

**EFFECTS OF FEEDING SPROUTED SORGHUM (*SORGHUM BICOLOR*) DIETS FORTIFIED  
WITH EXOGENOUS ENZYMES ON EGG PRODUCTION OF RED AND WHITE AMBERLINK  
LAYERS**

**By**

**Nkhumbuleni Remember Muavha**

**Student number: 11630015**

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

**Department of Animal Science  
Faculty of Science, Engineering & Agriculture  
University of Venda  
Thohoyandou  
South Africa  
2023**

**Supervisor: Dr. F. Fushai**

**Co-Supervisor: Dr. A.J. N. Netshipale**

## DECLARATION

I, Nkhumbuleni Remember Muavha student number 11630015, hereby declare that this dissertation for the Master of Science in Agriculture (MSCANS) as submitted to the Department of Animal Sciences, Faculty of Science, Engineering & Agriculture, at the University of Venda, has not been previously submitted for any degree at this or another university. It is original in design and in execution, and all reference material contained therein has been duly acknowledged.

Signature ...  ..... Date .....24/04/2023.....

## **ACKNOWLEDGMENT**

I want to acknowledge my supervisor, Dr. F. Fushai for his guidance and constant support. I want to acknowledge support by my co-supervisor, Dr. A.J. N. Netshipale. I hereby also express my deepest appreciation and to Mr W.M. Lubusi, Mr L.T. Mashaba, Mr O. Chabalala, Ms M. Ramukanda, Ms Q. Rambau and Ms N. Mariba for assistance during data collection, interpretation and feed formulation. I am equally appreciative to Mr M.E. Nyathi for his support with laboratory analyses and procurement. To the Publication Committee Fund (PCF), Blind S.A and National Research Foundation (NRF), I am greatly appreciative of the financial support for this research. Last, but not least, I appreciate the University of Venda Disability Unit (DSU), for their support to us students living with disabilities.

## ABSTRACT

The objective of the study was to investigate the effects of feeding sprouted sorghum (*Sorghum bicolor*) diets fortified with exogenous enzymes on egg production by Red and White Amberlink layers. Egg production by 216 Red and White Amberlink layers which were 16 weeks into production was evaluated over a six-week period. The layers were housed in a naturally ventilated battery house, placed in forty-eight 45 cm length × 45 cm width × 42 cm height cages, each stocked at 3 birds/cage. The birds were allocated to treatments in a randomised 3 (diets) × 2 (enzymes) × 2 (strain) factorial arrangement replicated six times. Experimental diets were Meadow Powerlay Late Lay (Product V16418) as a positive control (PC), and iso-nutrient (70 g/kg Crude Fibre, 130 g/kg CP, 5 g/kg Lysine), sprouted, and raw (negative control (NC) sorghum-soybean layer diets. A duplicate mix of each diet was fortified with 500 g/tonne of a custom multi-enzyme cocktail (xylanase Endo-1, 4-Beta-Xylanase (EC-3.2.1.8), 2440 U/kg, Endo-1, 3 (4)-Beta-Glucanase (EC-3.2.1.6), 304 U/kg, and 6-phytase (IUB 3.1.3.26), 1220 U/kg). Layers on the raw sorghum diet had low ( $P < 0.05$ ) feed intake. Interaction ( $p = 0.0038$ ) of the layer strain, diet and enzyme occurred for the laying rate. Highest ( $P < 0.05$ ) laying rate was attained when the Red Amberlink layers were on the commercial diet with enzymes, and when White Amberlink layers were on the same diet without enzymes, similar ( $P > 0.05$ ) to when both strains were on the enzyme supplemented, sprouted sorghum diet. Lower ( $P < 0.05$ ) laying rates were observed when both strains were on the raw sorghum, without ( $P > 0.05$ ) enzyme effect compared to other treatments. Laying rate in layers on the sprouted sorghum diet was not different ( $P > 0.05$ ) from the commercial diet, and significantly higher ( $P < 0.05$ ) raw sorghum-based diet with or without enzyme fortification. The net effect of treatments on laying rate was in the dietary order commercial feed > sprouted sorghum > raw sorghum ( $P < 0.05$ ). The Red Amberlink strain laid larger ( $P < 0.05$ ) eggs than the White strain. Expressed on both egg number and egg weight basis, the FCR were in the dietary order commercial feed > sprouted sorghum > raw sorghum ( $P < 0.01$ ). Strain\*enzyme interaction occurred for egg weight ( $P < 0.05$ ), whereby the enzymes reduced ( $P < 0.05$ ) egg weight in Red Amberlink layers when on the sprouted sorghum diet, which was quantitatively similar on all other treatments, except for opposite, quantitative enzyme effect on White Amberlink layers on the commercial diet. Though inferior to the commercial diet, the comparative egg production and FCR largely supported replacement of the commercial, with sorghum diets, more so when the sorghum is sprouted. Treatment interaction on egg production and size suggested both beneficial and deleterious enzyme action, likely the effects of unique dietary chemical matrices.

**Keywords:** Amberlink layers, exogenous enzymes, sprouted sorghum

## TABLE OF CONTENTS

<b>DECLARATION.....</b>	<b>i</b>
<b>ACKNOWLEDGMENT .....</b>	<b>ii</b>
<b>ABSTRACT .....</b>	<b>iii</b>
<b>Table of contents .....</b>	<b>iv</b>
<b>LIST OF TABLES.....</b>	<b>vi</b>
<b>LIST OF ABBREVIATIONS.....</b>	<b>vii</b>
<b>Chapter 1: INTRODUCTION.....</b>	<b>1</b>
1.1 Background.....	1
1.2 Statement of research problem .....	2
1.3 Justification .....	3
1.4 Main objective .....	3
1.5 Specific objectives.....	3
1.6 Research hypothesis.....	4
<b>Chapter 2: LITERATURE REVIEW.....</b>	<b>5</b>
2.1 Introduction .....	5
2.2 Origins and current production of Sorghum .....	5
2.3 Uses of sorghum .....	5
2.4 Nutritive value of sorghum.....	5
2.5 Anti-nutritional factors in sorghum .....	6
2.5.1 Phytate .....	6
2.5.2 Cyanogenic glycosides and tannins .....	6
2.6 Nutritional benefit of sprouting sorghum for layer diets .....	6
2.7 Effects of exogenous enzymes on layer performance .....	7
2.8 Summary of the literature review .....	7

<b>Chapter 3: RESEARCH METHODOLOGY .....</b>	<b>8</b>
3.1 Experimental conditions .....	8
3.2 Methods and materials .....	8
3.3 Sorghum preparation.....	8
3.4 Enzymes .....	8
3.5 Chemical analyses .....	8
3.6 Formulation and preparation of experimental diets .....	9
3.7 Housing, experimental birds, design, and management .....	10
3.8 Measurements .....	10
3.9 Ethical considerations.....	10
3.10 Statistical Analysis.....	11
<b>Chapter 4: RESULTS .....</b>	<b>12</b>
<b>Chapter 5: DISCUSSION, CONCLUSION &amp; RECOMMENDATIONS .....</b>	<b>15</b>
5.1 Discussion.....	15
5.2 Conclusion.....	16
5.3 Recommendations.....	17
<b>Chapter 6: References.....</b>	<b>18</b>

## LIST OF TABLES

Table 3.1: Ingredient composition (% as is) of sorghum diets .....	9
Table 3.2: Analysed ingredient and calculated dietary chemical composition. ....	10
Table 4.1: Effects of feeding sprouted sorghum diets fortified with exogenous enzymes on egg production of Red and White Amberlink layers.....	13

## LIST OF ABBREVIATIONS

ADF	Acid Detergent Fibre
AOAC	Association of Official Analytical Chemists
Ca	Calcium
CF	Crude Fibre
CP	Crude Protein
DM	Dry Matter
EE	Ether Extract (Crude Fat)
EW	Egg Weight
FCR	Feed Conversion Ratio
FI	Feed Intake
G	gram
Kg	Kilogram
M	meter
ME	Metabolisable energy
NDF	Neutral Detergent Fibre
NPN	Non-Protein Nitrogen
NRF	National Research Foundation
NSPs	Non-Starch Polysaccharides
P	Phosphorus
PCF	Publication Committee Fund
RS	Raw Sorghum
SAHDC	School of Agriculture Higher Degree Committee
SS	Sprouted Sorghum
UHDC	University Higher Degree Committee



## CHAPTER 1: INTRODUCTION

### 1.1 Background

The cost of the conventional poultry feedstuffs continues to escalate, with the feeding cost typically contributing up to 75% of the total cost of production (Alagawany *et al.*, 2018). In addition to feed cost, enterprise viability is also largely dependent on the dietary efficacy, which demands high precision feeding of least cost formulated diets. The nutrient balance of the conventional maize-soybean poultry diet is primarily anchored on both superior high nutritional quality and complementarity of these basal ingredients compared to the competing alternative energy and protein rich feedstuffs. Therefore, substitution of these basal feeds with the alternative feedstuffs risks potentially nutritionally inferior diets, which therefore requires careful ingredient selection, and effective processing.

Due to the high cost of dietary protein, much of current research in alternative low-cost poultry feedstuffs is biased towards the protein-rich legume grains. However, being the quantitative major component in the poultry diet, declining, erratic global maize production is also strongly driving the cost of stock feed (Mabelebele *et al.*, 2015). In sub-Saharan Africa, over the past few decades, genetic improvement for high yields and superior quality, and strong extension messaging favoured maize production at the expense of the more climate resilient traditional cereals (Mwadalu and Mwang, 2013). Given limited irrigation capacity, economic commercial dry land maize production has long been largely limited to areas which receive reliable, high rainfall (Travis *et al.*, 2006). However, globally, climate-change is shifting both the spatial and temporal rainfall distribution, effectively diminishing ecosystems previously suitable for maize production (Ayanlade *et al.*, 2018). Without large scale irrigation development, maize production in most arid areas will therefore significantly decline, disrupting the global trade (Ringler *et al.*, 2011). At the same time, the demand from an increasing human population is a strong push on the costs of maize (Linden, 2017).

In tropical developing regions, the consensus strategy to mitigate declining maize production is to promote the climate-resilient small grains (Muzerengi and Tirivangasi, 2019). A suitable candidate is sorghum (*Sorghum bicolor*) (Sedghi *et al.*, 2011). Compared to the millets, preference for sorghum is a trade-off based on the relative nutritive values as maize substitutes in poultry diets (Nyamambi *et al.*, 2007), edaphic and climatic adaptability (Sedghi *et al.*, 2011) and yield, in relation to the production cost (Dicko *et al.*, 2006). While competitive in the relative production cost, and containing beneficially bioactive simple phenolic acids, flavonoids, sorghum also contains high levels of soluble non-starch polysaccharides and condensed tannins (Dicko *et al.*, 2006). In the arid tropics, sorghum is considered a suitable dietary energy substitute for maize (Sedghi *et al.*, 2011), given better edaphic

and climatic adaptability (Sedghi *et al.*, 2011), comparable nutritive value (13.7 MJ ME kg<sup>-1</sup>, 9.5% CP versus 13.9 MJ ME kg<sup>-1</sup>, 10.1% CP). Whole sorghum protein is dominated (70%) by kafirins, which are relatively poorly digested proteins due to a high degree of polymerization, extensive disulfide bridges, and strong interaction with condensed tannins, resistant starches (Belton *et al.*, 2006) and phytate (Liu *et al.*, 2015). These antinutritive attributes may affect sorghum dietary efficacy in terms of both energy and protein utilisation (Liu *et al.*, 2015), and demand concerted investigation to identify the critical poultry (e.g. genotype, age, productive state) – sorghum (e.g. genotype, growth conditions) interactive factors within specific production environments.

Given the antinutrients, effective sorghum processing is critical to mitigate its nutritional limitations. In small-holder systems, germination is considered practical, cost effective, biologically efficient bioprocessing (Inyang and Zakari, 2008; Correia *et al.*, 2008). Subject to correct matching of the chemical matrix, optimized enzyme dosage, and depending on the type, age and strain of the birds (Cowieson *et al.*, 2006), exogenous enzymes might further enhance the dietary efficacy. While previous studies (Pasquali *et al.*, 2017; Gidago *et al.*, 2020) suggest multi-enzyme efficacy in raw sorghum-based broiler diets, enzyme supplements to either raw or germinated sorghum did not benefit broiler chick performance (Torki *et al.*, 2007).

Therefore, this study investigated the effects on Red and White Amberlink layer performance of sprouting sorghum and fortifying sorghum-based diets with a custom cocktail of xylanase, glucanase and phytase enzymes.

## 1.2 Statement of research problem

Climate change is negatively affecting maize production, resulting in the shortage of cheap, quality feed for small-scale farming sectors. Thus, farmers need to adapt by using low-cost raw materials to produce animal feeds. Sorghum can replace maize in poultry feed because of its high soluble carbohydrate content (Sauer *et al.*, 1978) and if the effects of tannin is reduced by improving its nutritive value (Nyachoti *et al.*, 1997). Maize and sorghum are more genetically related compared to wheat and barley (Ciacci *et al.*, 2007). Sorghum can tolerate drought better than maize. However, like other cereal grains, sorghum contains anti-nutrients such as phytate, complex carbohydrates, tannins and trypsin inhibitor which reduce nutritive value. Conversely, exogenous enzymes are essential in the reduction of anti-nutritional factors. However, there is limited research on the effects of sprouting sorghum and on the effects of supplementary exogenous enzymes for efficient feeding to different layer strains, which might respond differently to such novel diets. Therefore, the study investigated

egg production by different layer strains on a sprouted sorghum diet supplemented with exogenous enzymes.

### 1.3 Justification

The South African human population is increasing, driving the demand for animal by-products, while feeds are getting expensive, and the price of protein and energy sources is also escalating. Therefore, only the more competitive commercial farmers can afford to buy commercial feeds. Thus, small-scale farmers must find locally available alternative cereal grains such as sorghum to enhance their competitive advantage, with need for low-cost practical processing to remove anti-nutritional factors. Sorghum grows well in the Southern African warm climate. Sorghum grow well in many South African Provinces but with the Free State Province being the main producer (DAFF, 2010). Sorghum is produced globally and yearly on 41.31 million hectares (ha) of land with a total production of 59.83 million tons (USDA, 2019). In East Africa Uganda is the fourth leading producer following countries such as South Sudan, Tanzania and Ethiopia (FAOSTAT, 2018).

Sprouting is practical, low cost, effective method for processing grains which can be readily adopted by farmers on low resource settings. Germination metabolism during sprouting of the sorghum grain breaks down complex molecules into simpler forms, thereby detoxifying anti-nutrients and reducing secondary compounds to avail nutrients for efficient assimilation. Exogenous enzymes catalyse the breakdown of anti-nutritional factors found in sorghum grains such as tannins, to improve egg production.

### 1.4 Main objective

The main objective of the study is to determine the effects of sprouting, and of exogenous enzymes on the nutritive value of sorghum (*Sorghum bicolor*) as a low-cost substitute for maize in the commercial layer diet.

### 1.5 Specific objectives

To determine the effects on egg production of:

- i) Feeding a sprouted sorghum diet.
- ii) Fortifying sorghum diets with a custom multi-enzyme cocktail (xylanase Endo-1,4-Beta-Xylanase (EC-3.2.1.8), 2440 U/kg, Endo-1,3(4)-Beta-Glucanase (EC-3.2.1.6), 304 U/kg, and 6-phytase (IUB 3.1.3.26), 1220 U/kg) AXTRA® XAP enzymes.
- iii) Layer strain (Red, White Amberlink) when fed raw versus sprouted sorghum-based, enzyme fortified diets.

## 1.6 Research hypothesis

- i) Feeding a sprouted sorghum diet does not affect egg production.
- ii) Fortifying sprouted sorghum-based diets with exogenous enzymes does not influence egg production.
- iii) The layer strain (Red or White Amberlink) does not affect egg production when fed on enzyme fortified sorghum-based diets.

## CHAPTER 2: LITERATURE REVIEW

### 2.1 Introduction

The viability and development of the poultry industry largely depends on the quality of feed and prices (Adenjimi *et al.*, 2011). However, in developing countries, stock feeds are expensive and not readily available. Increasing demand and cost of maize justify exploration of alternative ingredients for poultry diets (Mehri *et al.*, 2009), with application of modern technologies (Oluka, 2000). The chemical composition, and consequently, the nutritive value of cereal grains is affected by factors such as climate, type of soil and genotype (Ebadi *et al.*, 2005). Sorghum (*Sorghum bicolor*) is poor in lysine, threonine, and tryptophan, but rich in leucine, proline and glutamic acid (Duodu *et al.*, 2003). Germination removes the anti-nutritional factors and improves nutrient availability from cereal and legume grains (Maidala, 2015). Exogenous enzymes can further improve nutrient digestion (Enwere, 1998).

### 2.2 Origins and current production of Sorghum

Sorghum falls in the grass family Poaceae (Kimber, 2000). Sorghum can be classified depending on the starch content, structure, and functional properties (Sang *et al.*, 2008). Globally, sorghum is the fifth most important crop after wheat, rice, maize, and barley (Bryden *et al.*, 2009).

### 2.3 Uses of sorghum

In Africa, the Middle East, Asia, and Central America sorghum is the traditional staple diet (CGIAR, 2009). Sorghum is genetically more related to maize when compared to wheat and barley (Ciacci *et al.*, 2007), which makes it the best nutritional substitute for expensive maize. It is used as human food and animal feed, such that its global production and trade are increasing due to increasing demand (FAO, 1995). Sorghum can also be used to produce ethanol biofuel (ICRISAT, 2009).

### 2.4 Nutritive value of sorghum

In poultry nutrition, energy is vital for the provision of body heat, maintenance, growth and production (Inaku *et al.*, 2011). Sorghum contains high energy (13.7 MJ ME/kg) and moderate protein (9.5% CP), which is comparable with the 13.9 MJ ME/kg and 10.1% CP content of maize (Nyamambi *et al.*, 2007). However, the chemical composition is highly variable. Rajashekher *et al.*, (2003) reported 12% CP, 75% starch, 4% fat and 4% minerals. Ensminger and Olentine (1978) reported 8.9 – 15%CP, crude 2.8% fat (ether extract), 1.5 – 1.7% ash, 2.1 – 2.3% crude fibre (CF). Starch is a primary carbohydrate in sorghum grains, though highly variable (32 to 79%), followed by the soluble sugars

and non-starch polysaccharides (Waniska and Rooney, 2002). The important fibrous carbohydrate components are cellulose, hemicellulose, lignin, and pectin (Waniska and Rooney, 2002). The protein value of sorghum is close to that of maize, though with less lysine, methionine, and threonine (Sikka and Johari, 1979).

## **2.5 Anti-nutritional factors in sorghum**

### **2.5.1 Phytate**

Phytate is found in all plant matter, and accounts for about two-thirds of total phosphorus in plant-based diets (Maenz, 2001). It is an anti-nutritional factor, because it binds minerals and makes them deficient to the animal (Oatway *et al.*, 2001). High dietary calcium in poultry diets induces significant reduction in the digestibility of phytate phosphorus (Tamim *et al.*, 2004). Since laying hens need large quantities of dietary calcium for constant egg production, there is research interest on the effect of phytase in poultry (Snow *et al.*, 2004). Exogenous phytases release phosphorus by destruction of the plant phytate, thereby removing the adverse effects of phytate on digestion and absorption of other minerals, amino acids, and energy (Selle and Ravindran, 2007).

### **2.5.2 Cyanogenic glycosides and tannins**

According to Traore *et al.* (2004), dried, malted red sorghum contains cyanogenenic glycosides and other anti-nutrients. The largest cyanogen found in sorghum is dhurrin (Rooney and Waniska, 2000). Cyanogen can be reduced through sprouting (Shayo *et al.*, 2001). Depending on the cultivar, sorghum contains variable condensed tannins and polyphenolic compounds that cause an unpleasant taste and reduce intake, digestibility, growth, and feed efficiency in livestock (Rooney and Waniska, 2000). Tannins decrease the digestibility and utilization of absorbed nutrients (Rooney and Waniska, 2000). Hassan *et al.* (2003) reported that high tannin sorghum reduces weight gain and feed intake for broiler chicks. Although decortication of sorghum grain can significantly reduce the tannin content (Rooney and Waniska, 2000), residual tannins still depressed growth rate, reduced feed intake, feed efficiency, nutrient digestibility, and compromised bone improvement (Nyachoti *et al.*, 1997).

## **2.6 Nutritional benefit of sprouting sorghum for layer diets**

Several processing methods are commonly used to improve the nutritive value of cereal grains. Processing methods such as soaking, boiling, microwave cooking and autoclaving decrease the tannin content of cereals (Gee and Harold, 2004). Shayo *et al.* (2001) observed that sprouting improved nutritional and functional quality of sorghum better than other processing methods. However, leaching of soluble components can occur during soaking in water for sprouting, while

enzyme activities during sprouting cause significant chemical changes to nutrients (Traore' *et al.*, 2004). Sprouting improved the protein content in low tannin sorghum (Hamid, 2001). Soaking and sprouting improved protein digestibility (Laetitia *et al.*, 2005). Fafiolu *et al.* (2006) reported an increase in body weight at levels up to 30% sorghum sprouts in the layer rations. Ebadi *et al.* (2005) reported significant increase in feed take when maize was substituted with sorghum grain in the layers' diet.

High dietary levels of up to 30% sorghum sprout did not affect yolk and albumin weight and Haugh units (Fafiolu *et al.*, 2006). Similarly, 25% replacement of maize with sorghum did not change the Haugh unit (Ebadi *et al.*, 2005), with increased yolk index and shell weight and improved yolk colour. Ebadi *et al.* (2005) reported decrease in shell thickness with sorghum replacement of maize.

## **2.7 Effects of exogenous enzymes on layer performance**

Exogenous enzymes are widely used in poultry feeding (Schedle *et al.*, 2012). Overall, there is limited, largely inconsistent information on the benefit of exogenous enzymes in layers. However, subject to correct matching of the chemical matrix, optimized enzyme dosage, and depending on the type, age and strain of the birds (Cowieson *et al.*, 2006), exogenous enzymes enhance dietary efficacy. While previous studies (Pasquali *et al.*, 2017; Gidago *et al.*, 2020) suggest multi-enzyme efficacy in raw sorghum-based broiler diets, enzyme supplements to either raw or germinated sorghum did not benefit broiler chick performance (Torki *et al.*, 2007). Exogenous proteinases improved broiler performance (Hajati, 2010). Phytases reduced phytic acid, releasing bound proteins and minerals (Shah *et al.*, 2011; Nourmohammadi *et al.*, 2012).

## **2.8 Summary of the literature review**

Literature review suggested unprocessed sorghum contains significant anti-nutritional factors which bind nutrients, making them deficient for birds, which necessitates processing prior to feeding to chickens. There are several methods of processing sorghum, but, in poor resource settings, sprouting is considered superior and more practical to other bio-processing methods. Previous studies suggested sprouted sorghum is a good source of protein, energy and other nutrients for laying birds, sufficient to maintain constant egg production. Subject to correct match to the dietary chemical matrix, fortifying sorghum based layer diets with cocktails of proteinase, phytase, amylase and complex carbohydrase enzymes potentially improves nutrient extraction and layer performance.

## CHAPTER 3: RESEARCH METHODOLOGY

### 3.1 Experimental conditions

The study was conducted at the University of Venda, School of Agriculture Experimental Farm. The location is on latitude 22°58'32" South, longitude 30°26'45" East, at altitude of 596 m above sea level, with maximum and minimum temperatures of 31°C and 18°C (Tadross *et al.*, 2005). Feed and diet samples were analyzed in the Animal Science Nutrition Laboratory. It was conducted over 6 weeks from the 1st week of February to week 3 of March 2020.

### 3.2 Methods and materials

This study evaluated the effects of sprouted sorghum as a substitute for maize in diets for different layer strains, and the effects of fortification of the sorghum-based diets with a cocktail of fibrolytic, protease and phytate degrading enzymes.

### 3.3 Sorghum preparation

Bulk red sorghum (*Sorghum bicolor*) grain was manually cleaned of dirt and screened for viable seeds before division into two equal lots. Sprouts of one lot were prepared by 30-minute initial grain soaking in 2% sodium hypochlorite solution, 12- hour soaking of washed grain in tap water, 5-day open-air (21 - 33°C) sprouting under manual irrigation, which was terminated by 5-day sun-drying of single layer grain spread on black plastic sheets [refuse plastic bags, size 750×950mm] on an open concrete slab. Representative dried samples were collected for chemical analyses.

### 3.4 Enzymes

A custom multi-enzyme cocktail which contained xylanase Endo-1,4-Beta-Xylanase (EC-3.2.1.8) 2440 U/kg, Endo-1,3(4)-Beta-Glucanase (EC-3.2.1.6) 304 U/kg, 6-phytase (IUB 3.1.3.26) 1220 U/kg was supplemented at inclusion rate at 500g/tonne.

### 3.5 Chemical analyses

The dry matter content was determined according to the AOAC (1990; method 930.15). Ash contents were determined by drying the sample at 550°C overnight (AOAC, 1990; method 942.05). The Nitrogen (N) content were evaluated using a Kjeldahl procedure (AOAC, 1990; method 984.13). Fat content was determined by soxhlet fat extraction (AOAC, 1990; method 930.15).



### 3.6 Formulation and preparation of experimental diets

Experimental diets equally met the minimum feeding standards for layer diets (National Research Council (NRC), 1994). A commercial diet (Meadow Feeds (Pty) Ltd, Delmas, South Africa Powerlay Late Lay diet, Product V16418) served as a control. Test diets were constituted using dehulled soybean (*Glycine max*), sprouted and raw sorghum (**Table 3.1**) which were first hammer-milled (Jacobson hammer mill, model P160 Teordrop 10HP, China) through a 3mm screen before 20-minute mixing (MORHLANG VERTA MIX, 1200VM, USA) in half-tonne lots along with micro ingredients into complete diets (**Table 3.2**). Each diet had a duplicate fortified with exogenous enzymes.

**Table 3.1: Ingredient composition (% as is) of sorghum diets**

Ingredients	Diets	
	Raw sorghum	<sup>1</sup> Sprouted sorghum
Soybean cake	12.13	13.23
Raw sorghum	72.35	-
<sup>1</sup> Sprouted sorghum	-	77.62
<sup>2</sup> Layer macro pack	2.88	3.11
Lysine	0.09	0.09
Limestone	10.37	11.62
Mono Dicalcium Phosphate	1.73	1.04
Sand	0.46	0.49
Total	100.00	100.00

<sup>1</sup>30-minute sterilisation in 2% aqueous sodium hypochlorite, 12 hours soaking in tap water, 5-day open-air sprouting, sun-dried.

<sup>2</sup>Composition per kg of premix; Vitamin A - 588235.300 UI, Vitamin D3 - 205882.400 UI, Vitamin E - 882.353 UI, Vitamin K-88.235 mg, Vitamin B - 117.647 mg, Vitamin B2 - 235.294 mg, Vitamin B6 -147.059 mg, Niacin -1647.059 mg, Calcium Pantothenate- 411.765 mg, Biotin - 1470.588 µg, Folic acid - 29.412 mg, Vitamin B12 - 1176.471 µg, Choline Chloride - 15278.820 mg, 6-Phytase -17647.060 FTU, Limestone (as carrier) - 32.353 g, Salt -235.294 g, Mono-dicalcium phosphate - 470.588g, Cobalt from cobalt sulphate - 29.412 mg, Copper from Copper sulphate - 352.941 mg, Iron from Ferrous sulphate -1764.706 mg Iodine (Potassium iodide) - 58.824 mg, Manganese (Manganese sulphate) - 4117.647 mg, Zinc (Zinc sulphate) - 1764.706 mg, Selenium (Sodium selenite) - 8.824 mg, Lysine -88.824 g, Methionine -109.412 g, Enzyme (Natuphos E 10000 G) - 1.765 g.

**Table 3.2: Analysed ingredient and calculated dietary chemical composition.**

	DM	Fat	ADF	NDF	Crude Protein	Ca	P
<i>Ingredients (g kg<sup>-1</sup> DM)</i>							
Raw Sorghum	923.1	31.5	97.0	307.9	98.4	0.2	0.3
<sup>1</sup> Sprouted Sorghum	916.7	28.3	118.5	244.9	142.6	0.3	0.41
Soybean Cake	940.3	22.2	153.7	297.4	485.2	2.5	0.64
<i>Diets (g kg<sup>-1</sup> DM)</i>							
Raw Sorghum	934.0	25.4	258.0	88.6	131.2	45.1	5.1
<sup>1</sup> Sprouted Sorghum	931.1	23.0	211.6	103.6	130.4	45.1	4.9

<sup>1</sup>30-minute sterilisation in 2% aqueous sodium hypochlorite, 12 hours soaking in tap water, 5-day open-air sprouting, sun-dried.

### 3.7 Housing, experimental birds, design, and management

Egg production was evaluated in a trial running over 42 days using 216 Red and White Amberlink layers which were at 16 weeks into production. The layers were housed in a naturally ventilated battery house, placed in forty-eight 45 cm length × 45 cm width × 42 cm height cages stocked at 3 birds /cage in a balanced, completely randomised 3 (diets) X 2 (enzymes) X 2 (strain) factorial with 6 replicates per treatment. Each pen was equipped with one nipple drinker and a width-long tube feeder. The trial was conducted under unrestricted feeding, automated 16:8 light: dark hour lighting regime. The layers received the recommended vaccination regimes for Newcastle, Infectious Bronchitis and Infectious Bursal Disease.

### 3.8 Measurements

Weekly cage feed intake, egg numbers and egg weights were recorded, and the feed conversion ratio calculated per dozen, and per kilogram eggs. Fresh eggs were collected, weighed, and counted per cage daily at 09:00. Egg production was calculated as both the laying rate (hen-day egg production (%)) and the egg output (laying rate x average egg weight (g/bird/day))

### 3.9 Ethical considerations

Ethical approval was acquired from the Ethics Committee of the University of Venda [Clearance Certificate SARDF/19/ANS/04/1305]. The housing specifications and the feeding and watering spaces complied with the statutory standards for the rearing of laying birds in battery systems.

### 3.10 Statistical Analysis

Egg production parameters were analyzed using the GLM procedure of Minitab software version 18 (2017) using the model:

$$Y_{ijkl} = \mu + \alpha_i + \beta_j + F_k + W_l + (\alpha\beta)_{ij} + (\alpha F)_{ik} + (\alpha W)_{il} + (\beta F)_{jk} + (\beta W)_{jl} + (FW)_{kl} + (\alpha\beta F)_{ijk} + (\alpha\beta FW)_{ijkl} + \varepsilon_{ijkl}$$

Where,  $Y_{ijkl}$  is the observed parameter value,  $\mu$  the overall mean,  $\alpha_i$  the effect of the  $i^{\text{th}}$  diet ( $i = 1, 2, 3$ ),  $\beta_j$  the effect of the  $j^{\text{th}}$  enzyme ( $j = 1, 2$ ),  $F_k$  the effect of  $k^{\text{th}}$  strain ( $k = 1, 2$ ),  $W_l$  the effect of the  $l^{\text{th}}$  week ( $l = 1, 2, 3, 4, 5, 6$ ),  $(\alpha\beta)_{ij}$ ,  $(\alpha F)_{ik}$ ,  $(\alpha W)_{il}$ ,  $(\beta F)_{jk}$ ,  $(\alpha\beta F)_{ijk}$ ,  $(\alpha\beta FW)_{ijkl}$  the respective diet, enzyme, strain and week interactions, and  $\varepsilon_{ijkl}$ , the residual error. Different treatment means were compared using the Tukey's test at  $p < 0.05$

## CHAPTER 4: RESULTS

Layer performance parameters are presented in Table 4. Layers on the raw sorghum diet had low ( $P < 0.05$ ) intake. All the measured egg production and quality parameters varied with the week of measurement ( $P < 0.001$ ). Highest ( $P < 0.05$ ) laying rate was attained when the Red Amberlink layers were on the commercial diet with enzymes, and when White Amberlink layers were on the same diet without enzymes, similar ( $P > 0.05$ ) to when both strains were on the enzyme supplemented, sprouted sorghum diet. Low ( $P < 0.05$ ) laying rates were observed when both strains were on the raw sorghum, without ( $P > 0.05$ ) enzyme effect. Laying rate was intermediate ( $P < 0.05$ ) in layers on the sprouted sorghum diet, without ( $P > 0.05$ ) enzyme effect. The net effect was laying rate in the dietary order commercial feed  $>$  sprouted sorghum  $>$  raw sorghum ( $P < 0.05$ ). The Red Amberlink strain laid larger ( $P < 0.05$ ) eggs than the White strain. Expressed on both egg number and egg weight basis, the FCR were in the dietary order commercial feed  $>$  sprouted sorghum  $>$  raw sorghum ( $P < 0.01$ ). Strain\* enzyme interaction occurred for egg weight ( $P < 0.05$ ), whereby the enzymes reduced ( $P < 0.05$ ) egg weight in Red Amberlink layers when on the sprouted sorghum diet, which was quantitatively similar on all other treatments, except for opposite, quantitative enzyme effect on White Amberlink layers on the commercial diet.

**Table 4.1: Effects of feeding sprouted sorghum diets fortified with exogenous enzymes on egg production of Red and White Amberlink layers**

Treatments			Intake	Laying Rate	Egg weight	Egg Output	FCR	FCR	
			(g/b/d)	%	(g)	(g /b/d)	(kg/doz)	(kg/kg)	
Strain(S)	Diet (D)	Enzyme (E)							
Red	Commercial	+	114.4	90.7 <sup>a</sup>	60.4	54.66	1.53	2.11	
		-	115.0	87.5 <sup>ab</sup>	61.6	53.78	1.60	2.16	
	<sup>1</sup> Sprouted Sorghum	+	114.4	84.9 <sup>abc</sup>	60.4	51.29	1.65	2.27	
		-	114.3	80.8 <sup>cde</sup>	61.9	49.95	1.74	2.34	
		Raw Sorghum	+	112.7	76.0 <sup>ef</sup>	61.0	46.46	1.83	2.51
			-	112.5	77.8 <sup>def</sup>	61.7	47.93	1.79	2.42
White	Commercial	+	114.4	84.9 <sup>abc</sup>	60.3	51.12	1.67	2.28	
		-	114.1	89.1 <sup>a</sup>	59.6	52.96	1.57	2.19	
	<sup>1</sup> Sprouted Sorghum	+	114.2	86.1 <sup>abc</sup>	59.9	51.55	1.63	2.26	
		-	113.9	82.7 <sup>bcd</sup>	60.3	49.83	1.69	2.34	
		Raw Sorghum	+	112.9	77.4 <sup>def</sup>	60.6	46.82	1.79	2.46
			-	112.7	72.2 <sup>f</sup>	60.8	43.80	1.93	2.64
SEM		4.93	11.13	2.446	6.577	0.291	0.394		
Strain (S)									
Red			113.86	82.94	61.15 <sup>a</sup>	50.68	1.69	2.30	
White			113.70	82.08	60.27 <sup>b</sup>	49.35	1.71	2.36	
SEM			4.914	12.232	0.246	7.141	0.308	0.413	
Diet (D)									
Commercial			114.46 <sup>a</sup>	88.05 <sup>a</sup>	60.45	53.13 <sup>a</sup>	1.59 <sup>c</sup>	2.18 <sup>c</sup>	
Sprouted Sorghum			114.18 <sup>a</sup>	83.63 <sup>b</sup>	60.64	50.66 <sup>b</sup>	1.68 <sup>b</sup>	2.30 <sup>b</sup>	
Raw Sorghum			112.70 <sup>b</sup>	75.84 <sup>c</sup>	61.04	46.25 <sup>c</sup>	1.83 <sup>a</sup>	2.51 <sup>a</sup>	
SEM			0.486	1.116	2.490	0.659	0.0291	0.0393	
Enzyme (E)									
+			113.81	83.33	60.45	50.32	1.68	2.31	
-			113.75	81.68	60.97	49.71	1.72	2.35	
SEM			4.915	12.211	2.484	7.165	0.307	0.414	
Week (W)									
1			112.03 <sup>c</sup>	85.12 <sup>ab</sup>	61.31 <sup>ab</sup>	52.04 <sup>a</sup>	1.61 <sup>b</sup>	2.19 <sup>c</sup>	
2			115.68 <sup>b</sup>	77.28 <sup>cd</sup>	61.56 <sup>a</sup>	47.51 <sup>bc</sup>	1.85 <sup>a</sup>	2.51 <sup>ab</sup>	
3			110.73 <sup>c</sup>	89.29 <sup>a</sup>	59.67 <sup>c</sup>	53.25 <sup>a</sup>	1.50 <sup>b</sup>	2.10 <sup>c</sup>	
4			117.72 <sup>a</sup>	80.65 <sup>bc</sup>	60.99 <sup>ab</sup>	49.09 <sup>b</sup>	1.79 <sup>a</sup>	2.44 <sup>b</sup>	
5			110.89 <sup>c</sup>	87.60 <sup>a</sup>	60.38 <sup>bc</sup>	52.80 <sup>a</sup>	1.54 <sup>b</sup>	2.12 <sup>c</sup>	
6			115.63 <sup>b</sup>	75.10 <sup>d</sup>	60.35 <sup>bc</sup>	45.40 <sup>c</sup>	1.90 <sup>a</sup>	2.63 <sup>a</sup> <sub>w</sub>	
SEM			4.140	11.133	2.430	6.597	0.268	0.363	
P Values									
S			0.776	0.551	0.002	0.115	0.551	0.224	
D			0.027	0.001	0.244	0.001	0.001	0.001	
E			0.910	0.252	0.074	0.472	0.264	0.488	
W			0.001	0.001	0.001	0.001	0.001	0.001	
S x D			0.838	0.314	0.775	0.345	0.442	0.495	
S x E			0.728	0.861	0.026	0.601	0.965	0.564	
S x W			0.928	0.954	0.134	0.892	0.881	0.843	
D x E			0.944	0.310	0.507	0.479	0.504	0.659	
D x W			0.961	0.089	0.001	0.069	0.297	0.216	
W x E			0.969	0.946	0.224	0.838	0.966	0.905	
S x D x E			0.925	0.038	0.484	0.098	0.068	0.106	
S x D x W			0.993	0.486	0.001	0.930	0.545	0.902	
D x W x E			0.996	0.774	0.396	0.827	0.947	0.942	
W x S x E			0.955	0.107	0.128	0.028	0.199	0.054	
W x S x D x E			0.942	0.010	0.043	0.010	0.035	0.051	

abcdef Means in the same column that are not sharing a common superscript are significantly different ( $P < 0.05$ ).

SEM- standard error of the means.

<sup>1</sup>30-minute sterilisation in 2% aqueous sodium hypochlorite, 12 hours soaking in tap water, 5-day open-air sprouting, sun-dried.

## CHAPTER 5: DISCUSSION, CONCLUSION & RECOMMENDATIONS

### 5.1 Discussion

The study investigated the effects of sprouting sorghum, and of dietary fortification with exogenous enzymes on the egg production of Red, compared to White Amberlink layers. Several factors influence the productivity of laying hens including the environment (Xu *et al.*, 2022) and genetics (Liu *et al.*, 2019). The experimental setup was typical of environmentally uncontrolled small-scale production. According to the breeder (Dekalb Poultry) specifications, compared to the White strain the Red Amberlink is larger (1950 vs 1725 g), consumes more feed (112 vs 108 g/day), with higher FCR (kg/kg) (2.12 vs 2.03), larger egg size (60.0 g vs 62.1 g) and more hen-housed egg production (479 vs 486) per cycle. In his study, overall egg production parameters approximated the strain standards. Feed intake is an important factor for low performance of layers (Li *et al.*, 2011). Given large variation in sorghum antinutrients such as the tannins, the effects on laying performance depend on the variety, feeding level, processing method, and the poultry genotype (Singh *et al.*, 2003). In this study, unlike the sprouted sorghum diet, the raw sorghum diet depressed feed intake. Previous studies reported low feed intake in chickens fed sorghum-based diets (Manu-Barfo *et al.*, 2013; Nortey *et al.*, 2013). Agunbiade *et al.*, (2016) similarly reported higher intake in maize-based diets compared to sorghum-based diets, with or without enzyme fortification.

In this study, all the measured egg quality parameters depended on the week of measurement, with some interaction with the main effects. The weekly variations were attributed to changes in nutrient requirements, and or ambient conditions during different weeks. Egg production was in the dietary order commercial feed >sprouted sorghum >raw sorghum, which suggested inferior sorghum diet efficacy, while confirming the benefit of sprouting sorghum. Sprouting is widely reported to increase the nutritive value of cereal grains (Muhammad *et al.*, 2013). In sorghum, the benefit is largely attributed to reduction of tannins, which increases intake, and the digestibility of protein, with enhanced bioavailability of amino acids (Baba *et al.*, 2012). In brown layers, Ochieng *et al.*, (2018) observed similar egg weights when maize was replaced by graded quantities of low tannin sorghum. Similarly, in White Leghorn hens, Abera *et al.* (2020) reported improved egg production and feed efficiency when raw, improved sorghum varieties replaced maize. In this study, significant interactions on egg production of the layer strain with the dietary factors (diet type, enzymes) were observed. The laying rate, and, quantitatively, the egg output was high when both the Red and White Amberlink strains layers were on the commercial diet regardless of supplementary enzymes, which suggested no enzyme benefit on the dietary matrix. Egg production on the enzyme-supplemented sprouted sorghum diet matched the commercial diet, which implied an additive benefit of sprouting sorghum, and of supplementary enzymes. Egg production was low on the raw sorghum, regardless of enzyme

fortification, confirming the inferior quality of the raw cowpea diet, and a mismatch to the supplementary enzymes, likely the effect of antinutrients. In this study, feeding layers raw or the sprouted sorghum diets did not affect egg size, much as sprouting of sorghum and enzymes did not benefit the egg size. Expressed on both the quantitative (egg number) and qualitative (egg weight) basis, the FCR were in similar dietary order commercial feed < sprouted sorghum < raw sorghum, which also confirmed the relative low efficacy of sorghum diet, and its improvement by sprouting the sorghum. Strain x diet x enzyme interaction occurred in similar pattern for the FCR when expressed both on the egg weight and egg number basis. Highest FCR was observed in White Amberlink layers on the raw sorghum diets and in Red Amberlink layers on the raw sorghum diet, regardless of the enzyme supplement. The least FCR were observed for the Red and White Amberlink layers on the commercial diet, followed by both strains sprouted sorghum diets, with intermediate values for the rest of other treatments. The strain and diet effects were regardless of the enzyme supplement. This trend of effects suggested less dietary efficacy in terms of the FCR for the sorghum diets, with benefit in sprouting sorghum, but no overall enzyme benefit on the efficiency of feed utilisation.

In this study, strain\* enzyme interaction was significant for egg weight. The enzymes reduced egg weight in Red Amberlink layers when on the sprouted sorghum diet, which was quantitatively similar on all other treatments, except for opposite, quantitative enzyme effect on White Amberlink layers on the commercial diet. Interactions on the laying rate and egg size resulted both beneficial and deleterious enzyme action, likely the effects of unique dietary chemical matrices. Across studies, the findings on enzyme benefit are inconsistent. Two previous studies (Pasquali *et al.*, 2017; Gidado *et al.*, 2020) suggested multi-enzyme efficacy in raw sorghum-based broiler diets. In contrast, enzyme supplements to either raw or germinated sorghum did not benefit broiler chick performance (Torki *et al.*, 2007). These contradictory findings confirm the need for correct matching of the chemical matrix, optimized enzyme dosages, and considerations for the type, age and strain of the birds (Cowieson *et al.*, 2006). In principle however, exogenous enzymes should increase the efficiency of sorghum digestion through breaking down of the anti-nutritional factors such as fibre, phytate and non-starch polysaccharide (NSP) (Adeola and Cowieson, 2011), to ensure consistent animal performance regardless of dietary chemical variations (Flores-Cervantes *et al.*, 2011; Lu *et al.*, 2013).

## 5.2 Conclusion

Though less efficient compared to the commercial diet, the comparative egg production and FCR largely supported replacement of the commercial, with sorghum diets, more so when the sorghum is sprouted. Treatment interactions on the laying rate and egg size were attributed to unique dietary chemical matrices.



### 5.3 Recommendations

Cost-benefit analyses is recommended to indicate the financial implications of substituting sprouted sorghum for maize in layer diets within different farming systems. Strain \* diet \* enzyme interactions on egg production justify research into diet tailored, more potent supplementary exogenous enzymes.

## CHAPTER 6: REFERENCES

- Abera, G., Urge, M., and Girma, M., 2020. Effects of replacing maize with different sorghum varieties on laying performance and egg quality of white Leghorn hens. *Livestock Research for Rural Development*, volume 32, article 170.
- Adejinmi, O.O., Hamzat R.A., Raji A.M and Owoyibo A.O (2011). Performance, nutrient digestibility and carcass characteristics of Broilers fed cocoa pod husks-based diets. *Nig. J. of Anim. Sci.* 13: 61-68.
- Adeola, O., and Cowieson, A.J., 2011. Opportunities and challenges in using exogenous enzymes to improve nonruminant animal production. *J. Anim. Sci.* 89:3189–218.
- Agunbiade, O.M., Ominisi, P.A., and Omaye, J.J., 2016. Performance of laying hens fed sorghum-based diets supplemented with enzymes. *Nigerian Journal of Animal Production*, 43 (2): 224-232.
- Alagawany, M., Elnesr, S.S., and Farag, M.R., 2018. The role of exogenous enzymes in promoting growth and improving nutrient digestibility in poultry. *Iranian journal of veterinary research*, 19(3), p.157.
- AOAC, 1990. *Official Methods of Analysis*. 15th Edition, Association of Official Analytical Chemists, Arlington.
- Ayanlade, A., Radeny, M., Morton, J.F., and Muchaba, T., 2018. "Rainfall variability and drought characteristics in two agro-climatic zones: An assessment of climate change challenges in Africa." *Science of the Total Environment*, vol. 630, pp. 728-737.
- Baba, G.M., Modu, S., Falmata, A.S., Hajjagana, L., and Ibrahim, Z., 2012. Evaluation of the nutritional value of sprouted sorghum fortified with sorghum and groundnut. *Scholarly Journal of Agricultural Science*, 2 (11): 292- 296.
- Belton, P.S, Delgadillo, I., Halford, N.G., and Shewry P.R., 2006. Kafirin structure and functionality. *Journal of Cereal Science*, 44 (3), 272–286.
- Bryden, W.L., Selle, P.H., Cadogan, D.J., Li, X., Muller, N.D., Jordan D.R., Gidley, M.J., and Hamilton, W.D., 2009. A review of the nutritive value of sorghum in broilers. *Rural Industry Res. Dev. Corp.* Kingston, Aust.
- CGIAR (Consultative Group on International Agricultural Research) (2009) website <http://www.cgiar.org/>.
- Ciacci, C., Maiuri, L., Caporaso, N., Bucci, C., Del Giudice, L., Massardo, DR., Pontieri, P., Di Fonzo, N., Bean, SR., Ioerger, B., and Londei, M., 2007. Celiac disease: in vitro and in vivo safety and palatability of wheat-free sorghum food products. *Clin Nutr*, 26: 799–805.
- Correia I., Nunes A., Barros A.S., and Delgadillo, I., 2008. Protein profile and malt activities during sorghum germination. *J Sci Food Agric.*; 88:2598–2605.

- Cowieson, A., Singh, D., and Adeola, O., 2006. Prediction of ingredient quality and the effect of a combination of xylanase, amylase, protease and phytase in the diets of broiler chicks 1 Growth performance and digestible nutrient intake. *British Poultry Science*, 47, 477–489.
- Department of Agriculture Forestry and Fisheries, 2010. Sorghum Production Guideline. Republic of South Africa. Department of Agriculture, Forestry and Fisheries.
- Dicko, M.H., Grupcage, H., Zouzouho, O.C., Traore, A.S., Van Berkel, W.J.H., and Voragen, A.G.J., 2006. Effect of germination on the activities of amylases and phenolic enzymes in sorghum varieties grouped according to food end – use properties. *Journal of the Science of Food and Agriculture*. 86:953-963.
- Duodu, K.G., Taylor, J.R.N., Belton, P.S., and Hamaker, B.R., 2003. Factors affecting protein digestibility. *Journal of Cereal Science* 38, 117-131.
- Ebadi, M. R., Pourreza, J., Jamalian, J., Edriss M.A., Samie A.H., and Mirhadi, S.A., 2005. Amino acid content and availability in low, medium and high tannin sorghum grain for poultry. *Int. Poult. Sci.* 4:27–31.
- Ensminger, M.E., Olentine Jnr, C.G., 1978. *Feeds and Nutrition – Abridged*. 1st Edition. The Ensminger Publishing Company, Covis, Carlifornia.
- Enwere, N.J., 1998. *Foods of Plant Origin: Processing and Utilization with Recipes and Technology Profiles*. Afro-Orbis Publications Ltd., Nsukka 301pp.
- Fafiolu, A.O., Oduguwa, O.O., Ikeobi, C.O.N., and Onwuka C.F.I., 2006. Utilization of malted sorghum sprout in the diet of rearing pullets and laying hens. *Archivos de Zootecnia*. 55: 361-371.
- FAO Corporate Document Repository (1995), “Chapter 6–Nutritional Inhibitors and Toxic Factors”, In: *Sorghum and Millets in Human Nutrition*, FAO Agriculture and Consumer Protection, available online at <http://193.43.36.103/DOCREP/T0818e/T0818E0j.htm>.
- FAO, 1995. *Sorghum and millet in human nutrition*. FAO. Food and nutrition series No. 27. Rome. Italy. P. 55-60.
- FAOSTAT, 2018. FAOSTAT statistical database. Rome: FAO (Food and Agriculture Organization of the United Nations).
- Flores-Cervantes, S., Casillas-Franco, J.C., Orozco-Hernandez, J.C., 2011. Effect of a multi-enzymatic mix in a sorghum-soybean meal-based ration on hen performance. *Italian Journal of Animal Science*, 10 (25): 131-133.
- Gee, M.C., and Harold, G.W., 2004. An assessment of the tannin content of wild sorghum. *Journal of Crop Science*, 43:1850 – 1870.
- Gidado, A.S., Oguntoye, M.A., and Akintunde, A.R., 2020. Evaluation of maize–sorghum mixture-based diets supplemented with exogenous enzyme on growth response, haematology and serum biochemical indices of starter broiler chickens. *Nigerian Journal of Animal Production*, 47(4), pp.158-166.

- Hajati H., 2010. Effects of Enzyme Supplementation on Performance, Carcass characteristics, Carcass Composition and Some Blood Parameters of Broiler Chicken. *American Journal of Animal and Veterinary Sciences*; 5(3):221-227.
- Hamid, F.H., 2001. The effects of germination and fermentation process on chemical composition and nutritional value of low – tannin grain sorghum. M. sc. Thesis, Faculty of Animal Production, University of Khartoum.
- Hassan, I.A., Elzubeir, E.A., and El Tinay, A.H., 2003. Growth and apparent absorption of minerals in broiler chicks fed diets with low or high tannin contents. *Trop. Anim. Health Prod.* 35:189-196.
- ICRISAT (International Crops Research Institute for the Semi-Arid Tropics) (2009), website <http://www.icrisat.org/>.
- Inaku, E.N., Bawa, G.S., Olugbemi, T.S., and Buba, W., 2011. Nutritive value of yam peel meal in broiler diet. In: Adeniji, A.A., Olatunji, E.A. and Gana, E.S. (Eds) *Value Reorientation in Animal Production: A Key to National Food Security and sustainable Economy*. Pp 481-483. Proceedings of 36th
- Inyang, C.U., and Zakari, U.M., 2008. Effect of germination and fermentation of pearl millet on proximate, chemical and sensory properties of instant “Fura”- a Nigerian cereal food. *Pakistan Journal of Nutrition*. 7 (1): 9-12.
- Kimber, C.T., 2000. Origins of domestication sorghum and its early diffusion to India and China. In: C.W. Smith, R.A. Fredericksen (Ed), *Sorghum Origin, History, Technology, & Production*. John Wiley & Sons inc, New York.
- Laetitia, M.M., Joseph, H.D., Joseph, D., and Christian, M., 2005. Physical, chemical and microbiological changes during natural fermentation of “gowe”, a sprouted or non-sprouted sorghum beverage from West Africa. *African Journal of Biotechnology* 4 (6): 467-496.
- Li, F., Xu, L.M., Shan, A.S., Hu, J.W., Zhang, Y.Y., and Li, Y.H., 2011. Effect of daily feed intake in laying period on laying performance, egg quality and egg composition of genetically fat and lean lines of chickens. *British Poultry Science*, 52 (2): 163-168.
- Linden, E. (2017, June). Remember the population bomb? It’s still ticking. *New York Times: Sunday Review*, 4. Google Scholar.
- Liu, S.Y., Fox, G., Khoddami, A., Neilson, K.A., Truong, H.H., Moss, A.F., and Selle, P.H., 2015. Grain sorghum: a conundrum for chicken-meat production. *Agriculture*, 5(4), pp.1224-1251.
- Liu, Z., Yang N., Yan, Y., Li, G., Liu, A., Wu, G., and Sun, C., 2019. Genome-wide association analysis of production performance in chickens across the whole laying period. *BMC Genetics* (2019) 20:67.
- Lu, H., Adedokun, S.A, Preynat A, Legrand-Defretin, V., Geraert P.A., Adeola, O., et al., 2013. Impact of exogenous carbohydrases and phytase on growth performance and nutrient digestibility in broilers. *Canadian Journal of Animal Science*; 93(2):243-249.

- Mabelebele, M., Siwela, M., Gous, R.M., and Iji, P.A., 2015. Chemical composition and nutritive value of South African sorghum varieties as feed for broiler chickens. *S. Afr. J. Anim. Sci.* 45, 207-213.
- Maenz, D.D., 2001. Enzymatic characteristics of phytases as they relate to their use in animal feeds. Pages 61-83 in *Enzymes in Farm Animal Nutrition*. M. R. Bedford, and G. G. Partridge, ed. CAB International, Wallingford, UK.
- Maidala, A., 2015. Nutritional evaluation of selected plant protein sources in the diets of broiler chickens. Unpublished Ph.D thesis. Animal production department. Abubakar Tafawa Balewa University, Bauchi
- Manu-Barfo, P.I., Nortey, T.N., Naazie, A.I., 2013. Effect of sorghum-barley brewer's scaget grain as a feed ingredient on broiler performance and carcass characteristics. *Bulletin of Animal Health and Production in Africa*, 16 (1): 67-74.
- Mehri, M., Pourreza, J., and Sadeghi, G., 2009. Replacing maize with pearl millet in laying hens' diets. *Trop ani. heal and pro.* 42 (3):439-444.
- Muhammad, S., Hussain, A., and Subhani, M., 2013. Use of sprouted grains in the diets of poultry and ruminants. *Indian Journal of Research*, 2 (10): 4-7.
- Muzerengi, T., and Tirivangasi, H.M., 2019. "Small grain production as an adaptive strategy to climate change in Mangwe District, Matabeleland South in Zimbabwe, Jambá." *Journal of Disaster Risk studies*, vol. 11, p. a652. Available: <https://doi.org/10.4102/jamba.v11i1.652>.
- Mwadalu, R., and Mwangi, M., 2013. "The potential role of sorghum in enhancing food security in Semi- Arid eastern Kenya: A review." *Journal of Applied Biosciences*, vol. 71, pp. 5786-5799.
- Nortey, T.N., Naazie, A., and Manu-Barfo, P., 2013. The effect of feeding sorghum-barley brewer's scaget grain (Sbbsg) on performance and egg quality of laying hens. *Bulletin of Animal Health and Production in Africa*, 16 (1):89-99.
- Nourmohammadi, R., Hosseini, S., Farhangfar, H., Bashtani, M., 2012. Effect of citric acid and microbial phytase enzyme on ileal digestibility of some nutrients in broiler chicks fed maize-soybean meal diets. *Italian Journal of Animal Science*; 11(1):36-40.
- NRC, 1994. *Nutrients requirements of poultry*. Ninth Revised Edition. National Academy Press. Washington, D.C.
- Nyachoti, C.M., Aktinson, J.L., and Leeson, S., 1997. Sorghum tannins: a review. *World's Poult. Sci. J.* 53:1-21.
- Nyamambi, B., Ndlovu, L.R., Naik, Y.S. and Kock, N.D., 2007. Intestinal growth and function of broiler chicks fed sorghum-based diets differing in condensed tannin levels. *S. Afr. J. Anim. Sci.* 37, 203-214.
- Oatway, L., Vasanthan, T., and Helm, J.H., 2001. Phytic acid. *Food. Rev. Int.* 17:419- 431.

- Ochieng, B.A., Owino, W.O., Kinyuru, J.N., Mburu, J.N., Gicheha, M.G., Kabuage, L., 2018. Effect of low tannin sorghum-based feed on physical and nutritional quality of layer chicken eggs. *Journal of Food Research*, 7 (4): 94-106.
- Oluka, S.I., 2000. Mechanical Characteristics of Local Maize Kernels in Quasi- Static Compressive Loading. *Journal of Science and Technology*. Vol 6: PP1-9.
- Pasquali, G.A. M., Fascina, V.B., Silva, A.L., Aoyagi, M.M., Muro, E.M., Serpa, P.G., Berto, D.A., Saldanha, E.S.P.B. and Sartori, J.R., 2017. Maize replacement with sorghum and a combination of protease, xylanase, and phytase on performance, nutrient utilization, litter moisture, and digestive organ size in broiler chicken. *Canadian Journal of Animal Science*, 97: 328 – 337.
- Rajashekher, A.R., Prasad, V.L.K. & Sudhakar, D., 2003. Sorghum and pearl millet for poultry feed. In: *Alternate uses of sorghum and pearl millet in Asia*. Common Fund for Commodities Technical Bulletin, 34. pp. 14-41. International Crops Research Ins.
- Ravindran, V., Ravindran, G., and Sivalogan, S., 1994. Total and phytate phosphorus contents of various foods and feedstuffs of plant origin. *Food Chem*. 50:133-136.
- Ringler, C., Zhu, T., Cai, X., Koo, J., and Wang, D., 2011. How African Agriculture Adapt to Climate Change can: *Climate Change Impacts on Food Security in Sub-Saharan Africa: Insights from Comprehensive Climate Change Modeling (No. 15 (20))*. International Food Policy Research Institute (IFPRI).
- Rooney, L.W., and Waniska, R.D., 2000. "Chapter 4.2 - Sorghum Food and Industrial Utilization", In: *Sorghum Origin, History, Technology, and Production*, Eds. C.W. Smith and R.A. Frederiksen. J. Wiley & Sons, New York, NY, pp 689-729.
- Sang, Y., Bean, S., Seib, P.A., Perderson, J., and Shi, Y.C., 2008. "Structure and Functional Properties of Sorghum Starches Differing in Amylose Content", *J. Agric. Food Chem*. Vol. 56, pp. 6680-6685.
- Sauer, D.B., Seitz, L.M., Burroughs, R., Mohr, H.E., West, J.L., Milleret, R.J., and Anthony, H.D., 1978. Toxicity of *Alternaria* metabolites found in weathered sorghum grain at harvest. *J. Agric. Food Chem*. 26:1380-1393.
- Schedle, K., Pecina, J., Punz, C., Mair, C., and Windisch, W., 2012. Inclusion of NSP-hydrolyzing enzymes in diets for grower-finisher pigs containing two levels of distillers dried grains with soluble. *Animal Review*; 18:129-134.
- Sedghi, M., Ebadi, M.R., Golian, A. & Ahmadi, H., 2011. Estimation and modelling true metabolizable energy of sorghum grain for poultry. *Poult. Sci*. 90, 1138-1143.
- Selle, P.H., and Ravindran, V., 2007. Microbial phytase in poultry nutrition. *Animal feed Science and Technology*, 135: 1–41.

- Shah, S.A., Zeb, A., Masood, T., Noreen, N., Abbas, S.J., Samiullah, M., Alim, M.A., and Muhammad, A., 2011. Effects of sprouting time on biochemical and nutritional qualities of mungbean varieties. *Afr J Agric Res*; 6(22):5091–5098.
- Shayo, N.B., Laswai, H.S., Tiisekwa, B.P.M., Nnko, S.A.M., Gidamis, A.B., and Njoki, P., 2001. Evaluation of nutritive value and functional qualities of sorghum subjected to different traditional processing methods. *Inter. J. Food Sc. Nutri.* 52: 117-126.
- Sikka, K.C., and Johari, R.P., 1979. Comparative nutritive value and amino acid content of different varieties of sorghum and effect of lysine fortification. *J. Agric Food Chem.* 27:962-965.
- Singh, S., Sai Prasad, S.V., and Katiyar, D.S., 2003. Genetic variability in the fodder yield, chemical composition and disappearance of nutrients in brown midrib and white midrib sorghum genotypes. *Asian-Aust. J. Anim. Sci.* 16:1303-1308.
- Snow, J.L., D.H. Baker, and C.M. Parsons. (2004). Phytase, citric acid, and 1  $\alpha$ -hydroxycholecalciferol improve phytate phosphorus utilization in chicks fed a cornsoybean meal diet. *Poult. Sci.* 83:1187-1192.
- Tadross, M., Jack, C., and Hewington, B., 2005. On RCM-based projections of change in Southern African summer climate. *Geophysical Research letters*, 32(10): 23713 -24460.
- Tamim, N.M., Angel, R., and Christman, M., 2004. Influence of dietary calcium and phytase on phytate phosphorus hydrolysis in broiler chickens. *Poult. Sci.* 83:1358- 1367.
- Torki, M. and Farahmand-Pour, M., 2007. Use of dietary enzyme inclusion and seed germination to improve feeding value of sorghum for broiler chicks. In *Proc. 16th European Symp. Poult. Nutr.* Strasbourg, France (pp. 643-646).
- Traore, T., Mouquet, C., Icard-Vernire, C., Traore, A.S., and Treche, S., 2004. Changes in Nutrient Composition, Phytate and Cyanide Contents and Alpha-amylase Activity during Cereal Malting in Small Production Units in Ouagadougou (Burkina Faso), *Food Chemistry Vol. 88*, pp. 105-114.
- Travis, D.K., Mitchell, R. T., and Joe, D.H., 2006. Variation in nutritional value of sorghum hybrids with contrasting seed weight characteristics and comparisons with maize in broiler chickens. *Journal of Crop Science*, 46: 695-699.
- USDA. 2019. World agricultural production. United States Department of agriculture. Foreign Agricultural Service, Circular Series WAP 5–19.
- Waniska, R.D., & Rooney, L.W., 2002. Structure and chemistry of the sorghum caryopsis. In: Smith, C.W. & R.A. Fredericksen (Ed), *Sorghum origin, history, technology, and production*. New York: John Wiley & Sons Inc.
- Xu, D., Shu, G., Liu, Y., Qin, P., Zheng, Y., Tian, Y., Zhao, X., and Du, X., 2022. Farm environmental enrichments improve the welfare of layer chicks and pullets: A comprehensive review. *Animals*, 2022 (12), 2610. 28 pages. <https://doi.org/10.3390/ani12192610>.