive or extensive conditions without any differences in meat quality attributes. Parameters affected by the production system included age at slaughter, live weight gain, carcass weight, carcass conformation and fatness scores. Lambs reared on cultivated pasture had better carcass classification (conformation and fatness), as well as carcass weights than those reared on semi-natural pasture. Further studies are needed to evaluate the existing recommendations of pH_{24} to be 5.7 for meat to identify if this recommendation applies for animal material and production systems used in this study. Studies are also needed on whether pH at 24 hours post mortem should be described as “final” or “ultimate” pH, since further pH decline in muscle beyond 24 hours has been observed. If pH measurements are to be carried out later than 24 hours after slaughter, possible correlations between pH and meat quality attributes, such as tenderness, should be examined.

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III

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Article

A Comparison of Two Different Slaughter Systems for Lambs. Effects on Carcass Characteristics, Technological Meat Quality and Sensory Attributes

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Simple Summary: Slaughter systems for lambs can differ in many ways, as they are optimised to meet site-specific conditions, and may affect carcasses differently. It is therefore important to assess whether different slaughter systems affect meat quality parameters such as colour and tenderness, which are important meat quality factors from a consumer perspective when buying a meat product. This study investigated whether two slaughter systems differing in stunning method, electrical stimulation of carcasses and chilling regime resulted in differences in quality attributes in meat from intact male lambs. It also examined whether breeding for carcasses with a higher incidence of lean muscle and less fat content has affected tenderness scores for Icelandic lambs over time. The results showed that the two slaughter systems tested did not affect meat quality parameters to any large extent. Further analysis showed an increase in mechanical tenderness values in meat samples from the Icelandic lamb population over time, which could be due to selective breeding for carcasses with higher muscle and less fat content.

Abstract: Two slaughter systems for lambs and their effects on meat quality in terms of texture, colour and sensory attributes were compared. The slaughter systems differed in methods for controlling *rigor mortis* and carcass chilling. One slaughter system (large-scale) used electrical stimulation and fast chilling of carcasses, while the other system (small-scale) did not use electrical stimulation and applied slower chilling, with carcass temperature decreasing over a longer period after slaughter. Ten pairs of ram lamb twins were selected, and one of each pair was slaughtered at the large-scale abattoir and the other at the small-scale abattoir. Carcass weight, conformation, fatness, pH and temperature were recorded. *Musculus longissimus thoracis et lumborum* was analysed for colour, cooking loss, Warner–Bratzler shear force and sensory attributes. For meat quality attributes, the only differences were found in meat colour L* (lightness; $p = 0.0073$), sensory attribute "appearance colour" ($p = 0.0089$) and "fatty flavour" ($p = 0.0554$). Meat from the small-scale abattoir was darker in colour and had a more fatty flavour than the meat from the large-scale abattoir. For sensory attributes (apart from colour), no significant differences were found between the two abattoir systems.

Keywords: lamb; pH; meat colour; cooking loss; Warner–Bratzler shear force; selected breeding; slaughter system

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1. Introduction

Lamb production is a traditional and economically important part of Icelandic agriculture. The production system for lambs in Iceland generally involves lambs being born in May and reared on summer pasture until gathering in August, when lamb slaughter starts.

The slaughter season usually ends in November. Lamb has high domestic food value and it is of critical importance to produce high-quality meat that satisfies consumer demands. An investigation in 2003 on the tenderness of Icelandic lamb found low shear force values (mean 1.8 kg/cm^2), indicating tender meat [1]. However, a more recent study indicated a decrease in tenderness of *Musculus longissimus dorsi* meat from Icelandic lamb during the past two decades [2]. Probable reasons for this trend are (i) ongoing breeding for increased muscle and decreased fat using progeny testing, based on ultrasound measurements of live animals for animal selection [3], and (ii) changes in slaughter practices, with faster chilling of lamb carcasses and low-voltage electrical stimulation. It is known that rapid chilling of carcasses can affect meat tenderness negatively, by increasing the risk of cold shortening [4]. Electrical stimulation of carcasses can prevent cold shortening when applied in combination with fast chilling of the carcass after slaughter [5]. To prevent cold shortening through accelerating pH decline, electrical stimulation within 30 min post-mortem can be applied [6]. Electrical stimulation can therefore be used to improve the eating quality of meat by controlling the pH decline in relation to muscle temperature [7]. However, growing interest in local produce and in procuring meat directly from farms is resulting in small local abattoirs using traditional slaughter methods being set up.

It is important to understand how the slaughter system can affect meat quality parameters, in order to optimise it and avoid negative effects on quality. It is also important to determine how breeding has affected meat quality attributes such as tenderness, in order to adapt or make changes in the breeding goals if necessary. Hence selection for lean meat yield could negatively affect meat quality as perceived by consumers [8]. The aims of this study were therefore to investigate whether small-scale slaughter systems affect quality attributes in lamb meat compared with large-scale slaughter systems and to assess whether breeding for carcasses with a higher incidence of muscle and less fat has affected lamb tenderness scores over time.

2. Materials and Methods

Ten pairs of intact ram lamb twins of the breed Icelandic sheep were included in the study. One of each pair was slaughtered in a large-scale abattoir slaughtering 2500 lambs/day and the other in a small-scale abattoir slaughtering 75 lambs/day. All lambs were raised on the same farm in south-east Iceland. The lambs were born in May and grazed with their mothers for the first weeks on cultivated pasture and then on natural pasture (aftergrass) until mid-September. Before slaughter, all lambs were kept in lairage for 10–12 h overnight at the abattoir without feed, but with free access to water. Lambs were on average 160 days at slaughter. All carcasses were visually assessed by trained personnel according to the EUROP scale for conformation (classes 1–5, with class 5 being the most swelling) and fatness (classes 1–6, with class 6 being the fattiest). All carcasses were hung by the Achilles tendon after dressing (removal of skin, legs, head and visceral organs).

2.1. Large-Scale Abattoir

Transport from the farm to the large-scale abattoir involved three hours in a ventilated lorry. The lambs were stunned electrically (head to back) with 110 V, 50 Hz for 5 s and slaughtered by exsanguination. After dressing and evisceration, the carcasses were electrically stimulated (10 A, 80 V for 60 s) before entering the chiller. The carcasses were then chilled at 2–4 °C for 30 h before sampling of *M. longissimus thoracis et lumborum* (LTL).

2.2. Small-Scale Abattoir

Transport time from the farm to the small-scale abattoir was 30 min. All lambs in that system were stunned with a captive bolt pistol and slaughtered by exsanguination. The carcasses were kept at 10–15 °C for the first six hours after slaughter and then chilled at 3–4 °C for 30 h until sampling of the LTL muscle.

2.3. Carcass Weight Loss during Chilling

The carcasses were weighed upon arrival at the chiller (hot carcass weight) and after chilling (cold carcass weight). The difference in weight was used for calculating carcass weight loss (%) as:

% Carcass weight loss = (Hot carcass weight (g) – Cold carcass weight (g))/Hot carcass weight × 100.

2.4. Sampling, Packaging and Handling of the LTL Muscle

The LTL muscle with subcutaneous fat was cut from the left side of all carcasses, between the last lumbar vertebrae and the seventh thoracic vertebrae, 24–30 h after slaughter. The whole muscle was labelled, weighed and vacuum-packed in 25 cm × 35 cm bags, and then aged for 6–7 days at 2–4 °C. All LTL samples were analysed fresh for both technological and sensory quality attributes. Unfortunately, two samples from the large-scale abattoir were frozen prior to analysis, and are thus not included in the results for colour, cooking loss, Warner–Bratzler shear force (WBSF) and sensory attributes.

2.5. pH Measurements

The pH and temperature value in all carcasses were measured 24 h after slaughter, by inserting a probe (Seven2Go pro, Metler Toledo, Schwerzenbach, Switzerland) into the LTL muscle between the 12th and 13th ribs.

2.6. Colour

Colour was measured before vacuum packaging on fresh, uncooked LTL one day after slaughter, using a Minolta CR-300 colorimeter (Konika Minolta, Tokyo, Japan) with a D65 light source. The instrument provided information about the lightness (L* value), redness (a* value) and yellowness (b* value) of the muscle samples. The LTL muscle was allowed to bloom for one hour before measuring. Each attribute was measured in triplicate for each muscle sample.

2.7. Warner–Bratzler Shear Force (WBSF)

Instrumental LTL tenderness, expressed as shear force, was analysed on cooked LTL, using a Warner–Bratzler knife (TA-7), with a guillotine block, connected to a texture analyser (TA.HD Plus Connect, Godalming, Surrey, UK), and a head speed of 2.5 mm/s. Samples with dimensions 1.0 cm × 1.0 cm × 3.0 cm (width × height × length), cut orthogonal to the fibre direction, were used for these measurements.

2.8. Sensory Evaluation

The sensory evaluation was carried out using a trained panel (Generic descriptive analysis) of a total of 11 panellists, with 6–10 panellists at each of five occasions depending on availability [9]. The software used for checking panel performance was PanelCheck V1.4.0 (Nofima, Tromsö, Norway) and that used for data collection was FIZZ (Version 2.50B, Biosystémes, Couternon, France). PanelCheck is a tool that uses several plots to evaluate results from descriptive analyses, which helps the user to identify the performance of individual assessors. The meat was evaluated in five sensory sessions, with four different samples per panellist tested in each. Each panellist tested one sample per LTL from the same location within the muscle. Hence, all LTL was divided into ten samples to provide each panellist with a sample of LTL from the same muscle location in each session. All samples were coded with random three digit numbers and presented to the panellists in a random order. Latin square order was used to obtain randomized order of samples. The meat was cooked sous vide (Anova Precision Cooker, Anova Culinary Inc., San Francisco, CA, USA) at 68 °C for one hour and then flash-fried in a dry pan at a high temperature for 30 s on each side. The cooked meat was cut into 2 cm thick slices, placed in aluminium containers and served to the panel while still warm. The panellists were placed in separate cubicles with standardised light. They were offered crackers and water to avoid residual flavours

between samples. Panellists evaluated each sample on a scale from 1 to 100 for odour (frying, sour, fatty and liver), appearance (colour), flavour (frying, sour, fatty, sweet and liver) and texture (softness, tenderness, juiciness and mushiness).

2.9. Cooking Loss

All LTL samples were weighed before and after cooking, to calculate cooking loss (%).

2.10. Statistics

Statistical analysis was performed using Proc Mixed of the Statistical Analysis Software (SAS) [10], with slaughter system as a fixed effect and twin pair of lambs as random effect, using model 1 below. A general Satterthwaite approximation for the denominator degrees of freedom was performed, using SATTERTH option in SAS. Differences were considered significant at $p \leq 0.05$ and a tendency for significance was assumed at $0.05 < p \leq 0.10$. Carcass characteristics and technological meat quality attributes are presented as mean and standard deviation, respectively.

$$\text{Model 1: } Y_{ij} = \mu + S_i + P_j + e_{ijk} \quad (1)$$

where Y_{ij} is the dependent variable, μ is the grand mean, S_i is the fixed effect of slaughter system, P_j is the random effect of twin pair, and e_{ijk} is the residual error.

3. Results

3.1. Carcass Characteristics and Technological Quality Attributes

There was a difference in fatness scoring ($p = 0.0002$) between the abattoirs, with the small-scale abattoir obtaining a higher score (3.5) than the large-scale abattoir (2.3) (Table 1). There was also a difference ($p = 0.0073$) in colour (L*) between the abattoirs, with the large-scale abattoir having a higher L* value than the small-scale abattoir (Table 1). A tendency for significance was found in temperature$_{24}$ where the small-scale abattoir had a bit lower temperature (4.3 °C) compared to the large-scale abattoir (5.0 °C). There were no significant differences between the slaughter systems for carcass conformation, hot carcass weight, cold carcass weight, carcass weight loss, pH$_{24h}$, cooking loss or WBSF (Table 1). Individual data for each animal regarding carcass characteristics and technological quality attributes can be found in Tables S1 and S2.

Table 1. Carcass characteristics, pH$_{24h}$, temperature$_{24}$, colour measurements and cooking loss of lamb slaughtered in the small-scale system ($n = 10$) and large-scale system ($n = 10/n = 8$ [1]; means ± stdev).

Parameters	Small-Scale	Large-Scale	SEM [2]	*p*-Value [3]
Carcass characteristics				
Hot carcass weight (kg)	19.8 ± 1.2	19.8 ± 1.7	0.46	0.9580
Cold carcass weight (kg)	19.4 ± 1.1	19.3 ± 1.7	0.46	0.9039
Carcass weight loss (%) [4]	2.44 ± 0.6	2.74 ± 0.2	0.13	0.1476
Conformation score [5]	3.9 ± 0.3	4.0 ± 0.5	0.13	0.5911
Fatness score [6]	3.5 [a] ± 0.5	2.3 [b] ± 0.5	0.16	0.0002
pH$_{24}$ [7]	5.65 ± 0.1	5.63 ± 0.1	0.04	0.7118
Temperature$_{24}$ (°C) [8]	4.3 ± 0.6	5.0 ± 1.0	0.25	0.0986
Meat quality attributes				
L*	36.5 [a] ± 1.5	38.3 [b] ± 1.4	0.47	0.0073
a*	18.8 ± 2.2	20.2 ± 1.0	0.51	0.4928
b*	4.2 ± 1.4	4.9 ± 0.7	0.44	0.5500
Cooking loss (%)	14 ± 3.4	16 ± 2.4	1.01	0.1231
WBSF (N) [9]	49 ± 15.3	43 ± 9.4	4.22	0.9651

[1] Regarding large-scale abattoir $n = 10$ for carcass characteristics; $n = 8$ for meat quality attributes. [2] Standard error of the mean. [3] Differences considered significant at $p < 0.05$ and tending towards significance at $0.05 < p \leq 0.10$. [a,b] = Mean values within rows with different superscripts differ significantly. [4] Difference between hot and cold carcass weight (%). [5] Scoring into five classes, with five being the highest (very good conformation) and one being the lowest (very poor conformation). [6] Scoring into six classes, with six being the highest (very high fat) and one being the lowest (very low fat). [7] pH at 24 h after slaughter. [8] Temperature at 24 h after slaughter. [9] Warner–Bratzler shear force measured in Newton.

3.2. Sensory Attributes

In line with the technological colour measurements, a difference (p = 0.0089) was found for the sensory parameter "colour appearance", with lamb from the small-scale abattoir obtaining a higher score (33, i.e., darker) than lamb from the large-scale abattoir (30) (Table 2). A difference (p = 0.0370) was also found for the parameter "fatty flavour", for which lamb from the small-scale abattoir had a higher score (18) than lamb from the large-scale abattoir (15) (Table 2). No differences were found for the different odour and texture attributes, or for frying, sour, sweet and livery flavour (Table 2). Individual data for each animal regarding sensory attributes can be found in Table S2.

Table 2. Warner–Bratzler shear force values and sensory analyses comparing lamb slaughtered in the small-scale system (n = 10) and large-scale system (n = 8).

Parameters		Small-Scale	Large-Scale	SEM [1]	p-Value [2]
Odour attributes [3]	Frying	33	33	2.58	0.5446
	Sour	13	12	1.08	0.1398
	Fatty	28	27	1.90	0.9660
	Liver	29	30	1.53	0.5053
Appearance attribute [3]	Colour	33 [a]	30 [b]	1.85	0.0089
Flavour attributes [3]	Frying	26	23	1.93	0.8533
	Sour	24	26	2.23	0.1054
	Fatty	18	15	1.36	0.0370
	Sweet	9	9	0.57	0.5045
	Liver	41	41	1.80	0.7850
Texture attributes [3]	Soft	47	53	4.67	0.8389
	Tender	46	50	5.24	0.8131
	Juicy	49	50	3.57	0.7513
	Mushy	14	17	1.39	0.3099

[1] Standard error of the mean. [2] Differences considered significant at $p \leq 0.05$. [a,b] = Mean values within rows with different superscripts differ significantly. [3] Sensory attributes were scored on a scale from 0–100, with 100 being the highest score.

4. Discussion

The two slaughter systems compared in this study differed in multiple ways, such as type of stunning, use of electrical carcass stimulation and chilling regime. Despite these major slaughter and carcass handling differences, only a few differences were found in technological and sensory meat quality attributes. This indicates that both systems investigated can be used in practice without compromising these meat quality attributes under the circumstances stated. This was an unexpected finding, as previous studies have shown that numerous factors during pre-slaughter, slaughter and post-slaughter can all contribute to variation in meat quality attributes, as reviewed by Sañudo et al. [11]. The results from the present study indicate good robustness for both slaughter systems tested and the different treatments within each system. A limitation of this study could thus be that all animals came from the same farm and got slaughtered on the same day. However, in this way, we could not only limit the genetic effects by using twin lambs, and we also reduced the effect of variation between farms as well as day of slaughter which let us focus on the effect of slaughter system.

The stunning method used in the abattoirs was either captive bolt or electrical stunning. Previous studies have found that the stunning method does not affect meat quality attributes [12]. It has also been shown that the use of electrical stunning head-to-leg, combined with low voltage electrical stimulation of the carcass, can increase the rate of pH decline post-slaughter, compared with electrical head-only and captive bolt stunning with low-voltage electrical stimulation. While not measured in this experiment, electrical stunning increasing pH decline could be interpreted as a positive effect prior to fast chill-

ing of carcasses shortly after stunning, to avoid cold shortening [13]. Therefore it can be concluded that the electrical stunning combined with the fast chilling approach used in the large-scale abattoir may be beneficial to avoid cold shortening.

The pairs of twin lambs included in this study were almost identical in carcass weight and conformation score, but there was a significant difference in fat score between the abattoirs. This difference probably arose because the classification was performed by different individuals in the two abattoirs [14], since a review by Craigie et al. [15] have shown that carcass scoring is not consistent between individual classifiers.

Both pH_{24} and cooking loss were unaffected by slaughter system. Both slaughter systems resulted in a pH_{24} value that met recommendations by Meat Standards Australia (MSA), with an upper limit of 5.7 [16]. Hence, based on recommendations from the MSA the pH_{24} values recorded in the present study would have positive effects on eating quality attributes such as tenderness. Cooking loss was unaffected by electrical stimulation (large-scale abattoir only), supporting previous findings [5,6,17]. It can therefore be concluded that the slaughter systems had similar effects on pH_{24} and cooking loss in the lamb samples.

Meat colour is a complex meat quality factor that can be affected by various factors linked to the production system, the slaughter system and the animal material [18]. The results obtained here for a* and b* were similar for both slaughter systems, supporting previous findings [6,14,19]. Hopkins and Ferrier [6] showed that, despite a more rapid pH decline in stimulated carcasses, there was no effect on meat colour. Even with similar pH_{24} values in lamb samples, a difference in L* was found between the slaughter systems in the present study, which is in line with findings by Pouliot et al. [5]. A darker colour was observed for meat samples from the small-scale abattoir compared with the large-scale abattoir. Differences in lightness have previously been explained by different myoglobin concentrations in muscle due to differences in animal age [20], or to changes during the first hour of meat blooming due to different temperatures in muscle at rigor development [21]. The reason for the difference in lightness in the present study is unclear since there was no difference in animal age. However, this difference was also detected on cooked meat by the sensory panel, which recorded a difference in the attribute "colour appearance" between the two abattoirs, with meat from the small-scale treatment being rated as darker than meat from the large-scale abattoir. At the point of purchase, based on previous findings, consumers would consider meat from both slaughter systems studied here to be of acceptable lightness [22]. Therefore it can be assumed that, while the difference in lightness might be visible to the consumer, it would probably not influence the acceptability of lamb from either of the slaughter systems compared here.

Another difference between the abattoirs was the use of electrical carcass stimulation, as low-voltage electrical stimulation of the carcasses was used only at the large-scale abattoir. Previous studies have compared different electrical settings (V, Hz, A) and different durations of electrical stimulation (s), with non-consistent results. Some studies have found differences in WBSF when using low-voltage electrical carcass stimulation compared with a control group [5,17,23–27], while other studies have found no differences [6,19,28]. A study by Polidori et al. [26] concluded that low-voltage electrical stimulation of lamb carcasses may reduce potentially negative risks to meat quality associated with fast chilling of carcasses immediately after dressing. This may be because low-voltage electrical stimulation induces glycolysis [29]. However, Pommier et al. [28] used low-voltage stimulation in their study and, based on their results, they suggest that high-voltage electrical stimulation is required to get a rapid pH decline that allows early freezing of meat without negative consequences for meat quality attributes. It is debatable whether high-voltage electrical stimulation is required to allow fast chilling in order to avoid cold shortening. The results from the present study suggest that low-voltage stimulation is sufficient to allow fast chilling of carcasses without affecting WBSF negatively, at least compared with the small-scale slaughter system without electrical stimulation and a slower chilling process.

When considering WBSF from the perspective of the values themselves, there were no significant differences between the slaughter systems (Table 1). The breeding goals

for Icelandic sheep (the only bread present in Iceland) in recent decades have focused on carcass composition traits, such as improved slaughter weight, more muscle and less fat [3]. This selection process has resulted in a change in carcass composition in Icelandic lambs, according to Eiríksson and Sigurdsson [3]. Based on the WBSF threshold of ≤5 kg (~49 N) set by Shorthose et al. [30], the meat from slaughter systems can be interpreted as acceptably tender. However, meat samples from both the large-scale and small-scale systems (WBSF = 49 N and 43 N, respectively), were near or at the threshold. In studies in 2003 of meat from male Icelandic lambs [1,31], lambs just over 4 months of age had WBSF values of 17.2 N (1.75 kg) [1] and 18.3 N (1.87 kg) [31] and 7-month-old lambs had a value of 27.6 N (2.81 kg) [1]. A study in 2018 reported WBSF values for Icelandic lamb of on average 48.2N (4.92 kg) [2], indicating an increase in WBSF due to selective breeding over recent decades. This statement is supported by previous findings that selection for lean meat yield can reduce the consumer eating quality of lamb [8]. In addition, others have found decreasing sensory scores for the traits tenderness, juiciness, flavour and overall liking as an effect of breeding [14].

Except for "colour appearance" and "fatty flavour", the attributes tested in the present study did not differ between the two slaughter systems. Previous studies have found differences in the sensory attributes firmness (less firm with electrical stimulation) [5], flavour (ovine flavour less intense with electrical stimulation) [5], palatability (more palatable with electrical stimulation) [25] and tenderness (more tender with electrical stimulation) [23–25] when comparing electrical stimulation to untreated carcasses. However, other studies have found no differences in tenderness [28], flavour [23,25,28] or juiciness [5,23,25,28]. Spanier et al. [32] detected a difference in flavour development in beef during post-mortem ageing, which could explain the tendency for the small-scale abattoir in the present study to have a higher score for fatty flavour. Another explanation for differences in flavour between electrical stimulation and control groups is suggested to be connected to decreased firmness of the meat following electrical stimulation [5]. The meat then requires less chewing and flavour compounds are less likely to be liberated, so the flavour perception is lower [5]. However, there was no significant difference in WBSF or perceived tenderness scores between the system with and without electrical stimulation in the present study, contradicting this suggestion.

5. Conclusions

The most important finding from this study was that the two slaughter systems compared, one with electrical stimulation and fast chilling of carcasses and one with electrical stimulation and slower chilling of carcasses, did not result in any differences in meat quality parameters. However, more research is needed to see if the results of this study persist with different animal material and production systems.

Supplementary Materials: The following are available online at https://www.mdpi.com/article/10.3390/ani11102935/s1, Table S1: Individual data on pH and carcass characteristics (Small-scale = Small-scale slaughter system, Large-scale = Large-scale slaughter system). Table S2: Individual data on technological meat quality attributes (colour, cooking loss and WBSF) and sensory attributes (Small = Small-scale slaughter system, Large = Large-scale slaughter system).

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treatment of the lambs was the same as of other lambs at the farm and in the abattoirs. They were from a normal production system and nothing was altered. We only sampled meat from the carcasses of the lambs the day after slaughter.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

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IV

animals

MDPI

Article

A Comparison of Fresh and Frozen Lamb Meat—Differences in Technological Meat Quality and Sensory Attributes

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Simple Summary: Freezing is used to extend the storage time of meat and is common practice in lamb meat production, since it maintains a steady supply of seasonal meat throughout the year and allows shipping over long distances. Fresh meat may also be purchased and frozen at home, to enable longer storage of the product before consumption. Freezing is the best preservation method, apart from chilling of fresh meat. However, differences in quality parameters between fresh and frozen meat may influence consumer choice and preferences. It is thus important to evaluate these differences, and how they are affected by conditions and animal handling during primary production, slaughter method and storage conditions before retail sale. This study examined the effect of freezing on technological meat quality and sensory attributes in lamb meat samples collected at two different slaughterhouses using different slaughter methods. Several differences between fresh and frozen-thawed meat were detected in terms of technological meat quality and sensory attributes, including colour, Warner-Bratzler shear force, cooking loss, flavour attributes and juicy texture.

Abstract: Technological meat quality and sensory attributes of fresh and frozen lamb meat were compared. Samples were collected from two abattoirs (one small-scale, one large-scale) that use different slaughter methods in terms of chilling regime and electrical stimulation. The fresh and frozen meat samples included products from both slaughter systems. Ten twin pairs of ram lambs were used in the study, with one of each twin slaughtered at each abattoir. Fresh meat was analysed after chilling and frozen meat was stored frozen for three months and analysed after thawing. The *Musculus longissimus thoracis et lumborum* was analysed for colour, cooking loss, sensory attributes, Warner-Bratzler shear force (WBSF) and distribution of water and lipid within each meat sample. Meat samples analysed after frozen storage were darker, less red and more yellow than the fresh meat. Freezing and frozen storage increased fluid loss and WBSF compared with the fresh meat, due to protein denaturation. Frozen storage affected sensory attributes by increasing fatty odour, frying flavour, sour flavour, fatty flavour and liver flavour, and by reducing juicy texture and mushy texture.

Keywords: meat colour; cooking loss; Warner-Bratzler shear force; tenderness; odour; texture; flavour; LF-NMR; water and lipid distribution

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1. Introduction

Lamb meat is traditionally regarded as a seasonal meat product in the Northern Hemisphere, due to the seasonal availability of the meat. The traditional lambing season is in the spring, due to the usual reproduction cycle of the ewe in temperate regions [1]. Hence, most of the lamb meat in northern hemisphere regions becomes available in autumn, and any surplus meat not sold fresh can be stored as frozen until the market for fresh meat has declined. Freezing of meat has given markets an opportunity to offer sales of lamb

all year round, which can result in a better market price for lamb meat. Furthermore, the surplus product does not have to be sold off at low prices when supply exceeds demand for fresh meat in the intensive slaughter season [2]. Therefore, freezing of meat can be used to stabilise the market and increase product flexibility [3,4], allowing producers to sell the meat at a better price and making lamb meat available to consumers all year around [5]. Meat is stored frozen to preserve the product and to keep the meat quality as high as possible [6]. The impact of freezing on meat quality has been reviewed by Leygonie et al. [7], who concluded that freezing has well-documented effects on moisture loss in meat, but that the literature is inconsistent about the combined effects of freezing and thawing on other parameters, such as colour and tenderness. Studies have also reported increased moisture losses with increasing frozen storage time in both lamb [8] and beef [9]. However, the effect of frozen storage on the sensory properties of lamb meat has been shown to be small when assessed by a trained panel, and should not affect consumer acceptance for the product [10]. Differences between fresh and frozen meat have been reported when untrained consumer panels assessed lamb meat for tenderness, flavour and overall acceptability [10,11]. When studying differences between fresh and frozen meat, it is important to consider how the freezing process affects meat quality attributes. For example, Luyet [12] showed that ice crystals are formed differently depending on whether the freezing is fast or slow. When the rate of freezing is slow, ice crystals form in large bundles between muscle fibres, whereas during rapid freezing ice crystals form both within and between cells [12]. Petrović et al. [13] observed a clear increase in cooking loss for both fast and slow freezing rates compared with unfrozen meat. In that study, rapid freezing procedures resulted in more intracellular ice crystals that were smaller in size, which thereby reduced the weight losses (thawing and cooking) of the meat compared with slower freezing procedures [13]. Differences in crystal formation contribute to the fluid loss after thawing and cooking, due to more or less disruption to the fibre structure, which enables water to leave the muscle cells [12]. This water cannot be rebound to proteins during the thawing process, and is hence lost as thawing loss [13]. Freezing has been shown to improve shear force values [14] or to cause no detectable differences in lamb meat [11]. However, the combined effect of chilling and frozen storage has not been fully evaluated according to a review by Coombs et al. [15], with few studies in particular analysing the effects of longer chilling storage combined with frozen storage exceeding 3–4 months. The aim of the present study was therefore to evaluate the combined effect of chilling and freezing storage, and whether frozen lamb meat displays differences in meat quality attributes compared with fresh meat. The hypothesis was that regardless of chilling regime, freezing would lead to a higher fluid loss which influences meat quality.

2. Materials and Methods

A detailed description of the experimental design, including animals, slaughter systems, sample treatments and experimental analyses, is provided in a companion paper by Stenberg et al. [16]. In brief, 10 pairs of intact ram lamb twins originating from the same farm and of the Icelandic sheep breed were used in this study. Ten lambs (one from each pair) were slaughtered at a large-scale abattoir and the remaining 10 were slaughtered at a small-scale abattoir. Both abattoirs kept the lambs in lairage for 10–12 h overnight before slaughter. During lairage, all animals had access to water, but no feed. All lambs were on average 160 days old at slaughter and had an average hot carcass weight of 19.8 $\pm$ 1.4 kg (mean $\pm$ st. dev.). All animals were hung by the Achilles tendon after dressing. Under Icelandic legislation (Slaughter Regulation Act 461/2003, Animal Welfare Act 55/2013, Quality Controlled Sheep Farming Act. 1160/2013), no ethical approval was needed before execution of the study.

2.1. Slaughter Facilities

Ten lambs were slaughtered at a small-scale abattoir slaughtering about 75 lambs/day, and the other 10 were slaughtered at a large-scale abattoir slaughtering 2500 lambs/day.

The small-scale abattoir used captive bolt stunning and kept the carcasses at 10–15 °C during the first six hours after slaughter, followed by chilling at 3–4 °C for 30 h, at which point samples of *M. longissimus thoracis et lumborum* (LTL) were removed. The large-scale abattoir used electrical stunning and applied electrical stimulation to all carcasses prior to entering the chiller, and the carcasses were hung for 30 h at 2–4 °C before sampling of LTL.

2.2. Sampling, Packaging and Handling of LTL Muscle

Both LTL muscles, including subcutaneous fat, were removed from all carcasses at the location between the last lumbar vertebra and the seventh thoracic vertebra. The LTL muscle from the left side was labelled as fresh and the muscle from the right side was labelled as frozen. All muscle samples were vacuum-packed in 25 cm × 35 cm bags. Two samples from the large-scale abattoir were unfortunately frozen by mistake and could not be analysed as fresh meat. Each LTL muscle, after removal of subcutaneous fat, was divided into parts to provide samples for the different analyses. Colour and NMR analyses were done on uncooked meat, and sensory analysis and WBSF were done on cooked meat. The cooking procedure was standardised as sous vide cooking for one hour (Anova Precision Cooker, Anova Culinary Inc., San Francisco, CA, USA) at 68 °C, and then flash-frying at a high temperature on each side for 30 s. Slices of 1.0 cm of the most posterior part of uncooked *M. longissimus lumborum* (LL) were used for NMR samples. Slices of 3.0 cm of cooked anterior par of *M. longissimus thoracis (LT)* were used for WBSF samples and the remaining part of LL was used for sensory analysis.

2.3. Fresh Samples

The fresh samples were stored at 2–4 °C and aged for 6–7 days before analysis.

2.4. Frozen Samples

The frozen samples were subjected to two different procedures after slaughter, based on standard operations at the two different abattoirs. The samples from the small-scale abattoir were aged for four days at 2 °C before samples were frozen at −24 °C and stored for three months. The samples from the large-scale abattoir were frozen on the day after slaughter, and then kept at −24 °C for three months. All frozen samples were thawed at 4 °C overnight before analysis.

2.5. Colour

Colour measurements were carried out on the fresh meat before vacuum packaging and cooking, and on the frozen meat after vacuum packaging and thawing. Each sample was allowed to bloom for one hour at 20 °C before the colour was measured in triplicate for each muscle sample. All samples were analysed with a Minolta CR-300 colorimeter (Konika Minolta, Tokyo, Japan) with a D65 light source. The colour measurements provided information about the lightness (L* value), redness (a* value) and yellowness (b*) of the muscle samples.

2.6. Thawing and Cooking Loss

All fresh samples were weighed before and after the cooking process, to calculate the loss after cooking (%). The frozen samples were weighed as frozen and after cooking, to calculate the combined loss of fluid from both thawing and cooking.

2.7. Warner-Bratzler Shear Force (WBSF)

Muscle tenderness was measured instrumentally on cooked samples of LTL by a Warner-Bratzler knife (TA-7) comprising a guillotine block coupled to a texture analyser (TA.HD Plus Connect, Godalming, Surrey, UK), which moved at a speed of 2.5 mm/second. The sample size for analysis was 1.0 cm × 1.0 cm × 3.0 cm (width × height × length). Samples were cut orthogonally to the muscle fibre direction, and each WBSF value was derived from quadruple measurements for each muscle sample.

2.8. Sensory Evaluation

A trained panel (generic descriptive analysis) [17] consisting of 6–10 panellists participated in each of a total of five testing occasions, depending on the availability of individuals. Each sensory session evaluated four different samples. To check panel performance and the performance of the individual assessors, PanelCheck V1.4.0 (Nofima, Tromsö, Norway) software was used. In each session, each panellist was given one 2 cm thick sample from the same location within each LTL muscle, which means that each LTL was divided into 10 different test samples. Samples were still warm when presented to the panellists in individual aluminium containers for each sample. The test samples were numerically coded with randomised numbers and were presented randomly to the panellists. Each panellist carried out their testing in a separate cubicle with standardised light, where they were given crackers and water to be consumed between samples to avoid residual flavour contamination. The evaluation was carried out for the following sensory traits: odour (frying, sour, fatty and liver), flavour (frying, sour, fatty, sweet and liver) and texture (softness, tenderness, juiciness and mushiness), which were rated on a linear scale from 1 to 100 for each trait. Before the actual test the panellists were calibrated at two panel training sessions to select and define the same specific characteristics and intensity of each individual sensory attribute. The defined sensory attributes to describe odour, appearance, flavour and texture were then used in the study to evaluate the meat. A more detailed description of the attributes can be found in Table S1 in the Supplementary Materials.

2.9. Low Field Nuclear Magnetic Resonance (LF-NMR)

Samples of the fresh meat and frozen-thawed uncooked meat were analysed by low field nuclear magnetic resonance (LF-NMR) at ambient temperature (20 ± 1 °C). A Minispec mq 20 benchtop NMR analyser (Bruker Optics, Rheinstetted, Germany) was used to analyse the water and lipid distribution in the meat muscle through transversal relaxation time analysis. Three samples (approximately 1 g) were cut from each muscle, transferred individually to 10 mm NMR sampling tubes and placed inside the magnet. The Carr-Purcell-Meibook-Gill (CPMG) pulse sequence [18,19] was applied, with an interpulse spacing of 100 µs, 8000 echoes, a recycle delay of 10 s and 16 repetitive scans. NMR relaxation time data were collected using Bruker Minispec software (Bruker Optics, Rheinstetten, Germany) and then further analysed in Matlab (Mathworks Inc., Natric, MA, USA).

2.10. Statistical Analyses

Technological meat quality and sensory data (Tables S2 and S3) was analysed by Proc Mixed in the Statistical Analysis Software (SAS) [20], with twin pair of lambs as random effect, in the model:

$$\text{Model: } Y_{ijk} = \mu + S_i + F_j + SF_{ij} + P_k + e_{ijkl}$$

where Y_{ijk} is the dependent variable, μ is the grand mean, S_i is the fixed effect of slaughter system, F_i is the fixed effect of fresh or frozen (sample treatment), SF_{ij} is the interaction between slaughter system and sample treatment, P_k is the random effect of twin pair, and e_{ijkl} is the residual error.

A general Satterwaite approximation for the denominator degrees of freedom was performed, using the SATTERTH option in SAS. Differences were considered significant at $p \leq 0.05$, and marginal significance was assumed at $0.05 < p \leq 0.10$.

The relaxation data (Tables S4 and S5) were maximum-normalised prior to further analysis. The normalised data were analysed by principal component analysis (PCA) in Unscrambler X® (Camo AS., Oslo, Norway), and then fitted to a multi-exponential curve using the Low-field NMR toolbox for Matlab, as described by Pedersen et al. [21]. PCA was performed in Unscrambler X (Camo AS, Oslo, Norway) on all data to find similarities and differences between samples from the two treatments. The data were centred and weighted with the inverse of the standard deviation of each variable, to correct for the use of different units.

Results presented directly in the text below are least squares means ± standard deviation.

3. Results

3.1. Technological Meat Quality Attributes

An effect of freezing compared with fresh meat was found in colour measurements, cooking loss and WBSF (Table 1). There was a tendency (p = 0.0654) for a significant difference in lightness (L*) between fresh and frozen meat, where the fresh meat was lighter, i.e., had higher L* value, than the frozen meat (Table 1). A difference (p = 0.0080) was also found in redness (a*), where the fresh meat was redder, i.e., had a higher value than the frozen meat (Table 1). The parameter cooking loss also showed a significant difference (p = <0.0001), with the frozen meat displaying a greater loss than the fresh meat (Table 1). There was a tendency for significance in WBSF, where the frozen meat had a higher shear force value than the fresh meat (Table 1).

Table 1. Colour parameter values, cooking loss and shear force values (WBSF) of lamb meat tested fresh and after frozen storage for three months (mean ± standard deviation).

Parameter	Fresh, n = 18	Frozen, n = 20	SEM [1]	*p*-Value [2]
Lightness (L*)	37.5 ± 1.7	36.7 ± 1.4	0.38	0.0654
Redness (a*)	19.5 ± 1.8	18.8 ± 1.4	0.37	0.0080
Yellowness (b*)	4.5 ± 1.2	6.0 ± 1.4	0.33	0.0006
Fluid loss (%)	15 ± 3.1	27 ± 3.3	0.70	<0.0001
WBSF (N) [3]	46 ± 13.1	51 ± 13.0	3.03	0.0837

[1] Standard error of the mean. [2] Differences considered significant at $p < 0.05$ and tending towards significance at $0.05 < p \leq 0.10$. [3] Warner-Bratzler shear force measured in Newtons.

Interactive effects were found for redness (a*) and WBSF. There were two interactions between slaughter system and meat treatment (fresh or frozen) on redness (a*) (p = 0.0011) and WBSF (p = 0.0269). The interactive effect on redness comprised an increase in redness in frozen (a* = 19.0) compared with fresh (a* = 18.8) meat for the small-scale abattoir and a decrease in redness in the frozen (a* = 18.5) samples compared with fresh meat (a* = 20.2) for the large-scale abattoir (Figure 1). The WBSF value from the small-scale abattoir decreased in frozen (47.6 N) compared to fresh (49.0 N) meat. However, an increase in WBSF was found in frozen (53.7 N) compared to fresh (43.4 N) meat from the large-scale abattoir (Figure 2).

3.2. Sensory Attributes

For some of the specific odour, flavour and texture attributes tested, an effect of freezing was observed (Table 2). A significant difference (p = 0.0229) was observed for the attribute fatty odour, where frozen meat had a higher score than fresh meat (Table 2). The attribute frying flavour was significantly (p = 0.0340) higher in the frozen than the fresh meat (Table 2). Similar differences were seen in the attributes sour flavour (p = 0.0204), fatty flavour (p = 0.0003) and liver flavour (p = 0.0222), which all showed higher scores for the frozen meat than the fresh (Table 2). There was also a difference in terms of juicy texture (p = 0.0001) and a tendency for a significant difference in mushy texture (p = 0.0714), with both attributes showing higher scores for fresh meat compared with frozen meat (Table 2).

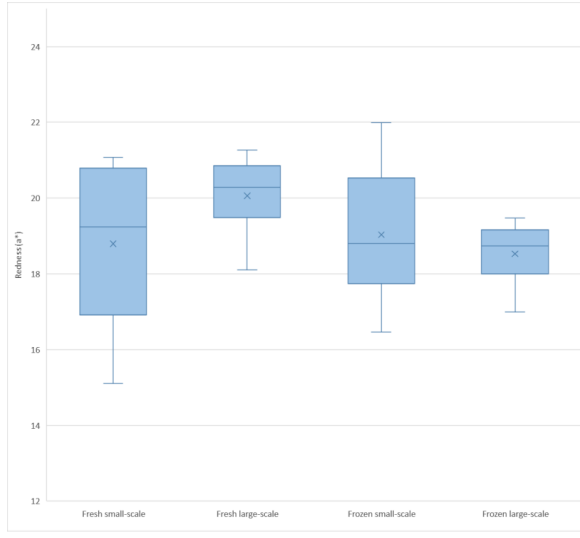

Figure 1. Boxplot showing redness (a*) for fresh and frozen meat samples from the small-scale and large-scale abattoirs.

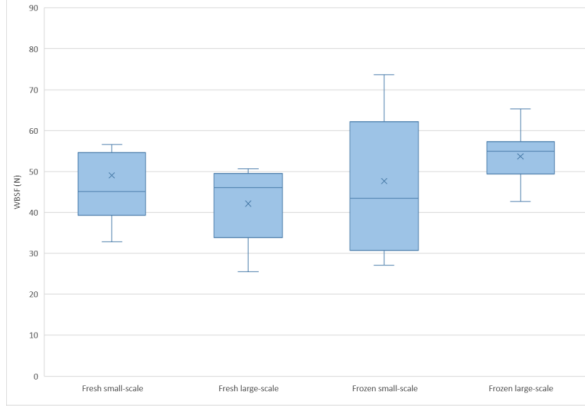

Figure 2. Boxplot showing Warner-Bratzler shear force (WBSF) in Newtons (N) for fresh and frozen meat samples from the small-scale and large-scale abattoirs.

Table 2. Results of sensory analyses comparing muscle samples from fresh meat and meat after frozen storage for three months.

Parameter		Fresh, n = 18	Frozen, n = 20	SEM [1]	*p*-Value [2]
Odour attributes [3]	Frying	33 ± 8.7	36 ± 6.4	1.78	0.2465
	Sour	12 ± 3.8	14 ± 2.9	0.82	0.1043
	Fatty	27 ± 3.2	32 ± 7.0	1.38	0.0229
	Liver	30 ± 4.6	32 ± 4.4	1.05	0.1665
Appearance attribute [3]	Colour	31 ± 4.7	30 ± 6.8	1.44	0.4780
Flavour attributes [3]	Frying	24 ± 4.5	29 ± 6.8	1.33	0.0340
	Sour	25 ± 5.1	31 ± 8.0	1.55	0.0204
	Fatty	16 ± 3.5	22 ± 4.8	0.98	0.0003
	Sweet	9 ± 1.6	9 ± 1.8	0.40	0.6240
	Liver	41 ± 5.2	45 ± 5.3	1.38	0.0222
Texture attributes [3]	Soft	50 ± 13.9	53 ± 13.8	3.22	0.5586
	Tender	48 ± 16.0	50 ± 14.8	3.71	0.6816
	Juicy	49 ± 9.6	35 ± 11.2	2.40	0.0001
	Mushy	16 ± 4.7	14 ± 3.7	1.00	0.0714

[1] Standard error of the mean. [2] Differences considered significant at $p \leq 0.05$. [3] Sensory attributes were scored on a scale from 0–100, with 100 being the highest score.

3.3. LF-NMR Analysis

The transverse relaxation time curves were collated and analysed by PCA, to identify sample similarities and variations in water and lipid characteristics and their distribution throughout the muscle samples (Figure 3). The first two principal components in the PCA plot explained 92% of the variation between samples. Sample groupings showed a clear distinction between fresh and frozen lamb meat samples (Figure 3). A more confined cluster of the frozen samples compared with the fresh samples indicated that the between-sample variation in water and lipid distribution and characteristics was smaller after freezing than in the fresh meat samples.

To explain the effects of freezing and to identify potential differences in water distribution in the meat samples, the relaxation time data were fitted to a multi-exponential curve. Proton relaxation analysis identified three proton populations in the meat samples (Table 3). In agreement with earlier LF-NMR studies on muscle-based foods [22–24], the origin and allocations of these proton populations were interpreted as follows: the proton distribution indicated the presence of a small water population A_{2b} (approximately 10% of all water) with translational relaxation parameter T_{2b} of approximately 10–16 ms in all meat samples (Table 3). This population is believed to relate to water molecules in close association with macromolecules in the meat. A dominant population A_{21}, corresponding to approximately 80% of the identified protons, with an approximate relaxation time of 40–50 ms was observed in all meat samples. This population mainly correlates to restricted water in myofibrillar cells and lipids within the muscle. Finally, a third population T_{22} was observed, contributing to 7–14% of the protons, with relaxation times in a wider range. This population is believed to correspond to less restricted water within the muscle, or extra-myofibrillar water [22–24].

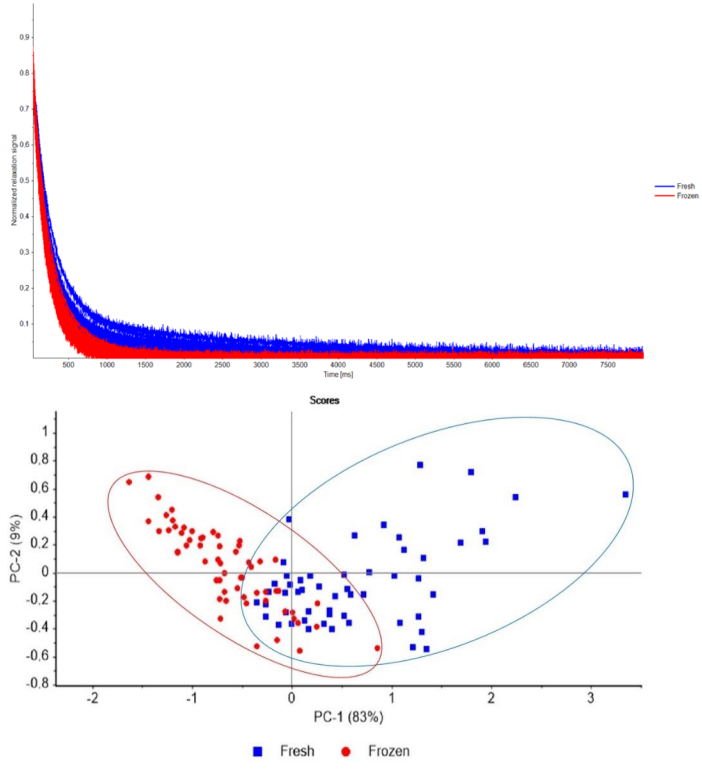

Figure 3. (**upper panel**) Normalised transversal relaxation curves obtained with LF-NMR for fresh and frozen lamb meat samples, and (**lower plot**) principal component analysis (PCA) plot of these relaxation curves. The blue ellipse indicates the grouping of fresh meat samples, and the red ellipse indicates frozen meat samples.

Table 3. Transverse relaxation times (T_{2i}, i = [b, 1, 2]) and apparent proton populations (A_{2i},) obtained through multi-exponential fitting of the LF-NMR relaxation data for the fresh and frozen lamb meat samples (mean ± standard deviation; n = 18 for fresh, n = 20 for frozen).

Parameters	Fresh, n = 18	Frozen, n = 18	SEM [1]	*p*-Value [2]
	Translational relaxation parameters			
T_{2b} (ms)	13.6 ± 6.9	12.1 ± 8.8	1.92	0.5972
T_{21} (ms)	48.7 ± 4.3	41.0 ± 4.3	1.05	<0.0001
T_{22} (ms)	224.1 ± 89.3	105.5 ± 20.4	15.8	<0.0001
	Water population within muscle sample			
A_{2b} (%)	9.1 ± 7.5	9.0 ± 14.5	2.80	0.9657
A_{21} (%)	81.5 ± 7.2	78.0 ± 13.2	2.59	0.3549
A_{22} (%)	9.4 ± 2.1	13.0 ± 3.9	0.75	0.0029

[1] Standard error of the mean. [2] Differences considered significant at $p < 0.05$ and tending towards significance at $0.05 < p \leq 0.10$.

Correlation analysis revealed medium negative correlations between the relaxation times and fluid loss ($r = -0.532$ and $r = -0.598$ to T_{21} and T_{22}, respectively) and medium positive correlations between the extra-myofibrillar water proportion A_{22} and fluid loss

($r = 0.466$). Correlation analysis of the LF-NMR and textural and sensory data revealed medium negative correlations between T_{2b} and soft ($r = -0.539$), and tender texture ($r = -0.496$).

4. Discussion

This study evaluated differences between fresh and frozen lamb meat in terms of technological meat quality and sensory attributes. The decision to include meat from large-scale and small-scale slaughter treatments in the fresh and frozen groups was based on common practice in current commercial production. This is therefore the most accurate way to assess how meat available on the market could be affected by freezing and frozen storage. The post mortem handling of samples before freezing differed between the two slaughter systems covered by the study. These differences in post-slaughter treatment could have had an effect on quality characteristics such as colour stability and water loss, based on findings by Choe et al. [25] in a study investigating the effect of ageing meat at different temperatures and for different periods before freezing. However, no such differences were detected in this study.

4.1. Fluid Losses

Fluid losses occurred, as combined thawing and cooking losses, and affected several sensory and physicochemical attributes, such as juicy texture. Fluid losses and juicy texture were both affected by freezing, with the frozen samples showing greater losses after thawing and cooking and perceived to be less juicy by the sensory evaluation panel. The sensory attribute juicy texture is discussed below together with the other sensory attributes. The literature describes different types of fluid losses, such as drip loss, thawing loss, cooking loss, fluid loss, water loss or changes in water-holding capacity (not defined further) or combinations of two or several of these parameters [8,9,26–29]. In the present study, losses of fluids were measured as combined thawing and cooking losses, although the individual effect of these parameters could be examined in future studies in order to distinguish between the fractions. The results in the present study are compared with those of previous studies reporting any type of loss of fluids, or not, in relation to freezing compared with unfrozen meat. Previous studies have shown that freezing can have an increasing effect on fluid losses from lamb meat [8,29], which is supported by the results in the present study. Similar findings have been reported for beef meat, by Vieira et al. [9] observing increased fluid losses as a results of freezing. Muela et al. [29] did not find differences in cooking loss between fresh and frozen meat, with the explanation that differences may have occurred in thawing losses instead of cooking losses. The thawing losses were greater for the frozen compared with the unfrozen meat samples in that study, and hence the total fluid losses were greater for the frozen samples [29]. Bhattacharya et al. [27] ascribed drip loss to protein denaturation by the high ionic strength of extracellular fluid as an effect of freezing, resulting in the protein losing its water-holding capacity [27,29]. Since protein denaturation affects water-holding capacity negatively, this may be an explanation for the increased fluid loss observed after freezing in the present study. Another explanation could be that ice crystallisation, which occurs when meat is frozen, can damage cell structures and lead to increased fluid losses when the meat is thawed [12,13].

4.2. Colour

When consumers are choosing meat in the supermarket, meat colour is one of the most important quality measures they consider. In particular, the redness (a* value) is often identified as the most important measure, since consumers often associate a certain degree of redness with meat that is safe to eat [30]. However, whether red colour can be used as the single most valid method for evaluating safe meat has not been established within the scientific community. The results in this study revealed a change in colour as an effect of freezing, as well as an interactive effect between slaughter systems and sample treatment (fresh/frozen) on redness. Comparing meat from the fresh and frozen groups,

the frozen and thawed samples tended to have darker, less red and more yellow colouring than the fresh meat. An effect of freezing on meat colour has been described in previous research [10,25,29,31]. The decrease in lightness (L*) in the thawed meat compared with the fresh in our study supports previous findings [29,31,32]. Farouk et al. [28] attributed this reduction in L* to higher thaw drip in slowly frozen beef samples, leading to greater light reflection and thereby a lighter colour compared with fast-frozen beef samples. Since the frozen meat in the present study had greater fluid losses after cooking, the explanation suggested by Farouk et al. [28] may be valid also for our results. The observed reduction in redness (a*) might be explained by reduced activity of metmyoglobin-reducing enzymes, leading to accumulation of metmyoglobin that was visually apparent as reduced redness in the frozen samples compared with the fresh [32]. The decrease in redness (a*) might therefore be due indirectly to the loss of fluids in the frozen samples after thawing and cooking. However, differences in redness (a*) are not always found between fresh and frozen meat, e.g., Muela et al. [29] did not find differences in redness (a*) between fresh and frozen meat when looking at different freezing methods and durations of frozen storage. The results from that study suggest that redness (a*) is not exclusively affected by freezing, a suggestion supported by Fernández et al. [33] who claim that freezing protects meat from decolouration, resulting in recovery of meat colour after thawing and thereby no visible differences in meat colour. When meat is less light (L*) and less red (a*), the yellow colour (b*) may be more prominent, and the increase in b* in our frozen samples could therefore be an indirect effect of the decrease in redness (a*). Another theory is that increased yellowness (b*) is due to oxidation and yellowing of fat, as suggested by Moore and Young [30] on examining the effect of storing meat under display film in terms of e.g., discolouration. The intramuscular fat content in the lamb meat analysed in this study was not measured, so we offer this as a speculative theory, rather than an explanation. Variation in yellowness (b*) in pork muscle has previously been described as an effect of variation in fractions of deoxymyoglobin and oxymyoglobin, as well as an effect of internal reflectance [34]. Since there was a change in both lightness (associated with internal reflectance) and redness (associated with the different myoglobin forms) between fresh and thawed samples in this study, this could explain the difference in yellowness. However, the numerical differences in L*, a* and b* between the fresh and frozen sample groups were quite small, and should have only a low impact when consumers are making an assessment with the human eye before prospective purchases. The differences in meat colour may affect consumer acceptance of lightness and redness, according to previous research [34]. Khliji et al. [35] found that when redness (a*) is $\geq$9.5 and lightness (L*) is $\geq$34, consumers would regard meat colour as acceptable based on colour parameters. The significant differences seen in the colour parameters examined in the present study lead us to conclude that there were differences between the fresh and frozen meat samples, but the small numerical differences indicate that these differences were not undesirable in either treatment from a consumer perspective. The same conclusions can be drawn on examining the effect of the interactive difference in redness. The differences in ageing between the two slaughter systems tested could have affected colour stability, as seen in previous work [30]. However, the numerical differences in redness between slaughter systems and sample treatments were small, and should not pose a risk of consumers reacting negatively when visually assessing the meat in a purchase situation.

4.3. WBSF

There was a tendency for a difference in WBSF between the fresh and frozen meat, with the frozen meat having a higher value. Previous research has found contrasting effects on WBSF on comparing frozen meat with fresh. For example, Duckett et al. [36] did not detect any difference in shear force (SF) between fresh and frozen meat from normal lambs, but observed a decrease in SF following freezing for meat from lambs with the callipyge gene. In contrast, Smith et al. [6] found an increase in SF in frozen meat compared with fresh and Muela et al. [31] observed an increase in WBSF in meat after frozen storage

for 15 or 21 months compared with fresh meat. However Muela et al. [31] observed no difference in WBSF in meat stored frozen for one or nine months compared with fresh meat [31]. Another study reported a decrease in WBSF values as an effect of increased frozen storage [9]. Hence, it may not be correct to state unequivocally that freezing and frozen storage increases or decreases WBSF, since many factors influence the outcome. Such factors can include frozen storage duration [31], freezing rate [3], and a combination of ageing before frozen storage and storage time [9]. With this in mind, it may not have been the freezing and thawing itself that affected the WBSF in frozen meat compared with fresh in this study. The effect seen in the present study was a tendency to significance for WBSF, which did not correspond to the results of the sensory testing where the panellists did not detect a difference between fresh and frozen meat in the attribute tender texture. It can therefore be concluded that the tendency to significance for WBSF may not have been detectable by consumers, which could be seen as a positive result when evaluating the freezing of meat.

Since WBSF is a method for evaluating tenderness, it is important to reflect on the numerical values obtained when discussing the results. As shown in Figure 2, all mean values of WBSF were higher than 40 N and some even over 50 N. Previously published work has suggested a threshold of $\leq$5 kg (~49 N) for meat to be considered acceptably tender [37]. With this information in mind, it is reasonable to assume that all meat samples from the present study except the frozen samples from the large-scale abattoir would be rated acceptably tender from a WBSF perspective. It could also be stated that the WBSF results indicate that all meat samples within the present study are in the upper range, or above the threshold for acceptably tender meat. The differences detected for WBSF were not reflected in the sensory attribute texture tenderness, for which the sensory panel did not find a difference between fresh and frozen meat samples. This could be interpreted as a broader tolerance than 49 N, since the mean WBSF value was 46 $\pm$ 13.1 N for fresh and 51 $\pm$ 13.0 N for frozen meat.

The interactive effect on WBSF, with an increase in WBSF after freezing in meat from the large-scale abattoir compared with fresh meat and a decrease in WBSF after freezing in meat from the small-scale abattoir compared with the fresh samples, may have been influenced by differences in the ageing regime before freezing the meat. Meat from the small-scale abattoir was aged for four days before frozen storage and for six days before the fresh meat was tested. In comparison, meat from the large-scale abattoir was aged for one day before frozen storage and six days before the fresh meat was tested. This difference in ageing may partly explain the differences in texture between the abattoirs. The difference in WBSF between frozen and fresh meat was greater for the large-scale abattoir, and might be explained by the shorter ageing time. The reason for using different ageing regimes in the different abattoirs was, as previously mentioned, to mimic current production and enable valid comparison. It is thus important to note that increased ageing time before freezing in the large-scale abattoir could improve meat quality in terms of WBSF values.

4.4. Sensory Attributes

Quite a few of the sensory parameters analysed were affected by freezing in the present study. The overall explanation for the increases in sensory properties, where flavour accounted for most attributes affected, could be loss of fluid after thawing and cooking of frozen meat. An explanation for the decreased juiciness scores proposed by Bueno et al. [38] is that freezing causes loss of water owing to disruption of cell structures, resulting in higher concentrations of flavour (in this study represented by frying, sour, fatty and liver) compounds in the samples. The decreased mushiness scores for the frozen samples may also have been an effect of the decreased juiciness due to loss of fluids. Another explanation relates to increased firmness and release of flavour compounds while chewing. According to Pouliot et al. [39], decreased firmness makes the chewing process shorter, which results in less release of flavour compounds. Since the WBSF values were quite high (51 $\pm$ 13.0 N) for the frozen group and there was also a decrease in juiciness, it may be reasonable to

assume that more chewing was required for these samples, increasing the release of flavour compounds, which may be the reason for elevated flavour scores of the thawed meat in this study. However, there were no differences in the sensory attributes of tenderness and softness between the samples, which could contradict the previous reasoning. Overall, the decreased juiciness may have caused the increase in flavour attributes in the present study, despite not influencing the scores for softness and tenderness in the frozen samples. Further, the sensory panellists could not detect any differences in the colour of the cooked meat, which could also be connected to the small numerical differences obtained for the colour measurements of lightness, redness and yellowness. The sensory attribute fatty odour increased in the frozen samples compared with the fresh, possibly due to lipid oxidation during frozen storage, supporting findings by Muela et al. [31] of an increase in lipid oxidation with increased storage time up to 15 months. Lipid oxidation was not measured in the present study, however, so the cause of the increased fatty odour for frozen samples cannot be established with certainty. It is possible that a form of lipid oxidation accrued during storage in the frozen samples and resulted in the increase in fatty odour when assessing the samples for sensory attributes. Other studies focusing on sensory properties of fresh and frozen lamb meat have found that most attributes are unaffected when the meat is assessed by a trained sensory panel [10,11]. The only difference found in previous work was for tenderness scores, which increased for nine months of storage compared with fresh meat and meat frozen for 1, 15 or 21 months [10]. The lack of effect on tenderness and juiciness in previous work was attributed to relatively fast freezing rates and also to the absence of temperature fluctuations during storage [11]. This theory is supported by findings on the effect of fluctuating temperatures during frozen storage, with negative effects in terms of rancidity if meat is stored frozen at higher temperatures (−5 or −10 °C) before the ultimate freezing temperature (−35 °C) is established [40]. Both Muela et al. [10] and Muela et al. [11] concluded that the lack of differences between fresh and frozen lamb meat in terms of sensory attributes means that consumers should not be concerned about buying frozen or thawed meat due to decreased sensory quality. Although freezing influenced several sensory attributes in the present study, this does not mean that the differences were negative (or positive) from a consumer point of view.

4.5. LF-NMR Analysis

Some minor shifting trends between the proton populations were observed in the fresh and frozen meat. However, the only statistically significant difference in A_{2i} parameters was an increase in A_{22} after freezing the samples, indicating that the proportion of water within the extra-myofibrillar space of the muscle increased during freezing. Further, the frozen storage increased restriction of water, as indicated mainly by the sharp decrease in the T_{22} relaxation time parameter, corresponding to extra-myofibrillar water. This indicates that freezing led to a proportional shift in water from the myofibrillar space to the extra-myofibrillar cells, but also that restriction of the extra-myofibrillar water increased during frozen storage.

An increase in the transverse relaxation rate ($R = T_2^{-1}$) has been shown to correlate well with heat-induced protein denaturation of whey proteins [41], and with protein aggregation and denaturation during cooking of shrimp [42]. The observed decrease in relaxation times (T_{21} and T_{22}) in this study thus indicates that the lamb muscle was partially degraded or denatured during frozen storage, and that the T_{22} parameter was especially sensitive to frozen storage-induced changes in muscle structure. Pearson's correlation analysis showed medium negative correlations between the relaxation times and cooking loss, and medium positive correlations between the extra-myofibrillar water proportion A_{22} and cooking loss, indicating that freezing had a reducing effect on the water-holding ability of the muscle. This agrees with findings by Straadt et al. [22] that cooking induces shrinkage of myofibrils and a concurrent increase in the extra-myofibrillar space due to heat-induced expulsion of water from the myofibrillar matrix. This could also be linked to the increase in fluid loss after frozen storage.

Correlation analysis of the LF-NMR and textural and sensory data showed medium negative correlations between T_{2b} and soft and tender texture as assessed during the sensory evaluation. However, no such correlations were seen between the NMR parameters and the instrumental texture analysis (WBSF), indicating that meat texture was not highly affected by the water distribution of the muscle, potentially due to the small variation in textural properties in the samples assessed in the study.

5. Conclusions

To freeze meat may cause a negative effect on several meat quality parameters. It would therefore be valid to recommend usage of fresh meat to avoid negative effects on meat quality caused by freezing. The practical usage of freezing to increase storage time of meat does however promote freezing as a method to use both today and in the future. It is therefore of utter importance to further study the effect of freezing of meat and how to optimize the procedures of frozen storage to promote a low negative impact on meat quality.

Supplementary Materials: The following are available online at https://www.mdpi.com/article/10.3390/ani12202830/s1. Table S1: Definitions of the sensory attributes tested. Table S2: Individual data on technological meat quality attributes of fresh meat (colour, cooking loss and WBSF) and sensory attributes (Small = small-scale abattoir, Large = large-scale abattoir). Table S3: Individual data on technological meat quality attributes of frozen meat (colour, cooking loss and WBSF) and sensory attributes (Small = small-scale abattoir, Large = large-scale abattoir). Table S4: Translational relaxation parameters and water population within muscle samples from fresh meat (Small = small-scale abattoir, Large = large-scale abattoir). Table S5: Translational relaxation parameters and water population within muscle samples from frozen meat (Small = small-scale abattoir, Large = large-scale abattoir).

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Institutional Review Board Statement: Under Icelandic legislation, no ethical approval was needed (Slaughter Regulation Act 461/2003, Animal Welfare Act 55/2013 and Quality Controlled Sheep Farming Act 1160/2013): Ethical review and approval were waived for this study since the treatment of the lambs was the same as for other lambs at the farm and in the abattoirs. All lambs were from a normal production system and nothing was altered. We only sampled meat from the carcasses of the lambs the day after slaughter.

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What is meat quality? There is no easy answer, as many factors from field to fork influence final eating quality. This thesis examined the effects of different factors with rearing, slaughter and meat handling on lamb growth and on carcass composition, instrument-measured technological meat quality and sensory attributes of meat from intact male lambs. Carcass and meat quality attributes were shown to be affected by differences in feeding, slaughter and storage method.

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