Bruising in slaughter cattle: its relationship with creatine kinase (CK) levels and meat quality

By

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Declaration

I, **Mpakama Thandiswa**, vow that this dissertation has not been submitted to any University and that it is my original work conducted under the supervision of Prof. V. Muchenje. All assistance toward the production of this work and all the references contained herein have been duly accredited.

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Abstract

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The objective of the study was to determine the effects of pre-slaughter conditions and anima-l related factors on bruising, creatine kinase (CK) and beef quality. Three hundred and twenty one cattle from three breeds (108 Bonsmara, 130 Beefmaster and 83 Brahman) were used in this study. The animals were grouped according to age categories as follows; Group 1(16months), Group 2 (18months) and Group 3 (24 months). Blood samples for CK determination were collected at exsanguisation using disposable vacutainer tubes. The *Muscularis longisimus thoracis et lumborum* (LTL) muscle was used to determine beef colour (L,* a* and b^*) and ultimate pH. Significant P (<0.05) breed effects were observed on bruising score and CK levels with Bonsmara breed having the highest percentage (80%) of bruising score and higher CK (705.3±80.57) values. The higher CK levels were also in winter season. The effect of breed, sex and age at slaughter on meat quality (pHu, L*, a*, b*) was also observed. There were positive correlations between distance travelled and meat quality, while there was no relationship observed between CK and distance travelled. Therefore, it was concluded that animal related factors had an effect on meat quality and CK levels.

Keywords: Age at slaughter, beef quality, bruising, colour, creatine kinase, pH, season, transportation.

List of abbreviations

a* = redness

b* = yellowness

CK = creatine kinase

L* = lightness

LTL = *Muscularis longisimuss thoracis et. Lumborum*

pHu = ultimate pH

SAS = Statistical Analysis System

U/1 = Units per litre

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Chapter 1. General Introduction

Transportation of live animals is usually a major stress. Beef cattle are exposed to physical and physiological stimuli stressors through human-animal or animal interactions when transported to slaughter, auction markets or to farms (Weeks *et al*., 2002). Common vehicles used to transport these animals are trucks. Besides the issue of welfare, a strong relationship exists between pre-slaughter cattle management and carcass damage that was reported by many authors resulting in the production of bruises and the release of some muscle enzymes such creatine kinase (Grigor *et al*., 1997; Knowles *et al.*, 1994). The important factors in relation both to animal welfare and meat quality are loading, stocking density and road condition (Tarant *et al*., 1992). In addition long distances travelled by the animal increases stress and physical damage such as bruising in animals (Villarroel *et al.,* 2001; Gallo *et al*., 2003; Hoffman *et al*., 2010). Of all the pre-slaughter events mentioned above, transportation causes more stress and even predisposes animals to bruising (Knowles, 1999).

A bruise is defined as a tissue injury with rupture of the vascular supply and accumulation of blood and serum (Gracey *et al*., 1999; Hoffman *et al*., 1998). Occurrence of bruising starts from the farm, during transportation, at the market and at the slaughter house (Jarvis *et al.*, 1995). It results from a physical blow from a stick, a stone, a metal projection or an animal fall (Chambers, Grandin, Heinz and Srisuvan, 2004). Muscle damage of animal during transportation results in the release of CK in the blood.

It means that bruising in animals can be detected by measuring the amount of CK present in the blood plasma (Braun, Medaille and Trumel, 2008). Creatine phosphokinase (CPK) is used as a prognostic indicator for the uterine torsion. It acts as a catalyst in the conversion of the creatine which consumes Adenosine Tri-Phosphate (ATP) to make Phosphocreatinine and Adenosine Di-Phospahte (ADP) (Radostits, Arundel, Gay, Hinchcli and Blood, 2000).

Bruising in cattle affects the quality of the carcass and results in meat of poor keeping quality because the bruised sites offer an environment suitable for microbial growth and the meat is undesirable to consumers (Gallo, 2008; Hoffman, 2010). It is also postulated that bruised cattle are stressed and would have an abnormally high pH because of glycogen depletion and the subsequent lower production of lactic acid in the muscles (Kannan, Chawan, Kouakou and Gelaye, 2002). Therefore high pH apart from favoring microbial growth and reduction of shelf life of meat, will also cause meat to be darker and tough (Chambers *et al*., 2004) . Therefore it is clear that some kinds of damage affecting welfare result in creatine kinase release, so this can be used in conjunction with other indicators as a welfare or bruising measure.

1.2. Problem statement

Research has been done to investigate the animal handling and welfare issues during transportation of animals from farms to commercial abattoirs. Although there is little or no information on the relationship between creatine kinase (CK) values with pH and beef colour. In addition many studies have been done to determine the occurrence of cattle bruises from the farm to the abattoir but most of them have not been able to link the level of bruising with creatine kinase (CK) values. Poor welfare of animals increases stress in animals before slaughter and may lead to increased mortality and bruises. Therefore, occurrence of bruising caused by poor handling practices and road transportation, result in poor meat quality due to injuries such as hemorrhagic muscles and broken bones. Meat that is bruised is wasted as it is not suitable for consumption because it is not acceptable to the consumers. It cannot be used for processing or manufacture, it decomposes and spoils rapidly as the bloody meat is an ideal medium for growth of contaminating bacteria. For the above reasons, it must be condemned at meat inspection. Furthermore, if secondary bacterial infection occurs on the bruised muscle, abscess is formed causing the entire carcass to be condemned. This results to economic losses to farmers, transporters and slaughter houses (Mota-Rojas *et al.*, 2006). Furthermore, the season of slaughter has been shown to influence the welfare of beef (Gosalvez *et al*., 2006). Extremes in summer temperatures increase the risk of bruises on arrival (DOAs), the risk of pale soft exudative (PSE) (Dalla Costa *et al*., 2007) meat in pigs and turkeys, as well as dark cutting beef in cattle (Kadim *et al*., 2009).

1.3. Justification

Cattle that come from farms are equally exposed to various handling and transport conditions during marketing and before slaughtering. When these conditions are associated with rough handling, violent impact of the animals against protuberant or sharp-edged surfaces, or with aggression between animals, mechanical damage to the animal tissues can occur which may develop into a bruise. In addition, as animals are transported to the abattoir for long hours, , stocking density and at the lairage they might get injured. These conditions cause bruising and some kinds of damage which affect welfare results in creatine kinase releases. Besides the issue of bruising meat quality parameters such as pH, colour and tenderness are affected (Muchenje *et* *al*., 2009). Therefore this study on pre-slaughter conditions will create a better understanding on how animals must be handled from the farm up to the slaughterhouse. It is also hoped that the mixing of unfamiliar animals, inadequate ventilation, exposure to extreme temperatures, lack of food and water and delays before unloading will be taken into consideration as they are the critical points to be monitored during pre-slaughter conditions. The study also aimed to improve beef production and meat quality in a short term and also aims at finding ways of predicting beef quality through the use of CK.

1.4. Objectives

The broad objective of the study was to determine the effect of bruising in slaughter cattle, its relationship with creatine kinase (CK) levels and meat quality.

1.4.1. Specific Objectives

- To determine the effects of pre-slaughter conditions and animal related factors on bruising and creatine kinase (CK) levels in slaughter cattle.
- To determine the effects of distanced travelled and animal related factors on CK levels and meat quality of beef.

1.5. Specifics Hypothesis

- There are no effects of animal related factors and distance travelled on bruising in slaughter cattle and CK levels.
- Distance travelled does not have any effect on the CK levels and meat quality of beef.

1.6. References

Braun, J. P., Medaille, C. and Trumel, C. (2008). Clinical interpretation of enzyme activities and concentrations: A review of the main metabolic factors affecting variation. *Israel Journal of Veterinary Medicine, 63 (1),* 1-12.

Broom, D. M. (2000). Welfare assessment and welfare problem areas during handling and transport. In: Grandin, T (Ed). Livestock handling and transport livestock, 2nd Edition CABI Publishing, New York, pp 43-61.

Chambers, P. G., Grandin, T., Heinz, G. and Arisuvan, T. (2004). Effects of stress and injury on meat and by-product quality. In : Guidelines for human handling transport and slaughter of livestock. FAO Corporate documentary repository.

Dalla Costa, O. A., Faucitano, L., Coldebella, A., Ludke, J. V., Pelosa, J. V., dalla Roza, D. and Paranhos da Costa, M. J. R. (2007). Effects of season of the year, truck type and location on truck on skin bruises and meat quality in pigs. *Livestock Science, 107,* 29-36.

Gallo, C. B. (2008). Using scientific evidence to inform public policy on the long distance transportation of animals in South America. *Veterinaria Italiana, 44,* 113-120.

Gallo, C., Lizondo, G. and Knowles, T. (2003). Effects of journey and lairage time on steers transported to slaughter in Chile. *Veterinary Record, 152,* 361-364.

Gosalvez, L. F., Averos, X., Valderlvira, J. J. and Herranz, A. (2006). Influence of season, distance and mixed loads on the physical and carcass integrity of pigs transported to slaughter. *Meat Science, 73,* 553-558.

Gracey, J. G., Collins, D. S. and Huey, R. J. (1999). Meat hygiene, 10th edition. London: Elsevier Health Science Publishers, United Kingndom, pp.

Grigor, P. N., Goddard, P. J., MacDonald, A. J., Brown, S. N., Fawcett, A. R., Deakin, D. W., and Jarvis, A. M., Selkirk, L. and Cockram, M. A. (1997). The influence of source, sex class and pre-slaughter handling on bruising of cattle at two slaughterhouses. *Livestock Poduction Science, 43*, 215-225.

Hoffman, D. E., Spire, M. F., Schwenke, J. R. and Unruh, J. A (1998). Effect of source of cattle and distance transported to commercial slaughter facility on carcass bruises in mature beef cows. *Journal of the American Veterinary Medical Association, 212,* 668-672.

Jarvis, A. M., Selkirk, L. and Cockram, M. S. (1995). The influence of source, sex class and pre-slaughter handling on bruising of cattle at two slaughterhouse. *Livestock Production Science, 43*, 215-224.

Kadim, I. T., Mahgoub, O., Al-Marzooqi, W., Khalaf, S., Al-Sinawi, S. S. H. and Al-Amri, I. S. Effects of transportation during the hot season and low voltage electrical stimulation on histochemical and meat quality characteristics of sheep longissimus muscle. Livestock *Science*, *126*, 154-161.

Kannan, G., Chawan, C. B., Kouakou, B. and Gelaye, B. (2002). Influence of packaging method and storage time on shear value and mechanical strength of intramuscular connective tissue of cattle. *Journal of Animal Science, 80*, 2383-2389.

Knowles, T. G.(1999). A review of the road transport of cattle. *Veterinary Record*, *114*, 197-201.

Knowles, T. G., Warriss, P. D., Brown, S. N. and Kestin, S. C. (1994). Long distance transport of exports lambs. *The Veterinary Record*, *134*, 107-110.

Mota-Rojas, D., Becerril, M., Lemus, C., Sanchez, P., Gonzalez, M., Olmos, S. A., Ramirez, R. and Alonso-Spilsbury, M. (2006). Effects of mid-summer transport duration on pre-and post- slaughter performances and pork quality in Mexico. *Meat Science, 73*, 404-412.

Muchenje, V., Dzama, K., Chimonyp, M., Strydom, P. E. and Raats, J. G. (2009). Relationship between stress responsiveness and meat quality in the three cattle breeds. *Meat Science*, *81*, 653-657.

Radostits, O. M., Arundel, J. H., Gay, C. C., Hinchcli, F. F. and Blood, D. C. (2000). 9th Edition, Veterinary Medicine: A textbook of diseases of cattle, sheep, pigs and horses. *Elsevier Health Sciences*, pp 1426-1430.

Tarrant, P. V., Kelly, F. J., Harrington, D. and Murphy, M. (1992). Long distance transportation of steers to slaughter: effect of stocking density on physiology behaviour and carcass quality. *Livestock Production Science*, *30*, 223-238.

Villarroel, M., Maria, G. A., Sanudo, C., Sierra, I., Garcia-Belenguer, S. and Gebresenbet, G. (2001). Critical points in the transport of cattle to slaughter in Spain that may compromise the animal's welfare. *The Veterinary Record*, *149*, 173-176.

Weeks, C. A., McNlly, P. W. and Warriss, P. D. (2002). Influence of the design of facilities at the auction markets and animal handling procedures on bruising in cattle. *Veterinary Record*, *150*, 743-748.

Chapter 2. Literature Review

2.1. Introduction

Transportation stress is complex stressor involving temperature fluctuations, withdrawal from food and water, mixing with unfamiliar animals, and motion (Sutherland *et al*., 2009). The stress response to road transport in cattle will vary depending on the type of animal and conditions during the journey (Fergusson and Warner, 2008). Increasing time from the farm to the abattoir usually has a negative effect on meat quality Warriss (2000), with longer transport times increasing stress indicators such cortisol, creatine kinase, and lactate (Grandin, 2000). Research has shown that cattle transported for distances less than 400km are unlikely to have carcasses with above normal pH values, while cattle transported for distance greater than 2000km or durations more than 24 hours are likely to show pH values above normal (Eldridge and Winfield, 1988; Tarrant, 1989). The distance travelled by cattle to the abattoir and the occurrence of bruises seem to be positively correlated, with the level of bruising increasing with the distance travelled (McNally and Warriss, 1996).

2.2. Characterization of bruising

A bruise is defined as a tissue injury with rapture of vascular supply and accumulation of blood and serum (Hoffman, 1998). As soon as the tissue is damaged, a region of localized hypersensitivity occurs around the injury area. The hypersensitivity of the bruised area minimizes movement of the individual and contact with injury, until healing has occured. Thus, it has been interred that pain is promoter of repair (Basbaum and Woolf, 1999).

Bruises in cattle occur during the anter-mortem period but they can only be seen at slaughter due to thickness of the bovine skin. Bruises assessment is therefore a post mortem function and it is a retrospective reflection of all physically damaging events that may have occured prior to slaughter (Strappin *et al.*, 2009). Bruising in cattle is not only an indication of poor welfare, but can cause heavy financial loses (Jarvis *et al*., 1995; Grandin, 2000; Gallo, 2008) since bruised meat must be trimmed, downgraded or condemned depending on the severity of the bruises.

2.3. Beef cattle bruising and its causes during handling and transport

A bruise develops when force is applied to the skin by use of a blunt object, such as stone, metal projection, a stick or when an animal falls (Strappin *et al.*, 2009). There are numerous risks associated with transport and handling which all have the potential to cause bruising and poor welfare to animals. Critical areas in the meat production chain where bruising can occur include: the farm, during road transportation, at livestock markets, during loading and unloading and even during stunning procedures (Jarvis *et al.,* 1995). Handling at livestock markets has a significant contribution to bruising (Knowles *et al.*, 1999).

Other main factors influencing bruising during handling and transportation are stockman-ship, facility and vehicle design, driving style, mixing of animals before transport or in lairage, presence of horned animals and animal temperament (Grandin, 2000). No aceration is caused on the skin and therefore blood and serum from the damage vessels accumulate in the tissue Hoffman *et al.* (1998) which leads to pain, swelling and tenderness (Blood and Studdert, 1988).

In the event of handling and transport most is the results of physical blows from driving instruments like sticks, projecting objects in facility and trucks and animal falling (Chambers *et al.*,2004). Therefore the location as well as frequence and appearance of bruises vary widely and all three aspects give valuable clues as the frequence with which certain bruises appear on slaughter are indicative of specific handling or transportation and can be utilized to determine problem areas in the transport chain (Grandin, 1990, 2000; Jago *et al.,* 1996). The size and discoloration of a bruise can be used to determine the age of a bruise as this changes over time (Gracey and Collins, 1992**).**

2.4. Aging the bruises

Although the presence of bruises at slaughter is apparent to the eye, knowledge of the exact time of infliction is necessary if steps are to be taken to prevent bruising (Grandin, 1993). At slaughter a bruise maybe dated approximately by physical criteria. Estimating the age of a bruise can provide for the identification of the place and time of livestock damage and provides information about the cause (Fisher *et al.,* 1998).

The age of a bruise can also be estimated by measuring the electrical conductivity of the tissue, which increases up to a maximum at 40 hours (Edridge *et al.,* 1984). Bruises are separated into two categories; fresh bruises and those that are several days or weeks old. Fresh bruises do not have yellow mucous. Old (several days) which is responsible for yellow colour or have clear yellow mucous (Grandin, 2000). According to Langlois and Gresham (1991) and Langlois (2007) the yellowness in a bruise is considered to be more informative in bruise aging than the colours. The yellow bruise which is more than 18 hours is considered to be old (Gracey and Collins, 1992; Grandin, 2000; Langlios, 2007).

2.5. Impact of bruising on meat quality

Carcass damages such as bruising, haemorrhages, skin blemishes, blood plash and broken bones are common occurrences found due to improper conditions and increased distance travelled. Bruises or contusions are described as superficial discoloration due to haemorrhage into the tissue from raptured blood vessels beneath the skin surface, without the skin being broken. In the contusions the blood accumulates in surrounding tissues, producing pain, swelling and tenderness (Blood and Studdert, 1988). In a bruise, haemorrhage, skin blemish and or blood splash the skin of the animal and blood vessels may accumulate excessive blood which has to be trimmed off during processing.

Trimming part of the carcass off will reduce meat yield and value, expensive and increase processing time. Untrimmed parts have poor appearance and can serve as substrates for microbial growth causing the meat to spoil earlier than normal. Broken bones may cause bone splinters in meat and this will be dangerous to consumer if not detected after deboning (Warriss, 2000). Bruising has been a major concern in beef cattle. More bruising occurs at the handling phase at auctions or at the processors (Hoffman *et al.*, 1998; McNally and Warriss, 1996) as opposed to during transit. Hoffman *et al.* (1998) reported that mature cows subjected to first-point testing at auctions and transported for long distances produced carcasses with more severe bruises and a greater number of bruises than ranch-derived cows or cows not subjected to first of testing.

2.6. Weather conditions

Climate change could affect meat quality in one of two ways. There could be an effect through changing farming or abattoir practices to adapt the climate change, and there could be a direct effect of the changing weather conditions on the animals' (Gregory, 2010). The indirect effects of the changing weather conditions on the animals may include; changing the genotype of animals by introducing more heat tolerant breeds; holding animals outdoors of housing them during winter, feeding low protein-high finisher rations to combat heat induced growth suppression; pre-condition animals to hot conditions so they will be better adapted to survive heat stress during transport to a processing plant (Gregory, 2010). Grandin (1992) reported that occurrence of Dark Firm Dry (DFD) is highest during very cold weather combined with precipitation, which increases the rate of body-heated loss and elicits shivering. Summer conditions have been associated with paler meat colour compared with winter time (Gregory, 2010). However Scanga *et al.* (1998) also reported the incidence of DFD beef is high in very warm weather or when large fluctuations in temperature occur over short periods of time.

2.7. Effect of animal factors on meat quality

The quality of beef is affected by many factors but the most important are type, breed, sex, feeding, growth rate and animal handling associated with marketing and slaughter of animals. The effect of breed of cattle on the incidence of DFD condition in the longissimus muscle was also studied by (Shackelford *et al*., 1994). The sex significantly influences carcass and neat quality. Bull carcass is characterized by higher meat content with simultaneous lower content of fat compared to heifer's carcass. Heifers are more fattened with lower share of bones. However, meat obtained from bull is often characterized by improper quality parameter especially high pH and dark colour, which has negatively affected its technological properties and limits suitability to culinary meat production (Weglarz *et al.,* 2002; Mach *et al.*, 2008).

2.8. Beef quality

Beef quality is the product which results from the transformation each muscle undergoes the different operations of meat processing. Meat quality is a function of tenderness, pH, colour, juiciness, flavour and nutritive value (Webb and O'Nell, 2008). Quality attributes primary affected by transport and handling in cattle include pH, colour, tender, texture and moisture and a degration of these variables is collectively referred to as dark-firm and dry, high pH, low glycogen meat (Tarrant, 1989).

2.9. Colour of meat

Pre-slaughter handling factors such as loading, transportation, distance travelled, unloading and time spent in the lairages, the procedure and method of slaughtering used to affect these attributes (Roth *et al.,* 2007).Colour has been a very important factor in beef marketing, since it is the first quality attribute seen by the consumer, and is an indicator of freshness and wholesomeness (Muchenje *et al.,* 2009a; Troy and Kerry, 2010). Meat purchasing decisions are influenced by colour more than any other quality factor because consumers use discoloration as an indicator of freshness (Mancini and Hunt, 2005; Rosenvold and Anderson, 2003). Colour of meat depends upon several individuals' factors and their interactions, and concentration of meat pigments, essentially myoglobin and the chemical state of myoglobin (Rosenvold and Anderson, 2003).

Differences in meat colour have been associated with variations in intramuscular fat and moisture content, age dependent changes in muscle myoglobin content and the pHu of the muscle (Muchenje *et al.,* 2008). Myoglobin is the basic pigment in fresh meat and its content varies with production factors such as species, animal age, sex, feeding systems, type of muscle and muscular activity. Meat colour can also be defined in terms of Hunter calorimetric co-ordinates with L*, a* and b* values (Kannan et al., 2003). Lightness (L*) co-ordinates indicates lightness of the meat and it ranges from 0 (all light absorbed) to 100 (all light reflected). The a* co-ordinate is the redness of meat and it ranges from -60 (green) to + 60 (red) and the b* co-ordinate ranges from -60 (blue) to +60 (yellow) Simela. (2005).

2.10. pH and quality of meat

Meat quality is influenced to a large extent by the rate of pH decline in the muscles after slaughter and by the ultimate pH and colour affect consumption (Simela. 2005). These attributes are affected by conditions that occur prior and post-slaughter from the farm to the abattoir. The pH of living is around 5.4 to 6.0 but after death the sugars in the muscles are converted to the lactic acid, lowering the pH (Hambrecht *et al.,* 2004). The pH below 5.4 leads to tougher muscles (Sen *et al*., 2004; Muchenje *et al*., 2008). This is due to the formation of bonds between the myosin head and actin resulting in the occurrence of rigor mortis as already allued to stressful handling facilities at the abattoir and the rate at which meat is cooled mainly affect pHu (Toohey and Hopkins, 2006). Nutritional stress as can occur during cattle auctions can results in dehydration, electrolyte imbalances, negatively energy balance, glycogen depletion in muscle and catabolism of protein and fat, ultimately increasing the pH (Dhanda *et al.,* 2003; Mushi *et al.,* 2009).

2.11. Characterization and functions of creatine kinase (CK)

CK is a key enzyme for the energetic metabolism of tissue with high and fluctuating energy demands in vivo, as it is the case of skeletal muscle tissue, which is the most important for the meat industry (Kunz, 2001). CK is used to show the response of animals to stress caused by transportation, handling, loading and off-loading. This enzyme is generally utilized as an indicator of physical stress and /or muscle damage in animal production. CK is a member of phosphoryl transfer enzymes usually called guanidine or phosphagen kinase (Radostits, *et al*., 2000; Brawn *et al*., 2008). CK is therefore used to indicate cellular injury, heart and liver damage (Zhang *et al.,* 2010). Pre-slaughter, muscular activity and damage is severe due to transportation and handling. This increases the CK levels in the blood (Kannan *et al.,* 2003). In addition when animals are transported for more than 6 hours, CK activity increases (Kannan *et al*., 2007). In goats, higher levels of CK in the plasma may also be caused by breed temperament, excitability and fighting against each other (Grzyb and Skorkowski, 2005).

This enzyme can be measured from the blood of the animal before and after transportation (Bertin *et al.,* 2007). CK activity is easy to determine when animals (sheep, pigs and cattle) are handled vigorously loaded and unloaded (Kannan *et al.,* 2003; Adenkole and Ayo, 2010). Therefore, physiological stress induced to an animal can be determined by using CK as an indicator of stress in order to improve meat quality. It is then separated into fractions that contain cells and serum. In the serum, tests are done to determine the amount of CK present. Any present is measured and reported in units (U) of enzyme activity per litre (L) of serum. CK continues to exert important functions after slaughter, participating in the transformation of the muscles into meat.

CK is also used to evaluate the synthetic capacity of an organ, to diagnose the adverse effects of toxic compounds which inhibit enzymes and to monitor the inductive activity of exogeneous compounds or enzyme activation by minerals or vitamins. In all these cases, interpretation is usually based on physic pathological data regarding input of the enzyme into body fluid, most often blood plasma and sometimes urine, digestive contents and cerebrospinal fluid. When interpreting the decrease in plasma activity of an enzyme used as a marker if organ damage, the plasma half-life of enzymes is sometimes used to address their clearance (Radostits *et al.*, 2000).

2.12. Conclusion

Pre-slaughter handling of cattle involves several activities that occur prior to animal slaughter. Transportation and pre-slaughter handling are very important, not only from a welfare point of view, but because they have effects on bruising and beef quality thereby resulting in negative economic implications. When cattle are stressed, the glycolysis rate increases which can result in poor meat quality i.e. dark firm and dry (DFD). DFD meat might be associated with long period of stress. In order to evaluate stress-inducing factors, behavioural and physiological measurements are used together with indicators of post-mortem muscles metabolism. Muscle metabolism results in CK being released to the blood, which also affects meat quality. Therefore to improve beef quality, there is a need to determine the effects of pre-slaughter beef handling on bruising, CK and meat quality.

2.13. References

Adenkola, A. Y. And Ayo, J. O. (2010). Physiological and behavioural responses of livestock to road transportation stress: A review. *African Journal of Biotechnology, 9 (31*), 4845-4856.

Basbaum, A. and Woolf, C. (1999). Pain. *Current Biology, 9,* 429-431.

Bertin, M., Pomponi, S. M., Kokhula, C., Lwasaki, N., Suzuki, T. And Ellington, W. R. (2007). Origin of genes for the isoforms of Creatine Kinase. Gene, 392-1-2.

Blood, D. C. and Studdert, V. P. (1988). Baillieres : Comprehensive Veterinary Dictionary Bailliere Tindall, 24-28. Oval Road London, NWI and DX, UK.

Brawn, J. P., Medaille, C. and Trumel, C. (2008). Clinically interpretation of enzyme activities and concentrations: A review of the main metabolic factors affecting variation. *Israel Journal of Veterinary Medicine, 63 (1),* 1-12.

Chambers, P. G., Grandin, T., Heinz, G. and Srisuvan, T. (2004). Effects of stress and injury on meat and by-product quality. In : Guidelines for human handling, transport and slaughter of livestock. FAO Corporate document respository.

Dhanda, J. S., Taylor, D. G. and Murray, P. J. (2003). Part 1: Growth, carcass and meat quality parameters of male goats: effects of genotype and live weight at slaughter. *Small Ruminant Research, 50,* 57-66.

Edridge, G. A. and Winfield, C. G. (1988). The behaviour and bruising of cattle during transport at different space allowances. *Australian Journal of Experimental Agriculture, 28,* 695-698.

Eldridge, G. A., Barnett, J. L., McCausland, L. P., Millar, H. W. C. and Vowles, W. J. (1984). Bruising and method of marketing cattle. *Animal Production Australian, 15,* 675.

Fergusson, D. M. and Warner, R. D. (2008). Have we underestimated the impact of pre-slaughter stress on meat quality in ruminants? *Meat Science, 80*, 12-19

Gracey, J. F. and Collins, D. S. (1992). *Meat hygiene.* Bailliere Tindall, London.

Grandin, T. (1990). Design of loading facilities and holding pens. *Applied Animal Behaviour Science, volume, 28*, 187-201.

Grandin, T. (1992). Problems with bruises and dark cutters in harvest steers/ heifers, In: Improving the consistency and competitiveness of beef- A blueprint for total quality management in the feed beef industry. The final report of the national Beef Quality Audit- 1991. Colorado State University, Fort Collins.

Grandin, T. (2000). Livestock handling and transport (2^{nd} edition). Wallingford, Oxon, UK: CABI Publishing.

Gregory, N. G. (1996). Welfare and hygiene during pre-slaughter handling. *Meat Science, 43,* 35-46.

Gregory, N. G. (2010). How climatic changes could affect meat quality. *Food Research International, 43*, 1866-1873.

Grzyb, K. and Skorkowski, E. F. (2005). Characterization of creatine kinase isoforms in herring (Clupea harengus) skeletal muscle. *Comparactive Biochemistry and Physiology, Part B, 140, 629*- 834.

Hoffman, D. E., Spire, M. F., Schwenke, J. R. and Unruh, J. A. (1988). Effect of source of cattle and distance transported to commercial slaughter facility on carcass bruises in mature beef cows. *Journal of the American Veterinary Medical Association, 212,* 668-672.

Jago, J. G., Hargreaves, A. L., Harcourt, R. G. and Matthews, L. R. (1996). Risk factors associated with bruising in Red Deer at a commercial slaughter plant. *Meat Science, 40,* 181-191.

Jarvis, A. M., Selkirk, L. And Cockram, M. S. (1995). The influence of source of source, sex class and pre-slaughter handling on the bruising of cattle at two slaughterhouses. *Livestock Production Science, 43,* 215-224.

Kannan, G., Kouakou, B., Terril, T. H. and Gelaye, S. (2003). Endocrine, blood metabolite and meat quality changes in goats as influenced by short-term, pre-slaughter stress. *Journal of animal Science, 81,* 1499-1507.

Kannan, G., Terrill, T. H., Kouakou, B. and Galipalli, S. (2007). Blood metabolite changes and live weight loss following brood seaweed extract supplementation in goats subjected to stress. *Small Ruminant Research, 73,* 228-234.

Knowles, T. G. (1999). A review of the road transport of cattle. *Veterinary Record, 144,* 197-201.

Kunz, W. S. (2001). Control of oxidative phosphorylation in skeletal muscle. *Biochimica et Biophysic Acta, 1504,* 12-19.

Langlois, N. E. I. (2007). The science behind the quest to determine the age of bruises- a review of the English language literature. *Forensic Science, Medical and Pathology, 3,* 241-251.

Langlois, N. E. I. And Greshman, G. A. (1991). The aging of bruises- a review and study of the colour changes with time. *Forensic Science International, 50,* 227-238.

Mach, N., Bach, A., Velarde, A. and Devant, M. (2008). Association between animal, rasportation, slaughterhouse practices and meat pH in beef. *Meat Science, 78,* 232-238.

Mancini, R. A . and Hunt, M. C. (2005). Current research in meat colour. A review . *Meat Science, 71,* 100-121.

McNally, P. W. and Warris, P. D. (1996). Recent bruising in cattle at the abattoirs. *Veterinary Record, 138,* 126-128.

Muchenje, V., Dzama, K., Chimonyo, M., Strydom, P. E and Raats, J. G. (2009b). Relationship between pre-slaughter stress responsiveness and beef quality in three cattle breeds. *Meat Science, 81*, 653-657.

Muchenje, V., Dzama, K., Chimonyo, M., Strydom, P. E. and Raats, J. G. (2008). Meat quality of Nguni, Bonsmara and Aberdeen Angus steers raised on natural pasture in the Eastern Cape, South Africa, *Meat Science, 79,* 20-28.

Muchenje, V., Dzama, K., Chimonyo, M., Strydom, P. E., Hugo, A. and Raats, J. G. (2009a). Some biochemical aspects pertaining to beef eating quality and consumer health. A review- *Food Chemistry, 112,* 279-289.

Mushi, D. E., Safari, J., Mtenga, L. A., Kifaro, G. C. and Eik, L. O. (2009). Effects of concentrate levels on fattening performance, carcass and meat quality attribute of Small

East African x Norwegian crossbred goats fed low quality grass hay. *Livestock Science, 124,* 148-155.

Radostits, O. M., Arundel, J. H., Gay, C. C., Hinchcl, F. F. and Blood, D. C. (2000). 9th Edition Veterinary Medicine: A textbook of the diseases of cattle, sheep, pigs and horses. *Elsevier Health Science,* pp, 1426-1430.

Ronsevold, K. and Anderson, H. J. (2003). Factors of significant for pork quality-a review *Meat Science, 64*, 219-337.

Roth, B., Imsland, A., Gunnarsson, S., Foss, A. and Schelvis-Smith, R. (2007). Slaughter quality and rigor contraction in farmed turbot (Scophthalmus maximum) between different stunning methods. *Aquaculture,* 272, 754-761.

Scanga, J. A., Belk, K. E., Tatum, J. D., Grandin, T. And Smith, G. C. (1998). Factors contributing to the incidence of dark cutting beef, *Journal of Animal Science, 76,* 2040-2047.

Sen, A. R., Santra, A. and Karim, S. A. (2004). Carcass yield, composition and meat quality attributes of sheep and goats under semiarid conditions. *Meat Science, 66,* 757-763.

Shacklelford, S. D., Koohmariane, M., Wheeler, T. L., Cundiff, L. V. and Dikeman, M. E. (1994). Effect of biological type of cattle on the incidence of the dark, firm and dry condition in the longissimus muscle. *Journal of Animal Science*, *72,* 337-343.

Simela, L. (2005). Meat characteristics and acceptability of chevon from South African indigeneous goats. University of Pretoria, South Africa.

Strappini, A. C., Metz, J. H. M., Gallo, G. and Kemp, B. (2009). Origin and assessment of beef cattle at slaughter. *Animal, 3,* 728-736.

Sutherland, M. A., Krebs, N., Smith, J. S., Dailey, J. W., Carroll, J. A. and McGlone, J. J. (2009). The effect of three space allowances on the physiology and behaviour of weaned pigs during transportation. *Livestock Science, 126*, 183-188.

Tarrant, P. V. (1989). Animal behaviour and environment in the dark –cutting condition . In S. U. Fabiansson, W. R. Shorthose, and R. D. Warner (Eds.), Dark cutting in cattle and sheep (pp 8-18). Sydney, *Australia: Australian Meat and Livestock Research Development Corporation.*

Tompson, J. (2002). Managing meat tenderness. *Meat Science, 62,* 295-308.

Toohey, E. S. and Hopkins, D. L. (2006). Eating quality of commercially processed hot boned sheep meat. *Meat Science, 72*, 660-665.

Troy, D. J. and Kerry, J. P. (2010). Consumer Perception and the Role of Science in the Meat Industry. *Meat Science, 86,* 214-226.

Warriss, P. D (2000). Meat Science: an introductory text, Wallingford, Oxon, UK: CABI Publishing.

Webb, E. C. and O'Neil, H. A (2008). The animal fat paradox and meat quality. *Meat Science, 80,* 28-36.

Weglarz, A., Zapletal, P., Gil, Z., Skrzynski, G. and Adamczyk, K. (2002). The effect of sex and age on beef quality. Zesz, Nauk, Prz. Hod., 62, 211-216 (in Polish, with English summary).

Zhang, M., Xin, L., Bao, E., Hartung, J. and Yue, Z. (2010). Variation in the expression of Hsp 27 αB-crystllin mRNA and protein in the heart and liver of pigs exposed to different transport times. *Research in Veterinary Science, 90 (3),* 423-438.

Chapter 3: Effect of distanced travelled and animal factors on bruising and creatine kinase levels (CK)

Abstract

By

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The objective of the current study was to determine the effect animal factors and distance travelled on bruising and creatine kinase (CK) levels. Three hundred and twenty one cattle from three breeds (108 Bonsmara, 130 Beefmaster and 83 Brahman) were used in the study. Blood samples for CK determination were collected at exsanguinations using disposable vacutainer tubes. Animal factors were significantly associated with bruise scores. There was a significant effect ($P<0.001$) of the season on CK levels. Significant breed, age and gender differences were also observed on CK levels. There was no relationship observed between pre-slaughter conditions such as distance, on bruising score and CK levels. Therefore, it was observed that pre-slaughter conditions did not have effect any on the bruising score and CK.

Key words: Animal welfare, breed, bruising score, creatine kinase, distance travelled, season.

3.1. Introduction

Bruising in slaughtered cattle is affected by intrinsic and extrinsic factors. The age of animal at slaughter, breed and sex of the animal are some of the intrinsic factors while extrinsic factors include pre-slaughter conditions (Muchenje *et al*., 2009a; Muchenje *et al*., 2009b). Pre-slaughter conditions are a chain of activities that occur before the animal is slaughtered such as loading and off-loading, transportation and lairage period. Transportation is an important activity of the farming industry because it involves several potential stressors such as poor handling and road conditions, unloading, mixing of unfamiliar animals, humidity, overcrowding and length of the journey (Buil *et al*., 2004; Verga *et al*., 2009). Poor transport conditions prior to slaughter can lead to stress, bruising and death in some cases of serious injury. Conditions associated with rough handling or with aggression between animals which lead to damage to the animal tissues which may develop into a bruise (Gracey *et al.*, 1999).

Bruises or contusions are described as superficial discoloration due to haemorrhage into tissue from raptured blood vessels beneath the skin surface, without the skin being broken (Blood and Studdert, 1998; Vimiso, 2010). In the contusions, the blood accumulates in surrounding tissues, producing pain and swelling (Blood and Studdert, 1998). These injuries which include bruises, blemishes and broken bones are associated with short or long transportation due to poor road conditions (Maria *et al.*, 2006; Minka and Ayo, 2007; Liste *et al*., 2008). Bruising occurs at the farm, during transportation and at the slaughterhouse (Jarvis *et al*., 1995).

Several authors; Yeh *et al*. (1978), McNally and Warris (1996) and Hoffman *et al*. (1998) emphasised the relationship between distance travelled and occurrence of bruising in bovines suggesting that level of bruising might increase with the distance travelled by animals and consequently the amount (kg) of bruise tissue trimmed per carcass (Wythes and Shorthose 1991). Level of bruising varies with sex and age of the animal (Yeh *et al*., 1978; Gallo *et al*., 1999), such that older animals are more prone to bruising (Strappini *et al*., 2008). Cattle are more likely to pass through a livestock market before arriving at the slaughterhouse. Pre-slaughter conditions such as transportation have been found to have deleterious effects on the physiology (lactate, glucose, CK and cortisol) of the animal. Long transportation increases the activity of muscle enzymes such as creatine kinase (CK), increases proportionately with the duration of the journey, but they also remain high for several days (Warriss *et al.*, 1995; Fazio and Ferlazo, 2003). CK is an enzyme found in the skeletal muscles. It is released into the blood plasma in response to stress and muscle damage (Knowles *et al*., 1999; Vojtic, 2000). The total activity of CK is used to measure the sum of enzyme activities from different organs. CK can be either being as M or a B monomer sub-unit. The muscle M-CK can also be used as S-CK found in the M-line of the zone in the sarcomere length (Hornikova *et al*., 2009). Therefore, when CK is found in the blood, it could indicate severe muscle damage. However there is limited information regarding the relationship between animal factors and distance travelled on the occurrence of bruises and the activity of plasma creatine kinase. The objective of the current study was therefore to determine the effect of animal factors and distance travelled on bruising and the creatine kinase levels.

3.2 Materials and Methods

3.2.1 Description of the study site

The study was conducted in East London Abattoir which is located in Buffalo City Municipality. The abattoir is situated at longitudes and latitudes 32.´2º S and 27.´5º E in the Amathole District of the Eastern Cape Province of RSA. The area has vegetation that ranges from grassland and thicket to forests and bush veld with *Themeda trianda and Digitaria eriantha* being the most dominant plant species. The place receives approximately 850 mm of rainfall per year of which most falls during the summer months. The day temperature ranges for the period of study were10 ºC to a high of 36º C with a mean temperature of about 22.3ºC. The topography of the area is generally flat with few steep slopes.

3.2.2 Animals management

Data was collected from 321 cattle over four seasons (winter, summer, autumn and spring) between the year 2011 and 2012. All cattle were slaughtered at the East London commercial abattoir in the Eastern Cape Province. The plant operated from 0600 to 1700 h Mondays to Fridays. On arrival at the abattoir the three breeds of cattle; Bonsmara, Brahman and Beefmaster were identified. The numbers of animals per breed were 108 Bonsmara, 130 Beefmaster and 83 Brahman. The animals used were male and females, and were grouped according to age categories as follows: Group 1(16 months), Group 2 (18 months) and Group 3 (24 months). The cattle were transported by the owners from the farms to the abattoir. Transportation time (departure, arrival time and distance travelled), lairage duration, number of animals per pen, slaughtered animals per day and daily temperature were all recorded.

On arrival at the abattoir, offloading activities were monitored and cattle were given water *ad-libitum* prior to slaughter. Cattle were then slaughtered using commercial standard procedures of the abattoir. The captive bolt method was used to stun the animals, using a voltage of 300 V, a frequency of Hz, a current of 5A in 40-45s at a pulse of 12/s.

3.2.3. Blood collection and plasma separation

The blood was collected from beef animals on the dressing floor at the point of bleeding soon after stunning using disposable vacutainer tubes. The blood samples were placed in ice until blood plasma was completely separated within two hours after collection. A centrifuge machine (model: 5403 Centrifuge Geratebay Eppendorf GmbH, Engelsdorp Germany) was used to centrifuge blood samples at 3550 rpm, 21^0C for 10. Plasma was then separated and inserted into1.5 ml Eppendorf tubes using pipettes and stored at -20^0C. The samples were then arranged for easy identification during transportation for determination of CK.

3.2. 4. Creatine Kinase level determination

Creatine kinase from the stored blood samples were analysed using a Model DXC 600 machine (Beckman Coulter Ireland) at the National Health Laboratory Services, Port Elizabeth. The contents included 2 x 61 ML CK Reagent, 1 preparation insert and the reactive ingredients included Creatine Phosphate, Disodium Salt 461 mmol/ L, Nadide 30:0mmol/L, Adenosine-5-Diphosphate, Monopotassium Salt, Dihydrate 36.0 mmol /L, Glucose 24.0 mmol /L, Glucose-6-Phosphate Dehydrogenase 46.1kU/L and Hexokinase 136kU/L. All the ingredients were added for quantitative determination of CK activity of units per litre (U/L) in plasma.

3.2.5. Bruise measurements

Only carcasses that had bruises were considered. The assessment was done using a method based on the Australian Carcass Bruise Score System (Anderson, 1979). Bruise scores were calculated for each load by multiplying the number of bruises in each size class by a weighting factor: slight 1, medium 3, and heavy 5, and adding these values (Anderson and Horder, 1979). These bruise scores were then divided by the number of cattle per load to give a mean bruise score per animal for each load. Bruise age was estimated using the method of Gracey *et al.* (1999) as described in Table 3.1.

Table 3. 1 Colour observations used to estimate the age of bruises

Observable colour of the bruise	Estimated age of the bruise in hours
Red and hemorrhagic (bright-red)	0-10 hours old
Dark- red colour	Approximately 24 hours old
Watery consistency	24-38 hours
Rusty orange colour, soapy to touch, clear yellow mucus	+72 hours (3 days old)

Gray *et al*. (1999).

3.2.6. Statistical Analysis

The PROC GLM of Statistical Analysis System (SAS) Institute (2003) was used to analyse the effect of breed, age, gender and season on the activity of creatine kinase and bruising score.

The model used was as follows

$У_{ijkl} = \mu + \alpha_i + \beta_j + \lambda_k + G_l + \varepsilon_{ijkl}$

Where ,

Y_{ijkl} = response variable (creatine kinase, bruising score and bruise age)

μ = constant mean common to all observations

α_i= effect of breed

β_j = effect of age group (Group 1, 2 and 3)

λ_k = effect of season

G_l= effect of gender

E_{ijkl} = random error

Significant differences between least square group means for CK and animal factors were compared using Duncan's procedure. The data was analysed using PROC GLM of Statistical Analysis System (SAS) Institute (2003) to determine the relationship between distance duration, CK and bruising score. Significant differences between least square group means for creatine kinase concentrate and animal related factors.

3.3. Results and Discussion

Bruising score and CK levels were not related to distance travelled **(**P>0.001). The activity of CK for Bonsmara breed was higher **(**705.3±80.57) than the Brahman breed (461.8±80.63) as shown in Table 3.2. The increase of CK activity in bonsmara breed is more susceptible to stress before slaughter and also could be due to having been roughly driven and mishandled before and after transportation (Broom *et al*., 1996; Averos *et al.,* 2008). The activity of CK increased in the blood after tissue damage, poor muscular tissue reperfusion, decreased heat and fatigue, apparently, were a result of increased in the permeability of muscle membrane induced by capture, loading and transportation (Warriss *et al*., 1995; Knowles *et al*., 1999; Lopez *et al*., 2006; Guardia *et al*., 2009). Transportation for several hours is a physically demanding factor; animals have to maintain balance and the contact between animals produces fatigue and bruising, affecting the permeability of the membranes and the liberation of the enzymes in the blood (Brown *et al.,* 1999; Lopez *et al*., 2006). Transportation and related handling procedures caused an increase in CK levels of animals. It was postulated that CK activity indicate the extent of muscular activity and damage during hauling and handling Kannan *et al*. (2000) and Miranda-de la Lama *et al.* (2010). Furthermore, other animals are susceptible to transport stress leading to highest levels of CK in the blood (Yu *et al*., 2009).

Table 3. 2 Least square means (±SE) for the effect of breed on creatine kinase levels of meat slaughtered at East London abattoir

Breed	Creatine kinase (U/1)
Bonsmara (n = 108)	705.3±80.57[a]
Beefmaster (n=130)	657.4±73.85[ab]
Brahman (n =83)	461.8±80.63[b]

[a,b] Means in the same column with different superscripts are significantly different at $^{*}P<0.05$

Male animals had the highest CK values (P<001) while the female animals had the lowest CK values Table 3.3. Male animals tended to be more excitable, aggressive and stress sensitive than females. The sex of animals has an effect on animal stress, with males being more stressed than females (Mota-Rojas *et al*., 2006). The higher the increase in the plasma activity of CK found in steers transported for 12-16 hours agrees with Warris *et al*. (1995) who reported that a direct relationship between the duration of transport and the rise in the activity of the enzyme.

Creatine kinase levels were higher in 18 months old animals than 24 months old animals Table 3.3. Young animals are very poorly adapted to cope with transportation resulting in high mortality rates and injuries. Steinhardt and Thielscher (1999) reported that young animals respond to transportation with increases in body temperature, heart rate and plasma cortisol concentration. These findings are supported by Parrot *et al.* (1994), whose findings were that young animals are at risk of more bruising than older animals resulting in high levels of CK. High levels of the enzyme CK in the blood plasma indicate physical exhaustion due to long journeys. Hence such animals should not be transported for long hours.

Table 3. 3 Least square means (±SE) for the effect of sex and age of animal on creatine kinase levels of meat slaughtered at East London abattoir

Sex	**Creatine kinase (U/1)**	**Age**	**Creatine kinase (U/1)**
Female (n=222)	579.3±13.65[b]	16 months(116)	636.6±76.83[b]
Male (91)	724.6±21.31[a]	18 months (85)	729.6±89.75[a]
		24 months (112)	524.1±78.19[c]

[a,b,c] Means in the same column with different superscripts are significantly different at [*]P<0.05

The results for the effect of season on the bruising score and CK levels are shown in Table 3.4. Creatine kinase was higher during the winter season (704.1±43.85) and lower CK values in the summer season (181.3±124.09). This is in agreement with the results by Knowles *et al*. (1998) who observed that in their study CK levels was higher in winter season due to poor pre-slaughter conditions. Zahner *et al*. (2004) found that an influence of cold climatic conditions on CK secretion in dairy cows. However other authors reported an increased CK values in lambs as a result of physical stress during summer (Minton and Blecha, 1990; Lowe *et al.,* 2002, Miranda-de la Lama *et al.,* 2011). Wilson *et al*. (1990); Knowles *et al*. (1998) and Liu *et al*. (2008) reported that vigorous physical activity at high temperatures had an effect on CK. Transportation of cattle during summer and winter months is associated with external air temperature and air humidity and result in increased CK levels. Muscle enzyme activity increases during transport because of increased muscle cell permeability or muscle cell damage, making CK a very sensitive indicator of muscular damage (Miranda-de la Lama *et al*., 2010).

Autumn had the highest bruising score (1.86±1.07) and the summer season had the least bruising score (1.12±0.11) Table 3.4. Seasonal factors mainly temperature and humidity also seem to play a role in the prevalence of bruises. Results from this study concur with Edridge and Winfield (1988), Gosalvez *et al*. (2006) and dalla Costa *et al*. (2007), who found that cold weather combined with precipitation, increased the mean number of bruises per animal, although the reasons are still unclear. More bruising in winter may be associated with falls, slips or sprains produced during pre-slaughter handling.

Table 3. 4 Least square means (±SE) for effects of season on bruising score and CK levels of meat slaughtered at East London abattoir

Season	Bruising score	Creatine kinase (U/1)
Spring	1. 32±0.54[b]	698.3±59.19[a]
Summer	1.12±0.11[c]	181.3±124.09[b]
Winter	1.44±0.13[b]	704.1.3±43.85[a]
Autumn	1.86±1.07[a]	658.0±116.25[a]

[a,b,c]Means in the same column with different superscripts are significantly different at [*]P<0.05, [***]P <0.001

Spring = (August, September and October)

Summer = (November, December and January)

Winter = (May, June and July)

Autumn = (February, March and April)

Figure 3.1 shows the effect of breed on the bruising scores of slaughtered beef animals. The Bonsmara had the highest percentage none bruised meat 88% and the Brahman had the lowest none bruised meat 49%. Bonsmara also had highest percentage 20% of slightly = bright red bruised meat, 0-10 hours old) while Brahman had lowest percentage of bright red bruised meat 4%. The stress response during handling also depends on the genetic differences within the breed. Susceptibility to bruising and temperament differs between individual animals. Fordyce *et al*. (1985) concluded that this was more important than breed differences. It appears that breed differences can be attributed to differences in behaviour and to being horned or hornless (Minka and Ayo, 2007).

Bonsmara had the highest percentage 10% medium = bruised dark red, 24 hours old with Brahman having the lowest bruised dark red meat 5%. This is due to transportation conditions. This could be due to several injuries associated with road transportation. The level of bruising might increase with the length of journey travelled by the animals (Weeks *et al.*, 2002). All the breeds Brahman and Beefmaster breeds had the lowest percentage on severe deep bleeding bruising or injuries (24-38 hours old). Occurrence of bruises can be attributed to breed differences.

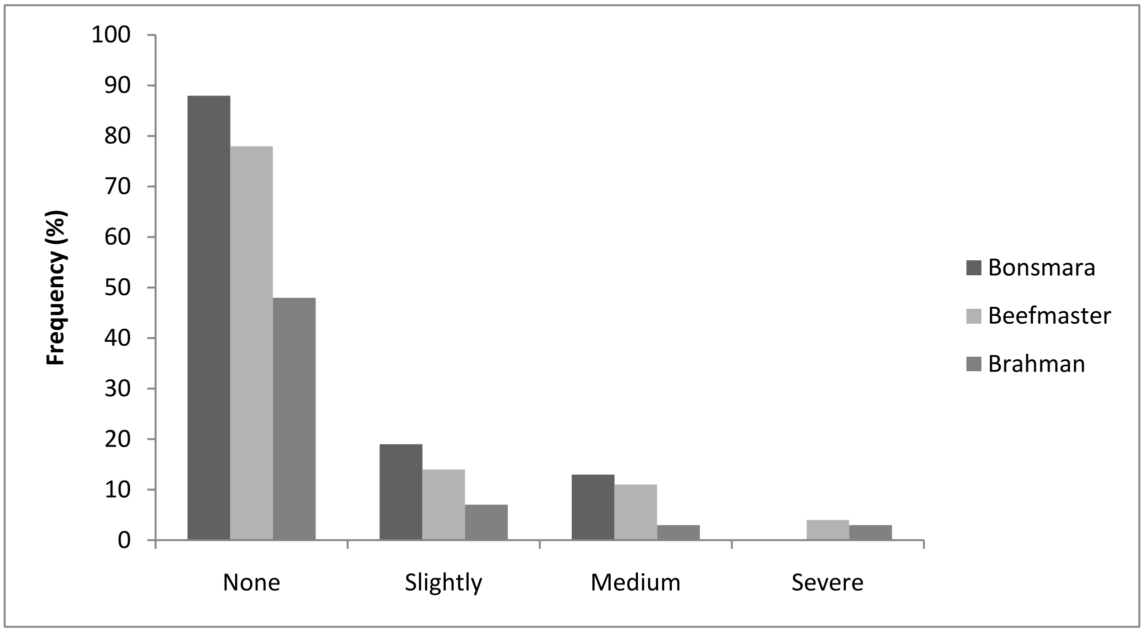

Figure 3. 1 Effect of breed on bruising scores of slaughtered beef animals.

None = 0 hours

Slightly = 0-10 hours old, Red and hemorrhagic (bright-red)

Medium = 24 hours old, Dark- red colour

Severe = 24-38 hours old, Watery consistency

The highest number of slightly bright red bruised meat was observed in 16 months and the 24 months old cattle had the lowest slightly bright bruised meat (Figure 3. 2). The highest medium of dark red bruises was observed in 16 months and the lowest medium dark red bruised as observed in the 24 months old cattle. The level of bruising in animals varies with age of the animal with young animals being more prone to bruising than old animals. The fact that young animals had more bruising may not only be due to age, but also due to increased poor handling (Strappini *et al*., 2009). These results disagree with Wythes and Shorthose (1991) who reported that heavy animals had more bruising than young animals – the mature and old cows and oldest steers of the group.

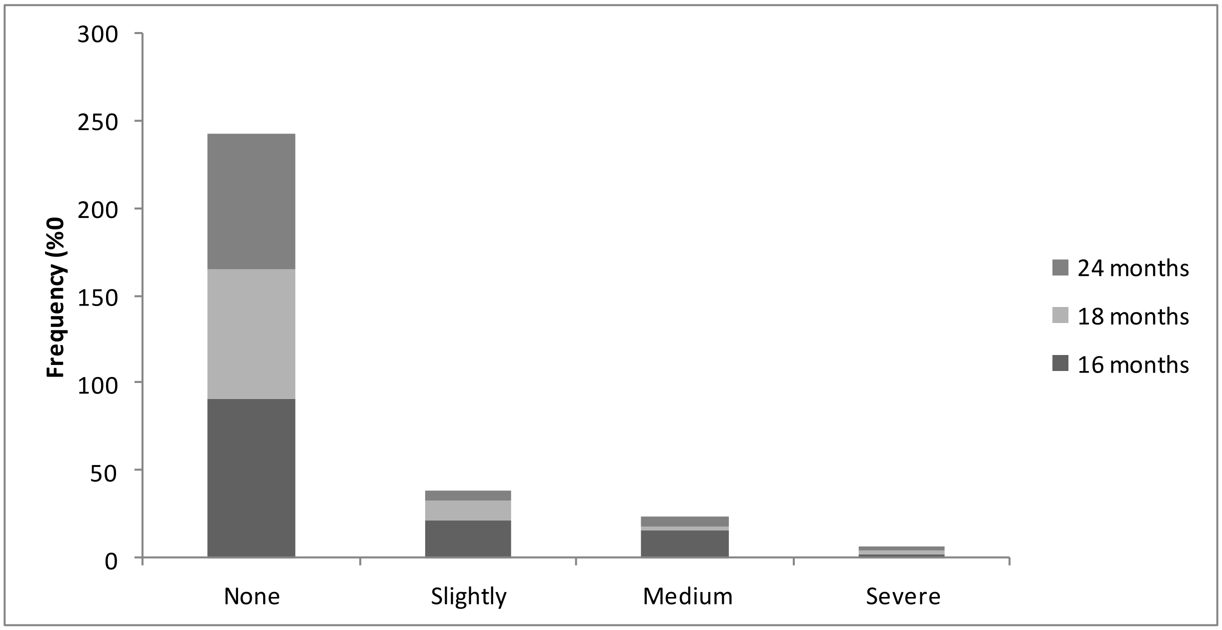

Figure 3. 2 Effect of age on bruising scores of slaughtered beef animals.

None = 0 hours

Slightly =0-10 hours old (Red and hemorrhagic bright-red)

Medium = 24 hours old (Dark- red colour)

Severe = 24-38 hours old (Watery consistency)

Highest levels of slightly bright bruised meat and medium dark bruised meat were observed in female animals, while male animals had the lowest slightly bright bruised meat and medium dark bruised meat (Figure 3.3). In literature there is evidence that female animals have higher chances for bruises than male animals (Yeh *et al*., 1978; Jarvis *et al*., 1995). Furthermore, only in cows the amount of bruising increased with increased duration of journey. Another reason is that female animals would bruise more easily due to their lack of fat cover (Knowles *et al.*, 1994; Grandin, 1998). Sex and age might explain the higher bruises susceptibility in female and older animals (Eguinoa *et al*., 2003). Another reason is that cows are treated with less care due to their lower commercial value compared with other types of animals (Grandin, 2007). Poor body condition of animals can be associated with higher proportion of bruises in cows compared to young heifers and steers (Gallo *et al.*, 1995; Grandin, 2002). These results agreed with Yeh *et al*. (1978) who reported that heifers had significantly more bruises than steers and also when animals kept as separate groups, cows bruise significantly more than steers and bulls. When heifers were completely separated from steers during transportation and handling, the mean number of bruises per animal differed significantly between sex classes. Furthermore, only in cows did the amount of bruising increase with increased duration of the journey. The levels of bruising from both male and female animals had a lower percentage on severe deep bruises.

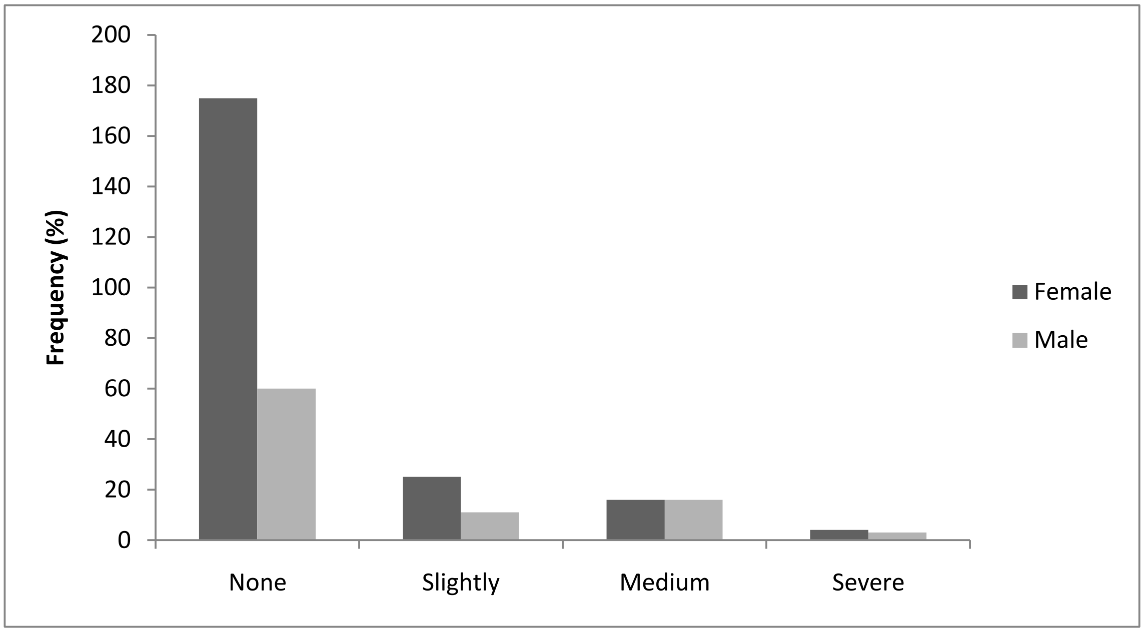

Figure 3. 3 Effect of sex on bruising scores of slaughtered beef animals.

None = 0 hours

Slightly = 0-10 hours old (Red and hemorrhagic bright-red)

Medium = 24 hours old (Dark- red colour)

Severe = 24-38 hours old (Watery consistency)

3.4 Conclusion

It is concluded that animal factors such as sex, breed and animal age at slaughter may contribute to the development of bruises and have an effect on CK levels, Bonsmara breed having higher CK levels and highest percentage of slightly bruised meat. Seasons have an effect on the bruising score and CK levels with winter months having higher CK levels. On the other hand pre-slaughter conditions such as distance travelled did not affect bruising score and CK levels.

3. 5. References

Anderson, B. (1978). The Australian Carcass Bruise Scoring System. *Proceedings of the Australian Society of Animal Production, 12,* 242-243.

Anderson, B. and Horder, J. C. (1978). The Australian Carcass Bruising Scoring System. *Queensland Agricultural Journal, 105,* 281-287.

Averos, A., Martin, S., Riu, M., Serratosa, J. and Gosalvez, L. F. (2008). Stress response of extensively reared young bulls being transported to growing-finishing farm under Spanish summer commercial conditions. *Life Science, 119,* 334-340.

Blood, D. C. and Studdert, V. P. (1988). Bailliere;s Comprehensive Veterinary Dictionary. BailliereTindall, 24-28 Oval Road London, NWI and DX, UK.

Broom, D. M., Goode, J. A., Hall, S. J. G., Lloyd, D. M. and Parrot, R. F. (1996). Hormonal and physiological effects of a 15- hour road journey in sheep: comparison with the responses to loading, handling and penning in the absence of transport. *Veterinary Research Communication, 27,* 515-518.

Brown, S. N., Knowles, T. G., Edwards, J. E. and Warriss, P. D. (1999). Relationship between food deprivation before transport and aggression in pigs held in lairage before slaughter. *Veterinary Record, 145, 630-634.*

Buil, T., Maria, G. A., Villaroel, M., Liste, G. and Lopez, M. (2004). Critical points in the transport of commercial rabbits to slaughter in Spain that may compromise animals'welfare. *World Rabbits Science*, *12*, 269-279.

Dalla Costa, , O. A., Fauctitano, L., Coldebella, A., Ludke, J. V., Pelso, J. V., Dalla, Roza, D. and Parranhos da Costa, M. J. R. (2007). Effect of the season of the year, truck type and location on truck on skin bruises and meat quality in pigs. *Livestock Science, 107,* 29-36.

Eguinoa, P., De la Fuenta, J., Sanchez, M., Perez, C., Lauzurica, S., Viera, C., Gonzalez de Chavarri, Brocklehurst, S., Arana, A., Mendizabal, J. A., Vernon, R. G. and Purroy, A. (2003). Lipogenic enzyme activities in different adipose depots of Pirenaican and Holstein bulls and heifers taking into account adipocyte size. *Journal of Animal Science, 81,* 432-440.

Eldridge, G. A. and Winfield, C. G. (1988). The behaviour and bruising of cattle during transport at different space allowances. *Australian Journal of Experimental Agriculture, 28,* 695-698.

Fazio, E. and Ferlazzo, A. (2003). Evaluation of stress during transport. *Veterinary Research Communications, 27,* 519-524.

Fordyce, G., Goddard, M. E., Tyler, R., William, C. and Toleman, M. A. (1985). Temperament and bruising of Bos Indicus cross cattle. *Australian Journal of Experiment Agriculture, 25,* 283-288.

Gallo, C., Carmine, X., Correa, J. and Ernst, S. (1995). Analisis del tiempo de transporte yespera, rentimiento de la canal de bovines transportadosdesde Osorno a Santigo. In.ProceedingsXXth Reenion Anual d la Sociedad Chilena de Production Animal.

Gallo, C., Caro, M. and Villarroel, C. (1999). Characteristics of cattle slaughtered within the Xth Region (Chile) according to the terms stated by the official Chilean standards for classification and carcass grading. *Archivos de Medical Veterinaria, 31, 81-88.*

Gosalvez, L. F., Averos, X., Valdevira, J. J. and Herranz, A. (2006). Influence of season distance and mixed loads on physical and carcass integrity of pigs transported to slaughter. Meat Science, *73,* 553-558.

Gracey, J. F., Collins, D. S. and Huey, R. J. (1999). *Meat Hygiene,* 10th edition . Elsevier Health Science Publishers, United Kingdom, pp.

Grandin, T. (1998). Handling methods and facilities to reduce stress on cattle. *Veterinary Clinics of North America-Food Animal Practice, 14,* 325-341.

Grandin, T. (2002). Behavioural Principles of Livestock Handling. *American Registry of Professional Animal Scientists, pp. 1-11.*

Grandin, T. (2007). Livestock Handling and Transport, 3rd Edition. CAB International. Wallingford Oxon, United Kingdom.

Guardia, M. D., Estany, J., Balasch, S., Oliver, M. A., Gispert, M. and Diestre, A. (2009). Risk assessement of skin damage due to pre-slaughter conditions and RYRL gene in pigs. *Meat Science, 81,* 745-751.

Hoffman, D. E., Spire, M. F., Schwenke, J. R. and Unruh, J. A. (1998). Effect of source of cattle and distance transported to a commercial slaughter facility on carcass bruises in mature beef cows. *Journal of the American Veterinary Medical Association, 212,* 668-672.

Hornikova, D., Herman, P., Mejsnar, J., Vecer, J. and Zurmanova, J. (2009). Creatine kinase structural changes induced by substrates. *Biochimica et Biophysica Acta, 1794,* 270-274.

in pregnant sheep. *Small Ruminant Research, 35,* 249-253.

Jarvis, A. M., Selkirk, L. and Cockram, M. S. (1995). The influence of source, sex class and pre-slaughter handling on the bruising of cattle at two slaughterhouses. *Livestock Production Science, 43*, 215-224.

Kannan, G., Terrill, Y. H., Kouakow, B., Gazal, O. S., Gelaye, S., Amoah, E. A. and Samake, S. (2000). Transportation of goats: effects on physiological stress responses and live weight loss. *Journal of Animal Science, 78*, 1450-1457.

Knowles, T. G. (1999). A review of the road transport of cattle. *Veterinary Record, 144,* 197-201.

Knowles, T. G., Maunder, D. H. and Warriss, P. D. (1994). Factors affecting the incidences of bruising in lambs arriving at one slaughter. *Veterinary Record, 134*, 44-45.

Knowles, T. G., Warriss, P. D., Brown, S. N. And Edward, J. E. (1998). Effects of stocking density on lambs being transported by road. *Veterinary Record, 142,* 505-509.

Liste, G., Maria, G. A., Garcia-Belenquer, S., Chacon, G., Gazola, P. and Villarroel, M. (2008). The effect of transport time, season and position on the truck on steers response in rabbits. *World Rabbit Science, 16,* 229-235.

Liu, Z. L., Chen, P., Li, J. M., Lin, S. B., Wang, D. M., Zhu, L. P. and Yang, D. P. (2008). Conjugated linoleic acids (CLA) moderate negative responses of heat stressed cows. *Livestock Science, 118*, 255-261.

Lopez, O., Marco, I., Montane, J. and Lavin, S. (2006). Transport stress in Southern chamois (rupricapra pyrenaica) and its modulation by acepromazine. *Veterinary Journal, 1720*, 347-355.

Lowe, T. E., Gregory, N. G., Fisher, A. D. and Payne, S. P. (2002). The effects of temperate elavaluation and water deprivation on lamb physiology welfare and meat quality. *Australia Journal of Agriculture Research, 53*, 704-714.

Maria, G. A., Buil, T., Liste, G., Villarroel, M., Sanudo, C. and Olleta, J. L. (2006). Effects of transport time and season on aspects of rabbit meat quality. *Meat Science, 72*, 773-777.

McNally, P. W. and Warriss, P. D. (1996). Recent bruising in cattle at abattoirs. *Veterinary Record, 138*, 126-128.

Minka, N. S. And Ayo, J. O. (2007). Effects of loading behaviour and road transport stress on traumatic injuries in cattle transported by road during hot dry season. *Livestock Science, 107*, 91-95.

Miranda –de la Lama, G. C., Sepulveda, W. S., Villarroel, M. and Maria, G. A. (2011). Livestock vehicle accidents in Spain: causes, consequences, and affects on animal welfare. *Journal of Applied Animal Welfare Science, 14*, 109-123.

Miranda-de la Lama, G. C., Villarroel, M., Liste, G., Escos, J. and Maria, G. A. (2010). Critical points in the chain of lamb in Spain that may compromise the animals welfare. *Small Ruminant Research, 90 (1-3)*, 174-178.

Morta-Rojas, D., Becerril, M., Lemus, C., Sanchez, P., Gonzalez, M., Olmos, S. A., Ramirez and Alonso-Spilsbury, M. (2006). Effects of mid-summer duration on pre-slaughter and post slaughter and pork quality in Mexico. *Meat Science, 73*, 404-412.

Muchenje, V., Dzama, K., Chimonyo, M., Strydom, P. E. and Raats, J. G. (2009a). Relationship between stress responsiveness and meat quality in three cattle breeds. *Meat Science, 81*, 653-657.

Muchenje, V., Dzama, K., Chimonyo, M., Strydom, P. E., Hugo, A. and Raats, J. G. (2009b). Some biochemical aspects pertaining to beef eating quality and consumer health: A review. *Food Chemistry, 112,* 279-289.

Parrot, R. F., Misson, B. H. and De la Riva, C. F. (1994). Differential stressors effects on the concentration of cortisol, prolactin and catecholamines in the blood of sheep. *Research Veterinary Science, 56*, 236-239.

SAS. (2003). In: *SAS/ STATT Guide to Personal Computers, Version 6*, Statistical Analysis System Institute Inc. Cary, NC.

Steinhardt, M. and Thielscher, H. H. (1999). Maturity of suckler calves and dairy calves at the second and third week of postnatal age and forms of reaction of the animals to transport by road. *Landbauforschung Volkenrode, 49*, 70-89.

Strappin, A. C., Metz, J. H. M., Gallo, G. and Kemp, B. (2009). Origin and assessment of beef cattle at slaughter. *Animal, 3*, 728-736.

Strappini, A. C., Frankena, K., Metz, J. H. M., Gallo, C. B. and Kemp, B. (2008). Presence of bruising in cattle beef carcasses in Chile. In Proceedings of 4th International Workshop on the Assessment of Animal Welfare at the Farm and Group Level (WALF), Ghent, Belgium.

Vegara, H. and Gallego, L. (2002). Effect of electrical stunning on meat quality of lamb. *Meat Science, 56 (4),* 345-349.

Verga, M., Luzi, F., Petracci, M. and Cavani, C. (2009). Welfare aspects in rabbit rearing and transport and handling. *Italian Journal of Animal Science, 8,* 191-204.

Vimiso, P. (2010). Effects of marketing channel on bruising, ultimate pH and colour of beef; and stakeholder perception on the quality of beef cattle slaughtered at a smallholder abattoir. MSc Thesis, University of Fort Hare, South Africa.

Vojtic, I. (2000). Macro CK type 1 as a major component of serum creatine kinase activity

Warriss, P. D., Brown, S. N., Knowles, T. G., Kestin, S. C., Edwards, J. E., Dolan, S. K. and Philips, A. J. (1995). The effects on cattle of transport by road for up to fifteen hours. *Veterinary Record, 136,* 319-323.

Weeks, C. A., McNally, P.W. and Warriss, P. D. (2002). Influence of the design of facilities at auction markets and animal handling procedures on bruising in cattle. Veterinary *Record, 150,* 743-748.

Wilson, B. W., Nieberg, P. S., Buhr, R. J., Kelly, B. J. and Shultz, F. T. (1990). Turkey muscle growth and focal myopathy. *Poultry Science, 69*, 1553-1562.

Wythes, J. R. and Shorthose, W. R. (1991). Chronological age and dentition of effects on carcass and meat quality of cattle in Northern Australia. *Australian Journal of Experimental Agriculture, 31,* 145-152.

Yeh, E., Anderson, B., Jones, P. N. And Shaw, F. D. (1978). Bruising in cattle transport over a long distance. *Veterinary Record, 103,* 117-119.

Yu, J., Tang, S., Bao, E., Zhang, M., Hao, Q. and Yue, Z. (2009). The effect of transportation on the expression of heat shock proteins and meat quality of M. longissimus dorsi in pigs. *Meat Science, 83,* 474-478.

Zahner, M., Schrader, L., Hauser, R., Keck, M., Langhans, W. and Wechsler, B. (2004). The influence of climatic conditions on physiological and behavioural parameters in dairy cows kept in open stables. *Animal Science, 78,* 139-147

Chapter 4. Effect of animal factors and distance travelled on creatine kinase levels and beef quality

Abstract

By

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The current study determined the effects of distance travelled and animal factors on creatine kinase (CK) and beef quality. Correlations among meat quality attributes (ultimate pH and colour of meat (L,* a* and b*) were also determined. Distance travelled, season, ambient temperature and lairage duration were recorded at the East London Abattoir. A total of 321 beef samples from Bonsmara, Beefmaster and Brahman were evaluated for physico-chemical meat quality attributes. The *Muscularis longissimus thoracis et. Lumborum* (LTL) muscle were used to determine ultimate pH (pHu), colour (L* - lighness, a* - redness and b* - yellowness). The Beefmaster breed had the highest values for pHu (6.3±0.05), while the Bonsmara had the highest L*(24.8±0.78), a* (17.5±0.53) and b* (12.8±0.53) values. Age of animal at slaughter (16, 18 and 24 months) had no effect on (pHu). Summer had the highest pHu values while the winter had the lowest pHu values. There were positive correlations between pre-slaughter conditions (distance travelled), pHu and colour (L^*, a* and b*) of beef. It was concluded that breed, season and age at which animals are slaughtered and distance travelled affect beef quality.

Keywords: Age at slaughter, distance, lightness, redness, season, ultimate pH.

4.1. Introduction

The quality of beef is likely to be affected by transport stress and animal related factors (Tarrant, 1981; Mitchell *et al*., 1988; Tarrant *et al.,* 1992; Vimiso, 2010). These animal factors include age of animal at slaughter, breed, and sex. Breed, sex and age of animal at slaughter being handled and exposed to stress is very essential in determining the extent to which the meat quality will be compromised. According to Bourguet *et al.* (2010) the animal stress starts at the farm with the preparation of animals for transportation and ends at the moment of slaughtering the animals (Chapter 3). During the pre-slaughter period, an animal becomes stressed because it experiences changes in the nearby environment, which can either cause it to respond to exogenous or stimuli (Ndou *et al*., 2011).

Cattle transported for short journeys less than 4 hours do not normally cause severe stress unless there is trauma (Grandin, 2000; Tarrant *et al*., 1988), and in literature there is evidence that even travelling short distances can reduce shrinkage, decrease glycogen reserves and increased meat temperature (Agness *et al.,* 1990**).** In addition, the distance travelled by animals from farm to slaughterhouse cause rapture in muscle cells, depositing some enzymes into the blood stream (Hoffman *et al.,* 1998). As transportation stress increases the activities of the enzymes such as creatine kinase also increases (CK) (Warriss *et al.,* 1998).

Creatine kinase (CK) is found in the skeletal muscles of animals and it is responsible for maintaining energy homeostasis at sites of high adenosine tri-phosphate (ATP) (Diene and Storey, 2009). Creatine kinase can be used to indicate cellular injury, heart and liver damage during transportation and poor handling (Zhang *et al*., 2010). Muscular activity and damage is due to transportation and handling which later affects the quality of beef.

The ultimate pH, colour and juiciness are the most important beef quality parameters affected by poor handling (Muchenje *et al*., 2008b). Several studies indicate that stress lowers post mortem pHu values due to repletion of glycogen in ruminants and produce tougher meat (Devine *et al.*, 2006; Gregory, 2010) on the other hand pre-slaughter stress resulted in higher pHu and darker cuts (Martinezo-Cerezo *et al*., 2005; Muchenje *et al.*, 2009a). In addition seasonal changes such as temperature affect the level of glycogen in muscles and ultimate pH after slaughter, and consequently the quality of meat (Kreikemeier *et al*., 1988; Muchenje *et al*., 2008). An increase in glycolysis results from excessive excitement, starving and stress caused by ambient temperature and long journey, which in turn leads to high post-mortem pH values (Honkavaara *et al*., 2003).

4.2. Material and Methods

4.2.1 Study site

Study site, animal management and biochemical determination of CK were described in Chapter **3.**

4.2.2. Meat quality measurements

Beef samples were collected 24 hours after slaughter for measurements of tenderness, ultimate pH and colour (L*, a* and b*). The *Muscularis longisimus thoracis et. Lumborum* (LTL) were used for measurements. The muscle was removed by cutting a sample between the 4th and 6th ribs of the loin region while the carcass was still hanging, then the samples were vacuum-packed, kept in a cooler box an hour after collection and further stored in the refrigerator at -4^{o}C.

4.2.2.1. Colour measurement

Colour of the meat (L =*lightness, a*= redness and b*= yellowness) was determined in the *Muscular longissimus thoraciset. Lumborum* (LTL) 24 hours after slaughter using a colour-guide 45/0 BYK-Gardener GmbH machine with a 20 mm diameter measurement area and illuminant D65-day light, 10^{0}standard observer. Three readings were taken by rotating the Colour Guide 90^{0} between each measurement, in order to obtain a representative average value of the colour. This guide was calibrated each day before taking measurements using the green standard.

4.2.2.2. pH measurements

Meat pH_{24} from Bonsmara, Beefmaster and Brahman of carcasses was measured in the *Muscularis longissimus thoracis et. lumborum* muscle at the level of the 10th rib 24 hours after slaughter in carcasses that were stored at 0 to 3˚C. The measurement was carried out using a portable pH meter (CRISON pH 25, CRISON Instruments, SA Spain).

The pH meter was calibrated using pH 4 pH 7 and pH 9 standard solutions (CRISON Instrument, SA Spain) before each day's measurement. The measurements were then performed with a sharpened metal sheath to prevent probe breakage and meat contamination.

4.3. Stastical analysis

The effect of breed, age group and season on CK, pHu, L,* a^* and*b* were analysed using Proc GLM of SAS (2003). The stastical model used as follows

$У_{ijk} = \mu + \alpha_i + \beta_j + \lambda_k + \varepsilon_{ijk}$

Where

$У_{ijk} =$ Response variables (CK, pHu, L, a, b and WBSF)

μ = Overall mean

α_i = breed effect (Bonsmara, Beefmaster and Brahman)

β_j = age group effect (Group 1, 2 and 3)

λ_k = seasonal effect (summer, winter, spring and autumn)

ε_{ijk} = random error term

The significant difference between least square group means for CK levels and beef quality parameters were compared using Duncan multiple range procedure. PROC GLM of Statistical Analysis System (SAS) (2003) procedure of correlations was competed to determine the relationship between pre-slaughter condition on CK and meat quality attributes.

4. 4. Results and Discussion

4.4.1 Effect of breed on meat quality

Breed had effects on beef pHu (Table 4.1). Beef from Beefmaster had the highest ($P<0.001$) pH values and the Bonsmara breed had the least pH values. The pH values were ranging from 6.3±0.05 in meat from Beefmaster and 5.6±0.05 in meat from Bonsmara. The pHu levels of Bonsmara were within the expected range and similar to those reported by Muchenje *et al.* (2008). Different temperament found within an individual breed can influence meat quality due to inherent genetic parameters. This study agrees with the findings by Grandin (1997), Mormede *et al.* (2002), Fazio and Ferlazo (2003) who reported that breed temperament influence the degree to which animals respond to stress. The lowest pHu values from Bonsmara breed might reflect to higher glycogen usage during the pre-slaughtering phase. Bonsmara is more susceptible to transportation, harsh environmental conditions and handling stress. Glucose undergoes glycolysis but in the absence of oxygen, lactic acid is formed which causes pH in muscle to drop (Muchenje *et al.*, 2009a).

However, pHu values from Beemaster breed were above the normal range. Beef with pHvalues higher than 6.0 is undesirable because of its dark colour (Muchenje *et al*., 2008). Beefmaster are known to have aggressive behaviour and which resulted in dark meat. The higher pHu values from beefmaster could be due to stress prior to slaughter.

Table 4. 1 Effects of breed on ultimate pH and colour of meat from cattle slaughtered at East London abattoir (±s.e.m).

	Breed		
Parameters	**Bonsmara**	**Beefmaster**	**Brahman**
N	108	130	83
CK(U/1)	705.3±80.57[a]	657.3±73.85[ab]	461.8±80.63[b]
pH_u	5.6±0.05[b]	6.3±0.05[a]	6.1±0.06[a]
L^*	24.8±0.78a[a]	24.6±0.71[a]	21.7±0.89[b]
a^*	17.5±0.53[a]	16.8±0.53[a]	13.2±0.66[b]
b^*	12.8±0.53[a]	11.0±0.49[a]	9.6±0.61[b]

[a,b]Means in the same row with different superscripts are significantly different at $^*P<0.05$,

pH_u = ultimate pH; L^* = lightness; a^* = redness and b^* = yellowness and CK = creatine kinase

It may as a result of transportation, rough handling, inclement temperatures, or anything that causes the animal to draw on its glycogen reserves before slaughter. Beefmaster are aggressive animals and are more prone to injuries resulting in carcass damage and poorer meat quality. The current results are in agreement with Voisinet *et al. (*1997) and King *et al.* (2006) who showed that cattle which are highly temperamental produced a high incidence of borderline dark cutters than calm cattle. The behaviour of animals prior to slaughter is related to heavy muscular work that influences glycogen content, on which final pH meat depends (Mach *et al*., 2008). Therefore lower glycogen amounts result in production of small amounts of lactic acid and thus high pHu. Ultimate pH found in muscle is one of the contributing factors to the quality of meat. Welglarz *et al*. (2002), Kogel (2005) and Mach *et al*. (2008) found that beef with high pH_u and dark colour had negative effects on meat quality and leads to DFD (Dark Firm Dry) meat.

Breed had no effects (P >0.001) on L* values, the darker meat (low L* values) from the three breeds: Bonsmara, Beefmaster and Brahman may be attributed by reduced muscle glycogen and increased myoglobin because of the light scattering properties of meat. The darker meat produced by the three breeds in comparisons to the improved breeds agrees with O'Neill *et al.* (2006). O'Neill *et al*. (2006) observed that indigenous breeds such as Bonsmara released more catecholamines than exotic breeds during the pre-slaughter period, causing the depletion of glycogen. Another reason could be due to the long journeys they travelled, poor handling before slaughter Commission International De I Eclairage (1976), found that in their study that dark meat is due to muscle glycogen that has been used up rapidly during the handling, transport and pre-slaughter period, after slaughter there is little lactic acid production which results in Dark-Firm-Dry (DFD) meat.

Klont *et al.* (1999), Lensik *et al*. (2001), Lensink (2002) and Zhang *et al*. (2005) observed that in their studies stress has a tendency to make meat darker and had significantly lower values of $L,^*$ a^* and b^*. Bonsmara had the highest a* values with Brahman having the least a* values. The a* values were ranging from 17.5±0.53 in meat from Bonsmara and 13.2±0.66 in meat from Brahman. The pH values from these breeds were within the normal range. This is because pH_u values were fairly normal and colour is not greatly affected by pre-slaughter condition such as transportation. This is supported by Abril *et al*. (2001) who found that colour is not affected by transport time but ageing decreased the redness and yellowness which accompanies the natural discolouration of fresh meat from red to brown.

Bonsmara had the highest b* values (12.8±0.53) and the lowest b* values (9.6±0.61) in the Brahman breed. This could be due to the degree to which animals are stressed during handling and transportation depends on their temperament, handling during the journey, condition of the animal, and duration of the journey resulting in pale colour.

4. 4. 2 Effect of age on meat quality

The effects of age on meat quality are shown in (Table 4.2). Meat from 16, 18 and 24 months old animals had no effect on pHu. The pHu values ranging from 6.1 in meat from 16 months old animals, 6.1 in meat from 18 months old cattle and 6.1 in meat from 24 months old cattle. The pHu values from 16, 18 and 24 months old cattle were above the normal pH range. The meat pH above 6.0 is a dark red colour (Bartos *et al*., 1993; Mounier *et al*., 2006), increased tenderness (Silva *et al*., 1999) and poor palatability (Viljoen *et al.*, 2002; Wulf *et al.*, 2002). These results are mostly likely to be due to the different state and concentrations of myoglobin of different ages. This can also be attributed to the fact that an animal's behaviour differs with an animal's age and the stage of growth (Terlouw, 2005).

Different age groups had no effect on L* values, this could be due to higher levels of pH obtained from the three age group categories. The meat often has sticky consistency and poorer taste and flavour. The pH of meat affects its colour (Abril *et al*., 2001; Weglarz, 2010). Meat colour may be influenced by the activity undertaken by the animal prior to slaughter. Therefore meat becomes darker with increasing animal age. These results are in agreement with Funghi *et al*. (1994), who found that L^*values decreased with the increase in slaughtering age. Zhang *et al.* (2005) found that high pH meat had lower L^*(lightness), a^*(redness) and b^* (yellowness) values than normal pH meat, indicating that high pH meat is darker and less brown than is normal pH meat.

Meat from 16 months old animals had the highest a* and b* (17.9±0.53) and (12.9±0.49) and the 24 months old animals had the least a*and b* values of 12.9±0.54 and 9.3±0.51. Differences in meat colour have been associated with variations in intramuscular fat and moister content, age dependent in muscle myoglobin content (Lawry, 1974). Animals that had fed on pasture have a yellow fat colour because of the high levels of beta-carotene contained by grass; consumers often perceive meat with yellow fat as having come from an old or diseased animal (Priolo *et al*., 2001).

Table 4. 2 Effects of age on ultimate pH and colour of meat from cattle slaughtered at East London abattoir (±s.e.m).

Parameters	**Age**		
	16months	**18months**	**24months**
N	120	87	114
CK (U/1)	636.9±76.83[b]	729.6±89.75[a]	524.1±0.63[c]
pH_u	6.1±0.05[a]	6.1±0.06[a]	6.1±0.05[a]
L^*	25.2±0.71[a]	26.6±0.83[a]	20.9±0.73[b]
a^*	17.9±0.53[a]	17.7±0.62[a]	12.9±0.54[b]
b^*	12.9±0.49[a]	11.5±0.58[b]	9.3±0.51[c]

[a,b, c]Means in the same row with different superscripts are significantly different at $^*P<0.05$

pH_u = ultimate pH; L^* = lightness; a^* = redness and b^* = yellowness and CK = creatine kinase

4. 4.3 Effect of season on meat quality

Spring season had the highest (P<0.001) pHu values and the autumn season had the lowest pHu values. This can be attributed to the variations in environmental conditions. The study is not in agreement with the findings by Gurdia *et al*. (2004) and Van der Perre *et al*. (2010) reported that Dark Firm and Dry meat is expected to be high in summer than other seasons. Stressful environment results in the reduction of glycogen levels in the muscles resulting in lower lactic acid production and high pHu. Winter had the highest L* values, while summer had the least L* values. The study contradict with the findings by the Maria *et al*. (2006) who found that the lowest L* values expected to be low in winter where there is a shortage of feed during the winter season causing beef dark cutting. This shows that the summer season was more stressful than winter season resulting in higher myoglobin and decreased glycogen results in darker meat (Priolo *et al*., 2001).

Winter season had the highest a* values and the lowest a* values in the summer season. Autumn had the highest b* values and the lowest b* values were observed in summer season. This is not in agreement with Kim *et al.* (2003) who indicated colour coordinates as affected by temperature, winter season and ante-post-mortem short term stressors as they change the normal muscle metabolism. However, b* values for other season were in the acceptable range as indicated by Muchenje *et al.* (2009).

Table 4. 3 Effects of seasons on ultimate pH and colour of meat from cattle slaughtered at East London abattoir (±s.e.m).

Parameters	Spring	Summer	Winter	Autumn
N	196	43	32	50
CK (U/1)	698.3±59.19[a]	181.3±124.09[b]	704.1±43.85[a]	658±116.25[a]
pHu	6.3±0.03[a]	6.0±0.07[b]	5.9±0.08[b]	5.4±0.06[c]
L*	24.6±0.52[b]	15.3±1.11[c]	30.3±1.28[a]	24.8±1.03[b]
a*	18.0±31[a]	4.7±0.66[b]	18.4±0.77[a]	16.9±0.61[a]
b*	10.5±0.39[c]	5.0.±0.66[d]	14.1±0.76[b]	18.1.±0.61[a]

[a,b,c,d] Means in the same column with different superscripts are significantly different at $^{*}P<0.05$,

pH_{u} = ultimate, L*= lightness, a*= redness, b* = yellowness and CK = creatine kinase

4 .4.4 Correlation between distance travelled and CK levels on meat quality attributes.

Correlation co-efficients between distance travelled and CK levels on meat quality attributes (L*, a* and b*) are presented in Table 4.4. From the results there was no correlation observed between CK levels and meat quality attributes (lightness (L*) and yellowness (b*) except for the redness (a*) of the meat. It was postulated that CK levels would be an indication of poor animal handling and therefore be linked to poor beef quality (Table 4.4). The study agrees with the findings by Warries *et al*. (1990) and Mach *et al.* (2008) indicated that creatine kinase was not related to any of the meat quality parameters, but can be linked to muscle exhaustion that occurs prior to slaughter. The CK activity in the blood due to poor handling negatively affects the quality of meat (Yu *et al.*, 2009).

The distance travelled had the negative relationship on ultimate pH and colour of beef. The longer the distance travelled by the animals to the abattoir the higher the pHu will be. This study agreed with the findings of Honkavaara *et al*. (2003), shorter or longer duration of animal transport to slaughterhouse had a great effect on high pH values of meat than when transport exceeded 12h. Pre-slaughter stress can affect several aspect of meat quality in livestock animals (Tarrant *et al.*, 1992). Several authors indicate that the distance travelled by animals from the farms to the slaughterhouse affect meat quality (Franc *et al*., 1990; Pipek *et al*., 2003; Jelenikova, 2008). This implies that transportation affects glycogen levels (Devine *et al*., 2006; Ferguson and Warner, 2008; Muchenje *et al.*, 2009b). Miranda-de la Lama *et al.* (2009) reported that rough roads cause stress but Kadim *et al.* (2009) reported stress on smooth road which then increases muscle pH and this is in agreement with the current study.

Table 4. 4 Correlations between creatine kinase level, distance and meat quality (pHu, L^*, a^* and b*) from Bonsmara, Beefmaster and Brahman slaughtered at East London abattoir

	L^*	a^*	b^*	CK level	Distance
L^*		0.21^{***}	0.15^{**}	0.06	0.35^{***}
a^*			0.45^{***}	0.193^{***}	0.26^{***}
b^*				0.05	0.44^{***}
CK level					-0.01
Distance					

Significantly correlated at $^{**}P<0.01$, $^{***}P<0.001$

pH_u = ultimate pH; L^*= lightness; a^*=redness and b^*= yellowness . CK =creatine kinase

4. 5 Conclusion

The study showed that animal related factors affect the quality of beef, with Beefmaster having the highest pHu values. In addition seasonal changes also affect the quality of beef, with spring season having highest pHu values. On the other hand distance had a negative relationship on beef quality (L*, a* and b*), however no correlation was observed between distance and CK levels. It was also concluded that there is no significant relationship between meat parameters (L,* a* and b*) and CK.

4 .6. References

Abril, M., Campo, M. M. A., Onemic Sanudo, C., Alberti, P. and Negueruela, A. I. (2001). Beef colour evolution as a function of ultimate pH. *Meat Science, 58*, 69-78.

Agness, F., Sartorelli, P., Borrow, H. A. and Locatelli, A. (1990). Effect of transport loading or noise on blood biochemical variables in calves. *American Journal of Veterinary Research, 51*, 1679-1681.

Bartos, L., France, C., Rehak, D. and Stipkova, L. (1993). A practical method to prevent dark cutting (DFD) in beef. *Meat Science*, *34*, 275-282.

Bourguet, C., Deiss, V., Gobert, M., Durand, D., Boissy, A. and Terlouw, E. M. C. (2010). Characterising the emotional reactivity of cows to understand and predict their stress reactions to the slaughter procedure. *Applied Animal Behaviour Science*, *125,* 9-21.

Commission International De I' Eclairage (1976). Calorimetry (2nd ed.). Vienna, Switzerland: CIE.

Devine, C. E., Lowe, T E., Wells, R W., Edwards, N. J. and Edwards, J. E. H. (2006). Pre-slaughter stress arising from on –farm handling and its interactions with electrical stimulation on tenderness of lambs. *Meat Science, 73*, 304-312.

Diene, C. A. and Storey, K. B. (2009). Creatine kinase regulation by reversible phosphorylation in frog muscle. *Comparative Biochemical and Physical, Part B*, *152,* 405-412.

Fazio, E. and Ferlazzo, A. (2003). Evaaluation of stress during transport. *Veterinary Research Communications*, *27 (Supplement),* 519-524.

Ferguson, D. M. and Warner, R. D. (2008). Have we underestimated the impact of pre-slaughter stress on meat quality in ruminants? *Meat Science, 80, 12-19.*

Franc, C., Bartos, L., Bouska, J., Burda, J. and Slanec, E. (1990). Prevention of the DFD defect of beef by housing of cattle in single-animal boxes at the slaughterhouse. In Czech. *Zivocisna Vyroba, 35,* 603-608.

Funghi, R., Pugliese, , C., Bozzi, R. and Lucifero, M. (1994). Evolution of performances of Chianina young bull from 6-24 months. Quality parameters of meat. 81-86 in Production. 7th International Congress on Italian beef cattle contest, Perugia, Italia.

Grandin, T. (1997). Assessment of stress during handling and transport. *Journal of Animal Science, 75*, 249-257.

Grandin, T. (2000). Livestock handling and transport, 2nd edition, CAB International Wallingford, Oxon, United Kingdom.

Gregory, N. G. (2010). How climatic changes could affect meat quality. *Food Research International, 43,* 1866-1873.

Guardia, M. D., Estany, J., Balasch, S., Oliver, M. A., Gispert, M. and Diestre, A. (2004). Risk assessment of PSE condition due to pre-slaughter conditions and RYRL gene in pigs. *Meat Science, 67,* 471-478.

Hoffman, D. E., Spire, M. F., Schwenke, J. R. and Unruh, J. A. (1998). Effect of source of cattle and distanced transported to a commercial slaughter facility on carcass bruises in mature beef cows. *Journal of the American Veterinary Medical Association*, *212,* 668-672.

Honkavaara, M., Rintasalo, E., Ylonen, J. and Pudas, T. (2003). Meat quality and transport stress of cattle. *Deutsche Tierarztliche Wonchenschrift, 110,* 125-128.

Jelenikova, J., Pipek, P. and Staruch, L (2008). The influence of ante-mortem treatment on relationship between pH and tenderness of beef. *Meat Science, 63,* 509-513.

Kadim, I. T., Mahgoud, O., Al-Marzooqi, W., Khalaf, S., Al-Sinawi, S. S.H. and Al-Asammi, I. S. (2009). Effects of transportation during the hot season and low voltage electrical stimulation on histochemical and meat quality characteristics of sheep *longissimus* muscle. *Livestock Science, 126,* 154-161.

Kim, Y. S., Yoon, S. K., Song, Y. H. and Lee, S. K. (2003). Effects of season on colour of Hanwoo (Korean native cattle) beef. *Meat Science, 63, 509-513.*

King, D. A., Schuehle Pfeiffer, C. E., Randel, R. D., Welsh, T. H, Oliphant, R. A., Baird, B. E., Curley, K. O. T. R., Vann, R. C. and Savell, J. W. (2006). Influence of animal temperament and stress responsiviness on the carcass quality and beef tenderness of feedlot cattle. *Meat Science, 74,* 546-556.

Klont, R. E., Bamier, V. M. H., Smulders, F. J. M., Van Dijk, A., Hoving-Bolink, A. H. and Eikelenboom, G. (1999). Post-mortem variation in pH, temperature, and colour profiles of carcasses in relation to breed, blood haemoglobin content, and carcass characteristics. *Meat Science, 53,* 195-202.

Kogel, J. (2005). Hereditary and environmental effects on the quality of beef. *Animal Science Papers and Reports, 23* (4), 281-302.

Kreikemeier, K. K., Unruh, J. A. and Eck, T.P. (1998). F actors affecting the occurrence of dark-cutting beef and selected carcass traits in finished beef cattle. *Journal of Animal Science, 76,* 388-395.

Lawry, R. A. (1974). Meat Science. 2nd edition. Oxford, Pergamon Press.

Lensink, B. J. (2002). The human-animal relationship in animal production. *First Virtual Global Conference on Organic Beef Cattle Production September, 02 to October,* 15-2002.

Lensink, B. J., Fernandez, X., Cozzi, G., Florand, L. And Veissier, I. (2001). The influence of farmers behaviour towards calves on animal's responses to transport and quality of veal meat. *Journal of Animal Science, 79*, 642-652.

Mach, N., Bach A., Velarde A. and Devant, M. (2008): Association between animal, transportation, slaughterhouse practices, and meat pH in beef. *Meat Science, 78, 232–2*

Maria, G. A., Buil, T., Liste, G., Villarroel, M., Sanudo, C. and Olleta, J. L. (2006). Effects of transport time and season on aspects of rabbit meat quality. *Meat Science, 72*, 773-777.

Martnezo-Cerezo, S., Sanudo, C., Panea, B., Medel, I., Delfa, R., Sierra, I., Beltran, J. A., Cepero, R. and Olleta, J. L. (2005). Breed, slaughter and ageing time effects on physic-chemical characteristics of lamb meat. *Meat Science, 69*, 323-333.

Miranda-de la Lama, G. C., Villarrol, M., Olleta, J. L., Alierta, S., Sanudo, C. and Maria, G. A. (2009). Effect of the pre-slaughter logistic chain on meat quality of lambs. *Meat Science, 83,* 606-609.

Mitchell, G., Hattingth, J. and Ganhao, M. (1988). Stress in cattle assessed after handling, after transport and after slaughter. *Veterinary Record, 123*, 201-205.

Mormede, P., Courvoisier, H., Ramos, A., Marissal-Avry, N., Ousova, O. and De 'saute's, C. (2002). Molecular genetics approaches to investigate individual variations in behavioural and neuroendocrine stress responses. *Phychoneuroendocrinology, 27*, 563-583.

Mounier, L., Dubroeucq, H., Andanson, S. And Veissier, I. (2006). Varriations in meat pH of beef bulls in relation to conditions of transfer to slaughter and previous history of the animals. *Journal of Animal Science, 84*, 1567-1576.

Muchenje, V., Dzama, K., Chimonyo, M., Raats, J. G. and Strydom, P.E. (2008). Meat quality of Nguni, Bonsmara and Aberdeen Angus steers raised on natural pasture in the Eastern Cape, South Africa, *Meat Science, 79,* 20-28.

Muchenje, V., Dzama, K., Chimonyo, M., Strydom, P.E. and Raats, J.G. (2009b). Relationship between pre-slaughter stress responsiveness and beef quality in three cattle breeds. *Meat Science, 81,* 653-657.

Muchenje, V., Dzama, K., Chimonyo, M., Strydom, P. E., Hugo, A. and Raats, J. G. (2008b). Sensory evaluation and its relationship to physical meat quality attributes of beef from Nguni and Bonsmara steers raised on natural pastures. *Animal, 2,* 1700-1706.

Muchenje, V., Dzama, K., Chimonyo, M., Strydom, P. E., Hugo, A. and Raats, J. G. (2009a). Some biochemical aspects pertaining to beef eating quality and consumer health: a review. *Food Chemistry, 112,* 279-289.

Ndou, S. P., Muchenje, V. and Chimonyo, M. (2011). Animal welfare in multipurpose cattle production systems and its implications on beef quality. *African Journal of Biotechnology, 10(7),* 1049-1064.

O'Neill, H. A., Webb, E. C., Frylinck, L. and Strydom, P. E. (2006). The stress responsiviness of three different beef breed types and the effect on ultimate pH and meat colour. In *Proceedings 52nd International Congress of Meat Science and Technology (*pp.181-182). 13-18, August 2006, Dublin, Ireland.

Pipek, P., Harberl, A., and Jelenikova, J. (2003). Influence of slaughterhouse handling on the quality of beef carcasses. *Czech Journal of Animal Science, 39,* 371-378.

Priolo, A., Micol, D. and Agabriel, J. (2001). Effects of grass feeding systems on ruminant eat colour and flavour: A review. *Animal Research, 50,* 185-200.

SAS. (2003). In : SAS/STAT Guide to Personal Computers, Version 6, Statistical Analysis System Institute Inc. Cary, NC.

Silva, J. A., Patarata, L. And Martins, C. (1999). Influence of ultimate pH on bovine meat tenderness during ageing. *Meat Science, 52*, 453-459.

Tarrant, P. V. (1981). The occurrence, causes and economic consequences of dark-cutting in beef- a survey of current information. In D. E. Hood, and P. V. Tarrant (Eds). *The problem of dark cutting in beef (pp. 3-34).* The Hague: Martinus Nijholf.

Tarrant, P. V., Kelly, F. J. and Harrington, D. (1988). The effect of stocking density during 4 hour transport to slaughter, on behaviour, blood constituents and carcass bruising in Friesland steers. *Meat Science, 24*, 209-224.

Tarrant, P. V., Kenny, F. J., Harrington, D. and Murphy. M. (1992). Long distance transportation of steers to slaughter: effect of stocking density on physiology behaviour and carcass quality. *Livestock Production Science, 30*, 223-238.

Terlouw, C. (2005). Stress reactions at slaughter and meat quality in pigs: genetic background and prior experience. A brief review of recent findings. *Livestock Production Science, 94*, 125-135.

Van de Perre Ceustermans, A., Leyten, J. and Geers, R. (2010). The prevalence of PSE characteristics in pork and cooked ham- Effects of season and lairage time. *Meat Science, 86*, 931-937.

Viljoen, H. F., De Kock, H. L. and Webb, E. C. (2002). Consumer acceptability of dark, firm and dry (DFD) and normal pH beef steaks. *Meat Science, 61*, 181-185.

Vimiso, P. (2010). Effects of marketing channel on bruising, ultimate pH and colour of beef; and stakeholder perception on quality of beef from cattle slaughtered at a small holder abattoir. MSc Thesis, University of Fort Hare, South Africa.

Voisinet, B. D., Grandin, T., O'Connor, S. F., Tatum, J. D. and Deesing, M. J. (1997). Bos indicus cross feedlot cattle with excitable temperaments have tough meat and a higher incidence of borderline dark cutters. *Meat Science, 46*, 367-377.

Warriss, P. D., Brown, S. N., Knowles, T. G., Edward, J. E., Kettlewell, P. J. and Guise, H. L. (1998). The effect of stocking density in transit on the carcass quality and welfare of slaughter pigs: 2. Results from the analysis of blood and meat samples. *Meat Science, 50 (4),* 447-456.

Warriss, P., Kestin, S. C., Young, C. S., Bevins, S. N. (1990). Effect of pre-slaughter transport on carcass yield and indices of meat quality I sheep. *Journal of the Science and Food Agriculture, 51*, 517- 523.

Weglarz, A. (2010). Beef quality depending on cattle category and slaughter season. *Czech Journal Animal Science, 55, (12),* 548-556.

Weglarz, A., Zapletal, P., Gil, Z., Skrznnsk, G. And Adamczk, K. (2002). The effect of sex and age on beef quality in Poilish, summary in English. *Zeszyty Nankowe Przeglada Hodowlanego, 62,*211-216.

Wulf, D. M., Emnett, R. S., Leheska, J. M. and Moeller, S. J. (2002). Relationship among glycolytic potential, dark cutting (dark, firm and dry) beef, and cooked beef palatability. *Journal of Animal Science, 78,* 2595-2607.

Yu, J., Tang, S., Bao, E., Zhang, M., Hao, O. And Yue, Z. (2009). The effect of transportation the expression of heat shock proteins and meat quality of M. longissimus dorsi in pigs. *Meat Science, 83,* 474-478.

Zhang, M., Xin, L., Bao, E., Hartung, J. and Yue, Z. (2010). Variation in the expression of Hsp27, αB-crystllin mRNA and protein in the heart and liver of pigs exposed to different times. *Research in Veterinary Science, 90 (3),* 432-438.

Zhang, S. X., Farouk, M. M., Young, O. A., Wieliezco, K. J. and Podmore, C. (2005). Functional stability of frozen normal and high pH beef. *Meat Science, 69*, 765-772.

Chapter 5: General Discussion, Conclusion and Recommendations

5.1 General Discussion

Poor handling and transportation of animals can lead to bruising and some in cases more serious injury or death. Transport and stress can have a negative effect on the colour, tenderness of beef, while bruising had a negative effect on industry due to the most expensive cuts of meat to be trimmed and sometimes the condemnation of the whole carcass. Bruising also results in the removal of some parts of the meat during carcass inspection which may assume economic relevance. Therefore, the focus of the current study was to determine the occurrence of bruising in slaughter cattle and its relationship with creatine kinase and meat quality. The effects of pre-slaughter conditions, animal related factors and their relationship with bruising and creatine kinase levels were determined in Chapter 3. In Chapter 4, the effects of distance travelled and animal factors on creatine kinase levels and meat quality were determined.

In chapter 3 the effect of pre-slaughter conditions, animal related factors and their relationship with bruising and creatine kinase CK levels were determined. It was observed that CK affected breed with Bonsmara recorded with the highest CK levels. Animals differ in their tolerance of stressful situations and consequently in their stress status (Bourguet *et al*., 2010). The CK activity increases when animals were roughly driven and mishandled before and after transport (Broom *et al*., 1996; Averos *et al.,* 2008). Temperature is associated with a season such that during cold a season (winter season) animals had higher CK levels than in the summer season. Increased CK levels in winter are associated with external air temperature and air humidity which also have a negative effect on the welfare of cattle during transportation.

Young animals are at the risk of more bruising than older animal resulting in high CK levels (Parrot *et al.*, 1994). Among the three age categories, group 2 (18 months) cattle was recorded with higher values for CK levels while group 1 (16 months) had the least CK values. In addition, male animals are more susceptible to stressful conditions. In the current study, it was recorded that male animals had higher CK levels than female animals. Eguinoa *et al.* (2003) indicated that the reason young animals are easily stressed is because of the organisation in fat deposition and subcutaneous tissue between cattle, sex and age (Jarvis *et al.*, 1995).

From this study animal factors such as age, sex and type of animal were found to affect the development of bruises in cattle. Bonsmara had the highest bruising score (Chapter 3). The Bonsmara was the most responsive to stress. The more the animal is handled, the greater the chance on injuries leading to carcass bruising. Bruising is evidence of poor handling (Gregory, 1996). Young animal had an effect on bruising such that 16 month animals had the highest bruising score than the older animals. Differences in age categories could be associated with rough handling, violent impact of animals against protuberant or sharp-edged surfaces, or with aggression between animals, mechanical damage to the animal's tissues can occur which may develop into a bruise.

In Chapter 4, among the three breeds, Beefmaster had higher values for pHu, which resulted in darker meat colour than Bonsmara and Brahman breeds. This could be attributed to the inherent genotype differences and, possibly, the differences in their response to pre-slaughter handling. A high pHu reflects a depletion of muscle glycogen due to stress or other factors (Dhana *et al*., 2003; Muchenje *et al.,* 2009; Mushi *et al*., 2009). However higher values for L*, a* and b* were observed in Bonsmara. This is due to the fact that animals have different growth rates that affect intramuscular fat content (Ndlovu *et al.,* 2009).

Season had a significant effect on pHu, such that summer had highest pH values. This may be attributed to the fact that there was more huddling together in the truck as well as more activity. The study agreed with the findings by Gregory (2010) and Dalla Costa *et al*. (2007) who reported that summer conditions have been found associated with pale meat colour compared with wintertime. Age at slaughter had no significantly effect on ultimate pH. However, the differences observed for the colour of meat (L*, a* and b*) among the age categories; 16, 18 and 24 months old were within the normal range. Meat from the group 1 (16 months), group 2 (18 months) and group 3 (24 months) can be used for consumption. There were negative correlations between pre-slaughter conditions and the quality of beef. Distance travelled was positively correlated with beef colour (L*, a* and b*). L* values and a* were strongly related with distance travelled. However, no correlation was observed between distance travelled and CK levels. Factors that also affect meat quality are pre-slaughter treatment (Santos *et al*., 2008) and pre-slaughter conditions. Therefore there is no relationship between pre-slaughter conditions and creatine kinase levels.

5.2 Conclusion

It was concluded that breed, season, age at which animals are slaughtered and pre-slaughter conditions contribute to bruising and affect ultimate pH and colour of beef. Breed, season, sex and age at slaughter also had an effect on CK levels. Among pre-slaughter stressors, the distance travelled had an effect on the quality of beef leading to a dark firm and dry meat and high pH levels, even though creatine kinase was not really affected by the distance travelled, but it was postulated that length of journey increase the activity of CK. Therefore, it can be stated that poor handling from the farm and during transportation should be taken into account in order to reduce stress before slaughter in order to improve better beef quality.

5.3 Recommendations

In the current study it is recommended that:

To prevent high chances of bruising due to poor handling, the study should aim on the following procedures:

1. Occurrence of bruising is it at the farm, during transportation or at the slaughterhouse? Seems to have serious effects on the welfare of animals and it can lead to significant loss of meat quality and production.
2. The way animals are handled at loading at the farm, during transportation, at unloading and during lairage.
3. Evaluation of facility conditions at the farm, livestock markets and transportation of animals.
4. There is a need to determine stress effect on meat quality and understand the relationship of glycogen and lactic acid to pH decline in meat after slaughter.
5. To evaluate physiological and biochemical changes or stress hormones and their relationship with technological attributes of beef at the farm, at arrival, and just before slaughter. This is necessary because physiological parameters such as CK affect meat quality due to high levels of CK.

5.4 References

Averos, A., Martin, S., Riu, M., Serratosa, J. And Gosalvez, L. F. (2008). Stress response of extensively reared young bulls being transported to growing- finishing farm under Spanish summer commercial conditions. *Life Science, 119,* 334-340.

Bourguet, C., Deiss, V., Gobert, M., Durand, D., Boissy, A. and Terlouw, E. M. C. (2010). Charaterising the emotional reactivity of cows to understand and predict their stress reactions to the slaughter procedure. *Applied Animal Behaviour Science, 125,* 9-21.

Broom, D. M., Goode, J. A., Hall, S. J.G., Lloyd, D. M. and Parrot, R. F. (1996). Hormonal and physiological effects of a 15- hour road journey in sheep: Comparison with the responses to loading, handling and penning in the absence of transport. *Veterinary Research Communication, 27,* 515-518.

Dalla Costa, O.A., Faucitano, L., Coldebella, A., Ludke, J.V., Peloso, J.V., dalla Roza, D. and Paranhos da Costa, M.J.R. (2007). Effects of the season of the year, truck type and location on truck on skin bruises and meat quality in pigs. *Livestock Science, 107,* 29-36.

Dhanda, J. S., Taylor, D.G. and Murray, P. J. (2003). Growth, carcass and meat quality parameters of male goats: effects of genotype and live weight at slaughter. Part 1. *Small Ruminant Research, 50,* 57-66.

Eguinoa, P., Brocklehurst, S., Arana, A., Mendizabal, J.A., Vernon, R.G. and Purroy, A. 2003. Lipogenic enzyme activities in different adipose depots of Pirenaican and Holstein bulls and heifers taking into account adipocyte size.*Journal of Animal Science, 81,* 432-440.

Gregory, N. G. (1996). Welfare and hygiene during pre-slaughter handling. *Meat Science, 43,* 35-46.

Gregory, N. G. (2010). How climatic changes could affect meat quality. *Food Research International*, *43*, 1866-1873.

Jarvis, A. M. and Cockram, M. S. (1995). Effect of handling and transport on bruising of sheep sent directly from farms to slaughter. *Veterinary Record*, *135,* 523-527.

Muchenje, V., Dzama, K., Chimonyo, M., Strydom, P. E. and Raats, J. G. (2009). Relationship between pre-slaughter stress responsiveness and beef quality in three cattle breeds. *Meat Science, 81*, 653-657

Mushi, D.E., Safari, J., Mtenga, L. A., Kifaro, G.C. and Eik, L. O. (2009). Effects of concentrate level on fattening performance, carcass and meat quality of small East African x Norwegian crossed goats fed low quality grass hay. *Livestock Science, 124,* 148-155.

Ndlovu, T., Chimonyo, M. and Muchenje, V. (2009). Monthly changes in body conditions score and internal parasites prevalence in Nguni, Bonsmara and Angus steers raised on sweetvels. *Tropical Animal Health and Production, 41*, 1169-1177.

Parrot, R. F., Misson, B. H. and De la Riva, C. F. Differential stressors effects on the concentration of cortisol, prolactin and catecholamines in the blood of sheep. *Research Veterinary Science, 56,* 236-239.

Santos, V.A., Siliva, J. A., Silverstre, A. M. D., Siliva, S. R. and Azevedo, J. M. T. (2008). The use of multivariate analysis to characterize carcass and meat quality of goat kids protected by the PGI" Cabrito de Barroso". *Meat Science*, *116,* 70-81.

Appendice 1 Cattle transport recording sheet

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Cattle transport recording sheet

Delivery date	**Loading/departure time**	**Arrival time**	**Slaughter date**	**Slaughter time**	**No. of cattle in vehicle**	**Farm to abattoir distance (km)**	**Day temp**

Appendice 2 Carcass record sheet for ultimate pH and colour

Carcass no.	Breed	Weight	Sex	Class	Age	pH24	Colour at 24h		
							L*	a*	b*

Appendice 3 Bruise score record sheet

Carcass no. ....   Slaughter date

Breed   ....   Sex

Left side   **Right side**