

**Fossil shell flour supplementation in diets of Dohne Merino rams: effects on feed
acceptability and heat stress tolerance**



University of Fort Hare
Together in Excellence

By

Mwanda Lwazi

(201307068)

**A dissertation submitted in fulfillment of the requirements for the degree of Master of
Science in Agriculture (Animal Science)**



Supervisor: Dr. C.T. Mpendulo
University of Fort Hare
Together in Excellence
Department of Livestock and Pasture Science

Faculty of Science and Agriculture

University of Fort Hare

P/Bag X 1314

Alice

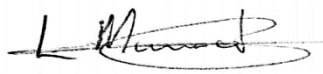
5700

February 2020

Declaration

I, Mwanda Lwazi, declare that this project is my original work and has not been submitted to any other University. The project was conducted under the supervision of Dr. C.T. Mpendulo. I also declare that, as required by these rules and conduct, I have fully cited and referenced all material and results that are not original to this work.

L. Mwanda ...



Date ...12 February 2020..

University of Fort Hare

Style and content approved by:

Dr C.T. Mpendulo ...



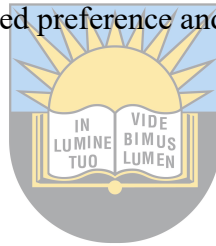
Date ...13 February 2020.....

Abstract

The objective of the current study was to evaluate the effects of feeding Dohne Merino rams with various inclusion levels of Fossil Shell Flour (FSF) as an additive supplement on feed preference and heat tolerance response of Dohne Merino rams. The FSF is used as an animal feed additive and is known to be rich in minerals. A total of twenty-four Dohne Merino rams (average 19.8 ± 0.43 kg body weight) were used to determine the feed preference of varying inclusion levels fossil shell flour (0g FSF/kg, 20g FSF/kg, 40g FSF/kg and 60g FSF/kg). The preference was assessed using feed intake and coefficient of preference. The feed intake, co-efficient of preference and physical attributes of feed (such as colour, smell and texture) was measured. The feed intake and coefficient of preference were higher in rams supplemented 60g FSF/kg ($P < 0.05$). The physical attributes of the experimental diets were adjusted based on colour, smell and texture. The colour of experimental diets were brown in 0g FSF/kg while in 20g FSF/kg, 40g FSF/kg and 60g FSF/kg the colour of experimental diets were light brown ($P < 0.05$). All the experimental diets had a pleasant smell ($P < 0.05$). The texture of diets with 0g FSF/kg was moderately firm while diets 20g FSF/kg, 40g FSF/kg and 60g FSF/kg had a firm texture. The order of preference of diets supplemented with fossil shell flour in terms of feed intake by Dohne Merino rams was: 60g FSF/kg > 40g FSF/kg > 20gFSF/kg > 0g FSF/kg. In the second experiment, a total of twenty-four ($n=24$) Dohne Merino rams of about 8 to 9 months old, weighing 19.8 ± 0.43 kg were confined in individual pens for a period of 100 days. The rams were subjected to four varying inclusion levels of fossil shell flour (0g/kg, 20g/kg, 40g/kg and 60g/kg. Water and feed intake were recorded throughout the period of successive feeding. Blood samples were collected on days 0, 45 and 90. The parameters studied included the average daily water intake (ADWI), average feed intake (ADFI), skin temperature (ST), respiration rate (RR), pulse rate (PR), rectal temperature (RT),

total plasma protein (TPP), mean corpuscular hemoglobin concentration (MCHC) and mean corpuscular volume (MCV). The inclusion levels of FSF affected both the ADWI and ADFI, showing a high-water intake and feed intake in rams subjected to 40g/kg of FSF followed by 60g FSF/kg, 20g FSF/kg and 0g FSF/kg ($P < 0.01$) respectively. All the physiological parameters ST, RT, RR, HR and PR decreased with an increase in the inclusion levels of FSF ($P < 0.01$). The TPP increased as the levels of FSF increased ($P < 0.01$). The MCHC was low in rams subjected to 60g FSF/kg and in 0g to 40g FSF/kg TPP increases as levels of FSF increases ($P < 0.01$). This study concluded that supplementing with various inclusion levels of FSF had a positive effect on feed preference. The FSF also showed the ability to mitigate heat stress in Dohne Merino rams.

Key words: Sheep, fossil shell flour, feed preference and heat tolerance



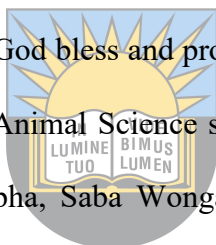
University of Fort Hare
Together in Excellence

Acknowledgements

First, I would like to thank God Almighty for keeping me up day by day and bless me to be where I am today. Success of this document was made possible due to contributions from various people of which I am obliged to acknowledge.

I would like to express my gratitude to my supervisor, Dr. C.T. Mpendulo, for his valuable guidance throughout this project. His guidance and understanding provided me courage and strength. Without his support this project would not be completed.

Deeply, I would also like to thank my research colleague: Pastor Ikusika (PhD Candidate) for his patience, encouragement, advice and guidance starting from the beginning to the successful completion of this research work; may God bless and prosper all that concern you in Jesus Name –Amen. This includes the fourth year Animal Science students that participated in this project: Xhongo Olothando, Siphosethu Mkhapha, Saba Wonga, Basana Lubabalo, Siseko Xashimba, Mbenga Yamkela, Cele Likhona, Magquphu Siphosethu, Nkasa Lona and Mdakane Bongekile.



University of Fort Hare
Together in Excellence

I would like to thank the National Health Laboratory Service at Victoria Laboratory Hospital for their relentless enthusiasm in analysing the blood samples and timely reporting of the results.

Special thanks to my colleagues and friends especially Ayanda Nyambali, Mbambalala, Lwando, Mfisi Khanyisa, Mase Amanda, Ndongeni Sibongele, Saho Samkelo and Siphango Nkosomzi. I would also like to thank the Department of Livestock and Pasture Staff members Mr Tikwayo and Mr Pepe for the technical support.

I sincerely acknowledge the financial support received from the South African Medical Research Council through Prof. Anthony I. Okoh. Grant number SAMRC/UFH/P790

Lastly, I would like to thank my parents and my sister (Fikiswa Mwanda), I am eternally grateful, words will not be enough to convey my heartfelt gratitude to you.



University of Fort Hare
Together in Excellence

Dedication

This dissertation is dedicated to my mother, Mrs. N.F. Mwanda and father, Mr. M.W. Mwanda for their prayers and encouragement.



University of Fort Hare
Together in Excellence

Table of Contents

Declaration	ii
Abstract	iii
Acknowledgements	v
Dedication	vii
CHAPTER 1: Introduction	1
1.1. Background	1
1.2. Problem statement	2
1.3. Justification	3
1.4. Objectives	3
1.5. Hypotheses	4
1.6. References	4
CHAPTER 2: Literature review	7
2.1. Introduction	7
2.2. General overview of sheep in South Africa	7
2.3 Sheep production systems	8
2.3.1. Extensive production system	8
2.3.2 Semi-intensive production system	10
2.3.3 Intensive production system	10
2.4 Description of Dohne Merino sheep	11
2.5 Nutritive value of value of FSF	12
2.6 Potential of FSF as animal performance enhancer	14
2.7 Factors affecting feed preference/ acceptability	17



University of Fort Hare
Together in Excellence

2.7.1 Quality of feed as factor affecting feed preference	17
2.7.2 Animal related factors	18
2.8. Effects of heat stress on sheep.....	19
2.9.1 Effect of heat stress on growth performance	19
2.9.2 Effect of heat stress on feed intake	20
2.10 Signs of heat stress.....	22
2.10.1 Behavioural pattern	22
2.10.2 Blood biochemistry: heat stress indicators.....	22
2.11 Heat stress prediction tools	23
2.12. Supplementation as a management practice of heat stress in sheep	24
2.13. Summary	25
2.14. Reference.....	26
Chapter 3: Feed preference of Dohne Merino rams fed varying levels of fossil shell flour.....	39
<i>Abstract</i>	39
3.1. Introduction	40
3.2. MATERIAL AND METHODS	41
3.2.1. Ethical consideration	41
3.2.3. Management of rams, feeding and experimental design	42
3.3. Measurements.....	56
3.3.1. Feed and Coefficient of preference	56
3.3.2. Proximate components	56
3.3.4. Diets evaluation	57
3.3.5 Feeding behaviour	57



University of Fort Hare
Together in Excellence

3.4 Data analyses	57
3.5. Results	59
3.5.1 Chemical composition of diets	59
3.5.2 Physical characteristics of diets	59
3.5.3 Feed intake and coefficient of preference.....	63
3.5.4 Feeding behaviour of rams	63
3.5. Discussion	63
3.6 Conclusion.....	68
3.7. References	69
Chapter 4: Effects of fossil shell flour supplementation on heat tolerance of Dohne Merino rams	
.....	75
<i>Abstract</i>	75
4.1. Introduction	76
4.2. Material and Methods.....	78
4.2.1. Ethical consideration	78
4.2.2. Study site	78
4.2.3. Experimental design, management and feedings of rams	78
4.3 Blood collection procedures.....	79
4.4. Measurements.....	81
4.4.1 Physiological parameters	81
4.4.2 Blood biochemical parameters	81
4.5 Statistical analyses.....	82
4.6. Results	84



University of Fort Hare
Together in Excellence

4.6.1. Feed and water intake	84
4.6.2. Physiological response	84
4.6.3. Blood biochemical response	88
4.7.1 Effects of FSF on feed and water intake.....	91
4.7.2 Physiological response	91
4.7.3. Blood parameters	92
4.8 Conclusion and recommendations	93
4.9 References	93
CHAPTER 5: General discussion, conclusions and recommendations.....	100
5.1 General discussion.....	100
5.2. Conclusion.....	101
5.3. Recommendations	102
5.4. References	103



University of Fort Hare
Together in Excellence

List of tables

Table 2.1: Mineral constituent of fossil shell flour (FSF).....	14
Table 3.1: Composition of experimental diets (DM %)	55
Table 3.2: Diets score (0 – 5 scale) based on physical characteristics	58
Table 3.3: Chemical composition of the experimental diets.....	60
Table 3.4: Physical characteristics of dietary treatments.....	61
Table 3.5: Experimental diets score (0 – 5 scale*) based on physical characteristic	62
Table 3.6: Preference of Dohne Merino ram for fossil shell flour.....	64
Table 3.7: Preference patterns exhibited by Dohne Merino rams fed fossil shell flour	65
Table 4.1: Mineral composition of fossil shell flour FSF.....	80
Table 4.2: Effect of fossil shell flour (FSF) on feed and water intake in Dohne merino rams.....	85
Table 4.3: Effect of fossil shell flour on skin temperature, respiration rate, rectal temperature and pulse rate.....	86
Table 4.4: Effect of FSF and day interaction on skin temperature and rectal temperature	87
Table 4.5: Effect of fossil shell flour on some blood heamatobiochemical parameters	89
Table 4.6: Effect of FSF and day interaction on TPP, MCV and MCHC	90

List of figure

Figure 2.1: Significance of various breeds as percentages of weaning weights contributed to

National Small Stock Improvement Scheme..... 9



University of Fort Hare
Together in Excellence

List of appendices

Appendix 1: Ethical clearance certificate	104
Appendix 2: List of papers submitted	106
Appendix 3: English editor's certificate	108
Appendix 4: Turnitin report	Error! Bookmark not defined.



University of Fort Hare
Together in Excellence

List of Abbreviations

ADFI	: Average daily feed intake
ADG	: Average daily gain
ADWI	: Average daily water intake
AOAC	: Association of Official Analytical Chemists
ARC	: Agricultural research council
CF	: Crude fibre
CoP	: Coefficient of preference
CP	: Crude protein
DE	: Diatomaceous earth
DM	: Dry matter
DMI	: Dry matter intake
E	: Energy
EE	: Ether extract
FR	: Feed refusal
FSF	: Fossil shell flour
g	: Grams
Hb	: Hemoglobin
HR	: Heart rate
Kg	: Kilograms
MCHC	: Mean corpuscular hemoglobin concentration
MCV	: Mean corpuscular volume
ME	: Metabolisable energy
n	: Sample size



University of Fort Hare
Together in Excellence

NBV	: Number of bite per visits
NRC	: National Research Council
NS	: Non-significant
NV	: Number of visits
°C	: Degrees Celsius
OM	: Organic matter
PCV	: Packed cell volume
pH	: Potential of hydrogen
PR	: Pulse rate
RBC	: Red blood cell
RR	: Respiration rate
RT	: Rectal temperature
SAS	: Statistical analysis systems
SEM	: Standard error of the mean
SPCA	: Society for the prevention cruelty to animals
ST	: Skin temperature
TPP	: Total plasma protein
TSF	: Time spent feeding
WBC	: White blood cell



University of Fort Hare
Together in Excellence

CHAPTER 1: Introduction

1.1. Background

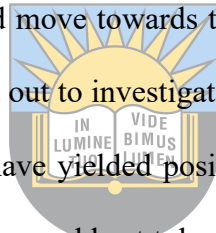
Sheep are important domestic animals compared to other domesticated livestock, because they produce multipurpose products such as meat, wool, milk and skin for household consumption (Miao *et al.*, 2005; Marino *et al.*, 2016). In modern day farming, farmers are faced with several challenges including an unstable climate, the increasing demand of agricultural products, lack of resources and increasing prices of feed stuff (Frelat *et al.*, 2016; Matlou, 2018). The combination of these factors has led to a lack of good quality feed for use in animal production and a thus a call for remedial strategies (Tripathi *et al.*, 2016). Easily accessible and affordable mineral supplements are vital in ensuring an improved and profitable animal production. Naturally occurring products such as fossil shell flour (FSF) is often a positive solution for animal feed supplementation during times where balanced ration provision is crucial but limited for sheep.

Among the factors that affect livestock production, the climate is unquestionably one of the most important. In addition, high ambient temperature and rainfall patterns are typical climatological factors that greatly impact the availability of pastures year-round. Abate (2009) reported that prolonged high temperatures and the delayed onset of rainfall result in poor regrowth of grasses and causes heat stress in livestock. However, in South Africa, livestock farmers suffer the most from climatological effect since large number of sheep are kept in an extensive farming system which depend solely on natural pasture to meet daily requirement.

The FSF, also known as Diatomaceous earth, is a naturally occurring mineral found in the sea as well as in lakes in the form of diatoms (Koster, 2013). It is a tiny particle and easily accessible compound that has a large porous surface which enables it to euthanize worms and some other internal parasites. The FSF is very rich in mineral elements silicon, when mined and milled, it

appears whitish in colour when dry and light than when wet. The FSF has high porosity and low density with a typical chemical composition of oven-dried diatomaceous earth being 80 to 90% silica, with 2 to 4% aluminium (attributed mostly to clay minerals) and 0.5 to 2% iron oxide (Ikusika *et al.*, 2019). Above all these, FSF has a relatively higher absorptive capability allowing it to be suitably used in animal feeding to improve the flow of the feed as well as improve feed mixing.

The different properties of FSF enable it to be used in improving livestock production (Ikusika *et al.*, 2019). The FSF is one of naturally available substances that can be used as a feed additive for animal feed in livestock production. The use of FSF as a feed additive rather than chemical-based feed additives can, therefore, be a good move towards the production of organic meat products. There are studies that have been carried out to investigate the effects of FSF as a feed additive on livestock performance. These studies have yielded positive results. However, there is a lack of information regarding the feed preference and heat tolerance of Dohne Merino sheep fed varying levels of FSF.



University of Fort Hare
Together in Excellence

1.2. Problem statement

High ambient temperatures impose direct and indirect stress on livestock. Some of the indirect effects of high ambient temperatures include scarcity of water, low availability of feed from grain by-products because of low grain production which also increases the cost of production (Mendelson *et al.*, 2006). Similarly, Houghton *et al.* (2001); Rust and Rust (2013) noted that, high ambient temperatures directly impact livestock production by reducing growth rate, wool production, milk production and reproduction. Therefore, there is a need to make use of cheap and available feed resources which can be advantageous to poor farmers thus increasing profitability .

Recently, there has been growing interest in using FSF as a mineral-rich feed additive in livestock (El Tazi *et al.*, 2014; Ikusika *et al.*, 2019). Furthermore, the knowledge about the use of mineral-rich feed additives to enhance feed acceptability and mitigate heat stress is common, but use of FSF is not yet explored. Therefore, there is a need to incorporate feeding and management practices as way of mitigating thermal stress to improve the productivity of the animals.

1.3. Justification

The FSF is a mineral-rich additive; it is cheap, available in abundance in many parts of the globe, and it is eco-friendly (Adebiyi *et al.*, 2009). It is vital to enhance sheep production performance such as feed acceptability and heat tolerance response. However, if feed acceptance increases due to nutrient composition of FSF in the diet of sheep, this results in the improvement of growth performance and productivity. Mineral-rich additives are advised to be included in the diet of heat-stressed animals to increase feed acceptance at the same time to mitigate heat stress. Due to global increase in climatic conditions that we experience nowadays, farmers are advised to use FSF as an alternative feed. Sordillo *et al.* (1997) noted that micronutrients that are found in FSF act as stress suppressors during high temperatures. Therefore, there is a need to determine the effects of varying levels of FSF as an additive supplement to improve Dohne Merino sheep productivity. Little information is available on the use of FSF as a feed additive in the diets of Done Merino to improve feed acceptability and heat tolerance response.

1.4. Objectives

The broad objective of the current study was to investigate the effects of varying levels of FSF as an additive supplement to improve Dohne Merino sheep productivity. The specific objectives are to:

- i. Determine feed acceptability of varying inclusion levels of fossil shell flour by Dohne Merino sheep, and to;
- ii. Determine the effects of varying inclusion levels of fossil shell flour supplementation on heat tolerance response of Dohne Merino rams

1.5. Hypotheses

The null hypotheses are:

- i. There are no effects of varying inclusion levels of fossil shell flour on feed acceptability by Dohne Merino sheep.
- ii. There are no effects of varying inclusion levels of fossil shell flour supplementation on heat tolerance response of Dohne Merino rams.



1.6. References

University of Fort Hare
Together in Excellence

- Abate, F.S.** 2009. Climate Change Impact on Livelihood. *Vulnerability and Coping Mechanisms in West-Arsi Zone, Ethiopia*. **28**(4): 681-686
- Adebiyi, O.A., Ologhobo, A.D and Ogunwale, O.A.** 2009. Growth and haematological variables of broiler chicken fed varying levels of diatomaceous earth as growth promoter. *International Journal of Current Research*, **2**: 062-066.
- El Tazi, S.M., Mukhtar, M.A., Mohamed, K.A. and Tabidi, M.H.** 2014. Effect of using black pepper as natural feed additive on performance and carcass quality of broiler chicks. *International Journal of Pharmaceutical Research and Analysis*, **4**:108-113.

- Frelat, R., Lopez-Ridaaura, S., Giller, K.E., Herrero, M., Douchamps, S., Djurfeldt, A.A., Erenstein, O., Henderson, B., Kassie, M., Paul, B.K. and Rigolot, C.** 2016. Drivers of household food availability in sub-Saharan Africa based on big data from small farms. *Proceedings of the National Academy of Sciences*, **113**(2): 458-463.
- Houghton, J.T., Ding, Y.D.J.G., Griggs, D.J., Noguer, M., van der Linden, P.J., Dai, X., Maskell, K. and Johnson, C.A.** 2001. Climate change 2001: the scientific basis. The Press Syndicate of the University of Cambridge, United Kingdom.
- Ikusika, O.O., Mpendulo, C.T., Zindove, T.J. and Okoh, A.I.** 2019. Fossil Shell Flour in Livestock Production: A Review. *Animals*, **9**(3): 70.
- Koster, H.** 2013. Diatomite in animal feed. <http://www.Agrisilica.co.za/pdf/eng/diatom>. Accessed 18 May 2019.
- Marino, R., Atzori, A.S., D'Andrea, M., Iovane, G., Trabalza-Marinucci, M. and Rinaldi, L.** 2016. Climate change: Production performance, health issues, greenhouse gas emissions and mitigation strategies in sheep and goat farming. *Small Ruminant Research*, **135**: 50-59.
- Matlou, N.Y.** 2018. Challenges faced by emerging farmers in the managing projects towards sustainable agricultural development in Capricorn District, Doctoral dissertation, University of Limpopo, Limpopo Province, South Africa.
- Mendelsohn, R., Nordhaus, W.D. and Shaw, D.** 2006. The impact of global warming on agriculture: a Ricardian analysis. *The American economic review*, **85**(6): 753-771.



University of Fort Hare
Together in Excellence

Miao, Z. H., Glatz, P. C. and Ru. Y. J. 2005. Free-range poultry production-A review. *Asian-Austrian Journal of Animal Science*, **18**(1): 113-132.

Rust, J.M. and Rust, T. 2013. Climate change and livestock production: A review with emphasis on Africa. *South African Journal of Animal Science*, **43**(3): 255-267.

Sordillo, L.M., Shafer-Weaver, K. and DeRosa, D. 1997. Immunobiology of the mammary gland. *Journal of dairy science*, **80**(8): 1851-1865.

Tripathi, A., Tripathi, D.K., Chauhan, D.K., Kumar, N. and Singh, G.S. 2016. Paradigms of climate change impacts on some major food sources of the world: a review on current knowledge and future prospects. *Agriculture, Ecosystems and Environment*, **216**: 356-373.



University of Fort Hare
Together in Excellence

CHAPTER 2: Literature review

2.1. Introduction

Resource-poor farmers in South Africa keep relatively high numbers of sheep in an extensive farming system (Mapiye *et al.*, 2009). This kind of farming system compromises the growth rate and reproductive performance during hot summer months as well as in the dry seasons. This eventually leads to severe economic losses by farmers. In the hot summer months high ambient temperatures resulted in heat stress on livestock coupled with rainfall patterns that greatly impaired the availability of good quality forage, especially during the dry seasons (Mlambo and Mapiye, 2015; Purwar *et al.*, 2017). Therefore, the provision of supplementary feed to sheep during critical periods should improve the performance of sheep to maximize productivity and gain profit (Young *et al.*, 2014). According to Adebisi *et al.* (2009) who reported that incorporating FSF in the diet of animals enhance feed acceptability and feed conversion while improving growth performance. Marai *et al.* (2007) reported that the provision of a cheaper mineral-rich feed additive can be vital in improving livestock production performance. Fossil shell flour as feed additive for livestock increases acceptability of feed and acts as a stress suppressor during heat stress (Sordillo *et al.*, 1997). This review, therefore, focused on the knowledge about FSF supplementation in the diet of Dohne Merino rams: effects on feed acceptability and heat stress tolerance.

2.2. General overview of sheep in South Africa

Sheep are important domesticated animals when compared to other domesticated livestock as they produce multipurpose products such as meat, wool, milk and skin for household consumption (Marino *et al.*, 2016). According to Johnson *et al.* (2005), raising lambs on high concentrate diets brings about great improvement in the daily gains, dressing percentage and carcass quality than

when the lambs are raised on a forage system. The differences in genetic potential among sheep breeds brings opportunity for the selection of breeds for better performance by breeding different types of sheep that possess good quality traits. For the improvement of sheep production, an assessment of all the available sheep genetic resources is vital in understanding their role under different sheep production systems. Among all the sheep breeds in South Africa Dohne Merino is dominant breed because of its adaptability to survive under harsh environmental conditions and veld conditions in the sour-grass of the Eastern Cape regions (Cloete *et al.*, 1951). Therefore, supplementing the diet of this kind of breed with FSF might improve daily feed intake, weight gain and feed efficiency. In addition, this breed is far better than other breeds in terms of contribution to National Small stock Improvement Scheme in weaning weights (Cloete *et al.*, 2014) as illustrated in Figure 2.1 below.



2.3 Sheep production systems

2.3.1. Extensive production system

University of Fort Hare
Together in Excellence

An extensive sheep production system relies on the communal land and in most cases, there is no hired labour. Sheep also graze on natural veld, crop by-product and the sheep hardly receive supplementary feed (Getu, 2015). Livestock in an extensive farming system are mostly dependent on natural pastures and is characterized by low inputs. This farming system indicates a high mortality rate, low rate of reproduction, weaning percentage and the turnover is very low (Dagnew *et al.*, 2017). These are the results of poor housing, poor health and poor nutrition (Tadesse *et al.*, 2015). To improve productivity in this kind of production system there is need to consider supplementing with cheap available feed stuff such FSF. Fossil shell flour is cost

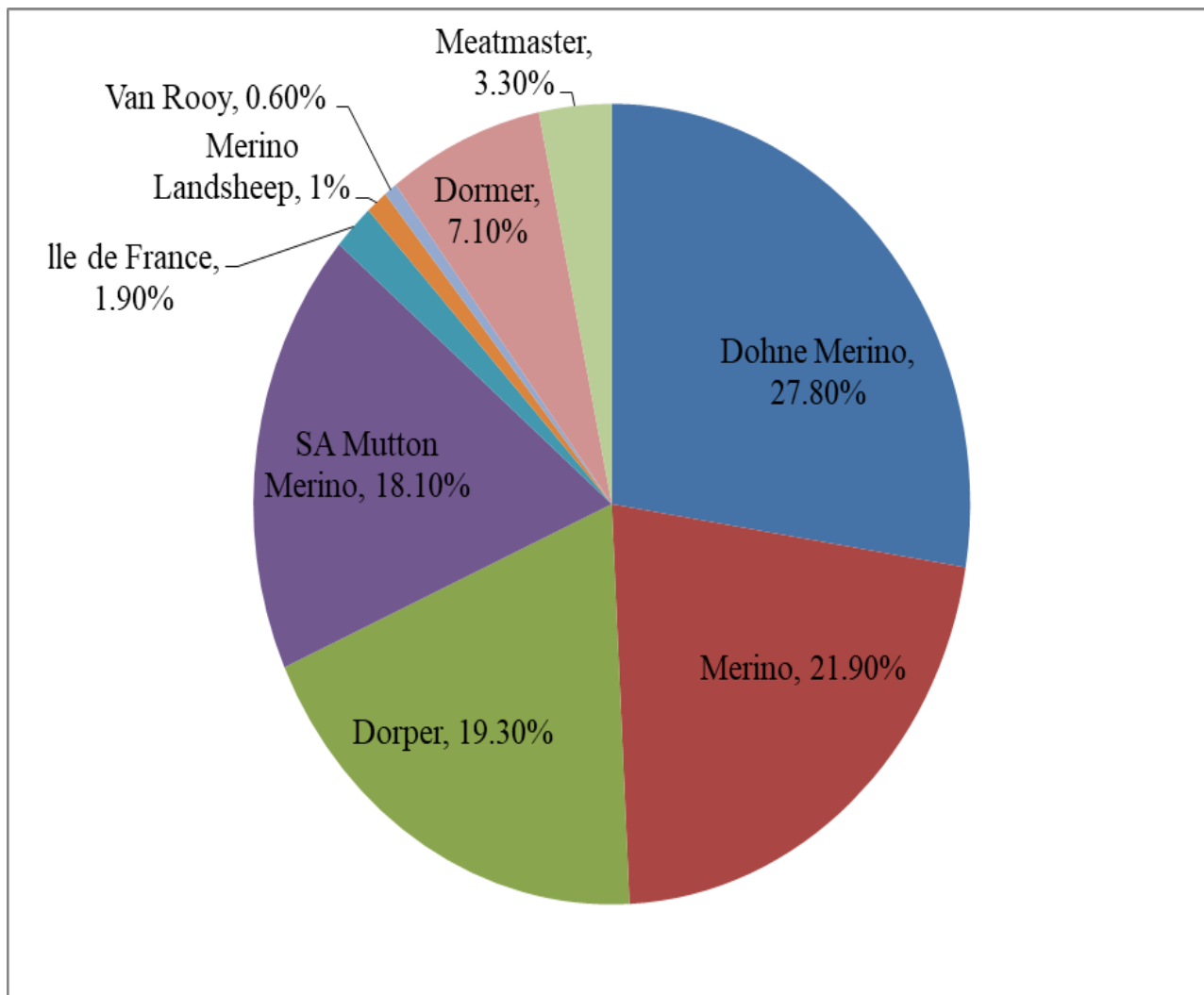


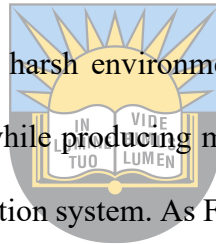
Figure 2.1: Significance of various breeds as percentages of weaning weights contributed to National Small Stock Improvement Scheme

Source: (Cloepe *et al.*, 2014)

effective, readily available, healthy and eco-friendly feed additives which can serve as animal feed supplement. It is a naturally occurring silicate rich substances that are readily available in many countries.

2.3.2 Semi-intensive production system

A semi-intensive production system includes large investment on hired labour, technology and better management practices. In this production system the selection of breeds for better performance is also involved (Castel *et al.*, 2003; De Rancourt *et al.*, 2006). Furthermore in this production system animals are allowed to roam freely and graze on natural pastures whilst they are also supplemented with a certain amount of formulated feeds during dry periods (Morris, 2017). Shelter is also provided during harsh environmental conditions, lactation and gestation period. To cut the cost of production while producing more is best way of making profit, hence FSF could fit best in this kind of production system. As FSF has a potential to increases daily feed intake and growth rate.



University of Fort Hare
Together in Excellence

2.3.3 Intensive production system

The intensive sheep production system is practised by producers who are market driven (Garnett, 2010). It involves the use of selected breeds with high genetic potential, innovations, technology, and better management practices which favour productivity. In addition, under this production system animals are completely confined, and ewes are mostly synchronised, lambing more frequently probably multiply offspring per year. In addition to that, in this type of production system all animal needs are catered for from feed provision to disease control (Garnett, 2010). Even in this production system using cheap readily feed stuff such as FSF could lower the cost of production. The improved average daily weight gain of 0.20 kg/day in confined West Africa dwarf

sheep fed 4% of FSF in the study conducted by Emeruwa (2016), makes fossil shell flour fit best also this production system.

Nevertheless, though Dohne Merino was bred under a semi-intensive system of production, it also performs brilliantly in other systems of sheep production. Dohne Merino can be reared both in extensive and intensive systems of production. In a study conducted by Nowers *et al.* (2017) looking at the production system in an extensive area where some Dohne Merino sheep were kraaled overnight . It was observed that there is no statistical effect found in the weight gain between Dohne Merino sheep kraaled (between 16:00hr and 8:00hr) and Dohne Merino that was unrestricted. Likewise, greasy fleece weight, fiber diameter and clean yield percentage for the two groups were similar. Therefore, from the above point of view, it clearly shows that Dohne Merino will successfully displace its genetical potential under any production system.

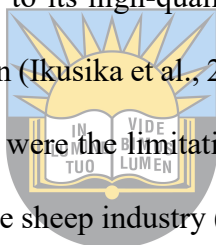
2.4 Description of Dohne Merino sheep

Dohne Merino sheep is a synthetic dual-purpose breed that is native to South Africa. It is an important breed that was bred in late 1939 at the Dohne Agricultural Research Station near Sutterheim in the Eastern Cape Province of South Africa. According to Swanepoel *et al.* (2007) this breed was established by interbreeding Merino ewes with German Merino (SA Sheep Merino) rams. The offspring were chosen for its adaptability to survive under harsh environmental conditions and veld conditions in the sour-grass of the Eastern Cape regions (Cloete and Durand, 2000). Additionally, Dohne Merino sheep was developed to meet market requirements such as high lamb survival rate, great quality fleece, high fecundity and ability to lamb within a short lambing interval (Van Wyk *et al.*, 2008). The main physical appearance of Dohne Merino sheep

includes plain white coat and amber coloured muzzle, sloping rump, long legs, straight back and both ewes and rams are polled (Cloete and Durand, 2000).

2.4.1 Important of Dohne Merino sheep to South Africa farming

The Dohne Merino is native to Africa and was bred in South Africa around 1939. Since then, the breed has gained popularity in South Africa and other African countries. Due to its ability to withstand the arid and semi-arid weather conditions in most parts of Africa, it has relieved farmers in these areas as they have resorted to rearing the Dohne Merino (Wyk et al., 2008). From South Africa, the breed has spread to other African countries due to its high fertility and fast lamb growth rate (Cloete, 2016). The Dohne Merino improved the wool industry in South Africa (around 40% of the SA wool sheep) and Africa; due to its high-quality wool and ease of management, most farmers have resorted to wool production (Ikusika et al., 2020). The breed has filled the gap of low fertility and high mortality rates, which were the limitations of other sheep breeds, including the Merino, and has resulted in a boost in the sheep industry (Swanepoel, 2006). Dohne Merino breed has help to meet the protein requirement of South African.



University of Fort Hare
Together in Excellence

2.5 Nutritive value of value of FSF

Minerals are substances that occur naturally and are required in minute (1 to 2500 mg per day) amounts by animals' bodies. Minerals are required for different functions which range from maintenance, production, growth to reproduction by livestock (Sharma *et al.*, 2009). Different animal species have different nutritional requirements such as mineral requirements. The compound FSF consists of different minerals which include copper, boron, titanium and manganese which are minerals that form a small composition, 2% iron, 3% magnesium 5% sodium and 33% silicon (Mvumi *et al.*, 2006).

These different minerals of FSF play different roles in animals. For instance, a mineral that is found in large quantities in FSF is silicon and plays a role in bone formation with the help of Calcium (Ca). Calcium is responsible for formation and preservation of bones (Soetan *et al.*, 2010). Bones are where both red blood cells and white blood cells in adult sheep are produced. For normal function of red blood cells found within the bones, iron must be present in adequate amounts in blood. Iron parttake in haemoglobin formation (Price *et al.*, 2013). Fossil shell flour consists of 5% sodium and the key role of sodium working with potassium, and chlorine is to uphold osmotic pressure (Galyean *et al.*, 1999). The mineral constituents of FSF are illustrated in Table 2.1 below.



University of Fort Hare
Together in Excellence

Table 2.1: Mineral constituent of fossil shell flour (FSF)

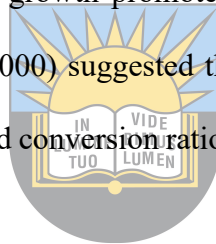
Element	Quantity
Calcium (Ca)	0.40
Sodium (Na)	0.26
Manganese (Mn)	0.0052
Iron (Fe)	0.72
Copper (Cu)	0.0019
Vanadium (V)	ppm 43.8
Sulfate Sulfur (S)	0.062
Phosphorus (as P2O5)	0.037
Potassium (K)	0.16
Chloride	0.074% or 740 ppm
Zinc (Zn)	0.0022
Titanium (Ti)	ppm 420
MgO (calculated from % Mg)	0.34
Strontium (Sr)	ppm 59.9
Boron (B)	0.0023
Magnesium (Mg)	0.21
% CaO (calculated from % Ca)	0.55
Aluminum (Al) %	0.65

Source: (Adebiyi *et al.*, 2009)

2.6 Potential of FSF as animal performance enhancer

2.6.1 FSF as animal performance enhancer

Generally, the value of livestock greatly depends on their body weight. The substances that are used to accelerate growth of animals are called growth promoters and enhancers. There is a stable increase in the market of growth promoters and enhancers. Li *et al.* (2013) reported that antibiotics are the most dominant group of growth promoters that are used in animal feed. There are other growth promoters other than antibiotics. These include vaccines, antimicrobials, essential oils, bacteriophages, innate defense, probiotics, prebiotics, organic acids, botanicals, antibodies, enzymes and phytochemicals (Abd El-Tawab *et al.*, 2016). The use of antibiotics in livestock feed has become objectionable due to chemical residues that remain in meat and meat products, and they create an antibiotic-resistant bacterial population in humans (Adebiyi *et al.*, 2009). Hence, there is still room for developing new growth promoters that are chemically free, available in abundance and cost-effective. Macy (2000) suggested the use of FSF in the diets of chicken in order to improve average daily gain, feed conversion ratio, growth performance and also to control parasites.



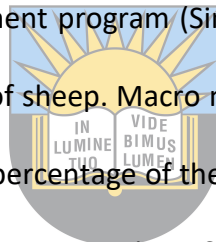
University of Fort Hare
Together in Excellence

Sarijit *et al.* (2016) reported improvement in terms of average daily gain, feed intake and efficacy were observed in animals fed 32% FSF, 11% activated carbon, 35% moisture, and 22% organic carbon. Osweiler and Carson, (1997) reported good acceptability of diet incorporated with 2.5 percent FSF in West African Dwarf sheep. Therefore, if FSF has good acceptability when it is included in the animal diet, that would lead to increased feed intake. This could mean that an increased feed intake results in better growth performance. Moreover, the study also revealed that animals did not display any harmful effects to their growth and health (Sarijit *et al.*, 2016). Similarly, in the study conducted by Adebiyi *et al.* (2009) where cockerels were fed 6% FSF for 6 weeks, they noted that there was an increase in growth performance and bone development. Modirsane *et al.* (2008) reported positive results on daily feed intake and the growth rate of piglets

fed 0.02% of FSF at the end of their weaning period. Therefore, fossil shell flour could bring about substantial economic gains for livestock producers, due to its ability to increase feed preference and to mitigate heat stress.

2.6.2. Fossil shell flour as source of minerals in Livestock Production

Minerals are essential nutrients for livestock, and they affect many aspects of an animal's health and performance. These elements play critical roles in the proper functioning of enzymes, hormones, and cells. Deficiencies can result in performance issues such as compromised rates of gain, milk production and reproduction. Trace mineral intake and absorption help livestock achieve optimal performance and their genetic potential, making trace mineral absorption integral to a successful herd management program (Sims and Garreth, 2010). There are many minerals that are required in the diet of sheep. Macro minerals are required in larger amounts, with that requirement expressed as a percentage of the diet or as gains per head per day. They include potassium, calcium, phosphorus, magnesium, Sulphur, and sodium. Micro minerals are minerals needed in very small quantities. They are also called trace minerals. The requirement by animals for these minerals are expressed in milligrams per head per day or in parts per million. They include Iron, copper, manganese, zinc and selenium (Galvayan et al., 1999). Mineral's deficiencies may be more prone in organic farming as farmers are to feed mostly home-grown pasture and feeds where soil may be deficient in minerals. There is a need to ensure that organically produced feeds are sufficient in any nutrients to keep the animals in full health and vigor at all stages of their productive life. Trace minerals that have been identified as important for normal immune function and disease resistance are zinc, manganese, selenium, and copper in many field conditions (Galvayan *et al.*, 1999). Zn and Mn are essential for the integrity of the



University of Fort Hare

Ngqoma, 2010

epithelial tissue such as gastrointestinal, urinary, reproductive and respiratory tract thereby reducing infiltration of pathogen through these epithelial lining and protect. Copper and zinc are important for both cell and humoral-mediated immunity through many molecular functions such as production of antibodies, neutrophils and lymphocytes replication, and antioxidant enzyme production (Adebiyi et al., 2009.) All these minerals are available in fossil shell flour (Ikusika et al., 2019)

2.7 Factors affecting feed preference/ acceptability

2.7.1 Quality of feed as factor affecting feed preference

Bennett *et al.* (2011) noted that FSF is very useful to add to animal feed, especially in sweet feed. Sweet feed contains sticky substances such as molasses and is stored in areas of high humidity that cause the feed to clump. Adebiyi *et al.* (2009) reported that FSF prevents clumping of feed, thus improving flowability and mixability of animal feed. This eventually becomes advantageous for animals feed on diet containing FSF. Fossil shell flour acts as an anti-caking agent, hence it prevents the clumping of feed particles therefore exposing the whole feed to both enzyme and bacteria and therefore more feed is actually digested and utilized. Moreover, FSF comprises of about fourteen trace minerals that are required by animals for their body maintenance and production. Therefore, this means that when FSF is included in the diet of sheep it is likely to influence the feed preference.

In general, the chemical and physical attributes of feed are the most important factors that affect an animal's feed preferences. The study conducted by Morand-Fehr (2003) revealed that goats prefer dry feeds that come in pellets over the same dry feeds in the form of flour. The preference of cows for dust feed can be improved by making the feeds to be in pellets (Meier *et al.*, 2012). In

a study that was conducted by Meier *et al.* (2012), it was reported that physical qualities of a feedstuff, such as structure, length and thickness, may impact the feed acceptance.

Ingredients with a low particle size in theory have higher chance of being mixed homogeneously. Sheep prefer feed with tiny particle sizes and the inclusion of feed additives in feeding diets increases feed intake if the particle size are small (Baumont *et al.*, 2000). Increase in feed preference results in increase in feed intake. Larger particle sized feeds are difficult to digest especially in young growing sheep. The chemical composition is regarded as a quality indicator of a feed and it is also found as an important factor that influences feed preferences in animals (Boufennara *et al.*, 2012). Ginane *et al.* (2003) found that quality attributes of fiber rich feed for dairy animals could be most preferred. In an examination on field with yearlings' calves Ginane *et al.* (2003) found that the calves favoured grass of good quality. According to Meier *et al.* (2012) good quality fiber of a feed enhances feed intake and feed preference. Also, it has been found that feed with high ash content is of low quality and therefore its palatability and preference is compromised (Meier *et al.*, 2012).



University of Fort Hare
Together in Excellence

2.7.2 Animal related factors

The previous exposure of animals to feedstuff can increase their feed preference and feed intake. Likewise, hunger also seems to be of significance in an animal's feed preferences. A hungry animal will in general eat the primary feed offered in the absence of its real preferred feed (Scharenberg *et al.*, 2007). Also, in a study conducted by Villalba and Provenza (1999), it was discovered that physiological status has an impact on feed preferences in lambs. It was reported that physiological status of lambs was more important for the feed preferences than the structure of the feeds. As hunger considered as the most important factor that influences feed acceptability, since FSF increases appetite in animals (Osweiler and Carson, 1997), therefore, it is vital to include FSF in

diet of animals to increase feed intake. Osweiler and Carson, (1997) further reported good acceptability of diet incorporated with 2.5 percent FSF in West African Dwarf sheep. Sheep prefer feedstuff that will meet their nutrient requirements.

Sheep require about 20 to 40 ounces each per day of NaCl, and the feed intake decreases when NaCl is not included in their diet (Digby *et al.*, 2011). Many researchers (Garmyn *et al.*, 2011; Desimone *et al.*, 2013) reported that incorporating minerals such as Mg, Na, K and Ca increase palatability which leads to increased feed preference. Nutritional requirements of an individual animal and the differences in stages of production including lamb development, gestation, lactation and dry period are aspects that may impact each animal's feed preference (Dikmen *et al.*, 2009). Even though ruminant animals share numerous things in common, many things regarding feeding behaviour differ between the different ruminant species (Meier *et al.*, 2012). Moreover, the findings of the study conducted by Atwood *et al.* (2001) have shown that even animals with same physiological status have different feed preferences.



University of Fort Hare
Together in Excellence

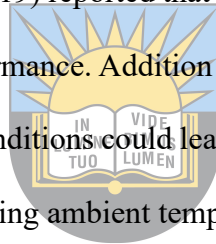
2.8. Effects of heat stress on sheep

Based on climate change, there is an increasing concern of the impact of heat waves on animal welfare as well as their production in both temperate and tropical regions (Nardone *et al.*, 2010). There are negative effects that are brought by heat stress on animal productivity and health (Bernabucci *et al.*, 2015). The heat stress effects are particularly negative in high productive animals due to the close relationship between metabolic heat generation and production levels.

2.8.1 Effect of heat stress on growth performance

Growth can be defined as cell duplication or in increase in the live weight of an animal which has been determined by the genetic composition of the animal and the environment. These factors

include feed intake, ambient temperature, hormones and enzymes (Marai *et al.*, 2007). Sheep performance is mainly affected by a thermal environment. One of the important signs for heat stress in sheep is elevated levels of respiration rate and body temperature. Increase in body temperature is directly proportional to the decrease in feed intake, redistribution in blood flow and changes in endocrine activities which adversely affect reproductive and productive performance in the sheep (Abouheif and Alsobayel, 1982, 1983; Eltawill and Narendran, 1990). Moreover, a decrease in the feed intake ultimately results in the poor growth performance of sheep. However, Calamari *et al.* (2007) stated that mineral supplements during high ambient temperature buffers the effect of high temperature and brings substantial improvement in average weight gain, feed conversion efficiency. Ikusika *et al.* (2019) reported that the inclusion of FSF in the diet of Dohne Merino wethers improve growth performance. Addition of FSF in the diet of Dohne Merino sheep which is to be adaptable under harsh conditions could lead to positive results, since FSF also plays a good role in mitigating heat stress during ambient temperature.



University of Fort Hare
Together in Excellence

2.8.2 Effect of heat stress on feed intake

Sheep is known as the species that have good capability of converting fibrous, low-quality feedstuff into meat and other products more than cattle (Hafez, 2013). Moreover, moderate heat stress results in reduction in feed intake and slow body development in newborn sheep, which consume high feed intake of medium quality roughage diets. Furthermore, relative responses to supplements that provide mainly fermentable metabolizable energy (ME), or similar amounts of fermentable ME and additional metabolizable protein are not affected by the moderate heat stress (Dixon *et al.*, 1999).

The immune system of sheep is improved by use of supplementation of brown seaweed (Tasco) to post-harvest fescue hay which in turn results in protecting the body against long heat-induced

oxidation stress (Saker *et al.*, 2004). Digestion is the process whereby the feedstuffs are broken down into smaller amounts so they can be absorbed into the bloodstream of the animal. A study conducted by Marai *et al.* (2001) has shown that there is a decrease in crude protein (CP), metabolizable energy (ME) ether extract (EE), energy (E) and digestibility of dry matter (DM), when the animals are exposed to high temperatures. Nutrient degradability and digestibility on a rumen increase with the increases of ambient temperature and low relative humidity (35°C and 50% humidity). However, decreases in ambient temperature and increase in relative humidity (30°C and 60%) lower the digestibility and nutrient degradability in adult Merino rams (Weniger and Stein, 1992). A study carried out by Shafie *et al.* (1994) showed that digestibility of DM, CP, crude fibre (CF), organic matter (OM), and nitrogen free extract (NFE) contents are lesser amongst rams kept at 35°C compared to those of the control.

The feed digestibility is affected by ambient temperature through a variation in the gastro-intestinal tract as well as the degree of passage of the digesta through the system. Digestibility is increased by the decrease in degree of passage or the increase in the retention time of the digesta. When the rams are kept at temperature of 35°C the rumen temperature and rumen volume are reported to be slightly high with lower levels of the rumen buffering capacity, protozoal count, total volatile fatty acid concentration and ammonia-NH₃ and pH being the non-effecting agent on heat stress but in return heat stress being affected (Shafie *et al.*, 1994). Even in this case to increase feed during heat stress you need to consider the usage of mineral rich feed supplements such as FSF. Rhoads *et al.* (2013) reported that supplementing diet with mineral additive that improve insulin sensitivity may be good tactic to increase likelihood of surviving lethal heat stress. Therefore, that will increase feed intake, feed conversion ratio and eventually lead to increased growth performance as mentioned in the above subheading.

2.9 Signs of heat stress

2.9.1 Behavioural pattern

The behaviour of livestock is linked to that of its health status. Livestock are said to underperform and decrease their production when they are under massive heat pressure (Ratnakaran *et al.*, 2017). Moreover, heat pressure may lead to change in the behavioral pattern of livestock. Habeeb *et al.* (2018) noted that when temperatures are between 26.7 to 32.2°C with humidity ranging from 50 to 90% moderate indications of heat stress might be observed in livestock. The most noticeable livestock behavior response towards heat stress includes an increased frequency of drinking, reduced feed intake, defecating, urinating frequency and shade seeking attitude. As temperatures increase above 37°C, animals start to display more critical signs of heat stress. They do this through open mouth breathing with panting and tongue hanging out. In severe cases, extreme environmental temperatures may result in death, especially when livestock is experiencing other stresses such as illness (Vitali *et al.*, 2009).



University of Fort Hare
Together in Excellence

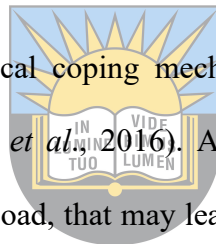
2.9.2 Blood biochemistry: heat stress indicators

Blood biochemical factors change when animals are heat stressed. Hemoglobin and packed cell volume increases during heat stress in livestock, and alterations are regarded as drastic change in adaptive mechanism features of livestock (Mazzullo *et al.*, 2014). Moreover, there are several hormones that are involved in the homeothermy regulation process in livestock. Animals decrease secretion of thyroid hormones to regulate metabolic activities and hence the production of body heat. Feed intake decreases under long-term heat stress and leads to poor feed utilization. This results in the reduction in quantities of erythrocytes and hemoglobin channels and ultimately

results in reduced red blood cells (Swenson and Reece, 2006). Al-Haidary (2004) reported that the values of PCV increase during heat stress due to the fact that it acts as adaptive mechanism for sheep during high temperatures, it provides water necessary for evaporation in the course of high temperatures. The concentration of hemoglobin (Hb) decreases during heat stress condition this might be since animals need more Hb to transport oxygen molecules to support the evaporative cooling mechanism. Gyanendra *et al.* (200) reported that high ambient temperatures reduce the number of white blood cells, red blood cell, mean corpuscular volume (MCV) and mean corpuscular hemoglobin concentration (MCHC).

2.10 Heat stress prediction tools

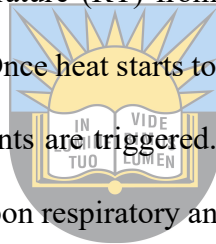
Livestock has a variety of physiological coping mechanisms that helps to lower their body temperature during heat stress (Yasha *et al.*, 2016). At the point where physiological coping mechanisms fail to lessen temperature load, that may lead to acceleration of body temperature to an extent that the metabolism of the animal effected and those eventual compromises health status of the animal. Body temperature shows consequences in the heat that is released by the body and the heat that is absorbed by the body; hence it is used as a tool to determine heat tolerance in animals. The major parameters that determine physiological adjustment mechanisms in small ruminants include changes in temperature of the rectum, respiration, and heart rate (Sejian *et al.*, 2017; Invernizzi *et al.*, 2016). The rectal temperature is a regular tool used in estimating animal temperature, for experimental justification (Dikmen *et al.*, 2011). Respiratory rate during extremely hot temperatures is the first thermoregulation system expressed by small ruminants to be able to reduce body heat. Moreover, various physiological reactions are known as sheep and goat's response to high environmental temperatures. These responses include panting, sweating



University of East Haven
Together in Excellence

rate, improved respiration rate, decreased metabolic rate and changes in endocrine role (Sejian *et al.*, 2012; Nejad and Sung, 2017). Hence in discussing complications and a suite of physiological changes is important because of heat stress reaction that varies in between animals.

Many studies have shown that rectal temperature and respiration rate are great determinants of heat stress and might be utilized to evaluate the danger of the hot environment. Gupta *et al.* (2013) discovered an increase in respiration rate and rectal temperature varying from 39.35 and 47.33°C to 38.97 and 43.66°C, in that order, when goats were subjected to hot temperatures for 6 hours. Correspondingly, the investigation by Hooda and Upadhyay (2014) revealed an increment in respiration rate (RR) and rectal temperature (RT) from 40, 42, and 44°C, as the animals were subjected to extreme condition (heat). Once heat starts to be beyond the normal thermal range, the heat loss evaporative cooling components are triggered. Mortola and Frappell (2000) noted that heat-stressed sheep are basically rely upon respiratory and related methods for cooling regulation.



University of Fort Hare
Together in Excellence

2.11. Supplementation as a management practice of heat stress in sheep

A supplement is a substance that is added to animal feed in order to improve the overall productivity of the animal through feed utilization (Kempton *et al.*, 1977). It is imperative important to make use feed supplement or additives that will increase the acceptability of feed. Osweiler and Carson, (1997) reported good acceptability of diet incorporated with 2.5 percent FSF in West African Dwarf sheep. Hence mineral-rich additives are advised to be included in the diet of heat stressed animals to increase feed acceptance at the same time to mitigate heat stressed. Heat stress negatively affects the metabolism of animals. Heat stress brings about huge loss of minerals of which does not only affect the ability of animals to adapt, it may also lead in illness in severe

cases. In addition, the reduced feed intake during heat stress also leads to reduced mineral intake therefore worsening the condition of mineral deficiencies (Damanhoury and Tayeb, 1992; Pirard *et al.*, 20050).

Low concentration of minerals in the body tissue weakens enzymes and hormonal functioning may be responsible for preventing heat stress, this may even lead to death of animal. Trace elements (particularly Se, Cu and Zn) are counted among the micronutrients that play a vital role during high ambient temperature (Sordillo *et al.*, 1997). A study conducted by Khorsandi *et al.* (2016) rereported that supplementation of heat stressed dairy cows under heat stress with multi-trace minerals reduces overall stress. Calamari *et al.* (2007) stated that mineral supplementation during high ambient temperature must not be considered as a means of covering up for some nutrient but, but also as a mean buffer the effect of high temperature. Rhoads *et al.* (2013) reported that supplementing diet with mineral additive that improve insulin sensitivity may be good tactic to increase likelihood of surviving lethal heat stress.



University of Fort Hare
Together in Excellence

2.12. Summary

Heat stress is counted among factors that hold back sheep from performing to their full genetically ability. High ambient temperatures directly impact livestock production by reducing feed intake and growth rate, therefore, that eventually lead to poor production. However, there are many strategies that have been employed to fight heat stress including provision of shade, use of sprinklers to cool down animal body temperature and air conditioner but they are not effective and sometimes not applicable during substantial need to install them. Hence using mineral supplements that is cheap, available in abundance seem too helpful to improve feed acceptability and mitigating heat stress at the same time.

2.13. Reference

- Abd El-Tawab, M.M., Youssef, M.I., Bakr H.A., Fthenakis G.C., Giadinis, N.D.** 2016. Role of probiotics in nutrition and health of small ruminants. *Polish Journal of Veterinary Sciences*.19:893–906.
- Abouheif, M.A. and Alsobayel, A.A.** 1982. Reproductive performance of Saudi Arabian black Najdi ewes under the local environment of the Riyadh area. *World Review of Animal Production*, **18**(9): 56.
- Abouheif, M.A. and Alsobayel, A.A.** 1983. Environmental and genetic factors influencing birth weight of black Najdi lambs. *World review of animal production*, **3**(1): 65-71.
- Adebiyi, O.A., Sokunbi, O.A. and Ewuola, E.O.** 2009. Performance evaluation and bone characteristics of growing cockerel fed diets containing different levels of diatomaceous earth. *Middle East Journal of Scientific Research*, **4**(1): 36-39.
- Al-Haidary, A.A.** 2004. Physiological responses of Naimey sheep to heat stress challenge under semi-arid environments. *International Journal of Agricultural Biology*, **2**:307-309.
- Atwood, S.B., Provenza, F.D., Wiedmeier, R.D. and Banner, R.E.** 2001. Influence of free-choice vs mixed-ration diets on food intake and performance of fattening calves. *Journal of Animal Science*, **79**(12): 3034-3040.
- Baumont, R., Prache, S., Meuret, M. and Morand-Fehr, P.** 2000. How forage characteristics influence behaviour and intake in small ruminants: a review. *Livestock Production Science*, **64**(1): 15-28.

- Bennett, D.C., Yee, A., Rhee, Y.J. and Cheng, K.M.** 2011. Effect of diatomaceous earth on parasite load, egg production, and egg quality of free-range organic laying hens. *Poultry science*, **90**(7): 1416-1426.
- Bernabucci, U., Basiricò, L., Morera, P., Dipasquale, D., Vitali, A., Cappelli, F.P. and Calamari, L.U.I.G.I.** 2015. Effect of summer season on milk protein fractions in Holstein cows. *Journal of dairy science*, **98**(3):1815-1827.
- Boufennara, S., Lopez, S., Bousseboua, H., Rodríguez, R.B. and Bouazza, L.** 2012. Chemical composition and digestibility of some browse plant species collected from Algerian arid rangelands. *Spanish journal of agricultural Research*, **6**(1): 88-98.
- Calamari, L., Abeni, F., Calegari, F. and Stefanini, L.** 2007. Metabolic conditions of lactating Friesian cows during the hot season in the Po valley. 2. Blood minerals and acid-base chemistry. *International journal of biometeorology*, **52**(2): 97-107.
- Castel, J.M., Mena, Y., Delgado-Pertúñez, M., Camúñez, J., Basulto, J., Caravaca, F., Guzmán-Guerrero, J.L. and Alcalde, M.J.** 2003. Characterization of semi-extensive goat production systems in southern Spain. *Small Ruminant Research*, **47**(2): 133-143.
- Chandra, G. and Aggarwal, A.** 2009. Effect of DL--Tocopherol Acetate on Calving Induced Oxidative Stress in Periparturient Crossbred Cows During Summer and Winter Seasons. *Indian Journal of Animal Nutrition*, **26**(3): 204-210.
- Chaudhary, S.S., Singh, V.K., Upadhyay, R.C., Puri, G., Odedara, A.B. and Patel, P.A.** 2015. Evaluation of physiological and biochemical responses in different seasons in Surti buffaloes. *Veterinary world*, **8**(6): 727.

- Cloete, S.W.P. and Durand, A.** 2000. Increasing lamb output by crossing commercial Merino ewes with South African Meat Merino rams. *Australian Journal of Experimental Agriculture*, **40**(1): 11-16.
- Cloete, S.W.P., Olivier, J.J., Sandenbergh, L. and Snyman, M.A.** 2014. The adaption of the South Africa sheep industry to new trends in animal breeding and genetics: A review. *South African Journal of Animal Science*, **44**(4): 307- 321.
- Collier, R.J. and Zimbelman, R.B.** 2007. February. Heat stress effects on cattle: What we know and what we don't know. In *22nd Annual Southwest Nutrition & Management Conference*, **2**: 76-83.
- Dagnew, Y., Urge, M., Tadesse, Y. and Gizaw, S.** 2017. Sheep production and breeding systems in north western lowlands of Amhara region, Ethiopia: implication for conservation and improvement of Gumz sheep breed. *Open Journal of Anim Science*, **7**(2): 179-97.
- Damanhour, Z.A. and Tayeb, O.S.** 1992. Animal models for heat stroke studies. *Journal of pharmacological and toxicological methods*, **28**(3): 119-127.
- De Rancourt, M., Fois, N., Lavín, M.P., Tchakérian, E. and Vallerand, F.** 2006. Mediterranean sheep and goats production: An uncertain future. *Small Ruminant Research*, **62**(3): 167-179.
- DeSimone, J.A., Beauchamp, G.K., Drewnowski, A. and Johnson, G.H.** 2013. Sodium in the food supply: challenges and opportunities. *Nutrition reviews*, **71**(1): 52-59.
- Digby, S.N.; Chadwick, M.A.; Blache, D.** 2011. Salt intake and reproductive function in sheep. *Animal*, **5**:1207–1216.



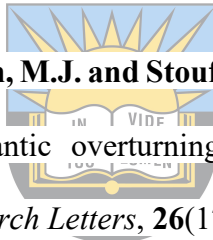
University of Fort Hare
Together in Excellence

Dikmen, S. and Hansen, P.J. 2009. Is the temperature-humidity index the best indicator of heat stress in lactating dairy cows in a subtropical environment.? *Journal of dairy science*, **92**(1): 109-116.

Dikmen, S., Orman, A. and Ustuner, H. 2011. The effect of shearing in a hot environment on some welfare indicators in Awassi lambs. *Tropical animal health and production*, **43**(7): 1327.

Dikmen, S., Ustuner, H., Turkmen, I.I. and Ogan, M. 2009. Fattening performance and feed source preference of native Awassi lambs fed individually in a cafeteria feeding system. *Tropical animal health and production*, **41**(4): 485-491.

Dixon, K.W., Delworth, T.L., Spelman, M.J. and Stouffer, R.J. 1999. The influence of transient surface fluxes on North Atlantic overturning in a coupled GCM climate change experiment. *Geophysical Research Letters*, **26**(17): 2749-2752.



University of Fort Hare

Eltawil, E.A. and Narendran, R. 1990. Ewe productivity in four breeds of sheep in Saudi Arabia. *World Review of Animal Production*, **25**(1): 93-96.

Emeruwa, C.H. 2016. Growth Performance of West African Dwarf Sheep Fed Diets Supplemented with Fossil Shell Flour, Doctoral dissertation, University of Ibadan, Nigeria.

Galyean, M.L., Perino, L.J. and Duff, G.C. 1999. Interaction of cattle health or immunity and nutrition. *Journal of Animal Science*, **77**(5): 1120-1134.

Garmyn, A.J., Hilton, G.G., Mateescu, R.G., Morgan, J.B., Reecy, J.M., Tait, R.G., Jr.; Beitz, D.C., Duan, Q., Schoonmaker, J.P. and Mayes, M.S. 2011. Estimation of relationships between mineral concentration and fatty. *J. Anim. Sci.* 2011, 89, 2849–2858.

- Garnett, S.T.** 2010. Constraints to effective adoption of innovative livestock production technologies in the Rift Valley (Kenya). *Journal of arid environments*, **96**:9-18.
- Getu, A.** 2015. Review on Challenges and Opportunities Sheep Production: Ethiopia. *African Journal of Basic & Applied Sciences*, **7**(4): 200-205.
- Ginane, C., Petit, M. and D'Hour, P.** 2003. How do grazing heifers choose between maturing reproductive and tall or short vegetative swards.? *Applied Animal Behaviour Science*, **83**(1): 15-27.
- Gupta, M., Kumar, S., Dangi, S.S. and Jangir, B.L.** 2013. Physiological, biochemical and molecular responses to thermal stress in goats. *International Journal of Livestock Research*, **3**(2): 27-38.
- Gyanendra, S., Hooda, O.K., Mahapatra, R.K., Meur, S.K. and Varshney, V.P.** 2009. Ameliorative effect of zinc and manganese supplementation in buffalo calves during hot climatic conditions. *Indian Journal of Animal Sciences*, **79**(11): 1153-1155.
- Habeeb, A.A., Gad, A.E. and Atta, A.M.** 2018. Temperature-humidity indices as indicators to heat stress of climatic conditions with relation to production and reproduction of farm animals. *International Journal of Biotechnology and Recent Advances*, **1**(2): 35-50.
- Hafez, E.S.E. and Hafez, B.** 2013. *Reproduction in farm animals*. John Wiley and Sons.
- Hooda, O.K. and Upadhyay, R.C.** 2014. Physiological responses, growth rate and blood metabolites under feed restriction and thermal exposure in kids. *Journal of Stress Physiology & Biochemistry*, **10**(2): 78.



University of Fort Hare

- Ikusika, O.O., Mpendulo, C.T., Zindove, T.J. and Okoh, A.I.** 2019. Fossil Shell Flour in Livestock Production: A Review. *Animals*, **9**(3): 70.
- Invernizzi, G., Modina, S., Corbani, D., Bronzo, V., Pisani, L.F., Caputo, J.M., Agazzi, A., Dell’Orto, V. and Savoini, G.** 2016. Hepatic and subcutaneous adipose tissue variations in transition dairy goats fed saturated or unsaturated fat supplemented diets. *Small ruminant research*, **144**: 211-219.
- Johnson, P.L., Purchas, R.W., McEwan, J.C. and Blair, H.T.** 2005. Carcass composition and meat quality differences between pasture-reared ewe and ram lambs. *Meat science*, **71**(2): 383-391.
- Kempton, T. J., Nolan, J. U. and Leng, R. A.** 1977. Principles for the use of non-protein nitrogen and bypass proteins in diets of ruminants. *Wid Anim. Rev.* 22,2-10.
- Khorsandi, S., Riasi, A., Khorvash, M., Mahyari, S.A., Mohammadpanah, F. and Ahmadi, F.** 2016. Lactation and reproductive performance of high producing dairy cows given sustained-release multi-trace element/vitamin ruminal bolus under heat stress condition. *Livestock Science*, **187**:146-150.
- Li, Y.X., Zhang, X.L., Li, W., Lu, X.F., Liu, B. and Wang, J.** 2013. The residues and environmental risks of multiple veterinary antibiotics in animal faeces. *Environmental monitoring and assessment*, **185**(3): 2211-2220.
- Macy, A.** 2000. Organic livestock handbook. Canadian Organic Growers Inc., Ottawa, Ontario, Canada.

Mapiye, C., Chimonyo, M., Dzama, K. 2009. Seasonal dynamics, production potential and efficiency of cattle in the sweet and sour communal rangelands in South Africa. *Journal of Arid Environments*, **73**: 529-536.

Marai, I.F.M., Ayyat, M.S. and El-Monem, U.A. 2001. Growth performance and reproductive traits at first parity of New Zealand White female rabbits as affected by heat stress and its alleviation under Egyptian conditions. *Tropical animal health and production*, **33**(6): 451-462.

Marai, I.F.M., Ayyat, M.S. and El-Monem, U.A. 2001. Growth performance and reproductive traits at first parity of New Zealand White female rabbits as affected by heat stress and its alleviation under Egyptian conditions. *Tropical animal health and production*, **33**(6): 451-462.



Marai, I.F.M., El-Darawany, A.A., Fadiel, A. and Abdel-Hafez, M.A.M. 2007. Physiological traits as affected by heat stress in sheep a review. *Small ruminant research*, **71**(1-3): 1-12.
Together in Excellence

Marai, I.F.M., El-Darawany, A.A., Fadiel, A. and Abdel-Hafez, M.A.M. 2007. Physiological traits as affected by heat stress in sheep - a review. *Small ruminant research*, **71**(1-3): 1-12.

Marai, J.W. 2011. The influence of flush feeding with different nitrogen sources on ovulation and conception rates in Dohne-Merino ewes. Dissertation submitted to the Department of Animal Science. University of Pretoria.

Marino, R., Atzori, A.S., D'Andrea, M., Iovane, G., Trabalza-Marinucci, M. and Rinaldi, L. 2016. Climate change: Production performance, health issues, greenhouse as emissions

and mitigation strategies in sheep and goat farming. *Small Ruminant Research*, **135**: 50-59.

Mazzullo, G., Rifici, C., Caccamo, G., Rizzo, M. and Piccione, G. 2014. Effect of different environmental conditions on some haematological parameters in cow. *Annals of Animal Science*, **14**(4): 947-954.

Meier, J.S., Kreuzer, M. and Marquardt, S. 2012. Design and methodology of choice feeding experiments with ruminant livestock. *Applied Animal Behaviour Science*, **140**(3-4): 105-120.

Mlambo, V. and Mapiye, C. 2015. Towards household food and nutrition security in semi-arid areas: What role for condensed tannin-rich ruminant feedstuffs. *Food Research International*, **76**: 953-961.



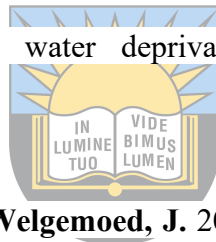
Modirsanei, M., Mansoori, B., Khosravi, A.R., Kiaei, M.M., Khazraeinia, P., Farkhoy, M. and Masoumi, Z. 2008. Effect of diatomaceous earth on the performance and blood variables of broiler chicks during experimental aflatoxicosis. *Journal of the Science of Food and Agriculture*, **88**(4): 626-632.

Morand-Fehr, P. 2003. Dietary choices of goats at the trough. *Small ruminant research*, **49**(3): 231-239.

Morris, W. 2017. Social media, an entrepreneurial opportunity for agriculture-based enterprises. *Journal of Small Business and Enterprise Development*, **4**: 141–143.

Mortola, J.P. and Frappell, P.B. 2000. Ventilatory responses to changes in temperature in mammals and other vertebrates. *Annual Review of Physiology*, **62**(1): 847-874.

- Mvumi, B.M., Stathers, T.E., Kaparadza, V., Mukoyi, F., Masiwa, P., Jowah, P. and Riwa, W.** 2006. Comparative insecticidal efficacy of five raw African diatomaceous earths against three tropical stored grain Coleopteran pests: *Sitophilus zeamais*, *Tribolium castaneum* and *Rhyzopertha dominica*. In *Proceedings of the 9th international conference on stored-product protection, Campinas, Sao Paulo, Brazil*, **6**:15-18.
- Nardone, A., Ronchi, B., Lacetera, N., Ranieri, M. S., Bernabucci, U.** 2010. Effects of climate changes on animal production and sustainability of livestock systems. *Livestock Science*, **130**: 57-69.
- Nejad, J.G. and Sung, K.I.** 2017. Behavioral and physiological changes during heat stress in Corriedale ewes exposed to water deprivation. *Journal of animal science and technology*, **59**(1): 13.
- Nowers, C.B., Nobumba, L.M. and Welgemoed, J.** 2017. The effect of overnight kraaling on sheep production in the Sourveld areas of the Eastern Cape Province, South Africa. *Applied Animal Husbandry and Rural Development*, **10**(1): 9-16.
- Osweiler T. and Carson S.** 1997. Evaluation of diatomaceous earth fed to sheep and goat. *Journal of Agriculture and Science*, **92**:499–503.
- Pirard, P., Vandentorren, S., Pascal, M., Laaidi, K., Le, A.T., Cassadou, S. and Ledrans, M.** 2005. Summary of the mortality impact assessment of the 2003 heat wave in France. *Eurosurveillance*, **10**(7): 153-156.



Together in Excellence

- Price, C.T., Koval, K.J. and Langford, J.R.** 2013. Silicon: a review of its potential role in the prevention and treatment of postmenopausal osteoporosis. *International journal of endocrinology*, **6**(4): 34.
- Purwar, V., Oberoi, P.S. and Dang, S.A.** 2017. Effect of feed supplement and additives on stress mitigation in Karan Fries heifers. *Veterinary World*, EISSN: 2231-0916
- Ratnakaran, A.P., Sejian, V., Sanjo Jose, V., Vaswani, S. and Bagath, M.** 2017. Behavioral responses to livestock adaptation to heat stress challenges. *Asian Journal of Animal Science*, **11**(1): 1-13.
- Rhoads, R.P., Baumgard, L.H., Suagee, J.K. and Sanders, S.R.** 2013. Nutritional interventions to alleviate the negative consequences of heat stress. *Advances in nutrition*, **4**(3): 267-276.
- Saker, K.E., Fike, J.H., Veit, H. and Ward, D.L.** 2004. Brown seaweed-(Tasco™) treated conserved forage enhances antioxidant status and immune function in heat-stressed wether lambs. *Journal of animal physiology and animal nutrition*, **88**(3-4): 122-130.
- Samer, M., Loebstin, C., Fiedler, M., Ammon, C., Berg, W., Sanftleben, P. and Brunsch, R.** 2011. Heat balance and tracer gas technique for airflow rates measurement and gaseous emissions quantification in naturally ventilated livestock buildings. *Energy and Buildings*, **43**(12): 3718-3728.
- Sarijit, J., Steve, S., Micheal, E. and Rodney, D.** 2016. Animal Feed Compositions Including Spent Filter Media Containing Diatomaceous Earth. U.S. Patent Application 15/095,608.
- Scharenberg, A., Arrigo, Y., Gutzwiller, A., Soliva, C.R., Wyss, U., Kreuzer, M. and Dohme, F.** 2007. Palatability in sheep and in vitro nutritional value of dried and ensiled sainfoin

(*Onobrychis viciifolia*) birdsfoot trefoil (*Lotus corniculatus*), and chicory (*Cichorium intybus*). *Archives of Animal Nutrition*, **61**(6): 481-496.

Sejian, V., Kumar, D., Gaughan, J.B. and Naqvi, S.M. 2017. Effect of multiple environmental stressors on the adaptive capability of Malpura rams based on physiological responses in a semi-arid tropical environment. *Journal of Veterinary Behavior*, **17**: 6-13.

Sejian, V., Maurya, V.P., Kumar, K. and Naqvi, S.M.K. 2012. Effect of multiple stresses (thermal, nutritional, and walking stress) on the reproductive performance of Malpura ewes. *Veterinary medicine international*, **12**(6): 78.

Shafie, M.M., Murad, H.M., El-Bedawy, T.M. and Salem, S.M. 1994. Effect of heat stress on feed intake, rumen fermentation and water turnover in relation to heat tolerance response by sheep. *Egyptian Journal of Animal Production*, **31**(2): 317-327.

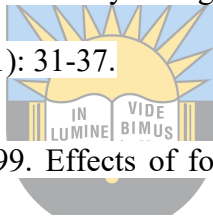
Sharma, M.C., Rupasi, T. and Pankaj, K. 2009. Area specific mineral mixture: need and importance. *Vets communications*, **4**(1): 3-8.

Smiles, U., Ezeano, C.I. and Ochiaka, C.D. 2018. Climate change and Adaptation Coping Strategies among Sheep and Goat Farmers in Ivo Local Government Area of Ebonyi State, Nigeria. *Sustainability, Agriculture, Food and Environmental Research*, **6**(2):34.

Soetan, K.O., Olaiya, C.O. and Oyewole, O.E. 2010. The importance of mineral elements for humans, domestic animals and plants: A review. *African Journal of Food Science*, **4**: 200–222.

Sordillo, L.M., Shafer-Weaver, K. and DeRosa, D. 1997. Immunobiology of the mammary gland. *Journal of dairy science*, **80**(8):1851-1865.

- Swanepoel, J.W., Van Wyk, J.B., Cloete, S.W.P. and Delpont, G.J.** 2007. Short communication inbreeding in the Dohne Merino breed in South Africa. *South African Journal of Animal Science*, **37**(3): 176-179.
- Swenson, M.J. and Reece, W.O.** 2006. Heat tolerance in Brazilian sheep: physiological and blood parameters. *Tropical Animal Health and Production*, **41**(1): 95-101.
- Tadesse., Negesse, T and Abebe, G.** 2015. Sheep production and marketing system in southern Ethiopia: the case of Awassazuria district. *Trop Anim Health Prod* (2015) 47:1417–1425
- Van Wyk, J.B., Swanepoel, J.W., Cloete, S.W., Olivier, J.J. and Delpont, G.J.** 2008. Across flock genetic parameter estimation for yearling body weight and fleece. *South African Journal of Animal Science*, **38**(1): 31-37.
- Villalba, J.J. and Provenza, F.D.** 1999. Effects of food structure and nutritional quality and animal nutritional state on intake behaviour and food preferences of sheep. *Applied Animal Behaviour Science*, **63**(2): 145-163.
- Vitali, A., Segnalini, M., Bertocchi, L., Bernabucci, U., Nardone, A. and Lacetera, N.** 2009. Seasonal pattern of mortality and relationships between mortality and temperature-humidity index in dairy cows. *Journal of Dairy Science*, **92**(8): 3781-3790.
- Weniger, J.H. and Stein, M.** 1992. Einfluss von Umgebungstemperatur und Luftfeuchte auf die Nahrstoffverdaulichkeit beim Schaf. Problemstellung, Durchfuehrung der Untersuchungen, Verdaulichkeit. *Zuchtungskunde*, **64**: 148-155.



University of Fort Hare

excellence

Yasha, A.; De La, F.; Luanna, S.; Batista, F.; Af, S.; Elb, S. 2016. Growth and reproduction hormones of ruminants subjected to heat stress. *Journal of Animal Behaviour Biometeorol.* **5**(1): 7-12.

Young, J.M., Tromp, J. and Thompson, A.N. 2014. The critical control points for increasing reproductive performance can be used to inform research priorities. *Animal Production Science*, **54**(6): 645-655.



University of Fort Hare
Together in Excellence

Chapter 3: Feed preference of Dohne Merino rams fed varying levels of fossil shell flour

(This manuscript is under review at Journal of Animal Production Science)

Manuscript ID: AN19589

Abstract

A total of twenty-four Dohne Merino rams (average 19.8 ± 0.43 kg body weight) were used to determine feed acceptability of varying inclusion levels of fossil shell flour (0g FSF/kg, 20g FSF/kg, 40g FSF/kg and 60g FSF/kg). The diet preferred was assessed from the coefficient of preference (CoP) value calculated from the ratio between the intakes of each individual diet divided by the average intake of the different diets. The feed intake and coefficient of preference was higher in rams supplemented with 60g FSF/kg ($P < 0.05$). Physical attributes of experimental diets were adjusted based on colour, smell and texture. The colour of experimental diet were brown in 0g FSF/kg while in 20 FSF/kg, 40g FSF/kg and in 60g FSF/kg were light brown ($P < 0.05$). All the experimental diets had a pleasant smell ($P < 0.05$). The texture of diets with 0g FSF/kg was moderately firm while diets 20g FSF/kg, 40g FSF/kg and 60g FSF/kg had a firm texture. The order of preference of diets supplemented with fossil shell flour in terms of feed intake by Dohne Merino rams was: 60g FSF/kg > 40g FSF/kg > 20gFSF/kg > 0g FSF/kg. Therefore, FSF has potential as a supplement for sheep showing better acceptability by Dohne Merino rams as inclusion levels of FSF increased in the diets.

Key words: acceptability, Dohne Merino rams, feed preference and fossil shell flour

3.1. Introduction

In most tropical countries, livestock is mostly reared under extensive production system, where animals in these countries mainly depend on natural pastures for their nutrients (Komwihangilo *et al.*, 2001). The disadvantage of using this kind of farming system is a scarcity of forage during the dry season which forces the animals to consume a forage of low quality (Dumont *et al.*, 1995; Mapiye *et al.*, 2009). In addition, an extensive farming system compromises the growth rate of the animal by reducing over 50% of its live weight and therefore increases morbidity and mortality, which in turn leads to severe economic losses (Ilori *et al.*, 2013). Furthermore, a number of researchers (Homann *et al.*, 2007; Kronberg and Malechek, 1997; Pfister And Malechek, 1986) have reported that there is a significant limitation on the productivity of domestic sheep regardless of their potential to survive in natural forage.



Over the years several strategies have been employed to reduce the lack of quality forage for livestock during the dry season. The use of roughages and concentrates was discovered as the most aiding strategy during such situations (Ilori, 2013). In addition, concentrates are usually far more expensive and feed accounts for 50% to 75% of total production costs (Herrero *et al.*, 2016). Thus there's a need to make full use of cheaper and available nutritious alternative sources of feed in order to maximize the livestock production. Several positive reports have been made about less popular silica-rich powder (fossil shell flour), which could possibly relieve the critical shortage of feed if given maximum research attention (Weaver *et al.*, 2013; Wiewiora *et al.*, 2015).

Recent studies (Ikusika *et al.*, 2019; Koster, 2013) have shown that apart from being a mineral-rich additive FSF can be used as an animal growth promoter, natural anthelmintic, and vaccine adjuvant in livestock. The FSF is a naturally occurring, silicon rich sedimentary rock made up of fossilized remains of millions of diatoms, a type of hard-shelled plant algae originally deposited

millions of years ago in the earth from dried up seas and lakes (Ikusika *et al.*, 2019; Koster, 2013). The FSF is cheap, it is available in abundance and it has nontoxic characteristics (Ikusika *et al.*, 2019). However, little research efforts have been stimulated to explore the potential benefits of FSF as dry season feed for ruminants.

Intake of feed voluntarily is relatively considered as one of the major indicators of feed quality and is well recognized as a potential way that showcase the performance of animals (Coleman and Moore, 2003). Moreover, many authors (Kalio and Larbi, 2006; Mtenga *et al.*, 1992) have reported that the feed intake of any diet is affected by what animals prefer as well as how acceptable the feed is and that related to the behaviour of the animal. Likewise, the degree of preference and acceptability of the diet is one of the key things to note when determining the best feedstuff (Ferguson *et al.*, 2002; Ganskopp *et al.*, 2019). Thus, the current study aimed to investigate the extent to which fossil shell flour can form part of a ration for Dohne Merino ram without having a negative effect on feed preference and intake. It was hypothesized that there is no significant effect of the FSF inclusion on the feed acceptability of Dohne Merino rams.

3.2. MATERIAL AND METHODS

3.2.1. Ethical consideration

The way in which the experiment was conducted complied with the standards set out by the Animal Ethics Committee of the University of Fort Hare (Ethical No: MPE011SMWA01/19/A). All trial procedures were conducted with the moral standards of experimentation given by the board of ethics on animal use of the Society for the Prevention of Cruelty to Animals (SPCA) (Constitutional Court of South Africa, 2013).

3.2.2. Study site

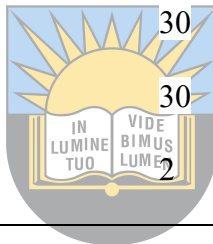
The study took place at the Honeydale Research farm of the University of Fort Hare, located in Alice, Eastern Cape Province, of South Africa. The farm lies at 32°46' S and 26°50' E, at an altitude of 535 metres above sea level. Honeydale Research farm has temperature of an approximate 24.6 °C maximum with the minimum temperature being 11.1 °C, and the average temperature is 17.8 °C. The annual rainfall of the area is 575mm and is often during the months of November to March also referred to as the summer months. The vegetation is dominated by grasses such as *Themeda triandra* and *Cymbopogon plurinodis* with woody plants such as *Acacia karroo* and shrubs encroaching the grazing lands (Mucina and Rutherford, 2006).

3.2.3. Management of rams, feeding and experimental design

A total of twenty-four Dohne Merino rams, aged 8 to 9 months with a weight range of 16 to 24 kg, averaging (19.8 ± 0.43 kg), were confined in individual pens. Each of the twenty-four rams was drenched with 2 mL of Maxicare to control gastrointestinal nematodes using a drenching gun prior to the commencement of the experiment. The preference test lasted for 21 days, where 7 days was allowed for adaptation. A total of four inclusion levels of FSF (0g/kg, 20/kg, 40g/kg and 60g/kg) were used to test feed acceptability in Dohne Merino rams. The lucerne and grass hay was purchased from Border Seed Distributors CC (Queenstown, South Africa). Lucerne and grass hay were chopped to pass through a 5 cm screen. The gross composition of experimental diets is shown on Table 3.1. The diets were formulated according to the recommended standards.

Table 3.1: Composition of experimental diets (DM %)

Ingredients	0g FSF/kg	20g FSF/kg	40g FSF/kg	60g FSF/kg
Maize	8	8	8	8
Sunflower oil cake	10	10	10	10
Molasses	5	5	5	5
Wheat bran	15	15	15	15
Limestone	1.6	1.6	1.6	1.6
Sheep mineral-vitamin premix	0.20	0.20	0.20	0.20
Salt	0.30	0.30	0.30	0.30
Ground Lucerne hay	30	30	30	30
Ground teff hay	30	30	30	30
Fossil shell flour	0	4	4	6

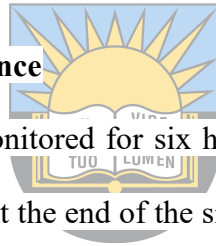


University of Fort Hare
Together in Excellence

set for the small stock by the National Research Council (2007). Rams were fed 3% of their body weight according to ARC (1994), where each diet was introduced to each ram approximately at the same time for a duration of six hours per day in four different feeding troughs. The position of the feeding troughs was interchanged daily to prevent bias and conditioning of the animals in recognizing part of the pen for a particular diet (Ogunbosoye, 2014). Fresh and clean water was made available throughout the successive feeding trial. Rams were assigned to four treatments (0g/kg, 20/kg, 40g/kg and 60g/kg) with six rams allocated to each treatment, making a total of twenty-four rams for the current experiment.

3.3. Measurements

3.3.1. Feed and Coefficient of preference



The amount of feed consumed was monitored for six hours per day after which rams were fed grass hay only for the rest of the day. At the end of the six hours, each of the feeds not consumed was measured by deducting remnants from the amount of feed offered. Preference co-efficient (CoP) was calculated as the ratio of diet intake by an individual to the average intake of all the diets. The percentage of preference was calculated as the ratio of feed intake by an individual to total intake multiplied by 100. Diet was considered acceptable for CoP when it is greater than one while ranking was based on the percentage of preference (Babayemi, 2007; Karbo *et al.*, 1993).

3.3.2. Proximate components

A total of sixteen (four per treatment) samples of the diets were taken for the purpose of laboratory analysis. Samples were oven-dried at 65 °C until a constant weight was obtained for dry matter determination after which they were milled and analyzed for crude protein, crude fibre, ether

extract and ash according to the procedures by (AOAC, 1990). Neutral-detergent fiber (NDF), acid detergent lignin (ADL) and acid-detergent fiber (ADF) were measured using Ankom fiber analyzer²⁰⁰.

3.3.4. Diets evaluation

Physical characteristics (colour, smell and texture) of the diets were assessed according to general procedures of (AOAC, 2005). The colour of the diets was assessed using a colour chart while the smell and texture of the diets were judged by 15 trained panelists. The scale (0 – 5) that was used is shown on Table 3.2.

3.3.5 Feeding behaviour

Feeding behaviour (time spent on feeding, number of visits and number of bites per visit) of twenty-four Dohne Merino ram were recorded with a digital video camera for 6 hours each day during feed preference test. The digital video camera was located on the top view of the pens where clear and adequate footage of each ram was obtained. Filmed feeding behavioural footage was analyzed by two observers who were trained to classify the visual observations of feeding behaviour.

3.4 Data analyses

The data for chemical composition, physical characteristics of diets, feed intake and behavioural patterns of rams were analyzed using the General Linear Model procedure of SAS (2010) and significant means were separated using Duncan's Multiple Range Test.

Table 3.2: Diets score (0 – 5 scale) based on physical characteristics

	Rankings					
	0	1	2	3	4	5
Observation	Very bad	Bad	Going bad	Moderate	Good	Excellent
Colour	Very dark	Dark	Dark brown	Deep brown	Brown	Light brown
Smell	Offensive	Poor	Almost pleasant	Fairly pleasant	Pleasant	Very pleasant
Texture	Slimy	Very soft	Soft	Moderate firm	Firm	Very firm

Source: Abegunde *et al.* (2019).



University of Fort Hare
Together in Excellence

3.5. Results

3.5.1 Chemical composition of diets

The data for the chemical composition of experimental diets is shown in Table 3.3. The dry matter composition (DM) was higher in 0g FSF/kg and 40g FSF/kg ($P<0.05$). Likewise, the crude protein in 20g FSF/kg and 40g FSF/kg ($P<0.05$). There was a significant difference among the values of Crude fibre (CF), where 40g FSF/kg and 60g FSF/kg showed the highest values ($P<0.05$). There was no significant difference ($P<0.05$) among the values of ether extract across the treatments while the value of ash was higher in 40g FSF/kg. The values of acid detergent fibre (ADF) and neutral detergent fibre (NDF) were both higher in 0g FSF/kg ($P<0.05$). There was no significant difference among values of acid detergent lignin (ADL) across the treatments ($P<0.05$).

3.5.2 Physical characteristics of diets

Results on the effects of fossil shell flour on colour, smell and texture are shown in Table 3.4. The colour of experimental diets ranged from light brown to brown. The treatment with 0g FSF/kg was brown whereas treatments with 20g FSF/kg, 40g FSF/kg and 60g FSF/kg were light brown in colour. There was no significant difference ($P<0.05$) among the smell of the experimental diets, for they all have a pleasant smell. The texture of the treatments was moderate firm in 0g FSF/kg while 20g FSF/kg, 40g FSF/kg and 60 FSF/kg had a firm texture. Table 3.5. Shows the physical characteristics of experimental diets.

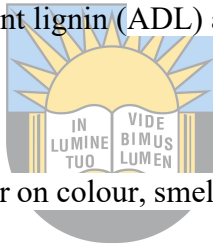


Table 3.3: Chemical composition of experimental diets

	Treatments				SEM	P - Value
Parameters	0g FSF/kg	20g FSF/kg	40g FSF/kg	60g FSF/kg		
Dry matter %	95.5 ^a	93.83 ^b	96.62 ^a	93.60 ^b	0.78	0.0001
Crude protein %	18.20 ^b	21.88 ^a	22.60 ^a	18.80 ^b	0.81	0.0001
Crude fibre %	13.75 ^b	14.13 ^{ab}	15.00 ^a	14.66 ^a	0.87	0.0001
Ether Extract %	1.70 ^a	1.72 ^a	1.80 ^a	1.76 ^a	0.87	0.0001
Ash %	9.40 ^b	9.44 ^b	10.30 ^a	9.33 ^b	0.78	0.0001
NDF%	44.26 ^a	39.2 ^b	39.53 ^b	38.74 ^b	0.90	0.0001
ADF%	40.16 ^a	31.52 ^b	31.64 ^b	31.08 ^b	5.94	0.0001
ADL%	11.07	11.52	11.30	11.34	1.20	0.0001

^{a,b,c}: Means with different superscripts in the same row differ significantly (P<0.05).

ADF: acid detergent fibre; ADL: acid detergent lignin; NDF: neutral detergent fibre; FSF – fossil shell flour and SEM: Standard error of the mean.

Table 3.4: Physical characteristics of dietary treatments

Parameters	Treatments			
	0g FSF/kg	20g FSF/kg	40g FSF/kg	60g FSF/kg
Colour	Brown	Light Brown	Light Brown	Light Brown
Smell	Pleasant	Pleasant	Very Pleasant	Very Pleasant
Texture	Moderately firm	Firm	Firm	Firm



University of Fort Hare
Together in Excellence

Table 3.5: Experimental diets score (0 – 5 scale*) based on physical characteristic

Parameters	Treatments				SEM	P- values
	0g FSF/kg	20g FSF/kg	40g FSF/kg	60g FSF/kg		
Colour	4.61 ^c	5.64 ^b	5.71 ^a	5.72 ^a	0.023	0.0001
Smell	4.03 ^c	4.42 ^b	5.01 ^a	4.43 ^b	0.023	0.0001
Texture	3.03 ^d	4.02 ^c	5.00 ^a	4.23 ^b	0.012	0.0001

*0 – very bad; 1 – bad; 2 – going bad; 3 – moderate; 4 – good; 5 – excellent

^{a,b,c}: Means with different superscripts in the same row differ significantly (P<0.05).

FSF – fossil shell flour and SEM – Standard error of the mean.



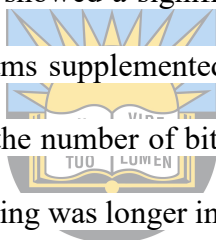
University of Fort Hare
Together in Excellence

3.5.3 Feed intake and coefficient of preference

Results of the effects of FSF on feed intake (FI) and coefficient of preference (CoP) of Dohne Merino rams are shown in Table 3.6. There were significant differences in FI and CoP with varying inclusion levels of fossil shell flour ($P<0.05$). The FI and CoP of diets was higher in rams supplemented by 60g FSF/kg ($P<0.05$). The order of preference was as follows: 60g FSF/kg > 40g FSF/kg > 20gFSF/kg > 0g FSF/kg.

3.5.4 Feeding behaviour of rams

Results of the effects of fossil shell flour on behavioural patterns exhibited by Dohne Merino rams are presented in Table 3.7. The results showed a significant difference ($P<0.05$) in behavioural patterns exhibited by Dohne Merino rams supplemented with fossil shell flour. The number of visits was higher in 60g FSF/kg while the number of bite per visit was lower in both 0g FSF/kg and 20g FSF/kg. The time spent on feeding was longer in 60g FSF/kg and the remaining feed was more in both 0g FSF/kg and 20g FSF/kg ($P<0.05$).



Fort Hare
Together in Excellence

3.5. Discussion

Though there were significant differences that were observed in the values of coefficient of preference (CoP) across the treatments, however, colour, smell and texture of the feed did not have much significant difference in visual inspection. Where colour ranged from light brown to brown, the smell ranged from pleasant to very pleasant while texture ranged from moderately firm to firm. However, the differences between the smell, colour and texture might not have been different to an extent of causing any difference in the preference of feed and intake of feed, due to the slight difference between these parameters.

Table 3.6: Preference of Dohne Merino ram for fossil shell flour

Parameters	Treatments				sem	P-value
	0g FSF/kg	20g FSF/kg	40g FSF/kg	60g FSF/kg		
Intake (kg)	1.96 ^d	2.88 ^c	3.36 ^b	4.80 ^a	0.012	0.0001
Coefficient of preference	0.60 ^d	0.89 ^c	1.03 ^b	1.48 ^a	0.034	0.0001
% Preference	15.08	22.15	25.85	36.92		
Preference ranking	4 th	3 rd	2 nd	1 st		

^{a,b,c}: Means with different superscripts in the same row differ significantly ($P < 0.05$).

FSF – fossil shell flour and SEM – Standard error of the mean.



University of Fort Hare
Together in Excellence

Table 3.7: Preference patterns exhibited by Dohne Merino rams fed fossil shell flour

Parameters	Treatments				SEM	P values
	0g FSF/kg	20g FSF/kg	40g FSF/kg	60g FSF/kg		
NV	2.96 ^d	3.96 ^c	4.64 ^b	6.75 ^a	0.455	0.0001
NBV	3.17 ^c	4.43 ^c	8.00 ^b	11.54 ^a	0.889	0.0001
TSF	1.96 ^c	2.89 ^b	3.36 ^b	4.82 ^a	0.270	0.0001
FR	381.18 ^a	363.14 ^{ab}	354.54 ^b	328.68 ^c	6.751	0.0001

^{a,b,c}: Means with different superscripts in the same row differ significantly ($P < 0.05$).

NV – number of visits, NBV– number of bites per visit, TSF – time spent on feeding, FR– feed remaining, FSF – fossil shell flour and SEM – Standard error of the mean.



University of Fort Hare
Together in Excellence

Moreover, it must also be considered that the pattern of feed intake (FI) and preference observed in the current study for the dry matter intake (DMI) and percentage of CoP were not corresponding with the differences observed in the smell, colour and texture attributes. The current observations are similar to results obtained by Brand *et al.* (2018), who noted that feed intake of grower ostriches fed different inclusion levels of *lupinus angustifolius* were not affected by colour of the feed. It is therefore relatively unclear whether the smell, colour and texture of the feed had an impact on the feed intake and preference of the rams. However, it is likely that some other elements apart from physical characteristics of diets may have had a more important influence for determining feed preference.

The results of the current study are in line with the study conducted by Brand *et al.*, (2008), who noted that the preference green plastic strip was not influenced by physical characteristics of feedstuff. However, the results are different to the findings of Bubier *et al.* (1996), who noted that feed intake and preference of chickens was influenced by the colour of feed. Forbes and Covasa (1995), noted that intake of feed in a system where there is free choice of feeding does not only rely on the physiological state or metabolic requirements of the livestock, but also can be influenced by factors which include previous experience and social interactions. Many researchers (Forbes and Covasa, 1995; Rose *et al.*, 1986), noted that the form, type and nutrient content of the feed greatly impact on diet preference. Brand *et al.* (2018) noted that factors such as breed, trough design, the position of the trough, genetics and management may also contribute to diet selection by livestock. However, in the current study grass hay was the only constituent of the diet to which the rams had access to previously. In addition to that all other factors that possibly could have an impact on the preference of feed were kept constant and the same for all the pens to reduce the risk of any outside influences on feed preference.

The results of the current study showed that the feed intake and preference of Dohne Merino rams were influenced by the inclusion of FSF in diets. Nonetheless, it was observed that the diets that demonstrated superior values in terms of the animals' behavioural patterns: the number of bites, number of visits, time spent feeding on the FSF and the number of bites observed per visit gave a concurrent rise in feed intake. This is vital because intake of feed is known as one of the criteria used to assess acceptability and preference of feedstuffs (Kalio *et al.*, 2006; Ganskopp *et al.*, 2019).

Moreover, it was observed that 40g FSF/kg and 60g FSF/kg were mostly preferred treatments. This may be since animals in free access to feed prefer the one that meets their nutrients requirements. This result showed that the inclusion of FSF to diets of Dohne merino up to 6% for a long time has a positive effect on the dry matter intake of the feed. This could be due to fact that the mineral content of FSF increases the palatability and acceptance of the diets (Da Silva *et al.*, 2016). The results of the current study are similar to those obtained by Emeruwa (2016) who recorded lowest feed intake value to 2% of FSF group (900 ± 9.0) and highest value to 6% of FSF inclusion group (1009 ± 9.0). Moreover, Digby *et al.* (2011) reported that feed intake of sheep increases when NaCl is included as mineral supplement and decreases when NaCl is not included in their diet. Many researchers (Garmyn *et al.*, 2011; Desimone *et al.*, 2013) reported that incorporating minerals such as Mg, Na, K and Ca increase palatability which leads to increased feed preference.

Steel (2006) noted goats that have free access to feed will show variation of feed intake depending on the nutrients available from the feed. Crude protein was relatively high in diets supplemented with 20g FSF/kg, 40g FSF/kg and 60g FSF/kg, hence the rams showed better preferences to these diets, may be due to favorable amount of crude protein. Despite the low levels of CP in the control diet (0g FSF/kg), the CP content in all the experimental diets were way above the critical threshold

value (7%) required for rumen microbial functioning and normal dry matter intake (Ikhimioya, 2008). This suggests that the CP levels of all experimental diets were enough to fulfill maintenance and production requirements of ruminants (Van Soest, 1994). Nevertheless, the current study revealed that Dohne Merino rams exhibited the least acceptance for 0g FSF/kg and 20g FSF/kg. This may be associated with the relatively low mineral value in the feed stuff due to low levels of silica (Apata and Ologhobo, 1994). Numerous studies conducted have shown that there is a preference of certain feedstuff when compared to others which have resulted in a higher ranking of feed preference by animals in the present study (Larbi *et al.*, 1993). The order of preference ranking is shown in Table 3.4: 60g FSF/kg > 40g FSF/kg > 20g FSF/kg > 0g FSF/kg.

3.6 Conclusion

The outcome of the current study showed that FSF has the potential for improving feed acceptability of Dohne Merino rams. With only FSF inclusion levels of 40g FSF/kg and 60g FSF/kg optimum results are obtained when fed to Dohne Merino rams. Therefore, FSF can be used as supplement to forages that have poor nutritional quality especially during the dry seasons.



3.7. References

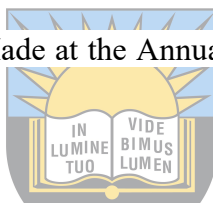
Abegunde, T.O., Odedire, J.A., Akande, C.O., Omoleke, R.T. and Adebayo, M.A. 2019.

Nutritive value and acceptability of dry banana leaves ensiled with cassava peels and urea by west african dwarf sheep. *International Journal of Agriculture and Environmental Research*, **5**(2): 216 - 230.

Apata, D.F. and Ologhobo, A.D. 1994. Biochemical evaluation of some Nigerian legume seeds. *Food Chemistry*, **49**(4): 333-338.

ARC. 1994. *Nutrient Requirements of Ruminant Livestock*. CAB International.

Association of Official Analytical Chemists. 1990. Official Methods of Analysis: Changes in Official Methods of Analysis Made at the Annual Meeting. Supplement, **15**. Association of Official Analytical Chemists.



Association of Official Analytical Chemists. 2005. Official methods of analysis of the Association of Analytical Chemists. Association of Official Analytical Chemists.
Together in Excellence

Babayemi, O.J. 2007. In vitro fermentation characteristics and acceptability by West African dwarf goats of some dry season forages. *African Journal of Biotechnology*, **6**(10).

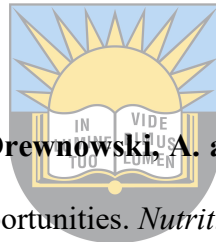
Brand, T.S., Engelbrecht, J.A., Van der Merwe, J. and Hoffman, L.C. 2018. Feed preference of grower ostriches consuming diets differing in *Lupinus angustifolius* inclusion levels. *South African Journal of Animal Science*, **48**(1): 170-185.

Brand, T.S., Kruger, A.C.M. and Aucamp, B.B. 2008. The effect of feeding management practices on the feed intake and production performance of newly hatched ostrich chicks. *Proceedings of the Ostrich Information day, Oudtshoorn, South Africa*.

Bubier, N.E., Lambert, M.S., Deeming, D.C., Ayres, L.L. and Sibly, R.M. 1996. Time budget and colour preferences (with specific reference to feeding) of ostrich (*Struthio camelus*) chicks in captivity. *British poultry science*, **37**(3): 547-551.

Coleman, S.W. and Moore, J.E. 2003. Feed quality and animal performance. *Field Crops Research*, **84**(1-2): 17-29.

Da Silva, I.F., de Souza Rodrigues, R.T., Queiroz, M.A.Á., Chizzotti, M.L., Zanetti, M.A., da Cunha, J.A. and Busato, K.C. 2016. Net requirements of calcium, phosphorus, magnesium, and sulphur for growth of non-descript breed hair lambs of different sex classes in the Brazilian semiarid conditions. *Tropical animal health and production*, **48**(4): 817-822.



DeSimone, J.A., Beauchamp, G.K., Drewnowski, A. and Johnson, G.H. 2013. Sodium in the food supply: challenges and opportunities. *Nutrition reviews*, **71**(1): 52-59.

University of Fort Hare

Digby, S.N., Chadwick, M.A. and Blache, D. 2011. Salt intake and reproductive function in sheep. *Animal*, **5**(8): 1207-1216.

Dumont, B., Meuret, M. and Prud'Hon, M. 1995. Direct observation of biting for studying grazing behavior of goats and llamas on garrigue rangelands. *Small Ruminant Research*, **16**(1): 27-35.

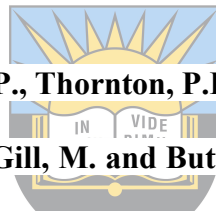
Emeruwa, C.H. 2016. Growth Performance of West African Dwarf Sheep Fed Diets Supplemented with Fossil Shell Flour, Doctoral dissertation, University of Ibadan, Nigeria.

Ferguson, N.S., Bradford, M.M.V. and Gous, R.M. 2002. Diet selection priorities in growing pigs offered a choice of feeds. *South African Journal of Animal Science*, **32**(2): 136-143.

Forbes, J.M. & Covasa, M. 1995. Application of diet selection by poultry with particular reference to whole cereals. *World's Poultry Science Journal*, **51**(2): 149-165.

Ganskopp, D., Myers, B., Lambert, S. and Cruz, R. 1997. Preferences and behavior of cattle grazing 8 varieties of grasses. *Journal of Range Management*, **6**: 578-586.

Garmyn, A.J., Hilton, G.G., Mateescu, R.G., Morgan, J.B., Reecy, J.M., Tait Jr, R.G., Beitz, D.C., Duan, Q., Schoonmaker, J.P., Mayes, M.S. and Drewnoski, M.E. 2011. Estimation of relationships between mineral concentration and fatty acid composition of longissimus muscle and beef palatability traits. *Journal of Animal Science*, **89**(9): 2849-2858.



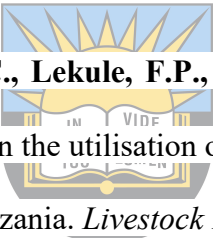
Herrero, M., Henderson, B., Havlik, P., Thornton, P.K., Conant, R.T., Smith, P., Wiersenius, S., Hristov, A.N., Gerber, P., Gill, M. and Butterbach-Bahl, K. 2016. Greenhouse gas mitigation potentials in the livestock sector. *Nature Climate Change*, **6**(5): 452.

University of Fort Hare

Homann, S., Van Rooyen, A.F., Moyo, T. and Nengomasha, Z. 2007. Goat production and marketing: Baseline information for semi-arid Zimbabwe. *International Crops Research Institute for the Semi-Arid Tropics*, **2**: 1–71.

Ikhimiya, I. 2008. Acceptability of selected common shrubs/tree leaves in Nigeria by West African Dwarf goats. *Livestock Research for Rural Development*, **20**(6):90.

Ikusika, O.O. Mpendulo, C.T. Zindove, T.J. and Okoh, A.I. 2019. Fossil Shell Flour in Livestock Production: A Review. *Animals*, **9**(3): 70.

- Ilori, H.B., Salami, S.A., Majoka, M.A. and Okunlola, D.O.** 2013. Acceptability and nutrient digestibility of West African Dwarf goat fed different dietary inclusion of Baobab (*Adansonia digitata*). *IOSR Journal of Agriculture and Veterinary Science*, **6**(3): 22-26.
- Kalio, G.A., Oji, U.I. and Larbi, A.** 2006. Preference and palatability of indigenous and exotic acid soil-tolerant multipurpose trees and shrubs by West African Dwarf sheep. *Agroforestry systems*, **67**(2): 123-128.
- Karbo, N., Barnes, P. and Rudat, H.** 1996. An evaluation of browse forage preferences by sheep and goats in the Northern Guinea Savannah zone, Ghana. In 2. *African Feed Resources Network (AFRNET) Workshop, Harare (Zimbabwe)*, 6-10 Dec 1993. AFRNET.
- Komwihangilo, D.M., Sendalo, D.S.C., Lekule, F.P., Mtenga, L.A. and Temu, V.K.** 2001. Number 6 Farmers' knowledge in the utilisation of indigenous browse species for feeding of goats in semi-arid central Tanzania. *Livestock Research for Rural Development*, **13**(6): 70.
- The logo of the University of Fort Hare, featuring a shield with a sunburst at the top, a book in the center, and the motto 'IN VIDE' below it. The shield is flanked by two lions.
- University of Fort Hare
Together in Excellence
- Koster, H.** 2013. Diatomite in animal feed. <http://www.Agrisilica.co.za/pdf/eng/diatom>. Accessed 18 May 2019.
- Kronberg, S.L. and Malechek, J.C.** 1997. Relationships between nutrition and foraging behavior of free-ranging sheep and goats. *Journal of Animal science*, **75**(7): 1756-1763.
- Larbi, A., Jabbar, M.A., Orok, E.J., Idiong, N.B. and Cobbina, J.** 1993. *Alchornea cordifolia*, a promising indigenous browse species adapted to acid soils in southeastern Nigeria for integrated crop-livestock agroforestry production systems. *Agroforestry Systems*, **22**(1): 33-41.

- Mapiye, C., Chimonyo, M. and Dzama, K.** 2009. Seasonal dynamics, production potential and efficiency of cattle in the sweet and sour communal rangelands in South Africa. *Journal of Arid Environments*, **73**(4-5): 529-536.
- Mtenga, L.A., Komwihangilo, D.M. and Kifaro, G.C.** 1992. December. Selectivity in sheep and goats fed Albizia, Gliricidia, Leucaena and Tamarind multipurpose trees. In *Proceedings of the Second Biennial Workshop of African Small Ruminant Network (SRNET)*, **6**: 7-11.
- Mucina, L. and Rutherford, M.C.** 2006. 'The vegetation of South Africa, Lesotho and Swaziland.' (South African National Biodiversity Institute).
- National Research Council (US).** 2007. Committee on Nutrient Requirements of Small Ruminants, National Research Council, Committee on the Nutrient Requirements of Small Ruminants, Board on Agriculture, Division on Earth and Life Studies. *Nutrient requirements of small ruminants: sheep, goats, cervids, and new world camelids*.
- Ogunbosoye, D. O. and Otukoya, F. K.** 2014. Evaluation of preference and intake of Browse species by West African Dwarf goats in Nigeria. *International Journal of Innovative Research and Development*, **3**(3): 168-176.
- Pfister, J.A. and Malechek, J.C.** 1986. Dietary selection by goats and sheep in a deciduous woodland of northeastern Brazil. *Journal of Range Management*, **50**: 24-28.
- Rose, S.P., Burnett, A. and Elmajeed, R.A.** 1986. Factors affecting the diet selection of choice-fed broilers. *British Poultry Science*, **27**(2): 215-224.
- SAS Institute** .2008. SAS/STAT; user's guide, version 9.2. SAS Inst., Inc., Cary

Steele, M. 2006. Goats. CTA-Macmillan Publishing Ltd., London and Basingstoke, **2**: 152.

Van Soest P. J., Robertson J. B., Lewis B. A. 1994. Methods of dietary fiber, neutral detergent fiber, and non-starch polysaccharides in relation to animal nutrition. *Journal Dairy Science*, **74**: 3583-3597.

Weaver, A., See, M., Hansen, J., Kim, Y., De Souza, A., Middleton, T. and Kim, S. 2013. The use of feed additives to reduce the effects of aflatoxin and deoxynivalenol on pig growth, organ health and immune status during chronic exposure. *Toxins*, **5**(7): 1261-1281.

Wiewiora, M.A., Lukasiewicz, M., Bartosik, J.U., Makarski, M.A. and Niemiec, T.O. 2015. Diatomaceous earth in the prevention of worm infestation in purebred pigeons. *Annals of Warsaw University of Life Sciences-SGGW. Animal Science*, **54**: 71- 96.



University of Fort Hare
Together in Excellence

Chapter 4: Effects of fossil shell flour supplementation on heat tolerance of Dohne Merino rams

(This manuscript has been published in Journal of Veterinary and Animal Science, vol. 10:100-133)

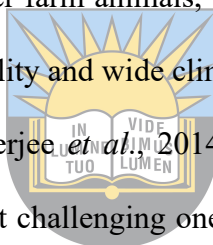
Abstract

The objective of the current study was to determine the effects of fossil shell flour (FSF) supplementation on heat tolerance of Dohne Merino sheep. A total of twenty-four (n=24) Dohne Merino rams of about 8 to 9 months old, weighing 19.8 ± 0.43 kg, were confined in individual pens for a period of 100 days. The rams were subjected to four varying inclusion levels of FSF (0g/kg, 20g/kg, 40g/kg and 60g/kg. Water and feed intake were recorded throughout the period of successive feeding. Blood samples were collected on days 0, 45 and 90. The parameters studied included the average daily water intake (ADWI), average feed intake (ADFI), skin temperature (ST), respiration rate (RR), pulse rate (PR), rectal temperature (RT), total plasma protein (TPP), mean corpuscular hemoglobin concentration (MCHC) and mean corpuscular volume (MCV). The inclusion levels of FSF affected water intake and ADFI, showing high ADWI and feed intake in rams subjected to 40g FSF/kg followed by 60g FSF/kg, 20g FSF/kg and 0g FSF/kg ($P < 0.01$) respectively. All the physiological parameters ST, RT, RR, HR and PR decreased with an increase in inclusion levels of FSF ($P < 0.01$). The TPP increased as the levels of FSF increased ($P < 0.01$). The MCHC was low in rams subjected to 60g FSF/kg and in 0 to 40g FSF/kg. It can be concluded that water and feed intake increased with the increasing levels of FSF while the physiological parameters decline as levels of FSF increased. Therefore, FSF can be used as a supplement to rams up to 40g FSF/kg to mitigate heat stress.

Key Words: Heat tolerance, blood metabolites, fossil shell flour, Dohne Merino rams

4.1. Introduction

Sheep are an important species of livestock, providing meat and wool. They play a vital role in the economy of millions of people who earn their livelihood either by directly rearing sheep or indirectly using products from sheep (Gupta *et al.*, 2013). Today, sheep farming is practiced in almost all the inhabited continents of the world, being easily adaptable and instrumental to several civilizations (Skapetas and Kalaitzidou, 2017). Al-Haidary *et al.* (2012) reported that sheep easily adapted under various geographical and environmental conditions even in extreme and harsh climates and performed better than other farm animals, though goats are more heat tolerant than sheep. However, despite sheep adaptability and wide climatic tolerance, the productivity of sheep often declines due to heat stress (Banerjee *et al.*, 2014). Among many types of stressors that animals undergo, heat stress is the most challenging one with the present climatic changes been experienced globally (Silanikove and Koluman, 2015).



University of Fort Hare
Together in Excellence

Heat stress is one most important stressor in semi-arid (Al-Dawood, 2015), and in arid (Silanikove, 2000). Gupta *et al.* (2013) defined heat stress as the perceived discomfort and physiological strain associated with an exposure to an extreme and hot environment. Srikanthakumar *et al.* (2003), Caroprese, (2008) and Al-Dawood (2015) gives temperature tolerance range for adult sheep as - 12 °C to 32 °C. Kiyatkin, (2005) reported that in order to avoid hyperthermia and other related stress in farm animals EU legislated law minimum temperature for each species for the purpose of animal welfare. Nevertheless, the degree of tolerance depends majorly on such factors as physiological condition of the animal as well as the anionic-cationic balance in the blood (Srikanthakumar *et al.* 2003; Cwynar *et al.*, 2014). Korczynski *et al.* (2004) and Al-Dawood (2017)

reported that it needs to strongly control heat stress syndromes in animals during high environmental temperatures. An animal's skin temperature (ST), respiration rate (RR), and rectal temperature (RT) should, therefore, be measured, as a vital and basic indicator of thermal homeostasis of the body.

De Rensis and Scaramuzzi (2003), and Cwynar *et al.* (2014) reported and recommended that rectal temperature and respiratory rate parameters are excellent indicators of thermal balance of the body and as such should be used as a research method. Srikanthakumar *et al.* (2003) and Cwynar *et al.* (2014) reported and confirmed a correlation between the environmental temperature and blood parameters. Several strategies have been used to mitigate heat stress in livestock production. Some of these include provision of shade, adequate provision of water, sprinklers, fans, and air conditioner. However, some of these strategies are expensive and cannot be afforded by communal and small-scale farmers while some are not practicable in certain regions of the world where good water is a scarce commodity. Thus, there is a need to explore the use of other alternative management practices such as the use of feed additives that could reduce massive heat load without affecting production and reproduction performance.

Recently, there is a growing interest in the use of fossil shell flour (FSF) as feed additive in livestock (El Tazi *et al.*, 2014). Fossil shell flour is a naturally occurring, silicon rich sedimentary rock made up of fossilized remains of millions of diatoms, a type of hard-shelled plant algae originally deposited millions of years ago in the earth from dried up seas and lakes (Koster, 2013; Ikusika *et al.*, 2019). Recent studies have shown that FSF can be used as animal growth promoter, natural anthelmintic, and vaccine adjuvant in livestock (Koster, 2013; Ikusika *et al.*, 2019). Fossil shell flour is rich in minerals, cheap, it is available in abundance and has nontoxic characteristics (Ikusika *et al.*, 2019). Though the knowledge on the animal response towards heat stress is

common, nutritional manipulation using feed additive such as FSF remains unexplored. It was hypothesized that the supplementation of Dohne-Merino rams' diets with fossil shell flour will act as a viable alternate strategy to reduce the effect of heat stress. Therefore, this study investigated the effects of fossil shell flour inclusion on heat stress indicators in Dohne-merino rams. It was hypothesized that there is no effect of varying levels of FSF on the heat tolerance response of Dohne Merino rams.

4.2. Material and Methods

4.2.1. Ethical consideration

The ethical considerations are described in Section 3.2.1.

4.2.2. Study site

The study site is described in Section 3.2.2.

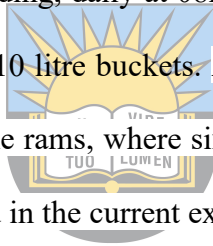


University of Fort Hare
Together in Excellence

4.2.3. Experimental design, management and feedings of rams

A total of twenty-four (n=24) Dohne Merino rams aged 8 to 9 months, ranging from 16 to 24 kg averaging (19.8 ± 0.43 kg), were confined in individual pens at the same facility in the same area under the same environmental conditions. The pens had similar temperatures (35.29°C), relative humidity (76.75%), dry bulb temperature (34.82°C), wet bulb temperature (31.15°C) and the mean temperature-humidity index (THI) was (88.09). Each of twenty-four rams was drenched with 2 mL of Maxicare to control gastrointestinal nematodes using a drenching gun. The experiment lasted for 100 days excluding the 10 days of adaptation period. The study was carried out in summer (from December 2018 till March 2019). The rams were randomly subjected to four

varying levels of FSF (0g/kg, 20/kg, 40g/kg and 60g/kg) in a Completely Randomized Design. The lucerne and grass hay was purchased from Border Seed Distributors CC (Queenstown, South Africa). Lucerne and grass hay were chopped to pass through a 5 cm screen. The chemical composition of fossil shell flour is shown on Table 4.1. Experimental diets were formulated using grass hay, fossil shell flour, wheat bran, sunflower oil cake, maize, molasses, limestone, lucerne hay, salt, and premix. The formulation of experimental diets was in line with recommendations for small stock according to standard set by the National Research Council, (2007). The chemical composition of the experimental diets is shown in Table 3.3. The rams were fed at 3% of their body weight according to Neary (1998). Water and feed were made available to the rams *ad libitum* throughout the period of successive feeding, daily at 08h00 and at 16h00. Fresh and clean water was made available to each ram using 10 litre buckets. A total of four treatments (0g/kg, 20/kg, 40g/kg and 60g/kg) were assigned to the rams, where six rams were allocated to each treatment, making a total of twenty-four rams used in the current experiment.



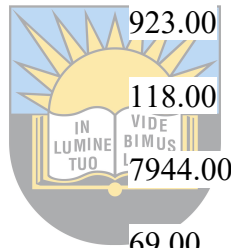
University of Fort Hare
Together in Excellence

4.3 Blood collection procedures

On day 0, 45, 90 seven milliliters (ml) of blood was collected from jugular vein using a graduated syringe and transferred into tubes for the determination of contents and concentrations of red blood cells (RBC), white blood cells (WBC), packed cell volume (PCV) hemoglobin (Hb), total plasma protein (TPP), mean corpuscular hemoglobin concentration (MCHC) and mean corpuscular volume (MCV). All tubes were placed immediately on ice and then transferred to the laboratory within two hours of collection. The plasma was extracted following centrifugation

Table 4.1: Mineral composition of fossil shell flour FSF

Items	Quantity (% or ppm)
DM %	93.00
Ca	0.40
% CaO (calculated from %Ca)	0.55
Mg	0.21
%MgO (calculated from %Mg)	0.34
K%	0.16
Cu (mg/kg)	30.00
Na (mg/kg)	923.00
Zn(mg/kg)	118.00
Fe(mg/kg)	7944.00
Mn(mg/kg)	69.00
P (as P ₂ O ₅)	0.037
Sulfate Sulfur (S)%	0.062
Aluminum (Al) %	0.065
Vanadium (V) %	0.00438
Boron (B) %	0.0023



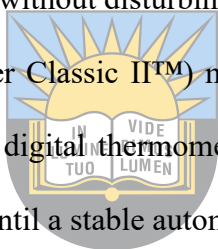
University of Fort Hare
Together in Excellence

at 3000 x g for 10 minutes and stored at -20°C pending analyses (Pambu-Gollah, *et al.*, 2000; Stella *et al.*, 2007).

4.4. Measurements

4.4.1 Physiological parameters

The skin temperature (ST), respiratory rate (RR), heart rate (HR) and rectal temperature (RT) were measured on day 0, 45, 90. In the current study ST was measured from four body parts (abdomen, scrotum, face and from animal's back) using infrared thermometer (ADTEMP™) using methods described by (Sejian *et al.*, 2017). The RR was recorded by counting flank movements per minute using a stopwatch from 5 m without disturbing the rams. The HR was measured using a stethoscope (3M™ Littmann® Master Classic II™) method described by (Verschuren *et al.*, 2008). The RT was measured using a digital thermometer (Uniontech). The thermometer was inserted about 2 to 3 cm in the rectum until a stable automated reading was obtained.



University of Fort Hare
Together in Excellence

4.4.2 Blood biochemical parameters

The packed cell volume (PCV) was estimated using methods described by Olayemi *et al.* (2015). Total red blood cells (RBC) and white blood cells (WBC) were determined using methods by Rana *et al.* (2014). The haemoglobins (Hb) genotypes were determined using cellulose acetate electrophoresis, following the preparation of the haemolysate as described in methods by Osita *et al.* (2019). The hematimetric parameters (Mean Corpuscular Volume – MCV and Mean Corpuscular Hemoglobin Concentration – MCHC) were determined by calculation. The concentration of total plasma proteins (TPP in g/ 100 mL) was determined using a refractometer and the plasma retained in a capillary tube.

4.5 Statistical analyses

The data for feed intake, water intake, physiological and blood biochemical response were analyzed using the repeated measures procedure of SAS/JMP (2018) by multivariate analysis of variance which include the effect of fixed factors, namely treatment (0g FSF/kg, 20g FSF/kg, 40g FSF/kg and 60g FSF/kg) and days (DAY: longitudinal time over which experiment was carried out on days 0, 45 and 90) and the $\text{Trt} \times \text{DAY}$ interaction of the variables studied were analyzed. Significant treatment mean values were compared using the Duncan Multiple Range Test of the same package. The level of statistical significance was set at $P < 0.05$.

The following model ADFI and ADWI was used:

$$Y_{ij} = \mu + D_i + E_{ij}, \text{ where}$$

$$Y_{ij} = \text{Observation (ADFI and ADWI)}$$



$$\mu = \text{Overall mean}$$

University of Fort Hare
Together in Excellence

$$D_i = \text{Effect of dietary treatments (0g FSF/kg, 2g FSF/kg, 4g FSF/kg, 6g FSF/kg)}$$

$$E_{ij} = \text{Random error}$$

While the following model for physiological and blood biochemical response was used:

$$Y_{ij} = \mu + T_i + D_j + (T + D)_{ij} + E_{ij}$$

Where:

Y_{ij} = observation of physiological and blood biochemical parameters (e.g. ST, RT, RR, PR, Hb, TPP, PCV, WBC, RBC, MCV, MCHC and GLU)

μ = the general mean or a common element of all individuals

T = the effect due to the i^{th} treatments (e.g. $i = 1, 2, 3, 4$)

D $_j$ = the effect due to number of days (e.g. $j = 0, 45, 90$)

(T + D) $_{ij}$ = Interaction between treatments and days

E = random error associated with individual observations



University of Fort Hare
Together in Excellence

4.6. Results

4.6.1. Feed and water intake

The results on the effects of fossil shell flour (FSF) on ADWI and the ADFI are shown in Table 4.2. The ADFI and ADWI were both higher in the rams subjected to 40g FSF/kg inclusion level ($P < 0.01$). The interaction of the effects of Trt $\times$ DAY was not significant ($P > 0.05$).

4.6.2. Physiological response

Results on the effects of fossil shell flour on skin temperature (ST), rectal temperature (RT), respiration rate (RR) and heart/pulse rate (PR) are shown in Table 4.3. The effects of treatment with inclusion levels of FSF on physiological responses tested were significantly different across all treatments tested ($P < 0.01$). The ST, RT, RR, and PR were highest in rams subjected to 0g FSF/kg inclusion level ($P < 0.01$). The effects of the days were significant across all physiological parameters tested ($P < 0.01$). The RT and PR were high on day 1 of the experiment ($P < 0.05$), whilst the ST was high on day 45 and on day 1 ($P < 0.01$). The RR was smallest on day 90 ($P < 0.01$). For the interaction of the effects of Trt $\times$ DAY, only on RT and RR were significant ($P < 0.01$). Results on the interaction of the effects of Trt $\times$ DAY on RT and RR are shown in Table 4.4. The RR was high in rams subjected to 0g FSF/kg and 20g FSF/kg, on day 90 ($P < 0.01$). In general, the RT was high on day 1 across treatments tested ($P < 0.01$).

Table 4.2: Effect of fossil shell flour (FSF) on feed and water intake in Dohne merino rams

Parameter	Treatment				SEM	Effect
	T1	T2	T3	T4		
Feed intake (g/day)	257.13 ^b	286.94 ^b	468.17 ^a	464.21 ^a	18.43	***
Water intake (L/day)	1.33 ^c	1.84 ^b	2.33 ^a	1.94 ^{ab}	0.14	**

T1; 0g FSF/kg, T2; 20g FSF/kg T3; 40g FSF/kg, T4; 60g FSF/kg, NS, nonsignificant;

TRT, treatment; SEM, standard error means;

*** $P < 0.0001$, ** $P < 0.01$, * $P < 0.05$



University of Fort Hare
Together in Excellence

Table 4.3: Effect of fossil shell flour on skin temperature, respiration rate, rectal temperature and pulse rate

Parameters	Treatments					Days			Effects			
	T1	T2	T3	T4	SEM	1	45	90	SEM	Trt	DAY	Trt x DAY
ST (°C)	35.67 ^a	32.42 ^b	31.35 ^b	28.17 ^c	0.46	32.95 ^a	33.24 ^a	29.51 ^b	0.40	***	***	NS
RT (°C)	38.94 ^a	38.02 ^b	36.80 ^c	34.08 ^d	0.27	38.07 ^a	36.96 ^b	35.86 ^c	0.23	***	***	***
RR (breath/minutes)	29.92 ^a	25.00 ^b	18.25 ^c	16.25 ^c	0.93	28.19 ^a	21.56 ^b	17.31 ^c	0.80	***	***	***
PR (beats/minutes)	76.83 ^a	76.58 ^a	72.67 ^b	70.08 ^c	0.69	74.88 ^a	72.81 ^b	74.44 ^{ab}	0.61	***	*	NS

T1; 0g FSF/kg, T2; 20g FSF/kg T3; 40g FSF/kg, T4; 60g FSF/kg, ST; skin temperature, RT; rectal temperature, RR; respiration rate, PR; pulse rate, NS, nonsignificant; Trt, treatment; SEM, standard error means; Trt x DAY, treatment and interaction.

***P< 0.0001, **P < 0.01, * P< 0.05.

Table 4.4: Effect of FSF and day interaction on skin temperature and rectal temperature

Parameters	TRT	Days			SEM
		0	45	90	
RR(Breath/minutes)	0g FSF/kg	33.05 ^c	37.60 ^a	36.35 ^b	0.80
	20g FSF/kg	30.05 ^c	34.37 ^a	32.82 ^b	0.80
	40g FSF/kg	28.70 ^d	32.85 ^a	32.50 ^a	0.80
	60g FSF/kg	26.23 ^c	28.15 ^b	30.13 ^a	0.80
RT (°C)	0g FSF/kg	39.15 ^a	39.30 ^a	38.38 ^b	0.47
	20 FSF/kg	38.55 ^a	37.93 ^b	37.58 ^b	0.47
	40 FSF/kg	37.40 ^b	37.65 ^b	35.35 ^b	0.47
	60 FSF/kg	37.18 ^a	32.95 ^b	32.13 ^b	0.47

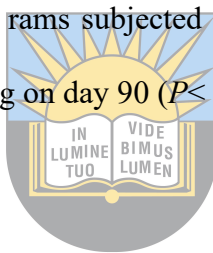
Means followed by different superscripts by row are significantly different at $P < 0.05$. RT; rectal temperature, RR; respiration rate, TRT, treatment; SEM, standard error means



University of Fort Hare
Together in Excellence

4.6.3. Blood biochemical response

The results for all measured blood biochemical parameters (Hb, TPP, Glucose, PCV, WBC, RBC, MCV and MCHC) are shown in Table 4.5. Only the effects of fossil shell flour on blood biochemical response parameters (TPP, MCV and MCHC) are shown in Table 4.6. The TPP was high on 0g FSF/kg and 60g FSF inclusion levels on day 1 ($P < 0.01$). The TPP was high in rams fed 20g FSF/kg and 60g FSF/kg on day 45 ($P < 0.01$). The MCV was high in rams fed 0g FSF/kg and 60g FSF/kg on day 1 ($P < 0.01$). On the other hand, the MCV was high in rams fed 20g FSF/kg and 40 FSF/kg on day 45 and on day 90 ($P < 0.01$). The MCHC was high in rams fed 0g FSF/kg on day 90 ($P < 0.01$). The MCHC was high amongst rams fed 20g FSF/kg on day 45 and on day 90 ($P < 0.01$). The MCHC was high in rams subjected to 40g FSF/kg on day 1 ($P < 0.01$). The MCHC was high in rams fed 60g FSF/kg on day 90 ($P < 0.01$).



4.7 Discussion

The current study focused on assessing the extent at which FSF inclusion levels can reduce heat stress in Dohne Merino rams by measuring ADWI, ADWFI, ST, RR, RP, TPP, MCV and MCHC. The results obtained in the current study have shown that FSF impacts feed intake, water intake, physiological responses and blood biochemical in Dohne Merino rams.

Table 4.5: Effect of fossil shell flour on some blood heamatobiochemical parameters

Parameters	Treatments					Days				Effects		
	T1	T2	T3	T4	SEM	0	45	90	SEM	TRT	DAY	TRT x DAY
Hb (g/100 mL)	9.25 ^a	8.83 ^{ab}	8.28 ^b	8.74 ^{ab}	0.29	10.13 ^a	8.38 ^b	7.83 ^b	0.25	NS	***	***
TPP (g/100 mL)	56.42 ^{bc}	57.67 ^b	63.53 ^b	58.80 ^a	1.17	58.25	58.75	59.95	1.01	**	NS	**
Glucose (mmol/L)	2.53	2.30	2.63	2.60	0.17	2.53	2.60	2.76	0.15	**	**	**
PCV (%)	0.27	0.25	0.23	0.29	0.03	0.23	0.29	0.25	0.03	NS	NS	NS
WBC (x10 ³ /mm ³)	3.60	3.69	3.72	3.79	0.38	2.85 ^b	3.65 ^{ab}	4.19 ^a	0.33	NS	*	NS
MCV (fl)	39.54 ^c	40.63 ^b	41.55 ^{ab}	42.22 ^a	0.59	41.63 ^a	39.55 ^b	41.77 ^a	0.51	**	**	***
MCHC (%)	46.03 ^{bc}	46.52 ^b	52.89 ^a	40.19 ^c	2.09	42.41	43.94	47.86	1.81	**	NS	**
RBC (x10 ⁶ /mm ³)	4.13 ^b	4.14 ^b	4.17 ^b	5.87 ^a	0.33	4.33 ^a	4.82 ^b	5.37 ^b	0.29	**	**	NS

T1; 0g FSF/kg, T2; 20g FSF/kg T3; 40g FSF/kg, T4; 60g FSF/kg, HEM; hemoglobin, TPP; total plasma protein, PCV; packed volume cell, WBC; white blood cells, MCV; mean corpuscular volume, MCHC; mean corpuscular hemoglobin concentration, RBC red blood cell, NS, nonsignificant; TRT, treatment; SEM, standard error means; TRT x DAY, treatment and interaction.

***P< 0.0001, **P < 0.01, * P< 0.05

Table 4.6: Effect of FSF and day interaction on TPP, MCV and MCHC

Parameters	Trt	Days			SEM
		1	45	90	
TPP (g/100 ml)	0g FSF/kg	59.25	57.25	58.75	2.03
	20g FSF/kg	57.00 ^b	61.00 ^a	53.50 ^c	2.03
	40g FSF/kg	54.00 ^b	59.00 ^a	57.50 ^a	2.03
	60g FSF/kg	69.50 ^a	57.75 ^c	63.25 ^b	2.03
MCV (fl)	0g FSF/kg	43.55 ^a	32.65 ^c	42.43 ^b	1.03
	20g FSF/kg	41.28 ^b	42.88 ^{ab}	43.28 ^a	1.03
	40g FSF/kg	39.58 ^b	42.80 ^a	42.27 ^a	1.03
	60g FSF/kg	41.68 ^a	39.88 ^b	39.10 ^b	1.03
MCHC (%)	0g FSF/kg	48.48 ^b	35.48 ^c	54.15 ^a	3.62
	20g FSF/kg	41.73 ^b	50.58 ^a	47.25 ^a	3.62
	40g FSF/kg	62.68 ^a	49.90 ^b	46.10 ^c	3.62
	60g FSF/kg	36.80 ^b	39.83 ^b	43.95 ^a	3.62

Means followed by different superscripts by row are significantly different at $P < 0.05$. TPP; total plasma protein, MCV; mean corpuscular volume, MCHC; mean corpuscular hemoglobin concentration, Trt; treatment, SEM; standard error means, fl; femtolitre.

4.7.1 Effects of FSF on feed and water intake

Feed and water intake are vital attributes in determining the adaptive ability of sheep (Minka and Ayo, 2009; Sejian et al., 2010). When animals are experiencing heat stress, they tend to reduce feed intake and increase their water consumption. The study observed that ADWI and ADFI increased as inclusion levels of fossil shell flour increased, and this could be because rams were not heat stressed since the fossil shell flour supplemented in their diet had opposing effect of high body temperatures. Hence, supplementation with FSF is one of the nutritional strategies to combat heat stress in animals. The FSF as a mineral feed additive helps the systems to delay oxidation by inhibiting the formation of free radicals or by interrupting propagation of free radicals by several mechanisms (Brewer, 2011). Thus supplementation of heat-stressed animals with FSF is required to correct their negative mineral imbalances, because heat stress induces a significant decrease in the dry matter intake (Marai et al., 2008). The results of the current study on ADWI and ADFI further justify the fact that mineral constituents found in FSF improve insulin sensitivity because good insulin response is a major factor to successful adoption of heat load. Therefore, supplementation of diet with FSF was an effective strategy to improve the likelihood of rams to survive heat load.

4.7.2 Physiological response

Physiological responses are regarded as first parameters to check whether the animal is coping with any adverse environmental temperatures (Marai et al., 2007). Many researchers (Otoikhian et al., 2009; Phulia et al., 2010; Sejian et al., 2014) have conducted research on physiological adaptive mechanisms such as RT, PR and RR. In the current study, there was a significant difference ($P < 0.01$) among physiological parameter measured (ST, RT, RR and PR) across all treatment groups, as presented in Table 4.3. Among the parameters, RR and RT have been used as a reliable heat stress indicators in sheep (Daramola and Adeloye, 2009; Marai et al., 2002; Sejian et al., 2010). The outsets of thermal stress in animals are best indicated by increase in rectal temperature and respiration rate (Al-Haidary, 2004; Marai et al., 2007).

This positive relationship between the RR and RT indicates the importance of respiratory mechanism for thermolysis and maintenance of homeothermia in the animals, to avoid an increase in body temperature (Gomes and Starling, 2003). The significant decrease in RT and PR as inclusion levels FSF increased in the present study indicates that the heat stress was mitigated by the inclusion of FSF in experimental diets tested. The decrease in RR and RT in rams supplemented with varying levels of FSF may be due to a favorable effect of these antioxidants on body thermoregulation (Webb *et al.*, 2010).

However, the observed significant decrease in PR as the levels of FSF increase across the treatment groups is possibly because of FSF supplement as it typically gives an adaptive mechanism of reducing a ram's body temperature. Many researchers support these results as Singh and Hooda, (2011) found a decrease in PR in the group supplemented with yeast as mineral additive in buffalo calves. Wrinkle *et al.* (2012) also found a reduction in RR and PR supplementation with rumen-protected niacin. The reduced ST, RT, RR, and PR in rams supplemented with FSF shows an improvement in thermo tolerance and better performance. The highest mean values of ST, RT, RR, and PR were noted in rams subjected to 0g FSF/kg (control group). However, the lowest values for ST, RT, RR, and PR across the rams supplemented with the FSF possibly suggest that FSF can be used as supplement on mitigating heat stress.

4.7.3. Blood parameters

Varying inclusion levels of FSF influenced some blood biochemical parameters (TPP, MCV and MCHC) whereas most of the other parameters (Hb, PCV, GLU, RBC and WBC) were not influenced by inclusion level ($P > 0.05$). Both Hb and PCV are regarded as reliable indicators of heat stress in livestock (McManus *et al.*, 2009). The findings that both Hb and PCV were not affected by diet inclusion of FSF indicate that the FSF induced anti-heat stress effects. Similarly to the results of the present study, Abdel-Samee,(1991), indicated an increase in TPP in goats

subjected to 0gFSF/kg, 20g FSF/kg and 40g FSF/kg on day 45 while Srikandakumar *et al.* (2003) reported similar results in sheep. This may explain the fact that FSF was able to decrease animal body temperatures, as it was noted by Purwar *et al.* (2017) that feed additive improves thermotolerance of animals.

The results obtained in this study are similar Silva and Starling (2003) who investigated cutaneous and respiratory evaporation in wool sheep. The results showed an increase in MCV in rams fed 20g FSF/kg and 40g FSF/kg on day 45 and day 90. This could suggest that FSF was able to mitigate heat stress in Dohne Merino rams. Also, the higher blood flow can increase cellular circulation (Ei-Nouty, 1990). However, in this present study, the values of TPP and MCV increased. This could be explained by the fact that FSF has the opposing effect on hemodilution because of its antioxidant content. It is good to note also, the silica, which is a major component of FSF, and the long period of feeding could be responsible to the results obtained on hematological parameters in this study

4.8 Conclusion and recommendations

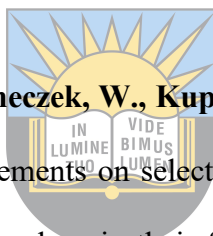
The findings from the current study showed that FSF supplementation was able to adjust physiological and biochemical response of Dohne Merino rams, as the best results were recorded in rams fed 40g FSF/kg. Moreover, the study also indicated that rectal temperature and respiration rate are important parameters that can be used by poor-resource farmers for heat tolerance assessment in sheep. Therefore, fossil shell flower can be used as a supplement to rams up to 40g FSF/kg to mitigate heat stress.

4.9 References

- Abdel-Samee, A.M.** 1991. Detection of heat adaptability of growing lambs in subtropics. *Zagazig Veterinary Journal*, **19**:719–731.
- Al-Dawood, A.** 2015. Adoption of agricultural innovations: Investigating current status and barriers to adoption of heat stress management in small ruminants in Jordan. *American-Eurasian Journal of Agricultural & Environmental Sciences*, **15**: 388–398.

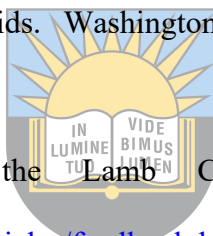
- Al-Haidary, A.A.** 2004. Physiological responses of Naimey sheep to heat stress challenge under semi-arid environments. *International Journal of Agriculture and Biology*, **2**: 307-309.
- Al-Haidary, A.A., Aljumaah R.S., Alshaikh, M.A., Abdoun, K.A., Samara, E.M., Okah, A.B., Aluraiji, M.M.** 2012. Thermoregulatory and physiological responses of Najdi sheep exposed to environmental heat load prevailing in Saudi Arabia. *Pakistan Veterinary Journal*, **32**: 515–519.
- Altan, O., altan, A.L.I., Çabuk, M. and Bayraktar, H.** 2000. Effects of heat stress on some blood parameters in broilers. *Turkish Journal of Veterinary and Animal Sciences*, **24**(2): 145-148.
- Banerjee, D., Upadhyay, R.C., Chaudhary, U.B., Kumar, R., Singh, S., Polley, S., Mukherjee, A., Das, T.K. and De, S.** 2014. Seasonal variation in expression pattern of genes under HSP70. *Cell Stress and Chaperones*, **19**(3): 401-408.
- Brewer, M. S.** 2011. Natural antioxidants: sources, compounds, mechanisms of action, and potential applications. *Comprehensive Review in Food Science and Food Safety*, **10**: 221–247.
- Caroprese, M.** 2008. Sheep housing and welfare. *Small ruminant research*, **76**(1-2): 21-25.
- Cwynar, P., Kolacz, R. and Czerski, A.** 2014. Effect of heat stress on physiological parameters and blood composition in Polish Merino rams. *Berl Munch Tierarztl Wochenschr*, **127**(5-6): 177-82.
- Daramola, J.O. and Adeloye, A.A.** 2009. Physiological adaptation to the humid tropics with special reference to the West African Dwarf (WAD) goat. *Tropical Animal Health and Production*, **41**(7): 1005-1016.
- De Rensis, F. and Scaramuzzi, R.J.** 2003. Heat stress and seasonal effects on reproduction in the dairy cow—a review. *Theriogenology*, **60**(6): 1139-1151.
- El Tazi, S.M., Mukhtar, M.A., Mohamed, K.A. and Tabidi, M.H.** 2014. Effect of using black pepper as natural feed additive on performance and carcass quality of broiler chicks. *International Journal of Pharmaceutical Research & Analysis*, **4**:108-113.

- El-Nouty, F.D., Al-Haidary, A.A. and Salah, M.S.** 1990. Seasonal variations in hematological values of high-and average-yielding holstein cattle in semi-arid environment. *Journal of King Saud University*, **2**(2): 173-182.
- Gomes, R., and Starling, J.M.C.** 2003. Evaporação cutânea e respiratória em ovinos sob altas temperaturas ambientes. *Revista Brasileira de Zootecnia*, **2**: 1956-1961.
- Gupta, M., Kumar, S., Dangi, S.S. and Jangir, B.L.** 2013. Physiological, biochemical and molecular responses to thermal stress in goats. *International Journal of Livestock Research*, **3**(2): 27-38.
- Ikusika, O.O. Mpendulo, C.T. Zindove, T.J. and Okoh, A.I.** 2019. Fossil Shell Flour in Livestock Production: A Review. *Animals*, **9**(3): 70-73
- Kiyatkin, E.A.** 2005. Brain hyperthermia as physiological and pathological phenomena. *Brain research reviews*, **50**(1): 27-56.
- Korczynski, M., Rojkowski, A., Janeczek, W., Kupczynski, R., Pogoda-Sewerniak, K.** 2004. The influence of microclimatic elements on selected indicators of physical thermoregulation and basic physiological factors in calves in their first weeks of life. *Science Books of Rural Academy of Wroclaw*, **501**: 113–118.
- Koster, H.** 2013. Diatomite in animal feed. <http://www.Agrisilica.co.za/pdf/eng/diatom>. Accessed 18 May 2019.
- Marai, I.F.M., El-Darawany, A.A., Fadiel, A. and Abdel-Hafez, M.A.M.** 2007. Physiological traits as affected by heat stress in sheep a review. *Small ruminant research*, **71**(1-3): 1-12.
- Marai, I. F. M., El- Darawany, A. A., Fadiel, A., Abdel-Hafez, M. A. M.** 2008. Reproductive performance traits as affected by heat stress and its alleviation in sheep. *Tropical and Subtropical Agroecosystems*, **8**: 209–234.
- Marai, I.F.M., El-Darawany, A.A., Fadiel, A. and Abdel-Hafez, M.A.M.** 2009. Physiological traits as affected by heat stress in sheep—A review, *Small Ruminant Research*, **71**: 1–12



University of Fort Hare
Together in Excellence

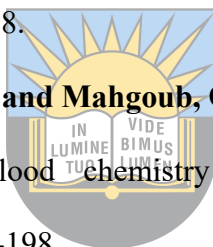
- Marai, I.F.M., Habeeb, A.M. and Gad, A.E.** 2004. Reproductive traits of female rabbits as affected by heat stress and lighting regime under subtropical conditions of Egypt. *Animal Science*, **78**(1): 119-127.
- McManus, C., Paludo, G.R., Louvandini, H., Gugel, R., Sasaki, L.C.B. and Paiva, S.R.** 2009. Heat tolerance in Brazilian sheep: physiological and blood parameters. *Tropical Animal Health and Production*, **41**(1): 95-101.
- Minka, N.S. and Ayo, J.O.** 2009. Physiological responses of food animals to road transportation stress. *African Journal of Biotechnology*, **8**(25): 7415-7427.
- Mucina, L. and Rutherford, M.C.** 2006. *The vegetation of South Africa, Lesotho and Swaziland*. South African National Biodiversity Institute.
- National Research Council.** 2007. Nutrient requirements of small ruminants: sheep, goats, cervids, and new world camelids. Washington, DC: The National Academies Press. <https://doi.org/10.17226/11654>.
- Neary, M.** 1998. "Feeding the Lamb Crop," Purdue University Extension, <http://ag.ansc.purdue.edu/sheep/articles/feedlamb.html>
- Olayemi, O.D., Jegede, O.C., Obudu, E. and Obeta, S.S.** 2015. Effect of dry and wet seasons on some haematological parameters in Sokoto Redgoat and Balami sheep. *Vom Journal of Veterinary Science*, **10**: 96-103.
- Osita, C.O., Ani, A.O., Ezema, C., Oyeagu, C.E., Uzochukwu, I.E. and Ezemagu, I.E.** 2019. Hematological and Biochemical Indices of West African Dwarf Sheep Fed Diets Containing Yeast (*Saccharomyces cerevisiae*), Grass, Grass/Legume (50: 50) and Legume. *Pakistan Journal of Nutrition*, **18**(1): 34-41.
- Otoikhian, C.S.O., Orheruata, J.A., Imasuen, J.A. and Akporhwarho, O.P.** 2009. Physiological response of local (West African Dwarf) and adapted Switzerland (White Bornu) goat breed to varied climatic conditions in South-South Nigeria. *African Journal of Geneneral Agriculture*, **5**(1): 1-6.



University of Fort Hare
Together in Excellence

- Pambu-Gollah, R., Cronje, P.B. and Casey, N.H.** 2000. An evaluation of the use of blood metabolite concentrations as indicators of nutritional status in free-ranging indigenous goats. *South African Journal of Animal Science*, **30**(2): 115-120.
- Phulia, S.K., Upadhyay, R.C., Jindal, S.K. and Misra, R.P.** 2010. Alteration in surface body temperature and physiological responses in Sirohi goats during day time in summer season. *Indian Journal of Animal Sciences*, **80**(4): 340-342.
- Purwar, V., Oberoi, P.S. and Dang, S.A.** 2017. Effect of feed supplement and additives on stress mitigation in Karan Fries heifers. *Veterinary World*, EISSN: 2231-0916
- Rana, M.S., Hashem, M.A., Sakib, M.N. and Kumar, A.** 2014. Effect of heat stress on blood parameters in indigenous sheep. *Journal of the Bangladesh Agricultural University*, **12**(4): 91-94.
- Sejian, V., Kumar, D., Gaughan, J.B. and Naqvi, S.M.** 2017. Effect of multiple environmental stressors on the adaptive capability of Malpura rams based on physiological responses in a semi-arid tropical environment. *Journal of Veterinary Behavior*, **17**: 6-13.
- Sejian, V., Maurya, V.P., Naqvi, S.M.K., Kumar, D. and Joshi, A.** 2010. Effect of induced body condition score differences on physiological response, productive and reproductive performance of Malpura ewes kept in a hot, semi-arid environment. *Journal of Animal Physiology and Animal Nutrition*, **94**(2): 154-161.
- Sejian, V., Singh, A.K., Sahoo, A. and Naqvi, S.M.K.** 2014. Effect of mineral mixture and antioxidant supplementation on growth, reproductive performance and adaptive capability of Malpura ewes subjected to heat stress. *Journal of animal physiology and animal nutrition*, **98**(1): 72-83.
- Silanikove, N.** 2000. Effects of heat stress on the welfare of extensively managed domestic ruminants. *Livestock production science*, **67**(1-2): 1-18.
- Silanikove, N. and Koluman, N.** 2015. Impact of climate change on the dairy industry in temperate zones: predications on the overall negative impact and on the positive role of dairy goats in adaptation to earth warming. *Small Ruminant Research*, **123**(1): 27-34.

- Silva, R.G. and Starling, J.M.C.** 2003. Evaporação Cutânea e Respiratória em ovinos sob altas temperaturas ambientes, *Revista Brasileira Zootecnia*, **32**: 1956–196.
- Singh, S.P., Hooda, O.K. and Kumar P.** 2011. Effect of yeast supplementation on feed intake and thermal stress mitigation in buffaloes. *Indian Journal of Animal Science* (India), **81**(9): 961-964.
- Singh, O. and Hooda, S.P.** 2011. Effect of yeast supplementation on feed intake and thermal stress mitigation in buffaloes. *Indian Journal of Animal Sciences*. **32**: 1956 – 1960
- Skapetas, B., Kalaitzidou, M.** 2017. Current status and perspectives of sector in the world. *Livestock Research for Rural Development*, **29**(2): 17 -19.
- Srikandakumar, A., Johnson, E.H. and Mahgoub, O.** 2003. Effect of heat stress on respiratory rate, rectal temperature and blood chemistry in Omani and Australian Merino sheep. *Small Ruminant Research*, **49**(2): 193-198.
- Srikandakumar, A., Johnson, E.H. and Mahgoub, O.** 2003. Effect of heat stress on respiratory rate, rectal temperature and blood chemistry in Omani and Australian Merino Sheep. *Small Ruminant Research*, **49**:193-198.
- Starling, J.M.C., Silva, R.G., Cerón-Muñoz, M., Barbosa, G.S. S.C. and Costa, M.J.R.P.** 2002. Analysis of some physiological variables for the evaluation of the degree of adaptation in sheep submitted to heat stress, *Revista Brasileira de Zootecnia*, **32**: 2070–2077.
- Stella, A.V., Paratte, R., Valnegri, L., Cigalino, G., Soncini, G., Chevaux, E., Dell’Otro, V. and Savoini, G.** 2007. Effect of administration of live *Saccharomyces cerevisiae* on milk production, milk composition, blood metabolites and faecal flora in early lactating dairy goats. *Small Ruminant Research*, **67**: 7-13.
- Verschuren, O., Ketelaar, M., Takken, T., Helders, P.J. and Gorter, J.W.** 2008. Exercise programs for children with cerebral palsy: a systematic review of the literature. *American Journal of Physical Medicine and Rehabilitation*, **87**: 404–417.



University of Fort Hare
Together in Excellence

- Webb, E.C., Van Niekerk, W.A., Lee, K. and Marais, W.J.** 2010. Reproductive performance of semi-intensively kept Döhne Merino ewes fed with different protein supplements. *South African Journal of Animal Science*, **40**(5): 451-454.
- Wrinkle, S.R., Robinson, P.H. and Garrett, J.E.** 2012. Niacin delivery to the intestinal absorptive site impacts heat stress and productivity responses of high producing dairy cows during hot conditions. *Animal Feed Science and Technology*, **175**(1): 33-47.



University of Fort Hare
Together in Excellence

CHAPTER 5: General discussion, conclusions and recommendations

5.1 General discussion

It can never be overemphasized as the way sheep are so important, since they play very important roles to the poor resource farmer (Saico and Abul, 2007). However, there are many production constraints that affect sheep production which include poor nutrition, diseases and parasites. Moreover, among the above-mentioned problems affecting sheep production high ambient temperature is one of the major factors that seriously affect sheep performance (Al-Haidary, 2004). Nevertheless, high ambient temperature does not always have a direct effect on sheep performance. For instance, long time exposure of pasture to high ambient temperature may possibly lead to reduced forage quality and that will consequently lead to poor growth, increased disease risks and reproductive failure in sheep.

Thamo *et al* (2017) noted that focusing on applying strategies such provision of shades and sprinkling animals does not necessarily help since getting correction is the only way out and therefore other mitigation approaches are required. However, high ambient temperatures directly affect the increasing body temperature and the respiration rate. Hence, the increased body temperature is linked with alterations in endocrine functions, redistribution in blood flow and decrease in feed intake which has a negative influence in the productive and reproductive performance of the sheep (Abouheif and Alsobayel, 1982, 1983; Eltawill and Narendran, 1990).

The current study was developed to investigate whether supplementation using FSF would influence feed acceptability and heat tolerance response of Dohne Merino rams. First the study was conducted to determine the effects of FSF on feed acceptability in Dohne Merino rams to understand the threshold at which rams can accept feed without any detrimental effect on their feed preference and feed intake. Also, the varying inclusion levels of fossil shell flour were determined to check at which threshold FSF can be added to significantly mitigate heat stress on rams without compromising their productivity. The broad objective of the current study was to investigate the effectiveness of varying levels of FSF as an additive supplement to improve sheep productivity.

In Chapter 3, the outcome of the current study showed that the FSF has the potential to improve feed acceptability of Dohne Merino rams. The diets with 40g FSF/kg and 60g FSF/kg inclusion levels of FSF showed better acceptability compared to other treatments. The reason for better acceptability in diets with 40g FSF/kg and 60g FSF/kg inclusion levels of FSF could be because fossil shell flour has minerals constituents that increases the palatability through aroma flavor in diets supplemented with 40g FSF/kg and 60g FSF/kg; hence rams showed better preferences.

In Chapter 4, the mean values of all physiological parameters skin temperature (ST), respiration rate (RR), pulse rate (PR), rectal temperature (RT), decreased with an increase in inclusion levels of fossil shell flour. The inclusion levels of FSF affected average daily water intake (ADFI) and average daily water intake (ADWI), showing high water intake and feed intake in rams subjected to 40g/kg of fossil shell flour (FSF) followed by 60g FSF/kg, 20g FSF/kg and 0g FSF/kg. Amongst all biochemical parameters tested only total plasma protein (TPP), mean corpuscular hemoglobin concentration (MCHC) and mean corpuscular volume (MCV) were influenced by fossil shell flour. The TPP increased as the inclusion levels of FSF increased. The MCHC was low in rams subjected to 60g FSF/kg and in 0 to 40g FSF/kg TPP increases as levels of FSF increases. It can be concluded that water and feed intake increase with increasing levels of fossil shell flour while the physiological parameters decline as levels of fossil shell flour increase. Therefore, this means that fossil shell flour can be used as a supplement to rams up to 40g FSF/kg to mitigate heat stress.

5.2. Conclusion

From the results obtained in the current study it can be concluded that;

The positive response in terms of improving feed acceptability of Dohne Merino rams indicated that with only FSF inclusion levels of 40g FSF/kg and 60g FSF/kg optimum results are obtained when fed to Dohne Merino rams. Therefore, this means that fossil shell flour can improve growth performance of Dohne Merino rams.

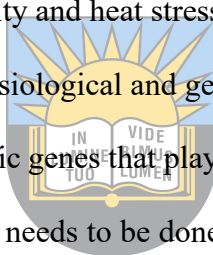
The inclusion level of 40g/kg of FSF in the diet of Dohne Merino rams was able to adjust the physiological and biochemical response of Dohne Merino rams.

Fossil shell flour can be used up to 40g FSF/kg as a supplement of Dohne Merino rams to mitigate heat stress.

Improvement was noted on feed intake, physiological and biological parameters of Dohne Merino rams fed with 40g/kg FSF.

5.3. Recommendations

The use of other common sheep breeds, other than Dohne Merino rams used in the current study, need attention as this broadens the scope of using FSF as a mineral supplement to help add information regarding feed acceptability and heat stress tolerance. Sheep respond to thermal stress in many ways such as behavioral, physiological and genetic based. Furthermore, there is a need to investigate candidate genes and specific genes that play a pivotal role in protecting animals against heat stress. However, further research needs to be done regarding adaptation of sheep to Southern African environmental condition.



University of Fort Hare
Together in Excellence

For good achievement in the organic animal feeding practices considering levels of FSF (40g FSF/kg to 60g FSF/kg) in the diet of Dohne Merino rams is advised.

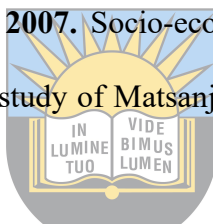
The feed intake and time spent on feeding clearly show that over the successive feeding trial there was an inclination towards a preference of rams to eat diet supplemented with 40g FSF/kg over the other treatments.

To adjust physiological and biochemical response of Dohne Merino rams during thermal stress, the inclusion level of 40g FSF/kg is recommended.

For future purposes, further research should be carried out on the reproductive performance of Dohne Merino rams supplemented varying levels of fossil shell flour to determine its effect on the sperm production and quality.

5.4. References

- Abouheif, M.A. and Alsobayel, A.A. 1982.** Reproductive performance of Saudi Arabian black Najdi ewes under the local environment in Riyadh area. *World Review of Animal Production*, 18: 9.
- Abouheif, M.A. and Alsobayel, A.A. 1983.** Environmental and genetic factors influencing birth weight of black Najdi lambs. *World Review of Animal Production*, 19: 51.
- Al-Haidary, A.A. 2004.** Physiological responses of Naimey sheep to heat stress challenge under semi-arid environments. *International Journal of Agriculture and Biology*, 2: 307-309.
- Eltawill, A. and Narendran, R. 1990.** Ewe productivity in four breed of sheep in Saudi Arabia. *World Review of Animal Production*, 25: 93.
- Saico, S.S. and Abul, S.A.L.A.M. 2007.** Socio-economic constraints on goat farming in the lowveld of Swaziland-A case study of Matsanjeni. *Journal of sustainable development in Africa*, 9(3).
- Thamo, T., Addai, D., Pannell, D.J., Robertson, M.J., Thomas, D.T. and Young, J.M. 2017.** Climate change impacts and farm-level adaptation: Economic analysis of a mixed cropping–livestock system. *Agricultural systems*, 150: 99-108.



University of Fort Hare
Together in Excellence

Appendix 1: Ethical clearance certificate



University of Fort Hare
Together in Excellence

ETHICAL CLEARANCE CERTIFICATE AREC-150311-008

Certificate Reference Number: MPE011SMWA01/19/A

Project title: **Fossil shell flour supplementation in the diet of ram: Effect on acceptability, performance, heat stress tolerance, ruminal fatty acid composition and microbial population.**

Nature of Project MSc in Animal Science

Principal Researcher: Lwazi Mwanda

Supervisor: Dr C.T. Mpendulo

On behalf of the University of Fort Hare's Animal Research Ethics Committee (AREC) I hereby give ethical approval in respect of the undertakings contained in the above-mentioned project and research instrument(s). Should any other instruments be used, these require separate authorization. The Researcher may therefore commence with the research as from the date of this certificate, using the reference number indicated above.

Special conditions:

- 1) With respect to the fistulation procedure, can we please get a weekly update on the health of the eight animals, which includes pictures of the fistulated area. Could we please also get a report from the veterinarian that is responsible for the fistulation at the half way stage and at the completion of this component of the project. This report needs to provide assurances that the animals welfare is being considered and that the animals are still healthy. Please also be aware that dependent on available time, members of the committee are likely to come and inspect the animals during the course of the study.



University of Fort Hare
Together in Excellence

Please note that the AREC must be informed immediately of

- Any material change in the conditions or undertakings mentioned in the document;
- Any material breaches of ethical undertakings or events that impact upon the ethical conduct of the research.

The Principal Researcher must report to the AREC in the prescribed format, where applicable, annually, and at the end of the project, in respect of ethical compliance. The AREC retains the right to

- Withdraw or amend this Ethical Clearance Certificate if
 - Any unethical principal or practices are revealed or suspected;
 - Relevant information has been withheld or misrepresented;
 - Regulatory changes of whatsoever nature so require;
 - The conditions contained in the Certificate have not been adhered to.
- Request access to any information or data at any time during the course or after completion of the project.
- In addition to the need to comply with the highest level of ethical conduct principle investigators must report back annually as an evaluation and monitoring mechanism on the progress being made by the research. Such a report must be sent to the Dean of Research's office.

The Animal Research Ethics Committee wishes you well in your research.

Yours sincerely



Dr. Craig Tambling
AREC Chairperson

07 April 2019



University of Fort Hare
Together in Excellence

Appendix 2: List of papers submitted

Manuscript 1

Animal Production Science - Manuscript AN19589



Animal Production Science <onbehalf@manuscriptcentral.com>

Wed 2019/10/09, 9:53 PM

Mwanda, Lwazi; Mpendulo, Thando Conference; IKUSIKA, OLUSEGUN OYEBADE; Tikwayo, Sizwe Edward; Yawa, Mandla; Gajana, Sabelo

Reply all

10-Oct-2019

Dear Mr Mwanda

Your manuscript entitled 'Feed preference of Dohne Merino rams fed varying levels of fossil shell flour' has been successfully submitted online and will be given full consideration for publication in Animal Production Science.

Your manuscript ID is AN19589. Please mention this manuscript ID in all future correspondence or when calling the office for questions.

If there are any changes in your street address or email address, please log in to ScholarOne Manuscripts at <https://mc.manuscriptcentral.com/csiro-an> and edit your details as appropriate. You can also view the status of your manuscript at any time by checking your Author Centre after logging in.

Thank you for submitting your manuscript to Animal Production Science.

Sincerely,

Editorial Office, Animal Production Science

Manuscript 2

Your manuscript LIVSCI_2019_1172 has been sent for review



Livestock Science <Evisesupport@elsevier.com>

Mon 2019/09/23, 11:11 AM

Mwanda, Lwazi

Reply all

This message was sent automatically.

Reference: LIVSCI_2019_1172

Title: Effects of fossil shell flour supplementation on heat tolerance of Dohne Merino rams

Journal: Livestock Science

Dear Mr. Mwanda,

I am currently identifying and contacting reviewers who are acknowledged experts in the field. Since peer review is a voluntary service it can take time to find reviewers who are both qualified and available. While reviewers are being contacted, the status of your manuscript will appear in EVISE® as 'Reviewer Invited'.

Once a reviewer agrees to review your manuscript, the status will change to 'Under Review'. When I have received the required number of expert reviews, the status will change to 'Read for Decision' while I evaluate the reviews before making a decision on your manuscript.

To track the status of your manuscript, please log into EVISE® and go to 'My Submissions' via: http://www.evisesupport.com/evise/faces/pages/navigation/NavController.jspx?_afRNLAACR=LIVSCI

Kind regards,

Livestock Science

Have questions or need assistance?

For further assistance, please visit our [Customer Support](#) site. Here you can search for solutions on a range of topics, find answers to frequently asked questions, and learn more about EVISE® via interactive tutorials. You can also talk 24/5 to our customer support team by phone and 24/7 by live chat and email.

Copyright © 2018 Elsevier B.V. | [Privacy Policy](#)

Elsevier B.V., Radarweg 29, 1043 NX Amsterdam, The Netherlands, Reg. No. 33156677.

P.O Box 2083
Butterworth
4960

To: Whom it May Concern.

**CONFIRMATION LETTER OF LANGUAGE EDITING FOR
DISSERTATION**

This letter serves to inform and confirm that the dissertation of Mr.
Mwanda Lwazi

Buthelezi was submitted for Language Editing and duly edited by
Mr Silusapho Nyanda, ID number 8905096002086.

The dissertation which was submitted in fulfilment of the
requirements for the Master of Science (Agriculture) degree in Animal
Science in the Department of Livestock and Pasture Science Faculty of
Science and Agriculture at the University of Fort Hare

The editor of this Dissertaion, Mr. Nyanda, is qualified to edit as he is an
experienced journalist and government communicator. With more than



University of Fort Hare
Together in Excellence

six year in the media and communication field Mr Nyanda is currently a
Assitant Director: Writer for Vuk'uzenze

For further queries you are welcome to contact Mr Nyanda on 067 876
0455 or via email on silusapho@gmail.com.

Regards,

Silusapho Nyanda



University of Fort Hare
Together in Excellence



University of Fort Hare
Together in Excellence