Meat quality characteristics of the plains zebra (*Equus quagga)*

by

Angelique Henn

Thesis presented in partial fulfilment of the requirements for the degree of

MASTER OF SCIENCE IN ANIMAL SCIENCE

In the Faculty of AgriSciences at Stellenbosch University

Supervisor*:* Prof. L.C. Hoffman

Co-supervisor: Dr. H. Lambrechts

March 2020

DECLARATION

By submitting this thesis electronically, I declare that the entirety of the work contained therein is my own, original work, that I am the sole author thereof (save to the extent explicitly otherwise stated), that reproduction and publication thereof by Stellenbosch University will not infringe any third party rights and that I have not previously in its entirety or in part submitted it for obtaining any qualification.

Date: March 2020

SUMMARY

This study presents the first available baseline information on the carcass contribution, physical meat parameters, chemical and mineral composition of plains zebra meat, and sensory profile and optimum ageing period of seven selected plains zebra (*Equus quagga*) muscles [*Longissimus thoracis et lumborum* (LTL), *semimembranosus (*SM), *biceps femoris (*BF), *semitendinosus (*ST), *infraspinatus (*IS), *supraspinatus* (SS), and *psoas major* (PM)]. The muscles were obtained from animals cropped during a wet winter season (n = 8) and a dry summer season (n = 12) in the Western Cape Province of South Africa.

The average undressed carcass weight of the winter-harvested stallions and summer-harvested stallions were 324.4 ± 5.55 kg and 291.50 ± 11.65 kg, respectively. Average cold carcass weight reported for the winter- and summer-harvested groups were 188.3 ± 4.03 kg and 164.5 ± 5.53 kg, respectively. The warm and cold dressing percentages were numerically higher for the winter-harvested animals (59.5 ± 0.55 % and 58.0 ± 0.60 %, respectively) than for the summer-harvested animals (58.1 ± 0.68 % and 56.60 ± 0.70 %, respectively). Harvest season did not influence the proportional contribution of the LTL, SM, BF, ST, IS, SS and PM to the cold carcass weight. A considerable amount of internal offal (21.8 to 22.5 % of the undressed cold carcass weight) has the potential to be used as a low-cost protein source.

All the physical parameters (pH_u, drip loss, cooking loss, shear force and colour coordinates) differed between the muscles and seasons respectively, except for the CIE a* and chroma. The ultimate pH (pH_u) of all the muscles fell within the biological normal range and was not classified as dark firm and dry meat. Most of the physical measurements (pH_u, drip loss, cooking loss and colour coordinates) were comparable to values characteristic to red meat, the exception being the high shear force values for samples obtained from both seasons. Meat samples from the winter-harvest animals were intermediate in terms of toughness, compared to summer-harvest samples that were characterised as tough. The ST and SS of the winter-harvested animals, and the IS of the summer-harvested animals, were intermediate in terms of toughness. The remainder of the respective muscles obtained from both groups had shear force values representative of tough meat. The CIE colour of the meat samples obtained from both groups corresponded to the intermediate range associated with game meat.

The selected muscles differed significantly in terms of moisture, protein, and intramuscular fat contents. The muscle protein content was the only component influenced by season of harvest, with winter-harvested samples having a higher protein content when compared to the summer-harvested samples (21.8 ± 0.18 g/100g vs. 20.7 ± 0.12 g/100g). Season-muscle interactions were reported for the intramuscular fat and ash content, while strong negative correlations were reported for pooled moisture and protein. The primary macro- and micro-minerals present in the LTL, SM, BF, liver, and rib included potassium, phosphorous, sodium and magnesium together with iron, zinc, copper, selenium (except in the rib), manganese and strontium. Muscle type influenced the sodium, iron, copper, manganese, and strontium levels. The significant differences for the proximate and mineral composition observed for each of the main effects were marginal, and therefore it is debatable whether it is of biological consequence in terms of human health when consumed.

The sensory profile of plains zebra meat can be characterised as game-like, beef-like, sweet-associated, and herbaceous aromas and flavours. The LTL, SM and BF muscles differed significantly in terms of sensory profile, and fatty acid content, with the BF having a distinct sensory profile as well as a higher fatty acid content.

An ageing trial was conducted to determine the optimum ageing period needed to reach the maximum meat tenderness for the LTL, SM and BF muscles. An improved bloomed surface colour and maximum tenderness was achieved at 14 days and 20 days post-mortem for summer-harvested samples, respectively. Colour stability of the muscles were high as no visual discolouration was observed up to day 32 of post-mortem ageing.

Findings from this study will contribute meaningfully to the establishment of meat production potential of plains zebra under controlled farming conditions, thus investigating the potential of this species to contribute to food security in South Africa.

OPSOMMING

Hierdie studie bied die eerste beskikbare basislyninligting oor die karkasbydrae, fisiese vleisparameters, chemise- en mineraalsamestelling, sensoriese profiel en optimale verouderingstydperk vir sewe geselekteerde vlakte zebra (*Equus quagga*) spiere [*Longissimus thoracis et lumborum* (LTL)), *semimembranosus* (SM), *biceps femoris* (BF), *semitendinosus* (ST), infraspinatus (IS), *supraspinatus* (SS) en *psoas major* (PM)]. Die spiere is versamel van diere wat tydens 'n nat winterseisoen (n = 8) en 'n droë somerseisoen (n = 12) in die Wes-Kaap Provinsie van Suid-Afrika geoes is.

Die gemiddelde dooie/intakte karkasgewig van die hingste wat in die winter geoes is en die hingste in die somer geoes is, was onderskeidelik 324,4 ± 5,55 kg en 291,50 ± 11,65 kg. Gemiddelde koue karkasgewig vir die winter- en somer-oesgroepe was onderskeidelik 188,3 ± 4,03 kg en 164,5 ± 5,53 kg. Die warm en koue uitslagpersentasie was numeries hoër vir die diere wat in die winter geoes is (onderskeidelik 59,5 ± 0,55% en 58,0 ± 0,60%), wanneer vergelyk word met die diere wat in die somer geoes is (onderskeidelik 58,1 ± 0,68% en 56,60 ± 0,70%). Die seisoen van oes het nie die proporsionele bydrae van die LTL, SM, BF, ST, IS, SS en PM tot die koue karkasgewig beïnvloed nie. 'n Aansienlike hoeveelheid interne afval (21,8 tot 22,5% van die intakte koue karkasgewig) kan potensieel as lae-koste proteïenbron benut word.

Al die fisiese parameters (pH_u, drupverlies, kookverlies, skeursterkte en kleurkoördinate) het onderskeidelik tussen die spiere en seisoene verskil, behalwe vir die CIE a * en chroma. Die finale pH (pH_u) van al die spiere het binne die biologiese normale waardes geval en vlakte zebra vleis is nie as donker, ferm en droë vleis geklassifiseer nie. Die meeste fisiese metings (pH_u, drupverlies, kookverlies en kleurkoördinate) was vergelykbaar met waardes wat kenmerkend is vir rooivleis, met die uitsondering van die hoë skeursterkte waardes vir monsters wat in beide seisoene geoes is. Vleismonsters van die winter-oesdiere was geklassifiseer as intermediêr in terme van taaiheid, in vergelyke met die somer-oesmonsters wat as taai geklassifiseer is. Die ST en SS van die diere wat in die winter geoes is en die IS van die diere wat in die somer geoes is, was intermediêr in terme van taaiheid. Die res van die onderskeie spiere wat van albei groepe verkry is, het skeursterktewaardes wat verteenwoordigend is van taai vleis, gehad. Die CIE-kleur van die vleismonsters wat van beide groepe versamel is, stem ooreen met die intermediêre waardes wat met wildsvleis assosieer word.

Die geselekteerde spiere het aansienlik verskil ten opsigte van vog-, proteïen- en binnespierse vetinhoud. Die spierproteïeninhoud was die enigste komponent wat beïnvloed is deur die seisoen van oes, met monsters wat deur die winter geoes is, wat 'n hoër proteïeninhoud in vergelyking met die somer-oesmonsters (21,8 ± 0,18 g / 100 g teenoor 20,7 ± 0,12 g / 100 g) gehad het. Seisoen-spierinteraksies is gevind vir die binnespierse vet- en asinhoud, terwyl sterk negatiewe korrelasies vir vog en proteïen gevind is. Die primêre makro- en mikro-minerale teenwoordig in die LTL, SM, BF, lewer en rib was kalium, fosfor, natrium en magnesium, tesame met yster, sink, koper, selenium (behalwe in die rib), mangaan en strontium. Die spiertipe het die natrium-, yster-, koper-, mangaan- en strontiumvlakke beïnvloed. Die beduidende verskille vir die proksimale- en mineraalsamestelling wat

by elk van die hoofeffekte waargeneem is, was marginaal en daarom is dit te betwyfel of dit van biologiese belang is in terme van verbruikersgesondheid.

Die sensoriese profiel van die vlaktesebravleis word gekenmerk deur ‘n wildagtige, beesagtige, soetgeassosieerde en kruidagtige aroma en geur. Die LTL-, SM- en BF-spiere het betekenisvol verskil ten opsigte van sensoriese profiel en vetsuurinhoud, met die BF wat 'n verskil het van die LTL en SM in terme van sensoriese profiel sowel as vetsuurinhoud.

'n Verouderingsproef het bepaal wat die optimale verouderingstydperk sal wees wat nodig is om die maksimum vleis sagtheid vir die LTL-, SM- en BF-spiere te bereik. 'n Verbeterde oppervlakkleur en 'n maksimum sagtheid is behaal op onderskeidelik 14 dae en 20 dae nadoods vir somer-oesmonsters. Die kleurstabiliteit van die spiere was goed, aangesien geen visuele verkleuring tot en met dag 32 van veroudering waargeneem is nie.

Bevindinge uit hierdie studie sal sinvol bydra tot die vasstel van die vleisproduksiepotensiaal van vlaktesebra onder beheerde boerderyomstandighede, om sodoende tot voedselsekerheid in Suid-Afrika by te dra.

ACKNOWLEDGEMENTS

I would like to express my sincerest appreciation towards the following individuals and institutions for their help, support, and contribution in the successful completion of this research project.

My Supervisor, Prof L. C. Hoffman, for believing in me and giving me the opportunity to do this research project. Thank you for enabling me to be on this amazing journey of self-discovery and it was always a true honour being associated with you. I will take the principles that you have taught me and know that it will serve me well in all my future endeavours. Thank you for teaching the most important lesson of all: Carpe Diem!

My Co-Supervisor, Dr H. Lambrechts, I will always be thankful for the valuable insights that you have delivered in shaping me as a researcher and for challenging me beyond my own capabilities and limitations.

My beloved husband, Connie Myburgh. Thank you for the endless encouragement, sometimes tough love, and never-ending support. Without your continual motivation, unconditional love and plethora of wisdom, the successful completion of this project would not be possible. Thank you for always being my sounding board, a listening ear on difficult days and for guiding me in an effortless manner. I am deeply grateful and proud that you stood by me through the long nights and for becoming an expert in shear force and all things meat related with me! A special thank you is also extended to our dogs, Bessie and Vinkie for being my partners in crime on the loneliest days of writing at home.

My best friend, Karla Pretorius. Thank you for accepting me as your friend from day one and for all the followed never ending laughs, spontaneous adventures, and lifelong memories. Thank you for always being my number one supporter in everything I do and for believing in my ability to overcome anything that stood in my way. Without your continuous encouragement and “good chat, good chat” moments my project would not have been possible!

All my other fellow postgraduate students and friends at the Department of Animal Sciences: Raoul du Toit, I deeply appreciate all the help on hunting trips and every coffee at the department which got met through difficult days. A special thank you goes towards **Andrea Ferreira** for believing in my capabilities and for being there with words of encouragement. Without the motivation and support of **Tenisha Roos** and **Anél du Plessis** I would not have been able to finish this thesis**. Bianca Silberbauer** and **Carmen Malan**, thank you for all the late nights, being my Stellenbosch family and what I needed at the most difficult time in the completion of this research project.

The Animal Sciences Department and technical staff, thank you for your continuous friendliness and support. **Adele Smith-Carstens**, thank you for always walking the extra mile to ensure everything runs smoothly and for being a listening ear when I needed one. **Lisa Uys**, thank you for all the

arrangements, orders and always being available for anything else that I needed. **Jonas Christiaan**, thank you for all the assistance in completing practical work in the meat lab. A special thank you also goes towards **Tannie Beverley Ellis**, **Michael Mlambo** and **Janine Booyse** for assisting me with proximate analysis

The Food Science Department, in particular the **Sensory Department** and staff for assisting me with sensory trials. My deepest appreciation also goes towards **Dr J. Marais** for being there whenever I had questions, needed advice, or sought reassurance.

Dirk Giliomee and **Elandsberg Nature Reserve** and the **Quagga project members**, for the use of your facilities to conduct research trials. Without the use of your farms and animals and the assistance from you, the research trials would not be possible.

Prof M. Kidd from the Centre for Statistical Consultation for performing statistical analysis on the data used in this thesis. Thank you for all your patience and willingness to continually assist me.

The South African Research Chairs Initiative (SARChI) funding as administered through the National Research Foundation (NRF). I wish to acknowledge the financial assistance of the NRF towards this study. Views and opinions expressed within this thesis are however those of the authors and not endorsed by the NRF.

My father (**Mark Henn**), mother (**Charmain Henn**) and brother (**Mark Henn**) for enabling and continually supporting my studies throughout my academic career. My deepest appreciation and gratitude also extend to the rest of my family (**Frikkie Myburgh, Anzel Myburgh, Stephan Myburgh and Retha Myburgh**). Thank you for all the interest in my project and being there throughout my studies to help and support me in whichever way I needed it.

Lastly, without our Heavenly Father I would not where I am today. Through His mercy alone I had the blessing of this opportunity. All recognition goes towards Him for surrounding me with these amazing people who supported, motivated, and loved me throughout the completion of my thesis. All honour, recognition and grace go to Him for enabling me to successfully complete this research project.

ABBREVIATIONS

Abbreviation	Expansion
◦C	Degree Celsius
%	Percentage
AI	Adequate Intake
ANOVA	Analysis of Variance
BF	*Biceps femoris* muscle
bw	Body weight
CEC	Commission of the European Communities
CIE	International Commission on Illumination
cm	Centimetre
DAFF	Department of Agriculture, Forestry and Fisheries
DFD	Dark, firm, dry
DNA	Deoxyribonucleic acid
DSA	Descriptive sensory analysis
EFSA	European Food Safety Authority
EU	European Union
FAME	Fatty acid methyl esters
FAO	Food and Agriculture Organization of the United Nations
FES	Free extensive system
FMD	Foot and mouth disease
g	Gram
GIT	Gastro-intestinal tract
ha	Hectare
IMF	Intramuscular fat
IPCS	International Programme on Chemical Safety
IUCN	International Union for Conservation of Nature
IS	*Infraspinatus* muscle
JECFA	Joint Expert Committee on Food Additives
kg	Kilogram
KNP	Kruger National Park
LAU	Large animal unit
LDL	Low-density cholesterol
LSMeans	Least square means
LL	*Longissimus lumborum* muscle
LOD	Limit of detection
LT	*Longissimus thoracis* muscle
LTL	*Longissimus thoracis et lumborum* muscle

Abbreviation	Expansion
M	Metre
mg	Milligram
mm	Millimetre
MUFA	Monounsaturated fatty acids
N	Newton
n	Number
nd	Not detected
n6:n3	Omega-6 to omega-3 ratio
OIE	World Organisation for Animal Health
pH_u	Ultimate pH
PM	*Psoas major* muscle
PTWI	Provisional Tolerable Weekly Intake
PUFA	Polyunsaturated fatty acids
PUFA:SFA	Polyunsaturated to saturated fatty acid ratio
r	Pearson's correlation coefficient
RA	*Rectus abdomini* muscle
RDA	Recommended daily allowance
RF	*Rectus femoris* muscle
SANparks	South African National Parks
SES	Semi-extensive system
SFA	Saturated fatty acids
SM	*Semimembranosus* muscle
SNP	Serengeti National Park
SS	*Supraspinatus* muscle
ST	*Semitendinosus* muscle
TB	*Triceps brachii* muscle
TDI	Total daily intake
TWI	Total weekly intake
v/v	Volume to volume ratio
VPN	The National Directorate Veterinary Public Health
WE	Western Europe
WHC	Water-holding capacity
WHO	World Health Organization
WBSF	Warner-Bratzler shear force
µl	Microliter

NOTES

This thesis is presented in the format prescribed by the Department of Animal Sciences, Stellenbosch University. The language, style and referencing format used are in accordance to the requirements of the journal of Meat Science. This thesis represents a compilation of manuscripts where each chapter is an individual entity and some repetition between chapters has, therefore, been unavoidable.

TABLE OF CONTENTS

DECLARATION i
SUMMARY ii
OPSOMMING iv
ACKNOWLEDGEMENTS vi
ABBREVIATIONS viii
NOTES x
CHAPTER 1 1
REFERENCES 3
CHAPTER 2 5
2.1 THE SOUTH AFRICAN GAME MEAT INDUSTRY – THEN AND NOW 5
2.2 THE PLAINS ZEBRA (*Equus quagga*) 8
2.2.1 Habitat and ecology 10
2.2.2 Social system and behaviour 11
2.3 MEAT PRODUCTION POTENTIAL OF THE PLAINS ZEBRA 12
2.3.1 Equine carcass characteristics 12
2.3.2 Physical meat quality 20
2.3.3 Chemical meat quality 34
2.4 CONCLUSION 42
2.5 REFERENCES 42
CHAPTER 3 55
3.1 INTRODUCTION 56
3.2 MATERIALS AND METHODS 57
3.2.1 Animals and study location 57
3.2.2 Plains zebra harvesting, dressing, and sampling 58
3.2.3 Statistical Analysis 59
3.3 RESULTS 59
3.3.1 Carcass yields 60
3.3.2 Offal yields 61
3.3.3 Muscle yields 63
3.4 DISCUSSION 65

3.4.1 Carcass yields 65
3.4.2 Offal yields 68
3.4.3 Muscle yields 70
3.5 CONCLUSION 71
3.6 REFERENCES 72
CHAPTER 4 78
4.1 INTRODUCTION 79
4.2 MATERIALS AND METHODS 80
4.2.1 Animals and study location 80
4.2.2 Plains zebra harvesting, dressing, and sampling 81
4.2.3 Physical Analysis 81
4.2.4 Statistical analysis 82
4.3 RESULTS 83
4.4 DISCUSSION 87
4.5 CONCLUSION 93
4.6 REFERENCES 94
CHAPTER 5 99
5.1 INTRODUCTION 100
5.2 MATERIALS AND METHODS 101
5.2.1 Animals and study location 101
5.2.2 Plains zebra harvesting, dressing, and sampling 101
5.2.3 Chemical Analysis 102
5.2.4 Mineral Analysis 102
5.2.5 Statistical Analysis 104
5.3 RESULTS 105
5.3.1 Proximate Analysis 105
5.3.2 Mineral Analysis 108
5.4 DISCUSSION 112
5.4.1 Proximate Analysis 112
5.4.2 Mineral Analysis 116
5.5 CONCLUSION 121
5.6 REFERENCES 121

CHAPTER 6 128
6.1 INTRODUCTION 129
6.2 MATERIALS AND METHODS 130
6.2.1 Animals and study location 130
6.2.2 Plains zebra harvesting, dressing, and sampling 130
6.2.3 Descriptive Sensory Analysis 131
6.2.4 Fatty acid Analysis 132
6.2.5 Statistical Analysis 133
6.3 RESULTS 136
6.3.1 Physical measurements 136
6.3.2 Sensory analysis 136
6.3.3 Fatty acid composition 140
6.4 DISCUSSION 142
6.5 CONCLUSION 147
6.6 REFERENCES 148
CHAPTER 7 153
7.1 INTRODUCTION 154
7.2 MATERIALS AND METHODS 155
7.2.1 Animals and study location 155
7.2.2 Plains zebra harvesting, dressing, and sampling 155
7.2.3 Physical analysis 156
7.2.4 Statistical Analysis 156
7.3 RESULTS 157
7.3.1 Winter group (2017) 157
7.3.2 Summer group (2018) 159
7.4 DISCUSSION 166
7.5 CONCLUSION 170
7.6 REFERENCE 170
CHAPTER 8 174
ADDENDUM I 177

CHAPTER 1

GENERAL INTRODUCTION

South Africa is a developing country with many socio-economic instabilities regarding its vulnerability to food insecurity (Altman, Hart, & Jacobs, 2009). The effect of climate change and unstable agricultural markets on food security in South Africa can be challenged by the adaptive and resilient capacity of indigenous efforts such as utilising neglected and underutilised species (FAO, 2018). The adverse effects of climate change and market instabilities on pastoral farming in South Africa has resulted in farmers searching for strategies that will assist them in coping with the challenges and financial constraints associated with climate change. A potential adaptive strategy is to convert either to production systems that include mixed livestock-game farming or game farming only, as some domestic livestock systems such as beef production is highly vulnerable to the extreme temperature or precipitation fluctuations currently experienced in various parts of South Africa (Otieno & Muchapondwa, 2016).

Game species are well adapted to arid and semi-arid African environments and are less affected by bush encroachment that result from land degradation and overgrazing (Oberem & Oberem, 2016). The inclusion of game animals, whether using a grazer, browser, and mixed feeder strategy, allows for the optimal use of the vegetation and higher stocking densities when mixed in a holistic system (Cooper & Van der Merwe, 2014). The high resilience of game species in conjunction with their low carbon footprint reduces not only the environmental impact but also the input cost as they are not reliant on grain-based feeds (Cooper & Van der Merwe, 2014) or as labour-intensive compared to cattle farming (Taylor, Lindsey, & Davies-Mostert, 2016). With this in mind, the conversion to game farming is further fuelled by the high commercial value of game animals as well as game farming owning its place as the sixth-largest contributor to the agricultural industry (Carruthers, 2008; Otieno & Muchapondwa, 2016). In comparison to traditional livestock farming, game farming generated revenue through many sources which are primarily through trophy and recreational hunting, breeding and live sales, ecotourism, and lastly meat production as a by-product (Child, Musengezi, Parent, & Child, 2012; Lindsey, 2011). These revenue sources known as the four pillars are primarily based on the “shuffling” of surplus game animals between farms and/or are either sold through hunting or auction purposes and when necessary culled for meat production to maintain population size and also to ensure breeding herds with high ranking animals (Taylor et al., 2016).

Recently, a decline in live game sales has been observed as the scarcity of high-valued species reduced due to the increased number of game farms and animals available for auction. The latter resulted in game prices decreasing from live game prices to hunting prices (Groenewald, 2019). The decline in selling prices has resulted in game farmers culling not only their conventional meat-producing game species to earn income from meat sales, but also their more unconventional game species such as the plains zebra. Nonetheless, the increasing number of game farms still advocate breeding of game and thus the production of protein to meet food demands in an environmentally friendly and sustainable manner.

Established game farms in South Africa are mainly located near rural communities that are characterised by low household incomes. Even though game meat is currently marketed as a niche product, approximately 70 % of the carcass consists of cheaper cuts including bone, meat, and red offal (liver and kidneys), which all is available to and affordable for lower-income households (Taylor et al., 2016). However, among consumers, there is a shared perception that game meat is dry and tough and needs to be addressed. Therefore, in order for game meat to directly contribute to the red meat supply of South Africa, standard operating methods need to apply to ensure products of high and consistent quality (Hutchison, Mulley, Wiklund, & Flesch, 2010; North & Hoffman, 2015).

The plains zebra is an excellent example of an unconventional meat-producing game species that is not consumed by most of South Africa's meat consumers (Hack, East, Rubenstein, & Gray, 2002), which can be ascribed to consumers not being knowledgeable on the production potential of plains zebra, as well as the sensory profile of meat form this species. Young adult plains zebra stallions that have attained sexual maturity that have not yet formed their own harems (i.e. occupy a low rank in bachelor groups) are typically removed from herds. This particular category of plains zebra presents an opportunity to be utilised for meat production purposes, while the relatively young age of the animals holds promise for the product in terms of meat quality traits. The plains zebra has a highly adaptable nature with an immunity to the livestock threatening foot and mouth disease as they are odd-toed. The spreading of this disease in South Africa leads to the prohibition of game meat exports to the European Union but does not apply to the export of zebra meat. The scarcity of plains zebra meat in the local market can be countered and the export of higher meat quantities can be promoted if information regarding the meat production potential and the quality thereof is available. Consequently, for plains zebra meat to compete with existing meat products, information on the carcass composition and yield, physical quality, nutritional quality, and the sensorial quality needs to be generated through reliable scientific research.

Research question, aims and objectives

The aim of this research was to determine baseline data for carcass yields, physical quality, mineral composition, sensory profile, and the optimum post-mortem ageing period for maximum meat tenderness of plains zebra stallions. The research also aimed to determine the effect of muscle type and season of harvest on these parameters as well as on the chemical and fatty acid composition of plains zebra meat harvested in the Western Cape Province, South Africa.

The objectives of this study were:

1. Evaluate available literature on the zebra, horse, and donkey species to determine the suitability of the plains zebra as a meat source as data is limited. (Chapter 2)
2. Determine the carcass yields of the plains zebra as influenced by the harvesting season –winter (June 2017) and summer (January 2018) (Chapter 3).
3. Determine the physical meat quality parameters of the plains zebra as influenced by muscle type and season of harvest (Chapter 4).
4. Determine the influence of muscle type and harvesting season on the chemical composition of plains zebra meat (Chapter 5)

5. Determine the mineral composition as influenced by muscle type (*Longissimus thoracis et lumborum*/LTL, *semimembranosus*/SM, and *biceps femoris*/BF) of plains zebra meat as well as the mineral composition in the liver and rib (Chapter 5)
6. Determine the sensory profile of the LTL, SM and BF plains zebra muscles through a descriptive sensory analysis (DSA) (Chapter 6).
7. Determine the fatty acid composition of the different muscles (LTL, SM and BF) (Chapter 6)
8. Determine the influence of post-mortem ageing on the physical meat quality of vacuum packed LTL, SM and BF steaks derived from both the winter and summer season for a 24-day and 32-day ageing trial, respectively, in order to determine the optimum ageing period for optimum meat tenderness (Chapter 7).

REFERENCES

Altman, M., Hart, T. G. B., & Jacobs, P. T. (2009). Household food security status in South Africa. *Agrekon*, *48*(4), 345–361. https://doi.org/10.1080/03031853.2009.9523831

Carruthers, J. (2008). “Wilding the farm or farming the wild”? The evolution of scientific game ranching in South Africa from the 1960s to the present. *Transactions of the Royal Society of South Africa*, *63*(2), 160–181. https://doi.org/10.1080/00359190809519220

Child, B. A., Musengezi, J., Parent, G. D., & Child, G. F. T. (2012). The economics and institutional economics of wildlife on private land in Africa. *Pastoralism*, *2*(1), 1–32. https://doi.org/10.1186/2041-7136-2-18

Cooper, S. M., & Van der Merwe, M. (2014). Game ranching for meat production in marginal African agricultural lands. *Journal of Arid Land Studies*, *24*(1), 249–252.

FAO, IFAD, UNICEF, WFP, & WHO. (2018). *The State of Food Security and Nutrition in the World 2018. Building climate resilience for food security and nutrition. Rome, FAO. Licence: CC BY-NC-SA 3.0 IGO*. https://doi.org/10.1093/cjres/rst006

Groenewald, Y. (2019). The exotic game market goes bang. Retrieved November 27, 2019, from The M&G Online website: https://mg.co.za/article/2019-01-11-00-the-exotic-game-market-goes-bang

Hack, M. A., East, R., Rubenstein, D. I., & Gray, E. (2002). Status and action plan for the plains zebra (*Equus burchellii*). In P. D. Moehlman (Ed.), *Equids: Zebras, Asses and Horses. Status Survey and Conservation Action Plan* (pp. 43–60). IUCN/SSC Equid Specialist Group. IUCN, Gland, Switzerland and Cambridge UK.

Hutchison, C. L., Mulley, R. C., Wiklund, E., & Flesch, J. S. (2010). Consumer evaluation of venison sensory quality: Effects of sex, body condition score and carcase suspension method. *Meat Science*, *86*(2), 311–316. https://doi.org/10.1016/j.meatsci.2010.04.031

Lindsey, P. (2011). An analysis of game meat production and wildlife-based land uses on freehold land in Namibia: Links with food security.*TRAFFIC East/Southern Africa*, Harare, Zimbabwe.

North, M. K., & Hoffman, L. C. (2015). Changes in springbok (*Antidorcas marsupialis*) *Longissimus*

thoracis et lumborum muscle during conditioning as assessed by a trained sensory panel. *Meat Science*, *108*, 1–8. https://doi.org/10.1016/j.meatsci.2015.05.004

Oberem, P., & Oberem, P. (2016). Species accounts. In *The New Game Rancher*. Biza Publications.

Otieno, J., & Muchapondwa, E. (2016). Agriculture and adaptation to climate change : The role of wildlife ranching in South Africa. *Economic Research Southern Africa*, 1–28.

Taylor, A., Lindsey, P., & Davies-Mostert, H. (2016). An assessment of the economic social and conservation value of the wildlife ranching industry and its potential to support the green economy in South Africa. The Endagered Wildlife Trust. Johannesburg, South Africa. Retrieved from https://www.sagreenfund.org.za/wordpress/wp-content/uploads/2016/04/EWT-RESEARCH-REPORT.pdf

CHAPTER 2

LITERATURE REVIEW

2.1 THE SOUTH AFRICAN GAME MEAT INDUSTRY – THEN AND NOW

At the onset of the 20th century, the game industry was recognised as a market and was yet to secure its valuable position in the agricultural sector in South Africa as it stands today. During that time, at least one species and two subspecies of mammals were extinct, due to uncontrolled hunting in the late 1800s. These species and subspecies respectively include the blue buck (*Hippotragus leucophaeus*), the quagga (*Equus quagga quagga*) and the Cape lion (*Panthera leo melanochaita*), with the black wildebeest (*Connochaetes gnou)*, bontebok (*Damaliscus pygargus pygargus*), and the Cape mountain zebra (*Equus zebra*) being nearly extinct (Meester, 1954). Game animals were classified as *res nullius* (i.e. without ownership) and was believed to be a carrier of diseases prone to infect livestock, thus having the potential to negatively influence livestock production (Carruthers, 2008). Game animals were therefore hunted as a disease control measurement, leading to overhunting and thus a rapid reduction in the total number of game animals (Carruthers, 2008; Van der Merwe, 2014).

Game meat started to receive interest locally as a protein source by the 1950s after World War II, as meat was a rationed commodity. Meat rationing resulted in an increased interest in game meat, and collectively, with the increase in total landowners (which were by the time protected by trespassing laws), game numbers started to stabilise by the late 1950s (Carruthers, 2008). Stabilisation of game numbers was aided by the awareness created by the International Union for Conservation of Nature (IUCN) conference about game farming (wildlife ranching) held in Belgian Congo in 1953, as well as the development of the South African Hunting and Game Conservation Association into a national association in 1957 (Hoven, 2015). During the 1960s, the progressive increase in the size of the game industry resulted in an increased interest by scientists in aspect pertaining to game farming (Carruthers, 2008), which resulted in the establishment of the South African Journal of Wildlife Research in 1970 (Hoven, 2015). The first game auction in South Africa was held in 1965 at Tshipise in the then Transvaal Province (Hoven, 2015). The growth of the market was further encouraged by the permitting of private ownership of game animals, as the Game Theft Act (105 of 1991) that was formulated and implemented, afforded the game farming industry with an opportunity to rapidly develop and grow (Van der Merwe, Saayman, & Krugell, 2004). This opportunity was further supported by the 1994 Constitution of South Africa, which embodied the notion of sustainable use of the environment (Oberem, 2012; Taylor, Lindsey, & Davies-Mostert, 2016). At this point, the main drivers of the industry were hunting and ecotourism, which was later accompanied by the breeding and live sales of high-value game species in the early 2000s.

The expansion of the market led to the identification of four interlinked pillars, i.e. hunting for trophy purposes and production of biltong, ecotourism, breeding and live sales, and meat production (Van der Merwe et al., 2004; Van Schalkwyk & Hoffman, 2010). According to Hoven (2015), private

game reserves or game farms in South Africa are managed to gain profit in either consumptive or non-consumptive categories of the four mentioned pillars. The Gross Domestic Product (GDP) of the game sector is currently classified as part of the tourism and agricultural sector, as it contributes to both. The tourism industry in total contributed 2.9% to the GDP of South Africa in 2016, which was more than agriculture itself during that period (Stats SA, 2018). Ecotourism consists out of the non-consumptive aspect of game farming (i.e. wildlife watching) and plays a vital role in supporting wildlife and conservation associations, as well as local communities in South Africa as tourists are interested in the "African experience" and in seeing species with unique features (horn types, colour variations and body size (Van der Merwe, 2014).

The establishment of intensive and semi-intensive game breeding operations to breed high-value species and colour variants is of great interest to game farmers as it is profitable in terms of live sales, trophy hunting, biltong hunting, ecotourism, and meat production. The intensification of breeding certain game species for live sales and trophy hunting purposes has resulted in a high turnover in these species, and consequently led to the production of disqualifying surplus animals (Taylor et al., 2016). The maintenance of surplus animals are not self-sustainable as their numbers are not controlled through natural predators, thus regular culling is required to control surplus animal numbers to adhere to the natural carrying capacity of the systems in question (Taylor et al., 2016). Using culling to control surplus animals has resulted in more readily available game meat for consumption purposes. For example, meat from ungulates hunted for trophy purposes are generally seen as a secondary product and is usually not taken by the trophy hunter. The meat can then be sold by the farmer to the local community to avoid wastage (Taylor et al., 2016). Meat resulting from the hunting of certain ungulate species is not preferred by consumers due to the lack of information of preparation of such meat, and little if any awareness about health benefits. Ungulates culled for trophy purposes include odd-toad ungulates such as the zebra and even-toed ungulates such as giraffe and various deer species. Currently, zebra is one of the top 10 species hunted for trophy purposes, alongside springbok, impala (*Aepyceros melampus*), greater kudu (*Tragelaphus strepsiceros*), blesbok (*Damaliscus pygargus phillipsi*), black (*Connocheates gnou*) and blue wildebeest (*Connochaetes taurinus*), warthog (*Phacochoerus africanus*), gemsbok (*Oryx gazella*), and red hartebeest (*Alcelaphus buselaphus caama*) in South Africa (Munzhedzi, 2018). Trophy hunting in South Africa has contributed to the growth of the total number of game reserves, from 10 reserves in the 1960s to an estimate of 11600 in 2015 owning up to 22 million hectares (18% of the land surface; Hoven, 2015). Live sales of game animals and trophy hunting have each contributed R1.7 billion to the economy in 2016 and 2015, respectively. Consumptive (biltong) hunting increased by 35% in the period from 2013 to 2015 and contributed R8.6 billion to the economy in 2015 (Munzhedzi, 2018). The latter had a growth rate of 35% between the year 2013 and 2015 (Munzhedzi, 2018), highlighting the fact that ecotourism, trophy hunting and live sale/breeding cannot promote the growth of the industry alone as it mostly a meat-producing sector (Van Schalkwyk & Hoffman, 2010).

The global demand for animal protein or meat products has increased rapidly over the past decade due to the tremendous increase in the world population. The world population is currently standing on 7.5 billion people and is estimated to reach 9.3 billion people by 2050 (Thomson, 2003).

However, with the higher demand for animal protein, the consumption of plant protein instead of animal protein is a popular and growing global food trend. Consumers are also becoming aware of the health implications of diets containing high levels of animal fats, which are known to contain high levels of saturated fatty acids (SFA) and low levels of polyunsaturated fatty acids (PUFA). Seeing that consumers are becoming aware that the consumption of SFA, found in red meat derived from livestock, are associated with lifestyle diseases creates an opportunity to produce or inform the modern consumer about alternative animal protein sources (Kearney, 2010). However, not only is the modern consumer concerned about these health-related problems but they are also interested in food products being fresh, organic, green and local, which has resulted in the reintroduction of trends such as the farmers' market, slow market and slogans such as "home-grown". South Africa is fortunate to have numerous game species suitable for sustainable culling to produce meat with the trending labels mentioned above as game meat is known to be an organic high protein source rich in iron and low in fat. Unlike most livestock, game animals are generally better adapted to harsh arid environments, parasites, parasites-borne diseases, and toxic plants, making them an ideal farming candidate.

According to the Food and Agriculture Organization (FAO) of the United Nations, Africa is the world's largest producer of game meat, with a yield of 1145920 tonnes in 2016. South Africa contributes 3.5% (40295 tonnes) to Africa's game meat production and ranks eighth in terms of the global exportation of game/venison (2681 tonnes) recorded in 2016. New Zealand is currently the leading venison/game meat exporter, with a yield of 10807 tonnes in the same year. Developed countries in Western Europe (WE) account for most of the total recorded game/venison meat imports in 2016, with a total of 50345 tonnes being imported. The top five importers of WE were Germany (18083 tonnes), Netherland (16383 tonnes), Belgium (6633), France (5956 tonnes), and Italy (4835 tonnes) (FAO, 2019). The low contribution of South Africa towards the supplying and exporting of game meat is a direct result of the game meat exportation embargo set by the European Union (EU) in 2011. The embargo was in effect for three years due to the outbreak and transmission of foot and mouth disease (FMD) in cloven-hoofed ungulates. During this period, the plains zebra (*Equus quagga*), classified as a meat-producing species by the Meat Safety Act no. 40 of 2000, were the only species available for exportation, as it is not susceptible to FMD.

The embargo has led to a significant setback in the game meat industry as South Africa is still struggling to regain its position as the second-largest exporter of game meat in 2009 (FAO, 2019). The Department of Agriculture, Forestry and Fisheries (DAFF) has since the embargo, agreed on regulations between South Africa and the EU to resume exports. The National Directorate Veterinary Public Health (VPN) on the "standard for the registration or re-registration of a game farm for export status" set in 2010 (VPN/05/2010-01) were updated and replaced in 2017 (VPN/05/2017-01) by DAFF in order to prohibit the spreading and exportation of infected game meat. Despite these efforts, the industry is still facing challenges, as there was a confirmed outbreak of FMD in the high surveillance area of the FMD-free zone in the Limpopo Province in cattle during the months of January and November 2019 (Louw-Carstens, 2019). This FMD outbreak has led to a suspension of the FMD-free status without vaccination in cloven-hoofed animals in South Africa by the official World Organisation for Animal Health (OiE, 2019; DAFF, 2019). Outbreaks such as the FMD epidemic result in uncertainties

regarding the production and export of game meat, since the plains zebra are the only meat-producing game species that are not affected by the spreading of FMD.

African horse sickness (AHS) is a seasonal viral disease of equids endemic in South Africa except for the controlled area in the Western Cape Province where only sporadic outbreaks has occurred in the last two decades. Outbreaks of AHS in South Africa has led to a non-tariff barrier to trade live equids to the EU and therefore the preservation of the controlled area in the Western Cape Province is of great importance (Porphyre & Grewar, 2019). Infections in zebra populations act as reservoir for the disease due to its asymptomatic nature in zebras specifically, creating a serious concern for the horse industry (Porphyre & Grewar, 2019; Zientara, Weyer & Lecollinet, 2015). However, due to the small zebra population located in the South African AHS controlled area it has been found to be highly unlikely that the small zebra populations in the Western Cape Province will be able to maintain a persistent infection in and around the AHS controlled area. However, the trading of plains zebras outside and inside the controlled areas needs to be included in the disease control and surveillance planning of South Africa to reduce the risk for AHS outbreaks (Porphyre & Grewar, 2019).

Trading of the plains zebra contributes to all four the pillars as it supports the value chain in terms of wildlife farming, wildlife-associated activities, and wildlife products. Zebras are famous for their unique phenotype, with their meat used especially for the making of salami (Hoffman, Geldenhuys, & Cawthorn, 2016). Commercial and non-commercial hunters, therefore, hunt them for their skins, meat, and trophies (Stears, Shrader, & Castley, 2016). The auction prices of the plains zebra can vary considerably, and prices fetched are determined by colour variation, gestation status, age, and sex. On average a plains zebra stallion is valued at R5500, and mares fetched up to R5000 at auctions held in 2019. However, zebras with unique features or unique colour variations obtain higher prices. For example, during auctions held in 2019, a blue-eyed golden zebra stallion fetched R750 000, and golden zebra between R75 000 – R150 000 (Wildswinkel, 2017)

When the farming of zebras for meat purposes is considered, limited information is available on the meat quality and production potential despite its high export potential. Therefore, it is of importance to determine the carcass yield and meat quality of the plains zebra as well as the factors, ante-mortem and post-mortem, that influences these attributes. As limited information is available on this equine species, the meat production potential in terms of other equines such as horse and donkey will be reviewed in this chapter.

2.2 THE PLAINS ZEBRA (*Equus quagga*)

The plains zebra is a large-bodied herbivorous odd-toad ungulate that belongs to the Genus: *Equus* and Family: *Equidae*. The plains zebra has distinct black and white stripes with shadow stripes superimposed on the white stripes of particularly the hindquarters. The plains zebra has an average shoulder height of 1.3 m, with an average mature live weight between 290-340 kg (Stuart, 2015). This wild African equid has six morphologically distinct subspecies with small genetic differentiation (Groves & Bell, 2004) based on differences in small cranial and tooth characteristics, body size, stripe width and stripe pattern (Hack, East, Rubenstein, & Gray, 2002). There is, however, controversy surrounding the use of the scientific name of the plains zebra species and its subspecies. Throughout this thesis, the

taxonomy system described by Groves & Bell (2004) will be used to describe the species and subspecies.

According to Groves & Bell (2004), the six subspecies of the plains zebra include the *Equus quagga boehmi* (Grant's zebra), *E. q. crawshaii* (Crawshay's zebra), *E. q. borensis* (Maneless zebra), *E. q. chapmani* (Chapman's zebra), *E. q. burchelli* (Burchell's zebra) and *E. q. quagga* (Cape quagga). The extinct Cape quagga has been determined through comprehensive DNA sequencing of museum specimens to be a colour variant of the plains zebra. However, earlier speculations have led to the launching of the Quagga Project in 1987 to restore the pelage characteristics of the extinct Quagga through selectively breeding a panel of selected plains zebras (Harley, Knight, Lardner, Wooding, & Gregor, 2009). Currently, the project is in its fourth generation of animals that adhere to the description of and are assigned the designation of the "Rau quagga", as they resemble a reduced striping pattern as observed in some museum quagga specimens (Harley, Lardner, Gregor, Wooding, & Knight, 2018).

The plains zebra is one of the most widely and abundantly distributed among most of the grazing animals found in South Africa (Stears et al., 2016). The population size of the plains zebra is challenging to calculate as they occur on private land, and in South African National Parks (SANParks) with the latter that have limited or out-dated data. Despite this, an estimation in 2002 noted the total population size of plains zebra to be around 663 212 plains zebras in South Africa (Hack et al., 2002). More recent estimates of the plains zebras in parks by Ferreira, Gaylard, Greaver, Hayes, & Cowell (2016) are presented in Table 2.1. The Endangered Wildlife Trust estimated that a minimum of 59 204 plains zebras occurred in 803 protected areas between 2010 and 2015 (Endangered Wildlife Trust unpubl. data cited by Stears et al., 2016).

Table 2.1 Estimates for plains zebra numbers in South African parks recorded between 2014 and 2016 (adapted from Ferreira et al., 2016).

Parks	Region	Number	Year of count	Count/Sampling method	Trend
Golden Gate	Arid region	1592	2016	Total counts	Increasing
Mokala	Arid region	358	2016	Total counts	Increasing
Addo – Main/Colchester	Frontier region	477	2016	Total counts	Increasing
Addo-Kuzuko	Frontier region	20	2016	Total counts	Decreasing
Addo- Nyathi	Frontier region	449	2016	Total counts	Increasing
Karoo	Frontier region	10-15	2014	Total counts	Decreasing
Mountain Zebra Park	Frontier region	0	2014	Total counts	Decreasing
Kruger National Park	Northern region	19850-30020	2014	Transect using distance sampling	Non-directional
Marakele	Northern region	966-1125	2015	Total counts	Increasing
Mapungubwe	Northern region	161	2014	Sampling surveys using fixed width transect	Unknown

According to the regional and national Red List status updated respectively in 2004 and 2016, the plains zebra is of least concern, with no major factors threating the existence of the species, making is suitable for production and harvesting purposes. However, to ensure that it is viable game species to farm with, overhunting and habitat degradation due to cattle encroachment needs to be managed optimally to maintain population numbers within stocking density guidelines, and thus ensure the sustainable use of available forage.

There are currently two subspecies of mountain zebra found in Southern Africa: The Cape Mountain zebra (*Equus zebra zebra*) and the Hartmann's Mountain zebra (*Equus zebra hartmannae*). The status of the Cape Mountain zebra is currently improving from being classified as endangered in 2007 to vulnerable in 2008, and finally as of least concern in 2016 (Hrabar et al., 2016). The Hartmann's Mountain zebra was classified as endangered in 2004, and due to the small population increase classified as vulnerable in 2016 (Novellie, King, Muntifering, Uiseb, & Child, 2016). Consequently, both subspecies do not present an opportunity to be produced and harvested for the game meat industry. The Hartmann's Mountain Zebra is currently restricted to certain areas in South Africa due to the Threatened or Protected Species Regulations (Taylor et al., 2016), which has resulted in a decline in its commercial value as farms located in the restrictive area is saturated with this particular species. As a result, they are harvested and replaced by higher valued wildlife species, threatening the Hartmann's Mountain zebra population even more (Taylor et al., 2016).

2.2.1 Habitat and ecology

The plains zebra is a bulk non-selective feeder with the ability to graze a variety of grass species of both poor and good quality (Estes, 2012; Oberem & Oberem, 2016). This ability allows them to have wide home ranges as they are easily adaptable to marginal lands such as the thicket biome and the eastern granite sandveld in the Kruger National Park (KNP; Furstenburgh, 2009), with grass heights between 100 - 350mm (Bothma, 2011). Plains zebras commonly roam on grassland, savannah woodlands, sweetveld and mixed veld. However, they tend to avoid deserts, dense forests, and wetlands (Estes, 2012; Oberem & Oberem, 2016).

Ungulates such as the plains zebra are hindgut fermenters, with the hindgut that contains symbiotic microorganisms which enable them to digest cellulose found in low-quality feeds (Duncan, 1992; Oberem & Oberem, 2016). They are thus able to effectively digest feed that is high in fibre and low in protein, by taking in an abundant amount (Furstenburgh, 2009). The plains zebra has an average intake of 7.8 kg dry forage per animal per day, which is double the volume that is reported for blue wildebeest. Water consumption is recorded to be on average of 7.8 litres of water per animal per day. The diet of the plains zebra consists of 93 % grass, 5 % browse and 2 % fruits (Bothma, 2011). Unlike antelope, zebras have a fully functional upper set of incisors and can easily bite off medium height grass, without uprooting grass, causing less damage to the veld (Furstenburgh, 2009). A study in the Nechisar plains indicated that the plains zebra demonstrated a high preference for *Themenda triadra* (red grass) and *Lintonia nutans* and favoured other grasses such as *Setaria sphacelata* (African bristlegrass), a species of muraina grass called *Ischaemum afrum, Chrysopogon aucheri* and *Cenchrus ciliaris* (dhaman grass) (Doku, Bekele, & Balakrishnan, 2007). The plains zebra has also shown a

preference for *Heteropogon*, *Cymbopogon, Stipagrostis, Hyparhennia* and *Aristida* species (Kingdon, 2015). They can also digest the devil's thorn (*Tribilus teristris*), which is toxic to sheep (Bothma, 2011).

The plains zebra enters grasslands in the early stages of flush growth, before other selective grazers, removing the older growth layer. This practice initiates new growth and enhances the overall quality of the vegetation for grazers such as wildebeest and antelope (Estes, 2012; Oberem & Oberem, 2016). The plains zebra is a mobile migrate species with an unstable home range. The plains zebra migrates due to the onset of rain, veld fires and food- and water availability (Smuts, 1975a). Given their ability to digest low-quality feed, they usually are also the first animal to graze on burnt veld (Furstenburgh, 2009). In the Serengeti, they migrate up to 200 km and in Namibia up to 160 km. Migration in the KNP is due to the availability of a distinct summer and winter grazing area. The disappearance of surface water typically drives the migration of the plains zebra from the winter grazing area to the summer grazing area. They favour areas in the KNP with biomass of approximately 1300 kg dry plant matter per hectare, whereas in Limpopo Province they inhabit areas with a moderate grass layer with 150 trees per hectare (Bothma, 2011).

2.2.2 Social system and behaviour

To commercially produce meat derived from wild species, such as the plains zebra, knowledge about their unique social behaviour and welfare level are essential to ensure sustainable and profitable production. By understanding the behavioural patterns, preventative measures can be put in place to minimise potential stress-causing situations, thereby ultimately optimising production outcomes (Cam, Kirikci, & Garipoglu, 2018). Plains zebras typically form several harems and bachelor groups, that all contribute to one sizeable social organisation, consisting out of hundreds of individuals. Harems are stable breeding groups consisting out of one sexually matured male (also referred to as a stallion) between 8-12 years of age and five to six unrelated females (also referred to as mares), with their offspring forming an undisturbed family (Klingel, 1972; Rubsentein, 2018). Studies in Danish and Dutch zoos indicated that the social organisation and behaviour of captive plains zebras are similar to the behaviour of wild plains zebras (Andersen, 1992; Schilder & Boer, 1987).

The hierarchy of the females is characterised by four levels, and can be established by fighting (Furstenburgh, 2009). In some studies, however, it was reported that, the hierarchy of females appeared to be established by age (Andersen, 1992; Pluháček & Bartoš, 2005) or the residency time in captive groups, as foaling does not change the ranking of mares (Pluháček & Bartoš, 2005). The first level is occupied by the dominant alpha mare of eight years and older, followed by the second level of two to three beta mares between the ages of five and eight years. The third level is represented by two to five mares between three and five years, and lastly the fourth level consists out of numerous sub-adults of both sexes generally under the age of three years (Furstenburgh, 2009). Therefore, it can be recognised that plains zebras are not territorial but rather family bonded (Klingel, 1969).

Bachelor groups are formed when solitary males join, reaching up to 50 individuals per group. These males consist of young stallions detached from their natal groups, and old unfit stallions that lost their harems due to rivalry (Estes, 2012; Hack et al., 2002; Klingel, 1969; Oberem & Oberem, 2016). Young stallions, regardless of their maturity, leave their natal harems between the ages of one and four

and half years of age. The reason for leaving is either due to their mother bonding with her new foal or to the lack of similar-aged males in the family, thus finding playmates in nearby bachelor groups (Klingel, 1969). Plains zebra stallions are sexually mature at the age of four to four and a half years but will only leave their bachelor group at the age of five years when they are strong enough to establish their own harem (Klingel, 1969). Low ranking stallions in these groups have the potential to be harvested for meat production, and through herd management excluded from breeding herds. These stallions tend to be young, and form harems by abducting fillies at the age of 13 -15 months during their first oestrus. The fillies will come into oestrus in monthly intervals for an estimate of five days for as long as a year, before they conceive. During this time, stallions will need to fight off other young stallions and probably lose their fillies to other males. Plains zebra mares are sexually matured at the age of two and a half years and will have their first foal at the age of three and a half years (Klingel, 1969). The gestational period of a plains zebra mare is 12 months, thus foaling year-round and peaking during the rainy season (Oberem & Oberem, 2016).

Younger stallions can replace stallions with harems due to death or being too old or unfit for future breeding (Klingel, 1972). Stallions in established families vigorously communicate with stallions passing by, indicating their capability of defending their harem, and therefore will seldom be challenged. A lack of communication will indicate that the stallion is ready to be replaced or is unfit, and such stallions will then be replaced by young stallions shadowing the harem, and that without a fight, will displace the old or unfit stallion from the group. The group of mares will stay intact and will gradually accept the new male (Estes, 2012). The new stallion will force out colts that are not part of his offspring and mares, regardless of their lactating status, will go into oestrus again. Lactating mares will stop weaning their foals, given that foals still suckling will typically be killed by the new stallion (Furstenburgh, 2009). As mentioned, these young bachelor stallions are ideal to be culled for meat although the meat quality of zebra is sparsely reported on, with Hoffman et al. (2016) being one of the few reports giving some necessary information on the meat quality of this equid species.

2.3 MEAT PRODUCTION POTENTIAL OF THE PLAINS ZEBRA

2.3.1 Equine carcass characteristics

The economic value of a carcass is determined by the quantity and quality of the resulting meat when utilising carcass weight and amount of marketable meat (Swatland, 1994). Growth and development of an animal upon slaughter form the basis of meat production, and thus, the distribution of carcass tissue is a significant factor used to determine carcass quality (Mahgoub & Lu, 1998). Carcass tissue distribution measurements are interrelated with the sensorial quality of meat which indirectly influences the quantity of meat cuts that will be acceptable according to consumer preferences. The same criteria used for carcass evaluation for domestic livestock and in equine species, apply for various game species such as zebra (Hoffman, 2000). Carcass yield, which includes undressed and dressed carcass weights, is an essential measurement as game animals are primarily sold per kg in South Africa (Hoffman & Wiklund, 2006).

2.3.1.1 Carcass yield

Information on the carcass yields of the plains zebra is limited to two studies conducted during the dry season in Kenya (Onyango, Izumimoto & Kutima, 1998) and South Africa (Hoffman et al., 2016). The carcass weight of 20 plains zebras recorded during the winter season in a summer rainfall region (i.e. Bushveld, Limpopo Province, South Africa), ranged between 106.0 kg to 190.6 kg, averaging at 138.2 kg. Onyango et al., (1998) recorded a similar average of 140 kg in plains zebra stallions harvested in Kenya. No literature is available on the variation in plains zebra carcass weight and dressing percentages as influenced by age, slaughter weight, sex, breed, production system or finishing diet, thus warranting further research. Some of these factors have been reported to influence the carcass weight of both game and other equine species and will be discussed.

The slaughter age of an animal can determine the meat yield and quality, as live weight is intrinsically dependent on the species-specific growth rate (Renecker, Renecker, & Mallory, 2005) and physiological maturity, and extrinsically on the harvesting season (Sookhareea, Taylor, Woodford, Dryden, & Shorthose, 1995). Knowledge on the growth curve and feed conversion efficiency in game species is crucial as it determines the maximum meat production potential and also the optimum slaughter age which is of economic importance (Von La Chevallerie, 1970).

As previously mentioned, there is no available literature on the undressed carcass weight of plains zebra (i.e. live body weight) at different ages, of for any of the subspecies. There is however, ample literature available on the live body weight of various horse breeds (De Palo, Maggiolino, Centoducati, & Tateo, 2013; Domínguez, Crecente, Borrajo, Agregán, & Lorenzo, 2015; Franco, García Fontán, Temperan, García Calvo, & Lorenzo, 2010; Juárez et al., 2009; Lanza et al., 2009; Litwińczuk et al., 2008; Lorenzo et al., 2014; Lorenzo, Sarriés, & Franco, 2013; Sarriés & Beriain, 2005; Znamirowska, 2005), and in Martina Franca donkeys specifically (Polidori, Vincenzetti, Cavallucci, & Beghelli, 2008; Polidori, Pucciarelli, Ariani, Polzonetti, & Vincenzetti, 2015). The respective studies reported accurate figures on live body weight in terms of months and years. Most of the studies focused on foals younger than 24 months of age (De Palo et al., 2013; Domínguez et al., 2015; Franco et al., 2010; Juárez et al., 2009; Lanza et al., 2009; Lorenzo et al., 2014; Lorenzo & Pateiro, 2013; Sarriés & Beriain, 2005), with only a few studies focusing on adult horses ranging from 6-12 years of age (Litwińczuk et al., 2008; Znamirowska, 2005). However, many of these studies did not necessarily use age as a research treatment but rather as information regarding the experimental design.

Species/breed-specific growth curves are plotted in relation to the age of an animal and are generally sigmoidal in terms of the growth rate of different tissues, maturity weight, and fat deposition (Swatland, 1994). The growth curve of the Burchell's zebra, a subspecies of the plains zebra, was first described by Smuts (1975b). Smuts (1975b) used the Von Bertalanffy growth equation to determine the theoretical asymptotic live weight, total length, vertebral column length, heart girth, shoulder height, head length, hindfoot length, tail length and ear length of both male and female zebras, based on pooled data over a period of approximately three years from the KNP, South Africa (Smuts, 1974). The theoretical Von Bertalanffy growth equations and the asymptotic ages for live weight, total length, and shoulder height recorded by the author, are presented in Table 2.2. Take note that the scientific name

used by Smuts (1975b), *Equus burchelli antiquorum*, was revised by Groves & Bell (2004) to be *Equus quagga burchelli*. The latter species name will be used when reference is made to the Burchell's zebra.

Table 2.2 Theoretical Von Bertalanffy growth equations, asymptotic ages and calculated weight for the live weight and calculated length for the total body length and shoulder height, for Burchell's zebra (adapted from Smuts, 1975b).

Measurement	n	Unit	Equation	Asymptotic age (years)	Asymptotic weight/length	12 months	18 months
Live weight	161	kg	$W_t = 316.9(1-e^{-0.99(t+0.651)})^3$	11	316.9	165.5	217.0
Total length	250	cm	$l_t = 279.0(1-e^{-1.04(t+0.733)})$	5	279.0	233.3	279.02
Shoulder height	263	cm	$l_t = 134.8(1-e^{-1.28(t+0.854)})$	3	134.8	122.3	128.2

Abbreviations: W_t= weight in kilograms (kg); l_t= length in centimetres (cm), t = age in years

Knowledge of the growth curve can be used to visually estimate the age of Burchell's zebra in the field, i.e. based on live weight and shoulder height. According to the asymptotic live weight equation, the Burchell's zebra would reach its theoretical asymptotic live weight of 316.9 kg at approximately 11 years of age, and adult body weight after 36 months of age (>292.0 kg). The adult shoulder height of the Burchell's zebra is attained after 36 months of age, thus indicating that an animal with a slightly shorter shoulder height than known adult animals, is possibly between 12 and 36 months old. However, there is only a five cm difference between zebras of approximately 24 months and adults, which makes it almost impossible to estimate age based on shoulder height after 24 months of age. In terms of live weight, an adult Burchell's zebra (i.e. older than 5 years) is approximately 64 kg heavier than a 24-month old animal, and thus visually the hindquarters of an adult will look noticeably broader than a sub-adult viewed from the rear. Viewed laterally, all zebras will appear to be adults as the greatest portion of the bulk is in the hindquarters (Smuts, 1974). This is of importance when selecting zebras with unknown ages for meat production and information on the plains zebra will contribute to meat production efficiency when culled for commercial purposes.

Zebras described as *Equus burchelli* (Gray; Sachs, 1967) or *Equus burchelli boehmi* (Ansell, 1971; revised as *Equus quagga boehmi* by Groves & Bell 2004) harvested in the Serengeti National Park (SNP), Tanzania had a noticeable lower live weight than the Burchell's zebra in the KNP. Stallions and mares from the KNP were respectively on average 22 % and 32 % heavier than the zebras in the SNP (Table 2.3; Smuts, 1975b). The Burchell's zebra of the KNP was also larger in terms of total length, vertebral column (excluding tail), shoulder height, heart girth and hindfoot length. However, zebras from eastern Zambia did not differ in live weight from the KNP zebras (Smuts, 1974). These differences indicate that the optimum age for meat production in the plains zebra may differ between taxonomy and region. With the focus on the plains zebra, a more accurate representation of the growth rate can be established when data is obtained in multiple locations to account for live weight differences.

Table 2.3 Comparison of live weights between adult zebras harvested from different locations in Africa adapted from Smuts (1975b).

Area and (sub) species	Adult stallions		Adult mares		Reference
	n	Mean Weight (kg)	n	Mean weight (kg)	
Kruger National park (Burchell's zebra)	57	318.5	51	321.6	Smuts (1975b)
Eastern Zambia (*Equus quagga boehmi*)	10	323.3	7	341.4	Smuts (1975b)
Serengeti National Park (*Equus quagga boehmi*)	13	247.3	8	241.5	Sachs (1967)

Smuts (1975b) concluded that that the optimum culling age of the Burchell's zebra for commercial exploitation is between 12 and 48 months, and for meat production at 12 months of age. According to the Von Bertalanffy theoretical growth curve, growth increments decreases after 12 months, and it is assumed that efficiency of production is thus the highest during the first 12 months of age (Smuts, 1675b). However, at 12 months, only 57 % of the mature live weight (292.0 kg) is attained, and 75 % of the mature live weight is only attained after 18 months of age. Between 12 and 18 months, there is only an 18 % increase in live weight which is a combination of meat, bone, fat, and other tissues (Smuts, 1675b). This raises the question of what the cost-efficiency is versus the meat yield in terms of dressing percentages for raising these zebras for an additional six months or more or instead be recommended to cull the Burchell's zebra after 18 months of age for meat production. Nevertheless, the study was conducted within the KNP where animals are not usually culled for commercial distribution outside of the KNP. Differences in mature live weight between subspecies and location will account for considerable differences in biomass calculations (Smuts, 1975b). Furthermore, the growth curves and recommendations made in the mentioned study were determined from 1969 to 1972, and the growth of the Burchell's zebra may have changed over the last 50 years due to environmental changes. The outdated data on the zebra highlights the need to be able to calculate the growth curve and optimum culling age for plains zebra farmed with in different regions, as it is currently the main meat-producing zebra species in South Africa.

Carcass characteristics (muscle growth and dressing percentages) and meat quality (tenderness and intramuscular fat) are not only influenced by age but by sex as well. A high degree of sexual dimorphism in body size is usually seen in most sexually segregating ungulates (Weckerly, 1998), whereas species that live in stable mixed-sex groups tend to have little or insignificant body weight dimorphism (Ruckstuhl & Neuhaus, 2002). The latter is mostly observed in species where females have a year-round oestrus cycle or in monogamous territorial species such as Kirk's dik-dik (*Madogua kirkii*) and steenbok (*Raphicerus campestris*; Neuhaus & Ruckstuhl, 2002). A lack of sexual dimorphism in live weight between male and female zebras have been observed, and is indicated in Table 2.3 (Sachs, 1967; Smuts, 1975b). The lack of sexual dimorphism is argued to be due to the

synchronised behaviour between zebra individuals within a family unit to ensure the stability of the harem, as no significant differences were observed in activity budgets between male and female plains zebras for grazing, standing or walking (Neuhaus & Ruckstuhl, 2002). The asymptotic undressed carcass weights recorded in the study of Smuts (1975b) for the Burchell's zebra, due to an absence of dimorphism, were pooled and indicated to be 316.9 kg for both stallions and mares. Shoulder heights of stallions were observed to be on average significantly 1.8 cm higher than in mares. Despite the significance, the difference is small and will be unnoticeable when observed and estimated in the field. To some extent, sexual dimorphism was observed for neck thickness in the Burchell's zebra and was observed to be only relevant in adults. The neck girth was on average, 8.1 cm greater in the stallions than in the mares (Smuts, 1975b). The influence of sexual dimorphism on undressed and dressed carcass weights have not been studied in the plains zebra and will be of value to obtain maximum meat production.

Information on the influence of nutrition, when related to the difference in nutritional value of vegetation between seasons, on the carcass yield of many game species is limited. Game meat is predominantly available during the winter hunting season (May to August) in summer rainfall areas in South Africa. Variations in live weight between seasons observed in game species, can potentially be ascribed due to a difference in nutrient availability and seasonal foraging behaviour of animals (Hoffman, Schalkwyk, & Muller, 2009). In game species as the impala, the live weight and meat quality is altered by the breeding seasons due to strenuous activities, such as fighting, restrictive feeding time and intake (Hoffman, 2000). In contrast, species such as the plains zebra with year-round oestrus cycles in stable harems with one stallion, are less likely to have a specific mating season as the reproduction potential of the group is determined by one stallion. Therefore activity budgets spent on grazing does not vary between stallions and non-pregnant mares year-round (Neuhaus & Ruckstuhl, 2002) and thus the variation in live weight between seasons may not be due to a specific breeding season but rather due to fluctuations in terms of diet components and nutritional differences in the diet. However, when the goal is meat production, stallions in bachelor groups are most likely to be harvested as they do not form part of the breeding harems. A study on activity budgets in plains zebra harems collected additional data on one bachelor group consisting out of seven stallions (Neuhaus & Ruckstuhl, 2002). The authors noticed that these bachelor stallions spent more time lying down and less time grazing than the studied harem mares and stallions (Neuhaus & Ruckstuhl, 2002). This can indicate that energy expenditure in bachelor zebras are less than harem stallions, as they are not responsible for keeping a breeding group intact.

Studies on the effect of seasonal changes on the carcass characteristics in other equine species are limited, and the effect of nutritional levels and production systems are instead studied. Horses are generally placed on pasture after weaning and fattened with a commercial feed indoors (Belaunzaran et al., 2017; De Palo et al., 2016, 2013; De Palo, Tateo, Maggiolino, & Centoducati, 2014; Domínguez et al., 2015; Franco & Lorenzo, 2014; Franco et al., 2011; Juárez et al., 2009; Lanza et al., 2009; Lorenzo, Crecente, et al., 2014; Lorenzo, Franco, Crecente, Sarriés, & Vázquez, 2014; Lorenzo & Gómez, 2012; Rossi et al., 2017; Sarriés & Beriain, 2005; Tateo, De Palo, Ceci, & Centoducati, 2008; Tateo, De Palo, Padalino, & Centoducati, 2016). In most of these studies, diets were supplemented

with a mineral/vitamin premix. Predominantly, one of two commercial feeds were fed which consisted either of corn, soybean meal/flour, wheat bran and rolled barley (rolled oats are sometimes added) (De Palo et al., 2016, 2013; Tateo et al., 2008) or of corn, soybean meal/flour, wheat bran, rolled barley, alfalfa, sugarcane molasses, beet, animal fats, calcium carbonate, sodium chloride and powder lactose (Domínguez et al., 2015; Franco & Lorenzo, 2014; Lorenzo, Crecente, et al., 2014; Lorenzo & Gómez, 2012). The influence of season is generally excluded as a significant effect when horses are raised for commercial meat production purposes. An increasing trend in the live weight and carcass weight has been observed in foals fed commercial feed and supplements indoors, however, no statistical differences were reported (De Palo et al., 2014; Daniel Franco, Crecente, Vázquez, Gómez, & Lorenzo, 2013a; Lorenzo, Crecente, et al., 2014).

2.3.1.2 Dressing percentages and offal yields

The production of meat involves the evaluation of the entire carcass, including the muscles, adipose tissues, bones, tendons, glands, and edible organs (Polidori & Vincenzetti, 2017). The meat production potential of an animal is determined by the calculation of the dressing percentage, which is the proportion of the hot and/or cold carcass weight relative to the live weight of the animal in question. Variations in the dressing percentages between species are evident; however, variations within a species can also occur as a result of differences in pre-slaughtering conditions, production system, the season of harvest, level of maturity and degree of finish. Prior to slaughter, domesticated equine and livestock are fasted to ensure gut emptying, whereas in game species this cannot be managed due to their non-domesticated nature and production system used. Gut fill needs to be considered when the live weight and dressing percentage are compared to that of domesticated species measured before slaughter (Hoffman, 2000). In addition, gut fill percentage not only affects the dressing percentage of a carcass, but also the inclusion of both the internal (organs and intestines) and external offal (head, skin, and legs). The size and weight of internal and external offal are dependent on the species, breed, maturity level and sex (for example the presence of horns in males in some species) of the animal slaughtered (Van Zyl & Ferreira, 2004). Additionally, the type of gastrointestinal tract also affects the dressing percentages with non-ruminant such as equids having lower offal contents in relation with their body size (Aganga, Aganga, Thema, & Obocheleng, 2003; Polidori & Vincenzetti, 2017).

Data on the dressing percentages of the plains zebra is limited; however, it was found to be 56 % by Onyango et al., (1998). As the latter was calculated from a small number of zebras in Kenya before 1998, plains zebras in the Western Cape, South Africa may depict different dressing percentages today due to environmental and regional differences. Nonetheless, the dressing percentages of the two plains zebras studied compares favourably to that of Galician Mountain x Hispano-Bretón crossbreed horse foals (46.90 -54.16 %) (Domínguez et al., 2015; Lorenzo, Crecente, et al., 2014), Galician Mountain foals (47.2-47.7 %) (Lorenzo, Sarriés, et al., 2013) and Martina Franca donkey foals (43.7 – 54.5 %) (Hernández-Briano et al., 2018; Polidori et al., 2015, 2008). The dressing percentages of different horse breeds and Martina Franca donkeys from various studies are presented in Table 2.4. The mean dressing percentages of commercially slaughtered horses and donkeys (Table 2.4.) were

found to have comparable or even higher dressing percentage and meat yields when compared to Nguni (52.1 %), Bonsmara (56.9 %), and Aberdeen Angus cattle (53.7 %) (Muchenje, Dzama, Chimonyo, Raats, & Strydom, 2008) as well as to the South African Mutton Merino (SAMM; 41.5 %) and Dormer sheep (44.2 %; Cloete, Hoffman, Cloete, & Fourie, 2004).

As no studies have investigated the difference in dressing percentage between male and female plains zebras, one can postulate that the lack of sexual dimorphism may result in similar dressing percentages. In support, no statistical differences for dressing percentages has been observed between male and female Italian Heavy Draft Horses (Tateo et al., 2016), Galician Mountain horses (Lorenzo, Sarriés, et al., 2013), Burguete horses (Sarriés & Beriain, 2005) and in slaughter horses from Mexico (González et al., 2006). Similarly, no significant differences were observed for red viscera weight, green viscera weight, skin weight, limbs weight and head weight between male and female horses slaughtered in Mexico (González et al., 2006).

No effect on the dressing percentages between horses raised with different nutritional levels in a semi-extensive system (SES) has been observed (De Palo et al., 2014; Lorenzo, Crecente, et al., 2014). However, it has been found that horses in an SES had significantly higher dressing percentages than horses raised in a free extensive system (FES; Lorenzo, Crecente, et al., 2014). The higher dressing percentages can be attributed to the higher amount of energy available for growth and development in SES horses, which resulted in the significant higher live weights, carcass weights and intramuscular fat percentages in these horses (Lorenzo, Crecente, et al., 2014). The dressing percentages of the plains zebra, therefore, might differ between types of production systems and regions, with the latter being related to rainfall patterns, the nutritional quality of vegetation between seasons, and also to the suitability of the plains zebra to the specific area.

Table 2.4 Mean dressing percentages of various horse breeds and the Martina Franca donkey as affected by research treatment (sex, age and breed and nutritional level).

Animal information	Treatment	Dressing percentage	Reference
Horse			
Italian heavy draft horse	Male	69.77**	#1
(11 months)	Female	68.25**	
Galician Mountain horse	Male	47.7*	#2
(15 months)	Female	47.2*	
Slaughter horses from	Male	59.21**	#3
Mexico	Female	59.30**	
Burguete	Male	63.43	#4
(16 months)	Female	63.88	
Burguete	Male	67.18	#4
(24 months)	Female	67.12	
Italian Heavy Draft Horse	6 months	70.03**	#5
	11 months	72.60**	
	18 months	73.90**	
Sanfratellano and Haflinger (15 months)	Sanfratellano	59.33**	#6
	Haflinger	59.66**	
Italian Heavy Draft Horse	Low NL[c]	73.71	#7
(11 months)	Medium NL	72.60	
	High NL	72.99	
Slaughter horses from Poland (10 years)	-	62.7* 65**	#8
Donkey			
Martina Franca	Gelding	45.1*; 46.6**	#9
	Female	43.7*; 45.6**	
Martina Franca	8 months	49.2**	#10
	12 months	53.9**	
Martina Franca (15 months)	-	53.3*; 54.5**	#11

[#1]Tateo et al. (2016), [#2]Lorenzo, Sarriés et al. (2013), [#3]González et al. (2006), [#4]Sarriés & Beriain (2005), [#5]De Palo et al. (2013), [#6]Lanza et al. (2009), [#7]De Palo et al. (2014), [#8]Litwińczuk et al. (2008), [#9]Hernández-Briano et al. (2018), [#10]Polidori et al. (2015), [#11]Polidori et al. (2008)

Abbreviations: [a]FES = free extensive system; [b]SES = semi-extensive system, [c]NL = nutritional level

*cold dressing percentage; **hot dressing percentage

2.3.2 Physical meat quality

The physical meat quality of domestic livestock, equine, and game species can be evaluated in terms of acidity (pH), water-holding capacity, tenderness, and meat surface colour following standardised methodologies as described by Honikel (1998). The physical meat quality of the plains zebra is limited to one study conducted on the loin and leg of two zebras in Kenya (Onyango et al., 1998). However, the physical meat quality and the effect of controllable and non-controllable treatments are yet to be quantified in the plains zebra and therefore merits further research. Nonetheless, the influence of intrinsic and extrinsic factors has been studied in other equine species and will be discussed.

2.3.2.1 Acidity (pH)

Ante-mortem, muscle pH ranges between 7.0-7.2 and will start to decline to 5.3-5.8 post-mortem, reaching the ultimate pH (pH_u; Huff-Lonergan, 2009). The extent of pH decline post-mortem is influenced by species, muscle type, and pre-slaughter/harvesting stress (Honikel, 2004). The conversion of muscle to meat is through the process of anaerobic glycolysis, where lactic acid derived from glycogen is a post-mortem metabolic end product. The concentration of lactic acid, and thus its dissociated H^+ is typically measured as muscle pH. The post-mortem pH_u influences the water-holding capacity, tenderness, colour, and shelf life of the meat (England, Matarneh, Scheffer, & Gerrard, 2017; Honikel, 2004). For this purpose, the pH is measured 24 hours (pH_{24h}) post-mortem (Swatland, 1994) to indicate the potential meat quality that can be expected (Warriss, 2000a). A pH_u value of 5.5-5.7 measured 24 hours post-mortem indicates that the muscle contains adequate amounts of glycogen as the animal itself was in a good physical state at slaughter. However, the variation in the final pH_u is primarily related to the muscle type and the glycogen concentration within a muscle (Honikel, 2004).

Stress experienced by animals can be acute (short-term) or chronic (long-term), with both types of stress resulting in variations in physical meat quality characteristics. Prolonged chronic stress will cause an influx of adrenaline over an extended period resulting in a glycogen depletion in muscle fibres (Maltin, Balcerzak, Tilley, & Delday, 2003), limiting the formation of lactate post-mortem (Swatland, 1994). The high induced pH can increase the water-holding capacity of meat, forming little or no exudate (Hoffman & Laubscher, 2009a). Chronic stress can, therefore, lead to dark firm and dry (DFD) meat (Hoffman, 2001; Maltin et al., 2003). In general, game animals are exposed to high levels of stress due to the physical effort exploited during hunting, consequently depleting the glycogen stores resulting to a higher pH_u value (pH >6) than usual, supporting the known tendency for game meat being DFD in appearance (Hoffman, Muller, Schutte, Calitz, & Crafford, 2005). In slaughter horses, significant increases in the lactate concentrations have been found due to stress of loading/unloading and transportation to the slaughterhouse. The stress response in these horses can be normalised through the effect of lairage and the time-bound to it (Werner & Gallo, 2008). While in game animals pre-slaughter normalisation of the stress response cannot be considered similarly due to a difference in production conditions, and stress can potentially be minimised by efficient culling/harvesting tactics specific to the species of interest (Hoffman & Laubscher, 2009).

Generally, shot placements located on the head are more effective as the animal collapses almost immediately. Shot placements in the shoulders, rib or back may initiate longer running times increasing ante-mortem stress levels impacting meat quality negatively. In addition to the species-specific tactics, an example is the preference of night culling for antelope species such as impala instead of day culling (Hoffman, 2000, 2001; Kritzinger, 2002; Von La Chevallerie & Van Zyl, 1971). With the plains zebra being categorised as an equine and game species, the effect of stress on muscle pH will be of interest when compared. A pH_{48h} has been reported to be 5.7 in the loin and 5.8 in the leg of the plains zebra (Onyango et al., 1998). The effect of stress on horse and donkey meat is limited. Despite the effect of stress, horse muscle has a favourable pH_u <6 with a low tendency towards DFD (Gill, 2005). The pH values of horse and donkey concerning the effect of slaughter age, sex and muscle type are presented in Table 2.5. All the horse meat pH measurements fell within the normal pH range for red meat, with the Martina Franca donkey meat being characterised by a pH higher than 6.0.

No effect of slaughter age on the muscle pH_{24h} has been observed in Galician Mountain x Burguete (13 vs 26 months; Domínguez et al., 2018), Galician Mountain x Hispano-Bretón (8 vs 11 months; Domínguez et al., 2015) and in Galician Mountain foals (9 vs 12 months, Franco et al., 2011). The effect of age was, however, observed in cold-blooded, Posavje and Crossbred horses from Slovenia with varying ages between seven and 35 months. The increase of age was accompanied by a significant decrease in pH values taken 14 days (β=-0.01 per month, $p<0.0001$; Kaić, Žgur, Luštrek, & Potočnik, 2018). The meat from twelve-month old Martina Franca donkeys had a lower pH than their 18-month-old counterparts (De Palo et al., 2017). No significant effects between male and female for pH_{24h} in 15-month-old Galician Mountain foals (Franco et al., 2011; Lorenzo, Sarriés, et al., 2013) and 11-month-old Italian Heavy Draft Horse (Tateo et al., 2008) were observed. Similarly, no effect was observed for meat pH four days and 14 days post-mortem in 24-month-old Burguete foals (Sarriés & Beriain, 2005) and in seven to 35-month-old cold-blooded, Posavje, and crossbred horses, respectively (Kaić et al., 2018). However, female 16-month-old Burguete foals had significantly higher pH than males taken four days post-mortem.

Differences between pH values were recorded for the *Longissimus thoracis* (LT) and *semitendinosus* (ST) muscle of cold-blooded, Posavje and crossbred horses from Slovenia (Kaić et al., 2018), and between ten retail cuts obtained from Jeju foals (Seong et al., 2016). The differences observed were attributed to differences in physical activity, stored glycogen levels, and thus, the degradation rate (Seong et al., 2016). In contrast, no effect of muscle type between the *Longissimus thoracis et lumborum* (LTL), *semimembranosus* (SM), *biceps femoris* (BF), ST, *triceps brachii* (TB), and *psoas major* and *minor* (PM) muscles were observed in Galician Mountain (Franco & Lorenzo, 2014; Lorenzo, Pateiro, & Franco, 2013) and Galician Mountain x Hispano-Bretón foal crossings (Franco & Lorenzo, 2014). Also, no effect was observed between the LTL, SM, BF, ST, and *rectus femoris* (RF) muscle for Italian Heavy Draft Foals (Tateo et al., 2008) and between the LL and ST of Poland slaughter horses (Litwińczuk et al., 2008). The contrasting results regarding slaughter age, sex and muscle type may be due to different pre-slaughter stresses experienced, such as transportation (Sarriés & Beriain, 2005).

Table 2.5 Mean (± standard error) of equine muscle pH regarding the effect of slaughter age, sex, muscle type and breed.

Animal and treatment	Age	Sex	Muscle Type	Time Post-mortem	pH	Reference
Age effect						
Galician Mountain x	13 months	Both	LT[a]	24 hours	5.61 ± 0.01	#1
Burguete horse	26 months				5.66 ± 0.01	
Galician Mountain x	8 months	Both	LTL[b]	24 hours	5.61 ± 0.01	#2
Hispano-Bretón horse	11 months				5.59 ± 0.01	
Galician Mountain horse	9 months	Both	LTL	24 hours	5.66 ± 0.01	#3
	12 months				5.62 ± 0.01	
Martina Franca donkey	12 months	-	LTL	24 hours	6.43 ± 0.08	#4
	18 months				6.11 ± 0.08	
Sex effect						
Cold-blooded, Posavje &	7-35 months	Male	LTL & ST[c]	14 days	5.57 ± 0.01	#5
Crossbred horses		Female			5.56 ± 0.02	
Galician Mountain horse	9 -12 months	Male	LTL	24 hours	5.64 ± 0.01	#3
		Female			5.59 ± 0.01	
Italian Heavy Draft Horse	11 months	Male	LTL, SM[d],	24 hours	5.59 ± 0.59	#6
		Female	BF[e], ST &		5.63 ± 0.59	
			RF[f]			
Galician Mountain horse	15 months	Male	LTL	24 hours	5.6 ± 0.01	#7
		Female			5.6 ± 0.01	
Burguete horse	16 months	Male	LTL	4 days	5.56	#8
		Female			5.63	
Burguete horse	24 months	Male	LTL	4 days	5.61	
		Female			5.58	
Muscle type						
Cold-blooded, Posavje &	7-35 months	Both	LTL	14 days	5.52 ± 0.02	#5
Crossbred horses			ST		5.61 ± 0.02	
Galician Mountain x	15 months		LTL	24 hours	5.65-5.64 ± 0.01	#9
Hispano-Bretón horse			SM		5.61-5.62 ± 0.01	
			BF		5.61-5.65 ± 0.01	
		-	ST		5.63-5.66 ± 0.01	
			TB[g]		5.66-5.69 ± 0.01	
			PM[h]		5.64-5.64 ± 0.01	
Galician Mountain horse	15 months		LTL	24 hours	5.59 ± 0.17	#10
			SM		5.61 ± 0.14	
			BF		5.64 ± 0.15	
		-	ST		5.67 ± 0.17	
			TB		5.69 ± 0.22	
			PM		5.64 ± 0.14	
Italian Heavy Draft Horse	11 months	Both	LTL	24hours	5.53 ± 0.64	#6
			SM		5.76 ± 0.64	
			BF		5.45 ± 0.64	
			ST		5.38 ± 0.64	
			RF		5.93 ± 0.64	

Table 2.5 *Continued.*

Animal and treatment	Age	Sex	Muscle Type	Time Post-mortem	pH	Reference
Breed effect						
Sanfratellano horse	15 months	-	LT	4-6 days	5.69 ± 0.02	#11
Haflinger horse					5.61 ± 0.02	
Galician Mountain horse	15 months	-	LTL	24 hours	5.68 ± 0.02	#12
Galician Mountain x Hispano-Bretón horse					5.61 ± 0.02	

[#1]Domínguez et al. (2018); [#2]Domínguez et al. (2015), [#3]Franco et al. (2011),[#4]De Palo et al. (2017), [#5]Kaić et al. (2018), [#6]Tateo et al. (2008),[#7]Lorenzo, Sarriés, et al. (2013), [#8]Sarriés & Beriain (2005), [#9]Franco & Lorenzo (2014), [#10]Lorenzo, Pateiro, & Franco (2013), [#11]Lanza et al. (2009), [#12]Franco et al. (2013a)

Abbreviations: [a]LT= *Longissimus thoracis*, [b]LTL= *Longissimus thoracis et lumborum*, [c]ST= *semitendinosus*, [d]SM= *semimembranosus*, [e]BF= *biceps femoris*, [f]RF= *rectus femoris,* [g]TB = *triceps brachii,* [h]PM= *psoas major* & *minor*

2.3.2.2 Water-holding capacity

The water-holding capacity of meat is the ability to retain its water or added water under an applied heat or force (Brewer, 2004). Muscles consist of 75 % water, which is located in the spaces between the thin actin and thick myosin fibres, mainly as a result of capillary forces within the myofibril structure (England et al., 2017; Huff-Lonergan, 2009; Huff-Lonergan & Lonergan, 2005). The water-holding ability is important to consider in terms of consumer preference, as it plays a determining role in the appearance and juiciness of meat. The water-holding capacity of meat is determined by the accumulation of liquid exudate in fresh and thawed uncooked meat and cooked meat (due to shrinkage). Water in muscles can either be bound, immobilised or in a free form. Bound water has low mobility and is highly resistant to external or applied forces. Immobilised water or entrapped water has partially limited mobility and migrate into the free water compartment during the conversion of muscle to meat and can, therefore, be lost as purge (England et al., 2017).

The water-holding capacity of meat is related to the pH_u and generally post-mortem glycolysis will reduce the pH_u to around 5.4-5.5 approaching the iso-electric point, which is considered the minimum point of water-binding by muscle proteins (Lawrie & Ledward, 2006). Consequently, high rates of post-mortem pH decline and a low pH_u can have deleterious effects on the water-holding capacity, resulting in undesirable high moisture losses. The amount of moisture loss in the form of drip build-up in packaged meat results in an unpleasant visual perception by consumers at the point of sale (Lawrie & Ledward, 2006). Also, low water-holding capacity results in significant weight losses in meat cuts and influences the serviceability of meat negatively (Troy & Kerry, 2010). Therefore, to avoid these potential economic losses, effort has been and are being made to minimise moisture loss in meat products (Troy & Kerry, 2010).

Another form of moisture loss influenced by similar factors as the drip loss, is the moisture loss of cooked meat. With the application of heat, the amount of moisture loss is time and temperature-dependent, as it changes the physical and chemical properties of meat. The effect of high cooking temperatures is interrelated with the level of protein denaturation and will significantly decrease the water-holding capacity (Troy & Kerry, 2010). In addition, the amount of shrinkage due to cooking is

associated with the level of juiciness perceived in cooked meat (Lawrie & Ledward, 2006). In general, a high moisture content is linked to improved physical meat quality characteristics such as tenderness, firmness, juiciness, and colour (England et al., 2017). The water-holding capacity of meat has an inverse relationship with the pH_u as meat with a higher pH_u tend to result in meat with decreased moisture losses (Lawrie & Ledward, 2006; Shange, Gouws, & Hoffman, 2019). To the best of our knowledge, no information is available regarding the correlation between the water-holding capacity and pH_u for equine meat. Research is thus essential to establish if inverse relationships between pH and water-holding capacity exist for all equine species such as horses, donkeys and zebras, and the effect of such correlations on equine meat quality. Significant negative correlations between the pH_u and both drip and cooking loss percentages have been observed in impala, kudu (Hoffman, Mostert, Kidd, & Laubscher, 2009), and springbok (Hoffman, Kroucamp, & Manley, 2007a).

Contradicting research for moisture loss in horsemeat was reported regarding the effect of slaughter age, sex, and muscle type. Inconsistent results reported in literature can potentially be attributed to several factors that were not standardised between studies, in particular, the breed type, production system, slaughtering practice, post-mortem processing, sample management and the method used to determine the drip and cooking loss percentages. These inconsistencies are reflected in the large variation observed between the percentage drip loss (range: 1.32 – 3.47 %) and percentage cooking loss (range: 14.94 -38.66 %) reported in horsemeat, as influenced by age, sex and muscle type (Table 2.6). Percentage drip loss in meat did not differ significantly between slaughter age, sex and muscle type between studies using the same method of analysis (Domínguez et al., 2015; Franco & Lorenzo, 2014; Franco et al., 2011). However, the meat of female horses (1.80 %) were found to have a significantly higher drip loss percentage than those of males (1.36 %) when the method described by Honikel (1998) were used (Kaić et al., 2018). Lepetit, Grajales, & Favier (2000) established that the cooking loss percentage of meat increases as the internal temperature increases. The cooking loss percentage was found to vary with 12 % between 70°C and 80°C (Lepetit et al., 2000) indicating that minor cooking temperature differences can alter the end result (Franco et al., 2011). The cooking loss of the plains zebra was determined to be 21.9 % by placing steaks in polythene bags in a heated water bath for 30 minutes at 70°C (Onyango et al., 1998).

Table 2.6 Mean values (± standard error) for the drip and cooking loss percentages in equine meat, as influenced by slaughter age, sex and muscle type.

Animal and treatment	Age	Sex	Muscle Type	Time Post-mortem	Drip loss (%)	Cooking loss (%)	Ref[a]
Age effect							
Galician Mountain x Burguete	13 months	Both	LT[b]	24 hours	-	23.3 ± 0.38	#1
	26 months				-	22.7 ± 0.38	
Galician Mountain x Hispano-Bretón	8 months	Both	LTL[c]	4 days	2.5 ± 0.13	20.0 ± 0.95	#2
	11 months				2.3 ± 0.13	15.7 ± 0.95	
Italian Heavy Draft Horse	6 months	Male	LTL	48 hours	-	37.2 ± 0.89	#3
	11 months				-	37.9 ± 0.89	
	18 months				-	38.7 ± 0.89	
Galician Mountain	9 months	Both	LTL	24 hours	1.4 ± 0.11	17.6 ± 0.92	#4
	12 months				1.3 ± 0.11	14.9 ± 0.92	
Martina Franca donkey	12 months	-	LTL	24 hours	-	41.4 ± 0.49	#5
	18 months				-	39.5 ± 0.49	
Sex effect							
Cold-blooded, Posavje & Crossbred	7-35 months	Male	LTL & ST[d]	14 days	1.4 ± 0.11	-	#6
		Female			1.8 ± 0.14	-	
Cold-blooded, Posavje & Crossbred	7-35 months	Male	LTL & ST	28 days	-	22.0	
		Female			-	21.7	
Galician Mountain	15 months	Male	LTL	24 hours	3.1 ± 0.2	21.5 ± 1.7	#7
		Female			3.9 ± 0.2	14.9 ± 1.7	
Galician Mountain	9 -12 months	Male	LTL	24 hours	1.4 ± 0.11	15.5 ± 0.92	#4
		Female			1.3 ± 0.11	16.5 ± 0.92	
Italian Heavy Draft Horse	11 months	Male	LTL, SM[e], BF[f], ST & RF[g]	10 days (thawed)	-	26.6 ± 1.02	#8
		Female			-	24.2 ± 1.02	
Muscle type							
Cold-blooded, Posavje & Crossbred	7-35 months	Both	LTL	14 days	1.51	-	#6
			ST		1.80	-	
Cold-blooded, Posavje & Crossbred	7-35 months	Both	LTL	28 days	-	19.5	
			ST		-	24.1	
Galician Mountain x Hispano-Bretón	15 months	-	LTL	24 hours	1.8 -2.1 ± 0.06	18.2-21.0 ± 0.33	#9
			SM		1.7-1.8 ± 0.06	17.2-18.7 ± 0.33	
			BF		1.8-2.2 ± 0.06	15.7-20.8± 0.33	
			ST		1.9-2.1 ± 0.06	14.6-15.2 ± 0.33	
Galician Mountain	15 months	-	LTL	24 hours	3.5 ± 0.80	19.3 ± 6.36	#10
			SM		2.2 ± 0.64	20.3 ± 4.29	
			BF		2.2 ± 1.12	17.2 ± 4.54	
			ST		2.0 ± 1.01	17.3 ± 4.87	
			TB[h]		3.0 ± 1.18	20.3 ± 5.05	
			PM[i]		2.3 ± 0.85	19.9 ± 5.35	

Table 2.6 *Continued*

Animal and treatment	Age	Sex	Muscle Type	Time Post-mortem	Drip loss (%)	Cooking loss (%)	Ref[a]
Muscle type							
Italian Heavy Draft	11 months	Both	LTL	10 days	-	25.4 ± 0.98	#8
Horse			SM	(thawed)	-	25.2 ± 0.98	
			BF		-	22.7 ± 0.98	
			ST		-	26.2 ± 0.98	
			RF			27.4 ± 0.98	
Breed effect							
Cold-blooded	7-35 months	Both	LTL &	28 days	-	21.79	#6
Posavje			ST		-	22.03	
Crossbred					-	21.6	
Sanfratellano	15 months	-	LT	4-6 days	-	24.67 ± 1.31	#11
Haflinger					-	25.94 ± 1.31	
Galician Mountain	15 months		LTL	4 days	2.17 ± 0.13	18.43 ± 0.71	#12
Galician Mountain x Hispano-Bretón		-			1.72 ± 0.13	21.33 ± 0.71	

[#1]Domínguez et al. (2018); [#2]Domínguez et al. (2015), [#3]De Palo et al. (2013), [#4]Franco et al. (2011), [#5]De Palo et al. (2017), [#6]Kaić et al. (2018),[#7]Lorenzo, Sarriés, et al. (2014), [#8]Tateo et al. (2008), [#9]Franco & Lorenzo (2014), [#10]Lorenzo, Pateiro, & Franco (2013), [#11]Lanza et al. (2009), [#12]Franco et al. (2013a)

Abbreviations: [a]Ref = Reference, [b]LT= *Longissimus thoracis*, [c]LTL= *Longissimus thoracis et lumborum*, [d]ST= *semitendinosus*, [e]SM= *semimembranosus*, [f]BF= *biceps femoris*, [g]RF= *rectus femoris*, [h]TB = *triceps brachii*, [i]PM = *psoas major & minor*

2.3.2.3 Tenderness

Tenderness of meat is currently considered as one of the most important physical parameters that influence the perception of the consumer about the eating quality or palatability of meat (Lawrie & Ledward, 2006; Troy & Kerry, 2010). Tenderness of cooked meat is determined by measuring the Warner-Bratzler shear force (WBSF), as lower shear force values is representative of improved meat tenderness. The individual and combined influence of pre- and post-slaughter factors can contribute to a variation in meat tenderness. Pre-slaughter factors include age, species, sex, breed, diet, and degree of stress prior to slaughter (Nowak, 2011). Post-slaughter, the interaction of physiochemical meat characteristics and alterations of the structural components within a muscle, influences muscle pH and consequently tenderness (King, Wheeler, Shackelford, & Koohmaraie, 2009). The structural components in question include structural proteins, the collagen content, the degree of muscle fibre shortening, and the rate and degree of post-mortem glycolysis (King et al., 2009; Troy & Kerry, 2010).

Significant variations in tenderness between muscle types and retail cuts reported for horse meat, can be attributed to the anatomical location of muscles and activity these muscles need to perform, which consequently affect the collagen content and sarcomere length of each individual muscle (Franco & Lorenzo, 2014; Seong et al., 2016; Tateo et al., 2008). A study by De Palo et al. (2016) determined the correlation between physical meat characteristics of the *Longissimus lumborum* (LL) and six selected muscles (SM, BF, ST, *supraspinatus,* and RF) in Italian Heavy Draft Horse foals slaughtered at 18 months of age. A positive correlation between the LL and the selected muscles were

reported, indicating that the ante-mortem and post-mortem conditions affected the ultimate pH of the entire carcass. A weak correlation for shear force values between LL and the selected muscles ($r < 0.279$, $p > 0.098$), with the RF ($r = 0.818$, $p < 0.000$) being the exception, and the average values of the selected muscles ($r = 0.442$, $p = 0.031$) were found. The authors concluded that the variations observed for WBSF values were influenced by biochemical changes in the body, instead of biochemical changes in individual muscle components, i.e. myofibril length, proteolytic enzyme activity and collagen content. The latter was supported by the lack of correlation ($p > 0.181$) for collagen between the LL, and the six selected muscles reported (De Palo et al., 2016).

The rate and extent of post-mortem glycolysis measured 24 hours post-mortem (pH_u) is an essential factor in determining meat tenderness. A curvilinear relationship between muscle pH_u and shear force value seems to exist due to the activity of proteolytic enzymes. The proteolytic activity reduces at an intermediate pH_u of 5.8 to 6.3, as it is not in the optimal pH range for the enzyme activity of calpains, which peaks at a neutral pH_u. Therefore, shear force values of meat tend to increase as the pH_u increase from 5.5-6.0. The effect is reversed, and meat tenderness improves (lower shear force values) as the pH_u further rises above 6.0 up to 7.0 (Purchas & Aungsupakorn, 1993; Watanabe, Daly, & Devine, 1996). Also, tougher meat can be found due to a rapid pH decline early post-mortem consequently resulting in the reduction of calpain activity combined with a relatively high calpastatin level (Troy & Kerry, 2010). Nonetheless, the influence of muscle pH on tenderness in horsemeat is evident, as Znamirowska & Stanislawczyk (2005) found a positive, although weak, correlation between the pH_u and shear force values. However, as the post-mortem time progressed a strong positive correlation was calculated at 48 hours ($r = 0.66$, $p \leq 0.05$) and 120 hours ($r = 0.72$, $p \leq 0.05$; Znamirowska & Stanislawczyk, 2005). The correlation obtained indicates that the decrease in muscle pH will lead to a decrease in shear force values and consequently, an increase in horsemeat tenderness. The mean tenderness values of horse concerning slaughter age, sex and the breed effect is summarised in Table 2.7.

Regarding the effect of age, an inverse relationship exists between meat tenderness and age (Segato, Cozzi, & Andrighetto, 1999), as it influences the amount of connective tissue and the formation of more insoluble or heat-stable collagen (Badiani, Nanni, Gatta, Tolomelli, & Manfredini, 1997; Lawrie & Ledward, 2006). The collagen content in horse muscle rapidly increases between the ages of 12 and 18 months, stabilising thereafter. The soluble collagen content in horsemeat reduces noticeably with age as it decreases from 40-50 % of the total collagen in 12-month-old horses to half the number (22-27%) in 30-month-old horses. There is a further reduction to 7-8 % in horses >10 years (Robelin et al., 1984, as cited by Lorenzo et al., 2014b). In agreement with the effect of age, Sarriés & Beriain (2006) found that the shear force values of the LTL muscle were higher in 24 - than in 16-month-old Burguete foals measured four- and eight-days post-mortem. The differences were speculated to be due to a reduced rate of muscle gain found in the older foals, which can be subsequently associated with lower protein synthesis and degradation (Sarriés & Beriain, 2006). Similarly, Kaić et al. (2018) found that an increase in age was significantly followed by an increase in horse meat WBSF values, after a 14-day ageing period of the LT and ST muscles.

In contrast, the effect of age on meat tenderness was also found to be non-significant in other horse meat studies in which meat tenderness was analysed 24 hours (Domínguez et al., 2015, 2018), 48 hours (De Palo et al., 2013) and four days post-mortem (Franco et al., 2011). The effect of age was also found to have no effect on the meat tenderness of Martina Franca donkeys 24 hours (De Palo et al., 2017), two days (Polidori, Beghelli, Cavallucci, & Vincenzetti, 2011) and four days post-mortem as well (Polidori et al., 2015). The contrasting differences may be a result of other pre- and post-slaughtering (sex, breed, muscle type, diet, and production system) effects that are not standardised throughout all these studies. Some of the studies that reported no significant effect used relatively young animals with small age intervals. It can be reasoned that these results may be due to the fact that an early slaughter age resulted in the sex steroid hormones not reaching adult serum levels, with these hormones that are linked to growth, development and maturation (Franco et al., 2011). When the effect of sex on meat tenderness was investigated in horses, various studies reported no significant effects in horses slaughtered at an early age (Franco et al., 2011; Kaić et al., 2018; Lorenzo, Sarriés, et al., 2013; Sarriés & Beriain, 2006; Tateo et al., 2008). The limited studies on the effect of horse breed type on meat tenderness has shown no effects between the selected breeds – Haflinger vs Sanfratellano (Lanza et al., 2009), Galician Mountain vs Galician Mountain x Hispano Bretón crossbreed (Franco, Crecente, Vázquez, Gómez, & Lorenzo, 2013b), and cold-blooded (Slovenian draft horse and Croatian draft horse) vs. Posavje (Croatian Posavina horse and Posavje horse) vs. crossbreeds horses (Kaić et al., 2018).

Seasonal changes that may affect meat tenderness are related to availability and quality of feed as well as breeding (Laubscher, 2009). Unlike game species, the effect of season on horsemeat tenderness is not of interest as many slaughter horses are finished indoors with controlled diets. For that reason, differences in horsemeat tenderness between productions systems and diet are more of interest. Significant effects between productions system (FES vs SES) (Franco et al., 2011), linseed supplementation (Domínguez et al., 2018) and the nutritional level (De Palo et al., 2014) on meat tenderness has been observed in horse foals. Foals from an SES had significantly lower WBSF values than foals from the FES. The latter can be attributed to the significantly higher intramuscular fat found in the SES, which can be related to the improved tenderness (Franco et al., 2011). The effect of diet was also observed by Domínguez et al. (2018) who reported lower meat shear force and pH_u values for foals on a linseed-rich concentrate diet (tender meat) than the control group (intermediate tenderness). De Palo et al., (2014) observed significant lower meat shear force values in horses fed on a low nutritive level (150 %) than horses fed on a medium (180 %) and high nutritive level (200 %). Data on the production system and diet differences on equine meat and game meat are scarcely available, and thus highlight a need for research on these aspects.

Table 2.7 Mean (± standard error) values for Warner-Bratzler shear force of equine meat regarding the effect of slaughter age, sex, and breed.

Animal and treatment	Slaughter age	Sex	Muscle type	Time Post-mortem	WBSF[a] (N)	References
Age effect						
Galician Mountain x Burguete	13 months 16 months	Both	LT[b]	24 hours	29.3-43.2 ± 1.60 36.7-40.5 ± 1.60	#1
Galician Mountain x Hispano-Bretón	8 months 11 months	Both	LTL[c]	24 hours	37.3 ± 1.23 34.5 ± 1.23	#2
Italian Heavy Draft Horse	6 months 11 months 18 months	Male	LTL	48 hours	50.0 ± 0.23 52.1 ± 0.23 55.3 ± 0.23	#3
Galician Mountain	9 months 12 months		LTL	4 days	26.2 ± 0.20 27.3 ± 0.20	#4
Burguete	16 months 24 months	Both	LTL	4 days	4.8-5.0 4.6-5.0	#5
Burguete	16 months 24 months	Both	LTL	8 days	4.8-5.0 4.6-5.0	
Martina Franca donkey	8 months 12 months	Male	LTL	4 days	54.0 ± 9.50 62.7 ± 11.40	#6
Martina Franca donkey	12 months 18 months	-	LTL	24 hours	54.9 ± 1.82 51.3 ± 1.28	#7
Sex effect						
Galician Mountain	15 months	Male Female	LTL	4 days	37.3 ± 0.5 29.2 ± 0.5	#8
Galician Mountain	9 and 12 months	Male Female	LTL	4 days	29.7 ± 0.20 24.8 ± 0.20	#4
Italian Heavy Draft Horse	11 months	Male Female	LTL, SM[d], BF[e], ST[f], RF[g]	10 days	51.4 ± 0.28 58.6 ± 0.28	#9
Burguete	16 and 24 months	Male Female	LTL	4 days	4.6-5.0 4.8-5.0	#5
Burguete	16 and 24 months	Male Female	LTL	8 days	3.5-4.3 3.7-4.5	
Breed effect						
Sanfratellano Haflinger	15 months	-	LTL	4-6 days	58.5 ± 0.44 55.8 ± 0.44	#10
Galician Mountain Galician Mountain x Hispano-Bretón	15 months	-	LTL	4 days	27.3 ± 0.17 34.1 ± 0.17	#11

#1Domínguez et al., 2018, **#2**Domínguez et al., 2015, **#3**De Palo et al., 2013, **#4**Franco et al., 2011, **#5**Sarriés & Beriain, 2006, **#6**Polidori et al. (2015), **#7**De Palo et al. (2017), **#8**Lorenzo, Sarriés, et al., 2014, **#9**Tateo et al., 2008, **#10**Lanza et al., 2009, **#11**Franco et al., 2013b

Abbreviations: [a]WBSF = Warner-Bratzler shear force, [b]LT = *Longissimus thoracis,* [c]LTL = *Longissimus thoracis et lumborum,* [d]SM = *semimembranosus,* [e]BF = *biceps femoris,* [f]ST= *semitendinosus,* [g]RF = *rectus femoris*

2.3.2.4 Surface colour

The visual observation of meat colour when purchasing a product, is the first parameter used by the consumer to gauge freshness. With red meat, consumers favour a stable, bright cherry-red colour instead of brown discolouration (Neethling, Suman, Sigge, Hoffman, & Hunt, 2017; Warriss, 2010), or iridescent green hues that are sometimes incorrectly considered as microbial spoilage (Swatland, 1984). The colour of meat is measured in accordance with the CIE Lab colour system that reports values in terms of lightness (CIE L*), red-green spectrum (CIE a*) and blue-yellow spectrum (CIE b*). From these values, the hue-angle (perceived colour) and chroma (saturation index) values are calculated (Hunt et al., 2012; Lawrie & Ledward, 2006). Changes in lightness, redness and yellowness indexes over time reflects the pigment concentration and redox state as it describes the colour deterioration of meat colour from red to brown (Mancini & Hunt, 2005).

The colour of equine and game meat has been described to be a dark red colour, not related to the eating quality of DFD (Gill, 2005; Hoffman, Muller, et al., 2005; Lorenzo, Sarriés, et al., 2014). Both species generally have smaller adipose tissue deposits than livestock species (Hoffman et al., 2016; Hoffman & Mcmillin, 2009; Lorenzo, Sarriés, et al., 2014), however, equine carcasses characteristically contain fat that has a distinctive yellow colour (Lorenzo, Sarriés, et al., 2014). Horsemeat is rich in myoglobin that has a high oxygen-binding ability. The oxidation of bright red oxymyoglobin to brown metmyoglobin when accelerated, reduces the red colour stability and shelf-life of fresh horsemeat (Badiani & Manfredini, 1994 cited by Lorenzo, Sarriés, et al., 2014). Discolouration of red meat occur with time as the interval when fresh meat will display a favourable surface colour, is relatively short lived. The colour of foal meat (24 months of age) deteriorated after three days when displayed under a gas-permeable film. Therefore, it was suggested not to keep foal meat under display for more than two days due to the development of an unattractive rusty colour (Sarriés, Beriain, & Insausti, 2011). Characterisation of colour stability in red meat species and between muscle types/retail cuts is therefore an essential measurement to ensure unnecessary revenue losses (Neethling, Suman, Sigge, & Hoffman, 2016).

A summary of studies on the colour measurement of horse and donkey meat to determine the influence of slaughter age, sex and breed are presented in Table 2.8. No significant differences between L*, a* and b* colour measurements in selected muscles exist between male and female Galician Mountain foals (Franco et al., 2011; Lorenzo, Sarriés, et al., 2013), Italian Heavy Draft Horse foals (Tateo et al., 2008), Burguete foals (Sarriés & Beriain, 2005, 2006), and Martina Franca donkey foals (Hernández-Briano et al., 2018). The absence of surface colour differences in horse and donkey meat may be due to similar muscle myoglobin concentrations (Franco et al., 2011; Lorenzo, Sarriés, et al., 2013; Sarriés & Beriain, 2005; Tateo et al., 2008), and similar levels of daily physical activity between males and females. No effect of breed type on the L*, a* and b* values were observed between the LTL muscles of Sanfratellano and Haflinger foals (Lanza et al., 2009) as well as between Galician Mountain and Galician Mountain x Hispano-Bretón foals (Franco et al., 2013a). Kaić et al. (2018) found no difference for the LT and ST (pooled data) between breed groups for L* and b* values, however, reported significant differences for a* values as the muscles from cold-blooded horses were more red than those obtained from Posavje and crossbred foals. However, all the colour measurements for the

rectus abdomini (RA) muscle was reported to be significantly higher in Burguete foals than in Hispano-Bretón foals. The significant differences observed were attributed to maturity level rather than breed differences, as the carcass measurements indicated that the Burguete foals were closer to their adult weight (Juárez et al., 2009).

Myoglobin concentration tends to increase with age, influencing the perceived surface colour of meat. Domínguez et al. (2015) and Lorenzo, Crecente, et al. (2014) reported a negative correlation ($r = -0.558$; $p < 0.01$ and $r = -0.576$; $p < 0.01$, respectively) between Fe-heme concentration and L* values in horse indicating that an increase in Fe-heme concentration results in a decrease in L* values (meat becomes darker). A significant inverse correlation between L*-values and myoglobin was also reported for plains zebra loin and leg (Onyango et al., 1998). Darkening of the LTL/LT muscle with age has been previously reported in horses (Domínguez et al., 2015, 2018; Kaić et al., 2018) and game species such as blue wildebeest (Van Heerden, 2018), kudu and impala (Mostert, 2007). Kaić et al. (2018) found that the L* value in the LT and ST muscles of horses (cold-blooded, Posavje and crossbred horses) between the ages of seven and 35 months decreases with 0.1 unit per month ($\beta = -0.10$; $p = 0.026$), indicating that these muscles get significantly darker with age. However, slaughter age did not influence meat colour in Galician Mountain foals (Franco et al., 2011) and Martina Franca donkey foals (De Palo et al., 2017; Polidori et al., 2015).

Sarriés & Beriain (2006) reported contrasting results for age, as the LTL muscle from 24-month-old Burguete foals which was characterised by significantly lighter meat (higher L* values) and lower myoglobin concentrations than their 16-month-old counterparts after four days post-mortem vacuum-packed ageing. They suggested that a more significant age range would be of value for better understanding regarding the effect of age. The higher L* values measured in the LTL of the 24-month-old foals were instead attributed to their extended stay on pasture outdoors before being placed on a finishing diet indoors (Sarriés & Beriain, 2006). However, the results were in contrast with the tendency that darker meat is observed in animals finished outdoors on pasture than animals finished indoors on concentrate diets due to differences in feeding level and activity level (Vestergaard, Oksbjerg, & Henckel, 2000). Nevertheless, several other factors are also responsible for observed colour differences in meat, such as pH and intramuscular fat (Neethling et al., 2017). The older foals had a higher intramuscular fat content which can contribute to the higher L* values observed as a positive correlation between the intramuscular fat content, and L* values were reported ($r = 0.45$; $p \leq 0.05$; Sarriés & Beriain, 2005).

Meat samples can be differentiated based on colour between productions systems, (Juárez et al., 2009; Neethling et al., 2017), production region (Hoffman, Kritzinger, & Ferreira, 2005), and harvest season (Neethling et al., 2017) as different dietary regimes characterise different systems, regions and seasons. Diet or more specifically feeding level can significantly influence the surface colour of red meat due to the eventual effect on muscle pH. Muscle pH_u and the rate of pH decline after slaughter is one of the primary reason for the colour differences observed between pasture- and grain-based feeding systems (Neethling et al., 2017) as it alters the water-holding capacity and light scattering of meat (Neethling et al., 2017; Shange et al., 2019). A higher pH_u was reported for pasture-raised cattle, reasoned to be a consequence of inadequate energy intake or to their heightened susceptibility to pre-

slaughter stress due to a lack of human contact in these types of feeding systems. Both consequences can be relevant in equid species as they are farmed extensively and semi-extensively in various regions and seasons. Low energy intake and pre-slaughter stress causes low glycogen levels in the muscle resulting in decreased lactic acid production and thus a high pH_u. High pH_u values influence meat colour by reducing the refractive index and consequently forming a closed muscle structure that absorbs light rather than reflecting light. The closed muscle structure reduces the penetration of oxygen into the muscle, resulting in a bright red surface layer of oxygenated myoglobin that is ruled out by an underlying purple layer of reduced myoglobin (Warriss, 2000b). Therefore, the meat will have a lower L*, chroma and hue-angle values and will visibly be less red and perceived as being dark (Warriss & Brown, 1993). In agreement, horse foals fed higher levels of concentrate (1.5 vs 3.0 kg fodder/foal-day) had significantly higher L* values as well as b* values in the LTL, SM, BF, ST, and TB muscles (Franco & Lorenzo, 2014). Foals finished on 3.0 kg commercial concentrate resulted in a higher intramuscular fat content in all the above-mentioned muscles, contributing to the lighter meat colour (Franco et al., 2013a; Franco & Lorenzo, 2014). Horses reared in a FES produced meat with a significantly higher pH_u, however, were significantly lighter with a more intense red colour than those raised in an SES. The animals raised in the FES were kept on pasture and not fattened with a grain-based finishing diet such as those in the SES (Lorenzo, Crecente, et al., 2014). These conflicting results merit further research to determine the influence of feeding level and production system on the meat surface colour in equine species.

Several studies have investigated the effect of muscle type on the colour measurements of horsemeat (Franco & Lorenzo, 2014; Kaić et al., 2018; Lorenzo, Pateiro, et al., 2013; P. N. Seong et al., 2016; Tateo et al., 2008). However, the comparison between these studies is difficult, as intrinsic, and extrinsic factors affecting meat surface colour differs between investigations. In general, the differences in surface colour between individual skeletal muscles can be due to the biochemical composition and myoglobin concentration (Neethling et al., 2017). The myoglobin concentration is dependent on the muscle fibre type as higher myoglobin contents is found in red (oxidative) muscle fibres than in white (glycolytic) muscle fibres (Russell, Hertz, & McMillian, 2011). Postural muscles are found to have higher levels of myoglobin and to be redder in colour, as slow-contracting fibres characterise it, compared to locomotive muscles (Warriss, 2000a). Significantly lower Fe-heme concentration in the ST muscle was observed (Franco & Lorenzo, 2014) and as expected the ST muscle (followed by the LTL) were reported to have significantly higher L*-values compared to the LTL, SM, BF, TB and PM Galician Mountain and Galician Mountain x Hispano-Bretón horse foal muscles (Franco & Lorenzo, 2014; Lorenzo, Pateiro, et al., 2013). The ST muscle, followed by the LTL muscle had the lowest a* values in comparison with the mentioned muscles (Franco & Lorenzo, 2014). The ST muscle was also observed in Italian Heavy Draft Horse foals to be the lightest muscles with the lowest myoglobin concentration compared to the LTL, SM and RF muscles (Tateo et al., 2008). The ST muscle was also found to be lighter and more yellow than the LT muscle in horses from Slovenia (Kaić et al., 2018).

Table 2.8 Mean values (± standard error) for meat surface colour measurements in equine meat regarding the effect of slaughter age, muscle type and sex.

Animal and treatment	Slaughter age	Muscle	Sex	Time post -mortem	L*	a*	b*	Ref[a]
Age effect								
Galician Mountain	13 months	LT[b]	Both	24 hours	38.6 ± 0.30	11.2 ± 0.23	10.6 ± 0.17	#1
x Burguete	16 months				34.4 ± 0.30	12.6 ± 0.23	9.1 ± 0.07	
Galician Mountain	8 months	LTL[c]	Both	24 hours	39.7 ± 0.38	12.3 ± 0.40	11.6 ± 0.23	#2
x Hispano-Bretón	11 months				37.9 ± 0.38	12.2 ± 0.40	10.9 ± 0.23	
Galician Mountain	9 months	LTL	Both	4 days	41.2 ± 0.45	10.8 ± 0.53	4.9 ± 0.25	#3
	12 months				41.1 ± 0.45	9.9 ± 0.53	4.3 ± 0.25	
Martina Franca	12 months	LTL	Both	24 hours	36.6 ± 0.71	17.0 ± 0.36	-1.5 ± 0.22	#4
Donkey	18 months				36.0 ± 0.71	18.2 ± 0.36	-0.6 ± 0.22	
Martina Franca	8 months	LTL	Male	4 days	33.6 ± 2.94	12.2 ± 0.48	8.8 ± 0.22	#5
Donkey	12 months				32.3 ± 2.36	11.5 ± 0.83	7.9 ± 0.13	
Sex effect								
Cold-blooded,	7 – 35	LTL &	Male	14 days	42.8 ± 0.47	20.0 ± 0.31	11.1 ± 0.36	#6
Posavje &	months	ST[d]	Female		41.5 ± 0.57	20.6 ± 0.37	10.7 ± 0.31	
Crossbred								
Galician Mountain	15 months	LTL	Male	24 hours	40.4 ± 1.10	17.6 ± 0.40	10.6 ± 0.30	#7
			Female		38.2 ± 1.10	17.1 ± 0.40	11.3 ± 0.30	
Galician Mountain	9 & 12	LTL	Male	4 days	40.7 ± 0.45	10.4 ± 0.53	4.9 ± 0.25	#3
	months		Female		40.5 ± 0.45	10.2 ± 0.53	4.4 ± 0.25	
Italian Heavy Draft	11 months	LTL, SM[e],	Male	72 hours	36.7 ± 1.36	11.7 ± 0.44	-1.7 ± 0.68	#8
Horse		BFf, ST	Female		36.1 ± 1.36	10.5 ± 0.44	1.0 ± 0.68	
		& RF[g]						
Burguete	16 months	LTL	Male	4 days	34.1	15.13	8.4	#9
			Female		34.2	13.79	7.3	
Burguete	24 months	LTL	Male	4 days	35.2	16.48	10.4	
			Female		35.7	16.90	10.6	
Burguete	16 months	LTL	Male	24 hours	49.5	12.28	14.3	#10
			Female		53.6	10.26	14.8	
Burguete	24 months	LTL	Male	24 hours	49.6	13.28	14.8	
			Female		50.1	12.79	13.52	
Martina Franca		ST	Male	45	35.4 ± 1.90	15.7 ± 1.10	4.6 ± 0.60	#11
Donkey			Female	minutes	35.4 ± 1.90	18.6 ± 1.10	3.9 ± 0.60	

Table 2.8 *Continued*

Animal and treatment	Slaughter age	Muscle	Sex	Time post -mortem	L*	a*	b*	Ref[a]
Breed effect								
Galician Mountain	15 months	LTL	Both	24 hours	37.0 ± 0.42	15.67 ± 0.28	8.6 ± 0.16	#12
Galician Mountain x Hispano-Bretón					37.4 ± 0.42	15.19 ± 0.28	8.6 ± 0.16	
Sanfratellano	15 moths	LTL	-	4 days	38.8 ± 0.71	15.46 ± 0.61	8.3 ± 0.44	#13
Haflinger					40.8 ± 0.71	17.02 ± 0.61	9.7 ± 0.44	
Burguete	24 months	RA[h]	Both	24 hours	34.6 ± 0.62	24.2 ± 0.74	9.6 ± 0.47	#14
Hispano-Bretón					28.4 ± 0.97	19.8 ± 0.59	7.0 ± 0.53	

Domínguez et al., 2018, [#2]Domínguez et al., 2015, [#3]Franco et al., 2011, [#4]De Palo et al., 2017, [#5]Polidori et al., 2015, [#6]Kaić et al., 2018, [#7]Lorenzo, Sarriés, et al., 2013, [#8]Tateo et al., 2008, [#9]Sarriés & Beriain, 2006, [#10]Sarriés & Beriain, 2005, [#11]Hernández-Briano et al., 2018, [#12]Franco et al., 2013b, [#13]Lanza et al., 2009, [#14]Juárez et al., 2009

Abbreviations: [a] Ref = Reference, [b]LT= *Longissimus thoracis*, [c]LTL= *Longissimus thoracis et lumborum*, [d]ST= *semitendinosus*, [e]SM= *semimembranosus*, [f]BF= *biceps femoris*, [g]RF= *rectus femoris*, [h]RA = *rectus abdomini*

2.3.3 Chemical meat quality

The nutritional value of meat is an essential factor to consider for the formulation of balanced diets in order to fulfil in human health requirements. Components such as high protein, low fat, wholesome fatty acids, minerals, vitamins, and the amino acid profile are all important to consider in the choice and consumption of meat products. There is an increasing trend towards meat products being leaner, and wholesome. As a result, the production of non-traditional or leaner species is being introduced into the South African market. Both game and equine meat are known for being a lean product, high in protein and low in fat, with an adequate mineral, vitamin, amino acid, and fatty acid profile when compared to domestic livestock species. However, to improve productivity and to produce meat products of high and consistent quality, it is necessary to evaluate the nutritive value and meat quality of each of these non-traditional meat-producing species.

The nutritive value and chemical meat quality of meat can be characterised by its calculated proximate components, consisting of 75 % moisture, 20 % protein, 1-10 % intramuscular fat, and 1% carbohydrates, vitamins, and minerals (ash content). These components of meat may vary depending on the influence of different intrinsic and extrinsic factors. The proximate composition of plains zebra meat is limited to two studies, namely Onyango et al., (1998) and Hoffman et al., (2016). The findings of both studies were in agreement; however, it should be noted that the sample size in the study of Onyango et al., (1998) was small, i.e. only two animals. The proximate composition of horse meat, as influenced by age, sex, breed, muscle type and diet, has been described by several authors (Table 2.9). Contrasting results were reported for the moisture, protein, intramuscular fat, and ash content in horsemeat studies, as influenced by the respective factors.

2.3.3.1 Moisture

The moisture content of the plains zebra loin, leg and LL were found to be 75.2 g/100g, 74.8 g/100g (Onyango et al., 1998) and 76.36 g/100g (Hoffman et al., 2016), respectively. The moisture content of the plains zebra LTL muscle is comparable to that of other game species, such as eland (75.6 g/100g; Needham, Laubser, Kotrba, Bureš, & Hoffman, 2019), kudu (75.7-75.8 g/100g; Hoffman, Mostert, Kidd, et al., 2009), blesbok (73.9-76.1 g/100g; Neethling, Hoffman, & Britz, 2014), springbok (73.24-73.39 g/100g; Hoffman, Kroucamp, & Manley, 2007b) and impala (74.0-75.0 g/100g; Hoffman, Mostert, Kidd, et al., 2009). The moisture content of plains zebra meat falls within the wide range reported for horses, i.e. from 68.11 g/100g in 6-month-old Italian Heavy Draft Horse foals (De Palo et al., 2013) to 77.40 g/100g in 15-month-old Galician Mountain foals (Lorenzo & Pateiro, 2013) which can be attributed to these influencing factors depicted in Table 2.9.

The effect of slaughter age was observed for the meat moisture content in Galician Mountain x Burguete foals (13 vs 26 months; Domínguez et al., 2018), Italian Heavy Draft Horse foals (6 vs 11 vs 18 months; De Palo et al., 2013) and in Galician Mountain foals (9 vs 12 months; Franco et al., 2011). Domínguez et al. (2018) observed a decrease in moisture content, whereas the remaining authors reported an increase in moisture content in horse meat with an increase in slaughter age. An effect of sex was observed by Tateo et al. (2008), who reported that the moisture content of meat obtained from 11-month-old Italian Heavy Draft Horse foals was higher in females than in males. However, no sex effect was reported for Galician Mountain (Franco et al., 2011; Lorenzo, Sarriés, et al., 2013) and in Burguete foals (Sarriés & Beriain, 2005). The effect of breed was reported between 24-month-old Sanfratellano and Haflinger horses (Lanza et al., 2009), however, was not reported for Burguete and Hispano-Bretón foals (Juárez et al., 2009). See Table 2.9.

Literature on the influence of season on the moisture content in horsemeat is limited, as formulated finishing diets are fed to commercial horses. The meat obtained from horses slaughtered in six different regions in northern Spain had a significantly higher mean moisture content in spring (73.9 g/100g) compared to winter (72.9 g/100g) (Belaunzaran et al., 2017). Regarding the effect of livestock production system, the LTL muscle in Galician Mountain foals raised in a FES (9 months of age) had a higher moisture content than those in an SES (12 months of age). The results were attributed to the three months extended finishing period of the SES foals, which led to higher intramuscular fat percentages (Franco et al., 2011). It is accepted that an inverse relationship between moisture and intramuscular fat typically exist in animals finished for more extended periods or when excess feed/ higher concentrate level is given (Franco, Bispo, González, Vázquez, & Moreno, 2009). In agreement, a significant inverse relationship ($r = -0.49$, $p < 0.01$) was observed by the aforementioned study. Similar results were reported by Lorenzo, Crecente, et al. (2014) regarding the effect of livestock production system and concentrate level.

2.3.3.2 Protein

The protein content of the plains zebra loin, leg and LTL was reported as 22.8 g/100g, 24.2 g/100g (Onyango et al., 1998) and 22.29 g/100g (Hoffman et al., 2016), respectively. The protein content of the LTL/loin is comparable to large-bodied game species such as eland (23.0 g/100g; Needham et al., 2019), blue wildebeest (22.3 g/100g; Van Heerden, 2018) and kudu (22.25 g/100g; Hoffman, Mostert, et al., 2009). It is also similar to that of SAMM (22.2 g/100g) and ~1.0 g/100g higher than Dormer sheep (21.9 g/100g; Cloete et al., 2004) and Aberdeen Angus (21.4 g/100g; Bureš, Bartoň, Kotrba, & Hakl, 2015). Horsemeat is known to have a high protein content which ranges up to 23.7 g/100g in the LTL muscle of horses originating from northern Spain (Table 2.9; Belaunzaran et al., 2017).

The protein content of horsemeat is influenced by sex, muscle type, livestock production system (Lorenzo, Sarriés, et al., 2014) and slaughtering age. The studies depicted in Table 2.9 did not observe any effect of slaughtering age or sex except for De Palo et al. (2013) and Tateo et al. (2008), respectively. Regarding the effect of livestock production system, the LTL muscle in Galician mountain ($p < 0.05$; Franco et al., 2011) and Galician Mountain x Hispano-Bretón foals ($p < 0.01$; Lorenzo, Crecente, et al., 2014) in an FES had a higher protein content those in an SES. A well-recognised inverse relationship exists between moisture and protein content (Browning, Huffman, Egbert, & Jungst, 1990; Hoffman, Mostert, & Laubscher, 2009; Hoffman, Kroucamp, & Manley, 2007; Rowe, Macedo, Visentainer, Souza, & Matsushita, 1999). However, a higher moisture content in the LTL muscle of horses raised in the FES as well was reported (Franco et al., 2011; Lorenzo, Crecente, et al., 2014).

2.3.3.3 Intramuscular fat

The intramuscular fat content of meat is a visual and gustatory sensorial quality parameter, evaluated upon purchase and after consumption due to its association with tenderness, juiciness, and flavour of meat. Recently, there has been a shift towards low-fat diets by the health-conscious consumer, thus intra- and intermuscular fat are considered as strong visual cues to consumers, with visible fat being associated with the meat being less healthy (Vermeulen, Schönfeldt, & Pretorius, 2015). However, game species such as the plains zebra, produces lean meat with barely any visible fat, and this meat is therefore perceived by consumers as a healthy alternative to other conventional meat types. The intramuscular fat content of the plains zebra in Kenya has been reported to be low, i.e. 0.3 g/100g in the loin and leg (Onyango et al., 1998), compared to the levels reported by Hoffman et al. (2016) for plains zebras in South Africa, i.e. 1.47 g/100g in the LL muscle.

Fat colour is also a key factor at the point of sale, as white fat is desired by consumers. Equine species, such as horse and zebra, are known to have fat with a peculiar yellow fat colour, placing a considerable strain on the development of these types of meats globally (Tateo et al., 2008). The intramuscular fat content varies largely with a range of 0.10-6.59 g/100g in the respective studies presented in Table 2.9. However, values up to 14.5 g/100g in forequarter cuts (Paleari, Soncini, Beretta, & Rossi, 1992) and 16.3 g/100g in the LT (Matsuoka, Takahashi, & Yamanaka, 1993) in horses have

been reported. The intramuscular fat content of meat from 24-month-old Burguete foals varied from 2.08 g/100g (Juárez et al., 2009) to 5.21 g/100g (Sarriés & Beriain, 2005). The high variability in the intramuscular fat content is reasoned to be due to the effect of slaughter age, sex, muscle type, breed and finishing diet (Lorenzo, Sarriés, et al., 2014).

Regarding slaughter age, Domínguez et al. (2018) found significantly higher intramuscular fat deposits in Galician Mountain x Burguete foals slaughtered at 26 months, when compared to those slaughtered at 13 months. However, no significant differences were found for intramuscular fat between eight and 11-month-old Galician Mountain x Hispano-Bretón (Domínguez et al., 2015) and between six, 11 and, 18-month-old Italian Heavy Draft Horse foals (De Palo et al., 2013). The effect of sex was observed by Tateo et al. (2008) reporting higher intramuscular fat values in male Italian Heavy Draft Horse foals whereas Lorenzo, Sarriés, et al. (2013) and Sarriés & Beriain (2005) found no effect of sex with female Burguete and Galician Mountain foals having a slightly higher intramuscular fat content than the males, respectively. Supporting the effect of livestock production system and diet, Galician Mountain foals raised in a FES had higher intramuscular fat than those in an SES. The latter is attributed to the more extended finishing period as previously mentioned (Franco et al., 2011). Similar results were reported for Galician Mountain foal x Hispano-Bretón foals (Lorenzo, Crecente, et al., 2014).

2.3.3.4 Ash and minerals

The ash content is the inorganic compound left after combustion and consists predominantly of mineral constituents (Perez & Andujar, 1981). The ash content of plains zebra meat was recorded to be 1.5 g/100g in the loin, 1.1 g/100g in the leg (Onyango et al., 1998) and 1.09 g/100g in the LL (Hoffman et al., 2016). It was observed in various game species that the LTL muscle has an ash content of approximately 1 g/100g (Hoffman et al., 2007b; Hoffman, Mostert, & Laubscher, 2009; Hoffman, Smit, & Muller, 2008; Laubser, 2018; Van Heerden, 2018). The ash content in horsemeat has a variability ranging from 0.98 g/100g in Avelignese (Rossi et al., 2017) to 4.03 g/100g in Burguete horse (Sarriés & Beriain, 2005), however, averaging around 1-2 g/100g (Table 2.9). The inter-species variation in the ash content highlights the importance of determining the influence of factors such species, breed, hormones, muscle type, sex, slaughter age, and diet differences, on the mineral content of equine and more specific plains zebra meat (Doyle, 1980; Keeton & Eddy, 2004).

Horsemeat is a valuable source of phosphorus, magnesium, iron, zinc, and copper (Badiani et al., 1997; Lorenzo, Sarriés, et al., 2014) with high levels of calcium and copper in horse bone meal (Lee et al., 2007). Lee et al. (2007) found that 100 g raw Jeju horsemeat contributes 24 % phosphorous, 2.5 % sodium, 6.7 % potassium, 21 % iron, 26 % zinc and 40 % copper to the daily recommended mineral intakes of male adults, however calcium and manganese levels were negligible. Research on the influence of abovementioned factors is limited to only a few studies on muscle type and finishing diet, as presented in Table 2.10.

Significant differences in the mineral content between horse muscles exist (Franco & Lorenzo, 2014; Lorenzo & Pateiro, 2013; Seong, Park, Kang, et al., 2016). The variation in mineral concentration can be ascribed to the difference in anatomical location of the related muscles (Zarkadas et al., 1987),

and physical activity that the muscles carry out and resultant fibre type composition (Doornenbal & Murray, 1982). Finishing diet intensity (1.5 kg vs 3.0 kg feed) was found to influence the mineral content of six muscles obtained from 15-month-old Galician Mountain foals. Calcium, potassium, phosphorus, copper, iron, and manganese levels were influenced by finishing diet, whereas magnesium, sodium and zinc levels were not affected (Franco & Lorenzo, 2014).

Table 2.9 Mean (± standard error) proximate values (g/100g meat) in equine meat in terms of slaughter age, sex, and muscle type.

Animal and treatment	Slaughter age	Sex	Muscle type	Moisture	Protein	Intramuscular fat	Ash	Reference
Age effect								
Galician Mountain x	13 months	Both	LT[a]	74.50 ± 0.21	22.50 ± 0.36	0.37 ± 0.64	1.26 ± 0.01	Domínguez et al.
Burguete	26 months			72.60 ± 0.21	22.70 ± 0.36	1.82 ± 0.64	1.38 ± 0.01	(2018)
Galician Mountain x	8 months	Both	LT	74.54 ± 0.14	20.41 ± 0.24	1.27 ± 0.09	1.20 ± 0.04	Domínguez et al.
Hispano-Bretón	11months			74.78 ± 0.14	21.05 ± 0.24	1.29 ± 0.09	1.41 ± 0.04	(2015)
Italian Heavy Draft	6 months	Males	LTL[b]	68.11 ± 0.55	23.63 ± 0.66	2.57 ± 0.32	1.97 ± 0.11	De Palo et al. (2013)
Horse	11 months			71.32 ± 0.55	21.24 ± 0.66	3.11 ± 0.32	1.38 ± 0.11	
	18 months			72.43 ± 0.55	20.13 ± 0.66	3.19± 0.32	1.25 ± 0.11	
Galician Mountain	9 months	Both	LTL	75.43 ± 0.14	20.61 ± 0.17	0.31 ± 0.05	-	Franco et al. (2011)
	12 months			75.93 ± 0.14	20.44 ± 0.17	0.16 ± 0.05	-	
Martina Franca donkey	12 months	Both	LTL	75.44 ± 0.40	19.54 ± 0.31	1.13 ± 0.12	1.27 ± 0.03	De Palo et al. (2017)
	18 months			72.64 ± 0.40	21.34 ± 0.31	1.94 ± 0.12	1.24 ± 0.03	
Martina Franca donkey	8 months	Male	LTL	77.30 ± 2.26	19.80 ± 0.24	1.76 ± 0.23	1.11 ± 0.25	Polidori et al. (2015)
	12 months			75.80 ± 1.64	21.00 ± 2.32	1.87 ± 0.18	1.33 ± 0.22	
Martina Franca Donkey	12 months	Male	LT	74.80 ± 3.31	21.40 ± 2.95	2.41 ± 0.71	1.04 ± 0.77	Polidori et al. (2011)
	18 months			72.50 ± 2.21	22.3 ± 3.01	3.71 ± 0.43	1.10 ± 0.89	
Sex effect								
Galician Mountain	15 months	Male	LTL	76.60 ± 0.20	22.30 ± 0.20	0.10 ± 0.10	1.20 ± 0.10	Lorenzo, Sarriés, et
		Female		76.20 ± 0.20	22.30 ± 0.20	0.10 ± 0.10	1.20 ± 0.10	al. (2013)
Galician Mountain	9 and 12	Male	LTL	75.67 ± 0.14	20.34 ± 0.17	0.24 ± 0.05	-	Franco et al. (2011)
	months	Female		75.76 ± 0.14	20.62 ± 0.17	0.21 ± 0.05	-	
Italian Heavy Draft	11 months	Male	LTL, SM[c],	70.35 ± 2.03	21.91 ± 1.38	4.52 ± 0.43	1.34 ± 0.19	Tateo et al. (2008)
Horse		Female	BF[d], ST[e] &	72.56 ± 2.03	19.99 ± 1.38	4.01 ± 0.43	1.03 ± 0.19	
			RF[f]					
Burguete	16 months	Male	LTL	68.37	19.91	3.01	3.34	Sarriés & Beriain
		Female		70.70	19.90	3.32	4.03	(2005)
Burguete	24 months	Male	LTL.	68.98	20.59	5.21	2.56	
		Female		71.37	20.50	4.76	3.16	

Table 2.9 *Continued*

Animal and treatment	Slaughter age	Sex	Muscle type	Moisture	Protein	Intramuscular fat	Ash	Reference
Muscle effect								
Galician Mountain	15 months	-	LTL	74.53-75.05 ± 0.18	20.67-20.99 ± 0.07	0.15-0.58 ± 0.03	1.13-1.21 ± 0.01	Franco & Lorenzo (2014)
Galician Mountain x Hispano-Bretón			SM	74.39-75.12 ± 0.18	21.74-22.34 ± 0.07	0.25-0.74 ± 0.03	1.20-1.29 ± 0.01	
			BF	74.64-74.77 ± 0.18	21.02-21.38 ± 0.07	0.49-1.11 ± 0.03	1.20-1.25 ± 0.01	
			ST	75.48-76.82 ± 0.18	21.11-21.19 ± 0.07	0.49-1.02 ± 0.03	1.18-1.21 ± 0.01	
			PM[g]	75.21-75.23 ± 0.18	20.10-20.36 ± 0.07	1.38-1.83 ± 0.03	1.23-1.28 ± 0.01	
			TB[h]	75.12 -75.35 ± 0.18	20.89-20.93 ± 0.07	0.31-0.75 ± 0.03	1.22-1.26 ± 0.01	
Galician Mountain	15 months	-	LTL	76.49 ± 0.09	22.31 ± 0.12	0.22 ± 0.02	1.25 ± 0.02	Lorenzo, Pateiro, et al. (2013)
			SM	76.69 ± 0.09	21.98 ± 0.12	0.15 ± 0.02	1.28 ± 0.02	
			BF	76.83 ± 0.09	21.67 ± 0.12	0.37 ± 0.02	1.22 ± 0.02	
			ST	77.24 ± 0.09	21.39 ± 0.12	0.31 ± 0.02	1.28 ± 0.02	
			TB	77.40 ± 0.09	21.44 ± 0.12	0.28 ± 0.02	1.27 ± 0.02	
			PM	77.24 ± 0.09	21.17 ± 0.12	0.67 ± 0.02	1.27 ± 0.02	
Italian Heavy Draft Horse	11 months	Both	LTL	69.51 ± 2.17	21.67 ±1.55	4.28 ± 0.19	1.14 ± 0.21	Tateo et al. (2008)
			SM	73.59 ± 2.17	19.57 ± 1.55	4.47 ± 0.19	1.09 ± 0.21	
			BF	68.39 ± 2.17	20.68 ± 1.55	4.23 ± 0.19	1.02 ± 0.21	
			ST	71.08 ± 2.17	21.80 ± 1.55	3.91 ± 0.19	1.50 ± 0.21	
			RF	69.15 ± 2.17	21.08± 1.55	4.31 ± 0.19	1.15 ± 0.21	
Poland slaughter horses	10 years	Both	LL[i]	69.78 ± 0.32	19.67 ± 0.19	6.59 ± 0.38	1.10 ± 0.03	Litwińczuk et al. (2008)
			ST	72.54 ± 0.23	20.17 ± 0.11	3.79 ± 0.22	1.12 ± 0.03	
Breed effect								
Avelignese Horse	12 months	Male	LL	-	20.69 ± 0.57	3.62 ± 0.10	0.99 ± 0.01	Rossi et al. (2017)
Martian Franca Donkey				-	20.03 ± 0.48	4.87 ± 0.10	0.97 ± 0.01	
Burguete	24 months	Male	LTL	72.32 ± 0.71	20.64 ± 0.73	2.08 ± 0.11	1.13 ± 0.06	Juárez et al. (2009)
Hispano-Bretón				70.58 ± 0.91	21.81 ± 0.68	2.22 ± 0.15	1.23 ± 0.05	
Sanfratellano	15 months	-	LTL	73.23 ± 0.36	-	2.29 ± 0.23	1.39 ± 0.13	Lanza et al. (2009)
Haflinger				72.80 ± 0.36	-	2.44 ± 0.23	1.57 ± 0.13	

Abbreviations: [a] LT= *Longissimus thoracis*, [b]LTL= *Longissimus thoracis et lumborum*, [c]SM= *semimembranosus,* [d]BF= *biceps femoris*, [e]ST= *semitendinosus*, [f]RF= *rectus femoris*, [g]PM = *psoas major & minor*, [h]TB = *triceps brachii*, [u]LL= *Longissimus lumborum*

Table 2.10 The mean (± standard error) mineral content (mg/100g) in equine meat in terms of tissue type.

Animal	Slaughter age	Tissue type	K	P	Na	Mg	Ca	Fe	Zn	Mn	Cu	Ref[a]
Bologna, Northern Italy	6-10 years	Thigh muscle	331.0 ± 8.00	231 ± 3.00	74.2 ± 2.70	28.9 ± 0.1	3.77 ± 0.05	3.89 ± 0.18	3.72 ± 0.21	-	0.20 ± 0.01	#1
Jeju horses	30-36 months	LTL[b]	315.5 ± 17.60	168.7 ± 6.70	38.1 ± 3.30	21.0 ± 1.30	6.3 ± 0.50	2.10 ± 0.40	2.30 ± 0.50	0.02 ± 0.00	-	#2
		Cannon bone	82.2 ± 13.80	5874.3 ± 369.5	549.2 ± 43.6	132.1 ± 12.3	10193.7 ± 3107.9	12.3 ± 5.20	4.70 ± 0.50	0.12 ± 0.03	0.79 ± 0.20	
Galician Mountain on 1.5 kg feed	15 months	LTL	220.1 ± 3.63	201.2 ± 1.96	46.03 ± 0.80	21.33 ± 0.29	3.45 ± 0.05	3.10 ± 0.04	2.55 ± 0.03	0.01 ± 0.41	0.11 ± 3.05	#3
		SM[c]	279.3 ± 3.63	228.9 ± 1.96	49.24 ± 0.80	27.74 ± 0.29	4.07 ± 0.05	3.95 ± 0.04	1.63 ± 0.03	0.02 ± 0.41	0.15 ± 3.05	
		ST[d]	281.6 ± 3.63	226.6 ± 1.96	56.88 ± 0.80	26.65 ± 0.29	4.30 ± 0.05	3.02 ± 0.04	2.02 ± 0.03	0.01 ± 0.41	0.15 ± 3.05	
		BF[e]	278.1 ± 3.63	207.8 ± 1.96	60.37 ± 0.80	28.15 ± 0.29	4.56 ± 0.05	4.83 ± 0.04	2.36 ± 0.03	0.02 ± 0.41	0.20 ± 3.05	
		TB[f]	288.3 ± 3.63	213.2 ± 1.96	57.45 ± 0.80	28.42 ± 0.29	4.10 ± 0.05	5.02 ± 0.04	2.65 ± 0.03	0.02 ± 0.41	0.21 ± 3.05	
		PM[g]	289.0 ± 3.63	202.6 ± 1.96	53.48 ± 0.80	26.80 ± 0.29	3.92 ± 0.05	4.32 ± 0.04	2.20 ± 0.03	0.02 ± 0.41	0.17 ± 3.05	
Galician Mountain on 3.0 kg feed	15 months	LTL	190.8 ± 3.63	186.1 ± 1.96	65.08 ± 0.80	27.10 ± 0.29	3.92 ± 0.05	2.78 ± 0.04	2.83 ± 0.03	0.01 ± 0.41	0.12 ± 3.05	#3
		SM	199.9 ± 3.63	197.5 ± 1.96	63.46 ± 0.80	31.08 ± 0.29	3.94 ± 0.05	3.18 ± 0.04	1.58 ± 0.03	0.01 ± 0.41	0.14 ± 3.05	
		ST	191.6 ± 3.63	198.6 ± 1.96	65.97 ± 0.80	29.13 ± 0.29	4.26 ± 0.05	2.76 ± 0.04	2.15 ± 0.03	0.01 ± 0.41	0.14 ± 3.05	
		BF	228.8 ± 3.63	184.9 ± 1.96	47.78 ± 0.80	23.87 ± 0.29	3.87 ± 0.05	3.50 ± 0.04	2.10 ± 0.03	0.01 ± 0.41	0.12 ± 3.05	
		TB	216.2 ± 3.63	185.3 ± 1.96	47.07 ± 0.80	22.63 ± 0.29	3.50 ± 0.05	3.79 ± 0.04	2.33 ± 0.03	0.01 ± 0.41	0.15 ± 3.05	
		PM	255.8 ± 3.63	202.8 ± 1.96	47.28 ± 0.80	23.32 ± 0.29	3.77 ± 0.05	3.72 ± 0.04	1.94 ± 0.03	0.01 ± 0.41	0.15 ± 3.05	
Galician Mountain	15 months	LTL	202.61 ± 2.34	187.28 ± 3.92	52.56 ± 1.20	42.32 ± 0.93	4.51 ± 0.07	2.65 ± 0.08	2.19 ± 0.01	0.01 ± 0.01	0.14 ± 0.01	#4
		SM	200.24 ± 2.34	195.91 ± 3.92	52.62 ± 1.20	41.06 ± 0.93	4.17 ± 0.07	3.14 ± 0.08	1.82 ± 0.01	0.01 ± 0.01	0.17 ± 0.01	
		ST	198.32 ± 2.34	190.58 ± 3.92	59.90 ± 1.20	40.07 ± 0.93	4.11 ± 0.07	2.56 ± 0.08	2.17 ± 0.01	0.01 ± 0.01	0.15 ± 0.01	
		BF	190.77 ± 2.34	186.20 ± 3.92	59.71 ± 1.20	41.52 ± 0.93	4.26 ± 0.07	3.72 ± 0.08	2.07 ± 0.01	0.02 ± 0.01	0.18 ± 0.01	
		TB	202.61 ± 2.34	196.39 ± 3.92	59.38 ± 1.20	43.31 ± 0.93	4.28 ± 0.07	3.90 ± 0.08	2.74 ± 0.01	0.02 ± 0.01	0.21 ± 0.01	
		PM	199.98 ± 2.34	196.50 ± 3.92	68.08 ± 1.20	38.70 ± 0.93	4.42 ± 0.07	4.04 ± 0.08	2.66 ± 0.01	0.02 ± 0.01	0.17 ± 0.01	
MF Donkey	15 months	LT[h]	343.7 ± 65.9	212.9 ± 56.7	52.5 ± 13.3	24.8 ± 6.71	8.65 ± 2.13	3.80 ± 1.01	3.67 ± 0.78	-	-	#5

[#1]Badiani et al. (1997); [#2]Lee et al. (2007); [#3] Franco & Lorenzo (2014); [#4] Lorenzo & Pateiro (2013); [#5]Polidori et al. (2008) Abbreviations: [a] Ref = references, [b]LTL= *Longissimus thoracis et lumborum*, [c]SM= *semimembranosus,* [d]ST= *semitendinosus*, [e]BF= *biceps femoris*, [f]TB = *triceps brachii*, [g]PM = *psoas major* & *minor*, [h]LT= *Longissimus thoracis*

2.4 CONCLUSION

Meat from game species is characterised by a favourable nutritional composition and can as such be labelled as a healthy and organic alternative meat product. However, a lack of product uniformity and the fact that game meat is collectively labelled under the generic term “venison”, complicate the formulation of guidelines for the industry in the farming of game species for meat purposes. The absence of such guidelines hampers the production of game meat products that are of consistent quality.

The plains zebra is a species that has not been the focus of many studies concerning meat quality, as South African consumers are sceptical about consuming equine meat. The potential for plains zebra meat production has recently gained attention due to their immunity against FMD, which impacts positively on the export potential of its meat. Limited information on the nutritional value of zebra meat indicated that plains zebra meat is high in protein, low in intramuscular fat, and has a favourable fatty acid composition.

2.5 REFERENCES

Aganga, A. A., Aganga, A. O., Thema, T., & Obocheleng, K. O. (2003). Carcass Analysis and Meat Composition of the Donkey. *Pakistan Journal of Nutrition*, *2*(3), 138–147. https://doi.org/10.3923/pjn.2003.138.147

Andersen, K. F. (1992). Size, design and interspecific interactions as restrictors of natural behaviour in multi-species exhibits. 1. Activity and intraspecific interactions of plains zebra (*Equus burchelli*). *Applied Animal Behaviour Science*, *34*(1–2), 157–174. https://doi.org/10.1016/S0168-1591(05)80064-4

Ansell, W. F. H. (1971). Order Perissodactyla. In J. Meesters & H. . Setzer (Eds.), *The mammals of Africa: an identification guide*. Washington: Smithsonian Institution Press.

Badiani, A., & Manfredini, M. (1994). The production of horse meat. *Italian Journal of Animal Science*, *20*, 5–43.

Badiani, A., Nanni, N., Gatta, P. P., Tolomelli, B., & Manfredini, M. (1997). Nutrient Profile of Horsemeat. *Journal of Food Composition and Analysis*, *10*(3), 254–269. https://doi.org/10.1006/jfca.1997.0540

Belaunzaran, X., Lavín, P., Barron, L. J. R., Mantecón, A. R., Kramer, J. K. G., & Aldai, N. (2017). An assessment of the fatty acid composition of horse-meat available at the retail level in northern Spain. *Meat Science*, *124*, 39–47. https://doi.org/10.1016/j.meatsci.2016.10.014

Bothma, J. du P. (2011). Burchell’s or plains zebra (*Equus burchelli*). *Game & Hunt*, 8–11.

Brewer, M. S. (2004). Water-Holding Capacity. In *Encyclopedia of Meat Sciences* (pp. 242–249).

Brown, K., & Gilfoyle, D. (2007). Introduction: Livestock diseases and veterinary science in Southern Africa. *South African Historical Journal*, *58*(1), 2–16. https://doi.org/10.1080/02582470709464742

Browning, M. A., Huffman, D. L., Egbert, W. R., & Jungst, S. B. (1990). Physical and compositional characteristics of beef carcasses selected for leanness. *Journal of Food Science*, *55*(1), 9–14. https://doi.org/10.1111/j.1365-2621.1990.tb06005.x

Bureš, D., Bartoň, L., Kotrba, R., & Hakl, J. (2015). Quality attributes and composition of meat from red deer (*Cervus elaphus*), fallow deer (*Dama dama*) and Aberdeen Angus and Holstein cattle (*Bos taurus*). *Journal of the Science of Food and Agriculture*, *95*(11), 2299–2306. https://doi.org/10.1002/jsfa.6950

Cam, M. A., Kirikci, K., & Garipoglu, A. V. (2018). To understand the animal behaviors is important for many aspects of animal husbandry. *Journal of Animal Health and Behavioural Science*, *2*(1), 1–3.

Carruthers, J. (2008). "Wilding the farm or farming the wild"? The evolution of scientific game ranching in South Africa from the 1960s to the present. *Transactions of the Royal Society of South Africa*, *63*(2), 160–181. https://doi.org/10.1080/00359190809519220

Cloete, J. J. E., Hoffman, L. C., Cloete, S. W. P., & Fourie, J. E. (2004). A comparison between the body composition, carcass characteristics and retail cuts of South African Mutton Merino and Dormer sheep. *South African Journal of Animal Sciences*, *34*(1), 44–51. https://doi.org/10.4314/sajas.v34i1.4040

DAFF. (2019). Agriculture, Forestry and Fisheries on foot-and-mouth disease outbreak | South African Government. Retrieved August 12, 2019, from https://www.gov.za/speeches/foot-and-mouth-disease-14-jan-2019-0000

De Palo, P., Maggiolino, A., Centoducati, P., Milella, P., Calzaretti, G., & Tateo, A. (2016). Is meat quality from *Longissimus lumborum* samples correlated with other cuts in horse meat? *Animal Science Journal*, *87*(3), 428–438. https://doi.org/10.1111/asj.12433

De Palo, P., Maggiolino, A., Centoducati, P., & Tateo, A. (2013). Slaughtering age effect on carcass traits and meat quality of italian heavy draught horse foals. *Asian-Australasian Journal of Animal Sciences*, *26*(11), 1637–1643. https://doi.org/10.5713/ajas.2013.13174

De Palo, P., Tateo, A., Maggiolino, A., & Centoducati, P. (2014). Effect of nutritive level on carcass traits and meat quality of IHDH foals. *Animal Science Journal*, *85*(7), 780–786. https://doi.org/10.1111/asj.12203

De Palo, P., Tateo, A., Maggiolino, A., Marino, R., Ceci, E., Nisi, A., & Lorenzo, J. M. (2017). Martina Franca donkey meat quality: Influence of slaughter age and suckling technique. *Meat Science*, *134*, 128–134. https://doi.org/10.1016/j.meatsci.2017.07.025

Doku, Y., Bekele, A., & Balakrishnan, M. (2007). Population status of plains zebra (*Equus quagga*) in Nechisar plains, Nechisar National Park, Ethiopia. *Tropical Ecology*, *48*(1), 79–86.

Domínguez, R., Crecente, S., Borrajo, P., Agregán, R., & Lorenzo, J. M. (2015). Effect of slaughter age on foal carcass traits and meat quality. *Animal*, *9*(10), 1713–1720.

https://doi.org/10.1017/s1751731115000671

Domínguez, R., Pateiro, M., Crecente, S., Ruiz, M., Sarriés, M. V., & Lorenzo, J. M. (2018). Effect of linseed supplementation and slaughter age on meat quality of grazing cross-bred Galician x Burguete foals. *Journal of the Science of Food and Agriculture*, *98*(1), 266–273. https://doi.org/10.1002/jsfa.8466

Doornenbal, H., & Murray, A. C. (1982). Effects of age, breed, sex and muscle on certain mineral concentrations in cattle. *Journal of Food Science*, *47*, 55–58. https://doi.org/10.1111/j.1365-2621.1982.tb11026.x

Doyle, J. J. (1980). Genetic and nongenetic factors affecting the elemental composition of human and other animal tissues- a review. *Journal of Animal Science*, *50*, 1173–1183.

Duncan, P. (1992). *Zebras, Asses, and Horses: An Action Plan for the Conservation of Wild Equids*.

England, E. M., Matarneh, S. K., Scheffer, T. L., & Gerrard, D. E. (2017). Perimortel muscle metabolism and its effects on meat quality. In *New aspects of meat quality* (pp. 63–81). Woodhead Publishing.

Estes, R. D. (2012). Zebra, asses, and horses: Family equidae. In *The behaviour guide of African mammals: including hoof mammals, carnivores, primates* (20th Anniv, pp. 235–248). London, England: University of California Press, Ltd.

Ferreira, S., Gaylard, A., Greaver, C., Hayes, J., & Cowell, C. (2016). *Large vertebrate abundance in Parks 2015/2016*. Skukuza, South Africa.

Franco, D., Bispo, E., González, L., Vázquez, J. A., & Moreno, T. (2009). Effect of finishing and ageing time on quality attributes of loin from the meat of Holstein-Fresian cull cows. *Meat Science*, *83*(3), 484–491. https://doi.org/10.1016/j.meatsci.2009.06.030

Franco, D., Crecente, S., Vázquez, J. A., Gómez, M., & Lorenzo, J. M. (2013a). Effect of cross breeding and amount of finishing diet on growth parameters, carcass and meat composition of foals slaughtered at 15 months of age. *Meat Science*, *93*(3), 547–556. https://doi.org/10.1016/j.meatsci.2012.11.018

Franco, D., Crecente, S., Vázquez, J. A., Gómez, M., & Lorenzo, J. M. (2013b). Effect of cross breeding and amount of finishing diet on growth parameters, carcass and meat composition of foals slaughtered at 15 months of age. *Meat Science*, *93*(3), 547–556. https://doi.org/10.1016/j.meatsci.2012.11.018

Franco, D., García Fontán, M. C., Temperan, S., García Calvo, L., & Lorenzo, J. M. (2010). Carcass characterization of Galician mountain foals. *Encyclopedia of Meat Sciences*.

Franco, D., & Lorenzo, J. M. (2014). Effect of muscle and intensity of finishing diet on meat quality of foals slaughtered at 15 months. *Meat Science*, *96*(1), 327–334. https://doi.org/10.1016/j.meatsci.2013.07.018

Franco, D., Rodríguez, E., Purriños, L., Crecente, S., Bermúdez, R., & Lorenzo, J. M. (2011). Meat quality of “Galician Mountain” foals breed. Effect of sex, slaughter age and livestock production

system. *Meat Science*, *88*(2), 292–298. https://doi.org/10.1016/J.MEATSCI.2011.01.004

Furstenburgh, D. (2009). Focus on the Plains Zebra (*Equus quagga*). *SA Hunter*, *08031*. Retrieved from https://www.researchgate.net/publication/316165914

Gill, C. O. (2005). Safety and storage stability of horse meat for human consumption. *Meat Science*, *71*(3), 506–513. https://doi.org/10.1016/j.meatsci.2005.04.030

González, H. G., Mota-Rojas, D., Becerril-Herrera, M., Casas-García, C., Zermeno, M., Ramírez-Necoecha, R., … Alonso-Spilsbury, M. (2006). Horse meat for human consumption in México: slaughter performance and carcass morphometry. *Journal of Food Technology*, Vol. 4, pp. 106–110.

Groves, C. P., & Bell, C. H. (2004). New investigations on the taxonomy of the zebras genus Equus, subgenus Hippotigris. *Mammalian Biology*, *69*(3), 182–196. https://doi.org/10.1078/1616-5047-00133

Hack, M. A., East, R., Rubenstein, D. I., & Gray, E. (2002). Status and action plan for the plains zebra (Equus burchellii). In P. D. Moehlman (Ed.), *Equids: Zebras, Asses and Horses. Status Survey and Conservation Action Plan* (pp. 43–60). IUCN/SSC Equid Specialist Group. IUCN, Gland, Switzerland and Cambridge UK.

Harley, E. H., Knight, M. H., Lardner, C., Wooding, B., & Gregor, M. (2009). The Quagga project : progress over 20 years of selective breeding. *South African Journal of Wildlife Research*, *39*(2), 155–163. https://doi.org/10.3957/056.039.0206

Harley, E. H., Lardner, C., Gregor, M., Wooding, B., & Knight, M. H. (2018). *The restoration of the Quagga: 24 years of selective breeding*.

Hernández-Briano, P., Ramírez-Lozano, R. G., Carrillo-Muro, O., López-Carlos, M. A., Méndez-Llorente, F., & Aguilera-Soto, J. I. (2018). Gender and live weight on carcass and meat characteristics of donkeys. *Ciencia Rural*, *48*(4), 1–7. https://doi.org/10.1590/0103-8478cr20170889

Hoffman, L. C. (2000). The yield and carcass chemical composition of impala (*Aepyceros melampus*), a southern African antelope species. *Journal of the Science of Food and Agriculture*, *80*(6), 752–756.

Hoffman, L. C. (2001). The effect of different culling methodologies on the physical meat quality attributes of various game species. *Proceedings of the 5th International Wildlife Ranching Symposium Sustainable Utilization, Pretoria, South Africa.*, 212–221.

Hoffman, L. C., Geldenhuys, G., & Cawthorn, D. M. (2016). Proximate and fatty acid composition of zebra (*Equus quagga burchellii*) muscle and subcutaneous fat. *Journal of the Science of Food and Agriculture*, *96*(11), 3922–3927. https://doi.org/10.1002/jsfa.7623

Hoffman, L. C., Kritzinger, B., & Ferreira, A. V. (2005). The effects of region and gender on the fatty acid, amino acid, mineral, myoglobin and collagen contents of impala (Aepyceros melampus)

meat. *Meat Science*, *69*(3), 551–558. https://doi.org/10.1016/j.meatsci.2004.10.006

Hoffman, L. C., Kroucamp, M., & Manley, M. (2007a). Meat quality characteristics of springbok (*Antidorcas marsupialis*). 1: Physical meat attributes as influenced by age, gender and production region. *Meat Science*, *76*(4), 755–761. https://doi.org/10.1016/j.meatsci.2007.02.017

Hoffman, L. C., Kroucamp, M., & Manley, M. (2007b). Meat quality characteristics of springbok (*Antidorcas marsupialis*). 2: Chemical composition of springbok meat as influenced by age, gender and production region. *Meat Science*, *76*(4), 762–767. https://doi.org/10.1016/J.MEATSCI.2007.02.018

Hoffman, L. C., & Laubscher, L. L. (2009a). A comparison between the effects of day and night cropping on greater kudu (*Tragelaphus strepsiceros*) meat quality. *South African Journal of Wildlife Research*, *39*(2), 164–169.

Hoffman, L. C., & Laubscher, L. L. (2009b). Comparing the effects on meat quality of conventional hunting and night cropping of impala (*Aepyceros melampus*). *South African Journal of Wildlife Research*, *39*(March), 39–47.

Hoffman, L. C., & Mcmillin, K. W. (2009). Improving the meat quality of venison and other exotic game. *Improving the Sensory and Nutritional Quality of Fresh Meat*, (1994), 447–475. https://doi.org/10.1533/9781845695439.3.447

Hoffman, L. C., Mostert, A. C., Kidd, M., & Laubscher, L. L. (2009). Meat quality of kudu (Tragelaphus strepsiceros) and impala (Aepyceros melampus): Carcass yield, physical quality and chemical composition of kudu and impala *Longissimus dorsi* muscle as affected by gender and age. *Meat Science*, *83*(4), 788–795. https://doi.org/10.1016/j.meatsci.2009.08.022

Hoffman, L. C., Mostert, A. C., & Laubscher, L. L. (2009). Meat quality of kudu (*Tragelaphus strepsiceros*) and impala (*Aepyceros melampus*): The effect of gender and age on the fatty acid profile, cholesterol content and sensory characteristics of kudu and impala meat. *Meat Science*, *83*(4), 737–743. https://doi.org/10.1016/j.meatsci.2009.08.026

Hoffman, L. C., Muller, M., Schutte, D. W., Calitz, F. J., & Crafford, K. (2005). Consumer expectations, perceptions and purchasing of South African game meat. *South African Journal of Wildlife Research*, *35*(1), 33–42.

Hoffman, L. C., Schalkwyk, S. van, & Muller, N. (2009). Effect of season and gender on the physical and chemical composition of Black Wildebeest (*Connochaetus gnou*) meat. *South African Journal of Wildlife Research*, *39*(2), 170–174. https://doi.org/10.3957/056.039.0208

Hoffman, L. C., Smit, K., & Muller, N. (2008). Chemical characteristics of blesbok (*Damaliscus dorcas phillipsi*) meat. *Journal of Food Composition and Analysis*, *21*(4), 315–319. https://doi.org/10.1016/J.JFCA.2007.12.003

Hoffman, L. C., & Wiklund, E. (2006). Game and venison - meat for the modern consumer. *Meat Science*, *74*(1), 197–208. https://doi.org/10.1016/j.meatsci.2006.04.005

Honikel, K. O. (1998). Reference methods for the assessment of physical characteristics of meat. *Meat Science*, *49*(4), 447–457. https://doi.org/10.1016/S0309-1740(98)00034-5

Honikel, K. O. (2004). Chemical and physical characteristics of meat - pH Measurement. In *Encyclopedia of Meat Sciences* (pp. 238–242). https://doi.org/10.1017/CCO9780511791581.016

Hoven, W. (2015). Private game reserves in southern Africa. In R. Van Der Duim, M. Lamers, & J. Van Wijk (Eds.), *Institutional arrangements for conservation, development and tourism in eastern and southern Africa: a dynamic perspective.* (pp. 101–108). Springer.

Hrabar, H., Birss, C., Peinke, D., King, S., Novellie, P., Kerley, G., & Child, M. (2016). *Equus zebra zebra* – Cape Mountain Zebra. *The Red List of Mammels of South Africa,Lesotho and Swaziland*, 1–14.

Huff-Lonergan, E. (2009). Fresh meat water-holding capacity. In *Improving the Sensory and Nutritional Quality of Fresh Meat* (pp. 147–160). https://doi.org/10.1533/9781845695439.1.147

Huff-Lonergan, E., & Lonergan, S. M. (2005). Mechanisms of water-holding capacity of meat: The role of postmortem biochemical and structural changes. *Meat Science*, *71*(1), 194–204. https://doi.org/10.1016/j.meatsci.2005.04.022

Hunt, M. C., King, A., Barbut, S., Clause, J., Cornforth, D., Hanson, D., … Mohan, A. (2012). AMSA meat color measurement guidelines. In *American Meat Science Association, Champaign, Illinois USA*. Retrieved from http://www.meatscience.org

Juárez, M., Polvillo, O., Gómez, M. D., Alcalde, M. J., Romero, F., & Valera, M. (2009). Breed effect on carcass and meat quality of foals slaughtered at 24 months of age. *Meat Science*, *83*(2), 224–228. https://doi.org/10.1016/j.meatsci.2009.04.022

Kaić, A., Žgur, S., Luštrek, B., & Potočnik, K. (2018). Physicochemical properties of horse meat as affected by breed, sex, age, muscle type and aging period. *Animal Production Science*, *58*(12), 2352–2357. https://doi.org/10.1071/AN17156

Keeton, J. T., & Eddy, S. (2004). Chemical and physical characteristics of meat/Chemical composition. *Encyclopedia of Meat Sciences*, (2002), 210–218. https://doi.org/10.1016/B0-12-464970-X/00118-5

King, D. A., Wheeler, T. L., Shackelford, S. D., & Koohmaraie, M. (2009). Fresh meat texture and tenderness. In J. P. Kerry & D. Ledward (Eds.), *Improving the sensory and nutritional quality of fresh meat* (pp. 61–88). Cambridge, UK: Woodhead Publishing Ltd.

Kingdon, J. (2015). *The Kingdon field guide to African mammals* (second). London, England: Bloomsbury Publishing Plc.

Klingel, H. (1969). The social organisation and population ecology of the plains zebra (*Equus Quagga*). *Zoologica Africana*, *4*(2), 249–263. https://doi.org/10.1080/00445096.1969.11447374

Klingel, H. (1972). Social Behaviour of African Equidae. *Zoologica Africana*, *7*(1), 175–185. https://doi.org/10.1080/00445096.1972.11447438

Kritzinger, B. (2002). *Meat quality parameters of the impala (Aepyceros melampus)*. Unpublished Master's thesis in Animal Science, Stellenbosch, South Africa.

Lanza, M., Landi, C., Scerra, M., Galofaro, V., & Pennisi, P. (2009). Meat quality and intramuscular fatty acid composition of Sanfratellano and Haflinger foals. *Meat Science*, *81*, 142–147. https://doi.org/10.1016/j.meatsci.2008.07.008

Laubscher, L. L. (2009). *The effect of different cropping methods on the meat quality of various game species*. Unpublished Master's thesis in Animal Science, Stellenbosch, South Africa.

Laubser, J. G. (2018). *Meat quality characteristics of male and female common eland (Tragelaphus oryx)*. Unpublished Master's thesis in Animal Science, Stellenbosch, South Africa.

Lawrie, R. A., & Ledward, D. A. (2006a). *Lawrie's meat science* (7th ed). CRC Press.

Lee, C., Seong, P., Oh, W., Ko, M., Kim, K., & Jeong, J. (2007). Nutritional characteristics of horsemeat in comparison with those of beef and pork. *Nutrition Research and Practice*, *1*(1), 70. https://doi.org/10.4162/nrp.2007.1.1.70

Lepetit, J., Grajales, A., & Favier, R. (2000). Modelling the effect of sarcomere length on collagen thermal shortening in cooked meat: Consequence on meat toughness. *Meat Science*, *54*(3), 239–250. https://doi.org/10.1016/S0309-1740(99)00086-8

Litwińczuk, A., Florelk, M., Skałecki, P., & Litwińczuk, Z. (2008). Chemical composition and physiochemical properties of horse meat from the *Longissimus lumborum* and *semitendinososus* muscle. *Journal of Muscle Foods*, *19*(3), 223–236. https://doi.org/10.1111/j.1745-4573.2008.00117.x

Lorenzo, J. M., Crecente, S., Franco, D., Sarriés, M. V., & Gómez, M. (2014). The effect of livestock production system and concentrate level on carcass traits and meat quality of foals slaughtered at 18 months of age. *Animal*, *8*(3), 494–503. https://doi.org/10.1017/S175173111300236X

Lorenzo, J. M., Franco, D., Crecente, S., Sarriés, M. V., & Vázquez, J. A. (2014). Effect of Cross Breeding With Hispano-Bretón and Burguete on Productive Parameter and Dynamics of Growth of " Galician Mountain " Foals. *60th International Congress of Meat Science and Technology*, 3–6. Punta del Este, Uruguay.

Lorenzo, J. M., & Gómez, M. (2012). Shelf life of fresh foal meat under MAP, overwrap and vacuum packaging conditions. *Meat Science*, *92*(4), 610–618. https://doi.org/10.1016/j.meatsci.2012.06.008

Lorenzo, J. M., & Pateiro, M. (2013). Influence of type of muscles on nutritional value of foal meat. *Meat Science*, *93*(3), 630–638. https://doi.org/10.1016/j.meatsci.2012.11.007

Lorenzo, J. M., Pateiro, M., & Franco, D. (2013). Influence of muscle type on physicochemical and sensory properties of foal meat. *Meat Science*, *94*(1), 77–83. https://doi.org/10.1016/j.meatsci.2013.01.001

Lorenzo, J. M., Sarriés, M. V., & Franco, D. (2013). Sex effect on meat quality and carcass traits of

foals slaughtered at 15 months of age. *Animal*, *7*(7), 1199–1207. https://doi.org/10.1017/S1751731113000189

Lorenzo, J. M., Sarriés, M. V., Tateo, A., Polidori, P., Franco, D., & Lanza, M. (2014). Carcass characteristics, meat quality and nutritional value of horsemeat: A review. *Meat Science*, *96*(4), 1478–1488. https://doi.org/10.1016/j.meatsci.2013.12.006

Louw-Carstens, M. (2019, November 8). Nuwe bek-en-klou-seer 'n "groot terugslag." *Netwerk 24*. Retrieved from https://www.netwerk24.com/Nuus/Omgewing/nuwe-bek-en-klou-seer-n-groot-terugslag-20191108#loggedin

Mahgoub, O., & Lu, C.D. (1998). Growth, body composition and carcass tissue distribution in goats of large and small sizes. *Small Ruminant Research*, *27*(3), 267–278.

Maltin, C., Balcerzak, D., Tilley, R., & Delday, M. (2003). Determinants of meat quality: tenderness. *Proceedings of the Nutrition Society*, *62*(02), 337–347. https://doi.org/10.1079/PNS2003248

Mancini, R. A., & Hunt, M. C. (2005). Current research in meat color. *Meat Science*, *71*, 100–121. https://doi.org/10.1016/j.meatsci.2005.03.003

Matsuoka, A., Takahashi, T., & Yamanaka, Y. (1993). Lipid and fatty acid composition of M. *Longissimus thoracis* of horse. *Japanese Journal of Dairy and Food Science*, *42*(3), 89–96.

Meester, J. (1954). Research on the mammals of South Africa. *Nature*, *174*, 1149–1150. Retrieved from https://www.nature.com/articles/1741149a0

Mostert, A. C. (2007). *Meat quality of Kudu (Tragelaphus strepsiceros) and Impala (Aepyceros melampus)*. Unpublished Master's thesis in Animal Science, Stellenbosch, South Africa.

Muchenje, V., Dzama, K., Chimonyo, M., Raats, J. G., & 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*(1), 20–28. https://doi.org/10.1016/J.MEATSCI.2007.07.026

Munzhedzi, S. (2018). Unlocking the socio-economic potential of South Africa's biodiversity assets through sustainable use of wildlife resources. *Department of Environmental Affairs*. Retrieved from www.environment.gov.za

Needham, T., Laubser, J. G., Kotrba, R., Bureš, D., & Hoffman, L. C. (2019). Sex influence on muscle yield and physiochemical characteristics of common eland (Taurotragus oryx) meat. *Meat Science*, *152*, 41–48. https://doi.org/10.1016/j.meatsci.2019.02.008

Neethling, J., Hoffman, L. C., & Britz, T. J. (2014). Impact of season on the chemical composition of male and female blesbok (*Damaliscus pygargus phillipsi*) muscles. *Journal of the Science of Food and Agriculture*, *94*(3), 424–431. https://doi.org/10.1002/jsfa.6281

Neethling, N. E., Suman, S. P., Sigge, G. O., & Hoffman, L. C. (2016). Muscle-specific colour stability of blesbok (*Damaliscus pygargus phillipsi*) meat. *Meat Science*, *119*, 69–79. https://doi.org/10.1016/j.meatsci.2016.04.015

Neethling, N. E., Suman, S. P., Sigge, G. O., Hoffman, L. C., & Hunt, M. C. (2017). Exogenous and endogenous factors influencing color of fresh meat from ungulates. *Meat and Muscle Biology*, *1*(1), 253. https://doi.org/10.22175/mmb2017.06.0032

Neuhaus, P., & Ruckstuhl, K. E. (2002). The link between sexual dimorphism, activity budgets, and group cohesion: The case of the plains zebra (*Equus burchelli*). *Canadian Journal of Zoology*, *80*(8), 1437–1441. https://doi.org/10.1139/Z02-126

Novellie, P., King, S., Muntifering, J., Uiseb, K., & Child, M. F. (2016). A conservation assessment of Equus zebra hartmannae. In M. F. Child, L. Roxburgh, E. Do Linh San, D. Raimondo, & H. Davies-Mostert (Eds.), *The Red List of mammals of South Africa, Lesotho and Swazilanded* (pp. 1–8). South African National Biodiversity Institute, Pretoria.

Nowak, D. (2011). Enzymes in tenderization of meat – The system of calpains and other systems. *Polish Journal of Food and Nutrition Sciences*, *61*(4), 231–237. https://doi.org/10.2478/v10222-011-0025-5

Oberem, P. (2012). Wildlife ranching in South Africa. Retrieved July 4, 2018, from Conservation, Hunting, Wildlife website: https://africageographic.com/blog/wildlife-ranching-in-south-africa/

Oberem, P., & Oberem, P. (2016). Species accounts. In *The New Game Rancher*. Biza Publications.

Onyango, C. A., Izumimoto, M., & Kutima, P. M. (1998). Comparison of some physical and chemical properties of selected game meats. *Meat Science*, *49*(1), 117–125. https://doi.org/10.1016/S0309-1740(97)00116-2

Paleari, M. A., Soncini, G., Beretta, G., & Rossi, M. T. (1992). Microbiological and chemical aspects of corned, cooked and vacuum packed horse- meat. *Italian Journal of Food Science*, *4*, 205–212.

Perez, D., & Andujar, G. (1981). Determination of ash content in meat products. *Meat Science*, *5*(3), 165–170. https://doi.org/10.1016/0309-1740(81)90001-2

Pluháček, J., & Bartoš, L. (2005). Further evidence for male infanticide and feticide in captive plains zebra, Equus burchelli. *Folia Zoologica*, *54*(3), 258–262. https://doi.org/10.1006/anbe.1999.1371

Polidori, P., Beghelli, D., Cavallucci, C., & Vincenzetti, S. (2011). Effects of age on chemical composition and tenderness of muscle *Longissimus thoracis* of Martina Franca donkey breed-Donkey meat quality at different age. *Food and Nutrition Sciences*, *02*(03), 225–227. https://doi.org/10.4236/fns.2011.23031

Polidori, P., Pucciarelli, S., Ariani, A., Polzonetti, V., & Vincenzetti, S. (2015). A comparison of the carcass and meat quality of Martina Franca donkey foals aged 8 or 12 months. *Meat Science*, *106*, 6–10. https://doi.org/10.1016/j.meatsci.2015.03.018

Polidori, P., & Vincenzetti, S. (2017). Quality and nutritional characteristics of donkey meat. In D. B. McCarthy (Ed.), *Meat and Meat Processing* (pp. 155–176). Nova Science Publishers, Inc.

Polidori, P., Vincenzetti, S., Cavallucci, C., & Beghelli, D. (2008). Quality of donkey meat and carcass characteristics. *Meat Science*, *80*(4), 1222–1224. https://doi.org/10.1016/j.meatsci.2008.05.027

Porphyre, T., & Grewar., J.D. (2019). Assessing the potential of plains zebra to maintain African horse sickness in the Western Cape Province, South Africa. PLoS ONE 14(10). https://doi.org/10.1371/journal.pone.0222366

Purchas, R. W., & Aungsupakorn, R. (1993). Further investigations into the relationship between ultimate pH and tenderness for beef samples from bulls and steers. *Meat Science*, *34*(2), 163–178. https://doi.org/10.1016/0309-1740(93)90025-D

Renecker, T. A., Renecker, L. A., & Mallory, F. F. (2005). Relationships between carcass characteristics, meat quality, age and sex of free-ranging Alaskan reindeer: a pilot study. *Rangifer*, *25*(2), 107–121.

Rossi, R., Ratti, S., Pastorelli, G., Maghin, F., Martemucci, G., Casamassima, D., … Corino, C. (2017). Effect of dietary plant extract on meat quality and sensory parameters of meat from Equidae. *Journal of the Science of Food and Agriculture*, *97*(14), 4690–4696. https://doi.org/10.1002/jsfa.8334

Rowe, A., Macedo, F. A. F., Visentainer, J. V., Souza, N. E., & Matsushita, M. (1999). Muscle composition and fatty acid profile in lambs fattened in drylot or pasture. *Meat Science*, *51*(4), 283–288. https://doi.org/10.1016/S0309-1740(98)00063-1

Rubsentein, D. I. (2018). Ecology and sociality in horses and zebras. In *Ecological Aspects of Social Evolution* (pp. 282–302). https://doi.org/10.2307/j.ctt7zvwgq.17

Ruckstuhl, K. E., & Neuhaus, P. (2002). Sexual segregation in ungulates: A comparative test of three hypotheses. *Biological Reviews of the Cambridge Philosophical Society*, *77*(1), 77–96. https://doi.org/10.1017/S1464793101005814

Russell, P. J., Hertz, P. E., & McMillian, B. (2011). Muscles, bones and other movements. In *Biology: the dynamic science* (2nd ed., pp. 937–952). Yolanda Cossio.

Sachs, R. (1967). Liveweights and body measurements of Serengeti game animals. *African Journal of Ecology*, *5*(1), 24–36. https://doi.org/10.1111/j.1365-2028.1967.tb00758.x

Sarriés, M. V., & Beriain, M. J. (2005). Carcass characteristics and meat quality of male and female foals. *Meat Science*, *70*(1), 141–152. https://doi.org/10.1016/j.meatsci.2004.12.006

Sarriés, M. V., & Beriain, M. J. (2006). Colour and texture characteristics in meat of male and female foals. *Meat Science*, *74*(4), 738–745. https://doi.org/10.1016/j.meatsci.2006.06.005

Sarriés, M. V., Beriain, M. J., & Insausti, K. (2011). Meat foal colour oxidation . *57th International Congress of Meat Science and Technology*, 24–27. Ghent-Belgium.

Schilder, M. B. H., & Boer, P. L. (1987). Ethological investigations on a herd of plains zebra in a safari park: Time-budgets, reproduction and food competition. *Applied Animal Behaviour Science*, *18*(1), 45–56. https://doi.org/10.1016/0168-1591(87)90253-X

Segato, S., Cozzi, G., & Andrighetto, I. (1999). Effect of animal morphotype, sex and age on quality of horse meat imported from Poland. *Recent Progress in Animal Production Science: Progress, XII*

Congress, 674–676.

Seong, P. N., Park, K. M., Kang, G. H., Cho, S. H., Park, B. Y., Chae, H. S., & Ba, H. Van. (2016). The differences in chemical composition, physical quality traits and nutritional horse meat as affected by various retail cut types. *Asian Australas Journal of Animal Science*, *29*, 89–99.

Seong, P., Park, K. M., Cho, S., Kang, G. H., Chae, H. S., Park, B. Y., & Van Ba, H. (2016). Effect of cut type and post-mortem ageing on the technological quality, textural profile and sensory characteristics of horse meat. *Animal Production Science*, *56*(9), 1551. https://doi.org/10.1071/AN14545

Shange, N., Gouws, P., & Hoffman, L. C. (2019). Changes in pH, colour and the microbiology of black wildebeest (Connochaetes gnou) *Longissimus thoracis et lumborum* (LTL) muscle with normal and high (DFD) muscle pH. *Meat Science*, *147*, 13–19. https://doi.org/10.1016/j.meatsci.2018.08.021

Smuts, G. L. (1974). Age determination in Burchell's zebra (Equus burchelli antiquorum) from the Kruger National Park. *Journal of South Africa Wildlife Management Assesment*, *4*(2), 103–115. Retrieved from https://journals.co.za/content/wild/4/2/AJA03794369_1959

Smuts, G. L. (1975a). Home range sizes for burchell's zebra equus burchelli antiquorum from the Kruger National Park. *Koedoe*, *18*, 139–146. Retrieved from http://www.koedoe.co.za/index.php/koedoe/article/viewArticle/918

Smuts, G. L. (1975b). Pre- and postnatal growth phenomena of Burchell's Zebra *Equus Burchelli Antiquorum*. *Koedoe*, *18*(1), 69–102. https://doi.org/10.4102/koedoe.v18i1.915

Sookhareea, R., Taylor, D., Woodford, K., Dryden, G., & Shorthose, R. (1995). The effect of slaughter age and sex type on meat quality of Javan rusa (*Cervus timoriensis*) male deer. *CSIRO Meat Industry Research Conference*, 7B 17-7B 20.

Stears, K., Shrader, A., & Castley, G. (2016). A conservation assessment of *Equus quagga*. In M. F. Child, L. Roxburgh, E. Do Linh San, D. Raimondo, & H. T. Davies-Mostert (Eds.), *The Red List of mammals of South Africa, Lesotho and Swaziland* (pp. 1–8). Retrieved from http://www.lynxeds.com/product/mammals-africa-volumes-1-6

Stuart, C. (2015). *Stuart's field guide to mammals of Southern Africa: Including Angola, Zambia and Malawi*.

Swatland, H. J. (1984). Optical Characteristics of Natural Iridescence in Meat. *Journal of Food Science*, *49*, 685–686.

Swatland, H. J. (1994). *Structure and developement of meat animals and poultry*. Technomic Publishing Company, Inc.

Tateo, A., De Palo, P., Ceci, E., & Centoducati, P. (2008). Physicochemical properties of meat of Italian Heavy Draft horses slaughtered at the age of eleven months. *Journal of Animal Science*, *86*(5), 1205–1214. https://doi.org/10.2527/jas.2007-0629

Tateo, A., De Palo, P., Padalino, B., & Centoducati, P. (2016). Quality of carcasses in I.H.D.H. foals reared in the province of Bari (Italy). *Italian Journal of Animal Science*, *4*(May 2014), 418–420. https://doi.org/10.4081/ijas.2005.2s.418

Taylor, A., Lindsey, P., & Davies-Mostert, H. (2016). An assessment of the economic social and conservation value of the wildlife ranching industry and its potential to support the green economy in South Africa. The Endagered Wildlife Trust. Johannesburg, South Africa. Retrieved from https://www.sagreenfund.org.za/wordpress/wp-content/uploads/2016/04/EWT-RESEARCH-REPORT.pdf

Thomson, K.J. (2003). World agriculture: towards 2015/2030: an FAO perspective. *Land Use Policy*, *20*(4), 375. https://doi.org/10.1016/S0264-8377(03)00047-4

Troy, D. J., & Kerry, J. P. (2010). Consumer perception and the role of science in the meat industry. *Meat Science*, *86*, 214–226. https://doi.org/10.1016/j.meatsci.2010.05.009

Van der Merwe, P. (2014). Introduction to game farm tourism. In P. van der Merwe & L. du Plessis (Eds.), *Game farm & hunting tourism* (pp. 1–24). Sun Media Metro.

Van der Merwe, P., Saayman, M., & Krugell, W. (2004). Factors that determine the price of game. *Koedoe*, *47*(2), 105–113. https://doi.org/10.4102/koedoe.v47i2.86

Van Heerden, A. M. (2018). *Profiling the meat quality of blue wildebeest (Connochaetes taurinus)*. Unpublished Master's thesis in Animal Science, Stellenbosch, South Africa.

Van Schalkwyk, D. L., & Hoffman, L. C. (2010). *Guidelines for the harvesting of game for meat export 2010*. Windhoek, Namibia: Ministry of Environment & Tourism.

Van Zyl, L., & Ferreira, A. V. (2004). Physical and chemical carcass composition of springbok (*Antidorcas marsupialis*), blesbok (*Damaliscus dorcas phillipsi*) and impala (*Aepyceros melampus*). *Small Ruminant Research*, *53*(1–2), 103–109. https://doi.org/10.1016/j.smallrumres.2003.08.017

Vermeulen, H., Schönfeldt, H. C., & Pretorius, B. (2015). A consumers perspective of the South African red meat classification system. *South African Journal of Animal Science*, *45*(3), 339–353. https://doi.org/http://dx.doi.org/10.4314/sajas.v45i3.11

Vestergaard, M., Oksbjerg, N., & Henckel, P. (2000). Influence of feeding intensity, grazing and finishing feeding on muscle fibre characteristics and meat colour of semitendinosus, Longissimus dorsi and supraspinatus muscles of young bulls. *Meat Science*, *54*(2), 177–185. https://doi.org/10.1016/S0309-1740(99)00097-2

Von La Chevallerie, M. (1970). Meat production from wild ungulates. *Proceedings of the South African Society for Animal Production, 9*, 73–87.

Von La Chevallerie, M., & Van Zyl, J. H. M. (1971). Some effects of shooting on losses of meat and meat quality in springbok and impala. *South African Journal of Animal Science*, *1*, 113–116.

Warner, R. D. (2014). Measurement of meat quality. In C. E. Devine & M. E. Dikeman (Eds.),

Encyclopedia of Meat Sciences (2nd ed., pp. 164–171). Oxford: Elsevier Academic Press.

Warriss, P. D. (2000a). *Meat science: an introductory text.* Wallingford: CABI Publishing.

Warriss, P. D. (2000b). The effects of live animal handling on carcass and meat quality. In *Meat science An introductory text* (pp. 131–154). Wallingford: CABI Publishing.

Warriss, P. D. (2010). *Meat Science: an Introductory Text.*

Warriss, P. D., & Brown, S. N. (1993). Relationships between the subjective assessment of pork quality and objective measures of colour. *Safety and Quality of Food from Animals*, (17), 68–71. https://doi.org/10.1017/S0263967X00001397

Watanabe, A., Daly, C. C., & Devine, C. E. (1996). The effects of the ultimate pH of meat on tenderness changes during ageing. *Meat Science*, *42*(1), 67–78. https://doi.org/10.1016/0309-1740(95)00012-7

Wildswinkel. (2017). *Statistics* (Version 5.1) [Mobile application software]. Retrieved from https://apps.apple.com/za/app/wildswinkel/id1079382333

Weckerly, F. W. (1998). Sexual-size dimorphism: Influence of mass and mating systems in the most dimorphic mammals. *Journal of Mammalogy*, *79*(1), 33–52. https://doi.org/10.2307/1382840

Werner, M., & Gallo, C. (2008). Effects of transport, lairage and stunning on the concentrations of some blood constituents in horses destined for slaughter. *Livestock Science*, *115*(1), 94–98. https://doi.org/10.1016/j.livsci.2007.12.023

Zarkadas, C.G., Marshall, W.D., Khalili, A.D., Ngujyen, Q., Zarkadas, G.C., Karatzas, C.N., & Khanizadeh, S. (1987). Mineral composition of selected bovine, porcine and avian muscles, and meat products. *Journal of Food Science*, *52*(3), 520–525.

Zientara, S., Weyer, C.T., Lecollinet, S., (2015). African horse sickness. *Revue Scientifique et Technique International Office of Epizootics,* 34(2), 315-327.

Znamirowska, A. (2005). Prediction of horse carcass composition using linear measurements. *Meat Science*, *69*(3), 567–570. https://doi.org/10.1016/j.meatsci.2004.10.008

Znamirowska, A., & Stanislawczyk, R. (2005). Effect of maturing process on changes in physico-chemical propertes of colt meat. *Acta Scientiarum Polonorum Technologia Alimentaria*, *4*(2), 79–88.

CHAPTER 3

THE YIELD AND CARCASS COMPOSITION OF THE PLAINS ZEBRA (*Equus quagga*) HARVESTED IN TWO SEASONS

ABSTRACT

This study presents baseline data on the meat production and consumable offal of plains zebras harvested over two seasons, and at different localities in the Western Cape Province, South Africa. Eight plains zebras were harvested in the winter season at Prinskraal farm, and 12 plains zebras in the summer season at Elandsberg Nature Reserve. The winter-harvested group had a mean undressed carcass (dead) weight of 324.4 ± 5.55 kg, compared to the 291.50 ± 11.65 kg for the summer-harvested group. The warm and cold dressing percentages were numerically higher for the winter-harvested animals (59.5 ± 0.55 % and 58.0 ± 0.60 %, respectively) than for the summer-harvested animals (58.1 ± 0.68 % and 56.60 ± 0.70 %, respectively). External offal weight was similar for the winter-harvested and summer-harvested groups, i.e. 41.98 ± 0.85 kg and 41.95 ± 1.33 kg, respectively. Regarding the internal offal weight, the winter-harvested animals (70.76 ± 21.8 kg) yielded heavier offal, when compared to the summer-harvested animals (66.13 ± 3.78 kg). To determine the proportional contribution of specific muscles to cold carcass weight, the *Longissimus et lumborum* (LTL), *semimembranosus* (SM), *biceps femoris* (BF), *semitendinosus* (ST), *psoas major* (PM), *infraspinatus* (IS), and *supraspinatus* (SS) muscles were removed from animals harvested during both seasons. Harvest season did not influence the proportional contribution of the respective muscles to the cold carcass weight, with the LTL being the heaviest, followed by the BF and then the SM. Pooled data of the percentage contribution of each muscle to cold carcass weight indicated that the LTL (3.5 ± 0.18 %), SM (1.6 ± 0.04 %), BF (2.7 ± 0.05 %) and ST (0.9 ± 0.02 %) muscles differed significantly from one another. No significant difference was recorded between the contribution of the IS (0.6 ± 0.02 %), SS (0.4 ± 0.03 %) and PM (0.6 ± 0.02 %) muscles. Carcass and muscle yields, as well as a low-cost protein offal yields from this study indicate that the plains zebra has the potential to be utilised as an alternative protein source to humans.

Keywords: Game meat, Carcass yields, Offal yields, Muscle yields

3.1 INTRODUCTION

Meat derived from game animals is a well-known protein source in South Africa, forming part of the consumptive use in game industry (Erasmus & Hoffman, 2017). Game species that are suitable for meat production are adapted to survive in harsh environments while producing quality meat (Oberem & Oberem, 2016). When farmed on marginal lands, game species produce meat with a higher return rate than conventional livestock farming (Cooper & Van der Merwe, 2014; Du Toit, Meissner, & van Niekerk, 2013). The plains zebra is physiologically and behaviourally adapted to survive in semi-arid conditions with low-quality forage (Estes, 2012).They achieve this by having a hindgut digestive system that enables them to digest forage low in protein and high in structural carbohydrates, i.e. the long, tough stems and grass in the early stages of flush (Estes, 2012). Plains zebra are capable of digesting cellulose at a faster rate than ruminants on the same terrain (Hack, East, Rubenstein, & Gray, 2002) and are able to extract more protein from poor quality grass than ruminants such as the wildebeest (Estes, 2012). The plains zebra is normally the first to venture into pastures that are wetter, more wooded and taller, acting as a pioneer for grassland biodiversity, and as a consequence of this, allows for the utilisation of forage by subsequent species such as antelope and wildebeest (Estes, 2012).

The plains zebra can thus be considered as a good candidate for mixed species farming and can be maintained at higher stocking densities than similar-sized ruminants in grasslands of a poor nutritional quality (Hack et al., 2002). A dry zebra mare of 4 years of age is equal to 1.55 Large Animal Units (LAU), and a dry mare 7 years of age is equal to 1.65 LAU. Compared to mares, a zebra stallion 4 years of age is equal to 1.40 LAU, while a zebra stallion 7 years of age is equal to 1.45 LAU (Meissner, Hofmeyr, Van Rensburg, & Pienaar, 1983). Based on these values, this amounts to an average of 1.52 zebras per LAU (Bothma, 2011; Meissner et al., 1983). A more refined stocking density given by Bothma (2011) for zebras is calculated to be approximately 0.76 zebras per grazer unit, or 0.63 zebras per browser unit.

The combined promotion of zebra meat and other game species can potentially contribute to the promotion of game meat production, which will assist in the development of both the local and export game meat market. The plains zebra is not a cloven-hoofed animal and is not susceptible to the endemic foot-and-mouth disease (FMD), and therefore they do not fall under the FMD control regulations. This makes them a suitable species for meat production purposes for export by game abattoirs and meat exporters (Scoones et al., 2010). However, for game meat to compete with existing meat products, the meat production potential of various game species needs to be established to inform, not only the consumers but meat processors and marketers as well. The first step to establishing the meat production potential of a game species is to obtain baseline information on slaughter traits such as carcass, muscle and offal yields to determine the potential of such species for meat production purposes. Knowledge of these attributes in combination with nutritional and sensory attributes of the meat will help predict the value and the potential markets that the product can reach and be exported to. Information on the meat production of game species is limited, especially for plains game such as the plains zebra.

According to Estes (2012), the plains zebra has the potential to be a commercially viable meat-producing animal due to its adaptation capability to various environments and large body size, with

males weighing an average of 220-322 kg and females 175-250 kg. Research on carcass characteristics of the plains zebra will enable marketers to be more informed about the potential of game meat as an alternative/substitute protein source, which will assist in marketing approaches that can aid in the development of new markets for game meat. The utilization of game as an additional protein source can also potentially benefit food security in South Africa in an environmental and economically sustainable manner (Cooper & Van der Merwe, 2014). South Africa produces an insufficient amount of protein annually and needs to expand the utilisation of alternative local protein sources to meet the high demand because of the fast-growing population. Mid-year estimates over the last decade for the South African population indicated an approximate increase of 18.56 % from 2008 to 2018 (StatsSA, 2008, 2018).

Game animals are typically culled by professional teams with a rifle on game farms, with carcasses that are then transported to processing plants, from where the meat is distributed for commercial use (Van Schalkwyk & Hoffman, 2016). South Africa has a strong hunting culture, which generates the potential for the utilisation of game meat, which in turn presents an economic incentive to the conservation of wildlife in South Africa (Hoffman, Muller, Schutte, Calitz, & Crafford, 2005). Not only is the plains zebra readily culled and hunted by professional teams and recreational hunters for their skins and meat but are also seen as a tourist attraction due to their unique phenotype. As a result, game animals such as the zebra, directly and indirectly, contribute to ecotourism, trophy hunting, breeding, game lodges, photographic safaris, and stock sales (Cooper & Van der Merwe, 2014).

The abovementioned information indicate that a study on the meat production potential of plains zebra, as quantified by carcass characteristics and meat quality parameters, is warranted to determine the potential contribution of this species to the economic viability and sustainability of the game industry in South Africa. Therefore, the aim of this study was to generate baseline information on carcass, muscle and offal yields in the plains zebra, which in turn may assist game farmers on the potential of this species to be used for meat production purposes.

3.2 MATERIALS AND METHODS

3.2.1 Animals and study location

3.2.1.1 Winter harvest: Prinskraal (June 2017)

Eight male plains zebras were culled during June 2017 on Prinskraal farm (24°37’ 45.1” S – 20°06’44.9” E) near Bredasdorp in the Western Cape Province, South Africa. The plains zebras were mature and free-roaming and were cropped as part of the standard annual population control protocol of the farm. The farm is located in the Fynbos biome of South Africa, which is characterised by the presence of Central Rûens Shale Renosterveld that consist out of low to medium-tall grasses and dominated by renosterbos (*Elytropappus rhinocerotis*). Vegetation in this biome also includes *Aspalathus, Athanasia* and *Rhus* species (Lasislav Mucina & Rutherford, 2006). The area receives an average annual precipitation of 300-480mm, peaking from late autumn to the winter months (May to August) when 49% of the annual rains are received. The region is characterised by a maximum mean daily temperature of

27.3°C in January, and a minimum of 5.6°C in July (Rebelo, Boucher, Helme, Mucina, & Rutherford, 2006).

Prior to the harvesting, the animals were maintained with 400 animals of various game species in a veld camp of approximately 800 ha in size. The zebras mostly foraged on Bermuda grass (*Cynodon dactylon*) and the natural vegetation as described above. They also foraged on oat pastures when available. In the dry months, the zebras were given supplementary mineral licks.

3.2.1.2 Summer harvest: Elandsberg Nature Reserve – Bartholomeus Klip (January 2018)

Twelve male plains zebras were culled during January 2018 in the Elandsberg Nature Reserve – Bartholomeus Klip (33°28'19.913'' S - 19°2'18.916" E), near Hermon in the Western Cape Province, South Africa. The plains zebras for this harvest group consisted of sub-adult and adult plains zebras that were culled as part of the annual cropping program of the Quagga Project (Harley, Knight, Lardner, Wooding, & Gregor, 2009). This study area is also located in the Fynbos biome but is dominated by the Swartland Alluvium Fynbos (SAF) veld type, ranging from the Elandskloof Mountains to the Limiet Mountains near Wellington. The SAF veldtype is known to be the wettest type of alluvium fynbos that is characterised by an average annual rainfall of between 320-980mm. The area receives rain from May to August (Rebelo et al., 2006). The region is also known to be the alluvium type of fynbos with the widest range of ambient temperatures, varying from an absolute maximum of 43°C to an absolute minimum of 2°C (Fu, Chang, & Patricola, 2017). The veld is dominated by asteraceous fynbos i.e. 0.7-0.9m small-leaved proteoid and reed-like restioid species and is characterised as being an evergreen fine-leaved shrub land with lengths varying from low to moderate (Fu et al., 2017; Rebelo et al., 2006).

The plains zebras grazed on the natural vegetation until four to five months prior to harvesting. They were relocated from the veld camps to camps approximately 10ha in size, and at a stocking density of four zebras per camp. Available forage in the camps consisted primarily of Bermuda grass (*Cynodon dactylon*), and to a lesser extent oat grass (*Avena sativa*) and ryegrass (*Lolium perenne*). The zebras were not given any supplementary feed or licks throughout the year. During the dry season (November to February) the grasses in the camps start to die off and becomes very fibrous with a low digestibility and protein content by January (when the plains zebras were harvested). However, there were enough bulk left to sustain the zebras.

3.2.2 Plains zebra harvesting, dressing, and sampling

Harvesting and evisceration procedures were conducted in accordance with the standard operating procedure (Ethical clearance number: 10NP_HOF02). Animals from both study locations were harvested during the day from a secure hunting vehicle by a sharpshooter with an appropriate rifle. The animals were culled with headshots to limit wounding and carcass damage/wastage (Von La Chevallerie & Van Zyl, 1971). Followed by the shot, all the animals were exsanguinated immediately and loaded onto the back of a designated vehicle. The time and date culled and additional notes regarding the shooting incident were documented to note pre-harvesting stress. The animals were transported to a nearby field slaughtering facility on the farm. At the slaughtering facility, the animals

were loaded from the vehicle and weighed with a calibrated hanging scale to determine the exsanguinated, undressed (bled) weight defined in the study as the dead weight of the animal.

The external, followed by the internal, offal were removed and weighed separately as described by van Schalkwyk & Hoffman (2010). The heart and kidneys were weighed after the removal of the surrounding fat layer. After evisceration, the carcasses were halved along the spinal column and divided between the second and last rib into four quarters separating the carcasses into two hindquarters (left and right) and two forequarters (left and right). The warm carcass quarters were weighed separately before entering a mobile chiller. The carcasses were placed in a suspended manner in the mobile chiller approximately 45 minutes post-mortem at ±4°C and transported back to the Department of Animal Science at Stellenbosch University. Upon arrival, the carcasses were transferred to a chiller (±4°C) located in the meat processing laboratory.

Plains zebra harvested at Prinskraal had a ~72-hour refrigeration period and plains zebras harvested at Elandsberg Nature Reserved had a ~24-hour refrigeration period before any carcass/muscle measurements were recorded. Following the refrigeration period, the cold carcass weights of each quarter and the ultimate pH were recorded. The carcasses were deboned, and seven commercially important muscles were excised from the left and right side of the carcasses. The weights of each muscle for the eight winter and 12 summer harvested plains zebra stallions were recorded. The selected muscles included the fillets – *psoas major* (PM), two shoulder muscles – *infraspinatus* (IS) and *supraspinatus* (SS), three hindquarter muscles – *semimembranosus* (SM), *biceps femoris* (BF) and *semitendinosus* (ST), and lastly the *longissimus thoracis et lumborum* (LTL) which extends from the forequarter to the hindquarter along the vertebral column. The LTL were excised separately into the *longissimus thoracis* (LT) located in the forequarter and the *longissimus lumborum* (LL) located in the hindquarter.

3.2.3 Statistical Analysis

The statistical analysis was conducted using Statistica 64 version 13.4 (2018) VEPAC model and Microsoft Excel (2016). Descriptive statistics were used as an indication of the expected variation for the carcass yields, offal yields and muscle yields per season. An exponential regression analysis was performed to determine the relationship between the offal yields and age for plains zebras harvested in the summer season as well as between the proportional contribution of each muscle and cold carcass weight from both seasons. The contribution for each selected muscle to the cold carcass weight from both seasons was pooled and analysed by means of univariate analysis of variance (ANOVA), with muscles as a fixed effect and animals as a random effect. A 5 % significance level was used.

3.3 RESULTS

Only observations on the carcass, offal, and muscle yields between the two seasons could be made due to the descriptive statistical method used to quantify the data. The ages of the winter group were unknown whilst that of the 12 stallions from the summer group were known as they were cull stallions from the Quagga breeding project; the latter allows for a comparison of the effect of age (even though the numbers are low) on the yields. Additionally, the plains zebra harvested in the winter season were

classified as physically mature adults over the age of three years (Mentis, 1972). The known ages for animals harvested in the summer season ranged from two to 13 years.

3.3.1 Carcass yields

Table 3.1 presents the carcass parameters of plains zebra stallions harvested during two seasons in the Western Cape Province of South Africa. The dead weight of animals harvested in the winter had a mean of 324.4 ± 5.55 kg with a minimum of 304.8 kg and a maximum of 353.7 kg. Animals in the summer season were found to have a numerically lower mean dead weight of 291.5 kg ± 11.65 kg with a minimum of 234.4 kg and a maximum of 347.8 kg. The winter harvesting group had a mean warm carcass weight of 193.1 ± 3.95 kg and a cold carcass weight of 188.3 ± 4.03 kg. As expected, the warm (168.9 ± 5.79 kg) and cold carcass weights (164.5 ± 5.53 kg) for the summer harvesting group were also found to be numerically lower than the winter harvesting group due to the lower dead weight found for the summer group. The winter harvesting group had a warm dressing percentage of 59.5 ± 0.55 and a cold dressing percentage of 58.0 ± 0.60. The summer harvesting group had a warm dressing percentage of 58.1 ± 0.68 and a cold dressing percentage of 56.6 ± 0.70. Both harvesting groups had a moisture loss between the warm and cold dressing percentage of ~1.5 %.

A regression was calculated to determine the exponential relationship between the carcass characteristics and age of the summer harvesting group. The coefficient of determination (R^2-value) and the exponential equation for all the carcass parameters are presented in Table 3.2. The dead, warm and cold carcass weights had weak R^2-values of 0.2311, 0.2369 and 0.2460, respectively. The warm and cold dressing percentage also had a weak R^2-values of 0.0378 and 0.0583, respectively. This indicates that the correlation between the carcass yields and age was low.

Table 3.1 Mean (±standard error) and the range of carcass yields from eight plains zebra stallions harvested in the winter season and 12 plains zebra stallions harvested in the summer season.

		Winter harvest (n=8)		Summer harvest (n=12)	
Carcass parameter		**Mean ± SE**	**Range**	**Mean ± SE**	**Range**
Intact carcass weight	**kg**	324.4 ± 5.55	304.8 – 353.7	291.5 ± 11.65	234.4 – 347.8
Warm carcass weight	**kg**	193.1 ± 3.95	176.0 – 210.1	168.9 ± 5.79	139.3 – 190.8
Cold carcass weight	**kg**	188.3 ± 4.03	169.6 – 205.0	164.5 ± 5.53	136.1 – 185.5
Warm dressing percentage	**%**	59.5 ± 0.55	57.7 – 2.0	58.1 ± 0.68	54.9 – 63.2
Cold dressing percentage	**%**	58.0 ± 0.60	55.6 – 60.7	56.6 ± 0.70	53.3 – 61.2

Abbreviations: SE= standard error

Table 3.2 The coefficient of determination (R^2-value) and the exponential equation to indicate the relationship between the respective carcass parameters and age of plains zebra stallions (aged between 2 and 13 years) harvested during a summer season in the Western Cape Province of South Africa.

Carcass component	**Exponential equation**	**R^2-value**
Dead weight	$y = 262.39e^{0.0167x}$	0.2311
Cold carcass weight	$y = 150{,}58e^{0.0142x}$	0.2369
Warm carcass weight	$y = 154.05e^{0.0148x}$	0.2460
Warm dressing percentage	$y = 58.709e^{-0.002x}$	0.0378
Cold dressing percentage	$y = 57.386e^{-2e-04x}$	0.0583

y = carcass component; x = age

3.3.2 Offal yields

The mean external and internal offal weights (kg) and its contribution to the dead weight (%) for plains zebra stallions harvested in the winter and summer season are presented in Table 3.3. Included in the external offal is the head, legs, skin and tail and was found to be numerically similar for the winter (41.980 ± 0.853 kg) and summer harvest (41.950 ± 1.334 kg) which contributed 13.0 ± 0.23 % and 14.5 ± 0.29 % to the dead weight, respectively. The filled gastrointestinal tract (GIT), liver, heart, trachea, lungs, kidneys, and spleen made up the internal offal component of the carcass. The internal offal of plains zebras harvested in the winter season weighed 70.760 ± 3.113 kg and contributed 21.8 ± 0.89 % to the dead weight. The summer harvest group had a numerically lower internal offal weight of 66.132 ± 3.782 kg with an almost similar contribution of 22.5 ± 0.50 % to the dead weight. The total skin weight and contribution hereof to the dead weight of the plains zebra harvested in the winter season (22.288 ± 0.533 kg; 6.9 ± 0.19 %) were found to be lower than for the plains zebras harvested in the summer season (24.046 ± 0.927 kg; 8.3 ± 0.18 %). The liver weight was found to be numerically similar for the plains zebras harvested in the winter (3.300 ± 0.130 kg) and summer season (3.320 ± 0.109 kg) which contributed 1.0 ±0.04 % and 1.2 ± 0.05 % to the dead weight, respectively. The kidneys had a numerically similar mean contribution for both seasons (0.2 ± 0.01 %) and weighed 0.740 ± 0.030 kg and 0.648 ± 0.032 kg for the winter and summer harvest, respectively. The total offal for the winter harvesting group was 112.740 ± 3.424 kg and for the summer was 108.081 ± 4.933 kg. The total offal

comprised 34.8 ± 0.94 % and 37.1 ± 0.44 % of the dead weight for the winter and summer harvesting group, respectively.

Table 3.4 represents the coefficient of determination (R^2-value) and the equation for an exponential regression drawn for the proportion of the offal (%) over the age of the 12 plains zebras in the summer harvesting group. Only 6.50 % (R^2 = 0.0650) of the variation in the total offal can be attributed to the exponential relationship with age.

Table 3.3 Mean (± standard error), minimum and maximum of offal yields from eight plains zebra stallions harvested in the winter season and 12 plains zebra stallions harvested in the summer season.

Carcass components	Winter harvest (n=8)			Summer harvest (n=12)		
	Mean ± SE (kg)	Mean ± SE (%[1])	Range (kg)	Mean ± SE (kg)	Mean ± SE (%[1])	Range (kg)
Dead weight	324.4 ± 5.55		304.8-353.7	291.5 ± 11.65		234.4-347.8
Head[2]	12.606 ± 0.322	3.9 ± 0.05	11.70-14.45	12.046 ± 0.344	4.17 ± 0.13	10.30-14.06
Legs	6.725 ± 0.182	2.1 ± 0.04	6.000-7.550	5.408 ± 0.159	1.867 ± 0.04	4.500-6.100
Skin	22.288 ± 0.533	6.9 ± 0.19	20.800-25.100	24.046 ± 0.927	8.3 ± 0.18	18.350-29.200
Tail[3]	0.362 ± 0.040	0.1 ± 0.01	0.241-0.600	0.449 ± 0.029	0.2 ± 0.01	0.314-0.645
Total external offal	41.98 ± 0.853	13.0 ± 0.23	39.39-46.39	41.95 ± 1.334	14.5 ± 0.29	34.08-49.93
GIT	60.150 ± 3.300	18.6 ± 0.97	48.000-76.200	56.050 ± 3.547	19.1 ± 0.54	37.450-74.850
Liver	3.300 ± 0.130	1.0 ± 0.04	2.750-3.750	3.320 ± 0.109	1.2 ± 0.05	2.600-3.900
Heart	1.788 ± 0.093	0.6 ± 0.03	1.350-2.100	1.727 ± 0.084	0.6 ± 0.02	1.309-2.266
Trachea & Lungs	3.850 ± 0.458	1.2 ± 0.13	2.600-6.000	3.417 ± 0.187	1.2 ± 0.06	2.500-4.550
Kidneys	0.740 ± 0.030	0.2 ± 0.01	0.661-0.904	0.648 ± 0.032	0.2 ± 0.01	0.487-0.860
Spleen	0.933 ± 0.043	0.3 ± 0.01	0.776-1.113	0.969 ± 0.074	0.3 ± 0.07	0.631-1.586
Total internal offal	70.760 ± 3.113	21.8 ± 0.89	59.393-86.558	66.132 ± 3.782	22.5 ± 0.50	46.453-86.360
Total offal	112.74 ± 3.424	34.8 ± 0.94	100.44-131.02	108.08 ± 4.933	37.1 ± 0.44	80.63-136.29

[1]Parameter as a percentage of the dead weight

[2]Head: Measured without skin and with tongue

[3]Tail: Measured without skin

Abbreviations: SE= standard error, GIT = Gastrointestinal tract

Table 3.4 The coefficient of determination (R^2-value) and the exponential equation to indicate the relationship between the respective offal components and age of plains zebra stallions (aged between 2 and 13 years) harvested during a summer season in the Western Cape Province of South Africa.

Offal components	Exponential equation	R^2-value
Head	$y = 4.2378e^{-3E-04x}$	0.0180
Legs	$y = 1.8479e^{0.0001x}$	0.0060
Skin	$y = 8.4970e^{-4E-04x}$	0.0767
Tail[3]	$y = 0.1588e^{-6E-04x}$	0.0244
Total external offal	$y = 14.754e^{-3E-04x}$	0.0494
GIT	$y = 18.078e^{0.0007x}$	0.1153
Liver	$y = 1.2215e^{-1E-03x}$	0.1092
Heart	$y = 0.5669e^{0.0006x}$	0.0784
Trachea & Lungs	$y = 1.0555e^{0.0014x}$	0.1731
Kidneys	$y = 0.2369e^{-1E-03x}$	0.2238
Spleen	$y = 0.3643e^{-0.002x}$	0.1472
Total internal offal	$y = 21.594e^{0.0006x}$	0.1350
Total offal	$y = 36.419e^{0.0002x}$	0.0650

y = offal proportion; x = age

3.3.3 Muscle yields

The mean muscle weights (kg) and their contribution to the cold carcass weight (%) for plains zebra stallions harvested in the winter and summer season are presented in Table 3.5. The mean weight is the combination of the muscles on the left and right side of the carcass. The muscles weights of the winter harvest group were numerically higher than the summer harvest group. The LTL muscle was found to have the highest ($p < 0.001$) contribution of 3.9 ± 0.39 % and 3.3 ± 0.30 % to the cold carcass weight for the winter and summer season, respectively. The SS muscle was found to have the lowest contribution and contributed numerically similar to the cold carcass weight for both the winter (0.4 ± 0.06 %) and summer (0.4 ± 0.01 %) seasons. The total mean contribution to cold carcass weight of all seven muscles was 11.0 ± 0.43 % and 9.9 ± 0.16 % for the winter and summer group, respectively.

A regression was calculated to determine the relationship between the muscle yields (kg and %) and the cold carcass weights for both harvesting groups. The coefficient of determination (R^2-value) and the exponential equation for all the muscle yields are presented in Table 3.6. The variation of the total muscle weight relative to the cold carcass weight taken into consideration by the equation was 78.61 % ($R^2 = 0.7861$).

Figure 3.1 represents the mean proportion (%) of each individual muscle to the mean cold carcass weight (176.4 kg) of 20 plains zebra stallions. The LTL (3.5 %), SM (1.6 %), BF (2.7 %) and ST (0.9 %) differed significantly from one another. The LTL muscle had the highest contribution followed by the BF and then SM. The proportion of the IS (0.6 %), SS (0.4 %) and PM (0.6 %) to the cold carcass weight were non-significant.

Table 3.5 Muscle yields from eight plains zebra stallions harvested in the winter and 12 plains zebra stallions harvested in the summer season. The results are reported as Means (± standard error), minimum and maximum of the muscle weight (kg) and contribution (%) to the cold carcass weight.

Carcass components	Winter harvest (n=8)			Summer harvest (n=12)		
	Mean ± SE (kg)	Mean ± SE (%[1])	Range (kg)	Mean ± SE (kg)	Mean ± SE (%[1])	Range (kg)
Cold carcass weight	188.3 ± 4.03		169.6-205.0	164.5 ± 5.53		136.1-185.5
LTL	7.455 ± 0.829	3.9 ± 0.39	4.675-11.025	5.410 ± 0.298	3.3 ± 0.10	3.825-7.275
SM	3.178 ± 0.093	1.7 ± 0.03	2.875-3.600	2.642 ± 0.120	1.6 ± 0.06	1.800-3.275
BF	5.271 ± 0.188	2.8 ± 0.07	4.625-6.075	4.215 ± 0.142	2.6 ±0.05	3.475-4.928
ST	1.819 ± 0.106	1.0 ± 0.04	1.425-2.200	1.490 ± 0.049	0.9 ± 0.02	1.300-1.725
IS	1.069 ± 0.053	0.6 ± 0.03	0.925-1.350	0.971 ± 0.042	0.6 ± 0.02	0.775-1.200
SS	0.841 ± 0.122	0.4 ± 0.06	0.600-1.675	0.633 ± 0.031	0.4 ± 0.01	0.475-0.800
PM	1.084 ± 0.039	0.6 ± 0.02	0.900-1.200	0.902 ± 0.044	0.6 ± 0.03	0.600-1.250
Total	20.72 ± 1.117	11.0 ± 0.43	16.77-24.15	16.26 ± 0.581	9.9 ± 0.16	13.53-19.68

[1]Parameter as a percentage to the cold carcass weight

Abbreviations: LTL= *longissimus thoracis et lumborum*, BF= *biceps femoris*, SM = *semimembranosus*, ST= *semitendinosus*, IS = *infraspinatus*, SS = *supraspinatus*, PM= *psoas major,* SE= standard error

Table 3.6 The coefficient of determination (R^2-value) and the exponential equation of the muscle yields drawn over the cold carcass weights for 20 plains zebras harvested in the winter and summer season.

Carcass components	Weight		Proportional contribution	
	Exponential equation	R^2-value	Exponential equation	R^2-value
LTL	$y = 0.8205e^{0.0114x}$	0.6501	$y = 1.3448e^{0.0054x}$	0.2914
SM	$y = 1.0904e^{0.0055x}$	0.4316	$y = 1.7871e^{-5E-04x}$	0.0067
BF	$y = 1.4203e^{0.0067x}$	0.7466	$y = 2.3278e^{0.0007x}$	0.0338
ST	$y = 0.5349e^{0.0063x}$	0.6431	$y = 0.8766e^{0.0003x}$	0.0043
IS	$y = 0.4640e^{0.0044x}$	0.3564	$y = 0.7605e^{-0.002x}$	0.0653
SS	$y = 0.1905e^{0.0074x}$	0.3213	$y = 0.3123e^{0.0014x}$	0.0170
PM	$y = 0.5438e^{0.0033x}$	0.1434	$y = 0.8913e^{-0.003x}$	0.1011
Total	$y = 4.494e^{0.0079x}$	0.7861	$y = 7.3654e^{0.0019x}$	0.1685

y = muscle yield or proportion; x = cold carcass weight

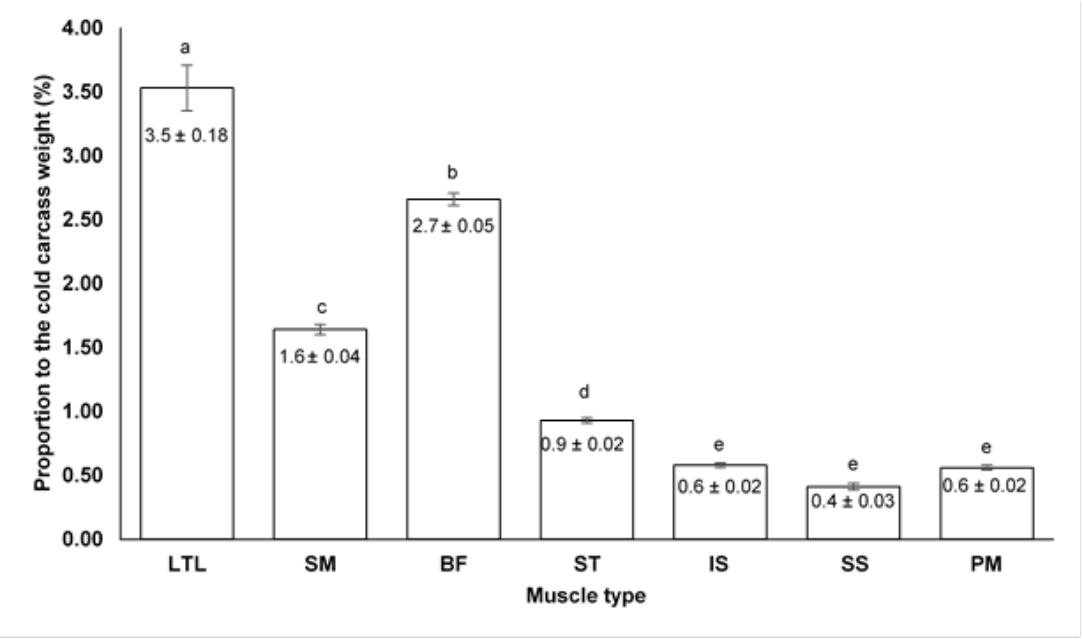

Figure 3.1 The percentage contribution (mean ± SE) of the *longissimus thoracis et lumborum* (LTL), *semimembranosus* (SM), *biceps femoris* (BF), *semitendinosus* (ST), *infraspinatus* (IS), *supraspinatus* (SS) and *psoas major* (PM) to the average cold carcass weight of plains zebra stallions harvested during two seasons in the Western Cape Province of South Africa.

3.4 DISCUSSION

3.4.1 Carcass yields

It was observed that the animals harvested in the summer season (291.5 ± 11.65 kg) had a lower dead (bled) weight than the animals harvested in the winter season (324.4 ± 5.55 kg; Table 3.1). The differences in dead carcass weight was reflected in the lower dressed carcass yields (warm and cold) observed for the summer-harvested animals (warm = 168.9 ± 5.79 kg; cold = 164.5 ± 5.53) compared to the winter-harvested animals (warm = 193.1 ± 3.95; cold = 188.3 ± 4.03 kg). Information on the carcass weight of the plains zebra is limited to two studies, i.e. Onyango et al., (1998) and Hoffman, Geldenhuys, & Cawthorn (2016). Both these studies reported lower carcass yields compared to the current study, which may potentially be attributed to the dry season the animals were harvested in. The lower carcass yields reported for the summer-harvested animals in the current study could be due to seasonal differences in the diet, harvest location and forage behaviour, altering the plain of nutrition and as a result the body composition. In the Western Cape Province, the rainy season is in the winter between May and August and the dry summer months between November and February. During the wet season, the availability of forage increases in biomass and consists of leafy grass with high nutrient concentrations. In contrast, the biomass during the dry season is reduced and the plains zebras are dependent on fibrous grass species with low nutritional quality (Owen-Smith, Le Roux, & Macandza, 2013). Additionally, the plains zebras harvested in the winter season, in this study, had access to a supplement lick during the dry months to restore depleted minerals. Furthermore, the plains zebra also tends to consume more forage in the rainy season, than in the dry season, due to natural grazing being

more abundant, leading to more grass bites per step taken (Havarua, Turner, & Mfune, 2014). This results in an increase of fat deposition in the rainy season and as observed in this study, the dead and dressed carcass weights were heavier for the plains zebras harvested in the winter season. Visually, it was noted in this study that the winter-harvested plains zebra carcasses were characterised by a higher degree of subcutaneous fat deposition, when compared to that of the summer-harvested plains zebras. However, it needs to be kept in mind that the difference in level of subcutaneous fat deposition may potentially also be ascribed to the mixed age composition of the summer-harvested group (i.e. sub-adults and adults), whereas the winter-harvested group consisted of only adult animals

Historically, meat-producing horses were slaughtered at the end of their working career, and only recently this practice was changed with foals being slaughtered for consumption (Tateo, De Palo, Ceci, & Centoducati, 2008). The slaughter of meat-producing horses is thus based on age which have a beneficial influence on carcass characteristics, with season not considered as these horses are provided with artificial feed year-round irrespective of the location or season (Lorenzo et al., 2014). In this study, to investigate the effect of age on dead, warm and cold carcass weights, and dressing percentage for plains zebra, an exponential regression analysis was carried out. A low coefficient of determination ($R^2 \leq 0.24$) was reported for all the carcass factors, indicating that less than 24 % of the variation in carcass yields can be ascribed to the influence of animal age. The low R^2 value potentially indicate that the summer-harvested animals already achieved maximum growth and thus were in the plateau phase on a typical sigmoidal curve at the point of harvest. Mentis (1972) found that free-roaming plains zebra reach their mature body weight at the age of 36 months. The youngest animal in this study was 24 months, and the advanced growth may potentially be ascribed to the fact that these animals were part of the Quagga project, i.e. would have received additional feed when required and thus not have been subjected to the challenges of seasonal forage availability and quality that free-roaming plains zebras experience. In this study thus, the differences found between the two groups can most probably be ascribed to the effect of the harvesting season on the subcutaneous fat deposition and consequently body composition.

To the best of our knowledge, there is little information available on the meat production potential of plains zebra, therefore findings in this study will be compared to related information about commercial horse meat breeds and/or donkey breeds. Specialised breeds for horse meat production such as the Burguete (675 kg) and Hispano-Bretón (HB) (715 kg) breed were found to have heavier adult body weights than the plains zebras. Higher slaughter and carcass weights were reported for the Burguete breed at 16 months (411.3 kg and 275.5 kg, respectively) (Sarriés & Beriain, 2005) and 24 months (395 kg and 258.9 kg, respectively) and the HB breed at 24 months (406 kg and 275.5 kg, respectively) (Juárez et al., 2009). Heavier slaughter and carcass weights were reported for the Sanfratellano breed (411 kg and 243.75 kg) slaughtered at 18 months. The Haflinger breed also slaughtered at 18 months has a comparable slaughter weight (349.83 kg) and carcass weight (207.83 kg) to the plains zebras in this study (Lanza, Landi, Scerra, Galofaro, & Pennisi, 2009). The plains zebra is heavier than Martina Franca donkeys (101-181 kg) between the ages of 8 and 15 months (Polidori, Cavallucci, Beghelli, & Vincenzetti, 2009; Polidori et al., 2008; Polidori et al., 2015) and domesticated donkeys found in tropical regions in Africa (such as Botswana), Central America and Asia (<150 kg)

(Aganga, Aganga, Thema, & Obocheleng, 2003). Compared to the meat-producing horse and donkey breeds, the plains zebras in this study are intermediate in terms of intact-, warm- and cold carcass weights.

Dressing percentage is an important parameter when the meat production potential of an animal or species is investigated (Van Zyl, Von La Chevallerie, & Skinner, 1969). In game animals, the dressing percentage is the proportion of the dressed weight relative to the undressed/dead carcass (bled) weight. Game animals cannot be fasted before harvesting and therefore may have a higher gut fill than domestic livestock species that are typically fasted in lairage for 24 hours before slaughter. Comparable results for the dressing percentage for both the winter (59.5 ± 0.55 %) and summer season (58.1 ± 0.68 %) were found in this study. The difference observed is attributed to the higher carcass yields observed and the higher subcutaneous fat level noted in the winter harvesting group. A lower dressing percentage of 56 % for plains zebra was reported by Onyango et al., (1998), however, the sample size of the study was very small (n=2) and animals were harvested in very dry, semi-arid conditions, with these conditions that may have contributed to the low carcass yields and dressing percentages reported.

Meat-producing horses typically yield high dressing percentages and dressing percentages, when compared to other domestic livestock species. The dressing percentage of the plains zebras in this study were higher than that reported for the HB x Galician Mountain crossbreed (52.8 %) and the Galician Mountain breed (50.3 %) (Franco, Crecente, Vázquez, Gómez, & Lorenzo, 2013), and are comparable to that of the Haflinger (59.6 %) and Sanfratellano horse breeds (59.3 %) (Lanza et al., 2009). However, horse breeds such as the Burguete (Juárez et al., 2009; Sarriés & Beriain, 2005), HB (Juárez et al., 2009), Italian Heavy Draft horse breed (Tateo, De Palo, Padalino, & Centoducati, 2016) and horses slaughtered in Poland (Litwińczuk, Florelk, Skałecki, & Litwińczuk, 2008) had higher dressing percentages (>63 %) than the plains zebras harvested in this study. The studies reporting high dressing percentages can potentially be ascribed to a low gut fill as a result of the animals subjected to pre-slaughter fasting, which subsequently resulted in higher dressing percentages. Another factor that can possibly influenced the dressing percentage reported for plains zebra in this study, is the degree of blood loss before weighing, which potentially resulting in a higher calculated dressing percentage. The dressing percentage of the plains zebras harvested in both seasons compared favourably to that of Martina Franca donkeys (49.2 – 57.5 %) found in various studies (Aganga, Aganga, Thema, & Obocheleng, 2003; Polidori, Vincenzetti, Cavallucci, & Beghelli, 2008; Polidori, Pucciarelli, Ariani, Polzonetti, & Vincenzetti, 2015).

When comparing the dressing yield of the plains zebra with other game species commonly found in South Africa, the dressing percentage compared favourably with impala (58.0 %;Hoffman, 2000), springbok (56 %; Hoffman & Mcmillin, 2009), blesbok (50.6-53.7 %, Hoffman & Mcmillin, 2009; Hoffman, Smit, & Muller, 2008) and large-bodied game species such as the greater kudu (58.3 %; Hoffman, Mostert, Kidd, & Laubscher, 2009), gemsbok (54 %; Onyango et al., 1998), eland (50.8-51 %; Hoffman & Mcmillin, 2009; Needham, Laubser, Kotrba, Bureš, & Hoffman, 2019), black wildebeest (53.19%; Hoffman, van Schalkwyk, & Muller, 2009) and African savannah buffalo (48-53 %; Hoffman, Hildebrandt, & Leslie, 2018). However, higher dressing percentages for male springbok (64.9%),

blesbok (62.9 %) and impala (63.4 %) have been reported by Van Zyl & Ferreira (2004). The differences in dressing percentages can be attributed to seasonal and location differences influencing the body composition of these species (Neethling, Hoffman, & Britz, 2014).

The dressing percentages of game animals are also comparable with domestic livestock species such as cattle, sheep, and goats. The dressing percentage of the plains zebra compares favourably to that of Nguni (52.1 %), Bonsmara (56.9 %) and Aberdeen Angus (53.7 %) cattle (Muchenje, Dzama, Chimonyo, Raats, & Strydom, 2008). The plains zebra values in this regard also compared favourably to sheep breeds such as the South African Mutton Merino (SAMM; 41.5 %), Dormer (44.2 %) (Cloete, Hoffman, Cloete, & Fourie, 2004), fat-tailed indigenous Damara (59.9 %) and also to goats such as Boer goats and indigenous goats (55.7 %; Tshabalala, Strydom, Webb, & De Kock, 2003).

3.4.2 Offal yields

Game animals produce by-products that are edible and non-edible. Edible by-products include tripe, liver, heart, lungs, kidneys, and spleen; and external offal which include the head, tail, and feet/lower legs (McCrindle, Siegmund-Schultze, Heeb, Zárate, & Ramrajh, 2013). Edible offal is marketed by informal suppliers and forms part of the traditional diet in South Africa (McCrindle et al., 2013) with a per capita consumption of 4.7 kg and 4.8 kg for the year of 2009 and 2013 respectively (Erasmus & Hoffman, 2017). Information on the quantity of edible offal of game species is limited and needs to be investigated since edible offal is a low-cost protein and nutrient-rich food source that has the potential to contribute to food security (Hoffman, Laubscher, & Leisegang, 2013; McCrindle et al., 2013). Non-edible offal, in game species, such as the skin and horns (as well as the head after removal of the cheek and other meat/muscles) are primary products collected during trophy hunting and are of high economic value in the game industry (McCrindle et al., 2013; Taylor, Lindsey, & Davies-Mostert, 2016). The plains zebra is a hindgut fermenter and lacks horns, unlike some game species, and should therefore rather be compared with other equine species in terms of head and GIT weights. Information on offal yields in horses and donkeys, however, is limited.

In this study, the highest contributing external offal component to the dead carcass weight in plains zebras harvested in both locations, was the skin followed by the head (Table 3.3). Presently, low quantities of plains zebra meat enter the formal market value chain in South Africa due to plains zebras being primarily hunted by recreational hunters for their skins with the meat seen as a secondary product. However, large numbers of zebras are culled by professional teams for the utilisation of their skin and meat. The meat is typically exported to the European Union as plains zebras are excluded from the FMD control regulation (Uys, 2015). It is important for the industry to note the contribution of the skin and head (as well as the legs under the knee joint with the hooves) to the carcass weight as these body parts remain attached to the carcass during transportation to the processing plants. This enables the marketers to achieve a top price for the skin since the skin on the head is also flayed with that of the body and lower legs. Another reason for keeping the skin on the carcass during transportation and chilling, is to reduce moisture loss since only a few game species have a subcutaneous fat layer to minimise this loss. However, in this study, the head and skin were removed prior to transportation and

chilling and the head of the plains zebras from both locations was weighed without the skin as the skin was flayed according to industry standards. A lower skin weight and contribution to the dead weight for animals harvested in the winter season (22.288 ± 0.533 kg; 6.9 ± 0.19 %) were found than for animals harvested in the summer season (24.046 ± 0.927 kg; 8.3 ± 0.18 %). The lower skin weight of animals harvested in the winter than in the summer can potentially be attributed to different skinning teams that were used for each season. A professional skinning team were used for the winter harvest, and a non-professional skinning team were used for the summer harvest, which may have resulted in variations in the subcutaneous fat left on the skin, thus influencing the contribution of the skin to the dead weight.

The head weight and its contribution to the dead weight were found to be very similar for the winter- (12.606 ± 0.322 kg; 3.9 ± 0.05 %) and summer-harvested animals (12.046 ± 0.344 kg; 4.2 ± 0.13 %). A weak relationship between animal age and head and skin weight, as evident in the low R^2 value obtained with the exponential regression analysis, is indicative of the animals that were mature at the point of harvest, which supports the conclusion for the carcass yields. These findings agree with the observations of González et al. (2006) that in male horses slaughtered in México the skin contributed 7.4 % and the head 4.5 % to the live weight (277.8 kg). The slaughter weight of these horses fell in the range of the dead weight of the plains zebras harvested in the summer season (Table 3.1). Catalan crossbreed donkeys with a live weight of more than 151 kg were found to have a similar head and lower skin contribution to the empty body weight (live weight – gastrointestinal content) than the plains zebras in the current study (Hernández-Briano et al., 2018). The lower skin contribution observed in the study of Hernández-Briano et al. (2018) can be postulated to be due to the skin weight not including the skin on the head and legs, as the study did not provide a detailed methodology.

When compared to smaller game species, the proportional weight of the skin of the plains zebra from both seasons was higher than found for male impala (3.9 %) and male fallow deer (6.6 %) with an dead weight of 49.9 kg and 47.4 kg, respectively (Fitzhenry et al., 2019; Hoffman, 2000). The skin weight and contribution can also be compared to large-bodied game species which will have heavier skins and body weights such as eland bulls (20.3 kg and 6.6 %) with an dead weight of 305.4 kg (Needham et al., 2019) and blue wildebeest bulls (13.6 – 17.9 kg and 7.9-8.4 %) weighing 168.8-208.2 kg (Van Heerden, 2018).

In terms of the internal offal, animals harvested in the winter season (70.760 ± 3.113 kg; 21.8 ± 0.89 %) produced heavier or similar yields when compared to animals harvested in the summer season (66.132 ± 3.782 kg; 22.5 ± 0.50 %). The GIT was the heaviest component measured for internal offal weight and had the highest contribution to the dead weight of animals harvested in the winter (60.150 ± 3.300 kg; 18.6 ± 0.97 %) and summer season (56.050 ± 3.547 kg; 19.1 ± 0.54 %). The lower GIT weight reported for the summer-harvested animals can potentially be attributed to a lower gut fill, since it has been shown by the exponential regression that the effect of age had a weak effect on the GIT proportion (R^2 = 0.1153; Table 3.4). It has been found that in the rainy (winter) season the plains zebra consumes more forage (by counting the grass bites per step taken) as a result of the natural grazing being more abundant (Havarua et al., 2014).

The liver represents a valuable source of vitamin A, vitamin B_1, and nicotinic acid. The liver and kidneys contain higher levels of iron, copper, and zinc than skeletal muscle, (Lawrie & Ledward, 2006),

making it a valuable low-cost protein source especially for the rural communities of South Africa (Hoffman et al., 2013). However, recently offal is rather processed into niche-market products that are seen as delicacies in upmarket restaurants (Hoffman et al., 2013). The liver weight and contribution to total carcass yield in the winter season (3.300 ± 0.130 kg; 1.0 ± 0.04 %) were comparable with that found for the summer season (3.320 ± 0.109 kg; 1.2 ± 0.5 %). The kidney weights in the winter season (0.740 ± 0.030 kg; 0.3 ±0.01 % of carcass weight) were also comparable with that found for the summer season (0.648 ± 0.032 kg; 0.2 ± 0.03 %). The contribution of the liver weight to the empty body weight (live weight – gastrointestinal content) in Catalan crossbreed donkeys with a live weight of >151 kg were found to be similar to the plains zebras when calculated in the same manner (Hernández-Briano et al., 2018).

The liver weight as a proportion to carcass weight is comparable to that of impala (1.3 %; Hoffman, 2000), blesbok (1.3 %; Van Zyl & Ferreira, 2004) and blue wildebeest (1.0 %; Van Heerden, 2018). When compared to livestock species such as the South Africa Mutton Merino (SAMM; 0.99 kg) and Dormer (1.13 kg) lambs (Cloete et al., 2004), the plains zebra displayed heavier livers. However, the contribution to the dead weight was similar to SAMM (1.9 %) and Dormer (2.0 %) sheep breeds (Cloete et al., 2004). The proportional weight of the kidneys relative to the live weight are comparable with impala (0.3 %; Hoffman, 2000) fallow deer (0.3 %; Fitzhenry et al., 2019), blesbok (0.3 %; Van Zyl & Ferreira, 2004) and blue wildebeest (0.2 %; Van Heerden, 2018).

3.4.3 Muscle yields

The same primal meat cuts used to section beef carcasses are also used for the sectioning of game meat species and meat-horse breeds. The game meat trade however tends to sell muscles as a whole rather than meat cuts that are frequently made up out of two or more muscles. In this study, seven muscles including the LTL (loin), SM (topside), BF (silverside), ST (eye of the round), PM (fillet) and two additional representative shoulder muscles, the SS and IS, were removed from the carcass and weighed to determine each muscle's contribution to the overall meat yield in plains zebra. To the best of our knowledge, no information is available on the contribution of the respective muscles to the meat yield in plains zebra.

Recorded weight for all seven muscles for the winter-harvested animals were numerically heavier when compared to the weights recorded for the summer-harvested group (Table 3.5). Notable numerical differences between the winter and summer harvest can be observed for the LTL (7.455 ± 0.829 kg and 5.410 ± 0.298 kg, respectively), SM (3.178 ± 0.093 kg and 2.642 ± 0.120 kg, respectively) and BF (5.271 ± 0.188 kg and 4.215 ± 0.142 kg, respectively) muscles. The difference in weight between the two seasons can be attributed to the lower carcass yields observed for the summer-harvested group (Table 3.1), due to the real muscle yield being dependent on the weight at slaughter and consequently the carcass weight. The remainder of the seven muscles had almost numerically similar weights and proportions when compared between seasons. Age did not have a strong influence on carcass or organ yields observed for the summer-harvested animals, and therefore an exponential regression analysis was carried for each muscle weight and its calculated proportion to the cold carcass weight. The analysis generated high R^2 values for the LTL (0.6501), BF (0.7466) and ST (0.6431), however, it is

more of value to consider the proportional contribution of each muscle to the cold carcass weight. The R^2-values showed a weak increase with the cold carcass weight with 29.1 % (highest R^2-value) of the variation in the LTL and 0.4 % (lowest R^2-value) of the variation in the ST explained by the exponential relationship with the cold carcass weight. The results indicate that the proportional contribution of the muscles is relatively similar irrespective of the lower or higher carcass weights observed in this study.

The mean proportion (%) of each individual muscle to the mean cold carcass weight of the 20 plains zebras stallions were pooled and presented in Figure 3.1. Significant differences ($p < 0.001$) were found between the hindquarter muscles; the LTL (3.5 %), SM (1.6 %), BF (2.7 %) and ST (0.9 %) which also significantly differed from the smaller sized muscles, the IS (0.6 %), SS (0.4 %) and PM (0.6 %). The differences found between muscles in terms of contribution (and weight) can be attributed to their anatomical location and function. The LTL was found to be the highest contributing muscle which is expected since the LTL is an epaxial muscle with many bundle fibres extending over the vertebral column from the forequarter to the hindquarter (Frandson, Wilke, & Fails, 2013). The BF was the second-highest contributing muscle followed by the SM and then the ST. These muscles are the proximal muscles on the pelvic limb and are larger than the muscles on the shoulder joint due to their locomotive and hip extension functions (Frandson et al., 2013; Payne, Hutchinson, Robilliard, Smith, & Wilson, 2005). The PM muscle is a small muscle involved in the flexion of the hip and is one of the most tender muscles on a carcass due to its low functional activity resulting in small fibre diameters and less connective tissue. The PM muscle was found to have a similar contribution as the two stabilizing shoulder muscles; the IS and the SS which are respectively involved in the flexion and extension of the shoulder (Frandson et al., 2013).

The muscle yields of the plains zebra can be compared to game species and various horse breeds. However, data on individual muscle yields in various horse breeds is limited, since yields are represented as primal meat cuts consisting of two or more muscles and therefore cannot be compared. The LTL, SM, BF and ST in the plains zebra were found to have a lower contribution to the the total carcass weight, when compared to findings reported for blue wildebeest (4.8-5.0 %, 3.8-3.9 %, 4.8-5.0 % and 1.5 %, respectively; Van Heerden, 2018) and fallow deer (7.5 %, 5.2 %, 6.1 % and 1.6 %, respectively; Fitzhenry et al., 2019). The LTL muscle had a higher contribution, the SM a lower contribution, the BF a higher contribution and the ST were found to have a similar contribution to the cold carcass weight when compared to eland (2.6 %, 2.2 %, 2.4 % and 0.9 %; Needham et al., 2019). Higher percentages for the IS and SS muscle were reported for fallow deer (1.3 % and 1.1 %, respectively; Fitzhenry et al., 2019), eland (0.7 % and 0.7 %, respectively; Needham et al., 2019) and blue wildebeest (1.3-1.4 % and 1.1-1.2 %, respectively; Van Heerden, 2018); results that may be related to a more active behaviour, e.g. like jumping more frequently by fallow deer and eland.

3.5 CONCLUSION

The study is first of its kind to provide baseline information on the carcass, offal and muscle yield potential of plains zebras harvested in two localities in the Western Cape Province of South Africa. The study included animals that were harvested during two seasons to establish a potential influence of seasons on the carcass characteristics of plains zebra. Carcass dressing percentage compared

favourably and in agreement with other equine species, game species and livestock species such as cattle and sheep. The considerable contribution of the edible by-products in this study and the high muscle yield, indicate that the plains zebra can potentially be used as a valuable protein source.

Further studies are warranted to determine the full production potential and economic viability of the plains zebra. Future studies should investigate forequarter and hindquarter commercial meat cuts in terms of weight and percentage to the cold carcass weight for a better comparison to horse, donkey, and cattle commercial meat cuts. Studies on the meat and bone yield and meat-to-bone ratios and percentages are also warranted, for there is no information available in literature on these aspects. Other factors that also need to be investigated include, the influence of production systems, season, ages, and sex on the meat production potential of plains zebra. As plains zebra meat is exported regularly to the EU it will be of benefit to establish the physical meat quality as well as nutritional and organoleptic attributes of the meat to promote further marketing as an alternative/substitute meat product.

3.6 REFERENCES

Aganga, A. A., Aganga, A. O., Thema, T., & Obocheleng, K. O. (2003). Carcass Analysis and Meat Composition of the Donkey. *Pakistan Journal of Nutrition*, *2*(3), 138–147. https://doi.org/10.3923/pjn.2003.138.147

Bothma, J. du P. (2011). Burchell's or plains zebra (*Equus burchelli*). *Game & Hunt*, 8–11.

Cloete, J. J. E., Hoffman, L. C., Cloete, S. W. P., & Fourie, J. E. (2004). A comparison between the body composition, carcass characteristics and retail cuts of South African Mutton Merino and Dormer sheep. *South African Journal of Animal Sciences*, *34*(1), 44–51. https://doi.org/10.4314/sajas.v34i1.4040

Cooper, S. M., & Van der Merwe, M. (2014). Game ranching for meat production in marginal African agricultural lands. *Journal of Arid Land Studies*, *24*(1), 249–252.

Du Toit, C. J. L., Meissner, H. H., & van Niekerk, W. A. (2013). Direct greenhouse gas emissions of the game industry in South Africa. *South African Journal of Animal Sciences*, *43*(3), 340–361. https://doi.org/10.4314/sajas.v43i3.8

Erasmus, S. W., & Hoffman, L. C. (2017). What is meat in South Africa? *Animal Frontiers*, *7*(4), 71. https://doi.org/10.2527/af.2017.0449

Estes, R. D. (2012). Zebra, asses, and horses: family equidae. In *The behaviour guide of African mammals: including hoof mammals, carnivores, primates* (20th Anniv, pp. 235–248). London, England: University of California Press, Ltd.

Fitzhenry, L. B., Cawthorn, D. M., Muchenje, V., Bureš, D., Kotrba, R., & Hoffman, L. C. (2019). Carcass composition and yields of wild fallow deer (*Dama dama*) in South Africa. *African Journal of Wildlife Research*, *49*(1), 100–110. https://doi.org/10.3957/056.049.0100

Franco, D., Crecente, S., Vázquez, J. A., Gómez, M., & Lorenzo, J. M. (2013). Effect of cross breeding and amount of finishing diet on growth parameters, carcass and meat composition of foals slaughtered at 15 months of age. *Meat Science*, *93*(3), 547–556. https://doi.org/10.1016/j.meatsci.2012.11.018

Frandson, R. D., Wilke, W. L., & Fails, A. D. (2013). *Anatomy and Physiology of Farm Animals* (7th ed.). Wiley-Blackwell.

Fu, D., Chang, P., & Patricola, C. M. (2017). Intrabasin Variability of East Pacific Tropical Cyclones during ENSO Regulated by Central American Gap Winds. *Scientific Reports*, *7*(1), 189–198. https://doi.org/10.1038/s41598-017-01962-3

González, H. G., Mota-Rojas, D., Becerril-Herrera, M., Casas-García, C., Zermeno, M., Ramírez-Necoecha, R., … Alonso-Spilsbury, M. (2006). Horse meat for human consumption in México: slaughter performance and carcass morphometry. *Journal of Food Technology*, Vol. 4, pp. 106–110.

Hack, M. A., East, R., Rubenstein, D. I., & Gray, E. (2002). Status and action plan for the plains zebra (*Equus burchellii*). In P. D. Moehlman (Ed.), *Equids: Zebras, Asses and Horses. Status Survey and Conservation Action Plan* (pp. 43–60). IUCN/SSC Equid Specialist Group. IUCN, Gland, Switzerland and Cambridge UK.

Harley, E. H., Knight, M. H., Lardner, C., Wooding, B., & Gregor, M. (2009). The Quagga project : progress over 20 years of selective breeding. *South African Journal of Wildlife Research*, *39*(2), 155–163. https://doi.org/10.3957/056.039.0206

Havarua, Z., Turner, W. C., & Mfune, J. K. E. (2014). Seasonal variation in foraging behaviour of plains zebra (Equus quagga) may alter contact with the anthrax bacterium (*Bacillus anthracis*). *Canadian Journal of Zoology*, *92*(4), 331–337. https://doi.org/10.1139/cjz-2013-0186

Hernández-Briano, P., Ramírez-Lozano, R. G., Carrillo-Muro, O., López-Carlos, M. A., Méndez-Llorente, F., & Aguilera-Soto, J. I. (2018). Gender and live weight on carcass and meat characteristics of donkeys. *Ciencia Rural*, *48*(4), 1–7. https://doi.org/10.1590/0103-8478cr20170889

Hoffman, L. C. (2000). The yield and carcass chemical composition of impala (*Aepyceros melampus*), a southern African antelope species. *Journal of the Science of Food and Agriculture*, *80*(6), 752–756.

Hoffman, L. C., Geldenhuys, G., & Cawthorn, D. M. (2016). Proximate and fatty acid composition of zebra (*Equus quagga burchellii*) muscle and subcutaneous fat. *Journal of the Science of Food and Agriculture*, *96*(11), 3922–3927. https://doi.org/10.1002/jsfa.7623

Hoffman, L. C., Hildebrandt, W. R., & Leslie, A. J. (2018). Chemical Composition of African Savanna Buffalo (Syncerus caffer) Meat. *African Journal of Wildlife Research*, *48*(1), 013003. https://doi.org/10.3957/056.048.013003

Hoffman, L. C., Laubscher, L. L., & Leisegang, K. (2013). Nutritional value of cooked offal derived from free-range rams reared in South Africa. *Meat Science*, *93*(3), 696–702. https://doi.org/10.1016/j.meatsci.2012.11.041

Hoffman, L. C., & Mcmillin, K. W. (2009). Improving the meat quality of venison and other exotic game. *Improving the Sensory and Nutritional Quality of Fresh Meat*, (1994), 447–475. https://doi.org/10.1533/9781845695439.3.447

Hoffman, L. C., Mostert, A. C., Kidd, M., & Laubscher, L. L. (2009). Meat quality of kudu (*Tragelaphus strepsiceros*) and impala (*Aepyceros melampus*): Carcass yield, physical quality and chemical composition of kudu and impala Longissimus dorsi muscle as affected by gender and age. *Meat Science*, *83*(4), 788–795. https://doi.org/10.1016/j.meatsci.2009.08.022

Hoffman, L. C., Muller, M., Schutte, D. W., Calitz, F. J., & Crafford, K. (2005). Consumer expectations, perceptions and purchasing of South African game meat. *South African Journal of Wildlife Research*, *35*(1), 33–42.

Hoffman, L. C., Schalkwyk, S. van, & Muller, N. (2009). Effect of season and gender on the physical and chemical composition of Black Wildebeest (*Connochaetus gnou*) meat. *South African Journal of Wildlife Research*, *39*(2), 170–174. https://doi.org/10.3957/056.039.0208

Hoffman, L. C., Smit, K., & Muller, N. (2008). Chemical characteristics of blesbok (*Damaliscus dorcas phillipsi*) meat. *Journal of Food Composition and Analysis*, *21*(4), 315–319. https://doi.org/10.1016/J.JFCA.2007.12.003

Juárez, M., Polvillo, O., Gómez, M. D., Alcalde, M. J., Romero, F., & Valera, M. (2009). Breed effect on carcass and meat quality of foals slaughtered at 24 months of age. *Meat Science*, *83*(2), 224–228. https://doi.org/10.1016/j.meatsci.2009.04.022

Lanza, M., Landi, C., Scerra, M., Galofaro, V., & Pennisi, P. (2009). Meat quality and intramuscular fatty acid composition of Sanfratellano and Haflinger foals. *Meat Science*, *81*, 142–147. https://doi.org/10.1016/j.meatsci.2008.07.008

Lawrie, R. A., & Ledward, D. A. (2006). Meat and human nutrition. In *Lawrie's Meat Science* (pp. 342–357). https://doi.org/10.1016/B978-0-08-100694-8.00003-0

Litwińczuk, A., Florelk, M., Skałecki, P., & Litwińczuk, Z. (2008). Chemical composition and physiochemical properties of horse meat from the *Longissimus lumborum* and *semitendinososus* muscle. *Journal of Muscle Foods*, *19*(3), 223–236. https://doi.org/10.1111/j.1745-4573.2008.00117.x

Lorenzo, J. M., Sarriés, M. V., Tateo, A., Polidori, P., Franco, D., & Lanza, M. (2014). Carcass characteristics, meat quality and nutritional value of horsemeat: A review. *Meat Science*, *96*(4), 1478–1488. https://doi.org/10.1016/j.meatsci.2013.12.006

McCrindle, C. M. E., Siegmund-Schultze, M., Heeb, A. W., Zárate, A. V., & Ramrajh, S. (2013). Improving food security and safety through use of edible by-products from wild game.

Environment, Development and Sustainability, *15*(5), 1245–1257. https://doi.org/10.1007/s10668-013-9436-2

Meissner, H. H., Hofmeyr, H. S., Van Rensburg, W. J. J., & Pienaar, J. P. (1983). Classification of livestock for realistic prediction of substitution values in terms of a biologically defined Large Stock Unit. *Technical Communication*, (175), 1–40.

Mentis, M. T. (1972). *Ungulate management on private land in Natal*. Natal Parks, Game and Fish Preservation Board, Pieter-maritzburg, Natal, R.S.A.:

Muchenje, V., Dzama, K., Chimonyo, M., Raats, J. G., & 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*(1), 20–28. https://doi.org/10.1016/J.MEATSCI.2007.07.026

Mucina, L., & Rutherford, M. C. (2006). The vegetation of South Africa, Lesotho and Swaziland. *Strelitzia 19*, 1–30. https://doi.org/10.1007/s

Needham, T., Laubser, J. G., Kotrba, R., Bureš, D., & Hoffman, L. C. (2019). Sex influence on muscle yield and physiochemical characteristics of common eland (*Taurotragus oryx*) meat. *Meat Science*, *152*, 41–48. https://doi.org/10.1016/j.meatsci.2019.02.008

Neethling, J., Hoffman, L. C., & Britz, T. J. (2014). Impact of season on the chemical composition of male and female blesbok (*Damaliscus pygargus phillipsi*) muscles. *Journal of the Science of Food and Agriculture*, *94*(3), 424–431. https://doi.org/10.1002/jsfa.6281

Oberem, P., & Oberem, P. (2016). Species accounts. In *The New Game Rancher*. Biza Publications.

Onyango, C. A., Izumimoto, M., & Kutima, P. M. (1998). Comparison of some physical and chemical properties of selected game meats. *Meat Science*, *49*(1), 117–125. https://doi.org/10.1016/S0309-1740(97)00116-2

Owen-Smith, N., Le Roux, E., & Macandza, V. (2013). Are relatively rare antelope narrowly selective feeders? A sable antelope and zebra comparison. *Journal of Zoology*, *291*(3), 163–170. https://doi.org/10.1111/jzo.12058

Payne, R. C., Hutchinson, J. R., Robilliard, J. J., Smith, N. C., & Wilson, A. M. (2005). Functional specialisation of pelvic limb anatomy in horses (Equus caballus). *Journal of Anatomy*, *206*(6), 557–574. https://doi.org/10.1111/j.1469-7580.2005.00420.x

Polidori, P., Cavallucci, C., Beghelli, D., & Vincenzetti, S. (2009). Physical and chemical characteristics of donkey meat from Martina Franca breed. *Meat Science*, *82*(4), 469–471. https://doi.org/10.1016/j.meatsci.2009.03.001

Polidori, P., Pucciarelli, S., Ariani, A., Polzonetti, V., & Vincenzetti, S. (2015). A comparison of the carcass and meat quality of Martina Franca donkey foals aged 8 or 12 months. *Meat Science*, *106*, 6–10. https://doi.org/10.1016/j.meatsci.2015.03.018

Polidori, P., Vincenzetti, S., Cavallucci, C., & Beghelli, D. (2008). Quality of donkey meat and carcass characteristics. *Meat Science*, *80*(4), 1222–1224. https://doi.org/10.1016/j.meatsci.2008.05.027

Rebelo, A. G., Boucher, C., Helme, N., Mucina, L., & Rutherford, M. C. (2006). Fynbos Biome. In L. Mucina & M. C. Rutherford (Eds.), *The vegetation of South Africa, Lesotho, and Swaziland* (pp. 53–219). South African National Biodiversity Institute, Pretoria.

Sarriés, M. V., & Beriain, M. J. (2005). Carcass characteristics and meat quality of male and female foals. *Meat Science*, *70*(1), 141–152. https://doi.org/10.1016/j.meatsci.2004.12.006

Scoones, I., Bishi, A., Mapitse, N., Moerane, R., Penrith, M. L., Sibanda, R., … Wolmer, W. (2010). Foot-and-mouth disease and market access: challenges for the beef industry in southern Africa. *Pastoralism*, *1*(2), 135–164. https://doi.org/10.3362/2041-7136.2010.010

StatsSA. (2008). Statistical release: Mid-year population estimates July 2008. Retrieved from http://www.statssa.gov.za/publications/P0302/P03022008.pdf

StatsSA. (2018). Statistical release: Mid-year population estimates July 2018. Retrieved from http://www.statssa.gov.za/publications/P0302/P03022018.pdf

Tateo, A., De Palo, P., Ceci, E., & Centoducati, P. (2008). Physicochemical properties of meat of Italian Heavy Draft horses slaughtered at the age of eleven months. *Journal of Animal Science*, *86*(5), 1205–1214. https://doi.org/10.2527/jas.2007-0629

Tateo, A., De Palo, P., Padalino, B., & Centoducati, P. (2016). Quality of carcasses in I.H.D.H. foals reared in the province of Bari (Italy). *Italian Journal of Animal Science*, *4*, 418–420. https://doi.org/10.4081/ijas.2005.2s.418

Taylor, A., Lindsey, P., & Davies-Mostert, H. (2016). An assessment of the economic social and conservation value of the wildlife ranching industry and its potential to support the green economy in South Africa. The Endagered Wildlife Trust. Johannesburg, South Africa. Retrieved from https://www.sagreenfund.org.za/wordpress/wp-content/uploads/2016/04/EWT-RESEARCH-REPORT.pdf

Tshabalala, P. A., Strydom, P. E., Webb, E. C., & De Kock, H. L. (2003). Meat quality of designated South African indigenous goat and sheep breeds. *Meat Science*, *65*(1), 563–570. https://doi.org/10.1016/S0309-1740(02)00249-8

Uys, G. (2015). Game exports after FMD. Retrieved August 25, 2019, from Farmer's weekly website: https://www.farmersweekly.co.za/agri-business/agribusinesses/game-exports-after-fmd/

Van Heerden, A. M. (2018). *Profiling the meat quality of blue wildebeest (Connochaetes taurinus)*. Unpublished Master's thesis in Animal Science, Stellenbosch, South Africa.

Van Schalkwyk, D. L., & Hoffman, L. C. (2010). *Guidelines for the harvesting of game for meat export 2010*. Windhoek, Namibia: Ministry of Environment & Tourism.

Van Schalkwyk, D. L., & Hoffman, L. C. (2016). *Guidelines for the harvesting & processing of wild game in Namibia 2016.* Ministry of Environment & Tourism.

Van Zyl, J. H. M., Von La Chevallerie, M., & Skinner, J. D. (1969). A note on the dressing percentage in the springbok (*Antidorcas marsupialus* (*Zimmermann*) and impala, (*Aepyceros melampus*).

Proceedings of the South African Society for Animal Production, *9*, 199–200.

Van Zyl, L., & Ferreira, A. V. (2004). Physical and chemical carcass composition of springbok (*Antidorcas marsupialis*), blesbok (*Damaliscus dorcas phillipsi*) and impala (*Aepyceros melampus*). *Small Ruminant Research*, *53*(1–2), 103–109. https://doi.org/10.1016/j.smallrumres.2003.08.017

Von La Chevallerie, M., & Van Zyl, J. H. M. (1971). Some effects of shooting on losses of meat and meat quality in springbok and impala. *South African Journal of Animal Science*, *1*, 113–116.

CHAPTER 4

INFLUENCE OF MUSCLE TYPE ON THE PHYSICAL MEAT QUALITY CHARACTERISTICS OF THE PLAINS ZEBRA (*Equus quagga*)

ABSTRACT

The study reports on the physical quality characteristics (pH_u, drip loss, cooking loss, Warner-Bratzler shear force, CIE colour coordinates) of plains zebra meat obtained from animals that were culled at two distinctly different location in the winter rainfall region of the Western Cape Province of South Africa. Six skeletal (*Longissimus thoracis et lumborum*/LTL, *semimembranosus*/SM, *biceps femoris*/BF, semitendinosus/ST, *infraspinatus*/SS, *supraspinatus*/SS) muscles were removed from a total of 20 carcasses, of which eight were obtained during the winter cropping session, and 12 during the summer cropping session, respectively. The pH_u and drip loss percentage of samples obtained from winter-harvested animals were significantly higher, whilst the cooking loss percentage, shear force values, CIE L*, CIE b* and hue-angle were significantly higher for the summer-harvested animals. All physical meat quality traits were influenced by muscle type. The IS (5.83) and SS (5.89) had the highest pH_u, and the LTL and SM were characterised by the lowest pH_u (5.54). Drip loss was the most pronounced in the three primal muscles, the LTL (1.50 %), SM (1.65 %) and BF (1.43 %). The SS muscle was characterised by the lowest drip loss (0.78 %); however, this muscle also demonstrated the highest cooking loss (40.24 %). For tenderness, meat samples obtained from the winter-harvested animals (52.22 N) were intermediate in terms of toughness, compared to samples from summer-harvested animals (63.76 N) where the toughness value exceeded the upper shear forced limit for tender meat. The ST (51.58 N) and SS (46.11 N) obtained from the winter-harvested animals, and the IS (49.68 N) obtained from the summer-harvested animals, were intermediate in terms of toughness. The remainder of the respective muscles obtained from both groups were characterised by shear force values higher than the suggested tenderness value. All colour measurements recorded for all six muscles as well as both groups corresponded to the intermediate range associated with game meat. Although significant differences were reported for certain meat quality traits in this study, it is debatable whether these differences will be of interest to consumers.

Keywords: Plains zebra, Game meat, Season, Surface colour, Tenderness

4.1 INTRODUCTION

Physical meat quality characteristics features prominently in determining consumer perception during purchasing, and acceptance of a product following consumption. Quality parameters evaluated prior to consumption can be categorised as “intrinsic” and “extrinsic” quality cues; with “experience” and “credence” quality attributes after consumption (Vermeulen, Schönfeldt, & Pretorius, 2015). The after-consumption experience will have a profound effect on re-purchasing behaviour. Intrinsic cues and experience attributes relate to the physical characteristics of a product, which may include amongst others visual attributes and organoleptic properties. Extrinsic cues include characteristics that are not physically related to the product, and include amongst others the price, brand name and meat label information. Credence attributes of a meat product are related to the welfare, slaughter age, traceability, and wholesomeness of a meat product, and cannot be fully evaluated by the consumer during purchasing or after consumption (Vermeulen et al., 2015).

Physical characteristics of red meat include aspects such as colour, moisture loss in the form of drip and during cooking, tenderness, and flavour (Honikel, 1998; Hughes, Oiseth, Purslow, & Warner, 2014). These aspects all influence consumer satisfaction, and thus ultimately the marketing potential of a product (Hughes, Oiseth, et al., 2014). Visual attributes such as the colour of raw meat products and the drip build up in packaged meat products when on the shelf in supermarkets or butcheries, are the two most important aspects that influence consumer decisions at the point of sale. Consumers’ associate meat colour with freshness and are attracted to bright red meat and will therefore be reluctant to buy meat with a dark red or pale colour (Hughes, Kearney, & Warner, 2014). Drip build up in packaged meat products is also associated with an unpleasant visual perception that is linked to inferior product quality (Hughes, Oiseth, et al., 2014). Additionally, the colour perceived and the degree of moisture loss, due to a low water-holding capacity, are of economic importance since unpleasant colour variations and decrease in the product weight will result in a low saleability and in turn economic losses (Hughes, Kearney, et al., 2014; Hughes, Oiseth, et al., 2014). Moisture loss during cooking and resulting tenderness and flavour of cooked meat are factors influencing the choice of the consumer to regularly purchase and/or consume the product (Bailey, 1972; Vermeulen et al., 2015).

Numerous studies on the physical meat quality of conventional livestock species such as cattle, sheep and pigs have been conducted, however, a limited number of studies were carried out on meat obtained from game species. Game meat is a popular choice of tourists visiting South Africa, and is positively perceived to be an indigenous, healthy, lean, and organic meat source (Hoffman & Wiklund, 2006). However, game meat is often assumed, due to an unattractive deep red colour, to be a tough and dry meat, which negatively impact on consumer choice (Hoffman, 2001; Hoffman, Muller, Schutte, Calitz, & Crafford, 2005). The colour of meat is determined by the chemical composition and myoglobin concentration present in the muscle, with the latter linked to the type and level of activity a muscle is responsible for (Lawrie & Ledward, 2006). The darker colour of game meat is postulated to be a result of pre-harvesting stress, and potentially also the fact that game animals are free-range and thus more active than domesticated livestock (Neethling, Suman, Sigge, Hoffman, & Hunt, 2017). The latter results

in a higher myoglobin concentration in the muscle, which is responsible for the darker meat colour of game animals (Hoffman, 2001; Hoffman, Mostert, Kidd, & Laubscher, 2009). Despite the assumption by consumers that game, in general, is a tough meat, several studies have reported on the intermediate to tender nature of game meat (Cawthorn et al., 2018; Hoffman, Diana, Muller, Van Rensburgh, & McMillin, 2019; Hoffman, Mostert, et al., 2009; Hoffman, Schalkwyk, & Muller, 2009).

South Africa is home to a wide variety of game species that are considered as acceptable meat-producing species, with meat from these species mostly purchased and processed by hunters as fresh raw meat cuts or as cured meat products such as biltong and droëwors (Hoffman & Cawthorn, 2012; Van der Merwe & Du Plessis, 2014). However, when the available literature on game meat quality is considered, to the best of our knowledge, no information on the meat quality characteristics and thus the meat production potential of the plains zebra (*Equus quagga*) is available.

The plains zebra is a good candidate as a meat-producing species due to its adaptability to harsh environments, and non-susceptibility to the livestock threating foot-and-mouth disease (FMD) affecting cloven-hoofed animals. The spreading of this disease in South African led to the prohibition of game animal exports to the European Union (EU) in 2011. However, the prohibition did not apply to zebras as they are not susceptible to the disease, simultaneously promoting the export of this species. Prior to the prohibition, low numbers of 300-600 zebras were harvested for exporting purposes which increased to 2927 zebras harvested for exporting in 2016 (De Villiers C. pers. comm. 2018). With the increase in numbers, information on the physical meat quality of raw and cooked zebra meat will assist game producers in providing information on the suitability of plains zebra to be farmed for meat production purposes.

The aim of this study therefore is to determine the physical meat quality of the plains zebra, as determined by surface colour, drip loss, cooking loss and tenderness. As the plains zebra is classified as game, the effect of season was considered to determine the optimal time of harvesting for this species for meat purposes.

4.2 MATERIALS AND METHODS

4.2.1 Animals and study location

A total of twenty plains zebra stallions in total were harvested from which eight were harvested in the winter season June at Prinskraal farm near Bredasdorp in the Central Rûens Shale Renosterveld vegetation unit and 12 in the summer season January at Elandsberg Nature Reserve-Bartholomeus Klip near Hermon in the Swartland Alluvium Fynbos vegetation unit. Both study locations formed part of the fynbos biome in the Western Cape Province. Plains zebra harvested in the winter season (Prinskraal farm) were kept in ~800 ha camp shared with about 400 other game from various species whereas plains zebras harvested in the summer season (Elandsberg Nature Reserve) were kept in smaller camps of 10 ha with four zebras per camp. The zebras in both locations were free-roaming and in an extensive production system however, animals harvested on the Prinskraal farm had longer running distances due to the larger camp size. Animals from both locations were self-sustained and minimal human interference was needed by ensuring functional water points and mineral licks when

needed. Detailed information regarding the description of vegetation per location can be found in the Materials and Methods of Chapter 3.2.1.

4.2.2 Plains zebra harvesting, dressing, and sampling

The plains zebra stallions from both locations were harvested during the day in a similar manner with an appropriate rifle (Ethical clearance number: 10NP_HOF02). The carcasses were exsanguinated and then transported to the on-farm slaughtering facilities, and skinned, eviscerated, and dressed according to Van Schalkwyk & Hoffman (2016). The dressed carcasses were subsequently stored suspended in a mobile chiller at ±4°C and transported to the Department of Animal Science at the University of Stellenbosch for further processing analysis. The refrigeration period was ~72 hours and ~24 hours for the winter- and summer-harvested groups, respectively. After the refrigeration period, all carcasses were deboned, and specific muscles were excised from the left and right half of each carcass. For the season and muscle type comparison, six commercially important muscles were used which includes two shoulder muscles – *infraspinatus* (IS) and *supraspinatus* (SS), three hind limb muscles – *biceps femoris* (BF), s*emitendinosus* (ST) and *semimembranosus* (SM), and lastly the *Longissimus thoracis et lumborum* (LTL) from the forequarter-hindquarter section.

4.2.3 Physical Analysis

The pH was recorded by placing the probe in the centre of the cranial end of each muscle upon removal. One portion of ~80 g with a thickness of ~2.0 cm from each muscle was cut perpendicular to the muscle fibres for physical measurements such as surface colour, cooking loss and shear force. Another portion between approximately 60-100 g with a thickness of ~1.5 cm was removed for the drip loss measurement. The portion for physical analysis in the LTL, SM and BF were randomly assigned in relation to the ageing time points (Chapter 7). A portion in the mid-section was selected for physical measurements in the IS, SS and ST. The pH was measured with a Crison pH25 portable pH meter (Alella Barcelona, Spain) calibrated with two standard buffers at pH 4 and 7. The glass electrode, measuring the pH and temperature, was rinsed between each measurement with distilled water.

The water-holding capacity of each muscle was measured by determining the moisture loss using the drip bag method (Honikel, 1998). The meat sample allocated to drip loss from each muscle were weighed to determine the initial weight. The sample was hooked onto a wire and suspended in an inflated polyethene bag without touching the top, bottom, and sides of the bag. After the 24-hour suspension in the chiller (2-4°C), the samples were removed, blotted dry with absorbent paper, and weighed to determine the final weight for the calculation of the moisture loss percentage in the form of drip.

Cooking loss was determined for the meat samples allocated for the physical analysis. The ~2.0 cm raw steak of each muscle were weighed to determine the initial weight. After weighing, each steak sample was placed in a polyethene bag and submerged in a preheated water bath set to cook at 80°C for 60 minutes. After the 60 minutes, the steak samples were removed from the water bath and

the water that collected in the bag, removed. The steak samples were then placed in a refrigerator (±4°C) overnight to cool down. Following the cool-down period, the steak samples were removed from the refrigerator, blotted dry with absorbent paper, and weighed determining the final weight to calculate the percentage of moisture loss during cooking.
The drip and cooking loss percentages were separately determined by the following equation:

Moisture loss % = [(Initial weight – Final weight)/ (Initial weight)] *100

The cooked steaks were used to determine the instrumental tenderness by means of the force required to shear the cooked samples. The shear force was measured with an Instron Universal Testing Machine (Instron UTM, Model 2519-107) with a Warner-Bratzler blade fitting on a load cell of 2 kN. The blade fitting had a triangular opening with a base length of 13 mm and a perpendicular height of 15 mm programmed to shear at a crosshead speed of 200 mm/min

Six rectangular 1 cm x 1 cm x 2 cm samples from each cooked steak were cut parallel to the muscle fibres with a 1 cm double-bladed scalpel. Each of the rectangularly shaped samples was individually cut perpendicular to the muscle fibres. The force needed to shear perpendicular through the muscle fibres were measured in Newton. The WBSF for each muscle was determined by calculating the average force needed to shear the six samples.

The surface colour of the ~2.0 cm thick steak cut perpendicular to the muscle fibres was measured instrumentally per muscle with a calibrated Colour-guide 45°/0° colorimeter (BYK- Gardner GmbH, Gerestried, Germany). Each of the allocated steaks was left to bloom for ~30 minutes before five measurements at random positions on the surface of the meat were measured. The measurements taken by the colorimeter is in accordance with the CIE Lab colour system reporting CIE L* (lightness), CIE a* (red-green value), and CIE b* (blue-yellow) values (Honikel, 1998). The hue-angle and the chroma values were calculated from the CIE a* and CIE b* values with the following equations:

Hue-angle (h_{ab}) $= tan^{-1}\left(\frac{b^*}{a^*}\right)$

Chroma $\left(C^*\right) = \sqrt{(a^*)^2 + (b^*)^2}$

4.2.4 Statistical analysis

The statistical analysis was performed using a VEPAC model of Statistica 64 version 13.4 (2018). The experimental design was a mixed model of analysis of variance (ANOVA), with animal number as a random effect, and season and muscle type as fixed effects. For post-hoc testing, Fisher LSD was used. A normal probability plot was compiled for each characteristic to determine any deviations from normality and possible outliers. Statistical differences were considered significant at a probability level of 5% ($p \leq 0.05$). The data are reported as the LSMeans and standard error for each characteristic as per season and as per muscle type.

4.3 RESULTS

Interactions occurred ($p \leq 0.05$) between season and muscle type for the WBSF, CIE L* and hue-angle (Table 4.1, Figure 4.1a, b and c). Significantly higher WBSF values were reported for almost all muscles (except for the SM muscle) obtained from the summer-harvested animals, when compared to corresponding muscles obtained from the winter-harvested animals (Figure 4.1a). The IS (36.20 N) obtained from the winter-harvested animals were the most tender and both the LTL (71.39 N) and BF (74.59 N) obtained from the summer-harvested animals were the toughest. The CIE L* values for all the muscles were higher in the summer than in the winter harvested group, however, the differences observed were not always of significance. The plains zebra harvested in the summer season had higher CIE L* values for the LTL, IS and SS muscles. The two forequarter muscles the IS and SS did not differ significantly within a season but did differ ($p \leq 0.05$) between seasons. In both seasons, the LTL and ST were associated with being the two lightest/brightest muscles. The hue-angle values for all the muscles were significantly higher in the summer when compared to the same muscle from the winter season. Similar to the CIE L*, the LTL and ST were associated with having the highest hue-angle values (>39) (Figure 4.1c).

The remaining physical attributes (pH_u, drip loss percentage, cooking loss percentage, CIE a*, CIE b* and chroma) significantly differed ($p \leq 0.05$) between muscle types. Also, the effect of season was significant in all the attributes except in two colour measurements: CIE a* and chroma. When comparing season as an effect, the pH_u was higher in the winter (5.83) than in the summer season (5.53). The latter can also be attributed to the level of stress experienced per season and will be discussed further in section 5.5. The highest pH_u was observed in both the IS (5.83 ± 0.06) and SS (5.89 ± 0.06), with the lowest pH_u in the LTL (5.5 ± 0.04) and SM (5.5 ± 0.03) muscles. The mean pH_u values for both main effects were below six. The drip loss percentage was higher for the winter harvesting group (1.57 ± 0.10 %) than for the summer harvesting group (0.81 ± 0.08 %). The LTL, SM and BF muscles had the highest and the remaining muscles the lowest (< 1%) drip loss percentage. In terms of the seasonal effect, the cooking loss percentage was significantly higher in the summer than in the winter season, even though the numerical difference for the mean values was ~2.07 %. Between muscle types, the ST (39.35 ± 0.61 %) and SS (40.24 ± 0.50 %) had the highest cooking loss percentage and the LTL (35.00 ± 0.64 %) and IS (35.03 ± 0.60%) the lowest. In terms of colour, the smaller muscles types (ST, IS and SS) were measured to be redder as the CIE a* values were significantly higher than the larger muscles types (LTL, SM and BF). The ST, IS and SS had CIE a* values of ~14 and the lowest value was 12.76 measured on the LTL muscle.

Table 4.1 The level of statistical significance (p-values) for the main effects; season and muscle type of the physical meat quality parameters for plains zebra meat.

Parameter	Season	Muscle	Muscle x Season
pH_u	**< 0.001**	**< 0.001**	0.664
Drip loss %	**< 0.001**	**< 0.001**	0.37
Cooking loss %	**0.006**	**< 0.001**	0.609
WBSF (N)	**< 0.001**	**< 0.001**	**< 0.001**
L* (lightness)	**0.035**	**< 0.001**	**0.021**
a* (redness)	0.316	**< 0.001**	0.591
b* (yellowness)	**0.021**	**< 0.001**	0.072
Chroma	0.672	**< 0.001**	0.392
Hue-angle	**0.003**	**< 0.001**	**0.024**

Table 4.2 Physical meat quality characteristics (LSMeans ± SE) of six skeletal muscles obtained from adult male plains zebra harvested during winter and summer in the Western Cape Province of South Africa.

Main effect		pH_u	Drip loss %	Cooking loss %	WBSF (N)	CIE L*	CIE a*	CIE b*	Chroma	Hue-angle
Season	**Winter**	$5.83^{a} \pm 0.04$	$1.57^{a} \pm 0.10$	$36.84^{b} \pm 0.51$	$52.22^{b} \pm 2.38$	$32.13^{b} \pm 0.74$	14.07 ± 0.42	$10.32^{b} \pm 0.33$	17.47 ± 0.46	$36.17^{b} \pm 0.92$
	Summer	$5.53^{b} \pm 0.03$	$0.81^{b} \pm 0.08$	$38.91^{a} \pm 0.42$	$63.76^{a} \pm 1.95$	$34.30^{a} \pm 0.60$	13.50 ± 0.34	$11.39^{a} \pm 0.27$	17.72 ± 0.37	$40.22^{a} \pm 0.75$
Muscle type	**LTL**	$5.54^{cd} \pm 0.04$	$1.50^{a} \pm 0.19$	$35.00^{c} \pm 0.64$	$62.98^{b} \pm 1.76$	$35.23^{a} \pm 0.35$	$12.76^{c} \pm 0.27$	$11.03^{b} \pm 0.20$	$16.94^{c} \pm 0.30$	$41.09^{a} \pm 0.56$
	SM	$5.54^{d} \pm 0.03$	$1.65^{a} \pm 0.21$	$38.89^{b} \pm 0.47$	$64.50^{b} \pm 1.39$	$32.20^{c} \pm 0.31^{c}$	$13.23^{bc} \pm 0.18$	$10.47^{cd} \pm 0.19$	$16.91^{c} \pm 0.23$	$38.26^{b} \pm 0.40$
	BF	$5.62^{bc} \pm 0.05$	$1.43^{a} \pm 0.17$	$38.75^{b} \pm 0.49$	$68.57^{a} \pm 1.35$	$33.01^{b} \pm 0.25$	$13.62^{b} \pm 0.16$	$10.35^{cd} \pm 0.16$	$17.14^{c} \pm 0.20$	$37.08^{c} \pm 0.36$
	ST	$5.64^{b} \pm 0.05$	$0.86^{b} \pm 0.08$	$39.35^{ab} \pm 0.61$	$55.23^{c} \pm 0.98$	$34.92^{a} \pm 0.36$	$14.45^{a} \pm 0.21$	$12.47^{a} \pm 0.15$	$19.15^{a} \pm 0.20$	$40.91^{a} \pm 0.50$
	IS	$5.83^{a} \pm 0.06$	$0.90^{b} \pm 0.13$	$35.03^{c} \pm 0.60$	$42.94^{d} \pm 1.38$	$32.01^{c} \pm 0.31$	$14.52^{a} \pm 0.19$	$10.62^{c} \pm 0.17$	$18.02^{b} \pm 0.23$	$36.13^{d} \pm 0.35$
	SS	$5.89^{a} \pm 0.06$	$0.78^{b} \pm 0.08$	$40.24^{a} \pm 0.50$	$53.70^{c} \pm 1.37$	$31.91^{c} \pm 0.32$	$14.13^{a} \pm 0.17$	$10.17^{d} \pm 0.15$	$17.44^{c} \pm 0.20$	$35.71^{d} \pm 0.36$

Abbreviations: LSMeans = Least square means, LTL= *Longissimus thoracis et lumborum*, BF= *biceps femoris*, SM = *semimembranosus*, ST= *semitendinosus*, IS = *infraspinatus*, SS = *supraspinatus*

[a-d] LSMeans with different superscripts in columns within main effects differ significantly at $p \leq 0.05$.

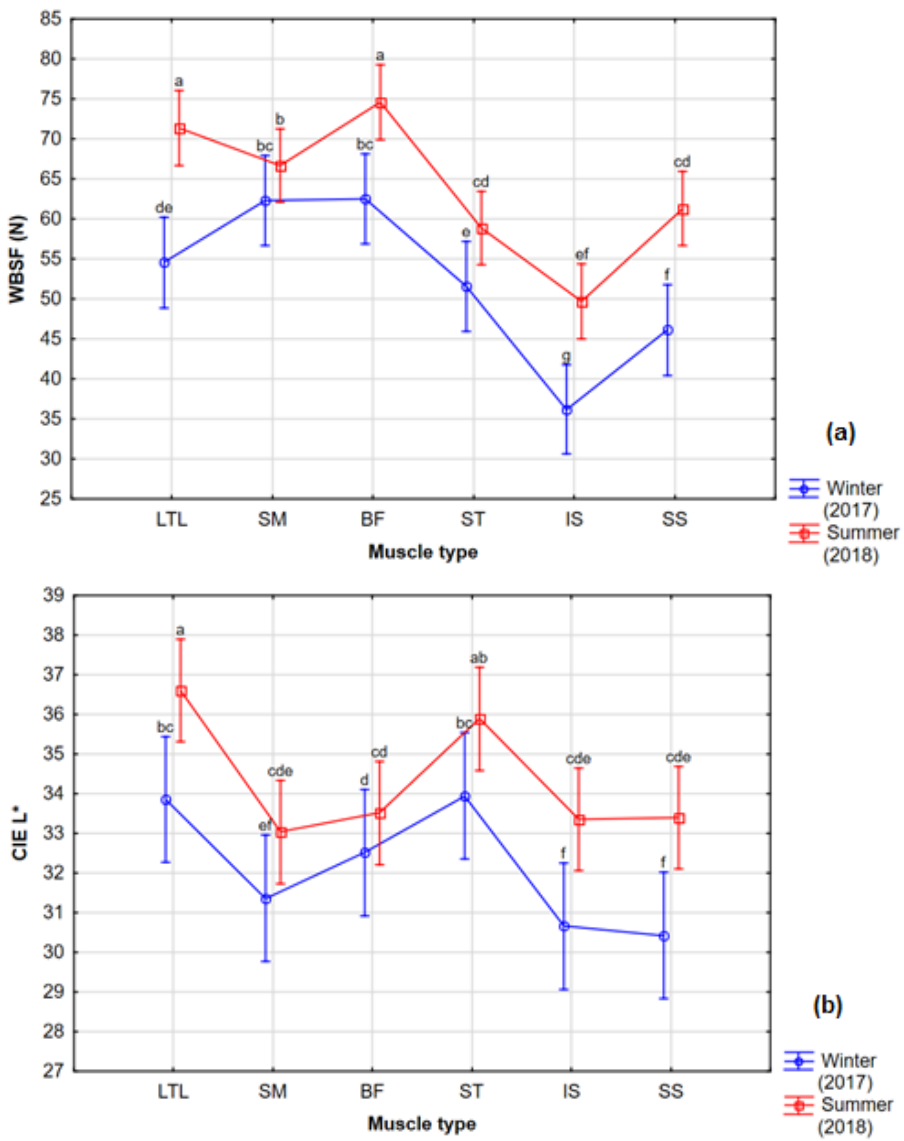

Figure 4.1 Interactions between season and muscle type for the WBSF (a), CIE L* (b) and hue-angle (c).

[a-g]LSMeans of the main effect with different superscripts differ significantly at $p \leq 0.05$.

LTL = *Longissimus thoracis et lumborum*, SM = *semimembranosus*, BF = *biceps femoris*, ST= *semitendinosus,* IS = *infraspinatus*, SS = *supraspinatus*

4.4 DISCUSSION

The aim of this study was to investigate the effect of season in which plains zebra stallions are harvested, on the physical meat quality characteristics of six specific skeletal muscles. The ultimate pH (pH_u) of the muscles was significantly influenced by the two main effects, i.e. season of harvest and muscle type (Table 4.1).

The variation in pH_u in the meat samples between the two seasons and six muscle types can potentially be ascribed to the glycogen content of the muscles at the point of harvest, with glycogen content that may vary considerably due to intrinsic and extrinsic factors (Lawrie & Ledward, 2006). Intrinsically, each individual muscle consists of a combination of red and white muscle fibres, which in turn is determined by the muscle's function and anatomical location (Cassens & Cooper, 1971). The location and function of a skeletal muscle play an important role in determining the quantity of glycogen that can be stored within the muscle (Kohn, Kritzinger, Hoffman, & Myburgh, 2005; Taylor, 2004). Extrinsically, the glycogen stored in each muscle type is influenced amongst others by the plane of nutrition and pre-harvest/slaughter activity and stress (Cassens & Cooper, 1971; Kohn et al., 2005).

The meat samples obtained in the winter season were found to have a significantly higher pH_u which fell into the higher end of the ideal range for an unstressed animal (5.5-5.8) when compared to those of the summer season (Immonen, Ruusunen, & Puolanne, 2000). The plains zebras harvested in the rainy (winter) season was on a higher plane of nutrition which is associated with higher availability of glycogen for post-mortem glycolysis entailing a relatively lower pH_u (Cassens & Cooper, 1971; Kohn et al., 2005). The unexpected relatively higher pH_u recorded can be attributed to the difference in farm layout between the two locations since the culling method for both seasons were similar and both over a period of two days (Chapter 3). The zebras from the winter harvest roamed in a relatively large 800 ha camp shared with about 400 other game animals whilst the zebras from the summer harvest roamed in multiple adjacent smaller 10 ha camps with 2-4 zebras per camp. Therefore, the high pH_u can be explained by the difference in camp size between the two farms and thus the higher daily activity of the zebra in the 800 ha camp in combination with the physical effort exploited due to stress during the hunting process. Contrasting to the summer harvest, when only one to two stallions were culled per camp, the stallions culled in the single 800 ha camp tended to flee together in their bachelor group from the hunting vehicle after every shot placement. This allowed all the selected stallions to flee together while simultaneously covering substantial longer running distances after each shot. Therefore, the winter group was more aware of the hunting vehicle resulting in extensive ante-mortem stress. The combination of extensive physical effort and stress of the winter harvest probably led to a rapid depletion of muscle glycogen stores, reducing the lactic acid formation, and therefore elevating the post-mortem pH_u as observed (Honikel, 2004; Lawrie & Ledward, 2006).

Regarding the muscle type comparison, this study reported a significant higher pH_u in the two small muscles measured in the forequarter (IS and SS). This indicates that both muscles had a decreased glycogen storage level than the remaining four muscles prior to harvest. The high pH_u found for these muscles is still acceptable as muscles with as $pH_u > 6.0$ are generally classified as being dark, firm, and dry (DFD) (Lawrie & Ledward, 2006; Shange, Gouws, & Hoffman, 2019). The pH_u found for

both main effects (5.54-5.89) (Table 4.2) was considered to be in the biologically normal range of 5.3-5.8 without having notable adverse effects on the meat quality such as any visual tendencies towards dark, firm and dry (DFD) meat despite the stress observed (Honikel, 2004; Shange et al., 2019). Studies on horse and donkey meat were primarily conducted on the LTL and hindquarter muscles, therefore, limiting data of the physical meat quality on forequarter muscles is available. It was, however, reported that horse meat muscles have an average pH_u < 6 (Gill, 2005) which may indicate that the higher pH_u found in this study is still acceptable as it did not exceed a mean pH_u of 6. The pH_u of the LTL, SM, BF and ST is comparable to that found for Italian Heavy Draft horses foals slaughtered at the age of 11 months (Tateo, De Palo, Ceci, & Centoducati, 2008) and Galician Mountain foals slaughter at 15 months of age (Lorenzo, Pateiro, & Franco, 2013). The pH_u of the LTL was also comparable to the LT muscle of Martina Franca donkeys (Polidori, Vincenzetti, Cavallucci, & Beghelli, 2008). In contrast, high pH_u values >6.0 for Martina Franca donkeys were observed by De Palo et al. (2017) compared to which the plains zebra's pH_u are favourably lower.

The water-holding capacity of plains zebra meat was found to be influenced by season of harvest and muscle type; however, no interaction was observed between season and muscle type for both drip and cooking loss percentages (Table 4.1). The water-holding capacity in the form of drip loss or cooking loss is an important factor to determine the percentage of weight loss of a meat product in the fresh and cooked form (Mostert & Hoffman, 2007). The water-holding capacity is also important to form a holistic opinion on the juiciness perceived during consumption as consumers' value succulent meat and often associate dry meat as being tough (Warriss, 2000). Factors influencing the water-holding capacity of meat is primarily the pH_u in relation with the isoelectric point of the meat, the rate of pH decrease, the muscle fibre type (Lefaucheur, 2010) and the level of intramuscular fat (Warriss, 2000).

A significant higher drip loss percentage was found in the meat for the winter harvest group in relation to the summer harvest group. This was unexpected due to the high pH_u reported for the winter harvest being above the isoelectric point (≥ 5.4 or 5.5) (Honikel, 2014; Huff-Lonergan, 2009). Proteins lose their ability to bind water as they start to denature at a pH_u below the isoelectric point, therefore negatively lowering the water holding capacity (Den Hertog-Meischke, Van Laack, & Smulders, 1997; Huff-Lonergan & Lonergan, 2005). In contrast, proteins are negatively charged above the isoelectric point which enhances their ability to bind water and as a result increasing the water-holding capacity (Huff-Lonergan & Lonergan, 2005). Even though the difference between seasons is nearly double, the difference found is still < 1 %. Therefore, although the drip loss percentage differed statistically between the seasons, the drip loss percentage may not necessarily differ notably with regards to physical meat quality attributes as experienced by consumers. The LTL and SM were reported to have a pH_u of 5.54 which is close to the isoelectric point and therefore as expected had the highest drip loss percentages compared to the other muscles (Miller, 2004). A high drip loss was also found in the BF muscle which did not significantly differ from the LTL and SM. This can be attributed to the fact that these muscles are large muscles with a higher surface area to volume ratios which results in the formation of more drip and also, being large muscles their cooling rate would have been slower than the smaller forequarter muscles resulting in higher drip loss (Van Heerden, 2018). Muscles with a high ratio of

glycolytic fibres or white fibres are known to have a high glycolytic capacity resulting in a lower pH_u. The SM is known to consist mostly out of glycolytic fibres and was found as expected to have a pH_u closer to the isoelectric point and thus the highest drip loss percentage in relation to the remaining muscles (Den Hertog-Meischke et al., 1997).

In general, the drip loss observed in this investigation was in the similar range for game species such as eland (Needham, Laubser, Kotrba, Bureš, & Hoffman, 2019), blue wildebeest (Van Heerden, 2018) and fallow deer (Cawthorn et al., 2018) although higher drip loss percentages were found for the blesbok (Neethling, Hoffman, & Britz, 2014). The drip loss percentage measured for the blesbok SM muscle (1.4 %) was lowest whilst the drip loss percentage measured for the plains zebra SM muscle (1.65 ± 0.21 %) was the highest. Similarly, to the plains zebra, Van Heerden (2018) also found in blue wildebeest that the SM had the highest drip loss and the IS and SS the lowest drip loss percentage. When compared with horse meat, the drip loss percentages of the plains zebra were generally lower than have been found for various horse breeds (Franco, Crecente, Vázquez, Gómez, & Lorenzo, 2013; Lorenzo, Crecente, Franco, Sarriés, & Gómez, 2014; Seong et al., 2016). Franco & Lorenzo (2014) found comparable drip loss percentage in the SM muscle for Galician Mountain and Galician Mountain x Hispano-Bréton crossbreed foals. However, similarly to blesbok, the drip loss for the SM in the latter study was found to the lowest when compared to the LTL, BF and ST.

The water-holding capacity of the meat derived from the plains zebras were further investigated by determining the quantity of water loss as a percentage to the initial weight during cooking. The cooking loss results did not coincide with that found for the drip loss (Table 4.2). In this study, the summer season was associated with the highest cooking loss, which was expected due to its low pH_u. This trend, however, differs to what was found for drip loss percentage which was the lowest for the summer season. This opposing trend can be also be seen with the effect of muscle type. The SS muscle was found to have the lowest drip loss percentage and the highest cooking loss percentage. In order to investigate the trend observed, a Pearson's correlation test was conducted and a negative but low correlation was calculated for the pooled drip and cooking loss values for plains zebra muscles ($r = -0.170$; $p = 0.051$); this could be linked to the fact that there is a limited amount of expressible/free water in any muscle sample which can be exuded via drip and/or cooking loss. The cooking loss percentage of muscles were found to be notably higher (approximately double) than reported for the Galician Mountain and Italian Heavy Draft horses (Lorenzo et al., 2013; Tateo et al., 2008).

At the expense of colour and taste/flavour, meat tenderness is seen as the most critical factor by consumers for the evaluation of the eating quality (Elzerman, Hoek, van Boekel, & Luning, 2011; Vermeulen et al., 2015). The main factors influencing meat tenderness related to muscle properties are the collagen and composition of muscle fibres whilst post-mortem factors include temperature, pH, sarcomere length and proteolysis, with species-related factors being growth rate and genotype (Maltin, Balcerzak, Tilley, & Delday, 2003). Significant differences were found between season and muscle type as well as an interaction between the two main effects for WBSF (Table 4.1). Meat is classified as being tender with as shear force value < 42.8 N and tough > 52.68 N. The winter harvest samples fell into the intermediate range of tenderness, although bordering on the tough classification whilst summer harvest fell above the suggested value for tough meat (Table 4.2). The winter season was found to have a lower

mean shear force value than the summer season which corresponds with the lower cooking loss as a high cooking loss is associated with less tender meat. The IS muscle in both seasons had a higher tenderness (lower WBSF value) which can be explained by the low moisture loss observed. The forequarter muscles are usually high in collagen causing these muscles to be tougher. The tenderness observed can be due to the measurement method used as the collagen did not form part of the sample sheared. Tenderness of meat is also subjective to the overall texture and grain of muscle itself which is influenced by the muscle fibre type and the size of the muscle fibre bundle. The muscle fibre type and bundle are in their turn related to the inherent function and intensity of muscle activity exploited by the individual muscle (Lawrie & Ledward, 2006). Large muscle fibre bundles are found in coarse-grained muscles such as the SM, while small muscle fibre bundles comprise fine-grained muscles such as the ST muscle (Lawrie & Ledward, 2006). Finely grained supportive muscles are usually more tender than muscles that are frequently used for exercise resulting to be stronger and therefore less tender (Sebsibe, 2008). The shear force values measured between the muscles of the plains zebras harvested in both seasons may be explained by the differences in skeletal muscle characteristics. All three primal muscles (LTL, SM and BF) except for the winter LTL was found to have a shear force value of >60 N. Similarities in shear force values could be drawn between the summer LTL and BF and also between the winter SM & BF. However, no significant differences were observed for the SM muscle with the season as an effect. The high shear force value obtained for the summer LTL can be attributed to the effect of stress over age seen in Figure 4.2. The latter figure is a representation of the shear force values obtained for the LTL for each individual zebra culled in the summer season in relation with their known age. There is an inverse relationship between meat tenderness and age with meat from older animals being perceived as less tender (Schönfeldt & Strydom, 2011; Segato, Cozzi, & Andrighetto, 1999). This is due to the amount of connective tissue as well as the formation of more insoluble or heat-stable collagen linkages with age (Badiani, Nanni, Gatta, Tolomelli, & Manfredini, 1997; Lawrie & Ledward, 2006). During the denaturation of this highly cross-linked intramuscular collagen, greater tension is thus created due to the collagen linkages being more thermally stable limiting the degree of shrinkage (Lawrie & Ledward, 2006). The figure illustrates the expected increase in toughness with age in the LTL muscle. However, a higher toughness was observed in two younger animals ~2-years and ~4-years old. Both animals were the last two to be culled on the second harvesting day. Although the mean pH_u of the summer group suggests an association with acute stress the animals could have experienced a degree of chronic stress as well. As both animals were in the near vicinity of the other animals harvested throughout the day (as well as the previous day) this could have contributed to the mobilisation of glycogen into the muscle itself, therefore, explaining the high shear force values observed. This is an example of where the effect of stress could be greater than the effect of age on meat tenderness. Meat tenderness has the tendency to decrease as the meat pH_u increases from 5.5 to 6.2. This phenomenon did not coincide with the results found in this study when season is considered as the meat tenderness of the winter season were generally lower than that from the summer season despite the pH_u. However, the shear force values obtained are still associated with tough meat.

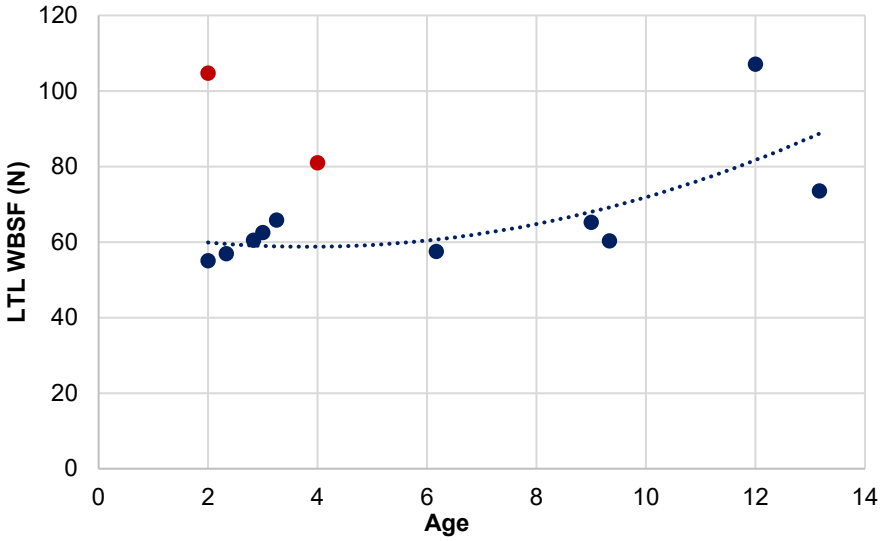

Figure 4.2 Warner Bratzler Shear Force (WBSF) of different aged plains zebra measured in the LTL (*Longissimus thoracis et lumborum*) muscle. (● Outlier animals due to stress).

Colour is an important parameter to be measured as it is the first attribute to influence the consumer's decision at the point of sale. The colour of meat is dependent on various factors and varies between species. Consumers tend to consider meat derived from game species as a single entity and classify the meat as being dark in colour when compared to that of domestic livestock species. The darker meat colour in game species can be explained by their natural higher physical activity levels as they are free-running animals (Hoffman et al., 2005). Consequently, this results in muscles containing a higher quantity of red oxidative fibres and thus a higher concentration of the pigment-containing compound myoglobin (Hoffman, 2001; Neethling, Hoffman, & Muller, 2016; Neethling et al., 2017). This is supported by the findings of Vestergaard et al. (2000) that physical activity has a greater impact on the meat colour than the diet itself with the higher physical activity resulting in a higher myoglobin concentration found within the muscle when compared to domestic livestock species (Hoffman, Kritzinger, & Ferreira, 2005; Kritzinger, Hoffman, & Ferreira, 2003). The latter is also applicable to muscles used for high endurance activities such as postural muscles (Taylor, 2004). The darker colour can also be explained by the fact that game species have a low intramuscular fat level and are also frequently exposed to higher levels of pre-slaughter/harvesting stress, thereby reducing the glycogen levels and thus elevating the pH_u and consequently resulting into meat leaning towards DFD. Shange, Gouws, & Hoffman (2019) reported that typical colour measurements for game meat at a "normal" pH can be described as CIE L* > 33, CIE a* > 13, CIE b* > ~10, chroma > 17 and hue-angle > 36 and DFD meat as CIE L* < 30, CIE a* < 11, CIE b* < 7, chroma < 13 and hue-angle < 32. Although this can be used as a guideline to compare colour measurements with, the authors did note that further research should be conducted as the inherent meat colour differs between various game species and their muscles.

The data obtained for all the colour measurements in this study fell into the normal to the intermediate range as described by Shange et al., (2019). As expected the colour parameters were

significantly influenced by muscle type due to the physiological differences between muscles, consequently, altering the pH_u (Brewer, Zhu, Bidner, Meisinger, & McKeith, 2001) and myoglobin concentration between muscles as well as the muscle fibre type ratio within and between different muscles types (Miller, 2002). The effect of season was observed in the CIE L*, CIE b* and hue-angle values. Significant interactions were also observed between the two main effects for CIE L* and hue-angle (Table 4.1). The CIE L* values measure the brightness of the muscle or meat sample in terms of light scattering. The latter is subjective to the structural attributes of the muscle itself related to the extent of protein denaturation which is enhanced by age which is again linked to muscle development (Hughes, Oiseth, et al., 2014). It was noted in a review by Lorenzo et al., (2014) that the colour of horsemeat is affected by age and they postulated that the CIE L* values decreased (muscles became darker) until a certain age had been reached, consequently following an increase in lightness again. To investigate the effect of age in the plains zebra, an exponential regression was drawn for the pooled CIE L* values for animals harvested in the summer season over age. A moderate coefficient of determination (R^2 = 0.4460) with a negative slope ($y= 36{,}344e^{-0{,}01x}$) was observed. This indicates CIE L* values, as expected, decreased with age (became darker) and that 44.60 % of the variation observed in the CIE L* values can be explained by the exponential relationship with age. The effect of age has the potential to be established further if a larger samples size with more defined age groups were to be analysed. The CIE L* values were higher ($p \leq 0.05$) in the summer harvest than in winter. This consequently results in similar differences being observed for the hue-angle (Table 4.2) as a strong linear correlation between the CIE L* and hue-angle values can be noted ($r = 0.7998$; $p < 0.001$). Figure 4.1b and 4.1c illustrate the interaction for CIE L* and hue-angle respectively between the two main effects. The LTL and ST muscle in both seasons in this study had the highest CIE L* (lighter) and hue-angle values and the IS and SS muscle in both seasons had the lowest CIE L* value (darker). The IS and SS in both seasons were also associated with low hue-angle values. With regards to the effect of muscle type on the CIE L* values, it is interesting to note that the two forequarter and SM muscles within a season did not differ significantly from one another, however, they differed between seasons (Figure 4.1b). In general, the LTL muscle was also associated with a high CIE L* which is expected as the pH_u (5.54) were observed to be at the protein's isoelectric point. A pH_u close to the isoelectric point increases the free water between muscle fibres which simultaneously increases the light scattering of a muscle. Similar to this study, lower CIE L* values (darker) were found for the LTL muscle in black wildebeest harvested in the winter season when compared to other seasons such as spring and autumn (Hoffman, van Schalkwyk, & Muller, 2009). The CIE L* values obtained in this study were lower than that found for horses slaughtered in Poland at the age of 10 years (Litwińczuk, Florelk, Skałecki, & Litwińczuk, 2008). It was also lower than found for the LTL in Hispano Breton x Galician Mountain foals slaughtered at 15 months age (Lorenzo & Gómez, 2012). This indicates that the muscles from both horse breeds were observed to be brighter than the plains zebra. The CIE L* values obtained are comparable to that found for eland (Needham et al., 2019), blue wildebeest (Van Heerden, 2018), kudu (Hoffman, Mostert, et al., 2009) and gemsbok (Hoffman & Laubscher, 2010).

The forequarter muscles (IS and SS) and the ST muscle had a higher mean CIE a* value with the ST further associated with having the highest CIE b*, chroma and hue-angle values. Both the CIE

a* and CIE b* values are used to calculate the chroma and hue-angle values and as expected, the pooled CIE a* (r = 0.886; p<0.001) and CIE b* (r = 0.828; p < 0.001) values both had a strong correlation with chroma. In general, the CIE a* value is positively associated with the myoglobin concentration within a muscle rather than the myoglobin structure (Vestergaard et al., 2000). The ST muscle of the plains zebra is thus characterised as having a more saturated red colour. Consequently, it can be postulated that the ST muscle in the plains zebra, as well as the forequarter muscles, have a higher myoglobin concentration in relation to the remaining muscles. This is contradictory to the fact that the ST muscle is generally referred to as a white muscle as it is normally high in glycolytic (type IIB) muscle fibres (Vestergaard et al., 2000). Furthermore, the IS and SS which are red stabilising muscles are typically referred to as muscles red in colour as they are characterised with type I and type IIA muscle fibres, rich in myoglobin. However, in this study, it can be noted that the forequarter muscles have a similar redness as the ST muscle. In previous studies on game meat, it was found that the SS muscle had a redder appearance than the other six selected muscles (Cawthorn et al., 2018; Hoffman, Mostert, et al., 2009; Needham et al., 2019; Van Heerden, 2018). It was observed by Onyango et al., (1998) that zebra loin and rump in comparison to that of beef and gemsbok were the dullest due to the low saturation observed (although only two animals from each species were used).

The CIE b* value is primarily associated with myoglobin structure/form and therefore higher CIE b* values are linked to higher amounts of metmyoglobin on the meat surface. As a result, meat with a higher CIE b* and hue-angle value will exhibit a more yellow/brown meat colour. The plains zebra ST muscle will, therefore, have a light-saturated red-brownish meat surface colour when bloomed. Overall, it can be noted that the meat colour for plains zebra is more red-brownish than purple. The colour measurements observed for donkey meat indicates a more red-purplish meat colour as the CIE a* values are higher than found for plains zebra and the CIE b* values are negative (De Palo et al., 2017). However, a significant difference in the visual observation of the brown colour by consumers has been suggested to be at the point when ~60 % of the myoglobin concentration contains the metmyoglobin structure (Lawrie & Ledward, 2006). Even though statistical differences were found in relation to the colour measurements in the plain zebra muscles, it still raises the question of whether the buyer/consumer will be capable of distinguishing between these.

4.5 CONCLUSION

The aim of this study was to establish the physical meat quality of six selected plains zebra muscles harvested in two seasons each at a different location. As the plains zebra is exported to the European Union on a regular basis the goal of this study is to promote the marketing of the plains zebra meat in the local and export market. The physical meat quality of the plains zebra is found to be in line with other game meat species with shear force values being slightly higher than favoured, although it can be expected that the shear force could decrease with ageing, an aspect that warrants further research. Furthermore, the pH_u obtained was in the normal range with no signs of DFD and are similar to that of various horse breeds and compared favourably to the Marina Franca donkeys. The pH_u, drip and cooking loss percentages, WBSF, CIE L*, CIE b* and hue-angle of selected plains zebra muscles were influenced by harvesting season. The most significant of the seasonal differences were WBSF as the

meat from the winter season was more tender than summer. Higher drip and cooking loss were associated with the animals harvested in the winter season. The colour measurements obtained for the plains zebra were comparable to that associated with game meat colour. The forequarter muscles measured as well as the ST muscle in the plains zebra had a redder colour than the remaining muscles. In general, the plains zebra meat colour can be described as a red-brownish meat colour. As the plains zebra can be exported at present, further research on the physical meat quality should be conducted to ensure more reliable results in relation to the effect of season; larger sample size and the inclusion of autumn and spring will be beneficial. The effect of age should also be investigated further by forming more defined age groups and by potentially including older mares that are not used for breeding purposes. Furthermore, the investigation of the muscle fibre type and collagen content in relation to the tenderness and colour observed will be of value. As the chemical composition of only the LTL muscle has been established to date, the effect of season and muscle type on the nutritional profile of the meat from this species will be beneficial for further marketing of the plains zebra as a meat-producing game species.

4.6 REFERENCES

Badiani, A., Nanni, N., Gatta, P. P., Tolomelli, B., & Manfredini, M. (1997). Nutrient Profile of Horsemeat. *Journal of Food Composition and Analysis*, *10*(3), 254–269. https://doi.org/10.1006/jfca.1997.0540

Bailey, A. J. (1972). The basis of meat texture. *Journal of the Science of Food and Agriculture*, *23*(8), 995–1007. https://doi.org/10.1002/jsfa.2740230811

Brewer, M. S., Zhu, L. G., Bidner, B., Meisinger, D. J., & McKeith, F. K. (2001). Measuring pork color: effects of bloom time, muscle, pH and relationship to instrumental parameters. *Meat Science*, *57*(2), 169–176. https://doi.org/10.1016/S0309-1740(00)00089-9

Cassens, R. G., & Cooper, C. C. (1971). Red and white mucles. *Advances in Food Research*, *19*, 1–74.

Cawthorn, D., Fitzhenry, L. B., Muchenje, V., Bureš, D., Kotrba, R., & Hoffman, L. C. (2018). Physical quality attributes of male and female wild fallow deer (*Dama dama*) muscles. *Meat Science*, *137*, 168–175. https://doi.org/10.1016/j.meatsci.2017.11.031

De Palo, P., Tateo, A., Maggiolino, A., Marino, R., Ceci, E., Nisi, A., & Lorenzo, J. M. (2017). Martina Franca donkey meat quality: Influence of slaughter age and suckling technique. *Meat Science*, *134*, 128–134. https://doi.org/10.1016/j.meatsci.2017.07.025

Den Hertog-Meischke, M. J. A., Van Laack, R. J. L. M., & Smulders, F. J. M. (1997). The water-holding capacity of fresh meat. *Veterinary Quarterly*, *19*(4), 175–181. https://doi.org/10.1080/01652176.1997.9694767

Elzerman, J. E., Hoek, A. C., van Boekel, M. A. J. S., & Luning, P. A. (2011). Consumer acceptance and appropriateness of meat substitutes in a meal context. *Food Quality and Preference*, *22*(3), 233–240. https://doi.org/10.1016/j.foodqual.2010.10.006

Franco, D., Crecente, S., Vázquez, J. A., Gómez, M., & Lorenzo, J. M. (2013). Effect of cross breeding and amount of finishing diet on growth parameters, carcass and meat composition of foals slaughtered at 15 months of age. *Meat Science*, *93*(3), 547–556. https://doi.org/10.1016/j.meatsci.2012.11.018

Franco, D., & Lorenzo, J. M. (2014). Effect of muscle and intensity of finishing diet on meat quality of foals slaughtered at 15 months. *Meat Science*, *96*(1), 327–334. https://doi.org/10.1016/j.meatsci.2013.07.018

Gill, C. O. (2005). Safety and storage stability of horse meat for human consumption. *Meat Science*, *71*(3), 506–513. https://doi.org/10.1016/j.meatsci.2005.04.030

Hoffman, L. C. (2001). The effect of different culling methodologies on the physical meat quality attributes of various game species. *Proceedings of the 5th International Wildlife Ranching Symposium Sustainable Utilization, Pretoria, South Africa.*, 212–221.

Hoffman, L. C., & Cawthorn, D. M. (2012). What is the role and contribution of meat from wildlife in providing high quality protein for consumption? *Animal Frontiers*, *2*(4), 40–53. https://doi.org/10.2527/af.2012-0061

Hoffman, L. C., Diana, L., Muller, N. M., Van Rensburgh, B. J., & McMillin, K. W. (2019). Carcass yields and physico-chemical meat quality characteristics of Namibian springbok (*Antidorcas marsupialis*) as influenced by muscle, sex and age. *Meat Muscle Biology*, 1–28. https://doi.org/10.22175/mmb2019.05.0016

Hoffman, L. C., Kritzinger, B., & Ferreira, A. V. (2005). The effects of sex and region on the carcass yield and M. *Longissimus lumborum* proximate composition of impala. *Journal of the Science of Food and Agriculture*, *85*(3), 391–398. https://doi.org/10.1002/jsfa.1936

Hoffman, L. C., & Laubscher, L. L. (2010). A comparison between the effects of day and night cropping on gemsbok (Oryx gazella) meat quality. *Meat Science*, *85*(2), 356–362. https://doi.org/10.1016/j.meatsci.2010.02.003

Hoffman, L. C., Mostert, A. C., Kidd, M., & Laubscher, L. L. (2009). Meat quality of kudu (*Tragelaphus strepsiceros*) and impala (*Aepyceros melampus*): Carcass yield, physical quality and chemical composition of kudu and impala *Longissimus dorsi* muscle as affected by gender and age. *Meat Science*, *83*(4), 788–795. https://doi.org/10.1016/j.meatsci.2009.08.022

Hoffman, L. C., Muller, M., Schutte, D. W., Calitz, F. J., & Crafford, K. (2005). Consumer expectations, perceptions and purchasing of South African game meat. *South African Journal of Wildlife Research*, *35*(1), 33–42.

Hoffman, L. C., Schalkwyk, S. van, & Muller, N. (2009). Effect of season and gender on the physical and chemical composition of Black Wildebeest (*Connochaetus gnou*) meat. *South African Journal of Wildlife Research*, *39*(2), 170–174. https://doi.org/10.3957/056.039.0208

Hoffman, L. C., & Wiklund, E. (2006). Game and venison - meat for the modern consumer. *Meat*

Science, *74*(1), 197–208. https://doi.org/10.1016/j.meatsci.2006.04.005

Honikel, K. O. (1998). Reference methods for the assessment of physical characteristics of meat. *Meat Science*, *49*(4), 447–457. https://doi.org/10.1016/S0309-1740(98)00034-5

Honikel, K. O. (2004). Chemical and physical characteristics of meat - pH Measurement. In *Encyclopedia of Meat Sciences* (pp. 238–242). https://doi.org/10.1017/CCO9780511791581.016

Honikel, K. O. (2014). Conversion of muscle to meat: Glycolysis. *Encyclopedia of Meat Sciences*, *1*, 314–318. https://doi.org/10.1016/B978-0-12-384731-7.00098-2

Huff-Lonergan, E. (2009). Fresh meat water-holding capacity. In *Improving the Sensory and Nutritional Quality of Fresh Meat* (pp. 147–160). https://doi.org/10.1533/9781845695439.1.147

Huff-Lonergan, E., & Lonergan, S. M. (2005). Mechanisms of water-holding capacity of meat: The role of postmortem biochemical and structural changes. *Meat Science*, *71*(1), 194–204. https://doi.org/10.1016/j.meatsci.2005.04.022

Hughes, J. M., Kearney, G., & Warner, R. D. (2014). Improving beef meat colour scores at carcass grading. *Animal Production Science*, *54*(4), 422. https://doi.org/10.1071/AN13454

Hughes, J. M., Oiseth, S. K., Purslow, P. P., & Warner, R. D. (2014). A structural approach to understanding the interactions between colour, water-holding capacity and tenderness. *Meat Science*, *98*(3), 520–532. https://doi.org/10.1016/j.meatsci.2014.05.022

Immonen, K., Ruusunen, M., & Puolanne, E. (2000). Some effects of residual glycogen concentration on the physical and sensory quality of normal pH beef. *Meat Science*, *55*(1), 33–38. https://doi.org/10.1016/S0309-1740(99)00122-9

Kohn, T. A., Kritzinger, B., Hoffman, L. C., & Myburgh, K. H. (2005). Characteristics of impala (*Aepyceros melampus*) skeletal muscles. *Meat Science*, *69*(2), 277–282. https://doi.org/10.1016/j.meatsci.2004.07.007

Kritzinger, B., Hoffman, L. C., & Ferreira, A. V. (2003). A comparison between the effects of two cropping methods on the meat quality of impala (*Aepyceros melampus*). *South African Journal of Animal Science*, *33*(4), 233–241.

Lawrie, R. A., & Ledward, D. A. (2006). *Lawrie's meat science* (7th ed). CRC Press.

Lefaucheur, L. (2010). A second look into fibre typing - Relation to meat quality. *Meat Science*, Vol. 84, pp. 257–270. https://doi.org/10.1016/j.meatsci.2009.05.004

Litwińczuk, A., Florelk, M., Skałecki, P., & Litwińczuk, Z. (2008). Chemical composition and physiochemical properties of horse meat from the *Longissimus lumborum* and *semitendinososus* muscle. *Journal of Muscle Foods*, *19*(3), 223–236. https://doi.org/10.1111/j.1745-4573.2008.00117.x

Lorenzo, J. M., Crecente, S., Franco, D., Sarriés, M. V., & Gómez, M. (2014). The effect of livestock production system and concentrate level on carcass traits and meat quality of foals slaughtered

at 18 months of age. *Animal*, *8*(3), 494–503. https://doi.org/10.1017/S175173111300236X

Lorenzo, J. M., & Gómez, M. (2012). Shelf life of fresh foal meat under MAP, overwrap and vacuum packaging conditions. *Meat Science*, *92*(4), 610–618. https://doi.org/10.1016/j.meatsci.2012.06.008

Lorenzo, J. M., Pateiro, M., & Franco, D. (2013). Influence of muscle type on physicochemical and sensory properties of foal meat. *Meat Science*, *94*(1), 77–83. https://doi.org/10.1016/j.meatsci.2013.01.001

Lorenzo, J. M., Sarriés, M. V., Tateo, A., Polidori, P., Franco, D., & Lanza, M. (2014). Carcass characteristics, meat quality and nutritional value of horsemeat: A review. *Meat Science*, *96*(4), 1478–1488. https://doi.org/10.1016/j.meatsci.2013.12.006

Maltin, C., Balcerzak, D., Tilley, R., & Delday, M. (2003). Determinants of meat quality: tenderness. *Proceedings of the Nutrition Society*, *62*(02), 337–347. https://doi.org/10.1079/PNS2003248

Miller, R. K. (2002). Factors affecting the quality of raw meat. In J. Kerry, J. Kerry, & D. A. Ledward (Eds.), *Meat processing* (pp. 27–63). Woodhead Publishing.

Miller, R. K. (2004). Chemical and physical characteristics of meat/Palatability. In *Reference Module in Food Science. Encyclopedia of Meat Sciences* (2nd ed., pp. 256–261).

Mostert, R., & Hoffman, L. C. (2007). Effect of gender on the meat quality characteristics and chemical composition of kudu (Tragelaphus strepsiceros), an African antelope species. *Food Chemistry*, *104*(2), 565–570. https://doi.org/10.1016/j.foodchem.2006.12.006

Needham, T., Laubser, J. G., Kotrba, R., Bureš, D., & Hoffman, L. C. (2019). Sex influence on muscle yield and physiochemical characteristics of common eland (*Taurotragus oryx*) meat. *Meat Science*, *152*, 41–48. https://doi.org/10.1016/j.meatsci.2019.02.008

Neethling, J., Hoffman, L. C., & Britz, T. J. (2014). Impact of season on the chemical composition of male and female blesbok (*Damaliscus pygargus phillipsi*) muscles. *Journal of the Science of Food and Agriculture*, *94*(3), 424–431. https://doi.org/10.1002/jsfa.6281

Neethling, J., Hoffman, L. C., & Muller, M. (2016). Factors influencing the flavour of game meat: A review. *Meat Science*, Vol. 113, pp. 139–153. https://doi.org/10.1016/j.meatsci.2015.11.022

Neethling, N. E., Suman, S. P., Sigge, G. O., Hoffman, L. C., & Hunt, M. C. (2017). Exogenous and endogenous factors influencing color of fresh meat from ungulates. *Meat and Muscle Biology*, *1*(1), 253. https://doi.org/10.22175/mmb2017.06.0032

Onyango, C. A., Izumimoto, M., & Kutima, P. M. (1998). Comparison of some physical and chemical properties of selected game meats. *Meat Science*, *49*(1), 117–125. https://doi.org/10.1016/S0309-1740(97)00116-2

Polidori, P., Vincenzetti, S., Cavallucci, C., & Beghelli, D. (2008). Quality of donkey meat and carcass characteristics. *Meat Science*, *80*(4), 1222–1224. https://doi.org/10.1016/j.meatsci.2008.05.027

Schönfeldt, H. C., & Strydom, P. E. (2011). Effect of age and cut on tenderness of South African beef. *Meat Science*, *87*(3), 206–218. https://doi.org/10.1016/j.meatsci.2010.10.011

Sebsibe, A. (2008). Sheep and Goat Meat Characteristics and Quality. *Meat Production and Quality, Sheep and Goat Production Handbook for Ethiopia*, 323–228.

Segato, S., Cozzi, G., & Andrighetto, I. (1999). Effect of animal morphotype, sex and age on quality of horse meat imported from Poland. *Recent Progress in Animal Production Science: Progress, XII Congress*, 674–676.

Seong, P. N., Park, K. M., Kang, G. H., Cho, S. H., Park, B. Y., Chae, H. S., & Ba, H. Van. (2016). The differences in chemical composition, physical quality traits and nutritional horse meat as affected by various retail cut types. *Asian Australas Journal of Animal Science*, *29*, 89–99.

Shange, N., Gouws, P., & Hoffman, L. C. (2019). Changes in pH, colour and the microbiology of black wildebeest (*Connochaetes gnou*) *Longissimus thoracis et lumborum* (LTL) muscle with normal and high (DFD) muscle pH. *Meat Science*, *147*, 13–19. https://doi.org/10.1016/j.meatsci.2018.08.021

Tateo, A., De Palo, P., Ceci, E., & Centoducati, P. (2008). Physicochemical properties of meat of Italian Heavy Draft horses slaughtered at the age of eleven months. *Journal of Animal Science*, *86*(5), 1205–1214. https://doi.org/10.2527/jas.2007-0629

Taylor, R. G. (2004). Muscle fibre type and meat quality. *Encyclopedia of Meat Sciences*, 876–882.

Van der Merwe, P., & Du Plessis, L. (2014). *Game farm and hunting tourism*. Sun Media Metro.

Van Heerden, A. M. (2018). *Profiling the meat quality of blue wildebeest (Connochaetes taurinus)*. Unpublished Master's thesis in Animal Science, Stellenbosch, South Africa.

Van Schalkwyk, D. L., & Hoffman, L. C. (2016). *Guidelines for the harvesting & processing of wild game in Namibia 2016.* Windhoek, Namibia: Ministry of Environment & Tourism.

Vermeulen, H., Schönfeldt, H. C., & Pretorius, B. (2015). A consumers perspective of the South African red meat classification system. *South African Journal of Animal Science*, *45*(3), 339–353. https://doi.org/http://dx.doi.org/10.4314/sajas.v45i3.11

Vestergaard, M., Oksbjerg, N., & Henckel, P. (2000). Influence of feeding intensity, grazing and finishing feeding on muscle fibre characteristics and meat colour of *semitendinosus*, *Longissimus dorsi* and *supraspinatus* muscles of young bulls. *Meat Science*, *54*(2), 177–185. https://doi.org/10.1016/S0309-1740(99)00097-2

Warriss, P. D. (2000). *Meat science: an introductory text.* Wallingford: CABI Publishing.

CHAPTER 5

INFLUENCE OF MUSCLE TYPE AND SEASON ON THE PROXIMATE COMPOSITION OF PLAINS ZEBRA (*Equus quagga*) MEAT

ABSTRACT

This study reports on the chemical composition of the meat from plains zebra stallions harvested in the Western Cape, South Africa. Proximate analysis were carried out using six muscles, i.e. *Longissimus thoracis et lumborum* (LTL), *semimembranosus* (SM), *biceps femoris* (BF), *semitendinosus* (ST), *infraspinatus* (IS) and the *supraspinatus* (SS), that were collected from animals hunted during winter and summer, respectively. Mineral analysis was carried out on the LTL, SM, BF, and the liver and ribs collected from the winter-harvested plains zebra. The proximate composition of the six muscles ranged between 75.5 – 77.4 g/100g for moisture, 20.4 -20.8 g/100g for protein, 1.5-2.0 g/100g for intramuscular fat, and 1.2-1.3 g/100g for the ash content. There was a significant interaction between season and muscle type for the intramuscular fat and ash content. The six muscles differed in terms of moisture, protein and intramuscular fat, content. The protein content was the only component influenced by season, with higher values reported for winter-harvested animals. The primary macro- and micro-minerals present in the LTL, SM, BF, liver, and rib included potassium, phosphorous, sodium and magnesium together with iron, zinc, copper, selenium (except in the rib), manganese and strontium. Muscle type influenced the sodium, iron, copper, manganese, and strontium levels. Even though differences were noted for the chemical components in plains zebra meat, these differences were small and therefore it is debatable whether it will impact on consumer preference, and thus acceptability of plains zebra meat significantly.

Keywords: Season, Muscle type, Plains zebra, Proximate, Minerals

5.1 INTRODUCTION

There has been a recent trend to move away from meat products and towards plant-based options due to health and sustainability-related concerns. Despite this trend, the adventure of consuming alternative red meat sources compared to conventional meat sources, such as beef, has also been increasing over the last few years (Lorenzo et al., 2014), especially amongst younger more informed consumers (Hoffman & Cawthorn, 2013). Exotic protein sources have become of interest in the modern era as younger consumers that are more adventurous and educated on health-related aspects, are more willing to consume these "new" types of protein sources (Hoffman & Cawthorn, 2013). An example of exotic protein sources local to South Africa include meat derived from various game species such as springbok (*Antidorcas marsupialis*), impala (*Aepyceros melampus*), blesbok (*Damaliscus pygargus phillipsi*), greater kudu (*Tragelaphus strepsiceros*), blue wildebeest (*Connochaetes taurinus*), eland (*Taurotragus oryx*), and plains zebra (*Equus quagga*) (Hoffman & Wiklund, 2006). Meat derived from these species are lean, protein-dense, and sought after by the health-conscious consumer (Hoffman & Wiklund, 2006; Hoffman, Muller, Schutte, & Crafford, 2004; Mostert & Hoffman, 2007). Furthermore, meat derived from these species has unique species-specific organoleptic properties that add to the marketing potential as being a healthy authentic meat product (Neethling, Hoffman, & Muller, 2016). Although game meat in general is a nutritious protein source, very limited information is available on several of these ungulate species, and even less on the various genetic and non-genetic factors that influence the nutritional profile of game meat.

Another alternative meat source that is not very popular amongst consumers is equine meat. Historically, horse and donkey meat were the result from old animals slaughtered at the end of their working lives and was considered as a meat source for low-income households due to its low nutritional and organoleptic quality (Badiani & Manfredini, 1994). Yet, these species and breeds have the potential to produce high-quality meat products when slaughtered as foals (Stanislawczyk & Znamirowska, 2005). A review on the nutritional aspects of horsemeat classified it as being a "dietetic" meat source as it can improve the omega-3 index, docosahexaenoic acid (C22:3n-6) and iron status due to its contribution to a lower intake of total- and LDL-cholesterol (Lorenzo et al., 2014). Horsemeat is also high in essential macro- and micro-minerals that play an important role in human health (Lorenzo et al., 2014). Various studies report on the carcass characteristics, physical and chemical composition of different horse breeds (and sexes) slaughtered at different ages and fed various diets. Even though this meat source has been compromised by the horsemeat adulteration scandal (Stanciu, 2015), the limited available scientific information has contributed to improve consumer perception and nutritional quality of equine meat (Lorenzo et al., 2014).

The *Equus* genus; inclusive of the plains zebra; has long been overlooked in South Africa as a game meat production species. The plains zebra is a hindgut fermenter, which enables them to digest low quality feed, by consuming higher densities, at a comparatively faster rate than other ruminants such as bovine and ungulates. The plains zebra is adapted not only physiologically but behaviourally and morphologically to occupy large geographical home ranges where course grasslands occur, which enables them to use this type of environments more effectively than other ungulates (Duncan, 1992). The latter makes them an ideal candidate for meat production as they can satisfy their protein and

nutrient requirements by ingesting a large amount of low-quality feed when necessary, which then undergo catalytic digestion in the large intestine (Hume, 2002). Furthermore, the meat from the plains zebra has a high potential for export, as the species is not susceptible to foot-and-mouth disease.

Despite these mentioned beneficial aspects, limited research has been conducted on the meat and nutritional quality of the plains zebra. Two studies that reported on the proximate composition of meat derived from the plains zebra mainly focused on the *Longissimus thoracis et lumborum* (LTL) muscle and the muscles of the entire leg. None of the studies has taken any genetic or non-genetic factors into account as the information was merely presented as baseline data. Onyango et al., (1998) compared the composition of the LTL and the whole leg from only two plains zebras to that of beef (Bos indicus), hartebeest/kongoni (Alcelaphus buselaphus) and East African oryx (Oryx beisa), harvested in Kenya. Hoffman, Geldenhuys, & Cawthorn (2016) determined the proximate and fatty acid composition of the LTL muscle and compared it to previous studies on horse and donkey meat. In both studies the plains zebra had a protein and intramuscular fat content described as a protein-dense foodstuff, low in intramuscular fat.

In South Africa, the harvesting of game animals for commercial use is an all-year round activity, with the meat from the plains zebra being exported to European countries (De Villiers C, pers. comm. 2019). The variation in nutritional plane due to the season of harvest and production region has been acknowledged to influence the chemical and sensorial attributes of different species (Neethling, 2014). However, as the plains zebra has the inherent ability to compensate for nutritional variations in forage it would be of value to determine the proximate composition of the meat with season of harvest as an effect while at the same time quantifying, if any, the differences in the proximate composition between selected muscles in the plains zebra. Information on the mineral levels of plains zebra tissues has not been determined, warranting baseline research.

The goal of this study is therefore to extend the knowledge of the chemical composition of plains zebra meat and simultaneously creating awareness of the nutritional benefits of consuming other equine species as well.

5.2 MATERIALS AND METHODS

5.2.1 Animals and study location

A total of 20 plains zebra stallions were culled in the Fynbos biome situated in the Western Cape Province, South Africa. Eight of the plains zebras were culled in the winter season (June 2017) at Prinskraal farm, near Bredasdorp and the remaining 12 in the summer season (January 2018) at Elandsberg Nature Reserve –Bartholomeus Klip, near Hermon. All the zebras harvested in the winter roamed in one camp of ~800 ha shared with ~400 other game animals. Whereas those harvested in the summer roamed in smaller camps of ~10 ha with four zebras per camp. Detailed information on the animals and study locations are described in Chapter 3.2.

5.2.2 Plains zebra harvesting, dressing, and sampling

Animals harvested at Prinskraal farm and Elandsberg Nature Reserve was culled, exsanguinated, and eviscerated according to Van Schalkwyk & Hoffman (2016). The carcasses were dressed and

eviscerated at a nearby slaughtering facility on both farms (Ethical clearance number: 10NP_HOF02). During evisceration, a liver sample (from the middle of the liver) from each zebra stallion harvested in the winter season was collected for mineral analysis. The liver samples were labelled and placed in a mobile chiller at ±4°C. The carcasses from both seasons were quartered and hung in the mobile chiller (±4°C) and transported to the University of Stellenbosch where deboning and sampling was continued. Upon arrival, the labelled liver samples from the winter season were stored in a freezer (-20°C) until further analysis. During the deboning procedure, the last rib on the right side of the carcasses from the winter season was collected, labelled, and stored (-20°C) for mineral analysis. See Chapter 3.2 for more detailed information on the harvesting, dressing, and sampling procedures for the plains zebras in both seasons.

5.2.3 Chemical Analysis

The proximate analysis includes the determination of the total moisture, total crude protein, total lipid, and ash content of the selected six muscles. Each analysis was carried out in duplicate, and the final value represents the average of the two. A five percent fault percentage were used, and an extra measurement was taken if the difference between the two values exceeded the fault percentage. The homogenised sample allocated for proximate analysis were thawed overnight at ±4°C before the analysis was conducted.

The moisture content (g/100g) was measured according to the Association of Official Analytical Chemists (AOAC) official method 934.01 (AOAC International, 2002c). The moisture content (g/100g) was determined by drying a 2.5 g homogenized sample for all six muscles at 100ºC for 48 hours. The ash percentage was determined by placing the moisture-free samples in a furnace at 500ºC for 6 hours in accordance with the AOAC method 942.05 (AOAC International, 2002a).

The total lipid content (g/100g) was determined by using 5 g homogenized sample from each muscle according to the chloroform-methanol extraction gravimetric method as described by Lee, Trevino, & Chaiyawat, (1996). The samples were expected to have a lipid content of less than five percent and therefore a 1:2 (v/v) chloroform/methanol solution was used for the lipid extraction.

The total crude protein (g/100g) was determined for each muscle by drying the lipid extracted sample at 60ºC. The lipid extracted and dried samples were separately finely ground and 0.1 g sub-sample analysed in a Leco Nitrogen/Protein Analyser (Leco Fp-528, Leco Corporation) in accordance to the AOAC 992.15 Dumas combustion method (AOAC International, 2002b). The Leco analyser determines the nitrogen percentage and thus the total crude protein was determined by multiplying the nitrogen percentage with 6.25.

5.2.4 Mineral Analysis

The mineral composition (n=8 animals from the winter cull) was determined by using a 5 g raw/wet homogenate sample of the LTL, SM BF muscle and liver, whereas a 5 g defatted and ashed sample was used to determine the mineral composition of the rib. The wet samples were stored in specimen vials at -20°C until thawed overnight at ±4°C prior to the acid digestion.

Upon analysis, the rib samples were thawed overnight at ± 4°C and cleaned of any tissue and cartilage. Each sample was dried at 100°C for 24 hours and the weight before and after drying were recorded. The dried bone was mixed with 100 mL petroleum ether and placed under a ventilator for a total of 48 hours. The fat layer was removed after 24 hours and another 100 mL petroleum ether was added for fat extraction for the remaining 24 hours. After the 48-hour defatting period, the defatted bone samples were dried for 24 hours at 100°C. The defatted bone weight before (1st drying weight) and after drying (2nd drying weight) was recorded. Following the drying procedure, the samples were ashed for 24 hours at 600°C, cooled and weighed. The cooled ashed samples were separately crushed and a 5 g sample was taken for acid digestion.

A 0.5 g accurately weighed sample from each tissue dissolved in ultra-pure nitric acid (HNO_3) and hydrogen peroxide (H_2O_2), were digested using a Mars microwave digester at elevated pressure and temperature (200°C). Following the digestion, the samples were cooled, and the extracts made up to a volume of 50 mL with deionised water in acid-cleaned Falcon tubes. The latter was then analysed for 27 elements from which five were major elements (calcium [Ca], magnesium [Mg], phosphorus [P], potassium [K] and sodium [Na]) and 22 trace elements (aluminium [Al], antinomy [Sb], arsenic [As], barium [Ba], beryllium [Be], boron [B], cadmium [Cd], chromium [Cr], cobalt [Co], copper [Cu], iron [Fe], lead [Pb], lithium (Li), manganese [Mn], mercury [Hg], molybdenum [Mn], nickel [Ni], selenium [Se], silicon [Si], strontium [Sr], vanadium [V] and zinc [Zn]). The major elements were analysed on a calibrated and validated Thermo ICap 6200 inductively coupled plasma atomic emission spectroscopy (ICP-AES) instrument (Thermo Fisher Scientific, Waltham, Massachusetts, USA). The instrument was calibrated with NIST-traceable standards (supplied by Inorganic Ventures Inc., Christiansburg Virginia, USA) to quantify the selected major elements. The NIST-traceable quality control standard (De Bruyn Spectroscopic Solutions, Bryanston, South Africa) was used to validate the accuracy of the calibration and also to monitor drift throughout the analysis. The trace elements were analysed on a calibrated and validated Agilent 7900 quadrupole inductively coupled plasma mass spectrometry (ICP-MS) instrument (Agilent Technologies, Santa Clara, California, USA) set to optimise sensitivity and minimise oxide formation (< 0.3 %). The instrument was optimised for the analysis in Agilent HMI mode where all the samples and standards were diluted with argon gas to minimise the matrix load in the analyser. The selected trace elements were measured by using helium as a collision cell gas. To monitor the instrumental drift and to correct for matrix differences between both samples and standards, an internal standard (ISTD) solution containing scandium, yttrium, germanium, rhodium, and indium was introduced (online). The same calibration and validation procedures for ICP-AES were used.

The results for the bone/rib tissue were corrected to account for the fat percentage extracted from the sample prior to mineral analysis. The wet tissue was expressed as mg/kg meat and liver on a wet-weight basis whilst the bone is expressed as mg/kg rib on a dry-weight basis.

The minerals of the tissues were analysed on two consecutive days with testing of the LTL on day one and the SM, BF, liver, and rib/bone on day two. Therefore, the limit of detection (LOD) for the selected minerals differs between the two days. The LOD values (ug/kg) for the LTL (day 1) and the remaining tissues (day 2) as well as the number of animals who had values above the LOD are depicted in Table 5.1.

Table 5.1 The limit of detection (LOD) value (µg/kg tissue) for each mineral and the total number of animals with mineral levels above the LOD values.

Mineral	Limit of detection (µg/kg tissue)		Number of animals above the limit of detection				
	LTL	SM/BF/Liver/Rib	LTL	SM	BF	Liver	Rib
Aluminium (Al)[1]	80	123	7	5	4	8	8
Antimony (Sb)[1]	1	1	8	8	5	1	8
Arsenic (As)[1]	3	4	8	3	0	3	4
Barium (Ba)[2]	2	3	0	0	0	0	8
Beryllium (Be)[1]	0.002	0.003	1	0	0	1	3
Boron (B)[1]	692	1065	0	0	0	1	8
Cadmium (Cd)[2]	1	2	3	0	1	8	8
Calcium (Ca)[3]	23	35	8	8	7	8	8
Chromium (Cr)[2]	39	61	7	2	6	3	8
Cobalt (Co)[4]	4	6	6	3	1	8	8
Copper (Cu)[4]	8	13	8	8	8	8	8
Iron (Fe)[4]	146	225	8	8	8	8	8
Lead (Pb)[1]	3	5	8	4	4	8	8
Lithium (Li)[4]	28	43	0	0	0	1	0
Magnesium (Mg)[3]	7	11	8	8	8	8	8
Manganese (Mn)[4]	1	1	8	8	8	8	8
Mercury (Hg)[1]	41	62	7	0	4	8	2
Molybdenum (Mo)[4]	37	57	4	0	0	8	6
Nickel (Ni)[1]	0.062	0.095	5	2	5	0	8
Phosphorous (P)[3]	26	41	8	8	8	8	8
Potassium (K)[3]	25	39	8	8	8	8	8
Selenium (Se)[4]	1	2	8	8	8	8	2
Silicon (Si)[1]	28	43	8	3	2	5	8
Sodium (Na)[3]	1	1	8	8	8	8	8
Strontium (Sr)[2]	1	2	8	8	8	8	8
Vanadium (V)[1]	1	2	8	2	1	8	8
Zinc (Zn)[4]	44	68	8	8	8	8	8

Abbreviations: LTL = *Longissimus thoracis et lumborum*, SM = *semimembranosus,* BF = *biceps femoris*
[1]Minerals with undefined functions or environmental contaminations, [2]Non-essential micro-minerals, [3]Essential macro-minerals, [4]Essential micro-minerals

5.2.5 Statistical Analysis

Statistical analysis analyses were performed using the VEPAC model of Statistica 64 version 13.4 (2018). The experimental design for the proximate composition was a mixed model repeated measures of analysis of variance (ANOVA) with animal number as a random effect and season and muscle type as fixed effects. Pearson's correlation coefficient was used to quantify correlations between pooled proximate parameters. The experimental design for the mineral composition was also a mixed model repeated measures of analysis variance (ANOVA) for the pre-dominant macro-minerals (potassium,

phosphorus, sodium, magnesium and calcium) and micro-minerals (iron, zinc, copper, selenium, manganese and strontium) with animal as a random effect and muscle type as a fixed effect. For post hoc testing, Fisher LSD was used. A normal probability plot was compiled for each characteristic to determine any deviations from normality and possible outliers. A level of 5 % was used to determine whether the effects were significant. Results are reported as least square means (LSMeans) and standard error (SE) for each characteristic as per season, and per muscle type where applicable. Descriptive statistics (mean, standard error, minimum and maximum) are reported for the muscle mineral content, and for all the minerals measured in the liver and rib. The mean of each mineral was calculated from the total animals with a detection value higher than the LOD values.

5.3 RESULTS

5.3.1 Proximate Analysis

The interaction and the effect of both season and muscle type on the proximate composition of the selected muscles are represented in Table 5.2 and Table 5.3, respectively. An interaction ($p \leq 0.05$) occurred between season and muscle type for the intramuscular fat and ash content (Table 5.2, Figures 5.1a and b). Almost all muscles, except for the ST and IS, were characterised by a higher intramuscular fat content in the winter than in the summer season, even though these differences were not always of significance. It needs to be noted that the significantly higher intramuscular fat of the BF obtained from winter-harvested animals, differed from the intramuscular fat content of all the other muscles obtained from animals harvested during both seasons. Irrespective of the two main effects, the intramuscular fat content for all the meat samples were below 2 g/100g muscle. For the interaction of the ash content (Figure 5.1b), the SS in the winter was significantly higher than that of all the remaining muscles, except for the BF muscle in the winter season. Nonetheless, the ash content for most of the muscles was ~1.2 g/100g muscle.

Season influenced the muscle protein content, with a higher protein content reported for muscles obtained from the winter-harvested animals, when compared to the summer-harvested group (21.8 ± 0.18 g/100g muscle vs. 20.7 ± 0.12 g/100g muscle: $p \leq 0.05$). The moisture and protein content were significantly influenced by muscle type. An inverse relationship was be observed between the moisture and protein content of corresponding muscles. The LTL and SM muscles had the lowest moisture content and the highest protein content. The forequarter muscles (IS and SS) had the highest moisture content and the lowest protein content.

Table 5.2 The level of statistical significance (p-values) for the main effects of season and muscle type of the chemical meat quality characteristics for plains zebra meat.

Characteristic	Season	Muscle	Muscle x Season
Moisture	0.053	**< 0.001**	0.845
Protein	**< 0.001**	**< 0.001**	0.646
IMF	0.111	**< 0.001**	**0.010**
Ash	0.054	0.392	**0.033**

Abbreviations: IMF = intramuscular fat

Table 5.3 LSMeans (± standard error) for the chemical composition (g/100g muscle) of the plains zebra with the season of harvest and muscle type as the two main effects.

Main effect		Moisture	Protein	IMF	Ash
Season	**Winter**	76.1 ± 0.17	21.8^{a} ± 0.18	1.9 ± 0.07	1.3 ± 0.03
	Summer	76.7 ± 0.13	20.7^{b} ± 0.12	1.6 ± 0.04	1.2 ± 0.02
Muscle type	**LTL**	75.8^{cd} ± 0.19	22.0^{a} ± 0.19	1.5^{b} ± 0.08	1.2 ± 0.03
	SM	75.5^{d} ± 0.23	22.0^{a} ± 0.27	1.6^{b} ± 0.07	1.3 ± 0.05
	BF	76.0^{c} ± 0.19	21.2^{b} ± 0.20	2.0^{a} ± 0.10	1.2 ± 0.03
	ST	76.7^{b} ± 0.16	21.2^{b} ± 0.19	1.6^{b} ± 0.06	1.2 ± 0.03
	IS	77.4^{a} ± 0.23	20.4^{c} ± 0.25	1.8^{a} ± 0.08	1.2 ± 0.04
	SS	77.1^{ab} ± 0.23	20.8^{bc} ± 0.31	2.0^{a} ± 0.13	1.3 ± 0.06

$^{a\text{-}f}$LSMeans within a main effect with different superscript differ significantly at $p \leq 0.05$
IMF = intramuscular fat, LTL = *Longissimus thoracis et lumborum*, SM = *semimembranosus*, BF = *biceps femoris*, ST= *semitendinosus,* IS = i*nfraspinatus*, SS = *supraspinatus*.

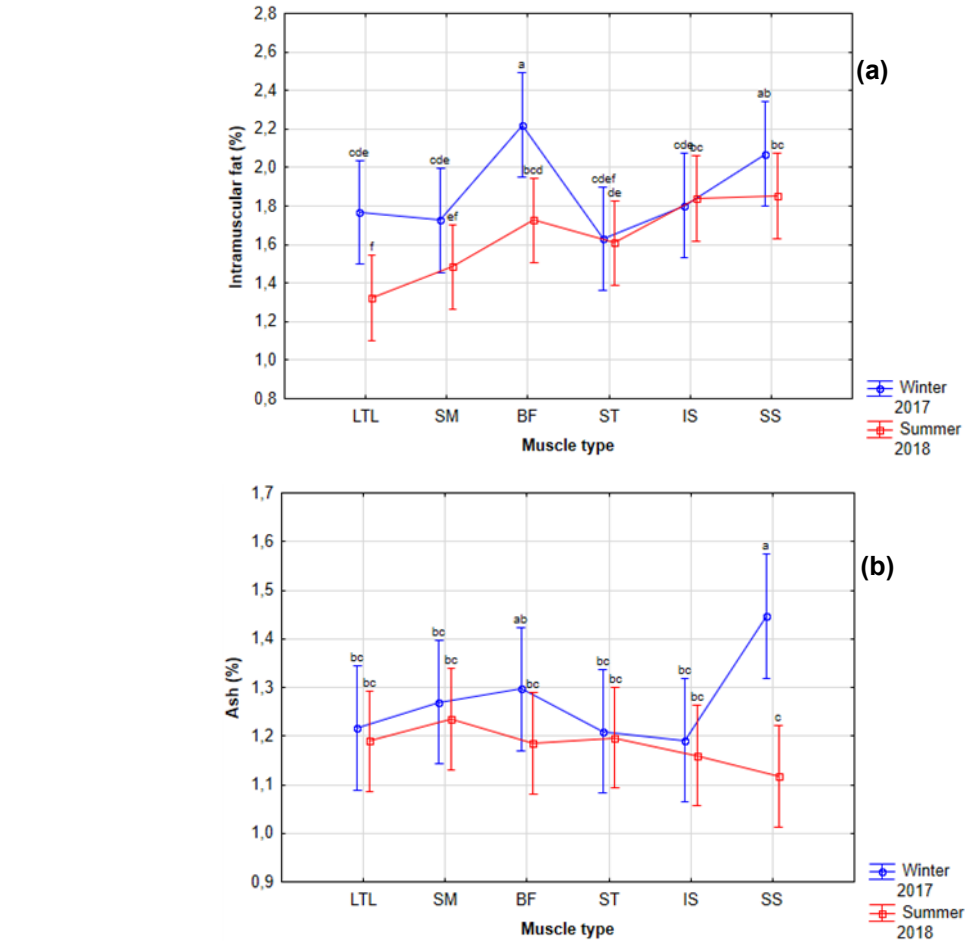

Figure 5.1 Interaction between the season of harvest and muscle type for the intramuscular fat (a) and ash percentage (b).

[a-f]LSMeans of the main effect with different superscript differ significantly at $p \leq 0.05$

LTL = *Longissimus thoracis et lumborum*, SM = *semimembranosus*, BF = *biceps femoris*, ST= *semitendinosus,* IS = i*nfraspinatus*, SS = *supraspinatus*.

5.3.2 Mineral Analysis

The mineral analysis was determined on the muscle, liver and rib tissues obtained from the eight plains zebra stallions harvested in the winter season. Refer to Table 5.1 for the total of zebras which represented an unknown value less than the limit of detection (< LOD). The LOD value for each of the minerals tested in the muscles, liver and rib are also presented in the aforementioned table. Only observations on the mineral content between minerals and tissue types can be made as a result of the descriptive statistical method used to quantify the data. The primary macro- and micro-minerals were selected based on the quantity and level detected in the meat samples obtained from the animals harvested during the winter season. The primary macro-minerals were potassium, phosphorous, magnesium, sodium and calcium and the primary micro-minerals iron, zinc, copper, selenium, manganese, and strontium. The effect of muscle type was only determined on the primary macro- and micro-minerals observed in the three selected muscle tissues (Table 5.4). The remaining minerals measured in the three muscles are presented in Table 5.5. All the minerals quantified in the liver and rib are presented in Table 5.6. The muscle tissue and liver are depicted as mg/kg wet-weight basis and the rib as mg/kg dry-weight basis.

Regarding the effect of muscle type, the BF muscle had a significantly higher mineral content for sodium, magnesium, iron, zinc, and copper compared to the LTL and SM muscles. The SM muscle had a lower manganese concentration ($p = 0.001$) and higher strontium concentration ($p = 0.002$) than both the LTL and BF muscles (Table 5.4). The muscle tissue was characterised by higher levels of potassium and magnesium, when compared to the levels detected in liver and rib. Phosphorous was observed to be the second-highest mineral in the muscle tissue. The phosphorous concentration in the muscles was lower than reported for the liver and rib samples. As expected, the concentration of calcium and sodium were numerically higher in the rib than in the liver and muscle tissue. Iron was the most abundant micro-mineral in the muscle tissue however it had the lowest value compared to the liver followed by the rib. The zinc and especially the strontium concentrations of the rib was higher than detected in the liver and muscle tissue. However, the maximum zinc concentration of the liver (59.661 mg/kg) was almost similar to the minimum zinc concentration of the rib (58.780 mg/kg. The copper concentration in the liver was higher than levels reported for the muscle tissue and rib. Likewise, the manganese of the liver was higher than that of the rib and muscle tissue.

The remaining minerals in the muscle tissue (Table 5.5), liver and rib (Table 5.6) are 10 minerals with undefined functions (aluminium, antimony, arsenic, beryllium, boron, lead, mercury, nickel, silicon and vanadium) which may be present as environmental contaminants, three non-essential (barium, cadmium and chromium) and three essential micro-minerals (cobalt, lithium and molybdenum). These minerals were, however, not present in any of the muscles analysed (Table 5.1). Boron and lithium were each present in only one liver sample (10.057 and 5.398 mg/kg, respectively) and beryllium in only one LTL (0.002 mg/kg) and liver sample (0.004 mg/kg), whilst not detected in any of the remaining soft tissue (muscle tissue and liver tissue) samples. Antimony was only detected in one liver sample (0.005 mg/kg), however it was detected in most of the muscle tissue.

Table 5.4 Mean (± standard error) of the main macro- and micro-minerals (mg/kg meat) of the plains zebra *Longissimus thoracis et lumborum, semimembranosus* and *biceps femoris* muscles as influenced by muscle type.

Minerals	**LTL**			**SM**			**BF**			**p-value**
(mg/kg meat)	**LSMeans ± SE**	**Min**	**Max**	**LSMeans ± SE**	**Min**	**Max**	**LSMeans ± SE**	**Min**	**Max**	
Macro-Minerals										
Potassium (K)	4527.303 ± 144.654	3654.037	4892.289	4536.914 ± 82.512	4277.298	5022.617	4254.485 ± 102.861	3624.929	4544.903	0.170
Phosphorous (P)	2435.078 ± 64.553	2044.748	2613.369	2544.183 ± 32.007	2441.821	2729.155	2494.037 ± 59.355	2135.155	2715.258	0.384
Sodium (Na)	451.300^{b} ± 20.667	346.819	520.548	539.379^{a} ± 17.974	482.002	602.673	519.425^{a} ± 9.833	455.752	541.257	**0.002**
Magnesium (Mg)	328.118^{b} ± 8.239	275.656	344.909	343.412^{b} ± 5.321	318.859	361.091	365.027^{a} ± 6.525	328.710	390.545	**0.006**
Calcium (Ca)	42.531 ± 2.937	31.739	57.592	47.349 ± 2.657	39.348	61.533	41.723 ± 0.904	38.832	46.033	0.21
Micro-Minerals										
Iron (Fe)	24.086^{b} ± 2.171	16.961	34.759	26.134^{b} ± 1.419	21.112	31.717	29.535^{a} ± 1.632	23.800	35.562	**0.001**
Zinc (Zn)	14.131^{b} ± 0.625	12.059	16.700	14.727^{b} ± 0.816	11.396	17.339	16.512^{a} ± 0.846	13.498	19.689	**0.004**
Copper (Cu)	1.285^{b} ± 0.086	0.905	1.651	1.351^{b} ± 0.057	1.141	1.659	1.644^{a} ± 0.083	1.217	1.886	**< 0.001**
Selenium (Se)	0.168 ± 0.006	0.139	0.198	0.175 ± 0.004	0.150	0.191	0.174 ± 0.005	0.149	0.192	0.527
Manganese (Mn)	0.100^{a} ± 0.014	0.051	0.168	0.065^{b} ± 0.008	0.038	0.097	0.108^{a} ± 0.012	0.062	0.166	**0.001**
Strontium (Sr)	0.054^{b} ± 0.004	0.037	0.070	0.066^{a} ± 0.004	0.055	0.087	0.046^{b} ± 0.0020	0.036	0.057	**0.002**

$^{a\text{-}f}$LSMeans within a row with different superscript differ significantly at $p \leq 0.05$

Abbreviations: LTL = *Longissimus thoracis et lumborum*, SM = *semimembranosus,* BF = *biceps femoris*

Table 5.5 Mean (± standard error) mineral composition (mg/kg meat) represented in plains zebra Longissimus thoracis et lumborum, *semimembranosus* and *biceps femoris* muscles.

Minerals	LTL			SM			BF		
(mg/kg meat)	Mean ± SE	Min	Max	Mean ± SE	Min	Max	Mean ± SE	Min	Max
Aluminium (Al)[1]	0.992 ± 0.354	0.208	3.178	1.250 ± 0.367	0.386	3.004	0.878 ± 0.271	0.148	1.896
Antimony (Sb)[1]	0.048 ± 0.017	0.011	0.157	0.010 ± 0.003	0.002	0.028	0.008 ± 0.002	0.003	0.016
Arsenic (As)[1]	0.009 ± 0.000	0.004	0.022	0.006 ± 0.001	0.004	0.007	< LOD	< LOD	< LOD
Barium (Ba)[2]	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD
Beryllium (Be)[1]	0.002 ± 0.00	0.002	0.002	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD
Boron (B)[1]	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD
Cadmium (Cd)[2]	0.002 ± 0.000	0.002	0.002	< LOD	< LOD	< LOD	0.004 ± 0.000	0.004	0.004
Chromium (Cr)[2]	0.305 ± 0.078	0.042	0.620	0.224 ± 0.020	0.185	0.264	0.367 ± 0.088	0.109	0.718
Cobalt (Co)[4]	0.010 ± 0.001	0.006	0.016	0.032 ± 0.013	0.010	0.075	0.008 ± 0.000	0.008	0.008
Lead (Pb)[1]	0.032 ± 0.013	0.003	0.119	0.014 ± 0.003	0.008	0.026	0.218 ± 0.143	0.013	0.826
Lithium (Li)[4]	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD
Mercury (Hg)[1]	0.001 ± 0.000	0.001	0.002	< LOD	< LOD	< LOD	0.002 ± 0.000	0.001	0.003
Molybdenum (Mo)[4]	0.057 ± 0.006	0.043	0.082	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD
Nickel (Ni)[1]	0.019 ± 0.027	0.112	0.281	0.079 ± 0.007	0.065	0.094	0.170 ± 0.035	0.061	0.308
Silicon (Si)[1]	3.412 ± 0.895	1.427	9.035	2.317 ± 0.039	2.230	2.441	3.088 ± 0.264	2.561	3.615
Vanadium (V)[1]	0.005 ± 0.002	0.002	0.019	0.002 ± 0.000	0.002	0.002	0.005 ± 0.000	0.005	0.005

Abbreviations: LTL = *Longissimus thoracis et lumborum*, SM = *semimembranosus',* BF = *biceps femoris,* < LOD = less than limit of detection

[1]Minerals with undefined functions or environmental contaminations, [2]Non-essential micro-minerals, [3]Essential macro-minerals, [4]Essential micro-minerals

Table 5.6 LSMeans (± standard error) mineral composition (mg/kg meat) represented in plains zebra liver and rib.

Minerals	Liver (mg/kg liver)			Rib (mg/kg dry-weight)		
	Mean ± SE	Min	Max	Mean ± SE	Min	Max
Aluminium (Al)[1]	1.398 ± 0.337	0.249	3.501	2.708 ± 0.410	1.510	5.260
Antimony (Sb)[1]	0.005 ± 0.000	0.005	0.005	0.161 ± 0.027	0.039	0.247
Arsenic (As)[1]	0.006 ± 0.000	0.005	0.007	0.005 ± 0.002	0.003	0.011
Barium (Ba)[2]	< LOD	< LOD	< LOD	12.967 ± 0.887	9.632	16.101
Beryllium (Be)[1]	0.004 ± 0.000	0.004	0.004	0.001 ± 0.000	0.001	0.001
Boron (B)[1]	10.543 ± 0.000	10.543	10.543	1.165 ± 0.212	0.761	2.588
Cadmium (Cd)[2]	0.244 ± 0.0199	0.181	0.354	0.003 ± 0.000	0.002	0.005
Calcium (Ca)[3]	81.843 ± 4.332	67.942	101.541	107720.042 ± 5343.651	89515.346	133031.296
Chromium (Cr)[2]	0.150 ± 0.021	0.094	0.215	0.289 ± 0.039	0.149	0.449
Cobalt (Co)[4]	0.105 ± 0.004	0.092	0.121	0.150 ± 0.011	0.124	0.226
Copper (Cu)[4]	10.057 ± 0.413	8.598	11.647	0.382 ± 0.034	0.198	0.501
Iron (Fe)[4]	109.041 ± 10.595	57.070	148.853	79.060 ± 10.926	37.843	125.245
Lead (Pb)[1]	0.150 ± 0.013	0.094	0.197	0.476 ± 0.022	0.371	0.566
Lithium (Li)[4]	5.398 ± 0.000	5.398	5.398	< LOD	< LOD	< LOD
Magnesium (Mg)[3]	235.603 ± 5.976	211.616	260.869	2589.058 ± 125.313	2193.645	3154.185
Manganese (Mn)[4]	3.180 ± 0.115	2.689	3.729	0.181 ± 0.011	0.134	0.232
Mercury (Hg)[1]	0.020 ± 0.003	0.009	0.032	0.001 ± 0.000	0.001	0.001
Molybdenum (Mo)[4]	1.479 ± 0.116	1.154	2.200	0.044 ± 0.005	0.044	0.067
Nickel (Ni)[1]	< LOD	< LOD	< LOD	0.172 ± 0.038	0.172	0.357
Phosphorous (P)[3]	3610.220 ± 56.743	3338.438	3787.107	84160.262 ± 3249.944	71423.088	95435.686
Potassium (K)[3]	2867.866 ± 139.893	2341.109	3363.237	907.494 ± 41.973	694.435	1049.823
Selenium (Se)[4]	0.630 ± 0.0128	0.583	0.683	0.019 ± 0.001	0.018	0.021
Silicon (Si)[1]	2.777 ± 0.407	1.696	4.326	7.047 ± 1.326	3.877	13.339
Sodium (Na)[3]	1314.866 ± 132.793	880.046	1776.594	4686.797 ± 124.825	4197.382	5136.988
Strontium (Sr)[2]	0.136 ± 0.010	0.107	0.185	365.763 ± 19.275	288.947	432.134
Vanadium (V)[1]	0.016 ± 0.003	0.008	0.032	0.004 ± 0.000	0.003	0.005
Zinc (Zn)[4]	52.713 ± 2.242	42.075	59.661	63.385 ± 1.299	58.780	67.378

Abbreviations: SE = standard error, LOD = Limit of detection; [1]Minerals with undefined functions or environmental contaminations, [2]Non-essential micro-minerals, [3]Essential macro-minerals, [4]Essential micro-minerals

5.4 DISCUSSION

5.4.1 Proximate Analysis

The proximate composition (%; g/100 g wet weight) of lean mammalian muscle consists of approximately 75 % moisture, 19 % protein, 2.5 % intramuscular fat and 3.5 % of various soluble non-protein components (Lawrie & Ledward, 2006). Variations in the proximate composition between different animals are the result of intrinsic (genetic) and extrinsic (environmental) influences (Dobranic, Njari, Miokovic, Cvertila, & Kadivc, 2009; Lorenzo et al., 2014), and therefore no species can be used as being representative of another. However, as game species are normally associated as one group in the minds of numerous consumers, an average range for game meat can be established (similar for domestic livestock), to a certain extent. Intrinsic and extrinsic factors that influence the proximate composition of meat includes amongst others the effect of species, breed/sub-species, age, sex, muscle function and location, level of physical activity, and the plane of nutrition. The latter three being applicable to the chemical variation found in this study as this influences not only the muscle fibre types and the connective tissue between muscles, but also the intramuscular fat which in turn has an inverse relationship with moisture (Pic, 2013; Sebranek, 2004).

The effect of season significantly influenced the muscle protein content, with the latter ascribed to the seasonal variation in the quantity and nutritional value of the forage consumed by the plains zebras culled in this study (Neethling, Hoffman, & Britz, 2014). Even though the two study locations included different veld types, the plains zebras primarily foraged on Bermuda grass (*Cynodon dactylon*) that dominated the veld and camps at Bredasdorp and Elandsberg Nature Reserve. Bermuda grass is a perennial C_4 grass species commonly utilised as forage for livestock species in South Africa. C_4 grasses are generally high in nitrogen which is translocated to storage components beneath the ground level as the plant mature, consequently, reducing the protein content and digestibility of the plant. With the onset of the rainy season, the biomass of Bermuda grass is promoted by new growth and in this case, by the natural foraging behaviour of the plains zebra. The plains zebra has the ability to effectively digest forage that is fibrous and low in protein due to their unique digestive physiology. Inherently, plains zebras shift their dietary patterns between seasons as a strategy to achieve their nutritional requirements for survival. During the dry season, the plains zebra consumes a large quantity of poorer quality (which is more available) forage to minimise their energy output to seek for higher quality grass which is sparsely available. Therefore, as expected the veld in both study locations was lush and green during the rainy season (winter) whilst fibrous with a low digestibility in the dry summer season. As a result, the plains zebras harvested in this study consumed larger quantities of the mature low-quality Bermuda grass in the summer season and nutrient-rich Bermuda grass in the winter season per step taken as the grass was in its growing phase. Even though the meat protein content differed significantly between seasons, the difference was ~1.1 g/100g, which may not be of nutritional significance in terms of human consumption.

Game meat derived from various species is characterised by a proximate composition that include ranges of 70-75 g/100g moisture, 20-24 g/100g protein, 0.2-0.25 g/100g intramuscular fat, and 1.0-2.4 g/100g ash content. As the plains zebra is classified as a wild equine species it is of interest to

compare the proximate composition with both horse and donkey breeds (Table 5.7) as they are frequently consumed in Europe (Gill, 2005) to which plains zebra meat is exported (Hoffman & Wiklund, 2006; Hoffman et al., 2016). A review on horse and donkey breeds noted the effect of age on the chemical meat quality characteristics (Lorenzo et al., 2014).

To investigate the effect of age on the chemical attributes of the plains zebra muscles, an exponential regression was calculated for the known ages of the animals harvested in the summer season. A low coefficient of determination (R^2-value) < 0.12, were observed for moisture, protein, intramuscular fat, and ash. It should be noted that the stallions used in this study were all be classified as mature males, and consequently the growth and related change in body weight would have entered the plateau stage of a typical sigmoidal growth curve. Due to their maturity level, a minor increase in intramuscular fat and decrease in moisture with age is expected. However, the indirect intramuscular fat to moisture relationship is typically observed when excess feed is available which is seldom in wild animals. This intramuscular fat to moisture relationship is thus particularly due to the huge effect of season on the nutrient composition of the feed and as a result the dietary intake of an animal.

The meat obtained from the plains zebra harvested in this study had a moisture content in the range reported for horse and donkey breeds and were similar to what has been found for large-bodied game species such as eland (Needham, Laubser, Kotrba, Bureš, & Hoffman, 2019), and blue (Van Heerden, 2018) and black wildebeest (Ndyoki, 2018). Meat from the latter two species and the plains zebra have a slightly higher moisture contents than other game species. The wide range reported for moisture content in horse meat needs to be noted as it can be attributed to factors such as breed differences, slaughter age and muscle type (Lorenzo, Sarriés, et al., 2014). The moisture content of the plains zebras meat samples was, therefore, more comparable to that of the Galician Mountain horse breed slaughtered at 9, 12 and 15 months of age (Franco et al., 2011; Lorenzo, Sarriés, & Franco, 2013; Lorenzo, Pateiro, & Franco, 2013). Moreover, in this study, significantly higher values were observed in the IS (77.4 ± 0.23 g/100g) and SS (77.1 ± 0.23 g/100g) compared to the remaining four muscles. The LTL (75.8 ± 0.19 g/100g) and SM (75.5 ± 0.23 g/100g) were found to be the lowest. Similar findings were found for fallow deer, blue wildebeest and eland harvested in South Africa where the LTL and the SM had the lowest moisture content with the SS being one of the highest (Fitzhenry, 2016; Needham et al., 2019; Van Heerden, 2018). The moisture content for the LTL is similar to what has previously found for the plains zebra LTL muscle by Onyango et al., (1998) and Hoffman et al., (2016).

The protein, intramuscular fat and ash content of the plains zebra meat was in the range reported for various game species, horse, and donkey breeds (Table 5.7). The South African Food Labelling regulation R146/200 has set the standard for protein and fat content in a final meat product to be > 19 g/100g and < 3 g/100g, respectively to be classified as lean. Meat derived from the plains zebra can, therefore, be regarded and marketed as a protein-dense foodstuff low in fat (Hoffman et al., 2016; Onyango et al., 1998).

The protein content was found to be the highest in the LTL (22.0 ± 0.19 g/100g) and SM (22.0 ± 0.27 g/100g) while the lowest protein value was recorded to be in the two forequarter muscles, the IS (20.4 ± 0.25 g/100g) and the SS (20.8 ± 0.31 g/100g). The latter results were expected as the LTL is

known as a muscle high in protein and the SS muscle lower in protein. A similar trend was found for Galician Mountain horse muscles where the LTL and the SM were characterised by a higher protein content, when compared to the BF and ST (Lorenzo et al., 2013). The protein content reported for plains zebra meat in this study further confirms that the protein content of game meat is higher than for domestic livestock. Therefore, the protein content obtained for the plains zebra is comparable to various horse breeds, eland, blue wildebeest, gemsbok, blesbok, kudu and black wildebeest. The results obtained are also similar to that found for plains zebra meat measured by Onyango et al., (1998) and Hoffman et al., (2016).

As mentioned, an inverse relationship has been established in multiple studies between moisture and intramuscular fat, as well as between moisture and protein content. This trend is well recognised where the increase of the moisture content is typically related to a decrease in the protein and intramuscular fat levels (Browning, Huffman, Egbert, & Jungst, 1990; Hoffman, Mostert, & Laubscher, 2009; Hoffman, Kroucamp, & Manley, 2007; Rowe, Macedo, Visentainer, Souza, & Matsushita, 1999). A negative correlation was calculated between the pooled moisture and protein values for the plains zebra muscles ($r = -0.8084$, $p < 0.001$), supporting the inverse relationship established between the two components in previous studies. In relation to intramuscular fat, an inverse linear correlation to moisture was found to be insignificant ($r = -0.010$, $p = 0.912$); this low correlation can be attributed to both the low lipid values as well as a low variation between the lipid values of the respective muscles obtained from the animals hunted during both seasons.

The intramuscular fat content of horse breeds also has a wide range as it has been reported to be influenced by slaughter age, breed, sex, muscle type and finishing diet (Lorenzo et al., 2014; Table 5.7). The intramuscular fat content reported for horse meat was 14.5 g/100g in cuts from the forequarter (Paleari, Soncini, Beretta, & Rossi, 1992) and 16.3 g/100g in the *Longissimus thoracis* (LT) (Matsuoka, Takahashi, & Yamanaka, 1993). The intramuscular fat content of the plains zebra can also be influenced by the abovementioned factors, and therefore further research is warranted to establish the effect of these factors (Hoffman et al., 2016). Nonetheless, in this study, an interaction between season and muscle type was observed (Table 5.2). Muscle type significantly determined the intramuscular fat content, with an average intramuscular fat content of ~1.8 g/100g when all the muscle types were taken into consideration (Table 5.3). The BF (2.22 g/100g) collected from winter-harvested animals was characterised by a significantly higher intramuscular fat content than the remainder of the muscle types obtained from both the winter- and summer harvested animals. The variation observed between muscle types can potentially be ascribed to location and function of the muscle, which differs between species. The intramuscular fat content of a muscle is influenced by the metabolic nature of the fibres as well as the contractile type (Lefaucheur, 2010; Maltin, Balcerzak, Tilley, & Delday, 2003). Red oxidative fibres (Type I) is characterised to have a higher concentration of lipids (intramuscular fat), and are found in high endurance muscles that support posture and in muscles used for low-intensity isometric training (Gunn, 1978; Kohn, Kritzinger, Hoffman, & Myburgh, 2005; Lefaucheur, 2010; R. G. Taylor, 2004). Therefore, apart from the BF, as expected the IS and SS had a relatively high intramuscular fat content as it is characterised as a stabilising red muscle type high in collagen and relatively lower in protein (Frandson, Wilke, & Fails, 2013). This finding is supported by the meat colour findings in Chapter 5, as

the IS and SS were found to be more red in colour than the LTL, SM and BF. Muscles with a higher ratio of white muscle fibres are expected to have lower intramuscular fat levels, e.g. athletic animals such as horses have more fast-contracting fibres than slow-contracting fibres in their limb muscles (Gunn, 1978) due to continuous activity altering the protein and moisture content accordingly. Inter-species differences can also be found as a result of the level of physical activity altering the muscle fibre type ratio within a muscle itself. Onyango et al., (1998) observed a notable lower intramuscular fat content of 0.3 g/100g in the LTL muscle of plains zebra, which potentially can be attributed to the small sample size (n=2) and to the animals not receiving supplementary feed during the dry season, i.e. in which the animals were culled. This assumption can be supported when considering the inter-carcass variation for the fat in both seasons (0.89-2.34 g/100g; Table 5.3) in this study and in the earlier study by Hoffman et al. (2016) (1.03-3.10 g/100g). Additionally, game meat is considered to have an intramuscular fat content < 3 g/100g which is supported by the results found for the plains zebra (Hoffman & Cawthorn, 2012; Hoffman & Wiklund, 2006).

The ash content of meat is an estimate of the smallest muscle fraction that comprises of the salty and inorganic mineral residues remaining after combustion (Perez & Andujar, 1981). The ash content did not significantly differ between season of harvest and muscle type, however, an interaction between the two main effects was observed. The interaction can be due to the significant seasonal difference observed in the SS muscle (Figure 5.1b). The ash values were approximately 1.2-1.3 g/100g and are comparable to that found for previous studies on the plains zebra (Hoffman et al., 2016; Onyango et al., 1998), and were in the range found for various game species, horse breeds and Martina Franca donkeys (Table 5.7).

Table 5.7 Proximate composition range of game species, horse and donkey breeds and plains zebra.

Species	Muscle	Moisture (g/100g)	Protein (g/100g)	Fat (g/100g)	Ash (g/100g)	References
Game	LTL	70-75	20-24	0.2-0.25	1.0–2.4	#1
Horse	Various	68.34-77.40	19.8-22.31	0.12-6.63	0.98-4.03	#2
Donkey	Various	72.50-77.30	19.80-22.80	1.13-3.71	1.01-1.33	#3
Plains zebra	LTL	74.90-75.50	22.1-23.5	0.29-0.31	1.39-1.61	#4
	Leg	74.60-75.00	23.7-24.7	0.24-0.36	1.08-1.12	
Plains zebra	LTL	74.1-77.90	21.39-23.30	1.03-3.10	1.01-1.26	#5

Abbreviation: LTL = *Longissimus thoracis et lumborum*
[#1]Hoffman & Cawthorn, 2013
[#2]Badiani, Nanni, Gatta, Tolomelli, & Manfredini, 1997; Franco, Crecente, Vázquez, Gómez, & Lorenzo, 2013; Franco et al., 2011; Juárez et al., 2009; Lanza, Landi, Scerra, Galofaro, & Pennisi, 2009; Lorenzo, Pateiro, et al., 2013; Lorenzo, Sarriés, et al., 2013; Sarriés & Beriain, 2005; Tateo, De Palo, Ceci, & Centoducati, 2008
[#3]De Palo et al., 2017; Polidori, Beghelli, Cavallucci, & Vincenzetti, 2011; Polidori, Cavallucci, Beghelli, & Vincenzetti, 2009; Polidori, Pucciarelli, Ariani, Polzonetti, & Vincenzetti, 2015; Polidori, Vincenzetti, Cavallucci, & Beghelli, 2008
[#4]Onyango et al., 1998
[#5]Hoffman, Geldenhuys, & Cawthorn, 2016

5.4.2 Mineral Analysis

Red meat is regarded as a valuable source of bioavailable essential macro- and micro-minerals required for normal growth and good human health (Williamson, Foster, Stanner, & Buttriss, 2005). Regarding essential macro-minerals, meat contains high levels of potassium and phosphorus, moderate levels of sodium and magnesium, and relatively low levels of calcium (Keeton, Ellerbeck, & Núñez de González, 2014). In agreement, the main macro-minerals detected in the three plains zebra muscles were potassium (4254.485 - 4536.914 mg/ kg), phosphorous (2435.078 - 2544.183 mg/ kg), sodium (451.300 - 539.379 mg/ kg), magnesium (328.118 – 365.027 mg/ kg) and calcium (41.723 – 47.349 mg/ kg) (Table 5.4). The large quantities of potassium and phosphorous in meat are needed as it contributes to essential processes in the body and in combination with calcium, it is also involved with the formation of bones and teeth (Karakök, Ozogul, Saler, & Ozogul, 2010). The potassium content in various horse breeds for these muscles varies between 190.77-315.50 mg/100g, which is substantially lower than found for plains zebra meat in the present study. The phosphorus content in various horse breeds for these muscles varies between 187.28-278.1 mg/100g (Badiani et al., 1997; Franco & Lorenzo, 2014; Lee et al., 2007; Lorenzo & Pateiro, 2013; Lorenzo et al., 2014) and were comparable to the plains zebra. When compared to the values of beef, the plains zebra meat was found to be higher in both potassium (3500 mg/kg beef) and phosphorous (1800 mg/kg beef) (Sales, 1995). The calcium content was comparable the LTL, SM and BF muscle of 15-month-old Galician Mountain foals (Lorenzo & Pateiro, 2013), however, it was lower than reported for Jeju foals (Lee et al., 2007) and Martina Franca donkeys (Polidori et al., 2008).

Meat is also an important source of essential micro-minerals such as dietary iron and zinc with useful amounts of copper, cobalt, chromium, and selenium (Williamson et al., 2005). The primary essential micro-minerals determined in the selected plains zebra muscles, included iron (24.086-29.535 mg/kg) and zinc (14.131-16. 512 g/kg), followed by copper (1.285-1.644 mg/kg) and lower levels of selenium (0.168-0.175 mg/kg), manganese (0.065-0.100 mg/kg) and strontium (0.046-0.054 mg/kg) (Table 5.4).

The mineral composition of muscle tissue in meat-producing species is influenced to intrinsic factors such as species, breed, sex, slaughter age and muscle type and extrinsic factors such as production region and mineral concentration of the diet (Doyle, 1980). As pertaining to muscle type, the BF muscle had the highest levels of magnesium, iron, zinc, and copper. Significantly, the LTL muscles were observed to have had the lowest sodium content with the SM muscle being the highest in strontium and the lowest in manganese (Table 5.4). The combination of certain minerals in skeletal muscles varies at different anatomical locations (Zarkadas et al., 1987) due to the differences in physical demands resulting in specific fibre type compositions (Doornenbal & Murray, 1982). Therefore, the variation in the mineral distribution observed can be ascribed to the differences in activity level and in contractile processes, muscle fibre type and the biological function of minerals for each of the three muscles (Doornenbal & Murray, 1982; Zarkadas et al., 1987). The BF and SM muscle in equine species are large proximal pelvic limb muscles with long fascicle lengths and specialise in doing work and achieving high force and power, when compared to other pelvic limb muscles (Payne, Hutchinson, Robilliard, Smith, & Wilson, 2005). Most of the locomotive muscle mass occurs in in the hind limb, therefore, it is

hypothesised that equine musculature is optimised for propulsion in the hind limb and for support in the thoracic limb (Payne, Veenman, & Wilson, 2004). Both magnesium and sodium are directly involved with ante-mortem muscle contraction (Keeton et al., 2014) and as expected found in higher quantities in the two pelvic limb muscles in the plains zebra. The sodium content of the selected muscles is comparable to 15-month-old Galician Mountain foals which range from 52.56 – 59.71 mg/100g (Lorenzo & Pateiro, 2013). In addition, Lorenzo, Sarriés, et al. (2014) observed that horsemeat tends to have advantageously slightly lower sodium levels than beef, pork, poultry, and lamb which is beneficial towards people such as hypertension patients aiming to consume a low sodium diet. The latter is therefore also relevant to plains zebra meat. However, the magnesium content of the plains zebra LTL, SM and BF muscles was found to be lower compared to the respective muscles of Galician Mountain foals which range from 410.6-423.2 mg/kg (41.06-42.32 mg/100g; Lorenzo & Pateiro, 2013). Nonetheless, the magnesium levels in the plains zebra LTL (328.118 ± 8.239 mg/kg) were ± 100 mg/kg higher than found in the LTL muscle of Jeju horses (Lee et al., 2007) and LT muscle of Martina Franca donkeys (Polidori et al., 2008). This highlights the effect of equine species and breed differences on the mineral content of meat; therefore, it is possible that the mineral composition may differ between different zebra sub-species as well. Zinc is thought to be present in higher quantities in muscles associated with higher activity levels during movement and was found to be the highest in the BF, which is reported to have a higher force-generating potential than the SM in horses (Payne et al., 2005). A higher iron level is normally found in muscles consisting of predominantly red muscles fibres and as a result a higher myoglobin content (Lawrie & Ledward, 2006). Similar to the plains zebra, the iron, copper, and manganese content of the BF muscle obtained from Galician Mountain foals were also significantly higher than in the LTL and SM muscle. However, the iron, zinc, copper, and manganese content of the Galician Mountain foal muscles were slightly higher in comparison to the corresponding plains zebra muscles in the present study (Lorenzo & Pateiro, 2013). Nonetheless, the iron content in the plains zebra can still be regarded as being high when compared to chicken breast (4.0 mg/kg), beef sirloin (20.7 mg/kg), lamb loin (22.3 mg/kg), pork loin (3.6 mg/kg) and ostrich fillet (24.3 mg/kg) (Lombardi-Boccia, Martinez-Dominguez, & Aguzi, 2002).

In game animals, research often overlooks the investigation of cheaper cuts including bone meat, liver and kidneys which makes up approximately 70 % of the carcass. These cuts have the potential not only to be sold to the lower end of the market but also to the higher speciality end of the market when obtained under a strict infrastructure. Consequently, value can then be added to these cuts when processed into niche products such as carpaccio, liver pâté, droëwors, etc. (Taylor, Lindsey, & Davies-Mostert, 2016). Edible offal tissue such as the liver form part of the traditional African diet as it is an excellent source of nutrients with generally higher concentrations of iron, zinc, copper and manganese than in the muscle tissue (Cymbaluk & Christensen, 1986; Lawrie & Ledward, 2006). In agreement to this study, plains zebra liver had higher amounts of iron (109.041 ± 10.595 mg/kg), zinc (52.713 ± 2.242 mg/kg), copper (10.057 ± 0.413 mg/kg) and manganese (3.180 mg/kg) than the selected muscle types.

A comparison of the selected minerals in the plains zebra muscles and liver with the recommended dietary allowance (RDA) or adequate intake (AI) for the relevant mineral aimed at human

health are presented in Table 5.8. The largest contribution that a 100 g portion of plains zebra meat and liver possibly can potentially make in reaching the RDA and AI requirements would be through the supply of phosphorus, iron, copper, and zinc, and to lesser extent potassium and magnesium. Furthermore, a 100 g liver portion obtained from the plains zebra has the potential to exceed the RDA and AI requirements for iron and copper.

Another tissue with a high mineral content and that remains left over after meat processing, is the skeletal bones of a carcass. Skeletal bones of game animals are a by-product, with the potential to be used as a plant fertilizer. Skeletal bone is a dynamic mineral storing tissue and serves as a reservoir for minerals such as magnesium, sodium and especially calcium and phosphorous (Frandson et al., 2013; McDonald et al., 2002). The rib from the plains zebra stallions was characterised by a high calcium (107720.042 ± 5343.651 mg/kg), phosphorous (84160.262 ± 3249.944 mg/kg), sodium (4686.797 ± 124.825 mg/kg), magnesium (2589.058 ± 125.313 mg/kg), potassium (907.494 ± 41.973 mg/kg), strontium (365.763 ± 19.275 mg/kg), iron (79.060 ± 10.926 mg/kg) and zinc (63.385 ± 1.299 mg/kg) content.

Muscle tissue, bone and especially offal such as the liver and kidney can bio-accumulate toxic metals such as arsenic, cadmium, mercury and lead which is detrimental to human health (Alonso et al., 2004). These metals are transferred to animals through direct exposure from polluted drinking water or pasture irrigated with polluted water (Sedki, Lekouch, Gamon, & Pineau, 2003). The analysis of these metals in animal consumable products is therefore important and is suggested to be determined routinely in order to monitor the minimum residue limit for the respective heavy metals that is considered safe for human consumption (Ambushe, Hlongwane, McCrindle, & McCrindle, 2012). This is especially important in South Africa as meat, offal (Magwedere et al., 2013) and even bone broth form part of the traditional diet. Concerning metal contaminants found in some of the plains zebra tissues analysed were aluminium, antimony, arsenic, cadmium, lead, and mercury. However, it was observed that the levels for all the contaminants in the plains zebra tissues were well below the maximum levels specified for each of the elements (Table 5.8). The latter findings were expected as plains zebras in South Africa does not roam in polluted areas. Aluminium was predominantly detected in the liver (1.398 mg/kg), rib (2.708 ± 0.410 mg/kg) and LTL muscle (0.992 ± 0.354 mg/kg) of the plains zebra. The European Food Safety Authority (EFSA) has set a tolerable weekly intake (TWI) of aluminium to 1.0 mg/kg body weight/week (EFSA, 2008) which equate to ~65 mg per week for a 65 kg individual. Therefore, the aluminium levels in the plains zebra soft tissue and rib is well below the TWI. The mean aluminium levels in all three tissues were lower than detected in fallow deer meat (3.170 – 5.763 mg/kg) (Fitzhenry, 2016) as well as many processed meat products, seafood, dairy products, some bakery products, cocoa products, some vegetables, tea leaves, herbs and spices which ranges between 5-10 mg/kg (EFSA, 2008). Antimony was present in all the LTL (0.048 ± 0.017 mg/kg), SM (0.010 ± 0.003 mg/kg) and rib samples (0.161 ± 0.027 mg/kg). Based on very limited studies the World Health Organization (WHO) recommended a total daily intake (TDI) of 0.006 mg/kg of body weight (WHO, 2003) equating to ~ 2.73 mg/kg per week for a 65 kg individual. Therefore, the mean antimony level of plains zebra muscle and rib can be considered to be very low, however, it is recommended that more studies need to be conducted in order to ensure a firm conclusion on the TDI for antimony (Belzile, Chen, & Filella, 2011).

The Joint Expert Committee on Food Additives (JECFA) of the Food and Agriculture Organization of the United Nations (FAO) and the WHO have set a Provisional Tolerable Weekly Intake (PTWI) for inorganic arsenic of 0.002 mg/kg body weight/day which will equate to ~0.91 mg per week for a 65 kg individual (FAO/WHO, 2011). As a result, the mean arsenic detected in the LTL muscle (0.009 mg/kg) can also be considered to be very low. Lead and cadmium are both metals that not only accumulate significantly in meat and liver but in the bone as well. This was found in agreement with this study as both elements were found in all three tissue types. Lead was detected in all the liver (0.150 ± mg/kg), rib (0.476 ± 0.022 mg/kg) and LTL muscle samples (0.032 ± 0.013 mg/kg) and in 50 % of both the SM (0.014 ± 0.003 mg/kg) and BF samples (0.218 ± 0.143 mg/kg). According to the regulations set by the Commission of the European Communities (CEC, 2006), the lead levels of the plains zebra fell way below the maximum levels set for bovine meat (0.10 mg/kg wet-weight basis) and offal (0.50 mg/kg wet-weight basis). Cadmium was predominantly detected in the liver (0.244 ± 0.0199 mg/kg) and rib (0.003 ± 0.000 mg/kg) and in one LTL (0.002 mg/kg) and in three BF muscle samples (0.004 mg/kg) of this study. The cadmium levels observed were also far below the maximum level set for horse meat (0.20 mg/kg wet-weight basis), bovine meat (0.050 mg/kg wet-weight basis) and for horse and bovine liver (0.50 mg/kg wet weight) (CEC, 2006). Mercury is a highly toxic metal which is not only naturally released but also released, into the environment through human activities (WHO, 2007). Mercury is commonly found in high levels in many fish species (CEC, 2006). A TDI for mercury was determined to be 0.0002 mg/kg body weight per day which will equate to ~0.091 mg per week for a 65 kg individual (IPCS, 2003). Low levels of mercury was detected in the LTL (0.001 ± 0.000 mg/kg; n=7) and BF (0.002 ± 0.000, n=4), however, higher levels of mercury was detected in the liver (0.020 ± 0.0003 WT; n =8) of the plains zebra. The latter indicates that a 65 kg individual needs to consume ~4.5 kg liver per week to reach the maximum tolerable level. Therefore, the mercury content of the liver can still be considered low.

Table 5.8 LSMeans (mg/kg) mineral content found in plains zebra muscles (*Longissimus thoracis et lumborum, semimembranosus* and *biceps femoris*) and liver compared to recommended dietary allowance or maximum intake levels.

Mineral	LTL	SM	BF	Liver	RDA/AI [(i)]	RDA/AI met by 100g LTL	RDA/AI met by 100g SM	RDA/AI met by 100g BF	RDA/AI met by 100g liver
	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/day)	(%)	(%)	(%)	(%)
Macro									
Potassium (K)	4527.303	4536.914	4254.485	2867.866	4700	9.6	9.7	9.1	6.1
Phosphorous (P)	2435.078	2544.183	2494.037	3610.220	700	34.8	36.4	35.6	51.6
Sodium (Na)	451.300	539.379	519.425	1314.866	1500	3.0	3.6	3.5	8.8
Magnesium (Mg)	328.118	343.412	365.027	235.603	310-400	8.2-10.6	8.6-11.1	9.1-11.8	5.8-7.6
Calcium (Ca)	42.531	47.349	41.723	81.843	1000-1200	0.35-0.43	0.39-0.47	0.35-0.42	0.68-0.81
Micro									
Iron (Fe)	24.086	26.134	29.535	109.041	8-18	13.4-30.1	14.5-32.7	16.4-36.9	60.6-136.3%
Zinc (Zn)	14.131	14.727	16.512	52.713	8-11	12.9-17.7	13.4-18.4	15.0-20.6	47.9-65.9
Copper (Cu)	1.285	1.351	1.644	10.057	0.9	14.3	15.0	18.3	111.7
Manganese (Mn)	0.100	0.065	0.108	3.180	1.8-2.3	0.43-0.56	0.28-0.36	0.47-0.60	13.8-17.7

Concerning metals found in the soft tissue of the plains zebra.

Mineral	LTL		SM		BF		Liver		Maximum level
	(mg/kg)	n	(mg/kg)	n	(mg/kg)	n	(mg/kg)	n	
Aluminium (Al)	0.992	7	1.250	5	0.878	4	1.398	8	1 mg/kg bw/week (TWI) [(i)]
Antimony (Sb)	0.048	8	0.010	8	0.008	5	0.005	1	0.006 mg/kg bw/day (TDI) [(ii)]
Arsenic (As)	0.009	8	0.006	3	-	0	0.006	3	0.002 mg/kg bw/day (PTWI) [(iii)]
Cadmium (Cd)	0.002	3	-	0	0.367	1	0.244	8	0.20 mg/kg horse meat; 0.50 mg/kg horse liver [(iv)]
Mercury (Hg)	0.001	7	-	0	0.002	4	0.020	8	0.0002 mg/kg bw/day [(v)]
Lead (Pb)	0.032	8	0.014	4	0.218	4	0.150	8	0.1 mg/kg bovine meat [(iv)]

Abbreviations: LTL = *Longissimus thoracis et lumborum*; SM = *semimembranosus*; BF = *biceps femoris*, RDA = Recommended dietary allowance; AI = adequate intake; TWI = Total weekly intake, TDI = Total daily intake, PTWI = Provisional Tolerable Weekly Intake, bw = body weight

(i) EFSA, 2008
(ii) WHO, 2003
(iii) FAO/WHO, 2011
(iv) CEC, 2006
(v) IPCS, 2003

5.5 CONCLUSION

The study presents the first ever baseline information on the chemical composition of plains zebra meat, taking season of harvest into account and as assessed for six specific skeletal muscles. Harvest season significantly influenced muscle protein content, which potentially may be ascribed to the hindgut fermenter feeding strategy of the plains zebra. The muscle type determined the moisture, protein, and intramuscular fat content. The study also reported on a small significant difference in the chemical attributes of muscles. It is recommended that a larger sample size, taking age and sex of the animals, and hunting season into account, and more hunting seasons be used in future studies to generate additional data and investigate the influence of these factors on the chemical composition of meat derived from the plains zebra.

The plains zebra meat obtained in this study can be regarded and marketed as a lean protein-dense foodstuff as it is high in protein and low in fat. Further research is required on the mineral and fatty acid content of plains zebra meat, and the potential influence thereof on the sensory profile of meat derived from the plains zebra.

5.6 REFERENCES

Alonso, M. L., Montaña, F. P., Miranda, M., Castillo, C., Hernández, J., & Benedito, J. L. (2004). Interactions between toxic (As, Cd, Hg and Pb) and nutritional essential (Ca, Co, Cr, Cu, Fe, Mn, Mo, Ni, Se, Zn) elements in the tissues of cattle from NW Spain. *BioMetals*, *17*(4), 389–397. https://doi.org/10.1023/B:BIOM.0000029434.89679.a2

Ambushe, A. A., Hlongwane, M. M., McCrindle, R. I., & McCrindle, C. M. E. (2012). Assessment of levels of V, Cr, Mn, Sr, Cd, Pb and U in bovine meat. *South African Journal of Chemistry*, *65*, 159–164. Retrieved from http://repository.up.ac.za/handle/2263/19556

AOAC International. (2002a). Ash of animal feed. AOAC official Method 942.05. In *Official methods of analysis* (17th ed.). Arlington, Virginia, USA: Association of Official Analytical Chemist Inc.

AOAC International. (2002b). Dumas combustion method. AOAC official method 992.15. In *Official methods of analysis* (17th ed.). Arlington, Virginia, USA: Association of Official Analytical Chemist Inc.

AOAC International. (2002c). Loss on drying (moisture) at 95 -100^{o}C for feeds. AOAC official methods 934.01. In *Official methods of analysis* (17th ed.). Arlington, Virginia, USA: Association of Official Analytical Chemist Inc.

Badiani, A., & Manfredini, M. (1994). The production of horse meat. *Italian Journal of Animal Science*, *20*, 5–43.

Badiani, A., Nanni, N., Gatta, P. P., Tolomelli, B., & Manfredini, M. (1997). Nutrient Profile of Horsemeat. *Journal of Food Composition and Analysis*, *10*(3), 254–269. https://doi.org/10.1006/jfca.1997.0540

Belzile, N., Chen, Y. W., & Filella, M. (2011). Human exposure to antimony: I. sources and intake. In *Critical Reviews in Environmental Science and Technology* (Vol. 41). https://doi.org/10.1080/10643381003608227

Browning, M. A., Huffman, D. L., Egbert, W. R., & Jungst, S. B. (1990). Physical and compositional characteristics of beef carcasses selected for leanness. *Journal of Food Science*, *55*(1), 9–14. https://doi.org/10.1111/j.1365-2621.1990.tb06005.x

CEC. (2006). Regulation 2006/1881 of the Commission of the European Communities of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs. *Official Journal of the European Union*, *2006*(1881), 5–24. Retrieved from http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2006:364:0005:0024:EN:PDF

Cymbaluk, N. F., & Christensen, D. a. (1986). Copper, zinc and manganese concentrations in equine liver, kidney and plasma. *The Canadian Veterinary Journal. La Revue Veterinaire Canadienne*, *27*(5), 206–210.

De Palo, P., Tateo, A., Maggiolino, A., Marino, R., Ceci, E., Nisi, A., & Lorenzo, J. M. (2017). Martina Franca donkey meat quality: Influence of slaughter age and suckling technique. *Meat Science*, *134*, 128–134. https://doi.org/10.1016/j.meatsci.2017.07.025

Dobranic, V., Njari, B., Miokovic, B., Cvertila, Z., & Kadivc, M. (2009). Chemical composition of horse meat. *Scientific and Professional Papers*, *XI*, 62–67.

Doornenbal, H., & Murray, A. C. (1982). Effects of age, breed, sex and muscle on certain mineral concentrations in cattle. *Journal of Food Science*, *47*, 55–58. https://doi.org/10.1111/j.1365-2621.1982.tb11026.x

Doyle, J. J. (1980). Genetic and nongenetic factors affecting the elemental composition of human and other animal tissues- a review. *Journal of Animal Science*, *50*, 1173–1183.

Duncan, P. (1992). *Zebras, Asses, and Horses: An action plan for the conservation of wild equids*.

EFSA (European Food Safety Authority). (2008). Safety of aluminium from dietary intake - Scientific Opinion of the Panel on Food Additives, Flavourings, Processing Aids and Food Contact Materials (AFC). *EFSA Journal*, *6*(7), 1–34. https://doi.org/10.2903/j.efsa.2008.754

FAO/WHO. (2011). Joint FAO/WHO standard programme codex committee on contaminants in foods. Fith Session. *Joint Fao/Who Food Standards Programme Codex Committee on Contaminants in Foods*, 1–89.

Fitzhenry, L. B. (2016). *Yields and meat-quality attributes of wild fallow deer (dama dama) in South Africa*. Unpublished Master's thesis in Animal Science, Stellenbosch, South Africa.

Franco, D., Crecente, S., Vázquez, J. A., Gómez, M., & Lorenzo, J. M. (2013). Effect of cross breeding and amount of finishing diet on growth parameters, carcass and meat composition of foals slaughtered at 15 months of age. *Meat Science*, *93*(3), 547–556. https://doi.org/10.1016/j.meatsci.2012.11.018

Franco, D., & Lorenzo, J. M. (2014). Effect of muscle and intensity of finishing diet on meat quality of foals slaughtered at 15 months. *Meat Science*, *96*(1), 327–334. https://doi.org/10.1016/j.meatsci.2013.07.018

Franco, D., Rodríguez, E., Purriños, L., Crecente, S., Bermúdez, R., & Lorenzo, J. M. (2011). Meat quality of “Galician Mountain” foals breed. Effect of sex, slaughter age and livestock production system. *Meat Science*, *88*(2), 292–298. https://doi.org/10.1016/J.MEATSCI.2011.01.004

Frandson, R. D., Wilke, W. L., & Fails, A. D. (2013). *Anatomy and Physiology of Farm Animals*.

Gill, C. O. (2005). Safety and storage stability of horse meat for human consumption. *Meat Science*, *71*(3), 506–513. https://doi.org/10.1016/j.meatsci.2005.04.030

Grace, N. D., Pearce, S. G., Firth, E. C., & Fennessy, P. F. (1999). Content and distribution of macro- and micro-elements in the body of pasture-fed young horses. *Australian Veterinary Journal*, *77*(3), 172–176. https://doi.org/10.1111/j.1751-0813.1999.tb11228.x

Gunn, H. M. (1978). Differences in the histochemical properties of skeletal muscles of different breeds of horses and dogs. *Journal of Anatomy*, *127*(3), 615–634.

Hoffman, L. C., & Cawthorn, D. M. (2012). What is the role and contribution of meat from wildlife in providing high quality protein for consumption? *Animal Frontiers*, *2*(4), 40–53. https://doi.org/10.2527/af.2012-0061

Hoffman, L. C., & Cawthorn, D. M. (2013). Exotic protein sources to meet all needs. *Meat Science*, *95*(4), 764–771. https://doi.org/10.1016/j.meatsci.2013.04.027

Hoffman, L. C., Geldenhuys, G., & Cawthorn, D. M. (2016). Proximate and fatty acid composition of zebra (*Equus quagga burchellii*) muscle and subcutaneous fat. *Journal of the Science of Food and Agriculture*, *96*(11), 3922–3927. https://doi.org/10.1002/jsfa.7623

Hoffman, L. C., Kroucamp, M., & Manley, M. (2007). Meat quality characteristics of springbok (*Antidorcas marsupialis*). 2: Chemical composition of springbok meat as influenced by age, gender and production region. *Meat Science*, *76*(4), 762–767. https://doi.org/10.1016/J.MEATSCI.2007.02.018

Hoffman, L. C., Mostert, A. C., & Laubscher, L. L. (2009). Meat quality of kudu (*Tragelaphus strepsiceros*) and impala (*Aepyceros melampus*): The effect of gender and age on the fatty acid profile, cholesterol content and sensory characteristics of kudu and impala meat. *Meat Science*, *83*(4), 737–743. https://doi.org/10.1016/j.meatsci.2009.08.026

Hoffman, L. C., Muller, M., Schutte, D. W., & Crafford, K. (2004). The retail of South African game meat: current trade and marketing trends. *South African Journal of Wildlife Research*, *34*(2), 123–134. https://doi.org/http://hdl.handle.net/10520/EJC117194

Hoffman, L. C., & Wiklund, E. (2006). Game and venison - meat for the modern consumer. *Meat Science*, *74*(1), 197–208. https://doi.org/10.1016/j.meatsci.2006.04.005

Hume, I. D. (2002). Digestive strategies of mammals: Application of chemical reactor theory to

digestion. *Acta Zoologica Sinica*, *48*, 1–19. https://doi.org/10.1002/jmor.20270

IPCS. (2003). *Concise International Chemical Assessment Document 50: Elemental mercury and inorganic mercury compounds: human health aspects. Geneva, World Health Organization, International Programme on Chemical Safety.* Retrieved from http://www.who.int/ipcs/publications/cicad/cicads

Juárez, M., Polvillo, O., Gómez, M. D., Alcalde, M. J., Romero, F., & Valera, M. (2009). Breed effect on carcass and meat quality of foals slaughtered at 24 months of age. *Meat Science*, *83*(2), 224–228. https://doi.org/10.1016/j.meatsci.2009.04.022

Karakök, S. G., Ozogul, Y., Saler, M., & Ozogul, F. (2010). Proximate analysis. Fatty acid profiles and mineral contents of meats: a comparative study. *Journal of Muscle Foods*, *21*(2010), 210–223. https://doi.org/10.1111/j.1745-4573.2009.00177.x

Keeton, J. T., Ellerbeck, S. M., & Núñez de González, M. T. (2014). Chemical Composition. In *Encyclopedia of Meat Sciences* (Vol. 1). https://doi.org/10.1016/B978-0-12-384731-7.00087-8

Kohn, T. A., Kritzinger, B., Hoffman, L. C., & Myburgh, K. H. (2005). Characteristics of impala (*Aepyceros melampus*) skeletal muscles. *Meat Science*, *69*(2), 277–282. https://doi.org/10.1016/j.meatsci.2004.07.007

Lanza, M., Landi, C., Scerra, M., Galofaro, V., & Pennisi, P. (2009). Meat quality and intramuscular fatty acid composition of Sanfratellano and Haflinger foals. *Meat Science*, *81*, 142–147. https://doi.org/10.1016/j.meatsci.2008.07.008

Lawrie, R. A., & Ledward, D. A. (2006). *Lawrie's meat science* (7th ed). CRC Press.

Lee, C. M., Trevino, B., & Chaiyawat, M. (1996). A simple and rapid solvent extraction method for detremining total lipids in fish tissue. *Journal of Association of Analytical Chemist International*, *79*, 487–492.

Lee, C., Seong, P., Oh, W., Ko, M., Kim, K., & Jeong, J. (2007). Nutritional characteristics of horsemeat in comparison with those of beef and pork. *Nutrition Research and Practice*, *1*(1), 70. https://doi.org/10.4162/nrp.2007.1.1.70

Lefaucheur, L. (2010). A second look into fibre typing - Relation to meat quality. *Meat Science*, Vol. 84, pp. 257–270. https://doi.org/10.1016/j.meatsci.2009.05.004

Lombardi-Boccia, G., Martinez-Dominguez, B., & Aguzi, A. (2002). Total Heme and Non-heme Iron. *Food Chemistry and Toxicology*, *67*(5), 1738–1741.

Lorenzo, J. M., & Pateiro, M. (2013). Influence of type of muscles on nutritional value of foal meat. *Meat Science*, *93*(3), 630–638. https://doi.org/10.1016/j.meatsci.2012.11.007

Lorenzo, J. M., Pateiro, M., & Franco, D. (2013). Influence of muscle type on physicochemical and sensory properties of foal meat. *Meat Science*, *94*(1), 77–83. https://doi.org/10.1016/j.meatsci.2013.01.001

Lorenzo, J. M., Sarriés, M. V., & Franco, D. (2013). Sex effect on meat quality and carcass traits of foals slaughtered at 15 months of age. *Animal*, *7*(7), 1199–1207. https://doi.org/10.1017/S1751731113000189

Lorenzo, J. M., Sarriés, M. V., Tateo, A., Polidori, P., Franco, D., & Lanza, M. (2014). Carcass characteristics, meat quality and nutritional value of horsemeat: A review. *Meat Science*, *96*(4), 1478–1488. https://doi.org/10.1016/j.meatsci.2013.12.006

Magwedere, K., Shimwino, J., Hemberger, Y., Hoffman, L. C., Midzi, E. M., & Dziva, F. (2013). Lead and Cadmium Levels in Liver, Kidney and Muscle of Harvested Wild Springbok (*Antidorcus marsupialis*) Under Extensive Management in Southern and Southeastern Namibia. *South African Journal of Wildlife Research*, *43*(1), 52–60. https://doi.org/10.3957/056.043.0110

Maltin, C., Balcerzak, D., Tilley, R., & Delday, M. (2003). Determinants of meat quality: tenderness. *Proceedings of the Nutrition Society*, *62*(02), 337–347. https://doi.org/10.1079/PNS2003248

Matsuoka, A., Takahashi, T., & Yamanaka, Y. (1993). Lipid and fatty acid composition of M. *Longissimus thoracis* of horse. *Japanese Journal of Dairy and Food Science*, *42*(3), 89–96.

McDonald, P., Edwards, R. a, Greenhalgh, J. F. D., Morgan, C. a, Sinclair, L. a, & Wilkinson, R. G. (2002). Animal Nutrition. In *Animal nutrition* (7th ed.). Retrieved from http://www.cabdirect.org/abstracts/19701406676.html

Mostert, R., & Hoffman, L. C. (2007). Effect of gender on the meat quality characteristics and chemical composition of kudu (*Tragelaphus strepsiceros*), an African antelope species. *Food Chemistry*, *104*(2), 565–570. https://doi.org/10.1016/j.foodchem.2006.12.006

Ndyoki, F. P. (2018). *Physicochemical and Microbiological Attributes of Black Wildebeest (Connochaetes gnou) Muscles*. Unpublished Master's thesis in Animal Science, Stellenbosch, South Africa.

Needham, T., Laubser, J. G., Kotrba, R., Bureš, D., & Hoffman, L. C. (2019). Sex influence on muscle yield and physiochemical characteristics of common eland (Taurotragus oryx) meat. *Meat Science*, *152*, 41–48. https://doi.org/10.1016/j.meatsci.2019.02.008

Neethling, J. (2014). *Impact of season on the chemical composition of male and female blesbok (Damaliscus pygargus phillipsi) muscles*. Unpublished Master's thesis in Food Science, Stellenbosch, South Africa.

Neethling, J., Hoffman, L. C., & Britz, T. J. (2014). Impact of season on the chemical composition of male and female blesbok (*Damaliscus pygargus phillipsi*) muscles. *Journal of the Science of Food and Agriculture*, *94*(3), 424–431. https://doi.org/10.1002/jsfa.6281

Neethling, J., Hoffman, L. C., & Muller, M. (2016). Factors influencing the flavour of game meat: A review. *Meat Science*, Vol. 113, pp. 139–153. https://doi.org/10.1016/j.meatsci.2015.11.022

Onyango, C. A., Izumimoto, M., & Kutima, P. M. (1998). Comparison of some physical and chemical properties of selected game meats. *Meat Science*, *49*(1), 117–125. https://doi.org/10.1016/S0309-

1740(97)00116-2

Paleari, M. A., Soncini, G., Beretta, G., & Rossi, M. T. (1992). Microbiological and chemical aspects of corned, cooked and vacuum packed horse- meat. *Italian Journal of Food Science*, *4*, 205–212.

Payne, R. C., Hutchinson, J. R., Robilliard, J. J., Smith, N. C., & Wilson, A. M. (2005). Functional specialisation of pelvic limb anatomy in horses (*Equus caballus*). *Journal of Anatomy*, *206*(6), 557–574. https://doi.org/10.1111/j.1469-7580.2005.00420.x

Payne, R. C., Veenman, P., & Wilson, A. M. (2004). The role of the extrinsic thoracic limb muscles in equine locomotion. *Journal of Anatomy*, *205*(6), 479–490. https://doi.org/10.1111/j.0021-8782.2004.00353.x

Perez, D., & Andujar, G. (1981). Determination of ash content in meat products. *Meat Science*, *5*(3), 165–170. https://doi.org/10.1016/0309-1740(81)90001-2

Pic, Y. (2013). Residue Analysis. *Encyclopedia of Meat Sciences (Second Edition)*, *6*(1995).

Polidori, P., Beghelli, D., Cavallucci, C., & Vincenzetti, S. (2011). Effects of age on chemical composition and tenderness of muscle *Longissimus thoracis* of Martina Franca donkey breed-Donkey meat quality at different age. *Food and Nutrition Sciences*, *02*(03), 225–227. https://doi.org/10.4236/fns.2011.23031

Polidori, P., Cavallucci, C., Beghelli, D., & Vincenzetti, S. (2009). Physical and chemical characteristics of donkey meat from Martina Franca breed. *Meat Science*, *82*(4), 469–471. https://doi.org/10.1016/j.meatsci.2009.03.001

Polidori, P., Pucciarelli, S., Ariani, A., Polzonetti, V., & Vincenzetti, S. (2015). A comparison of the carcass and meat quality of Martina Franca donkey foals aged 8 or 12 months. *Meat Science*, *106*, 6–10. https://doi.org/10.1016/j.meatsci.2015.03.018

Polidori, P., Vincenzetti, S., Cavallucci, C., & Beghelli, D. (2008). Quality of donkey meat and carcass characteristics. *Meat Science*, *80*(4), 1222–1224. https://doi.org/10.1016/j.meatsci.2008.05.027

Rowe, A., Macedo, F. A. F., Visentainer, J. V., Souza, N. E., & Matsushita, M. (1999). Muscle composition and fatty acid profile in lambs fattened in drylot or pasture. *Meat Science*, *51*(4), 283–288. https://doi.org/10.1016/S0309-1740(98)00063-1

Sales, J. (1995). Nutritional quality of meat from some alternative species. *World Review of Animal Production*, *30*, 47–56.

Sarriés, M. V., & Beriain, M. J. (2005). Carcass characteristics and meat quality of male and female foals. *Meat Science*, *70*(1), 141–152. https://doi.org/10.1016/j.meatsci.2004.12.006

Sebranek, J. G. (2004). Raw material composition analysis. *Encyclopedia of Meat Sciences*, (1990).

Sedki, A., Lekouch, N., Gamon, S., & Pineau, A. (2003). Toxic and essential trace metals in muscle, liver and kidney of bovines from a polluted area of Morocco. *Science of the Total Environment*, *317*(1–3), 201–205. https://doi.org/10.1016/S0048-9697(03)00050-0

Stanciu, S. (2015). Horse meat consumption − Between scandal and reality. *Procedia Economics and Finance*, *23*(October 2014), 697–703. https://doi.org/10.1016/S2212-5671(15)00392-5

Stanislawczyk, R., & Znamirowska, A. (2005). Changes in physio-chemical properties of horsemeat during frozen storage. *Acta Scientiarum Polonorum Technologia Alimentaria*, *4*(2), 89–96.

Tateo, A., De Palo, P., Ceci, E., & Centoducati, P. (2008). Physicochemical properties of meat of Italian Heavy Draft horses slaughtered at the age of eleven months. *Journal of Animal Science*, *86*(5), 1205–1214. https://doi.org/10.2527/jas.2007-0629

Taylor, A., Lindsey, P., & Davies-Mostert, H. (2016). An assessment of the economic social and conservation value of the wildlife ranching industry and its potential to support the green economy in South Africa. The Endagered Wildlife Trust. Johannesburg, South Africa. Retrieved from https://www.sagreenfund.org.za/wordpress/wp-content/uploads/2016/04/EWT-RESEARCH-REPORT.pdf

Taylor, R. G. (2004). Muscle fibre type and meat quality. *Encyclopedia of Meat Sciences*, 876–882.

Van Heerden, A. M. (2018). *Profiling the meat quality of blue wildebeest (Connochaetes taurinus)*. Unpublished Master's thesis in Animal Science, Stellenbosch, South Africa.

Van Schalkwyk, D. L., & Hoffman, L. C. (2016). *Guidelines for the harvesting & processing of wild game in Namibia 2016.* Windhoek, Namibia: Ministry of Environment & Tourism.

WHO. (2007). Preventing disease through healthy environments. Exposure to mercury: a major public concern. *World Health Organization*.

Williamson, C. S., Foster, R. K., Stanner, S. A., & Buttriss, J. L. (2005). Red meat in the diet. British Nutrition Foundation. *Nutrition Bulletin*, *30*, 323–335. https://doi.org/10.1111/j.1467-3010.2005.00525.x

World Health Organization. (2003). Antimony in drinking-water background document for development of WHO guidelines for drinking-water quality. *WHO Guidelines*, *2*(3), 1–11. https://doi.org/10.1016/j.kjms.2011.05.002

Zarkadas, C. G., Marshall, W. D., Khalili, A. D., Ngujyen, Q., Zarkadas, G. C., Karatzas, C. N., & Khanizadeh, S. (1987). Mineral composition of selected bovine, porcine and avian muscles, and meat products. *Journal of Food Science*, *52*(3), 520–525.

CHAPTER 6

SENSORY AND FATTY ACID PROFILE OF PLAINS ZEBRA (Equus quagga) MEAT, AS INFLUENCED BY MUSCLE TYPE

ABSTRACT

The aim of this study was to provide information on the descriptive sensory attributes of plains zebra *Longissimus thoracis et lumborum* (LTL)*, semimembranosus* (SM) and *biceps femoris* (BF) muscles. The fatty acid profile of the respective muscles was determined and related to the experience of a trained sensory panel that evaluated the sensory attributes of the meat. The overall aroma intensity had a strong positive correlation with a game-like aroma and flavour, liver-like aroma, metallic aroma and flavour, fatty aroma, and sweet associated taste. The discriminant analysis plot indicated that the plains zebra BF muscle had a sensory profile that differed distinctly from that of the plains zebra LTL and SM muscles, which was clustered together. The BF muscle had the lowest sensory score for beef-like flavour (35.1 ± 0.87) and tenderness (47.1 ± 1.55), and the highest sensory score for residue (18.2 ± 1.33) compared to the LTL and SM muscle. However, the BF muscle had a tendency to be higher in overall intensity aroma (73.3 ± 1.02), game-like aroma (70.7 ± 1.20) and flavour (70.3 ± 0.081), and sweet-associated taste (24.4 ± 0.66), and lower in beef-like aroma (33.2 ± 1.19) and mealiness (5.2 ± 0.93). The BF also had a higher fatty acid content (22.2 ± 1.05 mg/g meat) which can potentially be related to the higher intramuscular fat content of this muscle (2.2 ± 0.10 %) in comparison to its counterparts. Plains zebra meat had a desirable fatty acid profile, as the PUFA:SFA ratios (0.7 ± 0.06; pooled mean) and n6:n3 PUFA ratios (1.5 ± 0.33; pooled mean) were within the recommended guidelines, as well as a low intramuscular fat content (1.7-2.2 %). Findings of this study indicate that extensively produced plains zebra meat can be considered as a healthy red meat alternative.

Keywords: Plains zebra, Descriptive sensory analysis, Fatty Acids

6.1 INTRODUCTION

The introduction of alternative protein sources in the food and beverage industry is an increasing trend globally, as protein is viewed as a primary nutrient, supporting good human health. Protein-centric trends are focussed on plants and more recently on insects, with a movement away from conventional red meat sources (Van Huis, 2016; World Economic Forum, 2019). The catalyst behind reduced-meat diets labelled as vegan (exclusion of all animal products), vegetarian (exclusion of animal flesh and animal slaughter by-products), pescatarian (exclusion of poultry and red meat, includes fish) and flexitarian (mainly vegetarian but includes meat occasionally) can be ascribed to an increased awareness about planetary health (environmental and human health) and animal welfare (Marinova & Bogueva, 2019). Out of 264 individuals surveyed in the United States of America, approximately 80 % of vegans were driven towards veganism due to animal abuse, and only 20 % by its health benefits (Radnitz, Beezhold, & DiMatteo, 2015). It was speculated that ethical vegans transitioned much quicker to veganism as an emotional reaction rather than a health-orientated reaction and that the transition in health vegans was much more delayed and a consequence of research publications and the proliferation of meat substitutes (Radnitz et al., 2015). Therefore, ethical concerns have a higher possibility to result in the total relinquishment of meat while health concerns will rather result in reduced-meat diets or meat alternatives (Von Massow, Weersink, & Gallant, 2019).

Most of the global population is however, still consuming (red) meat from domesticated species as their primary protein source, and this is related to reasons being gustatory, health, traditional, and cultural in nature (Marinova & Bogueva, 2019; World Economic Forum, 2019). With the awareness surrounding animal welfare and planetary health, many omnivores, and flexitarians with a preference for red meat are also leaning towards sustainable red meat products with higher proportions of polyunsaturated fatty acids (PUFA). Red meat is a nutrient-dense commodity which satisfies consumers in terms of its positive sensory attributes (aroma, flavour, and texture) while simultaneously providing a high sense of gut fill, preventing the majority of the population from considering non-meat alternatives (World Economic Forum, 2019). Consequently, the need for alternative animal red meat products which are sustainable and free from medication, antibiotics, growth hormones and disease is progressively becoming more imperative by consumers (Bothma & Du Toit, 2016).

South Africa has the advantage of being able to utilise meat from game animals which are a sustainable, readily available natural resource that is healthy, substance-free, and ethical (Hoffman & Wiklund, 2006). South African red meat consumers consider the health benefits, ethics, availability and most importantly, the sensory profile when purchasing or re-purchasing game meat (Wassenaar, Kempen, & van Eeden, 2019). Game meat can be labelled as “humane” when professional culling teams are used for commercial culling as they are qualified to follow the industry’s harvesting guidelines aimed at animal welfare and the preserving of meat quality (Bothma & Du Toit, 2016; Hoffman & Wiklund, 2006). Game meat produced from an extensive or semi-extensive production system is considered to be one of the purest forms of available red meat rich in protein and low in fat (< 3 %) with an adequate mineral and fatty acid profile (Bothma & Du Toit, 2016).

A primary driver in the red meat industry is to deliver high-quality meat of consistent quality in order to promote repeat purchasing (Marescotti, Caputo, Demartini, & Gaviglio, 2019). A critical

determinant for repeat purchasing is the sensorial quality of a meat product that has the desired visual, aroma, flavour, juiciness, and textural characteristics (Neethling, 2016). However, game meat in South Africa is collectively labelled under the generic term “venison”, thus creating the perception amongst consumers of product irregularity as variations between species exist in terms of its sensory and fatty acid profile (Hoffman, Muller, Schutte, & Crafford, 2004; Neethling, 2016). Therefore to promote various game species as an alternative meat-based product it needs to be available, cost-effective, safe, produced in an humane manner, ethically produced and most importantly, meet the sensory and overall meat quality demands of consumers (Wassenaar et al., 2019). Consequently, research needs to focus on species-specific sensory attributes in game animals in order to advocate for improved marketing and to change the quality perspective among consumers.

The basic sensory profile of certain game species has been established; however, research on the influence of factors such as muscle type, age, breed/subspecies, season, and production system are still scant. The plains zebra (*Equus quagga*) is a species known for yielding a low-fat meat type that has an adequate fatty acid profile as far as health benefits are concerned (Hoffman, Geldenhuys, & Cawthorn, 2016). To the best of our knowledge, no studies have been carried out on the sensory profile of plain zebra meat. Meat derived from the plains zebra is often described as being sweet, similar to horse with a subtle game flavour; however, the flavour has never been established through a trained sensory panel (Powell, 2014). Therefore, this study aims to establish the basic sensory and fatty acid profile between muscle types of the plains zebra and to evaluate how the fatty acid content influences the sensory rating.

6.2 MATERIALS AND METHODS

6.2.1 Animals and study location

Eight mature plain zebra stallions were obtained from Prinskraal farm, near Bredasdorp in June during the winter of 2017. The farm is located in the Western Cape Province South Africa and is part of the Fynbos biome with Central Rûens Shale Renosterveld vegetation unit. The stallions grazed primarily on Bermuda grass (*Cynodon dactylon*) and the natural vegetation, which is a combination of the renosterbos (*Elytropappus rhinocerotis*) and *Aspalathus, Athanasia* and *Rhus* species (Mucina & Rutherford, 2006). For further information regarding the location and vegetation unit refer to the Material and Methods section of Chapter 3.2.

6.2.2 Plains zebra harvesting, dressing, and sampling

The plains zebras were harvested during the day with an appropriate rifle over two consecutive days due to logistical reasons. After shot placement in the head, the stallions were exsanguinated and transported to a nearby slaughtering facility on the farm itself to be weighed, skinned, and eviscerated as described in the Materials and Methods section of Chapter 3.2 (Ethical clearance number: 10NP_HOF02). The quartered carcasses were placed in a suspended manner in a mobile chiller at ± 4°C to undergo *rigor mortis*. The carcasses were then transported to the Department of Animal Sciences at Stellenbosch University for further sampling.

After a refrigeration period of ± 72 hours, the three selected muscles; *Longissimus thoracis et lumborum* (LTL), *semimembranosus* (SM) and the *biceps femoris* (BF) were excised from the right side for sensory analyses. All three muscles were trimmed from the epimysium and subcutaneous fat layer (if any) and subsequently cut into three portions. The first portion was allocated for the chemical/proximate analysis, the second portion for the training phase and the third portion for the testing phase of the descriptive sensory analysis. All three portions were separately vacuum packed, labelled, and frozen at -20°C for two and a half months until analysis.

6.2.3 Descriptive Sensory Analysis

6.2.3.1 Sample preparation

The three plains zebra muscles were thawed at ± 4°C for 36 hours after being stored at -20°C for two and a half months, before the training for the descriptive sensory analysis was performed. On each testing day, the LTL, SM and BF muscles of the allocated replicates were removed from their respective vacuum bags, blotted dry with absorbent paper, and weighed for the calculation of thaw loss. The thaw loss was calculated as a percentage of the muscle weight before freezing (AMSA, 2015). The entire muscle portion was used for cooking, and no salt (NaCl) or seasoning was added to the meat samples throughout the trial. After weighing, each sample was individually positioned onto an aluminium foil-covered stainless-steel grid and inserted into an oven bag (Glad®). In the geometric centre of each muscle a thermocouple probe, attached to a handheld digital temperature monitor (Hanna Instruments, South Africa), was inserted and firmly secured to the cooking bag through a twist tie. The prepared samples were then placed into a preheated industrial oven (163°C; Hobart, Paris, France) until an internal meat temperature of 73°C was recorded on the probe's temperature logger (AMSA, 2015).

Upon reaching the selected internal temperature, the meat samples were removed from the oven and cooking bags and allowed to rest at room temperature for 10 minutes. After the resting period, the cooked samples were blotted dry and weighed to calculate the cooking loss as a percentage of the thawed muscle weight (AMSA, 2015). The cooked samples were then cut from the middle into 1.0 cm^3 cubes and individually enfolded in aluminium foil. The cubes were then placed into three-digit coded glass ramekins (four cubes/ramekin) to ensure a randomised serving sequence and reheated for 10 minutes in a 100°C preheated oven before serving. The samples were evaluated within 10 minutes of serving. At each training/testing station, the ramekins with the meat cubes were placed into a water bath set to 70°C to ensure a constant temperature throughout the entire training or testing session (AMSA, 2015).

After the descriptive sensory analysis was conducted, the remaining portions of the cooked samples were cut with a 1.0 cm double-bladed scalpel parallel to the muscle fibres into six rectangular 1.0 x 1.0 x 2.0 cm cuboids for the measurement of Warner-Bratzler shear force (WBSF). The shear force was measured using the Instron Universal Testing Machine (Instron UTM, Model 2519-107) with a Warner-Bratzler blade fitting at a load cell of 2 kN. For detailed information on the Instron settings, refer to Materials and Methods of Chapter 4.2.

6.2.3.2 Descriptive Sensory Analysis

The descriptive sensory analysis was conducted at the Department of Food Science (University of Stellenbosch) by a panel of 12 judges that were selected based on their previous experience with sensory analysis of various meat types. The sensory analysis was done on three meat treatments namely the LTL (n = 8), SM (n = 7) and BF (n = 8) muscle, where one plains zebra per muscle represented one replication. The selected panellists were trained according to the sensory guideline and the consensus method described by AMSA (2015) and Lawless & Heymann (2010), respectively. The training of the panellist was performed over four consecutive days, consisting of two 50-minute sessions per day. In the first session on day one of training, each panellist received four 1.0 cm^3 meat cubes per reference sample (Table 6.1). The reference samples served as a baseline measurement for the panellist in order to clarify the 22 sensory descriptors depicted in Table 6.2. The sensory descriptors consisted of eight aroma descriptors, eight flavour descriptors and six textural descriptors. During the second training session on day one, the panellist received two replications of each muscle type with four 1.0 cm^3 meat cubes per muscle per replication. One replication of each muscle (four 1cm^3 meat cubes) was further evaluated by the panellist per 50 minutes session for the remaining training sessions. The panellist used an unstructured line scale ranging from zero (indicating “low intensity’) to 100 (indicating “high intensity”) to score the meat samples for each of the 22 sensory descriptors (AMSA, 2015). Testing was allowed after all the panellist were confident in the defining of the selected sensory descriptors.

Testing of the muscles was also conducted over four consecutive days, with two 50-minute sessions per day. One replicate of all three muscles was assigned in a randomised order to one session at a time, and the re-test method was used for the descriptive sensory analysis (AMSA, 2015). Each panellist was assigned to an individual tasting station equipped with a computer to which the Compusense® five software programme (Compusense, Guelph, Canada) was installed. The panellist was given still mineral water, Pink Lady apple slices and biscuits to use as palate cleansers between each sample.

6.2.4 Fatty acid Analysis

The fatty acid profiles for the LTL, SM and BF muscles from each of the eight plains zebra carcasses were determined. From the raw meat samples allocated for chemical analysis a 20 g portion was cut, trimmed from the epimysium and subcutaneous fat, homogenised, separately vacuum packed, and immediately frozen at -80°C until the fatty acid analysis could be performed.

Prior to the fatty acid analysis, the raw homogenised samples were thawed overnight in a refrigerator set at ± 4°C. From each sample, 1.0 g was weighed out for fat extraction using a chloroform: methanol (2:1; v/v) solution with 0.01 % butylated hydroxytoluene (BHT) as an antioxidant. After that, each sample was homogenised in the extraction solvent for 30 seconds with a polytron mixer (WiggenHauser, D-500 Homogenizer). Each muscle sample was quantified for the individual fatty acid by using heptadecanoic acid (C17:0) as the internal standard (catalogue number H3500, Sigma-Aldrich, Gauteng, South Africa). From the extracted fat solution, a 250 µL sub-sample was collected and trans-methylated in a water bath set to 70 °C for two hours, using methanol: sulphuric acid (19:1; v/v) solution

as the trans-methylated agent. The samples were removed after two hours and left to cool down at room temperature. Once the samples were cooled, water and hexane were used to extract the fatty acid methyl esters (FAME), resulting in the separation of the FAME-containing hexane solution and the distilled water. The top hexane phase was then transferred to a spotting tube and dried in a water bath under nitrogen. Fifty µL hexane was added to the dried sample in the spotting tube, which was then centrifuged. One µL of this mixture was injected into the gas chromatograph for the analysis of the FAME.

The FAME's of each plains zebra's LTL, SM and BF muscles were analysed using a Thermo TRACE 1300 series gas-chromatograph (Thermo Electron Corporation, Milan, Italy) fitted with a flame-ionisation detector (GC-FID), using a 30 m ZB-WAX Zebron 7HG-G007-11 capillary column with an internal diameter of 0.25 mm, a 0.25 µm film thickness (Cat. No. HY260M142P, Anatech, Cape Town, South Africa) and ~ 45 minutes runtime. Helium was used as carrier gas (1ml/min flow rate) as 1.0 µL of the sample was injected in a 5:1 split ratio at an injector temperature maintained at 260°C. The initial oven temperature was set to a 100°C and maintained for two minutes which was followed by a temperature increase rate of 10°C per minute for four minutes until an oven temperature of 140°C was reached. Upon reaching 140°C, the oven temperature was immediately increased with a 3°C per minute until 190°C was reached. Lastly, this was followed by a final temperature increase of 30°C per minute until the final temperature of 260°C was reached and maintained for a minimum of five minutes. The FAME of each plains zebra LTL, SM, and BF sample was identified by comparing the retention times with those of a standard FAME mixture (Supelco™37 Component FAME mi, Cat no 47885-U, Supelo, USA). The results were expressed as a percentage of the total FAME content per muscle.

6.2.5 Statistical Analysis

A completely randomised experimental design was used for this trial as eight male plains zebras were culled at random for each muscle type - *Longissimus thoracis et lumborum* (n = 8), *semimembranosus* (n = 7), and *biceps femoris* (n = 8). Muscle type served as the treatment, and the animals were the replicates, with eight replicates for the LTL and BF muscle and seven replicates for the SM muscle in each analysis (physical, sensory, or chemical). For the descriptive sensory analysis, PanelCheck Software (Version 14.0, www.panelcheck.com) was used to monitor the results provided by the panellist. After that, the data obtained for the descriptive sensory analysis including thaw loss percentage, cooking loss percentage, WBSF values and fatty acids of plains zebra meat were analysed with SAS software (Version 9.4; SAS Institute Inc., Cary, USA). The General Linear Models procedure was used to perform a univariate analysis of variance (ANOVA) and a t-test. Results were reported as LSMeans ± standard error. Deviation from normality for the standardised residuals was tested through the Shapiro-Wilk test (Shapiro & Wilk, 1965). No outlier values were removed for this trial. Fisher's least significant differences were calculated at a 5 % significance interval to compare muscle type (Lyman Ott & Longnecker, 2010). The associations between the sensory attributes were illustrated via a Principle Component Analysis plot (PCA) and Discriminant Analysis (DA) plot (XLSTAT, Version 2016, Addinsof, New York USA). Pearson's Correlation coefficient (*r*) was used to quantify correlations

between parameters, a probability level of 5 % ($p \leq 0.05$) and tendencies was considered significant for all tests. These correlations were compiled and presented in Addendum I.

Table 6.1 Reference standards used during the descriptive sensory analysis training phase of plains zebra meat.

Reference sample	**Reference for**	**Final internal temperature**	**Scale**
Aged beef rump (35 days)	Beef-like aroma and flavour, mealiness	73°C	0 = low intensity; 100 = high intensity
Aged beef rump (35 days) browningss	Sweet-associated aroma and taste	73°C	0 = low intensity; 100 = high intensity
Beef fillet	Initial juiciness, sustained juiciness, tenderness, residue	73°C	0 = low intensity; 100 = high intensity
Mutton[a]	Tenderness, residue	73°C	0 = low intensity; 100 = high intensity
Springbok[b]	Game-like aroma and flavour	73°C	0 = low intensity; 100 = high intensity
Ostrich[c]	Metallic aroma and flavour	73°C	0 = low intensity; 100 = high intensity
Liver[d]	Liver-like aroma, flavour, and texture	No probe used; pan fried.	0 = low intensity; 100 = high intensity
Zebra fat[e]	Fatty aroma	No probe used, melted	0 = low intensity; 100 = high intensity

[a]Mutton (*Ovis aries*) chops, [b]Springbok (*Antidorcas marsupialis*) *Longissimus thoracis et lumborum* muscle, [c]Ostrich (*Struthio camelus*) steaks, [d]beef-ox liver, [e]plains zebra (*Equus quagga*) subcutaneous fat.

Table 6.2 Definition and scale of descriptive sensory analysis attributes (aroma, flavour, taste, and texture) evaluated by the trained sensory panel.

Sensory attributes	Description of attributes	Scale
Aroma and Flavour		0 = low intensity; 100 = high intensity
Overall intensity	Intensity of the aroma in the first few sniffs	0 = low intensity; 100 = high intensity
Game-like*	Aroma/flavour associated with meat from wild animal species	0 = low intensity; 100 = high intensity
Beef-like*	Aroma/flavour associated with cooked aged beef	0 = low intensity; 100 = high intensity
Metallic*	Aroma/flavour associated with raw meat/blood like	0 = low intensity; 100 = high intensity
Liver-like*	Aroma/flavour associated with panned fried beef ox liver	0 = low intensity; 100 = high intensity
Herbaceous*	Aroma/flavour associated with vegetation on farms	0 = low intensity; 100 = high intensity
Fatty	Aroma associated with rancid meat	0 = low intensity; 100 = high intensity
Sweet-associated aroma	Aroma associated with the browning of cooked meat surfaces (Maillard reaction)	0 = low intensity; 100 = high intensity
Sweet-associated taste	Taste associated with sucrose solution	0 = low intensity; 100 = high intensity
Salty taste	Taste associated with sodium ions	0 = low intensity; 100 = high intensity
Sour taste	Taste associated with citric acid solution	0 = low intensity; 100 = high intensity
Texture		0 = low intensity; 100 = high intensity
Initial juiciness	Amount of fluid extruded on the surface of meat when pressed between thumb and forefinger (perpendicular to the fibres)	0 = low intensity; 100 = high intensity
Sustained juiciness	Amount of moisture perceived during mastication	0 = low intensity; 100 = high intensity
Tenderness	Impression of tenderness after mastication	0 = low intensity; 100 = high intensity
Residue	Residue tissue remaining after mastication (difficult to chew through)	0 = low intensity; 100 = high intensity
Mealiness	Disintegration of muscle fibres into very small particles (perception within the first few chews)	0 = low intensity; 100 = high intensity
Liver-like texture	Texture similar to that of pan-fried beef ox-liver (spongy/pasty)	0 = low intensity; 100 = high intensity

*Sensory attributes evaluated for aroma and flavour

6.3 RESULTS

6.3.1 Physical measurements

The influence of muscle type on the physical measurements of plains zebra meat is presented in Table 6.3. No significant differences were found between the three muscles for pH_u (5.7 ± 0.20; pooled mean), drip loss percentage (2.09 ± 0.03 %; pooled mean), thaw loss percentage (8.7 ± 0.47 %; pooled mean), cooking loss percentage (30.1 ± 1.76 %; pooled mean) and WBSF values (44.7 ± 1.33 N; pooled mean) at a 5 % level. A tendency was found for the pH_u, of the BF to be higher, when compared to that of the LTL and BF (p = 0.093).

Table 6.3 LSMeans (± standard error) of the physical measurements of mature plains zebra stallions as influenced by muscle type.

Parameter	Muscle type			P-value
	LTL	SM	BF	
pH_u*	5.7^{ab} ± 0.20	5.7^{b} ± 0.20	5.8^{a} ± 0.20	0.093
Thaw loss %	9.2 ± 0.45	8.4 ± 0.50	8.5 ± 0.45	0.587
Cooking loss %	29.1 ± 1.70	31.0 ± 1.87	30.3 ± 1.70	0.732
WBSF (N)	42.6 ± 3.63	43.8 ± 4.00	47.6 ± 3.63	0.567

[a,b]Means with different superscripts in the same row differ from one another (p ≤ 0.05)

*Determined in Chapter 4

6.3.2 Sensory analysis

Muscle type had an influence (p ≤ 0.05) on the beef-like flavour, tenderness, and residue. The BF muscle of the plains zebra had the highest residue (18.2 ± 1.33), the lowest beef-like flavour (35.1 ± 0.87), and tenderness (47.1 ± 1.55). Even though non-significant differences were observed in some attributes the BF muscle had the tendency to be the highest in overall intensity (73.3 ± 1.02), game-like aroma (70.7 ± 1.20), game-like flavour (70.3 ± 1.00), and liver-like flavour (3.9 ± 0.58) and also to be the lowest in beef-like aroma (33.2 ± 1.19) and mealiness (5.2 ± 0.93). Similarly, the LTL muscle tended to be the highest for initial (41.8 ± 1.22) and sustainable juiciness (48.1 ± 1.22). Salt and sour taste was not detected in any of the plains zebra muscles and were thus not included in Table 6.4.

Table 6.4 LSMeans (± standard error) of the sensory scores for plains zebra stallions as influenced by muscle type.

Sensory attributes	Muscle type			p-value
	LTL	SM	BF	
Aroma				
Overall intensity	70.1^{b} ± 1.02	70.5^{ab} ± 1.12	73.3^{a} ± 1.02	0.091
Game-like aroma	67.7^{ab} ± 1.20	66.91^{b} ± 1.32	70.7^{a} ± 1.20	0.085
Beef-like aroma	35.5^{ab} ± 1.19	37.4^{a} ± 1.31	33.2^{b} ± 1.19	0.091
Liver-like aroma	2.8 ± 0.70	2.2 ± 0.77	3.4 ± 0.70	0.515
Metallic-like aroma	15.7 ± 1.21	15.5 ± 1.33	18.1 ± 1.21	0.282
Sweet-associated aroma	22.8 ± 0.67	22.3 ± 0.73	21.7 ± 0.67	0.622
Herbaceous aroma	20.8 ± 0.57	22.2 ± 0.63	21.7 ± 0.57	0.287
Fatty aroma	9.1 ± 0.24	9.2 ± 0.26	9.5 ± 0.24	0.405
Flavour				
Game-like flavour	67.9^{ab} ± 1.00	66.4^{b} ± 1.11	70.3^{a} ± 1.00	0.081
Beef-like flavour	**38.0^{a} ± 0.87**	**38.9^{a} ± 0.98**	**35.1^{b} ± 0.87**	**0.034**
Liver-like flavour	3.1^{ab} ± 0.58	2.1^{b} ± 0.64	3.9^{a} ± 0.58	0.175
Metallic-like flavour	17.5 ± 0.87	17.2 ± 0.96	19.1 ± 0.87	0.328
Sweet-associated taste	22.5^{b} ± 0.66	22.8^{ab} ± 0.73	24.4^{a} ± 0.66	0.134
Herbaceous flavour	20.7 ± 0.54	20.7 ± 0.60	21.2 ± 0.54	0.805
Texture				
Initial juiciness	41.8^{a} ± 1.35	36.4^{b} ± 1.49	38.0^{ab} ± 1.35	0.061
Sustained juiciness	48.1^{a} ± 1.22	44.2^{b} ± 1.24	45.1^{ab} ± 1.22	0.063
Tenderness	**53.7^{a} ± 1.55**	**50.8^{ab} ± 1.71**	**47.1^{b} ± 1.55**	**0.030**
Mealiness	7.5^{ab} ± 0.93	8.3^{a} ± 1.03	5.2^{b} ± 0.93	0.094
Residue	**11.7^{b} ± 1.33**	**13.3^{b} ± 1.46**	**18.2^{a} ± 1.33**	**0.014**

$^{a\ b}$Means with different superscripts in the same row differ from one another ($p \leq 0.05$)

The correlation between the different sensory attributes for plains zebra meat is depicted in a PCA biplot (Figure 6.1). The combination of F1 and F2 explained 53.75 % of the total variance, of which F1 explains 38.86 %, and F2 explains 14.92 %. The discriminant analysis (DA) plot (Figure 6.2) describes 100.0 % of the total variance between treatments (muscle type); with F1 and F2 describing 82.98 % and 17.02 % of the variation between the treatments, respectively. According to Figure 6.2a, the treatments are separated with a slight overlap of the LTL and SM muscle. The BF muscle of the plains zebra is strongly associated with the sensory attributes on the right side of F1, while both LL and SM are strongly associated with the left side of F1.

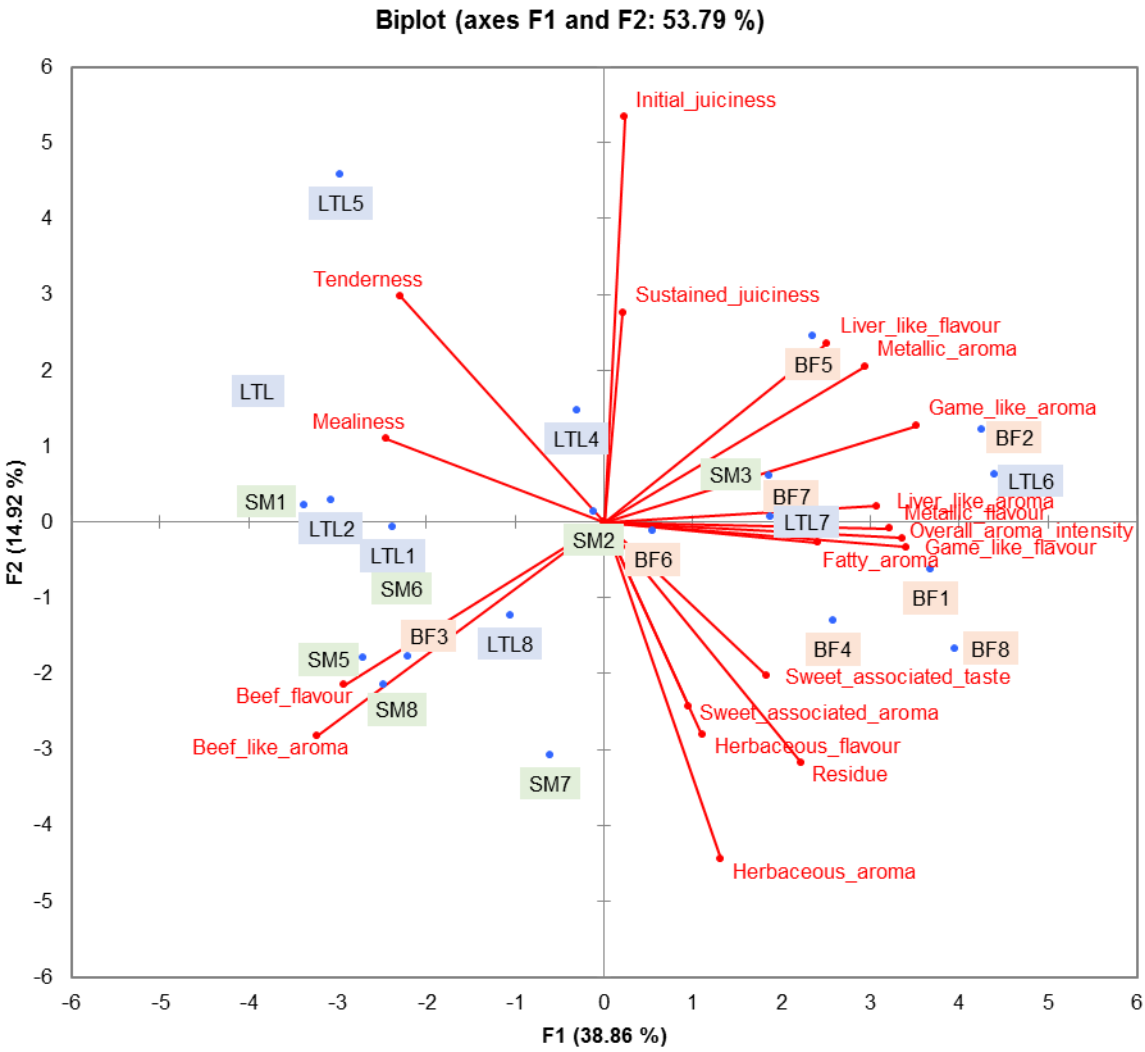

Figure 6.1 Principle component analysis (PCA) biplot depicting the means of the sensory attributes of male plains zebra *Longissimus thoracis et lumborum* (LTL), *semimembranosus* (SM) and *biceps femoris* (BF) muscles.

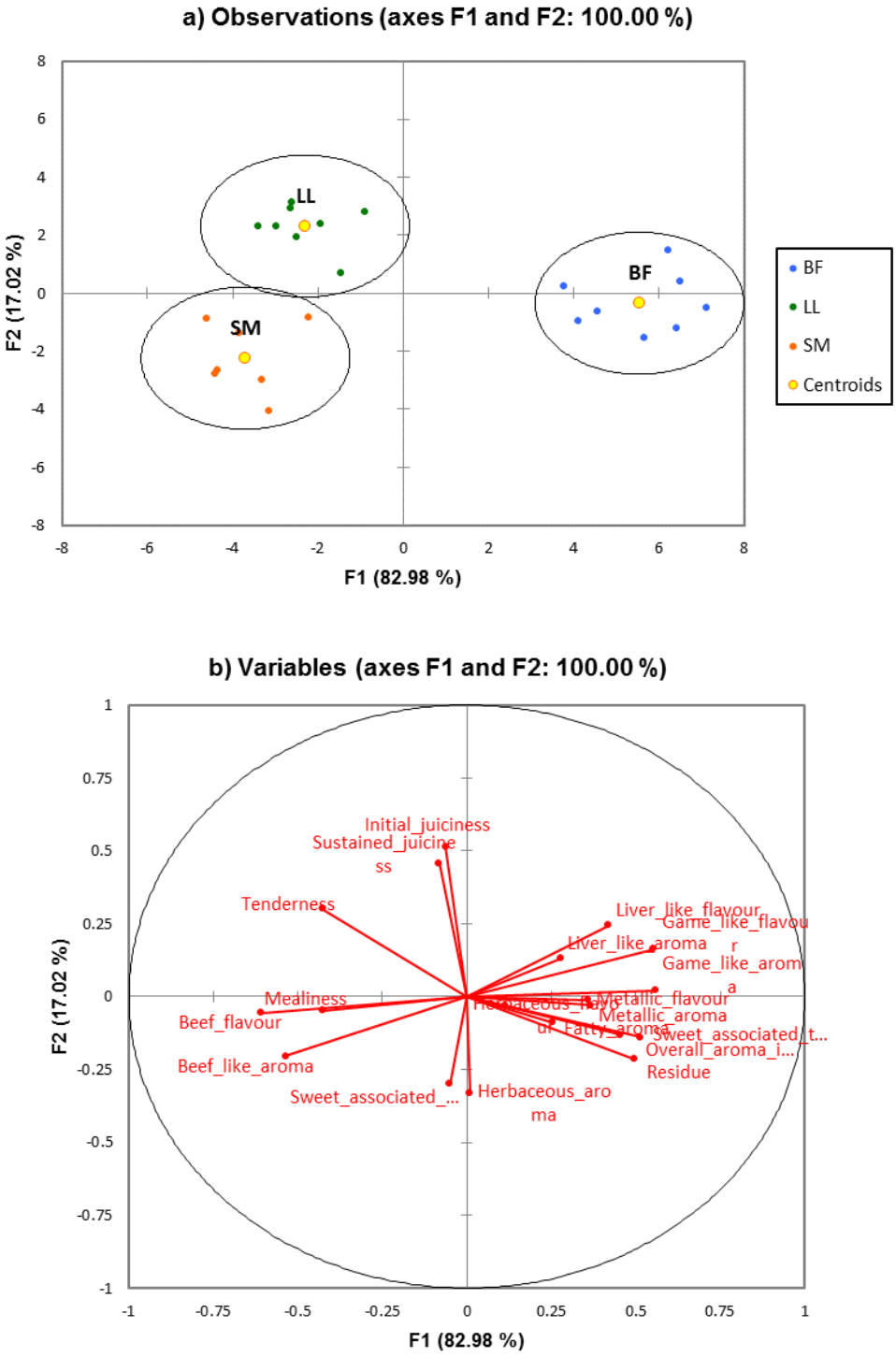

Figure 6.2 Discriminant analysis (DA) plot **(a)** and DA variable loadings plot **(b)** of the sensory characteristics of plains zebra stallions. In the DA plot, the LTL refers to *Longissimus thoracis et lumborum*, the SM to *semimembranosus* and the BF to *biceps femoris* muscles of the plains zebra.

6.3.3 Fatty acid composition

Table 6.5 presents the fatty acid profile (%) and the proximate intramuscular fat (g/100g) content of plains zebra stallions as influenced by muscle type. The highest ($p = 0.009$) intramuscular fat content (2.2 ± 0.10 g/100g) and total fatty acid content (22.2 ± 1.05 mg/100g) were found in the BF muscle, followed by both the LTL and SM muscles.

Regarding the saturated fatty acid content, the SM muscle had the highest ($p \leq 0.05$) C12:0 (Lauric; 3.4 ± 0.17 %), while the LTL and BF did not differ significantly from one another. The C24:0 (Lignoceric) was significantly the lowest in the BF muscle (1.0 ± 0.22 %) but did not differ between the remaining two muscles. Even though no significant differences were observed between muscle types for C6:0 (Hexanoic), C14:0 (Myristic), C15:0 (Pentadecyclic), C18:0 (Stearic), and C20:0 (Arachidic), the SM tended to be the highest in these SFA's. The latter also explains the tendency observed for the SM muscle to have the highest total SFA content (57.3 ± 2.93 %).

The SM muscle was observed to have the lowest ($p \leq 0.05$) C18:1n9c (Oleic; 7.4 ± 1.05 %) and total monounsaturated fatty acid (MUFA; 11.2 ± 1.59 %) content; with no differences between the LTL and BF muscle. The SM muscle also tended to have lower C16:1n7 (Palmitoleic) and C17:1 (Heptadecenoic) MUFA's than both the LTL and BF.

Of the 10 polyunsaturated fatty acids (PUFA) detected in plains zebra meat, only C18:3n3 (alpha-linolenic) was significantly influenced by muscle type, with a higher mean content found in both the LTL (6.6 ± 0.83 %S) and BF (5.9 ± 0.83 %) muscles than in the SM muscle (2.0 ± 0.83 %). However, there was also a tendency observed for the SM muscle to be higher in C18:3n6 (Gamma-linolenic) and C22:2n6 (Docosadienoic).

Muscle type did not have a significant influence on the content of PUFA:SFA ratio, n6-PUFA, n3-PUFA or the n6:n3 PUFA ratio. The pooled means for these calculations were determined as 0.7 ± 0.06 PUFA:SFA ratio, 18.9 ± 0.96 n6-PUFA, 14.0 ± 1.13 n-3 PUFA, and 1.5 ± 0.33 n6:n3 PUFA ratio. However, a high tendency for the SM to be lower in n3-PUFA than both the LTL and BF existed.

Table 6.5 LSMeans (± standard error) of the fatty acid profile (%) of adult plains zebra stallions as influenced by muscle type.

Fatty Acid	Total mean	Muscle type			p-value
		LTL	SM	BF	
Total fatty acids (mg/100g)	19.1 ± 1.05	**17.7^{b} ± 1.05**	**17.3^{b} ± 1.05**	**22.2^{a} ± 1.05**	**0.009**
Total IMF (g/100g)		**1.8^{b} ± 0.10**	**1.7^{b} ± 0.10**	**2.2^{a} ± 0.10**	**0.009**
C6:0 (Hexanoic)	2.0 ± 0.26	1.9^{ab} ± 0.26	2.5^{a} ± 0.26	1.5^{b} ± 0.26	0.059
C8:0 (Caprylic)	nd	nd	nd	nd	-
C10:0 (Capic)	2.2 ± 0.41	2.4 ± 0.41	2.1 ± 0.41	2.1 ± 0.41	0.821
C12:0 (Lauric)	2.9 ± 0.17	**2.7^{b} ± 0.17**	**3.4^{a} ± 0.17**	**2.6^{b} ± 0.17**	**0.008**
C13:0 (Tridecyclic)	0.6 ± 0.24	0.6 ± 0.24	0.6 ± 0.24	0.8 ± 0.24	0.684
C14:0 (Myristic)	2.8 ± 0.24	2.6^{b} ± 0.24	3.3^{a} ± 0.24	2.6^{ab} ± 0.24	0.086
C15:0 (Pentadecyclic)	0.9 ± 0.14	0.7^{b} ± 0.14	1.2^{a} ± 0.14	0.8^{ab} ± 0.14	0.059
C16:0 (Palmitic)	25.6 ± 1.84	22.7 ± 1.84	26.9 ± 1.84	27.1 ± 1.84	0.192
C18:0 (Stearic)	12.7 ± 1.22	11.0^{b} ± 1.22	14.7^{a} ± 1.22	12.3^{ab} ± 1.22	0.123
C20:0 (Arachidic)	1.2 ± 0.12	1.0^{b} ± 0.12	1.4^{a} ± 0.12	1.1^{ab} ± 0.12	0.089
C22:0 (Behenic)	nd	nd	nd	nd	-
C23:0 (Tricosylic)	nd	nd	nd	nd	-
C24:0 (Lignoceric)	1.5 ± 0.22	**1.7^{a} ± 0.22**	**1.9^{a} ± 0.22**	**1.0^{b} ± 0.22**	**0.032**
Total SFA	52.1 ± 2.93	47.1^{b} ± 2.93	57.3^{a} ± 2.93	51.9^{ab} ± 2.93	0.085
C14:1n9c (Myristoleic)	nd	nd	nd	nd	-
C15: 1n9t (Cis-10-pentadecenoic)	nd	nd	nd	nd	-
C16:1n7 (Palmitoleic)	2.3 ± 0.30	2.7^{a} ± 0.30	1.8^{b} ± 0.30	2.4^{ab} ± 0.30	0.122
C17:1 (Heptadecenoic)	1.8 ± 0.26	2.2^{a} ± 0.26	1.3^{b} ± 0.26	2.2^{ab} ± 0.26	0.065
C18:1n9c (Oleic)	10.1 ± 1.05	**11.6^{a} ± 1.05**	**7.4^{b} ± 1.05**	**11.3^{a} ± 1.05**	**0.022**
C20:1n9 (Gondoic)	0.8 ± 0.13	1.0 ± 0.13	0.8 ± 0.13	0.7 ± 0.13	0.260
Total MUFA	15.0 ± 1.59	**17.5^{a} ± 1.59**	**11.2^{b} ± 1.59**	**16.4^{a} ± 1.59**	**0.032**
C18:2n6c (Linoleic)	8.9 ± 1.27	9.9 ± 1.27	7.6 ± 1.27	9.1 ± 1.27	0.436
C18:3n6 (Gamma-linolenic)	3.7 ± 0.36	3.4^{ab} ± 0.36	4.5^{a} ± 0.36	3.3^{b} ± 0.36	0.064
C18:3n3 (Alpha- linolenic)	4.8 ± 0.83	**6.6^{a} ± 0.83**	**2.0^{b} ± 0.83**	**5.9^{a} ± 0.83**	**0.003**
C20: 2n6 (Eicosadienoic)	0.3 ± 0.02	0.3 ± 0.02	0.2 ± 0.02	0.3 ± 0.02	0.402
C20:3n6 (Dihomo-gamma-linolenic)	1.4 ± 0.22	1.6 ± 0.22	1.1 ± 0.22	1.4 ± 0.22	0.223
C20:3n3 (Eicosatrienoic)	2.6 ± 0.39	2.9 ± 0.39	2.4 ± 0.39	2.6 ± 0.39	0.738
C20:4n6 (Arachidonic)	1.1 ± 0.13	1.2 ± 0.13	0.9 ± 0.13	1.2 ± 0.13	0.250
C20:5n3 (Eicosapentaienoic)	0.7 ± 0.11	0.8 ± 0.11	0.7 ± 0.11	0.6 ± 0.11	0.317
C22:2n6 (Docosadienoic)	3.8 ± 0.74	2.8^{b} ± 0.74	5.0^{a} ± 0.74	3.6^{ab} ± 0.74	0.128
C22:6n3 (Docosahexaenoic)	5.4 ± 0.77	5.8 ± 0.77	6.0 ± 0.77	4.3 ± 0.77	0.283
Total PUFA	32.9 ± 1.86	35.4 ± 1.86	31.5 ± 1.86	31.7 ± 1.86	0.294
PUFA:SFA	0.7 ± 0.06	0.8 ± 0.06	0.6 ± 0.06	0.6 ± 0.06	0.144
n6-PUFA	18.9 ± 0.96	19.3 ± 0.96	19.1 ± 0.96	18.3 ± 0.96	0.767
n3-PUFA	14.0 ± 1.13	16.1^{a} ± 1.13	12.5^{b} ± 1.13	13.4^{a} ± 1.13	0.098
n6:n3 PUFA ratio	1.5 ± 0.33	1.2 ± 0.33	1.5 ± 0.33	1.8 ± 0.33	0.430

^{a,b}Means with different superscripts in the same row differ from one another ($p \leq 0.05$)

Abbreviations: SFA = saturated fatty acids (includes C6:0, C8:0, C10:0, C12:0, C13:0, C14:0, C15:0,C16:0, C18:0, C20:0, C22:0, C23:0 and C24:0); MUFA = monosaturated fatty acids (includes C14:1n9c, C15:1n9t, C16:1n7,C17:1, C18:1n9c and C20:1n9), PUFA = polyunsaturated fatty acids (includes C18:2n6c, C18:3n6, C18:3n3, C20:2n6, C20:3n6, C20:3n3, C20:4n6, C20, 5n3, C22:2n6 and C22:6n3); IMF = intramuscular fat; nd = not detected

6.4 DISCUSSION

This study compared the physical measurements, sensory characteristics, and fatty acid profile of three muscles (LTL, SM & BF) obtained from mature plains zebra stallions.

Fluctuations in muscle pH are attributed to the level of pre-slaughter stress and daily activity of the animal. The pH of meat is a critical quality measurement as it affects the water-holding capacity, flavour, and tenderness of meat (Honikel, 2004). The pH taken 72 hours post-mortem prior to freezing for all three muscles (5.7 ± 0.20; pooled mean) fell into the higher end of the biologically normal range of 5.3-5.8, and thus any negative associations regarding the water-holding capacity, flavour and tenderness due to the impact of pH can possibly be ruled out (Honikel, 2004).

Game species culled for meat production are usually deboned, vacuum-packed and sold as frozen products to prolong shelf-life to ensure safe consumption year-round as it is supplied throughout South Africa and internationally. Freezing and thawing of meat influences the water fraction by reducing the moisture content of the meat product in the form of exudate (thaw loss) and cooking loss (Leygonie, Britz, & Hoffman, 2012a). The meat used in this trial was, therefore, vacuum packed and kept frozen at -20°C for two and half months and thawed for 36 hours (due to its large size) in a temperature-controlled refrigerator at ± 4°C prior to sensory analysis. However, no differences or tendencies in the thaw and cooking loss percentages between muscle types were observed and may be the result of standardised freezing, thawing, and cooking processes. The cooking loss for the LTL (29.1 ± 1.70 %), SM (31.0 ± 1.87 %) and BF (30.3 ± 1.70 %) in this study was lower than found for the same muscles measured from fresh meat samples in Chapter 4 (33.2 ± 0.81 %, 38.1 ± 0.81 % and 37.9 ± 0.81 %, respectively). The lower cooking loss percentage is in contrast with previous game meat studies that reported either no differences between fresh and previously frozen game meat samples (Leygonie, Britz, & Hoffman, 2012b; Van Heerden, 2018) or reported higher cooking losses in frozen samples compared to fresh game meat samples (Needham et al., 2019). A strong negative correlation between cooking loss percentage and thaw loss percentage was observed in this study ($r = -0.503$; $p = 0.014$), indicating that a decrease in cooking losses would lead to an increase in thaw losses. A strong negative correlation ($r = -0.526$, $p = 0.010$) was also observed between cooking loss percentage and initial juiciness. No correlations between initial or sustained juiciness and thaw loss and pH_u, or between either juiciness attributes in plains zebra LTL, SM and BF existed. Sustained juiciness is often linked to the intramuscular fat of red meat (Lawrie & Ledward, 2006; Wood, Enser, Fisher, et al., 2008), however, this was not the case for plains zebra meat in the current study ($r = -0.102$; $p = 0.643$), possibly due to the low intramuscular fat content of plains zebra meat.

The fatty acid composition of lipids plays an important role in the development of the flavour of red meat (Wood, Enser, Fisher, et al., 2008; Wood et al., 2003) as various flavour precursors are produced from SFA, n-6 and n-3 PUFA's (Wood & Enser, 1997). High proportions of PUFA in game meat can lead to the development off-flavours as it is more susceptible to lipid oxidation (Wood et al., 1999, 2003). The game-like sensory attribute in meat has been suggested to be linked to high levels of PUFA in the intramuscular fat as influenced by the quantity and quality of the grazing material (Lawrie & Ledward, 2006; Wiklund, Manley, Littlejohn, & Stevenson-Barry, 2003). The diet of the plains zebra consisted of the natural vegetation (Chapter 3.2, Material and Methods) in the area, Bermuda grass

(*Cynodon dactylon*), and oat grass (*Avena sativa*), therefore, it was expected that the highest contributor to the overall aroma for plains zebra meat was game-like aroma (66.9-70.7) which had a large absolute intensity relative to the overall aroma intensity (70.1-73.3). A strong positive correlation was found between overall aroma and game-like aroma (r = 0.855; p =0 0.00) and flavour (r = 0.705; p = 0.000) in plains zebra meat. No correlations existed between α-linolenic acid and characteristics that are often perceived as negative sensory attributes by consumers (game-like, liver-like, and metallic) in plains zebra meat (Addendum I). However, negative correlations were present between the n-3 PUFA content and liver-like aroma (r = -0.497; p = 0.016) and metallic aroma (r = -0.527; p = 0.010).

Beef-like aroma (33.2-37.4) and flavour (35.1-38.9) were found to have the second highest score and was both negatively correlated with the overall aroma intensity (r = -0.631; p = 0.001 and r = -0.603; p = 0.002, respectively). Therefore, a high overall aroma intensity can rather be used as an indicator of game-like aroma/flavour intensities and a low overall aroma intensity as an indicator of beef-like aroma/flavour of plains zebra meat. Furthermore, moderate to positive correlations were also found between overall aroma and sensory attributes such as liver-like aroma, metallic aroma and flavour, fatty aroma, and sweet associated taste (Addendum I). The correlations between aromas and flavours indicate that flavour is related to the aroma released in the mouth during the consumption of game meat (Listrat et al., 2016; Radder & Le Roux, 2005) and therefore it was only necessary to test for overall aroma intensity instead of overall flavour intensity as well. The game-like aroma in the plains zebra can be characterised as a combination of both liver-like and metallic aroma which were strongly correlated with game-like aroma (r = 0.670; p = 0.000 and r = 0.700; p = 0.000, respectively). Similar, game-like flavour was also strongly correlated with liver-like flavour (r = 0.622; p = 0.002) and moderately correlated with metallic flavour (r = 0.470; p = 0.024). Liver-like and metallic aroma and flavour are undesirable sensory attributes and perceived negatively by consumers; however, both liver-like aroma and flavour scores in the plains zebra muscles were low, and should not have any major effect on the sensory profile of plains zebra meat. Similar correlations regarding liver-like and metallic aroma were reported for eland (Needham et al., 2019) and blesbok meat (Neethling, 2016). The score for game-like flavour intensity obtained for the plains zebra LTL (67.9 ± 1.00; Table 6.4) is slightly higher than that of blue wildebeest (63.0 ± 0.57; Van Heerden, 2018), gemsbok (60.4 ± 1.31), red hartebeest (61.7 ± 1.27), impala (62.6 ± 1.25) and kudu (62.1 ± 1.10) and lower than found for blesbok (70.7 ± 1.09) and springbok (76.5 ± 1.26; Neethling, 2016).

Muscle type had a significant influence on the beef-like flavour. However, tendencies towards muscle type differences for aromas and flavour were observed in overall aroma, game-like aroma and flavour, beef-like aroma, liver-like flavour, and sweet-associated taste. The DA plot (Figure 6.2a) clearly indicates the separation between the BF and both the LTL and SM muscles on the vertical axis of the plot. The slight overlap between the LTL and SM support these tendencies towards significance in some sensory attributes. The LTL and SM muscles are grouped on the left side of the DA plot, the LTL is grouped in the second quadrant (top left) and the SM in the third quadrant (bottom left) both slightly overlapping the horizontal axis. The BF, on the other hand, is grouped in the centre over the horizontal midline of the right quadrants. The discriminant loadings plot (Figure 6.2b) classifies the sensory

attributes while the DA plot (Figure 6.2a) illustrate the association of the sensory attributes to the different treatments (muscle types).

The tenderness of meat is one of the main deciding factors for re-purchasing by consumers as it is the most important parameter in determining meat quality. According to the p-value generated by the ANOVA analysis, significant differences existed between muscle type for tenderness ($p = 0.03$) and residue ($p = 0.01$). The BF muscle had the lowest tenderness and the highest score for residue, and thus, as expected, a very strong negative correlation between tenderness and residue existed ($r = -0.919$; $p < 0.001$). The DA plot of the observations (Figure 6.2a and b) supports the significant indications as the BF muscle is clearly separated from the LTL and SM muscle and is rather associated with the attributes on the right side of the figure. Also, a strong positive correlation existed between tenderness scores and mealiness ($r = 0.675$; $p < 0.001$) and a strong negative correlation existed between residue and mealiness ($r = -0.714$, $p < 0.001$). These correlations indicate that less tender plains zebra meat will have more residue but will be less mealy. Even though, both residue and mealiness are considered to be a negative sensory attribute, the scores for both in all three muscle types were low and can thus be postulated not to impact on the tenderness of plains zebra meat significantly. The residue and mealiness scores in the plains zebra was lower than found for other large-bodied game species such as eland (Needham et al., 2019) and blue wildebeest (Van Heerden, 2018).

No significant differences were observed between muscle types in terms of WBSF values in previously frozen meat. A negative non-significant correlation ($r = -0.328$) between the shear force and tenderness rating by the sensory panel were found. A similar non-significant correlation was also observed for blue wildebeest ($r = -0.341$; Van Heerden, 2018), which is contradictory to findings of previous studies that reported significant negative correlations between sensory tenderness scores and WBSF for springbok (Hoffman, Kroucamp, & Manley, 2007; North & Hoffman, 2015), eland (Needham et al., 2019) and impala (Engels, 2019). This trend may indicate that WBSF is not always a reliable predictor of sensory tenderness (Van Heerden, 2018).

The mean WBSF values measured for the LTL (42.6 ± 3.63 N), SM (43.8 ± 4.00 N) and BF (47.6 ± 3.63 N) muscles during the sensory analysis (Table 6.3) were substantially lower than those measured for fresh meat in Chapter 4 (54.6 ± 2.89, 62.3 ± 2.88 and 62.5± 2.87 N, respectively). The lower shear force values observed can be a consequence of freezing the meat, a longer thawing period (36 hours) and different cooking methods used between fresh and the previously frozen meat. Even though only the LTL muscle of the plains zebra had WBSF values below the 42.9 N threshold for tender meat, the SM and BF plains zebra muscles were only slightly higher and were still below the 52.7 N threshold to be classified as tough (Destefanis, Brugiapaglia, Barge, & Dal Molin, 2008). The SM and BF can, therefore, be classified as being intermediately tender.

Noticeably high sensory scores for sweet-associated aroma and taste was observed for plains zebra meat in comparison to game species such as impala (Engels, 2019; Neethling, 2016) springbok (Neethling, 2016; Neethling, Muller, van der Rijst, & Hoffman, 2018), gemsbok, blesbok, red hartebeest, and kudu (Neethling, 2016). Sweet-associated aroma and taste have been positively correlated to consumer liking of meat (Oltra et al., 2015), thus contributing to meat flavour (Spanier, Flores, McMillin, & Bidner, 1997). A higher score for sweet taste in red meat has been found to be linked to high pH_u

values associated with dark, firm and dry (DFD) meat (Byrne et al., 2001; Flores, Armero, Aristoy, & Toldra, 1999), however, the pH_u values of plains zebra meat in this study fell into the biologically normal range with an average of 5.7 (Table 6.3). No correlations between sweet-associated taste and pH_u were found in plains zebra meat. Volatile aroma compounds associated with sweet-associated aroma and taste in red meat are hexanal, heptanal, nonanal, 2,3-butanedione, butanoic acid and 2-pentylfuran (Madruga, Elmore, Oruna-Concha, Balagiannis, & Mottram, 2010; Specht & Baltes, 1994). Volatile aroma compounds in plains zebra meat have not yet been studied warranting further research. Sweet-associated sensory attributes in lamb have been associated with concentrations of glucose, adenosine monophosphate, inosine and to a lesser extent glucose-6-phosphate and inosine monophosphate, as these nucleotides are precursors for ribose which in its turn participates in the formation of meaty aroma compounds (Oltra et al., 2015). Equine meat is relatively rich in glycogen with initial glycogen levels in the muscles of horse reported to be double compared to beef (Lawrie & Ledward, 2006). Muscle tissues initially rich in glycogen will have a high concentration of residual glycogen and glucose in the muscle tissue post-rigor (Gill, 2005), which has been reported to possibly add to the higher sensory scores for sweet-associated attributes in horse meat (Litwińczuk, Florelk, Skałecki, & Litwińczuk, 2008; Stanislawczyk & Znamirowska, 2005). The LTL muscle of adult horse stallions was found to have a higher score for sweet smell (41.4 ± 2.42; Diaconu, Lazăr, Găină Diaconu, Ciobanu, & Boişteanu, 2015) than observed for the plains zebra meat in this study (22.8 ± 0.67; Table 6.4). Therefore, it will be of value to establish the glycogen concentrations and mentioned meaty aroma precursors and its association with the formation of sweet-associated sensory attributes in plains zebra and horse meat as studies are non-existent or limited. Therefore, it can be postulated that the plains zebra has higher energy reserves in their muscles than other game species which could, therefore, have contributed to a higher sweet associated-aroma and taste observed in the selected muscles. Furthermore, the sweet-associated aroma in this study had moderate negative correlations with palmitoleic acid, PUFA, n-3 PUFA and PUFA:SFA ratio (Addendum I). These negative correlations indicate that a higher intensity of sweet-associated aroma will lead to a decrease in palmitoleic acid, PUFA, n-3 PUFA and PUFA:SFA ratio in plains zebra meat. The sweet-associated taste was negatively correlated with hexanoic (C6:0), docosahexaenoic (C22:6n3) and positively correlated with the n6:n3 ratio in the plains zebra meat (Addendum I).

The high herbaceous sensory scores of the plains zebra meat may be associated with the vegetation type found on farm which consisted of grasses and fragrant Fynbos vegetation that comprised the diet of these animals. Higher ratings of herbaceous scores were also observed in extensively produced impala on the same vegetation type as the plains zebra in comparison with semi-extensive and intensive impala on Central Sandy Bushveld (Engels, 2019). No correlations between the herbaceous attributes and any fatty acids existed in this study. Distinctive herbaceous sensory characteristics were observed in the meat of extensively produced South African Dorper lambs as a result of their dietary regime, which consisted of aromatic Karoo bushes and shrubs (Erasmus, Hoffman, Muller, & van der Rijst, 2016). High concentrations of terpenes (volatile compounds with a strong aroma) in the Karoo vegetation were found to cause the unique herbaceous flavour in the meat of these Karoo lambs (Erasmus et al., 2017). The intense herbaceous characteristics in the plains zebra can thus also

be due to similar volatile compounds occurring in the digested Fynbos vegetation. However, as the fatty acid and volatile compounds of the natural vegetation and grasses consumed by the plains zebras were not determined, this statement can only be postulated, thus highlighting an area for future research.

As only one comparative study on the fatty acid profile of plains zebra harvested in the northern Bushveld (Limpopo Province, South Africa) during the winter (summer-rainfall region) could be found, it was thus similarly hypothesised that the fatty acid profile of the intramuscular fat will reflect the fatty acid profiles of the diet of the animals (Hoffman et al., 2016). The intramuscular fat and the fatty acid content was the highest in the BF when compared to that of both the LTL and SM muscles, which can be attributed to the anatomical location, activity level, metabolic nature and the contractile type of the muscle fibres found in this muscle (Lefaucheur, 2010). The predominate class of fatty acids in the intramuscular fat of the LTL, SM and BF muscles of the plains zebra in this study was SFA (52.1 ± 2.93 %, pooled mean) followed by PUFA (32.9 ± 1.86 %, pooled mean) and then MUFA (15.0 ± 1.59 %, pooled mean). In contrast, Hoffman et al. (2016) observed PUFA as the predominant fatty acid group in plains zebra LL with a wide range of 31.40-46.67 % and lower amounts of SFA (41.01 %, mean) than found in this study. The higher SFA's found for the muscles in this study can be attributed to the detection of hexanoic (C6:0), caprylic (C8:0) capic (C10:0), and lauric acid (C12:0) which was not analysed in the study of Hoffman et al. (2016). These fatty acids accounted for approximately 16.1 %, 15.0 % and 13.5 % of the SFA's in the muscles respectively in the present study. The resulted total MUFA of the LTL was in line with the LL measured in the previous study (Hoffman et al., 2016). The primary fatty acids present in the intramuscular fat of the plains zebra muscles in descending order were palmitic (C16:0; 25.6 ± 1.84 % pooled mean), stearic (C18:0; 12.7 ± 1.22 % pooled mean), oleic (C18:1n9c; 10.1 ± 1.05 % pooled mean) and linoleic (C18:2n6c; 8.9 ± 1.27 % pooled mean) acids. Palmitic, stearic, oleic, linoleic, and linolenic acids in the LTL fell into the lower end of the range observed for the plains zebra LL by Hoffman et al. (2016). Moreover, a higher intramuscular fat content in the plains zebra muscles was linked to a higher oleic acid content ($r = 0.520$; $p = 0.009$). The fatty acid profile of the plains zebra might differ between production system and regions being related to rainfall patterns, nutritional quality, and density of feed between seasons and also the suitability of the plains zebra to the specific area.

Stearic acid, classified as an SFA, is a desirable fatty acid as it is converted to oleic acid in the human body (Bender, 1992) and therefore it does not influence the plasma cholesterol levels unlike the other fatty acids in this group (Wood et al., 2008). All unsaturated fatty acids (MUFA and PUFAs) are also seen as desirable fatty acids as they can decrease the LDL cholesterol levels in the blood (Wood, Enser, Richardson, & Whittington, 2008). Stearic acid is the second largest SFA in the plains zebra LTL, SM and BF muscles indicating that 23.4-32.2% of the SFA in all three muscles in this study is desirable to consumers. The desirable fatty acid content, which is the combined value of stearic acid, MUFA and PUFA comprise 63.9 %, 57.4 % and 60.4 % of all fatty acids in the plains zebra LTL, SM and BF, respectively. The plains zebra meat can, therefore, be considered as a healthy food commodity with the capability of reducing plasma cholesterol levels (Hoffman & Ferreira, 2004).

The MUFA and PUFA were only significantly influenced by muscle type with regards to oleic acid (C18:1n9c), total MUFA and α-linolenic acid (C18:3n3) being significantly lower in the SM muscle.

Oleic acid predominated the MUFA class in the LTL, SM and BF muscles contributing 66.3%, 66.1% and 68.9% to the total MUFA of the respective muscles. Similarly, oleic acid was also the predominant MUFA in the previous study on the plains zebra LL comprising 89 % of the total MUFA (Hoffman et al., 2016). The n3 essential fatty acid α-linolenic acid (4.8 ± 0.83 %, pooled mean) in this study was found to be much lower compared to plains zebras harvested in the Limpopo Province, South Africa (11.78 ± 4.33 %; Hoffman et al., 2016). Alpha-linolenic acid can be elongated to long-chain n3 PUFAs namely eicosapentaenoic acid (C20:5n3) and docosahexaenoic acid (C22:6n3), which may explain the higher docosahexaenoic acid (5.4 ± 0.77 %, pooled mean) in the plains zebra meat in this study compared to 0.60 ± 0.24 % in the study by Hoffman et al., (2016). These differences may be related to the differences in the diet as the plains zebras in this study were harvested in the Fynbos biome with Central Rûens Shale Renosterveld vegetation and the plains zebra from the mentioned study in the Savannah biome northern Bushveld bioregion. It should be highlighted that the α-linolenic acid was much lower ($p = 0.003$) and docosadienoic acid (C22:2n6) was much higher ($p = 0.128$) in the SM compared to the other two muscle types. Docosadienoic acid (C22:2n6) was found to have a negative correlation ($r = -0.679$, p) to α-linolenic acid and consequently, plains zebras with lower α-linolenic acid contents will have higher docosadienoic acid contents, as observed in the plains zebra SM muscle.

Muscle type did not influence the PUFA:SFA ($p = 0.144$), n6-PUFA ($p=0.767$), n3-PUFA ($p = 0.098$), and n6:n3 PUFA ($p = 0.430$), however, the SM muscle had the tendency to be lower in n3-PUFA content which can be explained by the lower ($p = 0.003$) α-linolenic acid contents in the SM muscle as discussed. The PUFA:SFA ratio of the plains zebra muscles in this study were lower than found for the plains zebras studied by Hoffman et al., (2016), however, the n6-PUFA, n3-PUFA, and n6:n3 PUFA were comparable as it fell into the lower end of the range reported by the author. It is recommended that the PUFA:SFA ratio be a minimum of 0.4, with meat usually having a ratio of 0.1 (Wood et al., 2003). It was also recommended by the British Department of Health (1994) for the n6:n3 PUFA ratio to have a 4:1 upper limit. All three plains zebra muscles had a PUFA:SFA ratio above the recommended minimum (0.7 ± 0.06; pooled mean) and an n6:n3 PUFA below the recommended maximum (1.5 ± 0.33; pooled mean). The n6:n3 ratio found in the plains zebra LTL muscles in the present study was lower than found for the same muscle in game species such as blue wildebeest (4.2 ± 1.37; Van Heerden, 2018), eland (1.45 ± 0.15; Needham et al., 2019), kudu (2.22 ± 0.47; Hoffman, Mostert, & Laubscher, 2009) springbok (2.86 ± 0.19; Neethling et al., 2018) and impala (2.67-3.76; Hoffman et al., 2009 and Neethling, 2016). The n6:n3 PUFA ratio in the plains zebra BF was higher than recorded for eland BF (1.33 ± 0.11; Needham et al., 2019). With regards to the intramuscular fat content, the PUFA:SFA and the n6:n3 PUFA ratio, plains zebra meat in this study can also be considered and promoted as a health food commodity.

6.5 CONCLUSION

Muscle type had minor influences on the sensory properties of plains zebra meat, influencing only beef-like flavour, tenderness, and residue. Game-like aroma had the highest contribution to overall aroma intensity in plains zebra meat with game-like aroma defined as a combination of liver-like and metallic aroma. Liver-like and metallic aroma and flavour are undesirable sensory attributes negatively

perceived by consumers; fortunately, the score for liver-like sensory attributes was low and should not have any significant effects on the sensory profile of plains zebra meat. Overall aroma intensity was positively correlated with game-like aroma and negatively correlated with beef-like aroma, indicating that a high overall aroma can be associated with game-like sensory attributes. The high score for sweet-associated sensory attributes of the plains zebra meat may be caused by volatile compounds or by a high post-rigor muscle glucose level. The high score for herbaceous sensory attributes may be caused by the fragrant Fynbos vegetation consumed by the plains zebra. Therefore, it is recommended that future studies focus on the volatile compounds of the diet and the meat of the plains zebra to determine the correlation between the vegetation of the area and both the sweet-associated and herbaceous flavour in plains zebra meat. It is also recommended that the glucose concentration in plain zebra meat is determined to clarify its association, if any, to the sweet-associated sensory attributes. It was further observed that plains zebra meat classified as tender will have low amounts of residue but will be mealier than less tender meat. Nevertheless, the scores for residue and mealiness were low, possibly not influencing the sensory profile significantly.

Only a few fatty acid contents were influenced by muscle type with little correlations between the most abundant fatty acids and sensory attributes. The intramuscular fat and fatty acid content were significantly higher in the BF muscle and did not differ between the LTL and SM muscle. The plains zebra meat exhibited a desirable fatty acid profile with both the PUFA:SFA and n6:n3 PUFA ratios meeting the recommended guidelines set by the British Department of Health (1994). The low intramuscular fat content observed in all three muscle types in combination with the adequate PUFA:SFA and n6:n3 PUFA ratios promotes the marketing of plains zebra as an alternative healthy lean red meat source. This study aimed to generated baseline data on the sensory and fatty acid profile of three muscle types in the plains zebra, however, further research is still essential to evaluate the impact of intrinsic (slaughter age, sex and more muscle types) and extrinsic (seasons, dietary regime and location) factors on the sensory and fatty acid profile. It can also be recommended to use a larger samples size for improved quantification of these attributes and correlations.

6.6 REFERENCES

AMSA. (2015). Research guidelines for cookery, sensory evaluation, and instrumental tenderness measurements of meat. In *Chicago, USA: National Livestock and Meat Board.* Retrieved from http://www.meatscience.org

Bender, A. (1992). Meat and meat products in human nutrition in developing countries. In *FAO Food and Nutrition Paper (FAO)*. Retrieved from http://agris.fao.org/agris-search/search.do?recordID=XF9328643

Bothma, J. du P., & Du Toit, J. G. (Edss.). (2016). *Game ranch management* (6th ed). Pretoria: Van Schaik.

Byrne, D. V., Bredie, W. L. P., Bak, L. S., Bertelsen, G., Martens, H., & Martens, M. (2001). Sensory and chemical analysis of cooked porcine meat patties in relation to warmed-over flavour and pre-slaughter stress. *Meat Science*, *59*(3), 229–249. https://doi.org/10.1016/S0309-1740(01)00072-9

Destefanis, G., Brugiapaglia, A., Barge, M. T., & Dal Molin, E. (2008). Relationship between beef consumer tenderness perception and Warner-Bratzler shear force. *Meat Science*. https://doi.org/10.1016/j.meatsci.2007.05.031

Diaconu, E. C., Lazăr, R., Găină (Diaconu), N., Ciobanu, M. M., & Boişteanu, P. C. (2015). Research on assesment of sensory horse meat sluagtered in NE region. *Lucrări Ştiinţifice - Seria Zootehnie*, *16*, 118–123.

Engels, R. A. (2019). *Ante- and post-mortem factors influencing impala (Aepyceros melampus) meat quality*. Unpublished Master's thesis in Animal Science, Stellenbosch, South Africa.

Erasmus, S. W., Hoffman, L. C., Muller, M., & van der Rijst, M. (2016). Variation in the sensory profile of South African Dorper lamb from extensive grazing systems. *Small Ruminant Research*, *144*, 62–74. https://doi.org/10.1016/j.smallrumres.2016.07.020

Erasmus, S. W., Muller, M., Alewijn, M., Koot, A. H., van Ruth, S. M., & Hoffman, L. C. (2017). Proton-transfer reaction mass spectrometry (PTR-MS) for the authentication of regionally unique South African lamb. *Food Chemistry*, *233*, 331–342. https://doi.org/10.1016/j.foodchem.2017.04.131

Flores, M., Armero, E., Aristoy, M. C., & Toldra, F. (1999). Sensory characteristics of cooked pork loin as affected by nucleotide content and post-mortem meat quality. *Meat Science*, *51*(1), 53–59. https://doi.org/10.1016/S0309-1740(98)00097-7

Gill, C. O. (2005). Safety and storage stability of horse meat for human consumption. *Meat Science*, *71*(3), 506–513. https://doi.org/10.1016/j.meatsci.2005.04.030

Hoffman, L. C., & Ferreira, A. V. (2004). Chemical composition of two muscles of the common duiker (Sylvicapra grimmia). *Journal of the Science of Food and Agriculture*, *84*(12), 1541–1544. https://doi.org/10.1002/jsfa.1813

Hoffman, L. C., Geldenhuys, G., & Cawthorn, D. M. (2016). Proximate and fatty acid composition of zebra (*Equus quagga burchellii*) muscle and subcutaneous fat. *Journal of the Science of Food and Agriculture*, *96*(11), 3922–3927. https://doi.org/10.1002/jsfa.7623

Hoffman, L. C., Kroucamp, M., & Manley, M. (2007). Meat quality characteristics of springbok (*Antidorcas marsupialis*). 4: Sensory meat evaluation as influenced by age, gender and production region. *Meat Science*, *76*(4), 774–778. https://doi.org/10.1016/J.MEATSCI.2007.02.020

Hoffman, L. C., Mostert, A. C., & Laubscher, L. L. (2009). Meat quality of kudu (*Tragelaphus strepsiceros*) and impala (Aepyceros *melampus*): The effect of gender and age on the fatty acid profile, cholesterol content and sensory characteristics of kudu and impala meat. *Meat Science*, *83*(4), 737–743. https://doi.org/10.1016/j.meatsci.2009.08.026

Hoffman, L. C., Muller, M., Schutte, D. W., & Crafford, K. (2004). The retail of South African game meat: current trade and marketing trends. *South African Journal of Wildlife Research*, *34*(2), 123–134. https://doi.org/http://hdl.handle.net/10520/EJC117194

Hoffman, L. C., & Wiklund, E. (2006). Game and venison - meat for the modern consumer. *Meat*

Science, *74*(1), 197–208. https://doi.org/10.1016/j.meatsci.2006.04.005

Honikel, K. O. (2004). Chemical and physical characteristics of meat - pH Measurement. In *Encyclopedia of Meat Sciences* (pp. 238–242). https://doi.org/10.1017/CCO9780511791581.016

Lawless, H. T., & Heymann, H. (2010). *Sensory Evaluation of Food: Principles and Practices* (2nd ed.) [Book]. https://doi.org/10.1007/978-1-4419-6488-5

Lawrie, R. A., & Ledward, D. A. (2006). *Lawrie's meat science* (7th ed). CRC Press.

Lefaucheur, L. (2010). A second look into fibre typing - Relation to meat quality. *Meat Science*, Vol. 84, pp. 257–270. https://doi.org/10.1016/j.meatsci.2009.05.004

Leygonie, C., Britz, T. J., & Hoffman, L. C. (2012a). Impact of freezing and thawing on the quality of meat: Review. *Meat Science*, *91*(2), 93–98. https://doi.org/10.1016/j.meatsci.2012.01.013

Leygonie, C., Britz, T. J., & Hoffman, L. C. (2012b). Meat quality comparison between fresh and frozen/thawed ostrich M. iliofibularis. *Meat Science*, *91*(3), 364–368. https://doi.org/10.1016/j.meatsci.2012.02.020

Listrat, A., Lebret, B., Louveau, I., Astruc, T., Bonnet, M., Lefaucheur, L., … Bugeon, J. (2016). How muscle structure and composition influence meat and flesh quality. *Scientific World Journal*, Vol. 2016. https://doi.org/10.1155/2016/3182746

Litwińczuk, A., Florelk, M., Skałecki, P., & Litwińczuk, Z. (2008). Chemical composition and physiochemical properties of horse meat from the *Longissimus lumborum* and *semitendinososus* muscle. *Journal of Muscle Foods*, *19*(3), 223–236. https://doi.org/10.1111/j.1745-4573.2008.00117.x

Lyman Ott, R., & Longnecker, M. (2010). *An Introduction to Statistical Methods and Data Analysis*. Alexandria: Canada: Brooks/Cole, Cengage Learning.

Madruga, M. S., Elmore, J. S., Oruna-Concha, M. J., Balagiannis, D., & Mottram, D. S. (2010). Determination of some water-soluble aroma precursors in goat meat and their enrolment on flavour profile of goat meat. *Food Chemistry*, *123*(2), 513–520. https://doi.org/10.1016/j.foodchem.2010.04.004

Marescotti, M. E., Caputo, V., Demartini, E., & Gaviglio, A. (2019). Discovering market segments for hunted wild game meat. *Meat Science*, *149*, 163–176. https://doi.org/10.1016/j.meatsci.2018.11.019

Marinova, D., & Bogueva, D. (2019). Planetary health and reduction in meat consumption. *Sustainable Earth*, *2*(1), 1–12. https://doi.org/10.1186/s42055-019-0010-0

Mucina, L., & Rutherford, M. C. (2006). The vegetation of South Africa, Lesotho and Swaziland. *Strelitzia 19*, 1–30. https://doi.org/10.1007/s

Needham, T., Laubser, J. G., Kotrba, R., Bureš, D., Smyth, H., & Hoffman, L. C. (2019). Sensory characteristics of the *Longissimus thoracis et lumborum* and *biceps femoris* muscles from male

and female common eland (*Taurotragus oryx*). *Meat Science*. https://doi.org/10.1016/j.meatsci.2019.107918

Neethling, J. (2016). *Factors influencing the flavour of the meat derived from South African game species*. Unpublished Doctor's thesis in Food Science, Stellenbosch, South Africa

Neethling, J., Muller, M., van der Rijst, M., & Hoffman, L. C. (2018). Sensory quality and fatty acid content of springbok (Antidorcas marsupialis) meat: influence of farm location and sex. *Journal of the Science of Food and Agriculture*, *98*(7), 2548–2556. https://doi.org/10.1002/jsfa.8743

North, M. K., & Hoffman, L. C. (2015). Changes in springbok (*Antidorcas marsupialis*) *Longissimus thoracis et lumborum* muscle during conditioning as assessed by a trained sensory panel. *Meat Science*, *108*, 1–8. https://doi.org/10.1016/j.meatsci.2015.05.004

Oltra, O. R., Farmer, L. J., Gordon, A. W., Moss, B. W., Birnie, J., Devlin, D. J., … Farrell, D. (2015). Identification of sensory attributes, instrumental and chemical measurements important for consumer acceptability of grilled lamb Longissimus lumborum. *Meat Science*, *100*, 97–109. https://doi.org/10.1016/j.meatsci.2014.09.007

Powell, E. (2014). Zebra meat: Exotic and lean - But does it taste good? Retrieved November 14, 2019, from Independent website: https://www.independent.co.uk/life-style/food-and-drink/zebra-meat-exotic-and-lean-but-does-it-taste-good-9623700.html

Radder, L., & Le Roux, R. (2005). Factors affecting food choice in relation to venison: A South African example. *Meat Science*, *71*(3), 583–589. https://doi.org/10.1016/j.meatsci.2005.05.003

Radnitz, C., Beezhold, B., & DiMatteo, J. (2015). Investigation of lifestyle choices of individuals following a vegan diet for health and ethical reasons. *Appetite*, *90*, 31–36. https://doi.org/10.1016/j.appet.2015.02.026

Shapiro, A. S. S., & Wilk, M. B. (1965). An analysis of variance test for normality (complete samples). *Biometrika* 52, 591-611.

Spanier, A. M., Flores, M., McMillin, K. W., & Bidner, T. D. (1997). The effect of post-mortem aging on meat flavor quality in Brangus beef. Correlation of treatments, sensory, instrumental and chemical descriptors. *Food Chemistry*, *59*(4), 531–538. https://doi.org/10.1016/S0308-8146(97)00003-4

Specht, K., & Baltes, W. (1994). Identification of volatile flavor compounds with high aroma values from shallow-fried beef. *Journal of Agricultural and Food Chemistry*, *42*(10), 2246–2253. Retrieved from http://ci.nii.ac.jp/naid/10030334007/en/

Stanislawczyk, R., & Znamirowska, A. (2005). Changes in physio-chemical properties of horsemeat during frozen storage. *Acta Scientiarum Polonorum Technologia Alimentaria*, *4*(2), 89–96.

Van Heerden, A. M. (2018). *Profiling the meat quality of blue wildebeest (Connochaetes taurinus)*. Unpublished Master's thesis in Animal Science, Stellenbosch, South Africa.

Van Huis, A. (2016). Edible insects are the future? *Proceedings of the Nutrition Society*, *75*(3), 294–305. https://doi.org/10.1017/S0029665116000069

Von Massow, M., Weersink, A., & Gallant, M. (2019). Meat consumption is changing but it's not because of vegans. Retrieved November 12, 2019, from The Converstation website: https://theconversation.com/meat-consumption-is-changing-but-its-not-because-of-vegans-112332

Wassenaar, A., Kempen, E., & van Eeden, T. (2019). Exploring South African consumers' attitudes towards game meat—Utilizing a multi-attribute attitude model. *International Journal of Consumer Studies*, (February), 437–445. https://doi.org/10.1111/ijcs.12523

Wiklund, E., Manley, T. R., Littlejohn, R. P., & Stevenson-Barry, J. M. (2003). Fatty acid composition and sensory quality of Musculus *Longissimus* and carcass parameters in red deer (Cervus elaphus) grazed on natural pasture or fed a commercial feed mixture. *Journal of the Science of Food and Agriculture*, *83*(5), 419–424. https://doi.org/10.1002/jsfa.1384

Wood, J. ., Enser, M., Fisher, A. . ., Nute, G. R., Richardson, R. I., & Sheard, P. R. (1999). Animal Nutrition and Metabolism Group Symposium on "Improving meat production for future needs" Manipulating meat quality and composition. *Proceedings of the Nutrition Society*, *58*, 363–370. https://doi.org/10.1017/S0029665199000488

Wood, J. D., & Enser, M. (1997). Factors influencing fatty acids in meat and the role of antioxidants in improving meat quality. *British Journal of Nutrition*, *78*(1), S49–S60. https://doi.org/10.1079/bjn19970134

Wood, J. D., Enser, M., Fisher, A. V., Nute, G. R., Sheard, P. R., Richardson, R. I., … Whittington, F. M. (2008). Fat deposition, fatty acid composition and meat quality: A review. *Meat Science*, *78*(4), 343–358. https://doi.org/10.1016/j.meatsci.2007.07.019

Wood, J. D., Enser, M., Richardson, R. I., & Whittington, F. M. (2008). Fatty Acids in Meat and Meat Products. In C. K. Chow (Ed.), *Fatty Acids in Foods and their Health Implications* (3rd ed., pp. 87–107). https://doi.org/10.1201/9781420006902.ch5

Wood, J. D., Richardson, R. I., Nute, G. R., Fisher, A. V., Campo, M. M., Kasapidou, E., … Enser, M. (2003). Effects of fatty acids on meat quality: A review. *Meat Science*, 66, 21–32. https://doi.org/10.1016/S0309-1740(03)00022-6

World Economic Forum. (2019). *Meat: The Future Series - Alternative Proteins | World Economic Forum*. 31.

CHAPTER 7

POST-MORTEM AGEING OF THE THREE PRIMAL MUSCLES FROM THE PLAINS ZEBRA (E*quus quagga)*

ABSTRACT

The aim of this study was to establish the optimum ageing period for the *Longissimus thoracis et lumborum* (LTL), *semimembranosus* (SM) and *biceps femoris* (BF) muscles of plains zebra (*Equus quagga)* stallions. Eight animals were harvested in the winter season at Prinskraal farm Bredasdorp and another twelve animals in the summer season at Elandsberg Nature Reserve-Bartholomeus Klip near Hermon in the Western Cape Province of South Africa. The ageing trial was first conducted on the muscles obtained from the winter-harvested group but due to a lack of decrease in meat tenderness the trial was elongated and repeated on the muscles obtained from the summer-harvested animals. The muscles from the winter-harvested group was vacuum-aged at 4°C for 3, 5, 9, 12, 14, 16, 18, 20, 22 and 24 days, whilst the muscles from the summer-harvested group for 1,5, 9,12, 14, 16, 20, 24, 28 and 32 days. The cumulative purge loss increased from 3.0 % to 4.5 % over the 32-day ageing period. The pH increased and the cooking loss decreased specifically from day 24 onwards. Significant interactions were observed between muscle type and ageing period for CIE a*, CIE b*, hue-angle and chroma. Meat surface colour was less bright (with one unit) on day 1, 16 and 32 and became more red and yellow until day 14 and 16, respectively. Hue-angle values did not change over time, whereas chroma continued to increase until day 14 as well. Tenderness increased significantly for the LTL, SM and BF muscle over the ageing period. It seems that no special distinction is needed between the LTL, SM and BF muscles as all three muscles reached their maximum tenderness at day 20 post-mortem. Therefore, an ageing period of 20 days can be recommended to obtain optimum tenderness (34.1 ± 2.36 N; pooled mean), lightness (35.1 ± 0.63; pooled mean), and redness (14.2 ± 0.47; pooled mean) even though the moisture loss was still at is highest on day 20 (weep loss = 4.0 ± 0.31 %; cooking loss = 38.4 ± 0.33 %).

Keywords: Plains zebra, Post-mortem ageing, Tenderness, Game meat

7.1 INTRODUCTION

The *Longissimus thoracis et lumborum* (LTL), *semimembranosus* (SM) and *biceps femoris* (BF) muscles are among the largest and primal muscles on the carcasses of game species, and this presents an opportunity to exploit game species as an alternative red meat source as it can be sold fresh or processed into value-added products. The physical-chemical composition of each muscle type differs based on the anatomical location and thus physiological function, which in turn influence meat quality parameters and thus consumer experience (Ba, Park, Dashmaa, & Hwang, 2014). The eating quality of meat is evaluated by consumers in terms of colour, tenderness, and healthiness. These meat parameters can vary between muscle type and species and are also influenced by various ante-mortem and post-mortem factors. Arguably, meat tenderness is the most important quality characteristic as consumers are willing to pay a premium price for guaranteed tenderness (Miller, Ramsey, Hoover, Carr, & Crockett, 2001). However, on the point of purchase meat colour is the most important aspect for the consumer, for meat colour is considered an indicator for meat freshness.

The common perception that game meat has an unpleasant game-like flavour while being a dark, tough and dry meat can potentially be ascribed to a difference in hunting and processing protocols, as well as cooking methods, which result in non-uniform products and misperceptions about the quality among consumers (Radder & Le Roux, 2005). Recently, the focus of many game meat research studies has been on the improvement of physical meat quality parameters as well as Warner-Bratzler shear force values (WBSF) in high valued muscles through mechanisms such as post-mortem ageing. Refrigerated vacuum-packed ageing is a common method used to improve the eating quality of meat (Ba et al., 2014), with the ageing period being influenced by muscle type, species, age, sex, animal feeding regime and degree of pre-slaughter stress (Nowak, 2011). An adequate ageing period of red meat generally improves the colour and tenderness of cuts, while increasing the water-soluble flavour precursors, characteristic flavour, and taste intensity. With the progression of post-mortem ageing technological meat quality parameters can also be negatively influenced, such as increased weep loss and cooking losses and the development of undesirable rancid flavour and needs to be considered when the correct ageing time for meat is considered.

Wet post-mortem ageing is a common ageing method used to increase tenderness of meat derived from domestic livestock species and has been shown to increase to improve tenderness in many game species such as springbok (North, Frylinck, & Hoffman, 2015), blue wildebeest (Van Heerden, 2018), impala (Engels, 2019), buffalo (Van As, 2019), and eland (Needham, Laubser, Kotrba, Bureš, & Hoffman, 2020). The plains zebra is another species that can benefit from post-mortem ageing due to the intermediate to high shear forces values observed 24- and 72-hours post-mortem in Chapter 4. The plains zebra can be seen as an alternative species for meat production as it has been found to produce good quality healthy meat (Chapter 4 and 5) with high yields from each animal culled due to the high dressing percentages (Chapter 3). Therefore, the purpose of this study was to assess the changes in the physical characteristics of the plains zebra LTL, SM and BF muscles during post-mortem ageing. The goal is to determine the ideal ageing period, which improves tenderness while maintaining the other physical meat quality parameters.

7.2 MATERIALS AND METHODS

7.2.1 Animals and study location

For the ageing trail, eight male plains zebras were obtained during the winter in June 2017 from Prinskraal farm, located near Bredasdorp in the Western Cape Province. Another 12 male plains zebras were obtained during the summer in January 2018 from Elandsberg Nature Reserve – Bartholomeus Klip, located near Hermon in the Western Cape Province. Both locations were situated in the Fynbos region and grazed primarily on Bermuda grass (*Cynodon dactylon*) and the natural vegetation. The vegetation type at Prinskraal farm was Central Rûens Shale Renosterveld and at Elandsberg Nature Reserve was Swartland Alluvium Fynbos. The 20 plains zebras harvested in total were free-roaming and in an extensive farming system. The plains zebras at Prinskraal were raised in a ~ 800 ha camp shared with 400 other animals derived from various game species for field optimisation. The plains zebras at Elandsberg Nature Reserve were moved four to five months prior to harvesting to 10 ha camps with four zebras per camp. The plains zebras obtained in the winter season was classified as mature > 5 years. The known ages of those harvested in the summer season ranged from two years to 13 years and two months of age. Refer to the Material and Methods of Chapter 3.2 for detailed information regarding the location and vegetation.

7.2.2 Plains zebra harvesting, dressing, and sampling

All the plains zebras from both seasons were harvested during the day with an appropriate rifle. Following the shot placement, the plains zebra carcasses were exsanguinated and transported with a designated vehicle to the on-farm slaughtering facility (Ethical clearance number: 10NP_HOF02). On arrival, the carcasses were weighed, skinned, eviscerated, and dressed according to standard guidelines described by Van Schalkwyk & Hoffman (2016). After evisceration, the carcasses were cut through the midline and again between the second and third last rib separating the carcasses into two hindquarters and two forequarters. Thereafter, each quarter was hung separately in a suspended manner in a mobile chiller at ± 4°C and transported to the Department of Animal Science at the University of Stellenbosch for sampling. Detailed information on the harvesting and slaughtering procedures can be found in Chapter 4.

Plains zebra carcasses from the winter and summer season had a refrigeration period of ± 24 hours and ± 72 hours, respectively, before deboning of a total of six selected muscles. For the ageing trial, only the left *Longissimus thoracis et lumborum* (LTL), separated as the *Longissimus thoracis* (LT) and *Longissimus lumborum* (LL), *semimembranosus* (SM) and *biceps femoris* (BF) were used. Each muscle was divided into 10 equally cut steaks perpendicular to the longitudinal axis into approximately two centimetres each for different ageing time points in the winter and summer season. The ageing time points are represented in Table 7.1 per season. Thereafter, each allocated steaks was individually weighed and vacuum-packed in plastic bags (70 µm nylon and polyethylene; oxygen permeability of 30 cm3/m2/24h/1atm, carbon dioxide permeability of 105 cm3/m2/24h/1atm and moisture vapour transfer rate of 2.2 g/m2/24h/1atm) under 5mb residual pressure (according to the pressure reading of the machine gauge; Multivac, Model C200, Sepp Haggenmuller, Wolfertschwenden, Germany) and stored

in a refrigerator at 0-4°C until its allocated ageing day for physical analysis. The samples allocated as day 1 for the winter season and allocated as day 3 for the summer season were analysed on the day of deboning and were not vacuum packed and stored.

Table 7.1 Summary of the ageing time points per season for the LTL, SM and BF muscles of zebra stallions.

n	Season	Ageing time points (days post-mortem)								
8	Winter	1	5	9	12	14	16	18	20	24
12	Summer	3	5	9	14	16	20	24	28	32

7.2.3 Physical analysis

Physical analysis of all three muscles were conducted on each of the ageing time points for the winter and summer season as stipulated in Table 7.1. The cumulative weep/purge loss percentage, cooking loss percentage, muscle pH, raw meat colour (CIE colour coordinates L*, a*, b*, hue-angle and chroma) and Warner-Bratzler shear force measurements were recorded as described in Chapter 5.

On each ageing time point the LTL, SM and BF were removed from its vacuum packaging and blotted dry by absorbent paper towel to remove the excess moisture. Each steak was individually weighed to record the “after” weight for the determination of cumulative weep/purge loss percentage as the “initial” weight was recorded prior to vacuum packaging. Consequently, the weep loss percentage was determined by the following equation:

Weep loss % = [(Initial weight – Final weight)/ (Initial weight)] *100

7.2.4 Statistical Analysis

The statistical analysis for each group (winter and summer) were analysed separately as a mixed model univariate analysis of variance (ANOVA). Plains zebra number (animal) was used as a random effect and muscle type, day, and all interaction terms as fixed effects. The statistical analysis was analysed with Statistica 64 version 13.4 (2018) VEPAC model. For post hoc testing, Fisher least significant differences (LSD) was used. A normal probability plot was drawn for each characteristic to determine any deviations from normality and possible outliers. Statistical differences were considered significant at a probability level of 5% ($p \leq 0.05$). The data are reported as least square means (LSMeans) and standard error for each characteristic as per muscle type and per post-mortem ageing day.

7.3 RESULTS

7.3.1 Winter group (2017)

The mean values for the physical parameters (pH, weep loss, cooking loss and WBSF) and colour coordinates (L*, a*, b*, hue-angle and chroma) of the LTL, SM and BF muscles obtained from the animals harvested in the winter season are presented per main effect in Table 7.3 and 7.4, respectively. The effect of muscle type (Table 7.2) was observed for almost all the parameters except for pH_u (5.77 ± 0.039, pooled mean), CIE b*(11.4 ± 0.51, pooled mean) and chroma values (18.8 ± 0.74; pooled mean). The LTL muscle had the lowest shear force value (42.1 ± 2.82 N) and the highest CIE L* (33.4 ± 0.69), the SM had the highest weep (4.4 ± 0.31 %) and cooking loss (37.2 ± 0.32 %) percentages and lastly the BF muscle had the highest CIE a* (15.6 ± 0.57) and lowest hue-angle values (36.11 ± 0.57) (Table 7.3). The effect of ageing days (Table 7.2) was observed for meat pH, WBSF, CIE a*, CIE b* and chroma values. The pH of the muscles followed a linear increasing trend until day 22 post-mortem whereas the CIE a* and b* values only increased from day 3 to day 5 and plateaued until the last ageing day (Table 7.3). The WBSF values reported on day 3 was significantly higher than the WBSF values reported for each of the remaining 9 time points, which did not differ from one another. As there was no apparent trend observed throughout the 24-day ageing period and as the muscles did not reach optimum tenderness it was deemed necessary to do a follow-up trial with an extended ageing period on the summer group.

Table 7.2 Individual and interactive effects of muscle type and days post-mortem on physical characteristics of plains zebra stallions harvested in the winter season.

Main experimental effect	**p-values**								
	pH_u	**Weep loss %**	**Cooking loss %**	**WBSF (N)**	**Colour coordinates**				
					L*	**a***	**b***	**Hue**	**Chroma**
Muscle type	0.523	**0.006**	**<0.001**	**<0.001**	**0.006**	**0.012**	0.960	**<0.001**	0.110
Day	**<0.001**	0.140	0.147	**0.003**	0.101	**<0.001**	**<0.001**	0.421	**<0.001**

Abbreviations: WBSF = Warner-Bratzler shear force

Table 7.3 LSMeans (± standard error) of the physical characteristics of plains zebra stallions harvested in the winter season as per muscle type and ageing period.

Main effects		pH	Weep loss %	Cooking loss %	WBSF (N)
Muscle Type	LTL	5.77 ± 0.039	3.6^{b} ± 0.31	33.3^{b} ± 0.32	42.1^{b} ± 2.82
	SM	5.76 ± 0.039	4.4^{a} ± 0.31	37.2^{a} ± 0.32	52.1^{a} ± 2.82
	BF	5.78 ± 0.039	3.9^{b} ± 0.31	36.4^{b} ± 0.32	52.1^{a} ± 2.82
Ageing period (days post-mortem)	3	5.71^{d} ± 0.041	*	36.4 ± 0.51	59.8^{a} ± 3.48
	5	5.74^{cd} ± 0.041	3.6 ± 0.36	36.5 ± 0.51	47.8^{b} ± 3.48
	9	5.77^{bc} ± 0.041	3.7 ± 0.36	35.5 ± 0.51	48.7^{b} ± 3.48
	12	5.77^{bc} ± 0.041	3.9 ± 0.36	36.4 ± 0.51	50.0^{b} ± 3.48
	14	5.77^{bc} ± 0.041	3.7 ± 0.36	35.4 ± 0.51	50.4^{b} ± 3.48
	16	5.77^{bc} ± 0.041	4.1 ± 0.36	34.9 ± 0.51	48.5^{b} ± 3.48
	18	5.81^{ab} ± 0.041	4.4 ± 0.36	35.2 ± 0.51	45.2^{b} ± 3.48
	20	5.83^{a} ± 0.041	4.2 ± 0.36	35.4 ± 0.51	45.5^{b} ± 3.48
	22	5.79^{ab} ± 0.041	4.3 ± 0.36	35.2 ± 0.51	48.5^{b} ± 3.48
	24	5.77^{bc} ± 0.041	4.1 ± 0.36	35.2 ± 0.51	43.8^{b} ± 3.48

$^{a-d}$Means with different letters in the same column (within a main effect) differ from each other ($p \leq 0.05$).

*No weep loss was measured on day one as physical analysis were conducted prior to packaging.

Abbreviations: WBSF = Warner-Bratzler shear force, LTL = *Longissimus thoracis et lumborum*, SM = *semimembranosus* and BF = *biceps femoris*

Table 7.4 LSMeans (± standard error) of the colour coordinates of plains zebra stallions harvested in the winter as per muscle type and ageing period.

Main effects		L*	a*	b*	Hue	Chroma
Muscle Type	LTL	33.4^{a} ± 0.69	14.4^{b} ± 0.57	11.4 ± 0.51	38.2^{a} ± 0.57	18.4 ± 0.74
	SM	32.1^{b} ± 0.69	14.7^{b} ± 0.57	11.4 ± 0.51	37.6^{a} ± 0.57	18.7 ± 0.74
	BF	32.4^{b} ± 0.69	15.6^{a} ± 0.57	11.5 ± 0.51	36.11^{b} ± 0.57	19.4 ± 0.74
Ageing period (days post-mortem)	3	32.6 ± 0.73	13.5^{c} ± 0.57	10.1^{c} ± 0.53	36.7 ± 0.63	16.9^{c} ± 0.75
	5	32.3 ± 0.73	14.8^{b} ± 0.57	11.1^{b} ± 0.53	36.7 ± 0.63	18.5^{b} ± 0.75
	9	32.0 ± 0.73	15.2^{ab} ± 0.57	11.6^{ab} ± 0.53	37.2 ± 0.63	19.1^{ab} ± 0.75
	12	32.1 ± 0.73	15.1^{ab} ± 0.57	11.3^{ab} ± 0.53	36.8 ± 0.63	18.9^{ab} ± 0.75
	14	32.5 ± 0.73	15.2^{ab} ± 0.57	11.6^{ab} ± 0.53	37.2 ± 0.63	19.2^{ab} ± 0.75
	16	33.3 ± 0.73	14.7^{b} ± 0.57	11.4^{ab} ± 0.53	37.8 ± 0.63	18.7^{ab} ± 0.75
	18	33.1 ± 0.73	15.4^{a} ± 0.57	11.7^{a} ± 0.53	37.1 ± 0.63	19.4^{a} ± 0.75
	20	33.1 ± 0.73	15.0^{ab} ± 0.57	11.8^{a} ± 0.53	38.1 ± 0.63	19.2^{ab} ± 0.75
	22	32.9 ± 0.73	15.3^{ab} ± 0.57	11.7^{a} ± 0.53	37.3 ± 0.63	19.3^{ab} ± 0.75
	24	32.7 ± 0.73	15.0^{ab} ± 0.57	11.8^{a} ± 0.53	38.1 ± 0.63	19.1^{ab} ± 0.75

^{a,b,c}Means with different letters in the same column (within a main effect) differ from each other ($p \leq 0.05$).

Abbreviations: WBSF = Warner-Bratzler shear force, LTL = *Longissimus thoracis et lumborum*, SM = *semimembranosus* and BF = *biceps femoris*

7.3.2 Summer group (2018)

Significant second-order interactions were observed between muscle type and ageing period (days post-mortem) for CIE a* (p = 0.023), CIE b* (p = 0.013), hue (p = 0.001) and chroma (p = 0.032) of plains zebra meat. No interaction between muscle type and ageing days post-mortem was observed for pH_u, weep loss percentage, cooking loss percentage, Warner-Bratzler shear force values and CIE L*, thus the main effects are interpreted separately for these characteristics. Furthermore, significant differences between the muscle types and ageing days post-mortem were found for all the physical characteristics except for chroma which did not differ between muscle types and hue-angle which did not differ between ageing days (Table 7.5).

Table 7.5 Individual and interactive effects of muscle type and days post-mortem on physical characteristics of plains zebra stallions harvested in the summer season.

Main experimental effect	p-values								
	pH_u	Weep loss %	Cooking loss %	WBSF (N)	Colour coordinates				
					L*	a*	b*	Hue	Chroma
Muscle type	**0.004**	**0.003**	**0.000**	**0.001**	**0.000**	**0.040**	**0.021**	**0.000**	0.812
Day	**0.000**	**0.000**	**0.004**	**0.000**	**0.002**	**0.000**	**0.000**	0.360	**0.000**
Muscle type*day	0.081	0.081	0.302	0.667	0.177	**0.023**	**0.013**	**0.001**	**0.032**

Abbreviations: WBSF = Warner-Bratzler shear force

The pH was the lowest (p = 0.004) in the LTL while the SM and BF plains zebra muscles did not differ from one another. Significant differences for pH between ageing days also existed low pH values between 5.3-5.5 were observed between days 1 to 20 post-mortem with minimal fluctuations. However, a large peak in pH was observed in the meat at day 24 reaching a maximum mean pH of 5.75 (Table 7.6 and Figure 7.1).

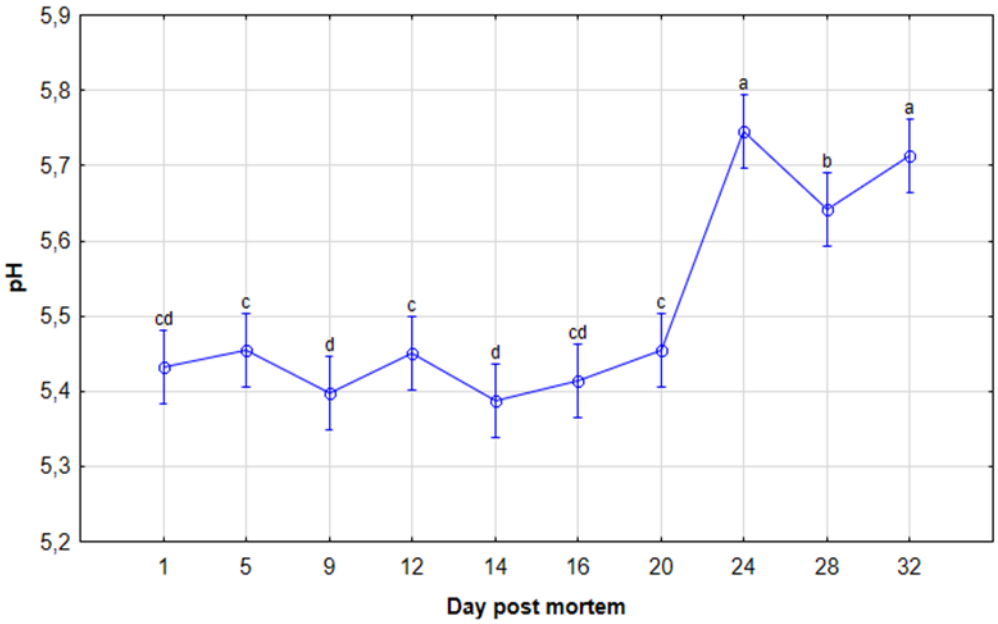

Figure 7.1 The change of pH in pooled muscles during ageing of plains zebra meat up to 32 days post-mortem. [a-d]Different letters indicate significance ($p \leq 0.05$) between mean values. Error bars indicate the standard error.

The weep loss percentage was observed to be the lowest ($p = 0.003$) in the LTL compared to both the SM and BF muscles. The ageing day effect ($p < 0.001$) existed as the weep loss were found to have a linear increasing trend with the ageing period. The lowest mean weep loss percentage was recorded on day 5 (3.0 ± 0.31 %) and the highest on day 32 (4.5 ± 0.31 %) post-mortem (Table 7.6).

The cooking loss percentage was influenced by both main effects and was significantly lower in the LTL compared to both the SM and BF muscles (Table 7.6). A relatively constant cooking loss percentage was observed between days 5 and 20, followed by a significant decrease in cooking loss at day 24 which remained constant until the last ageing day (37.3-37.7 %).

According to the WBSF observed in this study, the BF was significantly tougher than both the LTL and SM muscle, as indicated by higher shear force values. All three muscle types in the plains zebra had a linear decreasing trend from day 1 up to day 20 post-mortem (Table 7.6 and Figure 7.2). However, a slight increase in WBSF values was reported for day 24 which was followed by lower values during day 28 to 32. The WBSF values reported between day 28 and 32 was comparable to the WBSF values measured on day 20 post-mortem. All three muscles types can be classified as tender from day 14 to 32 post-mortem.

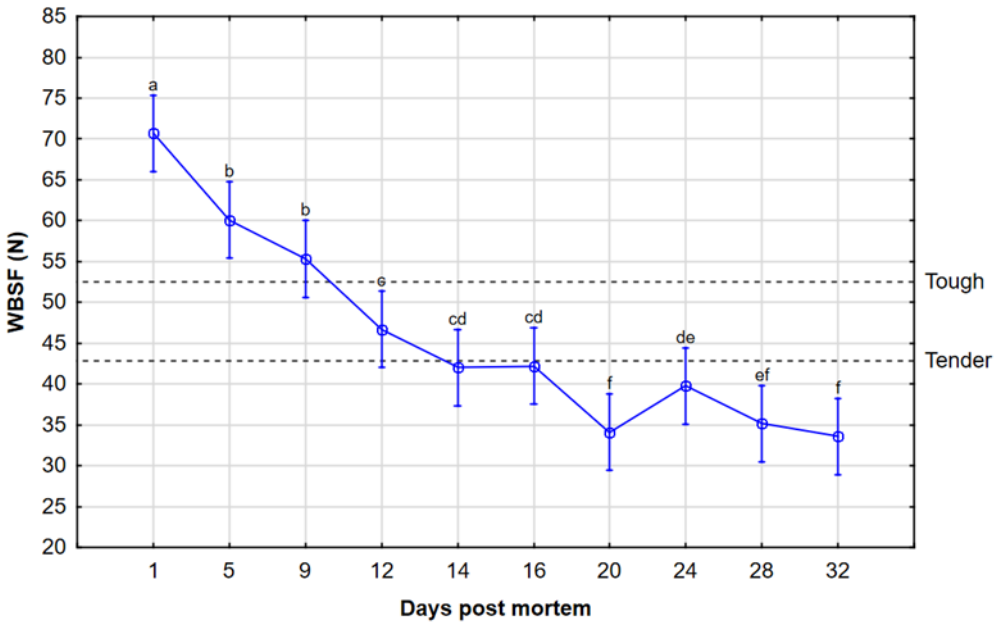

Figure 7.2 The change in the Warner-Bratzler shear force (N) of pooled muscles during ageing up to 32 days post-mortem. [a-f]Different letters indicate significance ($p \leq 0.05$) between mean values. Error bars indicate the standard error of the mean of each muscle group.

The colour measurements as influenced by muscle type and ageing day are presented in Table 7.7. No interaction between the main effects for CIE L* was observed. The LTL muscle of the plains zebra was lighter ($p < 0.001$) than both the SM and BF muscles. Even though the CIE L* differed ($p = 0.002$) between ageing days minimal fluctuations of < 1 unit were observed between days and no clear trend could be identified. The muscles were darker on day 1 (34.4 ± 0.63), day 16 (34.6 ± 0.63) and day 32 (34.6 ± 0.63).

An interaction between muscle type and ageing day existed for CIE a*, CIE b*, chroma and hue-angle (Table 7.5). The muscle type-day interaction ($p = 0.023$) for CIE a* is presented in Figure 7.3. The SM muscle was found to have significantly higher CIE a* values than the LTL muscle at days 5, 9 12 and 14 post-mortem. The LTL muscle was however found to have higher CIE a* values than both SM and BF muscles at days 20, 24 and 28 post-mortem. No significant differences existed between muscle types at days 1, 16 and 32 post-mortem. Despite the lack of significant differences between muscle types for the latter three ageing time points, a general increasing trend in CIE a* values for all three muscle types could be observed until day 14 which was followed by a plateau for the remainder of the ageing days. This trend is also apparent in the mean values for ageing days post-mortem presented in Table 7.7.

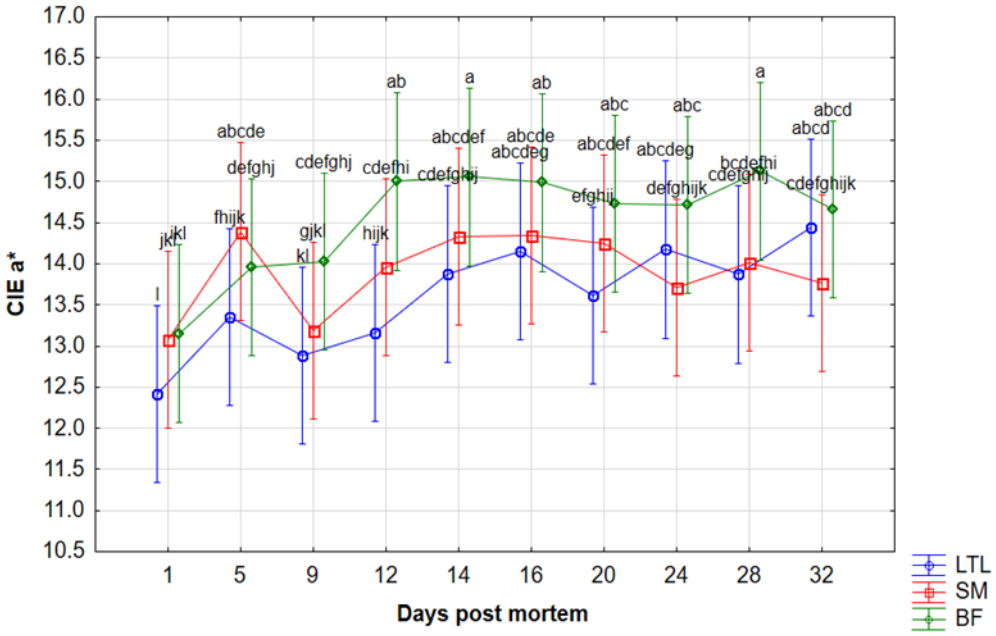

Figure 7.3 LSMeans (± standard error) of the a* values of plains zebra *Longissimus thoracis et lumborum* (LTL), *semimembranosus* (SM) and *biceps femoris* (BF) muscles during ageing up to 32 days post-mortem. [a-k]Different letters indicate significance ($p \leq 0.05$) between mean values. Error bars indicate the standard error.

The muscle type-day interaction ($p = 0.013$) for CIE b* is presented in Figure 7.4. The LTL muscle was significantly more yellow than the BF muscle on days 1, 9 and 32 post-mortem. The LTL was also more yellow than both the SM and BF muscle on day 14 with the BF being less yellow than both muscles on day 5 of post-mortem ageing. The trend for the LTL to be more yellow ($p = 0.021$) than the BF is also apparent for the mean values between muscle types (Table 7.7). No significant differences between muscle types were observed on days 12, 16, 20, 24 and 28 post-mortem. When evaluating the ageing period alone as main effect, the CIE b* values plateaued on day 16 to day 32 after a noticeable increase on day 14 (Table 7.7).

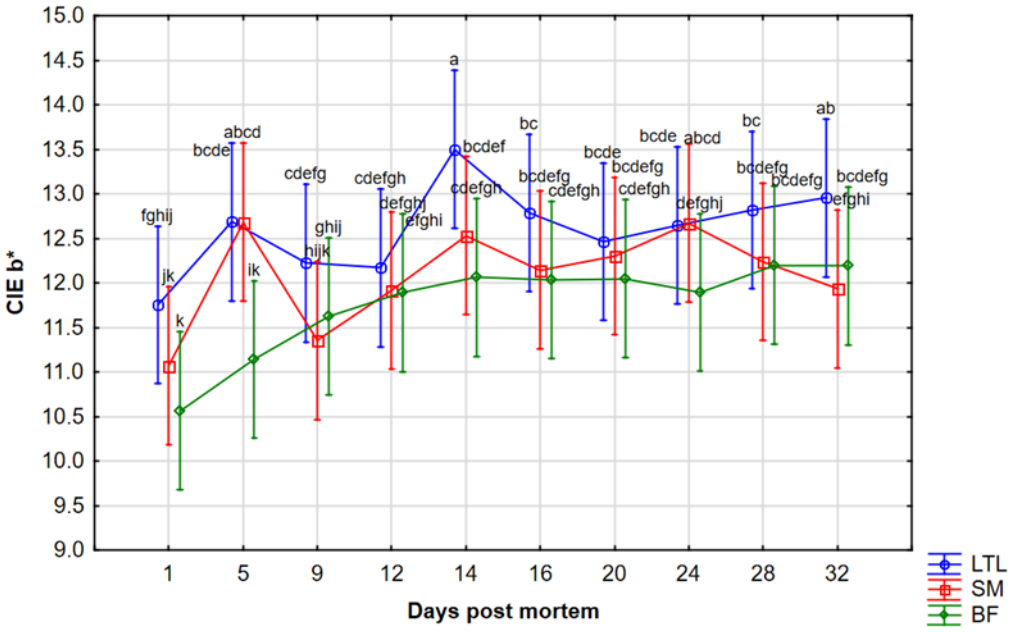

Figure 7.4 LSMeans (± standard error) of the b* values of plains zebra Longissimus thoracis et lumborum (LTL), semimembranosus (SM) and biceps femoris (BF) muscles during ageing up to 32 days post-mortem. [a-k]Different letters indicate significance ($p \leq 0.05$) between mean values. Error bars indicate the standard error.

Even though an interaction ($p = 0.001$) between the main effects was observed for hue-angle (Figure 7.5) no significant differences ($p = 0.360$) were observed between the ageing day means (Table 7.7). However, all three muscle means differed ($p < 0.001$) from one another with the highest value observed for the LTL (43.2 ± 0.90) and the lowest for SM (39.0 ± 0.90). Overall, the hue-angle stayed relatively constant for each individual muscle throughout the ageing period.

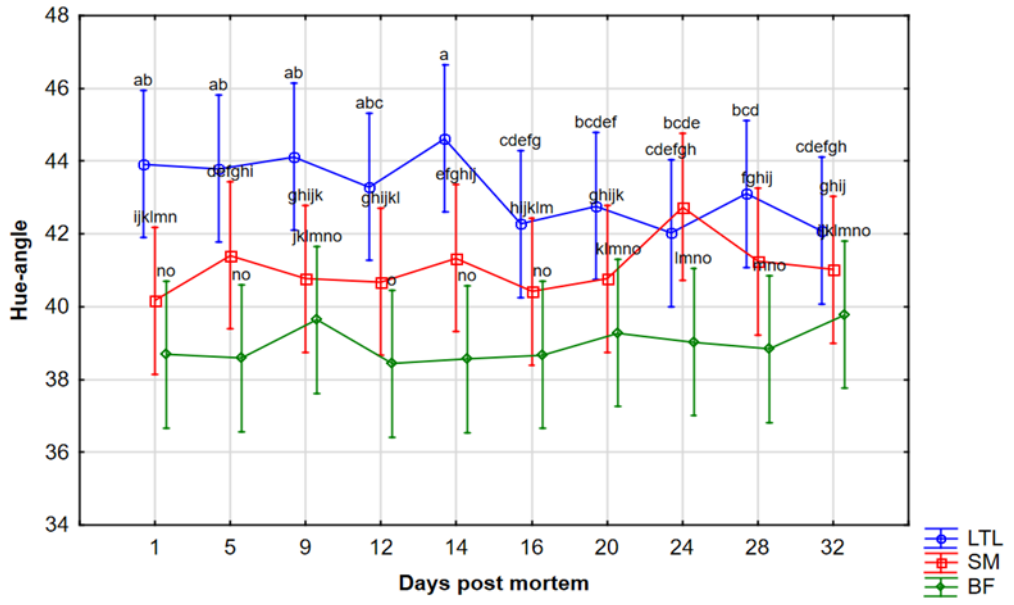

Figure 7.5 LSMeans (± standard error) of the hue-angle values of plains zebra *Longissimus thoracis et lumborum* (LTL), *semimembranosus* (SM) and *biceps femoris* (BF) muscles during ageing up to 32 days post-mortem. [a-o]Different letters indicate significance ($p \leq 0.05$) between mean values. Error bars indicate the standard error.

The interaction (p = 0.032) between the main effects for the chroma values in plains zebra meat is presented in Figure 7.6. The SM muscle had the highest chrome value on day 5 and the LTL on day 32 in comparison to their muscle counterparts. No significant differences between muscles were observed for days 9, 5, 12, 14, 16, 20, 24, and 28 of ageing. The interaction may be explained by the peak at day 5 and the drop at day 9 for the SM muscle and the peaks at days 5 and 14 and the drop at days 9 and 12 for the LTL muscle. Nevertheless, a plateau was reached for all three from day 14 onwards. The plateau reached from day 14 to day 32 is also evident in the mean ageing day chroma values presented in Table 7.7. Furthermore, no significant differences between the means for muscle type effect were observed (Table 7.5) indicating that the saturation index of all three muscle types was similar.

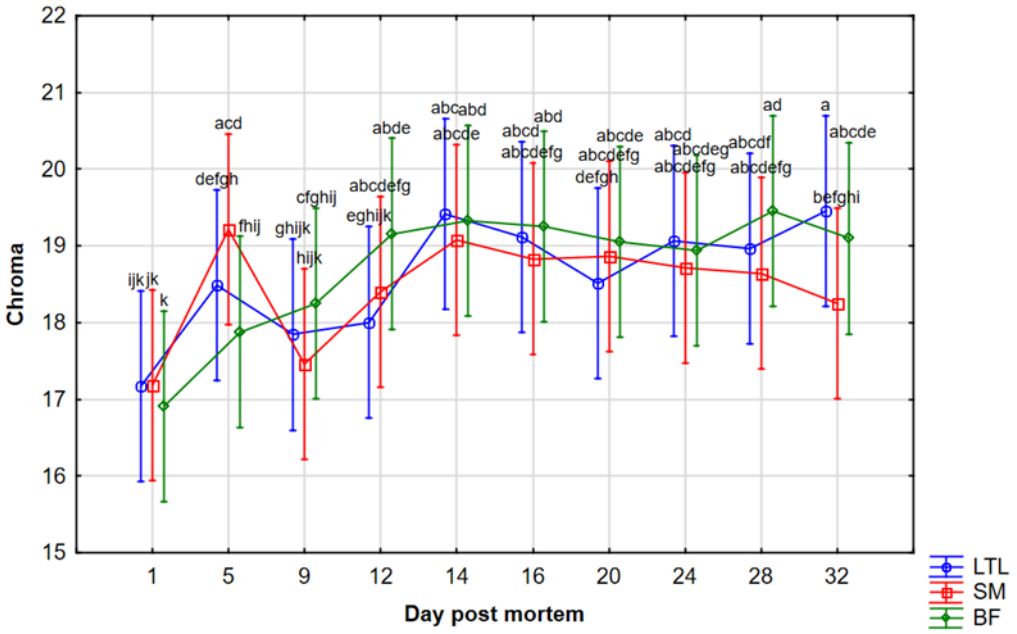

Figure 7.6 LSMeans (± standard error) of the chroma values of plains zebra *Longissimus thoracis et lumborum* (LTL), *semimembranosus* (SM) and *biceps femoris* (BF) muscles during ageing up to 32 days post-mortem. [a-k]Different letters indicate significance (p ≤ 0.05) between mean values. Error bars indicate the standard error.

Table 7.6 LSMeans (± standard error) of the physical characteristics of plains zebra stallions harvested in the summer season as per muscle type and ageing period.

Main effects		pH	Weep loss %	Cooking loss %	WBSF (N)
Muscle Type	LTL	$5.49^{b} \pm 0.018$	$3.4^{b} \pm 0.29$	$36.5^{b} \pm 0.27$	$42.8^{b} \pm 1.88$
	SM	$5.52^{a} \pm 0.018$	$4.0^{a} \pm 0.26$	$38.8^{a} \pm 0.27$	$45.0^{b} \pm 1.88$
	BF	$5.51^{a} \pm 0.018$	$3.8^{a} \pm 0.29$	$39.2^{a} \pm 0.27$	$50.1^{a} \pm 1.88$
Ageing period (days post-mortem)	1	$5.43^{cd} \pm 0.025$	*	$38.7^{a} \pm 0.33$	$70.7^{a} \pm 2.36$
	5	$5.45^{c} \pm 0.025$	$3.0^{f} \pm 0.31$	$38.4^{ab} \pm 0.33$	$60.1^{b} \pm 2.36$
	9	$5.40^{d} \pm 0.025$	$3.3^{ef} \pm 0.31$	$38.5^{a} \pm 0.33$	$55.3^{b} \pm 2.36$
	12	$5.45^{c} \pm 0.025$	$3.3^{ef} \pm 0.31$	$38.3^{abc} \pm 0.33$	$46.7^{c} \pm 2.36$
	14	$5.39^{d} \pm 0.025$	$3.6^{de} \pm 0.31$	$38.2^{abc} \pm 0.33$	$42.0^{cd} \pm 2.36$
	16	$5.41^{cd} \pm 0.025$	$3.7^{cde} \pm 0.31$	$38.7^{a} \pm 0.33$	$42.2^{cd} \pm 2.36$
	20	$5.45^{c} \pm 0.025$	$4.0^{bc} \pm 0.31$	$38.4^{ab} \pm 0.33$	$34.1^{f} \pm 2.36$
	24	$5.75^{a} \pm 0.025$	$4.0^{cd} \pm 0.31$	$37.3^{d} \pm 0.33$	$39.8^{de} \pm 2.36$
	28	$5.64^{b} \pm 0.025$	$4.4^{ab} \pm 0.31$	$37.6^{cd} \pm 0.33$	$35.2^{ef} \pm 2.36$
	32	$5.71^{a} \pm 0.025$	$4.5^{a} \pm 0.31$	$37.7^{bcd} \pm 0.33$	$33.6^{f} \pm 2.36$

[a-d]Means with different letters in the same column (within a main effect) differ from each other ($p \leq 0.05$).

*No weep loss was measured on day one as physical analysis were conducted prior to packaging.

Abbreviations: WBSF = Warner-Bratzler shear force, LTL = *Longissimus thoracis et lumborum*, SM = *semimembranosus* and BF = *biceps femoris*

Table 7.7 LSMeans (± standard error) of the colour coordinates of plains zebra stallions harvested in the summer as per muscle type and ageing period.

Main effects		L*	a*	b*	Hue	Chroma
Muscle Type	LTL	$36.9^{a} \pm 0.63$	$13.6^{b} \pm 0.48$	$12.6^{a} \pm 0.39$	$43.2^{a} \pm 0.90$	18.6 ± 0.55
	SM	$34.4^{b} \pm 0.63$	$13.9^{ab} \pm 0.48$	$12.1^{ab} \pm 0.39$	$41.1^{b} \pm 0.90$	18.5 ± 0.55
	BF	$34.0^{b} \pm 0.63$	$14.5^{a} \pm 0.48$	$11.8^{b} \pm 0.39$	$39.0^{c} \pm 0.90$	18.7 ± 0.55
Ageing period (days post-mortem)	1	$34.4^{d} \pm 0.63$	$12.9^{e} \pm 0.47$	$11.1^{e} \pm 0.38$	40.9 ± 0.90	$17.1^{d} \pm 0.53$
	5	$35.4^{a} \pm 0.63$	$13.9^{c} \pm 0.47$	$12.2^{bc} \pm 0.38$	41.3 ± 0.90	$18.5^{b} \pm 0.53$
	9	$35.3^{ab} \pm 0.63$	$13.4^{d} \pm 0.47$	$11.7^{d} \pm 0.38$	41.5 ± 0.90	$17.9^{c} \pm 0.53$
	12	$35.2^{abc} \pm 0.63$	$14.0^{bc} \pm 0.47$	$12.0^{cd} \pm 0.38$	40.8 ± 0.90	$18.5^{b} \pm 0.53$
	14	$35.1^{abc} \pm 0.63$	$14.4^{ab} \pm 0.47$	$12.7^{a} \pm 0.38$	41.5 ± 0.90	$19.3^{a} \pm 0.53$
	16	$34.6^{cd} \pm 0.63$	$14.5^{a} \pm 0.47$	$12.3^{abc} \pm 0.38$	40.5 ± 0.90	$19.1^{a} \pm 0.53$
	20	$35.4^{a} \pm 0.63$	$14.2^{abc} \pm 0.47$	$12.3^{bc} \pm 0.38$	40.9 ± 0.90	$18.8^{ab} \pm 0.53$
	24	$35.6^{a} \pm 0.63$	$14.2^{abc} \pm 0.47$	$12.4^{ab} \pm 0.38$	41.3 ± 0.90	$18.9^{ab} \pm 0.53$
	28	$35.2^{abc} \pm 0.63$	$14.3^{abc} \pm 0.47$	$12.4^{ab} \pm 0.38$	41.1 ± 0.90	$19.0^{ab} \pm 0.53$
	32	$34.6^{bcd} \pm 0.63$	$14.3^{abc} \pm 0.47$	$12.4^{abc} \pm 0.38$	41.0 ± 0.90	$18.9^{ab} \pm 0.53$

[a-d]Means with different letters in the same column (within a main effect) differ from each other ($p \leq 0.05$).

Abbreviations: WBSF = Warner-Bratzler shear force, LTL = *Longissimus thoracis et lumborum*, SM = *semimembranosus* and BF = *biceps femoris*

7.4 DISCUSSION

The main aim of this study was to determine when the point of maximum tenderness is reached in the different vacuum-packed plains zebra stallion muscles during post-mortem ageing at 4°C. The ageing trial was first conducted on the winter group over a period of 24 days and was repeated on the summer group over a period of 32 days. The trial was repeated as there was a lack of decrease in tenderness by the end of the ageing period in the first (winter) group indicating that a longer period might be advantageous. The lack of decrease in shear force values can potentially be ascribed to the undefined curvilinear relationship that seems to exist between meat tenderness and pH_u. The mean pH_u over the entire ageing period was 5.77, falling into the pH range of 5.5 to 6.1 with shear force values tending to increase with increase of pH (Purchas & Aungsupakorn, 1993). It has been speculated that the curvilinear relationship is the result of reduction in proteolytic enzyme activity between a pH_u of 5.8 and 6.3, as the activity of calpains that are involved in meat tenderisation, only starts to peak between a pH_u of 6 and 7 (Purchas & Aungsupakorn, 1993). The higher pH values observed for the winter-harvested samples compared to that of the summer-harvested samples can be due to the higher pre-harvest/slaughter activity as a result of the differences in farm layout which was discussed in Chapter 4.

Comparable weep, cooking and colour coordinates throughout both the ageing periods of the plains zebra muscles in the winter (Table 7.3 and 7.4) and summer-harvest group (Table 7.6 and 7.7) was reported. The colour coordinates measured on meat from the winter harvest group fell into the normal to intermediate range associated with game meat with minor fluctuations throughout the ageing period. The minor fluctuations observed indicated that the colour stability of plains zebra LTL, SM and BF was high throughout the 24-day post-mortem ageing period as no discolouration was observed. However, as a consequence of the shorter winter ageing period, only the extended period based on the summer group will be discussed further.

The results obtained for the summer harvested samples indicated that the mean pH (Table 7.6) values for the LTL, SM and BF remained in the acceptable range during the ageing process, however, significant differences between muscle types existed ($p = 0.004$). The LTL was characterised by the lowest pH with no differences between the SM and BF muscles, lower LTL pH may be indicative of higher post-mortem glycogen content, when compared to the SM and BF. However, the degradation of the LTL was similar to the SM and BF as no interaction between muscle type and ageing days were reported in this study ($p = 0.081$). The differences between pH of the muscle types can be attributed to differences in physical activity and thus muscle fibre type and stored glycogen levels (Seong et al., 2016). The pH values in all three muscles generally ranged between 5.43 and 5.45 measured from 24 hours to 20 days post-mortem, following a continuous peak on day 24, 28 and 32 (Table 7.6 and Figure 7.1). The low overall pH may be explained by the speculation made in Chapter 6 that plains zebras, similar to horses, have high levels of stored glycogen in their muscles ante-mortem. Adding to the latter is that plains zebras are morphologically built for large home ranges (Smuts, 1975) and the animals in this study grazed in relatively small 10 ha camps indicating a low daily activity and thus increased glycogen levels upon culling. The plains zebras were also used to human contact, as they formed part of the Quagga project, indicating that their familiarity with the hunting vehicle in combination with shorter

fleeing distances did not alter the glycogen levels to a large extent in the muscles upon culling. It will be of value if future research opportunities are created to determine how the glycogen content is correlated to stress/degree of physical activity and to the muscle fibres of various plains zebra muscles with regards to meat quality.

A significant peak in pH from 5.45 at day 20 to a mean intermediate pH of 5.75 on day 24, 5.64 at day 28 and 5.71 at day 32 was observed in this study. An gradual increase for pH during the ageing period is generally accepted (Lawrie & Ledward, 2006) and was found to increase from a pH 5.48 to 5.63 over a period of 20 days (with 10 day intervals) post-mortem ageing in vacuum packed striploins (*Longissimus lumborum,* 1st to 5th lumbar vertebrae) of Jeju crossbreed horses (Seong et al., 2016). An overall increase from pH 5.38 measured 24 hours post-mortem, to pH 5.56 measured 16 days post-mortem in vacuum packed beef LTL was reported and attributed to changes in the charge of meat proteins due to the activity of proteolytic enzymes (Boakye & Mittal, 1993). The changes in the meat protein charges, could provide an explanation for the sudden increase in the pH observed at day 24 in this study, however, the biochemical mechanism of this speculation is unknown (Engels, 2019).

Cumulative purge loss, also known as weep loss, in fresh meat is an unattractive side-effect of post-mortem ageing which is negatively perceived by consumers as a build-up of a bloody liquid in meat packaging (Huff-Lonergan & Lonergan, 2005). The ability of meat to retain water is influenced by the rate and extent of post-mortem pH decline and essentially its influence on the net charge of proteins involved with the water-holding capacity of meat. It is also dependent on the rate and extent of proteolysis, protein oxidation and the degradation of the cytoskeleton which leads to muscle cell shrinkage (Huff-Lonergan & Lonergan, 2005). The iso-electric point, reached at a pH of 5.4-5.5, is the minimum point of water-binding by the muscle proteins as the net charge is zero (Huff-Lonergan & Lonergan, 2005; Lawrie & Ledward, 2006). In the present study, the pH was between 5.4 and 5.5 from 1 to 20 days post-mortem, therefore in the range considered as the iso-electric point of meat protein, resulting in meat pH having an effect on the water-holding capacity. However, in the case of weep loss, contrasting results were observed as the percentage weep increased with an increase in pH over the post-mortem ageing period never reaching a plateau. The observed increase could be due to the variation in proteolysis of key cytoskeletal proteins in post-mortem muscle such as desmin, an intermediate filament protein, which may be related to cumulative weep production (Huff-Lonergan and Lonergan 2005).

A weep loss of one to two percent in vacuum-age meat is considered acceptable by consumers, where a weep loss of more than four percent is considered excessive, negatively impacting consumer perception on meat quality (Colle et al., 2015). In the current study, a mean weep loss up to 4.5 ± 0.31 % was observed at day 32 of post-mortem ageing with the LTL (3.4 ± 0.29 %) muscle having a significantly lower mean weep loss than both the SM (4.0 ± 0.26 %) and BF (3.8 ± 0.29 %) muscles (Table 7.6). Despite pH, muscle type differences can also be attributed to muscle function and structure (Honikel, 1987). It has been reported by Honikel (1987) that weep loss increase linearly with a decrease in sarcomere length in muscle cells. Unfortunately, the sarcomere lengths of the muscle cells in the selected plains zebra muscles were not measured in this study meritting further research. The weep loss percentage in the plains zebra muscles were higher than the acceptable 1-2% industry range, thus

bordering the classification for excessive weep loss. The higher weep loss in the plains zebra steaks for all three muscles was expected as the whole muscles were each divided into 10 steaks for each of the ageing time points, thus increasing the surface area to volume ratio during storage. The weep loss percentage of the plains zebra LTL was higher than found for the 1-2% maximum found for vacuum-aged beef steaks (Hodges, Cahill, & Ockerman, 1974; Lagerstedt, Enfält, Johansson, & Lundström, 2008), lower than 6.0 % for springbok (North et al., 2015), 6.5 % for impala (Engels, 2019) and 6.9 % for buffalo (Van As, 2019), and comparable to 3.5 % for blue wildebeest (Van Heerden, 2018) and eland (Needham et al., 2020). It needs to be noted that on day 1 of ageing no weep loss percentage was recorded as the physical analysis was conducted prior to vacuum packaging which included the measurement of drip loss percentage.

In contrast to the trend observed between pH and weep loss percentage over the 32-day ageing period, the cooking loss percentage as expected decreased with an increase in pH values. It seemed that a higher cooking loss was found on the ageing days when plains zebra meat was linked to lower weep loss, as more moisture was available to be released during the cooking processes. However, this was not the case regarding muscle type as the LTL, similar to its weep loss, had the lowest ($p < 0.001$) cooking loss percentage than both the SM and BF muscles (Table 7.6). The cooking loss of the muscles was significantly higher at the isoelectric point recorded between day 1 and 20 of the ageing period. The latter was followed by an anticipated significant decrease in cooking loss percentage in concurrence with the pH peak from day 24 onwards. The trend observed for the cooking loss in this study and its relationship with post-mortem pH of meat is well explained in literature (Huff-Lonergan & Lonergan, 2005; Lawrie & Ledward, 2006).

No interactions between muscle type and day were observed for meat tenderness, therefore the main effects will be discussed separately. The tenderness of meat is one of the most important physical parameters valued by consumers when considering purchasing or re-purchasing of red meat products. Tenderness is influenced by the alterations of the structural components within a muscle such as intramuscular connective tissue (total and insoluble collagen), structural proteins (myofibrillar and sarcoplasmic) and sarcomere length (King, Wheeler, Shackelford, & Koohmaraie, 2009; Purslow, 2005; Troy & Kerry, 2010). Plains zebra meat has been found to have a much higher “background toughness” (Chapter 5) than poultry and pork, thus post-mortem ageing is essential to improve tenderness of plains zebra meat by means of post-mortem proteolysis of structural proteins within muscle fibres. In this study, the BF muscle was significantly tougher than its counterparts over the 32-day ageing period (Table 7.6). The higher toughness in the plains zebra BF muscle corresponded with aged buffalo meat (Van As, 2019), aged blue wildebeest meat (Van Heerden, 2018), and aged springbok meat (North et al., 2015), where the BF muscle was reported to be tougher than the LTL muscle. The higher WBSF values observed in the BF can be attributed to the muscle’s “background toughness” which is dependent on the collagen composition and concentration, and cross-linkage of the collagen matrix (Colle et al., 2015; King et al., 2009; Sentandreu, Coulis, & Ouali, 2002). Insoluble and soluble collagen content in muscles are not affected or improved by ageing (Colle et al., 2015; Sentandreu et al., 2002; Silva, Patarata, & Martins, 1999) and therefore the higher values observed for the BF muscle can possibly be attributed to a higher insoluble collagen content compared to the LTL and SM muscle (North et al.,

2015; Rhee, Wheeler, Shackelford, & Koohmaraie, 2004). Unfortunately, the collagen content of the muscles was not quantified meriting further research. Furthermore, all three investigated muscles showed a similar gradual decreasing trend ($p < 0.001$) in mean WBSF values over the progression of the 32 days post-mortem ageing period with highest mean WBSF at day 1, and the lowest mean WBSF at day 20 and 32 (Table 7.6 and Figure 7.2).

The colour of red meat in vacuum-packaging in retail stores is one of the first factors influencing consumers' decision at the point of sale as it is a visual indicator of meat "freshness" (Hughes, Kearney, & Warner, 2014; Mancini & Hunt, 2005; Troy & Kerry, 2010). Discolouration of red meat is inherently subjected to the myoglobin concentration and its chemical state, lipid oxidative status, oxygen absorption rate (Neethling, Suman, Sigge, & Hoffman, 2016), and purge losses (Colle et al., 2015). Equine meat is characterised as having a high myoglobin concentration and are therefore more susceptible to oxidation, reducing the colour stability and shelf-life of fresh equine meat (Badiani and Manfredini as cited in Lorenzo et al., 2014). Therefore, to avoid revenue losses and to improve the meat quality, the characterisation of colour stability in plains zebra meat is essential as it is both a game and equine meat producing species.

According to the colour specifications for game meat established by Shange, Gouws, & Hoffman (2019), all three plains zebra muscles over the ageing period in the present study demonstrated colour coordinates after blooming typical to game meat (Table 7.7). The darker colour associated with game meat that is still acceptable by consumers has been characterised by a CIE L* value > 33 (Shange et al., 2019) and < 40 (Volpelli, Valusso, Morgante, Pittia, & Piasentier, 2003). The L* values (35.1 ± 0.63; pooled mean) of all three plains zebra muscles over the 32-day post-mortem ageing period demonstrated acceptable L* values. No interaction between the main effects were observed with L* values fluctuating between 34.4 (day 1) and 35.6 (day 24) with no clear trend. The meat was lighter on day 5, 16 and 32 post-mortem ageing, by approximately one unit. The small difference, however, may not be of significance in terms of consumer perception upon purchasing and consumption. The LTL muscle was found to be significantly lighter than the SM and BF plains zebra muscles.

An interaction was observed between muscle type and ageing days for a*, b*, hue-angle and chroma colour parameters. Despite the interactions, the general increase in a*, b* and chroma over the ageing period, indicated that all three muscles became redder, yellower, and more saturated. The redness and chroma values both reached a plateau from day 14 and yellowness from day 16. No significant differences were recorded between muscle types for chroma and between ageing days for hue-angle (Table 7.7). The hue-angle values for all three muscle types differed from one another with the LTL being the dullest, then the SM and lastly the BF. As seen in Figure 7.5, the hue-angle values stayed relatively constant over the ageing period for each muscle type indicating that the muscles did not become noticeably duller as the ageing period progressed. Therefore, as post-mortem ageing progressed all three muscle types reached acceptable colour values according to Shange et al. (2019) and Volpelli et al. (2003) with no noticeable discolouration, indicating that plains zebra meat has a high colour stability during vacuum-packed ageing over a period of 32 days.

7.5 CONCLUSION

The aim of this study was to determine the ageing period needed to ensure optimum tenderness of the *Longissimus thoracis et lumborum*, *semimembranosus*, and *biceps femoris* muscles in plains zebra stallions. The plains zebra vacuum-aged LTL, SM and BF steaks obtained from the summer-harvested animals could be classified as tender from day 14 onwards, with maximum tenderness being obtained on day 20 post-mortem at 4°C. The lack of decrease in shear force values observed in the samples obtained from the winter-harvested group demonstrated the effect of physical effort prior to culling (these zebra experienced more ante-mortem stress) on meat tenderness and needs to be taken into account when considering the effect of post-mortem ageing on meat tenderness. The inconsistent results obtained for meat tenderness between the two groups can potentially be attributed to several factors that differed between the two groups, i.e. the harvesting location, dietary regime, and farming practice. Therefore, it is recommended that further research is conducted in determining the effect of season, location, diet, and stress under controlled conditions. Nonetheless, the ageing period conducted on the summer-harvested muscles improved the bloomed surface colour of the steaks up until 14 days post-mortem, however, if necessary an ageing period up to 32 days can also be recommended as it did not result in visible discolouration of the steaks. However, microbial safety would need to be evaluated at this ageing period.

Furthermore, sensory meat quality attributes such as aroma, flavour and texture were not measured over the ageing period in this study. The effect of the ageing period on the sensory meat quality and microbial safety will be of value when considering vacuum-aged plains zebra steaks as a marketable product since both variables are altered by post-mortem ageing.

7.6 REFERENCE

Ba, H. Van, Park, K., Dashmaa, D., & Hwang, I. (2014). Effect of muscle type and vacuum chiller ageing period on the chemical compositions, meat quality, sensory attributes and volatile compounds of Korean native cattle beef. *Animal Science Journal*, *85*(2), 164–173. https://doi.org/10.1111/asj.12100

Boakye, K., & Mittal, G. S. (1993). Changes in pH and water holding properties of *Longissimus dorsi* muscle during beef ageing. *Meat Science*, *34*(3), 335–349. https://doi.org/10.1016/0309-1740(93)90082-S

Colle, M. J., Richard, R. P., Killinger, K. M., Bohlscheid, J. C., Gray, A. R., Loucks, W. I., … Doumit, M. E. (2015). Influence of extended aging on beef quality characteristics and sensory perception of steaks from the *gluteus medius* and *Longissimus lumborum*. *Meat Science*, *110*, 32–39. https://doi.org/10.1016/j.meatsci.2015.06.013

Engels, R. A. (2019). *Ante- and post-mortem factors influencing impala (Aepyceros melampus) meat quality*. Unpublished Master's thesis in Animal Science, Stellenbosch, South Africa.

Hodges, J. H., Cahill, V. R., & Ockerman, H. W. (1974). Effect of vacuum packaging on weight loss, microbial growth and palatability of fresh beef wholesale cuts. *Journal of Food Science*, *39*, 143–146. https://doi.org/10.1111/j.1365-2621.1974.tb01009.x

Honikel, K. O. (1987). How to measure the water-holding capacity of meat? Recommendation of standardized methods. In *Evaluation and Control of Meat Quality in Pigs* (pp. 129–142). https://doi.org/10.1007/978-94-009-3301-9_11

Huff-Lonergan, E., & Lonergan, S. M. (2005). Mechanisms of water-holding capacity of meat: The role of postmortem biochemical and structural changes. *Meat Science*, *71*(1), 194–204. https://doi.org/10.1016/j.meatsci.2005.04.022

Hughes, J. M., Kearney, G., & Warner, R. D. (2014). Improving beef meat colour scores at carcass grading. *Animal Production Science*, *54*(4), 422. https://doi.org/10.1071/AN13454

King, D. A., Wheeler, T. L., Shackelford, S. D., & Koohmaraie, M. (2009). Fresh meat texture and tenderness. In J. P. Kerry & D. Ledward (Eds.), *Improving the sensory and nutritional quality of fresh meat* (pp. 61–88). Cambridge, UK: Woodhead Publishing Ltd.

Lagerstedt, Å., Enfält, L., Johansson, L., & Lundström, K. (2008). Effect of freezing on sensory quality, shear force and water loss in beef M. *Longissimus dorsi*. *Meat Science*, *80*(2), 457–461. https://doi.org/10.1016/j.meatsci.2008.01.009

Lawrie, R. A., & Ledward, D. A. (2006). *Lawrie's meat science* (7th ed). CRC Press.

Lorenzo, J. M., Sarriés, M. V., Tateo, A., Polidori, P., Franco, D., & Lanza, M. (2014). Carcass characteristics, meat quality and nutritional value of horsemeat: A review. *Meat Science*, *96*(4), 1478–1488. https://doi.org/10.1016/j.meatsci.2013.12.006

Mancini, R. A., & Hunt, M. C. (2005). Current research in meat color. *Meat Science*, *71*, 100–121. https://doi.org/10.1016/j.meatsci.2005.03.003

Miller, M. F., Ramsey, C. B., Hoover, L. C., Carr, M. A., & Crockett, K. L. (2001). Consumer thresholds for establishing the value of beef tenderness quality and consistency - the issues. *Journal of Animal Science*, *79*(12), 3062–3068.

Needham, T., Laubser, J. G., Kotrba, R., Bureš, D., & Hoffman, L. C. (2020). Influence of ageing on the physical qualities of the *Longissimus thoracis et lumborum* and *bicess femoris* muscles from male and female free-ranging common eland (*Taurotragus oryx*). *Meat Science*, *159*, 107922. https://doi.org/10.1016/j.meatsci.2019.107922

Neethling, N. E., Suman, S. P., Sigge, G. O., & Hoffman, L. C. (2016). Muscle-specific colour stability of blesbok (*Damaliscus pygargus phillipsi*) meat. *Meat Science*, *119*, 69–79. https://doi.org/10.1016/j.meatsci.2016.04.015

North, M. K., Frylinck, L., & Hoffman, L. C. (2015). The physical and biochemical changes in springbok (*Antidorcas marsupialis*) *Longissimus thoracis et lumborum* and *Biceps femoris* muscle during ageing. *Meat Science*, *110*, 145–152. https://doi.org/10.1016/j.meatsci.2015.07.009

Nowak, D. (2011). Enzymes in tenderization of meat – The system of calpains and other systems. *Polish Journal of Food and Nutrition Sciences*, *61*(4), 231–237. https://doi.org/10.2478/v10222-011-0025-5

Purchas, R. W., & Aungsupakorn, R. (1993). Further investigations into the relationship between ultimate pH and tenderness for beef samples from bulls and steers. *Meat Science*, *34*(2), 163–178. https://doi.org/10.1016/0309-1740(93)90025-D

Purslow, P. P. (2005). Intramuscular connective tissue and its role in meat quality. *Meat Science*, *70*(3), 435–447. https://doi.org/10.1016/j.meatsci.2004.06.028

Radder, L., & Le Roux, R. (2005). Factors affecting food choice in relation to venison: A South African example. *Meat Science*, *71*(3), 583–589. https://doi.org/10.1016/j.meatsci.2005.05.003

Rhee, M. S., Wheeler, T. L., Shackelford, S. D., & Koohmaraie, M. (2004). Variation in palatability and biochemical traits within and among eleven beef muscles. *Journal of Animal Science*, *82*(2), 534–550. https://doi.org/10.2527/2004.822534x

Sentandreu, M. ., Coulis, G., & Ouali, A. (2002). Role of muscle endopeptidases and their inhibitors in meat tenderness. *Trends in Food Science & Technology*, *13*(12), 400–421. https://doi.org/10.1016/S0924-2244(02)00188-7

Seong, P., Park, K. M., Cho, S., Kang, G. H., Chae, H. S., Park, B. Y., & Van Ba, H. (2016). Effect of cut type and post-mortem ageing on the technological quality, textural profile and sensory characteristics of horse meat. *Animal Production Science*, *56*(9), 1551. https://doi.org/10.1071/AN14545

Shange, N., Gouws, P., & Hoffman, L. C. (2019). Changes in pH, colour and the microbiology of black wildebeest (*Connochaetes gnou*) *Longissimus thoracis et lumborum* (LTL) muscle with normal and high (DFD) muscle pH. *Meat Science*, *147*, 13–19. https://doi.org/10.1016/j.meatsci.2018.08.021

Silva, J. A., Patarata, L., & Martins, C. (1999). Influence of ultimate pH on bovine meat tenderness during ageing. *Meat Science*, *52*, 453–459. https://doi.org/10.1016/S0309-1740(99)00029-7

Smuts, G. L. (1975). Home range sizes for burchell's zebra *equus burchelli antiquorum* from the Kruger National Park. *Koedoe*, *18*, 139–146. Retrieved from http://www.koedoe.co.za/index.php/koedoe/article/viewArticle/918

Troy, D. J., & Kerry, J. P. (2010). Consumer perception and the role of science in the meat industry. *Meat Science*, *86*, 214–226. https://doi.org/10.1016/j.meatsci.2010.05.009

Van As, J. S. (2019). *Logistics around the meat supply chain in Kruger National Park : the African savanna buffalo (Syncerus caffer caffer) as model*. Unpublished Master's thesis in Animal Science, Stellenbosch, South Africa.

Van Heerden, A. M. (2018). *Profiling the meat quality of blue wildebeest (Connochaetes taurinus)*. Unpublished Master's thesis in Animal Science, Stellenbosch, South Africa.

Van Schalkwyk, D. L., & Hoffman, L. C. (2016). *Guidelines for the harvesting & processing of wild game in Namibia 2016.* Windhoek, Namibia: Ministry of Environment & Tourism.

Volpelli, L. A., Valusso, R., Morgante, M., Pittia, P., & Piasentier, E. (2003). Meat quality in male fallow

deer (*Dama dama*): effects of age and supplementary feeding. *Meat Science*, *65*, 555–562. https://doi.org/10.1016/S0309-1740(02)00248-6

CHAPTER 8

GENERAL CONCLUSION

The purpose of this study was to determine the overall meat production potential of the plains zebra (*Equus quagga*) by generating baseline data that can be valuable to the game meat industry in terms of product labelling, consumer education and marketing. The study also aimed to provide baseline information on the meat quality of plains zebra, for export of plains zebra meat is not prohibited due to the fact that plains zebra is not susceptible to foot-and-mouth disease. Plains zebra stallions in South Africa are often hunted for their skins or cropped for meat products as a management strategy to remove low ranking stallions in bachelor groups, and thus meat harvested in such a way has the potential to contribute to food security and economic stability of South Africa. The aim of this study was thus to investigate the influence of muscle type, season, and post-mortem ageing on the meat quality of plains zebra stallions obtained in the Western Cape, South Africa. The first harvesting took place during the wet winter season, June 2017 at Prinskraal farm in Bredasdorp and the second harvesting during the dry summer season, January 2018 at Elandsberg Nature Reserve- Bartholomeus Klip.

The plains zebras obtained from the winter-harvested group yielded heavier carcasses with heavier muscle weights than those obtained from the summer-harvested group. The difference in carcass weight can potentially be attributed to seasonal differences in the diet, harvesting location, and forage behaviour which alters the plane of nutrition, and as a result the body composition. However, the dressing percentages between the carcasses obtained from both seasons and the proportional contribution of the muscles to the cold carcass weight were relatively similar, irrespective of the carcass weights recorded. The corresponding dressing percentages indicate that the meat production potential is probably not influenced by season, thus promoting year-round cropping for the local and export markets. In relation to other equines, the plains zebra is larger than meat producing donkeys and predominantly smaller than meat-producing horse breeds in terms of slaughter and carcass weights. The plains zebras from both harvest groups contributed sizeable skins that has a unique value due to its aesthetic appeal, and a substantial volume of edible offal which can be utilised as a low-cost protein source.

Season of harvest influenced pH_u, drip, cooking loss, Warner-Bratzler shear force (WBSF), CIE L*, CIE b* and hue-angle of plains zebra meat. However, the most significant seasonal effect was observed was the pH_u, and in terms of consumer perception, the WBSF values. The meat obtained from the winter-harvested group fell into the intermediate range for tenderness, although bordering on the tough classification, whilst meat from the summer-harvested group fell above the suggested value for tough meat. The differences observed between the two groups can potentially be ascribed to a difference in physical activity and related stress prior to culling, as the animals from the summer-harvested covered substantial longer running distances (due to larger camps) after shot placement than their counterparts. Despite the potential higher stress levels in the summer-harvested group, the pH_u of meat was in the biologically normal range, without resulting in visible difference in tendencies inclined to be associated with dark, firm, and dry (DFD) meat.

Knowing the physical meat quality differences between different muscle types instead of meat cuts (frequently made up out of two or more muscles) is important in the game meat trade as whole muscles are generally sold or processed into various products. As expected, the physical meat quality differed significantly between muscle type, which can potentially be attributed to the differences in anatomical location, function, and structural properties. The knowledge gained on the variability in physical meat quality of different plains zebra muscles could be used as a guideline for determining which muscles are suited for fresh meat production, and which muscles may be more suitable for further processing into products to obtain maximum value from the entire plains zebra carcass. The physical meat quality of the plains zebra as recorded in this study, is similar to that reported for other game meat species, with shear force values being slightly higher than preferred. Despite the high shear force values, it can be expected to decrease with post-mortem ageing, and this was found to be the case for the *Longissimus thoracis et lumborum* (LTL), *semimembranosus* (SM) and *biceps femoris* (BF) plains zebra muscles.

The effect of muscle $pH_{u,}$ as a consequence of pre-slaughter/harvest physical activity, was observed in post-mortem vacuum-packed tenderisation of meat obtained from the winter- and summer-harvested groups. The ageing trial was first conducted on the three primal (LTL, SM and BF) muscles obtained from the winter-harvested animals over a period of 24 days, however, the muscles did not reach values associated with tender meat, which can possibly be attributed to undefined curvilinear relationship between meat tenderness and $pH_{u.}$ The trail was thus repeated using the same muscles from the summer-harvested animals but extending the trial period to 32 days. The vacuum-aged LTL, SM and BF steaks from the summer-harvested group were classified as tender at day 14, and the muscles achieved maximum tenderness on day 20 post-mortem. The ageing period used for the muscles from the latter group, improved meat surface colour up to day 14, however, no major discolouration was observed on the meat surface up to day 32 post-mortem. Therefore, it can be suggested that in the case of a high ultimate pH_u, plains zebra can potentially be aged until day 32 as no adverse effects on meat colour stability or shear force values were observed.

Season had a minor influence on the chemical meat quality of the plains zebra, with protein content being the only parameter that differed between the two groups, with higher protein values reported for the winter-harvested group. This difference, however, potentially is of little nutritional benefit to consumers. Moisture, protein, and the intramuscular fat content differed between muscle types, with the LTL and SM muscle being characterised by the lowest moisture and highest protein content. The two forequarter muscles, i.e. *infraspinatus* (IS) and *supraspinatus* (SS), and the BF muscle had the highest intramuscular fat content. Plains zebra meat can be considered lean and healthy as the muscles obtained from both groups were high in protein, and low in intramuscular fat with the concentrations of potassium, phosphorous, sodium, magnesium, calcium, iron, zinc, copper and manganese in plains zebra meat contributing notably to the human recommended dietary requirements.

Comparison of the LTL, SM and BF obtained from the winter-harvested group in terms of sensory profile and fatty acid analysis indicated no major differences between the three muscles. The BF muscle received the highest score for residue and the lowest for both beef-like flavour and tenderness. In general, the high overall aroma in plains zebra meat can be associated with game-like

sensory attributes and a low overall aroma with beef-like attributes. Plains zebra meat can also be characterised by sweet-associated sensory attributes. The saturated and polyunsaturated fatty acid ratios between muscle types did not differ, indicating that all three muscles can be considered as healthy by consumers. The three plains zebra muscles were characterised by a desirable fatty acid profile, with both the PUFA:SFA and n6:n3 PUFA ratios meeting the recommended guidelines set by the British Department of Health (1994).

Recommendations

As the study aimed to generate baseline data on the meat production and quality potential of the plains zebra, it is evident that there are various aspects that merits further research before the successful meat production potential of the plains zebra can be realised.

The plains zebra stallions obtained from both seasonal groups was determined to be mature as they had passed their maximum point of growth rate. Despite this assumption, it is recommended that the influence of age on the various meat quality factors evaluated is eliminated by harvesting animals of known age. The establishment of an accurate growth curve and feed conversion efficiency in the plains zebra is crucial, as it determines the maximum meat production potential and also the optimum slaughter age associated with marketable carcass yields and desirable physical, chemical and sensorial meat quality which is of economic importance. Plains zebra growth curves in males and females can be established by means of a serial slaughter study from multiple localities across South Africa as growth curves might differ due to differences in the dietary regime.

It is also recommended that the meat production and quality parameters of the plains zebra is evaluated in both males and females, even though species that live in stable mixed-sex groups such as the plains zebra tend to have little sexual dimorphism in terms of live weight and activity budgets. Distribution of carcass tissue is a significant factor used to determine carcass yields and research focussed on the meat to bone ratio and cutting test (block test) of plains zebra carcasses will ensure the correct allocation of carcass portions and as a result maximise the economic value thereof.

The baseline data generated with season as an effect warrants further investigation. As the plains zebra has the potential to be cropped year-round, all four seasons should be included in future studies focussed on the physical, chemical/nutritional, and sensorial meat quality. The differences found between muscles should be investigated further by determining muscle fibre type and collagen content of the six selected plains zebra muscles and, their association to meat quality.

It is further recommended to repeat the plains zebra ageing trial due to the inconsistent results obtained for meat tenderness between the two groups. For ease of comparison between main effects it is suggested to use similar ageing time points when determining the optimum post-mortem ageing period for maximal meat tenderness. It is also suggested to perform the ageing trial with meat from similar aged animals harvested on the same farm under the same ante- and post-mortem conditions to eliminate unintentional slaughter age, farming practice and dietary regime differences. However, to ensure marketable vacuum-aged plains zebra steaks research also needs to be conducted on the microbial safety and sensory parameters of the LTL, SM and BF muscles over the ageing period.

ADDENDUM I

CORRELATION MATRIX

Table I Correlation matrix for Pearson correlation coefficient (r) for the sensory characteristics and fatty acids obtained from plains zebra meat. Values in bold are significant at a level of $p \leq 0.05$.

Variable	1	2	3	4	5	6	7	8	9	10
1	**1**	**0.000**	**0.001**	**0.008**	**0.005**	0.098	0.219	**0.005**	**0.000**	**0.002**
2	**0.885**	**1**	**<0.001**	**0.000**	**0.000**	0.441	0.554	**0.006**	**0.000**	**0.000**
3	**-0.631**	**-0.783**	**1**	**0.003**	**<0.001**	0.982	0.893	0.058	**0.002**	**<0.001**
4	**0.537**	**0.670**	**-0.598**	**1**	**<0.001**	0.215	0.225	0.092	**0.002**	0.075
5	**0.562**	**0.700**	**-0.826**	**0.724**	**1**	0.730	0.337	0.112	**0.024**	**0.012**
6	0.354	0.169	0.005	0.269	0.076	**1**	0.050	0.245	0.126	0.785
7	0.266	0.130	-0.030	0.263	0.210	0.413	**1**	**0.043**	0.436	0.714
8	**0.561**	**0.556**	-0.402	0.359	0.341	0.253	**0.426**	**1**	**0.050**	0.061
9	**0.705**	**0.689**	**-0.613**	**0.622**	**0.470**	0.329	0.171	**0.414**	**1**	**0.000**
10	**-0.603**	**-0.672**	**0.731**	-0.379	**-0.513**	-0.060	0.081	-0.397	**-0.694**	**1**
11	0.373	**0.507**	**-0.695**	**0.483**	**0.449**	-0.118	-0.187	0.234	**0.585**	**-0.589**
12	**0.636**	**0.589**	**-0.629**	**0.543**	**0.611**	0.184	**0.454**	**0.484**	**0.655**	**-0.645**
13	0.145	0.100	-0.104	0.324	0.186	0.152	**0.426**	0.058	0.308	0.218
14	**0.587**	**0.476**	-0.186	**0.416**	0.264	**0.533**	**0.423**	**0.508**	0.399	-0.137
15	0.027	0.241	-0.333	0.085	0.291	-0.300	**-0.546**	0.048	0.025	-0.191
16	-0.061	0.028	-0.189	0.217	0.260	-0.036	-0.080	0.204	-0.064	0.086
17	-0.394	-0.372	0.230	-0.366	-0.216	0.071	-0.265	-0.128	**-0.494**	0.355
18	-0.361	**-0.419**	0.374	-0.397	-0.209	0.075	-0.206	-0.359	**-0.472**	0.413
19	0.332	0.331	-0.239	0.341	0.179	-0.090	0.256	0.175	**0.503**	-0.249
20	0.088	0.088	-0.056	0.018	0.002	-0.172	-0.277	-0.272	0.264	-0.269
21	0.297	0.196	-0.139	0.047	0.004	-0.040	-0.279	0.140	0.184	-0.301
22	-0.199	-0.285	**0.424**	-0.258	**-0.441**	0.318	0.197	-0.221	-0.175	0.396
23	-0.189	-0.236	-0.032	**-0.497**	-0.093	-0.233	-0.248	-0.361	-0.239	0.110
24	0.227	0.250	-0.225	0.303	**0.459**	0.119	-0.097	0.063	0.016	-0.130
25	0.116	0.126	-0.117	0.052	0.261	0.048	-0.245	-0.315	0.153	-0.119
26	0.224	0.201	-0.175	-0.131	0.123	-0.030	0.210	0.254	0.293	-0.304
27	-0.350	-0.333	0.256	-0.007	-0.239	0.010	-0.031	-0.059	**-0.418**	0.261
28	**-0.488**	**-0.533**	**0.461**	-0.322	-0.374	-0.065	-0.154	-0.408	-0.289	0.162
29	0.096	0.034	0.177	-0.067	-0.199	0.025	-0.048	-0.089	-0.056	0.333
30	-0.199	-0.303	**0.428**	-0.049	-0.091	0.113	-0.215	-0.353	-0.347	0.369
31	-0.326	-0.202	0.175	-0.011	-0.248	-0.064	-0.118	-0.161	-0.252	0.233
32	-0.035	-0.130	0.211	0.133	0.136	0.202	-0.096	-0.228	-0.213	0.212
33	0.014	-0.093	0.158	0.130	0.130	0.143	-0.106	-0.218	-0.025	0.006
34	0.348	0.261	-0.212	0.382	**0.507**	0.393	0.266	0.138	0.131	-0.092
35	0.170	0.072	-0.003	0.282	0.357	0.367	0.049	-0.001	-0.008	0.037
36	-0.079	-0.175	0.224	0.077	0.091	0.096	-0.218	-0.265	-0.131	0.112
37	**-0.462**	**-0.545**	**0.684**	**-0.567**	**-0.669**	-0.126	-0.109	-0.290	-0.311	**0.514**
38	-0.076	0.005	-0.083	-0.204	-0.272	**-0.418**	-0.006	0.017	0.043	-0.062
39	-0.033	0.054	-0.152	-0.178	-0.197	-0.406	0.063	0.090	0.093	-0.107
40	0.206	0.301	-0.392	0.000	0.067	-0.109	0.114	0.355	0.265	-0.332
41	-0.166	-0.133	0.123	-0.189	-0.346	-0.190	-0.165	-0.157	-0.201	0.162
42	-0.117	-0.045	0.021	-0.194	-0.295	-0.214	0.070	0.148	0.087	0.040
44	-0.156	-0.241	0.277	0.038	0.051	0.094	-0.229	-0.296	-0.246	0.211
45	-0.073	0.112	-0.211	-0.195	-0.139	-0.318	-0.110	0.212	0.076	-0.236
46	0.015	0.159	-0.186	0.154	0.203	0.064	0.361	0.223	0.308	-0.108
47	-0.208	-0.210	0.251	-0.197	-0.403	-0.291	-0.210	-0.314	-0.160	0.167
48	-0.128	-0.094	0.117	-0.175	-0.297	-0.299	-0.160	-0.123	0.014	0.121
49	-0.149	-0.100	0.119	-0.220	**-0.437**	-0.330	-0.225	-0.195	0.010	0.010
50	-0.317	-0.272	0.343	-0.377	**-0.509**	-0.230	-0.368	-0.286	-0.258	0.171
51	-0.042	-0.118	0.167	0.110	0.155	0.124	-0.091	-0.139	0.000	0.039
52	-0.331	-0.335	0.190	-0.262	-0.322	-0.161	-0.139	-0.367	-0.310	0.051
53	0.186	0.070	0.022	0.282	0.337	0.382	0.110	-0.051	-0.040	0.094
54	0.114	0.210	-0.303	-0.076	-0.057	-0.226	0.076	0.250	0.190	-0.246
55	**-0.427**	-0.324	0.256	-0.408	**-0.519**	**-0.433**	-0.261	-0.156	-0.117	0.078
56	-0.274	-0.159	0.092	-0.345	-0.402	**-0.438**	-0.211	0.008	-0.028	-0.019
57	-0.269	-0.248	0.262	-0.202	-0.397	-0.295	-0.140	-0.127	0.005	0.164
58	**-0.477**	-0.328	0.210	**-0.497**	**-0.527**	**-0.466**	-0.310	-0.152	-0.190	0.001
59	**0.513**	**0.416**	-0.262	**0.482**	**0.532**	0.273	0.072	0.211	0.192	0.009

Numbers in the first row correspond to numbers in the first row; 1, overall aroma intensity;2, game-like aroma; 3, beef-like aroma; 4, liver-like aroma; 5, metallic aroma; 6, sweet associated aroma; 7, herbaceous aroma; 8, fatty aroma; 9, game-like flavour; 10, beef-like flavour; 11, liver-like flavour; 12, metallic flavour; 13, herbaceous flavour; 14, sweet associated taste; 15, initial juiciness; 16, sustained juiciness; 17, tenderness; 18, mealiness; 19, residue; 20, Warner-Bratzler shear force; 21, thaw loss; 22, cooking loss; 23, drip loss, 24, pH; 25, moisture; 26, intramuscular fat; 27, protein; 28, C6:0; 29, C10:0; 30, C12:0; 31, C13:0; 32, C14:0; 33, C15:0; 34, C16:0; 35, C18:0; 36, C20:0; 37, C24:0; 38, C16:1; 39, C17:1; 40, C18:1nc; 41, C20:1; 42, C18:2n6c; 43, C18:3n6; 44, C18:3n3; 45, C20:2n6; 46, C20:3n6; 47, C20:3n3; 48, C20:4n6; 49, C20:5n3; 50, C22:2n6; 51, C22:6n3; 52, total SFA; 53,total MUFA; 54, total PUFA; 55, PUFA:SFA; 56, n6; 57, n3; 58, n6:n3; 59, total fatty acids. The non-shaded area indicates Pearson correlation coefficient (r); area shaded in grey indicates corresponding p-values for Pearson correlation coefficient (r).

Table I *Continued.*

Variable	11	12	13	14	15	16	17	18	19	20
1	0.079	**0.001**	0.508	**0.003**	0.901	0.783	0.063	0.090	0.121	0.688
2	**0.014**	**0.003**	0.649	**0.022**	0.268	0.898	0.080	**0.046**	0.123	0.691
3	**0.000**	**0.001**	0.637	0.394	0.120	0.388	0.291	0.079	0.273	0.801
4	**0.020**	**0.007**	0.132	**0.049**	0.700	0.320	0.086	0.061	0.112	0.934
5	**0.032**	**0.002**	0.396	0.224	0.177	0.230	0.321	0.339	0.415	0.993
6	0.592	0.401	0.489	**0.009**	0.164	0.871	0.746	0.734	0.684	0.434
7	0.392	**0.030**	**0.043**	**0.045**	**0.007**	0.715	0.222	0.346	0.239	0.200
8	0.282	**0.019**	0.794	**0.013**	0.829	0.349	0.561	0.092	0.426	0.209
9	**0.003**	**0.001**	0.153	0.059	0.908	0.772	**0.017**	**0.023**	**0.014**	0.223
10	**0.003**	**0.001**	0.319	0.533	0.382	0.697	0.096	0.050	0.252	0.215
11	**1**	**0.016**	0.948	0.365	0.388	0.899	0.111	**0.014**	0.100	0.175
12	**0.497**	**1**	0.321	0.284	0.982	0.636	0.054	**0.027**	0.095	0.204
13	-0.014	0.217	**1**	0.060	0.610	0.346	0.187	0.868	**0.042**	0.984
14	-0.198	0.234	0.398	**1**	0.722	0.370	0.718	0.712	0.562	0.313
15	0.189	-0.005	-0.112	-0.079	**1**	**0.014**	0.167	0.577	0.121	0.168
16	-0.028	0.104	0.206	0.196	**0.504**	**1**	0.051	0.848	0.210	0.388
17	-0.341	-0.407	-0.285	-0.079	0.298	0.411	**1**	**0.000**	**<0.001**	0.126
18	**-0.504**	**-0.459**	-0.037	-0.081	0.123	-0.042	**0.675**	**1**	**0.000**	0.599
19	0.352	0.357	**0.427**	0.128	-0.333	-0.271	**-0.919**	**-0.714**	**1**	0.303
20	0.293	0.275	0.004	-0.220	0.297	-0.189	-0.328	-0.116	0.225	**1**
21	0.085	0.207	-0.260	-0.026	0.217	0.161	-0.142	-0.208	0.029	0.045
22	-0.247	-0.294	0.212	0.058	**-0.526**	-0.391	0.050	0.334	0.037	-0.090
23	-0.146	**-0.448**	0.018	-0.262	0.000	-0.199	0.205	**0.440**	-0.175	-0.211
24	0.019	0.111	-0.068	0.296	0.387	0.266	0.072	0.099	-0.114	0.083
25	0.038	-0.100	-0.045	-0.016	0.275	-0.040	0.204	**0.415**	-0.278	0.165
26	-0.059	0.173	0.035	0.188	-0.113	-0.102	-0.293	-0.188	0.333	-0.031
27	-0.132	-0.128	-0.156	-0.141	-0.116	0.035	0.170	0.023	-0.198	-0.168
28	-0.204	-0.068	-0.107	**-0.440**	-0.058	0.041	0.141	0.086	-0.186	0.362
29	-0.163	-0.303	0.208	0.156	0.037	-0.147	-0.006	0.238	-0.045	-0.178
30	-0.328	-0.164	0.056	-0.018	0.170	0.228	0.214	0.334	-0.278	0.320
31	0.078	**-0.479**	-0.102	-0.067	-0.364	-0.057	-0.003	-0.204	0.063	**-0.459**
32	-0.182	0.058	-0.029	0.038	0.143	0.296	0.111	0.120	-0.149	0.385
33	-0.155	0.179	0.093	0.014	0.113	0.125	-0.085	0.144	0.020	**0.535**
34	0.051	0.385	0.045	0.294	0.040	0.151	-0.111	-0.047	0.075	0.353
35	-0.049	0.217	-0.056	0.141	0.092	0.242	0.081	0.092	-0.123	0.319
36	-0.176	0.064	-0.005	-0.040	0.198	0.224	0.077	0.195	-0.126	**0.498**
37	**-0.431**	**-0.435**	-0.082	-0.288	-0.117	-0.246	0.150	0.295	-0.180	0.025
38	0.124	-0.182	-0.033	-0.131	-0.108	-0.304	-0.065	-0.154	0.083	-0.288
39	0.118	-0.096	0.001	-0.078	-0.110	-0.270	-0.121	-0.219	0.164	-0.316
40	0.174	0.101	-0.095	0.109	-0.026	0.000	-0.068	-0.279	0.111	**-0.457**
41	0.124	-0.411	-0.211	-0.258	-0.071	-0.206	0.283	0.122	-0.348	-0.290
42	0.093	-0.177	0.229	-0.060	-0.205	-0.246	-0.116	-0.061	0.205	**-0.434**
43	-0.199	-0.020	-0.078	-0.076	0.187	0.283	0.169	0.174	-0.198	0.393
44	0.178	-0.033	-0.133	-0.051	0.172	-0.020	0.035	-0.170	0.079	-0.206
45	0.083	0.198	0.157	0.026	-0.181	-0.036	-0.076	-0.130	0.150	-0.322
46	-0.158	**-0.476**	-0.074	-0.218	-0.139	-0.352	0.126	0.299	-0.205	-0.209
47	0.012	-0.363	0.176	-0.138	-0.160	-0.330	-0.103	0.117	0.117	-0.340
48	0.166	-0.328	-0.063	-0.265	-0.075	-0.369	0.042	0.060	-0.094	-0.068
49	-0.161	**-0.618**	-0.222	-0.254	-0.022	-0.405	0.183	0.306	-0.277	-0.272
50	-0.136	0.202	0.033	0.022	0.119	0.197	-0.039	0.072	0.031	**0.518**
51	-0.091	-0.218	-0.051	**-0.419**	-0.074	-0.100	0.150	0.145	-0.239	-0.078
52	-0.082	0.199	0.018	0.186	0.059	0.187	0.010	0.093	-0.073	0.361
53	0.171	-0.008	-0.086	0.021	-0.060	-0.113	-0.057	-0.244	0.090	**-0.440**
54	-0.026	-0.331	0.051	-0.338	-0.043	-0.209	0.038	0.078	0.036	-0.190
55	0.019	-0.255	0.023	-0.224	0.020	-0.170	0.025	0.001	0.064	-0.287
56	-0.023	-0.272	0.332	-0.179	-0.161	-0.252	-0.104	0.132	0.165	-0.187
57	-0.024	-0.322	-0.169	-0.404	0.053	-0.143	0.139	0.024	-0.067	-0.161
58	0.061	0.175	0.197	**0.490**	0.179	0.168	-0.082	0.096	0.069	0.069
59	-0.059	0.173	0.035	0.188	-0.113	-0.102	-0.293	-0.188	0.333	-0.031

Numbers in the first row correspond to numbers in the first row; 1, overall aroma intensity;2, game-like aroma; 3, beef-like aroma; 4, liver-like aroma; 5, metallic aroma; 6, sweet associated aroma; 7, herbaceous aroma; 8, fatty aroma; 9, game-like flavour; 10, beef-like flavour; 11, liver-like flavour; 12, metallic flavour; 13, herbaceous flavour; 14, sweet associated taste; 15, initial juiciness; 16, sustained juiciness; 17, tenderness; 18, mealiness; 19, residue; 20, Warner-Bratzler shear force; 21, thaw loss; 22, cooking loss; 23, drip loss, 24, pH; 25, moisture; 26, intramuscular fat; 27, protein; 28, C6:0; 29, C10:0; 30, C12:0; 31, C13:0; 32, C14:0; 33, C15:0; 34, C16:0; 35, C18:0; 36, C20:0; 37, C24:0; 38, C16:1; 39, C17:1; 40, C18:1nc; 41, C20:1; 42, C18:2n6c; 43, C18:3n6; 44, C18:3n3; 45, C20:2n6; 46, C20:3n6; 47, C20:3n3; 48, C20:4n6; 49, C20:5n3; 50, C22:2n6; 51, C22:6n3; 52, total SFA; 53,total MUFA; 54, total PUFA; 55, PUFA:SFA; 56, n6; 57, n3; 58, n6:n3; 59, total fatty acids. The non-shaded area indicates Pearson correlation coefficient (r); area shaded in grey indicates corresponding p-values for Pearson correlation coefficient (r).

Table I *Continued.*

Variable	21	22	23	24	25	26	27	28	29	30
1	0.169	0.363	0.387	0.297	0.600	0.304	0.102	**0.018**	0.663	0.362
2	0.369	0.187	0.278	0.250	0.568	0.358	0.121	**0.009**	0.878	0.160
3	0.527	**0.044**	0.884	0.302	0.594	0.423	0.239	**0.027**	0.418	**0.042**
4	0.832	0.234	**0.016**	0.160	0.814	0.552	0.975	0.134	0.761	0.824
5	0.985	**0.035**	0.674	**0.028**	0.228	0.576	0.272	0.079	0.363	0.679
6	0.856	0.139	0.286	0.589	0.829	0.892	0.965	0.769	0.910	0.608
7	0.197	0.369	0.254	0.660	0.259	0.336	0.888	0.483	0.827	0.325
8	0.523	0.310	0.091	0.775	0.144	0.243	0.789	0.053	0.686	0.098
9	0.400	0.425	0.271	0.941	0.485	0.175	**0.047**	0.181	0.800	0.105
10	0.163	0.061	0.619	0.554	0.588	0.158	0.228	0.461	0.121	0.083
11	0.699	0.256	0.507	0.933	0.864	0.790	0.548	0.351	0.458	0.126
12	0.343	0.173	**0.032**	0.613	0.648	0.431	0.561	0.759	0.161	0.454
13	0.231	0.332	0.936	0.758	0.838	0.874	0.477	0.628	0.342	0.801
14	0.906	0.794	0.227	0.170	0.942	0.391	0.520	**0.036**	0.477	0.937
15	0.321	**0.010**	1.000	0.068	0.204	0.607	0.597	0.792	0.867	0.439
16	0.464	0.065	0.363	0.220	0.856	0.643	0.873	0.853	0.503	0.295
17	0.518	0.821	0.348	0.743	0.351	0.175	0.438	0.522	0.977	0.328
18	0.340	0.120	**0.036**	0.655	**0.049**	0.390	0.916	0.696	0.275	0.120
19	0.896	0.866	0.423	0.603	0.199	0.120	0.365	0.394	0.837	0.199
20	0.840	0.683	0.334	0.708	0.452	0.888	0.442	0.090	0.415	0.137
21	**1**	**0.014**	0.392	0.891	0.468	0.248	0.852	0.893	0.420	0.957
22	**-0.503**	**1**	0.570	0.585	0.562	0.538	0.818	0.488	0.077	0.857
23	-0.187	0.125	**1**	0.238	0.194	0.512	0.284	0.545	0.585	0.274
24	0.030	-0.120	-0.251	**1**	**0.006**	0.770	0.140	0.098	0.556	0.161
25	-0.159	0.128	0.275	**0.544**	**1**	0.718	**0.000**	0.259	0.144	0.521
26	0.251	-0.135	0.141	0.063	0.078	**1**	**0.000**	0.143	0.662	**0.010**
27	-0.041	-0.051	-0.228	-0.311	**-0.663**	**-0.699**	**1**	**0.035**	0.272	0.150
28	-0.030	-0.152	-0.130	-0.346	-0.240	-0.308	**0.433**	**1**	**0.018**	**0.003**
29	0.177	0.377	0.117	0.126	0.308	-0.094	-0.233	**-0.479**	**1**	0.862
30	-0.012	-0.040	-0.233	0.296	0.138	**-0.512**	0.303	**0.575**	0.038	**1**
31	-0.165	0.195	0.012	-0.125	-0.168	-0.086	0.186	-0.077	0.088	-0.161
32	0.017	-0.126	-0.388	**0.456**	0.109	-0.353	0.209	**0.467**	-0.205	**0.884**
33	0.113	-0.089	-0.089	0.120	0.096	-0.270	0.111	0.289	0.176	**0.591**
34	-0.093	-0.089	-0.350	**0.546**	0.130	0.068	-0.111	-0.005	-0.350	0.363
35	-0.012	-0.188	-0.347	**0.405**	0.098	-0.188	0.117	0.318	-0.391	**0.662**
36	0.074	-0.201	-0.307	0.350	0.127	-0.322	0.206	**0.592**	-0.264	**0.893**
37	0.065	0.229	0.150	**-0.609**	-0.215	-0.098	0.207	0.317	0.283	0.102
38	-0.030	0.012	0.362	**-0.474**	-0.136	0.152	-0.067	-0.313	0.243	**-0.718**
39	-0.001	-0.011	0.328	**-0.431**	-0.181	0.271	-0.110	-0.383	0.189	**-0.804**
40	0.273	-0.136	0.241	-0.303	-0.187	**0.520**	-0.250	**-0.550**	0.071	**-0.908**
41	-0.158	0.061	0.365	**-0.453**	0.018	-0.359	0.172	-0.024	0.220	-0.244
42	-0.090	0.317	0.320	**-0.464**	-0.139	0.113	-0.074	-0.350	**0.448**	**-0.597**
43	0.023	-0.214	-0.300	0.325	0.067	-0.403	0.303	**0.618**	-0.305	**0.907**
44	-0.059	0.117	0.151	-0.082	-0.062	0.335	-0.218	**-0.456**	0.037	**-0.771**
45	-0.271	0.139	0.038	-0.347	-0.054	0.252	-0.142	-0.216	-0.191	**-0.587**
46	-0.039	0.101	**0.466**	**-0.462**	0.098	-0.236	0.114	0.009	0.300	-0.179
47	0.006	0.278	**0.412**	-0.338	0.126	-0.057	-0.096	-0.285	**0.647**	-0.308
48	-0.064	0.139	0.331	**-0.464**	0.099	-0.133	-0.053	-0.054	0.348	-0.359
49	-0.043	0.183	**0.413**	-0.333	0.123	-0.261	0.157	-0.006	**0.437**	-0.135
50	0.060	-0.207	-0.378	0.249	-0.006	-0.131	0.161	**0.558**	-0.378	**0.752**
51	-0.043	0.004	0.261	-0.201	0.042	-0.349	0.242	**0.446**	0.019	0.150
52	-0.042	-0.076	-0.363	**0.515**	0.165	-0.181	0.047	0.233	-0.222	**0.695**
53	0.171	-0.087	0.312	-0.391	-0.176	0.401	-0.192	**-0.502**	0.143	**-0.901**
54	-0.094	0.214	0.312	**-0.492**	-0.109	-0.080	0.105	0.090	0.237	-0.306
55	-0.013	0.133	0.327	**-0.438**	-0.122	0.057	0.012	-0.138	0.264	**-0.507**
56	-0.096	0.277	0.263	**-0.415**	-0.024	-0.205	0.096	0.060	**0.452**	-0.054
57	-0.077	0.132	0.291	**-0.460**	-0.156	0.035	0.092	0.097	0.019	**-0.450**
58	0.054	-0.015	-0.198	**0.713**	0.381	-0.086	-0.222	**-0.468**	0.320	0.371
59	0.251	-0.135	0.141	0.063	0.078	**1.000**	**-0.699**	-0.308	-0.094	**-0.512**

Numbers in the first row correspond to numbers in the first row; 1, overall aroma intensity;2, game-like aroma; 3, beef-like aroma; 4, liver-like aroma; 5, metallic aroma; 6, sweet associated aroma; 7, herbaceous aroma; 8, fatty aroma; 9, game-like flavour; 10, beef-like flavour; 11, liver-like flavour; 12, metallic flavour; 13, herbaceous flavour; 14, sweet associated taste; 15, initial juiciness; 16, sustained juiciness; 17, tenderness; 18, mealiness; 19, residue; 20, Warner-Bratzler shear force; 21, thaw loss; 22, cooking loss; 23, drip loss, 24, pH; 25, moisture; 26, intramuscular fat; 27, protein; 28, C6:0; 29, C10:0; 30, C12:0; 31, C13:0; 32, C14:0; 33, C15:0; 34, C16:0; 35, C18:0; 36, C20:0; 37, C24:0; 38, C16:1; 39, C17:1; 40, C18:1nc; 41, C20:1; 42, C18:2n6c; 43, C18:3n6; 44, C18:3n3; 45, C20:2n6; 46, C20:3n6; 47, C20:3n3; 48, C20:4n6; 49, C20:5n3; 50, C22:2n6; 51, C22:6n3; 52, total SFA; 53,total MUFA; 54, total PUFA; 55, PUFA:SFA; 56, n6; 57, n3; 58, n6:n3; 59, total fatty acids. The non-shaded area indicates Pearson correlation coefficient (r); area shaded in grey indicates corresponding p-values for Pearson correlation coefficient (r).

Table I *Continued.*

Variable	31	32	33	34	35	36	37	38	39	40
1	0.129	0.873	0.948	0.104	0.437	0.720	**0.027**	0.731	0.883	0.345
2	0.355	0.554	0.674	0.230	0.745	0.424	**0.007**	0.980	0.806	0.162
3	0.424	0.335	0.473	0.332	0.989	0.304	**0.000**	0.707	0.490	0.064
4	0.959	0.544	0.556	0.072	0.193	0.726	**0.005**	0.350	0.417	0.999
5	0.254	0.536	0.554	**0.013**	0.095	0.680	**0.000**	0.209	0.368	0.760
6	0.773	0.356	0.514	0.064	0.085	0.664	0.566	**0.047**	0.054	0.621
7	0.591	0.662	0.632	0.219	0.826	0.318	0.621	0.979	0.776	0.603
8	0.464	0.296	0.317	0.531	0.997	0.222	0.179	0.940	0.684	0.096
9	0.245	0.329	0.909	0.550	0.972	0.552	0.148	0.845	0.672	0.222
10	0.285	0.330	0.977	0.678	0.868	0.610	**0.012**	0.780	0.628	0.121
11	0.724	0.407	0.480	0.818	0.823	0.422	**0.040**	0.574	0.593	0.428
12	**0.021**	0.792	0.413	0.070	0.320	0.773	**0.038**	0.405	0.664	0.646
13	0.644	0.897	0.673	0.839	0.801	0.981	0.710	0.880	0.996	0.666
14	0.760	0.864	0.949	0.174	0.522	0.858	0.183	0.552	0.723	0.620
15	0.088	0.514	0.608	0.858	0.675	0.366	0.597	0.623	0.618	0.905
16	0.796	0.170	0.570	0.491	0.265	0.305	0.257	0.158	0.212	1.000
17	0.990	0.616	0.700	0.613	0.713	0.727	0.494	0.768	0.581	0.759
18	0.350	0.586	0.511	0.830	0.678	0.373	0.172	0.484	0.316	0.198
19	0.775	0.496	0.927	0.734	0.576	0.566	0.412	0.708	0.455	0.614
20	**0.028**	0.070	**0.009**	0.099	0.138	**0.015**	0.911	0.183	0.141	**0.029**
21	0.452	0.937	0.608	0.671	0.955	0.736	0.768	0.894	0.997	0.207
22	0.372	0.566	0.686	0.686	0.390	0.358	0.294	0.956	0.961	0.537
23	0.955	0.061	0.679	0.093	0.096	0.144	0.486	0.082	0.118	0.258
24	0.559	**0.025**	0.576	**0.006**	**0.050**	0.093	**0.002**	**0.019**	**0.035**	0.150
25	0.433	0.614	0.655	0.545	0.650	0.553	0.313	0.526	0.397	0.383
26	0.688	0.091	0.201	0.753	0.379	0.125	0.648	0.479	0.199	**0.009**
27	0.385	0.326	0.606	0.605	0.586	0.335	0.331	0.755	0.611	0.238
28	0.722	**0.021**	0.171	0.981	0.130	**0.002**	0.132	0.136	0.065	**0.005**
29	0.682	0.337	0.410	0.094	0.059	0.213	0.180	0.253	0.377	0.743
30	0.451	**<0.001**	**0.002**	0.081	**0.000**	**<0.001**	0.634	**<0.001**	**<0.001**	**<0.001**
31	**1**	0.307	0.098	0.144	0.187	0.119	0.787	0.178	0.250	0.431
32	-0.218	**1**	**0.017**	**<0.001**	**<0.001**	**< .001**	0.491	**<0.001**	**<0.001**	**<0.001**
33	-0.346	**0.483**	**1**	0.562	0.182	**0.013**	0.857	0.056	**0.018**	**0.009**
34	-0.307	**0.711**	0.124	**1**	**<0.001**	**0.002**	**0.027**	**<0.001**	**<0.001**	**0.019**
35	-0.279	**0.893**	0.282	**0.880**	**1**	**<0.001**	0.266	**<0.001**	**<0.001**	**0.000**
36	-0.327	**0.938**	**0.498**	**0.588**	**0.830**	**1**	0.793	**<0.001**	**<0.001**	**<0.001**
37	-0.058	-0.148	0.039	**-0.451**	-0.236	-0.056	**1**	0.350	0.462	0.994
38	0.284	**-0.889**	-0.396	**-0.788**	**-0.910**	**-0.833**	0.200	**1**	**<0.001**	**0.000**
39	0.244	**-0.916**	**-0.480**	**-0.727**	**-0.901**	**-0.866**	0.158	**0.977**	**1**	**<0.001**
40	0.169	**-0.855**	**-0.520**	**-0.474**	**-0.684**	**-0.872**	-0.002	**0.707**	**0.790**	**1**
41	0.393	**-0.478**	-0.186	**-0.620**	**-0.514**	**-0.496**	0.226	**0.667**	**0.516**	0.262
42	0.298	**-0.863**	-0.231	**-0.844**	**-0.915**	**-0.799**	0.300	**0.785**	**0.779**	**0.596**
43	-0.232	**0.960**	**0.479**	**0.570**	**0.843**	**0.972**	-0.029	**-0.823**	**-0.863**	**-0.864**
44	0.098	**-0.761**	**-0.482**	**-0.507**	**-0.747**	**-0.758**	-0.034	**0.662**	**0.736**	**0.734**
45	0.087	**-0.505**	**-0.608**	-0.159	-0.245	**-0.483**	0.183	0.273	0.382	**0.519**
46	0.176	**-0.483**	-0.050	**-0.701**	**-0.573**	-0.357	0.330	**0.664**	**0.530**	0.201
47	0.329	**-0.656**	-0.071	**-0.832**	**-0.790**	**-0.545**	0.325	**0.687**	**0.625**	0.336
48	0.309	**-0.650**	-0.184	**-0.787**	**-0.755**	**-0.555**	0.264	**0.818**	**0.690**	0.330
49	0.374	**-0.511**	-0.160	**-0.805**	**-0.650**	-0.399	**0.423**	**0.634**	**0.519**	0.188
50	-0.381	**0.865**	**0.476**	**0.645**	**0.837**	**0.932**	0.019	**-0.843**	**-0.826**	**-0.749**
51	0.165	-0.100	0.140	**-0.483**	-0.332	-0.023	-0.063	0.285	0.165	-0.137
52	-0.257	**0.916**	0.349	**0.912**	**0.963**	**0.814**	-0.286	**-0.908**	**-0.905**	**-0.735**
53	0.237	**-0.931**	**-0.519**	**-0.630**	**-0.818**	**-0.927**	0.069	**0.872**	**0.917**	**0.961**
54	0.206	**-0.650**	-0.090	**-0.933**	**-0.837**	**-0.483**	**0.415**	**0.692**	**0.645**	0.316
55	0.170	**-0.793**	-0.223	**-0.929**	**-0.916**	**-0.649**	0.338	**0.800**	**0.789**	**0.536**
56	0.161	**-0.457**	0.181	**-0.806**	**-0.662**	-0.283	0.380	**0.505**	**0.428**	0.062
57	0.204	**-0.682**	-0.291	**-0.858**	**-0.819**	**-0.551**	0.365	**0.711**	**0.697**	**0.459**
58	-0.216	**0.509**	0.260	**0.651**	**0.566**	0.403	-0.402	**-0.549**	**-0.527**	-0.379
59	-0.086	-0.353	-0.270	0.068	-0.188	-0.322	-0.098	0.152	0.271	**0.520**

Numbers in the first row correspond to numbers in the first row; 1, overall aroma intensity;2, game-like aroma; 3, beef-like aroma; 4, liver-like aroma; 5, metallic aroma; 6, sweet associated aroma; 7, herbaceous aroma; 8, fatty aroma; 9, game-like flavour; 10, beef-like flavour; 11, liver-like flavour; 12, metallic flavour; 13, herbaceous flavour; 14, sweet associated taste; 15, initial juiciness; 16, sustained juiciness; 17, tenderness; 18, mealiness; 19, residue; 20, Warner-Bratzler shear force; 21, thaw loss; 22, cooking loss; 23, drip loss, 24, pH; 25, moisture; 26, intramuscular fat; 27, protein; 28, C6:0; 29, C10:0; 30, C12:0; 31, C13:0; 32, C14:0; 33, C15:0; 34, C16:0; 35, C18:0; 36, C20:0; 37, C24:0; 38, C16:1; 39, C17:1; 40, C18:1nc; 41, C20:1; 42, C18:2n6c; 43, C18:3n6; 44, C18:3n3; 45, C20:2n6; 46, C20:3n6; 47, C20:3n3; 48, C20:4n6; 49, C20:5n3; 50, C22:2n6; 51, C22:6n3; 52, total SFA; 53,total MUFA; 54, total PUFA; 55, PUFA:SFA; 56, n6; 57, n3; 58, n6:n3; 59, total fatty acids. The non-shaded area indicates Pearson correlation coefficient (r); area shaded in grey indicates corresponding p-values for Pearson correlation coefficient (r).

Table I *Continued.*

Variable	41	42	43	44	45	46	47	48	49	50
1	0.449	0.596	0.476	0.741	0.946	0.340	0.561	0.499	0.140	0.848
2	0.545	0.838	0.268	0.610	0.470	0.337	0.670	0.650	0.208	0.591
3	0.577	0.923	0.202	0.333	0.395	0.247	0.596	0.590	0.109	0.446
4	0.387	0.376	0.865	0.372	0.482	0.368	0.424	0.312	0.076	0.616
5	0.105	0.172	0.816	0.528	0.352	0.057	0.169	**0.037**	**0.013**	0.481
6	0.386	0.328	0.670	0.140	0.772	0.179	0.166	0.125	0.292	0.574
7	0.451	0.753	0.292	0.618	0.090	0.337	0.465	0.301	0.084	0.680
8	0.474	0.501	0.170	0.332	0.307	0.145	0.576	0.373	0.187	0.527
9	0.359	0.694	0.259	0.729	0.153	0.465	0.950	0.963	0.234	0.999
10	0.460	0.856	0.333	0.277	0.622	0.445	0.583	0.964	0.434	0.858
11	0.572	0.674	0.364	0.417	0.708	0.471	0.958	0.448	0.463	0.537
12	0.051	0.419	0.926	0.882	0.365	**0.022**	0.089	0.127	**0.002**	0.356
13	0.334	0.294	0.723	0.546	0.473	0.736	0.423	0.775	0.309	0.880
14	0.235	0.786	0.731	0.817	0.907	0.317	0.529	0.221	0.242	0.920
15	0.749	0.347	0.392	0.432	0.410	0.528	0.467	0.733	0.920	0.588
16	0.347	0.259	0.191	0.926	0.871	0.099	0.125	0.083	0.055	0.368
17	0.192	0.598	0.442	0.875	0.732	0.568	0.639	0.848	0.403	0.862
18	0.579	0.784	0.428	0.439	0.555	0.166	0.596	0.787	0.155	0.744
19	0.104	0.347	0.365	0.721	0.494	0.348	0.594	0.668	0.200	0.887
20	0.179	**0.038**	0.063	0.346	0.134	0.339	0.113	0.758	0.209	**0.011**
21	0.471	0.682	0.917	0.790	0.211	0.861	0.977	0.771	0.847	0.787
22	0.782	0.140	0.326	0.595	0.527	0.648	0.200	0.527	0.403	0.344
23	0.079	0.127	0.154	0.481	0.858	**0.022**	**0.046**	0.114	**0.045**	0.069
24	**0.026**	**0.022**	0.121	0.703	0.096	**0.023**	0.106	**0.022**	0.112	0.241
25	0.933	0.518	0.755	0.775	0.802	0.649	0.557	0.646	0.567	0.980
26	0.085	0.599	0.051	0.110	0.234	0.267	0.791	0.536	0.218	0.543
27	0.421	0.731	0.150	0.305	0.509	0.596	0.657	0.805	0.464	0.453
28	0.912	0.094	**0.001**	**0.025**	0.311	0.966	0.177	0.803	0.978	**0.005**
29	0.301	**0.028**	0.147	0.865	0.371	0.154	**0.001**	0.096	**0.033**	0.069
30	0.251	**0.002**	**<0.001**	**<0.001**	**0.003**	0.403	0.143	0.084	0.530	**<0.001**
31	0.057	0.158	0.276	0.649	0.685	0.411	0.116	0.142	0.072	0.067
32	**0.018**	**<0.001**	**<0.001**	**<0.001**	**0.012**	**0.017**	**0.000**	**0.001**	**0.011**	**<0.001**
33	0.383	0.276	**0.018**	**0.017**	**0.002**	0.818	0.742	0.390	0.455	**0.019**
34	**0.001**	**<0.001**	**0.004**	**0.011**	0.459	**0.000**	**<0.001**	**<0.001**	**<0.001**	**0.001**
35	**0.010**	**<0.001**	**<0.001**	**<0.001**	0.248	**0.003**	**<0.001**	**<0.001**	**0.001**	**<0.001**
36	**0.014**	**<0.001**	**<0.001**	**<0.001**	**0.017**	0.087	**0.006**	**0.005**	0.054	**<0.001**
37	0.288	0.154	0.891	0.876	0.392	0.115	0.121	0.213	**0.039**	0.928
38	**0.000**	**<0.001**	**<0.001**	**0.000**	0.196	**0.000**	**0.000**	**<0.001**	**0.001**	**<0.001**
39	**0.010**	**<0.001**	**<0.001**	**<0.001**	0.065	**0.008**	**0.001**	**0.000**	**0.009**	**<0.001**
40	0.216	**0.002**	**<0.001**	**<0.001**	**0.009**	0.346	0.109	0.115	0.379	**<0.001**
41	**1**	**0.025**	0.050	0.395	0.952	**<0.001**	**0.010**	**<0.001**	**<0.001**	**0.000**
42	**0.456**	**1**	**<0.001**	**0.001**	0.081	**0.016**	**<0.001**	**0.000**	**0.001**	**<0.001**
43	-0.404	**-0.813**	**1**	**<0.001**	**0.018**	0.085	**0.003**	**0.004**	0.064	**<0.001**
44	0.182	**0.620**	**-0.748**	**1**	0.059	0.581	0.139	0.083	0.248	**0.000**
45	-0.013	0.363	**-0.479**	0.391	**1**	0.820	0.538	0.986	0.988	0.181
46	**0.786**	**0.487**	-0.359	0.119	-0.049	**1**	**0.000**	**<0.001**	**<0.001**	**0.008**
47	**0.517**	**0.857**	**-0.588**	0.311	0.132	**0.690**	**1**	**<0.001**	**<0.001**	**0.001**
48	**0.850**	**0.659**	**-0.562**	0.361	0.004	**0.843**	**0.740**	**1**	**<0.001**	**0.000**
49	**0.731**	**0.612**	-0.383	0.245	-0.003	**0.815**	**0.791**	**0.808**	**1**	**0.004**
50	**-0.664**	**-0.765**	**0.899**	**-0.679**	-0.283	**-0.526**	**-0.629**	**-0.692**	**-0.570**	**1**
51	**0.505**	0.207	-0.014	-0.044	-0.345	**0.556**	0.342	**0.537**	**0.525**	-0.279
52	**-0.535**	**-0.899**	**0.812**	**-0.758**	-0.366	**-0.586**	**-0.746**	**-0.735**	**-0.650**	**0.791**
53	**0.459**	**0.711**	**-0.913**	**0.756**	**0.463**	0.403	**0.495**	**0.550**	0.382	**-0.853**
54	**0.461**	**0.833**	**-0.492**	**0.553**	0.172	**0.601**	**0.781**	**0.711**	**0.729**	**-0.514**
55	**0.443**	**0.890**	**-0.659**	**0.716**	0.272	**0.563**	**0.782**	**0.689**	**0.686**	**-0.651**
56	0.386	**0.781**	-0.331	0.184	0.014	**0.607**	**0.874**	**0.640**	**0.678**	-0.345
57	**0.435**	**0.717**	**-0.528**	**0.743**	0.267	**0.482**	**0.558**	**0.633**	**0.632**	**-0.553**
58	**-0.413**	**-0.444**	0.359	**-0.434**	-0.298	-0.380	-0.234	**-0.501**	**-0.408**	0.353
59	-0.359	0.113	-0.403	0.335	0.252	-0.236	-0.057	-0.133	-0.261	-0.131

Numbers in the first row correspond to numbers in the first row; 1, overall aroma intensity;2, game-like aroma; 3, beef-like aroma; 4, liver-like aroma; 5, metallic aroma; 6, sweet associated aroma; 7, herbaceous aroma; 8, fatty aroma; 9, game-like flavour; 10, beef-like flavour; 11, liver-like flavour; 12, metallic flavour; 13, herbaceous flavour; 14, sweet associated taste; 15, initial juiciness; 16, sustained juiciness; 17, tenderness; 18, mealiness; 19, residue; 20, Warner-Bratzler shear force; 21, thaw loss; 22, cooking loss; 23, drip loss, 24, pH; 25, moisture; 26, intramuscular fat; 27, protein; 28, C6:0; 29, C10:0; 30, C12:0; 31, C13:0; 32, C14:0; 33, C15:0; 34, C16:0; 35, C18:0; 36, C20:0; 37, C24:0; 38, C16:1; 39, C17:1; 40, C18:1nc; 41, C20:1; 42, C18:2n6c; 43, C18:3n6; 44, C18:3n3; 45, C20:2n6; 46, C20:3n6; 47, C20:3n3; 48, C20:4n6; 49, C20:5n3; 50, C22:2n6; 51, C22:6n3; 52, total SFA; 53,total MUFA; 54, total PUFA; 55, PUFA:SFA; 56, n6; 57, n3; 58, n6:n3; 59, total fatty acids. The non-shaded area indicates Pearson correlation coefficient (r); area shaded in grey indicates corresponding p-values for Pearson correlation coefficient (r).

Table I *Continued.*

Variable	51	52	53	54	55	56	57	58	59
1	0.123	0.396	0.605	**0.042**	0.206	0.214	**0.021**	**0.012**	0.304
2	0.118	0.750	0.336	0.132	0.468	0.253	0.127	**0.048**	0.358
3	0.386	0.920	0.160	0.238	0.677	0.227	0.336	0.228	0.423
4	0.227	0.192	0.730	0.053	0.107	0.356	**0.016**	**0.020**	0.552
5	0.135	0.116	0.798	**0.011**	0.057	0.061	**0.010**	**0.009**	0.576
6	0.463	0.072	0.299	**0.039**	**0.036**	0.171	**0.025**	0.208	0.892
7	0.528	0.618	0.730	0.229	0.334	0.523	0.150	0.745	0.336
8	0.085	0.817	0.249	0.478	0.973	0.564	0.488	0.333	0.243
9	0.150	0.857	0.385	0.596	0.899	0.980	0.386	0.381	0.175
10	0.817	0.669	0.257	0.723	0.932	0.455	0.997	0.967	0.158
11	0.681	0.710	0.435	0.906	0.933	0.916	0.914	0.781	0.790
12	0.318	0.363	0.970	0.123	0.241	0.210	0.134	0.424	0.431
13	0.819	0.933	0.698	0.816	0.918	0.121	0.441	0.367	0.874
14	**0.046**	0.395	0.923	0.114	0.303	0.414	0.056	**0.018**	0.391
15	0.739	0.789	0.787	0.846	0.928	0.462	0.809	0.413	0.607
16	0.651	0.393	0.608	0.337	0.439	0.246	0.515	0.444	0.643
17	0.495	0.964	0.798	0.864	0.911	0.637	0.528	0.709	0.175
18	0.511	0.673	0.262	0.724	0.995	0.549	0.912	0.664	0.390
19	0.272	0.742	0.682	0.870	0.773	0.451	0.760	0.754	0.120
20	0.722	0.090	**0.036**	0.386	0.184	0.394	0.462	0.755	0.888
21	0.847	0.849	0.437	0.671	0.953	0.664	0.727	0.807	0.248
22	0.985	0.729	0.694	0.326	0.546	0.200	0.548	0.947	0.538
23	0.219	0.082	0.138	0.138	0.118	0.214	0.167	0.354	0.512
24	0.346	**0.010**	0.059	**0.015**	**0.032**	**0.044**	**0.024**	**<0.001**	0.770
25	0.846	0.441	0.411	0.612	0.571	0.911	0.465	0.066	0.718
26	0.095	0.398	0.052	0.710	0.791	0.338	0.871	0.689	**<0.001**
27	0.254	0.827	0.369	0.625	0.954	0.654	0.669	0.297	**0.000**
28	**0.029**	0.273	**0.012**	0.676	0.521	0.781	0.652	**0.021**	0.143
29	0.928	0.297	0.505	0.265	0.213	**0.027**	0.928	0.127	0.662
30	0.483	**0.000**	**<0.001**	0.147	**0.011**	0.803	**0.028**	0.074	**0.010**
31	0.441	0.225	0.266	0.334	0.426	0.453	0.340	0.311	0.688
32	0.642	**<0.001**	**<0.001**	**0.001**	**<0.001**	**0.025**	**0.000**	**0.011**	0.091
33	0.516	0.094	**0.009**	0.675	0.295	0.397	0.168	0.220	0.201
34	**0.017**	**<0.001**	**0.001**	**<0.001**	**<0.001**	**<0.001**	**<0.001**	**0.001**	0.753
35	0.113	**<0.001**	**<0.001**	**<0.001**	**<0.001**	**0.000**	**<0.001**	**0.004**	0.379
36	0.915	**<0.001**	**<0.001**	**0.017**	**0.001**	0.180	**0.005**	0.051	0.125
37	0.771	0.176	0.748	**0.044**	0.107	0.067	0.079	0.052	0.648
38	0.177	**<0.001**	**<0.001**	**0.000**	**<0.001**	**0.012**	**<0.001**	**0.005**	0.479
39	0.441	**<0.001**	**<0.001**	**0.001**	**<0.001**	**0.037**	**0.000**	**0.008**	0.199
40	0.523	**<0.001**	**<0.001**	0.133	**0.007**	0.772	**0.024**	0.068	**0.009**
41	**0.012**	**0.007**	**0.024**	**0.023**	**0.030**	0.063	**0.034**	**0.045**	0.085
42	0.332	**<0.001**	**0.000**	**<0.001**	**<0.001**	**<0.001**	**<0.001**	**0.030**	0.599
43	0.949	**<0.001**	**<0.001**	**0.015**	**0.000**	0.114	**0.008**	0.085	0.051
44	0.839	**<0.001**	**<0.001**	**0.005**	**<0.001**	0.388	**<0.001**	**0.034**	0.110
45	0.099	0.079	**0.023**	0.421	0.199	0.949	0.208	0.157	0.234
46	**0.005**	**0.003**	0.051	**0.002**	**0.004**	**0.002**	**0.017**	0.067	0.267
47	0.102	**<0.001**	**0.014**	**<0.001**	**<0.001**	**<0.001**	**0.005**	0.271	0.791
48	**0.007**	**<0.001**	**0.005**	**<0.001**	**0.000**	**0.001**	**0.001**	**0.013**	0.536
49	**0.008**	**0.001**	0.065	**<0.001**	**0.000**	**0.000**	**0.001**	**0.048**	0.218
50	0.187	**<0.001**	**<0.001**	**0.010**	**0.001**	0.099	**0.005**	0.090	0.543
51	**1**	0.160	0.904	**0.019**	0.108	**0.027**	0.052	**0.018**	0.095
52	-0.296	**1**	**<0.001**	**<0.001**	**<0.001**	**0.001**	**<0.001**	**0.000**	0.398
53	0.026	**-0.854**	**1**	**0.018**	**0.000**	0.277	**0.003**	**0.020**	0.052
54	**0.474**	**-0.866**	**0.478**	**1**	**<0.001**	**<0.001**	**<0.001**	**0.000**	0.710
55	0.336	**-0.950**	**0.671**	**0.957**	**1**	**<0.001**	**<0.001**	**0.002**	0.791
56	**0.452**	**-0.646**	0.231	**0.867**	**0.779**	**1**	**0.002**	0.112	0.338
57	0.402	**-0.878**	**0.586**	**0.917**	**0.919**	**0.596**	**1**	**<0.001**	0.871
58	**-0.480**	**0.660**	**-0.471**	**-0.660**	**-0.602**	-0.333	**-0.798**	**1**	0.689
59	-0.349	-0.181	0.401	-0.080	0.057	-0.205	0.035	-0.086	**1**

Numbers in the first row correspond to numbers in the first row; 1, overall aroma intensity;2, game-like aroma; 3, beef-like aroma; 4, liver-like aroma; 5, metallic aroma; 6, sweet associated aroma; 7, herbaceous aroma; 8, fatty aroma; 9, game-like flavour; 10, beef-like flavour; 11, liver-like flavour; 12, metallic flavour; 13, herbaceous flavour; 14, sweet associated taste; 15, initial juiciness; 16, sustained juiciness; 17, tenderness; 18, mealiness; 19, residue; 20, Warner-Bratzler shear force; 21, thaw loss; 22, cooking loss; 23, drip loss, 24, pH; 25, moisture; 26, intramuscular fat; 27, protein; 28, C6:0; 29, C10:0; 30, C12:0; 31, C13:0; 32, C14:0; 33, C15:0; 34, C16:0; 35, C18:0; 36, C20:0; 37, C24:0; 38, C16:1; 39, C17:1; 40, C18:1nc; 41, C20:1; 42, C18:2n6c; 43, C18:3n6; 44, C18:3n3; 45, C20:2n6; 46, C20:3n6; 47, C20:3n3; 48, C20:4n6; 49, C20:5n3; 50, C22:2n6; 51, C22:6n3; 52, total SFA; 53,total MUFA; 54, total PUFA; 55, PUFA:SFA; 56, n6; 57, n3; 58, n6:n3; 59, total fatty acids. The non-shaded area indicates Pearson correlation coefficient (r); area shaded in grey indicates corresponding p-values for Pearson correlation coefficient (r).